Through the implementation of designable genetic circuits, probiotic strains of bacteria can be used as non-invasive diagnostic tools for the gastrointestinal tract. In order for these programmed cells to be able to detect and report disease biomarkers after exiting the gut, the genetic circuits need to be able to record these signals it received within the gut environment using genetically-encoded memory. Complex memory register circuits would allow for multiplex interrogation and detection of disease biomarkers. We have developed a computational approach for the scalable design of genetic circuits that contain memory, which are known as sequential logic circuits. Importantly, these sequential logic circuits can also be implemented for temporal programming of cells. The theory-based approach to design sequential circuits from simple NOT gate responses is robust and makes accurate predictions for standard cell growth conditions. However, the way in which circuit component performance varies for different bacterial strains and gut-relevant environments is poorly understood and could lead to loss of performance of the circuit. Here we aim to develop a computational approach for the design of robust sequential logic circuits for in vivo diagnostic and therapeutic applications. In this work, we use a TetR family of repressors to build NOT and NOR logic gates that can be composed into complex sequential circuits. The NOT gates were characterized in the probiotic strain Escherichia coli Nissle 1917. Using this data, we designed and predicted the behavior of larger circuit designs. We present a set of genetic circuits that encode combinational logic and sequential logic and show that the circuit outputs are in close agreement with our quantitative predictions from the design algorithm.
This project employed giant unilamellar phospholipid vesicles to study the attractive and repulsive interactions between solid membrane domains as plate-like inclusions both experimentally and theoretically using continuum models. In contrast to the well-known repulsive interactions in fluid-fluid phase separation, we found a significant qualitative departure in pairwise solid domain interactions by developing protocols to control the domain numbers per vesicle. Other than previous work has only observed and reported repulsive interaction or attractive interaction happen individually, we discovered that pairs of microscopic solid membrane coexisting (two per vesicle) within an Lα membrane fluid bilayer, exhibit long range potentials that can be both attractive and repulsive with a distinct minima or preferred separations. These interactions result from the shear elasticity of the solid domains, beyond simply enhanced bending stiffness and shear rigidity, solid domains tend to expel Gaussian curvature into the fluid membrane phase, which generically competes with the global spherical topology of the vesicle. We also discovered the ability to toggle interactions by osmotic adjustment or mechanical manipulation of the fluid membrane by manipulating the ratio of membrane area to vesicle volume and employing micropipettes or osmotic pressure.

This work investigates reversibly manipulation of pairwise interactions and adjusted separations between flat-shaped “2D” colloids dispersed in fluid membranes whose interactions are tunable through global vesicle properties, which suggested useful for positioning control of objects on surfaces or pattern formation in materials for a new class of highly flexible ultrathin materials with dynamic responsive functional patterns for a variety of applications.

(This work is collaborated with Dr. Hao Wu and Prof. Gregory Grason from Polymer Science and Engineering Department at UMass Amherst.)

References: