New technologies in biological plant protection and its localization

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Abstract. *The sharp increase in the use of pesticides in agrobiocenosis in the background of no-till and minimum tillage called: the growth of costs, the decline of soil fertility, the occurrence of resistance in harmful organisms and change in species composition, a number of other pressing environmental problems. In this regard, the most preferred and safe bipolarization of plant protection. The use of microorganisms in plant protection can reduce the number of harmful organisms in anthropogenic ecosystems, to improve the structure of soil due to increase of the antagonistic bacteria and fungi in it. These types of bacteria and fungi, it is advisable to use directly for plant protection. They cause maceration of the pathogens cell walls, produce a variety of physiologically active substances that stimulate plant growth, suppress the growth of pathogens and even insect-pests. Developed a new method of biological plant protection based on the system application (tank mix) of soil microorganisms bacteria and fungi. It is based on the use of various microorganisms: Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium (RU, Patent № 2539025).*

Consistency is the three mandatory conditions making -1) Processing plant residues, 2) seed treatment, 3) treatment of crops by a complex of microorganisms.

To reduce the cost of biologics applied the concept of culturing microorganisms in a simple culture medium (sugar and bran) in terms of its small tonnage production.

Bran are a source of protein, vitamins, polysaccharides and starch. They are also a source of minerals and calcium. Bran protein is the basis of cytoplasmic content of the cells. Sugar is an additional source of nutrition for bacteria and fungi. The microorganisms break down sugars by hydrolysis to simple sugars of glucose and fructose. Nutrient medium from the bran may contain amino acids. But some of the amino acids added in small amounts to intensify the growth of microorganisms. In this case, the amino acid is a catalyst for microorganisms.

Small-scale production has its own characteristics:

1. It is located near production fields.

2. It is a periodic process.

3. It belongs to seasonal productions.

4. The need of this production is determined by the necessity of carrying out of agrotechnical measures: vegetation treatment, seed treatment and stubble.

5. Cultivation of microorganisms associated with the enzymatic decomposition of complex components, typically from the group of sugars and/or polysaccharides.

In the first case we have treated wheat seeds with different microorganisms (titer of 104 to 107).

The control variant in winter wheat was without treatment and the third variant was treated with the fungicide Raxil (Tebuconazole + Prothioconazole + Metalaxyl) Then seeds were sown. Then in the phase of the third leaf of plants grown from treated seed microorganisms, were re-treated with a suspension of microorganisms (titer of 104 to 107).In addition, after harvesting fore crop residues were treated with a suspension of the above-mentioned complex of microorganisms.

The results showed that the yield in the control variant (without treatment) of 3.87 tons/ha, the third option (with the fungicide Raxil) - 4.38 tons/ha, and the first option (microbiological treatment) - 4,5 tons/ha.

Study of biometric parameters of seedlings showed that the long roots (cm) control variant ranged from 7 to 15, in the third variant (with the fungicide Raxil Ultra) - from 8 to 13 and in the first (microbiological treatment) - from 9 to 16.

Furthermore it is established that at low and medium infection of seed such processing can suppress the development of root rot agents, so in the absence of smut is appropriate to apply microbiological protection.

The abstract is often the only part of the paper to be read, so include your major findings in a useful and concise manner. Include a problem statement, objectives, brief methods, quantitative results, and the significance of your findings. The abstract should be no more than 350 words long.

Keywords. *biological plant protection, Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium*
List both specific and general terms that will aid in searches.

Main Body Instructions (main headings use the Heading 1 style)

The sharp increase of the pesticides uses in agroecosystems on the background of the "No" and "Minimum" tillage caused: cost increase, soil fertility reduction, increasing of pests' resistance and its species composition change, as well as a number of other sharp environmental problems. In this regard, the most preferable and safe is the biological plant protection. Application of the microorganisms in plant protection can reduce the number of harmful organisms in agrobiocenosis and improve soil structure due to amount increase of antagonistic fungi and bacteria in the soil.

Such antagonistic bacteria and fungi types, are reasonable to use directly for plant protection. Its causes maceration of the pathogens cell walls, produce various physiologically active substances, which stimulate plant growth, suppress the growth of pathogenic microorganisms and insects.

To meet these challenges, manufacturers of biological crop protection products offer a wide range of ready to use microbial agents, which, despite of the high quality, has major disadvantages: organisms in these media are at dormancy stage (which often leads to the lost of effectiveness because of long period for the transition period to a vegetative stage), the limited spectrum of action (on the certain pathogen or pest) and their high cost (which leads to the cost production increase).

