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Full Length Research Paper

Effects of salt stress on micropropagation of potato (*Solanum tuberosum* L.)

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Six cultivars of potato (*Solanum tuberosum* L.) Bartina, Spunta, Cardinal, Desirée, Timate and Fabula were assayed *in vitro* for salinity (NaCl) tolerance. A modified single-node cuttiang bioassay was used in which cultivars were exposed to a range of NaCl levels (0, 40, 80 and 120 Mm), in a Murashige and Skoog medium, for 1 month. Evaluations were performed twice for each cultivar at each salt level. Six vegetative growth parameters (shoot and root length, fresh and dry weights) were measured at the time of harvest and corrected for differences in cultivar vigor. Fresh weight of shoot and length of shoot decreased by given range levels of NaCl. The effect of all treatments was very drastic on length and weight of root. The cultivar Bartina exhibited greater tolerance to the highest salt doses. Plants of this cultivar had greater shoot length than other cultivars at all salt concentrations tested. The results indicated that cultivar "Bartina" is more salt tolerant than other cultivars studied.

Key words: Potato, *Solanum tuberosum* L, salinity, *in vitro*, micropropagation.

INTRODUCTION

Salinity is an abiotic factor that combines elements of water deficit and sodium (Na) toxicity (Bohnert et al., 1995), is among the most serious and widespread of agricultural problems contributing to lost crop yield and arable land. At least, 25% of the world's cultivated land area is occupied by salt-affected soil, primarily in semi arid and arid areas (Flowres and Yeo, 1995).

Under more favorable environments, where land is intensively cultivated, salinity problems may develop where quality irrigation water is limited or mismanaged or drainage is lacking (Morpurgo, 1991). In the past, major efforts to circumvent salinity were directed to soil reclamation and water desalination. Modifying the environment to suit the plant is increasingly expensive, which provide strong impetus to the development of plants with increased salinity tolerance (Zhang et al., 1993).

The cultivated potato (*Solanum tuberosum* L.) is an internationally important stable food crop. It is grown in 79% of countries and in volume of production rank fourth globally after rice (*Oryza sativa*), wheat (*Triticum aestivum*) and maize (*Zea mays*) (Woolfe, 1987). Potatoes are classified as moderately salt-sensitive (Maas and Hoffman, 1977) tolerance to irrigation with saline water evaluated on several potato cultivars and species in pot (greenhouse, screenhouse) or field trials in Pakistan (Ahmad and Abdullah, 1979), India (Paliwal and Yadav, 1980) Tunisia (Hannachi et al., 2004) and other countries (Bilski et al., 1988; Levy et al., 1988; Morpurgo, 1991). Difference in salinity tolerance existed among cultivars and species, salinity depressed yield and generally had a greater effect on tuber weight than tuber number. Assessment of salinity tolerance in the field is seasonally constrained, affected by climate and may lack reliability due to combined salinity and water stress problems. Variable salt composition and uneven distribution in the field may also tend to confound field screening (Shannon, 1984). More reliable and time-sav-ing selection techniques have been developed using tissue culture technology.

In vitro determination of salinity tolerance, utilizing nodal cuttings of tissue culture propagated (Micropropagated) plants enabled ranking of potato culti-vars and wild species (Pour et al., 2009; Silva et al., 2001; Morpurgo and Silva-rodriguez, 1987).

A highly significant correlation was found between *in vitro* growth parameters and the field performance of ten potato clone exposed to a high level of NaCl (Morpurgo, 1991). In this study, some of the morphological changes occurring under different salt stresses were studied for

