Soil treatment using natural biocoenosis

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Abstract. The paper considers the possibility of using different biocoenosis for soil treatment of oil contamination. It is indicated that the use of artificial biocoenosis is expensive and not always effective. It is shown that the use of natural biocoenosis, for example, in the form of activated sludge taken from the treatment facilities of oil refineries, is much cheaper and not inferior in effectiveness to the action of biological substance preparations (biological preparations) based on artificial biocoenosis.

Physical-chemical and biotechnological methods of soil treatment from oil pollution are most widely known, and they have proven to be effective and natural means of protecting the natural environment from pollution [1-20]. Most of the known biotechnologies are based on introducing microorganisms - destructors of oil hydrocarbons into the polluted environment [1 - 5].

Nowadays several domestic and foreign preparations for soil treatment of oil pollution have been developed and widely advertised: putidoil, devoroil, etc. [1-5]. The selection of microorganisms - destructors is based on their ability to grow intensively on nutrient media with hydrocarbons or crude oil as the only carbon sources.

In the practical application of such preparations, the main indicator of their effectiveness is the eventual result - soil treatment of oil pollution.

Currently, the effectiveness of the microbiological method for soil treatment of oil pollution is beyond doubt. However, as studies in recent years have shown, this method has fairly clear limitations. Firstly, this applies only to seasonal use (in the warm season with a temperature of at least 10-15°C), and secondly, when creating optimal cultivation conditions in the soil (moisture, the presence of necessary mineral elements, soil aeration, etc.) and, thirdly, the desired overseeding of cultures of microorganisms with the achievement of their content in the soil approximately 10⁵ - 10⁸ cells/g soil. Even under optimal conditions, this process is long-standing (lasting from about a month to several months).

As research results show, the activity of microorganisms in most cases increases in the state of immobilization. In this regard, some developers recommend introducing cultures of microorganisms and their associations in a state of immobilization (in adsorbed form). Recently, there is information about the use of biological substance preparations (biological preparations), including hydrocarbon-oxidizing microorganisms or their associations on adsorbents. Research in this direction continues.

As sources of mineral nutrition, we can use the following media (for example, the cultivation of yeast of the Candida microorganisms): (NH₄)₂SO₄ – 5.0 g/l; KH₂PO₄ – 0.85 g/l; K₂HPO₄ – 0.15 g/l; MgSO₄ 7H₂O – 0.5 g/l; NaCl – 0.1 g/l; CaCl₂ 4H₂O – 0.1 g/l; crude oil – 0.1 vol.%; H₂O up to 500 ml.
Using such nutrient media and a standard production scheme (Figure 1), we obtain experimental batches of biomass, which were used in natural conditions for soil treatment of oil pollution.

\[ \text{Figure 1. Apparatus and technological scheme for obtaining a biological preparation for soil treatment of petroleum products.} \]

Obtaining biomass according to the scheme shown in Figure 1 allows having a living producer that can be stored for several months.

The resulting producer and the above nutrient media are added to the oil-contaminated soil, which is also moistened and loosened.

The hydrocarbon-oxidizing Candida microorganisms that we used did not give the high expected result. Moreover, the high cost of biological preparations for soil treatment of petroleum products and certain difficulties in their development lead to the need to search for and use various wastes, for example, surplus activated sludge generated during biological water purification. In this regard, we proposed to use natural biocoenosis in the form of activated sludge taken from the treatment facilities of an oil refinery.

In this case, the technology of using activated sludge was implemented in the form shown in Figure 2.

\[ \text{Figure 2. Scheme of using surplus activated sludge as a biological preparation for soil treatment of petroleum products.} \]
Activated sludge, adapted to the oxidation of oil hydrocarbons, can be an effective biological preparation—a destructor of oil hydrocarbons since the microorganisms of such sludge are more efficient than sludge cultures of urban treatment facilities.

Comparing the technological methods of biological treatment of soil and grounds with the already developed technology of biological water treatment, it should be noted that the first is at the stage of creation, and the second is at the stage of improvement. In our opinion, a complex solution is also possible, when the activated sludge of biological treatment facilities of industrial waters, mainly oil refineries, adapted to the oxidation of petroleum products, is introduced into the contaminated soil or the soil in the form of a suspension. The transportation of the activated sludge suspension can be carried out both through pipelines and using special vehicles.

