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Wenjuan Qu^{ab}, Guoying Gao^{ab}, Bingbing Shi^{ab}, Peng Zhang^{ab}, Taibao Wei^{ab}, Qi Lin^{ab}, Hong Yao^{ab} & Youming Zhang^{ab}

^a Key Laboratory of Eco-Environment-Related Polymer Materials, Ministry of Education of China, College of Chemistry and Chemical Engineering, Northwest Normal University, Lanzhou 730070, China

^b Gansu Key Laboratory of Polymer Materials, College of Chemistry and Chemical Engineering, Northwest Normal University, Lanzhou 730070, China Published online: 06 Nov 2013.

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An easy prepared double naphthalene Schiff-base for highly selective sensing of cyanide via the dipolymer in aqueous solution

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^aKey Laboratory of Eco-Environment-Related Polymer Materials, Ministry of Education of China, College of Chemistry and Chemical Engineering, Northwest Normal University, Lanzhou 730070, China; ^bGansu Key Laboratory of Polymer Materials, College of Chemistry and Chemical Engineering, Northwest Normal University, Lanzhou 730070, China

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A highly selective chemosensor based on an easy-to-prepare double naphthalene Schiff-base (**R**) is reported for the colorimetric and fluorometric dual-channel sensing of cyanide by taking advantage of the dipolymer in DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution. The detection of cyanide was carried out via the nucleophilic attack of cyanide anion on the C==O of the probe's tautomer with a 1:1 binding stoichiometry, which could be confirmed by ¹H NMR and MS studies. The detection limit of **R** for the determination of cyanide was estimated to be 8.434×10^{-9} M. And other anions, including F⁻, Cl⁻, Br⁻, I⁻, AcO⁻, H₂PO₄⁻, HSO₄⁻, ClO₄⁻, SO₄²⁻ and N₃⁻, had nearly no influence on the probing behaviour. The test strips based on **R** were fabricated, which could act as convenient and efficient CN⁻ test kits.

Keywords: cyanide; chemosensor; compounds combined; HEPES solution; colour change

1. Introduction

Anion recognition is an area of growing interest in supramolecular chemistry on account of its important role in a wide range of environments, clinical, chemical and biological applications (1), and considerable attention has been focused on the design of artificial receptors that are able to selectively recognise and sense anion species (2-5). To be treated as one of the most toxic and deadly anions, cyanide is still being widely used in synthetic fibres, resins, herbicide and the gold-extraction process (6, 7). Unfortunately, cyanide is extraordinarily harmful for humans which damages by absorption through the lungs, gastrointestinal track and skin, and can kill mammals upon binding to a haeme unit (8), which interferes with electron transport, resulting in hypoxia (9). However, cyanide-containing salts are widespread chemicals found in surface water originating not only from industrial waste but also from biological sources (10). It is known that 0.5-3.5 mg/kg of body weight is fatal for humans (11). The highest allowable level of cyanide in drinking water is only 1.9 µM according to the World Health Organisation (WHO) (12-14). Therefore, sensitive and selective detection of cyanide is in great demand; easy and affordable sensors for CN⁻ are in great need for various situations.

Up to the present moment, a large number of chemosensors for cyanide have been invented (15-19). Of all the reported cyanide chemosensors (20, 21), fluorescent chemosensors present many advantages,

including high sensitivity, low cost, easy detection and suitability as a diagnostic tool for biological concern (22). Unluckily, only a few, if any, fluorescent cyanide anion sensors are capable of displaying high selectivity over other anions in DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution. Even like this, most of these sensors for cyanide rely on hydrogen-bonding motifs, and as a result they generally display moderate selectivity over other anions (such as F^- , AcO⁻ and H₂PO⁻₄) (23). In order to overcome the limitation of these sensors, many other detection methods for cyanide have been designed and investigated by utilising nucleophilic reaction of cyanide or demetallation of metal complexes with cyanide (24-25). But, drawbacks still exist for these CN⁻ sensors. For instance, the detection of cyanide cannot be done in aqueous solution in some cases, and some cannot work under mild conditions or the determination procedure is time consuming in other cases (26).

Our research group has a long-standing interest in molecular recognition (27-30). Herein, we report a simple chemosensor (**R**) for CN⁻, which was synthesised in one step by the facile Schiff-base condensation reaction of 2-hydroxyl-1-napthaldehyde and β -naphthylamine. The tautomer of **R** possesses a C=O and a C=N group, as we all know, C=O is easy to take place in addition reaction in which basic ions such as CN⁻ appear. The two naphthalene groups act as fluorophores, while the presence of hydroxyl and C=N in the same sensor molecule confers the

^{*}Corresponding author. Email: zhangnwnu@126.com



Scheme 1. (Colour online) Approach in this work for direct detection of cyanide.

coordination capacity required to coordinate cyanide anions. Importantly, probe **R** displays a very fast response (< 30 s) to cyanide at room temperature, and the fluorescence intensity increased by more than five times with the addition of cyanide.

