Studia Universitatis "Vasile Goldiş", Seria Ştiinţele Vieţii
Vol. 22, issue 4, 2012, pp. 543-547

INFLUENCE OF DIFFERENT POTASSIUM CONCENTRATIONS IN POTATO MICROTUBERIZATION

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ABSTRACT. In this study the effect of potassium nutrition on the induction and development of microtubers by in vitro tuberization was evaluated for two potato genotypes: Christian and Desiree. The objective of this experience was to investigate whether an increasing in potassium concentration would improve in vitro tuberization rate. It was studied the effect of three levels of potassium (10, 25 and 40 (mM/L) on in vitro tuberization. Increasing the potassium supply increased the size but decreased the number of microtubers.

Keywords: *in vitro* tuberization, micropropagation, production, Solarum tuberosum L., tissue culture, minicuttings

INTRODUCTION

In vitro tubers can be alternative propagating materials in a seed potato programme besides in vitro plantlets provided the tuberization rate (i.e. the number of in vitro tubers per plantlet) is satisfactorily high and provided the in vitro tubers can be stored for a long time without considerable loss (Lazányi et al., 1998).

Storability and sprouting characteristics of in vitro tubers could be improved if the size of tubers could be increased (Tábori et al., 1998).

Because of their small size and weight microtubers have tremendous advantages in terms of disease free, storage, transportation and mechanization (Kefi et al., 2000; Kanwal et al., 2006). Consequently, much attention has been focused on the in vitro production of virus-free potatoes (Djurdjina et al., 1997). Many in vitro techniques have been developed during the last decades for producing potato plantlets on nutrient media under aseptic conditions. Meristem culture and propagation of plantlets from nodal cutting techniques are useful in seed tuber production. In vitro propagated plantlets can produce microtubers throughout the year.

Microtubers are produced in vitro on complete plantlets or on plant organs by changing the nutrient medium and/or the external conditions.

Microtubers are more advantageous than microplant in respect of storing, carrying, handling and maintaining crop stand. Microtuberisation of potato is a highly complex developmental process regulated by plant growth regulators and inhibitors.

Potato is usually propagated asexually by means of tubers, that is, the underground stems of the plant. However, with the conventional method of vegetative propagation, potatoes are often prone to many pathogens such as fungi, bacteria and viruses, thereby resulting in poor quality and yields.

Potassium increases the size of tubers and not the number (Trehan et al., 2001, quoted by S. K. Bansal and S.P. Trehan, 2011).

Potassium increases the speed transport of assimilated products (starch translocation from leaves and place them in tubers); under the influence of potassium, increase the number and percentage of large tubers (Ianosi, 2002).

Potassium excess increases the tubers size exaggerated (Ianosi, 2002).

Potassium shows its promoting effect on microtuber number and size, and it is cultivar specific (Naik and Sarker, 1998). Number of microtubers gradually decreased with increasing levels of potassium. The results are similar to those of Naik and Sarker (1998), who reported that microtuber number declined with increasing potassium concentration.

MATERIALS AND METHODS

The experiment was conducted at the tissue culture laboratory of NIRDPSB Brasov, Romania. For production of microtubers in vitro, the potato varieties Christian and Desiree were used.

Tubers of potato cultivar Christian and Desiree were selected and washed with water. For potato tubers shooting, temperature was 180C, on light until the buds reach 2-3 cm long. Shoots were washed in tap water before in vitro culture, than were dipped 3 minutes in 96% ethyl alcohol and then 15 minutes were dipped in a 20 % Domestos solution plus two drops of Tween- 20, followed by 3 washings with double distilled autoclaved water, under sterile air laminar flow hood; sterilization of tubes were made in the drying chamber at 1800C, and the culture medium which had to be introduced into test tubes, was sterilized by autoclaving at 1200C. Growth chamber is equipped with racks, light regime is 4000 lux, the period of 16 hours light and 8 hours dark at a temperature of 200C day and 18 – 200C at night.

Micropropagation involves meristem development phase as a minicutting and then regeneration of this, as a plantlet with leaves, which is divided in minicuttings.

These minicuttings are developing themselves into plantlets (Chiru, 1998). After we had obtained obtained a large number of clones this were tested, by Elisa test and only healthy clones were multiplied. The meristem culture technique for virus elimination is essentially based on the principle that many viruses are unable to infect athe apical/axillary meristems of a growing plant and that a virus- free plant can be produced if a small (0.1-0.3 mm) piece of meristematic tisuue is propagated (Morel and Martin, quoted by Naik and Karihaloo, 2007). Micropropagation is a tissue culture (in vitro)method used for rapid and true to type multiplication of plants on artificial nutrient media under controlled environment (Naik and Karihaloo, 2007).

