J Fluoresc (2007) 17:257–264
DOI 10.1007/s10895-007-0167-3

ORIGINAL PAPER

Spectrofluorimetric Determination of Pentachlorophenol Based on Its Inhibitory Effect on The Redox Reaction Between Hydroxyl Radicals and Fluorescent Dye Rhodamine B

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Received: 13 October 2006 / Accepted: 1 February 2007 / Published online: 29 March 2007

Abstract Hydroxyl radicals that is generated by Fenton reagent reacts with rhodamine B, which makes the fluorescence quenching of rhodamine B. However, there is an inhibitory effect of pentachlorophenol on the reaction. Based on this observation, an inhibitory fluorimetric method is reported for the determination of trace pentachlorophenol. The fluorescent inhibition of rhodamine B is measured by fix-time method. On the optimum conditions of experimentation, the detection limit for pentachlorophenol is 3.0 ng/ml and the linear range of the determination is 4.0–240 ng/ml. Combined with the samples treating with ion exchange resins and XDA-1 absorption resin, the method has been used for the determination of pentachlorophenol in synthetic samples and natural water samples with satisfactory results. We have also discussed the possible mechanism of the reaction.

Keywords Hydroxyl radicals · Spectrofluorimetric · Pentachlorophenol · XDA-1 absorption resin · Natural water samples

Introduction

Pentachlorophenol (PCP) has been widely used in the industrial, agricultural, and domestic fields as a wood preservative to control mold and insects since its production around 1930s. It is the most toxic representative of the chlorophenols. Although it has been forbidden in several countries, as a fungicide, it is still widely used in wood preservation. The behavior of PCP in the environment is strongly determined by its physicochemical properties related to processes such as adsorption, leaching, vaporization and degradation [1]. The lipophilicity of chlorophenols (CPs) contributes to their bioaccumulation in the food chain. On the basis of the evidence from animal toxicity studies and human clinical data [2–4], and as it's harmful to human health, PCP has been classified as a probable human carcinogenic and listed among the 65 priority pollutants in waters by the United States Environmental Protection Agency (US EPA) [5]. The EPA has regulated the maximum contaminate level (MCL) of PCP in drinking water at 1 ng/ml [6], while European Union (EU) legislation requires the maximum admissible concentration of phenols in drinking water to be 0.5 ng/ml [7]. The World Health Organization (WHO) classifies PCP as a highly dangerous substance that can be absorbed through the respiratory or gastrointestinal systems with strong toxic effects [8]. Because of its environmental persistence, the acute toxicity, and its potential health threat combined with strict regulations of (US EPA) and (EU), special attention has been paid to the determination of PCP in the environment.

Nowadays, the methods most frequently described in the literature for the determination of chlorophenols in water are based on chromatographic analysis, such as gas chromatography/mass (GC-MS) spectroscopy [9–12] and liquid chromatography/mass (LC-MS) spectroscopy [13,14]. The detection of chlorophenols in environment using these techniques has been adequately reported at very low detection levels. However, these techniques need either inconvenient derivatisation approach or toxic organic solvents. At the same time, they are time consuming, expensive, and require trained technicians, which make the popularization difficult. The development of an inexpensive, fast and sensitive method for determination of PCP is of primary interest.

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Hydroxyl radicals that are generated by various approaches have very high reaction activity; they can join in series reactions just as the oxidant or the reducing agent. Of all, the Fenton reagent has extensive applications [15], which uses H_2O_2 as the precursor of the hydroxyl radicals, Fe(II) as the catalytic reagent [16,17]. So far the application of the oxidation of Fenton reagent most concentrated on the degradation of environmental organic pollutions [18–20], there is nearly no report on the analytical applications.

In the present work, combined with the spectrofluorimetry which offers excellent detection limits in the determination of trace amounts of many organic molecules, it have been found experimentally that hydroxyl radicals produced by improved Fenton reagent oxidized rhodamine B (RhB), causing the fluorescence quenching of RhB; the addition of trace PCP has an inhibitory effect on the reaction. Based on this observation, a novel method for determination PCP has been proposed. The method has good sensitivity and selectivity, compared with the chromatographic method, it is low cost, convenient and no need trained technicians. The proposed method has applied in the determination of PCP in 8 synthetic samples and 5 real water samples.

