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APPLE TREE (*Malus domestica* Borkh) NITROGEN SUPPLY OPTIMIZATION BY FERTIGATION AND BACTERIAL FERTILIZERS

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Abstract

Nitrogen is a special macronutrient as it comes into soil only with rainfall, remains of living organisms and fertilizers. Changes of apple (*Malus domestica* Borkh) nitrogen supply faster affect the fruit yield and quality as compared to other nutrients. Our paper it the first multivariate study of soil, leaf and fruit mineral status of apple trees on leached chernozem at different ways and sources of nitrogen supply which shows high environmental safety and efficiency of fertigation. The aim of the research was development of a model for optimal nitrogen provision to improve apple tree yielding and fruit quality. Tests with cv. Zhigulevskoe grafted on rootstock 62-396 were carried out in 2014-2016 in the experimental apple orchard planted in 2007 (Michurin Federal Scientific Centre, Tambov Region) at 4.5×1.0 m planting spacing. Each plot comprised 5 trees; all tests were arranged in triplicate. Obtained data were statistically processed by dispersion, correlation and regression analysis. Humus content and soil acidity, abundant of rhizosphere microorganisms, apple tree productivity, and the levels of essential elements in leaves and soil were determined, as well as vitamin C, sugars and organic acids content in fruits. Experimental variants were control 1 (no fertilizers and no irrigation), control 2 (no fertilizers, drip irrigation); subsoil placing of N_{60}, N_{90}, N_{120}, $N_{60}P_{20}K_{60}$, $N_{90}P_{30}K_{90}$, $N_{120}P_{60}K_{120}$; fertigation of N_{15}, N_{25}, N_{35}, $N_{15}P_{12}K_{15}$, $N_{25}P_{20}K_{25}$, $N_{35}P_{25}K_{35}$; Azovit, 4 l/h (cells and spores of *Azotobakter chroococcum* B-9029, 5×10^9 CFU/g), Azovit, 4 l/h + Phosphatovit, 4 l/h (cells and spores of *Bacillus mucilaginosus* B-8966, 0,129 CFU/g). In deep fertilizer placement, the complex NPK but not N increases the apple tree yielding. Fertigation with $N_{35}P_{25}K_{35}$ provides optimal level of all major nutrients in leaves and maximum fruit yield averaged 396.3 c/ha for three years. Single application of N fertilizers unbalances soil nutrient composition by increasing the content of easily hydrolyzed nitrogen, which, in turn, reduces the increase in yield. Complex fertilizing by fertigation and using bacterial preparations ensures the optimum N/P ratio in fruits (6.8), as well as K/N ratio (1.8-1.9). Fertilization by fertigation and in the tree trunk strips reduces the concentration of ascorbic acid in fruits at harvest maturity. N variants, despite the mode of fertilizer application, have the worst impact on vitamin C concentration which was 15-20 % lower compared to the complex fertilization, and the sugar-acid ratios also decreased to 10.7-12.8. Fertigation and use of bacterial fertilizers increase microbiological activity of the soil. As a result, we suggest the model for apple tree yield optimization based on proper use of nitrogen fertilizers. This study shows that the use of bacterial cultures as a temporary alternative to chemical fertilizer improves productivity in intensive apple orchards (up to 327.5 c/ha on average over 3 year experiment).

Keywords: *Malus domestica* Borkh, apple tree, nitrogen nutrition, fertigation, deep fertilizer placement, drip irrigation, yield, fruit quality

Nitrogen deficiency inhibits the growth of plant roots and aerial parts, which reduces their photosynthetic capacity and productivity [1] while optimal nitrogen intake activates enzyme proteins. It is therefore important to note that the nitrogen status regulates the ability to generate heat-shock proteins, which improves the heat resistance of plants [2]. Nitrogen intake activates photosynthesis and improves productivity [3]; it affects the growth of trees, their bodies, shoots, and leaves via protein synthesis [4, 5]. Optimal nitrogen intake has positive effects on roots, growth, root morphology and branching [6, 7]. According to Trunov [6], increased nitrogen dosage improves the intake of nitrogen, phosphorus, and potassium via roots. Nitrogen increases the number and longevity of fruit-bearing formations, causes more abundant flowering and blossoming of fruits, decreases the ovary reduction, and contributes to fruit growth, increase in size and yields [8-10]. Nitrogen surplus induces excessive shooting at the expense of productivity, causing larger but looser leaves to grow while also entailing protracted growth, slower ripening, and inhibited resistance to cold [11, 12]. Zn, Cu, and Fe deficit ensues, which results in physiological diseases [13]. Fruit ripening is delayed, which worsens their taste, shelf life, and appearance while making fruits more susceptible to physiological and infectious diseases during storage [14-16].

