



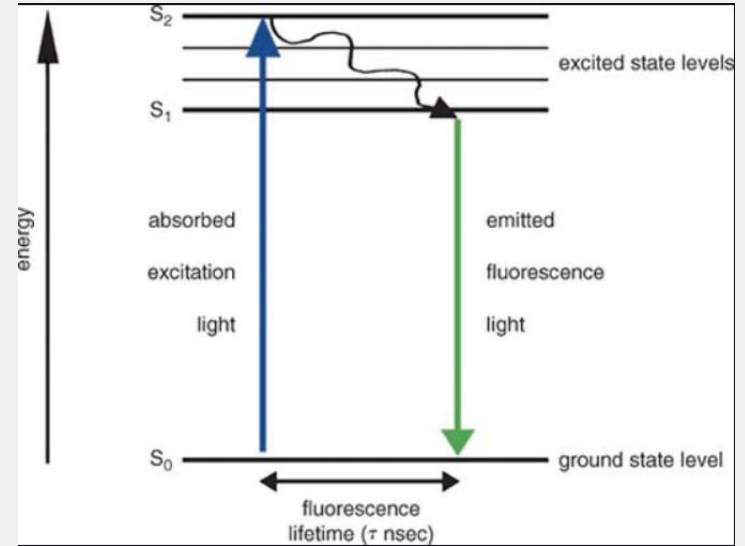
Chemical Fluorescence Probe

ALFA
CHEMISTRY

1

Fluorescence

Fluorescence is a phenomenon of photoluminescence. When some substances are excited by light, electricity, magnetism, etc., the electrons absorb energy from the ground state to the excited state, while the electrons in the excited state are unstable, and will return to ground state through transition. When the electron returns to the ground state from the lowest vibrational level of the first excited singlet state, the energy is released in the form of luminescence, and the emitted light is fluorescent.

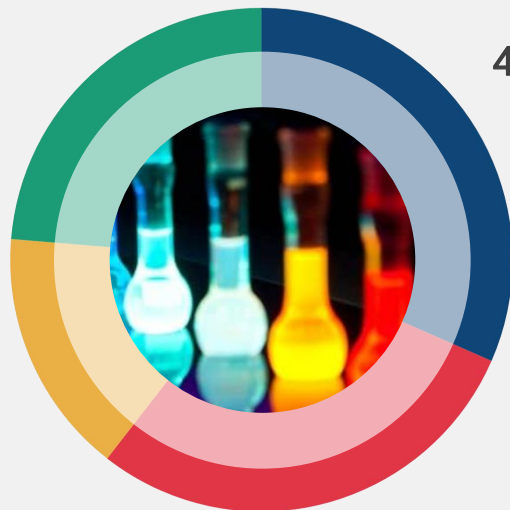


2

Characteristics of the Fluorescent Probe

1 Easy to synthesize and purify, high yield, safe and non-toxic.

2 Good stability and solubility



4 Higher fluorescence quantum yield, large molar extinction coefficient

3 Specifically combined with the label by physical or chemical action, the labeling conditions are mild, and the residue and by-products are easily removed

3

Composition of fluorescent molecular probes

**Receptor**

Receptor can selectively binds to a subject and causes a change in the chemical or biological microenvironment in which the probe is located.

**Fluorophore**

Fluorophores convert those chemical or biological microenvironmental change caused into human-perceivable or instrument-detectable signal like color change and fluorescence. .

**Spacer**

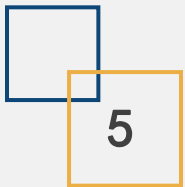
Spacer links the fluorophore and the recognition group to enable the identification information to be efficiently converted into a fluorescent signal (such as changes in fluorescence intensity, movement of the fluorescence spectrum)



4

Substances detected by fluorescent probes

| Proton (H⁺) | Free radicals | Gas signal molecules |
|---|--------------------------|--------------------------------|
| Heavy metal ions | Anions | Transition metal ions |
| Alkali and alkaline earth metal ions | Biomacromolecules | Small organic molecules |



5 Fluorescent probe



Ion channel fluorescent probes, acceptor fluorescent probes, mitochondrial labeled probes



