UDK 635.925:582.711.711:631.53.03

ELEMENTS OF INNOVATIVE TECHNOLOGY FOR SPIRAEA X VANHOUTTEI (BRIOT) ZAB. SEEDLINGS GROWING IN AN INDUSTRIAL NURSERY

O.E. Klimenko, N.I. Klimenko

Nikita Botanical Gardens - National Scientific Center, Yalta

Introduction

Vegetative propagation is important for plant introduction. In new soil and climatic conditions introduced plants can later come into generative stage, give defective seeds, fruit rarely or poorly, or even not bear fruits at all that makes it impossible to use seed propagation method. Besides, seed propagated plants do not always inherit decorative features that attract introducers and designers. Therefore, vegetative propagation is the most effective way to get planting material in sufficient quantity. It can be obtained from maternal plants, before the generative age. N.F. Dovbish [4] noted that it contributes to the further aromorphosic development of introduced species as the organisms get new opportunities for development of the environmenalt resources.

In soil and plant rhizosphere microorganisms capable to synthesize substances that improve nutrition, stimulate plant growth and development are common. These substances affect various aspects of plant life, regulate particular stages of their growth and development and make possible qualitative changes in the economically valuable plant features [1]. Use of active microorganisms (MO) strains in plant horticulture is based on symbiosis and associations of plant and MO. MO have complex effect on a plant: improve nitrogen nutrition of plants through nitrogen fixation, phosphorus nutrition – through phosphatmobilization [7, 9]; stimulate growth through secretion of growth regulators - indoleacetic acid, gibberellins, auxins et al. [8, 11]; improve the state of plants by allocating other biologically active substances - vitamins, enzymes, antiseptics and others [12].

At present, based on active bacterial strains microbial products (MP) have been created and successfully used for cereals, vegetables and forage crops [2]. MP are used for treating seed at sowing or soil bacterization during the growing season. They are environmentally friendly and help to decrease the use of fertilizers by 20 - 30%.

More rarely MP are used in ornamental plant growing. There is some information about using Fitostimofos preparation based on *Agrobacterium radiobacter* strain 2258 CMF to improve the survival rate of annual and perennial flower crops seedlings [1] and increase seed germination of medicinal and ornamental plants [3]. In fruit crops nursery Fosfoenterin was used to increase the output of peach and sweet cherry planting material [5]. Thus, using of useful microorganisms for increasing of growth intesity, plant nutrition and adaptation to abiotic stresses has great practical importance. Literature sourses have little information about MP use for ornamental shrubs seedling growing. So, investigations of active bacterial strains with stimulating and growth-regulatory effects that let to increase the output of ornamental crops planting material are important.

The aim of our researches was to evaluate the influence of MP on soil agrochemical properties, rooting rate, growth, plant development and output of standard seedlings *Spiraea x vanhouttei* (Briot) Zab. [13] in nursery industry.

Materials and Methods

Objects of the studies were *S. x vanhouttei* plants, MP and soil agrochemical properties. Investigations were carried out in the nursery in NBG (Department of Steppe

Plant Growing) on the small field plots in 2012 - 2014. Experimental varianst were MP: Azotobacterin (*Azotobacter chroococcum* 10702) with nitrogen-fixing properties. Fosfoenterin that is based on *Enterobacter nimipressuralis* 32-3strain and transforms hardly accessible phosphates into plant available state and Complex microbial preparations (CMP) that has plural effect on plants. This Complex is an equal parts mixture of nitrogen fixer - Diazofit (*Agrobacterium radiobacter* 204), Fosfoenterin and Biopolicid created on the basis of *Paenibacillus polymyxa* P strain - antagonist of pathogenic micromycetes.

MP have been developed and provided by the researchers of Microbiology Department at the Institute of Crimea Agriculture. Titer of preparations was 7 - 10 billion. CFU / ml. Working solutions of MP suspensions were prepared in the day of planting through diluting suspensions in 100 times with tap water. For better survival rate and bacteria nutrition on spirea cuttings combined use of MP and organo-mineral fertilizer "Component 2" (0.5% solution), which consisted of macro- and micronutrients set, humates and adhesive were investigated. This drug, according to the developers (NIC "Ecology"), also has immunomodulatory properties. Untreated plants were used as a control.