The new method for biological crop protection was developed based on the systemic application (as a tank mix) of soil microorganisms (bacteria and fungi). It is based on the application of different microorganism's complex: Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium [1]. Consistency is based of to three mandatory conditions - 1) treatment of plant residues, 2) seed treatment 3) crop ptotection.

To reduce biologics cost and producing the vegetative forms of microorganisms we offer to apply the concept of its production on a simple medium (sugar and bran), or even on saline mixture in a local small-tonnage production. This makes it possible to prepare preparations for the particular conditions of each farmer with improved logistics and apply the biologics in a complex (against diseases and pests, as well as to improve soil fertility).

Small-tonnage production has its own characteristics:

1. It is located near the production fields.

2. It is a periodical process.

3. It belongs to the seasonal production.

4. The demand for this production is determined by the needs of agricultural activities: vegetative treatment, seed treatment, stubble cultivation.

5. Cultivating of microorganisms related to enzymatic degradation of complex components, usually from the group of sugars and / or polysaccharides.

Materials and methods

As the object of study were taken prospective components of the nutrient medium for the cultivation of microorganisms in the conditions of small-tonnage production – bran, crushed grain, and sugar. Further more the following strains of microorganisms were used: Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium (from the collection of the Kuban State Agrarian University).

In a first option, our experimental wheat seeds were treated with various microorganisms (titer from 104 to 107). Control variant was untreated (winter wheat), while the third option has been

treated with fungicide (Raxil (Tebuconazole + Prothioconazole + Metalaxyl)). Then the seeds were sown in the soil. Then in the plant grows phase of 3-leaf the grown plants from treated seeds were treated again with the microorganism suspension (titer 104 to 107). Along with this, after harvesting of a previous crop residues were treated with a suspension of the mention above microorganisms' complex.

Research results.

In agricultural biotechnology with a high volumes of production of biological products, one of the main objectives is to ensure the right composition of the culture media by replacing expensive components: protein - peptone, vitamins, amino acids, yeast extract, salt on the various waste products containing right components for micro-organisms. That is why, as the basis of the nutrient medium for culturing microorganisms it was chosen different types of bran and sucrose (sugar). Bran was selected due to its nutritional value, content of all essential vitamins and minerals [2, 3].

Usage of bran can replace classical medium (modified or similar broths Saburo, Capek, timing-broth), used in classical microbiology or biotechnology, by a simple and less expensive media. According to the chemical composition, corn bran contained more dietary fiber than wheat and wheat bran containing nearly 2 times more protein and 4.6 times more fat. Furthermore, it should be noted that in the raw grain there is molybdenum that is important for bacterial enzymatic complex A. chroococcum - [4], as well as in soybean grain - about 99 mcg on 100 g [5]. Wheat bran content more potassium (1182 mg on 100 g) compared to its content in maize bran (44mg on 100g), which is required for the activity of enzymes involved in protein synthesis.

Sugar use as a component of nutrient medium especially inexpensive, available and properties - by hydrolysis (acidic or enzymatic), it easily forms monosaccharides (glucose and fructose).

To chose the raw materials used for preparation of the nutrient medium (bran), which provides activity of microorganisms, the growth rate was determined and gave a quantitative assessment (CFU / ml). Pure cultures of this microorganism were grown in vitro on nutritive media from corn or wheat bran (with varying its mass ratio to the amount of sugar). Microorganisms were cultured in these nutrient media for 7 days at 23 ± 2 ° C. Then dilutions of the culture fluids were made seeding by depth way on nutrient media №1, timing-agar (for bacteria) and Saburo, Capek (fungi). Further culturing was carried out on agar medium for 7 days at 30 ± 2 ° C for bacteria and 14 days at 23 ± 2 ° C for the fungi with a daily counting of the CFU number.

Results of these experiments showed that the growth of microbial cultures is possible on nutrient media, based on wheat, and corn bran. But practically in all the samples grown on corn bran, and passed for further study on the medium № 1, timing-agar for bacteria and Saburo, Czapek for fungi the beginning of development and growth of microorganisms was for one day later than in samples grown on wheat bran. A fungus such as M. anisopliae and B. bassiana gave a larger number of colonies on the culture medium from wheat bran. In addition to the culture medium from wheat bran growth of these fungi began after two or three days, while on the medium from corn bran only the third-fourth day. This can be explained by the fact that the coarse fiber content of corn bran polysaccharide fraction as a dietary fiber reaches 79%, which is 36.2% higher than in wheat bran (Table 1).

