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# Biodegradability and Risk Assessment of Biomass-based Polymeric Materials

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## Abstract

With the intention to solve environmental problems caused by synthetic plastics from petroleum resources, biodegradable polyurethane foams and thermosetting moldings were prepared from biomass, such as wood and wheat bran by liquefaction method. Biodegradability of these biomass-based polymeric materials was investigated. In activated sludge, polyurethane foams from liquefied wheat bran and thermosetting molding from phenolated wood were decomposed approximately 14% and 29% for 20 days, respectively. One of the wood fungi, *Coriolus versicolor* was able to grow without supplemental nutrition, only with distilled water and polyurethane foam as a nutrition source. Risk assessments were also conducted and results showed that estrogenicity, mutagenicity, and carcinogenicity were not observed in the extractives of biomass-based polymeric materials.

**Key Words:** biodegradability, polyurethane, risk assessment, *C. versicolor*, phenolated wood

## Introduction

A wide variety of plastics that supports our daily life have been synthesized from petroleum resources. These materials were not easily decomposed by microorganisms, causing various environmental problems. Thus, the world has made various countermeasures to protect the environment. For example, dumping plastics into the ocean was prohibited by an international treaty MARPOL 73/78 (Marine Pollution Act 73/78) because marine ecological systems are disrupted by plastics discarded at sea (Aguilar and Borrell 1994).

As another settlement against such environmental problems, biodegradable plastics have been actively developed in recent years. These biodegradable polymers are mainly the aliphatic polyesters produced by microbiological and chemical synthesis, natural polymer-based products, and their blends (Chandra and Rustigi 1998). However, most biodegradable plastics are generally expensive due to the high cost of their ingredients, and thus they are used only for limited purposes.

These circumstances have provided motivation for the authors to develop inexpensive biodegradable plastics by utilizing waste biomass materials, such as by-products in food and wood processed in industries. In this study, polyurethane foams and thermosetting moldings were prepared from waste biomass. The main purpose of the present study is to investigate the biodegradability and to assess the environmental risk of these materials.

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## Materials and Methods

### *Materials*

Wood was of 20 to 80 mesh of birch (*Betula maximomawiczii regel*) and wheat bran was used. They were dried in an oven at 105°C for 24 h and then kept in a desiccator at room temperature before use. Wood flour used as filler for molding was of 200 mesh pass size. All other chemicals used were obtained from commercial sources.

### *Methods*

#### Preparation of biomass-based polymeric materials

Polyurethane foams from wood and wheat bran were prepared by the method we have previously reported (Lee et al. 2000a) with modifications. Briefly, the liquefaction of biomass is conducted by the mixture of polyethylene glycol and glycerol in the presence of sulfuric acid (1 wt% based on the solvent weight) at 150°C for 60 min. The ratio of polyethylene glycol and glycerol was set to be 4. Biomass solid content in solvent was 33 wt%. It was not necessary to pretreat biomass materials except for drying. The obtained liquefied products were directly used to prepare polyurethane foam by reaction with diphenylmethane diisocyanate in the presence of catalyst, silicone surfactant, and water. Thermosetting moldings were prepared by hot-pressing the mixture of phenolated wood (37.7%), wood flour filler (49.5%), hexamine (9.4%), Ca(OH)2 (2.4%), and Zn stearate (1%) (Lee et al. 2000b).

#### Degradation test by activated sludge

Aerobic biodegradability by activated sludge was tested by the method of Japanese Industrial Standard (JIS) K6950 (Yagi 1988). The samples (9 mg) and standard activated sludge (30 mg) were added into the MITI culture solution (300 ml). As the blank, only standard activated sludge (30 mg) was added. These cultures were incubated at 25±1°C and the value of BOD was measured by BOD meter.

From the value of BOD, biodegradability of the sample was calculated by following equation,

$$D_B\,(\%) = \{S - B\}/ThBOD \times 100 \quad \cdots\cdots (1)$$

where DB is the degree of biodegradability (%); S the BOD value (mg) of the culture solution of sample; B the BOD value (mg) of the blank culture, and ThBOD the theoretical BOD value for oxidation of the sample from the composition of organic atoms, which was analyzed by Perkin Elmer 2400.

