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Determination of catalytic oxidation products of phenol by RP-HPLC

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Abstract A reversed-phase high-performance liquid chromatography (RP-HPLC) with ultraviolet detection was established for the determination of phenol, catechol, hydroquinone, and *p*-benzoquinone in the reaction solution of catalytic oxidation of phenol using hydrogen peroxide as the oxidant and copper-doped FeSBA-15 zeolite as the catalyst. Separation was accomplished on a reversed-phase C_{18} column, and the elution condition was optimized by changing the composition of the mobile phase. A good resolution of all of the relative components in the reaction solution was achieved when the mobile phase was methanol–water–1% acetic acid aqueous solution = 10:50:40 (v/v/v). The concentrations of phenol, catechol, hydroquinone, and *p*-benzoquinone were determined in 11 different reaction solutions by the external standard method. The proposed HPLC method was simple, accurate, reliable, and suitable for tracing the amount of target products during the catalytic oxidation reaction of phenol. The results can provide data support for evaluating the

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properties of catalysts, and, thus, guide the selection of catalysts for the industrial production of dihydric phenol.

Keywords Reversed-phase high-performance liquid chromatography (RP-HPLC) · Phenol · Catechol · Hydroquinone · *p*-Benzoquinone · Catalytic oxidation

Introduction

The catalytic oxidation of phenol has attracted considerable attention in recent years due to the important use of products generated from this reaction, namely, catechol and hydroquinone. Catechol and hydroquinone are basic organic chemical raw materials, intermediates, and additives of medicine, pesticide, dye, perfume, rubber, and photographic material. In addition, they find uses as auto-oxidation inhibitor, monomer polymerization inhibitor, oil anticoagulant, catalyst for ammonia, etc. [1–3]. The catalytic oxidation of phenol using hydrogen peroxide as an oxidant over catalyst (Fig. 1) has become a main method for the large-scale production of dihydric phenols, owing to its good reputation of "clean technology". Obviously, the catalyst plays a key role in this technology. The incorporation of transition metal ions into framework sites of SBA-15, a new type of mesoporous silica molecular sieve, has become a popular procedure for the preparation of catalysts in the oxidation of phenol [4–6]. Commonly, the conversion rate of phenol and yields of dihydric phenols are used for estimating the extent of the reaction. Therefore, an indispensable part of the catalytic oxidation of phenol is to develop an effective method for the determination of products and related compounds in this reaction.

The analytical methods for phenol and dihydric phenols include ultraviolet–visible (UV) spectrophotometry [7], thin-layer chromatography (TLC) [8], and fluorescence [9–12], while the most frequently used methods are gas chromatography (GC) [13–16], high-performance liquid chromatography (HPLC) [17–20], and electrochemical methods [21–24]. The UV spectra of phenol, catechol, and hydroquinone overlap with one another when using spectrophotometry, so software analysis such as the successive approximation method [25], multi-wavelength linear

