III MEĐUNARODNA NAUČNA KONFERENCIJA

"REMEDIJACIJA 2010"

3rd INTERNATIONAL SCIENTIFIC CONFERENCE

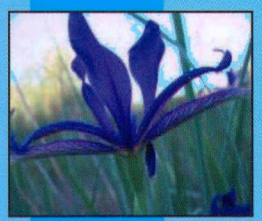
„REMEDIATION 2010"

SAVREMENE TEHNOLOGIJE REMEDIJACIJE ZA OČUVANJE I ZAŠTITU ŽIVOTNE SREDINE

CONTEMPORARY REMEDIATION TECHNOLOGIES FOR ENVIRONMENTAL CONSERVATION AND PROTECTION

ZBORNIK RADOVA

PROCEEDINGS



Privredna komora Srbije, Beograd
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EX SITU BIOREMEDIATION IN MOBILE FACILITY: OUR EXPERIENCE WITH MOBILE PRODUCTION BIOREACTOR FOR MICROBAL BIOMASS AND VARIOUS TYPES AND LEVELS OF POLLUTANTS

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Abstract

One of the consequences of development of industrial civilization is that the quantity of the generated waste exceeds the ability of the nature to clean itself. Therefore, it is necessary to perform the cleaning of polluted areas, to heal them and restore them back to their original condition, before the pollution, i.e. to conduct remediation. Bioremediation is a contemporary method by which the conditions for the growth of microorganisms that are present in an ecosystem are optimized for the purpose of increasing the speed of the biological decomposition of hydrocarbons and other pollutants.

This paper presents a review of three bioremediation procedures that our research team has successfully implemented on the industrial level. By bioremediation we have processed the soil contaminated by crude oil and oil derivatives, then off-balance mazut (masout) and finally, waste emulsions from metal processing industry by applying a multi-stage procedure followed up by bioremediation.

A mobile bioreactor is a new solution in the soil bioremediation technology and water polluted by organic substances, primarily oil and oil derivatives. By applying this solution it is possible to get and apply a large quantity of active microbial biomass that may transform and degrade harmful substances at the pollution site, i.e. at the soil treatment spot.

Key words: *ex situ* bioremediation, mobile bioreactor, pollutants

EX SITU BIOREMEDIJACIJA NA MOBILNOM POSTROJENJU: NAŠA ISKUSTVA SA MOBILNIM PROIZVODNIM BIOREAKTOROM ZA DOBIJANJE MIKROBNE BIOMASE I RAZLIČITIM VRSTAMA I NIVOIMA KONTAMINACIJE

Rezime

Posledica razvoja industrijske civilizacije je i da količina generisanog otpada prevazilazi sposobnost prirode za samoprečišćavanjem. Stoga je neophodno izvršiti čišćenje zagađenih prostora, izlečenje i njihovo vraćanje u stanje pre zagađenja, odnosno remedijaciju. Bioremedijacija je savremena metoda kojom se optimizuju uslovi za rast mikroorganizama prisutnih u ekosistemu da bi se povećala brzina biološke razgradnje ugljovodonika i drugih zagadjivača.

U ovom radu dat je pregled tri bioremedijaciona postupka koje je naša istraživačka grupa uspešno realizovala na industrijskom nivou. Bioremedijacijom su obrađeni zemlja kontaminirana sirovom naftom i naftnim derivatima, potom vanbilansni mazut, a višestepenim postupkom, koji uključuje bioremedijaciju i otpadne emulzije iz metalnoprerađivačke industrije.

Mobilni proizvodni bioreaktor je novo rešenje u tehnologiji bioremedijacije zemljišta i voda zagađenih organskim supstancama, prvenstveno naftom i naftnim derivatima, čijom je primenom moguće dobijanje i primena velike količine aktivne mikrobne biomase, sposobne da transformiše i degradira štetne supstance, na mestu zagađenja tj. na mestu tretmana bioremedijacijom.
Ključne reči: *ex situ* bioremijacija, mobilni bioreaktor, zagađivači

INTRODUCTION

Oil, oil derivatives and products of the petrochemical industry complex are key power sources and raw materials for production, and therefore, they are most commonly used in all fields of life and work.

