Evaluation of garden strawberry varieties against biochemical parameters and genetic aroma determinants

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Abstract. The paper presents the results of a long-term (2015-2020) study into a collection of domestic and foreign strawberry varieties cultivated in the Central Black Region, in terms of biochemical composition (content of soluble solids (SS), sugars, organic acids, ascorbic acid, anthocyanins) and genetic aroma determinants. The varieties with a high SS content (above 12.0%) herewith include Alena, Divnaya, Kupchikha, Olympic hope, Torpedo, Flora; sugars (above 9.0%) – Alena, Lastochka, Olimpiyskaya Nadezhda, Flora, Privlekatelnaya; ascorbic acid (above 80.0 mg/100 g) – Divnaya, Kupchikha, Sudarushka, Festival chamomile; anthocyanins (above 100.0 mg/100 g) – Alena, Fireworks. Among the varieties addressed, the Kubata, Tsaritsa (domestic selection), Kimberly, Marshall, Red Gauntlet, Vima Tarda (foreign selection) varieties are characterized by a combination of functional alleles of FaOMT and FaFAD1 genes for a complex aroma profile. The selected promising varieties can be used as sources of valuable quality attributes and biochemical composition of fruits.

1. Introduction
Today, the use of berries for the food and processing industry is of great importance due to new varieties. The garden strawberry is one of the most popular berries in the Central Black Earth Region [1-3].

The strawberries (*Fragaria × ananassa* Duch.) are very popular from among berry crops. There are more than 20 species and 600 varieties of strawberries that come in different in size, color, taste, aroma and biochemical profile of the fruit [1].

Strawberries are rich in vitamin C and other antioxidant compounds such as vitamin E, β-carotene, as well as phenolic components (phenolic acids, flavonols, flavan-3-ols, anthocyanins). The presence of phenolic components largely determines the quality, nutritional value and sensory properties of fresh fruits and processed strawberries. Moreover, the majority of strawberry varieties cultivated in the EU and beyond just slightly differ in biochemical composition of fruits, though with only a few of them to combine a high level of fruit quality with an increased accumulation of biologically active substances [2-5].

An important consumption attribute speaking well for the quality of strawberry fruits is their aroma, which is due to a large amount of volatile aromatic organic substances, the number of which exceeds 360 [6, 7]. An aromatic profile of strawberry fruits is greatly shaped by about 20 compound that include mesifuran (fruit and caramel aroma) and γ-decalactone (peach-like, fruity, sweet aroma)
The content of mesifuran in strawberry fruits is controlled by the \textit{FaOMT} gene [7], whereas \(\gamma\)-decalactone – by the \textit{FaFAD1} gene [5].

Many modern industrial strawberry varieties have a weakly expressed fruit aroma due to lower genetic diversity of alleles that control the content of volatile aroma-forming substances [8, 9]. With this in view, improving the biochemical composition and preserving the aroma of fruits is one of the challenges to be addressed by modern strawberry breeding programs.

The paper aims to determine the rate of core food and biologically active components accumulated in strawberry fruits under the conditions of the Central Black Earth Region, to identify genes for the aroma of fruits and to isolate valuable genotypes – sources of their high content.

2. Materials and methods

The studies were carried out in 2015-2020. The objects of research were 32 strawberry varieties of various genetic origins. Biochemical analyzes of fruits were carried out in accordance with standard methods: content of soluble solids – through refractometric method (RX-5000i refractometer, Atago, Japan); the content of sugars, organic acids, ascorbic acid – by titrimetric methods (automatic G20S titrator of the Titration Compact series, METTLER TOLEDO, Switzerland); anthocyanins – by the method of pH differential spectrophotometry (Genesy 10uv spectrophotometer, Thermo, USA) [10, 11]. Statistical processing of the results was carried out using the Microsoft Excel 2016 software package. The calculation included the determination of the sample mean, standard error, median, 25\textsuperscript{th} and 75\textsuperscript{th} percentiles.

The \textit{FaOMT-SI/NO} marker [7] was used to assess the allelic state of the \textit{FaOMT} gene, and the \textit{FaFAD1} marker was used for the \textit{FaFAD1} gene [5]. The reaction mixture in a total volume of 15 \(\mu\)l contained 1.5 mM Taq buffer, 2.0 mM deoxynucleoside triphosphate mixture, 2.5 mM magnesium chloride, 0.2 U Taq polymerase, 0.2 \(\mu\)M of each primer, and 20 ng of genomic DNA. Amplification was provided by a T100 thermal cycler manufactured by BIO-RAD in the modes described in the original publications [5, 7]. Amplification products were separated by electrophoretic method in agarose gel (agarose concentration – 2\%, buffer system – 1x TBE). Amplicons were sized with a Gene Ruler 100 bp DNA Ladder (Thermo Fisher Scientific).

