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RESULTS OF SUGAR ALCOHOLS INFLUENCE OVER DIFFERENT ROMANIAN POTATO VARIETIES

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Abstract

Plantlets of three cultivars Sarmis, Christian and Roclas were induced to microtuberized under dark conditions and at temperature of 17°C. In medium of tuberisation were applied two different sugar alcohols (sorbitol ans mannitol) for evaluate the influence of this under the number of microtubers obtained/plantlet and the average weight of a microtuber. It was used three concentrations of sugar alcohols (0.05; 0.11; 0.17 mol/l) which were compared with controlled medium in which was not added any type sugar alcohol.

Key words*: potato, plantlets, microtubers, hydric stress, manitol, sorbitol.*

INTRODUCTION

Increasing crop production in drought environment may be achieved through breeding crops that are more tolerant to drought (Rao S. and FTZ J., 2013). According to one of the stress concepts, stress is defined as an environmental factor, which can be potentially unfavorable to living organisms (Levitt, J., 1980, quote by Hassanpanah, 2009). Fresh water resources are limited and their use in agricultural production is expected to come under increasing constraints (Albiski K. et al., 2012). How plants cope with drought stress is a topic of an intense debate (Kacem N. S et al., 2017). Biotechnology like tissue culture technology offers rapid alternative in crop improvement. In recent years, tissue culture based *in vitro* selection has emerged as a feasible and cost-effective tool for developing stress-tolerant plants (Rao S. and FTZ J., 2013).

Potato is highly amenable to tissue culture (Espinoza et al., 1986, quote by Gopal J. and Iwama K., 2007) and micropropagation and microtuberization have become established methods of rapidly multiplying cultivars for seed production as well as for germplasm conservation and exchange (Roca et al., 1979; Ranalli et al., 1994; Gopal et al., 1998, 2002, 2005; Donnelly et al., 2003, quote by Gopal J. and Iwama K., 2007). *Solanum tuberosum* L. is sensitive to drought due to its shallow root system (Iwama and Yamaguchi, 2006, quote by Bundig C. et al., 2016). Mannitol or sorbitol have been used by several workers as osmotic stress agents for *in vitro* selection (Hassan N.M. et al., 2004; Mohamed M.A.H., 2000). A polyol is an alcohol containing multiple hydroxyl groups. Sugar alcohols include: sorbitol, glycerol, erythritol, maltitol, isomalt, mannitol, lactitol, threitol, arabitol, ribitol and xylitol (Acton A.Q., 2013).

MATERIALS AND METHODS

The starting point for obtaining a free material of potato viruses is the culture of meristems. The meristem is inoculated on test tubes with medium Murashige and Skoog (MS), 1962. After 6-8 months, after more subculture, in function of genotypes from meristems plantlets are developing. To evaluate the phytosanitary quality of the plantlets, ELISA test was made. The infected clones are eliminated. Biological material free of virus is *in vitro* multiplied. While the temperature of 20 ± 20°C is the necessary for plants micropropagation, the temperature required to obtain the microtubers is generally lower (17°C). Sucrose is the most decisive factor for *in vitro* tuber formation. Sucrose is a source of energy and at higher concentrations, is favoring the formation of microtubers. For the production of microtubers, sucrose concentrations are increased from 2% used for plant micropropagation to 8%. On culture recipients with developed plantlets (Figure 1) is put a liquid medium for

microtubersitation. The recipients are kept in dark conditions and after 3 month the microtubers (Figure 2) are harvested.
To studying the effect of sugar alcohol over microtuberisation 6 variants were analyzed in a bifactorial experience, 3 x 2, in 3 repetitions. The graduations of the studied factors were: experimental factor B, the variety, with three graduations: a_1 – Sarmis, a_2 – Christian, a_3 – Roclas; Experimental factor B, sugar alcohol, with two graduations: b_1 – sorbitol, b_2 – mannitol.
Microtuber production is an important rapid multiplication method for prebase stock formation as well as germplasm exchange.

