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"Federal Research Center for Agrobiotechnologies of the Far East Region of A.K. Chaika"

APPROVED

Director of FBSO "FRC of Agrobiotechnologies
of the Far East Region of A.K. Chaika"

_____Dr. A.N. Emelianov

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REPORT

OF SCIENTIFIC RESEARCH

"Study of Effectiveness of Bacterial Solution on Soybean Crops During Seed Treatment and Foliar Application upon Vegetation"

(Agreement No. 13-NIR, September 5th, 2017)

Research Leader:

Head of the Department of
Agronomy and Agrochemistry _____Dr. R.V. Timoshinov

Timiryazevsky town, 2018

LIST OF PERFORMERS

Performers:

Head of the Department	_____	Dr. R.V. Timoshinov
Senior Scientist	_____	E.J. Kushaeva
Scientist	_____	L.E. Babinec
Agrochemist of 2nd category	_____	A.A. Falileev
Seed Agronomist	_____	L.V. Ulenkova

BRIEF

Report of 13 pages, 11 tables, 1 figure.

BACTERIA, SOY, SEED TREATMENT, EFFECTIVENESS, GERMINATION, YIELD, PHYTOTOXICITY

CONTENT

INTRODUCTION

To successfully increase the yield of broadacre crops, it is necessary to have new products that would be more effective than existing ones. Recently, various rhizobacteria have been widely used to increase crop yields, improve nutrients delivery to the plants and increase resistance to adverse environmental factors on crops. However, when using new products, it is necessary to determine their phytotoxicity in different soil and climatic conditions on the basis of experimental scientific data on the effect of specific products on certain crops, as it varies significantly depending on the type of soil, acidity index, humus content and fertility level. To control the amount of phytotoxicity, a biomethod is used, which involves the use of test plants, which weigh and calculate the percentage changes to the control. Using the bioindication method, fairly accurate results are quickly obtained without expensive equipment and reagents.

For this purpose, in 2018 on the basis of the Federal Research Center for Agrobiotechnology of the Far East Region of A.K. Chaika" at the experimental fields of the Department of Agriculture and Agrochemistry carried out technological field studies of the bacteria provided by SOSBIO RESEARCH RUS LLC on soy in accordance with Appendix 3B to the contract No. 13-NIR dated September 5th, 2017 between SOSBIO RESERCH RUS LLC and FBSO "FRC of Agrobiotechnologies of the Far East Region of A.K. Chaika".

PROTOCOL OF THE FIELD TRIAL RESEARCH STUDY

1. Research study period: from May to October
2. Venue: Russian Federation, Primorsky Krai, Ussuriysk city, experimental fields of FBSO "FRC of Agrobiotechnologies of the Far East Region of A.K. Chaika"
3. Soil and climatic zone: 4th forest-steppe zone
4. Culture – soy
   4.1. Variety – Monsoon
   4.2. Seed planting rate - 100 kg / ha
   4.3. Date of Planting - May 24th
   4.4. Date of emergence of full shoots - June 9th
   4.5. Phase of plant development at the time of processing - 3rd true leaf
5. Type of scientific study: field production
6. Agrotechnics
   6.1. Soil - meadow-brown bleached, humus content - 3.3%, pHsol. - 5.3, P2O5 - 25 mg / kg, K2O - 80 mg / kg.
   6.2. Preceded by: wheat - plowing at 22-24 cm (plowing) MTZ-82 + PLN-3-35; closing moisture (harrowing) MTZ-1221 + SP-11U + BZTS-1.0; deep cultivation on 12-14 cm MTZ-82 + KRG-4.2; introduction of mineral fertilizers MTZ-82 + LRU-450; cultivation with rolling on 4-5 cm MTZ-1221 + KNK-7.2.
   6.3. Fertilizer - under cultivation before sowing, diammofoska N10R26K26 - 100 kg / ha, ammonium nitrate N34 - 100 kg / ha.
   6.4. Care measures - inter-row processing, introduction of a set of protective equipment soil application of the herbicide May 27th - Lapis lazuli 0.8 l / ha + Simba 1.2 l / ha; vegetation June 30th - Corsair 2.5 l / ha + Borey 0.1 l / ha + Allure 0.15 l / ha + Rakurs 0.3 l / ha, further July 6th foliar introduction of bacteria with a backpack sprayer, July 26th Spirit 0.4 l / ha
7. EXTREME WHETHER CONDITIONS - waterlogging in July-August
8. Plot size - 20 sqm
9. Number of replications - three.

