Medicinal Plants

Applied Biology of Domestication and Export

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Introduction

Since the time immemorial, Indian subcontinent has been reputed as the treasure house of valuable Medicinal Plants of the World on account of vast diversity in climatic condition (Padulosi *et.al.*, 2002, Jobhar *et.al.*, 2003). However, population explosion coupled within enviromental degradation has brought about a substantial depletion in the forest and natural vegetation. This has culminated in 'threat' to the very existence of several plant species of therapeutic value. Hence, an alarm has been raised to adopt effective strategies for conserving and further development of biodiversity of medicinal plants. (Fronz, 1993; Brucher, 1985; Schroder, 1998). Basic, application oriented basic and applied aspects of environmental biology of medicinal plants, form the foundation of such strategies (Govil *et.al.*, 2002). These scientific informations are considered of immense importance for domestication, cultivation and improvement of medicinal plants (Singh *et.al.*, 2003, Jakhar *et.al.*, 2003).

The scientific interest in such studies has been recently developed due to the importance of such invetigations in understanding the nature of biological processes involved in plant production. This discipline of the life sciences has also opened new vistas to understand the nature of inter relationship between organisms including plants and their physical, chemical and biotic environments (Larcher, 2002). The influence of environmental factors on the growth of plants has been observed for centuries. However, the interest in the quantification of such influences, at various stages of plant development, is of recent origin (Nilsen and Orcutt, 1996).

Relevance of such observations in agriculture and horticulture has been emphasized repeatedly by several workers. Villareal (1980)

realized that plant productivity in developing countries like India has been much lower than in the developed countries. This is mainly due to the fact that excessive priorities have been given to cereals, very little to fruits and vegetables and practically nil to plants having medicinal values. Views of Sen (1977), Brady (1980) and Coulter (1980) have further supported the aforementioned statements. Kramer (1980) emphasized that physiological processes of plants should be investigated in relation to environmental parameters so as to obtain production oriented results. Importance of such investigation becomes multifold when we consider ecofarming in tropics and sub-tropics where plants are often likely to be subjected to environmental stresses (Alvin and Kozlowski, 1977; Sen and Bansal, 1978; Larcher, 2002).

India's progress in improving traditional crops is a matter of pride for ourselves. However, little attention seems to have been given to medicinal plants which occupy a unique place in Indian socioeconomy (Jakhar *et.al.*, 2003). They advocated to undertake applied and fundamental research work on medicinal plants, several of which may attain the position of important cash crops for Indian farmers in future. With the exception of very few plants which have been brought under commercial cultivation recently, most of the plants used as raw materials in Indian pharmacy, are still predominantly extracted from their wild resources and a continuous extraction of these medicinal plants from Indian forests is depleting with an alarming rate this invaluable wealth of India (Gupta, 1977; Jain, 1979; Jain and Sastry, 1982; Sarin, 2003).

Atal and Kapur (1982) and Swaminathan (1982) have further advocated the need of bringing more species of therapeutically valuable plants under cultivation. For domestication and successful cultivation of medicinal plants, value of environmental biological investigation has been repeatedly emphasized (Kavaljian, 1980; Jain and Sastry, 1982), Krishnan (1980) has mentioned that after the identification of medicinal plants for pharmaceutical industry, the work of ecophysiologist immediately starts. He emphasized that the study of physiological constraints is the first task for medicinal plant domestication programme. According to him, out of several constraints, the studyon causes and cure of low and erratic seed germination, prolonged crop duration, low productivity and difficult propagation are to be dealt on priority basis. Giving the examples of a few medicinal plants (including *Atropa, Rauwolfia, Catharanthus and Papaver*), he emphasized that the study of basic physiological processes in relation to agro-climatic conditions of the locality in question and under

various environmental stresses, should be conducted. The information so obtained will be of immense importance for a plant improvement programme. Similar views have also been expressed by Sahu (1979), Nickell (1982), Parson (1982) and Govil *et.al.*, (2002).

Ashwagandha and red tephrosia enjoy a considerable therapeutic repute in Allopathic (Jain, 1979), Homeopathic (Sanyal, 1982), Unani (Israeli, 1982) and Ayurvedic (Dastur, 1970) systems of medicine. Barring a few areas, these two species are predominantly collected from wild sources for Indian pharmaceutical industry and foreign export trade. Screening of the published literature, personal visit to various organizations including Central Institute of Medicinal and Aromatic Plants (CIMAP), Trade Development Authority of India (TDAI), CSIR, ICAR and scientific discussions with the scientists of National Medicinal Plants Board (NMPB) have clearly revealed that no systematic and scientific studies seem to have been conducted on ecophysiological aspects of these two species. Therefore, investigations reported in these case studies were carried out with following aims and objectives in view :

1. To study the phenological behaviour of plant species in natural habitat of their occurrence.
2. To study the seed germination and early seedling growth responses of plants under environmental stresses.
3. To study the vegetative growth responses of plants to environmental stresses.
4. To study flowering, fruiting, seed setting and yield potential of plants under various environmental stresses.
5. To study the effect of certain plant growth regulators on seed germination, vegetative and reproductive growth and therapeutic yield.
6. To study the most suitable type of soil for plant growth and cultivation and to study the effect of certain macronutrients on plant performance under field conditions.
7. To review the present status of use of these plants in Indian pharmaceutical industry and their export potential.

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Historical Background

Such approaches are considered as the essential part of production oriented crop physiology and have recently attracted the attention of scientists all over the World (Larcher, 2002) Misra (1980) emphasized to study the plant under natural as well as in laboratory conditions as affected by environmental stresses. Many plant biologists emphasized the ecophysiological approaches for studying the frost resistance, drought resistance and resistance to salinity (Asana, 1975; Levitt, 1980, 81; Quisenberry, 1982; Nilsen and Orcutt, 1996). The scientific and systematic study of various plants–physiological processes in relation to environmental conditions, is the main goal of plant environmental biology. Eckardt (1977) has mentioned that such studies form the background for understanding the structure, function and distribution of plants on the earth. Street and Opik (1970) have also mentioned that all the plant physiological processes should be studied in relation to their environmental conditions. According to Larcher (2002) plant ecophysiology is concerned fundamentally with the physiology of plants as modulated by fluctuating external influences.

Plants and their environment form a complex system with the multiplicity of factorial interaction. It is due to the magnitude of fastly interacting external factors affecting the plant life. Larcher (2002) emphasized that it is the task of ecophysiology to study the dynamic interplay of environmental factors, the reaction and adaptation of organisms and the regulatory mechanism underlying these interactions. The advancement in agricultural sciences gave a new impetus to ecophysiological studies in India, specially when the importance of quantification of crop-weather relationship was realized. With the realization of numerous physiological constraints in plant

introduction, domestication, cultivation and production, agricultural scientists and planners started looking towards ecophysiological studies to be carried out in systematic and scientific manner. It has further been cautioned that, as there is considerable amount of complexity in the environmental influences and plant responses, research projects must involve carefully and cautiously constituted long-term planning, most comprehensive recording of observations and also a critical evaluation of data and consequently the most unbiased and genuine interpretation (Larcher, 2002).

It would be worth mentioning here that efforts have been made while reviewing the literature in this treatise to avoid the repetition of what has already appeared in various forms in literature including some text book (Levitt, 1980, 81; Larcher, 2002), proceedings of symposia/seminars, (Landsberg and Cutting, 1977), review articles (Bewley and Black, 1978; Lange *et.al.*, 1981; Koller and Hadas, 1982; Parson, 1982). The literature has been reviewed with particular emphasis on ecophysiological aspects, relevant to present objectives with special reference to non-traditional crop plants, cultivated, semi-cultivated or wild plants, especially those having therapeutical value in modern systems of medicine.

Though, plant ecological researches in India have made considerable advancement (Misra, 1980), ecophysiological studies on plants may be regarded as comparatively new. Levitt (1980, 81) recently mentioned that there is a whole new field in stress physiology waiting for investigation, specially, taking the whole plant system, as experimental material. A brief review of the work done on various aspects of environmental biology of plants in recent past is being given in the following paragraphs.

1. Plant Responses to Environmental Stresses

A. General

Treshow (1970) emphasized that no organism in the world is independent of the environment and every living being constantly influences or is influenced by its organic and inorganic surroundings. It was also documented that environment is a complex of factors and the interaction of factors is so complicated that it is impossible to isolate any single component of factors that does not influence the other. Daubenmire (1976), for the convenience of study of complex environment, subdivided it into units like physical environment and biotic environment. Physical environment contained temperature, light, water, soil and atmosphere. Levitt (1980) classified the stress into two main groups—biotic and abiotic. Infection of plants and competition

with other organisms were called as biotic stresses while physical factors influencing the plants were included under abiotic stresses. All these stresses were studied by many workers using different plant materials including chick pea, lettuce, rice, barley, wheat, corn, cotton and other crops.

Sarin and Gupta (1968) and Subrahmanyam and Murty (1968) have shown the importance of temperature in plant distribution. Rathore (1971) has expressed the role of temperature in plant distribution. Treshow (1970), Levitt (1980) and Larcher (1980) have also shown that the temperature affects the other eco-climatic factors and the physiology of plants is also affected.

Precipitation either in the form of rainfall, snow or dew and atmospheric humidity are closely interrelated with plant growth and development. Water is required for all life processes and often functions as the limiting factor for several ecophysiological plant processes (Mayer *et.al.*, 1974). Adaptability of plants is determined by the availability of water (Misra, 1980). All the physiological processes are influenced by water directly or indirectly (Leopold and Kriedemann, 1975).

Light, which influences the plants by virtue of its intensity, quality and periodicity, plays a vital role in determining the plant characteristics, distribution and survival. The main function of light in ecophysiology of plants is to regulate their physiological process such as photosynthesis, growth and productivity (Smith, 1966). Kaul (1967) studied the significance of light in the life of *Xanthium strumarium L.*, a promising medicinal plant. Nilsen and Orcutt (1996) also obtained similar results in fabaceous plants.

B. Seed Germination and Dormancy in Relation to Environmental Stresses

The scientific information on seed germination are comprehensively available for some field crop plants (Hadas, 1976) but such information on medicinal plants are scanty. Atal and Kapur (1982) have emphasized the need of cultivation of medicinal and aromatic plants but they have mentioned that for such efforts a knowledge of germination behaviour is necessary. This type of study is still inadequate particularly with reference to Indian medicinal plants (Krishnan, 1980). Koch (1977) has reviewed the literature on such aspects and has given some information on seed germination of plants which are weeds but may be exploited as the source of important drugs. Kozlowski (1972) also reviewed the literature on germination

and some other related aspects. Koller (1972) showed the importance of environmental factors controlling seed germination.

Contributions of Dr. D.N. Sen and his team studying seed germination, particularly of Indian desert plants, are worth mention. According to Sen (1977) the quantitative and qualitative nature of responses of seeds with reference to germination under various environmental stresses is highly variable depending upon several factors including age of seeds, viability, dormancy etc. He has further emphasized that a study of ecophysiology of seed germination is of paramount importance in understanding the growth and development of plants. The various factors affecting seed germination and various types of dormancies are important. Before starting the experimental investigations on breaking of dormancy, actual cause of dormancy must be worked out (Sharma and Sen, 1974). Ramakrishnan (1963) studied the nature of the dormancy in some grass seeds. Tripathi (1969) also conducted similar studies in *Asphodelus* sp. Impermeability of seedcoat to water was reported to be the prime cause of seed dormancy in plants (Bewley and Black, 1995).

The involvement of phytochrome in the control of light dependent dormancy in photoblastic seed was reported by Pandey (1965). Natural inhibitors of seed germination were reported by Kaufmann (1975) in seedcoat and embryo of seeds of *Cucurbita*. Such natural germination inhibitors have also been reported by Sharma and Sen (1974) in *Sesamum indicum* and by Bhandari and Sen (1972) in some *Citrullus* sp. A large number of chemicals were used by various workers for breaking seed dormancy (Sen, 1977). Naik (1954) used NAA for gram seed; Chatterji *et.al.*, (1966) applied 2, 4-D, IBA, GA_3 and coumarin for *Sesamum indicum*, while Vyas and Agrawal (1972) tested auxins on several plants.

Toole (1973) reviewed the literature on the action and interaction of light and temperature in influencing seed germination. Quality, quantity and periodicity of light have been reported to influence seed germination. Koller (1972) also studied the effect of light on seed germination. Shamsi and Whitehead (1974, 77) have made a detailed study of effect of temperature on seed germination in species of *Epilobium* and *Lathyrus* and reported that optimum temperature for above mentioned species were 10°C and 20°C respectively, if other conditions were kept constant. Popay and Roberts (1970) studied the effect of temperature on germination percentage in *Capsella* species. Levitt (1980) studied seed germination in relation to high and low temperature treatments in several species of winter annuals. Timson

(1965) in *Polygonum*, Singh (1982) in *Opium* species and Simon *et.al.*, (1976) in cucumber reported a variety of effects of varying temperature on seed germination.

Effects of moisture stress were investigated by several workers on germination. Hadas (1977) made an extensive study of the effects of moisture stress on seed germination in some leguminous plants and reported that the percentage of germination of seeds was declined with increasing moisture stress under field condition as well as in petridishes. Hegarty (1977a) also made similar studies. Singh and Singh (1981a,b; 82a,b; 83a,b), Singh and Afria (1985) and Singh *et.al.*, (1986) have also studied the effect of moisture stress on seed germination percentage in all the three major cereals, some legumes and medicinal plants.

Effect of several growth substances in promoting or inhibiting seed germination of some medicinal plants under normal and stress conditions has been studied by Jones (1969). Gibberellic acid in various concentrations has been shown to be a promoter of seed germination (Krishnamoorthy, 1975). Abscisic acid was shown to inhibit seed germination in a number of plants (Milborrow, 1974a,b). Miller (1956) reported that cytokinins in general promote seed germination. Ethylene has shown variable effects on seed germination process (Abeles, 1973). Amen *et.al.*, (1970) found that low temperature treatment in seeds of *Distichlis spicata* could successfully break the dormancy. Babu and Joshi (1970) found that the dormancy of *Borreria articularis* seed could be broken by gibberellic acid or low temperature treatment or by a prolonged washing of seeds.

Mayber *et.al.*, (1958) reported that GA_3 was capable of overcoming the dormancy induced by high temperature. Popay and Roberts (1970) reported that dormancy of seeds of *Capsella* could be overcome by stratification treatment. Rao and Reddy (1978) found that the seeds of *Indigofera linifolia* could be induced to germination by hot and cold temperature treatments. Villiers (1972) made a detailed study of various aspects of seed dormancy in many wild and cultivated plant species. Ovacharrow (1977) pointed out that in several cases considerable differences are observed in seed germination behaviour under laboratory and field conditions.

C. Seedling Growth in Response to Environmental Stresses

Sinha (1977) pointed out that little is known about the seedling growth of medicinal plants under environmental stresses. Seed germination alone is not important until seedling growth, vigour and establishment are properly studied. Seedling growth considerably

affects the crop stand density and final yield of the resultant crop (Down and Hillemers, 1975).

Hadas (1977) and Atal and Kapur (1982) pointed out that the scientific information on seed germination and subsequenty seedling growth of some crop plants are comprehensively available but such information about medicinal plants are scanty till now. Alvin and Kozlowski (1977) studied the germination and seedling growth of several plants under climatic stresses. Singh and Singh (1981a, b) also studied the nature of seedling growth of several plants under natural as well as stress conditions. Bewley *et.al.*, (2000) stated that the extent to which the seedling growth of the seedlings developed from the seeds treated with various physical and chemical methods is affected by such treatments, has not been studied with adequate attention. In certain cases, these treatments have been reported to influence variably the growth of resultant seedlings. The growth behaviour of seedlings in responses to environmental stresses such as water stress has been studied by Singh and Singh (1981a, b; 82a, b). Similar studies on the seedling growth and other aspects of growth in relation to water stress were also carried by Hsiao *et.al.*, (1976) using different plant materials. Recently, the importance of the study of the level of water stress which critically affects seed germination and the growth of subsequently developed seedlings, has been realized to make a physiological evaluation of drought resistance characters in plants (Hall *et.al.*, 1976; Carlson, 1980). The quantification of effects of the adverse environmental conditions is of considerable importance for raising a successful agricultural crop as they determine the limit of yield and productivity of crops (Gelmond, 1978; Agrawal, 1980).

Levitt (1980) reported that environmental conditions such as water, temperature and radiation stresses affect the seedling growth and seed germination of plants. Taylor *et.al.*, (1982) also studied the germination and seedling growth characteristics of three species of tomato as affected by water deficit and it was shown that shoot growth was affected to a greater extent than the root growth of seedlings by moisture stress. Dutta and Basu (1982) studied the environmental effects on seedling growth in *Cassia sophera*. The study of seedling growth as affected by environmental stresses, has also been undertaken by some other workers (Sen, 1980; Singh and Singh, 1982b). The adverse effects of temperature and radiation stresses on seedling growth were also reported by Bewley and Black (1995).

D. Vegetative Growth in Response to Environmental Stresses

Vegetative growth is the connecting phase between seed

germination - seedling growth and flowering and fruiting stages of the plants. The productivity and economic yield is affected upto a marked extent by the vegetative growth. Some aspects of vegetative growth such as shoot length, root length, number of leaves and leaf area etc. in relation to environmental stresses have been studied to a limited extent in medicinal plants (Atal and Kapur, 1982). These parameters of the vegetative growth produce cumulative effects on productivity of the plant (Leopold and Kriedemann, 1975). The effect of radiation stress on the vegetative growth is mediated through different physiological processes including photosynthesis, respiration and photorespiration (Campbell, 1981).

Temperature affects the physiology of plants by exerting a regulatory role in transpiration, translocation and assimilation (Leopold and Kriedemann, 1975). All these physiological processes ultimately affect the growth and yield of plants in various ways. Such investigations have also been carried out by Bohra and Sen (1976) and Levitt (1980, 81). Lange *et.al.*, (1981) gave a comprehensive account of the growth of plants in response to physical environment. Effect of weather on plant growth and development was quantified by Landsberg (1977) and Eckardt (1977). Dennis *et.al.*, (1970) reported that light inhibits the shoot length and increases the leaf area in straw berry. Root/shoot ratio was found to be greater under water stress in gram (Singh and Afria, 1985). Low temperature treatment in relation to shoot length and root length was studied by McDanial (1982) in *Citrus* species where shoot length was increased under low temperature treatment but the root length was adversely affected.

E. Flowering, Fruiting and Seed Setting in Relation to Environmental Stresses

Flowering, fruiting and seed setting have tremendous importance in determining the plant productivity (Larcher, 2002). Responses, concerning the reproductive capacity of plants have been investigated by some workers on some selected traditional crops (Gupta, 1978). Barton *et.al.*, (1973) emphasized that among various factors which influence the growth and floral development of plants, temperature and diurnal light period are most important. Pathak (1967) reported that seed setting is affected by the moisture, temperature and light stresses in *Tribulus terrestris*. Kaufman (1972) found that the moisture stress highly affects the reproductive stage which can be divided into three phases–flowering, fruit enlargement and ripening. Gupta (1977) reported that water stress decreases the flowering and fruiting both, in terms of number of these reproductive

parts and the percentage of retained reproductive units resulted from enhanced shedding of flowers and fruits and consequently the yield is adversely affected. Chinoy *et.al.*, (1965) expressed the view that flowering stage is a critical phase which is most susceptible to the environmental stresses.

Keeping in view the importance of flowering, fruiting and seed setting, Copper and Hammer (1996) studied the drought stress in relation to growth, development and yield. He reported the detrimental effects of internal moisture stress on photosynthesis and yield. Trivedi and Tripathi (1982) studied the effect of moisture stress in *Spergula arvensis* and *Plantago* species. They found that flowering and fruiting both were adversely affected by moisture stress. Harder *et.al.*, (1982) also obtained similar results in corn. Murty and Murty (1982) studied the effect of light intensity on different stages of growth and reported that the flowering and fruiting in rice (*Oryza sativa*) are increased under low light intensity. Gupta (1978) reported that in barley and wheat the promotion of flowering under low temperature treatment enhanced the yield of plants.

F. Productivity and Economic Yield in Relation to Environmental Stresses

The temperature, water and radiation stresses are generally reported to show an adverse effect on yield and productivity of plants (Levitt, 1981). Copper and Hammer (1996) laid an emphasis on the study of yield or productivity of the plants in relation to drought. Landsberg and Cutting (1977) pointed out that various conditions of environmental stresses influence the plant productivity in different ways.

Biscoe and Gallagher (1977) also revealed that weather is a major variable affecting the crop productivity in advanced agricultural system and advocated to study carefully the effect of weather on productivity. Yates (1968) accepted honestly his failure to evolve consistant relationship between crop yield and weather. Recently, some workers suggested that weather influences the physiological and developmental processes which ultimately determines the yield of the crops (Nosberger *et.al.*, 2001). The effect of climatic factors on the physiological processes which determine crop growth and yield, has also been studied by Biscoe and Gallagher (1977). The biomass and yield under various climates have also been studied by some workers in some medicinal plants including opium, castor, linseed, datura etc. (Singh *et.al.*, 2000). However, plant productivity of herbs of medicinal value as affected by environmental stresses has been

given little attention, particularly under Indian agroclimatic conditions.

The relationship of the economic yield with biological yield which is expressed as coefficient of effectiveness or harvest index was studied in cereal crops (Arnon, 1975; Gupta, 1977). Both the economic as well as biological yields are influenced by environmental stresses (Levitt, 1981). Recently, Singh *et.al.*, (2000) studied the effect of environmental stresses on the dry matter accumulation of some plants of therapeutic importance in India. Larson (1975) revealed that environmental stresses influence the crop yield by affecting the different stages of the plant development (floral initiation, inflorescence and seed formation) and also studied the drought stresses in relation to yield of plants. Singh and Purohit (2000) reported that the productivity of plants is influenced by environmental stresses in a complicated manner.

Kramer (1980) reported that the crop yield is controlled by a complex interaction between the genetic potential of crop plants and environment in which they grow. Variation in the genotype and in the environment including weather and cultural practices, act through physiological processes to control growth. Thus, the physiological processes of plants are the machinery through which environment affects the plant growth quantitatively and qualitatively. Asana and Sarin (1969) have carried out investigations on physiological factors associated with yield. Many aspects of production physiology of plants under marginal conditions have been recently reviewed by Nilsen and Orcutt (1996).

2. Plant Growth Substances In Relation to Plant Growth and Yield

The hormonal control of plant growth and development is most fascinating and fast growing field of plant physiology. Although, few plant hormones, classed as auxins, gibberellins, cytokinins, abscisic acid and ethylene, are well established during the last 30 to 40 years, a detailed study has recently been made on the action and chemistry of plant growth regulators (Krishnamoorthy, 1981). The studies on applied aspects of plant growth substances, specially their application in agriculture and horticulture is fastly gaining momentum due to a great potential of phytohormones in yield improvement (Weaver, 1972; Nickell, 1982).

The literature on the action and biochemistry of the growth regulators is comprehensively available (Milborrow, 1974; Moore, 1980; Leopold, 1982). Some growth regulators are extensively employed for promoting roots in cuttings, inhibiting the sprouting of tubers under

storage, controlling the developmental and maturation processes of fruits and a selective eradication of unwanted plants from cultivated fields. The use of growth regulators has revolutionized the agricultural practices, more particularly in horticulture in advanced countries (Sircar, 1971; Krishnamoorthy, 1981; Nickell, 1982; McLaren, 1982).

The role of gibberellic acid in germination of seeds of various crops including bean, wheat, barley etc. was discussed by Krishnamoorthy (1981). Sen Gupta and Chattopadhyay (1955) studied the effect of IAA, IBA and NAA on seed germination in *Corchorus capsularis, Hibiscus sabdarifolia* and *Crotolaria juncea*. Abscisic acid has been shown to act as dormancy imposing agent which inhibits the seed germination in crop plants (Milborrow, 1974). Ethylene was reported to act as the promoter of seed germination as well as a dormancy breaking agent in pine-apple by Abeles (1973).

Auxin promoted root elongation in lower concentrations and strongly inhibited the same in higher concentrations (Pilet *et.al.*, 1979). Chaudhary and Singh (1960) reported a promotery effect of gibberellic acid and auxin on tomato plant in relation to growth and development. Milborrow (1974) studied the inhibitory effect of abscisic acid in relation to shoot and root growth, leaf area and other aspects of growth in *Citrus* species. The role of ethylene in plant growth and development was reviewed by Abeles (1973). The variable effects of growth regulators on flowering, fruiting and seed setting behaviour of plants have been the subject of some recent reports (Leopold and Kriedemann, 1975; Krishnamoorthy, 1981).

3. Interaction of Growth Substances with Environmental Stresses

Growth and developmental processes in plants are controlled by many factors (both environmental and internal) and their interaction (Leopold and Kriedemann, 1975; Levitt, 1980). Witwer (1971), Arnon (1975), Bruinsma (1982) and Sacher (1982) have shown that the adverse effects of certain abiotic stresses may be reduced or eliminated by careful and timely use of certain plant growth substances. However, this seems to be a new challenge to plant physiologists who should provide field applicable and economic remedies after intensive experimentations (McLaren, 1982; Grierson *et.al.*, 1983).

4. Soil Types and N, P and K in Relation to Plant Growth and Yield

Murthy and Hirekerier (1980) reviewed the literature on soils found in India and point out that these may be categorized into four

major classes on the basis of dominant size of soil particles (clay, silt, sand and loam). Misra (1980) indicated that plant growth and yield are effected by special physical and chemical characteristic of the soil. The behaviour of soils was studied mostly in traditional crops but such effort have not been made in medicinal plants to the adequate extent. Saroha (1981) emphasized a close relationship between plants growth and soil type. Nitrogen, phosphorus and potash are considered most important nutrients out of 16 well established micro and macro nutrients (Lauchii, 1983). The mineral requirements of crops differ from species to species (Larcher, 1980). Atal and Kapur (1982) realized that mineral requirements of medicinal plants are more or less unstudied and only very few reports are available on the subject (Saroha, 1981)

On the basis of brief resume summarized in foregoing discussions and a careful screening of available literature, it may be indicated that a substantial gap of information exists in our understanding environmental biology of medicinal plants of India in general and *T. purpurea* and *W. somnifera* in particular. Such gap certainly hampers the productivity and subsequently the export potential of medicinal plants (Sarin, 2003, Rawat, 2003, Singh *et.al.*, 2003, Jakhar, *et.al.*, 2003). Hence, comprehensive investigations on such aspects are the utmost requirements in national and international R and D efforts. Case studies reported here are part and partial of such efforts.

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Experimental Technology for Applied Environmental Biology of Medicinal Plants

The medicinal value of plants is known since ancient past. Before the Christian era; Riguveda, Athurv veda, Charak Sanghita, Sushrut-Sanghita have given an excellent account of medicinal plants. A number of books on medicinal plants known as herbals were written by various herbalists (Fucus, Cordus, Lobel etc.). India was the leading country in the study of medicinal plant before sixteenth century. *Withania somnifera* L. (Dunal) and *Tephrosia purpurea* L. (Pers) occupy a reputable position in India pharmacology (Atal and Kapur, 1982). Both these species enjoy considerably therapeutic repute in Ayurvedic, Homeopathic, Unani and Allopathic systems of medicine (Bhattacharjee, 2001). All parts of *Withania somnifera* and *Tephrosia purpurea* have great medicinal importance in some ways or the other. Both these plants have been used as experimental system in exploring the applied aspects of environmental biology in present investigations.

Withania somnifera L. (Dunal) is an errect and branching undershrub upto 30 to 150 cm. in height. Stem and branches covered with minor star shaped hairs, nearly all parts more or less stillately tomentose, branches densely tomentose. Leaves petiolate, ovate, subacute, upto 10 cm. long. Flowers bisexual, greenish or lurid yellow, small about 1 cm. long usually 5 flowers borne together in short, axillary clusters in subsessile umbelliform cymes, berries with 6 to 7 mm. diameter enclosed in a much enlarged inflated somewhat 5 angled pubescent calyx, fruit globose, smooth red and seeds yellow scurfy. Root is the most important part with economic point of view. Roots vary in size and shape and light brown in colour. The species

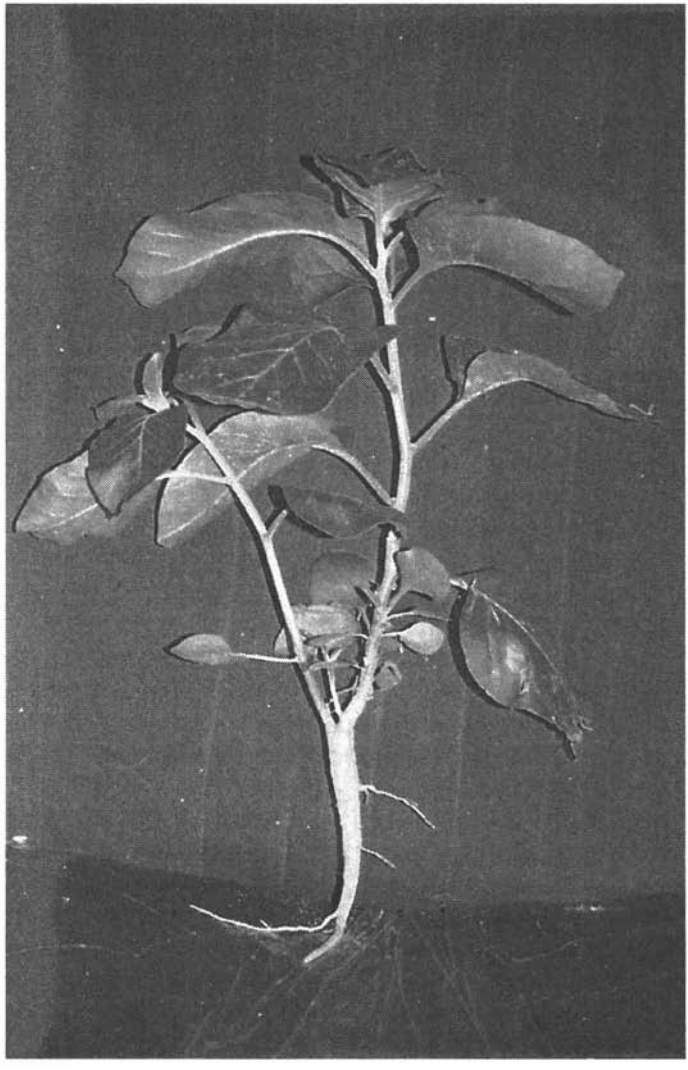
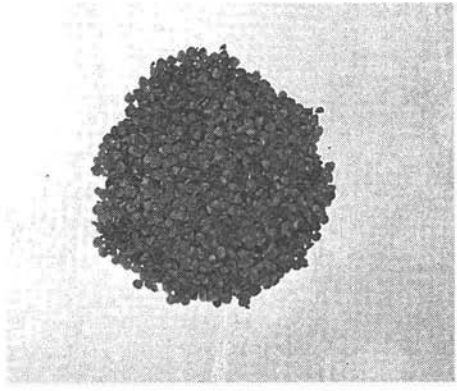
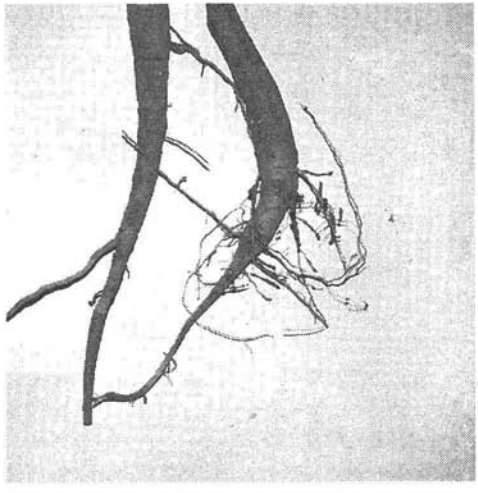


Fig. 3.1 : *Withania somnifera L.* (Dunal)



A. Seed



B. Root

Fig. 3.2 : *Withania somnifera L.* (Dunal)
Econòmic Parts

is found widey distributed in almost all the plain regions of India but the species is more densely concentrated in northern India, specially drier parts. Plant is found wild in hilly region upto an altitude of 1800 mt. The plant is also cultivated upto a limited extent in some parts of southern Rajasthan and northern Madhya Pradesh. However, the bulk of crude drug available in the market is reported to come from parts of Rajasthan including Nagaur, Bikaner and Pilani from the wild resources.

All parts of Ashwagandha have medicinal value. The leaves are anthelmintic and febrifuge. A fomentation of leaves is ued for sore eyes, boils and swollen hands and feet. A paste of leaves is locally applied to kill lice infesting the body and over carbuncles and syphilitic sores, an ointment prepared by boiling the leaves in fat is useful for bed sores and wounds. The leaves contain the alkaloid 'somniferine'. The root is tonic, stimulant alterative, aphrodisiac, narcotic, diuretic abortifacient and deobstruents. It is given in about 300 mg doses in general debility, rheumatic infections, dyspepsia. It is also consumed in loss of appetite, cough and dropsy, 2 gms of the root with milk or ghee is given as an aphrodisiac. 450 mg powder is given twice daily with sugar candy in Leucorrhoea and bloody discharges from uterus etc. Root is very efficacious for toning up the uterus of women who habitually miscarry. For this, decoction of the root is used with long pepper, ghee and honey. In scrofula, chest complaints and cold a decoction is recommended. A paste made of the root and leaves is applied over carbuncles, ulcer and swelling. The paste of the fresh root is used over scrofulous and other glandular swellings. The berries and seeds are diuretic. They are given in chest complaints. Recently the antibiotic and antibacterial activity of the root as well as leaves has been shown experimentally (Ahuja, 1965; Dastur, 1970).

The most important part of the plant is dried roots which are in demand in the international market. The crude drug (*W. somnifera* root) are predominantly collected from wild resources for domestic market as well as export purposes.

Tephrosia purpurea L. (Pers) is a much branched, erect, perennial shurb, glabrous or sparsely hairy, about 180 cm. in height, having an offensive smell. Leaves about 12 cm. long, compound, stipules erect or reflexed, leaflet 9 to 21, glabrous above, covered with silky hairs beneath, nerves conspicuous on both surface. Flowers red or purple in leaf apposed racemes which are about 15 cm. long. Calyx thinly silky, teeth triangular, corolla pubescent on the back. Pods linear,

slightly curved, seeds 5 to 8 in number per pod. Red tephrosia is found in almost all parts of India from hilly region to sourthern India and from Rajasthan to eastern parts of the country. It is cultivated upto a limited extent in southern India as forage crop. In the form of medicinal plant, it is not cultivated in India till now.

Red tephrosia has unique place in medicinal plants of India. All parts of this plant are commonly used as medicine in Ayurvedic system as well as Allopathic system of medicine. Tephroli group of medicines which are used for various disorders of blood forming system of human being are prepared from *Tephrosia purpurea*. Mainly the seeds are utilized for the extraction of active ingredient of the pharmaceutical products (Kapoor and Mitra, 1979).

The plant is tonic, laxative, cardiac blood purifier, deobstruent, diuretic, anthelmintic for children, antidotal, antipyretic and alterative. It is useful in diseases of the liver, spleen, heart and blood, bronchitis, bilious, febrile attacks, gonorrhoea, asthma, tumours, ulcer and piles. A decoction of the dried plant is given in cough, flatulence, diseases of the liver and spleen.

The root is diaphoretic, diuretic, blood purifier and bitter. It is useful in dyspepsia, tympenitis, chronic diarrhoea, bronchitis, asthma, inflammation, boils, pimples and disorder of the liver and spleen. In diarrhoea due to indigestion, a decoction of the root is given with long pepper. The decoction is given with black pepper in gonorrhoea. In fever and vomiting paste of the root made with water and ginger is given with honey. In cough and hiccup, a pill of the size of a gram seed made of the paste of root bark and black pepper is very efficacious as a blood purifier. A decoction of the root is very useful to the children as an anthelmintic. In diseases of the liver, a paste of the root mixed with butter and milk is given. In cough, the dried root is smoked. In hydrocoele, a powder of the root is given with water for about a month in doses of 100 mg to 450 mg.

For the cure of enlarged glands of the neck, a paste of root made with rice water is applied to the enlarged glands.

A powder of the root mixed with honey is applied to wounds. For calculous affections, the juice of the leaves in doses of 50 gm is given with sugar. For the cure of dry eczema, 15 gm of tender leaves macerated in water is taken. An ointment made of the fine powder of the leaves with some blend oil has healing properties. The oil extracted from the seeds is efficacious to local application for eczema.

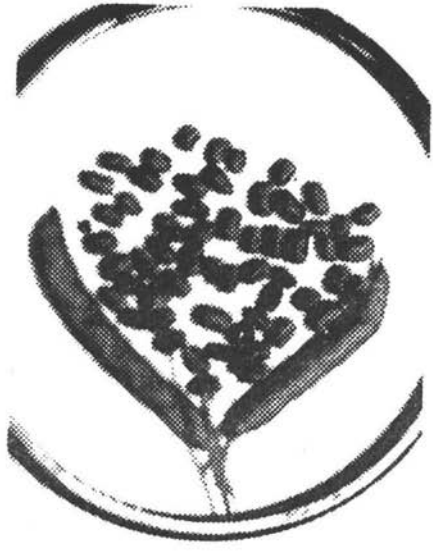
All the experiments reported in these case studies were

A. Plant in Vegetative Phase

B. Flowering and Fruiting Twig

Fig. 3.3 : *Tephrosia purpurea L.* (Pers)

C. Root



D. Economic Part (Seeds)

Fig. 3.4 : *Tephrosia purpurea L.* (Pers)

conducted on the seeds collected from wild plants. For this purpose, 10 sites were selected all around the University. These sites were different for both the species under investigations. On each site five plants showing approximately similar vigour were randomly tagged during preceding flowering and fruiting season for red tephrosia and for Ashwagandha. As these seed source plants were perennial, the ages of these plant could not be determined. Mature pods and berries from these specific plants were collected periodically and brought to laboratory. Sun dried seeds were separated from these fruits and bulked together for whole fruiting season. Now, seeds with uniform shape, size and colour were sorted and stored in glass stoppered bottles under ordinary laboratory conditions. These seeds were used for further investigations. The same procedure was repeated in second year so as to get fresh seeds of the species from the same parental stock and the probable experimental error due to the genetic variability could, thus, be minimized. Growth substances, tested in present investigations were procured from reputed firms, following the method already described in considerable details, (Singh, *et.al.*, 2000).

Standard abbreviations have been used for all the further descriptions.

NAA = α–Nephthalene acetic acid–100% pure
(Arnold Otto Mayer, Germany)

GA = GA_3 : Gibberellin : Gibberellic acid
(F. Holfmann Roche and Co. Ltd., California, U.S.A.)

BA = 6–Benzyl adenine-synthetic cytokinin
(SISCO Research Laboratory, Bombay, India)

CME = Chlorofluorenol methyl ester, a morphactin
(E. Merk A.G. Darmatodt. Germany–as gratis sample)

ETH = Ethrel : Ethephon : (2 chloroethyl phosphonic acid) (CEPA)
(SISCO Research Laboratory, Bombay)

ABA = Abscisic acid, synthetic cis-trans isomer mixture
(Sigma Chemical Co., St. Louis, U.S.A.)

PEG = Polyethylene glycol '6000'
(Willson and Co., Bombay, India)

Other abbreviations used in the text are as below :

CCC = Cycocel (Chlorocholine chloride)

IBA = Indole–3–butyric acid

IPA = Indole propionic acid

TIBA = Tri iodo benzoic acid

2, 4–D= 2, 4–Dichlorophenoxy acetic acid.

Specific experimental procedures adopted for particular aspects of the ecophysiology of plant materials have been described in respective chapters.

Standard terminology used for plant productivity and economic yield is defined below :

1. *Plant productivity* : This is the total dry matter accumulation of the whole plant harvested at maturity stage.
2. *Economic yield* : This includes the dry matter accumulation (dry weight) of economic plant parts. Economic plant part for *W. somnifera* was considered as dry roots at harvest while dried seeds of *T. purpurea* were taken as the economic part.
3. *Therapeutic yield* : This comprised the alkaloid content (mg) per gram dry weight of the dried roots of *W. somnifera* and per two grams of dried seeds of *T. purpurea*.
4. *Pharmaceutical yield* : This is the total alkaloid content (mg) obtained from the dried economic part in respective species.

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Phenology of Indian Medicinal Plants in Relation to Applied Environmental Biology

Phenology is the calender of events in the life history of plants and indicates the times of seedling appearance, vegetative growth, flowering, fruiting and maturity and seed dispersal of the plants under natural habitat (Misra, 1980). He emphasised on the phenological studies of the plant before starting the research work on uncultivated plants. The significance of phenological studies was realized as early as 1932 when Brown and Blanquet published the first text book on plant sociology. They emphasized that the study of periodic and rhythmic changes in the plant life under natural habitat gives the base for germination and other studies about plant cultivation. Oosting (1958) also studied the seasonal development of vegetative and reproductive phases in several plants.

Ambasht (1976) mentioned that biotic and abiotic environments considerably affect the phenology of plants. These qualitative and quantitative influences bring about dynamic changes in the biological events like germination, leaf fall, bud initiation and other reproductive stages. According to Daubenmire (1976) phenological studies also provide a convincing clue to the relative importance of moisture and temperature in different seasons. Keeping in view the importance of the phenological observations which give the basic tool for further research on plants, present study was undertaken to investigate the seasonal events of growth and development i.e. germination and seedling emergence, vegetative growth, flowering, fruiting and seed setting of the plant species.

Experimentia

Ten sites selected for seed collection purpose (Chapter 3) were also marked for phenological aspects of each species. Five plants of each species were tagged keeping adequate distance between plant units. Commencement of various phenological stages was studied following the method of Misra (1980). Seedling emergence in the close vicinity of each plant unit was considered as the initiating step for the study of various subsequent phenological events including vegetative growth (just prior to flower initiation), reproductive growth, seed/fruit dispersal etc.

Experimental Observations

Observations which were conducted for two consecutive years following the similar investigational approach, have revealed some interesting features given below :

(A) Withania somnifera L. (Dunal)

In naturally occurring plants, the initiation of seed germination and seedling growth was observed in late May and was found to be completed upto last week of June. Occasional and unevenly distributed premonsoon showers were noted to be stimulatory for seed germination and seedling growth. On the sites which had very scanty rainfall during this period, seedling emergence was hampered (Table 4.1). The vegetative growth of plants was completed between mid–June and the last week of November. At this time, plants were with well developed leaves and further growth of shoot in length was correlatively reduced because the flower bud development was just started.

First flower bud initiation which resulted in the termination of main shoot growth, was observed in the middle of November in these species. Flower formation commenced in late November. Soon after it, a large number of flowers appeared speedively on the plant resulting in the blooming of plants. Full bloom was observed between December to early March and was completed upto middle of March. Fruit formation was initiated in mid–December and fruiting continued for a long time upto the end of February (Table 4.1).

Fruit ripening was considered as the first stage of senescence. The ripening of fruits started from mid–February and continued upto mid–March (early summer) when all fruits reached maturity in the naturally growing plants. Maturation of seeds was observed upto late March. At this time seed shedding was seen. It completed within two months upto May and withering of leaves occurred in May and June.

All the old leaves completely withered down. Dormancy period in the seeds of Ashwagandha in natural habitat was found for about 11-14 months. The period of onset of almost all the phenological events over lapped frequently in plants growing on each site as well as among plants of different sites.

Table 4.1

Phenological observation on *W. somnifera* and *T. purpurea* in natural habitats

S.No.	Plant growth and developmental stage	Approx. Period	
		W. somnifera	*T. purpurea*
1.	Seed germination and seedling growth	25 May to 25 June	15 March to 10 April
2.	Vegetative growth	15 June to 30 November	1 April to 27 May
3.	Flower initiation	25 November to 5 December	21 May to 29 May
4.	Full bloom	10 December to 15 March	10 June to 15 July
5.	Fruiting initiation	15 December to 10 January	22 June to 14 July
6.	Fruit/pod setting	28 December to 25 February	1 July to 28 August
7.	Fruit/pod maturity	15 February to 15 March	5 August to 15 September
8.	Seed shedding	21 March to 5 May	1 September to 28 August
9.	Dormancy period	11 to 14 Months	4 to 6 Months
10.	Nature	Perennial	Perennial

***B. Tephrosia purpurea* L. (Pers)**

In red tephrosia seed germination and seedling emergence started in mid-March and continued to the first week of April. After 3 to 5 days of seedling emergence the first pair of leaves appeared. Between the first week of April and the last week of May, the vegetative growth of plants was completed. The shoot growth in length was arrested at flower initiation stage which commenced in the last week of May and the full bloom stage of plants was achieved in June to July.

Initiation of fruit formation was observed to commence in the last week of June. The maximum pod setting was recorded in July and August. Pods matured from August to mid–September. Deshiscence and seed sheding processes in the plants occurred in the first week of August to the last week to September. Dormancy period in seeds was found for about 4 to 6 months.

Interpretations and Applications

Though, the plant species under present investigations are quite distant and unrelated phylogenetically (Hutchinson, 1973), they have shown some interesting points of resemblance. Just matured and naturally available seeds showed variable span of dormancy under natural conditions. However, the period of dormancy was much longer in Ashwagandha than in red Tephrosia. The rate of seedling emergence and seed germination was acelerated in both the species by summer downpour. Both the species are perennial but tend to become annual if brought under cultivation. In case of red tephrosia, perennation took place by a dormant rootstock system and some part of the shoot remains viable having quiescent buds. Contrary to *T. purpurea, W. somnifera* retains leaf development activity throughout the year excluding the period of harsh temperature (mid–June to mid–July). When plants are brought under cultivation, the phenological and morphological behaviour of plants are drastically changed. This is specially true for Ashwagandha where such changes are more prevalent and some workers have given the name *Withania ashwagandha* due to certain characteristic features which are quite different from wild species.

In *Tephrosia purpurea* the number of leaflets and their size are increased on cultivation. However, there is little change in the time of commencement of various phenological manifestations in this species. In *Withania somnifera,* the time of flowering and fruiting is markedly reduced when the plant is subjected to cultivation practices (Atal and Schwarting, 1961). The phenological behaviour of *Withania somnifera* is close to its allies viz. *Physalis longifolia, P. pubiscence* and *P. vergeniana* (Anderson, 1968) and some wild species of *Solanum* including *S. corolinense, S. nigrum, S. triflora* (Steinbauer *et.al.,* 1955). Similarly, the phenological behaviour of red tephrosia has shown considerable resemblance with that of some leguminous species including *Alhagi pseudoalhagi, Abrus precatorius, Trifolium indicum* and *Crotolaria juncia* (Rao, 1968).

