Macromolecular Protein Assemblies from the Membrane and Solution Studied with LILBID-Laser Mass Spectrometry

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Biological molecules in membranes often form highly functional macromolecular assemblies, such as the complexes of the breath chain (I to V), ion-channels, transporters etc. Since they are partially hydrophobic their analysis represents a great challenge to mass spectrometry (MS). Nevertheless they are of greatest relevance and in their majority important drug targets. Solvable protein assemblies on the other hand are less demanding from the point of preparation but also often form highly functional, complicated macromolecular complexes or nanomachines such as ribosomes, polymerase, efflux pumps or oligomers of proteins such as those formed from Aβ–amyloid causing Alzheimer´s disease.
LLILBID (laser induced liquid bead ion desorption) allows to analyse biomolecules from on demand micro droplets (R=50 μm) in vacuum by laser ablation of preformed ions from solutionby means of a pulsed IR laser (3μm). The strength of interaction can be varied by the laser intensity. At low intensity large membrane molecules, solubilised in micelles from detergent, can thus be made "flying" and analyzed by TOF mass spectrometry (MS). At higher intensity noncovalently bound assemblies (complexes) may be fragmented into their subunits, allowing the analysis of their covalent building blocks. The method is very sensitive and requires only μl of solution at μM concentration for an analysis. Moreover it is tolerant to alkali salts, different buffers, detergents. doubly charged ions, pH etc. Therefore the ions may be studied in a more or less native environment, which is in general crucial for the formation of specific functional assemblies.
The talk will give an overview of the method, discuss results from complex I, from ATPases (complex V), and other nano machines and findings for the Aβ–oligomers and other soluble protein complexes etc.

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