Session 1: Talk 1

Hanna King

Purdue University
PULSe/Chemistry, Graduate Student (5th Year)
PI: Angeline Lyon

Identifying Inhibitors of Phospholipase C β4 to Regulate CD8+ T Cell Activation

The adaptive immune system is essential for cardiac repair after a heart attack. A central component of this response are cytotoxic T cells as they clear cardiac damage and promote cardiac remodeling. Cytotoxic T cell dysregulation can lead to a plethora of comorbidities and mortality. Prolonged activation leads to accumulation of granzyme B, increasing the risk of death the year following a heart attack. However, complete abrogation of cytotoxic T cell activity increases the risk of cardiac rupture post-heart attack. Reduction of cytotoxic T cell activation could minimize long-lasting damage after an acute cardiac injury.

Phospholipase C β4 (PLCβ4) is a potential target for reducing cytotoxic T cell activity, as cells lacking PLCβ4 are weakly activated upon T cell receptor (TCR) activation. PLCβ4 hydrolyzes phosphatidylinositol-4,5-bisphosphate (PIP2) at the plasma membrane, generating second messengers that increase intracellular Ca^{2+} and activate protein kinase C, leading to cytotoxic T cell activation. Inhibiting PLCβ4 activity could reduce T cell activation through the TCR pathway. The goal of this project is to identify the first generation of PLCβ4 inhibitors that regulate T cell activity for future use in mitigating cardiac tissue damage following myocardial infarction.

Session 1: Talk 2

Samuel Baffoe

Purdue University
Biology, Graduate Student (5th Year)
PI: Leifu Chang

AI-Guided Design of a Compact RNA-Guided Transposon for Programmable Gene Insertion

Programmable genome integration represents a frontier in synthetic biology, and CRISPR-associated transposon (CAST) systems provide a powerful framework for RNA-guided DNA insertion. However, their large multi-subunit architectures limit delivery and modularity.

This project seeks to create a compact, single-protein CAST platform by directly coupling a single effector such as dead Cas9 to the transposition adaptor TniQ through a rationally engineered protein–protein interface. We employed an AI-integrated structural modeling and protein sequence design pipeline aimed at reprogramming the dCas9 surface to dock with TniQ in a geometry conducive to transposition.

This work combines deep learning, protein design, and biochemical validation to address challenges in domain geometry and interface engineering, establishing a blueprint for next-generation programmable transposon systems with broad applications in genome engineering, synthetic biology, and therapeutic delivery.

Session 1: Talk 3

Amr El-Arby

University of Notre Dame
Chemistry and Biochemistry, Graduate Student (5th Year)
PI: Shahriar Mobashery

Outer Membrane–Peptidoglycan Anchoring in *Pseudomonas aeruginosa*

Outer membrane anchoring to peptidoglycan is a defining but poorly understood feature of the Gram-negative cell envelope. In *Pseudomonas aeruginosa*, this linkage is mediated by covalent attachment of the outer-membrane lipoprotein OprI to the peptidoglycan layer, yet the enzyme responsible for this reaction has remained unknown.

Here, we identify PA2854 as the enzyme that catalyzes OprI–peptidoglycan attachment in vivo. Using purified PA2854, OprI, and synthetic peptidoglycan substrates, we reconstituted the reaction in vitro and demonstrated site-specific coupling of the ε-amino group of OprI Lys83 to the peptidoglycan peptide stem.

Structural and biochemical analyses define the molecular mechanism of PA2854-mediated outer membrane anchoring and establish this non-redundant reaction as a key contributor to Gram-negative envelope integrity.

Session 1: Talk 4

Emmanuel Oluwarotimi

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PULSe/Biochemistry, Graduate Student (4th Year)
PI: Andrew Mesecar

Biochemical and Structural Characterization of *Candida* GTP Cyclohydrolase I

The growing burden of antifungal resistance and increasing mortality from *Candida* infections highlight the need for new therapeutic targets. GTP cyclohydrolase I (GCH1) catalyzes the rate-limiting step in tetrahydrofolate biosynthesis and is essential for fungal DNA replication and survival.

This study provides the first biochemical and structural characterization of GCH1 from multiple *Candida* species. Kinetic, thermal stability, mass photometry, cryo-EM, and mutagenesis analyses reveal conserved decameric architecture alongside species-specific oligomerization behavior.

These findings identify structural features that may be exploited for future antifungal drug development.

Session 1: Talk 5

Rohan Bhardwaj

Purdue University
Chemistry, Graduate Student (5th Year)
PI: Jonathan Schlebach

Surveying the Sequence Constraints of Folding on the Ribosome by Deep Mutational Scanning

Protein folding begins during the earliest stages of biosynthesis on the ribosome, yet relatively little is known about how the ribosome influences these initial folding pathways.

To address this challenge, we developed an arrest peptide-based fluorescence reporter and deep mutational scanning approach to explore the sequence constraints governing cotranslational folding of *Escherichia coli* dihydrofolate reductase (DHFR). A codon-saturated library of approximately 9,500 DHFR variants was generated and analyzed using fluorescence-activated cell sorting and deep sequencing.

This work provides a comprehensive view of the sequence constraints associated with late-stage cotranslational folding and establishes a generalizable strategy for mapping folding intermediates in diverse proteins.

Session 1: Talk 6

Shalini Iyer

Purdue University
Chemistry, Research Scientist
PI: Chittaranjan Das

Allosteric Rewiring of Actin-Activated Ceg14 by LegA11 Inhibits ATP-to-AMP Hydrolysis

Bacterial effectors often depend on host factors for activation. The Legionella effector Ceg14/SidL catalyzes ATP-to-AMP hydrolysis upon actin binding and plays a role in host cytoskeleton modulation. Another Legionella effector, LegA11, inhibits Ceg14 activity.

Using cryo-electron microscopy, we determined the structure of the ternary Ceg14–actin–LegA11 complex at 3.57 Å resolution, revealing the molecular basis of actin activation and LegA11-mediated inhibition. Actin binding stabilizes a catalytically competent active-site arrangement, whereas LegA11 inhibits activity through a distinct allosteric mechanism without displacing actin.

These findings reveal a novel example of effector–effector regulation achieved through non-competitive inhibition and provide new insights into bacterial manipulation of host-cell processes.