The differences observed between the phenology of plants naturally growing on study sites as evident from overlapping on

period of commencement of certain phenological manifestations of various sites, may be attributed to the variations in environmental factors at microclimatological level (Landsberg and Cutting, 1977; Mahala, 2002). The effects of environmental factors on phenological events have also been observed by Dikshit (1972) in *Chorchorus* species; Subrahmanyam and Murty (1968) on *Pisum sativum*. Singh and Gopal (1973) also made similar observations on two weeds namely *Anagalis arvensis* and *Chenopodium album*. These workers have further shown that cultural practices have brought about significant alteration in the period of commencement of certain phenological events. Mahala (2002) and Singh *et.al.*, (2003) selecting species of medicinal plants reported that phenology and phenometry are altered on domestication and cultivation including seed setting and production aspects. Such information are applied for collection and preservation of seed material for subsequent uses in experiments and nursery practices.

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Seed Dormancy : Status and Ameliorative Measures

The study of seed germination is fundamental for understanding the growth and development of plants and is an essential basis for the formulation of desirable means of determining the plant producing value of the species (Black *et.al.*, 2000). The mature seed which is made up of an embryo and variable amount of endosperm and the seedcoat or testa, is a compact independent biological entity with the capacity of germinating so as to develop into a new plant under favourable conditions. The successful cultivation of plants largely depends on the quality and germination behaviour of seeds (Nikolaeva, 1969). The mature seeds of some plants do not germinate even though, all the component of favourable environment is supplied to them. Such seeds are known as dormant. The period in which such seeds do not germinate, is called dormancy period (Misra, 1980).

Seed dormancy has ecological implications as the dormant seeds tide over the unfavourable conditions and germinate afterwards when the favourable environment is available (Wareing, 1969). The physiological aspects of various types of dormancy of seeds have been studied by some workers (Nikolaeva, 1969; Black, 2000; Singh and Purohit, 2000; Singh *et.al.*, 2001). Nikolaeva (1969) emphasized that the cause of dormancy must be worked out to study both fundamental and applied aspects of germination of wild as well as cultivated plants, Villiers (1972) used the term dormancy to describe the state of arrested development whereby the organ or organism by virtue of its structure and chemical composition may possess one or more mechanisms preventing its own germination.

Bishnoi (1997) and Mahala (2002) reported that among non-

cultivated arid zone plants, hard seedcoat, impermeable to water, is important cause of dormancy. Nilson and Orcutt (1996) emphasized that breaking of the dormancy and inducing seeds to germinate may be accomplished in a number of plant species by low temperature treatment. Hegarty (1973) reported that temperature treatment plays an important role in breaking dormancy in many plant species. Evenari (1965) studied germination of some species as affected by light treatment. Seed germination in relation to light and phytochrome was reviewed by Bewley and Black (1995).

Breaking the dormancy by chemical and mechanical scarification treatments was reported by Joshi and Nigam (1970). Sen (1977) observed the stimulation of seed germination by chemical scarification in *Trianthema* seeds. Anderson (1968) reported that a number of seeds of other plant species failed to show any response to such treatments. He also reported that a considerable extent of variability exists in the responses of seeds to treatment with inorganic chemicals.

Effects of growth substances in breaking the seed dormancy and stimulating the germination process have been studied by several workers (Weaver, 1972), and a lot of variability in results has been obtained, depending upon the species and concentration of growth substances, period for which treatment was given and stage of seed age (Sen, 1977). The role of growth regulators in breaking the seed dormancy was reviewed by Krishnamoorthy (1981). Abeles (1973) emphasized the importance of ethylene in germination of seeds and reported variable effects. Keeping in view all the facts mentioned above and the importance of such studies, present experiments were undertaken on seeds of medicinal plants which show long periods of dormancy.

Experimentia

Seeds stored following the methods already described (Chapter 3) were subjected to viability test by using standard T Z method (Agrawal, 1980). The test was employed at regular intervals to observe the period for which seeds could remain viable under ordinary laboratory conditions but failed to germinate due to dormancy. Various dormancy breaking measures including water soaking treatment, chemical scarification, hot water treatment, low temperature treatment (stratification) and treatments with various concentrations of selected growth substances, were given to seeds of the experimental species following the method of Anderson (1968) as modified by Shamsi and Whitehead (1974). Experiments for *T. purpurea* were conducted during March-April, months of first year of experimentation with two repetitions and then repeated again in the same months in the second

year. However, experiments on *W. somnifera* were conducted during May-June of first year with two repetitions and then repeated again in same months of following year. Data obtained on germination percentage were analysed biometrically following the methods of Chandel (1985) and Duncun (1955). Data presented in tables and figures are overall averages of two repetitions of experiments.

Experimental Observations

Seed viability persisted in both the species for atleast 3 years under ordinary laboratory conditions. No seed germination was observed in freshly harvested seeds and this condition was observed atleast for 5 months in red tephrosia and for ten months in Ashwagandha. In 6 or 7 months old seeds of *T. purpurea* only 17.5% seeds germinated when they were not subjected to any dormancy breaking treatment. This germination percentage was observed to be almost unchanged on dry storage for subsequent years. In *W. somnifera* only 30 per cent seeds germinated when no dormancy breaking treatment was given. Dormant seeds of experimental species responded variably to different dormancy breaking measures under laboratory trials. Success or failure of various physical and chemical methods to break the dormancy was based on their efficacy to enhance or inhibit the seed germination process of aged seeds. Features of special interest of these treatments have been given below in brief.

(A) Response to Water Soaking Treatment

It was observed that water soaking treatment upto a period of 48 hours enhanced the germination percentage over control in both the species under investigation. In *T. purpurea* as well as *W. somnifera* water soaking treatment for 24 hours proved most effective in enhancing the germination percentage over control. Water soaking for longer period reduced germination percentage significantly in both the species (Table 5.1).

(B) Response to Hot Water Treatment

Hot water treatment improved the germination percentage in both the species. However, this treatment for 30 minutes and above proved to be deteriorative for seed germination in *W. somnifera*. It was further observed that *T. purpurea* seeds responded more favourably than the seeds of *W. somnifera* of this treatment (Fig. 5.1).

(C) Response to Low Temperature Treatment

In this treatment seeds were subjected to low temperature (comparative to room temperature) under moist condition. Therefore, the treatment is designated as stratification treatment. Germination percentage could be promoted by all the degrees of low temperature

Table 5.1

Average and cumulative seed germination percentages of *W. somnifera* and *T. purpurea* under water soaking treatment for different periods

Plants	Treatment (hrs)	Average and cumulative seed germination %					Absolute change over control
		9th day	12th day	15th day	18th day	21st day	
W. somnifera	Control	7.5	13.1	21.9	30.7	30.7	—
	12 hrs.	8.9	18.3	30.2	36.3	36.6	+ 5.6
	24 hrs.	11.5	24.1	37.5	40.5	40.5	+ 9.8
	48 hrs.	12.8	26.3	28.5	36.5	36.5	+ 5.8
	72 hrs.	4.7	10.2	20.2	26.3	26.3	–4.4
					L.S.D.	3.984	
T. purpurea	Control	5.7	7.8	10.2	17.5	17.5	—
	12 hrs.	9.1	11.3	14.5	21.7	21.7	+ 4.2
	24 hrs.	14.3	17.3	21.5	26.9	26.9	+ 9.4
	48 hrs.	11.3	14.5	16.8	18.9	18.9	+ 1.3*
	72 hrs.	3.4	3.9	6.1	7.1	7.1	– 10.4
					L.S.D.	2.956	

Each figure is average of 5 replicates of 20 seeds each.

* Insignificant.

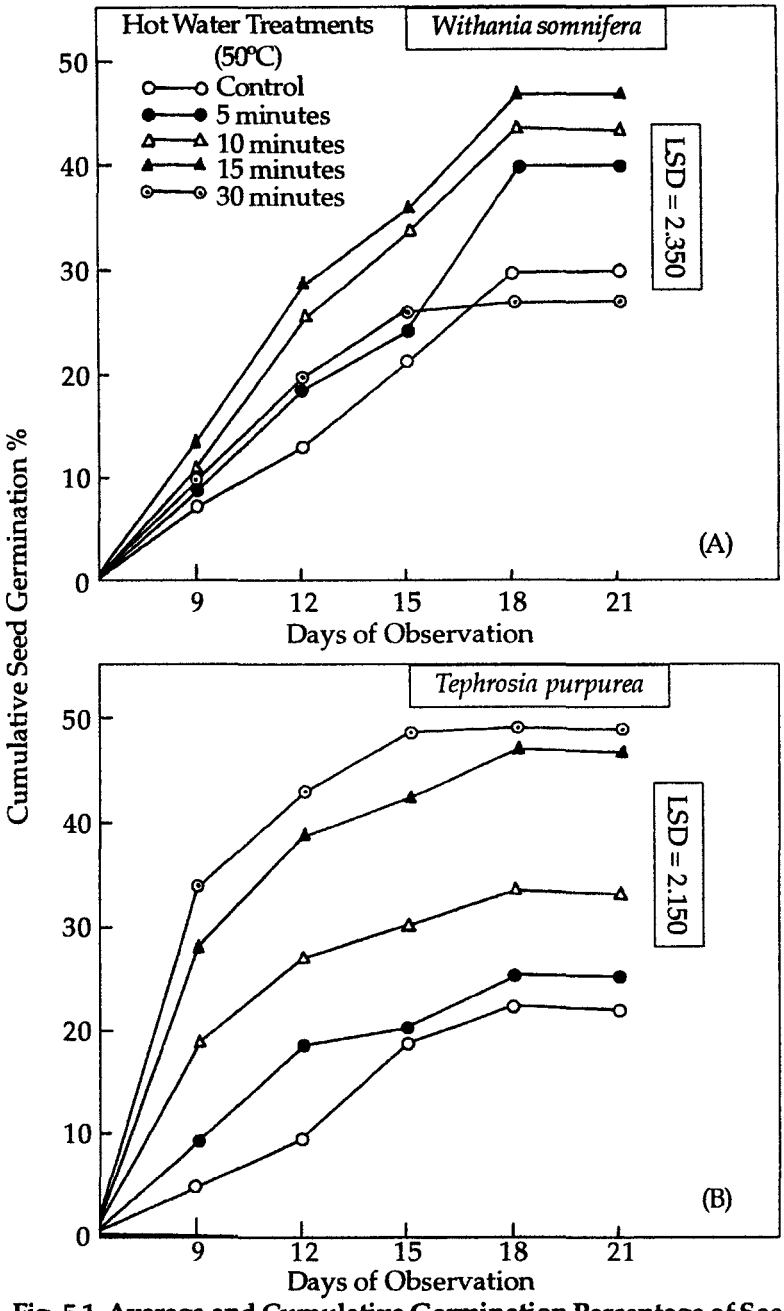

Fig. 5.1. Average and Cumulative Germination Percentage of Seeds of *W. somnifera* and *T. purpurea* under Hot Water Treatment

Table 5.2

Average and cumulative seed germination percentages of *W. somnifera* and *T. purpurea* under low temperature treatment for different periods

Plants	Treatment (°C)	Average and cumulative seed germination %					Absolute change over control
		9th day	12th day	15th day	18th day	21st day	
	Control	7.5	13.1	21.9	30.7	30.7	—
	0°C	—	—	—	—	—	- 30.7
W. somnifera	5°C	9.3	13.9	24.5	37.8	37.8	+ 7.1
	15°C	14.1	24.5	35.7	46.1	46.1	+ 15.4
	20°C	21.4	27.3	39.4	42.3	42.3	+ 11.6
					L.S.D.	4.356	
	Control	5.7	7.8	10.2	17.5	17.5	—
	0°C	—	—	—	—	—	- 17.5
T. purpurea	5°C	9.4	11.3	18.7	26.7	26.7	+ 9.2
	15°C	14.6	19.1	26.7	34.6	34.6	+ 17.1
	20°C	11.3	14.8	19.4	24.9	24.9	+ 7.4
					L.S.D.	5.420	

Each figure is average of 5 replicates of 20 seeds each.
Control = Ambient.

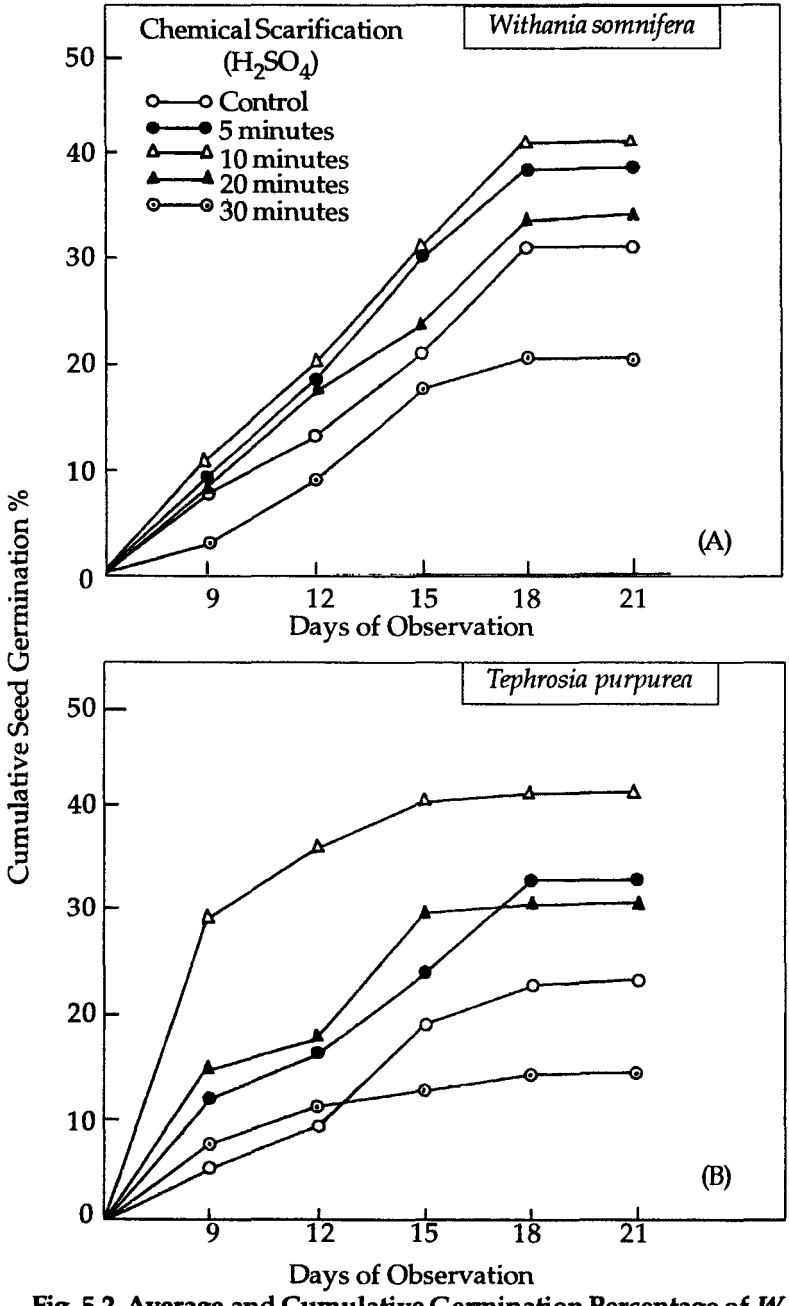

Fig. 5.2. Average and Cumulative Germination Percentage of *W. somnifera* and *T. purpurea* under Chemical Scarification Treatment

tested in both the species. 15°C temperature with moisture was found to be most favourable for enhancing seed germination percentage. There was no significant difference in their response to 15°C temperature treatment at specific level. Temperature lower than 15°C, though, enhanced germination percentage, was found less effective than 15°C. Temperature lower than 5°C did not allow to germinate any seed. However, seeds did not lose their viability (Table 5.2) under 5°C and 0°C temperatures.

(D) Response to Chemical Scarification

It was observed that chemical scarification with concerntrated sulphuric acid for different periods (in minutes) promoted the germination percentage over control in both the species, except 30 minute treatment in *W. somnifera*, whereby seed germination was deferred by this treatment significantly. Seeds of *T. purpurea* responded more favourably to this treatment than those of *W. somnifera*. Ten minute chemical scarification was found to be most suitable in both the species (Fig. 5.2).

(E) Response to Treatments with Growth Substances

Data obtained for these treatments were analysed by Duncun's Multiple Range Test which could bring into light certain interesting features of the effects of various concentrations of growth substances on seed germination percentage in both the test materials (Table 5.3).

(i) Effects of auxin : It was observed that all the tested concentrations of NAA promoted seed germination percentage in *W. somnifera* but the response was statistically insignificant to 10mg/*l* concentration in this species. 100 mg/*l* concentration was recorded to be the most effective and physiologically active with respect to the seed germination in this species and increasing the concentration of synthetic auxin (NAA) beyond 100 mg/*l* could not bring any significant change beyond the level of 100 mg/*l* concentration of NAA.

Contrary to *W. somnifera*, seed germination percentage was drastically reduced by 50, 100 and 200 mg/*l* concentrations of NAA in *T. purpurea*. 10 mg/*l* concentration of NAA could enhance significantly the germination percentage in this species too.

(ii) Effect of gibberellin : Gibberellic acid (GA_3) has stimulated the seed germination process in both the species. However, 10 mg/*l* concentration of this growth substance was found to be most effective in *W. somnifera* while 50 mg/*l* concentration was found to be most effective in *T. purpurea*. 200 mg/*l* concentration of GA_3 has reduced germination over control in *T. purpurea* but the effect was insignificant statistically.

(iii) Effect of cytokinin : In *W. somnifera* 1, 10, 20 mg/*l* concentrations of synthetic cytokinin-benzyl adenine (BA) have shown

Table 5.3

Average and cumulative seed germination percentages of *W. somnifera* and *T. purpurea* under low temperature treatment for different periods

Treatments Conc. (mg/*l*)		*W. somnifera* Average and cumulative seed germination percentages (Days)					Signi-ficance (Duncan)	*T. purpurea* Average and cumulative seed germination percentages (Days)					Signi-ficance (Duncan)
		9th	12th	15th	18th	21st		9th	12th	15th	18th	21st	
Control		7.5	13.1	21.9	30.7	30.7	a	5.7	7.8	10.2	17.5	17.5	a
NAA	10	9.5	15.4	18.3	33.9	33.9	b	8.4	12.6	18.6	29.6	29.6	b
	50	16.7	19.7	26.4	45.4	45.4	c	4.2	6.8	7.9	8.5	8.5	–b
	100	14.3	23.1	25.2	53.9	53.9	d	—	—	—	—	—	–c
	200	8.9	16.3	29.6	51.9	51.9	d	—	—	—	—	—	–c
GA_3	10	5.6	12.4	31.5	43.9	43.9	b	7.1	12.7	38.5	41.9	41.9	c
	50	8.8	14.4	38.1	54.4	54.4	c	19.1	26.5	41.3	48.3	48.3	d
	100	11.2	30.4	51.2	63.4	63.4	d	9.9	13.9	17.6	23.9	23.9	b
	200	9.5	28.3	41.4	56.6	56.6	c	3.5	11.6	15.4	18.3	18.3	a
BA	1	11.6	29.7	38.8	52.6	52.6	b	11.5	21.6	31.3	41.9	41.9	b
	10	17.3	34.5	50.4	57.7	57.7	c	24.3	28.7	38.4	44.3	44.3	b
	20	23.7	29.8	38.4	61.8	61.8	d	28.9	32.8	41.5	50.4	50.4	c
	50	19.6	23.5	37.4	60.6	60.6	d	21.2	33.5	47.4	49.3	49.3	c

Contd...

Contd....

Treatments	Conc. (mg/l)	*W. somnifera* Average and cumulative seed germination percentages (Days) 9th	12th	15th	18th	21st	Significance (Duncan)	*T. purpurea* Average and cumulative seed germination percentages (Days) 9th	12th	15th	18th	21st	Significance (Duncan)
CME	1	9.1	14.6	18.3	36.5	36.5	b	6.5	10.2	18.6	18.6	18.6	a
	10	11.2	18.7	25.4	41.3	41.3	c	17.4	21.4	32.3	44.9	44.9	b
	20	7.3	15.4	21.6	26.4	26.4	–b	5.4	6.9	11.4	13.7	13.7	–b
	50	1.2	9.2	14.9	19.8	19.8	–c	1.1	2.7	4.3	4.8	4.8	–c
ABA	0.1	7.3	11.4	21.5	26.7	26.7	–b	4.9	8.3	11.4	14.6	14.6	–b
	1	3.2	7.1	11.4	16.4	16.4	–c	4.3	5.9	8.6	10.3	10.3	–c
	10	2.4	4.5	7.5	12.5	12.5	–d	1.1	2.1	2.9	3.1	3.1	–d
	20	—	2.1	3.4	7.3	7.3	–e	—	—	—	—	—	
ETH	10	14.5	20.3	30.4	38.6	38.6	b	6.4	9.9	11.7	17.9	17.9	a
	50	18.7	25.3	48.4	55.6	55.6	c	19.6	26.7	31.3	33.4	33.4	b
	200	20.8	39.5	50.4	66.3	66.3	d	26.5	31.3	48.9	65.4	65.4	c
	500	15.3	21.4	23.6	26.5	26.5	–b	1.3	2.7	3.7	4.9	4.9	–b

Significance column represents 'range' according to Duncan's multiple range test.

The same letters for two means in the same column mean not significant to each other at 5% level.

progressively increasing promotory effect on seed germination process but the higher dose has not shown further improvement. Almost similar trend of results was recorded for *T. purpurea*.

(iv) Effect of morphactin : Morphactin (CME) has shown concentration dependent dual behaviour in affecting the seed germination in *W. somnifera* where 1 and 10 mg/l concentrations promoted while 20 and 50 mg/l concentrations reduced the germination percentage over control. In case of *T. purpurea* 10 mg/l concentration promoted significantly the seed germination percentage but higher concentration (50 mg/l) significantly reduced the germination. 50 mg/l concentration of morphactin adversely affected the viability of seeds in both the species.

(v) Effect of abscisic acid : With the exception of 0.1 mg/l concentration of synthetic abscisic acid (ABA) which could reduce germination percentage only insignificantly in *T. purpurea*, all the tested concentrations of abscisic acid have shown progressive effects in reducing the seed germination percentage in the plant materials used in present study. 20 mg/l concentration proved to be highly significant with this point of view. This concentration has shown deteriorative effect to such an extent that germination percentage was reduced to zero in *T. purpurea*, and to 7.3% in *W. somnifera*.

(vi) Effect of ethephon : It was interesting to note that the germination percentage was enhanced progressively and significantly by 10, 50 and 200 mg/l concentrations of ethephon (ETH) in *W. somnifera*. In case of *T. purpurea* 50 and 200 mg/l concentrations have also enhanced germination percentage progressively and significantly. 500 mg/l concentration of ethephon (a potent ethylene releaser) has drastically reduced the germination percentage in both the species which responded upto lesser extent than to ABA treatment with respect to germination process.

Interpretations and Applications

The germination behaviour of wild as well as cultivated plants differs considerably depending on the variety, species, genera, and their genetic make up, certain morphological and physiological characteristics and also on the set of environmental factors which constitute the environmental complex (Gelmond, 1978). In present investigations also, these facts are evident as both the species showed marked differences in their germination percentages when seed treatment was not given. As the viability could be observed atleast for 3 years but almost no germination was observed in seeds immediately after harvest and the same conditions were noted for 6-7 months in *T. purpurea* and 10-11 months in *W. somnifera*, it appears that thereis

primary dormancy in the seeds of both the species. This may be due to immature embryo at the time of harvest. At this time seeds were observed to imbibe sufficient amount of water due to relatively soft and water permeable seedcoat, even then they failed to germinate. The seeds required a period of dry storage under laboratory conditions. However, even after 6 and 11 months, respectively, the untreated seeds could show only poor germination particularly in comparison to seeds of other plants belonging to family Papilionaceae and Solanaceae, respectively. Another interpretation for no germinability immediately after harvest, may be the presence of endogenous seed germination inhibitors which might be responsible for such a constraint in seed germination. Although, the chemical nature of such germination inhibitors could not be established in present plant materials, there are few reports of such inhibitors or secondary metabolites in wild and cultivated Solanaceous plants (Anderson, 1968; Heydecker, 1973; Thompson, 1973; Hawkes *et.al.*, 1979). The species investigated by these workers included *Physalis, Solanum, Nicotiana, Atropa* and *Capsicum,* all belonging to family *Solanaceae.*

According to Black *et.al.*, (2000) dormancy is quite common in wild legumes due to hard seedcoat or due to the presence of germination inhibitors in seedcoat. This type of constraint in seed germination of leguminous plants was reported in groundnut, lentil and some other legumes (Singh *et.al.*, 2003). Improvement in seed germination percentage under water soaking treatment has also been reported by earlier workers (Popay and Roberts, 1970; Thompson, 1970). The promotory effect of water soaking in the present investigations may be attributed to the imbibition of water as stimulated by loosening of hard seedcoat and activation of hormonal action which lead to the enhancement of α-amylase activity and ultimately the germination processes (Ching, 1972; Gelmond, 1978). The inhibitory effect of prolonged water soaking treatment was also reported by Ching (1972) who has shown that prolonged water soaking with distilled water hampers the oxygen absorption which causes hypoxial condition for germinating seeds and ultimately retards the hydrolysis of reserve food substances. Saroha (1981) also reported similar findings in some wild medicinal plants.

Promotory effect of hot water treatment on seed germination was also reported by Anderson (1968) in several solanaceous and leguminous plants. Kauraw and Chakarbarti (1982) with rice cultivars and Thompson (1973) with *Silene* and *Lychnis* also reported similar results. The effect of hot water treatment for a shorter duration (5 minutes) was more prominant in *W. somnifera* than in *T. purpurea.*

Presence of a waxy seedcoat which requires hot water treatment for longer duration so as to make the same loose and permeable to water in *T. purpurea* was also been reported in other minor leguminous seeds including lentils (Singh and Singh, 1982b). In this species it was reported that the waxy seedcoat is loosened due to the partial melting of the seedcoat wax in response to hot water treatment. However, a prolonged hot water treatment always proved to be injurious to embryo leading to germination inhibition.

Anderson (1968) reported variable effects of temperatures below room temperature on seed germination in various species of *Solanum* and *Physalis* and some species of *Madicago* and *Melilotus*. Enhancement of seed germination by treatment with low temperature has also been reported by Khare (1978) with *Verginea indica*. Selecting a few medicinal plants, Saroha (1981) reported that low temperature treatment was considerably effective in enhancing the seed germination percentage. However, it present studies 0°C temperature drastically reduced the germination percentage. This may be attributed to the injurious effect of low temperature (freezing) on seed germination. Levitt (1980) has reported chilling injury of seeds in a number of plants including *Coleus, Solanum, Nicotiana* and *Phaseolus*.

Seed germination response to chemical scarification has indicated that hard seedcoat in *T. purpurea* causes physical resistance to the emergence of radical and thus inhibits seed germination. The enhancement of seed germination by chemical scarification applied for specific periods has also been reported by Ambasht (1963) in *Alhagi* (a close relative of *T. purpurea)*, Chatterji and Mohnot (1968) in *Prosopis*, Vyas and Agrawal (1972) in *Indigofera* and Saroha (1981) in *Argemone mexicana* and *Fumaria indica*. However, chemical scarification for longer duration proved to be injurious for living tissues of the seeds in various cases (Anderson, 1968; Wareing, 1969).

Concentration dependent dual behaviour of synthetic auxin, NAA in affecting seed germination percentage was also reported by Chaudhary and Singh (1960) in tomato and Kumar *et.al.*, (1984) in *Solanum melongena*. The inhibitory effect of higher concentrations of auxin on seed germination was also observed by Randhawa (1971) in some fruit crops. According to him higher concentrations of auxin function as antiauxin and stimulate the biosynthesis of massive endogenous ethylene leading to a suppression of enzyme system related to seed germination. The promotory effect of GA_3 on seed germination was also reported by Randhawa (1971), Chen (1975), Kaufman (1975), Saroha (1981) and Kumar *et.al.*, (1984). The promotory effect of GA_3 is ascribed to its capacity to accelerate α–amylase activity which promotes

hydrolysis of reserve carbohydrates ultimately leading to successful metabolism which is a pre-requisite for seed germination.

Promotory effect of cytokinins on seed germination was also reported by Sankhla and Sankhla (1972) in *Lacutuca sativa* and Saroha (1981) in *Argemone mexicana* and *Funaria indica*. Selecting kidney bean and corn as the experimental materials, Moore (1980) has shown that exogenously applied cytokinins are incorporated in transfer RNA which is specifically involved in the biosynthesis of α–amylase in seed germination.

The promotory effect of morphactin (specially lower concentration) on seed germination is in agreement with the findings of Schneider (1970, 72) and Bopp (1972). According to Schneider (1970) the effect of morphactin on germination processes is similar to that of ABA. The inhibitory effect of higher concentrations of morphactin (CME) in present studies supported the results of Tayal and Gopal (1976) with *Trigonella*. Parups (1983) mentioned that the mechanism of action of morphactin in relation to seed germination is still obscure.

Inhibitory effect of all the tested concentrations of ABA in both the plant materials in present investigation is in agreement with the findings of earlier workers (Addicott and Lyon, 1969; Milborrow, 1974a, b, c). Barendse (1983) reported that the inhibitory action of ABA in seed germination might be related to the suppression of α–amylase activitiy by ABA treatment.

Dual action of ethylene in regulating seed germination process is interpretable in the light of the findings of Katering and Morgan (1969) and Abeles (1973). The influence of ethylene on germinating seeds, specially the dormant seeds, involves a complex interaction of the gas with light, endogenous GA and cytokinin and also ABA. This complexity has forced Lieberman (1979) to accept that no satisfactory explanation is probably available for the mechanism of action of ethylene in a variety of plant growth and developmental processes including the seed germination.

It may be concluded from the above discussion that dormancy in *T. purpurea* as well as *W. somnifera* may be due to more than one factor including hard seedcoat impermeable to water and gaseous exchange and also causing mechanical impedence to embryo, waxy layer in seed coat, presence of some endogenous inhibitor either in seedcoat or endosperm or embryo itself. The chemical nature of these inhibitors (which may be hormonal or alkaloidal in nature) should be further investigated. The findings mentioned above are useful in bringing medicinal plant species under cultivation (Jakhar *et.al.*, 2003).

□□□

Seed Germination Responses under Abiotic Stresses

Seed germination phase of plants is considered critical for the cultivation of plants as it indirectly determines the crop stand density and consequently the yield of resultant crop (Gelmond, 1978). Sen (1977) emphasized that plants are often subjected to environmental stresses, specially under arid climates. Therefore, a critical evaluation and quantification of plant responses to such environmental stresses may be considered a subject of considerable theoretical and applied importance. Several other workers have also realized the importance of such investigations for plant domestication, their successful cultivation and plant production (Levitt, 1980; Lange *et.al.*, 1981, 82; Christiansen and Lewis, 1982; Nilsen and Orcutt, 1996).

Out of environmental stresses, moisture, temperature and radiation stresses are considered most important (Levitt, 1980). The information concerning the germination behaviour of seeds of certain crop plants under moisture stress are comprehensively available but such studies have been rarely conducted on medicinal plants (Hadas, 1976; Hegarty, 1977; Carlson, 1980; Brady, 1980; Singh and Singh, 1981 a, b; 1982 a, b and 1983 a, b, c). This scantiness of experimental data is specially evident for plants belonging to the members of Solanaceae (Hawkes *et.al.*, 1979), and Papilionaceae (Sinha, 1977; Upadhyay *et.al.*, 1981; Summurfield, 1981).

Plant responses to temperature stresses have been a major focus of agronomic (crop ecology) and physiological researches specially due to the importance of such studies for agricultural amelioration of the species. This is mainly due to the fact that temperature is an

equally important environmental stress under arid, semi-arid and tropical zones (Burke *et.al.*, 1976; Levitt, 1980). The effects of low as well as high temperature stresses on potential plant productivity has been a matter of several recent reports (Landsberg and Cutting, 1977; Stanhill, 1977; Bewley, 1979; McDanial, 1982).

The quality, quantity and periodicity of visible radiation is known to regulate a large number of processes of plant metabolism, growth and development (Leopold and Kriedeman, 1975; Blondon *et.al.*, 1977; Levitt, 1980). However, light to the extent of stress adversely affects various plant growth processes including seed germination (Cathey and Campbell, 1982). Christiansen (1982), who has shown that radiation stress may promote or inhibit or (no effect) the seed germination process depending upon the sensitivity of the species of light has used peanut, lettuce, corn, sunflower and vinca for this purpose. Keeping in view all the above mentioned facts, a critical study was conducted to quantify the effect of water, temperature and light stresses on seed germination processes in present plant materials.

Experimentia

Uniform sized seeds of *W. somnifera* and *T. purpurea* were subjected to moisture stress during germination using standard PEG 6000 method details of which have already been described by Singh and Singh (1982 a, b). These experiments were also conducted for two consecutive years in *T. purpurea* and *W. somnifera* as well. Data were analysed by Duncan's method (1955).

The effect of temperature stress was studied by subjecting the seeds (placed in sterilized glas petridishes on Whatman No. 2 filter papers moistoned with double distilled water) to a range of low and high temperatures c.f. room temperature. Seeds were kept under such temperatures for 24 hours under total darkness. For low temperature treatments BOD incubator was used while for high temperature treatments, water bath covered with black cloth was used. The BOD incubator was prestandardized by using a voltage stabilizer having dual range volt guard and thermostat for atleast 72 hours. Water bath was also standardized for 30, 40 and 50°C for 48 hours. Seeds were placed in small sterilized borosilicate glass test tubes having distilled water and medical absorbant cotton plugs. These treatments of various temperatures were given for 24 hours. Seeds kept at room temperature under dark conditions served as 'control'. All these seeds after treatment were brought to ordinary laboratory conditions and germination counts were made daily at 8 A.M. in the morning.

The effect of various levels of radiation stress was studied by

giving the radiation stress (high intensity, extremely low intensity or total dark and light deficit (shade); 120, 0 and 40 lux, respectively) by a careful manipulation of light intensity under laboratory conditions. The Philips fluorescent tube light W.C.L. of 40 W were used to provide high intensity light (120 lux) using the method as described by Cathey and Campbell (1975). The low radiation stress was given to another lot of seeds by keeping the seeds under dark. The light deficit (shade) treatment was also given following the method of Cathey and Campbell (1975).

Experimental Observations

(A) Seed Germination in Response to Moisture Stress

It was observed that decreasing external water potentials (ewp) progressively reduced the germination percentage in both the species (Table 6.1). However, –1.5 bars ewp proved an exception to the above mentioned generalization. This mild stress has very interestingly enhanced the cumulative germination percentage in *W. somnifera*. In *T. purpurea* –1.5 bar ewp could impose only an insignificant constraint on seed germination process. On an average, the seeds of *T. purpurea* proved to be more sensitive than those of *W. somnifera* to moisture stress. The mild moisture stress has also improved the speed (rate) of germination in *W. somnifera* over control.

(B) Seed Germination in Response to Temperature Stress

The low temperature stress in dark for 24 hours stimulated the seed germination process in both the species. 15°C temperature proved to be most effective in enhancing seed germination percentage in both the species. Temperature stress below the range of 5°C caused deleterious effect on seed germination in both the species upto such extent that no seed could germinate at 0°C. *T. purpurea* proved to be slightly more responsive than *W. somnifera* to low temperature stress. The high temperature stress has shown retarding effect on seed germination process in both the species. At 50°C, there was no evidence of seed germination in these species (Table 6.2).

(C) Seed Germination Response to Radiation Stress

Data represented by Fig. 6.1 indicated that high light intensity for 24 hours as well as 48 hours significantly reduced the seed germination percentage in both the species. It was more interesting to note that light deficit and total darkness have shown the stimulatory effect on seed germination.

W. somnifera seeds proved to be more responsive than

T. purpurea seeds to the stimulatory action of low right intensities in present investigation.

Table 6.1

Average and cumulative seed germination percentages of *W. somnifera* and *T. purpurea* in response to moisture stress (different external water potentials e.w.p.)

Treatments Conc. (mg/*l*)	*W. somnifera* Average and cumulative seed germination percentages					Signi-ficance	*T. purpurea* Average and cumulative seed germination percentages					Signi-ficance
	9th day	12th day	15th day	18th day	21st day		9th day	12th day	15th day	18th day	21st day	
Control	7.5	13.1	21.9	30.7	30.7	a	5.7	7.8	10.2	17.5	17.5	a
–1.5 bars	11.8	17.5	26.6	37.5	37.5	b	4.3	8.7	9.7	14.4	14.4	–b
–3.0 bars	5.4	11.3	18.3	21.8	21.8	–b	4.2	5.9	8.2	9.6	9.6	–c
–5.0 bars	—	5.6	10.7	16.3	16.3	–c	1.1	3.6	5.6	6.7	6.7	–d
–7.5 bars	—	—	4.2	6.5	6.5	–d	—	—	—	—	—	–e
–10.0 bars	—	—	—	—	—	–e	—	—	—	—	—	–e

Any two means in one column indexed by same letter are not significantly different from each other at 5% level of significance according to Duncan's multiple range test.

All the figures are average of 5 replicates of 20 seeds each.

Table 6.2

Average and cumulative seed germination percentages of *W. somnifera* and *T. purpurea* in temperature stress (below and above normal room temperature)

Treatments Conc. (mg/l)	*W. somnifera*						*T. purpurea*					
	Average and cumulative seed germination percentages					Signi-ficance	Average and cumulative seed germination percentages					Signi-ficance
	9th day	12th day	15th day	18th day	21st day		9th day	12th day	15th day	18th day	21st day	
Control (Room Temp.) (0°C)	7.5	13.1	21.9	30.7	30.7	a	5.7	7.8	10.2	17.5	17.5	a
0	—	—	—	—	—	–c	—	—	—	—	—	–c
5	9.1	18.6	26.5	37.8	37.8	b	8.1	16.4	22.3	26.7	26.7	b
15	14.4	24.7	31.6	46.1	46.1	d	14.6	24.3	28.8	34.6	34.6	c
20	11.3	21.4	29.8	42.3	42.3	c	16.9	21.4	24.5	24.9	24.9	b
30	6.6	17.5	21.9	34.1	35.1	b	6.8	11.5	18.3	25.7	25.7	b
40	—	—	4.3	11.8	11.8	–b	—	1.7	2.1	3.2	3.2	–b
50	—	—	—	—	—	–c	—	—	—	—	—	–c

Any two means in one column indexed by same letter are not significantly different from each other at 5% level of significance according to Ducan's multiple range test.

All the figures are average of 5 replicates of 20 seeds each.

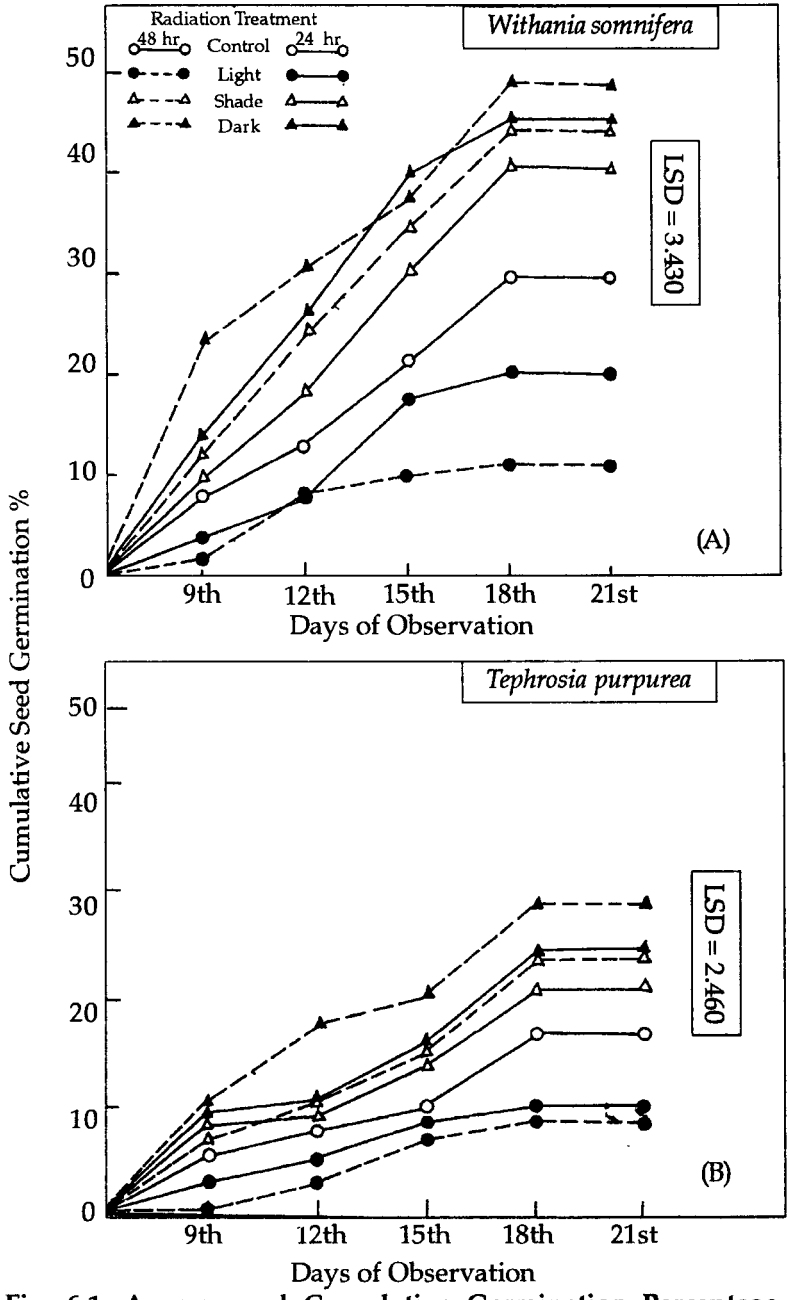

Fig. 6.1. Average and Cumulative Germination Percentage of *W. somnifera* and *T. purpurea* under different radiation stresses.

Interpretations and Applications

A reduction in the seed germination percentage in response to increasing moisture stress, as observed in the present studies, has also been reported by Hadas (1976) in gram and horsebean; cotton and rye; Hegarty (1977) in carrot. Kaufman and Ross (1970) in Lettuce reported that low seed germination percentage under moisture stress was correlated with reduced water uptake. Singh and Singh (1981 a, b; 82 a, b and 83 a, b, c) using similar experimental approach and a variety of plant materials including various composites of maize, different hybrids of wheat, cultivars of rice, macrospermae and microspermae lentils have reported that the suppressive effects of increasing moisture stress was primarily due to reduced mobilization of reserved carbohydrate resulted from suppressed α–amylase activity. Singh and Singh (1983b) further showed that simulated water stress stimulated the accumulation of free proline in germinating wheat seeds and this metabolic manifestation may be used as a good biochemical index of moisture stress tolerance in plants. However, a significant stimulation of seed germination process in one of the species (*W. somnifera*) under present investigation in response to lowest tested order of moisture stress, is difficult to interpret. There are very few reports where a stimulatory effect of mild moisture stress on seed germination has been reported (Singh and Singh, 1995). Hansen (1975) observed that the effectiveness of mild moisture stress in stimulating seed germination in wheat was correlated with accelerated activation of α–amylase by this treatment. The involvement of ethylene biosynthesis and its effect on endogensous gibberellin production which leads to the promotion of α–amylase activity was also reported by Hansen (1975).

The promotory effect of low temperature treatment on seed germination of some tropical plants including legumes and solanaceous species has also been reported by Anderson (1968) who used *Medicago, Melilotus, Alhagi, Solanum* and *Physalis* as the experimental material. He also concluded that a temperature lower than room temperature may be used to break dormancy of some seeds and to stimulate seed germination. Agakischicer (1962) in cotton, Pandey and Sinha (1978) in *Crotolaria* and Kaufman and Ross (1970) in lettuce also obtained similar results. Extremely low temperature (0°C) seemed to cause freezing injury to embryo leading to inhibition of the germination as also reported by Levitt, (1980). The effect of high temperature may also be interpreted in the same light. Kappen (1981) with *Commelina* and *Cladonia* and Chawan (1971) in *Sida* species have shown that high heat treatment is directly related to heat injury.

It was reported that high temperature treatment triggers abnormally high ethylene biosynthesis leading to a phytotoxic effect (Abeles, 1973).

Simon (1974) proposed that under a moderately lower temperature (but not extremely low such as 0°C) germinating seeds imbibed more moisture due to the increased membrane permeability. This permeability increment stimulates the hydration process at a higher rate and consequently hydrolytic processes are increased. However, McDanial (1982) pointed out that a commonly acceptable concept on the physiology of low temperature affecting seed germination process is yet to be experimentally confirmed. Durley (1983) with *Phaseolus* and *Nicotiana* has shown that moderate temperature stress releases bound gibberellins from the cell membrane due to the enhanced membrane permeability and consequently the seed germination process is accelerated.

The effect of radiation stress on seed germination depends upon the sensitivity of the seeds. In present investigation, the seeds of both the species have shown considerable sensitivity to high as well as low light stresses. The suppressive effect of high light intensities on seed germination was also observed by Abeles and Lonski (1969) in lettuce seeds. Curtis (1967) with *Sorghum* showed that high intensity of light stimulated excessive ethylene production by germinating seeds and this ethylene acts as anti-gibberellin. Abeles (1973) and Lieberman (1979) have shown that in light sensitive seeds, the effect of radiation stresses on seed germination is mediated through phytochrome system. Under higher radiation stress, Pr form of phytochrome is generated and thus regulates the biosynthesis of atleast two phytohormones viz. gibberellin and ethylene. Gibberellin remains in bound form and ethylene is released, therefore, germination processes is inhibited. Contrary to the above, in low radiation stress, Pfr form is generated. By this gibberellin is transformed from bound to free form and this form of gibberellin masks the inhibitory effect of stress ethylene and consequently the germination is promoted under dark or shade conditions. Cathey and Campbell (1982), Morgan and Smith (1981) have also given similar interpretation. On the basis of above discussion, this may be inferred that none of the species in present studies, was found to be resistant to moisture, temperature and radiation stresses, atleast with respect to seed germination process under laboratory conditions. These scientific information may be used for modulating the therapeutic indices of medicinal plants of Solanaceae and Fabaceae families.

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Seedling Growth of Medicinal Plants under Environmental Stresses

The seedling growth phase of plants is contributory for raising a successful crop because seed germination alone would not provide crop stand density unless seedling growth and establishment are promptly followed (Singh, *et.al.*, 2000). The scientific information on seedling growth are not satisfactorily available specially under the condition of environmental stresses (Singh and Singh, 1995). Such information on non-traditional crops are much more scanty (Mahala, 2002; Cooper and Hammer, 1996) emphasized to study seedling growth under various environmental conditions to understand the growth, development and adaptability of plants. Several other workers have also realized the importance of such investigations (Levitt, 1980; Christiansen and Lewis, 1982). The quantitative and qualitative behaviour of seedling growth under different climatic conditions markedly differs and environmental stress greatly affects this parameter of growth (Levitt, 1980, 81).

