

Chemical Engineering Colloquium

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12:00 PM – 1:00 PM/Goddard Hall 227

Engineering the Calcium-Dependent Beta Roll Peptide for Biotechnology Applications

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We have been exploring the beta roll forming repeats-in-toxin (RTX) peptide as a unique scaffold for protein engineering studies as it has the useful feature of being intrinsically disordered in the absence of calcium, and it folds into a well-defined 3-D structure in the presence of calcium. We have extensively characterized this tunable conformational change using a variety of techniques. We have explored the capping requirements for the scaffold, we have made synthetic peptides with a repeated consensus sequence, and we have concatenated beta rolls together to explore how this impacts the folding of the peptide. We have also immobilized the peptides to explore their functionality when tethered. Now we have begun to engineer the beta roll peptide for useful biotechnology applications. In the first line of work, we have engineered the faces of the beta roll with leucine side chains. This enables the beta roll to self-assemble in the presence of calcium and we have shown that this can serve as a stimulus-responsive cross-linking domain for use in protein hydrogel formation. We have also shown that a consensus sequence of beta roll domains reversibly precipitates in response to calcium and we have explored this as a novel protein purification tag that is more useful than the commonly used elastin-like peptide sequences. We have evolved beta roll mutants with affinities for target proteins using a variety of selection techniques. We have produced a mutant beta roll with calcium-dependent affinity for lysozyme and we have demonstrated the value of this peptide in affinity chromatography applications. We have inserted the RTX peptide into an enzyme to introduce calcium-dependent control over catalytic activity. And most recently we have been developing the scaffold for the binding and separation of rare earth elements. The most recent results of these efforts will also be presented.

