Regio- and Chemo-Selective Polymerization of Phenols Catalyzed by Oxidoreductase Enzyme and Its Model Complexes

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SUMMARY: Oxidative polymerizations of phenol derivatives have been performed using an oxidoreductase enzyme and its model complexes as catalyst to produce new functional polymers. Soluble polyphenols were synthesized using peroxidase catalyst in an aqueous methanol. Enzymatic polymerization of syringic acid involved elimination of carbon dioxide and hydrogen from the monomer to give poly(1,4-oxyphenylene) (PPO). Tyrosinase-model complexes catalyzed highly regioselective oxidative polymerization of a 2,6-unsubstituted phenol, 4-phenoxyphenol, to produce unsubstituted PPO showing crystallinity with a melting point. Chemoselective polymerization of phenols having an unsaturated group took place through peroxidase catalysis, yielding crosslinkable polyphenols.

Introduction

All the naturally occurring polymers are produced *in vivo* catalyzed by enzymes. Recently, reports have been appeared, which handle *in vitro* synthesis of not only biopolymers but also non-natural synthetic polymers through enzymatic catalysis[1-6)]. These enzyme-catalyzed polymerizations ("enzymatic polymerizations") receive much attention as new methodology of polymer syntheses, since in recent years structural variation of synthetic targets on polymers has begun to develop highly selective polymerizations for the increasing demands in the production of various functional polymers in material science. Main target macromolecules by the enzymatic polymerization are polysaccharides, polyesters, poly(amino acid)s, and polyaromatics. The former three polymers are often synthesized using hydrolases as catalyst. A hydrolase is an enzyme which catalyzes hydrolysis of polysaccharides, fats (triglycerides), or peptides. However, it can also act as a catalyst of the reverse reaction by selecting the reaction conditions, leading to the polymer production. As to the polyaromatics, enzymatic synthesis of polyanilines and polyphenols is reported.

Phenol-formaldehyde resins using prepolymers such as novolaks and resols are widely used in industrial fields. These resins show excellent toughness and temperature-resistant properties. However, toxic nature of formaldehyde has problems in their manufacture and use. Therefore,

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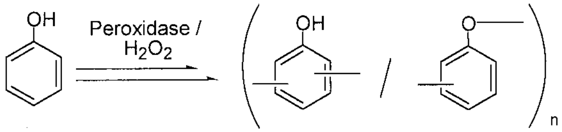
an alternative process for preparation of phenol polymers without using formaldehyde is strongly desired.

For the last decades, enzymatic synthesis of polyphenols has been extensively investigated1-7). In living cells, various oxidoreductases play an important role in maintaining the metabolism of living systems. So far, several oxidoreductases, peroxidase, laccase, bilirubin oxidase etc, have been reported to catalyze oxidation polymerization of phenol derivatives, and among them, peroxidase is most often used. Peroxidase is an enzyme whose catalysis is an oxidation of a donor to an oxidized donor by the action of hydrogen peroxide, liberating two water molecules. Horseradish peroxidase (HRP) is a single-chain β-type hemoprotein that catalyzes the decomposition of hydrogen peroxide at the expense of aromatic proton donors.

The present article deals with our recent progress on the oxidative polymerization of phenols using an oxidoreductase enzyme and enzyme-model complexes as catalyst.

Soluble Polyphenol

In an aqueous organic solvent, peroxidase can act as catalyst for oxidative polymerization of various phenol derivatives, yielding a new class of polyaromatics1-7). The polymerization proceeds at room temperature and during the polymerization, powdery polymers are often precipitated, which are readily collected after the polymerization. In the case of phenol, the simplest and most important phenolic compound in industrial fields, a polyphenol consisting of phenylene and oxyphenylene units was obtained (Scheme 1)8-11). Phenol is a multifunctional monomer for oxidative polymerization, and hence, conventional polymerization catalysts afford an insoluble product with non-controlled structure12). On the other hand, we found that the HRP-catalyzed polymerization in an aqueous methanol produced the DMF-soluble polymer with molecular weight of 2100-6000 in good yields10,11). Furthermore, the unit ratio (regioselectivity) could be controlled by changing the solvent composition; the polymer in the range of the phenylene unit from 32 to 66 % was obtained. The resulting polymer possessed relatively high thermal stability.