Alvin and Kozlowski (1977) reviewed the literature on the seedling growth of some crop plants under different agroclimatic conditions. Singh and Kakralya (2003) reviewed the nature of seedling growth of several plants under natural and stress conditions. The seedling response to moisture stress in certain plants has been studied by earlier workers (Hadas, 1976; Hegarty, 1977; Bewley, 1979; Singh and Singh, 1981 a, b; 1982 a, b). The work done on solanaceous as well as papilionaceous medicinal plants is specially inadequate (Hawks *et.al.*, 1979; Brady, 1980; Saxena, 1981; Summerfield, 1981). The temperature stress is recognized as most widely operative and

limiting factor for crop physiology and directly affects the seedling growth of plants in various ways and ultimately influences the crop production in various agroclimatic conditions. Several agronomists have, therefore, emphasized the importance of study of temperature stress in relation to crop physiology (Burke *et.al.*, 1976; Levitt, 1980, 81), low as well as high temperature both affect the seedling growth as reported by some workers in few traditional crops (Landsburg and Cutting, 1977; Bewley and Black, 1995).

The radiation stress has been given little attention, specially in the comparison of moisture and temperature stresses in affecting the crop physiology. The quantity and quality of light influence the seedling growth by regulating various aspects of processes of plant metabolism, growth and development (Leopold and Kriedeman, 1975; Levitt, 1980). The radiation stress has been reported to affect adversely the seed germination and seedling both (Cathey and Campbell, 1975). Christiansen (1982) has shown that radiation stress may enhance or retard the length of seedlings in lettuce and cauliflower. Due to the scantiness of such information in non-traditional crops, specially the medicinal plants, present investigation was carried out to make a critical appraisal of the influence of certain environmental stresses on seedling growth.

Experimentia

Seedling growth response of *W. somnifera* and *T. purpurea* to moisture stress (in terms of various external water potentials) were investigated following the method of Singh and Singh (1982 a, b) with a little modification which included the germination of seeds in sterilized Petridishes on Whatman No. 2 filter paper moistened with distilled water. Seeds with approximately 2 mm radicle length were carefully blotted dry and placed on sterilized Whatman No. 2 filter paper in another group of corning glass Petridishes. The filter paper in these Petridishes were moistened with PEG, '6000' osmoticum solutions of various external water potentials (ewp) at regular intervals. The shoot length and root length of seedlings were measured on 12th day after subjecting the seedlings of osmotic stress with PEG 6000. Seedling treated with distilled water at regular intervals served as control. Each treatment consisted of 5 replicates of 5 seedlings each.

The experiment on the effect of temperature stress on seedling growth were conducted following the method of Dolan (1972) with the modification that controlled temperature growth chamber was replaced by BOD incubator wherein various temperatures were

maintained. The day light was substituted by a cool fluorescent Philips tube light in each chamber of the incubator. Seedlings kept on ordinary room temperature with natural photoperiod served as control. All the temperature treatments were given for 24 hours only and then these treated seedlings were shifted to ordinary laboratory conditions in glass Petridishes having distilled water to avoid moisture stress. Data on shoot and root lengths were taken on 12th day.

For radiation stress studies, seeds were first germinated in glass Petridishes on Whatman No. 2 filter paper duly moistened with distilled water. Seedlings with uniform radicle lengths (about 2 mm) were transferred to another group of Petridishes having moistened filter paper. Seedlings in these Petridishes were subjected to various light intensities broadly categorized as high light intensity (120 lux), shade (median intensity–40 lux) and dark (low intensity—0.1 lux). All the treatments of light intensities were given only once either for 24 or 48 hours and seedling growth was recorded on 12th day. Arrangements for light, shade and dark treatments were made following the method of Cathey and Campbell (1975) as described in Chapter VI with little modifications. For this purpose, the laboratory space was portioned into three chambers. The first chamber was used to provide about 120.8 lux light intensity with the help of Philips 40 W fluorescent (WCL) tubes. This intensity was maintained on the surface of the wooden table having the growing seedlings arranged in glass Petridishes. The second chamber was used to give light intensity (41.2 lux) comparable with shade. The third chamber was maintained as dark room (light intensity about 0.0 lux) with low light stress. The slow moving ceiling fans were used to cool down the room chambers. Seedlings (in Petridishes with moistened filter papers) which received usual light intensity and photoperiod prevailing in well ventilated laboratory space served as 'control'. Seedlings which had been subjected to various light stresses for 24 and 48 hours were also shifted to the same space where control seedlings were placed ordinary laboratory conditions.

All these experiments were repeated four times (two times in first year and two times in final year of experimentation) similar results. The data being discussed here, were obtained from experiments conducted during March-April months for *T. purpurea* and May-June months for *W. somnifera*.

Experimental Observations

Analysis of data for the seedling growth depicted following features of interest.

(A) Seedling Growth in Response to Moisture Stress

It was observed that shoot length was adversely affected by increasing moisture stress (decreasing external water potentials) in both the species excepting the mild stress which promoted the shoot length significantly in *W. somnifera* only while the same external water potential decreased the length of shoot non-significantly in *T. purpurea*. The root growth in length in both the species has shown dual behaviour in response to increasing moisture stress. Upto –5.0 bars in *W. somnifera* and under –1.5 bars in *T. purpurea*, a significant enhancement of the root length was observed. All the other tested osmotic potentials decreased the root length significantly except –3.0 bar external water potential which could decrease the root length of *T. purpurea* only insignificantly. The moisture stress of highest order (–10.0 bars) proved to be most detrimental to seedling growth of *T. purpurea* where a cent per cent reduction of shoot and root lengths was recorded (Fig. 7.1).

(B) Seedling Growth in Response to Radiation Stress

The high light intensity given to the seedlings for 24 hours as well as 48 hours has shown adverse effect on shoot and root growth of both the species. Shoot and root of seedlings have shown almost identical responses quantitatively to this radiation stress treatment. However, the shade and dark both have shown a promotory effect on shoot and root growth in length. In *W. somnifera* shoot and root have shown similar responses to low radiation stress but in *T. purpurea* the shoot growth was promoted to a greater extent than the root growth. Shade and dark treatments for longer duration (48 hours) have given more favourable results in both the species under present investigations. *W. somnifera* and *T. purpurea* did not differ significantly from each other with reference to their responses to radiation stresses of different orders (Table 7.1).

(C) Seedling Growth in Response to Temperature Stress

Treatment of temperature stress has shown the dual action in controlling the shoot and root lengths. Low temperature treatment promoted the shoot and root length in both the species significantly excluding low temperature stress of 5°C on root length of *W. somnifera* which was decreased insignificantly. Below 5°C, seedlings could not be obtained due to the failure of germination of seeds at this temperature (Fig. 7.2). The higher range of temperature retarded the shoot and root lengths in both the species significantly. The 30°C temperature treatment proved to be an exception to this generalization in *W somnifera* where this temperature has shown a promotory effect

Table 7.1
Average seedling growth (shoot and root lengths) under various radiation stresses in *W. somnifera* and *T. purpurea*

Treatments	Period (hrs)	*W. somnifera*				*T. purpurea*			
		Shoot		Root		Shoot		Root	
		L (cm.)	ACL	L (cm.)	ACL	L (cm.)	ACL	L (cm.)	ACL
Control		4.5	—	5.2	—	5.3	—	6.4	—
High light intensity	24	3.1	–1.4	3.7	–1.5	4.1	–1.2	5.3	–1.1
(120 lux approx.)	48	2.8	–1.7	3.1	–2.1	3.4	–1.9	4.9	–1.6
Low light intensity	24	5.6	+ 1.1	6.6	+ 1.4	6.7	+ 1.4	7.9	+ 1.5
(40 lux approx.)	48	6.7	+ 2.2	7.3	+ 2.1	7.4	+ 2.1	7.5	+ 1.1
Low light intensity	24	8.9	+ 4.4	7.4	+ 2.2	7.8	+ 2.5	8.8	+ 2.4
(0 lux approx.)	48	9.9	+ 5.4	9.2	+ 4.0	9.8	+ 4.5	8.9	+ 2.5
L.S.D.			0.890		1.250		0.950		0.987

All figures are significant over control.

All the figures are average of 5 replicates of 5 seedlings each.

ACL = Absolute change in length; L = Length.

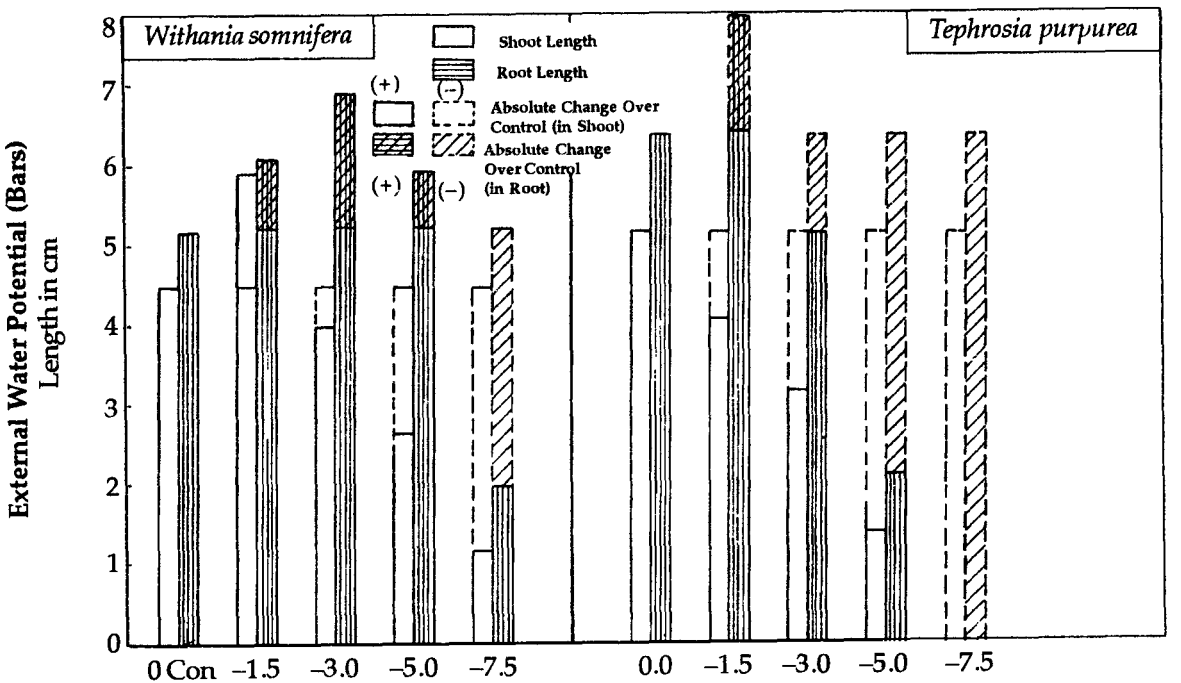

Fig. 7.1. Average Shoot and Root Length of 12 Days Old Seedlings of *W. somnifera* and *T. purpurea* which were subjected to different water potentials.

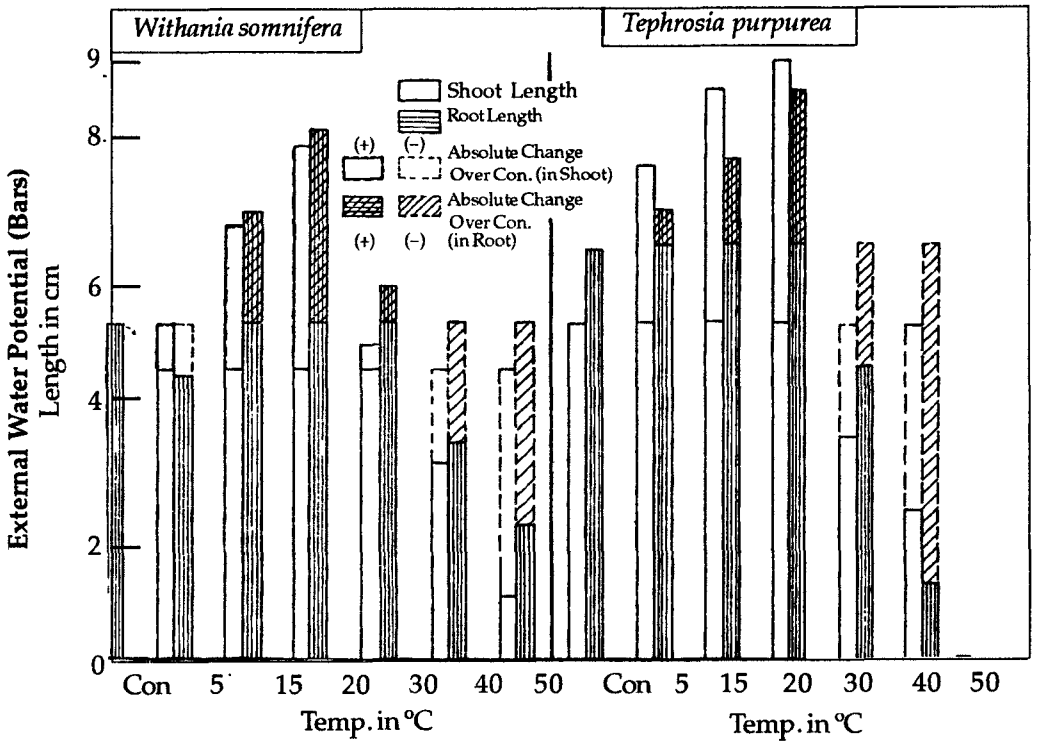

Fig. 7.2. Average Shoot and Root Length of 12 days old seedlings and *W. somnifera* and *T. purpurea* which were subjected to low and high temp. stress for 24 hours.

on shoot and root growth. The roots of both the species were found to be more responsive than shoots to the high temperature stress in general.

Interpretations and Applications

The retardation of seedling growth of plants by moisture stress under laboratory, pot and field conditions has also been observed by Parmer and Moore (1966) in corn and Lawlor (1969) in kidney bean. Recently, Singh and Singh (1981 a, b; 82 a, b; 83 a, b, c) selecting hybrids of wheat, composites of maize and macrospermae as well as microspermae lentils, have also shown a retarding effect of different external water potentials on seedling growth. Singh and Singh (1981a) have shown that the adverse effect of moisture stress on seedling growth may be attributed to the tendency of this treatment to suppress the α–amylase activity in germinating seeds and growing seedlings. This stimulates a reduction of mobilization of a reserve carbohydrates which are, otherwise, required for successful metabolism, an integral part of growth. Selecting several varieties and species of tomatoes as experimental plant materials, Taylor *et.al.*, (1982) made a comprehensive study on the effect of different external water potentials (PEG '6000') on seedling growth in laboratory conditions. They have shown that –4 to –8 bars external water potentials progressively retarded the shoot and root lengths of seedlings under laboratory conditions. Michel and Kaufman (1973) with some leguminous plants and Taylor *et.al.*, (1982) with tomato species also reported that mild moisture stress caused a slight promotion of seedling growth. Recently, Singh and Afria (1985) in macrosperma and microsperma varieties of gram also observed a promotory effect of moisture stress of lower order on seedling growth. This promotory effect may be attributed to the capacity of mild moisture stress to synthesize appropriate amount of ethylene which ultimately stimulates germination and seedling growth in lower concentrations. This action of ethylene is considered to be operative through the release of gibberellins from bound form to free form (Low, 1975). Singh and Singh (1995) and Singh *et.al.*, (2002) using several varieties of arid legumes as experimental material, showed that mild moisture stress may be employed to harvest beneficial responses in terms of seed and seedling conditioning. Mahala (2002) also obtained similar results in medicinal plants of Indian desert.

Results of effect of high radiation stress on seedling growth in present plant materials are in agreement to the findings of certain earlier workers (Cathey and Campbell, 1975). Penning *et.al.*, (1974)

using the similar experimental approach as employed for present investigation, have reported that high intensity of light given for a specific period, inhibited the growth of shoot and root of the seedlings of several varieties of *Phaseolus vulgaris, Arachis hypogea and Zea mays.* They attributed this inhibitory effect of light to the lowering of rate of certain biochemical processes which are otherwise, essential for normal seedling growth. Among these processes suppression of certain enzymes related to carbohydrate and protein metabolism were shown to be primarily affected by high light intensities (Cooper and McDonald, 1970; Penning *et.al.*, 1974). Curtis (1967) using sorghum seedlings as experimental system reported that the inhibitory effect of high light intensity on seedling growth was mediated through excessive ethylene production which stimulates the reduction of seedling growth when synthesized excessively under high radiation stress. Chung *et.al.*, (1981) also observed the promotory of low light intensities on seedling growth of *Dendrobium monele.* Cathey and Campbell (1975) with lettuce, corn flower and vinca have also reported a promotory effect of dark and very low light intensities on hypocotyl length. These results are similar to those obtained in present investigation. However, Cathey and Campbell (1982) have pointed out that seed germination and seedling growth responses to various light intensities are highly variable depending upon the species, cultivar, environmental conditions and also the conditions of the climate during preceding fruiting and seed development period of the parent plants. In this context, it is important to point out that seed germination responses of present plant materials are similar to the seedling growth responses of the same to various radiation stresses. This can be interpreted on the basis of observations of Cathey and Campbell (1982) who have shown that regardless of the requirements for seed germination very similar systems (enzymatic as well as phytochrome) were operative in controlling the growth of the seedlings developed from the germinating seeds. These systems are reported to continue to function atleast upto the stage when secondary leaves start their own photosynthesis and the seedlings attain autotrophy. Cooper and Hammer (1996); Nilsen and Orcutt (1996) and Nosberger *et.al.*, (2001) suggested to employ this technique (seedling response evaluation) for screening the germplasm of cultivated and wild plants for abiotic stress tolerance.

The promotory effect of the temperature lower than the room temperature as observed in present investigation, is explanable on the basis of the findings of some earlier workers, Pollock (1962) with bean, Christiansen (1978) with cotton and Young *et.al.*, (1981) with

Kochia, Dutta and Basu (1982) with *Cassia* also obtained similar results. However, temperature below 5°C caused a low temperature stress forseed germination as well seedling growth. The deleterious effect of such low temperature stress on seed germination and seedling growth has also been observed by Taylor *et.al.*, (1972) with cotton, Blacklow (1972) with maize and Wiesner and Grape (1972) with rye grass. The promotory effect of the temperature below the room temperature on early seedling growth is attributable to the accelerated synthesis of certain enzymes at these temperatures. Leopold and Kriedemann (1975), selecting corn seedlings, have shown that optimum temperature for these enzyme systems varies from variety to variety. However, temperature below a critical level (viz. 5°C or below in present studies) caused a low temperature stress in their investigations too. Gilman *et.al.*, (1973) made a comprehensive study of the effect of high temperature stress on seedling growth in 12 cultivars of soybean. It was shown that temperature above 25°C given for a longer duration was inhibitory for seedling growth. This finding is supported by the results of present investigation on *Withania somnifera* as well as *Tephrosia purpurea*. A mild stimulatory effect of 30°C (marginally above the room temperature) is difficult to explain Johnson *et.al.*, (1974) and Berry and Raison (1981) observed that the temperature optima for seedling growth differs from species to species under ordinary conditions. In present studies, this optima was observed at 32°C with reference to seed germination and seedling growth of *W. somnifera* while for *T. purpurea* temperature optima was recorded at 25°C. This indicated that high temperature stress tolerance was marginally higher in the solanaceous species as compared to leguminous medicinal plant. McDenial (1982) with cotton and certain other crop plants reported that high temperature stress was closely related to water status of germinating seeds and growing seedlings. Bishnoi (1997), Mahala (2002) and Singh *et.al.*, (2003) also reported the applicability such responses for germplasm evaluation and conservation and scientific exploitation of biodiversity of medicinal plants.

□□□

Vegetative Growth under Abiotic Stresses

The vegetative growth phase in which the plants undergo an exponential increase in size without which flowering process can not be readily induced, determines the economic and biological yields of plants culminating in crop production (Leopold and Kriedeman, 1975). The vegetative growth is influenced by environmental stresses in various ways (Biscoe and Gallagher, 1977; Levitt, 1980, 81). Many crop physiologists and agronomists have realized the importance of observing the environmental stresses influencing the vegetative stage (Landsberg and Cutting, 1977; Gupta, 1975, 84; Christiansen and Lewis, 1982; Bidinger *et.al.*, 1996). Among the environmental stresses, water, radition and temperature are most influensive, widely operative and limiting factors for vegetative growth of crop plants (Evans, 1975). Brady (1980) emphasized that such investigations are integral part of production oriented environmental biology.

Evans (1973) in maize and sugarcane and Saroha (1981) in *Fumaria arvensis* studied the effect of light on vegetative growth of plants and also advocated to undertake further researches on plants of different economic classes including medicinal plants. Palmer (1973) in maize, Hermellin (1973) in barley and Martin (1973) in wheat also studied the effect of temperature on vegetative growth of the crop plants. The study of effects of water stress on vegetative growth of plants have been undertaken by Slayter (1973) in cotton, Gupta (1975) in wheat and maize and Landsberg and Cutting (1977) in tomato, potato and certain other field crops and obtained some interesting results. However, very few reports are available on the influences of components of environmental stress on various parameters of

vegetative growth in medicinal plants (Atal and Kapur, 1982). Gupta (1980) and Singh *et.al.*, (2000) reported that in certain medicinal plants where vegetative organs constitute the economic part of the herb, such investigations may provide basic but valuable information for undertaking a medicinal plant cultivation programme. Swaminathan (1982) also emphasized the need of such investigations. Keeping in view the inadequacy of information on such aspects, present studies were conducted to observe the vegetative growth responses of two selected medicinal plants to certain environmental stresses.

Experimentia

Healthy seedlings (15 days old) were selected from seed beds and transplanted to the earthen pots containing normal garden soil. These pots were arranged in a randomized block design pattern for *W. somnifera* as well as *T. purpurea*. Five seedlings were transplanted in each pot meant for stimulating the moisture stress. Ten pots containing five seedling transplants each, constituted a single replicate and each treatment block consisted of five replicates. Watering was done on the intervals 3rd, 4th, 5th, 6th and 7th day in equal amount in each block. After 45 days of transplantation, five plants from each replicate were randomly selected for observation purpose. Data on shoot and root lengths were taken and leaf area per leaf and per plant for *W. somnifera* was determined by the formula given by Pearce *et.al.*, (1975). The formula is as follows :

$$\text{Area} = (L \times W) \times (0.75)$$

where L = Maximum length

W = Maximum width

The area obtained by this formula is for individual leaf on the plant. The total leaf area per plant was determined by summing up the area of all leaves of each plant. However, due to the compound nature of leaves of *T. purpurea* the traditional graphic method was employed to find out the leaf area in this species.

The seeds of experimental species were first germinated in small enamel trays having a layer of normal garden soil and adequate moisture. These trays after seedling emergence (15th day) were kept in BOD incubators maintained at different temperature *i.e.* 5, 15, 20°C (low temperature) and 30, 40, 50°C (high temperature). These temperature treatments were given only for 12 hours. The seedlings which were kept under ordinary laboratory condition served as 'control'. Out of these treated seedlings, seedlings of uniform size were transplanted in earthen pots having normal garden soil with identical composition. Each pot was having 10 seedlings in the

beginning but only four were retained and others were thinned out. Pots were arranged in randomized block design. Plants in pots were subjected to standard agronomic cultural practices and were harvested at 45 days after transplantation. Five plants from each replicate were randomly selected, soil particles were carefully washed away and observations for shoot and root lengths and leaf area were taken following the usual method as described above.

Plants used for the study of water stress influencing the vegetative growth, were protected from precipitation by covering the experimental space with heavy duty transparent polythene cover. However, those used for temperature and radiation stress studies were kept in open space and no care of precipitation was taken in these cases.

The low and high radiation stress was given at seedling stage (15 days old) for 24 hours in both the species. Healthy seedlings were subjected to radiation stresses by the method described in Chapter 7. These experiments were also conducted in two consecutive years for and *T. purpurea W. somnifera*. Seedlings treated with radiation stresses were also transplanted in earthen pots where these were subjected to ordinary conditions. 50 days old plants were harvested and brought to laboratory for further observations on shoot, root and leaf growth.

Experimental Observations

(A) Vegetative Growth in Response to Moisture Stress

The shoot and root lengths were progressively retarded by all the conditions of moisture stress in both the species except only mild moisture stress which enhanced the shoot and root lengths in *W. somnifera* only (Table 8.1). Data indicated that roots are less susceptible to moisture stress than shoots in both the species. The averaged total leaf area was also adversely affected by moisture stress in both the species excepting the lower stress. The total dry matter production as well as the fresh weight of whole plant were also similarly influenced by moisture stress. The repressive effect of moisture stress on vegetative growth was quite significant statistically except the effect of moisture stress simulated by watering plants on 5th day intervals. In this case, total dry matter accumulation could be influenced only insignificantly in *W. somnifera*. On an average *W. somnifera* was adversely affected to the less extent than *T. purpurea* with reference to vegetative growth by simulated moisture stress.

(B) Vegetative Growth in Response to Temperature Stress

The temperature stress, though was given to seedlings only for 12 hours, has shown dual and cumulative action in affecting the

Table 8.1

Vegetative growth responses to moisture stress treatments in *W. somnifera* and *T. purpurea*

Treatments (days)	Shoot growth				Root growth				Leaf area (cm^2)	Total FW (gm)	Total DW (gm)
	L (cm)	ALC	FW (gm)	DW (gm)	L (cm)	ACL	FW (gm)	DW (gm)			
Control (3rd)	38.9	—	5.600	1.680	22.6	—	3.300	1.650	384	11.940	4.010
4th	47.6	+ 8.7	6.410	1.970	26.4	+ 3.8	3.650	1.720*	463	13.750	4.540
5th	27.6	–11.3	3.520	1.160	24.8	+ 2.2*	3.120	1.210	313	10.020	3.587*
6th	19.4	–19.5	2.050	0.730	18.5	–4.1	1.910	0.750	244	5.850	1.730
7th	9.5	–29.4	1.320	0.450	9.2	–13.4	0.950	0.475	136	3.150	1.210
L.S.D.	3.450			0.215		2.960		0.325	41.5		0.428
Control (3rd)	45.4	—	8.500	3.400	29.8	—	2.100	1.050	274	15.900	6.040
4th	39.04	– 5.4	7.410	2.980	34.6	+ 4.8	2.420	1.250	213	14.050	5.390
5th	30.60	–14.8	3.820	1.320	21.4	– 8.4	1.450	0.750	176	8.650	2.940
6th	11.70	–23.7	2.560	1.110	10.4	–19.4	0.980	0.460	94	5.280	2.100
7th	—	–45.4	—	—	—	–29.8	—	—	—	—	—
L.S.D.	3.758			0.415		3.140		0.170	35.7		0.378

Each figure is secondary average of five replicates.

* Insignificant at 5% level of significance.

L = Length; ACL = Absolute change in length; FW = Fresh weight; DW = Dry weight.

vegetative growth of both the species. The low temperature stress stimulated by 5, 15, 20 and 30°C enhanced significantly the shoot and root lengths and leaf area in *W. somnifera*. These low thermal stresses also increased dry matter production in *W. somnifera*. However, the higher temperature treatment (40 and 50°C) showed adverse effects significantly on all the parameters of vegetative growth of *W. somnifera* (Table 8.2). In *T. purpurea* 5, 15 and 20°C temperature stresses enhanced the shoot length but 5 and 15°C temperatures reduced average root length in this species. The root length of *T. purpurea* was slightly promoted by the temperature stress stimulated by 20°C and 30°C temperatures. The shoot as well as root lengths were progressively retarded by 40 and 50°C temperature stresses in this species. The effect of low as well as high temperature stresses on fresh weight, dry weight, leaf area and total dry matter production was similar to that observed in *W. somnifera* (Table 8.2). On an average *T. purpurea* was adversely affected by temperature stress to a greater extent than *W. somnifera* with respect to vegetative growth.

(c) Vegetative Growth in Response to Radiation Stress

Radiation stress cumulatively influenced the vegetative growth in variable manner. High light intensity adversely affected the shoot and root length in both the species while under same intensity of light the leaf area was increased but dry weight of root was reduced in both the species (Fig. 8.1). The total dry matter production was reduced in both the species by high radiation stress but the mild intensity of light has shown favourable effects on vegetative growth excluding leaf area which was not enhanced by this treatment. The similar results were obtained in both the species. The low radiation stress adversely affected the vegetative growth including root and shoot growth, leaf area and dry matter production. Detrimental effect of high radiation stress (high light intensity) proved more pronounced than the adverse affect of low radiation stress in both the species with respects to shoot and root growth in length.

Interpretations and Applications

The simulation of moisture stress of different orders by withholding the water supply for different periods to the plants grown under phytotron, green house, earthen pots or field conditions has been a traditional method for the investigation of effects of moisture stress on vegetative and reproductive growth (Clark and Hiler, 1973). Several other workers have also used the method for similar purpose with appropriate modifications. In present investigation, though, the exact matric potential caused by withholding the water supply for 3, 4, 5, 6 days could not be measured, there are clear evidences that moisture stress of different orders could be stimulated. According to

Table 8.2
Vegetative growth responses to various temperature stress (low and high) treatments in *W. somnifera* and *T. purpurea*

Treatments (°C)	Shoot growth				Root growth				Leaf area (cm^2)	Total FW (gm)	Total DW (gm)
	L (cm)	ALC	FW (gm)	DW (gm)	L (cm)	ACL	FW (gm)	DW (gm)			
Control	38.9	—	5.600	1.680	22.6	—	3.300	1.650	384	11.940	4.010
5	41.5	+ 2.6	6.400	1.980	24.8	+ 2.2	2.720	1.360	426	12.340	4.195
15	53.4	+ 14.5	8.810	2.760	26.8	+ 4.2	3.430	1.740	484	15.850	5.170
20	47.6	+ 8.7	7.640	2.320	31.4	+ 8.8	3.810	1.950	525	15.450	4.950
30	40.2	+ 1.3	5.860	1.760*	28.6	+ 6.0	2.830	1.420	346	12.050	4.700
40	21.3	–17.6	3.540	1.080	11.6	–11.0	1.420	0.750	216	6.550	2.130
50	12.5	–26.4	2.450	0.750	8.8	–13.8	0.910	0.460	114	4.100	1.310
L.S.D.		2.250		0.254		1.980		0.108	32.4		0.180
Control	45.4	—	8.500	3.400	29.8	—	2.100	1.050	274	15.900	6.040
5	47.6	+ 2.2	8.910	3.580	21.4	–8.4	1.420	0.680	314	16.550	6.110*
15	51.4	+ 6.0	9.530	3.820	26.3	–3.5	1.850	0.890	346	17.850	6.440
20	57.4	+12.4	10.760	4.310	34.6	4.8	2.650	1.240	375	20.210	7.350
30	42.3	–3.1	8.150	3.620	30.5	0.7*	2.300	1.150*	306	16.120	5.870
40	21.5	–23.9	4.550	1.840	18.1	–11.7	1.600	0.760	146	7.930	3.370
50	—	–45.4	—	—	—	29.8	—	—	—	—	—
L.S.D.		1.950		0.136		2.140		0.180	29.8		0.125

Each figure is secondary average of five replicates. * Insignificant at 5% level of significance.
L = Length; ACL = Absolute change in length; FW = Fresh weight; DW = Dry weight.

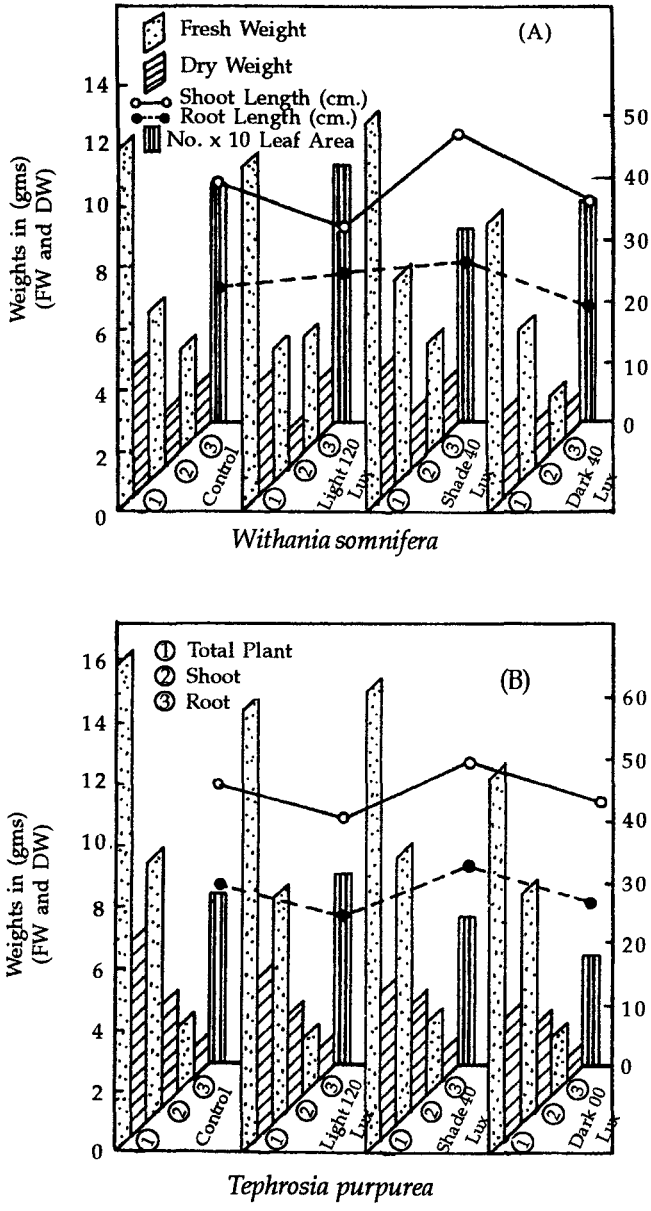

Fig. 8.1. Vegetative Growth Responses to Various Radiation Stress Treatment in *W. somnifera* and *T. purpurea*

Clark and Hiler (1973) plant responses to moisture stress are themselves good indicators of water deficit in soil and plant tissue. The retardation of shoot and root growth by moisture stress was also observed by Harris (1973) in cotton, Townsend and McGinnies (1973) in *Astragalus*, Slayter (1973) in wheat, maize and some millets, Johnson *et.al.*, (1974) in barley and Sen *et.al.*, (1979) in some arid zone weeds. Boyer (1976) in sunflower, Johnson *et.al.*, (1974)in wheat and barley, McGree and Davis (1974) in sorghum and Coulter (1980) in rice also reported the adverse effect of water stress on total leaf area/plant. Coulter (1980) selected several cultivars of paddy and reported that water stress in the soil beyond a critical level retarded considerably and progressively the shoot and root growth and leaf area and number. Bradford and Hsiao (1982) reported that the growth retarding effect of moisture stress is certainly mediated through a reduction of rates of vital physiological processes including the uptake of water and minerals by root, adverse effect on cell wall permeability of the root hairs, inhibition of protein synthesis and imbalance in the synthesis and transport of plant growth hormones including auxin, gibberellin, cytokinin, ethylene and abscisic acid.

However, it was interesting to note in present investigations that a mild water stress enhanced the vegetative growth. These results are in agreement to the findings of Vartapetain (1973) with white mustard and Milburn (1979) with tomato and brinjal. Milburn (1979) noted that it is difficult to explain this abnormal behaviour of plants under mild moisture stress. Findings of Abroni and Richmond (1978), Lange *et.al.*, (1982) also supported the views expressed by Milburn (1979).

It is to worth mention that temperature stress (either low or high) given for a limited period of 12 hours to seedlings of early age was sufficient to stimulate a growth promotion or retardation in subsequently growing seedlings/plants. The promotory effect of low temperature stress (but not below 5°C) on vegetative growth in present studies supported the findings of earlier workers (Dolan, 1972; Koller 1972; Guerrero and Williams, 1975). Reduction of vegetative growth by high temperature was also reported by Townsend and McGinnies (1972) in *Astragalus*, Goscho *et.al.*, (1973) in sugarcane, McGree and Davis (1974) in sorghum and Bjorkman *et.al.*, (1974) in *Atriplex*. Bjorkman *et.al.*, (1974) concluded that adverse effect of high temperature on vegetative growth were attributable to a reduction in relative growth rate, photosynthetic activity and low CO_2 uptake. They reported that environmental conditions prevailing at the time of seedling development influence not only the seedling growth but also the vegetative growth of subsequently developed plant. Bierhuizen (1973) with tobacco and lentil and Palmer (1973) with maize have also observed that the cumulative and long lasting effects of environmental factors were reflected not only into subsequent vegetative

growth but also continued upto flowering, fruiting and seed setting stages. And in certain cases the growth and reproductive behaviour of plants (second generation) developed from so formed seeds was found to be influenced by the previous generation treatments (Pulgar and Laude, 1974).

Selecting seedlings of different cultivars of *Rumex obtusifolius*. McLaren and Smith (1978) reported that high light intensity given by fluorescent tube to seedlings, stimulated a tremendous reduction of stem and root growth of resultant plants. Adverse effect of high light intensity on vegetative growth was also been reported by Gabe and Black (1979) in cucumber, Morgan and Smith (1979) in *Chenopodium*, Loveys (1979) in tomato. Loveys (1979) used various cultivars of tomato (3 species of *Lycopersicon*) and concluded that the detrimental effect of high radiation stress on vegetative growth varied considerably depending upon the age of seedlings at the time of treatment. Morgan and Smith (1981) indicated that the effect of high intensity radiation on seedling growth and subsequent developments of plants are certainly modulated by a phytochrome system functioning under red-far-red interaction but some indirect effects which are operative through water and temperature stresses can not be overlooked. The promotory effects of moderate radiation stress (shade) on growth of plants developed from treated seedlings has also been reported by some earlier workers such as Downs *et.al.*, (1957) with *Phaseolus vulgaris, Helianthus annus* and *Impomoea hydrodea*. Cathey and Campbell (1982) observed that under dark conditions, even for a single cyclic treatment (24 hours), the shoot and root lengths were abnormally increased but the growth of primary leaves was retarded. Durley (1983) and Kaufman (1983) interpreted such observations on the basis of gibberellin dominated hormonal interactions. It has been pointed out that under natural conditions, gibberellins are synthesized in germinating seeds and growing seedlings but when such seedlings are subjected to absolute dark for specific period, gibberellins are rapidly transformed into 'free forms' and these are subsequently exhausted in elongating the seedlings. However, such dark period stimulates an adverse effect on biosynthesis of fresh gibberellins and this set back in gibberellin biosynthesis culminates in poor growth of such seedlings when transplanted and kept under normal conditions. However, it should be clearly mentioned here that water, temperature and radiation stresses are always operative in an interrelated manner and considerably effect the plant responses stimulated by each of them (Lange *et.al.*, 1981, 82). Mahala (2002), Larcher (2002) and Singh *et.al.*, (2003) using plants of diverse economic groups interpreted that such studies are pivotal in domesticational approach and also in biodiversity conservation strategies.

□□□

Reproductive Development under Simulated Environmental Stresses

Reproductive growth of the plant comparises flowering, fruiting and seed setting characters (Kaufman, 1972). This is the most important and crucial stage of the plant life and directly determines the economic yield in majority of plants. This phase is highly susceptible to environmental stresses (Larcher, 2002). The flowering process is controlled by a complex interaction of genetics and environment. Various components of environmental complex are known to act upon the complexity of the processes involved in the formation of flowers, fruits and seed (Leopold and Kriedemann, 1975). Effect of temperature has been studied by many workers on the flowering process (Zeevaart, 1967; Downs and Hillmer, 1975). However, temperature, light and water are known to influence or regulate the reproductive growth of plants in an interrelated and correlative manner (Evans, 1971; Levitt, 1980, 81; Nilsen and Orcutt, 1996).

The effect of quantity and quality of the radiation on the flowering process in plants, especially the cultivated species, has been studied by several plant biologists under natural as well as phytotronic environments (Majur and Johnson, 1974; Gupta, 1978; Christiansen, 1982). The role of moisture stress experienced naturally by plants or induced artificially with some specific objectives at various stages of plant growth including seed germination, seedling development, vegetative growth, preflowering stage or even during flowering period, has been investigated by some workers (Bidinger *et.al.*, 1996; Singh and Purohit, 2000). Screening of relevant literature has clearly revealed substantial lacunae in our knowledge about the role of abiotic stresses in controlling the reproductive growth of plants. Furthermore, the quantitative and qualitative determination of effects

of radiation, temperature and water stresses on flowering, fruiting and seed setting processes has not been performed upto reasonable extent in medicinal plants, particularly in the species belonging to Solanaceae and Papilionaceae families. Therefore, present work was conducted to evaluate the aspect of reproductive growth as influenced by certain environmental stresses.

Experimentia

For studying the long lasting effects of moisture stress the seeds were germinated (by the method described in Chapter 6) under ordinary laboratory condition. 15 days old seedlings with uniform shoot and root lengths were placed on Whatman No. 2 filter paper in sterilized borosilicate glass Petridishes. Each Petridish was supplied with PEG 6000 solution of different e.w.p. upto the half depth before placing the seedlings in these Petridishes (Singh and Singh, 1982b). All the seedlings were then covered with circular pieces of Whatman No. 2 filter paper already dipped in PEG solutions of specific concentrations. Seedlings placed in distilled water in Petridishes, served as control. All these treatments were given for 48 hours in 2 cycles of 24 hours each but interrupted by a non-stress gap of 24 hours. Then, the seedlings were transplanted in earthen pots containing garden soil following the same method as described earlier. Plant developed from these stressed seedlings were watered at regular intervals (every 3rd days) and were not allowed to experience any water stress till the experimental was terminated.

For studying the effect of moisture stress given to pot cultured plants (during vegetative growth) on reproductive growth, 15 days old healthy seedlings were transplanted in earthen pots and moisture stress was stimulated by the method described in Chapter 8. The data on reproductive growth was recorded by counting periodically the number of retained flowers, number of fruits and number of seeds per plant and data were finally cumulated. The effect of low and high temperature stress was studied on plants developed from seedlings which had been treated with various thermal stresses employed the method described in Chapter 8 with the modification that each treatment was given for 24 hours.

Reproductive growth in response to radiation stress was investigated by treating plants at seedling growth stage (15 days old) as per method described in Chapter 8. The effect of radiation stress given to plants at vegetative growth stage (preflowering stage) on reproductive growth was also studied by treating potted plants at the stage when 5th leaf was fully unfolded under conditions simulated

for seedling treatments (Chapter 8). However, this treatment was given only for 48 hours partitioned into 2 cycles of 24 hours each with a non-stress gap of 3 days and then the potted plants were transferred to normal conditions. Experimental design and statistical analysis were the same as adopted for earlier experiments (Chapter 8). These experiments were also conducted for two consecutive years under identical simulated stresses.

Experimental Observations

(A) Reproductive Growth in Response to Moisture Stress

Moisture stress given to seedlings for 48 hours under laboratory condition has retarded the reproductive growth except mild moisture stress (–3.0 bars external water potential). The mild moisture stress has promoted all the parameters of reproductive growth including the number of flowers/plant, number of fruits/plant, number of seed/plant and total dry weight of seeds. The effect of –10.0 bar external water potential was so severe in both the species that seedlings for further studies could not be maintained. In *T. purpurea* no seedling could survive at –7.5 bars treatment due to cent per cent stimulated seedling mortality (Table 9.1). When moisture stress was given to the seedlings well established in pots by withholding the water supply for definite intervals, the reproductive growth was progressively retarded but the effect was comparatively less severe than that observed for seedling treatment on an average *T. purpurea* was affected to a greater extent than *W. somnifera* by the reproductive growth retarding action of moisture stress given to plants (cultured in pots) at vegetative growth stage.

(B) Reproductive Growth in Response to Temperature Stress

It was observed that low temperature stress (5, 15 and 20°C) given to seedlings for 24 hours under laboratory conditions, produced remarkable effect on the reproductive growth of plants developed from these seedlings. 20°C temperature was found to be most effective in enhancing the number of flowers, fruits, seeds and dry weight in *W. somnifera*. In this species 30°C temperature also promoted the reproductive growth quite significantly. In case of *T. purpurea* 15°C temperature proved to be most favourable for reproductive growth enhancement. In case of *T. purpurea* 30°C temperature reduced the reproductive growth in terms of number of flowers, fruits and seeds/plant and average dry weight of seeds obtained from single plant. Temperature above 30°C has caused detrimental effect in both the species. However, *T. purpurea* proved to be more susceptible than *W. somnifera* (Table 9.2). The long lasting effect of temperature stress was

Table 9.1

Effect of moisture stress (different external water potentials) on certain reproductive growth parameters in *W. somnifera* and *T. purpurea*

Treatment (bar)	Seedling stage treatment				Treatment (days intervals)	Pre-flowering stage treatment			
	No. flowers/ plants	Nc. fruits/ plant	No. seed/ fruit	Total seed dry weight (gm.)		No. flowers/ plant	No. fruits / plant	No. seed/ fruit	Total seed dry weight (gm)
W. somnifera									
Control	35.5 a	20.3 a	20.9 a	0.600 a	3rd	36.4 a	21.3 a	21.4 a	0.640 a
–3.0	47.8 b	29.8 b	24.6 b	1.144 b	4th	54.6 c	44.5 c	26.4 c	1.820 c
–5.0	27.4 –b	11.5 –b	20.7 a	0.420 –b	5th	47.5 b	31.3 b	24.3 b	1.550 b
–7.5	11.6 –c	5.2 –c	16.4 –b	0.150 –c	6th	26.3 –b	14.4 –b	18.5 –b	0.420 –b
–10.0	0.0 –d	0.0 –d	0.0 –c	0.0 –d	7th	16.3 –c	8.4 –c	16.6 –c	0.250 –c
T. purpurea									
Control	47.7 a	30.5 a	6.4 a	2.500 a	3rd	48.6 a	31.5 a	6.1 a	2.700 a
–3.0	31.4 –b	19.6 –b	5.3 –b	1.520 –b	4th	58.7 b	46.5 b	6.5 b	4.310 b
–5.0	20.3 –c	10.8 –c	4.2 –c	0.660 –c	5th	41.6 –b	29.5 –a	5.7 –b	2.330 a
–7.5	0.0 –d	0.0 –d	0.0 –d	0.0 –d	6th	29.5 –c	11.7 –b	4.4 –c	0.670 –b
–10.0	0.0 –d	0.0 –d	0.0 –d	0.0 –d	7th	0.0 –d	0.0 –c	0.0 –d	0.0 –c

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level according to Duncan's multiple range test. All the figures are average of five replicates.

