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# INFLUENCE OF SALINITY ON THE MORPHOLOGY AND PHYSIOLOGY OF *AMARANTHUS DUBIUS* [CALLALOO] AND *CAPISCUM CHINESE* VAR. SCOTCH BONNET.

Sasikala D.P. Potluri and P.V. Devi Persad
Department of Life Sciences, The University of the West Indies
Mona, Kingston 7, Jamaica

## ABSTRACT

The effects of salt stress on the morphology, growth and physiology of *Amaranthus dubious* [callaloo] and *Capsicum chinense* [var. scotch bonnet pepper] were investigated. Various concentrations of sea salt have been used to obtain different salinity levels of the soil from 0 to 10 dS $m^{-1}$. Low levels of salinity at 2 dS $m^{-1}$ actually enhanced the growth of the plants as measured by the shoot height. Concentrations higher than this inhibited the growth of callaloo. For pepper, salinity level of 4.0 dS $m^{-1}$ was also not that inhibitory. However, salt levels above that level inhibited growth and the older leaves first turned yellowish in colour. There was a positive correlation between the increase in the level of proline in the plant tissues and salt concentration. Proline content was higher in the shoot than the root. Soluble carbohydrates increased with increasing salinity levels in both callaloo and pepper. The protein levels decreased with increasing salinity levels in callaloo but remained unchanged in pepper. The activity of the enzyme nitrate reductase was inhibited above concentrations of 4.0 dS. $m^{-1}$ in both plants and the inhibition was severe in the roots. The results indicate that callaloo is more sensitive to salt stress than scotch bonnet pepper and slightly different mechanisms are involved in the salt tolerance in callaloo and pepper.

## INTRODUCTION

Salt stress is one of the principal factors causing reduction in plant growth and generally in agricultural production. Salinity can inhibit plant growth by reduced external water potentials, toxicity through excess ions and general imbalance of ions [Greenaway and Munns, 1980]. Plants differ in their response to salinity depending upon the genotype and environmental conditions. The salinity of Caribbean soils have been increasing due to the frequency of droughts and faulty irrigation systems. In Jamaica about 25% of the total arable land is believed to be under some sort of saline stress. The situation could be similar in other Caribbean countries. Considering the fact that most of these countries are hilly and the total arable land is relatively less [between 30 to 50%], salt stress does create serious problems for agricultural productivity. Therefore, it is important to identify the level of tolerance to salt stress in some important crops in the Caribbean so that currently underutilized/ unutilized lands could be brought in to production thereby increasing agricultural productivity in the Caribbean.

Callaloo is an important leafy vegetable and scotch bonnet pepper is an important commodity crop in Jamaica. The present work has been undertaken to study the effects of salt stress on these two crops.

## MATERIALS AND METHODS

Seeds of callaloo and scotch bonnet pepper were obtained from local farmers. They were germinated in small trays on sterile sand/soil mixture in the green house. Plastic pots 22cm in diameter and same depth were filled with 5kg of sandy loam sterile soil. Holes were drilled for drainage in these pots. Two seedlings of uniform size and at four-leaf stage were transplanted in to each pot. After one week, the pots were subjected to the following saline treatments: control, salinity levels [EC] of 2, 4, 6, 8, and 10dS $m^{-1}$ Salinity levels were obtained with the help of crude sea salt dissolved in tap water. Each treatment had 10 pots. The root medium salinity levels were maintained through leaching and replenishing with appropriate salt solution every three days. The plants were protected from rain but otherwise grown in open for 6 weeks.

## OBSERVATIONS

The following parameters were observed:

Shoot height [cm], proline content of the shoot and root [Bates et al. 1973], soluble carbohydrate content [Dubois et al. 1956], protein content [Lowry et al. 1951], sodium and potassium content [flame photometry] and the activity of the enzyme nitrate reductase [ Davison and Stewart, 1984].

## RESULTS

Shoot height: Shoot height actually showed a slight increase in both plants at salinity levels of 2dS $m^{-1}$. [Table 1] In callaloo, there was severe reduction above salinity levels of 4.0dS m-1 and the plants in 10dS $m^{-1}$ died with in 4 weeks. In scotch bonnet pepper, the plants were looking healthy up to 6.0dS $m^{-1}$ except for a slightly stunted appearance. Above these salt levels, plants showed brittle leaves, older leaf yellowing, and brittle stems. Shoot tips started showing necrosis, however, the plants survived even at the highest salt level used.

