Efficiency of Rice Bran for Removal of *p*-Dichlorobenzene from Water

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Rice bran was found to effectively adsorb *p*-dichlorobenzene. The amount of *p*-dichlorobenzene adsorbed was plotted against the equilibrium concentration of substsnces in solution on a logarithmic scale, and a linear relationship was obtained, indicating that the adsorption reaction was Freundlich-type. Adsorption of *p*-dichlorobenzene by rice bran was observed in the pH range 1–12. The removal of *p*-dichlorobenzene by rice bran was attributed to the uptake by intracellular particles called spherosomes.

Key words —— *p*-dichlorobenzene, spherosome, rice bran, Freundlich isotherm

INTRODUCTION

The dichlorobenzenes have been used for a number of years as insecticides. The *para* compound is employed on a very large scale against the clothes moth and against insects infesting hides, furs and museum speciments.[1)] Most of the toxicological information available relates to the *para* isomer.[2)]

In Japanese tap water, concentration of 5.8, 4.5 and 0.5 ng/l have been reported for the *para, ortho*, and *meta* isomers, respectively.[3)]

The industrial output of *p*-dichlorobenzene in Japan has increased in the past decade from 8800 tons in 1963 to 33000 tons in 2001.[4)] To protect water sources, it is important to keep the concentrations of this compound in ground water as low as possible.

The object of this work was to investigate several adsorbents for the effective removal of *p*-dichlorobenzene from water.

MATERIALS AND METHODS

Apparatus —— The assay of *p*-dichlorobenzene was performed on a Shimadzu (Kyoto, Japan) Model GC-14B gas chromatograph equipped with a flame ionization detector and a capillary column (ULBON (Shinwa Chemical lndustries, LTD., Kyoto, Japan) HR-52, 30 m × 0.53 mm). The column was maintained at 150°C, with both the injection port and detector were maintained at 200°C.

Materials —— Rice bran was purchased at a local market. The composition of rice bran is shown in Table 1.

Standards of *p*-dichlorobenzene were purchased from Wako Pure Chemical Industries Ltd. (Amagasaki, Japan). Wastewater samples were taken from a chamber storing wastewater collected from chemical laboratories (Kobe Pharmaceutical University).

Adsorption Experiment —— A 100 ml of sample solutions containing 5 mg/l of *p*-dichlorobenzene and rice bran (0.05–1.0 g) were placed into 100 ml glass stoppered Erlenmeyer flasks and mixed with a stirrer. The reaction mixture was filtered through filter paper (quantitative ashless No.5A Toyo Roshi, Ltd., Tokyo, Japan) to remove the rice bran. The initial 10 ml of filtrate was discarded because of the adsorption of *p*-dichlorobenzene by the filter paper. In control samples without rice bran, the subsequent filtrate after the discarded portion contained the same amount of adsorbent as the original solution. Fifty ml of this filtrate was placed in a separatory funnel and 5 ml of m-xylene was added to the solution. The mixture was shaken for 1 min and the

Table 1. Composition of Rice Bran and Spherosomes

Constituent	Concentration (g/100 g)	
	Rice bran	Spherosomes
Water	13.5	9.8
Protein	13.2	26.6
Lipid	18.3	3.9
Carbohydrate		
glucide	38.3	38.4
fiber	7.8	3.6
Ash	8.9	17.4

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separated m-xylene layer was subjected to gas chromatography (GC) to analyze *p*-dichlorobenzene. To assess the evaporation loss of *p*-dichlorobenzene, control experiments without rice bran were performed as above. Maximum loss was about 5% (4.7 ± 0.22%), although negligible loss was detected in most cases. The removal efficiency of rice bran was calculated after taking into account the evaporation loss of *p*-dichlorobenzene. Values are shown as means ± S.D.

