BIOTECNOLOGY

ANALYSIS OF INFLUENCE OF BIOHUMUS ON THE BASIS OF CONSORTIUM OF EFFECTIVE MICROORGANISMS ON THE PRODUCTIVITY OF WINTER WHEAT

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Abstract: The article considers the problem of recycling of poultry waste, in particular, feather-downy waste and chicken dung. Traditional ways of poultry waste disposal have been shown and their shortcomings have been described. The main advantages of use of bioprocessing methods to implement the process of waste conversion with the formation of effective products – biohumus – have been determined. Biofertilizers or biohumus is a modern innovative means applied not only for the purpose of an increase in productivity, but also for land recultivation and resuscitation. The authors have studied the biohumus obtained by means of processing of mixture of feather-downy waste and dung in the ratio of 8:2 with the application of the biological product containing the consortium of decomposer microorganisms: Bacillus pumilus AL16, Microbacterium terregens AC1180, Aeromonas sp. B5376, Arthrobacter globiformis AC1529, Streptomyces olivocinerus AC1169 and Acinetobacter sp. B390. An assessment of efficiency of application of various rates of the developed biohumus for winter wheat crops has been made. It is shown that the application of a comparison sample - the organic mineral fertilizer "Universal'noe" (the expense is 150 g/m\textsuperscript{2}) and the use of the developed biohumus (the expense is 150 g/m\textsuperscript{2}) provided an increase in its growth, compared with the control sample of the wheat "Skipetra", by 12 and 15 cm (the stem elongation stage), by 15 and 18 cm (the earing stage), by 21 and 30 cm (the milk stage) and by 19 and 30 cm (the firm ripe stage) respectively. The above values for the grade Zimushka were within the similar limits: an increase in its growth, compared with the control sample, by 17 and 18 cm (the stem elongation stage), by 19 and 21 cm (the earing stage) and by 19 and 31 cm (the milk stage and the firm ripe stage) respectively. An increase in the productivity of grades of the winter wheat "Skipetra" and "Zimushka" by 1.5 times (1.25 ± 0.25 t/haetare) has been established during the application of the obtained biohumus with the rate of 150 g/m\textsuperscript{2}. The results of studies of change of fractional composition of grain proteins testify that the application of biohumus provides an increase in gluten fractions of winter wheat grains. An assessment of economic efficiency has been performed, it has been established that the highest rates of profitability refer to the variant with the application of the developed biohumus (the expense is 150 g/m\textsuperscript{2}) – 63.2\%, the lowest rates are 39.2\% in the control sample.

Keywords: Biohumus, poultry waste, effective microorganisms, waste bioconversion, microbial utilization, biological product, productivity of wheat, biofertilizer

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INTRODUCTION

Recycling of waste and by-products in the branches of food and processing industry provides a number of environmental problems in the sphere of ensuring the sustainable development and protection of the environment [1–3].

Feather-downy waste in the form of by-product is formed in high quantities during commercial poultry processing and, in the chemical relation, is native keratin [2]. The high mechanical stability, hardness and a large number of disulfide bonds of native keratin makes it resistant to the degradation caused by proteolytic enzymes, such as pepsin, trypsin and papain [3, 4, 6]. Due to the high content of nitrogen the accumulation of the formed waste by their storage, burial or burning provides not only the destruction of surface soils, but also considerable losses of biological resources, such as proteins and enzymes [5].

The traditional way of dung removal is burning. However, it is not expedient either from the ecological, or the economic point of view [7]. One of problems of dung utilization by means of burning is a high exit of...
CO₂. Taking into account the pollution caused by the use and change of properties of the used lands, about 9% of evolutions of CO₂ [8] fall to the share of poultry-farming sector in the structure of human activity.

The foremost scientists in the field of ecology have proved that poultry waste burning is pernicious for the planet and mankind. It is stated in a report of the UN that poultry farming is the main source of pollution of water and soil – it provides the formation of more greenhouse gases, than that produced by cars, the intensive poultry farming provides the degradation of soils, the pollution of reservoirs and air [9–11].

The simplest of the existing technologies of utilization of mixtures of feather-downy raw materials and dung is composting. Vast free areas remote from settlements are needed to organize it in the open air. The process lasts from 3 to 6 months and demands periodic turning of heaps for the homogenization of mixture and the activation of activity of microbes [11–15].