**Table 1. Effect of various concentrations of salt onthe shoot height of Amaranth and Capsicum.**

| Salt concn. 0.0dS | 0.2 | 0.4 | 0.6 | 0.8 | 1.0 |
|---|---|---|---|---|---|
| *Plant sp.* | Shoot height in mm | | | | |
| Amaranth | 45+4.8 | 52+4.4 | 41+4.7 | 31+3.8 | 18+4.2 nm |
| Capsicum | 38+4.1 | 42+3.5 | 37+3.9 | 31+2.3 | 19+1.2 13+1.1 |
| nm = not measured | | | | | |

Proline Content: There was a positive correlation between the proline content and salt levels used in both plants and in shoots as well as roots [Table-2]. However, scotch bonnet pepper showed a higher increase in proline in the shoot than callaloo. The highest proline levels [38.8μ mol] were observed in pepper grown at 8 dS $m^{-1}$ while at the same concentration of salt, callaloo had a proline level of 23.9μ mols. In general, the proline content of the shoot was much higher than the roots.

**Table 2. Effect of salt concentrations on the Proline content of Amaranth and Capsicum.**

| Salt conc. [dS] | | 0.0 | 2.0 | 4.0 | 6.0 | 8.0 |
|---|---|---|---|---|---|---|
| *Plant sp.* | | Proline content (m mol. g-1 fresh wt) | | | | |
| Amaranth | Shoot | 3.8+0.2 | 4.1+0.2 | 18.8+0.9 | 23.6+0.8 | 23.9+0.8 |
| | Root | 1.2+0.1 | 1.3+0.1 | 3.7+0.2 | 4.5+0.2 | 4.5+0.1 |
| Capsicum | Shoot | 4.5+0.3 | 5.8+0.2 | 21.6+0.9 | 33.4+1.1 | 38.8+1.8 |
| | Root | 1.6+0.1 | 1.8+0.1 | 3.4+0.2 | 4.6+0.1 | 4.8+0.1 |

Protein content: The protein content of callaloo decreased from 11.6% in the control to 7.3% in plants grown at 8 dS $m^{-1}$ salt levels [Table 3]. There was a corresponding decrease in roots also but this was not as pronounced as in the shoot. On the other hand, protein levels in pepper have not changed markedly in responded to salt stress. There was only a slight decrease from 10.5% protein in the shoot in control plants to 9.7% in plants grown at 8 dS $m^{-1}$.

**Table 3. Protein content of sboot and root of Callaloo and peppersubjected various levels of salt stress.**

| Salt conc. [$dSm^{-1}$] | | 0.0 | 2.0 | 4.0 | 6.0 | 8.0 |
|---|---|---|---|---|---|---|
| ***Plant sp.*** | | **Protein content [% dry wt.]** | | | | |
| Amaranth | Shoot | 11.6+0.3 | 11.8+0.3 | 10.3+0.4 | 8.3+0.2 | 7.3+0.3 |
| | Root | 7.3+0.2 | 7.2+0.3 | 6.8+0.2 | 6.3+0.2 | nm |
| Capsicum | Shoot | 10.5+0.3 | 11.3+0.4 | 10.6+0.3 | 9.8+0.2 | 9.7+0.2 |
| | Root | 6.8+0.1 | 6.8+0.2 | 6.9+0.2 | 6.4+0.4 | 6.5+0.2 |

nm = not measured

Carbohydrate content: Soluble carbohydrate content increased from 18.8% in the shoot of callaloo control plants to 31.5% in the plants grown at 8 dS $m^{-1}$[Table 4]. A similar increase from 21.1 to 29.8% was seen in pepper. Roots also showed an increase in carbohydrate content but the increase was not substantial.

