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ORGANIC FERTILIZER'S ROLE IN THE IMPROVEMENT OF SOIL MICROFLORA AND BIOMETRIC VALUES IN FRUIT CROPS

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SUMMARY

Intensive farming has adversely affected soil fertility and ecosystems due to excessive chemicalization in the Kazakhstan Republic. The soil lacks organic matter, leading to declining fertility. Biologization has emerged to address this issue as a solution to improve soil fertility and enhance the growth and development of crop plants. This study aimed to determine the impact of organic fertilizers on the composition of soil microflora and biometric values in cultivating stone and pome fruits. The use of microbiological methods analyzed soil microflora and physiological bacterial groups. The research's novelty lies in developing a technology that utilizes organic fertilizers and biological products to improve the activities of soil microorganisms and plant roots' nutrition. Organic fertilizers 'Bio juice Energy Plus' and 'Agroflorin' positively influenced beneficial bacteria and fungi and significantly contributed to soil enrichment. Organic fertilizers and biological products support soil biological processes and replenish the nutrients, promoting sustainable agricultural productivity and reducing chemical dependency. The current study emphasizes the potential of organic fertilizers and natural products in enhancing soil fertility and supporting plant growth in Kazakhstan. Biologization also offers an eco-friendly approach to agricultural practices, ensuring agronomic productivity and the long-term good health of soils.

Keywords: Organic fertilizers, biological products, chemicalization, biologization, soil microorganisms, soil fertility, plant growth and development, stone and pome fruits

Key findings: The applied organic fertilizers, i.e., Agroflorin, Alkaral, and Bio juice Energy Plus, have proven efficient in improving the qualitative and quantitative composition of microbial association of soils under fruit crops. Research revealed that organic fertilizers boost the beneficial microorganisms and positively influence most physiological indicators in the apple tree culture compared with the control version. With positive results, the D-4 variant using organic fertilizer 'Bio juice Energy Plus' led and performed better.

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INTRODUCTION

In agriculture, crop and fruit production mainly depends on the widespread use of fertilizers and pesticides globally. In intensive farming, these chemicals' addition to the soil and environment sought to enhance crop production and meet global food demand. Fertilizers and pesticides have also become essential inputs for ensuring food safety. However, inorganic fertilizers and pesticides cause undesirable impacts on crops and the environment (Duddigan *et al*., 2020). These chemicals also remain in the soil and surroundings for a long time and have various residual effects in the form of biotic and abiotic factors.

The abovementioned stress conditions degrade the soil, adversely affecting other microflora organisms, the environment, and human health (Gu *et al*., 2019; Bastaubayeva *et al*., 2023). The undesirable properties of fertilizers and pesticides have compelled the search for an option that positively affects soil fertility, allowing for sustainable agriculture that gains positive attention from the farming community – organic fertilizers (Kumar and Madhu, 2016; Gusev *et al.,* 2022; Makenova *et al*., 2023). At the present stage of agriculture development in our country, with an enhanced demand for crop production volume, the soil deteriorates, and fertility problems arise on a large scale.

As a result, the question arises to address those soil fertility problems to achieve zero land degradation while enhancing and maintaining the organic matter content in soils. Globally, researchers are trying to raise the need for an annual increase in soil organic carbon of 0.4% (4 ppm) with an international goal of promoting and achieving sustainable soil management and climate change mitigation (Minasny *et al.*, 2018). The reproduction of organic matter in the soil is the basis of its existence as a natural resource.

A set of agricultural practices ably enriching the soil with plant biomass is necessary to reproduce and renew soil organic matter and optimize its qualitative characteristics. Such techniques include applying organic fertilizers (litter manure, peat, compost, and straw with nitrogen fertilizers) and sowing perennial grasses, stubble crops, and green dung (Shaji *et al*., 2021). It was also apparent that long-term use of biological products and agricultural practices reflected more in the qualitative characteristics of soil organic matter than in its gross content. In the soil, reduced mineralization of humic substances can happen by minimizing the mechanical tillage and selecting plants for crop rotation.

