Structural characterization of Coiled-Coil Protein Origami (CCPO) structures

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Coiled-coil protein origami (CCPO) uses modular coiled-coil building blocks for the de novo design of polyhedral protein nanostructures, using topological design principles distinct from natural globular proteins. While the CCPO strategy has proven successful in designing various protein topologies, obtaining high-resolution structural information has remained challenging due to the small size and high flexibility of these novel protein folds. To overcome these challenge, we employed specific techniques and approaches in our study. Our X-ray crystallography efforts yielded a high-resolution crystal structure of the triangular CCPO by implementing shorter linkers, more stable coiled-coil peptides, and incorporating a natural GCN homodimer. Additionally, we made progress with cryo-electron microscopy by utilizing nanobodies, which allowed us to obtain a highresolution structure of one vertex of the tetrahedral CCPO.

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