Soil and water become contaminated by oil and oil derivatives due to incidental spill outs in their exploitation, transportation, processing, storing and utilization. Only 10% of the pollution originates from large incidental spill outs that contaminate sea shores, lakes and water flows and attract significant media attention.

A difficult economic situation in our country, the application of outdated technologies, the bombing campaign which targeted oil refineries, non-existence and non-observance of legal provisions, all have resulted in numerous sites in our country that are contaminated by a wide spectrum of polluting substances, primarily oil derivatives [1].

According to the records kept by the Ministry of Environment and Spatial Planning in Serbia there are about 240 plants that produce hazardous waste and pollute our environment on a daily basis. The most jeopardized are the areas in the vicinity of thermo-electric power plants that have ash dumps, factories producing and processing various metals, chemical industry plants and mines. The level of environmental pollution in Serbia is great, mildly speaking [2].

REMEDIATION

Until recently it was deemed that natural recycling capacities were unlimited. However, the development of industrial civilization has resulted in quantities of the generated waste that exceed the ability of the nature to clean itself. Therefore, it is necessary to perform the cleaning of polluted areas, to heal them and restore them back to their condition before the pollution, i.e. to remedy them.

For these purposes numerous methods have been developed and applied: physical, chemical and physicochemical, by which it is attempted to extract, to exercise control over and to transform pollutants [3, 4].

However, these procedures may resolve the issue only partially.

For the majority of these methods it is typical that they generate, as a result of their application, waste which requires controlled disposal. Today in Serbia there are no industrial waste dumps, so that type of waste ends up in communal waste dumps. In these dumps people often perform the first step of manual recycling. It is not hard to imagine what might happen in a dump where the communal waste, i.e. the household waste is deposited and how short is the route from that dump back to the environment.

BIOREMEDIATION

The nature has its own self-cleaning mechanisms. When petrol hydrocarbons are found free in the environment they become subdued to numerous natural processes: dissolution, vaporization, dispersion, photochemical oxidation, emulsification, adsorp-

tion to suspended particles of materials and sedimentation. The primary mechanism of removal of hazardous substances from the environment is biodegradation of hydrocarbon by the natural population of microorganisms that are present at the contamination site. In that way, oil derivatives are transformed and eliminated from the environment [5].

Hazardous substances are food for microorganisms. Through the action of microorganisms that are naturally present in the soil in a serious of reactions they become substances that do not have toxic effects on humans and on the environment. Some microorganisms degrade a certain number of oil components, but the mixed population-consortium most often provides for a higher degree of degradation. Some components may be decomposed only co-metabolically – through a joint effect of several microorganisms.

Today it is generally accepted that bioremediation is a contemporary method by which the conditions for the growth of microorganisms that are present in an ecosystem are optimized in order to increase the speed of biological degradation of hydrocarbons. [6-8]. Bioremediation may be implemented at the very contamination site (*in situ*) as well as beyond that site when we talk about the *ex situ* bioremediation.

MOBILE PRODUCTION BIOREACTOR

Today, both worldwide and in our country, bioremediation procedures and optimized and improved on several levels:

- Laboratory selection, isolation and construction of new groups of microorganisms that are optimal for individual cases of environmental pollution;
- Development and application of new procedures, such as electro remediation that additionally stimulates bioremediation processes [9];
- Designing mobile production facilities in order to get the biomass of microorganisms directly at the contamination site, i.e. soil treatment.

The mobile production multi-functional bioreactor is a new solution in the bioremediation technology of the soil and waters polluted by organic substances, primarily oil and oil derivatives. This technology optimizes conditions for the growth of microorganisms that are present in an eco system in order to improve the speed of biological decomposition. The mobile bioreactor provides for the preparation of microbial biomass (a "biological agent") at the contamination site, thus improving the efficacy and speed of degradation of polluting substances, reducing the costs of the procedure at the same time.