Scheme 1.

The soluble polyphenols were enzymatically obtained from *m*-substituted phenols13). The HRP-catalyzed polymerization of *m*-cresol in an equivolume mixture of methanol and phosphate buffer (pH 7) produced the polymer with Tg of 204 °C in a high yield, which was readily soluble in polar solvents such as methanol, acetone, DMF, and DMSO. As to *m*-alkyl substituted phenols, the enzyme origin strongly influenced the polymer yield; HRP could readily polymerize the monomer having a small substituent, whereas in the case of large substituent monomers, the high yield was achieved by using soybean peroxidase (SBP) as catalyst. Bisphenol-A was polymerized by SBP catalyst to give a soluble polymer with molecular weight of several thousands14). Interestingly, the polymer was subjected to thermal curing at 150-200 °C.

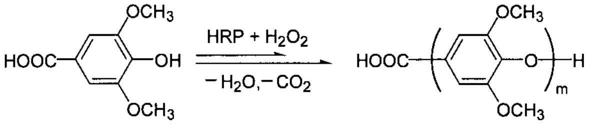
Oxidative Polymerization of Syringic Acid

Poly(2,6-dimethyl-1,4-oxyphenylene) (poly(phenylene oxide), PPO) is widely used as high-performance engineering plastics, since the polymer has excellent chemical and physical properties, e.g., a high glass transition temperature (ca. 210 °C) and mechanically tough property. PPO was first prepared from 2,6-dimethylphenol monomer using a copper/amine catalyst system15). 2,6-Dimethylphenol was also polymerized through HRP catalysis to give the polymer consisting of exclusively 1,4-oxyphenylene unit16), on the other hand, a small amount of Mannish-base and 3,5,3'5'-tetramethyl-4,4'-diphenoquinone units are contained in the commercially available PPO.

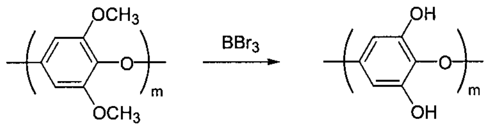
Syringaldehyde is abundantly present in plants as its glycosidic derivatives. We examined the peroxidase-catalyzed polymerization of syringic acid, an acidic form of syringaldehyde (Scheme 2)17,18). The polymerization was started by the addition of hydrogen peroxide (oxidizing agent). In the polymerization using HRP catalyst in a mixture of acetone/acetate buffer (pH 5) (40:60 vol%), polymeric materials were formed during the polymerization. The yield of the methanol-insoluble part was 79 %. The isolated polymer was soluble in common polar organic solvents and its molecular weight was 1.3×10^4. SBP catalyst also gave the polymer with higher molecular weight than HRP. It is to be noted that a polymer was not obtained from non-substituted 4-hydroxybenzoic acid under the similar reaction conditions and conventional oxidation catalysts could not induce the polymerization of syringic acid. Laccases derived from *Pycnoporus coccineus* and *Myceliophthore* were also active for the polymerization of syringic acid. In the polymerization in a mixture of acetone and acetate buffer (pH 5) (40:60 vol%), both enzymes afforded the polymer in more than 80% yield and the molecular weight was in the range of several thousands.

From NMR, IR, and MALDI-TOF mass analyses, the polymer was found to consist exclusively of 1,4-oxyphenylene unit and to have phenolic hydroxyl group at one terminal end and benzoic acid group at the other. Thermal properties were evaluated by using DSC and TG. In the DSC measurement under nitrogen, glass transition temperature (Tg) was observed at 169 °C and the polymer did not show clear melting point below 300 °C. TG analysis under nitrogen showed that temperatures at 5 and 10 weight % loss were 358 and 376 °C, respectively, and the residual ratio at 1000 °C was 19 %.

