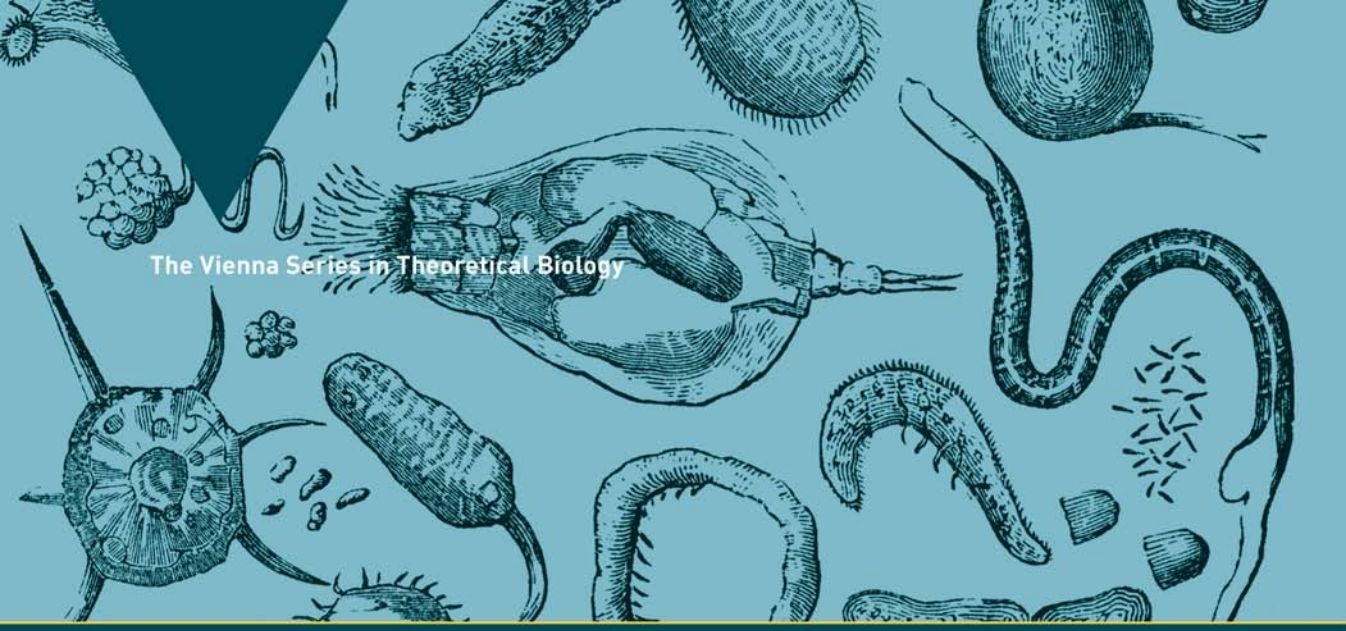


The Vienna Series in Theoretical Biology

The Major Transitions in Evolution Revisited

edited by
Brett Calcott and Kim Sterelny

Major Transitions in Evolution Revisited

Vienna Series in Theoretical Biology

Gerd B. Müller, Günter P. Wagner, and Werner Callebaut, editors

The Evolution of Cognition, edited by Cecilia Heyes and Ludwig Huber, 2000

Origination of Organismal Form: Beyond the Gene in Development and Evolutionary Biology, edited by Gerd B. Müller and Stuart A. Newman, 2003

Environment, Development, and Evolution: Toward a Synthesis, edited by Brian K. Hall, Roy D. Pearson, and Gerd B. Müller, 2004

Evolution of Communication Systems: A Comparative Approach, edited by D. Kimbrough Oller and Ulrike Griebel, 2004

Modularity: Understanding the Development and Evolution of Natural Complex Systems, edited by Werner Callebaut and Diego Rasskin-Gutman, 2005

Compositional Evolution: The Impact of Sex, Symbiosis, and Modularity on the Gradualist Framework of Evolution, by Richard A. Watson, 2006

Biological Emergences: Evolution by Natural Experiment, by Robert G. B. Reid, 2007

Modeling Biology: Structure, Behaviors, Evolution, edited by Manfred D. Laubichler and Gerd B. Müller, 2007

Evolution of Communicative Flexibility: Complexity, Creativity, and Adaptability in Human and Animal Communication, edited by Kimbrough D. Oller and Ulrike Griebel, 2008

Functions in Biological and Artificial Worlds: Comparative Philosophical Perspectives, edited by Ulrich Krohs and Peter Kroes, 2009

Cognitive Biology: Evolutionary and Developmental Perspectives on Mind, Brain, and Behavior, edited by Luca Tommasi, Mary A. Peterson, and Lynn Nadel, 2009

Innovation in Cultural Systems: Contributions from Evolutionary Anthropology, edited by Michael J. O'Brien and Stephen J. Shennan, 2010

Transformations of Lamarckism: From Subtle Fluids to Molecular Biology, edited by Snait B. Gissis, and Eva Jablonka, 2011

The Major Transitions in Evolution Revisited, edited by Brett Calcott and Kim Sterelny, 2011

The Major Transitions in Evolution Revisited

edited by Brett Calcott and Kim Sterelny

The MIT Press
Cambridge, Massachusetts
London, England

For information about special quantity discounts, please email special_sales@mitpress.mit.edu

This book was set in Times New Roman on InDesign by Asco Typesetters, Hong Kong. Printed and bound in the United States of America.

Library of Congress Cataloging-in-Publication Data

The major transitions in evolution revisited / edited by Brett Calcott and Kim Sterelny
p. cm. — (Vienna series in theoretical biology)
Includes bibliographical references and index.
ISBN 978-0-262-01524-0 (hardcover : alk. paper)
1. Evolution (Biology) 2. Biodiversity. 3. Genetic transformation. 4. Population genetics. 5. Maynard Smith, John, 1920– Major transitions in evolution. I. Calcott, Brett. II. Sterelny, Kim.
QH366.2.M34 2011
576.8—dc22 2010028052

10 9 8 7 6 5 4 3 2 1

Contents

	Series Foreword	vii
	Preface and Acknowledgments	ix
	Introduction: A Dynamic View of Evolution Brett Calcott and Kim Sterelny	1
I	**A BIG PICTURE OF BIG PICTURES OF LIFE'S HISTORY**	15
1	**The Miscellaneous Transitions in Evolution** Daniel W. McShea and Carl Simpson	19
2	**Alternative Patterns of Explanation for Major Transitions** Brett Calcott	35
3	**Biological Ontology and Hierarchical Organization: A Defense of Rank Freedom** Samir Okasha	53
4	**Darwinian Populations and Transitions in Individuality** Peter Godfrey-Smith	65
5	**Evolvability Reconsidered** Kim Sterelny	83
II	**THE PROKARYOTE'S TALE**	101
6	**To Be or Not To Be: Where Is Self-Preservation in Evolutionary Theory?** Pamela Lyon	105
7	**The Evolution of Restraint in Structured Populations: Setting the Stage for an Egalitarian Major Transition** Benjamin Kerr and Joshua Nahum	127

8	**Conflicts among Levels of Selection as Fuel for the Evolution of Individuality** Paul B. Rainey and Benjamin Kerr	141
III	**COMPLEXITY AND THE DEVELOPMENTAL CYCLE**	163
9	**Evolutionary Transitions in Individuality: Multicellularity and Sex** Richard E. Michod	169
10	**How Many Levels Are There? How Insights from Evolutionary Transitions in Individuality Help Measure the Hierarchical Complexity of Life** Carl Simpson	199
11	**Plant Individuality and Multilevel Selection Theory** Ellen Clarke	227
12	**Phylogenetic, Functional, and Geological Perspectives on Complex Multicellularity** Andrew H. Knoll and David Hewitt	251
13	**The Small Picture Approach to the Big Picture: Using DNA Sequences to Investigate the Diversification of Animal Body Plans** Lindell Bromham	271
IV	**CONCLUDING REMARKS**	299
14	**Concluding Remarks** Eörs Szathmáry and Chrisantha Fernando	301
	Contributors	311
	Index	313

Series Foreword

Biology is becoming the leading science in this century. As in all other sciences, progress in biology depends on interactions between empirical research, theory building, and modeling. However, whereas the techniques and methods of descriptive and experimental biology have evolved dramatically in recent years, generating a flood of highly detailed empirical data, the integration of these results into useful theoretical frameworks has lagged behind. Driven largely by pragmatic and technical considerations, research in biology continues to be less guided by theory than seems indicated. By promoting the formulation and discussion of new theoretical concepts in the biosciences, this series is intended to help fill the gaps in our understanding of some of the major open questions of biology, such as the origin and organization of organismal form, the relationship between development and evolution, and the biological bases of cognition and mind.

Theoretical biology has important roots in the experimental biology movement of early-twentieth-century Vienna. Paul Weiss and Ludwig von Bertalanffy were among the first to use the term *theoretical biology* in a modern scientific context. In their understanding the subject was not limited to mathematical formalization, as is often the case today, but extended to the conceptual problems and foundations of biology. It is this commitment to a comprehensive, cross-disciplinary integration of theoretical concepts that the present series intends to emphasize. Today, theoretical biology has genetic, developmental, and evolutionary components, the central connective themes in modern biology, but also includes relevant aspects of computational biology, semiotics, and cognition research and extends to the naturalistic philosophy of sciences.

The "Vienna Series" grew out of theory-oriented workshops, organized by the Konrad Lorenz Institute for Evolution and Cognition Research (KLI), an international center for advanced study closely associated with the University of Vienna. The KLI fosters research projects, workshops, archives, book projects, and the journal *Biological Theory*, all devoted to aspects of theoretical biology, with an emphasis on integrating the developmental,

evolutionary, and cognitive sciences. The series editors welcome suggestions for book projects in these fields.

Gerd B. Müller, University of Vienna and KLI
Günter P. Wagner, Yale University and KLI
Werner Callebaut, Hasselt University and KLI

Preface and Acknowledgments

As with other volumes in this series, this book had its roots in a KLI workshop, one organized around themes similar to those of the volume. KLI workshops are immensely productive and enjoyable intellectual experiences, but they are also very intense. The workshop participants spend three days in sustained, focused, and cumulative discussion. Each of the participants' talks is followed with a typical question and answer session, but that is just the beginning. The discussion is supplemented and extended with further (often probing) talk over coffee, lunch, dinner, and drinks. As the group interacts, points are made, ideas are thrashed out, and theories develop, with each session building on the previous one. All of this takes a great deal of time and intellectual energy, so we want to thank all the workshop participants for giving so generously: Lindell Bromham, Peter Godfrey-Smith, Ben Kerr, Andy Knoll, Michael Lachmann, Rick Michod, Samir Okasha, Alirio Roslaes, Carl Simpson, and Eörs Szathmáry. In addition, Werner Callebaut, Gerd Müller, and a number of visitors to the KLI were able to join in the discussion and contribute to the workshop.

Maintaining this intensity, and still managing to have a fantastic time, was possible only because of an enormous amount of work done by those at the KLI institute. Everyone went to great lengths to ensure that everything ran smoothly and easily, so we could all concentrate on major transitions (rather than, say, Internet connections or flight details). In particular, Eva Karner went far beyond any conceivable duty; she was endlessly helpful before, during, and after the workshop to us both. And once the day's papers were done and dissected, the Institute was generous and imaginative in making sure we all had a good time, and that wine, beer, and pork were never in short supply. We must especially thank Werner Callebaut for this. Werner was part of the intellectual life of the workshop, but he was also the prime mover of its social life.

The workshop was preceded and followed by our own collaborative work on major transitions and, more generally, on macroevolutionary dynamics and their relation to microevolution. That began when Brett was a graduate student, but we owe a great debt to the Templeton Foundation for the opportunity to develop and extend it, for that foundation funded Brett to work as a postdoctoral fellow on the evolution of complexity. Their support allowed Brett to work on the issues canvassed by the workshop and to prepare a presentation

for it. But it also enabled him to play a central role in organizing the workshop and editing the collection that has emerged from it. More recently, the philosophy program of the Australian National University (ANU) has supported Brett to work on these issues (as, along with Victoria University Wellington, it has long supported Kim).

This collection has been longer coming than we hoped. As always, not everyone who attended the workshop was able to deliver a chapter. We also wanted to broaden the range of the volume, and at Gerd Müller's suggestion, we solicited papers from other sources. All this took time, as did refereeing, revising, and updating. But the collection has come, and we hope it testifies to the originality and vitality of the extraordinarily fertile mind of John Maynard-Smith over his long career, and to the insight generated by his collaboration with Eörs Szathmáry.

Introduction: A Dynamic View of Evolution

Brett Calcott and Kim Sterelny

The Major Transitions in Evolution is part of an important tradition in evolutionary biology. This tradition attempts to identify large-scale patterns in life's history, and to relate those patterns to evolutionary mechanisms that can be studied empirically. Here, we sketch some of this history and give our take on the importance of these projects. But we also lay out the ways in which Maynard Smith's and Szathmáry's formulation contrasts with its predecessors, and explain the significance of those contrasts. These themes connect in many ways with individual chapters of this collection, and at times we identify those connections. But for the most part, we postpone specific discussion of the chapters to the section introductions, concentrating instead on very general themes.

We suggest that one crucial feature of the Maynard Smith and Szathmáry picture is that it is dynamic. In contrast to many others, their vision of life's history incorporates important changes in the evolutionary process itself. Their "Major Transitions" identified uncontroversially important episodes in evolutionary history, but each of these episodes also changes some key evolutionary factor: the construction of new individuals; the increasing bandwidth and fidelity of inheritance; the establishment of new inheritance channels; the development of open-ended sources of variation. These are all fundamental parts of the evolutionary process—change these, and we change the mechanisms of evolution. We review the importance of this dynamism in the context of other "big picture" visions of life's history, and suggest some ways in which their picture might be further developed.

Why History Matters

Evolutionary biology is, in part, a historical science. One of its aims is to explain the shape of life's history—its major episodes and developments. Such a project presupposes we know the features in life's history most in need of explanation. To explain the shape, we first need to decide what that shape is. A recurring and controversial suggestion is that life's history is marked by a directional trend. As a whole, the average value of some key parameter (diversity, complexity, adaptedness) increases over time. (For reviews, see: Jablonski

2007; Knoll and Bambach 2000; McShea 1998). Much of this work has grown out of the idea that the history of life is progressive. From simple origins, more advanced, better adapted, better designed forms have emerged, replacing their inferior predecessors. This idea has been at once influential and deeply problematic (Ruse 1996). Making the idea of progressive change empirically tractable, and purging it of anthropocentrism, has proved extraordinarily difficult. The problem of detoxifying the concept of progress has motivated attempts to decouple work on large-scale trends from directional and progressivist ideas of history. Instead, we have seen formulations of directionality focused on complexity, diversity, or some similar surrogate for progress , though each of these has its own problems (for example, Vermeij 1987, 1999).

In reflecting on this disciplinary history in their chapter, McShea and Simpson distinguish between two methods of investigating such trends. The first takes off from a distinctive account of evolutionary mechanisms. Having developed a picture of evolutionary process, theorists of this variety then ask: What will the history of life be like, if this really is how evolution works? So in this method, one begins with the resources of biological theory and asks what signature these supposed evolutionary mechanisms, if correctly identified, would have on the history of life. Two recent, theory-driven projects that roughly fit this theory-first approach to history are Mark Ridley's *Mendel's Demon*, which focuses on transitions in the fidelity of heritability (Ridley 2000), and Wallace Arthur's work on developmental drive (Arthur 2004). Arthur argues that there are biases in the supply of variation of selection, and that these will influence large-scale evolutionary trajectories in ways we might detect. Neither of these two projects depend on any pretheoretic conception of life's history.

A second method begins with an attempt to reconstruct the idea of progress: the pretheoretic intuition that living beings constitute a series from the simplest forms at one end to humans at the other. McShea and Simpson insist that this project is legitimate. While we have probably anthropocentrically projected a comforting human illusion onto the natural world, it is also possible that we are responding to a real structure in nature. But though legitimate, so far this method has been unsuccessful. McShea and Simpson think there have been near misses, but, as yet, no one (including Maynard Smith and Szathmáry) has given a coherent, theoretically well-motivated account of the history of life in which the evolution of humanlike creatures is the predictable outcome of a driven trend.

Despite the conceptual and empirical difficulties that face these projects, the program of identifying and explaining large-scale patterns in the history of life has never stopped. For example, contrasting hypotheses are defended by Geerat Vermeij and the late Stephen Gould. Vermeij argues that the history of life is dominated by an ecological arrow of time. The world's ecosystems have been restructured over time, as high-energy, high-impact keystone species replace those with lower energetic needs and consequently a lighter footprint on their world. The relationship between high-energy and low-energy species is often asymmetric: High-energy species tend to make the environment less friendly for low-

impact species but not vice versa (because they are low impact). So, although there are countless local exceptions and quiet corners, in Vermeij's model, the pace of life increases over time (Leigh, Vermeij, and Wikelski 2009; Vermeij 1999).

Stephen Gould has articulated an influential alternative framework—his model of "passive diffusion from the left wall" (Gould 1996). Gould accepts that, in some sense, life's history is directional. The complexity of the most complex organism extant tends to increase over time. But, Gould argues, this reflects no deep fact about the dynamics of evolution. This trend is a consequence of life's simple origins, and of the fact that while there is no maximum bound on an organism's complexity, there is a lower one. In such circumstances, many specific histories of origin and extinction, each different from and often independent of one another, will tend to sum to a trend of increasing maxima. There is a causal explanation of each data point, each twig in the history of life, but there is no unified history of the sequence as a whole (see McShea and Simpson, this volume).

As we noted earlier, these projects are partly driven by the intrinsic importance of questions about the large-scale shape of life. But taking a big picture view of life's history also plays a crucial role in testing and refining our understanding of how evolution works. History supplies smoking guns; phenomena that provide crucial evidence for one version of evolutionary theory over others. It is a source of puzzling cases that challenge and stretch the explanatory resources of different versions of evolutionary theory. Most famously and persistently, history poses the problem of scale: of whether large-scale patterns are nothing more than the accumulated results of well-understood microevolutionary processes playing out in local populations over a few generations. So, for example, our current best reconstruction of the history of life is stocked with examples of sudden bursts of evolutionary inventiveness and the evolution of extraordinary novelties. Yet it is also rich in examples of phenotypic conservatism. Arguably, both rapidly appearing novelties and stasis call into question uniformitarian views of evolutionary change (Gould 2002). Incorporating a larger expanse of history also enables us to explore the interplay between internal and environmental factors in driving evolutionary trajectories, for microevolutionary accounts can often assume a largely static environment. Knoll and Hewitt explore this interplay in their chapter on the evolution of multicellularity (see also Calcott and Sterelny submitted; Sterelny 2009).

Most important, deep history forces us to examine the origins of evolutionary agents whose essential characteristics are presupposed by many specific research agendas in evolutionary biology. Deep history requires us to ask about the origin not just of species but of genes, cells, organisms, and life itself. Even if evolution is, indeed, largely the history of gene change (Lynch 2007), we need an explanation of the origins of genes and of their replication. We noted previously that history manufactures smoking guns. Arguably, the major transitions are themselves such smoking guns. Among the major transitions are episodes of the creation of new kinds of evolutionary agent: eukaryotic cells; multicelled animals; social insects. These episodes of the evolution of individuality show that selection

acts on collectives of fitness-bearing agents, not just on those agents themselves, and that higher-level selection drives evolutionary trajectories.

Evolution Upgraded: A Dynamic Vision of Life's History

Many big picture models of the history of life have been static, conceiving of evolutionary possibility as fixed over time. In *Major Transitions in Evolution*, John Maynard Smith and Eörs Szathmáry bought a much more dynamic model to debates about the history of life. Instead of conceptualizing life as evolving through a fixed, though immense, space of organic design, and asking how that space is explored over time, Maynard Smith and Szathmáry conceived of the space of biological possibility as itself evolving (Maynard Smith and Szathmáry 1995, 1999; Szathmáry and Maynard Smith 1997). One way to think of changes in life's potential is to identify key innovations. So, for example, Andrew Parker argues that the invention of sight powered massive change in the Cambrian Explosion (Parker 2003) and Nick Lane identifies a set of these innovations—such as photosynthesis, movement, and hot blood—in his *Life Ascending: The Ten Great Inventions of Evolution* (Lane 2009). These innovations also led to great expansions of phylogenetic diversity: The evolution of photosynthesis made possible the evolution of vast numbers of new species. Each expands ecospace: Organisms make a living in very new ways, and in new places, as a result of these innovations. These all seem good candidates for possibility-expanding innovations.

Maynard Smith and Szathmáry have a more profound approach. Their key events are changes in the evolutionary process itself—this is how they identified their Major Transitions (see table 1 in Szathmáry and Maynard Smith 1995). In this view, the major transitions in evolution modify core elements of the evolutionary process itself. Like a robot that continually reprograms itself, or a factory that manufactures parts to change its own operation, evolution upgrades itself, amplifying the kinds of further change that are possible. This idea is developed in Peter Godfrey Smith's chapter and in his book on Darwinian populations (Godfrey-Smith 2009). Following Lewontin, he identifies a set of minimal conditions for evolutionary change, and then discusses ways in which those minimal conditions can be enriched to make large-scale, permanent change possible.

Two central themes in Maynard Smith and Szathmáry's book (1995) develop this idea of change in the conditions that make evolutionary change possible. One concerns the expansion of mechanisms of hereditary—where richer and more accurate systems of the intergenerational flow of information evolve. The other focuses on the evolution of new levels of biological individuality; an evolutionary change after which previously independent entities now reproduce together, sharing their evolutionary fate. Both mark out core features of the Darwinian process. One is a radical change in the kind of individual from which evolving populations and lineages are built. The other is a change in the processes

relating these individuals across generations. A third, less well-explored theme, concerns the generation of variation, which they touch on in their final chapter on language. Maynard Smith and Szathmáry are impressed by both the generative and the representational capacity of language, seeing a close analogy with the informational capacity of genes. Thus, we see three core features of the Darwinian process of change—the subject of change, how change is passed on, and ways in which further change is generated—are all themselves subject to modification.

This dynamic view contrasts strongly with Gould's view that morphospace has been explored by passive diffusion from a starting point of minimal complexity. It provides a different way to conceive of the bounds on complexity, and on how they have changed throughout life's history. Even if we concede that there is simply passive diffusion from the initial minimal conditions of life (and this is controversial), the Maynard Smith and Szathmáry view suggests there is an upper bound, too. Until problems of inheritance were solved, there was an upper limit on early replicator complexity. Until a method for inheriting somatic differences in genetically identical cells was established, there was an upper limit on the complexity of multicellular organisms. And until cooperation became manageable, there was an upper limit on the size and complexity of social groups of animals, including humans.

The lower bound on organismal complexity has also changed over time. Viruses depend on harnessing and subverting important mechanisms in more complex organisms for their own evolutionary ends. Without these more complex hosts, viruses would not be possible. Dan Dennett has pointed out that there is often more than one pattern in a given set of data. In many cases, we can see different patterns, depending on the grain of our analysis and our explanatory interests (Dennett 1991). Thus, one important and difficult issue is whether these alternative ideas of deep history are empirically distinct, competing models or just different heuristics, directing our attention in different ways to specific episodes and mechanisms. In any case, the major transitions model is heuristically powerful, for it forces us to ask important questions about the conditions of change and the stability of those conditions.

Evolution in Flux

By addressing the broad sweep of history—from early replicators through to the complexities of human language—and doing so with an eye to the changing nature of the evolutionary process itself, Maynard Smith and Szathmáry address and incorporate many challenges to the received view that have been raised in the last thirty years. Until the 1980s, virtually all of evolutionary biology focused on the causes and consequences of fitness differences among individual organisms over relatively short periods of time. The received view of evolutionary biology was organism-centered and microevolutionary. Evolutionary change took place through small changes in local populations, and these

changes could be studied over relatively few generations. Mayr, Dobzhansky, Stebbins, and their allies had forged these assumptions into a postwar synthesis of whole-organism biology with the population genetics of the 1930s. Their legacy was to convince the profession that a relatively direct relationship existed both between genetic and phenotypic change (so evolution could be conceptualized as change in gene frequency) and between change in local populations and the observable patterns in longer spans of evolutionary history.

That simple picture of the relationship between microevolutionary change and the macroevolutionary pattern has never been completely accepted (for an overview, see Depew and Weber 1995). But by the 1980s it had become openly controversial. In 1984, John Maynard Smith remarked that paleobiology was once more at the center of evolutionary biology, challenging the resources of the microevolutionary toolkit to explain patterns in the history of life (Maynard Smith 1984). The challenge originated with Gould's and Eldredge's "punctuated equilibrium" model of species evolution. It expanded in various ways: to questions about directional trends; to the role of mass extinction in shaping life's history; to the supposed large-scale stasis in basic morphology after the Cambrian Explosion; and to issues about the role of contingency in evolutionary history. No one showed that microevolutionary mechanisms *couldn't* explain macroevolutionary patterns; but there was no longer a consensus that these mechanisms sufficed.

At the beginning of this ferment, paleobiology and its distinctive evidence played the key role. But gradually, molecular data began to be important, both because it made reconstructions of phylogenetic history much more reliable and especially as it allowed evolutionary biology to begin to build data about small organisms into their big picture. Microbiology played almost no role in the establishment of the synthesis consensus. If it had, Mayr's biological species concept could hardly have dominated thinking for so long. The rise of molecular biology was an inestimable boom to microbiology, giving real tools through which microbial, and especially prokaryote, evolution could be studied. The results have reshaped our picture of life's history, with (i) the establishment of the symbiotic origin of eukaryotes (Clearly, at least one utterly pivotal event in life's history was a lineage fusion, not an incrementally diverging lineage fission.); (ii) the discovery that prokaryotes are divided into two deeply diverging and biochemically very different branches, the Achaea and the Eubacteria; and (iii) the discovery that horizontal gene transfer has played, and continues to play, a central role in prokaryote evolution. It gradually became clear that evolutionary biology, in focusing on multicelled animals and plants, had ignored the importance of horizontal exchange, and the fusing of distinct lineages with complementary parts into new individuals (Doolittle and Bapteste 2007; O'Malley and Dupre 2007; Woese 2008).

The central status of the organism as unit of selection also came under question. The challenge arose partly from theoretical concerns. In 1966, George Williams published his brilliant *Adaptation and Natural Selection*, arguing that the gene rather than the organism

was the true unit of selection. Richard Dawkins (especially, but not alone) championed this idea, arguing that genes deserve a central role for three reasons. First, genes have effects on their environment that make them more or less likely to be replicated. Second, genes are replicated in ways that make cumulative evolution possible. Third, while genes' advantageous environmental effects are usually effects on the organisms of which they are a part, that is not universally so. Dawkins's *The Extended Phenotype* put both the nature and the importance of biological individuality into question, arguing that the reach of the gene extended beyond the boundaries of the organism.

But the status of the organism also began to be questioned in broader contexts. One was historical. In 1987, Leo Buss's *Evolution of Individuality* argued that individuality was a derived character, and that the unit of selection could itself change over the course of evolution. At roughly the same time, the role of groups returned to the debating table and, more important, the importance of cooperation moved to center stage. Williams's critique of good-for-the-species explanations and Hamilton's kin-selected model of cooperative behavior jointly seemed to kill high-level models of selection stone dead. But from the late 1980s, driven largely by the persistence of David Sloan Wilson, multilevel selection models crept back into view. Wilson and others argued that there were cases resistant to kin selection explanations and that some versions of multilevel selection models were much less unrealistic in their assumptions than had been suggested. Researchers began talking about eusocial insects as superorganisms again (for example, in Seeley's *Wisdom of the Hive*).

These issues are still being debated, though with increasing consensus and theoretical clarity (West, Griffin, and Gardner 2007). Perhaps the most important result of this debate was the focus on the importance of cooperation, and its relevance at all levels in the hierarchy of life (Michod and Herron, 2006). In particular, it has become clear that cooperation has played a deep and important role in the evolution of life's complexity, resulting in alliances both within and across species, and producing stable, reproducing, units that can be treated as individuals in their own right within a Darwinian framework.

Last, claims that developmental biology had been "left out" of the modern synthesis began to emerge. An explosion in our understanding of how development works, including the discovery of so-called master genes, along with a wealth of information about species' genomes, provoked a reevaluation of the importance of the relationship between evolutionary and developmental biology. Exactly what this claim amounted to was often far from clear, but a central issue revolved around understanding the origins of variation. Although the synthesis relied on variation, it lacked a theory of the source of such variation (Kirschner and Gerhart 2005). Such a theory is important in evolutionary biology, for unless the supply of variation is rich and unbiased—providing a uniform variety to be selected—that supply will influence evolutionary trajectories (Arthur 2004). Importantly, many of the discoveries from developmental genetics suggested that we have no reason to expect the supply of variation to be, in general, unbiased and unlimited. So the simple, Fisherian model of variation is slowly being replaced by more realistic alternatives. Kirschner and

Gerhart have made one promising attempt to develop a general theory of variation, and while they emphasize multicellular development, their principles of developmental organization are formulated very generally. It is therefore possible that these principles of developmental organization—principles that result in biases in the supply of variation—could be used to characterize multiple levels in the biological hierarchy.

One response to these challenges, though a dull one, would be to maintain that the standard narrative is essentially true, but with exceptions. Selection acts on populations of organisms, and change over time is the summed result of these population-level events, unless those organisms are too old, too small, too social, or have too inefficient mechanisms for establishing a division between germline and soma. Maynard Smith and Szathmáry offer a much more interesting alternative. Instead, we should see the exceptions as transitions to (or in some cases, from—see Leroi, Koufopanou, and Burt 2003) the standard case. Understanding the exceptions, and why exceptions exist in some lineages, is crucial to understanding the scope and limits of the more familiar model of evolution as change in local populations of organisms.

What Do The Transitions Share?

No one will deny that the events identified by Maynard Smith and Szathmáry are important and interesting. But is that all they share? Why *these* transitions, and not others? McShea and Simpson (this volume) push the problem of unity hard; they doubt that Maynard Smith and Szathmáry's major transitions capture the same kind of evolutionary episode, even though each may be individually important. Of course, the transitions need not be unified in this strong sense for them to be worth studying as a group. As we have suggested, family resemblances might still exist between them, so that studying one offers heuristic insight into others, and they may be similar in that each offers a striking challenge to received models of evolutionary theory.

That said, there has been a vigorous tradition of seeing the transitions as unified; those who have followed up Maynard Smith and Szathmáry's ideas, especially, have taken the common thread to be the evolution of new kinds of evolutionary individuals (Michod 1999, in particular). This emphasis is reflected in the content of the current volume: The majority of the papers are concerned with some aspect of transitions in individuality. As we have suggested, there is a more expansive way to interpret these transitions—one that involves not just transitions in individuality, but any shift in the core components of the evolutionary process.

Peter Godfrey-Smith (this volume and Godfrey-Smith 2009) introduces a conceptual framework that is helpful in exploring this idea, and also in responding to the challenge from McShea and Simpson. Godfrey-Smith co-opts Richard Lewontin's classical formulation of the minimum conditions of evolution to introduce the concept of a Darwinian population. A Darwinian population is a population of agents that reproduce, but not with equal

prospects of success, and in such a way that descendents resemble their parents. Thus, Darwinian populations satisfy the minimum conditions for evolutionary change, and a Darwinian individual is simply a member of a Darwinian population. As Godfrey-Smith points out, there are central and peripheral instances of Darwinian populations, for there are clear and marginal cases of reproduction, clear and marginal cases of inheritance, and perhaps clear and marginal cases of fitness differences. Second, he points out that reproduction, fitness difference, and inheritance are only the *minimal conditions* for evolutionary change. If evolutionary change in a population is to generate complex outcomes, the population must satisfy extra conditions, though the identity of those extra conditions remains controversial. A *core case* of a Darwinian population, then, is a population that (i) clearly and unambiguously satisfies the minimum conditions for evolutionary change, and (ii) also satisfies (to an important extent) the extra conditions, whatever they may be, that make possible the evolution of complex traits.

At the beginning of a transition, then, we have a population of Darwinian agents interacting with others in fitness-affecting ways. But that population is itself structured: Interactions are patterned so that local groups interact with one another in ways that contrast with their interactions with agents outside that local group. So there are groups—collections—of interacting Darwinian agents. At the beginning of a transition, these collections are at best peripheral or marginal cases of Darwinian agents, and the metapopulation of groups is at best a peripheral example of a Darwinian population. The group members, on the other hand, are core Darwinian agents. By the end of a transition, the collections have become collectives. They are now core Darwinian agents in an unambiguously Darwinian population. Their members are still present, but they have become parts of collectives, and the evolutionary fate of those surviving descendents is now welded together. Often, they are no longer clear cases of Darwinian individuals in a Darwinian population. Thus, at the end of a transition, we see a new evolutionary agent—a collective. But that single agent is more structurally complex than those we saw interacting at the beginning of the process. The major transitions are episodes in which the *vertical complexity* of life has increased through the transformation of a collection or group into a collective.

Consider, for example, eukaryote origins. There is ongoing controversy about the identity of the partners and the process of fusion through which the eukaryote cell first appeared (de Duve 2007). But there is no doubt that the eukaryotes evolved through such a transition. The ancestors of the mitochondrion (and perhaps the nucleus)—now a part of the eukaryote—were once independent organisms, fitness bearers in their own right. They were once paradigm Darwinian individuals; they are now parts of a more structurally complex Darwinian individual, and are themselves less clear cases of Darwinian individuality. Many of the canonical transitions are naturally seen as the evolution of a new form of Darwinian individual, and as expanding the space of biological possibility as a result. We know very little about the earliest transitions identified by Maynard Smith and Szathmáry. But the aggregation of independent replicators into compartments and into protochromosomes

are such cases (if life as we know it was built via such stages). So, too, is the evolution of multicelled and social organisms.

There are two problems in thinking that the major transitions *just are* transitions in individuality. First, David Queller has pointed out that there seem to be two very different transitions in individuality: "egalitarian" and "fraternal" transitions (Queller 2000). Perhaps these should not be lumped together. Eukaryote evolution is the paradigm of an egalitarian transition, for the partnership that became the new Darwinian individual did not begin with an association between closely related individuals. In contrast, the evolution of multicelled organisms (and eusocial animals) is a fused alliance between close relatives. Explaining these two types of transition poses quite different challenges. In egalitarian transitions, differentiation between the partners, and hence the potential profits of specialization, come for free. But there is no automatic overlap of evolutionary interest, and (in the first instance, anyway) no possibility of a division of reproductive labor. And so there are potentially unmanageable problems of conflict. In fraternal transitions, there is an overlap of evolutionary interest (in clones, identity), so the problem of conflict is less pressing. But the profit of cooperation is more elusive, as differentiation does not predate partnership (see also Calcott 2008).

Second, as McShea and Simpson point out, neither the evolution of sex nor the evolution of the distinctive forms of human social life fits this model of the making of a major transition. Of course, the list is not sacred. Perhaps major transitions are just transitions in individuality, and Maynard Smith and Szathmáry simply got the membership wrong. It is also possible that appearances are deceptive. Michod (this volume) argues that sex does fit this model, because, at least in those lineages for which sex is essential for reproduction, the mating pair rather than the mating individuals are the real bearers of fitness. It is *the pair* that succeeds or fails. Human culture might fit the model, if, as is sometimes argued, language-mediated cultural learning, and that alone, explains why group selection has been a powerful force shaping human evolution (Boehm 2000; Richerson and Boyd 2001). Thus, it might be possible to shoehorn these examples into this conception of a transition after all, but it would be shoehorning.

One way of seeing a common feature among the major transitions is therefore to look to population structure and selective environment. Calcott (this volume) points out that most work on transitions in individuality takes this approach. This work poses the following question: What was it about the early-transition population structure and environment that made selection for cooperation so powerful that agents did not just evolve to cooperate, but evolved a ballistic commitment to cooperate, giving up the capacity to reproduce independently? Calcott suggests that there is a quite different strategy for identifying the unifying features of transitions—an engineering strategy that focuses on the structure not of populations but of prototypical individuals within them. Expansions in phenotype complexity (as when a collection of individuals fuse into a collective individual) pose engineering problems. Organisms must be constructed and maintained in different ways as they change in

size and complexity, especially given that organisms must function while they change (both in ontogeny, and over evolutionary time). For example, Knoll and Hewitt (this volume) explore the problem of resource flow, and the constraint that flow imposes on multicellularity. As multicelled lineages evolve from threads and films to organisms with internal structure, modest increases in size and three-dimensional complexity exceed the limits of systems dependent on diffusion. The evolution of new kinds of individuals typically involves changes in physical scale, not just solutions to cooperation dilemmas. While the mechanical details of engineering challenges will differ across transitions, Calcott suggests the general character of both challenge and solution may well be invariant. For example, all transitions to new complex individuals depend on managing differentiation and the division of labor without top-down control, transitions in increasingly complex management without managers.

One of Maynard Smith and Szathmáry's own ideas dovetails with Calcott's line of thought. We develop the idea briefly as an example of an alternative way of seeing common features across the transitions, for, while they certainly discuss transitions in individuality, they also suggest that the major transitions are transitions in the mechanisms of inheritance involving innovations that expand the fidelity and bandwidth of inheritance. We see these ideas of inheritance as importantly connected to transitions in individuality, for increasing vertical complexity increases the problem of developmental control. Cumulative evolution depends on high-fidelity inheritance, and high-fidelity inheritance depends on sending developmental signals across the generation with high bandwidth and fidelity, and sending them in ways that enable these signals to structure development in the next generation. Increasingly complex phenotypes can evolve only if gene replication with high fidelity and bandwidth also evolves. But that is not enough; genes must be turned on and off in the right sequence, and in predictable environments.

Expansions in vertical complexity therefore increase the demands on inheritance mechanisms. Multicelled organisms have evolved many times (Bonner 1998), but only in a few cases have these lineages generated impressive disparity and diversity. The evolution of complex multicellularity requires the evolution of a higher-level unit with its own fitness values. But it requires more—the evolution of a developmental cycle—and that in turn requires a major advance in mechanisms of inheritance. Protist genes never have to contribute to building afresh the critical inner cellular structures of protists. The reproduction of these crucial intercellular structures can largely be reduced to growth and fission. In contrast, organs and tissues do not exist in miniature in fertilized ova. Complex multicelled organisms exist only because there are developmental cycles in which key structures of adult organisms are rebuilt from scratch in the new generation. So the problem of cross-generation fidelity is much more pressing for macrobes than for microbes.

For this reason, Calcott and I have suggested that the Cambrian Explosion really does constitute a major transition by the lights of Maynard Smith and Szathmáry, even though it is not on their canonical list (see also Jablonka and Lamb 2006, but for somewhat different

reasons). We suggest that the egg is a major breakthrough, allowing the flow of genes across the generations to orchestrate development in a fine-grained and reliable way (Calcott and Sterelny submitted; Sterelny 2009). Such developmental control, we suggest, was essential to build complex bilaterians. This suggestion—admittedly, very speculative—develops an idea of Scott Gilbert (Gilbert 2001). He outlines a program for integrating developmental biology with ecology, taking into account ecological influences on development. But environments do not just happen to organisms; organisms help make their own environment and those in which their offspring develop. Termites, for example, develop in a world built by and for termites. As a consequence, their developmental environment has been stabilized. Compared to their presocial ancestors, termite genes are expressed in a narrowed range of developmental environments, and hence the phenotypic effects of those genes are more predictable. We suspect that such increased parental control of developmental environments was crucial to the Metazoan radiation. In discussing inheritance, Maynard Smith and Szathmáry emphasize transitions in the kind and quantity of information transmitted across the generations. But for genes to have stable phenotypic effects, they must be inserted into a sufficiently structured and predictable developmental environment. It is no use just sending genes; the parental generation must build an environment in which those genes are used in the right way. As the developmental pathway becomes more complex, the gene-reading environment becomes as important as signal quantity and fidelity. The egg *is* such a structured system. It is adapted both to function in an environment and to provide an initial set of triggers for gene expression.

The various transformations in the fidelity and bandwidth of inheritance thus suggest a largely unexplored facet of Maynard Smith and Szathmáry's *Major Transitions*. The common feature of the transitions is that they involve important innovations in inheritance and developmental control. Those innovations will vary immensely in detail in particular cases, but they all either enable more information to flow across the generations or increase the reliability and precision with which that information is used.

Looking Forward

John Maynard Smith and Eörs Szathmáry presented the Major Transitions as the beginning of a research program, not its culmination. As we shall see, in the decade and more that has followed, their legacy has been developed in important ways. Most obviously, important work has been done on specific transitions: on eusociality; the origins of eukaryote cells; and especially multicellularity. There has been a very rich development of the theory of multi-level selection, and its application to the problem of individuality. There has been less focus on identifying other conditions that make major evolutionary change possible. Some of the strengths of this legacy are explored and further developed in the chapters that follow. But there is also some attempt to make at progress on the less well traveled roads signposted within *Major Transitions in Evolution*.

References

Arthur, Wallace. (2004). *Biased Embryos and Evolution*. Cambridge: Cambridge University Press.

Boehm, C. (2000). Conflict and the evolution of social control. *Journal of Consciousness Studies* 7 (1–2): 79–101.

Bonner, John Tyler. (1998). Origins of multicellularity. *Integrative Biology* 1(1): 27–36.

Calcott, Brett. (2008). The other cooperation problem: Generating benefit. *Biology and Philosophy* 23(2): 179–203.

Calcott, Brett, and Kim Sterelny. (submitted). The Cambrian Explosion and the evolution of life. *Biological Theory*.

de Duve, Christian. (2007). The origin of eukaryotes: A reappraisal. *Nature Reviews Genetics* 8(May 2007): 395–403.

Dennett, D. C. (1991). Real patterns. *Journal of Philosophy* 87: 27–51.

Depew, D., and B. H. Weber. (1995). *Darwinism Evolving: Systems Dynamics and the Genealogy of Natural Selection*. Cambridge, MA: MIT Press.

Doolittle, W. Ford, and Eric Bapteste. (2007). Pattern pluralism and the tree of life hypothesis. *Proceedings of the National Academy of Science* 104(7): 2043–2049.

Gilbert, Scott. (2001). Ecological developmental biology: Developmental biology meets the real world. *Developmental Biology* 233: 1–12.

Godfrey-Smith, Peter. (2009). *Darwinian Populations and Natural Selection*. Oxford: Oxford University Press.

Gould, Stephen Jay. (1996). *Full House: The Spread of Excellence from Plato to Darwin*. New York: Harmony Press.

Gould, Stephen Jay. (2002). *The Structure of Evolutionary Theory*. Cambridge, MA: Harvard University Press.

Jablonka, Eva, and Marion Lamb. (2006). The evolution of information in the major transitions. *Journal of Theoretical Biology* 239(2): 236–246.

Jablonski, David. (2007). Scale and hierarchy in macroevolution. *Palaeontology* 50(1): 87–109.

Kirschner, Marc W., and John C. Gerhart. (2005). *The Plausibility of Life*. New Haven, CT: Yale University Press.

Knoll, Andrew, and Richard Bambach. (2000). Directionality in the history of life: Diffusion from the left wall, or repeated scaling on the right? In Douglas Erwin and Scott Wing (eds.), *Deep Time: Paleobiology's Perspective*. Lawrence, KS: The Paleontological Society, 1–14.

Lane, Nick. (2009). *Life Ascending: The Ten Great Inventions of Evolution*. London: Profile Books.

Leigh, Egbert G. Jr., Geerat J. Vermeij, and Martin Wikelski. (2009). What do human economies, large islands and forest fragments reveal about the factors limiting ecosystem evolution? *Journal of Evolutionary Biology* 22 (1): 1–12.

Leroi, A., L. M. Koufopanou, and Austin Burt. (2003). Cancer selection. *Nature Reviews Cancer* 3(3): 226–231.

Lynch, Michael. (2007). *The Origins of Genome Architecture*. Sunderland, MA: Sinauer.

Maynard Smith, John. (1984). Paleontology at the high table. *Nature* 309: 401–402.

Maynard Smith, John, and E. Szathmáry. (1995). *The Major Transitions in Evolution*. New York: W. H. Freeman.

Maynard Smith, John, and E. Szathmáry. (1999). *The Origins of Life: From the Birth of Life to the Origins of Language*. Oxford: Oxford University Press.

McShea, Daniel W. (1998). Possible largest-scale trends in organismal evolution: Eight "live hypotheses." *Annual Review of Ecology and Systematics* 29: 293–318.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality.* Princeton, NJ: Princeton University Press.

Michod, R. E., and M. D. Herron. (2006). Cooperation and conflict during evolutionary transitions in individuality. *Journal of Evolutionary Biology* 19: 1406–1409.

O'Malley, Maureen, and John Dupre. (2007). Size doesn't matter: Towards a more inclusive philosophy of biology. *Biology and Philosophy* 22(2): 155–191.

Parker, Andrew. (2003). *In The Blink of an Eye*. New York: Basic Books.

Queller, David. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions of the Royal Society B* 355: 1647–1656.

Richerson, Peter J., and Robert Boyd. (2001). Built for speed, not for comfort. *History and Philosophy of the Life Sciences* 23: 423–463.

Ridley, Mark. (2000). *Mendel's Demon: Gene Justice and the Complexity of Life*. London: Weidenfeld and Nicholson.

Ruse, M. (1996). *Monad to Man: The Concept of Progress in Evolutionary Biology*. Cambridge, MA: Harvard University Press.

Sterelny, Kim. (2009). Novelty, plasticity and niche construction: The influence of phenotypic variation on evolution. In Anouk Barberousse, Michel Morange, and Thomas Pradeu (eds.), *Mapping the Future of Biology. Evolving Concepts and Theories*. Dordrecht, Germany: Springer.

Szathmáry, E., and J. Maynard Smith. (1995). The major evolutionary transitions. *Nature* 374: 227–232.

Szathmáry, E., and J. Maynard Smith. (1997). From replicators to reproducers: The first major transitions leading to life. *Journal of Theoretical Biology* 187: 555–571.

Vermeij, Geerat J. (1987). *Evolution and Escalation: An Ecological History of Life*. Princeton, NJ: Princeton University Press.

Vermeij, Geerat J. (1999). Inequality and the directionality of history, *American Naturalist* 153(3): 243–253.

West, S. A., A. S. Griffin, and A. Gardner. (2007). Evolutionary explanations for cooperation. *Current Biology* 17(16): R661–672.

Woese, Carl. (2008). The domains of life and their evolutionary implications. In Michael J. Dunn, Lynn B. Jorde, Peter F. Little, and Shankar Subramaniam (eds.), *Encyclopedia of Genetics, Genomics, Proteomics and Bioinformatics*. New York: John Wiley & Sons.

I A BIG PICTURE OF BIG PICTURES OF LIFE'S HISTORY

Brett Calcott and Kim Sterelny

In the introduction, we suggested that the single most important feature of Maynard Smith and Szathmáry's Major Transitions was its dynamic approach: The major changes are those that affect the key elements in the process of evolution itself. Even if this is right, it still does not isolate a single line of investigation about major transitions, nor a single way of understanding how and why they might occur. The chapters in this section sample a number of approaches to the major transitions. Each chapter critiques or extends the major transitions framework in some way, but not in the same way, nor with the same goal. Okasha, for example, takes the existing framework for granted, and argue that it entails a conceptual shift in the way we think of organisms. McShea and Simpson, in contrast, are skeptical that there is unity within the major transitions as they are currently laid out. Some of these different approaches reflect ambiguities within the major transitions literature itself (McShea and Simpson do a particularly good job of identifying some of these problems). But the different approaches also reveal the fertile ground that exists for integrating, assessing, and applying work done on the major transitions with other ideas, both in biology (such as evolvability) and in philosophy of science (such as unification, and the nature of explanation).

McShea and Simpson argue that Maynard Smith and Szathmáry do not provide a coherent, well-motivated framework for thinking about the history of life. Their specific transitions do not cohere with their advertised framework, nor can the framework be modified in a principled way to fit their list of canonical major transitions. The chief problem they see is that the final transition to human societies is an outlier; whatever it is that unifies the other transitions, it is simply not the same thing going on in this final, human, case. McShea and Simpson survey a number of earlier, lesser known, attempts to find some property that underlies and unifies large-scale evolutionary change, contrasting Maynard Smith and Szathmáry's Major Transitions with Julian Huxley's investigation of higher and lower organisms and Stebbin's eight levels of organization. Each of these various frameworks is found wanting. McShea and Simpson don't simply dismiss such frameworks because they might be thinly disguised versions of a "great chain of being," which places humans at the topmost rung. They take seriously the idea that, in principle, there could be some property that

does increase over time, and is maximized in some way with humans. In their assessment, however, none of these frameworks captures a consistent, measurable property that justifies such an ordering. They conclude by suggesting we treat more seriously the demand for theoretical consistency in any such broad-sweeping framework.

Calcott argues that the major transitions literature has been too focused on identifying conditions in which there is selection for cooperation, despite the threat of defection. While this is undoubtedly important, Calcott proposes that there are alternative resources we can draw upon to help understand these transitions. Mayr, Tinbergen, and others have remarked that there are different kinds of explanation in biology, and Calcott suggests that these other kinds of explanation reveal different ways of identifying theoretical unity across major transitions (Mayr 1961; Tinbergen 1963). Using examples from the evolution of multicellularity in *Volvox carteri*, a green algae, he shows how different researchers have deployed very different kinds of explanation to pick out different factors important in enabling this particular transition. This model system may exemplify the distinctive selective pressures necessary for a major transition, but it also exemplifies the engineering challenges involved in an expansion of vertical complexity. In this case, the factors that allow these challenges to be met are specific to *V. carteri*; however, they suggest more general conditions, involving similar structural and organizational challenges, that are present across the major transitions. Identifying these organizational prerequisites for a growth in complexity, and understanding how they might interact with the more familiar levels of selection problems, provides one way of extending work on major transitions.

Okasha's chapter explores the implications of a framework that acknowledges multiple levels of organization, arguing for a displacement of the organism as a privileged level of analysis in evolutionary biology. Much work in multilevel selection is still conceptually anchored to the organism, conceptualizing genetic and cellular evolution as evolution "below the level of the organism." In contrast, but equally anchored in the organism, selection in hives, colonies, and other groups is the selection of "superorganisms." Okasha argues that anchoring multilevel selection to a specific rank in this way is arbitrary and misleading. He makes his argument by way of an analogy between the organizational hierarchy under investigation in the major transitions, and a different kind of biological hierarchy—phylogenetic systematics. He argues that the idea of rank freedom in systematics—that there is no strict meaning to any particular level in the hierarchy of species relations—has a natural analog in the hierarchy of biological organization, and that there is no privileged rank, or level, of organization that is privileged. As he points out, this idea is not as radical as it might seem, but merely makes explicit what underlies much of the theorizing currently taking place under the rubric of multilevel selection theory.

Godfrey-Smith analyzes the evolution of levels of organization by making the idea of a *Darwinian population* central. This idea derives from an abstract summary of the essential conditions for evolution by natural selection: There must be a population of individuals

that vary in characters, that have differential survival and reproduction due to these characters, and that produce offspring whose characters correlate with those of their parents. This kind of population constitutes what Godfrey-Smith calls a *minimal concept* of a Darwinian population. He considers the ways in which this concept can be enriched to explain complex and disparate evolutionary outcomes. Crucially, he also looks at how marginal cases (those that only approximate Darwinian populations) are important for understanding transitional cases. Godfrey-Smith compares his approach with that of Michod and Sterelny and, in particular, suggests that thinking in terms of Darwinian populations provides a more fruitful approach than working with the concept of a *replicator*, an approach that has been common in thinking about levels of selection. He connects these ideas to the major transitions by noting that we can recognize different Darwinian populations embedded within one another, each at a different level of organization. During a transition, when a new level of individuality arises, there is a tendency for the lower-level populations to become *de-darwinized*—to become more marginal Darwinian populations—while those at the higher level become less marginal. Godfrey-Smith's general approach demonstrates a subtle, but important, issue. Rather than trying to isolate rigid conditions that carve the biological world into distinct lumps, he explicitly incorporates fuzzy notions, partial cases, and multiple viewpoints into his framework. These features don't just recognize multiple levels of organization; they also give us ways of understanding the crucial transitional phases between them.

Sterelny connects the major transitions to evolvability and its evolution, beginning with a framework he has used to investigate multiple channels of inheritance (Sterelny 2000). His framework is related to Godfrey-Smith's ideas in the previous chapter (though Godfrey-Smith outlines some key differences) in that it begins with a minimal notion of the general conditions for evolution by natural selection. Sterelny then outlines three sets of additional conditions for enabling *enriched* Darwinian environments, conditions whose study might illuminate both how novel and complex phenotypes evolve, and why some lineages seems so much better at it than others. These *evolvability conditions* include anti-outlaw conditions, stability conditions, and conditions on the generation of variation. He then revises this framework by addressing two crucial issues left out of his original formulation: phenotypic plasticity and the interaction between the properties of individual developmental systems and the properties of populations of evolving individuals. These factors are often neglected in accounts of evolvability (such as Wagner 2005). Sterelny then uses this framework to outline a three-pulse model of the conditions that make the evolution of complexity possible, beginning with the establishment of fully equipped cells, continuing with microbial evolution, and finally a phase that includes multicellular development. The outline is a sketch, of course; the details are difficult to nail down. The aim is only to put the framework into action, showing how the interactions between population-level and individual-level properties are important to understanding the evolution of biological complexity.

References

Mayr, E. (1961). Cause and effect in biology. *Science* 134: 1501–1506.

Sterelny, K. (2000). Niche construction, developmental systems and the extended replicator. In R. Gray, P. Griffiths, and S. Oyama (eds.), *Cycles of Contingency* (pp. 333–350). Cambridge, MA: MIT Press.

Tinbergen, N. (1963). On the aims and methods of ethology. *Zeitschrift fur Tierpsychologie* 20: 410–433.

Wagner, A. (2005). *Robustness and Evolvability in Living Systems*. Princeton, NJ: Princeton University Press.

1 The Miscellaneous Transitions in Evolution

Daniel W. McShea and Carl Simpson

In Mark Twain's *Letters from the Earth*, his sardonic alter ego Puddin'head Wilson reflects that if the Eiffel Tower represented the history of the world, and the skin of paint atop the knob at the pinnacle were the portion of that history in which humans have existed, "anybody would perceive that that skin was what the tower was built for. I reckon they would, I dunno" (Twain 1962, 226).

Twain was being facetious, of course, but taken at face value the remark does reflect a common intuition that people are special, that their existence reflects something profound about the evolutionary process, that evolution somehow culminates in them, that they are what evolution at the largest scale—all life over its entire 3.5 billion-year history, the whole Eiffel Tower—is all about. In the spirit of Puddin'head's remark, a modern analyst could choose to scorn this intuition as an expression of our natural human infatuation with ourselves. Alternatively, but equally skeptically, a hard-headed contemporary might treat this intuition as a cultural residue of our two-thousand-year-old obsession with the Great Chain of Being, the notion—dating back to Aristotle—that there is an ordering among organisms, from lower to higher, from monad to man.

But another avenue is open to us. We could, instead, take the widely shared impression of an ordering seriously. We could conjecture that the Great Chain represents a direct but difficult-to-articulate insight—perhaps partly confused, or perhaps only partly correct—into a true natural ordering of some kind. Of course, a modern Great Chain would include a time component, identifying not just a set of organisms but a set of transitions: bacterium to protist to multicellular animal, or more recently, within chordates, fish to reptile to mammal to human. In other words, we could take seriously the idea that an updated Great Chain reflects an actual ascent of some kind, an increase in some objective and important property or quantity over the history of life. And we could then investigate to discover what that property is. Our project would be to discover what we will call the "theoretical unity" that links the transitions leading to people. So, for example, we would ask what is the variable that increases in the transitions from fish to human? On what scale do reptiles score higher than fish and lower than mammals? Is it intelligence, energy intensiveness, or maybe fitness? The search would be open-ended. The goal would be to determine what it is that our

intuition has grasped, to figure out how to say, in scientific language, what we already think we know prescientifically. If the Great Chain is real, even in part, what is it that is increasing as we move up it?

In this project, testing would be crucial. We would want to operationalize each of the candidate variables, and then to make some measurements. Probably most candidates would turn out to be false leads in the sense that they do not produce the right trajectory, that is, they do not increase consistently up the Great Chain. Or they might fail in not placing people at the top. For example, body size is a candidate variable, increasing over at least the early transitions. But it fails at the end because people are smaller than blue whales (and many other mammals). In this approach, many variables might have to be considered before we found one that actually did increase in a way that captured the central intuition. But finally, when we have located the right variable, we could proceed to investigate the dynamics of the trend, its causes, its various exceptions, and so on.

Alternatively, there is another project we could pursue, one that is more in keeping with the spirit of Puddin'head's remark. We could set aside the Great Chain and our intuition that people are special. Then, turning to evolutionary theory, we could ask what variable we *expect* to increase over the history of life. Again, the trend might be noisy, the pattern imperfect. And if that variable can be operationalized, we can then investigate its pattern of change. In this approach, there is no intuited a priori set of transitions to be connected somehow, no trend with humans at its endpoint. Instead, beginning with some variable chosen based on theory, we would determine *empirically* what the major steps upward have been. If theory predicts that maximum body size should increase in evolution, say, on account of the advantages of large size, then we would investigate to discover whether or not the predicted trend occurs, and if it does, what are its properties. We could do this independently for any number of variables—perhaps fitness and complexity, as well as body size—that are predicted to increase in evolution at the large scale. In this project, the question is not about the Great Chain, but about directionality more generally: "What is it that is increasing over the history of life?"

In this project, unlike the first, we would be undaunted by the discovery that humans are not the culmination of a trend. Indeed, this project leaves open the possibility that we are not the culmination of a trend in any variable, at least not in evolution at large scale. (At smaller scales, of course, many species arising later in time will turn out to be local maxima in some variable or other, if only because the evolutionary branching process is Markovian, making humans—and indeed every other species—the local maximum of *some* sort, in *some* variable.)

The Great Chain is in bad odor these days, at least officially, which would make it easy to scorn the first project as unscientific. But we think it is not. For one thing, our prescientific intuitions could be right. For another, there is nothing wrong with focusing on people and the steps leading to us. We are interesting to ourselves. And there is nothing unscientific about pursuing that interest, about seeking the evolutionary sources of human nature

in deep time. It would be of considerable general interest to know whether there is some variable that is changing directionally over the grand sweep of evolutionary time, some trend that makes humans—or something like us—likely. Of course, in a modern discussion, our notion of a trend would have to be updated. Evolution is not a linear ascent. Any trend, however central to the process as a whole, will experience local reversals and be subject to numerous exceptions. But the primary intuition underlying the Great Chain could be right. If so, it would be important to show that evolutionary theory predicts, however roughly, the series of transitions it represents. Such a project could be partially revisionary. Our prior, intuitive conception of the scale might have to be revised during the investigation.

It would also be easy to laud the second project as a kind of scientific ideal. What could be more in keeping with the spirit of objective inquiry than a priori agnosticism about the standing of humans, combined with dismissal of the Great Chain? But there is a concern that needs to be addressed here. Behind an official agnosticism, one suspects an agenda of demoting the importance of people, an agenda that could easily bias the investigator against a finding of a trend culminating in humans, even if one had actually occurred. And therefore in this project, the investigator would need to be careful to let the data speak, to leave open the possibility that humans might really be the culmination of a large-scale trend, at least for certain variables (perhaps, say, ability to control the environment, if it could be operationalized). Properly understood, the main point of this second project is not to understand the evolution of humans at all. But it does not rule out any particular finding about us. Insofar as this project would concern us at all, the point would be simply to find out objectively, neutrally, where we stand.

Two Projects

A possible example of the first project is Francisco Ayala's 1974 treatment of biological progress. Ayala is interested in what he calls "a 'ladder of life' rising from amoeba to man" (Ayala 1974, 339), an updated Great Chain (although he does not use this phrase). His project seems to be to examine a number of candidates for the critical variable that underlies ascent up the ladder, including increasing adaptedness and the accumulation of genetic information, ultimately rejecting them in favor what he calls "ability to obtain and process information about the environment" (Ayala 1974, 349). On this scale, Ayala argues, animals generally score higher than plants, vertebrates higher than invertebrates, mammals than reptiles, and humans than all other species. The match to the Great Chain would seem to be very good.

Now there are reasons to be skeptical of Ayala's result. He has not found a way to operationalize "ability to gather and process information" in a way that would make them objectively measurable. His assessments are impressionistic. But there is nothing wrong

with his project, even by modern standards, insofar as it represents an attempt to find the key variable that underlies an updated Great Chain sequence. It is a bit like the project of finding a formula underlying a sequence of numbers that we have prior reason to think are the result of some underlying process, such as 2, 3, 4, 8, 14, 21, 34, 52, which is very nearly a Fibonacci series. Both for evolution and for a number sequence, the search for an underlying formula seems scientifically reasonable, provided we are able to test our hypotheses objectively, and provided, too, we are prepared to give up a preferred hypothesis that on close study is discovered to fit the sequence poorly.

An example of the second project is George Gaylord Simpson's chapter on evolutionary progress in his mid-twentieth-century, semipopular *The Meaning of Evolution* (Simpson 1967). Essentially, Simpson was seeking the key feature of organisms that accounts for what he thought of as the obvious directionality in the history of life. He considered a number of possibilities, including adaptedness, efficiency, specialization, independence from the environment, control over the environment, complexity, energy intensiveness, and others. In the end, the project failed, and he despaired of finding a single variable (aside from the general tendency for life to expand), concluding that there was not one but many sorts of progress. Significantly, and consistent with his overall strategy, Simpson began his treatment by recognizing the potential for anthropocentric bias in such an investigation, but admonished us not to reject out of hand the possibility that humans may rank highest. He considered our status, like that of every other species, an open question, something to be discovered in the course of investigation. In this second project, the important species and the transitions to them are the outcome of the investigation, not—as in the first project—the standard by which the success of the investigation is to be judged.

Theoretical Unity versus History as One Damn Thing after Another

The two projects are very different, the first focusing on the Great Chain and the second on the history of life, ignoring the Great Chain. But they have something in common, namely, a search for theoretical unity, for a common thread running through the history of life. They share a hope that the search for a common thread will reveal something about the evolutionary process, about its robustness perhaps, its repeatability, or the nature of the forces guiding it. The first project starts with an intuited series of transitions and asks what is the theoretical unity behind them. The second starts with a theoretical unity—some variable predicted to increase by evolutionary theory—and asks what series of transitions (if any) it predicts.

Theoretical unity is a big part of what makes history interesting. History becomes much more intellectually satisfying when we can explain a series of apparently disparate events by citing a common underlying cause. Consider the following major events in mid-twentieth-century world history: the overthrow of the democratically elected government of Iran in 1953, the building of the Berlin Wall in 1961, the Soviet invasion of Czechoslo-

vakia in 1968, the U.S. involvement in Vietnam in the 1960s and 1970s, and the Soviet war in Afghanistan in the 1970s and 1980s. All of these events are explained by a common cause: the escalating Cold War between the United States and the Soviet Union from the end of World War II until the collapse of the Soviet Union. In other words, the Cold War provides the theoretical unity that explains and makes sense of an otherwise disparate series of events.

However, not all historical sequences have this unified structure. Let's move to a smaller time scale: I leave my house in a hurry one morning, forgetting my bag lunch at home. So for lunch I go to a restaurant instead, there by chance encountering a colleague who apprises me of a talk on campus later that afternoon. The talk is to be held in a building on a part of campus that is distant from my office, so I drive there, and after the talk head directly home, which requires me to take a different route home than usual. On my way home by this unusual route, I run over a rake on the highway puncturing two tires. This was an eventful day. Each event is directly connected to and depends on the one before it. Each is a necessary part of the explanation of my arrival home in a tow truck, with my incapacitated car riding atop, and alop, on its flatbed.

Now these events are causally connected and dependent on each other. But there is no theoretical unity. Unity does not demand a single unifying factor. Our Iran to Afghanistan macro-narrative would be unified even if the narrative depended on, say, an interaction between the Cold War and world population growth. But in the story of this eventful day, there is no unified analysis even of this more complex kind. Notice too that there are no general lessons to be learned from this story, say, about the importance of not forgetting one's lunch or about not taking unusual routes home. After all, forgetting my lunch could just as easily have eventuated in me finding a $20 bill on the sidewalk outside the restaurant, and no punctured tires later. From this history we learn only what happened to me that day, which while fascinating (and frustrating) to me, entertaining to my family over dinner, and briefly amusing to my friends perhaps, contains little of general interest. Each event has its own unique cause. Of course, history of this sort does have a sort of fascination, of the kind offered by certain adventure stories, page-turners that are hard to put down because of the rapid flow of improbable events. We gobble up such stories like we eat peanuts, never completely satiated and not at all edified. This is history without theoretical unity, history as a series of miscellaneous transitions, history as Henry Ford saw it, "one damn thing after another."

Of course, it could be that real life is really like this. Many historians study history under the assumption that there are no general principles. In biology, Gould (1989) has defended a view of evolution as the product of chance, with no governing large-scale regularities, at least in the features of organisms. And this view could be right. But surely it is too soon to decide that. In any case, both of the projects outlined above start with the view that the search for regularity is worthwhile, that there could be some theoretical unity in the history of life, and that the goal of discovering it, if it is there, is worthy.

The Miscellaneous Transitions

Since the mid-twentieth century, most studies of large-scale directionality have involved one of the two projects, or a combination of them. Many of these have revolved around the idea of progress. We have already mentioned Ayala's and Simpson's treatments. There is also Vermeij's (1987) argument that the history of life is characterized by organisms with ever-greater energy intensiveness, Van Valen's (1989) suggestion that absolute fitness increases, Knoll and Bambach's (2000) suggestion that what is increasing is diversity and occupation of ecospace, and others (see lists in McShea 1998 and Rosslenbroich 2006). In some cases, it is clear that the project is of the second sort, that the variable of interest arose from theory (e.g., absolute fitness). In others, it seems likely that the project is of the first sort, an attempt to find the variable underlying a set of preconceived transitions (e.g., Ayala). But all share the virtue of searching for theoretical unity.

However, there have also been some instructive exceptions, and we briefly outline three of them here. The first appears to be a case where a theoretical unity was sought, and almost—but not quite—found. (As it turns out, however, this project might be retrospectively salvageable.) The last two seem to us to be failures, cases in which theoretical unity was sought only halfheartedly, if at all. In both cases, none was found, not surprisingly. These projects do not seem salvageable.

Huxley and the Basis for "Higher" and "Lower"

In a chapter on evolutionary progress in his mid-twentieth century, semipopular *Evolution in Action*, Julian Huxley (1942) sought the variable that underlies our notions of "higher" and "lower." His goal was to find a theoretical unity, that is, to find the variable that changes directionally in evolution as higher organisms evolve from lower. His treatment was not explicitly about the Great Chain, but "higher" and "lower" are key words in Great Chain discourse, and it is clear throughout that the transitions he was seeking to explain are the standard ones—the more complex crustaceans succeeding the trilobites, the jawless fishes succeeding the marine arthropods, followed by the amphibians, reptiles, and so on. Huxley's Great Chain branched a great deal, as any modern version must, with insects pursuing their own ascent from higher to lower in parallel with the chordates, for example. But the location of people at the top, as the most dominant species of all time, was unquestioned and indeed was a big part of what was to be explained.

Huxley proceeded by considering two candidate variables, ultimately rejecting both of them: ability to survive (rejected because the organisms we recognize as higher seem not to be especially extinction resistant) and complexity (rejected because many lower organisms are quite complex). He then considered and accepted a variable he called "dominance," and argued that dominant groups (i.e., higher groups) might be those with traits that give them greater control over and independence from the environment. In the Paleozoic, large predatory arthropods, eurypterids, were more powerful swimmers than the tri-

lobites they evolved from. And later fish were more efficient swimmers than their earlier relatives. Both of these represent increases in a kind of environmental control, in the ability of these aquatic animals to manipulate or make use of the medium in which they live. And humans, of course, are the apotheosis of environmental control. In other cases, however, it is not environmental control that increases from "lower" to "higher" but internal homeostasis, that is, independence from the environment. Lower marine animals, he argues, are in diffusional equilibrium with the surrounding sea water, whereas fish are able to regulate their internal chemistry somewhat. The evolution of a shelled egg liberated reptiles from their ancestor's dependence on water in early development, and endothermy in birds and mammals produced some measure of independence from temperature variability in the environment.

If these examples, and others that Huxley cites, are granted, it would seem that he has found not *one* but *two* variables that increase up a modern Great Chain—control over the environment and independence from the environment—with some transitions marked by one and some by the other. And so we think we must judge this result a failure in the search for theoretical unity. In making this judgment, we do not deny that both environmental control and independence could be important in the history of life. Nor do we deny that one or the other underlies all of the major transitions picked out by the Great Chain, some transitions marked by increase in environmental control, some by increase in independence from the environment, and some by both. But his failure to find a single variable that underlies all transitions implies that "lower" and "higher" mean different things in different contexts, that the Great Chain is not unified. In effect there are at least two Great Chains, one for control and one for independence. This could be the case, a fact of the world, but we accept it at the price of rejecting, or at least revising, the intuition that motivated the project in the first place.

On the other hand, Huxley's result may yet be salvageable, if only we could find a way to unify control and independence. Is there some single factor that these two variables represent alternative manifestations of? The answer could be no. It's easy to see an exoskeleton, a shelled egg, or life cycle with resting-cyst stage as ways to achieve some degree of independence from the external environment. But it is difficult to see them as control over the external environment in the same sense in which, say, a beaver or a human building a dam is. On the other hand, both control and independence have to do with the organism-environment relationship, and more specifically with the ability of organisms to develop and function under conditions of their own making. Somewhere in that concept there may be a single variable that underlies the ascent from lower to higher (and, indeed, Laland, Odlong-Smee, and Feldman [2001] treat these as different aspects of so-called niche construction).

It is worth noting here that some promising conceptual work along these lines has been done by Rosslenbroich (in preparation; see also Rosslenbroich 2005, 2006). In particular, he develops a notion of organismal "autonomy," and shows in a compelling way how

autonomy seems to have increased in each of the transitions along a standard Great-Chain-like sequence, bacterium to protist to multicellular individual, and so on.

Stebbins and the Eight Major Levels of Organization

The results of Ledyard Stebbins's 1969 study of progress are less promising. Stebbins identifies eight "major levels of organization" in evolution. How are these levels to be understood? He wrote:

> In the long run, organisms repeatedly have evolved new ways of exploiting . . . environments. In doing so, their bodies have from time to time evolved new levels in the hierarchy of complexity from macromolecule to organelle, cell, tissue, organ, and organ system. Achieving these levels required the accumulation of new genetic information, concerned largely with the integration of development and metabolism and with regulating the translation of genetic information into form and function. (Stebbins 1969, 29)

And here is his list of levels (Stebbins 1969, 30):

1. Earliest self-reproducing organic systems (free-living viroids, none still living).
2. Surrounding cell membrane with selective permeability and active transport of metabolites (prokaryotes).
3. Division of labor between nuclear, cytoplasm organelles (flagellates, other protozoa (eukaryotes)).
4. Multicellular organisms with some cellular differentiation (sponges, algae, fungi).
5. Differentiated systems of organs and tissues (coelenterates, flatworms, higher plants).
6. Organized central nervous system, well developed sense organs, limbs (arthropods, vertebrates).
7. Homeothermic metabolism (warm blood) (mammals, birds).
8. Dominance of tool using and conscious planning (man).

Let us ask just what is increasing here. Stebbins uses the term "complexity," which he seems to understand as hierarchy, the number of levels of organization, or parts within wholes. Earlier in his book a short section describes the hierarchical structure of a simple muscle fiber, starting with a whole muscle tissue and descending in a series of steps through fiber group, single fiber, fibril, and down to a single actin-myosin subunit. Most of his list of eight levels makes sense if complexity is understood in precisely this way, as nested objects within objects or, in evolutionary terms, as the progressive origin of ever higher levels of aggregation. Certainly the transition from level 2, prokaryotic cell, to level 3, solitary eukaryotic cell, can be understood in this way, the eukaryotic cell having arisen historically as an association between (at least) two prokaryotes, an archaebacterium and a eubacterium. Likewise, the level 3 to level 4 transition—solitary eukaryotic cell to multicellular eukaryote—is obviously the origin of a new level. The next transition, from 4 to 5, is somewhat problematic, in that although a new level arises, it does so in a different way than in the earlier transitions. Tissues and organs arise not by aggregation of lower-level entities but by interpolation, so to speak, between existing levels, between the level of the multicellular whole and the level of the cell. Still, hierarchical structure does increase.

So far so good (or at least, good enough), but in the 5-to-6 transition, the scheme breaks down. Central nervous systems, sense organs, and limbs are just organs and organ systems, the same sort of entities that arose in the transition from 4 to 5. Undoubtedly, there is something special about them, but it is surely *not* that they represent either the addition or the interpolation of a new level. The same goes for the transition from level 6 to 7. Homeothermy may be an advance in some sense—for example, in Huxley's sense, providing increased independence from the environment—but it adds no new level of hierarchy. And as for the evolution of humans, it could be argued that they represent a higher level of nesting than a solitary multicellular individual, because we are social, and sociality is the aggregation of lower-level individuals. (And in that case, sociality should replace sense organs and limbs as level 6, and the list should end there.) But corals are also social, consisting of multiple multicellular polyps, and so are many insects and vertebrates, forming societies consisting of multiple multicellular individuals. Therefore, if the criterion for new levels is hierarchy, there is no obvious reason to pick humans as representative of the social level, tool use and conscious planning notwithstanding.

Stebbins left some wiggle room in his understanding of levels, with his mention of accumulation of genetic information, integration of development and metabolism, and increasing regulation of form and function. Possibly his claim is that the evolution of brains, homeothermy, and humans—levels 5, 6, and 7—involved increases in all or some of these. But these are things that he says are *required* for the evolution of new levels of organization, not constitutive of or definitive of them. Thus, even if these things could be shown to have increased in the last transitions, it would still remain to be shown the sense in which they constitute increases in hierarchy. Stebbins did not do so, and it is not at all obvious how it could be done.

On its face, Stebbins's list of eight levels looks like an attempt to justify the Great Chain, to find the variable that underlies it, project 1. If so, then this worthy project failed, because no common variable was found. On the other hand, suppose that his project was really to trace the trajectory of levels of organization wherever it would have led him, that is, project 2. But in that case, his project also failed, in that he abandoned the notion of levels at the higher levels. In particular, humans do not seem to be hierarchically above other social multicellulars. Either way, no theoretical unity has been found. If this were the only way of reconstructing history, it would be revealed to be a series of miscellaneous events, on the long road to human beings, one damn thing after another.

Maynard Smith's "Levels of Selection" and Maynard Smith and Szathmáry's "Major Transitions"

Two 1995 publications by John Maynard Smith and Eörs Szathmáry—a book and a *Nature* paper—have attracted considerable attention from molecular evolutionists, macroevolutionists, and philosophers of biology, and have helped propel a long-overdue resurgence of interest in the evolution of hierarchy (Griesemer 2001; Jablonka 1994; McShea 2001; Michod 1997, 1999; Michod and Roze 1997; Queller 2000). The issue they address is the

evolution of higher-level wholes from lower-level individuals, the emergence in evolution of new and higher levels of selection (Michod 1999; Michod et al. 2006). And they address it by examining what they call the "major transitions" in evolution, identifying eight of them, and discussing possible mechanisms by which they arose.

The *Nature* paper contains the clearer general statement of their project, so we will focus on that. It begins by directing the reader to a list of major evolutionary transitions in their table 1 (Szathmáry and Maynard Smith 1995, 228):

1. Replicating molecules to populations of molecules in compartments
2. Unlinked replicators to chromosomes
3. RNA as gene and enzyme to DNA and protein (genetic code)
4. Prokaryotes to eukaryotes
5. Asexual clones to sexual populations
6. Protists to animals, plants and fungi (cell differentiation)
7. Solitary individuals to colonies (non-reproductive castes)
8. Primate societies to human societies (language)

It then goes on to say:

> There are common features that recur in many of the transitions: (1) Entities that were capable of independent replication before the transition can only replicate as parts of a larger unit after it. For example, free-living bacteria evolved into organelles. (2) The division of labor: as [Adam] Smith pointed out, increased efficiency can result from task specialization. . . . For example, in ribo-organisms nucleic acids played two roles, as genetic material and enzymes, whereas today most enzymes are proteins. (3) There have been changes in language, information storage and transmission. Examples include the origin of the genetic code, of sexual reproduction, of epigenetic inheritance and of human language. (Szathmáry and Maynard Smith 1995, 227)

There follows a short section about complexity, especially genetic complexity. Unfortunately, there is no clear statement of how complexity is to be understood, and the connections among complexity, the major transitions, and the common features of the transitions are never discussed. Their discussion has a second theme, having to do with the flow of information across the generations, in particular the notion that the fidelity and bandwidth of inheritance has increased; but again the relationships with complexity and the common features of the transitions are not clear. In any case, complexity and bandwidth aside, it seems clear that their major interest is the first criterion above, the increase in hierarchical structure in evolution and the origins of new levels of selection. Almost all of the subsequent discussion in the literature has focused on this aspect of their project, taking the trend in levels to be its centerpiece. We will read their project this way, too.

This interpretation is consistent with three pieces of evidence: First, a discussion follows immediately of the first shared feature of the transitions, namely, "that entities capable of independent replication before the transition can only replicate as parts of a larger whole

afterwards." In other words, a higher-level entity arises from an integration of lower-level units, from either an ecological association or a clonal aggregate of them. And it is fairly clear that Szathmáry and Maynard Smith understand it to arise as the result of higher-level selection.

Second, a central concern in both the 1995 paper and the book is the problem of cheaters, lower-level individuals that pursue their Darwinian self-interest at the expense of the higher-level whole. The question raised is how are higher levels able to persist when selection would seem to favor cheaters that undermine them? To say this question has been focal in the literature on levels of selection would be an understatement. Interest in it has been almost obsessive, to the point that other questions have been nearly ignored (cf. Calcott 2008).

Third, Maynard Smith published a paper in 1988 that is undoubtedly an intellectual precursor to his part of the 1995 paper and book. And that earlier paper was explicitly devoted to the increase in "complexity" in evolution, this time more clearly conceived as the origin of new levels of selection. He also offered a table that is remarkably similar to the 1995 table. The earlier table differs in a few ways, notably in the identification of the penultimate level as occupied by "demes," as well as by social groups, and in the designation of an extra level—the species level—between demic-social and human culture. The inclusion of demes and species confirms the diagnosis. Maynard Smith understood the increase in complexity in the conventional way, as an increasing trend in hierarchy, understood as parts within wholes (Eldredge and Salthe 1984).

Assuming the conjecture is true, that the major transitions are to be understood mainly as increases in hierarchy, the decision to include human society as the eighth and last transition is a departure, a violation of Maynard Smith's own criteria for what the sequence of transitions represents. A human society would seem to be just that, a society, not an even higher level of selection. It would seem to occupy the same hierarchical level as a social insect society (achieved already in the seventh transition) or a primate society (present already at the start of the eighth). Each human is a multicellular eukaryotic individual, and a human society would appear to be just another association of multicellular eukaryotic individuals. To be sure, our societies are different in detail from ant and baboon societies. Ours are also unique in a number of ways. But social organization in every multicellular species is different in detail from every other, and more different with greater taxonomic distance. Likewise, every species is unique. But that does not make us a higher level.

What is the next highest level, the level above the social? From first principles, it would seem to be an association of societies, a metasociety or supersociety of some sort. Now among nonhuman animals, at least, there seem not to be any supersocieties, either extant or in the fossil record (McShea and Changizi 2003), at least none that are individuated to the extent that organisms or even societies are. There is, in the Recent, an invasive species of ant in which clusters of colonies can be identified, but there is no division of labor among colonies as there is among well-individuated members of colonies at the next level down.

What about humans? Identifying levels in human societies is problematic, because—at least in modern societies—we associate in so many different ways, as members of so many different social units, with no clean hierarchical structure to them. Associations are crosscutting, so that a member of a village can also be a member of a guild that includes people from a number of villages, and also a member of a family or kin group that seems to crosscut both. This does not rule out the possibility that humans form supersocieties; it simply means that there is some conceptual and empirical work to be done—finding ways to assess and locate hierarchical structure—before such a higher level can be claimed.

Another tactic might be to argue that our societies occupy the same level as ant and baboon societies but are more individuated at that level. In other words, all or most animal societies occupy the same level of selection, but in the human case, selection has operated more powerfully, more efficaciously, to produce greater individuation, evidenced perhaps by our greater division of labor among individuals, the development of intermediate-level associations such as teams and groups (the equivalent of tissues and organs), and so on. In other words, the suggestion is that humans are social just like ants and baboons, but that we are more *intensely* social, that we occupy the social level more fully. Again, arguing this requires some work, if only because, intuitively, the eusocial insects seem to be *more* intensely social than people, at least on account of having castes marked by phenotypic, as well as behavioral, differentiation. More generally, ants do seem to be more committed to social living than people.

There are other ways to try to argue this. One might say that human societies are more complex, not because we occupy the social level more fully, but because our societies involve more interactions than ant societies, more types of interaction, more social roles, and so on. But this is complexity in a very different sense from that being invoked in the rest of the list of major transitions. Unless one could defend a sharp distinction between kin selection and group selection, and argue that only humans are strongly group selected, human social complexity is not complexity in the sense of hierarchy, of higher levels of selection. Alternatively, one might focus on language and culture, as Maynard Smith and Szathmáry did, with both understood as novel modes of information transfer and inheritance. But while the human mode of cultural transmission is unique, it is not obvious that it produces a higher level of selection. No such argument has been made for other epigenetic information transmission mechanisms. For example, diffusible morphogens in animal development are certainly epigenetic, but their role is mainly to organize lower-level units (cells) within a higher-level whole (a multicellular individual). No one would claim that this mechanism produces an even higher individual, above that of the multicellular individual. Likewise, cultural transmission might be said to organize lower-level human individuals within a higher-level society. But it is hard to see how its produces a whole at an even higher level. (It is worth noting that Richerson and Boyd [2001] have argued that cultural transmission explains the between-group differences and within-group similarities that make group se-

lection important in human evolution. But this argument does not lead in any obvious way to a higher-level whole, that is, to a metasociety of any kind.)

In any case, though it might be possible to argue that we occupy the level of selection above other animal societies, or that our societies are more individuated at the social level, Maynard Smith and Szathmáry do not even attempt it. Indeed, in the 1995 paper and book, the ascent from one level of selection to another is claimed only for *many* of the transitions, not for all, in particular for numbers 2, 4, 5, 6, and 7. They seem aware of their theoretical inconsistency, aware that including transition number 8, the transition to humans, requires understanding the notion of a "major transition" in a different sense. It is a change in mode of information transmission, not an increase in hierarchy.

Finally, we note that, consistent with our claim of theoretical disunity, the features that separate the transition to human society from the ones just before it (change in mode of information transmission versus increase in hierarchy) have had different and independent impacts in the literature. And interestingly, these impacts roughly parallel the two projects outlined above. First, the Maynard-Smith and Szathmáry notion of change in mode of information transmission has had some impact on studies of the evolution of human language and culture (e.g., Laland et al. 2001). It fits nicely with current thinking about language and culture as epigenetic, as information-transmission systems that have been built "on top," so to speak, of the older, DNA-based genetic system. And in its strong focus on humans, this work is somewhat allied to project 1. The issue in this area is not hierarchy, or the common themes of the evolutionary process, generally. Instead it is that last and very special transition to humans. But the major impact has been on thinking about hierarchy, evident especially in the work of Michod (e.g., 1997, 1999) on what he calls evolutionary transitions in individuality and on how these transitions come about. This work is strongly allied to project 2, the search for large-scale directionality, with no particular focus on humans. For the trend in individuality, humans are relevant but they are definitely not the zenith.

In sum, we cannot find any theoretical unity in the Maynard Smith and Szathmáry list of major transitions. The list needs revision, or the project (or projects) needs to be redefined, and not in terms of hierarchical complexity. One could argue that theoretical unity is not their goal, that they are being held to an inappropriate standard. But then we are entitled to ask, what is the point of collecting these transitions together? Can it be just their importance as milestones on the road to us? In that case, the major transitions story would be like a "story of my day," how it all started with the forgetting of a lunch and how it ended with a trip home in a tow truck. It would be the story of how life began, some 3.5 billion years ago, what happened next, and next, and next. It is a story that takes us from the origin of life (genes, DNA) to the transitions in level of selection (bacterium, protist, multicellular individual, colony) to the origin of human beings (in particular, our language), a series of miscellaneous transitions with no obvious unity beyond their salience for us.

It could be, of course, that humans are special, independent of whether or not we are a higher level of selection. That skin of paint atop the Eiffel Tower could really be the most

interesting feature of the tower. (Is that skin of paint a different color from the rest of the tower? Is there a transmitter of some kind embedded in it?) And yet it could still be that no theoretical unity underlies both us and the other "major transitions," however defined. But in that case, treatments such as those of Stebbins and Maynard Smith and Szathmáry are not just failures to find a theoretical unity, they are distractions. To force humans into the same theoretical box with other transitions is to overlook whatever it is that actually makes us special.

Afterthoughts

There is something philosophically muddled and scientifically casual about these three treatments of evolution at the largest scale. And we would argue, somewhat impatiently, that the time for getting serious about evolution at the largest scale is long past. We have had the tools for understanding large-scale trends for decades. And for much longer, we have demanded theoretical consistency in the study of trends. No one in modern biology would study the evolution of body size in any taxonomic group without adopting a single, operational definition of body size and applying it in a consistent way to all specimens considered. And there is no reason to settle for a lower standard in our treatment of the history of life at the largest scale. To be clear, the villain has not been the Great Chain, for some version of it could reflect a real insight, and it could be treated seriously. Neither has the problem been the notion that the history of life might be governed by chance, that no unity in fact exists, because that too is a possible truth of the world. The fault has been the yielding to theoretical inconsistency. Enough is enough.

This discussion may seem uncharitable, especially in its treatment of Maynard Smith and Szathmáry, whose work has been so well received. So let us add that many aspects of the Maynard Smith and Szathmáry study may be real contributions, especially in the insights it offers into the possible mechanisms underlying some of the more puzzling big events in early evolution, such as the evolution of genetic code. And considerable praise is due to everyone who takes on the daunting project of searching for pattern at the scale of life's history. The goal of finding a unity in that history hovers above evolutionary discourse as the moon once did above Earth, inviting the bold to explore the limits of what we can know and learn. Whatever the faults of their findings, explorers in this area deserve credit for their audacity. If they deserve any blame, it is only for a last-minute faintness of heart, for conceding failure too easily after having boldly traveled so far.

Acknowledgments

This work was supported by a grant from the Cambridge Templeton Consortium.

References

Ayala, F. J. (1974). The concept of biological progress. In F. J. Ayala and T. Dobszhansky (eds.), *Studies in the Philosophy of Biology* (pp. 339–355). New York: Macmillan.

Calcott, B. (2008). The other cooperation problem: Generating benefit. *Biology and Philosophy* 23: 179–203.

Eldredge, N., and S. N. Salthe. (1984). Hierarchy and evolution. In R. Dawkins and M. Ridley (eds.), *Oxford Surveys in Evolutionary Biology*, Vol. 1. (pp. 184–208). Oxford: Oxford University Press.

Gould, S. J. (1989). *Wonderful Life*. New York: W. W. Norton.

Griesemer, J. (2001). The units of evolutionary transition. *Selection* 1(1): 67–80.

Huxley, J. (1942). *Evolution in Action*. New York: Harper.

Jablonka, E. (1994). Inheritance systems and the evolution of new levels of individuality. *Journal of Theoretical Biology* 170(3): 301–309.

Knoll, A. H., and R. K. Bambach. (2000). Directionality in the history of life: Diffusion from the left wall or repeated scaling of the right? *Paleobiology* 26(suppl.): 1–14.

Laland, K. N., J. Odling-Smee, and M. W. Feldman. (2001). Niche construction, biological evolution, and cultural change. *Behavioral and Brain Sciences* 23: 131–146.

Maynard Smith, J. (1988). Evolutionary progress and Levels of selection. In M. Nitecki (ed.), *Evolutionary Progress* (pp. 219–230). Chicago: University of Chicago Press.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution*. New York: Oxford University Press.

McShea, D. W. (1998). Possible largest-scale trends in organismal evolution: Eight "live hypotheses." *Annual Review of Ecology and Systematics* 29: 293–318.

McShea, D. W. (2001). The minor transitions in hierarchical evolution and the question of a directional bias. *Journal of Evolutionary Biology* 14(3): 502–518.

McShea, D. W., and M. A. Changizi. (2003). Three puzzles in hierarchical evolution. *Integrative and Comparative Biology* 43: 74–81.

Michod, R. E. (1997). Evolution of the individual. *American Naturalist* 150: S5–S21.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality.* Princeton, NJ: Princeton University Press.

Michod, R. E., and D. Roze. (1997). Transitions in individuality. *Proceedings of the Royal Society of London Series B*, *Biological Sciences* 264(1383): 853–857.

Michod, R. E., Y. Viossat, C. A. Solari, M. Hurand, and A. M. Nedelcu. (2006). Life-history evolution and the origin of multicellularity. *Journal of Theoretical Biology* 239(2): 257–272.

Queller, D. C. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions: Biological Sciences* 355(1403): 1647–1655.

Richerson, P., and R. Boyd. (2001). Built for speed, not for comfort. *History and Philosophy of the Life Sciences* 23: 423–463.

Rosslenbroich, B. (2005). The evolution of multicellularity in animals as a shift in biological autonomy. *Theory in Biosciences* 123: 243–262.

Rosslenbroich, B. (2006). The notion of progress in evolutionary biology—the unresolved problem and an empirical suggestion. *Biology and Philosophy* 21: 41–70.

Simpson, G. G. (1967). *The Meaning of Evolution*. New Haven, CT: Yale University Press.

Stebbins, G. L. (1969). *The Basis of Progressive Evolution*. Chapel Hill: University of North Carolina Press.

Szathmáry, E., and J. Maynard Smith. (1995). The major evolutionary transitions. *Nature* 374: 227–232.

Twain, M. (1962). *Letters from the Earth*. New York: Harper & Row.

Van Valen, L. M. (1989). Three paradigms of evolution. *Evolutionary Theory* 9(1): 1–17.

Vermeij, G. (1987). *Evolution and Escalation*. Princeton, NJ: Princeton University Press.

2 Alternative Patterns of Explanation for Major Transitions

Brett Calcott

Two central claims in Maynard-Smith and Száthmary's book are that (a) some events in evolutionary history are special because they changed what was subsequently possible to evolve, and (b) these events share many similar properties. The goal in this chapter is to explore this second claim, to assess what these claims of similarity amount to, and how and why we can make them.

My goal is different from that of McShea and Simpson (this volume), who look for similarity in the *outputs* of these transitions; they wonder, for example, whether all the transitions increase hierarchical complexity. Instead, I shall focus on similarities in the mechanisms responsible for bringing about the transitions identified by Maynard-Smith and Száthmary. Much of the subsequent literature has identified *the* common mechanism as an especially potent form of multilevel selection; potent enough to suppress the perennial threat of defection. The main point of this chapter is to highlight additional ways that these transitions might be unified by the causal mechanisms that explain them. Though not widely acknowledged, Maynard-Smith and Száthmary identified a number of similarities across the major transitions in addition to the problem of differing levels of selection (Maynard Smith and Szathmáry 1995, 12). The arguments I give here suggest that exploring these—and other possibilities—would provide a richer understanding of the major transitions.

To make good on the project, I need to outline a general model of explanation and show how, in biology, distinct explanatory projects coexist and can complement one another. I first show that claims of similarity across the transitions are best understood as *broad explanatory generalizations*. The properties that are similar pick out key factors that enabled particular transitions to occur, yet they do so in a way that remains abstract enough to apply across diverse events. In doing so, they both explain and unify the recurring pattern of transitions.

I then join this observation with a familiar idea: that there are different kinds, or *patterns*, of explanation in biology. Some well-known ways of carving these up are Mayr's proximate and ultimate explanations,[1] and Tinbergen's four questions (Mayr 1961; Tinbergen 1963). These distinctions capture an important fact: Even when we identify a particular

biological phenomenon, we can still go about explaining it in a number of ways. I show that different patterns of explanation are applicable to the kinds of evolutionary change identified as major transitions, too. I do this by examining three explanations for a single transition: the evolution of multicellularity in *Volvox carteri*.

Together, these two ideas show that there are multiple ways of identifying similarities that unify the various major transitions. Once alternative explanatory options are laid out, it is clear that the work on the major transitions concerning cooperation and the levels of selection has focused on a single pattern of explanation, while others have largely been ignored. I suggest that a richer account of major transitions is possible if we deploy multiple patterns of explanation, as these different explanatory hypotheses can interact and mutually constrain one another.

I begin the chapter with a brief summary of a manipulationist account of explanation, which is well suited to capturing how many explanations work in biology (Hitchcock and Woodward 2003; Woodward 2003; Woodward and Hitchcock 2003).[2] This preamble provides a unified foundation for thinking about explanation, for, although there are different patterns of explanation, they all share a core structure.

Explanatory Generalizations: A Short Plausible Account

In this section, I use a simple biomechanical example to examine the structure of explanatory generalizations. Once the basic structure is laid out, it will be clear how it fits other kinds of biological explanation. In the next section I show how it fits explanations for major transitions, too.

Let's say we want to explain how a kangaroo jumps. A typical response might mention a number of factors: how the tail is used as a counterbalance, how the alignment of the enlarged fourth toe with the leg bone serves to drive the jump, and how the elastic tendons in the ankle store and release the energy to aid the jump.[3] Given a *target of explanation* (how the kangaroo jumps), we respond with a set of *difference-makers* (the tail, the toe, and the tendons), and some generalizations about how these difference-makers affect the target of explanation.[4]

Why do these factors explain how a kangaroo jumps? Think what might happen if we changed or "manipulated" any of these features. A shortening of the tail, a misalignment of the central toe, or a weakening of the elasticity of the tendons; all of these changes would reduce a kangaroo's ability to jump. We could construct a detailed biomechanical model—calculating the precise relationships between these features—to ascertain more exactly the outcome of manipulating them. Yet even without such a model, simple verbal explanations pick out the same key properties that, if manipulated, would affect jumping behavior.

So, according to a manipulationist account of explanation, we explain some target phenomenon by showing what it depends on, and by showing the structure of this dependence.

A specification of this structure shows how the phenomenon to be explained (jumping), is changed by various manipulations of other properties (tendons, toes, and tails).

It is worth mentioning some further points concerning how well and how broadly these explanations apply. First, we don't require our biomechanical explanation to tell us how kangaroos jump under all conditions, or to deliver results with perfect precision. So these generalizations connecting the difference-makers and the target of explanation need not be lawlike. We're doing biology here, not physics, and as Woodward and Hitchcock suggest, we should *expect* explanations to vary in their depth, rather than there being a simple dichotomy between lawlike and accidental generalizations (Hitchcock and Woodward 2003).

Second, our explanation is meant to *generalize* across kangaroos.[5] We don't want to explain how a single kangaroo jumps, or how every animal jumps. So these explanations have an implicit *domain of application*: all kangaroos.

Third, we can vary the domain of application. Instead of all kangaroos, we could restrict the domain to just Eastern Grey Kangaroos—the kind you see hopping through the Canberra suburbs at night in winter. We could also broaden the domain to include (say) wallabies as well as kangaroos. So our explanations can become more or less general, depending on what we intend to apply them to.

Fourth, increasing or decreasing the generality in this way may have other implications for our explanation. It is probable that, by narrowing the range to particular species of kangaroo, we can provide a more accurate account of the relationships between the difference-makers and the explanatory target. After all, within-species differences are likely to be smaller than cross-species differences. In contrast, the variations in physiology between wallabies and kangaroos may mean our explanation is less accurate, or we may be forced to exclude some detail, making our explanation more abstract. So there may be trade-offs between the breadth of the domain we choose and depth of our explanation.

So much for kangaroos; what about other biological explanations? This kind of structure—identifying a target of explanation and providing difference-makers—applies equally well across an enormous variety of biological explanations. We explain how certain kinds of speciation occur by citing spatial structure and mating preferences, because changing these factors affects the probability of speciation (Gavrilets 2004). We explain why the Cambrian Explosion occurred by citing an increase in oxygen, or the arrival of some particular constellation of developmental mechanisms, because without these being present, the explosion would not have occurred (Marshall 2006). We explain the origin of eusociality in bees by appealing to kin selection, for the relatedness of colony individuals is the crucial factor that enabled a reproductive division of labor, and subsequent worker specialization (Hughes, Oldroyd, Beekman, and Ratnieks 2008). And we explain the increase in malaria—and subsequent malarial resistance—by citing the introduction of agriculture, because a side effect of agriculture was the provision of increased breeding grounds for malaria-carrying mosquitoes (Afrane et al. 2004; Wiesenfeld 1967). In each case, what counts as an explanation is the identification of some difference-making factor and an understanding of how it (putatively) effects a change in the target of explanation.

Although each of these explanations shares the same basic structure, there are differences between them: The kinds of targets vary, the domain of application varies, and the assumptions about background conditions also differ. I'll look at some of these differences presently. Before doing that, I'll show that much of the work describing and understanding major transitions has this explanatory structure.

Explanatory Generalizations for Major Transitions

One key feature common to many of the major transitions was an increase in the level of biological organization, or the arrival of a new kind of individual. This feature has become so central to subsequent work on major transitions that it is often thought to be definitive (Michod 1999), though Maynard-Smith and Száthmary actually identify "the way that information is transmitted between generations" as the central defining feature (Maynard Smith and Szathmáry 1995, 6). The evolution of multicellularity and the evolution of eusociality are paradigms of this kind of transition in organization. In each case, formerly solitary individuals got together to form some higher-level cohesive unit.

Attempts to explain how these transitions occurred fit well with the structure of explanation I laid out in the last section. The target of explanation is the origin of a new level of organization. The difference-makers are those factors that ensure evolutionary stability of cooperation for lower-level individuals. The stability is central because any transition to a higher level of organization must overcome a problem:

> Why did not natural selection, acting on entities at the lower level (replicating modules, free-living prokaryotes, asexual protists, individual organisms) disrupt integration at the higher level (chromosomes, eukaryotic cells, sexual species, multicellular organisms, societies) (Maynard Smith and Szathmáry 1995)?

Such transitions are made possible by features that solve this problem between the lower-level individuals; allowing integration at the higher level and, in some cases, the arrival of a new kind of individual. A number of factors stabilize cooperation: relatedness, iterated interactions and reciprocity, mutualisms, and punishment or conflict mediation. These are the difference-makers. Identifying them, and showing the effect they have in stabilizing cooperation, explains how a transition in a level of organization—a major transition—could have occurred.

These various factors—such as relatedness and iterated interactions—are thought to underwrite the stability of cooperation because manipulating these factors in simple models can switch the evolutionary outcome from noncooperative to cooperative behavior. These models show how the difference-makers affect whether or not a transition to a new level of organization occurs, just as a biomechanical model of kangaroo locomotion shows how changing various physiological factors would affect a kangaroo's jumping ability.

These models, like many others in evolutionary biology, abstract away from many biological details. For example, in some cases the individuals in the models are meant to represent cells; in other cases, they represent ants or bees. This abstraction allows the models to *generalize* across a broad domain—they are equally applicable to multicellularity as they are to eusociality.

Such simplified models may not explain everything about the transitions. Given they are generalizing across such a broad domain, we might expect them to have traded away some detail for this increase in breadth. Despite this, they identify important similarities across what appear to be very different events, telling us something very general about how these striking evolutionary changes could occur.

Three Tales about Green Algae

In this section, I'm going to ignore the unifying aspect of major transitions (we'll return to this in the next section). Instead, I'm going to pick one specific transition—the evolution of multicellularity in *V. carteri*—and look at three ways that it has been explained. These explanations differ significantly, but they are not (or not obviously) competing explanations. Rather, they form different *patterns of explanation*. I'll show that only one of them investigates the stability of cooperation. This suggests that, at least in *V. carteri*, we can explain something about the transition without referring to cooperation.

Once I've established that there is more than one kind of explanation for this particular transition, I'll return to the problem of unification. Here, I explore whether the patterns of explanation for *V. carteri* that did not mention cooperation might be made general enough to apply to other major transitions.

One reason to study green algae is because they can teach us something about the evolution of multicellularity. A number of related species in the Volvocalean family form a gradation of complexity between single-celled and simple multicellular organisms. The members of this family of algae differ in size, the number of cells they produce, and whether or not there is a split between germline and somatic cells. This split is thought to be central to understanding how a new level of individuality has evolved (though see Clarke, this volume, for a skeptical response to this idea). When a split does occur, there are also differences in the proportion of somatic (dead-end) cells produced. *V. carteri* is one of the more complex species, possessing a distinct split between germ and soma. I'll briefly go through three ways that this increase in complexity has been explained.

Explanation 1: Conflict Mediation

Michod explores the evolution of the split between germ and soma in *V. carteri* with a population genetics model that includes structured interactions (Michod 1999).[6] The model assumes that cells have already established a group-level life cycle. Each group begins

Table 2.1
Possible equilibria in two-locus modifier model

Equilibria	Description	Interpretation
Dm	All cells defect (D) and there is no germline sequestration (m)	Groups not stable because of defection. Single cells persist
DM	All cells defect (D), and germline sequestration (M)	Not of biological interest as it is never stable
Cm/Dm	Polymorphic for cooperation and defection (C/D), no germline sequestration (m).	Groups of cooperating cells, but no higher level functions
CM/DM	Polymorphic for cooperation and defection C/D), with germline sequestration (M)	New individuals: groups of cooperating cells with higher level functions

Modified from Michod, 1999.

with a single cell, which reproduces, and the offspring stay together. Mutations might occur during cell reproduction, so groups may not consist of identical members. The groups eventually split up, with some cells going on to found their own groups.

This model has two genes, each with two possible alleles. The first gene controls whether the cell cooperates (C) or defects (D). Cells that cooperate increase the fitness of all cells in the group, whereas cells that defect increase their own fitness (by self-reproduction) at the expense of others in the group.

The second gene stipulates whether a cell is capable of allocating a germline (M) or not (m). If it is, then after a splitting, one of the cell lineages will continue reproducing within the group, but will die off when the group splits up (the somatic line), and the other will undergo diminished reproduction within the group, but will contribute to subsequent generations of cell groups (the germline).

Michod assumes plausible values for the controlling parameters in the model, and then determines that there are a number of stable equilibria for this model (see table 2.1). Importantly, one of these equilibria includes cells that both cooperate and allocate a separate germline. Michod provides the following reasoning for why the germline split is important. In a cooperating lineage, putting aside a set of slowly reproducing cells for a germline concentrates most defector mutations in the somatic line, who thus have no long-term evolutionary future. So the evolution of a split between germ and soma helps stabilize cooperation, and selects for the evolution of cooperating cell groups rather than individual-living cells.

