Microchemical Journal 106 (2013) 27–33

Contents lists available at SciVerse ScienceDirect

Microchemical Journal

journal homepage: www.elsevier.com/locate/microc

A novel colorimetric and fluorescent sensor for fluoride and pyrophosphate based on fluorenone signaling units

T. Daniel Thangadurai [a,c], Chang Jun Lee [b], Su Hee Jeong [a], Sunghak Yoon [a], Yeong Geon Seo [d], Yong-Ill Lee [a,*]

[a] Anastro Laboratory, Department of Chemistry, Changwon National University, Changwon 641–773, Republic of Korea
[b] Department of Chemistry, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599, USA
[c] Department of Bio and Nano Chemistry, Kookmin University, 861–1 Jeongneung-dong, Seongbuk-gu, Seoul 136–702, Republic of Korea
[d] Department of Computer Science, Gyeongsang National University, Jinju 660–701, Republic of Korea

ARTICLE INFO

Article history:
Received 25 April 2012
Accepted 29 April 2012
Available online 3 May 2012

Keywords:
Fluorenone
Urea
Anions
Naked-eye detection
Fluorescence quenching
Time dependant density functional theory

ABSTRACT

In this study, a novel chromogenic receptor, 1-(naphthalen-1-yl)-3-(9-oxo-9*H*-fluoren-1-yl)urea (**1**), utilizing fluorenone and naphthalene moieties as signaling groups was designed and synthesized. The interaction and colorimetric sensing properties of receptor **1** with different anions were investigated by the naked eye, as well as UV-visible and fluorescence spectroscopy. The addition of 100 equiv. of fluoride and pyrophosphate as tetrabutylammonium salts to 1.25×10^{-4} M CH_3CN:DMSO (9:1, v/v) solution mixture of receptor **1** produced a wine-red color. The oxoanions and a variety of other anions were not capable of producing any significant color change with receptor **1** under similar experimental conditions. The corresponding UV-vis measurements showed a bathochromic shift of the 395 nm band of receptor **1** to ~500 nm for fluoride and pyrophosphate. Fluorescence emission changes indicate clearly that receptor **1** behaves like an ideal photo-induced electron transfer (PET) sensor upon complexation with anions. The limit of detection (LOD) of the sensor **1** is calculated to be ca. 250 and 110 nM for F^- and $HP_2O_7^{3-}$, respectively. The ^1H NMR titration studies shed further light on their mode of binding with receptor **1**. The quantum mechanical calculations through time dependant density functional theory (TD-DFT) using basis set B3P86/TZVP support our experimental findings with a good agreement.

1. Introduction

Due to many possible applications in analytical chemistry and biomedical research, a considerable amount of research attention has focused on the design of receptors with the ability to selectively bind cation, neutral and anionic species [1–3]. In particular, the design and synthesis of receptors capable of binding anionic guests is of critical importance, due to its potential applications in chemical, environmental, and biological processes [4]. Among the various anions, fluoride (F^-) and pyrophosphate ($HP_2O_7^{3-}$) are the most important because of their various roles in medicinal field [5] and cellular processes [6], respectively. For this obvious reason, various research groups are focusing on the detection and discrimination of these biological entities. There have been a variety of reports thus far concerning fluoride [7] and pyrophosphate [8] selective receptors using various spectroscopic detection methods.

The broad application of anions necessitates the development of easily synthesized receptors that can recognize and sense the anions at very low levels. Due to its simplicity and high sensitivity, fluorescence is becoming increasingly important as a method for chemical trace detection [9]. Receptors based on anion-induced changes in fluorescence appear to be particularly attractive because they offer the potential for high sensitivity at low analyte concentration [10]. Fluorosensors for a large number of biotic and abiotic analytes have been designed in the past decade by appending a fluorescent fragment to the envisaged receptor framework: in all cases, an efficient mechanism has to be provided for either quenching or reviving fluorescence, following substrate recognition. Fluorescent anion receptors utilizing photo-induced electron transfer (PET) [11], intramolecular charge transfer (ICT) [12], excited-state proton transfer [13], metal-to-ligand charge transfer [14], excimer/exciplex formation [15], and competitive binding [16] mechanisms have all been recently developed.