Isolation of Spherosomes —— Spherosomes were isolated using an improved method based on that of Moreau *et al*.[5] Samples of 1 g (dry weight basis) of rice bran were ground in 40 ml of grinding medium consisting of 20 mM sodium succinate, pH 5.6, containing 10 mM $CaCl_2$ with a mortar and pestle. The paste was filtered through four layers of cheesecloth, and the filtrate centrifuged at 30000 × *g* for 20 min. The spherosome pad was removed from the surface with a spatula and washed by resuspending in 40 ml of fresh medium. This suspension was recentrifuged at 30000 × *g* for 20 min. This process was repeated two more times, and the final pellet was used as the spherosome fraction. The composition of spherosome is shown in Table 1. Moisture content was determined by drying a sample for 6 hr at 110°C. Protein concentration was determined by the method of Kjeldahl.[6] Lipids were extracted by the Bligh and Dyer method.[7] The mass of the total lipid was determined by drying an aliquot of chloroform extract in a vacuum oven overnight and weighing the resulting lipid residue. Carbohydrate (glucide) was determined by Anthrone method.[8] Dietary fiber was determined by Association of Official Analytical Chemists (AOAC) method.[9]

RESULTS AND DISCUSSION

Adsorption Rate

Table 2 shows efficiency of rice bran for removal of *p*-dichlorobenzene at reaction time 90 min, because the removal efficiency became constant after 60 min treatment. The average removal efficiency for *p*-dichlorobenzene was 86.1%.

Adsorption Isotherm

The amount of *p*-dichlorobenzene adsorbed at equilibrium was plotted against the concentration of *p*-dichlorobenzene in solution on a logarithmic scale. Equilibrium was measured after at least three hours of contact. A linear relationship was obtained, indicating that the adsorption reaction was of a Freundlich type (Fig. 1).

Table 2. Removal Efficiency of Rice Bran for *p*-Dichlorobenzene

Substance	Concentration (mg/l)		Removal efficiency (%)
	Before treatment	After treatment	
p-Dichlorobenzene	5	0.6–0.8	86.1 ± 1.6[a]

a) Data represent the mean ± S.D. of three separate determinations. Rice bran, 10 g/l; Reaction time, 90 min.

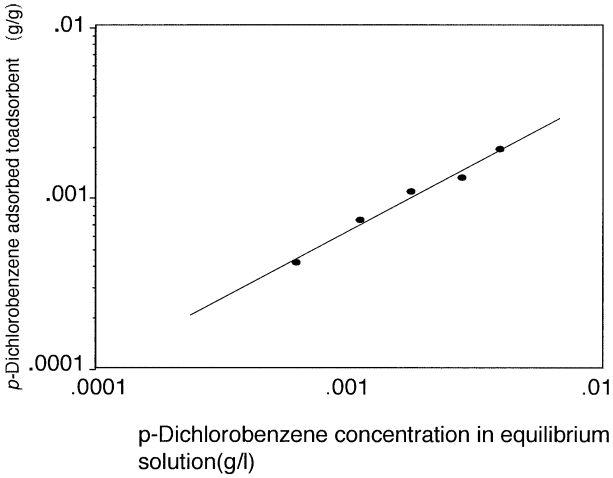

Fig. 1. Freundlich's Adsorption Isotherm of *p*-Dichlorobenzene

Data represent the mean ± S.D. of three separate determinations. Reaction time: 3 hr, *p*-Dichlorobenzene: 5 mg/l, pH: 7.

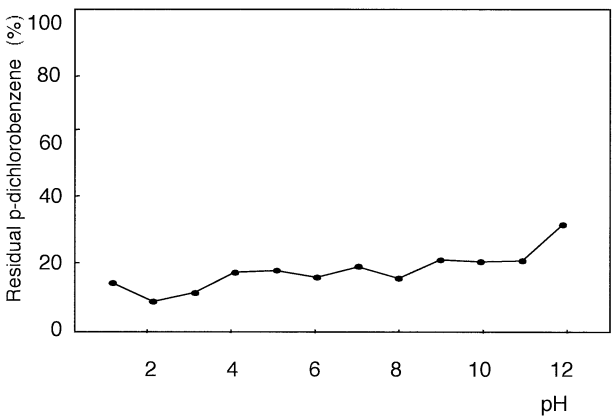

Fig. 2. Effect of pH on the Adsorption of *p*-Dichlorobenzene

Data represent the mean ± S.D. of three separate determinations. Rice bran: 10 g/l, *p*-Dichlorobenzene: 5 mg/l. Each solution of HCl, citric acid-phosphate buffer, and carbonate buffer was used for the preparation of pH 1–2, pH 3–7 and pH 8–12 solutions, respectively.

