International Journal of Advanced Research in Biological Sciences
ISSN: 2348-8069
www.ijarbs.com
DOI: 10.22192/ijarbs Coden: IJARQG(USA) Volume 5, Issue 1 - 2018

Research Article

DOI: http://dx.doi.org/10.22192/ijarbs.2018.05.01.015

Effect of osmotic inhibitors on reducing of growth rate and the preservation of viability of sterile cultures

Elena Kutas[1]*, Aleksandr Veyevnik[1], Vladimir Titok[1], Lyubov Ogorodnyk[2]
[1]Central Botanical Garden of the NAS of Belarus, 220072 Minsk, Surganova, 2v, Republic of Belarus.
E-mail: *vinogradova-kira@tut.by*
[2]Kyiv Taras Shevchenko University, 01601 MSP Kyiv, Volodymyrskastreet, 64, Ukraine.

Abstract

The paper presents the results of experimental studies on the effect of different concentrations of mannitol and sorbitol on activation the meristem, the growth rate and viability of regenerants of sterile cultures of 6 varieties of highbush blueberry and 3 varieties of cranberry ordinary. It was shown, that activation of meristems, the growth rate and viability of regenerants of investigated varieties depend on osmotic inhibitors, contained in the nutrient medium, their concentrations and variety affiliation of plants.

Keywords: mannitol, sorbitol, highbush blueberry, cranberry ordinary, viability, aseptic culture.

Introduction

In world practice for many years to maintain a collection of sterile cultures there were used a constant growing of them under optimal conditions for these plants. However, it turned out that such cultivation has some significant disadvantages: firstly, the possibility of appearance of somaclonal variation (due to violations of the genetic stability of the frequent transplanted material); secondly, the risk of contamination by alien genetic material and random loss of its own genetic aterial; in - third, the complexity of the processes (the necessity of regular transplantation on fresh medium); in -fourth, the high cost of nutrient medium components, required for frequent transfers.

You can achieve slowing of growth of the culture by several ways:

1) a decrease in cultivation temperature (Mullin and Schelegel, 1976; Coman and Neculae, 1981; Westcott, 1981a; Demenko and Trushechkin, 1988; Mohammed, 1998; Winthers, 1989; Dorion et al., 1994; Hae-Boong et al., 1996; Samarina et al., 2014; Ali et al., 2016);
2) changes in carbohydrate concentration in the medium (Samsonova, 1991; Vysotskaya, 1994; Homes et al., 1982; Rubluo and Kartha, 1985; Rolland et al., 2006);
3) induction of osmotic stress (Tukai et al., 1988; Codaccioni and Vescovi, 1987; Schilde-Renther et al., 1982; Wanas et al., 1986; Pua and Chong, 1985);
4) the use of retardants (Westcott, 1981b; Cathey, 1964; Zadontsev et al., 1973; Chailakhyan, 1967; Kende et al., 1963);
5) a decrease in atmospheric pressure in the culture vessels (Muromtsev et al., 1990; Winthers, 1979; Brindgen and Staby, 1981, 1983; Dorion et al., 1994).

The main obstacle in storage of the plant material of cowberry and blueberry is its rapid aging, leading, ultimately, to the death of the plant. To prevent this process, you should regularly, every 2-3 weeks, transplant regenerants on fresh nutrient media.

In our opinion, one way of solving of this problem is a slowing of growth with the help of osmotic inhibitors.

As the review of the literature on the subject, the information concerning the effect of osmotic inhibitors on the viability of regenerants in culture in vitro is scanty (Vysotskaya, 1994; Salmatova et al., 2009; San José et al., 2010; Rahmanet al., 2010; Ciobaniui and Constantinovici, 2012) and for plans of highbush blueberry and cranberry is absent no at all.

The aim of research was to study the effect of different concentrations of mannitol and sorbitol on the viability of introduced varieties of highbush blueberry and cranberry in culture *in vitro*.

