CHAPTER 2

Tripodal chemosensors in cation recognition
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Outline

Tripodal receptor contains multi arms with different functionalities which are reserved for the binding of analytes. It is considered that tripodal shaped receptors are between cyclic and acyclic ligands that can bind guest molecules more strongly in comparison to acyclic ones. The binding ability and the selectivity towards guest molecules largely depend on the cavity of the tripodal receptor and the flexibility of the tripodal arms. Selective binding of analytes by the tripodal receptors has a number of advantages in comparison to monopdal and bipodal receptors. This chapter deals with the molecules of this family.

Section A of this chapter addresses the status of tripodal shaped chemosensors with different topologies that express their ability in cation binding.

Section B describes the present work on tripodal shaped architectures that show selective sensing of different metal ions. Subtle variation in structure leads to different selectivity in the sensing process.
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Section A

Status on tripodal chemosensors in cation recognition

Outline

This section summarizes the critical survey of various reports and reviews on the recognition of cations by artificial tripodal shaped receptors.
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2A.1. Review on synthetic tripodal shaped chemosensors towards cation recognition.

Tripodal receptor contains multi arms with different functionalities which are tailored or attached for the binding of analytes. It is considered that tripodal shaped receptors are between cyclic and acyclic ligand that can bind guest molecules more strongly in comparison to acyclic ones.\(^1\) The binding ability and the selectivity towards guest molecules largely depend on the cavity of the tripodal receptor and the flexibility of the tripodal arms.\(^2\)\(^-\)\(^5\) Selective binding of analytes by the tripodal receptors has a number of advantages in comparison to monopodal and bipodal receptors. As the tripodal receptors contain flexible chain, consist of several donor atoms, can form stable complexes with many alkaline earth metals, alkali and even with transition metal ions.\(^6\)\(^,\)\(^7\) Tripodal receptors can form octahedral complexes by coordination of single metal centre by facial manner or can form polynuclear complexes by three dimensional binding approaches.\(^8\) Limited number of tripodal shaped receptor for sensing of cations is known in the literature. Some of them are described here. Shanzer and co-workers have shown that tripodal receptor \(2A.1\) is selective for \(\text{Ca}^{2+}\) ions.\(^9\) Trimethylolpropane- based \(2A.1\) also used in commercially available \(\text{Na}^+\) ionophore \(2A.2.\)\(^10\)

\[
\begin{align*}
2A.1 & \text{ R = NHCR'ON} & 2A.2 & \text{ R = N(CH}_3\text{)}_3C\text{H}_3 \\
2A.3 & \text{ R = R}_1 = \text{Et, } R_2 = \text{Et, } R_3 = \text{Et, } R_4 = \text{N(CH}_3\text{)}_3C\text{H}_3 & \\
2A.4 & \text{ R = R}_1 = \text{Et, } R_2 = \text{R}_3 = \text{Et, } R_4 = \text{N(CH}_3\text{)}_3C\text{H}_3 & \\
2A.5 & \text{ R = Et, } R_2 = \text{Et, } R_3 = \text{R}_4 = \text{N(CH}_3\text{)}_3C\text{H}_3 & \\
2A.6 & \text{ R = Et, } R_2 = \text{Et, } R_3 = \text{Et, } R_4 = \text{N(CH}_3\text{)}_3C\text{H}_3 & \\
\end{align*}
\]

Analogous designs such as \(2A.3, 2A.4\) and \(2A.5\) as established by Yin et. al were used for alkali and alkaline earth metals.\(^6\) Among the various metal salts, these three receptors showed better transporting ability towards \(\text{Li}^+, \text{Na}^+\) and \(\text{Ca}^{2+}\) ions.

Fan and co-workers synthesized tripodal shaped receptor \(2A.6\) that contains carboxylmethoxymethyl groups. It was noted to act as a perfect ionofore for the analysis of alkali and alkaline earth metal ions in PVC membrane electrode.\(^11\)
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Teasdale et al. reported the synthesis of tripodal oxa-amides and oxa-esters- based receptor 2A.7 and described a comparative study as ionophore in potentiometric ion-selective electrodes for alkali and alkaline earth cations. Considering the feature of 2A.7 in mind, they further synthesized compounds 2A.8 and 2A.9 for analysis of Li\(^+\), Na\(^+\), K\(^+\), Ca\(^{2+}\), and Mg\(^{2+}\) ions in PVC membrane electrode. Recognition of metal ions by tripodal shaped receptors containing N-acyl (thio)urea and picolin(thio)amide are also known. Garcia and coworkers reported a series of N-acyl (thio) urea based receptors 2A.10-2A.13 which were used for sensing and extraction of Ag\(^+\) ions. In the series, compound 2A.11 exhibited the highest ability to sense Ag\(^+\) ion.

Same group also introduced receptors 2A.14 and 2A.15. Pyridine amide-based structure 2A.14 intimated greater efficiency for Hg\(^{2+}\) ions in the presence of other metal ions, even in presence of soft Ag\(^+\) metal ions. In comparison, structure 2A.15 sensed Cu\(^{2+}\) ions. Tuntulani et, al synthesized aza crown ether calix[4]arenes based tripodal receptors 2A.16a and 2A.16b for selective binding of transition metal ions such as Co\(^{2+}\), Cu\(^{2+}\), Ni\(^{2+}\), Zn\(^{2+}\). They reported that receptor 2A.16 binds Co\(^{2+}\), Cu\(^{2+}\), Ni\(^{2+}\) in 1:1 stoichiometric fashion and Zn\(^{2+}\) binds in both 1:1 and 1:2 fashions.
Tripodal shaped structures 2A.17 and 2A.18 were synthesized by Hiratani et. al. using Claisen rearrangement as a key step. They were explored in metal ion sensing. They introduced three 8-hydroxyquinolyl moieties as binding sites in 2A.17 for selective binding of trivalent metal salts. Among various trivalent metal salts, 2A.17 exhibited selectivity for Ga$^{3+}$ ions. The same group used catechol units in receptor 2A.18 for binding of iron (III) trichloride hexahydrate.

Hexadentate tripodal ligand 2A.19, containing three 8-hydroxy-5-sulfoquinoline moieties was used for fluoregenic sensing of Al$^{3+}$ and Ga$^{3+}$ ions. They proved the "tripod effect" of 2A.19 in the binding of metal salt by synthesizing compound 2A.19a. Tripod 2A.19 gave higher binding with Al$^{3+}$ and Ga$^{3+}$ ions in comparison to the 2A.19a. The reason for performance of 2A.19 was further investigated by the same group. They synthesized 8-hydroxy-5-sulfoquinoline and two 5-sulfocatechol subunits containing receptor structure 2A.20. They reported that the key factor of fluorescence enhancement was the stoichiometry between Al$^{3+}$ and the bound bidentate subunits. The chelating groups and their formal charges turned the charge density of Al$^{3+}$ due to which...
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the photoinduced charge transfer quenched the fluorescence emission of the 8-hydroxyquinoline ligand.

Prodi et al. reported dansyl labeled tripodal receptor 2A.21, for selective sensing of metal ions.\(^\text{17}\) Receptor 2A.21 shows strong binding ability towards Cu\(^{2+}\), Co\(^{2+}\), Zn\(^{2+}\) and Cd\(^{2+}\) metal ions in semi aqueous solvent and the complexation features were documented from the pronounced change in fluorescence intensity. To prove the cooperative binding effect of tripodal arms compound 2A.22 was undertaken in the study. The tripod 2A.21 was selective for Cu\(^{2+}\) ion at neutral pH.

Malval and coworkers prepared a tripodal fluorescent probe 2A.23, by the incorporation of p-(N, N-dimethylamino) benzenesulfonamide and tris (2-aminoethyl) amine (tren) for selective binding of Zn\(^{2+}\) ions.\(^\text{18}\) Calixarene - based tripodal receptor containing diphenylphosphoryl acetamide was observed to function at the upper rim for selective binding of Actinides/ Lanthanides metal ions.\(^\text{19}\) They used carbamoylmethylphosphine oxide (CMPO) for selective binding as well as extraction of lanthanides and actinides metal ions. In their further work they used similar concept i,e modification of calix[4]arenes with carbamoylmethylphosphine oxide functions at the wide rim for recognition of metal salts.\(^\text{19-23}\) Like calixarene platform, Reinhoudt used cavitands -based platform containing CMP(O) as main binding units for recognition of actinides as well as lanthanides metal ions.\(^\text{24,25}\) These types of receptor molecules though contain four ligating arms but three CPMO units play vital roles in the binding process.

Scott et al. devised C\(_3\)-Symmetric Triphenoxymethane Platform in which three carbamoylmethylphosphine oxide (CMPO) moieties are anchored and preorganized for binding of metal ions.\(^\text{26}\) They reported that three CPMO moieties are oriented in such a way that three CPMO cooperatively bind through six donor oxygen atoms and one or two nitrate counter ions with metal ions. They show that between lanthanides and actinides, actinide metal ions bind more strongly and hence they are extracted from aqueous
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Like receptor 2A.27, tripodal shaped receptor 2A.28 showed greater ability to complex lanthanides metal ions. The crystal structure reveals that the lanthanide ion is eight-coordinated by two tetradentate ligands, forming distorted bicapped trigonal antiprism. As an explanation, this type of complex structure has been possible due to the presence of strong n- n interactions between the benzimidazole rings.

Compound 2A.29 having catechol and tris-methylaminocyclohexane units, choose trivalent metal salts at the core. During complexation, tripodal receptor 2A.29 formed rigid tripodal framework by intramolecular H-bonding. Cyclohexane unit in the design acts as a spacer which assists in effective binding of trivalent lanthanides.
Sasaki and co-workers synthesized substituted benzene tripodal derivative 2A.30 for recognition of tetrahedral NH$_4^+$ ions over spherical K$^+$ ions. They revealed that the pyrazole nitrogen is responsible for this recognition ability towards NH$_4^+$ ions over K$^+$ ions. They reported that receptor 2A.30 is comparable towards NH$_4^+$ ion recognition as like as the natural product nonactin, used as representative NH$_4^+$ ionophore. However, alkylammonium ions were found to be complexed at the core of the tris (oxazolines) based tripodal receptors (2A.33-2A.36).

Receptor 2A.34 was characterized to show selectivity towards n.BuNH$_3^+$ and t.BuNH$_3^+$. Compound 2A.36 containing bare oxazoline ring is more effective in the sensing of sterically hindered t.BuNH$_3^+$. Modification of framework by the introduction of 2, 4, 6- triethylbenzene instead of 2, 4, 6- trimethylbenzene led to the new structures 2A.37 and 2A.38 which gave more enhanced selectivity towards n.BuNH$_3^+$.

Kim and co-workers synthesized 1,3,5-trisphenoxy-2,4,6- triethylbenzenes (2A.39-2A.45) for effective cation binding. They show that upon introduction of electron donating units like OCH$_3$ and OCH$_2$Ph groups on the phenoxy units of 2,4,6-triethylbenzenes enhanced the cation binding ability. From experimental findings, they suggested that modification of the phenoxy units by changing adjacent cation-n-
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interactions as well as the direct hydrogen bonding interaction, influences the ammonium ion binding ability.

Abbastabar-Ahangar et al. synthesized 3, 5-dinitro-N-(tri-2-pyridylmethyl) benzamide labeled tripodal receptor 2A.46 for sensing and determination of Cadmium (II).\textsuperscript{35} It showed high sensitivity and stability towards accurate determination of low level of cadmium.

Kikkeri et al. addressed a series of bioinspired tripods based on fluorescent Phenol-oxazoline sites for binding of Fe(III) ions.\textsuperscript{36} They used (4S,5S)-2-(2-hydroxyphenyl)-5-methyl-4,5-dihydro-1,3-oxazole-4-carbonyl group 2A.47,

for its potential dual properties, serving as a selective iron(III) binder and simultaneously as a fluorophore. Cis-2A.48, cis- 2A.49 and trans -2A.49 varied in length of the spacers between the central carbon anchor and the ligating sites. During
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the titration of cis-2A.48 and cis- 2A.49 with Fe(III) ions, fluorescence intensity was completely quenched.

Tripodal receptor 2A.50 based on imine linkage was developed by Bhardwaj et al. for selective sensing of Ag\(^+\) ions.\(^{37}\) The fluorescence intensity was considerably enhanced upon interaction with only Ag\(^+\) ions.

Saluja et al. synthesized benzthiazole-based tripodal shaped chemosensor 2A.51 which sensed Ba\(^{2+}\) ions in semi aqueous solvent.\(^{38}\) Here also the emission intensity was enhanced upon addition of Ba\(^{2+}\) to the receptor solution. Naphthalene motif-based tripodal chemosensor 2A.52 exhibited selectivity towards Al\(^{3+}\)/Pb\(^{2+}\) metal ions over other transitional and alkaline earth metal ions.\(^{39}\) Addition of Al\(^{3+}\) or Pb\(^{2+}\) enhanced fluorescence intensity of 2A.52 more than 10-fold in comparison to its initial intensity.

