

BAG report_Wild (22/01/21 - 24/01/21)

HSPol Project Report

Our aim is to get a better understanding of heparan sulfate chain elongation, which is carried out by the EXT1/EXT2 enzyme complex. This hetero-dimeric complex catalyses the alternating addition of N-acetylglucosamine and glucuronic acid to the growing polysaccharide chain. In order to obtain a molecular insight into the complex architecture as well as into the catalyzed reaction mechanisms, we prepared a sample for cryo-EM analysis containing the EXT1/EXT2 protein complex and its UDP-N-acetylglucosamine and UDP-glucuronic acid substrates. A 24 hrs data acquisition took place on 26/10/21 – 27/10/21. A total number of 7046 images was collected at a pixel size of 0.84Å. To this date, data processing in Relion 3.1 provided a 3D-reconstruction at a nominal resolution of 2.7Å. We are currently building a structural model, which is straight forward in the very well ordered protein core, but more challenging in the rather flexible regions close to the active sites.