The effect of the solid fraction of pig manure on the biological activity of agrochernozem

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Abstract. The results of studies in 2016-2019 on the effect of the solid fraction of pig manure, applied at doses of 20-60 t/ha, on the indicators of the biological activity of the agrochernozem of loamy quasi-gley medium-arable medium with a low carbon content of organic matter (1.14-1.22%) are presented. The manure contained 0.59% of N, 1.13% of P₂O₅, 0.14% of K₂O, 82.1% of organic matter at pH = 7.4. In field experiments carried out at the experimental field of the Omsk State Agrarian University (Omsk), the cellulose-destroying capacity of the soil and the activity of the enzymes catalase, urease and invertase were studied. The soil of the control variant had a weak cellulose-degrading capacity and was poor in the content of the studied enzymes. The results of the experiments indicate a positive effect of pig manure on the activity of biological processes in the soil. The cellulose-degrading capacity of agrochernozem depended on the dose of manure and the hydrothermal conditions of the growing season. In the period from the first to the third years of manure aftereffect, the intensity of cellulose destruction increased in comparison with the control by 20-90% at a dose of 20 t/ha and by 70-230% at a dose of 60 t/ha. Organic fertilization increased the activity of urease by 34.6-68.0%, by 10.7-14.6% that of catalase and by 5.8-12.2% that of invertase.

1. Introduction

One of the urgent tasks of modern agriculture is the preservation and stabilization of the fertility and ecological state of soils [1,2]. To solve it, an essential role is played by the regulation of the flows of organic matter, macro- and microelements in soils of agrocenoses through the introduction of modern farming systems and the use of fertilizers [3-5]. An insufficient level of their use leads to a negative balance of plant nutrients and humus in the soils of different regions of Russia, including the Omsk Irtysh region [6-7]. Against the background of a decrease in the amount of applied mineral fertilizers, the preservation of soil fertility of agricultural lands is possible with the use of all available biological resources, including organic fertilizers [8]. In the Omsk region, simultaneously with the development of pig breeding, the output of pig manure is increasing. Its use makes it possible to simultaneously solve both the problems of increasing the productivity of agricultural crops and stabilizing soil fertility, and disposing of waste from livestock enterprises. At the same time, an agroecological assessment of the effect of its various doses on the state of soil properties is necessary. To diagnose the ecological state of the soil, it is customary to use indicators of biological activity, which include the cellulolytic and enzymatic activity of the soil [9-12].

Cellulose is one of the most widespread carbon compounds, which contains a significant amount of organic carbon in the biosphere. Decomposition of cellulose is a large-scale natural destructive process...
carried out by a variety of microorganisms and plays an important role in the return of carbon to the atmosphere in the form of carbon dioxide, which is necessary for the process of photosynthesis. In addition, according to the literature, the decomposition of cellulose is directly related to the formation of humic substances in the soil and the formation of the soil structure [11].

Along with cellulolytic activity, it is advisable to determine the enzymatic activity of the soil. According to a number of authors, the activity of soil enzymes is a stable and sensitive indicator for assessing the biological activity of soils [11-13]. The use of indicators of the biological activity of the soil makes it possible to substantiate the ecological expediency of various agrochemical methods for intensifying the production process of agricultural crops. However, there are no data on the biological activity of arable soils in the Omsk region under the conditions of the use of pig manure. In this regard, the aim of the study was to study the effect of pig manure on the indicators of cellulose-destroying and enzymatic activity of agrochernozem of the Omsk Irtysh region.

2. Materials and methods
The studies were carried out in field experiments at the experimental field of the Omsk State Agrarian University (Omsk) in the period from 2016 to 2019. The study area is located in the southern forest-steppe of Western Siberia. The experimental field is located on the second terrace above the floodplain of Irtysh river. The soil of the experimental site is represented by quasi-gley medium-arable medium loamy agrochernozem (Russian soil classification as of 2004). The humus content in the topsoil of the control variant over the years of research was low and varied from 1.97 to 2.10% [14]. The sum of exchangeable bases was 20.2 cmol/kg, the pH of the aqueous suspension was 6.75.

The scheme included the following variants: control; manure 20 t/ha; manure 30 t/ha; manure 40 t/ha; manure 50 t/ha; manure 60 t/ha. The organic fertilizer was delivered from RUSKOM-Agro LLC (Kormilovsky district of the Omsk region). The manure contained was 0.59% of N, 1.13% of P₂O₅, 0.14% of K₂O, 82.1% of organic matter, the pH indicator was 7.4. The manure was introduced in the spring before sowing, followed by incorporation for plowing to a depth of 0–20 cm. The cultivated crop was wheat of “Duet” variety.

