CHEMICAL ENGINEERING TRANSACTIONS Volume 24, 2011
Editor Sauro Pierucci
 ISBN 978-88-95608-15-0 ISSN 1974-9791
DOI: 10.3303/CET1124174

Kinetic Modeling of Bioremediation Processes Applied to Marine Sediments

Francesca Beolchini*, Laura Rocchetti, Antonio Dell'Anno

Dipartimento di Scienze del Mare, Università Politecnica delle Marche
Via Brecce Bianche, 60131, Ancona, Italy
f.beolchini@univpm.it

The use of bioremediation technologies aimed at the reduction of the contamination associated with marine sediments can be an effective eco-compatible alternative to the more expensive conventional treatments. Although in the literature there are many examples of bioremediation treatments, the innovative contribute of this work is the application of kinetic models to prokaryotic abundances and aliphatic hydrocarbon degradation during the application of clean up biotechnologies. With this aim, bioremediation experiments were performed to reduce hydrocarbon contamination present in harbor sediments, by means of the addition of inorganic nutrients and sand. The results of this study demonstrated that in the presence of inorganic nutrients and sand amendments the highest percentage of aliphatic hydrocarbon biodegradation was observed (>70%). A semi-empirical model was fitted to experimental data, and a yield coefficient of biomass of carbon produced/mass of carbon degraded was calculated, ranging between 0.09 and 0.73. This work highlighted the importance and usefulness of the application of a rather semi-empirical model for the prediction of the residual contaminant concentrations during bio-treatments.

1. Introduction

Bioremediation is a strategy aimed at the reduction of contamination in a matrix, often based on the introduction of inorganic nutrients to enhance bioremediation performance. In fact, for example, it is known that in marine environments biodegradation can be limited by nutrient availability (Leahy and Colwell, 1990).
Kinetic models able to predict the process performances are a fundamental tool for planning strategies of large scale site remediation. Several examples can be found in the literature for the prediction of contaminant residual concentrations during bioremediation (Li et al., 1995; Zhang et al., 1998). Nevertheless, when bioremediation strategies are applied, modeling often regards only contaminant degradation rather than also the prediction of prokaryotic abundances.
In this work, bioremediation experiments were carried out on contaminated sediments. Experimental results were then used to assess the suitability of a rather simple semi-empirical model to predict temporal changes of microbial growth and residual hydrocarbon concentrations during bioremediation experiments in order to provide a support tool when designing bioremediation strategies (Beolchini et al., 2010).

Please cite this article as: Beolchini F., Rocchetti L. and Dell'Anno A., 2011, Kinetic modeling of bioremediation processes applied to marine sediments, Chemical Engineering Transactions, 24, 1039-1044
DOI: 10.3303/CET1124174

2. Materials and Methods

2.1 Sample collection and characterization

The sediment used for bioremediation experiments was sampled from an Italian harbor in the Adriatic Sea. The sand and seawater samples used as amendments were collected from an uncontaminated site of the Adriatic Sea.

Total organic matter was determined by calcination. Carbon content was assumed as the half of total organic matter.

For grain size analysis, sediment samples were wet sieved on a 63 μm sieve.

2.2 Bioremediation experiments

Microcosm experiments were carried out in 250 mL flasks, filled with 20 g of wet harbor sediment and 100 mL of 0.2 μm-filtered seawater. K_2HPO_4, $(NH_4)_2SO_4$ and wet sand (1 g of dry sand for 10 g of dry sediment) were added to the microcosms to evaluate the factors that affected hydrocarbon removal (Table 1; for further details: Beolchini et al, 2010). K_2HPO_4 and $(NH_4)_2SO_4$, in a stoichiometric ratio of C:N:P equal to 100:10:1, were used to supply N and P for stimulating the growth of prokaryotes. Besides inorganic amendments, also the presence of an inoculum of sand was tested to improve material transfer. The flasks were incubated at a temperature of 30 °C in a shaking incubator for 35 days.

Table 1: Experimental conditions investigated in the present study and hydrocarbon degradation percentages at the end of treatments (35 days).

