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Efficiency of the Technology of Propagation of Cherries Under Conditions of in Vitro

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Abstract: *The article substantiates the relevance of the method of microclonal propagation and cultivation of plants in vitro as a cost-effective technology for the production of planting material, which allows producing a sufficient number of plants for scientific and industrial purposes. In order to establish the effectiveness of the new generation of growth stimulators previously not used in in vitro culture, to compare them with the standard substances used in clonal micropropagation, an economic assessment was made of the effectiveness of the technology for propagating promising stocks of cherry in intensive conditions in vitro, developed in the laboratory "In vitro and biotechnology »Scientific Research Institute of Horticulture, Viticulture and Winemaking named after Academician Mahmud Mirzaev. An economic assessment of the technology for the propagation of promising sweet rootstock in vitro in intensive conditions included the selection of an object for introducing innovations, a description of the technology, costing, and the determination of economic evaluation indicators. The methods used were based on a systematic approach and universally recognized proven methods used in scientific research for fruit crops. The results of experimental studies were processed using the program "BorlandC ++ Bulder6" and the electronic program "O`ITS" developed by the project participants to carry out calculations to assess the economic efficiency of innovative technologies used in agriculture. To determine the economic assessment of innovative technology, a methodology was developed that included economic indicators such as a resource conservation coefficient, a gross innovation growth rate, and a technology innovation degree. Based on the studies, it was discussed that the microclonal method of vegetative propagation of cherries has established itself as highly effective and economically viable.*

Keywords— innovations, cherries stocks, technology, prime cost, profit, resource saving ratio, innovation gross growth rate, breeding rate, efficiency

1. INTRODUCTION

The most important task in the development of the agricultural industry in the Republic of Uzbekistan is a significant increase in the production of fruit and vegetable products most demanded by the population. Among the fruit crops that are of particular value in solving these problems, one of the first places belongs to the sweet cherry. Sweet cherry is one of the most profitable fruit crops, which is important for cultivation in areas favorable for the realization of its great productivity potential.

Uzbekistan is one of the 40 countries in the world where cherries are grown in large commercial volumes. According to the Corporate Statistical Database of the UN Food and Agriculture Organization (FAOSTAT), today Uzbekistan is one of the ten world leaders in the cultivation of sweet cherries. Of the 100 thousand tons of cherries produced in Uzbekistan per year, at least 30 percent are exported abroad, including to the CIS countries. So, in 2018, Uzbekistan was among the four largest world exporters of cherries, yielding in terms of export volume only to three countries: Chile, the USA and Turkey [26].

Despite the presence of favorable conditions for growing fruit crops in Uzbekistan, global climate changes, problems of land and water resources in the republic, an increase in the susceptibility of many agricultural crops to diseases and pests necessitate the implementation of breeding programs aimed at obtaining highly adaptable technological varieties of intensive plantings. In addition, there is a large reserve for expanding the area of commercial production of fruits, including sweet cherries, in the steppe regions of many regions of the republic. This also becomes possible only with the use of the latest achievements of science and practice in the development of modern intensive cultivation technologies, the defining elements of which are varieties and rootstocks. Without new varieties, without new hybrids, without more sophisticated technologies, it is impossible to increase the output of agricultural products, ensure the country's food security and increase the export potential of agricultural producers.

Selection of the most effective variety-rootstock combinations of sweet cherries using new varieties and rootstocks is a necessary condition for the development of regional technology for cultivation of sweet cherries and the successful development of domestic horticulture. Today, a cost-effective technology for the production of planting material, which allows the production of plants in sufficient quantities for scientific and industrial purposes, is the method of micropropagation and in vitro plant growth.

Micropropagation - obtaining in vitro plants that are genetically identical to the original explant (method of vegetative propagation of plants in in vitro culture). Micropropagation is based on a unique property of a somatic

plant cell - totipotency - the ability of cells to fully realize the genetic potential of the whole organism [20].