The study of indicators of the biological activity of the soil was carried out in variants with manure doses of 20 and 60 t/ha. Soil samples were taken after harvesting the culture with a drill to a depth of 0–20 cm. In the samples taken, the invertase activity was determined according to V.F. Kuprevich with the final determination of sugars according to Bert rand; urease activity was determined according to Hoffman with colorimetric ending; the activity of catalase was determined by the gasometric method [13].

The cellulose-breaking capacity of the soil was determined in the field using the application method according to L.D. Tikhomirova [15]. Cotton cloths were laid vertically in the topsoil 0–20 cm in five replicates. The exposure time of the applicators in the soil was 30, 60 and 90 days.

3. Results
The decomposition of cellulose in soil is carried out by a large group of various microorganisms: aerobic bacteria and fungi, as well as anaerobic mesophilic and thermophilic bacteria [9]. The cellulose-degrading activity of agrochernozem was studied in the period of 1–3 years of the manure aftereffect. It was found that it depended on the exposure period, manure dose and hydrothermal conditions (air temperature and amount of precipitation) (Table 1).

The soil of the control variant in all the years of research was characterized by a weak cellulose-breaking capacity, as per the rating scale according to D.G. Zvyagintsev [9,15]. During the growing season, cellulose decomposition was 13.2-16.8%. The low activity of cellulose destruction in the studied agrochernozem is associated with the low carbon content of organic matter (1.16-1.22%), gross (0.11-0.13%) and mineral nitrogen. The reason for the low humus content of the soil is its susceptibility to processes of planar washout, as well as long-term use in arable land. In comparison with our data, the decomposition of cellulose in meadow chernozem soils with a high humus content (6.5-6.7%) in the stationary experiments of the Omsk Agrarian Scientific Center was significantly higher: from 27.4-31.4
to 47.6% [15]. The hydrothermal conditions of 2017 in the first year of the manure aftereffect were characterized by a lack of moisture and higher air temperatures in comparison with the long-term average data: the amount of precipitation from May to August was less by 46.9%, and the air temperature was more by 2.1 °C. In this regard, after 30 days of the experiment, the weight loss of the webs on the control was only 1.83% at a very low rate of cellulose decomposition (Table 2).

**Table 1. Influence of the aftereffect of pig manure on the intensity of cellulose decomposition in agrochernozem**

| Variant | 30 days | 60 days | 90 days |
|---------|---------|---------|---------|
| 2017, 1 yr. of manure aftereffect |         |         |         |
| Control | 1.83    | 8.29    | 13.2    |
| Manure 20 t/ha | 2.38    | 8.84    | 14.4    |
| Manure 60 t/ha | 2.68    | 9.75    | 22.5    |
| LSD05   | 1.00    | 1.25    | 2.76    |
| 2018, 2 yrs. of manure aftereffect |         |         |         |
| Control | 5.22    | 8.15    | 16.8    |
| Manure 20 t/ha | 6.82    | 12.5    | 20.8    |
| Manure 60 t/ha | 10.4    | 16.5    | 38.6    |
| LSD05   | 1.20    | 1.13    | 2.06    |
| 2019, 3 yrs. of manure aftereffect |         |         |         |
| Control | 3.53    | 7.22    | - a     |
| Manure 20 t/ha | 4.60    | 10.7    | -       |
| Manure 60 t/ha | 8.52    | 14.1    | -       |
| LSD05   | 1.01    | 1.12    | -       |

* no samples were taken

**Table 2. Cloth decomposition rate, % per day**

| Variant | 30 days | 60 days | 90 days |
|---------|---------|---------|---------|
| 2017, 1 yr. of manure aftereffect |         |         |         |
| Control | 0.06    | 0.22    | 0.16    |
| Manure 20 t/ha | 0.08    | 0.22    | 0.9     |
| Manure 60 t/ha | 0.09    | 0.24    | 0.42    |
| 2018, 2 yrs. of manure aftereffect |         |         |         |
| Control | 0.17    | 0.10    | 0.29    |
| Manure 20 t/ha | 0.23    | 0.19    | 0.28    |
| Manure 60 t/ha | 0.35    | 0.20    | 0.74    |
| 2019, 3 yrs. of manure aftereffect |         |         |         |
| Control | 0.12    | 0.12    | -       |
| Manure 20 t/ha | 0.15    | 0.20    | -       |
| Manure 60 t/ha | 0.28    | 0.19    | -       |

