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Synthesis of a New Family of Phenol Resin by Enzymatic Oxidative Polymerization

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Enzymatic oxidative polymerization of phenol has been performed in an aqueous organic solvent using horseradish peroxidase as catalyst to produce a new family of phenol resin. The resulting polymer was partly soluble in DMF and DMSO and the fraction soluble in DMF has molecular weight of $3.5\text{x}10^4$. From IR and ^{1}H NMR analyses, the polymer structure was found to be composed of a mixture of phenylene and oxyphenylene units.

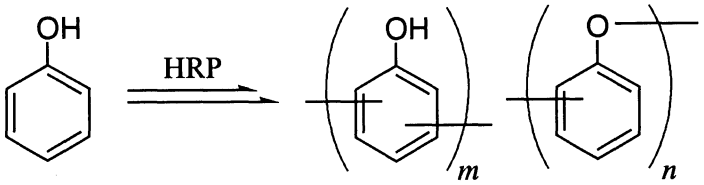
Enzymatic polymerizations provide a new synthetic method of polymers whose synthesis is otherwise difficult and possess much potential for synthesis of polymers with novel structures and properties.1) Typical examples are the first chemical synthesis of cellulose via the polymerization of β-D-cellobiosyl fluoride catalyzed by cellulase enzyme,2) and the oxidative polymerization of *o*-phenylenediamine catalyzed by horseradish peroxidase (HRP) to produce a polymer consisting mainly of an imino-2-aminophenylene unit, which was not able to be obtained by conventional oxidative polymerizations.3) Very recently, the enzymatic polymerization has been extended to a ring-opening polymerization of lactones by using lipase as catalyst.4)

The peroxidase is an enzyme which catalyzes the decomposition of hydrogen peroxide at the expense of aromatic proton donors. HRP is commercially available and is one of the most extensively investigated peroxidases in organic reactions. Phenol resins like novolak and resol resins are very important materials in the industrial field.5) The enzymatic syntheses of these resin derivatives by using HRP catalyst were reported; *p*-phenylphenol was polymerized to produce a high molecular weight polymer with complicated structures.6,7) However, the enzymatic polymerization of phenol itself has not been reported except the oligomer formation from phenol.6,8) This communication deals with preparation of a new phenol resin by the enzymatic polymerization using HRP as catalyst.

HRP is known to show the activity for the oxidation of phenol and aniline derivatives in a mixture of buffer solution and various water-miscible organic solvents such as 1,4-dioxane and acetone.[9,10] In this study, a mixture of 1,4-dioxane and phosphate buffer (pH 7.0) (80:20 vol%) was employed as solvent in order to increase the solubility of the resulting polymer and/or the reaction intermediates during the polymerization. The polymerization of phenol was performed by HRP catalyst at room temperature under air. The polymerization started by the addition of hydrogen peroxide as a source of oxidizing agent.[11] The reaction mixture turned black immediately, and the black and fine precipitates were formed. Then, the color of the reaction mixture gradually became brown and finally pale yellow. After 8 h, phenol was consumed in more than 95%. The polymer was isolated by filtration, followed by successively washing with water and methanol to remove unreacted phenol, oligomeric compounds, HRP, and buffer salts. The resulting polymer was partly soluble in DMF and DMSO, and insoluble in water, benzene, and THF. The yield (methanol-insoluble part) was almost constant in the reaction time longer than 8 h (~ 75%). The polymer obtained for 24 h was further separated by washing with DMF. The ratio of the DMF-soluble part was 25% and the molecular weight determined by gel permeation chromatography (GPC) was 3.5×10^4. As a controlled experiment, the polymerization of phenol in the absence of HRP enzyme did not occur. HRP is a hemoprotein containing iron(III) in the porphyrin ring. Hemin, iron(III) chloroporphyrin was used as a model catalyst for the polymerization of phenol and was found to give no polymeric materials. These data indicate that the present polymerization proceeds by the enzymatic catalysis. From differential scanning calorimetric (DSC) analysis, the resulting polymer possessed no clear melting and glass-transition points.

The effects of the nature of buffer on the polymerization have been examined. In 1,4-dioxane / acetate buffer (pH 4.0) (80:20 vol%) the polymer of molecular weight of 3.2×10^4 was obtained in 67% yield. The use of carbonate buffer (pH 10.0) lowered the yield and molecular weight of the polymer (yield = 20%, molecular weight = 1.9×10^4). The polymerization in a mixture of 1,4-dioxane and sulfite buffer (pH 7.0) afforded no polymeric materials. These data imply that the kind of buffer salt as well as the buffer pH affected the present polymerization. The reaction atmosphere, however, virtually did not affect the present polymerization; the yield and molecular weight of the polymer prepared under argon or oxygen were almost the same as those of the polymer obtained under air.

The structure of the polymer was estimated by IR and ^{1}H NMR spectroscopies. The IR spectrum of the DMF-soluble part shows the broad peak at 3290 cm^{-1} due to O-H bond and the peaks at 1590 and 1490 cm^{-1} ascribed to aromatic C-C bond as well as the strong peak at 1210 cm^{-1} due to C-O-C and/or C-OH bonds. The peak due to C-OH vibration of phenol monomer (1235 cm^{-1}) disappeared. The spectrum of the DMF-insoluble part was similar to that of the DMF-soluble part, implying that the insoluble part has higher molecular weight compared with the soluble part or slightly contains crosslinking structure. In the ^1H NMR spectrum of the DMF-soluble part, a multiplet peak around 6.2-8.0 ppm due to aromatic protons and a broad peak around 9.6 ppm ascribed to hydroxyl proton of phenol group were observed. The integrated ratio of the both peaks was 7.7:1. These data indicate that the structure of the polymer is composed of a mixture of phenylene and oxyphenylene units (the ratio of the phenylene unit to the oxyphenylene unit = 1.2). In contrast, the polymer obtained by the enzymatic polymerization of *p*-phenylphenol was of poly(phenylene) structure.[7)]



The present enzymatic polymerization of phenol by HRP catalyst afforded phenol resin having a new structure with relatively higher molecular weight. Further investigations on the mechanism of the present polymerization and on the detailed characterization of the resulting polymer are now in progress.

References

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8) In ref. 6, there is no detailed description concerning the experimental conditions of polymerization of phenol, the oligomer yield, and the structure of the oligomer from phenol. We have examine the effects of the polymerization conditions on the polymer yield and molecular weight. For example, the concentration of hydrogen peroxide in the reaction solution, i.e., the amount and the number of the addition of hydrogen peroxide greatly affected the polymerization behavior. The difference in the molecular weight

of the resulting polymer is probably due to that of such reaction conditions between the present study and Ref. 6.

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11) The polymerization was performed as follows. Under air, 0.47 g (5.0 mmol) of phenol and 10 mg of HRP were dissolved into 20 mL of 1,4-dioxane and 5mL of phosphate buffer (pH 7.0). By the addition of 28 μL (0.25 mmol) of hydrogen peroxide (30%), the polymerization started. The same amount of hydrogen peroxide was added further 19 times at a 15 minute interval to the polymerization mixture. After 24 h, the resulting polymeric powder was collected by filtration. The polymer was washed successively with water and methanol, followed by drying in vacuo to give 0.34 g of polyphenol (yield 72%).

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