The effect of mineral nutrition on the biochemical composition of Venyaminovskoe apple cultivar fruits

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Abstract. The results of a five-year study of the mineral fertilizers' effect on the chemical composition of Venyaminovskoe apple cultivar fruits are presented. It is shown that the use of soil and foliar application of nitrogen and potash fertilizers did not have a significant effect on the content of soluble solids, sugars, titrated acids, ascorbic acid, and phenolic compounds in fruits. It was established that the most significant factor affecting the chemical composition of fruits are the weather conditions of the growing season. At the same time, the content of ascorbic acid significantly exceeding the control over an average of five experimental years was found in the option with the introduction of nitrogen and potash fertilizers into the soil at a dose of N60K80 together with foliar top dressing. There is a tendency of increasing accumulation of leucoanthocyanins in fruits over the years. In 2021, there was a significant decrease in catechins with the use of mineral fertilizers at a dose of N60K80, as well as in the following options: without soil and minimal (N30K40) fertilization against the background of top dressing.

1 Introduction

Full nutrition is the basis of vital activity and one of the most important factors ensuring active longevity, participating in the formation and realization of the adaptive potential of the human body, contributing to reducing the risk of developing alimentary dependent pathologies [1].

Currently, it is especially important to improve the quality of agricultural products including fruits. Fruits and berries consumed in the native (not processed) state should have high consumer advantages: marketable appearance and rich biochemical composition. Obtaining high-quality fruit products is ensured primarily by the introduction of new selection varieties into production combined with agrotechnical measures, including the use of mineral fertilizers.

Among fruit crops, apple occupies one of the leading places in the world, second only to bananas and citrus fruits. Currently, apples are produced in 51 countries. The population of the world prefers apples to many of the fruits grown [2-6]. Apples also occupy a leading
position in the consumer basket of the Russian population. According to the areas of fruit plantations in the country, the apple trees account for 65% [7, 8].

Apple fruits are an important component of a healthy diet due to the wide range and high content of nutrients (sugars, organic acids) and biologically active compounds (ascorbic acid and phenolic complex) that the human body is unable to synthesize. Based on this, apples are particularly valuable products that have not only highly nutritious, but also therapeutic value [9, 10].

Since the nutritional value of apples is largely determined by the content of organic components, important tasks of apple agrotechnics are not only to increase the crop’s productivity, but also to obtain fruits with rich biochemical composition. The chemical composition of apples can be influenced by various factors: cultivar, age of trees, the timing of their ripening and harvesting, meteorological conditions of the growing season, etc.; with this, mineral nutrition plays an important role in the formation of yield and fruit quality [11-14].

However, fertilizers' introduction into the soil cannot always provide a sufficiently reliable effect due to the varying availability of elements, environmental conditions, delayed manifestation of fertilizers' effect on perennial crops. Mineral elements supplied with fertilizers take part in the formation of chemical indicators' complex of fruit quality [15-19].

The most significant factor determining a positive or negative effect on the accumulation of various biochemical substances in fruits is the cultivar. Each genotype reacts to different doses and methods of applying mineral fertilizers in different ways. Based on this, an in-depth and concretized selection of mineral nutrition for each cultivar is necessary to improve the chemical composition of fruits, in particular apples. In this regard, the goal was to study the effect of mineral fertilizers on the biochemical composition of Veniaminovskoe apple fruits, in particular, on the content of soluble solids, the amount of sugars, ascorbic acid, titrated acids, P-active catechins, and leucoanthocyanins.

2 Materials and methods

The research was conducted at the All-Russian Scientific Research Institute of Fruit Crop Breeding (VNISPK) (Orel region) in 2017...2021. The object of the study was the Veniaminovskoe apple cultivar grown in a field experiment to study the mineral fertilizers' efficiency in a medium-sized apple orchard. The research was carried out for five years (2017...2021) jointly by two laboratories of the Institute: fertilization – by the laboratory of agrochemistry, study of the biochemical fruits' composition – by the laboratory of biochemical and technological evaluation of varieties and storage. Mixed samples of picking-maturity fruits were taken from each experimental plot when recording the harvest.

The soil of the experimental site is medium-loamy agro-gray underlain by dolomitic limestones. Agrochemical indicators of the arable soil layer at the time of the experiment laying: pHKCl – 5.03...5.72, humus – 3.7 ±0.14%, mobile phosphorus – 110...200 mg/kg, exchange potassium – 110...170 mg/kg.

