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Microbial Degradation of Hydrophobic Compounds under Various Environmental Conditions

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Abstract
Human activities related to industry, agriculture and increasing human population have a great impact on the environment and easily lead to pollution jeopardizing natural habitats and ecosystems. Oil and its derivates from inadequate disposal and accidental spills are among the most common pollutants. Microbial degradation of these hydrophobic, organic compounds is a topic currently generating a great number of studies in environmental protection. The aim of this study is the comparison of two *Rhodococcus* strains isolated from soil and aqueous phase regarding the biodegradation efficacy in various environmental niches: minimal media, salted water and potting soil. Being aware of the changes of enzymes activity affected by various environmental conditions, we will be able to apply the most adequate microbial strains in bioremediation processes.

Introduction
Oil and oil-related pollutants from industrial activities and accidental spills are among the most recent environmental problems that humankind must cope with [1]. Biodegradation of hydrophobic compounds is in the focus of attention [2] and seems to be a very promising process. Applying bioremediation methods, various microorganisms and their microbial pathways can be exploited in environmental protection [3]. *Rhodococci*, which are ubiquitous, aerobic, Gram+ rods, belonging to the *Nocardiaceae* family of the order *Actinomycetales*, have the ability to utilize hydrocarbons as sole carbon and energy source [4].

Experimental
Several bacterial strains were isolated in our laboratory from oil-contaminated soil and even from dead oil. One of them was identified as a *Rhodococcus sp.* MK1. This isolate was compared to a *Rhodococcus erythropolis* PR4 strain (NBRC 100887, NITE) regarding oil degradation efficiency. This strain was isolated from sea water [5] and its whole genome was sequenced.
Since one of the strains was originated in soil and the other one in aqueous phase, the degradation activity might depend on these environmental conditions. The degradation experiments were performed in various niches: minimal medium, artificial sea water and potting soil. Contamination was modelled with hexadecane that was used as a sole carbon and energy source. Respiration and hydrocarbon consumption were monitored, carbon and oxygen balance were determined to compare the bacterial strains to each other concerning the efficiency of oil biodegradation.

Results and discussion

Based on the respiratory activity of bacterial cells, oxygen consumption in aqueous samples inoculated with *Rhodococcus erythropolis* PR4 was higher than those which were inoculated with *Rhodococcus sp*. MK1 (Figure 1.).

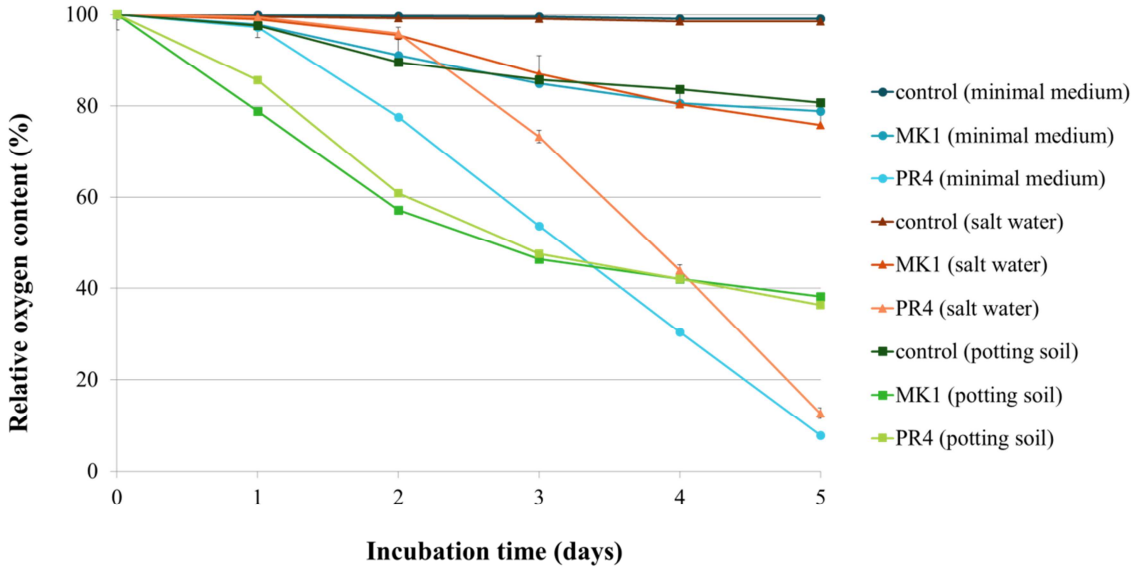

Figure 1. Changes in relative oxygen contents under variuos conditions

Biodegradation of hexadecane was most intensive in potting soil for both strains, but *Rhodococcus erythropolis* PR4 appeared to be more potent in water-based niches. The rate of hydrocarbon biodegradation showed a slight decrease under high salinity conditions compared to the activity in minimal medium (Figure 2).