Apples contain sugars, necessary acids, and nearly all the vitamins and micronutrients the human needs [17]; however, excess nitrogen intake reduces the consumer quality of fruits. Meanwhile, appropriate dosing and application of nitrogen fertilizers help control the productivity of plantations and the quality of yield while also reducing costs and negative environmental impact [10, 18].

Vitamin C content is what largely determines the nutritional value of apple fruits, especially in winter. Besides, ascorbic acid is important for long-term storage. Being an antioxidant, it is involved in oxidation and reduction processes, in the synthesis of hormones and sundry essential compounds [19]. Ascorbic acid is also needed to overcome and reduce the effects of oxidative stress [20, 21]. For example, sufficient amounts of ascorbic acid inhibit pulp browning to a great extent [21]. The mechanisms behind these processes are yet to be explained; however, it has been shown that the acid inhibits negative oxidation processes in membranes and prevents peroxides from destroying fats [22]. Ascorbic acid is crucial for curbing salinity stress [23]. For the best fruit taste, the sugar-acid ratio (SAR) must be within 15 to 25. At SAR > 25, fruits become blank-tasted and useless for further processing [24].

Shelf life and preservation of fruits are two important indicators that depend not only on the storage conditions but also on the content and ratio of minerals in fruits and leaves [25], i.e. they are also related to nutrient intake.

The authors hereof are the first to have evaluated how altering mineral and water intake by fertigation and drip irrigation affects the mineral status and productivity improvements observed in large-scale leached-chernozem apple plantations. The research team was able to describe particularities of using bacterial fertilizers (Azovit and Phosphatovit) and to establish approximate recommended region-specific fertigation rates.

The purpose of this research was to model apple yield and to optimize nitrogen intake for better productivity and fruit quality while using different fertilizers.

Techniques. Experiments were carried out in a test orchard of Michurin Federal Research Center, Michurinsk, Tambov Region, in 2014-2016 on *Malus domestica* Borkh trees, cv. Zhigulevskoye grafted on rootstock 62-396. The orchard was planted in 2007 with a 4.5 m×1 m planting layout and equipped with a fertigation and drip irrigation system; each plot comprised 5 trees in triplicates. The test-plot soil was leached, low-humus, medium-loam meadow chernozem on sands with pseudofibers. The humus content was 2.6 to 3.2 percent, the alkaline saturation was 70 to 90 percent, and the humus horizon depth was 40 to 50 cm on average. Upper-layer reaction was low-acidic at pH 5.7 to 5.9. It was a dusty, crumby, and grainy soil. Upper-horizon porosity reached 65%. Field moisture capacity of the arable layer was about 30%, the easily hydrolyzed nitrogen content was 152.8 mg/kg as found by I.V. Tyurin and M.M. Kononova's method; mobile phosphorus content was 146.0 mg/kg, while exchangeable potassium content was

167.6 mg/kg as found by the Chirikov method.

Fertilizers were incorporated annually in early spring at 10 to 15 cm below the surface; fertigation was performed throughout the vegetation period to meet the nutrient-specific needs of plants. Annual fertigation rates were divided into 10 irrigations. Sampling was as follows: Control 1 (no fertilizers, no irrigation); Control 2 (no fertilizers, drip irrigation); application of N_{60}, N_{90}, N_{120}, $N_{60}P_{20}K_{60}$, $N_{90}P_{30}K_{90}$; application of bacterial fertilizers: Azovit (4 l/ha, living cells and spores of *Azotobacter chroococcum* B-9029, 5×10^9 CFU/g), Azovit (4 l/ha) plus Phosphatovit (4 l/ha, living cells and spores of *Bacillus mucilaginosus* B-8966, 0.129 CFU/g). The bacterial fertilizers were produced by Industrial Innovations Limited, Russia. In case of drip irrigation, bacterial fertilizers were applied by irrigating the roots in spring.