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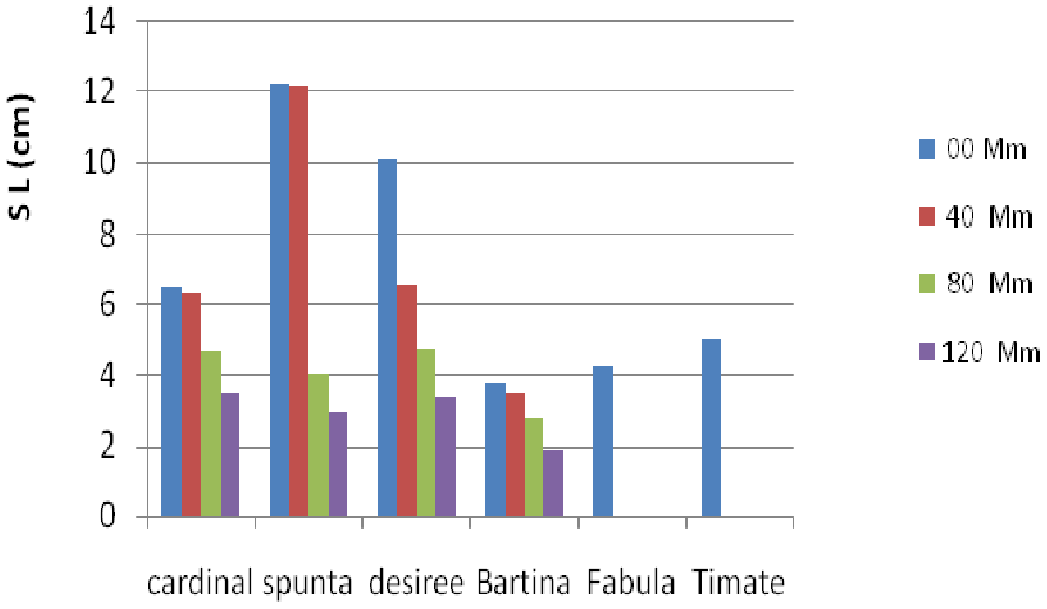

Figure 1. The effect of salinity on *Solanum tuberosum* cultivars' shoot length.

potato plants grown *in vitro*. A description of the plant's responses to NaCl stressing conditions was given.

MATERIALS AND METHODS

Plantlets of six cultivars of potato (*S. tuberosum* L.) Bartina, Spunta, Cardinal, Desirée, Timate and Fabula were used in this study. Nodal cutting were used as explants. The explants were surface sterilized with 70% ethanol for 1 min followed by sodium hypochlorite (2%) for 10 min and thoroughly washed with sterile distilled water for three times (Aazami et al., 2010a). The explants were cultured on semi solid MS (Murashige and Skoog, 1962) media containing 30 g sucrose and 8 g agar (pH 5.8) dispensed in test tubes. Test tubes were incubated at 25±2°C under 16 h illuminations (70 µ mol m^{-2} s^{-1}). Salinity was simulated by the addition of NaCl at four concentrations (0, 40, 80 and 120 Mm). After 30 day, the plantlets were destructively harvested and six vegetative growth parameters were measured. The shoot and root lengths were measured, from the point of emergence of the lateral bud on the original single-node cutting or the point of emergence of the roots to the tip of the organ, respectively. The shoot and root fresh and dry weights were measured at the time of harvest and after 60 h in a 60±2°C convention oven, respectively. Data were subjected to ANOVA and LSD analysis.

RESULTS

The results of this study show that the measured parameters of growth are inversely proportional with the NaCl concentration. The two cultivars, Fabula and Timate do not reveal any growth in the presence of salt even with weak concentrations. However, a relative deterioration was recorded on the growth at the other cultivars.

Shoot and root length and root but not shoot dry weights significantly decreased (p= 0.001) in the nodal cuttings challenged with salt, compared with the growth of control nodal cutting for the four cultivars.

Concerning the effect of the salinity of the medium over the length of the shoot, the statistical analyses revealed three groups (Figure 1). The first gathers the control medium (0 Mm NaCl) and the two mediums with 40 and 80 Mm where the length of the shoots was identical at the Bartina cultivar. In the second group, Spunta and Cardinal did not show a significant difference in their length on the control medium and the medium to 40 Mm of NaCl. On the other hand, the Désirée cultivar was classified only with the last group with significant differences in the length of its shoot between the various salts concen-trations.

Concerning the root length, one can divide the four varieties into three groups (Figure 2): plant with the least damaged in the concentrations which arrive at 120 Mm, it gather the cultivar Désirée, and the rate of reduction in growth does not exceed 38%; plants which showed a tolerance relative to 40 and 80 Mm of NaCl, they represent the cultivar Spunta and Cardinal; plants whose rate of reduction in growth reaches 40% with 40 Mm NaCl, they represent the Bartina.

It was observed that a major reduction in the fresh weight of the root system promotionnellement with the increase in the salt concentration (Figure 3). Three groups appeared: the first is consisted of two cultivars Spunta and Desirée which showed a difference in growth between the control medium and the mediums containing a salt concentration. The second group was represented by the Bartina cultivar of which the difference was not significant with the growth of its plants on the control medium and the medium with NaCl 40 Mm on the one hand and between the mediums with 80 and 120 mm of

