2,4-dinitrophenylhydrazine and 1,10-phenanthroline Conjugated Colorometric Receptor for Selective Sensing of F^-

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Abstract: A new chromogenic sensor, 1,10-phenanthroline affixed to the 2,4-dinitrophenylhydrazine moiety, was synthesized and characterized. Among anions added into it, F^-, AcO^- and $H_2PO_4^-$ ions resulted in the color changes from light brown to dark brown, while other tested anions did not induce color changes. Meanwhile, the anion-binding ability has been investigated by UV-vis and ^1H NMR titrations in DMSO-d_6. The sensor displays an even stronger binding to F^- over AcO^- and $H_2PO_4^-$ ions with 1:1 stoichiometry through hydrogen bonding.

Keywords: Colorimetric, anion sensor, supramolecular chemistry, 1,10-phenanthroline.

1. INTRODUCTION

With the development of the supramolecuar chemistry, anion recognition represents an interesting area of research because it plays a critical role in the biological, chemical and environmental fields [1]. In this regard, much effort has been devoted to design and synthesis of the artificial anion receptors [2,3], especially for realizing the visible anion recognition [4-6]. Generally, anion acceptors should contain the appropriate scaffold to accommodate the anionic guest through hydrogen bonding or electrostatic interactions. Notable examples of neutral anion receptors consist of amide [7,8], sulfonamide [9], pyrrole [10,11], urea [12,13], thiourea [14,15], and guanidinium [16] groups as binding sites that are able to establish $N\text{-}H\cdots A^-$ hydrogen bonds with anions. Meanwhile, the hosts of the compounds particularly comprising the chromophoric/fluorescent sites have attracted growing attention in recent years because this mechanism of anion sensing will help to achieve the so-called 'naked-eyes' detection without using any spectroscopic instrument. Thus, the functional groups attached with an appropriate chromophoric part either covalently or intermolecularly as a sensor can lead to either color changes, fluorescing or both. Actually, nitro-substitute phenylhydrazine as a hydrogen donor and chromophoric group has been reported lately. For instance, our group has investigated several anion receptors through coupling *2,4*-dinitrophenylhydrazine with different fragments, which showed significant color changes upon the presence of anions of interest [17].

On the other hand, anions also play a major role in physiology and in healthcare, which has been well documented in farming and in heavy industry. Some of them are harmful pollutants, for example, phosphates from fertilizers, etc. As a result, detection of anions (fluoride, carboxylate, phosphate, and so on) has become extremely significant. The carboxylate anions hold specific biochemical behaviors in the enzymes and antibodies and are critical components of numerous metabolic processes [18]. Phosphate is deemed to be a critical structural element in teeth and bones and it is an essential pollutant whose role in the eutrophication of waterways is well recognized [19,20]. In particular, the fluoride anion, an important anion, is often added to drinking water and toothpaste because of its beneficial effect in dental health. It is also used in the treatment of osteoporosis [21]. However, excessive fluoride ingestion can be problematic and can result in the development of dental or skeletal fluorosis [22]. Recently, Raposo *et al.* [23] group reported a serial of probes using phenylthioureas functionalized with heterocyclic moieties, in most of the cases, which showed highly selective with fluoride by UV-vis titrations and fluorescence study. Meanwhile, X. P. Bao [24] group also reported two simple colorimetric anion chemosensors containing OH and CONH groups. But only one receptor displayed only selectivity with fluoride anion which induced a noticeable color change and UV-vis spectrum changes.

Thus, the synthesis of efficient anion receptors not only frequently requiring elaborate design but also having highly selective with anion has received considerable attention. In this paper, novel anion receptors were designed and synthesized through reacting *2,4*-dinitrophenylhydrazine (binding sites) with *1,10*-phenanthroline group as a yellow solid in 55% yield (Scheme **1**). Consequently, the sensor 1 was sensitive to anions and presented special color deepened with the addition of F^-, $H_2PO_4^-$ and AcO^- anions in DMSO. In addition, the anion sensing was investigated by the UV–vis titration and ^1H NMR titration methods.

2. EXPERIMENTAL

2.1. Apparatus

^1H NMR spectra were obtained on a Varian UNITY Plus-400 MH_Z Spectrometer. ESI-MS was performed with a MARINER apparatus. C, H, N elemental analysis were made on an Elementar vanio-EL. UV–vis spectra were recorded on a Shimadzu UV-2450 Spectrophotometer with a quartz cuvette (path length = 1 cm) at 298.2±0.1K.