The fluorene family compounds are base materials for the production of dyes and optical brightening agents. Fluorenones have been used extensively as catalyst precursors for electro-catalyticoxidation [17], inhibition of DNA tumor viruses [18], light-emitting materials [19], etc., but have rarely been used as chemical sensors. 9-Fluorenone has been investigated as an attractive element in organic solar cells and display devices. This has led us to design and synthesize a fluorescent sensor containing a fluorenone framework that is particularly suitable for environmental and biological applications. In this paper, we describe a novel chromofluorogenic receptor, 1-(naphthalen-1-yl)-3-(9-oxo-9*H*-fluoren-1-yl)urea (**1**), which contains fluorenone as a chromophore/fluorophore and urea moiety as a

* Corresponding author. Tel.: +82 55 213 3436; fax: +82 55 213 3439.
E-mail address: yilee@changwon.ac.kr (Y-I. Lee).

0026-265X/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
doi:10.1016/j.microc.2012.04.012

binding unit for developing a colorimetric sensor for F^- and $HP_2O_7^{3-}$. Sensor **1** selectively detects anions such as F^- and $HP_2O_7^{3-}$ in CH_3CN: DMSO (9:1, v/v) solution via both colorimetric and fluorometric methods. The anion binding property of sensor **1** was assessed by monitoring the change of fluorescence emission intensity and using ^1H NMR titration techniques after the addition of anions in the form of tetrabutylammonium salts.

2. Experimental section

2.1. General

All of the anions were purchased in the form of tetrabutylammonium salts from Sigma–Aldrich. Solvents such as dimethylsulphoxide (DMSO) and acetonitrile (CH_3CN) were purified prior to use. Spectroscopic-grade solvents were used for the titration studies. Commercial-grade chemicals and solvents were used as such unless otherwise specified. Silica Gel G was used for thin-layer chromatography (TLC). 1-Amino-9*H*-fluorenone, 1-naphthyl isocyanate, and other reagents and solvents were purchased from Sigma–Aldrich and used as such.

All the experiments were conducted at room temperature. All new compounds were fully characterized via standard spectroscopic techniques. Microanalyses were conducted on a Perkin Elmer 2400 Series II CHNS/O Analyzer. Infrared spectra were recorded on a JASCO FTIR-6300 spectrometer. Electronic absorption spectra were recorded with an Agilent 8453 spectrophotometer. ^1H NMR and ^{13}C NMR spectra were generated on a Bruker 400 FTNMR spectrometer at 298 K. High-resolution mass spectra were obtained on a Micromass Platform II mass spectrometer. Fluorescent studies were conducted on a JASCO FP-6500 spectrofluorophotometer.

2.2. Synthesis of 1-(naphthalen-1-yl)-3-(9-oxo-9H-fluoren-1-yl)urea (**1**)

1-Napthylisocyanate (0.23 mL; 1.625 mmol) was added to a dry 10 mL acetonitrile solution of 1-amino-9*H*-fluorenone (0.050 g; 1.30 mmol) at room temperature. The reaction mixture was stirred overnight (~12 h). The completion of the reaction was monitored by TLC. Column chromatography was used to purify the product (90:10; Hex:EA). Recrystallization was done in a hexane/ethyl acetate mixture.

Yield: 80%; M. p.: 215 °C Anal. Cald. for $C_{24}H_{16}N_2O_2$; C, 79.11; H, 4.43; N, 7.69%; Found: C, 78.99; H, 4.34; N, 7.73%; IR (ν_{max}, 25 °C, KBr, cm^{-1}): 3281 3145, 1627, 1555, 1251, 1213, 1130, 780, 690; UV-visible (λ_{max}, 25 °C, CH_3CN:DMSO (9:1, v/v), nm): 255 (naphthalene), 315 (fluorenone); ESI-MS (*m/z*): 364.21; ^1H NMR (DMSO-d_6) 400 MHz, 25 °C, δ (ppm): 9.86 (s, 1H), 9.71 (s, 1H), 8.26–8.21 (q, 2 H, $J_1 = 8.56$ H_z, $J_2 = 5.00$ H_z, $J_3 = 8.16$ H_z), 7.99–7.97 (t, 1H, $J_1 = 6.76$ H_z, $J_2 = 1.60$ H_z), 7.81–7.78 (m, 3 H), 7.65–7.52 (m, 6 H), 7.43–7.38 (m, 2 H); ^{13}C NMR (DMSO-d_6) 100 MHz, 25 °C, δ (ppm): 194.3, 153.5, 137.1, 135.4, 129.8, 128.7, 126.5, 126.3, 126.2, 125.2, 123.9, 122.9, 121.6, 120.6, 114.7.