Materials and Methods

As objects of study there were used 6 varieties of highbush blueberry ('Bluecrop', 'Blueray', 'Dixi', 'Herbert', 'Rancocas', 'Scammel') and 3 varieties of cranberries ordinary ('Koralle', 'Masovia', 'Erntedank'), regenerated in culture in vitro by method of activation of axillary meristems. Experiments were carried out with mannitol and sorbitol in a concentration range of 1-3 $g \cdot L^{-1}$ on nutrient medium WPM (Lloyd and McCown, 1981), containing 4 mg $\cdot L^{-1}$indoleacetic acid and 15 mg $\cdot L^{-1}$izopenteniladenina at 16 h photoperiod, illuminance 4000 lux, a temperature of 25^0C.

During the experiments, the plants were cultivated without tansplantation on fresh nutrient medium for 12 months.* The viability of the material was evaluated by our method, comprising a five-point scale, according to which: 0 points - the death of plants, 1, 2, 3, - an intermediate state, 4 - maximum viability, and also we took into account the rate of activation of meristems and shoot growth. Findings are taken after 2, 4 and 12 months of cultivation. Experimental data are shown in Table. 1 - 3. The figures in the tables are the average values for the 10 regenerants of each sort.

* The term "viability" is used by us as a concept characterizing the potential duration of the life of plants under cultivation without renovation of nutrient media.

Results and Discussion

Figures in Table 1 indicate that the maximum activation of meristems was observed in meristems for all sorts without exception on the medium № 1, containing 1 $g \cdot L^{-1}$mannitol, during the first 2 months of cultivation, and was 50,6 – 80,1%. The viability of regenerants approached to the highest evaluation 4 numbers on the medium № 3, containing 2 $g \cdot L^{-1}$ of mannitol.

After 4 months of cultivation there were observed the maximum length of the shoots on the medium №1 for all varieties of highbush blueberry (0,39-0,53cm) and cranberries ordinary (1,30-1,41 cm). In this case their viability decreased to 2,89-3,23 points. In our opinion, the decrease of the viability of regenerants is not accidental, since the end of their growth is accompanied by the aging process.

For allvarieties was marked good viability, equal 3,41-3,84 numbers on the medium № 3, containing 2 $g \cdot L^{-1}$ of mannitol, despite their prolonged cultivation (4 months) without transfers. This concentration of mannitol in the medium promoted to reducing the growth rate, there by preventing the aging process. One might think that mannitol as osmotic inhibitor has an inhibitory effect on the flow of water and plastic substances - constitute an integral and complex process, what is the growth of regenerants.

An analysis of the experimental data, shown in Table 2, gives reason to believe that the medium № 3, containing 2 $g \cdot L^{-1}$ of sorbitol, is optimal for preserving of the viability of most varieties of highbush blueberry ('Blueray', 'Herbert', 'Rancocas', 'Scammel') and one variety of cranberry ordinary ('Erntedank') during their long cultivation (4 months) without transfers. For other varieties ('Koralle', 'Masovia', 'Dixi', 'Bluecrop') should be considered as the optimal medium № 1 containing 1 $g \cdot L^{-1}$ sorbitol.

Thus, the addition into the nutrient medium 2 $g \cdot L^{-1}$mannitol, or 1- 2 $g \cdot L^{-1}$ sorbitol promotes to reducing of growth rate of regenerants, slowing the aging process, the preservation of viability that provides an opportunity to increase the interval between transfers of up to 4 months instead of 2-4 weeks.

T a b l e 1. The effect of different concentrations of mannitol on the viability of regenerants of *Vaccinium corymbosum* L. and *Vaccinium vitis- idaea*L. in culture *in vitro*

