EFFECTS OF SEED PRIMING WITH PLANT GROWTH REGULATORS AND NUTRIENTS ON GROWTH AND YIELD OF COTTON (Gossypium herbaceum L.) UNDER SALINITY STRESS

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I. INTRODUCTION

Cotton remains the most miraculous fibre under the sun, even after 8,000 years. No other fibre comes close to duplicating all of the desirable characters combined in cotton. The fibre of a thousand faces and almost as many uses, cotton is noted for its versatility, appearance, performance and above all, its natural comfort. From all types of apparel, including astronauts in-flight space suits, to sheets and towels and tarpaulins and tents, cotton in today's fast-moving world is still nature's wonder fibre. It provides thousands of useful products and supports millions of jobs as it moves from field to fabric.

The most commonly cultivated species of cotton in the world include *Gossypium hirsutum*. Two additional cultivated species are *Gossypium arboreum* (which originated in the Indo-Pakistan sub-continent) and *Gossypium herbaceum* (from South Africa), which are also called “Old world” or “Asiatic cottons”. These two species of cotton with short staple length fibre are known for stress tolerance.

In India, cotton is primarily grown in dry tropical and subtropical climates at temperatures between 11 °C and 25 °C with a rainfall of 250-1500 mm. It is cultivated in India from sub-himalayan region of Punjab in the north to Tamil Nadu in South and from dry regions of Kutch to high rainfall areas of Manipur in east. It is cultivated on a large scale in Maharasthra, Gujarat, Andhra Pradesh, Karnataka, Madhya Pradesh, Punjab, Rajasthan, Haryana, Tamil nadu and Uttar Pradesh.

Cotton (*Gossypium spp.*) is an important fibre crop of India covering an area of 88.20 lakh hectares with production of 242.50 lakh bales and productivity of 467 kg lint per ha. In Karnataka, it is grown on an area of 3.81 lakh hectares producing 7.00 lakh bales with the productivity of 312 kg lint per hectare (Anon., 2006).

It is generally known that soil productivity changes based on its physical and chemical properties. The important factors effecting soil productivity are soil salinity, sodicity and ground water levels. Soil salinity and sodicity are mainly caused by natural and cultural (secondary salination) factors. While climate, topographic properties, parent material and distance to sea are natural factors; unscientific irrigation practices, irrigation water quality and poor land management are cultural factors. Saline, saline-alkali and alkali soils are usually seen in the arid and semi arid climatic conditions. In these areas, the capillary rise from shallow water table flooding and higher evapotranspiration than precipitation can cause salt accumulation at the soil surface (Wei Jang and Ronald, 2005).

The salinity effects on soil can be of chemical, physical and biological. Chemical effects are certain exchanges and interactions among the salts, whereas, the major physical effect is soil permeability (Farooqi et al., 2005). Biological effects are the changes in osmotic pressure and alteration of protoplasm and cell membrane permeability (Abd El-Samad et al., 2004).

Soil salinity has caused heavy loss of natural resources in India. Out of 329 million hectares of land in the country, about 175 million hectares (53%) are suffering from degradation in some form or the other and out of which, 7.65 million hectares is salt affected (Anon., 2006). It is mainly due to excessive and uncontrolled irrigation, accumulation of salts in the top layer due to higher evapotranspiration in arid conditions, excessive use of chemical fertilizers containing chlorides, sulfates etc. and poor drainage conditions. This problem is faced in 8 out of 15 agro-climatic zones of India, particularly in Haryana, parts of Punjab, Rajasthan, Uttar Pradesh, Gujrat, Maharashtra, Madhya Pradesh, Karnataka, Andhra Pradesh and Tamil Nadu. Substantial areas of good irrigated lands are affected by saline-alkali and water-logging problems (Irfan et al., 2005).

In Karnataka, the area under salt affected soils is estimated to be 179 thousand ha. Such salt affected soils are mainly found in Dharwad, Bijapur, Belgaum, Bellary, Chitradurga and Raichur districts. It is estimated that the saline soils are distributed in 35.9 per cent of taluks out of which 14.6 per cent have salts at injurious and critical levels for crop production.

Soluble salts in saline soils which impair soil productivity are chlorides, sulfates, carbonates and bicarbonates of calcium, magnesium, sodium and potassium. These salts cause “physiological drought” and reduce the availability of phosphorus and micronutrients such as iron, manganese, zinc, copper and cobalt to crop plants.

Cotton seeds are particularly vulnerable to salinity stress encountered between sowing and seedling establishment. Poor germination and seedling establishment are the results of soil salinity. It adversely affects the growth and development of many crop plants and results in low productivity. The effects of salinity on cotton range from reduction in
germination percentage, fresh and dry weight of shoots and roots, chlorophyll content, nitrate reductase activity to the uptake of various nutrient ions.

It is thought that depressive effect of salinity on germination could be related to declining in endogenous levels of hormone (Debez *et al*., 2001). However, pre-sowing seed treatments have been shown to enhance the vigour and synchronized stand established under stress conditions. Further pre-soaking seeds with optimal concentration of phytohormones has been shown to be beneficial to growth and yield of some crop species grown under saline conditions by increasing nutrient reserves through increased physiological activities and root proliferation (Singh and Darra, 1971).

Therefore, in order to overcome/mitigate the effect of salinity on cotton crop, an experiment was planned through seed priming with plant growth regulators and nutrients under field conditions having natural salinity gradients with the following objectives.

1. To study the effect of plant growth regulators and micronutrients on physiological and biochemical changes in cotton under various levels of salinity
2. To study the role of plant growth regulators and micronutrients in mitigating the salt stresses and their influence on growth, yield and yield attributes in cotton
II. REVIEW OF LITERATURE

Salinity is a major problem as millions of tons of salts are annually dumped on to the soil through irrigation water. The reduction in growth of many crop plants by salinity may result from its effects on dry matter production relations, water status, physiological processes, biochemical reactions or a combination of such factors (Zidan, 1991 and Malibari et al., 1993).

The effects of plant growth regulators and nutrients on physiological, biochemical, ion relations and yield and yield components of cotton genotypes under saline conditions are reviewed in this chapter.

2.1 BIOCHEMICAL CHARACTERS

2.1.1 Chlorophyll content

Singh and Jain (1981) observed that both chlorophyll ‘a’ and ‘b’ as well as chl ‘a’/’b’ ratio decreased with increase in salinity viz., 0, 4, 8 mMhos/cm both in fruit wall and seed in chickpea, however, growth regulators spray viz., kinetin (10 ppm) and GA$_3$ (25 ppm) enhanced the chlorophyll contents.

The concentration of chlorophyll, RNA content and photosynthesis decreased due to increase in concentration of Na$_2$CO$_3$ and NaHCO$_3$ salts and decrease was more in greengram compared to pea (Garg and Garg, 1985).

Malibari (1993) reported that when salt stressed (50, 100 and 150 mM NaCl) wheat plants were sprayed with kinetin (5 ppm) and abscissic acid (20 ppm) total chlorophyll content increased compared to control plants.

Both chlorophyll ‘a’ and chlorophyll ‘b’ contents gradually decreased with increasing salt intensity in mulberry, however, the rate of depletion was more with chl ‘a’ than chl ‘b’ (Ramanujulu et al., 1993). Similar reports were made by Sudhakar et al. (1991) and was attributed to the increased chlorophyllase activity and partly due to the interference of salt ions with denovo synthesis of proteins, the structural component of proteins.

Munjal and Goswami (1995) reported that NaCl (0, 3 and 9 dS m$^{-1}$ ECe) treatments decreased the total chlorophyll and carotenoid content in cotyledonary leaves of cotton. The reduction was attributed to destruction of chlorophyll and instability of pigments and proteins (Somani, 1991). Similar reduction in chlorophyll content under NaCl salinity has been reported in mungbean by Saha and Gupta (1998) and Singh et al. (1994).

Saha and Gupta (1998) reported that salinity (10 and 25 dS m$^{-1}$) decreased the accumulation of chlorophyll in mungbean plants. However, the plants treated with triazoles (5 ppm and 7 ppm) and cycocel 100 ppm showed lesser reduction in the levels of total chlorophyll content than the untreated plants grown under same salinity level.

Afria et al. (1998) reported that the cycocel spray (1000 and 1500 mg l$^{-1}$) increased the chlorophyll content significantly as compared to untreated seeds of Guar and application of saline water @ 6 and 12 dS m$^{-1}$ decreased the chlorophyll content significant as compared to control (0 dS m$^{-1}$).

The chlorophyll ‘a’ and ‘b’ and total chlorophyll contents were reduced significantly at higher concentrations of salts in mungbean cultivars and suggested that the specific enzyme which is responsible for synthesis of chlorophyll was suppressed by higher concentration of salt (Ashraf and Rasual, 1988).

Khan and Srivastava (2000) observed that maize plants raised with nitrates as a sole nitrogen source showed increase in chlorophyll content in leaves under salinity (100 mM NaCl) compared to control (0 mM NaCl).

Sahoo et al. (2001) found decreased accumulation of chlorophyll content in the leaves of both salt sensitive (Ratna) and salt tolerant (Geta) cultivars of rice with increased levels of salinity from control (0 mM NaCl) to 25, 50, 100, 200 mM NaCl. The trend was more pronounced in the susceptible cultivars than tolerant cultivars.

Salinity (0, 25, 50, 100 mM NaCl) decreased the amount of chlorophyll ‘a’ and ‘b’ and total chlorophyll content in leaves of pearl millet however, application of nitrate (2 and 10 mM) increased the chl ‘a’, ‘b’ and total chlorophyll content under salinity (Badr and Albassam, 2001).

Sivakumar et al. (2002) found that foliar spray of brassino steroid (0.1 ppm), mepiquat chloride (50 ppm), NAA (40 ppm), salicylic acid (100 ppm) and tricantanol (10 ppm) increased the chlorophyll content compared to control in pearl millet.

Sayed and Godallah (2002) found that in the absence of thiamin, chlorophyll ‘a’, ‘b’ and total chlorophyll content decreased progressively on salinization in sunflower plants.
However, this trend was modified by foliar spray of thiamin (5 and 10 mg l⁻¹) which indicated increase in the chlorophyll content in leaves.

Total chlorophyll and its fractions ‘a’ and ‘b’ contents decreased with increased salinity (0, 100 and 200 mol m⁻³ NaCl) in two spring wheat cultivars, Barani-83 (salt sensitive) and SARC-1 (salt tolerant). However, foliar application of GA₃ (100 mg l⁻¹) enhanced both the pigments in Borani-83 but total chlorophyll in both the lines (Ashraf et al., 2002).

Chlorophyll ‘a’, ‘b’ and total chlorophyll content in leaves of cucumber and melon decreased in normal irrigation water supplemented with 60 mM NaCl, however, these contents increased with supplementation of Ca(NO₃)₂ along with NaCl treatment in irrigation water (Cengiz et al., 2003). Similar results were obtained when K₂SO₄ (3 mM) was applied to the root zone along with 60 mM NaCl in tomato and pepper (Cengiz et al., 2002).

Cucumber plants grown under high salinity (100 mM NaCl) decreased the chlorophyll content, however, the concentration of chlorophyll was increased when the soil was supplemented with Ca(NO₃)₂ (Cengiz and David, 2002).

Garg et al. (2005) reported progressive and significant decrease in the chlorophyll concentration with increased salinity (0, 3, 6 and 9 dS m⁻¹), however, higher chlorophyll content was maintained in improved fertility (N₆₀, P₄₀) at all salinity levels both at vegetative and flowering stages.

El-Tayeb (2005) found that Barley seeds presoaked with 1mM salicylic acid (SA) under salinity (0, 50, 100, 150, 200 mM NaCl) increased the photosynthetic pigment like chl ‘a’, ‘b’ and caratenoids in shoots and roots of 15 day old seedlings compared to seedlings treated with NaCl alone.

2.1.2 Proline accumulation

Water stress induces characteristic change in the level of free amino acids, especially a marked increase in the amino acid proline. Accumulation of free proline under stress conditions has been taken as a criteria to screen genotypes for drought and salinity tolerance.

Plunneke and Johan (1972) observed that in cotton a marginal increase in proline content was noticed under salt stress. The total free amino acid concentration in NaCl treated cotton leaves was considerably higher than the plants grown under normal conditions. The data suggesting that sodium might have a role in synthesis of amino acids in cotton plants.

Steewart and Lee (1974) observed the accumulation of proline in halophytes and level of proline increased with increase in salinity, suggesting that capacity to accumulate proline was correlated with salt tolerance and also served as a source of solute for intercellular osmotic adjustment under saline conditions. Palfi et al. (1974) found the differential accumulation of proline in 27 species of halophytes.

Balasimha (1983) reported that the proline content in leaves of cocoa seedlings increased due to drought. Proline content was less in ABA (10 ppm) treated plants as compared to kinetin (10 ppm) and control. Abscisic acid treatment caused decreased accumulation of proline (Huber, 1979).

Singh and Singh (1999) reported that the proline content in increased with increase in salinity levels (0, 4, 8 dS m⁻¹) in shoots of tolerant genotypes (SG-11 and DHG-84-11) compared to susceptible genotypes (Pusa-256 and Phule G-5) in chickpea.

Sahoo et al. (2001) found that salinity levels at 0, 25, 50, 100, 150 and 200 mM NaCl significantly enhanced the rate of proline accumulation in the leaves of both salt sensitive (Ratna) and salt tolerant (Geta) cultivars of rice with increased salinity, the trend was more pronounced in the tolerant than the sensitive cultivar.

Badr and Albassam (2001) found increased free amino acids including proline content in salt-stressed plants (0, 25, 50 and 100 mM) compared to control, but decreased amount of free amino acid and proline was noticed when nitrate (10 mM) was applied with irrigation water to pearl millet.

Proline content increased under salt stress by 76, 217 and 119 per cent in leaf, root and nodule respectively over the control. Whereas, foliar spray of IAA, gibberlic acid and kinetin at 1.0 µm reduced the per cent increase in proline from 76 to 56, 41 and 21 respectively under salt stress in mungbean (Nandini and Subhendu, 2002).

Anuradha and Seeta Ram Rao (2002) found that the rice seedlings subjected to salinity stress showed increase in proline content and further supplementing NaCl (150 mM) with brassino steroids viz., 0.5, 1.0, 3.0 µM still enhanced the proline content.

Filiz et al. (2004) reported that free proline in seedlings of IR-28 rice cultivar increased under salt stress (120 mM NaCl), however, 24-epibrassinolide (3 µM) seed treatment caused significant decrease in free proline content.
2.1.3 Nitrate reductase activity

Nitrate is considered the primary source of nitrogen from the soil. The main function of enzyme nitrate reductase (NR) is to reduce nitrate to nitrite (Beevers and Hugeman, 1969). Being an adoptive enzyme, its activity is affected by several internal and external factors. Nitrate reductase activity was readily induced by the addition of nitrate substrate (Afridi and Hewitt, 1964) and increased as the concentration of nitrate was increased in the nutrient solution (Hageman and Flesher, 1969). The mean activity per gram fresh weight per hour of nitrate reductase was highest at the seedling stage and then declined thereafter in soybean (Liu and Hadly, 1971; Harper and Hageman, 1972 and Harper et al., 1972). The activity of nitrate reductase enzyme in cotton leaves is dependent on several variables in the growing conditions such as light, nutrition, water supply and plant age (Beevers and Hageman, 1969).

Klyshev et al. (1974) studied the effect of NaCl and Na₂SO₄ in pea seedlings on NRA and observed decreased activity of nitrate reductase due to salinity and increased accumulation of initial and final products in the chain of nitrate reduction and disrupted protein synthesis. At iso-osmotic concentrations of the salts, suppression of enzyme activity was greater with NaCl than Na₂SO₄ and a direct relationship between enzyme activity and protein content was observed.

Balasubramanian et al. (1974) observed a differential response of nitrate reductase activity in crop species. The in vivo NRA in excised leaves of Brassica campestris and safflower subjected to nitrate free treatment was lower than that in Barley and wheat. The enzyme in barley was more sensitive to stress than in wheat and there exists a genotypic difference in wheat and barley in response to salt and water stress. On the contrary Boucaud and Suos (1974) opined that NaCl greatly increased the activity of NR in Cochlearia anglica.

Khan (1996) found that nitrate reductase (NR) and nitrite reductase activities were substantially declined in leaves and roots of two soybean genotypes grown under varied amount of NaCl and Na₂CO₃ salts (0-12 dS m⁻¹). Sodium carbonate proved more inhibitory than NaCl to both the enzymes and further, leaves showed higher levels of nitrate reductase and nitrite reductase activity than roots, however, the salt induced inhibition was higher in leaves.

Khan and Srivastava (2000) reported that the inclusion of nitrate and inorganic ions like Cu²⁺, K⁺, Mn²⁺, Mg²⁺ and Zn²⁺ in assay medium increased the nitrate reductase activity in roots and leaves of maize under salinity (100 mM NaCl).

Badr and Albassam (2001) reported that the activities of nitrate reductase, nitrite reductase and glutamate synthetase were reduced under salinity (0, 25, 50 and 100 mM NaCl) but among them glutamate synthetase activity was less affected in pearl millet (Pennisetum typhoides). However, application of nitrate (10 mM) in irrigation solution partially restored the activities of above enzymes.

Sivakumar et al. (2002) reported that brassinosteroid (0.1 ppm) caused significant increase in nitrate reductase activity (24.35 µg g⁻¹ hr⁻¹) followed by NAA (23.41 µg g⁻¹ hr⁻¹). Salicylic acid (23.26 µg g⁻¹ hr⁻¹) compared to control (17.07 µg g⁻¹ hr⁻¹) at 60 DAS and 80 DAS in pearl millet.