To get microtubers belonging of these varieties were used minicuttings (figure 1) obtained from plantlets (figure 2), that were inoculated in recipients. Each recipient contains 15 minicuttings. In the first three weeks, the medium used was MS (supplemented with sucrose 20%, 8% agar, NAA 0.5 mg / l), then was added to each recipient 70 ml of ½ MS liquid medium supplemented with 80g / l sucrose, 5 mg / l BA. The recipients was incubated in complete dark at a temperature of 20±1 oC. Three levels of potassium (10, 25 and 40 mM K/l) were used.

After the tuberization period of 8 weeks (figure 3), potato plantlets were removed from culture recipients and microtubers were harvested (figure 4), washed to remove traces of medium and to avoid subsequent infections that may occur during their storage.

Fig. 1 Minicuttings

Fig. 2 Plantlets

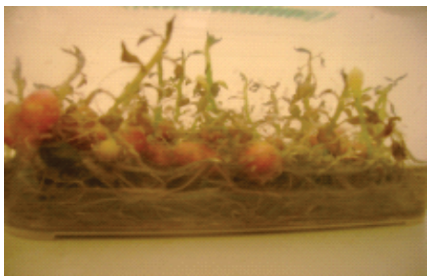

Fig. 3 In vitro tuberization

Fig. 4 Harvested microtubers

Experience was organized according to the protocol of producing microtubers, performed in laboratory, in special culture vessel, grouped into 3 groups (repetitions) (figure 5).

Legend:
- a – variety
- b – concentration level
- r – repetition

	a1			a2		
r1	b1	b2	b3	b1	b2	b3
r2	b1	b2	b3	b1	b2	b3
r3	b1	b2	b3	b1	b2	b3

Fig. 5 Location sketch of experimental variants made for the two varieties and three level of potassium concentration

From figure 5, it appears that the bifactorial experience with 2 experimental variants was performed using the following graduations:

- Experimental factor – a – variety with 2 graduations:
- a_1-Christian (control variety);
- a_2- Desiree.

As control, was fixed variant 1, representative for Romanian varieties.

- Experimental factor – b – tuberization liquid medium with 3 graduations:
- b_1 – tuberization medium with a concentration of potassium of 10 mM/L (control medium);
- b_2 – tuberization medium with a concentration of potassium of 25 mM/L;
- b_3 – tuberization medium with a concentration of potassium of 40 mM/L;

Observations and determinations made: determining the number and average weight of microtubers by weighing individual, depending by variety and potassium concentration used on medium of microtuberization.

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RESULTS AND DISCUSSIONS

Number of microtubers

From point of view of the influence of the variety (Table 1), by comparing with the control variety, Christian obtained a greater number of microtubers/ plantlets (1.24 microtubers/plantlets), compared with Desiree variety, which leads to a distinct significant, negative difference of -0.10 microtubers/ plantlet.

Table 1

Influence of variety on microtubers number

Variety (a)	Average number of microtubers/plantlets		Differences	Significance
	nr.	%		
Christian (Ct)	1.24	100.00	-	-
Desiree	1.14	91.94	-0.10	oo

LSD 5 % a = 0.032 microtub. LSD 1 % a = 0.074 microtub. LSD 0,1 % a = 0.234 microtub

In case of tuberization medium influence (Table 2), the obtained results indicate a negative very significant differences using in tuberization medium potasium with concentration of 25 mM/l and 40mL/L (by - 0.32 microtub. / plantlet and -0.48 microtub. / plantlet).

Table 2

Influence of potassium concentration on microtubers number

Potassium concentration (mM/l)	Average number of microtubers/plantlets	%	Dif. (g)	Signif.
10	1.46	100.00	-	
25	1.14	78.08	-0.32	ooo
40	0.98	67.12	-0.48	ooo

LSD 5 % b = 0.076 microtub. LSD 1 % b = 0.110 microtub. LSD 0,1 % b = 0.165 microtub

Statistical analysis for the combined influence of the varieties and different potassium concentration in microtuberization medium (Table 3) shows that the results obtained by *in vitro* tuberization induction were presented next: for Desiree variety, by comparison with Christian variety the differences were not significant for all the potassium concentrations; between potassium concentrations used in tuberization medium, limit differences obtained are very significant negative for Christian variety, on 40 mM potassium concentration used in tuberization medium, by comparing with 10 mM/L potassium concentration (-0.53 microtub./ plant) and for 25 mM/L by comparing with 10 mM/L potassium concentration (0.33 microtub./ plant); the same situation is for Roclas variety with difference of -0.42 microtub./ plant and 0.29 microtub./ plant for 40 mM potassium concentration and 25 mM/L.