Lignified cuttings of *S. x vanhouttei* 20 cm length were soaked in tap water for 24 hours before planting, then their basal ends immersed for 1-2 cm in the mixture of MP and Component 2 solutions for 20 minutes. For comparison, Zircon preparation was used as an analogue as it is widely used in ornamental horticulture as an immunomodulator and root stimulater. Before planting cuttings were soaked in 0.01% solution of Zircon for 14 hours.

Cuttings were planted in the first field of nursery in March, 10 - 15 samples per an account plot. Plot area was 0.7 - 1.0 m^2. Experiments had three times replications. Variants` location was rendomized within a row of nursery. Experience was designed for a two-year planting of seedlings. Cultural care of studied plants was carried out in accordance with accepted agricultural practices in the industrial nursery of Steppe Plant Growing Department at NBG.

Account of plants growth and state was carried out according to the procedure [6]. Seedlings quality was evaluated according to GOST 26869-86.

Soil in the plot is southern carbonate plantaged light clayey chernozem on red-brown Pliocene clay. Content of nutrients in the soil before the experiments was low in nitrate nitrogen (5.9 mg / kg) and mobile forms of phosphorus (8.5 mg / kg) and optimal in potassium (325 mg / kg). Humus content ranged from 3.02 to 3.24%, depending on soil and plot.

Soil samples for analysis were collected annually during the period of intensive shoot growth (late July - early August) according to the experiment variants in the layer 0 - 40 cm. Content of phosphorus mobile forms and potassium in soil determined according to Machigin (DSTU 4114-2002), nitrate nitrogen - potentiometrically GOST 26951-86, organic matter - according to Tiurin (DSTU 4289: 2004). Statistical analysis of the results was carried out by the program ANOVA. The level of significance was set at 5% ($p<0.05$).

Results and discussion

Content of nitrate nitrogen and available phosphorus in the soil under *S. x vanhouttei* seedlings was low in control and all other variants and potassium content was high (Table. 1).

Table 1

Content of humus, nitrate nitrogen, mobile forms of phosphorus and potassium in the soil (layer 0 -40 cm) under *S. x vanhouttei* seedlings after cuttings treatment with MP and Component 2, nursery NBG, the average for two years (2012 - 2013).

Variants	N – NO3	P2O5	K2O	Humus, %
	mg/kg soil			
Control	8.1±1.5	8.5±4.2	310±45	2.85±0.20

Azotobacterin	5.6±0.8	10.5±2.9	362±16	2,78±0.29
Azotobacterin + Component 2	5.6±1.0	18.9±7.9	328±94	2.44±0.24
Fosfoenterin	7.6±2.2	12.2±3.4	322±24	2.75±0.14
Fosfoenterin + Component 2	7.0±3.5	21.9±3.5	306±116	2.76±0.28
CMP	8.2±2.3	14.9±4.3	345±50	2.70±0.23
CMP + Component 2	4.7±0.1	15.0±5.0	362±66	2.66±0.24
Optimal [10]	15–20	28–38	211–270	–

On average, two years applied MP and their mixtures with Component 2 helped to reduce nitrate nitrogen in the soil under *S. x vanhouttei* seedlings and only CMP had not changed it, but Fosfoenterin and its mixture with Component 2 - reduced nitrate nitrogen to less extent than other preparations. This is probably due to the increase in the number of established cuttings under the influence of MP and increased consumption of soil nitrogen by rapidly growing seedlings of *S. x vanhouttei*. Therefore, growing ornamental plants using MP and under a low content of soil nitrate nitrogen in the first nursery plot needed low dose of nitrogen dressing to improve nutrition of growing seedlings.

Using of each MP resulted in greater amount of mobile phosphorus in soils. The largest increase of this element content occurred under using a mixture of Fosfoenterin and Component 2 in 13.4 mg / kg that corresponds to 60 kg / ha P_2O_5.

Content of exchangeable potassium in the soil was increased when using most of MP, significantly using Azotobacterin and CMP in combination with Component 2 in 52 mg / kg or 17% of control.

Humus content in the soil was high enough in control. Application of MP and its mixtures with Component 2 gave a tendency to decrease humus content in the soil. It was less when using Azotobacterin, Fosfoenterin and its mixtures with Component 2. However, any significant difference between control and other variants haven`t been found.

The study results in rooting of *S. x vanhouttei* cuttings treated with MP indicated that their survival rate considerably varied in control – from year to year, from 30 to 67% of the planted cuttings and the average for three years was 48% (Table. 2).