A sharp reduction in the activity in the activity of nitrate reductase and nitrogenase in shoots and roots of two maize cultivars (cv. 323 and cv. 324) was noticed under different salinity levels (-0.2, -0.6, -1.0 and -1.6 MPa). However, Azospirillum inoculation at seedling stage stimulated nitrate reductase and nitrogenase activity in both shoots and roots of both cultivars of maize (Abd El-Samad Hamidia et al., 2004).

The activity of nitrate reductase was adversely affected by increased salinity but it was consistently higher in improved fertility conditions as compared to low fertility plants at all salinity levels in sesame (Garg et al., 2005).

Priti et al. (2005) found that nitrate reductase activity (NRA) decreased under water stress in Gomati cultivar of Japanese mint (Mentha arvensis), however, when suckers were soaked in the solution of chlormequat chloride (500 ppm and 1000 ppm) for 24 h NR activity increased.

2.1.4 Ionic distribution in leaves

Janardhan et al. (1976a) studied the cationic composition of cotton leaves subjected to different levels of salinity. Pre-sowing salinisation affected the uptake of Na⁺ and K⁺ considerably. Sodium uptake of Varalaxmi and Bhagya was much lower than in Hampi and Laxmi and also the K/Na ratio in the tissue was higher (1.60) as compared to a lower K/ha ratio of 0.63 in Hampi and 0.45 in Laxmi at the salinity level of 12 mMhos/cm which provide the support to the opinion that K/Na ratio has a role in determining salt tolerance in plants.
Further, it was suggested that, the exclusion of sodium by some selective mechanism and a higher K/Na ratio in the tissue were responsible for the relatively high tolerance shown by Varalaxmi and Bhagya and is common to many salt tolerant crops (Syed and El-Swaify, 1973).

The ability of plant to maintain a high level of potassium even under saline conditions is one of the factors determining the salt tolerance of the plant (Sinha, 1978).

Iyenger et al. (1978) observed increase in the contents of Na\(^+\), Ca\(^{2+}\), Mg\(^{2+}\) and Cl\(^-\) ions with an increase in the salinity of sea water from 10,000 ppm and 15,000 ppm in all the three varieties of cotton. Application of sea water did not alter the K\(^+\) level as its uptake was lower and there was high proportion of Ca\(^{2+}\) accumulation, with an increase in salinity. It may be because of the higher amount of calcium present in the water applied and sodium content did not hamper the uptake of calcium.

Janardhan et al. (1979) observed that with increasing saline water concentration from 4 mmhos cm\(^{-1}\) to 16 mMhos cm\(^{-1}\), there was concomitant increase in sodium content and decrease in potassium content. This however holds true at early stages than at later stages. The K/Na ratios were considerably lower at early stages of growth and cultivars Varalaxmi and Bhagya accumulated less sodium, thereby maintained higher K/Na ratios as compared to other cultivars.

Joshi et al. (1978) observed marked differences among the genotypes of wheat for accumulation of sodium and depletion in potassium content with the rise in exchangeable sodium. In Khachia, a tolerant type, the depletion of potassium was comparatively lower than the sensitive varieties HD 4502 and HD 4530, and further, tolerant varieties had lower Na/K ratio.

Banuls (1991) found that application of Ca nutrient solution restricted uptake and subsequent translocation of Na\(^+\) to the leaves and increased K\(^+\) concentrations in both roots and leaves of citrus plants and Cl\(^-\) accumulation in leaves was reduced when external Ca\(^{2+}\) concentration increased, whereas, Cl\(^-\) concentration in roots remained constant.

Saha and Gupta (1998) observed that salinity levels of 10 and 15 mMhos cm\(^{-1}\) increased both Na and Cl ions, while potassium (K) ion decreased resulting in decrease in K/Na ion ratio. However, plant growth retardants viz., triazoles (LAB-150978 5 ppm, BAS-111 7 ppm) and cyccel (100 ppm) applied as a soil drench increased the accumulation of K ion and decreased the accumulation of Na and Cl ion under salinity conditions compared to control in mungbean plants.

Varshney et al. (1998) observed that salinity caused a significant increase in Na content of shoot while K remained almost unaltered in three chickpea cultivars. Besides K/Na ratio, Ca, Mg and total N contents also decreased. However, chickpea cultivar ICC 4948 having highest K/Na ratio was the most tolerant cultivar compared to ICC 4951 and ICC 6098.

Singh and Singh (1999) found that the sodium content in shoot increased in all the chickpea genotypes with increased salt stress from 0 to 8 dS m\(^{-1}\). The sodium content was higher in susceptible genotypes (Pusa-256 and Phule G-5) than that of tolerant genotypes (SG-11 and DH-G 84-11). The values of potassium content in shoot of tolerant genotypes were significantly higher than that of susceptible genotypes, whereas in roots, the values of potassium content in susceptible genotypes were significantly higher than that of tolerant genotypes.

Uma Singh et al. (2004) reported that when urd bean seeds treated with indole acetic acid (IAA) solutions of 0, 50, 100, 150 ppm for 6 hours and exposed to salinity levels of 0, 4 and 8 dS m\(^{-1}\), the mineral distribution recorded at 72 and 96 hours after seedling indicated that the level of NPK was reduced with increasing salinity in embryo-axis, whereas the concentration of Na\(^+\) increased both the embryo-axis and cotyledons, while the concentration of N, P and K increased in embryo axis and decreased in cotyledons.

The contents of Na\(^+\), Ca\(^{2+}\) and K\(^+\) increased markedly in shoots and roots of sunflower plants due to salinization but the K\(^+\) contents and the ratio of K'/Na\(^+\) decreased 3 and 5 fold in shoot and root, respectively. However, foliar spray of thiamin (5 and 10 mg l\(^{-1}\)) greatly reduced the accumulation of Na\(^+\), Ca\(^{2+}\) and Cl\(^-\) but increased the K\(^+\) content and K'/Na\(^+\) ratio in shoots and roots of salt-stressed plants over the range of solute potential from 0 to -1.0 MPa (Sayed and Gadallah, 2002).

Ashraf et al. (2002) found that two spring wheat cultivars. Barani-83 (salt sensitive) and SARC-1 (salt tolerant) when exposed to a salinity (0, 100 and 200 mol m\(^{-3}\) NaCl) caused a marked increase in the concentrations of Na\(^+\) and Cl\(^-\) in shoots and roots of both lines.
However, with the foliar application of GA$_3$ (100 mg L$^{-1}$) accumulation of Na$^+$ and Cl$^-$ was decreased whereas, Ca$^{2+}$ was increased in both shoots and roots of both wheat lines.

Cengiz and David (2002) reported that membrane permeability, sodium and chloride ions were increased in leaves and roots of cucumber in the elevated salinity (100 and 150 mM NaCl) treatment, however, supplemented soil with Ca(NO$_3$)$_2$ decreased the Na$^+$ and Cl$^-$ and increased the concentration of K$^+$, Ca$^{2+}$ and N in leaves and roots.

Nandini and Subhendu (2002) reported that foliar spray of IAA, gibberllic acid and kinetin at 0.1 to 1.0 µM decreased the sodium and chloride contents, whereas potassium and calcium content in leaf, root and nodule increased under NaCl stress (4 dS m$^{-1}$).

Cengiz et al. (2003) reported that membrane permeability increased in normal irrigation water supplemented with 60 mM NaCl treatment for both cucumber and melon. However, supplementary Ca(NO$_3$)$_2$ (5 mM) restored membrane permeability. Sodium (Na) concentration in plant tissues increased in leaves and roots in the elevated NaCl treatment and concentrations of Ca$^{2+}$, K$^+$ and N in leaves were decreased in the high salt treatment and fully restored by supplementary Ca(NO$_3$)$_2$. Similar, results were obtained when K$_2$SO$_4$ (3 mM) applied to the root zone along with 60 mM NaCl in tomato, cucumber and pepper (Cengiz et al., 2002).

Watanabe et al. (2003) reported that concentration of sodium (Na) decreased in roots and shoots of cotton plants treated with 5-aminolevulinic (ALA) under high salinity soil compared to plants under low salinity soil.

Abd El-Samad Hamdia et al. (2004) reported that the uptake and partitioning of Na$^+$, K$^+$ and Ca$^{2+}$ varied between the two maize cultivars (cv. 323 and cv. 324) with salt concentration and Azospirillum inoculation. Sodium concentration increased with the increased salinity (-0.2, -0.6, -1.0 and -1.6 MPa) and increase in Na$^+$ was much greater in the shoots of cv. 323 (salt sensitive) compared to cv. 324 (salt tolerant). Whereas, Ca$^{2+}$, K$^+$ and K'/Na$^+$ ratio decreased with increased salinity levels in both cultivars. However, azospirillum inoculation to seedlings restricted the Na$^+$ uptake and enhanced uptake of K$^+$ and Ca$^{2+}$ in both cultivars. Azospirillum inoculation stimulated K$^+$ and Ca$^{2+}$ in shoots in the two maize cv. which intern might increase Ca$^{2+}$ and reduces K$^+$ leakage from root cells (Cramer et al., 1985; Hamidia et al., 2000 and El-Koamy et al., 2003).

Garg et al. (2005) found gradual but progressive decline in K concentration and a sharp increase in Na concentration with increasing salinity levels under both the improved and low soil fertility conditions in sesame. However, under improved soil fertility, the plants maintained higher K : Na ratio than plants grown under low fertility at all salinity levels viz., K : Na ratio decreased from 10.91 (control) to 7.32, 3.85 and 2.98 at 3, 6 and 9 dS m$^{-1}$ salinity levels respectively in improved soil fertility plants while the decrease was from 8.41 (control) to 5.00, 2.76 and 1.99 in low soil fertility plants at the corresponding salinity levels.

Barley seeds presoaked with 1 mM salicylic acid (SA) under salinity (0, 50, 100, 150 and 200 mM NaCl) increased accumulation of potassium, calcium, phosphorus and decreased the accumulation of sodium in both shoots and roots of 15 day old seedlings with increased salinity level (El-Tayab, 2005).

Sylvic (2005) found that on increased level of sodium (Na') in roots of red osier dog wood plant exposed to Na$_2$SO$_4$ was recorded in the presence of supplemental Ca$^{2+}$, whereas, there was no change in potassium and Ca$^{2+}$ levels. In shoots of seedlings treated with Na$_2$SO$_4$, the addition of Ca$^{2+}$ did not affect Na', K' and Ca$^{2+}$ levels.

Anjum et al. (2005) found that the concentration of K' and Ca$^{2+}$ was inhibited with NaCl treatments (80 mM and 160 mM). While Na' and Cl' levels increased in the different plant parts of Senna, however, CaCl$_2$ treatment (5 mM and 10 mM) enhanced the K' and Ca$^{2+}$ concentration, while the combined treatments mitigated the adverse effect caused by NaCl. Thus, calcium could alleviate the NaCl-induced inhibited of plant growth via the maintenance of net K' to Na' selectivity.

Ndayiragije et al. (2006) reported that exogenous application of putrescine (1 and 10 mM) in rice in presence of NaCl (0, 150 and 300 mM) decreased the Na' and Cl' accumulation and increased the K' and CO$_2^+$ accumulation compared to control plants.

2.2 YIELD AND YIELD PARAMETERS

Sinha (1965) found that pre-soaking rice seeds with 75 ppm NAA and IAA each and 1 M KH$_2$PO$_4$ resulted in greater production of dry matter.

Puntamkar et al. (1970) reported that pre-soaking of seeds of wheat varieties with 3 per cent solution of Na$_2$SO$_4$ increased the yield by 351.0 and 217.3 per cent in 5.227 and K-
65 genotypes respectively when compared to control, while, soaking with increased concentration of NaHCO₃, MgCl₂ and MgSO₄ salts decreased the yield.

Singh and Darra (1971) found that pre-soaking of wheat seedling with GA and IAA enhanced the plant height, while, IBA and IAA increased root length in comparison with NAA in wheat. All the hormones more or less increased the dry weight of shoots and grain yield, the optimum concentration was 200 ppm and above supra optimal concentration the activity was either ceased or decreased.

Pre-soaking of seeds with hormone solution has been found to increase salinity and alkali tolerance in crop plants. Singh and Darra (1971) observed that pre-soaking of seeds with IAA (200 ppm) increased the yield of wheat. Similarly, Chippa and Lal (1985) obtained higher yields of wheat in Raj-911 variety under saline sodic conditions by pre-soaking the seeds in IAA (200 ppm) and IBA (200 ppm) for 18 hours. Under saline conditions the naturally occurring growth hormones in plant get suppressed and the pre-soaking treatments fulfill the requirements of the optimum amount of hormones normally required by the plant for better growth (Nieman and Benstein, 1959; O’leary and Prisoo, 1970; Singh and Darra, 1971).

Darra and Saxena (1971) reported that pre-soaking of maize seeds with 100 ppm of GA resulted in increased number of grains per cob, number of effective cobs per plant, grain yield under low salinity (< 4 milli mhos cm⁻¹). While, under high salinity levels (10 mmos cm⁻¹) 200 ppm of GA could be more useful in an increasing germination and yield and yield parameters.

Dwivedi (1979a) recorded higher yield of wheat in HP-2009 variety at ESP level ranged from 29 to 51 when seeds were pre-soaked with 3 per cent calcium nitrate for 12 hours. Chippa and Lal (1985) also noticed the higher yield of wheat (Raj-911) under saline sodic soils by pre-soaking with seeds with 3 per cent Na₂SO₄ solution.

Sawan et al. (1989) observed that foliar spray of nitrogen @ 108 kg ha⁻¹, calcium @ 50 mg l⁻¹ and micronutrients Cu, Zn, Fe and Mn @ 12.5 mg l⁻¹ on Egyptian cotton cultivar “Giza-75” at 70, 80 and 100 DAS increased the fresh weight and dry weight of seedlings and yield per plant.

Maliberi (1993) found that the interactive effects of various levels of salinity (50, 100 and 150 mM NaCl) and abscissic acid (20 ppm) and kinetin (5 ppm) on wheat showed positive relationship on dry matter accumulation.

Patra et al. (1995) observed that foliar spray of nutrient solution viz., KNO₃ 0.5 per cent Ca(NO₃)₂ 0.5 per cent and urea 2.0 per cent at 50 per cent flowering enhanced the LAI, CGR, pods per plant, shelling percentage and 100-kernel weight in groundnut.

Kalita et al. (1995) reported that combined foliar spray of 3 per cent P₂O₅, 100 ppm NAA in greengram resulted in higher total dry matter accumulation, seed yield and harvest index.

Afria et al. (1998) found that saline irrigation at 12 dS m⁻¹ resulted in significant decrease in the transpiration rate, straw yield, seed yield and harvest index in Guar whereas, under same salinity the seeds soaked with cycoceol (1500 mg l⁻¹) showed significant increase in leaf area, straw yield and seed yield with reduced transpiration and harvest index.

Saha and Gupta (1998) reported that plant growth regarded vitamin, triazoles (LAB-150978 (5 ppm), BAS-111 (7 ppm)) and cycoceol at 100 ppm applied as soil drench improved growth, photosynthetic activity plant height, plant biomass and number of pods per plant of mung bean and 10 mMhos cm⁻¹ salinity, though the decline in transpiration increment of stomatal conductance was not affected.

Khan and Srivastava (2000) reported that salinity (100 mM NaCl) concentration caused reduction in root, shoot and total dry weight in two week old maize plants raised with 0 and 6 mM nitrogen as sole nitrogen source.

Number of grains per spike and grain yield per plant, 1000-grain weight and number of spike per plant decreased significantly under salt stress (0, 100 and 200 mol m⁻³) in two spring wheat cultivars, Barani-83 (salt sensitive) and SARC-1 (salt tolerant) but a more adverse effect of salt was observed on Barani-83 than on SARC-1 with respect to yield attributes. However, application of GA₃ (100 mg l⁻¹) caused a slight decrease in grains per spike and increase in thousand grain weight in both the cultivars (Ashraf et al., 2002).

Foliar spray of brassinosteroid, salicylic acid, NAA and mepiquat chloride and 0.1, 100, 40, 50 ppm increased the grain yield in pearl millet. Among the treatments, brassinosteroid recorded the maximum yield of 3591 kg ha⁻¹ followed by tricontanol (3505), NAA (3484) and salicylic acid (3427) as compared to control (3018 kg ha⁻¹) (Sivakumar et al., 2002).
Cengiz and David (2002) found that cucumber plants grown under high salinity (100 mM NaCl) decreased total dry matter and fruit yield compared to control treatment. However, when soil was supplemented with Ca(NO$_3$)$_2$ increased the dry matter, fruit yield compared to the high salt treatment.

Cengiz et al. (2003) found that plants irrigated with water containing high NaCl treatment (60 mM) produced less dry matter, fruit yield than the normal irrigated water treatments in both cucumber (Cucumis sativus cv. Orlando) and melon (Cucumis melo cv. Ananas) species. However, supplementing irrigation water with 5 mM Ca(NO$_3$)$_2$ resulted in increased dry matter and fruit yield over plants irrigated with saline water.

Abd El-Samad et al. (2004) found lack of negative response to increase in NaCl concentration (-0.2, -0.6, -1.0 and -1.6 MPa) for water content, dry matter yield and leaf area of two maize cultivars (cv. 323 and cv. 324). The rate of decline in growth parameters was greater at higher salinity and in the sensitive cv. 323 compared to the salt tolerant cv. 324. However, maize seedlings were treated with Azospirillum reversed the inhibitory effect of salinity on growth of two maize cultivars.

Kirankumar et al. (2005) reported that foliar spray of NAA (20 ppm) increased the plant height, dry weight, rate of photosynthesis and seed cotton yield. While mepiquat chloride (50 ppm) sprayed at 90 DAS was more effective than chlomequat chloride (375 and 500 ppm) in reducing plant height and leaf area, higher photosynthesis boll weight and cotton yield.