The classical technology has been recently supplemented with a lot of modifications. For example, composting with the use of steam-gas plants or enzymatic composting where the speed of process increases several times [16]. In recent years the ways of dung utilization with the use of coprophages (larvae of Musca domestica S., specially grown earthworms Eisenia) have become popular. For example, a unique way of dung processing with the use of house fly larvae has been worked out in the All-Russian Research Institute of Livestock [17, 18]. During the growth of fly larvae the dung mass turns into a slightly damp friable mass with a slight smell [19, 20]. The final product of processing with the use of this way is the fertilizer which is capable to increase considerably the quality and productivity of soil [21–23]. Vermiculture is quite popular in the Western European countries where people have got used to using the line of earthworms Eisenia fetida, or "Red Californian hybrid" [24]. In Russia people use the following hybrids of Red Californian worm: Obinsk, Obolensk, Vladimir and Morevka hybrids [25]. However the method of dung utilization with the use of fly larvae and lines of hybrid earthworms has essential shortcomings: the complexity of maintenance of necessary parameters of temperature and humidity of egg laying and the cultivation of larvae and lines of worms [21–23]; the difficult design of recycling units (the existence of flap doors, a lot of feeders, the complex system of motion of trays along the racks) [26]; the complexity of production organization in the territory of Russia, considering rather low average winter temperatures [27, 28]; the considerable losses of organic substance in the final products: the increased speed of their mineralization and, as a result, the high losses of nitrogen and the formation of a large amount of ammonia [29]; the discrepancy of the final products to the requirements for the physical and chemical composition presented in the Technical specifications of the Russian Federation No. 94991, some international standards and agrochemical indicators [17].

Thus, the technologies of worm composting and the use of fly larvae, even considering all their advantages, are favorable for small farms or for use for one's own needs [25, 28, 29]. The searches of alternative technologies of efficient recycling of waste of the poultry-farming complex have provided the development of biotechnologies, i.e. the use of systems (consortia) of effective microorganisms (EM) capable to process in the course of their activity the organic and inorganic substances, which are part of waste, into efficient final products – fertilizers [8, 31].

In other words, EM provide quick composting – the biological oxidation of organic substrata which provide the enzymatic hydrolysis of keratin – the structural protein of a feather [32].

The activity of effective microorganisms determining the duration and depth of process of biotransformation [32, 33] is the basis of the vast majority of bioconversion technologies. The biological products, biostimulators or the obtained products – biofertilizers (biocomposts) applied in the similar technologies are communities of selected microorganisms, or contain a specific strain of microorganisms of a certain type [17, 33].

The application of biological products for organic waste processing allows to correspond to the latest requirements of production of organic fertilizers of livestock waste [31, 33]. Namely, the preservation of its biological potential, the exclusion of a possibility of presence of pathogenic microorganisms, the elimination of an unpleasant smell and the decrease in the risk of incidence of workers at the enterprises of agrarian and industrial complex [32].

Obtaining biofertilizers on the basis of a chicken feather has drawn attention of research scientists [34] since long ago. Feather meal is cheap and easily available source of nitrogen (15% of N) and can be used as an effective biofertilizer with high biogenous properties.

Biotechnologies are the development and expansion of set of processing methods the roots of which appeared [1] thousands years ago. The application of biotechnologies in agriculture, for example, the application of growth enhancing hydrolyzates of proteins is the perspective trend of development of branch and can effectively be applied in the production of fertilizers [9, 29].

The microbial degradation of feather-downy waste is an alternative to the existing technologies of production of nitrogen fertilizers. There are bacteria capable to use feather-downy waste as the main organic substratum and a source of carbon, nitrogen, sulfur and energy [32, 35]. Indolyl-3-acetic acid is the main growth regulator which increases the availability of nutrients for plants, and also provides the growth of roots [33–36].

The degraded product obtained on the basis of feather waste can become an abundant source of the corresponding amount of tryptophane which is the basis of synthesis of indoleacetic acid [1, 36]. The poultry waste biohumus is the product with a low prime cost and a high efficiency that has a number of advantages in comparison with the existing analogs, in
particular, the pathogenic microflora which is a special problem during efficient waste recycling is completely absent in the biohumus [21, 34, 37, 39].

