Infrared-Reflection-Absorption-Spectroscopy (IRRAS) of Lipid-Peptid Interactions in Monolayers at the Air-Water Interphase

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Infrared-reflection-absorption-spectroscopy (IRRAS) at the air-water interface is ideally suited to study the interaction of peptides and proteins with lipid monolayers. In contrast to transmission IR-spectroscopy, only the proteins bound to or incorporated into the lipid monolayers are detected by IRRAS. This facilitates the spectroscopic analysis of the amide bands of proteins caused by possible conformational changes upon binding. In addition, it is possible to analyze the conformation and the orientation of molecules in pure protein films or proteins bound to the lipid monolayers by recording spectra at different reflection angles and different polarizations of the IR light and comparing them with model calculations. Finally, a detailed analysis of the background absorption of water yields complementary information on the film thickness at the air-water interface and its changes.

Two examples for different binding modes of peptides and proteins to lipid monolayers will be presented. The first example is an amphipathic model peptide KLAL [1] which is highly surface active by itself and binds and incorporates strongly into lipid monolayers of negatively charged phospholipids. In bulk solution this peptide is random coil. When adsorbed to the air-water surface it is first α-helical and then converts to a β-sheet structure at the interface. This conversion is concentration dependent and can also be achieved by compressing a KLAL-film from very low surface pressure. Here, a decrease in pressure is observed when the peptide converts to the β-sheet structure. Bound to POPG films it is in the α-helical conformation and the helices are oriented parallel to the water surface. The α-helices penetrate the chain region, because the surface pressure increases on binding. Film expansion leads to a conversion of the α-helix to β-sheet. This conversion is reversible when the films are compressed again. Analogues of this peptide with double-D substituted amino acids behave differently.

The protein lactoferrin binds strongly to lipid A, the endotoxic principle of bacterial membranes [2]. The protein binding to lipid A monolayers is strong but occurs only peripherally to the headgroups. No increase in surface pressure is observed. The analysis of the IR spectra of the protein shows that no change in secondary structure is involved in the binding and that the number of bound proteins to the lipid A film is almost the same as the number bound to the air-water-surface.

References

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