P-21

Effect of lignin-degrading enzymes on the decomposition of large aromatic hydrocarbons: Coronene as a surrogate for powdered activated carbon (PAC).

Kojo Twum Konadu [1], Keiko Sasaki [1*], Kwadwo Osseo-Asare [2], Takashi Kaneta [3].

1. Kyushu University, Department of Earth Resources Engineering, Fukuoka, Japan.
2. Penn State University, Department of Materials Science & Engineering and Department of Energy & Mineral Engineering, Pennsylvania, USA.
3. Okayama University, Department of Chemistry, Okayama, Japan.

Abstract

Coronene was decomposed by the cell-free spent medium (CFSM) of *Phanerocheate chrysosporium*, which contained 1.12 mU/mL and 20.61 mU/mL of lignin peroxidase (LiP) and manganese peroxidase (MnP) respectively. The reaction was completed in three days, during which, about 13.6% of the initial 10 mg of coronene had been converted into primarily aliphatic compounds. The changes effected by lignin-degrading enzymes on the chemical bonds in the coronene and PAC residues were similar, which indicated that the coronene can function as surrogate for studying larger hydrocarbons.

1. Introduction

Lignin-degrading enzymes such as lignin peroxidase (LiP), manganese peroxidase (MnP) and laccase, are known to degrade aromatic hydrocarbons by cleaving the C=C and C=O bonds in materials like coal and powdered activated carbon (PAC) which are sometimes used as surrogates for the carbonaceous matter in refractory gold ores [1-3]. However, these substrates have complex molecular sizes and structures, making it difficult to adequately quantify the effects of the enzymes. Accordingly, coronene ($C_{24}H_{12}$) was selected as a surrogate for some larger hydrocarbons because it represents the smallest approximation to graphene sheets [4]. It was anticipated that by using this simpler aromatic carbon, it would be easier to characterize and make inferences about the effect of biodegradation on materials like PAC

2. Experimental

2.1 Fungal growth and enzyme secretion

P. chrysosporium was used for enzyme production in a liquid culture with the same composition as Tien [1]. The fungus was cultured in 500 mL of the medium in a 2000 mL sterilized flasks for 3 days at pH 4.0, 37ºC and no agitation. Following that, the cell-free spent medium (CFSM) was collected by filtration through a 0.2 µm filter and the activities of MnP [5] and LiP [1] were determined before being used for coronene degradation.

2.2 Coronene degradation

20 mL samples of CFSM were contacted with 10 mg of coronene (97%) at initial pH of 4.0 and 30ºC. The mixture was shaken at 120 rpm for 7 days and samples were taken periodically for analysis. The solid residue was washed with ultra-pure water before being dried under vacuum at room temperature. Afterwards, the dried solid was characterized by fluorescence spectrometry, SEM, XRD, FTIR and ^{1}H-NMR.

3. Results and discussion

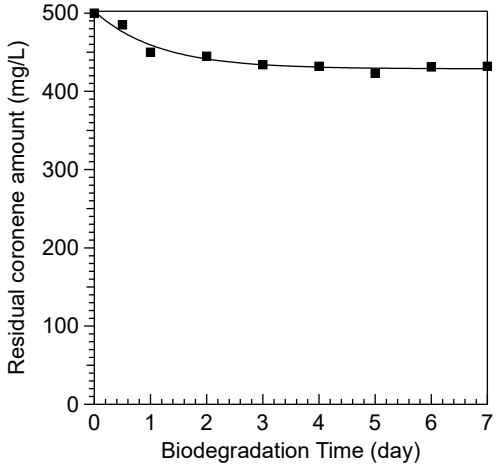

Fig. 1 Changes in residual coronene mass over bio-degradation time. Initial coronene mass 10 mg.