The prospects of the method of microclonal propagation of plants in the system of production of healthy planting material of the highest quality categories are beyond doubt. This is justified by numerous studies that date back to the middle of the 20th century. In particular, the first achievements in the field of clonal micropropagation were obtained in the late 50s of the XX century by the French scientist Georges Morel, who managed to obtain the first regenerant orchid plants. The achievement of success was facilitated by the technique of cultivation of the apical meristem of plants in vitro, already developed by that time [27].

As a rule, the researchers used the apical meristems of herbaceous plants as the primary explant: carnations, chrysanthemums, sunflowers, peas, corn, dandelions, lettuce, and studied the effect of the composition of the nutrient medium on the processes of regeneration and plant formation. J. Morel in his works also used the apex of the cymbidium (family orchid), consisting of a growth cone and two or three leaf primordia, from which, under certain conditions, he observed the formation of spherical spheres - protocorms. The formed protocorms could be divided and then cultivated independently on a newly prepared nutrient medium until the formation of leaf primordia and roots. As a result, he discovered that this process is endless and that high quality and genetically uniform, virus-free planting material could be obtained in large quantities [28].

2. RESEARCH METHODOLOGY

Today, in Uzbekistan there are laboratories at almost all major scientific institutions, where issues related to the problems of microclonal reproduction of plants and their recovery are studied. In particular, in the laboratory "Biotechnology" of the Scientific Research Institute of Horticulture, Viticulture and Winemaking named after Academician Mahmud Mirzaev, research is being carried out to select the optimal media and ways to reduce infection in vitro to obtain the largest number of full-fledged seedlings of sweet cherry varieties from intraspecific hybridization.

Recognizing the effectiveness and high importance of innovations in breeding, seed production and plant protection for the agricultural sector, it should be noted that the transition to the accelerated development of innovation requires the creation of a new organizational and economic mechanism aimed at defining priorities in the innovation sphere, implementing strategic planning for the development of a self-organization mechanism, organization of expertise and evaluation of innovative projects [17].

Measuring innovation for evaluating innovation projects is necessary, since the low ability to generate innovation in the agricultural sector is associated not so much with a lack of technology, but with the lack of adequate methodological tools necessary for making decisions in the field of innovation to determine the key factors that stimulate innovation in the agricultural sector.

Based on the foregoing and taking into account the fact that currently in horticultural production circles there is a discussion about the advisability of using virus-free planting material obtained by in vitro micropropagation due to its high cost to establish the effectiveness of growth stimulants that have not been previously used in in vitro culture. of the new generation, comparing them with the growth substances standardly used in clonal micropropagation, an economic assessment of the effectiveness of the technology of propagation of promising cherry rootstocks in intensive in vitro conditions was carried out, developed in the laboratory "In vitro and biotechnology" of the Research Institute of Horticulture, Viticulture and Winemaking named after Academician Mahmud Mirzaev [1.22].

The assessment was carried out on the basis of the methodology for assessing the economic efficiency of innovative technologies in agriculture, developed by us within the framework of the applied project PZ-20170928458 "Improving the use of innovative, resource-saving technologies in agriculture" (2018-2020) [18.19].

The research methodology was based on a systematic approach and generally recognized proven methods used in scientific research for fruit crops. In particular, laboratory work carried out in the course of the study was carried out according to the recommendations from the methodological manual of J. Driver “Artificial cultivation (in vitro) from tissues and cells in laboratory conditions.” [11] Phenological observations, biometric calculations, as well as laboratory theoretical and practical analysis was carried out according to the method of Kh. Ch. Buriev et al. [3], VL Vitkovsky [6], PL Feklistov and VV Khudyakov. [21] Cameral and variational-statistical processing of experimental data was carried out according to the method of BA Dospekhov. [10] The data obtained were registered with modern measuring instruments that passed state verification.

The results of experimental studies were processed using the "BorlandC ++ Bulder6" program and the "O`ITS" electronic program developed by the project participants to carry out calculations to assess the economic efficiency of innovative technologies used in agriculture[1].