Table 9.2

Effect of moisture stress (given at seedling stage for 24 hours) on certain reproductive growth parameters in *W. somnifera* and *T. purpurea*

Plant	Treatment (°C)	Number of flowers/plant		Number of fruits/plant		Number of seed/fruit		Total seed/dry weight (gm)	
W. somnifera	Control	36.4	a	21.3	a	21.4	a	0.640	a
	0	0.0	–d	0.0	–d	0.0	–d	0.000	–d
	5	38.6	b	27.6	b	19.8	–b	0.850	b
	15	43.4	c	32.3	c	22.6	a	1.120	d
	20	49.6	d	39.6	d	25.7	b	1.620	e
	30	39.3	b	27.5	b	24.6	b	0.980	c
	40	21.4	–b	11.4	–b	19.6	–b	0.320	–b
	50	17.3	–c	8.9	–c	17.8	–c	0.210	–c
T. purpurea	Control	48.6	a	31.5	a	6.1	a	2.700	a
	0	0.0	–c	0.0	–d	0.0	–d	0.000	–d
	5	52.3	b	40.3	b	6.3	b	3.400	b
	15	61.4	d	52.5	d	6.7	c	4.200	c
	20	56.6	c	43.4	c	6.5	b	3.600	b
	30	46.3	a	26.3	–b	6.0	a	1.600	–b
	40	21.5	–b	9.8	–c	5.3	–b	0.560	–c
	50	—	–c	—	–d	—	–c	—	–d

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level according to Duncan's multiple range test. All the figures are average of five replicates.

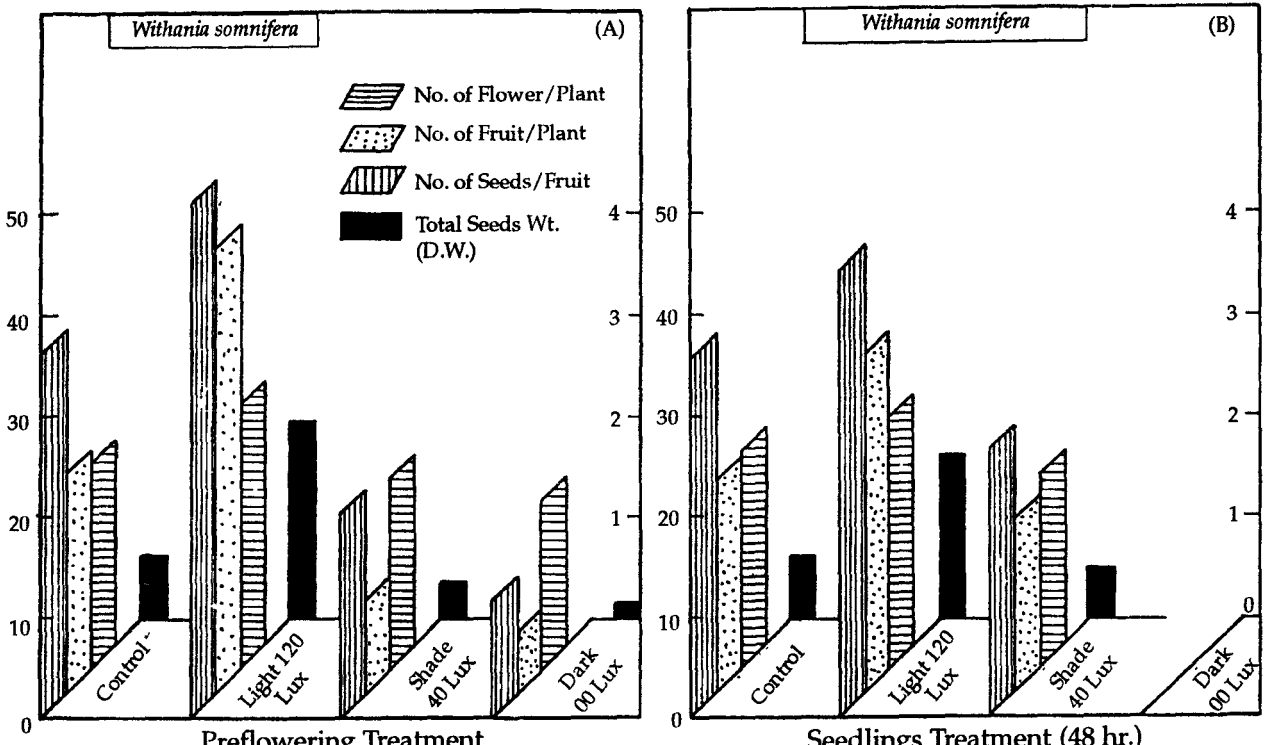

Fig. 9.1. A. Reproductive Growth Parameters As Affected by Various Radiation Stress Treatment *W. somnifera*.

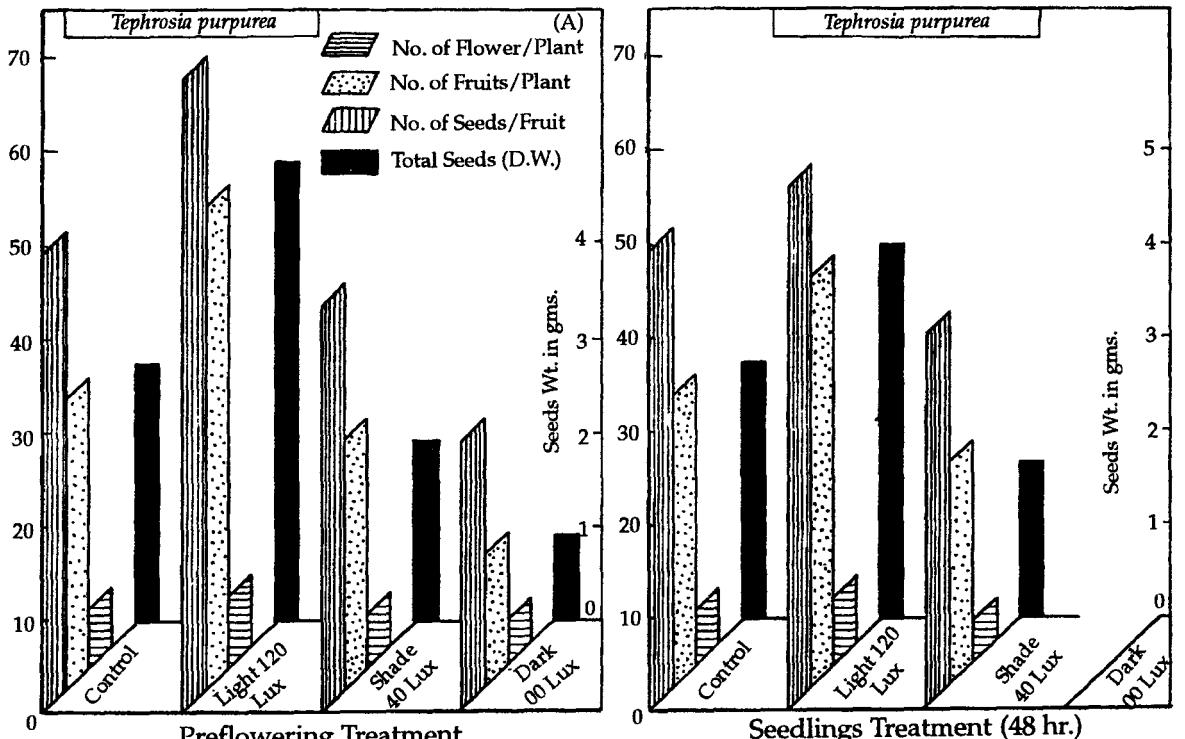

Fig. 9.1. B. Reproductive Growth Parameters as Affected by Various Radiation Stress Treatment in *T. purpurea*.

reflected in vegetative as well as reproductive growth both. However, efforts to subject the pot cultured and well established plants to temperature stresses of various degrees could not be successful because a growth chamber with regulated temperature and adequate space for treatment could not be constructed.

(C) Reproductive Growth in Responses to Radiation Stress

High light intensity (approx. 120 lux) given to seedlings for a period of 48 hours showed a long lasting effect on reproductive growth which was significantly enhanced in both the species. The light deficit stress (shade 40 lux) given to seedlings considerably reduced the reproductive growth in both the species. The absolute dark (0 lux) caused adverse effect upto such extent that seedlings could not survive instead of appropriate care. Both the species responded quantitatively similarly to these radiation stress treatments (Fig. 9.1 a, b). When the radiation stress was given to the plants at vegetative stage for 48 hours, the effects were variable. It was observed that the high radiation stress promoted the number of flowers, fruits and seeds and also the dry weight of the seeds in both the species. The light deficit and dark both have retarded the reproductive growth progressively. It was also noted that the stimulatory effect of high radiation stress was more pronounced when the same was given at vegetative stage. Contrary to this, the low radiation stresses were more effective when given at seedling stage. It was also observed that vegetative growth responses were in contrast to the reproductive growth responses in general to various radiation stresses (Fig. 9.1 A, B).

Interpretations and Applications

The cumulative and adverse effects of moisture stress given to seedlings or given to plants at vegetative stage on reproductive growth was also reported by earlier workers (Hsiao *et.al.*, 1976; Hsiao and Bradford, 1982). Hsiao and Bradford (1982) reported that high moisture stress during vegetative stage switched on a series of physiological reactions culminating in their effects on hormonal biosynthesis. Hartung and Witt (1968) with *Vicia faba* and *Halianthus annvus* and Hall *et.al.*, (1977) with tomato reported that excessive moisture stress tremendously reduced the endogenous levels of IAA, GA and cytokinin. Yang and Pratt (1978) suggested that high moisture stress stimulates ethylene production as well as ABA biosynthesis upto such level that these growth inhibitors reduce the number of reproductive primordia which culminates into a reduction of reproductive growth. Some workers interpreted that the detrimental

effect of water stress is mediated through its effect on adverse the photosynthesis and stomatal conductance (Jordan *et.al.*, 1975; Begg and Turnur, 1976; Pheloung and Barlow, 1981). The stimulatory effect of mild moisture stress on reproductive growth and yield has also been observed on farmers fields (Stanhill, 1977). However, very little systematic experimental work has been done on this abnormal response. Landsberg and Thorpe (1975) in barley also reported the stimulatory effect of moderate level of moisture stress on reproductive phase. They have further shown that flowering and fruiting was commenced earlier under mild moisture stress and the vegetative growth period was consequently reduced. Landsberg (1977) observed that hormones play an important part in the regulation of reproductive development under stress situation too. Landsberg and Thorpe (1975) suggested that this may be due to reduced gibberellin production and the ethylene is produced in such an amount that flowering is promoted. Otherwise, in higher concentrations (under high moisture stress) ethylene is inhibitory for the reproductive growth (Leiberman, 1979).

Harris and James (1969) with sunflower and castor, showed that a non-freezing low temperature stress (20, 10°C) given to seedlings or pot cultured plants exhibited a promotory effect on reproductive growth. Alden and Harmann (1977) called such behaviour of certain plants under low temperature (sub-freezing temperature) as a peculiar type of plant hardening response which was designated a very important phenomenon with agricultural point of view. They confirmed this fact by experimenting on large number of species of forest trees including Gymnosperms and Angiosperms. However, the degree of low temperature which proved to be stimulatory for reproductive growth, is highly variable depending upon the species, environmental conditions and time and cycles of low temperatures treatments. The actual physiological mechanism underlying this manifestation is still obscure (McDenial, 1982). The adverse effect of high temperature on reproductive growth of plants was also reported by earlier workers in some horticultural and agronomic crops (Anderson, 1977; Christiansen, 1978). McDenial (1982) concluded that long lasting but adverse effect of high temperature on reproductive growth is mediated through certain temperature sensitive enzymes which are converted into 'distorted forms' with disturbed three dimentional structure. However, the hormonal regulation of this response is yet to be worked out (Leopold, 1982).

Majur and Johnson (1974) with soybean cultivars also showed that increasing the light intensity upto a certain level, improved the quantitative aspects of reproductive growth under semicontrolled

condition. The number of flowers, fruits and seeds and dry weight of the seeds were increased. However, the light intensity above a critical level seemed to affect the reproductive growth adversely, Vergara *et.al.*, (1969) with rice cultivars also obtained similar results. Cathey and Campbell (1982) found that moderate to high radiation stress given to developing seedlings, triggers off a mechanism involving the phytochrome system which regulates the shifting of vegetative to generative stage and also the quantitative aspects of reproductive phase. They further reported that the intensity of light which enhanced the reproductive growth varied from species to species. This was confirmed by selecting poinsettia, chrysanthemum and soybean under similar environmental conditions. In this experiment, it was also shown that low light intensity and complete dark delayed or inhibited the flowering and fruiting processes. Franz (1993) with *Arnica montana* and Schroder (1998) with *Prunus africana,* medicinal plants of high export potential, showed that abiotic stresses exhibited a complex interaction in modulating the biological processes involved in biosynthesis of economically important secondary plant metabolites. Hence, such aspects should be critically and experimentally evaluated for determining the quality of herbal drugs.

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Plant Productivity and Economic yield as Modulated by Abiotic Stresses

Crops are generally selected for agricultural purposes by vertue of their productivity and yield which are usually recognized as biological yield and economic yield (Gupta, 1993). Many agronomists and physiologists have reported that biological as well as economic yield of the plant are highly affected by environmental stresses (Slayter, 1973; Levitt, 1980, 81; Gupta, 1993; Cooper and Hammer, 1996; Nosberger *et.al.*, 2001). Yield is a complex quantitative character which is the result of the complex interplay of the genetically operative hereditory make up of the plant and the environmental factors. In the set of prevailing conditions any one or all the factors may act to the extent of stress in plants. However, radiation, temperature and moisture stresses have been studied more frequently under field, green house and phytotronic conditions (Evans, 1973; Palmer, 1973; Singh and Gopal, 1973 etc.). The action of temperature stress in influencing the yield of plants was investigated by Bierhuizen (1973) in tomato and cucumber. Palmer (1973) in maize and Begg and Barton (1973) in pearl millet also conducted similar studies. Slayter (1973) concentrated his attention on the study of water stress in wheat crop. Fischer (1973) and Sen and Bansal (1978) studied some arid and semi-arid zone plants as affected by moisture stress.

The plant parts which provide the economic yield vary from crop to crop and may include any part of the plant or many parts simultaneously. The plant responses, therefore, in terms of productivity and economic yield differ considerably depending upon the biological nature of the economic yield providing parts. With pharmaceutical point of view, the yield generally comprises the dried plant parts

(powdered of intact). This is termed as crude drug yield (CDY). In modern pharmaceutical industry, therapeutic yield (TY) is considered as the amount of active principles (including alkaloids, glucosides, tannins, terpinoids and certain other 'so called' secondary plant products) present in dried crude drug (Atal and Kapur, 1982). However, the screening of published literature, has clearly revealed that information on crude drug yield and therapeutic yield responses of medicinal plants to environmental stresses are extremely scanty. Therefore, efforts were made to evaluate these yield parameters as influenced by certain environmental stresses in present plant materials.

Experimentia

Five plants from each treatment block were randomly selected for the study of productivity and economic yield of the plants under treatments. The treatments with radiation, temperature and moisture stresses were given as per procedure described in Chapter 8. The plants were carefully harvested with roots and washed with fresh water and then fresh weight of the whole plant and fresh weight of economic parts (root in *W. somnifera* and seeds in *T. purpurea)* were recorded and then these samples were dried in over at 80°C for 48 hours and dry weights of the whole plant and individual economic parts were noted. Productivity in all these descriptions refers the biological productively including the fresh and dry weights of the whole plant while economic yield refers the dry weight of the economic plant parts, the therapeutic and pharmaceutic yields have also been studied. Harvest index was determined as per standard formula given below:

$$HI = \frac{\text{D.W. of economic part}}{\text{D.W. of whole plant}}$$

The average crude drug yield (CDY) per plant was expressed as the dry weight (gm) of economic part while the therapeutic yield (TY) was expressed in terms of mg of total alkaloids content per unit dry weight of the economic part. The therapeutic yield was determined by estimating the total alkaloids content of specific economic part as per method described by Harborne (1973). Individual alkaloids of the economic parts of *W. somnifera* and *T. purpurea* were not determined because in Allopathic and Ayurvedic preparations, mixtures of alkaloids are used and not the partitioned principles. Each determination of total alkaloids content was conducted on 5 replicates of single plant each and all the experiments were repeated for two

consecutive years in the same months. However, no record of climatic condition was maintained.

Experimental Observations

(A) Productivity and Economic Yield in Response to Moisture Stress

Water deficit influenced the productivity in a depressive way. The dry as well as fresh weights were adversely affected by the moisture stresses of all the tested degrees in both the species significantly. However, the response to mild moisture stress was exception. This level of stress enhanced the economic yield of the *W. somnifera*. The therapeutic yield on the basis of per gram dry weight in *W. somnifera* and per two grams dry weight in *T. purpurea* was observed to increase with increasing moisture stress. Total pharmaceutical yield of the plant was decreased under the moisture stress caused by six days or more intervals of watering in *W. somnifera* but in case of *T. purpurea* pharmaceutical yield was reduced progressively by all the degrees of moisture stress (Table 10.1). Harvest index of the plant for *W. somnifera* has given abnormal results where roots constitute the economic part. Mild moisture stress reduced the harvest index and higher degrees of moisture stress enhanced the same. In *T. purpurea* harvest index was progressively reduced by all the tested moisture stress conditions.

(B) Productivity and Economic Yield in Response to Temperature Stress

Though, the low temperature stress was given to seedlings for a limited period, this was sufficient to enhance the fresh as well as dry weights of the whole plant at maturity stage in both the species. The sub-freezing low temperature (5°C) has shown a significant reduction of fresh and dry weights of economic part in *W. somnifera* while in *T. purpurea* the crude drug yield (CDY) was enhanced by all the degrees of low temperature stress. But when high temperature treatment was given to seedlings, the fresh and dry weights of economic parts of both the species were reduced significantly. The 30°C treatment has shown insignificant effect in both the species. Fresh weight and dry weight of the economic part in *W. somnifera* was decreased above 30°C while in *T. purpurea* fresh weight decreased at 30°C also (Table 10.2). Low temperature treatment (5, 15, 20°C) decreased the pharmaceutical yield in *W. somnifera* where this parameter was decreased under high temperature treatment (40 and 50°C) also.

Table 10.1

Effect of moisture stress on plant productivity and economic yield in *W. somnifera* and *T. purpurea*

Treatment (days)	Total Weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
W. somnifera							
Control	12.500 a	4.650 a	3.300 a	1.650 a	0.354 a	200 a	330 a
4th	14.940 b	5.540 b	3.654 a	1.720 a	0.310 –b	250 b	430 b
5th	10.580–b	4.120 a	2.120 –b	1.220 –b	0.530 c	310 c	682 c
6th	6.100 –c	1.940 –b	1.900 –b	0.750 –c	0.386 b	360 d	270 –b
7th	3.230–d	1.270 –c	0.950 –c	0.475 –d	0.374 b	400 d	190 –c
T. purpurea							
Control	18.600 a	7.950 a	2.700 a	1.900 a	0.238 a	310 a	294 a
4th	15.550–b	6.490 –b	1.510 –b	1.100 –b	0.171 –b	390 b	214 –b
5th	9.280 –c	3.450 –c	0.660 –c	0.450 –c	0.130 –c	420 c	94 –c
6th	5.380–d	2.140 –d	0.0 –d	0.0 –d	0.0 –d	0 –b	0 –d
7th	0.0 –e	0.0 –e	0.0 –d	0.0 –d	0.0 –d	0 –b	0 –d

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level of significance according to Duncan's multiple range test.

* Data for therapeutic yield for *T. purpurea* are on per two grams dry weight (of economic part) basis.

All the figures are average of five replicates.

Table 10.2

Effect of temperature stress on plant productivity and economic yield in *W. somnifera* and *T. purpurea*

Treatment (0°C)	Total Weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
W. somnifera							
Control	12.500 a	4.650 a	3.300 a	1.650 a	0.354 a	200 a	330 a
0	0.0 –d	0.0 –d	0.0 –e	0.0 –e	0.0 –d	0 –e	0 –f
5	13.260 a	4.960 a	2.710 –b	1.360 –b	0.274 –c	80 –d	108 –e
15	16.980 b	6.120 c	3.400 a	1.740 b	0.284 –c	110 –c	191 –c
20	16.930 b	5.750 b	3.810 b	1.950 c	0.339 a	160 –b	312 a
30	12.130 a	4.680 a	3.120 a	1.920 c	0.412 b	250 b	480 b
40	6.890 –b	2.410 –b	1.409 –c	0.750 –c	0.312 –b	310 c	223 –b
50	4.450 –c	1.500 –c	0.900 –d	0.460 –d	0.306 –b	380 d	174 –d
T. purpurea							
Control	18.600 a	7.950 a	2.700 a	1.900 a	0.238 a	310 a	294 a
0	0.0 –c	0.0 –c	0.0 –d	0.0 –d	0.0 –d	0 –d	0 –e
5	19.980 a	8.780 b	3.400 b	2.610 b	0.296 c	200 –c	260 –b

Contd....

Contd....

Treatment (0°C)	Total Weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
15	22.120 b	9.950 c	4.200 c	3.500 c	0.351 d	250 –b	437 c
20	23.910 c	10.120 c	3.610 b	2.810 b	0.276 b	270 –b	378 b
30	17.850 a	7.150 a	1.620 –b	1.200 –b	0.169 –b	340 a	204 –c
40	8.580–b	3.850 –b	0.560 –c	0.420 –c	0.049 –c	410 b	86 –d
50	0.0 –c	0.0 –c	0.0 –d	0.0 –d	0.0 –d	0 –d	0 –e

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level of significance according to Duncan's multiple range test.

* Data for therapeutic yield for *T. purpurea* are on per two grams dry weight (of economic part) basis.

All figures are averages of 5 determinations of one plant each.

The high temperature stress treatments have critically reduced the pharmaceutical yield in both the species. It was interesting to note that 30°C temperature has remarkably improved the total pharmaceutical yield in *W. somnifera*. Therapeutic yield was reduced by low temperature stress in both the species where high temperature stress improved the therapeutic yield. The harvest index was increased by low temperature (5 and 15°C) but decreased by high temperature (40 and 50°C) in both the species.

(C) Productivity and Economic Yield in Response to Radiation Stress

The high intensity radiation stress promoted the dry weight of economic parts, therapeutic yield and pharmaceutical yield in both the species. The low intensity of light (shade) and dark both decreased all the parameters of plant productivity excluding the fresh weight of *W. somnifera* in shade. Harvest index was improved by high radiation stress and light deficit both in *W. somnifera* where absolute dark reduced the harvest index. In *T. purpurea* also similar results were obtained. The maximum value of harvest index was recorded under shade in *W. somnifera* but under high radiation stress in *T. purpurea* (Table 10.3).

Interpretation and Applications

The reduction of crop yield in terms of dry matter production of the whole plant and of the particular economic part of plants under moisture stress has also been reported by earlier ecologists, agronomists and crop ecophysiologists including Slayter (1973) in cotton and Hsiao *et.al.*, (1976) in soybean, barley and oat Bradford and Hsiao (1982) mentioned that water deficit stimulated a retardation of crop growth and yield by including a reduction in photosynthesis and net assimilation rate, disturbing the hormonal balance in plants. The enhancement of plant productivity under simulated mild moisture stress condition in present studies is in agreements to the findings of Peters and Runkles (1967) with cotton, Kramer (1969) with soybean and Kapoor and Mitra (1979) with groundnut. The stimulatory effect of mild moisture stress on dry matter accumulation and crop yield is difficult to interpret, the actual mechanism (physiological) underlying this manifestation is not known (Bradford and Hsiao, 1982), Parson (1982) expressed the view that mild moisture stress switched on the biosynthesis of ethylene and the rate of ethylene production under such condition is regulated in such a manner that ethylene level becomes stimulatory for growth of root as well

Table 10.3

Effect of radiation stress on plant productivity and economic yield in *W. somnifera* and *T. purpurea*

Treatment (Lux) Approx.	Total Weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
W. somnifera							
Control	12.500 a	4.650 a	3.300 a	1.650 a	0.354 a	200 a	330 a
Light (120)	13.120 b	5.350 b	3.810 c	1.810 b	0.345 a	270 b	499 b
Shade (40)	13.600 b	4.610 a	3.620 b	1.750 a	0.380 –b	160 –b	280 –b
Dark (0)	9.600–b	2.990 –b	1.910 –b	0.910 –b	0.910 –b	120 –c	108 –c
T. purpurea							
Control	18.600 a	7.950 a	2.700 a	1.900 a	0.238 a	310 a	294 a
Light (120)	19.540 b	9.150 b	4.620 b	3.210 b	0.350 b	380 b	608 b
Shade (40)	17.150–b	5.950 –b	1.950 –b	1.350 –b	0.230 a	260 –b	175 –b
Dark (0)	12.850–c	4.300 –c	0.0 –c	0.0 –c	0.0 –b	0 –c	0 –c

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level of significance according to Duncan's multiple range test.

* Data for therapeutic yield for *T. purpurea* are on per two grams dry weight (of economic part) basis.

All the figures are average of five determinations.

reproductive parts. This view is also supported by the work of earlier researchers (Abeles, 1973; Lieberman, 1979). Milborrow (1974) reported that a mild moisture stress is operative through the closure of stomata regulated by ABA production. This leads to partial closure of stomata, reduction of water loss, enhanced photosynthesis and improved rate of assimilation.

The promotory effect of low temperature stress (above the sub-freezing temperature) on plant productivity and economic yield has also been a subject of few earlier investigations (Boyee *et.al.*, 1976; Shivraj, 1978; Berry and Raison, 1981). The injurious effect of high temperature stress on crop growth and yield was also observed by Berry and Raison (1981) who concluded that each species requires a specific range of temperature to reach specific stage of development and the deviation from these specific thermal conditions stimulates variable degree of responses in plants and these variations depend upon genetic make up of the species and environmental conditions. In present studies, the above facts is evident specially for low temperature stress. It may be seen that 5°C temperature proved to be stimulatory for yield in *T. purpurea* but in *W. somnifera* this temperature reduced the yield of the whole plant as well as its economic parts.

The stimulatory effect of high radiation stress on dry matter accumulation in both the species under present investigation indicated that these species are high light requiring plants (sun loving plants). These results are in agreement to the findings of Evans (1975) who reported that light is a key factor for reproductive process which determines the yield of plants. The adverse effect of light deficit on yield was also observed by Vergara (1978) with a number of rice cultivars. He concluded that decreasing the light intensity below a critical level always exerted on the adverse effect on crop yield. Similar results were also obtained by Shivraj (1978) and Levitt (1980, 81).

The promotory or inhibitory effects of environmental stresses on therapeutic yield, pharmaceutical yield and harvest index are difficult to interpret on account of the scantiness of available information on medicinal plants. In present investigation, the therapeutic yield is marked by total alkaloids content of roots in *Withania somnifera* where Withasomine is the active principle and by total alkaloids content of seeds in *T. purpurea* where Tephrine is the active principle. Some workers have studied the effect of environmental

stresses on alkaloid content in certain plants (Walter and Nawacki, 1978; Duke, 1982; Kaul and Jutshi, 1982; Yaniv and Palevitch, 1982). However, the pharmaceutical yield was reduced by moisture stress in present studies. This may be attributed to the adverse effect of water deficit on the dry matter of economic parts (please see earlier paragraph of this discussion for interpretation).

The results of effect of temperature stress on therapeutic yield and pharmaceutical yield are consistant with the findings of Walter and Nawacki (1978) and Svab *et.al.*, (1979). Bose (1984) also reported similar results in some Indian medicinal plants. These workers have studied the effect of radiation stresses on alkaloid content and obtained the results which are similar to the results of our experiments.

Arnon (1975) and Kaicker *et.al.*, (1978) expressed the view that when the yield is a chemical constituent (like alkaloids), the economically valuable component of the crop is only a small fraction of the total dry matter produced and moderate water/temperature/radiation stresses may show favourable effects on the pharmaceutical yield. They assumed that under these conditions, the decomposition of starch and proteins may favour the formation of so called 'secondary plant products' including alkaloids. Similar results were obtained by Svab *et.al.*, (1979) and Verzar *et.al.*, (1978).

Observations on harvest index in both the experimental plant species failed to furnish any conclusive information about the control of plant yield by environmental stresses. Data presented in table 10.1, 10.2 and 10.3 clearly indicated that the harvest index did not change in conformity with other parameters of yield (dry matter production of economic part, pharmaceutical yield and therapeutic yield) under moisture, temperature and radiation stresses. Arnon (1975) also indicated that the critaria of harvest index was not an absolute relationship and in several cases it has provided abnormal and opposite information. Raoson and Bremner (1974) with several varieties of wheat, barley and some other cereals and more recently Mann (1984) with several genotypes of durum wheat under field conditions also concluded that the harvest index showed weak associations with economic yield and therefore is a poor index of yield potential. It was further pointed out that the importance of determination of harvest index depended on the species, cultural practices and environmental conditions. The coefficient of effectiveness (harvest index) is more particularly a poor indicator of yield potential of the plant in the species where non-reproductive parts constitute the economic yield

(Kawakami, 1978). In present studies, also harvest index has given very limited information about the yield potential of even *T. purpurea* where seed constituted the economic yield. In case of *W. somnifera* this parameter practically failed to give any valuable information with pharmaceutical industry or agricultural point of view. Padulosi (2002) and Jakhar *et.al.*, (2003) selecting medicinal herbs of diverse growth habit also reported that samples of crude drugs collected from different niches exhibited tremendous variability in their contents of secondary metabolites. *Withania somnifera* also when collected from drier parts of Rajasthan exhibited differences in therapeutic yield. Hence, with export point of view, such investigations must be carried on priority basis.

□□□

Modulation of Plant Growth, Biomass and Economic Yield by Phytoregulants

The use of plant hormones and substances with hormone like activity in agriculture was first suggested of Cholodny (1936) who reported that hormonization of grains of wheat and oat resulted in an increase of crop yield upto 55%. Since then, a large number of reports have been published on fundamental and applied aspects of plant hormones (Malik, 1995). The principal growth regulators which have shown good potential in Agriculture and Horticulture include NAA, IAA, GA_3, 2, 4-D, ABA, ETH, BA, CME etc. (Krishnamoorthy, 1981; McLaren, 1982; Nickell, 1982; Purohit, 1983, 84; and Malik, 1995). Moolani and Kanitkar (1961) used growth regulators for breaking the dormancy in few traditional crops and observed that plant growth regulators have widely overlapping effects and functions. Sircar (1971) reported that IAA plays specific role in germination, seedling growth, shoot and root growth and in flowering also affects the yield of crops. Krishnamoorthy (1975) edited an excellent text on gibberellins and their regulatory roles in germination process, vegetative growth, flowering and fruiting and abscission of leaves and other plant parts. ABA was shown to act as an inhibitor of various processes of plant growth and development (Milborrow, 1974). Ethylene also plays an important role in seed germination and flowering (Abeles, 1973).

Cytokinins and morphactins are also, now-a-days, used for agricultural and horticultural purpose as some commercial formulation are now available. Naik (1954) used NAA for breaking the dormancy and enhancing the seedling growth in *Cicer arietinum* and *Pisum sativum*. GA was used for similar purpose in *Phaseolus*

aureus by Chakarvarti (1958). Ethylene was employed by Bisaria and Paliwal (1981) in *Triticale* and cytokinin by Harvey *et.al.*, (1974) in cucumber for similar purposes. Auxin and gibberellin were used for promoting the vegetative growth by Gopal and Singh (1982) in *Brassica juncia* while morphactin and cycocel in *Arabidopsis thialiana* by Sankhla (1970). Kinetin and IAA improved the growth and yield in *Pisum sativum* (Nandwal and Bharti, 1982). Weaver (1972), Papadakis and Gx (1978), Nickell (1982), Swaminathan (1982) and McLaren (1982) also emphasized the application of growth regulators in boosting the agricultural production. Atal and Kapur (1982) reviewed the literature on the cultivation and utilization of medicinal plants with particular refrence to Indian agroclimatic conditions. They advocated the application of new technology for medicinal plants cultivation including the use of growth regulators. Chadha (1976, 78) concluded that the application of growth regulators for improvement and cultivation of horticultural, plantation and medicinal plants is gaining momentum in India. Keeping in view, the scantiness of information on such aspects and the practical experience on the related field (Singh, 1979 in solanaceous crop plants), present studies were conducted with the objective to investigate the effect of growth regulators on various stages of growth, development and yield of two medicinal plants.

Experimentia

Uniform sized seeds of both the experimental species were treated with aqueous solutions of different concentrations of selected growth substances by keeping the seeds for 24 hours in glass Petridishes under ordinary condition of photoperiod and temperature. These treated seeds were quickly planted in earthen pots at uniform depth of garden soil. Seed germination was studied on the basis of emergence of cotyledons. Seedling growth was studied by measuring the shoot and root lengths of seedlings after 15 days of planting.

For studying the effect of growth substances on vegetative growth parameters, seedlings of uniform size were procured as per method described earlier in Chapter 8. These 15 days old seedlings were treated with various concentrations of growth substances by placing them in large Petridishes containing the test solutions (24 hours). The detail procedure was the same as adopted for studying the effect of external water potential. These treated seedlings were then transplanted into earthen pots and for subsequent period were subjected to normal environmental conditions. The procedure of investigation and cultural practices were the same as described in Chapter 8.

For studying the effect of growth substances on reproductive growth and plant productivity including the economic yield also, the treatments were given to 15 days old seedlings at laboratory conditions. These seedlings were transplanted in earthen pots as described for vegetative growth. Studies on various parameters of reproductive growth and yield were made by the method described in detail in Chapters 9 and 10. All these experiments were also repeated for two consecutive years and data were analysed by standard statistical measures (Duncan, 1955; Chandel, 1978).

Experimental Observations

(A) Growth Regulators in Relation to Germination, Seedling Growth and Vigour

NAA, GA_3 and BA showed promotory effects on germination and seedling growth in *W. somnifera* as well as in *T. purpurea* while CME and ABA showed inhibitory effect on the same. ETH promoted the germination but the seedling growth was retarded by the treatment. NAA promoted the germination percentage with increasing concentrations in *W. somnifera* while higher concentrations reduced the same in *T. purpurea*. NAA exhibited dual action in influencing the growth of plumule and radicle. The lower concentrations of NAA increased the same but higher doses reduced the seedling growth in both the species. GA_3 also exhibited promotory effect with increasing concentrations on seed germination and seedling growth in both the species. 1, 10 and 20 mg/*l* concentrations of BA progressively and significantly promoted the seed germination percentage and seedling growth in both the species, 50 mg/*l* concentration of this synthetic cytokinin, though, significantly promoted seed germination and seedling growth, the effect was not progressive and it was quantitatively similar to that caused by 20 mg/*l* (Table 11.1) of BA.

CME in low concentrations showed a progressive enhancement of germination process and seedling growth in both the species but higher concentrations of CME were found inhibitory for germination as well as seedling growth. All the tested concentrations of ABA showed inhibitory effect on all the parameters of germination and seedling growth in both the species excluding only lower concentration of ABA which promoted the radicle length in *W. somnifera* only. Ethylene enhanced the germination in both the species excepting only higher concentration which depressed the germination process in both the species under present investigations. The plumule experienced the adverse effect under this treatment while the radicle

length was increased by lower concentrations of ETH. However, higher concentrations of ETH showed inhibitory effects on the seedling growth (Table 11.1). The seedling vigour was promoted by tested concentrations of auxin, gibberellin and cytokinin in both the speices. However, 200 mg/*l* concentration of NAA proved to be inhibitory for seedling vigour. This parameter was promoted by lower concentrations of CME which reduced the vigour when applied in higher concentrations. The lowest tested dose of ABA showed a mild stimulatory effect on seedling vigour in both the experimental species. Lower concentrations of ethylene exhibited promotory effect on seedling vigour which was, otherwise, retarded by highest concentration of ethephon.

(B) Growth Regulators in Relation to Vegetative Growth

NAA (applied at seedling stage) showed cumulatively promoting effect on shoot and root lengths in lower concentrations while higher concentrations of NAA suppressed the parameters of vegetative growth in both the species. The leaf area was enhanced by the NAA treatment (excluding only the highest concentration which showed inhibitory effects in *W. somnifera* as well as in *T. purpurea*). The dry matter production was promoted by lower tested concentrations of NAA which in higher doses reduced the dry matter production. The most effective and physiologically active concentration of NAA proved to be 50 mg/*l* both the species (Table 11.2) in exhibiting a long lasting effect on vegetative growth. GA_3 exhibited a progressive enhancement of parameters of vegetative growth but increasing the concentration beyond 100 mg/*l* level did not bring any more fruitful results and thus, 100 mg/*l* concentration of GA_3 proved to be most effective in present investigations (Table 11.2). Benzyl adenine (BA), a synthetic cytokinin, showed the effect similar to that caused by GA_3 but relatively lower concentrations of BA could produce the similar effect and 20 mg/*l* concentration proved to be most effective (Table 11.2). On an average BA proved to be most potent promoter of various aspects of vegetative growth in *W. somnifera* and *T. purpurea* both.

Results of morphactin treatment were found to be interesting as 20 and 50 mg/*l* concentrations of CME drastically reduced the shoot and root growth, total leaf area per plant, fresh weight per plant and also the total dry weight per plant in *W. somnifera* and *T. purpurea* both. Lower tested concentrations of CME (1 and 10 mg/*l*) promoted the parameters of vegetative growth significantly and progressively in *W. somnifera* but in *Tephrosia purpurea* these concentrations have acted similar to the higher concentrations of morphactin (Table 11.2).

Table 11.1

Seed germination percentage, seedling growth and seedling vigour under treatments with different concentrations of growth substances in *W. somnifera* and *T. purpurea*

Treatments		*W. somnifera*						*T. purpurea*					
Conc. (mg/*l*)		Germination (%)	Seedling length (cm)			Seedling vigour		Germination (%)	Seedling length (cm)			Seedling vigour	
			Shoot		Root				Shoot		Root		
Control		37.4 a	5.7	a	6.9	a	258.6 a	42.4 a	6.4	a	7.3	a	308.5 a
NAA	10	41.7 b	6.3	b	7.8	b	325.2 b	57.6 c	7.6	b	8.4	b	483.8 b
	50	49.4 c	7.4	c	8.4	c	414.9 d	63.4 d	8.9	c	7.6	a	475.8 b
	100	58.6 e	8.6	d	6.4	a	375.0 c	52.4 b	7.1	b	5.6	–b	293.4 –b
	200	52.4 d	4.3	–b	4.9	–b	256.8 a	36.3 –b	5.3	–b	4.7	–c	173.6 –c
GA_3	10	48.5 b	6.1	b	7.4	b	356.9 b	55.3 b	7.5	b	8.6	b	475.5 b
	50	56.7 c	7.6	c	8.9	c	504.6 d	61.5 c	9.8	c	10.7	c	658.0 d
	100	63.8 d	9.9	e	10.8	d	689.0 e	67.6 d	12.9	e	8.4	b	567.8 c
	200	68.7 e	8.2	d	7.1	a	487.7 c	71.5 e	10.6	d	6.9	–b	636.3 d
BA	1	51.4 b	7.4	b	7.6	b	390.6 b	59.4 b	8.9	b	8.7	b	516.7 b
	10	57.3 c	8.9	c	7.7	b	441.2 c	68.7 c	9.7	c	9.1	c	635.3 d
	20	63.5 d	9.3	d	8.6	d	563.1 e	70.4 d	10.9	d	9.7	d	672.8 e
	50	60.7 d	8.8	c	8.0	c	485.6 d	67.3 c	9.6	c	8.8	b	592.2 c

Contd...

Contd...

Treatments		*W. somnifera*				*T. purpurea*			
Conc. (mg/*l*)		Germination (%)	Seedling length (cm) Shoot	Root	Seedling vigour	Germination (%)	Seedling length (cm) Shoot	Root	Seedling vigour
CME	1	43.6 b	7.5 b	7.3 b	318.2 b	54.5 b	8.2 b	9.1 b	495.9 c
	10	52.6 c	4.8 –b	5.1 –b	268.2 a	59.6 c	5.6 –b	5.9 –b	351.6 b
	20	21.3 –b	3.2 –c	4.3 –c	91.5 –b	39.3 –b	4.3 –c	3.4 –c	133.6 –b
	50	16.3 –c	2.4 –d	1.2 –d	19.5 –c	21.2 –c	2.1 –d	1.2 –d	25.4 –c
ABA	0.1	28.3 –b	4.6 –b	8.7 b	246.2 a	40.6 a	5.3 –b	8.1 b	328.8 b
	1.0	17.4 –c	3.5 –c	6.4 a	111.3 –b	31.4 –b	3.9 –c	6.3 –b	197.8 –b
	10.0	10.5 –d	2.4 –d	1.7 –b	17.8 –c	24.3 –c	2.1 –d	4.2 –c	102.0 –c
	20.0	8.3 –d	1.2 –e	1.1 –c	9.1 –d	11.4 –d	1.1 –e	9.7 –d	7.9 –d
ETH	10	57.6 b	4.8 –b	7.9 b	455.0 b	55.6 b	5.6 –b	8.9 b	494.8 c
	50	63.4 c	3.7 –c	8.7 c	551.5 c	68.7 c	4.7 –c	7.1 a	487.7 c
	200	71.5 d	2.4 –d	3.6 –b	257.4 a	74.3 d	3.2 –d	5.4 –b	405.2 b
	500	29.6 –b	1.1 –e	1.7 –c	31.3 –b	42.3 a	1.6 –e	1.4 –c	59.2 –b

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level according to Duncan's multiple range test.

All the figures are average of 5 replicates of 5 units each but data on germination percentage are averages of 5 replicates of 20 seeds each.

Table 11.2

Cumulative effect of treatments with growth substances (given to 15 days old seedlings) on vegetative growth parameters in *W. somnifera* and *T. purpurea*

W. somnifera

Treatments	Conc. (mg/l)	Shoot Length (cm)	Shoot ACL	Root Length (cm)	Root ACL	Total leaf area (cm^2)	Total FW/ Pl. (gm)	Total DW/Pl. (gm)	Germinability** (%)
Control		40.7	—	26.5	—	427	11.600	4.750	38.7
NAA	10	46.3	5.6	37.5	11.0	474	12.700	5.280	40.3
	50	61.7	21.0	48.3	21.8	493	15.600	6.650	45.4
	100	32.8	7.9	21.4	-5.1	348	10.340	3.240	45.4
	200	30.1	-10.6	18.6	-7.9	306	3.730	2.470	51.7
L.S.D.			2.8		3.1	36.4		1.240	2.11
GA_3	10	43.4	2.9	29.6	3.1	489	12.500	5.350	43.8
	50	53.6	12.9	47.6	21.1	546	15.740	6.690	46.3
	100	70.5	33.8	41.3	14.8	646	17.300	7.370	56.4
	200	68.6	27.9	32.4	5.9	572	13.560	5.560	58.7
L.S.D.			2.7		2.1	47.5		1.360	1.98
BA	1	48.7	8.0	31.4	4.9	524	13.300	5.680	47.3
	10	71.5	31.7	43.4	16.9	587	16.430	6.910	52.4
	20	77.4	36.4	48.3	21.8	643	19.920	8.210	58.9
	50	69.9	29.2	36.9	10.4	605	18.130	7.960	55.7
L.S.D.			3.6		3.4	49.3		1.140	2.31
CME	1	48.3	7.6	31.4	4.9	465	12.340	5.460	40.7
	10	51.4	19.7	36.6	10.1	564	20.400	7.310	44.5
	20	24.3	-16.4	21.3	-5.2	313	8.530	3.680	24.9
	50	17.5	-23.2	19.4	-7.1	204	4.910	2.150	21.9
L.S.D.			2.6		2.7	37.4		0.980	2.1
ABA	0.1	30.7	-10.0	36.7	10.2	323	10.800	4.310	33.4
	1	19.4	-21.3	21.4	-5.1	285	8.340	2.830	24.6
	10	11.3	-29.4	8.9	-17.6	214	6.430	2.570	19.8
	20	8.6	-32.1	6.7	-19.8	123	8.160	1.320	17.6
L.S.D.			3.0		3.6	41.4		0.650	1.73
ETH	10	34.4	-6.3	31.4	4.9	365	11.130	4.650	48.4
	50	21.3	-19.4	17.6	-8.9	324	9.340	3.980	51.3
	200	15.4	-25.3	14.5	-12.0	223	5.810	2.480	58.9
	500	11.3	-29.4	9.4	-17.1	163	3.470	1.450	40.7
L.S.D.			4.1		3.4	38.5		0.680	1.83

Contd...

Contd...

Treatments Conc. (mg/l)	*T. purpurea*							
	Shoot		Root		Total leaf area (cm^2)	Total FW/ Pl. (gm)	Total DW/Pl. (gm)	Germinability** (%)
	Length (cm)	ACL	Length (cm)	ACL				
Control	52.8	—	24.4	—	307	16.400	7.400	43.8
NAA 10	57.3	4.5	27.3	2.9	344	18.420	8.520	54.5
50	68.4	15.6	20.5	5.1	486	21.630	9.910	59.3
100	50.3	-2.5	21.4	-3.0	287	14.100	7.020	47.5
200	32.3	-20.5	16.3	-8.1	224	10.600	4.380	38.8
L.S.D.		2.3		2.6	27.2		1.430	1.9
GA_3 10	55.3	2.5	27.6	3.2	413	17.620	8.410	52.1
50	63.5	10.7	31.4	7.1	475	19.130	8.820	58.4
100	78.6	25.8	37.5	13.1	514	24.460	11.380	62.1
200	71.4	18.6	29.6	5.2	468	18.610	8.630	60.1
L.S.D.		1.9		2.7	29.4		0.950	2.24
BA 1	67.5	14.7	29.3	4.9	474	17.910	8.530	62.4
10	79.3	26.5	37.4	13.0	546	24.830	11.620	64.8
20	88.3	35.5	39.8	15.4	605	29.310	12.830	67.1
50	82.1	29.3	31.2	6.8	573	25.930	12.130	65.3
L.S.D.		3.2		2.3	39.5		1.120	1.8
CME 1	46.5	-6.3	20.8	-3.6	244	15.230	5.940	53.2
10	31.4	-21.4	19.2	-5.2	215	11.310	5.190	55.7
20	21.3	-31.5	12.5	-11.9	186	9.180	4.270	40.1
50	14.4	-38.4	8.4	-16.0	113	7.460	2.380	24.5
L.S.D.		3.8		2.9	29.5		0.850	1.46
ABA 0.1	39.3	-13.5	31.3	6.9	265	10.130	4.650	42.8*
1	24.4	-28.4	17.8	-6.6	214	7.340	3.490	35.1
10	18.6	-34.2	11.7	-12.7	176	6.530	2.950	28.1
20	9.8	-43.0	8.5	-15.9	94	3.980	2.110	16.3
L.S.D.		4.8		3.9	31.6		0.790	1.8
ETH 10	44.5	-8.3	29.6	5.2	273	17.600	7.910	58.4
50	38.6	-14.2	14.4	-10.0	262	14.530	6.940	60.5
200	27.3	-25.5	11.3	-13.1	216	8.310	3.830	66.2
500	16.7	-36.1	6.8	-17.6	168	5.710	2.680	37.4*
L.S.D.		3.7		2.8	33.4		0.650	3.1

Each figure is secondary average of 5 replicates.

