The content and characteristics of the accumulation of carotenoids in the grain of modern corn hybrids

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Abstract. The studies were carried out in 2018–2019 in the Belgorod region to identify hybrids and parental forms of corn with increased content of carotenoids in the grain. The material for the study was 4 domestic hybrids, 3 hybrids of the selection of Maizadur, 1 - Syngenta, 1 - Monsanto, 1 - Woodstock, standard - Belkorn 250 MB, as well as 4 parental components of the Decent SV hybrid. During the experiment, agricultural practices traditional for the region were used. The harvesting of the registration plots was carried out manually, considering the weight of the cobs and the harvesting moisture content of the grain. The content of carotenoids in grain was determined according to the developed method in terms of lutein. In 2018, a preliminary screening of a sample of hybrids was carried out. All studied hybrids, except for GIS 1 and GIS 3, surpassed the Belkorn 250 SV standard. The concentration of carotenoids in two domestic hybrids Resource SV (18.4 mg/kg) and Decent SV (19.8 mg/kg) surpassed the hybrids of the Maizadur selection by 35.9 and 40.4% of the Syngenta selection by 8.7 and 15.2%, Monsanto - by 26.6 and 31.8%, Woodstock - by 8.2 and 14.6%, respectively. On average, over two years, the highest content of carotenoids in grain and their harvest per unit area was observed for the corn hybrid Decent DM (21.8 mg/kg and 169.4 g / ha, respectively), in which the values of these indicators exceeded the Belkorn 250 MV standard in 2 times. Among the parental components, the self-pollinated lines EM 10 ЗС (26.6 mg/kg) and SB 5-1 SV (29.7 mg/kg) were characterized by a high concentration of carotenoids - the parental components of the Decent SV hybrid.

1. Introduction
The value of corn as a fodder crop has been known for a long time. Feed mixtures containing corn are high-energy products for feeding animals and poultry in specialized agricultural sectors. Modern selection and genetic research programs of domestic and foreign scientific centers, aimed at improving the quality indicators of grain, contribute to improving the fodder qualities of corn as a source of natural fodder [1, 2].

The grain used for feed purposes is of particular value if it is characterized by improved quality characteristics. With the help of breeding, you can improve the quality of the corn grain - starch, fat, sugars, protein, and carotenoids. Carotenoids as substances that play a special role in the metabolism of animals and humans are of interest for improving the corn grain complex.

The color of corn kernels is largely due to the presence of carotenoids - pigments of red, orange, and yellow colors [3]. At the beginning of the 20th century, an increase in weight gain was noted in
the USA due to the use of yellow-grain corn for feeding animals, in comparison with white-grain corn [4]. Feed enriched with provitamin A has a high effect on the productivity and safety of poultry, the rate of egg-laying, an increase in the average egg weight, and the saturation of yolk color [5]. At the same time, the accumulation of carotenoid pigments in corn grain depends to a large extent on the genotype [6].

The research aims to identify hybrids and parental forms of corn with increased content of carotenoids in the grain.

2. Materials and methods

The studies were carried out in 2018–2019, in the Belgorod region. To create hybrids of corn with increased content of carotenoids, several original self-pollinated lines were isolated in 2009. In targeted crossing in 2010, the best lines of the 6th inzukht (I6) were involved as maternal and paternal components of three-line hybrids, the best of which (Effective SV, Resource SV, Stable SV, and Decent SV) in 2013-2017 successfully passed the State test and are included in the register of breeding achievements approved for use in the Central Black Earth Region.

The material for the study was the indicated hybrids, their parental components, as well as samples of foreign selection (3 hybrids of the Maizadur company, 2 - Syngenta, 1 - Monsanto, 1 - Woodstock). The hybrid Belkorn 250 MB was used as a standard. In 2018, screening of maize hybrids of domestic and foreign selection was performed for the quantitative content of carotenoids in grain. In 2019, the study involved original maize hybrids and their parental components, distinguished by the number of carotenoids, for comparative analysis. Experiments were set up, records and observations were carried out according to the generally accepted method. Repetition - two, two-row plots with an area of 9.8 m2.

The soil of the experimental site is typical, medium-thick, low-humus, heavy loamy chernozem on loess-like loam with a humus content (according to Tyurin) 4.7 ... 5.6%. pH of the salt extract 5.8 ... 6.3, the content of mobile phosphorus and exchangeable potassium (according to Chirikov) - 67 ... 78 and 88 ... 112 mg/kg of soil, respectively, the degree of saturation with bases - 90%.

