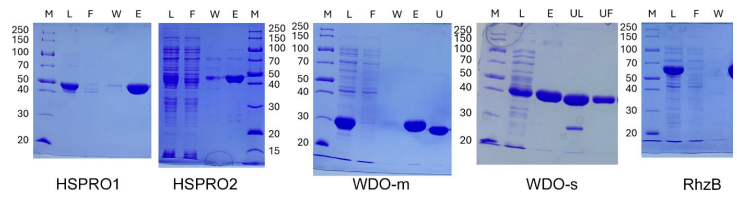

Figure 1. SDS-PAGE confirms the purity of IMAC-purified proteins. M: size marker (kDa); L: load; F: flow-through; W: wash; E: elution; U: untagged; UL: untagged load; UF: untagged flow-through.

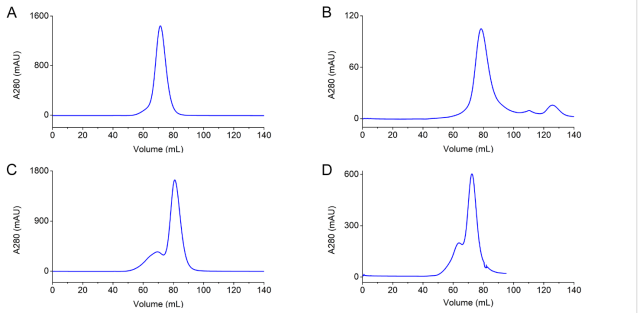

Figure 2. Size-exclusion chromatography confirms protein sample homogeneity. (A) HSPRO-1 (B) HSPRO-2 (C) WDO-m (D) WDO-s, using a Superdex 200 16/60 column (Cytiva).

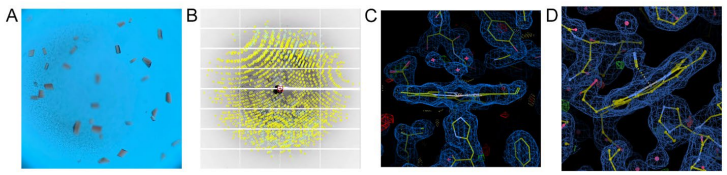

Figure 3. Crystal structures of WDO-m and WDO-s. (A) WDO-m crystal image. (B) WDO-m diffraction image. Heme active sites of WDO-m (C) and WDO-s (D) with $2F_o$-F_c electron density map at 1σ.

Figure 5. BIZON cryo-EM workstation with software

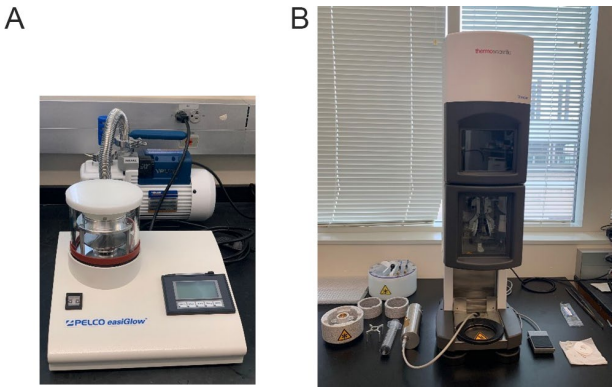

Figure 4. Photography of Pelco easiGlow (A) and ThermoFisher Vitrobot Mark IV (B)

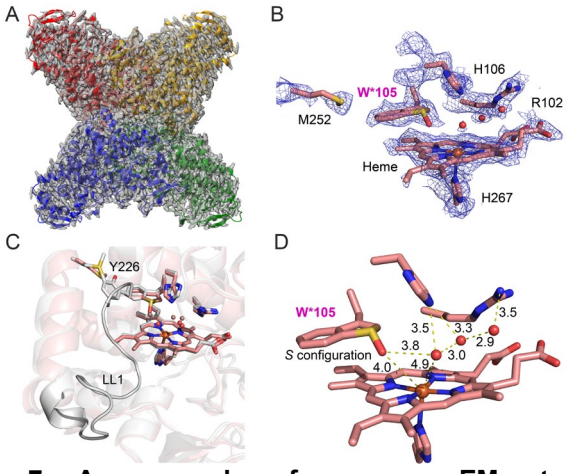

Figure 7. An example of our cryo-EM structural characterization (to be published)**.** (A) Cryo-EM density map and model of KatG S-Trp105 (W*) at 2.22 Å resolution, (B) Active site density map. S-Trp105 is surprisingly present as O=S-Trp in the *S* configuration, (C) Structural superposition of KatG S-Trp105 (salmon) and WT KatG (gray) with an RMSD of 0.440 Å for 611 aligned Cα atoms. (D) Detailed structural view of the active site, including the heme center illustrating key residues and their spatial relationships. Distances between selected atoms are indicated in angstroms (Å).

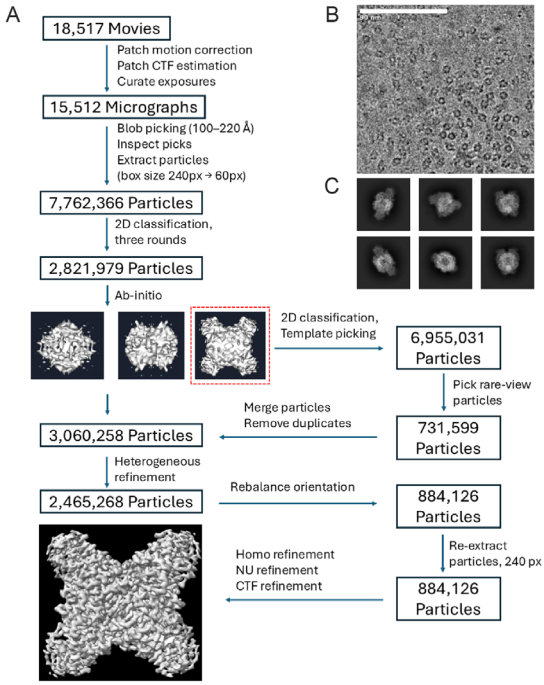

Figure 6. An example of our workflow of cryo-EM data processing for KatG with a genetically substituted S-Trp105 from Trp105 (to be published). (A) Flow chart of cryoEM data processing. (B) Representative cryoEM micrograph. (C) Representative 2D class averages.