3. RESEARCH RESULTS

Research on the development of an effective method for improving the nutrient medium and adapting them in vivo in order to increase the introduced promising low-growing sweet cherry rootstocks in vitro was carried out in the period

[1] Certificate No. DGU 06980 on the official registration of a program for electronic computers, issued by the Intellectual Property Agency under the Ministry of Justice of the Republic of Uzbekistan, registered in the state register of programs for electronic computers of the Republic of Uzbekistan on October 14, 2019.

from 2017 to 2019. The subject of the research was the types of nutrient media used for microclonal propagation of cherry rootstocks in vitro, the growth elements contained in them and their concentration.

The economic assessment of the propagation of promising cherry rootstocks under intensive in vitro conditions was carried out in the following sequence [2]:

1. Selecting an object for the implementation of innovation;

2. Description of technology;

3. Calculation of the cost;

4. Determination of indicators of economic assessment.

1. Selecting an object for the introduction of innovation. As objects of research, we used low-growing stocks of foreign cherry varieties Krymsky 5, Gizela 6, Gizela 5, Colt, introduced in the laboratory "In vitro and biotechnology" of the Research Institute of Horticulture, Viticulture and Winemaking named after Academician Mahmud Mirzaev.

As a basis for comparing this technology, the technology of growing cherry rootstocks in open field conditions was used. For growing seedlings from these rootstocks, 1 hectare of the area of the farm "Alisher Fayz" of the Tashkent region, 0.40 hectares of the area of the farm "Zaxriddin Flower Plantation" of the Zangiotinsky district of the Tashkent region, 0.70 hectares of the land plot of the farm "Tursunova Nafisa bogi uzumzori" and 0.50 hectares of the area of FH “Kordinal niktarin raspberry” in Pastdargom district of Samarkand region.

2. Description of technology. Shoots of rootstocks of introduced foreign varieties of sweet cherry VSL-2 (Krymsk 5), GISELA 5, GISELA 6, Colt were packed in plastic bags and transferred to the in vitro laboratory. All explants were washed for one hour in running water. At the next stage, they were kept for 30 seconds in a 70% ethanol solution. Surface sterilization of the starting material was provided with 0.1% and 0.2% sodium hypochlorite solution for 15-20 minutes. To remove the remaining solution from the sterilized shoots, they were washed three times with sterile distilled water.

For cultivation, the nutrient medium MS - Murashige-Skuga, ½ MS - a modified MS medium with vitamins, WPM - Woody Plant Medium, DKW - Driver and Kuniyuki were used [25].

To accelerate growth, growth regulators BAP and IBA were added to the nutrient medium. In particular, at the stage of proliferation, 6-benzylaminopurine (BAP) at a concentration of 0, 1, 2 mg / L + indolylbutyric acid (IBA) at a concentration of 0.01 mg / L were used. After 3-fold subculturing (with an interval of 21 days), the morphometric parameters of the plants were measured, which testified to the positive effect of the studied method of reproduction of all studied rootstocks in the experiment. In the phase of microshoot rooting, the IBA regulator was used at a concentration of 0, 1, 2 mg / L, and after 45 days the data were taken.

In all cases, the pH level of the nutrient medium was equated to 5.8, if necessary, it was balanced with solutions of 0.1 N KOH and HCl. The prepared culture medium was sterilized in an autoclave at a temperature of 1210C, a pressure of 105 Kra for 20 minutes.

The cultivation vessels were kept for 16 hours under photoperiodic conditions (40-45 MCE / m2 s), in an incubator at a temperature of 23 ± 1 ° C. The experiments were carried out in 4 different versions and 4 repetitions.

The next step was the cultivation of plants extracted from the culture vessels in a greenhouse in the soil and preparation for implementation.