2.2. Chemicals

All reagents for synthesis obtained commercially were used without further purification. In the titration experiments, all the anions were added in the form of tetrabutylammonium (TBA) salts, which were purchased from Sigma-Aldrich Chemical, stored in a vacuum desiccator containing self-indicating silica and dried fully before using. DMSO was dried with CaH_2 and then distilled in reduced pressure.

2.3. General Method

All experiments were carried out at 298.2±0.1K, unless otherwise mentioned. UV–vis spectra were measured using an ultraviolet-visible spectrophotometer, UV-2450 (Shimadzu Corp., Kyoto, Japan). A 5.0×10^{-4} $mol{\bullet}L^{-1}$solution of the compound 1 in DMSO was prepared and stored in the dry atmosphere. This solution was used for all spectroscopic studies after appropriate dilution. Solutions of 1.0×10^{-2} $mol{\bullet}L^{-1}$tetrabutylammonium salts of the respec-

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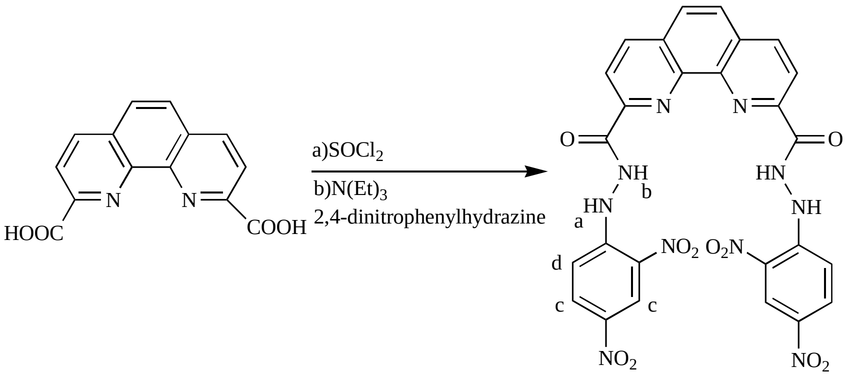


Scheme 1. The synthesis route of receptor 1.

tive anions were prepared in dried and distilled DMSO and were stored under a dry atmosphere.

^{1}H NMR titration experiments were carried out in the DMSO-d_6 solution (TMS as an internal standard). A 1.0 × 10^{-2} mol•L^{-1} solution of the compound 1 in the DMSO-d_6 was prepared. Then, the increased amount of fluoride anion (1.0 mol•L^{-1} in DMSO-d_6) was added to the solution mentioned above.

2.4. Synthesis of 1,10-phenanthroline-2,9-dicarbonyl-2',4'-dinitrophenylhydrazine (1)

The synthesis route of the receptor 1 was demonstrated in Scheme **1**. 0.268 g (1 mmol) *1,10*-phenanthroline-*2,9*-dicarboxylic acid was added into the freshly distilled thionyl chloride (25 mL) and the mixture solution was refluxed for 6h. Then, the solution was concentrated under reduced pressure. The slight yellow residue was dissolved in 25 mL dry CH_2Cl_2 followed by addition of a catalytic amount of triethylamine. *2,4*-dinitrophenylhydrazine (0.396 g, 2 mmol) were added slowly to the above-mentioned cold mixture solution, stirred for 3 days at room temperature, poured into water, filtered and washed to give 0.345 g pure yellow solid after recrystallization from DMF/CH_3CN. Yield=55%. ^{1}H NMR (DMSO-d_6): δ 11.03 (s, 2H,NH), 9.08 (s, 2H, NH), 8.36 (d, 4H, phenyl-H), 8.27 (s, 2H, phenyl-H), 7.72 (s, 2H, phen-H), 7.43(d, 2H, phen-H), 6.87 (d, 2H, phen-H); ^{13}C NMR (DMSO-d_6): 162.9 (C=O), 149.2, 147.4, 138.8, 136.2, 130.3, 128.4, 127.8, 122.9, 122.2, 115.6. ESI-mass: m/z = 1307.9 [M]$^-$, Elemental analysis calcd for $C_{26}H_{16}N_{10}O_{10}\cdot 3H_2O$: C, 45.75; H, 3.25; N, 20.52 Found C, 45.42; H, 3.42; N, 19.95.