The experimental orchard was planted according to the scheme 6.0 x 3.0 m in 2013. The annual spring application of increasing doses of nitrogen (in the form of ammonium nitrate – 33% a.s.) and potash fertilizers (in the form of potassium chloride – 40% a. s.) was started in 2015. Foliar fertilizing was carried out three times during the growing season: after flowering – 1% urea solution; in the phase of intensive shoots' growth (July) – a mixture of 1% urea solution and 0.3% potassium sulfate solution; 30-40 days before fruit picking – 0.3% potassium sulfate solution. The experiment's repetition was 4-fold, there were 5 trees in each repetition. The arrangement of options was systematic. Mixed samples of fruits were taken from each experimental plot when recording the harvest.
The content of soluble solids (SS) in fruits was determined by the refractometric method using a PAL-3 digital refractometer (ATAGO); sum of sugars – by the Bertrand method (GOST 8756.13-87); titrated acids (total acidity) – by titration of extracts with a decinormal solution of sodium hydroxide (GOST 25555.0 – 08); ascorbic acid – by titration of oxalic acid extracts with Tilmans dye (2,6 – sodium dichlorophenolindophenolate); determination of phenolic compounds (catechins and leucoanthocyanins) – by photometric method using a photometer FEC KFK–3–01–"ZOMZ" [20, 21].

Statistical processing of the obtained data was carried out by the method of two-way analysis of variance [22].

3 Results and discussion

Studies conducted earlier in the soil and climatic conditions of the Orel region showed a significant influence of the growing season's meteorological conditions on the biochemical composition of apple fruits [23, 24].

The average annual data and hydrothermal conditions of the growing season in the studied years are presented in Table 1.

Table 1. Weather conditions of the growing season, 2017...2021.

| Month | 2017 | 2018 | 2019 | 2020 | 2021 | Average long-term values |
|-------|------|------|------|------|------|--------------------------|
| Average monthly temperature, ºC | | | | | | |
| May   | 12.3 | 16.4 | 15.6 | 11.3 | 14.0 | 13.0 |
| June  | 16.0 | 17.0 | 20.5 | 19.9 | 19.7 | 16.9 |
| July  | 18.6 | 19.9 | 17.4 | 19.6 | 21.8 | 18.5 |
| August| 19.2 | 18.4 | 17.1 | 18.2 | 20.2 | 17.1 |
| Precipitation sum, mm | | | | | | |
| May   | 56.3 | 31.4 | 85.0 | 59.1 | 63.3 | 36.3 |
| June  | 59.6 | 18.2 | 20.7 | 46.4 | 99.6 | 65.1 |
| July  | 75.0 | 119.9| 49.8 | 111.6| 37.8 | 88.0 |
| August| 100.8| 11.2 | 54.7 | 26.0 | 29.0 | 65.7 |
| Σ     | 291.7| 180.7| 210.3| 243.1| 229.7| 255.1 |

The average monthly temperature of the growing season was higher compared to the average annual data in 2018, 2020, and 2021. The amount of precipitation significantly differed from the average annual values. The most humid growing season was in 2017 (291.7 mm), the driest season was in 2018 (180.7 mm). During the fruit ripening period (August), there was an insufficient amount of moisture in 2018, 2020, 2021, optimal (54.7 mm) – in 2019, excessive (100.8 mm) – in 2017.

For the period of 2017...2021 depending on the year and the research option, the SS content ranged from 12.80 to 15.15%, on average over the years – from 13.03 to 14.56% (Table 2). No significant differences were found depending on the applied doses of fertilizers. The weather conditions of the growing season had a favorable effect on the SS accumulation in fruits, the average experimental value of the indicator was significantly higher in 2019, when August (the period of fruit ripening) had an optimal temperature regime.
Table 2. The content of soluble solids (%) in the fruits of the Venyaminovskoe apple cultivar when applying fertilizers, 2017...2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|-------------------|-----------------------------|-----------|
|                   | 2017 | 2018 | 2019 | 2020 | 2021 |
| 1. Control        | 12.80 | 12.80 | 14.45 | 13.90 | 13.30 | 13.45 |
| 2. N30 K40        | 13.10 | 13.70 | 14.20 | 14.55 | 13.95 | 13.90 |
| 3. N60 K80        | 13.55 | 13.15 | 14.25 | 13.25 | 13.90 | 13.62 |
| 4. N90 K120       | 13.25 | 12.75 | 14.65 | 13.35 | 13.70 | 13.54 |
| 5. Control + fer.(top dr.) | 13.20 | 12.60 | 14.95 | 13.65 | 13.65 | 13.61 |
| 6. N30K40 + fer. (top dr.) | 13.15 | 13.20 | 15.15 | 13.20 | 14.35 | 13.81 |
| 7. N60K80 + fer. (top dr.) | 13.20 | 13.00 | 14.65 | 13.20 | 13.90 | 13.59 |
| 8. N90K120 + fer. (top dr.) | 13.80 | 13.10 | 14.25 | 13.45 | 13.20 | 13.56 |
| Average B         | 13.25 | 13.03 | 14.56 | 13.56 | 13.74 |