The following preparations were incorporated in the soil: ammonium nitrate containing at least 34.4% of ammonium- and nitrate-nitrogen; huminified superphosphate, a $Ca(H_2PO_4)_2 \cdot H_2O$ water-soluble phosphorus-based fertilizer with 26% P_2O_5, 5% N; and potassium sulfate, a concentrated chlorine-free fertilizer (46-50% K_2O). All the preparations were produced by OAO Buy Chemicals Factory, BHZ, Russia. For fertigation, the researchers used Amofoska, a complex NPK fertilizer (12% N, 15% P_2O_5, 15% K_2O, 14% S, 0.5% MgO) produced by OOO Mettorg, Russia; potassium monophosphate, a phosphorus and potassium compound fertilizer (52% P_2O_5, 34% K_2O) produced by OAO BHZ; and Master, a complex water-soluble fertilizer with chelated micronutrients ($N_{13}P_{40}K_{13}$, 0.070% Fe, 0.030% Mn, 0.010% Zn, 0.005% Cu, 0.020% B, 0.001% Mo) produced by Valagro SpA, Italy. The fertilizer composition for making a fertigation solution was adjusted to vegetation phase-specific needs.

The soil was sampled in late August (in June and August to measure microbial population) at a depth of up to 40 cm. Data included humus quantification, easily hydrolyzed nitrogen quantification by the Kjeldahl method [26]; mobile phosphorus quantification by a KFK-3-01 photometer (Zagorsk Optical and Mechanical Plant, Russia) and exchangeable potassium quantification by a Jenway PFP 7 flame photometer (Bibby Scientific, UK) by the Chirikov method [27]; soil pH in KCl extraction; total exchangeable bases by the Kappen-Hilkovic method; and the microbial population of the rhizosphere by the Krasilnikov method, using beef extract agar (BEA) [28].

Leaves were sampled in mid-August. Total potassium, phosphorus, and nitrogen (by the Kjeldahl method) content were evaluated in a single batch [27].

Monosaccharide and disaccharide content was determined by the Bertrand method; ascorbic acid content was measured by iodometry; total acidity was measured by titrometry [29]; dry content was measured after drying to constant weight [27]. Only picking-maturity fruits were used for these measurements. The yield was accounted when weighing fruits from accountable trees [30].

Data were processed statistically by analysis of variance, correlation and regression analysis [31, 32] using Microsoft Excel 2007 with the AgCStat add-in. Below are the mean values (M) with standard errors of means (±SEM), pairwise correlation coefficients (r), and least significant difference values at CI 95% ($t_{0.05}$).

Results. Nitrogen is extremely mobile in plants and soils; nitrate-nitrogen is easily leached into groundwater. Active drip irrigation and excessive application of nitrogen fertilizers may have negative effects, which is why clarifying the particularities of nitrogen intake is important for reducing environmental pollution [33].

In this experiment, nitrogen fertilizers applied without irrigation had no significant effect on the mean triennial yield (LSD_{05} 36.4 centers/ha), except the sample that had the maximum rate of N_{120} (see Fig. 1). Application of nitrogen fertilizers at 120 kg/ha, as well as incorporation of complex fertilizers in the neartrunk areas resulted in a significant increase in yield against the controls: the mean triennial yield rose from 260.2 to 317.0 (N_{120}), 321.2 ($N_{90}P_{30}K_{90}$) and 321 centners/ha ($N_{120}P_{60}K_{120}$), i.e. by 56.8 to 60.0 centners/ha (or by 21.8 to 23.1%) due to raising the soil nitrogen, phosphorus, and potassium content to the optimal levels. Fertigation enabled lowering the application rates; however, a significant increase in productivity was only noted in the case of complex fertilizers, as yield peaked when using the maximum dosage. Over 2014-2016, drip irrigation increased this indicator from 260.2 (Control 1) to 295.4 (Control 2) centners/ha on average, i.e. by 35.2 centners/ha or by 13.5%.

