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CULTIVATION OF PLANTS OF POTATO IN VITRO AT MICROCLONAL REPRODUCTION

The purpose. To determine optimum root formation in crop *in vitro* of medium-early cultivars of potato Nevskaya depending on acidity of nutrient medium (pH), intensity of illumination and photoperiod for increase of production of the initial improved planting stock. **Methods.** Complex use of laboratory, mathematical and statistical, calculation-comparative methods and system analysis. **Results.** Experimental data concerning influence of acidity of nutrient medium (pH), intensity of illumination and photoperiod on induction of root formation are brought at microclonal reproduction of the improved initial material. It is fixed that pH of nutrient medium influences intensity of root formation. **Conclusions.** The maximum productivity indexes and economic efficiency of plants *in vitro* of medium-early cultivar Nevskaya are gained at pH=5,3, photoperiod of 16 h and illumination of 2500 l. Intensity of root formation made 92,7%, mass of average microtuber — 667,7 mg, mass of microtubers for 1 plant — 617,3 mg, amount of microtubers in mass more than 350 mg — 79,1% at profitableness of production of 117 %, cost prices 7,37 hrn/microtuber, conditional net profit — 8,63 hrn/microtuber.

Key words: root formation, amount of interstitial sites, height of plants, acidity of nutrient medium, photoperiod, intensity of illumination, mass of microtubers.

The necessary basis for the development of the technology for obtaining micro- and minibools is the knowledge of the mechanism of tuber formation as a physiological and biochemical process and its methods of regulation. It is established that tuber formation in a plant is induced by a system of factors, :excess of assimilants, hormonal state of the plant, photoperiod, temperature decrease, nitrogen deficiency, change of tragacanth centers due to the damping of the activity of the apical meristem of the stalk in the direction of soboles and tubers, ontogenetic state of the plant [1 , 2]

[3]With the result that the process of tuber formation can be regulated by a number of endo- and exogenous factors, which are the basis for a number of micro and minitubers technologies in primary potato seed production, aimed at the long-term preservation of the reproductive properties of virus tested seed potatoes. [3] The success in cultivating cultures of cells, tissues and organs of plants is determined by the nutrient midst composition. [4,5,6] . For cultivating the potato culture the Murashige, Skoog (MS) [7] environments are used. They include macro- and micronutrients, iron, calcium, vitamins and growth stimulants. The acidity of this medium is important. It is known that in the native conditions a plant cell functions in the close variation limits of the hydrogen ions concentration. The relative stability of the pH value in the intracellular and environment of the cell is supported by buffer systems, in which the protein molecules play an essential role as ampholes. The structure and activity of biological macromolecules, preeminently proteins, especially enzyme proteins depend on the Ph value[4]. Also, the acidity of the medium determines the availability of nutrients for plants in vitro. . It is known that very acidic or alkaline media limit the flow of some elements as phosphorus and iron, making them relatively insoluble, limiting the growth of plants. At the same time, other elements with high acidity pass into the dissolved condition and become toxic to explants [9]. . It is very important that all processes go at a certain acidity. Especially it is necessary for biological catalysts as an enzymesthe to function properly(their activity can be sharply slowed down if they go through these limits). On the efficiency of the biotechnological method for obtaining qualitative source material is substantially influenced by other tuber factors formation such as the illumination intensity and the photoperiod affect on the efficiency of biotechnological method for receiving a qualitative source material. In such a way, investigation of above mentioned factors interaction is important for optimizing the process of potato tubers formation in vitro [6].

Purpose. The goal of the article is to determine the optimal mode of tuber formation in the culture *in vitro* medium early ripen potato variety Nevska,

depending on the acidity of the nutrient medium (pH), light intensity and photoperiod to increase the production of the original planting material disinfected.

Materials and research methods. The intensity of potato tuber formation of different varieties with the same acidity is different. The influence of the nutrient medium pH in interaction with the photoregulation over the Nevsky-type potato bulbous formation intensity in the culture in vitro was studied in the microclone laboratory. Three factors were put into the study: factor A - photoperiod (10 and 16 hours), factor B - light intensity (1500 and 2500 lux), factor C - acidity of the medium (pH = 4.3, 4.8, 5.3). Studies were performed according to generally accepted methods. To obtain the healed potato plants in vitro outcomes by biotechnological method the method of thermo-chemotherapy in combination with the apical meristem culture was used, in accordance with "Methodological recommendations for research on potatoes" [5], methodological recommendations "Improvement of potato in culture in vitro" [8], "Optimization of methods of rehabilitation, reproduction and protection of seed potatoes against viral infection" [10] and "Biotechnological methods for the receipt and evaluation of healed potatoes" [3]. The experiments were carried out according to generally accepted methods [11, 12]. The economic production efficiency of recovered source material in culture in vitro was calculated based on the actual cost of microtubers according to technological maps.