The stability degree of soil organic matter increases under perennial grasses and sharply decreases in black fallow (Masyutenko, 2019). Using modern methods expressed the intensification of microbiological processes and the mobilization of fertility elements, the efficiency of soil cultivation, and the root rhizosphere of fruit crops (Ivanov *et al*., 2017). The bacterial fertilizers' utilization enhances the microbiological processes in the soil, contributing to nutrient accumulation in an accessible form for crop plants (Zhang *et al*., 2019; Sivojiene *et al.*, 2021). After analyzing the total carbon, nitrogen, and nitrogen-15 content in organic fertilizers, soil pools, canopy biomass, and tree leaves, past studies revealed that using plants as a source of carbon and nitrogen improved soil quality and fertility, hence increasing crop productivity (Assefa and Tadesse, 2019).

Studies on the interaction of plants and microorganisms are currently of particular relevance because the sharp reduction in mineral and organic fertilizers and plant protection products' use in agriculture creates the need to search for additional sources of nitrogen nutrition for crop plants (Hoagland *et al*., 2006; Alhassan *et al*., 2019, 2021; Sarkar *et al*., 2021). Developing a technique for applying biological products is ongoing at the KazSRI of Horticulture LLP. This study aims to investigate the use of organic fertilizers and natural products, such as, Agroflorin, Alkaral, and Bio juice Energy Plus, and regulate the activity of beneficial soil microorganisms in cultivating pome and stone fruits.

MATERIALS AND METHODS

A study of the soil proceeded with using liquid organic fertilizers and preparations, i.e., Agroflorin, Bio juice Energy Plus, and Alkaral, under pome and stone fruit crops grown in the District Talgar, Almaty region, Republic of Kazakhstan. For microbiological analysis, collecting the soil samples for all variants followed the methodology of GOST-17.4.4.01-83. The statistical data and results compiled were according to the GOST-R ISO 10576-1 standard. Before planting pome and stone fruit crops, seedlings sustained solutions of the above-listed liquid organic fertilizers and biological products at the concentrations recommended by manufacturers. Applying fertilizers in appropriate concentrations during the vegetative growth of plants transpired once near the trunk, twice as a root top dressing, and twice as a foliar top dressing.

Purpose of the study

Determination of the microflora composition and the main physiological groups of soil bacteria (nitrogen-fixing, Pseudomonas, Bacillus, aerobic bacteria, and fungi) after growing the seedlings of apple, plum, and apricot using organic fertilizers was the goal. Accounting for the total number of microorganisms ensued on meat-peptone agar on starch-ammonia agar (AA), with actinomycetes and bacteria from 10 thousand dilutions taken into account. The dilution of the soil suspension was 10-3-10-5 degrees, the volume of the inoculum suspension per Petri dish was 100 µl, the recording time was 3–7 days, and cultivating in a thermostat at a temperature of 1 °C ± 29 °C. Pseudomonas bacteria detection occurred on the agar medium King B. The Azotobacter presence established in soils continued by seeding on Ashby's medium. The fungal flora accounting followed by sowing the soil suspension in the acidic environment of Czapek. The total microbial count (TMC) attained validity on meat-peptone agar, Czapek-Dox, Ashby, and starch-ammonia agar. Determining the number of nitrogen-fixing bacteria employed the method of agarized nitrogen-free medium (Zvyagintsev, 1991; Erismana, 2004; Lin *et al*., 2019).

RESULTS AND DISCUSSION

The data results indicated that various organic fertilizers and biological products enhanced the quantitative composition of microorganisms of different systematic groups in the soil. By growing pome and stone fruit crops using organic fertilizers, i.e., Agroflorin, Alkaral, and Bio juice Energy Plus, the quantitative composition of ammonifiers, aerobic pseudomonad bacteria, amylolytic bacteria, mold fungi, and aerobic nitrogen fixers underwent study using microbiological analysis in the soil.

The main physiological groups of soil microorganisms are visible in Figure 1. Various groups of soil microorganisms served as indicators. A statistical compilation of the obtained data ensued using the 8.0.14.0*80.00 mm version Scan_500® colony counter. The existing study showed the composition of soil micro biocenosis of the apple cultivar 'Golden Delicious' studied on the MPA universal medium. The number of bacteria dominated in the experimental variants, i.e., in the Agroflorin (G-2 variant) (2.4 ± 0.1 10-6 CFU/g), Bio juice Energy Plus (G-4 variant) (2.4 ± 0.1 10-6 CFU/g), in Alkaral (variant G-3) (2.2 ± 0.1 10-6 CFU/g) compared with the control variant (1.03 ± 0.1 10-6 CFU/g) (Figure 1).

