Chapter 1

Introduction
1.1 Chemistry of Soft Interactions

The existence of life on the Earth makes this planet a very special one in the universe. Among many factors which help the life to grow on the Earth, the non-covalent interactions are responsible to make the biochemical processes go on in a specific way as required which makes a thing ‘living’. With advances in science and technology we are still trying to understand the biological processes taking place in a cell, in a living system and thus in human body. It helps in making our life healthier. Understanding the biological processes help us to develop more specific and more potent medicine having minimum or no bad effects.

The smallest part of living organism is the cell. Number of reactions takes place in the cell simultaneously e.g. ion exchange, generation of energy, replication of DNA or RNA and many more with extreme accuracy and specificity. There is an auto reaction control and rate accelerations are observed. The base lies in the soft and reversible interactions, e.g. hydrogen bonding exists between two strands of DNA double helix which is a non-covalent interaction. The 2013 Nobel Prize in medicine is awarded for the work on machinery regulating vesicle traffic, a major transport system in our cell. The prize is shared by James E. Rothman, Randy W. Schekman and Thomas C. Sudhof. They have explained the precise control system for the transport and delivery of cellular cargo, which are mainly based on non-covalent interactions. Enzymes are made from proteins which self assemble for a specific task. Emil Fischer in 1894 gave lock and key model to explain enzyme action and provided the basis of molecular recognition. In short non-covalent interactions are basis of biochemical processes.

![Comparison of enzyme specificity with lock and key](image)
Natural receptors are quite flexible and can adopt a necessary change in their structure as well as function depending on the environmental need. Because of these fascinating characteristics 21st century research has more focused on materials involving soft chemistry.

The research in this area started long before with the observation of Paul Ehrlich that ‘molecules do not act if they do not bind’. The concept of receptor and substrate binding strengthened with lock and key model and resulted in enormous number of diverse molecules when Alfred Werner gave concept of coordination. In 1987 Jean Marie-Lehn, Donald J. Cram and Charles J. Pedersen got the Nobel Prize for their work in the area of soft interaction named as Supramolecular Chemistry: ‘Chemistry beyond the molecule bearing on the organized entities of higher complexity that result from association of two or more chemical species held together by non-covalent interactions.’

Supramolecular chemistry is also called the chemistry of molecular assemblies and of the intermolecular bond, chemistry of non-covalent bond and non-molecular chemistry.

Concept of covalent and supramolecular bonding

Fig. 1.2
Supramolecular entities are thermodynamically less stable, kinetically more labile and dynamically more flexible than covalently bonded molecules. The supramolecular chemistry deals with binding of a substrate to receptors where a receptor is generally a bigger component which is capable of binding smaller molecules. The substrate is usually a smaller molecule whose binding is being sought.

**Components of Supramolecular Chemistry**

**Fig. 1.3**

Molecular recognition, transformation and translocation represent the basic functions of supramolecular species. Recognition is the preferential binding of a given substrate by the receptor molecule. Generally, recognition and subsequent binding of a substrate by a receptor are reflected in terms of a change in the physical properties of the receptor molecule. When receptor bears a reactive functionality, it makes chemical changes to the bound substrate and transforms it to the more stable or more suitable substrate and thus
acts as a supramolecular catalyst. Sherman’s careplexes and hemicareplexes are appropriate examples of receptors having transformation capacities.⁶ Benzyn was trapped in the cavity of a careplex and reacted with the aromatic ring of the careplex receptor. A lipophilic membrane soluble receptor may act as a carrier effecting the translocation of the bound substrate. Supramolecular chemistry comprises of all types of species held by non-covalent interaction including oligomolecular species like macrocycles or cage compounds as well as polymolecular species.

Supramolecular chemistry is divided into two broad classes.⁴

I Supermolecules: “Supermolecules are well defined discrete oligomolecular species that result from the intermolecular association of a few components based on the principles of molecular recognition.”

II Supramolecular assemblies: “Supramolecular assemblies are polymolecular entities that result from the spontaneous association of a large undefined number of components into specific phase having more or less well-defined microscopic organization and macroscopic characteristics depending on its nature.”

