European Polymer Journal 48 (2012) 580–585

Contents lists available at SciVerse ScienceDirect

European Polymer Journal

journal homepage: www.elsevier.com/locate/europolj

Enzymatic polymerization of phenol catalyzed by horseradish peroxidase in aqueous micelle system

Lei Zhang [a,b], Wenshan Zhao [b], Zhanling Ma [b], Guangrui Nie [b], Yuanchen Cui [a,*]

[a] Key Lab of Special Functional Materials, Henan University, Kaifeng 475000, PR China
[b] College of Chemistry and Chemical Engineering, Henan University, Kaifeng 475000, PR China

ARTICLE INFO

Article history:
Received 23 September 2011
Received in revised form 15 November 2011
Accepted 13 December 2011
Available online 24 December 2011

Keywords:
Enzymatic polymerization
Phenol
Solubilization
Horseradish peroxidase
Buffer

ABSTRACT

Enzymatic polymerization of phenol catalyzed by horseradish peroxidase (HRP) has been carried out in aqueous micelle system using sodium dodecyl benzene sulfonate (SDBS) as a surfactant. The addition of SDBS to buffer greatly enhances the polymer yield. With a usage of SDBS over 0.4 g for 5 mmol of phenol monomer, the polymer could be obtained almost quantitatively. The number-molecular weight of the THF-soluble part determined by GPC is increased from 1100 to 2000 with increasing the dosage of SDBS. The phenol polymerization maintains high yields in aqueous micelle system over a wide pH range from 4 to 10. The activity of enzyme in buffer is so high that the phenol polymerization in aqueous micelle system could be completed only in 2 h with high yield. The resulting polymer is a kind of powdery material, which is partly soluble in DMF, DMSO and THF. IR analysis showed that the polymer structure contained a mixture of phenylene and oxyphenylene units. From TG analysis, the polymer was found to possess high thermal stability.

1. Introduction

Phenol–formaldehyde resins are widely used as surface coatings, adhesives, laminates, molding and friction materials. Generally, such resins are produced by curing treatment of novolaks and/or resols, which are obtained by polymerization of phenol with formaldehyde. However, the concern over the toxicity of formaldehyde has resulted in limitations on their production and use. Therefore, an alternative process for preparation of phenol polymers without using formaldehyde is desired.

An enzyme-catalyzed polymerization has been investigated extensively as a new methodology for polymer synthesis, which is expected to be an alternative route for the preparation of phenolic resins without using toxic formaldehyde [1–3]. So far, a new class of useful and high-performance phenolic polymers have been prepared by utilizing peroxidase as catalyst, most of which cannot be obtained by conventional chemical methods [4–14]. In the enzymatic polymerization of phenol and phenol derivatives, using a mixture of organic solvent and buffer often produces the polymer effectively [4–6,15]. Organic solvents that are miscible in water, for example, dioxane, acetone and methanol must be used, despite the decrease in the activity of peroxidase [4].

For the development of an environmentally benign process of polymer production, the use of organic solvents is not preferred. However, the enzymatic polymerization of phenol in a buffer gives the polymer in very low yield. Recently, it was reported that the enzymatic polymerization of phenol in water proceeded efficiently in the presence of the template, such as poly(ethylene glycol) [16,17], poly(ethylene glycol) monododecyl ether [18], poly(ethylene glycol)–poly(propylene glycol)–poly(ethylene glycol) [19] and cyclodextrin derivates [20–22]. The template enables the polymerization in water to produce the polymer in high yield. Carbon nanotube was also used as template

* Corresponding author. Tel./fax: +86 378 3881589.
E-mail address: yuanchencui@126.com (Y. Cui).

0014-3057/$ - see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.eurpolymj.2011.12.011

to synthesize phenolic polymer in water [23]. However, in these novel catalyst systems, the products are complexes of the phenolic polymers and the templates. It is difficult to obtain the pure polymers from the complexes.

While a growing literature exists on using water soluble polymers as templates in the enzymatic phenol polymerization in buffer, very little work has been done with the polymerization in aqueous micelle system [24]. In this paper, we report enzymatic oxidative polymerization of phenol in aqueous micelle system using HRP as catalyst. The pure phenol polymer could be obtained in high yield with moderate molecular weight. The effects of amount of SDBS, buffer pH value, amount of HRP and the addition pattern of hydrogen peroxide upon the polymerization were investigated systematically.

2. Experimental

2.1. Materials

Horseradish peroxidase (RZ = 2.5, activity = 200 U/mg) was purchased from Shanghai Guoyuan Biotechnology Co., Ltd. and used without further purification. All other Chemicals employed in this work were obtained from various commercial suppliers and were of the highest purity available.

