Journal of Luminescence 164 (2015) 138–145

Contents lists available at ScienceDirect

Journal of Luminescence

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

Synthesis and evaluation of a new colorimetric and ratiometric fluorescence probe for copper ions

Har Mohindra Chawla*, Priyanka Munjal, Preeti Goel

Department of Chemistry, Indian Institute of Technology Delhi, Hauz Khas, New Delhi 110016, India

ARTICLE INFO

Article history:
Received 18 October 2014
Received in revised form
16 March 2015
Accepted 24 March 2015
Available online 1 April 2015

Keywords:
Calix[4]arenes
Recognition
Ratiometric
Fluorescence
Copper

ABSTRACT

Synthesis and spectroscopic evaluation of compounds **3a, 3b** and **4** reveal that cone conformer of 25,27-bis(o-aminothiophenyl propyloxy) -tetra-*p*-*tert*-butylcalix[4]arene **3a** can function as a highly selective ratiometric and colorimetric fluorescence probe for copper ions.

1. Introduction

Copper ions play a significant role in both chemical and biological reactions especially in controlling the kinetics and transport of ions within the cell [1]. Major alteration in cellular level concentration of copper ions ushers serious neurodegenerative diseases which may include Alzheimer's and Parkinson diseases [2]. Copper is the third most abundant transition metal ion in the human body and is known to act as a co-factor for metalloenzymes as redox centre [3]. It is extensively used in chemical, electrical, textile and organic chemical industries and can be a cause for serious environmental concern warranting new approaches for its analysis and mitigation [4].

Calixarenes are macrocyclic metacyclophanes which have been extensively examined as molecular receptors. This is primarily due to their easy functionalization, hydrophobic cavity and manipulable conformational characteristics to confer selectivity for metal ion binding [5]. Calixarene scaffolds can be linked to fluorescence functions to offer changes in color and fluorescence characteristics on interaction with low concentrations of metal ions. Research on calixarene based systems has provided significant knowledge on their use, sensitivity and capacity to provide visual detection of analyte with fast response times [6]. General mechanisms involved in calixarene based systems for metal ion detection are mainly photo-induced electron transfer (PET) [7], fluorescence resonance energy transfer (FRET) [8], photon induced charge transfer (PCT) [9] or intramolecular charge transfer (ICT) protocols [10] for which considerable information is available for distinguishing different mechanisms involved in the recognition process.

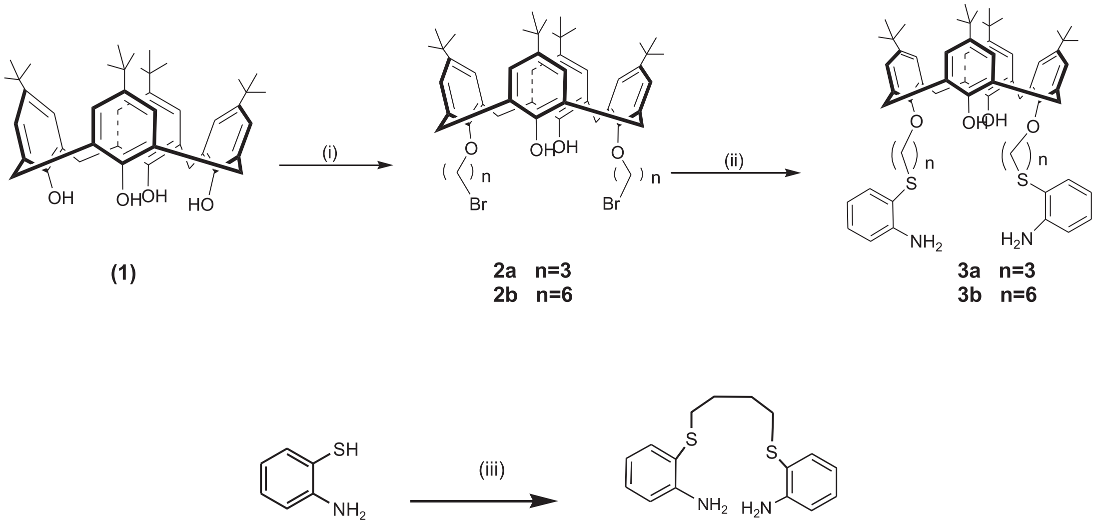
Though a number of fluorescence based methods have been reported for copper ion recognition, most of them are based on fluorescence quenching to reveal so called ‘on–off’ methodology [11,12]. A relatively less number of methods have been investigated which depend upon enhancement of fluorescence intensity on interaction with metal ions [13]. Sadly most of these methods are markedly affected by system perturbations and intermolecular interactions. Recently our group has also reported selective interaction of Schiff base appended calixarenes with copper ions [14]. All these advances in the literature suggest the need for continuation of research on the development of copper selective molecular probes that can function through a ratiometric change (which can show fluorescence changes at two different wavelengths) for better scrutiny and reliability as well as a colorimetric response with anticipated rapidity [15].

