THE STUDY OF N-ALKYL FUNCTIONALIZED INDOLENINE BASED SQUARAINES AS FLUORESCENT PROBES FOR PROTEINS DETECTION

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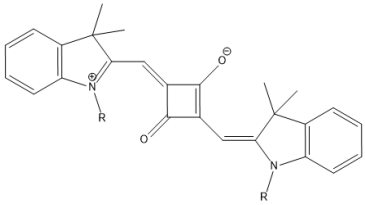
Squaraine dyes are widely studied as potential fluorescent probes for the labelling and detection of biomolecules, including proteins. Detection of proteins could be useful for chemical and biochemical analyses, biotechnology and immunodiagnostics. Serum albumins are multifunctional proteins present in the blood serum of animals, which bind and transport a variety of ligands. The study of interactions between dye and serum albumins is important for research as a simple diagnostic method for biochemical systems, because albumins concentration could be used as diagnostic parameter.

In previous work has been shown that squaraine dyes can specifically bind to proteins. Continuing the research we have synthesized novel N-alkyl functionalized indolenine based squaraines with the aim to create fluorescent probes for the detection of proteins, particularly albumins:



R= $CH_2(CH_2)_2SO_3Na$ (**sq1**), $(CH_2)_5COOH$ (**sq2**), $CH_2(CH_2)_5COOC_4H_9$ (**sq3**), $CH_2(CH_2)_5N^+(C_2H_5)_3$ (**sq4**)

We have studied fluorescent properties of N-alkyl functionalized indolenine based squaraines in the buffer solution and in the presence of bovine serum albumin (BSA), RNA and DNA by UV-vis absorption and fluorescence spectroscopies. The maxima of excitation spectra of the studied dyes in buffer are located at 620–640 nm with the fluorescence emission maxima lie between 638–645 nm. All of these dyes gave no significant fluorescent response upon addition of nucleic acids. For dyes – sq1 and sq2 (bearing sulfonate and carboxyl groups), binding to BSA resulted in the shift of excitation and emission maxima positions of fluorescence to the long-wavelength spectral region, up to 20 and 17 nm, respectively. Furthermore, these dyes showed a similar increase in fluorescence intensity with BSA (20 times for sq1 and 17 times for sq2). For dyes sq3 and sq4, in the presence of BSA excitation and emission spectral maxima of fluorescence weren't shifted relative to the corresponding spectra of the free dyes. The lowest emission intensity values, in the presence of BSA, were observed for sq4 dye. In the complex with BSA, the sq1 demonstrated increase in emission intensity of fluorescence in 22 times.

The quantum yield value for the dye sq1 in the presence of BSA increase up to 0.67 (while 0.065 in a free state). The wide linear detection range of BSA by dye sq1 is 0.03–5 mg/ml (R^2 = 0,96).

The change in spectral-luminescence properties of N-alkyl functionalized indolenine based squaraines dyes upon interaction with BSA makes them promising candidates for biomolecule detection.