10. Scheme of the experiment - the study of the effectiveness of the bacterial preparation on soybeans was carried out in accordance with the scheme of experimental study proposed by the customer (Table 1).

Table 1 – Scheme of Experimental Study

Experiment No.	Variant No.	Type of Treatment	Method of Treatment		Crop development stage	Dissolved liquid treatment rate, l/t, l/Ha	Plot size	Pure treatment rate, l/t(Ha)	Total pure treatment used, ml
			When	How					
1	1	Control	No treatment	-	(GS 00)	-	20	-	-
	2	Standard chemical treatment Oplot	Seed treatment before planting	Spraying	(GS 00)	10 l/t	20	0.5	1.5
	3	Standard chemical treatment Oplot + Bacteria	Seed treatment before planting	Spraying	(GS 00)	10 l/t	20	0.5 + 1	1.5+3
	4	Bacteria	Seed treatment before planting	Spraying	(GS 00)	10 l/t	20	1	3
2	1	Standard chemical treatment Oplot	Seed treatment before planting	Spraying	(GS 00)	10 l/t	20	0.5	1.5
	2	Bacteria	Upon Vegetation	Spraying	2-3 leaf	200 l/Ha	20	0.45	0.9

11. Accounting yield by harvester (accounted area 10 sqm), plant productivity by biometric analysis of the structure of the crop per 1 sqm.

RESULTS OF THE FIELD TRIAL RESEARCH STUDY

The weather conditions of 2018 were characterized by high air temperature, which was higher than the average long-term values throughout the growing season of soybeans. Precipitation in April, May and June was *21.9*; *110.9* and *75.4* mm respectively, which amounted to *54.8*; *217.5* and *93.1%* of the average annual indicators, and in July and August there was abundant precipitation and overmoistening of the soil. During this period, *138.8* and *347.7* mm fell, respectively, which amounted to *154.2* and *259.5%*. In September and October, the weather was dry and warm. Meteorological conditions during the growing season of 2018 are presented in Table 2.

Table 2 – Meteorological conditions during the growing season of crops in 2018 (data from the meteorological station of Timiryazevsky town)

Month	Decade	Air Temperature, t ^{0}C		Precipitation, mm	
		2018	Mean Annual Value	2018	Mean Annual Value
April	1	3.1	3.2	3.9	12
	2	6.6	5.9	18.0	11
	3	11.5	8.4	0.0	17
	Month	7.1	5.8	21.9	40
May	1	8.4	10.5	63.1	12
	2	16.2	11.8	9.8	18
	3	13.4	13.3	38.0	21
	Month	12.7	11.9	110.9	51
June	1	16.8	14.6	23.0	27
	2	13.5	16.0	25.0	31
	3	18.0	17.2	27.4	23
	Month	16.1	15.9	75.4	81
July	1	17.1	19.0	60.6	29
	2	21.0	20.1	69.9	31
	3	26.3	21.2	8.3	30
	Month	21.6	20.1	138.8	90
August	1	21.1	21.4	108.5	46
	2	20.6	21.2	70.2	43
	3	19.9	19.9	169.0	45
	Month	20.5	20.8	347.7	134
September	1	16.6	16.8	60.7	41
	2	16.6	14.9	12.9	38
	3	13.1	12.9	6.0	25
	Month	15.4	14.9	79.6	104
October	1	12.0	10.6	24.8	21

Studying the effect of bacteria on soybean plants is impossible without the use of a biological indication, which can be used to directly obtain information about the reaction of the studied organism to the treatments used. Bioindication allows to assess the condition of various parts of soybean plants after the application of the studied treatments. In this regard, a field experiment was laid to determine the effectiveness of bacteria in the soybean variety Monsoon.

