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ECOLOGY

EFFECT OF OIL CONTAMINATION ON MICROBIOLOGICAL PROCESSES IN SOILS

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The effect of oil contamination of natural microbiocenoses and biological activity of various types of soils is examined. It is shown that the biological activity indices and the amount of residual oil products depend on the properties of the original soil and the importation dose.
Key words: *oil contamination, microorganisms, soils.*

Preservation of natural and economic complexes and systems, maintenance of their integrity and life-support functions for stable development of society, improvement in the quality of life, enhancement of the health of people and the demographic situation, assurance of ecological safety of the countries and planet on the whole are referred to as strategic trends in the ecological politics of the world community.

At the present time, the problem of the protection and recultivation of soils that have been subjected to chronic oil spills has become a serious problem in our country. Oil contaminations are referred to as anthropogenic effects of a catastrophic order, evoking a rapid responsive reaction. Cases of soil contamination by oil and oil products, which results in loss of the earth's productivity, and thereafter also in complete degradation of landscapes, are increasing in number in the areas of oil fields and on lands traversed by pipelines.

The content of oil in a soil diminishes rapidly in the first three months, and thereafter, this process is slowed. The following are basic causes of reduction in oil content in soil: evaporation of volatile fractions, mineralization of the oil, physical carry-off by water flows, and lithification.

Disruption of the normal functioning of all components of the biogeocenoses, and also their integrity occurs in areas of the natural medium that have been contaminated by oil and oil products: on contact with oil, vegetation perishes immediately, or after two-three vegetative periods, and will not recuperate over the long term.

Oil contamination leads to a profound change in all links of the natural biocenoses. A common characteristic of all oil-contaminated soils is a change in the number of and range of specie diversity of pedobionts (soil meso- and microfauna and microflora). Here, the types of responsive reactions of various groups of pedobionts to

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the contamination will differ: the number of some will increase, others decrease, while the population of a third group will remain essentially constant. As the content of oil in the soil diminishes, their number will be restored to the initial level, but the number of oil-oxidating bacteria will still exceed the same groups in the uncontaminated soils over the long term. Changes in the status of the complex of soil microorganisms can be used for bioindication of soils contaminated with oil and oil products.

On the whole, the population of microorganisms in the soil profile decreases with diminishing concentration of hydrocarbons [1].

The microorganisms are characterized by high sensitivity to the condition and environment of their habitation, and vigorously react to changes that occur in the soil under the action of natural and anthropogenic factors. Moreover, changes may occur not only in the specie structure of microbe associations of the soil, but also in their biochemical activity [2].

The Caspian Scientific-Research Institute of Arid Agriculture has evaluated the effect of oil contamination on the status of various types of Russian soils; this made it possible to increase the volume of the sample and obtain their more objective comparative characteristics.

Table 1

Paraffins	Content in crude, wt. %
C_7H_{16}	4.12
C_8H_{18}	1.45
C_9H_{20}	1.11
$C_{10}H_{22}$	1.19
$C_{11}H_{24}$	1.85
$C_{12}H_{26}$	2.14
$C_{13}H_{28}$	2.49
$C_{14}H_{30}$	2.32
$C_{15}H_{32}$	2.13
$C_{16}H_{34}$	2.11
$C_{17}H_{36}$	1.84
$C_{18}H_{38}$	1.98
iso-$C_{19}H_{40}$	1.03
$C_{19}H_{40}$	1.90
iso-$C_{20}H_{42}$	0.72
$C_{20}H_{42}$	1.83
$C_{21}H_{44}$	1.56
$C_{22}H_{46}$	1.37
$C_{23}H_{48}$	1.33
$C_{24}H_{50}$	1.10
$C_{25}H_{52}$	1.08
$C_{26}H_{54}$	1.07
$C_{27}H_{56}$	1.02
$C_{28}H_{58}$	1.00
$C_{29}H_{60}$	0.97
$C_{30}H_{62}$	0.78
$C_{31}H_{64}$	0.76