Increasing salinity levels progressively and significantly decreased seed yield and dry matter production under both improved and low soil fertility conditions in sesame. However, the reduction was less under improved fertility as compared to low fertility condition at all levels of salinity (Garg et al., 2005).

Anjum et al. (2005) reported that biomass of the root, shoot and leaf decreased significantly with each NaCl treatment (80 mM and 100 mM) compared to control in Senna plants, however, the increased biomass was observed with CaCl$_2$ treatments (5 mM and 10 mM) under stress.
III. MATERIAL AND METHODS

An investigation was made with an objective to mitigate salt stress by soaking cotton seeds of var. RAHS-14 (*Gossypium herbaceum*) with growth regulators (IAA and GA) and nutrients (Na$_2$SO$_4$ and Ca(NO$_3$)$_2$) during 2005-06 at Agricultural Research Station (ARS), Gangavati, Koppal district, Karnataka under natural soil salinity levels of <2, 6 and 12 dS m$^{-1}$.

### 3.1 EXPERIMENTAL SITE

The experiment was conducted in B-8 block where the desired salinity levels (<2, 6 and 12 dS m$^{-1}$) existed.

#### 3.1.1 Soil and its characteristics

The experiment was carried out on a saline vertisol with clay (44%) texture. Soil depth ranged from 0.75 to 1.5 m. The site had the pH in the range of 8.2 to 9.19. Organic clay content was 0.25 per cent with CaCO$_3$ content in the range of 10.2 to 11.2 per cent.

#### 3.1.2 Climate

The rainfall data and monthly mean maximum and minimum temperature data during 2005-06 are depicted in Table 1.

#### 3.1.3 Experimental details

Cotton seeds were soaked with growth regulators and nutrients for 24 hrs, shade dried till to attain original seed moisture level and was tested under three salinity levels viz., <2, 6 and 12 dS m$^{-1}$ in the field at ARS, Gangavati under natural soil salinity gradient.

#### 3.1.3.1 Genotype

The relatively salt tolerant RASH-14 variety belonging to the species *G. herbaceum* was used for the study.

#### 3.1.3.2 Salinity levels (Main plot)

<table>
<thead>
<tr>
<th>Salinity levels (dS m$^{-1}$)</th>
<th>Electrical conductivity (EC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$S_1$</td>
<td>&lt; 2 dS m$^{-1}$ (control)</td>
</tr>
<tr>
<td>$S_2$</td>
<td>6 dS m$^{-1}$</td>
</tr>
<tr>
<td>$S_3$</td>
<td>12 dS m$^{-1}$</td>
</tr>
</tbody>
</table>

#### 3.1.3.3 Treatments (Sub plot)

The following growth regulators and nutrients were used for seed soaking for 24 hours with the proportion of 1:1 w/v and then shade dried till the seeds attain original seed moisture and then used for sowing.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>$G_1$ – IAA</td>
<td>200 ppm</td>
</tr>
<tr>
<td>$G_2$ – GA</td>
<td>10 ppm</td>
</tr>
<tr>
<td>$G_3$ – Na$_2$SO$_4$</td>
<td>3 per cent</td>
</tr>
<tr>
<td>$G_4$ – Ca(NO$_3$)$_2$</td>
<td>3 per cent</td>
</tr>
<tr>
<td>$G_5$ – Control</td>
<td>-</td>
</tr>
</tbody>
</table>

**Treatment combinations**

1. $S_1G_1$ (<2 dS m$^{-1}$ + IAA @ 200 ppm)
2. $S_1G_2$ (<2 dS m$^{-1}$ + GA @ 10 ppm)
3. $S_1G_3$ (<2 dS m$^{-1}$ + 3% Na$_2$SO$_4$)
4. $S_1G_4$ (<2 dS m$^{-1}$ + 3% Ca(NO$_3$)$_2$)
5. $S_1G_5$ (<2 dS m$^{-1}$ + Control)
6. $S_2G_1$ (6 dS m$^{-1}$ + IAA @ 200 ppm)
7. $S_2G_2$ (6 dS m$^{-1}$ + GA @ 10 ppm)
8. $S_2G_3$ (6 dS m$^{-1}$ + 3% Na$_2$SO$_4$)
9. $S_2G_4$ (6 dS m$^{-1}$ + 3% Ca(NO$_3$)$_2$)
10. $S_2G_5$ (6 dS m$^{-1}$ + Control)
11. $S_3G_1$ (12 dS m$^{-1}$ + IAA @ 200 ppm)
12. $S_3G_2$ (12 dS m$^{-1}$ + GA @ 10 ppm)
13. $S_3G_3$ (12 dS m$^{-1}$ + 3% Na$_2$SO$_4$)
14. $S_3G_4$ (12 dS m$^{-1}$ + 3% Ca(NO$_3$)$_2$)
15. $S_3G_5$ (12 dS m$^{-1}$ + Control)

#### 3.1.3.4 Design and plan of layout

The field was divided into blocks of 5.4 m length for each salinity level. The natural salinity level in each block was uniform. Treatment combinations were allotted to blocks in split plot design with a main plot size of 31.5 x 12.6 m (salinity levels) and sub plot size of 5.4...
Table 1. Weather data of Agricultural Research Station, Gangavathi during August 2005 to July 2006

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Months</th>
<th>Rainfall (mm)</th>
<th>Temperature (°C)</th>
<th>Mean relative humidity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Mean maximum</td>
<td>Mean minimum</td>
</tr>
<tr>
<td>1</td>
<td>August, 2005</td>
<td>88.5</td>
<td>28.81</td>
<td>21.24</td>
</tr>
<tr>
<td>2</td>
<td>September</td>
<td>108.75</td>
<td>30.71</td>
<td>20.74</td>
</tr>
<tr>
<td>3</td>
<td>October</td>
<td>59.5</td>
<td>30.65</td>
<td>22.28</td>
</tr>
<tr>
<td>4</td>
<td>November</td>
<td>-</td>
<td>29.60</td>
<td>17.06</td>
</tr>
<tr>
<td>5</td>
<td>December</td>
<td>-</td>
<td>31.29</td>
<td>16.88</td>
</tr>
<tr>
<td>6</td>
<td>January, 2006</td>
<td>-</td>
<td>31.48</td>
<td>17.11</td>
</tr>
<tr>
<td>7</td>
<td>February</td>
<td>-</td>
<td>33.17</td>
<td>16.60</td>
</tr>
<tr>
<td>8</td>
<td>March</td>
<td>-</td>
<td>35.80</td>
<td>20.60</td>
</tr>
<tr>
<td>9</td>
<td>April</td>
<td>-</td>
<td>41.20</td>
<td>25.30</td>
</tr>
<tr>
<td>10</td>
<td>May</td>
<td>30.0</td>
<td>43.51</td>
<td>26.56</td>
</tr>
<tr>
<td>11</td>
<td>June</td>
<td>12.0</td>
<td>36.29</td>
<td>24.32</td>
</tr>
<tr>
<td>12</td>
<td>July</td>
<td>15.0</td>
<td>34.57</td>
<td>25.46</td>
</tr>
</tbody>
</table>
x 4.0 m (treatments) and it was replicated thrice.

3.1.3.5 Land preparation
The land was ploughed with tractor and harrowed twice to bring the soil to fine tilth before sowing.

3.1.3.6 Fertilizer application and after care
Fertilizer (40:20:20 NPK kg ha\(^{-1}\)) application and after care operations were carried out as per the package of practices recommendation for the zone.

3.1.3.7 Sowing
Seeds were soaked with growth regulators and nutrients for 24 h and shade dried till they attain original seed moisture with the proportion of 1:1 w/v and were then used for sowing with a row spacing of 90 cm and an intra row spacing of 20 cm. After emergence, gap filling operation was carried out to maintain the required plant population.

3.2 OBSERVATIONS

3.2.1 Soil characters
The following soil characters were analyzed before sowing and later at 45, 90, 120 DAS and at the time of harvest of crop and are presented in Table 2.

3.2.1.1 Soil reaction (pH)
Soil pH was measured in soil water (1:2.5) suspension using glass electrode pH meter (Jackson, 1973).

3.2.1.2 Electrical conductivity (EC)
Electrical conductivity of clear soil solution was determined in 1:2.5 soil water suspension by using conductivity meter (Jackson, 1973) and converted to ECe using standard factor.

3.2.1.3 Determination of water soluble cations (me/l)
Preparation of 1:2.5 soil water extract

Twenty grams of soil was transferred to a conical flask and 50 ml of distilled water was added and kept on the mechanical shaker for half an hour and filtered. A known quantity of aliquot was taken for each of the following estimations.

3.2.1.3.1 Estimation of calcium and magnesium
Calcium and magnesium was determined in soil samples collected at before sowing, then at 45, 90, 120 DAS and at the time of harvest of crop, following method of Chapman and Pratt (1961).

a. Calcium plus magnesium
An aliquot of ten ml was pipetted into porcelain dish. To this dish ten drops of NH\(_4\)Cl + NH\(_2\)OH buffer (pH-10.00), 10 drops of potassium cyanide and five to six drops of Erichrome Black T (EBT) indicator was added. Mixed thoroughly with a glass rod and titrated against standard ethylene diamine tetra-acetic acid (EDTA) solution of 0.01 N till the wine red colour turned to sky blue. A blank was run without soil on for the sample and deducted the blank reading of Ca + Mg from sample reading. Calcium and magnesium content was expressed in milli equivalents per litre.

b. Calcium
A ten ml aliquot from soil water suspension was pipped into porcelain dish. To this dish five ml of NaOH (10%) was added to maintain pH 11 and ten drops of potassium cyanide and a pinch of meroxide indicator was added. Mixed thoroughly with a glass rod and titrated against standard EDTA solution of 0.01N till the orange red colour turned to purple or lavender. A blank was run without soil as for the sample similar to Ca + Mg determination and calcium content was expressed in milli equivalents per litre.

c. Magnesium
The difference of a and b was taken as magnesium content and expressed in milli equivalents per litre.

3.2.1.3.2 Estimation of sodium and potassium
Soluble sodium and potassium in known volume of aliquot was determined by running clear extract on flame photometer (Jackson, 1973).

3.2.1.3.3 Sodium adsorption ratio (SAR)
Sodium adsorption ratio of soil solution was calculated using the following formula.

\[
\text{SAR} = \frac{Na^+}{\sqrt{Ca^{2+} + Mg^{2+}}} \times 2
\]
3.2.2 Physiological and biochemical characters

The following physiological and biochemical parameters were analyzed in the leaves of cotton at 45, 90 and 120 DAS.

3.2.2.1 Total chlorophyll content

Chlorophyll content in the leaves of cotton of different treatments subjected to different salinity levels was determined colorimetrically as per the DMSO (Di-methyl sulfoxide) method of Shoaf and Lium (1976).

Fresh leaf tissue of 100 mg was cut into small pieces and incubated in 7.0 ml of DMSO at 65°C for 30 minutes. At the end of incubation period, supernatant was decanted and leaf tissue was discarded. Volume was made to 10 ml with DMSO and absorbance of extract was read at 645, 652 and 663 nm using DMSO as blank. The total chlorophyll contents were calculated using the formulae.

\[
\text{Chlorophyll 'a'} = 12.7 \times (A_{663}) - 2.69 \times (A_{645}) x \frac{V}{1000 \times W \times a}
\]

\[
\text{Chlorophyll 'b'} = 22.9 \times (A_{645}) - 4.68 \times (A_{663}) x \frac{V}{1000 \times W \times a}
\]

Total chlorophyll = Chlorophyll 'a' + Chlorophyll 'b'

Where,
- A = Absorbance at specific wave length (645 and 663 nm)
- V = Final volume of the chlorophyll extract (ml)
- W = Fresh weight of the sample (g)
- a = Path length of light (1 cm)

3.2.2.2 Determination of free proline content (µg g\(^{-1}\) fresh weight)

Free proline content in the leaves of cotton of different treatments subjected to different salinity levels was determined colorimetrically as per the method of Bates et al. (1973).

A known weight (0.5 g) of fresh leaf sample was ground well in a mortar using fine sand and extracted with ten ml of 3.0 per cent sulphosalicylic acid. The extract was filtered and 2.0 ml of the filtrate was used for proline estimation. To this 2.0 ml of filtrate, 2.0 ml of acid ninhydrin reagent (2.5 g of ninlydrin was dissolved in 40 ml of 6 M orthophosphoric acid and 60 ml of glacial acetic acid), 2.0 ml of glacial acetic acid were added and placed in a boiling water bath for one hour. Following this test tubes containing the samples were transferred to a separating funnel and 6.0 ml of toluene was added, shaken thoroughly and allowed for few minutes for separation of two layers.

The lower layer was discarded and the upper toluene layer containing the colour complex was read in colorimeter at 520 nm. The proline content was calculated using the following formula.

\[
\text{Proline content (µg g}\(^{-1}\) fresh weight) = \frac{36.2311 \times OD \times V \times d}{2 \times f}
\]

Where,
- OD = Optimal density at 520 nm
- V = Total volume of extract in ml
- d = Fresh weight/dry weight ratio
- f = Milligrams of fresh weight taken for proline estimation
- 2 = Volume of the extract taken for proline estimation

3.2.2.3 Estimation of nitrate reductase activity

The activity of nitrate reductase enzyme was estimated in leaf samples as described by Hageman and Hucklesby (1971).

Twenty leaf discs were collected randomly using a hand punch from all the leaves on a plant and five plants were selected for the purpose. These discs were floated on 0.1 M
**LEGEND**

Genotype : RAHS-14  
Salinity levels (dS m\(^{-1}\)) : Three (Main plot)  
\[ S_1 - < 2 \text{ dS m}^{-1} \text{ (control)} \]  
\[ S_2 - 6 \text{ dS m}^{-1} \]  
\[ S_3 - 12 \text{ dS m}^{-1} \]  

Treatments : Five (Sub plot)  
\[ G_1 \text{ – IAA @ 200 ppm} \]  
\[ G_2 \text{ – GA @ 10 ppm} \]  
\[ G_3 \text{ – Na}_2\text{SO}_4 \text{ 3 per cent} \]  
\[ G_4 \text{ – Ca(NO}_3\text{)}_2 \text{ 3 per cent} \]  
\[ G_5 \text{ – Control} \]  

Treatment combinations
1) \[ S_1G_1 \text{ (<2 dS m}^{-1} \text{ + IAA @ 200 ppm)} \]  
2) \[ S_1G_2 \text{ (<2 dS m}^{-1} \text{ + GA @ 10 ppm)} \]  
3) \[ S_1G_3 \text{ (<2 dS m}^{-1} \text{ + 3% Na}_2\text{SO}_4) \]  
4) \[ S_1G_4 \text{ (<2 dS m}^{-1} \text{ + 3% Ca(NO}_3\text{)}_2) \]  
5) \[ S_1G_5 \text{ (<2 dS m}^{-1} \text{ + Control)} \]  
6) \[ S_2G_1 \text{ (6 dS m}^{-1} \text{ + IAA @ 200 ppm)} \]  
7) \[ S_2G_2 \text{ (6 dS m}^{-1} \text{ + GA @ 10 ppm)} \]  
8) \[ S_2G_3 \text{ (6 dS m}^{-1} \text{ + 3% Na}_2\text{SO}_4) \]  
9) \[ S_2G_4 \text{ (6 dS m}^{-1} \text{ + 3% Ca(NO}_3\text{)}_2) \]  
10) \[ S_2G_5 \text{ (6 dS m}^{-1} \text{ + Control)} \]  
11) \[ S_3G_1 \text{ (12 dS m}^{-1} \text{ + IAA @ 200 ppm)} \]  
12) \[ S_3G_2 \text{ (12 dS m}^{-1} \text{ + GA @ 10 ppm)} \]  
13) \[ S_3G_3 \text{ (12 dS m}^{-1} \text{ + 3% Na}_2\text{SO}_4) \]  
14) \[ S_3G_4 \text{ (12 dS m}^{-1} \text{ + 3% Ca(NO}_3\text{)}_2) \]  
15) \[ S_3G_5 \text{ (12 dS m}^{-1} \text{ + Control)} \]
Fig. 1: Plan of layout of field experiment

Fig. 1 Plan of layout of field experiment
KNO₃ with their lower surface facing upward and kept under bright light for one hour to make the stomates to expand fully. From this ten discs were selected at random, their fresh weight recorded and again floated in a small, narrow mouthed tube containing 2.5 ml of 0.1 M KNO₃ and 2.5 ml of phosphate buffer (pH 7.5). The discs floated on the solution were infiltrated using a vacuum pump till the discs settled down (30 minutes), after which the solution along with discs was incubated for 30 minutes at 30°C. At the end of this period, leaf discs were removed and the solution was placed on a boiling water bath for ten minutes to terminate the reaction. This extract was used for the estimation of nitrate reductase activity.

An aliquot of 2.0 ml from each sample was pipetted out into another clean test tube to which 2.0 ml of 1 per cent sulphanilamide and 2.0 ml of 0.01 per cent N-Naphthalene Diamine Dihydrochloride (NNEDA) was added. The colour developed at room temperature was read in colorimeter at 540 nm and nitrate concentration in the enzyme extract was expressed in nanomoles using KNO₃ standard curve.

**Preparation of KNO₃ standard curve**

The KNO₃ solution was made from 0.2 to 1.0 nano mole/ml with water. From this, 7.0 ml was made each to having different concentrations to which 1 ml of each sulphanilamide and naphthalene diamine dihydrochloride (NNEDA) were added and allowed for to stand 20 minutes at room temperature. The absorbance were read at wave length of 540 nm and the graph was plotted by OD values verses with respective concentrations of KNO₃.

**3.2.2.4 Determination of sodium and potassium (%)**

Sodium and potassium were determined in leaf samples collected at 45, 90 and 120 days after sowing following the method of Chapman and Pratt (1961).