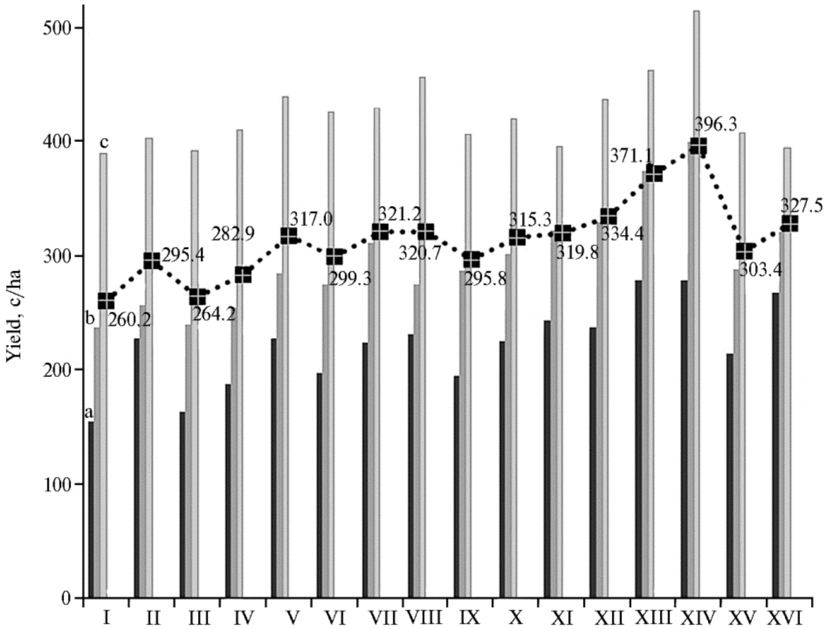

Fig. 1. ***Malus domestica*** **Borkh, cv. Zhigulevskoye yield on rootstock 62-396: results obtained by using different nitrogen fertilizing methods in 2014** (a)**, 2015** (b)**, and 2016** (c)**:** I for Control 1 (no fertilizers, no irrigation); II for Control 2 (drip irrigation); III for N60, IV for N_{90}, V for N_{120}, VI for $N_{60}P_{20}K_{60}$, VII for $N_{90}P_{30}K_{90}$, VIII $N_{120}P_{60}K_{120}$ (fertilizers); IX for N_{15}, X for N_{25}, XI for N_{35}, XII for $N_{15}P_{12}K_{15}$; XIII for $N_{25}P_{20}K_{25}$; XIV for $N_{35}P_{25}K_{35}$ (fertigation); XV for Azovit, XVI for Azovit + Phosphatovit. The figure shows the mean yield. LSD_{05}: 25.8 in 2014; 34.0 in 2015; 49.3 in 2016; the triennial mean is 36.4 centners/ha (Michurinsk, Tambov Province).

Azovit and Phosphatovit are preparations intended to replace or limit the use of mineral fertilizers while optimizing the assimilation of the essential nutrients, which is expected to produce eco-friendly products of better quality and yield. Yield increase in bacterial-fertilized samples was evaluated against Control 2. Azovit placement in the soil did not have a significant effect on this indicator. A combination of bacterial fertilizers did increase the yield, but the increase was commensurate with that resultant from a minimum concentration of a complex mineral fertilizer. Simultaneous application of Azovit and Phosphatovit increased the yield from 295.4 to 337.5 centners/ha, i.e. by 42.1 centners/ha or by 14.3%; this was due to increasing the bacterial colonization of the apple tree's rhizosphere and improving the availability of minerals. Fertigation with a mineral complex increased the mean triennial yield in 2014-2016 from 295.4 to 334.4 ($N_{15}P_{12}K_{15}$), 371.1 ($N_{25}P_{20}K_{25}$), and 396.3 centners/ha ($N_{35}P_{25}K_{35}$), i.e. by 39.0 to 100.9 centners/ha or by 13.2 to 34.2 percent, which is statistically significant (LSD_{05} = 36.4 centners/ha).

Fertilizing positively affected the soil nutrient content, see Table 1. Nitrogen and phosphorus fertilizing was of utmost significance in this experiment, as the content of these elements was suboptimal in the controls. Exchangeable-potassium content in the soil was higher, although, in some years, plants were deficient in potassium as well, perhaps due to precipitation-induced leaching and redistribution of root-absorbed potassium into the ripening fruits [34]. Such response greatly depends on the experimental conditions, primarily on the soil type. In a study by D. Malaguti et al. [35], potassium content was increased by fertigation, while nitrogen content was increased by incorporation in alluvial soils, Italy. However, high-dose fertigation may increase the heterogeneity of soils in an orchard [36]. In this experiment, unifactorial application of nitrogen increased its content to optimal levels (177.9 to 201.6 mg/kg of soil); however, phosphorus and potassium content remained unchanged (121.7 to 137.3 P_2O_5 and 128.9 to 150.7 K_2O, values in mg/kg of soil). The multifactorial application only increased yield when the nitrogen application rates were at max. Complex fertilizing optimized the soil nutrient content in nearly all incorporated-fertilizer samples. Comparing fertigation at down to 40 cm below surface against incorporation at 10 to 15 cm below surface, easily hydrolyzed nitrogen content was only 3 to 5 percent lower; meanwhile, such fertigation used 70 to 75 percent less nitrogen fertilizer(s).

