Contribution ID: **27** Type: **Talk (17 + 3 min)**

Structural dynamics of substrate processing by the PAN-proteasome complex in solution: a time-resolved small angle neutron scattering study

Monday 20 March 2023 15:40 (20 minutes)

A multitude of structural biology techniques, including crystallography, NMR and cryo-EM, as well as single molecule experiments, have recently provided new and exciting mechanistic insight into protein substrate degradation by AAA+ ATPases and the proteasome. However, direct structural information on the conformational changes of the working complex and on the respective substrate state(s) and populations, during the active unfolding and degradation process in solution, remains scarce.

We use time-resolved small angle neutron scattering (TR-SANS), in combination with selective macromolecular deuteration and solvent contrast variation, to obtain structural information on the respective components during the active degradation process in solution.

By using the PAN-proteasome complex from the hyperthermophilic archaeon Methanocaldoccus jannaschii, it was possible to temperature-active and fine-tune the unfolding and hydrolysis process. Combined with online fluorescence, we were able to obtain separate structural information on the conformational state of PAN and on the GFPssrA substrate during the active reaction in solution. We find that PAN undergoes a reversible conformational contraction during the substrate unfolding process. GFP aggregates rapidly in the presence of PAN alone but is being hydrolyzed very efficiently once the proteasome is added to the reaction.

Primary author: GABEL, Frank (Insitut de Biologie Structurale, Grenoble, France)

Presenter: GABEL, Frank (Insitut de Biologie Structurale, Grenoble, France)

Session Classification: From Membranes to Kinetics and Dynamics

Track Classification: Health and Life Sciences