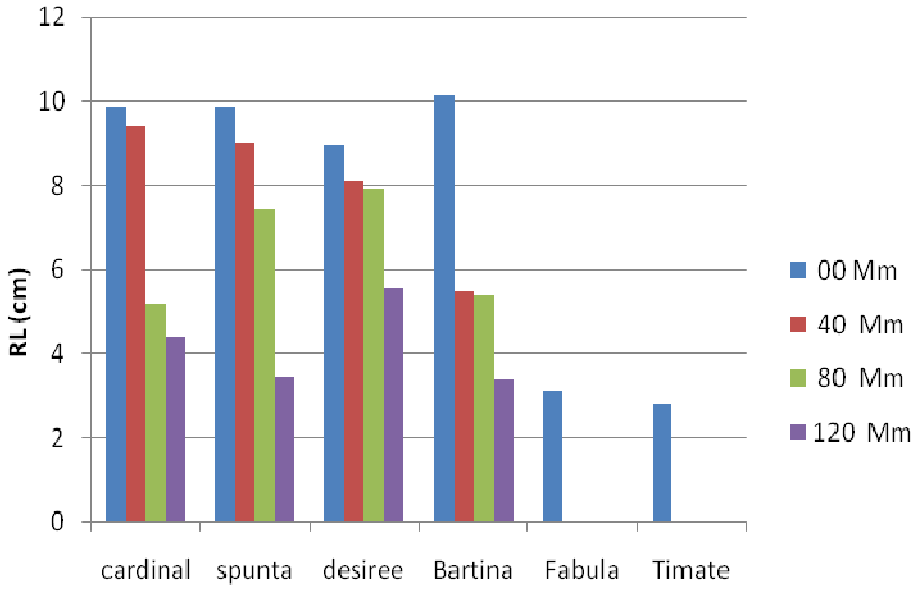

Figure 2. The effect of salinity on *Solanum tuberosum* cultivars' root length.

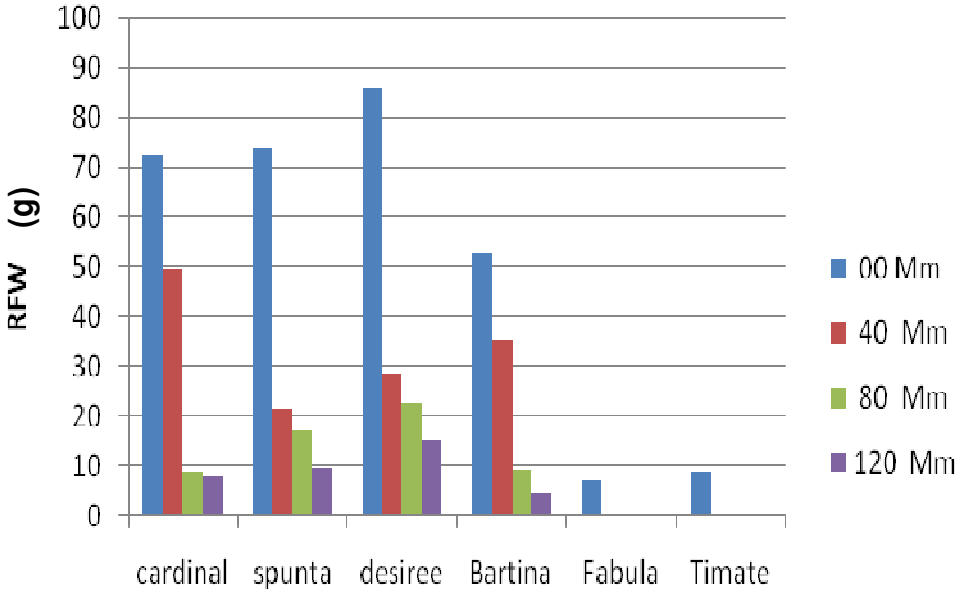

Figure 3. The effect of salinity on *Solanum tuberosum* cultivars' root fresh weight.

NaCl of another hand. In addition, in the group the three all cultivars showed same growth rate in fresh weight on the mediums with 80 and à120 Mm.

Concerning the effect of the salinity of the medium on growth rates in dry weights of the roots, the statistical analyses revealed a significant difference between the mediums at all the varieties except for Spunta (Figure 4).

According to the graph, the growth in root dry weight was proportionally related to the concentration of the medium out of NaCl and the intensity of the reduction in the dry weight according to the NaCl concentration is relatively related to the studied cultivars of potato. To 40 Mm of NaCl, the dry weight was little room of 37% in Cardinal, of 23% at Bartina and 51%, 73% at Désirée and Spunta, respectively. With 80 Mm NaCl the dry weights were reduced with more important rates which arrive at 66 and 79% at Spunta and Desiree, respectively and 89% at Bartina.