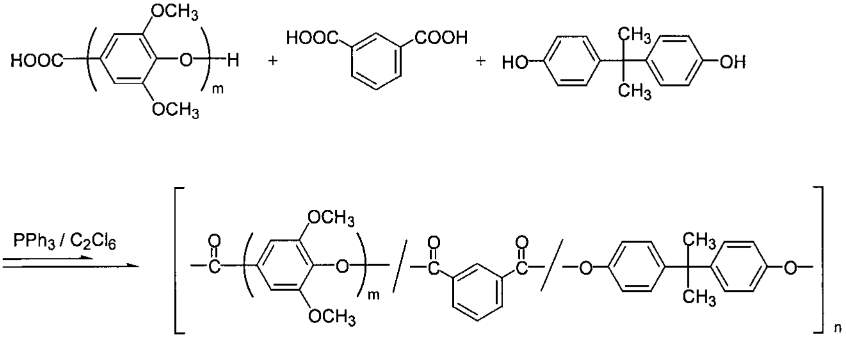
Oxidative Polymerization of Syringic Acid



Synthesis of Poly(2,6-dihydroxy-1,4-oxyphenylene)



Synthesis of Poly(phenylene oxide) - Aromatic Polyester Multiblock Copolymer



Scheme 2.

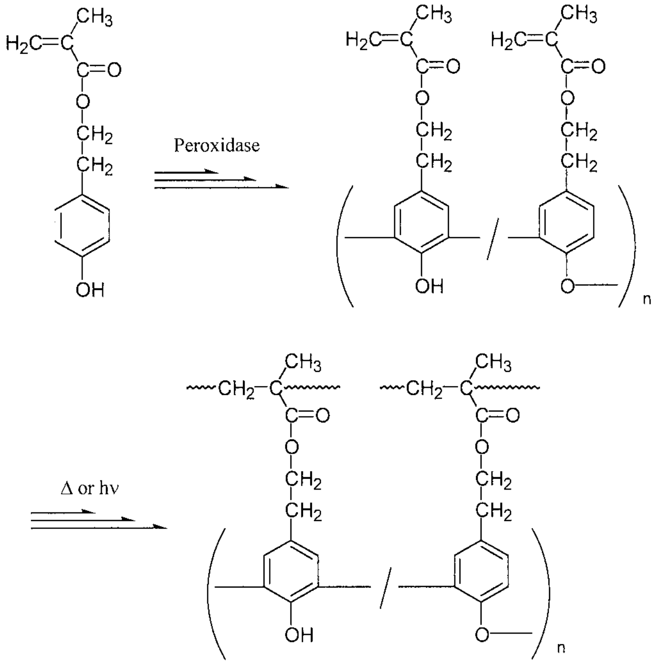
As a possible application, the polyether from syringic acid was converted to a new PPO derivative, poly(2,6-dihydroxy-1,4-oxyphenylene) by demethylation with an excess of boron tribromide (Scheme 2)[19]. The resulting polymer was soluble in DMF, DMSO, and acetone, however, insoluble in chloroform, methanol, and benzene. NMR and IR analyses showed that the extent of the demethylation was 93 % and the polymer was composed of 2,6-dihydroxy-1,4-oxyphenylene unit. The polymer was stable below 300 °C under nitrogen. The resulting polymer is useful as a starting material for synthesis of functional polymers since it has a reactive phenolic group and a PPO backbone having high thermal stability and chemical resistance.

By utilizing terminal heterogeneous two functional groups of the polymer, a new functional polymeric material containing PPO unit was synthesized[20]. Polycondensation of bisphenol-A, isophthalic acid, and the polymer in the presence of triphenylphosphine / hexachloroethane (coupling agent) afforded PPO - aromatic polyester multiblock copolymers (Scheme 2). From TG analysis, the multiblock copolymer was found to show relatively high thermal stability.

