

## Effect of Genotype, Substrate Combination and Pot Size on Minituber Yield in Potato (*Solanum tuberosum* L.)

<sup>1</sup>H. Vanaei, <sup>2</sup>D. Kahrizi, <sup>1</sup>M. Chaichi, <sup>3</sup>G. Shabani and <sup>4</sup>K. Zarafshani

<sup>1</sup>Agri-Jahad Organization, Hamadan Branch, Hamadan, Iran

<sup>2</sup>Department of Agronomy and Plant Breeding, Razi University, Kermanshah, Iran

<sup>3</sup>Deputy in Planning, State Department, Kermanshah, Iran

<sup>4</sup>Department of Agricultural Extension and Education, Razi University, Kermanshah, Iran

**Abstract:** Potato (*Solanum tuberosum* L.) is one of the major vegetable crops that are grown world-wide because of its economical importance. The main purpose of this study was to investigate the production of virus free plantlets from meristem culture in two cultivars of potato. The specific objectives was to study genotype (2 commercial cultivars of *S. tuberosum*, Marophona and Agria), substrate combination including 3 planting bed soil/sand/perlite (1:1:1), turb/perlite (1:1) and turb/rice hull/perlite (1:1:1), pot size with 19 cm (large) and 13 cm (small) diameters and their interactions on number and total weight of minitubers. A single excised meristem tip was carefully placed on the filter paper bridge of the culture tubes containing MS liquid and then semisolid medium. The virus-free plantlets were transferred to pots with above mentioned substrates and pots. Statistical analysis revealed that there are significant differences between potato cultivars, substrates, pots size and substrate × pot size interaction effects ( $p < 0.01$ ). However, there was no significant difference between other interaction effects on number and total weight of minituber. The Marophona cv. with 9 minituber/pot and 65 gr/plant averages was better than from Agria cv. with 7 minituber/pot and 57 gr/plant averages for number and total weight of minituber, respectively. Turb/perlite in large pots has the highest value for total weight (95gr/plant) and number of minituber (15minituber/plant). Furthermore the finding of this study indicated a positive relationship ( $r = +0.99$ ) between number of minituber and total weight of minituber (per plant).

**Key words:** Genotype % Substrate % Pot size % Minituber % Potato (*Solanum tuberosum* L.)

### INTRODUCTION

The potato (*Solanum tuberosum* L.) is the fourth ranked world crop which has a production of nearly 325 million tons annually. It is the most widely cultivated food crop after wheat, rice and maize. Therefore, it is considered as the most important dicotyledonous and tuber crop [1]. Moreover, potato is an important vegetable and a good source of antioxidants [2]. Potato production is being seriously hampered due to certain viruses, fungus and bacterial diseases. The total loss caused by diseases is estimated between 30 - 100 % during cultivation and in a period of 2 - 6 months of storage [3]. In terms of demand for potatoes, many countries lack an adequate supply of healthy potatoes, a staple crop capable of feeding many people [1].

Potatoes are normally propagated by planting the buds, or “eyes,” present on the tubers. This method of

propagation allows viruses to be transmitted to the new crop each year, resulting in diseases which in turn reduce yields [4-6]. During 1980s, with an advance in plant biotechnology new methods in plant pathology have been developed [6]. For example, micro-propagation of potatoes in laboratories has shown to eliminate virus diseases thus ensuring a virus-free material resulting in increased yields. New techniques such as meristem culture by *in vitro* has been introduced in order to develop virus-free varieties (Fig. 1) [7]. The main advantage of producing minitubers from *in vitro* plantlets is that it allows a faster multiplication rate in seed programs and reduces the number of field generations needed [5]. Moreover, transferring of virus-free plantlets from *in vitro* to *in vivo* and pot conditions are a critical period [7].

In previous reports the effects of plantlet density and soil mixture and on number, size and total weight of

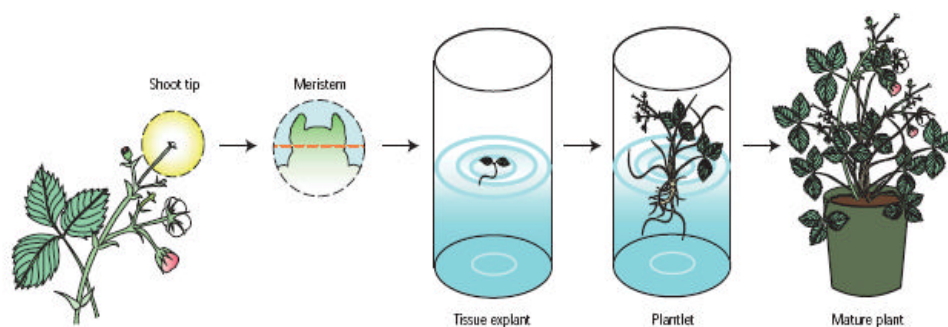


Fig. 1: Schematic representation of meristem culture of plants via tissue culture. Shoot tip meristems (the microscopic growing tips of shoots) can be dissected and grown in tissue culture to produce small plants that can then be transferred to soil and eventually to the field. This method is used extensively to propagate potatoes, strawberries, garlic and many ornamental plants. In addition to multiplying the original plant, this method often is effective in eliminating viruses [7].

minituber have been reported by several researchers [8-10]. This study takes a further step to analyze the effect of genotype, substrate combination and pot size on minituber yield in potatoes.

