Targeted Project / AY 2024 -2025

The early life of membrane-forming enzymes

Project Code: TRG-BIOC-CB

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Research area: Biochemistry, prebiotic chemistry, protein engineering, membrane engineering, biophysical chemistry

BBSRC DTP main strategic theme: Understanding the rules of life

BBSRC DTP secondary strategic theme: Transformative technologies

Project outline:

The advent of functional primitive membranes likely resulted from the synergistic emergence of self-assembling compartments and the catalysts responsible for synthesising the compartment precursors. It follows that the diversity of modern lipids might derive from the ability of ancestral enzymes to generate and modify membrane precursors. As such, identifying those ancestral enzymes (and their modern analogues) will lead us one step closer to understanding how functional primitive cells emerged and to developing new ways to engineer artificial life-like compartments. We will test the ability of (primitive) enzymes to generate membrane-forming components by using ultrahigh-throughput screening of >107 enzyme variants a day. Specifically, gene libraries (metagenomic or designed) will be compartmentalised in microfluidic droplets, followed by in vitro expression of the corresponding enzymes. The ability of these candidate enzymes to convert substrates into amphiphilic components will be tested at high/low temperatures and high/low pH values: only droplets generating components capable of enhancing membrane stability under extreme conditions will be selected. Screening of metagenomic libraries (e.g., from extreme environments) and primordial substrates will lead to the discovery of yet-unexplored chemical routes towards functional amphiphilic molecules boosting membrane stability. Directed evolution of enzymes that tailor small molecules to integrate into membranes will be possible with hydrolases, including phospholipases, which modify polar headgroups, thus creating surfactant properties. This approach will likely generate new clues vis-à-vis the origins of the universal organisation of biology, provide functional insight into hitherto unpredictable membrane-forming properties, and offer unprecedented insights into the molecular relationship between protocell structure and function.