Supramolecular chemistry is also known as host-guest chemistry where receptor is called host molecule which possesses convergent binding sites like Lewis basic donor sites and guest is the molecule which possesses divergent i.e. counter interacting binding sites like Lewis acidic acceptor site. Commonly the host is a large molecule or aggregate such as macrocyclic compound possessing a central hole or cavity of a definite size. Commonly the guest may be cation, anion or neutral molecule such as a hormone, drug, pheromone or neurotransmitter. Pederson’s crown ether complexation with metal ion is the first example of such man maid host-guest system. Sherman’s careplex and hemicareplex formation is another well studied host-guest system, where guest itself act as template to organize the assembly of the host molecule and gets trapped at the end of the synthesis. Hemicareplexes are more popular due to their application in stabilization of short lived reactive intermediates and to achieve direct reactions inside the cavity of the host molecule.⁶
1.2 Types of Supramolecular interactions\textsuperscript{4}

The forces that drive the non-covalent binding between host and guest can be classified into a) ion-ion interaction, b) ion-dipole interaction, c) dipole-dipole interaction, d) hydrogen bonding, e) cation-π interactions, f) π-π stacking, g) Van der Waals forces, h) hydrophobic effects and i) combinations of these interactions.

\begin{itemize}
  \item [\textbf{Ion-Ion interaction}]
  Bond energy of ion ion interaction between host and guest molecules ranges from 100 kJ/mole to 350 kJ/mole. The common example of such reaction is NaCl. Where Na\textsuperscript{+} ion is capable of organizing six Cl\textsuperscript{-} donor atoms around itself and Cl\textsuperscript{-} also attracts six Na\textsuperscript{+} ions around itself. Another example of ion-ion interaction is between supramolecular tris-(diazabicyclooctane) host which carries 3+ charge and an anion such as Fe(CN)\textsubscript{6}\textsuperscript{3-}.

  \begin{center}
  \textbf{Ion-ion interaction between tris-(diazabicyclooctane) host and Fe(CN)\textsubscript{6}\textsuperscript{3-} guest}
  \end{center}

  \begin{center}
  Fig. 1.4
  \end{center}

\end{itemize}

\begin{itemize}
  \item [\textbf{Ion-dipole interactions}]
  Bond energy of ion-dipole interaction between host and guest molecules ranges from 50 kJ/mole to 200 kJ/mole. The well known example is the binding of K\textsuperscript{+} with [18]-crown-6 (Fig. 1.5) or Zn\textsuperscript{2+} bound in octaaza-cryptand cavity (Fig. 1.6).
\end{itemize}
Dipole-dipole interactions

Dipole-dipole interactions are relatively weak interactions having bond energies of 5-50 kJ/mole. This interaction exists between the molecules having permanent dipoles as carbonyl functionalities. (Fig. 1.7)

Hydrogen bonding

“The hydrogen bond is an attractive interaction between a hydrogen atom from a molecule or a molecular fragment X–H in which X is more electronegative than H, and an atom or a group of atoms in the same or a different molecule, in which there is evidence of bond formation.”
It is a special type of dipole-dipole interaction in which a hydrogen atom attached to an electronegative atom is attracted to a neighbouring dipole on an adjacent molecule or functional group. ‘It is known as master key interaction in supramolecular chemistry.’

Hydrogen bonds are present in many supramolecular host-guest systems such as DNA double helix\(^2\) (Fig. 1.8) or in enzyme action on different substrates in the body.

![Hydrogen bonding between DNA double helix](image)

**Hydrogen bonding between DNA double helix**

**Fig. 1.8**

Many synthetic receptors undergo hydrogen bonding to make a stable supramolecular assemblies e.g. binding of barbiturate molecule with a macrocyclic host by six hydrogen bonds.\(^8\)

![Barbiturate ⊂ Aza Corand](image)

**Barbiturate ⊂ Aza Corand**

**Fig. 1.9**
When hydrogen atoms are attached to carbon rather than electronegative atoms and are attracted by neighboring dipole form **weaker hydrogen bonds**. This is possible with acidic hydrogens, where carbon containing hydrogen will be attached to electron withdrawing group. e.g. nitro methane encapsulated within the cavity of pyridine based crown ether. An example of such kind of weaker attraction is proposed when nitromethane gets encapsulated within the cavity of the pyridine based crown ether. (Fig. 1.10)

![Nitromethane and Pyridine linked Crown ether](image)

**Nitromethane**→**Pyridine linked Crown ether**

**Fig. 1.10**

- **Cation-π interactions**

In cation-π interaction, bond energy varies from 5 to 80 kJ/mole. The well known example is of sandwich molecule ferrocene, were Fe^{2+} ion is attracted by two aromatic π electron clouds. Pt^{2+} also forms such compounds due to cation-π interactions. When alkali or alkaline earth metal ions gets encapsulated in the aromatic cavity of supramolecular hosts or form complexes with carbon-carbon double bonds, the interaction involved is much more noncovalent or weak and becomes important in the supramolecular chemistry.