The oven dried plant sample was ground to a fine powder and 100 mg sample was digested with triacid mixture (HNO₃ : H₂SO₄ : HClO₄ in the proportion of 10:1:4 respectively). The digested sample was filtered through acid washed filter paper and the volume was made up to 100 ml with distilled water. The amount of sodium and potassium was estimated by using flame emission spectrophotometer. The total sodium and potassium was then calculated in percentage.

**3.2.2.5 Determination of calcium (%)**

Calcium was determined in the leaf samples collected at 45, 90 and 120 days after sowing following versenate titration of diacid digested samples (Chapman and Pratt, 1961).

The dried sample was ground to fine powder and digested with triacid mixture (HNO₃ : H₂SO₄ : HClO₄) in the proportion of 10:1:4 respectively). The digested sample was made to 100 ml with distilled water. From this 25 ml of the aliquot was taken into a 100 ml volumetric flask, to which 3 to 4 drops of bromocresol green (0.1%) and 2.0 ml of zirconyl oxichloride solution (2%) were added. The ammonium hydroxide (1:1) was added dropwise until the colour turned to blue, volume was made up to 50 ml with distilled water mixed thoroughly and filtered through Whatman No. 44 filter paper. From this 10 ml of the filtrate was pipetted out into a porcelain dish and diluted to about 25 ml. To this dish five drops of 4 N NaOH, 50 mg of murexide indicator and 3-4 drops of 1 per cent potassium ferrocyanide was added, mixed thoroughly with a glass rod and titrated against standard EDTA solution of 0.01 N till the orange red colour turned to lavender or purple. A blank (without plant material) was run as for the sample to check for contamination and purity of reagents. Calcium content was expressed in meq per 100 gram dry weight.

**3.2.3 Yield and its components**

**3.2.3.1 Yield of seed cotton per plant**

Cotton was picked separately from the tagged plants. The number of bolls picked during each picking were counted and weighed. From this mean yield of seed cotton per plant over all pickings was worked out.

**3.2.3.2 Seed cotton yield (kg ha⁻¹)**

Seed cotton obtained from net plot area from various pickings were considered for computation of cotton yield per hectare and expressed as kilograms per hectare.

**3.2.3.3 Total number of good bolls per plant**

At each picking, the total number of good bolls were counted from the tagged plants. From this mean total number of good bolls per plant was worked out.

**3.2.3.4 Total number of bad bolls per plant**

At each picking, the total number of bad bolls were counted from the tagged plants. From this mean total number of bad bolls per plant was worked out.
Table 2. The chemical properties of soil at different stages

<table>
<thead>
<tr>
<th>Time</th>
<th>Salinity levels</th>
<th>Soil depth (cm)</th>
<th>pH (1:2.5)</th>
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<th>Water soluble (1:2.5) cations (me l$^{-1}$)</th>
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<td>11.92</td>
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3.2.3.5 Number of bolls harvested per plant
At each picking, the total number of bolls picked from the tagged plants were counted and from this number of bolls harvested per plant was worked out.

3.2.3.6 Mean boll weight
It was worked out by dividing the seed cotton yield per plant by the number of bolls harvested per plant.

3.2.3.7 Harvest index (HI)
Harvest index was calculated by using the formula of Donald (1962) and expressed in percentage.

\[
\text{Harvest index (\%)} = \frac{\text{Economic yield}}{\text{Biological yield}} \times 100
\]

3.3 STATISTICAL ANALYSIS
Data recorded on various characters were subjected for Fisher's method of analysis of variance and interpretation of data was done as given by Gomez and Gomez (1984). The level of significance used in 'F' and 't' tests were $P=0.05$. Critical difference values were calculated whenever the F test was significant.

Correlation for various physiological, biochemical and yield and its components were worked out by the method as suggested by Steel and Torrie (1960).
IV. EXPERIMENTAL RESULTS

The experiment was conducted during the year 2005-06 to investigate the effect of seed priming with plant growth regulators (IAA and GA) and nutrients (\(\text{Na}_2\text{SO}_4\) and \(\text{Ca(NO}_3\text{)}_2\)) on RAHS-14 (\(\text{G. herbaceum}\)) cotton genotype under three salinity levels viz., \(<2.0, 6.0, 12.0\) dS m\(^{-1}\). The effect of these treatments on physiological and biochemical parameters and yield and yield components were analyzed. The data recorded on these parameters were statistically analyzed and experimental results obtained are presented in this chapter.

4.1 PHYSIOLOGICAL AND BIOCHEMICAL PARAMETERS

4.1.1 Chlorophyll content (mg g\(^{-1}\) fresh weight)

Significant differences were observed in the chlorophyll ‘a’ content among the treatments and salinity levels at all the stages. However, seed priming effects at 45 DAS and interaction effects at all stages were non-significant (Table 3).

Irrespective of salinity levels, seed priming with IAA had significantly higher chlorophyll ‘a’ (0.94 and 1.09 mg g\(^{-1}\)) at 90 and 120 DAS respectively. The treatments, GA (0.78) and \(\text{Na}_2\text{SO}_4\) (0.76) were on par with control (0.72) at 90 DAS whereas, at 120 DAS, the treatments GA (1.01), \(\text{Na}_2\text{SO}_4\) (1.04) and \(\text{Ca(NO}_3\text{)}_2\) (1.03) were on par with IAA (1.09). Least chlorophyll ‘a’ content was found in control at all the stages.

Chlorophyll ‘a’ content decreased significantly from a salinity level of S\(_1\) (<2.0 dS m\(^{-1}\)) to S\(_3\) (12.0 dS m\(^{-1}\)) to the tune of 25, 26 and 20 per cent at 45, 90 and 120 DAS respectively.

4.1.1.2 Chlorophyll ‘b’ content (mg g\(^{-1}\) fresh weight)

Significant differences for chlorophyll ‘b’ content were observed among the treatments and salinity levels. However, salinity levels at 90 DAS and interaction effect was found non-significant at all the three stages (Table 4).

The treatment IAA (G\(_1\)) had significantly higher chlorophyll ‘b’ content at 45, 90 and 120 DAS (0.46, 0.67 and 0.75, respectively) compared to control. However, the treatment GA (0.44), \(\text{Na}_2\text{SO}_4\) (0.42) and \(\text{Ca(NO}_3\text{)}_2\) (0.40) were on par with IAA at all the stages. In general, all the treatments had lesser chlorophyll ‘b’ content as compared to chlorophyll ‘a’ at all the stages.

The chlorophyll ‘b’ content decreased from 0.48 to 0.34 mg g\(^{-1}\), 0.66 to 0.55 and 0.80 to 0.60 mg g\(^{-1}\) fresh weight at 45, 90 and 120 DAS respectively with increase in salinity level from < 2.0 to 12.0 dS m\(^{-1}\).

The treatment IAA had lower reduction in chlorophyll ‘b’ content, whereas, control (G\(_5\)) had maximum reduction at all the stages.

4.1.1.3 Total chlorophyll content (mg g\(^{-1}\) fresh weight)

Significant differences among treatments and salinity levels were observed for total chlorophyll content at 45, 90 and 120 DAS (Table 5).

Irrespective of salinity, IAA had significantly higher total chlorophyll content of 0.95, 1.60 and 1.85 mg g\(^{-1}\) fresh weight at 45, 90 and 120 respectively. The seed priming with GA, \(\text{Na}_2\text{SO}_4\) and \(\text{Ca(NO}_3\text{)}_2\) were on par with IAA and at 45 and 120 DAS. However, at 90 DAS the treatment \(\text{Ca(NO}_3\text{)}_2\) (1.48) was on par with IAA. The control (G\(_5\)) treatment recorded lower total chlorophyll content at all the stages. The total chlorophyll content was maximum at 120 DAS compared to 30 and 90 DAS.

In general, as the salinity level increased from < 2 to 12 dS m\(^{-1}\), the total chlorophyll content decreased significantly from 1.01 to 0.74 mg g\(^{-1}\), 1.58 to 1.24 mg g\(^{-1}\) and 1.92 to 1.50 mg g\(^{-1}\) fresh weight at 45, 90 and 120 DAS respectively. The per cent reduction in chlorophyll content with increase in salinity was less in IAA, whereas, maximum reduction was noticed in control at all the stages.

Interaction effect was found to be non-significant at all the stages.

4.1.2 Proline content (\(\mu\)g g\(^{-1}\) dry weight)

Significant difference in the proline content was observed among treatments and salinity levels. However, interaction effect was non-significant among the treatments and salinity levels (Table 6).

The treatment IAA had significantly higher proline content at 45, 90 and 120 DAS (59.32, 95.23 and 148.74, respectively).
Table 3: Effect of plant growth regulators and micronutrients on chlorophyll ‘a’ content in leaf (mg g\(^{-1}\) fresh weight) at different plant growth stages under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S(_1)</td>
<td>S(_2)</td>
<td>S(_3)</td>
</tr>
<tr>
<td>G(_1)-IAA @ 200 ppm</td>
<td>0.56</td>
<td>0.48</td>
<td>0.41</td>
</tr>
<tr>
<td>G(_2)-GA @ 10 ppm</td>
<td>0.54</td>
<td>0.54</td>
<td>0.42</td>
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<td>G(_3)-Na(_2)SO(_4) @ 3%</td>
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<td>0.45</td>
<td>0.42</td>
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<tr>
<td>G(_4)-Ca(NO(_3))(_2) @ 3%</td>
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<td>0.47</td>
<td>0.39</td>
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<td>G(_5)-Control</td>
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<td>0.45</td>
<td>0.36</td>
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<tr>
<td>Mean</td>
<td>0.53</td>
<td>0.48</td>
<td>0.40</td>
</tr>
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</table>

For Comparing Treatments (G)  
Salinity (S)  
Interaction (G x S)

S\(_1\): < 2.0 dS m\(^{-1}\)  
S\(_2\): 6.0 dS m\(^{-1}\)  
S\(_3\): 12.0 dS m\(^{-1}\)
The treatment Ca(NO$_3$)$_2$ (57.16) was on par with IAA at 45 DAS. Similarly, at 90 and 120 DAS the treatment GA and Ca(NO$_3$)$_2$ were on par with IAA. Least proline content was observed in control (G$_0$) at all the stages.

Salinity had a positive impact on proline accumulation and it increased from 39.99 µg g$^{-1}$ at S$_1$ to 71.61 µg g$^{-1}$ at S$_3$ at 45 DAS. Similar trend was observed at 90 and 120 DAS. The per cent increase in proline content was 53 per cent at 120 DAS and 35 per cent at 90 DAS from S$_1$ to S$_3$ salinity level.

The interaction effect was non-significant at all the stages. However, IAA at S$_3$ salinity level recorded higher proline (172.55) at 120 DAS. Least proline content was observed in control S$_1$ (34.20) at 45 DAS.

4.1.3 Nitrate reductase activity (NRA) (nanomoles nitrate g$^{-1}$ fresh weight)

Significant differences for nitrate reductase activity was observed among treatments and salinity levels at 45, 90 and 120 DAS. Whereas, interaction effect was found non-significant at all three stages (Table 7).

All the pre-soaking treatments had significantly higher nitrate reductase activity at 45 DAS. Whereas, at 90 DAS G$_1$ (IAA @200 ppm) and G$_2$ (GA @ 10 ppm) and at 120 DAS G$_1$, G$_2$ and G$_3$ (Ca(NO$_3$)$_2$ 3 per cent) differed significantly as compared to control. However, at 90 DAS, G$_1$ and G$_2$ were on par and at 120 DAS G$_1$, G$_2$ and G$_3$ (Na$_2$SO$_4$ 3 per cent) were on par with each other.

In general, NRA decreased with increase in salinity levels at all the growth stages. The reduction was to the extent of 16 per cent at 45 DAS, 33 per cent at 90 DAS and 29 per cent at 120 DAS respectively in S$_3$ salinity level as compared to control.

The reduction in nitrate reductase was minimum in pre-soaking treatments as compared to control at all the salinity levels and growth stages.

4.1.4 Calcium content in leaf (%)

Significances in the calcium content of leaf at 45, 90 and 120 DAS were observed among the soaking treatments and salinity levels. Whereas, interaction effect was found non-significant at all stages (Table 8).

At 45 DAS, leaves of IAA treatment had significantly higher calcium content (3.32), while, control had the least (2.98) as compared to other treatments which were on par with each other. Similarly, at 90 and 120 DAS, IAA pre-soaking treatment had significantly higher calcium content (3.59 and 3.84 respectively) and was on par with Ca(NO$_3$)$_2$ (3.51 and 3.75 respectively) and GA (3.46 and 3.68 respectively) as compared to control (3.05 and 3.26).

Calcium content of leaves decreased as the salinity level increased at 45 DAS, lower salinity level S$_1$ (3.54) had significantly higher calcium as compared to S$_3$ (2.94). Similarly, at 90 and 120 DAS the calcium content decreased from 3.96 to 2.81 and from 4.34 to 2.96 respectively with salinity gradients from S$_1$ to S$_3$.

Interaction effect was found to be non-significant at all the stages. However, IAA seed priming treatment had highest calcium content compared to other treatments at all salinity levels.

4.1.5 Sodium content in leaf (%)

Significant differences in the sodium content of leaf was observed among genotypes and salinity levels at all the stages, whereas, interaction effect was significant only at 45 and 120 DAS (Table 9).

Leaf sodium content was significantly higher in control (0.71) and was on par with Na$_2$SO$_4$ (0.65) and GA$_2$ (0.62) at 45 DAS, however, significantly least sodium was noticed in IAA soaked treatment (0.42). Similarly, at 90 and 120 DAS, control had significantly higher (0.36 and 0.26 respectively) sodium content as compared to all other treatments. However, at 90 and 120 DAS, IAA (0.24 and 0.17 respectively) maintained least sodium content but was on par with Ca(NO$_3$)$_2$ (0.25 and 0.18 respectively).

Increase in salinity increased the sodium content in leaf. The per cent increase in sodium content with increase in salinity was less at 90 and 120 DAS compared to 45 DAS. The salinity level S$_3$ (12.0 dS m$^{-1}$) had significantly higher leaf sodium content of 1.11, 0.46 and 0.28 per cent at 45, 90 and 120 DAS respectively as compared to S$_1$ salinity level (<2.0 dS m$^{-1}$) which had significantly lower leaf sodium content of 0.20, 0.17 and 0.12 per cent at 45, 90 and 120 DAS respectively. Steep increase in sodium content was observed at S$_3$.
Table 4. Effect of plant growth regulators and micronutrients on chlorophyll ‘b’ content in leaf (mg g\(^{-1}\) fresh weight) at different plant growth stages under different salinity levels in cotton

<table>
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<tr>
<th>Salinity/Seed Priming Treatments</th>
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<td>G(_2)-GA @ 10 ppm</td>
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<th>CD at 5%</th>
<th>S.Em±</th>
<th>CD at 5%</th>
<th>S.Em±</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.03</td>
<td>0.08</td>
<td>0.03</td>
<td>0.07</td>
<td>0.03</td>
<td>0.08</td>
</tr>
</tbody>
</table>

For Comparing Salinity (S)

<table>
<thead>
<tr>
<th>CD at 5%</th>
<th>CD at 5%</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.01</td>
<td>NS</td>
<td>0.01</td>
</tr>
</tbody>
</table>

For Comparing Interaction (G x S)

<table>
<thead>
<tr>
<th>NS</th>
<th>NS</th>
<th>NS</th>
</tr>
</thead>
</table>

S\(_1\) < 2.0 dS m\(^{-1}\)
S\(_2\) 6.0 dS m\(^{-1}\)
S\(_3\) 12.0 dS m\(^{-1}\)
Table 5. Effect of plant growth regulators and micronutrients on total chlorophyll content in leaf (mg g\(^{-1}\) fresh weight) at different plant growth stages under three salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S(_1)</td>
<td>S(_2)</td>
<td>S(_3)</td>
</tr>
<tr>
<td>G(_1)-IAA @ 200 ppm</td>
<td>1.08</td>
<td>0.94</td>
<td>0.82</td>
</tr>
<tr>
<td>G(_2)-GA @ 10 ppm</td>
<td>1.04</td>
<td>1.00</td>
<td>0.77</td>
</tr>
<tr>
<td>G(_3)-Na(_2)SO(_4) @ 3%</td>
<td>1.04</td>
<td>0.85</td>
<td>0.78</td>
</tr>
<tr>
<td>G(_4)-Ca(NO(_3))(_2) @ 3%</td>
<td>1.01</td>
<td>0.86</td>
<td>0.72</td>
</tr>
<tr>
<td>G(_5)-Control</td>
<td>0.89</td>
<td>0.75</td>
<td>0.60</td>
</tr>
<tr>
<td>Mean</td>
<td>1.01</td>
<td>0.88</td>
<td>0.74</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  
Salinity (S): S\(_1\) < 2.0 dS m\(^{-1}\), S\(_2\) = 6.0 dS m\(^{-1}\), S\(_3\) = 12.0 dS m\(^{-1}\)
LEGEND

Salinity levels (dS m\(^{-1}\))

S\(_1\) - < 2 dS m\(^{-1}\) (control)
S\(_2\) - 6 dS m\(^{-1}\)
S\(_3\) - 12 dS m\(^{-1}\)

Treatments

G\(_1\) – IAA @ 200 ppm
G\(_2\) – GA @ 10 ppm
G\(_3\) – Na\(_2\)SO\(_4\) 3 per cent
G\(_4\) – Ca(NO\(_3\))\(_2\) 3 per cent
G\(_5\) – Control
Fig. 2. Effect of PGR’s and micronutrients on chlorophyll ‘a’, ‘b’ and total chlorophyll Scontent in leaf (at 90 DAS) under three salinity levels in cotton.
salinity level i.e. 2.3 times more at 45 DAS, 1.8 times more at 90 DAS, and 1.25 times at 120 DAS as compared to S2.

