

A Long Wavelength Excitable Fluorophore; Chloro Phenyl Imino Propenyl Aniline (CPIPA) for Selective Sensing of Hg (II)

Kadriye Ertekin · Ozlem Oter · Mustafa Ture ·
Serpil Denizalti · Engin Cetinkaya

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Abstract In this study, a very sensitive and highly selective irreversible optical chemical sensor (optode) for mercury ions was described. The sensing scheme was based on the interaction of Hg (II) with a newly synthesized fluoroionophore; chloro phenyl imino propenyl aniline (CPIPA) in plasticized PVC membrane. The sensor membranes were tested for the determination of mercury ion in aqueous solutions by batch and flow-through methods. The optodes allow determination of Hg (II) in the working range of 1.0×10^{-9} – 1.0×10^{-5} M with a detection limit of 4.3 ppb. The sensor exhibited excellent selectivity for Hg (II) with respect to several common alkali, alkaline earth and transition metal ions. The association constant of the 1:1 complex formation for Hg (II) was found to be $K_a = 1.86 \times 10^5 \text{ M}^{-1}$. The CPIPA exhibited high fluorescence quantum yield, long excitation and emission wavelength and high Stokes' shift values in the solid matrix which makes it compatible with solid state optics.

Keywords Optical chemical sensor · Hg(II) sensing · Mercury sensor · Spectrofluorometry

Introduction

Pollution of mercury and mercury compounds is a major problem in the vast decays due to their increasing utilization in industry and agriculture. They are notable for being toxic elements in all lists of priority pollutants, in different regulations and guidelines and the accurate analysis of them is of great importance. The methods for the determination of trace levels of different mercury species include atomic absorption or emission spectrometry, mass spectrometry, voltammetry and chromatography with spectrometric detectors [1, 2]. However, these methods are not suitable for field screening or on-site assay. So, the design of new, sensitive and accurate sensors allowing field analysis for mercury ions is of great interest in different areas. In these designs, different dyes have been used for spectral Hg (II) detection either in free or in immobilized form [3–6] Ros-Lis and co-workers performed selective signaling of Hg (II) in aqueous media at nanomolar levels employing a squaraine-based chemosensor [3].

Chiang et al. reported an oligonucleotide-based fluorescence probe for sensitive and selective detection of Hg (II) in aqueous solution. They used TOTO-3 fluorescent dye and the polythymine oligonucleotide T-33-, together. The fluorescence intensity increased in the presence of Hg (II) [4]. Neutral ionophores such as dithiocarbamates were employed in immobilized form [5]. Murkovic et al. used a lipophilic borate salt as a reagent for Hg (II), along with an amphiphilic carbocyanine dye as the optical transducer in

K. Ertekin (✉) · O. Oter · M. Ture
Faculty of Arts and Sciences, Department of Chemistry,
University of Dokuz Eylul,
35160 Tinaztepe,
Izmir, Turkey
e-mail: kadriye.ertekin@deu.edu.tr

S. Denizalti · E. Cetinkaya
Faculty of Sciences, Department of Chemistry, University of Ege,
35100 Bornova,
Izmir, Turkey

Present Address:
K. Ertekin
Dokuz Eylul Universitesi Fen-Edebiyat Fakultesi, Kimya Bolumu,
35160 Tinaztepe Buca,
Izmir, Turkey

plasticized PVC [6]. They reported 30 min response time for 100 nM concentration levels. Coskun et al. offered bis (2-pyridyl)-substituted borotriazaindacene dye as an NIR-emitting chemosensor for Hg (II) [7]. They performed their measurements in acetonitrile for 1–20 μM concentration range of Hg (II). Rhodamine B molecules were employed together with gold-nanoparticles (AuNP) for detecting mercury ions in aqueous solutions [8]. They improved the selectivity of the probe by modifying the AuNP surfaces with thiol ligands. Coskun and Akkaya reported a solution phase ratiometric fluorescent chemosensor for Hg (II) employing a boradiazaindacene dyad for the concentration range of 0–25 μM Hg (II) [9]. Yang et al. offered a quenching based sensor, working in the concentration range of 4.0×10^{-8} – 4.0×10^{-6} mol L⁻¹ [10] (See Table 1). Guo and coworkers designed an optical sensor membrane by using tetraphenylporphyrin in an organically modified sol–gel matrix. They reported a detection limit of 3.6×10^{-6} mol L⁻¹ for Hg (II) [11]. Kuswandi and Narayanaswamy designed an optode for mercury ion based on the reagent safranin-iodide solution trapped behind the Nafion membrane. The optode had a detection limit of 5 μM toward Hg (II) ions, with only Ag (I) as the main interferent [12]. Nolan and Lippard synthesized a Hg(II) binding, red-emitting, water soluble chemosensor. This sensor exhibited both turn-on and ratiometric fluorescence responses to Hg (II) with a high degree of selectivity [13]. Yusof et al. designed an optical test strip based on the use of Br-PADAP in sol–gel thin film. Interference studies showed that Al (III), Co (II) and Ni (II) significantly interfered during the determination [14]. Dolci et al. presented a solid-state chemosensor for mercuric ions, at micromolar concentration, working in water. The sensor exhibited a

remarkable selectivity for Hg²⁺ over Cu²⁺, Cd²⁺, Pb²⁺ and Zn²⁺ ions [15].

