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Seeding in Cubo: A Novel Approach for Growing Membrane Protein Crystals. Justin Robert, Kazumi Shimono, Ehud M. Landau, Kazumi Shimono, Javier Navarro, Membrane Protein Laboratory, Sealy Center for Structural Biology, The Univ. of Medical Branch, Galveston, TX 77555.

Growing well-ordered three dimensional crystals of membrane proteins for X-ray diffraction is one of the major challenging tasks in structural biology. In the mid 1990s a lipid matrix based on bicontinuous lipidic cubic phases was introduced for the crystallization of integral membrane proteins. The lipidic cubic phases display many features of native membranes including curved lipid bilayers delineating aqueous compartments. Most importantly, integral membrane proteins in lipidic cubic phases are stable. This procedure has been successfully applied for the crystallization and structural determination of few bacterial membrane proteins including bacteriorhodopsin, halorhodopsin, sensory rhodopsin II and anabaena sensory rhodopsin. The crystallization of membrane proteins from lipidic cubic phases present a number of experimental problems, such as a limited number of lipids that form cubic phases in the range of temperature for crystallization, and identification of colorless crystals in the lipid matrix. We have developed a novel technique for growing crystals of an unstable sensory rhodopsin II mutant from lipidic cubic phase using as seeds wild-type crystals.