Michod's model *explains* the transition in *V. carteri* by showing that a split between germ and somatic line prevents the accumulation of defectors in the population. Without such a split, the defectors are more likely to build up and displace any cooperators. The advent of germline allocation is thus a difference-maker, whose manipulation affects the evolutionary trajectory of the population, increasing the probability of a transition to a new level of organization.

As I have noted, the suppression of defection is essential to all transitions to new levels of individuality, so we have a candidate unifying mechanism; perhaps reproductive specialization is a common element in transitions of this kind, and for this reason.

Explanation 2: Generating Benefit

Solari and colleagues provide a different explanation for the germ-soma split, and the transition to multicellularity in *V. carteri* (Solari, Nedelcu, and Michod 2003). To stabilize cooperation, there clearly needs to be a way of preventing defection, and this has been the focus of most work on the evolution of cooperation. But it must also be true that individuals in groups do better than individuals by themselves. This raises a different question about the synergy of cooperation: How is it that group behavior can *generate benefit* (Calcott 2008)? It is this second problem that Solari et al. address, and the germ-soma split is an important part of the solution.

Solari et al. tell us *why* being in a group is better. *V. carteri* benefits because increased size allows more phosphate to be stored in the intracellular matrix. But getting bigger is not a simple matter; cell groups cannot simply increase in size without limit. As they become larger, they hit constraints, and it is these constraints that require a division of labor between germ and somatic cells.

Solari et al.'s model explains *how V. carteri* overcomes these constraints. They identify two constraints; I discuss only the first of these here, as this is sufficient to make my point. Solari et al. dub this first constraint the *flagellation constraint*. The ancestors of *V. carteri* went through two separate life stages. They began their life having flagella, so they could stay afloat. They then lost these flagella, and began to reproduce (by undergoing mitosis). Due to some shared cell mechanics involved in both these processes, it turns out that they must either swim *or* reproduce; they cannot do both simultaneously. As the group of cells becomes larger, it becomes necessary for some cells to specialize in swimming, so that the group, now much larger and taking longer to grow, does not sink. Thus, the benefits of being larger are obtained by a division of labor among the cells.

In Solari et al.'s model, the upper limit on size is possible without somatic specialization. They suggest that data from *V. carteri* and its close relatives map closely to the values in their model, and that this provides some evidence for the model.

Solari et al. *explain* the transition to complex multicellularity in *V. carteri* by showing how an increase in internal complexity (a division of labor) was necessary to enable the group to sustain a larger and more advantageous size. The split between germ and soma is a difference-maker, for, as their model shows, only individuals with a specialized soma can safely continue reproducing long enough to achieve this larger size.

The flagellation constraint is, of course, specific to this example. But it is likely to be an instance of a more general principle: The generation of benefit from cooperation often

depends on a division of labor that includes reproductive specialization, as individuals specialized for reproduction will often be unsuited for somatic tasks.

Explanation 3: Origins of Traits

Kirk describes a twelve-step program for evolving multicellularity and a division of labor (Kirk 2005). These twelve steps are each features on an "evolutionary pathway leading from a unicellular ancestor to multicellular organisms with a division of labor between different cell types." (p299) The steps include the partial inversion of the embryo; the establishment of organismic polarity; a full division of labor between germ and soma; and a bifurcated cell division program.

Kirk's project is quite different from the two previous explanations. He sets out to explain a series of related historical features, asking himself what needed to be added at each stage to produce something like *V. carteri*. In order to reconstruct a plausible series of prior mechanisms, Kirk looks at the properties of the extant, less complex, relatives, treating them as proxies for some common ancestor. The more distantly related the relative, the deeper the shared ancestral function.

Kirk explains the transition to complex multicellularity by enumerating a series of critical differences in developmental function on the pathway between a single-celled ancestor and *V. carteri*. Kirk's project is not merely descriptive, for although part of his goal is to describe a plausible series of stages leading to *V. carteri*, he also isolates plausible difference-makers whose change could have bought about each new functional stage.

For example, the inversion in *V. carteri* requires the gene *invA*, which codes for a protein affecting the microtubules that line certain key cells. This protein changes the relative movement of nearby cells connected by the cytoplasmic bridge. An ortholog of this gene is found in the simpler relatives of *V. carteri*, and in one case, this ortholog can cure the inversionless phenotype of a *V. carteri* mutant.

In each of the stages, Kirk documents the new mechanism and its function. He then identifies a homologous part (often a gene) that is present in a relative that does not possess this function. In some cases, Kirk documents the ancestral function of the homologous part, too. He thus identifies a series of mechanisms, explaining how each works, and showing how one might plausibly be changed into another by a small modification of the mechanism.

I've called such explanations "lineage explanations" (Calcott 2009). They are important because, though it is widely accepted that there is an incremental constraint on evolutionary change, in many cases it is difficult to see exactly how a progressive set of small changes could produce the differences between ancestral and derived phenotypes. Identifying the existence of these small changes does important explanatory work. It explains the origins of new phenotypic functions that, in this case, were essential for the transition to multicellularity.

Notice that Kirk has little to say about *why* these particular changes might have been fitness-increasing, or what kinds of selection regime might have led to their fixation. But his explanations still identify difference-makers, the key changes in individual mechanisms that were plausibly present in the ancestors of *V. carteri*. Without these key additions, *V. carteri* would not have evolved complex multicellularity.

Again, although the specific explanation is unique to the Volvocaleans, the explanatory scheme is both general and of critical importance, as it discharges the assumption of gradual change that underlies a selective explanation, by identifying the ancestral mechanism whose modification could, in fact, lead to the novel adaptive behavior.

Populations, Individuals, and Explanations

Each of the three explanations have the same core structure—providing difference-makers and showing how they affect a target of explanation. But they also differ in important ways. I shall outline one important difference in detail, and then briefly mention some others.

In some biological explanations, the target of the explanation refers to properties of a *population*, such as the stability or trajectory of some trait within that population. The difference-makers pick out those features that may alter this stability or trajectory. The theory of kin selection is like this. It identifies a key property—relatedness—whose manipulation determines whether the evolutionary outcome is a transition to sociality or not. These explanations relate population-level properties, such as average trait fitness or population size, to population-level targets, such as rate of fixation in a population.

In other cases, the target of biological explanations are properties of *individuals*, and the difference-makers pick out what features make possible a particular behavior, development, or physiological function. Notice that the explanation may be applied to many individuals (all kangaroos, for example), but the explanation itself refers to properties of individuals (tendons, tails, and toes).

So, some explanations are about populations, and these explanations generalize over many different populations. Other explanations are about individuals, and these explanations generalize over some class of relevantly similar individuals.[7]

These two types of target provide quite different ways of answering questions, because they demonstrate different kinds of contrasts. The first explains why some population might have evolved the way it did by contrasting different possible evolutionary outcomes. It tells us about the factors that, if changed, would have produced a different evolutionary outcome. The second explains individual mechanisms by contrasting the outcomes that various manipulations could have. It tells us how certain changes would affect the performance, development, or behavior of an individual. We can now see how this contrast applies to the three explanations I gave earlier.

Explanation 1 in the previous section has a population as an explanatory target. It told us what features were important to enable a stable equilibrium in which cooperation and a

Table 2.2
Summary of some key differences in the explanatory patterns used for the evolution of complex multicellularity in *V. carteri*

Explanation	Conflict Mediation	Generating Benefit	Origins of Traits
Target	Population change	Individual differences	Individual differences
Manipulation affect	Population make-up	Production of group benefit	Plausibility of change
Selection	Central	Assumed to optimize	Ignored
Defection	Central	Ignored	Ignored
Phenotypic detail	None	To assess fitness	To understand development

split between germ and soma was present. A stable equilibrium is a population-level property, and the difference-makers identified by Michod change whether or not this equilibrium occurs, and what kind of equilibrium it is.

Explanations 2 and 3 for *V. carteri* refer to properties of individuals. The first shows the differences required to enable a small mass of cells to become a large mass of cells. If we assume that fitness is increased by being bigger, it tells us why such a larger mass of cells might have evolved, and what enabled them to do so. Similarly, Kirk's explanation shows the differences required to add new functionality to some putative ancestor of *V. carteri*. It provides a plausible explanation for the origin of certain variations, giving us a picture of how a small change, or the co-option of a prior mechanism, led to some new, important functionality necessary for the evolution of multicellularity.

The contrast between population level and individual level properties brings with it some other noteworthy differences (summarized in table 2.2). To begin with, selection plays a very different role in each of these explanations. For the first explanation, selection is specifically modeled; it plays a central and focal role. The fitness of each type, and the frequency-dependent interactions between them, determine the equilibria, which are the target of the explanation. In the second explanation, selection is not explicitly modeled at all. Instead, the focus is on understanding the mechanisms that enable individuals to grow in size. Selection is off-stage, assumed to act on size, bringing about the large, more organized, multicellular types. In the last case, fitness is ignored. No doubt, Kirk thinks that natural selection played an important part in the evolution of multicellularity in *V. carteri*. But his explanations concern the developmental and physiological differences between an ancestral single-celled organism, and the relatively complex *V. carteri*. He shows these differences could have occurred through a series of small changes, but does not consider their selective advantage. His goal is, instead, to document the origins of these variations—elucidating where they came from, rather than why they stuck around.[8]

The physical detail in these explanations also differs. The first explanation ignores much about how *V. carteri* develops and functions, whereas the second two explanations rely on quite explicit physiological and developmental details. In the first case, it is the flagellation constraint that forces the introduction of the division of labor, and in the second case,

specific developmental details are required to understand the origins of each of Kirk's steps.

Despite these differences, each of these explanations identifies specific difference-makers for the evolution of the transition from single cell to complex multicellularity in *V. carteri*. Michod explains what, in part, enabled the stability of cooperation and how it required a split between germ and somatic lines, Solari et al. explains why getting larger was better and how a division of labor made it possible, and Kirk explains the probable origins of a series of developmental mechanisms essential to the final complex multicellular organism.

Moreover, these explanatory ideas are not unique to *V. carteri*: All of these authors are interested in these examples as a model system for understanding the evolution of multicellularity in general. In turn, multicellularity exemplifies a more general phenomenon: the formation of related collectives that can lead to new levels of individuality.

Alternative Patterns of Explanation for Major Transitions

As I have said, most work on major transitions has sought to explain the stability of cooperation, and the resulting generalizations have dealt with population-level properties. In contrast, when we look at one well-studied transition (the evolution of complex multicellularity in *V. carteri*), we see very different kinds of explanations, whose targets are individual-level properties. For now, I've only shown that such explanations can be given for a particular transition. But can such kinds of explanations be made general enough to identify important patterns in major transitions?

As I have noted, the particular explanations that were invoked for *V. carteri* won't work—not all major transitions concern flagellated pond inhabitants. The explanations need to be specified far more abstractly to be applicable across a greater domain of major transitions. Models of cooperation underwent exactly this kind of abstraction, so they could be made general enough to apply across the biological hierarchy:

> What began as the study of animal social behavior some forty years ago has now embraced the study of social interactions at all levels in the hierarchy of life. Instead of being seen as a special characteristic clustered in certain lineages of social animals, cooperation is now seen as the primary creative force behind ever greater levels of complexity through the creation of new kinds of individuals (Michod and Herron, 2006).

Perhaps the same kind of move is possible with these kinds of individual-level explanations. To show this, we need to return to this issue of why explanations in biology can apply across a broad domain.

Generalizations in Biology

Similarity due to biological relatedness is one obvious reason we can apply explanations across some domain. Our explanation for jumping generalizes to all kangaroos because all

kangaroos are pretty much alike. And kangaroos are all pretty much alike because they are related by common descent. Biological properties that are conserved through heredity provide similarities that allow us generalize across broad domains. For example, the near ubiquity of the genetic code enables us to make some *very* broad biological generalizations.

This kind of similarity is not the reason behind the most common generalizations about major transitions. The kinds of properties mentioned are not simply those that are shared due to relatedness—the evolution of eusociality and the evolution of multicellularity are not made possible because of some trait that is shared by common descent. Relatedness, for example, is important in many fraternal transitions. But the relatedness of Metazoan cells is not homologous to that of bees in a hive.

The generality of claims about major transitions comes about for a different reason, and it is the same reason that many models in population biology can be applied to populations of vastly different organisms. These models are very general because they make claims based on assumptions about fitness, yet how these fitnesses are actually produced does not matter. For example, a model of sexual selection will rely on various parameters, including mating probabilities and the fitness of some trait. But, depending on the species being studied, the actual trait may be as different as tail length, fur pattern, or the model of car owned. What matters for generality is that the population structure is the same, even though the individuals that compose the population, and the ways they behave, may be very different. As Sober says, "Even a race of robots or organisms from another planet whose mechanism of heredity is based on a structure other than DNA would potentially fall within the scope of these theories" (Sober 1984).

These generalities are about the properties of populations. Given certain assumptions about population size, degrees of heredity, mating preferences, and so forth, we can explain (and sometimes predict) the trajectories, or probable trajectories, of evolutionary processes. We can make these generalizations in part *because* we can ignore the physical facts of how fitness is realized.

Thus, the first explanation for the evolution of complex multicellularity in *V. carteri* abstracts away from physical details, and simply assumes that certain traits are heritable, and that these traits are associated with a particular fitness structure. By ignoring detailed physical facts, the resulting explanation is one that could easily be generalized to other kinds of organisms, and at other levels of organization.

I've now suggested two reasons that generalizations in biology can be made. The first is simply that the domain includes aspects of organisms that are alike *because* they are related. The second reason is that many generalizations about population processes rely on particular kind of fitness relations, even if the particular traits that produce these fitnesses are very different. Neither of these ways to make generalizations fits the bill if we want to generalize individual-level explanations to major transitions. We cannot rely on similarity due to common descent (as we could with kangaroos), for the individual properties of interest do not have a common origin. And we cannot abstract away from biological details

by using fitness, for the kinds of similarities we derive by doing this are about population-level processes. What we need is a third way to make generalizations, one that identifies similarities across individual-level properties that are not reliant on common descent.

Individual organisms may possess similar features for reasons other than relatedness—these similarities are analogies, rather than homologies. Dennett helpfully labeled such similarities as "forced moves" and "good tricks" (Dennett 1995). One source of forced moves is the universality of physical constraints—"the ways in which the world of organisms bumps against a non-biological reality" (Vogel 1988). For example, "animals have repeatedly evolved a complex branching hierarchy of vessels approximating a globally optimal system that minimizes the costs of the construction and maintenance of the fluid transport system" (LaBarbera 1990).

The actual fluid being transported may be as different as blood or sap, and the building material may also differ. But when stated in a very general fashion we have an explanation that holds across a wide variety of organisms, and does so because of physical constraints on how to best construct a particular kind of biological mechanism.

Physical constraints don't always produce a single optimal phenotype. Phenotypes are often compromises, making trade-offs between a number of physical constraints, and there may be several trade-offs that can be made. Karl Niklas has produced a series of models identifying several optimal plant shapes. These different optima exist because the plant shape must simultaneously perform several tasks, such as intercepting light, maintaining mechanical stability, dispersing spores, and conserving water (Niklas 1994, 2004). The different optima are produced because there are different ways of trading off each of these mutually constraining demands. The plants produced at each optimum also bore a strong resemblance to extant plants. Models such as these can generalize about similarity (where unrelated plants take on similar shapes) and diversity (why different plant shapes exist).

Good tricks can also result in striking similarities across diverse organisms. For example, finding your way home quickly and accurately without retracing your steps is an important life skill for many organisms. *Path integration* is a very simple, and very widely used, navigational strategy that has been discovered multiple times, in organisms as different as ants, rats, hamsters, crabs, bees, and spiders (Vickerstaff and Di Paolo 2005). Animals using path integration maintain a home vector that encodes the distance and direction to the nest or burrow. This vector is updated as the animal moves away from its home, by incorporating the distance moved and any change in direction—the equivalent of a technique in human navigation known as dead reckoning (Andel and Wehner 2004). Even after a circuitous outgoing search for food, organisms using this strategy can head directly home along the shortest route. Path integration itself appears to be a very general phenomenon, though its specific implementation may vary: In ants, updating the distance is done by counting steps; in bees, it is done by measuring optic flow (Wittlinger, Wehner, and Wolf 2006).

These examples show that broad generalizations about individual-level properties across nonrelated organisms are possible.[9] But it is important to note that these kinds of generalizations rely on a much more heterogeneous set of facts than the other kinds of generalizations. The similarities in the domain of explanation are not straightforward as they are in the case of relatedness in population level generalizations. Nor is this abstraction dependent on a single general factor (as is the case when the generalizations rely on knowing fitness, but ignoring how fitness might be generated). Rather, these cases require discovering the particular physical or organizational details that force particular solutions, or by recognizing what simple, effective solutions are available to problems that repeatedly occur across many organisms.

A Richer Understanding of Major Transitions

Here is what I have argued for thus far. First, claims about structural similarities across major transitions can be captured as broad, but otherwise ordinary, explanatory generalizations. Second, the kinds of generalizations central to most discussions of major transitions have been about population-level properties; notably, the identification of those factors that enable cooperation to be stabilized. Third, by examining one particular transition (in *V. carteri*), we see that other patterns of explanations—about individual-level properties—can be deployed to explain a transition. Finally, in the last section, I suggested that it is possible for these kinds of explanations to made general as well. This suggests that different patterns of explanation might yield other similarities across major transitions.

What would these explanations look like? The previous discussion of *V. carteri* suggests two places we might find such generalizations. First, consider Solari et al.'s explanation. Their model described how a division of labor was necessary to enable the evolution of a simple, but advantageous, increase in size. John Tyler-Bonner has argued for many years about the importance of getting bigger in general, so this is not an isolated incident (Bonner 2006). But as Solari et al. demonstrate, *simply getting bigger* is often not an option. Getting bigger requires reorganizing how the organism operates, often to something requiring more complexity (see Knoll and Hewitt, this volume, for more on what an increase in size might require). Just how this reorganization takes place may require new abilities—in this case, a division of labor was necessary.

Division of labor was a property that Maynard-Smith and Száthmary noted in many of the major transitions (Maynard Smith and Szathmáry 1995). It is also a property that Haim Ofek suggests as one example of a convergent social structure—a "good trick" from the social world. He urges us to look for further social adaptations that "repeat themselves in widely separate species and, for that reason, can be clearly ascribed to analogy (as distinct from homology)" (Ofek 2001, p96). One possible area for the further discovery of such convergent social structures may be group decision-making procedures. As groups become larger, the problems of aggregating information and making fast, informed decisions becomes more difficult. This kind of problem, like the division of labor, is expressible in abstract formalisms that may be applied across many different organisms, much like the work

on cooperation (see List 2004 for a simple example of applying formal work to animal groups).

Kirk's explanation provides another source for seeking very general explanations. Kirk's general approach is to understand what prior mechanisms needed to be in place to enable a transition. One possibility for a very general property that is important for transitions in levels of organization is plasticity. Carl Schlichting has argued that plasticity may play an important part in the origins of multicellularity, as differential expression in the variable microenvironments created by cell aggregation could explain the origin of cellular differentiation (Schlichting 2003). More recently, Michod has pointed out that the differential expression of a trait over an individual's life history might be coopted to be simultaneously expressed as a division of labor in a group (see Michod, this volume). These explanations resemble Kirk's—they seek to understand the origin of important properties that were essential to the operation of organized collectives. But the general implication is that plasticity is important to other transitions, such as the advent of eusociality. These ideas are not about populations. Rather, they explain the origin of particular properties of the higher-level individuals in terms of the prior abilities of lower-level individuals.

These suggestions identify very general principles that may provide alternative ways of unifying the mechanisms underlying the major transitions. If such analogies identify difference-makers that underwrite the stability or complexity of social structures *in general*, then these properties—like those identified by investigations into the stability of cooperation—can help us understand how new levels of organization evolve.

Last, these alternative explanatory patterns can provide another advantage, when used in conjunction with one another. As Tinbergen noted, different kinds of explanations can constrain and inform each other (Tinbergen 1963). Some of these interactions are evident in table 2.2. In the conflict mediation explanation, defection is a key component, as this must be overcome for cooperation to be stable and for groups to evolve. Just why being in a group is advantageous in the first place is not explained, for no phenotypic detail is provided. Contrast this with the generating benefit explanation. Here, explicit phenotypic details are provided, telling us that size is important, and explaining how this size is achieved. But in this case, the problem of defection is ignored—the individuals making up the collective are simply assumed to cooperate perfectly. What one explanation assumes, the other explains. Understanding the interactions between the various assumptions can provide a richer understanding of major transitions, by building a more complete and connected picture of the various factors that enable new levels of organization to arise.

Acknowledgments

Thanks to Kim Sterelny for many helpful suggestions. This work was funded by a Cambridge-Templeton Consortium grant (General Mechanisms Underlying Transitions in Complexity: An interdisciplinary approach), which I gratefully acknowledge.

Notes

1. Mayr's original distinction was between proximate and ultimate *causation*. This contrasts with Tinbergen, who posed his distinctions as alternative questions one could ask. The difference between an account of causation and an account of causal *explanation* is something I won't address here. Instead, I shall interpret Mayr's claims as offering different kinds of explanations (as others, such as Queller 2006, have done).

2. Here are some reasons for its suitability: (a) explanations can be given at any level of organization—they need not appeal to laws, or be reduced to more basic physical/chemical processes; (b) it can be used to explain classes of phenomena (generalizations) as well as single cases; (c) much of the technical apparatus associated with the account (such as in Pearl 2000) is designed to extract causal information from correlative information in situations where there are complex relationships between processes, as is often the case in biology. This account has also formed the basis for other more particular theories of explanation in biology (Calcott 2009; Craver 2007; Glennan 2005).

3. Like here: <http://animals.howstuffworks.com/mammals/kangaroo-hopping.htm>.

4. Philosophers refer to the target of explanation as the *explanandum* and difference-makers as the *explanans*. I always forget which way round they go.

5. I'm taking "kangaroo" to refer just to the genus *Macropus*, which excludes tree kangaroos. They can jump, too, but not in the same way as the iconic kangaroos.

6. Michod's later work (summarized in this volume) includes a much broader approach, incorporating aspects of all three types of explanation I outline here. The division of labor model is a more general version of Explanation 2, and Michod's discussion of the *regA* gene is a detailed example of what Kirk attempts in Explanation 3.

7. The contrast between explanations that focus on individual-level properties and those that focus on population-level properties is one way to view the difference between Mayr's proximate and ultimate explanations (Ariew 2003) suggests something like this.

8. Often summarized as "the arrival of the fittest, rather than the survival of the fittest" (DeVries 1904).

9. For a large array of extraordinary convergences at all levels of organization, see Conway Morris's book "Life's Solution" (Morris 2004). One can appreciate the ubiquity of convergent phenomenon without accepting his conclusions.

References

Afrane, Y. A., E. Klinkenberg, P. Drechsel, K. Owusu-Daaku, R. Garms, and T. Kruppa. (2004). Does irrigated urban agriculture influence the transmission of malaria in the city of Kumasi, Ghana? *Acta Trop 89*(2):125–134.

Andel, D., and R. Wehner. (2004). Path integration in desert ants, Cataglyphis: how to make a homing ant run away from home. *Proceedings of the Royal Society of London Series B, Biological Sciences 271*(1547):1485–1489.

Ariew, A. (2003). Ernst Mayr's "ultimate/proximate" distinction reconsidered and reconstructed. *Biology and Philosophy 18*:553–565.

Bonner, J. T. (2006). *Why Size Matters: From Bacteria to Blue Whales.* Princeton, NJ: Princeton University Press.

Calcott, B. (2008). The other cooperation problem: Generating benefit. *Biology and Philosophy* 23:179–203.

Calcott, B. (2009). Lineage explanations: Explaining how biological mechanisms change. *British Journal for the Philosophy of Science* 60(1):51–78.

Craver, C. F. (2007). *Explaining the Brain.* Oxford: Clarendon Press.

Dennett, D. C. (1995). *Darwin's Dangerous Idea: Evolution and the Meanings of Life.* New York: Simon & Schuster.

DeVries, H. (1904). *Species and Varieties: Their Origin by Mutation.* Chicago: Open Court.

Gavrilets, S. (2004). *Fitness Landscapes and the Origin of Species.* Princeton, NJ: Princeton University Press.

Glennan, S. S. (2005). Modeling mechanisms. *Studies in History and Philosophy of Science Part C: Studies in History and Philosophy of Biological and Biomedical Sciences* 36(2):443–464.

Hitchcock, C., and J. Woodward. (2003). Explanatory generalizations, part 2, Plumbing explanatory depth. *Nous* 37(2):181–199.

Hughes, W. O., B. P. Oldroyd, M. Beekman, and F. L. Ratnieks. (2008). Ancestral monogamy shows kin selection is key to the evolution of eusociality. *Science* 320(5880):1213–1216.

Kirk, D. L. (2005). A twelve-step program for evolving multicellularity and a division of labor. *Bioessays* 27(3):299–310.

LaBarbera, M. (1990). Principles of design of fluid transport systems in zoology. *Science* 249(4972):992.

List, C. (2004). Democracy in animal groups: A political science perspective. *Trends in Ecology and Evolution* 19(4):168–169.

Marshall, C. R. (2006). Explaining the Cambrian "explosion" of animals. *Annual Review of Earth and Planetary Sciences* 34:355–384.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution.* Oxford; New York: W.H. Freeman/Spektrum.

Mayr, E. (1961). Cause and effect in biology. *Science* 134: 1501–1506.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality.* Princeton, NJ: Princeton University Press.

Michod, R. E., and M. D. Herron. (2006). Cooperation and conflict during evolutionary transitions in individuality. *Journal of Evolutionary Biology* 19(5):1406–1409.

Morris, S. C. (2004). *Life's Solution: Inevitable Humans in a Lonely Universe.* New York: Cambridge University Press.

Niklas, K. (1994). Morphological evolution through complex domains of fitness. *Proceedings of the National Academy of Sciences* 91(July):6772–6779.

Niklas, K. (2004). Computer models of early land plant evolution. *Annual Review of Earth and Planetary Sciences* 32:47–66.

Ofek, H. (2001). *Second Nature: Economic Origins of Human Evolution.* Cambridge; New York: Cambridge University Press.

Pearl, J. (2000). *Causality.* Cambridge and New York: Cambridge University Press.

Queller, D. C. (2006). Sex ratios and social evolution. *Current Biology* 16(17):R664–668.

Schlichting, C. D. (2003). Origins of differentiation via phenotypic plasticity. *Evolution & Development* 5(1):98–105.

Sober, E. (1984). *The Nature of Selection: Evolutionary Theory in Philosophical Focus.* Cambridge: MIT Press.

Solari, C. A., A. M. Nedelcu, and R. E. Michod. (2003). Fitness and complexity in volvocalean green algae, in H. Lipson, E. K. Antonsson, and J. R. Koza (eds.), *Computational Synthesis: From Basic Building Blocks to High-Level Functionality* (pp. 218–225). Menlo Park, CA: AAAI Press.

Tinbergen, N. (1963). On aims and methods of ethology. *Zeitschrift für Tierpsychologie* 20:410–433.

Vickerstaff, R. J., and E. A. Di Paolo. (2005). Evolving neural models of path integration. *Journal of Evolutionary Biology* 208(17):3349–3366.

Vogel, S. (1988). *Life's Devices: The Physical World of Animals and Plants.* Princeton, NJ: Princeton University Press.

Wiesenfeld, S. L. (1967). Sickle-cell trait in human biological and cultural evolution. Development of agriculture causing increased malaria is bound to gene-pool changes causing malaria reduction. *Science* 157(793):1134–1140.

Wittlinger, M., R. Wehner, and H. Wolf. (2006). The ant odometer: Stepping on stilts and stumps. *Science* 312(5782):1965.

Woodward, J. (2003). *Making Things Happen: A Theory of Causal Explanation.* New York: Oxford University Press.

Woodward, J., and C. Hitchcock. (2003). Explanatory generalization, part 1, A counterfactual account. *Nous* 37(1):1–24.

3 Biological Ontology and Hierarchical Organization: A Defense of Rank Freedom

Samir Okasha

This chapter deals with the ontology of biology systems, with particular reference to hierarchical organization. That biological systems exhibit hierarchical structure is a commonplace: larger biological units, such as multicelled organisms, are composed of smaller biological units (e.g., cells), which themselves contain still smaller units (e.g., chromosomes).

Commonplace though this observation is, it is unclear exactly how the biological hierarchy should be conceptualized. Is the biological hierarchy strictly nested, or does it permit overlapping? What determines the hierarchical level that a given biological unit occupies? What biological relation(s) bind(s) the smaller biological units into larger units? Moreover, it is unclear whether there is a *single* hierarchy, subsuming all biological units. Eldredge (1985, 2003) argues that there are actually two hierarchies, genealogical and ecological; Sarkar (1998) contrasts the "abstract" genetic hierarchy with the "spatial" hierarchy; while Brandon (1988) discerns a dual hierarchy of replicators and interactors. So there is clearly plenty to be said about the nature of hierarchical organization in biology.

Building on the ideas of Eldredge, I examine the notion of hierarchy as it is featured in two quite different areas of modern biology. The first is the study of the "major evolutionary transitions," *sensu* Maynard Smith and Szathmáry (1995), and the related discussion of multilevel selection. The second is phylogenetic systematics. Hierarchical organization is central to both of these areas of biology, though quite different types of entities are involved. In the major transitions case, new hierarchical levels are created when free-living biological units, capable of surviving alone, become integrated into a larger collective. In the phylogenetics case, new hierarchical levels are created through repeated cladogenesis, or lineage splitting, which leads to a nested hierarchy of monophyletic groups.

A central idea in modern phylogenetic systematics is that of a *rank-free* hierarchy, that is, a hierarchy in which the various levels have no absolute meaning, unlike the traditional Linnaean hierarchy. I argue that the idea of rank freedom, which has proved so fruitful in phylogenetic systematics, can be extended to the quite different biological hierarchy that arises from evolutionary transitions. Applied to the latter hierarchy, rank freedom involves rejecting the idea that some biological units are organisms while others are suborganismic

or superorganismic; rather, all entities in the hierarchy are on a par, for there are no ranks. This fits well with the way that the concepts of "individual" and "group" are understood by multilevel selection theorists, and permits a useful new perspective on the old question, "What is an organism?"

Hierarchy in Two Areas of Biology

Beginning with Buss (1987), the literature on major transitions in evolution, also known as evolutionary transitions in individuality, has burgeoned (cf. Frank 1995; Maynard Smith and Szathmáry 1995; Michod 1999, 2005; Michod and Nedelcu 2003; Queller 2000; Reeve and Keller 1999; Sober and Wilson 1998). This is because biologists have come to realize that the coalescing of smaller biological units into larger ones is something that has occurred repeatedly in the history of life, generating the hierarchical complexity that we see in modern biological systems. A partial list of such transitions includes: single RNA replicators → networks of replicators, individual genes → chromosomes, prokaryotic cells → eukaryotic cells, single-celled organisms → multicelled organisms, solitary animals → integrated colonies. The challenge is to understand such transitions in Darwinian terms (as well as to piece together the actual sequence of stages in each transition). Why was it advantageous for the smaller biological units to sacrifice their individuality and form themselves into a corporate body? And how could such an arrangement, once evolved, be stable against invasion by cheats? These are the questions that a theory of evolutionary transitions must answer.

The study of evolutionary transitions has led to a reassessment of the traditional levels of selection question, familiar from the sociobiology debates of the 1960s and 1970s (Okasha 2005, 2006). Clearly, in any evolutionary transition, the potential exists for selection to act at more than one hierarchical level. For example, in the transition to multicellularity, selection could act on variant cell types within the emerging multicellular aggregate, and also on the aggregates themselves. Though the traditional levels of selection debate did not explicitly deal with evolutionary transitions, many themes and lessons from the former have proved useful for understanding the latter (Michod 1999; Queller 2000)—for example, that individual and group interests can pull in opposite directions, that high relatedness favors the evolution of cooperation, and that "policing" can promote group cohesion are all sociobiological themes that have reappeared in the recent literature on evolutionary transitions.

Clearly, hierarchy is central to the evolutionary transitions—for they lead smaller biological units to become nested within larger ones, giving rise to new "evolutionary individuals," in the terminology of Michod (1999). The resulting hierarchy corresponds to what Eldredge (1985) called the "ecological hierarchy." In this hierarchy, interaction among the smaller units is the source of the part-whole structure, that is, ecological interaction is the biological relation that binds the smaller units into a larger one. By contrast, in

Eldredge's genealogical hierarchy, genealogical relatedness, not ecological interaction, is what binds the smaller units into a larger unit. Admittedly, in certain of the evolutionary transitions—which Queller (2000) calls fraternal transitions—the smaller units that coalesce into the larger unit are closely related, or even clones (e.g., the transition to multicellularity").[1] However, in other transitions the coalescing units are unrelated (e.g., the formation of eukaryotic cells by the union of unrelated prokaryotes). And even in the fraternal transitions, it is not *by virtue* of being genealogically related that the smaller units constitute part of a larger unit; rather, it is by virtue of the fitness-affecting interactions that they engage in, for example, cooperation, division of labor, mutual policing, and so on. So the relevant hierarchy is ecological, not genealogical.

In phylogenetic systematics, a quite different area of biology, the notion of hierarchy is also central; but here the relevant hierarchy is genealogical. The main tasks of systematics, as usually conceived, are two: first, to reconstruct the branching tree of life, and second, to devise a coherent way of classifying extant species into higher taxa. The question of what the correct methodology for biological classification should be was extensively debated in the 1960s and 1970s (Hull 1988). Though the debate goes on, more and more biologists have now come around to the phylogenetic, or cladistic, point of view. The key principle of phylogenetic systematics is that all taxa should be monophyletic, that is, they should consist of an ancestral species and all and only its descendent species. Monophyletic taxa are real entities that exist independently of our classification scheme, cladists argue, whereas para- and polyphyletic taxa are not.

The concept of monophyly is best illustrated graphically. In the phylogenetic tree depicted in figure 3.1, which depicts the phylogenetic relations among seven extant species, A to E, all the monophyletic groups have been ringed. Notice that the monophyletic groups are nested inside each other, giving rise to a hierarchical pattern. This is not an accident: If the underlying phylogeny is branching rather than reticulate (i.e., if the branches never join up), then as a matter of logic, all monophyletic groups will be strictly nested. Cladistic principles thus give a clear justification for why biological classification should be hierarchical.

It is worth emphasizing that this phylogenetic hierarchy, which consists of monophyletic groups of species nested inside each other, is totally different from the hierarchy generated by the major transitions, which consists of evolutionary individuals nested inside each other. Of course, there is a tradition of arguing that species, and monophyletic taxa more generally, are *themselves* individuals (Ghiselin 1974; Hull 1978; Mishler and Brandon 1987); but this involves a quite different notion of individual from that at work in the evolutionary transitions literature. In the latter, the point of calling something an evolutionary individual (or organism) is to stress that it is functionally integrated, that is, its constituent parts work for the good of the whole. (It is in this sense that eusocial insect colonies, for example, are sometimes said to be evolutionary individuals.) No such implication is contained in the idea that species and clades are individuals; they are obviously not

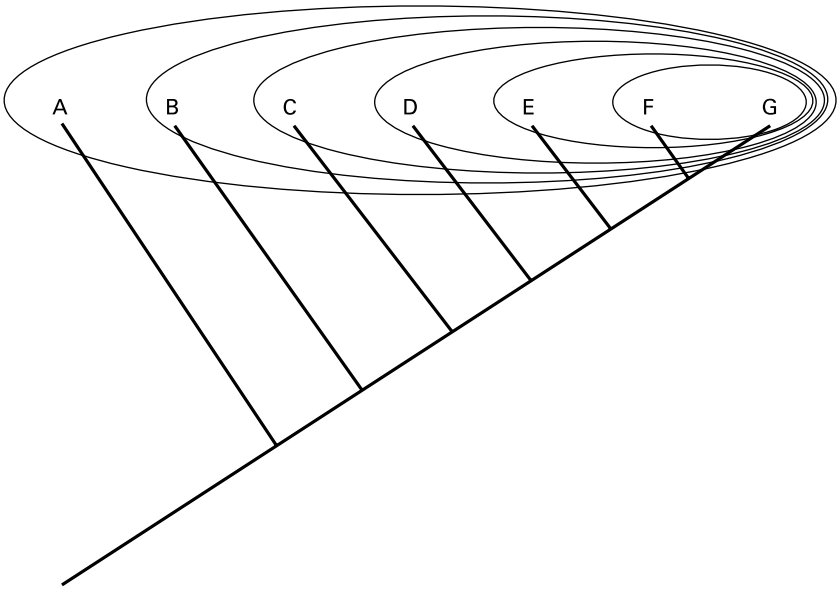


Figure 3.1
Phylogenetic tree with monophyletic groups ringed.

functionally integrated units. This highlights the fact that the ecological and genealogical hierarchies are quite different in kind.

Despite this difference, I believe that an interesting parallel can be drawn between the two hierarchies. Specifically, I argue that the concept of a rank-free hierarchy, which plays a key role in modern phylogenetic systematics, can be transposed to the major transitions hierarchy, with interesting consequences.

Rank Freedom in Phylogenetic Systematics

The concept of rank freedom arose out of attempts to reconcile phylogenetic-based classification with the traditional Linnaean classification system. As we have seen, the requirement that all taxa be monophyletic, the key principle of phylogenetic systematics, leads automatically to a nested hierarchy of units. The Linnaean system is also hierarchical: a number of species belong to a single genus, a number of genuses to a single family, a number of families to a single order, and so on. So it seems as if a rapprochement between the two classification systems should be possible. This was the view of many early phylogenetic systematists, who believed that the Linnaean system could basically be retained, subject to the requirement of universal monophyly. If the Linnaean system recognized a taxon,

be it a genus, family, class, or whatever, that turned out not to be monophyletic, the taxon must be rejected; but where monophyly was satisfied, the Linnaean classification system could peacefully coexist with phylogenetic-based classification. This peaceful coexistence was welcome because it was relatively nonrevisionist—cladists did not need to begin the project of biological classification anew, nor to rename existing taxa.

However, the ideal of peaceful coexistence has increasingly fallen on hard times, because it faces a crucial problem. How is one supposed to determine which Linnaean rank a given monophyletic group of species occupies (if any)? For example, in figure 3.1, what determines whether the group {D, E, F, G}, for example, constitutes a genus, a family, or an order? This question arises because the Linnaean hierarchy is a *ranked* hierarchy, whereas the phylogenetic-based hierarchy, constructed according to the principle of strict monophyly, is not. So if a rapprochement between the two hierarchies is to be achieved, there must be a way of assigning monophyletic groups to Linnaean ranks. Many modern cladists believe that there is no principled way of doing this and thus reject the idea of rapprochement, advocating instead that the Linnaean system be abandoned (Ereshefsky 2001). The only theoretically defensible classification system, they argue, must be both phylogenetically based and *rank free*. In a rank-free system, just two types of units are recognized: basal taxa (i.e., species) and monophyletic groups of species, of various degrees of inclusiveness. These monophyletic groups form a nested hierarchy, but no ranks are recognized. The question of whether a given taxon is a genus or a family, for example, is rejected as meaningless. The well-known *Phylocode* project, which codifies the principles of phylogenetic systematics, explicitly incorporates rank freedom.[2]

The advantages of a rank-free classification system are numerous. First, it is ontologically simpler, positing fewer types of entity. Second, it avoids the need for ad hoc modifications to the Linnaean system, such as adding ranks like superfamily and subphylum, which theorists had been driven to in their attempt to make the system work. Third, it faces up to the fact, long an embarrassment to taxonomists, that the Linnaean ranks had always seemed somewhat arbitrary, in that all the taxa designated as orders, for example, did not appear to have anything deep in common. Finally, rank freedom steers an interesting middle course between realism and conventionalism about higher taxa. Consider a traditional Linnaean taxon, for example, the primate order. Is this a real or a conventional classification? According to rank-free phylogenetic systematics, the answer depends on whether we are talking about grouping or ranking (cf. Mishler and Brandon 1987). There is an objective, mind-independent fact about whether the species we count as primates are monophyletic; if so, "primate" designates a real taxon, but if not, it does not. But there is no objective fact about whether primates are an *order*, rather than a class or a phylum, for ranks are mere conventions. So proponents of rank freedom argue that we should be realists about grouping, but conventionalists about ranking. This helps clarify the kernel of truth contained in the old taxonomist's adage "species are real units, higher taxa are arbitrary classes."

Rank Freedom and Evolutionary Transitions

Can the idea of rank freedom be extended from systematics to the quite different biological hierarchy that results from evolutionary transitions? I believe that it can and should, for it sheds light on a number of aspects of the discussion of major transitions and multilevel selection.

To see how the idea of rank freedom can apply to evolutionary transitions, recall the essential features of the transitions. A number of smaller biological entities, originally capable of surviving and reproducing alone, form themselves into a corporate body and sacrifice their individuality, giving rise to a new entity (or evolutionary individual) and thus an increase in hierarchical complexity. The hierarchy that results from this transitional process is often conceptualized in a ranked way. One manifestation of this is the idea that "organism" denotes an absolute level in this hierarchy; we refer to entities such as cells and organelles as "suborganismic," entities such as groups and colonies as "superorganismic." Thus, we talk as if there is an objective fact not just about whether any given biological unit is a genuine evolutionary individual, but also about what rank in the hierarchy the unit occupies.

However, it is difficult to see the rationale for designating some entities as organisms but not others; just as it is difficult to see the rationale for designating some taxa as orders, others as phyla. I suggest that a rank-free approach makes more sense. In a rank-free approach, no absolute meaning attaches to the various levels in the ecological hierarchy. Any entity that exhibits sufficient functional integration, and whose parts work (mostly) for the good of the whole, constitutes a genuine evolutionary individual, or organism, and thus belongs in the hierarchy. But there is no further question about what rank the entity occupies, nor about whether the entity is "really" an organism.

In a discussion of social insect evolution, David Queller remarks: "if it seems to be time for a revival of the view that social insect colonies can be superorganisms, I would suggest that it is more consistent to simply view them as organisms" (2000, 1653). Queller's suggestion may seem purely semantic, but in fact it touches on a deep philosophical issue, that he explores at length in his earlier review of Maynard Smith and Szathmáry's *Major Transitions in Evolution*. In that review, Queller argues that the modern understanding of major transitions requires us to rethink the concept of an organism. He writes:

> Our tendency to think of the organism as one of the levels in the hierarchy of life does not stand up to scrutiny. We find prokaryotic organisms, eukaryotic assemblages, multicellular eukaryotes, and organismal colonies. We designate something an organism, not because it is *n* steps up on the ladder of life, but because it is a consolidated unit of design. . . . You cannot pick a level above the organism and expect to see all cooperation and no conflict. If that is what you see, as arguably you do in some social insect colonies, then to maintain consistency I would suggest that you are looking at an organism. (Queller 1997, 18)

The idea that Queller expresses here, and the semantic recommendation about the use of the term "organism" to which it gives rise, makes perfect sense from a rank-free perspective. In effect, Queller is arguing that our tendency to read absolute meaning into the different levels in the ecological hierarchy is a mistake, in just the way that phylogenetic systematists regard it as mistaken to read absolute meaning into the various levels in the genealogical hierarchy. In both cases, we find a nested hierarchy of units, but in neither case does the hierarchy have ranks. It makes no more sense to ask whether a particular biological entity (e.g., an ant colony), occupies the rank of "organism" than it does to ask whether a particular monophyletic taxon occupies the rank of "family." Both questions must be rejected as resting on a false presupposition.

This is not to say that the concept of an organism should be abandoned, or rejected as meaningless. On the contrary, I take the concept to be fairly clear; it applies to any biological unit with a high degree of functional integration that is capable of reproduction, so has a life cycle, and whose parts work (mainly) for the good of the whole. The point is just that organism doesn't denote a rank in the ecological hierarchy; rather, *all* entities in that hierarchy, at all levels of inclusiveness, are organisms, or at least approximate that status. In the rank-free approach that I am advocating, therefore, we still get an objective answer to the question of whether or not a given entity is an organism (though, of course, some cases are borderline). But it offers no objective answer about whether a given entity occupies the *rank* of organism in the ecological hierarchy, as opposed, for example, to the rank of superorganism, for the hierarchy has no ranks.

Adopting a rank-free approach to either hierarchy involves a kind of egalitarianism: Entities at all hierarchical levels are treated alike, rather than assigned to determinate ranks. Of course, this does not imply that it is impossible to sort the entities into scientifically interesting categories, or kinds, for most such kinds are not ranks. A rank is a special *sort* of kind: Each entity in the hierarchy is meant to occupy exactly one rank, and if two entities belong to the same rank, they cannot bear the part-whole relation to one another (for then the part would have to be ranked below the whole). In arguing that the ecological hierarchy is rank free, I mean that the entities in the hierarchy cannot be assigned to ranks meeting these criteria; this is not to say that they cannot be divided into *any* kinds that are natural and/or scientifically interesting.

It is striking that one standard way of describing the ecological hierarchy, in discussions of evolutionary transitions, involves the use of *genealogical* descriptors to distinguish the levels from one another. For example, in the above quotation, Queller refers to "prokaryotic organisms, eukaryotic assemblages, multicellular eukaryotes" as three consecutive levels in the hierarchy. But the terms "prokaryote" and "eukaryote" are usually treated as phylogenetically defined. At least in the first instance, an entity counts as a eukaryote not by virtue of its particular internal properties, but by virtue of its ancestry, that is, its position in the tree of life.[3] This practice of using phylogenetic nomenclature to refer to the levels in the ecological hierarchy is widespread. I suggest that it provides further evidence for the

fact that the ecological hierarchy is actually rank free. If each entity in that hierarchy really belonged to a definite rank, there would be little point in using a nomenclature based on the entities' positions in a quite different hierarchy to identify the ranks.

Another argument for rank freedom stems from considerations relating to multilevel selection theory. As noted in the Hierarchy in Two Areas of Biology section, multilevel selection is intimately bound up with evolutionary transitions; for in a transition, natural selection can potentially act at (at least) two hierarchical levels. Multilevel selection theorists generally characterize the two levels as the group and the individual; transitions therefore require that group selection be strong enough to trump individual selection. (This terminology partly reflects the fact that multilevel selection theory grew out of the group selection debates of the 1960s.) However, most multilevel selection theorists are explicit that group and individual do not denote absolute levels in the biological hierarchy, but are purely relative designations (cf. Okasha 2006, 40–6). An entity that counts as a group in one context may count as an individual in another, and vice versa. Hamilton (1975) made this point clearly, in his famous demonstration of how Price's equation, suitably expanded, can describe selection at indefinitely many hierarchical levels. More recently, Michod (1999) expressed awareness of the same point, in emphasizing that the very existence of multicelled organisms, which are groups of cooperating cells, refutes the argument that group selection is invariably a weaker force than individual selection.

The fact that "group" and "individual" are used in this purely relative way tallies well with the idea of a rank-free hierarchy. For if the terms had absolute meanings, this would imply the existence of ranks in the hierarchy: There would be an objective fact about whether a given entity, such as a cellular slime mold, occupies the rank of group, individual, or neither. I suspect that most theorists of evolutionary transitions would deny there is such a fact; rather, all entities in the hierarchy, apart from those at the very bottom, are simultaneously groups and individuals, depending on our choice of focal level. This further supports the idea that the hierarchy generated via the coalescence of smaller reproducing units into larger ones is rank free.

To summarize, the idea of rank freedom, as applied to the ecological hierarchy, makes good sense of a number of facets of the recent discussion of evolutionary transitions and multilevel selection.

How Deep Does the Analogy Run?

Like all analogies, the preceding analogy between rank freedom in phylogenetic systematics and in evolutionary transitions does not hold in every respect. It is therefore important to ask how deep the analogy runs, and whether it is illuminating.

Recall that in rank-free phylogenetic systematics, monophyly is the source of the part-whole structure, that is, it is by virtue of possessing the property of monophyly that a given collection of basal units (species) constitutes a higher taxon. What is the analog of mono-

phyly in the evolutionary transitions case? The answer is functional integration, cooperation among parts, division of labor, mutual policing and other mechanisms of conflict suppression. These are the features that a collection of smaller units must exhibit to count as a genuine higher-level evolutionary individual (or organism). (A wolf pack, for example, does not count as an evolutionary individual, for it is not predominantly cooperative, whereas a honeybee colony may well do.)

At this point a possible disanalogy may suggest itself. For cooperation, functional integration, and other characteristics obviously come in degrees. Even paradigmatic evolutionary individuals such as modern metazoans aren't perfectly functionally integrated; intraindividual and intragenomic conflict is an ever-present threat, and occurs quite often. Similarly, social insect colonies form a continuum from the highly cooperative and integrative to ones in which conflict, rather than cooperation, is the norm. By contrast, monophyly seems to be an all-or-nothing affair. A given collection of species is either monophyletic or not; there is no gray area, or zone of indeterminacy (though, of course, we may be *uncertain* whether a given taxon is monophyletic). Therefore, entities in the phylogenetic hierarchy seem to have much sharper criteria of identity than those in the ecological hierarchy.

This disanalogy, however, is actually more apparent than real. It is true that *relative to any given phylogenetic tree*, monophyly becomes a yes/no matter. Given the phylogeny depicted in figure 3.1, for example, it is a determinate fact that {D, E, F, G} is monophyletic. But there is a potential source of indeterminacy here that has been swept under the carpet. For lineage splitting is not a perfectly determinate matter; in many cases, it may be unclear whether a particular lineage has split into two, or when the split occurred. So it may be indeterminate which of two competing phylogenetic trees is the better representation of reality. Therefore, the status of any group as monophyletic may itself be indeterminate, which means that the identity criteria of the units in the genealogical hierarchy are not necessarily sharper than of those in the ecological hierarchy.

In phylogenetic systematics, the grouping/ranking distinction helps clarify the old question of whether higher taxa are "real" or "conventional," as discussed in the section on rank freedom in phylogenetic systematics. In the evolutionary transitions case, an analogous clarification ensues in relation to another old question, namely, whether certain biological collectives count as organisms. A wide range of cases exist—social bacteria colonies, marine invertebrate colonies, slime molds, certain cooperative breeding groups, certain insect colonies—in which theorists disagree about whether the collectives in question count as real organisms (individuals), or mere aggregates.[4] The grouping/ranking distinction helps clarify what this question amounts to. The serious issue is whether the collectives exhibit sufficient functional integration to operate as evolutionary units. Do the parts mostly work for the good of the whole collective? Is there extensive division-of-labor among the parts? Are mechanisms in place to regulate the selfish tendencies of the parts and align their interests with those of the whole? Since these features come in degrees, we should not look for a hard-and-fast line. But the important point is that there is no *additional* question about

whether the collectives are "really" organisms; such a question would make sense only if "organism" denoted a rank in the hierarchy. In a rank-free approach, the question of whether something counts as an organism is just the question of whether it satisfies the criteria for inclusion in the hierarchy, at any level, in the first place.

My proposal—that rank freedom be exported from its original home in phylogenetic systematics and applied to the major evolutionary transitions—is not intended as a radical one, nor offered in a revisionist spirit. Rather, I believe that the idea of rank freedom is *already* at work, implicitly, in much theorizing about evolutionary transitions and/or multilevel selection. My suggestion is just that the idea be brought into the open, and the analogy with rank freedom in phylogenetic systematics made explicit. This may not make a huge practical difference, but it could help clarify the ontological commitments of our current biological theorizing. Similarly, the "species are individuals" thesis of Hull (1978) and Ghiselin (1974) did not make much practical difference, but it certainly helped clarify the biology of the day, as numerous biologists have recognized.

Acknowledgments

Thanks to Kim Sterelny, Brett Calcott, Peter Godfrey-Smith, John Wilkins, and Paul Griffiths for comments and discussion. Research for this chapter was supported by a grant from the U.K. Arts and Humanities Research Council, grant no. AH/F017502/1, which I gratefully acknowledge.

Notes

1. This assumes, as is standard, that multicellularity first arose when a single cell divided and its daughter cells failed to detach, rather than by the aggregation of unrelated cells.
2. See <http://www.ohiou.edu/phylocode>.
3. See Sapp (2005) for a detailed history of the eukaryote/prokaryote distinction.
4. Sterelny and Griffiths (1999, 166–177) discuss these problem cases under the heading "collective individuals."

References

Brandon, Robert N. (1988). Levels of selection: A hierarchy of interactors, in Henry C. Plotkin (ed.), *The Role of Behaviour in Evolution*. Cambridge, MA: Cambridge University Press.

Buss, Leo. (1987). *The Evolution of Individuality*. Princeton, NJ: Princeton University Press.

Eldredge, Niles. (1985). *Unfinished Synthesis: Biological Hierarchies and Modern Evolutionary Thought*. New York: Oxford University Press.

Eldredge, Niles (2003). The sloshing bucket: How the physical realm controls evolution, in James P. Crutchfield and Peter Schuster (eds.), *Evolutionary Dynamics* (pp. 3–32). Oxford: Oxford University Press.

Ereshefsky, Mark. (2001). *The Poverty of the Linnean Hierarchy*. Cambridge: Cambridge University Press.

Frank, Steven A. (1995). Mutual policing and repression of competition in the evolution of cooperative groups, *Nature* 377: 520–522.

Ghiselin, Michael T. (1974). A radical solution to the species problem, *Systematic Zoology* 23: 536–544.

Hamilton, William D. (1975). Innate social aptitudes in man: An approach from evolutionary genetics, in Robin Lane Fox (ed.), *Biosocial Anthropology* (pp. 133–155). New York: Wiley.

Hull, David L. (1978). A matter of individuality, *Philosophy of Science* 45: 335–360.

Hull, David L. (1988). *Science as a Process*. Chicago: University of Chicago Press.

Maynard Smith, John, and Eörs Szathmáry. (1995). *The Major Transitions in Evolution*. Oxford: Oxford University Press.

Michod, Richard E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality*. Princeton, NJ: Princeton University Press.

Michod, Richard E. (2005). On the transfer of fitness from the cell to the multicellular organism, *Biology and Philosophy* 20: 967–987.

Michod, Richard E., and Aurora Nedelcu. (2003). On the reorganization of fitness during evolutionary transitions in individuality, *Integrative and Comparative Biology* 43: 64–73.

Mishler, Brent D., and Robert N. Brandon. (1987). Individuality, pluralism and the phylogenetic species concept, *Biology and Philosophy* 2: 397–414.

Okasha, Samir. (2005). Multi-level selection and the major transitions in evolution, *Philosophy of Science* 72: 1013–1025.

Okasha, Samir. (2006). *Evolution and the Levels of Selection*. Oxford: Oxford University Press.

Queller, David C. (1997). Cooperators since life began, *The Quarterly Review of Biology* 72: 184–188.

Queller, David C. (2000). Relatedness and the fraternal major transitions, *Philosophical Transactions of the Royal Society B* 355: 1647–1655.

Reeve, Hudson Kern, and Laurent Keller (eds.). (1999). *Levels of Selection in Evolution*. Princeton, NJ: Princeton University Press.

Sapp Jan. (2005). The prokaryote-eukaryote dichotomy: Meanings and mythology, *Microbiology and Molecular Biology Reviews* 69: 292–305.

Sarkar, Sahotra. (1998). *Genetics and Reductionism*. Cambridge: Cambridge University Press.

Sober, Elliott, and David S. Wilson. (1998). *Unto Others: The Evolution and Psychology of Unselfish Behaviour*. Cambridge, MA: Harvard University Press.

Sterelny, Kim, and Paul E. Griffiths. (1999). *Sex and Death: An Introduction to the Philosophy of Biology*. Chicago: University of Chicago Press.

4 Darwinian Populations and Transitions in Individuality

Peter Godfrey-Smith

John Maynard Smith and Eörs Szathmáry (1995) used the phrase "major transitions" to refer to a set of evolutionary events with particular importance in the history of life. In their original count, eight such transitions were recognized. In many of their cases, though not all, the "transition" involved the appearance of a new kind of entity or biological unit, formed by the merging or combination of simpler ones. These new biological entities include eukaryotic cells, multicellular organisms, and insect societies. Michod (1999) uses the phrase "transitions in individuality" to refer to this particular kind of transition, and those are the ones I discuss in this chapter. But I will approach that topic by way of a general discussion of evolutionary processes.

There is a long tradition of giving abstract summaries of what is essential to evolution by natural selection. These summaries have two roles. One is describing the core of evolutionary theory—or rather, one aspect of the core—in a concise way. The second is guiding the application of evolutionary concepts to new phenomena and problem cases. We see that second goal in the most-cited summary of this kind, given by Lewontin (1970). Lewontin summarizes what he calls "Darwin's scheme" in the form of a recipe for change that can be applied, in principle, to systems of all kinds.

> As seen by present-day evolutionists, Darwin's scheme embodies three principles. . . .
>
> 1. Different individuals in the population have different morphologies, physiologies, and behaviors (phenotypic variation).
>
> 2. Different phenotypes have different rates of survival and reproduction in different environments (differential fitness).
>
> 3. There is a correlation between parents and offspring in the contribution of each to future generations (fitness is heritable).
>
> These three principles embody the principle of evolution by natural selection. While they hold, a population will undergo evolutionary change. (1970, 1)

This is one of a family of such summaries and recipes (see also Endler 1986, Maynard Smith 1988). All known formulations are subject to counterexamples or problems of other kinds (Godfrey-Smith 2007). This is because they tend to be the products of a trade-off

between two theoretical goals that pull in opposite directions. One goal is capturing all genuine cases. The other is describing a causally transparent machine. Nonetheless, the approach I take continues this tradition, as I think the Lewontin-style summaries are on the right track. The first idea I will use is what I call the *minimal concept* of a *Darwinian population*. This is a population in which the constituent individuals show variation in character, differences in reproductive output, and heritability (in the statistical sense). Though the presence of these features does not, as it is sometimes thought, guarantee that change will occur, we can think of these as the basic components in any Darwinian process. The term "Darwinian individual" will be used for any member of a Darwinian population.

The minimal concept has a problematic concept at its heart: reproduction. We need to know what reproduction is in order to work out whether there are fitness differences and heritability, but the concept of reproduction is one surrounded by puzzle cases. Some of those are discussed later. It is worth noting right away, though, that I do not use the concept of a *replicator*, the reproduction-like concept often favored in this sort of discussion. Maynard Smith and Szathmáry themselves discussed evolutionary transitions using the concept of a replicator. Sometimes replicator-based analyses are treated as more-or-less equivalent to three-part summaries in the style of Lewontin, and sometimes they are seen as competitors, depending on how narrowly the idea of "replication" is understood. Maynard Smith and Szathmáry understand the concept broadly, but I believe that the Lewontin-style summaries provide a better starting point. Replication is one kind of parent/offspring relationship, but not the only one that suffices for evolution by natural selection (Godfrey-Smith 2000). The conception of reproduction being developed by Griesemer (2000, 2005), which requires "material overlap" between generations and the capacity for development, is too narrow to use here as well.

The minimal concept gives us a definite starting point, but my aim is to develop a framework that emphasizes gradient concepts, approximations, and the idea that theoretical concepts often work by picking out paradigm cases that are surrounded by a cloud of more marginal ones. This is especially so in a Darwinian context. This idea can be illustrated by looking at a criticism sometimes directed at such formulations as my minimal concept. This criticism is seen in one form in Sterelny and Griffiths's *Sex and Death* (1999). According to this criticism, familiar three-part summaries do not capture the features of evolution by natural selection that make the process scientifically important, because they include both the powerful processes that give us eyes and brains and dull and trivial cases of sorting of fixed types. Therefore, our summary of natural selection should maybe be designed to capture a narrower class, something like the category of *cumulative* selection processes. These involve high heritability, ample variation, a fitness gradient, and perhaps other things as well.

My response is to hang onto the minimal concept, which has its own role, but locate it within a family of concepts. I use the following terminology: *Paradigm* Darwinian populations are those that generate novel, complex, and adaptive traits. The *minimal* concept is a

broader category, picking out all populations with variation, fitness differences, and heritability, including the paradigms but also many others. I also use a third concept, that of a *marginal* Darwinian population. These are not the "dull" cases within the minimal category. Rather, they are cases that do not clearly satisfy but only approximate the minimal requirements. Darwinian patterns of description get some purchase on phenomena that do not pass a classical test, though Darwinian descriptions can also be misleading in those cases. The distinction between minimal-sense, paradigm, and marginal cases can be applied to Darwinian individuals as well.

A Darwinian Space

One way to investigate the relations between these concepts is with a spatial approach, locating different Darwinian populations in an abstract space characterized by evolutionarily important parameters. This requires that we find a range of features that can be represented numerically. Each feature is associated with one dimension of the space. A population, by virtue of how it scores on each dimension at a time, occupies a point in the space. We can then ask whether the paradigm cases cluster in one part of the space and the marginal cases in another. The minimal criteria are supposed to pick out a large region, covering the paradigm cases and shading into the marginal ones. Once the space is constructed, though, the specific categories of paradigm, minimal, and marginal should fade in significance.

The list of dimensions we could include, the smorgasbord offered by recent work on evolutionary transitions and evolvability, is huge. I will pick just a few to look at here. I won't discuss some obviously important factors, like population size and structure, sexual as opposed to asexual reproduction, and various others. Some features are not easily representable in numerical terms, and those are not thereby less important.

In figure 4.1, I include one very familiar dimension, one often discussed but handled differently here, and a newer one. I imagine this as a three-dimensional projection of a higher-dimensional space.

H is an absolute measure of the fidelity of inheritance (not the comparative measure used in the minimal concept). When *H* is high, "cumulative" selection is possible. When *H* is low, the products of one round of evolution tend to be lost on the next, and do not reliably reappear later on. In the case of a sexual and polymorphic population like our own, it is sometimes said that there is low-fidelity inheritance at the level of organisms, but high-fidelity inheritance of genes. I resist the idea that we, as organisms, are low-fidelity inheritors in the sense relevant here. The contrast is with cases in the top-center of figure 4.1, where an "error catastrophe" makes it impossible to retain what evolution has built (Eigen and Schuster 1979). Clearly, we do not see that with humans. The faithful copying of stretches of genetic material is part of the mechanism by which these organism-level properties are achieved.

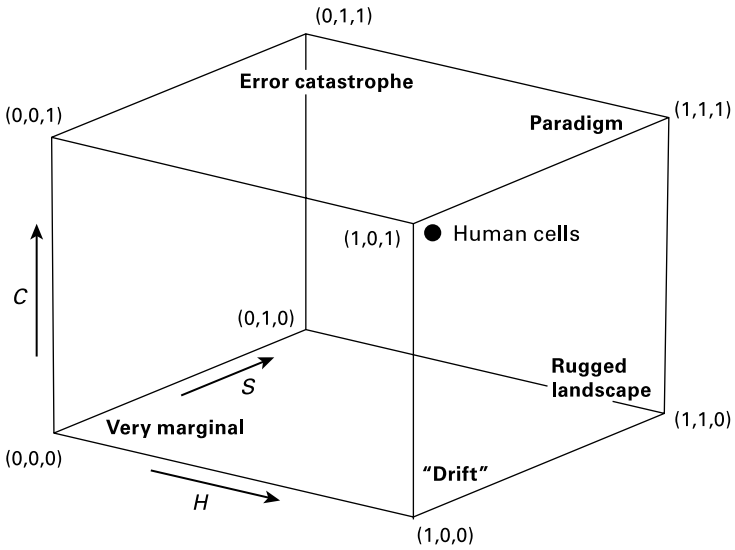


Figure 4.1
Three dimensions of the Darwinian space (*H*,*S*,*C*).

Dimension *S* is newer. I define *S* as the extent to which differences in realized fitness (actual reproductive output) in a population depend on differences in the *intrinsic* character of the members of the population. Intrinsic features—basically, internal make-up and structure—are contrasted with *extrinsic* ones, which involve relations to other things. Location is an example of an extrinsic property (though location may be a causal consequence of an intrinsic property, such as a preference).

Here is an initial justification for including *S*. Mutation and recombination, the paradigm sources of evolutionarily important variation, affect the intrinsic properties of organisms. To the extent that reproductive differences in a population are dissociated from intrinsic characteristics, they cannot be tracking the subtle intrinsic differences that mutation generates. Extrinsic properties such as location are themselves variable among organisms, can generate reproductive differences, and can be heritable (Mameli 2004; Odling-Smee, Laland, and Feldman 2003). Extrinsic factors of organisms are obviously very important in evolution; where an organism lives and who it interacts with largely determine *which* intrinsic features are worth having. But the evolutionary role of extrinsic phenotypic features *per se* is limited. In figure 4.1, the occupant of the area corresponding to a low value of *S* but high values of the other parameters is labeled "human cells." I will explain this later in the section on transitions in individuality.

The third dimension, *C*, represents, roughly speaking, the smoothness of the fitness landscape (Gavrilets 2004; Wright 1932). *C* is high when near-variants in phenotype are similar

with respect to (realized) reproductive output. The symbol comes from Lewontin's term "continuity," used in an early discussion of these issues (1985).

The evolutionary importance of continuity, or reasonable smoothness of fitness landscape, is widely recognized. But *S* and *C* also make possible a new treatment of the relation between selection and "drift." I assume that in real populations, differences in realized fitness will almost always exist, and will have some causal basis or other. Given that there are reproductive differences in a population, the question is where they come from, and what they are correlated with. Suppose *S* and *C* are both very low. As *S* is low, reproductive differences are largely due to extrinsic differences. These extrinsic factors are real features of the organisms; they are part of what fitness is treated as a function of, in the determination of *C*. Extrinsic factors, like intrinsic ones, may have a role that is robust, or one that is fine-grained and chaotic. When *S* and *C* are both very low, reproductive differences are due to factors that are both extrinsic and chaotic. These are the cases that induce talk of "drift"; recall the familiar example of two intrinsically similar organisms, one struck by lightning. (The label "drift" can also seem appropriate to some extent when only one of *S* or *C* is low, but those are not the clearest cases.)

So *S* and *C* have general importance; their use is motivated mainly by what happens at nonextreme values. But then we can note that at extremely low values, we find one famous kind of non-Darwinian phenomenon, "drift"-like change.

Figure 4.1 does not include any measure of the abundance of variation, but this is obviously crucial and several different measures of variation seem relevant. One is the amount of variation present at a time—the immediate raw material of evolutionary change. Another is the dispositional property a system may have, of reliably tending to produce new variation as it is consumed by selection. As discussed later, it is also common to emphasize some kind of "quasi-independence" of variation, or "modularity" in the effects of variation on the phenotype. Other important features are the size of a typical deviation and the absence of bias. But here I will make use of only the most basic of these features. *V* will represent the amount of variation in a population at a time.

Some parameters we would want to include are not independent of each other, either at extreme values or more generally. For example, if *H* is perfect, no new variation appears, though existing variation will be retained. A variety of causal relationships also exist between the features.

In this section I have shifted emphasis away from the discrete categories introduced earlier (paradigm, minimal, marginal) to a network of relations between populations, with respect to their evolutionarily important parameters. And as a population evolves, it not only changes the characteristics of the organisms within it, it also changes how it evolves in the future. Populations move through the space. A population can evolve a higher or lower *H*, by evolving better or worse suppression of mutation via DNA editing. It can evolve heat-shock proteins that increase *C*. It can move to a new environment full of capricious and lethal risks that reduce both *C* and *S*.

Forms of Reproduction

Earlier I noted the role of the concept of reproduction, which figures in the minimal concept. What *is* reproduction? We might give an initial analysis by saying that reproduction involves (i) the production of a new individual, (ii) of the same general kind as other members of the population, (iii) primarily via the causal role of particular preexisting individuals in that population. Reproduction can then be contrasted with (i) growth of the same individual, (ii) the production of waste and artifacts, and (iii) production of a new individual without it being the product of particular parents. This is a "shallow" analysis of reproduction. It uses terms that are themselves problematic, and may have to be revised under the influence of theory later on. But it is a start.

There are two well-known families of problem cases to grapple with.

1. *Reproduction versus growth* When is the production of new biological *material* the production of a new biological *individual*? The problem is most acute with plants and colonial organisms, and in the absence of sex.

2. *Collective entities* When does the production of a new collective entity (such as a colony, herd, or symbiotic association) count as reproduction at that higher level?

In addressing these problems, I avoid making any particular criteria essential to reproduction. Instead, I apply the gradient approach used in the previous section. Owing to the role of several biological features, cases of reproduction can be clearer or more marginal. This analysis may then be connected to the analysis used in the previous section; marginal Darwinian processes often involve marginal forms of reproduction.

Returning to the contrasts between reproduction and other phenomena introduced earlier, a case of reproduction might be marginal because (i) it is not clear that new individuals are being produced, as opposed to new parts of old individuals, or because (ii) the population is made of dubious higher-level entities that have little standing as individuals in their own right. Alternatively, it might be because (iii) the causal relation linking "parent" and "offspring" is not clear. (That last possibility applies often in the case of cultural evolution.) At different places in the tree of life, we find lots of different reproduction-*like* phenomena. This is for Darwinian reasons; the forms taken by the creation of new biological material from old are consequences of contingencies of ecology and history in different parts of the tree.

In this chapter, I discuss three reproduction-related features. They are designed to deal with the families of problems raised earlier: distinguishing growth from reproduction, and recognizing reproduction in higher-level entities. These two problems are linked, as growth-versus-reproduction problems usually involve either modular organisms or colonies (aspen, corals, fungi, etc.), and certainly such problems usually arise when at least multicellularity is present. So the treatment given here may apply generally to all cases of

"collective reproducers" (Godfrey-Smith 2009), though the framework may not be appropriate for some other sorts of cases.

The first parameter I use is *B*, which stands for "bottleneck." A clear case of reproduction, at least when dealing with collectives rather than things like cells, narrows to mark a divide between generations. This might be understood absolutely, or as some measure of the relation between propagule and adult size. A bottleneck enables a process of growth and development to begin anew, so a localized mutation can have a multitude of downstream effects (Dawkins 1982; Harper 1977). I understand *B* as existing in all degrees, not as marking a distinction between one-celled beginnings and everything else.

The second feature is *G*, or "germ/soma," which is understood more broadly as the degree of reproductive specialization in the collective's parts. When *G* is high, many parts of a reproducing entity are unable to become the basis of a new entity of the same kind. Only a few parts have that role. This feature seems particularly relevant when analyzing reproduction in colonies, as will be discussed further later. The third feature I will discuss is "integration," or *I*. This one is vaguer. It involves such features as division of labor (aside from the kind involved in *G*), mutual dependence of parts with respect to viability, and maintenance of a boundary between individual and environment.

In another work (2009) I provide a three-dimensional chart that categorizes various cases according to *B*, *G*, and *I*. Here I discuss just two of these dimensions, *G* and *I*, and offer an example with special relevance to questions about evolutionary transitions.

The colonial green algae in the clade that includes the *Volvox* organisms are often seen as a very informative case for the study of the evolution of multicellularity (Kirk 1998, 2005; Michod 2005; Michod et al. 2003). These organisms, commonly found in ponds, perform both sexual and asexual reproduction. A sexually produced cell divides repeatedly to produce a colony, which may be of various sizes and degrees of organization. The colonies swim using their members' flagella, migrating to shallow water during the day and collecting nutrients in deeper water at night. When food is plentiful, the colonies reproduce asexually; new colonies are formed inside the old from single initiating cells. The new colonies bud off or are released from inside the old colony. When food becomes scarce, they enter a sexual cycle, producing "zygospores," which lie dormant until times are better. Here, I consider only their asexual mode of reproduction.

In figure 4.2, all the colonies are formed from a single-celled bottleneck—all share a high value of *B*. They differ with respect to *G* and *I*. (A version of this figure was sketched initially by Rick Michod. That should not be taken to imply that he endorses the framework used here.)

The distinctions between GS, GS/S, and G/S states on the *G* axis follow Michod's framework (with apologies for my double use of the letter "S"—I have not italicized the letter in this second use). A GS colony has no reproductive division of labor at all; all cells carry out a similar mix of "somatic" and reproductive roles, though not simultaneously. In these organisms, a central nonreproductive function is locomotion, via the action of the flagella. A

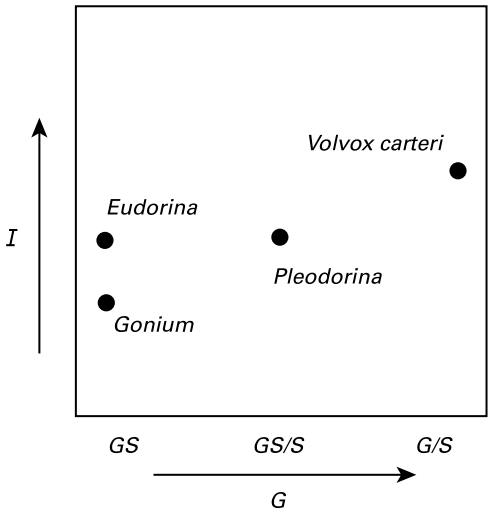


Figure 4.2
(*G*,*I*) comparisons for some colonial green algae.

particular cellular structure (basal body) is needed for both cell division and use of the flagella, so this determines a trade-off (Buss 1987), which is temporally organized in GS organisms. In the GS/S case, we have somatic cells plus cells that play both roles, Typically, the dual-role cells act "somatically" in locomotion first and then become reproductive. In the G/S cases on the right, there is a definite distinction between somatic and germline cells.

Gonium, on the far left, consists of loosely organized colonies of 8 to 16 cells. *Gonium* is basically a flat clump of cells, but with some spatial organization of flagella action. *Eudorina*, in contrast, has 32 cells and is spherical, with a distinction between inside and outside. Both organisms are GS.[1] *Pleodorina* colonies consist of 64 to 128 cells. They have the partial GS/S reproductive division of labor, and the somatic cells are localized to one part of the structure. *Volvox carteri*, on the right, is larger (2^{12} cells) and more integrated—many would say it counts clearly as an organism. It also has a clear germ/soma distinction, with the vast majority of cells allocated to a purely somatic role early in development.

It would be possible to add more cases, though their plotting on the chart might be controversial. *Volvox rousseletii* is larger than *V. carteri*, as it has 2^{15} cells. These colonies are GS/S, like *Pleodorina*, but might be distinguished with respect to dimension *G* because the reproductive cells in *V. rousseletii* have flagella only for a very short time, less than a day, before abandoning this set of functions. *V. rousseletii* is a more adept swimmer than *V. carteri*. It also has cytoplasmic bridges linking the cells when mature, which are lost during development in the case of *V. carteri*. These might be seen as a mark of higher integration

than we find in *V. carteri* (though Michod, personal communication, is wary of that comparison). So it would be possible to plot *V. rousseletii* as higher than any other case with respect to *I* and intermediate in *Pleodorina* with respect to *G*. But perhaps it should be intermediate between *Pleodorina* and *V. carteri* with respect to *G*, and it is not clear that it is higher than the *V. carteri* with respect to *I*. So I included only the clearer cases in the figure.

Comparison with Sterelny

In this section I compare my framework to one that Sterelny has discussed in several papers (2001, 2004), and revisits in his contribution to this volume. Sterelny's aim is to describe "enriched Darwinian environments . . . the characteristics of individuals, populations and environments in which the evolution of novelty and disparity is possible." Sterelny is more focused on the "high end" of the range of cases that I am discussing, and treating that region in more detail. But there are some relevant points of comparison. His conditions for "enrichment" of Darwinian processes are as follows:

Anti-Outlaw Conditions

1. Replicators should be transmitted vertically. Replicators should flow from parents to offspring, and to them alone.

2. Replicators should be transmitted simultaneously.

3. The transmission of the replicator set should not be biased. Either all an organism's replicators are transmitted to each descendent, or each replicator has an equal chance of being transmitted to each descendent.

Stability Conditions

4. The copy-fidelity of the generation of replicators from generation to generation should be high.

5. The replicator/organization map should be robust. To the extent that the causal channel from replicator to organization depends on context, both internal and external, that context should be stable and predictable.

Generation of Variation

6. The array of possible replicator sets should be very large; possibly even unbounded.

7. The effect of a replicator on the biological organization of its carrier should normally be well-behaved. That is, the replicator/organization map should be smooth. A map is smooth if a small change in the replicator set generates a small change in biological organization; and the smoother the map, the more evolvable the lineage using that inheritance channel. Moreover, the relationship between phenotype and fitness should be smooth: if T* is fitter than T, and T# is phenotypically intermediate between T* and T, its fitness should be intermediate, too.

8. The generation of biological organization from the replicator set should be modular. The replicators as a whole should not generate the biological organization of the organism as a whole. Rather, replicators, or small sets of replicators, should be designed so that they make a distinctive contribution to the generation of one or a few traits, and relatively little distinctive contribution to others.

One difference between us is that Sterelny assumes a replicator framework—not that the organisms in an evolving population are replicators, but that their inheritance mechanisms work through the physical transfer of replicators. If this is an application of the idea that Darwinian phenomena in general require replicators, then I think it is a mistake, even within a liberal conception of replication. Alternatively, Sterelny might be assuming, in the background, that *significant* evolutionary processes all involve replicators. In that case, "inheritance works via the transfer of replicators" becomes his condition zero, the first big distinction between different Darwinian phenomena. Either way, the treatment of replicators has the consequence that, from my point of view, Sterelny is discussing the relations between two Darwinian populations at once: a population of organisms and a population of underlying replicators which that undergo their own evolutionary activities. His "outlaw conditions" are concerned with such relations. There is no mention of bottlenecks or germ lines here; later I will discuss their role as "anti-outlaw" mechanisms. In fact, his third condition must be understood carefully if it is not to be incompatible with a germline requirement; it must mean all the replicators *initially* present. There is trouble if the replicators transferred are representative of changes during ontogeny.

Sterelny's Conditions 4 and 5 together yield high *H* in my sense. We have faithfully copied replicators, with stable effects on phenotype. So Sterelny breaks *H* into two components. His sixth condition is a standard condition on variation that I would also include in some form.

Conditions 7 and 8 are both linked to my *C*. Specifically, the last part of Condition 7 *is* my *C*, and the early parts of 7 are *means* to *C*. This recalls Lewontin's original discussion of "continuity"; he said that similar phenotypes should yield similar "ecological relations," in turn yielding a smooth fitness landscape (Lewontin 1985).

Condition 8 seems to me to duplicate part of 7. This is because modularity has the consequence that a small change to what is inherited implies a local change to phenotype. So I worry that Sterelny may be treating parts of a causal chain involved in *C* as if they were distinct requirements. Modularity is one means to a situation in which small changes to inherited resources imply small changes to phenotype. That, in turn, is a good way to ensure that similar organisms have similar fitness. Some of these connections are near to deductive, others are looser. Given the loose connections, Sterelny may be right that many of these factors are worth listing separately. But I suspect there is probably some partial duplication of the criteria having to do with smoothness of fitness landscape, through separately requiring both the outcome and some of its preconditions.

Transitions in Individuality

I now turn to the "major transitions in evolution." Most of these transitions, but especially what Michod calls "transitions in individuality," involve the appearance of new Darwinian

populations from old. Often this occurs via collective entities acquiring a genuine status as Darwinian individuals, that is, as members of Darwinian populations.

The analysis I have in mind works like this: We take a permissive attitude to the concept of a Darwinian population itself. Many can be recognized—some inside of others, some that look like mere byproducts of the activities of lower-level populations, some with odd shapes and boundaries. In many cases, it may appear that the "real" evolutionary action is going on somewhere else. And action elsewhere may indeed by going on. But these dubious cases will often nonetheless pass or at least approximate the minimal Darwinian criteria discussed earlier.

So Darwinian populations exist at many levels.[2] Returning to the spatial analysis used earlier, there are all sorts of dubious and apparently artifactual cases that can be located *somewhere* in such a space. But populations also move through the space, as a consequence of the evolution of new genetic and phenotypic features in the individuals themselves. This may include movement from marginal to paradigm status, or from paradigm to marginal. Movement of that kind is often a consequence of evolutionary change to how reproduction works. What were formerly mere collections of lower-level entities can become significant Darwinian individuals in their own right. As I will argue, movement of a higher-level population into "better" parts of the space often occurs through movement of another population—the lower-level one—into a "worse" part of the space.

Let us imagine a schematic evolutionary transition, having the flavor of a transition to multicellularity, of the kind discussed by Maynard Smith, Szathmáry, Michod, and others. We assume a population of lower-level entities that come to interact in collectives, either by association or by failure to separate after reproduction. Their association may initially be loose. But cooperation develops. Cooperation requires overcoming subversion problems. This may lead to the appearance of a series of adaptations that suppress lower-level competition.

Two characteristics are often discussed in this connection: bottlenecks and germlines (Buss 1987; Grosberg and Strathmann 1998; Michod 1999; Wolpert and Szathmáry 2002). Features like these can be visualized as producing two kinds of movement in a space defined by Darwinian parameters. First, the population of collectives, formerly marginal, moves closer to the paradigms. High values of B and G are associated with clearer instances of reproduction. The second change concerns the lower-level entities, which here we assume to be cells. The evolution of higher values of B and G at the collective level suppresses or curtails evolutionary activities at the cell level. These changes turn cells into a *less* significant Darwinian population, pushing them away from the paradigm region.

This happens in several ways. First, bottlenecks impose uniformity at the start of the life cycle of the collective entities. This reduces the scope for evolution at the lower level. This is a reduction in variation (V) for the population of cells found within a multicelled organism. The only genetic variation that can arise has to appear by mutation from an initially

uniform genotype, or perhaps by mitotic recombination or lateral gene transfer. Epigenetic variation can accumulate much more quickly.

Higher values of *B* reduce the *scope* for low-level evolution. Evolution of a germline has the further effect of making certain kinds of within-collective evolution *irrelevant* to longer-term evolutionary processes.

In my framework, this is a reduction in *S* at the cell level. To see this, think about the situation fairly late in ontogeny for a multicellular organism like one of us. There is an array of cells with different genotypes as a consequence of mutation (and different epigenotypes as well). Which cells are fitter than others? Some may reproduce faster than others, commandeering more resources, and so on. But if we ask which cells have the chance of giving rise to a long lineage of descendents, then these intrinsic differences have limited importance. What matters, instead, is location, the extrinsic property of being, or not being, in the germline.

So from the lower-level point of view, bottlenecks and germlines are "de-Darwinizing" elements. They make the collection of lower-level entities into a less significant Darwinian population. Whether or not this is the evolutionary *function* of bottlenecks and germlines, in the historical sense, it is something they in fact *do*.

Here, I will not take sides on the many interesting questions about the evolution of bottlenecks and germlines, but I will discuss some features of the landscape of possibilities in a bit more detail. In an asexual population, there can in principle be either without the other. A bottleneck without a germline is possible if there is spore-like or apomictic asexual reproduction. Reproductive specialization of the germ/soma kind without a bottleneck is also possible in principle; a large multicellular propagule might derive from a specialized reproductive structure. We might wonder whether there is much point in high *G* without high *B*, however. Then there may be genetic variance within the propagule, even though somatic evolution after sequestration is irrelevant. The result is the continuation in a small and special arena of the same sort of competitive process that germ/soma specialization usually acts to suppress (Michod and Roze 2001). An organism would carry into the next generation a competition inherited from the previous one.

Things are different in a sexual population with fusion of gametes. In a sexual population of this kind, bottlenecks without germlines are still possible. But a germline without a bottleneck seems even more implausible than in the asexual case (Grosberg and Strathmann 1998; Wolpert and Szathmáry 2002). Even if all the cells in a particular parent contributing gametes to a large propagule were themselves very genetically similar, the gametes themselves would differ, owing to the many separate events of segregation and recombination. Sex magnifies the problem of internal conflict discussed earlier.

Both pairs of authors cited above raise other possible problems with the possibility of a large sexual propagule, unrelated to the problem of internal competition. Grosberg and Strathmann suggest mechanistic problems with the coordination of cells during syngamy and early development. Wolpert and Szathmáry claim that it would be hard for such organ-

isms to have a coherent developmental program, as the organized differentiation of cell lineages in development requires that "all the cells have the same set of genes and obey the same rules" (745). Slime molds are, when we think of the multicellular fruiting body as the organism, highly sexual with many parents. But their developmental sequence is very simple, indeed. Wolpert and Szathmáry do not think that this argument from the possibility of complex developmental programs involves an inappropriate appeal to long-term consequences rather than short-term advantage, though they do gesture toward something like the culling of clades, rather than ordinary microevolution, for organisms with small propagules to prevail.[3]

Sex (in some form) is ancestral in all major multicellular lineages, though Grosberg and Strathmann do not think this itself imposes too tight a constraint, and hold that a functional rationale for bottlenecks should still be sought. In any case, though all these discussions are presented as speculative, the upshot is that various arguments make it unsurprising that bottlenecks without high reproductive specialization are common, but not the converse.

In sum, some key adaptations on the road to complex multicellularity have the effect of moving collective entities toward paradigm status as Darwinian individuals, by giving them a clearer form of reproduction, and moving lower-level entities away from paradigm status. The lower-level entities are partially "de-Darwinized" by the transition process. In many cases (such as cells) the lower-level entities still engage in a clear form of reproduction, but they come to score lower in other ways.

Comparison with Michod

I will contrast the preceding treatment with some of Michod's recent ideas about evolutionary transitions (2005, 2006, this volume). Michod gives a foundational treatment in quite different terms from me, and the contrasts are informative about the different possible ways of setting things up.

Michod claims that an evolutionary transition in individuality (ETI) involves the "transfer" or "export" of fitness from the lower level to the level of the collective. The evolution of multicellularity is his main example.

> During ETIs, the heritability of fitness for the new higher level must increase, while, at the same time, it must decrease for the lower-level units. This requires the *reorganization of fitness* . . . , by which we mean the transfer of fitness from the lower-level units to the new higher-level unit and the specialization of lower-level units in the fitness components of the higher-level unit. (2005, 968)
>
> . . . The evolution of cooperation is fundamental to ETIs, because it exports fitness from the lower level (e.g., its costs to cells) to the higher level (its benefits to the group) and in this way cooperation may create new levels of fitness. (2005, 969)

My first point is that this talk of export and transfer seems rather metaphorical. What is meant in more literal terms? So let us look at a more specific argument Michod makes

about what happens to lower-level fitnesses as a transition occurs. When reproductive specialization occurs in a transition to multicellularity, the fitness of the lower-level entities (cells) goes to zero. That means they cannot engage in a Darwinian process.

> Consider the case of multicellular organisms with complete G-S specialization. The germ cells specialize completely in reproductive functions and the somatic cells specialize completely in vegetative functions. The cell fitness of all cells must be zero (since fitness is the product of viability and reproduction and one of these is zero by the assumption of complete G-S specialization). Therefore, the fitness of the group is zero under MLS1, yet group fitness may be quite high under MLS2. (2005, 970)

> . . . Once the specialization is complete and the lower level units are specialized in one of the two major fitness components (viability or fecundity), they have no fitness by themselves and so group fitness in the sense of MLS1 is null, while group fitness in the sense of MLS2 may be quite high. (2005, 976)

For Michod, it is a general fact that fitness is a combination of viability and fecundity, as discussed in life-history models. Specifically, it is the product of these two factors, so if either is zero, overall fitness is zero. The argument is that, when we have complete germ/soma specialization, the fecundity of the somatic cells is zero, and the viability of the germline cells is zero. So no fitness is present at the lower level, and hence no Darwinian process at that level.

I do not think it is true that when germ/soma differentiation occurs, these cell-level viabilities and fecundities go to zero, on any natural kind of accounting. Germline cells do live for a time, and divide at the end of a period of remaining viable. Somatic cells do, in many cases, divide. They do not divide in the case of the *Volvox carteri* that Michod studies, but I assume he does not intend his argument to depend on that fact. I assume he sees the argument as applying generally to organisms with germ/soma specialization. But in general terms, surely both germline and somatic cells do the same sort of thing from a Darwinian point of view: Both kinds of cells will live for a while, and then divide or die.

Of course, Michod does not intend to deny any those assertions about what germline and somatic cells do. He accepts that both kinds of cells in organisms like us are able to live and reproduce. What Michod is guided by is a functional difference between germline and somatic cells. But that functional contrast has to do with the effects of different cells on whole-organism fitness: The somatic cells aid in maintaining whole-organism viability (though not only that—they also support reproductive efforts). Germline cells do not contribute to maintenance of viability. This functional contrast has to do with the cells' relations to the collective's fitness, not their own.

Given that, I suggest that when we are considering the fitness and other Darwinian properties of lower-level entities, such as cells, we should assess the cells' relations to the standard criteria for being a Darwinian population *in their own right*, on their own terms. The functional contrast discussed earlier does not prevent cells in a multicellular organism from

meeting, for a time, the criteria for making up a Darwinian population. It does not stop them from actually living, varying, reproducing, and potentially passing on—for a while at least—favored traits.

There are, in fact, two ways of looking at cells in such a case. We can think of the cells within each organism as making up a small and short-lived Darwinian population, and we can also collect all human cells together and treat them as making up one larger population. Both kinds of analysis are possible. It is true that the cells in an organism like us depend on other cells to stay alive and reproduce; they have come to inhabit a particular kind of environment. But in that environment, they do in fact survive and reproduce—they engage in ordinary Darwinian activities. Some are fitter than others. With respect to their relationships to the basic Darwinian criteria, human cells are no different from bacterial cells and protists. When we look in more detail at what *kind* of Darwinian population the cells within you, or the totality of human cells, form, then we do find they have special features of the kind discussed in the previous section. But to acknowledge these evolved features is not to say that cells in organisms like us, despite their ability to survive and reproduce, have no fitness at all.

Michod's models are informative. They feature a germ/soma specialization pathway that does not go via the subversion problem, but via ecological trade-offs between reproduction and other functions. Might it be that subversion prevention is sometimes an incidental by-product of a germ/soma specialization that tends to come about for other reasons? We are already used to that sort of byproduct explanation, as noted earlier, as a result of thinking about bottlenecks and their consequences. The models might be taken on board independently of Michod's commentary about them.

Conclusion

The project of this chapter has been to start from a general and independently motivated analysis of Darwinian processes, extend it so it can accommodate some extra distinctions (paradigm and marginal cases, etc.), and then apply it to some problems raised by evolutionary transitions. Many earlier discussions in this area have used the concept of a replicator, rather than a three-part summary in the more traditional style exemplified by Lewontin (1970). The three-part summaries are, I suggest, both superior in general and also quite useful for thinking about the major transitions. I have argued that attention to the concept of reproduction is particularly fruitful here. In many cases, the mechanisms of reproduction are crucial to the evolutionary properties of both higher-level Darwinian individuals, like multicellular organisms, and the lower-level Darwinian individuals from which they arose. Bottlenecks and germlines, in particular, have complementary roles for each kind of entity, contributing at once to the Darwinization of the higher-level individuals and the partial de-Darwinizing of the lower.

Notes

1. Kirk qualifies this. Under most circumstances all *Eudorina* cells divide, but "under some circumstances," the four anterior ones remain somatic (2005, 305).

2. When I talk of "levels" of selection here, roughly speaking, I mean this in what is known as the "MLS2" sense, not the "MLS1" sense (Damuth and Heisler 1988; Okasha 2006). Group selection, for example, requires reproduction *of groups*, and heritability at the group level. We apply the standard Darwinian criteria at all levels and in all cases. So differential fitness as a feature of entities at level *N* involves differential reproduction of entities at level *N*.

3. "[T]aking the broad view in evolutionary terms, organisms that develop from an egg would displace those that do not" (2002, 745).

References

Buss, L. W. (1987). *The Evolution of Individuality*. Princeton, NJ: Princeton University Press.

Damuth, J., and I. L. Heisler. (1988). Alternative formulations of multilevel selection. *Biology and Philosophy* 3: 407–430.

Dawkins, R. (1982). *The Extended Phenotype: The Gene as the Unit of Selection*. Oxford: W. H. Freeman.

Eigen, M., and P. Schuster. (1979). *The Hypercycle: A Principle of Natural Self-Organization*. Berlin: Springer-Verlag.

Endler, J. A. (1986). *Natural Selection in the Wild*. Princeton, NJ: Princeton University Press.

Gavrilets, S. (2004). *Fitness Landscapes and the Origin of Species*. Princeton, NJ: Princeton University Press.

Godfrey-Smith, P. (2000). The replicator in retrospect. *Biology and Philosophy* 15: 403–423.

Godfrey-Smith, P. (2007). Conditions for evolution by natural selection. *The Journal of Philosophy* 104: 489–516.

Godfrey-Smith, P. (2009). *Darwinian Populations and Natural Selection*. Oxford: Oxford University Press.

Griesemer, J. (2000). The units of evolutionary transition. *Selection* 1: 67–80.

Griesemer, J. (2005). The informational gene and the substantial body: On the generalization of evolutionary theory by abstraction. In M. Jones and N. Cartwright (eds.), *Idealization XII: Correcting the Model, Idealization and Abstraction in the Sciences* (pp. 59–115). Amsterdam: Rodopi.

Grosberg, R. K., and R. R. Strathmann. (1998). One cell, two cell, red cell, blue cell: The persistence of a unicellular stage in multicellular life histories. *Trends in Ecology and Evolution* 13: 112–116.

Harper, J. L. (1977). *Population Biology of Plants*. London: Academic Press.

Kirk, D. L. (1998). *Volvox: Molecular-Genetic Origins of Multicellularity and Cellular Differentiation*. Cambridge, New York: Cambridge University Press.

Kirk, D. L. (2005). A twelve-step program for evolving multicellularity and a division of labor. *Bioessays* 27: 299–310.

Lewontin, R. C. (1970). The units of selection. *Annual Review of Ecology and Systematics* 1: 1–18.

Lewontin, R. C. (1985). Adaptation. In R. Levins and R. C. Lewontin (eds.), *The Dialectical Biologist* (pp. 65–84). Cambridge, MA: Harvard University Press.

Mameli, M. (2004). Nongenetic selection and nongenetic inheritance. *British Journal for the Philosophy of Science* 55:35–71.

Maynard Smith, J. (1988). Evolutionary progress and levels of selection. In M. H. Nitecki (ed.), *Evolutionary Progress* (pp. 219–230). Chicago: University of Chicago Press.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution*. Oxford: W. H. Freeman.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality*. Princeton, NJ: Princeton University Press.

Michod, R. E. (2005). On the transfer of fitness from the cell to the multicellular organism. *Biology and Philosophy.* 20:967–987

Michod, R. E. (2006). The group covariance effect and fitness trade-offs during evolutionary transitions in individuality. *Proceedings of the National Academy of Sciences USA* 103: 9113–9117

Michod, R. E., A. M. Nedelcu, and D. Roze. (2003). Cooperation and conflict in the evolution of individuality IV: Conflict mediation and evolvability in *Volvox carteri*. *BioSystems* 69: 95–114.

Michod, R. E., and D. Roze. (2001). Cooperation and conflict in the evolution of multicellularity. *Heredity* 81: 1–7.

Odling-Smee, F. J., K. N. Laland, and M. W. Feldman. (2003). *Niche Construction: The Neglected Process in Evolution*. Princeton, NJ: Princeton University Press.

Okasha, S. (2006). *Evolution and the Levels of Selection*. Oxford: Oxford University Press.

Sterelny, K. (2001). Niche construction, developmental systems and the extended replicator. In R. Gray, P. Griffiths, and S. Oyama (eds.), *Cycles of Contingency: Developmental Systems and Evolution* (pp. 333–349). Cambridge, MA: MIT Press.

Sterelny, K. (2004). Symbiosis, evolvability, and modularity. In G. Schlosser and G. P. Wagner (eds.), *Modularity in Development and Evolution* (pp. 490–518). Chicago: University of Chicago Press.

Sterelny, K., and P. E. Griffiths. (1999). *Sex and Death: An Introduction to the Philosophy of Biology*. Chicago: University of Chicago Press.

Wolpert, L., and E. Szathmáry. (2002). Multicellularity: Evolution and the egg. *Nature* 420:745.

Wright, S. (1932). The roles of mutation, inbreeding, crossbreeding and selection in evolution. *Proceedings of the Sixth International Congress of Genetics* 1: 257–266.

5 Evolvability Reconsidered

Kim Sterelny

Darwinian Environments

The history of life is not just a history of evolution; it is a history of extraordinarily fecund evolutionary change. Many lineages have seen the evolution of complex adaptive structures, including completely novel structures: complex morphological innovations in the macrobes (sensory systems; locomotion; internal structural systems for circulation and support) and metabolic innovations in the microbes (nitrogen fixation, photosynthesis). Others lineages have histories of extraordinary diversification; most obviously the legendarily speciose beetle lineage. Perhaps most strikingly of all, there has been directionality in evolutionary history. As *Major Transitions* documents, there has been an evolutionary trend of a special kind: the evolution of new forms of organization—the eukaryotic cell, multicellularity, eusociality. These transitions expand the space of biological possibility (Maynard Smith and Szathmáry 1995). The upper bound on complexity is not fixed; rather, it is moved by such key innovations (Sterelny 1999).

So taken as a whole, life on earth has been impressively evolvable. It is disparate in basic metabolic style, in species richness, in morphology, in behavior, and in ecological role. But lineages are not *equally evolvable*. Some of the great, long-lived lineages of animals have been astonishingly diverse and disparate: most obviously, the arthropods. Others have been much less so. The tardigrades, for example, come in at not much more than 400 species, with limited morphological and ecological variation (Brusca and Brusca 1990); the placozoa are even more limited in (current) diversity and disparity. The Metazoan radiation itself has resulted in a lineage incomparably more diverse and disparate than most other multicelled lineages. Multicelled life has arisen many times, but from these twenty-plus lineages, there have been only three immense radiations of diversity (Bonner 1998). To shift from the macrobes to the microbes, almost all the great metabolic innovations—aerobic respiration, sulfur-oxidation, photosynthesis—have been bacterial innovations. The equally ancient Archaea seem to have been limited to methanogenesis, except when they have imported bacterial innovations by horizontal gene transfer (Woese 2008). Moreover, evolutionary possibility has varied over time: Bacterial associations may be ancient, but for the

first few billions years of life, no complex, enduring multicelled systems evolved. Thus, lineages vary in their capacity to generate complex novelty, taxonomic diversity, and new levels of organization. Lineages seem to vary in *evolvability*. That is, they vary in their existing reservoirs of variation available to selection, and, very plausibly, in their potential to generate new variation. There is an issue, though, as to whether these differences in the pattern of evolution across time and across lineages have a systematic underlying cause; whether there is a single difference (or small set of interrelated differences) between the disparate, diverse lineages and the also rans. One aim of a theory of evolvability is to identify just such a systematic underlying cause.

One approach to a theory of evolvability is to upgrade an existing project within evolutionary biology. There is an impressive history in evolutionary biology of attempts to characterize the general conditions under which evolution takes place. These projects characterize "Darwinian environments"; that is, they specify relationships between individuals, populations and environments that are necessary and sufficient for a population in such an environment to evolve. Perhaps the most famous specification comes from Richard Lewontin (1970). According to his recipe, a population that contains individuals that vary from one another; that reproduce with differing prospects for success, and that reproduce in such a way that offspring resemble their parents, will evolve. It turns out that characterizing the conditions on evolution in their full generality is demanding, and that Lewontin's conditions are best seen as an approximate specification of a Darwinian environment (Godfrey-Smith 2009; Okasha 2006).

Even setting these complications aside, Lewontin's approach is deliberately minimalist. It is an attempt to capture conditions necessary for any form of evolutionary change. They certainly do not guarantee that the conditions for the evolution of complexity are satisfied. So, for example, these conditions do not require that variations will continually arise, nor that the selective environment will be well behaved. So one way a theory of evolvability has been built is by enriching these minimal conditions; a theory of evolvability identifies the extra elements that need to be added to these minimal conditions to explain how the evolution of a complex, disparate, and highly adapted biota is possible. One form of a theory of evolvability is a specification of *enriched Darwinian environments*, identifying the characteristics of individuals, populations and environments in which the evolution of novelty and disparity is possible.

In my own recent work, I have attempted to characterize such enriched Darwinian environments; later, I reproduce a set of such "evolvability conditions" from these attempts (Sterelny 2001, 2004). Importantly, this set of conditions was not intended to be original or controversial. Rather it was intended to be a consensus synthesis of the existing literature. The conditions fall into three clusters. The first set characterize inheritance systems that would be insulated from subversion by selfish genetic elements (and their equivalent in other replication systems). As Godfrey-Smith points out, the components of a Darwinian agent can themselves be Darwinian agents forming a Darwinian population. Notoriously,

when they do, the results of within-agent competition and evolution tend to degrade the fitness of the agent. So in Godfrey-Smith's framework, these first three conditions are "de-Darwinizing" conditions; when satisfied, they ensure that the elements responsible for development and heredity do not themselves form a Darwinian population (Godfrey-Smith 2009; this volume). The second and third sets specify ideal conditions on the supply of usable variation of inherited resources, and on the use of those resources.

Anti-Outlaw Conditions

C1. Replicators should be transmitted vertically. Replicators should flow from parents to offspring, and to them alone.

C2. Replicators should be transmitted simultaneously.

C3. The transmission of the replicator set should not be biased. Either all an organism's replicators are transmitted to each descendent, or each replicator has an equal chance of being transmitted to each descendent.

Stability Conditions

C4. The copy-fidelity of the generation of replicators from generation to generation should be high.

C5. The replicator/organization map should be robust. To the extent that the causal channel from replicator to organization depends on context, both internal and external, that context should be stable and predictable.

Generation of Variation

C6. The array of possible replicator sets should be very large; possibly even unbounded.

C7. The effect of a replicator on the biological organization of its carrier should normally be well behaved. That is, the replicator/organization map should be smooth. A map is smooth if a small change in the replicator set generates a small change in biological organization; and the smoother the map, the more evolvable the lineage using that inheritance channel. Moreover, the relationship between phenotype and fitness should be smooth: if T* is fitter than T, and T# is phenotypically intermediate between T* and T, its fitness should be intermediate, too.

C8. The generation of biological organization from the replicator set should be modular. The replicators as a whole should not generate the biological organization of the organism as a whole. Rather, replicators, or small sets of replicators, should be designed so that they make a distinctive contribution to the generation of one or a few traits, and relatively little distinctive contribution to others.

As I now see it, this characterization of evolvability is misleading in three ways. First, it leaves out something very important: developmental plasticity. Second, the approach is too static: These conditions themselves have an evolutionary history that a theory of evolvability needs to capture. Finally, without saying anything false, this formulation masks the fact that evolvability (as we shall see) depends on interactions between individual organisms, populations, and environments. The importance of evolvability as an interaction effect is buried in this set of conditions. As with many discussions of evolvability (recently, Brookfield 2009; Pigliucci 2008), these conditions focus largely on the characteristics of individual developmental systems. Let's begin with the missing ingredient, phenotype plasticity.

Plasticity, Evolvability, and Modularity

There is a crucial connection between developmental and evolutionary plasticity, as Mary-Jane West-Eberhard and Marc Kirschner in collaboration with John Gerhart; have shown. They argue that the developmental plasticity of *organisms* explains the evolutionary plasticity of *lineages* (Gerhart and Kirschner 1997; Kirschner and Gerhart 1998, 2005; West-Eberhard 2003); for an early and important work on this connection, see Schlichting and Pigliucci (1998).