Research results. During the observing the growth and development of plants, it was found that the average increase in plant height depended on the photoperiod. So, on the 20th cultivation day at sixteen-hour illumination the plants in vitro were higher averagely by 0,2 cm than at ten-hour (Table 1).

On the 40th day of observation the height growth of plants during the 16-hour photoperiod was also higher by 44.4% than at 10 hours and was 2.6 cm. On the 20th and 40th day of cultivation the number of internodes was higher during the 16-hour photoperiod and averagely was , 4.1 and 5.8 pcs. vs 3.8 and 5.2 pcs. on 10-hour illumination. Nevsky is a medium-sized potato variety. Therefore, on the 20th

day of observation on the process of sixteen-hour lighting, only 5.1% of the microtubers were formed, but at the ten-hour - 3.6%. On the 40th day of cultivation the photoperiod almost did not affect on the microtubers formation: 12.1% of plants formed microtubers at sixteen hours, and at ten - 12.8%. During the interaction of the photoperiod and the intensity of the light, the height of the plants is almost the same in the 16-hours photoperiod on the 20th day - 4.2 and 4.3 cm at 1500 and 2500 lux, respectively, and at 10 hours and 1500 lux increase was 4.6 cm, which is 31.4% more than at 2500 lux. On the 40th day of cultivation the height growth of plants within the photoperiods is different. As a result, at 10 hours, it is 1.5 cm at 1500 lux, against 2.1 cm at 2500 lux. During the 16-hour photoperiod the height growth of plants amounted to 2.8 and 2.3 cm at 1500 and 2500 lux, respectively. On the 20th day of observations 5.3% of microtubers were obtained against 1.9% at 1500 lux during the 10-hour photoperiod and at 2500 lux. At 16 hours - 4.6 and 5.7%, respectively. On the 40th day of cultivation, the intensity of tuber formation is higher at a higher intensity of illumination. Thus, for ten hours and the intensity of light, 1500 lux was formed by 4.9% of microtubers - it is less than at 2500 lux. At sixteen hours and in the illumination of 1500 lux 9.8% of plants formed microtubers, which is 1.5 times less than at the 2500 lux.

1. The acidity (pH) Influence of the growing medium, photoperiod and quality of illumination on tuber formation of the Nevsky potato in the culture in vitro.

photoperid, hours	quality of illumination, lux	growing medium pH	On the day of cultivation									
			20th				40th				60th	80th h
			Plant height, cm	number of internodes.	The number of plants, which produced,%		height growth of plants, cm	number of internodes,.	The number of plants, which produced,%			
					soboles	tubers			soboles	tubers	tubers	tubers
10	1500	4,8	5,2	4,2	99,7	0,3	1,7	5,8	91,0	9,0	19,7	55,7
		4,3	4,3	3,9	98,3	1,7	1,5	5,1	92,7	7,3	14,0	30,0
		5,3	4,0	3,7	96,3	3,7	1,3	4,9	85,3	14,7	25,3	45,7
	2500	4,8	3,3	3,5	91,3	8,7	1,9	4,8	74,3	25,7	47,3	83,0
		4,3	3,5	3,9	95,0	5,0	2,3	5,5	91,7	8,3	9,3	59,7
		5,3	3,6	3,4	97,7	2,3	2,0	5,1	88,3	11,7	22,0	83,7
16	1500	4,8	3,5	3,5	97,7	2,3	3,3	5,8	94,0	6,0	13,0	45,0
		4,3	4,9	4,4	95,0	5,0	2,7	6,4	90,7	9,3	19,3	49,7
		5,3	4,2	4,2	90,3	9,7	2,4	5,6	86,0	14,0	25,7	74,0
	2500	4,8	4,5	4,3	98,7	1,3	2,4	6,0	90,7	9,3	28,0	76,7
		4,3	4,4	4,1	94,3	5,7	2,2	5,7	85,3	14,7	42,3	71,7
		5,3	3,9	3,8	93,3	6,7	2,3	5,4	81,0	19,0	43,7	92,7