Treatment	Inorganic nutrients	Sand	Total aliphatics	C>12-C24 aliphatics	C>24-C40 aliphatics
control	no	no	19.8%	32.2%	3.1%
nutrients	yes	no	44.0%	47.1%	39.7%
sand	no	yes	38.8%	28.8%	52.4%
nutrients + sand	yes	yes	73.6%	74.0%	73.2%

2.3 Chemical analysis

Aliphatic hydrocarbons were extracted according to EPA 3546 method and quantified according to EPA 8015D method by gas chromatography.

2.4 Determination of total prokaryotic abundance

Total prokaryotic abundance was determined according to standard protocols (Danovaro et al., 2002) by means of epifluorescence microscopy.

2.5 Statistical analysis

Analysis of variance (ANOVA) was carried out on the percentage of hydrocarbon degradation for testing differences among treatments.

2.6 Kinetic modeling

The following semi-empirical equations have been fitted to experimental data describing temporal changes of prokaryotic abundance (X) and total aliphatic hydrocarbons (C):

$$\begin{cases} \dfrac{dX}{dt} = kX(1-\beta X) + K_0 \left| \int_0^t X(t)dt \right| \\ \dfrac{dC}{dt} = \dfrac{1}{Y} kX(1-\beta X) \end{cases} \qquad X(0) = X_0;\ C(0) = C_0 \qquad (1)$$

where a modified version of the logistic equation was used for microbial growth rate dX/dt, adding a term that takes into account the population history (Bailey and Ollis, 1986). The contaminant degradation rate dC/dt was supposed to be associated only with the logistic term of microbial growth through a yield factor, Y. Equations (1) have been fitted to experimental data and the four adjustable parameters k, K0, β and Y have been estimated through a nonlinear regression technique.

3. Results and Discussion

3.1 Sediment characterization

Sediments used for bioremediation experiments had a pelitic fraction of 77%. Total organic matter content was 30±5 mg g^{-1}. Aliphatic hydrocarbons consisted of both the low molecular weight compounds, with C>12-C24 (LMW, 290±10 μg g^{-1}) and the high molecular weight compounds, with C>24-C40 (HMW, 210±10 μg g^{-1}).

3.2 Experimental bioremediation performances

The highest hydrocarbon biodegradation percentage was >70%, in the presence of both inorganic nutrients and sand, as confirmed by analysis of variance (Table 1; Figure 1). The residual concentrations of C>24-C40 hydrocarbons were lower in the experiments containing sand (Figure 2). Nutrients are known to increase hydrocarbon degradation efficiency (Swannell et al., 1996; Xu et al., 2004). Sand had a positive effect on the biodegradation of high molecular weight aliphatic hydrocarbons, more recalcitrant to biodegradation, probably increasing the solid/liquid interface in the sedimentary matrix, enhancing oxygen diffusion and material transfer.

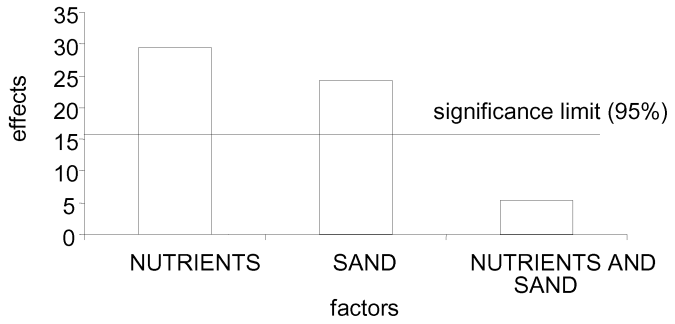

Figure 1: Results of the analysis of variance for aliphatic hydrocarbon degradation.