The uniqueness of the mobile bioreactor lies in the fact that it provides for the production of biomass of microorganisms that are specific for each individual pollution incident, designed for every individual need and customer.

By using a mobile bioreactor this may be done at the very contamination site or at the site of the contaminated soil treatment. The procedure is more effective, faster and cheaper, because there is no transportation or import of microorganisms.

The procedure and steps that ensure successful bioremediation through the utilization of the mobile bioreactor are:

- From the polluted environment the most capable microorganisms that can use pollutant as a carbon and energy source are first isolated and selected.

- Isolated microorganisms are then multiplied in a mobile bioreactor at the contamination site.
- The biomass that is obtained is returned back to the contaminated field, after the additional treatment of its strengthening, and then it performs in a few months or weeks what otherwise would require a few years under natural conditions.

By bioremediation the natural processes of healing and refining the environment are speeded up.

EXAMPLES OF THE SUCCESSFULLY IMPLEMENTED BIOREMEDIATIONS

The processes of bioremediation performed by our research team speak in favour of the successful application of this technology on the industrial level. Via this procedure, very high concentrations of various contaminants may be reduced to the legally acceptable level.

Example No. 1: Bioremediation of the soil contaminated by oil derivatives on Oil Refinery in Pančevo (Serbia) [10-13].

In the circle of the Oil Refinery in Pančevo, on the waterproof layer the bioremediation biopile («an open bioreactor») was made out of soil that was contaminated to various degrees by oil and oil derivatives. The volume of a biopile was 150 m^3. The natural aeration was stimulated by the perforated tube system. Humidity was maintained by the technological water from the tank, having the volume of 1 m^3 (it was also used to add up biomass) - by manual bedewing performed with the pump of up to 100 % water retention capacity. Leach solutions were collected by the drainage system and placed in the second tank, with the automatic control of the level through the pump connected to the water tank, so that the system was completely closed in reference to its impacts on the environment. Homogenization and additional aeration of the biopile contents were ensured by the manual mixing of the whole quantity every two weeks. The biopile was protected against direct impacts coming from the outside by a "polyethylene greenhouse". The experiment lasted 6 months.

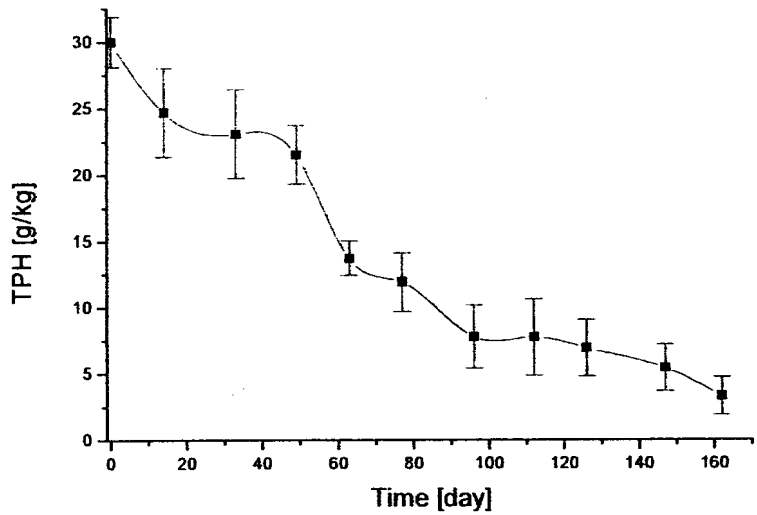

Figure 1. Reduction in the concentration of the total petroleum hydrocarbons in the soil during bioremediation

The change in the composition of the oil pollutant during the course of time is shown in figure 1 and it is the most illustrative example of the successfulness of biodegradation. In the soil contaminated by oil the concentration of the total petroleum

hydrocarbons (TPH) was reduced by bioremediation from the initial 30 g/kg to 3.3 g/kg of the dry substance.

Example No. 2: Multi-stage processing of off-balance emulsions from the metal processing industry followed up by bioremediation for the Lubricant Factory in Kruševac (Serbia) [14].