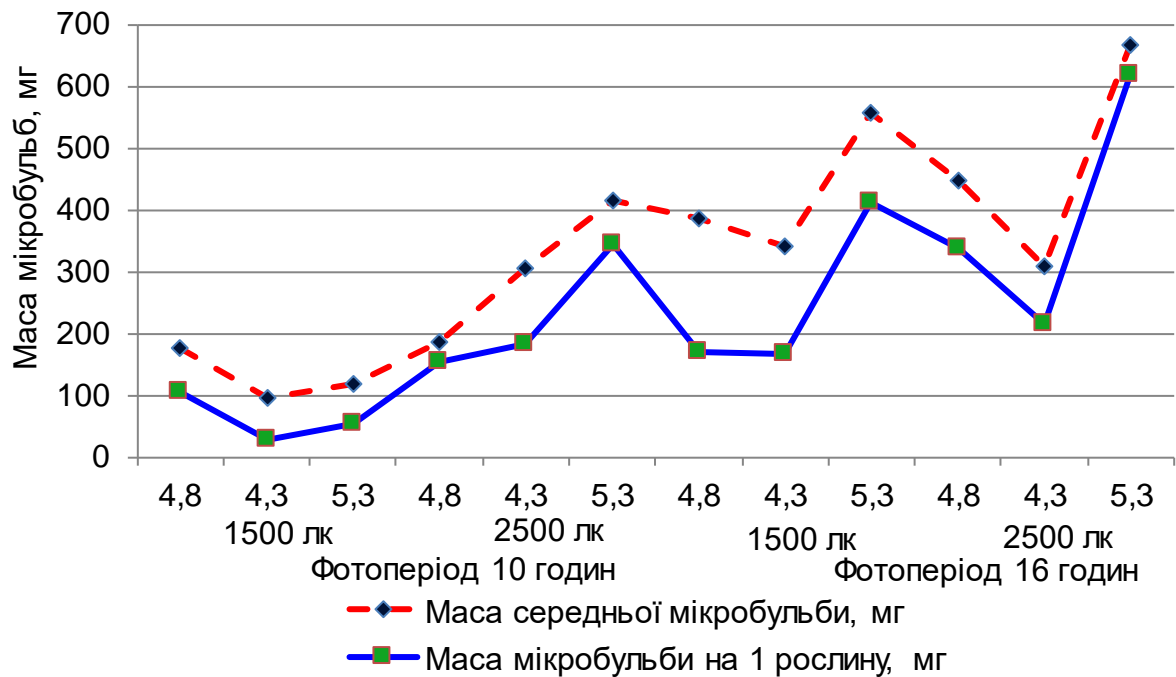
The nutrient medium acidity on the 20th and 40th day of observations slightly influenced over the height growth of plants and the number of internodes. On the 20th day the height growth of plants is 3.9; 4.3 and 4.1 cm (pH = 5.3, 4.3, 4.8); number of nodes -3.8; 4.1 and 3.9 pcs, respectively. On the 40th day the height growth of plants is 2.0; 2.2 and 2.3 cm, respectively, and the number of internodes - 5.3; 5.7 and 5.6 pieces (pH = 5.3, 4.3, 4.8). The tuber intensity on the 20th and 40th day of cultivation was higher at the medium pH of 5.3 - 5.6% and 14.8% vs. 4.3 and 9.9% and 3.2 and 12.5% at the pH 4.3; 4.8. On the 60th day of observation, the best indicators of tuber formation during the 10- hours photoperiod and the intensity of illumination at 2500 lux and pH amount to 4.8 - 47.3%, and at sixteen hours at 2500 lux illumination and pH to 5.3-43.7%. On the 80th day of cultivation, the intensity of tuber formation has increased significantly. Thus, at ten

hours of the photoperiod, 2500 lux and pH = 5.3, the tuber intensity was 84.0%, and at sixteen hours, 2500 lux and pH = 5.3 - 91.0%. At 16 hours of the photoperiod, the mass of the average microtubers was 450.7 mg, the weight of microtubers on one plant was 320.0 mg, which is 2.1 and 2.2 times more than at the ten-hour photoperiod (Table 2). The tuber formation process was significantly influenced by the intensity of illumination. At 2500 lux, 77.9% of plants formed microtubers vs 50.0% at 1500 lux. The mass of the average microtubers and the mass of microtubers per plant at 2500 lux illumination amounted to 387.9 and 308.2 mg, respectively, which is 108.8 and 151.9 mg more than at 1500 lux, respectively. There was a significant difference between the bulb intensity, the mass of the average microtubers and the mass of microtubers per plant at various pH values in the nutrient midst. The lowest tuber formation intensity was observed at pH 4.3 and amounted to 52.8%, which was 12.3 and 21.2% lower than the acidity of 4.8 and 5.3, respectively. The experiment was noted the mass reduction of the average microtubers at the nutrient medium pH which is 4.3 - 262.7 mg versus 298.5 mg and 439.2 mg at pH 4.8 and 5.3, respectively. At a lower nutrient midst pH the mass of microtubers per plant was 148.4 mg, which is 43.5 and 208.0 mg less than at pH 4.8 and 5.3, respectively. If we compare the tuber formation with different intensity of light, , the mass of the average microtuber within the ten-hour period of the photoperiod and at 2500 lux is 2.4 times more than at 1500 lux and amounts to 301.8 mg, but the mass of microtubers per plant at 2500 lux is 3, 6 times more than 1500 lux illuminate and amounts to 226.0 mg (Fig. 1). In percentage terms, the tuber intensity at 2500 lux is 75.5% versus 43.8% at 1500 lux.