3. Calculation of the cost. All costs associated with the implementation of this technology consist of production costs (DKW medium with vitamins, Murashige & Skoog Basal Medium with Vitamins, 6-Benzylaminopurine (BA), Indole-3-Butyric Acid (IBA), Gibberellic Acid, Nutrient Agar, D - (+) Glucose, Anhydrous, D-Sucrose, Flower Pot, peat, Ethanol 96%, 20% hydrogen peroxide, work gloves, sterile masks, gauze, scalpel, tweezers) and administrative costs (electricity costs, natural gas, wages). Calculations for all costs are presented in tables 1-3.

Table 1

The cost of growing 70 thousand cherry rootstocks in an in vitro laboratory (1 ha)

№	Funds names	Unit of measurement	Number	Cost, sum
	Laboratory costs			
1	DKW environment with vitamins	gram	2327	2 152 000
2	Fe Na-EDDHA	gram	34	40800
3	Murashige & Skoog with Vitamins (MS)	gram	130	1 115 625
4	6-Benzylaminopurine (BA)	mg	438	2000
5	Indole-3-Butyric Acid (IBA)	mg	182	6000
6	Kinetin	mg	400	2000
7	Naphthylacetic acid NAA	mg	90	2000
8	Gibberellin	mg	100	2000
9	Agar agar	Kg	3.226	646000
10	Sucrose	Kg	16	80000
11	Cassette (x88)	pieces	796	796000
12	Peat (Agrobalt 250 l)	Kg	800	5 168 000
13	70% ethanol alcohol	liter	10	300000
14	20% hydrogen peroxide	liter	2	40000
15	Sterile gloves	pieces	100	120000
16	Sterile masks	pieces	50	25 000
17	Sodium hypochlorite	liter	1	25 000
18	Gauze	meter	2	3000
19	Scalpel	pieces	10	30 000
20	Tweezers	pieces	10	100 000
21	Sterile wipes	pack	4	100
	Total			**10755425**

Note. Calculations were made in national currency - soum. For recalculation, you can use the rate of the Uzbek soum according to the Central Bank of the Republic of Uzbekistan for the settlement period (as of 01.11.2019): 1 USD = 9463.37 sum, 1 RUB = 148.40 sum. At the rate of the Central Bank of the Russian Federation: 67.4152 rubles for 10,000 Uzbek soums; 63.7748 rubles for 1 US dollar.

Table 2

Greenhouse costs

№	Funds names	Unit of measurement	Number	Cost, sum
1	Vessels	штук	70 000	3 850 000
2	Organic fertilizer	тонна	1	500 000
3	Mineral fertilizer (NPK-20-20-20)	кг	25	600 000
4	Plant protection products (insect-acaracide)	л	1	870 000
Total				**5 820 000**

Table 3

Administrative expenses

№	Funds names	Cost, sum
1	Electricity costs for 6 months	1 500 000
2	Labor costs for 6 months	24 000 000
3	CAP 25% (from the wage fund)	600 000
	Total	**26 100 000**
	Total costs	**41 699 425**
	Production losses 5%	2 084 975
	Total costs (including losses)	**43790400**

For a comparative analysis, the costs of production for growing cherry rootstocks by the traditional method and in vitro per 1 ha of land area are given (Table 4). According to the technological map, labor costs for workers when growing cherry rootstocks in the traditional way amounted to 7364.7 million soums, labor costs for in vitro production, calculated on the basis of a tariff scale, amounted to 24600 soums. When growing cherry rootstocks in the traditional way, 200 kg of seeds are consumed, at a price of 55 thousand soums per kilogram, the total cost was 11 million soums. As a plant protection agent, an insect-acaracid preparation (difen super 55% n. Cook) was used, the cost of which per hectare under in vitro conditions amounted to 870 thousand soums, and with the traditional method - 1.2 million soums. Other expenses include the cost of devices, in this case, 3850 thousand soums were spent on the purchase of 70,000 laboratory vessels.

4. Determination of indicators of economic assessment. The results of calculating the economic efficiency of cherry rootstocks grown by the in vitro method are presented in Table 5-6. This technology was used to grow cherry rootstocks of varieties Krymskiy 5, Gizela 6, Gizela 5, Colt in a nutrient medium MS, DKW, modified MS, WPM, while the MS medium was taken as a control.