* Insignificant; ** Germinability of seeds obtained from plants which were treated with growth substances at seedling growth stage.

Synthetic ABA and ethrel (ETH) acted in similar manner in retarding the vegetative growth of both the species. However, 0.1 mg/l concentration of ABA and 10 mg/l concentration of ETH promoted the root length in both the species (Table 11.2).

(C) Growth Regulators in Relation to Reproductive Growth

The number of flowers produced/plant was promoted by NAA concentrations progressively in *W. somnifera*. However, in *T. purpurea* 200 mg/l concentration of NAA significantly reduced the number of flowers borne, number of fruits/plant and the number of seeds/fruit/plant. In both the species seeds obtained from NAA treated (lower concentrations) plants have shown better germination than control plants. Gibberellic acid treatment given to seedlings has also shown promotory and long lasting effect on all the aspects of reproductive growth. 100 mg/l concentration of GA proved to be most effective and physiologically active in this respect. Seeds obtained from GA_3 treated plants showed better germinability when tested under laboratory conditions. All the tested concentrations of BA showed stimulatory effect on parameters of reproductive growth and germinability of seeds obtained from BA treated plants. 20 mg/l concentration of BA proved to be most effective. The long lasting effect of growth substances (NAA, GA_3 and BA) were compared and it was observed that BA proved to be most potent growth regulator with reference to long lasting effects on reproductive growth.

Morphactin proved to be a retardant of reproductive growth in both the species except 1 mg/l concentration which promoted number of flowers, fruits and seeds per plant in *W. somnifera*. In *T. purpurea*, cent per cent flowers dropped prematurely on plants treated with 50 mg/l concentration of CME at seedling stage resulting in total loss of reproductive parts and yield. ABA and ETH proved to be general retardants of reproductive growth for both the species. On an average *T. purpurea* proved to be more responsive to reproductive growth retarding action of abscisic acid and ethephon. Seeds obtained from such plants exhibited poor germination potential under laboratory test.

(D) Growth Regulators in Relation to Productivity and Economic Yield

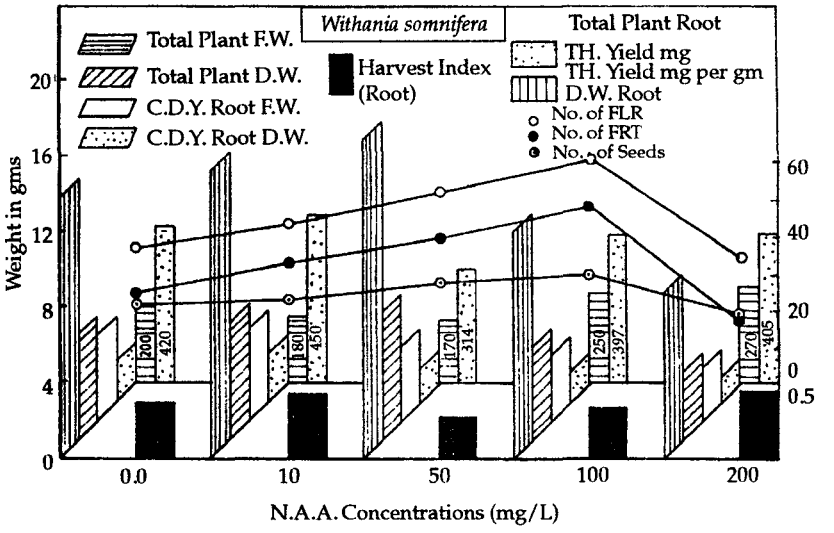
In general, lower concentrations of NAA enhanced the dry matter production and higher concentrations reduced the same in both the species. Similar results were obtained for yield of economic parts in present studies. 10 and 50 mg/l NAA enhanced the harvest index in *W. somnifera* where all the higher doses reduced the

parameter. In case of *T. purpurea* harvest index was promoted by all the tested doses of NAA but most satisfactory results were obtained with 50 mg/l concentration of NAA in this species. The therapeutic yield (total alkaloids content) was reduced by 10, 50 and 100 mg/l concentrations of NAA in *W. somnifera* where 200 mg/l concentrations of NAA promoted the total alkaloids content. However, the pharmaceutical yield could be enhanced by 10 mg/l concentration only of NAA in *W. somnifera* while all the other tested concentrations of NAA reduced this aspect of economic yield in this solanaceous species. The therapeutic as well as pharmaceutical yields were recorded to be lowered down by NAA concentration in *T. purpurea*.

GA_3 treatment enhanced the dry matter production for the whole plant as well as for economic parts in both the species. Harvest index was found to be highest in response to 100 mg/l of GA treatment. The therapeutic yield was decreased with increasing concentrations of GA_3. 100 mg/l GA_3 in *T. purpurea* and 50 mg/l GA_3 in *W. somnifera* gave highest pharmaceutical yield. BA proved to be a general promoter of plant productivity in both the experimental species but the therapeutic yield of both the species was considerably reduced by this treatment. All the tested concentrations of BA were recorded to be suppressive for the pharmaceutical yield in both the species except 20 mg/l of BA in *T. purpurea*. In this case the pharmaceutical yield was significantly promoted.

CME decreased the dry matter production in both the species except a mild stimulatory effect of lower concentrations of CME in *W. somnifera*. The dry weight of economic parts was also decreased by CME in both the plant species. However, in *W. somnifera* dry weight of economic part was improved by lower concentrations. Highest harvest index was observed under 50 mg/l CME treatment in *W. somnifera* but in *T. purpurea* the peak value of harvest index was obtained at 1 mg/l concentration of morphactin. Morphactin in lowest concentration decreased the pharmaceutical yield and therapeutic yield in *W. somnifera* while all the other (higher) doses were observed to be promotory for such parameters in this species. 1, 10, 20 mg/l concentrations of CME were found to favour the therapeutic yield in *T. purpurea* where the pharmaceutical yield was increased by 1 mg/l concentration only.

ABA proved inhibitory for productivity response in general in both the species excluding only pharmaceutical yield which was promoted by 0.1 and 1.0 mg/l of ABA in *W. somnifera* but 10 and 20 mg/l concentrations of ABA drastically reduced the pharmaceutical yield in *T. purpurea*. All the higher concentrations of ETH were found



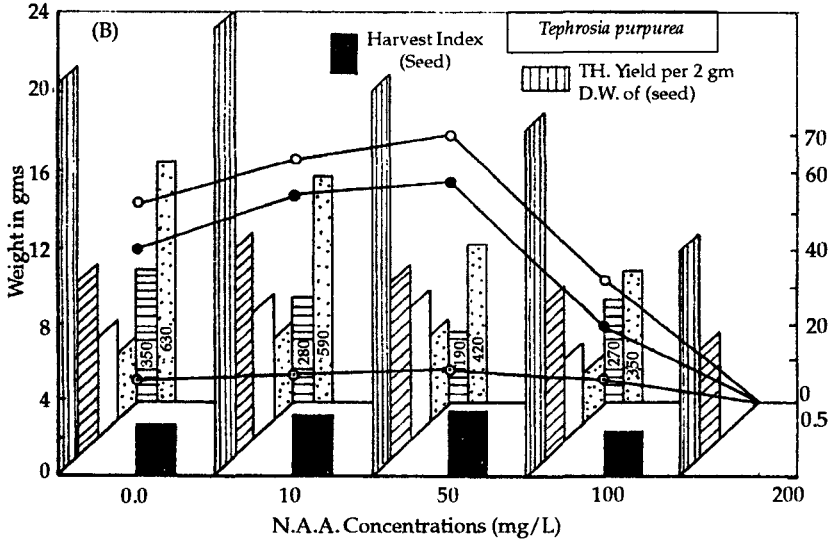


Fig. 11.1 A. Productivity and Economic Yield Parameters as Effected by NAA Treatment.

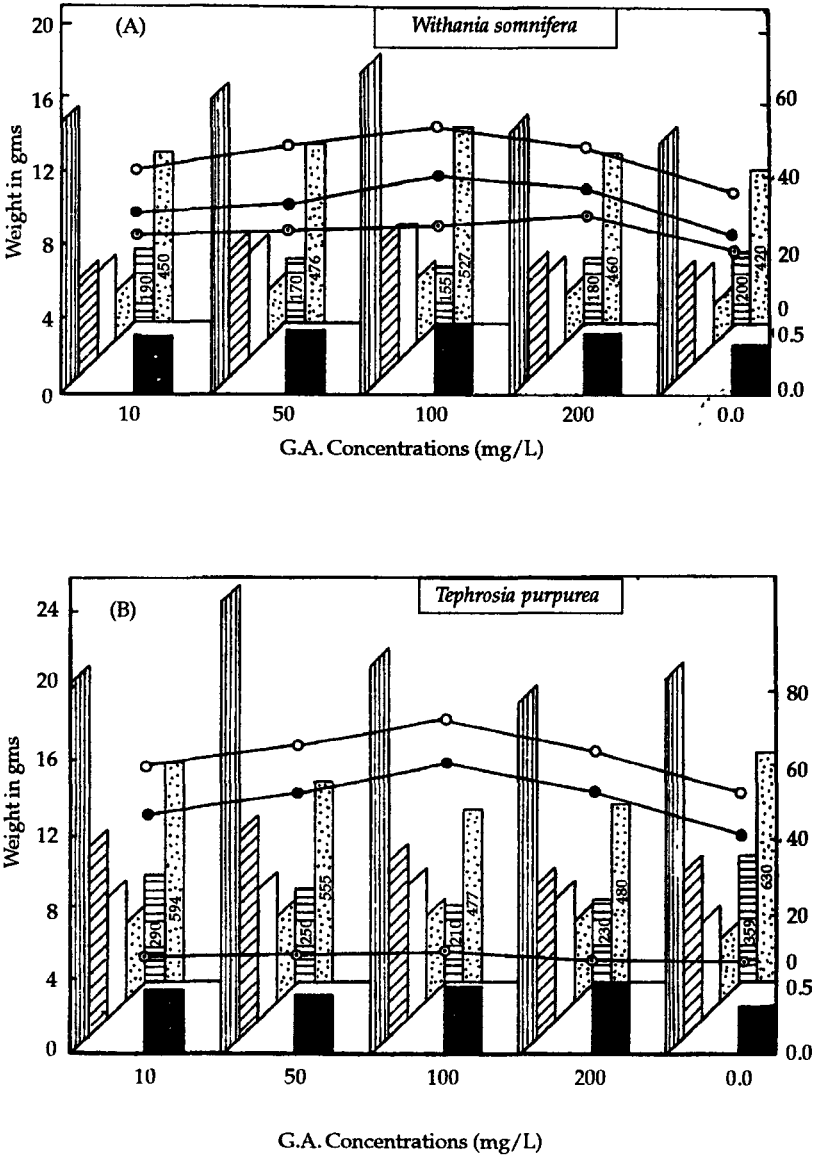


Fig. 11.1 B. Productivity and Economic Yield Parameters as Affected by Gebberellin Treatment.

* Index as per Fig. 11.1 A

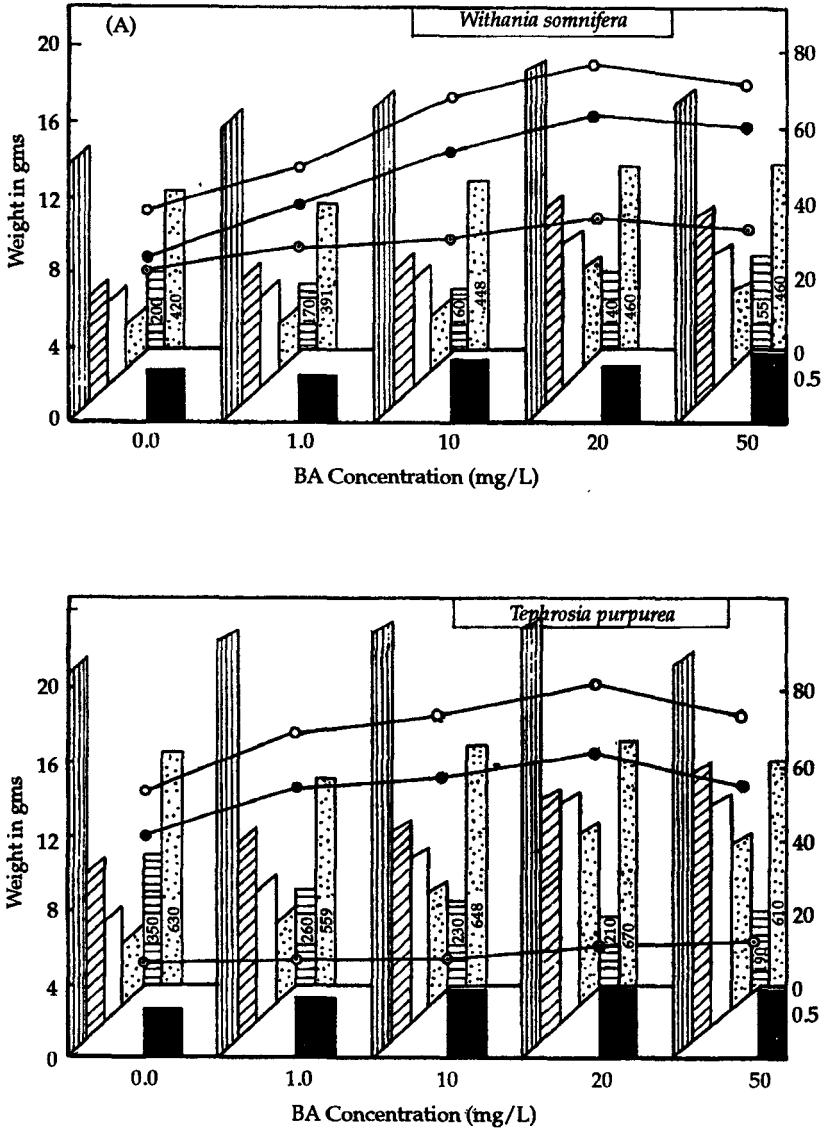


Fig. 11.1 C. Productivity and Economic Yield Parameters as Affected by BA Treatment.

* Index as per Fig. 11.1 D.

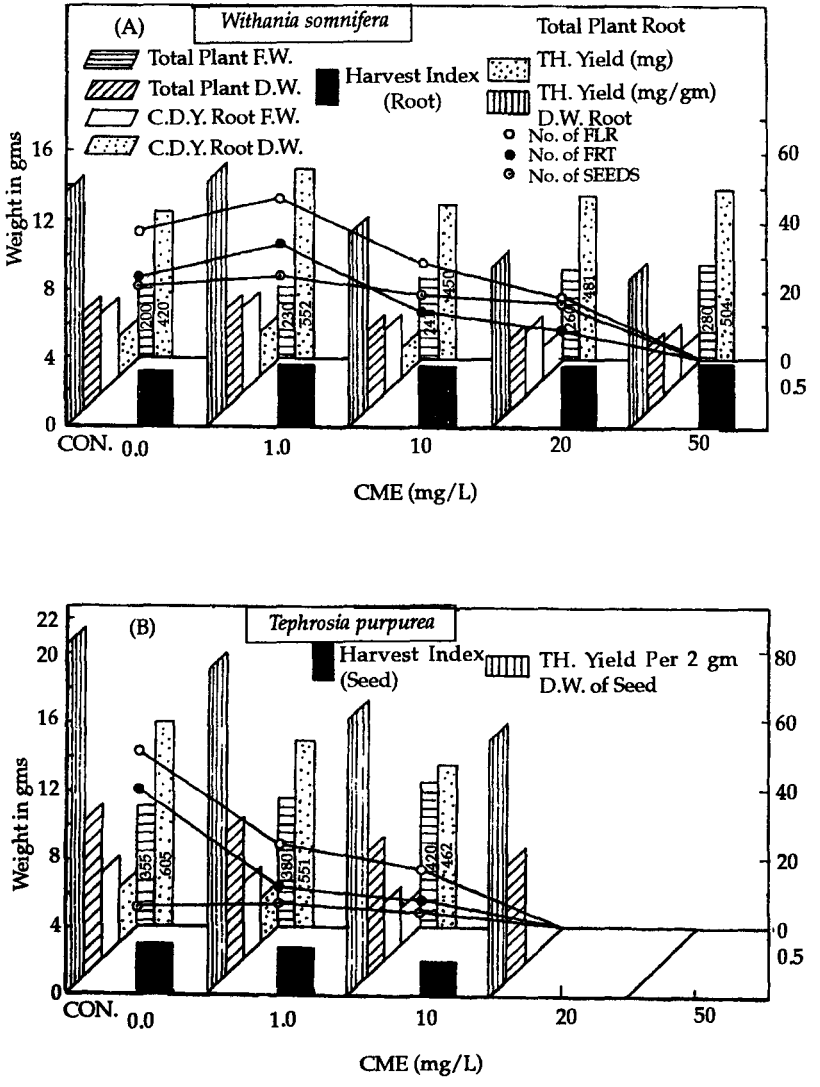
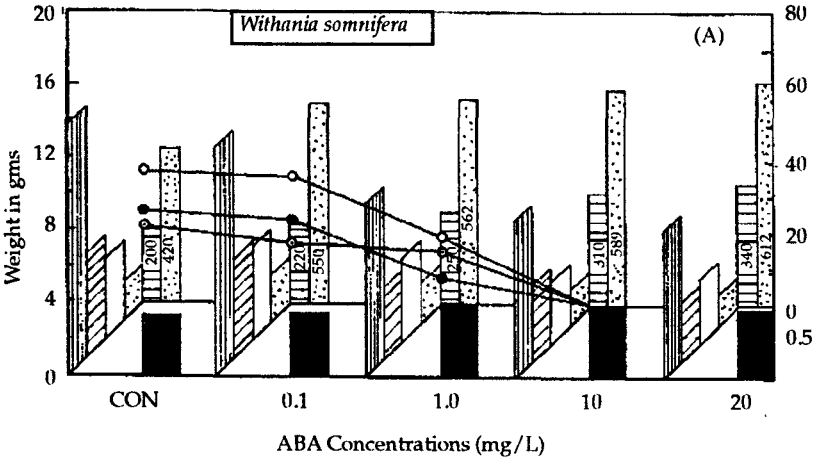


Fig. 11.1 D. Productivity and Economic Yield Parameters as Affected by Morphactin Treatment.



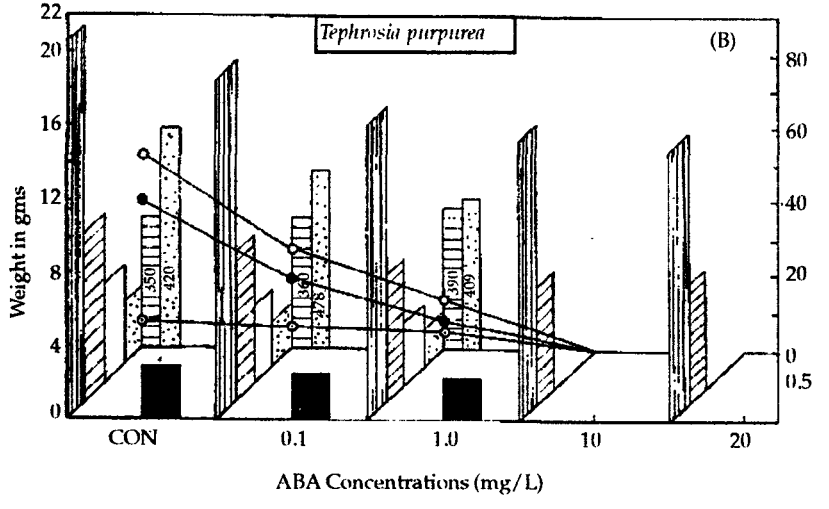


Fig. 11.1 E. Productivity and Economic Yield Parameters as Affected by BA Treatment.

* Index as per Fig. 11.1 D.

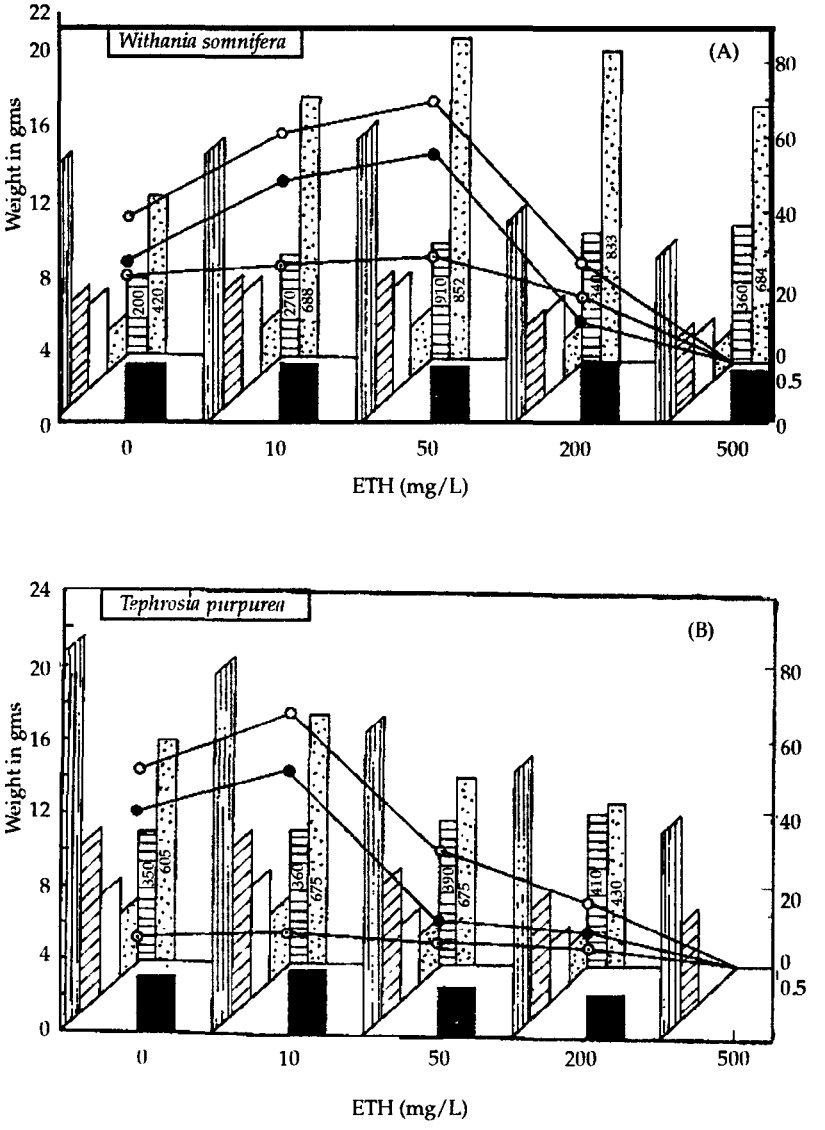


Fig. 11.1 F. Productivity and Economic Yield Parameters as Affected by Ethylene Treatment.

* Index as per Fig. 11.1 D.

to be inhibitory for dry matter production of the whole plant and of economic parts in *W. somnifera* and *T. purpurea*. Lower concentrations of ETH increased the same in both the species. Therapeutic yield was however, increased by ethephon treatment in both the species except very high concentration in *T. purpurea*. The pharmaceutical yield was enhanced by 10 and 50 mg/*l* concentrations of ETH in both the species. However, 500 mg/*l* of ETH proved to be inhibitory with this point of view. Data for such observations are represented by the Figures 11.1 a, b, c, d, e and f.

Interpretations and Applications

The germination behaviour of the seeds treated with distilled water (control) and then planted in the soil in pots considerably differed from the germination responses of similar seeds when kept for germination of Whatman No. 2 filter paper in Petridishes under laboratory conditions Chapter 4 and 5. The improvement in germination percentage of seeds when planted in soil is attributable to more than one factors including soil reaction and pressure. The stimulatory effect of edaphic pressure on germination of certain seeds was also reported by Anderson (1968) in *Physalis, Solanum* and *Trifolium*. Furthermore, the germination inhibitors present in seeds are leached out from germinating seeds and absorbed by the soil colloids. This action of the soil on seeds has also been observed by Lewak (1984). Promotion of seed germination by physiologically active concentrations of NAA was also reported by Rypack and Kamanicka (1982) in *Vibrum* species. The promotory effect of gibberellin on seed germination of certain medicinal plants has been a subject of few earlier reports (Chen, 1975; Black, 1980; Shrivastava and Day, 1982). The promotory effect of benzyl adenine on seed germination is in agreement to the findings of Shrivastava and Dey (1982) in *Helianthus annuus* L. The concentration dependent dual action of morphactin on seed germination was also reported by Schneider (1970, 72), Bopp (1972) and Sankhla *et.al.* (1975). Tayal and Gopal (1976) using *Trigonella* as the experimental material, have shown the inhibitory effect of higher concentrations of morphactin on seed germination and early seedling growth. Germination responses of present plant materials to the ETH and ABA treatment supported the observations of Abeles (1973), Milborrow (1974) and Moore (1980).

Lewak (1984) interpreted the effects of several growth regulators on seed germination by inhibitor-promoter equilibrium concept. He mentioned that cytokinins plays a key role in the

regulation of this complex interaction of hormones in which other hormones exhibit their effects by increasing or decreasing the endogenous level of natural cytokinins. Lewak (1984) also presented experimental evidences in support of his concept.

Promotion of seedling growth and vigour by relatively lower concentrations of NAA and inhibition by higher concentrations of the same, seemed to be the result of persistence of the action of NAA upto seedling growth stage. Similar result were also reported by Kumar *et.al.*, (1984) in some brinjal cultivars. The promotory effect of gibberellin and cytokinin on shoot and root growth of subsequently developed seedlings was also reported by Paleg (1965) in lettuce, Skoog and Armstrong (1970) in *Phaseolus* and Borkowska and Rudnicki (1975) in apple, Bopp (1972) also reported the shoot and root growth retarding effect of morphactins in tomato, brinjal, pea and soybean. Parups (1983) showed that the growth retarding activity of morphactins may be attributed to the inhibition of endogenous auxin synthesis and transport in germinating seeds and growing seedlings. The inhibitory effect of almost all the concentrations of ethephon and abscisic acid on seedling growth and vigour is interpretable of the basis of findings of Abeles (1973). Milborrow (1974) and Shimokowa (1984) concluded that the inhibitory effect of ethylene as well as abscisic acid is mediated through their adverse effects on DNA, RNA and protein biosynthesis and also to their inhibitory effect on enzyme biosynthesis, specially those enzymes which are related to successful metabolism, a part of seedling growth.

Choudhary and Chakarvarti (1982) carried out detailed studies on the effects of synthetic auxin on *Citrus* and observed a dual action of NAA on shoot and root growth, leaf area and dry matter production of the vegetative parts. Low (1975) also reported the growth promotory effects of gibberelins on shoot and root length and leaf area in some crop plants. The mode of action of gibberellins in promoting various aspects of vegetative growth, has been a matter of critical evaluation by some workers. Low (1975) and Lewak (1984) concluded that the effect of gibberellin treatments at early stage of plant development may continue till vegetative and reproductive growth stages and may be reflected even in the physiological behaviour of seeds developed from such treated plants. Fox (1969) selecting several members of Cruciferae, reported the stimulatory effect of BA on vegetative growth of plants. Leopold (1982) and Sen (1984) also concluded that most of the effects of cytokinins on growth and development are mediated through other hormones including

auxins and gibberellins. It was also reported that externally applied synthetic cytokinins enhanced the chlorophyll and protein biosynthesis, promoted the net assimilation rate and reduced the degradative processes such as senescence (Purohit, 1983). Sen (1984) also showed that cytokinin also defer the biosynthesis of ethylene and abscisic acid, Lurssen (1982, 84) reported that ethephon in various concentrations is readily absorbed by plant tissues and may continue to release ethylene endogenously on cent per cent basis for a longer duration. Selecting various cultivars of bean, he showed that the long lasting effect of externally applied ethephon on vegetative growth may be attributed to the promotion of the biosynthesis of abscisic acid. ABA itself when applied externally, is well reported to retard various parameters of vegetative growth (Milborrow, 1974).

Through, the mode of action of morphactins in affecting various aspects of vegetative and reproductive growths is not clearly understood, their are evidences to indicate that morphactins function as the antagonists of endogenous gibberellins, cytokinins and auxins (Corcoran, 1975). Singh (1979), Singh and Murty (1983) also reported some long lasting effects of morphactins (CME) on some correlative growth phenomena in solanaceous crop plant.

Zeevaart (1978) made extensive studies on the effects of externally applied plant hormones on flower initiation and development, fruit and seed setting in several plants. The mechanism by which the endogenous level of plant hormones in relation to flowering, is altered by the application of plant hormones was also investigated in some species of Xanthium, soybean, sunflower, cotton, Pharbitis, Chrysanthemum, white musturd and tomato. Using tobacco cultivars, Bose and Harada (1970) reported that IAA in lower concentrations was promotory for flower formation and fruit setting. However, higher doses of the same proved to be inhibitory. This was also reported that the effect of auxin is mediated through cytokinins which are endogenously involved in flowering process (Tran-Thanhvan *et.al.*, 1974a). Tomato and Harada (1984) viewed that phytologists would have to conduct more intensive experimental investigations so as to determine the chemical nature of the flowering hormones. However, they have reported that the application of plant growth regulators at early stages of plant development in *Cichorium, Chrysanthemum, Pharbitis* and *Althnea,* switched on certain biochemical changes which persists upto reproductive stage and exhibit their effects on flowering and fruiting behaviour of plants. Singh (1979) and Murty and Singh (1983) using certain crop plants reported that

the enhancement of reproductive growth by NAA, GA_3 and BA treatments may be attributed to a reduction of drop (abscission) of reproductive unit and consequently the number of flowers and fruits retained is increased. Addicott (1981) also reported similar effects of aforementioned growth substances in the control of reproductive growth.

In present studies, tested concentrations of CME, higher concentrations of ABA and ethephon seemed to act as the antagonists of endogenous growth promoters involved in flowering and fruiting processes. Corcoran (1975), Durley (1983) and Parups (1983) also reported such behaviour of morphactins. Milborrow (1974), Singh (1979) and Addicott (1981) also observed that externally applied higher concentrations of ABA and ethephon are always inhibitory for the reproductive growth. Lieberman (1979) mentioned that this action of ethylene was primarily due to a block of cell division and DNA synthesis in meristematic tissues.

The effect of externally applied plant growth substances on therapeutic yield of few species has been investigated by some workers. The reduction of alkaloids content by physiologically active concentrations of growth promoters is attributable to their tendency to enhance the normal metabolism of plants (Sircar, 1971). Chatterji *et.al.*, (1979) with *Solanum* and certain other medicinal plants, Biesboer and Mahlberg (1979) with *Euphorbia* sp. and Basu and Chakravarti (1984) with *Solanum khasium* (an important medicinal plant) also reported similar effects of IAA, NAA, 2, 4-D, GA_3, KN and BA on alkaloids content (secondary products) of the plants. However, the pharmaceutical yield by lower concentrations of NAA, GA_3 and BA could be enhanced because the dry weights of economic parts were promoted by these treatments. The promotory effect of morphactin, ABA and etheral on the therapeutic yield has also been reported by Gupta and Madan (1975) in *Datura* and Basu and Chakarvarti (1980) in *Vinca* Leopold (1982), Barendse (1983) and Sen (1984) remarked that the mechanism by which the metabolism of secondary plant products incuding alkaloids is regulated by externally applied or endogenously produced plant growth hormones, could not be understood so far, Bell (1980) expressed the view that plant hormones which retard the growth of plant parts, enhanced alkaloid biosynthesis by diverting certain protein and non-protein amino acids towards a 'biway' leading to elevated alkaloid biosynthesis. He also concluded that some part of the accumulated alkaloids may come through protein degradation process under certain hormonal treatments and treatments with certain conditions of environmental stresses

(interpreted in next chapter). Though the biochemical and molecular biology of responses to PGRs applications is yet to be understood, there are substantial and convincing evidences to mention that PGRs application are effective convenient and economically viable for booting the plant productivity (Malik, 1995; Singh and Singh, 1995; Kakralya *et.al.*, 2003). Rodemacher (2000), Mok *et.al.*, (2001), Buhan (2000) and Mahala (2002) also advocated the practical utility of such investigation in enhancing plant productivity under normal and stress environments.

□□□

Interactions of Growth Regulators and Stresses in Modulating Economic Yield and Phytomass Productivity

Environmental stress adversely affects the productivity of plants directly by altering the biochemical pathways of metabolism (Levitt, 1980-81; Larcher, 2002). The deteriorative effects of simulated moisture, thermal and radiation stresses on plant productivity and economic yield have been discussed in Chapter 10 of this book. Tumonov (1969) reported that damages caused by drought and frost may be deferred or reduced by simultaneous or sequential applications of some synthetic auxins. Later on Abdulla and Verkerk (1970) and Hicks and Stricker (1972) also observed that adverse effects of high temperature and radiation stresses may be counteracted partially or completely by the application of some growth regulators. Papadakies and Gx (1978) quoted several examples where plant resistance to multi-adversity conditions was corroborated by the use of growth regulators. Earlier to this, Witwer (1971), Arnon (1975), Ashour (1977), Alden and Hermann (1977) had also reported similar results. Therefore, Papadakies and Gx (1978) and Levitt (1980-81) and Larcher (2002) suggested the screening of growth substances for lowering down or mitigating the adverse effects of environmental stresses on plant growth, development, productivity and yield.

Keeping in view the suggestions of these workers, an effort was made to study the interaction of plant growth substances and environmental stresses in affecting the plant productivity in medicinal plant materials. One more objective of such investigation was to work out the most appropriate combination of water and temperature stresses with growth substances suitable for increasing the therapeutic

yield of plants. The undertaking of such studies was recently suggested by Singh *et al.*, (2000); Sankhla (2002) and Mahala (2002) for medicinal plants.

Experimentia

Seedlings were obtained by germinating seeds under ordinary laboratory conditions in large and deep enamel trays which were three quarter filled with white sand moistened with distilled water at regular intervals. When seedlings were 15 days old, these tray were watered with fix amount of PEG 6000 solution of –5.0 bar external water potential for the simulation of moisture stress for 2 cycles of 24 hours each with an intermission of 48 hours of ordinary watering. Procedure was same as described earlier in Chapter 8. Similarly another group of 15 days old seedlings was subjected to high temperature stress (40°C) by keeping them in BOD incubator for 2 cycles of 6 hours each with a gap of ordinary temperature for 2 hours when watering was done so as to avoid dessication of seedling. Out of these two groups of seedlings (one treated with moisture stress and another with high temperature stress) uniform size seedlings were transplanted in earthen pots kept in a protected place as described in Chapter 8. Established plants were thinned to uniform height. These plants were spray treated with one most effective (determined by earlier experiments, Chapter 11) concentration of NAA (100 mg/l), GA_3 (100 mg/l), BA (20 mg/l), CME (10 mg/l), ABA (10 mg/l) and ETH (200 mg/l) at the stage when 5th leaf was fully unfolded (20 days in *T. purpurea* and 25 days in *W. somnifera* after transplantation). All these spray treatments were again repeated when the flower formation was just initiated. Experimental design, method of investigations on plant productivity and economic yield were the same as described in Chapter 11.

Experimental Observations

Inferences regarding the apparent and probable interactions of externally applied plant growth substances with moderate moisture and high temperature stresses in present studies, are based on the comparative analysis of data obtained for total fresh weight, dry weight of the whole plant at maturity stage, fresh and dry weights of economic parts of the species and harvest index. Effect of growth hormones on the therapeutic yield per unit dry weight of economic parts and the pharmaceutical yield was also investigated on the plants which had been subjected to moisture stress and temperature stress at the seedling stage.

It was observed that moisture stress of –5.0 bars (external

water potential order) remarkably reduced the plant productivity. However, the therapeutic yield was enhanced by this level of moisture stress in both the species as compared to their respective untreated controls 100 mg/*l* concentration of NAA reduced the deteriorative effect of moisture stress on productivity in both the species. It was interesting to note that the pharmaceutical yield in *W. somnifera* where roots constitute the economic part, was augmented by NAA treatment upto such extent that it exceeded even the 'control'. However, in *T. purpurea* plant productivity and economic yield were drastically reduced by NAA + moisture stress treatment (Table 12.1). Gibberellin and benzyl adenine have also shown moisture stress antagonizing action on productivity and economic yield in both the species.

CME exhibited synergistic action with the moisture stress and stimulated a loss of productivity and economic yield in both the species. The pharmaceutical yield was consequently reduced by a combination of CME with moisture stress. Contrary to the total productivity and pharmaceutical yield, the therapeutic yield was remarkably enhanced by CME treatment in plants pretreated with moisture stress. ABA and ethephon have further retarded the productivity and pharmaceutical yield of moisture stressed plants. ABA (10 mg/*l*) was found to be more effective than ethephon (200 mg/*l*) was found to be more effective than ethephon (200 mg/*l*) in synergising the deteriorative effect of moisture stress in combination with the later. However, the therapeutic yield was considerably enhanced beyond the level of stressed plant in *W. somnifera*. In case of *T. purpurea*, abscission of most of the vegetative and reproductive parts took place at some or the other stage of their development (Table 12.1) when treated with a combination of moisture stress and ethephon.

The high temperature stress reduced the dry matter accumulation and total pharmaceutical yield to a greater extent than the moisture stress in both the species. However, the therapeutic yield was enhanced by high temperature stress in both the species (Table 12.2). NAA, BA and GA_3 showed similar sort of interactions with temperature stress, the adverse effect of which could be successfully counteracted by such treatments. However, in *T. purpurea* NAA treatment did not allow the reproductive parts to develop leading to a total loss of pharmaceutical and therapeutic yields. 20 mg/*l* of BA was found to be more effective than 100 mg/*l* of GA in reducing the adverse effect of temperature stress. CME, ABA and

Table 12.1

Plant productivity and economic yield attributes as affected by combined treatments of moisture stress and plant hormones in *W. somnifera* and *T. purpurea*

Treatments conc. mg/*l* + MS (–5 bar)	Total weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Thera-peutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
W. somnifera							
Control (P)	12.500 a	4.600 a	3.300 a	1.650 a	0.354 a	200 a	330 a
Control (MS)	7.410 –c	2.620 –b	2.110 –b	0.900 –b	0.346 –b	300 c	270 –c
NAA 100 + MS	11.750 –b	4.110 a	3.210 a	1.590 a	0.387 b	220 b	349 b
GA_3 100 + MS	13.900 b	5.400 b	4.820 b	2.300 b	0.426 c	180 –b	414 c
BA 20 + MS	14.750 c	6.310 c	5.430 c	2.800 c	0.444 c	150 –c	420 c
CME 10 + MS	5.800 –c	2.120 –d	1.750 –c	0.810 –c	0.380 b	325 d	260 –c
ABA 10 + MS	4.540 –d	2.110 –d	1.900 –d	0.700 –d	0.333 –c	350 e	245 –d
ETH 200 + MS	4.800 –d	2.400 –c	2.100 –e	0.800 –c	0.381 b	360 e	288 –b
T. purpurea							
Control (P)	18.800 a	7.950 a	2.700 a	1.900 a	0.238 a	310 a	294 a
Control (MS)	11.500 –b	4.910 –b	1.110 –b	0.850 –b	0.173 –b	400 b	170 –c

Contd....

Contd....

Treatments conc. mg/l + MS (–5 bar)	Total weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Thera-peutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
NAA 100 + MS	8.150 –c	3.910 –b	0.0 –d	0.0 –d	0.0 –d	0.0 –e	0.0 –e
GA_3 100 + MS	21.450 b	9.110 b	4.210 b	3.100 c	0.340 c	250 –c	387 b
BA 20 + MS	24.340 b	9.810 b	4.900 c	3.500 d	0.350 d	240 –d	420 c
CME 10 + MS	9.520 –c	4.120 –b	0.850 –c	0.600 –c	0.146 –c	410 b	123 –d
ABA 10 + MS	7.850 –d	3.530 –c	0.0 –d	0.0 –d	0.0 –d	0.0 –e	0.0 –e
ETH 200 + MS	6.130 –e	3.150 –c	0.0 –c	0.0 –d	0.0 –d	0.0 –e	0.0 –e

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level according to Duncan's multiple range test.

* Data for therapeutic yield for *T. purpurea* are on per two grams dry weight (of economic part) basis. Data on productivity are average of 5 replicates of 5 units each.

Therapeutic yield—of 5 replicates of single unit each. MS = Moisture stress.

Table 12.2

Plant productivity and economic yield attributes as affected by combined treatments of Temperature stress and plant hormones in *W. somnifera* and *T. purpurea*

Treatments conc. mg/l + TS (40°C)	Total weight / Plant (gm)		Weight of Economic Part/Plant (gm)		Harvest index	Thera-peutic yield/unit* gm dry wt. (mg)	Pharma-ceutical yield/ plant economic part (mg)
	FW	DW	FW	DW			
W. somnifera							
Control (P)	12.500 a	4.600 a	3.300 a	1.650 a	0.354 a	200 a	330 a
Control (TS)	6.150 –b	2.310 –b	1.250 –b	0.710 –b	0.304 –c	300 c	210 –b
NAA 100 + TS	14.740 b	4.910 b	3.100 a	1.750 b	0.357 a	240 b	420 b
GA_3 100 + TS	14.730 b	5.320 c	3.850 b	2.100 c	0.390 b	210 a	441 c
BA 20 + TS	15.440 c	4.800 d	4.460 c	2.500 d	0.430 c	190 –b	475 d
CME 10 + TS	5.870 –c	2.100 –c	1.160 –b	0.650 –b	0.307 –c	320 d	208 –b
ABA 10 + TS	5.330 –c	1.900 –d	0.910 –c	0.480 –d	0.305 –c	330 d	191 –c
ETH 200 + TS	5.420 –c	1.950 –d	0.950 –c	0.610 –c	0.315 –b	325 d	198 –c
T. purpurea							
Control (P)	18.800 a	7.950 a	2.700 a	1.900 a	0.238 a	310 a	294 a
Control (TS)	8.160 –b	3.800 –b	0.520 –c	0.410 –c	0.107 –b	400 b	82 –c

Contd....

Contd....

Treatments conc. mg/l + TS (40°C)		Total weight / Plant (gm)				Weight of Economic Part/Plant (gm)				Harvest index		Thera-peutic yield/unit* gm dry wt. (mg)		Pharma-ceutical yield/ plant economic part (mg)	
		FW		DW		FW		DW							
NAA	100 + TS	7.180	–c	3.100	–c	0.0	–d	0.0	–d	0.0	–c	0.0	–c	0.0	–d
GA_3	100 + TS	21.500	b	8.500	b	4.750	b	2.900	b	0.340	b	300	a	435	b
BA	20 + TS	24.350	c	10.210	c	5.260	c	3.400	c	0.333	b	280	–b	476	c
CME	10 + TS	7.150	–c	2.900	–d	2.140	–b	1.200	–b	0.401	c	430	c	285	–b
ABA	10 + TS	6.150	–d	2.800	–d	0.0	–d	0.0	–d	0.0	–c	0.0	–c	0.0	–d
ETH	200 + TS	6.400	–d	2.130	–e	0.0	–d	0.0	–d	0.0	–c	0.0	–c	0.0	–d

Any two means in the same column indexed by the same letter are not significantly different from each other at 5% level according to Duncan's multiple range test.

* Data for therapeutic yield for *T. purpurea* are on per two grams dry weight (of economic part) basis. Data on productivity are averages of 5 replicates of 5 units each.

Therapeutic yield data are averages of 5 replicates of one unit each.

TS = Temperature Stress.

ETH have shown sinergistic effects with temperature stress. CME, ABA and ETH have shown sinergistic effects with temperature stress in both the species in reducing the plant productivity which was reduced to a level lower than the plants which were treated only with temperature stress. *T. purpurea* was found to be more susceptible then *W. somnifera* to a combined action of temperature stress with ABA and ETH both (Table 12.2).

Interpretations and Application

Selecting bean, cotton, tomato and various varieties of garden pea and some cultivars of potato, Levitt (1981), reported that after germination, seedling growth is the most susceptible stage of plants which are adversely affected by water and temperature stresses given to the seedlings at the early stage. It was further shown that the pace of metabolism, once disturbed by externally applied environmental stresses is maintained throughout the life of the plant and considerably effects the yield and productivity. The effect of water and high temperature stresses on the hormonal content of plant parts was also reported by (Larcher, 2002; Mahala, 2002; Nilsen and Orcutt, 1996), however, indicated that very few growth substances have been tried as the protective regulators against the water and temperature stresses. Buhan (2000) using many plants as experimentalmaterials reported that CKs function as the protective substance against water and high temperature stresses. He also pointed out that externally applied growth substances including gibberellins and auxins lower down the damages caused by environmental stresses and increases the plant tolerance to environmental stresses. Galston *et.al.*, (1963) with pea, Moore (1970) with wheat and Kaufman with some grasses also reported that synthetic auxins and some gibberellins may be tested as the protective substances against the moisture and temperature stresses. Leopold (1982) has however, cautioned to undertake systematic and intensive investigation before the practical utility of such substances could be established. Milborrow (1981) mentioned that ABA and ethephon in physiologically active concentrations enhanced the effects of water stress and high temperature stress by reducing the endogenous levelof natural cytokinins. This reduction of cytokinins level permits a depression of various ongoing biochemical processes and consequently growth and productivity are reduced. The effect of morphactins in augumenting the deteriorative effect of simulated moisture and temperature stresses is interpretable on the basis of findings of Corcoran (1975) and Parups (1983) who have interpreted that most of the morphactins when applied externally, acted as the

antagonists of endogenous gibberellins and cytokinins and consequently the level of ABA is increased. This increased ABA level (and also the ethylene) was considered to be responsible for aforementioned effects of morphactin. Lieberman (1979), Milborrow (1981), and Levitt (1980-81) also expressed the view that drought and temperature stresses induce the biosynthesis of abscisic acid and ethylene which caused an irreversible and unrepairable loss to the seedling growth and also cumulatively to the subsequently developed plants which indicated the suppression of their growth and developmental processes. Observations of Shimokowa (1984), Lurssen (1984) and Ludwig (2000) also support the findings of present studies. As alkaloids are secondary plant products and they are usually synthesized under unfavourable or stressed conditions, the therapeutic yield showed opposite trend than the pharmaceutical yield. This means that the treatments which usually promote the growth and productivity, adversely affect the total alkaloids content on per unit dry weight basis. Similar results were also obtained by Bell (1980) in a number of plant materials. However, the actual mechanism by which applications of growth regulators promote or reduce the adverse influences of environmental stresses is yet, to be worked out (Sen, 1984; Singh and Afria, 1985; Judwig, 2000; Buhan, 2000; Sankhla, 2002; Mahela, 2002). Similar findings of some other workers also support the view of Singh *et.al.*, 2000) and Jobhar *et.al.*, (2003) who advocated that plant growth regulators and their commercial formulation may be practically applied for enhancing the economic yield and phytomass productivity of medicinal plants. Some new PGRs (including polyamines, pachlobutrazol, pauryl, mixtalol etc.) should be experimented on medicinal plants keeping in view the concepts of plant biodiversity conservation and development.