Table 1 - Comparative evaluation of the number of microorganisms (cfu / ml) made on the culture media of wheat and corn bran

The ratio of the sugar and bran g / 100 ml of medium	Cultures of microorganisms				
	Azotobacter chroococcum	*Bacillus megatherium*	*Trichoderma viride*	*Metarhizium anisopliae*	*Beauveria bassiana*
Corn bran					
0,5:0,5	$4x10^5$	$3x10^5$	$4x10^4$	$7x10^3$	$6x10^3$
0,75:1,0	$4,5x10^5$	$4x10^5$	$4,5x10^4$	$7x10^3$	$7x10^3$
1,0:1,5	$1x10^6$	$9x10^5$	$1x10^5$	$2x10^4$	$7x10^3$
1,25:2,0	$1,2x10^6$	$1x10^6$	$1,2x10^5$	$3x10^4$	$1x10^4$
1,5:2,5	$3x10^6$	$3x10^6$	$3x10^5$	$4x10^4$	$1x10^4$
2,0:3,0	$3,1x10^6$	$5x10^6$	$6x10^5$	$4x10^4$	$9x10^4$
Wheat bran					
0,5:0,5	$3x10^6$	$5x10^6$	$5x10^4$	$2x10^5$	$9x10^4$
0,75:1,0	$5x10^6$	$9x10^6$	$2x10^5$	$2x10^5$	$9x10^4$
1,0:1,5	$1x10^7$	$3x10^7$	$7x10^6$	$2,6x10^5$	$7x10^5$
1,25:2,0	$2x10^7$	$1x10^8$	$8x10^6$	$2,6x10^5$	$7,2x10^5$
1,5:2,5	$3 x10^7$	$2x10^8$	$8,5x10^6$	$2,6x10^5$	$8x10^5$
2,0:3,0	$3,5x10^7$	$2,5x10^8$	$9x10^6$	$2,6x10^5$	$8,5x10^5$

A similar result was obtained in samples with *A. chroococcum* in a smaller number of colony forming units and growth delay for a day compare to corn bran. Beginning of the growth of these bacteria in a corn bran medium - on the third day. This explain the absence of cellulolytic enzyme activity of these microorganisms, i.e. they are only able to use low molecular weight polysaccharides. Thus *T. viride*, *B. megatherium* (having cellulolytic activity) rising substantially at the same rate in media of wheat bran and corn bran. Moreover, these environments achieved approximately the same value of CFU / ml. The absence of cellulolytic activity in *A. chroococcum, M. anisopliae* and *B. bassiana* confirmed by test to determine the fermentation of carbohydrates using peptone water with indicator Andrade, in absence of color change of the indicator to red, where wheat bran was added. This indicates that the decomposition of bran does not occur. While the samples with introduction of *T. viride, B. megatherium* changed its color.

That is indicating on the decomposition of high-molecular polysaccharides, change of peptone water pH, the accumulation of acids by enzymatic hydrolysis of bran.

Thus, we selected components as the main raw material for the preparation of the culture medium from wheat bran and sugar that provide the vital activity of cultured microorganisms.

Due to the fact that the bran is the main component of the nutrient medium, and entering of yeast microflora inevitably, it became necessary to change the classical technological scheme, where cultures of microorganisms immediately introduced in to the nutrient medium of the bran and sugar. To solve this problem, it has been hypothesized - to divide the introduction of bran and primary cultures of microorganisms over time so that till the time when extraneous microflora will begin to develop in the environment, the main culture already will move from a lag phase of growth to the exponential phase, i.e. divide actions in a time.

In laboratory conditions all the components of the nutrient medium and culture microorganisms were added simultaneously by the first process scheme using bran. To do this, 1 ml of suspension cultures prepared from microorganisms with microbial load $1x10^7$ CFU / ml and yeast cells with a microbial load $1x10^3$ CFU / ml, were introduced in to a sterile liquid nutrient medium, prepared from sugar and wheat bran. Sterilization of the nutrient bran medium were done at 121 ° C for 15 minutes in the laboratory conditions was conducted in order to prevent introduction and influence of other microflora introduced with bran and artificially create it by inoculating yeast microflora.