In the fall, after harvesting the predecessor (winter wheat), processing was carried out with a disk mulcher DM-3,2 and plowing with a PLN 5-35 plow to a depth of 22 ... 24 cm. In spring, when the physical ripeness of the soil was reached, harrowing was carried out with light harrows BZSS-1 to close the moisture; the introduction of azophoska (16:16:16) at a dose of 400 kg/ha using the SZ-3.6 seeder; pre-sowing cultivation to a depth of 5 ... 7 cm using the AKSH-6 cultivator. Sowing (May 5) was carried out with a special selection seeder with six portioned cone seeding devices. Immediately after sowing, soil herbicide Difiline was applied at a dose of 1.6 l/ha. In the phase of 5 leaves, the insurance herbicides Dublon (1.5 l/ha) and Ballerina (0.4 l/ha) were used in the complex, after 2 leaves, single inter-row cultivation was carried out without hilling KRN-5.6. To control the European corn moth (Ostrinia nubilalis), the natural entomophage of the genus Trichogramma (T. euproctidis) was dispersed twice at a dose of 1 g/ha.

Harvesting was carried out manually with the determination of the weight of the ears on the plot. The moisture content of the grain was determined using an average sample of three ears using a Wile-65 moisture meter, the content of carotenoids in corn seeds was determined according to the developed method in terms of lutein (Tretyakov M. Yu., 2010).

The obtained experimental data were statistically processed by the method of one-way analysis of variance (Dospekhov, 1985) on a personal computer using the Statistica 6.0 program.

In 2018, the growth and development of maize took place against the background of an increased sum of effective temperatures - 1723.7 °C (123% of the average annual) and an abundance of precipitation - 362.8 mm (138% of the norm).

Good thermal and moisture supply in the first and second ten days of May made it possible to obtain seedlings at the optimum time (8 ... 10 days). In the period from germination to flowering, climatic indicators fluctuated slightly, without causing stress in corn plants. Flowering took place against the background of a combination of a large amount of precipitation (515.5% of the average...
annual) with the sum of effective temperatures close to normal. Filling the grain (2d ... 3d decade of August) and grain ripening (1st ... 2d decade of September) the temperature was above average, precipitation was almost completely absent. In general, the conditions of the growing season in 2018 were favorable for the growth and development of corn plants. A feature of the year was the possibility of selecting maize genotypes capable of withstanding arid conditions at the final stage of grain filling (Table 1).

Table 1. Distribution of the sum of effective temperatures and precipitation during the growing season of maize (2018)

| Month    | Decade | The sum of effective temperatures, °C | Precipitation, mm |
|----------|--------|--------------------------------------|-------------------|
|          |        | Average 2001-2017 | 2018 | Average 2001-2017 | 2018 |
| May      | I      | 57,6 | 119,8 | 11,6 | 2,0 |
|          | II     | 68,2 | 96,3 | 15,6 | 31,9 |
|          | III    | 98,2 | 106,1 | 25,0 | 10,5 |
| July     | I      | 110,1 | 123,7 | 20,1 | 6,0 |
|          | II     | 125,7 | 117,1 | 27,0 | 217,1 |
|          | III    | 138,3 | 165,7 | 19,8 | 24,3 |
| August   | I      | 126,4 | 144,6 | 9,5 | 0,0 |
|          | II     | 118,3 | 138,6 | 15,3 | 0,0 |
|          | III    | 104,0 | 143,8 | 15,0 | 0,0 |
| September| I      | 70,0 | 124,5 | 12,8 | 2,0 |
|          | II     | 59,8 | 90,1 | 10,9 | 0,5 |
|          | III    | 39,2 | 37,4 | 19,9 | 23,5 |
| Amount   |        | 1400,5 | 1723,7 | 262,7 | 362,8 |

The corn vegetation in 2019 took place in contrasting conditions. The sum of effective temperatures exceeded the average annual rate by 10% and amounted to 1571.9°C (Table 2). The period of increased temperatures was observed from the 3rd decade of May to the 3rd decade of June, which, against the background of a shortage of precipitation, led to the manifestation of signs of air and soil drought.

During the flowering period of maize plants (II decade of July), the situation with moisture supply, due to precipitation and a temporary decrease in temperature, was generally favorable. After flowering, a simultaneous increase in temperature and a decrease in precipitation was noted (2d, 3d August, and the first ten days of September) in comparison with the average long-term indicators. This combination of weather factors negatively affected the drought-sensitive maize genotypes and led to a drop in the inherent potentials of grain products, and in drought-resistant forms, it allowed the formation of a good grain yield in combination with reduced harvest moisture. The conditions of 2019 contributed to the realization of productivity by the genetic system of drought tolerance.
Table 2. Distribution of the sum of effective temperatures and precipitation during the growing season of maize (2019)