Shamsipur et al. employed dansylamidopropyl pendant arm derivative of 1-aza-4,10-dithia-7-oxacyclododecane in plasticized PVC membrane for Hg (II) sensing. The response of the sensor was based on the fluorescence quenching of the dye at pH 3.3. The proposed sensor responded to Hg (II) with a moderate relative signal change in the concentration range of 1.0×10^{-4} – 5.0×10^{-12} M. However, they reported a 14% signal drift after 6 weeks in the fluorescence intensity [16]. Cano-Raya et al. designed a new optical disposable sensor to determine mercury with a porphyrin proton-selective fluoroionophore [17].

Some of the mercury sensitive molecular probes suffer from lack of sensitivity [11, 12, 18, 19], selectivity [6, 12, 14, 17] or limited storage time [16] which are desirable properties for a chemo-sensor (See Table 1). Therefore, search for new sensing materials that can respond selectively and sensitively to the analyte is still necessary. Here we present the first application of the stable and long wavelength excitable fluorophore; chloro phenyl imino propenyl aniline (CPIPA) dye having high quantum yield for highly selective and sensitive determination of Hg (II) at nanomolar concentration levels. The dynamic range of the proposed sensor has been determined to be between 10^{-9} and 10^{-5} M Hg (II), i.e. from nanomolar to micromolar levels which may make this technique alternative to cold-vapor atomic absorption spectrometry or flame emission methods and was sensitive than most of the sensors reported in the literature [6, 10–12, 14, 17–19]. The offered dye has the advantages of long term photostability, high relative signal change and a large working range.

Table 1 Comparison of the offered method with present literature information regarding spectroscopic determination of mercury ions

Lit.	Optode	Linear Conc. Range (mol L ⁻¹)	Limit of detection (mol L ⁻¹).	Reversibility	Selectivity or main interferents
[6]	theamphiphilic oxacarbocyaninedye (DiO&(3)	40–650 ppb	20 ppb	irreversible	Zn(II),Ag(I)
[10]	tetra (<i>p</i> -dimethylaminophenyl) porphyrin	4.0×10^{-8} – 4.0×10^{-6}	8.0×10^{-9}	reversible	selective
[11]	(5,10,15,20-tetraphenylporphyrin, H2tp)	5.0×10^{-6} – 1.0×10^{-4}	3.6×10^{-6}	reversible	selective
[12]	Safranin-iodide solution	0 – 5×10^{-5}	5×10^{-6}	irreversible	Ag(I)
[14]	2-(5-bromo-2-pyridilazo)-5-diethylamino	0.5–2.5 ppm	6.63 ppb	irreversible	Al(III), Co(II) Ni(II)
[17]	5,10,15,20-tetraphenyl-21H,23H-porphine	1.0×10^{-7} – 2.6×10^{-6}	25 ppb	irreversible	Cl-
[18]	hexathiacyclooctadecane	2.1×10^{-7} – 1.2×10^{-4}	2.0×10^{-7}	irreversible	selective
[19]	4-hydroxy salophen	1.0×10^{-6} – 1.0×10^{-2}	1.3×10^{-7}	reversible	selective
Ours	Chloro phenyl imino propenyl aniline	1.0×10^{-9} – 1.0×10^{-5}	2.2×10^{-10} (4.3 ppb)	irreversible	Fe(III)

Experimental

Materials

The polymer membrane components, polyvinyl chloride (PVC) (high molecular weight) potassium tetrakis-(chlorophenyl) borate (PTCPB), and the plasticizer, bis-(2-ethylhexyl) phthalate (DOP), were obtained from Fluka. Absolute ethanol (EtOH), tetrahydrofuran (THF) and nitric acid (HNO₃) were of analytical grade. Solvents for the spectroscopic studies were used without further purification. Millipore ultra pure water was used throughout the studies.

Buffer components were of analytical grade (Merck and Fluka). For metal ion tests, AAS standard solutions (Zn²⁺, Sn²⁺, Ca²⁺, Bi³⁺, Na⁺, Ni²⁺, Co²⁺, Cu²⁺, Pb²⁺, Al³⁺, Cr³⁺, Mn²⁺, K⁺ and Fe³⁺, 1,000 mg/L, Merck) were employed. All standards were diluted with 0.01 M acetic acid/acetate buffer of pH 4.0. Solutions of Hg (I) and Hg (II) were prepared from the respective metal nitrates. The pH values of the solutions were checked using a digital pH meter (Orion) calibrated with standard buffer solutions of Merck. All of the experiments were carried out at room temperature; 25 °C. The synthesis of CPIPA dye (Chloro Phenyl Imino Propenyl Aniline) has been performed in laboratories of University of Ege and published earlier [20]. Schematic structure of the employed dye molecule is shown in Fig. 1.