Kirschner and Gerhart begin by pointing out that adaptive phenotypic plasticity is essential for complex organisms. Complexity depends on buffering against contingency. Developing embryos will be exposed to differing environmental fluxes; they will be supplied with differing nutrient packages; and the gene networks that control development act in varying genetic environments. Such variations in internal and external environment affect developmental trajectories.[1] Subsystems—organs, tissues, structural supports—develop and function in somewhat different internal environments. Unlike factory-built machines, organisms are not made of fully standardized parts. The exact shape, location, and structure of components cannot be predicted in advance. Yet organ systems must be appropriately connected to one another, and typically the systems of signaling, coordination, and linkage in development cope with such variation. The precise layout of the blood circulation system cannot be prespecified, for the network depends on bone and muscle growth. Cells must never be more than a few cell diameters away from a capillary. Capillaries are provided by oversupply coupled with selective attrition. The oversupply is pruned less vigorously where muscle development enhances the demand for oxygen flow (see Turner 2006). This mechanism of oversupply coupled with selective attrition seems to be the way one system adjusts to contingencies elsewhere (it is an example of Calcott's general engineering principles; which play a role in many of the transitions; see Calcott, this volume).

The general idea is familiar from discussions of robustness and homeostasis. Homeostasis depends on flexibility. If body temperature is homeostatically stabilized, the mechanisms that maintain invariance, and that return the system to a stabilized point after disturbance, must themselves change in response to variation and disturbance (Godfrey-Smith 1996). West-Eberhard shows that mechanisms of mutual adjustment are surprisingly powerful. Her flagship example is a two-legged goat, which was born with only its hind legs functional, but through a combination of behavioral, physiological, and morphological adjustments acquired the capacity to move like a goatish kangaroo. The goat died (accidentally) without issue, and is such an extreme example it might not be directly relevant to evolutionary issues. But successful adjustments to less fundamental perturbations have occurred. For example, in human populations there are many pathologically developed hearts, with arteries, veins, and valves in nonstandard places. These developmental pathologies are not

desirable, but they are not instantly fatal. Human developmental mechanisms can successfully connect these aberrant organs to our circulatory and respiratory system.

These mechanisms of adjustment to the internal environment make possible phenotypic adjustment to genetically driven novelty elsewhere in the phenotype. Adjustments will not require correlated genetic change. If sexual selection on a horned beetle increases the horn's length and mass, there is no need for further genetic changes to ensure that the horn is adequately supported structurally and that the beetle has the power to wield it. Indeed, in many horned beetle species, the development of the horn is facultative, depending on the initial food supply to the larvae, and hence the rest of the developmental system must be able to adjust to different horn mass and shape (Moczek 2005). Kirschner and Gerhart point out that such mechanisms of adaptive plasticity are central to the evolvability of genome structure. The mechanism in mitosis that ensures that each daughter cell receives the right chromosome complement ("spindle formation") is adaptively plastic. Before mitosis, genetic material is scattered through the nucleus, so no mechanism could be preprogrammed with information about the location of the chromosomes in the dividing mother cell. The microtubes that usher to them to the daughter cells thus explore from the centriole. If they connect with a chromosome, they stabilize; if not, they are reabsorbed, with new microtubes forming in their stead (Kirschner and Gerhart 1998). Without such a system, mutations that increase chromosome number would be fatal.

The general point is that unless there were a mechanism that mediated the adjustment of one structure in response to a change in another, a coordination problem would severely constrain adaptive change, for change in one structure will typically necessitate change in others. Phenotypic accommodation reduces the problem of correlated change. A genetically caused modification in one system need not wait for a genetically caused change in associated systems, even when both organ systems must change for either change to be adaptive. Mechanisms of phenotypic plasticity support phenotypic adjustment to genetically caused changes in an organism. These mechanisms act as *change amplifiers*. Genetic changes that directly affect only one component of an organism can result in a suite of adaptively correlated changes. Thus, a small genetic change can map onto a large phenotypic change through these knock-on effects. These considerations point to a critical connection between developmental and evolutionary plasticity.

The importance of plasticity has consequences for the role of modularity in evolution. It is now received wisdom that modularity increases evolutionary plasticity (for recent restatements of this view, see Pigliucci 2008; Wagner, Pavlicev, et al. 2007). If a trait is entrenched, that is, if in development it is connected to many other traits, the trait is evolutionarily inflexible. This is because a gene change that alters the target trait advantageously is likely to have disadvantageous consequences elsewhere in the phenotype (Wimsatt 2007, ch. 7). Only if the development of a trait is relatively modular can it respond freely to selection. However, Wimsatt's generative entrenchment model assumes

that *developmental interconnection* is undirected in just the way *mutation* is. If X and Y are two traits interconnected in development, and if X changes in ways relevant to Y's development, those effects on Y, like those of mutation, are undirected with respect to fitness. But given the existence of mechanisms of adaptive plasticity, this picture of modularity is too simple. Organs that develop together can be connected so that each responds appropriately to changes in the other.

Evolvability thus does not depend on a developmental quarantine, a system in which each organ develops independently of the developmental trajectory of other systems. Such a developmental organization would be immensely vulnerable to developmental mismatch driven by environmental and genetic noise, and would lead straight to the evolutionary coordination problem discussed earlier. We need to fine-tune our concept of modularity, for some developmental mechanisms that link the development of two systems enhance evolutionary plasticity rather than limit it. It follows that the idea of developmental autonomy somewhat mischaracterizes modularity. We should think of quasi-independence as some combination of autonomy with adaptive mutual adjustment. There are limits to mutual adjustment, but within those limits, in a noisy world, codeveloping systems of necessity have evolved to respond appropriately to one another's changes.

There is an analogous oversimplification in the literature on developmental robustness. Critical traits must develop robustly. It is very important that their development not be derailed by genetic or developmental noise. Hence, we should expect crucial aspects of phenotype to be canalized. Within population genetics, this idea of robustness has been captured (in part) by the notion that the development of a trait is less subject to genetic noise if its development depends on fewer genes (Wagner, Pavlicev, et al. 2007). Lenski, Barrick, and Ofria point out that robustness so conceived can reduce evolvability by shrinking a trait's mutational target. There are fewer genetic levers whose position affects that trait in question (Lenski, Barrick, et al. 2006). That is why it is less vulnerable to mutational noise, but also less available for adaptive change. Their point is well taken. But robustness can also be achieved not by reducing the mutational target but through genetic redundancy, and hence through many/one mappings of genotype to phenotype; mappings that can preserve significant amounts of potentially important genetic variation in a population (Rutherford 2000; Wagner 2007). Moreover, robustness is not invariance. Crucial behavioral, morphological, and metabolic traits must, indeed, develop robustly, but the precise pace and form of development must be sensitive to the rest of the phenotype. As noted earlier, homeostasis in one character requires plasticity in the systems that support the stabilized trait (Godfrey-Smith 1996, 3.4).

Evolvability: A Brief and Speculative History

In my view, the consensus characterization of rich Darwinian environments left out the crucial role of plasticity and its connection with modularity, and underplayed the interac-

tive basis of evolvability. Perhaps as important, this characterization poses the problem statically: It characterizes the destination rather than the route. The destination is important; we want to understand the rich Darwinian environments that make possible the evolution of complex macrobes, eusocial insect colonies, and the like. But the synthesis leaves out the evolution of evolvability. It is also important to explain the incremental construction of the enabling conditions of these necessarily late-evolving novelties. As Godfrey-Smith notes in this volume, populations do not automatically have a stable location in the space of Darwinian environments.

So let's suppose that a suitably modified set of conditions captures the evolutionary regimes in which novelty, disparity, and new levels of organization are likely to appear. How do such rich regimes come into existence and how are they maintained? I shall sketch a three-phase model: life up to the evolution of fully equipped prokaryote cells; a phase of microbial evolution; and a third phase that sees the evolution of complex development. Possibility-expanding novelties, including those that permit the evolution of new levels of biological individuality, have evolved throughout the history of life. But the conditions that permit novelty have changed over time. What follows is something between a how-possibly explanation of the evolution of evolvability and a first-approximation how-actually explanation. The main function of this section is to illustrate the idea that evolvability has an informative history. But this illustration is made more plausible by being constructed from genuine, though clearly very speculative, hypotheses about the course of that history.

I concentrate most on the first phase (as this material is the least familiar and most problematic). But I begin with the overall idea of a rough, three-phase model of the evolution of evolvability. The first of the phases is evolution before the cell. In understanding this phase of history, the key problem is to identify the conditions that made the evolution of the cell possible, to understand the initial bootstrapping of evolvability. The second phase is that of unicell evolution. The cell exists, together with the mechanisms of gene regulation, translation, and transcription. Horizontal gene transfer is important in prokaryote evolution. But at least on microevolutionary time scales, inheritance is predominantly vertical. Most of the time, all the genes in a cell succeed or fail together. But the causal chain between gene expression and cell phenotype is short and direct. Indeed, the prevalence of horizontal transfer depends on the fact that as a rule of thumb, if a gene does move between lineages, it will have the same phenotypic effect in its new home as it had in its old one. Prokaryote evolution continued apace after the emergence of complex macrobes. But the third phase is initiated by the evolution of a developmental cycle, and with it the invention of long-causal chain development.

One might therefore see rich Darwinian environments as established by two thresholds—those of Darwin and Maynard Smith. A "Darwinian threshold" marks the beginnings of an evolutionary regime dominated by high fidelity, largely vertical inheritance, and hence the evolutionary exploration of cellular lineages (Fox Keller 2009).

Maynard Smith's threshold is reached when development becomes complex, and hence when gene effects are, in his sense, arbitrary. Genes connect to phenotypes via complex causal chains mediated by much other biological equipment. The complexity of the chains, and their dependence on other biological systems, makes available multiple points of intervention. Thus, the one protein product can have different phenotypic effects in different tissues and organisms. The route from gene to phenotype becomes indirect, complex, arbitrary; molded by selection rather than the biochemistry of translation and transcription. As this happens, Maynard Smith argues, the genome comes to semantically specify the phenotype; the relationship becomes genuinely informational (Maynard Smith 2000). I shall briefly elaborate on this contrast between prokaryote and multicellular evolvability, before returning to the deeply obscure problem of precellular evolution.

Prokaryote Evolvability

Microbial evolution, and especially prokaryote evolution, is extraordinarily evolutionarily fecund. That fecundity is illustrated by the diversity of metabolic styles and the profusion of Archaean extremophiles. Prokaryotes explore nearby regions of phenotype space very effectively, through a combination of rapid generation time and large population sizes. If superior versions—potential improvements—are close in gene space to current genomes, their population size creates a large target. They are also fecund because one lineage will sometimes import innovations forged in another by horizontal gene transfer. As a consequence, the genetic resources available in the local environment are relevant to the evolutionary potential of the lineages in that environment. Indeed, these genetic resources are so important that in some views there is a prokaryote "mobilome" of genetic elements that move readily between lineages. This mobilome consists of bacteriophages; plasmids, transposable elements, and genes that are attached to them. Indeed, it is arguable that horizontal gene transfer is so important to prokaryote evolution that it undermines the tree of life model (Koonin and Wolf 2008).

There is no doubt that horizontal gene transfer has been important in the deep history of both eubacteria and Archaea. There has been horizontal transfer within bacterial lineages, and across domains as Archaea borrowed crucial metabolic innovations from bacteria. However, the extent and interpretation of gene migration across cellular lineages remains controversial (O'Malley and Dupre 2007). In one view, successful gene migration is a rare event. It has happened many times, and many genes in the genome of bacterial cell lineages have their original ancestor in other lineages. But that is true only because we are looking at ancient lineages—at the summed results of rare events over 3.5 billion years. Almost all the genes in any given prokaryote clone-line have been replicating together for millions of generations; the expected fitness of one is the expected fitness of all (Lerat, Daubin et al. 2005). In another view, lateral gene transfer is pervasive on evolutionarily significant time scales, but it is very rare for core "informational genes"—those responsible for the machinery of replication, transcription, and translation (Brown 2003; Woese 2008). In the

most extreme view, it is such a pervasive feature of prokaryote evolution as to undermine the very idea that prokaryote evolution is best thought of as a branching tree (Bapteste and Boucher 2008; Doolittle and Bapteste 2007; Martin and Embley 2004).

While these issues are of core importance, they are primarily about phylogeny rather than selective regime. It is one thing to claim that transfer is common enough over evolutionary history to undermine the idea that we best represent microbial evolution as a branching tree. It is another to say that it is so common that bacterial genomes in a cellular lineage are loose alliances of independent replicator lineages rather than a single linked genome with a shared fate. No one says that about bacteria or Archaea. As we shall see, that idea is very much on the table when the evolution of the cell itself is in question.

Maynard Smith's Transition

Prokaryotes are extraordinarily complex biochemical systems; that is why it is so hard to understand how they evolved. Even so, to the extent that we can talk of prokaryote developmental systems at all, they are vastly simpler than those of eukaryote multicelled lineages. (Eukaryote unicells are an interesting and difficult intermediate case.) Their genomes are stripped down (arguably, optimized for rapid replication and fast reproduction). Their genomes have few introns, and genes are often continuous stretches of DNA adjacent to their regulatory regions. So prokaryote genome organization is smaller and simpler than that of eukaryotes, as is gene control (Koonin and Wolf 2008; Lane 2005). So, whereas prokaryote gene effects are clearly complex, the genotype/phenotype relationship is very different in multicelled organisms. Genome size is typically larger. Notoriously, genome architecture is messily complex. It is more fragmented, with more complex control, and has a transcription-translation system that is faced with a high intron load and the need to export instructions across a nucleus boundary. Most critically, though, in multicell systems, development routinely builds from scratch new structures in each generation. Tissues, organ systems, support structures, and circulatory systems all have to be built anew. That is not true when a prokaryote splits into two daughter cells. Its cell walls and many intercellular structures are continuously present and available through the process of gene replication and cell fission.

In contrast to prokaryotes (and perhaps unicellular eukaryotes), therefore, multicell systems have a developmental cycle. Dawkins argues that the developmental cycle is essential to a certain kind of novelty, as an early acting mutation can rejig the structure of the whole organism, as it is rebuilt in the next generation (Dawkins 1982). I have argued, with Brett Calcott, that complex metazoan developmental systems are made possible not just by the evolution of higher-fidelity DNA replication, as Mark Ridley suggests (Ridley 2000), but also by the evolution of the control of the environment in which genes are used (Calcott and Sterelny submitted; Sterleny 2009). Eggs and seeds are highly structured and stabilized environments. In such environments, and only in such environments, development can have a predictable outcome despite depending on long, complex causal chains. After the

evolution of a developmental cycle and the evolution of systems that structure and stabilize the environments in which genes are expressed, the relationship between gene and phenotype is very indirect. Thus, Maynard Smith argues that the intrinsic chemical properties of the gene do not fix, or even much constrain, its ultimate effect on the phenotype of its bearer; the gene-phenotype relationship is arbitrary (Maynard Smith 2000). Gene reading systems expand the space of biological possibility by expanding the range of phenotypes that can be built from a set of genes.

A Darwinian Transition?

Prokaryote evolution is impressively fluid. But it presupposes the cell, and as Carl Woese rightly points out, the cell is itself the Mother of All Key Innovations. Was it put together by gradual change in an alliance of high-fidelity, coadapted replicators traveling together and sharing a common fate? He thinks not (see Goldenfeld and Woese 2007; Vetsigian, Woese, et al. 2006; Woese 2002, 2004). Woese thinks that *after* the evolution of the cell, evolution produces a treelike pattern. But that is not true of the evolution of the cell itself. As Woese pictures it, the evolution of the cell is, in effect, an egalitarian major transition (Queller 2000). Prior to its final invention, protocell-like structures were more like ecological communities than organisms, with genelike elements shifting regularly between quasi-cells.

In these quasi-cells, genelike elements are too mobile to have a common fate; they are not fated to succeed or fail together. For much of their evolution, these protocells were communities with somewhat porous boundaries. They were less integrated than the prokaryotes they evolved into, and so horizontal gene transfer was pervasive rather than occasional. The genes in the community only came to share a common fate and become a genome in the contemporary sense *after* the evolution of the full suite of prokaryote cell structures. As integration increased, symbiotic associations became more integrated and obligatory, and the balance between vertical and horizontal flow changed. As Woese sees it, then and only then can we think of selection on the cells (rather than on the previously mobile constituents) as fully Darwinian. For only then do cells reproduce, fully and paradigmatically. Thus, he thinks of this as the time when life—life organized into organisms, of which the cell is the minimal kind—crossed a "Darwinian threshold." LUCA—the Last Universal Common Ancestor—is not the first cell; it is the establishment of a cell whose genes share a common fate.

If anything like this picture is right, we need some explanation of why protogene interactions were, at least in some circumstances, cooperative enough for temporary and conditional alliances to become unconditional and permanent ones, as genuine cells evolved. Models of network evolution, conjoined with a recent hypothesis about the role of hydrothermal vents in early evolution, suggest a possible way forward here. There might be more routes to cooperation than the consensus conditions suggest, for it has recently been suggested that LUCA predates the full evolution of autonomous cell structure. The idea is that

the RNA-world to DNA-world transition and the evolution of the transcription-translation system *predates* the evolution of cell walls. This is because the cell wall chemistry of the Archaea is significantly different from that of the eubacteria, suggesting that their cell walls evolved independently, after LUCA. These early quasi-cells were bounded not by membranes but by inorganic structures, which were formed in an expanding foam of iron pyrites created as warm undersea vents (not black smokers) interacted with the ancient seas (Martin, Baross, et al. 2008). Geologically similar though chemically different structures exist today (today's seas lack iron). These structures are long lasting (for perhaps over 30,000 years), but they are dynamic, and their inorganic compartments have cell-like diameters. The compartments are a barrier to free movement of materials; they ensure that interaction is predominantly local. The output of some primitive form of protein synthesis would not simply diffuse away into the structure as a whole. But the components are not fully sealed: Materials move through these towers, and so genelike elements (and ensembles of such elements) from persisting cellular communities would be able to colonize vacant compartments formed by new growth and emptied ones created by internecine interactions within compartments (Koonin and Martin 2005).

If this idea were roughly right, evolution in these towers of physical compartments would be a natural counterpart of evolution in networks that Ben Kerr and others have explored (Kerr, Neuhauser, et al. 2006). Kerr shows that in network evolution, locally constrained migration selects for cooperative behavior, and movement through the towers was indeed likely to have been locally constrained. If the hydrothermal vent hypothesis is right, then cell evolution depended on physical scaffolding in the local environment, and was not completed until after the Archaean-eubacteria split. If, in contrast, Woese's model is right, the essential components of the cell were in place before the bacteria-Archaea split. On both views, protocells were somewhat more like ecological communities than organisms.

Hence, the idea of a transition to a Darwinian world. Woese and his allies think such protocell evolution precedes a Darwinian transition, for the phenotype of a pretransition protocell depends more on its neighbors and those neighbors' immediate ancestors than on the protocell's own distant ancestors (Vetsigian, Woese, et al. 2006). Their idea can be captured most clearly through a contrast. Despite horizontal gene transfer, a contemporary bacterium closely resembles its ancestor of a thousand generations back, because transmission is mostly vertical. Perhaps a plasmid for antibiotic resistance will have been lost or picked up. But most of the genes remain the same. At least in microevolutionary time frames, a dominating vertical historical signal explains phenotype character. As Woese sees it, the establishment of this dominating signal was the Darwinian transition of 3.5 billion years ago (Woese 2002, 2008). Before that, protocell phenotypes were not nearly so history dependent. Rather, they were as dependent on the supply of transferable elements in the current and recent past environments. Genomes (or what were to become genomes) were fluid on ecological, not just deep evolutionary, time frames.[2]

Indeed, it was only through this fluidity that the cell could evolve, and come to have a genome with a largely shared fate. The cell is an immensely complex system to establish from scratch, with its membranes, microtubular machinery, the mechanisms that control gene replication and use, cell wall structures, and the like. No single alliance of replicators in an early protocell could have generated all the necessary innovations. That is especially plausible since, throughout much of this process, gene replication would have been less accurate and the gene-protein mappings less precise and reliable. Some crucial cellular technology must have been imported, Woese suggests, by horizontal gene transfer. The search space was explored collectively, and from different starting points. The early suite of protocells would have differed one from another. Some would have had a better cell wall starting point, others better initial gene replication machinery. The cell was a product of collective search. A world with independent search, especially limited by the inefficiencies of early replication and translation systems, with strict vertical inheritance would not have found the cell.

Woese's model may well be wrong-headed. But if it is even close to being right, the evolutionary potential of a lineage does not depend solely on the intrinsic properties of the organisms that compose it. The evolutionary potential of protocell lineages depends in part on innovations available for import from other lineages in the local environment. The same is true if LUCA lacked cell walls, as the vent hypothesis supposes. If there really was a Darwinian threshold in early cell evolution, importing innovations from neighbors has changed from being a routine microevolutionary event to a more unusual aspect of biological change. But it has certainly not ceased. Horizontal gene transfer continues to be an important mechanism in prokaryote evolution. Lateral transfer as a mechanism of generating genetic diversity is less important in eukaryotes. But eukaryotes important innovations from other lineages via the incorporation of symbiotic passengers. While the incorporation of symbiotic associates is rare on microevolutionary time frames, it is a major source of innovation on macroevolutionary time scales (Kutschera and Niklas 2005; Moran 2007; Sterelny 2004). The resources available in the local environment continue to be an important determinant of evolutionary potential. This theme becomes the focus of the next section. The take-home message of this section is that while evolvability is not a trait of individual organisms (still less an adaptation of individual organisms) it does have an incremental history. The preconditions of novelty have been built gradually, over deep time, and unevenly in different lineages.

Lineages, Populations, and Environments

Evolvability is a property of lineages. But it is often discussed as if the evolvability of a lineage is a simple product of the developmental mechanisms of the organisms that form the lineage. While this is most obvious in the papers that float adaptationist models of evolvability (Earl and Deem 2004; Radman, Matic, et al. 1999), it is a pervasive trend in

the literature. That is not surprising. One aspect of evolvability is just the capacity of a lineage to generate novel traits, especially complex novel traits. But, of course, the developmental mechanism on which such novelties depend—the germline/soma split; the control of differentiation; the evolution of sex; DNA repair mechanisms—are themselves instances of novelty. So one standard way of thinking about evolvability is to see it as the project of explaining the evolution and unequal distribution through lineages of such crucial developmental innovations (Hendrikse, Parsons, et al. 2007; Pigliucci 2008). Explaining the evolution of evolvability turns into the project of explaining the origin and distribution of special developmental mechanisms, themselves novelties on which other novelties depend.

This project is important and challenging. The challenge arises because it is hard to combine the *distributional* and *incremental* requirements in a good explanation of developmental novelty. Standard evolutionary explanations explain the distribution of characters in populations; typically, changes in population profiles, as fitter variants become more common at the expense of less fit variants. Distributional factors are relevant to the appearance of novelty, for a given novelty will be more likely to appear given some trait distributions than others. While distributional explanations contribute to a theory of origins, they need to be supplemented by *lineage explanations* (Calcott 2008; this volume). A lineage explanation shows how the appearance of type T makes the appearance of T* more probable, by displaying a natural variational path from T to T*. A lineage explanation constructs a sequence of prototypical organisms, each of which has a modified, modifiable, and functioning variant of some focal trait.

A famous example of a lineage explanation is Nilsson and Pelger's celebrated model of the evolution of the eye (Nilsson and Pelger 1994). The model explains the origin of a morphological novelty by constructing a route from a simple light-sensitive patch to a focused lens eye. This route meets two conditions. First, adjacent links in the lineage are structurally similar, similar enough to shift from one to the next in one step. Second, the availability of each link as input to the next is compatible with the distributional processes acting on the population. Let E, E*, and E** represent three successive eyes at a stage in which the focus of the lens is gradually improving. Obviously, E* will not be a plausible intermediate between E and E** if an individual with an E*-class eye would have poor prospects in a population of agents equipped with E-class eyes. The Nilsson-Pledger model meets this condition because each step improves the resolution of the eye.

Individual variations that increase evolvability seem likely to be filtered out rather than preserved, and hence in the evolvability literature, satisfying the distribution criterion has been problematic. This issue was visible at the end of the second section of this chapter. Selection for robustness—canalizing currently adaptive traits—seemed to reduce the likelihood of further change. While selection could favor increases in replication fidelity, for mutations are typically neutral or worse, it is hard to see how changes that increased variation could survive selection. Most notoriously of all, if the evolution of new levels of

individuality requires agents to sacrifice their own capacity to reproduce, it has been hard to formulate plausible pictures of regimes that would select for such sacrifice.

Although I agree that explaining the evolution of these individual developmental novelties is central to explaining evolutionary plasticity, it is not sufficient. The supply of variation depends as well on properties of populations and environments. So I will conclude by beginning the project of characterizing the *collective profile* of a highly evolvable population, and briefly reviewing reasons for thinking that such collective properties are important. The take-home message is simple: Evolvability depends on interactions between individual developmental mechanisms, populations, and environments. We saw that particularly vividly in the last section, in discussing a couple of conjectures about cell evolution. As we shall see, there are less exotic cases that make the same point, albeit less dramatically.

Environmental Homogeneity and Heterogeneity

Variation is a property of populations, not individuals. It arises out of interactions between individual developmental mechanisms, population structure, and the environment. For example, variation in the environment bears on variation in the population. It is no surprise that heterogeneous environments can help maintain heterogeneity in a population, by selecting for different phenotypes in different patches of the population's range. Environmental complexity can also preserve variation, by imposing competing selective demands on a population, and hence allowing the evolution of a range of distinct, but equi-fit phenotypes. Chris Marshall and Karl Niklas argue that complex selective regimes in which agents must balance many competing demands keep heterogeneity in the population, for as the number of trade-offs goes up, so too does the number of equally fit, different phenotypes (Marshall 2006; Niklas 2002).

More surprising is the fact that homogeneous environments sometimes purge variation less effectively than one might expect. Even when a homogeneous environment imposes a uniform phenotype, genotype variation can survive. Importantly, variation that is cryptic in one environment can be revealed in others, thus powering evolutionary response to environmental change, for variation in the environment determines the fraction of the reaction norm that is expressed and thus exposed to selection. More uniform environments allow cryptic genetic variation to survive unexpressed. Suzanna Rutherford has developed this idea recently. Populations have unexpressed genetic variability; for example, in natural *Drosophila melanogaster* populations, she argues, there are on average hundreds of thousands of base pair differences between the average haploid genotype. Yet these populations are phenotypically uniform. This variation can be unmasked by environmental or genetic change. It is *unexpressed* difference, not *inexpressible* difference. For example, a mutant form of the heat-shock protein *Hsp90* in *Drosophila* unmasks mutations in other genes that would otherwise be silent (Rutherford 2000). Andreas Wagner has developed a similar idea (Wagner 2005). So factors that tend to make the experienced environment of organisms

more uniform can mask genetic variation, allowing it to accumulate in populations. Many-one relations between genotype and phenotype enable populations to store variation.

Population Structure

Woese's model of the collective evolution of the cell is a spectacular example of a population-centered approach to evolvability. According to his model, the evolution of the cell depended on there being a population of disparate protocells that could import innovations from one another. Woese's model is very conjectural. But horizontal gene transfer continues to be important in prokaryote evolution, with rich horizontal transfer of ready-made genetic material. Plasmids, phage DNA, and transposons are different-sized packets of genetic material that can move horizontally. Given the ubiquity of horizontal gene transfer, the richness of the local genetic resources is obviously important to the evolutionary possibilities of prokaryote lineages (Carroll 2002). Plant lineages, too, often hybridize, importing genetic resources from outside. More generally, sex makes the local supply of genetic resources relevant to macrobes, too, though "local" is defined less inclusively. Sex also makes metapopulation structure important. Microevolutionary change takes place within local populations, and if these are isolated from one another, there may well be potentially important gene combinations that are unavailable, because relevant variants have arisen in different populations. The evolutionary response of a lineage will depend in part on the variation spread across the populations from which it is built and the kinds of flow between them.

Population structure is also crucial to multilevel selection. The selection of cooperation in mixed populations depends on cooperating agents preferentially associating with other cooperating agents. It depends, in other words, on an appropriately viscous population structure. It follows that the mechanisms that result in biased associations are central to the evolvability of new levels of individuality, since multilevel selection probably plays a crucial role in the evolution of new levels of biological organization (Kerr and Godfrey-Smith 2002; Michod 1999, 2007; Okasha 2006).

Most novelties, including most developmental novelties, are constructed piecemeal, and hence their origin involves an interaction between collective characteristics—population structure, variation storage, the protection of local adaptation—and individual developmental mechanisms. The synthesis rightly insists that significant evolutionary change typically depends on the accumulation of small changes . This accumulation, in turn, depends on the existence of ratchets that protect each small advance. There is a ratchet that connects individuals in a lineage: high-fidelity replication preserves rare favorable variations. But while organism-level mechanisms that project change down the generations are necessary for evolvability, they are not sufficient. Potentially favorable change must be *preserved* by individual-level inheritance processes. But these must then be *amplified* in the population; their frequency must increase to levels that protect them from extinction by local ill chance. It is very easy for a rare variant to drift to extinction. The favorable change must also be

protected by population-level processes from being swamped by immigration. Local adaptations are easily lost by homogenizing effects of migration, so if a local variant is both isolated and amplified to high local frequency, a further mutational change and a further iteration is more likely. Selection is not just a consumer of variation: It creates variation. When a phenotypic change from P to P* depends on a sequence of genetic changes from $G_0 \Rightarrow G_1 \Rightarrow G_2$, selection can make G_2 vastly more probable by making G_1 common. A lineage explanation explains why the pathway from P to P* is available at all, by analyzing the developmental system that leads to P, and showing how that system can be modified in a small way without disaster. The distributional explanations explain why that system is repeatedly available for modification and why that modification, in turn, is made available for further change. This interplay between distribution and the generation of variation underscores the fact that evolvability is not a characteristic of individuals. It emerges from an interplay between (i) individuals and their developmental systems, (ii) the populations and lineages of which they are a part, and (iii) the environments within which they are embedded. To understand it, we need to combine distributional with lineage explanations.

In summary, this chapter has three take-home messages. First, the project of identifying rich Darwinian environments is important and difficult, for those environments make possible novelties that expand evolutionary possibility. I have suggested ways in which standard pictures of such environments need to be fine tuned. Second, that project should be historicized. It needs to be linked to a model of how such environments evolve, and when and why they are stable once evolved. The major transitions, to the extent that they are transitions in heritability, are part of this history. Third, it is important to explicitly identify the interactive character of these conditions. Individual properties of developmental mechanisms are important to evolvability, but they are not all that is important.

Notes

1. For an amusing example, see Ed Regis's description of the squat, Ramboesque chickens raised in a centrifuge that simulated life in a Jovian gravitational field (Regis 1990).

2. Thanks to Peter Godfrey-Smith for forcing me to think more clearly about this idea that a Darwinian threshold was crossed in pre-prokaryote evolution.

References

Bapteste, E., and Y. Boucher (2008). Lateral gene transfer challenges principles of microbial systematics. *Trends in Microbiology* 16(5): 200–207.

Bonner, J. T. (1998). Origins of multicellularity. *Integrative Biology* 1(1): 27–36.

Brookfield, J. (2009). Evolution and evolvability: Celebrating Darwin 200. *Biology Letters* 5(1): 44–46.

Brown, J. R. (2003). Ancient horizontal gene transfer. *Nature Reviews Genetics* 4: 121–132.

Brusca, G. J., and R. C. Brusca. (1990). *Invertebrates*. Sunderland, MA: Sinauer.

Calcott, B. (2008). Lineage explanations: Explaining how biological mechanisms change. *British Journal for the Philosophy of Science* 60: 51–78.

Calcott, B., and K. Sterelny. (submitted). The Cambrian Explosion and the evolution of life. *Biological Theory*.

Carroll, R. L. (2002). Evolution of the capacity to evolve. *Journal of Evolutionary Biology* 15: 911–921.

Dawkins, R. (1982). *The Extended Phenotype*. Oxford: Oxford University Press.

Doolittle, W. F., and E. Bapteste. (2007). Pattern pluralism and the tree of life hypothesis. *Proceedings of the National Academy of Science* 104(7): 2043–2049.

Earl, D. J., and M. W. Deem. (2004). Evolvability is a selectable trait. *Proceedings of the National Academy of Science* 101(32): 11531–11536.

Fox Keller, E. (2009). Self organisation, self-assembly and the origin of life. In A. Barberousse, M. Morange, and T. Pradeu (eds.), *Mapping the Future of Biology* (pp. 131–140). New York: Springer.

Gerhart, J. C., and M. W. Kirschner. (1997). *Cells, Embryos and Evolution*. Malden, MA: Blackwell Science.

Godfrey-Smith, P. (1996). *Complexity and the Function of Mind in Nature*. Cambridge: Cambridge University Press.

Godfrey-Smith, P. (2009). *Darwinian Populations and Natural Selection*. Oxford: Oxford University Press.

Goldenfeld, N., and C. Woese. (2007). Biology's next revolution. *Nature* 445(25 January): 369.

Hendrikse, J. L., T. E. Parsons, et al. (2007). Evolvability as the proper focus of evolutionary developmental biology. *Evolution & Development* 9(4): 393–401.

Kerr, B., and P. Godfrey-Smith. (2002). Individualist and multi-level perspectives on selection in structured populations. *Biology and Philosophy* 17(4): 459–476.

Kerr, B., C. Neuhauser, et al. (2006). Local migration promotes competitive restraint in a host-pathogen "tragedy of the commons." *Nature* 442(7098): 75–78.

Kirschner, M., and J. Gerhart. (1998). Evolvability. *Proceedings of the National Academy of Science, USA* 95: 8420–8427.

Kirschner, M. W., and J. C. Gerhart. (2005). *The Plausibility of Life*. New Haven, CT: Yale University Press.

Koonin, E. V., and W. Martin. (2005). On the origin of genomes and cells within inorganic compartments. *Trends in Genetics* 21(12): 647–654.

Koonin, E. V., and Y. Wolf. (2008). Genomics of bacteria and archaea: The emerging dynamic view of the prokaryotic world. *Nucleic Acids Research* 36(21): 6688–6719.

Kutschera, U., and K. Niklas. (2005). Endosymbiosis, cell evolution and speciation. *Theory in Biosciences* 124: 1–24.

Lane, N. (2005). *Power, Sex, Suicide: Mitochondria and the Meaning of Life*. Oxford: Oxford University Press.

Lenski, R., J. Barrick, et al. (2006). Balancing robustness and evolvability. *PLoS Biology* 4(12): 2190–2192.

Lerat, E., V. Daubin, et al. (2005). Evolutionary origins of genomic repertoires in bacteria. *PLoS Biology* 3(5): 807–814.

Lewontin, R. C. (1970). The units of selection. *Annual Review of Ecology and Systematics* 1: 1–14.

Marshall, C. (2006). Explaining the Cambrian "Explosion" of animals. *Annual Review of Earth and Planetary Sciences* 34: 355–384.

Martin, W., J. Baross, et al. (2008). Hydrothermal vents and the origin of life. *Nature Reviews Microbiology*, advance online publication, 29(September 2008) doi: 10.1038.

Martin, W., and T. M. Embley. (2004). Early evolution comes full circle. *Nature* 104(7): 134–137.

Maynard Smith, J. (2000). The concept of information in biology. *Philosophy of Science* 67(2): 177–194.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution*. New York: W. H. Freeman.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality*. Princeton, NJ: Princeton University Press.

Michod, R. E. (2007). Evolution of individuality during the transition from unicellular to multicellular life. *Proceedings of the National Academy of Science* 104: 8613–8618.

Moczek, A. (2005). The evolution and development of novel traits, or how beetles got their horns. *BioScience* 55(11): 937–951.

Moran, N. (2007). Symbiosis as an adaptive process and source of phenotypic complexity. *Proceedings of the National Academy of Science* 104(May 15): 8627–8633.

Niklas, K. (2002). Computer models of early land plant evolution. *Annual Review of Earth and Planetary Sciences* 32: 47–66.

Nilsson, D.-E., and S. Pelger. (1994). A pessimistic estimate of the time required for an eye to evolve. *Proceedings of the Royal Society of London, B* 256: 53–58.

Odling-Smee, J., K. Laland, et al. (2003). *Niche Construction: The Neglected Process in Evolution.* Princeton, NJ: Princeton University Press.

Okasha, S. (2006). *Evolution and the Units of Selection*. Oxford: Oxford University Press.

O'Malley, M., and J. Dupre. (2007). Size doesn't matter: Towards a more inclusive philosophy of biology. *Biology and Philosophy* 22(2): 155–191.

Pigliucci, M. (2008). Is evolvability evolvable? *Nature Reviews Genetics* 9(January): 75–82.

Queller, D. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions of the Royal Society B* 355: 1647–1656.

Radman, M., I. Matic, et al. (1999). Evolution of evolvability. *Annals of New York Academy of Sciences* 870: 146–155.

Regis, E. (1990). *Great Mambo Chicken and the Transhuman Condition: Science Slightly Over the Edge*. New York: Addison-Wesley.

Ridley, M. (2000). *Mendel's Demon: Gene Justice and the Complexity of Life*. London: Weidenfeld and Nicholson.

Rutherford, S. (2000). From genotype to phenotype: Buffering mechanisms and the storage of genetic information. *Bioessays* 22(12): 1095–1105.

Schlichting, C., and M. Pigliucci. (1998). *Phenotypic Evolution: A Reaction Norm Perspective*. Sunderland, MA: Sinauer.

Sterelny, K. (1999). Bacteria at the high table. *Biology and Philosophy* 14(3): 459–470.

Sterelny, K. (2001). Niche construction, developmental systems and the extended replicator. In R. Gray, S. Oyama, and P. E. Griffiths, *Cycles of Contingency* (pp. 333–350). Cambridge, MA: MIT Press.

Sterelny, K. (2004). Symbiosis, evolvability and modularity. In G. Schlosser and G. Wagner (eds.), *Modularity in Development and Evolution* (pp. 490–516). Chicago: University of Chicago Press.

Sterelny, K. (2009). Novelty, plasticity and niche construction: The influence of phenotypic variation on evolution. In A. Barberousse, M. Morange, and T. Pradeu (eds.), *Mapping the Future of Biology. Evolving Concepts and Theories*. Dordrecht, Germany: Springer.

Turner, J. S. (2006). *The Tinkerer's Accomplice. How Design Emerges from Life Itself*. Cambridge, MA: Harvard University Press.

Vetsigian, K., C. Woese, et al. (2006). Collective evolution and the genetic code. *Proceedings of the National Academy of Science* 103(28): 10696–10701.

Wagner, A. (2005). Robustness, evolvability, and neutrality. *FEBS Letters* 579(8): 1772–1778.

Wagner, A. (2007). Robustness and evolvability: A paradox resolved. *Proceedings of the Royal Society B* 275(1630): 91–100.

Wagner, G. P., M. Pavlicev, et al. (2007). The road to modularity. *Nature Reviews Genetics* 8(December): 921–931.

West-Eberhard, M. J. (2003). *Developmental Plasticity and Evolution*. Oxford: Oxford University Press.

Wimsatt, W. C. (2007). *Re-Engineering Philosophy for Limited Beings: Piecewise Approximations to Reality*. Cambridge, MA: Harvard University Press.

Woese, C. (2002). On the evolution of cells. *Proceedings of the National Academy of Science* 99(13): 8742–8747.

Woese, C. (2004). A new biology for a new century. *Microbiology and Molecular Biology Reviews* 68(2): 173–186.

Woese, C. (2008). The domains of life and their evolutionary implications. In M. J. Dunn, L. B. Jorde, P. F. Little and S. Subramaniam (eds.), *Encyclopedia of Genetics, Genomics, Proteomics and Bioinformatics*. New York: John Wiley & Sons.

II THE PROKARYOTE'S TALE

Brett Calcott and Kim Sterelny

Microbiology played almost no role in the establishment of the synthesis consensus (O'Malley and Dupre 2007). If it had, Mayr's biological species concept would not have dominated thinking about species and lineages for so long (Cohan 2002). The rise of molecular biology was an inestimable boon to microbiology, giving real tools through which microbial, and especially prokaryote, evolution could be studied. The results have been so surprising that they reshaped our picture of life's history. The depth and disparity of prokaryote evolution has been wholly unexpected, with the traditional "three kingdoms" model of the history of life being replaced by a picture recognizing the fundamental differences between the Achaea and the Eubacteria. The evolution of multicelled plants and animals is, from this perspective, just a couple of late-evolving side branches in one domain of the history of life (Ford Doolittle and Brown 1995; Woese 2008).

Most important, it has gradually became clear that evolutionary biology, in focusing on multicelled animals and plants, has ignored many typical processes of evolution, for in addition to radically reconfiguring our conception of the history of life, our new understanding of microbial evolution also forced a radical revision of received conceptions of mechanisms of evolution. As we noted in the introduction, the confirmation of the symbiotic origin of eukaryotes showed that one pivotal event in life's history was the formation of a cooperative alliance via lineage fusion, not an incrementally diverging lineage fission. Moreover, molecular systematics has clearly shown that horizontal gene transfer has played, and continues to play, a central role in prokaryote evolution (Ochman, Lerat et al. 2005). There no longer remains any doubt that horizontal gene transfer has been pervasive through the evolution of the bacteria and the Archaea. The live debate concerns whether horizontal gene transfer has been so pervasive as to undermine the branching tree model of prokaryote evolution (Cavalier-Smith 2010; Woese 2004), and whether horizontal gene transfer was even more pervasive and important in the origins of prokaryote cells (issues that are further discussed in Sterelny's Evolvability Reconsidered).

As a result of the molecular revolution, no one doubts that microbiology, like palaeontology, deserves its place at the high table of evolutionary theory. For one thing, prokaryotes are the nearest we have to a model of, arguably, the most challenging of all the major

transitions to understand, the evolution of the cell. As Woese has so vigorously argued, even the simplest living cell is extraordinarily complex, persisting only with the aid of an array of complex biochemical adaptations (Woese 2002). Lyon's main research project has been in cognitive biology: to argue that we can understand and explain the sophistication and subtly of prokaryote behavior only by modeling them as cognitive systems. As a consequence of this perspective, her chapter in this short section essentially underlines and reinforces Woese's message about cellular complexity. She points to the phenotypic distance between a putative, hypothetical ur-replicator or ur-chromosome and anything that can function as a cell, that is, that can effectively respond to its environment in ways that maintain its metabolic and physiological integrity. A first cell is a minimal evolutionary agent; a naked replicator, or chained set of replicators, is not. As Lyon notes, we are a very long way from having any candidate lineage explanation of an evolutionary trajectory from replicator to cell, a fact that some widespread metaphors (she notes) tend to suppress. Her chapter, then, is essentially a cautionary tale.

The next two chapters are more hopeful, and exploit a positive feature of the microbiological revolution for evolutionary theory: the use of bacteria to study evolution in action. The large population sizes, rapid growth rates, and evolutionary plasticity of prokaryotes make it possible to experimentally drive major evolutionary changes in bacterial lineages. Paul Rainey and Ben Kerr both run experimental labs to explore the circumstances in which bacteria evolve cooperative interactions, and the circumstances in which such interactions collapse. These two chapters review recent work done in these labs, and show how productively bacteria can be used as model systems for exploring the selective and developmental preconditions of cooperation, and its vulnerability to uncooperative invaders of various kinds.

Working with Joshua Nahum, Kerr reviews some recent and surprising work on the importance to cooperation of nontransitive competitive interactions, suggesting that rock-paper-scissors interactions may play a more important role than the occasional quirk in odd corners of ecology. If As beat Bs and Bs beat Cs and Cs beat As, tolerant As—those that make it possible for Bs to survive—do better than greedy, B-displacing As. For the greedy As allow Cs to swell in number by removing their enemy, thus eventually cutting their own throat, as those Cs displace the As. Of course, selection for tolerant As is effective only if the population structure is clumped, allowing the tolerant to enjoy the benefits of their B-ed and hence C-less neighborhoods. The mechanism is intuitive once explained, but Kerr and Nahum show not only that it is possible in principle, but that nontransitive competitive structures are probably both common and of great biological importance, in particular for the evolution of cooperation between unrelated lineages, such as the evolution of eukaryote cells.

In his joint chapter with Ben Kerr, Paul Rainey, likewise, reviews results from his experimental program and draws theoretical lessons from it. The experimental program probes both the environmental and genetic conditions under which bacteria evolve into

cooperative, quasi-multicellular films—in the right circumstances, a cooperative phenotype that allows them access to both nutrients and oxygen. But he also explores the breakdown of cooperation, in the course of developing a heterodox idea on the origins of the germ-soma distinction. Rainey suggests that this distinction might arise not as a form of cooperative division of labor (as Michod supposes, for example) but as a form of successful defection. Early germline specialists are successfully defecting against somatic suckers. As such, they may be precursors of a developmental cycle, for Rainey shows that the cooperative, film-forming morph readily re-evolves from the defecting, swimmer morph. In turn, swimming defectors easily re-evolve from second-generation film-making bacteria, and so on. Rainey and Kerr thus show a repeated biological sequence from multicelled films to one-celled dispersing swimmers to multicelled films. The sequence seems intriguingly like a developmental cycle, except that the stages are triggered by mutation rather than switching genes off and on. So in their different ways, both Rainey and Kerr show how productively bacteria can be used as model systems for exploring the selective and developmental preconditions of cooperation, and its vulnerability to uncooperative invaders of various kinds.

References

Cavalier-Smith, T. (2010). Deep phylogeny, ancestral groups and the four ages of life. *Philosophical Transactions of the Royal Society B* 365: 111–132.

Cohan, F. M. (2002). What are bacterial species? *Annual Review of Microbiology* 56: 457–487.

Ford Doolittle, W. F., and J. Brown. (1995). Tempo, mode, the progenote and the universal root. In W. M. Fitch and F. A. Ayala (eds.), *Tempo and Mode in Evolution: Genetics and Paleontology 50 Years After Simpson* (pp. 3–24). Washington, DC: National Academy of Sciences.

Ochman, H., E. Lerat, et al. (2005). Examining bacterial species under the specter of gene transfer and exchange. *Proceedings of National Academy of Sciences* 102(May 3): 6595–6599.

O'Malley, M., and J. Dupre. (2007). Size doesn't matter: Towards a more inclusive philosophy of biology. *Biology and Philosophy* 22(2): 155–191.

Woese, C. (2002). On the evolution of cells. *Proceedings of the National Academy of Sciences* 99(13): 8742–8747.

Woese, C. (2004). A new biology for a new century. *Microbiology and Molecular Biology Reviews* 68(2): 173–186.

Woese, C. (2008). The domains of life and their evolutionary implications. In M. J. Dunn, L. B. Jorde, P. F. Little and S. Subramaniam (eds.), *Encyclopedia of Genetics, Genomics, Proteomics and Bioinformatics*. New York: John Wiley & Sons.

6 To Be or Not To Be: Where Is Self-Preservation in Evolutionary Theory?

Pamela Lyon

Cooperation, it is commonly said, is a puzzle for evolutionary biology because of the intrinsic selfishness of living things (Queller 1997; Sachs et al. 2004). So foundational is selfishness to the contemporary theory of natural selection that the most influential account of the evolution of cooperation and altruism, kin selection, is based on it (Lehmann et al. 2007; Sober and Wilson 1998). Organisms are willing to moderate or even sacrifice their own existential imperatives to be, to grow, and to reproduce for kin, first and foremost, because kin are the closest to self, genetically speaking. In this chapter, I argue that the evolution of selfishness is itself a puzzle, one rarely apprehended much less addressed. I am not concerned here with the psychological implications of the term selfishness, the units of selection debate, or any other battleground of the sociobiology conflict. Rather, I am concerned with the simple idea that at some point, in the history of life on earth, a self-ordering, autocatalytic chemical system developed characteristics to which selfishness could be attributed.

The various hypotheses of the origin of life and the major transitions of evolution currently on offer, henceforward referred to as origins and transitions narratives—whether replicator-first (Maynard Smith and Szathmáry 1995), metabolism-first (Wächtershäuser 1988), RNA world (Gilbert 1986), lipid world (Segré et al. 2001), peptide world (Nelson et al. 2000), virus world (Koonin et al. 2006), communal, gene-swapping progenotes (Woese 1998), biological big bang (Koonin 2007), or panspermia (Hoyle and Wickramasinghe 2000)—all share the assumption, usually tacit, that somehow selfishness entered the world. Put another way, they assume the emergence of the sort of self-preserving, self-organized complexity that provides a minimal basis for attributing selfishness to a system. So potent was the emergence of this trait that evolutionary theorists still ponder and debate about how it was overcome, because overcome it must have been (so it is said) to produce the routine wonders of evolution that abound today.

Selfishness cannot simply be assumed, however. That a chemical system developed the impetus not merely to replicate, like a crystal, but to replicate specific sequences of complex molecules and persist against existential threat, are phenomena that demand explanation. None of the inorganic self-ordering, autocatalytic, dissipative structures known to

contemporary science does this (Abel and Trevors 2006; Kauffman 2000; Orgel 1992). A crystal, a candle flame, a hurricane, or a Bénard cell does not seek resources when the material conditions for continued catalysis runs out; they cease. Living things do so until all options are exhausted. Some of the simplest organisms engage in surprisingly elaborate behaviors to forestall cessation.

In *On the Origin of Species*, Charles Darwin observed that it is "so easy . . . to think that we give an explanation when we only restate a fact" (1996, 389). Darwin's admonition was aimed at those who uncritically invoked "such expressions as the 'plan of creation,' [and] 'unity of design,' " to explain the abundant diversity of life and the neat adaptive fit between an organism and its environs. I claim that Darwin's admonition applies to many current theoretical deployments of selfishness in evolutionary biology. So routinely and uncritically is selfishness invoked that it does seem to pass for explanation. This is a mistake, I believe, for two reasons.

First, assuming selfishness as a universal brute trait of prebiotic and biological entities obscures a major transition in the history of life on earth. This is the transition from a self-replicating, autocatalytic chemical system to what Godfrey-Smith (2009; this volume) calls a "Darwinian individual." Following Lewontin, Godfrey-Smith sets out three characteristics of a Darwinian individual: it is a member of a population of entities that (a) differ from one another in some character relating to morphology, physiology, or behavior (variation), in ways that (b) make a difference to the individual's survival and/or reproductive success (fitness) and (c) can be passed to offspring (heritability).

A Darwinian individual, so defined, need not be actively self-preserving and/or self-extending. In *biological* evolution, however, the characteristics of active self-preservation and self-extension via growth and reproduction are simply facts about whatever biological entity is under consideration. Viruses and prions are exceptions to the growth condition, but the rapid evolvability of the viral protein coat provides evidence of self-preservation. Thus, the characteristics of self-preservation and self-extension are typically taken for granted when selfishness is invoked in simulations and/or explanations of biological evolution.

The propensity of an individual entity to actively preserve and extend itself also tends to be assumed in the origins and transitions narratives currently on offer. Often the gap is subsumed under the (ill-defined) trait of selfishness or the equally abstract concept of competition. Such assumptions are only legitimate, I believe, if they are promissory notes for later explanation. For the most part, they do not appear to be. It would seem that "everybody knows" selfishness is a given, so theorizing typically moves on from there. The problem is, what everybody knows often turns out to mask unseen complications (McShea 1991).

The uncritical assumption of selfishness creates a fundamental instability in much current work on the evolutionary transitions and origins of life. Some of the leading transitions narratives assume selfishness explicitly (Michod and Roze 1997) or implicitly (Maynard

Smith and Szathmáry 1995) at their foundation. All current origin-of-life scenarios of which I am aware also appear to presume the emergence of selfishness, or the kind of self-organized complexity that supports the attribution of similar concepts, at a very early stage, well before cells appear. Since this assumption is unsupported, origins narratives remain incomplete until the emergence of self-preserving behavior is explained.

This chapter is organized into two parts. The first part sketches what self-preserving and self-extending behavior in a chemical system minimally involves, and demonstrates how this kind of behavior is elided in several origins and transitions narratives but principally in the RNA World hypothesis and *The Major Transitions in Evolution*. The second part advances an explanation of why this has gone largely unremarked. My claim is that Richard Dawkins introduced a new concept into biology with publication of *The Selfish Gene*. The concept of Dawkinsian selfishness profoundly changed the way many biologists think about their subject (Grafen and Ridley 2007) by making cooperation seem so problematic, but it also encouraged many to think that self-centered or self-serving behavior is not problematic. Dawkinsian selfishness thus has served as a sort of sticking plaster covering up an evolutionary transition.

Before we begin, let me be clear: I do not doubt that self-preservation—the inchoate striving to maintain system integrity against threat and to perpetuate existence via reproduction—is the default existential condition of living things, present and past. But the emergence of that default state needs to be explained. This is the truck-sized gap in origins and transitions narratives that Dawkinsian selfishness obscures. Once a plausible account of self-preservation emerges, I suspect that developing the trait of selfishness is relatively straightforward. A plausible account of self-preservation relies, in turn, on a genuinely explanatory account of *biological* self-organization. This we also do not have and badly need (Abel and Trevors 2006; Kauffman 2000). Most of the literature on biological self-organization trades heavily on the patent capacity of organisms to structure and continually manufacture the components and processes that comprise them, and includes a great deal of hand-waving in the direction of self-organizing processes in the physical sciences. These processes, in particular nonclassical thermodynamic approaches, provide a platform for biological organization, but no more. As others have suggested (e.g., Corning 2005; Rosen 2000), new concepts and ideas very likely will be needed to clear the remaining distance. Biological self-organization cannot be dealt with adequately here, however. Finally, I have no answers to these problems, only many questions. But that is a start.

Self-Preservation and the Evolutionary Transitions

The task in this section is to describe a void and demonstrate its existence. How to do this? To describe the absence, I will first describe what should be present. To demonstrate the existence of the void in contemporary origins and transitions narratives I will sketch four examples: the currently dominant RNA World scenario for the origin of life, as described

by one of its founders (Orgel 1968, 1992, 2003, 2004); Maynard Smith and Szathmáry's influential, information processing–based account of major evolutionary transitions (Maynard Smith and Szathmáry 1995); and, briefly, two accounts of the origin of cells (Koonin et al. 2006; Woese 1998).

Before we can characterize a self-preserving system, we must know something about the contrast case, a garden variety dynamical, self-organizing chemical system. I beg the reader's patience with this highly simplified presentation, which is no more than an enumeration of necessary entities and their relations.[1] It lays the foundation for later discussions.

Chemical Systems and Self-Preservation

In brief, a chemical *system* is a set of interacting or interdependent *molecules* (relatively stable aggregations of two or more atoms) that form a more or less integrated whole. Molecules are held together internally by *covalent bonds*, which involves electron sharing between atoms, and by the *van der Waals force*, which generates attraction or repulsion at the quantum level mainly through electrostatic interactions and atomic polarization. Molecules are joined with one another by van der Waals forces and ionic/polar bonds, which involve electron transfer between atoms that affect charge and thus the capacity to attract or repel other atoms. *Chemical reactions* occur when different chemical substances—atoms or molecules called reactants, or substrates—are brought into contact with one another, resulting in their conversion into one or more chemical products, which typically have a different structure and different reaction properties. *Catalysts* are chemical substances that influence the rate of chemical reactions; they can be inorganic (i.e., metals) as well as organic atoms and molecules. Classically, catalysis involves the motion of electrons in forming and breaking chemical bonds.

A chemical reaction is said to be *autocatalytic* if a product of the reaction provides a substrate, the thing the catalyst acts on, for the continuation of the reaction. An example is the spontaneous breakdown of aspirin into salicylic acid and acetic acid, which results in the vinegary smell of very old aspirin in sealed containers. An *autocatalytic set* is a collection of interacting chemical entities, each of which can be catalyzed by other members of the set, such that the collection as a whole is capable of catalyzing its own production, or sustaining itself, given adequate inputs of energy and matter. *Interaction* in a chemical system is simply the process by which change occurs, and a *dynamical system* is one whose behavior changes over time, continuously, in discrete increments, or a combination of both.

Many autocatalytic systems are *self-organizing*. In physics and chemistry, self-organization refers to increasing complexity of internal structure via the processes of attraction and repulsion arising from intermolecular interactions within the system itself under prevailing conditions (temperature, pressure, etc.). A wide variety of physical and chemical phenomena self-organize: crystallization, superconductivity, lasers, cyclones and *dissipative structures,* such as convective instabilities (e.g., Bénard cells). Such order-forming chemical reactions are called "dissipative" because they exist far from thermody-

namic equilibrium—the molecules that comprise them are kinetically active and chemically reactive. They generate heat and dissipate entropy. Many self-organizing chemical systems exhibit *emergent properties*, or characteristics qualitatively different from those exhibited by their constituents alone. Classic examples of emergent properties are the multiple forms (liquid, gas, ice), densities and solvent properties of water, which cannot be predicted from the properties of hydrogen and oxygen alone. Vitality is a complex emergent property.

In all known cases of self-ordering, dissipative systems that display emergent properties, autocatalysis continues until the conditions for the system's continuation cease. The major difference between such nonbiological systems and biological systems is not the ability to replicate—given inputs of matter and energy, autocatalytic sets will replicate if split into physically separated spaces. Rather, the major difference is the ability of a biological system to "reach out and manipulate the world on its own behalf" (Kauffman 2000, 49). A candle flame gutters and dies when the wax is exhausted; it doesn't search for more. The same is true for a Bénard cell, a laser, a hurricane or a Belousov-Zhabotinsky reaction when the conditions for their existence cease.

By contrast, in conditions of nutrient limitation, a population of *Bacillus subtilis* will synthesize enzymes to scavenge alternative sources of nutrients (Msadek 1999). If that fails, a portion of the bacterial population suicides through autolysis, which means individual cells break open their membrane and literally spill their guts, providing a temporary source of food for the remaining members of the population in the event conditions are about to change (Gonzales-Pastor, Hobbs, and Losick 2003). If conditions remain dire, the population commits to sporulation, an irreversible, do-or-die process in which cells transform, over eight hours, into spores capable of resisting injury by heat, chemicals and ultraviolet radiation and remaining dormant (sometimes for years) until life-sustaining conditions return.

As are all living systems, *B. subtilis* is self-preserving. Replication, although the principal concern of evolutionary biologists computing change in the frequency of alleles or phenotypic traits over time, is not the organism's sole *raison d'etre.* The bacterium acts as if it has interests in its own persistence, not merely in its offspring, and will go to great lengths to preserve itself. Strictly speaking, spores are not progeny but the product of metamorphosis, like butterflies from caterpillars.

Stuart Kauffman (2000), an early proponent of autocatalysis as a key to life's origin, observes that vitality as we know it involves production of thermodynamic work cycles enabled by feedback-regulated constraints, allowing the *controlled* acquisition and release of energy. The behavior of nonbiological chemical systems is not shaped by such constraints. Thus, such systems demand "a new concept of 'organization' that is not covered by our concepts of matter alone, energy alone, entropy alone, or [Shannon] information alone" (Kauffman 2000, 4). The observation that biological organization is qualitatively different from the kind of order generated in inorganic chemical systems has a long history (for modern examples, see Rosen 1985; Rosenblueth, Wiener, and Bigelow 1943; von

Bertalanffy 1968). In sum, what is missing from contemporary origins and transitions narratives is the shift from dynamical, self-ordering autocatalysis to a kind of organization that involves the construction of constraints to prolong and preserve autocatalysis, that is, system-preserving behavior.

The RNA World

The origin of life is, notoriously, a chicken-and-egg problem involving a macromolecule that copies itself (the nucleic acid DNA) in dependence on protein enzymes, long chains of amino acids capable of catalyzing reactions, which are themselves synthesized from nucleic acids (RNA). Life's origin thus initially resolves into a question of which came first, nucleic acids or proteins (Orgel 1968). The Miller-Urey experiments in the early 1950s showed that amino acids could be synthesized in conditions believed to simulate prebiotic earth, thus providing empirical support for a protein-first hypothesis. By the late 1960s, however, three researchers in different disciplines—microbiologist Carl Woese (1967), geneticist Francis Crick (1968), and chemist Leslie Orgel (1968)—independently concluded that nucleic acids were the more likely progenitor. Specifically, they hypothesized that the DNA-and-protein enzyme world of contemporary biochemistry was preceded by a form of life based on RNA genes and supposed RNA-based enzymes, which were then only theoretical entities. The unexpected discovery in the early 1980s of RNA enzymes (ribozymes) provided the necessary mechanism for the RNA World, as the hypotheses became known (Gilbert 1986). Today, it is the leading hypothesis for the origin of life.

The main challenge for the RNA World scenario is to explain the prebiotic synthesis of a relatively large complex of molecules capable of reproducing itself as a unit with a reasonable degree of fidelity. Such a macromolecule must be capable of carrying information about its sequence. According to Orgel, "If RNA was the first carrier of 'biological' information, then the task of prebiotic chemistry is to understand the way in which ribonucleotides were formed abiotically on the primitive earth and the way in which they were polymerized to form the very first strands of self-replicating RNA" (Orgel 2003, 211). For Orgel, who pursued the RNA World hypothesis vigorously but critically for four decades, the concept of information transfer in replication was key to demonstrating that "a chemical system . . . is capable of undergoing [D]arwinian evolution" by natural selection (Joyce 2007, 627). Once an informational macromolecule emerged, Orgel believed, the rest of evolution "is just history."

The degree of empirical support for the RNA World hypothesis is not central to my argument. However, Orgel's pessimistic estimation of the evidence supporting his hypothesis places in useful perspective unarticulated assumptions about system-preserving behavior in the development of life, so I will briefly outline it here. In his last review of progress on the RNA World hypothesis before his death in 2007, Orgel reports that "there is at present no convincing, prebiotic total synthesis of *any* of the nucleotides" necessary to support

RNA synthesis (Orgel 2004, 108; my italics). Further, no evidence has yet emerged linking ribozymes, "the smoking gun that . . . led to the more general acceptance of the RNA World hypothesis," to genetic transcription or RNA replication (Orgel 2004, 113). "[A]biotic synthesis of RNA is so difficult," Orgel concludes, "that it is unclear that the RNA World could have evolved *de novo* on the primitive Earth" at all (100), a conclusion first advanced by Cairns-Smith 30 years ago. If this improbable yet still conceivable de novo evolution did occur, Orgel writes, "it erects an almost opaque barrier between biochemistry and prebiotic chemistry," because the advent of RNA and biochemistry would have changed the chemical landscape profoundly and forever (100). One "casualty" of this reading of the RNA World, Orgel observes, is that resemblance arguments from contemporary biochemistry to the origin of life—"the majority of speculations"—lose their power (2003, 212).

Thus, in the opinion of one of its leading advocates,[2] scientific investigation of the RNA World hypothesis has yielded much interesting chemistry in the past 40 years but little progress toward an adequate explanation for the origins of informational molecules, much less life. The language Orgel uses to sketch "some consequences of the RNA World hypothesis" is all the more notable, therefore, because an informational molecule is still only a hope, not a reality. It is the sort of language used commonly in origins and transitions narratives, however, so I will quote several passages from Orgel's paper (2003) to illustrate the point. Italics have been added.

> *Pre-RNA organisms* must at least have been able to carry out ribose synthesis and phosphoester bond formation. (213)
>
> More positively, if some *catalytic functions* of the RNA world were never taken over by ribozymes, it might after all be possible to make some tentative inferences about the pre-RNA world . . . (214)
>
> An organism using the earlier genetic polymer synthesizes the monomers of the second system because they *confer some advantage* unrelated to replication. (214)
>
> However, in any *organized biological world* the properties of a vast library of "secondary metabolites" would be explored . . . (214)
>
> If the RNA World hypothesis is correct, RNA must have *picked and chosen* among the freely available prebiotic [amino acids], if any were available, and added different [amino acids] that it "*learned*" to synthesize. (212)

Organism was the name Aristotle gave to living things because their heterogeneous, ordered structure enables activities that ensure "the good" of the thing. In biology, task-discharging biochemical processes are called *functions*. Self-organization *in biology* thus requires not simply self-ordered complexity but function, a process or structure that materially contributes to operation of a larger system. Function requires chemical processes that rely on cybernetic (feedback-based) control, Kauffman's constraints, and ultimately algorithmic rule-following of the type *if in state X, do Y* (Trevors and Abel 2004).

The concept of function in biology is notoriously polysemic, however. Mahner and Bunge (2001) identify five meanings, each appropriate to a different explanatory context.

Two meanings refer to the value of the activities to the organism as a whole, considered in terms of their current value or history of selection. The remaining meanings refer to activities, usually in relation to some other, without reference to value and are sometimes called "effects." These meanings come closest to that used in chemistry, where a group of atoms responsible for a characteristic reactive property of a molecule is called a *functional group.* In biology, however, such activities remain—always—within the context of a self-preserving system, whether or not the explanatory target is the entire system, whether or not the activities are being studied in vivo or in vitro. System preservation—minimally, the controlled acquisition and release of energy and matter—depends on regulated processes. Without regulated processes, system preservation of the kind associated with life is impossible. System preservation is the overarching existential function of biochemical processes, at least until reproductive imperatives gain primacy.

Common modifiers in evolutionary theory such as "advantageous" and "fitness-enhancing" thus often imply a degree of biological organization to which existential goals can plausibly be attributed. What is advantageous to an informational macromolecule? Removed from the context of the cell, RNA does nothing functional in a biological sense. RNA World experiments designed to select for polymerization activity directly from random-sequence RNA (giving prebiotic chemical evolution a leg up, as it were) so far "have yielded only ribozymes that decorate themselves inappropriately with tagged nucleotides" (Johnston et al. 2004, 1324)—inappropriately, that is, compared to RNA within a self-preserving biological entity.

Function matters only where there is an implicit benchmark against which advantage and fitness mean anything at all. In the sort of organization subject to Darwinian selection of which we are aware, the benchmark is, first, system preservation and, second, system replication. System preservation is a plausible reason why all "known paradigm cases of evolution by natural selection depend on the high-fidelity copying of genetic material" (Godfrey-Smith 2009, 145), and why DNA repair mechanisms are so efficient. Although he might not agree with my argument about self-preservation, Godfrey-Smith concludes (as I do) that "genes themselves in most cases are marginal Darwinian individuals" (145).

In short, Orgel assumes that the emergence of an informational macromolecule is the hard part in arriving at an adequate explanation of the prebiotic origins of life. He may be right. However, we have no reason to expect that system-preserving behavior comes for free with informational replication. As we will see in the following sections, Orgel is not alone in his assumptions.

Major Evolutionary Transitions and Information

In *The Major Transitions in Evolution* (MTE), Maynard Smith and Szathmáry provide a detail-rich framework for theorizing about life's origins and the growth of biological complexity based on innovations in information storage and transmission between generations,

from the birth of replicating molecules to human language. Of the eight transitions they identify, the first four are germane to our discussion:

1. Replicating molecules → Populations of molecules in compartments
2. Independent replicators → Chromosomes
3. RNA as gene and enzyme → DNA + protein (genetic code)
4. Prokaryotes → Eukaryotes (Maynard Smith and Szathmáry 1995, 6)

By the time an entity has emerged that warrants the label "prokaryote," system-preserving behavior (e.g., cybernetic constraints, algorithmic processes) clearly has also emerged. None of the remaining four transitions in MTE refer explicitly to self-preserving behavior, either. However, as MTE's transitions are concerned with innovations in information storage and intergenerational transmission, it must be assumed that a system exists to which fidelity of transmission and/or increased storage capacity makes an existential difference. Herein lies the problem.

As with the RNA World, the plausibility of MTE is not important for our purposes. What is germane is the role that competition plays, right from the beginning of the account, as a result of the authors being "committed to the gene-centred approach outlined by Williams (1966) and made still more explicit by Dawkins (1976)" (Maynard Smith and Szathmáry 1995, 8). What does this commitment mean? Williams's and Dawkins's presentations are not equivalent, and Maynard Smith and Szathmáry are not entirely clear about what they are taking on board. Of the two accounts, Williams's is, in his seminal work at least, by far the most perspicuous and constrained. Against selection at the species or group level, Williams argues that natural selection "arises from a reproductive competition among individuals, and ultimately among genes," which are selected "on one basis only, [their] average effectiveness in producing individuals able to maximize the gene's representation in future generations" (Williams 1966, 251). Natural selection targets individuals and, ultimately, an individual's genes, particularly those that contribute to reproductive success. In the language of the RNA World, competition requires an informational molecule and something like a cell, which can make its way in a Darwinian scenario involving variation, fitness and heredity.

Dawkins takes Williams's basic argument considerably further in *The Selfish Gene* (1989), by combining it with developments in the gene's-eye view of the evolution of social behavior by Hamilton, Axelrod, and Trivers, mixing in Maynard Smith's application of mathematical game theory to evolutionary biology, and finally adding to this melange his own Hobbesian intuitions about (nonhuman) biological nature. The account begins with a "creation myth" (Sterelny 2001) about the emergence from the prebiotic soup of a self-copying molecule Dawkins calls a *replicator,* a terminology subsequently widely adopted, including in MTE. According to Dawkins, "a predominant quality" to be expected of a successful gene and its replicator ancestors is "ruthless selfishness" of a sort exhibited by

"Chicago gangsters," evident in their survival in a "highly competitive world" (Dawkins 1989, 2). We will discuss Dawkinsian selfishness in greater detail in the next section.

According to Dawkins, in the early stages of developing life, natural selection principally favored replicators possessing one or a combination of three stability-enhancing capacities: (1) persistence in a hostile environment (longevity); (2) speedy replication (fecundity); and (3) relatively faithful replication (accuracy). Competition among primordial selfish replicators thus involves increasingly elaborate "ways of increasing [their own] stability and of decreasing rivals' stability" (Dawkins 1989, 19). Replicators endowed with these characteristics grow most numerous and are the victors in the struggle for existence.

The point to notice here is that a Dawkinsian replicator is an agent; it has implicit interests in the outcome of the struggle. We may say that Dawkins's muscular prose is simply metaphorical, to underscore the tumultuous conditions under which the original replicator would have had to persist and evolve, but the metaphor obscures a significant transition in evolution: from a self-ordering, autocatalytic system to a self-preserving one, at least the beginnings of one. Because this suggestive but unelaborated property emerges at the beginning of informational replication—that and the complex substrate copied are what set the replicator apart from mere crystals—it need not be accounted for thereafter. System-preserving behavior thus is as hard-wired into the genes that succeed the primordial replicator as anything can be. Whatever else might be said about Dawkins's account, this move is problematic, as we have already seen.

Maynard Smith and Szathmáry adopt Dawkins's general scenario, albeit unadorned with the rhetoric of selfishness. They interpret the gene-centered view as imposing an explanatory constraint and engendering an important entailment at a very early stage of the emergence of life on earth. The constraint requires that each transition "be explained in terms of immediate selective advantage to individual replicators" (Maynard Smith and Szathmáry 1995, 8). The entailment is that, somehow, competition among replicators must be suppressed from the get-go to make each transition possible. Cooperation must evolve at each stage of innovation—starting from the transition from replicating molecules to compartmentalized replicating molecules—so that individual interests are subsumed within higher-order interests.

The necessary subordination of competitive behavior is accomplished by invoking Hamilton's rule, or kin selection: Cooperation can evolve where the genetic relatedness between cooperator and beneficiary is high and, generally speaking, the cost to the cooperator is not too onerous (Hamilton 1964a,b). While Hamilton proposed his mechanism specifically with respect to the evolution of social behavior among individual organisms, Maynard Smith and Szathmáry take the principle "to be quite general" (Maynard Smith and Szathmáry 1995, 8). They believed it significant, for example, that in the development of multicellular organisms, "At some point in the life cycle, there is only one copy, or very few copies, of the genetic material; consequently, there is a high degree of genetic relatedness between the units that combine in the higher organism" (8).

Adoption of the Dawkinsian replicator creates a tension in MTE, of which (I think) the authors are aware, at least in subsequent work (Szathmáry and Maynard Smith 1997). An example of what I mean is their treatment of the chemoton model of an early prebiotic system, proposed by Tibor Gantí. While the chemoton model is based on the existence of an informational molecule, they note that Gantí was "the first to call attention to autocatalytic cycles from the point of view of evolution. He pointed out that if two cycles are operating in the same environment, then the one with the larger kinetic rate constant can outgrow the other. This process is analogous to the competitive replacement of one species by another" (Maynard Smith and Szathmáry 1995, 34).

The implication is that it is useful to think of prebiotic autocatalytic systems as competing with one other. Yet evolution requires "more than autocatalysis"; it requires that "occasionally, a new variant chemical compound—a mutant—should arise, and, once arisen, should be replicated" (Maynard Smith and Szathmáry 1995, 35). But how does autocatalysis give rise to a "mutation"? Wouldn't that just be a change in the products of a cyclic chemical reaction, which (if stable and the necessary inputs keep coming) then become part of the cycle? Such "mutation" would be growth or differentiation rather than the production of a new and variant individual. Maynard Smith and Szathmáry have already pointed out that autocatalysis is "an important first step towards replication, but it is not the whole road" (1995, 21). Yet there is a more or less continual back and forth between talk of autocatalysis and talk of replicators, as though they are equivalent.

Another example is Maynard Smith and Szathmáry's use of the peroxisome as an example of a *simple replicator,* which they define as "a structure that can arise only if there is a preexisting structure of the same kind" (Maynard Smith and Szathmáry 1995, 41). A peroxisome is a lipid bilayer membrane enclosing enzymes that replicates by enlargement and simple division. It is an organelle (sans DNA) found in most eukaryotic cells, and is believed to be the vestige of an ancient symbiotic event. This means that a peroxisome is thought to have once been a free-living, self-preserving organism. How, then, does the existence and replicating behavior of a peroxisome provide any leverage at all on our understanding of how a simple informational molecule evolved? The simple answer is, it doesn't. Secondary simplification is no model of simple origination. The example illustrates *something*, but it doesn't do the work the theorists intend.

While "simple replication" is insufficient for the kind of information storage needed for life as we know it, according to Maynard Smith and Szathmáry, it is essential for their thesis that major transitions require the suppression of competition and the evolution of cooperation. "Once replicating molecules existed, relatedness between neighbours, arising because of limited movement, could have been important. . . . But prior to the origin of replication, relatedness is a meaningless concept" (Maynard Smith and Szathmáry 1995, 35). There is no explanation of why replication might make relatedness suddenly meaningful, but for competitive Dawkinsian replicators, there is no need to do so.

There is no question that Szathmáry and Maynard Smith find the Dawkinsian replicator concept "extremely useful in analysing evolutionary questions" (Maynard Smith and Szathmáry 1997, 558). Charting the transition from replicator to reproducer, they observe that the first replicators

> must have been relatively small molecules, because, in the absence of specific replicases, copying would have been inaccurate, and large molecules would have accumulated errors. This raises the central problem of how cooperating groups of small replicators could have arisen, and of how they could have been protected against invasions by molecular parasites (Szathmáry and Maynard Smith 1997, 555–556).

But how does an existential imperative arise in a group of autonomous, self-ordering chemical systems such that protection against parasites is necessary? How is such protection to be effected? And what, exactly, are such parasites parasitic on? These are nontrivial questions. Again, what we don't notice is the segue from an autocatalytic, self-ordering chemical system to one that competes. Moreover, this competitive system has sufficient interest in the outcome that this putatively normal behavior—the behavior that got it to this point in evolution—has to be *suppressed* to enable transition to the next stage of (system-preserving) complexity. Indeed, to some evolutionary biologists, this suppression provides the "unmistakable footprints of . . . natural selection" (Leigh 1999, 30). Yet the genesis of this highly significant property-to-be-constrained remains deeply obscure, a brute fact.

Other origins and transitions narratives, however, fare no better on this point. Woese proposes a scenario for the evolution of cells in which "[t]he universal ancestor is not a discrete entity, but, rather a diverse community," composed of very simple entities called "progenotes," which "survives and evolves as a biological unit" (Woese 1998, 6854). The progenotes of this early communal stage have inaccurate "information processing systems" and engage in promiscuous horizontal gene transfer. Woese envisions a period when "genetic temperatures" are "very high" but then "cool" in a process similar to physical annealing, from which stable new structures emerge. After this "genetic annealing" protocells evolve, differentiate, and depart the commune to make their way. Where or how system-preserving behavior emerges in these new, more stable entities remains obscure.

Unlike Woese's progenotes, the replicating entities that populate the gene-swapping virus world proposed by Eugene Koonin and colleagues (Koonin, Senkevich, and Dolja 2006), however promiscuous and communal, are explicitly described as selfish. These include both "parasitic elements" that become viruses and "selfish cooperatives" that become cells. What selfish means is unclear; that the entities are so is simply a given. Eventually, individuals "escape" from the commune "to preserve selective advantage," but what this means is also unclear. Selective advantage to "do" what? Why?

In sum, MTE assumes that the emergence of a replicating molecular complex coincides with the emergence of structures with self-preserving characteristics. Perhaps by contemporary standards, this is not a fault. But the two characteristics are not the same. Moreover, as Godfrey-Smith argues, being an informational replicator in a population of such replica-

tors probably doesn't qualify a system as a Darwinian individual displaying the three characteristics of variation, fitness, and heredity (Godfrey-Smith 2009). In the final section, I conjecture a reason why the "missing transition" has not been missed.

A Conjecture about Selfishness

The failure to account for the emergence of self-preservation has passed unremarked, I believe, largely as a result of the influence of *The Selfish Gene*, which introduced the concept of universal biological selfishness to evolutionary discourse. While Dawkins's extreme genic selectionist views (since moderated) have been subject to criticism from the book's publication to more recent times (Lewontin 1977; Rose and Rose 2000), Dawkinsian selfishness, by contrast, has been thoroughly integrated into contemporary thinking about evolution (Sober and Wilson 1998). The following statements are utterly uncontroversial:

> Darwinians begin with a bias toward seeing life as fundamentally selfish because competition is a logically necessary part of selection, while cooperation is not (Queller 1997, 187).
>
> [T]he essential theoretical problem is to elucidate how cooperative behaviour can originally evolve in a selfish world and how, thereafter, it can be maintained against invasion by selfish individuals. (Killingback, Doebeli, and Knowlton 1999, 1723)

Moreover, Dawkins's original intellectual contribution is now reflexively attributed to the more scientifically rigorous, less popular sources that influenced him, for example:

> . . . Hamilton (1964a,b) and George Williams (1966) explained how natural selection was intrinsically selfish, and that cooperative acts were likely to evolve only under restrictive conditions. (Sachs et al. 2004, 136)
>
> As evolutionary biologist W. D. Hamilton showed 40 years ago, selfish genes can lead to cooperation and altruism. (Queller 2004, 975)

I wish to show that Hamilton did no such thing, nor did Williams, Maynard Smith, or Darwin—theorists Dawkins cites as key influences. I claim that there is a clear boundary in discourse about the nature of the biological world pre-Dawkins and post-Dawkins. Pre-Dawkins, there were advocates of the individual and the gene as the basic unit of selection, but no one made the explicit inference: "the fundamental unit of selection, and therefore of self-interest" (Dawkins 1989, 11). Dawkins's equation of the unit of selection with self-interest was unique.

Dawkins claims *The Selfish Gene* is wholly consistent with Darwin's theory and declares its provenance in four major developments in the 1960s and early 1970s: (1) William's gene-centered approach to evolution; (2) Hamilton's kin selection model of the evolution of social behavior; (3) Trivers's game theoretic account of the evolution of reciprocal altruism; and (4) Maynard Smith's development (with and without Price) of evolutionary game

theory. I will demonstrate that, at the time he wrote the book at least, Dawkins could not have justified universal biological selfishness on the basis of these sources.

While altruism clearly was a puzzle for Darwin, it wasn't because of the selfishness or self-interest of organisms, terms he apparently reserved for human personality traits. Word searches for "selfish," "selfishness," and "self-interest" in the online editions of *The Origin of Species* (first and sixth editions), *The Descent of Man,* and *The Voyage of The Beagle* (see www.Literature.org) disclosed one reference each in *Descent* of "selfish" and "selfishness" and two references for "self-interest" (one each in *Voyage* and *Descent*), all referring to human behavior. The struggle for existence was enough to stand self-sacrificing behavior in need of explanation; selfishness wasn't required. For Darwin, selfishness—which in standard dictionary meaning implies action at the expense of others or with reckless disregard for the interests of others—was an extreme behavior.

While Hamilton, even more than Williams, is credited with showing how natural selection favors the selfish except in limited circumstances, this is not how Hamilton saw his own work. Initially, Hamilton's problem was to account for altruistic behavior—which he believed to be real, not some sort of delusion (Hamilton 1975)—in the context of Fisher's "genetical" approach to natural selection, which emphasized the individual as the unit of selection (Hamilton 1963). However powerful, Fisher's approach couldn't counter the claim, then pervasive, that altruism evolves because "natural selection favour[s] the most stable and cooperative groups" (Hamilton 1963, 354). Hamilton's solution was *inclusive fitness*, the idea that the fitness of interacting individuals—roughly, their chances of survival and reproduction—includes the fitness of their close genetic kin. Altruism, and cooperative sociality generally, could thus evolve among relatives because they share genes. To the extent that "genes favoring altruism" enhanced fitness, such genes would become more prevalent (Hamilton 1964a,b).

In his seminal papers, Hamilton uses selfish in its ordinary meaning: Selfish traits induce behavior that *harms* the interests of conspecifics at a fitness benefit to the agent; traits are altruistic that induce behavior that *benefits* the interests of conspecifics at a fitness cost to the agent. Hamilton consistently contrasts altruistic traits with selfish traits (Hamilton 1964a, 13–16), where both characteristics are extremes, not the behavioral norm. The first example Hamilton cites of a selfish genetic trait is the "killer" trait of *Paramecium aurelia* (Hamilton 1964a, 13), now known to be a bacterium that produces a toxin fatal to the protist's uninfected conspecifics. Also cited as selfish is the laying of male eggs by worker ants. By contrast, Hamilton does *not* regard as selfish occasional female egg laying by *Apis mellifera* workers, because they don't "try to get their eggs cared for in queen-cells" (Hamilton 1964b, 35).

A subsequent paper shows that mere individual competitive advantage in the struggle for existence doesn't typically equate with selfishness in Hamilton's early work. Hamilton (1970) addresses the evolution of selfish and spiteful behavior on grounds that his 1964 model cannot adequately explain them. "Biological selfishness" is defined by extreme ex-

ample: "Incidents in which an animal attacks another of the same species, drives it from a territory, or even kills and devours it" (Hamilton 1970, 1218). In sum, while Hamilton rightly may be said to have introduced selfishness into theorizing about the evolution of social behavior, he did so in a limited way.[3] Moreover, his deployment of the term, which remained consistent with ordinary usage, highlighted a class of behavior—selfish and spiteful behavior—that he believed required explanation just as much as altruistic behavior does, precisely because it is extreme. Hamilton's work thus cannot properly be said to have "explained how natural selection is intrinsically selfish."

Even more telling is Hamilton's review of two books the year before *The Selfish Gene* appeared, a brilliant little gem of artful, well-reasoned prose that contains many passages that could easily have referred to Dawkins's book. I will quote just one at length, to show that Hamilton was no believer in universal selfishness. Taking issue with Ghiselin's (1974) thesis that, "The economy of nature is altogether individualistic, and altruism is a metaphysical delusion," Hamilton observes that Ghiselin would have us accept that organisms

> adapt on much the same principles as men are supposed to act in the pages of Adam Smith, towards the maximization of individual advantage in every case. . . . But just as the *laissez faire* model has shown inadequacies in economics, so even more obviously the idea of total individualism is inadequate for the rest of the living world . . . The economic model is certainly a useful guide to insight and has been recognized as such by various biologists since Darwin . . . *but no analogy can fully substitute for careful thought about the particulars of a problem*. (Hamilton 1975, 176; my italics)

Williams is another kettle of fish. As did so many, Williams had a conversion experience on encountering *The Selfish Gene*, declaring himself "a proponent of the more extreme contemporary view of natural selection as a process for maximizing selfishness" (Williams 1988, 399). In the work that influenced Dawkins, however, Williams resiled from such loaded rhetoric. In a discussion of social adaptations, in fact, Williams argued vigorously against the use of terms "burdened with value judgment and emotional flavour," such as altruism, and proposed a value-neutral terminology (Williams and Williams 1957, 32–33). "Altruist" and "cooperator" were to be replaced with "social donor" or, simply, "donor." Noncooperators—now routinely labeled as selfish or "cheaters"—were "nondonors." This usage appears throughout *Adaptation and Natural Selection* (1966). I believe Williams used the term "selfish" once in his masterwork, in a passage aimed at deflating the rhetoric of Nature romantics. "Although attempts have been made . . . to interpret territoriality and intimidation as ultimately benign and biotically adaptive," Williams wrote, "I will assume . . . the basically selfish nature of such behaviour is accepted by most biologists" (Williams 1966, 194).

Evolutionary game theory is today regarded by many as a major breakthrough in biology, especially regarding the evolution of cooperation. Mathematical game theory is the study of "the ways in which *strategic interactions* among *rational players* produce *outcomes* with respect to the *preferences* (or *utilities*) of those players" (Ross 2006; author's italics).

The extension of game theoretic analysis to nonhuman biological players, first attempted in detail by Trivers (1971), provided a means for "thinking about evolution at the phenotypic level when the fitnesses of particular phenotypes depend on their frequencies in the population," such as sex ratios (Maynard Smith 1982, 2).

It is commonly asserted that evolutionary game theory assumes individual organisms normally "act in order to maximize, as best they can, their own self-interest" (Alexander 2003). Maynard Smith's enumeration of the changes to traditional game theory needed to fit it for the evolutionary context suggests otherwise, however. According to Maynard Smith, in evolutionary game theory "the criterion of rationality is replaced by that of population dynamics and stability, and *the criterion of self-interest* by Darwinian fitness" (Maynard Smith 1982, 2; my italics). In other words, fitness is not conceptually equivalent to self-interest but is, rather, a variable in a formula that in a different context is occupied by another concept.

Selfishness does not figure noticeably in *Evolution and the Theory of Games* or at all in the first work on the evolutionarily stable strategy (Maynard Smith and Price 1973). Nevertheless, the game theoretic vocabulary is replete with terms (e.g., defector, deception, cheater, bully, retaliator, limited war, war of attrition) that reflect its origins in human action for maximizing self-interest. Examples of animal contests, the evolution of sex ratios, and territorial behavior also make it easy to think in brutish terms. When Richard Lewontin suggested, apparently before anyone else, that game theory could benefit evolutionary biology, he noted its roots in purposive human behavior required that much of the terminology would have to be "discarded or very carefully redefined" for application in "mechanistic" biology (Lewontin 1961, 384). In *The Selfish Gene* not only was the agentive terminology retained, its teleological flavor was considerably amplified.

There is one context in which selfishness, post-Dawkins, appears to have been put to good use in the genetic context. In molecular biology, *selfish genetic elements* (SGEs; not all are genes) operate within a genome in a particular way: "[T]hey use manipulative strategies to maximize their own transmission relative to competing genome components, often undermining the organism's fitness in the process" (Martin 2009, R129). The best known of these elements are autonomously replicating, transposable segments of DNA called transposons, or mobile DNA. Transposons were the target of the original "selfish DNA hypothesis" simultaneously advanced by Orgel and Crick (1980) and Doolittle and Sapienza (1980). Note that the selfish DNA hypothesis relates not to genes in general but to genetic sequences embedded in genomes that display two properties. First, they arise and spread autonomously by making additional copies of themselves, such as tandem repeats (fairly commonplace) or random insertions in multiple sites. Second, these extra copies make no specific contribution to the organism's phenotype and thereby its survival and/or reproductive success. Such sequences were labeled "selfish" because they exploit the body's resources to copy and insert themselves without making a contribution to the organism's existential needs, or even at the expense of those needs. At a minimum they are free-

riders, at worst pathogenic. In short, they exhibit the central criterion of selfishness in ordinary use: action without regard to, or at the expense of others' needs.

SGEs of various types are ubiquitous in eukaryotic genomes, and are believed to be important factors in eukaryotic evolution (Hurst and Werren 2001). *Segregation distorters* act in meiosis or gametogenesis to increase the likelihood of their transmission above fifty percent, usually by skewing the sex ratio of offspring. *Postsegregation distorters* kill or weaken offspring whose viability doesn't benefit them. The *Medea* locus in the flour beetle *Tribolium castaneum* is a maternal allele that kills progeny that don't carry the sequence. Enzymatic *homing endonucleases* recognize certain sequences of DNA, cut the double strand at that point and insert a copy of the gene that makes the enzyme.

Many inherited microbial symbionts also manipulate host reproduction to facilitate their genetic transfer. The bacterium *Wolbachia*, a pervasive parasite of invertebrates, induces cytoplasmic incompatibility in the eggs of infected females; unless the mating male is also infected viable zygotes won't form. Replication of the nuclear material of mitochondria in metazoans and some plant organelles (i.e., chloroplasts) can also lead to selfish results, in the sense that their replication may harm the cell or the organism of which they are a part (Hurst and Werren 2001). The simple pursuit of an existential imperative is not enough to qualify as selfish; the pursuit must stand in a particular relation to the pursuits of other entities, namely, the whole genome and/or the organism. It is not appropriate to say that replicating liver cells are selfish relative to kidney cells, for example, whereas cancerous liver cells are selfish relative to other liver cells because they hijack nutrients for their own growth and compromise the functioning of the organ (indeed, the entire body) of which they are a part.

The evolution of SGEs is today regarded as virtually inevitable due to the ease with which DNA replicates and the genomic milieu, in which replication is an ongoing necessity (Doolitte and Sapienza 1980). The genomes of eukaryotes, including humans, are chockfull of transposon-induced DNA. Importantly, however, the characteristics of DNA alone are not what make SGEs virtually inevitable but, rather, the *context of the genome* in which the DNA functions—and a particular sort of genome at that. Prokaryotic genomes are remarkably free of SGEs in comparison with eukaryotes. The advent of sexual reproduction—meiotic chromosomal division and gamete fertilization—appears to have kick-started the evolution of the kinds of SGEs known today. Indeed, SGEs are believed to be major contributors to the sex-based "genetic conflict" increasingly regarded as "an inherent feature" of sexual reproductive systems (Hurst and Werren 2001, 605).

In sum, the use of the adjective "selfish" in relation to SGEs makes good sense. The usage conforms to, and the behavior described accords with common understanding of the term. It does work in biological and evolutionary explanations, and its explanatory usefulness has increased with time and new discoveries. However, research into SGEs provides no comfort for those who regard selfishness as the default state of biological entities or believe the trait emerged at an early stage of evolution. On the contrary, the research

suggests that selfish DNA of the kind included under this description required a set of fairly advanced evolutionary circumstances to arise and thrive: the evolution of eukaryotes and meiosis.

Conclusion

The widespread assumption in evolutionary biology that natural selection acts on intrinsically selfish individuals needs rethinking. There are fields in molecular and evolutionary biology in which the concept of selfishness does genuine explanatory labor, but I have argued that these circumstances are relatively limited. While all organisms are intrinsically self-preserving and self-extending, except in unusual circumstances, they are not all intrinsically self-serving, as other evolutionary theorists realized. Dawkins's rhetorically potent conflation of selfishness with competitiveness in the Darwinian struggle for existence simultaneously erased the distinction between self-preserving and self-serving behavior, and obscured a major transition in the history of life on this planet, for no tangible explanatory returns. How did a self-organizing, autocatalytic chemical system come to persist in such a way that it could be described as self-preserving, to say nothing of selfish? We do not know. Moreover, we do not appear to be overly concerned that we do not know. The answer cannot be, *it just did*. That this question lies at the juncture where, historically, vital essences and divine intervention have entered the origin-of-life narrative is cause for caution but not for fear. The problem of self-preservation is imposing but scientifically surmountable, or so I believe. It cannot be resolved, however, until it emerges from behind the curtain of selfishness.

Acknowledgments

My thanks to my colleague Jon Opie for invaluable discussion, especially about chemical systems; to Kim Sterelny and Brett Calcott, for finding the kernel in the rhetorical dross and forcing me to make this a better piece of work than it otherwise would have been; and to the Australian Research Council, for funding the work (DP0880559).

Notes

1. The following account is extracted from the unexceptional portions (usually the first paragraph) of the Wikipedia entries relating to each of the italicized terms. The compilation, for better or worse, is my own. None of the unreferenced statements in this and the next two paragraphs should be controversial, however.

2. One could dismiss these as the conclusions of a disappointed old man departing a field he pioneered, the shining promise of which did not come even close to being fulfilled. However, Orgel remained optimistic about the future of prebiotic chemistry, and indeed the RNA World hypothesis—just not as nature's first attempt at an informational molecule.

3. An exception to Hamilton's careful usage appears at the end of (Hamilton 1964b), where he describes the huddling of brooding penguins as "selfish."

References

Abel, D. L., and J. T. Trevors. (2006). Self-organization vs. self-ordering events in life-origin models. *Physics of Life Reviews* 3: 211–228.

Alexander, J. M. (2003). Evolutionary game theory. *Stanford Encyclopedia of Philosophy* Last accessed Feb. 23, 2009 at <http://plato.stanford.edu/entries/game-evolutionary>.

Corning, P. (2005). *Holistic Darwinism: Synergy, Cybernetics, and the Bioeconomics of Evolution*. Chicago: University of Chicago Press.

Crick, F. H. C. (1968). The origin of the genetic code, *Journal of Molecular Biology* 38: 367–379.

Darwin, C. (1996). *The Origin of Species* (Oxford World's Classics). Oxford: Oxford University Press.

Dawkins, R. (1989). *The Selfish Gene* (new ed.). Oxford: Oxford University Press.

Doolittle, W. F., and C. Sapienza. (1980). Selfish genes, the phenotype paradigm and genome evolution. *Nature* 284: 601–603.

Ghiselin, M. T. (1974). *The Economy of Nature and the Evolution of Sex*. Berkeley: University of California Press.

Gilbert, W. (1986). The RNA World, *Nature* 319: 618.

Godfrey-Smith, P. (2009). *Darwinian Populations and Natural Selection*. Oxford: Oxford University Press.

Gonzalez-Pastor, J. E., E. C. Hobbs, and R. Losick. (2003). Cannibalism by sporulating bacteria. *Science* 301: 510–513.

Grafen, A., and M. Ridley. (2007). *Richard Dawkins: How a Scientist Changed the Way We Think*. New York: Oxford University Press.

Hamilton, W. D. (1963). The evolution of altruistic behaviour. *The American Naturalist* 97: 354–356.

Hamilton, W. D. (1964a). The genetical evolution of social behaviour, I. *Journal of Theoretical Biology* 7: 1–16.

Hamilton, W. D. (1964b). The genetical evolution of social behaviour, II. *Journal of Theoretical Biology* 7: 17–52.

Hamilton, W. D. (1970). Selfish and spiteful behaviour in an evolutionary model. *Nature* 228: 1218–1220.

Hamilton, W. D. (1975). Gamblers since life began: Barnacles, aphids, elms. *Quarterly Review of Biology* 50: 175–180.

Hoyle, F., and N. C. Wickramasinghe. (2000). *Astronomical Origins of Life: Steps Toward Panspermia*. Berlin: Springer.

Hurst, G. D. D., and J. H. Werren. (2001). The role of selfish genetic elements in eukaryotic evolution. *Nature Reviews Genetics* 2: 597–606.

Johnston, W. K., P. J. Unrau, M. S. Lawrence, M. E. Glasner, and D. P. Bartel. (2004). RNA-catayzed RNA polymerization: Accurate and general RNA-templated primer extension. *Science* 292: 1319–1325.

Joyce, G. F. (2007). Obituary: Leslie Orgel (1927–2007). *Nature* 450: 627.

Kauffman, S. (2000). *Investigations*. New York: Oxford University Press.

Killingback, T., M. Doebeli, and N. Knowlton. (1999). Variable investment, the continuous prisoner's dilemma, and the origin of cooperation. *Proceedings of the Royal Society of London, Series B* 266: 1723–1728.

Koonin, E. V. (2007). The Biological Big Bang model for the major transitions in evolution. *Biology Direct* 2: 21.

Koonin, E. V., T. G. Senkevich, and V. V. Dolja. (2006). The ancient Virus World and evolution of cells. *Biology Direct* 1: 29.

Leigh, E. G., Jr. (1999). Levels of selection, potential conflicts, and their resolution: The role of the "common good." In L. Keller (ed.), *Levels of Selection*. Princeton: Princeton University Press.

Lehmann, L., L. Keller, S. West, and D. Roze. (2007). Group selection and kin selection: Two concepts but one process. *PNAS* 104: 6736–6739.

Lewontin, R. C. (1961). Evolution and the theory of games. *Journal of Theoretical Biology* 1: 382–403.

Lewontin, R. C. (1977). Caricature of Darwinism. *Nature* 266: 283–284.

Mahner, M., and M. Bunge. (2001). Function and functionalism: A synthetic perspective, *Philosophy of Science* 68: 75–94.

Martin, O. Y. (2009). Sexual selection: Selfish genetic element encourages polyandry. *Current Biology* 19: R129–R131.

Maynard Smith, J. (1982). *Evolution and the Theory of Games*. Cambridge: Cambridge University Press.

Maynard Smith, J., and G. R. Price. (1973). The logic of animal conflict. *Nature* 246: 15–18.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution*. Oxford: W. H. Freeman.

McShea, D. W. (1991). Complexity and evolution: What everybody knows. *Biology and Philosophy* 6: 303–324.

Michod, R. E., and D. Roze. (1997). Transitions in individuality. *Proceedings of the Royal Society of London B* 264: 853–857.

Msadek, T. (1999). When the going gets tough: Survival strategies and environmental signalling networks in *Bacillus subtilis*. *Trends in Microbiology* 7: 201–207.

Nelson, K. E., M. Levy, and S. L. Miller. (2000). Peptide nucleic acids rather than RNA may have been the first genetic molecule. *PNAS* 97: 3868–3871.

Orgel, L. E. (1968). Evolution of the genetic apparatus. *Journal of Molecular Biology* 38: 381–393.

Orgel, L. E. (1992). Molecular replication. *Nature* 358: 203–209.

Orgel, L. E. (2003). Some consequences of the RNA World hypothesis. *Origins of Life and Evolution of the Biosphere* 33: 211–218.

Orgel, L. E. (2004). Prebiotic chemistry and the origin of the RNA World. *Critical Reviews in Biochemistry and Molecular Biology* 39: 99–123.

Orgel, L. E., and F. H. C. Crick. (1980). Selfish DNA: The ultimate parasite. *Nature* 284: 604–607.

Queller, D. C. (1997). Cooperators since life began: Review of *The Major Transitions in Evolution*. *The Quarterly Review of Biology* 72: 184–188.

Queller, D. C. (2004). Kinship is relative. *Nature* 430: 975–976.

Rose, H., and S. Rose (eds.) (2000). *Alas, Poor Darwin: Arguments Against Evolutionary Psychology*. London: Jonathan Cape.

Rosen, R. (1985). *Anticipatory Systems: Philosophical, Mathematical, and Methodological Foundations*. Oxford and New York: Pergamon Press.

Rosen, R. (2000). *Essays on Life Itself*. New York: Columbia University Press.

Rosenblueth, A., N. Wiener, and J. Bigelow. (1943). Behavior, purpose, and teleology. *Philosophy of Science* 10: 18–24.

Ross, D. (2006). Game theory in *Stanford Encyclopedia of Philosophy* Last accessed March 2, 2009 at <http://plato.stanford.edu/entries/Game-theory>.

Sachs, J. L., U. G. Mueller, T. P. Wilcox, and J. J. Bull. (2004). The evolution of cooperation. *The Quarterly Review of Biology* 79: 135–160.

Segré, D., D. Ben-Eli, D. W. Deamer, and D. Lancet. (2001). The Lipid World. *Origins of Life and Evolution of Biospheres* 31: 119–145.

Sober, E., and D. S. Wilson. (1998). *Unto Others: The Evolution and Psychology of Unselfish Behavior*. Cambridge, MA: Harvard University Press.

Sterelny, K. (2001). *Dawkins vs. Gould: Survival of the Fittest*. Cambridge: Icon.

Szathmáry, E., and J. Maynard Smith. (1997). From replicators to reproducers: The first major transitions leading to life. *Journal of Theoretical Biology* 187: 555–571.

Trevors, J. T., and D. L. Abel. (2004). Chance and necessity do not explain the origin of life. *Cell Biology International* 28: 729–739.

Trivers, R. L. (1971). The evolution of reciprocal altruism. *Quarterly Review of Biology* 46: 35–57.

Von Bertalanffy, L. (1968). *General System Theory: Foundations Development Applications*. Harmondsworth, England: Penguin Books.

Wächtershäuser, G. (1988). Before enzymes and templates: Theory of surface metabolism. *Microbiological Reviews* 52: 452–484.

Williams, G. C. (1966). *Adaptation and Natural Selection: A Critique of Some Current Evolutionary Thought*. Princeton, NJ: Princeton University Press.

Williams, G. C. (1988). Huxley's *Evolution and Ethics* in sociobiological perspective. *Zygon* 23: 383–407.

Williams, G. C., and D. C. Williams. (1957). Natural selection of individually harmful social adaptations among sibs with special reference to social insects. *Evolution* 11: 32–39.

Woese, C. R. (1967). *The Genetic Code: The Molecular Basis for Genetic Expression*. New York: Harper & Row.

Woese, C. R. (1998). The universal ancestor. *PNAS* 95: 6854–6859.

Woese, C. R. (2002). On the evolution of cells. *PNAS* 99: 8742–8747.

7 The Evolution of Restraint in Structured Populations: Setting the Stage for an Egalitarian Major Transition

Benjamin Kerr and Joshua Nahum

Chance fights ever on the side of the prudent.
—Euripedes

The prolific reef-building capacity of hermatypic corals depends on their association with single-celled photosynthetic endosymbionts called zooxanthellae (Knowlton 2001). One might think of the reef itself as a magnificent signature of an interspecific union. A "major transition" in evolution has transpired: Formerly independent entities have come to rely on one another as a "higher-level" entity for continued existence. If we dig a bit deeper evolutionarily, the coral contains a layered series of such transitions. There is evidence that a nonphotosynthetic ancestor of zooxanthellae engulfed a red algal cell, which eventually generated its photosynthetic plastid (Bhattacharya et al. 2007; Keeling 2004). And this was not the first of such maneuvers: A nonphotosynthetic ancestor to the red algae engulfed a cyanobacterium, eventually giving rise to the algal chloroplast. Furthermore, the mitochondria in both coral cells and zooxanthellae come from a proteobacterium that was engulfed by an ancestor common to these present-day symbionts (Emelyanov 2003). Thus, the coral is an elaborate story of serial and parallel symbiotic transitions.

Why would separate entities sacrifice their autonomy and live together so intimately? A popular answer invokes an increased reproductive efficiency of the higher-level unit when lower-level units are able to focus on different tasks (e.g., Maynard Smith and Szathmáry 1995). For instance, under the hydrogen hypothesis for the origin of the eukaryotic cell, a hydrogen-dependent autotrophic host cell engulfed a hydrogen-producing heterotrophic bacterium (Martin and Muller 1998). These authors suggest that what started as an exchange of nutrients evolved into metabolic specialization, with the host performing nutrient acquisition and the proto-mitochondrion focusing on energy production.