6

Design mechanism of fluorescent probe



Photoinduced Electron
Transfer (PET)



Intramolecular charge
transfer (ICT)



Resonance Energy
Transfer (FRET)



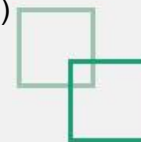
Excited Intramolecular
Proton Transfer
(ESIPT)



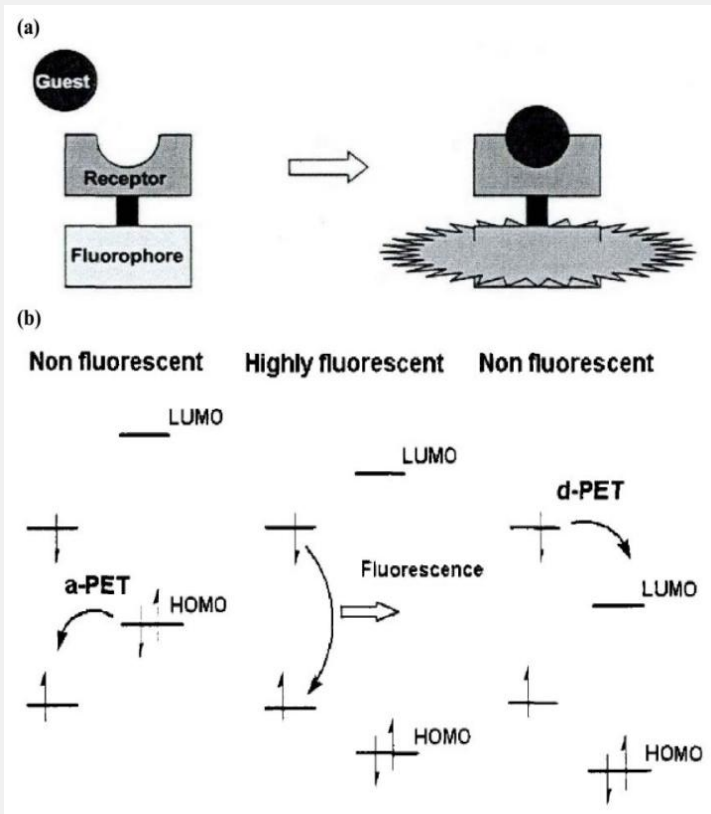
Excimer/Exciplex
Formation



Emerging
mechanisms include
Aggregation Induced
Emission (AIE)



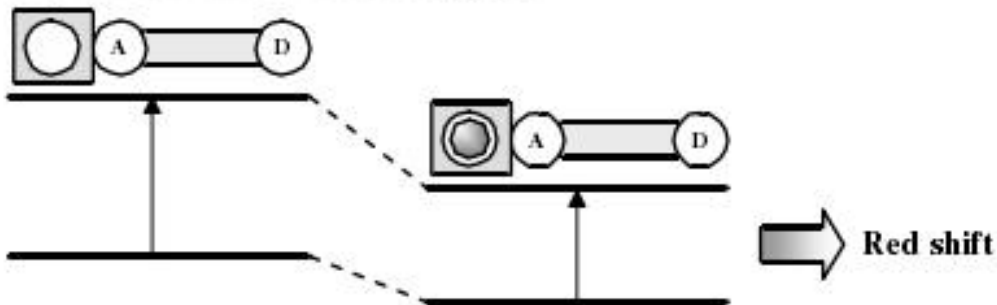
Photoinduced Electron Transfer (PET)