- **π-π staking**: Bond energy of this interaction varies from 0 to 50 kJ/mole. When one aromatic ring is electron rich and other is electron poor, there exists a weak electrostatic interaction, known as π-π staking. It is of two general types: a) edge to face b) face to face (Fig. 1.11)
Hydrophobic effects

Hydrophobic effects are considered as exclusion from polar solvents such as water, alcohol etc. When few oil droplets are added to water they are repelled by water molecules. There is a strong attraction already present between the surrounding water molecules which squeeze out the oil droplets and agglomeration of such droplets takes place which is known as hydrophobic effect. In supramolecular chemistry the host such as cyclodextrins, cyclophanes etc are soluble in polar solvents but their interior is hydrophobic. When organic guest molecules are added to their aqueous solutions, water molecules from the cavity are thrown out and organic molecules are stabilized in hydrophobic cavity of such host molecules. (Fig. 1.12)

![Diagram of PABA and α-cyclodextrin with edge to face π-π staking and face to face π-π staking](image)

**PABA** = p-aminobenzoic acid \( \equiv \) α-cyclodextrin

(Fig. 1.12)
Van der Waals forces:

The Van der Waals interactions are very weak having bond energy less than 5kJ/mole.

“Weak electrostatic attractions occur between dipoles and instantaneous dipoles created by polarization of the electron cloud of one atom/molecule by the nucleus of the adjacent atom/molecule are known as van der Waals forces.” They are present in noble gases. p-tert-Butyl-calix[4]arene encapsulate toluene due to this interactions. (Fig. 1.13) Interaction mainly depends on distance between the two dipoles and attraction decreases with increase in the distance rapidly.

![Toluene = p-tert-Butyl-calix[4]arene](image)

1.3 Classification of supramolecular interactions⁴:

The supramolecular interactions can be classified as summative or multiplicative interactions. In summative interaction the total stabilization energy is arithmetic sum of the individual stabilizations resulting from individual binding sites. While in case of multiplicative interactions the total stabilization energy of a supramolecular system is synergistically greater than the sum of individual stabilizations. Multiplicative interactions are found in most of the supramolecular species and comprise of chelate effect and macrocyclic effect.

Chelate effect: Metal complexes of bidentate or polydentate ligands are more stable than corresponding monodentate ligands. e.g Ethylenediamine complexes. (Fig. 1.14)
When ethylenediamine is added to the copper hexamine complex this chelating bidentate ligand replaces six ammonia molecules to form a chelate complex of copper and ethylene diamine, thus increases the entropy of overall reaction as total entities on reactant side are four while on product side the total entities are seven. This makes $\Delta S$ positive and $\Delta G$ negative which ensures the feasibility of the reaction.

**Macrocyclic effect**

Macrocyclic ligands are more stable than the corresponding acyclic analogous. This is due to presence of number of binding sites of the ligands at proper positions to chelate their guests. Such supramolecular entities are further stabilized by the macrocyclic effect. During the synthesis of macrocycle, the enthalpic penalty associated with bringing donor atom lone pairs in juxta positions with respect to one another in order to bind the guest molecule has been already paid in advance. The macrocyclic hosts such as corands are up to a factor of $10^4$ times more stable than their acyclic analogous podands with similar binding sites. In case of acyclic molecules more number of host-solvent bonds are required to be broken to achieve a stable host-guest complex compared to the cyclic host, as cyclic nature of the host makes minimum interaction with the bulk solvent. This provides the enthalpic stabilization. Macro cyclic host losses less degree of freedom when gets complexed which is entropically favoured process.
1.4 Classification of Supramolecular Host-guest Compounds

There are two types of supramolecular host-guest complexes. a) clathrates b) cavitates

**Clathrates:** “Clathrate is a kind of inclusion compound in which two or more components are associated without ordinary chemical union but through complete enclosure of one set of molecule in a suitable structure formed by another.” Here host is known as clathrand which possess extramolecular cavity. These species are stable in crystalline or solid state only. Cyclotriveratrylene host (CTV) forms clathrates with acetone involving van der Waals interactions or gets stabilized due to crystal packing.