| Month | Decade | The sum of effective temperatures, °C | Precipitation, mm |
|-------|--------|--------------------------------------|-------------------|
|       |        | Average 2001-2017 | 2018 | Average 2001-2017 | 2018 |
| May   | I      | 61,0 | 52,5 | 11,1 | 19,2 |
|       | II     | 69,8 | 72,6 | 16,5 | 3,5 |
|       | III    | 98,6 | 128,2 | 24,2 | 30,7 |
| June  | I      | 81,9 | 132,6 | 14,9 | 7,0 |
|       | II     | 97,1 | 153,2 | 17,5 | 4,5 |
|       | III    | 107,4 | 138,0 | 26,9 | 1,0 |
| July  | I      | 110,8 | 96,9 | 19,3 | 22,4 |
|       | II     | 125,2 | 104,8 | 37,6 | 3,2 |
|       | III    | 139,8 | 139,5 | 20,0 | 41,9 |
| August| I      | 127,4 | 92,6 | 9,0 | 2,6 |
|       | II     | 119,5 | 129,4 | 14,5 | 0,0 |
|       | III    | 106,2 | 123,9 | 14,2 | 0,0 |
| September | I | 73,0 | 116,3 | 12,2 | 0,0 |
|       | II     | 61,5 | 62,1 | 10,3 | 3,8 |
|       | III    | 39,1 | 29,7 | 20,1 | 26,3 |
| Amount|        | 1418,4 | 1571,9 | 268,2 | 166,1 |

3. The study of the content of carotenoids

The analysis of the results of exploratory screening of the quantitative content of carotenoids of the grain complex in maize hybrids, carried out in 2018, showed significant superiority of the original hybrids of the selection of Belgorod Federal Agrarian Scientific Center of the Russian Academy of Sciences over the Belkorn 250 MV standard (LSD05 = 3.3), as well as over most hybrids foreign selection (Fig. 1).

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The concentration of carotenoids in domestic hybrids Resource SV (18.4 mg/kg) and Decent SV (19.8 mg/kg) was higher than in the samples of the Maizadur selection, by 35.9 and 40.4%. Syngenta - by 8, 7, and 15.2%, Monsanto - by 26.6 and 31.8%, Woodstock - by 8.2 and 14.6%, respectively.

Two-year trials of the Decent CB hybrid and its parental components confirmed the results of the exploratory screening. At the same time, significant differences were revealed in the total content of carotenoids in parental components and hybrids created on their basis. The smallest expression of the trait, on average, was distinguished by the maternal form of a simple corn hybrid Orion C - self-pollinated line Bq 8-1 C (11.8 mg/kg), the greatest - paternal forms of hybrids Orion C and Decent SV - EM 10 zC (26.6 mg/kg) and SB 5-1 CB (29.7 mg/kg), respectively (Table 3). A comparative analysis of these maize hybrids and their crossing components suggests that the paternal self-pollinated lines EM 10 3C and SB 5-1 CB make a significant contribution to the formation of an increased concentration of grain complex carotenoids in the Decent SV hybrid.
Figure 1. The content of carotenoids in the grain of domestic and foreign maize hybrids, mg / kg (in terms of lutein), 2018

Table 3. The content of carotenoids in the grain complex of the corn hybrid Decent SV and its parental components (in terms of lutein)

| Parent component / hybrid | The number of carotenoids, mg / kg grain | 2018  | 2019  | average |
|--------------------------|-----------------------------------------|-------|-------|---------|
| Bq 8-1 S (R1)            |                                         | 10.6  | 13.1  | 11.8    |
| EM 10 3C (P2)            |                                         | 25.8  | 27.4  | 26.6    |
| Orirn S (P1 × P2)        |                                         | 18.7  | 21.5  | 20.1    |
| SB 5-1 SV (P3)           |                                         | 27.1  | 32.3  | 29.7    |
| Decent SV (P1 × P2) × P3 |                                         | 19.8  | 23.8  | 21.8    |
| Average                  |                                         | 20.4  | 23.6  |         |
| LSD_0.05                 |                                         | 6.1   | 4.7   |         |

Along with the study of the number of carotenoids in the grain of the studied maize hybrids, their yield per unit area, depending on the size of the yield, is of interest. On average, over 2 years of research, as a result of the prevailing arid conditions that fell on the critical phases of plant development, no significant differences in grain productivity were noted.

However, the quantitative collection of carotenoids per unit area varied greatly (\(V = 25.0\%\)). Thus, in hybrids Effective SV and Stable SV, it was higher than that of the standard, by 36.9 and 39.4\%, respectively. The Resource SV hybrid exceeded the standard in terms of this indicator by 1.8 times, Decent SV - by 2.0 times (Table 4).

It should be noted that the maternal form of the Effective SV, Resource SV, and Stable SV hybrids was a highly productive sterile simple hybrid - Sirius C. At the same time, they differ in the content of carotenoids, which is probably due to the influence of the paternal form.

