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Fabrication of a novel natural cellulose-based paper chemodosimeter via grafting-to of Rhodamine B moieties for detection of Hg^{2+}

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ABSTRACT

A cellulose-based paper chemodosimeter was designed and prepared by reactions of Rhodamine B with natural cellulose paper. Its sensing property to Hg^{2+} was investigated by fluorescence spectroscopies. While the Hg^{2+}-indicating paper was immersed into the Hg^{2+} solution, the ring of the rhodamine spirolactam opened and the thiosemicarbazide moiety would undergo an irreversible desulfurization reaction to form its corresponding oxadiazole structure, a colorful and fluorescent product. The color change can be discerned by a naked eye under the irradiation of 365 nm UV light. The Hg^{2+}-indicating paper displayed excellent selective toward Hg^{2+} over other commonly metal ions and the sensitive detection of Hg^{2+} was not interfered by other heavy metal ions. The results suggested that the Hg^{2+}-indicating paper would serve as a practical fluorescent chemodosimeter for rapid and convenience detection of Hg^{2+}.

1. Introduction

Mercury is one of the most dangerous environmental pollutants. The widespread pollution of mercury in global environment is mainly from two sources: the natural release such as volcanic emission and the weathering of rocks, and the anthropogenic emissions in the production process of industrial. The total global mercury emissions from both natural and human-generated can reach nearly 7500 tons per year, as estimated by US Environmental Protection Agency (EPA) [1]. Humans exposed to the mercury pollution are primarily through nature water [2,3], even at very low concentration, mercury could bring serious harm to humans. Further, both elemental and ionic mercury can be converted into methylmercury by bacteria in the environment [4], which could enter the food chain and accumulates in the higher organisms, thus causing serious damage to the central nervous and endocrine systems [5].

This serious situation caused by mercury pollution have accelerated the development of Hg^{2+} detection techniques [6]. Many techniques have been accomplished for Hg^{2+} screening, including atomic absorption spectroscopy [7] and inductively coupled plasma mass spectrometry [8], etc., otherwise, these techniques often require expensive instruments, complex sample preparations and highly trained personnel. Therefore, convenient, rapid and efficient detection technique of Hg^{2+} is essential for environmental monitoring of waters and evaluating the safety of aquatic foods.

Fluorescent molecule probes have a remarkable advantage over other detection systems due to its low detection limit and simple instrumentation. Considerable efforts have been devoted to design fluorescent chemosensor or chemodosimeter for detecting Hg^{2+}. Some of the fluorescent probes are based on the principle of fluorescence quenching (turn-off) [9,10]. However, most of the fluorescent probes are based upon the principle of fluorescence enhancement (turn-on) [11–16]. Especially, rhodamine-based fluorescent chemosensors or chemodosimeters have recently developed for the sensing of Hg^{2+}[17]. The rhodamine derivative is suitable for the construction of "turn-on" fluorescent chemosensor or chemodosimeters due to its particular structural property. As the molecular structure of rhodamine derivative with spirolactam structure is nonfluorescent, whereas ring-opening of the spirolactam would give rise to a strong fluorescence emission ("turn-on" process). Furthermore, the fluorescence emission wavelength of rhodamine is longer than 550 nm, which is preferred to serve as a reporting group to avoid the influence caused by the background fluorescence [18,19]. To date, several rhodamine based fluorescent chemosensors or chemodosimeters for Hg^{2+} detection have been developed. For examples, Mahato et al. [20] demonstrated a fluorescence resonance energy transfer (FRET) system for the detection of Cr^{3+} and Hg^{2+} by a naphthalimide-rhodamine 6G based fluorescent chemosensor. Quang et al. [21] reported a rhodamine 6G based fluorescent chemosensor to detect Hg^{2+} by connecting rhodamine 6G and 4-diethylaminosalicylaldehyde. Huang et al. [22] synthesized rhodamin B derivant N-acryloyl-N′-rhodamine B hydrazinethiourea and discussed the influence of intermolecular hydrogen bonding on fluorescence. However, the most of the Hg^{2+} detection reported above were measured in organic