□□□

Soil Conditions and Productivity of Medicinal Plants

Out of the environmental factors, which influence plant productivity and economic yield, soil conditions are of paramount importance (Misra, 1980). He indicated that the heterogeneity in the texture, structure and composition of the soil impart some specific physical and chemical characteristics to the soil which affect the growth behaviour of plants. Kawakami (1978) showed that physiological aspects of plants are affected by soil characteristics in more than one ways. Pandeya *et.al.*, (1968) and Barlow *et.al.*, (1976) also realized the importance of the study of soil in relation to the parameters of growth and development including germination, seedling growth, vegetative growth, flowering, fruiting and productivity of plants. Murthy and Hirekerier (1980) reviewed the literature on soils of India and pointed out that these may be conveniently categorized into few classes on the basis of dominant size of soil particles comprising the soil. These soil types are clay, silt, sand (very fine, fine, medium and coarse sands) and loam. These soil types exhibit considerable variations in supporting the plant growth and yield (Misra, 1980). Therefore, the study of most suitable soil type for a particular species of plants is very important specially for medicinal plants intended to be cultivated (Schroder, 1998). Keeping in view the importance of such investigations, present studies were therefore, conducted taking into account the principal soil types in which the present experimental plant materials are reported to grow in nature.

Experimentia

Various types of soil were collected from different locations

in the vicinity of the institution. These were brought to laboratory, powdered and oven dried so as to remove moisture from each of them. The humus was procured from a progressive farmer who prepares the well decomposed compost manure from cow dung. This was also powdered and dried as usual. Garden soil was taken from experimental garden of the institution and oven dried. This garden soil was considered as the control (Table 13.1)

Table 13.1

Characteristics of garden soil which was used as 'control' (Average two years data)

(A)	*Mechanical Properties*	
	(a) Sand	
	(i) Coarse Sand	9.40%
	(ii) Fine sand	53.00%
	(b) Silt	10.25%
	(c) Clay	27.86%
	(d) Texture	Sandy loam
(B)	*Chemical Properties*	
	(a) Total N (%)	0.048 (%)
	(b) Available Phosphorus	0.00055 (%)
	(c) Available Potassium	0.0047 (%)
	(d) Organic Carbon	0.516 %
	(e) pH	7.15
	(f) EC (mm mhos/cm at 25°C).....	0.676

Preparation of soils for experimental purpose, their categorization and filling in equal amount in large earthen pots upto a uniform height in each pot, was practised following the standard technique (Thompson, 1957). Uniform sized and unsoaked seeds of experimental plant materials were sown at uniform depth and distance in each pot. Each pot was having 20 seeds and each treatment of all the soil types consisted of 10 such pots. Pots were watered on 3 days intervals till the experiment were terminated. The number of seeds that germinated (criteria being 1.5 cm seedling emergence from soil surface) was observed. The number of seedlings that survived out of total germinated seeds in each pot was also counted. These data were transformed into percentages. Eighteen days after seed sowing, about half the number of 'in situ' survived seedlings with approximately uniform size was transplanted in other pots having soils of similar types and the percentage of

seedlings which could survive and got established after the transplantation, was also determined. Five plants were randomly selected out of those survived from each of the soil types at maturity stage and data on various aspects of productivity and economic yield were recorded following the method already described (Chapter 10).

Experimental Observations

With respect to germination as determined by seedling emergence, clay soil proved most unsuitable for both the species. *W. somnifera* showed maximum seed germination in sand while the peak value of seed germination of *T. purpurea* was obtained in a 50 : 50 sand + garden soil mixture (Table 13.2). However, only a very poor percentage of seedlings 'in situ' could survive under pure sand in both the species. It was interesting to note that maximum survival percentage of 'in situ' seedlings was obtained under humus conditions but this value was not significantly higher than recorded for sand and garden soil mixture in *T. purpurea*. No seedling transplanted in sand could survive inspite of maximum care in either species. Sand and garden soil mixture proved to be most supportive for the survival of transplanted seedlings followed by humus in *W. somnifera* as well as in *T. purpurea* (Table 13.2; Fig. 13.1).

The dry matter accumulation was found to be maximum in plants which were retained in sand + garden soil mixture and humus. The clay soil was observed to be least supportive to the accumulation of dry matter in the whole plant as well as in economic parts of both the species (Table 13.2). The maximum therapeutic yield was obtained from clay soil grown plants of both the species and the yield of total alkaloids content in terms of therapeutic yield was lowest in humus. However, the total pharmaceutical yield was obtained from plants retained in garden + sand mixture. The most unsuitable soil with reference to total pharmaceutical yield in present investigations proved to be humus for *W. somnifera* and clay for *T. purpurea* (Table 13.2; Fig. 13.1).

Interpretations and Applications

Patten and VanDoren (1970), selecting corn as the experimental material in green house conditions, showed that seed germination and seedling emergence were considerably higher in sandy soil as compared to compact clay soil. Parihar and Chaudhary (1977) concluded that poor germinability of crop plants under compact soil may be attributed to the imposed hypoxial condition (lack of O_2). Both the plant materials in present investigation proved to be

Table 13.2

Effect of soil types on germination, seedling survival, plant productivity and economic yield in *W. somnifera* and *T. purpurea*

Soil Types	Germination %	Seedling survival		Total wt./ Plant (gm)		Wt. of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/ unit* gm dry wt. (mg)	Pharmaceutical yield/ plant economic plant (mg)
		In situ (%)	Transplantation (%)	FW	DW	FW	DW			
W. somnifera										
Control (Field)	40.7	75.4	60.8	13.580	5.410	3.250	1.670	0.310	200	334
Humus	52.4	85.7	69.7	18.620	8.410	4.300	2.180	0.260	120	261
Sand	61.3	20.7	—	—	—	—	—	—	—	—
Clay	28.4	46.8	20.8	8.470	3.900	2.850	1.480	0.380	240	355**
Sand + Garden soil	54.7	80.4	73.3	15.800	7.800	4.950	2.450	0.340	160	392
L.S.D.	3.540	4.65	2.16	1.950	1.110	0.350	0.190	—	10.3	29.6

Contd....

Contd....

Soil Types	Germination %	Seedling survival		Total wt./ Plant (gm)		Wt. of Economic Part/Plant (gm)		Harvest index	Therapeutic yield/ unit* gm dry wt. (mg)	Pharmaceutical yield/ plant economic plant (mg)
		In situ (%)	Transplantation (%)	FW	DW	FW	DW			
T. purpurea										
Control (F)	31.4	60.4	49.3	21.080	7.800	4.330	2.640	0.300	310	396
Humus	43.7	70.9	50.5**	26.200	9.800	6.820	3.220	0.400	210	451
Sand	54.1	37.5	—	—	—	—	—	—	—	—
Clay	17.4	56.4**	19.8	15.20	5.100	1.875	1.220	0.240	400	240
Sand + Garden soil	69.8	67.3	50.5**	25.100	9.900	5.150**	3.380	0.360	270	469
L.S.D.	4.65	5.8	3.4	1.450	1.800	1.790	0.530	—	15.4	26.8

Each figure is secondary average of 10 replicates of 20 seeds each for germination. Other figures are secondary averages of 5.

* Data for therapeutic yield for *T. purpurea* are on per two gram dry weight (of economic part) basis.

** Non-significant at 5% level of significance.

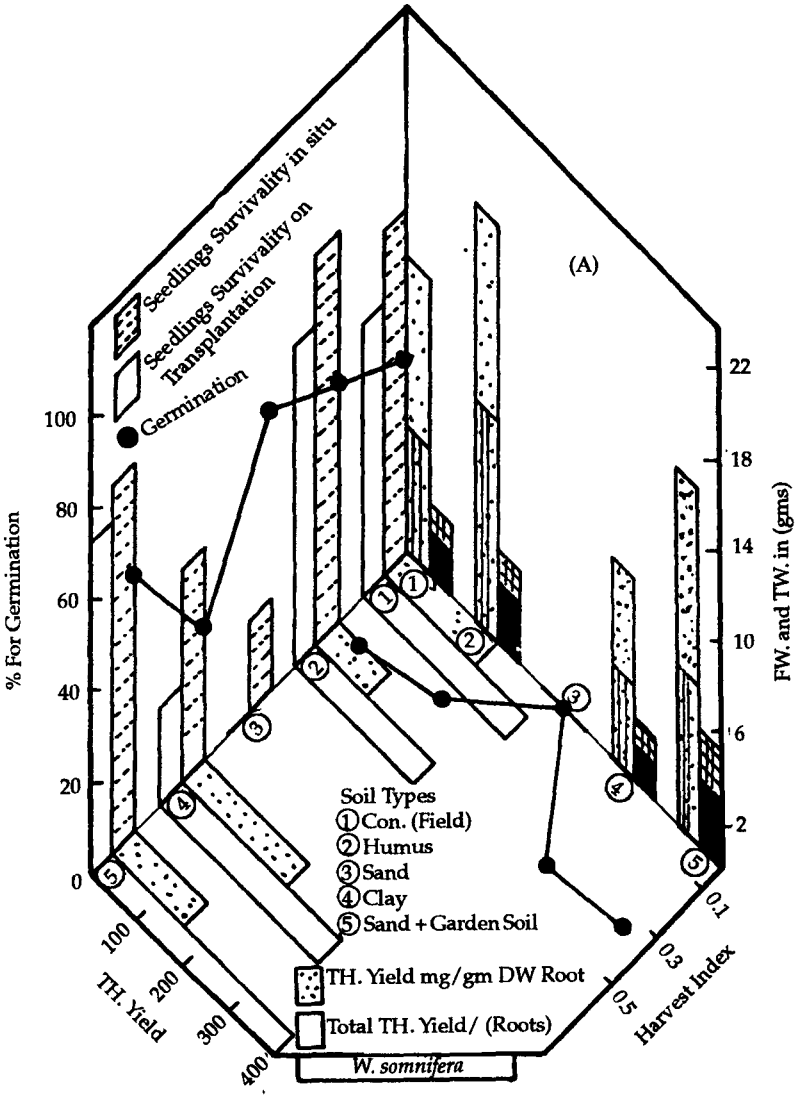


Fig. 13.1 A. Plant Productivity and Economic Yield as Affected by Various Soil Types in *W. somnifera*

Indices in Fig. No. 13.1A & B are common

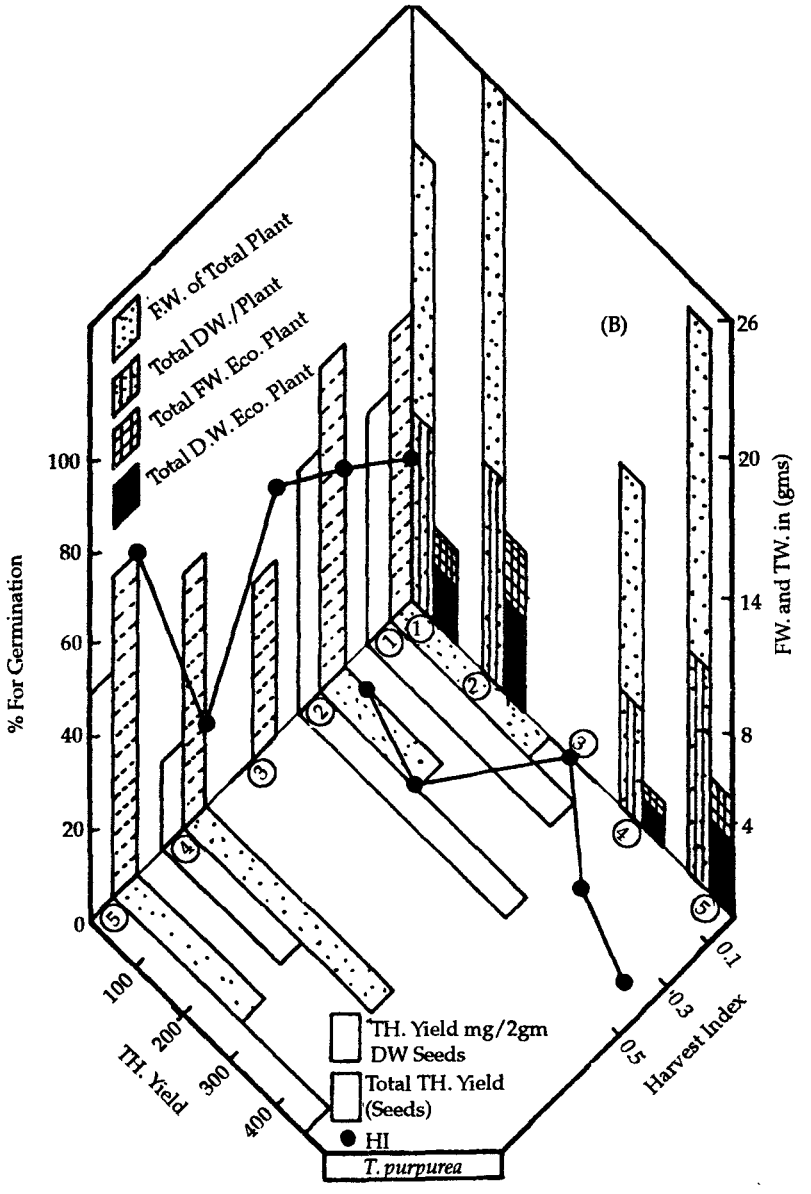


Fig. 13.1 B. Plant Productivity and Economic Yield as Affected by Various Soil Types in *T. purpurea*

Indices in fig 13.1 A & B are common

sensitive to low oxygen tension during germination and early seedling growth. Bhargava *et.al.*, (1979) also obtained similar result in *Capsicum annuum*. However, the poor 'in situ' survival percentage of the seedlings in sand appears to be due to the poor water holding capacity of sand. Jensen *et.al.*, (1972) also obtained similar results with various types of soil and seedlings of various forage crops.

Humus and sandy loam (sand + garden soil) supported most favourably the survival of 'in situ' seedlings as well as those transplanted in the same type of soil presumably due to a suitable proportion of moisture retention and nutrients. Townsend (1972) with various cultivars of gram, Mehta and Parihar (1973) with cotton and soybean also obtained similar results. The pure sand was unable to support further growth and development of plants of both the experimental species, probably due to the increased moisture stress and also due to the nutritional stress. The adverse effect of too much porous soil (sandy soil) on plant productivity and yield are attributable to a combined action of moisture and nutritional stresses. Taylor and Ratliff (1969) in cotton and groundnut also obtained similar results. These workers have shown that such adverse conditions hamper the growth rates of the roots leading to reduced biosynthesis of cytokinins which are, otherwise, essential for normal growth and development. Govil *et.al.*, (2002) also reported the results of studies on the germination, seedlings growth and survival, vegetative growth, plant productivity and yield under various types of soil in several crop plant species including wheat, rice, barley, jute, pea and cotton. On the basis of present investigation, it was realized that sandy loam soil is most suitable for the cultivation of both these species. Data obtained for the distribution of these species in various parts of India, also support the aforementioned findings. (Sarin, 2003). Farooqi and Sreeramu (2001) with Babchi, Datura, Ipecac and Senna and Jakhar *et.al.*, (2003) within Aloe, Isabgol, Opium and Basil also demonstrated the correlative effects of soil conditions on plant phytomass and economic yield. In their natural habitats, both these species show better acclimation index under sandy loam soil and in such soil condition the alkaloids are synthesized with more pace. And presumably it is because of these facts that Ashwagandha Nagauri and Red Tephroli and prefered in International trade.

□□□

Application of Macronutrients for Enhancing the Production Potential of Medicinal Plants

Out of 17 well established mineral nutrients which are essential for normal plant growth and development, three macronutrients viz. nitrogen, phosphorus and potassium, are considered most important (Lauchii, 1983). On the basis of extensive investigations, it has been repeatedly emphasized that the requirements of plants for these elements quantitatively differ from species to species (Larcher, 2002). The study of the critical level of N, P and K is, therefore, of vital importance for successful plant cultivation. Atal and Kapur (1982) and Farooqi and Shreeramu (2001) realized that the mineral requirements of most of the medicinal plants remained unexplored and only a few reports are available indicating the agronomic need of medicinal plants for N, P and K (Jakhar *et.al.*, 2003).

Kaul and Jutshi (1982) with some solanaceous species of medicinal value, Sobti and Kaul (1982) with *Datura* and Gulati *et.al.*, (1982) with belladona studied the effect of N, P and K on growth, productivity and active principles including alkaloids. It was indicated that the dose of the fertilizers, suitable time and mode of application, vary from crop to crop and such studies are of paramount importance for successful cultivation of medicinal plants (Sarin, 2003).

Keeping in view the above mentioned facts, the present investigation were undertaken to study the effect of N, P and K on productivity and economic yield of the species under field conditions.

Experimentia

The four sets of earthen pots filled with preweighed and sundried garden soil mixed with 50% white sand were arranged for different treatments of N, P and K at different stages of plants. 50

seeds were sown in each earthen pot at the uniform depth. In one set, different doses of N, P and K were mixed with soil as the basal dose. Ammonium nitrate was used as the nitrogen source while P_2O_5 (single super phosphate) for phosphorus and K_2O (murate of potash) for potassium were used as sources. In second set of pots, fertilizers were applied in soil when the plants were 20 days old. In the 3rd set N, P and K were mixed with pot soil when plants were 35 days old. All these plants were raised as untransplanted. In the final set of experiment. N, P and K fertilizers were sprayed on plants following the method used by Saroha (1981). Other cultural practices were the same as described in Chapter 10.

Experimental Observations

(A) Germination Responses to N, P and K Applications

As is evident by seedling emergence, the germination behaviour of plants was found to be variably influenced by different doses of N, P and K. The basal dose of nitrogen progressively promoted the germination percentage in both the species with increasing doses. However, highesttested dose of nitrogen reduced the germination in *W. somnifera*. The germination was also promoted by the soil application of phosphorus with increasing doses in both the species and the treatment with phosphorus was found more effective than nitrogen treatment for germination process in *W. somnifera*. It was observed that at lower doses of potash, the germination was enhanced while higher doses reduced the germination in both the species. It was further observed that 10 gm dose of potassium proved to be most effective in both the species with reference to germination. Seed germination percentage under this treatment even exceeded the 100 gm doses of nitrogen and phosphorus (Table 14.1 and Fig. 14.1). As the CDY was harvested in the form of roots in *W. somnifera* and the same was obtained from seeds in *T. purpurea*, both the species revealed considerable differences in there responses to treatments with N, P and K. Data for yield responses of *W. somnifera* are presented in Table 14.1 while data for such observation for *T. purpurea* have been represented by Fig. 14.1.

(B) Yield Responses to N, P and K Applications in W. somnifera

It was observed that the crude drug yield was improved by all the doses of nitrogen irrespective of the time of the application of nitrogen. 50 gm dose of nitrogen proved to be most effective in all the treatments except the spray treatment where CDY was improved upto 100 gm level of nitrogen in *W. somnifera*. The total pharmaceutical yield was significantly promoted by the nitrogen

Table 14.1

Effect of various chemical fertilizers on germination and plant productivity in *W. somnifera*

Treatment doses (gm)	Germination (%)	Seed dressing			Broadcasting (20 days)			Broadcasting (35 days)			Spray treatment (35 days)		
		CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)
Control	40.4	1.800	200	360	1.800	200	360	1.800	200	360	1.800	200	360
Nitrogen													
5	43.7*	2.710	160	432	2.300	145	335	2.100	155	325	1.950*	165	321
10	47.6	3.810	120	456	2.920	116	319	2.550	130	331	2.410	145	348
50	53.4	4.430	100	443	3.410	90	306	2.890*	115	332	2.750	130	351
100	30.5	1.910	160	304	2.250	120	270	2.400	130	312	2.980	240	715
L.S.D.	3.41	0.210	14.4	22.4	0.310	28.4	24.3	0.215	15.7	23.3	0.150	28.3	21.2
Phosphorus													
5	45.6	2.910*	175	508	2.650	150	397	2.310	160	368	1.850*	165	305*
10	49.4	3.400	150	510	3.100	135	418	2.800	150	420	2.200	155	341
50	56.4	3.910	130	507	3.410	110	375	3.220	130	418	2.900	135	391
100	62.7	4.420	110	486	3.900	80	351	3.510	120	421	3.100	120	372
L.S.D.	4.210	0.150	18.4	31.3	0.420	34.3	26.4	0.175	22.4	21.2	0.110	23.5	27.3

Contd....

...Contd.

Treatment doses (gm)	Seed dressing				Broadcasting (20 days)			Broadcasting (35 days)			Spray treatment (35 days)		
	Germination (%)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph yield/ economic part (mg)	CDY DW (gm)	Th. yield/ gm dry wt. (mg)	Ph. yield/ economic part (mg)
Potassium													
5	49.6	2.850*	190*	541	2.540	160	406	2.130	170	362	2.100	165	346*
10	59.6	3.610	170	613	3.100	150*	465	2.800	160	448	2.410	155	379
50	31.3	4.800	280	864	4.300	190	817	3.700	180	666	2.750	170	459
100	19.4	2.700	320	864	4.600	240	1104	3.900	220	858	3.100	190	580
L.S.D.	3.12	0.120	12.8	27.4	0.428	21.4	21.3	0.175	14.4	27.3	0.190	21.5	23.5

Each figure is secondary average of 5 replicates (for germination) of 20 seeds each. For other aspects each figure is an average of 5 replicates.

* Insignificant.

Th. = Therapeutic yield; Ph.= Pharmaceutic yield; CDY = Crude drug yield.

treatments when the same were given as the seed dressing only and the spray treatment was not found much suitable with respect to total pharmaceutical yield. Applications of nitrogen at vegetative and preflowering stages drastically reduced the pharmaceutical yield in this species. The therapeutic yield was considerably reduced by nitrogen applications except the spray treatment of heavy doses (100 gm) which significantly promoted the therapeutic yield as well pharmaceutical yield.

It was observed that the CDY was promoted by all the treatments of phosphorus progressively. Most suitable mode of phosphorus application for maximum advantage (CDY) proved to be the basal given at the time of seed sowing. The pharmaceutical yield was constantly promoted by phosphorus treatments given at the time of seed sowing, at vegetative stage and also at preflowering stage. Under higher doses, the therapeutic yield was reduced by phosphorus treatment irrespective of the time and mode of application. The crude drug yield was also promoted by tested doses of potassium, basal dose being the most suitable. The total pharmaceutical yield was most favourably increased by potassium treatment. It may be concluded that potassium treatment proved to be most beneficial from pharmaceutical point of view in this species. Higher doses of potassium enhanced the therapeutic yield, the most favourable mode of treatment being seed dressing. It was found that potassium application at the time of seed sowing was comparatively crucial (cf. nitrogen and phosphorus) for increasing the CDY and pharmaceutical yield (Table 14.1).

(C) Yield Responses to N.P.K., and Applications in T. purpurea

It was observed that the CDY was promoted by various doses of nitrogen when applied either at seed sowing or vegetative stage or preflowering stage, 50 gm dose being the most suitable. Delaying the application of N in *T. purpurea* proved advantageous for enhancing the crude drug yield excepting the spray treatment with higher doses which reduced CDY. Pharmaceutical yield as well as the therapeutic yield, were promoted by nitrogen application but most suitable mode of treatment was the spray treatment with 10 gm dose (10 gms of ammonium nitrate dissolved in 1 litre water and sprayed on 20 plants planted in five pots) at the time of ready-to-flower stage (Fig. 14.1).

The CDY was promoted by phosphorus application in all the doses and at all the times of application, the most suitable, mode and time of treatment being the spray treatment at ready-to-flower

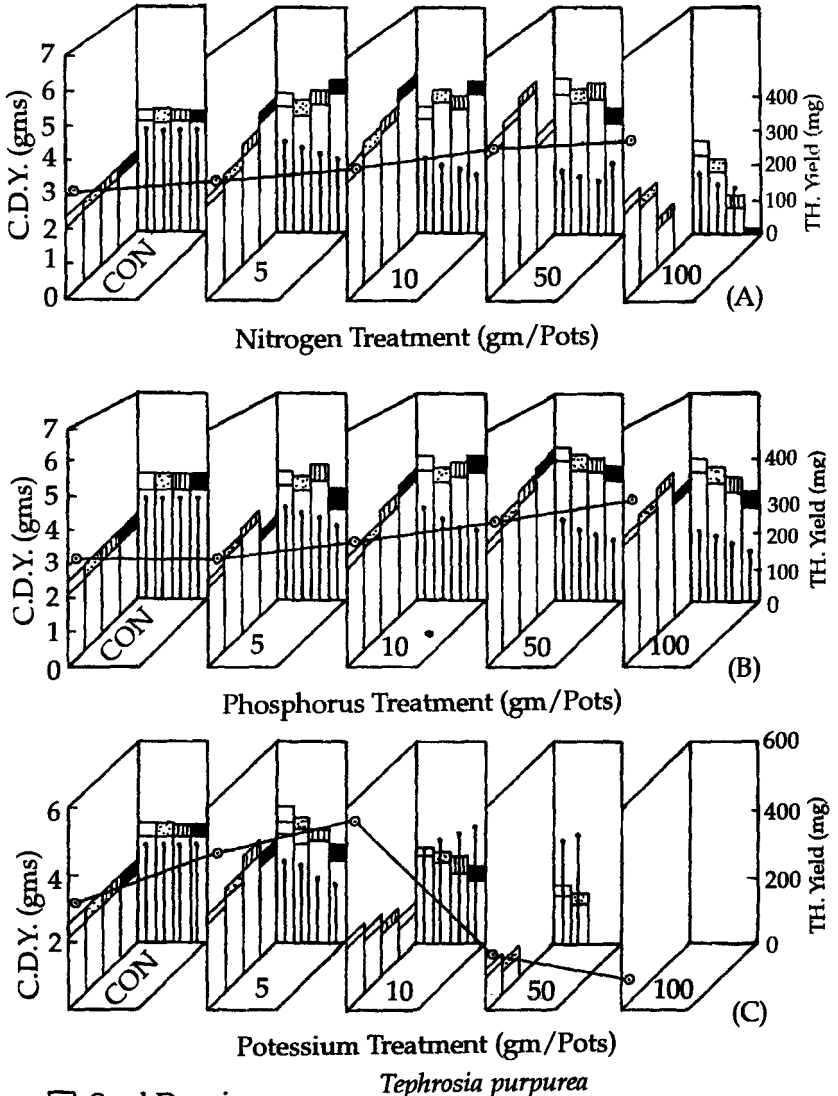


Fig. 14.1 Plant Productivity and Economic yield as Affected by N, P and K Applications in *T. purpurea*

stage. The pharmaceutical yield was also enhanced by phosphorus application but spray treatment was not found suitable from this point of view. The therapeutic yield was decreased by phosphorus application irrespective of time of application and mode of treatment. Doses of potassium proved deteriorative for yield attributes in this species.

Inter Pretations and Applications

The method employed in the present investigation seemed to be crude in the sense that the soil composition and the nutritional status of the soil could not be analysed before at nutrient applications. However, results obtained in these experiments have given preliminary information regarding the yield responses of experimental species to treatment with various doses of N, P and K at various stages of plant development. The improvement of seed germination and seedling emergence under moderate soil fertility conditions by the applications of adequate doses of nitrogen was also reported by Valish (1980) with wheat, barley and sugarbeet. However, the inhibitory effect of higher doses of nitrogen on seed germination may be attributed to the salt stress caused by the application of higher doses of nitrogen. The stimulation of such a stress in soil was also reported by Gupta (1977) who showed that presowing nitrogen application in higher doses delayed the seedling emergence and reduced germination percentage in bean, tomato and pepper leading to a poor cropstand density. Similarly, the higher doses of potash inhibited the seed germination and seedling emergence by stimulating a salt stress.

The adverse effect of the application of macronutrients on the alkaloids content per unit dry weight of the economic part may be attributed to the tendency of these macronutrients to favour the growth of the plants. It has been generally observed that the concentrations of macronutrients which retard the vegetative growth by causing the salt stress, proved promotory for alkaloids biosynthesis (Padulosi, 2002).

Ramanathan (1978) conducted detailed study of the effect of nitrogen on the alkaloids content of opium poppy and concluded that physiologically favourable doses of nitrogen reduced the alkaloids content in the cultivars of opium. Similar results were also reported for phosphorus applications in *Datura* by Afaq *et.al.*, (1979).

Thus, the result obtained in the present investigations supported the aforementioned findings and it may be emphasized that the application of N, P and K would be more profitable at the time of

ready-to-flower stage in *T. purpurea*. These application would be beneficial for farmers as well as for pharmaceutical industry.

Singh *et.al.* (2000), Singh *et.al.* (2003) and Jakhar *et.al.* (2003) using several plants of therapeutic significance demonstrated that excess fertilizers (and additional irrigations too) adversely affect the total alkaloid contents in economically important parts of medicinal plants. Therefore, only the judicious use of chemical fertilizers (and water) is most important requirement of the cultivation programme of medicinal plants. The efficacy of soil and folior applications of micro nutrients in the production of medicinal plants is yet to be established.

□□□

Export, Import and Marketing Scenario : Role of Environmental Biology of Medicinal Plants

Plants which possess curative properties have been employed for human welfare since the dawn of the civilization. WHO has compiled a list of 2000 medicinal plants used in various parts of the world but over 100 species have a consistently large demand in world trade of medicinal plants. In the present context of *"back to nature"* in health care, it is pertinent to conserve, domesticate and cultivate the plants so as to meet the increasing demand and also to reduce over dependence on wild resources. The international market of medicinal plants in over US dollars 60 billion per annum. This trade is increasing at the rate of 7%. India at present exports herbal materials and drugs to the tune of Rs. 446.3 crores only per year which can be raised upto Rs. 3000 crores by 2005 and 10,000 crores of rupees upto 2010 (targeted estimates). China and India are two global giants with respects to medicinal plants having about 40% biodiversity. China besides meeting its domestic requirements is earning US $ 5 billion per year from herbal trade. Therefore, ther is an emormous scope for India also to emerge as a major player in the global herbal market. However, this needs a comprehensive strategic plan embodying a hollistic view of the entire situation to boost the export upto Rs. 10,000 crores by 2010 with simultaneous efforts to minimize the import of medicinal herbs (Anonymous, 2000).

Scientific efforts in this direction must therefore include on equilibrated and interdisciplinary approach. In this co-ordinated approach basic, application oriented basic and applied aspects of

biology of therapeutically valuable plants must be investigated with the objective of domestication of plants, their cultural practices coupled with biodiversity conservation and developments. Some case studies reported in this book (treatise) are only a segment of the gamut. Singh *et.al.* (2003) Jakhar *et.al.* (2003) and Sharma (2003) reviewed the literature on these aspects in reasonable details Cracker and Simon (2002) and Hartmann *et.al.* (2003) also complied the literatures on propagational techniques for domestication and cultivation of medicinal plants. An outline of export, import and marketing of medicinal plants is described below :

Export of Medicinal Plants

The annual export of vegetable drugs is consistently increasing some data have been collected and presented in tables. However, figures available over the year have shown some degree of variations depending upon source. Table 15.1 indicates that export of Ayurvedic and Unani medicine was only Rs. 33.18 crores in 1991-92 which increased upto Rs. 54.74 crores in 1999-2000. The export of Homoeopathic medicines has shown a fluctuating trend.

Table 15.1

Export of vegetable drugs and alkaloids from India

Year	Ayurvedic & Unani Medicines		Homeopathic Medicines		Alkaloid	
	Quantity (in Tonnes)	Cost (Rs. lakhs)	Quantity (in Tonnes)	Cost (Rs. lakhs)	Quantity (in Tonnes)	Cost (Rs. lakhs)
1991-92	3360.56	3318.48	—	567.19	—	40.68
1992-93	4117.25	5022.43	—	164.56	—	68.17
1993-94	3112.34	5361.13	—	517.34	—	119.02
1994-95	4173.46	9338.94	—	180.02	—	115.01
1995-96	3716.29	9644.97	—	352.51	—	402.95
1996-97	12986.70	15503.58	56.32	128.53	154.80	179.39
1997-98	8939.52	6499.84	51.99	309.51	271.49	1989.65
1998-99	10898.79	7451.59	32.56	37.47	73.18	613.11
1999-2000	10399.20	5474.00	121.96	67.39	122.45	899.04

Source : Sharma, 2002.

The export of plant wise vegetable drugs has been summarized in Table 15.2 during 1995-96. The plant-wise export during 1999-2000 has been given in Table 15.3.

Table 15.2
Export of medicinal plant commodities from India, during 1995-96

Sl. No.	Item	Quantity (tonnes)	Value (in lakhs Rs.)
	Vegetable drugs		
1.	Bellandonna		
	Leaves	3.162	0.993
	Roots	1.000	0.268
2.	Galangal : rhizomes and roots including greater galangal	83.470	22.284
3.	Ginseng powder/chips	36.500	15.000
	Ginseng roots (others)	3072.110	1627.557
4.	Ipecac : dried rhizome and roots	3.866	4.347
5.	Liquorice roots	0.957	0.702
6.	Locust beans	7.200	48.551
7.	Poppy flowers and unripe dried heads poppy	6.551	4.577
	Poppy seeds	32.600	22.014
8.	Psyllium husk (Isabgul husk)	1686.230	14768.289
	Psyllium seed (Isabgul)	2258.230	672.720
9.	Sarsaparilla	1.100	0.305
10.	Senna leaves and pods	6279.771	1391.611
11.	Serpentina roots	3.937	3.198
12.	Tukmaria	216.220	48.826
13.	Unnab (Indian Jujube or Chinese dates)	29.450	13.444
14.	*Vinca rosea* (herbs)	191.924	64.990
15.	Zedovary roots	30.000	3.740
16.	Neem oil	151.729	217.481
	Total	**14096.007**	**18930.877**

Several organizations establishments are contributing in enhancing the export of medicinal plants from Indian Central Institute of Medicine and Aromatic Plants, Lucknow plays a pivotal role in boosting up the production of export oriented medicinal plants. Trade Development Authority of India regulates the qualitative and

Table 3
Export of medicinal plants during 1999-2000

Sl. No.	Plant Name	Export	
		Quantity (tonnes)	Value (in lakhs Rs.)
1.	*Glycirrhiza glabra*	70.39	81.93
2.	*Rauwolfia serpentina*	09.03	05.73
3.	*Panas ginseng*	1379.46	1015.14
4.	*Atropa pelladona*	22.74	8.19
5.	*Plantago ovata*	15295.31	10815.18
6.	*Swertia chirayata*	50.29	37.74
7.	*Cassio angustifolia*	7466.33	2254.20
8.	*Catharanthus roseus*	541.54	213.39
9.	*Hemidesmus indicus*	14.71	6.22
10.	*Juniperus communis*	0.80	0.79
11.	*Santalum album*	260.97	1759.62
12.	*Eucalyptus globules*	108.44	377.48
13.	*Myristica fragrans*	28.49	278.02
14.	*Artemissia pallens*	3.85	638.63

(Figures of other plants during 1999-2000 not available.)

quantitative aspects of export of medicinal plants. National Medicinal Plants Board, New Delhi has been establish to intensify the production and export oriented R&D activities throughout the country (Rawat, 2003). The board has identified 32 species of medicinal plants to be cultivated on priority basis for the purpose of domestic market as well as export purpose. These include Withania, Amla, Ashok, Aconite, Bael, Brahmi (*Bacopa monnieri*), Santalum, Swertia, Tinospora, Gudmar, Commiphora, Plantago, Jatamasi, Andrographis, Kolibari, Kokum, Kuth, Nightshade, Glycirrhyza safed musli (*Chlorophytum*), *Piper longum*, Daru haldi (*Berberis aristata*), Sarpgandha (*Rauwolfia*), Senna (*Cassia angustifolia*), Satawasi (*Asparagus racemosus*), Basil, Via Vidang (*Embelia rides*) etc. 25 State Medicinal Plants Boards have also been founded throughout the country so as to corroburate the efforts of NMPB. The details of export, import and marketing of medicinal plants may be obtained from NMPB as addressed below :

The Chief Executive Officer
National Medicinal Plants Board, Department of Indian Systems of Medicine and Homoeopathic, Government of India
Chandralok Building
36, Janpath, New Delhi,
Telephones : (011) 23319360, 23319255, Fax : (011) 23319360,
E-mail: nmpb22@indiatimes.com.

Import of Medicinal Plants

The pharmaceutical industry in India has been developed upto the extent that medicinal plants produced in India are not able to meet the industrial requirement. Therefore, import of some vegetable drugs becomes unavoidable. Data given in Table 15.4 depicts that during a period of nine years (1991-92 to 1999-2000) the import in terms of quantity of vegetable drugs and amount in rupees has shown a fluctuating trend. In this period, 1992-93 witnessed the lowest quantity import (914.95 tonnes only). In 1999-2000 the highest import was observed (3934.49 tonnes). The critical appraisal of such data also indicated the active efforts being made at various levels are to yield satisfactory results so far. Some valuable information on import of medicinal plants have been summarized in Table 15.5 and 15.6. However, the share of plant material in terms of cultivated and that procured from wild resources in foreign countries (from where the import was materialized) is not known.

Table 15.4

Import of Vegetative Drugs

Year	Ayurvedic & Unani Medicines		Homeopathic Medicines		Alkaloid	
	Quantity (Tonnes)	Cost (Rs. lakhs)	Quantity (Tonnes)	Cost (Rs. lakhs)	Quantity (Tonnes)	Cost (Rs. lakhs)
1991-92	1732.50	583.61	—	447.21	—	0.28
1992-93	914.95	442.83	—	427.53	—	—
1993-94	2201.95	268.14	—	581.67	—	1.65
1994-95	1814.29	236.94	—	524.91	—	11.96
1995-96	1287.37	1270.26	—	485.61	—	3.34
1996-97	3640.05	3395.02	126.21	496.96	—	—
1997-98	1637.19	507.04	102.13	572.49	6.41	97.94
1998-99	3761.57	1863.54	171.63	936.42	0.63	53.99
1999-2000	3934.49	3956.77	146.06	799.69	0.07	0.27

Source : Sharma, 2002.

Table 15.5
Import of medicinal plant commodities from India, during 1995-96

Sl. No.	Item	Quantity (tonnes)	Value (in lakhs Rs.)
1.	Agar Agar W/N modified	78.452	223.297
2.	Agarwood	12.089	11.889
3.	Ayurvedic and Unani NES	1287.373	179.887
4.	Belladonna extracts	0.400	5.355
5.	Chirata	58.224	14.610
6.	Galangal rhizomes and roots including greater galangal	55.300	6.344
7.	Ginseng extracts	5.764	323.398
8.	Ginseng power/chips	1.150	19.913
9.	Ginseng roots	210.626	38.702
10.	Liquorice roots	581.150	49.030
11.	Mint (others)	816.106	182.230
12.	Mint including leaves (all spices)	0.750	0.893
13.	Other ginseng roots	324.298	24.103
14.	Pyrethrum	132.500	89.304
15.	Sap and extracts of liquorice	50.827	39.974
16.	Sarsaparilla	3.500	1.076
17.	Sweet flag rhizome	2.500	3.266
18.	Unab (Indian jujube or Chinese dates)	25.186	1.711
19.	Vegetable saps and extracts	182.419	695.463
	Total	**3838.614**	**1910.445**

Table 15.6
Import of few medicinal plants in India during 1999-2000

Sl. No.	Plant Name	Export	
		Quantity (tonnes)	Value (in lakhs Rs.)
1.	*Glycirrhiza glabra*	581.15	49.03
2.	*Rauwolfia serpentina*	—	—
3.	*Panas ginseng*	324.30	14.69

Contd....

... Contd.

Sl. No.	Plant Name	Export	
		Quantity (tonnes)	Value (in lakhs Rs.)
4.	*Plantago ovata*	—	—
5.	*Swertia chirayata*	58.22	14.67
6.	*Cassio angustifolia*	—	—
7.	*Catharanthus roseus*	—	—
8.	*Hemidesmus indicus*	3.50	1.08
9.	*Myristica fragrans*	672.01	1225.00
10.	*Abelmoschus moschatus*	—	—
11.	*Santalum album*	—	—
12.	*Eucalyptus globules*	—	393.00

Plants indicated by blank (–) have been replaced by Indian vegetable drugs.

Marketing

The cultivation of medicinal plants in India is getting momentum but speed is not satisfactory. Farmers are being encouraged by various R&D agencies for domestication and cultivation of medicinal plants. Seed and planting material is given to farmers alongwith technical know-how. Inensive training programme are being regularly conducted and farmers are also acquainted with marketing scenario. Such programmes will meet success only with *Bye Back Guarantee* or with contract farming system. The marketing of medicinal plants is being strengthened in Rajasthan by the efforts of Rajasthan State Medicinal Plants Board (II Floor, Pantkrishi Bhawan, Jaipur). Marketing of Medicinal Plants in Madhya Pradesh has gained momentum. Mandis of Neemach, Khandwa, Bhopal and Jabalpur are well developed with respect to medicinal plants.

The relevant information on such aspects are given in Table 15.7, 15.8 and 15.9. Latest information collected from Rajasthan State Medicinal Plants Board have indicated that about 68000 hectare are in under medicinal plant cultivation out of which major share goes to Isabgol (about 62000 hectars). Due to efforts of various organization, the area is increasing but marketing board should also come forward for this purpose. National Institute of Agricultural Marketing (NIAM), Jaipur has also takes some initiatives in this direction.

Table 15.7
Estimated consumption of imported medicinal plants under cultivation

Sr. No.	Botanical Name	Vernacular Name	Parts Used	Estimated average consumption (kg)	% of Pharmacies consuming
1	2	3	4	5	6
1.	*Piper nigrum*	Kalimari	Fruits	55,200	43
2.	*Glycyrrhiza glabra*	Jethimadh	Roots	46,200	43
3.	*Swertia chirata*	Kariyatu	Whole plant	28,300	31
4.	*Berberis aristata*	Daru-Haldi	Bark	19,500	32
5.	*Picrorhiza kurroa*	Kadu, Kutaki	Roots	19,000	32
6.	*Pluchea lanceolata*	Rasna	Roots	13,200	24
7.	*Cinnamomum zeylanica*	Tamal patra	Leaves, Bark	10,700	46
8.	*Acorus calamus*	Ghoda vaj	Rhizomes	10,500	21
9.	*Ellataria cardamomum*	Elaichi	Seeds	8,600	21
10.	*Inula racemosa*	Pushkarmool	Roots	8,300	24
11.	*Hyoscyanus niger*	Khursaniajamo	Seeds	6,800	07
12.	*Saussurea lappa*	Kath, uplet	Roots, Whole Plant	6,200	12
13.	*Myristica fragrans*	Jaifal	Fruits, Flowers	5,200	24
14.	*Hedychium spicatum*	Kaupur kachali	Rhizomes	4,600	18
15.	*Valeriana jatamansi*	Tagarganth	Rhizomes	4,000	18
16.	*Nigella sativa*	Kalonji	Seeds	3,300	07
17.	*Eugenia caryophyllata*	Laving (Clove)	Flowers Bud	3,100	26
18.	*Anacyclus pyrethrum*	Akkalkaro	Whole plant	2,800	18

Contd....

...Contd.

1	2	3	4	5	6
19.	*Ferula narthrex*	Hing	Gum	2,750	19
20.	*Scindapsus officinalis*	Gajpiper	Fruits	1,450	07
21.	*Jasminum auriculatum*	Jui, Chameli	Whole plant	1,970	03
22.	*Viola odorata*	Banfasa	Whole plant	965	06
23.	*Carum roxburghii*	Bodi ajmod	Fruits	900	02
24.	*Rosa centifolia*	Gulab	Flowers	850	10
25.	*Amomum subulatum*	Elcho	Seeds	800	06
26.	*Citrus aurantifolia*	Santra	Fruits	755	02
27.	*Parmelia perfoliata*	Shaileyak	Whole Plant	750	06
28.	*Garcinia pendulata*	Amalvetas	Fruits	700	10
29.	*Croton tiglium*	Jamalgota	Fruits	400	06
30.	*Lilium polyphylum*	Kshirkakoli	Bulbs	175	04
31.	*Polygonatum cirrhifolium*	Menda	Root stock	150	06
32.	*Cannabis sativa*	Bhang	Seeds	140	02
33.	*Polygonatum verticillatum*	Mahamenda	Root stock	115	04
34.	*Areca catechu*	Sopari	Seeds	110	03
35.	*Papaver somniferum*	Khaskhas	Seeds	105	02
36.	*Fritillaria roylei*	Kakoli	Bulbs	65	04
37.	*Eulopia campestris*	Salampanjo	Roots	40	03
38.	*Crocus sativus*	Keshar	Stigma	35	04
39.	*Malaxis muscifera*	Rushbhak	Swollen stem	15	03
40.	*Malaxis accuminata*	*Jivak*	Swollen stem	15	04
41.	*Callicarpa macrophylla*	Priyanguful	Flowers	10	02

Contd....

...Contd.

1	2	3	4	5	6
42.	*Colchicum luteum*	Suranjan	Roots	10	02
43.	*Lubunga scandens*	Sugandhkokla	Fruits	10	02
44.	*Withania coagulence*	Kaknaj	Roots	10	02

Table 15.8

Plants culivated exclusively as medicinal crop

Plant	Part used	Areas where cultivated	Demand
Acorus calamus	RH	Karnataka*	High
Alpinia galanga	RH	Assam, W. Bengal, Karnataka and Kerala*	Med.
Aloe vera	LF (Juice)	Coastal areas of Saurashtra (Gujarat)	V. High
Ammi majus	FR	Jammu, Punjab and Western UP*	Med.
Andrographis paniculata	WP	UP, Bihar, WB, MP and Maharashtra*	High
Asparagus racemosus	RT	Neemuch (MP), Bundelkhand (UP)*	High
Atropa belladonna	RT/ LF	Tangmarg and Kashmir Valley (J&K)	Low
Carum carvi	FR	Lahaul and Kinnaur (HP), Kumaon (UA)*	High
Cassia angustifolia	LF; FR	Tirunelvelli, Ramnathpuram Distt. (TN)	High
Catharanthus roseus	RT; HB	Peninsular and southern coastal region*	V. High
Cephaelis ipecacuanha	RT	Mungpo (WB)	Med.
Chlorophytum borivilianum	RTS	Udaipur, Sikar (Rajasthan), Jalgaon (Maharashtra)*	High
Claviceps purpurea	Scler-otia	Nilgiri Hills, Bangalore and Jammu	High
Cincnona sps.	STBK	Nungpo (W. Bengal), Nilgiri Hills (TN)	High

Contd...

