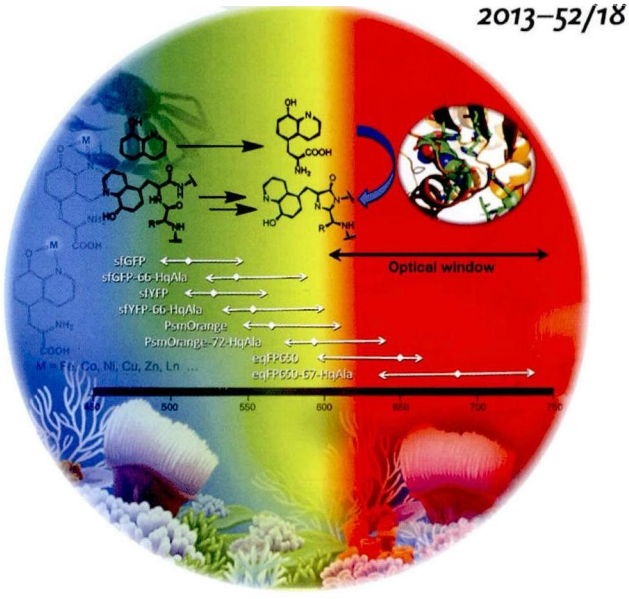
# Advances in the interface of photochemistry and bioinorganic chemistry

Genetic incorporation of metal-binding unnatural amino acids (UAA) is a powerful tool for protein sensor design, metalloenzyme engineering, and protein NMR. However, this method is currently underexploited by chemical biologists, due to the complex synthetic routes of UAA. Recently, Prof. Wang Jiangyun's group at the Institute of Biophysics, Chinese Academy of Sciences (CAS) published a paper entitled "Significant Expansion of the Fluorescent Protein Chromophore through the Genetic Incorporation of a Metal-chelating Unnatural Amino Acid" (*Angew Chem Int Ed* **2013**, 52: 4805—9). In this article, Wang and coworkers reported a one-step, high-yielding enzymatic route for the synthesis of a novel UAA-HqAla bearing the 8-hydroxyquinoline group. By substituting Tyr of the fluorophore in diverse fluorescent proteins (FP) with HqAla, Wang and coworkers showed for the first time that UAA incorporation results in significantly red-shifted excitation and emission spectra. Importantly, eqFP650-67-HqAla has the most far-red excitation peak (680 nm) of all GFP-like fluorescent proteins reported to date. This property can be extremely useful for deep tissue imaging. The crystal structure of superfolder GFP (sfGFP) bearing HqAla in its chromophore was reported in this article, revealing the formation of a novel 8-hydroxyquinolin-imidazolinone (HQI) chromophore which has a significantly larger conjugated π-system in comparison to the parental 4-(p-hydroxybenzylidene)-5-imidazolinone (HBI) chromophore found in *Aequorea victoria* green fluorescent protein (GFP). These results indicate that HqAla incorporation into the FP fluorophore bestows it with unique metal-chelating and metal ion sensing abilities. Among all biologically relevant metal ions, only Zn (II) binding to HQI causes a significant increase (7.2 fold) in fluorescence. This selective turn-on FP sensor was then applied for Zn(II) sensing *in vivo*. Since HqAla can be synthesized through a pure enzymatic route in one step and with high yield and its 8-hydroxyquinoline motif binds strongly to most transition metal ions including lanthanide ions, the technological barriers which have limited the application of the genetic incorporation of metal-chelating UAA have now been broken. With this technology, the future is now even brighter for GFP engineering, metalloprotein engineering, and protein NMR using paramagnetic metal ions.




***A metal-chelating unnatural amino acid ...***

This work was supported by NSFC.