At the present time, one of the urgent issues is meeting the need of the population for food, and first of all it includes grain and bakery products. Based on this fact, as for efficiency, winter wheat is considered to be potential grain crops. Due to a long vegetative period it is capable to use most fully the solar energy, the nutrients contained in soil and moisture during the autumn and spring period [36–38].

The application of high rates of mineral, in particular, nitrogen fertilizersto receive heavy crops of winter wheat grains when sowing them after the late removed predecessors, which is not always justified from the economic and ecological point of view [2].

Thus, the application of new biological fertilizers is an alternative to the new chemical fertilizers that need to be thoroughly checked [40–44].

**OBJECTS AND METHODS OF STUDY**

At the previous stage of studies a biological product on the basis of consortium of decomposer microorganisms was developed: **Bacillus pumilus AL16, Microbacterium terregens AC1180, Aeromonas sp. B5376, Arthrobacter globiformis AC1529, Streptomyces olivocineres AC1169 and Acinetobacter sp. B390**. The assessment of the compliance obtained with the application of a biological product, biohumus, with the requirements of regulatory documents for fertilizers of livestock and poultry waste, in particular, nitrogen fertilizers is capable to use most fully the solar energy, the nutrients contained in soil and moisture during the autumn and spring period [36–38].

The studies were carried out on chernozem soils of the land plots of the Leninsk-Kuznetsky district of the Kemerovo region (Russia). According to the mechanical and agrochemical analyses, this type of chernozem is referred to the soils, heavy in mechanical composition, because of the high content of physical clay (the fraction of 0.01 mm) – up to 72% and bad agrophysical properties – low air and water permeability [39]. The predecessor of crops is the corn prepared as silage.

The released frost-resistant grades of the winter wheat "Skipetr" and "Zimushka" which steadily provide with heavy crops are chosen as the test crops to assess the efficiency of the developed biohumus.

All the grades of winter wheat impose high requirements for soil properties: high fertility, structure, with the enough content of nutrients, such as nitrogen, phosphorus and potassium. The acidity of soil solution must not exceed the indicator of 7.5, therefore, the medium must be alkalescent or acidic.

Chernozems are considered to be the best soils for winter wheat. They have enormous soil and humus horizons and contain a significant amount of humus (from 8 to 12%).

**The agrotechnology standard for the area of studies.** The tests were carried out on chernozem soils of the land plots of the Leninsk-Kuznetsky district of the Kemerovo region (Russia). According to the mechanical and agrochemical analyses, this type of chernozem is referred to the soils, heavy in mechanical composition, because of the high content of physical clay (the fraction of 0.01 mm) – up to 72% and bad agrophysical properties – low air and water permeability [39]. The predecessor of crops is the corn prepared as silage.

The studied grades of winter wheat were sown in narrow rows with the sowing rate of 6.0 million of viable seeds per hectare (~ 260 kg/hectare). The experiment was performed on the plots of 50 m². The experiment was made with a triple frequency.

Design of the experiment. Fig. 1 provides the design of experiment of the study of influence of various rates of biohumus on the grades of winter wheat.

Proceeding from the laboratory results the design of experiment consists of the following stages: the biohumus obtained at the first stage of studies is applied for the plots with a crop of winter wheat with the following rates: 1 – the developed biohumus (the expense is 100 g/m²); 2 – the developed biohumus (the expense is 150 g/m²); 3 – the developed biohumus (the expense is 200 g/m²). The following was used as a comparison: 4 – the organic mineral fertilizer "Universal'noe" (the expense according to the instruction is 150 g/m²); 5 – the control (without the application of biohumus).

**Determination of protein and protein fractions in grains.** The protein in the grain obtained according to the design of the experiment was determined in accordance with GOST 10846-91 "Grain and products of its processing. Method for determination of protein" with the use of Kjeldahl flask. The protein fractions of grain were determined using the method of gel penetrating planar chromatography (GPPC).

| Biopreparation | Waste (feather-downy : dung 8 : 2) | Biohumus | Fertilizer "Universal'noe" 150 g/m² |
|----------------|----------------------------------|----------|----------------------------------|
| Wheat "Skipetr" |                                  |          |                                  |
| Wheat "Zimushka" |                                 |          |                                  |
| Application rates (g/m²) | 0 100 150 200 | 0 100 150 200 |

**Fig. 1.** Design of the experiment.
RESULTS AND DISCUSSION

Table 1 provides the results of a study of determination of the optimum norm of application of the developed biohumus for winter wheat of the grades “Skipetr” and “Zimushka”.