Table 2

Soil	Oil content in soil, liters/m^2								
	2.5			5			10		
	1 extraction	2 extraction	3 extraction	1 extraction	2 extraction	3 extraction	1 extraction	2 extraction	3 extraction
Bottomland	4.1	4.4	4.2	7.4	8.1	8.0	27.0	26.6	26.4
Light-chestnut	5.6	5.4	5.6	16.5	15.4	16.1	24.0	29.5	29.8
Light-chestnut, heavily alkaline	3.2	3.1	3.1	9.8	9.9	9.94	24.6	29.8	26.25
Brownish	3.6	3.6	3.4	4.8	5.7	5.6	23.6	23.1	21.24
Meadow	5.1	5.0	5.2	13.7	15.5	15.25	27.0	27.1	27.6
$LED_{0.95}$*	0.2	0.1	0.1	0.3	0.2	0.2	0.1	0.1	0.3

Note. *LED – the least essential divergence.

Table 3

Oil dose, liters/m^2	Population of microorganisms (depth of from 0 to 25 cm)							
	fungi		nonspore-bearing bacteria		bacilli		actinomycetes	
	millions per 1 g of soil	percent of initial	millions per 1 g of soil	percent of initial	millions per 1 g of soil	percent of initial	millions per 1 g of soil	percent of initial
$LED_{0.95}$	0.03		0.1		0.1		0.06	
Soddy-podzolic soil								
0	0.2	100	1.5	100	0.4	100	0.4	100
2.5	0.3	150	1.7	113	0.2	50	0.4	100
5	0.6	300	2.1	140	0.3	75	0.3	75
10	0.6	300	1.8	120	0.2	50	0.1	25
Black earth								
0	0.2	100	2.2	100	1.3	100	0.8	100
2.5	0.5	250	2.2	100	1.5	115	1.1	137
5	0.5	250	2.7	122	1.3	100	0.8	100
10	0.8	400	3.1	140	1.3	100	0.4	50
Light-chestnut soil								
0	–	–	3.4	100	2.7	100	1.2	100
2.5	–	–	3.5	103	2.6	96	1.7	141
5	–	–	4.5	132	3.1	114	1.4	116
10	–	–	2.9	85	2.9	107	0.8	66

The following Northern Caspian typical soils were the subjects of our investigation: brownish, light-chestnut, strongly alkaline, meadow and bottomland, as well as soddy-podzolic soil of the Moscow Administrative Region and black earth from the Tambov Administrative Region.

The experiment was performed in lysimetric vessels [3] with an area of 0.25Ч0.25 m. Crude oil in the following different doses was poured into the vessels containing the soil specimens: 2.5, 5, and 10 liters/m^2. The crude for the analyses was recovered from the Doleansk field in the Liman zone of the Astrakhan' Administrative Region. Parameters of the paraffin hydrocarbons of the oil employed for the experimental investigations are presented in Table 1. The species and quantitative composition of microorganisms in the test samples of the soils were subsequently studied.

The first set of soil samples was taken after one year, the second after two years, and a third after four years. The samplings were repeated four times. The soil samples and crudes were tested in a testing laboratory maintained by the FGU State Center of the Agrochemical Service "Astrakhanskii." The experiment concerning the effect of all oil and oil products on the microbiologic activity of soils was conducted at the Criminal Examination center of the Ministry for Internal Affairs.

The primary purpose of the study was to ascertain laws and mechanisms governing the transformation of the organic matter of soils from European Russia when subjected to oil contamination, and determine laws governing the variation of basic indicators of the physical and physico-chemical properties of the soils, as well as evaluate the overall trends of the effect of oil on their microbiological activity.

The change in the content of hydrocarbons (g/kg) was observed in the soils (Table 2).

Based on results of this experiment, the brownish soil exhibited the highest sensitivity to the action of petroleum hydrocarbons and capacity for self purification, since after importation of the three different dozes of oil to the soil, the hydrocarbon content was low, suggesting that the soil had coped with the indicated loading that it had received. The lowest rate of transformation of petroleum hydrocarbons was noted in the light-chestnut and meadow soils.

Overall trends in the effect of oil and oil products on the microbiological activity of the soils were observed under the conditions of this experiment. The population of microorganisms was determined over the two-month period after the contamination by universally accepted methods.