...Contd.

Plant	Part used	Areas where cultivated	Demand
Digitalis lanata	LF	Nilgiri and Pulney hills (TN), Bangalore	Low
Dioscorea floribunda	RH	Goa, Bangalore, Nungpo (WB) and Tripura	High
Embica offieinalia	FR	Bundlekhand and Eastern UP, Nimar (MP) and Bihar*	V. High
Eucalyptus globulus	LF. OIL	Nilgiri hills (TN)	High
Gloriosa superba	RT/ SD	Tiruchirapalli (TN)	Med.
Inula racemosa	RT	Lahaul (HP)*	Low
Kaempferia galangal	RH	Karnataka, TN and Kerala*	Low
Matricaria chamomilla	FL	Kullu (HP)*	Low
Papavar somniferum	Latex	Ghazipur (UP), Mandsaur (MP)	V. High
Pimpinella anisum	FR	Haryana, UP and Punjab*	High
Piper longum	FR/ RT	Bihar, Guntur (AP), Tura and Shillong (Meghalaya)*	High
Plantago ovata	SD/ Husk	Mehsana and Banaskantha (Gujarat)	V. High
Rauvolfia serpentina	RT	Hazaribagh (Jharkhand)	High
Saussurea costus	RT	Lahaul (HP)	High
Withania somnifera	RT	Manasa (MP)	High

* Small holding over scattered areas.
Abbreviations same as in Tables 15.1-15.4.

Future Prospects, Opportunities and Constraints

The World Health Organization (WHO) has emphatically advocated to strengthen the indigenous system of medicine in all countries of the World. This system has attracted the attention of

Table 15.9
Classification of drug sources based on plant parts

Sl. No.	Parts used as drug source	No. of Species used	% of Species used	Annual Consumption	% consumption of plant parts
1.	Whole Plant	48	15	789265	21
2.	Root	62	20	896200	24
3.	Leaf	24	08	254310	07
4.	Fruit and Seed	95	30	1071351	28
5.	Bark	36	12	265355	07
6.	Stem and Wood	18	06	297350	08
7.	Flower	16	05	34132	01
8.	Exudates (Gum resin)	11	04	147480	04
	Total	**310**	**100**	**3755443**	**100**

developed and developing countries both. Medicinal plants still play a major role in it. In several cases extracting the drug from medicinal plants in Cheaper than synthetic process (Farooqi and Sreerâmu, 2001). The variability in agroclimatic conditions in India has been a major factor in founding a great biodiversity of medicinal plants and we should make all effort to harvest this gift of nature but in a judicious and scientific manner. The demand of vegetable drugs of Indian origin has tremendously increased in International market and we must promptly utilize this situation. Many advanced countries have shown reasonable interest in Ayurvedic and Siddha system of medicine and it in our turn to utilize this trend and it may booast up our earning of foreign exchange.

However some constraints are coming in the way. These constraints include derth of scientific manpower for undertaking research on medicinal plants, their genetic improvement, agronomic practices, quality seed availability and agricultural economics. In Indian market there are several crude drugs which have identical name (like Brahami, Safed Musli, Kanoj) in local language. Their specific plants must be properly marked and for meeting the problem of adulteration, proper steps must be taken (Pharmacognostic

Studies). Many more plants which show extraordinary curative and healing properties are waiting for conservation, domestication and cultivation.

Foregoing discussion indicates that there are tremendous opportunities in India to become a world leader in medicinal plants trade due to its vast biodiversity of plants and indigenous knowledge we must exploit our plants but our climatic conditions also suits to the cultivation and introduction of several species of plants from temperate and tropical parts of the world.

□□□

Summary

Ashwagandha (*W. somnifera*) (L.) Dunal., family-Solanaceae) and red tephrosia (*T. purpurea*) (L.) Pers., family–Papilionaceae) were identified as the species of considerable therapeutic repute in Allopathic, Ayurvedic, Homeopathic and Unani systems of medicine. Screening the available and published literature revealed that reports on the systematic and scientific work aiming at their cultivation in Indian agroclimatic conditions were extremely scanty. Therefore, the present project was undertaken to investigate various environmental biological aspects of these species under laboratory, pot and field conditions.

The available and published literature on various ecobiological aspects of plants was updatively reviewed. Emphasis was given on the available information on the aspects of growth and development relevant to present studies. These aspects included environmental stresses in relation to germination, seedling growth, vegetative growth, reproductive growth,plant productivity and economic yield. A brief description of soil types affecting plant productivity and economic yield along with some macronutrients was also given. The role of growth substances in the regulation of various parameters of plant growth, development, productivity and economic yield was also briefly described along with the interaction of growth substances with environmental stresses in influencing plant productivity and economic yield. The existence of certain lacunae in our knowledge about the above mentioned aspects were pointed out so as to clarify the scope of present investigations. As far as possible, non-traditional crop plants and wild plants with determined therapeutic value were preferably quoted in the reviewed literature.

Experimental Technology contained information on general morphological, botanical and taxonomical characteristics of the experimental species. The present status of these species in India including their distribution, was also described. The therapeutic importance of specific plant parts was also elaborated with particular emphasis on the consumption of such parts by human beings in India. Seeds were collected from wild parental stock and stored under ordinary laboratory conditions. This chapter also included the standard abbreviations of selected growth substances along with their sources of procurement.

Phenological observations were undertaken on the plants under natural conditions and specific locations identified and marked for the purpose. It was observed that under natural habitat, seed germination and seedling emergence started in the month of May-June in *W. somnifera* where these processes were accelerated by occasional summer rains. Vegetative growth stage was followed by flowering and fruiting which were completed latest by December which was also the period of plant senescence and seed maturation. If allowed to grow plants of the same parental stock continued further vegetative and reproductive growth in the following years. In *T. purpurea,* germination and seedling emergence started in March and by the end of April these processes were completed. Various stages of vegetative growth are completed upto last week of May. The reproductive growth including seed setting and the beginning of pod maturation are completed upto mid–September in this species.

Under ordinary laboratory conditions of storage of seeds, dormancy was observed for a period of 11-14 months in *W. somnifera* but in *T. purpurea* this period was relatively shorter (6 to 9 months). Experimental evidences were obtained to indicate that dormancy in *W. somnifera* as well as *T. purpurea* appeared to be due to hard seed-coat. In *T. purpurea* a waxy seed coat was responsible for non-imbibition of water by the seeds. In both the species, presence of germination inhibitors was known on the basis of indirect evidences. Various physical and chemical treatments were employed to break the dormancy of seeds. It was observed that water soaking, hot water, low temperature and chemical scarification treatments for specific periods were successful for breaking the dormancy of seeds in both the species. Physiologically active concentrations of NAA, BA, GA_3 and ETH also showed promotory effects on seed germination percentage, indicating their efficacy in breaking the dormancy of

seeds. However, ABA and CME imposed (prolonged) the dormancy of seeds as indicated by the inhibition of germination process.

Seed germination studies on both the species were conducted under moisture stress, low and high temperature stresses and low and high radiation stresses under laboratory conditions. It was observed that increasing moisture stress drastically reduced germination percentage in both the species. *T. purpurea* proved to be relatively more sensitive to moisture stress than *W. somnifera* at seed germination stage. Low temperature stress improved the seed germination percentage while high temperature hampered the seed germination process in both species. Low intensity radiation (dark and shade conditions) satisfactorily promoted seed germination process while the high radiation stress reduced the germination percentage in both the species. Seedling growth of *W. somnifera* and *T. purpurea* under various environmental stresses was also studied under laboratory conditions. It was observed that decreasing external water potentials reduced the shoot and root lengths in both the species. Low temperature stress (20, 15°C) promoted the seedling growth in length while high temperature stress proved to be remarkably inhibitory for seedling growth of both the experimental species. Radiation stress of lower order tremendously improved the seedling growth which was, otherwise, reduced by high radiation intensity.

Effect of moisture stress on various parameters of vegetative growth was studied in pot cultured plants which were subjected to moisture stress at the seedling stage. It was observed that a mild moisture stress stimulated the vegetative growth in *W. somnifera* but all the other orders of moisture stress progressively and significantly retarded the vegetative growth in both the species. *T. purpurea* was found to be more susceptible to moisture stress. Vegetative growth was also studied by subjecting the seedlings to low and high temperature stresses and then transplanting them in pots. It was observed that low temperature stress even for a limited period produced long lasting but favourable effects on vegetative growth of plants. Vegetative growth responses to high temperature stress treatment were opposite to those observed for low temperature stress. The light deficit showed a promotory effect on various parameters of vegetative growth. However, total dark and high intensity of light have caused variable effects on vegetative growth in present plant materials. It was interesting to observe that the effect of temperature and radiation stresses, if given at the seedling stage, persisted upto vegetative growth stage of plants.

Simulated moisture stress of different orders reduced the reproductive growth to a remarkable extent in both the species and such adverse effects of moisture stress were observed in the plants obtained from the stressed seedlings and also pot cultured plants subjected to moisture stresses. The seedling stage was found to be more responsive to the adverse effect of moisture stress than the vegetative growth stage. A moderate low temperature stress resulted in profused flowering and fruiting in present investigation while high temperature stress exerted a deteriorative effect on reproductive growth. The high radiation stress promoted the reproductive growth in both the species. However, light deficit caused a reduction in reproductive growth.

The effect of simulated moisture stress which the plants experienced at vegetative growth stage was studied on plant productivity and economic yield. It was observed that moisture stress reduced the dry matter accumulation and the economic yield in general. However, a mild moisture stress promoted the plant productivity and economic yield in *W. somnifera*. The total alkaloids content per unit dry weight of the economic parts was found to be increased by the increasing order of the moisutre stress upto a critical level of the moisture stress. The long lasting effect of low and high temperature stresses was also reflected in plant productivity and economic yield and it was observed that the low temperature stress promoted dry matter accumulation and economic yield in both the species. However , the therapeutic yield was reduced by this treatment. The high temperature stress reduced the dry matter production and economic yield. The high intensity of light, though given to seedlings for a limited period, maintained its stimulating effect on plant productivity and economic yield. However, the deficit of light gave contrary results.

Seeds of experimental species were pretreated with various concentrations of auxin, gibberellin, cytokinin, morphactin, abscisic acid and ethephon, separately. The germiation behaviour and seedling emergence were studied under ordinary soil conditions in pots followed by the study of growth, development and reproduction. The cumulative effect of seed treatments on plant productivity and economic yield was also investigated. It was observed that NAA showed concentration dependent dual behaviour on seed germination, seedling growth and vigour. Comparatively lower concentrations promoted while higher concentrations of NAA suppressed these parameters. All the tested concentrations of GA_3 and BA improved seed germination, seedling growth and vigour.

BA (20 mg/*l*) provided most favourable results. The lowest tested concentration of CME (1 mg/*l*), a morphactin, remarkably promoted seed germination, seedling growth and seedling vigour under soil conditions. However, its higher doses almost progressively suppressed these parameters. Both ABA and ETH in general showed inhibitory effect on seed germination, seedling growth and vigour. However, the lower concentration of ETH promoted seed germination in both the species.

The dual action of NAA was also observed in affecting the vegetative growth as indicated by the promotory effect of relatively lower concentrations of NAA on shoot and root growth and leaf area and a retarding effect of higher concentrations of the same on these parameters. Various parameters of vegetative growth were promoted by GA_3 and BA, the latter being he most effective PGR with this point of view. With the exception of 1 and 10 mg/*l* concentrations of CME for *W. somnifera* only, CME, ABA and ETH proved to be general retardants of vegetative growth. NAA concentrations upto 100 mg/*l* showed promotory effects on reproductive growth while higher concentrations of NAA showed herbicidal effect on reproductive growth. GA_3 and BA maintained their long lasting and promotory effects on reproductive growth of both the experimental species. The reproductive growth was retarded by CME (except very low concentration in *W. somnifera*), ABA and ETH in general.

NAA also showed concentration dependent dual action in affecting plant productivity and economic yield. Higher concentrations proved to be inhibitory but lower concentrations promoted the same. However, the therapeutic yield was contrarily promoted by higher concentrations of NAA in *W. somnifera*. The dry matter accumulation in the whole plant as well as in economic parts, was enhanced by gibberellin and cytokinin treatments. However, the therapeutic yield was reduced by both these growth substances but the total pharmaceutical yield was recorded to exceed the control level because of recovery of more economic yield (CDY) from economic parts under such treatments. The plant productivity and the economic yield, were reduced by ETH, ABA and CME treatments in general. The therapeutic yield was, however, promoted by these growth retardants.

Experimental evidences were obtained to point out that NAA, BA and GA partly or completely counteract the adverse effects generated by various stresses. A combination of moisture stress with growth promoters, or of temperature stress with growth promoters,

furnished advantageous results from pharmaceutical point of view. However, a combination of NAA with the above mentioned environmental stresses was not found suitable in *T. purpurea* with this point of view. Evidences were obtained to indicate that combination of moisture and temperature stresses with growth retardant like CME, ABA and ETH was excessively disadvantageous with economic yield point of view.

Simple experiments were conducted to find out suitable soil type for the cultivation of experimental species in hand. It was observed that garden soil mixed with white sand in the ratio of 50 : 50 was most suitable for successful cultivation of *W. somnifera* as well as *T. prpurea*. Similarly simple experiments were also conducted to get preliminary information about the mineral requirements of the experimental species with special reference to three major elements N, P and K. It was observed that a basal dose of 50 gm/pot (10 kg soil) of nitrogen was most suitable for *W. somnifera*. Basal dose gave the best results, specially in comparison to other modes and times of treatments in this species. However, in *T. purpurea*, spray treatment at pre-flowering stage in lower doses proved to be more beneficial. In *W. somnifera*, a basal application of phosphorus at the rate of 100 gm of P_2O_5 per 10 kg soil-sand mixture and a basal dose of potassium (K_2O) at the rate of 50 gm/10 kg soil sand mixture were found to be most suitable for successful plant cultivation. However, in *T. purpurea*, spray application of phosphorus at the time of ready-to-flower stage was found to be most suitable. With reference to economic yield, a lower dose of potash at ready-to-flower stage was found most suitable. All the results obtained from these experiments were interpreted in the light of recent literature. However, it should be mentioned that the explanations furnished in this book may be tentative because experiments were not conducted in phytotronic and ideal environments and, therefore, interactions of environmental factors with various treatments could not be eliminated. But efforts were made to minimize experiments error by giving identical environments to treated and untreated (control) plants and also by following suitable experimental designs and statistical methods for data processing. Applicability of experimental observation and interpretations were pointed out in each chapter of the book. Present scenario of export-import and marketing of medicinal plants in general and experimental species in particular has been described in a separate chapter (Chapter 15).

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General Conclusions and Recommendations

The main objective of present investigations was to make efforts for exploring the possibility of the cultivation of two therapeutically important but agriculturally overlooked plant species *i.e. Withania somnifera* and *Tephrosia purpurea*. In their natural habitats, these plants are often subjected to various types of environmental stresses. Therefore, a study of the growth, development and productivity responses of these species under simulated environmental stresses was considered a subject of paramount importance for a plant domestication oriented programme. The extent to which a plant species responds to better cultural practices depends upon the nutritional requirements of the species and its growth characteristics under various water regimes and soil types. The present investigations were carried out keeping in view all these facts.

Extensive screening of the available literature revealed various lacunae in our knowledge about seed germination and seedling growth behaviour, type and causes of dormancy vegetative growth and reproductive growth behaviour and pharmaceutical and therapeutic yields. This was seen particularly true for the medicinal plants of India. Most of the medicinal plants are still predominantly obtained from their wild resources, particularly forests, and consequently these natural resources are depleting at an alarming rate. Therefore, the need for bringing the medicinal plants under cultivation has been repeatedly emphasized by various scientists. It was also realized that for such efforts, basic information on the

ecophysiological aspects of plants should be collected by further experimentations.

Phenological observations in present studies revealed that under natural conditions, seeds of both the species exhibit dormancy upto a maximum period of 14 months in *W. somnifera* and 9 months in *T. purpurea*. It was also observed that remarkable changes may be brought about in morphology, growth and developmental patterns of plants when these are subjected to cultivation practices. For example, plants of *Withania somnifera* when brought to cultivation are changed morphologically to such an extent that some authors have given a separate specific name to the cultivated forms of *W. somnifera* and the name of *W. ashwagandha* was suggested. However, it was noted that *W. somnifera* is cultivated to a very limited extent in parts of Rajasthan and Madhya Pradesh.

Various physical and chemical treatments could be successful only partly as the germination percentage could not exceed 58.4% in *W. somnifera* and 57.4% in *T. purpurea* even under most effective treatment in the present investigations. Laboratory tests have, however, indicated that ungerminated seeds were still viable. This indicated the presence of some endogenous germination inhibitor in high concentration in both the species. Although, the chemical nature of these inhibitors could not be investigated in present studies, certain alkaloids might be responsible for such a constraint. This conclusion is based on indirect evidences including response to the application of growth promotors like cytokinins which are known to reduce the level of endogenous germination inhibitorsof hormonal nature like ABA. There are reports that certain alkaloids in the seeds are reduced when the species is brought under cultivation. In present studies also, seed germination percentage of the seeds obtained from cultivated plants (I generation) was higher indicating that alkaloids probably function as germination inhibitors. Furthermore, plants which were treated with growth promoters yielded the seeds with higher germination percentage but reduced alkaloids content. However, the chemical characteristics of the actual germination inhibitors of *W. somnifera* and *T. purpurea* should be worked out in future.

The suppressive effect of moisture stress on seed germination process under pot, laboratory and field conditions, has been a subject of some reports. However, the feature of special interest in our studies was the stimulatory effect of mild moisture stress on seed germination in *W. somnifera*. This was explained on the basis of some published reports which pointed out that a mild moisture stress

accelerated ethylene biosynthesis upto such extent that endogenous gibberellins are activated or synthesized *'de novo'*. Exploration of the effect of such a mild stress is, therefore, recommended for other cultivated and wild plants. This may furnish agriculturally valuable information. The low and high temperature stresses promoted and retarded, respectively, the germination percentage in both the species. These studies encouraged the author to suggest that "hardening" responses to low and high temperature should be worked out in detail so as to quantity the tolerance of plants to less favourable conditions of temperate and tropical climates. Seed germination responses to radiation stress revealed that seeds of neither of the species in question, were photoplastic and dark conditions or light deficit proved to be promotory for seed germination.

Seedling growth was found to be remarkably sensitive to moisture stress which adversely affected various aspects of seedling growth. It was primarily due to the reduction of the mobilization of reserve carbohydrates of seeds. This metabolic constraint on successful metabolism is caused by reduced α–amylase activity which is known to be primarily regulatory for such a physiological event. Low temperature stresses upto a certain level stimulated seedling growth. Similarly low radiation stresses also showed stimulatory effects on seedling growth. *T. purpurea* was found to be more susceptible to moisture, thermal and radiation stresses at seed germination and early seedling growth stages than *W. somnifera*.

The study of moisture stress influencing the vegetative growth of plants has been traditionally conducted employing the controlled irrigation treatments. Such studies were found of sufficient value for irrigation scheduling. In present studies, though, experiments were conducted under pot conditions, results could indicate that an interval of more than five days between two consecutive irrigations would drastically reduce the vegetative growth of plants. The feature of special interest here was the effect of mild moisture stress which enhanced the vegetative growth of plants. The seedling growth stage was found to be considerably susceptible to radiation and temperature stresses and the effect of these treatments on seedling stage culminated in a reduced vegetative growth. It may be suggested that the effect of treatments of plant with radiation and thermal stresses at early and late vegetative stages on further growth and developmental processes, should also be worked out. This may provide more clear picture of the degree of tolerance of plant species.

The reproductive growth was also adversely affected by

simulated environmental stresses. However, the outstanding feature of results was the observation that mild moisture stress produced promotory effect on reproductive growth in *W. somnifera*. There are evidences to point out that a mild moisture stress allowed to generate ethylene upto such level that the same was just sufficient to enhance endogenous gibberellin biosynthesis. However, –3 bar external water potential did not prove a mild moisture stress for *T. purpurea*. Such a stimulatory level of moisture stress varies from species to species. It should also be critically evaluated for other species because thisprovides valuable information with pharmaceutical point of view. The present studies have also shown that the critical level of low temperature which stimulated reproductive growth, should also be worked out for other medicinal plants. In present investigation 20°C temperature proved to be most stimulatory for *T. purpurea* and below this temperature beneficial effects started to decline. Both the species were found to be similar with respect to their reproductive growth responses to high as well as low radiation stresses which caused long lasting effect upto the generative phase. It may be concluded that ready-to-flower stage was more susceptible than earlier stages of plant growth with respect to the adverse effect of moisture and radiation stresses.

The gamut of all these studies seems to indicate that the temperature optima for growth, development and yield differs for both the species. This conclusion is based on indirect experimental evidences. It has been noted that 30°C temperature given to seeds during germination or given to 15 days old seedlings by a careful manipulation of experimental procedure, produced different effects on two species. This temperature was found to be promotory for *W. somnifera* but inhibitory for *T. purpurea*. This difference indicated that for *W. somnifera* 30°C temperature simulated a mild low temperature stress while for *T. purpurea* a mild high temperature stress. But to find out the critical level of the specific optimum temperature for both the species, needs further experimentation.

Limited information are available to point out the correlation between the active principles of medicinal plants with the environmental components. On the basis of present studies it may be concluded that combination of moisture, temperature and radiation stresses may improve the market quality of the crude drug. This is examplified by *W. somnifera*. The roots collected from Bikaner range (Nagore, Pali etc.) of Rajasthan were considered superior with respect to therapeutic qualities as compared to the roots collected from other parts of India (Pratapgarh area of southern Rajasthan). Analysis of

climatological data of these areas indicated that Bikaner area experiences comparatively high temperature, more solar radiation and low rainfall. All these conditions together provide more suitable environment for Withania alkaloids biosynthesis.

Present studies also indicated that harvest index is not a good indicator of the economic yield specially for such medicinal plants where non-reproductive parts constitute the economic yield and the chemical constituents accumulated in these parts determine the pharmaceutical yield. It may be further concluded that the use of selected plant growth substances in suitable concentrations may ameliorate the cultivation of medicinal plants. However, the agricultural economics of application of cytokinin and abscisic acid should be worked out before rcommending their application for farming purposes. Unfortunately, both these bioregulants, although showing potent action, are very costly to use in agriculture. Organic chemists should come forward to evaluate the technology so as to reduce their cost of production as they have done in case of gibberellins which are now available on reasonable rates for agricultural purposes.

Application of NAA, GA and BA in physiologically active concentrations may provide fruitful results with pharmaceutical point of view. However, morphactin, ABA and ETH enhanced the therapeutic yield. The total pharmaceutical yield under a particular treatment is determined by the effect of treatment on the dry matter accumulation in the economic part of plant and on the therapeutic yield of that particular part. Therefore, it is difficult to generalize the information for specific and most suitable concentrations which vary from species to species and also depend upon the interaction of environmental factors with the genotypic expression. Therefore, it is recommended that further studies should be conducted on the use of growth regulators for amelioration of medicinal plants.

Growth promotors and retardants tested in the present investigations have shown antagonistic as well as synergistic interactions with environmental stresses. It may be concluded that moisture stress coupled with simultaneous or successive applications of BA and GA_3, may provide valuable results because moisture stress enhances the therapeutic yield but BA or GA_3 stimulate dry matter accumulation upto such extent that overall pharmaceutical yield considerably exceeds the level of untreated control plants. This situation is favourable both for farmers and pharmaceutical industry. Suitable soil type was found a prerequisite for the cultivation of present plant materials. It was also noted that both these species

responded well to N, P and K, applications in suitable doses and by appropriate method and at optimum time of application.

The endogenous level of natural growth hormones controls plant productivity. However, the quantitative correlation of these bioregulants with yield components still remain uninvestigated. In present studies too, only an indirect evidence (by external application) could be obtained. This evidence indicated that growth hormones are involved in affecting both the therapeutic and pharmaceutical yields. Effect of certain growth regulators has shown similarity with the effects caused by environmental stresses. This indicated that environmental stresses probably acted through a "hormonal promotion-inhibition balance". This information may be employed agriculturally by giving an appropriate quantum of environmental stress resulting in the enhanced plant productivity of medicinal herbs. The use of critical level of environmental stress, specially the water stress at suitable time would be cheaper and convenient as compared to hormonal application.

On the basis of these studies and a critical appraisal of published literature, a tentative working hypothesis may be given concerning the control of biosynthesis of secondary plant products (alkaloids etc.) which is most controversial issue in plant biology. The most probable mode of alkaloid biosynthesis in plants has been described by Roos and Luckner (1979). They described that two mechanisms are commonly operated for alkaloid biosynthesis-biway and high-way. Results of the present investigations seem to indicate the biosynthesis through that the probability a biway mechanism is very poor in present plant species because under favourable conditions application of stimulants of protein biosynthesis further augumented the alkaloids biosynthesis. This indicated that synthesis of alkaloids directly from amino acids is least probable way, especially at flowering stage. At the same time the demerits of a probable highway biosynthesis mechanism were also evident because protein biosynthesis is retarded under moisture stress and in such condition the total alkaloids content should have been reduced which was not found in the present case. Therefore, it may be concluded that some other mechanism may be operative for the biosynthesis of alkaloids under hormonal or environmental stress treatments. There are indirect evidences to point out that under low temperature stress, highway mechanism provides a more satisfactory explanation while under moisture stress condition a biway process explains it more satisfactorily. Further studies should be conducted on these aspects. Such

investigations may provide more useful information both with academic as well as application points of view.

The systematic and convincing information generated through these case studies, should be used for introduction, domestication and cultivation of these medicinal plants and few other species of Solanaceous and Fabaceous plants in non-traditional areas. These efforts should be part and partial of alternative agriculture and organic farming coupled with export promotions.

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Literature Cited

Abdulla, A.A. and Verkerk, K. (1970). Growth, flowering and fruiting in tomatoes in relation to temperature, cycocel and GA_3. *Natl. J. Agric. Sci. 18* : 105-110.

Abeles, F.B. (1973). *Ethylene in Biology.* Academic Press, New York, pp. 102-109.

Abeles, F.B. and Lonski, J. (1969) Stimulation of lettuce seeds germination by ethylene. *Plant Physiol. 44* : 277-278.

Addicott, F.T. (1981). *Abscission,* University of California Press, California, pp. 89-92.

Addicott, F.T. and Lyon, J.L. (1969). Physiology of abscissic acid and related substances. *Ann. Rev. Plant Physiol. 20* : 139-164.

Afaq, S.H., Wasiuddin and Afridi, M.M.R.K. (1978). Effect of different levels of phosphorus on growth and alkaloid content of *Datura innoxia* Mill. *Indian J. Pharm. Sci. 40* : 227-228.

Agakishicer, D. (1962). Accelerating effect of low temperature of germination of cotton seed in solarized solution. *Fizeol. Rest. 8* : 505.

Agarwal, R.L. (1980). *Seed Technology,* Oxford and IBH Publishing Co., New Delhi, pp. 449-507.

Ahroni, M. and Richmond, A.E. (1978). Endogenous gibberellins and abscisic acid content as related to senescence of detached lettuce leaves. *Plant Physiol. 62* : 224-228.

Alden, J.N. and Hermann, R.K. (1977). Cold injury and resistance in crop plants. In : *Physiological Aspects of Crop Nutrition and Resistance.* (Ed.) U.S. Gupta, Atma Ram and Sons, Delhi, pp. 163-180.

Alvin, P. de T. and Kozlowski, T.T. (1977). *Ecophysiology of Tropical* Crops. Academic Press, London, New York, pp. 111-117.

Amen, R.D., Carter, G.E. and Kelly, R.J. (1970). The nature of seed dormancy and germination in the salt marsh grass *Distichlis spicata. New Phytol. 69* : 1005-1013.

Anderson, R.N. (1968). Germination and Establishment of Weeds for Experimental Purpose, Weed Science Society of America : Illinois. pp. 68-80.

Anonymous 2000, *Progress Report* : Central Task Force on Medicinal Plants, GOI, New Delhi.

Arnold, A.C. and Krochmal, C. (1973). A Guide to the Medicinal Plants of United State. N.Y.T. Quadrangle, New York, pp. 63-90.

Arnon, I. (1975). Physiological principles of dryland crop production. In : *Physiological Aspects of Dryland Farming.* (Ed.) U.S. Gupta, Oxford and IBH Publishing Co., New Delhi, pp. 1-73.

Asana, R.D. (1975). Physiological Approaches to Breeding of Drought Resistant Crops. ICAR, New Delhi, pp. 35-41.

Asana, R.D. and Sarin, M.N. (1969). Crop Physiology in India. ICAR, New Delhi, pp. 20-22.

Ashour, N.I. (1977). Crop response to combined nutrients—regulars spray. In *Physiological Aspects of Crop Nutrition and Resistance* (Ed. U.S. Gupta, Atma Ram and Sons, New Delhi, pp. 181-195.

Aspinall, D., Nicholson, P.B. and May, C.H. (1964). The effect of soil moisture stress on the growth of barley. *Aust. J. Agric. Res. 15*: 729-734.

Atal, C.K. and Kapur, B.M. (1982). Cultivation and Utilization of Medicinal Plants. RRL (CSIR), Jammu Tawi.

Babu, V.R. and Joshi, M.C. (1970). Studies on physiological ecology of *Borreria articularis.* A common weed of bajra fields I. seed production, germination and seedling survival. *Trop. Ecol. 11*: 126-136.

Bair, W. (1973). Crop weather analysis model : Review and model development. *J. Appl. Meteorol. 12* : 937-947.

Barendse, G.W.M. (1983). Hormonal regulation of enzyme synthesis and activity. In *Aspects of Physiology and Biochemistry of Plant Hormones.* (Ed.) S.S. Purohit, Kalyani Publishers, New Delhi and Ludhiana, pp. 1-68.

Barlow, E.W.R., Boestma, L. and Young, J.L. (1976). Root temperature and soil water potential effects on growth and soluble carbohydrate concentration of corn seedlings. *Crop. Sci. 16* : 59-62.

Barton, R.A., Verbeck, R., Ellis, R. and Khan, A.A. (1973). Abscisic acid inhibition of gibberellic acid and cyclic AMP induced amylase synthesis. *Physiol. Plantarum. 29* : 186-189.

Basu, D. and Chakarvarti, R.K. (1980). Exploration and improvement of *Catharanthus roseus.* Zinn. G. Don. *Proc. 67th Session, Indian Sci. Congress, Calcutta,* Part III, Abstr. pp. 161.

Basu, D. and Chakarvarti, R.K. (1984). Changes in the growth and development of the berries of *Solanum khasianum,* Clarke by gibberellic acid. *Acta Bot. Indica, 12* : 41-44.

Begg, J.E. and Barton, G.W. (1973). Comparative study of five genotypes of bulrush millet (*Pennisetum typhoides*) under a range of photoperiods and temperatures. In : *Plant Responses to Climatic Factors* (Ed.) R.O. Stalyer, *Proceedings of the Uppsala Symposium,* UNESCO, Paris, pp. 141-144.

Begg, J.E. and Turner, N.C. (1976). Crops and Water deficits. *Adv. Agron. 28* : 161-217.

Bell, E.A. (1980). Secondary plant products. In : *Encyclopedia of Plant Physiology,* New Series Vol. 8 (Eds.) O.L. Lange, P.S. Nobel, C.B. Osmond and H. Ziegler : Springer Verlag, Berlin and New York, pp. 229-257.

Berry, J.A. and Raison, J.K. (1981). Responses of macrophytes to temperatue. In : *Physiological Plant Ecology.* Part I (Eds.) O.L. Lange, P.S. Nobel, C.B. Osmond and H. Ziegler, Springer Verlag, Berlin and New York, pp. 203-232.

Bewley J.D. and Black, M. 1995. *Seeds : Physiology of Development and Germination,* Springer Verlag, Berlin and New York.

Bewley, J.D. (1979). Physiological aspects of dessication tolerance. *Ann. Rev. Plant Physiol. 30* : 195-238.

Bewley, J.D. and Black, M. (1978). Physiology and Biochemistry of Seeds in Relation to Germination. Vol. 1 : Development, germination and growth. Springer Verlag, Berlin.

Bhandari, M.C. and Sen, D.N. (1972). Growth regulation activity exhibited by substances present in fruit pulp of *Citrullus lanatus* (Thumb). *Man-sf. Zeit. Natur forsch. 27* : 72-75.

Bhargava, A., Trivedi, P.C., Bahl, H.M. and Mathur, K.M. (1979). Effect of soil texture on multiplication of melidiogyne and the plant growth of *Capscium annum L. Udyanika. 3* : 26-27.

Bhattacharjee, S.K., 2001. *Handbook of Medicinal Plants,* III Edition. Pointer Publishers, Jaipur, India.

Bierhuizen, J.F. (1973). The effect of temperature on plant growth,

development and yield. In : *Plant Responses to Climatic Factors.* Proceeding of the Uppsala Symposium, UNESCO, Paris (Ed.) R.O. Slayter, pp. 89-95.

Biesboer, D.D. and Mahlberg, D. (1979). Effect of midlum modification and selected precursors on sterol production by short-term callus cultures of *Euphorbia tirucalli. J. Nat. Prod. 42* (6) : 648-657.

Bisaria, A.K. and Paliwal, N.K. (1981). Effect of ethephon on seed germination and seedling growth and sugar in *Triticale. Acta Bot. Indica. 9* : 148-150.

Biscoe, P.V. and Gallagher, J.N. (1977). Weather, dry matter production and yield. In : *Environmental Effects on Crop Physiology.* (Eds.) J.J. Landsberg and C.V. Cutting, Academic Press Inc. London, New York, pp. 75-100.

Bishnoi, S.K., 1997. Seed and stress physiological aspects of bioremediating plant species. MSc (Ag.) Thesis, Rajasthan Agril. Univ., Bikaner, India.

Bjorkman, M. Nobs, Mooney, H. Trowghton, J., Berry, J. Nicholson, F. and Ward, W. (1974). Growth responses of plants from habitats with contrasting thermal environments. Transplant studies in the Death Valley, *Experimental Garden, Carnegie Inst. Year Book, Washington,* 1973 : pp. 748-757.

Black M., Bradford K.J. and Vazquez Ramos, J. 2000. *Seed Biology: Advances and Applications.* CABI Publishing, Wallingford, Oxon, UK.

Black, M. (1970). Seed germination and dormancy. *Sci. Progr. 58* (231): 379-393.

Black, M. (1980). The role of endogenous hormones in germination and dormancy. *Israel J. Bot. 29* : (104) : 181-192.

Blacklow, W.M. (1972). Influence of temperature on germination and elongation of the radicle and shoot in corn (*Zea mays* L.). *Crop. Sci. 12* : 647-650.

Blondon, F., Jacques, M., Jacques, R. and Moussequ, M. (1977). The combined action of light and temperature on development : Some examples analysed in phytotron. In : *Environmental Effects on Crop Physiology.* (Eds.) Landsberg, J.J. and C.V. Cutting, Academic Press, London, New York, pp. 309-320.

Bohra, P.N. and Sen, D.N. (1976). Behaviour of some arid zone plants under water stress. Abstr. All India Symposium on Advancement of Ecology, Muzaffarnagar, pp. 29.

Bopp, M. (1972). On the effects of morphactins. In : *Hormonal Regulation of Plant Growth and Development.* (Eds.) Kaldway, H. and Y. Varder, Verlag, Chemie. Berlin, pp. 103-113.

Borkowska, B. and Rudnicki, R. (1975). Change in the level of cytokinins in apple seeds during stratification. *Fruit Sci. Rep. 2* : 1-6.

Bose, K.T. (1984). Pharmacopoeia. Bishan Singh and Mahendra Pal Singh, Dehradun, pp. 171-186.

Bose, K.T. and Harada, H. (1970). Effect of temperature and growth regulating substances on flowering of *Pharbitis nil. Bot. Mag. Tokyo. 83* : 228-284.

Boyee, K.G., Cole, J.F. and Chilkote, D.O. (1976). Effect of temperature and dormancy of germination of tall fescue. *Crop. Sci. 16* : 15-18.

Boyer, J.S. (1976). Water deficit and photosynthesis. In : *Water Deficit and Plant Growth.* Vol. IV (Ed.) T.T. Kozlowski, Academic Press, London and New York, pp. 153-190.

Bradford, K.J. and Hsiao, T.C. (1982). Physiological responses to moderate water stress. In : *Physiological Plant Ecology.* Part II (Eds.) Lange, O.L., P.S. Nobel, C.B. Osmond and H. Ziegler, Springer Verlag, Berlin, pp. 265-324.

Brady, N.C. (1980). The evaluation and removal of constraints in crop production. In : *Linking Research to Crop Production.* (Eds.) Staples, R.C. and R.J. Kuhr. Plenum Press, New York, London, pp. 11-34.

Brucher, H. 1985. Rich genepool of forgotton economically useful plants in tropics. *Plant Res. and Develop* 22 : 109-120.

Bruinsma, J. (1982). Plant growth regulators in field crops. In : *Chemical Manipulation of Crop Growth and Development.* (Ed.) J.S. McLaren, Butterworths, London, pp. 3-11.

Buhan T. 2000. The use of Benzyl adenine in orchard fruit growths. Mini Review. *Plant Growth Regulation,* 32 : 381-390.

Burke, M.J., Gupta, L.V., Quamme, H.A., Weiser, C.J. and Li, P.H. (1976). Freezing injury in plants. *Ann. Rev. Plant Physiol. 27* : 507-28.

Campbell, L.E. (1981). Fundamentals of radiation and temperature relations. In : *Physiological Plant Ecology—I.* (Eds.) Lange, O.L., P.S. Nobel, C.B. Osmond and H. Zigler, Springer Verlag, Berlin, pp. 11-55.

Carlson, P.S. (1980). *The Biology of Crop Productivity.* Academic Press, New York and London, pp. 39-51.

Cathey, H.M. and Campbell, L.E. (1975). Effect of light stress on plant growth of tomato. *J. Am. Soc. Hort. Sci.* 100-165.

Cathey, H.M. and Campbell, L.E. (1982). Plant responses to light quality and quantity. In : *Breeding Plants for Less Favourable Environments.* (Eds.) Christiansen, M.N. and C.F. Lewis, John Wiley and Sons, New York, pp. 213-258.

Chadha, K.L. (1976). Improvement of Horticultural, Plantation and Medicinal Plants. Proc. III Intn. Symp. on Trop. and Sub. Trop. Hort. Vol. I, Today and Tomorrow's Printers and Publishers, New Delhi, pp. 176-194.

Chadha, K.L. (1978). Improvement of Horticultural, Plantation and Medicinal Plants. Proc. III Intn. Symp. on Trop. and Sub. Trop. Hort. Vol. III, Today and Tomorrow's Printers and Publishers, New Delhi, pp. 186-194.

Chandel, S.R.S. (1995). A Handbook of Agricultural Statistics. Achal Prakashan Mandir, Kanpur (U.P.) India.

Chatterji, S.K., Nandi, R.P., Chaudhary, R. and Chatterji, S. (1979). Effect of external and internal factors on productivity, alkaloids and steroids in some commercially cultivated medicinal plants of Darzeeling Hills. *Planta Medica, 36* (3) : 235-236.

Chatterji, U.N. and Mohnot, K. (1968). Ecophysiological investigations on the *imbibition* and germination of seeds of *Prosopis juliflora* Linn. *Proc. Recent. Adv. Trop. Ecol.* Part I : pp. 261-268.

Chaudhari, B. and Singh, S.N. (1960). Seed treatment with plant growth regulators and their effects on growth and yield of tomato. *Ind. J. Hort. 7* : 48-51.

Chaudhary, B.K. and Chakravarti, V.R. (1982). Effect of seed treatments using some chemicals on the shoot and root growth of *Citrus limonia. J. Maharashtra Agril. Univ. 7* (1) : 66-68.

Chawan, D.D. (1971). Role of high temperature pretreatments on seed germination of desert species of *Sida* (Malvaceae) *Oecologia* (Berl.), 6 : 343-349.

Chen, S.S.C. (1975). Role of gibberellic acid in dormancy and seed germination. In : *Gibberellins and Plant Growth* (Eds.) Krishnamoorthy, H.N., Wiley Eastern Ltd., New Delhi, pp. 91-100.

Ching, M.T. (1972). Metabolism of germinating seeds. In : *Seed Biology*

(Ed.) Kozlowski, T.T., Vol. II, Academic Press, New York, pp. 103-124.

Christiansen, M.N. (1978). The physiology of plant tolerance to temperature extremes. In : *Crop Tolerance to Sub-optimal Land Conditions. Am. Soc. Agron. Wisconsin.* pp. 111-174.

Christiansen, M.N. (1982). World environmental limitations to food and fiber culture. In : *Breeding Plants for Less Favourable Environment.* (Eds.) Christiansen, M.N. and C.F. Lewis, John Wiley and Sons, New York, pp. 1-12.

Christiansen, M.N. and Lewis, C.F. (1982) Breeding plants for less favourable environments. John. Wiley and Sons, New York, pp. 435-440.

Chung, J.D., Chung, C.K., Suh, Y.K. and Byun, S.Y. (1981). Studies on aseptic culture of *Dendrobium monele* seeds. IV–Effect of light/dark treatments on seed germination and seedling growth. *J. Kore. Soc. Hort. Sci. 22* (2) : 139-145.

Clark, R.W. and Hiler, E.A. (1973). Plant measurements as indicators of crop water deficit. *Crop. Sci. 13* : 466-469.

Cooper, C.S. and McDonald, P.W. (1970) Energetics of early seedling growth in corn (*Zea mays* L.). *Crop. Sci. 10* : 136-139.

Cooper, M. and Hammer, G L. 1996. *Plant Adaptation and Crop Improvement.* CABI Publishing Wallingford, Oxon, UK.

Corcoran, M.R. (1975). Gibberellin antagonists and antigibberellins. In : *Gibberellins and Plant Growth.* (Ed.) Krishnamoorthy, H.N., Wiley Eastern Ltd., New Delhi, pp. 289-332.

Coulter, J.K. (1980). Crop improvement. In : *Linking Research to Crop Production.* (Eds.) Staplex, R.C. and R.J. Kuhr, Plenum Press, New York and London, pp. 195-205.

Cracker, L.E. and Simon, J.E. 2002. Herbs, Spices and Medicinal Plants Vol. 1, 2, 3 and 4—*Recent Advances In : Botany, Horticulture and Pharmacology.* CBS Publishers and Distributors, New Delhi.

Curtis, H.J. (1967). Radiation and ageing. In : *Symposia of the Society for Experimental Biology.* XXI. Cambridge at the University Press, pp. 51-67.

Dastur, J.F. (1970). Medicinal Plants of India and Pakistan. D.B. Tarapore Vala Sons and Co. Pvt. Ltd., Bombay, pp. 178-180.

Dennis, F.G., Jipecki, J.J. and Kiang, C.L. (1970). Effect of photoperiods of flowering and runner development of three strawberry cultivars, *J. Am. Soc. Hort. Sci. 95* : 750-54.

Dikshit, A.P. (1972). Gene ecological studies of a wild jute species II–

Phenology, reproductive capacity, dispersal and germination of seeds in nature. *Plant Sci.* 4 : 72-74.

Dolan, D.D. (1972). Temperature photo period and light intensity effects on growth of *Pisum Sativum*, Crop. Sci. 12 : 60-62.

Down, R.D. and Hillemers, H. (1975). *Environmental and Experimental Control of Plant Growth.* Academic Press, London, pp. 39-58.

Duke, J.A., (1982). Ecosystematic data on medicinal plants. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) Atal, C.K. and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 13-23.

Duncan, D.B. (1955). Multiple range and multiple F tests. *Biometrica, 11* : 1-42.

Durley, R.C. (1983). Biosynthesis of gibberellins in higher plants. In: *Aspects of Physiology and Biochemistry of Plants Hormones.* (Ed.) Purohit, S.S., Kalyani Publishers, New Delhi and Ludhiana, pp. 93-124.

Dutta, S.C. and Basu, R. (1982). Seed germination in *Cassia sophera L. Acta Agronomica Academic Scientiarum Hungareaceae, 31* (3/4) : 332-334.

Eckardt, F.E. (1977). Physiological behaviour in relation to the Environment : A Common comparison between crops and species of the natural ecosystem. In : *Environmental Effects on Crop Physiology.* (Eds.) Landsberg, J.J. and C.V. Cutting, Academic Press, London and New York, pp. 157-172.

Egger, K. (1981). Ecofarming in the tropics : Characteristics and potentialities. *Plant Research and Development. 13* : 96-106.

Evans, L.T. (1971). Flower indunction and the florigen concept. *Ann. Rev. Plant Physiol.* Vol. 22.

Evans, L.T. (1973). The effect of light on plant growth, development and yield. In : *Plant Responses to Climatic Factors,* Proc. of the Upsala Symposium (Ed.) Slayter, R.O. UNESCO, Paris, pp. 21-35.

Evans, L.T. (1975). *Crop Physiology.* Cambridge University Press, London, pp. 273-284.

Evenari, M. (1965). Light and seed dormancy. *Encyclopedia of Plant Physiol.* Vol. II (Ed.). Ruhland, W., Springer Verlag, Berlin, pp. 804-847.

Farooqi, A.A. and Sree Ramu B.S. 2001. *Cultivation of Medicinal and Aromatic Plants.* Universities Press (India) Ltd., Hyderabad, India.

Fischer, R.A. (1973). The effect of water stress at various stages of development on yield processes in wheat. In : *Plant Response to Climatic Factors.* (Ed.) Slayter, R.O., Proc. of the Upsala Symposium, UNESCO, Paris, pp. 233-239.

Fox, J.E. (1969). The cytokinins. In : *Physiology of Plant Growth and Development.* (Ed.) Wilkins, M.B., McGraw-Hill Book Co., London, pp. 63-87.

Franz, C. 1993. Domestication of Wild growing medicinal plants. *Plant Res. and Develop* 37 : 101-111.

Gabe, V. and Black, M. (1979). Two separate photoreceptor controls of hypocotyl growth in green seedlings. *Nature,* 278 : 51-54.