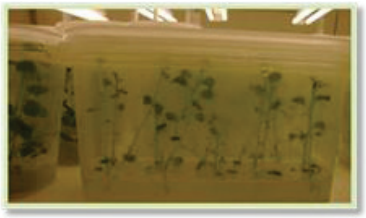

Figure 1. Developed plantlets

Figure 2. Microtubers

RESULTS AND DISCUSSIONS

From the Table 1 we can observe that mannitol determined obtaining a higher number of microtubers/ plantlets (1.07), even if the difference between the two agents is not significant.
From the Table 1 we can see that with the increase in concentration of agent water stress inducing, the number of microtubers/plantlets decreased.
At concentration of 0.05 mol/l sugar alcohol (Table 2), the difference is distinctly significant (-0.23 microtubers/pl), negative, to control, statistically assured. For the other concentrations (0.11; 0.17 mol/l) very significant, negative differences are obtained (-0.33; -0.49 microtubers/pl).
From the analysis of the average values of the number of microtubers/plantlet/variety (Table 3), it is observed that the differences are small, not significant, by 0.16 and 0.17 statistically assured (for Christian and Roclas), distinctly significant, positive.
The statistical interpretation of the combined influence of the two factors (Table 4), respectively the variety and the inducer of hydric stress *in vitro* shows that sorbitol concentrations of 0.05 and 0.011 mol/l present distinctly significant differences (-0.305 and -0.358), negative. Sorbitol concentration of 0.17 presents a very significant difference -0.604. When it is compared the two inductors of hydric stress, mannitol presents better results for 0.17 mol/l concentration with a positive difference, 0.23. Sorbitol presented a stronger osmotic pressure, causing a lower number of microtubers.
Another parameter studied was average weight of a microtuber.
Statistical analysis of the influence of the variety on weight (Table 5) of a microtuber shows that the difference (0.02 g) for mannitol comparative with sorbitol is not significant.
The statistical analysis of the influence of the concentrations of sugar alcohols shows us that 0.05, 0.11 and 0.17 mol/l determined an average weight of a microtuber with very significant (Table 6), negative differences (-0.17, -0.23, -0.28 g).
From the statistical analysis of the influence of the variety and of the sugar alcohols (Table 8) we may have observed that on concentration of 0,05 mol/l the differences are distinctly significant for both sugar alcohols, but negative (-0.17 g for sorbitol and -0.18 for mannitol) comparative with control (nutritive medium to which was not added sugar alcohols).
To next concentrations 0.11 and 0.17 mol/l the differences are very significant for both sugar alcohols, negative, statistically assured (-0.22 g and -0.26 g for sorbitol and -0.25 and -0.31 g for mannitol). It can be seen with increasing of sugar alcohols concentration in nutritive medium, this has as effect decreasing the average weight of a microtubers/plantlet. Regarding the differences between the two

sugar alcohols used with different concentrations, it can be noticed that there are no significant differences for the weight of a microtuber. The average weight of a microtuber is higher when mannitol it is used, so sorbitol has a higher osmotic potential.

Table 1. Influence of sugar alcohols on the average number of microtubers obtained/plantlet

Nutritive medium Murashige Skoog supplemented with sugar alcohols	Average number of microtubers obtained/plantlet		Dif.	Sign.
	Nr.	%		
sorbitol (Ct)	0.96	100.00	-	-
mannitol	1.07	111.58	0.11	ns

Table 2. Influence of sugar alcohols concentrations on the average number of microtubers obtained/plantlet

Concentrations of sugar alcohols (mol/l)	Average number of microtubers obtained/plantlet		Dif.	Sign.
	Nr.	%		
0.00 (Ct)	1.27	100.00	-	-
0.05	1.04	81.92	-0.23	oo
0.11	0.95	74.36	-0.33	ooo
0.17	0.79	61.62	-0.49	ooo

DL 5% = 0.13 DL 1% = 0.18 DL 0.1% = 0.26

Table 3. Influence of variety on the average number of microtubers obtained/plantlet

