cases causes problems during complementary SEM/EDS analyses when the sample is not transferred onto a different sample holder with regard to minimization of risk of its damage, loss or contamination. Therefore monocrystalline silicon plates with indicated conductivity of 5 $\Omega.cm^{-1}$ were tested in practice. This value is sufficient for SEM (based on measurements – carbon conductive strips specially intended for SEM have the conductivity of approx. 500 kΩ and higher). The plates with thickness of 300μm (±15μm) can also be used for subsequent direct FTIR analysis even in transmission mode, without the need of plate exchange. A method employing the image analysis system was introduced for precise sample adjustment for microdiffraction and the choice of the area to be measured. The trace of the primary beam on a fluorescent disc was scanned for different angles 2theta and subsequently saved as binary overlay images. A camera reads the live image of the sample through the focusing microscope and the operator can choose a spot for the analysis in ergonomic conditions (the second monitor was placed directly into the diffractometer chamber). This procedure is very convenient for heterogeneous samples, various abrasions, etc.

The system is further used for quantitative drug analysis. Both Rietveld method or method with the scale factor and the RIR (Reference Intensity Ratio) values (also called I/Ic values) are used. The results are cross-checked by standard methods of organic analysis (GCMS, FTIR), XRD analysis, in addition, enables precise phase analysis of inorganic components.

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Keywords: forensic, microdiffraction, microanalysis

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Strategies in the crystallization of a glycoallergen and a F1-ATPase complex

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Hypersensitivity reactions mediated by immunoglobulins E (IgE) are considered an important public health problem in developing countries. The IgE response against an allergenic protein is the sum of specific responses against each of its allergenic epitopes, which vary in intensity. Natural rubber latex (NRL) from *Hevea brasiliensis* contains several proteins and glycoproteins involved in this type of allergy. In general, these are involved in cross-reactivity against fruits, pollens and insect venoms. The structural information about this type of proteins is scarce and the cascade of events leading to allergy symptoms is also poorly understood. Among NRL allergens there are β-1,3-glucanases (Hev b 2), lectins (Hev b 6.02), class I chitinases (Hev b 11) and profilin (Hev b 8). We recently solved the structure of two polymorphic crystals of one glycosylated natural form of the Hev b 2 [1], at 2.5 Å and 2.8 Å resolution. One of them presented clear density for the oligosacharides. Interestingly, two glycosylation sites were found in the electron density maps. Appropriate buffers modified the solubility of the proteins. We have also solved the structure of two natural isoforms of Hev b 6.02 and three-dimensional studies for a recombinant Hev b 11 and Hev b 8 is underway.

We are also interested in the study of F1-ATPase from mitochondria, which is regulated by a low molecular weight inhibitor protein IF1. It has been reported that the endogenous complexes of F1 with IF1 exhibit a behavior that is different from that of complexes that had been reconstituted with exogenously added IF1. To get insight into the mechanism of action of IF1 we crystallized the endogenous complex F1-IF1 using two different approaches. Homogeneous preparation was fundamental; some preliminary results will be discussed.

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Lipid-Driven molecular complexes

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We describe three different self-assembled structures that are formed by lipid sequestration. The first two are designed lipopeptides that assemble into cylindrical micelles. The third is a two-component system consisting of an amphipathic protein forming a belt around a lipid assembly in discoidal lipoprotein particle.

Lipopeptide detergents (LPDs) are 25 residue peptides designed to form an alpha-helix with fatty acyl chains coupled to each end of the helix. LPDs form small 25-30 kDa micelles with aggregation number of 8-10, depending on the peptide sequence and the length of the acyl chains. We have previously described the 1.20 Å structure of LPD-12 [1], which forms octomeric micelles with all anti-parallel helices. We now report the 2.0 Å structure of LPD5Q-14, which is based on a different peptide sequence coupled to 14-carbon acyl chains. Crystals of LPDQ-14 contain 18 molecules per asymmetric unit, arranged into two 9-mer micelles. Both micelles have similar structures, and are made of up three units of "up-up-down" trimers. There are considerable differences between the LPD-12 and LPD5Q-14 micelles, but both consist of an outer alpha-helical shell surrounding an acyl chain interior, as designed.

We also report the 1.9 Å structure of saposin A in a discoidal assembly with interally-bound detergent molecules. Saposin A is an alpha-helical protein that exists in both soluble and lipid-bound states, and has superficial similarities to the exchangeable apolipoproteins. Our structure show a belt of two "open-state" saposin A molecules forming a belt around 40 detergents arranged in a bilayer-like arrangement. The structure gives insight into the biological functions of the saposins, as provides a high resolution view of a discoidal lipoprotein particle.

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