**Table 4: Soluble carbohydrate content of Callaloo and pepper subjected to various levels of salt stress.**

| Salt conc. [$dSm^{-1}$] | | 0.0 | 2.0 | 4.0 | 6.0 | 8.0 |
|---|---|---|---|---|---|---|
| ***Plant sp.*** | | **Carbohydrate content [% dry wt.]** | | | | |
| Amaranth | Shoot | 18.8+0.8 | 21.4+0.7 | 24.3+0.7 | 29.9+0.8 | 31.5+0.6 |
| | Root | 14.4+0.7 | 14.8+0.5 | 15.3+0.6 | 18.8+0.6 | nm |
| Capsicum | Shoot | 21.1+0.5 | 23.8+0.8 | 26.4+0.6 | 28.9+0.9 | 29.8+0.6 |
| | Root | 15.5+0.3 1 | 5.8+0.6 | 17.5+0.4 | 17.7+0.6 | 17.8+0.5 |

nm – not measure

Potassium and sodium levels: Sodium levels were much higher in roots than in shoots in callaloo [table 5]. However, in pepper, there were more or less similar. There was a sudden jump in the sodium levels of the shoot between plants grown at 4dS $m^{-1}$ and 6 dS $m^{-1}$ salt levels, from 28 mg.$kg^{-1}$ to 67 mg.$kg^{-1}$ in callaloo and from 33 to 71 in pepper. Potassium levels also increased but gradually in response to increasing salt levels.

Nitrate reductase: The activity of this enzyme showed a decrease in both plants. The decrease was more in the shoots of callaloo, decreasing from 1.5 µmol. $gm^{-1}$ in control to 0.5 µmol.$g^{-1}$in plants grown at 6 dS $m^{-1}$ salt levels. In pepper, the corresponding decrease was less, from 1.3 to 0.8 [Table 6]. However, activity of nitrate reductase was very severely inhibited in the roots of pepper compared to the roots of callaloo.

## DISCUSSION

The results clearly indicate that low levels of salinity is actually beneficial for the growth and metabolism of both plants – Amaranthus dubious [callaloo] and Capsicum chinense [var. scotch bonnet]. The growth parameters and biochemical composition indicate that there is no substantial decrease in any of the useful components of the two plants up to 4 dS m-1 salt levels.

**Table 5: Effect of salinity stress on the sodium and potasium content of Callaloo and pepper [expressed as mg.Kg$^{-1}$].**

| Salt concn. [dS.m$^{-1}$] | Concn. of sodium and potassium [mg. Kg$^{-1}$ dry st.] | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 0 | | 2 | | 4 | | 6 | |
| *Plant sp.* | Na | K | Na | K | Na | K | Na | K |
| Amaranth Shoot | 7+0.4 | 26+1.3 | 16+0.9 | 48+2.1 | 28+3.2 | 71+4.4 | 67+3.8 | 94+5.4 |
| Root | 8+0.3 | 28+1.8 | 18+1.4 | 44+2.2 | 43+3.4 | 56+4.3 | nm | nm |
| Capsicum Shoot | 8+0.3 | 28+1.2 | 18+0.9 | 54+3.6 | 33+2.2 | 71+4.3 | 71+4.5 | 101+5.4 |
| Root | 9+0.4 | 22+2.1 | 17+1.8 | 34+3.4 | 39+4.3 | 73+4.4 | nm | nm |

nm = not measured

**Table 6. Effect of salt stress on the activity of the enzyme nitrate reductase in Callaloo and pepper.**

| Salt Concn .[dS] | Nitrate Reductase Activity [mmol nitrite. g$^{-1}$ fresh wt.] | | | |
|---|---|---|---|---|
| | 0 | 2 | 4 | 6 |
| Callaloo Shoot | 1.5+0.03 | 1.7+0.03 | 1.1+0.02 | 0.5+0.01 |
| Root | 0.5+0.02 | 0.7+0.1 | 0.4+0.1 | 0.15+0.01 |
| Pepper Shoot | 1.3+0.02 | 1.6+0.03 | 1.2+0.03 | 0.8+0.02 |
| Root | 0.4+0.01 | 0.4+0.01 | 0.35+0.01 | 0.02+0.01 |

Most of the work on the effects of salt stress on Capsicums has been carried out on *Capsicum annum* and *Capsicum fruitescens* and to a lesser extent on a few other species but not on C. chinense. Hasheem et al. [1991] studied the germination ability of seeds of C. annuum . They found that at higher concentrations of sea salt, seedling growth was inhibited while germination was not. Cornillon and Palliox [1997] studied the influence of NaCl on four cultivars of C. annuum and observed that cv. Yolo wonder [sweet pepper] was most sensitive. *Capsicum chinense* var. scotch bonnet in the present study showed a similar behavior in that salt levels up to 4.0dS m$^{-1}$ were actually beneficial and even at 6.0dS m$^{-1}$, the growth was not severely affected. In this respect, C. chinense appears to be more tolerant than sweet pepper varieties.