4.1.6 Potassium content in leaf (%)

The data on potassium content in leaf observed at 45, 90, and 120 DAS are presented in Table 10. Treatments and salinity levels were found significant for potassium content in leaf at all the three stages except for pre-soaking treatments at 120 DAS but interaction effect was found to be significant at all stages. In general, potassium content was higher at 45 DAS as compared to 90 and 120 DAS.

At 45 DAS significantly higher potassium content was observed in IAA (2.98) as compared to all other treatments followed by Ca(NO$_3$)$_2$ (2.80) and GA (2.78) and Na$_2$SO$_4$ (2.74) which were on par with each other and significantly lower potassium content was observed in control (2.55). Similar trend was observed at 90 DAS whereas, at 120 DAS, numerically higher potassium content (1.40) was observed in IAA priming treatment followed by GA (1.31) and Ca(NO$_3$)$_2$ (1.24) and least was in control (1.15).

The salinity levels had a negative impact on potassium content of leaf at all the three stages. Significantly higher potassium content of 3.15, 2.28 and 1.38 per cent was observed at S1 salinity level at 45, 90 and 120 DAS respectively as compared to S2 and S3 salinity levels.

Interaction effect was found non-significant at all stages. Accumulation of potassium in leaves was significantly higher in soaking treatment, IAA (3.41) in S1 at 45 DAS, whereas, least accumulation was observed in control (0.97) in S3 salinity level at 120 DAS.

4.1.7 K/Na ratio in leaves

Significant differences among genotypes, salinity levels and interaction were observed for K/Na ratio in leaves at all the three stages and data are presented in Table 11.

Irrespective of salinity level, significantly higher K/Na ratio was observed in IAA (11.43) at all the stages as compared to other treatments followed by Ca(NO$_3$)$_2$ (9.18) which were on par with each other at 45 DAS. Significantly lower K/Na ratio was observed in control (5.93, 4.85 and 5.49) at 45, 90 and 120 DAS, respectively.

With increase in salinity level from S1 (<2.0 dS m$^{-1}$) to S3 (12.0 dS m$^{-1}$), K/Na decreased significantly from 16.66 to 2.28 at 45 DAS, from 13.93 to 3.52 and from 11.41 to 4.22 at 90 and 120 DAS respectively. In general, S1 level of salinity maintained significantly higher ratio whereas, S3 salinity level recorded lowest ratio at all the stages.

Interaction effect of G x S was significant with treatment IAA (22.73, 20.07 and 15.2) at 45, 90 and 120 DAS respectively as compared to other interaction and lowest ratio was found in control (1.52) in S3 salinity at 45 DAS. Similarly, at 90 and 120 DAS, control treatment maintained lower K/Na ratio of 2.22 and 2.69 respectively at S3 salinity level.

4.1.8 Na/Ca ratio in leaves

Significant differences were found among genotypes, salinity levels and interactions for Na/Ca ratio in leaves at all the three stages and data are presented in Table 12.

Significantly higher Na/Ca ratio was observed in control (0.25) at 45 DAS. Whereas, the treatment Na$_2$SO$_4$ (0.21) was on par with GA (0.20) and Ca(NO$_3$)$_2$ (0.19) treatment at 45 DAS. At 90 and 120 DAS the treatment control (0.13 and 0.09, respectively) had higher Na/Ca ratio. The treatment IAA (0.07) was on par with Ca(NO$_3$)$_2$ (0.08) at 90 DAS and no other treatment was found to be on par with control at 120 DAS which recorded significantly higher Na/Ca ratio (0.09). Significantly lower Na/Ca ratio was observed in IAA (0.13, 0.07 and 0.05) at all the stages.

Na/Ca increased from 0.06 to 0.38 with increase in salinity level from S1 (<2.0 dS m$^{-1}$) to S3 (12.0 dS m$^{-1}$) at 45 DAS. Similarly, Na/Ca ratio increased from 0.04 to 0.16 and from 0.03 to 0.10 at 90 and 120 DAS respectively with increase in salinity level from S1 to S3. S3 salinity level had significantly higher ratio of Na/Ca, whereas, S1 salinity level had lowest ratio at 45, 90 and 120 DAS.

Significant differences were found among salinity and treatment at all stages. The treatment control (0.50, 0.20 and 0.14) had higher Na/Ca ratio at S3 salinity level (12.0 dS m$^{-1}$) at 45, 90 and 120 DAS respectively. Least Na/Ca ratio was noticed in IAA (0.02) in S1 (<2.0 dS m$^{-1}$) salinity level at 120 DAS. Similarly, at 45 and 90 DAS also IAA maintained lower Na/Ca ratio (0.04 and 0.03 respectively) at S1 salinity level.

4.2 YIELD AND YIELD COMPONENTS

The data on yield and yield components are presented in Table 13, 14 and 15.

4.2.1 Seed cotton yield (g plant$^{-1}$)
Table 6. Effect of plant growth regulators and micronutrients on proline content in leaf (\text{g g}^{-1} \text{dry weight}) at different plant growth stages under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S&lt;sub&gt;1&lt;/sub&gt;</td>
<td>S&lt;sub&gt;2&lt;/sub&gt;</td>
<td>S&lt;sub&gt;3&lt;/sub&gt;</td>
</tr>
<tr>
<td>G&lt;sub&gt;1&lt;/sub&gt;-IAA @ 200 ppm</td>
<td>45.39</td>
<td>55.24</td>
<td>77.34</td>
</tr>
<tr>
<td>G&lt;sub&gt;2&lt;/sub&gt;-GA @ 10 ppm</td>
<td>40.39</td>
<td>52.26</td>
<td>71.39</td>
</tr>
<tr>
<td>G&lt;sub&gt;3&lt;/sub&gt;-Na&lt;sub&gt;2&lt;/sub&gt;SO&lt;sub&gt;4&lt;/sub&gt; @ 3%</td>
<td>37.34</td>
<td>48.69</td>
<td>68.58</td>
</tr>
<tr>
<td>G&lt;sub&gt;4&lt;/sub&gt;-Ca(NO&lt;sub&gt;3&lt;/sub&gt;)&lt;sub&gt;2&lt;/sub&gt; @ 3%</td>
<td>42.62</td>
<td>53.55</td>
<td>75.32</td>
</tr>
<tr>
<td>G&lt;sub&gt;5&lt;/sub&gt;-Control</td>
<td>34.20</td>
<td>44.40</td>
<td>65.39</td>
</tr>
<tr>
<td>Mean</td>
<td>39.99</td>
<td>50.83</td>
<td>71.61</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  
S<sub>1</sub>: S<sub>.Em±</sub> 0.95  CD at 5% 2.77  
S<sub>2</sub>: S<sub>.Em±</sub> 0.92  3.60  0.55  2.17  2.31  9.06  
S<sub>3</sub>: S<sub>.Em±</sub> 1.64  NS  3.43  NS  4.98  NS

S<sub>1</sub>: &lt; 2.0 dS m<sup>-1</sup>  
S<sub>2</sub>: 6.0 dS m<sup>-1</sup>  
S<sub>3</sub>: 12.0 dS m<sup>-1</sup>
LEGEND

Salinity levels (dS m\(^{-1}\))

\(S_1\) - < 2 dS m\(^{-1}\) (control)

\(S_2\) - 6 dS m\(^{-1}\)

\(S_3\) - 12 dS m\(^{-1}\)

Treatments

\(G_1\) – IAA @ 200 ppm

\(G_2\) – GA @ 10 ppm

\(G_3\) – Na\(_2\)SO\(_4\) 3 per cent

\(G_4\) – Ca(NO\(_3\))\(_2\) 3 per cent

\(G_5\) – Control
Fig. 3. Effect of PGR's and micronutrients on proline content (at 90 DAS) under three salinity levels in cotton.
Table 7. Effect of plant growth regulators and micronutrients on nitrate reductase activity in leaf (nanomoles nitrate g⁻¹ fresh weight) at different plant growth stages under three salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S₁</td>
<td>S₂</td>
<td>S₃</td>
</tr>
<tr>
<td>G₁-IAA @ 200 ppm</td>
<td>181.38</td>
<td>167.70</td>
<td>153.60</td>
</tr>
<tr>
<td>G₂-GA @ 10 ppm</td>
<td>175.76</td>
<td>163.67</td>
<td>149.14</td>
</tr>
<tr>
<td>G₃-Na₂SO₄ @ 3%</td>
<td>173.28</td>
<td>160.51</td>
<td>142.87</td>
</tr>
<tr>
<td>G₄-Ca(NO₃)₂ @ 3%</td>
<td>179.53</td>
<td>165.29</td>
<td>151.69</td>
</tr>
<tr>
<td>G₅-Control</td>
<td>167.16</td>
<td>150.18</td>
<td>139.47</td>
</tr>
<tr>
<td>Mean</td>
<td>175.42</td>
<td>161.47</td>
<td>147.35</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)         | S.Em± | CD at 5% | S.Em± | CD at 5% | S.Em± | CD at 5% |
| Salinity (S)                       | 3.13  | 9.12     | 3.05  | 8.92     | 2.15  | 6.26     |
| Interaction (G x S)                | 5.41  | NS       | 5.29  | NS       | 3.72  | NS       |

S₁: < 2.0 dS m⁻¹                         S₂: 6.0 dS m⁻¹                       S₃: 12.0 dS m⁻¹
LEGEND

Salinity levels (dS m\(^{-1}\))

\( S_1 \) - < 2 dS m\(^{-1}\) (control)
\( S_2 \) - 6 dS m\(^{-1}\)
\( S_3 \) - 12 dS m\(^{-1}\)

Treatments

\( G_1 \) – IAA @ 200 ppm
\( G_2 \) – GA @ 10 ppm
\( G_3 \) – Na\(_2\)SO\(_4\) 3 per cent
\( G_4 \) – Ca(NO\(_3\))\(_2\) 3 per cent
\( G_5 \) – Control
Fig. 4. Effect of PGR’s and micronutrients on nitrate reductase activity (at 90 DAS) under three salinity levels in cotton.
Table 8. Effect of plant growth regulators and micronutrients on calcium content in leaf (%) at different plant growth stages under three salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S&lt;sub&gt;1&lt;/sub&gt;</td>
<td>S&lt;sub&gt;2&lt;/sub&gt;</td>
<td>S&lt;sub&gt;3&lt;/sub&gt;</td>
</tr>
<tr>
<td>G&lt;sub&gt;1&lt;/sub&gt;-IAA @ 200 ppm</td>
<td>3.65</td>
<td>3.27</td>
<td>3.05</td>
</tr>
<tr>
<td>G&lt;sub&gt;2&lt;/sub&gt;-GA @ 10 ppm</td>
<td>3.58</td>
<td>3.24</td>
<td>3.01</td>
</tr>
<tr>
<td>G&lt;sub&gt;3&lt;/sub&gt;-Na&lt;sub&gt;2&lt;/sub&gt;SO&lt;sub&gt;4&lt;/sub&gt; @ 3%</td>
<td>3.49</td>
<td>3.18</td>
<td>3.00</td>
</tr>
<tr>
<td>G&lt;sub&gt;4&lt;/sub&gt;-Ca(NO&lt;sub&gt;3&lt;/sub&gt;)&lt;sub&gt;2&lt;/sub&gt; @ 3%</td>
<td>3.62</td>
<td>3.21</td>
<td>2.97</td>
</tr>
<tr>
<td>G&lt;sub&gt;5&lt;/sub&gt;-Control</td>
<td>3.34</td>
<td>2.92</td>
<td>2.68</td>
</tr>
<tr>
<td>Mean</td>
<td>3.54</td>
<td>3.16</td>
<td>2.91</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)
- S.E±: 0.06
- CD at 5%: 0.16

For Comparing Salinity (S)
- S.E±: 0.03
- CD at 5%: 0.14

For Comparing Interaction (G x S)
- S.E±: 0.10
- NS

S<sub>1</sub>: < 2.0 dSm<sup>-1</sup>
S<sub>2</sub>: 6.0 dSm<sup>-1</sup>
S<sub>3</sub>: 12.0 dSm<sup>-1</sup>
Table 9. Effect of plant growth regulators and micronutrients on sodium content in leaf (%) at different plant growth stages under three salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S1</td>
<td>S2</td>
<td>S3</td>
</tr>
<tr>
<td>G1-IAA @ 200 ppm</td>
<td>0.15</td>
<td>0.36</td>
<td>0.74</td>
</tr>
<tr>
<td>G2-GA @ 10 ppm</td>
<td>0.22</td>
<td>0.51</td>
<td>1.13</td>
</tr>
<tr>
<td>G3-Na2SO4 @ 3%</td>
<td>0.19</td>
<td>0.54</td>
<td>1.21</td>
</tr>
<tr>
<td>G4-Ca(NO3)2 @ 3%</td>
<td>0.17</td>
<td>0.44</td>
<td>1.12</td>
</tr>
<tr>
<td>G5-Control</td>
<td>0.26</td>
<td>0.53</td>
<td>1.35</td>
</tr>
<tr>
<td>Mean</td>
<td>0.20</td>
<td>0.48</td>
<td>1.11</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  
S.Em±  CD at 5%  
0.03  0.09  
0.02  0.07  
0.05  0.16  

For Comparing Salinity (S)  
S.Em±  CD at 5%  
0.012  0.036  
0.003  0.013  
0.022  NS  

For Comparing Interaction (G x S)  
S.Em±  CD at 5%  
0.005  0.014  
0.005  0.021  
0.008  0.024  

S1: < 2.0 dSm⁻¹  
S2: 6.0 dSm⁻¹  
S3: 12.0 dSm⁻¹
Table 10. Effect of plant growth regulators and micronutrients on potassium content in leaf (%) at different plant growth stages under three levels of salinity in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S1</td>
<td>S2</td>
<td>S3</td>
</tr>
<tr>
<td>G1-IAA @ 200 ppm</td>
<td>3.41</td>
<td>2.88</td>
<td>2.64</td>
</tr>
<tr>
<td>G2-GA @ 10 ppm</td>
<td>3.07</td>
<td>2.86</td>
<td>2.45</td>
</tr>
<tr>
<td>G3-Na2SO4 @ 3%</td>
<td>3.12</td>
<td>2.65</td>
<td>2.46</td>
</tr>
<tr>
<td>G4-Ca(NO3)2 @ 3%</td>
<td>3.23</td>
<td>2.83</td>
<td>2.34</td>
</tr>
<tr>
<td>G5-Control</td>
<td>2.91</td>
<td>2.69</td>
<td>2.05</td>
</tr>
<tr>
<td>Mean</td>
<td>3.15</td>
<td>2.79</td>
<td>2.39</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)

<table>
<thead>
<tr>
<th>Salinity (S)</th>
<th>CD at 5%</th>
<th>Interaction (G x S)</th>
<th>CD at 5%</th>
<th>Salinity (S)</th>
<th>CD at 5%</th>
<th>Interaction (G x S)</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>S1</td>
<td>&lt; 2.0 dSm⁻¹</td>
<td>S2: 6.0 dSm⁻¹</td>
<td>S3: 12.0 dSm⁻¹</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table 11. Effect of plant growth regulators and micronutrients on K/Na ratio in leaf at different plant growth stages under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S₁</td>
<td>S₂</td>
<td>S₃</td>
</tr>
<tr>
<td>G₁-IAA @ 200 ppm</td>
<td>22.73</td>
<td>8.00</td>
<td>3.57</td>
</tr>
<tr>
<td>G₂-GA @ 10 ppm</td>
<td>13.95</td>
<td>5.61</td>
<td>2.17</td>
</tr>
<tr>
<td>G₃-Na₂SO₄ @ 3%</td>
<td>16.42</td>
<td>4.91</td>
<td>2.03</td>
</tr>
<tr>
<td>G₄-Ca(NO₃)₂ @ 3%</td>
<td>19.00</td>
<td>6.44</td>
<td>2.09</td>
</tr>
<tr>
<td>G₅-Control</td>
<td>11.19</td>
<td>5.08</td>
<td>1.52</td>
</tr>
<tr>
<td>Mean</td>
<td>16.66</td>
<td>6.00</td>
<td>2.28</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)
- S.Em±: 0.40, 1.15, 0.23, 0.67, 0.12, 0.35
- CD at 5%: 0.08, 0.33, 0.11, 0.44, 0.09, 0.36

For Comparing Salinity (S)
- S.Em±: 0.68, 1.99, 0.40, 1.16, 0.21, 0.60
- CD at 5%: S₁ < 2.0 dSm⁻¹, S₂: 6.0 dSm⁻¹, S₃: 12.0 dSm⁻¹
LEGEND

Salinity levels (dS m\(^{-1}\))

- \(S_1\) - < 2 dS m\(^{-1}\) (control)
- \(S_2\) - 6 dS m\(^{-1}\)
- \(S_3\) - 12 dS m\(^{-1}\)

Treatments

- \(G_1\) – IAA @ 200 ppm
- \(G_2\) – GA @ 10 ppm
- \(G_3\) – \(\text{Na}_2\text{SO}_4\) 3 per cent
- \(G_4\) – \(\text{Ca(NO}_3\text{)}_2\) 3 per cent
- \(G_5\) – Control
Fig. 5 Effect of PGR’s and micronutrients on potassium, sodium and K/Na (at 90 DAS) content in leaf under three salinity levels in cotto.
In general, the seed cotton yield (Table 13) decreased significantly from 40.76 to 24.77 g plant\(^{-1}\) with increase in salinity from S\(_1\) (<2.0 dS m\(^{-1}\)) to S\(_3\) (12 dS m\(^{-1}\)) level. Among the treatments IAA recorded significantly higher yield of 36.53 g plant\(^{-1}\) as compared to other treatments followed by GA (33.32) and GA (32.41) which were on par with each other. The treatment control had the lowest seed cotton yield of 28.76 g plant\(^{-1}\) which was on par with Na\(_2\)SO\(_4\) (29.24).