Variety	Two months of cultivations					
	medium № 1		medium № 2		medium № 3	
	A	**V**	**A**	**V**	**A**	**V**
1	2	3	4	5	6	7
Vaccinium corymbosum						
'Bluecrop'	70,1	3,24±0,39	40,4	3,54±0,29	29,4	3,76±0,17
'Blueray'	64,4	3,47±0,64	39,3	3,49±0,39	30,7	3,54±0,71
'Dixi'	67,1	3,39±0,17	36,1	3,43±0,41	31,3	3,51±0,63
'Herbert'	69,2	3,38±0,60	31,3	3,46±0,58	30,2	3,65±0,45
'Rancocas'	58,5	3,48±0,64	29,9	3,49±0,43	37,7	3,53±0,47
'Scammel'	50,6	3,29±0,73	27,1	3,56±0,81	24,1	3,76±0,29
Vaccinium vitis-idaea						
1	2	3	4	5	6	7
'Koralle'	70,9	3,23±0,24	50,4	3,81±0,39	50,7	3,89±0,19
'Erntedank'	76,4	3,76±0,29	53,9	3,74±0,26	51,3	3,81±0,39
'Masovia'	80,1	3,71±0,31	53,9	3,74±0,39	51,3	3,87±0,45

Variety	Four months of cultivation					
	medium № 1		medium № 2		medium № 3	
	Length of shoots on 1 plant, cm	V	length of shoots on 1 plant, cm	V	length of shoots on 1 plant, cm	V
Vaccinium corymbosum						
'Bluecrop'	0,40±0,13	2,89±0,43	0,41±0,15	3,18±0,67	0,39±0,13	3,75±0,29
'Blueray'	0,53±0,26	3,01±0,87	0,51±0,19	3,24±0,43	0,29±0,11	3,49±0,34
'Dixi'	0,39±0,24	3,09±0,94	0,44±0,17	3,31±0,46	0,35±0,15	3,47±0,71
'Herbert'	0,46±0,17	2,97±0,53	0,48±0,13	3,29±0,37	0,41±0,17	3,61±0,60
'Rancocas	0,49±0,41	3,17±0,46	0,54±0,21	3,18±0,60	0,47±0,17	3,41±0,45
'Scammel'	0,43±0,20	2,98±0,60	0,51±0,29	3,36±0,58	0,48±0,21	3,59±0,58
Vaccinium vitis-idaea						
'Koralle'	1,30±0,78	3,23±0,63	0,75±0,39	3,60±0,45	0,80±0,60	3,84±0,73
'Erntedank'	1,41±0,67	3,41±0,45	0,89±0,46	3,27±0,71	0,29±0,74	3,76±0,67
'Masovia'	1,37±0,53	3,35±0,24	0,74±0,49	3,44±0,41	0,77±0,29	3,81±0,24

Notes: A - activation of axillary and apical meristems in percentages; V – viability, points; medium № 1 - WPM + 1 g $\cdot L^{-1}$mannitol; medium № 2 - WPM + 3 g $\cdot L^{-1}$mannitol; medium № 3 - WPM + 2 g $\cdot L^{-1}$mannitol; WPM - the main medium for clonal micropropagation of highbush blueberry and cranberry ordinary : macro- and micronutrients in WPM , mesoinositol - 100 mg $\cdot L^{-1}$, adenine sulfate - 80 mg $\cdot L^{-1}$, B_1 - 1 mg $\cdot L^{-1}$, B_6 – 0,5 mg $\cdot L^{-1}$, glycine - 2 mg $\cdot L^{-1}$, IAA - 4 mg $\cdot L^{-1}$, 2iP - 15 mg $\cdot L^{-1}$.

T a b l e 2. The effect of different concentrations of sorbitol on the viability of regenerants of *Vaccinium corymbosum* L. and *Vaccinium vitis-idaea* L. in culture *in vitro*

Variety	Two months of cultivation					
	medium № 1		medium № 2		medium № 3	
	A	V	A	V	A	V
Vaccinium corymbosum						
'Bluecrop'	64,5	3,01±0,21	56,5	2,84±0,51	57,1	2,93±0,36
'Blueray'	69,3	2,99±0,34	61,3	2,73±0,60	64,3	2,81±0,14
'Dixi'	58,4	3,16±041	52,8	2,99±0,16	57,1	3,01±0,13
'Herbert'	66,7	3,38±0,56	60,7	3,01±0,22	61,4	3,15±0,12
'Rancocas'	54,2	3,27±0,18	50,3	3,03±0,46	51,1	3,21±0,17
'Scammel'	62,3	3,03±0,60	58,4	2,89±0,39	59,4	2,91±0,09
Vaccinium vitis-idaea						
'Koralle'	65,9	3,25±0,25	62,9	2,79±0,35	63,4	2,84±0,59
'Erntedank'	70,4	3,41±0,19	61,7	3,01±0,61	65,2	3,21±0,29
'Masovia'	67,3	3,34±0,20	59,4	2,99±0,24	64,4	3,15±0,11