Experimental

Reagents and standards

PCP stock solution (1.000 mg/ml) was obtained by dissolving 0.500 g of solid PCP (analytical reagent grade, Shanghai, China) in 10 ml 10% (w/v) sodium hydroxide solution, then diluting in 500 ml volumetric flask with water and stored in the dark at 4°C. Working standards, 0.010 mg/ml or 0.001 mg/ml, were freshly prepared by diluting the stock solution with water before use. EDTA-Fe(II) stock solution (0.05 mol/l) was obtained by mixing the same volume of 0.1 mol/l EDTA-Na and 0.1 mol/l $FeSO_4$, in which the EDTA-Na and $FeSO_4$ solution were obtained by dissolving given amounts solid EDTA-Na or $FeSO_4$ in given volume of new boiled and cooled distilled water. 1.0×10^{-3} mol/l stock solution of RhB was prepared by dissolving given amounts of RhB in a given volume of water. 0.1 mol/l hydrochloric acid solution; 0.6% hydrogen peroxide solution. All the working solutions were freshly prepared by appropriate dilution of the stock solution. All the other chemicals used were of analytical reagent grade unless otherwise stated and doubly distilled water was used throughout.

Apparatus

Fluorescence spectra were obtained with an FP-6200 spectrofluorometer (JASCO, Japan). Fluorescence intensity measurements were carried out on a Model 930A spectrofluorimeter (Shanghai, China). The fluorescence intensities of solutions were obtained using 1 cm quartz cells. Professional software Origin version 5.0 was used for data processing.

Sample processing

Synthetic samples were prepared by mixing several different common phenolic compounds in different concentration, diluting into volumetric flask with water, adjusting to the pH 3.0 with 0.01 M nitric acid, then passing the solution through XDA-1 absorption resin, the leachates were adjust to pH 7.0 with 0.01 M sodium hydroxide solution. Several environmental water samples such as tap water, rainwater, snow water, surface water as well as river water were fetched and passed through the anion exchange resin (Strong-base OH^- form, Xi'an, China), cation exchange resin (Strong-acid H^+ form, Xi'an, China), and XDA-1 adsorption resin (Xi'an, China) orderly in the pH 3.0, then the leachates were adjust to pH 7.0 (just like above) for analysis.

Procedure

A 25 ml flask was added the reagents in the following sequence: 0.6 ml of 1.0×10^{-4} mol/l RhB solution, 0.5 ml of 10^{-3} mol/l EDTA-Fe(II), 0.5 ml of 0.6% hydrogen peroxide, an appropriate amount of PCP working solution, and 0.6 ml of 0.6 mol/l perchloric acid. The mixture was diluted to the mark with water and shaken until homogeneous, then keep in the room temperature for 10 min. The fluorescence intensity, F was determined at an excitation wavelength of 556.0 nm and emission wavelength of 576.0 nm. The fluorescence value, F_0 of the blank sample without PCP was obtained under the same conditions; then the value of $\Delta F = F\text{-}F_0$ was calculated.

Results and discussion

Spectral characteristics

RhB can emit very strong blood-red fluorescence in aqueous solutions. Its excitation and emission spectra at different experimental conditions were scanned using FP-6200 spectrofluorometer (see Fig. 1). Just like rhodamine 6G, when it oxidized by oxidizers, its molecular structure is destroyed and the fluorescence disappears [21]. In this work, RhB was oxidized by hydroxyl radicals which were generated by Fenton reagent (Fig. 1, 5–5'); the presence of the trace PCP has obvious inhibitory effect on the reaction and results in the reaction rate decrease [Fig. 1, 4–4']. When EDTA-Fe(II) is absent, both the blank reaction and the sample reaction are very unconspicuous (Fig. 1, 3–3' and 2–2'), which indicated that the EDTA-Fe(II) influences the production of hydroxyl