Apparatus

Absorption spectra were recorded using a Shimadzu UV-1601 UV-Vis spectrophotometer. Steady-state fluorescence emission and excitation spectra were measured using Varian Cary Eclipse Spectrofluorometer with a Xenon flash lamp as the light source. Metal response measurements were carried out with fiber optic probe (2 m long) and solid sample tip accessories constructed on the spectrofluorometer. For instrumental control, data acquisition and processing the software package of the

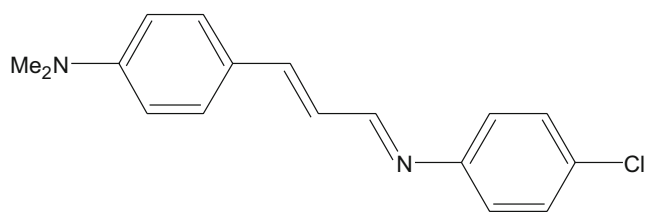


Fig. 1 Structure of the CPIPA (Chloro Phenyl Imino Propenyl Aniline) or 4-((1E, 3E)-3-[(4-chlorophenyl)imino]-1-propenyl)-N,N-dimethylaniline dye

spectrofluorometer was used. The tip of the bifurcated fiber optic probe was interfaced with a sensing film in a buffer containing homemade 300 μ L flow cell. The flow cell was equipped with a four channel Ismatec Reglo Analog peristaltic pump. Flow rate of the peristaltic pump was kept at 2.4 mL min⁻¹. Analyte solutions or buffers were transported via tygon tubing of 2.06 mm i.d. Instrumental set-up used for dye-doped thin film measurements was shown in Fig. 2.

Cocktail preparation protocols and the spectral characterization

The optode membranes were prepared to contain 120 mg of PVC, 240 mg of plasticizer (DOP), 1.17 mg of CPIPA dye (≈ 2.5 mmol dye kg⁻¹ polymer), two fold of potassium tetrakis(4-chlorophenyl) borate (in terms of mole number) and 1.5 mL of THF. The amount of potassium tetrakis(4-chlorophenyl) borate and the plasticizer was optimized with some preliminary experiments. The resulting cocktails were spread onto a 125 μ m polyester support (Mylar TM type) in order to obtain the sensing films. The films were kept in a desiccator in order to avoid the damage from the ambient air of the laboratory. The film thicknesses of the sensing slides were measured with Tencor Alpha Step 500 prophyliometer and were found to be 6.12 ± 0.071 μ m.

Results and discussion

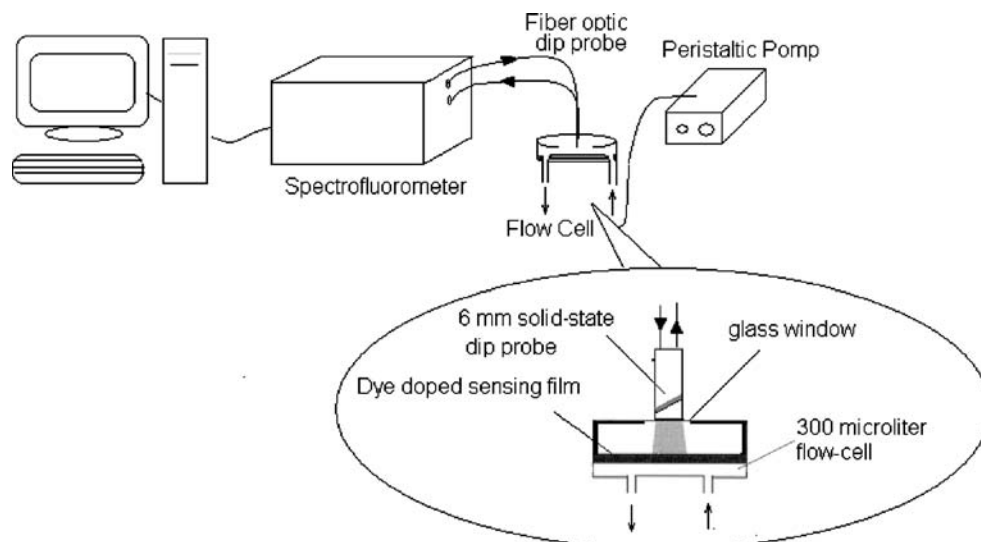
Choice of dye

In this work, sensitivity of the CPIPA dye to Hg (I), Hg (II) and many other polyvalent metal ions was investigated both in EtOH and plasticized PVC. Spectral characterization data of CPIPA in the solvents of EtOH, DCM, THF and in solid matrix of PVC was published earlier [20].

The dye was chosen due to the high molar extinction coefficients ($\lambda_{\max} = 400$ nm, $\epsilon = 28,000$ M⁻¹ cm⁻¹ in EtOH and $\lambda_{\max} = 547$ nm, $\epsilon = 56,000$ M⁻¹ cm⁻¹ in PVC) and quantum yield values in the employed matrices. The quantum yield in PVC matrix is 0.37 for the CPIPA dye in reference to Rose Bengal. The long wavelength excitation and emission characteristics ($\lambda_{\max}^{\text{ex}} = 556$ nm and $\lambda_{\max}^{\text{em}} = 593$ nm) make the dye compatible with the solid-state optical components (in particular with LED's emitting in the wavelength range of 550–570 nm and fiber optics) which can be useful in construction of inexpensive and field available instrumentation.