According to the data presented in Table 1, the application of the organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) and the use of the developed biohumus (the expense is 150 g/m²) provided an increase in its growth, compared with the control sample of the wheat “Skipetr”, by 12 and 15 cm (the stem elongation stage), by 15 and 18 cm (the earing stage), by 21 and 30 cm (the milk stage) and by 19 and 30 cm (the firm ripe stage) respectively.

The use of these biohumus variants has provided the best indicators, both for the grade “Skipetr”, and for the wheat of the grade “Zimushka”. The above values for the grade “Zimushka” were within the similar limits: an increase in its growth, compared with the control sample, by 17 and 18 cm (the stem elongation stage), by 19 and 21 cm (the earing stage) and by 19 and 31 cm (the milk stage and the firm ripe stage) respectively.

The results provided in the experiment in the study and analysis of dynamics of accumulation of dry weight when applying the developed biohumus and the organic mineral fertilizer “Universal’noe” were unequal. Thus, at the stem elongation stage the largest amount of dry weight is noted in the samples with the application of the organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) and with the use of the developed biohumus (the expense is 150 g/m²). The maximum gain at the earing stage was noted in the sample with the application of the developed biohumus (the expense is 150 g/m²), and was 340 g per 100 plants (the grade “Zimushka”).

The dry biomass in the control sample (without the use of biohumus) and in the variant with the application of the developed biohumus (the expense is 200 g/m²) is nearly 1.5–2.0 times lower than that in the variant with the application of the developed biohumus (the expense is 150 g/m²). This regularity in the dynamics of dry biomass is characteristic of the milk stage and the firm ripe stage.

The maximum gain in the noted phases is also in the variants with the application of the developed biohumus (the expense is 150 g/m²) and the organic mineral fertilizer “Universal’noe”.

It is known that the volumes of future harvest are characterized by two basic indicators – the density of productive plant stand and the mass of grain of one ear. These components are the generalizing characteristics of a harvest of winter wheat grains of any grade. Table 2 presents the results of calculation of the above-mentioned indicators during the application of biohumus.

Table 1. Dynamics of the growth (cm) and the accumulation of dry biomass (g/100 sol.) of winter wheat of the grades “Skipetr” and “Zimushka” in the plants depending on the rates of the applied biohumus or fertilizers (the average for 2014–2015)

| Variant of experiment | Growth dynamics (cm) / dry biomass accumulation (g/100 sol.) | Stem elongation stage | Earing | Milk stage | Complete ripeness |
|-----------------------|-------------------------------------------------------------|----------------------|--------|------------|-------------------|
|                       |                                                             | 75/143               | 78/181 | 95/245     | 94/245            |
| “Skipetr”             | Developed biohumus (the expense is 100 kg/m²)                |                      |        |            |                   |
|                       | Developed biohumus (the expense is 150 g/m²)                 | 88/155               | 91/225 | 110/336    | 110/328           |
|                       | Developed biohumus (the expense is 200 g/m²)                 | 87/133               | 89/167 | 99/267     | 99/273            |
|                       | Organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) | 85/133               | 88/170 | 101/289    | 99/289            |
|                       | Control (without the application of biohumus)               | 73/80                | 73/133 | 80/135     | 80/135            |
|                       | “Zimushka”                                                  |                      |        |            |                   |
|                       | Developed biohumus (the expense is 100 kg/m²)                | 79/143               | 82/183 | 99/257     | 99/257            |
|                       | Developed biohumus (the expense is 150 g/m²)                 | 92/157               | 96/228 | 115/340    | 115/332           |
|                       | Developed biohumus (the expense is 200 g/m²)                 | 90/136               | 94/170 | 103/277    | 103/277           |
|                       | Organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) | 91/137               | 94/170 | 103/297    | 103/297           |
|                       | Control (without the application of biohumus)               | 74/83                | 75/135 | 84/142     | 84/142            |
Table 2. Influence of various rates of biohumus on the indicators of structure of a harvest of winter wheat of the grades “Skipetr” and “Zimushka” (the average for 2014–2015)