LSD_{0.5} A = 0.62, LSD_{0.5} B = 0.49, LSD_{0.5} AB = 1.40

In the literary data, there is information on an increase in the mass SS fraction in apples when applying nitrogen and potash fertilizers [25, 26]; however, these data are not mathematically confirmed. In these studies, the soil and foliar application of nitrogen and potash fertilizers also did not have a significant effect on the SS accumulation in the fruits of the Venyaminovskoe cultivar.

The average content of the sum of sugars in all study options varied depending on the year from 11.22 to 12.26% (Table 3) and was significantly higher in 2020 and 2021 with a similar temperature and humidity regime of the ripening period of both years. In 2017 and 2018, there was a significant decrease in this indicator.

SS is an indirect indicator of the sugar content in fruits, which is in close correlation with the latter. In this study, the use of nitrogen and potash fertilizers did not have a significant effect on the sugar content in the fruits of the Venyaminovskoe cultivar, as well as on the content of SS, although there was a tendency to increase the level of sugars in fruits that received additional top dressing nutrition.

Table 3. The content of sugar sum (%) in the fruits of the Venyaminovskoe apple cultivar when fertilizing, 2017-2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|-------------------|-----------------------------|-----------|
|                   | 2017 | 2018 | 2019 | 2020 | 2021 |
| 1. Control        | 10.58 | 11.73 | 11.80 | 12.48 | 11.41 | 11.60 |
| 2. N30 K40        | 11.30 | 11.66 | 10.99 | 12.81 | 12.24 | 11.80 |
| 3. N60 K80        | 11.63 | 11.08 | 11.95 | 11.89 | 12.13 | 11.73 |
| 4. N90 K120       | 11.04 | 11.40 | 12.07 | 11.90 | 12.34 | 11.75 |
| 5. Control + fer.(top dr.) | 11.08 | 11.86 | 11.45 | 12.86 | 11.69 | 11.78 |
| 6. N30K40 + fer. (top dr.) | 10.93 | 12.37 | 12.88 | 11.95 | 12.24 | 12.07 |
| 7. N60K80 + fer. (top dr.) | 11.53 | 10.81 | 11.86 | 12.15 | 12.56 | 11.78 |
| 8. N90K120 + fer. (top dr.) | 11.65 | 11.66 | 11.71 | 12.09 | 11.89 | 11.80 |
| Average B         | 11.22 | 11.57 | 11.84 | 12.26 | 12.09 |

LSD_{0.5} A = 0.62, LSD_{0.5} B = 0.49, LSD_{0.5} AB = 1.40

An important indicator that has a strong influence on the taste of apples (to a greater extent than sugar) is acidity, moreover, not the total acid content, but the titrated acidity due to the presence of free acids. In apple fruits it is malic acid. Under the influence of
meteorological conditions of the growing season and the place of growth, the titrated acidity varies more widely than the sugar content.

The average content of titrated acids in the fruits of the Veniaminovskoe cultivar varied significantly – depending on the option and the research year from 0.31 to 0.86% and on average over the years - from 0.41 to 0.78% (Table 4).

**Table 4.** The content of **titrated acids** (%) in the fruits of the Veniaminovskoe apple cultivar when applying fertilizers, 2017-2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|------------------|-----------------------------|-----------|
| 1. Control       | 2017 0.76 2018 0.54 2019 0.41 2020 0.41 2021 0.45 | 0.52      |
| 2. N30 K40       | 2017 0.78 2018 0.42 2019 0.36 2020 0.44 2021 0.56 | 0.51      |
| 3. N60 K80       | 2017 0.78 2018 0.46 2019 0.38 2020 0.46 2021 0.49 | 0.52      |
| 4. N90 K120      | 2017 0.86 2018 0.58 2019 0.46 2020 0.52 2021 0.59 | 0.60      |
| 5. Control + fer.(top dr.) | 2017 0.75 2018 0.52 2019 0.41 2020 0.46 2021 0.57 | 0.54      |
| 6. N30K40 + fer. (top dr.) | 2017 0.78 2018 0.46 2019 0.37 2020 0.48 2021 0.58 | 0.53      |
| 7. N60K80 + fer. (top dr.) | 2017 0.83 2018 0.51 2019 0.39 2020 0.53 2021 0.56 | 0.56      |
| 8. N90K120 + fer. (top dr.) | 2017 0.74 2018 0.56 2019 0.49 2020 0.31 2021 0.57 | 0.53      |
| Average B        | 2017 0.78 2018 0.51 2019 0.41 2020 0.45 2021 0.55 | 0.55      |
| LSD0.5 A= 0.07   |                             | LSD0.5 B= 0.05 | LSD0.5 AB= 0.15 |

