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EXTENDED ABSTRACT

Biodegradation of Benzene and Toluene by Streptomyces Species Isolated From the Soil of Tabriz Refinery, Eastern-Azerbaijan and Investigation of the Kinetic Model

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1. Introduction

Aromatic hydrocarbons are an important group of environmental pollutants that are produced from different sources such as refineries and petrochemical, pharmaceutical, paint, and plastics industries. These environmental pollutants can be removed by different methods such as evaporation, chemical degradation, biological degradation, and adsorption. Biological methods are preferred selective methods because of their simplicity, utilization of environmental-friendly microorganisms, and production of safe materials such as water and carbon dioxide (Singh et al. 2007). Bioremediation is a process for the conversion of toxic materials into non-toxic and safe ones based on the metabolic activity of microorganisms capable of using organic and petroleum pollutants as carbon and energy sources. It is a useful technology with no environmental disruption (Sungpetch 1998). Streptomyces species, which belong to the category of Actinomycetes, are gram-positive and non-moving strains. Most of them produce spores and produce most of the clinically useful antibiotics of natural origin. These species have a different genus, and some of them can degrade aromatic compounds such as benzene, toluene, and even complex petroleum compounds (Claessen et al., 2006).

2. Methodology

2.1. Experimental study

In this work, biodegradation of benzene and toluene by Streptomyces species isolated from petroleum-contaminated soil in Tabriz Refinery was studied. Solutions containing bacterial strains were prepared using a Tryptic Soy Broth culture medium and adding a certain number of strains to it based on half McFarland standard and incubated for about 3 h (Weinstein 2018). In this study, four parameters, including pollutant type, its initial concentration, different Streptomyces species, and incubation time, were examined. Benzene and toluene were prepared separately in a sterilized liquid medium with concentrations of 50 and 100 mg/L and were added into the culture medium containing bacteria. Five different Streptomyces species were tested. The solution samples were incubated in a shaker incubator for 10 days at a temperature of 28 °C and a pH of 7.6. Each day a sample was obtained from tubes containing incubated samples and degradation rates of pollutants in each sample were obtained by double-beam spectrophotometer. The resulting metabolite with the highest

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degradation rate, was analyzed by GC-MS apparatus. It was found that Streptomyces ambofaciens Azar411 species had the maximum biodegradation percentage compared to other strains.

2.2. Modeling

Also, kinetic modeling of the biodegradation process of benzene and toluene was assessed in order to reveal degradation kinetics and to predict process performance at different initial concentrations of pollutants. The following mole balance equation can be written for the degradation of the substrate (benzene or toluene) by bacteria.

$$\frac{dC_S}{dt} = -\left(-r_S\right) \tag{1}$$

Where C_S (mo1L^{-1}) is the concentration of substrate, t is time in days, and $-r_S$ (mo1L^{-1} day^{-1}) is the rate of consumption of substrate by bacteria. Conventional models including first order, Michaelis-Menten Inhibition (MMI), and Michaelis-Menten Activation (MMA) models (Farzi et al. 2019) were utilized as reaction rate equations and their parameters were calculated using linear regression.

3. Results and discussion

Fig. 1 shows results of biodegradation of benzene and toluene by *Streptomyces ambofaciens Azar411* species for 10 days of incubation. As can be seen, more than 70% of benzene and toluene were degraded with an initial pollutant concentration of 50mg/L was degraded by the bacteria. But the degradation efficiency was much lower for higher initial concentrations of substrates which can be due to the inhibition effect of substrates and also the reduction of space for bacteria to degrade the pollutants.