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solvent–water solutions. As the organic solvent which has low boiling point was volatile, if the detection system was placed for a long time before use, the concentration changes of the liquid detection system should affect the detection accuracy. In addition, the detection was often manipulated in transparent glassware, such as in a test tube. We should take many brittle glasswares along with the liquid detection system if we want to conduct on-site detection. Hence this liquid detection system was not convenient to conduct on-site detection for the detection of Hg^{2+}. In the Ma's [23] and Wu's [24] work, the filter paper was immersed into the rhodamine derivatives solution in order to immobilize the rhodamine derivatives onto the filter paper surface, respectively. They purposed to prepare a kind of portable and convenient Hg^{2+}-indicating paper. However, the probes were just only physically adsorbed onto the substrate, so the probe should be easy washed away or dissolved by the solvent when one conduct the detection. This would affect the detection accuracy, even leading to the failure of the detection system. As the result, a portable, convenient and a accurate-measureable system for the mercury ion detection will be required to meet the practical on-site application.

Herein we report the design and preparation of a cellulose-based paper fluorescent chemodosimeter for the detection of Hg^{2+}. The Rhodamine B derivative was covalently grafted to the cellulose paper through grafting-to technique and the Hg^{2+}-indicating paper was prepared. While the Hg^{2+}-indicating paper was immersed into the Hg^{2+} solution, the fluorescence enhancement and the color change can be discerned by naked eyes under the irradiation of 365 nm UV light. We investigated in detail the Hg^{2+}-sensing capabilities of the Hg^{2+}-indicating paper. To the best of our knowledge, there was no information available in literatures about Rhodamine B-grafting cellulose paper to prepare a fluorescent and colorimetric chemodosimeter for the detection of Hg^{2+}.

2. Experimental

2.1. Materials

Rhodamine B (AR, 98%) was purchased from Aladdin-reagent. Ally isothiocyanate (AR, 97%) was supplied by Shijiazhuang Lida Chemicals Co., Ltd. Tetrakis-(triphenylphosphine) palladium (AR, 99.5%) was obtained from Synwitech. 4-Bromobenzyl bromide (AR, 98%) was provided by Beijing Coupling Technology Co., Ltd. Acetonitrile (MeCN) and N,N-Dimethylformamide (DMF) were dried over CaH_2 and distilled just prior to use. Other reagents and solvents obtained from commercial suppliers were used without further purification.

2.2. Instrumentation

^1H spectra were recorded on a Bruker AVANCE DRX 400 NMR spectrometer. FT-IR spectra were obtained using a Bruker TENSOR 27 with samples prepared as KBr disk pellets. PL spectra (at room temperature) were recorded on Hitachi F-4500 fluorescence spectrophotometer. X-ray photoelectron spectroscopy data were acquired with an ESCALab220i-XL electron spectrometer from VG Scientific using 300 W Al Kα radiation. Column chromatography was performed with silica gel (100–200 mesh).

2.3. Synthesis

2.3.1. Synthesis of Rhodamine B hydrazine ($RhBNH_2$)

To a 100 mL flask, Rhodamine B (2.40 g, 5 mmol) was dissolved in 50 mL ethanol. 6 mL hydrazine hydrate was then added slowly dropwise with vigorous stirring at room temperature. Then the stirred mixture was heated to 85 °C and refluxed for 2 h. After cooling to the room temperature, the above mixture solution was dried by reduced pressure distillation for about 30 min until the vacuum degree of the instrument was higher than 0.08 MPa so that there was no clear liquid. 1 mol/L HCl (75 mL) was added to the resultant in the flask to generate a transparent red solution. After that, 1 mol/L NaOH (80 mL) was added slowly with stirring until the pH value of the solution reached 9–10. The resulting precipitate was filtered and washed 3 times with distilled water, and then the precipitate was collected and dried in a vacuum oven for 48 h to provide a pink solid (2.024 g, yield: 88.9%). ^1H NMR (d-DMSO, 400 MHz, ppm):δ1.10(t, 12H, NCH_2CH_3), 3.33(q, 8H, NCH_2CH_3), 4.28(s, 2H, NH_2), 6.34(q, 4H, Xanthene-H), 6.37(d, 2H, Xanthene-H), 7.00(q, 1H, Ar-H), 7.48(m, 2H, Ar-H), 7.77(q, 1H, Ar-H).