The shoot fresh weight was the parameter which shown the least affected by the presence of salt; some is

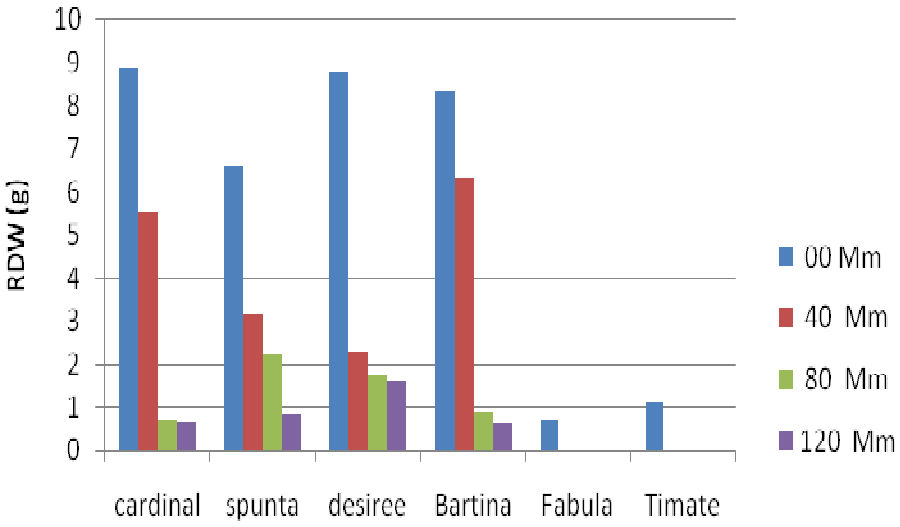

Figure 4. The effect of salinity on *Solanum tuberosum* cultivars' root dry weight.

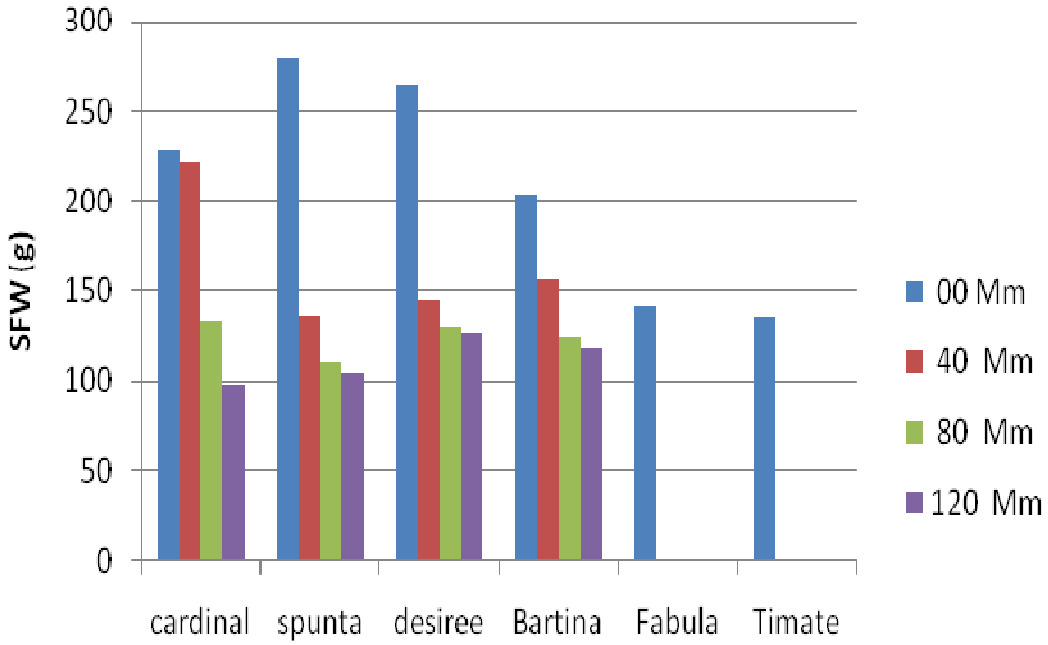

Figure 5. The effect of salinity on *Solanum tuberosum* cultivars' shoot fresh weight.

its concentration except for the Cardinal variety which presented a reduction of 40 and 46% to 80 and 120 Mm, respectively (Figure 5).

The effect of salinity on the dry weight of the shoot was accentuated in Cardinal and Désirée, whereas Spunta and Bartina do not present a significant difference (Figure 6).Three groups arose from the statistical analyses. The first represented by cardinal with a growth identical on the medium to 0 and 40 Mm of NaCl on the one hand and on the mediums with 80 and 120 mm of NaCl of another hand. The second, represented by Spunta and Désirée, showed a significant difference between the control medium and the saline medium. In the last group represented by Bartina an identical growth on all the studied mediums was recorded.