Effect of pH on Adsorption

Figure 2 shows the effect of pH on the ad-

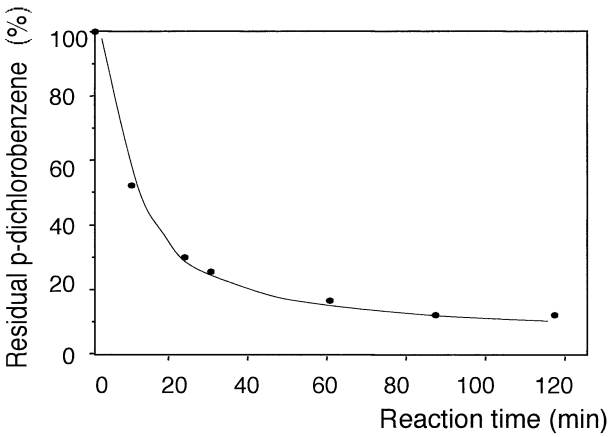

Fig. 3. Removal Efficiency of Rice Bran for *p*-Dichlorobenzene Added to Chemical Wastewater

Data represent the mean ± S.D. of three separate determinations. Rice bran: 10 g/l. *p*-Dichlorobenzene at 50 mg was dissolved in chemical wastewater, and the solution was extended to 1000 ml with chemical wastewater. In addition, it was diluted 10-fold, and 100.0 ml was used for the experiment.

sorption of *p*-dichlorobenzene by rice bran using buffer solutions at reaction time 90 min. Adsorption was observed over the range of pH 1–12. Therefore, it can be applied for the treatment of industrial wastewater over a wide pH range.

Application to Wastewater Treatment

When rice bran was applied to wastewater containing 5 mg/l of *p*-dichlorobenzene (Fig. 3), the percent removal was slightly better than that in pure water because the pH of the wastewater was 10. This observations indicate that rice bran can be used for treatment of wastewater.

Adsorption Mechanism

We investigated the mechanism of removal. We have previously reported that rice bran was effective in removal of organochlorine compounds such as chloroform, dichloromethane and benzene. Furthermore, it was confirmed that the spherosomes isolated from rice bran were effective in removing these organic compounds.[10)]

Analytical and laser microscopic data have confirmed that the removal of organochlorine compounds and benzene is dependent on the uptake of these compounds into intracellular particles called spherosomes.[10)] Spherosomes are intracellular particles about 10 μm in diameter and widely distributed among plants and fungi.[11)] Neither the function of spherosomes nor its analysis is well understood.

Spherosomes are organelles rich in lipid, and they differ in morphology and origin from large oil bodies.[12)] Table 3 shows the removal efficiency of *p*-dichlorobenzene by spherosomes isolated from 1 g (dry wt basis) of beer bran. The removal by spherosomes was similar to that of rice bran. We regarded the special membranes to be related to the uptake of chemical compounds into spherosomes. The chemical nature of the spherosomes is uncertain. Based on the result, we concluded that removal by rice bran is dependent on the uptake into spherosomes.

Table 3. Removal Efficiency of Spherosome Isolated from Various Amount of Rice Bran for *p*-Dichlorobenzene

Amount of rice bran used for isolating spherosome	*p*-Dichlorobenzene concentration (mg/l)		Removal efficiency (%)
	Before treatment	After treatment	
0.5 g	5	2.8–3.5	38.4 ± 5.6[a)]
1.0 g	5	0.9–1.8	73.4 ± 8.8[a)]
2.0 g	5	0.2–0.6	93.4 ± 8.8[a)]

a) Data represent the mean ± S.D. of three separate determinations. Reaction time, 90 min.

Taken together, the findings of this study suggested that the use of rice bran as an adsorbent is an efficient and cost-effective method for removal of *p*-dichlorobenzene from wastewater.

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