Among the salinity levels, S\(_1\) had significantly highest yield of 40.76 g per plant and lowest yield (24.77) was observed at S\(_3\) salinity level. Treatment and salinity interaction was found non-significant.

### 4.2.2 Seed cotton yield (kg ha\(^{-1}\))

Significant differences among treatments and salinity levels were observed for seed cotton yield (kg ha\(^{-1}\)) and data are presented in Table 13.

The treatment IAA produced significantly higher seed cotton yield of 2029.3 kg ha\(^{-1}\) which was on par with GA (1850.9) treatment, whereas, treatment control had recorded significantly lower seed cotton yield as compared to all other treatments.

As the salinity level increased, the yield decreased drastically. S\(_1\) salinity level had significantly higher yield (2264.5 kg ha\(^{-1}\)) as compared to the highest salinity level of S\(_3\) (1376.1 kg ha\(^{-1}\)).

Interaction effect between salinity and treatments was found non-significant, however, the seeds pre-soaked with IAA produced numerically higher yield (2476.1) at S\(_1\) (<2.0 dS m\(^{-1}\)) salinity level and least was observed in control (1151.5) at S\(_3\) (12.0 dS m\(^{-1}\)) salinity level.

### 4.2.3 Total dry matter at harvest (g plant\(^{-1}\))

Treatments and salinity level were found significant with regard to total dry matter production at harvest (Table 13).

The treatment with pre-soaking of IAA @ 200 ppm had higher total dry matter (114.25 g plant\(^{-1}\)) followed by Ca(NO\(_3\))\(_2\) (112.71) and GA (110.38) which were on par with each other, whereas, control treatment produced significantly lower total dry matter (95.27) at harvest.

With increase in salinity level from S\(_1\) to S\(_3\) total dry matter decreased significantly. S\(_1\) salinity level recorded maximum dry weight of 126.48 g plant\(^{-1}\), whereas, S\(_3\) salinity level had significantly lower dry matter yield (83.67).

Non-significant interaction effect was observed. However, IAA (132.13) at S\(_1\) salinity recorded numerically maximum dry matter and least was observed in control (69.09) at S\(_3\) salinity level.

No interaction effect was found between treatments and salinity. However, IAA at S\(_1\) (<2.0 dS m\(^{-1}\)) recorded higher harvest index (33.7%) and least was observed in Ca(NO\(_3\))\(_2\) (23.8) at S\(_2\) salinity level.

### 4.2.4 Total number of good bolls per plant

Significant differences in total number of good bolls per plant was found for treatments and salinity levels with non-significant differences for interaction (Table 14).

The treatment with IAA recorded significant more number of good bolls (19.83) over other treatments but was on par with Ca(NO\(_3\))\(_2\) (18.97) and GA (18.16). The least number of good bolls were observed in control (15.30 bolls/plant).

Lower salinity of S\(_1\) (<2.0 dS m\(^{-1}\)) had higher good bolls/plant (21.59). With increase in salinity levels number of good bolls/plant decreased from S\(_1\) (21.59) to S\(_3\) salinity level (13.41). S\(_1\) salinity level had 61.0 per cent more bolls/plant compared to S\(_3\).

Interaction effect was found non-significant. However, the highest number of good bolls were observed in IAA pre-soaked treatment at S\(_1\) salinity (23.61).

### 4.2.5 Total number of bad bolls per plant

Treatments and salinity levels were found significant for total number of bad bolls per plant (Table 14).

Significantly higher number of bad bolls were observed in control (4.69) compared to all other pre-soaking treatments. The treatment IAA pre-soaking recorded least number of bad bolls per plant (3.11) followed by Ca(NO\(_3\))\(_2\) (3.24) which were on par each other.

With increase in salinity level the number of bad bolls per plant increased from S\(_1\) (2.27) salinity level to S\(_3\) (5.13) salinity level.

Interaction effect was found non-significant. However, significantly higher number of bad bolls per plant was observed in control (6.80) at S\(_3\) salinity level and least was found in IAA treatment (1.89) at S\(_1\) salinity level.
Table 12. Effect of plant growth regulators and micronutrients on Na/Ca ratio in leaf at different plant growth stages under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>45 DAS</th>
<th>90 DAS</th>
<th>120 DAS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S&lt;sub&gt;1&lt;/sub&gt;</td>
<td>S&lt;sub&gt;2&lt;/sub&gt;</td>
<td>S&lt;sub&gt;3&lt;/sub&gt;</td>
</tr>
<tr>
<td>G&lt;sub&gt;1&lt;/sub&gt;-IAA @ 200 ppm</td>
<td>0.04</td>
<td>0.11</td>
<td>0.24</td>
</tr>
<tr>
<td>G&lt;sub&gt;2&lt;/sub&gt;-GA @ 10 ppm</td>
<td>0.06</td>
<td>0.16</td>
<td>0.38</td>
</tr>
<tr>
<td>G&lt;sub&gt;3&lt;/sub&gt;-Na&lt;sub&gt;2&lt;/sub&gt;SO&lt;sub&gt;4&lt;/sub&gt; @ 3%</td>
<td>0.05</td>
<td>0.17</td>
<td>0.40</td>
</tr>
<tr>
<td>G&lt;sub&gt;4&lt;/sub&gt;-Ca(NO&lt;sub&gt;3&lt;/sub&gt;)&lt;sub&gt;2&lt;/sub&gt; @ 3%</td>
<td>0.05</td>
<td>0.14</td>
<td>0.38</td>
</tr>
<tr>
<td>G&lt;sub&gt;5&lt;/sub&gt;-Control</td>
<td>0.08</td>
<td>0.18</td>
<td>0.50</td>
</tr>
<tr>
<td>Mean</td>
<td>0.06</td>
<td>0.15</td>
<td>0.38</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  

<table>
<thead>
<tr>
<th></th>
<th>S.Em±</th>
<th>CD at 5%</th>
<th>S.Em±</th>
<th>CD at 5%</th>
<th>S.Em±</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>G&lt;sub&gt;1&lt;/sub&gt;-IAA @ 200 ppm</td>
<td>0.012</td>
<td>0.034</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>G&lt;sub&gt;2&lt;/sub&gt;-GA @ 10 ppm</td>
<td>0.005</td>
<td>0.020</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>G&lt;sub&gt;3&lt;/sub&gt;-Na&lt;sub&gt;2&lt;/sub&gt;SO&lt;sub&gt;4&lt;/sub&gt; @ 3%</td>
<td>0.02</td>
<td>0.06</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

For Comparing Salinity (S)  

<table>
<thead>
<tr>
<th></th>
<th>S.Em±</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>S&lt;sub&gt;1&lt;/sub&gt; &lt; 2.0 dSm&lt;sup&gt;-1&lt;/sup&gt;</td>
<td>0.007</td>
<td>0.022</td>
</tr>
<tr>
<td>S&lt;sub&gt;2&lt;/sub&gt; 6.0 dSm&lt;sup&gt;-1&lt;/sup&gt;</td>
<td>0.003</td>
<td>0.008</td>
</tr>
<tr>
<td>S&lt;sub&gt;3&lt;/sub&gt; 12.0 dSm&lt;sup&gt;-1&lt;/sup&gt;</td>
<td>0.003</td>
<td>0.008</td>
</tr>
</tbody>
</table>

For Comparing Interaction (G x S)  

<table>
<thead>
<tr>
<th></th>
<th>S.Em±</th>
<th>CD at 5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>G&lt;sub&gt;1&lt;/sub&gt;-IAA @ 200 ppm</td>
<td>0.007</td>
<td>0.022</td>
</tr>
<tr>
<td>G&lt;sub&gt;2&lt;/sub&gt;-GA @ 10 ppm</td>
<td>0.003</td>
<td>0.008</td>
</tr>
<tr>
<td>G&lt;sub&gt;3&lt;/sub&gt;-Na&lt;sub&gt;2&lt;/sub&gt;SO&lt;sub&gt;4&lt;/sub&gt; @ 3%</td>
<td>0.003</td>
<td>0.008</td>
</tr>
</tbody>
</table>

S<sub>1</sub>: < 2.0 dSm<sup>-1</sup>  
S<sub>2</sub>: 6.0 dSm<sup>-1</sup>  
S<sub>3</sub>: 12.0 dSm<sup>-1</sup>
LEGEND

Salinity levels (dS m\(^{-1}\))

\[ S_1 \leq 2 \text{ dS m}^{-1} \text{ (control)} \]

\[ S_2 \leq 6 \text{ dS m}^{-1} \]

\[ S_3 \leq 12 \text{ dS m}^{-1} \]

Treatments

\[ G_1 \text{ – IAA } @ \text{ 200 ppm} \]

\[ G_2 \text{ – GA } @ \text{ 10 ppm} \]

\[ G_3 \text{ – Na}_2\text{SO}_4 \text{ 3 per cent} \]

\[ G_4 \text{ – Ca(NO}_3)_2 \text{ 3 per cent} \]

\[ G_5 \text{ – Control} \]
Fig. 6. Effect of PGR’s and micronutrients on sodium, calcium and Na/Ca (at 90 DAS) content in leaf under three salinity levels in cotton.
Table 13. Effect of plant growth regulators and micronutrients on seed cotton yield and total dry matter (g plant\(^{-1}\)) under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>Seed cotton yield (g plant(^{-1}))</th>
<th>Seed cotton yield (kg ha(^{-1}))</th>
<th>Total dry matter (g plant(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S(_1)</td>
<td>S(_2)</td>
<td>S(_3)</td>
</tr>
<tr>
<td>G(_1)-IAA @ 200 ppm</td>
<td>44.57</td>
<td>36.52</td>
<td>28.50</td>
</tr>
<tr>
<td>G(_2)-GA @ 10 ppm</td>
<td>41.45</td>
<td>33.03</td>
<td>25.48</td>
</tr>
<tr>
<td>G(_3)-Na(_2)SO(_4) @ 3%</td>
<td>38.76</td>
<td>26.36</td>
<td>22.59</td>
</tr>
<tr>
<td>G(_4)-Ca(NO(_3))(_2) @ 3%</td>
<td>42.61</td>
<td>28.07</td>
<td>26.56</td>
</tr>
<tr>
<td>G(_5)-Control</td>
<td>36.43</td>
<td>29.12</td>
<td>20.73</td>
</tr>
<tr>
<td>Mean</td>
<td>40.76</td>
<td>30.62</td>
<td>24.77</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  
S.Em\(\pm\) CD at 5%  
0.90  5.43  
1.29  5.07  
1.55  2.61

Salinity (S)  
S.Em\(\pm\) CD at 5%  
154.94  10.91  
53.98  2.78  
114.21  NS

Interaction (G x S)  
S.Em\(\pm\) CD at 5%  
65.94  5.43  
53.98  2.78  
114.21  NS

S\(_1\):< 2.0 dS m\(^{-1}\)  
S\(_2\): 6.0 dS m\(^{-1}\)  
S\(_3\): 12.0 dS m\(^{-1}\)
LEGEND

Salinity levels (dS m\(^{-1}\))

- \(S_1\) - < 2 dS m\(^{-1}\) (control)
- \(S_2\) - 6 dS m\(^{-1}\)
- \(S_3\) - 12 dS m\(^{-1}\)

Treatments

- \(G_1\) – IAA @ 200 ppm
- \(G_2\) – GA @ 10 ppm
- \(G_3\) – \(\text{Na}_2\text{SO}_4\) 3 per cent
- \(G_4\) – \(\text{Ca(NO}_3)_2\) 3 per cent
- \(G_5\) – Control
Fig. 7. Effect of PGR's and micronutrients on seed cotton yield and total dry matter at harvest under three salinity levels in cotton.
Table 14. Effect of plant growth regulators and micronutrients on number of good bolls, bad bolls and total number of bolls per plant under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>Number of good bolls per plant</th>
<th>Number of bad bolls per plant</th>
<th>Total number of bolls per plant</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S₁</td>
<td>S₂</td>
<td>S₃</td>
</tr>
<tr>
<td>G₁-IAA @ 200 ppm</td>
<td>23.61</td>
<td>20.15</td>
<td>15.73</td>
</tr>
<tr>
<td>G₂-GA @ 10 ppm</td>
<td>22.48</td>
<td>18.54</td>
<td>13.47</td>
</tr>
<tr>
<td>G₃-Na₂SO₄ @ 3%</td>
<td>20.40</td>
<td>17.57</td>
<td>12.33</td>
</tr>
<tr>
<td>G₄-Ca(NO₃)₂ @ 3%</td>
<td>23.13</td>
<td>19.41</td>
<td>14.36</td>
</tr>
<tr>
<td>G₅-Control</td>
<td>18.33</td>
<td>16.41</td>
<td>11.17</td>
</tr>
<tr>
<td>Mean</td>
<td>21.59</td>
<td>18.42</td>
<td>13.41</td>
</tr>
<tr>
<td>For Comparing Treatments (G)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S.Em±</td>
<td>0.71</td>
<td>2.06</td>
<td></td>
</tr>
<tr>
<td>CD at 5%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>For Comparing Salinity (S)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S.Em±</td>
<td>0.38</td>
<td>1.51</td>
<td></td>
</tr>
<tr>
<td>CD at 5%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>For Comparing Interaction (G x S)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S.Em±</td>
<td>1.22</td>
<td>NS</td>
<td></td>
</tr>
<tr>
<td>CD at 5%</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

S₁: < 2.0 dS m⁻¹  
S₂: 6.0 dS m⁻¹  
S₃: 12.0 dS m⁻¹
4.2.6 Total number of bolls per plant
The observation on total number of bolls indicated significant differences among salinity levels and treatments and no interaction effect was found (Table 14).

The treatment IAA (22.94) recorded significantly higher number of total bolls over other control but it was on par with Ca(NO$_3$)$_2$ (22.20) and GA (21.66) treatments. Least number of bolls was observed in control (20.00) and was on par with Na$_2$SO$_4$ (20.62).

Salinity levels influenced the total number of bolls per plant. At $S_1$ (<2.0 dS m$^{-1}$) salinity level, higher number of bolls/plant (23.86) was observed. With increase in salinity, the total number of bolls/plant decreased from 23.86 to 18.54 at $S_1$ and $S_3$ salinity levels respectively.

Interaction effect was found to be non-significant, however, total number of bolls were highest at $S_1$ salinity level in IAA (25.50) treatment.

4.2.7 Mean boll weight (g boll$^{-1}$)
The data showed significant differences between salinity level whereas, treatment and interaction effects showed non-significance (Table 15).

Higher mean boll weight was observed in IAA (2.15) followed by Ca(NO$_3$)$_2$ (2.13) and GA (2.06) as compared to control (1.99).

With increase in salinity levels, there was decrease in boll weight and $S_3$ salinity level had lowest boll weight of 1.73 g boll$^{-1}$ and significantly with $S_1$ (2.25) and $S_2$ (2.27) salinity levels.

4.2.8 Harvest index
Significant differences in harvest index was observed for treatments, salinity levels and interactions (Table 15). Among the treatments, IAA had higher harvest index (31.8%) and was significantly different from all other treatments.

In general, the harvest index was high at lower salinity level of $S_1$ (32.2%) whereas, $S_2$ salinity level had significantly lower harvest index.
Table 15. Effect of plant growth regulators and micronutrients on mean boll weight (g boll\(^{-1}\)) and harvest index under different salinity levels in cotton

<table>
<thead>
<tr>
<th>Salinity/Seed Priming Treatments</th>
<th>Mean boll weight (g boll(^{-1}))</th>
<th>Harvest index (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S(_1)</td>
<td>S(_2)</td>
</tr>
<tr>
<td>G(_1)-IAA @ 200 ppm</td>
<td>2.32</td>
<td>2.25</td>
</tr>
<tr>
<td>G(_2)-GA @ 10 ppm</td>
<td>2.27</td>
<td>2.16</td>
</tr>
<tr>
<td>G(_3)-Na(_2)SO(_4) @ 3%</td>
<td>2.19</td>
<td>2.14</td>
</tr>
<tr>
<td>G(_4)-Ca(NO(_3))_2 @ 3%</td>
<td>2.34</td>
<td>2.21</td>
</tr>
<tr>
<td>G(_5)-Control</td>
<td>2.15</td>
<td>2.27</td>
</tr>
<tr>
<td>Mean</td>
<td>2.25</td>
<td>2.27</td>
</tr>
</tbody>
</table>

For Comparing Treatments (G)  
S.Em±  CD at 5%  

|  | 0.06  | NS    | 0.55  | 1.59 |
|  | 0.06  | 0.23  | 0.46  | 1.80 |
|  | 0.10  | NS    | 0.94  | 2.75 |

S\(_1\): < 2.0 dS m\(^{-1}\)  
S\(_2\): 6.0 dS m\(^{-1}\)  
S\(_3\): 12.0 dS m\(^{-1}\)
V. DISCUSSION

Salinity is one of the world’s most environmental problems in agriculture. Salinity stress affects many metabolic aspects of plants and induces anatomical and morphological changes resulting in reduced growth. This reduction in growth may result on dry matter allocation, ion relations, water status, biochemical reactions or a combination of many physiological factors.

The use of plant growth regulators and nutrients is an alternative approach to ameliorate the effect of salinity in crops. Though cotton being recognized as the most salt tolerant of all crops, but has not received much attention in this regard. The present investigation deals with the effect of plant growth regulators and micronutrients under varying levels of salinity and their relative performance to physiological, biochemical parameters and the cause for differences in yield and attributes. The results obtained on these aspects in the present study are discussed in this chapter.

A field experiment was carried out during the year 2005-06 to find out ameliorative effects of plant growth regulators and micronutrients on different physiological, biochemical and yield and yield attributes in cotton.