Two important questions arise when considering division of labor:

1. How do the differentiated roles of lower-level entities originate?
2. How does the higher level persist given role defection at the lower level?

The first question concerns the origin of a differentiated system and the second question concerns its maintenance in the face of lower-level selfishness. Transitions to higher-level

units often depend on both the differentiation and self-restraint of lower-level units. Thus, an understanding of major transitions requires answers to the preceding questions.

In searching for answers, we make use of a dichotomy introduced by Queller (1997, 2000). He separated major transitions based on whether the higher-level entity was formed through the aggregation of different lower-level entities ("egalitarian" transitions) or through the "sticking together" of related lower-level entities ("fraternal" transitions). For instance, fraternal transitions were instrumental to the origins of multicellularity and eusociality in animals. Egalitarian transitions include the compartmentalization of different molecules in protocells, linking of different genes on a chromosome, and the endosymbiotic origin of mitochondria within the eukaryotic cell. While both fraternal and egalitarian transitions share important features, there are important differences between the two transitions regarding questions 1 and 2.

In figure 7.1, we illustrate two hypothetical sequences leading to a differentiated higher-level unit. In both cases, dotted lines are drawn around "individuals." Reproduction is central to the first path. Before the transition occurs, an entity gives rise to two daughter entities through division (white arrows). The transition occurs when these daughters re-

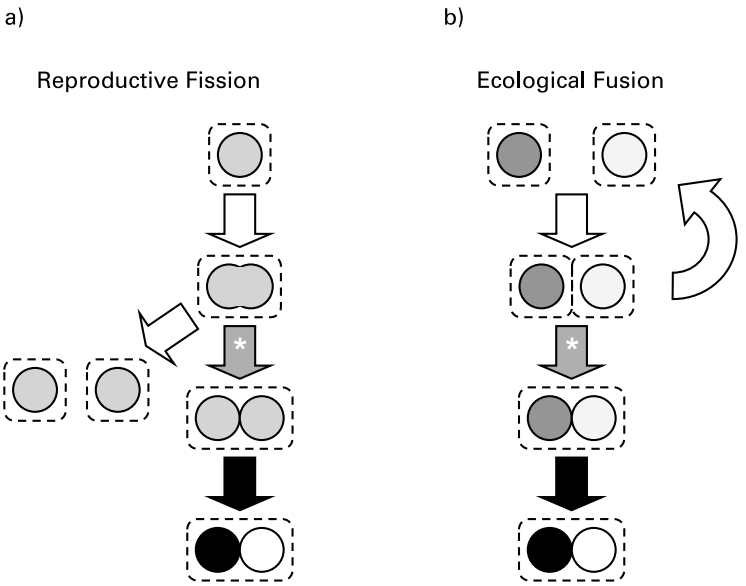


Figure 7.1
Two pathways to a major transition. (a) A type of fraternal transition termed "reproductive fission." Before the transition, an individual reproduces through division (white arrows). At some point (the arrow marked with an asterisk) the daughters of this division remain physically associated. Differentiation of these daughters occurs next (black arrow). (b) A type of egalitarian transition termed "ecological fusion." Before the transition, two individuals interact ecologically but maintain separate identities (white arrows). At some point (the arrow marked with an asterisk) these individuals become intimately (and physically) associated. Further evolution of the former individuals occurs next (black arrow).

main together (gray arrow with asterisk) and differentiate (black arrow). The second path coopts ecological interaction. Before the transition occurs, one entity interacts with another distinct entity (e.g., through predation, parasitism, competition). As these ecological interactions occur, the entities exist as separate individuals (white arrows). The transition occurs when these entities associate intimately (gray arrow with an asterisk) and evolve further (black arrow). We call the first path "reproductive fission" and the second path "ecological fusion." These paths are types of fraternal and egalitarian transitions, respectively.

Returning to the two questions, we see important differences between these paths. For ecological fusion, differentiation is already present before the transition. Though the roles of lower-level entities will presumably change further over evolutionary time, we note that the higher level starts with the raw ingredients for division of labor. In essence, some work has already been done in answering question 1. In contrast, the reproductive fission path starts with identical lower-level units that need to differentiate. In a sense, we start from scratch in answering question 1 (although presumably the answer will involve epigenetic plasticity to internal and external environmental factors).

Regarding question 2, reproductive fission immediately addresses defection. Because the lower-level units are relatives, cooperative action is explained by kin selection theory (Hamilton 1964; Maynard Smith 1964). For ecological fusion, question 2 requires some thought: How is it that independent entities with their own interests come to cooperate, and why don't they take advantage of one another once in association?

A case can be made that ecological fusion is simply a form of mutual exploitation between lower-level units that produces a functional higher-level unit. In the same vein, Bronstein (2001) discusses ecological mutualisms as "reciprocally exploitative interactions that provide net benefits to both partner species." In some symbioses, increased exploitation of one member by another may be selectively advantageous, and the higher-level unit could cease to function. Similarly, defectors in mutualisms have been reported that threaten the cooperative partnership (see Pellmyr, Leebens-Mack, and Huth 1996 for an example involving yucca moths). Given such possibilities, what selects for the restraint necessary to ensure higher-level functioning?

In this chapter, we address the evolution of restraint in simple ecosystems. When certain types of ecological interactions occur in spatially structured habitats, a form of restraint evolves. We present both simulation and experimental data on the evolution of restraint. These results are meant to address question 2 for the case of ecological fusion. Consequently, we argue that spatial structure may play an important role in setting the stage for egalitarian major transitions.

The "Rock-Paper-Scissors" Game and the Evolution of Restraint

In this chapter, we focus on the evolution of restraint in *nontransitive* ecological communities. A nontransitive community contains members whose ecological interactions violate

mathematical transitivity. One simple example involves three competitors engaged in a game of rock-paper-scissors. In this children's game, rock loses to paper, paper loses to scissors, and (in a violation of transitivity) scissors loses to rock. This dynamic is broadly distributed phylogenetically—the game has been described in bacteria (Kerr et al. 2002; Kirkup and Riley 2004), fungi (Paquin and Adams 1983), plants (Lankau and Strauss 2007), and animals (Buss and Jackson 1979; Sinervo and Lively 1996). For a concrete example, we will focus on a nontransitive bacterial system involving toxin production.

Nearly every major bacterial lineage contains strains that produce narrow-spectrum toxins, or *bacteriocins*, active against related sensitive bacterial strains (Riley and Wertz 2002a, 2002b). Bacteriocin production is often interpreted as an anticompetitor strategy (Chao and Levin 1981; Riley 1998; Riley and Gordon 1999), as the killing of toxin-sensitive strains opens up space and resources for toxin-immune producer strains. As an interesting wrinkle to the story, the producing cell *kills itself* as it releases the toxin in some cases. Thus, it is the neighboring (quiescent) clones of the active producer that benefit from its altruistic suicide.

Sensitive strains can evolve resistance, which adds a third player to the community. Resistance is different from the immunity found in producer cells. Immunity involves constitutive production of an immunity protein that binds and neutralizes the toxin, whereas resistance often involves alteration or loss of a membrane protein that binds or translocates the toxin (Feldgarden and Riley 1998; James, Kleanthous, and Moore 1996; Riley and Gordon 1999). Because these membrane proteins are also involved in other cell functions such as nutrient uptake, resistance often involves a concurrent growth cost. In certain cases, this cost of resistance is less than the cost of bacteriocin production, which includes the cost of immunity and the small probability of *lethal* toxin release. In these cases, the sensitive strain outgrows the resistant strain, the resistant strain outgrows the producer, and the producer kills the sensitive strain. Thus, we have a microbial game of rock-paper-scissors.

This nontransitive dynamic was demonstrated in vitro (Kerr et al., 2002) and in vivo (Kirkup and Riley, 2004) with strains of *Escherichia coli* that produce bacteriocin E2. Kerr and associates (2002) demonstrated that spatial structure was critical to the maintenance of all three strains. When these strains formed a patchwork on the surface of a Petri dish, all persisted as part of a "fluid mosaic"—a patch of each strain chased a patch of one other strain and, in turn, was chased by the third strain. However, in a well-mixed environment (e.g., in a shaken flask), the sensitive strain was immediately destroyed by the circulating toxin and then the resistant strain eventually replaced the producer.

In figure 7.2, photographs of the Petri dishes in the spatially structured treatment show the boundaries between patches of the different strains changing over time. The producer patches are fainter due to a lower density, whereas both the resistant and sensitive patches are thicker due to a higher density. As a patch of producer cells advances into territory previously claimed by sensitive cells, small pockets of de novo resistant cells are left be-

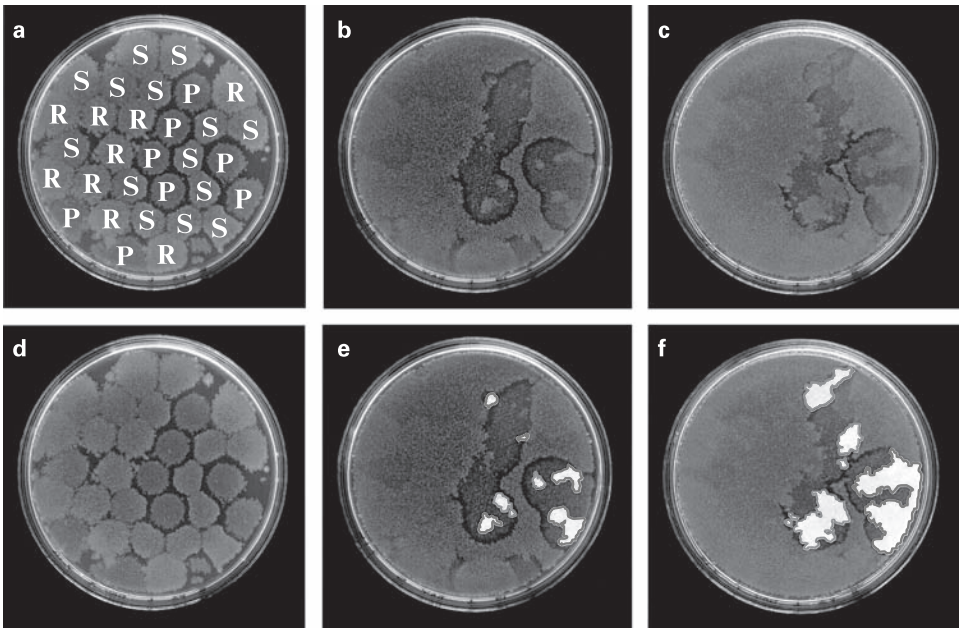


Figure 7.2
A biofilm rock-paper-scissors community on the surface of a Petri dish. The community is initialized by placing droplets of pure cultures of the producer (P), sensitive strain (S), and resistant strain (R) randomly in a hexagonal lattice pattern. The community is shown after three days (a), five days (b) and seven days (c), where replica plating transfers took place daily. Because producers are poor growers, they form patches of lower density, which allows boundaries between producer patches and patches of the other strains to be followed over time. Careful inspection of these boundaries reveals that P chases S and R chases P. In the bottom panel, we show the same three time points but have highlighted de novo resistant patches that arise within the producer patches as they move into areas formerly occupied by sensitive patches. These de novo resistant patches spread over time (i.e., from e to f). (Reproduced from Kerr et al., 2002, *Nature* 418: 171–174.)

hind. These cells eventually form patches themselves, swelling inside the producer patches over time (emphasized in gray in figure 7.2e and f). It has been demonstrated that the cost of resistance can vary widely from strain to strain (Feldgarden and Riley 1998, 1999). Also, resistant strains can ameliorate the cost of resistance through secondary "compensatory" mutations. Given the observation of de novo resistant cells in the experiment and the potential for compensation, one might ponder the long-term evolutionary trajectory for the cost of resistance. A reasonable forecast would be that resistant cells evolve to *minimize* the cost of resistance.

When the resistant strain is permitted to evolve in computer simulations of the full community (by allowing the cost of resistance to mutate), this strain does *not* minimize the cost of resistance in a spatially structured habitat (figure 7.3). Control runs in which the resistant strain evolves alone or with the other strains in a well-mixed habitat demonstrate that the resistant strain *can* evolve to minimize its growth cost (Kerr, 2007; Prado and Kerr, 2008).

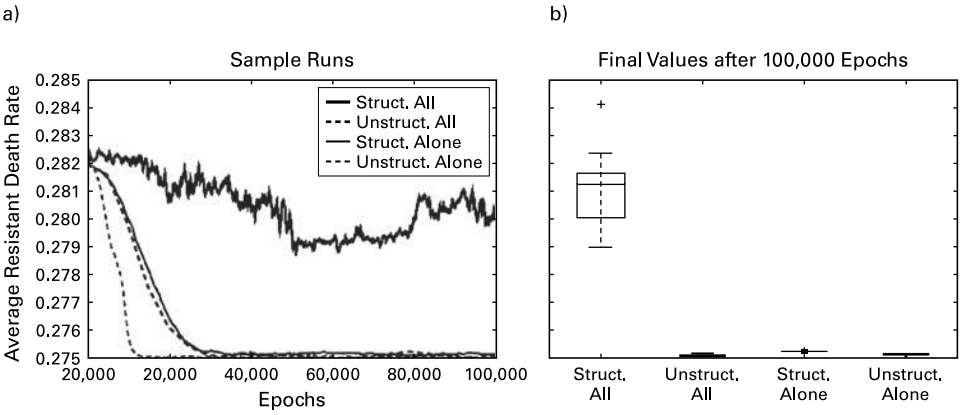


Figure 7.3
The evolution of restraint. We show the results of lattice-based simulations in which the resistant strain can evolve its death rate (which measures the cost of resistance). (a) Sample trajectories of resistant death rate. (An epoch is a unit of time equal to the average updating turnover of the lattice). When the resistant strain evolves alone (in a structured or unstructured habitat), its death rate evolves to the minimum allowed value. When the resistant strain evolves in an unstructured (i.e., well-mixed) habitat with the other two strains (producer and sensitive: the "full" community), its death rate again evolves to the minimum value. Note that, in this unstructured simulation with all community members, both producers and sensitive cells had to be continually "reseeded" because an unstructured community does not maintain diversity (see Prado and Kerr 2008 for details). In a structured community with all three strains, the resistant strain does *not* evolve to minimize its death rate. That is, the resistant strain evolves restraint in its growth rate. (b) The final average death rate from ten simulations for each community type. Horizontal lines in the boxes represent the upper, median, and lower quartile values, and vertical lines extending from each box cover all data points within 1.5 units of the interquartile range beyond the box. Outliers appear as plus signs beyond the vertical lines. (Reproduced from Prado and Kerr, 2008, *Evolution* 62: 538–548.)

Why does this strain not maximize its growth rate when competing with the other two members of this nontransitive community in a structured habitat?

Within a patch of resistant cells, any mutant that lowers its cost of resistance has an immediate selective advantage. It can outgrow its fellow resistant neighbors and more quickly invade a neighboring patch of producer cells. However, myopic gain is not synonymous with long-term gain in this community. The idea is captured well by the adage "the enemy of my enemy is my friend." By replacing your victim faster, you are more likely to come head to head with your enemy (the victim of your victim). Thus, a patch filled with faster-growing resistant cells is more likely to end up surrounded by sensitive cells (as it more quickly burns through its neighboring producer patches; see figure 7.4). Patches of resistant cells filled with slower growers end up surviving longer. By backing off the enemy of their enemy, such restrained cells ensure a longer tenure for their lineage. Consequently, the population of resistant cells remains restrained. Interestingly, restraint is also predicted if toxicity evolves. In simulations of a structured nontransitive community, producers do not evolve to maximize their toxicity (Prado and Kerr, 2008). In both cases, negative feedback from short-term adaptation favors restraint in this nontransitive structured community in the long term.

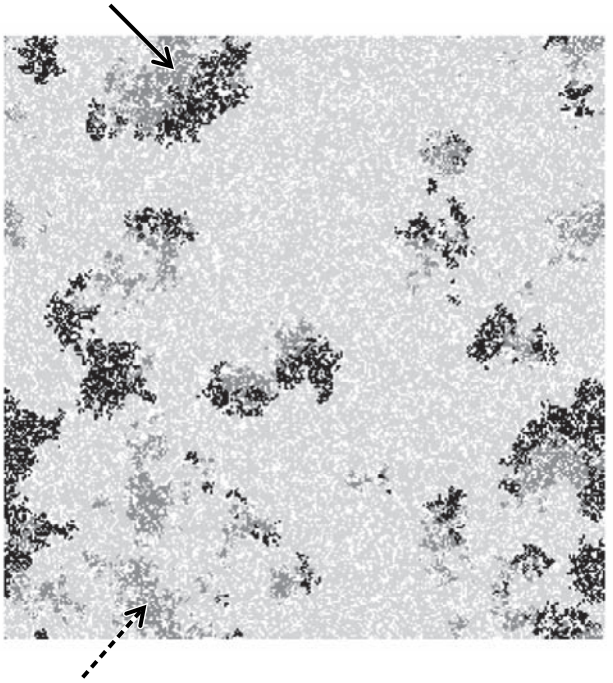


Figure 7.4
Snapshot of the lattice with all three strains in a structured community. The sensitive strain is light gray, the producer is black, and the resistant strain is dark gray (empty lattice points are white). Strains form clusters that chase one another around the lattice. Patches with restrained resistant cells tend to keep a "buffer" of producer cells between themselves and the sensitive cells (e.g., solid arrow). Patches with unrestrained resistant cells tend to "burn through" this buffer and end up surrounded by sensitive cells (e.g., dashed arrow). (Reproduced from Prado and Kerr, 2008, *Evolution* 62: 538–548.)

The "Tragedy of the Commons" and the Evolution of Restraint

The evolution of restraint described in the last section is predicted from simulations of virtual competitors. In this section, we discuss empirical results demonstrating that another combination of spatial structure and nontransitivity can favor the evolution of restraint. We shift focus from interacting individuals within a population to interacting subpopulations within a metapopulation. Consider a victim-exploiter community (prey-predator, host-pathogen, plant-herbivore, etc.) with the following two assumptions: (i) the exploiter population exhausts its victim population over a finite amount of time, and (ii) the victim population can sustain itself indefinitely in the absence of the exploiter. In a single population with both victims and exploiters, these assumptions would ensure extinction of the victim followed by extinction of the exploiter. However, in a metapopulation, both victim and exploiter can be maintained. Migration between subpopulations continually moves victims to sites lacking exploiters. Similarly, migration moves the exploiter from

decimated subpopulations to subpopulations with previously unexploited victims. Unoccupied sites for victims to colonize are generated as the exploiter goes extinct locally, following decimation of its victim. These transitions (colonization, exploitation, and extinction) occur simultaneously in different subpopulations. Thus, asynchrony within the metapopulation allows both members of this community to persist.

This persistence depends on a form of nontransitivity present in this system. We have three types of subpopulations: unoccupied subpopulations (U), subpopulations with victims only (V), and subpopulations with exploiters (E). Migration of victims from a V subpopulation into a U subpopulation transforms the latter into a V subpopulation (V beats U). Migration of exploiters from an E subpopulation into a V subpopulation transforms the latter into an E subpopulation (E beats V). Finally, without migration, E subpopulations transform into U subpopulations because exploiters go extinct after exhausting their local supply of victims (in a sense, U beats E). These transitions are similar to a game of rock-paper-scissors.

Kerr and associates (2006) performed real-time evolution experiments with a victim-exploiter metapopulation that satisfied the preceding assumptions. The victim was the bacterium *E. coli*, and the exploiter was a virus, T4 phage, which infects and kills the bacterium figure 7.5a). In the experiment, the phage and bacteria were serially propagated as a metapopulation in multiwell microtiter plates (figure 7.5b). Phage and bacteria did not coexist within a single well over an incubation period (if sufficiently abundant, the phage destroyed the bacterial population). However, migration between the wells allowed both bacteria and phage to coexist. Kerr and associates (2006) experimentally manipulated the form of migration. In one treatment, migration was spatially restricted, occurring only between neighboring wells. In a second treatment, migration was unrestricted, potentially occurring between any two wells in the metapopulation. Thus, the restricted treatment represents a metapopulation with a higher degree of structure.

The authors found that "rapacious" phage evolved in the unrestricted treatment. Rapacious phage had a higher rate of attachment to their host cells and a shorter latent period within their host before cell death (Eshelman et al., 2010). That is, the phage had evolved to become more infective and more virulent. In the restricted treatment, the phage evolved a relatively prudent strategy (lower infectivity and lower virulence). Under the conditions of this experiment, such prudence led to greater productivity of the phage within a subpopulation over an incubation period, whereas rapacity went hand-in-hand with better competitive ability (see figure 7.5c and d). When both phage types are present within a subpopulation, prosocial (prudent) use of host resources is locally disadvantageous. Thus, the advent of rapacity is an instance of the "tragedy of the commons" (Hardin 1968). In this experiment, we see that restricted migration can favor the type of prudence needed to avert the tragedy of the commons.

The reasons for these evolutionary results are similar to those in the case of the bacteriocin community discussed in the previous section. Rapacious phage subpopulations are less

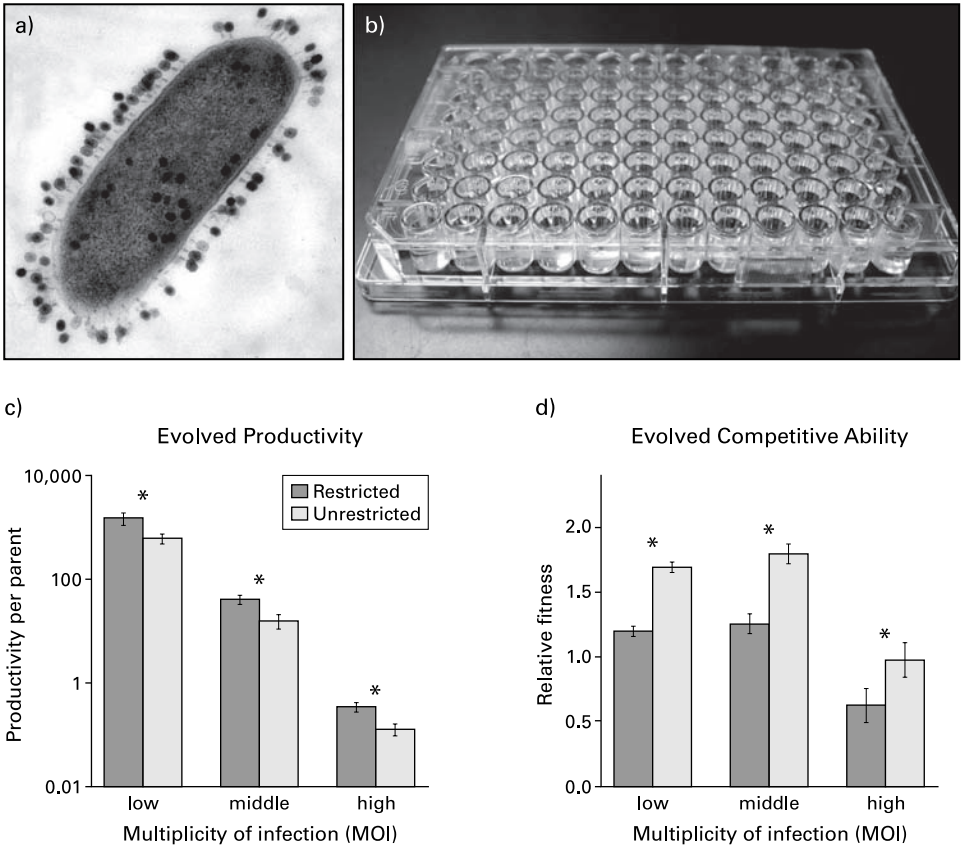


Figure 7.5
An experiment with bacteria and phage. (a) A micrograph of an *E. coli* cell being attacked by several T4 phage particles (courtesy of J. Wertz). (b) The microbial metapopulation. Each well within this microtiter plate is a subpopulation. The pattern of migration between these wells (accomplished by micropipetting from one well to another) was the experimental variable. Phage evolving in metapopulations in which migration was restricted to occur between neighboring wells evolved higher productivity (c) and lower competitive ability (d) compared to phage evolved in metapopulations in which migration was spatially unrestricted. The productivity and competitive ability assays were performed at three different multiplicities of infection (the ratio of phage to bacteria), but the results were the same in every case (asterisks denote statistically significant differences at $P < 0.05$). (Reproduced from Kerr et al., 2006, *Nature* 442: 75–78.)

productive. Because propagation of the metapopulation involves serial dilution to fresh medium, it takes fewer dilutions for a rapacious phage subpopulation to shift to an unoccupied state (an E → U transition). Given restriction to migration, this phage is limited in its access to host cells. Thus, the myopic advantage of rapacity within a subpopulation leads to long-term demise. Here, patches of rapacious E subpopulations are being "chased" faster by U subpopulations. Thus, we see that prudent exploiters, with their longer tenure, are favored in structured metapopulations. This experiment shows that spatial structure can favor the evolution of restraint.

Discussion

In egalitarian transitions, different entities come together to form a new entity. Thus, before the transition, these entities presumably interact ecologically. We note that in many cases this interaction likely has antagonistic components (competition, exploitation, etc.). Somehow, such antagonism is rerouted into a productive partnership. As stated in the introduction, there are two hurdles to jump to accomplish such a major transition. The first concerns a division of labor. Because the different entities may have very different ecological roles, some differentiation is already present from the start. The second hurdle concerns the ever-present threat of defection. In this chapter, we have argued that some ecological communities naturally promote self-restraint. Specifically, a nontransitive network of interaction can favor prudence when ecological interactions are localized.

How common are nontransitive networks in biological systems? Some authors have argued that rock-paper-scissors dynamics are common in natural communities (Sinervo and Calsbeek, 2006). Competitive nontransitivity has been described in side-blotched lizards (Sinervo and Lively 1996), allelopathic plants in the family Brassicaceae (Lankau and Strauss 2007), sessile marine invertebrates (Buss and Jackson 1979), and epiphytes of intertidal alga (Stebbing 1973). In principle, nontransitivity follows from the interaction between the following three types: a harming type, a type sensitive to the harm, and a type resistant to the harm. As long as the cost of harming is more than the cost of resistance, a nontransitivity results. The "harm" in this scenario could take any one of many forms (allelopathy, predation, parasitism, etc.). Thus, the conditions necessary for a nontransitive network may be fairly generic. Sinervo and Calsbeek (2006) suggest that nontransitivity may be important in many ecological contexts, including mutualism, resource competition, altruistic interaction, and Batesian mimicry.

Although one can debate the prevalence of nontransitivity, there is little doubt that most populations are spatially structured (Dieckmann, Law, and Metz 2000; Tilman and Kareiva 1997). Local interactions are especially relevant for species in which one part of the life cycle is sessile (e.g., plants, some marine invertebrates, and some microbes in biofilms). However, even populations of highly mobile organisms possess some degree of spatial structure. Indeed, spatial structure could have been a key ingredient in the origin of life on

this planet as mineral surfaces may have played a critical role in the catalysis of biopolymers in the early RNA world (Ferris, 2006).

Previous theoretical work has foreshadowed some of the main conclusions of this chapter. Johnson and Seinen (2002) explored a generic rock-paper-scissors community in which spatial structure favored the evolution of competitive restraint. However, the evolution of restraint may also occur in structured systems with other types of ecological interactions. One example concerns theoretical work on the evolution of hypercycles. The hypercycle (Eigen and Schuster 1977) is a cyclical network of autocatalytic reactions (e.g., a network of RNA strands, where each catalyzes the replication of the next in a looped chain). Here, there is ecological feedback through the cyclic architecture of the network. One question is why any component of this system should evolve to promote the replication of *other* components in the chain (Maynard Smith 1979). In particular, a hypercycle should be vulnerable to disintegration due to the threat of parasitic components, which receive improved catalytic help from the previous member of the cycle but provide poorer catalytic support to the next member of the cycle. Maynard Smith (1979) suggested that compartmentalization of the hypercycle provides a solution (i.e., a population of competing protocells, each containing the components of a hypercycle). Boerlijst and Hogeweg (1991) demonstrated that hypercycles in spatially structured environments self-organize into rotating spirals, which play a role in preventing the spread of parasites. Thus, a form of restraint in the components of a hypercycle can evolve in spatially structured populations without discrete compartmentalization.

In this chapter, we have illustrated that restraint evolves in structured ecosystems with cyclic networks of interaction. This is predicted to occur in the case of allelopathic interactions between bacterial competitors. Here, the scale of interaction is between individual cells. We have also presented empirical data in which bacteriophage pathogens evolved restraint when patterns of migration were spatially restricted within a metapopulation. Here, the cyclic network occurred between subpopulations of victims and exploiters. Incorporating the theoretical work on hypercycles, we see that restraint can evolve in a number of ecological contexts at a number of scales.

For some major transitions, restraint involves complete reproductive sacrifice (e.g., sterility of somatic tissue in multicellular organisms, or of workers in eusocial insect colonies). However, complete reproductive restraint is likely associated with *fraternal* transitions (Queller 2000). As we discussed in the introduction, fraternal transitions involving the type of "reproductive fission" in figure 7.1a start with a solution to the defector problem, as cooperators are naturally clumped. A worthwhile question (but not the subject of this chapter) is how higher-level units achieve lower-level differentiation through the reproductive fission route. Egalitarian transitions involving the type of "ecological fusion" in figure 7.1b face the inverse of these problems. Specifically, ecological fusion begins with a partial solution to the problem of differentiation (as the ecological players presumably had differentiated preexisting roles). However, there is a residual concern about the evolution of

restraint given ecological fusion of *unrelated* entities. Whereas complete reproductive restraint is not expected, some restraint is absolutely essential to the functioning of the higher-level unit. In this chapter, our central point has been that such restraint may occur naturally for nontransitive networks in a spatially structured habitat.

Throughout the preceding discussion, we have focused on systems in which lower-level units are distributed contiguously in space, as opposed to distribution into discrete compartments. We do not intend this as an argument that boundaries around higher-level units are unimportant. Indeed, boundaries may be essential to defining higher-level units (see Godfrey-Smith 2008 for a discussion of this point in the context of the levels of selection debate). Rather, we simply propose that compartments are not necessary for self-restraint to get off the ground. Such restraint is a form of cooperation and may be critical to setting the stage for an egalitarian transition. If, as Euripedes claims, chance fights ever on the side of the prudent, spatial structure may play a key role in promoting egalitarian transitions. We end with a broad description as to how such a sequence of events might unfold, which is taken from the pioneering work on major transitions:

> [T]he first stages of cooperation between replicators may have originated simply because they were neighbours on a surface: only later were discrete compartments . . . formed, within which cooperation could evolve further. (Maynard Smith and Száthmary 1995)

Acknowledgments

We thank Kim Sterelny and Brett Calcott for the invitation to an extremely fruitful KLI workshop. We thank Puneet Aulakh, Brittany Harding, Silja Heilmann, Brooks Miner, and Sterling Sawaya for helpful comments on previous drafts of this manuscript. BK was supported by NIH grant 07-004309 and NSF grant DEB0717486. This material is also based in part on work supported under a National Science Foundation Graduate Research Fellowship to JN.

References

Bhattacharya, D., J. M. Archibald, A. P. Weber, and A. Reyes-Prieto. (2007). How do endosymbionts become organelles? Understanding early events in plastid evolution. *BioEssays* 29: 1239–1246.

Boerlijst, M. C., and P. Hogeweg. (1991). Spiral wave structure in pre-biotic evolution: Hypercycles stable against parasites. *Physica D* 48: 17–28.

Bronstein, J. L. (2001). The exploitation of mutualisms. *Ecology Letters* 4: 277–287.

Buss, L. W., and J. B. C. Jackson. (1979). Competitive networks: nontransitive competitive relationships in cryptic coral reef environments. *The American Naturalist* 113: 223–234.

Chao, L., and B. R. Levin. (1981). Structured habitats and the evolution of anticompetitor toxins in bacteria. *Proceedings of the National Academy of Sciences USA* 78: 6324–6328.

Dieckmann, U., R. Law, and J. A. J. Metz. (2000). *The Geometry of Ecological Interactions: Simplifying Spatial Complexity*. New York: Cambridge University Press.

Eigen, M., and P. Schuster. (1977). A principle of natural self-organization. *Naturwissenschaften* 64: 541–565.

Emelyanov, V. V. (2003). Mitochondrial connection to the origin of the eukaryotic cell. *European Journal of Biochemistry* 270: 1599–1618.

Eshelman, C. M., R. Vouk, J. L. Stewart, E. Halsne, H. A. Lindsey, S. Schneider, M. Gualu, A. M. Dean, and B. Kerr. (2010). Unrestricted migration favours virulent pathogens in experimental metapopulations: Evolutionary genetics of a rapacious life history. *Philosophical Transactions of the Royal Society B* 365: 2503–2513.

Feldgarden, M., and M. A. Riley. (1998). High levels of colicin resistance in Escherichia coli. *Evolution* 52: 1270–1276.

Feldgarden, M., and M. A. Riley. (1999). The phenotypic and fitness effects of colicin resistance in *Escherichia coli* K-12. *Evolution* 53: 1019–1027.

Ferris, J. P. (2006). Montmorillonite-catalysed formation of RNA oligomers: The possible role of catalysis in the origins of life. *Philosophical Transactions of the Royal Society B, Biological Sciences* 361: 1777–1786.

Godfrey-Smith, P. (2008). Varieties of population structure and the levels of selection. *British Journal for the Philosophy of Science* 59: 25–50.

Hamilton, W. D. (1964). The genetical evolution of social behaviour. I. *Journal of Theoretical Biology* 7: 1–16.

Hardin, G. (1968). The tragedy of the commons. *Science* 162: 1243–1248.

James, R., C. Kleanthous, and G. R. Moore. (1996). The biology of E colicins: paradigms and paradoxes. *Microbiology* 142: 1569–1580.

Johnson, C. R., and I. Seinen. (2002). Selection for restraint in competitive ability in spatial competition systems. *Proceedings of the Royal Society of London. Series B, Biological Sciences* 269: 655–663.

Keeling, P. J. (2004). Diversity and evolutionary history of plastids and their hosts. *American Journal of Botany* 91: 1481–1493.

Kerr, B. (2007). The ecological and evolutionary dynamics of model bacteriocin communities. In M. A. Riley and M. A. Chavan (eds.), *Bacteriocins: Ecology and Evolution* (pp. 111–134). Heidelberg: Springer.

Kerr, B., C. Neuhauser, B. J. M. Bohannan, and A. M. Dean. (2006). Local migration promotes competitive restraint in a host-pathogen "tragedy of the commons." *Nature* 442: 75–78.

Kerr, B., M. A. Riley, M. W. Feldman, and B. J. M. Bohannan. (2002). Local dispersal promotes biodiversity in a real-life game of rock-paper-scissors. *Nature* 418: 171–174.

Kirkup, B. C., and M. A. Riley. (2004). Antibiotic-mediated antagonism leads to a bacterial game of rock-paper-scissors in vivo. *Nature* 428: 412–414.

Knowlton, N. (2001). The future of coral reefs. *Proceedings of the National Academy of Sciences USA* 98: 5419–5425.

Lankau, R. A., and S. Y. Strauss. (2007). Mutual feedbacks maintain both genetic and species diversity in a plant community. *Science* 317: 1561–1563.

Martin, W., and M. Muller. (1998). The hydrogen hypothesis for the first eukaryote. *Nature* 392: 37–41.

Maynard Smith, J. (1964). Group selection and kin selection. *Nature* 201: 1145–1147.

Maynard Smith, J. (1979). Hypercycles and the origin of life. *Nature* 280: 445–446.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution.* Oxford: W. H. Freeman.

Paquin, C. E., and J. Adams. (1983). Relative fitness can decrease in evolving asexual populations of *S. cerevisiae*. *Nature* 306: 368–371.

Pellmyr, O., J. Leebens-Mack, and C. J. Huth. (1996). Non-mutualistic yucca moths and their evolutionary consequences. *Nature* 380: 155–156.

Prado, F., and B. Kerr. (2008). The evolution of restraint in bacterial biofilms under nontransitive competition. *Evolution* 62: 538–548.

Queller, D. C. (1997). Cooperators since life began. *The Quarterly Review of Biology* 72: 184–188.

Queller, D. C. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* 355: 1647–1655.

Riley, M. A. (1998). Molecular mechanisms of bacteriocin evolution. *Annual Review of Genetics* 32: 255–278.

Riley, M. A., and D. M. Gordon. (1999). The ecological role of bacteriocins in bacterial competition. *Trends in Microbiology* 7: 129–133.

Riley, M. A., and J. E. Wertz. (2002a). Bacteriocin diversity: Ecological and evolutionary perspectives. *Biochimie* 84: 357–364.

Riley, M. A., and Wertz, J. E. (2002b). Bacteriocins: Evolution, ecology, and application. *Annual Review of Microbiology* 56: 117–137.

Sinervo, B., and R. Calsbeek. (2006). The developmental, physiological, neural, and genetical causes and consequences of frequency-dependent selection in the wild. *Annual Review of Ecology, Evolution, and Systematics* 37: 581–610.

Sinervo, B., and C. M. Lively. (1996). The rock-paper-scissors game and the evolution of alternative male strategies. *Nature* 380: 240–243.

Stebbing, A. R. (1973). Competition for space between epiphytes of Fucus serratus L. *Journal of the Marine Biological Association of the United Kingdom* 53: 247–261.

Tilman, D., and P. Kareiva. (1997). *Spatial Ecology*: *The Role of Space in Population Dynamics and Interspecific Interactions*. Princeton, NJ: Princeton University Press.

8 Conflicts among Levels of Selection as Fuel for the Evolution of Individuality

Paul B. Rainey and Benjamin Kerr

A fundamental feature of contemporary biological life is its hierarchical organization. Consider just one colony of leaf-cutting ants in a South American forest. This colony is composed of multiple individual ants, differentiated for different tasks. However, each ant is also composed of a set of differentiated cells. And each cell houses a diverse array of genes. Although multiple layers of nested organization characterize many living systems, these hierarchical structures were presumably reduced or absent in our most ancient ancestors. How then do biological hierarchies come to be?

This question motivates a consideration of the "major transitions in evolution," which describe the shift from autonomous lower-level entities to differentiated and integrated higher-level entities (Maynard Smith and Szathmáry 1995). At their core, many major transitions involve a hierarchical shift in *individuality*. Specifically, each transition is characterized by the emergence of individuality at a new level of organization. This occurs as a consequence of subjugation and coordination of lower-level units (Michod 1999).

Evolutionary transitions have been variously categorized (Buss 1987; Jablonka and Lamb 2006; Maynard Smith and Szathmáry 1995). An informative distinction concerns the nature of the alliance among the lower-level entities (Queller 2000). The so-named *egalitarian* transitions are characterized by a fairness in reproduction and mutual dependence: the coming together of disparate entities in a symbiotic association for the benefit of both partners. Examples include the transition from independently replicating nucleic acids to chromosomes, and the transition from prokaryotes to eukaryotes, in which—upon completion—the mitochondrion (once a free-living prokaryote) replicates as part of the host cell.

Unlike the egalitarian transitions, the *fraternal* transitions, such as the transition from single cells to multicellularity, and from multicellular organisms to societies (e.g., eusociality in certain insects), originate with an alliance of entities that at the outset were most likely identical (or highly similar) and where a division of labor arose through epigenesis—a common developmental program expressed differently in different units. Explaining the fraternal transitions poses special challenges because of the need to explain the evolution of the ultimate in self-sacrificial behavior, namely, reproductive altruism: the evolution of entities, such as soma (in a multicellular individual) and sterile workers (in a

eusocial insect colony), that forgo reproduction and serve solely to enhance fitness of the germline. While genetic relatedness (Hamilton 1964a, b) is without doubt a central feature of fraternal transitions (Okasha 2006; Queller 2000)—and conditional sterility a necessary factor (Charlesworth 1980)—the nature of the selective events and mechanistic details underpinning the fraternal transitions remain unclear.

Multicellularity

In this chapter we focus on the evolutionary transition from single cells to multicellular individuals—a transition that has been important for many taxa. The panoply of plant and animal forms owes a great deal to the multicellular foundations of these groups (Conway Morris 1998). From a genetically diverse range of starting positions, independent unicellular lineages have made the transition to multicellularity (Bonner 2000). The most ancient transitions occurred in the major lineages of large multicellular eukaryotes approximately one billion years ago (Wray 2001). However, multicellularity has also arisen in the ciliates, slime molds, diatoms, certain groups of prokaryotes, and, most recently, the volvocine algae (Bonner 1998; Herron and Michod 2008; Kirk 1998). Although certain benefits of multicellularity seem clear, such as the division of labor, the evolutionary causes and mechanistic details underlying this transition remain unknown.

Multicellularity also illustrates the fundamental tension inherent in any major transition, which involves the potential for dissonant interests of entities at different levels in the hierarchy. Although evolutionary transitions involve the exchange of some lower-level autonomy for higher-level functionality, entities at the lower level are not left entirely bereft of individuality (Buss 1987; Michod 1999). From the perspective of multilevel selection theory, natural selection may act simultaneously at different levels within the hierarchy (e.g., genes, cells, multicellular organisms, groups), and selection at one level may oppose selection at another level (Buss 1987; Sober and Wilson 1998). Certain kinds of cancers in vertebrates provide a case in point: Cancer is clearly maladaptive at the level of the organism; nonetheless, natural selection favors individual cells that become cancerous despite the negative consequences for the higher unit of selection (Frank 2007). With respect to major transitions, this conflict between levels is generally seen as a hurdle to be overcome. However, our aim in this chapter is to suggest that this inevitable conflict may sometimes play a productive role in the completion of a major transition (Rainey 2007; Rainey and Kerr 2010). (In discussing plant evolution, Clarke, in chapter 11, also suggests that selection at different levels can play a productive role in the evolution of multicellularity.)

What Needs Explaining?

A useful starting point is to consider what needs explanation. As Okasha (2006, 218) summarizes, "The challenge is to understand . . . transitions in Darwinian terms. Why was it

advantageous for the lower level units to sacrifice their individuality and form themselves into a corporate body? And how could such an arrangement, once first evolved, be evolutionarily stable?" This captures much that is central, but we argue that the essence of the problem resides not so much in why the lower-level units sacrifice their individuality, but in how individuality emerges at the level of the corporate body. In placing the emphasis on individuality at the higher level (Michod 1999), we recognize that individuality is a derived character and one that requires an evolutionary explanation (Buss 1987). The key issue, then, is to explain how variation in lower-level individuals generates a corporate individual with Darwinian characteristics (Dennett 1995).

From a conceptual perspective, a theoretical framework within which to consider transitions in individuality is provided by multilevel selection (MLS) theory (Damuth and Heisler 1988; Heisler and Damuth 1987; Okasha 2006; Sober and Wilson 1998). Here, we focus on the transition to multicellularity. Presumably, the early stages of such a transition involved the formation of simple groups by individual cells. Such groups may have depended on cooperation among individual cells (e.g., the costly production of adhesive polymers that enable cells to stick together after reproduction; see Rainey and Rainey 2003; Velicer and Yu 2003). Provided the group confers some advantage on the constituent cells that offsets the cost of group living, then cooperative cell-level traits that lead to group formation will be favored by selection (Rainey and Rainey 2003).

During this initial stage, the focus is on individual cells and the spread of a trait—in this instance cooperation. The transition to multicellularity, however, is far more than the evolution of cooperation. Critical for the evolution of multicellular organisms is the evolution of group-level adaptations including group reproduction, mechanisms to suppress cheating, and the emergence of development and differentiation. The focus of attention thus shifts from traits that are defined by the properties of individual entities to traits that are the properties of *groups* of cells. This shift marks a significant alteration in perspective and a move to the MLS-2 framework (Damuth and Heisler 1988). However, in MLS-2, group fitness is defined independently of particle fitness. The most successful groups are thus those that contribute the greatest number of group offspring to the next generation, irrespective of the number of cells those groups contain. Thus, fitness is different in MLS-1 versus MLS-2 contexts: In the MLS-1 context, fitness is the number of offspring particles, whereas in MLS-2, the number of offspring collectives defines fitness. Though this makes intuitive—and theoretical—sense it does not amount to an explanation; just how individuality transfers from particles to collectives is a deeply profound problem.

Theoretical studies of Michod and colleagues have made important contributions—particularly the concept of fitness decoupling: the need, during an evolutionary transition, for fitness at the higher level to become decoupled from the fitness of lower level entities (Michod and Nedelcu 2003). This is an important insight, but the mechanism by which it comes about is unclear. For example, Michod (1999) uses a simple model for the evolution of multicellularity that begins with "adult" organisms composed of two cell types

(cooperate and defect). Although the adult organisms are capable of producing offspring propagules, the production of propagules is not a consequence of adult functionality, but rather, depends on the average fitness of the individual entities of which each adult is composed. As Okasha (2006) remarks, this is "a sort of grey area between MLS-1 and MLS-2." Gradually, as the transition proceeds, fitness becomes decoupled from the lower level and, with this, individuality emerges at the level of the adult, to the point where the capacity to leave offspring propagules is a product of adult functionality and independent of the reproductive properties of the individual cells. While such a scenario describes plausible changes in selection pressures during a transition, the model assumes that the capacity to leave group offspring is already in place. But how such a new level of reproduction emerges requires explanation.

The Emergence of Group Reproduction

From a theoretical perspective, the shift from MLS-1 to MLS-2 captures the sense of an evolutionary transition in individuality. The transition completes when the higher-level entities become Darwinian individuals, that is, when populations of these organisms display variation, heritability, and reproduction. Thus, one critical trait that marks individuality at the higher level is the capacity for groups to leave offspring groups. This capacity is precisely what MLS-2 group fitness measures and is *not* the focus when considering MLS-1 groups. Thus, the MLS-2 context underlines one central problem in the evolution of higher-level individuality—the problem of the evolution of collective reproduction—and it is this that we feel most pressingly requires an evolutionary explanation. Here is where things become difficult.

Reproduction of collectives requires development and a life cycle such that offspring contain parts that were previously part of their parents. This is not something that newly formed groups are necessarily born with (Griesemer 2000). When considering the evolutionary origins of such a capability—particularly via natural selection—problems arise. The evolution of traits adaptive at a given level of biological organization requires the existence—at that level—of the necessary prerequisites for Darwinian individuality. When the trait whose origin we wish to explain is reproduction, we face a dilemma: Appeals to natural selection would seem to presuppose the existence of collective reproduction—the very trait whose evolution requires explanation. Griesemer foresaw precisely this problem when he argued that explaining the emergence of a new level of organization is necessary before invoking the evolution of adaptations specific to that new level (Griesemer 2000).

It is interesting to pause at this point. If the MLS framework is the appropriate context within which to consider evolutionary transitions (we believe that it is) and if the paradox is real, then there is a suggestion that the evolution of higher-level individuality is, in some way, tied to a property of individual cells. We return to this notion later, but at this stage we draw attention to the fact that in multicellular organisms, individuality is ultimately a prop-

erty of individual cells that, as a consequence of developmental control, give rise to a multicellular organism each generation.

The Inadequacy of Viability Selection

The absence of a means of collective reproduction does not mean that selection cannot act on collectives, but its capacity to do so is limited to selection at the level of collective *viability*. Provided that simple undifferentiated groups can evolve repeatedly from the ancestral state (which is readily envisaged), selection will favor the most viable groups (figure 8.1a). Although such groups are seen by selection, the connection between the consequences of selection at the level of groups at one point in time and the properties of groups at a latter point in time is lacking. The only connection is through the lower-level entities. It is difficult to see how viability selection alone could result in the evolution of true group-level traits such as the capacity for group reproduction, let alone, self-policing, development, and differentiation. We do, nonetheless, consider such a possibility in the penultimate section.

Imagine, however, that the viability process operates in tandem with a process by which groups are created from the lower-level parts of preexisting groups (see figure 8.1b). For selection to work creatively—and potently—on the higher level, it is crucial for groups to beget groups. But this returns us to the paradoxical situation described earlier: namely, that the capacity of groups to beget groups requires groups to have evolved this capacity.

Insights from Experiments

It is the goal of all evolutionists to understand the origins of the behaviors, morphologies, genetics, and so forth of the organisms that interest them. For much of the time, studies are indirect, relying on historical, theoretical, or comparative approaches to draw inferences regarding the evolutionary events that occurred in the distant past.

Some insight into the causes of multicellularity would emerge were it possible to explore the evolution of individuality in real time. Although seemingly fanciful, experimental bacterial populations do provide some opportunities. But more fundamentally, by starting with a population of organisms that lack all semblance of higher levels of organization, the experimenter is forced to confront issues that can be overlooked when the focus of investigation is an organism that has already made the transition to a higher level of organization—no matter how simple that transition may appear today.

The De Novo Evolution of Simple Undifferentiated Groups

Our own work uses experimental populations of the bacterium *Pseudomonas fluorescens*. When propagated in a spatially structured environment (a simple glass tube containing a

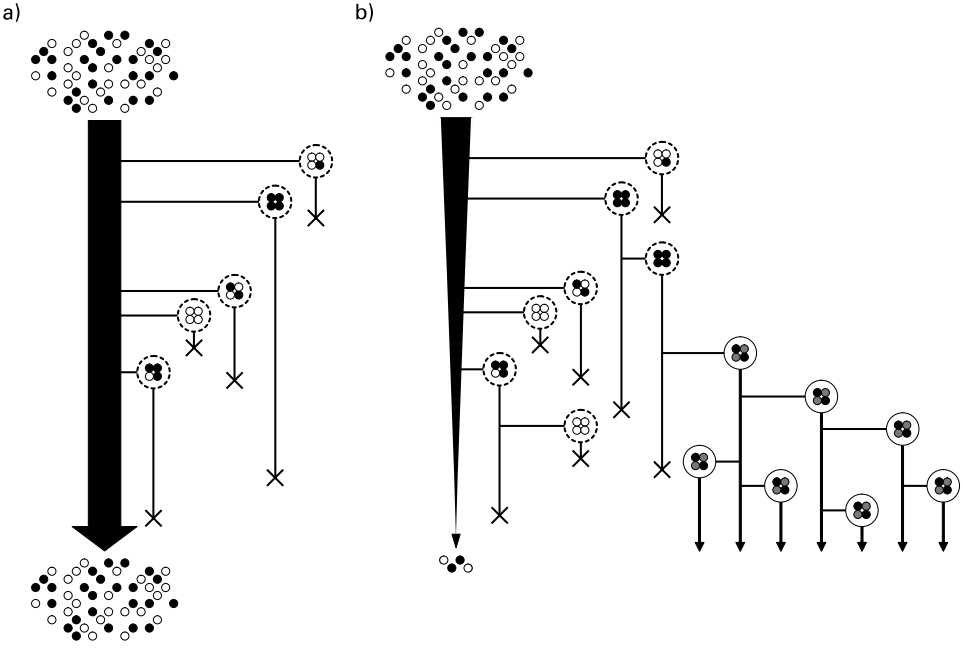


Figure 8.1
The role of group reproduction in group adaptation. (a) A scenario is shown in which loose groups form from individual cells (black and white circles). These groups do not beget new groups, nor do they contribute individual cells back to the cell population. Natural selection can certainly act on these groups. For example, in the picture, groups with more black cells live longer, and therefore the frequency of black cells within groups remains high (this occurs even though the black cells are at a frequency equal to the white cells within the "free cell" population). However, there is no way for evolutionary innovations at the group level to propagate through this form of group viability selection (given finite group lifetimes). For example, it is not the case that groups with black cells are more likely to form in future generations because they have a viability advantage at the group level. (b) A scenario is shown where group reproduction occurs. This opens the door for fecundity selection at the level of groups. In this picture, if a group possesses an innovation that improves its survival or reproduction, then the innovation can be passed on to daughter groups. For example, the production of specialized cell types (shown in gray) leads to a proliferation of groups with these specialized cells. Such a scheme requires both group reproduction and heredity of the developmental program. In this figure, we surround the constituent cells with a solid outer circle as they now have some of the properties associated with a higher-level individual (i.e., differentiation of parts and capacity to reproduce). If these groups compete with their free cell cousins and group formation confers advantages, then this population could shift from lower-level individuals to higher-level individuals, thereby accomplishing a major transition. (From Rainey and Kerr 2010, copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.)

Figure 8.2
The rise, the fall, and the outright destruction of a simple undifferentiated group. (Left) The wrinkly spreader mat is the cumulative product of the cooperative interactions of millions of cells. By working together, the cells in the mat colonize the air-liquid interface—a niche that is unavailable to the ancestral (broth colonizing) type. In colonizing this new niche, the cells of the mat are rewarded with an abundance of oxygen. (Middle) When the mat becomes too heavy, it collapses into the broth (it is not buoyant). The collapse is hastened by the presence of cheating genotypes that grow like a cancer within the mat, adding no structural strength, but reaping the benefits (access to oxygen). (Right) A mat is far more than the sum of the individual parts. This photo was taken immediately after a microcosm with an intact mat was disturbed (with a brief shake). The mat breaks into many pieces (just visible on the bottom) and does not spontaneously reform. Although a mat will eventually reemerge, it will do so by a process of growth and development from just a single cell. (From Rainey and Kerr 2010, copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.)

rich broth medium and incubated without shaking), the ancestral bacterium rapidly diversifies, producing a range of niche specialist genotypes (Rainey and Travisano 1998). Among the numerous emergent forms is a class of genotypes collectively known as wrinkly spreader (WS). WS genotypes are so named because of their distinctive wrinkly colony morphology on agar plates, but the important and relevant phenotype is that which manifests in the broth-filled microcosm in which they evolve. In this environment, WS genotypes form a self-supporting mat at the air-liquid interface (figure 8.2).

WS genotypes arise from a wide range of simple mutations that result in overactivation of adhesive factors (a cellulosic polymer and a proteinaceous factor) (Spiers et al. 2002, 2003; Spiers and Rainey 2005). The overproduction of these glues causes cells to remain attached after cell division. Although there is a significant fitness cost to each individual WS mutant (Knight et al. 2006; Maclean, Bell, and Rainey 2004; Rainey and Rainey 2003), WS cells nonetheless increase in frequency, ultimately outcompeting the ancestral genotype. They achieve this because the cost to individual cells is traded against a benefit that accrues to the group of WS cells. It works as follows: The production of adhesive glues means that, upon binary fission, daughter cells remain linked. Continuing cell division causes the population of cells to expand in a single cell layer across the air-liquid interface, ultimately joining and becoming attached to the edge of the glass vial. Once the surface is colonized, the mat grows in thickness, becoming a robust structure that is the cumulative product of the cooperative interactions of many millions of cells. By working together, the cells in the mat colonize a niche unavailable to the ancestral type. In colonizing this new

niche, the cells of the mat are rewarded with an abundance of oxygen (Rainey and Rainey 2003).

The evolution of WS is thus an example of the evolution of cooperation—de novo and in real time—from an ancestral state that is asocial and unicellular. The spread of polymer production is readily explained by kin selection (Hamilton 1964a,b). Baring mutation, clonal reproduction means that WS mats are composed of individuals whose relatedness is complete, the mat being a clone of genetically identical cells. Given mutation, the evolution of cheating (selfish) types is to be expected. Such types arise rapidly from the cooperating WS cells by mutation and grow as a cancer within the mat. Cheats do not produce adhesive polymers and therefore grow rapidly. Provided they arise within the fabric of the mat (or better still on the mat surface), they reap the benefits of group membership (access to oxygen) while forgoing the cost associated with polymer production; of course, in so doing they make no contribution to the network of polymeric strands required for maintenance of mat integrity. As might be anticipated, the cancerous growths ultimately compromise strength of the WS mat, which ultimately collapses (Rainey and Rainey 2003; see figure 8.2): a classic tragedy of the commons (Hardin 1968).

Adaptive Evolution of WS Groups

Having observed the emergence of groups, it is of interest to consider the possibility of the further adaptive evolution of the groups themselves. Indeed, anyone considering this possibility is likely, at first glance, to appeal to standard (MLS-1) group selection models, but it quickly becomes apparent that such models fail to fit with the biological reality of newly formed WS groups.

Standard group selection models most effectively explain the maintenance of cooperation in the face of selfish types that emerge as a consequence of selection at the lower level. In the absence of population structure, selfish types ultimately outcompete cooperating types, causing their extinction. If population structure (subdivision) exists, then cooperating types can be maintained provided cells periodically disperse into a global population, reassort, and then form new groups (Maynard Smith 1964; Wilson 1975).

Though theoretically sound, the maintenance of cooperation requires that the cells within each group periodically (and in a coordinated fashion) switch off traits that determine social behavior and then reactivate their expression (in a coordinated manner) to form new groups. This requires the existence of developmental control at the group level, and such control is highly unlikely to arise de novo in newly formed groups of cells; it certainly does not exist in WS groups. In the absence of a means of regulating social behavior, newly formed groups of cooperating cells will be driven extinct through the action of selection on selfish types.

Despite these reservations, we conducted initial experiments in a manner analogous to previous group selection experiments (Goodnight 1985; Swenson, Wilson, and Elias 2000, reviewed in Kerr 2009; Wade 1977) in which selection was imposed for a property of WS

groups, namely, mat strength. Indeed, the presence of substantial heritable phenotypic variation among different WS genotypes makes such group selection experiments possible (Bantinaki et al. 2007). Selection for mat strength does provoke a rapid response, although such a response is short-lived due to the ultimate triumph of cheating genotypes that, despite continued selection for mat strength, erode the success of even the strongest mats (McDonald and Rainey, unpublished). Experiments such as this demonstrate beyond doubt the capacity of selection to act on groups. However, in conducting standard group-selection experiments, the experimenter defines the group-level trait on which to select: The experimenter is also the vehicle of group reproduction. This more or less precludes the evolution of groups in ways that would see a genuine improvement in the Darwinian fitness of groups. The essential ingredient that is absent is any endogenous capacity for group reproduction—for groups to leave group offspring. In the absence of group reproduction an essential component of Darwinian individuality is lacking and, though selection can operate on viability, it is unable to act on group fecundity. In its absence, the evolution of traits adaptive at the group level is unlikely.

One possible way forward would be for group reproduction to be effected by an external factor, for example, stochastic disturbance of the broth in which the mats grow. The groups would therefore be endowed with individuality of a kind, but it is difficult to see how this haphazard means of reproduction would be effective. Dawkins (1982) comes to a similar conclusion about the difficulty of organismal adaptation given reproduction through a type of slapdash fissioning. For Dawkins, adaptive evolution at the level of the multicellular organism requires a developmental cycle (e.g., multicellular differentiation from a single cell origin in each generation). Returning to our WS mats, even if disturbance events could assist in mat reproduction, any newly emergent group will face the threat of extinction unless it has some way to maintain selfish types at low frequency. This, of course, requires a means of regulating sociality—itself a group-level trait and one that is difficult to envisage evolving without selection acting at the level of the group. This returns us to the problematic situation referred to earlier.

A New Hypothesis

Grappling with the problem of how WS groups might evolve traits that are adaptive at the group level—in a manner analogous to the adaptive evolution of individual cells—led eventually to the acceptance that such groups are incapable of evolving, except in an MLS-1 sense (provided the experimenter acts as the means of group reproduction), because of the inability of groups to leave offspring collectives. Like soma, WS groups appear to be an evolutionary dead end. Experimental observations of the evolution and demise of WS groups, however, suggest a possible solution to the origin of group-level reproduction.

The genetic architecture underlying the evolution of WS genotypes is evolutionarily flexible: a product of the modularity of the underlying regulatory systems (Beaumont et al. 2006; McDonald et al. 2009). Ancestral genotypes readily, and rapidly, give rise to WS genotypes, which in turn can lose the mat-forming phenotype (by definition these are cheats) by simple mutations that suppress production of the adhesive cellulosic polymer. The effects of these suppressor mutations can be readily reversed by mutations at additional loci (Beaumont et al. 2009). Our experimental studies show that there exists an almost inexhaustible supply of mutational routes for transitioning between group and individual cell states with minimal deleterious effects on fitness (Beaumont, Kost, Ferguson, Farr, and Rainey, unpublished).

Viewed in terms of cooperation and conflict, such findings suggest little opportunity for anything other than an ecological arms race between the groups and the cheats. However, viewed from a different perspective, the repeated evolution of groups from cheats, and cheats from groups, is analogous to a simple life cycle (figure 8.3). Since cell lineages can mutate so readily between a cooperative and a selfish morph, we can think of the unglued defectors who leave the mat as propagules of the mat itself. It is likely, though not certain, of course, that when ecological conditions again favor the WS mat-building morph, one or more of these cell-line lineages will re-evolve that phenotype and a new mat will develop. The germline is chosen by a form of mutational lottery, and heritability is not of high fidelity, as return to the mat phenotype depends on further mutation. But we do have a protolife cycle. As if by a sleight of hand, this change in perspective—combined with appropriate ecological conditions for the life cycle to take place—places the newly emergent WS groups in an MLS-2 framework. Groups that were seemingly incapable of leaving collective copies are now endowed with this capacity thanks to the cheating genotypes that serve in a manner analogous to a primitive germline. There is a clear irony in this altered perspective: The cheating genotypes—those types typically viewed as the greatest impediment to evolutionary transitions—stand as the savior of the groups. In fact, there is an even deeper irony: The cheating genotypes, though the nemesis of the group, are also its savior. In the context of the WS, when the mat collapses, the cheats, which are not glued to the mat fabric, are liberated. In an MLS-1 sense, the cheats cause the demise of the group, but from an MLS-2 perspective, the cheats are the means by which the group leaves offspring. Individuality, of a sort, at the higher level thus emerges from nothing more than the tension that exists between levels of selection. Rather than hinder the transition to higher levels of individuality, we suggest that there is creative potential in this tension—a tension that may have played an important role in certain transitions in individuality.

Toward Individuality

As emphasized previously, for traits adaptive at the group level to emerge, it is necessary for the group to be a Darwinian individual. How much further have we progressed with this

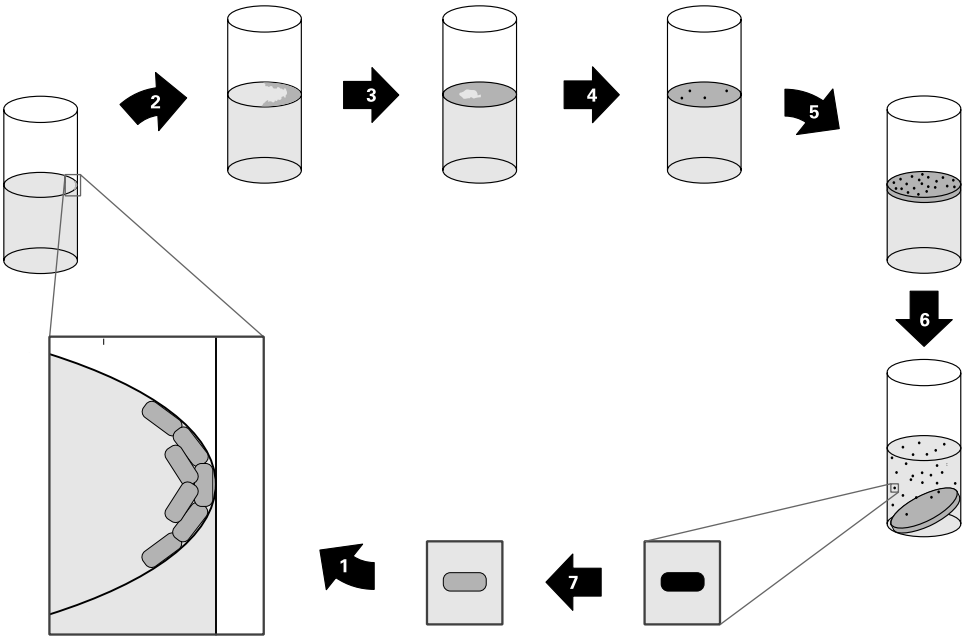


Figure 8.3
A putative life-cycle for mat-forming bacteria. We start with a single bacterium (given in gray) capable of producing an extracellular adhesive. (1) It reproduces at the interface between liquid and air (in the case shown, starting at the surface of a glass tube). Daughter cells stick together because of the adhesive they produce. (2, 3) The resulting biofilm spreads over the liquid's surface as a single cell layer. (4) Due to prime access to oxygen, a robust mat forms. Mutation generates "cheats" (cells that do not produce any adhesive polymer and grow faster as a consequence—represented in black). (5) These cells spread like a cancer within the mat and contribute to (6) the collapse of the mat. Because the cheats do not produce the adhesive, they are liberated from the mat upon collapse. (7) Back mutation from one of these cheats to a mat-producing cell completes the life cycle. Of course, we don't imagine such a life cycle playing out in an environment where only a single mat can form (like a single tube). Rather, the back mutants from the liberated cheats could establish mats in different locations from their parent mat. Here, the cell type leading to the death of the group also leads to its rebirth. The cheats form a germline, arising de novo from the mat-forming soma of an incipient multicellular individual. (From Rainey and Kerr 2010, copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.)

altered perspective—the notion of cheat as germline? On one hand, a great deal of progress is made, but true individuality remains some way off. The most obvious shortcoming is in the life cycle itself: It is clearly not a life cycle as we know it, not even compared to the simplest of life cycles such as those encountered in primitive volvocine algae such as *Gonium* (Kirk 2005). The major difference is the dependence on mutation (rather than developmental control) to transition between the soma and germline states. In addition to the crudeness, limitations, and constraints imposed by this mutational requirement, there is the thorny issue of heredity. The problem is not as great as it may first seem, however. The critical issue, as we will demonstrate mathematically, is the rate of transition between states, and this rate is heritable.

The reliance on mutation to transition between different stages of the life cycle need not, indeed cannot, be permanent. There is no reason why selection working at the level of group fecundity could not eventually find a way to bring the life cycle under developmental control. In principle, such control could come through a very small number of mutations that might, for example, bring polymer biosynthesis under the control of an extant oxygen-sensing regulator. In the presence of oxygen (the oxygen-replete air-liquid interface), cells would activate polymer biosynthesis and form WS mats. As the mat develops, a steep oxygen gradient forms across the mat, with cells on the underside suffering anoxia; polymer production would thus cease, allowing cells on the underside of the mat to swim away to once again activate group formation after a suitable (oxygen replete) niche is identified.

This scenario might seem fanciful, however, a recent selection experiment in which *P. fluorescens* cells were "forced" to transition rapidly between groups gives reason for optimism. After just four cycles between WS group and cheat, in two (of twelve) replicate lines, genotypes arose that had evolved the capacity to switch stochastically between states by an epigenetic mechanism (Beaumont et al. 2009).

A Model for Adaptive Mat Development

To explore the evolution of multicellular development in the *Pseudomonas* system, we introduce a discrete-time theoretical model. In this model, there are two types of cells, mat-formers and swimmers (cheats). We begin by describing the population dynamics of these cell types within a single mat. We assume that every mat is initialized by a single mat-former cell. Over time, mutation can generate swimmers. Let the $m(t)$ and $s(t)$ be the sizes of the mat-former and swimmer populations, respectively, in a single mat at time t. Populations within a mat grow according to the following branching process (Haccou, Jagers, and Vatutin 2005):

$$m(t+1) = \sum_{i=1}^{m(t)} [X_i - F_i(X_i)] + \sum_{j=1}^{s(t)} G_j(Y_j) \tag{8.1a}$$

$$s(t+1) = \sum_{j=1}^{s(t)} [Y_j - G_j(Y_j)] + \sum_{i=1}^{m(t)} F_i(X_i) \tag{8.1b}$$

The sets $\{X_1, X_2, X_3, \ldots\}$ and $\{Y_1, Y_2, Y_3, \ldots\}$ contain i.i.d. Poisson-distributed random variables ($X \sim \text{Poisson}(\beta_m)$ and $Y \sim \text{Poisson}(\beta_s)$). The i^{th} mat-former has X_i offspring cells, whereas the j^{th} swimmer has Y_j offspring cells. In this model, β_m and β_s are the average number of offspring cells per mat-forming cell and swimmer cell, respectively, per unit of time. Thus, we can think of these βs as birth factors. Because swimmers reproduce without contributing to the integrity of the mat, we assume that these cells have a birth rate advantage—that is, $\beta_s > \beta_m$.

The sets $\{F_1, F_2, F_3, \ldots\}$ and $\{G_1, G_2, G_3, \ldots\}$ contain i.i.d. binomially distributed random variables $[F(n) \sim \text{Binomial}(n, \mu_{m,s})$ and $G(n) \sim \text{Binomial}(n, \mu_{s,m})]$. Of its X_i offspring, the i^{th} mat-former has F_i swimmer mutants. And of its Y_j offspring, the j^{th} swimmer has G_j mat-former mutants. For simplicity, we let the probability of mutation from mat-former to swimmer ($\mu_{m,s}$) and from swimmer to mat-former ($\mu_{s,m}$) be equal: $\mu_{m,s} = \mu_{s,m} = \mu$.

The cell dynamics within a microbial mat are given by equations 8.1a and b. In addition, we assume that any mat has a finite lifetime. Specifically, the probability that a mat will collapse at time t is given by:

$$P_C(t) = 1 - \exp\{-(\alpha_m m(t) + \alpha_s s(t))\} \tag{8.2}$$

Thus, as the number of cells in a mat increase, the mat is more likely to collapse. Again, because swimmers do not contribute to mat integrity, swimmer cells have a disproportionately negative effect on the lifetime of the mat—that is, $\alpha_s > \alpha_m$.

For a collapsed mat, swimmer cells are the only way to "cash in" reproductively. Swimmer cells that survive mat collapse become the single mat-formers (after mutation) that give rise to new mats. As a consequence, mats face a trade-off between viability and fecundity. Production of swimmers makes a mat more fecund, but these very same swimmers shorten the lifetime of the mat. The way a mat consigns cells to different categories defines its developmental program. In turn, this developmental program yields the life history of the mat. Specifically, the investment in different cell populations affects when the mat expires and how much the mat invests in reproduction. In order to determine the optimal developmental program for a mat, we identify parameters in the model that affect this development.

One model parameter that influences the production of swimmers from mat-formers and vice versa is the mutation rate (μ). Of course, mutation is a stochastic process, and thus mat development is "noisy." That is, even if two mats have the same "program" (the same μ value), they are likely to produce different cell distributions due to the stochasticity inherent in development. Nonetheless, a change in the mutation rate will affect on the likelihood of different cell distributions. In this way, the mutation rate can affect mat fecundity and viability.

Before we discuss how to find the optimal developmental program (i.e., the best mutation rate), we need to specify fitness at the level of mats. Mat-level fitness is related to the ability of the mat to generate offspring mats (true to the MLS-2 conception of group fitness). Thus, it would seem that mat-level fitness should be proportional to the number of swimmer cells contained in the mat on its collapse. This fitness metric is fully adequate if mats always have the same generation time. However, the generation time of a mat is specified (at least probabilistically) *by its developmental program*.

The issue of variable mat lifetime potentially complicates a simple measure of mat-level fitness. All else being equal, a shorter generation time is beneficial within a growing population of mats. However, because swimmer cells simultaneously contribute to mat

reproduction and expiration, all else is not equal. For instance, if a slightly longer-lived mat can have many more swimmer cells upon collapse, then it may be advantageous to live longer. In some cases, a proper consideration of mat fitness will take into account the rate of growth of a *population of mats* that share the same developmental program. In the same way we would talk about cell populations displacing one another inside a mat, we can talk about mat populations (each characterized by a developmental program) displacing one another in their environment.

Different ecological circumstances will favor different developmental programs—that is, different investments in fecundity and viability. Here, we consider two ecological conditions. In the first condition (which we label *r*-selection), sites for mat formation are always available, so there is a premium on a short mat generation time. Production of swimmers should be adjusted as to maximize growth rate within an expanding *population* of mats. In the second condition (which we label *K*-selection), sites for mat formation rarely open up and there is pressure to lengthen mat generation time to maximize the *absolute number* of swimmer cells a mat produces (we assume these cells "lie in wait" for sites to open). We can use our model to identify the optimal mutation rate under *r*- and *K*-selection.

Assume a given mat collapses at t^*. There are $s(t^*)$ swimmer cells in the mat at this time, which we label s^*. Under *r*-selection, we wish to maximize the growth rate of mats within a mat population. To do this, we must consider the joint distribution of t^* and s^*. Specifically, for any mat, we have

$$\text{Prob}(t^* = T \text{ and } s^* = S) = \pi(T, S)$$

Armed with this distribution, the long-term growth rate (r) of a mat population with a specified developmental program is given by the solution to the Euler-Lotka equation (Euler 1760; Fisher 1930; Lotka 1925):

$$\sum_{T=0}^{\infty}\sum_{S=0}^{\infty}\pi(T,S)\mu\, Se^{-rT} = 1$$

For simplicity, we assume that a fraction μ of the swimmers mutate back to mat-formers directly after the mat collapses.

Here, we use a Monte Carlo simulation approach to generate the joint distribution π. Specifically, we generate 50,000 points $(t^*, s^*)_i$ using equations 8.1a and b and 8.2. An example of this joint distribution is shown in figure 8.4. In the figure, we see the life history trade-off faced by the mat: Higher fecundity tends to require a longer generation time. Once we have this joint distribution, we simply solve the following equation for r:

$$\sum_{i=1}^{50000}\frac{\mu s_i^* e^{-rt_i^*}}{50000} = 1$$

We then look for the mutation rate (μ) that maximizes r.

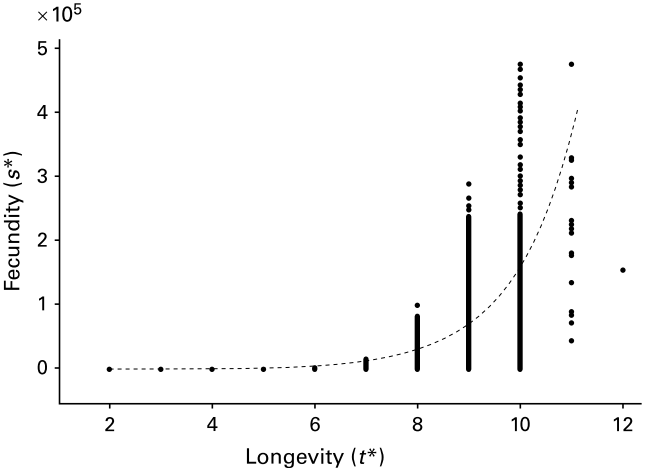


Figure 8.4
The joint distribution of mat longevity and mat fecundity. These points were generated from simulations of mat development given by equations 8.1 and 8.2 ($\beta_m = 4.0$, $\beta_s = 6.0$, $\alpha_m = 10^{-6}$, $\alpha_s = 10^{-5}$). (From Rainey and Kerr 2010, copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.)

For *K*-selection, we are looking for the mutation rate that maximizes *s**. We can employ the same Monte Carlo approach to generate 50,000 *s** values. Then we look for the mutation rate that maximizes the average *s** value.

Figure 8.5 shows the results of our analysis. We see that under *r*-selection, high mutation rates are favored, whereas under *K*-selection, lower mutation rates are favored. Under *r*-selection, longevity is sacrificed for a quick investment in swimmers, allowing a rapid explosion of mats. Under *K*-selection, longer-lived mats are selectively favored to maximize swimmer output.

Consideration of Alternative Hypotheses

The mutationally driven life cycle outlined above is but one example of a life cycle that emerges at the same instant that individuality transitions between levels. As such, it is possible to understand the evolutionary emergence of the life cycle without the need to invoke group reproduction as a precondition for its own evolution. Here, we consider an alternative hypothesis in which the germline is uninterrupted by mutation. From the outset, such a model is appealing because it removes the potentially restrictive requirement of mutation for the transition between stages of the life cycle. Once again we make use of the model *Pseudomonas* populations as a vehicle for our ideas, but this time take as the focus of interest the lower-level (cheating) entities.

Consider the cheating type as a totipotent germline. Imagine that during the course of its growth it produces, by chance mutation, a cell type with which it interacts, either directly,

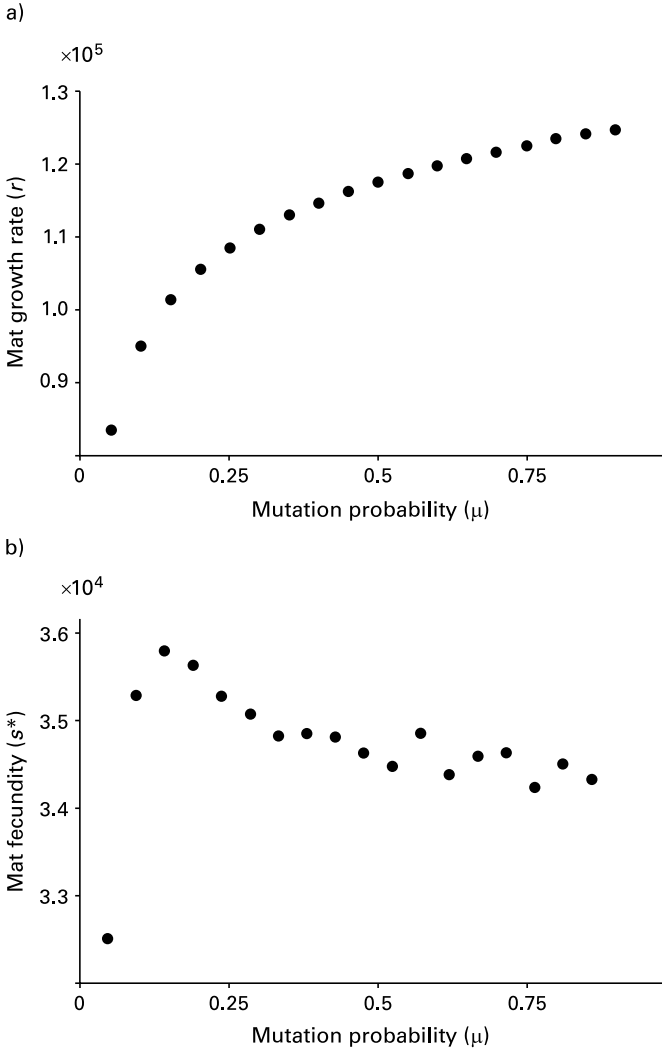


Figure 8.5
Optimal mutation rates in mat development. (a) Long-term growth (in an *r*-selected environment) is shown as a function of the mutation probability. Here, we see higher mutation rates yield faster growth of a lineage of mats. (b) Mat fecundity (favored in a *K*-selected environment) is maximized at lower rates of mutation. In parts a and b, the parameters of the model are the same as those in figure 8.4. (From Rainey and Kerr 2010, copyright Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission.)

or indirectly, and which, via that interaction, aids its own reproductive output. We might consider this a "helper" type; indeed, we might consider the WS genotype an exemplar of such a helper, although in so doing we add a level of complexity (and selection) that is not necessary. The helper may be any kind of reproductive altruist. An interesting example is provided by the suicidal altruists of *Salmonella typhimurium* that die while preparing the ground for infection (Ackermann et al. 2008). Nonetheless, returning to the familiar WS: as the mat forms, it becomes infiltrated by cells of the germline, which reap the advantage that accrues from growth at the air-liquid interface. Eventually, the mat collapses and the WS lineage goes extinct; nonetheless, the germline remains and in time gives rise to further WS types that it again exploits for its own advantage. Such a scenario captures elements of an earlier hypothesis for the origin of the germline in which the germline originates as a consequence of "other cell lineages altruistically removing themselves from the reproductive line to perform some somatic benefit to the organism" (Queller 2000). In this example, however, there is no altruism on the part of the WS—at least not in the sense of some kind of indirect benefit; rather, the WS aid the germline because their helping phenotype is set by the germline. From one perspective, the WS can be seen as an extreme altruist, sacrificing its life for the germline (altruism being an indirect consequence of the short-term advantage gained from colonization of the oxygen-replete air-liquid interface). From another perspective, the WS can be seen as an unfortunate pawn, sacrificed *by* the germline. This second perspective bears resemblance to parasites such as *Toxoplasma gondii* that control the behavior of their host in order to advance their own interests (Webster 2001).

Thus, from different starting positions we arrive at essentially the same end point: In both interrupted and uninterrupted models, potential exists for the evolution of a life cycle, and with that potential to arrest in the germline stage. Individuality in an MLS-2 sense is apparent. There are, however, some differences. For example, the interrupted model carries with it the initially burdensome requirement for mutation to mediate the transition between different stages of the life cycle, whereas the uninterrupted model requires only one-way mutation (to dead-end helper cells). In this sense, the uninterrupted model seems to offer a lower hurdle for an evolutionary transition. However, things get more interesting when one considers a second distinguishing feature, namely, the origin of multicellular differentiation. The uninterrupted model requires the emergence of extreme altruism via mutation in the presence of would-be "cheats." On the other hand, the interrupted model may involve nothing more than the advent of cheats in the face of cooperation. All else being equal, it seems the advantage goes to the interrupted model regarding the ease of such differentiation. Subtle differences in the "quality" of individuality at the MLS-2 and the requirements for the higher level of individuality in both instances also raise some interesting issues (we consider these fully elsewhere; see Rainey and Kerr 2010).

In outlining these two models our intention has been to portray possible scenarios for the evolution of life cycles—particularly the selective conditions favouring ecologically distinct phenotypes—that might eventually evolve to come under regulatory (developmental)

control. The molecular details by which such control could emerge are unknown but are likely to depend on nonadaptive processes such as mutation and genetic drift (Lynch 2007), opportunities for co-option (True and Carroll 2002) (facilitated by mutation and drift) and the existence of plasticity (West-Eberhart 2003). Under some circumstances it is even possible that the plasticity inherent in the genomic and regulatory organization of certain unicellular entities might be sufficient to produce a simple life cycle with minimal involvement from selection. For example, single cells driven to group formation as a mechanism of predation-avoidance might—given an appropriately organised and preprepared regulatory system—be capable of utilizing gradients generated across the colony as a means of regulating the transition between clumping and dispersing behaviors (Hochberg, Rankin, and Taborsky 2008). An idea like this involving co-option of a life history gene has been suggested to explain the evolution of reproductive altruism in the higher volvacine algae (Nedelcu and Michod 2006). The central idea is that in the ancestral (unicellular) state expression of the life history gene is conditioned on an environmental cue, but during the transition to multicellularity it evolves to come under the control of spatial (developmental) signals. Such a scenario makes a good deal of sense and is even supported by studies of *regA* expression (a regulator of chloroplast expression) in unicellular versus multicellular volvacine algae (Nedelcu and Michod 2006; see also chapter 9 by Michod, in this volume). However, just how such a change comes about—particularly the change necessary to bring differentiation under the control of endogenous signals—still requires an evolutionary explanation (see Michod 2006 and Michod et al. 2006, for a possible mechanism based on a viability-fecundity trade-off).

Additional scenarios for the evolution of life cycles that might effect the transition from MLS-1 to MLS-2 can be envisaged and the potential range of factors influencing the route taken are numerous, but arguably the most important factors will be ecological—the particular selective environment experienced by the evolving populations. Here is not the place to further explore the issues raised by comparisons of different models, but there are reasons for suspecting that the ultimate outcome of evolution under each will be distinctive.

Conclusion

Darwinian transitions in individuality, particularly those originating from a fraternal alliance among lower level entities, pose some of the most tantalizing problems in biology. Here we have drawn attention to the need to explain, in mechanistic terms, how variation in lower level individuals generates a corporate entity with Darwinian characteristics. Our emphasis on this issue stems from recognition that any explanation for the evolution of multicellularity from unicells—for the transition between MLS-1 and MLS-2—is dependent upon explaining how collectives evolve the capacity to leave collective offspring. The

life cycle, we argue, is the critical innovation: Life cycles decouple fitness—they transition individuality.

The unconventional life cycles described above that span the MLS-1 to MLS-2 juncture are founded in experimental reality. The interrupted life cycle model can operate in experimental *Pseudomonas* populations and, via its operation, WS mats can assume the role of "organisms"—organisms whose fitness is measured, not by the number of bacterial cells within each mat, but by the number of mat-offspring left by parents.

In advocating this model as one route to a proto-life cycle we recognize the irony. Tensions between levels of selection are typically viewed as significant impediments to evolutionary transitions (Michod 1996), but our altered perspective reveals a creative role for conflict. This conflict generates in a single step a means of collective reproduction, a life cycle, the basis of a self-policing system, and ecological circumstances possibly conducive to the eventual emergence of development. In addition, the hypothesis provides a plausible scenario for the origin of a soma/germline distinction, and for sequestration of the germline by soma—the latter arising from the fact that WS "soma" is under strong selection to check increased replication of cheating "germline" types. In this context it is interesting to note recent ideas on the evolution of ageing as a deprivation syndrome (Heininger 2002) driven by the tension between soma and germline—a tension that perhaps, at least for some evolutionary transitions, may have an ancient past.

Ideas are readily come by, but ideas translated into experiment have potency. Arguably the most important aspect of the hypothesis presented here is its experimental tractability. The focus of our own work remains bacterial populations and accordingly, in the absence of sex, pliable cell walls, and the energetic benefits attributable to the mitochondria, we do not expect to see a transition to multicellularity as evident in eukaryotic organisms, but that does not mean that early stages in the evolution of individuality cannot be explored, or indeed, the emergence of traits adaptive at the group level directly witnessed.

Acknowledgments

We thank members of the Rainey lab and particularly Katrin Hammerschmidt, Eric Libby, Peter Meintjes, and Caroline Rose for valuable discussion. Brett Calcott and Kim Sterelny provided insightful comments on drafts of the manuscript.

References

Ackermann, M., B. Stecher, N. E. Freed, P. Songhet, W. D. Hardt, M. Doebeli. (2008). Self-destructive cooperation mediated by phenotypic noise. *Nature* 454: 987–990.

Bantinaki, E., R. Kassen, C. Knight, Z. Robinson, A. J. Spiers, and P. B. Rainey. (2007). Adaptive divergence in experimental populations of *Pseudomonas fluorescens*. III. Mutational origins of wrinkly spreader diversity. *Genetics* 176: 441–453.

Beaumont, H. J., J. Gallie, C. Kost, G. C. Ferguson, and P. B. Rainey. (2009). Experimental evolution of bet hedging. *Nature* 462: 90–93.

Beaumont, H. J. E., R. Kassen, C. G. Knight, J. Malone, A. J. Spiers, and P. B. Rainey. (2006). The genetics of phenotypic innovation. In N. Logan (ed.), *Prokaryotic Diversity: Mechanisms and Significance.* Cambridge: Cambridge University Press.

Bonner, J. T. (1998). The origins of multicellularity. *Integrative Biology* 1: 27–36.

Bonner, J. T. (2000). *First Signals: The Evolution of Multicellular Development.* Princeton, NJ: Princeton University Press.

Buss, L. W. (1987). *The Evolution of Individuality.* Princeton, NJ: Princeton University Press.

Charlesworth, B. (1980). Models of kin selection. In H. Markl (ed.), *Evolution of Social Behaviour.* Weinheim: Verlag Chemie.

Conway Morris, S. (1998). *The Crucible of Creation.* Oxford: Oxford University Press.

Damuth, J., and I. L. Heisler. (1988). Alternative formulations of multi-level selection. *Biology & Philosophy* 3: 407–430.

Dawkins, R. (1982). *The Extended Phenotype.* Oxford: Oxford University Press.

Dennett, D. C. (1995). *Darwin's Dangerous Idea: Evolution and the Meanings of Life.* London: Penguin Books.

Euler, L. (1760). Recherches générales sur la mortalité: La multiplication du genre humain. *Memoires de l'academie des sciences de Berlin* 16: 144–164.

Fisher, R. A. (1930). *The Genetical Theory of Natural Selection.* Oxford: Oxford University Press.

Frank, S. A. (2007). *Dynamics of Cancer: Incidence, Inheritance and Evolution.* Princeton, NJ: Princeton University Press.

Goodnight, C. J. (1985). The influence of environmental variation on group and individual selection in a cress. *Evolution* 39: 545–558.

Griesemer, J. (2000). The units of evolutionary transition. *Selection* 1: 67–80.

Haccou, P., P. Jagers, and V. Vatutin. (2005). *Branching Processes: Variation, Growth and Extinction of Populations.* Cambridge: Cambridge University Press.

Hamilton, W. D. (1964a). The genetical evolution of social behaviour. 1. *Journal of Theoretical Biology* 7: 1–16.

Hamilton, W. D. (1964b). The genetical evolution of social behaviour. 2. *Journal of Theoretical Biology* 7: 17–52.

Hardin, G. (1968). The tragedy of the commons. *Science* 162: 1243–1248.

Heininger, K. (2002). Aging as a deprivation syndrome driven by a germ-soma conflict. *Aging Research Reviews* 1: 481–536.

Heisler, I. L., and J. Damuth. (1987). A method for analyzing selection in hierarchically structured populations. *The American Naturalist* 130: 582–602.

Herron, M. D., and R. E. Michod. (2008). Evolution of complexity in the volvocine algae: Transitions in individuality through Darwin's eye. *Evolution* 62: 436–451.

Hochberg, M. E., D. J. Rankin, and M. Taborsky. (2008). The coevolution of cooperation and dispersal in social groups and its implications for the emergence of multicellularity. *BMC Evolutionary Biology* 8: 238.

Jablonka, E., and M. J. Lamb. (2006). The evolution of information in the major transitions. *Journal of Theoretical Biology* 239: 236–246.

Kerr, B. (2009). Theoretical and experimental approaches to the evolution of altruism and the levels of selection. In T. Garland, and M. R. Rose (eds.), *Experimental Evolution: Concepts, Methods, and Applications of Selection Experiments.* Berkeley: University of California Press.

Kirk, D. L. (1998). *Volvox: Molecular Genetic Origins of Multicellularity and Cellular Differentiation.* Cambridge: Cambridge University Press.

Kirk, D. L. (2005). A twelve-step programme for evolving multicellularity and a division of labour. *Bioessays* 27: 299–310.

Knight, C. G., N. Zitzmann, S. Prabhakar, R. Antrobus, R. Dwek, H. Hebestreit, and P. B. Rainey. (2006). Unravelling adaptive evolution: How a single point mutation affects the protein co-regulation network. *Nature Genetics* 38: 1015–1022.

Lotka, A. J. (1925). *Elements of Physical Biology.* Baltimore: Williams & Watkins.

Lynch, M. (2007). The frailty of adaptive hypotheses for the origins of organismal complexity. *Proceedings of the National Academy of Sciences USA* 104(Suppl 1): 8597–8604.

Maclean, R. C., G. Bell, and P. B. Rainey. (2004). The evolution of a pleiotropic fitness tradeoff in *Pseudomonas fluorescens*. *Proceedings of the National Academy of Sciences USA* 101: 8072–8077.

Maynard Smith, J. (1964). Group selection and kin selection. *Nature* 201: 1145–1146.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution.* Oxford: W. H. Freeman.

McDonald, M. J., S. M. Gehrig, P. L. Meintjes, X. X. Zhang, and P. B. Rainey. (2009). Adaptive divergence in experimental populations of Pseudomonas fluorescens. IV. Genetic constraints guide evolutionary trajectories in a parallel adaptive radiation. *Genetics* 183: 1041–1053.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality.* Princeton, NJ: Princeton University Press.

Michod, R. E. (2006). The group covariance effect and fitness trade-offs during evolutionary transitions in individuality. *Proceedings of the National Academy of Sciences USA* 103: 9113–9117.

Michod, R. E., and A. M. Nedelcu. (2003). On the reorganization of fitness during evolutionary transitions in individuality. *Integrative & Comparative Biology* 43: 64–73.

Michod, R. E., Y. Viossat, C. A. Solari, M. Hurand, and A. M. Nedelcu. (2006). Life-history evolution and the origin of multicellularity. *Journal of Theoretical Biology* 239: 257–272.

Nedelcu, A. M., and R. E. Michod. (2006). The evolutionary origin of an altruistic gene. *Molecular Biolology of Evolution* 23: 1460–1464.

Okasha, S. (2006). *Evolution and the Levels of Selection.* Oxford: Oxford University Press.

Queller, D. C. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions of the Royal Society of London, B* 355: 1647–1655.

Rainey, P. B. (2007). Unity from conflict. *Nature* 446: 616.

Rainey, P. B., and B. Kerr. (2010). A new hypothesis for the evolution of individuality during the transition from single cell organisms to multicellularity. *BioEssays* 32: 872–880.

Rainey, P. B., and K. Rainey. (2003). Evolution of cooperation and conflict in experimental bacterial populations. *Nature* 425: 72–74.

Rainey, P. B., and M. Travisano. (1998). Adaptive radiation in a heterogeneous environment. *Nature* 394: 69–72.

Sober, E., and D. S. Wilson. (1998). *Unto Others: The Evolution and Psychology of Unselfish Behaviour.* Cambridge, MA: Harvard University Press.

Spiers, A. J., J. Bohannon, S. M. Gehrig, and P. B. Rainey. (2003). Biofilm formation at the air-liquid interface by the *Pseudomonas fluorescens* SBW25 wrinkly spreader requires an acetylated form of cellulose. *Molecular Microbiology* 50: 15–27.

Spiers, A. J., S. G. Kahn, J. Bohannon, M. Travisano, and P. B. Rainey. (2002). Adaptive divergence in experimental populations of *Pseudomonas fluorescens*. I. Genetic and phenotypic bases of wrinkly spreader fitness. *Genetics* 161: 33–46.

Spiers, A. J., and P. B. Rainey. (2005). The *Pseudomonas fluorescens* SBW25 wrinkly spreader biofilm requires attachment factor, cellulose fibre and LPS interactions to maintain strength and integrity. *Microbiology* 151: 2829–2839.

Swenson, W., D. S. Wilson, and R. Elias. (2000). Artificial ecosystem selection. *Proceedings of the National Academy of Sciences USA* 97: 9110–9114.

True, J. R., and S. B. Carroll. (2002). Gene co-option in physiological and morphological evolution. *Annual Review of Cell and Developmental Biology* 18: 53–80.

Velicer, G. J., and Y. T. N. Yu. (2003). Evolution of novel cooperative swarming in the bacterium *Myxococcus xanthus*. *Nature* 425: 75–78.

Wade, M. J. (1977). Experimental study of group selection. *Evolution* 31: 134–153.

Webster, J. P. (2001). Rats, cats, people and parasites: The impact of latent toxoplasmosis on behaviour. *Microbes and Infection* 3: 1037–1045.

West-Eberhart, M. J. (2003). *Developmental Plasticity and Evolution*. Oxford: Oxford University Press.

Wilson, D. S. (1975). A theory of group selection. *Proceedings of the National Academy of Sciences USA* 72: 143–146.

Wray, G. A. (2001). Dating branches on the tree of life using DNA. *Genome Biology* 3: 1–7.

III COMPLEXITY AND THE DEVELOPMENTAL CYCLE

Brett Calcott and Kim Sterelny

It is easy to overlook the metabolic, biochemical, and behavioral complexity of microbes, as they live and interact on a scale that makes observation challenging. Our sense of the extraordinary complexity and disparity of the multicellular world may depend, in part, on scale and perspective. Even so, understanding multicellularity is genuinely a special challenge. Multicellular organisms have developmental cycles: Organisms as enormous and as complex as a whale or a mountain ash were once a single fertilized cell. Tissues, organs, and organ systems have to be built from scratch in every generation. In a seed, roots, lignum, leaves, and flowers do not exist in miniature or in partial, fragmentary form, to act as templates or guides as a tree develops. With microorganisms, reproduction is fission, and many crucial structures are literally inherited from the parent cell. Nothing like this explains the stability and reliability of the developmental cycle of multicelled organisms. Multicelled organisms are typically huge compared with microorganisms, containing within each cell much of the complexity of a free-living microbe (though see McShea 2002), and with the extra complexities of their novel structures. In addition, they pose a vastly expanded and transformed problem of developmental control; a developmental cycle must be initiated and choreographed in each generation.

Likewise, understanding the division of labor in multicelled organisms seems to pose a special challenge (albeit one posed by eukaryotes as well). Prokaryotes have specialized parts. We might think of the flagella or the cell walls of a bacterium as miniature organs. But they are not organs that derive from independent organisms, nor are they composed of atoms that derive from free-living organisms. Complex multicellular life depends on the cooperative interaction of differentiated components, components whose ancestors were once Darwinian individuals with their own fitness interests and evolutionary fate. In many corporate organisms, the individual cell-line lineages have irretrievably lost their independent fate (though perhaps not as often as one might have supposed; see Leroi, Koufopanou et al. 2003). But even if the common fate of the cooperating cells of a macrobe is now frozen in place, it is hard to understand the origination and early stabilization of cooperation and differentiation, given divergent fitness differences and the opportunity to defect.

The solution to this problem, in broad outline, is now familiar. Selection can act at more than one level, and if there is structure to the population so that cooperative variants interact more regularly with other cooperators, the superior productivity of cooperative associations will inject more cooperators into the future, even though noncooperators do better in cooperator/noncooperator interactions (Okasha 2006). The problem has been to turn this broad picture into a realistic and testable scenario and, in particular, to explain the transition from MLS-1 to MLS-2 regimes (Okasha 2005). As we have noted earlier, one common thread in this collection is to conceptualize a major transition as a trajectory from collectives that vary in their contribution to the individual fitness of their members to corporate entities whose characteristics evolve as a consequence of corporate survival and reproduction. But it is one thing to identify and refine that theoretical framework, another to identify a lineage of incremental, selectable changes that will take us from collectives to corporations.

Paul Rainey and Ben Kerr's chapter (in the previous section) and Rick Michod's chapter on Evolutionary Transitions in Individuality differ in important ways; Michod sees the suppression of defection as essential, whereas Rainey and Kerr think that defection is coopted in transitions in individuality. But they all see the establishment of a reproductive division of labor as the foundation of this trajectory from MLS-1 to MLS-2. Thus, they build on an important tradition. Leo Buss's *Evolution of Individuality* took the suppression of selection based on variation in individual fitness at the atomic level to be crucial to the evolution of collective individuals. A germline-soma distinction is a crucial tool for suppressing such selection, as variation among somatic individuals has no downstream evolutionary consequence (Buss 1987).

Michod advances these issues by combining formal models of how the fitness of a collective can become decoupled from the fitness with more empirical work on the volvocine algae. Michod's key idea in explaining the origins of specialization is that, in an important class of cases, reproductive effort and somatic function trade off against each other. If this trade-off has the right shape, selection can favor specialization. Michod's idea of exploring these ideas by using the *Volvox* clade as a model system makes a lot of sense, for the clade includes free-living singled-celled organisms, simple aggregates with little or no differentiation, and larger organisms with a clear soma-germline separation and a full developmental cycle. Moreover, reproductive specialization pays for the *Volvox*, because this clade faces a very sharp trade-off between somatic and reproductive activity. The crucial somatic activity is powered movement using flagella. But the cellular machinery that build and use flagella is also used in mitosis, therefore a cell cannot both move and split. This lineage illustrates the trade-off that is pivotal to the formal models. It is less clear that the *Volvox* case generalizes: The sharp trade-off depends on an idiosyncratic feature of *Volvox* biology.

Two chapters in this section challenge and extend this understanding of the evolution of multicellularity. Carl Simpson argues that the MLS-1 to MLS-2 model of a major transition is incomplete because it overlooks a crucial component of fitness. He argues that the evo-

lution of individuality literature has failed to account for expansive fitness (roughly, the discretionary income an organism controls, which can be used for growth)—and that expansive fitness differences play an important role in the transition to regimes sensitive to the fitness of the corporate agent. Moreover, while accepting the MLS-1 to MLS-2 model, he thinks it is too coarse grained: There is a difference between life in a mere aggregate where your neighbors affect your fitness, and a life in which you have structured and systematic interactions (like making and responding to alarm calls). So he distinguishes between aggregates and groups, and suggests that the evolution of structured groups is an essential intermediary to the evolution of a corporate individual.

Ellen Clarke raises a different challenge. While metazoan history supports the idea that the germ-soma distinction is of foundational importance, the egalitarian transitions show that there is another route to corporate individuality. The eukaryotic alliance has evolved toward something like a division of reproductive labor, with the loss of many mitochondrial genes whose functions are taken over by the nuclear genome (Lane 2005). But this division of labor is a consequence, not a precondition, of a long history of intimate association as a complex, corporate agent. Clarke's "plant individuality and multilevel selection theory" develops the idea that the germ-soma split and the suppression of individual fitness differences within the corporate entity are not always essential steps in the evolution of corporate individuals. She shows that plant evolution does not fit this model.

Identifying individuals in plant populations is known to be fraught and complex (Harper 1977; Janzen 1977). One way of counting plants, the genet, defines individuality by genetic identity. An individual plant, counted this way, is a clade of genetically identical cells that descend from a single event of meiotic fusion. An individual genet need not correspond to a single plant in the gardener's sense, both because some physiologically cohesive plants are genetic mosaics and because vegetative reproduction and physiological accident can lead to a set of genetically identical but physiologically distinct structures. ("Identity" here is obviously a fuzzy notion; replication errors ensure that no large multicelled system really consists of a set of cells with identical DNA.) The ramet is the contrasting way of identifying evolutionary individuals: The individual is identified by appeal to developmental history and physiological integration. These contrasting ways of thinking about plant individuality are well known, in part because Richard Dawkins explored the issue at length, ultimately arguing that we should count by ramets because they are the products of a developmental cycle, and so are potential sources of evolutionarily consequential, selectable variation (Dawkins 1982).

Clarke's chapter reviews these issues but, more importantly, adds a third candidate: the meristem, or, more exactly, structural modules within plants that derive from a single meristem. A meristem, in turn, is a cell that retains its capacity to develop into soma or germ cells. In higher plants, meristems are typically distributed throughout the plant, rather than being sequestered in a special organ. Since somatic mutation is not uncommon, variation and fitness difference can arise between the different modules of the one plant as they

compete within the plant for space and other resources. Since many plants can reproduce vegetatively (by runners and other offshoots), and since most plants have no rigid germline-soma distinction, the mosaic of modules within the organism may not have the same evolutionary fate. Critically, Clark suggests that this competition can improve the vitality and fitness of the plant, as this internal competition enables the plant as a whole to reinforce the sites at which environmental and internal factors favor growth. Module competition within a plant is not like an unsuppressed cancer in a metazoan.

The other two chapters in this section focus less on these general issues of models and theory, and more on a specific, though truly spectacular example, the evolutionary radiation of the bilaterian metazoa in the Cambrian Explosion. Knoll and Hewitt develop a subtle model that integrates environmental and internal factors. They suggest that the explosion did have an external trigger; most probably a rise in atmospheric oxygen. But not all lineages radiated exuberantly in response; those that did were primed with existing capacities that could be coopted to the new challenges. Their chapter reminds us of the special physiological challenges posed by increased size in three dimensions. Internal cells need resources from the outside world, and they need to eject their waste into that world. As organisms grow large, diffusion no longer suffices for import and export. Modest increases in size and three-dimensional complexity exceed the limits of systems dependent on diffusion, and so transitions to complex multicellularity depend on the prior presence of mechanisms that allow the resource transport problem to be solved. Plumbing is unlikely to be the key to understanding the other transitions, but we suspect that Knoll and Hewitt's method of integrating environmental triggers and internal facilitators will productively generalize to other cases.