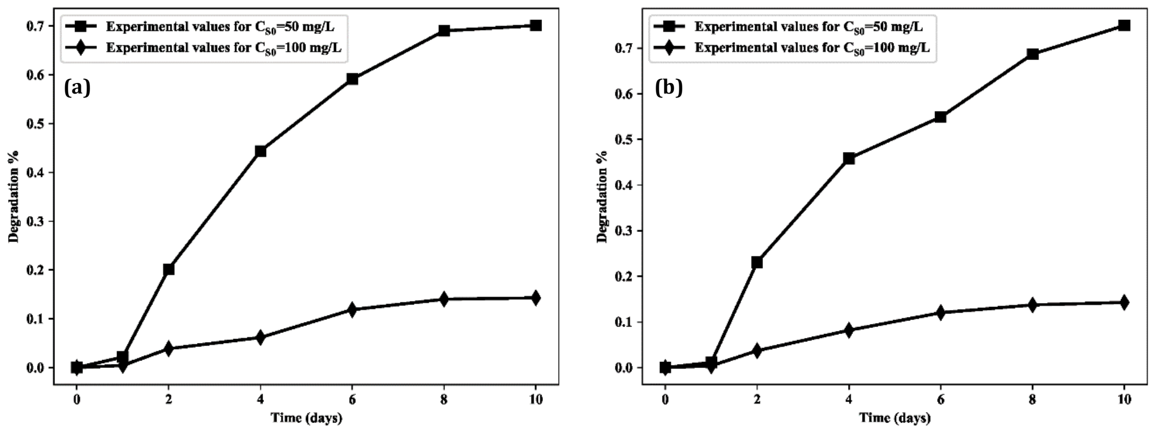

Fig. 1. Biodegradation percent of: a) benzene, b) toluene by Streptomyces ambofaciens Azar411 species with respect to time

GC-MS analysis of the metabolite after incubation revealed that no highly toxic components were produced during biodegradation process. Fig. 2 shows the results of kinetic modeling of the biodegradation process of benzene and toluene using three different models discussed above. As can be seen, Michaelis-Menten Inhibition Model resulted in the best fit on experimental results for both pollutants, which validates the claim about the inhibition effect of toxic pollutants on bacterial species.

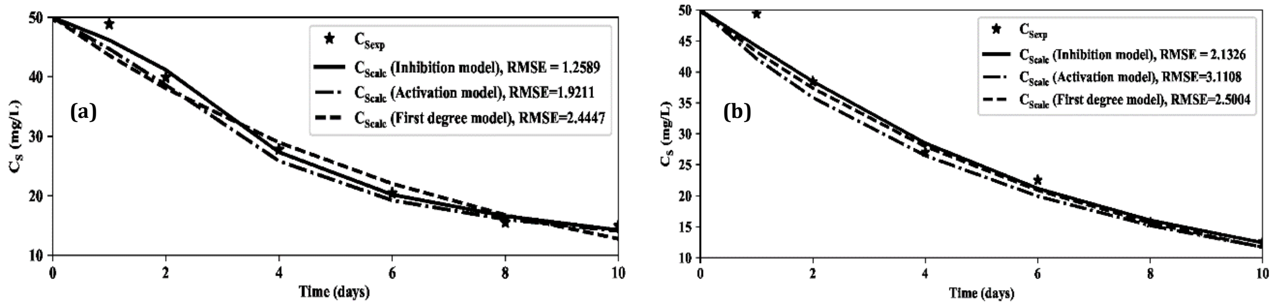

Fig. 2. Kinetic modeling results for biodegradation of: a) benzene, b) toluene by Streptomyces species

4. Conclusions

In this work, the biological degradation of two organic pollutants, including benzene and toluene using different Streptomyces species was investigated. It was observed that, more than 70% of the pollutants were degraded after 10 days of incubation for pollutant initial concentration of 50 mg/L. It was also revealed that increasing the initial concentration of the pollutant sharply decreased degradation efficiency. GS-MS analysis of the most degraded sample showed that no highly toxic components were produced and the process is environmentally safe, though it is slow compared to other methods. Also, for a better understanding of the degradation process, it was modelled kinetically using different conventional models and the results revealed that the Michaelis-Menten Inhibition model best fitted experimental results. Based on the results of this work, it can be concluded that Streptomyces species are a good choice for biodegradation of organic and petroleum pollutants because of their ease of access, production of non-toxic materials, and good performance in harsh environments.

5. References

Claessen D, de Jong W, Dijkhuizen L, Wösten HAB, “Regulation of Streptomyces development: reach for the sky!”, Trends in Microbiology, 2006, 14 (7), 313-319.

Farzi A, Dehnad A, Fotouhi AF, “Biodegradation of polyethylene terephthalate waste using Streptomyces species and kinetic modeling of the process”, Biocatalysis and Agricultural Biotechnology, 2019, 17, 25-31.

Singh A, Van Hamme JD, Ward OP, “Surfactants in microbiology and biotechnology: Part 2. Application aspects”, Biotechnology Advances, 2007, 25 (1), 99-121.

Sungpetch A, “Bioremediation of crude oil and standard hydrocarbons in soil”, PhD Thesis, Faculty of Graduate Studies, Mahidol University, 1998.

Weinstein MP, “Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard”, USA, CLSI, 2018, 11 ed.