Gelmond, H. (1978). Problems in crop seed germination. In : *Crop Physiology.* (Ed.) U.S. Gupta, Oxford, IBH Publishing Co., New Delhi, pp. 1-78.

Gilman, D.F., Fehr, W.R. and Barries, J.S. (1973). Temperature effects on hypocotyl elongation of soybean. *Crop. Sci. 13* : 246-249.

Goscho, G.J., Ruelke, O.C. and Wert, S.H. (1973). Residual effect of germination temperature in sugarcane. *Crop Sci. 13* : 274-276.

Govil, J.N., Pandey, J., Shivakumar, B.G., Singh, V.K., 2002. Recent progress in medicinal plants. Vol. V Crop improvement, production technology, trade and commerce, pp. 1-42, Sci. Tech. Publ. Co. L.L.C. Texas, USA.

Grierson, W., Saule, J. and Kawada, K. (1983). Beneficial aspects of physiological stress. *Hort. Rev. 4* : 179-209.

Guerrero, F.P. and Williams, W.A. (1975). Influence of temperature on the growth of *Erodium botrys* and *Trifolium subterianum. Crop Sci. 15* (4) : 533-556.

Gulati, B.C., Quereshi, N.A. and Taj-ud-din (1982). Cultivation of balladona in Kashmir. In : *Cultivation and Utilization of Medicinal Plants in India* (Eds.) Atal, C.K. and B.M. Kapur, RRL (CSIR) Jammu Tawi, pp. 262-269.

Gupta, R. (1977). Medicinal plants component of Indian forests. Indian Farming Special, Vol. I (Agriculture and Forestry) *XXVI* (II) : 96-98.

Gupta, R. (1980). Medicinal and aeromatic plants. In : *Handbook of Agriculture.* ICAR, New Delhi, 1188-1224.

Gupta, S. and Madan, C.L. (1975). Application of growth regulators like B-9, CCC and morphactin to *Dutura metal* L. *Indian J. Pharma. 38* : 44-47.

Gupta, U.S. (1975). Physiological Aspects of Dryland Farming. Oxford, IBH Pub. Co., New Delhi.

Gupta, U.S. (1977). Physiological Aspects of Crop Nutrition and Resistance. Atma Ram and Sons., Delhi.

Gupta, U.S. (1978). Crop Physiology. Oxford and IBH Publishing Co. New Delhi.

Gupta, U.S. (1984). Crop Physiology. Advancing frontiers. Oxford and IBH Publishing Co., New Delhi.

Hadas, A. (1969). Effect of soil moisture stress on seed germination. *Agron. J. 61* : 325-327.

Hadas, A. (1970). Effect of soil moisture stress on seed germination. *Israel. J. Agric. Res. 20* : 3-44.

Hadas, A. (1976). Water uptake and germination of leguminous seeds under changing external water potential in osmoticum solution. *J. Exp. Bot. 27* : 480-489.

Hadas, A. (1977). Water uptake and germination of leguminous seeds in soil of changing matric and osmotic water potentials. *J. Exp. Bot. 28* : 988-995.

Hall, A.E., Schulze, E.D. and Lange, O.L. (1976). Current perspectives of steady state stomatal responses to environment. In : *Water and Plant Life : Problems and Modern Approaches.* (Eds.) Lange, O.C., L. Kappen and E.D. Schulze, Springer Verlag, New York, pp. 169-188.

Hall, M.A., Kapuya, J.A., Sivakumaran, S. and John, A. (1977). The role of ethylene in responses of plants to stress. *Forest Sci. 8*: 217-223.

Hansen, P. (1975). Carbohydrate allocation. In : *Environmental Effects on Crop Physiology.* (Eds.) Landsberg, J.J. and C.V. Cutting, Academic Press, London and New York, pp. 247-258.

Harborne, J.B. (1973). Phytochemical Methods. Chapmann and Hall, London, New York, pp. 192-195.

Harder, H.J., Carlson, R.E. and Shaw, R.H. (1982). Yield and yield components and nutrient content of corn grain as influenced by post silking moisture stress. *Agron. J. 74* (2) : 275-278.

Harris, D.C. (1973). Photosynthesis, diffusion resistance and relative plant water content of cotton as influenced by induced water stress. *Crop. Sci. 13* : 570-573.

Harris, P. and James, A.T. (1969). Effect of low temperature on fatty acid biosynthesis in seeds. *Biochem. Biophys. Acta 187* : 13-18.

Hartmann, H.T., Flocker, W.J. and Kofranek, A.M. (1981). Plant

Science: Growth Development and Utilization of Cultivated Plants. Hall, Inc. New Jersey, U.S.A., pp. 202-243.

Hartmann, H.T., Kester, D.E., Davis, F.T. and Geneva, R.L. 2002, *Plant Propagation : Principles and Practices.* Prentice Hall, New Jersey (USA) 7 in Edition.

Hartung, W. and Witt, J. (1968). Uber, den, Einflub, der Bodenfekchtigkeit auf den Wuchsstoffgehalt von. Anastatica Liero Chuntica und *Helianthus annus. Flora Jene* (Abst.), 157 : 603-614.

Harvery, B.M.R., Lu, B.C. and Flatcher, R.A. (1974). Benzyl adenine and enzyme activity in cucumber cotyledons. *Can. J. Bot. 52:* 2581-86.

Hawkes, J.G., Lester, R.N. and Shieding, A.D. (1979). The Biology and Taxonomy of the Solanaceae. Academic Press, London and New York, pp. 367-392.

Hegarty, T.W. (1973). Temperature relations of germination in the field. In : *Seed Ecology.* (Ed.) Heydecker, W., Butterworth, London, pp. 166-201.

Hegarty, T.W. (1977a). Seed and seedling susceptibiliy to moisture stress in soil. *J. Exp. Bot. 28* : 659-668.

Hegarty, T.W. (1977b). Seed activation and germination under moisture stress. *New Phytol. 78* : 349-359.

Hermellin, T. (1973). Environmentally conditioned effects of acute gamma irradiation on growing barley plants. In : *Plant Responses to Climatic Factors.* (Ed.) Slayter, R.O. Proc. Uppsala Symposium, UNESCO Paris, pp. 121-126.

Heydecker, W. (1973). *Seed Ecology,* Butterworth, London, pp. 224-267.

Hicks, R.R. and Stricker, R.E. (1972). Plant density effect on grain yield of corn. *Agron. J. 64* : 484-487.

Hsiao, T.C. and Bradford, K.J. (1982). Physiological consequences of production (Eds.) Taylor, H.M., W.R. Jordan and Sinclear. Am. Soc. Agron. Wisconsin, U.S.A., 11-32.

Hsiao, T.C., Fereres, E., Acevedo, E. and Anderson, D.W. (1976). Water stress and dynamics of growth and yield of crop plants. In : *Water and Plant Life : Problems and Modern Approaches.* (Ed.) Lange, O.L., Springer Verlag, Berlin, pp. 176-203.

Hutchinson, J. (1973). *The Families of Flowering Plants.* 3rd ed. Oxford, Univ. Press, London, pp. 239-240.

Israeli, A.H. (1982). Unani system of medicine. In : *Cultivation and*

Utilization of Medicinal Plants (Eds.) Atal, C.K. and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 527-532.

Jain, S.K. (1979). Medicinal Plants. National Book Trust, New Delhi, pp. 157-159.

Jain, S.K. and Sastry, A.R.K. (1982). Threatened Plants of India. Botanical Survey of India and MAB, National Committee on Environmental Planning and Coordination, Deptt. of Sci. and Technology, New Delhi.

Jakhar, M.L., Singh Kazan and Kakralya, B.L. 2003, Prospective strategy for biodiversity conservation and development in medicinal and aromatic plants. pp. 145-156. In *Environmental Conservation, Depleting Resources and Sustainable Development* (Eds.) H.S. Sharma and T.I. Khan. Aavishkar Publishers, Distributors, Jaipur, India.

Jensen, E.M., Frelic, J.R. and Gifford, R.O. (1972). Emergence force of forage seedlings. *Agron. J. 64* : 635-639.

Johnson, R.R., Prey, N.M. and Moss, D.N. (1974). Effect of water stress on photosynthesis and transpiration of flag leaves and spike of barley and wheat. *Crop. Sci. 14* : 728-29.

Jones, R.L. (1969). Ethylene enhanced release of α–amylase from barley endosperm. *Plant Physiol. 43* : 442-444.

Jordan, W.R., Brown, K.W. and Thomas, J.C. (1975). Leaf age as a determinant in stomatal control of water loss from cotton during water stress. *Plant Physiol. 56* : 595-599.

Joshi, M.C. and Nigam, B.C. (1970). Autoecological studies on Rajasthan desert plants : 3. Seed output and germination in *Trianthema portulacastrum* Linn. *Trop. Ecol. 11* : 140-147.

Kaicker, U.S., Saini, H.C., Singh, H.P. and Chaudhary, B. (1978). Environmental effects on morphin content in opium poppy, (*Papaver somniferum*). *Bull. Narcotics, 30* : 69-74.

Kakralya, B.L., Baberwal, M.L. and Singh Kazan, 2003. Seed invigoration of wheat through plant growth regulation. In *Proc. International Congress on Plant Physiology* (Eds.) G.C. Srivastava (In Press).

Kapoor, S.L. and Mitra, R. (1979). Herbal Drugs in Indian Pharmaceutical Industry, N.B.R.I., Lucknow, pp. 64-67.

Kappen, L. (1981). Ecological significance of resistance to high temperature treatment. In : *Physiological Plant Ecology.* I–(Eds.) Lange, O.L., P.S. Nobel, C.B. Osmond and H. Ziegler, Springer Verlag, New York, pp. 166-189.

Katering, D.L. and Morgan, P.W. (1969). Ethylene as a component of the emanation for germinating peanut seeds and its effects on dormant Virginia type peanut seeds. *Plant Physiol. 44* : 326-330.

Kaufman, B.P. (1983). Physiology and biochemistry of gibberellins in developing shoot of grasses. In : *Aspects of Physiology and Biochemistry of Plant Hormones.* (Ed.) Purohit, S.S., Kalyani Publishers, Ludhiana and New Delhi, pp. 125-133.

Kaufman, M.R. (1969). Effect of water potential on germination of lettuce and cucurbits seeds. *Can. J. Bot. 47* : 1761-1764.

Kaufman, M.R. (1972). Water deficit and reproductive growth, In : *Water Deficit and Plant Growth.* Vol. III (ed.) Kozlowski, T.T., Academic Press, New York, pp. 91-124.

Kaufman, M.R. and Ross, K.J. (1970). Water potential, temperature and kinetin effects on seed germination in soil and solute system. *Am. J. Bot. 57* : 413-419.

Kaul, B.L. and Jutshi, U. (1982). Cultivation of *Solanum* sp. for steroids. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) Atal, C.K. and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 98-106.

Kaul, M.L.H. (1967). Studies on ecotypes and acads of *Mecardonea dianthera* II–Seed germination, seedling growth, reproductive and aggressive capacity. *Proc. Natl. Acad. Sci. India, 37* : 361-364.

Kauraw, L.P. and Chakarbarti, N.K. (1982). Thermo sensitivities of rice seeds. *International Rice Research News Letter 7* : 5-7. Central Rice Research Instt., Cuttack (India).

Kavaljian, L.G. (1980). Some current trends in Economic Botany. In: *Trends in Botanical Research.* (Eds.) Nagraj, M. and C.P. Malik, Kalyani Publishers, Ludhiana and New Delhi, pp. 275-282.

Kawakami, K. (1978). Physiology of yield of underground storage organs. In : *Crop Physiology.* (Ed.) Gupta, U.S., Oxford and IBH Publishing Co., New Delhi, pp. 269-301.

Khare, L.J. (1978). Some factors affecting seed germination of *Virgenia indica.* Kunth. : Effect of temperature and root exudation of maize on germination. *Acta Bot. Indica, 6* : 93-97.

Koch, W. (1977). Diseases, Pests and Weeds in Tropical Crops. Verlag Paul Porey, Berlin, pp. 28-55.

Koller, D. and Hadas, A. (1982). Water relation in germination of seeds. In : *Encyclopedia of Plant Physiology.* New series, Vol. 12:

B (Eds.) Lange, O.L., P.S. Nobel, C.B. Osmond and H. Ziegler, Springer Verlag, Berlin, pp. 401-431.

Koller, H.R. (1972). Leaf area and leaf weight relationships in the soybean. *Crop. Sci. 12* : 180-183.

Kozlowski, T.T. (1972). Seed Biology. Vol. I, Academic Press, London, pp. 69-83.

Kozlowski, T.T. (1972). Seed Biology. Vol. II, Academic Press, London, pp. 104-109.

Kramer, P.J. (1980). The role of physiology in crop improvement. In: *Linking Research to Crop Production.* (Eds.) Staples, R.C. and R.J. Kuhr, Plenum Press, New York and London, pp. 51-62.

Krishnamoorthy, H.N. (1975). Gibberellins and Plant Growth. Wiley Eastern Limited, New Delhi, pp. 115-144.

Krishnamoorthy, H.N. (1981). Plant Growth Substances including Their Application in Agriculture. Tata McGraw-Hill Publishing Co. Ltd., New Delhi, pp. 69-83.

Krishnan, R. (1980). Domestication of medicinal plants : Problems and prospects. In : *Current Trends in Botanical Research.* (Eds.) Nagraj, M. and C.P. Malik, Kalyani Publishers, New Delhi and Ludhiana, pp. 289-298.

Kumar, S., Singh, K. and Maheshwari, D.K. (1984). Seed germination and early seedling growth responses of brinjal to treatment with certain nematicides and growth substances. *Adv. Bio Sc.* 3.

Landsberg, J.J. (1977). Effect of weather on plant development. In: *Environmental Effects on Crop Physiology.* (Eds.) Landsberg, J.J. and C.V. Cutting, Academic Press, London, New York.

Landsberg, J.J. and Cutting, C.V. (Eds.) (1977). Environmental Effects on Crop Physiology, Academic Press, New York, pp. 1-5.

Landsberg, J.J. and Thorpe, M.R. (1975). The mechanism of apple bud morphogenesis : Analysis and a model. *Ann. Bot. 39* : 689-699.

Lange, O.L., Nobel, P.S., Osmond, C.B. and Ziegler, H. (Eds.) (1981). Physiological Plant Ecology–I : Responses to the Physical Environment. Springer Verlag, Barlin and New York, pp. 23-31.

Lange, O.L., Nobel, P.S., Osmond, C.B. and Ziegler, H. (Eds.) (1982). Physiological Plant Ecology–II : Water relations and carbon assimilation. Springer Verlag, Berlin and New York, pp. 1-4.

Larcher, W. (2002). *Physiological Plant Ecology.* Springer Verlag, Berlin and New York, pp. 295-308.

Larcher, W. 2002. *Physiological Plant Ecology.* pp. 295-308. Springer Verlag Berlin and New York.

Larson, K.C. (1975). Draught injury and resistance in crop plants. In: *Physiological Aspects of Dryland Farming.* (Ed.) Gupta, U.S., Oxford and IBH Publishing Co., New Delhi, pp. 147-165.

Lauchii, A. (1983). Inorganic plant nutrition. In : *Encyclopedia of Plant Physiology,* New series, Vol. 15 (Ed.) Lauchii, A. Springer Verlag, Berlin and New York, pp. 193-241.

Lawak, S. (1984). Hormones in seed dormancy and germination. In: *Hormonal Regulation of Plant Growth and Development.* Vol. I (Ed.) Purohit, S.S., Agro. Botanica Publishers Bikaner, (India), pp. 95-144.

Lawlor, D.W. (1969). Plant growth in polyethylene glycol solutions in relation to osmotic potential of root medium and leaf water balance. *J. Exp. Bot. 20* : 895-911.

Leopold, A.C. (1982). Hormonal regulating system in plants. In : *Recent Development in Plant Science.* (Ed.) Sen, S.P., Today and Tomorrow's Printers and Publishers, New Delhi, pp. 43-50.

Leopold, A.C. and Kriedemann, P.E. (1975). Plant Growth and Development. Tata McGraw Hill Publish. Co. Ltd., New Delhi, pp. 107-432.

Levitt, J. (1980). Plant responses to environmental stress. Vol. I. Chilling freezing and high temperature. Academic Press, New York, pp. 41-89.

Levitt, J. (1981). Plant responses to environmental stress–II. Water relations salt and other stresses. Academic Press, New York, pp. 57-107.

Lieberman, M. (1979). Biosynthesis and action of ethylene. *Ann. Rev. Plant Physiol. 30* : 533-540.

Loveys, B.R. (1979). The influence of quality of light on levels of abscisic acid in tomato plants and evidence for a novel abscisic acid metabolite. *Physiol. Plant. 46* : 79-84.

Low, V.H.K. (1975). Role of gibberellins in root and shoot growth. In: *Gibberellins and Plant Growth.* (Ed.) Krishnamoorthy, H.N., Wiley Eastern Ltd., New Delhi, pp. 101-114.

Ludwig, M.J. 2000. Indolebutyric acid in plant growth and development, *Plant Growth Regulation,* 32 : 219-230.

Lurssen, K. (1982). Manipulation of crop growth by ethylene and some implications of mode of generation. In : *Chemical Manipulation of Crop Growth and Development.* (Ed.) McLaren, J.S., Butterworths, London, pp. 47-90.

Lurssen, K. (1984). Plant responses to ethylene and ethylene releasing compounds. In : *Hormonal Regulation of Plant Growth and Development.* (Ed.) Purohit, S.S., Agro. Botanica Publishers, Bikaner, (India), pp. 307-351.

Mahala, S. 2002. Environmental Physiology of Domestication and Cultivation of Phytoremediative Halophytes. Ph.D. Thesis. Rajasthan Agricultural University, Bikaner, India.

Majur, D.J. and Johnson, D.R. (1974). Effect of light intensity on the development of field grown soybean. *Crop. Sci. 14* (6) : 839-841.

Malik, C.D. 1995. Plant Growth Regulators, Softwares for Crop and Plant Productivity. Presidential Address. Botany Section. Indian Science Congress Association, Calcutta, India.

Mann, M.S. (1984). Genetic analysis of yield and its components in durum wheat. Ph.D. thesis, Sukhadia Univ., Udaipur (Raj.).

Martin, Z. (1973). Vernalization and photoperiodism of common wheat as related to general and specific adaptability of varieties. In : *Plant Responses to Climatic Factors.* (Ed.) Slayter, R.O., Proc. of the Upsala, Symposium, UNESCO, Paris, pp. 153-161.

Mayber Poljakoff, I., Evanari, A.M. and Neumann, G. (1958). Effect of red light and GA_3 on the temperature inhibited germination in lettuce seeds. *Bull. Res. Council Israel. 6* (2) : 99-102.

McDanial, R.G. (1982). The physiology of temperature effects on plants. In : *Breeding Plants for Less Favourable Environment.* (Eds.) Christiansen, M.N. and C.F. Lewis, John-Wiley and Sons, New York, pp. 13-46.

McGree, K.J. and Davis, S.D. (1974). Effect of water stress and temperature on leaf size and number of epidermal cells in grain of Sorghum. *Crop. Sci.* 14 (5) : 751-754.

McLaren, J.S. (1982) (Ed.). Chemical Manipulation of Crop Growth and Development. Butterworths, London, pp. 1-27.

McLaren, J.S. and Smith, H. (1978). Phytochrome control of the growth and development of *Rumex obtusifolius* under simulated canopy-light environment. *Plant Cell Environ. 1* : 61-67.

Mehta, A.P. and Parihar, S.S. (1973). Seedling emergence in soybean and cotton as affected by seed bed characteristics and surface mulches. *Indian J. Agric. Sci. 43* : 45-49.

Michel, B.F. and Kaufman, M.R. (1973). The osmotic potential of polyethylene glycol '6000'. *Pl. Physiol. 51* : 914-16.

Milborrow, B.V. (1974a). Chemistry and biochemistry of abscisic acid. In : *The Chemistry and Biochemistry of Plant Hormones : Recent*

Advances in Phytochemistry. Vol. 7 (Eds.) Runeckless, V.C., E. Sondheimer and D.C. Walton, Academic Press, London, pp. 21-29.

Milborrow, B.V. (1974b). The chemistry and physiology of abscisic acid. *Ann. Rev. Plant Physiol. 25* : 259-307.

Milborrow, B.V. (1974c). Biosynthesis of abscisic acid by a cell free system. *Phytochemistry, 13* : 131-136.

Milborrow, B.V. (1981). Abscisic acid and other hormones. In : *Physiology and Biochemistry of Drought Resistance. In Plants.* (Eds.) Paleg, D. and A. Aspinall, Academic Press, London and New York, pp. 63-109.

Milburn, J.A. (1979). (ed.) Water Flow in Plants. Longman, London, New York, pp. 140-149.

Misra, K.C. (1980). Manual of Plant Ecology. Oxford and IBH Publishing Co., New Delhi, pp. 107-117.

Misra, R. and Das, R.R. (Eds.) (1971). Proceedings of the School in Plant Ecology. Oxford and IBH, New Delhi, 1-8.

Moewus, E. and Moewus, L. (1952). Sensitivity of grass roots to indole acetic acid. *Nature, London, 170* : 372-75.

Mok, D.W. and Mok, M.C. 2001. Cytokinin metabolism and action. *Annual Review of Plant Physiology and Plant Molecular Biology,* 52 : 89-119.

Moolani, M.K. and Kanitkar, U.K. (1961). Effect of treatments with 2, 4–D on germination and growth of gram (*Cicer arietinum*). *Proc. Indian Sci. Cong.* 38 (III) : 152.

Moore, R.P. (1970). Tetrazolium Testing Handbook : Contribution No. 29 to the Handbook of Seed Testing : Association of Seed Analysis, U.S.A., pp. 167-192.

Moore, T.C. (1980). Biochemistry and Physiology of Plant Hormones. Narosa Publish. House, New Delhi, pp. 112-126.

Morgan, D.C. and Smith, H. (1979). A systematic relationship between phytochrome controlled development and species habitat for plants grown in natural radiation. *Planta, 150* : 95-101.

Morgan, D.C. and Smith, H. (1981). Non-photosynthetic responses to light quality. In : *Physiological Plant Ecology–I.* (Eds.) Lange, O.L., P.S. Noble, C.V. Osmond and H. Ziegler, Springer Verlag, Berlin and New York, pp. 109-130.

Murthy, R.S. and Hirekerier, L.R. (1980). Soils. In : *Handbook of Agriculture.* ICAR, New Delhi, 5-25.

Murty, P.S.S. and Murty, K.S. (1982). Effect of reduced light at

different growth stages on the spikelet sterility in rice. *Madras Agric. Jour. 69* (2) : 121-123.

Murty, Y.S. and Singh, K. (1983). Flower abscission responses of *Vicia faba* to treatments with growth substances. *Acta Bot. Indica, 11*: 65-68.

Naik, S. (1954). Effect of α–naphthaline acetic acid on the rate of germination in Leguminous seeds. *J. Indian Bot. Soc., 33* : 153-161.

Nandwal, A.S. and Bharti, S. (1982). Effect of KN and IAA on growth, yield and nitrogen fixing efficiency of nodules in pea (*Pisum sativum*). *Indian J. Plant Physiol. 4* : 358-363.

Nickell, L.G. (1982). Plant Growth Regulators and their Agricultural Uses. Springer Verlag, New York, pp. 12-20.

Nikolaeva, M.G. (1969). Physiology of Seed Dormancy in Seeds, Israel Programme for Scientific Translation, Jerusalum, pp. 98-109.

Nilsen, E.T. and Orcutt, D.M. 1996. *Physiology of Plants under Stress Abiotic Factor.* John Wiley and Sons Inc., New York and Singapore.

Nosberger J., Geiger, H.H. and Struik, P.C. 2001. *Crop Science: Progress and Prospects.* CABI Publishing, Wallingford Oxon, U.K.

Oosting, H.J. (1958). The Study of Plant Communities. W.H. Freeman and Co., San Francisco, pp. 41-49.

Ovacharrow, K.E. (1977). Physiological Basis of Seed Germination. Amerind Publ. Co., New Delhi, pp. 81-89.

Padulosi, D., Leaman, D. and Quick, F.D. 2002. Challenges and opportunities in enhancing biodiversity conservation and uses of medicinal plants. *Herbs Spices and Medicinal Plants* 9 : 243-279.

Palmer, A.F.E. (1973). Photoperiod and temperature effects on a number of plant characters in several races of maize grown in the field. In : *Plant Responses to Climatic Factors.* (Eds.) Slayter, R.O., Proc. of the Upsala Symposium, UNESCO, Paris, pp. 113-118.

Pandey, B.N. and Sinha, R.P. (1978). Effect of storage period and temperature on germinability of the 4 species of *Crotolaria. Acta Bot. Indica,* 6 (Supp.) : 78-80.

Papadakies, J. and Gx, A.I. (1978). Toxins and crop growth. In : Crop Physiology. (Ed.) Gupta, U.S., Oxford and IBH Publishing Co., New Delhi, pp. 202-225.

Parihar, S.S. and Chaudhary, M.R. (1977). Crop responses to soil compaction : Seedling emergence and root growth. In : *Physiological Aspects of Crop Nutrition and Resistance.* (Ed.) Gupta, U.S., Atma Ram and Sons, New Delhi, pp. 289-310.

Parmer, M.J. and Moore, R.P. (1966). Effect of simulated drought of PEG solution on corn (*Zea mays L.*). Germination and seedling development. *Agron. J. 58* : 391-392.

Parson, L.R. (1982). Plant responses to water stress. In : *Breeding Plants for Less Favourable Environment.* (Eds.) Christiansen, M.N. and C.F. Lewis, John Wiley and Sons, New York, pp. 175-192.

Parups, E.V. (1980). Effect of morphactins on certain plant growth substances in bean roots. *Physiol. Plant. 29* : 56-58.

Parups, E.V. (1983). Physiology and biochemistry of morphactins. In: *Aspects of Physiology and Biochemistry of Plant Hormones.* (Ed.) Purohit, S.S., Kalyani Publisher, New Delhi, Ludhiana, pp. 255-77.

Pathak, P.S. (1967). Ecology of *Tribulus terristris* L. a medicinal plant, Ph.D. thesis, B.H.U. Varanasi.

Patten, G.P. and VanDoren, D.M. (1970). Effect of seed orientation on emergence and growth of corn. *Agron J. 62* : 592-595.

Pearce, R.B., Moek, J.J. and Bailey, T.B. (1975). A rapid method for estimating leaf area per plant. *Crop Sci. 15* : 691-694.

Penning, de Vries, F.W.T., Bruneting, A.H.M. and VanLaar, H.H. (1974). Products requirement and efficiency of biosynthesis : A qualitative approaches. *J. Theor. Biol. 45* : 339-377.

Peters, D.B. and Runkles, J.R. (1967). Shoot and root growth as effected by water availability. In : *Irrigation of Agricultural Lands.* (Eds.) Hagan, R.M., R.H. Haise and T.W. Edminster. Agro. Series : Am. Soc. Agron. Madison, Wisconsin, pp. 373-86.

Pheloung, P. and Barlow, E.W.R. (1981). Respiration and carbohydrate accumulation in water stressed wheat apex. *J. Exp. Bot. 32* : 921-931.

Pilet, P.E., Elliot, M.C. and Moloney, M.M. (1979). Endogenous and exogenous auxins in the control of root growth. *Plants, 146*: 405-408.

Pollock, B.M. (1962). Temperature control of physiological dwarfing in pea seedlings. *Plant Physiol. 37* : 190-197.

Popay, A.I. and Roberts, E.H. (1970). Factors involved in the dormancy

and germination of *Capsella bursapastoris L. Medik L. J. Ecol. 58*: 103-122.

Pulgar, C.E. and Laude, H.M. (1974). Regrowth of alpha-alpha after heat stress. *Crop Sci. 14* : 28-30.

Purohit, S.S. (1984). Hormonal regulation of plant growth and development. Vol. I. Agro Botanica Publishers, Bikaner (India), pp. 408-412.

Quisenberry, J.E. (1982). Breeding for drought resistance and plant water efficiency. In : *Breeding Plants for Less Favourable Environment.* (Eds.) Christiansen, M.N. and C.F. Lewis, John Wiley and Sons, New York, pp. 193-212.

Ramanathan, V.S. (1980). Effect of plant growth regulators on yield and morphin content in opium poppy. *Indian J. Agric. Res. 12* (4) : 246-250.

Randhawa, G.S. (1971). Plant growth regulators and gibberellins. ICAR, New Delhi, pp. 71-93.

Rao, P.N. and Reddy, B.V.N. (1978). Autoecological studies in *Indigofera linifolia* Retz. II. Germination behaviour of the seeds. First Botanical Conference. *J. Indian Bot. Soc.* 57 (Suppl.) : 78-79 (Abst.).

Raoson, W.M. and Bremner, P.M. (1974). The effect of individual grains removal on others. *Aust. J. Biol. Sci. 26* : 54-56.

Rathore, H.S. (1971). Weather and climatic factors as functions of solar radiation and other external influences. In : *Plant Ecology* (Eds.) Misra, R. and R.R. Das, Oxford and IBH Calcutta, pp. 4-16.

Rawat, R.B.S. 2003. National Medicinal Plants Board (NMPB). *Indian Forester,* 129 : 109-118.

Robertson, G.W. (1974). Wheat yield in relation to weather. *Can. J. Plant Sci. 54* : 625-50.

Rodemacher, Sankhla, N. 2002. Facing the flash flood of new knowledge and innovations : A plant physiologist in biotechnological wonderland. In : *Seeds, Bioregulants and Applied Plant Biotechnology* (Eds.) K.K. Bora, Karan Singh and Arvind Kumar, pp. 94-101. Pointer Publishers, Jaipur, India.

Rodemacher, W. 2000. Growth retardants : Effects on gibberellin biosynthesis and other metabolic pathways. *Annual Review of Plant Physiology and Plant Molecular Biology,* 51: 501-531.

Rypack, M. and Kamanicka, A. (1982). Effect of some growth substances on seed germination in *Malonia* sp. and *Viburnum lantum. Folia dendra Logica, 9* : 135-148.

Sacher, R.M. (1982). Strategies to discover plant growth regulators for agronomic crops. In : *Chemical Manipulation of Crop Growths and Development.* (Ed.) McLaren, J.S., Butterworth, London, pp. 13-18.

Sahu, M.B. (1979). Rauwolfia Serpentina-Sarpgandha. Vol. I. Botany and Agronomy. Today and Tomorrows' Printers and Publisher, New Delhi, pp. 76-102.

Sankhla, D. and Sankhla, N. (1972). Lettuce seed germination : Angatagonism between ABA and Ethrel. *Biol. Plant. 16* : 325-328.

Sankhla, N. (1970). Comparative study of the effect of CCC, morphactin and gibberellin on growth and development of *Arabidopsis theatiana* Arbid. *Info. Serv. 7* : 20-21.

Sanyal, P.K. (1982). Homeopathic pharmacy in India. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) Atal, C.K. and B.M. Kapur, RRL, CSIR, Jammu Tawi, pp. 61-82.

Sarin, Y.K. (1982). Indian raw materials for the production of tropane alkaloids *ibid.* pp. 251-258.

Sarin, Y.K. (2003). Medicinal plants raw materials for Indian drugs and pharmaceutical industry. *Indian Forester,* 129 (1) : 3-23.

Sarin, Y.K. and Gupta, S. (1968). A note on the distribution pattern of *Dioscorea deltoidea* Wall, a medicinal plant. In : *Proc. Symp. Recent Adv. Trop. Ecol. Part I.* (Ed.) Misra, R. and B. Gopal, pp. 245-247.

Saroha, A.P.S. (1981). Ecophysiological studies of some medicinal plants. Ph.D. thesis, Univ. of Udaipur (Raj.).

Saxena, M.C. (1981). Agronomy of lentils. In : *Lentils.* (Eds.) C. Webb and G. Hautin, C.A.B. Slough, England, pp. 111-129.

Schneider, G. (1970). Morphactins : Physiology and performance, *Ann. Rev. Plant Physiol. 21* : 499-536.

Schneider, G. (1972). Morphactins and plant growth regulation. In: *Hormonal Regulation of Plant Growth and Development.* (Eds.) H. Kaldeway and Y. Vergar. *Proc. Adv. Study Inst.* Izmir (1971), Verlag-Chemie. Weinkeim, pp. 317-331.

Schroder, J.M. (1998). Non-timber forest products and their potential for preserving tropical humid forests. *Plant Research and Development,* 47-48 : 108-117.

Sen, D.N. (1977). *Environment and Seed Germination of Indian Plants.* The Chronica Botanica Co., New Delhi.

Sen, D.N. (1980). Environment and Root Behaviour. Geobios International. Jodhpur (Raj.).

Sen, D.N. and Bansal, R.P. (1978). Environmental Physiology and Ecology of Plants (Prof. U.N. Chatterjee Commemoration Volume). Bishan Singh Mahendra Pal Singh (Dehradun).

Sen, D.N., Bohra, T.N. and Bhatti, P.R. (1979). Water regime and drought tolerance of three arid zone weeds. *J. Indian Bot. Soc. 58* : 120-28.

Sen, S.P. (1984). The molecular basis of hormone action. In : *Hormonal Regulation of Plant Growth and Development.* (Ed.) S.S. Purohit Agro. Bishan Publishers, Bikaner, India, pp. 1-40.

Sengupta, J.C. and Chattopadhyay, S.K. (1955). Effect of presowing seed treatment with hormones on germination of some common Indian fibre crops. *Bull. Bot. Soc. Bengal 9* : 10-13.

Shamsi, S.R.A. and Whitehead, F.H. (1977). Comparative eco-physiology of *Epilobium hirsutum* L. and *Lathyrus salicasia L.* IV–Effect of temperature and internal factors. Completion and concluding discussion. *J. Ecol. 65* : 71-84.

Shamsi, S.R.A. and Whiteland, F.H. (1974). Comparative eco-physiology of *Epilobium hirsutum* and *Lathyrus salicasia L.* I–General biology, distribution and germination. *J. Ecol. 62*: 269-280.

Sharma Ravinder, (2002). *Agro-techniques of Medicinal and Aromatic Plants* (Hindi), Daya Publishing House, Delhi.

Sharma, S.S. and Sen, D.N. (1974). A new report on secondary dormancy in certain arid zone seeds. *Curr. Sci. 43* : 486-487.

Shimokowa, K. (1984). Physiology and biochemistry of ethylene. In: *Hormonal Regulation of Plant Growth and Development.* I (Ed.) S.S. Purohit, Agro. Bot. Publi. India, Bikaner, pp. 279-306.

Shivraj, A. (1978). An Introduction to Physiology of Field Crops. Oxford and IBH Publishing Co., New Delhi.

Shrivastava, A.K. and Dey, S.C. (1982). Physiology of seed dormancy in sunflower (*Helianthus annuus L.*). *Hundariceae, 31* (1/2) : 70-81.

Simon, E.W. (1974). Phospholids and plant membrane permeability. *New Phytol. 73* : 377-384.

Simon, E.W., Minchen, A. and McMenomen, M.M. (1976). The low temperature limit for seed germination. *New Phytol. 77* : 310-311.

Singh, H.G. (1982). Cultivation of opium poppy. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) C.K. Atal and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 120-138.

Singh, H.G. and Saroha, M.S. (1970). Note on the time and methods of urea application to maize grown on soil with undulating topography. *Indian J. Agric. Sci. 40* (5) : 470-473.

Singh, K. (1979). Abscission studies in *Capsicum annuum* L. Ph.D. thesis, Meerut University, Meerut, India.

Singh, K. and Afria, B.S. (1985). Stress physiological aspects of seed germination and early seedling growth of Macrospermae and Microsperma grams. *Seed Res. 13* : 1-9.

Singh, K. and Murty, Y.S. (1983). Petiolar abscission responses to hormonal treatments in *Capsicum annum* L. varieties. *Proc. Indian Acad. Sci.* (Plant Sci.), *92* : 41-49.

Singh, K.P. and Gopal, B. (1973). The effect of photoperiods and light intensity on the growth of some weeds of crop fields. In : *Plant Responses to climatic factors.* (Ed.) R.O. Slayter. *Proc. Uppsalla Symp.* UNESCO Paris, pp. 461-73.

Singh, K.P. and Singh, K. (1981a). Stress physiological studies on seed germination and seedling growth in maize composites. *Acta Bot. Indica, 9* : 141-143.

Singh, K.P. and Singh, K. (1981b). Some biochemical changes during germination and seedling growth of maize composites in response to moisture stress. *Indian J. Agric. Chem. 14* : 173-176.

Singh, K.P. and Singh, K. (1982a). Stress physiological studies on seed germination and seedling growth of some wheat hybrids. *Indian J. Plant Physiol. 25* : 180-186.

Singh, K.P. and Singh, K. (1982b). Seed germination and seedling growth responses of lentils to moisture stress. *J. Environ. Biol. 3* : 137-150.

Singh, K.P. and Singh, K. (1983a). Seed germination and seedling growth responses of some rice cultivars to water potential treatments. *Indian J. Plant Physiol. 26* : 182-188.

Singh, K.P. and Singh, K. (1983b). Influence of simulated water stress on free proline accumulation in *Triticum aestivum L. Indian J. Plant Physiol. 26* : 319-321.

Singh, K.P. and Singh, K. (1983c). Water uptake and germination of wheat seeds under different external water potentials in osmoticum solutions. *Seed Res. 11* : 13-19.

Singh, K.P., Kumar, S. and Singh, K. (1986). Seed germination and

seedling growth responses of some Indian medicinal plants to moisture stress. *Seed Res. 14* (1) : 29-35.

Singh, Karan, Kakralya B.L. and Balraj Singh, 2000. Production physiology of Indian Medicinal Plants under Abiotic Stress. In : *Plant Productivity Under Environmental Stress.* (Eds.) K. Singh and S.S. Purohit, pp. 55-66. Agro. Bios., Jodhpur, India.

Singh, Karan and Kakralya, B.L. (2004). *Modern Phytology of Propagation. Applied and Basic* (In Hindi). Pointer Publishers, Jaipur, India.

Singh, Karan and Purohit, S.S. (2000). Plant Productivity under Environmental stress. Agro-bios, Jodhpur, India.

Singh, Karan and Singh, S. (1995). Seed invigoration in Chikpea. *Acta Botanica Indica,* 23 : 197-202.

Singh, S.J., Singh, Karan, Gupta, S.C. and Kakralya, B.L. 2003. Seed propagule banking system and applied cryobiology : Potential tools for plant biodiversity conservation and development. pp. 271-312. In : *Environmental Conservation, Depleting Resources and Sustainable Development* (Eds.) H.S. Sharma and T.I. Khan. avishkar Publishers, Distributors, Jaipur, India.

Sinha, S.K. (1977). Food Legumes : Distribution, Adaptability and Biology of Yield. FAO, Rome, *9* : 141-143.

Sircar, S.M. (1971). Plant Hormone Research in India, ICAR, New Delhi.

Skoog, F. and Amrstrong, D.J. (1970). Cytokinins : *Ann. Rev. Plant Physiol. 21* : 359-384.

Slayter, R.O. (1973). Effect of short period of water stress on leaf photosynthesis. In : *Plant Responses to Climatic Factors. Proc. Upsala Symposium,* UNESCO, Paris, pp. 202-211.

Smith, R.L. (1966). Ecology and Field Biology. Harper and Row Publishers, New York, pp. 77-82.

Sobti, S.N. and Kaul, B.L. (1982). Cultivation of *Dutura innoxia* and *D. metel.* In India. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) C.K. Atal and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 259-265.

Stanhill, G. (1977). Quantifying weather crop relationships, In : *Environmental Effects of Crop Physiology* (Eds.) J.J. Landsberg and C.V. Cutting, Academic Press, London, and New York, pp. 23-28.

Street, H.E. and Opik, H. (1970). The Physiology of Flowering in Plants : Their Growth and Development. Elsevier Publishing Company, Inc. New York.

Subrahmanyam, V.P. and Murty, Y.S. (1968). Phenological observation on peas. *Trop. Ecol. 4* : 53-60.

Summerfield, R.J. (1981). Adaptation to environment. In : *Lentils* (Eds.) C. Webb and G. Hawtin, CAB Slugh, England, pp. 91-110.

Svab, J., Maregal, G., Verzarpetri, G. and Rajki, E. (1979). Freezing treatment effects on flower and volatile substances of chammomille (*Mattricaria chammomilla L.*). Abst. of Paper presented at 27 Ann. Meeting of the Soc. Med. Plant Res., Budapest, Hungary *36* (3) : 246.

Swaminathan, M.S. (1982). Agricultural systems in the Himalayas. In : *Cultivation and Utization of Medicinal Plants.* (Eds.) C.K. Atal and B.M. Kapur, RRL, (CSIR), Jammu Tawi, pp. 29-44.

Tayal, M.S. and Gopal, R. (1976). Synergistic and antagonistic behaviour of maleic hydrazide, morphactin and gibberellic acid with reference to the seed germination in fenugreek (*Trigonella foenum graecum*). *Indian J. Plant Physiol. 19* : 71-75.

Taylor, A.G. Motes, J.E. and Kirkhan, M.B. (1982). Germination and seedling growth characteristics of three species of tomato affected by water deficit. *J. Am. Soc. Hort. Sci. 107* (2) : 282-285.

Taylor, A.G., Gipson, H.M. and Christetler, J.T. (1972). Plants under climatic stress, III. Low temperature and high light effects on photosynthetic products. *Plant Physiol. 49* : 798-802.

Taylor, H.M. and Ratliff, L.F. (1969). Root elongation rates of cotton and peanuts as a function of soil strength and soil water content. *Soil. Sci. 108* : 113-119.

Thompson, P.A. (1970). A comparison of the germination characters of species of Caryophyllaceae collected in Central Germany. *J. Ecol. 58* : 699-711.

Thompson, P.A. (1973). Geographical adaptation of seeds. In : *Seed Ecology.* (Ed.) W. Heydecker, Butterworth, London, pp. 37-41.

Tonimoto, S. and Harada, H. (1984). Hormonal regulation of flowering. In : *Hormonal Regulation of Plant Growth and Development.* I (Ed.) S.S. Purohit, Agro. Bot. Publisher, India, pp. 41-93.

Toole, V.K. (1973). Effects of light, temperature and their interaction on the germination of seeds. *Seed Sci.* and *Technol. 1* : 339-396.

Townsend, C.E. (1972). Influence of seed size and depth of planting on seedling emergence of two milkvetch species. *Agron. J. 64*: 627-630.

Townsend, C.E. and McGinnies, W.J. (1972). Mechanical scarification of gram and milkvetch seeds. *Crop. Sci. 12* : 392-394.

Townsend, C.E. and McGinnies, W.J. (1973). Factors influencing the vegetative growth and flowering in *Astragalus cicer L. Crop. Sci. 13* : 262-264.

Tran-Thanhvan, K.M., Dien, N.T. and Chlyah, A. (1974a). Regulation of organogenesis in small explants of superficial tissue of *Nicotiana tabaccum. Planta, 119* : 149-159.

Treshow, M. (1970). Environment and Plant Responses. McGraw Hill Book Co., London.

Tripathi, R.S. (1969). Ecology of *Cyperus rotundus* L. V. Reproduction by seeds. *Proc. Natl. Acad. Sci. India, 39*: 164-168.

Trivedi, S. and Tripathi, R.S. (1982). The effect of soil texture and moisture on reproductive strategies of *Spergula arvensis L.* and *Plantago major L. Weed Res. 22* (1) : 41-49.

Tumonov, I.I. (1969). Physiology of plants not killed by frost. *Izv. Akad. Nauk USSR, Ser. Biol. 4* : 464-480.

Upadhyay, U.C., Subhaish, R. and Joshi, C.R. (1981). Indian Literature on Pulses. Metropolitan Book Co., New Delhi, 6-27.

Valish, Y.A. (1980). Effect on mineral fertilizer on germination of field crops. Wissen Schoftliche Beitrage Martin Luther University, Halle Wittenberg, *20* : 249-259.

Vertapetain, B.B. (1973). Role of endogeneous metabolic water in plants under conditions of water deficit. In : *Plant Responses to Climatic Factors.* (Ed.) R.O. Slayter, *Proc. Upssala Symp.* UNESCO, Paris, pp. 227-233.

Verzar, Petri, G., Kiet, D.H. and Szoke, E. (1978). The alkaloid production in *Datura innoxia. Acta Bot. Acad. Sci. Hungary, 24* (3-4) : 351-362.

Villareal, R.O. (1980). Linking basic research to crop improvement: Progress in developing countries. In : *Linking Research to Crop Production.* (Eds.) R.C. Staples and R.J. Kuhr. Plenum Press, New York, London, pp. 7-17.

Villiers, T.A. (1972). Seed dormancy. In *Seed Biology.* Vol. II (Ed.) T.T. Kozlowski, Academic Press, London, pp. 19-24.

Walter, G.R. and Nawacki, E.K. (1978). Alkaloid Biology and Metabolism in Plants : Environmental factors and their effects on alkaloids. Plenum Press, New York, England, pp. 363-379.

Weaver, R.J. (1972). Plant Growth Substances in Agriculture, W.H. Freeman and Co., San Fransisco.

Wiesner, L.E. and Grape, D.F. (1972). Effect of temperature preconditioning and cultivation on rye grass (*Lolium* sp.) seed dormancy. *Crop. Sci. 12* : 760-764.

Witwer, S.H. (1971). Growth regulants in agriculture. *Outlook on Agriculture, 6* : 205-217.

Yang, S.F. and Pratt, H.K. (1978). The physiology of ethylene in wounded plant tissue. In : *Biochemistry of Wounded Plants Tissue* (Ed.) G. Kahl, De Gneyter, Berlin, pp. 595-622.

Yaniv, Z. and Palevitch, D. (1982). Effect of drought on the secondary metabolites of medicinal and aromatic plants. In : *Cultivation and Utilization of Medicinal Plants.* (Eds.) C.K. Atal and B.M. Kapur, RRL (CSIR), Jammu Tawi, pp. 1-2.

Young, J.A., Evans, R.A., Stevens, R. and Everctt, R.L. (1981). Germination of *Kochia prostrata* seeds. *Agron. J. 73* (6) : 957-961.

Zeevaart, J.A.D. (1967). The relation of growth regulators to flowering: Growth retardents. In : *Physiology of Flowering in Pharbitis nil.* (Ed.) G.C. Imamura, *Jap. Soc. Pl. Physiol.*, Tokyo, pp. 112-119.

Zeevaart, J.A.D. (1978). Phytohormones and flower formation. In : *Phytohormones and Related Compounds* : A comprehensive treatise. (Eds.) D.S. Lethom, P.F. Goodwin and T.J.V. Higgins, Elsevier North Holland, Amsterdam, pp. 291-327.

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