Like Knoll and Hewitt, Lindell Bromham's chapter, The Small Picture Approach to the Big Picture, is also focused on the Cambrian Explosion; in particular, on the issue of empirical support. It is one thing to build hypotheses and scenarios; it is another to test them. While all the chapters in this collection reflect on, and respond to, the issues of evidence, Bromham's chapter is the only one to focus on testability. Despite the antiquity of this instance of the multicellular transition, she argues that comparative molecular data impose serious constraints on macroevolutionary hypotheses, especially when we consider data from the so-called minor phyla. These enable us to constrain hypotheses about both the basal form of disparate phyla and their potential for large-scale change. Using these data, she argues that macroevolutionary hypotheses that depend on a qualitative distinction between the evolution of basic body plans and their further modification are not supported. Bromham's methodological case is convincingly argued, and rich phylogenetic information is available about lineages that evolved sociality and eusociality. It is less clear that we can use these comparative methods to test hypotheses about the more ancient transitions. But perhaps the signals of extensive horizontal gene flow at the base of the tree of life (discussed in Sterelny's chapter) hint that phylogenetic methods can reach very deep into history.

References

Buss, L. (1987). *The Evolution of Individuality.* Princeton, NJ: Princeton University Press.

Dawkins, R. (1982). *The Extended Phenotype.* Oxford: Oxford University Press.

Harper, J. L. (1977). *The Population Biology of Plants*. New York: Academic Press.

Janzen, D. H. (1977). What are dandelions and aphids? *The American Naturalist* 111: 586–589.

Lane, N. (2005). *Power, Sex, Suicide: Mitochondria and the Meaning of Life.* Oxford: Oxford University Press.

Leroi, A., L. M. Koufopanou, et al. (2003). Cancer selection. *Nature Reviews Cancer* 3(3): 226–231.

McShea, D. W. (2002). A complexity drain on cells in the evolution of multicellularity. *Evolution* 56: 441–452.

Okasha, S. (2005). Multilevel selection and the major transitions in evolution. *Philosophy of Science* 72(4): 1013–1025.

Okasha, S. (2006). *Evolution and the Units of Selection.* Oxford: Oxford University Press.

9 Evolutionary Transitions in Individuality: Multicellularity and Sex

Richard E. Michod

Reorganization of Fitness during Evolutionary Transitions in Individuality (ETIs)

Levels of Selection and Levels of Complexity

The challenge of explaining evolutionary transitions (ETs) was initially posed by Maynard Smith in two papers (1988, 1991) and later in a more systematic and comprehensive way in his book with Szathmáry (1995). The list of "levels of complexity" first offered by Maynard Smith (1988, table 2) focused on the levels of selection: replicating molecules, replicators in compartments, prokaryotic cells, eukaryotic cells, multicellular organisms, demes and social groups, species, and groups with cultural inheritance. Maynard Smith noted that it was debatable whether the last three were levels of selection; indeed, the level of species was left out of his list in the later paper and sex was added as an ET in the book with Szathmáry. Maynard Smith used the *levels of selection* as the defining framework of evolutionary transitions between different *levels of complexity*. The problem posed by Maynard Smith in both papers (1988, 1991) was "How did natural selection bring about the transition from one stage to another, since at each transition, selection for 'selfishness' between entities would tend to counteract the change . . . how is it that selection at the lower level does not disrupt integration at the higher level?" (1988, 222–223). It is this question that my colleagues and I have been trying to answer, using a combination of mathematical models and a model experimental system, the volvocine green alga.

Transfer of Fitness and ETIs

I have referred to Maynard Smith's original list as involving evolutionary transitions in individuality (ETIs; see Michod 1999). Let us first distinguish between ETIs and ETs in the sense of major events in the history of life. There are a number of immensely important events without which life as we know it would be vastly different, including such major events as the origin of the genetic code, language, oxidative photosynthesis, and the Cambrian Explosion. Understanding these events is critical for the field of evolutionary biology and for understanding life on earth. These events are not, however, ETIs. ETIs comprise

a common set of problems and solutions involving levels of selection and the integration of evolutionary units. Samir Okasha, at the KLI workshop, suggested ETIs constitute a natural kind, a natural grouping of phenomena involving common problems and sharing common solutions.

In addition to the familiar levels of selection, the list of ETs offered by Maynard Smith and Szathmáry (1995, 6) include, sex, the genetic code, and language; language was included because of its importance to cultural evolution and inheritance, and sex because members of a sexual population cannot reproduce without a mate. For many workers, sex and language are the odd members of the list. I argue here that sex belongs on the list, because the mating pair is the new level of selection, a new kind of individual as far as fitness is concerned. I am not so sure about the genetic code and language; I won't say anything more about them.

What are the essential properties of an ETI in the original levels-of-selection sense of Maynard Smith? During a transition from a lower to a higher level, we expect the fitness of the higher level to increase and the fitness of the lower level to decrease. Fitness may be defined as expected reproductive success at either the cell or group level. This expectation is illustrated in the panels in figure 9.1, which give the output of two completely different kinds of mathematical models of the origin of multicellular individuals from groups of cells. Panel A gives the results of a two-locus population genetic modifier model of the evolution of conflict mediators. Conflict mediators are genetically encoded properties that reduce the opportunity for within-group selection and/or enhance the opportunity for between-group selection. Panel B gives the results of an optimization model of the evolution of division of labor among cells when selection is at the cell-group level. These two models are vastly different, and are intended to study different aspects of ETIs' yet both models illustrate an ETI—the decrease in fitness at the lower (cell) level and the increase in heritable fitness of the new higher (multicellular group) level.

Cooperation among cells is central to evolutionary transitions in individuality and to the results shown in figure 9.1. Cooperation may be costly, as in the case of altruism, or not costly, as in the case of synergism (see the discussion below of sculling and rowing games taken from Maynard Smith and Szathmáry 1995). As altruism evolves, the costs of altruism reduce the fitness of cells and the benefits of altruism increase fitness of the cell groups. Therefore, it is expected that conflict mediators that increase the opportunity for altruism increase the fitness at the group level and reduce the fitness at the cell level (figure 9.2A).

What may not be expected is that the relative fitness at the group level compared to the cell level also increases, when conflict mediators evolve in response to noncostly or synergistic forms of cooperation. In synergistic forms of cooperation, there is no cost at the lower level, and the fitnesses of both levels increase during an ETI. However, the fitness at the group level increases more than the fitness at the cell level (figure 9.2B). Conflict modifiers in these models increase by virtue of being associated with more fit genotypes

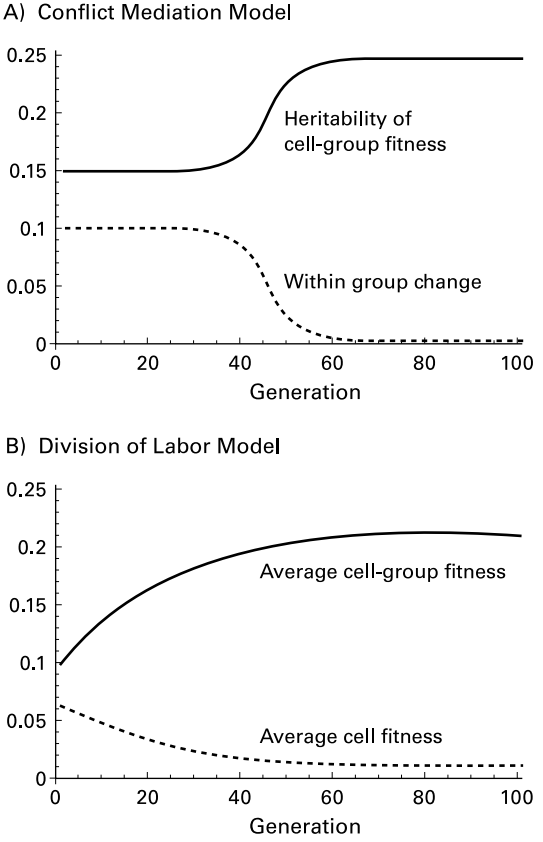


Figure 9.1
Transfer of fitness during an ETI. Panel A is from a conflict mediation two-locus population genetics model (Michod and Roze 1997), and panel B is from a life-history division of labor model (Michod 2006).

and by increasing the heritability of fitness of these types. When cooperation is maintained in a population in the face of deleterious mutation to noncooperation or defection, cooperating zygotes must be more fit than defecting zygotes, because the fitness of cooperating zygotes must compensate for directional mutation toward defection. Deleterious mutation to noncooperation reduces the functionality and fitness of the cell group. Modifiers increase by virtue of increasing the heritability of fitness of the already more fit cooperating genotype and by hitchhiking along with these more fit types. The transfer of fitness from the cell to group level during an ETI illustrated in figures 9.1 and 9.2 is not a built-in assumption of the models nor is it an interpretation of the models; rather, it is a consequence of the dynamics of the model.

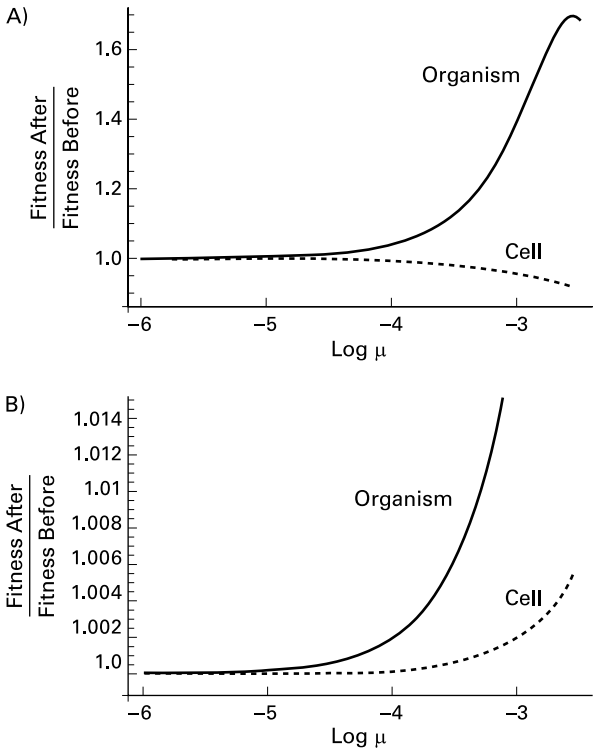


Figure 9.2
Ratio of fitness after an ETI to before, as a function of the deleterious mutation rate per cell division during development. Fitness ratios are plotted for the two levels of selection: the cell and the cell group or organism. Panel A is for altruistic cooperation and panel B is for synergistic forms of cooperation. Adapted from figures 13 and 14 of Michod and Roze (1999), which gives the details of the two locus conflict mediator model and analysis. A brief introduction to these models and results is given in the text.

The origin of fitness differences at a new level of organization (e.g., the cell group) is the central project of ETIs. In the words of Griesemer (2000, 70), "Selection can occur at a level only if there are entities at that level which are capable of being units of selection, i.e. the kinds of things that can have variance in fitness. It is the project of a theory of evolutionary transition to explain the evolutionary origin of entities with such capacity."

Specialization in Fitness Components

I have argued that, in addition to the transfer of fitness to the group level, a necessary component of an ETI is specialization of group members in the fitness components, reproduction and survival, of the group (Michod 2005, 2006). Why is division of labor at reproduction and survival fundamental to individuality and ETIs? When cells completely specialize at one of the two basic fitness components, reproduction or viability, they lose

their overall fitness and capacity to function as evolutionary individuals in their own right. As a consequence, the fitness of the group is no longer the average of the fitness of the cells. By virtue of their specialization, cells have low cellular fitness, whereas the fitness of the group may be quite high (see figure 9.1).

Maynard Smith and Szathmáry put the matter of reproductive specialization and ETIs this way (1995, 4): "entities that were capable of independent replication before the transition can replicate only as part of a larger whole after it." As Griesemer points out (2000, 26), this criterion for an ETI implies division of labor and specialization of some of the lower-level entities at reproduction. Consequently, it is through the reorganization of fitness (fitness transfer from lower to higher level and specialization of group members in the fitness components of the higher level) that the group becomes indivisible and hence a new individual.

What are the factors that lead to fitness reorganization? We have modeled two kinds of selective processes—conflict mediation (Michod 1996, 1997, 1999, 2003; Michod and Nedelcu 2003a; Michod, Nedelcu, and Roze 2003; Michod and Roze 1999, 2001) and division of labor (Michod 2006; Michod et al. 2006)—and applied these models to the origin of multicellularity in the volvocine green algae lineage (Herron and Michod 2008; Michod 2007a, 2007b; Michod and Nedelcu 2003a; Michod, Nedelcu, and Roze 2003; Nedelcu and Michod 2003a, 2006; Shelton and Michod 2010). Under certain conditions, both models result in the transfer of fitness between levels, but they differ in their reasons for specialization of the members of the group. In both kinds of models, specialization arises first through the evolution of cooperative interactions in the group. The evolution of cooperation is the central problem of social evolution. Cooperation is also fundamental to ETIs, because, as already mentioned, altruism and other forms of cooperation lead to the transfer of fitness from the lower level (the costs of altruism) to the group level (the benefits of altruism). Thus, the evolution of cooperation is the first stage in an ETI. In the following sections, we discuss the evolution of altruism in the volvocine green algae.

Conflict Mediation and Reproduction of the Group

Development is interpreted as the conversion of a propagule into an adult cell group through repeated cell division, in such a way that the adult group acquires the capacity to reproduce itself (Griesemer 2000). We have modeled this process using two locus population genetic modifier methods (Michod 1999). In our models, groups are created from propagules. Propagules may contain any number of cells sampled from an adult group or from several adult groups, as in the case of aggregation. In addition, sex may occur in the case of single-celled propagules that fuse with propagules from other groups to start a new group. The capacity of a group to reproduce itself may be measured by the degree to which a group created by a propagule resembles the group the propagule came from. Alternatively, since the group is made from a propagule, and the recurrence equations are in terms of the gene and genotype frequencies at the propagule stage, we may also measure the

capacity for reproduction as the degree to which the propagules produced by a group are similar to the propagule(s) that created the group.

The first locus in our models has two alleles, C and D, which express cooperation and defection, respectively, among cells. During development to make the adult form, there is the possibility of mutation from C to D at each cell division (back mutation is ignored, as there are many more ways to lose a functional trait like cooperation than to create it). These deleterious mutations disrupt the functioning of the adult cell group and increase the variance and opportunity for selection at the cell level. After the adult form is made, a propagule is formed. In our models, the propagule may be produced sexually or asexually, it may be composed of a single cell or multiple cells (if produced vegetatively), or it may be produced by aggregation of cells (Michod and Roze 2001). Depending on the parameters of development, which include the mutation rate, cell replication and death rates, number of cell divisions, the costs and benefits of cooperation, and the mode of propagule formation, a polymorphism may be maintained at the C-D locus by mutation selection balance. This polymorphism sets the stage for the evolution of conflict mediation. Conflict mediation affects the degree to which the propagule produced by an adult has the same properties as the propagule that founded the adult. In other words, the evolution of conflict mediators addresses the central issue of how groups acquire the capacity to reproduce themselves (Griesemer 2000).

The way in which development affects the reproduction of the group involves many factors; in our models, these factors involve specializations of cells that mediate the potential for the conflict inherent in the cooperation and defection behaviors. Examples of conflict mediation include germ-soma specialization and cell policing, both of which reduce the potential for conflict at the cell level and increase the heritability of fitness at the cell group level. Why do these conflict modifiers during development evolve, and how do they lead to the capacity of a group to reproduce itself?

The evolution of cooperation at the C-D locus implies the evolution of cells specializing at group beneficial functions (the benefits of cooperation). Further specialization is achieved through the evolution of alleles at a second "conflict mediator" modifier locus (M/m), which affects the parameters of development, opportunity for defection, and/or means and mode of propagule formation (and hence reproduction of the group). By changing these parameters, the modifier allele affects the between- and within-group variance and opportunity for selection at the two levels, and in this way may create the capacity of the group to reproduce itself (Griesemer 2000).

Under certain conditions, modifiers evolve (by hitchhiking with the more fit C allele at the polymorphic C-D locus) that have the effect of increasing the between-group variance and decreasing the within-group variance, thereby increasing the level of cooperation and the fitness and reproduction of the group. An example of this process has been plotted in figures 9.1A and 9.2. The details of the model are described elsewhere (Michod and Roze 1997). Examples of conflict modifiers we have studied include a germline, reduced muta-

tion rate, policing, programmed cell death, passing the life cycle through a single-cell zygote stage, and fixed group size. It is in this way that conflict mediation during development may enhance the capacity of group to reproduce itself.

Fitness Trade-offs

As already mentioned, fitness involves two basic components: viability and reproduction. An evolutionary unit needs to have both effectiveness at each of these components and a balance between them. This is difficult because energy, resources, and effort expended at one component often detract from the other component, resulting in trade-offs among fitness components. Fitness trade-offs drive the evolution of diverse life-history traits in extant organisms (Roff 2002; Stearns 1992). These same fitness trade-offs gain special significance during ETIs for several related reasons, which we discuss now in regard to the evolution of multicellularity (Michod 2006, 2007b; Michod et al. 2006).

1. Fitness trade-offs are significant to ETIs because they provide a basis for the origin of cooperative and altruistic behavior among cells (e.g., reproductive altruism or soma) within the cell group (Michod 1999, 2006; Michod and Roze 1999; Nedelcu and Michod 2006). For example, an important component of viability in the volvocine algae discussed later is flagellar motility, but reproduction and cell division interfere with flagellar motility (Koufopanou 1994; Solari, Kessler, and Michod 2006). In the unicellular members of this lineage, selection will presumably optimize the allocation of time and energy to these two processes, but, in a group, cells that spend more time flagellated divide less frequently than cells that spend less time flagellated. As flagellar action of cells benefits group motility, cells with a greater propensity to remain actively flagellated are altruistic relative to cells that spend less time flagellated (because the latter cells reproduce more).

2. The curvature of the trade-off between survival and reproduction (i.e., the way in which changing survival/viability affects reproduction) is known to be a central issue in life-history evolution. In the case of the origin of multicellularity, if the fitness trade-off is of convex curvature, evolution will result in cells specialized for either reproductive or survival-related functions of the group; that is, germ-soma, or G-S, specialization will be the optimal group strategy and will evolve (Michod et al. 2006). This is similar in effect to the well-known principle in life-history theory that convex fitness trade-offs select for specialization in time at viability and reproduction—that is, "big bang" or semelparous reproduction, in which organisms put all their effort into viability, maintenance and, growth until just before the end of life, when all effort is put into reproduction (Michod 1978; Schaffer 1974).

3. Fitness trade-offs can enhance the fitness of the group through a covariance effect, by which group fitness is augmented beyond the average fitness of cells according to the covariance of cellular contributions at viability and fecundity (Michod 2006; see equation 9.1 below).

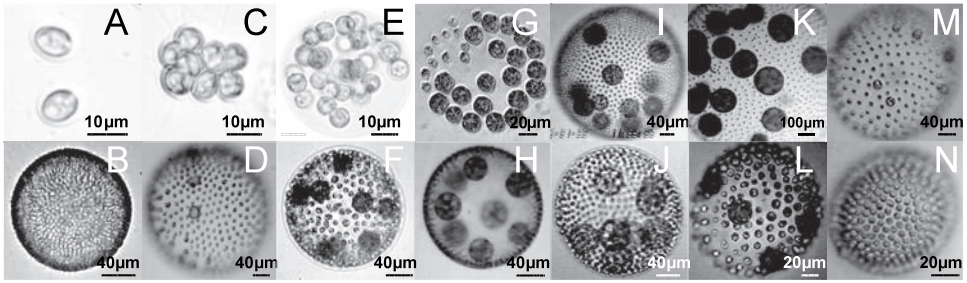


Figure 9.3
Volvocine green algae showing differences in cell number, body size, and degree and mode of G-S specialization (contrast enhanced). All cells in all panels are haploid. In the species where two cell types can be identified, the smaller cells are sterile and somatic and the larger are reproductive. (A) *Chlamydomonas reinhardtii*, (B) *Volvox rousseletii*, (C) *Gonium pectorale*, (D) *Volvox aureus*, (E) *Eudorina elegans*, (F) *Volvox tertius*, (G) *Pleodorina starrii*, (H) *Volvox obversus*; (I–N) are all *V. carteri*: (I) grown at high light, (J) at lower light showing dec. in fecundity, (K) mature adult, (L) *regA*$^-$ mutant, (M) *lag*$^-$ mutant, (N) *gls*$^-$/*regA*$^-$ mutant. Picture credits: C. Solari, M. Herron.

These three processes underlie the reorganization of fitness, by which I mean the transfer of fitness from the old lower-level individual to the new higher level and the specialization of lower-level units in fitness components of the new individual (Michod 2005, 2006). The outcome of this process has been illustrated in figure 9.1B, for a model described it more detail below (Michod 2006).

Multicellularity and Individuality in Volvocine Green Algae

Introduction to the Volvocine Green Algae

For the major multicellular lineages, the factors underlying their origins and evolution of mode of reproduction lay hidden deep in their evolutionary past, obscured by hundreds of millions of years of subsequent evolution. The volvocine green algae (figure 9.3) provide a unique window into the origin of multicellularity, division of labor, and the evolution of sexual reproduction.

The volvocine green algae are biflagellated, photosynthetic, facultatively sexual, predominately haploid (a diploid zygote is formed during sex) eukaryotes, comprising both unicellular species (in *Chlamydomonas*, shown in figure 9.3A, and *Vitreochlamys*) and colonial forms, with varying degrees of complexity involving differences in colony size, structure, and degrees of germ-soma specialization (figure 9.3). Among the colonial forms, organization can be as simple as clumps of four *Chlamydomonas*-like cells that fail to separate after cytokinesis (*Basichlamys*, *Tetrabaena*). Colonial forms comprising 8 to 32 undifferentiated cells can be organized as flat or slightly curved sheets in a single layer (*Gonium*; figure 9.3C), as spherical colonies (*Pandorina*, *Volvulina*, *Eudorina*, in

figure 9.3E, and *Yamagishiella*), or as a flat double layer of undifferentiated cells (i.e., a flattened sphere; *Platydorina*). Larger colonies, consisting of 32 to 128 cells, are all spherical and may exhibit both undifferentiated (*Chlamydomonas*-like) and terminally differentiated (somatic) cells (*Pleodorina*, in figure 9.3G, and *Astrephomene*). The largest forms are spherical and consist mostly of somatic cells and a much smaller number of specialized, unflagellated germ cells (various *Volvox* species and forms, figure 9.3I–N).

Several other model systems are being used to investigate the origins of multicellularity, including choanoflagellates (King and Carroll 2001), cellular slime molds (Foster et al. 2002; Queller 2003; Strassmann, Zhu, and Queller 2000), and myxobacteria (Shimkets 1990; Velicer, Kroos, and Lenski 2000). The volvocine algae exhibit a number of features that make them especially suitable for studying the transition from unicellular to multicellular life as well as the transition to sexual reproduction. (i) Volvocine algae exhibit a diverse array of multicellular forms (see figure 9.3). (ii) They also exhibit a bewildering array of sexual forms and systems (including homothallic and heterothallic, monoecious and dioecious, differing degrees of sexual dimorphism such as anisogamy, isogamy, oogamy, and sexual induction, via a sexual pheromone or not) (Zeiger, Gollapudi, and Spencer 2005). (iii) They can easily be obtained from nature and maintained in the lab under realistic conditions that allow for an ecophysiological and ecodevelopmental framework. Uni- and multicellular forms coexist in transient, quiet bodies of water and in large, eutrophic lakes (during early summer blooms). (iv) Many aspects of their biology have been studied (cytology, biochemistry, development, genetics, physiology, natural history, ecology, and life history) (Kirk 1998). (v) Unlike in the cellular slime molds and myxobacteria, cell groups develop from a single cell, so the cells in the group are related. This aspect of the life cycle is basic to the questions we investigate and is shared by the more complex multicellular forms we wish to understand. (vi) The genomes of the two most representative members—the unicellular *C. reinhardtii* and the multicellular *V. carteri*—have been sequenced and several genes involved in the ETI have been identified.

Evolution of Altruism

The first stage in both conflict mediation and division of labor models (figure 9.1) is the evolution of altruism. The evolution of reproductive altruism, that is, the evolution of soma, involves the evolution of cells with increased effort at functions that enhance the viability of the group at the expense of the reproduction of the cell. We have presented evidence that reproductive altruism in the multicellular green alga *V. carteri* (figure 9.3I–N) evolved via the cooption of a life-history gene whose expression in the unicellular ancestor was conditioned on an environmental cue (as an adaptive strategy to enhance survival at an immediate cost to reproduction) by shifting its expression from a temporal (environmentally induced) to a spatial context (developmental), as summarized in figure 9.4 (Nedelcu and Michod 2006). This example is perhaps the only example of a social gene

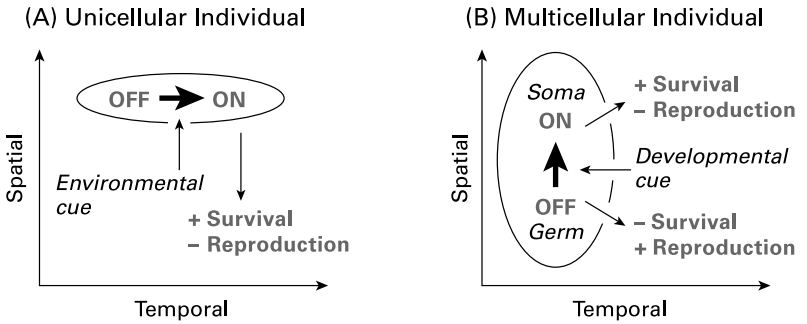


Figure 9.4
Change in expression of a life-history gene in space and time. Expression of genes is indicated by thick arrows. The effect on fitness when the gene is ON and OFF specified. (A) In a unicellular individual, the gene is expressed in response to an environmental cue in a temporal context and has the effect of increasing survival while decreasing effort at reproduction. (B) This same gene is expressed in a spatial context within a multicellular individual in response to a developmental cue. The cells in which the gene is expressed increase their effort at survival and decrease effort at reproduction. Adapted from Nedelcu and Michod (2006).

specifically associated with reproductive altruism, whose origin can be traced back to a solitary ancestor.

Volvox carteri consists of approximately 2,000 permanently biflagellated somatic cells and up to 16 nonflagellated reproductive cells (figure 9.3I–N). Terminal differentiation of somatic cells in *V. carteri* involves the expression of *regA*, a master regulatory gene that encodes a transcriptional repressor (Kirk et al. 1999) thought to suppress several nuclear genes coding for chloroplast proteins (Meissner et al. 1999). Consequently, cell growth (dependent on photosynthesis) and division (dependent on cell growth) of somatic cells are suppressed. Because they cannot divide, somatic cells do not produce offspring and remain flagellated, thus contributing to the survival and reproduction of the colony through flagellar action (Short et al. 2006; Solari et al. 2006; Solari, Kessler, and Michod 2006). In other words, the somatic cells express an altruistic behavior, and *regA*, whose expression is necessary and sufficient for this behavior (Kirk et al. 1999), is an altruistic gene. Which cells express *regA* and differentiate into somatic cells is determined early in development through a series of asymmetric cell divisions. The asymmetric divisions ensure that some cells (i.e., the germline precursors) remain above the threshold cell size associated with the expression of *regA* (Kirk 1995).

As with all forms of cooperation, this altruistic behavior is also susceptible to defection and selfish mutants; indeed, mutations in *regA* result in the somatic cells regaining reproductive abilities, which in turn results in them losing their flagellar capabilities (Kirk et al. 1987). figure 9.3L shows a *regA* mutant colony before dedifferentiation of the somatic cells. Soon, after dedifferentiation, the smaller cells in figure 9.3L begin growing, and the whole colony becomes a large reproductive mass. Since motility is important for these

algae (flagellar activity is required to maintain an optimum position in the water column relative to sunlight intensity), the fitness of these mutant colonies is negatively affected (Solari, Kessler, and Michod 2006).

How can an altruistic gene such as *regA* originate, and can its evolutionary origin be traced back to the unicellular ancestor of this group (similarly to *C. reinhardtii* in figure 9.3A)? To address the possibility that *V. carteri regA* evolved from a gene already present in a unicellular ancestor, we searched the *C. reinhardtii* genome for sequences with similarity to this gene. We found several sequences with similarity to *regA* (indicating the presence of a gene family) and identified the member that is most closely related to the *V. carteri regA* (Nedelcu and Michod 2006). As *V. carteri regA*'s expression is associated with the suppression of chloroplast biogenesis (Meissner et al. 1999), we reasoned that the *regA*-like gene in *C. reinhardtii* is expressed when photosynthesis should be down-regulated. To simulate such conditions, we grew *C. reinhardtii* in the dark, and investigated the expression of both *regA* and a nuclear gene coding for a chloroplast protein—thought to be a RegA target in *V. carteri* (Meissner et al. 1999). As predicted, we found that *regA* is specifically induced in the dark, and its expression coincides with the down-regulation of the chloroplast protein. Thus, our result that the *C. reinhardtii regA*-like gene is induced in the dark, an environment in which chloroplast biogenesis is restricted and the reproductive rate is significantly reduced, suggests that the *regA*-like gene in *V. carteri*'s unicellular ancestor was likely induced under environmental conditions when the temporary down-regulation of chloroplasts was beneficial in terms of survival though costly in terms of immediate reproduction. In other words, our results are consistent with one of the themes of this chapter which is that altruism may evolve from genes involved in life-history trade-offs.

Division of Labor

Having explained *how* a gene for reproductive altruism may originate, that is, by cooption of an existing life-history gene, let us return to *why* germ-soma specialization may evolve, that is, what are the selective factors favoring specialization at reproductive and vegetative functions? Using a optimality modeling approach based on the life-history concept of reproductive effort (Michod 1978; Schaffer 1974), we studied the evolution of specialization at reproduction and viability of cells belonging to cell groups (Michod 2006; Michod et al. 2006). This modeling approach assumes selection occurs only at the cell group level. This is likely the case in the volvocine algae, due to their mode of colony formation: All the cells in a colony are derived mitotically from a single cell. Hence, the cells in a colony are highly related genetically. Even though this mode of colony formation and, hence, high degree of genetic relatedness among cells in the group, holds for all volvocine species, reproductive altruism and germ-soma specialization evolves only in the larger members of the lineage which have more cells (see figure 9.3).

The phenotype of cells, in our model, is described by their effort at reproduction (fecundity), with the remainder of effort put into the viability. The viability, V, and fecundity, B,

of the group are assumed to be the arithmetic averages of the cell efforts at the two fitness components, viability and fecundity, v, and, b, respectively, or $V = \sum_i v_i/N$, and $B = \sum_i b_i/N$. In addition to the assumption of additivity, this formulation implies a kind of isomorphism between fitness components at the two levels, because we are assuming that the activities of the cell at cellular viability and cellular fecundity contribute, respectively, to group viability and group fecundity. Related to this issue is the assumption that the two fitness components of the group, viability and fecundity, are first composed separately from cell properties, and then combined (multiplicatively) to generate the fitness of the group, W (taken as the product of V and B). Without this assumption, evolution of specialization at activities that trade off with one another at the lower level would not be possible. If cells specialized in one activity or the other, then the fitness of specialized cells would be zero and so would be the group fitness, if group fitness was composed directly out of cell fitnesses (as opposed to being composed out of cell fitness components). In this way, the group may break through the trade-off constraints imposed at the cell level. Finally, we assume that the total fitness at either level is the product of viability and fecundity, as is appropriate for organisms with discrete generations. We further discuss these assumptions later with regard to the volvocine green algae.

A central result of the model is the group covariance effect given in equation 9.1, which shows that the fitness of the cell group, W (taken as the product of V and B), is greater than the average fitness of member cells, $\overline{w} = \sum_i v_i b_i/N$, by an amount equal to the negative covariance of the fitness components at the cell level (viability, v, and fecundity, b).

$$W = VB = \overline{w} - Cov[v, b] \tag{9.1}$$

If the covariance between fitness components is itself negative, as it is when fitness components trade off with one another, there is an enhanced fitness at the group level from what would be expected were group fitness composed as the average fitness of cells. The covariance effect given equation 9.1 translates the negative covariance of fitness components of group members into a benefit at the group level. Alternatively, if fitness components were to covary positively, fitness of the group would decrease from that expected by the average cell fitness. High fitness for any unit at any level of organization requires a balance of fitness components at that level, because the components are multiplied together to give total fitness (in the case of discrete generations). The covariance effect translates a lack of balance at the lower cell level into an advantage at the group level, especially under conditions of convexity of the trade-offs. Convexity of the trade-off curvature allows for enhanced effectiveness at each fitness component for cells that specialize.

The particular mathematical representation of the covariance effect given in equation 9.1 depends on additivity of effects on the viability and reproduction components of fitness as described earlier. Additivity of fitness effects is the simplest assumption possible; nevertheless, the assumption of additivity of the contributions of cells to the viability of the group may be relaxed and the general points still hold (Michod et al. 2006). It may be reasonable

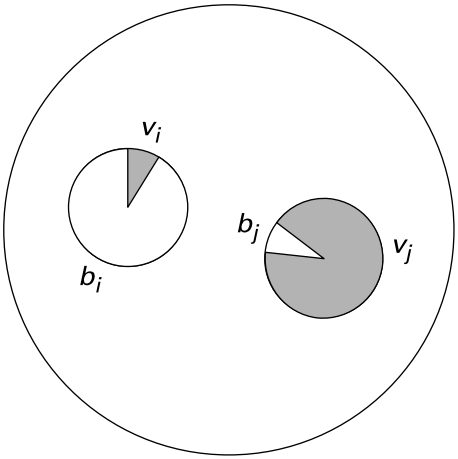


Figure 9.5
Two cells specializing in different fitness components, reproduction (white) and viability (gray). Cell *i* specializes in reproduction, with reproductive effort b_i , with less effort put into viability functions, v_i. Cell *j* does the reverse. Alone, they would each have low fitness because they are unbalanced and high fitness requires a balance at the two components. However, together, they may constitute a good team and bring high fitness to the group if the trade-off between reproduction and viability is convex.

to assume additivity in reproductive efforts. As illustrated in figure 9.5, what is required is if one cell has a high reproductive effort (and hence a low viability and a low cell fitness), this may be compensated for by another cell with high viability (and hence a low fecundity and also a low cell fitness) (Michod et al. 2006). Consequently, even though each of these cells by themselves would have a low fitness, together they can bring a high fitness to the group, especially under conditions of convexity of the trade-off. This kind of joint effect, whereby multiple cells may contribute more to the group than could each alone, does not require additivity (Michod et al. 2006) and would not be possible if group fitness were simply assumed to be the average of the cell fitnesses. In effect, such oppositely specialized cells complement one another and constitute a good and integrated "team" under conditions of convexity of the trade-off curve. This kind of joint effect is a first step toward integration of the group.

That these effects can lead to specialization of cells at the components of fitness of the group may be illustrated by beginning with a random uniform distribution of cells and sampling cells into groups of, for example, six cells, and allowing the output of the group to the cell pool to be given by group fitness, taken to be the product *VB*. In the computer simulation, each generation cells are sampled from the cell pool to make groups, and the process is iterated over time. Sampling cells in this way is a poor representation of development; most organisms are formed by mitotic division from a single cell, not by random sampling from a cell pool. Nevertheless, the simulation illustrates the main results of the

model, which is described in detail elsewhere (Michod 2006). An example of the results has already been given in figure 9.1B, which shows how the fitness of the group increases and the average fitness of the cells declines, as the cells within groups specialize at either viability or reproduction (in about equal proportions).

Germ-Soma Specialization in Volvocine Algae

Let us consider the applicability to the volvocine algae of the division of labor model of multicellularity described earlier (equation 9.1 and figure 9.1B). Two related assumptions of this model require further clarification with respect to the algae. First, the viability- and reproduction-enhancing activities of the cell contribute to similar capacities at the group level. Second, the fitness components of the group are composed separately (from cell properties) and then combined (multiplicatively) to generate the fitness of the group.

Let us note, again, that volvocine groups are created from a single cell that divides mitotically to produce the cells in the group. The assumption of many group selection models (Wade 1978) of the MLS-1 variety (see following text) that groups are formed by sampling from a migrant or propagule pool (to which many parent groups contribute) is not relevant to this lineage, nor is it relevant to many of the multicellular lineages we wish to understand. Along with others (Bonner 2000), I believe a general way to form multicellular groups is simply by the products of mitotic cell divisions sticking together and not separating. In such cases, the members of the group are highly related genetically, being clonal descendents of a single cell. Furthermore, the cells in a group are a result of a developmental process in which the efforts at activities underlying fitness may be jointly adjusted as required by the model discussed in figure 9.5. This is critically important, for without the possibility of such joint (and integrated) adjustments, cells could not specialize in activities that are assumed to interfere and trade off with one another. The cells would be constrained by the assumed cellular trade-offs. However, by virtue of belonging to a group, the cell as a group member may break through the constraints that govern the lives of single cells.

The common and ancestral mode of reproductive cell division in the volvocine lineage involves "multiple fission"; a reproductive cell grows 2^k-fold before dividing k times to produce a group of $N = 2^k$ cells. This multiple fission aspect of volvocine reproduction is not generally applicable, indeed, even certain derived species of *Volvox* have evolved a different developmental mode based on the more common binary fission means of cell division (for example, *V. rouselettii* in figure 9.3B). The important point, I think, is that a colony comes from a single reproductive cell. In addition, for all volvocine colonies, the colony's fecundity is the number of reproductive cells it contains. As required in the model discussed earlier, the total effort of cells at reproduction, $\sum_i b_i$, is the fecundity the group receives, B.

Viability is more complicated, but still the algae generally fit the model described above. Flagellar action is critical for viability in the volvocine algae. Motility is especially important for survival to avoid sinking and to reach light and nutrients (Solari, Kessler, and Mi-

chod 2006). In lakes, these algae perform daily vertical migrations in the water column to access resources that are heterogeneously distributed in space (surface/bottom) and time (day/night) (Sommer and Giliwicz 1986). Flagellar action is not only crucial for motility (Solari 2005), but it also facilities effective transport of nutrients and waste for the colony by mixing the water surrounding the colony (Short et al. 2006; Solari et al. 2006). The total flagellar force of the colony, say Z, is a cumulative function of the flagellar forces generated by single cells, say $\sum_i z_i$, although it is probably not exactly additive.

Though it may be natural to view the group property of flagellar force as a simple function of cell efforts at flagellar action, it is not clear how exactly to compose the function, V, that expresses viability of the group as a function of group flagellar action, $V = V(Z, E)$. This function would necessarily involve the environment, E, as the contribution of flagellar action to viability would depend on the local medium, for example, whether the medium is well stirred or calm. Alternatively, we need not begin with the viability of the group, but rather view group viability as the average of the viability of its member cells and the motility of the group as a contextual property (Heisler and Damuth 1987; Okasha 2006) of the cell's viability, writing $v_i = v(Z, E)$, with $V = \sum_i v_i$.

However viability is construed, the important condition with regard to the evolution of specialization is that the components of fitness be composed separately; otherwise, the kind of joint effect hypothesized in figure 9.5 cannot be realized. It is biologically correct, in terms of the life cycle of these algae, to compose the group flagellar force as a function of the cellular flagellar forces before the cells within the colony begin dividing to reproduce the colony. Since these algae have discrete generations, fitness is naturally the product of viability and reproduction, VB.

Individuality

The evolution of multicellular organisms is the premier example of the integration of lower-level individuals (cells) into a new, higher-level individual. The word "individual" comes from the Latin word *individuus* meaning "not divisible." Individuals are wholes and cannot be divided into smaller parts that maintain critical properties of the whole; the critical property of interest here is the capacity for reproduction and continued evolution. In philosophy, individuals are defined in terms of distinctness in both space and time. In biology, the criteria of genetic homogeneity, genetic uniqueness, and physiological autonomy and unity provide for different concepts of individuality (Santelices 1999). These different notions of individuality have merits and shortcomings. Clonally reproducing organisms (such as bacteria, many protists and fungi, and some animals and plants), organisms with high levels of within-organism change resulting from somatic mutation and selection (such as long lived plants), and highly social organisms (such as wasps and insects) pose counter examples to the criteria of genetic uniqueness, genetic homogeneity, and physiological autonomy, respectively, as necessary defining characteristics of individuality. These entities seem to be individuals, yet they fail to satisfy one of the criteria.

An evolutionary perspective may help resolve some of these difficulties. Individuals must be units of selection and so possess the Darwinian properties of heritable variation in fitness (Lewontin 1970). Individuals, when made up of lower level units as in the case of multicellular organisms, possess properties that restrict within-group selection and enhance between group selection. We have tried to understand the processes by which evolutionary individuals are created during ETIs, and these results may be used to further clarify the notion of individuality. According to the models described here and illustrated in figure 9.1, an ETI involves the reorganization of fitness, by which we mean the transfer of fitness from the lower level units to the group and the specialization of group members (who were once individuals) in the fitness properties of the group. Once the group contains members specialized in necessary fitness components, the members no longer possess all of the Darwinian properties required to evolve on their own and so are no longer evolutionary individuals.

How does the concept of individuality apply to the volvocine green algae? All members of the lineage share certain properties; especially relevant is the fact that all cells within a colony in all species are derived from a single cell. Although mutations undoubtedly occur during DNA replication, as reproductive cells grow and divide, the cells in colonies are clonally descended from a single cell and genetically related. For this reason we may expect lower levels of genetic variation between cells within the colonies and higher levels of variation between colonies. From the perspective of Griesemer's three stages of group reproduction during ETIs (2000), clonal cell groups are somewhere between stage 1 (the origin of a new level, the group) and stage 2 (a group with mechanisms of conflict mediation in place). The mechanism of conflict mediation in this case is reproducing through a single cell (Michod and Roze 2000; Roze and Michod 2001; Wolpert and Szathmáry 2002). Even with single-cell reproduction, significant levels of conflict may exist in clonal cell groups, depending on the generation time, mutation rate, and rate of division of selfish cells, and this may set the stage for the further evolution of other conflict modifiers such as a germline and cell policing (Michod 1996, 1997, 1998a,b; Michod, Nedelcu, and Roze 2003; Michod and Roze 1999). The single-cell mode of group formation also means that groups are produced from a developmental process that allows for the cells within colonies to be integrated. Even in the simplest eight- or sixteen-celled species, *Gonium* (figure 9.3G), the orientation of the flagella and eyespots depend on spatial location within the colony. The colony is a primary level of selection in this lineage. However, not all levels of selection are evolutionary individuals. Selection occurs simultaneously at multiple levels, and, if conflict mediators are not in place, lower levels of selection may take hold and destroy the higher level. Individuality refers to the capacity of the group to resist selection at lower levels as well as the capacity to be distinct from other such groups.

Reproductive specialization of certain cells in the capacity to produce the group is a key component of individuality of the group. As already mentioned, germ-soma specialization is present in the larger members of the lineage, from *Pleodorina* (64 to 128 cells, figure

9.3G) to and including all *Volvox* species, though to varying degrees. For example, in some *Volvox* species, such as *V. rouselettii* (figure 9.3B), germ cells have functional flagella for a period of time before they differentiate as germ, while in other species, such as *V. carteri* (figure 9.3C), germ cells are completely specialized and never have flagella (Kirk 1998). In addition, some species of *Volvox* maintain cytoplasmic bridges in the adult stage.

The relation between group fitness and cell fitness is fundamental to the distinction between the two kinds of multilevel selection (MLS), termed MLS-1 and MLS-2 (Damuth and Heisler 1988; Okasha 2004, 2006). Under MLS-1, the fitness of the group is the average fitness of the cells, whereas under MLS-2, group fitness is no longer proportional to average cell fitness. Under MLS-1, the relevant ancestor descendent lineages are at the cell level, whereas under MLS-2 the relevant lineages occur at the group level: Groups reproduce other groups. Under MLS-1, the focus of interest to evolutionary change is the level of the cell—the group is relevant as a context for cells—whereas under MLS-2, the focus of interest to evolutionary change is the group. The evolution of individuality can be viewed as the transition between MLS-1 and MLS-2 (Okasha 2004, 2006).

Let us consider a specific example, *Gonium* (figure 9.3C), and see how the MLS-1 and MLS-2 criteria apply (see also Shelton and Michod 2006). All of the cells in a *Gonium* colony are first flagellated and continue to grow until they lose their flagella and divide, to produce a daughter colony containing the same number of cells as the parent colony. There is no specialization at reproductive or vegetative functions. Nevertheless, because of their clonal mode of group formation, *Gonium* colonies have some of the properties of MLS-2. In particular, *Gonium* colonies reproduce other colonies and the relevant evolutionary change and interest is at the colony level. In addition, there is partial integration of cells at flagellar functions, as already mentioned, the flagella and eyespots are oriented differently depending on the location of the cell in the colony. A *Gonium* colony is a level of selection and adaptation, but with few means of conflict mediation other than reproducing through a single-cell stage.

The main point detracting from *Gonium*'s individuality would be its lack of cell specialization at reproduction. As already mentioned, Griesemer (2000) pointed out that Maynard Smith and Szathmáry's definition of an evolutionary transition (Maynard Smith and Szathmáry 1995, 227) implies some division of labor and specialization of cells at reproduction of the group. Without such division of labor, there is no real distinction between cell fitness and the fitness of the group, because all cells reproduce daughter groups; the fitness of the group is simply the average fitness of the member cells. For this reason, *Gonium* fitness follows the MLS-1 formulation. In addition, it is hard to imagine what cheating would look like in *Gonium*. Spending less time or energy on flagellar action is a likely candidate; however, this would likely cost the cell as well as the group. The orientation of the flagella in a *Gonium* colony are more akin to rowing games than to sculling games (Maynard Smith and Szathmáry 1995). In rowing games, members specialize by rowing on opposite sides, so if one member of the boat cheats and rows less, the boat goes in circles and everybody pays.

In contrast, in sculling games, members do not specialize, and instead every member paddles on both sides of the boat. In sculling, if one of the member of a crew cheats and paddles less, the boat will still move forward (although more slowly). The evolutionary transition to *Gonium* is more akin to panel B in figure 9.2 than to panel A. The fitness of both cells and groups increases during the transition.

Individuality is clearly a continuum, with partially integrated units of evolution and adaptation like *Gonium* being partway but not yet fully emerged as an evolutionary individual.

Sex and Individuality

Sex as an ETI

Understanding the evolution of sex remains one of the great problems in biology; however, no conceptual framework has yet been achieved that embraces the many different perspectives (Barton and Charlesworth 1998; Bell 1982; Birky 1993; Maynard Smith 1978; Michod 1995; Michod and Levin 1988; Otto 2003; Peters and Otto 2003), and few experimental systems exist which can address this problem. The problem of explaining the evolution of sex is that sex is common in all the major groups of life and hence should have clear fitness benefits over asex. However, what is most clear about sex is that it is costly to fitness, and there is not wide agreement on what benefits might offset these costs. In bacteria and viruses, sex (genetic exchange) is not associated with reproduction as it is in more complex organisms. Whether associated with reproduction or not, sex requires the coordination of two individuals, and, so implies the existence of a higher-level unit, the reproductive pair. With obligate sexual reproduction (in contrast with facultative sex), individual organisms cannot reproduce without a partner, and furthermore, reproductive success depends on the properties of both partners (likely in a complex and interactive way). Consequently, fitness is a property of the reproductive pair, not of individual organisms. In this sense, the reproductive pair is the real evolutionary individual in obligate sexual species; it is for this reason that the evolution of sex has been viewed as an ETI (Maynard Smith and Szathmáry 1995. 6), and studying sex as an ETI may help integrate the different perspectives on the evolution of sex.

Though I argue that viewing sex as an ETI may help integrate different perspectives on the problem of the evolution of sex, there are several senses in which the evolution of sex is not an ETI in the same sense as, say, multicellularity. First, as Samir Okasha pointed out to me, in the case of sex, the new unit, the mating pair, does not participate in higher-level transitions; in other words, the mating pair does not appear as a nested unit in the hierarchy of life (or any other hierarchy, such as a hierarchy of reproductive systems) the way cells or multicellular organisms are. Second, as Kim Sterelny pointed out to me, there is no pair-dependent heritability of mating pair fitness with sex. In a sexual population, there is not necessarily a correlation between the fitness of a mating pair and the fitness of the repro-

ductive pairs derived from the mating. Thus, though it is clear that the mating pair or group is a new unit of fitness, it is also clear that this fitness unit does not participate in evolutionary transitions as, say, cell groups do.

When discussing the transition from asexual to sexual reproduction, there are several stages to consider, with obligate sexual reproduction at one end of a spectrum of sexual systems. As already mentioned, most prokaryotes (bacteria and viruses) have sex in the sense defined below, of recombination with outcrossing, but for them sex is not associated with reproduction (which is clonal). Eukaryotes have facultative sex, in which case organisms reproduce sexually or asexually and the transition between the two forms of reproduction occurs in real time in the same population. This is the case for the volvocine algae.

During an evolutionary transition of a group into an individual, four general stages can be identified (Michod and Nedelcu 2003a): *initiation* and *stabilization* of the interactions among group members in time and space, functional *integration* of the group members, and *emergence* of fitness heritability of the group. Concerning the evolution of sexual reproduction among mating pairs, this framework raises two questions: (1) What was the impetus for the initial interactions among sexual partners? and (2) How did this interaction evolve into an new evolutionary unit, the obligate mating pair of sexually reproducing organisms?

As with any interaction, the initial interactions among sexual partners may be cooperative or conflictual. Based on our studies of the other ETIs, we expect that the transition to full sexual reproduction involved a series of cooperation-conflict and conflict mediation cycles (Michod 1999; Michod and Nedelcu 2003a, b). We believe that viewing sex as an ETI in the cooperation-conflict framework can help integrate different perspectives on the evolution of sex. An example is given later for the different theories for sex based on its role of coping with mutation, damage, or within-group change. These selective factors are just different kinds of genetic error (conflict) that sex serves to mediate.

What is sex? We consider sex to involve two basic components: recombination and outcrossing. By "recombination" we mean exchange of genetic information (usually by physical breakage and rejoining), and by "outcrossing" we mean that the DNA molecules involved in recombination come from different parents. In the diploid life cycle, recombination occurs during meiosis to make haploid gametes, followed by fusion (outcrossing) to restore the dominant diploid stage. In the haploid life cycle—for example, in the volvocine green algae discussed below—fusion (outcrossing) occurs among haploid gametes to produce a diploid stage, and recombination occurs during meiosis when the diploid stage germinates to produce the dominant haploid stage. We have hypothesized that recombination evolved for the function of DNA repair (Bernstein et al. 1984, 1985; Michod 1995), and the evidence that recombination is an adaptation for DNA repair is substantial (Bernstein and Bernstein 1991; Bernstein, Hopf, and Michod 1988; Birdsell and Wills 2003; Cox 1991, 2001; Michod 1995).

The problem of explaining the outcrossing aspects of sex is much more challenging, because the evolution of outcrossing is connected with the evolution of haploid and diploid life cycles and of reproductive systems generally (Hopf, Michod, and Sanderson 1988; Mable and Otto 1998). From the point of view of the advantages of DNA repair, in haploid life cycles, outcrossing (through fusion of haploid cells) is necessary to produce the diploid stage within which homologous recombinational repair can occur. For diploid life cycles, however, two genomes are always present in most cells, so outcrossing is not necessary to create the diploid stage. We have proposed that masking and complementation of deleterious recessive or partially recessive mutations is an important force maintaining outcrossing (Bernstein et al. 1985; Hopf, Michod, and Sanderson 1988; Michod and Gayley 1992, 1994) and the conditions under which this selective force operates have been clarified (Roze and Michod 2010). A variety of other theories emphasize the capacity of sex to help cope with deleterious mutation, in both finite and infinite populations, through the effects of both segregation and recombination (Agrawal 2006; Barton and Charlesworth 1998). Since the focus here is on haploid organisms, we will not consider further the challenging problem of explaining the evolution of outcrossing in diploids.

In modeling the DNA repair hypothesis for the origin of sex in haploids using explicit population dynamical models, we assumed the advantage of sex was recovery of gene-damaged cells (Long and Michod 1995; Michod 1998b; Michod and Long 1995). Several different costs and benefits of haploidy and diploidy were considered. We also studied several forms of interaction among sexual partners, in particular, "cooperative" sex (in which cells mate even if undamaged) and "selfish" sex (in which cells mate only if they are damaged). We found, as expected from the cooperation conflict framework, that cooperative sex is unstable to the increase of selfish sex (Michod 1998b).

A question we have not yet addressed in our models is how the conflict created by selfish sex may be mediated to lead to stabilization, integration, and the emergence of fitness heritability of a reproductive pair. Although sex creates the opportunity for conflict, we found that it also can serve to mediate conflict and thereby increase fitness heritability (figure 9.6). This result is similar to the role of sex in coping with genetic errors such as mutation and DNA damages. Using a Price analysis of a between- and within-group selection model, we studied the regression of cell group fitness on propagule gene frequency as a function of increasing deleterious selfish mutation rate (Price 1970). Some results are given in figure 9.6, which show how sex maintains higher fitness heritability under conditions of increasing conflict generated by selfish mutation and within-group change during the origin of multicellularity. Selfish mutants are mutants that are advantageous at the cell level but disadvantageous at the group level. Sex organizes variability and heritability of fitness of the new emerging unit in a way not possible in asexual populations. Viewed from the cooperation-conflict framework, the result in figure 9.6 is not surprising. As already mentioned, from the point of view of the group, within-group change is a kind of error, and

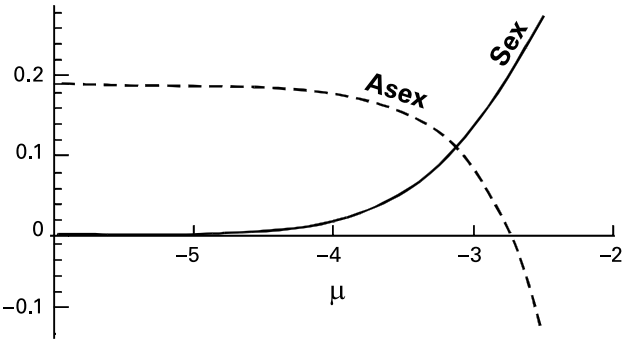


Figure 9.6
Effect of sex on heritability of fitness for increasing selfish mutation rate. The advantage of selfishness at the cell level is 5 percent and the advantage of cooperation at the group level is 2. Generation time (proportional to cell group size) is thirty cell divisions. Intermediate dominance of mutations. Two-locus conflict modifier model of the type described in the text (Michod 1999, appendix A).

sex has been shown to help cope with error from a variety of sources, DNA damage (Bernstein, Byers, and Michod 1981), mutation (Kondrashov 1988; Lynch et al. 1993; Muller 1932), and, in figure 9.6, within-group change.

The main point of this section is the transition from asexual to sexual populations may be viewed as an ETI, which allows us to see different theories about the evolution of sex in a common framework involving common elements. It remains to be understood what other similarities and differences exist between the asexual to sexual transition and the other transitions. We have already discussed several differences. In addition, we have just begun to explore the other stages of the asex to sex transition in an ETI framework. We continue with the problem of the evolution of sex in the next section, applying the cooperation/conflict framework to the volvocine green algae as a model experimental system.

Sex in *Volvox*

Sex in the volvocine algae is facultative in a predominately haploid life cycle. This means that differentiation into the sexual phase along with gamete production occurs in the haploid phase without meiosis. Mating of gametes produces a diploid zygote, which is dormant until the conditions are right for germination, at which time it undergoes meiosis to produce the haploid phase of the life cycle. In nature, while environmental conditions are favorable, haploid volvocine algae proliferate asexually in soil, temporary pools, freshwater lakes, or ponds. As conditions deteriorate (e.g., the pond dries up or nitrogen concentration in the soil declines), they become sexual and produce haploid gametes that fuse to form a resistant diploid zygote, which enters a period of dormancy of unpredictable length. When favorable conditions return, the zygote undergoes meiosis to produce haploid cells that divide mitotically to form asexual haploid individuals.

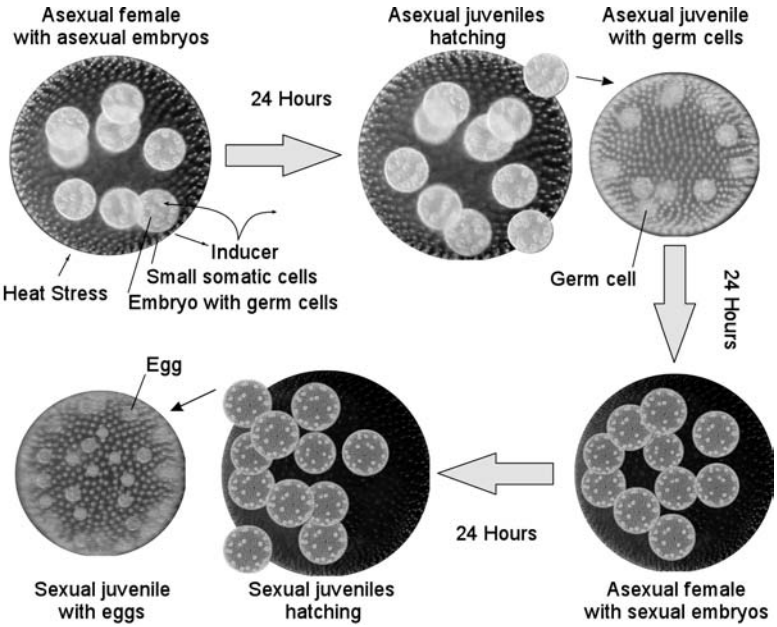


Figure 9.7
The initiation of the sexual phase in the life cycle of a *V. carteri* female following heat stress. During the asexual phase, *V. carteri* females consist of 2,000 small flagellated somatic cells and up to 16 large reproductive germ cells. In response to environmental stress, the somatic cells produce and release a sexual inducer (SI). The inducer acts on the germ cells and alters their developmental pathway such that in the next generation sexual females with up to 45 smaller and denser eggs are produced. All cells in all colonies in this figure are haploid. Not shown: A diploid zygote is formed by fusion of eggs and sperm (sperm comes from a male colony, not shown). Figure assembled from pictures provided by C. Solari.

Life depends on a delicate balance of reduction/oxidation chemistry. Stress upsets that balance. It has become clear that, although the factors cells perceive as stress are multiple and diverse, all these factors alter the cellular redox balance toward the formation of oxidizing molecules ($O_2.^-$, HO., and H_2O_2) called reactive oxygen species or ROS (e.g., Mittler 2002; Nedelcu, Marcu, and Michod 2004; Nedelcu and Michod 2003b). It is well known that ROS damages DNA. Recently, we (Nedelcu, Marcu, and Michod 2004; Nedelcu and Michod 2003b) hypothesized that the mechanistic connection between stress and sex in facultatively sexual lineages involves ROS, possibly reflecting the ancestral role of sex as an adaptive response to DNA damage (Bernstein et al. 1984). We explored this hypothesis in the facultatively sexual *V. carteri* (figure 9.7).

Facultatively sexual eukaryotes provide a unique perspective; for them, sex is not necessary for reproduction, and the transition from the asexual stage to the sexual stage occurs in real time. The impetus for sex is stress (see figure 9.7). Why should stress induce sex in facultatively sexual species?

In response to environmental stress—for example, heat—the somatic cells of *V. carteri* produce and release a sexual inducer (figure 9.7). The inducer acts on the germ cells and alters their developmental pathway such that in the next generation sexual females with up to forty-five smaller and denser eggs are produced instead of asexual germ cells (figure 9.7). The sexual inducer is also produced by the so-called spontaneous sexual males (Kirk and Kirk 1986), which occur spontaneously in culture. Results (Nedelcu 2005; Nedelcu, Marcu, and Michod 2004; Nedelcu and Michod 2003b) (i) show that the ability of both heat stress and SI to induce sex in *V. carteri* is drastically reduced in the presence of antioxidants; (ii) show that stress genes have been coopted into the sexual process, (iii) suggest that sex, cell cycle arrest, and programmed cell death (PCD) are alternative responses to stress; and (iv) suggest that the transition to sex involves DNA damage. Obtaining conclusive evidence for or against the role of DNA damage in the evolution of sex in *V. carteri* is the goal of our ongoing work. It is already known that DNA-damaging agents such as glutaraldehyde (Loshon et al. 1999), formaldehyde (Starr and Jaenicke 1988), and ultraviolet (UV) light induce sex in *V. carteri* (Adams et al. 1990; Coleman 1979; Darden 1968; Darden and Sayers 1969; Desnitski 2000, 2002; Starr 1968; Starr and Zeikus 1993).

How can sex help cope with DNA damage? In haploid facultatively sexual organisms, we hypothesize that sex is the most adaptive way to create a stress-resistant diploid spore (as opposed to an asexual haploid spore) because of its contribution (in terms of both mechanism and timing) to the eventual repair of stress-induced DNA damage. Specifically, the fusion of gametes creates the diploid state and diploidy provides for the genetic redundancy that allows for recombinational repair of DNA damages during germination, at the end of dormancy and the start of a new generation. We hypothesize that during dormancy, often associated with unpredictable and stressful environments, the level of DNA damage increases. This requires effective DNA repair when good conditions return so that offspring start life with a healthy genome. We are currently testing these hypotheses.

If the impetus for sex is the need to deal with stress-induced DNA damage, two scenarios can be envisioned in the cooperation conflict framework discussed previously (Long and Michod 1995; Michod 1998b; Michod and Long 1995): Either both partners are damaged (and thus sex competent), or only one partner is damaged (and thus sex competence must be induced in the other partner). The former scenario involves a cooperative interaction (as the benefit is reciprocal) and makes sense in the context of DNA damage induced by environmental stress, as many organisms in the population will likely be damaged. In contrast, the latter scenario describes a conflictual selfish interaction (as the interaction is beneficial for one but costly for the other) and assumes metabolically induced DNA damage in certain individuals (Michod 1998b). The occurrence of spontaneous sexual forms in *V. carteri* (Starr 1969) may be an example of selfish sex. In addition, we have found that, not only does the mechanism of production of the sexual inducer involve oxidative stress, but also

oxidative stress is involved in the mechanism of action of the sexual inducer in the partner (Nedelcu and Michod 2003b). In other words, the induction of sex in a partner involves a kind of manipulation, in which stress is induced in a partner presumably so that the partner becomes sex competent. This manipulation is a new kind of conflict that must be mediated on the path to obligate sex. Thus, we see in the volvocine green algae several cooperation, conflict, and conflict mediation cycles, during the transition from both asexual to sexual reproduction and unicellular organisms to multicellular individuals (Herron and Michod 2008).

Conclusion

During evolutionary transitions in individuality, new major levels of organization are created. Groups of previously existing individuals become new individuals. How and why this occurs is the major focus of the research discussed here. ETIs do not comprise all of life's major events; rather, they consist of a natural set of common problems and solutions involving transitions among levels of fitness and organization. During an ETI, fitness must be reorganized because the individuals that were the focus of evolution and adaptation before the ETI become components of a new higher-level individual after the ETI. This reorganization of fitness involves two major processes. First, fitness is increased at the new group level and reduced at the lower level. Second, the previously existing individuals, which are now members of the higher-level group, specialize in the fitness components (reproduction and viability) of the higher-level group. Using the examples of multicellularity and the evolution of sex, and as illustrated by the volvocine green algae, we have argued that the reorganization of fitness during these ETIs involves a number of cycles of cooperation, conflict, and conflict mediation.

The kinds of conflict, conflict mediation, and cooperation discussed in this chapter are quite varied. Examples of conflict involve trade-offs between fitness components, trade-offs between fitness effects at different levels of selection, stress, genetic error, sex, and sexual manipulation. Conflict mediation can involve a variety of group and developmental characteristics of the group including kinship, germ-soma differentiation, sex, determinate growth, programmed cell death, and other aspects of development such as the immune system or policing. Most fundamentally, conflict mediation makes group reproduction possible so that groups are able to reproduce similar groups with heritable properties. Cooperation in multicellular lineages involves the evolution of soma. As a repair system, sex may be cooperative or selfish. In the volvocine algae, we argued that sex is likely selfish. Examples of cooperation in the volvocine algae are the extracellular matrix and flagellar action. Conflict through its mediation allows for more cooperation. This cycle of cooperation, conflict, and conflict mediation drives evolutionary transitions in individuality.

Acknowledgments

Thanks to Samir Okasha, Kim Sterelny, and Deborah Shelton for their thoughtful comments on the manuscript.

References

Adams, C. R., K. A. Stamer, J. K. Miller, J. G. McNally, M. M. Kirk, and D. L. Kirk. (1990). Patterns of organellar and nuclear inheritance among progeny of two geographically isolated strains of *Volvox carteri. Current Genetics* 18: 141–153.

Agrawal, A. F. (2006). Evolution of sex: Why do organisms shuffle their genotypes? *Current Biology* 16: R696–R704.

Barton, N. H., and B. Charlesworth. (1998). Why sex and recombination. *Science* 281: 1986–1989.

Bell, G. (1982). *The Masterpiece of Nature: The Evolution and Genetics of Sexuality*. Berkeley: University of California Press.

Bernstein, C., and H. Bernstein. (1991). *Aging, Sex and DNA Repair*. San Diego: Academic Press.

Bernstein, H., H. C. Byerly, F. A. Hopf, and R. E. Michod. (1984). Origin of sex. *Journal of Theoretical Biology* 110: 323–351.

Bernstein, H., H. C. Byerly, F. Hopf, and R. E. Michod. (1985). Genetic damage, mutation and the evolution of sex. *Science* 229: 1277–1281.

Bernstein, H., G. S. Byers, and R. E. Michod. (1981). Evolution of sexual reproduction: Importance of DNA repair, complementation and variation. *American Naturalist* 117: 537–549.

Bernstein, H., F. A. Hopf, and R. E. Michod. (1988). Is meiotic recombination an adaptation for repairing DNA, producing genetic variation, or both? In R. E. Michod and B. Levin (eds.), *The Evolution of Sex: An Examination of Current Ideas* (pp. 139–160). Sunderland, MA: Sinauer.

Birdsell, J. A., and C. Wills. (2003). The evolutionary origin and maintenance of sexual recombination: a review of contemporary models, In R. J. MacIntyre and M. T. Clegg (eds.), *Evolutionary Biology* (pp. 27–138). New York: Kluwer Academic/Plenum Publishers.

Birky, J. C. W. (1993). American Genetics Society Symposium for the Evolution of Sex. *Journal of Heredity. American Genetics Society Symposium for the Evolution of Sex*, Vol. 84.

Bonner, J. T. (2000). *First Signals: The Evolution of Multicellular Development*. Princeton, NJ: Princeton University Press.

Coleman, A. W. (1979). Sexuality in colonial green flagellates. In M. Levandowsky and S. H. Hutner (eds.), *Biochemistry and Physiology of Protozoa*, 2nd Ed., Vol. 1 (pp. 307–340). New York: Academic Press.

Cox, M. M. (1991). The RecA protein as a recombinational repair system. *Molecular Microbiology* 5: 1295–1299.

Cox, M. M. (2001). Historical overview: Searching for replication help in all of the rec places. *Proceedings of the National Academy of Sciences (USA)* 98: 8173–8180.

Damuth, J., and I. L. Heisler. (1988). Alternative formulations of multilevel selection. *Biology & Philosopy* 3: 407–430.

Darden, W. H. (1968). Production of a male-inducing hormone by a parthenosporic *Volvox aureus*. *Journal of Protozoology* 15: 412–414.

Darden, W. H., and E. R. Sayers. (1969). Parthenospore induction in *Volvox aureus* D5. *Microbios* 2: 171–176.

Desnitski, A. G. (2000). Development and reproduction of two species of the genus *Volvox* in a shallow temporary pool. *Protistology* 1: 195–198.

Desnitski, A. G. (2002). Dormant stages of the green flagellate *Volvox* in a natural habitat. *Russian Journal of Developmental Biology* 33: 107–109.

Foster, K. R., A. Fortunato, J. E. Strassmann, and D. C. Queller. (2002). The costs and benefits of being a chimera. *Proceeding of the Royal Society of London B, Biological Science* 269: 2357–2362.

Griesemer, J. (2000). The units of evolutionary transition. *Selection* 1: 67–80.

Heisler, I. L., and J. Damuth. (1987). A method for analyzing selection in hierarchically structured populations. *The American Naturalist* 130: 582–602.

Herron, M. D., and R. E. Michod. (2008). Evolution of complexity in the volvocine algae: Transitions in individuality through Darwin's eye. *Evolution* 262: 436–451.

Hopf, F. A., R. E. Michod, and M. J. Sanderson. (1988). The effect of reproductive system on mutation load. *Theoretical Population Biology* 33: 243–265.

King, N., and S. B. Carroll. (2001). A receptor tyrosine kinase from choanoflagellates: Molecular insights into early animal evolution. *Proceedings of the National Academy of Sciences USA* 98: 15032–15037.

Kirk, D. L. (1995). Asymmetric division, cell size and germ-soma specification in *Volvox*. *Seminars in Developmental Biology* 6: 369–379.

Kirk, D. L. (1998). *Volvox*: Molecular-genetic origins of multicellularity and cellular differentiation. Cambridge: Cambridge University Press.

Kirk, D. L., G. J. Baran, J. F. Harper, R. J. Huskey, K. S. Huson, and N. Zagris. (1987). Stage-specific hypermutability of the *regA* locus of Volvox, a gene regulating the germ-soma dichotomy. *Cell* 18: 11–24.

Kirk, D. L., and M. M. Kirk. (1986). Heat shock elicits production of sexual inducer in *Volvox*. *Science* 231: 51–54.

Kirk, M. M., K. Stark, S. M. Miller, W. Muller, B. E. Taillon, H. Gruber, et al. (1999). *RegA*, a *Volvox* gene that plays a central role in germ-soma differentiation, encodes a novel regulatory protein. *Development* 126: 639–647.

Kondrashov, A. S. (1988). Deleterious mutations and the evolution of sexual reproduction. *Nature* 336: 435–440.

Koufopanou, V. (1994). The evolution of soma in the Volvocales. *The American Naturalist* 143: 907–931.

Lewontin, R. C. (1970). The units of selection. *Annual Review of Ecology and Systematics* 1: 1–18.

Long, A., and R. E. Michod. (1995). Origin of sex for error repair. I. Sex, diploidy, and haploidy. *Theoretical Population Biology* 47: 18–55.

Loshon, C. A., P. C. Genest, B. Setlow, and P. Setlow. (1999). Formaldehyde kills spores of Bacillus subtilis by DNA damage and small, acid-soluble spore proteins of the alpha/beta-type protect spores against this DNA damage. *Journal of Applied Microbiology* 87: 8–14.

Lynch, M., R. Burger, D. Butcher, and W. Gabriel. (1993). The mutational meltdown in asexual populations. *Journal of Heredity* 84: 339–344.

Mable, B. K., and S. P. Otto. (1998). The evolution of life cycles with haploid and diploid phases. *BioEssays* 20: 453–462.

Maynard Smith, J. (1978). *The Evolution of Sex*. London: Cambridge University Press.

Maynard Smith, J. (1988). Evolutionary progress and levels of selection, in M. H. Nitecki (ed.), *Evolutionary Progress* (pp. 219–230). Chicago: University of Chicago Press.

Maynard Smith, J. (1991). A Darwinian view of symbiosis, in L. Margulis and R. Fester (eds.), *Symbiosis as a Source of Evolutionary Innovation* (pp. 26–39). Cambridge, MA: MIT Press.

Maynard Smith, J. and E. Szathmáry. (1995). *The Major Transitions in Evolution*. San Francisco: W.H. Freeman.

Meissner, M., K. Stark, B. Cresnar, D. L. Kirk, and R. Schmitt. (1999). *Volvox* germline-specific genes that are putative targets of RegA repression encode chloroplast proteins. *Current Genetics* 36: 363–370.

Michod, R. E. (1978). Evolution of life histories in response to age-specific mortality factors. *The American Naturalist* 113: 531–550.

Michod, R. E. (1995). *Eros and Evolution: A Natural Philosophy of Sex.* Reading, MA: Addison-Wesley.

Michod, R. E. (1996). Cooperation and conflict in the evolution of individuality. II. Conflict mediation. *Proceedings of the Royal Society of London, B* 263: 813–822.

Michod, R. E. (1997). Cooperation and conflict in the evolution of individuality. I. Multi-level selection of the organism. *The American Naturalist* 149: 607–645.

Michod, R. E. (1998a). Commentary, evolution of individuality. *Journal of Evolutionary Biology* 11: 225–227.

Michod, R. E. (1998b). Origin of sex for error repair. III. Selfish sex. *Theoretical Population Biology* 53: 60–74.

Michod, R. E. (1999). *Darwinian Dynamics, Evolutionary Transitions in Fitness and Individuality*. Princeton, NJ: Princeton University Press.

Michod, R. E. (2003). Cooperation and conflict mediation during the origin of multicellularity, in P. Hammerstein (ed.), *Genetic and Cultural Evolution of Cooperation* (pp. 261–307). Cambridge, MA: MIT Press.

Michod, R. E. (2005). On the transfer of fitness from the cell to the multicellular organism. *Biology & Philosopy* 20: 967–987.

Michod, R. E. (2006). The group covariance effect and fitness trade-offs during evolutionary transitions. *Proceedings of the National Academy of Sciences USA* 103: 9113–9117.

Michod, R. E. (2007a). Evolution of individuality during the transition from unicellular to multicellular life, in J. C. Avise and F. J. Ayala (eds.), *In the Light of Evolution: Volume 1. Adaptation and Complex Design* (pp. 129–144). Washington, DC: The National Academies Press.

Michod, R. E. (2007b). Evolution of individuality during the transition from unicellular to multicellular life. *Proceedings of the National Academy of Sciences USA* 104: 8613–8618.

Michod, R. E., and T. W. Gayley. (1992). Masking of mutations and the evolution of sex. *The American Naturalist* 139: 706–734.

Michod, R. E., and T. W. Gayley. (1994). Genetic error, heterozygosity and the evolution of the sexual cycle, in M. Kirkpatrick (ed.), *Some Mathematical Questions in Biology: Evolution of Haploid and Diploid Life Cycles*, Vol. 25 (pp. 97–110). Providence, RI: American Mathematical Society.

Michod, R. E., and B. R. Levin. (1988). *Evolution of Sex: An Examination of Current Ideas*. Sunderland, MA: Sinauer.

Michod, R. E., and A. Long. (1995). Origin of sex for error repair. II. Rarity and extreme environments. *Theoretical Population Biology* 47: 56–81.

Michod, R. E., and A. M. Nedelcu. (2003a). Cooperation and conflict during the unicellular-multicellular and prokaryotic-eukaryotic transitions, in A. Moya and E. Font (eds.), *Evolution: From Molecules to Ecosystems* (pp. 195–208). Oxford: Oxford University Press.

Michod, R. E., and A. M. Nedelcu. (2003b). On the reorganization of fitness during evolutionary transitions in individuality. *Integrative and Comparative Biology* 43: 64–73.

Michod, R. E., A. M. Nedelcu, and D. Roze. (2003). Cooperation and conflict in the evolution of individuality IV. Conflict mediation and evolvability in *Volvox carteri*. *BioSystems* 69: 95–114.

Michod, R. E., and D. Roze. (1997). Transitions in individuality. *Proceedings of the Royal Society of London, B* 264: 853–857.

Michod, R. E., and D. Roze. (1999). Cooperation and conflict in the evolution of individuality. III. Transitions in the unit of fitness, in C. L. Nehaniv (ed.), *Mathematical and Computational Biology: Computational Morphogenesis, Hierarchical Complexity, and Digital Evolution*, Vol. 26 (pp. 47–92). Providence, RI: American Mathematical Society.

Michod, R. E., and D. Roze. (2000). Some aspects of reproductive mode and the origin of multicellularity. *Selection* 1: 97–109.

Michod, R. E., and D. Roze. (2001). Cooperation and conflict in the evolution of multicellularity. *Heredity* 81: 1–7.

Michod, R. E., Y. Viossat, C. A. Solari, A. M. Nedelcu, and M. Hurrand. (2006). Life history evolution and the origin of multicellularity. *Journal of Theoretical Bioogy* 239: 257–272.

Mittler, R. (2002). Oxidative stress, antioxidants and stress tolerance. *Trends in Plant Science* 7: 405.

Muller, H. J. (1932). Some genetic aspects of sex. *The American Naturalist* 66: 118–138.

Nedelcu, A. M. (2005). Sex as a response to oxidative stress: Stress genes co-opted for sex. *Proceedings of the Royal Society of London, B, Biological Science* 272: 1935–1940.

Nedelcu, A. M., O. Marcu, and R. E. Michod. (2004). Sex as a response to oxidative stress: A two-fold increase in cellular reactive oxygen species activates sex genes. *Proceedings of the Royal Society of London, B* 271: 1591–1596.

Nedelcu, A. M., and R. E. Michod. (2003a). Evolvability, modularity, and individuality during the transition to multicellularity in volvocalean green algae, in G. Schlosser and G. P. Wagner (eds.), *Modularity in Development and Evolution* (pp. 466–489). Chicago: University of Chicago Press.

Nedelcu, A. M., and R. E. Michod. (2003b). Sex as a response to oxidative stress: The effect of antioxidants on sexual induction in a facultatively sexual lineage. *Proceedings of the Royal Society of London, B* (Suppl.) 270: S136–S139.

Nedelcu, A. M., and R. E. Michod. (2006). The evolutionary origin of an altruistic gene in *Volvox carteri*. *Molecular Biology and Evolution* 8: 1460–1464.

Okasha, S. (2004). Multi-level selection and the major transitions in evolution. *Proceedings of the Philosophy of Science Association*, 19th Biennial Meeting—PSA2004, Austin, Texas.

Okasha, S. (2006). Evolution and the levels of selection. Oxford: Oxford University Press.

Otto, S. P. (2003). The advantages of segregation and the evolution of sex. *Genetics* 164: 1099–1118.

Peters, A. D., and S. P. Otto. (2003). Liberating genetic variance through sex. *BioEssays* 25: 533–537.

Price, G. R. (1970). Selection and covariance. *Nature* 227: 529–531.

Queller, D. C., E. Ponte, S. Bozzaro, and J. E. Strassmann. (2003). Single-gene greenbeard effects in the social amoeba *Dictyostelium discoideum*. *Science* 299: 105–106.

Roff, D. A. (2002). *Life History Evolution*. Sunderland, MA: Sinauer.

Roze, D., and R. E. Michod. (2001). Mutation load, multi-level selection and the evolution of propagule size during the origin of multicellularity. *The American Naturalist* 158: 638–654.

Roze, D., and R. E. Michod. (2010). Deleterious mutations and selection for sex in finite diploid populations. *Genetics* 184: 1095–1112.

Santelices, B. 1999. How many kinds of individual are there? *Trends in Ecology & Evolution* 14: 152–155.

Schaffer, W. M. (1974). Selection for optimal life histories: The effects of age structure. *Ecology* 55: 291–303.

Shelton, D. E., and R. E. Michod. (2010). Philosophical foundations for the hierarchy of life (Book review of *Evolution and the Levels of Selection* by Samir Okasha). *Biology and Philosophy* 25: 391–403.

Shimkets, L. J. (1990). Social and developmental biology of the myxobacteria. *Microbiology Review* 54: 473–501.

Short, M. B., C. A. Solari, S. Ganguly, T. R. Powers, J. O. Kessler, and R. E. Goldstein. (2006). Flows driven by flagella of multicellular organisms enhance long-range molecular transport. *Proceedings of the National Academy of Sciences USA* 103: 8315–8319.

Solari, C. A. (2005). Seminar: Multicellularity and the functional interdependence of motility and molecular transport. Department of Ecology and Evolutionary Biology, University of Arizona.

Solari, C. A., S. Ganguly, J. O. Kessler, R. E. Michod, and R. E. Goldstein. (2006). Multicellularity and the functional interdependence of motility and molecular transport. *Proceedings of the National Academy of Sciences USA* 103: 1353–1358.

Solari, C. A., J. O. Kessler, and R. E. Michod. (2006). A hydrodynamics approach to the evolution of multicellularity: Flagellar motility and germ-soma differentiation in volvocalean green algae. *The American Naturalist* 167: 537–554.

Sommer, U., and Z. M. Giliwicz. (1986). Long-range vertical migration of *Volvox* in tropical Lake Cahora Bassa (Mozambique). *Limmology and Oceanography* 31: 650–653.

Starr, R., and L. Jaenicke. (1988). Sexual induction in *Volvox carteri f. nagariensis* by aldehydes. *Sexual Plant Reproduction* 1: 28–31.

Starr, R. C. (1968). Cellular differentiation in *Volvox*. *Proceedings of the National Academy of Sciences USA* 59: 1082–1088.

Starr, R. C. (1969). Structure, reproduction and differentiation in *Volvox carteri f. nagariensis* Iyengar, strains HK 9&10. *Archiv fur Protistenkunde* 111: 204–211.

Starr, R. C., and J. A. Zeikus. (1993). Utex—the culture collection of algae at the University of Texas at Austin 1993 list of cultures. *Journal of Phycology* 29: 1–106.

Stearns, S. C. (1992). The evolution of life histories. Oxford: Oxford University Press.

Strassmann, J. E., Y. Zhu, and D. C. Queller. (2000). Altruism and social cheating in the social amoeba *Dictyostelium discoideum*. *Nature* 408: 965–967.

Velicer, G. J., L. Kroos, and R. E. Lenski. (2000). Developmental cheating in the social bacterium *Myxococcus xanthus*. *Nature* 404: 598–601.

Wade, M. J. (1978). A critical review of models of group selection. *Quarterly Review of Biology* 53: 101–114.

Wolpert, L., and E. Szathmáry. (2002). Multicellularity: Evolution and the egg. *Nature* 420: 745.

Zeiger, E., B. Gollapudi, and P. Spencer. (2005). Genetic toxicity and carcinogenicity studies of glutaraldehyde—a review. *Mutation Research* 589: 136–151.

10 How Many Levels Are There? How Insights from Evolutionary Transitions in Individuality Help Measure the Hierarchical Complexity of Life

Carl Simpson

How the vast range of spatial and temporal scales on which biological processes interact and relate to each other is a fundamental problem in evolutionary biology. The standard solution was given by Darwin and formed the core of the modern synthesis. Observed processes of organisms interacting with their environment (which includes other organisms) produce all the patterns at all spatial and temporal scales. From this perspective, biology is uniformitarian, patterns at all scales are thought to be caused by the observable local processes. Unfortunately, observations at large scales do not seem to be reducible to local patterns in all cases. The most critical of these observations are those transitions in evolution known as evolutionary transitions in individuality (Buss 1987; Maynard Smith and Szathmáry 1995; Michod 1999). What occurs during these transitions is the passing of the dominant fitness components from one level of organization to a higher, more inclusive level, and with it new levels of organization, evolution, and ecological interaction emerge. That is to say, as the new level of individuality emerges, we find new agents that not only have fitness values, the fitness of these new agents dominates the evolutionary trajectory of them and their components.

From the view of some standard theory, these transitions are impossible. The lowest levels of selection are thought to always dominate because of higher heritability, shorter generation times, and other factors that seem to stack the cards in favor of the lowest levels. Yet, the transition from individuated cellular to well-individuated multicellular life occurred approximately sixteen times in largely unrelated groups (Bonner 2001; Maynard Smith and Szathmáry 1995), and that from individuated multicellular life to partially individuated colonial or social life occurred multiple times in ten animal phyla (Wilson 1975). That may not seem like a lot considering those occur over 4.6 billion years and among countless other origination events, including the origin of life, but though these events are rare, they also seem to be patterned: Often these transitions occur in clusters of independent events like during the Cambrian Explosion. Clearly, these are not just statistical possibilities.

Since the modern synthesis, only conceptual work on the hierarchical nature of life has tried to explicitly incorporate the diversity of processes at the varying spatial and temporal scales in which organisms live and evolve. Curiously, all the hierarchies are rigid, having

only a few discrete levels of fixed rank. And there are often two hierarchies, one for organisms and one for their interactions. There are the replicator/interactor hierarchy (Hull 1980), the replicator/vehicle hierarchy (Dawkins 1982), the genealogical/ecological hierarchy (Eldredge 1985; Eldredge and Greege 1992), the codical/material hierarchy (Williams 1992), and the fraternal/corporate hierarchy (Queller 2000). It is curious, at least when viewed with a desire to unify phenomena, that when hierarchy theory encounters biology, it splits its effort. After all, the level both hierarchies have in common is the organism.

Because the majority of work on hierarchies did not realize the importance of the major transitions, most existing hierarchies are themselves static, unevolving, and synchronic. After Buss (1987), we know that any hierarchy must be diachronic and evolvable. The emergence of new hierarchical levels during a major transition explicitly allows for the hierarchy as a whole to evolve over time (Buss 1987; Maynard Smith and Szathmáry 1995; Michod 1999). But while theories of the major transitions are dynamic, they have not tried to incorporate ecology and evolution, which leaves the dual hierarchies, flawed as they may be, as the only hierarchy that takes ecology seriously.

Buss's notion that individuality is derived can teach us an additional lesson about the emergence of new levels. Individuality is derived, but poorly individuated organisms are still common today, yet they can themselves undergo additional major transitions. The sponges and cnidarians provide the best examples. Though they originated over a period of more than 550 million years, only a few lineages have increased their internal integration, but never as much as a simple bilaterian. Yet, both form aggregations and have many colonial species. Is a sleaze of sponges or a coral colony at the same level as colonial or social groups that evolved from highly individuated organisms such as social insects or mammals? There is no clear answer, but individuality itself cannot be the sole criterion for a new level. The hierarchical levels included in the major transitions and transitions in individuality seem to be only those that are highly derived, where it is easy to distinguish between levels.

These problems with current concepts in hierarchy and the major transitions do not allow us to answer some important questions about the hierarchical history of life: How many levels are there now and how many have there been in the past? Over time, we presume that the number of levels only increases cumulatively from the primitive prokaryotic level, through the eukaryotic, multicellular levels, and finally to the colonial level. But the levels in this list are independently derived many times with no constraint on what level the ancestor was at. The levels on this list also ignore ecological levels. A measure of the hierarchical complexity over time should include all types of levels.

Three phases can be identified in transitions in individuality. The aggregate phase is the least individuated, the group phase intermediately individuated, and the individual phase is the most. Each phase is characterized by a dominant fitness component. Differential expansion is the component associated with aggregates, differential persistence is associated with groups, and differential reproduction is characteristic of paradigm individuals. Evolu-

tionary transitions to more individuated phases require the accumulation of additional fitness components, but new levels are attained once the expansive component of fitness is attained. This allows us to know that organisms in each of the three phases of individuality are at the same level if they share a common ancestor.

The aggregate phase has never been considered important in major transitions, but turns out to be essential in the emergence of a new level of selection. The recognition of the aggregate phase allows for the precise identification of a new level and is general enough to incorperate both ecological and genealogical levels as aggregates of organisms from any number of species.

Transitions in Individuality

The View from Current Theory

Transitions in individuality occur when a lineage of organisms of a particular hierarchical level of organization evolves such high integration among organisms that the aggregations eventually become individuals at a new higher level. An explanation of transitions in individuality must describe the emergence of a new level and the subsequent individuation at that level.

It is generally understood that the major challenge a lineage faces when undergoing a major transition in evolution involves the emergence of fitness at a new level (Michod 1999; Okasha 2006). Traditionally, fitness in this context is taken to be reproductive-output measurable by the number of offspring produced. If a new level of fitness is the number of offspring groups produced by a group, it is easy to see how difficult it would be for selection among groups to operate: The number of descendent groups would be tiny compared to the number of offspring produced by constituent organisms. This seems to limit the efficacy of high levels of selection to very particular circumstances, the most important being a decrease in reproductive effort at the lower level (Wade 1978; and see Rice 1995, for other interesting examples).

There is more to fitness than the production of offspring. At the very least, there is also differential viability, or persistence, since some organisms live longer than others. Sometimes living longer allows the organism to produce more offspring, but more importantly, the frequencies of phenotypes in the future are affected just by the existence of long-lived organisms. Michod and collaborators (Herron and Michod 2008; Michod 2006, 2007; Michod et al. 2006; Michod and Herron 2006; Roze and Michod 2001) developed models showing that a simple trade-off between cell reproduction and viability in Volvocacean algae can contribute to a successful transition in individuality—as is indicated by the presence of a germline in the derived colonial members like *Volvox*.

Fitness in Michod's models is the product of viability and fecundity. The fitness of a single cell may be quite low if it specializes in reproduction or viability at the expense of

the other. Each cell has a life history describing its partition of fitness into reproductive and viability components. The colony as a whole has a large number of cells with variation in their life histories. In Michod's model the fitness of the colony, w, can be greater than the average fitness of cells ($\overline{w}_p$, subscripts are explained in table 10.1) if the covariance between each constituent cell's commitment to growth (v) and reproduction (b) is less than zero. Michod defines colony level fitness as $w = \overline{w}_p - \text{cov}(v, b)$ (Michod 2007).

Michod's model requires that somatic cells tend not to undergo cell division, which in plants and Volvocaceans is what is observed. However, this model does not generalize to animals. At the organismal level, animals have a huge range of somatic cell types that can produce germ cells even after they undergo considerable cell division (Buss 1987; Nieuwkoop and Sutasurya 1981). At the colonial level, clonal growth and sexual reproduction commonly co-occur. Animals have undergone transitions in individuality from cellular to multicellular as well as from multicellular to colonial levels at least fifteen times. It seems that at least half of the examples of transitions in individuality do not satisfy Michod's requirement that $\text{cov}(v, b) < 0$.

In animals, cell division is critical for developmental differentiation and growth. Animals seem to happily ignore the theoretical difficulties with the emergence of a new level of fitness, they are all highly individuated at the organismal level with apparently high fitnesses at the cellular and organismal level. To illustrate the issue, an individual of the cnidarian *Hydra* has 121 cells of 15 types (Bell and Mooers 1997) and a large number of offspring. The dog *Canis familiaris* has $10^{13.7}$ cells and 99 cell types (Bell and Mooers 1997), and on average 42 offspring across 7 litters, assuming 6 puppies a litter. An animal starts its life with a single cell and ends with potentially trillions of cells. Almost simultaneously new offspring can be produced, but the number of successful offspring can be orders of magnitude lower than the numbers of cells. Cell division and organismal reproduction are clearly decoupled.

Since the majority of transitions in individuality occur in animals, first from single cellular to multicellular transitions in the origin of the Metazoa, and subsequently in a number of colonial transitions in a number of animal phyla, we must understand how reproduction can evolve at a new level. A complex trait like reproduction to evolve requires the prior emergence of other components of fitness at the high level.

Expansion Is a Third Component of Fitness

Though it is not widely known, significant conceptual work on levels of selection has been done by Leigh Van Valen since the early 1970s, though with a much different focus than standard multilevel selection theory. As a consequence of trying to understand the implications of the Red Queen's hypothesis, Van Valen (1976) proposed a very general interpretation of fitness; fitness is best understood as the amount of energy an evolutionary entity controls that is available for expansion. This energy could be quantified as the number of reduced

carbon atoms available for oxidation, or the number of calories stored in an organism's tissue. For example, an organism controls some amount of energy from food sources, some of which is used to repair damaged tissues, the remainder can be used either to grow or to produce offspring. The expansive energy is that which can be used for growth or reproduction.

To see how the notion of expansive energy is useful, we can translate it to a more traditional counting-based notion of fitness where fitness is the number of individuals an organism produces. But what is an individual? The problem is that biological individuality corresponds roughly to how countable a type of organism is. Not all organisms are countable. Some, like snails, come in roughly the same size and have a discreet boundary, so that it is easy to understand what we mean by ten snails. But most sessile organisms in the marine benthos and a wide variety of plants are poorly individuated solitary and colonial organisms that can be of almost any size (see Clarke, this volume, for a discussion of plants in the context of transitions). A single large bryozoan colony can easily contain as many zooids as one hundred smaller colonies. What, then, do we mean by ten bryozoan colonies? Harper (1977) proposed counting genetic individuals called genets or alternatively counting physically defined modules called ramets. So ten bryozoan colonies would consist of ten genets, and the number of ramets would be equal to the number of zooids in all the colonies. The numbers of genets and ramets are essentially estimates of expansive energy. If an organism produced no offspring, instead spending all its expansive energy on growth (which includes clonal reproduction), the number of ramets would be directly proportional to expansive energy (in units of calories, for example). Alternatively, all expansive energy could be used for the production of offspring. The number of genets would be directly proportional to the expansive energy.

The expansive energy notion of fitness allowed Van Valen (1976) to distinguish three components to fitness: differential expected expansion (or growth), differential expected persistence (or viability), and differential expected multiplication (or fecundity). All three can change the frequencies of traits in a population over time. They are standard values, so that even if one does not accept the energy notion of fitness itself, the importance of these three components can be understood in standard theory.

The Three Phases of Transitions in Individuality

Conventionally, the fitness of a biological entity is the product of its reproductive output (multiplication) and its viability (persistence) (Michod 2007). But this notion of fitness is inadequate for understanding transitions in individuality, because in the earliest phase, nothing like reproduction in the usual sense occurs. Nor does the most primitive aggregate persist in any obvious way, because they may continuously break apart and form anew. In fact, the traditional fitness components are themselves built at each new level of

individuality (as is individuality itself) (Buss 1987). A more fundamental notion of fitness is required (Van Valen 1976), which includes three components: expansion, persistence, and multiplication. In this more general sense, an entity that expands more than another is more fit. A bamboo that covers a field by sending out clonal runners is more fit than one that consist of only a single shoot. Likewise, a bristlecone pine alive for thousands of years is more fit than another lasting a century.

In the following subsections I use a multilevel expansion of Sean Rice's (2008) stochastic derivation of the Price equation (Frank 1998; Hamilton 1975; Okasha 2006; Price 1972; Rice 2004) to describe multilevel evolution during the three phases of transitions in individuality. Rice's stochastic equation is useful because it treats phenotypes and fitness as random variables, which allows us to describe evolution prior to the origin of reproduction in the new whole, when random fragmentation predominates. Treating both fitness and phenotype as random variables, we can describe how both the phenotype and number of offspring depend on the size of the propagules formed in addition to the phenotype and size of the parent. In other words, the offspring of a parent with a specific phenotype will be very different phenotypes depending on the number and size of offspring produced. This contrasts with the standard Price equation, where the parent can produce only a set number of offspring of a particular type as a function only of its own phenotype.

The form of a hierarchical expansion of the stochastic Price equation is similar to other multilevel expansions (Frank 1998; Hamilton 1975; Okasha 2006; Price 1972; Rice 2004). It is most similar to the version of Arnold and Fristrup (1982) developed for studying species selection, however, because by recognition of expansion, a clear distinction between the multiplication of parts and the multiplication of wholes is possible.

Damuth and Heisler (1988) distinguish between multilevel selection type 1 and type 2, where either multiplication is of members of a whole (MLS 1), or multiplication of the wholes themselves occurs (MLS 2). Although they intended to clarify the differences between species selection and group selection, it has become clear that the key to understanding major transitions is understanding how MLS 1 evolves into MLS 2 (Okasha 2006). An understanding of the evolution of reproduction using the multilevel stochastic "Rice equation" allows us to understand how MLS 1 can transition into MLS 2 (table 10.1).

The basic form of the Price equation tracks the change over time in the mean of a trait in a population ($\Delta\overline{\phi}$). The mean trait value changes due to selection and changes that occur during the process of reproduction, including a lower level of selection. Selection is described by the covariance between fitnesses, w, and traits, ϕ: (cov(w, ϕ)). Changes due to other processes can be summarized by the expected change in traits between offspring and parent ($\phi^o - \phi = \overline{\delta}$), weighted by the fitnesses associated by those traits, E($w\overline{\delta}$). Adding the values of those two terms and scaling them by the average fitness ($\overline{w}$) describes the change in the mean trait value over time. The basic single level Price equation is

Table 10.1
Symbols and notation for the hierarchical expansion of Rice's (2008) stochastic version of the Price equation.

Level	Symbol	Meaning
Whole	N	Population size
	ϕ	Phenotype of a whole
	ϕ^o	Mean phenotype of a whole's offspring
	δ	$\phi^o - \phi$
	$\hat{\delta}$	Expected mean value of δ in the population
	m	Reproductive output of a whole
	v	Persistance (viability) of a whole
	w	Demographic fitness of a whole; equal to mv
	$\overline{w}$	Expected demographic fitness in the current environment
	Ω	$\frac{w}{\overline{w}}$ conditional on $\overline{w} \neq 0$
	E	Expansive fitness of a whole; equals $n_g \overline{w}_p$
Parts	n_g	Number of parts in whole g
	ϕ_p	Phenotype of a part
	ϕ_p^o	Mean phenotype of a part's offspring
	δ_p	$\phi_p^o - \phi_p$
	$\hat{\delta}_p$	Expected mean value of δ_p in the population
	w_p	Demographic fitness of a part
	$\overline{w}_p$	Expected demographic fitness of parts within a whole
	Ω_p	$\frac{w_p}{\overline{w}_p}$ conditional on $\overline{w}_p \neq 0$
	$\overline{w}_{p_t}$	Average fitness of parts across all groups

$$\Delta\overline{\phi} = \frac{1}{\overline{w}}[\mathrm{cov}(w, \phi) + E(w\overline{\delta})] \tag{10.1}$$

Rice's stochastic version has one additional variable, Ω, which is equal to $w/\overline{w}$, where w is the fitness of an individual and $\overline{w}$ is the average fitness of the population; Ω is conditional on $\overline{w} \neq 0$. Variables with a hat, that is, $\hat{\delta}$ and $\hat{\Omega}$, indicate the expected value of the random variable in question. Variables with a bar are the average values of those variables. The stochastic Price equation (Rice 2008) is

$$\Delta\overline{\phi} = \mathrm{cov}(\phi, \hat{\Omega}) + \mathrm{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\mathrm{cov}_i(\delta, \Omega)} + \overline{\delta} \tag{10.2}$$

Equation 10.2 contains two more terms than equation 10.1. The first, $\mathrm{cov}(\hat{\delta}, \hat{\Omega})$, measures the covariance between expected fitness and the expected difference between parent and offspring. This term will be positive if offspring with high expected fitness are consistently different from their parents. The covariance is calculated over the entire population. The

second additional term, $\overline{\text{cov}_i(\delta, \Omega)}$, measures the covariance between the number of offspring a single parent produces and the difference between offspring and parent. This term will be nonzero if the number of offspring an individual produces is related to the phenotypes that are produced. Several biological processes could be described by this term, including "offspring-size/clutch-size tradeoffs" (Charnov and Ernest 2006), and importantly for transitions in individuality, any relationships between propapgule size and offspring phenotype.

The term, $\overline{\delta}$, is the average difference between ancestors and descendents. Many processes can be incorporated into the value of this term, but importantly, evolution at a lower level of selection directly effects $\overline{\delta}$. Since the difference between ancestors and descendents can be taken over any time interval, $\overline{\delta}$ takes the same form as $\Delta\overline{\phi}$, but with selection and other processes occurring at a lower level; $\overline{\delta}$ itself can be described by some form of the Price equation (Arnold and Fristrup 1982; Frank 1998; Hamilton 1975; Okasha 2006; Rice 2004; Simpson 2010). The form underlying the Price equation describing $\overline{\delta}$ depends on the type of group level reproduction that is occurring. In the primitive case, there is no group reproduction per se, but each group (or patch) may have its own inherent rate of population growth. Assuming that selection and mutation are the only sources of change within groups, and that group phenotypes and group growth rates are random variables, $\overline{\delta}$ equals

$$\overline{\delta} = \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p} \tag{10.3}$$

The covariance term measures the effects of selection, while $\overline{\delta_p}$ measures the average difference between parent and offspring members. The variable Ω_p is the ratio between the group-specific growth rate, $\overline{w}_p$, and the growth rate of the whole population of groups, $\overline{w}_{p_t}$.

The expected change between ancestors and descendents ($\hat{\delta}$) is also needed in two terms in the equation 10.2. The importance of this value increases during the later phases of transitions because the expected offspring values depend on the mechanisms that produce offspring. Rice (2008) also provides a form of his equation written as the expected change in mean phenotype ($\widehat{\Delta\overline{\phi}}$), which is incorporated in a hierarchical expansion, by noting that $\widehat{\Delta\overline{\phi}} = \hat{\delta}$ and recursively expanding:

$$\hat{\delta} = \text{cov}(\hat{\phi}_l^o, \widehat{\Omega_p}) + \overline{\text{cov}(\phi_l^o, \Omega_p)} + \overline{\delta_p} \tag{10.4}$$

During evolution through the three phases of transitions, $\hat{\delta}$ becomes increasingly important because its values are deeply coupled with the mechanism of emerging level reproduction.

Substituting equation 3 (but not equation 10.4, for clarity) into equation 10.2 gives the full hierarchical expansion. Where the term $\hat{\delta}$ occurs, equation 10.4 can be substituted. The terms are shifted vertically to visually indicate their level of operation. We can identify selection at the level of wholes, interaction between the behavior of the parts during the reproduction of the wholes (where wholes and parts interact), and evolution among parts. The change in group phenotype over time is given by

whole *wholes and parts interact* *parts*

$$\Delta\bar{\phi} = \mathrm{cov}(\phi, \hat{\Omega}) + \mathrm{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\mathrm{cov}_i(\hat{\delta}, \Omega)} + \mathrm{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p} \quad (10.5)$$

Remember that the form of this multilevel description is constant throughout evolution during the three phases. Each term takes on new values as the mechanism of reproduction of wholes evolves. The first phase begins when organisms aggregate in some way.

Phase I: The Aggregate Phase

Despite the unfamiliarity of expansion as a fitness component, it is a direct reflection of the conventional components of the fitness of the members when viewed from the level of the aggregate or group. Indeed, differential expected expansion of aggregates changes the frequencies of members in a population in just the same way as differential multiplication of the members of the aggregates does; the only difference is the focal level (figure 10.1). The standard theory assumes that the average fitness of members ($\overline{w_p}$) is equal to the fitness of the group (w) (Frank 1998; Okasha 2006; Rice 2004). I find that definition difficult to accept because fragmentation of an aggregation (multiplication at the higher level) is independent of the reproductive output of the members, even in primitive examples. It is more parsimonious to treat the average member fitness, $\overline{w_p}$, as what it is—the population growth rate. In the expansive framework, expansive aggregate-level fitness (E) equals the average member fitness ($\overline{w}$) multiplied by the group size (n_g): $E = n_g\overline{w_p}$. Of course, these two definitions of group-level fitness are mathematically similar. The important aspect to remember is that, by equating the average fitness of members with the expansive fitness of aggregates, we do not assume that the aggregates themselves produce descendent *aggregates*, only that the aggregates themselves change in size or extent.

