Productivity of biotechnological sugar beet hybrids

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Abstract. A brief overview of the state of research on selection of new constant biotechnological lines – donors of glyphosate resistance and prospects of creating biotechnological hybrids of sugar beet are presented. Experimental data on nature of inheritance of glyphosate resistance in different sugar beet genotypes (mmSxxzz, mmNxxzz, MMTOp, etc.) were obtained. For the first time, separate and joint fruit lines-donors with 90-100% glyphosate resistance were obtained. In competitive and environmental tests, the first glyphosate-tolerant biotechnological hybrid of sugar beet TG-944 (1385) exceeded statistically the standard of yield and sugar volume from 1 ha. With mechanized (combine) harvesting, the average scoring yield of the hybrid was 65.5 t/ha. The purchasing costs for herbicides to protect sugar beet from weeds when growing glyphosate-resistant hybrids, compared to conventional commercial hybrids per 1 hectare on average for 2017-2019, amounted to 2447 and 8869 rubles, respectively, with an economic efficiency of 6422 rubles per 1 hectare of sowing.

1 Introduction

Sugar beet refers to intensive farming crops that require significant money expenditures and material resources during cultivation [5, 6, 8].

The level of yield of root crops of new hybrids, which reflects not only the output of products per unit area, but also the level of development of agricultural technology remains the main indicator of intensification of crop production [4, 9, 10]. At the same time, the following activities play an important role:

- accelerated creation of new cost-effective hybrids, including biotechnological ones, obtained on the basis of modern methods of biotechnology and genetic engineering;
- organizations of the system of primary and reproductive seed production of new sugar beet hybrids;
- introduction of resource-saving agricultural technologies of production, ensuring the reduction of material costs of a pesticide burden on environment.

Traditional breeding has not exhausted its potential to increase the productivity of sugar beet, but it can no longer provide a radical restructuring of a plant. Therefore, the use of

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biotechnology methods to create greater genetic diversity and to select forms with target characteristics and properties is of particular importance [1, 2, 3, 7, 10].

In the Russian Federation, in 2012, tasks on genetic engineering were included in the Comprehensive Program of Biotechnology Development for the period up to 2020 – "BIO-2020".

On November 28, 2018, President of the Russian Federation Vladimir Putin signed Decree No. 680 on faster development of genetic technologies. The Government was instructed to develop and approve the Federal Scientific and Technical Program of Development of Genetic Technologies for 2019-2027 within three months. In April 2019, the Government issued Resolution No. 479 "On approval of the Federal Scientific and Technical Program of Genetic Technologies Development for 2019-2027", in which genomic editing of an organism, genetically modified organisms are legalized in our country and the goal is to create "biosafety and ensuring the technological independence of the country".

The relevance of our researches is that the greatest damage to productivity of sugar beet hybrids is caused by weeds. Sugar beet, in comparison with other field crops, is susceptible to weeds at all stages of growth and development. Herbicides, as strong inhibitors of metabolism, destroying weeds, are toxic to sugar beet for 7-10 days. Therefore, the problem of creating sugar beet hybrids resistant to herbicides should be recognized as one of the priorities.

The aim of our researches is to create sugar beet lines tolerant to glyphosate as resistance donors on the basis of domestic, previously obtained self-fertile separate O-type lines, and joint-fruited pollinators. The practical result is the production of cost-effective, glyphosate-resistant biotechnological hybrids, which when grown, significantly reduce the risks of harm to sugar beets and environment by reducing the herbicide load.

2 Materials and methods

The researches were carried out in accordance with the State task "To create fundamentally new forms of sugar beet".

At the first stage of the researches, the task was solved by classical selection and genetic methods to create new separate mm lines and joint MM lines with tolerance to cercosporosis and glyphosate.

At the second stage, selection work continued to study the biological and economic characteristics of genetically combinationally modified lines, their hybridization, comprehensive testing and selection of the most valuable hybrids. The decision was made to use the most productive and cost-effective hybrids after a thorough analysis of digital data.

When selecting the material and research methods for determining the genotype of sugar beet on the basis of glyphosate tolerance, Mendelian ideas about dominance and recessivity were used. It was assumed that tolerance was controlled by the dominant resistance gene and that RR is a homozygote by the dominant, rr is a homozygote by the recessive, and Rr is a heterozygote for tolerance. Plants with signs of tolerance to glyphosate were designated as "T-forms", for example, T-pollinators, TO-types, TMS-lines, TG-hybrids.

At the first stage of the researches there was used self-pollination of the assumed T-forms, in seed material of which the plants of the 1st and 2nd years of life either died or were preserved after spraying with glyphosate in certain concentrations. At the same time, it was assumed that constant lines of dominant (RR) plants can be obtained only by successive (at least three times) self-pollination, and by selection in generations to track their heterozygosity.

