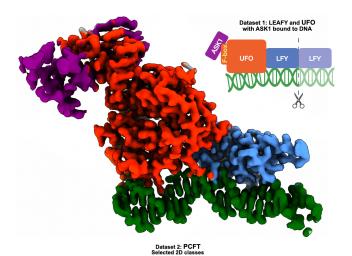
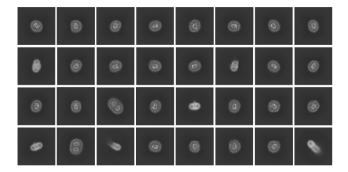
UFO sample

LEAFY is a plant-specific transcription factor that regulates flower development. UFO is a member of the F-box gene family in plants that acts as a LEAFY cofactor. A third protein (ASK1) that stabilizes UFO and a short ds DNA molecule that contains a specific sequence recognized by LEAFY-UFO are completing a complex of roughly 100 kDa. A first structure of this complex has been recently obtained at our Glacios 200kV-K2 summit system (Rieu et al. Nat. Plants 2023, 9, 315-329), although at medium resolution range (4Å). The aim of this project was to obtain a high-resolution structure of this complex in order to better understand the LEAFY-UFO synergistic role in transcription regulation. A first dataset was collected on CM01 on June 2022, on a similar grid as the one collected on the Glacios, but these data didn't yield the desired outcome mainly due to a severe preferential orientation problem, but also due to complex instability that caused many particles to lose at least one of the 5 partners/subunits. However, with the dataset 1 of this session that was our second attempt, we managed to reach our aim. After some optimization of the complex, we removed a few bases of the DNA molecule in order to reduce the copies of LEAFY to only one and also mutated some bases in order to increase the UFO/LEAFY affinity for the DNA molecule. This time we obtained a 2.5Å reconstruction that offers unprecedented insights into the interactions of UFO with LEAFY and of the complex of both with the DNA. We are currently building the model of the full complex, while preparing a manuscript that includes this structure.





PCFT sample

PCFT is a 40 kDa membrane transporter that carries folic acid into the human cells and is also the target of chemotherapy drugs. The aim of this project was to solve the structure of this tiny protein embedded in LMNG micelles. Our preliminary analysis of a Glacios dataset indicated that the particles could be efficiently aligned in 2D giving class averages with visible secondary structure features. Originally, we planned to acquire only a small number of movies as dataset 2 on the Krios in order to evaluate the benefits of a stronger set up combined with an energy filter. Since we saw that only a couple of thousands of micrographs of the dataset 1 was sufficient to obtained a high-resolution structure for the UFO/LEAFY complex, we decided to collect more data on dataset 2 and hopefully obtain a 3D reconstruction of PCFT as well. Circa 21 000 micrographs were acquired for dataset 2 and are currently being processed.