Okasha (2006) made a similar conceptual distinction following Damuth and Heisler (1988). He identifies two classes of group-level fitnesses: collective fitness and particle fitness. Particle fitness is the number of offspring constituent *particles* a collective aggregate produces, and collective fitness is the number of *collectives* a collective produces (which Okasha calls collective fitness$_1$ and collective fitness$_2$, respectively). During the aggregate phase of transitions, aggregates have only particulate fitness (collective fitness$_1$). Viewed from the focal level of members, the change in frequencies of types can be driven by any of the three fitness components among members (fitnesses at the member level could be a combination of fitness$_2$ and fitness$_1$ from the level below). Moving up, so that the focus is on the aggregate level, selection among aggregates appears to be driven only by the expansive component of fitness, their differential changes in size. At the focal level of wholes,

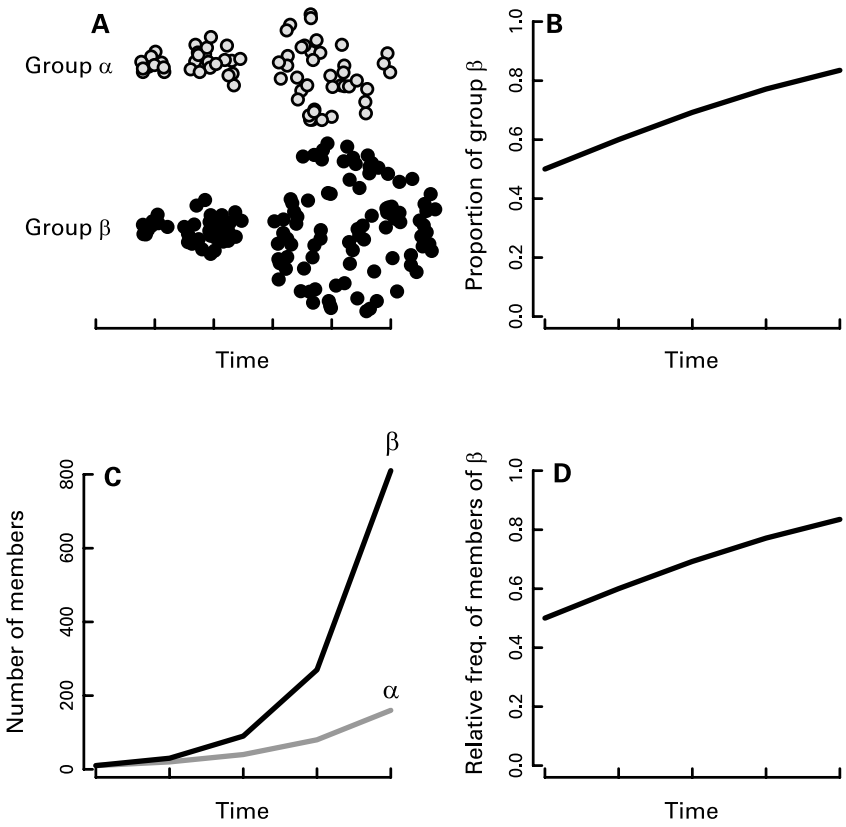


Figure 10.1
The expansive component of fitness is the change in size of units at the focal level. From a hierarchical perspective, differential expansion is caused by differential multiplication of constituent units. Panel A illustrates a hypothetical example of evolution by differential expansion. Group α expands at a lower rate than group β. Over time, there is a greater proportion of group β in the environment, as shown in panel B. The number of members of type α and β is tracked in panel C. The change in frequency of type β members shown in panel D matches the change in proportion of group β in the environment shown in panel B.

the expansive fitness is its change in size, while the demographic fitness of the whole is the differential multiplication and persistence of wholes.

The more traditional demographic fitness of aggregates (collective fitness, or fitness$_2$), w, is the product of the reproductive output (m) of collectives and their persistence (v), so that $w = mv$. In the aggregate phase, aggregate-level reproduction does not occur at all, or at best it is not systematic. If no reproduction occurs, m, the reproductive output at the aggregate level, is equal to zero and w is also equal to zero. There must be some viability, so v can never be equal to zero. If aggregate viability were equal to zero, they would have no staying power, instantly disaggregating. In the aggregate phase, I assume v follows a uni-

form distribution, so there is variation in how long each aggregate can last, but that variation is not correlated with anything. Because $m = 0$, w also equals zero. If there is some degree of fragmentation, I assume it is uncorrelated with the phenotype of the aggregates. By noting that $\text{cov}(x, y) = \beta_{x,y} \text{var}(x)$, we can see that if both the variance in reproductive output is zero (where $\Omega = 0$) and the aggregate phenotype and aggregate reproduction is uncorrelated (e.g., m has a uniform distribution), all three covariance terms involving Ω in equation 10.5 will equal zero because $\beta_{\phi,\Omega} = 0$.

When the first three terms of equation 10.5 that contain Ω, are also equal to zero, we can see that the change in mean group phenotype is governed exclusively by the evolution within groups:

$$\Delta\bar{\phi} = \text{cov}(\phi, \hat{\Omega}) + \text{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\text{cov}_i(\delta, \Omega)} + \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p}$$

$$\Delta\bar{\phi} = 0 + 0 + 0 + \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p} \tag{10.6}$$

$$\Delta\bar{\phi} = \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p}$$

All that is required in the aggregate phase is for membership in an aggregate of other organisms to have an effect on fitness, positive or negative, so that there is a nonzero covariance between the aggregate trait and the fitness of members. There seems to be no limit on the components of the aggregate. Other species or conspecifics could influence the fitness of others in an aggregate. Therefore, the aggregate phase may be quite common in nature. If, in a patch of ground, earthworms till the soil particularly well so that the vegetation grows lush, giving the earthworms more to feed on, all organisms in the patch benefit. If the members of the productive patch then reproduce more than members of other patches, even with random and independent dispersal members of the productive patch will increase over the landscape. In this example, aggregate phenotype is productivity, which all members contribute to in their own way.

Phase II: The Group Phase

It is important to recognize that the fitness components that characterize the three phases are not mutually exclusive. A lineage entering into the group phase does not require natural selection by expansive fitness to be turned off. On the contrary, transitions between the three phases occur by the *accumulation* of new components of fitness.

In the aggregate phase described earlier, the persistence and multiplication components of fitness have no selective traction because their variation is uncoupled from the phenotypes of the aggregates. However, once some aspect of aggregate-level phenotype (e.g., frequencies of types of members) become correlated with persistence or viability, so too does multiplication, and only the causal connection between the multiplication component of fitness and phenotypes is initially absent; the multiplication component sorts (Vrba and

Gould 1986), but it does not select, because phenotypes do not directly cause the number of offspring.

Members of groups can directly influence the viability or persistence of groups in any number of ways. A classic example is the alarm calls in squirrels; despite the high cost to the individual making the call, more group members survive, and therefore the group as a whole persists longer if the number of altruists is high (Sherman 1985). Any arbitrary function of the phenotype $f(\phi)$ could conceivably specify the actual relationship between persistence and phenotype.

Groups themselves do not form offspring per se, but they can fragment and form new groups by fission. Primitive groups must divide by fission if they divide at all, and the details of group fission have consequences for the efficacy of selection. We can understand the potential for evolution in such groups by modeling group reproduction as random sampling without replacement from finite parental colonies. A group reproduces by forming small propagule groups consisting of random subsets of the members.

Consider a group with N members. Beneficial traits occur in this group at a proportion, q, and the number of members in a propagule derived from that group is n, its propagule size. The number of members of that propagule that are beneficial, either altruistic or not, is equal to k/N. In the group are a total of j defectors, occurring at a frequency of $1 - q$. The probability of the offspring consisting of exactly k members of type q, given their proportion in the parent and the size of the propagule follows the hypergeometric probability:

$$Pr(k;n) = f(k;N,m,n) = \frac{\binom{j}{k}\binom{N-j}{n-k}}{\binom{N}{n}} \tag{10.7}$$

Equation 10.7 gives the probability that the resulting propagule has the phenotype k/n, independent of its viability. Figure 10.2 illustrates reproduction by random sampling for the case where $k = n$. The probability of forming a propagule with all members of type k is highest when the propagule size is equal to 1.

If the viability of the propagule (v) is a function of the proportion of benificial members in the propagule (k/n), so that $v = f(k/n)$, and the minimum viability for a propagule occurs if it contains at least k members of the beneficial type in a frequency greater than or equal to a threshold $X \cdot n$. The probability of producing a viable offspring with the phenotype k is

$$Pr(k;n) = \frac{\binom{j}{k}\binom{N-j}{n-k}}{\binom{N}{n}} \cdot f\left(\frac{k}{n}\right) \tag{10.8}$$

The probability of producing a viable offspring of any viable type ($k \geq X \cdot n$) is

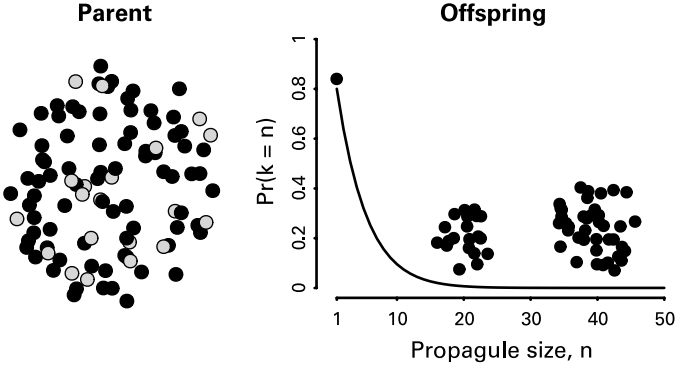


Figure 10.2
Group-level reproduction is modeled as sampling without replacement. Here, I show the probability of producing an offspring where all members of a founding propagule of size n are fixed for a particular trait. The number of propagule members containing the trait is equal to k. The probability of producing offspring that contains only members with the trait is equal to $\Pr(k = n)$. If frequency of the traits in the population is equal to p, then the maximum value of $\Pr(k = n)$ occurs when n equals 1 and is equal to p. In this example, p in the parent equals 0.8; members containing the trait are black. Members that don't are gray.

$$Pr(X \cdot n \le k \le n) = \sum_{k=X \cdot n}^{n} \left[\frac{\binom{j}{k}\binom{N-j}{n-k}}{\binom{N}{n}} \cdot f\left(X \le \frac{k}{n} \le 1 \right) \right] \tag{10.9}$$

Three factors influence the probability of forming a viable offspring: propagule size (n), tolerance for deleterious members (measured by the range of phenotypes that are viable: $X \cdot n \le k \le n$), and the magnitude of the viability function ($f(k/n)$). When there is high tolerance for deleterious members ($X \cdot n$ is much smaller than n) and the variation in viability is low, then the probability of producing a viable offspring is high, no matter what the propagule size (figure 10.3, B and E). When X is large, the probability of producing a viable offspring decreases as propagule size gets larger (figure 10.3, B and E). Things get interesting when the effects of viability are stronger. Figure 10.3 H shows the effects of a monotonic fitness function (figure 10.3 G) on the probability of producing a viable offspring. Larger propagule sizes always lower the probability of producing a viable offspring because the highest viability is found in phenotypes that are uniformly of type k. When propagules have only one member, the chance that a propagule is fixed for k is the highest.

Of the three important parameters, propagule size has the greatest effect on the probability of forming viable offspring. Moreover, propagule size directly influences the total number of possible offspring a group can form. Since propagules are formed by fission, the maximum number of propagules a parent group produces, i, occurs when the propagule size is 1. The number of offspring decreases as a function of the propagule size (n). When

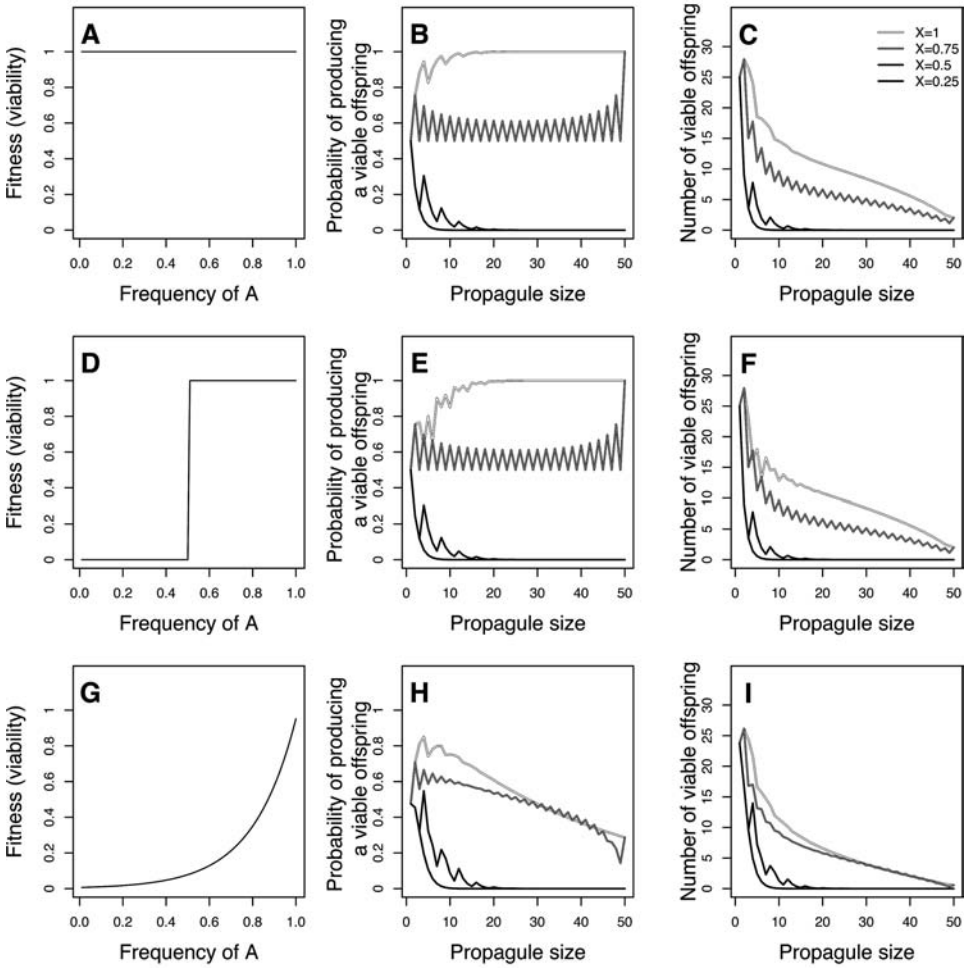


Figure 10.3
The demographic fitness of a group is a function of the viability function, the probability of forming a viable offspring by random sampling of the parent group, and the maximum propagule size of an offspring. Here, I explore various combinations of the three influences on fitness for a parent group consisting of fifty members, half of which are deleterious ($j = 0.5$), and the other half are beneficial. The viability of offspring propagules is a function of the frequency of beneficial members in the propagule and shown in panels A, D, and G. The probability of producing a viable propagule is plotted in the center column (panels B, E, and H), the panel in each row is for the viability function in the same row. The probability of forming a viable offspring depends on the tolerance of deleterious members, which can range from a quarter ($X = 0.25$) to none of the propagule ($X = 1$). The legend in panel C applies to all center and right column panels. Panels C, F, and I show the demographic fitness of parent groups and is the product of the probability of producing a viable offspring and the expected number of offspring with uneven partitioning, given the maximum propagule size.

Table 10.2
The enumerated partitions of a group with five members

5	4	**3**	**3**	2	2	1
	1	**2**	**1**	2	1	1
			1	1	1	1
					1	1
						1

the formation of propagules is random, any size propagule can form; parent groups may produce offspring of a variety of propagule sizes. Parent groups are partitioned into offspring. The number of possible partitions becomes quite large even if the size of the parent group is small. A group of ten members can be partitioned in forty-two different ways. A group with five members can be partitioned into propagules in seven ways, shown in table 10.2.

Maximum propagule size can be considered to be the maximum number of members in a particular partition. In the case in table 10.2, there are two possible partitions for a propagule size of 3: 3,2 and 3,1,1, consisting of two and three offspring, respectively. In this example, when maximum propagule size is 3, the average number of offspring is 2.5.

I calculated the average number of offspring for a group from a direct enumeration of the partitions of a group consisting of fifty members for each propagule size between one and fifty. Alternatively, even partitions can be made of a group, where the group is split into as many propagules as possible of the same size. The number of offspring (i) is given by the number of members, N, and the propagule size, n:

$$i = \frac{N}{n} \tag{10.10}$$

As there are often several ways to partition a group with the same maximum propagule size, the partitioning estimate gives a larger expected number of offspring than an even partitioning of equal-size propagules given by equation 10.10 (figure 10.4).

Propagule size is also important because it directly affects the multiplication component of group-level fitness. The multiplication component of group-level fitness is the actual number of viable offspring produced by a parent group and is determined by the product of equations 9 and 10, assuming offspring with constant propagule size:

$$Pr(X \cdot n \le k \le n) = \sum_{k=X \cdot n}^{n} \left[\frac{\binom{j}{k}\binom{N-j}{n-k}}{\binom{N}{n}} \cdot f\left(X \le \frac{k}{n} \le 1 \right) \right] \tag{10.11}$$

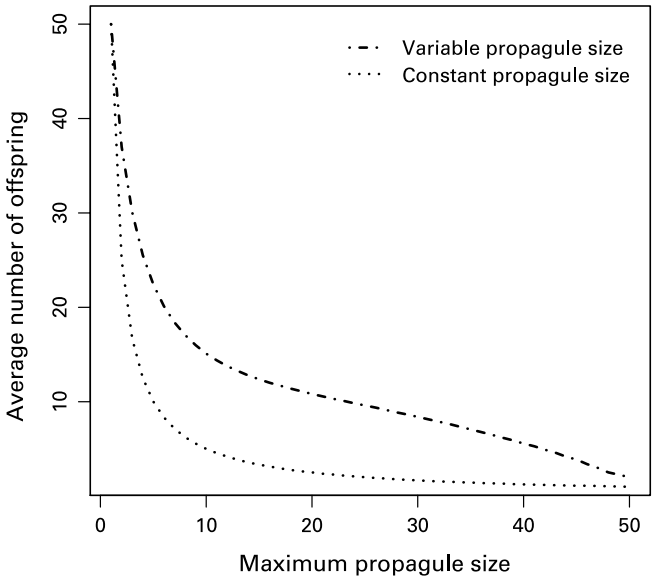


Figure 10.4
Propagules can be formed either by partitioning the parent into propagules with variable numbers of members or by subdividing the parents into propagules with constant numbers. Variable propagule size is achieved by partitioning the offspring into all possible sized propagules, given a maximum propagule size. The maximum propagule size contains the largest number of members. All possible partitions were enumerated for a parent with fifty members. The average number of offspring is calculated from the average number of offspring with the same maximum propagule size. (See table 10.2 and the worked example in the text.) The number of offspring produced, i, when propagule sizes are constant among offspring is given by N/n, where N is the number of members in the parent, and n is the propagule size.

Figure 10.3 (panels C, F, and I) shows the expected group-level fitness as a function of the maximum propagule size when partitioning is variable. As expected, when maximum propagule size is small, the number of offspring is much higher.

Importantly, even if there is no tolerance for deleterious members in a propagule (where $X = 1$), intolerant groups will still have comparable fitness to very tolerant groups. Further, if the partitioning of groups into propagules is actually unspecified, the majority of partitions include a large number of single-member propagules. Since possessing small propagules results in such strong sorting, those traits that are possible to inherit with a single propagule will rapidly increase in frequency.

The model of reproduction outlined earlier shows how group-level reproduction and the change in phenotypes during the reproduction process are tightly coupled, leading to nonzero values in two additional covariance terms in the stochastic Price equation (equation 5). Recall that the term $\mathrm{cov}_i(\hat{\delta}, \Omega)$ describes the average covariance between the number and phenotypes of the offspring of a single parent. Small propagules increase the numbers of offspring, w, so Ω is also affected by propagule size. If a parent group has a phenotype with mixed members, small propagules will have the effect of segregating the members, so

that propagules will tend to consist of only one type. Since propagule size influences both fitness and phenotype, $\text{cov}_i(\hat{\delta}, \Omega)$ will be positive. The among-group covariance between the change over reproduction and fitness, $\text{cov}(\hat{\delta}, \hat{\Omega})$, will also be positive because of variation in propagule size. Since propagule size is initially unspecified, the group phenotype is uncorrelated with number of offspring, the fist term in equation 5, $\text{cov}(\phi, \hat{\Omega})$ consequently equals zero.

Evolution in the group phase, as characterized by equation 5, is influenced by the evolution among members and the interaction between the reproduction of groups and the emerging group-level fitness within and among groups:

$$\Delta\overline{\phi} = \text{cov}(\phi, \hat{\Omega}) + \text{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\text{cov}_i(\delta, \Omega)} + \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p}$$

$$\Delta\overline{\phi} = 0 + \text{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\text{cov}_i(\hat{\delta}, \Omega)} + \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p} \qquad (10.12)$$

$$\Delta\overline{\phi} = \text{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\text{cov}_i(\hat{\delta}, \Omega)} + \text{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p}$$

The two key features of the group phase are differential viability of members with respect to the group and random fragmentation of groups. Both features can be expressed in a large number of ways. Members can either be inviable or cheaters, both of which negatively affect the group as a whole. Random fragmentation is more variable still. There are two ways that propagule membership can form. The simplest is direct fragmentation, so that a single organism can be a member of multiple groups over its life. For example, buffalo herds can temporarily fragment and reassemble with no evolutionary significance, or stromatolite colonies can fragment. Additionally, propagule members can be produced by the same mechanism as other members are, either by cell division, asexual reproduction, or sexual reproduction, as long as any group member could be a parent.

Phase III: The Individual Phase

Again, there are no discrete boundaries between phases. The advantages of a small propagule are many: It increases the ability to maintain rare beneficial traits by isolating them from deleterious traits and it increases the multiplication component of group-level fitness. As long as the ability of the group to divide is maintained, groups can expand before a further bout of reproduction occurs. If expansion is too slow, further reproduction by the group will produce fewer offspring. So the evolution of a small propagule size necessitates a change in the relative generation times between groups and members, because many generations of parts may be needed to expand the group sufficiently for the group to reproduce without a loss in fitness. Even though the rapid multiplication of parts unfortunately increases the risk of deleterious mutants originating, a small propagule successfully purges those mutants.

The benefits of small propagules are not limitless. A consequence of the evolution of a small propagule size is a uniformity in members. It is commonly expected that a division

of labor among group constituents benefits the group with the increased efficiency that the ability to perform multiple tasks simultaneously allows (Bonner 2001; Harvell 1994). If a division of labor is to arise among constituents, it must contend with the constraints the random reproduction of the group imposes. Any variation within a group after its founding by a small propagule or by differentiation must be easily reversible to a totipotent state. Otherwise not all variants can be inherited. The limit on inheritance of variation and the overall uniformity of groups produced by group-level selection constrains the evolution of division of labor. Only the origin of specified reproductive members will break the constraint imposed by group-level selection, opening the door to extensive division of labor. The deviation from unbiased random sampling distinguishes evolution in the third phase of transitions.

In the hypergeometric model of reproduction outlined in the previous subsection, the expected phenotype of a propagule, $\hat{\phi}'$, is equal to nk/N. Members of type k and other types all have the same probability of being sampled. Fisher's noncentral hypergeometric distribution allows for different sampling probabilities for each member type and would be appropriate to substitute into equation 11 in place of the hypergeometric. However, if propagule size is already small and the group generation times are already long compared to those of the members, nonrandom reproduction can no longer be modeled by a sampling process.

Instead, it is the partitioning of life history into growth and reproduction that is important to the evolution of individuals. An individual controls a finite amount of energy, ψ, in which some fraction is used for maintenance ($\varepsilon(\psi)$).

The rest, equal to its expansive fitness, can be further partitioned into growth ($\pi(\psi)$) or reproduction ($1 - \pi(\psi)$). The efficiency of reproduction is given by, η_r. The reproductive output of an organism with a given growth strategy is equal to (Baudisch 2008)

$$m(\psi) = (1 - \pi(\psi))^{\eta_r}\varepsilon(\psi) \tag{10.13}$$

A wide range of partitioning strategies are possible, from almost constant growth to a phase of rapid growth followed by only reproduction. It may seem strange that such variety in life-history strategy affects reproductive output. Many organisms have unlimited growth, but all real examples, from sponges to coral colonies, reproduce, each spawning millions of gametes into the oceans. Equation 13 describes reproduction as the fraction of energy not used for growth or maintenance. Only if $\pi(\psi)$ and $\varepsilon(\psi)$ are zero could reproductive output be maximized, so in individuals that grow continuously at a high rate, the actual energy expenditure on offspring will have to be low. Even if each offspring is cheap, so that η_r is high, it is important to remember that $m(\psi)$ is only the multiplicative component of fitness. A huge selective filter occurs in the settlement of corals, dramatically increasing the number of juvenile recruits only when the percentage of gravid corals is close to 100 percent (Hughes et al. 2000). Therefore, extremely low values of the persistence component of fitness v could be quite common. Since w is the product of m and v, the demographic

fitness of corals and sponges can be quite low. But since the overall fitness includes expansive and demographic components, those individuals that focus on growth can still have high fitness.

The first covariance term in equation 10.5 finally takes on a value. The phenotype of the individual can directly influence fitness, both by growth and directly, so $\mathrm{cov}(\phi, \hat{\Omega})$ has a nonzero value:

$$\Delta\overline{\phi} = \mathrm{cov}(\phi, \hat{\Omega}) + \mathrm{cov}(\hat{\delta}, \hat{\Omega}) + \overline{\mathrm{cov}_i(\delta, \Omega)} + \mathrm{cov}(\phi_p, \widehat{\Omega_p}) + \overline{\delta_p} \tag{10.14}$$

Organisms and colonies in the individual phase are common. All metazoan organisms are in the individual phase because the organisms themselves are reproductive. However, organisms vary in their degree of individuality. Variation in individuality is correlated with the degree to which reproductive members are determined in the organism (Simpson in review). Three different grades of individuality are well known: the poorly individuated cellular grade (Hyman 1940) with largely somatic embryogenesis (Buss 1983, 1987; Nieuwkoop and Sutasurya 1981), intermediately individuated tissue grade organisms (Hyman 1940) with a mix of epigenetic and preformistic germ speficification (Buss 1983, 1987; Nieuwkoop and Sutasurya 1981), and highly individuated organisms of the organ-system grade (Hyman 1940), also with a mix of epigenetic and preformistic germ speficification (Buss 1983, 1987; Nieuwkoop and Sutasurya 1981).

Evolution and Ecology Within Transitions in Individuality

The expansive and demographic fitness components are clearly associated with different life-history strategies that determine ecological specialization. Organisms that have a significant component of expansive fitness specialize in growth and the occupation and control of space. Corals, bryozoans, sponges, and a number of plant groups have a large expansive component of fitness. Evolution in snails and other motile organisms with approximately determinate growth is dominated by the demographic mode of fitness. It is difficult to describe examples of *organisms* that have only an expansive component of fitness, because there are none, at any level. But recognizing the expansive component of fitness has the benefit of allowing the boundedness of entities be undefined. (The conceptual issues of boundedness in hierarchical levels is well reviewed in section 2.1 of Okasha 2006.) Trait groups, patches, or other aggregations can all change in their spatial extent. A coral colony, a coral reef, as well as a reef community type can all expand in their spatial extent and density of occupation, even if they don't all possess a reproductive capacity.

Because the persistence and multiplication of aggregates is random (following a uniform distribution, for example), the aggregate phase may seem to be rare since many processes that would generate more elaborate underlying stochastic distributions are common. Several physical factors, however, may limit the potential multiplication and differential persistence of aggregates. The most important of these is the relative size of the aggregate compared to

the total habitable area. If an aggregate (think of a highly dispersed trait group) covers nearly the same area as the possible habitat, there can be no net multiplication. No dispersal is possible because there is nowhere new to go. But when aggregates are small relative to the potential habitable area, no purely geometric limits on multiplication exist.

Even with the natural limits, the aggregate phase of transitions in individuality could well be the most diverse phase in terms of the number of distinct aggregates and the number of independent transitions. This is because aggregates require only a context where a set of organisms experience an increase of demographic fitness. It is possible for communities of all scales to satisfy this criterion. Wilson (1980) explored this basic phenomenon in general, but interest has waned, I believe, because the mechanism suffers from an inability to form adaptations of any complexity. Natural selection always leads to adaptation (Van Valen 2009), but natural selection that is primarily expansive has adaptive limits, even within organisms.

The limits to adaptation are imposed mainly by the mechanisms that produce variation between aggregates. All variation within and among aggregates is produced by the constituent organisms, even if the aggregate is at a much higher level. New variants can be expressed within a single aggregate if the potential for aggregate-level multiplication is low, or in a descendent aggregate if multiplication is common. Since the potential for multiplication is largely controlled by the size of the aggregate relative to the habitat area, small aggregates should have a greater potential to differ from each other. Large aggregates will be so few that what variation does occur will generate little selective effect among aggregates.

Of course, in most aggregates, variation is often deleterious, and so the evolutionary pathway tends to pass through the three phases of transitions if the level of adaptations is to increase. But in those aggregates that are composed of multiple species, there is a natural source of preexisting variation, offering an alternative path of a sort to division of labor. Multispecies communities, if they multiply to some degree, have the trouble that members disperse separately, so the heritability of community structure is low.

Coordinated dispersal is fairly common in coral reef communities in the form of multispecies mass spawning (Harrison et al. 1984). It is common for the spawning of one organism to induce spawning in others nearby, resulting in all members of all species spawning simultaneously (Strathmann and Fernald 1987). This means that even if communities are assembled randomly (e.g., Hubbell 2001), any coordinated timing in reproduction will lead to a higher chance of a community reassembling.

The demographic openness of coral reef communities varies according to spatial scale and member dispersal ability (Knowlton and Jackson 2001). At the local scale, reef-dwelling species with larvae that don't disperse well are demographically closed; all members of those types originate locally. Other member species with planktonic larvae can disperse well enough, so that most members are derived from elsewhere and therefore demographically open. At larger scales, the limits of larval dispersal bound communities

(Caley et al. 1996). Historically, reef-building corals, which are mostly colonial, have been observed to have higher extinction rates than the largely solitary reef-dwelling corals (Simpson and Kiessling 2010). As a consequence of these higher extinction rates, large-scale diversity-stability relationships are observed (Simpson and Kiessling 2010).

A single species is a member of aggregates at each of the spatial scales, from locally to larger scales. Each of the scales can be a level of aggregation in its own right. Natural selection is possible at any one of these levels, but I suspect its efficacy is limited by the conflicts among multiple levels. The existence of multiple simultaneous levels of aggregation can undermine patterns so that no consistent patterns are observed as studies cross spatial scales (as described in Jablonski 2008). Any successful coordination among the members of an aggregate will therefore often not translate to other constituent or subsuming levels of aggregation. One way to think about this is in terms of constraints. In multispecies communities, there is no way to channel variation in such a way that optimal solutions to conflicting problems can be found.

Even with all the problems with adaptive community evolution outlined earlier, some multispecies communities have overcome them. The origin of eukaryotes by symbiosis and lichens are the most striking examples, but there are countless other examples of symbioses (Moran 2006) that would qualify as aggregations. In lichens, fragments of the thalus can disperse both the fungal and algal bionts (Budel and Scheidegger 1996; Honegger 1998; Walser 2004).

The origin of eukaryotes is special, partly because these organisms become the building blocks of so much diversity. Mostly, though, they are the clearest example of a multispecies aggregation—that due to the internalized nature of their aggregations—and have solved the problems associated with dispersal and covariation that are inherent in aggregations. Their small size means that they do not face a geometric limit to their reproductive capacity. For example, growth and division of mitochondria can be linked with cell division. Integration is high enough that, in metazoans, strange cross-level effects are common. One interesting example is that mitochondrial genes are integral to the functioning of programmed cell death (Danial and Korsmeyer 2004) in metazoans.

Aggregates and the Number of Hierarchical Levels

The ease with which ecological hierarchical levels fit into the aggregate phase suggests that it may be not be necessary to distinguish ecological and evolutionary hierarchies after all. The hierarchy of ecological interactors—organisms, avatars, local ecosystems, and regional ecosystems (Eldredge and Greege 1992)—can now be seen as a terse list of some common levels of aggregation (figure 10.5). The ecologically organized units are potential, or incipient, higher-level evolutionary individuals. Spatial and temporal scales vary continuously, but a new level does not occur at each scale. Instead, levels correspond to spatial and temporal scales *where dynamics occur*. These are many, but they are not all equal.

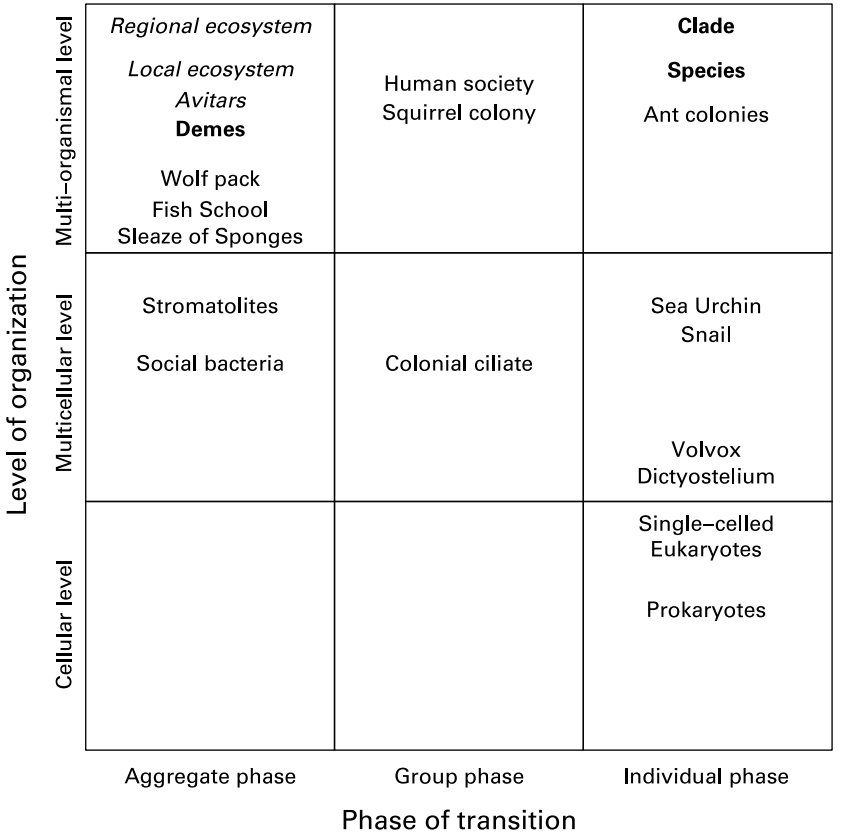


Figure 10.5
The three phases of transitions can be mapped to the hierarchical level of organization. Groups of organisms at various levels of organization can aggregate into new levels. With the right conditions, the aggregations can evolve high degrees of individuality by entering new phases of transitions. Ecological and evolutionary members of the dual hierarchies (e.g., Eldredge and Greege 1992) are mapped equally well into this table. Levels in the ecological hierarchy are shown in *italics*, while levels in the geneological hierarchy are shown in **bold**. Solitary, social, and colonial organisms can occur in any cell, and examples are plotted in normal text. Limits on the adaptive evolution of communities keep them from entering the group and individual phases to a great extent, so they tend to fill up the aggregate phase. The vertical positioning of each is based on the level of organization and further ranks are the qualitative rank order according to spatial and temporal scale. The phase of transitions is based on the dominant component of fitness as estimated by the proportion of the entity that is dedicated to reproduction (Simpson 2009b).

It is possible that we can observe this heterogeneity in meta-analyses of scale in ecological processes. Often, small-scale local patterns do not predict observations made at larger scales (reviewed in Jablonski 2007). From the view I argue for here, each new scale is a potential new level. The level status is attained only when something about the context of the scale leads to an increase in expansive energy of the members. The energetic criteria is important because there is—at least to a first approximation—a limit to the energy available (Van Valen 1976). This limitation in energy is what drives dynamics: Evolution, like economics, is driven by scarcity.

The difficulty in identifying a new level is also due to the lack of clear differences in rank. A coral reef community in the Caribbean may not share precisely the same rank as one in the Red Sea. From a strict constitutive hierarchy view, both are at the same level because they consist of a set of organisms. But because of dispersal and the spatial arrangement of habitable area, the Caribbean reefs are closed demographically, consisting of endemic species, and therefore have a different evolutionary potential from reefs of comparable size in the Red Sea. Those Red Sea reefs are connected by dispersal to the rest of the Indo-Pacific, and so the important scale that determine dynamics is much larger than that of the Caribbean reefs. The rank of the Caribbean level of selection is lower than that for the Red Sea and Indo-Pacific together, because the Caribbean consists of a set of reefs, whereas the Red Sea consists not only of its constituent reefs but also of members from other geographic provinces. This lack of clear boundaries between levels has always been used as an argument against hierarchy. I hope we have learned from Buss (1987), and the three phases of transitions discussed earlier, that when new levels emerge they may not be clearly demarcated; nonetheless, they do occur.

Recognition of the importance of aggregates makes the distinction between scale and hierarchy that Jablonski (2007) and others have advocated even more important. Scale, of course, is just an arbitrary metric. But such hierarchical levels tend not to consist of individuals in the biological sense (where the whole is well bounded, countable, and multiplies). The conceptual difficulty in distinguishing scale and hierarchy is present because they are largely correlated. Jablonski (2007) makes the distinction by recognizing that levels have the characteristics of philosophical individuals, because the constituents of a level are connected in some way. I believe my focus on dynamics and energy control accomplishes the same thing, with a focus on what drives evolution.

Perhaps the complex picture of countless levels, both emergent and individuated, can be best illustrated with the example of humans. One of the key differences between the major transitions in evolution of Maynard Smith and Szathmáry (1995) and the evolutionary transitions in individuality of Michod (1999) is in their treatment of humans, human culture, and language. Maynard Smith and Szathmáry (1995) say that, once language occurs, everything changes and a new level of evolution is attained. Michod (1999) is more specific; the new level must be that of human groups. But these don't have the status of full individuals. One of the only steadfast rules of hierarchy is that rates of change decrease as

you ascend levels in the hierarchy (Salthe 1985, 1993).[1] If the evolution at group level occurs in humans, the rate of change should be slow relative to morphological change. Human culture does just the opposite. The rate of cultural change is much faster than subordinate levels.

If there is significant group-level evolution, large numbers of groups would be required, so how many human groups are there? This question is surprisingly difficult to answer. Humans can be partitioned into groups based on cultural, religious, linguistic, political, geographic, genetic, subcultural, and other possible ways to criteria. But a partition on one criterion will rarely match partitions on others because humans naturally belong to several groups. Instead of giving up and saying there are no groups, we can recognize that a multitude of groups coexist, with each at a potentially different level because of differences in the scale of inclusiveness. Each group will vary in its components of fitness, as will other subordinate and subsuming groups. And the directions of selection may not coincide. This is the same situation as we find in ecological hierarchies. Effectively, an infinite number of levels exists in the interstices between obvious organizational levels. The obvious levels, like integer numbers, are those that have proceeded through to the individual phase of transitions—cellular, multicellular, and multiorganismal (colonial or social).

The path a lineage takes through the phases of transitions is not fixed, but determined by ecology. Expansive fitness, though it straddles levels, can be a potent component of high-level fitness because it directly contributes to the ability to control and occupy space. Whole animal phyla, like the bryozoans, are dedicated to this mode of life, and so are those only partially individuated at both the organismal and colonial levels (Simpson in review). The existence of multiple adaptive peaks that represent particular ecological life-history strategies also means that there is no way to be stalled midtransition (Stearns 2007), even if transitions are not instantly complete.

New aggregates constantly form and most probably dissipate before they attain a significant amount of expansive fitness. Even in *Volvox*, incipient levels can be observed, when a colony contains within itself three generations of offspring. Deciding whether *Volvox* is an alga with parental care or a colony of colonies is not as important as recognizing that even here we can see how levels of aggregations spontaneously arise. The raw material for entering into another aggregate phase of transitions is already present.

Even in the early fossil record, between 2.6 to 2.3 billion years ago, we can see complex spatial structuring in stromatolitic reef complexes (Grotzinger 1989), and even if the stromatolites are not biogenic themselves (Grotzinger and Knoll 1999), the spatial structure would still influence the life in these reef complexes.

Once ecology occurs, there is no upper limit on the potential hierarchical complexity of life. Of course, the first life forms had ecology. The only limits are on the potential for those levels and degrees of aggregations to evolve. Since the origin of life, it seems that the hierarchical complexity of life has always been infinite. What we think of as the transitions in

individuality, and an increase in hierarchical complexity, are only instances where the individual phase has been reached.

Acknowledgments

Many thanks to Brett Calcott and Kim Sterelny for organizing an exciting and successful meeting. I am grateful to Eva Karner and Werner Callebaut at KLI for the wonderful experience at Altenberg. Thanks to Dan McShea and Wolfgang Kiessling for commenting on this paper and to Dave McCandlish and Lauren McCall for discussing many of the ideas in this paper with me. This work was funded by the Cambridge-Templeton Foundation, grant # 1038 to D. McShea.

Note

1. This rule seems to work in both physical and evolutionary hierarchical systems. Geological processes continuously change the surfaces and possibly the internal structure of planets while the basic organization of the solar system changes considerably more slowly. The number of types of ant colonies, if measured by patterns of social organization is dwarfed by the number of ant species. Although not conclusive, this pattern suggests that the evolution of colony types is much slower than the evolution of organismal traits. Variation in rate of change across levels is largely a consequence of a many-to-one relationship between lower-level parts and the whole.

References

Arnold, A. J., and K. Fristrup. (1982). The theory of evolution by natural selection: A hierarchical expansion. *Paleobiology* 8(2): 113–129.

Baudisch, A. (2008). *Inevitable Aging?: Contributions to Evolutionary-Demographic Theory*. Demographic Research Monographs 4. Berlin: Springer.

Bell, G., and A. O. Mooers. (1997). Size and complexity among multicellular organisms. *Biological Journal of the Linnaean Society* 60(3): 345–363.

Bonner, J. T. (2001). *First Signals: The Evolution of Multicellular Development*. Princeton, NJ: Princeton University Press.

Budel, B., and C. Scheidegger. (1996). Thallus morphology and anatomy. *Lichen Biology* 37–64.

Buss, L. W. (1983). Evolution, Development, and the Units of Selection. *Proceedings of the National Academy of Sciences* 80(5): 1387–1391.

Buss, L. W. (1987). *The Evolution of Individuality*. Princeton, NJ: Princeton University Press.

Caley, M. J., M. H. Carr, M. A. Hixon, T. P. Hughes, G. Jones, and B. A. Menge. (1996). Recruitment and the local dynamics of open marine populations. *Annual Review of Ecology and Systematics* 27: 477–500.

Charnov, E. L., and S. K. M. Ernest. (2006). The offspring-size/clutch-size trade-off in mammals. *The American Naturalist* 167(4): 578–582.

Damuth, J., and I. L. Heisler. (1988). Alternative formulations of multilevel selection. *Biology and Philosophy* 3(4): 407–430.

Danial, N., and S. Korsmeyer. (2004). Cell death critical control points. *Cell* 116(2): 205–219.

Dawkins, R. (1982). *The Extended Phenotype: The Gene as the Unit of Selection*. San Francisco: Freeman.

Eldredge, N. (1985). *Unfinished Synthesis: Biological Hierarchies and Modern Evolutionary Thought*. New York: Columbia University Press.

Eldredge, N., and M. Greege. (1992). *Interactions: The Biological Context of Social Systems*. New York: Columbia University Press.

Frank, Steven A. (1998). *Foundations of Social Evolution*. Princeton, NJ: Princeton University Press.

Grotzinger, J. P. (1989). Facies and evolution of Precambrian carbonate depositional systems: Emergence of the modern platform archetype. In P. Crevello, J. Wilson, J. Sarg, and F. Read (eds.), *Controls on Carbonate Platform and Basin Development*. Society of Economic Paleontologists and Minerologists Special Publication 44: 79–106.

Grotzinger, J. P., and A. H. Knoll. (1999). Stromatolites in Precambrian carbonates: Evolutionary mileposts or environmental dipsticks? *Annual Review of Earth and Planetary Sciences* 27(1): 313–358.

Hamilton, W. D. (1975). Innate social aptitudes of man: An approach from evolutionary genetics. In R. Fox (ed.), *Biosocial Anthropology* (pp. 133–155). New York: Wiley.

Harper, J. L. (1977). *Population Biology of Plants*. New York: Academic Press.

Harrison, P., R. Babcock, G. Bull, J. Oliver, C. Wallace, and B. Willis. (1984). Mass spawning in tropical reef corals. *Science* 223(4641): 1186–1189.

Harvell, C. D. (1994). The evolution of polymorphism in colonial invertebrates and social insects. *Quarterly Review of Biology* 69(2): 155–185.

Herron, M., and R. Michod. (2008). Evolution of complexity in the volvocene algae: Transitions in individuality through Darwin's eye. *Evolution* 62(2): 436–451.

Honegger, R. (1998). The Lichen symbiosis—What is so spectacular about it? *Lichenologist* 30(3): 193–212.

Hubbell, S. (2001). *The Unified Neutral Theory of Biodiversity and Biogeography*. Princeton, NJ: Princeton University Press.

Hughes, T. P., A. Baird, E. Dinsdale, N. Motschaniwskyj, M. Pratchett, J. Tanner, and B. Willis. (2000). Supply-side ecology works both ways: The link between benthic adults, fecundity, and larval recruits. *Ecology* 81(8): 2241–2249.

Hull, D. L. (1980). Individuality and selection. *Annual Review of Ecology and Systematics* 11: 311–332.

Hyman, L. H. (1940). *The Invertebrates: Protozoa through Ctenophora*. New York: McGraw-Hill.

Jablonski, D. (2007). Scale and hierarchy in macroevolution. *Palaeontology* 50(1): 87–109.

Jablonski, D. (2008). Biotic interactions and macroevolution: extensions and mismatches across scales and levels. *Evolution* 62(4): 715–739.

Knowlton, N., and J. B. C. Jackson. (2001). The ecology of coral reefs. In J. F. Bruno, M. D. Bertness, S. D. Gaines, and M. E. Hay (eds.), *Marine Community Ecology* (pp. 395–422). Sunderland, MA: Sinauer.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution*. San Francisco: W.H. Freeman.

Michod, R. (2006). The group covariance effect and fitness trade-offs during evolutionary transitions in individuality. *Proceedings of the National Academy of Sciences* 103(24): 9113–9117.

Michod, R. (2007). Evolution of individuality during the transition from unicellular to multicellular life. *Proceedings of the National Academy of Sciences* 104(suppl 1): 8613.

Michod, R. E. (1999). *Darwinian Dynamics: Evolutionary Transitions in Fitness and Individuality*. Princeton, NJ: Princeton University Press.

Michod, R. E. (2001). Cooperation and conflict in the evolution of multicellularity. *American Zoologist* 41(6): 1526–1526.

Michod, R. E., Y. Viossat, C. A. Solari, M. Hurand, and A. M. Nedelcu. (2006). Life-history evolution and the origin of multicellularity. *Journal of Theoretical Biology* 239(2): 257–272.

Moran, N. (2006). Symbiosis. *Current Biology* 16(20): 866–871.

Nieuwkoop, P. D., and L. A. Sutasurya. (1981). *Primordial Germ Cells in the Invertebrates: From Epigenesis to Preformation*. New York: Cambridge University Press.

Okasha, S. (2006). Evolution and the Levels of Selection. Oxford: Oxford University Press.

Price, G. R. (1972). Extension of covariance selection mathematics. *Annals of Human Genetics* 35(4): 485–490.

Queller, D. C. (2000). Relatedness and the fraternal major transitions. *Philosophical Transactions: Biological Sciences* 355(1403): 1647–1655.

Rice, S. H. (1995). A Genetical Theory of Species Selection. *Journal of Theoretical Biology* 177(3): 237–245.

Rice, S. H. (2004). *Evolutionary Theory: Mathematical and Conceptual Foundations*. Sunderland, MA: Sinauer.

Rice, S. H. (2008). A stochastic version of the Price equation reveals the interplay of deterministic and stochastic processes in evolution. *BMC Evolutionary Biology* 8: 262.

Roze, D., and R. E. Michod. (2001). Mutation, multilevel selection, and the evolution of propagule size during the origin of multicellularity. *American Naturalist* 158(6): 638–654.

Salthe, S. N. (1985). *Evolving Hierarchical Systems: Their Structure and Representation*. New York: Columbia University Press.

Salthe, S. N. (1993). *Development and Evolution*. Cambridge, MA: MIT Press.

Sherman, P. W. (1985). Alarm calls of Belding's ground squirrels to aerial predators: nepotism or self-preservation? *Behavioral Ecology and Sociobiology* 17(4): 313–323.

Simpson, C. (2010). Species selection and driven mechanisms jointly generate a large-scale morphological trend in monobathrid crinoids. *Paleobiology* 36(3): 481–496.

Simpson, C., and W. Kiessling. (2010). The role of extinction in large-scale diversity-stability relationships. *Proceedings of the Royal Society B: Biological Sciences* 277: 1451–1456.

Simpson, C. (in review). The evolution of division of labour.

Stearns, S. (2007). Are we stalled part way through a major evolutionary transition from individual to group? *Evolution* 61(10): 2275–2280.

Strathmann, M., and R. Fernald. (1987). *Reproduction and Development of Marine Invertebrates of the Northern Pacific Coast: Data and Methods for the Study of Eggs, Embryos, and Larvae*. Seattle: University of Washington Press.

Van Valen, L. M. (1976). Energy and evolution. *Evolutionary Theory* 1(7): 179–229.

Van Valen, L. M. (2009). How ubiquitous is adaptation? A critique of the epiphenomenist program. *Biology & Philosophy* 24: 267–280.

Vrba, E. S., and S. J. Gould. (1986). The hierarchical expansion of sorting and selection—sorting and selection cannot be equated. *Paleobiology* 12(2): 217–228.

Wade, M. J. (1978). Critical review of models of group selection. *Quarterly Review of Biology* 53(2): 101–114.

Walser, J. C. (2004). Molecular evidence for limited dispersal of vegetative propagules in the epiphytic lichen *Lobaria pulmonaria*. *American Journal of Botany* 91(8): 1273–1276.

Williams, G. C. (1992). *Natural Selection: Domains, Levels, and Challenges*. New York: Oxford University Press.

Wilson, D. S. (1980). *The Natural Selection of Populations and Communities*. Menlo Park, CA: The Benjamin/Cummings Publishing Company.

Wilson, E. O. (1975). *Sociobiology: The New Synthesis*. Cambridge, MA: Belknap Press of Harvard University Press.

11 Plant Individuality and Multilevel Selection Theory

Ellen Clarke

Individuality in plants seems as obscure and ambiguous as in animals it appears clear and simple.
—Gray (1849), in White (1979, 113)

Gray's statement may seem an exaggeration to the modern reader. Although philosophers of biology have become accustomed to worrying over whether genes or species are real units of selection, it is generally taken as uncontroversial that organisms, at least, are individuals. Even multilevel selection theorists, who may acknowledge the challenges presented by things such as outlaw genes or eusocial insect colonies, don't tend to include plants among their list of entities that warrant serious philosophical concern. Yet in the nineteenth century such fears were commonplace among biological thinkers. Even before Charles Darwin was discussing the possibility of group selection, his grandfather Erasmus was discussing some of the peculiarities that can prompt confusion over the status of plants. He considered plant buds to be like babies growing on their parent stem. Many writers then subscribed to the view that plants and trees are not individuals at all, but rather metapopulations, or collections of unit parts. The eruption of green shoots and leaves each spring is not mere growth after a dormant period, but the birth of a new generation.

A tree is therefore a family or swarm of individual plants. (Erasmus Darwin 1800, quoted in White 1979, 109)

This chapter explores the motivation behind such views and derives some consequences for multilevel selection theory. The first section explores the problem of individuating plants and the suggestion that individuality should be settled using genetic homogeneity. The second section argues that, in some lineages, high somatic mutation rates might actually be favored, and selection processes acting on these mutations could actually be adaptive for the higher-level individual. The third section concludes that genetic heterogeneity, and the intraorganismal selection it can give rise to, does not always undermine a higher level of selection. Individuals can, given certain conditions, have competition among their parts.

The lesson I draw is that multilevel selection theorists are wrong to assume genetic heterogeneity necessarily results in evolutionary conflict that must be suppressed in order for

higher-level individuals to persist as units of selection. Under particular circumstances (circumstances satisfied by many plants) competition at a lower level can be beneficial for a higher-level individual.

Plants and Individuality

Modularity—The Plant as a Metapopulation

All vascular plants, including ferns, conifers, and flowering plants, grow by the accumulation or iteration of smaller constructional units. When a coconut palm grows, it does so by producing a new leaf at its crown. As every new leaf appears, an older leaf below will die and fall away, leaving its stem to contribute to the trunk. You can clearly see cross-sectional marks all along its trunk where these units have been repeated. Other plants iterate more than one unit at once. An oak tree develops by growing new shoot units in a forked or branching pattern. Clonal plants such as bracken or aspen iterate whole plants, by growing them from the ends of underground runners. All such growth patterns can be called modular. Modular growth is open ended and does not progress toward any fixed adult form, in contrast to development in so-called unitary organisms, which is determinate.

A modular organism grows by the repetition of some unit or module. These modules are self-reproducing, which is part of what prompted Erasmus Darwin and others to say that a modular organism should be viewed as a collection of individuals—reproductive ability is often thought to be the kind of property that only individuals possess. Yet there has to be more to it than that, because even humans are composed of smaller parts that are capable of reproducing themselves—cells. We will see later that it is very important to be clear on what *kind* of reproduction an entity must be capable of, if we are to use that as an indicator of individuality.

A couple of distinctions need to be made. First, clonal growth of the sort described is vegetative. Another type of cloning is parthenogenesis, which occurs even in vertebrate lineages and in which an organism self-fertilizes one of its gametes. For the purposes of this chapter, cloning by parthenogenesis or selfing is less interesting because it involves a single-celled stage, precluding many of the interesting consequences of multicelled propagation that I will be looking at later. Specifically, reproduction by means of a multicellular propagule has the potential to transmit multiple genotypes to the offspring, whereas unicellular propagules will always sample only one genotype from any variance in the parent. All references to "clones" in this chapter should therefore be understood as referring to an organism that has been produced vegetatively rather than parthenogenetically or by selfing.

Second, structural modularity, where an organism is built up out of the repetition of semiautonomous subparts, should be distinguished from developmental modularity. All complex life forms are probably, in part, developmentally modular. Developmental modu-

larity describes the partitioning of ontogenetic or embryonic processes into separate subprocesses, which develop to some degree autonomously of one another. Evolutionary modularity describes yet another separate but related phenomenon, in which parts or subprocesses within a lineage of organisms vary at different rates over evolutionary time (Schlosser and Wagner 2004). For the purposes of this work, whenever I refer to modularity, I am picking out the structural sense of the term, in which parts of the mature organism are iterated vegetatively and operate autonomously (in some respect and to some degree) over the course of the life cycle. Arthropods, for example, with their repeating body segments, are often described as developmentally modular, but I do not include them as being structurally modular.

It is important to define the meanings of some words I will be using:

A *module* is a self-reproducing and semiautonomous unit. In plants, it usually contains one or many meristems in a bud, shoot, or root.

A *meristem* is a special kind of plant cell that can differentiate into both germ and soma.

A *ramet* is a mitotically produced collection of modules that forms a physically coherent structural entity (a tree, or bush, for example.)

A *genet* is the collection of all those modules or ramets that have developed from a single zygote, that is, all the products of a single sexual reproductive event.[1]

When ramets iterate themselves we say the organism is *clonal*.

Clonality and modularity occupy a single spectrum, differentiated primarily by the degree of physical separation between modules. Some organisms, such as grasses, switch between modular and clonal modes according to environmental conditions. (The different modes are described as phalanx and guerrilla strategies, respectively.[2]) Many organisms are clonal as well as modular. For example, bracken ferns, strawberries, and aspen are modular (in fact, all higher plants are) in that their bodies are built by the iteration of root and shoot units. They are *also* clonal because they send out runners that grow into whole new genetic copies of the plant.

Some organisms are more modular than others. The English oak, or *Quercus robur,* has a high degree of differentiation of its parts. The root system differs from the shoot system. The uppermost leaves are dependent on the rest of the tree for water and nutrients. The roots are dependent on the rest of the tree for energy from sunlight. An oak tree is a unitary organism to a large extent. However, it retains a much greater degree of modularity than any metazoan. This is because all plants are developmentally plastic. Cells taken from just about any part of the tree can, given the right conditions, be grown into a whole new and sexually fertile tree. You cannot do this with most metazoans. In metazoans, the cells capable of growing a new organism are usually carefully hidden in the ovaries or testes (this is known as germ-soma separation). In the majority of cases, only sexual fertilization can

start a new life cycle. But plants grow new parts using meristem cells, and these are not sequestered (isolated) as they are in metazoans, but are scattered around the plant body, often remaining dormant. Thus, although the mature oak is differentiated, that specialization can be reversed, so that nearly all parts of the tree retain autonomy and independence, at least potentially, throughout the lifetime of the tree.

One organism we're going to keep coming back to is Quaking aspen, *Populus tremuloides*. Aspen are more modular than oak. Aspen trees look similar to oak trees in that there is a trunk, a root system, and a shoot system. But aspen also send runners underground, and from the ends of these runners grow new trees. There are known aspen genets that are 50,000 ramets strong and estimated to be over 10,000 years old. In fact, all clonal genets are potentially immortal. A forest of aspen trees has the same branching structure as a single aspen tree, except that while the trunk and roots of a tree are visible above ground, those of the forest are hidden under the soil. Of course, an aspen forest isn't perfectly analogous to an enormous and partially buried oak. First, the parts of the forest are less differentiated and more independent of each other than are the parts of an oak. Second, whether or not the aspen trees remain connected to each other is largely a matter of chance, because parts of the connective root structure are known to rot and decay, leaving parts of the network isolated. Land subsidence and burrowing animals likewise threaten the coherence of the structure. This usually fails to impact the health of the isolated trees, whereas severing the branches of an oak from their stem would cause certain death. This suggests that the trees are more autonomous than the shared root connections may suggest, although it may be that younger trees depend more on the network, and its usefulness as a resource declines with age. Aspen ramets certainly exchange water and other nutrients via their shared root system and will frequently graft with roots from other genets (see, e.g., Jelinkova, Tremblay, and De Rochers 2009).

. . . And Other Modular Animals

Higher plants are all modular, but they are not the only modular organisms. Fungi are modular and so are many animals, especially marine invertebrates. Well-known examples include corals—reef-building colonies of tiny coral polyps—and hydroids, such as the Portuguese man-o-war jellyfish, which is really a floating colony of thousands of individual zooids. Therefore, although I will mostly be limiting the discussion here to plants, it is important to realize that the implications reach far beyond that kingdom. All in all, modular organisms are not an insignificant subsection of the living world—in fact they make up well over fifty percent of the earth's biomass. And yet, as we will see, no one can quite agree what exactly these organisms are, or, more pragmatically, how exactly to incorporate them into our current evolutionary theory. An evolutionary theory that covers only a part of the biological diversity that has been generated since life began is not the kind of evolutionary theory we are looking for.

The Units Debate

How can one steer a middle course between indefinite subdivision and indefinite expansion. . . . Which member of the series deserves pre-eminently the title of individual? (Braun, 1853; quoted in White 1979, 134)

In order to test predictions generated using modern evolutionary theory, biologists need to measure fitness. There is a lot of controversy regarding the correct understanding of the fitness concept, but no matter what interpretation of fitness you favor, measuring fitness requires being able to count individuals. Being fit is about contributing to later generations, so measuring fitness requires the ability to differentiate between generations. We need to know what it means for an individual to count as being of a new generation rather than a mere part of its parent, and we need to be able to tell the difference between having a single offspring and having many.

There are three competing characterizations of the individual with respect to aspen (and other modular and clonal organisms.) As we will see, a biologist's preference for one view over the others is affected by their preference for one or another foundational criterion of individuality—sex, heritability, or object-hood. The genet view (Janzen 1977) says that the whole clone or forest is the individual, because an individual is just the developmental product of a zygote. The ramet view (Fagerström 1992; Harper 1977; Pan and Price 2002) holds that the aspen *trees* are individuals, and the module view (Pedersen and Tuomi 1995; Tuomi and Vuorisalo 1989a; Wikberg 1995; Winkler and Fischer 1999) holds that the true individuals are to be found at an even smaller scale—the root and shoot modules or the meristems.

Each of these views has merits as well as serious problems. The genet view allows us to talk about the capacity for clonal (vegetative) growth being an adaptation at the level of the genet. The idea is that the aspen spreads itself out in order to exploit a wider range of environmental resources and distribute its risk of mortality. On the other hand, individuals are commonly presumed to have a life cycle, but genets seem to lack these; it is ramets that reach maturity and senesce (Watkinson and White 1986). Only ramets show specialization of parts. The ramet view is also a fairly intuitive response to the problem, because trees *look like* organisms. Familiar organisms are proper objects or particulars; their parts are physically connected to each other and separated from everything else.[3] While the spatial contiguity of the genet is vague and arbitrary, according to what has or has not decayed in the root structure, ramets have nice clear edges wrapped in bark, ending in leaves at one end and roots at the other. However, although aspen ramets are relatively easy to delineate, because their propagating runners are underground, this apparent obviousness may be just an illusion of scale. If we were able to see underground, the aspen forest would appear not as a collection of discrete trees at all, but as a single stalked mass, topped with branches, something like a head of broccoli (Bouchard 2008). Another attribute thought to belong to

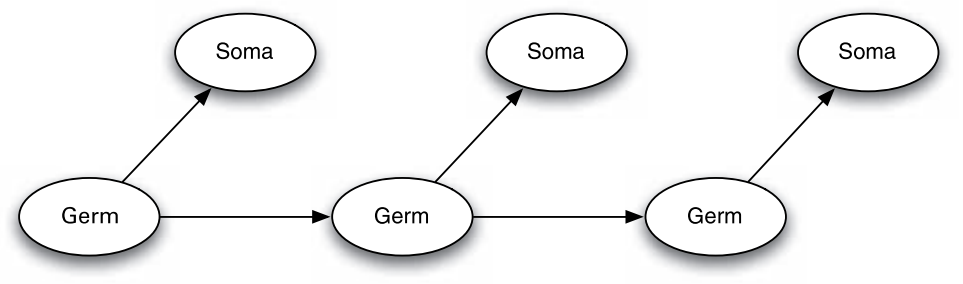


Figure 11.1
Germ-soma separation (reproduced from Buss 1983).

individuals is reproductive autonomy (Santelices 1999). Based on this criterion, modules are the real unit of selection.

With this overview of the different conceptual routes that are available for individuating plants, we are now in a position to summarize the theoretical commitments that determine our path:

1. *Continuity of the germ (heritability)*. Weismann's doctrine of the continuity of the germ plasm (Weismann 1893) states that although the germline is continuous, so that traits are inherited from ancestor germlines, and these are passed on to the soma, there is no transmission of traits from soma back to germ. Germ-soma separation is commonly schematized as in figure 11.1.

The arrows in the figure show the direction of heritability, or the transmission of traits. So while both somatic cells and germ cells can be said to reproduce, in that they mitotically divide, Weismann's barrier ensures that only germ, not soma, has the potential for long-term evolution. To count a unitary organism, such as a pig, we use Weismann's barrier to help us decide which pig parts to count. Only the germ pig cells, hidden away in the ovaries or testes, can influence the traits of descendent pigs. Somatic pig cells are evolutionary dead ends, because their traits are not heritable. So when we are thinking about evolution, the genes in the somatic cells can be ignored. Even pig germ cells don't need to be counted separately, because they can only influence the traits of descendents when paired in the right way with complementary germ cells, and as soon as this happens they form part of a new individual. Furthermore, pig germ cells are entirely helpless without the soma. All the different cells and organs and other parts are cooperating in the production of a new piglet, so we don't need to count them all separately. We just count the pig once.

Plants, on the other hand, show somatic embryogenesis. Plant germ cells, meristems, are not sequestered at all, but are distributed around the body of a plant throughout its lifetime. They are usually concentrated at the apices of root and shoot tips, but dormant meristem cells can be found all over the stem, and anyway plant cells are so developmentally plastic that almost all of them can, given the right treatment, be persuaded to be-

come totipotent, start a new developmental cycle, and propagate genetic units to a new generation. Weismann's barrier does not give us reason against counting plant parts separately.

2. *Sexual reproduction*. We can tell the difference between producing piglets and merely producing new pig cells because piglets are always produced sexually. Piglets always start out as single-celled zygotes, so we can say that everything after this single-celled stage counts as a new individual. Sexual recombination of the genome also guarantees that the new individual will be genetically unique, allowing us recourse to genotypes as a method of distinguishing individuals. But modular organisms like aspen reproduce asexually via runners, as well as sexually. Plant cells can pass on their traits to new plants even in the absence of sex.[4]

3. *Boundaries*. Last, pigs have edges. We do not need to worry about where one pig ends and another begins because they have reasonably clear boundaries. Aspen lack clear spatial boundaries because whether or not a tree remains physically connected to the rest of the genet is vague and arbitrary.

In pigs, these three sets of criteria converge to provide a single verdict on pig individuality, but for modular organisms they support separate views. Which view you choose regarding individuality therefore depends largely on which of these three features you see as most important. If you think that sexual reproduction is the most important thing about an individual, then you are likely to opt for the genet view, in which an individual is just the whole product of a sexual reproductive event. If you think that continuity of the germ (heritability) is key, then you will probably be moved to choose a module or meristem view. Finally, if you think that individuality is usually something that can be settled in a straightforward, less theoretically laden way on the basis of boundaries, then some kind of ramet view will most appeal to you.

The Gene's-Eye View

It should be noted that moving from fitness of individuals to fitness of alleles won't help, for how do we know which genes to count? And how often should we count them? The gene's-eye view collapses onto the organism view when you realize that geneticists use generations to define their time intervals. What is more, even with continuous time models, they use Weismann's doctrine of germ-soma separation to decide which genes to count. If a cow eats a lot of grass, it gets bigger because the number of cells in its body increases. Each cell contains a set of genes, so whenever a cow gets fatter, there are more copies of its alleles. Yet we wouldn't say that any of those alleles have raised their fitness. We don't count every single copy of an allele that exists when we count gene frequencies; we only count the ones that matter—that is, those that make it into zygotes. So we count organisms. What about organisms that don't make zygotes? Is the aspen that founds a new aspen grove, stretching to thousands of trees, just like a cow getting fatter? Or should we count

those trees separately because they are new life forms, capable of independent life and reproduction? The gene's-eye view won't tell us.

Clinging to the Genetically Homogenous Individual

> The existence of multiple genetic lineages within an organism creates a breeding ground for conflicts and cheating where lineages pursuing their own interests increase their returns relative to other lineages while decreasing the fitness of the organism. (Pineda-Krch and Lehtilä 2004, 1171)

Many authors prefer the view that a true individual is going to be genetically homogeneous—all of its parts will possess one common genotype. The thought is usually that homogeneity is required in order to prevent outbreaks of conflict among an individual's parts. The necessity of homogeneity to individuality is frequently taken as a fundamental underlying assumption in the major transitions literature.

The claim is that cooperation can be achieved only at higher levels of organization (such as the level of the multicellular organism) if there is some mechanism or condition that prevents the lower-level individuals from doing better by cheating (Buss 1983; Frank 1995; Maynard Smith and Szathmáry 1997). Specific policing mechanisms will have to evolve to prevent those individuals from competing at all (Frank 2003). Alternatively, the free rider problem is solved automatically whenever the lower-level individuals are genetically identical to one another. Hamilton's explanation of this is in terms of kin selection: If your genes are identical to mine, then my doing well at your expense is identical, in terms of allele propagation, to your doing well at my expense. Self-interest and altruism fail to be separate alternatives.

Dawkins (1982) defends homogeneity in these terms in his *Extended Phenotype*. He says that, for something to function as a unit of selection, it must not contain too much genetic variation. He argues that true individuals must be produced by means of single-celled bottleneck life stages, partly because single-cell stages ensure that only a single genome is inherited. Anything that is produced by a multicellular runner, such as a strawberry plant or an aspen ramet, is not a unit of selection because it will contain too much variation. He says "a geneticist will not discern a population of plants at all. The whole mass of straggling vegetation will have to be regarded as a population of cells, with cells of any one genotype being untidily peppered across the different plant." (Dawkins 1982, 260). The key point is that, in order for individuals to function as units of selection, there must be no lower-level selection. Genetic homogeneity ensures that there will be no competition among an individual's parts.

There is a serious limitation to the argument for using genetic homogeneity as the key criterion on which to delimit individuals. Homogeneity fails to obtain. All organisms contain genetic variation, because it is produced at a fairly steady rate by mutation during mitotic divisions. Replication is not perfect (if it were, of course, we would not be here), and despite there being lots of evolved mechanisms for minimizing heterogeneity (genetic variance) within humans (e.g., developmental bottlenecks, apoptosis, and policing systems

such as the immune system and the histocompatibility complex), it is well known that we tend to show a high degree of somatic mutation.[5] Yet we also know that somatic mutation causes a large number of problems, cancer being the most obvious. Cancer can be viewed as a free-rider problem—the lower-level individual (cell) is behaving selfishly and replicating itself at the expense of the higher-level individual. Plants and other modular organisms are known to show much, much higher genetic heterogeneity, because they lack mechanisms for minimizing it. The presumption that seems to follow, in the minds of Dawkins and others, is that plants and other modular organisms must face proportionally more serious free-rider problems. In fact, the problems must be so bad that the higher-level entity gets completely undermined and does not function as an individual at all.

In the next section, I present a challenge to this assumption, and suggest evidence that genetic heterogeneity may not always be a barrier to successful functioning as a higher-level individual.

Intraorganismal Selection

Evolution has classically been viewed as acting on variation among individuals within a population. Variation within individuals tends to be ignored. In the previous section, we began to see how Weismann's doctrine can explain why somatic mutation has not been treated as significant. Here, I introduce the theoretical idea of intraorganismal selection (IOS) and show how mechanisms that support it might be adaptive. First of all, we need to learn a little about the phenomenon of genetic heterogeneity.

Mutations and Mosaics

> The sheer number of cells that must be produced during the development of all but the smallest of organisms ensures that almost every individual is a genetic mosaic. (Otto and Hastings 1998, 509)

Previously we saw how intraorganismal genetic heterogeneity is seen by many to be an aberration, bringing deleterious effects. Maynard Smith and Szathmáry (1997, 244) argue that selection within an individual can be ignored because all the cells within a multicellular organism share a recent common ancestor (the zygote), so there is little or no genetic variation between cells. In fact, genetic heterogeneity is common because of the mutations, crossing over, and gene conversions that can happen during cell mitosis. Organisms can fail to be genetically homogeneous in two main ways: They can be chimeric or they can be mosaic. These states are differentiated in terms of the functional origin of the variation (although some authors use the terms as if they are equivalent). Chimerism is the term for an individual composed by two or more fused genotypes that came from different zygotes. A mosaic individual is composed of two or more genotypes that originated from a single zygote but diverged during mitotic (somatic) growth. In both cases, there is a single structural or functional individual in which different areas carry a different genotype.

Plants can become chimeric when they are grafted together. Seaweeds are known to coalesce (Monro and Poore 2004). Chimerism also commonly occurs in cellular slime molds (Dictyostelium), sponges, corals, and tunicates (and aspen root networks). In other organisms, it is prevented by histocompatibility mechanisms. More frequently, plants are mosaic. Here, we focus exclusively on mosaic heterogeneity because chimerism is not generally heritable.[6] In mosaics, "the branches of an individual tree or parts of a clone represent an archipelago of similar but distinct genetic islands" (Whitham and Slobodchikoff 1981, 287). Many of the plants in cultivation came originally from mosaics; mutations gave us pink grapefruits, seedless grapes, and navel oranges to name just a few (Otto and Hastings 1998). In fact, all the pink grapefruit in existence originate from a single branch of a single tree that was discovered in 1906 (Whitham and Slobodchikoff, 1981).

The level of mutation in mosaics depends on several things; the rate of mutation, the number of cell divisions (dependent on the lifetime of the organism), and the rate of purging. Modular organisms tend to have higher degrees of mosaicism than unitary organisms because of their long lifespan, unsequestered germline, and multicellular propagation. Plants, especially clonal plants, have very long lives; in fact, they do not senesce at all. Genets are, at least potentially, immortal, because they only die if every single one of their ramets dies. They do not always limit heterogeneity during ramet iterations by forcing development through a single-celled bottleneck; in aspen ramet propagation occurs by means of a multicellular runner, and this runner will pass on any heterogeneity that it happens to contain. Modular and clonal organisms are, again, special cases because iteration occurs at several levels and is often multicellular at every level; multicellular modules are iterated *and* ramets are iterated via multicellular runners, too. Furthermore, runners do not spring from a specially sequestered site, protected from mutation. They can arise from all over the root system. Any mutation occurring in any of the dormant meristems found throughout the root system can potentially be passed on to a new ramet, either vegetatively or sexually.

So clonal plants can be expected to show a huge degree of heterogeneity. Many authors have argued that this should be bad for them. Muller's ratchet[7] style explanations for the maintenance of sexual reproduction in an overwhelming majority of organisms center on the idea that sexual recombination purges mutations and thus protects sexual species from "mutational meltdown" (Klekowski 2003). This is the supposed advantage that makes the "twofold cost of sex" worth paying. Without recombination, asexual clonal plants accrue a lethal level of mutations.

> In the short term vegetative reproduction can clone genotypes that may be adaptively superior. In the long term, prolonged vegetative reproduction can lead to slow genotypic degradation through the stochastic fixation of deleterious somatic mutations. (Klekowski 2003, 61)

Or so say the theorists. The problem is that botanists and ecologists actually looking at clonal and asexual plants will not agree. Clonal and asexual plants thrive. Dorken and Eckert (2001) have done extensive fertility analyses and found that there is no association

between sexual fertility and plant vigor. Indeed, complex traits like sex are often degraded by mutation when they no longer increase fitness.

> Where field biologists associate vegetative vigour with clonality, theoretical biologists view clonality in a different way. (Klekowski 2003, 65)

IOS or Somatic Selection

> If somatic mutation of the genome is a frequent feature in modular organisms, natural selection is to be expected within genets and between modules. (Watkinson and White 1986, 47)

I follow convention in calling this "intraorganismal selection" (IOS), although the foregoing discussion should have made it clear that relativizing selection to the organism is not an unproblematic way of singling out the focal level in question. When the organism is clonal and forms physically connected genets, then IOS can be seen as selection between ramets. For nonclonal organisms, IOS will describe selection at a level lower than the ramet, that is, between modules or meristem cells. The basic point of IOS is that evolutionary change can take place within an individual, as well as between successive generations of individuals. The focal level of selection is shifted down so that the individual acts in some ways as a population, and evolutionary change occurs between successive generations of that individual's subparts. Of course, this could be said to be commonplace. After all, an individual wracked with cancer will also show a shift in frequencies of a particular allele over time. The difference has to do with long-term evolutionary consequences. In plants, the intraorganismal evolution is *heritable* because the victors of selective battles can be transmitted via both sexual and asexual routes. A better human analogy may be the immune system; this involves evolution during the lifetime of an individual, but it is also partly heritable—through breast milk, for example—between generations of individuals.

IOS (also called somatic, diplontic, or cell-lineage selection; see Buss 1983, Hughes 1989, Otto and Hastings 1998) occurs when genetic differences between cells or other subunits cause their differential survival or proliferation during an individual's development, and it has been discussed as a theoretical possibility since at least 1965.[8]

> Once genetic differences exist between parts of the same plant there is the opportunity for natural selection to modify the gene frequencies within an individual by the process of differential growth. If the parts possessing the mutation grow faster the mutation may spread. If the mutation prevents successful growth then it will be eliminated. (Sutherland and Watkinson 1986, 305)

Mutations within the soma are subjected to immediate selection by the environment as they compete with the wild-type soma. Mutations in sequestered germlines, on the other hand, face only gametic selection, only once per generation. Buss (1983) schematizes the difference between somatic and gametic selection like this:

Somatic selection = mutation → selection → propagation → selection

Gametic mutation = Mutation → propagation → selection

There is one extra round of selection for somatic over gametic mutations. This extra phase of selection potentially increases the capacity for rapid evolutionary change among modular lineages. Buss says that IOS leads to "the disproportionate proliferation of those variants favoured by environmental demands" (Buss 1983, 1390). What is more, when heritable genetic variance exists between cells or other subunits, selection between them can result in within-organism evolution—gene frequency change within a generation.[9] The displacement of the wild type by a mutant in a mitotic cell lineage is evolution.

Somatic selection might be particularly important in the evolution of plants and other modular organisms, because of their high rates of mutation and the way they distribute resources around their parts. Although somatic selection may occur in any multicellular individual, it is especially prevalent in modular organisms because they are composed of a hierarchy of subunits, all of which can undergo selection. Darwinian populations can be found at multiple levels—groups whose members have heritable variation in character, which leads to differences in reproductive output (Godfrey-Smith 2009, 39).[10] In aspen, therefore, the following are all levels at which competition may take place simultaneously: between cells in a meristem stratum (in each stratum), between meristem strata, between modules/buds/shoots, between branches, between trees/ramets, between genets/forests.

Although many authors focus on conflict between levels, it is much more likely that selection at the various levels will act concordantly (Otto and Hastings 1998). In plants, somatic selection is supposed to work as follows: Multicellular transmission, as occurs in propagation by runners and in module iteration, preserves a high prevalence of mutations. Mutated cells are not segregated but can propagate themselves. During development or growth, mutant and wild-type lineages may have different growth rates. This means that different cell lineages can compete. How do they compete? In higher plants, they compete for *apical dominance*. Successful, that is, energy-efficient, modules (leaf-meristem units) produce hormones (auxin and others) that promote cell division while suppressing growth in other modules (Haukioja 1991). This allows plants to direct resources to their most successful parts. Fast-growing mutations can spread throughout the whole ramet or even genet by outcompeting the inferior alleles, that is, by producing sufficient auxin to cause the slower-growing modules or lineages to die. The developing buds of a plant compete for sunlight and nutrients as well as apical dominance. The poorest competitors will lose out and eventually die.

> Since a plant is a population of competing buds which grow at different rates and regenerate each year the gene frequency of the plant or clone can change over a period of years and the parent genotype may be completely lost. (Whitham and Slobodchikoff 1981, 289)

Evidence for IOS

Although IOS is not universally accepted as having long-term evolutionary consequences for plants, there is sufficient evidence for its existence and importance that its consequences for evolutionary theory need to be acknowledged.

A well-known example of IOS at work comes from variegated maple (*Acer platanoides drummondii*). The white edging on the leaves arises by somatic mutation and is deliberately preserved by horticulturists for its aesthetic appeal. The white parts contain no chlorophyll and so don't photosynthesize, lessening the leaf's overall efficiency. Often, buds will appear that contain wild-type mutations. Wild-type buds produce normal green leaves with a higher rate of photosynthesis. Left alone, the more efficient wild type will spread throughout the whole ramet (this is known as "reversion"), so horticulturists must continually remove wild-type buds to preserve the plant's variegation. This shows that the gene frequencies of the living plant tissues change over the lifetime of the plant. What is more, because the mutations can end up being propagated to new ramets, either sexually, if they end up in flowering parts, or vegetatively, this change in gene frequencies can have long-term evolutionary consequences.

Munch and Braun argued that the shape of trees provides evidence of interbranch selection, for if the branches were growing entirely independently of one another instead of competing for light and other resources, tree crowns ought all to take the shape of a witch's broom (White 1979). More evidence for IOS can be found in American goldenrods, or *Solidago missouriensis*. In one particular patch, the average age of goldenrod clones is 200 to 400 years, with some being as old as 1,000 years. The clones contain more than 10,000 ramets, each renewed annually, and are well adapted to local conditions as well as to closely related species. Whitham and Slobodchikoff note that a clone like this, according to the theory of diplontic selection, ought to have a high potential for genetic change over the lifespan of a clone. An individual clone certainly lives long enough to span evolutionary time. Somatic mutation during asexual ramet propagation could produce enough heritable variation for the clonal individual to adapt to local conditions. In fact, because there has only been time for eight to twenty-five generations of clones since the last glaciation in the area, sexual reproduction cannot reasonably account for their adaptive plasticity. Adaptive plasticity at the level of an individual clone can be better explained by adaptive evolution among the ramets—the physiological modules—from which the clone is composed (Whitham and Slobodchikoff 1981, 289).

There is more evidence for the evolutionary significance of IOS, and in order to understand it we need to look in closer detail at the structure of plants.

A Closer Look at Meristems

> The rate of diplontic selection can be increased by raising the number of initials or the number of generations of cell divisions. Accordingly, the progression from structured apical meristems with few initials to stochastic meristems containing many initials is a general trend both in the ontogeny and phylogeny of higher plants.[11] (Hughes 1989, 257)

Hughes suggests that the pattern of plant evolution shows us that the capacity for IOS (intraorganismal selection) may have been selectively favored. In this section, I introduce some details about plant meristem structure to explain what he means.

The rate of mutational meltdown, Klekowski (2003) explains, depends on the particularities of meristem organization. There are three different kinds of meristem organization in vascular plants.

Phylogenetically most ancient is the monopodial case, where a single apical meristem is the ultimate source of all the cells in the shoot. In these determinate meristems, each initial[12] has a permanent role, so that after mitosis of the cell one daughter cell always remains undifferentiated and functions as the subsequent initial. Thus an effective single-celled bottleneck exists in meristem iteration. This type of meristem is found in *Pteridophytes* (ferns, horsetails, and lycopods). Any mutation occurring in this meristem will be transmitted to all descendent cells. There is no diplontic selection because there is no pool of meristem cells able to compete with one another. Vegetative reproduction is common in pteridophytes, and mutational loads are very high.

A more phylogenetically recent meristem type is found in gymnosperms (e.g., cycads, gingko, and conifers). These stochastic meristems have a population of initial cells from which a few cells are probabilistically assigned to continue as the next generation of initials, whereas the others differentiate. "The apical initials divide mitotically a number of times giving rise to a pool of daughter cells from which subsequent initials are randomly sequestered" (Pineda-Krch and Fagerström 1999, 682). Klekowski points out that, where the shoot lacks permanent initials, diplontic selection will be more common. However, extensive vegetative reproduction is quite rare in this group, possibly because it is too easy for deleterious mutations to spread throughout the meristem.

Last, in angiosperms (flowering plants), meristems are stratified. This is the largest group of vascular plants, and also one in which vegetative or clonal growth is extremely common and has evolved separately multiple times. Many of the most invasive weeds are in this group and are spread vegetatively. Angiosperm meristems are totally different from those of gymnosperms and pteridophytes, and these differences impact on the retention and distribution of mutations in these plants.

Angiosperm meristems are organized into tunica-corpus systems. The outer parts (tunica) consist of one or many discrete layers of cells that divide anticlinally (perpendicular to the plane of the outer surface). Underneath these is the corpus, containing cells that divide in all planes and mostly differentiate into leaves and stem. "A shoot apical meristem consists of relatively isolated subpopulations of meristematic cells" (Klekowski 2003, 62).

Each meristem stratum contains several totipotent initials, so diplontic selection is maximized within the subpopulations. But this structure also allows stable periclinal (i.e., with layers parallel to surface) mosaics to form. A mutant can spread through its whole layer and stay there, so one will often find that each layer has a different mutation fixed within it. This stratification often results in the perpetuation of mosaic ramets containing mutated tissues, as well as the production of mutant gametes. Mutants can persist through many cycles of vegetative growth as periclinal mosaics, because the separation between layers protects them from diplontic selection. Each layer is a mini-Darwinian population, in Godfrey-

Smith's terms. Klekowski and Kazarina-Fukshansky (1984) note that the meristems in higher plants are actually not very adept at losing deleterious mutations, and meristem stratification may even promote the long-term accumulation of mutations. "Thus, paradoxically, angiosperm shoot apical meristems have evolved characteristics that reduce diplontic selection against defective somatic mutants in the short term" (Klekowski 2003, 63).

The layers, however, aren't totally isolated from each other. Anticlinal divisions within the corpus sometimes displace cells, by effectively injecting a cell from one layer into another. Herbivory is especially effective at upsetting the divisions between layers. When this happens, any variation that has been lying dormant is released, prompting diplontic selection between the layers.

Marcotrigiano (2000) hypothesizes that this could be particularly advantageous, allowing the plant to keep some variation up its sleeve and exploit it exactly when it is needed—to adapt to an environmental threat or simply regrow parts that have been eaten. Though Klekowski can offer no explanation for the evolution of stratified meristems (in fact, he says they appear "maladaptive from the viewpoint of buffering against disadvantageous mutations"; Klekowski, Kazarinova-Fukshansky, and Mohr 1985, 1794), Pineda-Krch and Fagerström (1999) provide a quantitative model demonstrating how stratified meristems enable an efficient and rapid elimination of deleterious mutant lineages while resulting in an increased probability of long-lived mosaic states.

Angiosperms, the newest and most successful of all plants, have evolved stratified meristem regions, which make intraorganismal selection maximally effective in three ways: They ensure that deleterious mutations are rapidly purged, they allow high fitness mutations to displace the wild type, and they preserve genetic variance over the long term (Pineda-Krch and Lehtilä 2002). This is what prompted Hughes to infer that the maintenance of IOS has actually been favored in plant evolution by higher-level selection.

Cell lineage selection is most effective in the absence of bottlenecks. Plant module iteration in monopodial plants is via a bottleneck. As you move upwards in the family tree of plant species, you see that meristems first became multicellular, and then stratified to produce several multicellular compartments within the module. Plants had bottlenecks, but they lost them, and then lost them even more. Why? A reasonable answer is that it encouraged cell lineage selection. In the next section, we will see why this might offer benefits.

Benefits of IOS

> Differences in the patterns of organization of organisms may lead to different patterns of evolution, genetics and ecology. Plants and animals differ in their fundamental patterns of organization. Plants may be able to take advantage of somatic mutations in ways that are not available to animals. (Whitham and Slobodchikoff 1981, 287)

It might be beneficial to an organism to have competition among its parts. Intraorganismal variation is essential to the functioning of the mammalian immune system, underlying the changes in B cells essential to acquired immunity to disease (French, Laskov, and Scharff,

1989). Cell competition may also play an important part in metazoan development, ensuring only the fittest cells make it into the germline (Khare and Shaulsky 2006). But the biggest advantages of somatic selection can be reaped in modular organisms like plants. IOS has been acknowledged as a way of eliminating deleterious somatic mutations that might otherwise accumulate, especially in clonal plants (Buss 1983; Gill 1986; Klekowski 1988; Otto and Orive 1995; Sutherland and Watkinson 1986; Whitham and Slobodchikoff 1981). So IOS may solve the puzzle of why clonal and asexual plants do not struggle under their mutational load—somatic selection is a mechanism that allows them to purge mutations without sex.

Under the genetic mosaic hypothesis (GMH) (Gill et al. 1995) it is proposed that mosaicism is favored in plants, especially, because it provides the individual with a broader phenotypic repertoire when dealing with pests and herbivores. A single plant may represent a mosaic of genotypes that prevents herbivores from evolving specific metabolic pathways to overcome plant defenses. Mosaicism thereby offers an advantage in the red queen race against pests and parasites. Clonal success deserves a parallel justification to those offered for the success of sexuality, and IOS seems to offer it. The GMH proposes to explain coevolution between long-lived plants and their short-lived enemies by saying that IOS allows an intragenerational response to herbivore pressure. For every resistant gene in wheat, for example, the Hessian fly must possess a corresponding gene for virulence. Changes at a single locus can thus have a drastic effect on plant-herbivore interactions (Whitham and Slobodchikoff 1981, 290).

IOS also endows modular organisms with phenotypic plasticity on an ecological time scale, allowing them to respond to changing environmental conditions (Monro and Poore 2004). For example, adventitious[13] buds are usually formed after a plant suffers some kind of physical damage. "This suggests that the derivation of these buds from a different meristematic layer may be an adaptive response in that it presents an alternative genotype in a changing environment" (Whitham and Slobodchikoff 1981, 291). Unitary organisms are very conservative in comparison.

Monro and Poore 2009 show that, given intraclonal variation, mitotic cell lineages rather than sexual offspring may act as units of selection. They describe an experiment in which intraclonal genetic variation allowed a red seaweed to evolve adaptively in response to a changing environment, in the absence of sex.

Much Ado about Nothing?

Several authors (Harper 1988; Hutchings and Booth 2004) have expressed skepticism about the evolutionary importance of IOS, arguing that it is a theoretical possibility that has yet to be empirically observed. Although more recent studies have since provided further empirical support (e.g., Khare and Shaulsky 2006; Monro and Poore 2004, 2009), the idea has yet to gain widespread acceptance within biology. Proponents of IOS (Fagerström 1992; Fagerström, Briscoe, and Sunnucks 1998; Pineda-Krch and Lehtilä 2004b; Poore

and Fagerström 2000; Santelices 1999; Tuomi 2004) have speculated that underlying theoretical biases are to blame for this, such as a misplaced allegiance to Weismann's barrier, or a limited focus on the metazoan phyla. To sound an appropriate note of academic caution, I mention that the status of IOS as a mechanism with the potential for long-term evolutionary consequences is still controversial.[14] This chapter should therefore not be viewed as an argument for the existence of IOS. That is an empirical matter. What I do want to argue is that *if* IOS does occur, *then* there are some important adjustments to be made in the way we talk about multilevel selection. But the case discussed here shows that this is not idle speculation. There is serious evidence for IOS, and thus the conflict assumption ought not to be uncritically accepted by multilevel theorists.

IOS and Multilevel Selection Theory

In this section, I look at how multilevel selection theory may need to be revised in light of the possibility of lower-level selection offering higher-level adaptive advantages.

The Contrast Again

> One lesson of multi-level theory is that the evolution of cooperative wholes requires suppression of competition among the parts. (Okasha 2006, 150)
>
> Conflicts must be mediated for the new higher-level unit (the multicellular group) to become a true individual. (Michod and Nedelcu 2003, 64)
>
> Group adaptation . . . only obtains if within-group selection is completely abolished. (Gardner and Grafen 2009, 666)

Most people are now happy to accept the possibility of multiple levels of selection, yet many still think that the presence of multiple levels entails some sort of conflict. It is assumed that the direction of fitness at different levels within a multilevel selection model will always be in opposition.[15] Not all authors endorse the claim that genetic homogeneity is required for higher-level entities to act as biological individuals. Homogeneity is rather just *one way* in which the threat of lower-level subversion can be averted. Gardner and Grafen (2009) insist that, in order for something to exhibit group-level individuality, there must *either* be relatedness of one between the group members, *or* some sort of policing mechanisms that totally suppresses selection between them. Policing mechanisms are often accepted as an alternative to genetic homogeneity, so long as they ensure that no within-group competition takes place at all. For example, Frank (2003) discusses the role of fair meiosis in the evolution of chromosomes, and worker policing behavior in insect colonies, while Michod views a complete reproductive division of labor as an adaptation for suppressing within-group competition. Michod (2005) in particular goes so far as to make the suppression of lower-level conflict part of the definition of what it is to be a higher-level individual.

In direct contrast to this, we saw in the earlier section that lower-level selection can, in the right circumstances, offer adaptive benefits at the higher level. We saw that modular plants tend to be genetic mosaics, and that there is plenty of scope for selection between the different parts of the plant, especially between cells found in different meristem compartments. There are no mechanisms in place for eliminating competition between different cell lineages. Nonetheless, angiosperm meristem structure seems to bear all the hallmarks of a higher-level adaptation, evolved for the benefit of the higher-level plant. Plants seem designed at the highest level for the purpose of maximizing their fitness (Gardner and Grafen 2009, 660).

Why Are Plants Different?

It is the modular structure of plants that means IOS can be adaptive for them. Having a modular organization makes something less vulnerable to mutations, because "problems" are contained within the cell lineage within which they arise. Mutations can proliferate by means of cell divisions, but they cannot spread to affect unmutated cell regions. Because modules are by their very essence semiautonomous or independent, they are able to continue carrying out their functions and life processes in the face of considerable deviation in the behavior of the other modules with which they may share a stem, root system, and so on. In a nutshell, problems in one part of a plant will not always impact the health or success of other parts.

Plant parts are more autonomous of one another than are the parts of metazoans and most other nonmodular organisms, largely because they have rigid walls that preclude moving around within the body of the plant. This usually prevents cancer from being fatal in plants (Doonan and Hunt 1996). But there are other features of plant physiology that limit the damage deleterious or free-riding mutations can do. We saw in detail how angiosperm meristems are structurally arranged so that mutations are compartmentalized, limiting their spread by somatic selection. Auxin and other hormones act to regulate the growth of plant modules relative to each other, favoring the fast-growing parts. Stoloniferous plants, in particular, are even able to move their parts around, away from outlaws. As we saw earlier, IOS might allow plants to purge deleterious mutations. Plants can even purge mutations by self-pruning to remove inefficient parts. For example, leaves drop off and die on becoming shaded by higher branches.

In fact, as long as a plant retains a few meristem cells supplied with resources, it can afford the death of all its other parts. Unitary organisms, in contrast, are much more vulnerable to interorganismal conflict. Their parts are very differentiated, so that if one part becomes defective there is overall loss of function and the whole suffers. IOS will often lead to cancers, and bottleneck life cycles mean that even minor developmental disruptions can be fatal. This is probably one reason why metazoans have evolved germ-soma separation and mechanisms that limit mutation. IOS is too risky for metazoans. Plants are a different matter, however. Their uniformity, and the retention of reproductive autonomy by

Table 11.1
Different possible combinations of benefit and cost at two levels

Module	Ramet	Example
+ Beneficial	+ Beneficial	Increased metabolic rate
+ Beneficial	– Deleterious	Flower degradation, cancers
− Deleterious	+ Beneficial	Germline segregation
− Deleterious	− Deleterious	Reduced metabolic rate
Neutral	Neutral	Most mutations

clonal ramets and meristems—their modularity, in effect—allows them to afford within-genet selection. Plants essentially don't need conflict suppression mechanisms because it is just not that easy to free ride in a plant. Plants can capitalize on this by using competition among plant modules to ensure that the fittest modules spread their genes throughout the organism, without risking the overall collapse of the organism.

In any case, not all mutations will be deleterious. We can construct a two-by-two classification scheme (table 11.1) to represent the effects of traits at two levels—the ramet and the module; +/+, +/−, −/+, −/− with the effect of the mutation on the cell given to the left and the effect of the mutation on the ramet given to the right (Otto and Hastings 1998).

The −/− mutations will go extinct in competition with higher fitness modules. The +/+ mutations will sweep through the module population if they are positive enough. Most mutations will be either +/+ or −/−, or have no effect whatsoever. Although there are well-known exceptions, such as cancers, most mutations will have an effect that is concordant between the lower and higher levels. The +/− and −/+ cases tend to be in the minority. Otto and Hastings (1998) explain this in terms of cell function. There are more ways in which function can be lost than ways in which it can be gained, and this tends to be detrimental at both the cellular and the higher level. Mutations that improve the efficiency of metabolic pathways, on the other hand, will be beneficial at both levels. What is good for the cell is good for the higher-level unit in most cases. In fact, angiosperm meristem structure raises the frequencies of all combinations, but then it filters all except the +/+ and +/− kind out. As indicated in these examples, some mutations will be good for the module but bad for the ramet. Flower degradation—loss of sexual fertility—is commonly found in plants with a high degree of clonal vigor, as is to be expected if rates of IOS are high (Dorken and Eckert 2001). Degradation of flowers (or any other mutation that adversely affects the capacity for sexual fertilization) is deleterious at the level of the ramet *only if* it is the case that sexual reproduction forms a necessary component of fitness. Cancers tend to be relatively unproblematic in plants because of the rigidity of cell walls. Still, there will be +/− cases that slip through the net. Fast growth rates may be achieved at the expense of investment in protection against herbivory, for example.

The contention here is that structure within the individual (specifically meristem structure in plants) may be organized in such a way that makes finding +/+ combinations easier. Selection within the individual, then, acts as a sieve, eliminating deleterious mutations and increasing the frequency of beneficial ones. This sets IOS apart from other modifiers of mutation, because they tend to alter the mutation rate in the same way regardless of the effect of the mutation. Altering the strength of IOS will increase the mutation rate for mutations that are beneficial to cell function and decrease it for mutations that are deleterious to cell division or replication. My contention is that IOS can act as a mechanism that increases the chances of finding novel +/+ mutations.

This point needs emphasizing because other authors assume that lower-level selection will come into conflict with higher-level selection for at least some, if not all, traits (Andy Gardner, personal correspondence). In the case of plants, I am not denying that what is best for the module (or cell) will sometimes diverge from what is best for the ramet (or genet), as the third row of table 11.1 indicates. Nonetheless, my argument is that so long as this happens in a minority of cases, and the consequences when it does are never very serious, the mechanism that encourages intraorganismal selection can still benefit the higher-level individual overall.

Homogeneity and Individuality

I have argued that there are some reasons to view plants as individuals at the highest ramet or genet levels, even though their parts may undergo selective processes. I have tried to refute the idea that stoloniferous genets cannot function as units of selection because they contain too much genetic heterogeneity and no conflict suppression mechanisms, and so will be riven by internal conflict and free-riding cell lineages. We saw that, thanks to their modular structure, plants can maintain higher-level functionality in the face of intraorganismal selection. In fact, meristem structure in angiosperms might constitute a higher-level adaptation for the very purpose of enhancing lower-level selection. So genetic heterogeneity and IOS do not provide evidence against higher-level individuality in plants.

The lesson I draw out is that multilevel selection theorists are wrong to use genetic homogeneity and conflict suppression mechanisms as the central criteria of individuality in plants and modular organisms. Michod is wrong to base the definition of individuality on conflict suppression mechanisms; Dawkins is wrong to tie it to genetic homogeneity. We should accept the possibility that biological individuals can have selection among their parts.

One consequence of rejecting genetic homogeneity as a criterion of individuation is that it leaves the way open for us to include as parts of an individual all sorts of genetically distinct entities such as symbiotes, organelles, and perhaps even parasites that have standardly been kept separate. Authors such as Dupre and O'Malley (2007) argue that gut flora and other microbes ought to be reconceptualized as comprising a genuine part of the human organism. My arguments imply that genetic disparity alone does not constitute an argument against such claims.

Where are we left regarding the individuality problem for modular organisms? To what unit—the module, ramet, genet, or perhaps all of the above—should fitness be ascribed? I argued that we ought not to use IOS as a reason against individuating plants at higher levels. The presence of genetic variance within a unit gives us no valid evolutionary reason for ruling out that unit as a level of selection. So at least one argument against individuating aspen at the higher genet level has been eliminated. I do not rule out the possibility of finding other arguments against individuating plants at higher levels. Neither do my claims constitute an argument against endorsing lower-level units of selection in plants, such as modules or meristems. The modular view conflicts with the genet view only if multiple simultaneous levels of selection are ruled out. In most real-life cases it will be reasonable to view multiple units as acting as simultaneous levels of selection. In this case, the best solution is to use something like Pedersen & Tuomi's hierarchical multilevel selection model (1995), which acknowledges and partitions multiple levels of selection.

In this section, I showed why it might be a mistake to think that homogeneity and conflict reduction mechanisms are at the center of our understanding of what it is to be an individual. Once we see that lower-level conflict does not necessarily undermine individuality at a higher level, we can reject arguments from the existence of genetic conflict to individuation at low levels of biological organization. The way is then open to build hierarchical selection models that fully incorporate multiple levels of selection. Plants and other modular organisms are ideal subjects for such models.

Conclusion

In plants and other modular organisms, there may be selection between higher-level units (such as genets and ramets) as well as selection between lower-level units (such as modules and meristem cells) and, *contra the usual assumptions of multilevel selection theory,* these two levels of selection need not be in opposition to each other.

Levels-of-selection theorists are wrong to assume that the central problem in transitions is always that of minimizing within-group competition. Evidence of intralevel conflict does not qualify as evidence against the existence of a higher level of selection.

Finally, plants and other modular organisms such as aspen may be hierarchical individuals, by which I mean that ramets and genets and modules are all simultaneous levels of selection.

Acknowledgments

I would like to thank the Arts and Humanities Research Council for their generous financial support. I also give many thanks to Samir Okasha, Peter Godfrey-Smith, Brett Calcott, Kim Sterelny, Torbjörn Fagerström, and Andy Gardner for commenting on various drafts.

Notes

1. Other authors mean the term "genet" to imply genetic homogeneity, but I use a developmental definition because, as we will see later, the unit that develops from a zygote rarely stays genetically homogeneous for long.

2. For example, see Ye, Yu, and Dong (2006).

3. De Sousa says individuals are spatiotemporally bounded and continuous (De Sousa 2005).

4. Although, of course, Janzen, who simply defines individuality via sex, would deny that vegetative propagation produces new individuals at all.

5. Furthermore, heterogeneity or variance need not be genetic—selection may also act on nongenetic differences between cells, such as methylation patterns. There is even evidence that such patterns may be heritable (Jablonka 2005).

6. Although see Foster et al. (2002) for discussion of how chimerism seems not to impede individuality in slime mold slugs and insect colonies.

7. Muller's ratchet is the name for a process by which the genome of an asexual population accumulates deleterious mutations over time. Muller suggested this as an explanation for the adaptive value of sex. His argument was that sexual recombination exposed deleterious alleles to selection, allowing them to be purged from the population, and reversing the action of the ratchet.

8. By "as a theoretical possibility" I mean to convey that, while it is universally accepted as a tautological truth, there is no consensus as to whether it acts in such a way that it has long-term evolutionary consequences.

9. Presuming that a mother cell and a mitotically derived daughter cell can be said to belong to different generations. Of course, it should be understood that gene frequencies can change over the course of these generations without any selection taking place, as a consequence of mutation alone. Once variation exists, however, evolution by somatic selection can occur.

10. There is room for further discussion about the extent to which each level constitutes a paradigm rather than marginal Darwinian population, in Godfrey-Smith's terms.

11. There is room for further discussion about the extent to which each level constitutes a paradigm rather than marginal Darwinian population, in Godfrey-Smith's terms.

12. An "initial" is simply a totipotent meristem cell. The word is used to distinguish these from cells in the meristem area that are on their way to becoming differentiated as soma.

13. "Adventitious" buds are those that develop on the roots, leaves, or stem—that is, anywhere other than the tip of a shoot.

14. The *Journal of Evolutionary Biology* devotes the whole of issue 17 to discussing intraorganismal genetic heterogeneity.

15. D. S. Wilson (1980) is an important exception, who has long argued that selection at different levels can act in either opposing or harmonious directions. See also Hadany (2001).

References

Bouchard, F. 2008. Causal processes, fitness, and the differential persistence of lineages. *Philosophy of Science* 75: 560–570.

Buss, L. W. (1983). Evolution, development and the units of selection. *Proceedings of the National Academy of Science USA* 80: 1387–1391.

Dawkins, R. (1982). *The Extended Phenotype*. Oxford: W.H. Freeman.

De Sousa, R. (2005). Individuality. *Croatian Journal of Philosophy* 5: 195–218.

Doonen, J., and T. Hunt. (1996). Why don't plants get cancer? *Nature* 380: 481–482.