1. Soil and leaf nutrient content in apple trees *Malus domestica* Borkh (cv. Zhigulevskoye, rootstock 62-396) for different nitrogen fertilizing methods (M±SEM, Michurinsk, Tambov Region, 2014-2016)

Variant	Content					
	in the soil, mg/kg			in leaves, % wet		
	easily hydrolyzed N	P_2O_5	K_2O	N	P	K
No fertilizers						
Control 1 (no irrigation)	107,2	131,4	138,8	1,17	0,32	0,76
Control 2 (irrigated)	87,4	114,5	114,6	1,37	0,21	0,98
Mean	97,3±4,3	123,0±4,2	126,7±4,7	1,27±0,067	0,27±0,008	0,87±0,041
Incorporation of fertilizers						
N_{60}	127,4	121,7	150,7	1,48	0,24	1,21
N_{90}	177,9	137,3	128,9	1,55	0,22	1,09
N_{120}	201,6	123,2	135,6	1,77	0,26	1,25
$N_{60}P_{20}K_{60}$	146,5	176,4	173,9	1,87	0,33	1,34
$N_{90}P_{30}K_{90}$	184,6	185,5	206,1	1,95	0,41	1,26
$N_{120}P_{60}K_{120}$	196,8	193,1	219,7	2,14	0,39	1,48
Mean	172,5±8,1	156,2±9,4	169,2±8,8	1,79±0,094	0,31±0,011	1,27±0,57
Fertigation						
N_{15}	156,3±7,8	102,9	118,4	1,60	0,28	1,09
N_{25}	173,6	107,7	128,9	1,69	0,18	1,19
N_{35}	177,4	115,7	107,7	1,96	0,32	0,95
$N_{15}P_{12}K_{15}$	156,3	142,5	168,9	1,88	0,44	1,48
$N_{25}P_{20}K_{25}$	166,4	154,9	175,8	2,07	0,49	1,51
$N_{35}P_{25}K_{35}$	182,7	169,7	171,2	2,41	0,55	1,30
Mean	168,8±7,7	132,2±7,3	145,2±8,1	1,94±0,114	0,38±0,012	1,25±0,055
Bacterial fertilizers						
Azovit, 4 l/ha	162,4	103,9	141,3	1,70	0,31	1,20
Azovit, 4 l/ha + Phosphatovit, 4 l/ha	175,3	172,7	182,2	2,27	0,43	1,33
Mean	168,9±8,2	172,7±8,9	182,2±9,2	2,0±0,115	0,37±0,09	1,3±0,057
LSD_{05}	22,2	18,3	15,1	0,18	0,07	0,12
Optimal content (according to literature))						
	151-200 [37]	151-200 [38]	121-180 [38]	1,8-2,5 [39]	0,3-0,5 [39]	1,2-1,8 [39]

The need for nitrogen and its availability depend on the water intake as well as on the presence and content of other nutrients in the soil. Leaves indicate the availability of nutrients to plants. However, nutrient content in leaves does not correlate directly with that in the soil; it is only nitrogen that displays explicit correlations [40]. Despite more easily hydrolyzed nitrogen being present in the soil in case of unifactorial nitrogen fertilizing, leaf nitrogen content was suboptimal. This was typical for incorporation and for fertigation alike. Only complex fertilizing

2. Chemical composition and some quality indicators of apple fruits (*Malus domestica* Borkh, cv. Zhigulevskoye, rootstock 62-396) depending on fertilizing methods (*M*±SEM, Michurinsk, Tambov Province, 2014-2016)