During the course of the experimental investigations, insignificant fluctuation in the number of individual species of microorganisms was exposed under the weak contamination (oil dose of 2.5 liters/m^2). Actinomycetes are referred to these microorganisms. Changes in the composition and rate of change in microbiological processes were observed at the median level of contamination (oil dose of 5 liters/m^2) (Table 3). At the high level of contamination (oil dose of 10 liters/m^2), development of individual species and suppression of others (actinomycetes of the Niger group) were noted.

Structural changes in the functioning of ecosystems under various soil-ecological conditions were determined by the participation of various groups of soil microorganisms in biochemical processes.

There are also specie characteristics of microorganisms in the functioning of various ecosystems; this is confirmed by our investigations (Table 4). Thus, species that participate in the decomposition of organic matter in the early stages, for example, the bacilli Bac. agglomeratus, Bac. cereus, and Bac. virgulus, emerge as dominants in soils with a low activity of gum-formation processes (soddy-podzolic). Deeper transformation of organic matter takes place with the participation of Bac. idosus, Bac. mesentricus, etc.

A sharp reduction in the population of the group of actinomysetes, and an increase in the number of fungi were characteristic features of all the soil types investigated when the oil dose was increased. As for the bacilli and nonspore-bearing bacteria, however, their population decreased with a contamination of 5 liters/m^2, increased,

Table 4

Species of bacilli	Oil dose, liters/m^2				$LED_{0.95}$
	0	2.5	5	10	
Soddy-podzolic soil					
Bac. agglomeratus	8.5	9.8	12.8	9.0	0.4
Bac. idosus	25.1	27.5	20.4	18.2	1.0
Bac. megatherium	27.0	25.1	28.8	23.5	1.2
Bac. mesentericus trevisan	1.2	1.4	1.8	1.2	0.1
Bac. mesentericus niger	–	–	–	–	–
Bac. cereus	6.2	7.4	9.0	6.3	0.3
Bac. virgulus	1.3	2.7	2.2	1.0	0.2
Bac. asterosporus	3.5	3.3	3.0	3.0	0.2
Bac. gasificans	–	–	–	–	–
Black earth					
Bac. agglomeratus	0.6	1.5	2.7	1.2	0.2
Bac. idosus	25.3	29.3	20.4	18.0	0.8
Bac. megatherium	43.5	54.5	36.2	36.0	1.4
Bac. mesentericus trevisan	8.6	9.9	12.0	7.2	0.3
Bac. mesentericus niger	–	–	–	–	–
Bac. cereus	5.9	8.7	10.5	7.1	0.3
Bac. virgulus	1.2	3.1	2.5	1.0	0.2
Bac. asterosporus	1.4	1.8	2.0	1.6	0.1
Bac. gasificans	–	–	–	–	–
Light-chestnut soil					
Bac. agglomeratus	0.3	1.5	1.8	1.0	0.2
Bac. idosus	14.7	18.0	11.7	8.3	0.6
Bac. megatherium	25.9	24.6	27.9	16.6	1.0
Bac. mesentericus trevisan	22.0	29.4	32.4	15.0	0.9
Bac. mesentericus niger	9.1	13.5	18.6	14.2	0.5
Bac. cereus	1.4	2.6	3.7	2.0	0.3
Bac. virgulus	–	–	–	–	–
Bac. asterosporus	4.3	6.9	5.0	2.8	0.5
Bac. gasificans	0.9	1.4	1.9	1.9	0.2

and then, decreased at a contamination of 10 liters/m^2. It must be noted that in the light-chestnut soils, almost no fungi were observed. The specie composition of the bacilli (thousands per 1 g of the soils) is presented in Table 4.

Contamination of the soils with the oil led to a change in the population and correspondence between the various groups of microorganisms. Thus, importation of oil in a dose of 10 liters/m^2 in the soddy-podzolic soil increased the fungi (by a factor of three) and the nonspore-bearing bacteria (by a factor of 1.2) populations, but did not alter the overall bacilli population, and reduced the number of actinomycetes (by a factor of four).

