



DIFFERENTIAL RESPONSE OF POTATO UNDER SODIUM CHLORIDE STRESS CONDITIONS *IN VITRO*

M H Rahman*, R Islam, M Hossain and S A Haider

Department of Botany, University of Rajshahi, Rajshahi-6205, Bangladesh

Abstract

In vitro evaluations of salinity (NaCl) effects on three potato cultivars (Atlanta, Shepody and Shilbilaty) were investigated with five NaCl levels (0, 25, 50, 75 and 100 mM) by using single node explants. Significant differences were noticed among the cultivars followed by different NaCl levels. Salinity stress gradually depressed plant growth and root development with increased NaCl concentration in MS media. All the cultivars survived at high NaCl (100 mM) containing MS media with exhibiting different growth status. The results indicate that Shilbilaty performed better in shoot length and shoot fresh mass than Shepody and Atlanta. The Atlanta performed better in root growth than Shepody and Shilbilaty at different NaCl media. Highest salinity level drastically inhibits root growth in all the cultivars tested. The control and 25 mM NaCl containing MS media did not affect the growth traits of *in vitro* potato plantlets. The control was found superior in growth characterized than rest of the tested NaCl levels.

Keywords: Potato, single node, NaCl stress, growth response

Introduction

Salinity stress is a critical environmental constraint to crop productivity especially in arid and semiarid regions. The most of the crop plants is intolerable to high salinity conditions resulting decreased yield. Generally, plants are stressed in three ways in saline soils a) low water potential of the root medium leads water deficit, b) the toxic effects of the Na⁺ and Cl⁻ and c) nutrient imbalance by depression in uptake and/or shoot transport (Munns and Termaat 1986, Chapin 1991, Marschner 1995). Toxic accumulation of Na⁺ and Cl⁻ in the leaves has also been correlated with stomatal closure and reduction of total chlorophyll content in leaves both of which limit the amount of photosynthetic production (Romero-Aranda and Syvertsen 1996). Cell and tissue culture techniques together with conventional breeding and genetic engineering have been considered as the potential approaches for the development of plants with increased tolerance to environmental stresses in general and for salt stress in particular. The successful selection of mutant lines from cultured cells and the regeneration of whole plants from such cells have stimulated many attempts for the development of salt-tolerant plants. Potato is considered moderately salt sensitive compared with other crops (Maas and Hoffman 1977). A range of developmental responses in potato is affected by salinity. A small number of potato genotypes has been reported in salinity tolerance under outdoor, greenhouse or *in vitro* conditions. Field trials (Ahmad and Abdullah 1979, Barnes and Peele 1958, Bernstein and Ayers 1951, Bouaziz 1980, Levy 1992, Paliwal and Yadav 1980) and greenhouse pot trials were used to examine genotype salinity tolerance under NaCl or sodium salt irrigation solutions and were based on either tuber yield (Bruns and Caesar 1990, Heuer and Nadler 1995, Nadler and Heuer 1995), relative reduction of foliage dry weight (Bilski *et al.* 1988) or haulm fresh weight (Naik and Widholm 1993). *In vitro* evaluations of NaCl or mixed salt stress effects on potato genotypes were recently proposed as alternatives to the costly, labour-intensive and sometimes problematic field based traits. Salinity stress of potato have been reported by single-node cuttings (Arslan *et al.* 1987, Naik and Widholm 1993, Zhang *et al.* 1993), five-node cuttings

* Corresponding author: Email: hasanur7@yahoo.com

(Morpurgo 1991, Morpurgo and Rodriguez 1987), root tip segment or suspension cultures (Naik and Widholm 1993) *in vitro*. Morpurgo (1991) found a correlation between root fresh weights of *in vitro* plantlets and tuber fresh weights under saline irrigation in the field. Naik and Widholm (1993) found a correlation between *in vitro* root tip segments and haulm fresh weight in greenhouse plants

The objective of this study was to find out the effect of different levels of NaCl on *in vitro* growth traits of three potato cultivars by using single-node cuttings and to evaluate salt-tolerant cultivars effective for future use under salinity conditions.

Materials and Methods

Single-node cutting bioassay: Single node cuttings of potato (*Solanum tuberosum* L.) plantlets derived from meristem culture were subjected to initiate this experiment. Four week grown *in vitro* plantlets of Atlanta, Shepody and Shilbilaty (Bangladeshi Indigenous cultivar) were prepared with shoot apex and root discarded and single nodes (1cm) were separated and cultured in MS medium having 3% sucrose, 0.8% agar with a range of NaCl (0, 25, 50, 75 and 100 mM) treatments. Potato plantlets grown in high NaCl (100 mM) media were recultured in the same concentration of NaCl media and NaCl-free MS (Murashige and Skoog 1962) media to check their growth consistence and their tolerance potential *in vitro*. All the cultures were incubated at 25±2°C with 16/8 h D/N cycle at 15µmol m⁻² s⁻¹ photon flux density (cool white fluorescent light). The pH of the medium was adjusted to 5.8 and autoclaved at 121°C for 20 min. Three nodal segments were implanted in each culture vessel and four replications of each genotype at each salinity level were employed. The experiment ended on 4 weeks and the growth response were noted on shoot length (SL), root length (RL), shoot fresh mass (SF) and root fresh mass (RF). Growth data were evaluated at each salinity level with control (no NaCl trace).