The CPIPA in PVC exhibited quite good long term photostability for at least 8 months. The tested signal drift

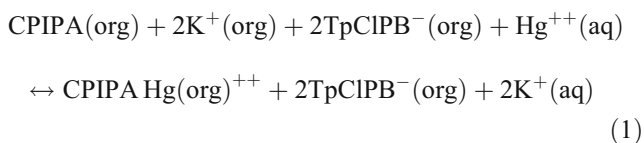
Fig. 2 Instrumental set-up used for dye-doped thin film measurements



was found to be only 2% which is another advantage of the employed dye over most of the existing mercury sensitive fluorescent probes [20].

Response to Hg (II) ions

When doped into plasticized PVC along with the anionic additive; potassium tetrakis-(4-chlorophenyl) borate; the CPIPA dye becomes a Hg (II) selective probe. In this system, Hg (II) ions are selectively extracted into the optode membrane by the anionic additive meanwhile potassium ions diffuse from the membrane into the aqueous phase due to the mechanism of ion-exchange. Physical aspect of the response mechanism of CPIPA dye can be explained by the following ion-exchange pathway.



The steady state fluorescence emission spectra of PVC-doped CPIPA were recorded before and after exposure to Hg (II) solution. The sensing membrane was also exposed to a flow of solutions containing Hg (II) in different concentrations in a flow cell. The excitation wavelength was set at 556 nm and the fluorescence intensity at 593 nm was continuously recorded. Continuous and irreversible decrease in fluorescence on exposure to Hg (II) was observed, so that the relative change of fluorescence measured after a certain period of exposure (10 min) served as the analytical signal. Figure 3, I and II reveal emission based response of the PVC doped CPIPA dye to different

concentrations of Hg (II) and related calibration plot at pH=4.0. The sensor membranes allow for the determination of Hg(II) at nano molar concentration levels in the linear concentration range of 1.0×10^{-9} to 1.0×10^{-5} M Hg (II) with a regression coefficient; $r^2=0.9893$. The detection limit of the sensor membrane (the concentration of the metal ion giving a signal equal to the blank signal

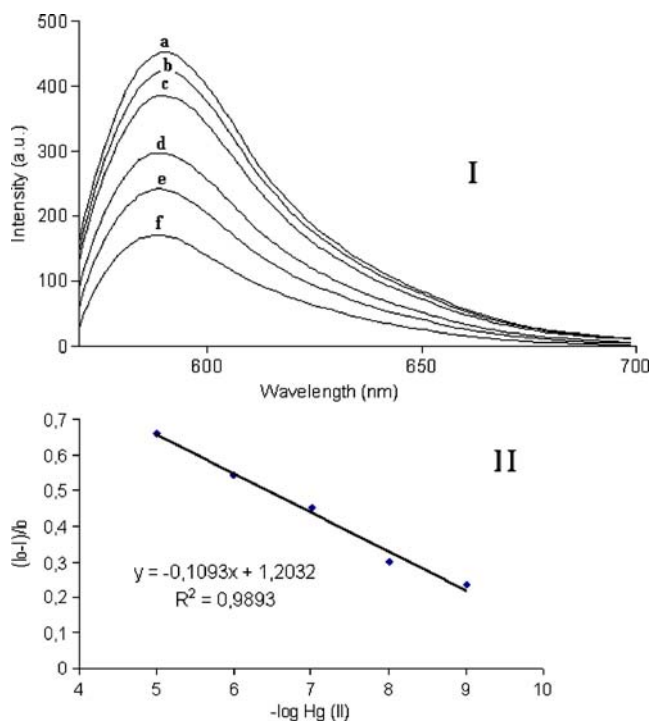


Fig. 3 I: Emission based response of the CPIPA doped PVC membrane to different concentrations of Hg (II). II: Calibration curve at pH=4.0 (λ_{ex} : 556 nm, a: buffer, b: 10^{-9} , c: 10^{-8} , d: 10^{-7} , e: 10^{-6} , f: 10^{-5} mol L $^{-1}$)

plus three standard deviation of the blank) was found to be $<10^{-9}$ mol L $^{-1}$ (4.3 ppb), which is sufficiently low for the detection of the sub-nanomolar concentration range of Hg (II) ions found in many chemical and biological systems. Table 1 gives a comparison of our sensor with the present fluorescence based Hg (II) sensors in terms of working range, limit of detection and selectivity.

Effect of pH and ionic strength on Hg (II) response

The pH dependency of the metal free form of the CPIPA dye was reported earlier. The pKa value of 10.25 makes the dye stable in the acidic and near neutral region of the pH scale [20].

The effect of pH on the complexation of CPIPA with Hg (II) was investigated between pH 4.0–7.0 at fixed metal concentration; 10^{-3} mol L $^{-1}$. As can be seen from Fig. 4, the relative signal change; $(I_0-I)/I_0$ produced by the Hg (II) ions was high enough and stable in the pH range of 4.0–6.0. According to the chemical equilibrium software programme (Visual MINTEQ), the distribution of the chemical species in natural waters at pH 4.0 is as follows: Hg(OH) $_2$ (93.067%), Hg $^{2+}$ (1,424%) and HgOH $^+$ (5.508%). However, Hg exists in solution in anion form because of complexation with acetate component of the employed buffer. When pH was higher than 6.0, the relative signal change has dramatically decreased in BES buffered solutions due to the formation of Hg(OH) $_2$ to a large extent, which implies reduced complexation of CPIPA with Hg (II). Therefore, for further studies pH 4.0 was chosen.