The metal processing industry creates various types of waste waters as a result of the production, processing and maintenance of metal parts. Oils used in this industry are composed of the mineral base and the appropriate amount of additives that are added in order to improve their characteristics. Very often oils are used in the form of emulsions, stable mixtures of water and oil, with the presence of detergent, biocides and small organic molecules as additives.

These emulsions are used for cooling and lubricating machine parts and cutters and for the removal of metal sawdust from the cutting zone. By being used emulsions loose their desired features due to thermal degradation and contamination and they have to be replaced by the new ones. The obtained waste emulsion contains 2-5 % of oil that is soluble in water.

The good practice applied in the world and in our industry as well implies the return of waste emulsions to the manufacturer who is then obligated to take care of them in accordance with the law.

In this project the waste emulsion from the metal processing industry is processed in a multi-stage procedure (figure 2). Since it originated from various workshops and factories, the emulsion had the varying presence of impurities and levels of degradation. Since the composition of water amounted to 95-99 %, the first goal was to separate the extra water, to purify it and let it into the water flow in accordance with local regulations.

The emulsion was first homogenized and after the adjustment of the pH value, the stability of the emulsion was disturbed by the flocculation and coagulation method. The extraction of the organic non-polar component, mainly of higher hydrocarbons from the mineral oil was then performed. Flocculation and coagulation were achieved by the mixture of non-organic salts, base, amphoteric polyelectrolyte and sedimentation agent.

By this procedure, 99 % polluting substances were extracted from water.

The second stage was the two-step adsorption/filtration for the purpose of separating small polar organic molecules, such as amino and hydroxyl compounds, aliphatic, cyclic and compounds with heteroatoms.

The reduction in the waste water volume was achieved by sand filtration and adsorption, with the utilization of polyfunctional filling. The wastewater processed in such a way was tested by competent institutions and let into the sanitary sewerage system in accordance with the local regulations.

During the processing, the most significant parameter that was monitored in the water was chemical oxygen demand – COD that reveals the presence of hydrocarbons. The initial COD value in the non-treated waste emulsion was about 100000 mg O_2/L. After the sand filter, the COD value was reduced to 5000-10000 mg/L. At the exit of the column with polyfunctional adsorbents COD amounted to about 100 mg/L. The very value confirms the efficacy of the applied procedure.