In addition to the above examples, some other interesting tripodal structures are illuminated below. They are mainly responsible for sensing of Zn\(^{2+}\), Cu\(^{2+}\), Co\(^{2+}\) and Hg\(^{2+}\) ions.

Lee et al. reported tripodal structure 2A.53 for ratiometric fluorescent determination of Zn\(^{2+}\) ions.\(^{40}\) The key factor of this sensor was the sp\(^2\) nitrogen and carbonyl groups from amide linkages which took part in simultaneous binding of a metal ion involving stable five-membered ring.
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From our laboratory anthracene labeled simple tripodal shaped receptor 2A.54 has been reported for selective sensing of Zn$^{2+}$ ions over other metal ions.\textsuperscript{41} Ghosh et al. published quinoline and naphthalene- based tripodal shaped sensors 2A.55 and 2A.56 for recognition of metal ions.\textsuperscript{42} Upon interaction of 2A.55 with Hg$^{2+}$ ions significant change in Uv- vis spectra was recorded.

In addition, interaction caused greenish fluorescence upon UV light irradiation of receptor- Hg$^{2+}$ mixed solution. Receptor 2A.56 which contains naphthalene in lieu of quinoline, did not show any selectivity towards any metal considered in the study.

Thus the survey of the literature demonstrates that the tripodal receptors provide suitable platform for metal ion binding starting from alkali metal ions to transition metal ions. The selectivity can be tuned upon modifying the binding arms. At the same time, the structures can be converted to the good sensors on adding suitable probes in the close vicinity of the binding site. The flexibility of the binding core of the structures also plays key role in giving selectivity in the binding event. All these features impart efficacy to the tripodal shaped molecules to act as good hosts in cation recognition.
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2A.2. References

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CHAPTER 2
Section B

Tripodal chemosensors in cation recognition

Outline

This section deals with the metal ion recognition using anthracene and pyrene fluorophores - based tripodal chemosensors. The tripodal sensors have been synthesized through a common strategy. The strategy is unique as the tripods of variable structures can be obtained easily. The metal ion binding properties of the tripods have been discussed in detail. The subtle variation in structure keeping the binding site unchanged results in the change in selectivity for a particular metal ion.
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2B.1 Objective
The discussion on the tripodal receptors in metal ion recognition in section 2A reveals that the flexible nature and variable binding units of the tripods make them promising candidates for selective recognition of metal ion. Considering the different characteristic features of tripodal receptors we framed our objective as to synthesize new tripodal shaped luminescent receptors through a common strategy for selective sensing of metal ions of biological significance.

2B.2. Present work
In order to fulfill the objective, the tripodal shaped molecules 2B.1 - 2B.6 were designed and synthesized. In all the designs, the alcoholic -OH group being one of the other functional groups, is disposed around the tertiary aliphatic nitrogen. In this context, our strategy was to place fluorophore, alcoholic and amide functional groups around a trivalent nitrogen centre to create functional receptor module for metal ions.

Also we intended to couple two tripodal centers to give another set of receptor module that may bind cationic substrates effectively through cooperative involvement of the two
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tripodal centers. The designed tripodal receptors 2B.1 - 2B.6 contain either anthracene or pyrene as fluorophore to report the recognition event.

2B.2.1. Syntheses of 2B.1 - 2B.4

Anthracene - based molecules 2B.1 - 2B.4 were synthesized according to the Scheme 2B.1. Condensation of 9-anthraldehyde with ethanolamine yielded Schiff base 2B.7a, which was reduced in situ to the amine 2B.7b using NaBH₄ in MeOH.

Scheme 2B.1. (i) Dry MeOH, reflux, 9 h.; (ii) NaBH₄, dry MeOH, reflux, 3 h.; (iii) Ethyl 2-chloroethanoate, K₂CO₃, dry Me₂CO, reflux, 7 h.; (iv) Amine 2B.11, THF, stirring, 4 h; (v) Amine 2B.13, THF, stirring, 4 h; (vi) NaH, imidazole, THF, CS₂, CH₃I; (vii) Amine 2B.15, THF, stirring, 8 h; (b) (i) o-phenylenediamine, DCC, DMAP, stirring in CH₂Cl₂ for 19 h; (ii) AcOH, heat, 2 h; (c) (i) 50% TFA in CH₂Cl₂, stirring, 3 h; (ii) NaH, dry THF, n-butyl bromide, reflux, 8 h; (iii) 50% TFA in CH₂Cl₂, stirred for 3 h; (iv) NaH, dry THF, 1, 3- dibromomethyl benzene, reflux, 8 h; (v) 50% TFA in CH₂Cl₂, stirred for 3 h.
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Scheme 2B.2. (i) Dry Benzene, reflux, 9 h.; (ii) NaBH₄, dry MeOH, reflux, 4 h.; (iii) Ethyl 2-chloroethanoate, K₂CO₃, dry Me₂CO, reflux, 7 h.; (iv) Amine 2B.11, THF, stirring, 4h; (v) Amine 2B.13, THF, stirring, 4h.

Reaction of 2B.7 with ethyl 2-chloroethanoate in the presence of K₂CO₃ in dry acetone under refluxing condition afforded the 4-substituted morpholin-2-one 2B.8 in 77% yield (Scheme 2B.1a). We achieved the synthesis of this 4-substituted morpholin-2-one in a simple way although other methods, in this context, are known in the literature.¹ Most of them were not straightforward and required more steps. However, in the present case, the intermediate 4-substituted morpholin-2-one 2B.8 was next reacted with various mono- and di-amines in THF to achieve the corresponding mono and diamides 2B.1-2B.4, as outlined in Scheme 2B.1. The benzimidazole – based mono- and diamines which were used in the reaction with 4-substituted morpholin-2-one, were obtained according to the Scheme 2B.1c. O-phenylenediamine was reacted with Boc-protected glycine to afford compound 2B.9 which upon heating in the presence of AcOH led the amine protected benzimidazole 2B.10 in appreciable yield. This compound was next reacted with either n-butyldibromide or m-xylene dibromide to give substituted benzimidazoles such as 2B.12 and 2B.14 respectively. The removal of the Boc- group in the presence of TFA gave desired amines 2B.11, 2B.13 and 2B.15.

Pyrene – based molecules 2B.5 and 2B.6 were synthesized (Scheme 2B.2) according to the same method as followed for anthracene in Scheme 2B.1. All the compounds were fully characterized by spectroscopic techniques.

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2B.2.2. Complexation studies on receptors 2B.1- 2B.4

The ability of the anthracene – labeled chemosensors 2B.1- 2B.4 and pyrene labeled chemosensors 2B.5- 2B.6 toward the sensing of different metal ions was investigated in CH₃CN containing DMSO using fluorescence and UV-vis spectroscopic tools. Prior to study the interaction of all the receptors with the metal ions, we initially studied compound 2B.1. The interaction behavior was followed using UV-vis, fluorescence and NMR spectroscopic tools.

Receptor 2B.1 showed selective sensing of Cu²⁺ by exhibiting significant enhancement in emission in CH₃CN containing 0.04% DMSO. While Cu²⁺ ion showed strong interaction with 2B.1, other important cations such as Zn²⁺ and Hg²⁺ ions exhibited moderate binding. Sensing of Hg²⁺ and Zn²⁺ is also equally important like Cu²⁺. Hg²⁺ is a well known pollutant. Its exposure to human body can lead to neurological diseases.²,³ On the other hand, zinc ion is an abundant component in the human body and plays important role in various fundamental biological processes, such as gene transcription, regulation of metalloenzymes, mammalian reproduction and neural signal transmission.⁴ Therefore sensing of such multiple metal ions by a single chemosensor is attractive. The present example belongs to this class although the chemosensor 2B.1 is more effective in sensing Cu²⁺. Figure 2B.1 shows the change in emission of 2B.1 in the presence of

![Figure 2B.1](image-url)

**Figure 2B.1.** Change in fluorescence ratio of 2B.1 (c = 4.10 x 10⁻⁵ M) upon addition of 15 equiv. amounts of cations at 418 nm.
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15 equiv. amounts of the different metal ions in CH$_3$CN containing 0.04 % DMSO on excitation at 370 nm. It is evident from Figure 2B.1 the receptor is more sensitive to Cu$^{2+}$ ion. Other metal ions except Zn$^{2+}$ and Hg$^{2+}$ merely perturbed the emission of 2B.1.

Upon gradual addition of Cu(ClO$_4$)$_2$ solution to the solution of 2B.1 (c = 4.10 x $10^{-5}$ M) in CH$_3$CN containing 0.04 % DMSO, the structured emission centered at 418 nm was enhanced to the significant extent without showing any other change in the emission spectra. During progression of titration a red shift of 3 nm of the emission peak at 418 nm was observed. Figure 2B.2a, in this regard, shows the change in emission titration spectra with Cu$^{2+}$ ions. Other metal ions except Zn$^{2+}$ and Hg$^{2+}$, revealed relatively insignificant responses in the region manifesting the pronounced OFF - ON type of Cu$^{2+}$-selectivity of 2B.1. In this connection, it is well known that Cu$^{2+}$ is paramagnetic ion with an unfilled 'd' shell and could strongly quench the emission of the fluorophore near it via electron energy transfer. But in our case, the enhancement of emission of 2B.1 after binding of Cu$^{2+}$ ion is quite interesting and mentionable along with other few
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related existing systems. To our opinion, such significant change in emission of 2B.1 in the presence of Cu$^{2+}$ is attributed to the better coordination of Cu$^{2+}$ by aliphatic amine nitrogen, benzimidazole ring nitrogen and amide, alcohol functionalities in the open cavity for which either change in geometry or the flexibility of the molecule is reduced. The consequence of which the metal-fluorophore interaction is modulated$^{8a-8b}$ and the photo induced electron transfer (PET) from the binding sites to the excited state of anthracene is inhibited. In this context, the role of amide carbonyl as blocking moiety for PET can not be ruled out.$^{9a-9c}$ This is also true for Zn$^{2+}$ and Hg$^{2+}$ ions. The difference is

![Figure 2B.3. Fluorescence titration curves (Guest]/[Host] vs change in emission) of 2B.1 (c = 4.10 x 10$^{-5}$ M) measured at 418 nm in CH$_3$CN containing 0.04% DMSO.](image)

Figure 2B.4. UV-vis Job plot for receptor 2B.1 ([H] = [G] = 4.45 x 10$^{-5}$ M) with Cu$^{2+}$ at 365 nm.

![Figure 2B.4. UV-vis Job plot for receptor 2B.1 ([H] = [G] = 4.45 x 10$^{-5}$ M) with Cu$^{2+}$ at 365 nm.](image)

![Figure 2B.5. UV-vis Job plot for receptor 2B.1 (c = 4.45 x 10$^{-5}$) with (a) Zn$^{2+}$, (b) Hg$^{2+}$, measured at 366 nm.](image)
only lying with the extent of change. We believe that it is due to the difference in coordinating ability of the binding site towards Cu$^{2+}$, Zn$^{2+}$ and Hg$^{2+}$ ions.

In the interaction process, the stoichiometries of the Cu-complex, Zn-complex and Hg-complexes were found to be 1:1, as evident from the break of the titration curve (Figure 2B.3) and also Job plots $^10$ (Figure 2B.4 for Cu$^{2+}$ and Figure 2B.5 for Zn$^{2+}$ and Hg$^{2+}$). From the titration data, association constants ($K_a$) for the formation of 2B.1-Cu$^{2+}$, and 2B.1-Hg$^{2+}$ complexes were estimated by nonlinear curve fitting procedure$^{11}$ and found to be $(7.16 \pm 0.473) \times 10^3$ M$^{-1}$, $(5.67 \pm 0.88) \times 10^2$ M$^{-1}$, respectively. We were unable to fit the titration data for Zn$^{2+}$ by nonlinear curve fit method to determine the binding constant value. In addition, we determined the fluorescence enhancement factor (relative indicator of binding strength) of 2B.1 at the emission 418 nm in the presence of 15 equiv. amounts of each metal ion in CH$_3$CN containing 0.04% DMSO to realize the selectivity in the binding process.

![Figure 2B.6](image1.png)

**Figure 2B.6.** Fluorescence enhancement factor (Z) of receptor 2B.1 ($c = 4.10 \times 10^{-5}$ M) upon addition of 15 equiv. amounts of various metal ions.

![Figure 2B.7](image2.png)

**Figure 2B.7:** Change in emission of receptor 2B.1 ($c = 4.10 \times 10^{-5}$ M) upon addition of Cu$^{2+}$ in the presence and absence of other metal ions.