By the end of the second exposure period, the rate of cellulose destruction increased significantly to 0.22% per day. An increase in the intensity of the cellulolytic activity of the soil was associated with an increase in the amount of precipitation in July (their amount approximately corresponded to the norm), as well as with an increase in the activity of microbiological processes. During the third term of the experiment, there was a slight decrease in the rate of destruction of cellulose to 0.16% per day.
In the variants with the application of manure, the activity of cellulose decomposition was higher at all observation periods. In 2017, after 30 days, it was 30-46% higher compared to the control. In the second period, the differences in the values of cellulose destruction on the fertilized variants compared with the control were less; a significant increase in the cellulolytic capacity (by 17.6%) was established only with the maximum dose of manure. After 60 days, the rate of cellulose decomposition in all variants of the experiment was similar (0.22-0.24% per day) and slightly higher at a fertilization dose of 60 t/ha.

After 90 days of the experiment, the decomposition rate slightly decreased in the control and at a manure dose of 20 t/ha compared to the previous period. However, in the variant with a manure aftereffect of 60 t/ha, it reached a maximum and exceeded the values in the control by 2.6 times. The cellulose-breaking capacity of the soil in this variant was 70.3% higher than in the control.

In the second year of aftereffect (2018), the process of cellulose destruction took place in generally better conditions, but with uneven soil moisture at the corresponding normal temperatures. The amount of precipitation from May to August exceeded the norm by 15%, in May it was 2 times more than the average long-term data. At the same time, July was dry. Due to weather conditions, the decomposition of cellulose in the first 30 days was more active compared to this period in 2017, as evidenced by the values of the rate of decomposition and decrease in the weight of the cloths. In the next period of exposure, their values decreased, probably due to drier conditions. By the end of 90 days of the experiment, the cellulose activity increased again. During all periods of research, there was a significant increase in the cellulose-decomposition capacity of the soil under the influence of manure. At a dose of 20 t/ha, it ranged from 24.4 to 53.9% compared to the control; at 60 t/ha by the middle and at the end of the growing season it was 2-2.3 times higher than in the control. The decomposition rate of cloths at the highest fertilizer dose was maximum and reached 0.74% per day.

In 2019 (the third year of aftereffect), observations of the cellulose-degrading activity of the soil were carried out in two periods due to low air temperatures in May and late sowing of wheat. Before the cloths were laid in June, precipitation fell above the norm by 30 mm (55.6%); however, July and August were drier. For 60 days, the amount of decomposed cellulose in the control was 7.2%, which is slightly less than in previous years. In variants with the aftereffect of manure, a higher cellulolytic activity remained: at a dose of 20 t/ha it exceeded the control by 30.3-47.9%, at a dose of 60 t/ha it exceeded the control by 1.95-2.4 times.

Thus, the results of the field experiment indicate that the solid fraction of pig manure in 1-3 years of aftereffect significantly increases the cellulose-breaking capacity of the soil. The enhancement of biological activity was associated with a change in a complex of factors: microbiological and nutrient regimes of the soil. In the literature, it is noted that the use of manure is accompanied by a significant increase in the number of all groups of microorganisms, including cellulolytics. This occurs both due to the creation of more favorable nutritional conditions for microflora, and due to the content of microbes in the manure itself, in which anaerobic cellulose-decomposing bacteria of the genus Clostridium and others live. When manure is introduced into the soil, the number of bacteria increases, especially ammonifying and cellulose decomposing bacteria, actinomycetes and fungi [15].

According to our data, the use of pig manure contributed to a significant increase in the carbon content of organic matter by 7-33.6% and mobile humic substances by 38-46% [14]. In addition, the content of mobile forms of nutrients in the soil increased, which enhance the development of cellulose-destroying microflora. It was also found that a higher cellulolytic activity of the studied agrochernozem was observed in periods and years with good moisture.

