IMAGING NATIVE MEMBRANES WITH THE ATOMIC FORCE MICROSCOPE

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The cell membrane separates the inside of a cell from the outside world. Embedded in the lipid bilayer, the membrane proteins are the doors of the cells. With typical dimensions of 5-10 nm membrane proteins are nanomachines that fulfill key functions such as energy conversion, solute transport, secretion and signal transduction. Their central role in a wide range of diseases and their function as interface between the extracellular space and the cytosol may explain the fact that 70% of all drug targets are membrane proteins. The atomic force microscope (AFM) produces images with an outstanding signal-to-noise ratio and addresses single molecules under native conditions, keeping the sample in buffer solution.

Progress in sample preparation and instrumentation has led to topographs that reveal sub-nanometer details of biomolecules' surface structure (Engel & Müller 2000). The AFM can therefore be used to study structure and dynamics of membrane proteins embedded in native membranes. Alternatively, two-dimensional crystals of purified membrane proteins are assessed by AFM as well as electron crystallography to elucidate their structure at atomic resolution. Examples presented concern the prototype G-protein coupled receptor rhodopsin (Fotiadis et al. 2003) (Fig. 1), bacterial photosynthetic machinery (Fotiadis et al. 2004; Siebert et al. 2004) (Fig. 2) and members of the medically significant aquaporin family (Agre et al. 2002; Fotiadis et al. 2002).

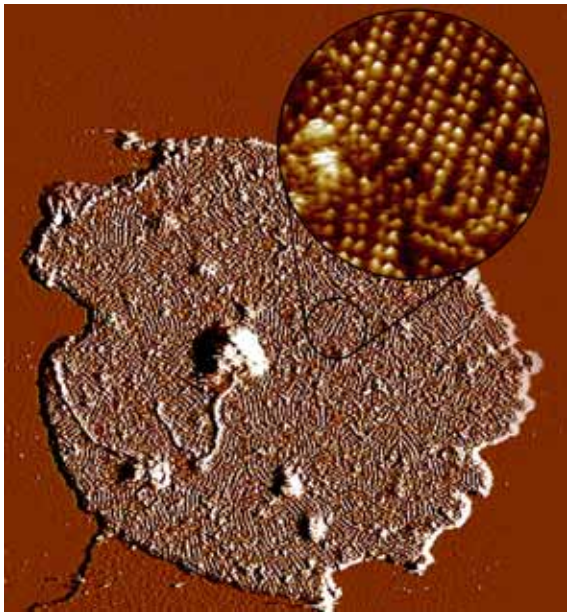

Fig. 1. Rhodopsin molecules are the light detectors of vertebrate's visual system and are densely packed in disk membranes of rod outer segments. Topographs of native murine disk membranes recorded in buffer solution using the AFM reveal the packing arrangement of rhodopsin molecules in rows of dimers. The diameter of the disk shown is 700 nm and the peak-to-to peak distance in a dimer is 3.8 nm. The disk overview image has been acquired in the deflection mode, whereas the high-resolution scan is a height image. Both images have been recorded in the constant force mode.

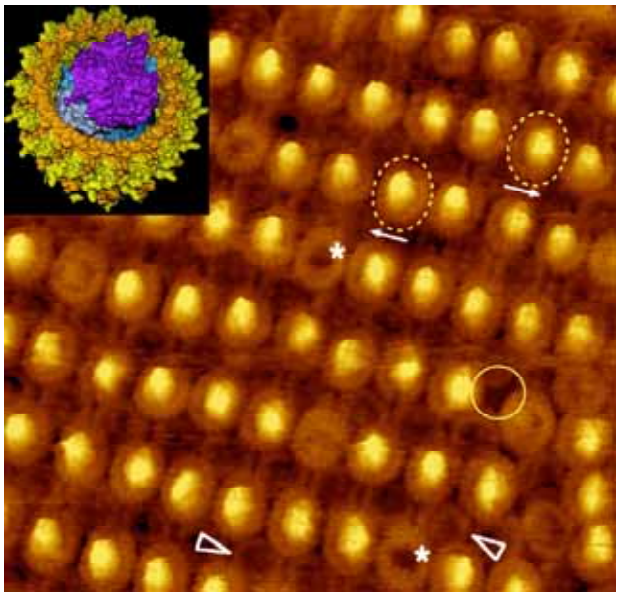

Fig. 2. High-resolution AFM topograph of a two-dimensional crystal of *R. rubrum* RC-LH1 complexes. The crystal is composed of alternating rows of complexes exposing either the high and prominent cytoplasmic side (bright rows) or the low periplasmic side (dark rows). A lipid region is marked by the solid circle. Asterisks denote "empty" LH1 complexes completely lacking the RC. The inset top left shows the atomic model produced from the atomic structures of RC and LH1. The scale bar represents 40 nm and the cytoplasmic side protrudes 4 nm from the bilayer.

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