Figure 2B.6, in this regards, shows the plot of fluorescence enhancement factor (Z) in the presence of the different metal ions. From the plot it is evident that the response of
the simple sensor 2B.1 towards Cu\textsuperscript{2+} is significant and also selective over Zn\textsuperscript{2+} and Hg\textsuperscript{2+} ions.

However, to comprehend the selectivity in the sensing of Cu\textsuperscript{2+} by 2B.1, we recorded the emission spectra of 2B.1 upon adding 5 equiv. amounts of Cu\textsuperscript{2+} in the presence of 5 equiv. amounts of other metal ions, examined in the present study. Figure 2B.7 displays the relative view on the change in emission of 2B.1 in the presence of Cu\textsuperscript{2+} when the other metal ions are absent and present in the receptor solution. Small increase in emission upon addition of Cu\textsuperscript{2+} to the solution of 2B.1 containing other metal ions (Figure 2B.7) demonstrates the lower selectivity in the binding process. When the same experiment was done in the absence of Zn\textsuperscript{2+} and Hg\textsuperscript{2+} ions, a better increase in emission of 2B.1 was noted (Fig. 2B.8). This underlined the fact that both Zn\textsuperscript{2+} and Hg\textsuperscript{2+} ions interfere in the binding of Cu\textsuperscript{2+} towards the binding site of 2B.1.

![Figure 2B.8](image_url)

**Figure 2B.8.** Change in emission of 2B.1 (c = 4.10 x 10\textsuperscript{-5} M) upon addition of Cu\textsuperscript{2+} in the presence and absence of other metal ions. a) Receptor 2B.1; b) 2B.1 + 5 equiv. of Cu\textsuperscript{2+}; c) 2B.1 + 5 equiv. of all metals (except Cu\textsuperscript{2+}); d) Receptor 2B.1 + 5 equiv. of all metals + 5 equiv. of Cu\textsuperscript{2+}; e) Receptor 2B.1 + 5 equiv. of all metals (except Cu\textsuperscript{2+}, Zn\textsuperscript{2+}, Hg\textsuperscript{2+}); f) Receptor 2B.1 + 5 equiv. of all metals (except Zn\textsuperscript{2+}, Hg\textsuperscript{2+}) + 5 equiv. of Cu\textsuperscript{2+}; g) Receptor 2B.1 + 5 equiv. of all metals (except Cu\textsuperscript{2+}, Hg\textsuperscript{2+}); h) Receptor 2B.1 + 5 equiv. of all metals (except Hg\textsuperscript{2+}) + 5 equiv. of Cu\textsuperscript{2+}; i) Receptor 2B.1 + 5 equiv. of all metals (except Cu\textsuperscript{2+}, Zn\textsuperscript{2+}); j) Receptor 2B.1 + 5 equiv. of all metals (except Cu\textsuperscript{2+}, Zn\textsuperscript{2+}) + 5 equiv. Cu\textsuperscript{2+}.

Furthermore, to investigate the effect of the counter anion of copper salt in the binding process, change in emission of 2B.1 in CH\textsubscript{3}CN containing 0.04 % DMSO was recorded upon gradual addition of Cu(NO\textsubscript{3})\textsubscript{2} solution (Fig. 2B.9a). The changes in emission (Figure 2B.9a) and absorbance (Figure 2B.9b) of 2B.1 upon addition of Cu(NO\textsubscript{3})\textsubscript{2} were identical.

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to that of Cu(ClO₄)₂. This experimental finding revealed that in the binding of Cu²⁺ ion into the tripodal core of 2B.1, the counter anion has no effect.

**Figure 2B.9**: Change in (a) emission, (b) absorbance of receptor 2B.1 (c = 4.10 x 10⁻⁵ M) upon addition of Cu(NO₃)₂ solution.

To get insight into the enhancing of emission behavior in the 2B.1-Cu²⁺ complex, we investigated the fluorescence decay behavior of 2B.1 itself and in the presence of equiv. amount of Cu²⁺ ion (Figure 2B.10).

**Figure 2B.10**: Fluorescence decays (at λ_max = 418 nm) of receptor 2B.1 upon addition of 1 equiv. of Cu(ClO₄)₂ ([H] = 4.10 x 10⁻⁵ M, [G] = 8.21 x 10⁻⁴ M) in CH₃CN containing 0.04% DMSO.

<table>
<thead>
<tr>
<th>Compound 2B.1 and its complex with Cu²⁺</th>
<th>τ₁ ns (a₁)</th>
<th>τ₂ ns (a₂)</th>
<th>τ₃ ns (a₃)</th>
<th>χ²</th>
</tr>
</thead>
<tbody>
<tr>
<td>2B.1</td>
<td>0.916</td>
<td>4.53</td>
<td>0.035</td>
<td>1.00</td>
</tr>
<tr>
<td></td>
<td>(11.40)</td>
<td>(46.58)</td>
<td>(42.03)</td>
<td></td>
</tr>
<tr>
<td>2B.1 + Cu²⁺ (1:1)</td>
<td>0.967</td>
<td>6.24</td>
<td>0.043</td>
<td>0.997</td>
</tr>
<tr>
<td></td>
<td>(8.30)</td>
<td>(76.63)</td>
<td>(15.07)</td>
<td></td>
</tr>
</tbody>
</table>
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Table 2B.1 summarizes the exponential fit results. Compound 2B.1 (c = 4.10 x 10^{-5} M) exhibited a triexponential fluorescence decay. Upon addition of equiv. amount of Cu^{2+} (c = 8.21 x 10^{-4} M), while the fast decay components were less affected, the time constant (t_2) of the lifetime decay component was greatly affected and contributed to the total emission with a larger preexponential factor.

Furthermore, we investigated the interaction of 2B.1 with the metal ions in aq. CH_3CN and no characteristic change in emission was observed (Fig. 2B.11).

In relation to this we also noticed the interaction of 2B.1 in more polar non aq. solvent such as DMSO. Interestingly, the light red colored solution of 2B.1 in DMSO (c = 2.2 x 10^{-3} M) was discharged in the presence of Cu(ClO_4)_2 and resulted in light green color of the solution. Other metal ions did not bring any detectable color change of the solution of 2B.1.

Figure 2B.11. Change in emission of 2B.1 (c = 3.75 x 10^{-5} M) upon addition of 28 equiv. of a) Zn^{2+}; b) Cu^{2+}; c) Hg^{2+} in CH_3CN containing 11.11% water (in all cases [cation] = 8.21x10^{-4} M) upon excitation at 370 nm.

Figure 2B.12. Change in color of 2B.1 (c = 2.2 x 10^{-3} M) upon addition of 10 equiv. amounts of different metal ions (c = 8.9 x 10^{-4} M) in DMSO: (a) Receptor 2B.1 and with (b) Ni^{2+}; (c) Mn^{2+}; (d) Co^{2+}; (e) Cd^{2+}; (f) Cu^{2+}; (g) Mg^{2+}; (h) Fe^{2+}; (i) Zn^{2+}; (j) Hg^{2+}; (k) Pb^{2+}.
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The appearance of color upon complexation of Cu$^{2+}$ in DMSO is presumably attributed to the charge transfer occurring in between the benzimidazole ligand and metal ion. It is noteworthy that this change in color is pronounced in the high concentration level of 2B.1 ($\sim 10^{-3}$ M) (Figure 2B.12). At the concentration level $\sim 10^{-5}$ M of 2B.1, no change in color was noticeable. However, at the concentration range $\sim 10^{-3}$ M of 2B.1 we carried out the fluorescence titration upon gradual addition of Cu$^{2+}$ ion in DMSO. It is mentionable that the Cu$^{2+}$ induced change in emission of 2B.1 was less compared to the change in CH$_3$CN containing 0.04 % DMSO and thereby demonstrated the role of polar solvent in the interaction process. DMSO being more polar than CH$_3$CN, reduces the interaction of Cu$^{2+}$ at the tripodal core of 2B.1. Similar is the case with Zn$^{2+}$ and Hg$^{2+}$ ions (Fig. 2B.13).

![Figure 2B.13. Change in emission of 2B.1 (c = 4.10 x 10$^{-5}$ M) with a) Zn$^{2+}$, b) Cu$^{2+}$, c) Hg$^{2+}$ in DMSO (in all cases [cation] = 8.21x10$^{-4}$ M) upon excitation at 370 nm.](image)

The UV-vis study of 2B.1 in the presence of all the metal ions except Cu$^{2+}$, under similar conditions, showed minor change in absorbance. For example, Figure 2B.14 shows the

![Figure 2B.14. Change in absorbance of 2B.1 (c = 4.10 x 10$^{-5}$ M) upon gradual addition of (a) Cu$^{2+}$; (b) Zn$^{2+}$; (c) Hg$^{2+}$ ion.](image)
change in absorbance upon gradual addition of \( \text{Cu}^{2+} \), \( \text{Zn}^{2+} \) and \( \text{Hg}^{2+} \). During the interaction with \( \text{Cu}^{2+} \) ions small red shift (\( \Delta \lambda = 5 \text{ nm} \)) of the absorption peak for anthracene occurred. Decrease in absorption with significant red shift of 2B.1 with \( \text{Cu}^{2+} \) is presumably due to cation - \( \pi \) interaction when the metal ion interacts in the tripodal core of 2B.1 in a 1:1 stoichiometric fashion under the guidance of alcohol, amide and benzimidazole groups. The metal coordination property of benzimidazole as well 2-substituted benzimidazole is well established.\(^{12}\)

\[ \text{Figure 2B.15.} \]  Suggested mode of binding of 2B.1 for the metal ions.

\[ \text{Figure 2B.16.} \]  AM1 optimized geometry of 2B.1 in gas phase (\( \mathcal{E} = -194.067 \text{ au} \); the closest distance between anthracene and benzimidazole is 4.89 Å).

It is worth mentioning that the 2-benzimidazole derivatives allow coordination towards metal ions through a variety of sites, with groups bearing nitrogen, oxygen and sulfur atoms and the coordination takes place through imidazolic nitrogen.\(^{12}\) This enable us to presume a binding mode (Figure 2B.15) where the metal ion is anchored at the tripodal core involving the benzimidazole ring nitrogen, aliphatic amine, amide and alcoholic groups altogether.

This is clearly understood from the disposition of the binding moieties in AM1 optimized\(^{13}\) geometry of 2B.1 (Figure 2B.16). The Mulliken charge densities on the different centers are also shown in Figure 2B.16. Closely spaced benzimidazole and anthracene moieties possibly partially shield the space in between them so that the metal ion (\( \text{Cu}^{2+} \)) –
fluorophore interaction is minimized and the PET process from the binding site to the excited state of anthracene is blocked.

We further recorded the $^1$H NMR of 2B.1 in the presence of equiv. amount of Cu$^{2+}$ ion in CDCl$_3$ containing 0.1% d$_6$-DMSO to be acquainted with the involvement of the binding site for complexation (Figure 2B.17). But the NMR spectrum upon addition of Cu$^{2+}$ ions resulted in broadening particularly for the resonances of the benzimidazolyl amide and alcoholic moeties of 2B.1. The anthracenyl ring protons of types 'b' and 'c' moved to the downfield direction by 0.02 ppm and other protons of types 'a' and 'd' were positionally unaffected. The benzimidazole ring protons of types 'e' and 'f' indicated a greater downfield chemical shift ($\Delta\delta = 0.20$ ppm).

Figure 2B.17. Partial $^1$H NMR (400 MHz) of (i) 2B.1 (9.16 x 10$^{-3}$ M); (ii) with 1 equiv. amount of Cu(ClO$_4$)$_2$ in CDCl$_3$ containing 0.1% d$_6$-DMSO.

In addition, the signals for the methylene protons adjacent to the aliphatic amine nitrogen underwent downfield movement by 0.04 – 0.06 ppm. This intimated the information that Cu$^{2+}$ ion shows the preferential coordination at the aliphatic amine nitrogen centre involving the aid of benzimidazoly amide and the alcoholic groups.

Thus the tripodal shaped compound 2B.1, comprising anthracene as fluorophore, benzimidazolyl amide, aliphatic amine and alcohol functionalities as binders/chelators, functions as a new chemosensor for Cu$^{2+}$ in preference to a variety of other metal ions studied except Hg$^{2+}$ and Zn$^{2+}$ ions. Although the tripodal cavity of 2B.1 preferentially binds Cu$^{2+}$ ion, it shows moderate sensing behavior towards Zn$^{2+}$ and Hg$^{2+}$ ions. Cu$^{2+}$ ion binding induced significant enhancement of emission of 2B.1 in CH$_3$CN containing
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0.04% DMSO, and also the change in colour in pure DMSO upon complexation of Cu\(^{2+}\) are the insights into this study for selective sensing of Cu\(^{2+}\) by the new candidate 2B.1.

