



Hydrocarbon degrading genes monitoring to assess potential of soils to self-restoration after oil pollution

Liliya Biktasheva, Polina Galitskaya, Valentin Fomin, and Svetlana Selivanovskaya

Kazan Federal University, Institute of Environmental Sciences, Department of Applied Ecology, Kazan, Russian Federation
(biktasheval@mail.ru)

Oil pollution of soil and water is one of the most serious environmental problems nowadays. In soils, processes of self-restoration highly depend not only on the level of pollution but also on the characteristics of indigenous microbial community, in particular on the presence of species able to degrade hydrocarbons (whose ability is encoded by corresponding genes) and their dynamics.

In the present study, pollution of three soil types sampled in Tatarstan Republic (Russia) was simulated, and further dynamics of hydrocarbons content as well as microbial community, in terms of hydrocarbon degradation encoding genes, was observed during 4 months laboratory experiment. The three soil types used were eutric podzoluvisols, haplic greyzem and haplic chernozems. Each soil was spiked by three doses of crude oil (6, 12 and 25% w/w). The total hydrocarbon content was estimated using gravimetric and gas chromatography methods. The numbers of 16S rRNA genes as well as bacterial catabolic genes, encoding alkane-monoxygenase related to the genera *Pseudomonas* spp., *Rhodococcus* spp., *Burkholderia* spp. and *Amycolatopsis* spp. (*alkB* and *alkB1* genes), and the *alkB* genes belonging to the species *Pseudomonas putida* and genus *Stenotrophomonas* spp, were estimated using qPCR method. Also we detected genes encoding catechol 2,3-dioxygenase - *xylE* (*Sphingobium*-like), *nahH* (related to the genera *Pseudomonas*, *Sphingomonas* and *Bacillus*) and *nah* gene encoding naphthalene dioxygenase.

It was found, that total hydrocarbon content decreased for all samples for 4-9%, excluding two samples with 25% of oil content and one sample with 12% oil content. The most effective biodegradation was in all soil types with 6% of oil contamination. In all the samples, saturated alkanes were biodegraded most efficient. It was revealed, that in all soil types investigated, independent on the level of contamination or time of sampling, total bacterial numbers were in range from 7.2105 to 6.2106 16S rRNA gene copies g⁻¹ soil. Number of *alkB*, *alkB1*, *xylE* and *nah* genes increased after soil contamination, however it was not the case for *nahH* gene. The abundance of *alkB* and *alkB1* genes related to genera *Pseudomonas* spp., *Rhodococcus* spp., *Burkholderia* spp. and *Amycolatopsis* spp. increased right after contamination in first 3 days from 104 to 105 gene copies g⁻¹ soil. That group of genes is usually more abundant than the other hydrocarbon degrading genes. Abundance of *xylE* gene increased during the first month of experiment from 101 to 104 gene copy number g⁻¹ soil. Number of *nahH* gene was within 10-102 gene copy numbers g⁻¹ soil, *nah* gene and *alkB* genes belonging to the species *Pseudomonas putida* and genus *Stenotrophomonas* spp. were on level of 102 gene copy number g⁻¹ soil.

It can be concluded that the tree soil types investigated had comparable ability to hydrocarbon decomposition in terms of initial hydrocarbon degrading species abundance. Number of hydrocarbon degrading genes increased after soil contamination, while alkane degradation encoding genes numbers raised faster (immediately after contamination) as compared with those of dioxygenase degrading genes.