5.1 PHYSIOLOGICAL AND BIOCHEMICAL PARAMETERS

5.1.1 Chlorophyll content

Chlorophyll is known to influence the photosynthetic rate and inturn influence growth and development of cotton (Krasichkova et al., 1989). However, under saline conditions there will be degradation in pigment composition, which induce decrease in chlorophyll content. Salinity stress may also lead to destruction of fine structure of chloroplast and instability of pigment protein complex (Lapina and Papov, 1970).

Chlorophyll ‘a’ and chlorophyll ‘b’ and total chlorophyll content of leaf decreased with increase in salinity at all stages, whereas, it increased from 45 DAS to 120 DAS. Decrease in chlorophyll content has been reported by several authors. Garg and Garg (1985) in greengram and Prakash and Prathapasenan (1989) in rice. Ashraf and Rasul (1988) attributed the reduction in chlorophyll content under salinity stress to suppression of enzymes responsible for synthesis of chlorophyll under higher salt concentrations. Increased chlorophyllase activity and interference of salt ions with de novo synthesis of structural component proteins of chloroplast are also known to influence the chlorophyll content under salinity stress (Rao and Rao, 1981; Reddy and Vora, 1986; Sudhakar et al., 1991).

In the present investigation, seed priming with IAA had maintained significantly higher total chlorophyll contents and its fractions ‘a’ and ‘b’ at all stages followed by Ca(NO$_3$)$_2$ and GA treatments compared to control which recorded highest reduction. Similar increase in chlorophyll content has been reported by several authors, Saha and Gupta (1998) in mungbean, Ramanujulu et al. (1993) in Mulbarry, Afria et al. (1998) in Gaur, El-Tayeb (2005) in barley and Cengiz et al. (2002) in tomato. The increase in chlorophyll content in these treatments was attributed to decrease in chlorophyllase activity and de novo synthesis of structural component of proteins which are responsible for chlorophyll degradation (Subater and Rodriguez, 1978).

5.1.2 Free proline content

The physiological significance of proline accumulation is poorly understood. Proline has been assigned the role of cytosolue, a storage compounds or a protective agent for cytoplasmic enzymes and cellular structure (Demir, 2000; Pandey and Ganapathy, 1985).

Hanson and Hitz (1982) suggested that proline accumulation is a consequence of stress induced damage to cells. In plants, the role of proline may not be restricted to that of a compatible osmolyte. For example, proline synthesized during water deficit and salt-stress may serve as an organic nitrogen reserve that can be utilized during recovery (Trotel et al., 1989).

In the present study, the proline content in leaves of RAHS-14 cotton genotype increased with increase in salinity levels and seed priming treatments differed significantly in their ability to accumulate free proline under salinity stress. Seed priming treatments, IAA and CO(NO$_3$)$_2$ had more proline accumulation at all stages especially at higher salinity levels compared to Na$_2$SO$_4$ and control treatments, which accumulated less proline at all the three stages. More pronounced effect of salinity on proline accumulation was observed at 120 DAS than at 45 and 90 DAS particularly at higher salinity levels. Similar results were found by Badr and Albassam (2001) in pearl millet, Anuradha and Seeta Ram Rao (2002) in rice, Nandini and Subhendu (2002) in mungbean and Filitz et al. (2004) in rice.
The increased accumulation of proline in seed priming treatments with PGRS (IAA and GA) under salinity stress is attributed to effects of NaCl on the action of pyrroline-5-carboxylate reductase enzyme (Huber, 1974), slower utilization of proline for protein synthesis (Steward, 1972) and stimulation of glutamate conversion to proline during stress are also responsible for the accumulation of proline under salt stress. Higher proline accumulation in seed treatment with IAA and GA with higher capacities for excluding sodium from the shoots has suggested direct relationship between proline accumulation and yield in tomato (Alfocea et al., 1993). The increase in accumulation of free proline plays an important in imparting certain degree of salt tolerance through osmoregulation (Deleuney and Verma, 1991 and Samanas et al., 1995).

5.1.3 Nitrate reductase activity (NRA)

It is suggested that the key enzymes controlling metabolic pathways may limit the crop growth and thereby limit the yields (Priti et al., 2005). The main function of the enzyme nitrate reductase is to reduce nitrate to nitrite thus has role in the nitrogen metabolism and the activity of nitrate reductase enzyme is dependent on several variables in the growing conditions such as light, nitrogen, water supply and plant age (Garg et al., 2005).

The present findings revealed that NRA at higher salinity levels (12.0 dS m\(^{-1}\)) decreased as compared to lower salinity level (<2.0 dS m\(^{-1}\)) at all stages. The NRA was maximum at early stages (45 DAS) and reduced at peak flowering stage (120 DAS). Among the pre-soaking treatments, IAA and Ca(NO\(_3\))\(_2\) treatment showed higher NRA activity in <2.0 dS m\(^{-1}\) salinity level at 45 DAS. In control and Na\(_2\)SO\(_4\) treatments the activity was less due to salinity (Plaut, 1974) with a concurrent decrease in protein content to a greater extent (Klyshev et al., 1974).

The stimulated NR activity in seed priming treatments compared to control plants might be due to enhanced nitrogen uptake by plants (Muthuchelian et al., 1994). The positive effect of IAA and Ca(NO\(_3\))\(_2\) on NRA was due to its possible role in the activation of the inactive nitrate reductase protein and prevention of enzyme degradation by proteolysis. This might also be involved in the enhancement of enzyme synthesis or its maintenance in the active form and thus, has a protective role on nitrate reductase activity (Richard and Stanley, 1981).

5.1.4 Ionic content

5.1.4.1 Calcium

Calcium is an essential element for maintenance of selectivity and integrity of cell membrane (Fageria, 1985) and to delay the process of senescence and abscission (Pooviah and Lespald, 1975). It is also known to maintain or increase the rate of cell multiplication in salt stressed cotton roots (Kurth et al., 1986). Another role of calcium in plants is that, it acts as a second messenger in the transactions of extracellular signals (Owne, 1988).

Addition of sodium seems to have a very significant effect in depressing the calcium content of plants (Janardhan et al., 1979). In the present study, calcium content decreased with increasing salinity levels of external medium, particularly at higher salinity levels. Similar to K, the decline in Ca concentration in tissues of salt treated plant may result from an antagonistic effect of Na and/or of Mg on Ca uptake (He and Crammer, 1993).

Calcium content in leaf differed significantly among salinity levels and treatments. Mean values indicated that the seed priming treatment viz., IAA, Ca(NO\(_3\))\(_2\) and GA had higher Ca content in leaves, whereas, control and Na\(_2\)SO\(_4\) had lower Ca content at all three stages. With increase in salinity the calcium uptake was decreased at all the stages. However, calcium content was high in IAA and Ca(NO\(_3\))\(_2\) treatments in < 2 dS m\(^{-1}\) salinity level at 120 DAS. Comparatively higher Ca content observed in leaves of IAA and Ca(NO\(_3\))\(_2\) treatments, which might have inhibited K uptake resulting in lower K/Na ratio (Janardhan et al., 1976a). It has been reported that, a number of divalent cations including calcium affects potassium uptake promoting at some concentrations and inhibiting at other concentrations (Malibiari, 1993). The Ca content in leaf in all treatments increased compared to control under all salinity levels. Similar results were found by Saha and Gupta (1998), Sayed and Gadallah (2002), Uma Singh et al. (2004), Garg et al. (2005) and El-Tayeb (2005). An increase in cytosolic Ca\(^{2+}\), as second messenger, might induce further physiological responses including the expression of osmotic response genes for salt adaptation in plants (Pardo et al., 1998).

5.1.4.2 Sodium

Sodium exerts its ill effects on growth by its direct involvement on metabolism and nutritional imbalance. Differential content of sodium in plant parts among the genotypes indicates selectivity in different plant parts to accumulate sodium under salinity stress.
(Greenway, 1965b). In the present study, Na⁺ content in leaves increased with increase in salinity level which agrees with the findings of other workers (Banuls, 1991; Sayed and Gadallah, 2002; Ashraf et al., 2002; Cengiz and David, 2002; Cengiz et al., 2003; El-Tayeb, 2005; Anjum et al., 2005 and Ndayiragijic and Lutts, 2006). With an advance in the age of plant Na⁺ content of leaves decreased which might be attributed to decreased ion uptake or could be due to ion loss from plants (Greenway, 1965a).

Sodium content in plant parts changed significantly with salinity levels and a sharp increase was observed between < 2.0 dS m⁻¹ and 12.0 dS m⁻¹ salinity levels at 45 DAS. The higher mean values for sodium content in leaves were observed in control followed by Na₂SO₄ at all salinity levels at 45 DAS. The percent accumulation of sodium in leaves was more in control at 45 DAS, with increase in salinity levels from S₁ (<2.0 dS m⁻¹) to S₃ (12.0 dS m⁻¹). Similar observations were also found at 90 and 120 DAS. However, the accumulation of sodium was less in seed priming treatments with IAA and Ca(NO₃)₂ with increase in salinity levels at all stages. These findings are also in agreement with those of Banuls (1991), Cengiz and David (2002), Cengiz et al. (2003), Anjum et al. (2005), Sylvic (2005) and Ndayiragijic and Lutts (2006). This clearly indicates that pre-treatment with plant growth regulators and micronutrients induced reduction in Na absorption and toxicity, which is further reflected in low membrane injury, high water content and more dry matter production (Zhang et al., 1999 and Indira and Ramanujam, 1985).

5.1.4.3 Potassium

Potassium is an important nutritional requirement of crop along with N, P and Ca required for chlorophyll development, maintenance of water content in leaves and maintains C:N ratio in plants (Lefeuvre et al., 2001). Potassium is essential in activating some enzymes involved in the synthesis of certain peptide bonds and carbohydrate metabolism. It is known to maintain integrity of cell membrane (Lutts et al., 2004) and in needed for protection of plant against excessive concentration of sodium (Ghoulam et al., 2001). Under a conditions an increase in K resulted in better utilization of applied nitrogen (Harper and Balke, 1981). Improvement of K uptake under saline conditions may then serve as a mechanism of salinity tolerance by better utilization of nitrogen.

In general, K content decreased with increased salinity in leaves and significant differences were found between treatments at all three stages. The general decrease in K content was accompanied by corresponding increase in Na content showing apparent antagonism between sodium and potassium. The present findings are in conformity with those of Banuls (1991), Cengiz and David (2002), Sayed and Gadallah (2002), Cengiz et al. (2003), Anjum et al. (2005) Sylvic (2005), El-Tayeb (2005) and Ndayiragijic and Lutts (2006).

Though the inhibition of potassium uptake with salinity was common to all the treatments, but treatments differed significantly in their ability and maintain sufficient concentration of potassium under salt stress. The pre-soaking treatments of IAA, Ca(NO₃)₂ and GA registered lesser decrease in potassium content whereas Na₂SO₄ recorded moderate reduction while, control recorded higher reduction in potassium content. The treatments IAA, Ca(NO₃)₂ and GA were having higher potassium content at higher salinity levels indicating that these treatments have better ability to maintain K content and uptake. These results are in accordance with Nandini and Subhendu (2002), Saha and Gupta (1998) and Garg et al. (2005) who reported efficient Na and Cl regulation and high K absorption capacity and maintenance of stomatal conductance and transpiration rate under saline conditions. On the other hand, the treatment control and Na₂SO₄, which had higher sodium at high salinity level maintained low potassium levels indicating very low K⁺ versus Na⁺ selectivity. Thus, the concentration of essential elements like K may play a role in salt tolerance of cotton (Rathert, 1983) and capacity to maintain sufficient K content may impart a degree of salt tolerance (He and Crammer, 1993).

5.1.4.4 K/Na

As the salinity increases, the plant loses its ability to keep out sodium and maintain favourable concentrations of other ions particularly potassium, resulting in lower K/Na ratios. The main barrier to passive Na⁺ flow into shoots and leaves of plasma membrane of suberized root endodermis cell. It is observed that, with an increase in sodium concentration in rooting medium, leaf sodium content increases. This may be due to break down of root membrane integrity under salinity stress (Bohnert et al., 1995 and Hamida and El-Komy, 1998).

Susceptibility to loss of ionic control in membrane may be a physiological feature of salt sensitivity. However, regulation of Na transport can be achieved by an efficient K-Na exchange at plasma lemma and may help plant to survive salinity stress (Jeschke, 1984).
has been shown that an efficient salt exclusion by roots is correlated with high level of K/Na selectivity ratio in Chenopodium (Reimann, 1991) and also many authors have suggested to use relatively higher K/Na ratio as a criterion for evaluating salt tolerance of plants (Hung and Manns, 1980; Greenway and Manns, 1980).

In the present investigation, K/Na ratio computed in leaves differed significantly among seed priming treatments and increased with age due to relatively high K⁺ and low Na⁺ accumulation. The K/Na ratios in leaves decreased from 9.72 to 1.34, 8.51 to 2.06 and from 6.72 to 2.46 at 45, 90 and 120 DAS from S₁ (<2.0 dS m⁻¹) salinity level to S₃ (12.0 dS m⁻¹) salinity level, respectively. The seed priming treatment IAA maintained maximum K/Na ratio followed by Ca(NO₃)₂ and GA. Similar observations were made by several authors Ashraf et al. (2002) Cengiz and David (2002), Cengiz et al. (2002), Sayed and Godallah (2002), Cengiz et al. (2003) and Anjum et al. (2005). The maintenance of high K/Na ratio has been reported to be associated with higher salt tolerance of crops (Greenway and Manns, 1980).

Since the treatment control had low K⁺ and high Na⁺ in leaves the K/Na ratio was less at all salinity levels. The decrease in K/Na ratio with an increase in soil salinity was mainly due to increased uptake of Na and decreased uptake of K⁺ indicating thereby mutual antagonism between those two cations (Balaguru and Khanra, 1982 and Bohra and Deorffling, 1993).

5.1.4.5 Na/Ca ratio

In general, salinity caused increased sodium content and discrimination of calcium resulting in the increased Na/Ca ratios in plants. In the present study also Na/Ca ratios in the leaves increased due to salinity. The Na/Ca ratio was more in control and Na₂SO₄ treatments at all three salinity levels with increase in the age of plant. Such high Na/Ca ion ratio disturbs ionic as well as nutritional balance and this might lead to poor growth and yield under salinity (Gill, 1992). The Na/Ca ratio was reduced in all seed priming treatments at all salinity levels. The treatments IAA and Ca(NO₃)₂ had lower Na/Ca compared to other treatments at all salinity levels and at all stages. The maintenance of lower Na/Ca ratios in other treatments is attributed to the increased or higher Ca²⁺ contents even under saline conditions. Thus calcium could alleviate the NaCl induced inhibition of plant growth via the maintenance of net K⁺ to Na⁺ selectivity (Anjum et al., 2005).

5.2 SEED COTTON YIELD AND ITS COMPONENTS

The data on seed cotton yield and its components indicated significant treatment differences at all the salinity levels. A linear reduction in yield with increase in salinity levels was noticed. Reduction of seed cotton yield due to salinity have been reported by El-Saidi (1973), Ahmad et al. (1991), Munk and Roberts (1995), Vulkan et al. (1998) and Phogat et al. (2001).

In the present study, among five seed priming treatments, IAA recorded maximum yield followed by Ca(NO₃)₂ and GA treatments and differed significantly with other treatments and least yield was recorded in control.

Similar to seed cotton yield, yield components also showed significant variations among different treatments. Among the treatments, IAA (19.83) and Ca(NO₃)₂ (18.97) recorded higher number of bolls per plant compared to control (15.30) irrespective of salinity levels. These results in accordance with several workers, Puntamkar et al. (1970), Darra and Saxena (1971), Singh and Darra (1971), Afria et al. (1998), Ashraf et al. (2002), Cengiz and David (2002) and Cenizy et al. (2003).

The beneficial effects of plant growth regulators and micronutrients may be attributed to an increased root activity and plasticity of cell wall to provide greater absorption of water and nutrients and pre-sowing treatments fulfills the requirement of optimum amounts of hormones normally required by the plants for better growth (Burman et al., 2002; Lutts et al., 2004).

The treatments, IAA (2.15) and Ca(NO₃)₂ (2.13) recorded maximum single boll weight as well as least per cent reduction in boll weight (19% and 21%, respectively). Similarly, IAA and Ca(NO₃)₂ recorded lesser number of bad bolls per plant (3.11 and 3.26 respectively) and control (4.69) recorded higher number of bad bolls per plant. Similar results have been quoted by Latif and Khan (1976), Janardhan et al. (1979), Sharma et al. (1991) and Subbaiah et al. (1995).