We next investigated whether other anions could influence the distinguishing of cyanide. To evaluate the selectivity of **R**, we measured the fluorescence intensity of **R** in the presence of various anions (F^- , Cl^- , Br^- , I^- , AcO⁻, H₂PO₄⁻, CN⁻, HSO₄⁻, ClO₄⁻, SO₄²⁻ and N₃⁻). The fluorescence maximum intensity of **R** was dramatically increased almost five times after the addition of cyanide, whereas the fluorescence intensity changes were not observed by other anions but were just comparable to **R** itself. Competitive assay also showed consistent evidence that only **R** was transformed by cyanide anions. The fluorescence intensity of **R** and other anions was restored as much as that of **R**--CN by addition of cyanide to the mixture of **R** and other anions.

2. Experimental

2.1 Materials and physical methods

Fresh double-distilled water was used throughout the experiment. All other reagents and solvents were commercially available at analytical grade and were used without further purification.¹H NMR spectra were recorded with a Mercury-400BB spectrometer at 400 MHz. ¹H chemical shifts are reported in ppm downfield from tetramethylsilane (δ scale) with the

solvent resonances as internal standards. Electrospray ionisation mass spectra (ESI-MS) were measured on an Agilent 1100 LC-MSD-Trap-VL system. UV-vis spectra were recorded on a Shimadzu UV-2550 spectrometer. Photoluminescence spectra were recorded on a Shimadzu RF-5301 fluorescence spectrophotometer. Melting points were measured on an X-4 digital melting point apparatus (uncorrected). The infrared spectra were recorded on a Digilab FTS-3000 FT-IR spectrophotometer.

2.2 Synthesis of sensor molecule R

Compound **R** can be readily prepared by a simple and lowcost Schiff base reaction of 2-hydroxy-1-naphthaldehyde with β -naphthylamine (Schemes 1 and 2). 2-Hydroxy-1naphthaldehyde $(0.346 \text{ g}, 2 \text{ mmol}), \beta$ -naphthylamine (0.358 g, 2.5 mmol) and catalytic amount of acetic acid (AcOH) were combined in hot absolute ethanol (30 ml). The solution was stirred under reflux for 4 h. After cooling to room temperature, the yellow precipitate was filtered, washed three times with hot absolute ethanol, then recrystallised with EtOH/H₂O to give a yellow powder product **R** (1.7 mmol) in 85% yield (melting point 149° C), ¹H NMR (DMSO- d_6 , 400 MHz) δ 15.99/ppm (s, 1H, OH), δ 9.83 (s, 1H, N=CH), 7.03–8.55 (m, 13H, ArH). ¹³C NMR (DMSO-d₆, 100 MHz) δ/ppm 171.10, 155.14, 141.07, 137.04, 136.98, 133.52, 131.51, 129.40, 129.01, 128.94, 127.77, 127.63, 126.86, 126.56, 125.84, 123.48, 122.31, 120.30, 119.59, 117.78, 108.56. IR (KBr) v: 1589 (CH=N), 1558 (C=C), 3059 (ArH), 3438 (O-H) cm⁻¹. Anal. calcd for C₂₁H₁₅NO: C, 84.84; H, 5.05; N, 4.71. Found C 84.81, H 5.05, N 4.71. ESI-MS calcd for $C_{21}H_{15}NO + H^+298.3$, found 298.3 (31).

2.3 General procedure for UV-vis experiments

All the UV–vis experiments were carried out in DMSO– H_2O (8:2, v/v) HEPES buffer (pH 7.21) solution on a Shimadzu UV-2550 spectrometer. Any changes in the UV–vis spectra of the synthesised compound were recorded on addition of tetrabutylammonium salts while keeping the ligand concentration constant (2.0×10^{-5} M) in all experiments. Tetrabutylammonium salt of anions (F⁻, Cl⁻, Br⁻, I⁻, AcO⁻, H₂PO₄⁻, CN⁻, HSO₄⁻ and ClO₄⁻) was used for the UV–vis experiments.



2.4 General procedure for fluorescence spectra experiments

All the fluorescence spectroscopy was carried out in DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution on a Shimadzu RF-5301 spectrometer. Any changes in the fluorescence spectra of the synthesised compound were recorded on addition of tetrabutylammonium salts while keeping the ligand concentration constant $(2.0 \times 10^{-5} \text{ M})$ in all experiments. Tetrabutylammonium salt of anions $(F^-, Cl^-, Br^-, I^-, AcO^-, H_2PO_4^-, CN^-, HSO_4^-$ and ClO_4^-) was used for the fluorescence experiments.