We next modified the structure 2B.1 by introducing the butyl chain onto the benzimidazole ring nitrogen. This introduced the new structure 2B.2. The characterization of this compound was done by \(^1\)H NMR, \(^{13}\)C, mass etc. The single crystal X-ray\(^{14}\) of 2B.2 further confirmed its structure. Figure 2B.18a shows the ORTEP plot with atom numbering scheme of 2B.2. In the structure two water molecules which may come either from solvent or from atmosphere, were found. This inclusion water exhibited key role in the packing of the molecules. It bridges the molecules involving hydrogen bonding with the benzimidazole ring nitrogen, hydroxyl and amide oxygen centers and gives nice H-bonded polymeric structures.

![Figure 2B.18. (a) ORTEP plot of 2B.2 and packing views down crystallographic (b) b- axis; (c) a- axis (non polar hydrogens are omitted for clarity).](image)

Further modification of the alcoholic part by converting into the xanthate group led to another structure 2B.3. The metal ion binding studies of these two new tripods were performed in the similar way as done for 2B.1. In the ground state the binding was realized by recording the absorption spectra of 2B.2 in the presence of the same metal salts taken for 2B.1. All the metal ions except Co\(^{2+}\), Cu\(^{2+}\) and Zn\(^{2+}\) brought about minor change in absorbance and confirmed their weak interaction. The changes in absorbance in the presence of Co\(^{2+}\), Cu\(^{2+}\) and Zn\(^{2+}\) are shown in Figure 2B.19. In each case the absorption intensity decreases upon gradual addition of metal perchlorate.
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The extent of change in the presence of different metal salts is presumably due to exact fitting of metal ions into the receptors cavity and also the softness or hardness of the binding centers and metal ions. Like 2B.2, the absorption intensity of 2B.3 was measurably perturbed in the presence of Zn$^{2+}$, Co$^{2+}$ and Cu$^{2+}$ (Fig. 2B.20). In both 2B.2 and 2B.3, the absorption peak for anthracene underwent slight red shift on complexation of Cu$^{2+}$ and Co$^{2+}$ ions.

The measurable change in absorbance for 2B.2 and 2B.3 in the presence of Co$^{2+}$ and Cu$^{2+}$ is attributed to the cation - π interaction when the metal ion resides at the core of the tripodal cavity. Such situation is only probable when the alcohol/xanthtate moiety, amide and benzimidazole groups are cooperatively involved in coordination of the metal ion. Interestingly, during interaction the colorless solution of 2B.2 became faint bluish in
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color in the presence of Cu(CIO₄)₂ (Figure 2B.21a). In this color change reaction, the solution of 2B.3 also turned into light green in the presence of Cu²⁺ solution (Figure 2B.21b). Other metal ions did not bring any color change of the solution of 2B.2 and 2B.3. Like 2B.1, the color change of the receptor solutions of both 2B.2 and 2B.3 is possibly due to the charge transfer occurring in between benzimidazole ligand and metal ion.

![Figure 2B.21. Change in color of (a) 2B.2 (c = 2.41 x 10⁻³ M); (b) 2B.3 (c = 2.5 x 10⁻³ M) upon addition of 10 equiv. amounts of different metal ions (c = 7.6 x 10⁻⁴ M) in DMSO.](image)

The fluorescence titration was carried out to understand the excited state selectivity for a metal ion. In this context, Figure 2B.22 illustrates the fluorescence ratios of 2B.2 and 2B.3 in the presence of 15 equiv. amounts of the different metal ions in CH₃CN.

![Figure 2B.22. Change in fluorescence ratio of (a) Receptor 2B.2; (b) Receptor 2B.3 upon addition of 15 equiv. of cations (λ exc = 370 nm).](image)
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containing 0.04 % DMSO. It is evident from Figure 2B.22a that the receptor 2B.2 is more selective to Co\(^{2+}\) ion. Other metal ions except Zn\(^{2+}\) merely perturbed the emission of 2B.2. In case of 2B.3, the maximum change occurred upon addition of Cu\(^{2+}\) ions.

Figure 2B.23. Change in emission of 2B.2 (c = 4.10 \(\times\) 10\(^{-5}\) M) with a) Co\(^{2+}\); b) Zn\(^{2+}\); c) Cu\(^{2+}\) in CH\(_3\)CN containing 0.04% DMSO (in all cases [cation] 8.2\(\times\)10\(^{-4}\) M) upon excitation at 370 nm.

The emission titration spectra for 2B.2 with the metal ions such as Co\(^{2+}\), Cu\(^{2+}\) and Zn\(^{2+}\) are shown in Figure 2B.23. Closure look reveals that the tripod receptor 2B.2 being structurally similar to 2B.1 prefers Co\(^{2+}\) ion in lieu of Cu\(^{2+}\) ion. Only a small change in the structure alters this selectivity.

We believe that the butyl chain in 2B.2 changes the electron density of the imidazole part as well as controls the steric feature. Combination of these possibly brings a subtle change in the cavity dimension due to which the Co\(^{2+}\) ion fits in the cavity according to the same mode as shown in Figure 2B.15.

In contrast, the chemosensor 2B.3 containing xanthate ester showed preferential binding of Cu\(^{2+}\) involving quenching of fluorescence. A marked change in emission spectra of 2B.3 was also noted in the presence of Ni\(^{2+}\) ions (Figure 2B.24). The quenching of emission in these cases is attributed to the activation of PET process occurring in between the binding site and the excited state of anthracene. To our belief, the softness of the xanthate ester moiety has an effect on the selection of Cu\(^{2+}\) and Ni\(^{2+}\) ions.

In the interaction process, the stoichiometry of the Cu-complex and Co-complex were found to be 1:1 both in excited and ground states, as evident from the Job plots (Fig. 2B.25).
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Figure 2B.24. Change in emission of 2B.3 (c = 4.23 x 10^{-5} M) with a) Cu^{2+}; b) Ni^{2+}; c) Fe^{2+} in CH_{3}CN containing 0.04% DMSO (in all cases [cation] 8.46 x 10^{-4} M) upon excitation at 370 nm.

Figure 2B.25. UV-vis Job plots for (a) 2B.2 with Co^{2+} measured at 365 nm ([H] = [G] = 4.10 x 10^{-5} M) and (b) 2B.2 with Cu^{2+} measured at 365 nm ([H] = [G] = 4.23 x 10^{-5} M).

From the titration data, association constants (K_a) for the formation of 2B.2-Co^{2+} and 2B.2-Cu^{2+} complexes were estimated by nonlinear curve fitting procedure\textsuperscript{11} and were found to be (1.61 \pm 0.164) \times 10^{4} \text{ M}^{-1}, (6.03 \pm 1.1) \times 10^{3} \text{ M}^{-1} respectively. We were unable to fit the titration data for other metal ions by nonlinear curve fit method to determine the binding constant value.\textsuperscript{11} Thus on moving from structure 2B.1 to 2B.3, the selectivity profile towards the recognition of metal ions is changed. This happens due to the steric feature of the individual binding arm around the trivalent nitrogen that modulates the cavity size of the tripodal molecules. Also the softness/hardness feature of the binding site contributes to the selectivity of a particular metal ion.
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We recorded the $^1$H NMR of 2B.2 and 2B.3 in the presence of equiv. amount of Co$^{2+}$ and Cu$^{2+}$ ions, respectively in CDCl$_3$ containing 0.1% d$_6$-DMSO to be familiar with the involvement of the binding site for complexation. But the NMR spectra of both the compounds in the presence of the said metal ions became too broad to draw any conclusion.

In order to be acquainted with the sensing behavior of the compound where the structure 2B.1 is present in the multiple forms, we designed and synthesized compound 2B.4. A similar study was performed with this compound and we found a different selectivity. In the design two molecules of 2B.1 are coupled through $m$-xylene spacer involving the benzimidazole ring nitrogen. Thus in 2B.4, two tripodal centres are present in the close vicinity.

Figure 2B.26. Change in fluorescence ratio of 2B.4 upon addition of 2 equiv. amounts of cations.

The metal ion binding properties of 2B.4 towards the metal ions such as Hg$^{2+}$, Cu$^{2+}$, Cd$^{2+}$, Fe$^{2+}$, Mg$^{2+}$, Co$^{2+}$, Ni$^{2+}$, Zn$^{2+}$, Ag$^+$ and Pb$^{2+}$ (taken as their perchlorate salts) were investigated in CH$_3$CN. Interestingly, the compound 2B.4 selectively recognized Zn$^{2+}$ ion in CH$_3$CN by exhibiting a drastic increase in emission of anthracene. In the study no other metal ions except Cd$^{2+}$ showed significant interaction. Compound 2B.4 exhibited weak emission at 412 nm in CH$_3$CN upon excitation at 370 nm. Upon titration with different metal ions, the emission of 2B.4 was changed to the different extents. Figure 2B.26, shows a comparative view when different metal ions were added in four equiv. amounts to the solution of 2B.4 in CH$_3$CN. Figure 2B.27a is the emission titration spectra with Zn$^{2+}$ ions. The inset of Figure 2B.27a represents the associated colour change of the solutions under illumination of UV light. Figure 2B.27b, describes the emission change with concentration of Zn$^{2+}$ in CH$_3$CN. While the emission of 2B.4 at 412 nm increased dramatically in the presence of Zn$^{2+}$, it was moderately perturbed in the
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presence of Cd$^{2+}$. Figure 2B.28 displays the change in emission spectra of 2B.4 in CH$_3$CN upon gradual addition of some selected metal ions. During the titration with Zn$^{2+}$ ions the emission intensity at 412 nm progressively increased with red shift ($\Delta \lambda = 4$ nm). Such significant change in emission of 2B.4 in the presence of Zn$^{2+}$ is attributed to the better coordination of Zn$^{2+}$ by amine nitrogen and amide, alcohol functionalities in the open cavity due to which rigidification of the molecule results in and the photo induced electron transfer (PET) from the binding sites to the excited state of anthracene is inhibited. As Zn$^{2+}$ has closed-shell d-orbitals, energy or charge transfer processes cannot take place.

Figure 2B.27. (a) Change in emission of receptor 2B.4 (c = 2.29 x 10$^{-5}$ M) with Zn$^{2+}$ in CH$_3$CN ([Zn$^{2+}$] 4.58x10$^{-4}$ M, upon excitation at 370 nm); Inset: Colour change of the receptor solution under illumination of UV light; (b) Fluorescence change of 2B.4 with [Zn$^{2+}$] (c = 2.29 x 10$^{-5}$ M).

Figure 2B.28. Change in emission of 2B.4 (c = 2.29 x 10$^{-5}$ M) with (a) Cd$^{2+}$; (b) Hg$^{2+}$; (c) Cu$^{2+}$ in CH$_3$CN ([cation] 4.58x10$^{-4}$ M, upon excitation at 370 nm.)
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However, we determined the fluorescence enhancement factor (Z) of \( \text{2B.4} \) at 412 nm in the presence of 2 equiv. amounts of each metal ion in CH\(_3\)CN. The emission increase is a relative indicator of binding strength. Figure 2B.29, in this regard, shows the plot of fluorescence enhancement factor (Z) in the presence of the different cations. From the plot it is clearly understood that the response of \( \text{2B.4} \) towards Zn\(^{2+}\) is significant and also selective over other metal ions. Such selectivity in the binding process is attributed to the coordination behavior of the alcoholic and amide functional groups along with the trivalent amine nitrogen in \( \text{2B.4} \).

![Figure 2B.29](image)

**Figure 2B.29:** Fluorescence enhancement factor of receptor \( \text{2B.4} \) (c = 2.29 \( \times \) 10\(^{-5}\) M) upon addition of 2 equiv. various metal ions.

To understand the selective sensing of Zn\(^{2+}\) by \( \text{2B.4} \), we recorded the emission spectra of the receptor upon adding 2 equiv. amounts of Zn\(^{2+}\) in the presence of 2 equiv. amounts of other metal ions examined. Figure 2B.30 displays the comparative view on

![Figure 2B.30](image)

**Figure 2B.30.** Fluorescence response of \( \text{2B.4} \) (c =2.29 \( \times \) 10\(^{-5}\) M) to Zn\(^{2+}\) (c = 4.58 \( \times \) 10\(^{-4}\) M) over the selected metal ions (c = 4.58 \( \times \) 10\(^{-4}\) M).
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the change in emission of 2B.4 in the presence of Zn$^{2+}$ when the other metal ions are absent and present in the receptor solution. The greater increase in emission upon addition of Zn$^{2+}$ to the solution of 2B.4 containing other metal ions (Figure 2B.30) corroborates the selectivity in the binding process.