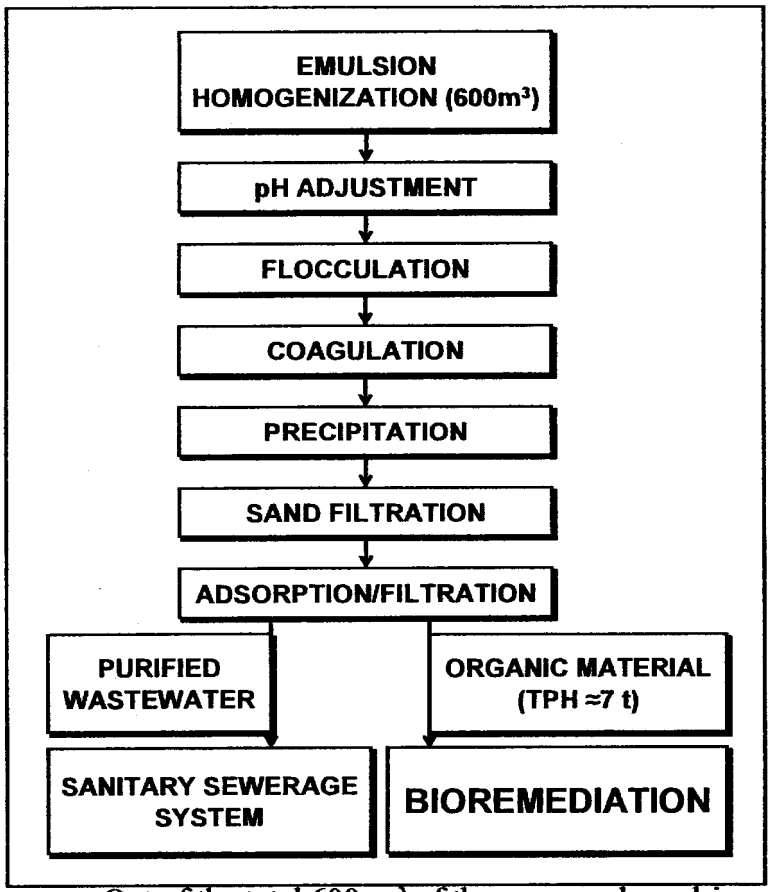

Figure 2. The scheme of the applied procedure for the multi-stage processing of off-balance emulsions

Out of the total 600 m³ of the processed emulsion, we obtained about 60 t of wet material from saturated filters and adsorption units. The quantity of the TPH in that material amounted to about 7150 kg. That material was subdued to bioremediation. It is important to emphasize that the adequate zymogene consortium of microorganisms was used to improve the mineralization process.

In the end of the process, the share of microorganisms that decomposed the petroleum hydrocarbons amounted to 95 % of the total number of microorganisms.

Example No. 3: Bioremediation of off-balance masout for the Belgrade Electric Power Plants-BEPP (Serbia) [15].

"Mazut", (heavy residual oil, heating oil) is used as a power source in heating plants, boiler rooms or for other purposes. Its multi-year storing and usage leads to the creation of reservoir hydrocarbon sludge with the high content of various mechanical impurities and water. The removal of the deposited sludge is performed periodically. The separated sludge is removed till further treatment. For the purpose of BEPP more than 650 t of sludge was processed by bioremediation at the plant for the *ex situ* treatment by bioremediation. The waste hydrocarbon material was mixed with non-purified sand as the bearing material, softwood and sawdust as the alternative source of carbon and the substrate that increased the water holding capacity and looseness of the soil. Natural processes were stimulated by adding the sources of nitrogen, phosphorous and potassium and they were injected by reinoculation every thirty days, by adding a large quantity of microorganisms highly specialised for hydrocarbon substrate. Microorganisms had been previously isolated and selected in the lab conditions from the waste mazut and multiplied in a mobile bioreactor at the implementation site of the *ex situ* bioremediation.

The figure 3 shows comparative gas chromatograms of the TPH samples in the beginning and at the end of the bioremediation process when in 120 days more than 200 t of waste mazut material was processed.