Studies on the effect of treatment of soybean seeds with a bacterial preparation under the prevailing conditions in 2018 showed that, starting from the plant development phase of the 3rd true leaf, in the variant with the joint use of Oplot chemical fungicide and bacteria, the largest root system weight and root length are observed, the increase was respectively 12.5 and 12.6%. With further consideration of July 13th in the variants with the use of bacteria, inhibition of the accumulation of the mass of roots and the length of the roots was not revealed (Tables 3, 4).

Table 3 – Dynamics of weight change of the root system of soy

Variant	Weight of the root system		Inhibition of the mass of roots increase, %
	gram	% to Control	
Record Date June 27th (first trifoliate)			
Control (no treatment)	0.49	100	-
Oplot fungicide	0.32	65.3	-34.7
Oplot + Bacteria	0.35	71.4	-28.6
Bacteria	0.37	75.5	-24.5
Record Date July 6th (3rd leaf)			
Control (no treatment)	0.64	100	-
Oplot fungicide	0.47	73.4	-26.6
Oplot + Bacteria	0.72	112.5	**+12.5**
Bacteria	0.51	79.7	-20.3
Record Date July13th (branching 4-5 leaf)			
Control (no treatment)	0.64	100	-
Oplot fungicide	0.76	118.8	+18.8
Oplot + Bacteria	0.79	123.4	**+23.4**
Bacteria	0.79	123.4	**+23.4**

Table 4 – The dependence of the length of soybean root per treatment used

Variant	Average root length		Inhibition of the root growth, % (±)
	cm	% to Control	
Record Date June 27th (first trifoliate)			
Control (no treatment)	10.4	100	-
Oplot fungicide	8.6	82.7	-17.3
Oplot + Bacteria	8.5	81.7	-18.3
Bacteria	10.4	100	0
Record Date July 6th (3rd leaf)			
Control (no treatment)	14.3	100	-
Oplot fungicide	13.4	93.7	-6.3
Oplot + Bacteria	16.1	112.6	**+12.6**
Bacteria	14.6	102.1	+2.1
Record Date July13th (branching 4-5 leaf)			
Control (no treatment)	14.8	100	-
Oplot fungicide	15.2	102.7	+2.7
Oplot + Bacteria	16.2	109.5	**+9.5**
Bacteria	16.3	110.1	**+10.1**

The greatest increase in the mass of soybean aerial organs was noted in the phase of the first trifoliate leaf and branching during seed treatment with bacteria (Table 5). The use of the studied treatments had a negative impact on the height of soybean plants, however it is necessary to note the least negative impact of the bacteria used in its pure form (Table 6, Figure 1).

Table 5 – Dynamics of weight change of the aerial organs of soy

Variant	Weight of aerial organs		Inhibition of weight changes, % (±)
	gram	% to Control	
Record Date June 27th (first trifoliate)			
Control (no treatment)	1.83	100	-
Oplot fungicide	1.56	85.3	-14.7
Oplot + Bacteria	1.54	84.2	-15.8
Bacteria	1.56	85.3	**14.7**
Record Date July 6th (3rd leaf)			
Control (no treatment)	2.47	100	-
Oplot fungicide	1.81	73.3	-26.7
Oplot + Bacteria	1.93	78.1	-21.9
Bacteria	1.98	80.2	**-19.8**
Record Date July13th (branching 4-5 leaf)			
Control (no treatment)	3.38	100	-
Oplot fungicide	3.70	109.5	+9.5
Oplot + Bacteria	2.84	84.0	-16.0
Bacteria	3.84	113.6	**13.6**

Table 6 – Dynamics of height change of soybean plants

Variant	Height of Plants		Inhibition of plant growth, % (±)
	cm	% to Control	
Record Date June 27th (first trifoliate)			
Control (no treatment)	10.3	100	-
Oplot fungicide	8.0	77.7	-22.3
Oplot + Bacteria	8.4	81.6	-18.4
Bacteria	10.0	97.1	-2.9
Record Date July 6th (3rd leaf)			
Control (no treatment)	13.7	100	-
Oplot fungicide	11.0	80.3	-19.7
Oplot + Bacteria	11.2	81.8	-18.2
Bacteria	13.0	94.9	-5.1
Record Date July13th (branching 4-5 leaf)			
Control (no treatment)	16.4	100	-
Oplot fungicide	15.2	92.7	-7.3
Oplot + Bacteria	14.1	86.0	-14.0
Bacteria	15.8	96.3	**-3.7**

Figure 1 – Oplot fungicide is in the center of the plot, control is on the left-hand side, and bacterium treatment is on the right-hand side of the field trial experiment

The greatest field germination was noted for the control; however, the combined use of chemical fungicide and bacterial seed treatment provides the greatest safety of soybean plants for harvesting (Table 7). That, ultimately, a beneficial effect on the yield increase. Usage of bacteria together with chemical agent led to the highest yield of 16.3 centner / Ha (Table 8).