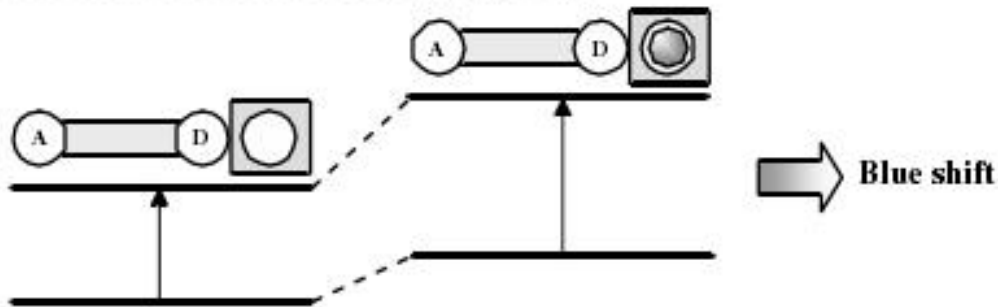
- Generally speaking, there are two types of photoinduced electron transfer processes. One is the transfer of electrons from the electron donor to the excited fluorescent group; the other is the transfer of electrons from the excited fluorescent group to the electron acceptor, and the oxidation of the excited fluorescent group results in fluorescence quenching. After binding to the guest, the process of PET is inhibited and the fluorescent group emits fluorescence.

