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EXPERIMENT REPORT

We diffracted crystals of bacterial wild-type FmtA, and also of the inactive mutant S127A, both crystallized or soaked with ribitol as a protomer of the wall teichoic acid substrate and also with ampicillin, since it is a protein of the penicillin binding protein family. We also diffracted crystals of Arabidopsis thaliana uracil UPP cro-crystallized with the substrate uracil.

In total, we tested ~55 crystals and collected 23 datasets. Best FmtA crystals diffracted to an improved resolution of 1.9–2.0 Å. This has allowed us to refine the wild-type model to higher resolution since previous datasets were collected at a resolution limit of 2.2 Å, and we also solved the structure of the inactive S127A mutant for the first time. However, we have not identified extra electron density that could be attributed to the bound ligands.

UPP crystals, on the other hand were of poor quality and grew in bundles that could not be easily separated while fishing in the loops. The datasets had superimposed diffraction patterns of multiple crystals and could not be indexed and processed correctly.

On the other hand, the experiments with HptCTH were uneventful. This is indeed a difficult protein that will require more work on the crystallization.

Overall, the performance of the beamline was excellent, and we solved a new structure of an important FmtA mutant. We did not achieve the expected results of FmtA or UPP in complex with ligands, and to do so, we will have to work on the crystallization conditions. The new data on S127A adds to the previous data collected at ESRF for the wild-type protein, which we expect to publish along 2023.

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