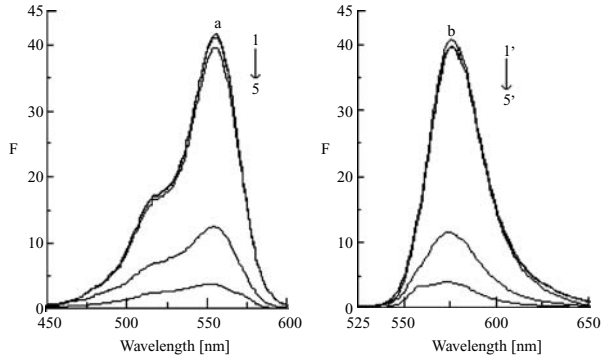

Fig. 1 Excitation (a) and emission (b) spectra of RhB in the presence of different reagents: (1–1'), RhB + $HClO_4$; (2–2'), RhB + $HClO_4$ + H_2O_2; (3–3'), RhB + $HClO_4$ + H_2O_2 + PCP; (4–4'), RhB + $HClO_4$ + H_2O_2 + EDTA-Fe(II) + PCP; (5–5'), RhB + $HClO_4$ + H_2O_2 + EDTA-Fe(II). RhB, 2.4×10^{-6} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; H_2O_2, 0.012%; EDTA-Fe(II), 2.0×10^{-5} mol/l; PCP, 0.08 μg/ml; Time, 10.0 min

radicals significantly. Furthermore, it was observed that there is a linear relationship between ΔF and the concentration of PCP (Fig. 2). The optimum excitation and emission wavelength are 556.0 nm and 576.0 nm.

Effects of variables

To take full advantages of the procedure, the reagent concentrations and reaction conditions must be optimized. Various experimental parameters were studied in order to obtain an optimized system. These parameters were optimized by setting all parameters to be constant and optimizing one each time. (In the all conditions' studies, the concentration of PCP was kept in 0.08 μg/ml.) Each experiment was replicated three times or more.

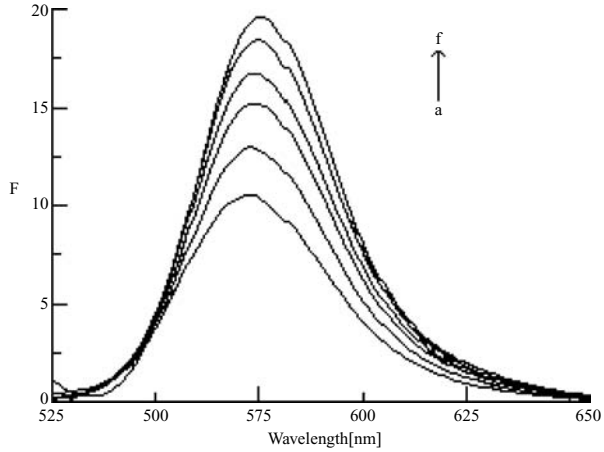

Fig. 2 The fluorescence spectra of different concentration of PCP: Conditions: RhB, 2.4×10^{-6} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; H_2O_2, 0.012%; EDTA-Fe(II), 2.0×10^{-5} mol/l; Time, 10 min; PCP (μg/ml), a, 0; b, 0.08; c, 0.16; d, 0.24; e, 0.32; f, 0.40

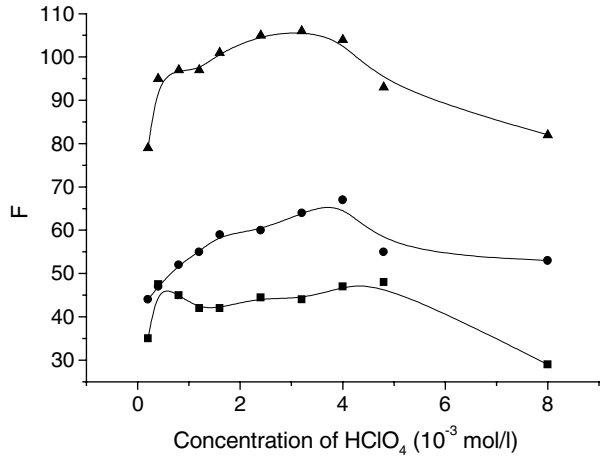

Fig. 3 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of $HClO_4$ concentration. Conditions: RhB, 2.4×10^{-6} mol/l; H_2O_2, 0.012%; EDTA-Fe(II), 2.0×10^{-5} mol/l; PCP, 0.08 μg/ml; Time, 10.0 min

The following media have been tried in the present experiments: hydrochloric acid, sulfuric acid, phosphoric acid and perchloric acid. It was found that there was nearly no inhibitory reaction in phosphoric acid; the sensitivity of the reaction is very low in sulfuric acid and hydrochloric acid. The inhibition effect of PCP is striking only in perchloric acid; furthermore, the relative standard deviation (RSD) is 1.85% for 11 determinations. Therefore, perchloric acid was selected as the reaction medium.