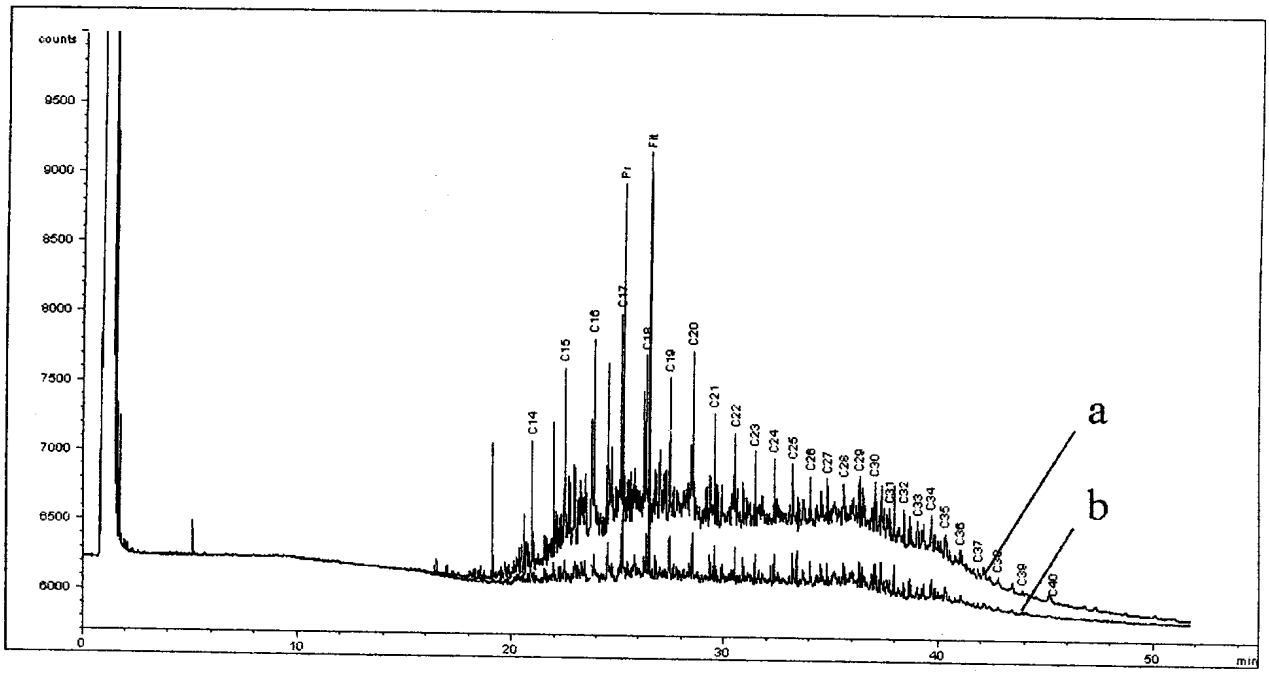

Figure 3. Gas chromatograms of the total petroleum hydrocarbons in the beginning (a) and upon the completion of bioremediation (b).

The world remediation market is worth about € 2 billion! About 20-25 % of all the remediation procedures in the world are performed today by the very bioremediation.

In the first decade of the 21th century, the cost of bioremediation in Europe ranged from 10 to 80 €/m^3, incineration 100-400 €/m^3, solidification 20-180 €/m^3! The advantage of bioremediation is evident as compared to the two most frequently applied technologies, even in regard to the price.

The key to the successful bioremediation is the following:

- isolation, selection and adaptation of zymogene microorganisms and getting an active consortium (*bioaugmentation*);
- achieving as high level of homogenization as possible by permanent mixing, which ensures the required quantity of the molecular oxygen – aeration (*bioventilation*);
- maintenance of the optimal wetness and the addition of biogenic elements in the required quantities, primarily nitrogen and phosphorous (*biostimulation*).

CONCLUSION

The need to heal and clean the environment is great and beyond any doubt. Neglect that lasted for many decades, the non-performance of the required actions and many other reasons have resulted in a large number of locations contaminated by various polluting substances. One of the technologies that marks remarkable success in the world and that is applied in the improvement of pollutions made by oil is bioremediation. Bioremediation enables the reutilization of the decontaminated soil, i.e. its rec-

ycling, thus creating the value. By the bioremediation procedure new waste is not generated that will end up in the communal waste

dump, but the soil and waters, as renewable resources are cleaned and recycled. By using the mobile bioreactor it is possible to get a large quantity of active biomass which may transform and degrade harmful substances, at the pollution site, i.e. at the stop of the oiled soil treatment. Besides that, in the obtained soil, after bioremediation, microorganisms have produced useful substances such as chemical acids and other compounds that will stimulate the increase of microbial diversity in the very soil and the growth of plants in it. Such revitalised soil may even be used to regulate the circle of the factory - pollution generator.

In accordance with the obtained results, it may be concluded that:

- The procedure of the *ex situ* bioremediation of soil contaminated by high concentrations of oil and oil derivatives in the industrial – real circumstances may be efficiently implemented.
- For the successfulness of the procedure, the key step is to prepare the active biomass of microorganisms, which in the described parameters of bioremediation has been proven once again by working on one's own groups of microorganisms that were isolated, selected and adapted from microbiogeocenosis that was the subject to biodegradation of contaminants of the anthropogenous origin.
- Biostimulation and bioventilation are a must and compatible elements of the successful bioremediation.
- Our results also confirm that bioremediation is a bio(geo)technological procedure by which the recycled soil may be obtained. Therefore, one does not get non-hazardous waste that may be discarded to a waste dump, but the soil that has its usable value.

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