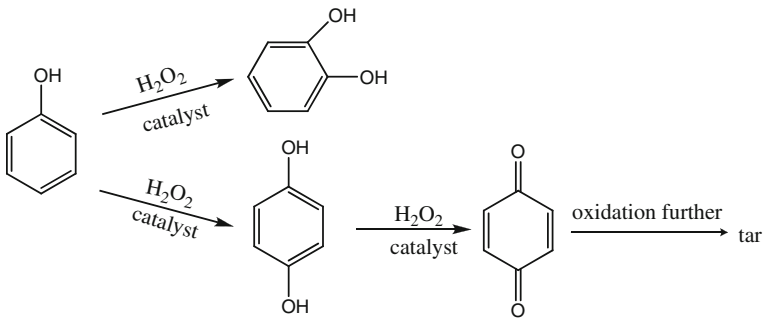

Fig. 1 Catalytic oxidation reaction of phenol

regression-differential coefficient spectrophotometry [26], and factor analysis-differential coefficient spectrophotometry [27] must be used in order to distinguish them. However, these analyses are complex and of low sensitivity. The selection of developing solvent is time-consuming in TLC analysis. Conventional fluorometry cannot determine dihydric phenols simultaneously. Yang et al. [12] obtained a better result when the synchronous-polarization-derivative technique was used. The common electrochemical method to determine phenol and dihydric phenols is to employ a modified electrode. However, chemical modification to prepare electrodes is complex, expensive, and of low reproducibility. The accuracy of using GC to determine phenols is low, due to the poor stability and reliability of GC combined with the oxidation property of phenols under operation temperature. Comparatively, reversed-phase (RP)-HPLC is simple, rapid, and accurate. However, the previously reported RP-HPLC methods focused on a single determination or two or three simultaneous determinations of phenol and dihydric phenols. There has been no report on the simultaneous determination of phenol, catechol, hydroquinone, and *p*-benzoquinone to date.

Based on the actual situation of impurities in samples, an RP-HPLC–UV method was established, which can accurately determine phenol, catechol, hydroquinone, and *p*-benzoquinone simultaneously. The obtained data intuitively reflected the extent of the reaction; thereby, it provided the basis for evaluating the catalytic performance of catalysts, and then it directed the technology optimization of the catalytic oxidation of phenol to dihydric phenols.

Experimental

Instruments and reagents

The HPLC instrumentation included a Waters Alliance 2695 Separations Module equipped with a vacuum degasser, a quaternary pump, and an auto–sampler, and was connected to a 996 UV–Vis photodiode–array (PDA) detector and an Empower chromatography manager system (Waters, Milford, MA, USA).

Reference substances (RSs) of phenol (≥98.0%), catechol (≥98.0%), hydroquinone (≥98.0%), and *p*-benzoquinone (≥99.0%) were purchased from Shanghai Lingfeng Chemical Reagent Co., Ltd. (Shanghai, China), Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China), Nanjing Chemical Reagent Co., Ltd. (Nanjing, China), and Shanghai Yuming Industrial Co., Ltd. Yuanhang Reagent Factory (Shanghai, China), respectively. Acetic acid (≥99.5%, analytical reagent) was purchased from Sinopharm Chemical Reagent Co., Ltd. Methanol (HPLC grade) was purchased from Merck (Darmstadt, Germany). Wahaha purified water (Wahaha Group Co., Ltd., Hangzhou, China) was used throughout the experiment.

The samples used in the present experiment were the reaction solutions of phenol oxidized by hydrogen peroxide with FeSBA-15 zeolite doped with different amounts of copper as catalysts.

Chromatographic conditions

HPLC separation was carried out on an Agela Venusil XBP C_{18} column (250 × 4.6 mm i.d., 5 μm) (Agela Technology Inc., USA). The column temperature was maintained at 30 °C. An isocratic elution with methanol, water, and 1% (vol.%) acetic acid solution (10:50:40, v/v/v) was employed at a flow rate of 1.0 mL/min. The injection volume was 10 μL. The effluent was monitored by the PDA detector set at 244 nm for *p*-benzoquinone, and 275 nm for phenol, catechol, and hydroquinone.

Results and discussion

Standard curves and detection limits

The individual stock solutions of phenol, catechol, hydroquinone, and *p*-benzoquinone used for calibration purposes were prepared by separately weighing a certain amount of RS into four different 10-mL volumetric flasks, among which phenol, catechol, and hydroquinone were dissolved and made up to the mark with water and *p*-benzoquinone with methanol. A proper amount of each stock solution was transferred into a 10-mL volumetric flask, mixed, and water was added to the volume. The concentrations of phenol, catechol, hydroquinone, and *p*-benzoquinone in mixed stock solution were 1.03, 0.154, 0.101, and 0.0112 mg/mL, respectively. Mixed standard solutions were obtained by diluting the mixed stock solution step by step with water and storing in the dark at low temperature. Each mixed standard solution was injected two times consecutively. The linearity of the calibration curve was obtained from the regression of the peak area versus the concentration of the standard. The correlation coefficient (*r*) of *p*-benzoquinone was lower presumably because of the lower concentration level than the others. The limit of detection (LOD) was defined as the concentration where the peak height was three times the background level (Table 1).