2.2. Measurements

IR spectra were performed on an Avatar 360 Fourier Transform Infrared FT-IR spectroscopy (Nicolet Company, American). Thermogravimetric (TG) analyses were performed on a TGA/SDTA851e instrument (Mettler-Toledo Company, Switzerland) at a heating rate of 10 °C/min under nitrogen or under air. Differential scanning calorimetry (DSC) was obtained on a DSC822e instrument (Mettler-Toledo Company, Switzerland) at a heating rate of 10 °C/min under nitrogen. Polymer molecular weight and polydispersity index (PDI) were estimated by gel permeation chromatography (GPC) using a Waters 515 apparatus with THF as the eluent at a flow rate of 1.0 mL/min. The calibration curves for the GPC analysis were obtained using polystyrene as the standard. The phenol concentrations during the enzymatic polymerization were analyzed by HPLC (Agilent 1100) using a C18-reverse phase column (2.1 mm × 150 mm, 5 μm). The mobile phase was composed of acetonitrile, distilled water and acetic acid with ratio of 45:55:0.1 at a flow rate of 0.4 mL/min. Phenol was measured at 280 nm using a UV absorbance detector.

2.3. Enzymatic polymerization of phenol in aqueous micelle system

SDBS and phenol (5 mmol) were dissolved in 0.1 M phosphate buffer (45 mL, pH 7.0). Then, an enzyme solution of HRP (1 mg) in 0.1 M phosphate buffer (5 mL, pH 7.0) was added. To this solution, hydrogen peroxide (5% aqueous solution, 0.25 mL) was added every 5 min for 14 times with general stirring at room temperature. The mixture was stirred for an additional 0.5 h. After that, the black mixture was vacuum-filtered and washed with water thoroughly to remove HRP, SDBS, residual hydrogen peroxide and phenol. The black filtrate was dried in vacuum at 60 °C to give the phenol polymer.

2.4. Titration of hydroxyl groups in phenol polymer

The ratio of phenylene to oxyphenylene units (Ph/Ox) was determined by titration method. Phenol polymer (0.10 g) was dissolved in 5.0 mL of pyridine containing 2.5% acetic anhydride. The solution was kept at 95 °C for 1 h with gentle stirring. After cooling to room temperature, water (0.50 mL) was added to the reaction mixture, then, the mixture was again heated at 95 °C for 10 min. The solution was titrated with 0.20 M sodium hydroxide in ethanol in the presence of phenolphthalein as an indicator [25].

3. Results and discussion

3.1. HRP-catalyzed polymerization of phenol in aqueous micelle system

At first, we set out to assess the influence of aqueous micelle on HRP-catalyzed phenol polymerization. SDBS was employed as surfactant, which is widely used in various fields. We examined the HRP-catalyzed polymerization of phenol in phosphate buffer (pH = 7) containing 0.15 g SDBS. The phenol conversions during the enzymatic polymerization were presented as Fig. 1 (curve *a*). The polymerization of phenol catalyzed by HRP in the absence of SDBS was also checked and the conversion changes were shown as cure *b* in Fig. 1. It can be seen that the polymerization in a phosphate buffer (pH 7) did not performed effectively; the final conversion of phenol was less than 6.0%. The similar results were also observed by other groups [4,8,15,23]. On the other hand, the polymerization in a mixture of the phosphate buffer and 0.15 g of SDBS gave polymer effectively and the conversion of phenol was up to 50%.

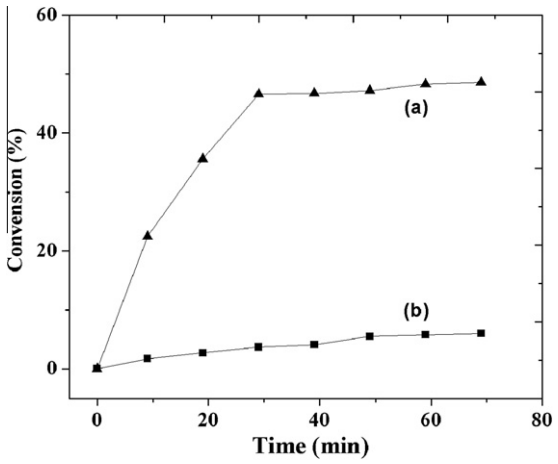

Fig. 1. Plots of conversion of phenol versus reaction time with different dosage of SDBS; (a) dosage of SDBS is 0.15 g, (b) dosage of SDBS is 0 g.