Response of the sensor membrane was also tested by making the ionic strength of the buffer solution 135 mM with NaCl. After exposure to different concentrations of saline Hg (II) solutions, the relative signal change of the emission spectrum of the sensor membrane was dropped from 69% to 50%.

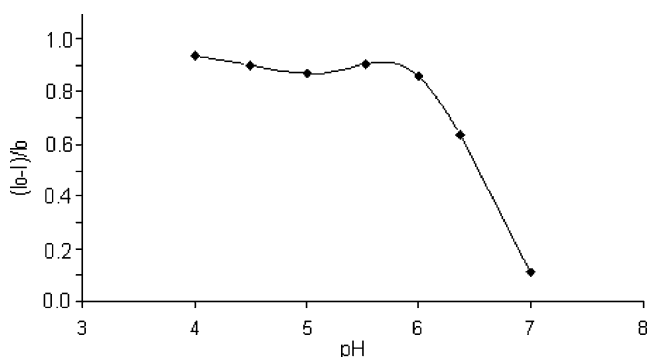


Fig. 4 The membrane response upon exposure to fixed concentrations (10^{-3} mol L $^{-1}$) of Hg (II) at different pH values

Mechanism of quenching and complex formation between CPIPA and Hg(II)

In many instances the fluorophore can be quenched both by collisions and by complex formation. The intensity based data (I_0/I) or $(I_0-I)/I_0$ exhibiting an upward-concave curvature toward the y-axis is the evidence of combined quenching both by collisions (dynamic quenching) and by complex formation (static quenching) with the same quencher [21, 22].

In case of dynamic quenching because the collision between the quencher and the fluorophore affects only the excited state of the fluorophore, no changes in the absorption spectrum are expected. On the contrary, the formation of ground-state complex in static quenching will perturb the absorption spectra of the fluorophore. Thus, by careful examination of the absorption spectrum, one can attempt to distinguish static and dynamic quenching.

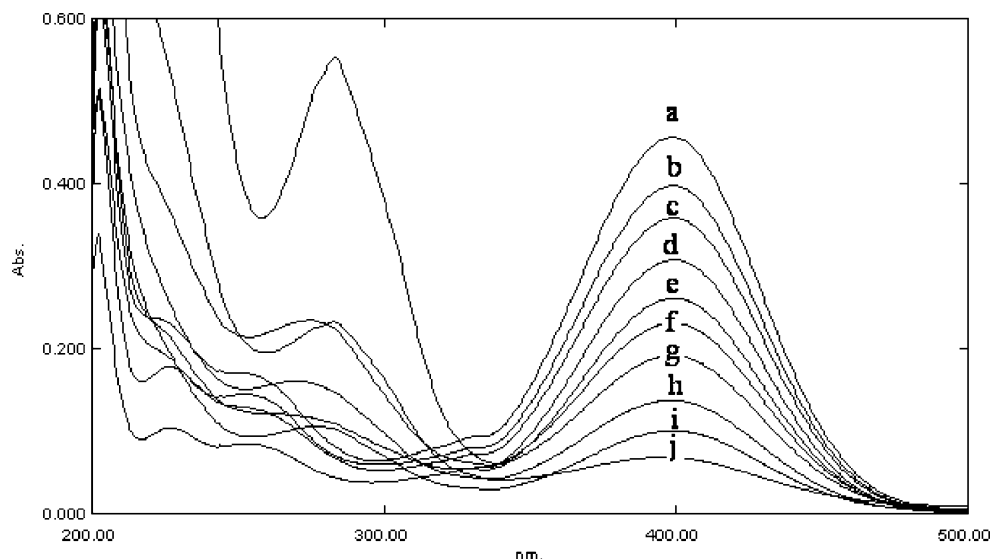
Figure 5 shows the absorption spectrum of CPIPA dye in EtOH solution in the absence and presence of Hg (II) ions (See spectrum “a” and others). The absorption band at 289 nm exhibited an increase in signal intensity upon exposure to Hg (II) and an accompanying spectral shift. Variation of absorption spectra and an accompanying discernible color change of CPIPA dye from pink to colorless reveal the formation of a non-fluorescent complex with Hg (II) in the ground state. When the linear shape of the fluorescence intensity based response and absorption spectroscopy related data were evaluated together, the quenching mechanism between CPIPA and Hg (II) can be concluded as “static quenching”.

The data in Fig. 5 was also used to determine the association constant (K_a) of the CPIPA-Hg complex [23]. The K_a was calculated by plotting $(A-A_0)/(A_x-A_0)$ versus $[Hg^{2+}]$, where the slope is the association constant, A_x was the measured absorbance intensity at any metal concentration around mid point, A_0 and A are the absorption intensities of the film when indicator dye in its unloaded and loaded forms, respectively. For our data such a plot is linear over the whole concentration range and the association constant was found to be $K_a=1.86 \times 10^5$ in ethanol. The plot can be defined by the equation; $y = 185507x - 0.1353$; and regression coefficient; $R^2 = 0.9916$ (See Fig. 6). Metal ion binding constant (K_a) value is dependent on many factors, including pH, temperature, viscosity and the presence of other ions. The response may vary significantly in complex environments such as seawater or blood.