Variant	Basic nutrients, % wet			Ratio in fruits		Content			SAR
	N	P	K	N/P	N/K	ascorbic acid, mg%	total sugars, %	organic acids, %	
No fertilizers									
Control 1 (no irrigation)	0.19	0.046	0.69	4.1	3.6	15.70	10.5	0.66	15.9
Control 2 (irrigated)	0.23	0.034	0.79	6.8	3.4	14.36	10.1	0.76	13.3
Mean	0.21±0.01	0.040±0.02	0.74±0.04	5.5±0.3	3.5±0.2	15.03±0.75	10.3±0.6	0.71±0.04	14.6±0.8
Incorporation of fertilizers									
N_{60}	0.19	0.033	0.85	5.8	4.5	13.48	9.2	0.83	11.1
N_{90}	0.42	0.059	0.67	7.1	1.6	12.78	8.6	0.73	11.8
N_{120}	0.55	0.053	1.04	10.4	1.9	11.85	8.1	0.76	10.7
$N_{60}P_{20}K_{60}$	0.28	0.073	1.06	3.8	3.8	13.20	10.0	0.68	14.7
$N_{90}P_{30}K_{90}$	0.39	0.070	1.24	5.6	3.2	14.83	11.6	0.77	15.1
$N_{120}P_{60}K_{120}$	0.48	0.078	1.14	6.2	2.3	14.07	11.7	0.82	14.3
Mean	0.39±0.02	0.061±0.003	1.00±0.05	6.5±0.4	2.9±0.1	13.37±0.71	9.6±0.6	0.77±0.04	13.0±0.8
Fertigation									
N_{15}	0.33	0.062	0.69	5.3	2.1	13.64	8.7	0.68	12.8
N_{25}	0.40	0.033	0.76	12.1	1.9	12.83	8.7	0.82	10.6
N_{35}	0.57	0.044	0.73	13.0	1.3	11.94	8.6	0.77	11.2
$N_{15}P_{12}K_{15}$	0.40	0.069	1.07	5.8	2.6	15.41	12.4	0.74	16.8
$N_{25}P_{20}K_{25}$	0.41	0.055	1.13	7.5	2.8	14.75	12.5	0.93	13.4
$N_{35}P_{25}K_{35}$	0.48	0.071	1.15	6.8	1.9	14.87	12.9	0.76	17.0
Mean	0.43±0.02	0.056±0.03	0.92±0.005	8.4±0.05	2.1±0.1	13.91±0.68	9.6±.05	0.74±0.05	13.6±0.7
Bacterial fertilizers									
Azovit, 4 l/ha	0.34	0.052	0.69	6.5	2.3	14.09	9.6	0.84	11.4
Azovit, 4 l/ha + Phosphatovit, 4 l/ha	0.48	0.071	0.86	6.8	1.8	15.72	11.8	0.74	15.9
Mean	0.41±0.02	0.062±0.003	0.78±0.04	6.7±0.4	2.1±0.1	14.91±0.91	10.7±0.7	0.79±0.05	13.7±0.8
LSD_{05}	0.05	0.007	0.12	0.9	0.3	1.92	1.3	0.08	1.76
Comparative figures (according to literature)									
	0.3-0.5 [37]	0.07-0.10 [37]	0.8-1.2 [37]	6.5-7.0 [unp]	1.8-2.2 [но]	10.1-15.0 [41]	9.2-12.2 [41]	0.51-0.80 [41]	13.0-20.0 [41]

Note. SAR stands for the sugar-acid ratio. The Table presents optimal N, P, and K content [37]; fruit means of total sugars, organic acids, and SAR [41]; unp is the results of the authors' unpublished studies.

aimed to optimize the content of all the nutrients under analysis was able to raise leaf nitrogen content to optimal levels.

Compared to leaves, the concentration of elements in fruits has a weaker correlation with that in the soil. However, fruit chemistry data enable quality analysis and shelf-life forecasts. Too high fruit nitrogen content coupled with relatively low calcium content may result in numerous physiological diseases during storage [21].

Increasing the nitrogen fertilizer application rates resulted in accumulating excessive amounts of nutrients, primarily nitrogen itself, see Table 2. Unifactorial nitrogen fertigation raised its quantity above optimum. Meanwhile, such high nitrogen content was coupled with suboptimal concentrations of other elements, especially phosphorus. Exchangeable-potassium availability in the soil was sufficiently high; however, its content in fruits largely depended on the fruit load.

Complex fertilizing can optimize the availability of elements, the ratios whereof are crucial for quality. Such ratios largely depend on the variety and on the region of cultivation; unpublished data suggest that N/P = 6.5 to 7.0 and N/K = 1.2 to 2.2 is optimal for Central Chernozemye. Optimal N/P ratios were observed at peak fertilizing rates in both incorporation and fertigation, in Control 2 and in both bacterial-fertilized samples. When applying the maximum amount of complex mineral fertilizers by incorporation, when applying nitrogen fertilizers and maximum complex by fertigation, as well as when applying bacterial fertilizer, N/K was optimal or close to optimal. Both N/K and N/P were only optimized by maximum-rate incorporation ($N_{120}P_{60}K_{120}$) and fertigation ($N_{35}P_{25}K_{35}$). Apparently, this was resultant from the improved microbial activity of the soil, which enhanced the absorption of nitrogen and sundry elements alike.