Variety	Four months of cultivation					
	medium № 1		medium № 2		medium № 3	
	length of shoots on 1 plant, cm	V	length of shoots on 1 plant, cm	V	length of shoots on 1 plant, cm	V
Vaccinium corymbosum						
'Bluecrop'	0,31±0,21	2,44±0,29	0,22±0,07	2,01±0,18	0,29±0,11	2,31±0,14
'Blueray'	0,44±0,07	2,21±0,16	0,39±0,16	2,14±0, 27	0,41±0,17	2,25±0,19
'Dixi'	0,32±0,09	2,36±0,31	0,28±0,19	1,99±0,19	0,29±0,09	2,14±0,22
'Herbert'	0,37±0,13	2,15±0,32	0,32±0,11	2,44±0,29	0,34±0,19	2,61±0,17
'Rancocas'	0,41±0,21	2,24±0,60	0,37±0,14	2,36±0,17	0,39±0,12	2,53±0,31
'Scammel'	0,35±0,16	2,09±0,45	*0,33±0,14*	2,17±0,07	0,33±0,16	2,35±0,24
Vaccinium vitis-idaea						
'Koralle'	1,11±0,41	2,54±0,22	0,31±0,19	2,02±0,16	0,54±0,11	2,39±0,37
'Erntedank'	1,17±0,60	2,65±0,31	0,37±0,21	2,56±0,21	0,61±0,17	2,81±0,31
'Masovia'	1,15±0,45	2,71±0,44	0,35±0,24	2,14±0,25	0,60±0,26	2,43±0,21

Notes: A – activation of axillary and apical meristems in percentages; V – viability, points;
medium № 1 – WPM + 1 g·L^{-1} of sorbitol; medium № 2 – WPM + 3 g·L^{-1} sorbitol; medium № 3 – WPM + 2 g·L^{-1} of sorbitol;
WPM – the main medium for clonal micropropagation of highbush blueberry and cranberry ordinary: macro- and micronutrients in WPM , mesoinositol – 100 mg·L^{-1}, adenine sulfate – 80 mg·L^{-1}, B_1 – 1 mg·L^{-1}, B_6 – 0,5 mg·L^{-1}, glycine – 2 mg·L^{-1}, IAA – 4 mg·L^{-1}, 2iP – 15 mg·L^{-1}.

T a b l e 3.The viability (V) of introduced varieties of *Vaccinium corymbosum* L. and *Vaccinium vitis-idaea* L. on different nutrient media, in 12 months after cultivation.