The enzymatic activity of the soil is one of the most important diagnostic indicators of the impact of anthropogenic loads on agroecosystems. Determination of the activity of soil enzymes is important for assessing the effect of agricultural practices on the rate of biological processes in the soil. In our experiments, we studied the activity of soil enzymes belonging to the class of hydrolases and oxidoreductases and playing an important role in the formation of soil fertility (Table 3). Invertase and urease are involved in the hydrolytic decomposition of organic substances, catalase is involved in redox reactions. They take part in the main processes of soil humification, so their activity is an indicator of soil cultivation.
According to D.G. Zvyagintsev [9, 15], the soil of the control variant was assessed as poor in the content of catalase (1.57-1.87 (cm$^3$/min)/g) and invertase (4.9-6.9 mg/g) and very poor in the content of urease (0.19-0.26 mg/g). At the same time, over the years of research, the dynamics of enzyme activity was noted.

Table 3. Enzymatic activity of agrochernozem under conditions of using solid fraction of pig manure

| Variant         | Catalase, O$_2$ (cm$^3$/min)/g | Urease, mg/g | Invertase, mg/g |
|-----------------|--------------------------------|--------------|-----------------|
|                 | 2016, manure effect             |              |                 |
| Control         | 1.70                            | 0.19         | 6.90            |
| Manure 20 t/ha  | 1.85                            | 0.22         | 7.30            |
| Manure 60 t/ha  | 1.93                            | 0.32         | 7.30            |
|                 | 2017, 1 yr. of manure aftereffect|              |                 |
| Control         | 1.57                            | 0.26         | 6.30            |
| Manure 20 t/ha  | 1.72                            | 0.26         | 6.30            |
| Manure 60 t/ha  | 1.80                            | 0.35         | 7.00            |
|                 | 2018, 2 yrs. Of manure aftereffect|              |                 |
| Control         | 1.87                            | 0.21         | 4.90            |
| Manure 20 t/ha  | 1.83                            | 0.30         | 5.50            |
| Manure 60 t/ha  | 2.07                            | 0.23         | 5.50            |

The highest level of catalase content was noted in 2018, that of urease, in 2017, that of invertase, in 2016 For comparison, it should be noted that the studied soil is close in the activity of urease and catalase, but significantly inferior in the activity of invertase to medium-humus meadow-chernozem soils in stationary experiments of ASC “Omsk” [15].

In variants with manure, an increase in the activity of enzymes was observed, depending on the dose of fertilizer, its duration and aftereffect. In the year of manure effect, a significant increase in the action of catalase and urease was noted. The catalase activity, as compared to the control, was 8.8 and 13.5% higher at doses of 20 and 60 t/ha, respectively. The urease activity increased most significantly: up to 68.4% in the variant with manure of 60 t/ha. The smallest changes were found for invertase activity. Under the influence of manure, it increased by only 5.8%, while it did not depend on the fertilizer dose.

In the first year of the aftereffect of manure, its positive effect on catalase activity persisted. An increase in the activity of urease and invertase occurred only at the maximum dose of manure. At the same time, the increase in urease activity was less than in the year of action (by 34.6%), while invertase, on the contrary, was somewhat higher (11.1%). It should be noted that the arid conditions of the 2017 growing season could have affected the enzyme activity.

In the second year of aftereffect, an increase in catalase activity was noted in the variant with a manure dose of 60 t/ha. Urease and invertase activity was higher for all fertilized variants. At the same time, the urease activity was maximum at a manure dose of 20 t/ha. The effect of invertase, with its general decrease in 2018, did not depend on the fertilizer dose and exceeded the control by 12.2%. In general, it should be noted that the enzymatic activity of agrochernozem during fertilization with manure remained weak; however, the activation of the activity of all enzymes was noted.

4. Conclusion

Thus, the studies carried out have shown that the use of solid fraction of pig manure is a significant factor in increasing the biological activity of the soil. The increase in the content of organic matter carbon, mobile humic substances and nutrients in the soil led to an increase in the cellulolytic and enzymatic activity of the soil. The degree of cellulose decomposition during the growing season in 1–2
years of manure aftereffect increased from 13.2-16.8% in the control to 14.4-20.8% at a dose of 20 t/ha and up to 22.5-38.6% at a dose of 60 t/ha. The positive effect of manure on the destruction of cellulose persisted in the third year of aftereffect. The most stable and significant effect on the activity of urease was exerted by manure at a dose of 60 t/ha in the year of effect and the first year of aftereffect. Organic fertilizer had a weaker effect on the activity of catalase and invertase, increasing it by 10.7-14.6 and 5.8-12.2%, respectively. In general, the studies carried out indicate a positive effect of manure on biological processes in agrochernozem.

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