Table 2

Survival rate of *S. x vanhouttei* cuttings in NBG nursery (% of planted) under using MP and Component 2, 2011 - 2013.

Variants	2011	2012	2013	Average	% of the control
Control	67±5	30±10	47±12	48.0	100
Zircon	67±7	27±3	–*	47.0	97.9
Azotobacterin	53±2	27±7	60±12	46.7	97.3
Azotobacterin + Component 2	50±3	43±7	60±15	51.0	106.2
Fosfoenterin	43±3	37±9	67±12	49.0	102.1
Fosfoenterin + Component 2	67±7	80±10**	70±2	72.3	150.6
CMP	56±9	33±3	63±13	50.7	105.6
CMP+ Component 2	76±4	85±5**	60±15	73.7	153.5
Notes: * in 2013 Zircon haven`t been used, ** significant difference with the control					

Applied MP changed the survival rate in different ways, and the intensity of their influence depended on the year. So, in 2011, when the survival rate of cuttings was high in control, most preparations either unchanged or somewhat reduced it and only mix of CMP and Component 2 stimulated significant increase of accustomed cuttings. In 2012 and 2013 treatment with most of preparations enhance the survival rate of cuttings, especially in 2012, when it was low in control. This is probably due to increase of plant resistance to adverse environmental factors during the period of rooting (lack of moisture, high temperature, etc.).

In the same year, Component 2, as an immunomodulator, significantly enhanced MP effect. For example, using of Fosfoenterin increased cuttings survival by 7% of planted ones, and adding thereto Component 2 – by 50% compared with the control. Under CMP applying this increase was 3 and 55%, respectively. In 2013 this trend was true for Fosfoenterin. An average of three years studies impact of Zircon and Azotobacterin on the rooting of *S. x vanhouttei* cuttings corresponded to the level of control. Fosfoenterin and CMP slightly increased the rate (by 2 - 6%) respectively to control. The combined use of these preparations and Component 2 stimulatted significant increase in the number of rooted plants by 51 - 54% compared with the control.

The results show that MP affected *S. x vanhouttei* growth and shoot formation in the nursery (Table. 3).

Variants	Number of annual shoots	Total length of annual shoot, cm	The average length of shoot, cm
Control	2.0	15.0	7.5
Azotobacterin	2.7	73.7	27.3
Azotobacterin + Component 2	3.8	70.0	18.4
Fosfoenterin	3.0	60.7	20.2
Fosfoenterin + Component 2	4.3	74.3	17.3
CMP	3.0	27.0	9.0
CMP + Component 2	3.7	73.2	19.8
НСР 05	1.0	35.0	9.2

Thus, using of MP increased number of shoots formed. The highest number of shoots formed under the influence of Fosfoenterin and Component 2 combination which exceeded control in more than half.

Total length of shoots growth was low in control and increased under the influence of MP. Adding of Component 2 to Fosfoenterin and CMP enhanced MP effect on the overall shoot growth; especially significantly Component 2 increased CMP effect - by 2.7 times compared with using CMP only. Total shoot growth increased not only by increasing their number, but also due to increasing a shoot length under MP use. The average shoot length was most significantly increased by Azotobacterin, by 20 cm, compared with the control. Addition of Component 2 to this preparation, as well as to Fosfoenterin had little effect on shoot length, but significantly stimulated shoot growth when used in composition with CMP, by 12.3 cm compared with the control.

In the second year after planting flowering plants were observed (Fig. 1, 2).