2.5 General procedure for ¹H NMR experiments

For ¹H NMR titrations, the solution of **R** was prepared in DMSO- d_6 and the appropriate concentrated solution of guest was prepared in double-distilled water. Aliquots of the two solutions were mixed directly in NMR tubes.

3. **Results and discussion**

The absorption spectral change of \mathbf{R} in the presence of various anions was studied in DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution and is shown in Figure S1 of the Supplementary Information, available online. Among these anion species tested, only cyanide responded to \mathbf{R} with a decrease at 448 nm. And the fluorescence spectra showed that only \mathbf{R} had a high selectivity for cyanide, other anions almost caused no change in fluorescence even after 180 min. As shown in Figure 1, when excited at 356 nm, it has a remarkable emission intensity at 413 nm. It is concluded that cyanide attacked probe **R** which caused a strong emission signal. Soon afterwards, the colour change from colourless to bright blue could be distinguished by naked eyes as shown in Figure 2.



Figure 1. (Colour online) Fluorescence spectra upon excitation at 356 nm in DMSO-H₂O (8:2, v/v, 0.01 M HEPES, pH 7.21) solution of **R** (2×10^{-5} M) and in the presence of other anions (1 $\times 10^{-3}$ M).



Figure 2. (Colour online) (a) Bar chart of fluorescence spectra upon excitation at 356 nm in DMSO-H₂O (8:2, v/v, 0.01 M HEPES, pH 7.21) solution of **R** (2×10^{-5} M) and in the presence of other anions $(1 \times 10^{-3} \text{ M})$. (b) Visual fluorescence emissions of probe **R** (2 × 10⁻⁵ M) after the addition of F⁻, Cl⁻, Br⁻, I⁻, AcO⁻, $H_2PO_4^-$, CN^- , HSO_4^- , CIO_4^- , N_3^- and SO_4^{2-} in DMSO; H₂O (8:2, v/v, 0.01 M HEPES, pH 7.21) solution on excitation at 356 nm using UV light at room temperature.

To validate the selectivity of sensor \mathbf{R} , the same tests were carried out in the presence of F⁻, Cl⁻, Br⁻, I⁻, AcO^- , $H_2PO_4^-$, CN^- , HSO_4^- , ClO_4^- , SO_4^{2-} and N_3^- anions. None of these anions interfered with the high selectivity of cyanide. It is depicted in Figure 3 that significant fluorescence enhancement was observed only after the



Figure 3. (Colour online) Normalised change in the emission intensity of **R** after addition of the cyanide anion $(1 \times 10^{-3} \text{ M})$ in the presence of other anions $(2 \times 10^{-5} \text{ M})$.

addition of cyanide in the mixed solution, while other anions induced negligible fluorescence enhancement for R under the same conditions. The selectivity of cyanide over other anions, especially CN^- , AcO^- , and $H_2PO_4^-$, is important because many reported cyanide sensors suffer from deleterious interference of these anions. Thus, compound **R** shows high selectivity towards cyanide.

Fluorescence monitoring of the cyanide addition reaction was also carried out by using a 20 µM solution of **R** in DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution at room temperature. Figure 4 shows the fluorescence spectra of R after addition of different amounts of cyanide. As anticipated, the solution of **R** displayed a weak emission signal in the absence of cyanide. However, addition of cyanide anions increased the fluorescence of the R-CN system steadily. For instance, the fluorescence intensity increased by more than five times when the concentration of cyanide reached 410 µl in the solution of R. Indeed, such fluorescence enhancement can

be distinguished with naked eyes. The inset of Figure 4 (a) shows photos of solutions of **R** in the absence and presence of cyanide under UV light (365 nm) illumination. By fluorescence titration, the binding constant between \mathbf{R} and cyanide was determined to be about $1.8 \times 10^5 \,\mathrm{M}^{-1}$ and the detection limit of R towards cyanide was obtained as 8.434 $\times 10^{-9} \text{ M}^{-1}$ as shown in Figure 4 (b) (32).

In addition, the combination of 1:1 of **R** and cyanide was further confirmed by the appearance of a peak at m/z321 and m/z 644, assignable to $[\mathbf{R} + CN^{-} - 2H^{+}]$ and $[2R + 2CN^{-} - 2H^{+}]$ in the ESI-MS (Figure S3 of the Supplementary Information, available online). The ESI-MS display of two strong peaks explains that \mathbf{R} -CN formed as dipolymer, and there may be a $\pi - \pi$ stacking effect. Furthermore, an analysis of the IR spectra of R and after cyanide addition proved informative (1:1, solid complex, KBr). Figure 5 shows a rapid peak decrease at 1597 and new peaks at 2239; the changes of these peaks indicated that C=N was broken and CN⁻ was added to C=O, while the peak at 3426 referenced to the stretching vibration absorption of the N-H bond.