The rate of redox reactions is correlated with redox potential, which is strongly pH dependent [17, 22]. Therefore, the effect of perchloric acid concentration was studied in the range of 2.0×10^{-4}–8.0×10^{-3}mol/l. The results are shown in Fig. 3. In the range of 8.0×10^{-4}–3.2×10^{-3}mol/l, when increasing the perchloric acid concentration, the fluorescence intensity of both the blank reaction (F_0) and sample reaction (F) increased slowly and just keep the ΔF constant. Therefore, a final concentration of 2.4×10^{-3} mol/l perchloric acid was selected as optimum.

In the experiment, the conventional Fenton reagent (Fe(II) + H_2O_2) was tried to be used to generate hydroxyl radicals, however, when the single Fe(II) was studied, it was observed that although the fluorescence of RhB can be quenched obviously, Surprisingly, there is no linear relationship between the ΔF and the adding PCP. When Fe(II) was chelated with EDTA in same molar ratio, the same advantage as Fe(II) can be obtained; in addition, there is good linear relationship between the ΔF and the adding PCP. It is likely that the adding of chelating reagent EDTA increase the solubility of the ferrous salt [23], and EDTA plays a key role in the quantificational generation of free radicals [24]; furthermore, the complexation constant of EDTA-Fe(II) and EDTA- Fe(III) is $\lg k_1 = 14.32$ and $\lg k_2 = 25.1, \lg k_2 > \lg k_1$

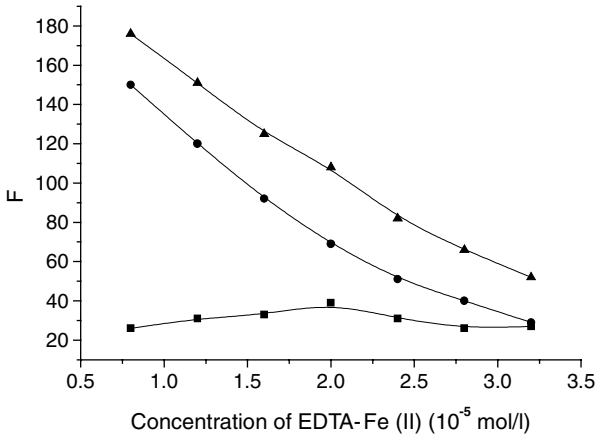

Fig. 4 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of EDTA-Fe(II) concentration. Conditions: RhB, 2.4×10^{-6} mol/l; H_2O_2, 0.012%; $HClO_4$, 2.4×10^{-3} mol/l; PCP, 0.08 μg/mL; Time, 10.0 min

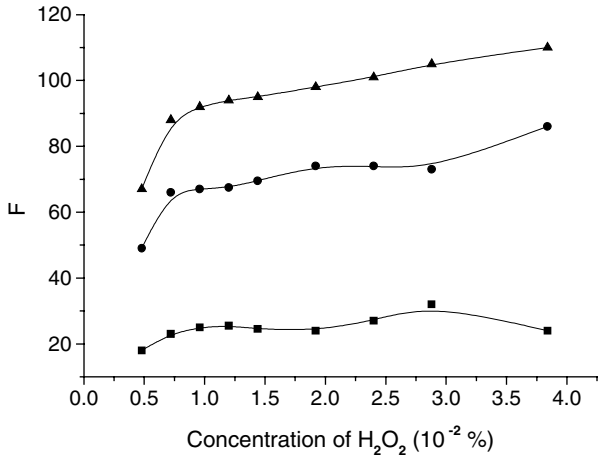

Fig. 5 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of H_2O_2 concentration. Conditions: RhB, 2.4×10^{-6} mol/l^{-1}; EDTA-Fe(II), 2.0×10^{-5} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; PCP, 0.08 μg/mL; Time, 10.0 min