Dorken, M. E., and C. G. Eckert. (2001). Severely reduced sexual reproduction in northern populations of a clonal plant, Decodon verticillatus (Lythraceae). *Journal of Ecology* 89: 339–350.

Dupre, J., and M. O'Malley. (2007). Metagenomics and biological ontology. *Studies in History and Philosophy of Science Part C: Studies in History and Philosophy of Biological and Biomedical Sciences* 38(4): 834–846.

Fagerström, T. (1992). The meristem-meristem cycle as a basis for defining fitness in clonal plants. *OIKOS* 63: 449–453.

Fagerström, T., D. A. Briscoe, and P. Sunnucks. (1998). Evolution of mitotic cell lineages in multicellular organisms. *Trends in Ecology and Evolution* 13: 117–120.

Foster, K. R., A. Fortunato, J. E. Strassmann, and D. C. Queller. (2002). The costs and benefits of being a chimera. *Proceedings of the Royal Society B, Biological Sciences* 269: 2357–2362.

Frank, S. A. (1995). Mutual policing and repression of competition in the evolution of cooperative groups. *Nature* 377: 520–522.

Frank, S. A. (2003). Repression of competition and the evolution of cooperation. *Evolution* 57(4): 693–705.

French, D. L., R. Laskov, and M. D. Scharff. (1989). The role of somatic hypermutation in the generation of antibody diversity. *Science* 244: 1152–1157.

Gardner, A., and A. Grafen. (2009). Capturing the superorganism: A formal theory of group adaptation. *Journal of Evolutionary Biology* 22: 659–671.

Gill, D. E. (1986). Individual plants as genetic mosaics: Ecological organisms versus evolutionary individuals. In M. J. Crawley (ed.), *Plant Ecology* (pp. 321–343). Oxford: Blackwell.

Gill, D. E., L. Chao, S. L. Perkins, and J. B. Wolf. (1995). Genetic mosaicism in plants and clonal animals. *Annual Review of Ecology, Evolution, and Systematics* 26: 423–444.

Godfrey-Smith, P. (2009). *Darwinian Populations and Natural Selection.* Oxford: Oxford University Press.

Hadany, L. (2001). A conflict between two evolutionary levels in trees. *Journal of Theoretical Biology* 208: 507–521.

Harper, J. L. (1977). *Population Biology of Plants*. New York: Academic Press.

Harper, J. L. (1988). An apophysis of plant population biology. In A. J. Davy, M. J. Hutchings and A. R. Watkinson (eds.), *Plant Population Ecology* (pp. 435–452). Oxford: Blackwell.

Haukioja, E. (1991). The influence of grazing on the evolution, morphology and physiology of plants as modular organisms. *Philosophical Transactions of the Royal Society B, Biological Sciences* 333: 241–247.

Hughes, R. N. (1989). *A Functional Biology of Clonal Animals. Functional Biology Series.* London: Chapman and Hall.

Hutchings, M. J., and D. Booth. (2004). Much ado about nothing . . . so far? *Journal of Evolutionary Biology* 17: 1184–1186.

Jablonka, E. (2005). *Evolution in Four Dimensions*. Cambridge, MA: MIT Press.

Janzen, D. H. (1977). What are dandelions and aphids? *The American Naturalist* 111: 586–589.

Jelinkova, H., F. Tremblay, and A. De Rochers. (2009). Molecular and dendrochronological analysis of natural root grafting in *Populus tremuloides*. *American Journal of Botany* 96: 1500–1505.

Khare, A., and G. Shaulsky. (2006). First among equals: Competition between genetically identical cells. *Nature Reviews Genetics* 7: 577–583.

Klekowski, E. J. (1998). *Mutation, Developmental Selection, and Plant Evolution*. New York: Columbia University Press.

Klekowski, E. J. (2003). Plant clonality, mutation, diplontic selection and mutational meltdown. *Botanical Journal of the Linnaean Society* 79: 61–67.

Klekowski, E. J., and N. Kazarinova-Fukshansky. (1984). Shoot apical meristems and mutation: Fixation of selectively neutral cell genotypes. *American Journal of Botany* 71: 22–34.

Klekowski, E. J., N. Kazarinova-Fukshansky, and H. Mohr. (1985). Shoot apical meristems and mutation: Stratified meristems and angiosperm evolution. *American Journal of Botany* 72: 1788–1800.

Marcotrigiano, M. (2000). Herbivory could unlock mutations sequestered in stratified shoot apices of genetic mosaics. *American Journal of Botany* 87: 355–361.

Maynard Smith, J., and E. Szathmáry. (1997). *The Major Transitions in Evolution*. Oxford: Oxford University Press.

Michod, R. E. (2005). On the transfer of fitness from the cell to the multicellular organism. *Biology and Philosophy* 20: 967–987.

Michod, R. E., and A. M. Nedelcu. (2003). On the reorganization of fitness during evolutionary transitions in individuality. *Integrative and Comparative Biology* 43: 64–73.

Monro, K., and A. Poore. (2004). Selection in modular organisms: Is intraclonal variation in macroalgae evolutionarily important? *The American Naturalist* 163: 564–578.

Monro, K., and A. Poore. (2009). The potential for evolutionary responses to cell-lineage selection on growth form and its plasticity in a red seaweed. *The American Naturalist* 173: 151–163.

Okasha, S. (2006). *Evolution and the Levels of Selection*. Oxford: Oxford University Press.

Otto, S. P., and I. M. Hastings. (1998). Mutation and selection within the individual. *Genetics* 102/103: 507–524.

Otto, S. P., and M. E. Orive. (1995). Evolutionary consequences of mutation and selection within an individual. *Genetics* 141: 1173–1187.

Pan, J. J., and J. S. Price. (2002). Fitness and evolution in clonal plants: The impact of clonal growth. *Evolutionary Ecology* 15: 583–600.

Pedersen, B., and J. Tuomi. (1995). Hierarchical selection and fitness in modular and clonal organisms. *OIKOS* 73: 167–180.

Pineda-Krch, M., and T. Fagerström. (1999). On the potential for evolutionary change in meristematic cell lineages through intraorganismal selection. *Journal of Evolutionary Biology* 12: 681–688.

Pineda-Krch, M., and K. Lehtilä. (2002). Cell lineage dynamics in stratified shoot apical meristems. *Journal of Theoretical Biology* 219: 495–505.

Pineda-Krch, M., and K. Lehtilä. (2004a). Costs and benefits of genetic heterogeneity within organisms. *Journal of Evolutionary Biology* 17: 1167–1177.

Pineda-Krch, M., and K. Lehtilä. (2004b). Challenging the genetically homogeneous individual. *Journal of Evolutionary Biology* 17: 1192–1194.

Poore, A., and T. Fagerström. (2000). Intraclonal variation in macroalgae: Causes and evolutionary consequences. *Selection* 1(1–3): 123–133.

Santelices, B. (1999). How many kinds of individual are there? *Trends in Ecology and Evolution* 14: 152–155.

Schlosser, G., and G. P. Wagner. (2004). *Modularity in Development and Evolution*. Chicago: University of Chicago Press.

Sutherland, W. J., and A. R. Watkinson. (1986). Do plants evolve differently? *Nature* 320: 305.

Tuomi, J. (2004). Genetic heterogeneity within organisms and the evolution of individuality. *Journal of Theoretical Biology* 17: 1182–1183.

Tuomi, J., and T. Vuorisalo. (1989a). What are the units of selection in modular organisms? *OIKOS* 54: 227–233.

Tuomi, J., and T. Vuorisalo. (1989b). Hierarchical selection in modular organisms. *Trends in Ecology and Evolution* 4: 209–213.

Watkinson, A. R., and J. White. (1986). Some life-history consequences of modular construction in plants. *Philosophical Transactions of the Royal Society B, Biological Sciences* 313: 31–52.

Weismann, A. (1893). *The Germ Plasm: A Theory of Heredity*. London: Walter Scott, Ltd.

White, J. (1979). The plant as a metapopulation. *Annual Review of Ecology, Evolution, and Systematics* 10: 109–145.

Whitham, T. G., and C. N. Slobodchikoff. (1981). Evolution by individuals, plant-herbivore interactions and mosaics of genetic variability: The adaptive significance of somatic mutations in plants. *Oecologia* 49: 287–292.

Wikberg, S. (1995). Fitness in clonal plants. *OIKOS* 72: 293–297.

Wilson, D. S. (1980). *The Natural Selection of Populations and Communities*. Menlo Park, CA: Benjamin/Cummings.

Winkler, E., and M. Fischer. (1999). Two fitness measures for clonal plants and the importance of spatial aspects. *Plant Ecology* 141: 191–199.

Ye, X. H., F. H. Yu, and M. Dong. (2006). A trade off between guerrilla and phalanx growth forms in Leymus secalinus under different nutrient supplies. *Annals of Botany* 98: 187–191.

12 Phylogenetic, Functional, and Geological Perspectives on Complex Multicellularity

Andrew H. Knoll and David Hewitt

Of all the events nominated as major transitions in evolution, none has received more attention than the rise of multicellularity. In part, this is because the subject can be approached from a number of perspectives, including systematics, developmental genetics, and the fossil record. And in part, of course, it is because multicellularity shapes our perceived biological landscape and, indeed, ourselves. In this chapter, we begin with a brief discussion of the phylogenetic distribution of multicellular organisms in general and complex multicellular life in particular, clarifying the important distinction between the two. We argue that multicellularity per se, the transfer of individuality from one cell to a coordinated group of attached cells (Michod 2007), was necessary for the emergence of complex multicellularity, but it was not sufficient. In the ensuing discussion we offer a biophysical answer to the question of what features distinguish the biomass- and species-rich clades of complex multicellular organisms from their simpler and less diverse sister groups. Only the animals, streptophyte green algae, florideophyte red algae, brown algae, and three clades of fungi have evolved active transport mechanisms for oxygen, nutrients, and signaling molecules that circumvent the strong constraints imposed by diffusion. A review of the Proterozoic fossil record shows that the radiations of complex multicellular animals and algae can be linked to an Ediacaran increase in the oxygen content of the atmosphere and oceans, confirming the historical link between diffusion and the evolution of complex multicellular organisms.

Phylogenetic Context

The first observation to make about multicellularity is that it does not occur in most eukaryotic clades. Patterson (1999) recognized seventy major clades of eukaryotes. Fifty-three of these are populated exclusively by unicellular organisms, predominantly motile predators on bacteria and other particle feeders, phytoplankton, and parasites. Most of these groups have few known species, conspicuous exceptions being the ciliates (which include some simple multicellular species), dinoflagellates, and granuloreticulosids (mainly

foraminiferans). Even the most diverse of these groups, however, fall two orders of magnitude short of the species richness documented for the most diverse multicellular clades.[1] Given the ecological and evolutionary success of single cells in these groups, the presence of multicellularity in Patterson's remaining seventeen clades requires functional explanation. Nine of the seventeen groups include simple filaments, hollow balls, sheets, or colonies organized by branching stalks. Patterson (1999) discussed stramenopiles (a large clade that includes the brown algae, diatoms, chrysophytes, and a number of other groups, both algal and heterotrophic), Viridoplantae (all green algae and their descendents, the embryophytic land plants), and opisthokonts (animals, fungi, and their close protistan relatives) as single entities, and subdivision of these megaclades increases this number to thirteen or more. Most are algal, although stalked colonial particle feeders have evolved at least three times, and simple filamentous fungi are absorptive heterotrophs. The widely discussed selective advantages of predator avoidance and maintaining position on a substrate or in the water column provide a reasonable functional explanation for most. In colonial heterotrophs such as the stalked ciliate *Epistylis* or mixotrophic chrysophytes, feeding may be facilitated by the coordinated beating of flagella. Complex multicellular organisms evolved from simple multicellular ancestors, but not all groups with simple multicells gave rise to complex descendents. In fact, most did not.

An additional four groups have achieved multicellularity via aggregation during one stage of the life cycle. Five others (two algal, three heterotrophic) have adopted coenocytial organization to achieve quasi-multicellularity and, in some cases, macroscopic size. There is no evidence that any of these evolved from simple multicellular ancestors; nor have any spawned complex multicellular descendents.

Collectively, simple multicellular organisms form a heterogeneous grouping that defies simple phylogenetic, functional, or developmental categorization. Indeed, we might choose to define many of these groups out of the discussion, depending on whether we insist that multicellularity include intercellular connections and/or cell differentiation. We prefer to deal them in, as they record important comparative biological information about the roads to (and in some cases, away from) complex multicellularity.

Most groups with simple multicellularity share several properties. Adhesive molecules (or, as in some filamentous diatoms, simple interlocking of wall protuberances) connect adjacent cells. Even with these physical attachments, however, communication and transfer of resources among cells and cellular differentiation are limited in some groups, absent in others. The multicellular state is both functional and persistent in these groups, and it appears to confer selective advantage. The penalty for loss of constituent cells, however, is commonly small. Michod (2006) modeled the emergence of multicellularity in terms of a downwardly convex curve relating fecundity and viability. Initially, increasing cell number may promote viability by, say, deterring predation; as long as most or all cells retain the capacity to reproduce, mean cell fitness will remain high. With increasing differentia-

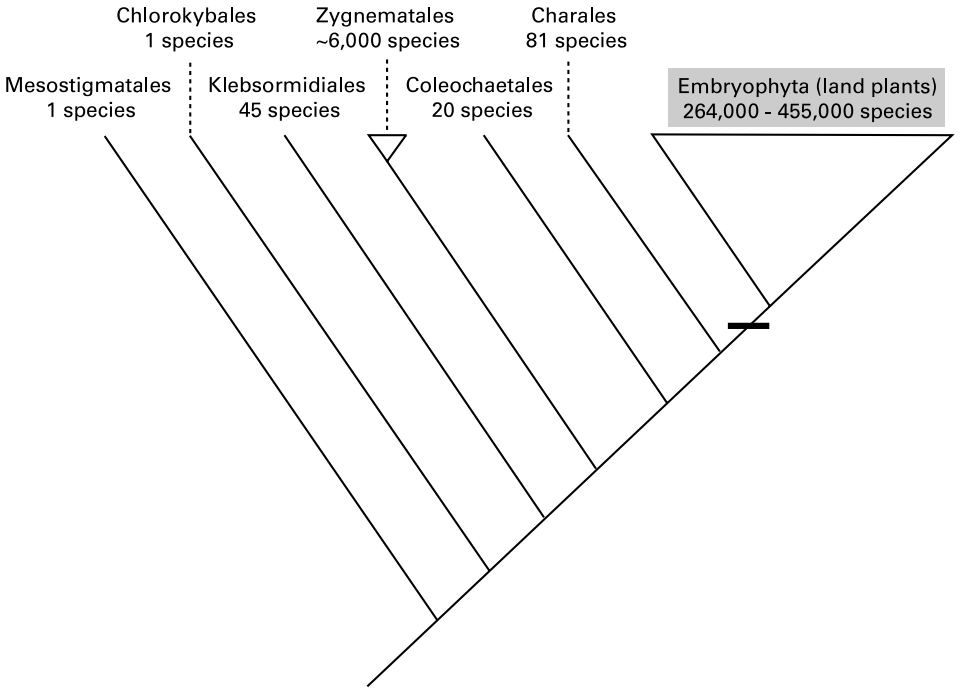
tion, however, mean cell fitness can decline even as mean cell viability continues to increase, due to reduction of per-cell fecundity—hence the convex shape of Michod's curve. As long as viability gain exceeds fecundity loss, overall fitness will increase, favoring multicellularity.

Importantly, for reasons discussed later, every cell in simple multicellular organisms is in direct contact with the external environment, at least during phases of the life cycle characterized by nutrient acquisition and active metabolism.

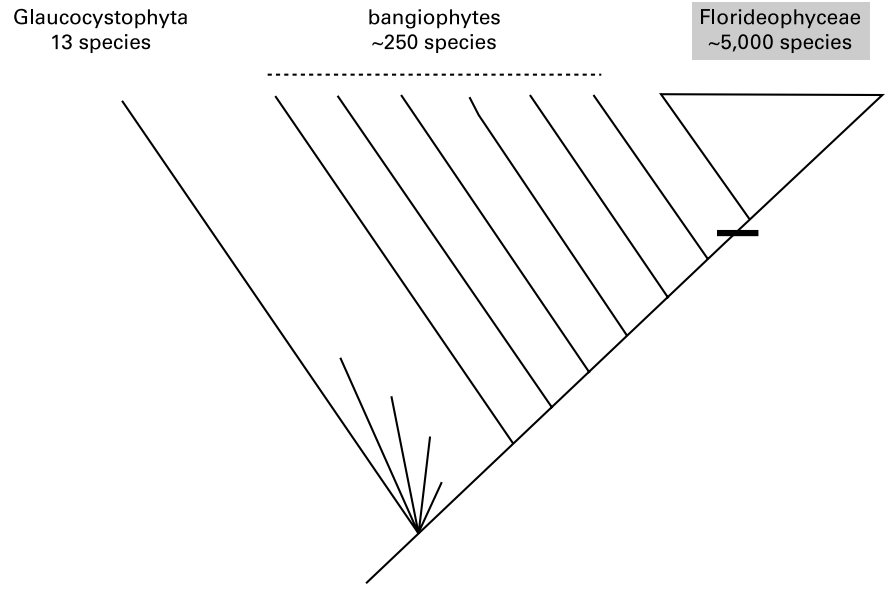
In contrast, complex multicellularity has evolved in animals, fungi (at least three times; Hewitt 2007; Schoch et al. 2009), green algae, red algae, and brown algae. Species in these clades show evidence of cell-cell adhesion, intercellular communication (e.g., gap junctions, plasmodesmata), and cellular and tissue differentiation mediated by networks of regulatory genes. Programmed cell death occurs in a number of these groups (as it does in some unicellular clades; see, e.g., Vardi et al., 1999), but unprogrammed cell or tissue loss can be lethal for the entire organism—perhaps more so in metazoans than in other groups with persistent stem cells. Notably, organisms in these clades display a multicellular organization in which only some cells are in direct contact with the environment.

The presence of exterior and interior cells is, in our view, critical. In consequence, cells do not all have equal access to nutrients and therefore will not accumulate biomass at a uniform rate—unless a mechanism evolves for the transfer of resources from one cell to another. Also, interior cells no longer receive signals directly from the environment, even though response to environmental dynamics remains key to growth, reproduction, and survival. Complex multicellular organisms, therefore, require mechanisms by which environmental signals can be received by surficial cells and transduced to interior cells, where genes will be up- or down-regulated in response. Of course, development in complex multicellular organisms can be defined as up- or down-regulation of genes in response to molecular signals from surrounding cells. In animals or plants, the effective environment of most cells is cellular, but signaling between adjacent cells may derive from fundamental mechanisms of signal transduction evolved in response to a need for life-history regulation (Knoll and Bambach 2000; Schlichting 2003; see below).

Only active transfer processes will free multicellular organisms with interior and exterior cells from the sharp constraints of molecular diffusion. Inspection of the three plots in figures 12.1 and 12.2 shows that, indeed, the circumvention of diffusion by active transport of metabolites and molecular signals differentiates biomass- and species-rich clades of eukaryotes from their less diverse sister groups. Within the Streptophyta (figure 12.1a), for example, the three basal clades of unicells (Mesostigmatales), sarcinoid colonies (Chlorokybales), and simple filaments (Klebsormidiales) have only a few dozen species among them. More distal branches include the Zygnematales [ca. 6,000 species of filamentous and (derived?) unicells] and their sister group, the megaclade characterized by complex branching, intercellular connections via plasmodesmata, and, in some cases at least, cells that


Chlorokybales
1 species
Zygnematales
~6,000 species
Charales
81 species
Mesostigmatales
1 species
Klebsormidiales
45 species
Coleochaetales
20 species
Embryophyta (land plants)
264,000 - 455,000 species

A


Glaucocystophyta
13 species
bangiophytes
~250 species
Florideophyceae
~5,000 species

B

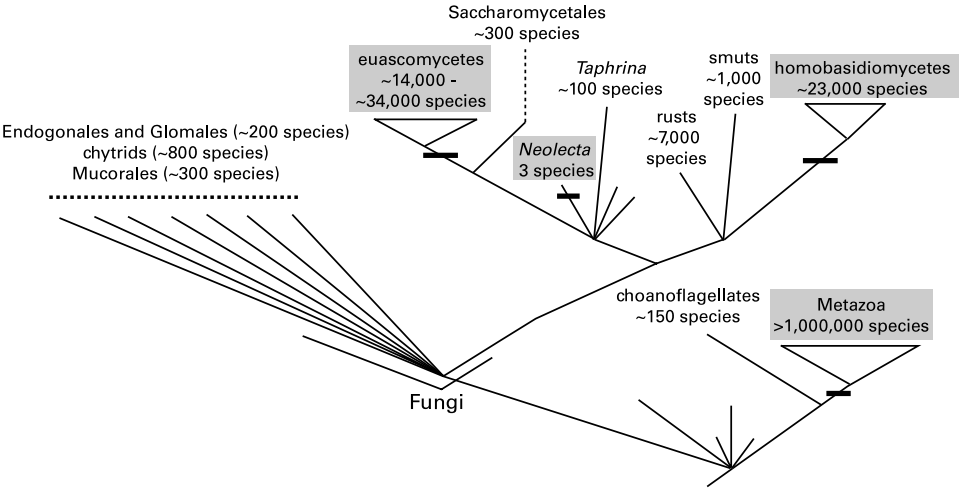


Figure 12.2
The distribution of species richness within the opisthokont clade. Bars indicate character evolution of three-dimensional multicellularity, with active transport of metabolites. Species-rich clades are characterized by complex multicellularity. Euascomycetes include the majority of the fruitbody-forming ascomycetes; homobasidiomycetes include the fruitbody-forming basidiomycetes. Phylogeny adapted from Hibbett et al. (2007); diversity values for fungal lineages from Mueller and Schmit (2007), except for *Taphrina* diversity, which is from Rodrigues and Fonseca (2003); metazoan diversity from Wilson (1999); choanoflagellate diversity from Maldonado (2004). Estimates are of named species and do not extrapolate to unnamed diversity.

are completely surrounded by other cells. This clade includes the Coleochaetales (ca. 20 species), Charales (81 species), and the most derived and three-dimensionally complex streptophytes, the embryophytes, with some 400,000 species and a collective biomass larger than all other eukaryotes combined. Similar patterns of species richness and biomass characterize the red (figure 12.1b) and brown algae, metazoans and their sister groups, and fungi (figure 12.2). High abundance and diversity reflect the functional possibilities of three-dimensional organization (Marshall 2005; Niklas 1994), shaped by evolvable networks of regulatory genes (Gerhart and Kirschner 2007), but fundamentally made possible by active transport mechanisms.

◀ **Figure 12.1**
The distribution of species richness within the Archaeplastida. Bars indicate character evolution of three-dimensional multicellularity, with active transport of metabolites. Species-rich clades characterized by complex multicellularity are noted by gray boxes. (A) Streptophytes, phylogeny adapted from Lewis and McCourt (2004), diversity values for algal streptophyte lineages from Turmel et al. (2007) and embryophyte diversity from Govaerts (2001; seed plant diversity: ~422,000; lower bound estimate cited as ~231,000), Schneider et al. (2004; fern diversity: ~10,000) and Renzaglia (2007; liverwort diversity: ~5,200, moss diversity: ~12,800, hornwort diversity: ~5,000). (B) Species diversity in the rhodophyte lineage; phylogeny adapted from Saunders and Hommersand (2004); diversity values from Algaebase taxonomic database (2007); Glaucocystophyte diversity from Andersen (1992). Estimates are for named species and do not extrapolate to unnamed diversity.

Diffusion and Its Circumvention

As noted earlier, three-dimensional anatomical structure has the functional consequence that many cells within the individual will not be in direct contact with their physical environment. Interior cells will not have access to the same nutrients or environmental signals as surface cells, and the extent of interior privation will be a direct function of diffusion.

Metabolism

Bacteria are metabolic machines adapted for a world of intermittent resource availability (e.g., Knoll and Bauld 1989). Fitness in bacteria is a strong function of metabolic rate when resources are present, and metabolic rate, in turn, reflects resource diffusion into the cell. Commonly, therefore, bacterial cells tend toward a high ratio of surface area to volume (by being small) and are everywhere in contact with the surrounding environment. Archaea appear to take this a step further, being adapted for environments where energy availability is limited (Valentine 2007). Under these conditions, it is not surprising that cell differentiation and complex, three-dimensional multicellularity are poorly developed in the prokaryotic domains.

Diffusion also constrains function in eukaryotic organisms, with P_{O2}, P_{CO2}, availability of fixed nitrogen, and concentrations of sugars or other metabolizable organic substrates all imposing potential constraints on size. Oxygen provides a particularly illuminating example, and is in fact unique as there are no transmembrane pumps that permit O_2 concentrations to build internally against a concentration gradient. Catling and colleagues (2005) have spelled out in detail why oxygen is a universal requirement for large multicellular organisms. Only the oxidation of organic molecules by O_2 provides sufficient energy to build a food chain that includes energy-intensive, macroscopic heterotrophs, and only oxygen in concentrations approaching those of the present day permits the oxygenation of interior cells in macroscopic organisms limited by diffusion.[2] Indeed, on this planet, no other oxidant exhibits both comparable abundance and energy gain from respiration; for large complex heterotrophs, there is no metabolic alternative to oxygen.

The constraints placed by oxygen diffusion on organism size were recognized nearly a century ago (Graham 1988; Krogh 1919). The thickness of cells or tissues that can be oxygenated by diffusion is a function of mass-specific metabolic rate (how rapidly oxygen is used) and the partial pressure of oxygen in the ambient environment (how much oxygen is available for use). In shallow marine waters in direct contact with today's atmosphere, O_2 diffusion limits tissue thickness to about 1 mm to 1 cm, depending on metabolic rate (figure 12.3; Raff and Raff 1970; Runnegar 1991). Severe or prolonged hypoxia causes cell death in animals, and it also appears to activate signaling pathways (Blackstone 2001), including those that cause unprogrammed cell proliferation (Harris 2001; Saul and Schwartz 2007).

Inward diffusion of metabolizable organic molecules will also limit tissue thickness in heterotrophs, and CO_2, light, and nutrient (e.g., nitrate or ammonia) penetration will do the

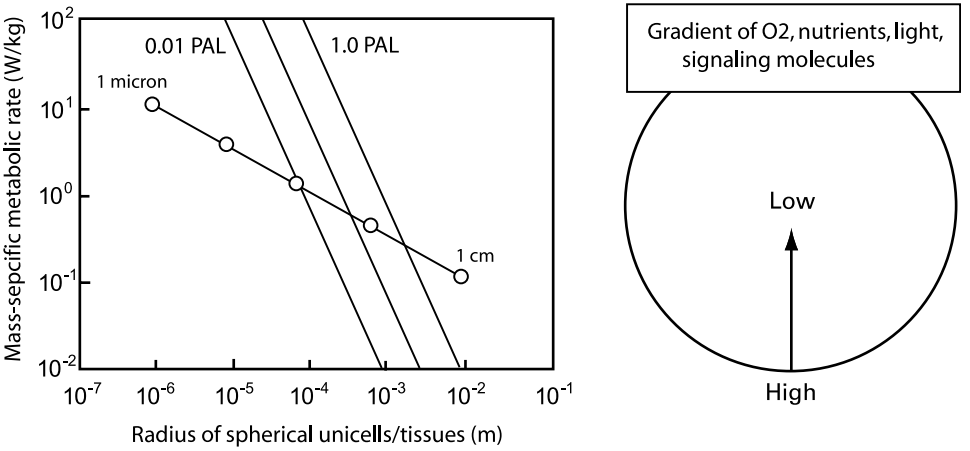


Figure 12.3
Diagram showing the empirically derived relationship between mass-specific metabolic rates for aerobically respiring organisms and the maximum radius of a spherical organism dependant on diffusion for the oxygenation of internal cytoplasm/cells; the high angle lines indicate the predicted limits on metabolic rate and size when (from right to left) P_{O2} is 1.0 times present atmospheric level (PAL), 0.1 PAL and 0.01 PAL (redrawn from Runnegar 1991). The cartoon on the right emphasizes the inward-decreasing gradient of metabolites and signaling molecules expected for diffusion-limited organisms.

same in autotrophs. (Even in the simple multicellular alga *Volvox*, coordinated beating of flagella is required to circumvent diffusive limitation by modifying the surface boundary layer; see Short et al. 2006.) The key point is that three-dimensionality potentially opens up a universe of new functional possibilities, but this potential can be realized only if resources needed for growth and reproduction can be transported among cells.

In green, red, and brown algae, intercellular transport is facilitated by plasmodesmata, or pit connections, strands of cytoplasm that contain endoplasmic reticulum, connecting adjacent cells via pits in cell walls (table 12.1). Algae also have membrane pumps that can move bicarbonate and other simple molecules across cell boundaries against a concentration gradient. Fungi commonly have incomplete cell walls that facilitate intercellular transfer of metabolites, whereas animals, lacking cell walls, have gap junctions and other protein-modulated modifications of cell membranes to govern molecular traffic between cells. Within these clades, some groups have evolved specialized cell and tissue types for long-distance transfer of metabolites: phloem in vascular plants, trumpet hyphae in brown algae, rhizomorphs in fungi (Cairney 2005) and complex circulatory systems in animals (table 12.1). Most fungi have the capacity for metabolite transfer across short distances, and fossils of meter-scale fungi that stood out like giants in Devonian landscapes contain networks of wide tubular cells that likely facilitated transfer of metabolites from mycelia to fruiting bodies (Boyce et al. 2007). Animals have also elaborated specialized surfaces (Graham 1988) to facilitate oxygen intake and oxygen-binding pigments that are transported throughout the body by fluids.

Table 12.1
Cell structures and cell/tissue types that promote active transport of metabolites and signaling molecules in complex multicellular eukaryotes

Taxon	Cell-Cell Connections	Differentiated Cells/Tissue
Streptophyte green algae/ embryophytes	Plasmodesmata	Sieve cells/phloem; leptoids, hydroids
Florideophyte red algae	Pit connections	
Brown algae	Plasmodesmata	Trumpet hyphae
Fungi	Septal pores, plasmodesmata	Rhizomorphs, vessel hyphae
Metazoans	Gap junctions	Circulatory system

Development

Schlichting (2003) recognized the consequences of three-dimensional growth for cell differentiation and, hence, development. As in the case of metabolism, these consequences stem from the fact that in three-dimensional multicellular organisms, only a subset of all cells is in direct contact with the environment. Many (but not all) of the signaling molecules employed by plants and animals occur in unicellular organisms found on closely related branches of phylogenies. For example, a number of gene families implicated in cell signaling and adhesion have been discovered in choanoflagellates (Abedin and King 2008; King, Westbrook and Young 2008; Segawa et al. 2006). Similarly, BIP2-like proteins known to regulate development in seed plants have been identified in the unicellular streptophyte *Mesostigma* (Nedelcu, Borza, and Lee 2006). MicroRNAs (miRNAs) that function in the regulation of vascular plant development have also been reported from the unicellular green alga *Chlamydomonas* (Molnar et al. 2007). For the most part, the functions of these protistan genes remain unknown, but many may be active in mediating morphological and cytological changes associated with life-cycle variation (King 2004; Knoll and Bambach 2000; Ruiz-Trillo et al. 2007; Schlichting 2003). An elegant example of this was provided by Nedelcu and Michod (2006), who showed that the gene *regA*, which regulates terminal somatic differentiation in *Volvox carteri*, has a homolog in its close unicellular relative *Chlamydomonas reinhardtii* that may function in life-cycle differentiation. Other signals adapted for multicellular development may have originated for signaling among cells in local populations, as seen, for example, in transient colony formation by the basal opisthokont *Sphaeroforma* (Ruiz-Trillo et al. 2007).

As Schlichting (2003) explained, three-dimensional growth has the consequence that interior cells will modify gene expression in response to external environmental change only if signals can be transduced to the interior from surficial cells that directly perceive environmental change. Moreover, the perceived physical and chemical environment of interior cells will differ from that of surface cells because nutrients, oxygen, and light will all be attenuated at depth. In a number of protists, nonmotile, commonly sexual stages of the

life cycle are induced by nutrient or oxygen privation, raising the possibility that simple cell differentiation in multicellular organisms might originally have occurred as a direct consequence of three-dimensional multicellularity. In this context, it is important to note that differentiation into two cell types is not an evolutionary milestone of multicellular organisms. Many unicellular organisms have life cycles in which different cell types alternate in time—the innovation of multicellular organisms is the spatial (and not only temporal) differentiation of cells.

Reactive oxygen species (ROS) act as developmental signals in both macroscopic and microscopic eukaryotes (Aguirre et al. 2005; Blackstone 2000), and, in animals at least, hypoxia-induced ROS can trigger the proliferation of cancerous cells (Saul and Schwartz 2007, and references therein). Thus, in early organisms with multiple cell layers, active regulation may have been necessary not so much to induce differentiation but to prevent, or at least control, spontaneous differentiation along an unprogrammed signaling gradient.

In effect, in three-dimensionally complex organisms, the ambient environment of interior cells is other cells, and genetic up- and down-regulation reflects perceived environmental gradients and signals from surrounding cells—the very definition of development. Moreover, signals generated by surface cells will diffuse inward, setting up a molecular gradient that induces different genetic responses in cells along the gradient—again the essence of development in plants and animals (Schlichting 2003). Active transport [for example, auxin transport in plants (Sieberer and Leyser 2006) or thyroid hormone action in animals (Feldt-Rasmussen and Rasmussen 2007), not to mention bulk transport by circulatory systems or vascular tissue] will increase the distance over which signaling can occur, again circumventing the limitations of diffusion and introducing topologically specific patterns of differentiation along the path of signal transport.

A Positive Feedback Loop

The obvious chicken-and-egg problem is whether size increase reflects or promotes the active transfer of nutrients, oxidants, and cell signals. The solution may be to consider the system of size, metabolism, and differentiation as a positive feedback loop.

Under a given set of environmental conditions (e.g., ambient oxygen and nitrogen concentrations), the size of an emerging multicellular organism will be limited, in the first instance, by diffusion. Plasmodesmata, gap junctions, or other mechanisms will increase the allowable thickness of tissues by facilitating the transfer of metabolites and signaling molecules between adjacent cells. The length scale on which such transfer will be effective, however, is small. Moreover, the degree to which oxygen and diffusible nutrients will be available to interior cells will remain a function of diffusion.

It is important, here, to reiterate that there are no transmembrane pumps that permit O_2 concentrations to build internally against a concentration gradient; therefore, oxygen is a special case among molecular requirements, in that bulk flow of dissolved oxygen is required for long-distance transport. As discussed later, this means that ambient oxygen

levels must have exerted an important control on size and shape in emerging multicellular organisms.

At small sizes, direct cell-cell connections may be sufficient to support interior cells in three-dimensional organisms. Increasing thickness, however, will enlarge the distance between surface and interior, setting up an increasingly strong gradient of nutrients and signaling molecules. As noted earlier, this gradient may itself promote cell differentiation, and cells that differentiate in ways that facilitate molecular transfer will potentiate further size increase (e.g., Bell and Mooers 1997). In this way, size and cell differentiation can populate a positive feedback loop, governed by metabolic and signaling gradients, that will eventually carry populations across a functional threshold—to the size and complexity of vascular plants, for example, or bilaterian animals.

A Geological Perspective

The fossil record for the first half of earth's history is poor. An Archaean origin for eukaryotes has been postulated on the basis of geochemical signatures for methanogenic Archaea in rocks as old as 2.7 billion years (Ga) and small subunit rRNA gene trees that place eukaryotes as sister to the archaeans. Consistent with this, steranes of probable eukaryotic derivation have been reported from 2.7-Ga rocks (Brocks et al. 1999). Both the phylogenetic relationships of Archaea and eukaryotes (Cox et al. 2008) and the syngenicity of Archaean steranes (Rasmussen et al. 2008; but see Waldbauer et al. 2009) remain topics of active debate. Moreover, at least some (hotly contested) models of eukaryogenesis rely on ur-symbiosis between a methanogen and facultatively aerobic proteobacterium. According to these models, eukaryotes cannot predate the origin of methanogenic archaeans, but could, in principle, postdate them by a long interval. Regardless of preferred scenario, however, the establishment of protists with mitochondria specialized for aerobic respiration—which appears to have occurred in stem group eukaryotes—postdates the initial accumulation of oxygen in the atmosphere and surface ocean some 2.4 Ga.

When the curtain rises on the fossil record later in the Paleoproterozoic Era, eukaryotes are already present (figure 12.4). Large ornamented unicells with wall ultrastructures known only among eukaryotes occur in 1800- to 1600-million-year-old (Ma) rocks, as do simple filaments of possible eukaryotic origin (Knoll et al. 2006). The oldest putative eukaryotic macrofossil, *Grypania*, is best known from 1600- to 1200-Ma deposits but occurs in rocks as old as 1,900 Ma (Han and Runnegar 1992). Based on the assumption that this organism was structurally and functionally similar to the extant green alga *Acetabularia*, Runnegar (1991) concluded that *Grypania* could have lived in shallow marine environments with oxygen levels only a few percent of present-day levels.

Bangiophyte red algae in about 1200-Ma cherts simultaneously record simple filamentous multicellularity, limited cell differentiation (including a cellular holdfast), and the

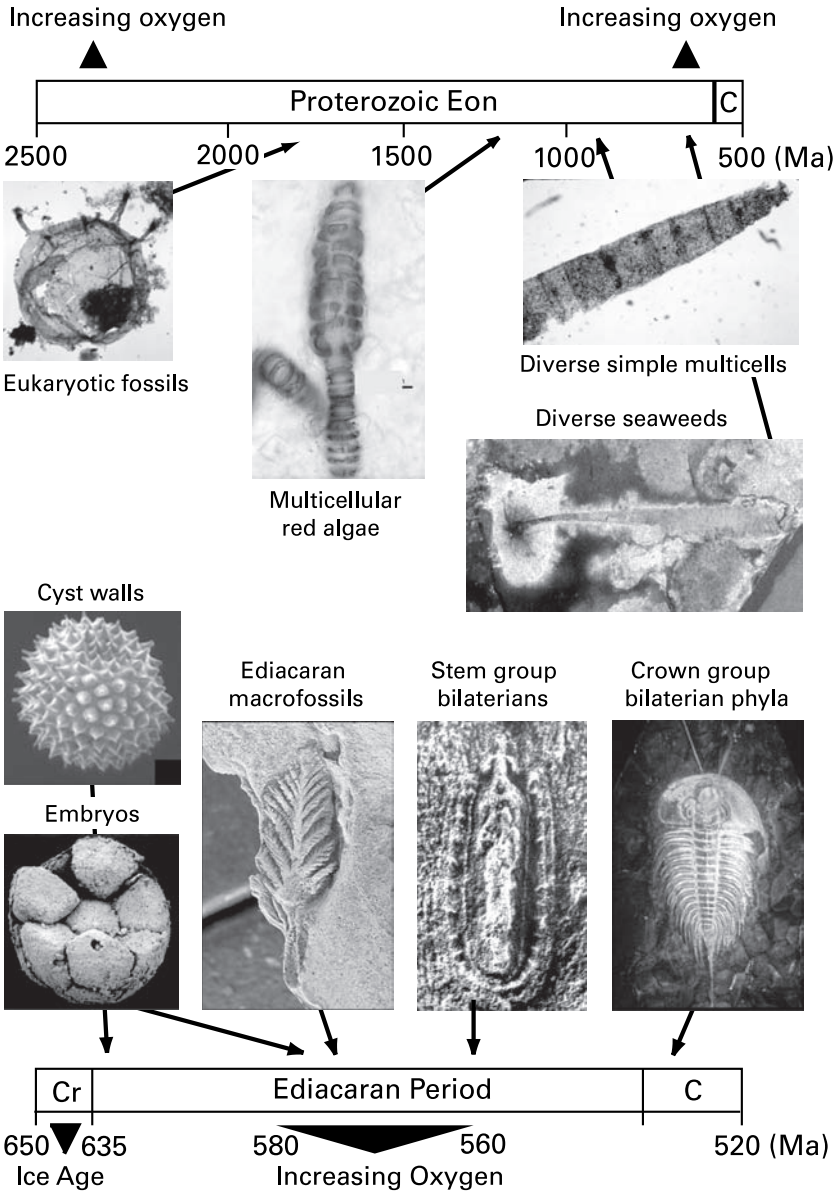


Figure 12.4
Diagram showing the stratigraphic record of simple and complex multicellular eukaryotes; see text and Knoll et al. (2006) for discussion.

appearance of a recognizable crown group member of a eukaryotic phylum (Butterfield 2000). In fact, simple eukaryotic filaments are common in fossil assemblages 1000 Ma and younger, and organisms with a three-dimensional arrangement of cells were present as early as about 800 million years ago (Knoll et al. 2006). Consistent with phylogenetic inference, such fossils suggest that the requirements for simple multicellularity were met early and often among eukaryotes (Knoll et al. 2006).

Despite the early appearance of simple multicellularity, records of complex multicellular organisms begin only near the end of the Proterozoic Era. Simple bangiophyte red algae may occur in 1200-Ma rocks, but florideophytes with three-dimensional anatomy are first recorded in 600- to 560-Ma phosphatic rocks of the Doushantuo Formation, in China (Xiao et al. 2004). Although simple filaments assigned to the cladophoran green algae occur in 800- 750-Ma rocks from Spitsbergen (Butterfield, Knoll, and Swett 1994), possible macroscopic greens first appear in 580- to 550-Ma shales (Steiner 1994; Xiao et al. 2002). Green algae with complex three-dimensional (coenocytic) architecture enter the record only in the Cambrian Period (Satterthwait 1976), whereas unequivocal embryophytes first appear in the Ordovician (Wellman and Gray 2000). More generally, macroscopic seaweeds with complex morphology began to spread across marine shelves and platforms only around 580 to 560 Ma (Xiao et al. 2002). On land, putative ascomycetes postdate the first appearances of embryophytes, but still occur in Lower Silurian rocks (Pratt, Phillips, and Dennison 1978; Sherwood-Pike and Gray 1985). Unequivocal records of complex multicellular fungi begin with silicified fossils in the earliest Devonian Rhynie Chert (Taylor, Hass, and Kerp 1999).

The earliest macroscopic heterotrophs are Ediacaran fossils preserved in basinal strata from Newfoundland, well dated at around 575 Ma (Narbonne 2005; figure 12.4). Many of these organisms have a distinctive architecture based on iterated tubes or cylinders. Their phylogenetic relationships are widely disputed, but stem group animals, or eumetazoans, constitute the current best estimate for systematic placement. As in many modern cnidarians, metabolically active tissue in these organisms may have been limited to a thin epithelium surrounding an inert fluid-filled or gelatinous interior. Thus, the "metabolic thickness" supported by inward diffusion of oxygen may have been considerably less than the whole-body thickness estimated from fossil casts and molds. Well-dated rocks from northern Russia place the origin of motile bilaterian animals a bit before 555 Ma (Martin et al. 2000), providing a minimum date for centimeter-scale animals with dense muscle tissues. Animals with biomineralized skeletons expanded globally at around 548 Ma (Grotzinger, Watters, and Knoll 2000), but only in the Cambrian did bilaterian animals with body plans comparable to those in present-day oceans diversify (Budd and Jensen 2000).

No unambiguous animal macrofossils predate 575 Ma, but microfossils push the metazoan record further back into time, and molecular biomarkers may push it even further. Phosphatic sedimentary rocks in southeastern China preserve the eggs and early cleavage-stage embryos (Xiao and Knoll 2000) of early, perhaps stem group (Hagadorn et al. 2006),

animals. These embryos occur stratigraphically below the first macroscopic Ediacaran heterotrophs seen regionally; they may or may not be older than the first Ediacarans in Newfoundland. The Chinese embryos occur in the same beds as abundant and diverse cysts of unusual size and both morphologic and ultrastructural complexity. These cysts are distinct from those known to be produced by algae, but share many similarities with cysts formed by animals that have a resting stage in their life cycles (Cohen, Kodner, and Knoll 2009). Indeed, Yin and colleagues (2007) recently discovered embryos within one type of cyst. These cysts have been found in rocks well dated at 632 Ma, deepening the geological record of animals by nearly 60 million years.

Still older animals are recorded by steranes, the geologically stable carbon skeletons of sterol molecules found in about 650-Ma shales from Oman (Love et al., 2009). Not all steranes are taxonomically diagnostic, but sponges constitute the most likely Neoproterozoic source of sterols with 24-isopropylcholestane skeletons (Kodner et al. 2008; Love et al. 2009). Found in some abundance in Oman shales, these molecules preserve a biological signature even though body fossils are absent.

We can argue forever about molecular clocks and depths of diversification they suggest (e.g., Rogers and Hug 2006; Bronham, this volume), but rapidly accumulating geochemical evidence indicates that pre-Ediacaran environments were, at best, only intermittently conducive to large aerobic heterotrophs (e.g., Fike et al. 2006; Canfield, Poulton, and Narbonne 2007; Canfield et al. 2008). Geochemical data suggest that the oxygen content of surface seawater was at least an order of magnitude below current levels during most of the Proterozoic Era; empirically, the oxygen minimum layer in Proterozoic oceans had a strong statistical tendency to become euxinic (e.g., Anbar and Knoll 2002; Arnold et al. 2004; Brocks et al. 2005; Canfield 1998; Scott et al. 2008; Shen, Knoll, and Walter 2003). Oxygen may have begun its second phase of increase early in the Neoproterozoic Era, but available evidence suggests that any Neoproterozoic trajectory was not monotonic. The global distribution of early diagenetic siderite in 800- to 580-Ma basins (Canfield et al. 2008; Johnston et al. 2010) strongly suggests that both oxygen and sulfate, the other great oxidant in modern oceans, were low at this time. Geochemical data further indicate that larger sulfate reservoirs and more persistently oxic deep waters began to appear about 580 Ma and stabilized within about 20 million years (Canfield et al. 2007, 2008; Fike et al. 2006; see figure 12.4), signaling the advent of oceans with relatively modern redox chemistry.

Observation (Rhoads and Morse 1971) and biophysical models (Runnegar 1991) indicate that the abundances of oxygen likely for surface water masses in pre-Ediacaran oceans (<10% present atmospheric level, PAL) were too low to support even millimeter-scale cell/tissue thickness in early, diffusion-limited animals. This doesn't require absence of metazoans from these oceans, but only means they would have to be small (or at least very thin). Given the impressive complement of animal signaling and adhesion molecules in sponges and cnidarians (Nichols et al. 2006; Putnam et al. 2007) and hypotheses that derive eumetazoans from larval sponges (Maldonado 2004), a good deal of genomic

evolution and divergence could have occurred within the animal clade before the appearance of macroscopic metazoan fossils.

As noted earlier, large animals appear by 575 Ma and motile bilaterians with elevated energy requirements are known to have existed soon thereafter. Interestingly, algae with complex multicellularity radiate at the same time, and biomarker molecules independently indicate that eukaryotes became major contributors to primary production only in the late Neoproterozoic Era (Knoll et al. 2007). Why should low oxygen have limited the evolution of complex multicellularity in algae, which, after all generate oxygen? One obvious consideration is that algae also respire aerobically and do so at night, when they are not generating oxygen. Thus, they are subject to the same constraints of oxygen diffusion that limit heterotrophs. Moreover, oceans with redox structure like that inferred for the Proterozoic Eon would have a nitrogen cycle far different from the one we know at present. Before 580 Ma, fixed nitrogen was likely in low supply in most marine photic zones (Anbar and Knoll 2002; Fennel, Follows, and Falkowski 2005); in its absence, algae would have competed poorly against nitrogen-fixing photosynthetic bacteria.

The foregoing observations suggest a close biophysical relationship between earth's redox chemistry and the emergence of complex multicellular organisms. Prior to the Ediacaran Period, P_{O2} and, additionally for autotrophs, nutrient availability limited the number of functioning cell layers in eukaryotic organisms. Rising oxygen levels alleviated this constraint by 580 to 560 Ma. The order of magnitude increase in P_{O2} inferred for this interval would have allowed the maximum volume of diffusion-limited organisms to increase by up to three orders of magnitude (see figure 12.2), providing ample raw material for increased anatomical complexity (cf. Bell and Mooers 1997). That is, the latest Proterozoic rise in oxygen would have given the nudge required to set in motion the positive feedback loop of size, transport, and differentiation.

In short, the long apparent lag between the appearance of simple multicellularity in eukaryotes and the radiation of groups with complex multicellular organization has an environmental component that can be linked back to the consequences of life with interior and exterior cells. Put another way, the physical environment within which natural selection occurs has changed through time in ways that greatly altered the selective value of mutations favoring three-dimensional multicellularity, with a latest Proterozoic state-shift changing the adaptive landscape irrevocably.

Discussion and Conclusion

Having outlined the biophysical requirements for complex multicellularity, we end where we began—with phylogeny. Within the eukaryotes, not all unicellular clades gave rise to simple multicells, and not all groups with simple multicellularity gave rise to organisms with complex multicellularity. Yet, complex multicellular eukaryotes dominate both species diversity and biomass within the domain.

Why have only a few eukaryotic clades evolved complex multicellularity? Understanding why some things don't evolve is challenging, as exemplified by Richard Lewontin's (2003) famous question: Why are there no grass-eating snakes? In combination with phylogeny, however, the positive feedback scenario outlined in this chapter may be instructive.

Complex multicellularity requires adhesion between cells, communication between cells, and a genetic program for cell differentiation. The capacity of cells to respond to external signals by remodeling is widespread among unicellular eukaryotes, suggesting that at least the rudiments of developmental program predate multicellularity. Molecules that promote cell-cell adhesion also occur widely in originally unicellular clades, where these molecules may well serve other functions (e.g., King, Westbrook and Young 2008; Sebe-Pedros et al. 2010).

Given these distributions, two key steps stand out as central to the emergence of complex multicellularity. First was the establishment of mechanisms for active molecular transport between adjacent cells. Of the seventeen eukaryotic clades known to include simple multicellular organisms, only five have plasmodesmata, gap junctions, or incomplete cell walls that facilitate cell-cell communication, and all complex multicellular organisms belong to these five clades.

The second required step, achieved in all groups with intercellular transport mechanisms, was the differentiation of mechanisms for the bulk transfer of oxygen through tissues. The circumvention of diffusion made possible by these innovations provided the jumping-off point for functionally and anatomically complex multicellular organisms.

This perspective reemphasizes a point made earlier by McShea (2002) and others: the evolutionary transition from unicells to complex multicellular organisms has several steps; it is a corridor, not a door. The entrance is marked by genetically mediated, geometrically regular multicellular colonies that derive some fitness benefit from their geometry but pay little penalty for ontogenetic mistakes. Marcot and McShea (2007) have rightly viewed this as a minor transition, as it is commonly reversible.[3] Simple multicells appear to be prerequisites for complex multicellular organisms, but they are not sufficient. The required boost comes from three-dimensionality. Positive feedbacks involving size, signal and materials transfer, and development propel clades toward the exit of the corridor, marking the truly major transition to complex multicellularity. Development underpins the establishment of "irreducible complexity" (Michod 2007), where unprogrammed cell proliferation or death can impose a severe penalty on fitness.

Of course, development also makes possible the innovations in structural support, food acquisition, and reproduction that collectively explain the high species richness and extensive biomass of complex multicellular organisms.

In conjunction with biophysical observation and modeling, the geologic record suggests that another door had to be opened along the path to complex multicellularity, this one environmental. Organismic fitness depends on environment as well as phenotype, and physical environments conducive to large size and developmental complexity may not

have appeared until life on earth was already more than 3 billion years old. By evolving active transport mechanisms that circumvent the strong biophysical constraints of diffusion, complex multicellular organisms reshaped the biosphere.

Acknowledgments

We thank participants in the KLI workshop on major transitions for stimulating discussion, and both Kim Sterelny and Jochen Brocks for helpful reviews of our manuscript. Research leading to this chapter was supported in part by NSF grant EAR-0420592.

Notes

1. Very likely, protistan diversity remains largely undocumented, with many experts estimating true species numbers an order of magnitude larger than those already described (e.g., the diatoms, with 10,000 described and 100,000 estimated species, many of them simple filaments; Kooistra et al. 2007). The same, however, is true of the animals and complex fungi, so that protistan diversity would need to be underdescribed by more than two orders of magnitude for unicellular and simple multicellular species to match the diversity of complex multicellular species.

2. Under the specific conditions where fermentable substrates are present in high concentration, the high rates of ATP production by fermenters may provide selective advantage over respirers in the same environment (Pfeiffer, Schuster, and Bonhoeffer 2001), but at the time(s) when multicellularity arose, such environments must have been a small subset of available habitats. Yeasts, for example, are thought to have evolved only 120 to 165 million years ago, in response to the evolution of sugar-rich angiosperm fruits (Thomson et al. 2005).

3. Diatoms provide an illustrative example. Many diatom species occur in nature as chains (or, less commonly, fans) of cells, presumably to evade predation and/or to maintain a particular position with respect to their environment. Breakdown of chains in culture is common, causing no ill effects on populations. Interestingly, the one major diatom clade made up almost exclusively of unicells is the raphid pennates, a highly derived clade characterized by flagellar motion of vegetative cells (Kooistra et al. 2007).

References

Abedin, M., and N. King. (2008). The premetazoan ancestry of cadherins. *Science* 319: 946–948.

Aguirre, J., M. Ríos-Momberg, D. Hewitt, and W. Hansberg. (2005). Reactive oxygen species and development in microbial eukaryotes. *Trends in Microbiology* 13: 111–118.

Algaebase Taxonomic Database. (2007). Accessed August 2007, at <http://www.algaebase.org>.

Anbar, A. D., and A. H. Knoll. (2002). Proterozoic ocean chemistry and evolution: a bioorganic bridge? *Science* 297: 1137–1142.

Andersen, R. A. (1992). Diversity of eukaryotic algae. *Biodiversity and Conservation* 1: 267–292.

Arnold, G. L., A. D. Anbar, J. Barling, and T. W. Lyons. (2004). Molybdenum isotope evidence for widespread anoxia in mid-Proterozoic oceans. *Science* 304: 87–90.

Bell, G., and A. O. Mooers. (1997). Size and complexity among multicellular organisms. *Biological Journal of the Linnean Society* 60: 345–363.

Blackstone, N. W. (2000). Redox control and the evolution of multicellularity. *BioEssays* 22: 947–953.

Blackstone, N. W. (2001). Redox state, reactive oxygen species and adaptive growth in colonial hydroids. *Journal of Experimental Biology* 204: 1845–1853.

Boyce, C. K., C. L. Hotton, M. L. Fogel, G. D. Cody, R. M. Hazen, A. H. Knoll, and F. M. Hueber. (2007). Devonian landscape heterogeneity recorded by a giant fungus. *Geology* 35: 399–402.

Brocks, J. J., G. A. Logan, R. Buick, and R. E. Summons. (1999). Archean molecular fossils and the early rise of eukaryotes. *Science* 285: 1033–1036.

Brocks, J. J., G. D. Love, R. E. Summons, A. H. Knoll, G. A. Logan, and S. Bowden. (2005). Biomarker evidence for green and purple sulfur bacteria in an intensely stratified Paleoproterozoic ocean. *Nature* 437: 866–870.

Budd, G. E., and S. Jensen. (2000). A critical reappraisal of the fossil record of the bilaterian phyla. *Biology Review* 75: 253–295.

Butterfield, N. J. (2000). *Bangiomorpha pubescens* n. gen., n. sp.: Implications for the evolution of sex, multicellularity and the Mesoproterozoic/Neoproterozoic radiation of eukaryotes. *Paleobiology* 26: 386–404.

Butterfield, N. J., A. H. Knoll, and K. Swett. (1994). Paleobiology of the Neoproterozoic Svanbergfjellet Formation, Spitsbergen. *Fossils and Strata* 34: 1–84.

Cairney, J. W. G. (2005). Basidiomycete mycelia in forest soils: Dimensions, dynamics and roles in nutrient distribution. *Mycological Research* 190: 7–20.

Canfield, D. E. (1998). A new model for Proterozoic ocean chemistry. *Nature* 396: 450–453.

Canfield, D. E., S. W. Poulton, T. Goldberg, A. H. Knoll, G. M. Narbonne, G. Ross, and H. Strauss. (2008). Ferruginous conditions dominated later Neoproterozoic deep-water chemistry. *Science* 321: 949–952.

Canfield, D. E., S. W. Poulton, and G. M. Narbonne. (2007). Late-Neoproterozoic deep-ocean oxygenation and the rise of animal life. *Science* 315: 92–95.

Catling, D. C., C. R. Glein, K. J. Zahnle, and C. P. McKay. (2005). Why O_2 is required by complex life on habitable planets and the concept of planetary "oxygenation time." *Astrobiology* 5: 415–438.

Cohen, P. A., R. Kodner, and A. H. Knoll. (2009). Ediacaran acritarchs as animal resting cysts. *Proceedings of the National Academy of Sciences USA* 106: 6519–6524.

Cox, C. J., P. G. Foster, R. P. Hirt, S. R. Harris, and T. M. Embley. (2008). The archaebacterial origin of eukaryotes. *Proceedings of the National Academy of Sciences USA* 105: 20356–20361.

Feldt-Rasmussen, U., and A. K. Rasmussen. (2007). Thyroid hormone transport and actions, in G. E. Krassas, S. A. Rivkees, and W. Kiess (eds.), *Diseases of the Thyroid in Childhood and Adolescence, Vol. 11: Pediatric Adolescence Medicine*, pp. 80–103. Basel: Karger.

Fennel, K., M. Follows, and P. G. Falkowski. (2005). The co-evolution of the nitrogen, carbon and oxygen cycles in the Proterozoic ocean. *American Journal of Science* 305: 526–545.

Fike, D. A., J. P. Grotzinger, L. M. Pratt, and R. E. Summons. (2006). Oxidation of the Ediacaran ocean. *Nature* 444: 744–747.

Gerhart, J., and M. Kirschner. (2007). The theory of facilitated variation. *Proceedings of the National Academy of Sciences USA*, 104(Suppl.): 8582–8589.

Govaerts, R. (2001). How many species of seed plants are there? *Taxon* 50: 1085–1090.

Graham, J. B. (1988). Ecological and evolutionary aspects of integumentary respiration: Body size, diffusion, and the Invertebrata. *American Zoologist* 28: 1031–1045.

Grotzinger, J. P., W. A. Watters, and A. H. Knoll. (2000). Calcified metazoans in thrombolite-stromatolite reefs of the terminal Proterozoic Nama Group. *Namibian Paleobiology* 26: 334–359.

Hagadorn, J. W., S. Xiao, P. C. J. Donoghue, et al. (2006). Cellular and subcellular structure of Neoproterozoic animal embryos. *Science* 314: 291–294.

Han, T. M., and B. Runnegar. (1992). Megascopic eukaryotic algae from the 2.1-billion-year-old Negaunee Iron-Formation, Michigan. *Science* 257: 232–235.

Harris, A. L. (2001). Hypoxia—a key regulatory factor in tumour growth. *Nature Reviews: Cancer* 2: 38–47.

Hewitt, D. (2007). Rise Above: Origins, Diversity, and Development of Complex Multicellularity in the Ascomycota. PhD thesis, Harvard University, Cambridge, MA.

Hibbett, D. S., et al. (2007). A higher-level phylogenetic classification of the Fungi. *Mycological Research* 111: 509–547.

Johnston, D. T., S. W. Poulton, C. Dehler, S. Porter, J. Husson, D. E. Canfield, and A. H. Knoll. (2010). An emerging picture of Neoproterozoic ocean chemistry: Insights from the Chuar Group, Grand Canyon, USA. *Earth and Planetary Science Letters* 290: 64–73.

King, N. (2004). The unicellular ancestry of animal development. *Developmental Cell* 7: 313–25.

King, N., et al. (2008). The genome of the choanoflagellate *Monosiga brevicollis* and the origin of metazoans. *Nature* 451: 783–788.

Knoll, A. H., and R. K. Bambach. (2000). Directionality in the history of life: Diffusion from a left wall or repeated scaling of the right? *Paleobiology* (Suppl.) 26(4): 1–14.

Knoll, A. H., and J. Bauld. (1989). The evolution of ecological tolerance in prokaryotes. *Transactions of the Royal Society of Edinburgh* 80: 209–223.

Knoll, A. H., E. J. Javaux, D. Hewitt, and P. Cohen. (2006). Eukaryotic organisms in Proterozoic oceans. *Philosophical Transactions of the Royal Society, B* 361: 1023–1038.

Knoll, A. H., R. E. Summons, J. Waldbauer, and J. Zumberge. (2007). The geological succession of primary producers in the oceans. In P. Falkowski and A. H. Knoll (eds.), *The Evolution of Primary Producers in the Sea* (pp. 133–163). Burlington, MA: Elsevier.

Kodner, R. B., R. E. Summons, A. Pearson, N. King, and A. H. Knoll. (2008). Sterols in a unicellular relative of the metazoans. *Proceedings of the National Academy of Sciences USA* 105: 9897–9902.

Kooistra, W. H. C. F., R. Gersonde, L. K. Medlin, and D. G. Mann. (2007). The origin and evolution of the diatoms: Their adaptation to a planktonic existence. In P. Falkowski and A. H. Knoll (eds.), *The Evolution of Primary Producers in the Sea* (pp. 207–249). Burlington, MA: Elsevier.

Krogh, A. (1919). The rate of diffusion of gases through animal tissues, with some remarks on the coefficient of invasion. *Journal of Physiology* 52: 391–408.

Lewis, L. A., and R. M. McCourt. (2004). Green algae and the origin of land plants. *American Journal of Botany* 91: 1535–1556.

Lewontin, R. (2003). Four complications in understanding the evolutionary process. *Santa Fe Bulletin* 18: 20–26.

Love, G. D., et al. (2009). Fossil steroids record the appearance of Demospongiae during the Cryogenian period. *Nature* 457: 718–721.

Maldonado, M. (2004). Choanoflagellates, choanocytes, and animal multicellularity. *Invertebrate Biology* 123: 1–22.

Marcot, J. D., and D. W. McShea. (2007). Increasing hierarchical complexity through the history of life: phylogenetic tests of trend mechanisms. *Paleobiology* 33: 182–200.

Marshall, C. R. (2005). Explaining the Cambrian "explosion" of animals. *Annual Review of Earth and Planetary Sciences* 34: 355–384.

Martin, M. W., D. V. Grazhdankin, S. A. Bowring, D. A. D. Evans, M. A. Fedonkin, and J. L. Kirschvink. (2000). Age of Neoproterozoic bilaterian body and trace fossils, White Sea, Russia: Implications for metazoan evolution. *Science* 288: 841–845.

McShea, D. W. (2002). A complexity drain on cells in the evolution of multicellularity. *Evolution* 56: 441–452.

Michod, R. E. (2006). The group covariance effect and fitness trade-offs during evolutionary transitions in individuality. *Proceedings of the National Academy of Sciences USA* 103: 9113–9117.

Michod, R. E. (2007). Evolution of individuality during the transition from unicellular to multicellular life. *Proceedings of the National Academy of the Sciences USA* 104(Suppl.): 8563–8676.

Molnar, A., F. Schwach, D. J. Studholme, E. C. Thuenemann, and D. C. Baulcombe. (2007). MiRNAs control gene expression in the single-cell alga *Chlamydomonas reinhardtii*. *Nature* 447: 1126–1129.

Mueller, G. M., and J. P. Schmit. (2007). Fungal biodiversity: What do we know? What can we predict? *Biodiversity and Conservation* 16: 1–5.

Narbonne, G. M. (2005). The Ediacara biota: Neoproterozoic origin of animals and their ecosystems. *Annual Review of Earth and Planetary Sciences*, 33: 421–442.

Nedelcu, A. M., T. Borza, and R. W. Lee. (2006). A land plant-specific multigene family in the unicellular *Mesostigma* argues for its close relationship to Streptophyta. *Molecular Biology and Evolution* 23: 1111–1015.

Nedelcu, A. M., and R. E. Michod. (2006). The evolutionary origin of an altruistic gene. *Molecular Biology and Evolution* 23: 1460–1464.

Nichols, S. A., W. Dirks, J. S. Pearse, et al. (2006). Early evolution of animal cell signaling and adhesion genes. *Proceedings of the National Academy of Sciences USA* 103: 12451–12456.

Niklas, K. J. (1994). Morphological evolution through complex domains of fitness. *Proceedings of the National Academy of Sciences USA* 91: 6772–6779.

Patterson, D. J. (1999). The diversity of eukaryotes. *The American Naturalist* 154(Suppl.): S96–S124.

Pfeiffer, T., S. Schuster, and S. Bonhoeffer. (2001). Cooperation and competition in the evolution of ATP-producing pathways. *Science* 292: 504–507.

Pratt, L. M., T. L. Phillips, and J. M. Dennison. (1978). Evidence of non-vascular land plants from Early Silurian (Llandoverian) of Virginia, USA. *Review of Palaeobotany and Palynology* 25: 121–149.

Putnam, N. H., et al. (2007). Sea anemone genome reveals ancestral eumetazoan gene repertoire and genomic organization. *Science* 317: 86–94.

Raff, R. A., and E. C. Raff. (1970). Respiratory mechanisms and the metazoan fossil record. *Nature* 228: 1003–1005.

Rasmussen, B., I. R. Fletcher, J. J. Brocks, and M. R. Kilburn. (2008). Reassessing the first appearance of eukaryotes and cyanobacteria. *Nature* 455: 1101–1104.

Renzaglia, K. S. et al. (2007). Bryophyte phylogeny: Advancing the molecular and morphological frontiers. *Bryologist* 110: 179–213.

Rhoads, D. C., and J. W. Morse. (1971). Evolutionary and ecologic significance of oxygen-deficient marine basins. *Lethaia* 4: 413–428.

Rodrigues, M. G., and A. Fonseca. (2003). Molecular systematics of the dimorphic ascomycete genus *Taphrina*. *International Journal of Systematic and Evolutionary Microbiology* 53: 607–616.

Roger, A. J., and L. A. Hug. (2006). The origin and diversification of eukaryotes: Problems with molecular phylogenetics and molecular clock estimation. *Philosophical Transactions of the Royal Society, B* 361: 1039–1054.

Ruiz-Trillo, I., G. Burger, P. W. H. Holland, N. King, B. F. Lang, A. J. Roger, and M. Gray. (2007). The origins of multicellularity: A multi-taxon genome initiative. *Trends in Genetics* 23: 13–118.

Runnegar, B. (1991). Precambrian oxygen levels estimated from the biochemistry and physiology of early eukaryotes. *Paleogeography, Palaeoclimatology, Palaeoecology* 97: 97–111.

Satterthwait, D. F. (1976). Paleobiology and Paleoecology of Middle Cambrian Algae from Western North America. PhD thesis, University of California, Los Angeles.

Saul, J. M., and L. Schwartz. (2007). Cancer as a consequence of the rising level of oxygen in the Late Precambrian. *Lethaia* 40: 211–220.

Saunders, G. W., and M. H. Hommersand. (2004). Assessing red algal supraordinal diversity and taxonomy un the context of contemporary systematic data. *American Journal of Botany* 91: 1494–1507.

Schlichting, C. D. (2003). Origins of differentiation via phenotypic plasticity. *Evolution & Development* 5: 98–105.

Schoch, C. L. et al. (2009). The Ascomycota tree of life: A phylum-wide phylogeny clarifies the origin and evolution of fundamental reproductive and ecological traits. *Systematic Biology* 59: 224–239.

Schneider, H., E. Schuettpelz, K. M. Pryer, R. Cranfill, S. Magallon, and R. Lupia. (2004). Ferns diversified in the shadow of angiosperms. *Nature* 428: 553–557.

Scott, M. C., T. W. Lyons, A. Bekker, Y. Shen, S. W. Poulton, X. Chu, and A. D. Anbar. (2008). Tracing the stepwise oxygenation of the Proterozoic ocean. *Nature* 452: 456–458.

Sebé-Pedros, A., A. J. Roger, F. B. Lang et al. (2010). Ancient origin of the integrin-mediated adhesion and signaling machinery. *Proceedings of the National Academy of Sciences USA* 107: 10142–10147.

Segawa, Y., H. Suga, N. Iwabe, C. Oneyama, T. Akagi, and M. Okada. (2006). Functional development of Src tyrosine kinases during evolution from a unicellular ancestor to multicellular animals. *Proceedings of the National Academy of Sciences USA* 103: 12021–12026.

Shen, Y., A. H. Knoll, and M. R. Walter. (2003). Evidence for low sulphate and deep water anoxia in a mid-Proterozoic marine basin. *Nature* 423: 632–635.

Sherwood-Pike, M. A., and J. Gray. (1985). Silurian fungal remains—probable records of the Class Ascomycetes. *Lethaia* 18: 1–20.

Short, M. B., C. A. Solari, S. Ganguly, T. R. Powers, J. O. Kessler, and R. E. Goldstein. (2006). Flows driven by flagella of multicellular organisms enhance long-range molecular transport. *Proceedings of the National Academy of Sciences USA* 103: 8315–8319.

Sieberer, T., and O. Leyser. (2006). Auxin transport, but in which direction? *Science* 312: 858–860.

Steiner, M. (1994). Die neoproterozoischen Megaalgen Südchinas. *Berliner geowissenschaftliche Abhandlungen (E)* 15: 1–146.

Taylor, T. N., H. Hass, and H. Kerp. (1999). The oldest fossil ascomycetes. *Nature* 399: 648.

Thomson, J. M., E. A. Gaucher, M. F. Burgan, D. W. De Kee, T. Li, J. P. Aris, and S. A. Benner. (2005). Resurrecting ancestral alcohol dehydrogenases from yeast. *Nature Genetics* 37: 630–635.

Turmel, M., J. F. Pombert, P. Charlebois, C. Otis, and C. Lemieux. (2007). The green algal ancestry of land plants as revealed by the chloroplast genome. *International Journal of Plant Sciences* 168: 679–689.

Valentine, D. L. (2007). Adaptations to energy stress dictate the ecology and evolution of the Archaea. *Nature Reviews of Microbiol*ogy 5: 316–323.

Vardi, A., I. Berman-Frank, T. Rozenberg, O. Hadas, A. Kaplan, and A. Levine. (1999). Progammed cell death of the dinoflagellate *Peridinium gatunense* is mediated by CO_2 limitation and oxidative stress. *Current Biology* 9: 1061–1064.

Waldbauer, J. R., L. S. Sherman, D. Y. Sumner, and R. E. Summons. (2009). Late Archean molecular fossils from the Transvaal Supergroup record the antiquity of microbial diversity and aerobiosis. *Precambrian Research* 169: 28–47.

Wellman, C. H., and J. Gray. (2000). The microfossil record of early land plants. *Philosophical Transactions of the Royal Society, B* 355: 717–731.

Wilson, E. O. (1999). *The Diversity of Life*. Cambridge, MA: Harvard University Press.

Xiao, S., and A. H. Knoll. (2000). Phosphatized animal embryos from the Neoproterozoic Doushantuo Formation at Weng'an, Guizhou Province, South China. *Journal of Paleontology* 74: 767–788.

Xiao, S., A. H. Knoll, X. Yuan, and C. M. Pueschel. (2004). Phosphatized multicellular algae in the Neoproterozoic Doushantuo Formation, China, and the early evolution of florideophyte red algae. *American Journal of Botany* 91: 214–227.

Xiao, S., X. Yuan, M. Steiner, and A. H. Knoll. (2002). Macroscopic carbonaceous compressions in a terminal Proterozoic shale: A systematic reassessment of the Miaohe biota, South China. *Journal of Paleontology* 76: 345–374.

Yin, L., M. Zhu, A. H. Knoll, X. Yuan, J. Zhang, and J. Hu. (2007). Doushantuo embryos preserved within diapause egg cysts. *Nature* 446: 661–663.

13 The Small Picture Approach to the Big Picture: Using DNA Sequences to Investigate the Diversification of Animal Body Plans

Lindell Bromham

The Adaptive Radiation of the Metazoans

The Metazoa (animal kingdom) is divided into approximately three dozen phyla (figure 13.1). The first undisputed fossils of around half of the animal phyla appear in the Cambrian, the geological period that runs from around 543 million years ago (Myr) to 488 Myr. At least a third of animal phyla have no fossil record to speak of (Valentine 2004), but we can infer from phylogenetic relationships that many of these lineages must be at least Cambrian in age. On the basis of this fossil evidence, it has been suggested that all of the major kinds of animals were generated in a period of around 10 to 15 million years (e.g., Carroll 2005; Levinton 2001; Valentine 2004). This inferred explosive radiation of animals in the Cambrian has been considered the signature of a phenomenal rise in diversity and complexity of animal life, and creating more complex ecosystems (e.g., Bambach, Bush, and Erwin 2007).

An earlier Precambrian fauna, known as the ediacarans, were relatively simple, soft-bodied creatures (Xiao and Laflamme 2008). With the possible exception of *Kimberella*, which has been interpreted as having a muscular foot and scraping radula like a mollusc (Fedonkin and Waggoner 1997), none of the ediacarans show clear evidence of appendages specialized for locomotion, and there are relatively few complex trace fossils (marks made in the sediment) that would bear witness to directed bilaterian movement in the Precambrian period (Jensen, Droser, and Gehling 2005). Due to the general lack of evidence of mouths, claws, teeth, eyes, or other equipment associated with hunting or foraging, the ecology of ediacaran communities has also been regarded as being fairly simple (Bambach et al. 2007; Xiao and Laflamme 2008). By contrast, the Cambrian fauna provides abundant evidence of animals with sense organs, appendages for locomotion and feeding, and defensive structures. Unlike the floor-bound ediacarans, some Cambrian animals moved down into the sediment by active burrowing, and some moved up into the water column by directed swimming. For the first time, there is clear evidence of specialized, mobile animal predators.

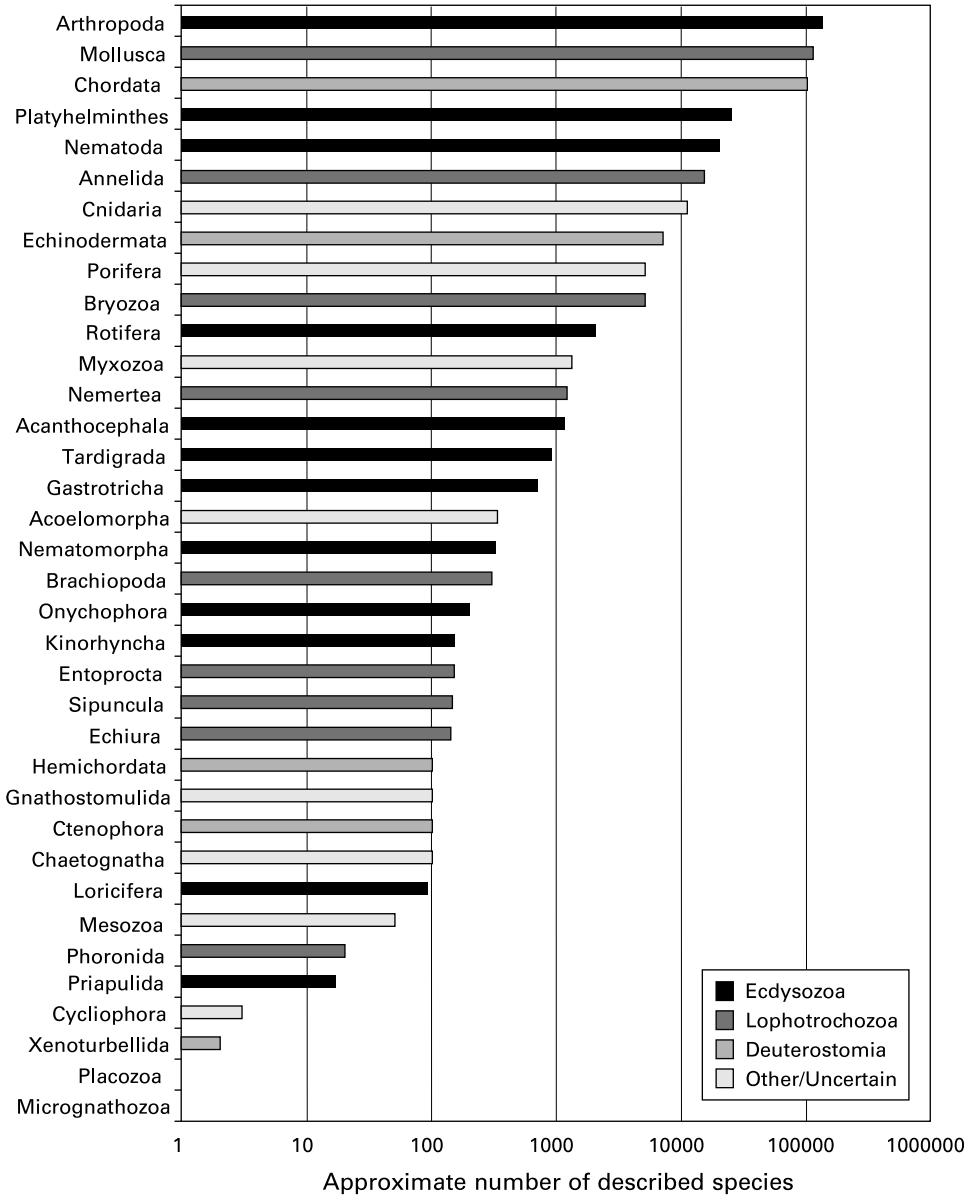


Figure 13.1
Approximate number of described species per metazoan phylum. There is no central database for described animal species, so these figures are open to debate. In most cases, it is a fair assumption that the number of described species will underestimate the actual diversity of the phylum, and in some cases only a relatively small proportion of the species thought to exist have been described. In addition, there is no definitive list of animal phyla, because there is disagreement over which taxa should be given phyletic status. Some taxa listed here are contained within other phyla in some systematic treatments (e.g., Echiura within Annelida). Some single species or genera have been elevated to phylum level on the basis of morphology (Micrognathozoa) or phylogeny (Xenoturbellida). Here, phyla are labeled according to superphyletic groupings (see figure 13.2). Assignment to superphyla is controversial in some cases. For example, Myxozoa (an important group of parasites of fish and other animals) have variously been classified as protists, cnidarians, or as a separate bilaterian phylum.

The metazoan radiation itself is not surprising. The great diversity of animals today must have been ultimately derived from a simple common ancestor, so we know the metazoans made the transition from blobs to bugs at some point. Adaptive radiations are common throughout evolutionary history, where a single ancestral lineage diversifies rapidly to produce a wide range of ecologically specialized descendents. We see evidence of rapid adaptive radiations on many oceanic islands, where a colonizing lineage radiates to fill a variety of ecological niches. For example, in as little as 10 million years, a single ancestral lineage of Hawaiian honeycreepers has radiated into more than fifty separate species, with a wide variety of colors and shapes, which occupy a range of niches including insectivores, seed-eaters, frugivores, nectarivores, and snail-eaters (Grant 2001; Lovette, Bermingham, and Ricklefs 2002). What is remarkable about the Cambrian radiation of metazoans is its apparent suddenness and uniqueness: It seems that more fundamental evolutionary change in animal complexity and diversity occurred in this relatively short period than in any equivalent time period before or since the Cambrian. In the time that it took Hawaiian honeycreepers to change the shape of their beaks or the color of their plumage, whole new body plans appeared in the fossil record.

Many hypotheses have been put forward to explain the sudden burst of animal diversity and disparity in the Cambrian. Some suggest an environmental trigger for the diversification: For example, animal evolution may have been constrained in earlier periods by a lack of environmental oxygen, so the rise in oxygen could have simultaneously released all metazoan lineages to develop large size and complex morphology (see Knoll, this volume). Others suggest that the driver of change was a kind of arms race in morphological or ecological complexity: For example, as some lineages became mobile predators, others had to develop defensive structures (see Bengston 2002). In this chapter, I want to consider only one particular kind of explanation for the Cambrian radiation, based on an "internal" trigger for the explosive evolution of diversity and disparity: that major innovations in body plan were generated from relatively few genetic changes of large phenotypic effect, particularly in the function of key conserved developmental genes such as those in the Hox cluster.

It is important to note that the various hypotheses for the cause of the metazoan radiation are not mutually exclusive; they may have all operated in concert to generate an extraordinary period of evolutionary change. However, here I concentrate only on a critical examination of the developmental genetic hypothesis of the origin and maintenance of body plans, which has garnered enthusiastic support in the last decade or two, because it has important implications for understanding macroevolutionary patterns.

Microevolution vs. Macroevolution: How Do Differences in Body Plan Arise?

The debate over the origin and evolution of animal body plans has a key role to play in evaluating claims about macroevolution. Broadly speaking, "microevolution" is used to describe the change in representation of heritable variants in a population over generations,

where new variants enter the population by mutation or migration, then rise or fall in frequency by selection and drift until one variant replaces all others. "Macroevolution" represents those changes that are not observable at the population level, but are detected by comparing different evolutionary lineages: Examples include the origins of major evolutionary adaptations, and differences in diversification rate. Darwin's genius was to connect the two: He explained macroevolution (differences between lineages, both past and present) using microevolutionary mechanisms (the observable change in frequency of variants in contemporary populations). He did this by demonstrating that variation is ubiquitous in populations; that there was a continuum of differences between populations, races, varieties, and species; and, most famously, by providing a plausible explanation of how the variation in populations could lead to the differences between lineages by the gradual accumulation of small changes over very long periods of time.

Darwin's argument rested on two related principles: uniformitarianism and gradualism. Darwin adapted Sir Charles Lyell's uniformitarian approach to geology, in which "the present is the key to the past" (Lyell 1830). The massive changes of the past, like building mountains or changing courses of rivers, could be explained by the continuous action over long time periods of forces we can witness today, such as uplift and erosion. This was Darwin's strategy for linking microevolution to macroevolution: The changes we can observe in populations today are sufficient, given immense time periods, to generate different lineages (Darwin 1859). But unlike geology, where occasional catastrophes can create sudden large changes, species-level differences do not tend to arise in contemporary populations. So to make the uniformitarian argument plausible, Darwin had to rely strongly on an argument from gradualism, such that large-scale changes are achieved by the accumulation of many small differences over long time periods:

> As natural selection acts solely by accumulating slight, successive, favourable variations, it can produce no great or sudden modifications; it can act only by very short and slow steps. Hence the canon of 'Natura non facit saltum' . . . We can plainly see why nature is prodigal in variety, though niggard in innovation. (Darwin 1859, 489)

Darwin's insistence that macroevolution could be explained in terms of microevolution was his most controversial claim. Even die-hard supporters of Darwin, such as Thomas Henry Huxley and Alfred Russel Wallace, did not fully support this claim, suggesting that there may be some evolutionary changes that did not fit this framework. Nonetheless, this key aspect of Darwin's theory of evolution became the foundation of the neo-Darwinian synthesis, which bolstered Darwin's theory by basing it on population genetics (both theory and observation), strengthened by observations of natural selection in wild and experimental populations, and evidence of gradual change from paleontology. Thus, mainstream opinion in evolutionary biology has been that lineage differences can be explained in terms of population genetic processes: All evolution is microevolution, and macroevolution is a level of observation, rather than a separate process.

But there have always been challenges to this view. Two major arguments have been made against the "macroevolution equals microevolution plus time" hypothesis. First, the principle of gradualism has been challenged on the basis that the fossil record suggests, in some cases, discontinuities in the origin of lineage differences, rather than a continuous accumulation of small changes. Second, the principle of uniformitarianism has been challenged on the grounds that large changes to phenotype, rarely if ever witnessed in contemporary populations, may occasionally generate lineage-level changes, particularly in periods when lineages are somehow more responsive to such large changes. Both of these challenges are most evident in debates about the Cambrian explosion, where some researchers have explained the disjunction in forms in the animal fossil record in terms of large changes to phenotype generated through changes to developmental gene expression, which no longer arise in modern populations due to developmental and genetic canalization.

Recent advances in the understanding of the way changes in the genome are translated to different phenotypes through the process of development have fueled this challenge to neo-Darwinian gradualism. Studies of genes that play a fundamental role in early embryonic patterning have revealed surprising and exciting results. In particular, a common "toolkit" of developmental genes has been found in a wide range of metazoans. Sometimes these genes perform similar functions in very different organisms, such as the *Pax6* gene, which initiates eye formation in species as divergent as flies, humans, and flatworms. In other instances, the same genes perform different tasks, or are expressed in different places or times (see Garcia-Fernandez 2005). In some cases, changes in the expression patterns of these genes correspond with key differences in body plan, such as segment identity in arthropods with different numbers and types of appendages (Tour and McGinnis 2005). The role played by these genes in determining the development of body plan in animal embryos has led to suggestions that these genes were instrumental in the evolution of different body plans. This claim has given rise to the hypothesis that the evolution of the "toolkit" itself, or the changing patterns of usage of toolkit genes, triggered the Cambrian explosion (e.g., Garcia-Fernandez 2005; Gellon and McGinnis 1998). In particular, it has been suggested that changes to developmental genes offer a way of generating very different phenotypes from relatively few changes to genotype.

The role of developmental genes in the evolution of animal body plans has been interpreted in a number of ways that are relevant to the issue of whether macroevolution is wholly explained by microevolutionary mechanisms. In some discussions, there is an implicit or explicit assumption that evolutionary changes in development follow the same microevolutionary patterns as any other trait (e.g., Budd 1999; Carroll 2005). But in other cases, the claim is made that consideration of the influence of key developmental genes suggests that the evolution of major body plan changes occurred by a discrete macroevolutionary process, not by the microevolutionary processes that we can witness in action today (e.g. Arthur 2000; Baguna and Garcia-Fernandez 2003; Budd 2006; Carroll 2000). If it is

true that the Cambrian explosion is an example of a discrete evolutionary event that marks a jump to a new level of complexity, then processes we observe in contemporary organisms and ecosystems may not give a full account of macroevolution (Erwin 2004). For example, Butterfield (2007) interprets the Cambrian explosion as a sign that macroevolution before the Cambrian was of a fundamentally different type than that which occurred after the Cambrian, and therefore concludes that a uniformitarian approach to studying macroevolution is not appropriate. More generally, the emerging view of the link between developmental genes and body plan evolution has led to the idea that the neo-Darwinian synthesis has been critically incomplete without information on development or body plan change (e.g., Carroll 2000; Pigliucci 2007; Telford and Budd 2003). These claims are important because if they are true, we have been doing things wrong for quite a while. It is therefore important to test the hypothesis that past metazoan evolution was of a different type from currently observable microevolutionary processes, by considering the available evidence.

One way to explore the idea that the Cambrian explosion was caused by large changes to body plan, particularly through the action of developmental genes, is to ask why the Cambrian explosion is unique in animal evolution. The fossil record does not record any other periods of such radical change in form and complexity of so many animal lineages simultaneously. Why do body plan differences all seem to originate in this particular evolutionary period, and why have none arisen since? If major innovations in body plan can be generated from few genetic changes in key genes, then why do these changes not occur throughout animal history, generating new phylum-level differences after the Cambrian explosion? Should we not see such variants arising in contemporary populations? One common explanation is that animal evolution in the Cambrian period was in some way permissive, and that body plan variants either could not be generated (due to genetic canalization) or could not persist (due to competition) at later stages (e.g., Davidson and Erwin 2006; Erwin 2007; Levinton 2001). If this was true, then we should expect that once body plans were formed, they were unable to give rise to new body plans. This process may be analogous to the developmental canalization of stem cells that, once committed to becoming a specialized cell line like muscle, heart tissue, or bone marrow, can no longer return to the pluripotent state nor give rise to a fundamentally different kind of cell.

If we wish to know whether animal evolution in the Cambrian was by a special mechanism that was not able to operate in later ages, then it would be helpful to know how body plan variation is generated, and whether the capacity for body plan change has been limited or absent since the Cambrian. There are several ways of approaching this issue, and we could not hope to form a complete picture of body plan evolution without information from paleontology, population genetics, development, physiology, and so on. But here I wish to focus on just one line of evidence that has proved valuable in unraveling metazoan evolution: molecular phylogenetics (using DNA or protein sequences to uncover the evolutionary relationships between contemporary species).

Much of the focus on the use of molecular phylogenies to understand the Cambrian explosion has been on molecular dating. Most estimates of divergence dates made from molecular data point to a substantial Precambrian history of the major metazoan lineages. These molecular dates are increasingly being interpreted as being compatible with the fossil record of ediacarans and Precambrian metazoan embryos (e.g., Budd 2008; Peterson et al. 2008). However, the large degree of variation between published estimates have made molecular dates difficult to interpret (Bromham 2006): Molecular dates have been presented as being both compatible (e.g., Aris-Brosou and Yang 2003) and incompatible (e.g., Blair and Hedges 2005) with an explosive radiation of animal phyla in the early Cambrian. Any interpretation of molecular date estimates must be done with a weather eye to the potential for imprecision and error in molecular estimates, and the results must be considered in light of evidence from all other lines of inquiry (Bromham 2006; Wray 2001).

Less controversially, molecular phylogenies provide an alternative record of the relationships between metazoan lineages, independent of traditional systematics based on morphological and developmental characters. These molecular phylogenies have an important role to play in testing ideas about body plan evolution. Molecular phylogenies have dramatically reshaped ideas about animal evolution. In particular, molecular phylogenetic analyses have split the animal kingdom into four main groups (figure 13.2). The first group, the diploblasts, contains the oldest extant phyla of the animal kingdom, Porifera (sponges) and Cnidaria (jellyfish, corals, etc). The Cambrian explosion is generally considered to represent the earliest diversification of the remaining phyla, collectively referred to as the Bilateria. Molecular phylogenies have been used to group the bilaterian phyla into three superphyla: the Lophotrochozoa (annelids and molluscs and their kin), Ecdysozoa (arthropods, nematodes, and relatives) and Deuterostomia (echinoderms, chordates, and friends). Each of these superphyla contains a diversity of forms and ways of life.

In addition to revealing novel superphyletic groupings in the animal tree, molecular data has also revolutionized the systematics of the "minor phyla": types of animals that are recognized as being representatives of ancient lineages, yet whose modern members are typically neither diverse nor disparate. Many of these minor phyla consist of, to be blunt, fairly unexciting little marine worms. In fact, a good proportion of metazoan phyla consist of dull little worms of one kind or another, even if explanations of the Cambrian Explosion tend to focus on the more charismatic body plans. Molecular phylogenetic analyses have been crucial in reconstructing the origins of the minor phyla and their relationships to the sexier phyla. To quote Levinton (2001, 465):

> Until the advent of molecular sequencing techniques, it was often difficult to establish relationships among apparently distantly related groups; this falsely highlights the multiple weirdo-evolutionary lawn hypothesis. No set of groups has been more victimized by this shortcoming than the "wormy" groups.

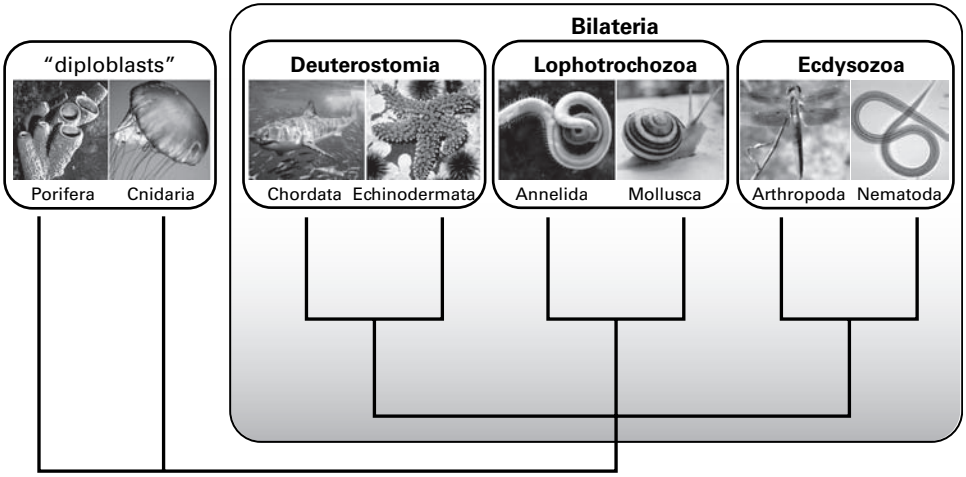


Figure 13.2
The relationships between some of the most species-rich metazoan phyla. Molecular phylogenies have caused the animal kingdom (Metazoa) to be divided into four superphyletic groups. The relationships between the basal groups, including Porifera (sponges) and Cnidaria (including jellyfish and corals), is uncertain (for example, there has been some suggestion that Porifera is paraphyletic and Cnidaria is descended from the bilaterian lineage). The bilaterian phyla (not all of which are bilaterally symmetrical) are divided into three superphyla. The Deuterostomia includes the chordates, echinoderms, and a number of minor phyla such as Hemichordata. The Lophotrochozoa are named for the lophophore (a tubelike feeding appendage found in some of the phyla, such as bryozoans, brachiopods, and phoronidans) and the trochophore type of larvae. Lophotrochzoan phyla, most of which show evidence of spiral cleavage in early embryological cell divisions, include annelids, molluscs, sipunculids (peanut worms), echiurans (spoon worms), pogonophorans (tube worms), and nemerteans (ribbon worms). The Ecdyzosozoa are named for the habit of molting their outer cuticle (a process referred to as ecdysis). The major ecdysozoan phyla are the arthropods and nematodes, but an array of other phyla are also part of this group, such as tardigrades, onycophorans, and rotifers.

The Evolutionary Lability of Body Plans

Phylogenies that reveal the evolutionary relationships between major animal groups are essential for placing both body plans and developmental genes in an evolutionary framework, to allow prediction of ancestral states, reconstruction of patterns of character evolution, and establishment of homology of traits (e.g., Baguna and Garcia-Fernandez 2003; Fortey, Briggs, and Wills 1996; Jenner 2000; Telford and Budd 2003). Molecular phylogenies play a particularly useful role because they provide a means of avoiding the circularity of inferring phylogeny from body plan characters, then using those same phylogenies to infer patterns of body plan evolution (see Jenner 2003). However, molecular phylogenies are not without error, as any comparison of published phylogenies of metazoan phyla will attest. Therefore, the use of molecular phylogenies in testing ideas about body plan evolution must be done within a statistical framework that assesses the robustness of the conclusions (e.g., Bromham and Degnan 1999). Inclusion of minor phyla is essential to

completing our picture of metazoan evolution, as these minor phyla often provide the intermediates that make the "unbridgeable gaps" between the major phyla look less severe, and help to reconstruct the order of acquisition of key body plan characteristics.

In order to illustrate how relatively modest systematic or comparative case studies can shed light on the big picture of animal evolution, I will present some cases studies of body plan evolution. I will focus specifically on the question of the fixity of animal body plans, rather than their origins, since this seems the most tractable part of the mystery to explore. The case studies presented here are not necessarily the most fascinating or conclusive case studies, they just happen to be the ones I have a passing familiarity with because members of my research group have worked on these taxa. No doubt someone else would choose a different set of taxa, and different body plan characteristics. As it happens, we will consider some of the less lovable metazoans: peanut worms, acorn worms, and cockroaches.

Lessons from Acorn Worms: Body Plans Are Not Immutable

Deuterostomia, one of the three bilaterian superphyla, contains the chordates, echinoderms, and a number of minor phyla. The members of the deuterostome superphylum are united by key developmental features, although the adult body plans of the two major deuterostome lineages could hardly be more different. The chordates, including our good selves, have a head-and-tail body plan, with the brain, sense organs, mouth, and breathing apparatus located up one end of the body, a muscularized post-anal tail at the other end, with a hollow dorsal nerve cord running down the back. They get their name from the notochord, a stiffened internal rod that runs from the head to the tail (this is replaced during development by the backbone in vertebrates). Echinoderms, on the other hand, have no head and no tail. Instead, they develop by pentaradial growth, giving rise to the iconic five-pointed symmetry of many echinoderms, such as starfish. Echinoderms do not have a linear nerve cord or a brain, but a ring-shaped nervous system (thus, reminiscent of a zombie movie, some echinoderms are efficient mobile predators with no brains). Echinoderms also have a water-vascular system that is used for both circulation and locomotion.

What would the ancestor of two such wildly different body plans have looked like? It is commonly assumed that the deuterostome ancestor possessed the basic bilaterian characteristics such as a front and back end, but none of the specific body plan characteristics of the modern deuterostome phyla, like a dorsal nerve cord or a water vascular system. Under this scenario, only after the split of the deuterostome stem lineage did the two major branches develop the characteristic body plans we see today: The chordates retained bilateral symmetry and developed a pharynx and dorsal nerve cord, the echinoderms lost their heads and developed pentameral symmetry, radial nervous system and water vascular system. But molecular studies of the minor phyla in the deuterstomes have challenged this picture.

The echinoderms and chordates account for around 95 percent of all deuterostome species. But there are a number of less charismatic lineages in the superphylum, including the

cephalochordates (lancelets), urochordates (sea-squirts), and hemichordates (acorn worms and pterobranchs). As their names suggest, it has generally been considered that these three groups arose from the chordate lineage, based on the shared chordate-like features of the dorsal nerve cord and pharynx (basically a head with gill slits). Whether the urochordates and cephalochordates should be considered phyla in their own right or subphyla of the Chordata is a matter of debate (Cameron, Garey, and Swalla 2000). This debate over phyletic status serves as a healthy reminder that while phyla are typically considered to represent discrete body plans separated by unbridgeable gaps, ongoing debate about whether certain lineages represent phyla or not illustrates that, in at least some cases, the distinction between "body plans" is less obvious. An extreme example is the deuterostome genus, *Xenoturbella*, consisting of two species of marine worms with virtually no distinguishing features (no brain, no central nervous system, no through-gut, no excretory system, not even any gonads), which was elevated to phylum status by phylogenetic studies that suggested it was a basal lineage within the deuterostome superphylum (Bourlat et al. 2006). The debate over phyletic status of deuterostome lineages illustrates that phyla (and thus "body plans") are not always discrete and obvious, but can grade into each other, an observation that could be interpreted as support for a gradualist model of body plan evolution in these lineages.

Hemichordates have traditionally been regarded as an early-branching lineage of chordates on the basis of shared chordate-like features, particularly the dorsal nerve cord and pharynx. But molecular sequence data consistently places hemichordates on the echinoderm lineage of the deuterostomes (figure 13.3). This conclusion has been supported by virtually all molecular phylogenetic analyses, including a large-scale study of 170 nuclear genes (Bourlat et al. 2006) and analysis of whole mitochondrial genomes (Castresana et al. 1998). So DNA sequence data provide statistically significant support for the hemichordates and echinoderms sharing a more recent common ancestor than either does with the chordates, a conclusion supported by some gene expression data (see Bromham and Degnan 1999).

The reader could be forgiven for thinking this is a bewilderingly dull example to include in a book on the rather more exciting topic of major transitions. Who really cares where acorn worms fit in the big scheme of things? But determining the phylogenetic position of hemichordates can tell us a lot about the evolution of the extremely different body plans in the major deuterostome lineages, Chordata and Echinodermata. If both of the major branches of the deuterostomes clade contain phyla with the classic chordate body plan features of a pharynx and dorsal nerve cord, then it implies that these features were present in the common ancestor (or gained independently in two lineages, which seems less likely given the shared developmental patterns; see Bromham and Degnan 1999; Hinman and Degnan 2000). This means that the ancestor of all deuterstome phyla had a chordatelike body plan (figure 13.3). The corollary of this is that the echinoderm lineage began with one body plan, lost those body plan features, and gained an entirely new set. Whenever this

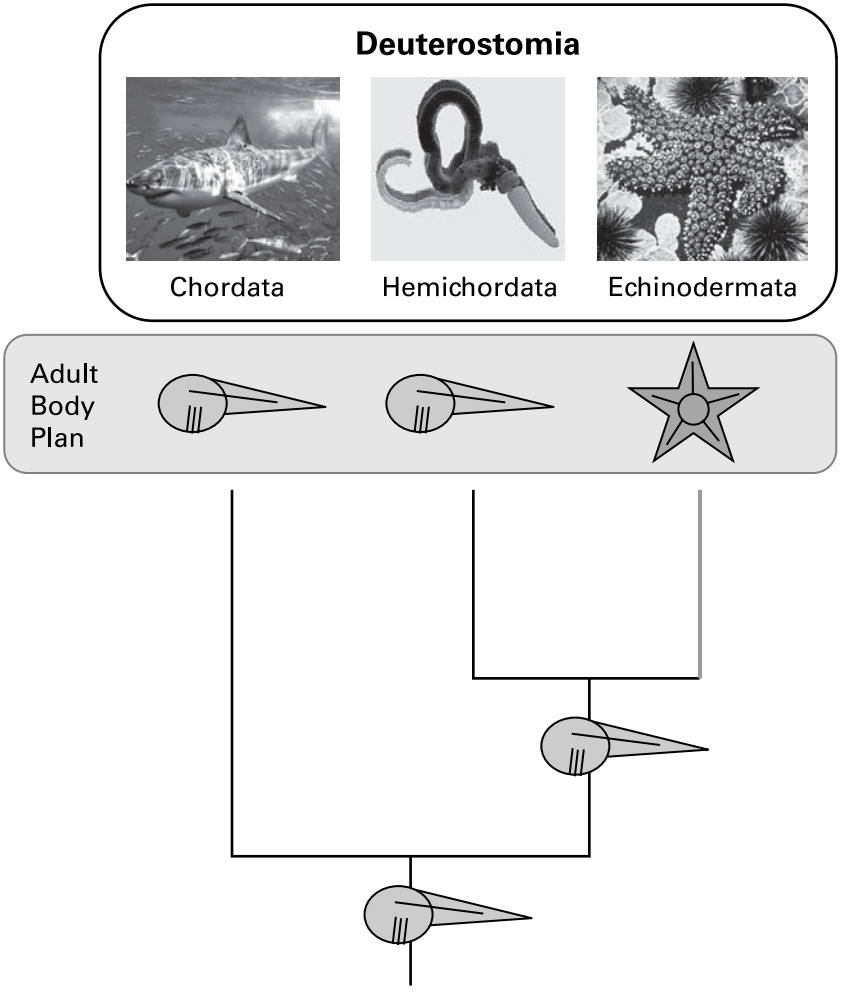


Figure 13.3
Chordates and hemichordates share key elements of their adult body plans, most notably the pharynx (throat with gill slits) and a dorsal nerve cord. So when taxa are grouped on the basis of shared body plan characters, the Chordata and Hemichordata have been considered to be more closely related to each other than either is to the pentamerally symmetrical echinoderms, which have no pharynx or dorsal nerve cord. However, molecular data unambiguously supports a grouping of hemichordates and echinoderms, suggesting that the chordate body plan features might be ancestral for this superphylum.

transition happened, it runs counter to the prevailing notion that, once a body plan has evolved, it would not have the evolutionary flexibility to give rise to an entirely different body plan (e.g., Davidson and Erwin 2006; Levinton 2001). Yet echinoderms seem to have done it in spades. While this is only one example, it does suggest that we cannot make the general claim that body plan characteristics were formed in a discrete event, or by some discontinuous mechanism, then were resistant to further change.