To explore the sensing mechanism of sensor R to CN^{-} , the ¹H NMR titration was investigated, which illustrated the characteristic structural changes occurring upon interaction with CN⁻. ¹H NMR titration displayed the chemical shift changes of R upon addition of CN-. As shown in Figure 6, sensor R showed two single peaks at 15.99 and 9.83 ppm in DMSO, we confirmed that they correspond to the protons of OH and CH=N (Ha), respectively. The intramolecular hydrogen bond was broken upon addition of CN⁻ by water solution; in the corresponding NMR spectra, the signal at 15.99 ppm (OH) disappeared, while at the same time, a new signal was found at 6.407 ppm which is ascribed to aliphatic amino. And then with the addition of CN⁻, the signal at 9.83 ppm (Ha) shifted upfield to a stable value at 9.43 ppm, and the

Figure 4. (Colour online) (a) Fluorescence titration spectra of CN⁻ (410 µl) in DMSO-H₂O (8:2, v/v, 0.01 M HEPES, pH 7.21) solution upon addition of increasing concentration of CN $(\lambda_{ex} = 356 \text{ nm})$. Inset: photograph showing the change in the fluorescence from left to right of **R** and **R**-CN. (b) Calibration curve $1/I - I_0$ of **R** (20 µM) in DMSO-H₂O (8:2, v/v, 0.01 M HEPES, pH 7.21) solution as a function of concentration of CN⁻.

1.5

Figure 5. (Colour online) IR spectra of compound R and R-CN in KBr discs.

2171

2000

Wavelength (cm⁻¹)

1500

1000

500

CN



R+CN

CN

406

1000

800

600

400

200

0.12

0.10

0.08

0.04

0.02

0.00

0.0

0.5

1.0

1/[1-1] 0.06 400

 $K_{a} = 1.8 \times 10^{6}$ $R^2 = 0.996$

Fluorescence intensity (a)



5 15.0 14.5 14.0 13.5 13.0 12.5 12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6

Figure 6. (Colour online) Partial ¹H NMR spectra of \mathbf{R} (0.05 M, DMSO- d_6) and in the presence of varying amounts of \mathbf{CN}^- (N=CH (Ha)).

aromatic hydrocarbon signals from 8.11 ppm to 6.5 ppm shifted upfield significantly.

From the results of ¹H NMR spectroscopic titration, FT-IR spectra and mass spectra, it can be partially proved that the reaction between **R** and CN^- has taken place in two steps (Scheme 2): first, CN^- attacks the tautomer of R and an addition reaction occurs with C==O, and then the compounds form as the dipolymer (Figure 7).

In order to demonstrate the practicality of **R** for the detection of cyanide, a preliminary paper test strip system was constructed as shown in Figure 8. After dropping a DMSO-H₂O (8:2, v/v) HEPES buffer (pH 7.21) solution of **R** on the neutral filter paper and being dried, an orange rectangle with colourless emission under UV light (365 nm) was formed. Careful exposure of the modified filter paper to the aqueous solution containing cyanide quickly switched the emission colour from colourless to bright blue under UV light (365 nm). However, such obvious emission colour variation was not detected for the

filter paper containing **R** after interaction with DMSO– H_2O (8:2, v/v) HEPES buffer (pH 7.21) solution containing other anions.

4. Conclusion

In conclusion, we have presented a simple, facile, low-cost and efficient Schiff-base example of a highly selective chemosensor for CN^- . The sensor gives an immediate response to the cyanide anion by fluorescence turn-on response. And the distinct colour changes under UV light (365 nm) and rapid fluorescence enhancement can be detected by naked eyes. The detection of CN^- by using **R** is found to be totally free of interference from any other anions, and the mechanism of the reaction was investigated by various means (such as ¹H NMR spectroscopic titration, FT-IR spectra and mass spectral analysis). Taking advantage of the dipolymer, as well as the successful application in paper testing, we believe that



Figure 7. (Colour online) Proposed sensing mechanism of sensor \mathbf{R} for the detection of cyanide.



Figure 8. (Colour online) Photographs of **R** of test papers of only **R** and after immersion into $DMSO-H_2O$ (8:2, v/v, 0.01 M HEPES, pH 7.21) solution with CN^- under irradiation at 365 nm.

the test strips could act as a convenient and efficient CN^- test kit.

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Supplemental data

Electronic Supplementary Information available: [Complete experimental procedures and some of the spectroscopic].

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