made it more easily of Fe(II)'s oxidation to Fe(III) corresponding to the easier generation of hydroxyl radicals. Especially speaking, it was observed that in the room temperature, when did not control the reaction, the fluorescence intensity F and F_0 decreased simultaneously and just kept the ΔF constant in 1h, so EDTA-Fe(II) was chosen to as the catalytic solution instead of single Fe(II) and nothing measurement to stop the reaction. In the system, EDTA-Fe(II) joined in the reaction as the catalytic reagent, there is a close relation between the concentration of EDTA-Fe(II) and hydroxyl radicals. The effect of EDTA-Fe(II) on both the sample and the blank reactions was studied in the range of $8.0 \times 10^{-6} - 3.2 \times 10^{-5}$ mol/l. The fluorescence intensity change for both reactions decreased with increasing EDTA-Fe(II) concentration. The results are displayed in Fig. 4. The plot of the difference between fluorescence intensity change for blank and sample reactions (ΔF) *vs*. Fe(III) concentration shows a maximum at 1.6×10^{-5} mol/l EDTA-Fe(II). Therefore, a final concentration of 1.6×10^{-5} mol/l EDTA-Fe(II) was considered to be the best choice.

As the precursor of the hydroxyl radicals, the concentration of hydrogen peroxide greatly influenced the hydroxyl radicals' concentration. The influence of concentration of hydrogen peroxide has been investigated in the range of 0.0048–0.0384%, it is shown that with the increase of hydrogen peroxide concentration, the fluorescence intensity of both the sample and blank reactions increased, however, ΔF *vs*. H_2O_2 concentration shows a constant maximum in the range of 0.0072 –0.0192%, just can be seen in Fig. 5. The concentration of H_2O_2 was chosen 0.012% as the optimum condition.

As the fluorescent indicator of the reaction, the effect of RhB concentration was studied in the range of 8.0×10^{-7}–4.0×10^{-6} mol/l. The results showed that an increase in RhB concentration caused an increase in the fluorescence intensity change of both the blank reaction (F_0) and sample reaction (F) (Fig. 6), however, ΔF *vs*. RhB concentration shows a constant maximum in the range of 1.6×10^{-6}–2.8×10^{-6} mol/l, so a final concentration of 2.4×10^{-6} mol/l RhB was used in the work.

The effect of temperature on the fluorescence intensity change of both the blank reaction (F_0) and sample reaction (F) in the range of 15–50°C was investigated. The results are shown in Fig. 7. It was observed that the an increase in temperature caused a decrease in the fluorescence intensity change of both the blank reaction (F_0) and sample reaction (F), however, ΔF *vs*. temperature shows a constant maximum, so room temperature was selected as the reaction temperature. Furthermore, the effect of reaction time was also studied in the range of 0–60 min, and it was observed

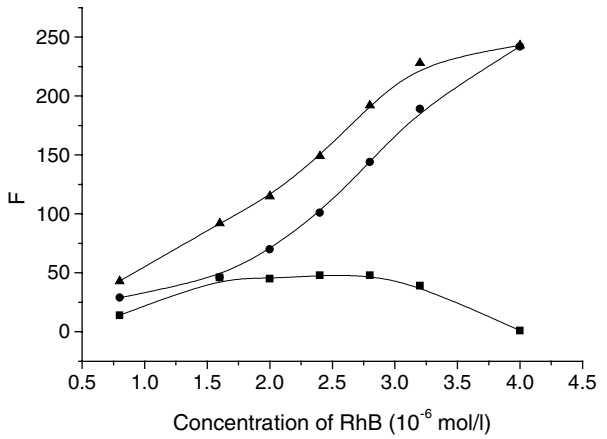

Fig. 6 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of RhB concentration. Conditions: EDTA-Fe(II), 2.0×10^{-5} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; H_2O_2, 0.0144%; PCP, 0.08 μg/ml; Time, 10.0 min