3. Test-site soil characterization (layer 0 to 40 cm) **in experiments with *Malus domestica* Borkh (cv. Zhigulevskoye, rootstock 62-396) under various fertilizing methods** (*M*±SEM, Michurinsk, Tambov Province, 2014-2016)

Variant	pH	Humus content, %	Total exchangeable bases, mmol/100 g of soil
No fertilizers			
Control 1 (no irrigation)	5.5	3.0	26.2
Control 2 (irrigated)	5.5	2.9	26.0
Mean	5.50±0.14	2.95±0.11	26.10±1.14
Incorporation of fertilizers			
N_{60}	5.6	2.9	26.1
N_{90}	5.5	3.0	24.4
N_{120}	5.6	2.8	23.8
$N_{60}P_{20}K_{75}$	5.5	2.9	25.9
$N_{90}P_{30}K_{90}$	5.4	3.1	26.8
$N_{120}P_{60}K_{120}$	5.4	3.0	26.8
Mean	5.50±0.23	2.95±0.14	25.63±1.46
Fertigation			
N_{15}	5.5	3.0	24.2
N_{25}	5.4	3.1	27.0
N_{35}	5.4	2.9	25.1
$N_{15}P_{12}K_{15}$	5.4	3.0	27.3
$N_{25}P_{20}K_{25}$	5.4	2.8	26.9
$N_{35}P_{25}K_{35}$	5.4	2.9	23.4
Mean	5.42±0.18	2.95±0.07	25.65±1.62
Bacterial fertilizers			
Azovit, 4 l/ha	5.5	2.8	26.1
Azovit, 4 l/ha + Phosphatovit, 4 l/ha	5.6	2.9	25.8
Mean	5.55±0.17	2.95±0.11	25.95±1.38
LSD_{05}	0.14	0.19	1.19

Fertilization had negative effects on the ascorbic acid content in the picking-maturity fruits. The mean triennial peak in this indicator was noted in Control 1 and when applying the complex bacterial fertilizers to the soil. $N_{90}P_{30}K_{90}$, $N_{120}P_{60}K_{120}$ and complex fertigation samples had slightly lesser (yet within statistical error) vitamin C content regardless of the application rate. Fertilizing the soil with nitrogen had a significant negative effect on the ascorbic acid content, which was reduced at greater fertilizing rates.

Introduction and fertigation with complex mineral and bacterial fertilizers resulted in greater sugar content compared to the controls. Meanwhile, unifactorial application of nitrogen fertilizers resulted in a significant decrease in this indicator. The degree of the negative effect did not correlate with the application rates or methods; only in the case of Azovit, the effect did not occur. Organic-acid content in picking-maturity fruits was greater in case of using mineral fertilizers; however, no unambiguous correlation with the unifactorial application of nitrogen was identified. Since the unifactorial application of nitrogen fertilizers had negative effects on fruit sugars content, the increased organic-acid content in these samples entailed a considerable drop in the sugar-acid ratio (SAR).

Soil humus content and acidity varied insignificantly from sample to sample. No unambiguous correlation was identified in such variations. Total absorbed bases in the 0 to 40 cm layer did not depend on the fertilizer type or on whether irrigation was used.

Complete NPK fertigation and application of bacterial preparations led to a significant increase in the microbial population of the rhizosphere, as compared to the controls, see Figure 2; NPK fertigation increased the microbial population in June, while bacterial fertilizers did so in August. Microbial population was comparable to the controls in case of top-dressing application and incorporation of complete NPK or N-only fertilizers. Nitrogen fertigation resulted in a significant increase in the microbial activity of soils, but only in June. Unifactorial application of mineral nitrogen fertilizers by incorporation (N_{120}) did not have a significant effect on the microbial population; however, unifactorial application of Azovit did increase the microbial activity.