#### Degradation test by wood rot fungi

The degradation tests by wood rot fungi (*C. versicolor*) were conducted as based on JIS A-9032. Wood rot fungi was inoculated in Potato Dextrose Agar (PDA) medium and the sample of polyurethane foam (20x20x10 cm) was placed on PDA medium. The culture was carried out at 28°C for 2 months. The culture of wood rot fungi in the absence of the PDA medium was also conducted. For the test, polyurethane foam was powdered by freezing crushing under liquid nitrogen with Cryogenic Sample Crusher JFC-300 (Japan Analytical Industry Co., Ltd.). The sample powder (1 g) was put into an Erlenmeyer flask (300 ml) and wood rot fungi was inoculated on to it. Distilled water was sprayed on the sample powder to moisten and it was incubated at 28°C for 2 months.

#### Extraction

The powder (0.2 g) from the polyurethane foam and the molding was extracted with 5 mL of distilled water at 100°C for 15 min and at 65, 40, and 28°C each for 24 h. Extraction by ethanol was conducted at 83°C for 30 min. The extracts were freeze-dried, dissolved with 20 ml of $H_2O$ or 10% dimethylsulfoxide, and diluted at various concentrations for the assays.

#### Risk assessments

The carcinogenicity and mutagenicity of the extracts were assayed by umu-test (Oda et al. 1985) using a commercial kit (umu-lac, Japan Immunoresearch Laboratories, Co., Ltd., Takasaki, Gunma, Japan), which included the bacterial strain (S. *typhimurium* TA 1535/pSK1002) and culture solution. The assay for estrogen receptor binding capacity of the extracts was performed by the method of Tamaya et al. (1984) using rabbit endometria. [2,4,6,7-3H(N)]-estradiol.

## Results

### *Biomass-based polymeric materials*

Polyurethane foams and moldings were successfully ob-

tained from liquefied wood and wheat bran. In polyhydric alcohol liquefaction, biomass is decomposed by heterolitic sovolysis and the decomposed components react with polyhydric alcohols or self-condense (Yu and Lee 2014). The phenolated wood for moldings can be obtained by the phenolysis of wood and the phenolation and condensation of its decomposed components, such as cellulose and lignin (Lin et al. 1997) Therefore, the liquefied components of biomass do not play a role as the filler, but react chemically in the polyurethane foams and the moldings. Formaldehyde was not used in preparing the moldings.

Table 1 shows the properties of biomass-based polymeric materials prepared in this study. They have comparable properties to those prepared from synthetic resins.

**Table 1.** Properties of biomass-based polymeric materials

| Sample | Density (g/cm$^3$) | Elastic modulus (MPa) |
|---|---|---|
| Polyurethan foams from liquefied wood and wheat bran | 0.028～0.045 | 1.5～10.1 |

| Sample | Flexural strength (MPa) | Flexural Modulus (MPa) | Flexural Toughness (MPa) |
|---|---|---|---|
| Moldings from phenolated wood | 81 | 8,590 | 32 |
| Molding from commercial resin | 86 | 9,005 | 37 |

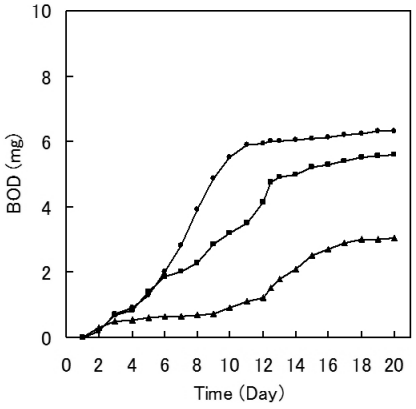



**Fig. 1.** BOD values of polyurethane foam and molding from biomass by activated sludge. ● Molding from phenolated wood, ■ Polyurethane foam from liquefied wheat bran, ▲ Blank.

### *Biodegradation by activated sludge*

Fig. 1 shows the variation of the BOD value with time. The BOD values of the samples were larger than the value of blank, indicating that the decomposition of the sample was advanced. The degree of the biodegradability converted from BOD values is shown in Fig. 2. In the case of polyurethane foam from liquefied wheat bran, the degree of biodegradability increased after 1 day and reached about 13% for 10 days. After that it gradually increased. On the other hand, the degree of biodegradability of the molding sample increased to 28% for 11 days. These results proved that biomass-based polymeric materials were decomposed by the microorganisms of activated sludge.