**Cavitates:** “Cavitate is a kind of inclusion compound in which one or more guest molecules are completely encapsulated within the cavity of host molecule.” Here host is known as cavitand which possesses intramolecular cavities. These are stable in solid as well as solution state. Macroyclic hosts possessing intramolecular cavity are capable of binding the guest molecule(s) by non-covalent interactions. When host is dissolved in a solvent, the solvent molecules get bound in the cavity of the host. If a suitable guest is added, the bonds between solvent and the host cavity gets broken and new bonds between host and guest molecules are formed. This happens only when the host has preferential selectivity to the added guest rather than a solvent molecule. Further this process should be entropically favoured.
Cavitates can be further classified on basis of the structures of host molecules. Fig. 1.16 represents various types of cavitates and the interaction presents between the host-guest molecules.
Classification of Cavitates

Fig. 1.16a
Catenane and Rotaxane

Charge Transfer

Carceplexes and Hemicarceplexes

Van der Waals interactions

Classification of Cavitates

Fig. 1.16b
1.5 Molecular recognition: Binding and selection of substrate(s) by a given receptor molecule is called molecular recognition. Mere binding is not recognition, but selective binding with a particular purpose is recognition. Recognition is due to geometrical and interactional complementarity between two or more associating entities.

Recognition depends on

- **Steric factors**: The best fit between receptor ‘ρ’ and substrate ‘σ’ is a steric complementarity i.e. presence of correct concave and convex domain at an appropriate positions on receptor and substrate

- **Interactional complementarity**: It means presence of complementary functionalities on ‘ρ’ and ‘σ’. Generally receptors possess convergent binding sites which include presence of dipoles, hydrogen bond donor sites, negative or positive charges etc. which need to be matching to the binding functionalities on substrate molecules like positive or negative charges, dipoles, hydrogen bond acceptors in correct position on ‘ρ’ and ‘σ’.

- **Large surface overlaps**: A maximum overlap is required between receptor and substrate to have multiple interaction sites.

In order to achieve efficient recognition, both high stability and selectivity are required. The natural receptors are flexible and can undergo conformational changes to bind a required substrate. These complexes are often less stable as the energy is utilized in making the conformational changes. Rigid receptors form more stable host-guest complexes but they lack in flexibility. Biological processes like exchange, regulation, transformations and transportations require built-in flexibility where a receptor needs to adapt and respond to the changes.
1.6 Chiral recognition

Biochemical systems involve enantiospecific receptor-substrate binding. Cram’s group in 1976 reported a new class of macrocyclic chiral compound named chiral crown ether in enantiomerically pure form (R,R). It does enantioselective recognition of chiral ammonium salts. It is useful in resolution of many drug molecules and thus has potential applications in pharmaceutical industry.

![Chiral crown ether for enantiospecific binding of amino acids](image)

**Fig. 1.17**

The (R,R) chiral crown ether displays enantioselective binding with one of the enantiomers of chiral amino acids. Generally it is observed that this (R,R) host binds with D- amino acids and their esters much more strongly than the L-guest. The host-guest complexes can be extracted by chloroform from water. The D isomer selectively binds to the host having better fit and gets preferentially extracted.

Stoddart and co-workers in synthesized a number of chiral corands derived from mannitol and tartaric acid derivatives which were employed for resolution of various amino acids.

Chiral recognition of amines and amino alcohols was possible by using azophenolic crown ethers (Fig. 1.18) where cis-1-phenylcyclohexane-1,2-diol was a chiral subunit which was responsible for chiral recognition while 2,4-dinitrophenyl-azophenol was introduced to generate a specific response in UV-Vis spectrum.
Resolution of racemic mixture of various aminoacids including methionine, phenylglycine, isolucine etc was done using [18]-crown-6-tetracarboxylicacid host. The chiral discrimination was studied with the help of their single crystal X-ray structures.

Chiral recognition with Podands and lariat ethers

Chiral recognition of amino acids with aromatic side chain (e.g. tryptophan and phenylalanine) can also be achieved with the help of chiral podands and lariat ethers.

Chiral, light responsive azophenolic crown ether

Fig. 1.18

Recognition of L-tryptophan in (S,S) host
Fig. 1.19

Recognition of L-phenylalanine in (S,S) host
Fig. 1.20
1.7 Methods of synthesis of supramolecular structures:

1.7.1 Template assisted synthesis

Template: “The chemical specie which organizes an assembly of atoms, with respect to one or more geometric loci, in order to achieve a particular linking of the atoms is known as template”

Ether formation reaction between –OH and –Cl functional groups is a base catalyzed reaction. When triethylamine is used as a base in the reaction of dihalide with diol, an intermolecular reaction resulted in an oligomeric species, which may further react with diols and dihalides to form a polymer. When specific alkali metal carbonate is used, it orients the reactants diol and dihalide around itself in an appropriate geometry to achieve
macrocyclization. Here alkali metal ion like Na\(^+\) is called template or more specifically a kinetic template and the specific macrocyclization in presence of cation or anion or a neutral molecule is known as template effect.