The UV-vis study of 2B.4 in the presence of the same metal ions under similar conditions showed minor change in absorbance, which indicated that the chemosensor 2B.4 behaves as an ideal PET system as interpreted by de Silva et al. For example, Figure 2B.31 shows the change in absorbance upon gradual addition of some metal ions such as Zn$^{2+}$, Cd$^{2+}$ and Cu$^{2+}$.

During interaction small red shift of the absorption peak for anthracene occurred. Decrease in absorption for anthracene (centered at 365 nm) with significant red shift

Figure 2B.31. Change in absorbance of 2B.4 (c =2.29 x 10$^{-5}$ M) upon gradual addition of (a) Zn$^{2+}$; (b) Cd$^{2+}$; (c) Cu$^{2+}$.

Figure 2B.32. UV Job plots for receptor 2B.4 (c = 2.29 x 10$^{-5}$ M) with (a) Zn$^{2+}$ and (b) Cd$^{2+}$ measured at 370 nm.
was noticed during the titration of 2B.4 with Cu$^{2+}$. This is presumably due to cation - $\pi$ interaction that possibly interplays when the metal ion resides in the tripodal cavity of 2B.4 under the guidance of alcohol and amide functional groups. In the ground state interaction, the stoichiometry of the Zn-complex was found to be 1:1 as determined by UV Job plot$^{10}$ (Figure 2B.32a). Similar composition (host: guest = 1:1) of the complexes of 2B.4 with Cd$^{2+}$ was observed in the ground state (Figure 2B.32b).

Interestingly, during the titration the colourless solution of 2B.4 in CH$_3$CN ($c = 8.57 \times 10^{-4}$ M) turned into light yellow and green colors in the presence of Zn(ClO$_4$)$_2$ and Cu(ClO$_4$)$_2$, respectively (Fig. 2B.33a).

![Figure 2B.33. (a) Change in color of 2B.4 in the presence of Zn$^{2+}$ and Cu$^{2+}$ ions in CH$_3$CN; (b) Suggested mode of binding of 2B.4 for the metal ions.](image)

Other metal ions did not bring any detectable color change of the solution of 2B.4. Here also the appearance of color upon complexation of Zn$^{2+}$ and Cu$^{2+}$ in CH$_3$CN is presumably attributed to the charge transfer occurring in between the benzimidazole ligand and metal ion. It is mentionable that this color change is prominent in the high concentration of 2B.4 ($\sim$10$^{-2}$ M).

Binding constant values for Zn$^{2+}$ and Cd$^{2+}$ ions ($K_a$) were found to be (2.80 x 10$^4$ M$^{-1}$) and (2.5 x 10$^4$ M$^{-1}$) respectively. Due to minor change in emission we were unable to determine the binding constant values for other metal ions.

We recorded the $^1$H NMR of 2B.4 in the presence of equiv. amount of Zn$^{2+}$ ion in CDCl$_3$ containing 0.1% d$_6$-DMSO. Upon complexation, the signals for the $-\text{CH}_2-$ groups adjacent to nitrogen centre at the core moved slightly to the downfield direction ($\Delta\delta = 0.02$-0.03 ppm). Other signals did not show any change in their positions. Thus, here also we presumed a binding structure according to the mode shown in Figure 2B.33b.

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The experimental findings reveal that the structural variations in the tripodal structure may result in different sensing behaviors towards the metal ions. The color change of the solutions in some cases is noted to be promising in naked eye detection of the metal ions.

The structural variation in the tripodal structure was next done by replacing the anthracene with pyrene motif. Accordingly, the structures 2B.5 and 2B.6 were designed and synthesized. The synthetic scheme for these two substrates is delineated in scheme 2B.2.

2B.2.3 Complexation studies on receptors 2B.5 and 2B.6

The foregoing discussion describes that alteration of the binding groups in the examples (2B.1-2B.4) gives the different selectivities and sensitivities towards different metal ions. In this section, the observation will be focused on the receptors 2B.5 and 2B.6 that are identical with the receptors 2B.1 and 2B.2 having pyrene as the fluorophore in lieu of anthracene. The characterization of these compounds was done by $^1$H NMR, $^{13}$C, mass etc. The single crystal X-ray$^{16}$ of 2B.6 further confirmed its structure (Figure 2B.34).

Figure 2B.34. (a) ORTEP plot of 2B.6 with atom numbering scheme and (b) packing view down crystallographic b-axis (non polar hydrogens are omitted for clarity).
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While the Figure 2B.34a shows the ORTEP plot of 2B.6 with atom numbering scheme, Figure 2B.34b represents the packing of the molecules down the crystallographic b-axis. In the packing, the π-stacking interaction between the pyrene units is noteworthy. However, in order to evaluate the interaction properties of 2B.5 and 2B.6, the same metal ions (taken as their perchlorate salts) were considered in CH$_3$CN containing 0.1% DMSO (DMSO was used for homogeneity of the solution). The receptor solutions were excited at 340 nm in fluorescence. The emission spectra of 2B.5 showed structured emission. Upon addition of metal salts to the solution of 2B.5, the emission of pyrene increased to the different extents. Figure 2B.35 shows the fluorescence ratio at 393 nm for 2B.5 in the presence of 20 equiv. amounts of different metal perchlorates. Under similar condition, the change in fluorescence ratio for 2B.6 with the same metal ions is

**Figure 2B.35.** Change in fluorescence ratio of 2B.5 (c = 5.93 x 10$^{-5}$ M) upon addition of 20 equiv. of metal ions at 393 nm.

**Figure 2B.36.** Change in fluorescence ratio of 2B.6 (c = 2.43 x 10$^{-5}$ M) upon addition of 16 equiv. of cations at 394 nm.

**Figure 2B.37.** Change in emission spectra of 2B.5 (c = 5.93 x 10$^{-5}$ M) upon addition of Zn$^{2+}$ (c = 1.18 x 10$^{-3}$ M, as perchlorate salt) in CH$_3$CN containing 0.1% DMSO ($\lambda_\text{ex} = 340$ nm).
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represented in Figure 2B.36. While the chemosensor 2B.5 is selective to Zn$^{2+}$ ion, the chemosensor 2B.6 shows a preference for Co$^{2+}$ ion as evident from Figures 2B.35 and 2B.36. Other metal ions weakly perturbed the emission in both cases. Figure 2B.37 displays the change in emission of 2B.5 in CH$_3$CN containing 0.1% DMSO upon gradual addition of zinc perchlorate. While the emission at 393 nm increased dramatically in the presence of Zn$^{2+}$ with the appearance of an unstable nonstructural band at 475 nm, it was negligibly perturbed in the presence of Mg$^{2+}$, Cd$^{2+}$ and Cu$^{2+}$ ions (Figure 2B.38).

![Figure 2B.38](image)

**Figure 2B.38.** Change in emission spectra of 2B.5 ($c = 5.93 \times 10^{-5}$ M) upon addition of (a) Mg$^{2+}$, (b) Cu$^{2+}$ and (c) Cd$^{2+}$ (in all cases concentration of metal salts = $1.18 \times 10^{-3}$ M) in CH$_3$CN containing 0.1% DMSO ($\lambda_{ex} = 340$ nm).

![Figure 2B.39](image)

**Figure 2B.39.** Change in emission spectra of 2B.6 ($c = 2.43 \times 10^{-5}$ M) upon addition of one equiv. of Co$^{2+}$ ion at 394 nm.
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Figure 2B.39 shows the emission titration spectra of **2B.6** upon gradual addition of Co²⁺ ions. The fluorescence intensity of **2B.6** was moderately perturbed in the presence of Zn²⁺. Other cations brought about minor change in emission. Figure 2B.40, in this regard describes the emission titration spectra for **2B.6** in the presence of some selected metal ions.

![Emission spectra](image)

**Figure 2B.40.** Change in emission spectra of **2B.6** (c = 2.43 x 10⁻⁵ M) upon addition of (a) Zn²⁺; (b) Mg²⁺; (c) Cu²⁺ (in all cases concentration of metal salts = 4.86 x 10⁻⁴ M) in CH₃CN containing 0.1% DMSO (λₑₓ = 340 nm).

We also determined the fluorescence enhancement factor (Z) of **2B.5** and **2B.6** at emission 394 nm in the presence of each metal ion in CH₃CN containing 0.1% DMSO.

![Fluorescence enhancement factor](image)

**Figure 2B.41:** Fluorescence enhancement factor of receptor (a) **2B.5** (c = 5.93 x 10⁻⁵ M), (b) **2B.6** (c = 2.43 x 10⁻⁵ M) upon addition of 15equiv. various metal ions.

Figure 2B.41, in this regard, shows the plot of fluorescence enhancement factor (Z) of both the receptors in the presence of the different cations. From the plot it is clearly...
understood that the response of 2B.5 and 2B.6 is significant towards Zn\(^{2+}\) and Co\(^{2+}\), respectively.

**Figure 2B.42.** Fluorescence Job plot of receptor (a) 2B.5 ([G] = [H] = 5.93 x 10\(^{-5}\) M) for Zn\(^{2+}\) at 393 nm, and (b) 2B.6 ([G] = [H] = 2.43 x 10\(^{-5}\) M) for Co\(^{2+}\) at 394 nm.

In the interaction process, the stoichiometry of the Zn\(^{2+}\) - 2B.5 complex and Co\(^{2+}\) - 2B.6 complexes were found to be 1:1, as confirmed by Job plots\(^\text{10}\) (Figure 2B.42). From the titration data, association constants (K\(_a\)) for the formation of 2B.5-Zn\(^{2+}\), and 2B.6-Co\(^{2+}\) complexes were estimated by nonlinear curve fitting procedure\(^\text{11}\) and found to be (2.29 ± 0.43) x 10\(^4\) M\(^{-1}\), (2.86 ± 0.81) x 10\(^4\) M\(^{-1}\), respectively (Fig. 2B.43).

**Figure 2B.43.** Nonlinear curve fitting of the fluorescence titration data for 2B.5 (c = 5.93 x 10\(^{-5}\) M) with (a) Zn\(^{2+}\) and (b) 2B.6 (c = 2.43 x 10\(^{-5}\) M) with Co\(^{2+}\) in CH\(_3\)CN containing 0.1% DMSO.
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Binding constant of other metal complexes were also calculated and found to be less than Zn$^{2+}$ and Co$^{2+}$ complex with receptor 2B.5 and 2B.6 respectively (Table 2B.2).

Table 2B.2. Association constants ($K_a$) by fluorescence method

<table>
<thead>
<tr>
<th>Guests$^a$</th>
<th>$K_a$ in $M^{-1}$</th>
<th>Receptor 2B.5</th>
<th>Receptor 2B.6</th>
</tr>
</thead>
<tbody>
<tr>
<td>$Zn^{2+}$</td>
<td>(2.29 ± 0.43) x $10^4$; $R= 0.981$</td>
<td>(2.02 ± 0.47) x $10^3$; $R= 0.953$</td>
<td>b</td>
</tr>
<tr>
<td>$Cd^{2+}$</td>
<td>(3.17 ± 1.7) x $10^3$; $R= 0.893$</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Cu^{2+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Co^{2+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Mg^{2+}$</td>
<td>(5.46 ± 1.7) x $10^3$; $R= 0.992$</td>
<td>(3.02 ± 0.166) x $10^3$; $R= 0.996$</td>
<td>b</td>
</tr>
<tr>
<td>$Ni^{2+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Fe^{2+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Pb^{2+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
<tr>
<td>$Mn^{3+}$</td>
<td>b</td>
<td>b</td>
<td>b</td>
</tr>
</tbody>
</table>

a. Perchlorate salts were used, b. Not determined due to small change.

The respective OFF - ON type of Zn$^{2+}$ and Co$^{2+}$- selectivities of 2B.5 and 2B.6 was established from fluorescence at 394 nm. Figure 2B.44, in this regard, describes the Zn$^{2+}$ and Co$^{2+}$ ion binding induced change in emission of 2B.5 and 2B.6 in the presence and absence of 5 equiv. amounts of other metal ions. As can be seen from Figure 2B.44, the interference of the metal ions considered in the present study, is established to be negligible.

Figure 2B.44. Fluorescence response of (a) 2B.5 ($c = 5.93 \times 10^{-5}$ M) to Zn$^{2+}$ ($c = 1.18 \times 10^{-3}$ M) over the selected metal ions ($c = 1.18 \times 10^{-3}$ M); (b) 2B.6 ($c = 2.43 \times 10^{-5}$ M) to Co$^{2+}$ ($c = 4.86 \times 10^{-4}$ M) over the selected metal ions ($c = 4.86 \times 10^{-4}$ M).
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As before, to our opinion, such significant increase in emission of both $\textbf{2B.5}$ and $\textbf{2B.6}$ in the presence of $\text{Zn}^{2+}$ and $\text{Co}^{2+}$, respectively is attributed to the best fit of them into the open cavities of the tripodes due to which rigidification results in. As a consequence, the PET is inhibited.$^{17\text{a,b}}$

On closer look it is observed that receptor $\textbf{2B.5}$ is similar to that of receptor $\textbf{2B.1}$ in structure; only the difference lies with the fluorophore pyrene instead of anthracene. Such subtle change in structure brings about different selectivities. While $\textbf{2B.1}$ is selective for $\text{Cu}^{2+}$ ion, receptor $\textbf{2B.5}$ is for $\text{Co}^{2+}$ ions. Thus this difference in selectivity is not only for the binding zone but also for the influences of n- surfaces of fluorophore.