The highest significant content of titrated acids was in 2017 – 0.78%. The content of titrated acids significantly exceeding the control on average for five years of research was in the option with the introduction of the highest dose of nitrogen and potash fertilizers into the soil (N90K120). Foliar fertilizing with urea and potassium sulfate for five years did not affect the concentration of titrated acids in the fruits of the Veniaminovskoe cultivar.

According to long-term data, the Veniaminovskoe apple cultivar is not highly vitamin-rich in ascorbic acid, which accumulates in its fruits from 2.6 to 5.2 mg/100 g [9, 27]. During the research period, this indicator varied in the range of 2.60...7.70 mg/100 g and significantly changed under the combined influence of weather conditions during the growing season and soil nutrition conditions. The greatest accumulation of ascorbic acid was noted in 2017 and 2021. In 2019 and 2020, there was a significant decrease in this indicator (Table 5).

**Table 5.** Ascorbic acid content (mg/100 g) in fruits of the Veniaminovskoe apple cultivar when fertilizing, 2017-2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|------------------|-----------------------------|-----------|
| 1. Control       | 2017 6.00 2018 5.30 2019 2.60 2020 3.05 2021 4.85 | 4.36      |
| 2. N30 K40       | 2017 6.00 2018 3.05 2019 3.50 2020 3.05 2021 6.15 | 4.35      |
| 3. N60 K80       | 2017 6.55 2018 5.75 2019 2.60 2020 3.95 2021 2.70 | 4.31      |
| 4. N90 K120      | 2017 7.02 2018 6.15 2019 3.55 2020 5.25 2021 7.20 | 5.83      |
| 5. Control + fer.(top dr.) | 2017 5.55 2018 4.80 2019 3.95 2020 2.20 2021 6.35 | 4.57      |
| 6. N30K40 + fer. (top dr.) | 2017 5.20 2018 5.30 2019 4.85 2020 7.45 2021 5.25 | 5.53      |
| 7. N60K80 + fer. (top dr.) | 2017 6.50 2018 6.15 2019 3.05 2020 7.06 2021 7.70 | 6.09      |
| 8. N90K120 + fer. (top dr.) | 2017 4.00 2018 3.50 2019 3.95 2020 3.95 2021 7.00 | 4.48      |
| Average B        | 2017 5.85 2018 5.00 2019 3.50 2020 4.49 2021 5.90 |           |
There was a significant increase in the content of ascorbic acid in fruits in the options of joint application to the soil and foliar top dressing in doses of N$_{30}$K$_{40}$ and N$_{60}$K$_{80}$ in 2020. The content of ascorbic acid significantly exceeding the control for an average of five experimental years was found in the option with the introduction of nitrogen and potash fertilizers into the soil at a dose of N$_{60}$K$_{80}$ together with foliar top dressing. Foliar top dressing with urea and potassium sulfate in 2017, 2018, 2019, and 2021 had no effect on the concentration of ascorbic acid in the fruits of the studied variety.

In our experiment, phenolic compounds – leucoanthocyanins and catechins - were studied in the fruits of the Venyaminovskoe cultivar.

The average content of leucoanthocyanins depending on the year of research and the experimental option ranged from 110.2 to 521.5 mg/100 g (Table 6). During the period of 2017...2021, there was a tendency of increasing accumulation of leucoanthocyanins in fruits over the years from 143.7 to 377.9 mg/100 g.