3. RESULTS AND DISCUSSION

3.1. UV-*vis* Spectral Titrations

The anion recognition properties of the receptor 1 were evaluated by analyzing the changes in UV-vis spectrum, which are displayed in Fig. (**1a**). The free Receptor 1 (c = 2.0 × 10^{-5} mol•L^{-1}) in

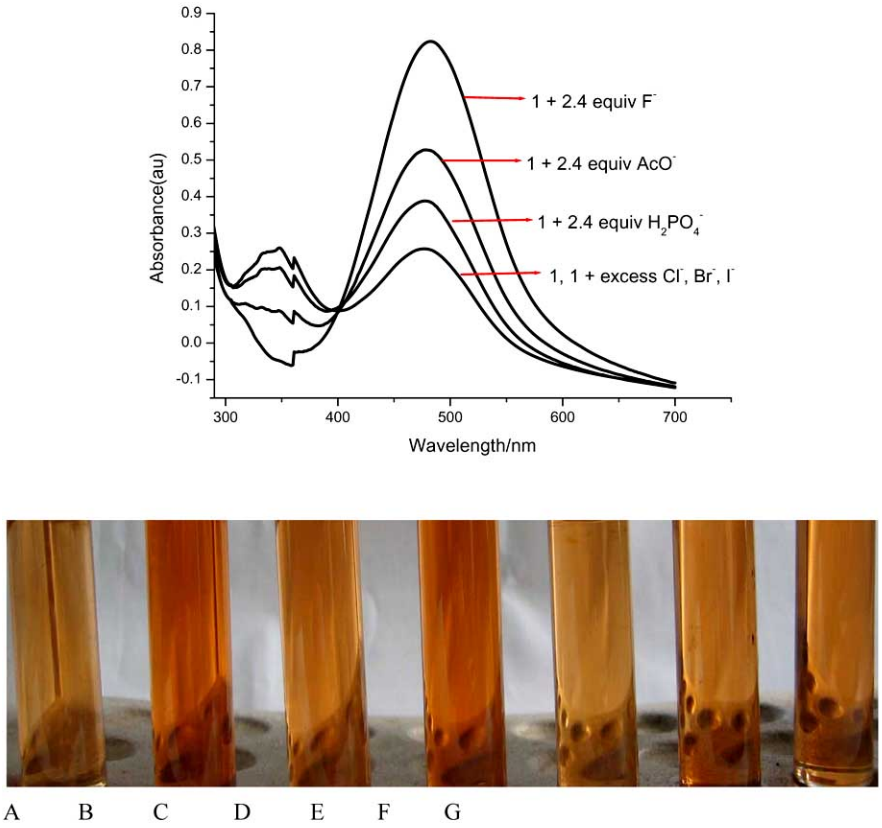


Fig. (1a). UV-vis spectral changes induced by different anions tested. (**1b**) Color changes of sensor 1 in DMSO (2×10^{-5} mol•L^{-1}) after addition of 10 eq. anions. A: 1 only; B: 1 + F$^-$; C:1 + AcO$^-$; D: 1 + $H_2PO_4^-$; E: 1 + Cl$^-$; F: 1 +Br$^-$; G: 1 + I$^-$.

DMSO shows an intense absorption band centered at 475 nm and a mild peak about at 348nm. As shown in spectra 1a, 1a exhibits remarkable changes upon addition of 2.4 equiv of F^-, $H_2PO_4^-$ and AcO^- (all as their tetrabutylammonium salts), respectively, whereas negligible responses are observed in the presence of other anions (Cl^-, Br^- and I^-). Meanwhile, on the addition of the F^-, $H_2PO_4^-$ and AcO^-, the solution color changes from light brown to dark brown while the solutions display little color response by adding other anions in DMSO (See Fig. **1b**). It is apparent that the compound 1 has higher selectivity for F^- anion than other anions because the absorption spectrum more obviously changed upon addition of fluoride. To investigate the anion binding properties of sensor 1 with the ions of the F^-, $H_2PO_4^-$, AcO^-, Br^-, Cl^- and I^-, UV-vis spectral titrations were further carried out in DMSO. During the titration the sensor 1 with F^-, the intensity of the absorption band centered at 480 nm obviously enhanced while the intensity of the absorption at 348 nm substantially decreased (Fig. **2**). Obviously, as shown from the Fig. (**2**), there was a clear isobestic point at 389nm, which indicated the host and the fluoride anion formed a type of complex by hydrogen bond. Meanwhile, The electronic properties of the chromophore were impacted by the formation of the hydrogen bonds leading to a color change, which was implemented through the charge transfer between the electron abundant nitrogen atom of NH unit and the electron deficient *2,4*-nitrophenyl moiety.

