

Development of a shell-focused approach to 3D particle picking in alpha-carboxysomes

R. Gray, L. Oltrogge, D. Savage, L.A. Metskas

When analyzing the arrangement of Rubisco in tomograms of alpha-carboxysomes, the Rubisco proteins must first be identified. The samples are heterogeneous and so the particles of interest must be separated out from the particles we are not interested in. The identification must be accurate and complete. False positives and especially false negatives will diminish the value of the data. These errors are not random and will affect the biophysical and chemical analysis that can be performed. The goal of this project was to develop a new method of isolating the Rubisco from the rest of the particles present in the tomogram. The previous method involved searching for Rubisco particles through classification using a Rubisco reference. The resulting classes would then have to be screened manually in a time consuming process. In the proposed method, the initial classification will target the component with the strongest signal, in this case, the protein shell of the carboxysome. This method results in clear, unambiguous classification of the shell particles which were able to be removed for further rounds of classification focusing on the Rubisco.