Chemoselective Polymerization of Phenol Derivatives to Crosslinkable Polyphenols

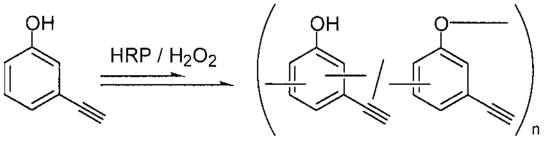
Chemoselective polymerization of a monomer having more than two polymerizable (reactive) groups is expected to afford a new class of reactive polymer having polymerizable or crosslinkable groups in the side chain. In case of such a monomer having an unsaturated polymerizable group, however, it is often difficult to achieve the chemoselective polymerization without involving reaction of the unsaturated group because of high reactivity toward various polymerization catalysts.

We have found that HRP catalysis induced a chemoselective polymerization of a phenol derivative having methacryloyl group21). Only the phenol moiety was polymerized without involving vinyl polymerization of methacryloyl to give a polymer having the methacryloyl group in the side chain (Scheme 3). The resulting polymer was subjected to thermal and photochemical curings.



Scheme 3.

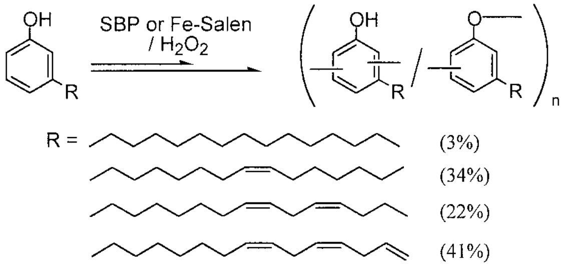
A phenol with an acetylenic substituent in the meta position was also chemoselectively polymerized to give the polyphenol having the acetylenic group (Scheme 4)[22]. The resulting polymer was converted to carbonized polymer in a much higher yield than enzymatically synthesized poly(m-cresol) and is expected to have potential applications as a reactive starting polymer.



Scheme 4.

Cardanol, a main component obtained by thermal treatment of cashew nut shell liquid (CNSL), is a phenol derivative mainly having the meta substituent of a C15 unsaturated hydrocarbon chain mainly with one to three double bonds. Since CNSL is nearly the one third of the total nut weight, much amount of CNSL is formed as by-product from mechanical processes for the edible use of the cashew kernel. Only a small part of cardanol obtained in the production of cashew kernel is used in industrial field, though it has various potential industrial utilizations such as resins, friction lining materials, and surface coatings. Therefore, development of new applications for cardanol is strongly desired.

We synthesized a new crosslinkable polymer by the SBP-catalyzed polymerization of cardanol (Scheme 5)[23]. The polymerization in a mixture of acetone/buffer of pH 7 (75:25 %) produced oily products and the yield and molecular weight of the methanol-insoluble part were 21 % and 4800, respectively. The polymer was soluble in common polar organic solvents. FT-IR analysis showed that the carbon-carbon unsaturated group in the side chain of cardanol was not reacted during the polymerization.



Scheme 5.

Recently, we reported that iron-*N,N'*-ethylenebis(salicylideneamine) (Fe-salen) complex efficiently catalyzed an oxidative polymerization of 2,6-dimethylphenol and *p-t*-butylphenol[24,25]. The catalyst is soluble in some organic solvents, whereas peroxidase is not soluble in common organic solvents. Therefore, Fe-salen was suitable as catalyst for oxidative polymerizations of hydrophobic phenolic monomers such as cardanol. The polymerization proceeded in 1,4-dioxane to give the polymer in good yields26). The yield and molecular weight increased as a function of the catalyst amount. The addition of pyridine slightly improved the yield and molecular weight.

