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Application of biopreparation "Rhoder" for remediation of oil polluted polar marshy wetlands in Komi Republic

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Abstract

This paper describes the testing and corresponding results of the preparation "Rhoder" in comparison with several other bioremediation variants during the field trials in Komi Republic throughout 2002–2003. All bioremediation trials were performed on one vast polar marshy wetland polluted by accidental crude oil spill and uncovered by grass. After application of the "Rhoder" at the site, with an area of ~2000 m^2, during the cold and rainy summer of 2002 (1.5 months), the level of oil contamination decreased by 20–51%, depending on initial oil pollution (458–738 g/kg dry weight of soil). In the middle of September 2002, the treated site was covered by 70–85% with green grass. Though, during 2003, the "Rhoder" treatment was not practiced, at the end of August 2003, the site was already covered by 85–95% with green grass and the level of oil contamination further decreased by 54–79% from the initial level of oil pollution at the beginning of 2002. These results were much better compared to those from other bioremediation variants applied at this spill.

Keywords: Bioremediation; Oil polluted polar marshy wetland; Komi Republic; Preparation "Rhoder"

1. Introduction

Most of functioning oil fields in Russia are situated in the northern regions of the European part of Russia (Komi Republic) and in Western Siberia. Severe climatic and geological conditions make not only oil production and transportation difficult, but also the application of remediation methods for accidental oil spills. A specific climate with short and cold summers lasting only 1–1.5 months and long and severe winters lasting 6–7 months with temperature fluctuations from plus 30–35 °C in July to minus 45–55 °C in January causes pipeline ageing and damage even for the modern pipelines with special anticorrosion covering. As a result, accidental oil spills unfortunately are frequent in these regions of Russia. There are more than 200,000 ha of contaminated soils with a thickness of oil layer greater than 5 cm in Western Siberia (Green Peace, 1998). Thus, extensive areas of tundra, forest–tundra and taiga are heavily contaminated and need remediation works. However, vast and impenetrable wetlands and marshy soils made impossible an application of the majority of conventional methods for land reclamation such as landfarming (ex-situ), bioventing, composting, etc., there. Harsh geological and climatic conditions in such region in Komi Republic impelled the company "Lukoil" to seek for the most effective and economically sound technologies for remediation of vast oil polluted territories. This paper describes the results of the preparation "Rhoder" applied to field tests on the polar marshy wetland during 2002–2003 in comparison with other bioremediation variants.

2. Materials and methods

The preparation "Rhoder" (Certificate No. 77.99.04. 515D.004855.08.01 issued by the Russian Ministry of

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Health) developed by us during the 1990s was successfully tested in Western Siberia on different types of oil contaminated wetlands, marshy soils and water surfaces between 1995 and 2000 (Murygina et al., 2000), as well as in laboratory and pilot tests in Komi Republic during 2001 (Mouryguina and Kalyuzhnyi, 2001).

The preparation "Rhoder" consists of two Rhodococcus oil degrading strains and represents a concentrated suspension of living bacterial cells with concentration of 10^9–10^{10} cells/ml that are ready for application without a preliminary activation, Murygina et al. (2000), Mouryguina and Kalyuzhnyi (2001). The working suspension of "Rhoder" for treatment contained 10^6–10^7 oil degrading cells/ml in addition to fertilizer $N_{15}/P_{15}/K_{15}$ (0.2% solution).

The field tests were performed in 2002 on the oil polluted wetland about 2 ha (accidental spills occurred 8–10 years before) in the polar region, in a cold and humid zone of the high northern taiga of the Komi Republic. All plants died at this wetland. The depth of oil contamination (458–738 g/kg dry soil) fluctuated from 15 to 25 cm. The area was divided into nine sites (~2000 m^2 each, size~25×80 m), two sites from those were used as negative controls and seven ones were chosen to test several bioremediation variants. One of these sites was provided for testing the preparation "Rhoder". About 5–7 cm of the upper layer of oil-contaminated peat was cut off previously. Each site was tilled twice by a ponton-stepped-machine and lime was added (400 kg/site) to increase the pH from 6.0 to 7.0. The Moscow State University (MSU) experimental site was treated three times (once every 2 weeks) with the preparation "Rhoder" and the fertilizer by sprinkling, using vacuum pump machinery. Before the second treatment with "Rhoder", a biosorbent (~4 kg on 0.02 ha of soil) was put on some pieces of crude oil, left after site tilling. Before the third treatment with "Rhoder", the Gramineae seeds of Avena sativa and timothy-grass (16 and 5 kg per 0.2 ha, respectively) were sowed by hand, 1 month later than at the other sites. The seeds were preliminarily activated