The population values of the Pseudomonas bacteria groups on the King B solid medium in the Bio juice Energy Plus (G-4 variant) was 4.0 ± 0.01 10-6 CFU/g, in the Agroflorin variant (2.2 ± 0, 1·10-6 CFU/g), and in the control variant (1.2 ± 0.01·10-6 CFU/g).

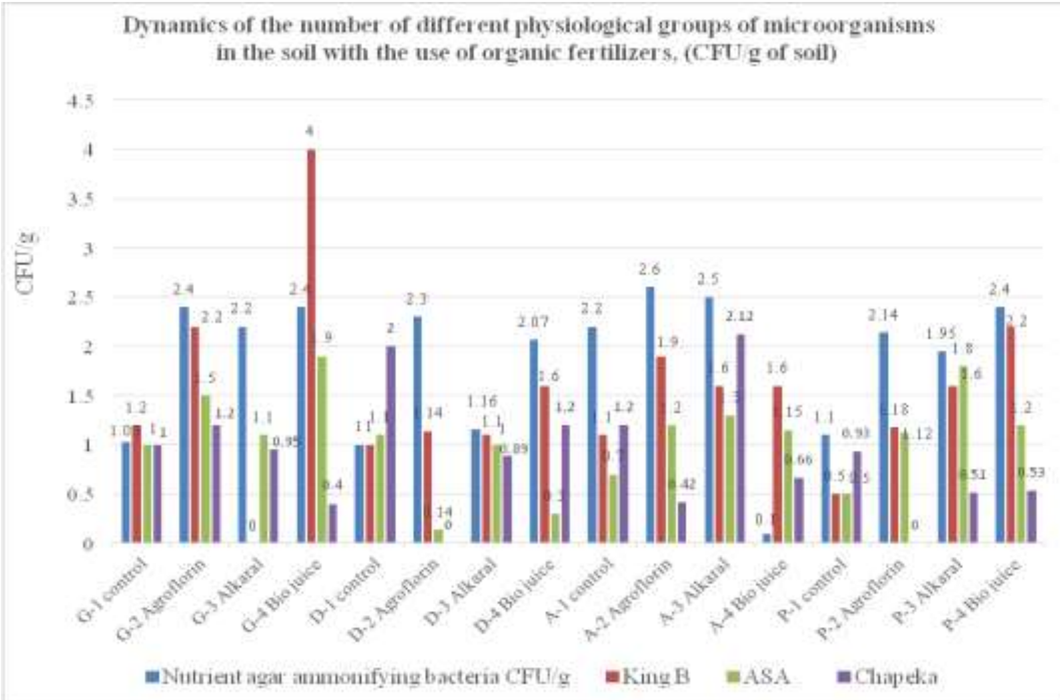

Figure 1. Dynamics of the number of various physiological groups of microorganisms in the soil under the apple cultivar 'Golden Delicious' with the use of organic fertilizers (CFU/g of soil).

However, in the Alkaral fertilizer variant, the Pseudomonas growth was absent. Regarding amylolytic bacteria, in the variant of Agroflorin (G-2) by 0.5 ± 0.01 10-5 CFU/g, and in the variant Alkaral (G-3) this bacterium was higher by 0.1 ± 0.01 10-6 CFU/g than the control (D-1) (1.0 ± 0.01 10-6 CFU/g) (Table 1). Amylolytic bacteria on starch-ammonia agar medium were evident in the variant of Bio juice Energy Plus (G-4) (0.9 ± 0.01 10-5 CFU/g).

The Golden Delicious apple tree soil with biological products and organic fertilizers, such as Agroflorin, Alkaral, and Bio juice Energy Plus, the acid-resistant bacteria count significantly exceeds the control variant number. Such an increased content of microorganisms correlates with the release of amino acids and carbohydrates (sugars, starch) on the surface of the roots, which are substrates for these microorganisms. On Czapek's agar medium in the variant with the use of Agroflorin (G-2) in the soil of Golden Delicious apple trees, the composition of soil fungi exceeds by 0.2 ± 0.1 10-4 CFU/g than the control (G-1) by 1, 0 ± 0.1 10-4 CFU/g, while in the Alkaral variant (G-3), this indicator was 0.95 ± 0.1 10-4 CFU/g and in the Bio juice Energy Plus variant (G-4) the soil fungi value was 0.4 ± 0.1 10-4 cfu/g (Table 1). In the rhizosphere soil, the microflora performs essential ecological functions to decompose the organic compounds, and it is also an antagonist for pathogenic organisms, providing natural protection.