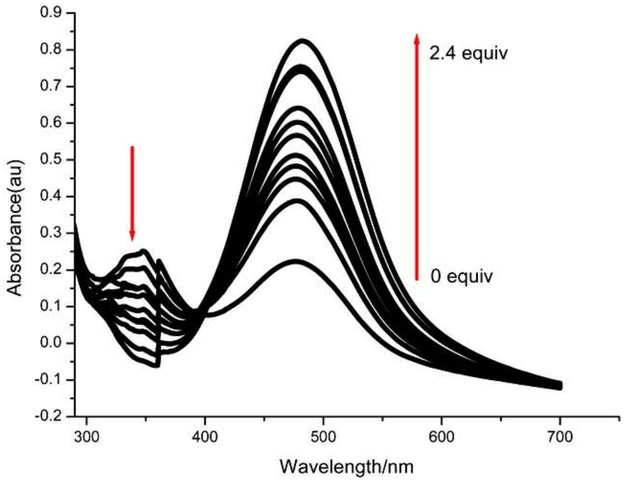


Fig. (2). Spectra taken over the course of the titration of DMSO solution of 1 (2×10^{-5} mol•L^{-1}) with a standard solution of $[Bu_4N]F$.

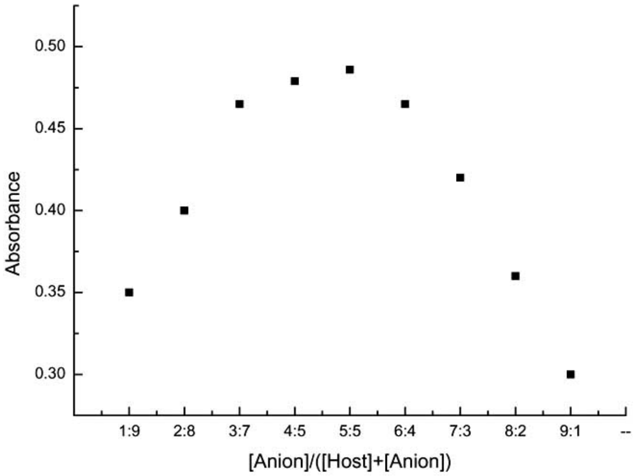


Fig. (3). The stoichiometry analysis of complex 1•F^- by Job plot analysis.

3.2. Determination of Affinity Constants

To determine the stoichiometry of the host-guest complex, job plots were obtained according to the method reported by K.A. Connors [25]. As an example (F^-), Fig. (**3**) demonstrated the formation of 1:1 stoichiometry host-fluoride complex. The association constants of the host 1 for anionic species, which were shown in Table **1**, were determined by non-linear fitting analyses of the titration curves according to the equation (1), for 1:1 host-guest complexation reported by B. Valeur [26].

$$A=A_0 + (A_{lim}-A_0)\{c_H+c_G+1/K-[(c_H+c_G+1/K)^2-4c_Hc_G]^{1/2}\}/2c_H \qquad (1)$$

Where, c_G and c_H are the concentration of guest and host, respectively and A is the intensity of absorbance at certain concentrations of the host and the guest. A_0 is the intensity of absorbance of host alone and A_{lim} is the maximum intensity of absorbance of host when guest is added. K is the affinity constant.

