## SUMMARY AND CONCLUSIONS

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Salinity in soil or water presents a stress condition for crop plants that is of increasing importance in agriculture. According to an FAO study salt-affected soils occupy nearly 7% of the world's land area (Massoud, 1974) and it is estimated that, in India, about 12 million hectares of land have been afflicted by the problems of soil salinity or alkalinity (Sharma and Gupta, 1986). As in other countries, in India also, the problem of soil salinity is becoming increasingly severe because an additional 15-20 million hectares of land in the canal irrigated tracts already run the risk of being degraded through the influence of salt (Abrol, 1986).

An increase in population demands an increase in agricultural production. Thus the need to produce more food grains, mainly due to population explosion, is continually pushing agriculture farther onto marginal lands often characterised by soils and waters with a high degree of natural salinity. Though a number of studies have been carried out with regard to salinity tolerance in plants (Levitt, 1980) the mechanism(s) by which salt inhibits plant growth is not yet clearly understood. Since it is not feasible to modify the saline environment to suit the crop plants, a thorough understanding of the mechanism(s) which salt inhibits growth would help in evolving some chemical treatments capable of modifying the plants to suit saline condition while maintaining reasonable and reliable yields.

Rice is one of the most widely cultivated cereal crops of India and it accounts for 40% of the total food grain production. It is found that growth and yield of most of the varieties of rice are adversely affected by salt stress. The present studies were, therefore, undertaken with a view to elucidating the mechanism(s) by which NaCl inhibits growth of rice (<u>Oryza sativa</u> L. var GR-3) and also to evolve some chemical treatments which would render rice tolerant to saline stress.

Polyamines (putrescine, spermidine and spermine) have recently been implicated in a number of stress-mediated responses of plants (Flores et al. 1984). Among the polyamines, putrescine has been shown to increase (Flores et al. 1984) or decrease (Priebe and Jager, 1978; Guye et al. 1986) in plant tissues under stress condition and the increase in putrescine content, as suggested by Slocum et al. (1984) may be of a protective nature conferring selective advantage to the stressed cells. Keeping this in view, the potential of putrescine in ameliorating the toxic effects of NaCl on growth and yield of rice has been examined. Earlier studies from this laboratory (Acharya, 1983) have shown that gibberellic acid (GA<sub>3</sub>) is also capable of reducing NaCl-induced inhibition of seedling growth of rice and hence the role of GA<sub>2</sub> on growth and yield of rice under saline condition has also been examined.

The effect of NaCl salinity (12 dS/m) and putrescine  $(10^{-5} \text{ M})$  or GA<sub>3</sub> (10 ppm) on growth and yield of rice has

been studied for a period of 60 days by culturing plants in pots. It has been found that NaCl at 12 dS/m salinity level decreased the extension growth, fresh weight and dry weight of shoot system to 55, 49 and 35 % respectively of the control on day 60 following salinization. Similarly the length, fresh and dry weights of root system were also reduced to 64, 53 and 43 % respectively of the control on day 60.

Application of putrescine, however, enhanced the extension growth of shoot and root system of salinized plants by 35 and 23 % respectively over the salt control on 60th day. The shoot and root systems of salt-stressed plants, as a result of putrescine application, produced 49 and 25 % respectively more fresh weight than the salt control at the end of 60 day period of the experiment. Saimilarly putrestinetreated salt-stressed plants showed an increase in their dry weight of shoot and root systems by 44 and 29 % respectively over the salt control on day 60.

 $GA_3$  administration, however, resulted in a more pronounced improvement in growth of salinized plants compared with putrescine-treated salt-stressed plants. On day 60, the linear growth, fresh weight and dry weight of shoot system of  $GA_3$ -treated salinized plants were, respectively 66, 70 and 85 % more than the salt control. The growth of root system of  $GA_3$ - treated salinized plants was much less compared with shoot growth. On day 60, the linear growth, fresh and dry weights of the root system of  $GA_3$ -treated

salinized plants registered an increase of 30, 45 and 37% respectively over the salt control. Non-salinized control plants also registered a significant increase in shoot growth in response to GA<sub>3</sub> application.

Soil salinity reduced the total leaf area to 58% of the control on day 55. However application of putrescine and  $GA_3$  resulted in a considerable alleviation (about 20 and 50% respectively more than that of the salt control on day 55) of salt-induced inhibition of leaf growth inhibition by NaCl and its partial reversal by putrescine and  $GA_3$ , the following parameters were examined:

- i) Contents of total chlorophyll, Na<sup>+</sup>, Cl<sup>-</sup>, K<sup>+</sup>, proline, total quaternary ammonium compounds, IAA, GA-like substances, ABA, polyamines @only in the case of control, salt-stressed, GA<sub>3</sub> - treated salt - stressed and GA<sub>3</sub>treated non-stressed plants) and total protein during different stages of shoot and root growth.
- activity of IAA oxidase, amylase, invertase, proline
  oxidase and agmatime deiminase during different stages
  of shoot and root growth.
- iii) extension growth of leaf.
- iv) contents of total chlorophyll and IAA and the activity of cellulase and pectin lyase during leaf growth.

It has been observed that there is a marked reduction in the content of total chlorophyll in salt-stressed plants compared with control. Putrescine and GA<sub>3</sub> considerably increased the chlorophyll content in salinized as well as in control plants. A dramatic increase in the levels of Na<sup>+</sup>, Cl<sup>-</sup>, proline and total quaternary ammonium compounds with a concomitant. decrease in the amount of K<sup>+</sup> were evident in salt-affected plants compared with control. Putrescine appreciably improved the ionic balance of the shoot system of salinized plants by diminishing the levels of Na<sup>+</sup> and Cl<sup>-</sup> and enhancing the amount of K<sup>+</sup>, even though it failed to make a similar trend in the root system.