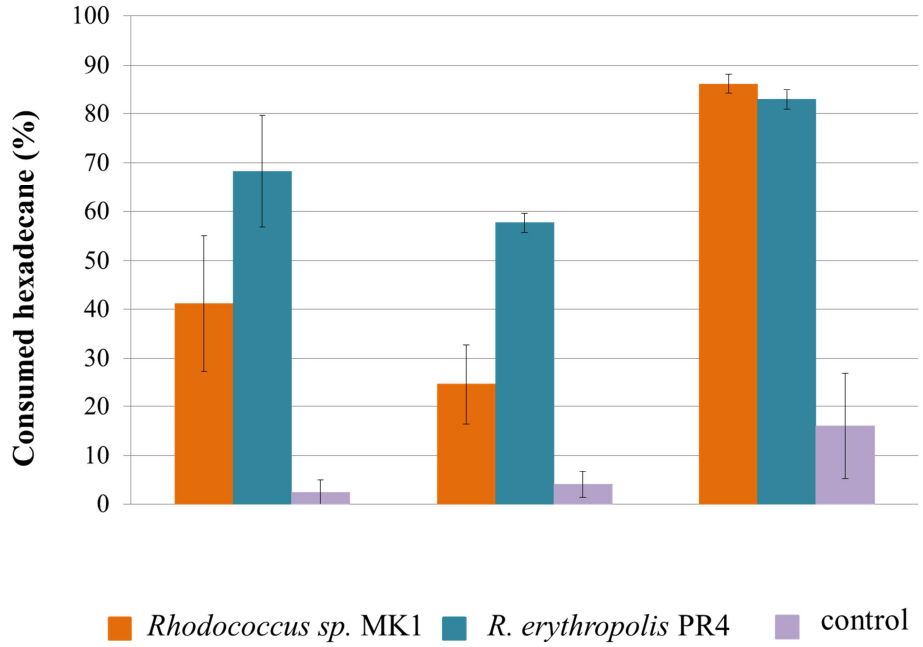

Figure 2. Consumed n-hexadecane in 5 days of incubation

During the hexadecane biodegradation process, hexadecane is converted by monooxygenases into hexadecanol, then into an aldehyde and further into a carboxylic acid. Carbon and oxygen balance can be estimated using the values of oxygen consumption, carbon dioxide production and hexadecane consumption. Based on these data, the results suggested that hexadecane was mainly utilized for biomass production (Figure 3).

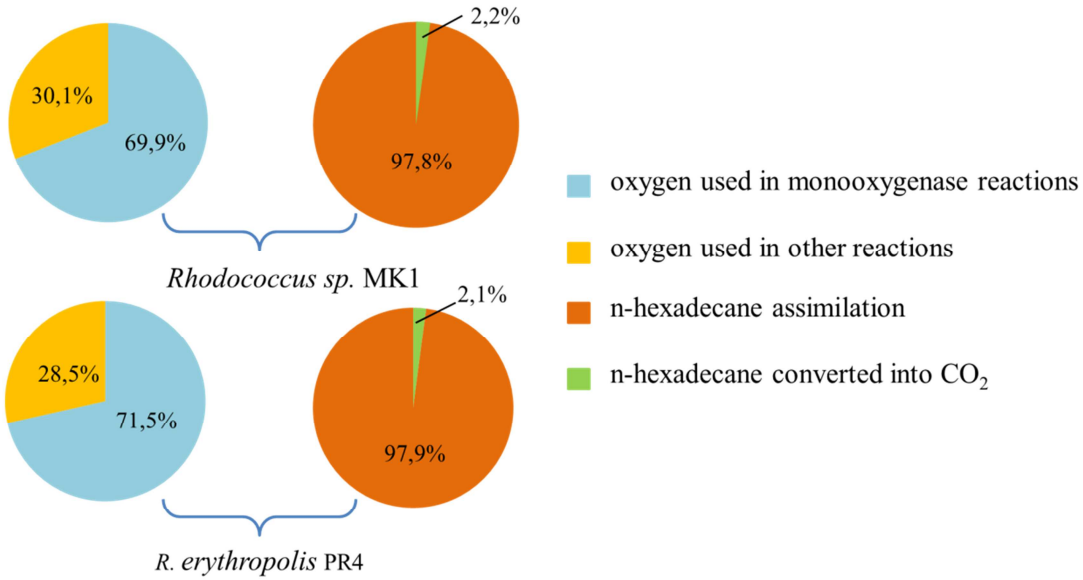

Figure 3. Carbon and oxygen balance of the hexadecane biodegradation in potting soil

Conclusion

Our experiments demonstrate that both of the *Rhodococcus* strains are able to utilize hydrocarbons as sole energy source under various conditions that makes them effective microbial tools for environmental remediation processes like *in situ* bioremediation (even in sea water e.g. the Gulf of Mexico). Better performance in potting soil suggests the presence of biofilm on the surface of the soil particles which might have beneficial effect for bacterial cells.

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