The yield in the control environment MS from the Krymsky 5 variety was 70,000 stocks, Colt - 68,000, Gizela 5 - 70,000 and Gizela 6 - 63,000.

When determining the efficiency of each cherry rootstock using generally accepted indicators of profit and profitability, it was revealed that when growing rootstocks of the Krymsky 5 variety per 1 hectare of area, production costs amounted to 53,675.4 thousand soums, the cost price - 766.8 thousand soums, received 121,324.6 thousand sum of profit, the level of profitability was 226.0%.

Table 4

Production costs for growing cherry rootstocks in the traditional way and in vitro per 1 ha of area (thousand sum)

	Indicators	Technology	
		Traditional	*In vitro*
1	Wage	7364,7	24600
2	Seed costs	11000	-
3	Laboratory costs	-	10755,4
4	Mineral and organic fertilizers	4000	1100
5	Plant protection products	1200	870
6	Electricity costs	-	10000
7	Natural gas costs	-	1500
8	Water costs	-	1000
9	Fuel costs	714	-
10	Other costs	1500	3850
	Total	**25778,7**	**53675,4**

When growing Colt rootstocks from 1 hectare, a profit of 116324.6 thousand soums was received, the level of profitability was 216.7%, and the cost of one rootstock was 789.3 soums. Accordingly, the profit when growing Gizela 5 and Gizela 6 from 1 hectare amounted to 121,324.6 thousand soums and 103,824.6 thousand soums, the level of profitability - 226% and 193.4%, and the cost price - 766.8 soums and 852.0 sum. As you can see, the indicators of rootstocks of varieties Krymskiy 5 and Gizela 5 are the same. It is worth noting that the relatively low indicators of Gisela 6 do not affect the production efficiency of this rootstock variety, especially since all other previously tested clonal cherry rootstocks grown in the traditional way do not even reach the level of Gisela 6.

A comparative analysis of the economic efficiency of growing sweet cherry rootstocks in vitro and in the traditional way per 1 hectare of area is shown in Table 5.

As you can see, production costs for growing cherry rootstocks in the traditional way amounted to 25778.7 thousand soums, in vitro - 53,675.4 thousand soums, the cost of the product per 1 ha was 380.4 thousand soums with the traditional method and 715 , 7 thousand soums when grown in vitro, i.e. difference of 335.29 thousand soums. At the same time, the gross revenue and profit for growing in vitro, respectively, are 139,500 thousand soums and 111,603.3

thousand soums more than with the traditional method, which of course affects the level of profitability, which reached 249.3% in vitro. The economic effect from the use of innovative technology, in this case, expressed in the increase in profit, amounted to 111603.3 thousand soums.

Table 5

Indicators of economic efficiency of production of cherry rootstocks per 1 hectare of land area (thousand soums)

№	Indicators	Traditional	*In vitro*	Difference (+,-)
1	Rootstock exit	75000	75000	
2	Production costs	25778,7	53675,4	27896,7
3	Production cost	380,4	715,7	335,29
4	Selling price	0,80	2,5	1,7
5	Income	48000	187500	139500
6	Profit	22221,3	133824,6	111603,3
7	Profitability	86,2	249,3	163,1
8	Economic effect		111603,3	
9	Resource saving ratio		2,08	
10	Innovation gross growth rate		0,83	
11	Reproduction factor	60%	95%	

For the organizational and economic assessment, the cultivar with the highest multiplication factor was selected, while taking into account the percentage of rooting and survival of microshoots rooted in vitro (95%) and ex vitro (60%).

One of the most important mechanisms contributing to the growth of the efficiency of management of innovative activities of enterprises is the development of a resource conservation program. This is especially important for the agricultural sector due to its specific features and in connection with the limited land and water resources. Therefore, a significant result of the introduction of innovative technologies, which characterizes their effectiveness, is the conservation of production resources. As a result of the introduction of resource-saving technologies, it manifests itself in two forms: either as an increase in the output of final products from a certain volume of resources, or as a reduction in the consumption of production resources for the manufacture of a certain volume of final products.