3. Results and Discussion

The target varieties greatly differed in the way in which they accumulated the core chemical components in fruits (Table 1). Soluble solids varied from 8.0\% (Kubata) to 14.9\% (Alena) with an average value of 10.9\%. The highest content of soluble solids (11.8\%, which is above the 75\textsuperscript{th} percentile) was found in the Alena, Divnaya, Kupchikha, Olimpiyskaya Nadezhda, Privlekatelnaya, Torpeda, Flora cultivars. The fresh and processed fruits of these varieties can be consumed.

The average accumulation of sugars in the target varieties was 7.7\% with a minimum content of 5.7\% (Kubata), a maximum – 10.7\% (Alena). High-sugar varieties (accumulating more than 8.4\% of the total sugar, which is above the 75\textsuperscript{th} percentile) include the following varieties: Alena, Lastochka, Olimpiyskaya Nadezhda, Privlekatelnaya.

The accumulation of organic acids in fruits varied from 0.67\% (Alena) to 1.31\% (Zenith) with an average value of 1.0\% for varieties. 46.9\% of the target varieties had the optimal acidity, not exceeding 1.0\%.

The amount of ascorbic acid ranged from 31.2 (Troitskaya) to 95.2 mg/100 g (Sudarushka), with an average value to make up 66.1 mg/100 g. Pursuant to breeding requirements, modern strawberry varieties should have ascorbic acid not less than 80-100 mg/100 g. Thus, 12.5\% of the target varieties correspond to this criterion, including Divnaya, Sudarushka, Festival chamomile, Marshall.

The variations in the amount of anthocyanins in fruits with an average value of 54.6 mg/100g ranged from 17.1 (Olimpiyskaya Nadezhda) to 107.4 mg/100g (Fireworks), i.e. the differences in varieties for this trait reached more than 6 times. More than half (53.1\%) of the target varieties accumulated anthocyanins less than 50.0 mg/100 g. The number of genotypes with an anthocyanin content in the range of 50.0-80.0 mg/100 g was 37.5\%. High (80.0-100.0 mg/100 g) and very high
(over 100.0 mg/100 g) accumulation of anthocyanins was found to be present in 3.1% and 6.3% of the varieties, respectively. The Alena, Torpeda and Fireworks varieties had a dark fruit color and, consequently, a high content of anthocyanins (above 90.0 mg/100 g). Moreover, by this indicator, the Privlekatelnaya (anthocyanin content 76.8 mg/100 g), Krymchanka 87 (73.6 mg/100 g), Flora (74.2 mg/100 g) varieties are of interest.

Table 1. The chemical profile of strawberries (2015-2020)

| Variety          | Soluble solids, % | Amount of sugar, % | Titratable acidity, % | Vitamin C, mg/100 g | Anthocyanin, mg/100 g |
|------------------|-------------------|--------------------|-----------------------|---------------------|-----------------------|
| Alena            | 14.9              | 10.7               | 0.67                  | 51.9                | 124.5                 |
| Bohemia          | 10.4              | 6.6                | 1.07                  | 58.5                | 24.8                  |
| Divnaya          | 12.8              | 7.6                | 1.0                   | 86.4                | 21.4                  |
| Zenith           | 11.0              | 7.8                | 1.31                  | 65.8                | 66.7                  |
| Krymchanka 87    | 9.8               | 6.9                | 1.10                  | 49.1                | 73.6                  |
| Kubata           | 8.0               | 5.7                | 1.0                   | 54.2                | 50.1                  |
| Kupchikha        | 12.4              | 6.3                | 0.60                  | 89.6                | 59.7                  |
| Lastochka        | 11.6              | 8.9                | 0.63                  | 56.6                | 44.3                  |
| Neznakomka       | 10.4              | 7.3                | 0.97                  | 71.6                | 67.3                  |
| Olimpiyskaya     | 13.5              | 9.8                | 0.93                  | 75.3                | 17.8                  |
| Nadezhda         |                   |                    |                       |                     |                       |
| Privlekatelnaya  | 11.9              | 9.0                | 0.98                  | 78.6                | 76.8                  |
| Sudarushka       | 11.0              | 8.2                | 1.30                  | 95.2                | 44.6                  |
| Torpeda          | 12.7              | 9.4                | 1.17                  | 74.3                | 93.6                  |
| Troitskaya       | 10.5              | 7.4                | 0.91                  | 31.2                | 25.4                  |
| Urozhainaya CGL  | 11.4              | 8.0                | 1.03                  | 61.7                | 43.7                  |
| Fireworks        | 10.3              | 7.0                | 0.88                  | 56.4                | 107.4                 |
| Festival         | 10.2              | 7.6                | 1.30                  | 78.0                | 35.2                  |
| Festival chamomile| 10.1              | 7.1                | 1.10                  | 80.0                | 42.6                  |
| Flora            | 12.6              | 9.2                | 1.04                  | 65.0                | 74.2                  |
| Tsaritsa         | 11.6              | 8.2                | 1.02                  | 65.1                | 59.9                  |
| Tsarskoselskaya  | 9.5               | 7.1                | 1.33                  | 67.6                | 42.3                  |
| Yarkaya          | 11.8              | 8.6                | 1.09                  | 61.3                | 40.3                  |
| Elianny          | 9.1               | 6.7                | 0.68                  | 33.2                | 33.4                  |
| Gigantella Maxim.| 9.6               | 6.5                | 0.97                  | 68.4                | 68.5                  |
| Korona           | 10.5              | 7.4                | 1.06                  | 64.6                | 48.3                  |
| Kimberly         | 11.2              | 8.4                | 0.94                  | 68.5                | 34.5                  |
| Maryshka         | 10.7              | 7.4                | 1.10                  | 66.8                | 60.7                  |
| Marshall         | 11.0              | 7.1                | 0.99                  | 85.0                | 70.9                  |
| Red Gauntlet     | 9.0               | 6.6                | 1.02                  | 65.4                | 37.4                  |
| Troubadour       | 8.8               | 6.7                | 1.10                  | 62.4                | 48.7                  |
| Vima Zanta       | 10.4              | 7.7                | 0.95                  | 59.1                | 45.7                  |
| VimaTarda        | 9.6               | 6.6                | 1.01                  | 68.8                | 61.9                  |
| Average (M±m)    | 10.9±0.26         | 7.7±0.20           | 1.0±0.03              | 66.1±2.49           | 54.6±4.27             |
| Median           | 10.6              | 7.4                | 1.02                  | 65.6                | 48.5                  |
| Percentile 25th  | 9.9               | 6.8                | 0.94                  | 58.7                | 38.1                  |
| 75th             | 11.8              | 8.4                | 1.10                  | 75.1                | 68.2                  |