Table **1** demonstrated the apparent association constants of the sensor 1 with different anions. From the Table **1**, it is shown that the association constant of the receptor 1 for fluoride anion is greater than other anions. Actually, the trend of selectivity of binding affinities of 1 for anions is $F^- > AcO^- > H_2PO_4^- >> Cl^- \sim Br^- \sim I^-$. At present, two principles could illuminate the differences among the all kinds of the anions for anion recognition. Firstly, it is necessary to hold the multiple hydrogen-bonding interactions between the hosts containing high affinity binding parts and the anionic guests. Secondly, another important factor is the charge and shape complementarity between the sensor and the anions. Obviously, the hosts possess of four NH groups, which can donor the H atom to the anionic guests like F^-, $H_2PO_4^-$ and AcO^-, however, these different anions exist obvious differences in shape complementarity that results in the unlike association affinities. As expected from the basicity of anions, F^-, AcO^- and $H_2PO_4^-$ gave stronger complexation than other anions, and could well be taken as model anions to demonstrate the signal mechanism. Furthermore, the fluoride anions was spherical and the smallest guest among them and might be the best one to fit for the sensor 1 with four NH binding sites, to form multiple hydrogen-bonding interaction (Scheme **2**). The $H_2PO_4^-$ was tetrahedral configuration and the acetate anion was a plane and triangular, respectively, which could not well match with four hydrogen atoms in the receptor. That is why the association constant K_{ass} for fluoride anions is the highest among the anions tested.

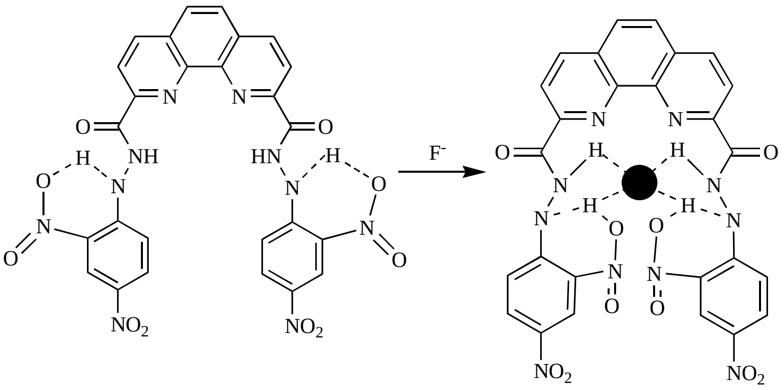
3.3. ^{1}H NMR Titrations

To further understand the character of the receptor–anion interactions, the ^{1}H NMR titrations were performed to monitor the changes of the receptor 1 (1×10^{-2} mol L^{-1}) upon addition of the fluoride anion in DMSO-d_6. Obviously observed from the Fig. (**4**), addition of 2 equiv of the tetrabutylammonium salts of fluoride anion to sensor 1 resulted in negligible shifts for the signals of two NH groups at 11.03 ppm (Ha) and 9.08 ppm (Hb), respectively, indicating one of the NH groups (Ha) of hydrazine moiety estab-

Table 1. Association Constants (logK_{ass}, M^{-1}) of the Sensor 1 with Anions in DMSO at 298.2±0.1K

Anions[a]	**F^-**	**AcO^-**	**$H_2PO_4^-$**	**Cl^-**	**Br^-**	**I^-**
logK_{ass}[b]	$5.12\pm0.25\times10^5$	$4.01\pm0.42\times10^4$	$3.83\pm0.33\times10^4$	ND[b]	ND	ND

[a] All the anions were added in the form of tetra-n-butylammonium (TBA) salts.
[b] ND: The association constant could not be determined due to very weak complexation.



Scheme 2. The proposed host–guest binding mode in solution.

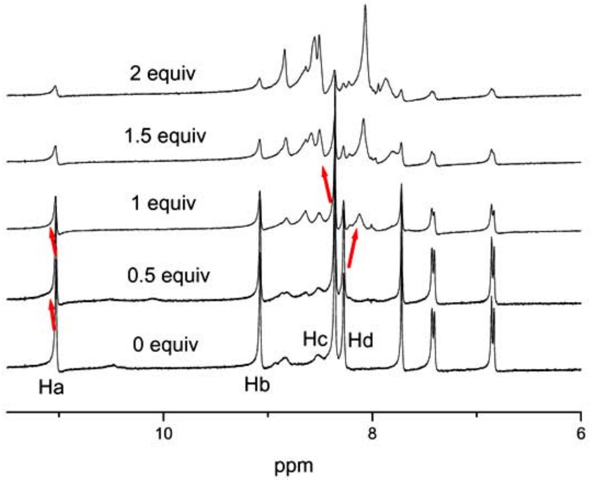


Fig. (4). Changes in ^1H NMR spectrum of the receptor 1 in the absence and presence of F^- ions.