On the other hand, receptors $\textbf{2B.2}$ and $\textbf{2B.6}$ are structurally similar having different fluorophores. But they show similar selectivity i.e towards $\text{Co}^{2+}$ ions. Although the selectivity was same, careful analysis reveals that the $\text{Co}^{2+}$- induced change in emissions of $\textbf{2B.2}$ and $\textbf{2B.6}$ (Fig. 2B.22a and 2B.36) are different. Receptor $\textbf{2B.6}$ exhibited greater change in intensity upon binding with $\text{Co}^{2+}$ ions in comparison to $\textbf{2B.2}$. Both the

![Figure 2B.45. Change in absorbance of (i) $\textbf{2B.5}$ ($c = 5.93 \times 10^{-5}$ M) upon gradual addition of (a) $\text{Zn}^{2+}$; (b) $\text{Cu}^{2+}$; (c) $\text{Co}^{2+}$ ions (ii) $\textbf{2B.6}$ ($c = 2.43 \times 10^{-5}$ M) upon gradual addition of (a) $\text{Co}^{2+}$; (b) $\text{Cu}^{2+}$; (c) $\text{Cd}^{2+}$ ions.](image)
selectivity and enhancement factors (Z) for 2B.6 were greater than that of 2B.2 which also underlined the key role of n-surface of the fluorophore in the binding. Similar to the previous cases UV-vis study of 2B.5 and 2B.6 in the presence of the same metal ions under similar conditions showed minor change in absorbance. For example, Figure 2B.45 shows the change in absorbance upon gradual addition of some selected metal ions.

In the interaction process, 1:1 stoichiometry of the Zn\(^{2+}\) - 2B.5 and Co\(^{2+}\) - 2B.6 complexes in ground state were found to be similar as observed in excited state. The colorless solution of 2B.5 in CH\(_3\)CN containing DMSO (c = 1.2 \times 10^{-3} \text{ M}) was discharged in the presence of Zn(ClO\(_4\))\(_2\) and Cu(ClO\(_4\))\(_2\) and resulted in light yellow and green color of the solution, respectively. Other metal ions did not bring any detectable color change of the solution of 2B.5. The appearance of color upon complexation of Zn\(^{2+}\) and Cu\(^{2+}\) is presumably attributed to the charge transfer occurring in between the benzimidazole ligand and metal ion. It is noteworthy that this change in color is pronounced in the high concentration level of 2B.5 (\(\sim 10^{-3} \text{ M}\)) (Figure 2B.46). Compound 2B.6 did not participate in the color change in the presence of Co\(^{2+}\) ions.

\[\text{Figure 2B.46. Change in colour of 2B.5 in the presence of Zn}^{2+}\text{ and Cu}^{2+}\text{ ions.}\]

\(^1\text{H}\) NMR of 2B.5 in the presence of equiv. amount of Zn\(^{2+}\) ion in CDCl\(_3\) containing 0.1% d\(_6\)-DMSO indicated only a weak downfield movement of the signals (\(\Delta \delta = 0.02\text{-}0.04 \text{ ppm}\)) for the pyrene ring protons. This underlines the fact that Zn\(^{2+}\) ion enters into the cavity according to the suggested mode shown in Figure 2B.15 where the pyrene ring imparts a strong cation-\(\pi\) interaction. Beside this, no other change in the \(^1\text{H}\) NMR spectra
was marked. For 2B.6, we were unable to record the $^1$H NMR spectra with equiv. amount of Co$^{2+}$ ion.

In order to be acquainted with the different selectivities of the tripods, we optimized the geometries of 2B.2, 2B.3, 2B.5 and 2B.6 by AM1 method$^{13}$ and compared with 2B.1 in Figure 2B.16. As can be seen from Figure 2B.47, the change of substituents alters the Mulliken charge densities on the coordinating centers and the cavity space in each case appears to be different. These factors presumably contribute to the interaction process due to which metal sensing behaviors of the tripods has become a function of structural diversity. The detail calculation by DFT method may give more insight and it is in progress.

Figure 2B.47. AM1 optimized geometries of (a) 2B.2, (b) 2B.3, (c) 2B.5 and (d) 2B.6 in gas phase.
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2B.3. Summary and outlook

In conclusion, a series of tripodal-shaped fluoreceptor comprising of amide, alcohol and xanthate ester functionalities have been synthesized. Their binding studies in solution exploiting fluorescence technique towards a series of metal perchlorate have been performed. Fluorescence studies interpret that the molecules are highly efficient to report the presence of a particular metal ion in solution. The sensing behavior has been found to vary with the change in interactional functionalities as well as the fluorophore present around the nitrogen centre of the core of the tripods. It has also been noticed that the tripodal molecules with similar binding sites having different fluorophores exhibit the different binding characteristics in solution. The comparative study, in this regard, has been described thoroughly. The theoretical investigation has been done partially. The details (DFT calculation) along this direction are in progress. We hope a correlation can be made after accumulating the theoretical data. To our belief, the different selectivities of the receptors in the excited state rather than the ground state are due to their structural diversity that controls the dimension of the binding core.

2B.4 Experimental

General

Reagents and solvents were purified using standard techniques. Solvents were dried over the appropriate drying agent following standard procedure. Solvents for spectroscopic measurements were of spectroscopic or HPLC grade. Infrared spectra were recorded on Perkin Elmer L120-00A spectrophotometer. $^1$H NMR spectra were recorded at 400 MHz using Bruker instrument. Mass spectra were recorded on API 2000 LCMS/MS instrument. Fluorescence measurements were done using Perkin Elmer LS 55 and UV-vis absorption spectra were recorded at room temperature using Perkin Elmer Lambda 25.

2-[[Anthracen-9-ylmethyl]-amino]-ethanol (2B.7a):

To a solution of 9-anthraldehyde (1 g, 4.85 mmol) in dry methanol (30 mL) was added ethanolamine (0.3 mL, 4.97 mmol) and the reaction mixture was refluxed for 9 h. The solution was cooled and stirred at 0 °C for 2 h to give yellow precipitate. The precipitate
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was filtered off and washed with methanol for several times and finally dried under vacuum to give foam like yellow solid (1.05 g, 87%), mp: 118-120 °C.

\[^1\text{H} \text{NMR (400 MHz, CDCl}	ext{3): }\delta 9.49 (s, 1H), 8.50 (d, 2H, J = 8 \text{ Hz}), 8.48 (s, 1H), 8.02 (d, 2H, J = 8 \text{ Hz}), 7.55 - 7.47 (m, 4H), 4.09 (s, 4H), 1.57 (s, 1H); \text{FT-IR } \nu \text{ cm}^{-1} (\text{KBr}): 3400, 3254, 2929, 2839, 1640, 1446, 1383, 1059; \text{MS (ESI): } m/z 250.2 [\text{M+H}]^+, 205.2, 179.2.\]

4-(Anthracen-9-ylmethyl)morpholin-2-one (2B.8):

To a stirred solution of Schiff-base 2B.7a (0.8 g, 3.21 mmol) in dry methanol (30 mL) NaBH\textsubscript{4} (0.305 g, 8.03 mmol) was added portion wise at 0 °C under nitrogen atmosphere. The reaction mixture was refluxed for 3 h. The solvent was removed under vacuum; water was added and extracted with CH\textsubscript{2}Cl\textsubscript{2} (30 mL x 3). The organic layer was separated and dried over Na\textsubscript{2}SO\textsubscript{4} and the solvent was removed in a rotary evaporator. Finally, the crude product was purified by column chromatography using 2% methanol in CHCl\textsubscript{3} as eluent to give brownish solid 2B.7b (0.62 g, 77%), mp: 84-86 °C. FTIR: \( \nu \text{ cm}^{-1} (\text{KBr}) \) 3307, 2922, 2850, 1623, 1445, 1050.

To a solution of 2B.7b (2.0 g, 7.96 mmol) in dry acetone (40 mL), K\textsubscript{2}CO\textsubscript{3} (1.54 g, 11.14 mmol) was added followed by addition of ethyl 2-chloroethanoate (1.37 g, 11.14 mmol). The reaction mixture was refluxed for 7 h under nitrogen atmosphere. After completion of the reaction, K\textsubscript{2}CO\textsubscript{3} was filtered off and the filtrate was evaporated on a rotary evaporator. Water was added to the residue and the aqueous layer was extracted with CH\textsubscript{2}Cl\textsubscript{2} (30 mL x 3). The combined organic layer was dried over anhydrous Na\textsubscript{2}SO\textsubscript{4} and concentrated in vacuo. Purification of this crude product by column chromatography gave the lactones 2B.8 (1.80 g, 77%) as light yellow solid, mp 159 °C.

\[^1\text{H} \text{NMR (400 MHz, CDCl}	ext{3): }\delta 8.46 (s, 1H), 8.36 (d, 2H, J = 8 \text{ Hz}), 8.02 (d, 2H, J = 8 \text{ Hz}), 7.56-7.46 (m, 4H), 4.52 (s, 2H), 4.24 (t, 2H, J = 4.8 Hz), 3.56 (s, 2H), 2.74 (t, 2H, J = 4 Hz); \text{^13C NMR (100 MHz, CDCl}	ext{3): }167.5, 131.3, 131.2, 129.2, 128.3, 127.0, 126.2, 125.0, 124.3, 69.0, 56.1, 52.7, 48.2; \text{FT-IR } \nu \text{ cm}^{-1} (\text{KBr}): 3050, 2916, 2846,1747, 1734, 1622; \text{MS (ESI): } m/z 292.2 [\text{M + H}]^+.\]
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tert-butyl (1H-benzo[d]imidazol-2-yl)methylcarbamate (2B.10):

To a stirred solution of Boc-protected glycine acid (0.323 g, 1.85 mmol), in dry CH₂Cl₂ (30 mL), DCC (0.383 g, 1.85 mmol) and a catalytic amount of DMAP were added at 0°C. The solution was stirred for 30 min. and a solution of o-phenylenediamine (1.0 g, 9.25 mmol) in dry CH₂Cl₂ was added to it under nitrogen atmosphere. After stirring for 19 h, the reaction mixture was filtered off to remove insoluble DCU. The filtrate was removed in vacuo and the crude product was purified by silica gel column chromatography using 1% methanol in chloroform to afford the compound 2B.9 (0.321 g, yield 65.4%), FT-IR ν cm⁻¹ (KBr): 3420, 3339, 3257, 3044, 2980, 1703, 1678, 1538.

Compound 2B.9 (0.2 g, 0.754 mmol) was heated in acetic acid (0.3 ml) at 60°C for 2 h. Then the reaction was quenched by addition of aq solution of NaHCO₃ (15 mL). The reaction mixture was next extracted with CHCl₃-MeOH mixture solvent (CHCl₃: MeOH = 3:1 v/v; 15 mL) and the organic layer washed with brine, dried over Na₂SO₄. The residue was purified by silica gel column chromatography using 40% petroleum ether in ethyl acetate to afford the compound 2B.10 (0.165 g, yield 88.5%), mp 174°C. ¹H NMR (400 MHz, CDCl₃): δ 10.30 (s, 1H), 7.70 (br s, 1H), 7.52 (br s, 1H), 7.25 - 7.23 (m, 2H), 5.55 (br t, 1H), 4.51 (d, 2H, J = 8 Hz), 1.30 (s, 9H); FT-IR: ν cm⁻¹ (KBr): 3348, 2978, 2931, 2751, 1688, 1529; MS (ESI): m/z 248.1 [M + H]⁺.

tert-butyl (1-butyl-1H-benzo[d]imidazol-2-yl)methylcarbamate (2B.12):

To a stirred solution of 2B.10 (0.3 g, 1.2 mmol) in THF (20 mL), NaH (0.029 g, 1.2 mmol) was added and the reaction mixture was refluxed. After ½ h butyl bromide (0.182 g, 1.3 mmol) added dropwise and the mixture was refluxed further for 4h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 15% petroleum ether in ethyl acetate as eluent yielded the product 2B.12 (0.3 g, 82%), mp 126-128°C. ¹H NMR (400 MHz, CDCl₃): δ 7.72 (dd, 1H, J₁ = 8 Hz, J₂ = 4 Hz), 7.35 (dd, 1H, J₁ = 8 Hz, J₂ = 4 Hz), 7.29-7.26 (m, 2H), 5.58 (br t, 1H), 4.60 (d, 2H, J = 8 Hz), 4.16 (t, 2H, J = 8 Hz), 1.82 - 1.74 (m, 2H), 1.47 (s, 9H), 1.42 - 1.37 (m,
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2H), 0.95 (t, 3H, \( J = 4 \) Hz); FTIR: \( \nu \text{ cm}^{-1} \) (KBr): 3328, 3040, 2941, 2888, 1916, 1797, 1683, 1615, 1506, 1457.