 $GA_3$ , on the other hand, proved to be more effective than putrescine in improving the ionic balance under stress condition.  $GA_3$  substantially decreased the Na<sup>+</sup> and Cl<sup>-</sup> accumulation and noticeably increased the K<sup>+</sup> level in saltstressed plants compared with salt control. Proline build up of shoot system was slightly reduced by putrescine while no change was detected in the root system. Application of  $GA_3$ , however, resulted in a more conspicuous fall in the proline content of shoot system of salinized plants whereas root system registered only a small reduction in its proline content. Growth regulator treatments, except in the shoot system of  $GA_3$ treated salt-stressed plants, could not bring about any considerable change in the content of quaternary ammonium compounds in stressed or in non-stressed plants.

A marked decline in the levels of IAA and GA-like substances with a dramatic increase in the amount of ABA was detected in salt-stressed plants compared with control. The reduction in the level of IAA and GA-like substances by NaCl was partially reversed by putrescine treatment. The enhancement of IAA in salt-stressed plants by gibberellin application was, however, more conspicuous than that obtained with putrescine. NaCl induced accumulation of ABA was significantly reduced by putrescine as well as GA<sub>3</sub> treatments. But GA<sub>3</sub> was found to be more effective in checking the ABA build up in NaCl exposed plants than putrescine.

Salinization of plants led to a stimulation of IAA oxidase and an inhibition of amylase, invertase and proline oxidase activities. Putrescine treatment reversed the effect of salinity on the above enzyme systems in the shoot tissue whereas noticeable alteration in the enzyme activities in the root system was discernible only in the early part of the growth. GA3-treated salt-stressed plants, however, registered a higher reduction in the IAA oxidase activity than the putrescine-treated ones. Activity of amylase, invertase and proline oxidase was also much higher in GA3-treated salinized plants than the putrescine administered ones. GA3 increased activity of amylase and invertase in the shoot system of non-stressed plants too. Total protein content was found decreased as a result of salinization. Both the growth regulators, however, considerably enhanced the proline content in salinized plants as compared to the salt control.

The study of leaf growth revealed that salinity adversely affects linear growth, chlorophyll and IAA contents and the activity of cellulase during leaf growth. However no significant change in the activity of pectin lyase was discernible as a result of salinization. A noticeable improvement in the linear growth, levels of chlorophyll and IAA as well as in the activity of cellulase were recorded in the leaves of putrescine-treated salt-affected plants compared with salt control. When GA<sub>3</sub> was given exogenously a substantial increase in the leaf growth, chlorophyll and IAA content and the activity of cellulase was observed in salt-stressed plants compared with the salt control.GA<sub>3</sub> notably increased the chlorophyll content and the activity of cellulase as well as pectin lyase in the control leaves also.

As observed earlier (Priebe and Jager, 1978; Guye <u>et</u> <u>al</u>.1986) in other plants the polyamine content was found decreased in rice upon salinization. The reduction in putrescine and spermidine was significantly overcome by the exogenously supplied  $GA_3$ . This increase in putrescine and spermidine contents was found to be due to the enhanced activity of agmatine deiminase. No sizeable alteration in the content of spermine was, however, detected in response to  $GA_3$ treatment. The levels of putrescine and spermidine as well as the activity of agmatine deiminase in the control plants

were also enhanced by GAz.

Like vegetative growth grain yield too was greatly reduced by NaCl. Salt stress reduced the total number and weight of filled grains per plant and 1000 grain weight to 25, 20 and 76 % respectively of the control values. Application of putrescine increased the total number and weight of filled grains per plant and weight of 1000 grains by 54 and 67 and 8% respectively over the corresponding values of the salt control.  $GA_3$  had a more pronounced effect in enhancing the total number of filled grains per plant (85 % more than that of salt control) under saline condition. Total weight of filled grains was doubled by gibberellin treatment under saline condition. 1000 grain weight was also higher in GAz-treated salt-stressed plants (12 % more than the salt control) than in putrescinetreated salinized plants. However the grain yield was much less in putrescine or  $GA_3$ -treated salinized plants compared with control.

It is concluded from the present studies that NaCl inhibits growth and reduces the yield of rice by

- i) decreasing the contents of chlorophyll, K<sup>+</sup>, IAA, GA like substances, protein and polyamines as well as
  the activity of amylase, invertase and agmatine
  deiminase in shoot and root tissues.
- ii) decreasing the contents of IAA and chlorophyll and the activity of cellulase during leaf development.

iii) increasing the amounts of Na<sup>+</sup>, Cl<sup>-</sup>, ABA, free proline, quaternary ammonium compounds and the activity of IAA oxidase in shoot and root tissues.

Putrescine was found to increase growth and yield by enhancing the extension growth, fresh and dry weights of shoot and root systems and total leaf area under saline condition. This improvement in growth and yield in response to putrescine treatment under saline condition could be due to

- i) the improved ionic balance (putrescine considerably decreased the net accumulation of Na<sup>+</sup> and Cl<sup>-</sup> and enhanced the K<sup>+</sup> content in salt-stressed plants, especially in the shoot system).
- ii) the considerable reduction in the content of ABA with a concomitant increment in the amounts of IAA and GAlike substances.
- iii) the diminution of IAA oxidase and enhancement of amylase, invertase and cellulase activities.
- iv) the reduction in the content of free proline and the build up of protein content.

Interestingly, all the parameters exhibited a better improvement under the influence of  $GA_3$  than with putrescine in salt-affected plants. Further,  $GA_3$  increased the activity of agmatine deiminase and the production of polyamines which also might have augmented the growth and yield of rice under saline condition.