Response of Amaranth to salinity stress

by

Elizabeth Nabwile Omami

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Supervisor: Prof. P. S. Hammes

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DECLARATION

I, Elizabeth Nabwile Omami, hereby declare that this thesis for the degree Ph.D. in Horticulture at the University of Pretoria, South Africa, is my own work and has never been submitted at any other university.

Elizabeth Nabwile Omami

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Responses of Amaranth to salinity stress

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Elizabeth Nabwile Omami

Supervisor: Prof. P. S. Hammes

Department: Plant Production and Soil Science

Degree: Ph.D.

ABSTRACT

Salinity continues to be one of the world's most serious environmental problems in agriculture. The increasing world population and urbanization are forcing farmers to utilize marginal lands as well as poor quality water. One of the strategies in dealing with salinity is growing salt tolerant plants and there has been increased need to understand the effects of salinity on crops. Owing to its high nutritive value and wide adaptability to diverse environments, amaranth is considered a promising crop for marginal lands and semiarid regions. The objective of the study was to investigate the response of amaranth to salinity stress and evaluate stress amelioration by calcium and seed priming.

Salinity tolerance during germination and early seedling growth was examined for six genotypes of amaranth (*Amaranthus* species) at different salt concentrations ranging from 0 to 200 mM NaCl or Na_2SO_4. Enhancement of germination was observed at 25 mM, while increasing salt concentrations reduced the germination percentage as well as germination rate. *A.tricolor* and Accession '83 were able to germinate in 200 mM NaCl while there was no germination at 200 mM Na_2SO_4 in all the genotypes. Overall, Accession '83 was the most resistant and *A. hybridus* the most sensitive genotype, particularly at high salt concentrations. Inhibition of germination was greater in Na_2SO_4

than in NaCl salinity treatments. Amaranth was more salt tolerant at germination than at seedling growth. Seedling emergence, survival and growth were reduced by salinity and at much lower concentrations than at seed germination. Differences in salt tolerance were noted among the genotypes.

Salinity stress was initiated at different growth stages (cotyledon stage, 2-leaf stage and 4-leaf stage) in order to determine whether tolerance of amaranth differs with the stage of development. The treatment either continued until termination of the experiment or for 14 days at each stage. Amaranth plants were less sensitive to salinity when the stress was initiated at the 4-leaf stage. Lower salt concentrations had less detrimental effects than higher concentrations when applied at the cotyledon stage. Application of low salt concentration at cotyledon stage for 14 days did not have any effect on plant growth. The results indicate that it is feasible to use saline water for growing amaranth with minimum yield losses if salt concentration, duration of exposure and time of salinization are carefully managed.

Differences in salinity tolerance among amaranth genotypes were analyzed in terms of plant survival, growth, gas exchange, water use and leaf anatomical changes. *A. hypochondriacus* and *A. cruentus* showed greater tolerance to salinity since they survived in 200 mM NaCl treatment and the reduction in growth at 50 and 100 mM was lower than that of *A. tricolor* and Accession '83. *A. hypochondriacus* and *A. cruentus* were more efficient water users and partitioned photosynthates towards shoot growth as opposed to the other two genotypes. Photosynthetic rate, stomatal conductance, stomatal density and apertures were reduced by salinity but were higher in *A. tricolor* than in *A. cruentus.* Salinity resulted in *A. cruentus* developing thicker leaves compared to *A. tricolor*. Productivity on saline soils can be increased by growing genotypes more tolerant to salinity.

The interactive effect of salinity and water stress on amaranth plant growth was evaluated. It was found that the reduction in shoot growth was greater in plants submitted

to water stress than in those submitted to salt or salt + water stress. Water use efficiency was increased while leaf water and osmotic potentials were reduced by the salinity stress treatments. In drying soil plants previously salinized had a greater degree of osmotic adjustment, so that plants were able to continue growth for a longer period compared to water stressed plants.

The effect of calcium in ameliorating salt stress was investigated. Supplementary calcium, either as $CaSO_4$ or $CaCl_2$ ameliorated the negative effects of salinity on growth, gas exchange, membrane permeability and mineral uptake. In a separate experiment it was shown that it is feasible to mitigate the adverse effects of salinity on amaranth seed germination, seedling survival and growth by seed priming and that the positive effect of priming persisted to vegetative growth stage. Priming with $CaSO_4$ + NaCl showed a greater positive response than priming with the individual salts.

Key words: *Amaranthus*, calcium, gas exchange, germination, growth, membrane permeability, photosynthesis, salinity stress, salt tolerance, seed priming, water relations, water stress, water use efficiency (WUE).

INTRODUCTION

Salinity is one of the world's most serious environmental problems in agriculture. It is estimated that about one-third of the world's cultivated land is affected by salinity (Perez-Alfocea *et al*., 1996). The National Academy of Sciences of the USA includes salinization of soils and waters as one of the leading processes contributing to a possible worldwide catastrophe (Francois and Maas, 1994). The increasing world population, especially in arid and semi-arid regions, food shortages, and land scarcity are compelling the use of lands not utilized because of salinity and other soil stresses. Salinity and sodicity problems are characterized by an excess of inorganic salts and are common in the arid and semi-arid lands (ASAL) where they have been naturally formed under the prevailing climatic conditions and due to the high rates of evapotranspiration and lack of leaching water (Mengel and Kirkby, 1982; Shannon *et al*., 1994). In the arid and semi-arid parts of Africa, for instance, salinity and alkalinity are major problems affecting about 24% of the continent (Reich *et al*., 2004). According to Eswaran *et al*. (1997) about 30% of the population of Africa or about 250 million people are living on or are dependent on this type of land.

