ABSTRACT

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TITLE: Synthesis and Photophysical Aspects of Fluorescent Biomolecular Probes for Serum Albumins Recognition

Serum albumins (SAs), a family of thiol-containing proteins such as bovine and human serum albumins (BSA and HSA), play a variety of key biological functions, including the removal of free radicals, regulating the pH balance of plasma, fluid transportation, maintenance of the blood osmotic pressure, preventing platelet aggregation, etc. A healthy adult's blood plasma normally contains 35 to 55 g/L of HSA, but in urine it drops below 30 mg/L. It has been reported that the levels of HSA in body fluids (blood or urine) are closely associated with the progression of many diseases. As a valuable indicator, HSA is commonly measured and has been recognized as an important diagnostic indicator for patients with critical illnesses, such as chronic hepatitis, liver cirrhosis, acute renal failure, diabetes mellitus and hypertension. Therefore, it is crucial to develop an effective chemosensor for selective recognition and quantification of HSA levels in biofluids, which is highly useful for the clinical applications. Various analytical techniques (like colorimetric, radiolabeling, immunoassays, electrophoresis, fluorometry, electrochemical, and LC–MS proteomics-based methods) have been developed effectively for the recognition of serum albumin, but, fluorometry is regarded as the most widely used technique in term of its easy operation, non-destructive nature, good selectivity, high sensitivity, quick response, and noninvasive benefits along with high resolution for real-time imaging *in vitro* and/or *in vivo*.

So, in this research endeavour, we have developed some simple fluorescent molecular probes, which can selectively and sensitively detect HSA/BSA over other biological interferences in purely aqueous medium. First, a TICT mechanism-based self-assembled molecular probe HJRA, was synthesized by incorporating a '*donor-* π *-acceptor*' system with julolidine as the electron donor and rhodanine as the electron acceptor moieties. This probe selectively binds at site II of HSA by undergoing disassembly, and emits intense red fluorescence at 582 nm (~298-fold) with a LOD value of 1.13 nM. Additionally, HJRA is also least cytotoxic, cell permeable and suitable for imaging of endogenous and exogenous HSA in living cells. Secondly, an ICT-based microenvironment sensitive fluorescent probe DCI-MIN was also developed for the selective and sensitive detection of HSA in complex biofluids. DCI-MIN has excellent photostability and exhibits a clear HSA induced large enhancement in emission intensity (~78-fold at 602 nm) with a significant (~126 nm) Stokes shift and very low LOD value of 1.01 nM. Thirdly, an NBD embedded olanzapine derivative, OLA-NBD was synthesized, which displays a ~5-fold fluorescence intensity enhancement upon addition of HSA with an association constant of $(9.87 \pm 0.02)\times10^{4}$ M^{-1} and the estimated LOD value is 59.4 nM. The fluorescence quenching study persuaded by OLA-NBD indicates the presence of a static quenching mechanism. Last one is a phenanthrene-pyrene-based fluorescent probe, PPI that can selectively bind with BSA protein and exhibits a fluorescence intensity enhancement at 457 nm with a LOD value of 6.87 nM. The study on fluorescence quenching induced by PPI reveals the occurrence of both static and dynamic quenching mechanisms.

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