2.3.2. Preparation of Bromobenzene-modified cellulose paper (1)

The clean filter paper was cut to be 18 × 18 mm per piece. And then the cut papers were washed with sequence of 0.5 mol/L H_2SO_4, 0.5 mol/L NaOH, EtOH and distilled water, then the papers were dried in the oven for the next reaction.

To a 50 mL flask, 6 pieces of cellulose paper with a dimension of 18 × 18 mm were immersed into 30 mL dry DMF. 0.25 g NaH was added and the mixture was degassed for 15 min. Then 4-Bromobenzyl bromide dissolved in 5 mL dry DMF was slowly added dropwise to the flask, and the solution was stirred at room temperature under N_2 for 20 h. Then compound 1 were all taken out and washed twice with sequence of acetone, DMF, distilled water and EtOH. At last, the sample was Soxhlet extracted for 24 h and dried in a vacuum oven.

2.3.3. Preparation of Ally isothiocyanate-modified cellulose paper (2)

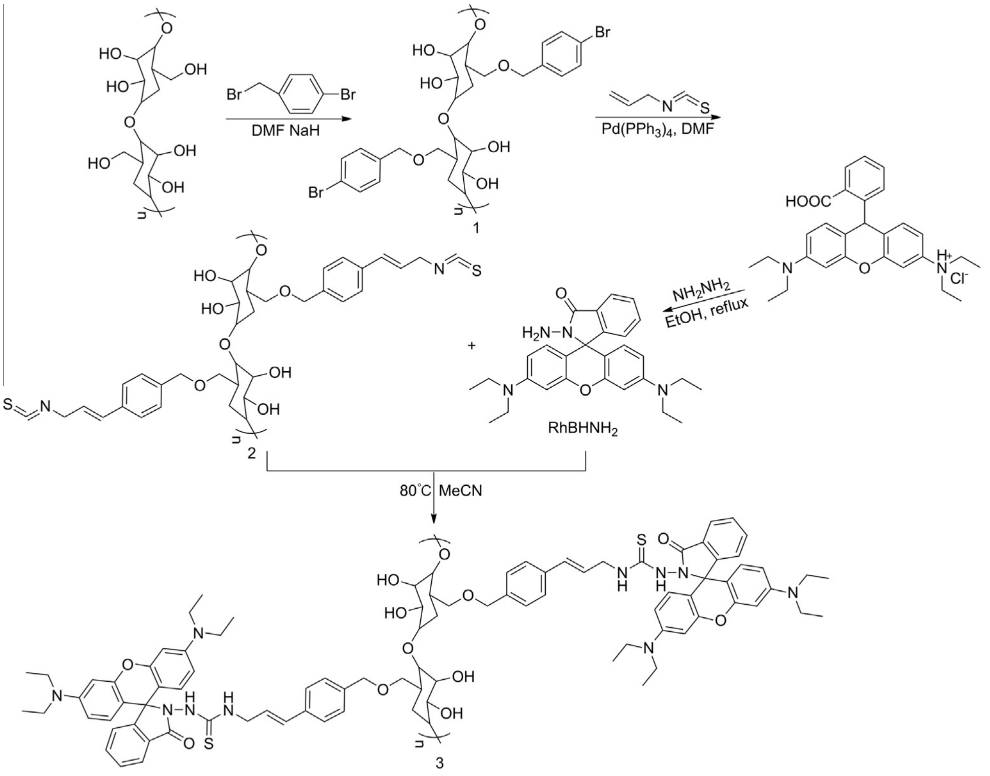
To a 50 mL flask, compound 1 (18 × 18 mm, 6 pieces) were immersed into 30 mL dry DMF. Aqueous solution of potassium bicarbonate (0.2 M, 3 mL) and 0.1 mL ally isothiocyanate were added and the mixture was degassed for 15 min and then $Pd(PPh_3)_4$ (2 mol%, 24 mg) was added under N_2 protection. The above mixture was refluxed at 80 °C under N_2 for 24 h. After cooling to RT, compound 2 were all taken out and washed twice with sequence of acetone, DMF, distilled water and EtOH. At last, the sample was Soxhlet extracted for 24 h and dried in a vacuum oven.

