Analysis of microheterogeneity of human serum albumin using a specific fluorescent probe and subnanosecond fluorescent spectroscopy. Experiments and medical applications.

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Human serum albumin (hSA, a globular protein, Mw approx. 67 kDa) is the main carrier and transport protein of extracellular human body fluids. Albumin executes a number of important functions (osmotic, ligand binding, detoxication, antioxidant, etc.) which are instantly related to special parts of its globule, binding sites. These sites are very sensitive to conditions within the body tissues and, as has been shown by our fluorescent experiments, are easily modified under either physiological or pathological conditions, e.g. during development of diseases. These essential physical-chemical modifications of the hSA result in microheterogeneity of albumin molecule population.

To study hSA binding sites, an original albumin-specific fluorescent probe CAPIDAN (a carboxyphenyl naphthalimide derivative) was synthesized. The probe-hSA interaction was studied in isolated hSA as well as in human serum. Some probe properties were estimated using ab initio quantum chemical calculations. Microheterogeneity of the hSA binding sites has been studied using subnanosecond fluorescence spectroscopy (single-photon counting technique). To characterize the hSA microheterogeneity quantitatively, an original method of fluorescence decay amplitude analysis was used. The results of experiments have shown that the hSA microheterogeneity is due to state of the main albumin binding region and can be described by six fluorescent parameters

These parameters are very stable in norm. However, in some diseases (peritonitis, pancreatitis, acute poisoning) they are sensitive to severity and outcome of the illness reflecting disease-induced hSA modifications. This hSA microheterogeneity was dramatically changed in mental disorders (paranoid schizophrenia, severe depression). We believe that fluorescent analysis of the hSA binding region microheterogeneity can be useful for recognizing severity of pathological processes and monitoring of the efficiency of their treatment. Clinical trials are in progress.

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