This rule was also noted in the black-earth specimens; here, the number of fungi increased by a factor of four, and the nonspore-bearing bacteria by a factor of 1.4, while the population of actinomycetes decreased by a factor of two. In the light-chestnut soil, the oil dosage of 10 liters/m^2 resulted in a small increase in the population of the fungi (by anonspore-bearing bacteria (by a factor of 1.2), and a decrease in the number of actinomycetes (by a factor of 1.5). No fungi were observed in this soil. It should be pointed out that importation of an oil dosage of 2.5 liters/m^2 in the black-earth and light-chestnut soil specimens gave rise to an increase in antinomycete population of 1.4 times.

Rules governing specie variation in the bacilli were noted under the different oil-contamination conditions. An oil dosage of 2.5 liters/m^2 stimulated growth in the population of the majority of representatives of the species investigated (with the exception of Bac. megatherium) in all soils. An oil dose of 5 liters/m^2 grew the population of Bac. agglomerates, Bac. mesentericus trevisan, Bac. cereus, and Bac. virgulus in the soddy-podzolic soil; Bac. agglomerates, Bac. mesentericus trevisan, Bac. cereus, Bac. virgulus, and Bac. asterosporus in the black earth; and, Bac. agglomerates, Bac. megatherium, Bac. mesentericus trevisan, Bac. cereus, Bac. asterosporus, Bac. gasificans, and Bac. mesentericus Niger in the light-chestnut soil. An oil dose of 10 liters/m^2 increased the number of Bac. agglomerates in the soddy-podzolic soil; and, Bac. agglomerates, Bac. cereus, Bac. mesentericus Niger, and Bac. gasificans in the light-chestnut soil. In contrast to the soddy-podzolic soils and black earth, the bacilli Bac. mesentericus Niger and Bac. gasificans, the population of which increased with increasing oil dosage of up to 5 liters/m^2 in the soil, were present in the light-chestnut soil.

In the soils with a weak flow of transformation processes (for example, in the soddy-podzolic soils), the species participating in the early stages of the decomposition of organic matter (Bac. cereus, Bac. virgulus, and Bac. agglomerates) emerged as dominants.

The reproductive function of the anctinomycetes was disturbed under the influence of the contamination; this was expressed as an increase the asporogenic forms, and the appearance of actinomycetes of the Niger group, as well as phytopathogenic fungi. The phytopathogenic fungi occupied the ecological niches that had been vacated.

When the soils were contaminated with oil, the activity of the formation of spore-bearing organs of penicillin increased, and a direct relationship was observed between the sharp increase in the number of spore-bearing fungi and the increase in the dosage of oil (Table 5).

Investigation of the soddy-podzilic and light-chestnut soils and black earths with different degrees of oil contamination exposed the direct effect of oil concentrations on the population of various groups of microorganisms in these soils. Low doses of oil had a stimulating effect on the activity of microorganisms, while the higher doses of oil (10 liters/m^2) inhibited the activity of the soil biota.

The reproductive functions of the actinomycetes were disturbed under the influence of the contamination; this was expressed as an increase in the percentage of their asporogenic forms, and the appearance of actinomycetes of the Niger group, as well as phytopathogenic fungi (Table 5).

Table 5

Oil doze, liters/m^2	Number of spore-bearing fungi of genus Penicillium		
	soddy-podzolic soil	black earth	light-chestnut soil
0	6.8	4.5	–
2.5	40	22	–
5	80	50	–
10	67	85	–

When contaminated with oil, therefore, profound disturbances occur in the structure of the complex of soil microorganisms; this finds expression as a change in the population and specie composition of the latter, and, consequently, also in the level of biochemical activity of the soils. Here, a change in the functional integrity of the microbiota, which provides for stability of the microbiocenoces that are accumulating in the soil, leads to its partial or complete degradation.

Based on the experimental data obtained and analysis of natural characteristics of various types of Russian soils, it is possible to conclude an increase in risk of accumulation of oil products in topsoil in the direction from south to north. Here, the properties of the soil, landscape-geochemical situation, and the buffering action of all ecosystems are of major significance.

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