2.3.4. Preparation of Hg^{2+}-indicating paper (3)

To a 50 mL flask, compound 2 (18 × 18 mm, 6 pieces) were immersed into 30 mL dry MeCN. $RhBNH_2$ (23 mg, 0.05 mmol) was added and then the mixture was refluxed at 80 °C for 24 h. After cooling to RT, compound 3 were all taken out and washed twice with sequence of acetone, DMF, distilled water and EtOH. At last, the sample was Soxhlet extracted for 24 h and dried in a vacuum oven (Scheme 1).

2.3.5. Synthesis of N-allyl-N′-Rhodamine B hydrazinethiourea (RhBCH)

To a 50 mL flask, $RhBNH_2$ (0.457 g, 1 mmol) was dissolved in 10 mL of dry MeCN. Ally isothiocyanate (0.149 g, 1.5 mmol) in dry MeCN (10 mL) was then added slowly dropwise with stirring at room temperature. The stirred mixture was heated to 80 °C for 24 h. After cooling to the room temperature, the above mixture solution was dried by reduced pressure distillation for about 30 min until the vacuum degree of the instrument was higher than 0.08 MPa so that there was no clear liquid. The resultant was purified by column chromatography with hexane–ethyl acetate (4:1, v/v) to provide a pink solid (0.309 g, yield:70.1%). ^1H NMR (d-DMSO, 400 MHz, ppm):δ1.09(t, 12H, NCH_2CH_3), 3.35(q, 8H, NCH_2CH_3), 4.81(d, 2H, $CH{=}CH_2$), 5.38–5.39(m, H, $CH{=}CH_2$), 6.33(q, 6H, Xanthene-H), 7.08–7.61(m, 4H, Ar-H), 7.88(d, 1H, NH), 9.01(s, H, -CSNH) (Scheme S1).



Scheme 1. Synthesis route of compound 3.

3. Results and discussion

3.1. Synthesis and characterization of Hg^{2+}-indicating paper

The strategy for the functionalization of cellulose paper is outlined in Scheme 1. The $RhBNH_2$ was first prepared under the reaction between Rhodamine B and hydrazine hydrate. And then the cellulose paper was chosen as a convenient, readily available substrate. The paper was cut to be 18 × 18 mm per piece. And the prepared papers were washed by Sulfuric acid solution, NaOH solution, EtOH and distilled water respectively before performing the reaction in order to remove the impurities adsorbed on the filter paper. The hydroxyl groups of the cellulose paper were etherified by reacting with 4-Bromobenzyl bromide to give the compound 1 containing the bromo-phenyl group. It was well known that the cellulose was composed of glucose, the “grafting-to” reaction mainly occurred with the glucose on the surface of cellulose, each glucose unit contained three hydroxyl groups, it was assumed that the reaction took place in the C-6 position hydroxyl (Scheme 1), however, the hydroxyl groups at C-2 and C-3 positions also had the opportunity to participate in the reaction. And then the bromo-phenyl group of the modified cellulose paper 1 was chemically modified by reacting with ally isothiocyanate under typical Heck coupling conditions to give the compound 2. At last, compound 2 was grafted by $RhBNH_2$ between the $–NH_2$ group and the thiocyanate group to give the target product compound 3. In order to minimize the possible influence of the physical adsorption, in each step after the preparation, the resulting modified paper were Soxhlet extracted for 24 h.

The structure of $RhBNH_2$ was determined by FTIR and ^1H NMR spectroscopy. In the FTIR spectrum (Fig. S1), the two absorption peaks at 3324 cm^{-1} and 3200 cm^{-1} were attributed to the stretching vibration of $—NH_2$, and the strong absorption peak at 1615 cm^{-1} was indicative of the bending vibration of $—NH_2$ group.