Variety	Average number of microtubers obtained/plantlet		Dif.	Sign.
	Nr.	%		
Sarmis (Ct)	0.90	100.00	-	-
Christian	1.06	117.55	0.16	Ns
Roclas	1.07	118.95	0.17	Ns

DL 5% = 0.59 DL 1% = 0.79 DL 0.1% = 1.05

Table 4. Combined influence of sugar alcohols and their concentrations on the average number of microtubers obtained/plantlet

Concentrations of sugar alcohols (mol/l)	Sorbitol		Dif.	Sign.	Mannitol		Dif.	Sign.	a2-a1	Sign.
	Nr.	%			Nr.	%			Nr.	
0.00 (Ct)	1.274	100.00	-	-	1.274	100.00	-	-	0.000	ns
0.05	0.969	76.05	-0.305	oo	1.119	87.80	-0.155	ns	0.150	ns
0.11	0.916	71.88	-0.358	oo	0.979	76.85	-0.295	oo	0.063	ns
0.17	0.670	52.58	-0.604	ooo	0.900	70.66	-0.374	ooo	0.230	*

DL 5% = 0.182
DL 1% = 0.256
DL 0.1% = 0.361

DL 5% =0.229
DL 1% = 0.405
DL 0.1% =0.940

Table 5. Influence of sugar alcohols on the average weight of a microtuber

Nutritive medium Murashige Skoog supplemented with sugar alcohols	Average weightof a microtuber		Dif.	Sign.
	(g)	%		
sorbitol (Ct)	0.25	100.00	-	-
mannitol	0.27	110.00	0.02	ns

DL 5% = 0.14 g DL 1% = 0.33 g DL 0.1% = 1.04 g

Table 6. Influence of sugar alcohols concentrations on the average weight of a microtuber

Concentrations of sugar alcohols (mol/l)	Average weight of a microtuber		Dif.	Sign.
	(g)	%		
0.00 (Ct)	0.43	100.00	-	-
0.05	0.26	59.59	-0.17	Ooo
0.11	0.20	45.64	-0.23	Ooo
0.17	0.15	34.63	-0.28	Ooo

DL 5% = 0.07 g, DL 1% = 0.09 g, DL 0.1% = 0.13 g

Statistical analysis of variety influence indicates differences not significant between varieties (Table 7).

Table 7. Influence of variety on the average weight of a microtuber

Variety	Average number of microtubers obtained/plantlet		Dif.	Sign.
	(g)	%		
Sarmis (Ct)	0.29	100.00	-	
Christian	0.26	88.11	-0.03	Ns
Roclas	0.23	78.32	-0.06	Ns

DL 5% = 0.59 g, DL 1% = 0.79g, DL 0.1% = 1.05g

Table 8. Combined influence of sugar alcohols and their concentrations on the average weight of a microtuber

Concentrations of sugar alcohols (mol/l)	Sorbitol		Dif.	Sign.	Mannitol		Dif.	Sign.	a2-a1	Sign.
	(g)	%			g	%			(g).	
0.00 (Ct)	0.43	100.00	-		0.43	100.00	-	-	0.000	ns
0.05	0.25	61.58	-0.17	oo	0.27	57.61	-0.18	oo	0.017	ns
0.11	0.18	49.15	-0.22	ooo	0.21	42.12	-0.25	ooo	0.030	ns
0.17	0.12	40.55	-0.26	ooo	0.17	28.70	-0.31	ooo	0.051	ns

DL 5% = 0.10 g
DL 1% = 0.14 g
DL 0.1% = 0.19 g

DL 5% = 0.16 g
DL 1% = 0.30 g
DL 0.1% = 0.79 g

CONCLUSIONS

Medium with different concentrations in which was added sugar alcohols very significantly reduced the number of microtubers/plantlets and weight of a microtuber compared with the medium to with no these osmotic agents (with 0.00 mol /l sugar alcohols).

Even though there are no significant differences for numbers/plantlets and weight of microtubers between the two sugars, lower values are obtained to sorbitol (0.96 microtubers/pl and 0.25 g) meaning that this is an inducer of *in vitro* drought more powerful than mannitol.

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