Plants of experimental T-forms, test hybrids and numbers (samples) from analyzing and saturating crosses were treated with glyphosate in the phase of the first and/or second pair of real leaves, and then in a greenhouse (or in a field, depending on a purpose of an experiment) in the phase of the testis rosette at the 2nd year of life. The dead specimens were assigned to the "rr" genotype. The surviving plants did not differ phenotypically and were most likely of the "Rr" or "RR" type by genotype. It was impossible to determine which genotypes prevailed among them, and the plants were selected for further research by their phenotype. The safety of plants was taken into account in 5, 7 and 10 days after spraying with glyphosate. Observations, accounting and analysis of digital data were carried out according to generally accepted methods. The test control (standard) in all experiments was the commercial hybrid Kuban MS 95.

3 Results and discussion

Table 1 shows the results of the competitive test of the most glyphosate-tolerant sugar beet hybrids in 2019. The hybrid with the catalog number 875 significantly exceeded the average rates for the experience, yield and sugar volume from 1 ha were 115 and 112%, respectively.

Table 1. Productivity of the most valuable TMS hybrids of sugar beet according to the competitive test in 2019

Catalog hybrid number	Density, th./ha	Productivity of root crops, biological		Sugar content		Sugar beet harvest		Glyphosate resistance %
		t/ha	in % to average on experience	%	in % to average on experience	t/ha	in % to average on experience	
931 MS12173xTOp3-99	97	72,6	109	17,4	100	12,6	109	86
941 TMS1-93xTOPKr-24	91	74,8	112	17,2	99	12,9	111	94
882 TMS8-93xTOp2-110	95	72,7	109	17,0	98	12,4	107	89
875 TMS8-93xTOp2-94	98	76,4	115	17,0	98	13,0	112	86
Average on experience	95	66,5	100	17,4	100	11,6	100	85
NSP 05	-	7,0	-	0,5	-	-	-	-

Note: In the competitive test (series 5), 36 TMS hybrids were studied, the repetition was sixfold, and the placement of plots was randomized.

Four biotechnological MS hybrids resistant to glyphosate with catalog numbers TG935 (1382), TG937 (1383), TG944 (1385) and TG946 (1386) were studied in the environmental test. The records were carried out in two terms on September 10 and October 8 with manual digging (Tab.2) and under mechanized combine harvesting on October 25 (Table 3).

When harvesting on September 10, the hybrid with the catalog number TG 944 (1385) exceeded the average values in terms of crop productivity, sugar content and sugar volume from 1 ha.

In the first period of harvesting, the ranging in rates among hybrids was:

on crop productivity 53,7 – 88,0 t/ha;

on sugar content 15,4 – 17,1 %;

on sugar volume 9,9 – 14,6 t/ha.

In the second harvest period the hybrid TG944 (1385) confirmed its productivity advantages, the ranging in rates was as follows:

on crop productivity 81,6 – 102,9 t/ha;

on sugar content 15,6 – 17,3 %;

on sugar volume 12,7 – 15,9 t/ha.

For the period from September 10 to October 8, the average increase in hybrids was: in productivity 26.2 t / ha (39%), sugar volume 4.2 t / ha (38%). There were no significant changes in quality of purified juice. The resistance of sugar beet hybrids to glyphosate varied from 87 to 96%.

The maternal form of TMS 8-93 and the paternal form – the pollinator TOp Kr-24 had the greatest impact on the level of productivity of hybrids.

Hybrids with catalog numbers TG 937 (1383) and TG 944 (1385), as the most resistant to glyphosate and showing high productivity, are planned to include them for study in environmental and production tests.

Table 2. Productivity of test glyphosate-tolerant MS sugar beet hybrids according to environmental testing data in 2019 (Biological productivity, manual digging of root crops at testing sites)

