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Single-molecule peptide sequencing - directed evolution of ClpS for an N-terminal amino acid binder

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A high-throughput method of peptide sequencing would be of great benefit to proteomics, personalized medicine and peptide drug discovery. A major hurdle in oligopeptide sequencing is discriminating positional information when identifying amino acids. Doing so currently requires analytical techniques that complicate high-throughput methodology. However, there are proteins found in nature capable of discerning amino acids with a c-terminal bond and a free n-terminal amine. We have adapted the bacterial protein *A. tumefaciens* ClpS, part of the N-end rule degradation pathway to a yeast surface display selection scheme, with the intent of selecting mutants able to tightly bind peptides in an N-terminus selective manner. We use a combination of a benchtop yeast pull-down assay and surface plasmon resonance to demonstrate our success.

Biography

Nicholas Callahan completed his PhD in the Biophysics Program of the Ohio State University, in 2015. He now works as a Postdoctoral Researcher for IBBR, an institute jointly funded by the University of Maryland and the National Institute of Standards and Technology, as part of the Kelman Group. The focus of his work is developing NAABs as part of a peptide sequencing collaboration.

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