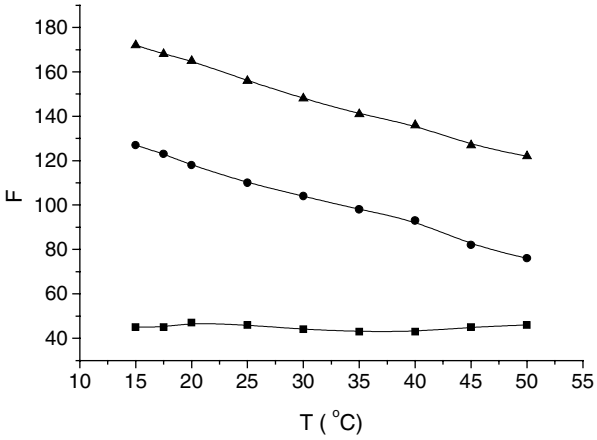

Fig. 7 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of Temperature. Conditions: RhB, 2.4×10^{-6} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; H_2O_2, 0.012%; EDTA-Fe(II), 2.0×10^{-5} mol/l; PCP, 0.08 μg/ml; Time, 5 min

that with the prolong of the reaction time, both the fluorescence intensity change of blank reaction (F_0) and sample reaction (F) decreased, nevertheless, the ΔF *vs*. Time shows a constant value in the range studied, in order to keep the consistent and comparable of the determination value, the reaction time of 10 min was chosen.

Analytical parameters

Under the optimum conditions defined, the calibration graph for PCP was obtained by a fixed time method. For the different concentration of work solution 0.001 mg/ml and 0.010 mg/ml, the graph was linear in the range of 0.024–0.08 μg/ml and 0.02–0.24 μg/ml, respectively; the regression equation is $\Delta F = 16.22 + 395.87\ C$ (μg/ml), with a correlation coefficient of 0.9999 and $\Delta F = 26.39 + 188.94\ C$ (μg/ml), with a correlation coefficient of 0.9999, respectively. The linear relationship was divided into two sections with the different PCP concentration; in addition, the condition of lower PCP concentration has a higher sensitivity. The possible reason is that in the higher PCP concentration, PCP itself can make the fluorescence quenching of the system.

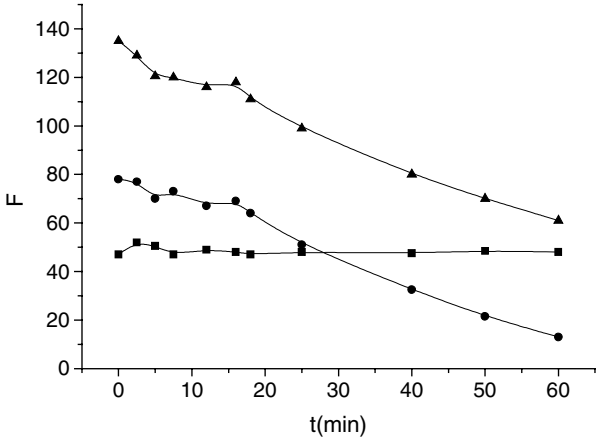

Fig. 8 Fluorescence change for (▲) sample and (●) blank reaction and (■) their difference as a function of Time. Conditions: RhB, 2.4×10^{-6} mol/l; $HClO_4$, 2.4×10^{-3} mol/l; H_2O_2, 0.012%; EDTA-Fe(II), 2.0×10^{-5} mol/l; PCP, 0.08 μg/ml

The determination limit (LOD $= 3S_b/k$, S_b is the standard deviation of the regent blank ($n = 11$), k is the slop of the calibration graph.) and the quantification limit (LOQ $= 10\ S_b/k$) [25] of the method are 0.8 ng/ml and 2.8 ng/ml, respectively.

To evaluate the accuracy and precision of the methods, two independent standard samples were used in the system, the relative error and the RSD is 2.11% and 1.13%, 0.80% and 0.92% for the PCP concentration of 0.012 μg/ml and 0.22 μg/ml, respectively. So the method has well accuracy and precision.

Selectivity

To study the selectivity of the proposed method, the effect of a series of foreign substances on the determination of 0.08 μg/ml PCP was tested under the optimum conditions. The results are summarized in Table 1. It can be seen from Table 1 that except for HPO_4^{2-}, $H_2PO_4^-$, Ag^+, Hg(II) and some phenolic compounds, most of the studied common ions and organic substances do not interfere with the determination. However, the ions influences could be completely removed by using a cation exchange resin (Strong-acid H^+ form, Xi'an, China) and an anion exchange resin (Strong-base OH^- form, Xi'an, China); through the research of synthetic samples, it is found that the influences of the phenolic compounds which were studied could be removed by XDA-1 absorption resin (Xi'an, China) (the procedure can be seen in the section of sample processing).

