NCCAT proposal for Chameleon usage:
Adaptation of the structure and function of Photosystem II to the high light stress condition photoinhibition

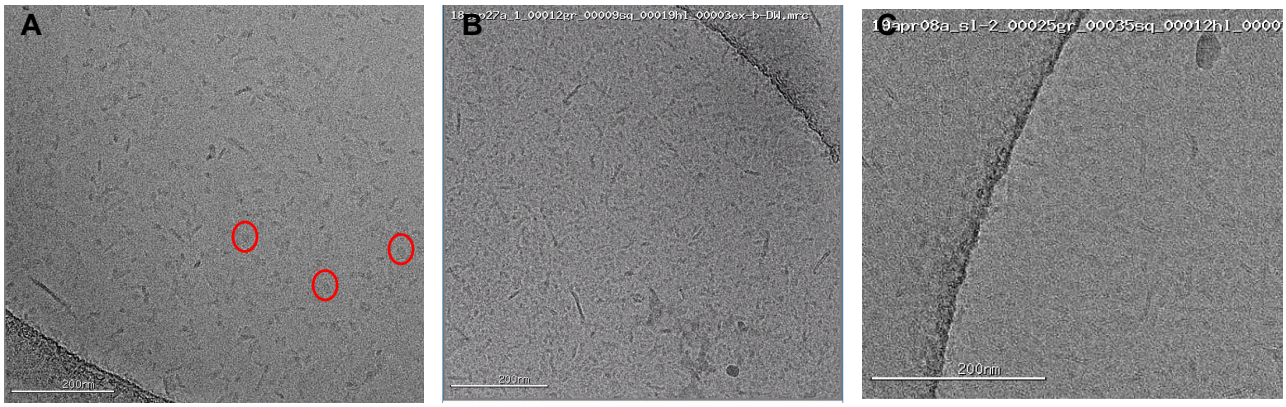

Fig. 1. Photoinhibition is associated with release of PSII extrinsic subunits. In order to study the structure under conditions relevant to photoinhibition, extensive testing of cryo-EM grid preparation conditions was performed for three different PSII samples. Micrographs of (A) the highly active PSII complex containing the three extrinsic subunits PsbO, PsbP, and PsbQ. Examples of individual PSII complexes are circled. (B) PSII depleted of PsbP and PsbQ while retaining PsbO, and (C) PSII depleted of all three extrinsic subunits. Data was collected on the Titan Krios at Florida State University with a K3 detector. The three representative micrographs show a reasonable number of particles. The particles with higher contrast are aggregates and particles that have aligned end-to-end or side-by-side. Chameleon tests are requested to overcome issues with low particle contrast due to the background, aggregation, and preferred orientation. The scale bars correspond to 200nm.

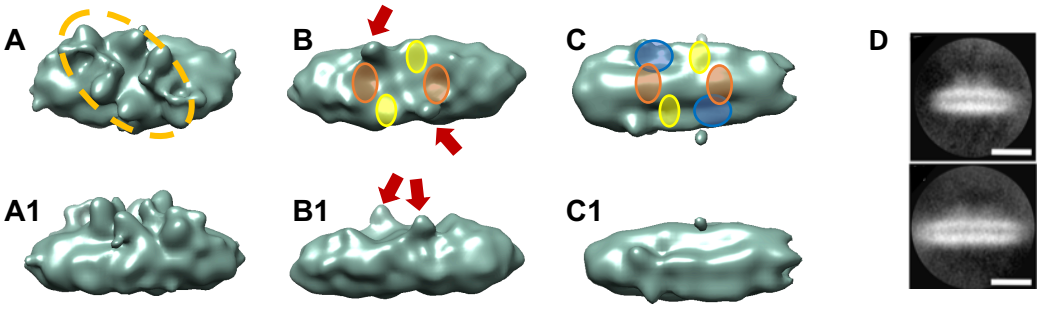

Fig. 2. Preliminary low-resolution models verify successful biochemical sample preparation targeting removal of specific subunits in order to understand key events during high light stress. (A) The sample is highly active and contains the extrinsic subunits PsbO, PsbP and PsbQ (within orange dashed oval). The model is shown perpendicular to the plane of the membrane. (A1) shows the model in (A) rotated by 90°. (B) A model of PSII shows that subunits PsbP and PsbQ (approximate previous location indicated by yellow and orange transparent ovals, respectively) have been successfully removed. The density for PsbO is indicated by red arrows. (C) The three extrinsic subunits have been removed (the previous location of PsbO is indicated by a blue oval). (D) Class averages of complexes with different lengths due to variation in peripheral subunits within the transmembrane region (scale bars correspond to 10nm). Chameleon usage may dramatically improve grid conditions and allow us to understand the details of conformational changes important under high light stress conditions.