Chromatograms of mixed standard solution are shown in Fig. 2. The elution order of the four compounds was hydroquinone, *p*-benzoquinone, catechol, and

Table 1 Standard curves and detection limits

Component	Linear range (mg/mL)	Standard curve	Correlation coefficient (*r*)	Detection limit (mg/mL)
Hydroquinone	$6.00 \times 10^{-4} \sim 0.10$	$A = -1{,}403.17 + 8.51 \times 10^6 C$	0.9999	2.00×10^{-4}
p-Benzoquinone	$1.50 \times 10^{-4} \sim 0.01$	$A = 5{,}525.63 + 5.72 \times 10^6 C$	0.9845	5.00×10^{-5}
Catechol	$9.00 \times 10^{-4} \sim 0.15$	$A = -357.26 + 1.19 \times 10^7 C$	1.0000	3.00×10^{-4}
Phenol	$7.50 \times 10^{-3} \sim 1.03$	$A = -2{,}996.75 + 6.99 \times 10^6 C$	1.0000	2.50×10^{-3}

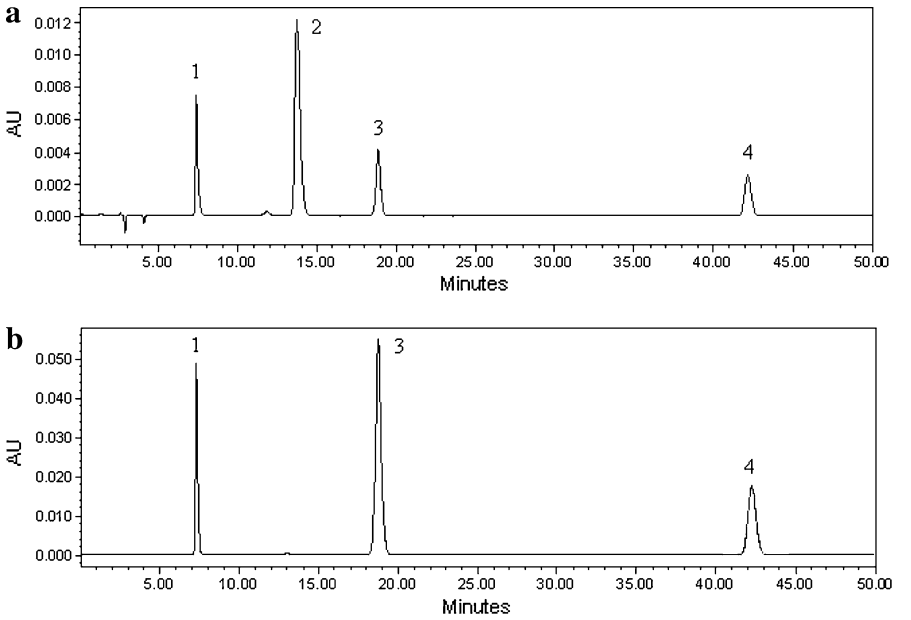

Fig. 2 Chromatograms of mixed standards. Column: Agela Venusil XBP C_{18}, 250 × 4.6 mm i.d., 5 μm; column temperature: 30 °C; mobile phase: methanol–water–acetic acid (1%) aqueous solution = 10:50:40 (v/v/v); flow rate: 1.0 mL/min; injection volume: 10 μL; wavelength for UV detection: 244 nm (**a**) and 275 nm (**b**). Peaks: *1* hydroquinone (0.0101 mg/ml), *2* *p*-benzoquinone (0.00112 mg/mL), *3* catechol (0.0154 mg/mL), *4* phenol (0.103 mg/mL)

phenol, which is associated with the hydrophobic interaction mechanism of RP-HPLC. The hydrophobic group is the same for the four substances, that is, benzene ring, therefore, the elution order is determined by the strength of the hydrophilic group. Hydroquinone has two hydrophilic –OH groups which are in para-position, and hydroxyl oxygen apt to form hydrogen bonds with water in the mobile phase. The two carbonyl oxygen of *p*-benzoquinone also can form hydrogen bonds with water, which is a little weaker than that of hydroxyl oxygen. The intra-molecular hydrogen bond between two ortho-position –OH in catechol reduces the hydroxyl oxygen number to form hydrogen bonds with water. Phenol has only one hydroxyl oxygen to form hydrogen bonds. So, the hydrophobic interaction of hydroquinone with the stationary phase is the weakest and it was eluted first, while the retention of phenol on the hydrophobic stationary phase is the strongest.