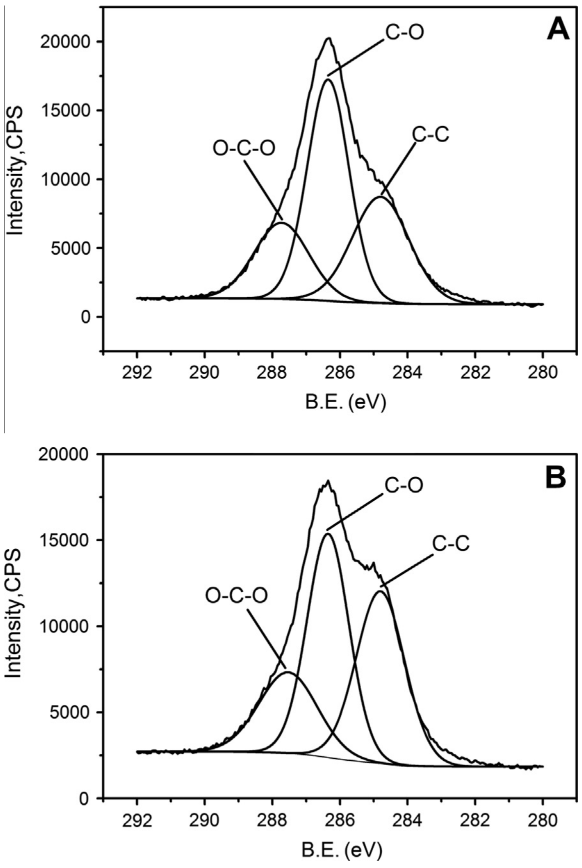


Fig. 1. A: C1s peaks of cellulose paper and B: C1s peaks of compound 3.

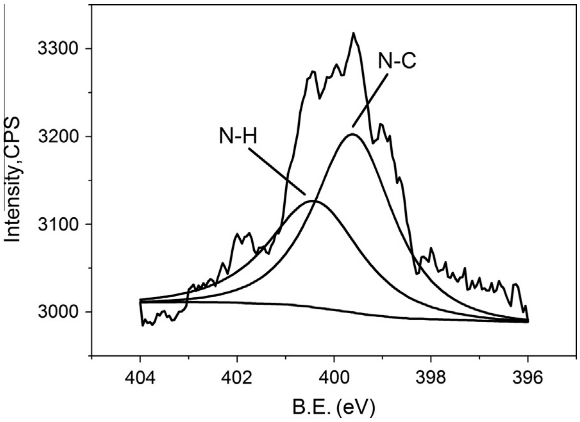


Fig. 2. N1s peak of compound 3.

The ^{1}H NMR spectrum of RhBNH$_2$ was shown in Fig. S2. The characteristic NMR signal at 4.3 ppm was assigned to the —NH$_2$ group. The FTIR spectral analysis and ^{1}H NMR spectroscopy show that the Rhodamine B was converted to RhBNH$_2$.

X-ray photoelectron spectroscopy (XPS) measurements were performed to determine the composition of the as-produced compound 3. The XPS spectra of the starting material cellulose paper and the resultant compound 3 were shown in Fig. S5. A N 1s peak at 399.8 eV was observed for compound 3, whereas no N signal was detected on the cellulose paper, and a weak S 2p peak at 162 eV appeared for compound 3 compared with the cellulose paper. We could think that the RhBNH$_2$ had been grafted to the cellulose paper. The elemental composition was measured to be [O] = 34.8 at.%, [N] = 0.49 at.%, and [S] = 0.11 at.%, which corresponds with atomic ratios of [N]:[S] = 4.455 (according to the formula of compound 3 is expected to 5). As the reaction mainly occurred on the surface of cellulose, the degree of surface substitution could be estimated according to the relative percentage of O and S elements by XPS analysis, each molecular structure of RhBNH$_2$ contained one S elements and two O elements, and every glucose unit contained four O elements, therefore, the degree of surface substitution was calculated to be 0.42%.

The C 1s spectra of the cellulose paper and compound 3 were shown in Fig. 1, the full width at half maximum (FWHM) widened obviously. The percentage of C—C in the cellulose paper was 31%, and the percentage of C—C in compound 3 was 37%, the increase was according to the graft of C-rich RhBNH$_2$ to the cellulose paper. In contrast, the percentage of C—O decreased from 47.6% to 42.7%, and the percentage of O—C—O also decreased from 21.4% to 20.3% due to the C—O bond and O—C—O bond in Rhodamine B moiety were little compared with C—C bond. The N 1s peak (Fig. 2) further indicated that the RhBNH$_2$ had been grafted to the cellulose paper.