### *Biodegradation by wood rot fungi*

Fig. 3 shows the degradation of the polyurethane foam

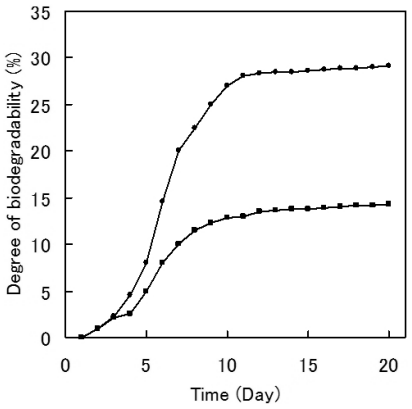
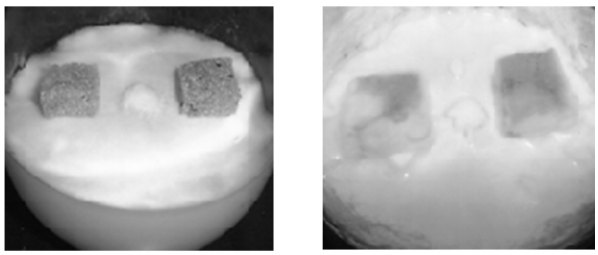



**Fig. 2.** Degree of biodegradability of polyurethane foam and molding from biomass by activated sludge. ●Molding from phenolated wood, ■ Polyurethane foam from liquefied wheat bran.



(Control) (After 2 months)

**Fig. 3.** A wood rot fungus, *Coriolus versicolor* IFO 30388, grew in polyurethane foam in the presence of PDA medium.

by *Coriolus versicolor*, a wood rot fungi, in the presence of PDA medium. At the beginning, the weight loss of the sample would have been measured, but because fungus propagates in the cell of the foam, it was not possible to accurately measure it. As shown in Fig. 3, the sample was fully covered by fungus, indicating that fungus was actively propagated. Fig. 4 shows the hypha of fungus propagated in the cell of polyurethane foam. However, under the condition with PDA medium, which includes rich organic nutrients, it is difficult to determine accurately whether fungus decomposes the sample or not. Therefore, the experiment, which uses only the foam sample as a carbon resource, was carried out. Fig. 5 shows the appearance of the samples after culturing for 2 months. The growth of the fungus was confirmed, indicating that fungus uses foam as a nutrition source.

### *Risk assessments*

Early warning systems of environmental hazards have become more important in recent years. Concern has been growing about potential adverse effects of genotoxins on public health. The umu-test is a sensitive and simple system for the screening of mutagenic and carcinogenic activities from environmental samples, and its sensitivity to toxic xenobiotics is equal to or higher than that of the Ames test (Shinagawa et al. 1983; Oda et al. 1985). Table 2 shows the results of umu-test in the extracts of polyurethane foams and moldings. Bisphenol A and furylfuramid were used as a positive control in both the presence and/or absence of the S9 fraction, and 2-aminoanthracene was used as a positive control in the presence of S9 mix. All extracts of the samples showed negative values in both the presence and absence of the S9 fraction, indicating that there were no mutagens or carcinogens in the extracts of the samples.

To investigate whether endocrine disrupting activity is derived from the samples, we assessed the estrogenicity of the extracts from them by the inhibition of binding between 3H-labeled estradiol and estrogen receptors (Korach et al. 1987). Bisphenol A and diethylstilbestrol were used as a positive control and clearly inhibited the binding of estrogen to its receptors. However, little inhibition activity was detected from the extracts of the samples.

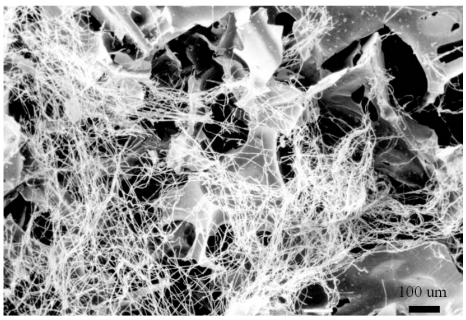
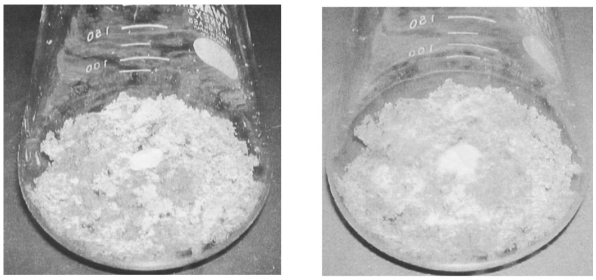