Complex stoichiometry

Information on the stoichiometry of the complex was obtained from the continuous variation method (Job’s

Fig. 5 Variations in the absorption spectra during the complex formation of CPIPA dye ($1 \times 10^{-5} \text{ mol l}^{-1}$), with the Hg (II) ions in ethanol. *a*: CPIPA dye solution, *b*: 1×10^{-6} , *c*: 2×10^{-6} , *d*: 3×10^{-6} , *e*: 4×10^{-6} , *f*: 5×10^{-6} , *g*: 6×10^{-6} , *h*: 7×10^{-6} , *i*: 8×10^{-6} , *j*: $9 \times 10^{-6} \text{ mol L}^{-1}$ Hg (II)



method). In Job's method different amounts of stock solutions of metal and ligand are mixed varying the mole ratio of reactants.

The principle of the method is as follows: the absorbance (Abs) is measured for a series of solutions containing the ligand and the cation such that the sum of the total concentrations of ligand and cation is constant:

$$C_L + C_M = C = \text{constant}$$

The position of the maximum of absorbance (A_{max}) is then related to the ratio M/L, as shown below.

$$X = \frac{C_M}{C_M + C_L}$$

Figure 7; Job's plot for the employed dye and metal; reveals complex stoichiometry between CPIPA and Hg (II) in ethanol. From the Job's plot, stoichiometry of the metal–ligand complex of Hg(II)–CPIPA was found to be 1:1 at the employed metal or dye concentration; $1 \times 10^{-5} \text{ mol L}^{-1}$.

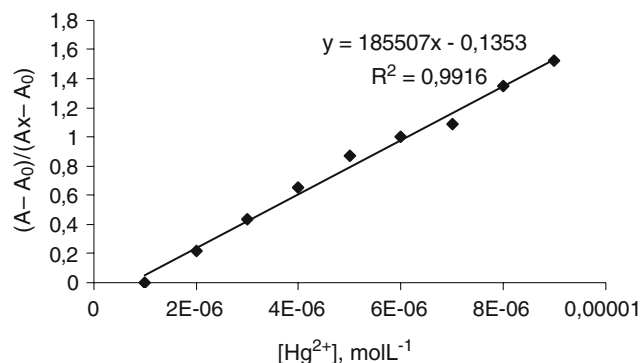


Fig. 6 Absorption based relative signal change $[(A-A_0)/(A_x-A_0)]$ of CPIPA dye in EtOH versus $[\text{Hg}^{2+}]$ concentration

Selectivity studies

Measuring Hg (II) concentrations in blood, groundwater or environmental samples with indicators becomes difficult due to competitive binding of Ca^{2+} , K^+ , Mg^{2+} and other cations. In order to examine the response of the CPIPA to possible interfering cations, the sensor films were treated with 10^{-3} M concentrations of Y^{3+} , Mo^{3+} , Sb^{3+} , Zn^{2+} , Sn^{2+} , Ca^{2+} , Bi^{3+} , Na^+ , Ni^{2+} , Co^{2+} , Cu^{2+} , Pb^{2+} , Al^{3+} , Cr^{3+} , Mn^{2+} , K^+ , Hg^+ , and Fe^{3+} ions in pH 4.0 acetic acid/acetate buffer solutions. From Fig. 8, it can be concluded that, the sensing membrane is capable of determining mercury ions (Hg^{2+}) with a high selectivity over Y^{3+} , Mo^{3+} , Sb^{3+} , Zn^{2+} , Sn^{2+} , Ca^{2+} , Bi^{3+} , Na^+ , Ni^{2+} , Co^{2+} , Cu^{2+} , Pb^{2+} , Al^{3+} , Cr^{3+} , Mn^{2+} , K^+ . However, the response to Fe^{3+} is markedly different from the response to other metal ions and fluorescence of CPIPA is affected appreciably by Fe^{3+} at concentrations of $\geq 10^{-3} \text{ mol L}^{-1}$.

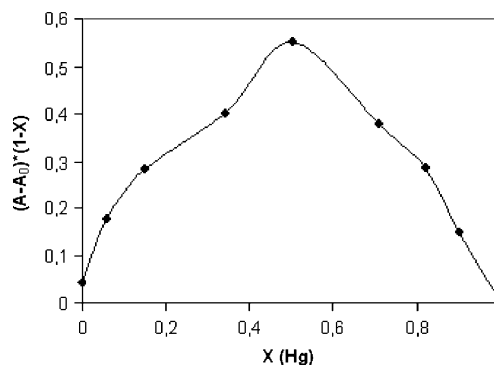
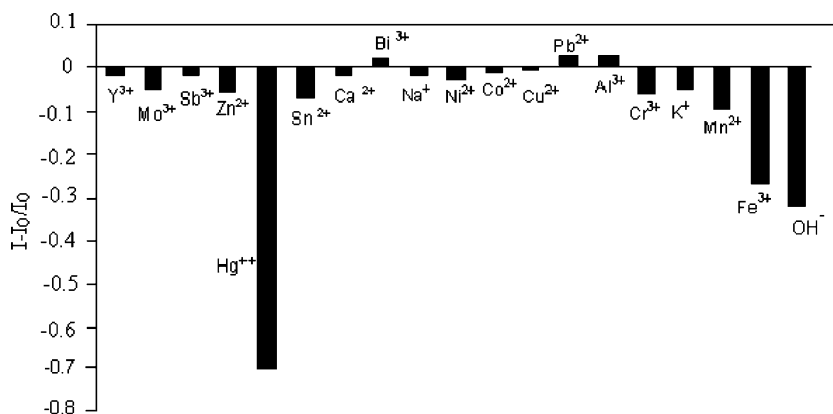


