Poster Presentation

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Cooperativity of folding in multidomain surface adhesins containing intramolecular cross-links.

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Gram-positive bacteria are decorated by many surface proteins including single molecule wide, elongated adhesins that form projections from the bacterial surface with "sticky" ends to provide adherence to host cells, other bacteria, and extracellular surfaces.

The C. perfringens adhesin Cpe0147 contains 11 repeat Ig-like domains that form a long stalk that projects a "sticky" adhesin domain from the bacterium surface. Each of the repeat domains contains an intramolecular ester bond formed spontaneously between the side chains of threonine and a glutamine residues [1,2]. These cross-links provide greatly enhanced mechanical, thermal, and often proteolytic stability to the entire adhesin. We have previously shown, using thermal stability experiments and circular dichroism, that single domain mutants unable to form cross-links, appear to be either unfolded or highly dynamic in solution. This poses a conundrum: how can these critical intramolecular ester bonds form if the protein domains are inherently unstable and unfolded until the bond is formed?

Our working hypothesis is that while single domains are inherently unfolded or dynamic, when part of the full polymer, adjacent domains cooperatively induce folding and bond formation even though they are separated in space by ~20 Å [1]. We are examining a combination of domain constructs using size exclusion chromatography with multi-angle laser light scattering (SEC-MALLS), differential scanning calorimetry, nuclear magnetic resonance, small-angle X-ray scattering (Fig. 1), and X-ray crystallography experiments to understand the potential for cooperative folding in these elongated, multi-domain surface proteins that allow bacteria to exist and flourish in a harsh host environment.

[1] Kwon, H., Squire, C.J., Young, P.G. & Baker, E.N. (2014). Proc Natl Acad Sci U S A., 111, 1367-1372.
[2] Young, P.G., Yosaatmadja, Y., Harris, P.W.R., Leung, I.K.H., Baker, E.N. & Squire, C.J. (2017). Chemical Communications, 53, 1502-1505.

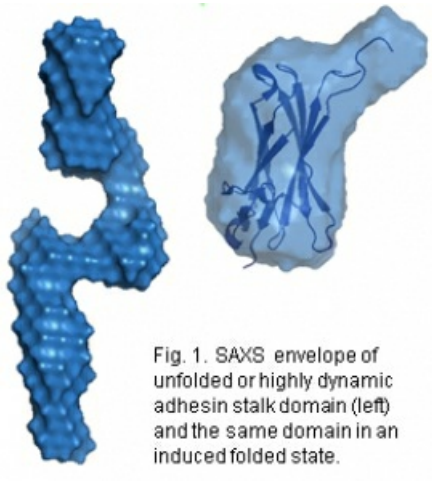

Fig. 1. SAXS envelope of unfolded or highly dynamic adhesin stalk domain (left) and the same domain in an induced folded state.

Keywords: cooperativity of folding, bacterial surface adhesin, intramolecular cross-link