Variety	medium № 4 (sorbitol – 1g·L^{-1})		medium № 5 (sorbitol - 2 g·L^{-1})		medium № 6 (sorbitol - 3 g·L^{-1})	
	lengthofshootson 1 plant, cm	V, point	length of shoots on 1 plant, cm	V, point	length of shoots on 1 plant, cm	V, point
Vaccinium corymbosum						
'Bluecrop'	2,15±0,52	1,39±0,29	1,97±0,30	0,99±0,16	2,07±0,27	1,29±0,19
'Blueray'	2,47±0,47	1,54±0,32	2,17±0,32	1,27±0,25	2,33±0,35	1,43±0,29
'Dixi'	2,91±0,39	1,12±0,51	2,59±0,21	0,84±0,30	2,81±0,40	1,07±0,31
'Herbert'	2,39±0,54	1,65±0,39	2,36±0,50	1,17±0,35	2,64±0,49	1,44±0,47
'Rancocas'	2,05±0,46	1,08±0,35	2,84±0,50	0,76±0,19	2,51±0,43	0,89±0,25
'Scammel'	2,91±0,43	1,39±0,40	2,51±0,49	1,01±0,27	2,43±0,40	1,27±0,33
Vaccinium vitis-idaea						
'Koralle'	4,15±0,73	2,05±0,32	4,01±0,69	1,37±0,29	3,15±0,50	1,34±0,39
'Erntedank'	3,92±0,60	1,94±0,25	3,27±0,54	1,15±0,19	3,07±0,47	1,41±0,20
'Masovia'	4,25±0,65	1,65±0,30	4,09±0,60	1,29±0,27	3,44±0,39	1,37±0,26
Variety	medium № 7 (mannitol – 1g·L^{-1})		medium № 8 (mannitol – 2g·L^{-1})		medium № 9 (mannitol - 3 g·L^{-1})	
	length of shoots on 1 plant, cm	V, point	length of shoots on 1 plant, cm	V, point	length of shoots on 1 plant, cm	V, point
Vaccinium corymbosum						
'Bluecrop'	3,21±0,54	1,17±0,35	3,15±0,50	2,01±0,50	3,17±0,51	2,38±0,40
'Blueray'	3,87±0,69	2,31±0,40	3,67±0,59	2,04±0,35	3,84±0,67	2,65±0,39
'Dixi'	3,91±0,71	2,15±0,29	3,51±0,60	2,71±0,34	3,25±0,59	2,83±0,37
'Herbert'	4,01±0,74	2,01±0,27	3,91±0,65	2,58±0,39	3,01±0,53	1,07±0,18
'Rancocas'	3,09±0,65	2,54±0,45	3,37±0,70	1,89±0,31	3,67±0,78	2,27±0,33
'Scammel'	3,23±0,68	1,95±0,31	3,18±0,60	2,12±0,49	3,24±0,59	2,19±0,11
Vaccinium vitis-idaea						
'Koralle'	4,21±0,69	2,51±0,37	4,81±0,75	2,61±0,40	4,37±0,70	2,89±0,47
'Erntedank'	4,39±0,71	2,66±0,40	4,15±0,69	2,84±0,51	4,25±0,67	2,25±0,36
'Masovia'	4,57±0,80	2,19±0,36	4,37±0,68	2,39±0,44	4,08±0,59	2,91±0,50

The highest viability as a result of direct culture material on fresh culture media for 12 months was marked for varieties of cranberry 'Masovia' − 2,91 points on a medium № 9, containing 3 g · L^{-1}mannitol (Table 3); lowest – 0,76 points − for a variety of blueberry 'Rancocas' on the medium № 5, containing 2 g · L^{-1} sorbitol. The remaining varieties took an intermediate position.

This is an indication of dependence of the viability, how on variety belonging of material and from the inhibitor, contained in the culture medium.

The optimum variant for long-term cultivation in the 12 months of studied varieties of highbush blueberry should be considered media (№ 7 - № 9), containing mannitol: for 4 varieties − 'Bluecrop', 'Blueray', 'Dixi', 'Scammel' – medium № 9, containing 3 g · L^{-1} of mannitol, for 'Herbert' − medium №8, containing 2 g · L^{-1} of mannitol, for 'Rancocas' − medium №7, containing 1 g · L^{-1}mannitol. In this case, the mannitol has an inhibitory effect on the growth rate of highbush blueberry, thereby preventing aging, which resulted in a high rate of viability.

Media containing sorbitol,can not be recommended for long-term cultivation (12 months) of introduced varieties of highbush blueberry and cranberry ordinary.

Comparative analysis of the viability of the material depending on his varietal and species identification showed that the viability of varieties of cranberries is higher than of varieties of blueberries. Probably, this is connected with the functional features, inherent in the evergreen shrubs, to which cranberry belongs. This demonstrates about dependence of the viability of regenerants not only on the osmotic inhibitors in the medium but also from a plant genotype.

Conclusion

The results of experimental research allow us to state that the addition of osmotic inhibitors into nutrient medium promotes to reducing of the growth rate, the prevention of aging and provision of viability of the material for 12 months without transfers it on fresh nutrient medium. It is necessary to take into account the variety and species specificity of the investigated material, clearly manifested by the action of inhibitors and also selective action of the inhibitors themselves.

