Peroxidase activity and hydrogen peroxide in phytoremediation of Bisphenol A.

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1. Introduction

Bisphenol A (BPA) is widely used in the plastic industry as a plasticizer or monomer for polycarbonate and epoxy resin, and its contamination in water and soil has been reported. The amounts of wastewater containing BPA have increased with increasing industrial production (Sakurai et al. 2004). BPA has an endocrine disrupting effect, therefore, effective methods for the removal of BPA from wastewater and contaminated sites are required. The objective of this study was to investigate the possible effect of plant peroxidase and hydrogen peroxide (H_2O_2) on the removal of BPA by aquatic plants.

2. Materials and methods

Plants of *Ceratophyllum demersum*, *Limnobium laevigatum* and *Salvinia auriculata* were cultivated in identical glass vessels containing 10% Hoagland's nutrient solution (pH 6.0), where light intensity was kept at 350 µmol photons/m^2/s. These plants were acclimatized to laboratory conditions for more than 20 days, and were used for experiments. These plants were selected because of their wide distribution in aquatic environment. One vessel without plant was prepared and used as a reference. The initial concentration of BPA was set at 100 µg/L. In addition, cell wall-bound peroxidase activities and plant endogenous H_2O_2 were measured and relations to removal rates of BPA were investigated.

Aquatic plants tissue were collected into liquid nitrogen and stored at -20 ºC. The measurements of H_2O_2 concentration and peroxidase activities were made according to former studies (Uchida et al., 2002; Pandolfini and Gabbrielli, 1992).

3. Results and discussion

BPA was effectively removed by every aquatic plant. Figure 1 shows time course changes of BPA by different plants, where *C. demersum* showed highest removal performance. Endogenous levels of H_2O_2 decreased with time. *L. laevigatum* and *C. demersum* showed relatively higher levels of H_2O_2 (Figure 2). In the presence of toxic substance or under stressed conditions, plants may increase the production of reactive oxygen species such as superoxide and H_2O_2, as a result of aerobic metabolism.

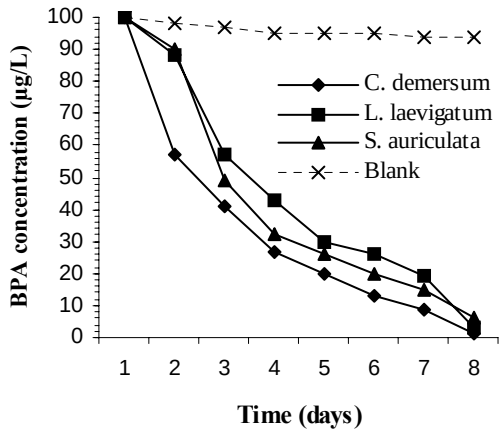

Figure 1. Time course changes of BPA using aquatic plants.

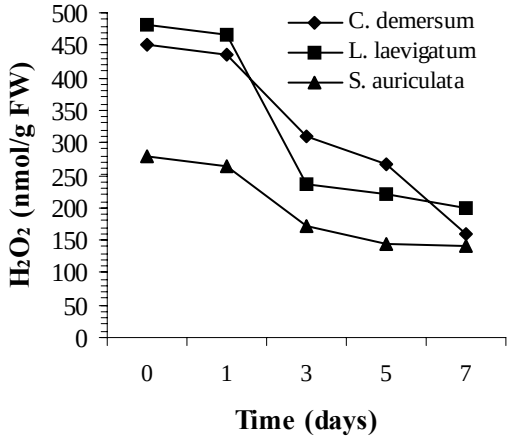

Figure 2. Time course changes of H_2O_2 in aquatic plants.

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It was thought that H_2O_2 might be consumed by a complex enzymatic antioxidant system including catalase and peroxidase.

Figure 3 (A-C) shows time course changes of plant peroxidase activity, demonstrating the enzyme activity differed significantly among the plants. Most peroxidase activities were kept constant or slightly decreased with time. *C. demersum* had relatively larger soluble and ionically cell wall-bound peroxidase activities (IPO). On the other hand, *S. auriculata* increased the activities of IPO and covalently cell wall-bound peroxidase (CPO) during the initial phase of experiment. The increase of peroxidase activity has been observed in many plant cell cultures, which is considered as a metabolic response under various stress conditions.

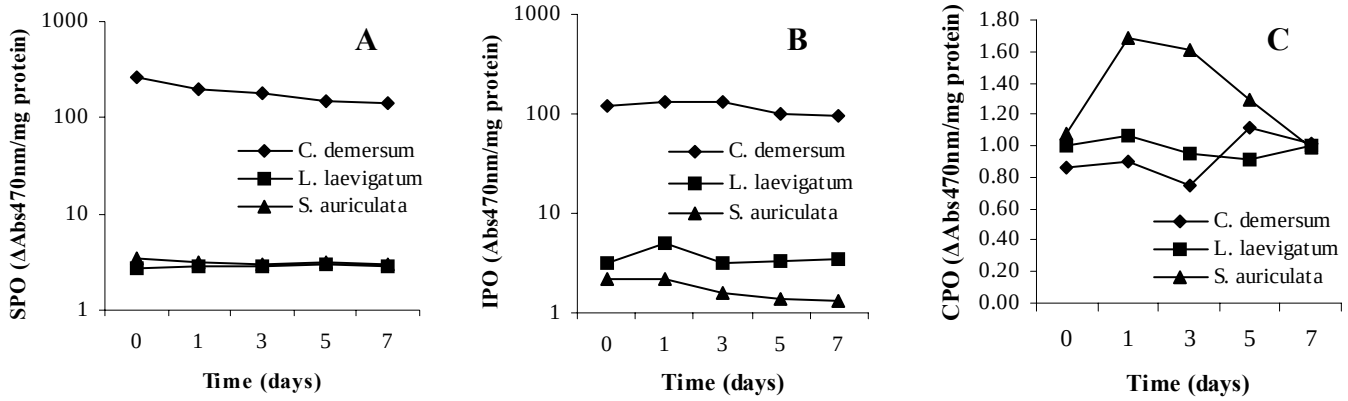

Figure 3. Time course changes of peroxidase activity, where A) Soluble peroxidase activity (SPO); B) Ionically cell wall-bound peroxidase activity (IPO), and C) Covalently cell wall-bound peroxidase activity (CPO), respectively.

Peroxidase enzymes are produced in cells of many microorganisms and plants, and are involved in secondary cell wall formation, lignification, oxidation of a wide range of toxic aromatic compounds including phenols, bisphenols and related heteroaromatic compounds through the reduction of H_2O_2 (Nicell 2003). Hydrogen peroxide may be produced through the photochemical or respiration reaction in plant itself (Bartoli et al. 2004).

Kinetic study will be needed to quantify the relations among the removal rates of BPA, peroxidase activities, and H_2O_2 concentration in aquatic plants.

4. Conclusion

Experimental results demonstrated BPA was effectively removed by different types of aquatic plants. In addition, it was thought the removal rate was affected by endogenous H_2O_2 concentration and peroxidase activity.

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