Catalog hybrid number	Combination of crossing, ecological station	Registration date	Density th./ha	Crop productivity biol.	Sugar, %	Sugar volume. t/ha	DB of juice, %	resistance to glyphosate, %
TG 935 (1382)	TMS 8-93xTOp 3-99 Experimental station, Gulkevichi	10.09	116	59,6	15,9	9,5	88,0	87
	LLC «Agrosakhar», field №34, the Uspensky district	10.09 08.10	115 88	73,0 81,6	15,4 15,6	11,2 12,7	87,8 87,5	91
Average			106	71,4	15,6	11,1	87,8	
TG 937 (1383)	TMS 3-127xTOp3-99 Experimental station, Gulkevichi	10.09	123	58,1	17,1	9,9	88,5	85
	LLC «Agrosakhar», field №34, the Uspensky district	10.09 08.10	122 110	76,0 102,9	15,5 15,8	11,8 16,3	88,1 87,5	96
Average			118	79,0	16,1	12,6	88,0	
TG 944 (1385)	TMS 8-93xTOp Kr24 Experimental station, Gulkevichi	10.09	123	53,7	16,1	8,7	88,2	96
	LLC «Agrosakhar», field №34, the Uspensky district	10.09 08.10	107 94	88,0 101,1	16,6 15,7	14,6 15,9	87,5 88,0	94
Average			108	81,0	16,1	13,1	87,9	
TG 946 (1386)	TMS 3127xTOpKr24 Experimental station, Gulkevichi	10.09	116	58,9	16,3	9,6	89,0	84
	LLC «Agrosakhar», field №34, the Uspensky district	10.09 08.10	111 115	74,0 90,0	16,2 17,3	12,0 15,6	88,1 88,5	87
Average			114	74,3	16,6	12,4	88,5	
Average on experience, 1st registration date		10.09	117	67,7	16,1	10,9	88,1	90
Average on experience, 2nd registration date		08.10	102	93,9	16,1	15,1	87,9	90

Table 3 shows the productivity of first glyphosate-tolerant sugar beet hybrids during combine harvesting.

Table 3. Productivity of the most valuable glyphosate-tolerant sugar beet hybrids according to the environmental test data in 2019. (Mechanized harvesting, credited crop productivity)

Catalog hybrid number	Combination of crossing	Registration date	Density th./ha	Crop productivity biol.	Sugar, %	Sugar volume. t/ha	DB of juice, %	resistance to glyphosate, %
ТГ 937 (1383)	TMS 3-127 x TOp 3-99	25.10	109	66,5	15,8	10,5	85,9	90,5
ТГ 944 (1385)	TMS 8-93 x TOpKr 24	25.10	107	64,6	16,7	10,8	86,4	92,3
Average on experience			108	65,5	16,2	10,6	86,1	91,4
Average on LLC «Agrosakhar»		25.10	-	54,0	-	-	-	0
Average on the Uspensky district (Agricultural review)		25.10	-	49,5	-	-	-	0
Average on Krasnodar Territory		25.10	-	52,6	-	-	-	0

The experiment was conducted in the foothill zone of Krasnodar Territory in the Uspensky district in LLC "Agrosakhar", field No. 34. The predecessor - winter wheat, sowing was carried out on April 24 with a seeding rate of 1.5 sowing units of seeds per 1 ha. The seeds were prepared manually in laboratory conditions of the experimental station. Spraying was carried out with the herbicide Total 480 in the development phase of sugar beet plants – two pairs of real leaves, at the rate of 2 liters per 1 ha. No re-spraying was required. The harvesting was carried out by a mechanized combine harvester "Greeme" on October 25. Sugar content was determined in 20 root samples in 3-fold repetition in the laboratory of the Uspensky Sugar Factory. The results of calculations carried out in the automated mode are presented in Table 5. The most fully used indicator – the average productivity of root crops (credited) in glyphosate-tolerant hybrids was significantly higher compared to the average yield on LLC "Agrosakhar" and the excess was 11.5 t/ha.

4 Conclusion

1. Applying traditional methods of genetics and breeding, we have obtained encouraging results in the creation of fundamentally new biotechnological lines and hybrids of sugar beet. The schemes of inheritance of glyphosate resistance in combination - capable separate and join lines are clarified.

2. The first glyphosate-tolerant sterile lines TMS 8-93 and TMS 3-127 were created.

3. A high degree of glyphosate resistance was confirmed in cercospore- resistant donor lines TOp 3-99 and TOpKr 24.

4. In competitive and environmental tests, the high combinational ability was shown by the separate lines TMS 8-93 and TMS 3-127.

5. In the environmental test with mechanized harvesting, the average test yield of biotechnological hybrids was 65.5 t/ha.

6. The cost of purchasing herbicides to protect sugar beets from weeds when growing glyphosate-resistant hybrids compared to conventional commercial hybrids per 1 ha in average for 3 years amounted to 2447 and 8869 rubles, respectively, with economic efficiency of 6422 rubles per 1 ha of sowing.

7. The possibility of increasing the economic efficiency of sugar beet production due to significant reduction in costs of purchasing herbicides, more effective control of weeds and high productivity of biotechnological hybrids are shown.

8. The results of step-by-step studies allow us to consider the methods of classical breeding in the process of creating biotechnological glyphosate-tolerant sugar beet hybrids as one of the most effective tools in hands of breeders in practical selection.

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