| Variant of experiment | Tilling capacity | Ear length, cm | Quantity of cones per ear, pcs | Quantity of grains per ear, pcs |
|-----------------------|------------------|----------------|-------------------------------|-------------------------------|
|                       | Total productive |                |                               |                               |
| “Skipetr”             |                  |                |                               |                               |
| Developed biohumus (the expense is 100 kg/m²) | 486.0 | 300.0 | 5.6 | 11.0 | 24.0 |
| Developed biohumus (the expense is 150 g/m²) | 566.0 | 307.0 | 6.2 | 13.5 | 28.0 |
| Developed biohumus (the expense is 200 g/m²) | 474.0 | 266.0 | 5.0 | 10.5 | 24.0 |
| Organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) | 562.0 | 303.0 | 6.1 | 13.8 | 26.0 |
| Control (without the application of biohumus) | 332.0 | 280.0 | 4.6 | 10.1 | 16.0 |
| “Zimushka”            |                  |                |                               |                               |
| Developed biohumus (the expense is 100 kg/m²) | 493.0 | 305.0 | 5.9 | 12.0 | 25.0 |
| Developed biohumus (the expense is 150 g/m²) | 575.0 | 312.0 | 6.6 | 14.0 | 29.0 |
| Developed biohumus (the expense is 200 g/m²) | 555.0 | 299.0 | 6.1 | 13.7 | 26.5 |
| Organic mineral fertilizer “Universal’noe” (the expense is 150 g/m²) | 570.0 | 308.0 | 6.5 | 14.0 | 27.0 |
| Control (without the application of biohumus) | 337.0 | 284.0 | 4.9 | 10.6 | 15.9 |

The highest tilling capacity is shown in the variants of experiments with the application of the developed biohumus (the rate is 150 g/m²) of the grade “Skipetr” (the total is 566.0 pieces/m, the productive capacity is 307.0 pieces/m); of the grade “Zimushka” (575.0 and 312.0 pieces/m respectively) and of the organic mineral fertilizers “Universal’noe” (the expense is 150 g/m²), of the grade “Skipetr” (the total is 562.0 pieces/m, the productive capacity is 303.0 pieces/m) and of the grade “Zimushka” (570.0 and 308.0 pieces/m respectively).

Thus, proceeding from the data presented in Tables 1 and 2 it is possible to draw a conclusion that biohumus has a positive effect on the indicators of harvest of the grades of winter wheat “Skipetr” and “Zimushka” under conditions of the Kemerovo region.

Fig. 2 presents the photo of result of use of biohumus for a crop of winter wheat of the grade “Skipetr” in comparison with the control sample (without the use of fertilizers).

As a result of the studies it has been established that the developed biohumus with the application rate of 150 g/m² has the highest positive effect.

The application of biohumus allows to increase the quality indicators of composition of a wheat harvest (Table 3).
Table 3. Change of productivity and quality indicators of winter wheat grains caused by the use of biohumus, t/hectare (the average for 2014–2015)

| Variant of experiment | Productivity, t/hectare | Increase, ± t/hectare | Thousand grain weight, g | Grain unit, g/l | Protein, % | Starch, % | Ash, % |
|-----------------------|-------------------------|-----------------------|--------------------------|-----------------|------------|-----------|--------|
| “Skipetr”             |                         |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.15                    | 0.89                  | 40.00                    | 770.00          | 14.15      | 68.54     | 1.70   |
| (the expense is 100 kg/m²) |                      |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.63                    | 1.25                  | 44.00                    | 783.00          | 15.02      | 67.00     | 1.63   |
| (the expense is 150 g/m²) |                      |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.13                    | 0.92                  | 41.00                    | 772.06          | 14.25      | 63.34     | 1.54   |
| (the expense is 200 g/m²) |                      |                       |                          |                 |            |           |        |
| Organic mineral fertilizer “Universal'noe” | 4.23                  | 1.12                  | 44.00                    | 783.00          | 14.76      | 68.55     | 1.73   |
| (the expense is 150 g/m²) |                      |                       |                          |                 |            |           |        |
| Control (without the application of biohumus) | 3.18                  | –                     | 39.00                    | 758.00          | 13.20      | 63.70     | 1.94   |
| “Zimushka”            |                         |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.33                    | 1.05                  | 42.00                    | 784.00          | 14.56      | 70.70     | 1.90   |
| (the expense is 100 kg/m²) |                      |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.42                    | 1.15                  | 45.00                    | 789.00          | 15.10      | 71.54     | 1.62   |
| (the expense is 150 g/m²) |                      |                       |                          |                 |            |           |        |
| Developed biohumus    | 4.13                    | 1.13                  | 43.00                    | 786.00          | 14.58      | 70.79     | 1.70   |
| (the expense is 200 g/m²) |                      |                       |                          |                 |            |           |        |
| Organic mineral fertilizer “Universal'noe” | 4.53                  | 1.11                  | 44.00                    | 787.00          | 15.00      | 69.03     | 1.69   |
| (the expense is 150 g/m²) |                      |                       |                          |                 |            |           |        |
| Control (without the application of biohumus) | 3.17                  | –                     | 41.00                    | 769.00          | 14.32      | 66.64     | 2.01   |