\textbf{N-\{(1H-benzo[d]imidazol-2-yl)methyl\}-2-\{\text{anthracen-9-ylmethyl}\}(2-hydroxyethyl)amino \text{ acetamide}} (2B.1):

Compound 2B.10 (0.15 g, 0.60 mmol) was dissolved in 50% TFA in CH\(_2\)Cl\(_2\) and the solution was stirred for 3 h. After that, the reaction was quenched by the addition saturated aq solution of NaHCO\(_3\) (15 mL). The aqueous solution was next extracted with CHCl\(_3\)-MeOH mixture solvent (CHCl\(_3\)-MeOH = 3:1 v/v; 15 mL) and the organic layer was washed with brine, dried over Na\(_2\)SO\(_4\) to afford the desired amine 2B.11 (0.085 g, yield 95%) which was almost pure to use in the next step.

To a stirred solution of lactones, 2B.8 (0.18 g, 0.62 mmol) in THF (20 mL), amine 2B.11 (0.091 g, 0.617 mmol) dissolved in THF (10 mL) was added dropwise. Stirring was continued for 4 h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl\(_3\) (25 mL x 3) and dried over anhydrous Na\(_2\)SO\(_4\). Purification of the crude mass by silica gel column chromatography using 3% CH\(_3\)OH in CHCl\(_3\) as eluent yielded the product 2B.1 (0.24 g, 88%), mp 122°C. \(^1\)H NMR (400 MHz, CDCl\(_3\) containing two drops of \( d_6 \)-DMSO): \( \delta \) 11.56 (s, 1H), 8.58 (d, 2H, \( J = 8 \) Hz), 8.41 (s, 1H), 8.29 (t, 1H, \( J = 8 \) Hz), 7.98 (d, 2H, \( J = 8 \) Hz), 7.53 (t, 3H, \( J = 8 \) Hz), 7.42 (t, 3H, \( J = 8 \) Hz), 7.22 - 7.18 (m, 2H), 4.75 (s, 2H), 4.22 (d, 2H, \( J = 4 \) Hz), 3.95 (t, 2H, \( J = 4 \) Hz), 3.11 (s, 2H), 3.07 (t, 2H, \( J = 4 \) Hz), 1.30 (br s, 1H); \(^13\)C NMR (100 MHz, \( d_6 \)-DMSO): 171.0, 152.0, 142.0, 134.0, 130.9, 129.5, 128.7, 127.4, 126.0, 124.9, 124.8, 123.0, 122.0, 119.0, 111.0, 58.8, 57.4, 56.5, 50.5, 36.4 (one carbon in the aromatic region is unresolved); FT-IR \( \nu \text{ cm}^{-1} \) (KBr): 3243, 3054, 2949, 2853, 1644, 1624; MS (ESI): \( m/z \) 439.1 [M + H]^+.

\textbf{N-\{(1H-benzo[d]imidazol-2-yl)methyl\}-2-\{\text{anthracen-9-ylmethyl}\}(2-hydroxyethyl)amino \text{ acetamide}} (2B.2):

Compound 2B.12 (0.17 g, 0.581 mmol) was dissolved in 50% TFA in CH\(_2\)Cl\(_2\) and the solution was stirred for 3 h. After that, the reaction was quenched by the addition saturated aq solution of NaHCO\(_3\) (15 mL). The aqueous solution was next extracted with
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CHCl₃-MeOH mixture solvent (CHCl₃-MeOH = 3:1 v/v; 15 mL) and the organic layer washed with brine, dried over Na₂SO₄ to afford the desired amine 2B.13 (0.22 g, yield 95%) which was almost pure to use in the next step.

To a stirred solution of lactone 2B.8 (0.18 g, 0.62 mmol) in THF (20 mL), amine 2B.13 (0.1 g, 0.528 mmol) dissolved in THF (10 mL) was added dropwise. Stirring was continued for 4 h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 3% CH₃OH in CHCl₃ as eluent yielded the product 2B.2 (0.24 g, 82%), mp 122 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.64 (d, 2H, J = 8 Hz), 8.43 (s, 1H), 8.37 (t, 1H, J = 8 Hz), 8.0 (d, 2H, J = 8 Hz), 7.75 (dd, 1H, J₁ = 8 Hz, J₂ = 4 Hz), 7.50 (t, 2H, J = 8 Hz), 7.40 (t, 2H, J = 8 Hz), 7.29- 7.27 (m, 3H), 4.77 (s, 2H), 4.22 (d, 2H, J = 4 Hz), 4.01- 3.94 (m, 4H), 3.13- 3.11 (br s, 4H), 1.69- 1.62 (m, 2H), 1.32- 1.26 (m, 2H), 0.88 (t, 3H, J = 8 Hz) (alcoholic –OH not found due to broadening); ¹³C NMR (100 MHz, CDCl₃): δ 172.1, 151.2, 141.4, 134.9, 131.4, 131.3, 129.1, 128.6, 128.1, 126.3, 124.9, 124.5, 122.7, 122.3, 119.0, 109.5, 61.2, 58.7, 56.6, 51.1, 43.5, 34.9, 31.7, 20.1, 13.6; FT-IR ν cm⁻¹ (KBr): 3551, 3286, 3085, 2956, 2868, 1778, 1660, 1623, 1533, 1474; MS (ESI) m/z 495.1 [M+H]⁺, Anal. Calcd for C₃₁H₃₄N₄O₂: C, 75.28; H, 6.93; N, 11.33 Found: C, 75.33; H, 6.96; N, 11.32.


A mixture of 2B.2 (0.15 g, 0.303 mmol), sodium hydride (60%; 0.37 mmol), and imidazole (0.07 mmol) in THF (20 mL) was stirred for 3 h under nitrogen atmosphere at room temperature. Carbon disulfide (0.115 g, 1.52 mmol) was added and the mixture was stirred for 1/2 h. Then, CH₃I (0.205 g, 1.45 mmol) was added and stirring was continued for 1/2 h. After completion of reaction, THF was evaporated off and water was added to the residue. The aqueous layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 10% petroleum ether in ethyl acetate as eluent gave the product 2B.3 (0.14 g, 80%), mp 68-70 °C.
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^1H NMR (400 MHz, CDCl₃): δ 8.34 (d, 2H, J = 8 Hz), 8.30 (s, 1H), 7.83 (d, 2H, J = 8 Hz), 7.75 (dd, 1H, J₁ = 8 Hz, J₂ = 4 Hz), 7.47 (br t, 1H), 7.41 (t, 2H, J = 8 Hz), 7.30-7.29 (m, 2H), 7.24-7.20 (m, 3H), 4.94 (t, 2H, J = 4 Hz), 4.74 (s, 2H), 4.36 (d, 2H, J = 4 Hz), 3.54 (t, 2H, J = 4 Hz), 3.31 (t, 2H, J = 4 Hz), 3.22 (s, 2H), 2.47 (s, 3H), 1.26-1.19 (m, 2H), 1.10-1.03 (m, 2H), 0.66 (t, 3H, J = 8 Hz); 13C NMR (100 MHz, CDCl₃): δ 170.6, 149.7, 142.3, 134.1, 131.2, 131.0, 129.0, 128.3, 127.6, 127.2, 126.7, 124.9, 123.7, 122.5, 121.8, 119.7, 109.7, 70.7, 56.7, 55.1, 50.9, 43.1, 35.8, 31.3, 29.7, 19.8, 13.4; FT-IR ν cm⁻¹ (KBr): 3565, 3092, 3071, 2933, 2690, 1983, 1853, 1791, 1763, 1598, 1517; MS (ESI): m/z 585.1 [M+H]^+, Anal. Calcd for C₃₃H₃₆N₄O₂S₂: C, 67.78; H, 6.20; N, 9.58, Found: C, 67.82; H, 6.29, N, 9.60.

2-[Anthracen-9-ylmethyl-(2-hydroxy-ethyl)-amino]-N-{1-[3-[2-{[anthracen-9-ylmethyl-(2-hydroxy-ethyl)-amino]-acetylamino}-methyl]-benzoimidazol-1-ylmethyl]-benzyI}-1H-benzoimidazol-2-ylmethyl)-acetamide (2B.4):

To a stirred solution of 2B.10 (0.4 g, 0.67 mmol) in THF (20 mL), NaH (0.020 g, 0.81 mmol) was added and the reaction mixture was refluxed. After ½ h, 1, 3- bis (bromo methyl) benzene (0.083 g, 0.304 mmol) was added dropwise. The solution mixture was refluxed for further for 4h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 20% petroleum ether in ethyl acetate as eluent yielded the product 2B.14 (0.16 g, 88 %), mp 182 °C.

^1H NMR (400 MHz, CDCl₃): δ 7.76 (d, 2H, J = 8 Hz), 7.26-7.21 (m, 2H), 7.17 (t, 2H, J = 8 Hz), 7.07 (d, 2H, J = 8 Hz), 6.88- 6.83 (m, 4H), 5.53 (br, 1H), 5.37 (s, 4H), 4.56 (d, 4H, J = 4 Hz); 13C NMR (100 MHz, CDCl₃): δ 155.7, 151.6, 142.2, 138.7, 135.6, 128.8, 128.6, 126.9, 123.3, 123.0, 122.3, 119.5, 110.0, 79.9, 47.0, 37.7, 28.3; FT-IR ν cm⁻¹ (KBr): 3351, 3053, 2935, 1681, 1614, 1534; MS (ESI): m/z 597.3 [M+H]^+.

Compound 2B.14 (0.15 g, 0.251 mmol) was then dissolved in 50% TFA in CH₂Cl₂ and the solution was stirred for 3 h. After that, the reaction was quenched by the addition saturated aq solution of NaHCO₃ (15 mL). The aqueous solution was next extracted with CHCl₃-MeOH mixture solvent (CHCl₃-MeOH = 3:1 v/v; 15 mL) and the organic layer washed with brine, dried over Na₂SO₄ to afford the desired amine 2B.15 (0.085 g, yield 86%) which was almost pure to use in the next step.
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To a stirred solution of lactone 2B.8 (0.124 g, 0.428 mmol) in THF (20 mL), amine 2B.15 (0.085 g, 0.214 mmol) dissolved in THF (10 mL) was added dropwise. Stirring was continued for 4 h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 3% CH₃OH in CHCl₃ as eluent yielded the product 2B.4 (0.24 g, 88%). *H NMR (400 MHz, CDCl₃): δ 8.58 (d, 4H, J = 8 Hz), 8.39 (s, 2H), 8.30 (t, 2H, J = 4 Hz), 7.95 (d, 4H, J = 8 Hz), 7.77 (d, 2H, J = 8 Hz), 7.48 (t, 4H, J = 8 Hz), 7.38 (t, 4H, J = 8 Hz), 7.21 (t, 1H, J = 8 Hz), 7.11-7.07 (m, 4H), 7.02 (d, 2H, J = 8 Hz), 6.77 (d, 2H, J = 8 Hz), 6.62 (s, 1H), 5.06 (s, 4H), 4.76 (s, 4H), 4.06 (d, 4H, J = 4 Hz), 3.96 (s, 4H), 3.10 (s, 8H), 1.81 (br s, 2H); ¹³C NMR (100 MHz, CDCl₃): δ 172.0, 151.5, 141.2, 136.1, 134.8, 131.4, 131.3, 129.7, 129.1, 128.5, 128.1, 127.2, 126.3, 125.8, 124.9, 124.5, 124.2, 123.2, 122.6, 119.1, 109.5, 60.8, 58.7, 56.6, 51.0, 46.4, 35.0; FT-IR ν cm⁻¹ (KBr): 3341, 2870, 1668, 1522, 1464, 1447; MS (ESI): m/z 979.7 [M+H]+, Anal. Calcd for C₆₂H₅₈N₈O₄: C, 76.05; H, 5.97; N, 11.44, Found: C, 76.10; H, 6.01; N, 11.41.