According to the second technological scheme the sugar is dissolved in water and then made inoculation medium with a suspension of microorganisms with the load $1x10^7$ CFU / ml, and sterilized wheat bran and the suspension of yeast cells from isolated culture with microbial load $1x10^3$ CFU / ml, were introduced into the nutrient media every 48 hours (fungi) and 24 hours (for bacteria).

The choice of the time intervals was determined by us, based on our previous research, which showed that it was after 48 hours of culturing fungi and after 24 hours of culturing bacteria growth of colony-forming units of the main crops were observed. The number of primary culture and yeast inoculum was 1 ml per 1000 ml of medium.

These microbial load were obtained by the preparation of the initial suspensions of cultures with subsequent dilution in saline for turbidity standard №5 and №10 EM, conditionally accepting that 10 EM corresponds to $1x10^9$ CFU/ml. Load data were selected for inoculating microorganisms with the liquid medium, due to the fact that the titer of microorganisms used for the culture liquid preparations, should be at least $1x10^7$ CFU/ml.

The number of yeast microflora introduced with bran and isolated from the culture medium, during multiple studies reached $1x10^3$ CFU / ml. The total time in culture medium supplemented with the bran was 7 days at 23 ± 2 ° C. Sowing of the liquid medium bran on Czapek agar medium were made after 2 days of growth (before introducing in to the medium of the wheat bran and yeast culture inoculation), as well as on the 7 day of culturing.

Sowing from culture liquids dilutions were made by profound way in a culture medium for bacteria №1, Saburo and Čapek for fungi. Tempering cultivation of microorganisms on agar media was carried out at a temperature of 30 ± 2 ° C for bacteria and 23 ± 2 ° C for fungi. The research results showed that is in general in all variants of the experiment the quality improve of biologics was achieved, expressed in a substantial increase of the titer (Table 2).

Table 2 - Indicators of the quality of microbiological preparations for various biotechnological process scheme

Trial variant	At first the technological scheme			At the second technology scheme			
	Number of CFU / g (ml) 168 hours of growth	pH	The residual amount of reducing sugars, %	Number of CFU / g (ml) 1 day for bacteria; 2 -day fungi (After the lag phase)	Number of CFU / g (ml) 168 hours of growth	pH	The residual amount of reducing sugars, %
A.chroococcum							
1	$9,9x10^6$	5,80	13,00	$7,8x10^5$	$5,7x10^8$	5,90	12,70
2	$1,1x10^7$	5,80	12,80	$8,2\ x10^5$	$6,3x10^8$	5,95	12,50
average	$1,0x10^7$			$8x10^5$	$6x10^8$		
B. megatherium							
1	$6,3x10^7$	6,00	9,82	$8,9x10^5$	$1,5x10^8$	6,30	9,13
2	$5,7x10^7$	6,24	10,0	$9,1x10^5$	$9,5x10^7$	6,27	9,20
average	$6,0x10^7$			$9,0x10^5$	$1,0x10^8$		
T viride							
1	$1x10^5$	4,2	5,78	$1,3x10^5$	$7,6x10^6$	4,93	8,50
2	$1x10^5$	4,3	6,00	$9,7\ x10^4$	$7,3x10^6$	4,90	8,49
average	$1x10^5$			$1x10^5$	$7,5x10^6$		
M. anisopliae							
1	$2,6x10^5$	4,8	5,00	$3,7x10^5$	$1,4x10^7$	5,43	9,33
2	$2,4x10^5$	4,8	5,20	$3,3x10^5$	$1,0x10^7$	5,30	10,00
average	$2,5x10^5$			$3,5x10^5$	$1,2x10^7$		
B. bassiana							
1	$2,0\ x10^5$	4,9	5,33	$3,7x10^5$	$9,0x10^6$	5,20	8,30
2	$2,0\ x10^5$	4,7	6,00	$3,9x10^5$	$9,0x10^6$	5,32	8,00
average	$2,0\ x10^5$			$3,8x10^5$	$9,0x10^6$		
yeast culture							
average, CFU / ml for whole trial	$1x10^4$	-	-	-	$2x10^2$	-	-

Yeasts are undesirable microflora in the process, developing throughout the culturing process of the main microflora, which is 7 days from the first variant, and 5 days for the second processing scheme for growing fungus cultures, when they were introduced into the culture medium with bran on the second day. And 6 days for growing bacterial cultures, where the bran are introduced after 1 day of growth of the main crops.