lishes intramolecular hydrogen bonding with the oxygen of electron-deficient NO_2 moiety by forming a six-member ring structure, and the other NH (Hb) part undergoes intramolecular hydrogen bonds with the fluoride anion as well, because the ^1H NMR spectra of Ha and Hb almost appeared to shift and broaden. Meanwhile, the aromatic signals of phenyl at 8.36ppm (Hc) and at 8.24ppm (Hd) were changed slightly due to the electron donating resonance affecting the charge of the phenyl group. From now on, two effects would be considered to operate the formation of hydrogen bonding between the hosts and anionic guests: (1) A shielding effect is induced by the increase of electron density in the phenyl rings through bond transmitting that should promote an upfield shift of ^1H NMR peak. (2) A deshielding effect caused by the polarization of the C-H bonds through a space effect that creates the partial positive charge on the proton and leads to a downfield shift [27]. Thus, it appeared that the ^1H NMR spectra reasonably implicated the anion recognition process *via* hydrogen-bonding interaction between NH groups of hydrazine and fluoride.

CONCLUSIONS

In conclusion, the new colorimetric anion receptor containing NH parts of hydrazine units was synthesized in a simply way. The sensor displays a strong binding to fluoride ions in DMSO through hydrogen bonds over other anions tested and can act an anionic recognition event by UV-vis spectrum and ^1H NMR titration. The receptor has better selectivity to F^- and the order is $F^- > AcO^- > H_2PO_4^- >> Cl^- \sim Br^- \sim I^-$.

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REFERENCES

[1] Hirsch, K. H.; Fischer, F. R.; Diederich, F. Phosphate recognition in structural biology. *Angew. Chem. Int. Ed*., **2007**, *46*, 338-352.

[2] Sessler, J. L.; Gale, P. A.; Cho, W.S. *Anion Receptor Chemistry*, Royal Society of Chemistry: Cambridge, **2006**.

[3] Nabeshima, T.; Saiki, T.; Iwabuchi, J.; Akine, S. Stepwise and dramatic enhancement of anion recognition with a triple-site receptor based on the Calix[4]arene framework using two different cationic effectors. *J. Am. Chem. Soc.,* **2005**, *127*, 5507-5511.

[4] Lin, C.; Simov, V.; Drueckhammer, D. G. Interaction of halide and carboxylate ions with 4, 5-Diacetamidoacridine-9(10H)-one: Thermodynamics of association and deprotonation events. *J. Org. Chem*., **2007**, *72,* 1742-1746.

[5] Maeda, H.; Kusunose, Y.; Mihashi, Y.; Mizoguchi, T. BF_2 Complexes of β-Tetraethyl-Substituted Dipyrrolyldiketones as anion receptors: potential building subunits for oligomeric systems. *J. Org. Chem*., **2007**, *72*, 2612-2616.

[6] Quinlan, E.; Matthews, S. E.; Gunnlaugsson, T. Colorimetric recognition of anions using preorganized tetra-amidourea derived Calix[4]arene sensors. *J. Org. Chem.,* **2007**, *72*, 7497-7503.

[7] Chmielewski, M. J.; Zieliński, T.; Jurczak. Synthesis, structure, and complexing properties of macrocyclic receptors for anions. *Pure Appl. Chem*., **2007**, *79*, 1087-1096.

[8] Kang, S. O.; Begum, R. A.; Bowman-James, K. Amide-Based ligands for anion coordination. *Angew. Chem., Int. Ed.,* **2006**, *45*, 7882-7894.

[9] Kondo, S.I.; Suzuki, T.; Toyamaand, T.; Yano, Y. Anion recognition by 1,3-benzenedisulfonamide derivatives bearing phenolic hydroxy groups in MeCN-d_3. *Bull. Chem. Soc. Jpn.,* **2005**, *78*, 1348-1350

[10] Sessler, J. L.; Camiolo, S.; Gale, P. A. Anion and ion-pair receptor chemistry: highlights from 2000 and 2001. *Coord. Chem. Rev*., **2003**, *240*, 17-55.

[11] Gale, P. A. Amidopyrroles: from anion receptors to membrane transport agents. *Chem. Commun.,* **2005**, 3761-3772.