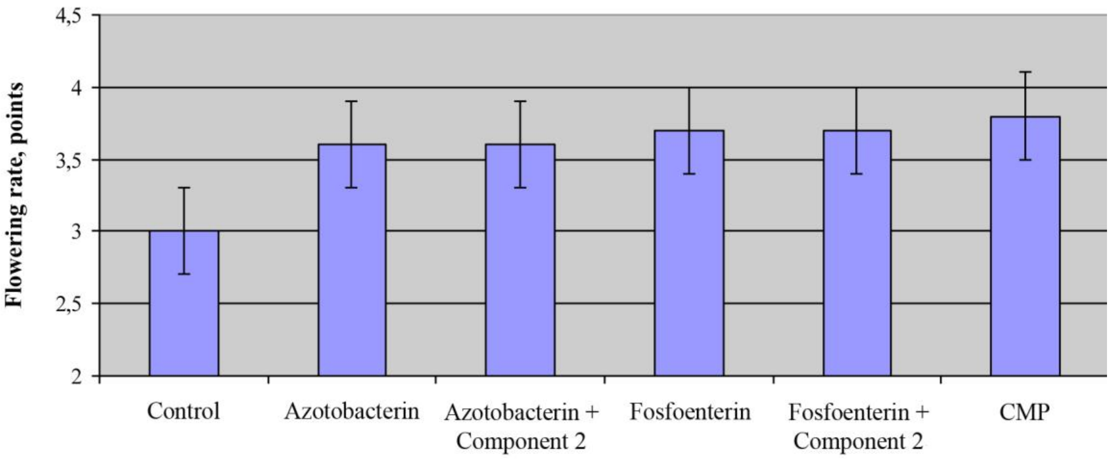

Fig. 1 Effect of MP and Component 2 on flowering rate of *S. x vanhouttei* seedlings in the second year after planting (2013)

Flowering was middle (3 points). All MP increased the intensity of flowering, Fosfoenterin and CMP significantly by 0.7 - 0.8 points or 23 - 27% relatively to control. Therefore, MP affected the rate of flower buds formation in *S. x vanhouttei*.

A B

Fig. 2 Rate of flowering in *S. x vanhouttei*: A – control, B – Fosfoenterin + Component 2, NBG-NSC nursery, 2012.

The main indicator of nursery productivity is the output of standard seedlings. An average index of three year researches for *S. x vanhouttei* was low in control - 35.3% of planted cuttings with changes from year to year from 10 to 51% (Table. 4).

Table 4

***S. x vanhouttei* standard seedlings output in NBG nursery under MP and Component 2 using, 2012 - 2014.**

Variants	2012	2013	2014	An average for three years	
	% of planted cuttings			% of planted cuttings	% of control
Control	51	10	45	35.3	100
Zircon	46	20	not found	33.0	94
Azotobacterin	53	20	60	44.3	126
Azotobacterin + Component 2	50	40	60	50.0	142
Fosfoenterin	42	20	57	39,7	112
Fosfoenterin + Component 2	51	70	70	63.7	180
CMP	53	10	55	39.3	111
CMP + Component 2	57	60	60	59.0	167
NCR 05	Fф < F05	12	15	–	–

Minimum output of seedlings was in control in 2013; however, under MP using maximum increase of seedlings output by 200 - 700% of the control was noticed this year. In the other two, the output of *S. x vanhouttei* seedlings was approximately the same, but MP increased the output of seedlings in 2014 more significantly. This may indicate the influence of particular year environmental conditions on the rate of bacteria development and intensity of their effect. On average for three years the best combination, which obtained maximum additional amount of standard seedlings, was Fosfoenterina combined with Component 2, the excess relative to the control was 80%. High excess amount compared to seedlings obtained from control was also in the variant with CMP combined with Component 2 - by 67% compared with the control.

All applied MP exceeded Zircon on the effects on *S. x vanhouttei* standard seedlings output in the nursery.

Conclusions

1. It was found that for *S. x vanhouttei* two-year seedlings growing in a nursery treatment of lignified cuttings basal ends by Fosfoenterin and CMP combined with Component 2 in spring before planting was favourable for increasing amount of phosphorus mobile forms and potassium in the soil, and stimulated seedlings growth and shoot formation, increased the rate of flowering. Their influence was more intensive than standard growth stimulator Zircon.

2. It has been demonstrated that using of Fosfoenterina and CMP in combination with Component 2 increased number of rooted *S. x vanhouttei* cuttings by 50% and number of standard seedlings by 67 - 80% compared with standard technology. These combinations can be recommended for wide production testing in nurseries in the southern chernozems.

3. On soils with low content of nitrogen mobile forms under MP using nitrogen fertilizing for plants is required in the first field of nursery. To avoid loss of humus content an introduction of field with green manure (busy couple) or perennial grasses to the crop rotation in a nursery is needed to increase content of organic matter in soil.

References

1. Voinilo N.V., Timofeeva V.A., Suhovitskaia L.A., Safronova G.V., Melnikova N.V. Vlianie biopreparatov fitostimos I rizobakterin na dekorativnye kachestva tsvetochnyh kultur // Sovremennoe sostoianie i perspektivy razvitia mikrobiologii i biotehnologii: materialy 7 mezhdunarodnoi konferentsii (Minsk, 31 maia – 4 iunia 2010). – Minsk: Belaruskaia navuka, 2010. – S. 227 – 228.