2.3. Colorimetric and absorption experiments

The colorimetric studies of receptor **1** towards various anions can be easily observed by the naked eye in the CH_3CN:DMSO (9:1, v/v) mixture at a concentration of 0.125 mM. The anions are in the form of tetrabutylammonium salts at a 12.5 mM concentration (host: guest = 1:100). The solutions for the colorimetric test were used to record UV-visible absorption spectra. In the absence of anions, the spectrum of host **1** was characterized by two peaks (315 and a shoulder at 395 nm) (Supplementary material, Fig. A1).

2.4. Fluorescence studies

To detect the anions, we conducted a fluorescence experiment for receptor **1** upon the addition of anions (1:100 equivalent ratios). Stock solutions of receptor **1** (15 μM) and the tetrabutylammonium salts of anions (1500 μM) were prepared in a CH_3CN: DMSO (9:1, v/v) mixture and used for the detection experiments. Titration experiments were conducted by measuring the changes in fluorescence emission that occurred upon the addition of anions to the degassed CH_3CN: DMSO (9:1, v/v) solution of **1** (5 μM). For all receptor **1** measurements, excitation was at 315 nm; both excitation and emission slit widths were 3 nm for detection and 5 nm for titration experiments. The initial volume of receptor **1** was 2 mL. Every titration was repeated at least twice until consistent values were obtained.

2.5. ^1H NMR studies

The solution of receptor **1** (0.001 M in DMSO-d_6) was titrated by adding known quantities of concentrated solution of anions (0.004 M) in the form of tetrabutylammonium salts. The chemical shift changes of the -NH protons of the urea moiety in the receptor were monitored. All titrations were repeated at least twice to obtain consistent values. DMSO-d_6 was purchased from Sigma–Aldrich and dried over molecular sieves (4 Å) prior to use. The anion salts were dried for at least a day in dynamic vacuum, prior to the experiments.

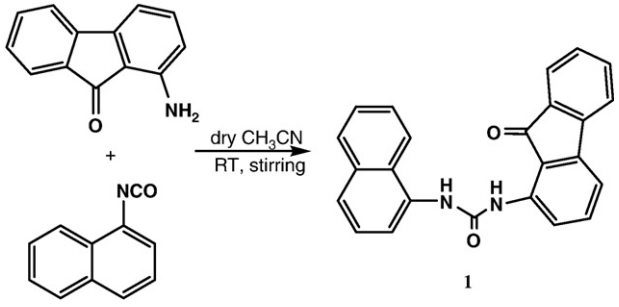
3. Results and discussion

3.1. Design and synthesis of probe **1**

In molecular recognition chemistry, the cavity size and binding sites play a critical role in determining the sensitivity and specificity of the sensor. The conscious modification in these parameters can result in interesting supra-systems. In general, most positively charged anion receptors have amide, pyrrole, urea, and ammonium or guanidinium groups as binding sites, which form N-H--A^- hydrogen bonds [20]. As they offer appropriate binding sites for the guests and stabilize the complexes via non-covalent interactions such as hydrogen and ionic bonding, urea-based anion sensors are generally considered to be of utmost importance [21]. Reaction of 1:1 molar ratio of 1-amino-9*H*-fluoren-9-one and 1-naphthyl isocyanate in dry CH_3CN yielded receptor **1** as pale yellow microcrystals (Scheme 1). The structure of receptor **1** was elucidated by elemental analyses, NMR (^1H and ^{13}C), and mass analysis (Supplementary material, Figs. A2 and A3).

3.2. Colorimetric naked-eye detection

The colorimetric anion sensing ability of receptor **1** was studied by some visual color changes observed upon mixing with various anionic guests (1:100 equiv. ratio) in the form of tetrabutylammonium salts in CH_3CN:DMSO (9:1, v/v) mixture. It was observed that the host solution undergoes significant color changes from pale yellow (395 nm) to wine red (~500 nm) upon addition of F^- and $HP_2O_7^{3-}$ (Fig. 1A). This is associated with deprotonation of more acidic urea proton and the subsequent charge transfer phenomenon [22,23]. As the receptor bound to



Scheme 1. Synthetic route for preparing the receptor (**1**), 1-(naphthalen-1-yl)-3-(9-oxo-9*H*-fluoren-1-yl)urea.