By studying the number of bacteria in soils in the variants of the apple cultivar tree Damira, it was apparent that, on average, the number of ammonifying bacteria was higher in the experimental variants than in the control variant (D-1) (1.0 ± 0.1 10-6 CFU/g). However, in the variant Agroflorin (D-2), this indicator was 2.3 ± 0.1 10-6 CFU/g, in the variant Bio juice Energy Plus (D-4) (2.07 ± 0.1 10- 6 CFU/g), and Alkaral variant (D-3) (1.16 ± 0.1 10-6 CFU/g). Pseudomonas group bacteria showed higher concentrations in the D-4 variant-Bio juice Energy Plus (1.6 ± 0.1 10-6 CFU/g), followed by Agroflorin (D-2) variant (0.14 ± 0.1 10-6 CFU/g), and Alkaral variant (D-3) (0.1 ± 0.1 10-6 CFU/g).

Table 1. Dynamics of the number of various physiological groups of microorganisms in the soil with the use of organic fertilizers (CFU/g of soil).

Fruit type and cultivars	Variants	Microorganisms (CFU/g)			
		Nutrient agar ammonifying bacteria	King B Pseudomonads	ASA Amylolytic bacteria	Chapeka moldy fungi
Apple (Golden Delicious)	G-1 Control	$1.03 \pm 0.1 \cdot 10^{-6}$	$1.2 \pm 0.01 \cdot 10^{-6}$	$1.0 \pm 0.01 \cdot 10^{-6}$	$1.0 \pm 0.1 \cdot 10^{-4}$
	G-2 Agroflorin	$2.4 \pm 0.1 \cdot 10^{-6}$	$2.2 \pm 0.1 \cdot 10^{-6}$	$1.5 \pm 0.01 \cdot 10^{-5}$	$1.2 \pm 0.1 \cdot 10^{-4}$
	G-3 Alkaral	$2.2 \pm 0.1 \cdot 10^{-6}$	0.00	$1.1 \pm 0.01 \cdot 10^{-6}$	$0.95 \pm 0.1 \cdot 10^{-4}$
	G-4 Bio juice Energy Plus	$2.4 \pm 0.1 \cdot 10^{-6}$	$4.0 \pm 0.01 \cdot 10^{-6}$	$1.9 \pm 0.01 \cdot 10^{-5}$	$0.4 \pm 0.1 \cdot 10^{-4}$
Apple (Damira)	D-1 Control	$1.0 \pm 0.1 \cdot 10^{-6}$	$1.0 \pm 0.1 \cdot 10^{-6}$	$1.1 \pm 0.1 \cdot 10^{-6}$	$2.0 \pm 0.1 \cdot 10^{-3}$
	D-2 Agroflorin	$2.3 \pm 0.1 \cdot 10^{-6}$	$1.14 \pm 0.1 \cdot 10^{-6}$	$0.14 \pm 0.01 \cdot 10^{-6}$	0.00
	D-3 Alkaral	$1.16 \pm 0.1 \cdot 10^{-6}$	$1.1 \pm 0.01 \cdot 10^{-6}$	$1.0 \pm 0.01 \cdot 10^{-5}$	$0.89 \pm 0.01 \cdot 10^{-4}$
	D-4 Bio juice Energy Plus	$2.07 \pm 0.1 \cdot 10^{-6}$	$1.6 \pm 0.1 \cdot 10^{-6}$	$0.3 \pm 0.01 \cdot 10^{-5}$	$1.2 \pm 0.1 \cdot 10^{-4}$
Apricot	A-1 Control	$2.2 \pm 0.1 \cdot 10^{-6}$	$1.1 \pm 0.01 \cdot 10^{-6}$	$0.7 \pm 0.01 \cdot 10^{-6}$	$1.2 \pm 0.44 * 10^{-3}$
	A-2 Agroflorin	$2.6 \pm 0.1 \cdot 10^{-6}$	$1.9 \pm 0.1 \cdot 10^{-6}$	$1.2 \pm 0.01 \cdot 10^{-5}$	$0.42 \pm 0.1 \cdot 10^{-4}$
	A-3 Alkaral	$2.5 \pm 0.1 \cdot 10^{-6}$	$1.6 \pm 0.01 \cdot 10^{-5}$	$1.3 \pm 0.01 \cdot 10^{-5}$	$2.12 \pm 0.1 \cdot 10^{-4}$
	A-4 Bio juice Energy Plus	$0.1 \pm 0.1 \cdot 10^{-6}$	$1.6 \pm 0.1 \cdot 10^{-6}$	$1.15 \pm 0.1 \cdot 10^{-6}$	$0.66 \pm 0.1 \cdot 10^{-4}$
Plum	P-1 Control	$1.1 \pm 0.1 \cdot 10^{-6}$	$0.5 \pm 0.01 \cdot 10^{-6}$	$0.5 \pm 0.01 \cdot 10^{-5}$	$0.93 \pm 0.1 \cdot 10^{-4}$
	P-2 Agroflorin	$2.14 \pm 0.1 \cdot 10^{-6}$	$1.18 \pm 0.1 \cdot 10^{-6}$	$1.12 \pm 0.01 \cdot 10^{-6}$	0.00
	P-3 Alkaral	$1.95 \pm 0.1 \cdot 10^{-6}$	$1.6 \pm 0.01 \cdot 10^{-6}$	$1.8 \pm 0.01 \cdot 10^{-6}$	$0.51 \pm 0.1 \cdot 10^{-4}$
	P-4 Bio juice Energy Plus	$2.4 \pm 0.1 \cdot 10^{-6}$	$2.2 \pm 0.1 \cdot 10^{-6}$	$1.2 \pm 0.1 \cdot 10^{-6}$	$0.53 \pm 0.1 \cdot 10^{-4}$