2-(pyrene-4-ylmethylamino) ethanol (2B.17):

To a solution of pyrene-4-carbaldehyde (0.4 g, 1.74 mmol) in dry benzene (30 mL) ethanolamine (0.3 ml, 4.97 mmol) was added and the reaction mixture was refluxed for 15 h. The solution was cooled and stirred at 0 °C for 2 h to give yellow precipitate. The precipitate was filtered off and washed with methanol for several times and finally dried under vacuum to give foam like yellow solid 2B.16 (0.4 g, 84%). Without characterization, compound 2B.16 (0.4 g, 1.46 mmol) was stirred at rt in dry CH₃OH (30 mL). Then NaBH₄ (0.305 g, 8.03 mmol) was added portion wise at 0 °C under nitrogen atmosphere. The reaction mixture was refluxed for 3 h. The solvent was removed under vacuum; water was added and extracted with CH₂Cl₂ (30 mL x 3). The organic layer was separated and dried over Na₂SO₄ and the solvent was removed in a rotary evaporator. Finally, the crude product was purified by column chromatography using 2% methanol in CHCl₃ as eluent to give brownish solid 2B.17 (0.62 g, 77%), mp: 84-86 °C. *H NMR (400 MHz, CDCl₃): δ 8.32 (d, 1 H, J = 8 Hz), 8.16 (d, 2H, J = 8 Hz), 8.10 (d, 2H, J = 8 Hz), 8.02-8.01 (m, 2H), 7.99-7.94 (m, 2H), 4.47 (s, 2H), 3.68 (t,
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4-(pyrene-4-ylmethyl)morpholine-2-one (2B.18):

To a solution of 2B.10 (0.4 g, 1.45 mmol) in dry acetone (40 mL), K₂CO₃ (1.54 g, 11.14 mmol) was added followed by addition of ethyl 2-chloroethanoate (0.27 g, 2.18 mmol). The reaction mixture was refluxed for 10 h under nitrogen atmosphere. After completion of the reaction, K₂CO₃ was filtered off and the filtrate was evaporated on a rotary evaporator. Water was added to the residue and the aqueous layer was extracted with CH₂Cl₂ (30 mL x 3). The combined organic layer was dried over anhydrous Na₂SO₄ and concentrated in vacuo. Purification of this crude product by column chromatography gave the lactone 2B.18 (0.3 g, 66%) as light yellow gummy solid. ¹H NMR (400 MHz, CDCl₃): 8 8.43 (d, 1H, J = 8 Hz), 8.23- 8.19 (t, 2H, J = 8 Hz), 8.17- 8.12 (m, 2H), 8.07-8.02 (m, 2H), 7.96- 7.91 (m, 2H), 4.33 (t, 2H, J = 4 Hz), 4.24 (s, 2 H), 3.51 (s, 2H), 2.74 (t, 2H, J = 4 Hz); FT-IR ν cm⁻¹ (KBr): 3432, 2988, 2957, 2846, 2766, 1877, 1728, 1648, 1588, 1465; MS (ESI): m/z 316.3 [M+H]⁺.

N-((1 H-benzo[d]imidazol-2-yl)methyl-2-((2-hydroxyethyl)pyrene-4-ethyl)amino)acetamide (2B.5):

To a stirred solution of lactone 2B.18 (0.16 g, 0.50 mmol) in THF (20 mL), amine 2B.11 (0.074 g, 0.50 mmol) dissolved in THF (10 mL) was added dropwise. The reaction mixture was stirred for further 4 h. After completion of reaction, monitored by TLC, THF was evaporated off and water was added to the residue. The aq. layer was extracted with CHCl₃ (25 mL x 3) and dried over anhydrous Na₂SO₄. Purification of the crude mass by silica gel column chromatography using 3% CH₃OH in CHCl₃ as eluent yielded the product 2B.5 (0.198 g, 84 %), mp 165-168 °C. ¹H NMR (400 MHz, CDCl₃ containing few drops of d₆-DMSO): 8 11.63 (s, 1H), 8.75 (d, 1H, J = 8 Hz), 8.51 (t, 1H, J = 8 Hz), 8.17 (d, 2H, J = 8 Hz), 8.12- 8.04 (m, 5H), 8.01- 7.96 (m, 2H), 7.66 (br s, 1H), 7.21- 7.20 (m, 2H), 4.44 (s, 2H), 4.30 (d, 2H, J = 4 Hz), 3.92 (d, 2H, J = 4 Hz), 3.20 (s, 2H), 3.01 (s, 2H), (Signal for –OH not observed due to broadening); ¹³C NMR (100 MHz, d₆-DMSO): 8 171.0, 152.1, 142.0, 134.0, 132.4, 130.7, 130.3,
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130.2, 129.2, 128.5, 127.3, 127.1, 127.0, 126.1, 125.1, 125.0, 124.5, 124.0, 123.9, 123.8, 121.3 (2 C unresolved), 118.2, 111.2, 58.7, 57.5, 57.4, 57.0, 36.6; FT-IR $\nu$ cm$^{-1}$ (KBr): 3294, 3226, 2954, 2807, 1662, 1622, 1603, 1531; MS (ESI): m/z 463.0 [M+H]$^+$. Anal. Calcd for C$_{29}$H$_{26}$N$_4$O$_2$: C, 75.30; H, 5.67; N, 12.11. Found: C, 75.33; H, 5.71; N, 12.09.

1, N-((1-butyl-1H-benzo[d]imidazol-2-yl)methyl)-2-(((3,6-dihydropyren-4-yl)methyl)(2-hydroxyethyl)amino)acetamide (2B.6):

To a stirred solution of lactone 2B.18 (0.15 g, 0.47 mmol) in THF (20 ml), amine 2B.13 (0.97 g, 0.047 mmol) dissolved in THF (10 ml) was added dropwise and the reaction mixture was stirred for 4 h. After completion of reaction, monitored by TLC, THF was evaporated and water was added to the residue. The aqueous layer was extracted with CHCl$_3$ (25 mL x 3) and dried over anhydrous Na$_2$SO$_4$. Purification of the crude mass by silica gel column chromatography using 3% CH$_3$OH in CHCl$_3$ as eluent yielded the product 2B.6 (0.20 g, 82 %), mp 130-132 °C. $^1$H NMR (400 MHz, CDCl$_3$): $\delta$ 8.82 (d, 1H, $J = 8$ Hz), 8.51 (br t, 1H), 8.17- 8.11 (m, 2H), 8.05 (s, 2H), 8.02- 8.0 (m, 2H), 7.97- 7.93 (m, 2H), 7.78- 7.76 (m, 1H), 7.30- 7.27 (m, 3H), 4.47 (s, 2H), 4.17 (d, 2H, $J = 4$ Hz), 3.98 (t, 2H, $J = 4$ Hz), 3.84 (t, 2H, $J = 4$ Hz), 3.20 (s, 2H), 3.08 (t, 2H, $J = 4$ Hz), 1.80 (br s, 1H), 1.60- 1.48 (m, 2H), 1.25- 1.14 (m, 2H), 0.80 (t, 3H, $J = 8$ Hz); $^{13}$C NMR (100 MHz, CDCl$_3$): $\delta$ 171.9, 151.1, 141.4, 134.8, 131.5, 131.3, 131.2, 130.7, 130.0, 128.7, 127.7, 127.4, 127.3, 125.9, 125.2, 125.1, 125.0, 124.6, 124.5, 124.1, 122.8, 122.4, 119.0, 109.5, 61.0, 58.9, 57.2, 43.3, 35.0, 31.5, 19.9, 13.5 (1C in the aliphatic region is not found presumably due to overlapping); FT-IR $\nu$ cm$^{-1}$ (KBr): 3672, 3356, 3221, 2935, 2875, 1688, 1650, 1538, 1475; MS (ESI): m/z 519.7 [M+H]$^+$. Anal. Calcd for C$_{33}$H$_{34}$N$_4$O$_2$: C, 76.42; H, 6.61; N, 10.80. Found: C, 76.47; H, 6.59; N, 10.77.

General procedure of fluorescence titration

Stock solutions of the hosts and guests were prepared in CH$_3$CN or CH$_3$CN-DMSO and 2 mL of the individual host solution was taken in the cuvette. The solution was irradiated at the excitation wavelength maintaining the excitation and emission slits. Upon addition of guest, the change in fluorescence emission of the host was noticed. The corresponding emission values during titration were noted and used for the determination of binding constant values.
General procedure of UV-vis titration

The receptors were dissolved in dry UV grade CH₃CN or CH₃CN-DMSO and 2 ml of the individual host solution was taken in the cuvette. Then, guests, dissolved in dry CH₃CN were individually added in different amounts to the receptor solution.

Method for Job plot

The stoichiometry was determined by the continuous variation method. In this method, solutions of host and guests of equal concentrations were prepared in dry CH₃CN or CH₃CN-DMSO. Then host and guest solutions were mixed in different proportions maintaining a total volume of 3 mL of the mixture. The related compositions for host:guest (v/v) were 3:0, 2.8:0.2; 2.5:0.5, 2.2:0.8, 2:1, 1.8:1.2, 1.5:1.5, 1:2, 0.8:2.2, 0.5:2.5, 0.2:2.8. All the prepared solutions were kept for 1 h with occasional shaking at room temperature. Then emission and absorbance of the solutions of different compositions was recorded. The concentration of the complex i.e., [HG] was calculated using the equation \([HG] = \Delta I/I_0 \times [H]\) or \([HG] = \Delta A/A_0 \times [H]\) where \(\Delta I/I_0\) and \(\Delta A/A_0\) indicate the relative emission and absorbance intensities. [H] corresponds the concentration of pure host. Mole fraction of the host \((X_H)\) was plotted against concentration of the complex \([HG]\). In the plot, the mole fraction of the host at which the concentration of the host-guest complex concentration \([HG]\) is maximum, gives the stoichiometry of the complex.

Determination of fluorescence enhancement factor \((Z)\)

Fluorescence enhancement factor \((Z)\) was calculated based on the equation \(Z = (F/F_0) [(V_0 + V)/V_0]\) where \(F\) = observed fluorescence, \(F_0\) = fluorescence of sample before guest addition, \(V_0\) = volume before addition of guest, \(V\) = volume after addition of guest.

Binding constant determination
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Binding constant values for receptors with the anionic guests were determined by nonlinear curve fitting procedure. The non linear fit was done using the equation 1.

\[ I = I_0 + \frac{(I_{\text{lim}} - I_0)/2}{C_H} \left\{ C_H + C_G + \frac{1}{K_a} - \left[ \left( C_H + C_G + \frac{1}{K_a} \right)^2 - 4C_H C_G \right]^{1/2} \right\} \]

Where \( I \) represents the intensity; \( I_0 \) represents the intensity of pure host; \( C_H \) and \( C_G \) are corresponding concentrations of host and anionic guest; \( K_a \) is the binding constant. The binding constant \( K_a \) and correlation coefficients (R) were obtained from a non-linear least-square analysis of \( I \) versus \( C_H \) and \( C_G \).

X-ray crystal

Data were collected with Nonius KappaCCD diffractometer. Programs used: data collection COLLECT,\textsuperscript{19} data reduction Denzo- SMN,\textsuperscript{20} absorption correction Denzo,\textsuperscript{21} structure solution SHELXS-97,\textsuperscript{22} structure refinement SHELXL-97,\textsuperscript{23} graphics XP (Bruker AXS, 2000). \( R \)- values are given for the observed reflections, \( WR^2 \)- values for all reflections.
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2B.5. References

2. (a) Harris, H.H.; Pickering, I.J.; George, G.N. Science, 2003, 301, 1203.
13. AM1 calculation was performed using minimal valence basis as STO 3G in Argus Lab 4.0.1, copyright (c) 1972- 2004 Mark Thompson and Planaria Software LLC, http://www.Arguslab.com.
14. Crystal data of 2B.2: C_{31}H_{34}N_{4}O_{2} * 2H_{2}O, triclinic, P-1 (No.2), a = 9.7055(1) Å, b = 10.5518(4) Å, c = 15.6162(4) Å, α = 94.348(2)°, β = 92.796(2)°, γ = 113.157(1)°, V = 1460.78(7) Å³, T = 223(2) K, Z = 2, D_{saxd} = 1.206 mg m⁻³, 20127 total reflections of which 4988 were independent, 3919 observed [I > 2σ (I)]. Structure solution and refinement with SHELXS-97 and SHELXL-97, final refinement against F² with 370 parameters, R₁ [I > 2σ (I)] = 0.0463, wR² = 0.1144.
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16. Crystal data of 2B.6: C₃₃H₃₄N₄O₂, triclinic, P-1 (No.2), a = 9.2847(4) Å, b = 10.6148(2) Å, c = 16.1534(5) Å, α = 71.762(1)°, β = 89.626(3)°, γ = 64.890(2)°, V = 1354.29(8) Å³, T = 223(2) K, Z = 2, D_calk = 1.272 mg m⁻³, 16088 total reflections of which 4562 were independent, 3996 observed [I > 2σ (I)]. Structure solution and refinement with SHELXS-97 and SHELXL-97, final refinement against F² with 357 parameters, R, [I > 2σ (I)] = 0.0509, wR² = 0.1297.