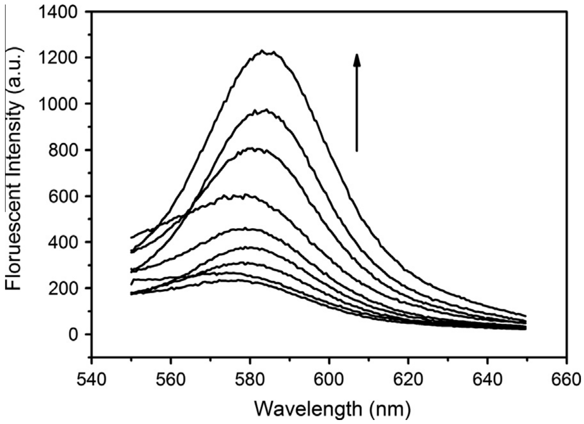


Fig. 3. Fluorescence for compound 3 upon addition of different concentrations of Hg^{2+} (0–1 mM). All the spectra were recorded after the addition of different Hg^{2+} concentrations for 10 min.

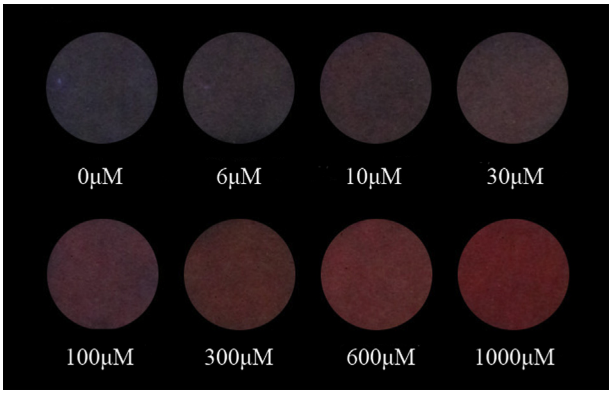


Fig. 4. Optical photographs recorded under the irradiation of 365 nm UV light for compound 3 upon addition of different concentration of Hg^{2+}.

3.2. Synthesis and characterization of RhBCH

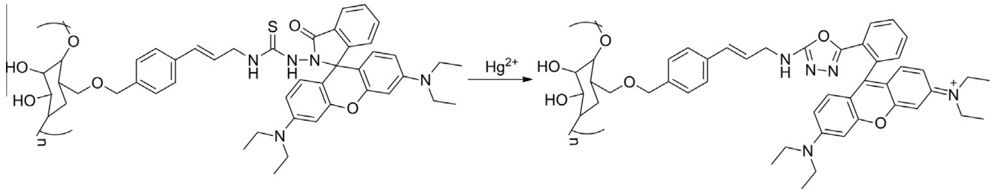
We also synthesized the RhBCH for the preparation of the adsorption-type test paper for the comparison (Scheme S1). The RhBNH$_2$ was reacted with ally isothiocyanate to produce the target product RhBCH.

The structure of RhBCH was also determined by FTIR spectral analysis and ^{1}H NMR spectroscopy. As compared with RhBNH$_2$, the two absorption peaks at 3324 cm^{-1} and 3200 cm^{-1} disappeared, and a new single peak at 3350 cm^{-1} appeared, which was attributed to the stretching vibration of —NH—, as shown in FTIR spectra (as shown in Fig. S3). The ^{1}H NMR spectrum of RhBCH was shown in Fig. S4. The peak at 4.3 ppm disappeared. The peaks at 4.81 ppm and 5.41 ppm were attributed to the —CH=CH$_2$ group. And the peaks at 7.89 ppm and 9.01 ppm were indicated the —NH— group. The FTIR spectral analysis and ^{1}H NMR spectroscopy indicated the formation of RhBCH.

3.3. Fluorescence "turn-on" effect of compound 3

To investigate the sensing capability of the novel chemdosimeter compound 3 to Hg^{2+}, A series of fluorescence measurements upon addition of Hg^{2+} were carried out. All the spectra were recorded after the addition of different Hg^{2+} concentrations for 10 min. The remarkable enhance of compound 3 emission intensity with the increase of the Hg^{2+} concentration illustrate that the fluorescence intensities of compound 3 have been effectively switched on (Fig. 3). The color change of compound 3 upon addition of different concentration of Hg^{2+} can be visualized by a naked eye under the irradiation of 365 nm UV light (Fig. 4).