Time reduction of development and culturing external microflora represented by yeast for 1-2 days, make it possible to do the process more profitable from an energy point of view. In this case during this period the sucrose in the culture medium, is consumed by the grown microflora rather than yeasts. In general, as the results shown (table 2), the use of two-phase processing circuit provides a culture liquid, with higher titer (especially fungal drugs,) as compared to a single-phase scheme in which inoculation of yeast microflora and grown cultures of microorganisms occurs simultaneously with bran. Titer of fungal microflora was increased: *T. viride, B. bassiana* and *M. anisopliae* from 10^5 to 10^6 - 10^7, and for bacteria *A. chroococcum, B. megatherium* from 10^7 to 10^8.

Preparations based on bacterial cultures were substantially free of yeast microflora even in a single-phase variant. This is because the development of *A. chroococcum, B. megatherium* introduce in to the culture liquid fungicidal substance that may inhibit the development of yeast. Biotechnological process for the first processing variant, especially in the samples with *T. viride, B. bassiana* and *M. anisopliae* has provided substantially lower pH (4,2-4,9) compared with the process flow diagram for a second variant (4.9 to 5 , 43).

In this case residual content of reducing sugars obtained by titration glucose was also lower - 5,0-5,78 for single-phase flow scheme against 8,30- 9,33 for a two-phase flow scheme. This indicates that the symbiosis in the medium of cultured fungi and external microflora - yeast leads to the fact that sucrose is consumed (decomposed to glucose and fructose) not only by the main microflora, but also consumes by yeast (and in low aeration – even fermented). The pH of samples with bacteria and the residual amount of reducing sugars by two technological schemes were almost equal. These changes in flow diagram form the introduction of bran in 24 hours in to the medium for bacteria and 48 hours in to the medium for fungi has solved the basic problem of biotechnological process - increasing purity of the preparation (culture fluid) and titer increase of the final products.

Thus, the technological scheme is represented in the Figure 1.

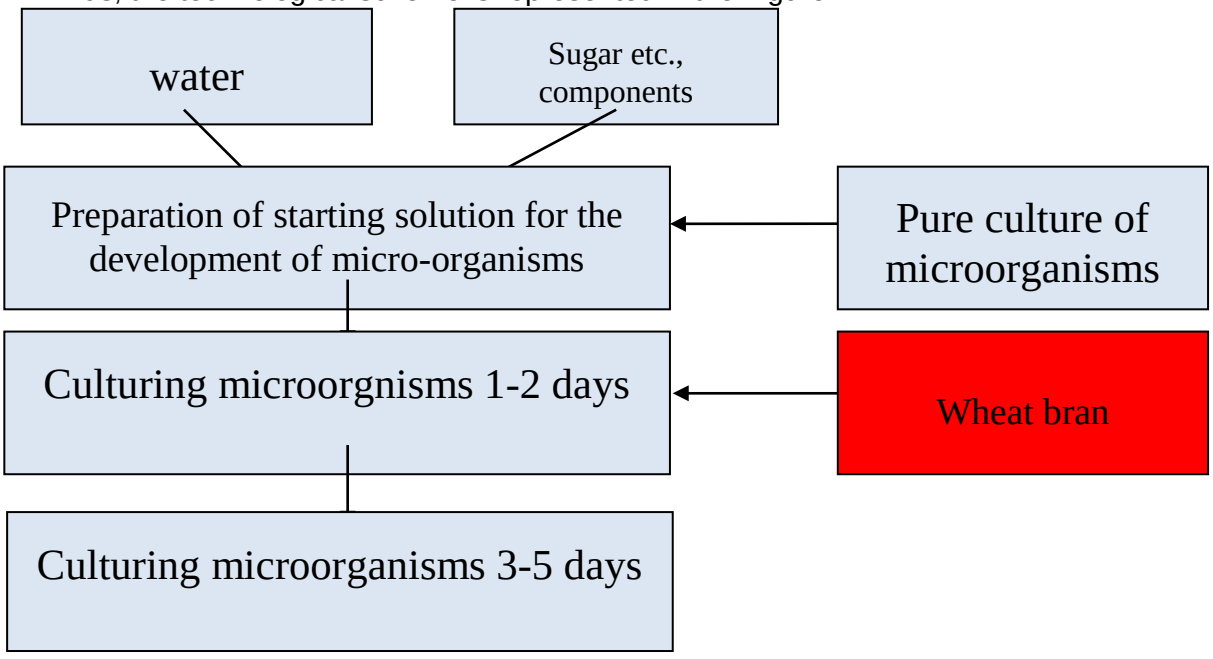

Figure 1 - Two-phase flow scheme of the production of microbiological preparations

Using of this biotechnology system in the conditions of farms (as well as transmitting of dormant cells into vegetative forms of microorganisms on physiological saline liquid) was effective (Table 3).