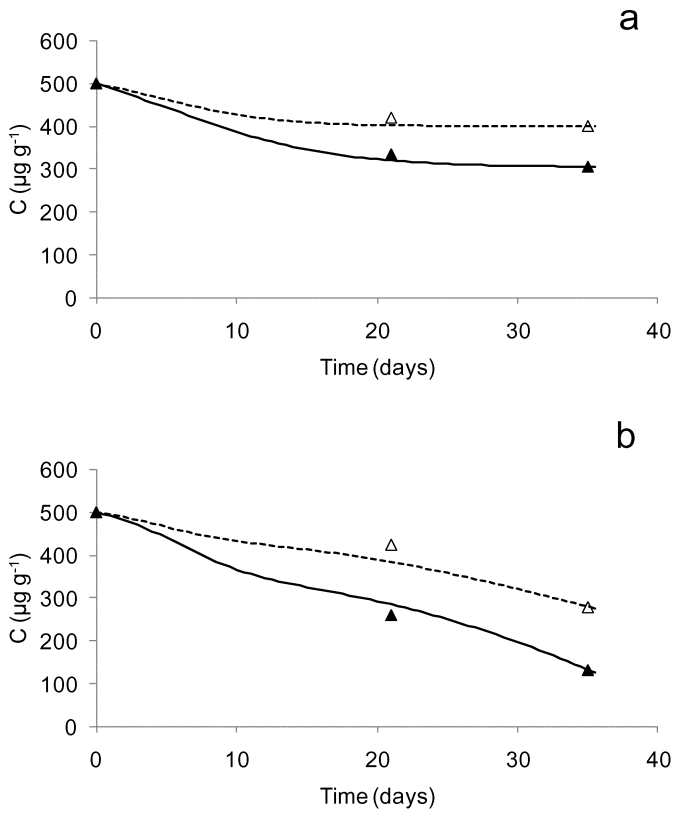

Figure 2. Temporal changes of total aliphatic hydrocarbon concentrations (converted in carbon equivalents) predicted by Equations (1) in the absence (a) and in the presence (b) of nutrient supply (Beolchini et al., 2010). Open triangles represent data obtained from microcosms without sand, and solid triangles from microcosms with sand. Reported are the interpolation lines of experimental data obtained by Equations (1).

The highest microbial abundances were observed in the microcosms containing inorganic nutrients, regardless the presence or absence of sand (Figure 3). In the treatments with inorganic nutrients, the highest prokaryotic abundance was observed on day 14, with $2.6 \pm 0.1 * 10^9$ cells g^{-1}. After this peak, cell abundances decreased up to quite constant values. Lower microbial abundances were observed in the microcosms without nutrient amendment, with a similar trend both in the microcosms with and without sand. Prokaryotic abundance increased from the beginning of the experiments up to day 14 reaching an asymptote, indicating that the nutrients present in the sediment and seawater could allow the existence of a stationary phase and support microbial metabolism.