Application

To evaluate the applicability of the proposed method, it was applied to the determination of 8 synthetic samples and 5 natural water samples (the preparation of the samples shows in the 2.2. Sample processing), the results show in Table 2 and Table 3. The recoveries for the water samples determination are in 90.5%–103.9%, and the relative errors for the synthetic analysis are from -1.44%–$+3.63\%$, which indicates that there is no serious interference through the proposed sample preparation.

The possible reaction mechanism

From the exciting and emission spectra (Fig. 1, 2–2', 3–3', 4–4', 5–5'), it can be seen that PCP has little inhibitory

Table 1 Effect of foreign substances on the determination of 0.08 μg/ml PCP

Foreign substance	Ratio[a]	Change of fluorescence intensity (%)	Foreign substance	Ratio[a]	Change of fluorescence intensity (%)
Na^+	1.4×10^4	+4.3	HCO_3^-	2.5×10^2	−6.7
K^+, NO_3^-	3.0×10^4	+5.7	Ag^+	3.2	+6.0
Cl^-	6.1×10^2	+4.6	Br^-	1.4×10	+7.0
SO_4^{2-}	3.8×10	+5.9	CH_3COO^-	2.2×10	+7.0
NH_4^+	1.8×10^4	+7.4	ClO_3^-	6.6×10	+4.6
Ca^{2+}	6.0×10^3	+6.0	BrO_3^-	3.2×10^3	+4.6
Mg^{2+}	2.4×10^3	+4.5	Hg(II)	4.0	+5.4
Cu^{2+}	1.3×10^2	−4.0	Sucrose	1.0×10^2	+5.6
Zn^{2+}	3.0×10^3	+3.4	Glucose	10	+4.2
Pb^{2+}	2.1×10^3	+4.8	Formic acid	2.0×10^2	+6.5
Cd^{2+}	5.6×10^4	+5.8	Bisphenol A	1.0	+4.1
HPO_4^{2-}	4.0	+4.8	2,4-dichlorophenol (2,4-DCP)	2.5	+5.0
$H_2PO_4^-$	4.6	+4.6	Phenol	2.5×10	+3.9
Ba^{2+}	1.4×10^2	+4.8	o-Nitrophenol (oNP)	1.0	+4.8
Mn^{2+}	1.1×10^2	+5.3	m-Nitrophenol (mNP)	10	+5.1
CO_3^{2-}	3.0×10^2	−5.0	p-Nitrophenol (pNP)	Serious effect	–

[a]The ratio of concentration between the interfering substance and PCP, i.e. [ion]/[PCP].

effect on the oxidation reaction of H_2O_2 and RhB, but it has obvious inhibitory effect on the oxidation reaction of Fenton reagent with RhB.

The well-known Fenton's reaction:

$$Fe(II) + H_2O_2 \rightarrow Fe(III) + \cdot OH + OH^- \quad (1)$$

constitutes a source of hydroxyl radicals' production by chemical means [26], which are often used to carry out chemical oxidation. Hydroxyl radicals have very high reaction activity and are very powerful oxidizing agents. During the work, the EDTA-Fe(II) was used to instead of single Fe(II), and the addition of the little molecule ligands doesn't change the reaction process, but only influence the reaction rate [27].

RhB emits very strong fluorescence, in the experiment, in the acid medium, after the addition of Fenton reagent, the fluorescence of RhB quenched was observed. The possible reason is that the structure of RhB was broken by the powerful oxidizing reagents hydroxyl radicals which were produced by Fenton's reaction, then results in the fluorescence of RhB quenched.