2. Volkogon V.V., Nadkernichna O.V., Kovalevska T.M., Tokmakova L.M., Kopilov E.P. Mikrobni preparaty u zemlerobstvi. Teoria i praktika / Za nauk. red. V.V.Volkogona – K.: Agrarna nauka, 2006. – 312 s.

3. Golubeva V.S., Dishuk N.G., Suhovitskaia L.A., Safronova G.V., Melnikova N.V. Primenenie biologicheskogo preparata fitostimos v lekarstvennom I dekorativnom rastenievidstve // Sovremennoe sostoianie i perspektivy razvitia mikrobiologii i biotehnologii: materialy 7 mezhdunarodnoi konferentsii (Minsk, 31 maia – 4 iunia 2010). – Minsk: Belaruskaia navuka, 2010. – S. 230 – 231.

4. Dovbysh N.F. Priemy uskorennogo razmozhenia introdutsirovannyh rastenij steblevymi cherenkami // Introduktsia i akklimatizatsia rastenij. – 1999. – Vyp. 32. – S. 71 – 79.

5. Klimenko O.E., Klimenko N.I., Kameneva I.A., Kulikova T.D. Vozdeistvie mikrobnyh preparatov na rost i razvitie plodovyh sazhentsev // Silskogocpodarska mikrobiologia: Mizhvidomch. temat. nauk. zb. – Chernigiv: Chernigivskij CNTEI, 2008. – Vyp. 8. – S. 90 – 103.

6. Kondratenko P.V., Bublik N.A. metodika provedenia polevyh issledovanij s plodovymi kulturami. – K: Agrarna nauka, 1996. – 95 s.

7. Muromtsev G.S., MZarshunova G.N., Pavlova V.F., Zolnikova N.V. Rol pochvennyh mikroorganizmov v fosfornom pitanii rastenij // Uspehi mikrobiologii. – 1985. – T. 20, № 2. – S. 174 – 198.

8. Patyka V.F., Omelianets T.G. Ekologichni osnovi zastosuvannia biologichnih zasobiv zahistu roslin iak alternative himichnim pestitsidam // Agroekol. Zhurn. – 2005. – № 2. – S. 21 – 24.

9. Patyka V.P., Tihonovich I.A., Filipev I.D., Gamaiunova V.V., Andrusenko I.I. Mikroorganizmi ta alternativne zamlerobstvo. – K.: Urozhai, 1993. – 176 s.

10. Rekomendatsii po primeneniu udobrenii v plodovyh i jagodnyh nasazhdeniah. Tsentralnyi institute agrohimicheskogo obsledovania selskogo hoziastva (CINAO). – M., 1983. – 42 s.

11. Chaikovska L.O., Gamaiunova V.V. Fosfatmobilizuiuchi bakterii ta ih vpliv na produktivnist roslin // Biologichni nauki i problem roslinitstva / Zb. nauk. prats Umanskogo derzh. agrouniversitetu. – Uman, 2003. – S. 220 – 225.

12. Smith S.E, Read D.J. Mykorhizal Symbiosis: 2nd ed. – London, Academic Press, 1997. – 605 p.

13. The plant list. A working list of all plant species. – http://www.ipni.org/ipni/idPlantNameSearch.do?id=742599-1&back_page=. – Searched on 14 November 2014.

Klymenko O.E., Klymenko N.I. Elements of innovative technology of seedlings Spiraea x vanhouttei (Briot) Zab. growing in industrial nursery // Works of the State Nikit. Botan. Gard. – 2014. – V. 139 – P. 170 – 176.

The data of triennial researches in using of various microbal preparations and organic and mineral fertilizer growing Spiraea x vanhouttei (Briot) Zab. seedlings in industrial nursery have been presented in the article. It was found that the introduction of these elements into the Spiraea x vanhouttei seedlings growing technology increased survival ability of ligneous cuttings, improves their growth, led to the formation of laterals, intensified flowering and increased the standard seedlings output. The best results according to all these parameters were obtained by Fospfoenterin and Complex of microbal preparations in combination with Component 2 treating ligneous cuttings before planting.

Keywords*: microbal preparations, Spiraea x vanhouttei (Briot) Zab., industrial nursery, technology elements.*