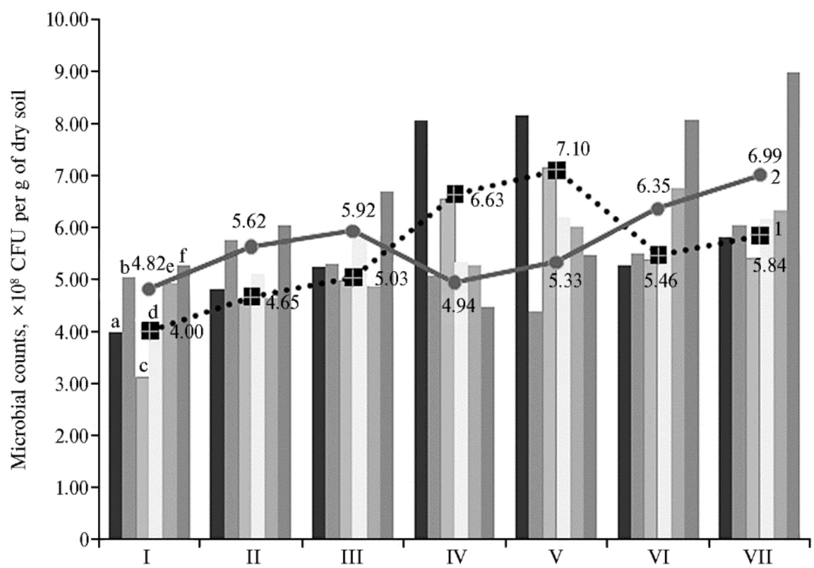

Fig. 2. Microbial population of *Malus domestica* Borkh (cv. Zhigulevskoye, rootstock 62-396) rhizosphere as affected by nitrogen fertilizers in June (a) **and August** (b)**, 2014; in June** (c) **and August** (d)**, 2015; and in June** (e) **and August** (f)**, 2016:** I for Control 1 (no fertilizers, no irrigation), II for N_{120}, III for $N_{120}P_{60}K_{120}$, IV for N_{35}, V for $N_{35}P_{25}K_{35}$; VI for Azovit, 4 l/ha, VII for Azovit, 4 l/ha + Phosphatovit, 4 l/ha; 1 and 2 for the mean microbial population in June and August, respectively. LSD_{05}: 1.06×10^6 in June 2014; 7.65×10^5 in August 2014; 5.53×10^5 in June 2015; 3.40×10^5 in August 2015; 3.51×10^5 in June 2016; 6.93×10^5 in August 2016; the triennial means were 3.58×10^5 for June and 8.38×10^5 for August, respectively. The figures are given in CFU/g (Michurinsk, Tambov Province, 2014-2016).

The authors hereof had previously indicated that the microbial population of the apple rhizosphere had a direct positive correlation with the plant productivity [42]. Applying a mineral-organic complex fertilizer in combination with Extrasol (*Basillus subtilus* Ch 13 strain, liquid form) improved the apple yield and the microbial activity of soils in the Moscow Region [43]. Using a bioorganic fertilizer in China (Linfen, Shaanxi) brought about a significant improvement in yield and productivity, as well as in the physicochemical and enzymatic activity of soils [44].

Data processing reveals that the leaf nitrogen content positively correlates with the easily hydrolyzed nitrogen in the soil (r = 0.73) as well as with yield (r = 0.72). The easily hydrolyzed nitrogen content in the soil correlates with that in fruits, albeit such correlation is weaker at r = 0.61.

Based on these results, the authors hereof have developed an apple yield model to optimize nitrogen fertilizing in the context of other factors: $Y = 1.37 + 0.0922x_1 - 0.0521x_2 + 9.01x_3 + 108.95x_4 + 1.91x_5 - 1.98x_6 + 0.61x_7 + 0.63x_8 + 0.21x_9$, where Y is yield, centners/ha; x_1 is the nitrogen fertilizer application rate, pn kg/ha; x_2 is the easily hydrolyzed nitrogen content in the soil; x_3 is the leaf nitrogen content, dry matter %; x_4 is the fruit nitrogen content, dry matter, %; x_5 is the soil humidity, %; x_6 is the phosphorus fertilizer application rate, pn kg/ha; x_7 is the potassium fertilizer application rate, pn kg/ha; x_8 is the available phosphorus content in the soil, mg/kg of soil; x_9 is the exchangeable-potassium content in the soil, mg/kg of soil; pn stands for primary nutrient.

Therefore, unifactorial application of nitrogen to soils helps optimize its accumulation in leaves at very high fertilizing rates; however, this causes a nutritional imbalance while exposing the environment to a greater chemical load. Fertigation has the greatest effect on apple yield and fruit quality while being eco-friendlier, as it requires 70 to 75 percent less fertilizer than incorporation. It does improve the microbial activity of soils, which in its turn improves the absorption of nutrients by plants, thus enhancing the physiological status of trees.

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