Both of these forms of resource conservation express the economy of production resources. Here it is worth making clarifications: saving resources is associated with a decrease in their costs compared to the costs of functioning technologies, and saving is associated with the introduction of resource-saving technologies.

Consequently, to assess the effectiveness of resource-saving technologies, the results of their implementation (saving resources) should be compared with the actual results of using resources in the technologies they replace.

Proceeding from this, in determining the economic efficiency of the technology for growing sweet cherry rootstocks in vitro, the resource saving coefficient ($R_{r.s.}$) was calculated, which is determined by the ratio of the total costs of innovative technologies to the total costs of traditional technologies.

$$R_{r.s.} = \frac{PC_{inn.}}{PC_{tr.}}$$

$R_{r.s.}$ - coefficient of resource saving;

$PC_{inn.}$ - production costs of innovative technologies;

$PC_{tr.}$ - production costs of traditional technology.

When growing cherry rootstocks using the in vitro method, the resource saving factor was 2.08, which indicates higher costs for obtaining cherry rootstocks in vitro than with the traditional method. But the result of the introduction of resource-saving technologies in our case manifested itself in the form of an increase in the output of final products from a certain amount of spent resources, i.e. this technology is effective from the point of view of resource conservation by increasing the proceeds from the sale of this seedlings.

To do this, we calculated another indicator reflecting the effectiveness of innovation, the coefficient of the cumulative increase in innovation (Ki), which is determined by the ratio of the increase in gross profit to the gross profit from innovative technology. This indicator shows how much the gross profit from the introduction of innovative technology has increased.

$$K_i = \frac{\Delta GP}{GP_{inn}}$$

The gross innovation growth rate was 0.74. That is, due to the fact that the increase in gross profit from innovation was more than 50%, it is possible not only to cover the high costs of growing rootstocks in vitro, but also to provide additional marginality. As mentioned above, resource conservation occurs due to additional profit.

4. CONCLUSIONS

Based on the studies carried out, it can be concluded that the microclonal method of vegetative propagation of sweet cherry has established itself as highly effective and economically justified.

As a result of the analysis of the experiments, the following advantages of microclonal propagation of cherry rootstocks in vitro have been revealed:

- the possibility of rapid reproduction and obtaining thousands of seedlings in a short time;

- work in laboratory conditions, in an absolutely sterile environment, where a product made healthier from various diseases and viruses is produced;

- the ability to grow plants in any quantity all year round, regardless of the season and weather conditions;

- saving the time required for growing seedlings in the traditional way, in particular, with microclonal propagation from seeds, it is possible to accelerate the transition from the juvenile to reproductive phase of development, which will shorten the production period to 1-3 years, when propagating from cuttings, the rooting period is reduced to 1 -2 years old;

- implementation of constant monitoring of each type of plant and operational change of the environment, if necessary;

- the possibility of long-term preservation of the surplus product in vitro;

- complete preservation of the genetic properties, quality and characteristics of the grown product;

- easy and fast propagation of plants in vitro, which is impossible or costly to propagate in the traditional way;

- savings in areas for planting material.

Using a healthy (virus-free) planting material for planting gardens in combination with the optimal scientifically grounded zoning of crops and varieties, scientifically grounded schemes for placing plants in plantations, as well as adhering to a high level of agricultural technology, you can get a higher yield and an earlier entry of plants into the marketable period. fruiting, thus providing a quick return on investment and getting a higher income compared to using conventional planting material.

It is important to note here that the clonal micropropagation method has great advantages; at the same time, it is a laborious and expensive procedure, therefore, its application on an industrial scale is currently limited. The technology of in vitro clonal propagation at the laboratory level has been developed in the world for more than 2,400 plant species, but the method is most often used for plants that are difficult to reproduce by conventional methods, as well as for solving problems related to breeding or basic research.

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