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Determination of Water Content in Organic Solvents by Naphthalimide Derivative Fluorescent Probe

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A new fluorescence dye, *N*-amino-4-(2-hydroxyethylamino)-1,8-naphthalimide (AHN), based on 4-bromo-1,8-naphthalic anhydride was synthesized as a fluorescence probe for determining water content in organic solvents. Changing from a non-polar to a polar solvent increases the solvent interaction with excitation spectrum and emission spectrum shifts to longer wavelengths. The fluorescence intensity of the probe decreased with the increasing concentration of water. In the range of 0.00 – 4.00% (v/v), the AHN fluorescence intensity changed as a linear function of water content. The detection limits were 0.019, 0.038, and 0.060% for dioxane, acetonitrile and ethanol, respectively.

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Introduction

In many organic solvents, water is generally considered as an impurity, and therefore its measurement in solvents is of importance for many industries and technologies. The traditional method for the quantitative measurement of water in organic solvents is the Karl Fisher titration.[1] Although this approach has several useful characteristics, some disadvantages such as the requirement for skilled personnel and specialized equipment, the complex of sample manipulations and interference from other co-existing species limit its wider application. Therefore, low-cost, rapid, simple, and sensitive alternative methods for the determination of trace amounts of water in organic solvents are a very appealing goal.

The polarity proticity of solvents have great influence on the fluorescence of a fluorescent molecule.[2,3] It is commonly acknowledged that solvent-dependent spectral changes can arise from either general or specific solvent effects.[4] The first effect results from interactions of the dipole moment of the fluorophore with the reactive field induced in the surrounding solvent. The latter results from the short-range interactions between the fluorophore and solvent molecules in its first solvation shell such as hydrogen bonding.[5] Hydrogen bonds between a protic solvent and a fluorescent molecule may form under certain conditions, which can considerably affect the molecular orbital energy levels and accordingly influence the photophysical properties.[6]

As a fluorescent dyes with high stability and strong intensity, naphthalic anhydrides and their derivatives have wide range of applications. These include use as semiconductor oligomer,[7] liquid crystal displays,[8] intercalators to cleave plasmid DNA,[9] and a monoboronic acid probe to determine glucose.[10] They are considered good candidates for new fluorescence dye probes. The introduction of the amino group at the 4-position greatly enhances the fluorescence of the molecule. This group of compounds seems quite promising as fluorescent carriers in the preparation of probes. A solvatochromic fluorophore 4-*N*,*N*-dimethylamino-1,8-naphthalimide[11] was synthesized and used for the detection of a wide range of biomolecular interactions. But to the best of our knowledge, there are few reports about their application in detecting water content.[12,13] As demonstrated in our previous papers,[14,15] naphthalimide are excellent candidates for preparing fluorescence sensors.

In this research, the authors synthesized a new fluorescence water probe for the determination of water content in organic solvents. A derivative of naphthalimide, *N*-amino-4-(2-hydroxyethylamino)-1,8-naphthalimide (AHN), was reported and used as a fluorescence indicator due to its favorable sensitivity to solvent polarity. It is observed experimentally that, with increasing of water content, the AHN fluorescence intensity changed dramatically and the spectra exhibited a red shift. This solvatochromic feature means AHN is promising as a fluorescent probe in the deteremination of water content. The AHN fluorescence probe results with dioxane, acetonitrile and ethanol solutions of different water content are displayed. The successful fabrication of the proposed probe presented satisfactory sensitivity and low detection limits and is a useful example of the use of naphthalimide derivatives for fluorescence probes to detect water.

Experimental

Reagents

4-Bromo-1,8-naphthalic anhydride, purchased from Taizhou Chemicals (Zhejiang, China), was recrystallized twice from chlorobenzene (m.p. 217 – 219°C). Hydrazine and 2-aminoethanol purchased from Damao Chemicals (Tianjin, China) were of analytical reagent grade. All organic solvents were of analytical reagent grade and were used as received or were dried to eliminate any water residue before being used in the experiments. Dioxane was pre-dried over KOH, then decanted and distilled at