References

Ali, S., Khan, N., Mehmood, A., Haider, S. Raheem,A. 2016.In vitro conservation of exotic potato genotypes through different incubated temperatures, aerophilic and micro-aerophilic conditions. International Journal of Biodiversity and Conservation.8(7): 147-152.

Brindgen, M.P., Staby, G. L. 1981. Low Pressure and Low Oxigen Storage of Nicotianatabacum and *Chrysanthemum morifolium*.Tissue cultures. Plant Sci. Lett. 22 (1): 117-185.

Brindgen, M.P., Staby, G. L. 1983. Protocols of low-pressure storage. In: Handbook of Plant cell culture. Ed. Evens. Sharp. Ammirato, Jamada, Macmillan publ. company. N. Y. 1: 816-827.

Cathey, H. M. 1964. Physiology of growth retarding chemicales. Ann. Rev. Plant of Physiol. 15: 271-302.

Chailakhyan, M.H. 1967. Action of retardants on plants.Chemistry in Agriculture. 5 (9): 26-30.

Chernrts, A.M., Abramenko, N.M., Stakanova, R.V. 1988. Development of long-term storage *in vitro* method of virus-free clones of fruit trees and strawberry.Biology of cultivationed cells and biotechnology. Novosibirsk. Part 2:392.

Ciobaniui, I.B., Constantinovici, D. 2012. The effects of sorbital and conservation period on the *in vitro* evaluation of Solanumtubersoum L. plantlets.CercetăriAgronomiceîn Moldova.45:(3):151.

Codaccioni, M., Vescovi, P. 1987. Croissance in vitro du *Menthaviridis* (L.) en presence de mannitol: Influence des conditions de culture anterieuresauxquellesontetesoumis les vitroplantsayantfourmi les explantats. Bull. Soc. bot. Lett. bot. 134 (3): 269-274.

Coman, T., Neculae, L. 1981. Date preliminigermoplasmi de capsum. Probl.Genet.Apl. Bucurest. 13 (6): 397-401.

Demenko, V.I., Trushechkin, V.G. 1988. Storage of strawberry plants. Biology of cultured cells and biotechnology. Novosibirsk. Part 2:393.

Dorion, N., Regnard, J.L., Serpette, I., Bigot, C. 1994. Effects of temperature and hypoxic atmosphere on preservation and further development of *in vitro* shoot of peach ('Armking') and peach x almond hybrid ('GF 677 '). Scientia Hort. 57: 201-213.

Hae-Boong, J., Ssang-Hee, H., Kwang-Yoon, K. 1996. In vitro preservation method of culturing shoot tip at low temperature in strawberry.RDA. J. Agri. Sci. 38 (1): 284-289.

Homes, I., Dubus, E., Bourdan, I. 1982.Cold Storage of Plant Tissue Culture. V. Inter. Congr. of Plant Tissue and Cell. Culture. Japan. P. 801.

Kende, H., Ninnemann, H., Lang, A. 1963. Inhibition of gibberelline acid biosynthesis in *Fusarium moniliforme* by AMO-1618 and CCC. Naturwissenschaften. 1: 50-55.

Lloyd, G., McCown, B. 1981. Commercially feasible micropropagation of mountain laurel, *Kalmia latifolia*, by use of shoot tip culture. Proc. Intern. Plant Prop. Soc. 30: 421-427.

Mohammed, A.I. 1998. Micropropagation, long-term deposits and cryopreservation *in vitro* of red raspberry. Author. diss. cand. biol. sciences. Moscow. Nauka: 17.

Mullin, P.H., Schelegel, D.E. 1976. Cold Storage Maintenance of Strawberry Meristem Plantlets. Hort. Sci. 11 (2): 100-105.

Muromtsev, G.S., Butenko, R.G., Tihonenko, T.I., Prokofiev, M. 1990. Fundamentals of agricultural biotechnology. Moscow. Nauka: 384.