**Table 6.** The content of leucoanthocyanins (mg /100 g) in the fruits of the Venyaminovskoe apple cultivar when fertilizing, 2017-2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|-------------------|-----------------------------|-----------|
|                   | 2017 | 2018 | 2019 | 2020 | 2021 |
| 1. Control        | 153.0 | 192.6 | 142.5 | 326.2 | 388.5 | **240.5** |
| 2. N$_{30}$K$_{40}$ | 134.0 | 110.2 | 245.0 | 422.0 | 177.5 | **217.7** |
| 3. N$_{60}$K$_{80}$ | 112.5 | 173.2 | 182.5 | 271.0 | 521.5 | **252.1** |
| 4. N$_{90}$K$_{120}$ | 157.8 | 196.2 | 270.0 | 327.1 | 422.4 | **274.7** |
| 5. Control + fer.(top dr.) | 173.2 | 156.2 | 271.5 | 377.5 | 525.0 | **300.7** |
| 6. N$_{30}$K$_{40}$ + fer. (top dr.) | 140.8 | 155.2 | 218.0 | 374.7 | 472.2 | **272.2** |
| 7. N$_{60}$K$_{80}$ + fer. (top dr.) | 146.2 | 163.3 | 211.5 | 356.6 | 268.6 | **229.2** |
| 8. N$_{90}$K$_{120}$ + fer. (top dr.) | 132.7 | 110.2 | 187.5 | 229.5 | 247.5 | **181.4** |
| Average B         | 143.7 | **157.1** | **216.0** | **335.5** | **377.9** |

In option 5 (control + foliar top dressing), the content of leucoanthocyanins significantly exceeded the control on average over the five years of the study and was significantly lower in options with the introduction of minimum mineral fertilizers’ doses N$_{30}$K$_{40}$ and the highest doses N$_{90}$K$_{120}$ together with foliar top dressing. In 2021, a significant decrease in the content of leucoanthocyanins was found in the option with a minimum dose of mineral fertilizers – N$_{30}$K$_{40}$.

Depending on the year and the research option, catechins in the fruits of the Venyaminovskoe cultivar were accumulated in the range from 74.9 to 252.0 mg/100 g and on average from 126.8 to 151.8 mg/100 g from year to year (Table 7).

In 2017, 2018, 2019, and 2020, the content of catechins in all experimental options with the use of mineral fertilizers was significantly higher compared to the control. In 2021, there was a significant decrease in catechins in the option with the use of mineral fertilizers at a dose of N$_{90}$K$_{80}$, as well as in the following options: without soil and minimal (N$_{30}$K$_{40}$) fertilization on the background of top dressing.
Table 7. Catechin content (mg/100 g) in fruits of the Veniaminovskoe apple cultivar when fertilizing, 2017-2021.

| Option (Factor A) | Years of research (Factor B) | Average A |
|-------------------|-----------------------------|-----------|
|                   | 2017 | 2018 | 2019 | 2020 | 2021 |         |
| 1. Control        | 83.0 | 120.9 | 101.8 | 92.3 | 252.0 | 130.0   |
| 2. N₃₀ K₄₀        | 112.4 | 245.0 | 136.0 | 108.1 | 137.5 | 147.8   |
| 3. N₆₀ K₈₀        | 126.0 | 129.3 | 120.5 | 176.1 | 103.3 | 131.0   |
| 4. N₉₀ K₁₂₀       | 183.8 | 136.2 | 172.0 | 117.0 | 245.3 | 170.9   |
| 5. Control + fer.(top dr.) | 132.1 | 134.1 | 135.3 | 112.9 | 92.8 | 121.4   |
| 6. N₃₀ K₄₀ + fer. (top dr.) | 156.0 | 105.8 | 142.5 | 132.8 | 74.9 | 122.4   |
| 7. N₆₀ K₈₀ + fer. (top dr.) | 151.7 | 122.5 | 165.1 | 144.7 | 124.6 | 141.7   |
| 8. N₉₀ K₁₂₀ + fer. (top dr.) | 140.4 | 87.0 | 148.7 | 130.8 | 184.0 | 138.2   |
| Average B         | 135.7 | 135.1 | 140.2 | 126.8 | 151.8 |         |
| LSD₀.₅ A          | 58.1 |
| LSD₀.₅ AB         | 129.8 |
| LSD₀.₅ B          | 45.9 |

4 Conclusions

According to the results of five-year studies, the use of soil and foliar application of nitrogen and potash fertilizers did not have a significant effect on the content of soluble solids, sugars, titrated acids, ascorbic acid, and phenolic compounds in the fruits of the Veniaminovskoe cultivar. It was established that the most significant factor affecting the chemical composition of fruits are the weather conditions of the growing season. At the same time, the content of ascorbic acid significantly exceeding the control over an average of five experimental years was found in the option with the introduction of nitrogen and potash fertilizers into the soil at a dose of N₆₀ K₈₀ together with foliar top dressing. There is a tendency of increasing accumulation of leucoanthocyanins in fruits over the years. In 2021, there was a significant decrease in catechins with the use of mineral fertilizers at a dose of N₆₀ K₈₀, as well as in the following options: without soil and minimal (N₃₀ K₄₀) fertilization against the background of top dressing.

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