Title: Programmed Lysis of Cells in Response to Electrogenetic Inputs

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Project Goals: The LLNL Secure Biosystems Design Scientific Focus Area (SFA) aims to develop robust biocontainment mechanisms at the sequence, cellular, and population levels to safeguard the deployment of genetically engineered, soil bacteria. In this portion of the project, we are focused on developing robust and generalizable cellular and population level containment mechanisms for improving the safer use of plant benefiting microbes in the rhizosphere.

Abstract Text: Cell death and lysis play an important role in a wide array of biological niches. By genetically "programing" microbial cell lysis, we might provide a new methodology for ensuring biocontainment of cells within natural ecosystems as well as for engineered microbial consortia. Understanding how lysis and its control give rise to observed population dynamics is challenging as few methods exist that enable precise measurement and control. To this end, we have developed tools that link an externally applied electronic potential to initiate, measure, and even control genetic circuits that biologically mediate cell lysis. We first explore direct electronic activation of cell lysis within a clonal population of cells through OxyR-mediated expression of lysis protein E (here abbreviated LysisE), a lytic protein from φX174 phage that disrupts peptidoglycan synthesis. This genetic circuit can plug directly into electrogenetic control through detection of hydrogen peroxide produced at a gold electrode surface. Next, we show that by transforming electronically produced hydrogen peroxide into native biological signals that induce quorum sensing, and, in turn, LysisE expression in a separate population, we can dramatically enhance cell lysis overall, preventing significant regrowth or remodeling of the overall population for at least 6 hours. Lastly, we show that by combining the hydrogen peroxide induced synthesis of quorum sensing signals and the quorum sensing induced expression of LysisE in the same population, we can create a self-lysing transmitter of lytic signals that can simultaneously lyse neighboring bacteria. Each of these methods should enable deployment in agricultural (i.e., rhizosphere) or industrial process settings.

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