Pua, E. C., Chong, C. 1985. Regulation of in vitro shoot and Root Regeneration in Maspure Apple by Sorbioto: (D-glucitol) and Related Carbon Sources. J. Am. Soc. Hort. Sci. 110 (5): 705-709.

Rahman, M.H., Islam, R., Hossain, M., Islam, M.S. 2010.Role of sucrose, glucose and maltose on conventional potato micropropagation. J. Agric. Technol. 6(4):733-739.

Rolland, F., Gonzalez, E.B., Sheen, J. 2006. Sugar sensing and signaling in plants: conserved and novel mechanisms. Annu. Rev. Plant Biol. 57: 675-709.

Rubluo, A., Kartha, K.K. 1985. *In vitro* Culture of Shoot Apical Meristems of Various Phaseollus Spesies and Cultivars. J. Plant Physiol. 119 (5): 425.

Salmatova, I.S., Dunayeva, S.E., Sharova, E.I., Shchipachev, S.M., Medvedev, S.S., Gavrilenko, T.A. 2009.Features of micropropagation and dynamics of morpho-physiological indicators of some representatives of the genera Rubus and Fragaria (Rosaceae) at the storage *in vitro*.Plant.resources. 45 (4): 1-12.

Samarina, L. S., Choudhary, R., Kolomiets, T. M., Abilfazova, Y. S., Saran, P. L. 2014. *In vitro* Conservation Technique for Russian Citrus limon. Agricultural Research. 3 (4): 279-283.

Samsonova, O.N. 1991. Long-term preservation of test-tube plants of strawberry. Author.dis. cand. biol. sciences. Moscow. Nauka: 19 p.

San José, M.C., Janeiro, L.V., Corredoira, E. 2010. Micropropagación del alisocomúnpara la conservación de sugermoplasma. Spanish J. Rural Develop. 1 (3): 31-38.

Schilde-Renther, L., Espinosa, N., Estrada, R., Lizarrga, R. 1982.*In vitro* Storage and Distribution of Potato Germplasm. Plant Tissue Culture. V. Inter. Congr. of Plant Tissue and Cell. Japan: 781.

Tukai, S., Morii, M., Hasahuru, O. 1988.Storage of Chrysanthemum (*Dendranthema grandiflorum* (Ramal.)Kitamura) plantlets *in vitro*. 5 (1): 20-25.

Vysotskaya, O.N. 1994. Long-term preservation in vitro of collection of strawberry plants. Plant Physiology. 41 (6): 935-941.

Wanas, W. H., Collow, J. A., Lindsey, A. W. 1986.Growth Limitation for the Conservation of Pea Genotypes. Plant Tissue Culture and its Agricultural Applications. London: Butterworths: 285-291.

Westcott, R.J. 1981a. Tissue culture storage of potato germplasm. 1. Minimal growth storage. Potato Research. 24: 331-342.

Westcott, R.J. 1981b. Tissue culture storage of potato germplasm. 2. Use of growth retardants. Potato Research. 24: 343-352.

Winthers, L.A. 1989. Cryopreservation and storage of the genofond. Proc.: Plant Biotechnology: cell culture. Moscow. Nauka: 204-233.

Winthers, L.A. 1979. Treeze preservation of somatic embryos and clonal plantlets of carrot (*Daucus carota* L.). Plant Physiol. 63: 460-467.

Zadontsev, A.I., Pikush G.R., Grinchenko, A.R. 1973. Chlorholyn chloride in plant-growing. Moscow. Nauka: 360.

Access this Article in Online

Website: www.ijarbs.com

Subject: Agricultural Sciences

Quick Response Code

DOI:10.22192/ijarbs.2018.05.01.015

How to cite this article:

Elena Kutas, Aleksandr Veyevnik, Vladimir Titok, Lyubov Ogorodnyk. (2018). Effect of osmotic inhibitors on reducing of growth rate and the preservation of viability of sterile cultures. Int. J. Adv. Res. Biol. Sci. 5(1): 84-90.

DOI: http://dx.doi.org/10.22192/ijarbs.2018.05.01.015