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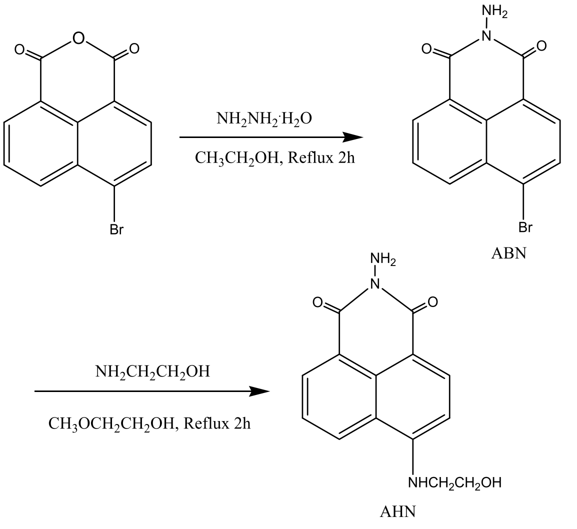

Fig. 1 Synthesis of AHN.

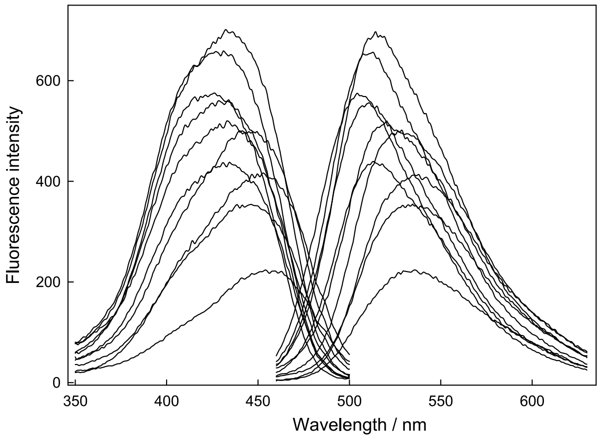

Fig. 2 Fluorescence excitation (left) and emission (right) spectra of the probe on exposure to different solvents (from top to bottom: tetrahydrofuran, ethyl acetate, dioxane, dichloromethane, acetonitrile, ethanol, acetone, glycol, methanol, water).

Table 1 Spectral properties of the probe in various solvents

Solvent	λ_{ex}/ nm	λ_{em}/ nm	$\Delta\nu$/ cm^{-1}	ε	n	Δf	I_{max}
Tetrahydrofuran	432	516	3768	7.39	1.107	0.3396	1.000
Ethyl acetate	431	513	3594	6.03	1.372	0.2000	0.935
Dioxane	427	503	3538	2.21	1.421	0.0200	0.811
Dichloromethane	432	511	3579	8.90	1.424	0.2169	0.795
Acetonitrile	435	519	3721	37.50	1.344	0.3055	0.746
Ethanol	443	527	3598	24.30	1.360	0.2890	0.717
Acetone	431	510	3709	21.49	1.360	0.2851	0.629
Glycol	448	537	3699	37.10	1.423	0.2771	0.580
Methanol	447	536	3715	32.60	1.329	0.3084	0.504
Water	454	547	3745	78.50	1.333	0.320	0.3199

reduced pressure; ethanol was at reflux over CaH_2 for 2 h, and distilled prior to use; acetonitrile was heated at reflux over P_2O_5 and distilled prior to use. Britton-Robinson (B-R) buffer solutions in the range of pH 2.0 to 11.5 were prepared by mixing appropriate amounts of phosphoric, acetic and boric acids of the same concentration (0.04 mol L^{-1}) and adjusting to desired pH with 0.20 mol L^{-1} NaOH. Outside this range, the pH was adjusted by addition of HCl or NaOH solutions. A stock solution of fluorescence probe (1.00×10^{-3} mol L^{-1}) was prepared by dissolving 27.1 mg of AHN in 100 ml of acetone. Unless otherwise stated, all other reagents were of analytical reagent grade and were used without purification or treatment. Doubly distilled water was used throughout.

Synthesis of N-amino-4-(2-hydroxyethylamino)-1,8-naphthalimide (AHN)

AHN was prepared according to the synthetic scheme shown in Fig. 1. *N*-Amino-4-bromo-1,8-naphthalic anhydride (ABN) was first synthesized according to the literature.[16] ABN was obtained as a yellow solid in a nominal yield of 88.6%: m.p. 212 - 213°C; m.s.: molecular ion peak, 292(M^+).