Table 7 – Field germination and preservation of soybean plants, Monsoon variety

Variant	Field germination, %	Number of plants, *thouthands/Ha*		Preservation of plants for harvesting, %
		Seedlings	Before harvesting	
Control (no treatment)	93.1	512	360	70.3
Oplot fungicide	80.9	445	330	74.2
Oplot + Bacteria	76.7	422	365	86.5
Bacteria	86.0	473	360	76.1

Table 8 – Soybean yield, Monsoon variety

Variant	Yield, *centners/Ha*	Deviation from the Control, ±	Increase delivered by bacteria, *centners/Ha*
Control (no treatment)	13.4	–	–
Oplot fungicide	14.7	+1.3	–
Oplot + Bacteria	16.3	+2.9	+1.6
Bacteria	15.0	+1.6	+1.6
$HCP_{(0,95)}$	1.1		

The combined use of bacteria and chemical fungicide on soybean of Monsoon variety led to an increase in the number of grains per plant and a mass of 1000 seeds (Table 9).

Table 9 – Biometric indicators of soybean plants, Monsoon variety

Variant	Plants height, cm	Bottom bean attachment height, cm	No. of beans per plant	No. of grains per plant	Weight of 1000 grains, *gram*
Control (no treatment)	94	17	23	41	178
Oplot fungicide	91	18	23	44	175
Oplot + Bacteria	92	16	23	46	179
Bacteria	93	16	24	45	167

The research study discovered that foliar application of bacteria during the growing cycle of soybean plants of Monsoon variety does not provide an increase in biometric indicators and yield (table 10, 11).

Table 10 – Biometric indicators of soybean plants treated by the bacterium during the growing cycle, Monsoon variety

Variant	Plants height, cm	Bottom bean attachment height, cm	No. of beans per plant	No. of grains per plant.	Weight of 1000 grains, gram
Control (no treatment)	94	17	23	41	178
Bacteria	97	16	24	42	175

Table 11 – The influence of the bacterial treatment of soybean during the growing cycle on the yield centners / Ha, Monsoon variety

Variant	No. of soybean plants before harvesting *thousands/Ha*	Yield, *centners/Ha*	Deviation from the Control, ± *centners/Ha*	Increase delivered by treatment, %
Control (no treatment)	360	13.4	–	–
Bacteria	350	13.3	-0.1	–
$HCP_{(0,95)}$	-	0.3	–	–

Table 12 – Summary of the improvement delivered by the bacterial seed treatment, in percentage

Indicator	Variant		Improvement delivered by the bacterial seed treatment comparing to the standard fungicide, %
	Oplot fungicide	Oplot + Bacteria	
Weight of the root system, gram	0.64	0.79	23.0
Root length, cm	14.8	16.2	9.5
Preservation of plants for harvesting, %	70.3	86.5	23.0
Yield, centners	13.4	16.3	21.6
No. of grains per plant	41	46	12.2
Weight of 1000 grains, gram	178	179	0.56

CONCLUSION

Thus, in the conditions of 2018, the use of the bacterium provided by SOSBIO RESEARCH RUS LLC for seed treatment on soy, together with Oplot fungicide, had a **significant** impact on the formation of soybean yield. Yield increase was 2.9 centners per hectare. However, the foliar application of bacteria during the growing cycle of soybean plants of the Monsoon variety showed no effect. Analysis of the effect of the bacteria tested in this study on soybean plants using the bioindication method showed that the joint application of Oplot fungicide and bacteria for seed treatment provides the greatest increase in the mass of the root system and the root length – 12.5 and 12.6% respectively.