The data were analyzed by IRRISTAT program and the differences among the treatments were tested with least square difference (LSD) at 5% level of significance.

Results and Discussion

Plantlet growth was not affected by 25 mM NaCl containing MS media and generally it was almost similar to control levels whereas 75 and 100 mM NaCl media significantly reduced plantlet growth compared with the control (Table 1). Shoot length, shoot fresh mass, root length and root fresh mass were gradually decreased with the increase of NaCl level in cultivar Atlanta and Shepody and the only exception to this was Shilbilaty (Table 1). At 100 mM of NaCl media, Shilbilaty yielded highest shoot length (2.52 cm) and shoot fresh mass (20.5 mg) than the Atlanta and Shepody while the root length and root fresh mass were much reduced in Shepody. The average root length (13.05 cm) and root fresh mass (45.70 mg) were noted maximum in Atlanta followed by Shepody and Shilbilaty. The control was always superior compared to the treatments employed for all the growth traits and cultivars. The significance level showed that cultivars and treatments were significantly different among each other except shoot fresh mass where the cultivars were not significantly different (Table 1).

Correlation analysis for the relationship of different growth parameters showed that significant correlation existed among each other in response to different NaCl levels. It was noticed that the shoot length decreased with increased root length and root fresh mass. On the other hand shoot fresh mass and root fresh mass increased with the decreased NaCl levels in the media. The increasing root length resulted significantly increased root fresh mass. The mean performances of Shilbilaty resulted best in shoot length and shoot fresh mass over the treatments applied than the Atlanta and Shepody while Atlanta gave highest root length and root fresh mass than the rest of two cultivars tested. It was noticed that the growth consistence remained better in Shilbilaty than Atlanta and Shepody by reculturing in high NaCl (100 Mm) media (data not shown).

Table 1. Shoot length (SL), shoot fresh mass (SF), root length (RL) and root fresh mass (RF) of *in vitro* potato plantlets as affected by NaCl and cultivars after 4 weeks of incubation.

Cultivars	Treatment NaCl (mM)	SL (cm)	SF (mg)	RL(cm)	RF(mg)
Atlanta	25	4.12a	30.80ab	17.33a	78.00a
	50	2.17b	25.3bc	17.75a	31.50b
	75	1.75bc	20.0cd	11.10b	28.75b
	100	0.85c	16.0d	4.05c	7.00c
	Control	5.30a	35.5a	15.03a	83.25a
Mean		2.84	25.5	13.05	45.70
Shepody	25	4.80a	29.8a	12.75a	29.75a
	50	4.95a	26.5a	6.90b	18.83ab
	75	2.20b	29.5a	3.30c	12.15ab
	100	1.50b	9.3b	1.10c	4.30b
	Control	5.17a	29.3a	12.00a	32.00a
Mean		3.72	24.9	7.21	19.41
Shilbilaty	25	3.82b	13.5c	6.25b	11.00b
	50	4.10b	60.0a	11.50a	33.75a
	75	2.35c	28.8b	8.55ab	36.25a
	100	2.52c	20.5bc	2.25c	8.25b
	Control	6.25a	15.3c	10.50a	18.50ab
Mean		3.81	27.6	7.81	21.55
LSD (P=0.05)		1.19	8.40	3.25	19.27
Source of variation		F Value			
Cultivar (C)		8.17**	1.18ns	39.70**	23.39**
Treatment(T)		44.21**	21.01**	43.36**	14.35**
C × T		2.81*	18.33**	6.23**	8.32**
r =0.313 (SLxSF)	r =0.458 (SLxRL)	r =0.416 (SLxRF)	r =0.449 (SFxRL)	r =0.494 (SFxRF)	r =-0.784** (RLxRF)

**= Significant at 1% level, *= Significant at 5% level. Means followed by a common letter are not different at the 5% level by DMRT.

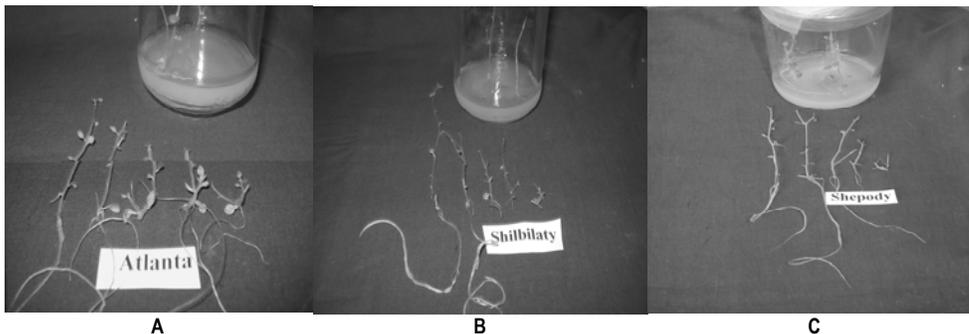


Plate 1. Salinity stress gradually affects the plantlets growth of three potato cultivars: A) Atlanta , B) Shilbilaty and C) Shepody