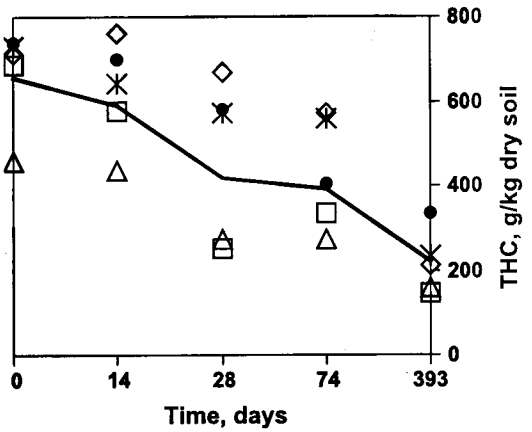

Fig. 1. Decrease of THC content during the bioremediation with the preparation "Rhoder", line—mean values of five samples.

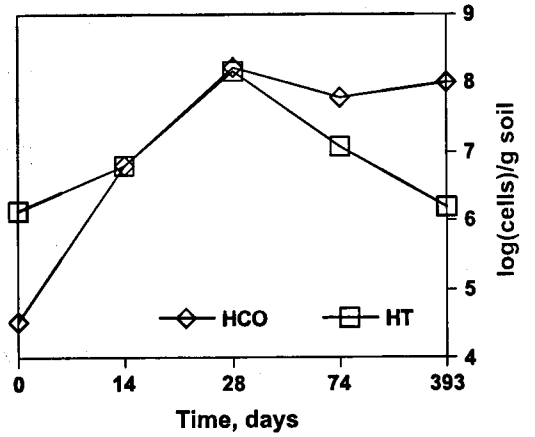

Fig. 2. Microorganism counts during bioremediation with the preparation "Rhoder".

by the anti-stress agent "Symbiont" (4 mL for all seeds). The other six bioremediation variants were performed according to the authors instructions: (1) agricultural engineering method without addition of any preparation ("Komimeliovodhozproekt"); (2) oil-degrading preparation "Universal" (Institution of Biology of RAS); (3) lignosorbent with immobilized oil-degrading preparation "Unirem" (company "Recultivation"); (4) oil-degrading preparation "Petrolan" (Company "Priborservice"); (5) peat-compost preparation "Bamil" (company "Nika"); (6) oil-degrading preparation "Deconta" (company "Deconta", Czech Republic). By the end of June 2002, when comparison tests started, the seeds mentioned above were sowed in all experimental sites, excepting the MSU site. The weather in summer 2002 was cold (+6 to +10 °C), rainy and windy, it was permafrost under a depth of 20–25 cm of oil-contaminated peat. The warm weather lasted only for 10–12 days in the middle of July. Then, it was rainy and cold again. In 2003, dry fertilizers (≈20 kg/site) were spread out without any addition of preparations at the experimental sites.

In 2002, samples from the MSU site were taken for analyses before each treatment as well as 1.5 months after the last application, according to a 5 point scheme (as a rule, one sample is taken from the centre of the site and four others are taken along the edges of the site). The total

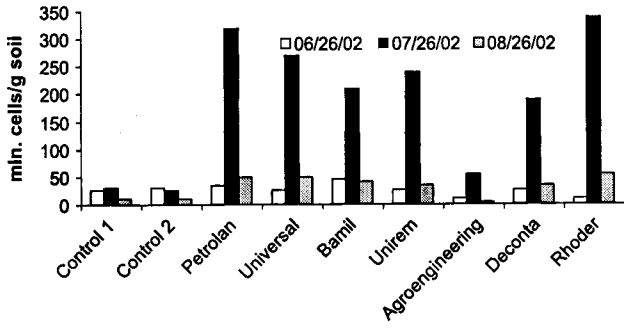

Fig. 3. MPN of nitrifying bacteria from soil of treated sites.

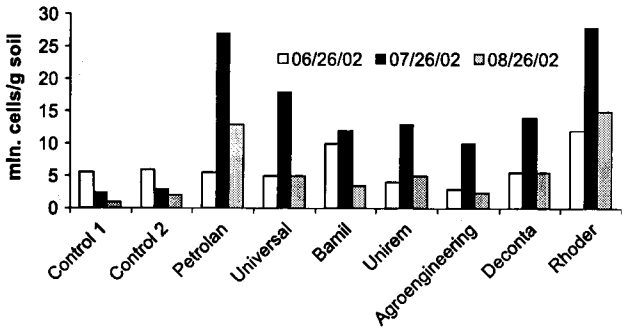

Fig. 4. MPN of ammonifying bacteria from soil of treated sites.