Determination of related substances in samples

A quantity of 1.00 mL of catalytic oxidation reaction solution 1 was accurately transferred to 10-mL volumetric flasks in five duplicates, for the intra-day measurement. The first sample solution was injected five times consecutively, and the determined concentrations of hydroquinone, catechol, and phenol were 0.151, 0.240, and 1.94 mg/mL, respectively, with relative standard deviations (RSDs) of

1.6%, 1.8%, and 1.2%, respectively. *p*-Benzoquinone was not detected. The rest of the four sample solutions were all injected twice. Inter-day measurement was performed by the analysis of five duplicates of catalytic oxidation reaction solution 1 diluted ten times in five consecutive days. The results with intra- and inter-day RSDs listed in Table 2 displays a good precision of the established RP-HPLC method for all of the substances determined. On this basis, the concentrations of the four substances in the other ten catalytic oxidation reaction solutions were determined subsequently, and the results are listed in Table 3. For the 11 reaction solutions, *p*-benzoquinone was always not detected. From Tables 2 and 3, it can be seen that the concentration of phenol is the lowest and that of dihydric phenols is the highest in catalytic oxidation reaction solution 8. That is to say, a higher yield of dihydric phenols can be produced when using the catalyst of solution 8.

Recovery test

Catalytic oxidation reaction solution 1 was prepared in triplicate. Each sample was spiked with different amounts of mixed standard solution and diluted to the mark with water. The recoveries were in the range of 97.6–104.5% (Table 4).

Optimization of the chromatographic conditions

Products in the 11 catalytic oxidation reaction solutions are almost the same, owing to the similar reaction conditions, except for the catalysts used. Therefore, only one of them, spiked with *p*-benzoquinone reference, was chosen as the test solution in the optimization of the chromatographic condition.

Selection of the mobile phase

Because of the weak acidity of phenol, catechol, and hydroquinone, acetic acid was chosen as the ion suppressor in the mobile phase. The proportion of 1% acetic acid aqueous solution was kept at 40%, and separation was optimized by changing the content of methanol and water. The separation effect at different methanol proportions is shown in Fig. 3. A good separation with a suitable run time was obtained when the proportion of methanol was 10%. Figure 4 shows the chromatograms of the test solution spiked with *p*-benzoquinone reference under

Table 2 Determination of the four related substances in catalytic oxidation reaction solution 1

Component	Hydroquinone	*p*-Benzoquinone	Catechol	Phenol
Intra-day ($n = 5$)				
Concentration (mg/mL)	0.151	ND	0.240	1.94
RSD (%)	1.6	–	1.8	1.2
Inter-day ($n = 5$)				
Concentration (mg/mL)	0.154	ND	0.235	1.91
RSD (%)	2.1	–	2.6	2.4

Table 3 Determination of the four related substances in the other ten catalytic oxidation reaction solutions

Reaction solution	Concentration (mg/mL)			
	Hydroquinone	*p*-Benzoquinone	Catechol	Phenol
2	0.161	ND	0.241	1.90
3	0.186	ND	0.273	1.87
4	0.190	ND	0.279	1.87
5	0.185	ND	0.268	1.89
6	0.204	ND	0.301	1.80
7	0.203	ND	0.294	1.81
8	0.207	ND	0.305	1.79
9	0.206	ND	0.304	1.80
10	0.167	ND	0.255	1.88
11	0.185	ND	0.294	1.86

Table 4 Recoveries of the four related substances in catalytic oxidation reaction solution 1

Component	Original Concentration (mg/mL)	Added	Detected	Recovery (%)
Hydroquinone	0.01510	0.00507	0.02005	97.6
		0.01014	0.02502	97.8
		0.02028	0.03578	102.0
p-Benzoquinone	ND	0.00056	0.00058	103.6
		0.00112	0.00117	104.5
		0.00224	0.00219	97.8
Catechol	0.02403	0.00772	0.03161	98.2
		0.01544	0.03930	98.9
		0.03088	0.05589	103.2
Phenol	0.1942	0.0516	0.2450	98.4
		0.1031	0.2995	102.1
		0.2062	0.3958	97.8

the mobile phase of methanol–water–acetic acid (1%) aqueous solution = 10:50:40 (v/v/v).