To confirm the assumptions about the peculiarities of the formation of the total amount of carotenoids during hybridization of maize and to identify the sources of this trait in 2018–2019, a comparative analysis of the parental components of the hybrids was carried out. The accumulation of carotenoids in 2018 was less intense than in 2019, which is possibly caused by the prevailing meteorological conditions. At the same time, the maxima and minima of their content in parental components and maize hybrids did not change over the years as a whole. The largest amount of
carotenoids was noted in the grain of the self-pollinated line EM 10 3C (26.6 mg/kg), the lowest in the self-pollinated line Bq 207 C (9.0 mg/kg). The maternal form, a simple hybrid Sirius C, and the paternal form of the three-line hybrid ResourceVY SV, a self-pollinated line RVP 12-1 SV, 22.5 and 21.9 mg/kg, respectively, were characterized by increased accumulation.

Table 4. Grain harvest of corn hybrids and collection of carotenoids per unit area (in terms of lutein)

| Hybrid             | Grain yield, t / ha | The number of carotenoids in grain, mg / kg | Harvest of carotenoids, g / ha |
|--------------------|--------------------|--------------------------------------------|--------------------------------|
|                    | 2018    | 2019    | average | 2018    | 2019    | average | 2018–2019 |
| Belkorn 250 MV (St.) | 8,5     | 6,9     | 7,7      | 9,7     | 11,9    | 10,8    | 84,7      |
| Effective SV       | 8,3     | 6,9     | 7,6      | 14,0    | 18,0    | 16,0    | 121,6     |
| Resource SV        | 8,9     | 6,4     | 7,7      | 18,5    | 21,1    | 19,8    | 152,5     |
| Stable SV          | 7,8     | 6,8     | 7,3      | 15,1    | 18,9    | 17,0    | 124,1     |
| Decent SV          | 8,5     | 6,9     | 7,7      | 19,8    | 23,8    | 21,8    | 169,4     |
| Average            | 8,6     | 6,8     | 7,6      | 15,4    | 18,7    | 17,1    | 130,8     |
| LSD05              | 0,79    | 0,68    | 0,7      | 2,36    | 3,41    | 2,6     | 6,6       |
| V, %               | 2,3     |         |          | 24,5    |         | 25,0    |           |

Table 5. The content of carotenoids in the grain complex of maize hybrids and their parental components (in terms of lutein)

| Parent component / hybrid | The number of carotenoids, mg / kg grain |
|---------------------------|----------------------------------------|
|                           | 2018 | 2019 | average |
| Bq 207 S (R1)             | 8,4  | 9,6  | 9,0     |
| EM 10 3C (P2)             | 25,8 | 27,4 | 26,6    |
| Sirius C (P1 × P2)        | 21,2 | 23,9 | 22,5    |
| RVP 23-3 SV (R3)          | 17,6 | 18,4 | 18,0    |
| RPV 12-1 SV (R4)          | 20,1 | 23,6 | 21,9    |
| Bq 108-2 SV (R5)          | 16,7 | 19,1 | 17,9    |
| Effective SV (P1 × P2) × P3 | 14,0 | 18,0 | 16,0    |
| Resource SV (P1 × P2) × P4 | 18,5 | 21,1 | 19,8    |
| Stable SV (P1 × P2) × P5  | 15,1 | 18,9 | 17,0    |
| Average                   | 17,5 | 20,6 | 19,1    |
| LSD05                     | 2,97 |     | 3,69    |

In general, the maternal form of a simple sterile hybrid "Sirius C" - Bq 207C with a total amount of carotenoids of 9.0 mg/kg, was inferior to the paternal EM 103C - 26.6 mg/kg. In turn, the paternal forms of three-line hybrids - RVP 23-3 SV and Bq 108-2 SV - were characterized by their lower content (18.0 and 17.9 mg/kg, respectively), and RVP 12-1 SV was comparable (21.9 mg/kg) with the maternal form of the Sirius C hybrid, which, as a result, affected the number of carotenoids in three-line maize hybrids - Effective SV, Stable SV, and Resource SV, respectively. This fact indicates the contribution of the paternal component of the hybrid to the accumulation of carotenoid pigments in the corn grain.
4. Conclusion
Thus, according to the results of the exploration studies carried out, a significant differentiation in the total amount of carotenoids in the grain among the corn hybrids cultivated in the region was revealed. New zoned maize hybrids, created at the Belgorod FASC RAS, significantly exceed the standard in terms of this indicator. On average, over two years, the highest content of carotenoids in grain and their harvest per unit area was observed for the corn hybrid Decent SV (21.8 mg/kg and 169.4 g / ha, respectively), in which the values of these indicators were higher than those of the Belkorn standard. 250 MV by 2 times.
As a result of a comparative analysis of the studied parental components that make up the formula of the hybrids of the Belgorod Scientific Center, significant differences were noted in the total content of carotenoids in the grain. It was the highest in the self-pollinated lines EM 10ЗС (26.6 mg/kg) and SB 5-1 SV (29.7 mg/kg) - the parental components of the Decent SV hybrid.

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