Although more frequent in arid lands, salt-affected soils are also present in areas where salinity is caused by poor quality of irrigation water and increases markedly during the dry season (De Pascale *et al*., 1997; Sifola and Postiglione, 2002). As competition for fresh water increases due to increasing population, water of better quality is used primarily for domestic purposes, whereas water of lower quality e.g. saline or polluted water (drainage water generated by irrigation agriculture, marginal-quality waters generated by municipalities) often is used for irrigation (Khroda, 1996; Oster, 2000; Bouwer, 2002). Wanjogu *et al*. (2001) reported that land degradation by salinization is on the increase where the use of poor quality irrigation water is a common practice in arid and semi-arid lands. However, there seems to be a general lack of information on the prevalence and composition of saline aquifers in sub-Saharan Africa (Karlberg and Penning de Vries, 2003). Although some countries, such as South Africa, Botswana and

Zimbabwe have documented the presence of saline aquifers, Karlberg and Penning de Vries (2003) reiterated that information as to what extent saline water is being used for irrigation in sub-Saharan Africa is lacking.

From the perspective of plant productivity, salinity problems accentuate year after year as a result of repeated irrigation with poor quality water. The concentration of salt in the soil rises due to evaporation. Similarly, the salt concentration in plant tissues increases as water is lost through the process of transpiration. Salt problems unrelated to irrigation are also known. Excessive fertilizer application, susceptible soil types, and drought can combine to play a major role in accentuating the toxic effect of salts on crop yield (Parker *et al*., 1983).

Agricultural production in arid and semiarid regions of the world, which depends on irrigation, faces a serious challenge because it must increase or at least maintain crop productivity while coping with ever more saline irrigation water. Irrigation with saline water is successfully practiced in many countries such as Israel, Italy and the US (Rhoades *et al*., 1992). The success of using such water is dependent on advances in the knowledge of the many factors involved in plant salt tolerance.

Salt accumulation in soils induces physiological and metabolic disturbances in crops affecting development, growth, yield and quality of crops (Pardossi *et al.*, 1999; Mavrogianopoulos *et al*., 1999; del Amor *et al*., 2000; Mer *et al*., 2000; Silveira *et al*., 2001). Reduction in growth results from salinity effects on dry matter allocation, ion relations, water status, biochemical reactions or a combination of many physiological factors. However, the severity of salt damage has been found to be dependent on the meteorological conditions, soil type (Shannon *et al*., 1994), species and cultivar (Rhoades *et al.*, 1992; Vicente *et al*., 2004), growth stages of the plant (Yeo *et al.*, 1991; Botia *et al*., 1998; Carvajal *et al*., 1998), time interval between irrigations, amount of water distributed and time of exposure to saline water (Oster, 1994). Such variability suggests that environment and species-specific assessments of plant salt tolerance are both required in order to obtain conclusive information regarding the cultivation of a certain species using saline water of specific concentration. This is due to the fact that general guidelines for agricultural management may not respond to the species and/or environment-specific crop requirements for optimum production (Rhoades *et al*., 1992; Dalton *et al*., 2000).

The degree of salt stress on a plant depends on three responses:

- Shoot dehydration through the low water potential caused by osmotic stress (Dubey, 1997; Carvajal *et al*., 1999);
- Nutritional imbalances caused by the interference of saline ions with essential nutrients in both uptake and translocation processes or partitioning within the plant (Liu and Zhu, 1998; Grattan and Grieve, 1999);
- Specific ion toxicity due to the accumulation of ions, particularly Na^+ and Cl^- in the cytoplasm (Greenway and Munns, 1980; Yeo, 1998; Wahome *et al*., 2001).

These three effects often coexist in soils exposed to salinization, and can persist throughout the growth season (De Pascale *et al*., 2003a, b).

Sustained and profitable production of crops on salt-affected soil is possible if appropriate on-farm management practices are carried out. To be successful, growers require an understanding of how plants respond to salinity, the relative tolerances of different crops and their sensitivity at different stages of growth, and how different environmental conditions affect salt-stressed plants. A widespread practice to reduce the salt content in soils is leaching. However, with the rising cost of water, this may not continue to be a feasible method for the future. Increasing salinity has increased the need to understand the effects of salinity on crops, and genetic improvement of salt tolerance has become an urgent need for the future of agriculture in arid and semi-arid regions (Shannon, 1984; Owens, 2001). Breeding of salt tolerant crop varieties will require a clear understanding of plant response to salinity and the complex mechanisms of salt stress tolerance (Zhu, 2001; Apse and Blumwald, 2002).

As the problems of salinity become more severe Aronson (1985) and NRC (1990) have proposed the growing of alternative plants and crops suited to moderately saline conditions with the option of introducing under-exploited, salt-tolerant minor crops. Salt tolerant plants may provide a logical alternative for many developing countries.

Amaranth is used for its grain and is also consumed as a cooked vegetable in many parts of the world. Owing to its high nutritive value and a wide adaptability to diverse environments, amaranth has been considered a promising crop for marginal lands and semiarid regions (Cunningham *et al*., 1992; Allemann *et al*., 1996). The prospects for future cultivation of salt-tolerant, high-yielding genotypes of amaranth are very encouraging. However, despite a substantial amount of literature on responses of plants to salinity stress, little information is available on amaranth. The general objectives of this study were to better understand the response of amaranth to salinity stress by investigating:

- Differences in salinity tolerance in different amaranth genotypes at seed germination and vegetative growth stages.
- Salt tolerance of amaranth as affected by salt concentration and timing of salinity imposition after seedling emergence.
- Morphological and physiological traits that contribute to salinity tolerance in amaranth.
- Interactive effects of salinity and drought stress on amaranth growth and development.
- The ameliorative effects of calcium nutrition and seed priming on growth, mineral uptake and photosynthesis of salt-stressed amaranth.