The rhodamine moiety of compound 3 was spirolactam structure when there was no addition of Hg^{2+} upon compound 3. And the atomic radius of sulfur was larger, the electron withdrawing ability of sulfur was really weak. The lone electron pair of the N atom on rhodamine derivative could transfer along the rhodamine moiety, which could cause the Photoinduced Electron Transfer (PET) process and make the fluorescent emission of rhodamine derivative quenched. As a result, compound 3 displayed weak fluorescence. While Hg^{2+} was added, the Hg^{2+} should promote the ring-open of the corresponding spirolactam and the desulfurization reactions, along with the formation of a more stable five-membered heterocyclic structure 1,3,4-oxadiazole (Scheme 2). The



Scheme 2. The desulfurization of the Hg^{2+}-indicating paper with Hg^{2+}.

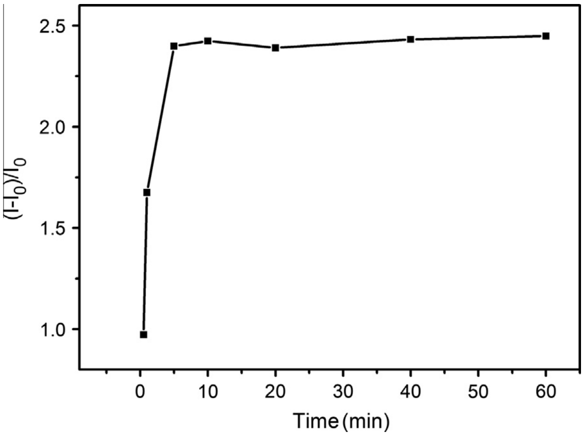


Fig. 5. Effect of time on the fluorescence intensity of compound 3 in the presence of Hg^{2+} (300 μM).

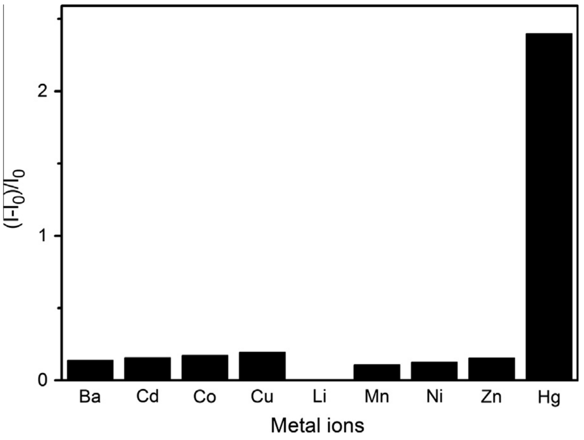


Fig. 6. Fluorescence intensity of compound 3 upon addition of 300 μM of different metal ions. All the spectra were recorded after the addition of different Hg^{2+} concentrations for 10 min.

co-planar structure which was not conductive to the electron transport was destroyed, and the PET process was blocked. As a result, compound 3 gave rise to strong fluorescence [17,25]. We could also discern a slight red-shift from 580 nm to 585 nm for the maximum emission. This was mainly due to the change of molecular structure after the addition of Hg^{2+} that the electron-donating group diethylamino converted to electron-withdrawing group 1,3,4-oxadiazoles [26]. The visual detection limitation for Hg^{2+} was determined to be 10 μM (2 ppm) according to Fig. 3. There was no obvious color change could be observed while the concentration of Hg^{2+} was lower than 10 μM. Wu et al. [27] had prepared the test paper by adsorbing rhodamine 6G derivative to filter paper, and the detection limit for Hg^{2+} was 9 ppm; in the Cheng' work [28,29], they had also immobilized the chemodosimeter to filter paper by adsorptive method, and the detection limit for Hg^{2+} were all determined to be 100 μM. It can be seen that, the detection capability of compound 3 which we had fabricated was better than that of the test strips previous reported. Although the detection limit was not as low as the liquid detection system, it was a new idea for the rapid colorimetric detection of Hg^{2+}. Therefore, compound 3 was convenient to meet the practical application.

We also prepared the adsorption-type test paper. Twenty pieces of cut paper (18 × 18 mm) were immersed into RhBCH solution (10^{-4} M) for 24 h to make sure the rhodamine derivatives were immobilized onto the filter paper surface, and then the test paper were removed and dried in an oven before performing the detection tests. The fluorescent intensity was measured, and the result was shown in Fig. S6. The result of fluorescence enhancement was highly irregular. We can estimate that the RhBCH was physically adsorbed on the paper. While the Hg^{2+} water solution was added dropwise, the RhBCH on the paper surface could be dissolved or washed away by water, which should cause the record inaccuracy.