Template may be an organic molecule, a metal cation or an anion. Cs\(^+\) is very effective as a template for the formation of cyclic products in good yields. The phenomenon is called ‘caesium effect’. Although template assisted synthesis gives products in good yields, it is often observed that the template gets trapped into the cavity of the synthesized host. Open hosts like crown ethers are exceptions. It is also seen that some so-called template assisted synthesis proceeds similarly even in absence of a template e.g. synthesis of porphyrins and macrocycle formed by condensation between acetone and ethylenediamine.\(^{15}\)

To obtain a template free host, high dilution technique is employed in absence of any specific ion or molecule to act as a template.

**1.7.2 High Dilution Synthesis\(^{4}\)**

![Diagram of template free high dilution synthesis of aza-crown ether](image)

Template free high dilution synthesis of aza-crown ether

**Fig. 1.22**
When an appropriate template is not available for macrocyclic host formation or for metal free synthesis is to be done, reaction is carried out in excess of solvent, which is known as high dilution synthesis. Here the reactants are dissolved in an excess of solvent and simultaneously added drop wise at a very slow rate from addition funnels to a round bottom flask containing an excess of solvent. This ensures a very low concentration of reactants as well as intermediate products at a time and reduces chance of polymerization favouring intramolecular cyclization to yield the desired macrocycle.

If rate of cyclization of reactant A-B, \( r_c = k_c [A-B] \) is compared with rate of polymerization \( r_p = k_p[A-B]^2 \)

\[
\frac{r_c}{r_p} = \frac{k_c [A-B]}{k_p[A-B]^2} = k_c / k_p [A-B]
\]

The lesser is the concentration of A-B, more is the rate of cyclization.

**Template assisted Synthesis**

**High dilution Synthesis**

Cryptand formation under High dilution as well as template assisted synthesis\(^{15,16}\)

Fig. 1.23
As depicted in Fig. 1.23 synthesis of TREN capped cryptand can be achieved using either lanthanide cations or under high dilution condition. In template assisted synthesis the yields are higher while in high dilution synthesis though the yields are lower, metal free cryptand is achieved.

1.8 Binding constant

Binding constant is a specific numerical value assigned to the host guest entities reflects stability of the host guest system. Binding constant is also known as stability constant or protonation constant. Binding constant is quantification of supramolecular host-guest complexation. It is a result of mathematical operations on experimental observations. Normally the experimental observations are based on titration methods using different spectroscopic techniques. For titration most often the guest is gradually added to the solution of host molecule. Supramolecular interactions between the two components if results in a change in the spectroscopic behavior of the host, the binding between the two can be detected and the technique can be employed for determination of binding constant. Alternatively potentiometric titration can also be used for determination of binding constant especially when metal ions are involved.

➢ Determination of binding constants.

The most common approach to find out binding constant is the supramolecular titration method. Here one component (most often a guest) is gradually added to the solution of the other (Host) while monitoring a change in physical property such as chemical resonance in $^1$H NMR, $^{13}$C NMR or absorption band in UV that is sensitive to the supramolecular interactions of interest. The resulting information is further processed by chemical models and their mathematical equations to calculate association constant $K_a$ also known as binding constant, Energetic and stoichiometry. Some of the most frequently used techniques for determining binding constant are outlined as follows.
1.8.1 Determination of Binding constant: pH measurement

Ligands are weak bronsted bases. In aqueous solutions the ligand can form metal-ligand complex or it can be protonated. There is a competition between metal ion and proton which can be monitored by PH-measurements or pH titration. This method was developed by Bjerrum. Method is very precise and can be applied to various metal ions and ligands.

The most common procedure for binding constant determination is pH titration. A particular concentration of ligand solution is prepared to which stoichiometric amount of metal ion and acid is added under nitrogen atmosphere to prevent aerial oxidation or reaction with carbon dioxide. A standard base is added drop wise to a magnetically stirred solution and pH is measured.

1.8.2 Determination of Binding constant: NMR measurement

The most informative technique which is applicable to a wide range of organic ligands is protone NMR measurements at different host-guest concentration. Relative shifts in NMR monitored titration can give qualitative information such as changes in symmetry of the molecule, stoichiometry of guest to the host molecule and quantitative information such as binding constant for the guest molecule.

In case of host-guest complex having (1:1) stoichiometry, chemical shift ($\delta$) of the signal of interest is assumed to be the weight average of the free host and the bound host complex. This is true for the equilibria that are fast on NMR time scale. For a slow exchange of complexed and uncomplexed host, the binding constant may be evaluated by simple integration of NMR signals.