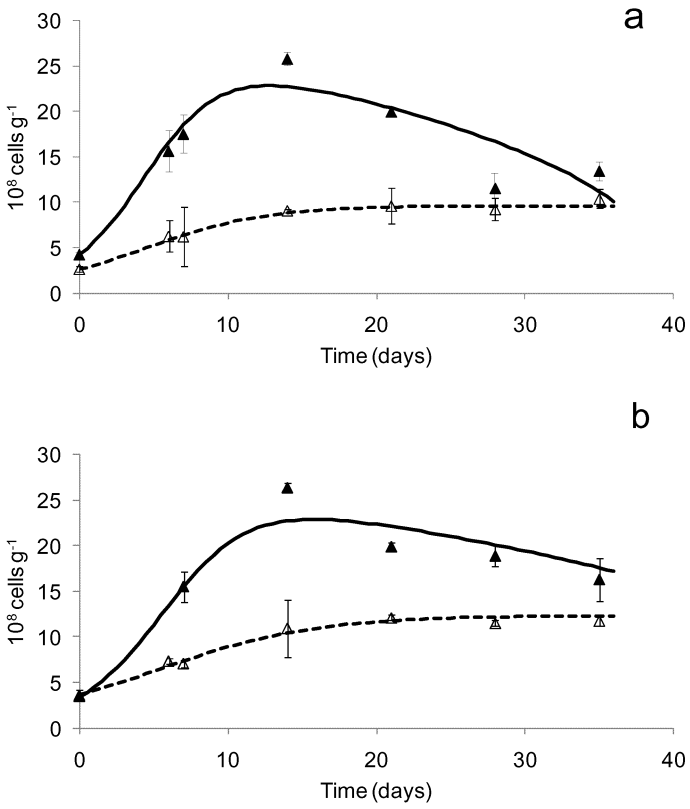

Figure 3: Temporal changes of prokaryotic abundances during bioremediation in the absence (a) and in the presence (b) of sand (Beolchini et al., 2010). Open triangles represent results obtained from microcosms without inorganic nutrients, and solid triangles from microcosms with inorganic nutrients. Reported are the interpolation lines of experimental data obtained by Equations (1).

3.3 Kinetic modeling

Table 2 shows the estimated values for parameters and the coefficient of determination R^2. The relatively high value ($\geq$ 0.9) of such coefficient confirms the suitability of Equations (1) to mathematically describe the bioremediation treatments investigated in the present work. It can be observed that the estimated values for the kinetic constant k increased with the presence of nutrients. K_0 was equal to 0 in the absence of nutrients, where the prokaryotic abundance reached a stationary phase with no decline.

Table 2: Estimated parameters of the models in Equations (1).

Experimental systems	K (d^{-1})	K_0 (d^{-2})	β (g) * (10^8 cells)$^{-1}$	Y (10^8 cells) * (μg hydrocarbons)$^{-1}$	R^2
control	0.24	0.0000	0.10	0.07	0.97
nutrients	0.39	-0.006	0.04	0.31	0.85
sand	0.18	0.0000	0.08	0.04	0.90
nutrients + sand	0.35	-0.004	0.04	0.14	0.90

Equations (1) were used to predict the contaminant temporal changes during all the investigated bioremediation treatments (Figure 2). Without nutrient supply(Figure 2a), the model predicted that hydrocarbon biodegradation took place only for the first 15 days of treatment, reaching a limit value in the range 300-400ppm (20-40% biodegradation), according to the presence of sand. These limit values were achieved when the microbial consortia reached their maximum stationary abundance (Figure 3). On the contrary, in the presence of inorganic nutrients (Figure 2b) the model predicted that hydrocarbon biodegradation progressively decreased during the whole treatment, even when microbial abundances showed a decline (Figure 3).
The results obtained in the experimental conditions investigated in present study offer a methodological approach that can be useful when designing bioremediation strategies.

References

Bailey J.E. and Ollis D.F., 1986, Biochemical Engineering Fundamentals. McGraw-Hill, Singapore.

Beolchini F., Rocchetti L., Regoli F. and Dell'Anno A., 2010, Bioremediation of marine sediments contaminated by hydrocarbons: experimental analysis and kinetic modelling, Journal of Hazardous Materials, 182, 403-407.

Danovaro R., Manini E. and Dell'Anno A., 2002, Higher abundance of bacteria than of viruses in deep mediterranean sediments, Applied and Environmental Microbiology, 68, 1468-1472.

Leahy J.G. and Colwell R.R., 1990, Microbial degradation of hydrocarbons in the environment, Microbiology Reviews, 54, 305-315.

Li K.Y., Zhang Y. and Xu T., 1995, Bioremediation of oil-contaminated soil-a rate model, Waste Management, 15, 335-338.

Swannell R.P.J., Lee K. and McDonagh M., 1996, Field evaluations of marine oil spill bioremediation, Microbiology Reviews, 60, 342-365.

Xu R. Lau N.L.A., Ng K.L. and Obbard J.P., 2004, Application of a slow-release fertilizer for oil bioremediation in beach sediment, Journal of Environmental Quality, 33, 1210-1216.

Zhang W.-X., Bouwer E.J. and Ball W.P., 1998, Bioavailability of hydrophobic organic contaminants: effects and implications of sorption-related mass transfer on bioremediation, Ground Water Monitoring and Remediation, 18, 126-138.