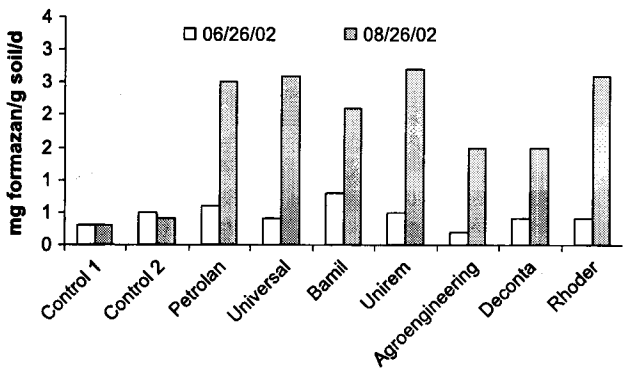

Fig. 6. Dehydrogenase activity of soil from treated sites.

hydrocarbon content (THC) was determined by a gravimetric method. Concentrations of microorganisms in the treated soils were estimated by MPN method using the meat-pepton agar for heterotrophic bacteria (HT) and Raymond's medium with oil for hydrocarbon-oxidizing bacteria (HCO) (Nazina et al., 1988).

The efficiency of each bioremediation variants was evaluated with indexes of bacterial trophic groups, enzymatic soil activity, length of grass and percentage of grass covering each site (Gerhardt, 1984).

3. Results and discussion

The results obtained (Fig. 1) showed that the application of the preparation "Rhoder" decreased the THC by 20–51%, depending on the initial (458–738 g/kg) oil contamination on the site during a short warm period in 2002. The supplementary THC decrease (residual THC content 147–336 g/kg) was observed in 2003 without addition of the "Rhoder" due to phytoremediation and fertilizers supply (Fig. 1). The MPNs of HCO and HT bacteria during the bioremediation in 2002 were rather high and only decreased slightly in 2003 (Fig. 2). By the middle of September 2002, the treated MSU site was covered with 70–85% of green grass. This grass covering increased until 85–95% in August 2003. At the same period, at the sites where the other bioremediation variants were applied, grass covering accounted only for 50–70%, except at the site of the company "Priborservice", where such covering was 90–95%. However, the quantity of seeds was tenfold higher in comparison with the other bioremediation technologies.

The results concerning specific characteristics of soil restoration (indexes of bacterial trophic groups, enzymatic soil activity) showed that the application of the oil-degrading preparations ("Rhoder", "Universal", "Petrolan", "Unirem") was more efficient than the use of preparation "Bamil" developed as an enhancer for agricultural soils or tilling with addition of fertilizers and seeds only (Figs. 3–6), especially when the initial THC was about 740 g/kg dry soil.

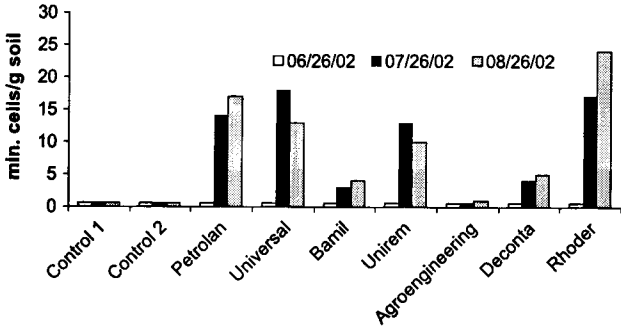

Fig. 5. MPN of HCO bacteria from soil of treated sites.

4. Conclusions

The results obtained during the field tests in the polar region of Komi Republic throughout 2002 showed that the triple application of the preparation "Rhoder" was able to degrade a spilled crude oil by 20–51% (mean value—40%) under the initial THC of 458–738 g/kg dry soil even when the weather was unfavourable for bioremediation. In this case, it was not expected that the bioremediation would be completed during one season. Phytoremediation and introduction of fertilizers during the summer of 2003 further decreased the THC by 9–50% (147–336 g/kg dry soil). It meant that the total decrease of THC accounted for 55–79%, mean value—67% for 2002–2003. Generally, the bioremediation technology with using the preparation "Rhoder", the biosorbent and the anti-stress agent for the seeds showed superior results compared to the other options tested at this spill.

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