Principal Investigator: Jochen Zimmer
Grant Title: Structural and functional biology reveal the mechanism of hyaluronan synthesis

Abstract

Hyaluronan (HA) is a multi-functional extracellular glycosaminoglycan that is ubiquitously expressed in the human body. It is a linear polysaccharide consisting of alternating glucuronic acid and N-acetylglucosamine units that can reach thousands of sugar units in length. In vertebrates, HA is synthesized by a membrane-embedded processive glycosyltransferase, the HA-synthase (HAS), that recognizes UDP-activated sugars as substrates and couples HA synthesis with its translocation across the plasma membrane. HA translocation occurs through a pore formed by the membrane-spanning region of the enzyme.

We compared HA biosyntheses by bacterial, viral, and vertebrate enzymes and discovered (1) that bacterial HAS forms stable dimers in a native lipid bilayer environment while other enzymes do not, and (2) that the direction of chain elongation differs in bacterial and viral enzymes (*1*).

HAS' quaternary structure was determined using photo-induced bleaching of fluorescently-labeled enzymes during TIRF microscopy. Further, by imaging HAS and HA simultaneously, we were able to show that the quaternary structure of HAS does not change during HA biosynthesis and is therefore likely important for catalysis. In agreement with these observations, bacterial HAS elongates the reducing end of HA whereas viral and likely also vertebrate HASs extend the non-reducing end. Thus, our results suggest a fundamentally different mechanism of HA biosynthesis in bacteria compared to other model systems and our future studies will focus on a detailed characterization of human HAS.

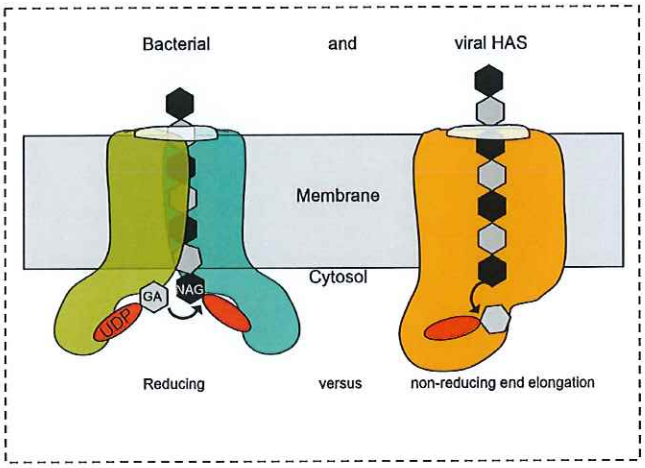

Figure 1: Bacterial and viral HAS utilize a different HA biosynthesis mechanism. Photo-bleaching of fluorescently-labeled HAS revealed that bacterial HAS dimerizes in a native-like lipid bilayer environment, whereas viral HAS does not. Further, only bacterial HAS elongates the reducing end of HA, enzymes from other model systems extend the non-reducing end (*1*).