Fluorescence spectrometry was used to quantify the amount of degraded coronene by the CFSM over the 7-day period. The findings in Fig. 1 appear to show that the reaction was completed around the 3rd day, with about 1.36 mg of the coronene being converted into other substances by 22.4 mU of LiP and 414.4 mU of MnP. 3D fluorescence analysis of the solid residue did not show any other compounds mixed with coronene. Thus, indicating that the oxidized products formed by the enzymes were either fluorescent inactive or released into solution. An observed increase in acidity of the residual liquid supernatant illustratesd the formation of some water-soluble compounds.

FTIR and ^{1}H-NMR showed an increase in the aliphatic functional groups C-H, C=C-H and carbonyl C=O relative to the C=C in the solid residue. Comparatively, the coronene residues and PAC residues from our earlier work [3] exhibited similar changes in chemical bonding after CFSM treatment, with the exception of the pH change, indicating that both substances might have followed C=C bond cleavage pathway for aromatic carbon degradation by lignin-degrading enzymes.

The physical structure of PAC and coronene residues were also analyzed, focusing on the pore structure of the solids. N_2 adsorption experiments conducted on the PAC residue showed that the specific surface area was significantly decreased after the treatment. This change was ascribed to the destruction of the microporous structure to form larger pores, however, analysis of the coronene residue by XRD (Fig. 2) showed that the layer structure was progressively compressed, resulting in a decrease in the d-spacing of the slits. This observation illustrates that it is possible that not all the micropores in the PAC residue were enlarged but that some might have become inaccessible through compression.

4. Conclusions

Further tests on the suitability of coronene as a surrogate for PAC will be conducted including Au(CN)2- uptake, but, based on the currently available data, coronene can serve as an easier characterization tool for larger, insoluble aromatic hydrocarbons.

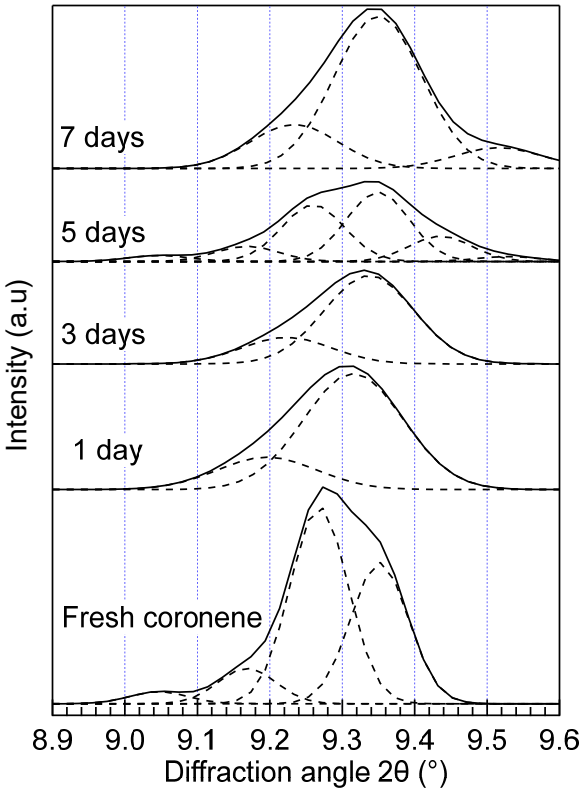

Fig. 2 XRD patterns of the fresh coronene and the bio-treated residues in the regions of 8.9-9.6° for the 001 peak.

Reference

[1] Ofori-Sarpong, G., Tien, M., Osseo-Asare, K. Hydrometallurgy, 102, (2010), 66-72.

[2] Tien, M., Kirk, T. K. Meth. Enzymol. 161, (1988), 238-299.

[3] Konadu, K. T., Sasaki, K., Osseo-Asare, K., Kaneta, T. Hydrometallurgy, 168, (2017), 76–83.

[4] Bartolomei, M., Pirani, F., Marques, J.M.C. J. Phys. Chem. C, 121, (2017), 14330–14338.

[5] Wariishi, H., Valli, K., Gold, M. H. J. Biol. Chem., 267, (1992), 23688-2369.