Supramolecular Chemistry-I

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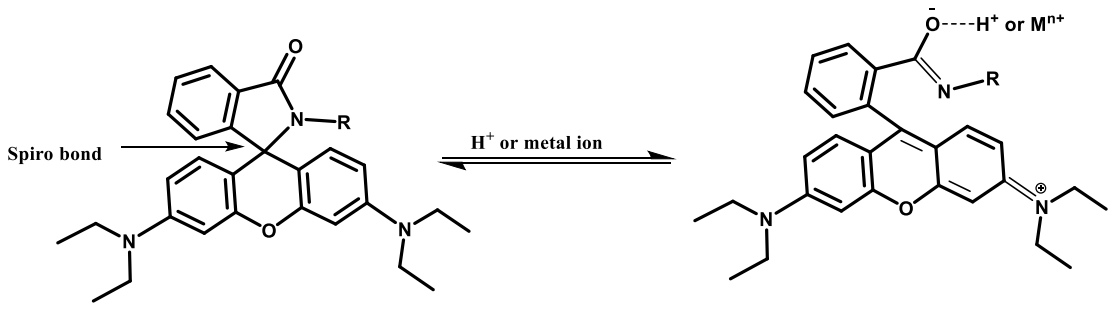
Week - 06

Lecture - 26

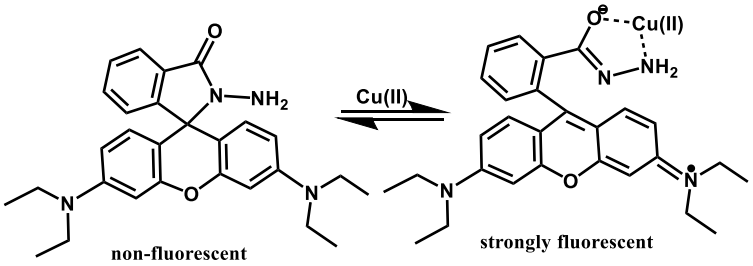
Hello, welcome back to the class. I have been showing you different mechanisms for fluorescence enhancement in presence of metal ions. So, here is another very important mechanism. Let me first draw it, then I will explain it. So, this is a particular type of compound with a spiro linkage. This particular compound called a spirolactum.



In fluorescence signaling, this type of compound is very important. How? When the spiro bond is intact, then the color will be white and it is non fluorescent. However, in presence of proton or a metal ion, this spiral bond breaks as shown. Metal ion is a Lewis acid, so it breaks the bond as well. So, what are the changes? This spiro bond breaks and the color changes to pink. Also, it is reversible. In the basic condition and absence of a metal ion, the spiro bond is formed again and it will become white in color.

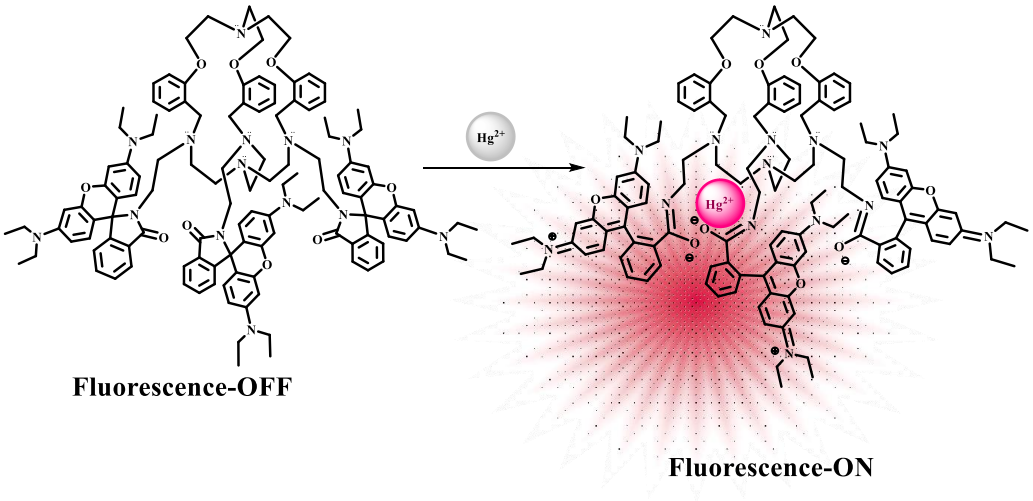
When the color is pink, we see an extended conjugation, and it is very highly fluorescent. As we can change the R group by many other groups, this receptor site can accept different metal ions and the compound can show high fluorescence. These compounds are photostable and they change from zero fluorescent to highly fluorescent. So, they are very very sensitive and can be easily detected even with naked eye because you give white color and then you give a metal ion its color turns to pink. So, by changing the R group we can get different receptors specific for different metal ions and it goes from white to pink color with high fluorescence, this systems can be excellent in fluorescence signaling of metal ions.