Table 3

Influence of variety and potassium concentration over microtubers number

Variety/ Potassium concentration (mM/l)	10		25		40		Differences		
	no.	Dif. Signif.	No.	Dif. Signif.	No.	Dif. Signif.	b_2-b_1	b_3-b_1	b_3-b_2
Christian	1.53	-	1.20	-	1.00	-	-0.33 ooo	-0.53 ooo	-0.20 oo
Desiree	1.38	-0.15 ns	1.09	-0.11 ns	0.96	-0.04 ns	-0.29 ooo	-0.42 ooo	-0.13 o

LSD 5 % a*b = 0.153 microtub
LSD 1 % a*b = 0.307
LSD 0,1 % a*b = 0.848

LSD 5 % b*a = 0.098 microtub
LSD 1 % b*a = 0.143 microtub
LSD 0,1 % b*a = 0.214 microtub

Microtubers weight

In the case of bifactorial statistical analysis (table 4), we can say that regarding the influence of microtubers weight, Desiree cultivar recorded a not significant difference, statistically assured by -0.04g, compared with Christian variety.

Table 4

Influence of variety on microtubers weight

Variety (a)	Average weight of a microtuber		Differences	Significance
	g	%		
Christian (Ct)	0.68	100.00	-	-
Desiree	0.64	94.12	-0.04	ns

LSD 5 % a = 0.080 g LSD 1 % a = 0.184 g LSD 0,1 % a = 0.586 g

Table 5

Influence of potassium concentration on microtubers weight

Potassium concentration (mM/l)	Average weight of a microtuber (g)	%	Dif. (g)	Signif.
10	0.46	100.00	-	
25	0.65	141.30	0.19	***
40	0.87	189.913	0.41	***

LSD 5 % b = 0.071 g LSD 1 % b = 0.103 g LSD 0,1 % b = 0.154 g

About potassium concentration influence (taking as control 10 mM/l potassium concentration), we may say that concentrations of 25 and 40 mM/L were very significantly influenced in the positive way the weight of microtubers obtained in the experience (Table 5), with differences by 0.19 g and 0.41 g.

Table 6

Influence of variety and potassium concentration over microtubers weight

Variety/ Potassium concentration (mM/l)	10		25		40		Differences		
	g	Dif. Signif.	g	Dif. Signif.	g	Dif. Signif.	b_2-b_1	b_3-b_1	b_3-b_2
Christian	0.48	-	0.68	-	0.88	-	0.2 **	0.40 ***	0.20 **
Desiree	0.44	-0.04 oo	0.62	-0.06 oo	0.85	-0.03 o	0.18 **	0.41 ***	0.23 ***

LSD 5 % a*b = 0.0016 g
LSD 1 % a*b = 0.0399 g
LSD 0,1 % a*b = 0.1093 g

LSD 5 % b*a = 0.0998 g
LSD 1 % b*a = 0.1452 g
LSD 0,1 % b*a = 0.2178 g

Examining the results through the combined influence of varieties and potassium concentration used in microtuberization medium was seen that in case of 10 mM/L and 25 mM/L potassium concentrations, for Desiree variety, comparing with Christian variety, distinctly significant, negative differences were obtained (respectively -0604 g and -0.06g). For the same variety, but for potassium concentration of 40 mM/L, was obtained a difference significant of -0.03 g, comparing with the control variety. From this table it can be observed that the higher value of microtuberts weight was obtained for Christian variety, when it was used in microtuberization medium potassium convcentration of 40 mM/L. Examining the differences obtained between the three concentrations of potassium are found weight values bigger for potassium concentration of 40 mM/L by comparing with concentration of 10 mM/L, for both cultivars, with negative differences of 0.40 g and 0.41 g.

CONCLUSIONS

In the present experience was demonstrated that a higher concentration of potassium in microtuberization medium determined obtaining greater microtubers. Microtuber number decreased with increasing of potassium concentration.