Detection wavelength

The UV absorption spectra of the four compounds obtained by the online PDA detector are shown in Fig. 5. Hydroquinone, catechol, and phenol have similar absorption, with characteristics approximately in 220 and 270 nm, due to their similarity in structure. *p*-Benzoquinone, which is largely different to the others in structure, displays a unique absorption with characteristic of 244 nm. However, in this wavelength, the other three compounds have very low absorption. It is

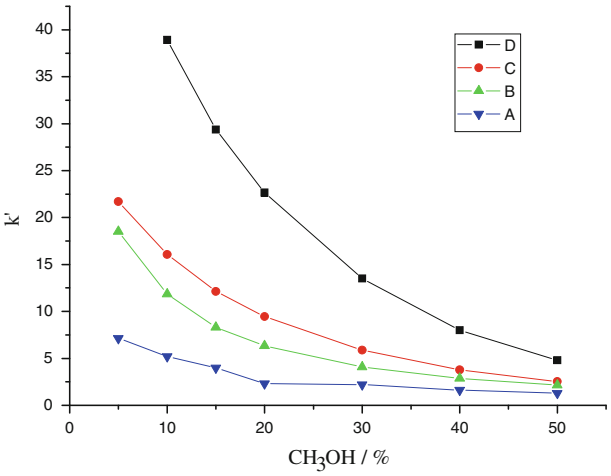

Fig. 3 Effect of the methanol proportion on the retention factor. *A* hydroquinone, *B* *p*-benzoquinone, *C* catechol, *D* phenol

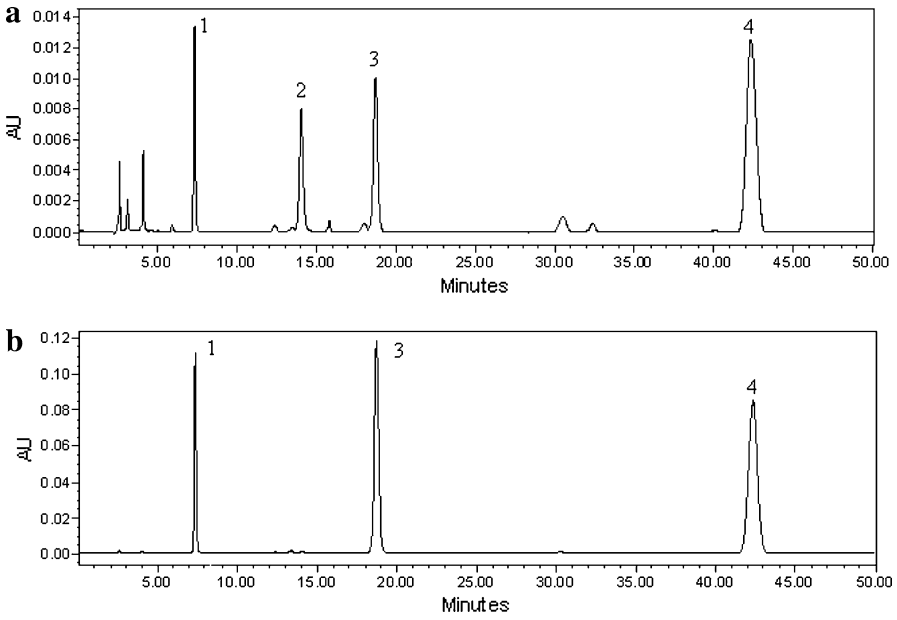

Fig. 4 Chromatograms of the test solution, added by *p*-benzoquinone reference, under 244 nm (**a**) and 275 nm (**b**). Peaks: *1* hydroquinone, *2* *p*-benzoquinone, *3* catechol, *4* phenol

difficult to choose a proper wavelength suitable for all of the compounds monitored. So, dual-wavelength detection was selected, in order to increase the detection sensitivity. Finally, *p*-benzoquinone was detected under 244 nm, and hydroquinone, catechol, and phenol under 275 nm.