Let me show an example, now. When the **R** group is hydrazine derivative, it becomes Cu^{2+}



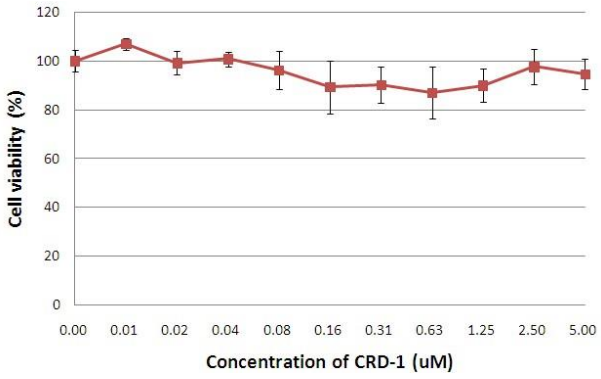
specific receptor. The non-fluorescent and white spirilactum breaks as shown in presence of Cu^{2+} ion. Color of the solution changes to pink and a strongly fluorescent solution is formed.

We have another cryptand based system where three spiro compounds are attached. It is quite pale in color and is not fluorescent. But specifically in presence of Hg^{2+} ion, the spiro bond is broken and it becomes dark purple and highly fluorescent.

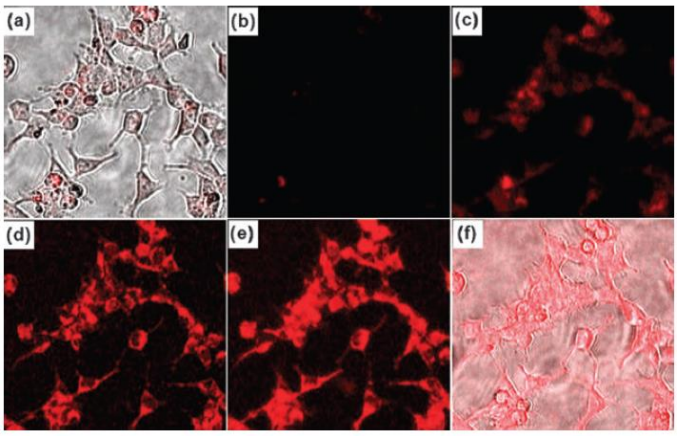


We called the system as CRD-1. Now 3 Hg^{2+} ions will bind because there are three spiro compounds attached. Mercury is not present in our body but we can get it from outside and it is one of the most poisonous elements known. So, during Hg^{2+} poisoning, how it spreads in the body and how much is the concentration are important to know before any

treatment. First thing will be to ascertain that the fluorophore signaling system is bio-compatible. For that an experiment as follows should be carried out.



Here, we find that even under very high concentration of CRD-1, the cells are not damaged and so we can use this compound. We inject a solution of CRD-1 to the poisoned victim and see the result with Confocal Microscope and get following type of images. From analysis of these images we can know how far poisoning has progressed and how much is the concentration of Hg^{2+} present.



Confocal fluorescence images of Hg^{2+} in **HEK 293** cells (Zeiss LSM 510 META confocal microscope X40 objective lens). (a) Bright-field transmission image and (b) fluorescence image of **HEK 293** cells incubated with the signaling system (1.0 μM). Further incubation with addition of $Hg(ClO_4)_2$ gives images (c)-(e).

So, we can calculate the detection limit. We find that it is 3 ppb (3 parts per billion). As per the EPA (environment protection agency) of the United States of America which most countries recognize the limitation of $Hg^{2=}$ in water is 6 ppb that means this particular system CRD-1 can be safely used to monitor the HG^{2+} content in drinking water.

Therefore, we first test the toxicity of the dye in the human cell using a technique called called MTT assay. And then we inject into human that will detect mercury present. What about other metal ions present in the human body? Well, this system is mercury specific and so other metal will not interfere. Once we remove then we again test and see mercury absent alright. Thank you for today.