**Fig. 4.** SEM micrograph of the hypha of wood rot fungus inside polyurethane foam.



(Control) (After 2 months)

**Fig. 5.** A wood rot fungus, *Coriolus versicolor* IFO 30388, grew in polyurethane foam in the absence of medium.

**Table 2.** Detection of mutagenicity and carcinogenicity by umu-test

| Samples | Extractive solvent | Temperature (°C) | Without S9 mix | With S9 mix |
|---|---|---|---|---|
| Polyurethane foam from Wood | Water | 28 | - | - |
| | | 40 | - | - |
| | | 65 | - | - |
| | | 100 | - | - |
| | Ethanol | 85 | - | - |
| Polyurethane foam from wheat bran | Water | 28 | - | - |
| | | 40 | - | - |
| | | 65 | - | - |
| | | 100 | - | - |
| | Ethanol | 85 | - | - |
| The molding from phenolated wood | Water | 28 | - | - |
| | | 40 | - | - |
| | | 65 | - | - |
| | | 100 | - | - |
| | Ethanol | 85 | - | - |
| Bisphenol A | (Control) | | + | + | + |
| Furylfuramid | (Control) | | + | + | + |
| 2-aminoanthracene | (Control) | | - | - | + |

**Table 3.** Inhibition of estrogen binding with estrogen receptor

| Samples | Extractive solvent | Temperature (°C) | Concentration of [$^3$H] labeled estrogen | | |
|---|---|---|---|---|---|
| | | | 0.1 nM | 1.0 nM | 5.0 nM |
| | | | Inhibition activity (%) | | |
| Polyurethane foam from wood | Water | 28 | 0 | 0 | 0 |
| | | 40 | 0 | 0 | 0 |
| | | 65 | 0 | 0 | 0 |
| | | 100 | 0 | 0 | 0 |
| | Ethanol | 85 | 0 | 0 | 0 |
| Polyurethane foam from wheat bran | Water | 28 | 0 | 1.3 | 0 |
| | | 40 | 0 | 0 | 0 |
| | | 65 | 0 | 0 | 0 |
| | | 100 | 0 | 0 | 0 |
| | Ethanol | 85 | 0 | 0.2 | 0 |
| The molding from phenolated wood | Water | 28 | 0 | 0 | 0 |
| | | 40 | 0 | 0 | 0 |
| | | 65 | 0 | 0 | 0 |
| | | 100 | 0 | 0 | 0 |
| | Ethanol | 85 | 0 | 0 | 0 |
| 25 nM Bisphenol A | Control | | 90.1 | 79.0 | 60.8 |
| DES (200-fold excess molar) | Control | | 100 | 100 | 100 |

## Discussion

Polymeric materials obtained in this study were degraded in activated sludge (Fig. 2). Furthermore, *C. versicolor* was able to grow with polyurethane foam as its sole nutrition source (Fig. 5). It has been found that polyurethane foams prepared from liquefied cellulose, bark and starch can be degraded in soil (Hatakeyama 1996; Ge et al. 2000; Lee et al. 2002). It has been also reported that synthetic polyester-type polyurethane foams are decomposed by a number of fungi and bacteria (Daby and Kaplan 1968; Crabbe et al. 1994; Nakajima-Kambe et al. 1995), and this degradation is generally initiated by hydrolysis of the ester bond with hydrolytic enzymes such as esterase. Many groups reported the purification and characterization of those enzymes (Pathirana and Seal 1984; Kay et al. 1991). However, it is known that polyether-type polyurethane foam is difficult to degrade. Even if polyurethane foams obtained in this study are polyether-type, they degrade in activated sludge. This may be mainly attributed to the degradation of the liquefied component of the biomass.

Phenolated wood has a chemical structure in which the phenol is combined to wood constituent decomposed by phenolysis. These wood components can be also biodegraded. To clarify the mechanisms of degradation of liquefied biomass, further investigation and characterization are required.

It is well known that bisphenol A, a raw material used in epoxy resin and polycarbonate resin, is an endocrine disrupting chemical. Additionally, other materials of synthetic resins such as phthalates and nonylphenols are also endocrine disrupting chemicals (Colborn et al. 1993; Steinmez et al. 1997), which are causing various environmental problems. However, estrogenicity, mutagenicity, and carcinogenicity were not detected from the extracts of the polyurethane foams and the moldings obtained in this study (Tables 2 and 3). Although further chemical analysis is needed, these materials can be regarded as environmentally safe.

## Acknowledgement

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