In case of the remoteness of objects where it is necessary to carry out biological treatment of soil or grounds, activated sludge is supplied in the form of dried biomass with a moisture content of no more than 10%. The technology for obtaining dry biomass of activated sludge with a moisture content of no more than 10% was developed by us [2]. This technology includes the following main stages. A suspension of surplus activated sludge from secondary clarifiers of biological treatment facilities with a concentration of about 0.8 – 1.0% for absolutely dry substances (ADS) is thickened using a pressure flotation unit to 2.5 - 3.5% for ADS and the thickened concentrate is sent to further thickening into a vacuum-evaporator installation. The resulting thickened suspension of activated sludge with a concentration of about 8-10% by ADS is fed to a spray dryer. The resulting activated sludge biomass has a moisture content of no more than 10%. The storage time of such biomass is at least six months.

The disadvantage of this technology is that some of the microorganisms die during dehydration. This is undoubtedly a negative factor, but taking into account that activated sludge is a waste, part of the dead biomass can be considered as a substrate for the vital activity of living organisms. There are other, more gentle methods of dehydration of microorganisms, including activated sludge. This primarily relates to the method of drying a suspension of microorganisms. It is more rational to use vacuum freeze or other methods that allow the vast majority of microorganisms to be kept alive during drying [1-32].

The use of surplus activated sludge from biological treatment facilities, mainly of oil refineries, as preparation for treating soil and ground from oil pollution, will, within certain limits, reduce the price of biological preparations of this type. In addition, this technical solution will facilitate the disposal of such a large-tonnage waste as surplus activated sludge from biological treatment facilities of oil refineries, the capacity of which has increased in recent years.

The method of processing oil-contaminated plots using biological preparations includes the following main stages of work.

1. Determination of the required dose of the introduced biological preparation according to the initial values of the concentration of petroleum products.
2. Creating a biological preparation for use by mixing biomass with water (diluting the biomass).
3. Loosening of the soil and grounds and the introduction of loosening components and mineral nutrients into them, as well as moistening the soil and grounds.
4. Direct introduction of the biological preparation in the form of an aqueous suspension into the soil and grounds.

The sequence of performing these works should be strictly observed, while the planned measures should, if possible, be performed on sunny days when no precipitation is observed. The latter circumstance is very important since a significant part of the applied preparation can be washed away by storm streams, thus, the effectiveness of soil and ground treatment with a biological preparation will be minimized.

After determining the area of the contaminated plots, the dose of the biological preparation necessary for the treatment of oil-contaminated soils and grounds is established. Then based on the consumption rate of 10 - 20 kg/ha, depending on the degree of contamination of the plot the calculation is carried out. In the considered case, according to the results of primary observations, the rate of soil and ground cultivation was proposed, which is equal to 15 kg/ha. This amount of biological
preparation was diluted (mixed with water) in a container with a volume of 180 liters. As it was diluting, the mixture of the biological preparation was stirred with nitroammophos (ammonium nitrate phosphate fertilizer) at the rate of 30 g per 1 m² of area. After two hours, the prepared mixture was applied to the soil by hand spraying.

At the same time, the mineral feed was added to the prepared mixture at the rate of 100 kg per 1 hectare of the surface. The composition of the mineral feed was selected according to the data in Table 1. The preparation and mineral salts were periodically mixed. The working solution was applied to the contaminated surfaces using a pump - a dispersing nozzle in doses by Table 2. At the same time, for a uniform distribution of the biological preparation, all the water was intensively mixed. Humidity in the plot treated with a biological preparation should be 60 - 80%. In dry weather, watering the treated plot is necessary. If the soil is heavily contaminated with petroleum products, secondary treatment with a biological preparation is possible without the introduction of fertilizers.

Table 1. Dose of applying mineral fertilizers to the contaminated soil surface.

| Mineral fertilization                      | The dose of fertilization, kg/m³ of working solution |
|--------------------------------------------|-----------------------------------------------------|
| Ammophos (monoammoniumphosphate)          | 3                                                   |
| Azophoska (nitrogen-phosphorus-potassium fertilizer) | 3                                                   |
| Ammoniumsulfate + ammophos                | 1 + 1                                               |
| Ammonium sulfate + simple superphosphate (monocalcium phosphate) | 1 + 3                                               |
| Ammoniumsulfate + doublesuperphosphate    | 1 + 1                                               |

Table 2. Dose of biological substance preparation application to the contaminated soil surface.