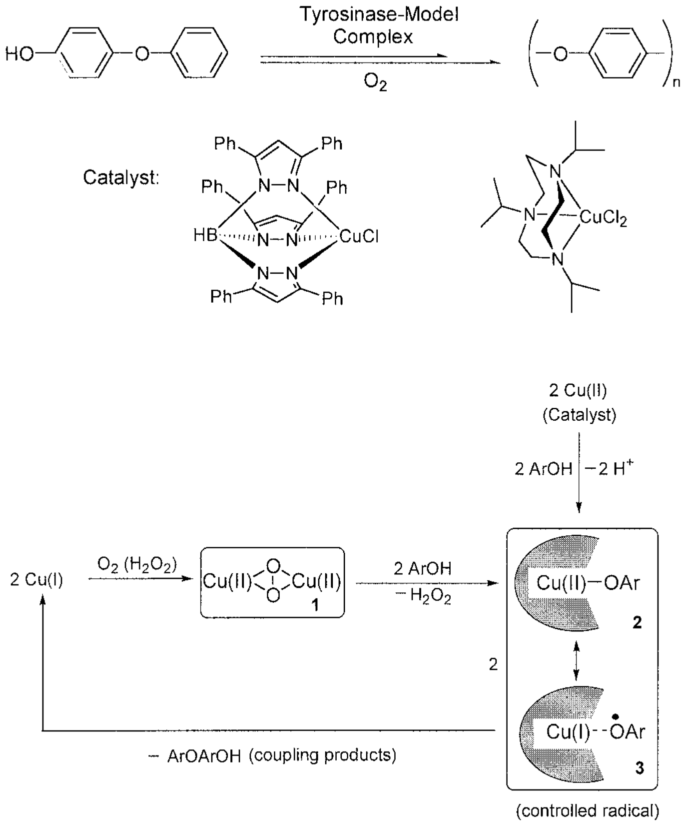
The curing of poly(cardanol) was examined by using cobalt naphthenate as catalyst. The mixture of the polymer and catalyst spread on a glass slide under air. Within 1 h, the polymer was cured to give the crosslinked film with high gloss surface, and the hardness measured by dynamic microhardness tester reached 120 N/mm^2 after 26 days. Under the similar conditions, the cardanol monomer was not subjected to hardening. The crosslinking also took place by the thermal treatment at 150 °C for 30 min. The monitoring of the curing by using FT-IR spectroscopy shows that the mechanism of the hardening reaction was similar to that of autoxidation of oils.

Regioselective Polymerization Using Tyrosinase-Model Complexes as Catalyst

Regioselective oxidative polymerization of phenol leading to *2,6-unsubstituted* PPO has not been achieved. This is due to the low selectivity in "electrophilic" or "free" phenoxy radical coupling. It has been found that polymerization of 4-phenoxyphenol using tyrosinase model complexes as catalyst regioselectively proceeded to give unsubstituted crystalline PPO with melting points (Scheme 6)27,28). The high selectivity is explained as follows. "Nucleophillic" μ-η^2:η^2-peroxo dicopper(II) complex **1** is generated as the sole active oxygen complex from the catalyst, and abstracts proton (not hydrogen atom) from the monomer to give phenoxo–copper(II) complex **2**, equivalent to phenoxy radical–copper(I) complex **3**. Intermediates **2** and/or **3** are not "free" radicals, but "controlled" radicals, and hence, regioselectivity of the subsequent coupling is regulated. The same catalyst system induced an oxidative polymerization of 2,5-dimethylphenol, giving rise to poly(2,5-dimethyl-1,4-phenylene oxide) having melting point of 275-308 °C29).

Conclusion

As described above, production of functional materials from phenols using an oxidoreductase enzyme or enzyme-related catalysts has advantages as follows: (i) the polymerization of phenols proceeds under mild reaction conditions without use of toxic reagents; (ii) phenol monomers having various substituents are polymerized to give a new class of functional polyaromatics; (iii) the structure and solubility of the polymer can be controlled by changing the reaction conditions, e.g., solvent composition; (iv) the procedures of the polymerization as well as the polymer isolation are very convenient. Furthermore, oxidative polymerizations



Scheme 6.

are environmentally benign processes owing to a small amount of by-products (often, only water). Therefore, oxidative polymerizations using an enzyme or enzyme model complex as catalyst possess a large potential for friendly production processes of functional polymeric materials in industrial fields.

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