5.2.1 Total dry matter

Total dry matter at harvest differed significantly among treatments and salinity levels. The treatment IAA maintained significantly higher total dry matter at all salinity levels. Moderate total dry matter was observed in Ca(NO₃)₂ and GA, where as least total dry matter...
Table 16. Correlation coefficients between seed cotton yield, total dry matter at harvest and biochemical characters at various stages in cotton

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Parameters</th>
<th>1</th>
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<td>0.466*</td>
<td>0.750*</td>
<td>0.907*</td>
<td>0.795*</td>
<td>0.855*</td>
<td>0.930*</td>
<td>0.731*</td>
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<td>0.762*</td>
<td>0.860*</td>
<td>0.840*</td>
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<td>0.828*</td>
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<td>0.990*</td>
<td>0.889*</td>
<td>0.817*</td>
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<td>0.896*</td>
<td>0.943*</td>
<td>0.847*</td>
<td>0.903*</td>
<td>0.870*</td>
<td>0.795*</td>
<td>0.774*</td>
<td>0.816*</td>
<td>0.777*</td>
<td>0.765*</td>
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<td>0.844*</td>
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<td>Chlorophyll 'b' content at 120 DAS</td>
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<td>0.980*</td>
<td>0.975*</td>
<td>0.919*</td>
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<td>0.931*</td>
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<td>Total chlorophyll content at 45 DAS</td>
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<td>0.908*</td>
<td>0.821*</td>
<td>0.910*</td>
<td>0.870*</td>
<td>0.766*</td>
<td>0.753*</td>
<td>0.795*</td>
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<td>Total chlorophyll content at 90 DAS</td>
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<td>0.900*</td>
<td>0.920*</td>
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<td>9</td>
<td>Total chlorophyll content at 120 DAS</td>
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<td>0.878*</td>
<td>0.892*</td>
<td>0.903*</td>
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<td>0.912*</td>
<td>0.859*</td>
<td>0.773*</td>
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<td>10</td>
<td>NR activity at 45 DAS</td>
<td>1.00</td>
<td>0.930*</td>
<td>0.968*</td>
<td>0.990*</td>
<td>0.982*</td>
<td>0.997*</td>
<td>0.977*</td>
<td>0.883*</td>
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<tr>
<td>11</td>
<td>NR activity at 90 DAS</td>
<td>1.00</td>
<td>0.988*</td>
<td>0.920*</td>
<td>0.930*</td>
<td>0.933*</td>
<td>0.937*</td>
<td>0.950*</td>
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<td>12</td>
<td>NR activity at 120 DAS</td>
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<td>0.967*</td>
<td>0.974*</td>
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<td>13</td>
<td>Proline content at 45 DAS</td>
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<td>0.997*</td>
<td>0.992*</td>
<td>0.976*</td>
<td>0.976*</td>
<td>0.918*</td>
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<td>14</td>
<td>Proline content at 90 DAS</td>
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<td>0.985*</td>
<td>0.935*</td>
<td>0.897*</td>
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<td>15</td>
<td>Proline content at 120 DAS</td>
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<td>0.995*</td>
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<tr>
<td>16</td>
<td>TDM at harvest</td>
<td>1.00</td>
<td>0.905*</td>
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<tr>
<td>17</td>
<td>Seed cotton yield</td>
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<td>0.905*</td>
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* Significant at 0.05 level
Table 17. Correlation coefficients between seed cotton yield, total dry matter at harvest and ionic content at various stages in cotton

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<td>1</td>
<td>Calcium content at 45 DAS</td>
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<td>0.921*</td>
<td>0.991*</td>
<td>0.746*</td>
<td>0.770*</td>
<td>0.927*</td>
<td>0.776*</td>
<td>-0.869*</td>
<td>-0.924*</td>
<td>-0.877*</td>
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<td>0.773*</td>
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<td>0.953*</td>
<td>0.964*</td>
<td>0.805*</td>
<td>0.852*</td>
<td>0.967*</td>
<td>0.874*</td>
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<td>-0.979*</td>
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<td>0.943*</td>
<td>0.843*</td>
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<td>Calcium content at 120 DAS</td>
<td>1.00</td>
<td>-0.866*</td>
<td>-0.984*</td>
<td>-0.954*</td>
<td>0.943*</td>
<td>0.907*</td>
<td>0.868*</td>
<td>0.855*</td>
<td>0.952*</td>
<td>0.930*</td>
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<td>-0.990*</td>
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<td>Sodium content at 45 DAS</td>
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<td>0.832*</td>
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<td>Sodium content at 90 DAS</td>
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<td>-0.843*</td>
<td>-0.854*</td>
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<td>Sodium content at 120 DAS</td>
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<td>-0.911*</td>
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<td>0.976*</td>
<td>-0.932*</td>
<td>-0.848*</td>
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<tr>
<td>7</td>
<td>Potassium content at 45 DAS</td>
<td>1.00</td>
<td>0.933*</td>
<td>0.896*</td>
<td>0.926*</td>
<td>0.990*</td>
<td>0.918*</td>
<td>-0.991*</td>
<td>-0.961*</td>
<td>-0.881*</td>
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<td>0.913*</td>
<td>0.843*</td>
<td>0.773*</td>
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<td>8</td>
<td>Potassium content at 90 DAS</td>
<td>1.00</td>
<td>0.760*</td>
<td>0.771*</td>
<td>0.926*</td>
<td>0.752*</td>
<td>-0.879*</td>
<td>-0.900*</td>
<td>-0.825*</td>
<td>0.843*</td>
<td>0.773*</td>
<td>0.908*</td>
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<tr>
<td>9</td>
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<td>0.785*</td>
<td>0.846*</td>
<td>0.896*</td>
<td>-0.899*</td>
<td>-0.853*</td>
<td>-0.797*</td>
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<tr>
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<td>K/Na ratio at 45 DAS</td>
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<td>11</td>
<td>K/Na ratio at 90 DAS</td>
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<td>0.926*</td>
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<td>-0.879*</td>
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<td>12</td>
<td>K/Na ratio at 120 DAS</td>
<td>1.00</td>
<td>-0.950*</td>
<td>-0.954*</td>
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<td>0.958*</td>
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<tr>
<td>14</td>
<td>Na/Ca ratio at 90 DAS</td>
<td>1.00</td>
<td>-0.983*</td>
<td>-0.857*</td>
<td>0.905*</td>
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<tr>
<td>15</td>
<td>Na/Ca ratio at 120 DAS</td>
<td>1.00</td>
<td>-0.983*</td>
<td>-0.857*</td>
<td>0.905*</td>
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<td>TDM at harvest</td>
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<tr>
<td>17</td>
<td>Seed cotton yield</td>
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* Significant at 0.05 level
Table 18. Economics of cotton as influenced by PGR's and nutrients under different salinity levels

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Total cost of cultivation (Rs./ha)</th>
<th>Gross returns (Rs./ha)</th>
<th>Net returns (Rs./ha)</th>
<th>B:C ratio</th>
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<tr>
<td>S₁G₁</td>
<td>13,046</td>
<td>49,526</td>
<td>36,480</td>
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<td>S₁G₂</td>
<td>12,966</td>
<td>46,057</td>
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<td>S₁G₃</td>
<td>12,964</td>
<td>43,069</td>
<td>30,105</td>
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<tr>
<td>S₁G₄</td>
<td>12,976</td>
<td>47,343</td>
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<td>S₁G₅</td>
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<td>27,533</td>
<td>2.13</td>
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<tr>
<td>S₂G₁</td>
<td>13,046</td>
<td>40,579</td>
<td>27,533</td>
<td>2.11</td>
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<td>S₂G₂</td>
<td>12,966</td>
<td>36,698</td>
<td>23,732</td>
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<td>29,286</td>
<td>16,322</td>
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<td>28,308</td>
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<td>S₃G₄</td>
<td>12,976</td>
<td>28,512</td>
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<td>S₃G₅</td>
<td>12,946</td>
<td>23,031</td>
<td>10,085</td>
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</table>

S₁r < 2 dS m⁻¹ (control), S₂r 6 dS m⁻¹, S₃ - 12 dS m⁻¹
G₁ – IAA 200 ppm, G₂ – GA 10 ppm, G₃ – Na₂SO₄ 3 per cent, G₄ – Ca(NO₃)₂ 3%,
G₅ – Control
Cost :
IAA – Rs. 250/g
GA – Rs. 100/g
Na₂SO₄ – Rs. 150/500 g
Ca(NO₃)₂ – Rs. 250/500 g
Seed cotton yield – Rs. 2000/q
was observed in control. The treatment control had (42%) the highest reduction of dry matter reduction from $S_1$ (<2.0 dS m$^{-1}$) to $S_2$ (12.0 dS m$^{-1}$) salinity level. In general, the treatment, which recorded comparatively less reduction in dry matter, recorded higher yield. These results are in agreement with those of Ghoulam et al. (2001), Singh and Usha (2003) and Khodary (2004).

5.3 CORRELATION STUDIES

The correlation studies indicated significant positive correlation with chlorophyll contents, proline content and nitrate reductase activity with total dry matter and seed cotton yield (Table 16).

The calcium and potassium contents and K/Na ratio in leaf showed significant positive correlation with total dry matter and seed cotton yield whereas, sodium content and Na/Ca ratio had at negative correlation at all stages (Table 17). These results are in agreement with those of Janardhan et al. (1979), Alfocea et al. (1993) and Bohra and Doerfling (1993).

5.4 ECONOMICS

The results on total cost of cultivation, gross returns, net returns and B:C ratio indicated significant differences between treatments and salinity levels (Table 18). Among the seed priming treatments, IAA and Ca(NO$_3$)$_2$ had higher total cost of cultivation (Rs.13,046 and 12,976), gross returns (Rs.49,526 and 47,343), net returns (Rs.36,480 and 34,367) and B:C ratio (2.79 and 2.64) respectively at $S_1$ (<2 dS m$^{-1}$) salinity levels compared to other treatments. In higher salinity level of $S_2$ (6.0 dS m$^{-1}$) and $S_3$ (12.0 dS m$^{-1}$), B:C ratio showed higher in seed priming treatments. IAA (2.11 and 1.43) and Ca(NO$_3$)$_2$ (1.40 and 1.27), respectively.

FUTURE LINE OF WORK

Salinity is a common wide spread problem in present day agriculture. Much work has been done on alleviation of problem with plant improvement and physical and chemical methods. But they are highly expensive, time consuming and does not suffice the objective. Hence, the future line of work advocates the use of PGR’s and nutrients to overcome the effects of salt stress on physiological, biochemical and yield component in crop plants. The results obtained in the present investigation help as mentioned below for further strengthening of research work on salt stress tolerance studies in cotton.

1. Testing of PGR’s and nutrients on large number of cotton genotypes under saline conditions for studying genotypic response
2. Study the ameliorative effects of PGRs and nutrients under drought stress in cotton
3. Study the ameliorative effects of PGRs and nutrients on germination percentage, seedling vigour, leaf area and on biophysical characters in cotton
4. Study the effect of PGR’s and nutrients on antioxidant enzyme activity under salinity stress in cotton
VI. SUMMARY

Environmental stresses are among the most limiting factors to plant productivity. Among these, salinity is one of the most detrimental factor. Salinization potentially limits the future of agriculture in most productivity areas of the world. Even though cotton is recognized as the most salt tolerant of all crops, its productivity can still be enhanced through use of plant growth regulators and nutrients as an alternate approach to mitigate the effects of salinity. Hence, the present investigations were carried out with seed priming through plant growth regulators (IAA and GA) and nutrients (Ca(NO\(_3\))\(_2\) and Na\(_2\)SO\(_4\)) and these were tested under three levels of salinity i.e. <2, 6.0 and 12.0 dS m\(^{-1}\) in RAHS-14 cotton genotype belonging to *Gossypium herbaceum* during 2005-06 at Agricultural Research Station, Gangavati. It was intended to study the ameliorative effects of PGR's and nutrients for the changes in growth, physio-biochemical and yield potential of cotton under saline conditions so as to know the salt tolerance mechanism.

1. In the present investigation, total chlorophyll content and its fractions a and b decreased with an increase in salinity, at 45, 90 and 120 DAS. In general, seed priming treatment with IAA and Ca(NO\(_3\))\(_2\) showed less reduction in total chlorophyll and ‘a’ and ‘b’ fraction whereas, control (G5) showed higher reduction followed by Na\(_2\)SO\(_4\) treatment.

2. Free proline content increased in all treatments with an increase in the salinity. The treatment IAA and Ca(NO\(_3\))\(_2\) accumulated more proline, while control (G5) accumulated least proline in leaves.

3. The nitrate reductase activity was maximum at 45 DAS followed by 90 and 120 DAS. With increase in salinity the nitrate reductase enzyme activity was decreased. Among the treatments, IAA showed higher activity at all the stages followed by Ca(NO\(_3\))\(_2\) and least enzyme activity was observed in control.

4. The treatment differences in ionic composition (Na\(^+\), K\(^+\) and Ca\(^{++}\)) were apparent under saline conditions in RAHS-14. There was general increase in Na\(^+\) content of leaves with increase in the salinity levels. Among the treatments, control (G5) and Na\(_2\)SO\(_4\) accumulated more sodium in leaf, whereas, IAA and Ca(NO\(_3\))\(_2\) had lower sodium accumulation at higher salinity levels at all the three stages. Moderate accumulation of Na\(^+\) was noticed in GA treatment.

5. On the other hand, potassium content decreased in leaf with increase in the salinity. However, the treatment IAA was found to possess higher potassium, moderate in Ca(NO\(_3\))\(_2\) and GA treatments and least in control.

6. The K/Na ratio was lowered due to salinity. Marked differences in K/Na ratio in leaves was observed among treatments. The seed soaking treatments with IAA and Ca(NO\(_3\))\(_2\) maintained higher K/Na ratio in leaves. While control and Na\(_2\)SO\(_4\) had lower K/Na ratio. Sodium content had negative relationship with seed cotton yield and total dry matter at harvest. Whereas, potassium content and K/Na ratio showed significant positive relationship.

7. Calcium content generally decreased with an increase in the salinity level particularly at higher salinity levels. The treatments IAA and Ca(NO\(_3\))\(_2\) had higher calcium content in leaves, whereas, the treatment control and Na\(_2\)SO\(_4\) had lower calcium content. Calcium content in leaf was found to have significant positive relationship with total dry matter and seed cotton yield.

8. Na/Ca ratio generally increased with increased levels of salinity. The treatment IAA had lower Na/Ca ratio in leaves followed by Ca(NO\(_3\))\(_2\). Whereas, the treatment control had higher Na/Ca ratio with increased levels of salinity, particularly at higher levels. The Na/Ca ratio had significant negative relationship with seed cotton yield and total dry matter content.

9. Total dry matter at harvest was reduced significantly due to salinity. The treatments IAA and Ca(NO\(_3\))\(_2\) recorded higher total dry matter at all salinity levels. While, GA and Na\(_2\)SO\(_4\) had moderate accumulation of dry matter at all salinity levels. Maximum reduction in total dry matter at harvest was observed in control.

10. Salinity reduced the seed cotton yield to an extent of 25 and 39 per cent at 6.0 and 12.0 dS m\(^{-1}\) salinity levels respectively as compared to control (<2.0 dS m\(^{-1}\)). Among the treatments, IAA had highest yield followed by Ca(NO\(_3\))\(_2\). The per cent reduction in seed cotton yield was least in IAA and highest in control (G5).

11. Yield components such as, number of bolls and boll weight were also affected by salinity. The treatment IAA and Ca(NO\(_3\))\(_2\) had higher boll number at all the salinity
levels while, control had least boll number. Similarly, boll weight was also more in IAA and Ca(NO$_3$)$_2$ treatment and less in control.

Based on the information generated from the present investigation, it can be concluded that treatments differed widely in their response to salinity and different plant growth regulators and nutrients may have different ameliorative mechanisms against salinity stress. Thus, information obtained would be useful in overcoming the salt stress by using PGRs and nutrients in cotton. Based on this study, among the plant growth regulators IAA (200 ppm) and nutrients Ca(NO$_3$)$_2$ (3%) would be better in mitigating the salinity stress and enhancing the cotton yield. From the investigations it could be concluded that higher yields obtained in IAA and Ca(NO$_3$)$_2$ treatments might be due to the mechanism of salt tolerance by maintenance of higher chlorophyll, proline, NR activity, potassium, calcium, K/Na ratio and lower sodium and Na/Ca ratio.
VII. REFERENCES


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ANONYMOUS, 2006b, Cotton Advisory Board, CCRI Report.


ASHRAF, M., FAKHRA, K. AND RASUL, E., 2002, Interactive effects of gibberellic acid (GA$_3$) and salt stress on growth, ion accumulation and photosynthetic capacity of two spring wheat (Triticum aestivum L.) cultivars differing in salt tolerance. Plant Growth Regulation, 36: 49-59.


APPENDIX I

Preparation of phosphatic buffer (0.1 M)

A: 0.2 M solution of Na$_2$HPO$_4$·7H$_2$O (35.61 g in 1000 ml of DW)

B: 0.2 M solution of Na$_2$H$_2$PO$_4$·2H$_2$O (31.21 g in 1000 ml of DW)

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EFFECTS OF SEED PRIMING WITH PLANT GROWTH REGULATORS AND MICRONUTRIENTS ON GROWTH AND YIELD OF COTTON (*Gossypium herbaceum* L.) UNDER SALINITY STRESS

SURESHER BABU S. 2006 Dr. B. S. JANAGOUDAR MAJOR ADVISOR

ABSTRACT

An investigation was made with an objective to mitigate salt stress by soaking cotton seeds of *Gossypium herbaceum* var. RAHS-14 with growth regulators (IAA and GA) and micronutrients (Na$_2$SO$_4$ and Ca(NO$_3$)$_2$) and tested under natural soil salinity levels of <2, 6 and 12 dS m$^{-1}$ during 2005-06 at Agricultural Research Station, Gangavati.

Seed priming treatments IAA and Ca(NO$_3$)$_2$ increased the yield and its components compared to other treatments under all salinity levels tested. The major contributing factors for enhanced yields are decrease in sodium content and Na/Ca ratio with increase in salinity and increase in potassium, calcium and K/Na ratio especially at higher salinity levels. The K/Na ratio was maintained in seed priming treatments IAA and Ca(NO$_3$)$_2$ by restricting the Na$^+$ uptake and increased K$^+$ uptake in shoot indicating the induced mechanism of salt tolerance in these treatments.

Among the biochemical parameters, chlorophyll 'a' and 'b' and total contents decreased with increase in salinity level. However, these contents increased in seed priming treatments with IAA and Ca(NO$_3$)$_2$ compared to rest of the treatments.

Similarly, the enzyme activity of nitrate reductase decreased with increase in salinity, whereas, free proline content increased. However, seed priming with PGR's and micronutrients increased the NR activity and free proline content under salinity stress considerably.

Based on the investigations, it could be concluded that among the plant growth regulators, IAA (200 ppm) and among the micronutrients, Ca(NO$_3$)$_2$ (3%) were found to be better in overcoming the effects of salinity stress through maintenance of higher chlorophyll, proline, nitrate reductase activity, potassium, calcium, K/Na ratio and reduced sodium content and Na/Ca ratio in leaf even at higher salinity levels.