The positive effect on the indicator of productivity depending on the use of biohumus, as for the grade “Skipetr”, was 0.89, 1.25, 0.92 and 1.12 t/hectare relating to the control samples, and 1.05, 1.15, 1.13 and 1.11 t/hectare for the grade “Zimushka”.

The high weight of 1000 grains characterizes one of the main components of high productivity of wheat winter grains.

A significant amount of authors studied the dependence of productivity and quality indicators of winter wheat grains on the weight of 1000 grains [43–46]. The analysis of results of studies in these works shows that the weight of 1000 grains of the grades of winter wheat studied by the authors changes depending on the features of the grade, the conditions of cultivation of winter wheat seeds and the climatic and weather characteristics of the area.

The studied variants of biohumus with various application rates considerably differ concerning this indicator – the difference in the weight of 1000 grains is on average 5 g. The highest weight of 1000 wheat grains was in the experiment variant where the developed biohumus with the application rate of 150 g/m² – 44 g and the organic mineral fertilizer “Universal'noe” (the expense is 150 g/m²) – 44 g, as well, for the grade “Skipetr” and 45 and 44 g for the grade “Zimushka”, respectively, were used.

Confirming the data submitted in the studies [42–47], we draw a conclusion that the change of weight of 1000 grains depending on experiment variants is characterized by the biological features of a grade of winter wheat and the environmental conditions, however the relative fineness of grain remains.

In the made experiments the grain variants obtained with the application of biohumus had an unequal unit the indicator of which fluctuated within 758–783 g/l for the grade “Skipetr” and a high grain unit was noted in the grade “Zimushka” – within 769–789 g/l. On average, the experiment variants with the application of the developed biohumus with the rate of 150 g/m² and the organic mineral fertilizer “Universal'noe” (the expense is 150 g/m²) have the highest indicators of unit for the grade “Skipetr” and “Zimushka”. Thus, as a result of the studies a considerable positive influence of the developed biohumus on the productivity of wheat grades “Skipetr” and “Zimushka” with the application rate of 150 g/m² has been established.

Table 4 presents the results of determination of protein in the grains of the grades “Skipetr” and “Zimushka” depending on the rates of the applied biohumus.

According to the data presented in Table 4 the application of all rates of the developed biohumus and also the use of the organic mineral fertilizer “Universal'noe” for the crop of winter wheat provides an increase in biosynthesis processes and consequently the growth of percentage of protein in grains.
Table 4. Change of protein content in winter wheat grains caused by the use of biohumus, % (over a period of 2014–2015)

| Variant | Years of studies | Years of studies |
|---------|-----------------|-----------------|
|         | “Skipet”        | “Zimushka”      |
|         | 2014            | 2015            | 2014            | 2015            |
| Developed biohumus (the expense is 100 kg/m²) | 10.95 | 12.84 | 12.24 | 13.27 |
| Developed biohumus (the expense is 150 g/m²) | 11.77 | 14.25 | 13.09 | 14.45 |
| Developed biohumus (the expense is 200 g/m²) | 11.52 | 12.37 | 13.05 | 14.23 |
| Organic mineral fertilizer “Universal'noe” (the expense is 150 g/m²) | 11.52 | 11.91 | 13.15 | 14.01 |
| Control (without the application of biohumus) | 10.78 | 10.86 | 11.35 | 11.43 |

On average, the percent of protein in the grains of the grade “Skipet” with the application of the developed biohumus increased by 1.87%, and by 1.11% for the grade “Zimushka” over a period of 2014 and 2015. The use of the organic mineral fertilizer “Universal'noe” has provided an increase in protein by 0.39 and 1.36% respectively. Presumably, the application of biohumus accelerates the exchange of nitrogenous substances, thus, starting the process of nitrogen reduction.