When the PCP was added, combined with the experiments results it can be said that PCP possibly join in the reaction that competed to hydroxyl radicals, which has obvious inhibitory effect on the reaction and then slows the reaction rate of hydroxyl radicals with RhB. As concerning of the reaction of the PCP with $\cdot$OH, generally hydroxyl radicals can react on aromatic compounds by three types of reactions: addition on a double bond, abstraction of a hydrogen atom (if the molecule has a side group -C*n*H*m*X) or electron transfer. In the case of PCP, all the positions of the aromatic cycle being occupied, so, it is probably $\cdot$OH has an *ipso*-attack on the chlorine's position, and the chloride ions were released in the process [28–30] (so in the medium of chloride acid, because of the addition of chloride ions, the reaction of PCP with $\cdot$OH was inhibited, which resulted in the lower sensitivity. On the other hand, in the medium of perchloric acid, perchloric ions reacted with the production of chloride ions which accelerated the the reaction of PCP with $\cdot$OH, so higher sensitivity was obtained.).

Table 2 The results of water samples determination ($n = 4$)

Sample	Found (ng/ml)	PCP added (ng/ml)	PCP found[a] (ng/ml)	Recovery (%)	R.S.D. (%)
Tap water	–	80	77.6	97.0	3.22
Surface water	–	80	79.8	99.8	2.71
Rainwater	–	80	83.1	103.9	1.73
Snowwater	–	80	72.4	90.5	1.59
River water	–	80	81.6	102.0	0.88

[a]Average of four determinations.

Table 3 The results of synthetic samples determination ($n = 4$)

Sample	Composition of synthetic samples (μg/ml)						C_{totle}	$C_{PCPmesured}$	Relative error (%)
	PCP	Phe	2,4-DCP	pNP	mNP	oNP			
A	0.1600	100.00					100.16	0.1652	3.25
B	0.1600		100.00				100.16	0.1612	0.75
C	0.1600	1000.0	100.00				1100.16	0.1594	− 0.38
D	0.1600			10.00			10.16	0.1577	− 1.44
E	0.1600				10.00		10.16	0.1602	0.13
F	0.1600					10.00	10.16	0.1658	3.63
G	0.1600			10.00	10.00	10.00	30.16	0.1580	− 1.25
H	0.1600	100.00	100.00	10.00	10.00	10.00	230.16	0.1589	− 0.69

Combined with the data reported in literatures [26], the possible reaction mechanism was suggested as follows:

$$Fe(II) + H_2O_2 + H^+ \rightarrow Fe(III) + \cdot OH + H_2O \quad (1)$$

$$\cdot OH + RhB \rightarrow P_1 \quad (2)$$

$$\cdot OH + PCP \rightarrow P_2 \quad (3)$$

The Eq. (1) is the redox reaction of Fenton reagent; then hydroxyl radicals are produced; the reaction (2) of hydroxyl radicals' oxidation of RhB results in the fluorescence disappears [31], P_1 is the product of the oxidized RhB. When the PCP is added, reaction (3) occurs, P_2 stands for the series products that PCP reacts with hydroxyl radicals [28–30]. In a definite condition, the concentration of adding PCP has a linear relationship with the recovery of the system's fluorescence.

Conclusions

Combined the conventional Fenton oxidation technique which is mostly applied in water research with sensitive fluorimetric method, we provided a novel method for determination of environmental endocrine-disrupting PCP, this is the first time.

The method has a high sensitivity with a LOD of 0.8 ng/ml and LOQ of 2.8 ng/ml, low RSD, fast operation, low cost, good accuracy and precision as well as high selectivity. The samples were prepared with previous separations which through anionic and cationic exchange resin and XDA-1 absorption resin, then direct determination with satisfactory results in the determination of pentachlorophenol, at ng/ml levels; in diverse water samples, obtaining recoveries between 90.5 and 103.9% in all the cases.

The possible reaction mechanism is that pentachlorophenol has a competition on the redox reaction of rhodamine B with hydroxyl radicals in perchloric acid medium. Especially speaking, in the studies, as a catalytic reagent, EDTA-Fe(II) was found has better stabilization than single Fe(II); also it indicated that the addition of EDTA changed the characteristic of the system, which is no linear relationship previous into a system that has good linear relationship. The EDTA possibly plays a key role in the quantificational generation of free radicals.

Compared with the chromatography methods, the proposed fluorimetic method is rapid, inexpensive and simpler operation, easier to popularization.

Acknowledgments The work was supported by the Natural Science Foundation of Henan Province (No. 0511021200) and Henan Key Laboratory for Environmental Pollution Control.

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