Structural Studies of Secreted Autotransporter Toxin, A Serine- Protease Autotransporter of Enterobacteriaceae

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Secreted autotransporter toxin (SAT), a member of the Serine Proteases Autotransporters of Enterobacteriaceae (SPATE) family, has proteolytic effects against complement proteins of the host. SPATE's are associated with gastrointestinal and urinary infections and SAT is highly prevalent among E.coli strains. The N-terminal signal sequence exports SAT through the inner membrane and the active passenger domain is translocated to the outer membrane by the C-terminal β-domain. All SPATE's are cleaved at the linker domain between the passenger and β-domain after secretion while some other autotransporters remain intact. To date, most structures of autotransporters show the passenger domain is structurally conserved folding with a β-helix spine. However, SPATE's show weak sequence homology and there are structural exceptions of an α-helical domain, with no β-helix domain. Purification of SAT show a higher molecular weight band that may have the linker domain intact. We used negative stain electron microscopy and dynamic light scattering to screen for monodispersity of SAT. Our data show that SAT may form a high order oligomer, an exception to the conventional structural homology of SPATE's. Cryogenic electron microscopy (cryo-EM) will be utilized to determine if SAT forms an oligomeric complex. X-ray crystallography will be used to determine SAT if it forms a conventional SPATE monomer.
Further structural analysis will be reported.