Table 3 - Cost-effectiveness of the application of various crop protection drugs (average 2013-2014)

Index	Trial variants			
	Without protection	The proposed technology (local production)	Ready biological product	Pesticide technology (seed treatment, fungicide, insecticide)
Yield t / ha	3,825	4,225	4,300	4,050
Cost of 1 liter of the preparation, rub	-	24,58	200,00	1250,00*1 7400,00*2 529,00*3
Production cost of units	6270,00	6697,69	20300,00	30207,00
Profit, rub/ha	46706,25	51818,56	39255,00	25885,20
Profitability, %	537,08	**557,82**	168,47	61,78

*1 – seed treatment *2 – fungicide *3- insecticide

For these purposes, equipped simple fermenters with aerators. Dealing with such production of microbiological preparations produced directly by farmers for their specific needs and for a certain period, and the control process is carried out on-line by specialists of the head company, with periodic sampling for microbiological analysis.

These drugs are marked with colored stickers for usability by staff, packed and delivered directly in to the field, where the tank-mixture is prepared immediately before application. Work for its application in the field are carried out only at night. Seed treatment is carried out just before sowing for better storage and viability of not spore microorganisms.

Results of field experiments has shown that biometrics index were maximal in the variant with the microbiological seed treatment. In the variant, the root length varied - from 9 to 16 cm, in the control variant, from 7 to 15 cm and in the variant with the fungicide " Raxil (Tebuconazole + Prothioconazole + Metalaxyl) " - from 8 to 13 cm (Table 3). Also it was founded that at low and medium infection level such seed treatment can suppress the development of root rot pathogens, so in the absence of smut infection it is advisable to apply the microbiological protection. This resulted in a higher yield of grain in the variant with the microbiological treatment - 4.5 t / ha, while in the control variant 3.87 tons / ha was reached and with fungicide "Raxil (Tebuconazole + Prothioconazole + Metalaxyl) " - 4.38 t / ha.

A further strategy is based on the use of the mention above complex of microbial agents that can act effectively (Table 5).

Table 5 – Influence of the application of fluids of microorganisms cultures on biometrics of wheat plants in the tillering phase and on the root rot defeat (vegetation experiments)

Variant	Root rot defeat, %	Root length, cm	Number of leaves	The volume of the root system, cm3
Control	50	34,4	2,1	6,5
T. viride	10*	32,2	2,5	6,4
B. megatherium	8*	39,0*	3*	8,0*
A. chroococcum	15*	34,8	3*	6,5

* Deviations from the control are significant at p = 0.95

However, for the 2-3rd treatment of crops during the growing period (at a temperature above + 18 ° C) in a tank mix is more appropriate to use a range of microorganisms: *Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium* and besides in this complex *A. chroococcum* appropriate to apply with *B. megatherium*, since being a nitrogen-fixing microorganism, it needs immediate power circuit phosphorus compounds which produce *B. megatherium*. So you can further realize the symbiotic relationship of these bacteria. Using cultures of *A. chroococcum, B. megatherium* and *(B. subtilis)* in the tank mixture compensates decreasing of plant biometrics associated with the use of *T. viride*, which is the destructor of polymers and provides the remaining microorganisms with available carbohydrates (sugars). The feasibility of such a tank mixture was confirmed in a production test on a total area of 90 hectares (where there were done experience with the area on the third part of the field):

1) control - without treatment (wheat seeds and vegetating plants) ;

2) biologics - wheat seeds and crops during the growing season treated by microbiological preparations;

3) pesticides - wheat seeds dressed with the fungicide "Raksil Ultra", and during the growing season crops treated with fungicide "Lamador" (protikonazol+tebuconazole) and insecticide "Dimethoate-400."