The number of microscopic fungi on Czapek's medium in the control variant in the soils of the apple cultivar Damira was two times higher than in the other variants. In the Agroflorin (D-2) variant, no microcenosis manifested, and the same also resulted in the studied soil samples; the infected background of the fungal ecosystem received no infection. Past studies have shown that with sufficient nutrition and hydration conditions, the vital activity of microorganisms was more energetic, with organic residues quickly destroyed and ammonifying bacteria multiplied more actively with organic fertilizers' use (Ivanov *et al*., 2017).

The obtained data on stone fruit crops showed that the soil microflora of the apricot cultivar Nikitinsky red-cheeked dominates with bacilli that used both organic nitrogen and mineral nitrogen. The results also revealed that in the rhizosphere of apricot trees, in the variant Agroflorin (A-2) and Alkaral (A-3), the population was higher (2.6 ± 0.1 10-6 CFU/g and 2.5 ± 0.1 10-6 CFU/g, respectively) than other variants. It also confirmed a positive relationship between the microorganisms' physiology and the properties of their environment.

The presence of bacteria of the genus *Pseudomonas* in the studied variants of apricot with Agroflorin (A-2), Alkaral (A-3), and Bio juice Energy Plus (A-4) was 1–1.5 times higher than the control (1.1 ± 0.01·10-6 CFU/g). The efficiency of amylolytic bacteria turned out to be higher with a difference of 0.45-0.6 ± 0.01·10-6 CFU/g compared with the control variant (0.7 ± 0.01·10-6 CFU/g). It was also the highest indicator of the effectiveness of using biological products. Micromycetes with a high content in the variant of Alkaral (A-3) (2.12 ± 0.1·10-4 CFU/g) can account for an exceptionally unfavorable factor from an ecological and agronomic point of view. In other options, the soil appeared to have a healthy microbiota.

During the experiment in plum cultivar Stanley soil, it was noticeable that the variants of Agroflorin (C-2) and Bio juice Energy Plus (C-4) contained a richer microflora and a higher number of ammonifying bacteria (pseudomonads and amylolytic) than with the control variant (C-1). The presence of several filamentous fungi was in the acceptable range in all soil samples, except for the absence of pathogenic microorganisms in the variant of Agroflorin (C-2). These dynamics of the number of microorganisms showed minor fluctuations in various types of biological products for fruit types and their cultivars compared with the control variant, expressed as values of the same order. According to past studies, soil microflora, which constitute a large group of soil micropopulation, play an influential role in soil structure formation (Chebotarev *et al*., 2019; Holík *et al*., 2019; Lavrenchuk and Ermoshin, 2019; Bamdad *et al*., 2022).