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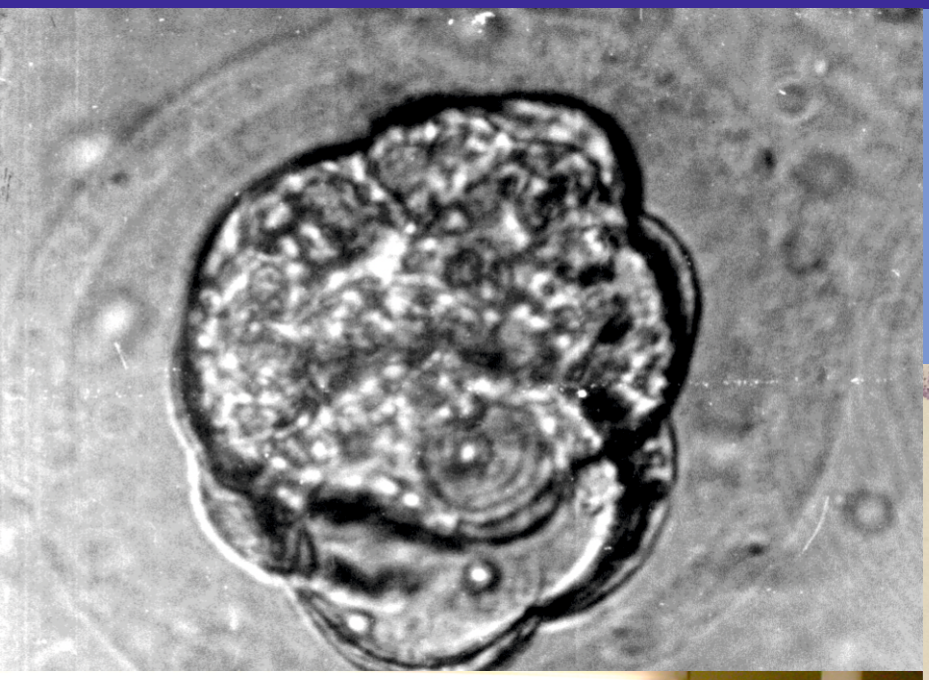
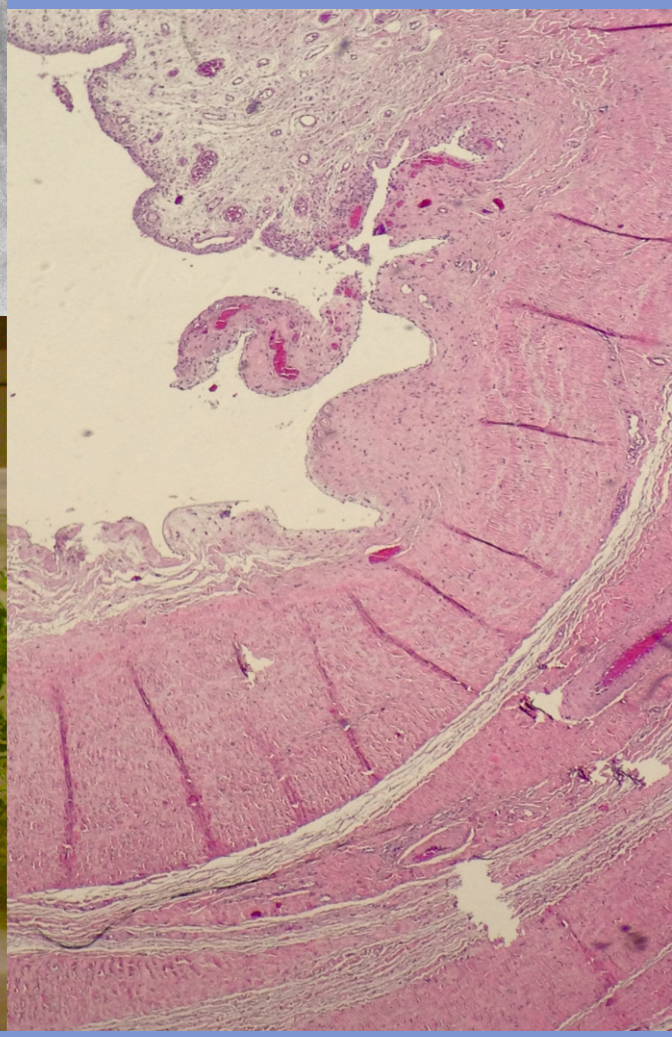


SERIA ȘTIINȚELE VIEȚII
(LIFE SCIENCES SERIES)
vol. 22, issue 4
Oct.-Dec. 2012



"VASILE GOLDIȘ" UNIVERSITY PRESS
ARAD 2012

ISSN 1584-2363
e-ISSN 1842-7863
ISSN-L 1584-2363
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