8 Intramolecular Charge Transfer (ICT)

(a) interaction with the donor group

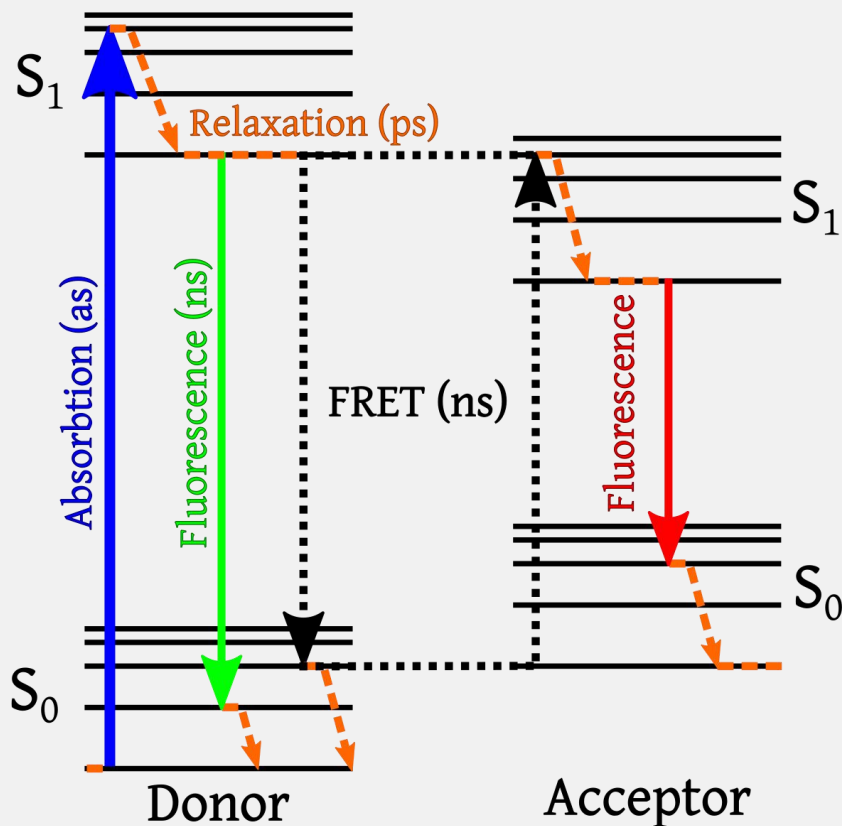


(b) interaction with the acceptor group



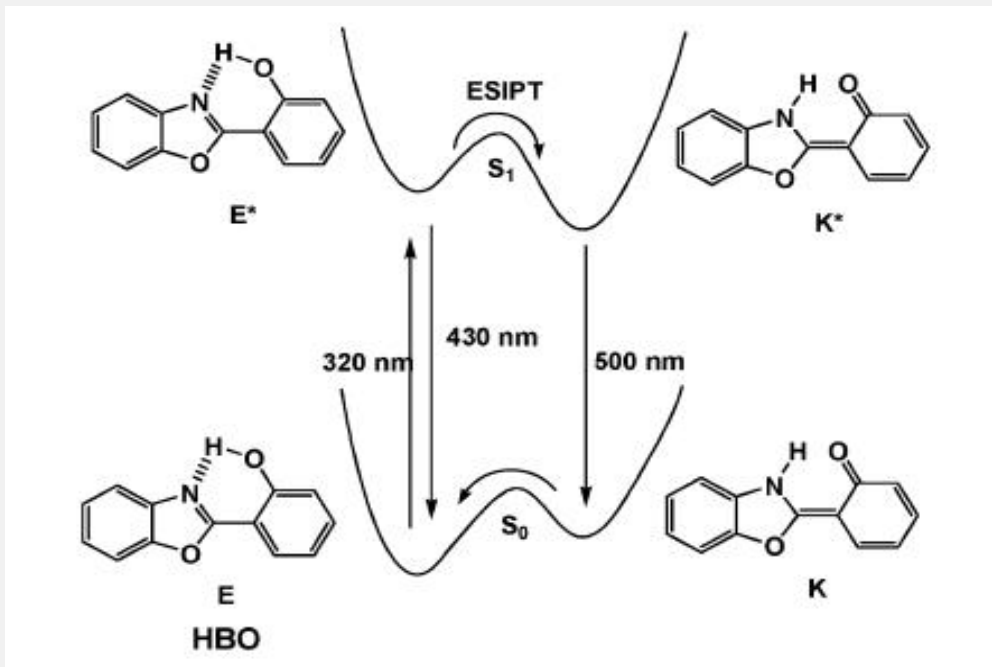
- Intramolecular charge transfer, also known as photoinduced Charge Transfer (PCT), is also an important principle of designing proportional fluorescent probes. The recognition groups of these fluorescent probes are directly connected with the fluorescent groups, and can also be understood as some atoms or groups that make up the fluorescent groups are directly involved in the recognition of the objects.

Fluorescence Resonance Energy Transfer (FRET)



- Fluorescence resonance energy transfer (FRET) is a kind of energy transfer. Energy transfer refers to the transfer of intramolecular energy from donor chromophore to receptor chromophore. ET can be divided into electron energy transfer (EET) and fluorescence resonance energy transfer (FRET) according to the interaction distance between energy donor and receptor.

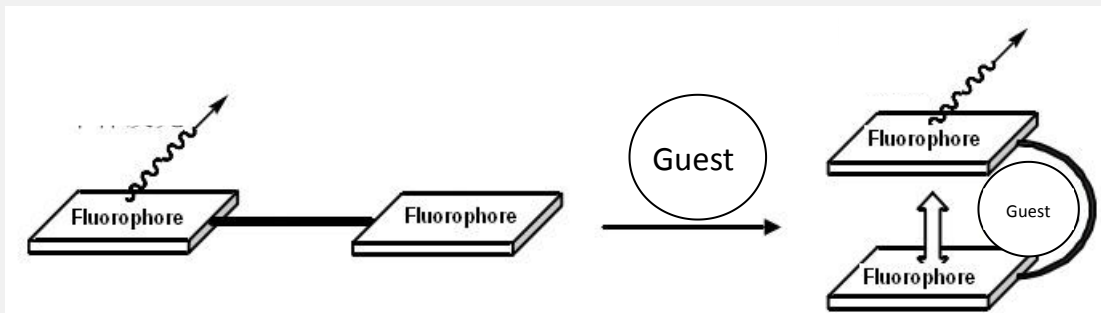
Excited Intramolecular Proton Transfer (ESIPT)



- ESIPT phenomena refer to the process in which the hydrogen nuclei of a group in a molecule are transferred from the ground state to the excited state through intramolecular hydrogen bonds to the adjacent nitrogen, sulfur, oxygen molecule to form corresponding tautomers.

11

Excimer/Exciplex Formation



- Excimer can be defined as an association formed by the interaction of an excited state fluorescent group with a ground state fluorescent group of the same structure. Similarly, if the fluorescent group in the excited state forms a complex with the fluorescent group with different structure in the ground state, it is called the Exciplex.

12

Aggregation-induced emission (AIE)



- Aggregation-induced emission (AIE) is an abnormal phenomenon that is observed with certain organic luminophores. Most organic compounds have planar structures and higher photoemission efficiencies in solution than in the solid state. When these luminophores aggregate or crystallize, which restricts those rotations, they can become very fluorescent or emissive, and the photoluminescence efficiency increases.

Thank You!

