Structure analysis of transcription related complexes and installation of cryo-EM in KEK

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Transcription is fundamental process for withdrawing genetic information stored in the genome. In eukarya, multi-subunit complexes, known as RNA polymerase II, general transcription initiation factors, and chromatin factors, carry out this reaction. To elucidate the mechanisms of the reaction in detail, their tertiary structural information is indispensable. Here, we show the large-scale purification of eukaryotic transcription-related complexes for structural analysis. Preliminary cryo-EM analysis showed that these complexes seemed to be disrupted due to harsh condition during cryo-grid preparation. Further optimization of cryo-grid preparation conditions is required.

March 2018, our institute obtained 200kV cryo-EM and prepared pipeline for solving high resolution structures of protein complexes. From October 2018, our cryo-EM were open to public. One remarkable result is a single particle analysis of 110kDa enzyme at 2.85 angstrom resolution, suggesting that we had established a proper protocol for cryo-EM operation and single particle analysis. As a next step, we will continue to support external users and carry out cryo-EM analysis of transcription-related complexes.