## MATERIALS AND METHODS

The potato tubers were cultured for supplying of meristem explants in a greenhouse under 16/8 hrs dark/light cycle at 20° C, 80% relative humidity and intensity 60-80  $\mu$  EmG<sup>2</sup> sG<sup>1</sup>. Shoot tips of potato cultivars were collected from 20-25 days old greenhouse grown plants and washed thoroughly under running tap water and then treated with 1.0% savlon and 1-2 drops of tween-80 for about 20 minutes. This was followed by successive three washing with distilled water to make the material free from savlon. Surface sterilization was carried out with 0.1% HgCl<sub>2</sub> for 2 minutes followed by gentle shaking. Furthermore, the sterilized shoot tips were immediately washed several times with sterile distilled water in order to remove all traces of HgCl<sub>2</sub>. After sterilization, the explants were laid on the sterile like using sterile forceps. Shoot tips were kept in one hand under stereomicroscope with a pair of forceps while the immature leaves and primordial leaves were snapped with slight pressure from the needle. Then the exposed meristem tips that appeared as a shiny dome were gently isolated with a sharp blade. A single excised meristem tip was carefully placed on the filter paper bridge of the culture tubes containing MS [11] liquid medium with the growth regulator of KIN and GA<sub>3</sub>. The culture tubes were placed in the growth chamber under 16/8 hrs dark/light cycle at 20±2°C. After 20-25 days the culture tube were transferred to MS semisolid medium with the

growth regulator (KIN+GA<sub>3</sub>) in which after four weeks the plantlets reached the highest of 9-10 cm. The plants were removed from the test tube over a sterile tile using a pair of forceps and transferred to *in vivo* conditions with suitable acclimatization using an experimental design. The experimental design was factorial on basis of completely randomized design (CRD) with three factors and five replications. The factors were potato genotype (2 commercial cultivars of *S. tuberosum*, Marofhona and Agria), substrate combination including 3 planting bed soil/sand/perlite (SSP) (1:1:1), turb/perlite (TP) (1:1) and turb/rice hull/perlite (TRP) (1:1:1) and pot size with 19 cm (large) and 13 cm (small) diameters.

The plants of a potato being initiated for tissue culture were tested in an accredited laboratory for freedom from the following: PVA, PVS, PVM, PVY, PVX, PLRV and PALCV. Tests were carried on a minimum of ten plantlets of each cultivar selected at random. ELISA method was used for virus testing.

Analyses of variance (ANOVA) were carried out in order to determine the effect of above mentioned treatments and the Duncan's multiple range tests was used to compare the mean performance.

## RESULTS AND DISCUSSION

Meristems of potato cultivars were collected from greenhouse grown plants after surface sterilization and it was transferred to tube tests and regenerated. The effects of different treatments on minituber yield of potato are presented in Table 1. Statistical analysis of data showed that there are significant differences between potato genotypes, substrates, pot size and substrate × pot size interaction effects (p<0.01). However, there were no

Table 1: The mean squares analysis of different treatment effects for number and total weight of minituber in potato

Source of variation	Df	No. of minituber (per pot)	total weight of minituber (per pot)
Cultivar (A)	1	16.6**	678.80**
Substrate (B)	2	15.3**	621.3**
Pot size (C)	1	18.7**	795.8**
AB	2	1.6 <sup>ns</sup>	125.6 <sup>ns</sup>
AC	1	1.2 <sup>ns</sup>	122.5 <sup>ns</sup>
BC	2	14.8**	735.9**
ABC	2	1.6 <sup>ns</sup>	107.8 <sup>ns</sup>
Error	48	0.78	60.6
CV%		11.0	13.9

Table 2: Mean performance of potato genotypes for number and total weight of minituber

genotypes	No. of minituber (per plant)	total weight of minituber (per plant)
Marophona	9±0.03 <sup>a</sup>	65±0.26 <sup>a</sup>
Agria	7±0.02 <sup>b</sup>	57±0.23 <sup>b</sup>

Table 3: Mean performance of substrate combination and pot size interaction for number and total weight of potato minituber. Substrate combination including 3 planting bed soil/sand/perlite (SSP) (1:1:1), turb/perlite (TP) (1:1) and turb/rice hull/perlite (TRP) (1:1:1) and pot size with 19 cm (L) and 13 cm (S) diameters

interaction effects	No. of minituber (per plant)	total weight of minituber (per plant)
TP × L	15.0±0.07 <sup>a</sup>	95±0.65 <sup>a</sup>
TP × S	9.0±0.06 <sup>c</sup>	68±0.55 <sup>c</sup>
TRP × L	12.0±0.08 <sup>b</sup>	84±0.66 <sup>b</sup>
TRP × S	9.0±0.02 <sup>c</sup>	66±0.65 <sup>c</sup>
SSP × L	2.0±0.04 <sup>d</sup>	16±0.68 <sup>d</sup>
SSP × S	1.0±0.07 <sup>e</sup>	9±0.65 <sup>e</sup>

observed significant difference between other interaction effects on number and total weight of minituber. This result is in agreement with those obtained by Ahmed Ali *et al.* [12].