[12] Boiocchi, M.; Del Boca, L.; Gomez, D. E.; Fabbrizzi, L.; Licchelli, M.; Monzani, E. Nature of urea−fluoride interaction: incipient and definitive proton transfer. *J. Am. Chem. Soc*., **2004**, *126*, 16507-16514.

[13] Kwon, J. Y.; Jang, Y. J.; Kim, S. K.; Lee, K. H.; Kim, J. S.; Yoon, J. Y. Unique hydrogen bonds between 9-Anthracenyl hydrogen and anions. *J. Org. Chem.,* **2004**, *69*, 5155-5157.

[14] Gunnlaugsson, T.; Davis, A. P.; Hussey, G. M.; Tierney, J.; Glynn, M. Design, synthesis and photophysical studies of simple fluorescent anion PET sensors using charge neutral thiourea receptors. *Org. Biomol. Chem*., **2004**, *2*, 1856-1863.

[15] Gunnlaugsson, T.; Davis, A. P.; O'Brien, J. E.; Glynn, M. Synthesis and photophysical evaluations of charge neutral thiourea or urea based fluorescent PET sensors for bis–carboxylates and pyrophosphate. *Org. Biomol. Chem.,* **2005**, *3*, 48-56.

[16] Schug, K. A.; Lindner, W. Noncovalent binding between guanidinium and anionic groups: focus on biological- and synthetic-based arginine/guanidinium interactions with Phosph[on]ate and Sulf[on]ate residues. *Chem. Rev.*, **2005**, *105*, 67-113.

[17] Shao, J.; Lin, H.; Lin, H. K. A simple and efficient colorimetric anion receptor for $H_2PO_4^-$. *Spectrochim. Acta: A*, **2008**, *70*, 682-685.

[18] Gunnlaugsson, T.; Davis, A. P.; O'Brien, J. E.; Glynn, M. Fluorescent sensing of pyrophosphate and Bis-carboxylates with charge neutral PET chemosensors. *Org. Lett.*, **2002**, *4*, 2449-2452.

[19] Kuo, L.J.; Liao, J. H.; Chen, C. T.; Huang, C. H.; Chen, C. S.; Fang, J. M. Two-arm ferrocene amide compounds: synclinal conformations for selective sensing of dihydrogen phosphate ion. *Org. Lett.*, **2003**, *5*, 1821-1824.

[20] Yoon, J.; Kim, S. K.; Singh, N. J.; Lee, J. W.; Yang, Y. J.; Chellappan, K.; Kim, K. S. Highly effective fluorescent sensor for $H_2PO_4^-$. *J. Org. Chem.*, **2004**, *69*, 581-583.

[21] Riggs, B. L. *Bone and Mineal Research*; Annual 2; Elsevier: Amsterdam, **1984**; pp. 366-393.

[22] Aaseth, J.; Shimshi, M.; Gabrilove, J. L.; Birketvedt, G. S. Fluoride: A toxic or therapeutic agent in the treatment of osteoporosis? *J. Trace Elem. Exp. Med.*, **2004**, *17*, 83-92.

[23] Raposo, M.; Calero, P. Synthesis and Study of the use of heterocyclic Thiosemicarbazones as signaling scaffolding for the recognition of anions. *J. Org. Chem.*, **2010**, *75*, 2922-2933.

[24] Bao, X. P.; Zhou, Y. H. Synthesis and recognition properties of a class of simple colorimetric anion chemosensors containing OH and CONH groups. *Sens. Actuators B.*, **2010**, *147*, 434-441.

[25] Connors, K. A. *Binding constants: The measurement of molecular complex stability*, USA Wiley, **1987**, pp. 25-28.

[26] Bourson, J.; Pouget, B.; Valeur, Ion-responsive fluorescent compounds. Effect of cation binding on the photophysical properties of a coumarin linked to monoaza- and diaza-crown ethers. *J. Phys. Chem.*, **1993**, *97*, 4552-4556.

[27] Boiocchi, M.; Del Boca, L.; Gomez, D. E.; Fabbrizzi, L.; Licchelli, M.; Monzani, E. Nature of urea−fluoride interaction: incipient and definitive proton transfer. *J. Am. Chem. Soc.*, **2004**, *126*, 16507-16514.

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