DISCUSSION

The response to salt of the cultivars used in this study was different. Nevertheless, all the cultivars showed reduction in shoot length, reduced root system develop-

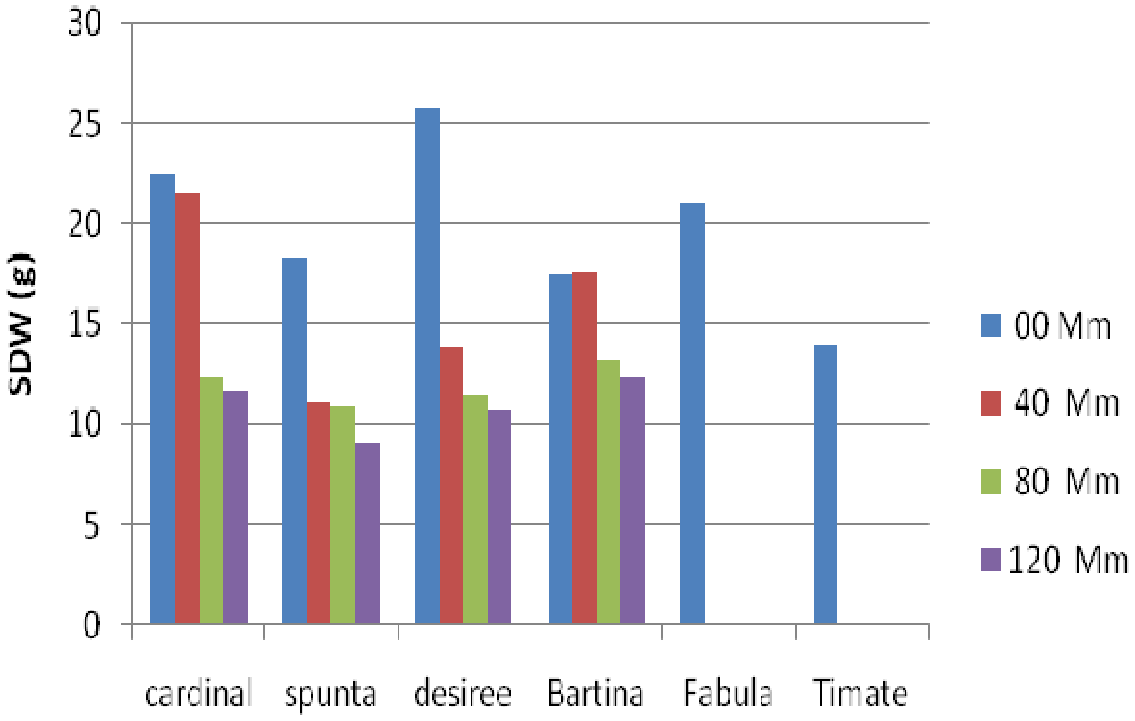

Figure 6. The effect of salinity on *Solanum tuberosum* cultivars' shoot dry weight.

ment due to salinity was also shown under field condition for some cultivars of potato by Levy et al. (1988).

The addition of NaCl to the culture media decreased the osmotic potential of the media inducing salinity stress that adversely affected the plants growth of potato cultivars. Several authors reported the use of NaCl for *in vitro* salinity screening in potato and different plants (Zhang et al., 1993; Pour et al., 2009; Aazami et al., 2010b). Plants growing in the presence of increasing NaCl concentrations decreased their shoot and root length in all potato cultivars (Zhang et al., 1993; Pour et al., 2009). Sasikala and Prazad (1994) and Zhang et al. (1993) reported that shoot and root but not shoot dry weight significantly decreased in the nodal cutting challenged with salt, compared with the growth of the control. With the increase in NaCl, the root fresh and dry weight of all cultivars significantly decreased. The rates of decrease were different depending on cultivars and NaCl levels. The results are in agreement with Levy et al. (1988), Zhang et al. (1993) and Morpurgo and Rogriguez (1987). Farhatullah and Raziuddin (2002) reported that salt stress had the divers' effect on the rooting and root growth in potato. This could be the main reason why all the character under study responded negatively because rooting play a major role in the supply of the nutrient to the plantlets.

Khrais et al. (1998) showed a positive relationship between shoot length and NaCl tolerance in potato. From the results obtained and discussed in this study, Cardinal cultivars showed better resistance in all NaCl treatments and it seems that this cultivar is more salt tolerant than other cultivars studied.

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