Lessons from Cockroaches: The Developmental Basis of Body Plan Is Labile

The evolution of segmentation provides a useful illustration of how molecular phylogenies can shed new light on the evolution of animal body plans. Segmentation refers to the repetition of structural units, either in the formation of body parts in the embryo or in features of the adult body plan; thus, it is one of the most fundamental aspects of body plan. One of the most obviously segmented phyla is Annelida (earthworms and their kin), where the tubelike body consists of repeated "rings." Chordates (the phylum containing the vertebrates) are less obviously segmented, but skeleton and muscles form in the embryo by the development of segmented blocks of tissues called somites. Segmentation in arthropods has been cited as one of the reasons for their evolutionary success, by providing a flexible way of patterning bodies that leads to diversification of limb morphology. But while these "big three" are often considered the only truly segmented (eusegmented) phyla, there is no clear definition of what should be called segmentation and what shouldn't. Chitons (basal molluscs), for example, have rows of plates down their backs, which some have interpreted as segments. Furthermore, even in the eusegmented taxa, some parts of the body are segmented, and some parts aren't: For example, in chordates, skeleton, muscles and skin form from a series of segments (somites), but other body parts, such as the internal organs, do not.

Some earlier animal taxonomies grouped segmented phyla together on the grounds that sharing such a fundamental body plan character must reveal a shared ancestry. However, molecular phylogenies do not group the three "eusegmented" phyla together. Instead, the eusegmented phyla (chordates, arthropods, and annelids) occur in each of the major lineages of animals (deuterostomes, ecdysozoans, and lophotrochozoans; see figure 13.4). This observation has led to the suggestion that segmentation was a feature of the last common ancestor of all bilaterian phyla, because it is assumed that the presence of this fundamental body plan character in all three major superphyla must reveal common inheritance of an ancestral segmented body plan. Thus segmentation is often cited as one of the features of the urbilaterian, the ancestral bilaterian possessed of all the genetic and developmental equipment necessary to give rise to the radiation of animal phyla (e.g., Balavoine and Adoutte 2003).

But there are fundamental differences in the way that members of these eusegmented lineages build their embryos (see Tautz 2004). A chick (chordate) embryo generates waves

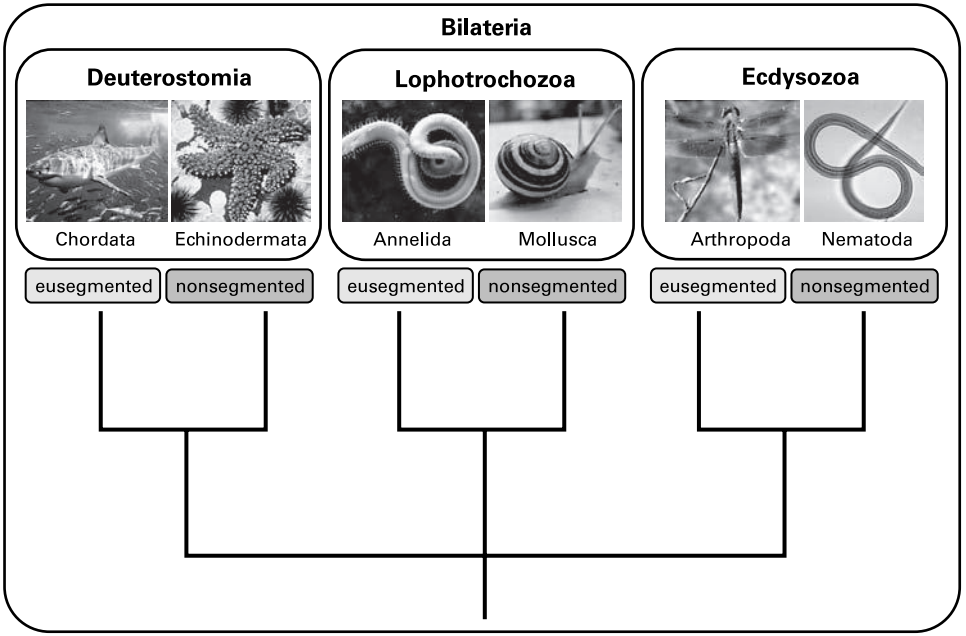


Figure 13.4
Three phyla are widely considered to be "eusegmented," though there are other phyla that show some degree of segmental construction. These eusegmented phyla occur in each of the bilaterian superphyla.

of gene expression, and as each new wave moves along the embryo, it creates an additional segment boundary. In a leech (annelid) embryo, new segments are defined by rounds of cell division. The fruitfly (arthropod) embryo uses an "inelegant" and complex interaction between the expression patterns of gap, pair-rule, and segment polarity genes to divide the embryo into stripes that develop specific segment identities (Akam 1989; Peel, Chipman, and Akam 2005). So at first glance it seems that each of the major bilaterian lineages has a unique way of creating a segmented body.

But filling in the gaps between these developmental exemplar species blurs the boundaries between the ways of forming the basic ground plan of the body. It transpires that the mechanism of segmentation in the fruitfly, the darling of arthropod genetics, is not typical of other arthropods. For example, the spider *Cupiennius salei* employs a mode of segmentation that is more similar to that of vertebrates than flies, in which waves of expression create sequential segments (Stollewerk, Schoppmeier, and Damen 2003). The spider even uses some of the same genetic pathways (e.g., the *Notch-Delta* pathway) to drive this "clock and wave-front" method of segmentation. Studying the pattern of segmentation in other arthropod lineages might shed light on the evolutionary process that changed the underlying mechanism of segmentation in fruitflies (Peel et al. 2005).

So why study cockroaches? The phylogenetic position of the Blattodea (cockroaches) makes them informative for studying the evolution of segmentation, since they sit between the wave-front spiders and the pair-rule flies. Blattodea contains at least 4,500 species, and, despite their bad reputation, many of them are quite charming creatures. I once kept a very attractive little native Australian cockroach as a pet, a teardrop-shaped animal less than a centimeter long, with a segmented carapace, delicate yellow stripes, and cute little cerci (the sensory appendages that stick out of the back end and detect air movements, a distinctive feature of blattodeans). I asked an entomologist what these native cockroaches ate, and he said no one knew, so I fed it lasagne. So not all cockroaches are shudder-inducing nuclear-holocaust-surviving global pests.

It turns out that American cockroaches (*Periplaneta americana*), like spiders and millipedes, use the *Notch* pathway to produce segments (Pueyo, Lanfear, and Couso 2008). This suggests that the wavefront method of segmentation is ancestral to the arthropods and insects, but that some insect lineages, including the flour-beetle *Tribolium castanateum* and the fruitfly *Drosophila melanogaster,* then evolved a new developmental pathway to make segmented embryos (figure 13.5). In other words, these lineages have rewired the developmental pathways used to make their segmented bodies, even though the end product looks much the same as in other insects. In fact, different parts of arthropod bodies are segmented by different means: For example, the *Notch* pathway has been coopted into specifying the development of leg segmentation in flies (see Pueyo et al. 2008).

This lability of the genetic architecture underlying segmentation tells us that even this fundamental aspect of body plan has been able to change since the Cambrian explosion. Flies and beetles are post-Cambrian creatures, descendents of the metazoan colonization of the land and sky. The first fossil insects are Devonian (approximately 420 to 360 Myr), winged insects do not appear until the Carboniferous (approximately 360 to 300 Myr), and fossil flies don't appear until the Triassic (approximately 250 to 200 Myr). If the basic developmental processes underlying body plan formation were able to change so dramatically in these lineages long after the Cambrian explosion, it would seem that the genetic architecture underlying body plans was not set during the Cambrian and has been immutable since.

Lessons from Peanut Worms: Body Plan Characters Come and Go

The examples given in the previous sections show that, on the one hand, there is evidence that members of two superphyla, Ecdysozoa and Deuterostomia, share aspects of the developmental basis of body plan formation because of the common use of the *Notch* pathway in determining segmentation in chordates and some arthropods. But, on the other hand, some arthropods have ditched this fundamental aspect of developmental genetics and use an entirely different way of making segments. What can we learn from looking at the third superphylum, the Lophotrochozoa, which contains the eusegmented annelids?

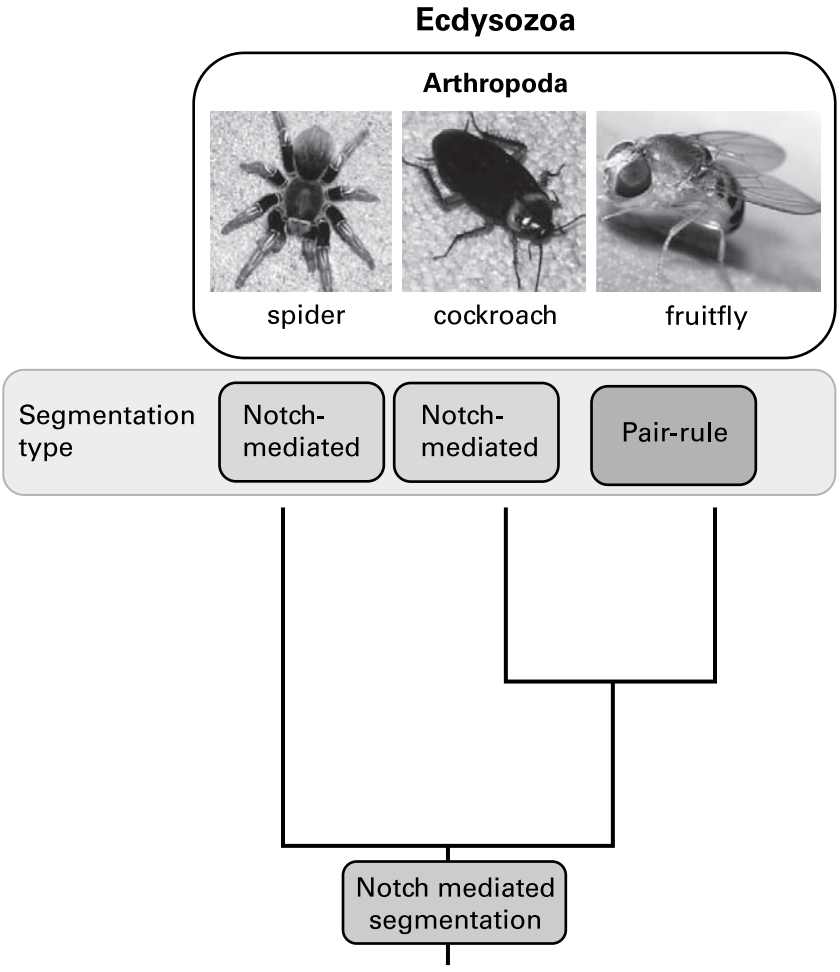


Figure 13.5
Spiders (*Cupiennius salei*) and cockroaches (*Periplaneta americana*) both employ the Notch-signaling pathway to define segments in the growing embryo. This pathway is also used in segmentation in vertebrate embryos, suggesting that either Notch-mediated segmentation is ancestral for two of the bilaterian superphyla, or that the Notch pathway has been independently coopted into defining segments in more than one lineage. Flies and beetles have evolved an entirely different means of defining body segments, independent of the Notch pathway.

Like the deuterostomes, the lophotrochozoans are united by shared developmental traits despite the fact that the major lineages—Mollusca and Annelida—have very different adult body plans. The molluscs are a very diverse phylum, including snails, bivalves (e.g., clams), and cephalopods (squid and octopus). The soft part of the molluscan body (the mantle) supports a muscular foot, a rasping radula (feeding appendage) and, in many taxa, secretes a calcareous shell. Annelids—including marine polychaetes, terrestrial earthworms, and leeches—all have a generally vermiform (wormy) construction, essentially a tube with a hole at each end, but their key defining feature is segmentation. Annelid bodies are built from a series of "rings," each of which contains a portion of the internal structures such as muscles, nerves, and digestive organs. So one of the major lineages of the Lophotrochozoa (Annelida) is highly segmented, and the other major lineage (Mollusca) is essentially unsegmented. Based on this information alone, it is difficult to predict whether the ancestral lophotrochozoan was segmented or not. But consideration of the minor lophotrochozoan phyla such as Echiura (spoonworms) and Sipuncula (peanut worms) might make the path of evolution of segmentation clearer.

Peanut worms have been described as "astonishingly unprepossessing creatures" (Tudge 2000). Shaped essentially like a shelled peanut with a tail, they generally live in shallow marine sediments. The sipunculid body plan is predominantly taken up with the digestive tract: The tentacle-surrounded mouth at the end of the "tail" leads to the gut, which is twisted around within the "peanut," with simple eyespots, chemoreceptors, muscles, and a brain (if fused pair of cerebral ganglia can be called a brain; Cutler 2001). Developmental studies show that, although sipunculan larvae show serially repeated structures, like muscle rings, these form simultaneously, not by the serial addition of segments (Wanninger et al. 2005). So sipunculan development does not follow the same pattern as annelids, where the segments are added sequentially to the growing embryo. At this point, you may well be tempted to say "well who cares how sipuncula grow their wormy little bodies?" But the importance of this finding becomes evident when it is combined with molecular phylogenetic data.

Although sipunculans have classically been considered to be most closely related to molluscs, particularly on the basis of similarity in the pattern of cells in early embryonic stages (referred to as the "molluscan cross"), molecular phylogenies tend to group sipunculans with the echiurans and annelids (Boore and Staton 2002; Schulze, Cutler, and Giribet 2007; Wanninger et al. 2005). If Sipuncula are more closely related to annelids than they are to molluscs, then there are two ways of explaining the patterns of segmentation in the metazoan tree. One possible explanation is that segmentation is a shared ancestral feature of bilaterians, present in the lophotrochozoan ancestor, then two or three lophotrochozoan phyla all independently lost annelid-style segmentation (Mollusca, Sipuncula, and Echiura; see Bleidorn 2007; Stuck et al. 2007). New results suggesting a role for the *Notch* pathway in annelid segmentation may support this hypothesis (Rivera and Weisblat 2008). The alternative explanation is that the lophotrochozoan ancestor was unsegmented, and

segmentation was independently derived in the annelid lineage (figure 13.6), thus not homologous to segmentation in arthropods and chordates (see discussions in Jenner 2000; Seaver and Kaneshige 2006; Tautz 2004). Either way, these case studies suggest that segmentation, a fundamental body plan character, seems to evolve along lineages just as other characteristics do.

What Is Special About Body Plan Characters?

Each of the case studies described in this chapter has a relatively small focus, such as determining the phylogenetic position of a fairly unsexy marine worm, so it may seem that they have little to tell us about the big picture of the evolution of complex animals. However, if the results of the case studies are reliable, then they point toward some important conclusions. The hemichordates (acorn worms) suggest that animal body plans were not fixed once they arose; instead, a lineage can begin with one body plan, then evolve an entirely different one. The Sipuncula (peanut worms) tell us that fundamental body plan characters such as segmentation can evolve along lineages, being lost, gained, or remodeled, just like other aspects of phenotype or development. The cockroaches show that the developmental mechanisms that specify fundamental body plan characters can change dramatically, so were not all fixed in the early diversification of animal phyla.

Of course, I may have chosen examples where body plan characters are mutable, and ignored cases where they appear to have a discrete origin in the Cambrian. But it is difficult to think of an example where body plan characters do not behave as any other character does, being conserved in some lineages and changed in others. For example, genes underlying the formation of the anterior-posterior body axis formation are remarkably conserved in function in flies and vertebrates, so these genes have been considered fundamental to the bilaterian body plan (hence the rainbow-colored diagram comparing hox gene expression in fly and mouse embryos now found in every biology textbook). Because phyla may differ in their *Hox* gene complements or patterns of expression, changes in *Hox* number or expression have been proposed as a mechanism for generating different body plans in animal phyla (e.g., Tour and McGinnis 2005). But *Hox* genes were not invented in the Cambrian explosion: They were present in the metazoan lineage from the beginning (Garcia-Fernandez 2005). Nor are changes in *Hox* gene number, expression patterns, or functions peculiar to the Cambrian explosion, as all of these aspects of *Hox* genes have changed before, during, and after the Cambrian explosion (e.g. Lanfear and Bromham 2008).

Hox genes, though clearly important and fascinating, do not appear to have a pattern of evolution that suggests their creation or modification provided a special basis for the evolution of body plans, generating large changes during a particular evolutionary period and then being immune to subsequent change. The same may be said for other developmental genes: while some are highly conserved, it is possible to find enough exceptions to show that they are evolutionarily labile, even for fundamental patterns set very early in

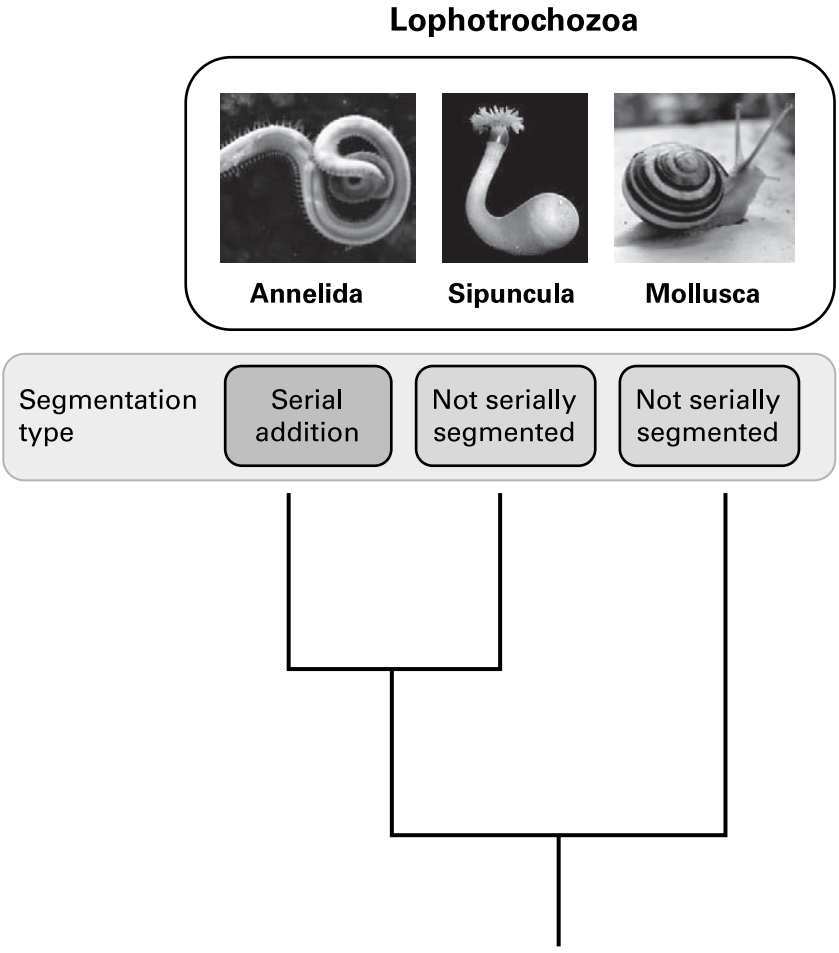


Figure 13.6
The molecular evidence for the phylogenetic placement of the Sipuncula is, like many minor phyla, still equivocal; however, several studies suggest that they are more closely related to annelids than to their traditional allies, molluscs. If true, this suggests that the annelid-style segmentation, formed by the serial addition of segments in the embryo, has been derived within the Lophotrochozoa and is not homologous to eusegmentation in the arthropods and chordates.

embryonic development. For example, one of the key developmental genes involved in dorsoventral patterning in flies is highly conserved among insects, but no homologs of this gene have been found in other arthropods (Copley 2008). This suggests a new gene that has arisen in insects has formed the basis of the adoption of an entirely new way of forming the fundamental axes of the embryo. So, although we cannot fail to be impressed by the conservation of developmental genes between disparate phyla, even very early embryonic developmental programs are able to change dramatically between related animal lineages.

Lessons from Eyes: Body Plans and Developmental Genes Are Normal Evolutionary Traits

The effect of conservation and change on body plan characters can be illustrated by considering the evolution of the wonderfully metazoan invention, the complex eye ("complex" refers to some level of spatial vision, where the direction and intensity of light can be detected, as opposed to simple light sensitivity). The pre-Cambrian ediacarans show no sign of having eyes, but many Cambrian animals have gloriously large and complex eyes (though, of course, many have no obvious eyes at all). The appearance of sight in the Cambrian has even been proposed as the primary cause of the animal radiation (Parker 1998). The most notable feature of metazoan eyes is their remarkable diversity. At first glance, the body plans of the different phyla seem to be characterized by fundamentally different kinds of eyes, such as the single-chambered eyes of vertebrates or the compound eyes of arthropods. But a closer inspection reveals a pattern of evolution like any important trait: a great deal of conservation, yet a surprising frequency of change. For example, compound "flies-eyes" are a key feature of Arthropoda, but arthropod visual systems have been remarkably labile. Not only has the form of the compound eye varied substantially between lineages (see the following discussion of lepidopteran eyes), but some arthropod lineages have independently evolved entirely different forms of eyes, such as the mirror-based eyes of the deep-sea ostracod *Gigantocypris,* or the camera-type (single chamber) eyes in spiders. New types of eyes appear at all levels of the arthropod phylogeny, from species to subphyla. Conversely, compound eyes have evolved independently in a number of other phyla, and can be found in a family of tube worms, some genera of bivalves, and also in some starfish (see Land and Nilsson 2002).

How could such a fundamental and highly engineered aspect of body plan as the complex eye evolve from one kind to another? Land and Nilsson (2002) present an extreme example. Most butterflies and some moths have classic "apposition" compound eyes, where the eye is made of many separate units, each with a lens that forms a separate image. But some moths and butterflies have an entirely different arrangement, the "superposition" compound eye, where the single retina is deep within the eye and multiple lenses work together to form a single erect image. "It is not very easy to see how it is possible to get from one type of eye to the other, without going through an intermediate that doesn't work." Yet this switch between apposition and superposition eyes has happened many

times in the Lepidoptera (butterflies and moths), and also several times in other insect groups. Land and Nilsson describe a possible path between the two types of eyes that involves incremental changes: "[T]o become nocturnal, the powers of the distal and proximal lenses must become more equal, the receptor later moves to a deeper location, and gradually more and more facets contribute to the image. There are no blind intermediaries." Complex eyes that seem discontinuously variable are apparently as able to be altered by evolution as other key aspects of phenotype.

The developmental genetics of eyes shows a similar pattern of conservation and change to that of the morphology. Much excitement has been generated by the demonstration that some of the genes that trigger eye development are conserved between animal phyla, most notably the homeobox-containing *Pax6* gene. The homeobox sequence of the *Pax6* gene is sufficiently conserved between taxa that the sequence from one species can trigger eye formation in a very distantly related species. For example, the *Pax6* sequence of a mouse can cause the formation of eye tissue in flies. The *Pax6* transfer experiments produced some of the most exiting scientific images of the past century, with unfortunate flies with ectopic (out-of-place) eyes on their limbs, antennae, wings, foreheads, and wherever else *Pax6* was expressed (Halder, Callearts, and Gehring 1995).

But *Pax6* is not a simple "master switch" for eye formation (Wilkins 2002). A number of other developmental genes are required to generate functioning eyes; there are at least half a dozen genes in *Drosophila* that can trigger ectopic eye formation, and, in both flies and vertebrates, part of the eye can still develop even when *Pax6* is knocked out (Pichaud and Desplan 2002). Interestingly, while the expression of *Pax6* may promote eye development, in some cases the presence of an eye can promote *Pax6* expression: *Pax6* expression can be induced in eyeless cavefish by transplanting a lens from a related species with a functional eye (Yamamoto and Jeffrey 2000).

The conservation of the *Pax6* gene across phyla is striking, but not unusual. The *Pax* gene family plays many important roles in nervous system development and organogenesis, so it is not surprising that these genes tend to be well conserved. But it is only the 60-amino-acid active site that is conserved between phyla; the rest of the gene sequence has acquired so many changes that it has lost any recognizable similarity (Morgan 2004). Many other genes with important roles in metabolism, physiology, and development show much greater levels of sequence conservation than *Pax6*, otherwise we would not be able to use DNA sequence data to uncover metazoan relationships. For example, there is a thirteen-amino-acid sequence in one of the active sites of DNA polymerase that has been stable for billions of years, such that it is virtually identical in a wide range of bacteria and similar in most other prokaryotes and eukaryotes (Bromham 2000; Patel and Loeb 2000). And although the transfer of *Pax6* between phyla is mightily impressive, the ability to move genes between distantly related species and have the genes function normally is not confined to developmental genes, as can be noted from frequent horizontal gene transfers, both natural and artificial. Genetic engineering would be a nonstarter if this were not so.

Nonetheless, the strikingly similar role of *Pax6* in eye development in many disparate metazoans has been taken as evidence that this gene is part of a developmental toolkit that evolved in the bilaterian stem lineage that enabled the evolution of complex eyes. But the invention of *Pax6* was neither necessary nor sufficient for the evolution of complex eyes in metazoans. Cnidarians have *Pax*-like genes that appear to be related to the *Pax* genes in bilateria. Indeed, two different *Pax* genes (*paxB* and *paxC*) from the coral *Acrospora* can cause the formation of ectopic eyes in *Drosophila* (despite the fact that these genes do not, of course, trigger eye formation in *Acrospora* itself; Miller et al. 2000). Conversely, the cnidarian hydromedusa produces a complex lens eye without the involvement of its *paxB* and *paxC* genes (Sun et al. 2001). If cnidarians have *Pax* genes that can trigger eye formation, but they can develop complex eyes without them, then this suggests the acquisition of a bilaterian developmental toolkit was not a prerequisite for the formation of complex eyes. Furthermore, post-explosion metazoans that have inherited the *Pax6* gene can find alternative ways of directing eye formation; for example, the development of adult eyes in the polychaete *Platynereis dumerilii* (Arendt et al. 2002) and eye regeneration in planarians (Pineda et al. 2002) are apparently *Pax6* independent. The association of *Pax6* with eye development in different metazoans is truly fascinating, but it seems to have all the characteristics of a "normal" evolutionary trait: conserved in many related taxa, yet changed in others.

Is the Cambrian Explosion Incompatible with Darwinian Gradualism?

When we stand back and look at the big picture of animal evolution, we see unbridgeable gaps between the phyla that appear to have their origin in a single evolutionary event. This has led some researchers to the conviction that body plan characters had a discontinuous origin, forming by the acquisition of a few large changes rather than the gradual accumulation of many small changes. Here, I have argued that it is sometimes helpful to take a small-picture approach, seeing if the predictions of the discontinuous hypothesis hold true for particular case studies, in addition to fitting the overall big picture. The small pictures presented here suggest it is possible for a lineage to begin with one body plan and evolve an entirely new one, and that even fundamental body plan characters like segmentation can evolve along lineages. These small pictures call into question some explanations for the suddenness and uniqueness of the metazoan radiation by showing that body plan characters and the genetic architecture that governs their development did not all arise in a single evolutionary event and then were unable to change further.

Conserved similarities in the ground plan of members of a phylum may impress, but conservation exists at all levels of the taxonomic hierarchy. We can pick any level of metazoan organization and choose characters that are largely invariant within groups at that level. All metazoans have wall-less cells joined by particular kinds of cell junctions. Within the metazoans, all ecdysozoans have a cuticle that is molted as the animal grows. Within the ecdysozoans, all arthropods have a chitinous exoskeleton. Within the arthropods, all

insects have three body segments, the middle of which bears three pairs of jointed appendages. Within the insects, all flies are defined by having one set of wings and one set of halteres, and so on. This hierarchy of conservation and change produced by continuous evolution is the basis of systematics and phylogenetics. To classify organisms, we select characters with a rate of change appropriate to the depth of divergence we wish to delineate. This is most obvious with molecular systematics: Choose a fast-changing gene to distinguish populations, a slow-changing gene to distinguish phyla, and an extremely highly conserved gene to study the relationships between kingdoms. But it is also the basis of morphological systematics: Choose highly labile traits like color, mating call, or bristle number to distinguish species; choose conserved traits like number of legs, shell material, or embryological characters to define phyla. Traits that define animal phyla are those traits that change at an appropriate rate so that they tend to differ between phyla but are more likely to be constant within phyla. There is no reason to be surprised that body plan characters are conserved within phyla, since that is commonly how they are defined in the first place (see also Budd 1999).

The continuous scale of conservation and change can be seen for most evolved characters, including genes and developmental patterns. The homeobox-containing genes are by no means unusual in their level of conservation between phyla, as a great many genes have this level of conservation between animal phyla, involved in metabolism, physiology, cell function, and so forth. Not surprisingly, these are the genes typically selected for phylum-level phylogenetic studies. Yet, as far as I know, nobody is proposing that we need a macroevolutionary mechanism to explain the conservation of form and function of metabolic genes. All we need to do is assume that these enzymes are so important that changes are rare. As with any other evolutionary character, we cannot assume that differences in genes or expression patterns between lineages played a causal role in the formation of the lineages, as they may have accumulated subsequent to lineage divergence. Similarly, it is possible to trace the history of languages by comparing the pattern of shared words (Gray and Atkinson 2003); we do not expect that the origin of new words actually caused the division of humans into separate language groups, but that language differences are simply an inevitable consequence of population divergence.

Recognition of distinct differences between the way arthropods, annelids, echinoderms, and chordates are put together does not mean that body plan characters evolved in a distinctly different way to other aspects of phenotype or development. Indeed, "unbridgeable gaps" between ancient lineages are expected under a gradual model of divergence, due to the extinction or modification of the intermediate lineages. As lineages diverge, some traits remain similar and some change, whether by drift or by selection. The longer two lineages have been separated, the more different they will be, and the more likely that lineages showing gradations between them will have gone extinct. Long-separated lineages are likely to have some conserved traits in common, potentially some convergent traits, and very many traits that differ. The impression of unbridgeable gaps may also be heightened

by the way that body plan differences are categorized. Body plan characters tend to be discrete, such as number of limbs, presence of eyes, type of skeleton, developmental origins of organs, and so on. Discrete traits such as these can, by definition, change only by whole numbers or entire categories, so may appear to evolve by "jumps."

Conclusion

There is clearly a remarkable increase in animal complexity from the Precambrian to the Cambrian periods. The rate of body plan evolution across the Precambrian-Cambrian boundary may have been high, but there is currently a lack of evidence to suggest that the pace of evolutionary change during this period depended critically on the formation of a genetic architecture that then constrained future innovation in body plan or development.

Raff (1996) explores the notion of inflexible body plans with the following thought experiment:

> Suppose that through some incredibly bad luck, all animal phyla became extinct except echinoderms. Could the survivors eventually evolve into new phyla? They would start from within a unique and unpromising body plan with tube feet, a water vascular system, a circular "brain" and pentameral symmetry. Could they evolve bilaterian symmetry and various features that we associate with the other phyla, or would they go on munching algal mats until the Sun swells into a red giant and parboils the entire lot? There is a lot of variability within the echinoderms, and they have done some remarkable things. One group of sea urchins, the heart urchins, has evolved a secondary bilateral symmetry. Who in Paleozoic times would have predicted that one group of crinoids, the comatulids or feather stars, would lose their stems and become highly motile, swimming or walking with their arms and clinging to their perches with cirri that resemble multiple articulated appendages? I don't know the answer to this question, but I'm sure that in the right circles a lot of beer and peanuts could be consumed while it was being debated.

Could one body plan give rise to a radically different one? Yes, the chordate body plan gave rise to the echinoderm body plan. Could bilateral symmetry evolve again? Yes, it has several times in the deuterostomes (e.g., sea cucumbers, heart urchins). Could aquatic creatures with a water vascular system ever hope to colonize the land? Well, plenty of other aquatic lineages did so in the past (and some are in the process of doing so today if the land-based crayfish that live in the mud in some Australian forests are anything to go by).

How, then, do we explain the near-simultaneous appearance of phylum-level differences in the Cambrian, and the failure to generate any similar level of variation since? Under a gradualist model, there is no need to worry about the lack of new phyla arising since the Cambrian, because this is explained by the positive relationship between time and divergence: A phylum is the amount of change you expect to accumulate over half a billion years or so, therefore they cannot appear overnight (but come back half a billion years from now, and those Australian crayfish may have given rise to a new phylum). But the gradualist

model still requires an explanation of why the speed of the increase in animal diversity and disparity over the Cambrian boundary is so much greater than that witnessed at other times.

Three key areas of investigation are needed to explore the adequacy or otherwise of a gradualist model of phylum divergence. One is the timing and duration of the radiation. Both paleontological and molecular studies have been used to suggest that bilaterian lineages arose and began diversifying well before the Cambrian. This makes the radiation of metazoan lineages look less explosive but raises more unanswered questions: If many bilaterian lineages existed in the Precambrian, then why did obvious body plan features like legs, eyes, and shells not appear in the fossil record until the Cambrian? Similarly, we cannot attribute differences between these lineages to a causal role in the original speciation event that created the two lineages without a way of determining which of the differences occurred at the original divergence and which were accumulated afterwards. Second, we need an expectation of how much change we could expect under a gradualist model in this time period, so that we can judge what observed level of change would be incompatible with a continuous model. This is not easy, but some attempts have been made to create biologically-informed models of body plan character changes, such as the evolution of a complex eye (Nilsson and Pelger 1994). If we wish to reject a neo-Darwinian explanation of patterns of conservation and change, we need a clearer picture of what any given period of microevolutionary divergence can produce, rather than relying on gut feeling. Third, we need to know if body plan–level characters could arise today and be perpetuated. This seems intuitively unlikely, but a dispassionate exploration would need a taxonomy-free definition of what constitutes a body plan character.

Any investigations of body plan evolution rely critically on using an unbiased way of recording change between lineages over evolutionary time, to avoid the circularity of defining as body plan difference only those that have a discontinuous distribution between phyla. We need a definition of a body plan that is not tied up with taxonomic level or depth of divergence. For example, a naïve observer might suggest that the sausagelike sea cucumbers, flowerlike crinoids, spiky sea urchins, and five-pointed starfish all had different basic body plans. Of course, a zoologist would counter that these animals all do share the common features of the Echinodermata. But the examples given in this chapter demonstrate that, at least in some cases, the relationship between body plan and phylum can become blurry when the minor phyla are considered. Hemichordates, for example, have some of the body plan characteristics of chordates, and some of echinoderms. Sipuncula are currently classed as a phylum, but some consider that they should be subsumed within the phylum Annelida. Insects have body plan innovations not seen in other lineages, such as wings, yet are nested within the phylum Arthropoda. Consideration of the unsexy minor phyla tempers some of the apparent "unbridgeable gaps" between phyla and counters some of the claims about discontinuous body plan evolution.

An unbiased assessment is also required when testing the consequences of the evolution of certain body plan features. For example, the three eusegmented phyla—Arthropoda,

Chordata, and Annelida—are among the most diverse and ubiquitous of the metazoan phyla, so segmentation has sometimes been considered a key innovation that, at least in part, accounts for the evolutionary success of these lineages, by giving them a kind of developmental flexibility that has allowed the evolutionary of many forms and ways of life. But then the molluscs, nematodes, and platyhelminths are just as successful, and they are not eusegmented (see figure 13.1). In fact, the two "preexplosion" metazoan phyla, the diploblasts Porifera (sponges) and Cnidaria (corals, jellyfish, and allies), both make it into the top ten most diverse phyla, despite their presumed lack of any inventions that could have triggered the bilaterian radiation. Molecular data, which records evolutionary history essentially independently of the record in phenotypes, developmental patterns, or fossil forms, might provide a level playing field in which all taxa are equal and body plan characters can be traced as they evolve along the evolutionary tree.

Acknowledgments

Many thanks to Brett Calcott, Rob Lanfear, Kim Sterelny, Andy Knoll, and Matt Phillips for their help with this chapter. Also thanks to John Maynard Smith, in memoriam, for his encouragement and enthusiasm: lunchtimes will never be the same again.

Note added in proof: This chapter was accepted for publication in 2008.

References

Akam, M. (1989). *Drosophila* development: Making stripes inelegantly. *Nature* 341: 282–283.

Arendt, D., K. Tessmar, M. I. de Campos-Baptista, A. Dorresteijn, and J. Wittbrodt. (2002). Development of pigment-cup eyes in the polychaete *Platynereis dumerilii* and evolutionary conservation of larval eyes in Bilateria. *Development* 129: 1143–1154.

Aris-Brosou, S., and Z. Yang. (2003). Bayesian models of episodic evolution support a late Precambrian explosive diversification of the Metazoa. *Molecular Biology and Evolution* 20: 1947–1954.

Arthur, W. (2000). The concept of developmental reprogramming and the quest for an inclusive theory of evolutionary mechanisms. *Evolution & Development* 2: 49–57.

Baguna, J., and J. Garcia-Fernandez. (2003). Evo-Devo: The long and winding road. *International Journal of Developmental Biology* 47: 705–713.

Balavoine, G. and A. Adoutte. (2003). The segmented *Urbilateria*: A testable scenario. *Integrative and Comparative Biology* 43: 136–147.

Bambach, R. K., A. M. Bush, and D. H. Erwin. (2007). Autecology and the filling of ecospace: Key metazoan radiations. *Paleontology* 50: 1–22.

Bengston, S. (2002). Origins and early evolution of predation. *Paleaontological Society Papers* 8: 289–317.

Blair, J. E., and S. B. Hedges. (2005). Molecular clocks do not support the Cambrian explosion. *Molecular Biology and Evolution* 22: 387–390.

Bleidorn, C. (2007). The role of character loss in phylogenetic reconstruction as exemplified for the Annelida. *Journal of Zoological Systematics and Evolutionary Research* 45: 299–307.

Boore, J. L., and J. L. Staton. (2002). The mitochondrial genome of the sipunculid *Phascolopsis gouldii* supports its association with Annelida rather than Mollusca. *Evolution* 19: 127–137.

Bourlat, S., T. Juliusdottir, C. Lowe, R. Freeman, J. Aronowicz, M. Kirschner, et al. (2006). Deuterostome phylogeny reveals monophyletic chordates and the new phylum Xenoturbellida. *Nature* 444: 85–88.

Bromham, L. (2000). Conservation and mutability in molecular evolution. *Trends in Ecology & Evolution* 15: 355.

Bromham, L. (2006). Molecular dates for the Cambrian explosion: is the light at the end of the tunnel an oncoming train? *Palaeontologia Electronica* 9: 2E.

Bromham, L. D., and B. M. Degnan. (1999). Hemichordates and deuterostome evolution: robust molecular phylogenetic support for a hemichordate+echinoderm clade. *Evolution & Development* 1: 166–171.

Budd, G. E. (1999). Does evolution in body patterning genes drive morphological change—or vice versa? *BioEssays* 21: 326–332.

Budd, G. E. (2006). On the origin and evolution of major morphological characters. *Biological Reviews* 81: 609–628.

Budd, G. E. (2008). The earliest fossil record of the animals and its significance. *Philosophical Transactions of the Royal Society B–Biological Sciences* 363: 1425–1434.

Butterfield, N. J. (2007). Macroevolution and macroecology through deep time. *Palaeontology* 50: 41–55.

Cameron, C. B., J. R. Garey, and B. J. Swalla. (2000). Special feature: Evolution of the chordate body plan: New insights from phylogenetic analyses of deuterostome phyla. *Proceedings of the National Academy of Sciences USA* 97: 4469–4474.

Carroll, R. C. (2000). Towards a new evolutionary synthesis. *Trends in Ecology & Evolution* 15: 27–32.

Carroll, S. B. (2005). *Endless Forms Most Beautiful: The New Science of Evo-Devo and the Making of the Animal Kingdom.* New York: W. W. Norton.

Castresana, J., G. Feldmaier-Fuchs, S. Yokobori, N. Satoh, and S. Paabo. (1998). The mitochondrial genome of the hemichordate *Balanoglossus carnosus* and the evolution of deuterostome mitochondria. *Genetics* 150: 1115–1123.

Copley, R. R. (2008). The animal in the genome: Comparative genomics and evolution. *Philosophical Transactions of the Royal Society B, Biological Sciences* 363: 1453–1461.

Cutler, E. B. (2001). Sipuncula, in *Encylopedia of Life Sciences*. John Wiley & Sons.

Darwin, C. (1859). *The Origin of Species by Means of Natural Selection*. London: John Murray.

Davidson, E. H., and D. H. Erwin. (2006). Gene regulatory networks and the evolution of animal body plans. *Science* 311: 796–800.

Erwin, D. H. (2004). One very long argument. *Biology and Philosophy* 19: 17–28.

Erwin, D. H. (2007). Disparity: morphological patterns and developmental context. *Palaeontology* 50: 57–73.

Fedonkin, M. A., and B. M. Waggoner. (1997). The late Precambrian fossil *Kimberella* is a mollusc-like bilaterian. *Nature* 388: 868–871.

Fortey, R. A., D. E. G. Briggs, and M. A. Wills. (1996). The Cambrian evolutionary "explosion": Decoupling cladogenesis from morphological disparity. *Biological Journal of the Linnean Society* 57: 13–33.

Garcia-Fernandez, J. (2005). The genesis and evolution of homeobox gene clusters. *Nature Reviews Genetics* 6: 891–992.

Gellon, G., and W. McGinnis. (1998). Shaping animal body plans in development and evolution by modulation of Hox expression patterns. *BioEssays* 20: 116.

Grant, P. R. (2001). Reconstructing the evolution of birds on islands: 100 years of research. *Oikos* 92: 385–403.

Gray, R. D., and Q. Atkinson. (2003). Language-tree divergence times support the Anatolian theory of Indo-European origin. *Nature* 426: 435–439.

Halder, G., P. Callearts, and W. J. Gehring. (1995). Induction of ectopic eyes by targeted expression of the *eyeless* gene in *Drosophila*. *Science* 267: 1788–1792.

Hinman, V., and B. D. Degnan. (2000). Retonoic acid perturbs *Otx* gene expression in the ascidian pharynx. *Development Genes and Evolution* 210: 129–139.

Jenner, R. A. (2000). Evolution of animal body plans: The role of metazoan phylogeny at the interface between pattern and process. *Evolution & Development* 2: 208–221.

Jenner, R. A. (2003). Unleashing the force of cladistics? Metazoan phylogenetics and hypothesis testing. *Integrative and Comparative Biology* 43: 207–218.

Jensen, S., M. L. Droser, and J. G. Gehling. (2005). Trace fossil preservation and the early evolution of animals. *Palaeogeography, Palaeoclimatology, Palaeocology* 220: 19–29.

Land, M. F., and D.-E. Nilsson. (2002). *Animal Eyes*. Oxford: Oxford University Press.

Lanfear, R., and L. Bromham. (2008). Statistical tests between competing hypotheses of Hox cluster evolution. *Systematic Biology* 57: 708–718.

Levinton, J. S. (2001). *Genetics, Palaeontology and Macroevolution*. Cambridge: Cambridge University Press.

Lovette, I. J., E. Bermingham, and R. E. Ricklefs. (2002). Clade-specific morphological diversification and adaptation in Hawaiian songbirds. *Philosophical Transactions of the Royal Society B, Biological Sciences* 269: 37–42.

Lyell, C. (1830). *Principles of Geology*. London: John Murray.

Miller, D. J., D. C. Hayward, J. S. Reece-Hoyes, I. Scholten, J. Catmull, W. J. Gehring, P. et al. (2000). Pax gene diversity in the basal cnidarian *Acropora millepora* (Cnidaria, Anthozoa): Implications for the evolution of the Pax gene family. *Proceedings of the National Academy of Science USA* 97: 4475–4480.

Morgan, R. (2004). Conservation of sequence and function in the Pax6 regulatory element. *Trends in Genetics* 20: 283–287.

Nilsson, D.-E., and S. Pelger. (1994). A pessimistic estimate of the time required for an eye to evolve. *Philosophical Transactions of the Royal Society B–Biological Sciences* 256: 53–58.

Parker, A. R. (1998). Colour in Burgess Shale animals and the effect of light on evolution in the Cambrian. *Philosophical Transactions of the Royal Society B, Biological Sciences* 265: 967–972.

Patel, P. H., and L. A. Loeb. (2000). DNA polymerase active site is highly mutable: Evolutionary consequences. *Proceedings of the National Academy of Science USA* 97: 5095–5100.

Peel, A. D., A. D. Chipman, and M. Akam. (2005). Arthropod segmentation: beyond the *Drosophila* paradigm. *Nature Reviews Genetics* 6: 905–916.

Peterson, K. J., J. A. Cotton, J. G. Gehling, and D. Pisani. (2008). The Ediacaran emergence of bilaterians: Congruence between the genetic and the geological fossil records. *Philosophical Transactions of the Royal Society B*, *Biological Sciences* 363: 1435–1443.

Pichaud, F., and C. Desplan. (2002). Pax genes and eye organogenesis. *Current Opinion in Genetics & Development* 12: 430–434.

Pigliucci, M. (2007). Do we need an extended evolutionary synthesis? *Evolution* 62: 2743–2749.

Pineda, D., L. Rossi, R. Batistoni, A. Salvetti, M. Marsal, V. Gremigni et al. (2002). The genetic network of prototypic planarian eye regeneration is Pax6 independent. *Development* 129: 1423–1434.

Pueyo, J. I., R. Lanfear, and J. P. Couso. (2008). Ancestral Notch-mediated segmentation revealed in the cockroach *P. americana*. *Proceedings of the National Academy of Science USA* 105: 16614–16619.

Raff, R. A. (1996). *The Shape of Life: Genes, Development, and the Evolution of Animal Form.* Chicago: University of Chicago Press.

Rivera, A. S., and D. A. Weisblat. (2008). And Lophotrochozoa makes three: Notch/Hes signaling in annelid segmentation. *Development Genes and Evolution* 219: 37–43.

Schulze, A., E. B. Cutler, and G. Giribet. (2007). Phylogeny of sipunculan worms: A combined analysis of four gene regions and morphology. *Molecular Phylogenetics and Evolution* 42: 171–192.

Seaver, E. C., and L. M. Kaneshige. (2006). Expression of "segmentation" genes during larval and juvenile development in the polychaetes *Capitella sp.* I and *H. elegans*. *Developmental Biology* 289: 179–194.

Stollewerk, A., M. Schoppmeier, and W. G. M. Damen. (2003). Involvement of Notch and Delta genes in spider segmentation. *Nature* 423: 863–865.

Stuck, T. H., N. Schult, T. Kusen, E. Hickman, C. Bliedorn, D. McHugh, and K. M. Halanych. (2007). Annelid phylogeny and the status of Sipuncula and Echiura. *BMC Evolutionary Biology* 7: 57.

Sun, H., A. Rodin, H. Zhou, D. P. Dickinson, D. E. Harper, D. Hewett-Emmett, and W-H. Li. (2001). Isolation of *Clodonema Pax-B* genes and studies of the DNA-binding properties of cnidarian Pax paired domains. *Molecular Biology and Evolution* 18: 1905–1918.

Tautz, D. (2004). Segmentation. *Developmental Cell* 7: 391–312.

Telford, M., and G. E. Budd. (2003). The place of phylogeny and cladistics in Evo-Devo. *International Journal of Developmental Biology* 47: 479–490.

Tour, E., and W. McGinnis. (2005). Hox genes and body plan: Evolution In *Encyclopedia of Life Sciences*. New York: John Wiley & Sons.

Tudge, C. (2000). *The Variety of Life: A Survey and Celebration of All the Creatures that Have Ever Lived.* Oxford: Oxford University Press.

Valentine, J. W. (2004). *On the Origin of Phyla.* Chicago: University of Chicago Press.

Wanninger, A., D. Koop, L. Bromham, E. Noonan, and B. M. Degnan. (2005). Nervous and muscle system development in *Phascolion strombus* (Sipuncula). *Development Genes and Evolution* 215(10): 509–518.

Wilkins, A. S. (2002). *The Evolution of Developmental Pathways*. Sunderland, MA: Sinauer.

Wray, G. A. (2001). Dating branches on the Tree of Life using DNA. *Genome Biology* 3(1): reviews 0001–0001.7.

Xiao, S., and M. Laflamme. (2008). On the eve of animal radiation: Phylogeny, ecology and evolution of the Ediacaran biota. *Trends in Ecology & Evolution* 24: 31–40.

Yamamoto, Y., and W. R. Jeffrey. (2000). Central role for the lens in cave fish eye degeneration. *Science* 289: 631–633.

IV CONCLUDING REMARKS

14 Concluding Remarks

Eörs Szathmáry and Chrisantha Fernando

The book *The Major Transitions in Evolution* was published in 1995. In 2003 the authors were planning a major revision of the work, which sadly did not materialize due to the death of John Maynard Smith in 2004. Aside from a general update of the content, we wanted to add some brand new items, including evolvable nanotechnology, artificial cells, and the nervous and immune systems. Some of these topics will be covered in another volume of the KLI book series, since many of the relevant insights also belong to what is now called an extended evolutionary synthesis (cf. Fernando and Szathmáry 2009a).

Origin of Living Systems

Artificial cells are important because, if the attempts meet with success, they reach into the domain of systems chemistry that deals with the analysis and synthesis of coupled autocatalytic chemical systems. Several of these systems belong to the category of infrabiological systems, showing some but not all important features of living systems (Szathmáry 2005; Fernando, Santos, and Szathmáry 2005). In the final analysis, a minimalist autonomous living system consists of three autocatalytic chemical systems: a metabolic engine, template replication, and membrane formation. The system as a whole is also autocatalytic, and can undergo spatial reproduction (Gánti 2003; Griesemer and Szathmáry 2009). We will not go into detail here, but we want to stress that the origin of evolvability is as much a problem in chemistry as in biology. As the renowned organic chemist Albert Eschenmoser said, the most important minds in chemistry should now deal with the origin of life. This entails dealing with not only the origin of biomolecular structures, but also the relevant and indispensable dynamics. Living systems are not in being, but in happening.

The emerging field of systems chemistry is dealing with establishing cooperative links between different types of nonenzymatic, autoacatalytic systems. If one considers autocatalytic metabolic networks, template replication, and membrane growth as the basic structural-dynamical building blocks, one can arrive at three different doublets, the experimental realization of which is a more immediate goal than that of the chemoton comprising

all three subsystems. As we shall see later, a systematic consideration of the three subsystems opens the door to an extended view of the major transitions.

Neuronal Replicators

As Jablonka and Lamb (2006) emphasized, the origin of the nervous system is a forgotten transition. They call attention to the fact that "the evolution of a nervous system not only changed the way in which information was transmitted between cells and profoundly altered the nature of the individuals in which it was present, it also led to a new type of heredity—social and cultural heredity—based on the transmission of behaviorally acquired information" (p. 236). We could not agree more; in fact, we (JMS and ES) planned to include the origin of the nervous system in the revised book. But there is a more radical view why the nervous system could merit special consideration within this transitions framework. This is because, at least in mammals and maybe also in birds, the nervous system may sustain a bona fide evolutionary mechanism, just as the adaptive immune system in vertebrates realizes real-time evolution for the generation of antibodies (Jerne 1985).

Of course, so-called Darwinian approaches to cognition (James 1890) and the dynamics of the nervous system (Changeux, Courrège, and Danchin 1973; Dawkins 1971; Edelman 1987) are not new, but James, in psychology, remained at the metaphorical level, and the biologists, in fact, always postulated purely selectionist rather than real evolutionary systems, in that the crucial element of multiplication (cf. Maynard Smith 1986) has always been missing from their expositions (see Fernando and Szathmáry 2009a,b for a detailed description of history and mechanisms). Spectacular cumulative adaptations and real cognitive novelties can arise, we believe, only in a full evolutionary system. Before our attempt, only Bill Calvin (1996) presented a sketchy model for the replication of neuronal activity patterns, but he did not consider how neuronal connectivity could be copied.

We presented the first model, including the latter aspect as well, very recently (Fernando, Karishma, and Szathmáry 2008). Without going into much detail, there are a number of possible approaches to describe evolution in, rather than of, the nervous system. It can be shown that dynamics at the level of synapses (for example, synapse strengthening in Hebbian learning) can be described in terms of natural selection (Adams 1998; Fernando and Szathmáry 2009b). Units of evolution also seem to be feasible at the level of neuronal assemblies (groups): First, one can copy, at a slower pace, in an activity-dependent fashion, the local connectivity of one assembly to the other (Fernando et al. 2008); second, one can copy much faster just the activity pattern (in simplest form the on-off states) of an array of neurons to another array of neurons (Fernando et al. 2009). The interesting question, of course, is whether such conceivable mechanisms really exist and what they bring to cognition. We suggest the most rewarding problems for application are insight problems, hypothesis formation, and complex thinking—in fact, any problem involving structured search. The proposed systems would work because they are envisaged to be coupled to the

reward system (including the effect of dopamine; see Dehaene and Changeux, 2000) that gives certain neuronal "hypotheses" feedback as to what act is behaviorally useful. Provided real evolutionary units exist in the brain, the reward would be exactly analogous to fitness.

One can argue that this line of thought is relevant for the major transitions since we face another instance of evolutionary units. This is true, but perhaps another consideration goes deeper than this. Can one image major evolutionary transitions within brain dynamics (Balázs Gulyás, personal communication)? We believe that the answer is affirmative. A mechanism to create higher-level units of evolution provides extra scope for solving complex problems by neuronal selection. Richard Watson's group has been at the forefront of delineating the algorithmic capabilities of various types of natural selection (Watson 2006). They have shown that, in a wide range of implementations, allowing symbiosis (major transition) of independently evolved partial solutions can solve problems in polynomial time that would take exponential time for a stochastic hill-climber. The kinds of problems these symbiotic algorithms excel in are those with interdependencies (epistasis), and structure that can be exploited. This rules out Kauffman's NK landscapes for they have epistasis, but no structure (Kauffman 1993). The archetypical problem of this type is the *h*ierarchical *if*-and-only-i*f* (HIFF) problem, which contains fractal local optima on which a hill-climber can get stuck, but is also highly structured (Watson, Hornby, and Pollack 1998). Symbiosis can solve problems that recombination cannot, for it does not depend on tight linkage, that is, it does not depend on the physical linkage on a chromosome corresponding to the functional linkage of alleles in solution space. Recently, in collaboration with Watson's group, we have shown that Hebbian learning can also be combined with neuronal replicators to provide structuring of exploration distributions (Toussaint 2003). An activity vector is evolved using a 1+1 evolutionary strategy (Beyer 2001) on a difficult problem such as HIFF. Once a local optimum is reached, Hebbian learning between the identical parent and child copies of the solution is undertaken. The activities are then randomized and evolution is restarted, but this time, replication occurs through the Hebbian matrix that was learned on the previous local optimum. If the learning rate for Hebbian learning is set low enough, the replication operation can have a memory of many previous local optima and channel the exploration distribution in this direction (Toussaint 2003). For certain kinds of problem, this allows the global optimum to be found more effectively (Fernando et al. 2009).

We know that complex problems are broken down into simpler ones, and we also know that some of these simpler problems are being worked on in parallel. One could assume that this strategy could yield perfect results; the solutions to the component subproblems fall into place by themselves. This is typically unlikely to be the case, however. Cutting the problem into subproblems may be suboptimal, and even if this is not the case, the joints between the subsolutions likely need to be streamlined for a better mutual fit. To use an analogy from another transition, although the acquisition of mitochondria and plastids for energy production and photosynthesis by an ancient eukaryotic cell was an undoubtedly

spectacular "jump" in evolution, an engulfed bacterium is a long way from a proper organelle, even if the bacteria involved are full of exaptations (see Maynard Smith and Szathmáry 1995 for details).

It is useful to follow the distinction made by Queller (1997) between the egalitarian and the fraternal major transitions. In the case of egalitarian transitions, the units that come together to form the higher-level unit are not alike, the initial advantage is combination of functions, and the greatest hurdle is the competition among the unlike units. The previously mentioned case of engulfed bacteria-turned-organelles is an egalitarian transition. In the case of fraternal transitions, the units coming together are alike, thus kin selection can operate, reproductive division of labor can arise, and the greatest hurdle is the initial advantage. If there is an initial advantage, it is likely to come from the economy of scale. The origins of animal, fungal, and plant multicellularity were fraternal transitions.

Before considering how these transitions may apply to neuronal replicators, we must stress again that evolution in the brain is more constrained than evolution in the wild, since, by virtue of past natural selection of the relevant genes, the brain acts as a "breeder" of ideas and the underlying neuronal representations. As we know, there is room during the transitions for the combination of functions, and since in the case of the brain this is organized by cutting the problem into subproblems, such a synergy is more likely to happen than in ordinary evolution, where egalitarian transitions were rare (the examples being the formation of protocells from unlinked genes, the formation of the eukaryotic cells with symbiotic organelles, and to some degree human groups with language). The conflicts among the unrelated replicators may also be less of a problem in the brain if the different neuronal replicators seeking the solution of different subproblems are provided limited habitats by a value system (the nature of which we leave open in this chapter).

How can the different component replicators be dynamically linked together? Whereas in natural evolution this is achieved by various forms of population structure (passive, as in evolution on a surface with limited mobility; or active, as in protocellular compartmentation of genes; cf. Könnyű, Czárán, and Szathmáry 2008; Szabó et al. 2002; Szathmáry and Demeter 1987), this is probably rarely the case for the nervous system. Occasionally, topographically adjacent but qualitatively different neuronal replicators might be "linked" together in the sense that they start multiplying together, but the form of "cellular" compartmentation, after which the whole group would freely move around, is certainly not an option in nerve tissue. Moreover, more often than not, complex thinking would presumably require linking together replicators from different brain areas. How could this linkage (binding) be achieved? A feasible, and appealing, idea that can be applied here is solving the binding problem by synchronous firing and/or oscillations (Singer 1998; Yu et al. 2008). First, there is an important structural property: In the cat visual cortex, and very possibly cortexwide, the functional connectivity forms a small world; there is "coexistence of local and global computations: feature detection and feature integration or binding" (Yu et al. 2008, p. 2891). One could say that the small world property is ideal for the coexis-

tence of local and integrated neuronal replicators: local replicators for subproblems and higher replicators for the whole problem. Second, as was previously pointed out, "evolved brains use assembly codes for the representation of contents and that these assemblies become organized through transient synchronization of the discharges of associated neurons" (Singer 1998, p. 1829). A higher-level neuronal replicator could partly be defined by such an assembly code.

It is easy to see that this cannot be a full solution of the whole problem, since it is not sufficient to link the two *populations,* A and B (for the two subproblems), together; one must establish links between *individuals* in the two populations, such as a1b2, a3b2, a4b8, and so on, and let these higher-level units multiply and then perform selection on them. The question is how to understand the formation of alternative "neuronal chromosomes" and how to understand the assignment of differential fitness to such linked entities rather than different lower-level replicators, analogous to genes. Uhlhaas and colleagues (2008) summarize evidence in favor of the idea that synchrony at multiple time scales serves different functions. Electroencephalograms and related investigations distinguish between theta (4 to 7 Hz), alpha (8 to 12 Hz), beta (13 Hz), and gamma (30 and 200 Hz) bands of oscillation. Theta and alpha seem to be active in top-down control and long-range synchronization. The beta band is active in long-range synchronization and sensory gating. The gamma band seems to ensure local synchronization. Both theta and gamma are involved in synaptic plasticity at different time scales. The linking of the different bands can be achieved in various ways; an interesting option is *n: m* phase synchrony, which indicates amplitude-independent phase locking of *n* cycles of one oscillation to *m* cycles of another oscillation (Palva, Palva, and Kaila 2005). Evidence is accumulating in favor of this mechanism (cf. Palva and Palva 2007). Undoubtedly, theta and alpha can serve consciousness and the establishment of a "global workspace" (Changeux and Michel 2006), but they may contribute to the linking of different replicators in various parts of the brain, that would locally replicate using the gamma band. Synaptic plasticity linked to the theta and gamma bands could also serve higher- and lower-level replication, respectively.

The previous paragraph makes it plausible that binding of distant neuronal replicators is possible, but still does not explain how different "chromosomes" could be maintained. However, it may not be necessary to follow the chromosome analogy very closely: It may be sufficient that, occasionally (in the theta band), successful replicators from the qualitatively different local arenas would be sampled, represented, and assembled, and the assemblies assessed in the global workspace, with a critical role of the prefrontal cortex. Analysis of the assemblies would then seed the local arenas with novel tasks (allowing for modifying the frame, so critical for insight problems), until the problem is solved or abandoned. Local evolution of the higher-level unit is also possible since the gamma band is found in all cortical structures. The process may or may not be conscious, but as we know from experience, the higher-level assemblies occasionally rise to consciousness during the task, and they certainly "pop into it" when the task is solved.

A possible function of neuronal "chromosomes" is suggested by recent work in the field of reinforcement learning (Sutton and Barto 1998). Andrew Barto, one of the inventors of reinforcement learning, states that today's machine learning algorithms "do not have the generative capacity required to significantly extend their abilities beyond initially built-in representations" (Barto et al. 2004). Typically, the most critical decision in applying temporal-difference methods is to determine how the state-action space is to be represented so that values can be assigned efficiently to state-action pairs, such that generalization to novel states and novel actions is possible. Because in large state-action spaces, it is not possible to exhaustively represent all possible pairs, supervised function approximation approaches have been used to design low dimensional parameterized spaces that represent the higher dimensional space of state-action pairs, for example, gradient-descent methods (Sutton and Barto 1998). More sophisticated methods of defining similar states can be used, for example, two states are similar in Kanerva coding if they agree on enough dimensions, even if they totally differ in others (Sutton and Barto 1998). The importance of forming low-dimensional representations of state-action pairs with which to associate value has also become evident in the field of intrinsic value systems (Oudeyer et al. 2007). These are approaches that generate internal rewards based on observation of learning progress on a given task. Such algorithms highlight the importance of state-action representation because they require comparison with previous performance on the same *task,* that is, for actions with the same *goal*. For such a comparison to be possible, improvement must be measured against past performance on the *same* and not a *different* problem. Therefore, there must be some mechanism of associating values with the problem they are trying to solve. For example, "intelligent adaptive curiosity (IAC)" incrementally divides the state-action space into regions, each containing a set of episodic memories. Initially, there is only one region. Splitting is done so that the sum of variances of the outcomes arising from executing the state-action pairs in both parts (to be) is minimized. That is, regions delineate distinct state-action pair types with distinct effects (Oudeyer et al. 2007). With each region is associated an expert that tries to predict the next state for that region alone. Learning progress is defined as the rate of error reduction in this prediction, and reward is defined as the learning progress. Q-learning is used for action selection based on this intrinsic reward signal. In a separate framework developed by Barto and colleagues, one part of the algorithm values novelty (i.e., promotes the execution of actions that have high prediction error), while the other part learns a value function (i.e., decreases prediction error), resulting in the emergent behavior that the overall algorithm also tends to produce continual increases in "predictivity" (Barto et al. 2004).

Returning to the problem of representations, this algorithm also incorporates a method of hierarchical definition of actions in the form of "options," a closed-loop control rule that can call other options (Sutton et al. 1999). Options say which states they can be executed in, they define a policy, and they say when they should terminate and what value should be assigned to particular kinds of terminal state. An "ecology" of options arises in the course

of experience (Minsky 1986), resulting in a process called "many-layered learning" (Utgoff and Stracuzzi 2002). Options are generated when (innately defined) salient stimuli are observed (Singh et al. 2005). These two methods (IAC and Options) both have the aim of structuring, assembling, and connecting action sequences, so that value can be effectively assigned, and used in action selection. We propose that the recombination of neuronal replicators may be the underlying mechanism by which such representations are formed and modified. Evolutionary methods have already been shown to be capable of producing representations for value functions that improves the performance of reinforcement learning (Whiteson et al. 2007). It is difficult to concieve how similar processes would not be necessary in the brain.

There are somewhat similar problem-solving architectures in the machine learning and evolutionary algorithms literature. For example, Jacobs and colleagues (1991) developed the so-called mixture of experts system, in which similar or different neural nets cooperate to find a solution, influenced by a gating network. In our case, the experts would be the local neuronal replicator populations, and gating would be given by the prefrontal cortex. Another similar abstract system is the cooperative coevolution architecture, which deliberately aims at producing coadapted subcomponents for problem solving (Potter and De Jong 2000). Here, the subpopulations are also isolated, but there is a central "domain model," which evaluates contributions to the "big problem," as it were, as follows. Individuals in the subpopulations evolve separately, but fitness values are given by the domain model. All subpopulations are evaluated in turn. For a chosen subpopulation, each individual is evaluated by the domain model that takes representative (best or randomly chosen) individuals from the other (currently not evaluated) subpopulations. This process is being carried out for all individuals of all subpopulations. A similar (but not necessarily identical) algorithm may work in human brains. Note the similarity of this algorithm to the symbiotic model discussed previously.

Clearly, the preceding speculations may turn out to be false or only vaguely correct, but we think that the topic is so important that it deserves every attention and the combination of expertise from widely different approaches.

Extended Major Transitions

Clearly, if there are neuronal replicators, and they can be active at various levels, this is a significant extension of the original scope. Let us offer a further extension that rests on Gánti's (2003) insight into the triple nature of life, with metabolism, boundary, and genome as the major key components. This offers us a chance to look at major transitions in these subsystems and how they are coupled together. As it is known, *The Major Transitions* focused mainly on the informational aspects of evolution. It may be time to take a broader view. Lenton, Schellnhuber, and Szathmáry (2004) provide a stimulus to this extension, linking major transitions to Earth history. It is not our task to present a full account of the

transitions in the three domains here, but we would like to illustrate what kinds of transitions we have in mind. For example, the transition to oxygenic photosynthesis, by the coupling of two independent photosystems to bridge the wide gap in redox potential, is a marked transition in metabolism. Incidentally, it allowed the appearance of the eukaryotic cell that now seems to have been intimately linked to the origin of the mitochondrion consuming oxygen in respiration. Also, plastids are descendents of engulfed cyanobacteria, who produced oxygen in the first place. Another major transition in metabolism is the origin of the closed circulation system we now find in vertebrates, annelids, and cephalopods (thus, this trait evolved independently at least twice). The rigid cell wall is an innovation in the boundary system, just as the formation of an exoskeleton in the Ecdysozoa (including arthropods). A coevolutionary analysis of the major transitions in all three key organismal components is a major and exciting research program for the future.

The major transitions idea has generated a lot of research by many able people, just as the original authors had hoped for. It can be extended in depth and breadth. An example of the former extension would be to consider how evolvability or the recent idea of niche construction (e.g., Laland, Odling-Smee, and Feldman 1999) is linked to the transitions. The idea of the emergence and cooperation of novel evolutionary units can be generalized to subjects where such a view has not yet been taken seriously. The origin of the immune systems awaits closer scrutiny, for example. In this chapter, we have indicated that an extension to the problem of competing and cooperating neuronal replicators may turn out to be rather spectacular.

References

Adams, P. (1998). Hebb and Darwin. *Journal of Theoretical Biology* 195: 419–438.

Barto, A. G., S. Singh, N. Chentanez. (2004). Intrinsically motivated learning of hierarchical collections of skills. *Proceedings of the Third International Conference on Development and Learning.*

Beyer, H.-G. (2001). *The Theory of Evolution Strategies*. Berlin: Springer.

Calvin, W. H. (1996). *The Cerebral Code. Thinking a Thought in the Mosaics of the Mind.* Cambridge, MA: MIT Press.

Changeux, J.-P., P. Courrège, and A. Danchin. (1973). A theory of the epigenesis of neuronal networks by selective stabilization of synapses. *Proceedings of the National Academy of Sciences, USA* 70: 2974–2978.

Changeux, J.-P., and C. M. Michel. (2006). Mechanisms of neural integration at the brain-scale level. In S. Grillner and A. M. Graybiel (eds.), *Microcircuits. The Interface between Neurons and Global Brain Function* (pp. 347–370). Cambridge, MA: MIT Press.

Dawkins, R. (1971). Selective neuron death as a possible memory mechanism. *Nature* 229: 118–119.

Dehaene, S., and J. P. Changeux. (2000). Reward-dependent learning in neuronal networks for planning and decision making. *Progress in Brain Research* 126: 217–229.

Edelman, G. M. (1987). *Neural Darwinism. The Theory of Neuronal Group Selection.* New York: Basic Books.

Fernando, C., K. K. Karishma, and E. Szathmáry. (2008). Copying of neuronal topology by spike-time dependent plasticity and error-correction. *PlosONE* 3(11): e3775.

Fernando, C., M. Santos, and E. Szathmáry. (2005). Evolutionary potential and requirements for minimal protocells. *Topics in Current Chemistry* 259: 167–211.

Fernando, C., and E. Szathmáry. (2009a). Chemical, neuronal and linguistic replicators. In M. Pigliucci and G. Müller (eds.), *Evolution: The Extended Synthesis* (pp. 209–249). Cambridge, MA: MIT Press.

Fernando, C., and E. Szathmáry. (2009b). The neuronal replicator hypothesis. In V. Goel, A. von Müller, and B. Glatzeder (eds.), *Towards a Theory of Thinking* (pp. 291–322). Berlin: Springer-Verlag.

Fernando, C., R. Goldstein, E. Szathmáry. (2010). The neuronal replicator hypothesis. *Neural Computation* 22(11): 2809–2857.

Gánti, T. (2003). *The Principles of Life.* Oxford: Oxford University Press.

Griesemer, J., and E. Szathmáry. (2009). Gánti's chemoton model and life criteria. In S. Rasmussen, M. A. Bedau, L. Chen, D. Deamer, D. C. Krakauer, N. H. Packard, and P. F. Stadler (eds), *Protocells: Bridging Nonliving and Living Matter* (pp. 481–512). Cambridge, MA: MIT Press.

Jablonka, E., and M. Lamb. (2006). The evolution of information in the major transitions. *Journal of Theoretical Biology* 239: 236–246.

Jacobs, R. A., M. I. Jordan, S. J. Nowlan, and G. E. Hinton. (1991). Adaptive mixture of experts. *Neural Computation* 3: 79–89.

James, W. (1890). *The Principles of Psychology.* Mineola, NY: Dover Publications.

Jerne, N. (1985). The generative grammar of the immune system. *Science* 229: 1057–1059.

Kauffman, S. A. (1993). *The Origins of Order.* New York: Oxford University Press.

Könnyű, B., T. Czárán, and E. Szathmáry. (2008). Prebiotic replicase evolution in a surface-bound metabolic system: Parasites as a source of adaptive evolution. *BMC Evolutionary Biology* 8: 267.

Laland, K. N., J. Odling-Smee, and M. W. Feldman. (1999). Evolutionary consequences of niche construction and their implications for ecology. *Proceedings of the National Academy of Scences. USA* 96: 10242–10247.

Lenton, T. M., H. J. Schellnhuber, and E. Szathmáry. (2004). Climbing the coevolution ladder. *Nature* 431: 913.

Maynard Smith, J. (1986). *The Problems of Biology.* Oxford: Oxford University Press.

Maynard Smith, J., and E. Szathmáry. (1995). *The Major Transitions in Evolution.* Oxford: Freeman.

Minsky, M. (1986). *The Society of Mind*. New York: Simon and Schuster.

Oudeyer, P.-Y., F. Kaplan, et al. (2007). Intrinsic motivation systems for autonomoys mental development. *IEEE Transactions on Evolutionary Computation* 11(2): 265–286.

Palva, J. M., S. Palva, and K. Kaila. (2005). Phase synchrony among neuronal oscillations in the human cortex. *Journal of Neuroscience* 25: 3962–3972.

Palva, S., and J. M. Palva. (2007). New vistas for α-frequency band oscillations. *Trends in Neuroscience* 30: 150–158.

Potter, M. A., and K. A. De Jong. (2000). Cooperative coevolution: An architecture for evolving coadapted subcomponents. *Evolutionary Computation* 8: 1–29.

Queller, D. C. (1997). Cooperators since life began. *Quarterly Review of Biology* 72: 184–188.

Singer, W. (1998). Consciousness and the structure of neuronal representations. *Philosophical Transactions of the Royal Society of London, B* 353: 1829–1840.

Singh, S., A. G. Barto, and N. Chentanez. (2005). Intrinsically motivated reinforcement learning. *Proceedings of Advances in Neural Information Processing Systems* 17: 1281–1288.

Sutton, R. S., D. Precup, et al. (1999). Between mdps and semi-mdps: A framework for temporal abstraction in reinforcement learning. *Artificial Intelligence* 112: 181–211.

Sutton, S. R., and A. G. Barto. (1998). *Reinforcement Learning: An Introduction*. Cambridge, MA: MIT Press.

Szabó, P., I. Scheuring, T. Czárán, and E. Szathmáry. (2002). *In silico* simulations reveal that replicators with limited dispersal evolve towards higher efficiency and fidelity. *Nature* 420: 360–363.

Szathmáry, E. (2005). Life: in search of the simplest cell. *Nature* 433: 469–470.

Szathmáry, E., and L. Demeter. (1987). Group selection of early replicators and the origin of life. *Journal of Theoretical Biology* 128: 463–486.

Toussaint, M. (2003). *The Evolution of Genetic Representations and Modular Adaptation.* ND 04, 44780, PhD thesis, Institut für Neuroinformatik, Ruhr-Universiät Bochum, Germany.

Uhlhaas, P. J., C. Haenschel, D. Nikolic, and W. Singer. (2008). The role of oscillations and synchrony in cortical networks and their putative relevance for the patho-physiology of schizophrenia. *Schizophrenia Bulletin* 34: 927–943.

Utgoff, P. E., and D. J. Stracuzzi. (2002). Many-layered learning. *Neural Computation* 14: 2497–2539.

Watson, R. A. (2006). *Compositional Evolution: The Impact of Sex, Symbiosis, and Modularity on the Gradualist Framework of Evolution*. Cambridge, MA: MIT Press.

Watson, R. A., G. S. Hornby, and J. B. Pollack. (1998). Modelling building-block interdependency. *PPSN-V* 1498: 97–106.

Whiteson, S., M. E. Taylor, et al. (2007). Empirical studies in action selection with reinforcement learning. *Adaptive Behavior* 15: 33–50.

Yu, S., D. Huang, W. Singer, and D. Nikolic. (2008). A small world of neuronal synchrony. *Cerebral Cortex* 18: 2891–2901.

Contributors

Lindell Bromham
Centre for Macroevolution and Macroecology, Evolution, Ecology, and Genetics, Research School of Biology, Australian National University

Brett Calcott
Research School of Social Sciences, Australian National University; and Centre for Macroevolution & Macroecology, Research School of Biology, Australian National University

Ellen Clarke
Department of Philosophy, University of Bristol

Chrisantha Fernando
Institute for Advanced Study, Collegium Budapest; Department of Informatics, University of Sussex; and Division of Mathematical Biology, National Institute for Medical Research, London

Peter Godfrey-Smith
Department of Philosophy. Harvard University

David Hewitt
Department of Organismic and Evolutionary Biology, Harvard University

Benjamin Kerr
Department of Biology, University of Washington

Andrew H. Knoll
Department of Organismic and Evolutionary Biology, Harvard University

Pamela Lyon
Discipline of Philosophy, University of Adelaide

Daniel W. McShea
Department of Biology, Duke University

Richard E. Michod
Department of Ecology and Evolutionary Biology, University of Arizona

Joshua Nahum
Department of Biology, University of Washington

Samir Okasha
Department of Philosophy, University of Bristol

Paul B. Rainey
New Zealand Institute for Advanced Study, Massey University

Carl Simpson
Museum für Naturkunde, Leibniz Institute at the Humboldt University Berlin

Kim Sterelny
Research School of Social Sciences, Australian National University; and Department of Philosophy, Victoria University, Wellington

Eörs Szathmáry
Collegium Budapest (Institute for Advanced Study); and Parmenides Foundation, Munich, Germany

Index

Abstraction, 35, 37, 39, 45–48, 65–66
Achaea, 6, 101
Adaptation, 1–2, 21–22, 66, 83–84, 94, 97–98, 106, 119, 152, 192, 218–220, 274, 302
  coadaptation, 92, 307
  group-level, 143–144, 146, 149, 243–244,
Additivity, 180–183
Ageing. *See* Life history
Aggregates, 9–10, 26–27, 54, 61–62, 128, 164–165, 173–174, 200–222, 252
Agriculture, 37
Alga, 26, 39–43, 71–72, 127, 142, 151, 176–192, 222, 251–264
  Astrephomene, 177
  Basichlamys, 176
Algorithms, 111, 113, 303, 306–307
Altruism, 105, 117–119, 130, 136, 141, 157–158, 170, 172–173, 175, 177–179, 210, 234. *See also* Cooperation
Amoeba, 21
Amphibians, 24
Analogy, 47–48. *See also* Convergence; Homology
Angiosperms, 240–241, 244–246
Anisogamy, 177
Annelids, 272, 277–278, 282–288, 292–295, 308
Anoxia. *See* Oxygen
Anthropocentrism, 2, 19, 22
Anti-outlaw. *See* Outlaw
Antibiotics, 93
Antioxidants. *See* Oxidation
Ants, 30, 47, 118, 141
Apoptosis, 234
Archaea, 83, 90–93, 101, 256, 260
Archaebacteria. *See* Bacteria
Archaeplastida, 255
Arthropods, 24–26, 83, 229, 275, 277–278, 282–284, 287–295
Ascomycetes, 255, 262
Aspens, 70, 228–238, 247
Asynchrony, 134
Atmosphere, 166, 251, 256–257, 260, 263
Autoacatalytic. *See* Catalyst
Autolysis, 109

Baboons, 29–30
Bacteria, 28, 83, 90–91, 101–103, 118, 127, 130, 134–135, 145, 151, 186, 256, 304
  archaebacteria, 26, 93
  bacteriocin, 130
  cyanobacteria, 127, 308
  eobacterium, 127
  eubacteria, 6, 26, 90, 93, 101
  myxobacteria, 177
Bangiophyte, 260, 262
Basichlamys. *See* Alga
Basidiomycete, 255
Benefits, 114, 118–121
  generation of, 41–42, 44, 49,
  of group living, 147–148, 175
  of multicellularity, 142
  of sex, 186
Between-group. *See* Selection
Bilaterians, 12, 166, 200, 260, 262, 264, 272, 277–279, 282–287, 291, 293–295
Biofilms, 131, 151
Biogenesis, 179, 222
Biomarkers, 262, 264
Biomechanics, 36–38
Bivalves, 286, 289
Blattodea, 284
Bootstrapping, 89
Bottlenecks, 71, 75–77, 234, 236, 240–241, 244
Boundaries, 71, 130–131, 138, 203, 215, 221, 233, 307–308
Brachiopods, 278
Brains, 27, 279–280, 286, 303–307
Brassicaceae, 136
Bryozoans, 203, 217, 222, 278
Buffering, 86, 241. *See also* Robustness
Buss, L., 7, 54, 141–143, 164, 200, 221, 237–238

Cairns-Smith, G., 111
Canalization, 88, 95, 276
Cancer, 142, 235, 244–245, 259
Carboniferous, 284
Catalysts, 108, 110–111, 137
  autoacatalytic, 105–110, 114–116, 137, 301
Cells, 3, 9, 11, 26, 28, 54, 71–72, 75–77, 86–90, 97, 102, 108, 116, 143, 169–192, 253, 258, 265,
  artificial, 301
  protocells, 92–94, 97, 116, 128, 137, 304
Cephalochordates, 280
Cephalopods, 286, 308
Cheating, 29, 54, 119–121, 143, 147–159, 234. *See also* Defection; Exploitation
Chimerism, 235–236
Chitons, 282
Chloroplasts, 121, 127, 178–179
Choanoflagellates. *See* Flagellates
Chordates, 277–288, 292–295
Chromosomes, 28, 87, 113, 121, 243
Ciliates, 142, 251–252
Clades, 55, 77, 165, 265
  cladistics, 55, 57
  cladogenesis, 53
Clones, 28, 90, 130, 148, 182–185, 202–204, 228–231, 236–245
Cnidaria, 200, 272, 277–278, 291, 295
Coevolution. *See* Evolution
Coadaptation. *See* Adaptation
Co-option, 44, 49, 158, 164, 166, 177, 191, 284–285
Coregulation. *See* Regulation
Cockroaches, 282, 284–285, 287
Coenocytic, 252, 262
Cognition, 302–303
Colonies, 16, 28, 37, 54, 58, 61, 70–72, 141–142, 176–177, 182–185, 199–203, 252, 265
Communication, 252–253, 265
Communities, 92–93, 116, 129–137, 217–221
Compartmentalization, 9–10, 28, 93, 113, 128, 137, 241, 304
Competition, 75–76, 85, 102, 106, 113–122, 130–138, 227–228, 234, 238, 241–247, 304
Complexity, 1–3, 5, 9–11, 16–17, 26–31, 39–50, 83–86, 105–112, 163–166, 169, 200, 222–223, 228, 264–265, 273, 276, 293
Conflict, 10, 39–41, 44, 61, 121, 141, 170–175, 184–192, 227–228, 243–247
Consciousness, 26–27, 305
Conventionalism, 57, 61
Convergence, 48, 292
Cooperation, 5, 7, 10–11, 38–49, 60–61, 75, 77, 92–93, 97, 105, 114–119, 129, 143–144, 148, 150, 157, 163–164, 170–192, 234, 307–308
  noncooperation, 38, 119, 164, 171 (*see also* Cheating; Defection; Exploitation)
Coordination, 76–77, 86–88, 141, 186, 218–219
Copying, 67, 112, 114, 116, 120–121. *See also* Replication
Coral, 27, 127, 200, 216–221, 230, 236, 277–278, 291, 295
Crick, W., 110, 120
Culture, 10, 29–31, 70, 169–170, 221–222, 302
Cyanobacteria. *See* Bacteria
Cybernetics, 111, 113
Cycads, 240

Darwin, C., 65, 89, 106, 117–119, 199, 274
Darwin, E., 227–228
Darwinian
  environments, 83–98
  individual, 106, 112, 117, 144, 149–150
  neo-Darwinian, 274–276, 294
  populations, 65–79, 238
  space, 67–69
Dawkins, R., 7, 91, 107, 113–119, 122, 149, 234–235, 246
Defection, 35–49, 127, 129, 136–137, 150, 163–164, 171, 174, 178. *See also* Cheating; Exploitation
Demes, 29, 169
Demographic. *See* Fitness
Dennett, D., 5, 47, 143
Deuterostomes, 279–280, 293
Development, 7–8, 11–12, 71, 76–77, 91–92, 94–98, 103, 143–159, 163–166, 173–178, 182, 202, 204, 253, 256, 258–259, 265
  ecodevelopmental, 177
  genes, 275–295
  modularity, 228–229
  plasticity, 85–88, 229, 232
Diatoms, 142, 252
Dictyostelium, 236
Difference maker, 36–38, 40–45, 49–50
Dinoflagellate. *See* Flagellates
Dioecious. *See* Gamete
Diploblasts, 272, 277–278, 295
Diploidy, 187–191
Directionality, 1–3, 6, 20–22, 24, 31, 83
Diversification, 83, 263–264, 271–274, 277, 282, 287, 292–294
Diversity, 1–2, 4, 11, 24, 83–84, 106, 219, 252, 255, 271–273, 294–295
Division of labor, 11, 26, 28, 30, 41–42, 48–59, 55, 61, 71–72, 127–129, 136, 163, 170–173, 179, 182, 215–216
  reproductive, 10, 37, 71–72, 164–165, 243
DNA, 28, 69, 91, 110, 113, 120–122, 187–191, 271
  DNA-world, 93
Dobzhansky, T., 6
Dominant groups, 24–25
Down-regulation. *See* Regulation
Drift, 69
Drosophila, 96, 284, 290–291

Ecdysozoa, 277–278, 284, 308
Echinodermata, 277–282, 292–294

Echiura, 272, 278, 286
Ecodevelopmental. *See* Development
Ecology, 12, 70, 136–138, 150, 154, 157–159, 200, 217, 221, 273
  ecological fusion, 128–129,
  ecological hierarchy, 54–56, 58–60, 200, 219–222
  ecospace, 4, 24
  ecosystems, 2, 129, 137, 219, 271, 276
Economics, 119, 221
Ediacara, 251, 262–264, 271, 277, 289
Eigen, M., 67, 137
Eldredge, N., 6, 53–55
Elegans, 176
Embryos, 86, 262–263, 275, 277, 282–289
  embryogenesis, 217, 232
  embryophytes, 252, 255, 258, 262
Emergence, 109
Endosymbiosis. *See* Symbiosis
Endothermy, 25
Engineering, 10–11, 16, 86, 289–290
Entrenchment, 87–88
Entropy, 109
Enzymes, 28, 109–113, 121, 292
Eobacterium. *See* Bacteria
Epigenetic, 28, 30–31, 152, 217
Epiphytes, 136
Eubacteria. *See* Bacteria
Eukaryote, 6, 9–10, 26, 94, 121–122, 165, 219, 251, 258–265
  eukaryogenesis, 260
Eumetazoan. *See* Metazoa
Eusociality. *See* Sociality
Evolution, 1–12, 19–21, 32, 65–67, 70–71, 73–74, 83–85, 89, 101, 221, 235, 274
  coevolution, 242, 307–308
  macroevolution, 6, 27, 94, 166, 273–276, 292
  microevolution, 3, 5–6, 77, 89, 93–94, 97, 273–276, 294
Evolvability, 15, 17, 67, 73, 83–98, 301, 308
Explanation, 3, 5–7, 15–16, 22–23, 35–49, 89, 95, 106, 111–112, 142–144, 286–287, 292
  byproduct, 75, 79
  lineage, 42, 95, 98, 102
  proximate, 35, 50
Exploitation, 120, 129, 133–134, 136–137, 157. *See also* Cheating; Defection
Extremophiles, 90
Eyes, 95, 275, 289–294

Fairness, 141
Fecundity, 78, 114, 153–156, 179–182, 203, 252–253. *See also* Viability
  group, 146, 149, 152
Feedback, 109, 111, 132, 137, 259–260, 264–265
Fertility, 236–237, 245
Fertilization, 121, 229–230, 245
Fidelity. *See* Copying
Fisher, R., 7
Fission, 11, 147, 149, 163, 182
  group, 210–211
  lineage, 6, 101
  reproductive, 128–129, 137
Fitness, 5, 9–11, 46–48, 65–69, 77–79, 112, 120, 143–144, 163–166, 169–173, 180, 192, 199–207, 231–232, 247, 305
  demographic, 205, 208, 212, 216–218, 221
  group, 149, 185, 213
  inclusive, 118
  landscapes, 68–69, 73–74, 76, 111, 209, 251, 257, 264, 303
  trade-offs, 175–176
Flagellates, 26, 41, 44, 71–72, 163–164, 175, 178–179, 182–192
  choanoflagellates, 177, 255, 258
  dinoflagellates, 251
Flowers, 239, 245
Free-riders, 234–235, 244–246. *See also* Cheating; Exploitation; Defection
Frequency-dependent. *See* Selection

Game theory, 119–120, 129–130, 185–186
Gametes, 76, 187, 191, 237–238
  dioecious, 177
  gametogenesis, 121
Genes, 3, 6–7, 12, 86–97, 112–114, 117–120, 227, 233–234, 253, 255, 275, 292
  alleles, 109, 174, 233–234, 237–238, 303
  gene-reading, 12
  gene-swapping, 105, 116
Genealogy, 53–56, 59
Generality, 8, 23, 35–50, 69–70, 78, 84, 180, 182, 187, 201–202, 282, 306
Genets, 165, 203, 229–233
Genetics, 6, 39, 88, 145, 171, 241, 251, 274, 276, 283–284, 290
Genomes, 7, 87, 90–94, 120–121, 177, 188, 263–264, 275, 307
Genotype, 76, 88, 91, 96–97, 228, 233–238, 242
Germline, 8, 39–40, 75–79, 150–151, 155–159, 164, 229, 232, 236–237, 244–245
Gradualism, 94–95, 274–275, 280, 291–294
Group selection. *See* Selection
Groups, 9, 29–30, 40–41, 44, 48–49, 54, 58, 60, 112, 116, 150, 158, 165, 187, 201, 207–217, 220, 243, 302
  reproduction, 144–149, 173–175, 206
  social, 5, 29, 118, 169, 200
Growth, 11, 70–71, 106, 115, 154–156, 178, 202–203, 216–219, 228, 231, 237–240, 244–245, 258
Gymnosperms, 240

Hamilton, W., 7, 60, 113–114, 117–119
Hemichordata. *See* Chordate
Herbivory, 241–242
Heterogeneity, 227–228, 234–236, 246
  environmental, 96, 183, 221

Heterotrophs, 252, 256–257, 262–264
Heuristics, 5, 8
Hierarchy. *See* Levels
Histocompatibility, 235–236
Hitchhiking, 171, 174
Homeobox, 290, 292
Homeostasis, 25, 86–88
Homeothermy, 26–27
Homogeneity, 183, 227, 234–235, 243, 246–247
  environmental, 96
Homology, 42, 46–48, 278, 287–289
Hormones, 238, 244, 259
Host-pathogen interactions, 5, 121, 133–136
Humans, 2, 10, 15–16, 19–22, 27–32, 221–222
Hydroids, 230, 258
Hydromedusa, 291

Individuality, 3–4, 7–12, 39, 41, 45, 54, 65, 74–77, 97, 141–145, 149–151, 157, 164–165, 169–173, 183–186, 192, 199–206, 217–223, 227–228, 231–234, 243, 246–247, 251
Information, 21, 26–28, 30–31, 38, 90, 108–116, 302
Inheritance, 1, 5, 9, 11–12, 67, 89, 170, 216, 232
  horizontal, 6, 83, 89–90, 92–94, 97, 101, 116, 290
  vertical, 73, 85, 89, 92–94
Innovations, 4, 11–12, 83, 90–97, 112–113, 146, 159, 259, 265, 273, 276, 294–295, 308
Insects, 58, 61, 183, 284, 289–290, 292, 294
Insectivores, 273
Intelligence, 19, 306
Introns, 91
Isogamy, 177

Jablonka, E., 11, 27, 141, 248, 302
Jellyfish, 230, 277–278, 295

K-selection. *See* Selection
Kauffman, S., 109, 111, 303
Kin, 30, 118
Kin selection. *See* Selection

Laws, 37
Lenski, R., 88, 177
Lepidoptera, 290
Levels, 4, 7–8, 89, 127, 169–173, 234, 243
  of organization, 15–17, 26–27, 38, 49, 53, 58–60, 199–202, 220
  of selection, 4, 8, 16, 27–31, 54, 141, 164, 227
Lewontin, R., 8, 65–66, 74, 84, 120, 265
Life history, 75, 78, 144, 150–154, 171, 175, 177–179, 216, 231
Linnaean, 53, 56–57
Lophotrochozoa, 272, 277–278, 282, 284, 286, 288
Lower-level. *See* Levels
Lyell, C., 274

Macrobes, 11, 83, 89, 97, 163
Macroecology. *See* Ecology
Macroevolution. *See* Evolution
Macrofossils, 260, 262
Macromolecule, 26, 110, 112
Maladaptive. *See* Adaptation
Malaria, 37
Mammals, 19–21, 25–26, 302
Maynard-Smith, J., 27–32, 35, 38, 48
Mayr E., 6, 16, 35, 101
Mechanisms, 1–2, 28, 35, 41–47, 67, 101, 292
  conflict suppression, 184, 246–247
  developmental, 94–98, 287
  of inheritance, 11, 30, 74
  and lineage explanations, 42
  microevolutionary, 6, 274–275
  policing, 234, 243
  reproduction, 206–207
Meiosis, 121–122, 187, 189, 243
Melanogaster, 96, 284
Meristems, 165, 229–233, 236–242, 244–248
Metabolism, 26–27, 83, 88, 90, 127, 242, 245, 255–260, 262, 301, 307–308
  metabolism-first, 115 (*see also* Origins of life)
Metamorphosis, 109
Metazoa, 12, 83, 91, 165, 202, 217, 219, 229–230, 242–244, 258, 262–264, 271–273, 275–279, 286–291, 294–295
Methanogenesis, 83, 260
Methylation, 248
Microbes, 11, 83, 89–91, 121, 135–136, 163, 246
Microbiology, 6, 101–102
Microevolution. *See* Evolution
Microfossils, 262
Microtubules, 42, 87, 94
Mimicry, 136
Mitochondria, 9, 121, 127–128, 219, 260, 303–304
Mitosis, 87, 232, 235, 238
MLS1/2. *See* Selection
Models, 4–5, 36, 38–39, 45–46, 92–95, 102, 177
Modularity, 69, 73–74, 86–88, 228–236, 244–246
Mollusca, 271, 278, 286
Morphology, 6, 95, 272–273, 277, 282, 292
  morphogenesis, 30
  morphospace, 5
Multi-level. *See* Selection
Multicellularity, 11, 38–39, 41–49, 54, 70–71, 75, 114, 128, 142–143, 145, 158, 163–166, 170, 175–178, 182–184, 188, 199–202, 251–255, 261–262, 264–266
Multigene. *See* Gene
Mutation, 68, 75–76, 87–88, 95, 115, 150–158, 171–172, 174, 188, 227, 234–242, 244–246
Mutualism, 38, 129, 136, 141
Myxobacteria. *See* Bacteria
Myxozoa, 272

Nectarivores, 273
Neighborhoods, 93–94, 102, 130–138

Nematoda, 277–278, 295
Neo-Darwinian. *See* Darwinian
Neoproterozoic, 263–264
Noise, 20, 88, 153
Non-Darwinian. *See* Darwinian
Nonmutualistic. *See* Mutualism
Nonreproductive. *See* Reproduction
Nonadaptive. *See* Adaptation
Noncooperation. *See* Cooperation
Notochord, 279
Nucleus, 9, 26, 87, 91

Offspring, 17, 65–66, 70, 73, 84–85, 106, 152–153, 201–207, 228, 231
  group, 143–144, 149–150, 210–216
  offspring-size vs. clutch size, 206
  sex ratio, 121
Ontogeny, 76, 229, 239, 265
Ontology, 53, 57, 62
Onycophorans, 278
Oogamy, 177
Ophotrochozoa, 278
Opisthokont, 252, 255
Optimality, 47, 153–156, 179, 219, 303
  optimization, 44, 91, 170, 175
Organelles, 26, 28, 58, 115, 121, 246, 304
Organization, 16–17, 38, 40, 44–50, 71–73, 77, 83, 158, 176, 180, 192, 222, 234, 246–247, 252–255, 264
  developmental, 8, 88
  hierarchical, 53–61, 141, 219–220,
  levels of, 15–17, 26–30, 38, 144–145
  modular, 244
  self-organization, 105, 107–109, 111 114, 116, 122, 137
Organogenesis, 290
Orgel, L., 110–112, 120
Origins of life, 105
Outcrossing, 187–188
Outlaw, 73–74, 244
  anti-outlaw, 17, 73–74, 85
Ovaries, 229, 232
Oxygen, 37, 166, 251, 256–265, 273, 308
  antioxidants, 191
  anoxia, 152
  oxidation, 169, 190–192, 203, 256, 259, 263
  oxygen-binding, 257
  oxygen-replete, 152, 157
  oxygenation, 256–257

Paleobiology, 6
Paleontology, 274, 276, 294
Paleozoic, 24
Panspermia, 105
Parasites, 116, 121, 129, 136–137, 157, 242, 246, 272
Parthenogenesis, 228
Pax, 275, 290–291
Phenotype, 3, 6, 10–12, 17, 30, 42, 47, 65, 68–69, 73–74, 86–92, 96–98, 120, 147, 157, 204–206, 209–211, 214–217, 242, 273, 275, 287–292
Pheromones, 177
Philosophy, 15, 32, 58, 183
Photosynthesis, 4, 83, 127, 169, 303–304, 308
Phylocode, 57
Phylogeny, 4, 6, 16, 53–62, 91, 166, 239–240, 251–255, 262, 264–265, 271–272, 276–278, 280, 282, 284, 286–289, 292
  monophyly, 55–61
  paraphyly, 278
  polyphyly, 55
Plasticity, 17, 49, 85–88, 158, 229, 232, 239, 242
  synaptic, 305
Policing, 54–55, 61, 174–175, 184, 192, 234–235, 243
Polymorphism, 40, 67, 174
Precambrian, 271, 277, 293–294
Predation, 24–25, 129, 136, 251–252, 271, 273, 279
Preformation, 217
Primates, 28–29
Prions, 106
Prokaryotes, 6, 26, 28, 38, 58–59, 84–92, 94, 97, 101–102, 113, 121, 141–142, 163, 187
Propagation, 136, 231–234, 236–239
  propagules, 76–77, 144, 150, 173–174, 182, 188, 210–216, 228
Prosocial, 134. *See also* Altruistic
Proterozoic, 251, 262–264
Protists, 11, 28, 38, 118, 183, 258–260
Protocell. *See* Cell
Protozoa, 26
Proximate. *See* Explanation
Punctuated equilibrium, 6
Punishment, 38

Quasi-independence, 69, 88. *See also* Modularity
Quasi-multicellular. *See* Multicellular
Queen cells, 118
Queller, 10, 55, 58–59, 117, 128, 304

R-selection. *See* Selection
Radiation, 12, 83, 271, 273, 277, 282, 289, 291, 294–295. *See also* Diversification
Randomness, 181, 204–206, 209–210, 212–213, 215–218, 303
Rank-freedom, 16, 53, 56–58, 60–62
Rationality, 120
Recombination, 68, 76, 187–188, 233, 236, 307
Redundancy, 88, 191
Red Queen hypothesis, 202, 242
Regulation, 25–27, 89, 91, 112, 150, 157–158, 178, 253, 255
  down-regulation, 179, 253, 259
  of growth, 244, 258–259
  of social behavior, 148–149
  up-regulation, 253

Relatedness, 37–38, 43, 45–48, 54–55, 114–115, 142, 148, 179
Replication, 11, 28, 66, 94, 109–112, 114–115, 121, 137, 234, 303
Replicators, 5, 9, 17, 53–54, 66, 73–74, 85, 94, 102, 113–116, 200, 302–305, 307–308
Representation, 5, 304–307
Reproduction, 9–11, 66, 70–71, 75–77, 121, 128, 153–155, 163, 175–187, 200–204, 214–216, 228, 233, 236, 238–240, 301
 asexual, 28, 38, 67, 71, 76, 174, 186–192, 215, 233, 236–237, 239, 242, 248
 group, 143–146, 148–149, 173–174, 206–211
 nonreproductive, 28
 vegetative, 165–166, 228–229, 231, 236–237, 239–240
Resemblance, 8, 111
RNA, 28, 54, 137
 -world, 105, 110–112
Robustness, 22, 69, 73, 85–86, 88, 95. *See also* Buffering

Scaffolding, 93
Schlichting, 49, 258–259
Segmentation, 229, 275, 282–288, 291–292
Segregation, 76, 121, 188, 245
Selection, 6–8, 27–31, 35–38, 54, 65–67, 69, 92, 95–98, 110, 112–114, 117–119, 142, 164–165, 169–170, 199, 201–202, 206–210, 218–219, 227, 234–235, 237–247, 274, 302–307
 between-group, 30, 170, 174
 diplontic, 237–241
 frequency-dependent, 44, 120
 group-selection, 10, 30, 60, 113, 148–149, 182, 227
 K- and *r*-selection, 154–156
 kin-selection, 7, 37, 43, 114, 129, 148, 234
 MLS1/2, 78, 143–144, 148–150, 157–159, 164–165, 185, 204
 multi-level, 16, 35, 60, 97, 142–143, 243, 246–247 (*see also* Levels)
 sexual, 46, 87
 viability, 145
 within-group, 30, 170, 174, 184, 187–189, 243, 247
Self-organization. *See* Organization
Self-preservation, 105–108, 112
Selfishness, 105–107, 113–114, 116–122, 148–150, 178–179, 188–189
 self-interest, 29, 234
Sequestration, 40, 159, 236–237, 240
 unsequestered, 165, 230, 232, 236
Sex, 10, 76–77, 97, 169–170, 173, 186–192, 233, 236–237
 ratios, 120–121
 sexual dimorphism, 177
Sober, 46
Sociality, 27, 29–30, 118
 eusociality, 7, 30, 37–38, 55, 141–142
Sociobiology, 54, 105
Soma, 8, 39–42, 71–72, 76, 78, 151, 159, 164, 172–186, 190–192, 232, 235–239, 241–242, 244
Specialization, 10, 28, 41–42, 71, 76–79, 127, 146–147, 163–164, 172–177, 179–186, 217, 257, 271. *See also* Division of labor
Speciation, 37, 46, 294
Sponges, 200, 216–217, 263, 277–278
Spores, 76, 109, 191
Stability, 5, 17, 73, 85, 114, 120, 187–188
 cooperation, 38–41, 45, 48–49, 163
 environment, 91–92
Stebbins, L., 6, 26–27, 32
Streptophyta, 251, 253, 255, 258
Stress, 190–192
Stromatolite, 215, 222
Subversion, 75, 79, 84, 243. *See also* Cheating; Defection; Exploitation
Suicide, 109, 130, 157
Superorganisms, 7, 16, 29–30, 54, 58–59
Symbiosis, 6, 92, 94, 115, 121, 129, 141, 219, 246, 303–304
 endosymbiosis, 127–128
Synergy, 41, 170, 172, 304
Syngamy, 76

Taxonomy, 55–61, 263, 272, 281–282, 294–295
Termites, 12
Trade-offs, 47, 72, 79, 164, 179–182, 206
 fitness, 175–176
 theoretical, 37, 65
 viability-fecundity, 153–154, 158, 201
Transcription, 89–91, 111, 178
Transitions, 1–5, 8–12, 15–17, 19–32, 35–36, 38–42, 45–46, 48–49, 53–56, 58–62, 65–67, 74–75, 77–79, 91–93, 105–108, 110–117, 127–129, 136–138, 141–146, 150–152, 157–159, 164–166, 169–170, 172–173, 185–187, 189–192, 199–204, 206–207, 209, 216–218, 220–222, 265, 301–304, 307–308
 asexual to sexual, 187
 egalitarian vs. fraternal, 10

Unicellularity, 89, 91, 158, 175–179, 228, 251, 253, 258–260, 264–265. *See also* Multicellularity
Uniformitarian, 3, 199, 274–276
Unselfish. *See* Selfish
Unsequestered. *See* Sequestered
Urbilateria, 282
Urchins, 293–294

Variation, 1–2, 5, 7–8, 17, 65–69, 73–76, 83–86, 88, 95–98, 106, 113, 117, 143–144, 149, 158, 164–165, 184, 202, 209, 211, 215–219, 223, 234–235, 238–239, 241–242, 248, 258, 274, 276–277, 293
Vegetative function, 185
Vehicles, 200. *See also* Replicator
Viability, 78, 145–146, 153–154, 173, 175, 179–183, 201–203, 208–212, 252–253. *See also* Fecundity
Virulence, 134, 242

Virus, 5, 106, 134, 186–187
viroids, 26
virus-world, 116. *See also* Origins of life
Volvox, 16, 39, 71–72, 158, 164, 173, 176–180, 182, 184–185, 187, 189, 192, 201–202, 222, 258

Wallace, A. R., 274
Weismann, 232–233
West-Eberhard, M. J., 86
Within-group selection. *See* Selection
Workers, 37, 118, 137, 141–142, 243. *See also* Specialization; Division of labor

Xenoturbella, 272, 280

Yeast, 266

Zooids, 203, 230
Zooxanthellae, 127. *See also* Symbiosis
Zygotes, 171, 189–190, 229, 231, 233, 235