According to the results of the studies of protein fractions of winter wheat grains, the use of the developed biohumus for the grades “Skipet” and “Zimushka” changes the ratio of protein fractions. The data are presented in Table 5.

Table 5. Change of fractional composition of protein of winter wheat grains depending on the rates of the applied fertilizers, mg/kg (the average for 2014–2015)

| Variant | Albumins | Globulins | Prolamins | Glutelins | Total protein |
|---------|----------|-----------|-----------|-----------|---------------|
| “Skipet” |          |           |           |           |               |
| Developed biohumus (the expense is 100 kg/m²) | 9.17 | 26.53 | 54.12 | 33.91 | 123.73 |
| Developed biohumus (the expense is 150 g/m²) | 9.85 | 27.12 | 55.99 | 34.90 | 127.86 |
| Developed biohumus (the expense is 200 g/m²) | 9.12 | 26.17 | 53.56 | 33.69 | 122.51 |
| Organic mineral fertilizer “Universal'noe” (the expense is 150 g/m²) | 9.54 | 27.04 | 54.89 | 33.72 | 125.19 |
| Control (without the application of biohumus) | 8.83 | 26.17 | 52.65 | 32.64 | 120.29 |
| “Zimushka” |          |           |           |           |               |
| Developed biohumus (the expense is 100 kg/m²) | 9.78 | 29.13 | 54.09 | 33.01 | 126.01 |
| Developed biohumus (the expense is 150 g/m²) | 10.39 | 28.45 | 57.79 | 38.13 | 134.76 |
| Developed biohumus (the expense is 200 g/m²) | 10.19 | 28.41 | 57.61 | 36.89 | 133.10 |
| Organic mineral fertilizer “Universal'noe” (the expense is 150 g/m²) | 9.98 | 29.29 | 54.59 | 35.23 | 129.09 |
| Control (without the application of biohumus) | 9.63 | 27.64 | 53.89 | 33.55 | 124.71 |
Table 6. Economic efficiency of cultivation of winter wheat depending on the rate of application of biohumus (using an example of the grade "Skipetr")

| Parameter                      | Control | Biohumus (the expense is 100 kg/m²) | Biohumus (the expense is 150 g/m²) | Biohumus (the expense is 200 g/m²) | Organic mineral fertilizer "Universal'noe" (the expense is 150 g/m²) |
|--------------------------------|---------|------------------------------------|------------------------------------|------------------------------------|---------------------------------------------------------------|
| Productivity, t/hectare        | 2.97    | 3.96                               | 4.43                               | 4.22                               | 4.15                                                          |
| Production costs, rub/hectare  | 21790   | 29220                              | 33010                              | 32040                              | 32360                                                         |
| Cost per unit of output, rub/hctare | 15230   | 18990                              | 20180                              | 19930                              | 19440                                                         |
| Product unit cost, rub/centner | 510.3   | 445.3                              | 423.8                              | 435.6                              | 437.8                                                         |
| Product sales profit, rub/hectare | 5990    | 11020                              | 12760                              | 12410                              | 12170                                                         |
| Level of profitability, %      | 39.3    | 58.0                               | 63.2                               | 62.2                               | 62.6                                                          |

The analysis of economic efficiency has shown that the largest investments are required during the cultivation of wheat with use of the organic and mineral fertilizer "Universal'noe" – 32360 rub/hectare and the smallest in the variant without the application of biohumus or any other fertilizers – the control variant – 21790 rub/hectare. However, in view of various productivity of grains of wheat which changes depending on the influence of various rates of biohumus, the product unit cost considerably changes and is within the limits of 423.8 (biohumus, the expense is 150 g/m²) – 510.3 rub/centner (control). The variant with the use of the developed biohumus (the expense is 150 g/m²) has the highest level of profitability – 63.2%, the control sample has the smallest level – 39.2%.

Thus, at the level of profitability of 63.2% it is necessary to grow up winter wheat of the grade "Skipetr" with the use of biohumus with the application rate of 150 g/m² on chernozem soils of the land plots of the Kemerovo region for receiving heavy grain crops (4.4–4.7 t/hectare) with high quality indicators.

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