Analyzing the obtained results, one can note that among the rhizosphere microorganisms, there are bacteria that can stimulate plant growth. In the studied samples, the microflora incurred dominance by non-spore-forming forms of bacteria. It proves that applying organic fertilizers increased the population growth of Pseudomonas bacteria as these microorganisms were pioneers in developing organic matter (Münster, 2018; Kozlova *et al*., 2019; Ye *et al*., 2020).

As observed in pome crops, based on the content of microorganisms in the experimental groups, the Bio juice Energy Plus variant was distinct, and it might be because this organic fertilizer contains microorganisms. In contrast, there were no other microorganisms in the Agroflorin fertilizer. Nevertheless, the Agroflorin fertilizer creates suitable conditions for developing microorganisms that benefit the soil. The organic fertilizer Alkaral was less effective than other organic fertilizers in producing soil microorganisms, most likely because Alkaral fertilizer resulted from fermentation.

In addition to studying the abundance of ammonifying bacteria, Pseudomonas, amylolytic bacteria, and micromycete flora, studies also occurred on the richness of nitrogen-fixing bacteria (Emcov and Mishustin, 2023; Sun *et al*., 2023). Of particular interest was the study of the influence of soil optimization on the number of bacteria involved in the nitrogen cycle since the nitrogen reserve of the soil largely depends on these microorganisms. In the studied soil samples, the number of aerobic nitrogen fixers appears in Table 2. The table also indicated that the ratio of the number of bacteria that consume organic forms of nitrogen in the original and optimized soil was unequal. The predominance of the former over the latter indicates a relatively high content of readily available organic compounds in the ground due to the use of organic fertilizers.

The best results were also notable in terms of the content of azotobacters in the apricot trees soil sample using Agroflorin (A-2) with 99%, followed by Bio juice Energy Plus (A-4) with 93%, which favorably affects the biological activities in the soil, its phytosanitary state, and plant growth and development (Figure 2). In the soil samples of the apple cultivar Golden Delicious, the variants Alkaral (G-3), Bio juice Energy Plus (G-4), and Agroflorin (G-2) contained more nitrogen-fixing microorganisms than the control (G-1) (75%) (Table 2). The applied fertilizers increase the biodiversity of the composition of microbial associations with the use of tillage for fruit crops (Table 1).

The data results' authentication entirely assessed the dynamics of biological activity and soil fertility state in the soils of the District Talgar, Almaty region, Kazakhstan (Figure 1). Moreover, the effect of organic fertilizers on the vegetative growth and development of the apple cultivars (Star Earlis, Golden Delicious, and Damira) also received scrutiny. The biometric indicators of pome fruits have attained identification. The length of growth shoots measured with a ruler began from the branch base to the apical bud. The trunk diameter measuring was at a height of 0.3 m from the soil surface. Measuring the trunk circumference continued in the first 10 days of June 2022 and 2023 (Table 3).

The biometric measurements and records of the first year of planting showed that organic fertilizers positively impact most physiological indicators in the apple cultivars (Star Earlis, Golden Delicious, and Damira) compared with the control versions of the fruit trees. With extra positive results, the D-4 variant using organic fertilizer 'Bio juice Energy Plus' led and performed better.