ABN (1.0 g) was mixed with 2 mL of 2-aminoethanol and added in small amounts to $CuSO_4·5H_2O$, then heated at reflux for 2 h in 30 mL of ethylene glycol monomethyl ether. The reaction mixture was poured into 150 mL of water, and this solution was filtrated. The solvent was evaporated under vacuum and the product was separated by column chromatography on silica gel with ethylacetate as eluent. The product was dried under vacuum at 60°C for 10 h, with a nominal yield of 76.8%: m.s.: molecular ion peak, 271(M^+). IR (KBr) ν_{max} cm^{-1}: 3315, 1664, 1583, 1387, 1248, 1068. ^{1}H-NMR (400 MHz, $CDCl_3$): δ 2.1 (s, 1H); δ 2.3 (s, 2H); δ 3.70 (m, 2H); δ 3.87 (m, 2H); δ 6.81 (d, J = 8.8 Hz, 1H); δ 7.68 (m, 1H); δ 7.73 (t, J = 5.6 Hz, 1H); δ 8.25 (d, J = 8.8 Hz, 1H); δ 8.43 (d, J = 7.2 Hz, 1H); δ 8.69 (d, J = 8.8 Hz, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 46.8; 61.1; 105.5; 114.6; 122.3; 124.5; 125.5; 127.7; 129.4; 137.2; 138.3; 137.4; 161.2; 161.1.

Instrumentation

All fluorescence measurements were conducted on a Perkin-Elmer LS-55 spectrofluorometer with both excitation and emission slits set at 5 nm; the procedure was controlled by a personal computer data processing unit. The light source was a pulsed Xe lamp. Solution pH was measured by use of a pHS-3B pH meter (Shanghai Precision & Scientific Instrument Co. Ltd., Shanghai, China). All fluorescence measurements were performed at room temperature (20°C) and atmospheric pressure.

Operating procedure

The sample solutions with different water contents in the pure organic solvents were freshly prepared prior to each measurement according to the following procedure. A 1.00-mL portion of AHN stock solution (1.00×10^{-4} mol L^{-1}) was povred into a 10-mL volumetric flask and evaporated to dryness. Then 1.00 mL of a different organic solvent was added, and a certain bulk volume of doubly distilled water was added by micro or macro transfer pipettes. A different organic solvent was then added into a 10-mL volumetric flask to the mark. The water contents in the prepared samples were standardized with the Karl Fisher method.

Results and Discussion

Spectral characteristics in different solvents and response mechanism

By observing Fig. 2 and Table 1, one can easily notice that the

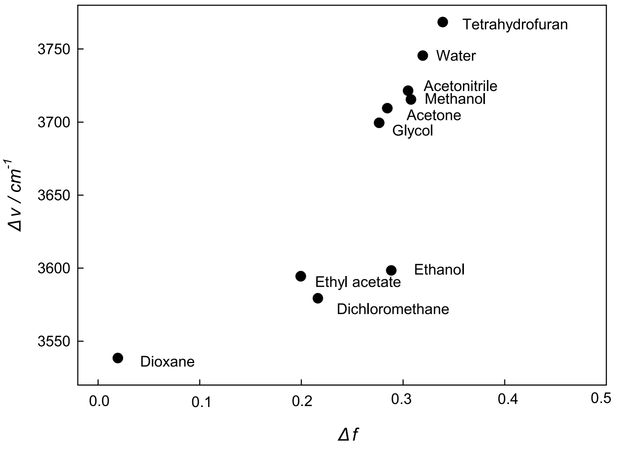

Fig. 3 The Stokes shift of the probe in various solvents.

solvent exerted an evident influence on the fluorescence intensities (from 697 in tetrahydrofuran to 224 in water) and excitation and emission spectral maxima positions (from 427 and 503 nm in dioxane to 454 and 547 nm in water). Like the behavior found in the 4-phenoxy-1,8-naphthalimide,[13] the dominant electronic transition of AHN to the first excited state should be of $\pi \rightarrow \pi^*$ character and the charge transfer process is possible due to the electron-donating ability of the naphthalimide group at C-4 position. The electronic charge distribution of $\pi \rightarrow \pi^*$ excited state is more extended than the ground state and the excited state is more polarizable. Changing from a non-polar to a polar solvent increases the solvent interaction and moves the excitation and emission spectra to longer wavelengths (red shift).[17]