It has been well established that environmental stress, specially water stress and salt stress, results in the accumulation of organic compounds like proline in many plants [Dix and Pearce, 1981; Katz and Tal, 1980; Pandey and Ganapathi, 1985; Stewart and Lehrer, 1980, Aspinal and Paleg, 1981, Devi Prasad and Potluri, 1993, 1996]. Proline accumulation is generally believed to be as an osmo-regulation process and general protection for cytoplasmic enzymes [Aspinall and Paleg, 1981]. In both Amaranthus dubious and Capsicum chinense in the present study, proline levels increased due to salt stress, though more proline accumulation was evident in pepper than in Callaloo. The accumulation was more in the shoot than in the root suggesting that there was more protection in the shoot against salt stress. The proline levels did increase in the roots but to a lesser extent. Roots are in direct contact with the soil solution and therefore would normally be expected to develop protective mechanisms to withstand the osmotic shock. If they cannot protect the membrane system, then more ion accumulation, especially that of Na, will result. The root system is poorly developed at higher concentrations and more Na is accumulated in the shoot system in both callaloo and scotch bonnet pepper. It appears that while the root system in general is inhibited, the xylem transport is not that hampered as higher ion accumulation is observed in the shoot system. Al-Bahrany [1994] and Gunes et al. [1996] observed increased levels of proline in the leaf tissue of *C. annum.*

Carbohydrates are also important for withstanding the osmotic shock and several salt stressed plants accumulate higher amounts of carbohydrates[ Misra and Dwivedi, 1995, Devi Prasad and Potluri, 1993, 96, ]. Very little information is available on the accumulation of carbohydrates due to salt stress on both Amaranths and Capsicums. In the present study, soluble carbohydrates increased in both plants.

The accumulation was higher in the shoot but in the roots, the accumulation was not inhibited compared to the control. This suggests salt stress did not cause transport problems for carbohydrates.

In Amaranth it is possible that higher carbohydrate accumulation was at the expenses of proteins as lower protein levels were observed. But in scotch bonnet pepper, protein content was not affected by salt stress. The accumulation of carbohydrate therefore must be either due to increased photosynthetic activity or at the expense of some other growth parameter. Since the plants were stunted at higher concentrations of salt, it is possible that the unavailability of carbohydrate for growth may be the reason for reduction in shoot height.

The levels of Na and K as affected by salt stress have been used to determine the level of salt tolerance in many plants. In the present study, levels of both Na and K increased in callaloo and pepper, though the increase was more in Na content. The ratio between K and Na changed in favor of Na with the increase in the level of salt. The levels of these ions were higher in shoot than the root, indicating that transport was not a problem. Similar higher levels of both Na and K in leaves and stems were found in Artiplex [Aslam et al. 1986], in tomato [Gill, 1990], in Capsicum annum [Al-Bahrany, 1994, Gunes et al. 1996, Gomez et al. 1996] and in Amaranths caudatus [Breus et al. 1994].

Studies on the effects of salt stress on enzymes is usually concentrated on polyphenol oxidase, however, N nutrition is an important factor in plant growth. Nitrate is usually transported to shoot and reduced there by the enzyme nitrate reductase. Comparatively lower levels of NR activity is found in roots. This was confirmed in the present results with both plants. In callaloo, the shoot NR activity was more affected.

This would increase metabolic cost to the plant, as roots will have to reduce nitrate for their use instead of receiving from leaves. The opposite happened in pepper, where the root NR was severely inhibited compared to the shoot. As overall protein content in callaloo declined, decrease in NR activity may partly be attributed to the inhibition of synthesis of the enzyme, which is a protein. However, the inhibition in pepper seems to be at the activity level than the synthesis of the enzyme, as protein content did not decrease significantly.

The present results clearly establish that the scotch bonnet pepper is more tolerant of salt stress than callaloo and that both plants follow slightly different mechanisms of tolerance. Callaloo can be grown successfully up to EC levels of 4 $dSm^{-1}$.

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