In the collection of strawberry addressed, the *FgOMT* gene was identified in 87.5% of the varieties, while the *FaFAD1* gene – in 25.0% of the varieties. The Olimpiyskaya Nadezhda and Sudarushka varieties that accounted for 4.9% of the total number of the target varieties had no functional alleles of both loci (Table 2).
Table 2. Grouping of strawberry varieties by the presence of genes for the aromatic set of fruits

| Analyzed loci are absent | Alleles of genes are present | FaOMT | FaFAD1 | FaOMT+FaFAD1 |
|-------------------------|-------------------------------|-------|--------|--------------|
| Olimpiyskaya Nadezhda, Sūdarushka | Alena, Bohemia, Divnaya, Zenith, Krymchanka 87, Lastochka, Neznakomka, Privlekatelnaya, Torpeda, Troitskaya, Urozhainaya CGL, Fireworks, Festival, Festival chamomile, Flora, Tsarskoselskaya, Yarkaya, Elianny, Korona, Maryshka, Troubadour, Vima Zanta | Kupchikha, Gigantella Maxim | Kubata, Tsaritsa, Kimberly, Marshall, Red Gauntlet, Vima Tarda |

In our earlier studies, the FaOMT gene was detected in 86.1% [12] and 93.7% [13] of the target varieties, while the FaFAD1 gene – in 25.6% [12] and 23.8% [14] genotypes, which correlates with the results presented.

Among the domestic strawberry, the proportion of varieties with the FaOMT and FaFAD1 genes was 85.7 and 14.3%, respectively. Among the foreign strawberry, the target genes were identified in 90.9% and 45.4% of the varieties.

The aroma of strawberries depends on a combination of many volatile aroma-forming substances, the content of which is attributed to various genetic factors. In this regard, the most promising varieties by a “fruit aroma” attribute are genotypes that combine several genes of aroma in the genome. Among the target varieties, the combination of functional alleles of the FaOMT and FaFAD1 genes characterizes the Kubata, Tsaritsa varieties (domestic selection), the Kimberly, Marshall, Red Gauntlet, Vima Tarda (foreign selection) varieties.

4. Conclusion

Following a review of the collection of strawberry varieties, genotypes with a high level of accumulation of the core biochemical components were identified: sugars (more than 9.0%) – Alena, Lastochka, Olimpiyskaya Nadezhda, Flora, Privlekatelnaya, ascorbic acid (more than 80.0 mg/100 g) – Divnaya, Kupchikha, Sūdarushka, Festival chamomile, anthocyanins (more than 100.0 mg/100 g) – Alena, Fireworks. The Kubata, Tsaritsa, Kimberly, Marshall, Red Gauntlet, Vima Tarda varieties have a combination of functional alleles of the FaOMT and FaFAD1 genes for the aromatic set of fruits.

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