2. Productivity of Nevsky potato plants in the culture in vitro depending on the acidity of the nutrient midst(Ph), photoperiod and illumination intensity.

photoperiod, hours	quality of illumination lux	growing medium pH	the average weight of microtubers, mg	The microtuber mass, mg/plant	Microtuber yield over 350 mg, %	The number of plants, which produced microtubers, %	The number of microtubers per one plant
10	1500	4,8	177,6	106,1	13,2	55,7	0,58
		4,3	96,3	29,2	0,0	30,0	0,30
		5,3	117,7	53,6	0,0	45,7	0,49
	2500	4,8	185,1	152,6	10,0	83,0	0,81
		4,3	306,4	182,3	35,8	59,7	0,60
		5,3	413,9	343,0	54,6	83,7	0,84
16	1500	4,8	384,7	171,1	46,0	45,0	0,45
		4,3	340,7	166,1	40,9	49,7	0,50
		5,3	557,3	411,5	71,6	74,0	0,72
	2500	4,8	446,7	337,9	58,3	76,7	0,79
		4,3	307,3	215,8	34,9	71,7	0,70
		5,3	667,7	617,3	79,1	92,7	0,91

Within the photoperiods of medium influences the pH influences the tuber formation. At 10 hours, the average weight of microtubers amounted to 5.3 is 265.8 mg, which is 84.4 and 64.4 mg more than at pH 4.8 and 4.3, respectively. At 16 hours the mass of the average microtubers at a pH of 5.3 - 612.5 mg versus 415.7 mg and 324.0 units. at pH 4.8 and 4.3, respectively. The mass of microtubers per plant at 10 hours is significantly higher at pH 5.3- 198.3 mg, which is 68.9 and 92.5 mg more than at pH 4.8 and 4.3. During the 16-hours photoperiod at pH 5.3, the mass of microtubers per plant is 514.4 mg, which is 2.0 and 2.7 times higher than at pH 4.8 and 4.3.



Pict. 1 The Influence of nutrient medium photoregulation and acidity on the microtubers formation of the Nevsky potato variety in the culture in vitro.

The cost of one microtubers in applying sixteen-hour illumination increases by 16.0%, compared with ten hours, on average by factor (Table 3). Using the intensity of the illumination of 2500 lux during the plant in vitro cultivation the cost was reduced by 35.6%, while at pH of the nutrient midst it was 8.7 and 29.7% lower than at pH 4.8 and 4.3.

Conclusions. Maximum indices of productivity and economic efficiency of middle-class Nevsky plants in vitro were obtained at pH = 5.3; also during the 16-hour photoperiod and illumination 2500 lux: the tuber formation intensity was 92.7%, the weight of the average microtubers - 667.7 mg, the mass of microtubers per plant - 617.3 mg, the number of microtubers weighing more than 350 mg - 79.1% with the profitability of production 117%, the cost of 7.37 UAH / microtubers, the conditional net profit was 8.63 UAH / microtubers.

3. Growing productiveness of the Middle Class Nevsky potatoes microtubers in the culture in vitro depending on photoregulation and acidity of the nutrient midst.

photoperiod, hours, (A)	quality of illumination lux (B)	growing medium pH (C)	The number of microtubers per one plant	Costs per plant, UAN	Prime cost, UAN / microtuber	conditional net profit or loss, UAN / microtuber	profitability, %
10	1500	4,8	0,56	6,00	10,71	5,29	49
		4,3	0,30	6,10	20,33	-4,33	-21
		5,3	0,46	6,20	13,48	2,52	19
	2500	4,8	0,83	6,35	7,65	8,35	109
		4,3	0,60	6,45	10,75	5,25	49
		5,3	0,84	6,55	7,80	8,20	105
16	1500	4,8	0,45	6,30	14,00	2,00	14
		4,3	0,50	6,40	12,80	3,20	25
		5,3	0,74	6,50	8,78	7,22	82
	2500	4,8	0,77	6,65	8,64	7,36	85
		4,3	0,72	6,75	9,38	6,63	71
		5,3	0,93	6,85	7,37	8,63	117

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