Notes

s8a.m4.o5 **Crystallization of the reaction center of photosystem ii and of reaction center components.** N. Adir, R. Anati, V. Cheredman, Y. Dobrovetzki and N. Lerner. *Department of Chemistry and Institute of Catalysis, Science and Technology, Technion, Technion City, Haifa 32000 Israel.*
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Photosynthesis is the process by which plants transform the energy of the sun into useful chemical energy. The primary reactions occur between a variety of cofactor molecules bound to large membrane protein complexes called reaction centers. Of the different reaction center types, one of the most complex is that of Photosystem II, found in all higher plants, green algae and cyanobacteria. It is the site of the initiation of linear electron flow, proton gradient formation and oxygen evolution. Reaction center II (RCII) contains 8 trans- membrane proteins, a membrane associated protein subunit, 7 different cofactor types and has a molecular weight of about 280,000. RCII, isolated from either spinach or pea photosynthetic membranes, have been crystallized [1]. The crystals were grown from RCII monomers in the presence of mixtures of detergents at higher detergent/ protein ratio than previously reported for other membrane proteins. The crystals grew as hexagonal rods with dimensions of up to 1 x 0.3 x 0.3 mm and diffracted to a maximum resolution of about 6.5Å, and belong to a hexagonal space group with unit cell dimensions of a = 495Å, b = 495Å, c = 115Å, $\alpha = \beta = 90^{o}$, $\gamma = 120^{o}$. Size exclusion chromatography, dynamic light scattering and thermoluminescence were used to probe the RCII detergent interactions and identify source of heterogeneity prior to and after crystallization.

Because of the large size and complexity of RCII, we are also trying to isolate various protein components and determine their structure separately: *i*. Oxygen evolution in RCII is dependent on the presence of a tetramanganese cluster, protected by a membrane-associated protein called the manganese-stabilizing protein (MSP). It has been proposed that the MSP belong to the class of "natively unfolded " proteins. MPS has been isolate from spinach and purified to homogeneity. The MSP has been crystallized in the presence of calcium. MSP crystals were found by MALDI-TOF mass spectrometry to contain only a dimeric form which may stabilize the protein conformation. *ii*. RCII is connected to the main antenna, LHCII by a number of intermediate membrane bound antenna molecules. One of them, CP29, has been isolated and crystals have been obtained. *iii*. Within the core of all RCII complexes is a unique dipeptide 15kDa transmembrane b-type cytochrome called cytochrome b_{559}. Overexpression of a unique fused gene product, reconstitution and crystallization trials will be described.

[1] Adir, N. (1999) *Acta Crys.* D**55**, 891-894. (1999).
[2] Anati, R. and Adir, N. *Photosynth. Res. In press.*