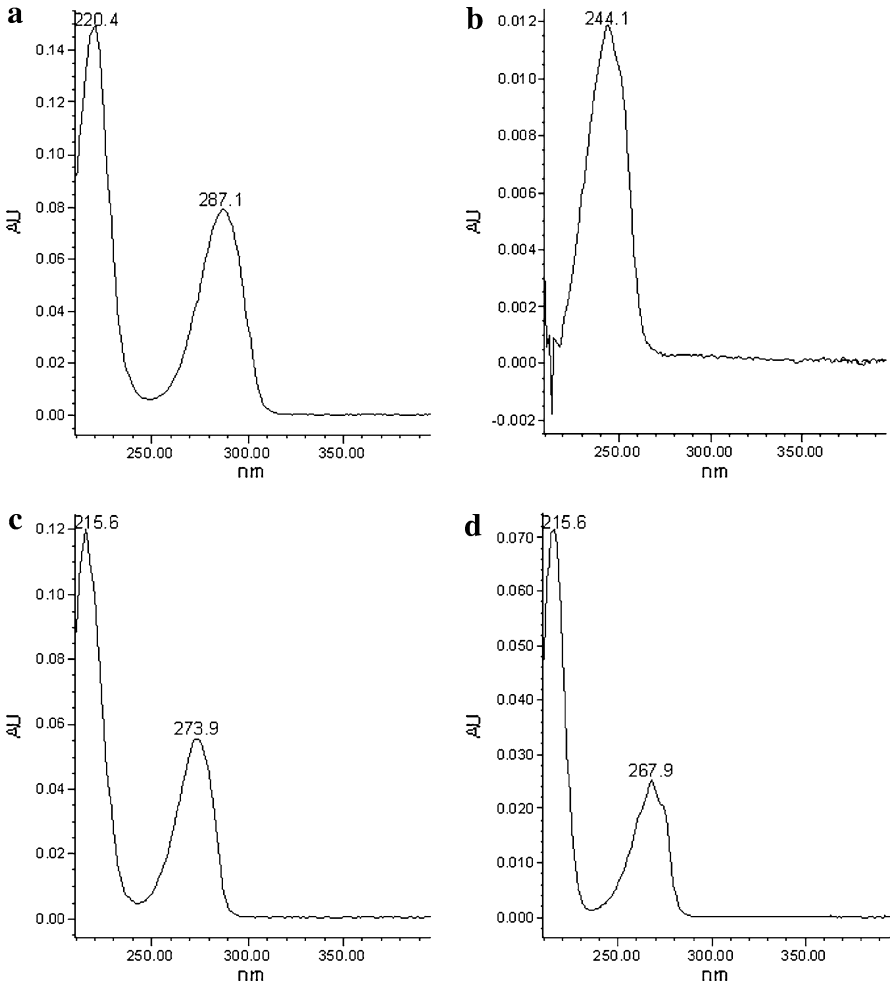

Fig. 5 UV absorption spectra for the four relative substances: **a** hydroquinone, **b** *p*-benzoquinone, **c** catechol, **d** phenol

Conclusions

A reversed-phase high-performance liquid chromatography (RP-HPLC) method was developed for the detection of phenol, catechol, hydroquinone, and *p*-benzoquinone based on the actual situation of impurities in the oxidation reaction solution of phenol with copper-doped FeSBA-15 zeolite as the catalyst. Two detection wavelengths were employed, i.e., 244 nm for *p*-benzoquinone and 275 nm for phenol, catechol, and hydroquinone. The four related compounds in 11 different reaction solutions were analyzed, respectively. The method is accurate and reliable. It can effectively evaluate the extent of the catalytic oxidation reaction and provide

the basis for the selection of catalyst, thereby, guiding the optimization of the process of the catalytic oxidation of phenol.

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