The experiment confirmed positive effect of the developed microbiological drugs in a tank mixture in comparison with chemical fungicides and insecticides (Table 6). This was manifested in the increase of grain yield due to the increase in tillering, number of grains in an ear and weight of 1000 seeds with a low-grade lesions of Fusarium root rot (15-18%). Under the same seeding rate with 4.5 million germinating grains per 1 ha, the most important elements are density of productive stems, number of spikelets per spike higher in the variant where plants were treated with microbiological preparations. Thus the number of stems per 1 m^2 somewhat higher in samples with the use of microbiological treatments compare to the samples with chemical plant protection, and in comparison with the control variant. So in the samples with the chemical plant protection, the number of plants with two or more stems lower than in the samples where microbiological protection scheme was used. This is realized in yield increase in the variant with biologics compared to chemical plant protection at 0.1-0.2 t / ha.

Table 6 - Effect of seeds and plants treatment and biological products on the damages of plants by root rot, grain yield and the elements of its structure (production test OOO SHP "Temizhbekskaya")

Variant of seed and plant treatment	Damages by root rot, %	The number of stems per 1 m2	Ear length, cm	Number of grains in a ear	Weight of 1000 grains, g	Grain yield, t / ha
2013						
Control	51	180	6,7	24,4	36,2	3,8
Biologics	23*	312*	7*	30,6*	47,5*	4,15*
Pesticides	15*	307*	7*	29*	47,2*	4,08*
2014						
Control	57	190	6,3	23	38,2	3,85
Biologics	26*	330*	7*	30,8*	47,9*	4,3*
Pesticides	18*	305*	7*	29,3*	46,8*	4,05*

* Deviations from the control are significant at p = 0.95

Production tests were done in the big agro producer – Group of companies "Steppe" (Russia, Kranodar region) and the results showed that three-year of use of biologics has increased the number of suppressive soil microflora in relation to pathogenic (Table 7).

Table 7 - Influence of seed treatment by biological products on biometric indicators of germination and yield of winter wheat

Variant	Root length, cm	Damages by root rot,%	Grain yield, kg / ha
Control	7-15	18,20	3,87
Fungicide	8-13	8,75	4,38
Biological drugs	9-16	9,00	4,50

This provided a reduction of the volume of fungicides application by 70% and 85% of insecticides. In addition, these activities have increased soil fertility levels. For example, in the enterprise "Rodina" (Russia, Volgograd region), the humus content in the soil in 2015 increased by 0.2% compared with 2012, and in the enterprise "Trud" - by 0.05%, the content of available phosphorus increased almost 1.5 times and potassium for about 1.2 times (table 8,9).

Table 8 - Value of pathogenic microorganisms to suppressive in the biocenosis on the lend of Groups of companies "Steppe"

Company	2012	2013	2014

«Rodina»	46/52	12/89	27/75
«Trud»	51/49	5/90	10/75

Table 9 - Influence of microbiological preparations application on soil fertility improvement in biocenosis on the lend of Groups of companies "Steppe"

Company	Humus content,%		The content of available phosphorus, units		The content of available potassium, units	
	2012	2015	2012	2015	2012	2015
«Rodina»*	3,8 (3,7-4,9)	4 (3,9-4,1)	32,8	47,8	477	596
«Trud»**	3,8 (3,7-3,9)	3,85 (3,8-3,9)	32,9	45,9	474	568

* Average of 10 fields, ** average of 9 fields

Discussion of research results.

Small-tonnage production of microbiological preparations (directly at the farm), with a pretty high quality is more then reality. For this we proposed usage of inexpensive components of the nutrient medium - sugar and wheat bran, and the formation of a two-phase scheme of preparing of biotech culture medium (the first stage - settling culture with the microorganism and the second - the introduction of partially sterile wheat bran) or saline.

Such kind of production is improving logistics and use of vegetative forms of microorganisms and the low price of the final product. Systemic application of microbiological preparations prepared by this production way (for the treatment of seeds, crops and crop residues) with the microbial complex developed (*Trichoderma viride, Beauveria bassiana, Metarhizium anisopliae, Azotobacter chroococcum, Bacillus megatherium*) were effective. This ensures not only the plant protection against diseases and pests, but improves soil suppressant and increases its fertility, increases the productivity of biocenosis. In general, this concept is being successfully implemented in Russia (in the regions: Krasnodar, Stavropol, Volgograd, Voronezh, Ural , Rostov regions, and in the Crimea).

Reference

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