In order to quantify the response of AHN to solvent polarity, we recorded the spectral properties of AHN compound in organic solvents of different polarity. The excitation wavelength (λ_{ex}), emission wavelength (λ_{em}), Stokes shift ($\Delta\nu$), dielectric constant (ε), refractive index (n), orientation polarizability (Δf) and relative fluorescence intensity (I_{max}), which was in relation to the most intensive emission measured in tetrahydrofuran ($I = 1.00$), were reported in Table 1.

Usually, general solvent effects on the fluorescence spectra can be estimated from a Lippert-Mataga equation:

$$\Delta\nu = \nu_a - \nu_f = \frac{2}{hc}\left(\frac{\varepsilon-1}{2\varepsilon+1} - \frac{n^2-1}{2n^2+1}\right)\frac{(\mu^*-\mu)^2}{a^3} + \text{constant} \quad (1)$$

Here ν_a and ν_f are wave numbers (cm^{-1}) of the absorption and emission, h is Planck's constant, c is the velocity of light, a is the radius residence cavity and μ^* and μ are excited and ground state dipole moments. $\Delta f = (\varepsilon - 1)/(2\varepsilon + 1) - (n^2 - 1)/(2n^2 + 1)$ accounts for the spectral shifts due to reorientation of the solvent molecules, called the orientation polarizability. Figure 3 shows the Stokes shift in various solvents *vs.* orientation polarizability. The Stokes shifts increase with the solvent polarity for the probe. Nevertheless, the Stokes shifts in dichloromethane and ethanol generated significant deviations from the correlations. These variances may be due to the effect of polymer backbone or the formation of hydrogen bonding. Such HBD (hydrogen-bond donor) solvents can easily form hydrogen bonds, binding to the carbonylic oxygens of the AHN molecules. These results are similar to those found for 1,8-naphthalimide derivatives.[13,18]

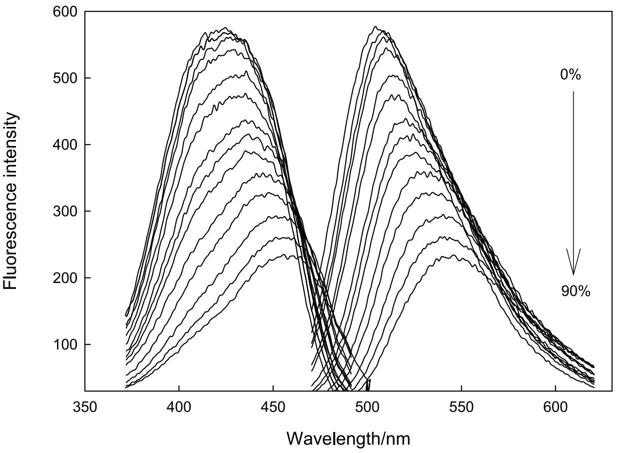

Fig. 4 Fluorescence excitation (left) and emission (right) spectra of the probe contacted with different water contents in dioxane in the range of 0 to 70% (v/v): (1) 0.00, (2) 0.20, (3) 0.60, (4) 1.00, (5) 2.00, (6) 3.00, (7) 5.00, (8) 8.00, (9) 10.0, (10) 20.0, (11) 30.0, (12) 50.0, (13) 70.0, and (14) 90.0%.

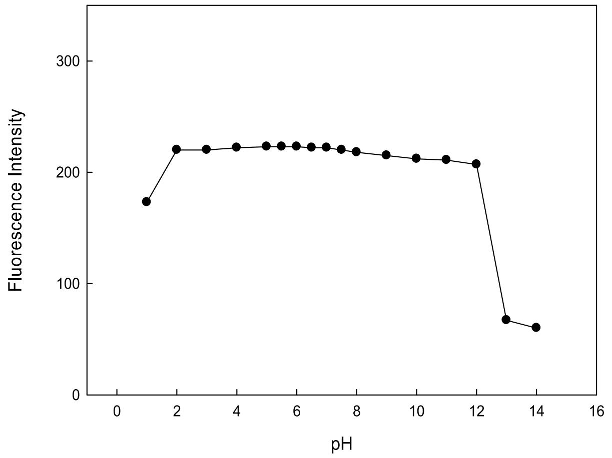

Fig. 5 Effect of pH on the AHN (1×10^{-5} mol L^{-1}) fluorescence intensity.