| Degree of surface contamination with petroleum products, % | Dose of biological preparation introduction, kg |
|------------------------------------------------------------|-----------------------------------------------|
| >5                                                         | 1 – 8                                         |
| 6 – 20                                                     | 10 – 18                                       |
| < 21                                                       | 18 – 25 and more                              |

Visual determination of the effectiveness of the preparation and the resulting destruction of petroleum products on the soil surface for the initial assessment was determined by the change in color from gray to dark reddish. The aggregate composition of petroleum products in the soil varied from a viscous liquid to easily decomposable solid particles with a putrescent smell while wet. Pieces of soil with residues of decomposed oil were not combustible.

The biological preparation was used after the suspension of microorganisms of activated sludge was thickened in a pressure flotation unit. The concentration of biomass in the thickened product was 3-4 times higher than in the initial suspension. The thickened product was used to treat the soil contaminated with petroleum products. Control over the content of petroleum products was carried out once a week. The results of studying the effect of soil treatment with a biological preparation in the form of a condensed product of biomass of microorganisms are presented in Table 3.

The results presented in Table 3 show that when treated with a biological preparation (a thickened product of the biomass of microorganisms of activated sludge) of soil contaminated with petroleum products, the content of the latter decreases in the soil much faster than their content in the soil not treated with the biological preparation.

The experience of microbiological soil treatment allows drawing some conclusions and make recommendations.

1. Neutralization of soils and grounds contaminated with petroleum products using a biotechnological method sharply accelerates the degradation of oil hydrocarbons while optimizing the sphere of vital activity of microorganisms (nutrient medium, humidity, temperature, etc.)
The most favorable for the microorganism’s vital activity is soil moisture in the range of about 50 - 80%, the temperature that is not lower than 8 - 10 °C, the presence of sources of potassium, nitrogen, phosphorus, and trace elements in the soil along with oil hydrocarbons.

Under optimal conditions, the processes of degradation of oil hydrocarbons are accelerated as a result of the vital activity of microorganisms present in the soil.

2. To maintain a high rate of degradation of oil hydrocarbons, it is necessary to maintain the presence of microorganisms in the soil in optimal concentrations (approximate biomass consumption is 10 - 80 kg per 1 ha).

**Table 3.** Change in the content of petroleum products in the soil when it is treated with a condensed biomass product of microorganisms of activated sludge in the soil without treatment.

| Number of days after the start of treatment | Concentration of petroleum products in soil, g/kg |
|--------------------------------------------|-------------------------------------------------|
|                                            | Treated area | Untreated area |
| 0 (start)                                  | 7.3          | 7.3            |
| 5                                          | 4.1          | 7.15           |
| 10                                         | 2.9          | 7.07           |
| 15                                         | 2.1          | 6.93           |
| 20                                         | 1.6          | 6.80           |
| 25                                         | 0.9          | 6.71           |
| 30                                         | 0.4          | 6.62           |
| 35                                         | 0.2          | 6.54           |
| 40                                         | 0.15         | 6.47           |
| 45                                         | 0.11         | 6.39           |

For carrying out practical measures, we recommend the following approximate plan (Table 4).

**Table 4.** Approximate plan of measures for soil treatment contaminated with oil and petroleum products.

| №  | Measure                                                                                           |
|----|---------------------------------------------------------------------------------------------------|
| 1  | Determination of the area of plots and drawing up a diagram of their location.                    |
| 2  | Inspection of contaminated soil (or water surface). Determination of control points (at least five) |
| 3  | Commission sampling for the content of petroleum products (100 g each)                           |
| 4  | Analysis of soil samples                                                                         |
| 5  | Determination of the dose of a biological preparation and the total need for a biological preparation |
| 6  | Determination of the need for mineral fertilizers and their delivery                             |
| 7  | Loosening the surface of oil-contaminated soil                                                   |
| 8  | Provision of technological machines for creating and application of the working solution        |
| 9  | Irrigation of soil plots with a working solution of a biological preparation with mineral fertilizers, 4 - 5 m³/ha (in the absence of atmospheric precipitation) |
| 10 | Sampling and analysis of soil (water) samples to determine the concentration of hydrocarbons after treatment with a biological preparation (after one to two weeks, one month, etc.) |

The presented plan (Table 4) was implemented with positive results when testing the soil treatment method in the Tula region of the Russian Federation.

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