In this communication, we report the synthesis of o-aminothiophenyl appended calix[4]arene derivative which selectively and sensitively interacts with copper ions to direct more research into the approach suggested. The selection of calixarene scaffold with useful functionalities based upon a study of active sites of copper containing proteins (e.g., plastocyanin) has been envisaged for ensuring measurement of analyte concentration with built in corrections for environmental and equipment flaws [16]. The designed molecular probe is different from recently investigated fluorophores

---

* Corresponding author. Tel.: +91 11 265 91517; fax: +91 11 265 81102.
E-mail addresses: hmchawla@chemistry.iitd.ernet.in, hmchawla@chemistry.iitd.ac.in (H.M. Chawla).

http://dx.doi.org/10.1016/j.jlumin.2015.03.027
0022-2313/



(1)

2a n=3
2b n=6

3a n=3
3b n=6

(4)

Scheme 1. Synthesis of novel receptor **3a**. Reagents and conditions: (i) 1,3-dibromopropane, CH_3CN, K_2CO_3, reflux; (ii) NaH, DMF, $TBAHSO_4^-$, o-aminothiophenol, stirring and (iii) 1,4-dibromobutane, NaH, DMF, $TBAHSO_4^-$, o-aminothiophenol, stirring.

like coumarin, 1,8-naphthalimide, quinoline and anthraquinone derivatives [17] and takes into cognizance of active site of plastocyanin which is known to play a prominent function in the initiation of biological processes. Since many copper containing proteins and enzymes deploy sulfur and nitrogen containing amino acids separated by a significant hydrophobic pocket in their constitution [18], we envisaged that if we could have nitrogen and sulfur binding sites appended to a hydrophobic cone or partial cone conformer of calix[4]arene (to take care of stereochemical and hydrophobic factors and a propyloxy chain to ensure adequate rigidity/flexibility ratio), the resulting molecular receptor would be able to interact with copper ions in a specific manner. This hypothesis was contemplated since calixarenes have been known to provide useful molecular receptors and sensor materials for transition metal ions.

The synthesis of the designed molecular receptor **3a** has been accomplished by minor modification of chemical reactions (Scheme 1) while its evaluation has been done by spectroscopic investigations for ionic recognition [19]. A reference compound **4** with similar binding sites and signaling units as those present in **3a** was also synthesized for determining the role of hydrophobic calixarene cavity. The design of **4** was adjusted in such a way that it contained the functionalites available in **3a** for interaction with copper ions. The size of calix[4]arene cavity and spacer length was also varied by synthesizing another calixarene derivative (**3b**) with appended hexyl chain in place of propyl chain. The synthetic compounds were investigated by UV–vis, NMR and fluorescence spectroscopy to determine the role of calixarene, spacer unit and conformation. The results obtained on evaluation of these molecular receptors indicate that 25,27-bis(o-aminothiophenyl propyloxy) tetra *p-tert*-butylcalix[4]arene (**3a**) can act as an efficient molecular probe for copper ions and cavity dimensions are important for copper interactions, thereby paving the way to design more efficient plastocyanin based copper ion receptors.

2. Results and discussion

2.1. Synthesis of the molecular receptors *3a* and *3b* and their characterization

The synthesis of the calixarene based molecular receptor **3a** was achieved through selective alkylation of *p-tert*-butylcalix[4]arene with 1,3-dibromopropane in the presence of potassium carbonate followed by a nucleophilic substitution reaction with o-aminothiophenol (Scheme 1). The reaction mixture on work up and column chromatography using ethyl acetate/hexane (2:1) as the eluent, gave creamish white solid (65% yield), with a m.p. 75–78 °C. It exhibited strong IR absorptions at υ_{max} 3343 (NH_2, str.), 1607 (NH, bend), 1361 (C–N, str.). In its ^1H NMR spectrum in DMSO-d_6, compound **3a** revealed the appearance of protons at δ 3.27 (–SCH_2; 4H) and D_2O exchangeable singlets at δ 5.30 and δ 8.57 for NH_2 (aminothiophenol) and hydroxyl protons of calix[4]arene respectively. The disappearance of a triplet at δ 3.97 ppm for –$BrCH_2$ present in the precursor **2a** confirmed the introduction of di-substituted aminothiophenol at the lower rim of calix[4]arene (Fig. 1) which was further confirmed by the appearance of a molecular ion peak at m/z 979.548 $[M+H]^+$ in its high resolution mass spectrum.

Compound **3b** was synthesized by using a similar method as adopted for **3a** except that tetrakis-(p-tert-butyl) 26,28-bis(hexyloxybromo)calix[4]arene (**2b**) was taken instead of tetrakis-(p-tert-butyl) 26,28-bis(propyloxybromo)calix[4]arene (**2a**). In its ^1H NMR spectrum in $CDCl_3$, compound **3b** revealed the appearance of protons at δ 3.01 (–SCH_2; 4H) and disappearance of a triplet at δ 3.53 for –$BrCH_2$ present in the precursor **2b** which confirmed the formation of the compound **3b**. The conservation of the original fixed cone conformation of both the derivatized calix[4]arenes was confirmed by the presence of two sharp doublets of methylene bridges at δ 4.4 and δ 3.3 in the ^1H NMR spectrum and a signal at δ 33 in the ^{13}C NMR spectrum.

2.2. Synthesis of the reference compound *4* and its characterization

Compound **4** was synthesized by using the same method as adopted for the synthesis of **3** when 1,4-dibromobutane (0.250 g, 0.25 mmol) was taken instead of tetrakis-(p-tert-butyl) 26,28-bis(propyloxybromo)calix[4]arene **2.** In its ^1H NMR spectrum, compound **4** revealed the appearance of protons at δ 2.62 (–SCH_2, 4H) and D_2O exchangeable singlet at δ 4.0 for NH_2 (aminothiophenol). The introduction of di-substituted aminothiophenol at 1,4-dibromobutane was confirmed by the appearance of a molecular ion peak at 305.1179 in its high resolution mass spectrum.