Experiment is carried out in deuterated solvents where known quantity of host is taken and small aliquots of the guest are added and spectrum is monitored as a function of guest concentration or host-guest ratio. Titration curve is monitored by computer programme such as EQNMR.
1.8.3 Determination of Binding constant: By UV-Visible measurement

Spectrophotometric method is convenient as compared to potentiometric method and less expensive as compared to NMR titration method. It can also be used for non-basic ligands such as halides. It is necessary to determine stoichiometry of the complex before calculation of binding constant.

Absorbance is measured at a wavelength where complex absorbs strongly but metal as well as ligand’s absorbance is minimum or ideally they do not absorb at all.

In mole-ratio method a series of solutions is prepared where concentration of any one of the reactant is held constant and other’s is varied.

Absorbance is plotted against concentration of the component whose concentration is varied. If only one complex of high stability is formed, the graph consists of two linear intersecting parts. Intersection point refers to the stoichiometry of the complex.

**Fig. 1.24**
Calculation of binding constant assuming 1:1 stoichiometry

\[ K_a = \frac{[HG]}{[H][G]} \]  
Equation-1

\[ [G] = \frac{1}{2} \left( \frac{1}{G_0} - H_0 - \frac{1}{K_a} \right) - \sqrt{\left( \frac{G_0 - H_0 - \frac{1}{K_a}}{2 + \frac{4G_0}{K_a}} \right)^2} \]  
Equation -2

\[ [HG] = \frac{1}{2} \left( \frac{1}{G_0} + H_0 + \frac{1}{K_a} \right) - \sqrt{\left( \frac{G_0 + H_0 + \frac{1}{K_a}}{2 + 4[G_0][H_0]} \right)^2} \]  
Equation-3

\[ \Delta A_{obs} = \varepsilon_{HG} ([HG]) \]  
Equation-4

From eq-3 and eq-4 binding constant ‘\( K_a \)’ can be calculated.

1.8.4 Determination of Binding constant: Fluorescence measurement

Fluorescence is one of the most popular, relatively cheap and most sensitive techniques. Due to its high sensitivity it can work with submicromolar or even nanomolar solutions.

Ideally fluorescence titrations are to be carried out with such a concentration where absorbance is less than 0.05 at a wavelength of excitation.

The best situation under which fluorescence experiments can be carried out is one where host and guest both are fluorescent inactive but complex is fluorescent active. But it may not be the case always still if either of the host or guest is fluorescent active and a complexation quenches the fluorescence, the binding constant can be measured. It can be static quenching or dynamic quenching.

Following equation is used for determination of binding constant.

\[ \Delta F_{obs} = K_{AHG} ([HG]) \]
1.9 Applications of supramolecular entities:

➢ Supramolecular hosts as Sensors:

“A sensor is a converter that measures a physical quantity and converts it into a signal which can be read by an observer or by an instrument.” 22

Supramolecular chemosensors are based on chemical changes which reflect the encapsulation of a particular guest into a well designed host molecule which acts as a sensor. Many techniques are employed to analyze the target entity including mass spectrometry and atomic absorption spectroscopy, however these methods are not popular as they are more time consuming or require sophisticated instrumentation. UV and fluorescence spectroscopy have been powerful tools due to their simplicity, high detection limit and application to bio imaging. These are fast, non-destructive, highly sensitive and suitable for high-throughput screening applications. Quantification is possible here with quite good efficiency. 23

A number of supramolecular sensors have been developed to sense cations, anions as well as organic molecules such as carboxylic acids, esters etc. Metal cations especially transition metal ions are one of the widely used substrates because of their biological concern and presence in the environment. 24 Organic macrocyclic host molecules serve as efficient receptors for such metal ions and these ions could get bound in their cavity. If binding of these metal ions to the receptors result in a trivial colour change or affect the fluorescence properties by change in fluorescence intensities, can act as a colorimetric chemosensors or fluorescence chemosensors respectively.
1.9.1 Design principle for fluorescence chemosensors for detection of metal ions\textsuperscript{25}.

Depending on their mechanism of action they are classified in five different ways.

1. **Fluorescent ligands:** They possess receptor unit capable of emitting fluorescence upon binding of the metal ion.

\[
\text{Fluorescent ligand as metal ion sensor}
\]

Fig. 1.25

Here an aromatic moiety which is a good fluorophore, is attached to the macrocyclic host or substituted with particular functional groups like amine, amide, imine, phenolic or carboxylic group. These functional groups help in binding the substrate which is generally a metal ion. Binding and selectivity depends strongly on number, type and geometrical arrangement of functional groups attached to the aromatic moiety. It is observed that paramagnetic ions are more strongly bound than the diamagnetic ions.