Spectral changes induced by addition of water content in organic solvents

The prepared fluorescence probe is really useful in the determination of water content in organic solvents. The fluorescence probe of AHN with dioxane solutions of different water content are displayed in Fig. 4. With the addition of water content from 0.00 to 90.0% (v/v), the AHN fluorescence intensity decreased dramatically and the excitation and emission peaks underwent a red shift. The excitation and emission peaks were 427 and 503 nm for AHN in dioxane, respectively. Similar behavior was observed in other organic solvents.

The effect of pH

The effect of the pH on the fluorescence intensity of the probe was investigated in aqueous solutions of different pH valves. The acidity of solutions was maintained by use of B-R buffer solutions in the range 2.00 to 11.50, and outside this range the pH was adjusted by the addition of HCl or NaOH. As shown in Fig. 5, the fluorescence intensity increased steadily when pH

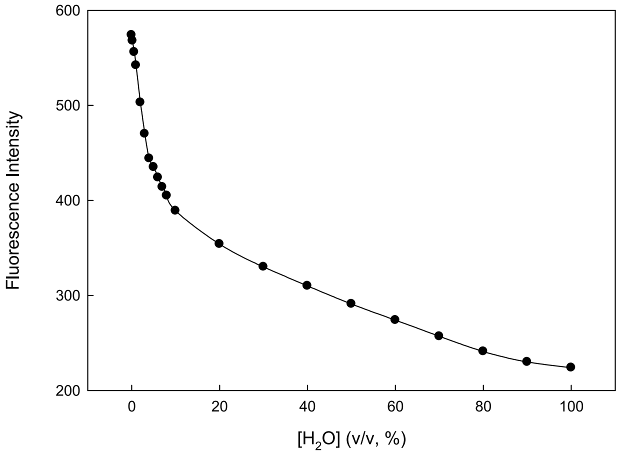

Fig. 6 The relative fluorescence intensity as a function of water content (v/v %) in the dioxane solution.

was between 2.00 and 12.00, and declined markedly when the pH was less than 2.00 or greater than 12.00.

Quantification and detection limit

The experimental response curves of the probe for water content in three different organic solvents (dioxane, acetonitrile and ethanol) were investigated. The excitation and emission wavelengths were 427 and 503 nm for dioxane, 435 and 519 nm for acetonitrile, 443 and 527 nm for ethanol, respectively. Figure 6 shows the experimental response curve of the probe for water content in dioxane solution. Similar response curves of probe in acetonitrile and ethanol were observed (not shown). The linear response of the probe covered the water content from 0.00 to 4.00% (v/v) for the three organic solutions. The following equations were obtained by fitting the experimental data obtained:

$$\text{Dioxane} \qquad I = -33.20[\mathrm{H_2O}] + 573.94 \quad (R = 0.9955) \tag{2}$$

$$\text{Acetonitrile} \qquad I = -16.77[\mathrm{H_2O}] + 519.58 \quad (R = 0.9943) \tag{3}$$

$$\text{Ethanol} \qquad I = -10.51[\mathrm{H_2O}] + 498.61 \quad (R = 0.9987) \tag{4}$$

These calibration equations can serve as the quantitative basis for the determination of trace water content in some organic solvents. The detection limits were 0.019, 0.038, and 0.060% for dioxane, acetonitrile and ethanol, respectively. The fluorescence intensity also keeps a good linearity in the range of water content from 20 to 80% for dioxane.

Conclusion

AHN, a derivative of naphthalimide, has been shown as a feasible fluorophore for the preparation of fluorescence water probes. The probe shows fluorescence intensity decreases by addition of water in organic solvents. The detection limits of the probe are sufficiently low to allow the detection of trace water. The proposed probe provides an alternative and convenient approach for determination of water content in organic solvents.

Acknowledgements

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