Diagram

Description automatically generated with medium confidence

**Figure 2-figure supplement 1. Structural determination of the Cel48S** **enzyme.** (A) A typical micrograph depicting the major cellulosomal exoglucanase, Cel48S. (B) The final 2D classification showing different orientations of this asymmetric protein. (C) The workflow of image processing for Cel48S. After several rounds of 2D classifications removing false positives, 362,956 particles were selected for initial 3D model building. The particles were subjected to a couple rounds of 3D classifications and refinement. The percentage of particles within the resultant 3D classes and the respective resolution of the 3D classes are visualized. Further refinement with 55,206 particles is shown in the black circle. After final refinement with B-factor of −110 Å2, a map of 3.66 Å was resolved. The map was post-processed with a solvent mask, and the particles were subjected to Bayesian polishing and CTF refinement. These operations improved the map to 3.38 Å. (D) The angular distribution of all 55,206 particles that contributed to the final Cel48S map is depicted. The height of the cylinder bars (from blue to red) is correlated with the number of particles in this view. (E) Local resolution variations in the cryo-EM map. The resolution ranges from 3.4-3.8 Å as calculated by the Relion local resolution option. (F) Fourier-shell correlation (FSC) plots of the final Cel48S density map. The plot shows the unmasked (green), masked (blue), phase randomized (red) and masking-effect-corrected (black) FSC curves. The resolution at which the gold-standard FSC curve drops below the 0.143 threshold is indicated (arrow).