2. **Intrinsic fluorescent probes:** In case of intrinsic fluorescence probes, binding site and chromophore are in direct electronic conjugation.

\[
\text{Intrinsic fluorescent probes as metal ion sensors}
\]

Fig. 1.26
This is achieved by connecting electron donating and electron accepting fragments by butadienyl, stilbenyl or phenyl groups. It can also be framed as donor-acceptor-donor type of system.

When donor-π-acceptor type of probe is excited at a particular wave length it goes to Frank-Condon excited state which undergoes charge transfer from donor to acceptor subunit of the probe. In charge transfer state as charge density separation increases, the dipole moment increases and emission takes place at higher wavelength. Now when metal binding takes place, the receptor uses its electrons in binding with the metal ion and hence its delta negativity and electron donating capacity decreases. This leads to a blue shift in absorption spectrum of the complex as compared to that of the unbound probe. When such complex is to be excited to Frank Condon state, higher energy is required.

After excitation, charge transfer process still proceeds which make the donor like a radical cation. As this donor site is bound to a metal ion an electrostatic repulsion takes place which weakens the coordinative bond, resulting in a very little blue shift observed in emission spectrum as compared to the unbound probe.
When there is a large energy gap between Frank Condon excited state and charge transfer state, the rapid and quantitative population of the weakly emissive charge transfer state is achieved. Here energy level position of charge transfer state is still lower than the other available non-emissive states, and thus the conversion from Frank Condon excited state to charge transfer state still dominates. This is reflected in the emission spectrum where very little shift is observed. As a result, all complexes of such donor acceptor probes are highly fluorescent than the free probes.

If the donor-acceptor probe is designed in such a way that different energy states are very close to one another, than complexation promotes fluorescence quenching due to reduction in charge transfer characteristic of probe after complexation. Here energy level of charge transfer state is raised, which results in conversion from Frank Condon excited state to other lower energy non radiative states. This results in fluorescence quenching.

Examples of such probes are shown in Fig. 1.28:

Azacorwn based intrinsic fluorescent probes

Fig. 1.28
3. Composite fluorophore-spacer-receptor systems:

Here ligand and fluorophore are electronically decoupled by a spacer.

![Diagram of composite fluorophore-spacer-receptor system as metal ion sensor]

**Composite fluorophore-spacer-receptor system as metal ion sensor**

*Fig. 1.29*

In most of the fluorophore-spacer-receptor systems highest occupied molecular orbital (HOMO) of receptor has energy in-between that of HOMO and LUMO of excited fluorophore. When excitation of electron from HOMO of such a probe takes place to its LUMO, simultaneous electron transfer takes place from free receptor to HOMO of the excited fluorophore. This results in fluorescence quenching. When complexation is achieved, loan pair of the receptor is bound with the cation, which prevents photo induced electron transfer to HOMO of the excited fluorophore. This allows electron from LUMO of the excited fluorophore to come to its HOMO followed by fluorescence enhancement (Fig. 1.30). Such probes are generally used as switch-ON type fluorescence sensors.
Examples of fluorophor-spacer-receptor systems are shown in Fig. 1.3.\textsuperscript{1,2,8,29}

**Photoinduced electron transfer**  
**Inhibition of photoinduced electron transfer (PET) after complexation**

**Fig. 1.30**

Examples of fluorophor-spacer-receptor systems are shown in Fig. 1.31.\textsuperscript{28,29}

**Fluorophore-spacer-receptor type of ligands**

**Fig. 1.31**
4. Exciplex or excimer forming probes:

Here binding and signalling units can form an intramolecular exciplex or excimer and encapsulation of the metal ions causes strong conformational changes which result in increasing or decreasing ratio of excimer-to-monomer emission. (Fig. 1.32)

Design of excimer forming probes

**Fig. 1.32**

Exciplex or excimer forming probes are designed in such a way that their geometrical arrangement gets changed on cation binding which results in fluorescence enhancement or quenching.

When one or two fluorophores are attached to cyclic or acyclic receptor via short alkyl spacers, they prevent PET in excited state after complexation as well as adopt particular geometrical conformation which results in excited state photo physical processes like intramolecular excimer or exciplex formation. Due to this phenomenon a new red shifted band in emission spectrum is observed due to charge transfer interactions in polar solvents. These are capable of switch-on the fluorescence after complexation.