Fig. 7 Job's plot for complex formation of CPIPA with Hg^{2+} in ethanol ($\lambda_{\text{abs}}=284 \text{ nm}$, $C_{\text{sum}}=1 \times 10^{-5} \text{ mol L}^{-1}$). Absorption based data was extracted from short-wavelength maximum of the absorption spectra of Hg^{2+} complex

Fig. 8 Metal-ion response of CPIPA for 10^{-3} mol L⁻¹ concentrations of Y³⁺, Mo³⁺, Sb³⁺, Zn²⁺, Sn²⁺, Ca²⁺, Bi³⁺, Na⁺, Ni²⁺, Co²⁺, Cu²⁺, Pb²⁺, Al³⁺, Cr³⁺, Mn²⁺, K⁺ and Fe³⁺ ions in pH 5.0 acetic acid/acetate buffer. Results were plotted as relative fluorescence changes; $(I-I_0)/I_0$



The fluorescence was dramatically quenched in the presence of Hg (II) at 590 nm exhibiting a relative signal change (RSC) ratio of 72%. Except that of Fe (III), other alkali, alkaline earth, and transition metal ions produced insignificant responses where RSCs were between 2–10%. Under the employed conditions, Fe³⁺ ions induced a somewhat quenched fluorescence intensity (RSC=28%) and appeared to interfere with the Hg²⁺-selective responses of CPIPA.

A selectivity comparison of Hg (I) over Hg (II) in separate solutions was also investigated. The response of the CPIPA dye upon interaction with Hg (I) and Hg (II) was measured at fixed Hg (I) or Hg (II) concentrations at pH 4.0. The steady state fluorescence emission spectra were recorded before and after exposure to 10^{-3} mol L⁻¹ buffered Hg (I) and Hg (II) solutions (See Fig. 9, I and II). The CPIPA dye was found to be effectual in selective binding of both Hg (I) and Hg (II) with respect to other possible interferants. However, a speciation was not possible.

Response time and regeneration

The response time (τ_{90}) for sensing slides was found to be approximately 10–20 min in buffer solutions. However, the

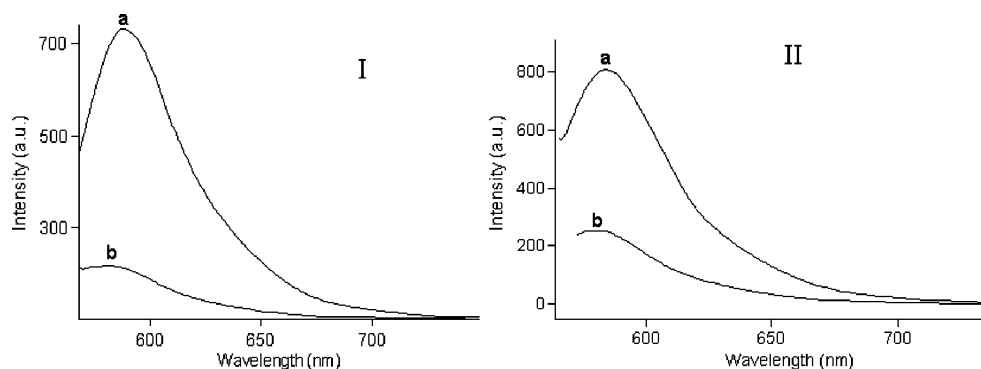
upper limit of response time can be improved. In our case, film thickness is approximately 6 μ m, which can be reduced and shorter response times can be reached.

The reversibility of the optode was checked by washing the used optodes with 0.1 mol L⁻¹ EDTA solutions at different pHs. 0.1 mol L⁻¹ HCl, HNO₃ and potassium tetrakis-(chlorophenyl) borate solutions were also tested for regeneration purposes. The results showed that the optodes could not be regenerated and each optode can be used as a disposable sensor for single shot analysis.

Conclusion

The work described here demonstrates a selective fluorescence based molecular probe for Hg (II) ions. To our knowledge the CPIPA dye was used for the first time as a fluoroionophore in the optical mercury sensing. The quantum yield of the dye in PVC is reasonably high (0.37) and could be excited at 556 nm with commercially available cheap LEDs. The CPIPA dye can be used at pH 4.0 for quantitative determination of Hg (II) in the concentration range of 1.0×10^{-9} to 1.0×10^{-5} mol L⁻¹. A quite good LOD (4.3 ppb) was reached.

