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An Energy Transfer Fluorescent Probe System for Glucose Sensor at Biomimetic Membrane Surface

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Abstract : Concanavalin A (conA) is a protein has been widely used in sensor system based on its specific binding to α-D-Glucose or α-D-Manose. For glucose sensor using conA, either fluoresence based techniques with intensity based or lifetime based are used. In this research, liposomes made from phospholipids were used as a biomimetic membrane system. In a first step, novel building blocks containing perylene labeled glucose units were added to the system and used to decorate the surface of the liposomes. Upon the binding between rhodamine labeled con A to the glucose units at the biomimetic membrane surface, a Förster resonance energy transfer system can be formed which combines unique fluorescence properties of perylene (e.g., high fluorescence quantum yield, no triplet formation) and its high hydrophobicity for efficient anchoring in membranes to form a novel probe for the investigation of sugar-driven binding reactions at biomimetic surfaces. Two glucose-labeled perylene derivatives were synthesized with different spacer length between the perylene and glucose unit in order to probe the binding of conA. The binding interaction was fully characterized by using high-end fluorescence techniques. Steady-state and time-resolved fluorescence techniques (e.g., fluorescence depolarization) in combination with single-molecule fluorescence spectroscopy techniques (fluorescence correlation spectroscopy, FCS) were used to monitor the interaction with conA. Base on the fluorescence depolarization, the rotational correlation times and the alteration in the diffusion coefficient (determined by FCS) the binding of the conA to the liposomes carrying the probe was studied. Moreover, single pair FRET experiments using pulsed interleaved excitation are used to characterize in detail the binding of conA to the liposome on a single molecule level avoiding averaging out effects.

Keywords : concanavalin A, FRET, sensor, biomimetic membrane
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