When the interaction effects are significant, we are not permitted to carry out mean comparison of main effects [13]. Therefore, mean comparison was conducted for main effect of genotype and substrate × pot size interaction effects.

The mean of number and total weight of minituber for potatoes genotypes are presented in Table 2. The genotype Marophona had the highest frequency for number (9 minituber/plant) and total weight (65 g /plant) of minituber per plant whereas the genotypes Agria had lower performance (7 minitubers/plant and 57 gr/plant

respectively above). This result is not consistent with findings of Rashidi [14].

The means of number and total weight of minituber for substrate combination and pot size interaction effects are presented in Table 3.

As shown in Table 3, turb/perlite in large pots (TP × L) has the highest value for total weight (95 g./plant) and number of minituber (15 minituber/plant). This result is in agreement with the finding of Silva *et al.* [15], but in disagreement with findings of Rashidi [14]. The substrates with soil combination are not suggested for minituber production because our results showed that they have the lowest value for number of minituber (per plant) and total weight of minituber (per plant). The interesting point in Table 2 is the interaction effect for number of minituber (per plant) and total weight of minituber (per plant). The results also indicated a significant positive correlation between number of minituber (per plant) and total weight of minituber (per plant) ( $p < 0.1$ ) ( $r = 0.99$ ).

#### ACKNOWLEDGMENT

The authors would like to thank Dr. Ali Deljoo (Faculty of Agriculture, Buali Sina University, Hamadan, Iran) for his assistance in laboratory trial.

#### REFERENCES

1. World Book, 2000. Potato. In: World Book Millennium 2000, World Book International. hydroponic greenhouse facility utilizing nutrient film technology.
2. Chen, Q., J. Su, S. Nandy and G. Kereliuk, 2007. Screening potato genotypes for antioxidant capacity and total phenolics. Plant Canada Congress.
3. Anonymous. 1992. Potatoes: Improving disease resistance and quality. Biotechnology and Development Monitor. 12: 3-5.
4. Espinoza, N., R. Estrada, P. Tovar, J. Bryan and J.H. Dodds, 1984. Tissue Culture Micropropagation, Conservation and Export of Potato Germplasm. International Potato.
5. Rannali, P., F. Bassi, G. Ruaro, P. Del Re, M. Dicandilo and G. Mandolino, 1994. Microtuber and minituber production and field performance compared with normal tubers. Potato Research, 37: 383-391.

6. Kahrizi, D., A.H. Salmanian, A. Afshari, A. Moieni and A. Mousavi, 2007b. Simultaneous substitution of Gly96 to Ala and Ala183 to Thr in 5-enolpyruvylshikimate-3-phosphate synthase gene of *E. coli* (k12) and transformation of rapeseed (*Brassica napus* L.) in order to make tolerance to glyphosate. *Plant Cell Rep.*, 26: 95-104.
7. Kahrizi, D., A. Arminian and A. Masumi Asl, 2007a. *In vitro* Plant Breeding. Razi University Press (in Persian).
8. Lommen, W.J.M. and P.C. Struik, 1984. Field performance of potato minitubers with different fresh weights and conventional seed tubers: Crop establishment and yield formation. *Potato Research*, 37: 301-313.
9. Gopal, J., J.L. Minocha and H.S. Dhaliwal, 1998. Microtuberization in potato (*Solatum tuberosum* L.). *Plant Cell Reports*, 17: 794-798.
10. Gopal, J., R. Kumar and G.S. Kang, 2002. The effectiveness of using a minituber crop for selection of agronomic characters in potato breeding programmes. *Potato Research*, 45: 145-151.
11. Murashige, T. and F. Skoog, 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Plant Physiology*, 15: 473-497.
12. Ahmed Ali, S., M.M. Alam and V. Souza Machado, 1994. Potato minituber production from nodal cuttings compared to whole *in vitro* plantlets using low volume media in a greenhouse. *Potato Research*, 38: 69-76.
13. Westhof, E., 1999. *Practical Statistics for Experimental Biologists*. 2nd edition by Wardlaw A.C.. John Wiley & Sons, Chichester, pp: 255.
14. Rashidi, T., 2004. The effect of different seedling density which were the result of tissue culture on growth analysis and minituber production in two potato cultivars. MSc Thesis.
15. Silva, E.C., A.B. Giusto, J. Alberto and C.S. Dias, 2006. Production of potato minituber by sprouts grown in several substrate combinations. *Horticultura Brasileira*, 24: 12-18.