Fig. 9 The response of the CPIPA dye upon interaction with Hg (I) and Hg (II) at fixed Hg (I) or Hg (II) concentrations (Hg (I) or Hg (II)= 10^{-3} mol L⁻¹, pH=4.0)



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References

1. Costley CT, Mossop KF, Dean JR, Garden LM, Marshall J, Carroll J (2000) Determination of mercury in environmental and biological samples using pyrolysis atomic absorption spectrometry with gold amalgamation. *Anal Chim Acta* 405:179–183
2. Clevenger WL, Smith BW, Winefordner JD (1997) Trace determination of mercury: a review. *Crit Rev Anal Chem* 27:1–26
3. Ros-Lis JV, Martinez R, Rurack K, Sancenon F, Soto J, Spieles M (2004) Highly selective chromogenic signaling of Hg²⁺ in aqueous media at nanomolar levels employing a squaraine-based reporter. *Inorg Chem* 43(17):5183
4. Chiang CK, Huang CC, Liu CW, Chang HT (2008) Oligonucleotide-based fluorescence probe for sensitive and selective detection of mercury(II) in aqueous solution. *Anal Chem* 80(10):3716–3721
5. Lerchi M, Reitter E, Simon W, Pretsch B, Chowdhury DA, Kamata S (1994) Bulk optodes based on neutral dithiocarbamate ionophores with high selectivity and sensitivity for silver and mercury cations. *Anal Chem* 66:1713–1717
6. Murkovic I, Wolfbeis OS (1997) Fluorescence-based sensor membrane for mercury (II) detection. *Sens Actuators B* 39:246–251
7. Coskun A, Yilmaz MD, Akkaya EU (2007) Bis(2-pyridyl)-substituted boratriazaindacene as an NIR-emitting chemosensor for Hg (II). *Org Lett* 9:607–609
8. Huang CC, Chang HT (2006) Selective gold-nanoparticle-based Turn-On fluorescent sensors for detection of mercury (II) in aqueous solution. *Anal Chem* 78:8332–8338
9. Coskun A, Akkaya EU (2006) Signal ratio amplification via modulation of resonance energy transfer: proof of principle in a novel emission ratiometric Hg(II) sensor. *J Am Chem Soc* 128:14474–14475
10. Yang Y, Jiang J, Shen G, Yu R (2009) An optical sensor for mercury ion based on the fluorescence quenching of tetra(*p*-dimethylaminophenyl)porphyrin. *Anal Chim Acta* 636:83–88
11. Guo L, Zhang W, Xie Z, Lin X, Chen G (2006) An organically modified sol–gel membrane for detection of mercury ions by using 5, 10, 15, 20-tetraphenylporphyrin as a fluorescence indicator. *Sens Actuators B* 119:209–214
12. Kuswandi B, Narayanaswamy R (2001) Selective pool optode for mercury ion sensing in aqueous solution. *Sens Actuators B* 74:131–137
13. Nolan EM, Lippard SJ (2007) Turn-on and ratiometric mercury sensing in water with a red-emitting probe. *J Am Chem Soc* 129:5910–5918
14. Yusof NA, Kadir WARWA (2009) Optical test strip for trace Hg (II) based on doped sol–gel film. *Spectrochim Acta Part A* 72:32–35
15. Dolci LS, Marzocchi E, Montalti M, Prodi L, Monti D, Natale CD, D'Amico A, Paolesse R (2006) Amphiphilic porphyrin film on glass as a simple and selective solid-state chemosensor for aqueous Hg²⁺. *Biosens Bioelectron* 22:399–404
16. Shamsipur M, Hosseini M, Alizadeh K, Alizadeh N, Yari A, Caltagirone C, Lippolis V (2005) Novel fluorimetric bulk optode membrane based on a dansylamidopropyl pendant arm derivative of 1-aza-4, 10-dithia-7-oxacyclododecane ([12]aneNS2O) for selective subnanomolar detection of Hg(II) ions. *Anal Chim Acta* 533(1):17–24
17. Cano-Raya C, Fernandez-Ramos MD, Gomez-Sanchez J, Capitan-Vallvey LF (2006) Irreversible optical sensor for mercury determination based on tetraarylborate decomposition. *Sens Actuators B* 117:135–142
18. Ensafi AA, Fouladgar M (2009) A sensitive and selective bulk optode for determination of Hg(II) based on hexathiacyclooctadecane and chromoionophore V. *Sens Actuators B: Chemical*. doi:10.1016/j.snb.2008.12.029
19. Ensafi AA, Far AK, Meghdadi S (2008) Highly selective optical sensor for mercury assay based on covalent immobilization of 4-hydroxy salophen on a triacetylcellulose membrane. *Sens Actuators B* 133:84–90
20. Derinkuyu S, Ertekin K, Oter O, Denizalti S, Cetinkaya E (2007) Fiber optic pH sensing with long wavelength excitable schiff bases in the pH range of 7.0–12.0. *Anal Chim Acta* 588:42–49
21. Oter O, Ertekin K, Kirilmis C, Koca M (2007) Spectral characterization of a newly synthesized fluorescent semicarbazone derivative and its usage as a selective fiber optic sensor for copper (II). *Anal Chim Acta* 584:308–314
22. Lakowicz JR (1999) Principles of fluorescence spectroscopy. Kluwer Academic/Plenum, New York
23. Connors KA (1987) Binding constants: the measurement of molecular complex stability. Wiley, New York