The present experiment reported show that the single node cuttings bioassay of three potato cultivars responded to different NaCl treatments and their efficiency was evaluated *in vitro*. The growth data were used to predict cultivars tolerance at higher salinity levels *in vitro* and the consistence of their subsequent field performances of salinity tolerance study is still needed. It appears that salinity tolerance of potato genotypes can be successfully evaluated *in vitro* as a promising substitute for conventional field evaluations. The single-node cutting bioassay is simpler to perform than the root tip segment (Yinling and Donnelly 1997). There is merit in evaluation at a range of salinity levels, since different genes are apparently expressed at different stress levels *in vitro* as described *in vivo* (Tal 1994). We recommend taking four growth parameter measurements at 0, 25, 50, 75, and 100 mM NaCl for further micropropagation scheme and consequently it can be used as a source of salt-tolerant line for field transplantation where salinity level is a major obstacle to potato production.

References

- Ahmad R and Abdullah Z U N (1979) Salinity induced changes in the growth and chemical composition of potato. *Pakistan J. Bot.* **11**: 103-112.
- Arslan N, Mix G and Bassam N E I (1987) Ermittlung der Salztoleranz *in vitro* bei Wildarten und Kultursorten der Kartoffel. *Landbauforschung Volkenrode* **37**: 128-131.
- Barnes W C and Peel T C (1958) The effect of various levels of salt in irrigation water on vegetable crops. *Proc. Am. Soc. Hort. Sci.* **72**: 339-342.
- Bernstein L and Ayers A D (1951) Salt tolerance of six varieties of green beans. *Proc. Am. Soc. Hort. Sci.* **57**: 243-248.
- Bilski J J, Nelson D C and Conlon R L (1988) The response of four potato cultivars to chloride salinity, sulphate salinity and calcium in pot experiments. *Am. Potato J.* **65**: 85-90.
- Bouaziz E (1980) Tolerance a la salure de la pomme de terre. *Physiologie Vegetale* **162**: 11-17.
- Bruns S and Caesar K (1990) Sprobenentwicklung und Knollenertrige mehrerer Kartoffelsorten bei hohen Salzkonzentrationen zu verschiedenen Entwicklungsstadien. *Potato Res.* **33**: 23-32.
- Chapin E S (1991) Integrated responses of plants to stress. A centralized system of physiological responses. *Bioscience* **40**: 29-31.
- Heuer B and Nadler A (1995) Growth and development of potatoes under salinity and water deficit. *Aust. J. Agri. ReS.* **46**: 1477-1486.
- Levy D (1992) The response of potatoes (*Solanum tuberosum* L.) to salinity: Plant growth and tuber yields in the arid desert of Israel. *Ann. Appl. Biol.* **120**: 547-555.
- Maas E V and Hoffman G J (1977) Crop Salt Tolerance - Current Assessment. *Journal of the Irrigation and Drainage Division. Proc. Am. Soc. Civil Engr.* **103**: 115-134.
- Marschner H (1995) Saline soils. In: *Mineral Nutrition of higher plants*. Academic Press, New York. pp. 657-680.
- Morpurgo R (1991) Correlation between potato clones grown *in vivo* and *in vitro* under sodium chloride stress conditions. *Plant Breeding* **107**: 80-82.
- Morpurgo R and Silva-Rodriguez D (1987) *In vitro* differential response of the potato (*Solanum tuberosum* L.) under sodium chloride stress conditions. *Rivista di Agricoltura Subtropicale e Tropicale* **81** : 73-77.
- Murashige T and Skoog F (1962) A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Physiologia Plantarum* **15**: 473-497.
- Munns R and Termaat A (1986) Whole plant responses to salinity. *Aust. J. Plant. Physiol.* **13**: 143-160.

- Nadler A and Heuer B (1995) Effect of saline irrigation and water deficit on tuber quality. *Potato Res.* **38**: 19-123.
- Naik P S and Widholm J M (1993) Comparison of tissue culture and whole plant responses to salinity in potato. *Plant Cell, Tissue and Organ Culture* **33**: 273-280.
- Paliwal K V and Yadav B R (1980) Effect of saline irrigation water on the yield of potato. *Indian J. Agricultural Sci.* **50**: 1-33.
- Romero-Aranda R and Syvertsen J P (1996) The influence of foliar applied urea nitrogen and saline solutions on net gas exchange of Citrus leaves. *J Amer. Soc. Hort. Sci.* **121**: 501-506.
- Tal M (1994) *In vitro* selection for salt tolerance in crop plants: Theoretical and practical considerations. *In vitro Cell Developmental Biology* **30**: 175-180.
- Yinling Z and Donnelly D J (1997) *In vitro* bioassay for salinity tolerance screening of Potato. *Potato Res.* **40**: 285-295
- Zhang Y, Brault M, Chalavi V and Donnelly D (1993) *In vitro* Screening for Salinity Tolerant Potato. In: Biometeorology. Proc. 13th International Congress of Biometeorology, September 12-18, 1993. Calgary, AB, Canada. Part **2**, **2**: 491-498.