Reverse is possible when geometry of the probe is such that in unbound state they form excimer or exciplex, but cation binding changes the geometry and prevents excimer or exciplex emission. Examples of exciplex or excimer forming probes are shown in Figure 1.33.
Fluorescent receptors with anthracenyl and pyrene fluorophore
Fig. 1.33

5. Chemodosimeter:
Weakly fluorescent probe reacts with redox active metal ions which greatly enhances its fluorescent activity. (Fig. 1.34)
Here a receptor which itself is non-fluorescent reacts with a metal ion to yield a fluorescent product and acts as switch-ON type fluorescent chemosensor. Selectivity amongst different metal ion depends on specificity of a reaction employed. The extent of fluorescence enhancement depends on reaction yield. Supramolecular hosts not only detect ions but are also used as molecular sensors.

1.9.2 Molecular Sensors
Explosives like trinitrotoluene (TNT) do need to be detected at ppm or ppb levels due to security reasons as well as due to environmental concerns. The macrocyclic host (Fig. 1.35) is found to bind 2,4-DNT and TNT molecules in its cavity. The films of different thickness were made by surface casting method based on the micrometric nanofibres of the macrocycle. These films show intense fluorescence which is immediately quenched on exposure to saturated TNT or 2,4-DNT vapours. The fluorescence can be slowly recovered on arial exposure or it can be immediately recovered on exposure to hydrazine vapours.
Another fluorescence sensor for TNB and TNT is pyrene derivative of amidocalix[4]arene-[15]crown-5. (Fig. 1.36) It gives two emission bands at 375 nm and 450 nm when excited at 343nm. TNB and TNT quench the fluorescence significantly while moderate quenching is observed with 2,6-DNT or 1,3-DNB with detection limit of 1ppb. It can also colorimetrically detect TNT and TNB. CHCl₃ solution of host gives intense bands at 320-360nm interval due to pyrene functionality. The host solution in chloroform appears colourless but turns yellow on addition of TNT and turns reddish orange on addition of TNB.  

Pyrene based hybrid fluorescent probe for TNB and TNT sensing

Fig. 1.36
1.9.3 Stabilization of reactive intermediates:

Supramolecular structures are also known for stabilization of reactive intermediates. The molecules like benzyne, cyclobutadiene etc. are extremely reactive and thus do not survive under normal reaction conditions. They can be formed and trapped within the interiors of macrocyclic cages where they are not affected by bulk reactants (Figure 1.37). This provides an opportunity for their complete characterization.

Stabilization of cyclobutadiene in carceplex cavity

Fig. 1.37
1.9.4 Nano reaction chamber:
Benzocyclobutenedione was encapsulated within the hemicarcerand by heating empty hemicarcerand in molten benzocyclobutenedione. Subsequent photolysis of the encapsulated guest converted it to benzocyclopropenone which on irradiation at 77K was converted it to benzyne.\(^3\) (Fig. 1.38) The hemicarcerand not only stabilizes benzyne in the cavity but also undergoes Diels-Alder reaction with it at ambient temperature. This reveals that the macrocyclic cavity not only acts as a container but also transforms the encapsulated guest.\(^3\)

![Generation of benzyne within the isolated interior of hemicarceplex](image)

*Fig. 1.38*
The other most versatile hosts known for supporting the reactions to undergo in its cavity without influence of the solution environment are cyclodextrins (CDs). Imidazole derivatized β-CD encapsulated and efficiently hydrolysed the phosphodiester 120 times faster as compared to the hydrolysis of the phosphodiester in the NaOH solution without β-CD. The reaction inside the cavity of β-CD also resulted in 99% selectivity for one of the two possible products (Fig. 1.39).

![Diagram](https://example.com/diagram.png)

**Highly selective hydrolysis of phosphodiester within the cavity of β-CD**

Fig. 1.39
Similar to the covalently bonded macrocyclic hosts, the host which are stabilized by non-covalent interactions like hydrogen bonding also provide an isolated interior to the encapsulated guests for their reactions. The hydrogen bonded resorcarene capsule encapsulated phenyl acetylene and phenyl azide molecules which underwent 1,3-dipolar cycloaddition reaction with efficient rate enhancement.\(^{39-40}\)

\[ \text{Capsule host} \]

\[ \text{Guests} \]

\[ \text{1,3-dipolar cycloaddition reaction within the resorcarene capsule} \]

**Fig. 1.40**
1.9 References:


2. See the Nobel Prize in Physiology or Medicine 2013, at http://www.nobelprize.org

3. The image has been taken from the website Creativecommons.org.