The time-dependence of fluorescence intensity with Hg^{2+} was also studied. As shown in Fig. 5, the fluorescence intensities of the system increased within few minutes and stabilized as the time continued. It was shown that the reaction rate of compound 3 with Hg^{2+} was very rapid.

In the next step, in order to study the selectivity of compound 3 to Hg^{2+}, the experiments of compound 3 upon other metal ions were carried out. The experiments on other various metal ions were also conducted under the same conditions as the Hg^{2+}, such as Ba^{2+}, Cd^{2+}, Co^{2+}, Cu^{2+}, Li^{+}, Mn^{2+}, Ni^{2+}, Pb^{2+}, Zn^{2+} were also studied. Fig. 6 showed the fluorescence changes for compound 3 at various metal ions. We could see that there was no significant fluorescence increment on adding other metal ions. Although some other metal ions can also trigger the desulfurization reaction, we assumed that the selectivity of metal ions for compound 3 was related to the solubility of the corresponding metal sulfides generated by the desulfurization reaction and the stability of the corresponding sulfide ligand. The Solubility Product Constants K_{sp} value of HgS ($\log K_{sp} = -52.4$) [30] was the lowest compared with other metal ions (Table 1), the more stable HgS can precipitate out from the water and promote the reactions. Therefore, the desulfurization reaction triggered by Hg^{2+} was the most efficient. The selectivity of compound 3 toward Hg^{2+} was higher over other metal ions.

Table 1
Solubility product constants of the metal sulfide.

	Hg	Ba	Cd	Cu	Li	Mn	Ni	Zn	Co
$\log K_{sp}$	−52.4	–	−26.10	−35.20	–	−12.60	−18.5	−21.60	−20.40

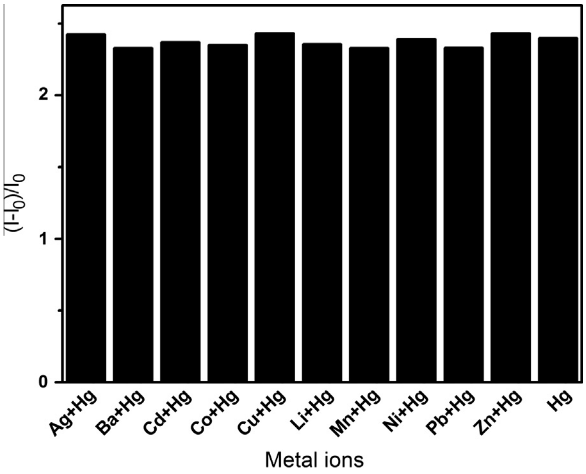


Fig. 7. Fluorescence intensity of compound 3 upon addition of Hg^{2+} (300 μM) and various metal ions (300 μM). All the spectra were recorded after the addition of different Hg^{2+} concentrations for 10 min.

For the better understanding of the selectivity, we then carried out some fluorescence measurements for the compound 3 while Hg^{2+} was coexistent with other metal ions (Fig. 7). The fluorescence intensity of compound 3 toward Hg^{2+} cannot be influenced by other metal ions, as the thiourea group of compound 3 responded to Hg^{2+} through irreversible desulfurization reaction, which prevented other metal ions from interference in real time detection. Herein, compound 3 can serve as a highly selective "turn-on" fluorescent chemodosimeter for the detection of Hg^{2+}.

4. Conclusions

In summary, the functionallization of cellulose paper by Rhodamine B-grafting has been carried out and structurally characterized via XPS spectra. The modified cellulose paper should be used as a fluorescent chemodosimeter for Hg^{2+} detection using chemoselective Hg^{2+} promoted ring-open of spirolactam and the desulfurization of thiourea to form a five-membered heterocyclic system as a design strategy. As demonstrated, the Hg^{2+}-indicating paper displayed excellent sensitivity and selectivity toward Hg^{2+} and its visual limitation was determined to be 10 μM. Notably, a colorimetric change made it possible to detect Hg^{2+} by the naked eye. Taken together, the Hg^{2+}-indicating paper was of particular interest because of its convenience and excellence for the detection of Hg^{2+}.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.reactfunctpolym.2013.08.009.

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