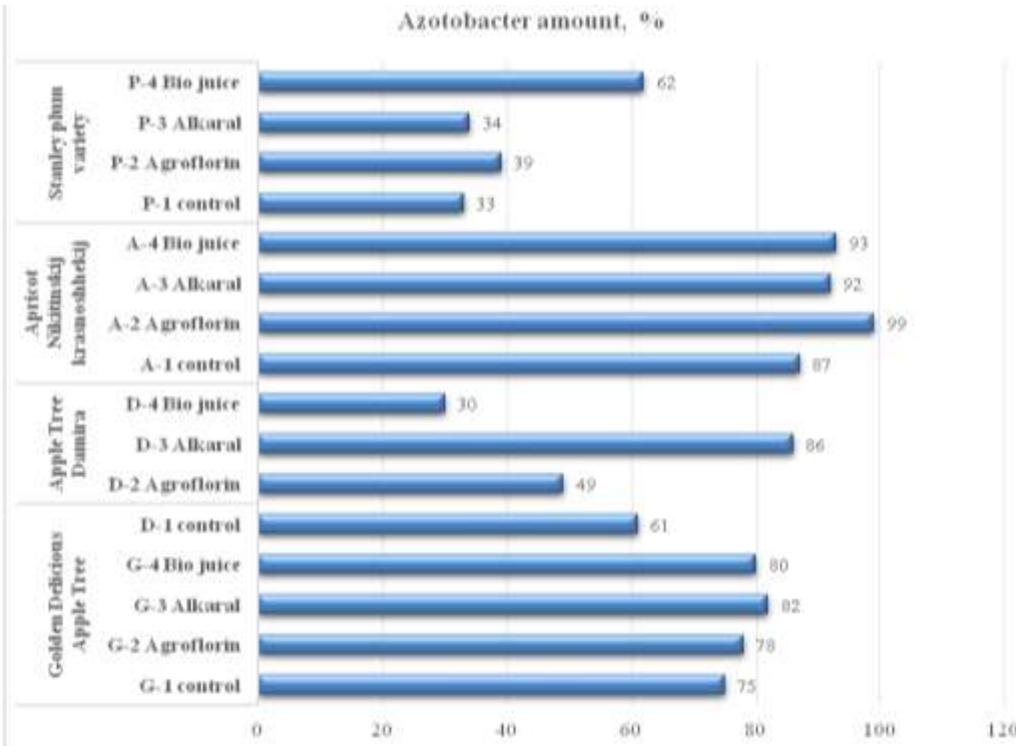

Figure 2. The content of nitrogen in the population of the genus *Azotobacter* in the soil of pome and stone fruit crops.

Table 2. Nitrogen content of the population of the genus *Azotobacter* in the soil of pome and stone fruit crops.

Variants	Fruit types and cultivars			
	Apple (Golden Delicious)	Apple (Damira)	Apricot (Nikitinsky red-cheeked)	Plum (Stanley)
G-1 Control	75	61	87	33
G-2 Agroflorin	78	49	99	39
G-3 Alkaral	82	86	92	34
G-4 Bio juice Energy Plus	80	30	93	62

Table 3. Biometric indicators of stone fruit crops based on an apple cultivar Damira.

Study years and variants	Growth parameters				
	Tree height (cm)	Trunk diameter (cm)	Trunk height (cm)	S-section of trunk (mm)	Growth (cm)
2022					
Control	1.32	1.60	43.25	2.02	41.20
Agroflorin	1.22	1.55	44.25	1.97	33.75
Alkaral	1.32	1.62	49.00	2.01	35.50
Bio juice Energy Plus	1.71	1.69	46.50	1.81	41.50
2023					
Control	1.63	1.82	47.33	2.86	41.67
Agroflorin	1.49	1.61	53.00	2.52	39.67
Alkaral	1.40	1.67	49.33	2.62	37.33
Bio juice Energy Plus	1.77	1.70	51.33	2.36	41.33

The obtained results on the biometric indicators of the second year based on the use of organic fertilizers and preparations confirm the positive effects on vegetative growth and productivity on the hibiscus of the fruits, viz., apple, plum, and apricot, compared with their control trees. The biometric indicators in Table 3 also showed that the use of organic fertilizers positively affects the vegetative growth and development of the apple tree compared with their control variant. The gathered data on biometric indicators of the second year with organic fertilizers' application and preparations indicate a favorable influence on vegetative growth and development of apple tree hibiscus compared with the control version (Table 3).

The organic fertilizers influence was more prominent on biometric indicators compared to the first year of cultivation of stone fruit crops. The two-year-long experiment conducted in an orchard using organic manure by comparing with control revealed nonsignificant differences in growth parameters; however, organic fertilizers showed a significant improvement in the quality indicators of the fruit yield (Tojnko *et al.*, 2007; Sun *et al*., 2023). Higher concentrations of bio-organic fertilizers significantly impacted soil microorganisms by reducing plants' incidence and increasing the activities of microorganisms (Zhang *et al*., 2019; Viba *et al*., 2020; Wang *et al*., 2022).

CONCLUSIONS

Soil microorganisms improve the structure of the soil, accumulate nutrients, and mineralize various organic compounds, turning them into easily digestible nutrition components, as evidenced by the positive results obtained by increasing the quantitative composition of soil microorganisms under pome and stone fruit crops. The applied organic fertilizers (Agroflorin, Alkaral, and Bio juice Energy Plus) have proven efficient in enhancing the microbial associations' qualitative and quantitative composition in the soils under fruit crops. Biologization is a controlled process of growing crop plants and increasing soil fertility based on the complex interaction between the ground with various types of plants, animals, and microorganisms, ensuring their biological protection from diseases, pests, and weeds.

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