ACS SyntheticBiology

In This Issue

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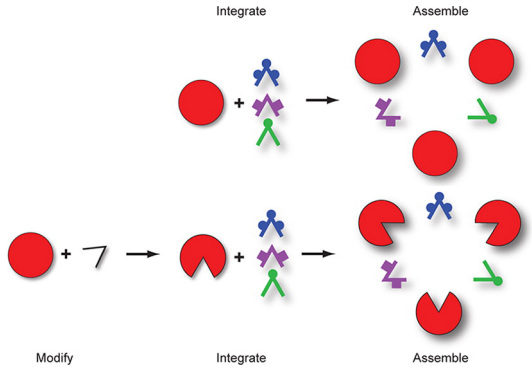
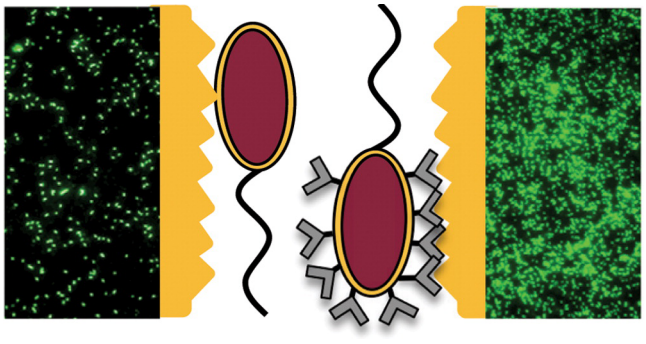
PROMOTERS FROM BIOBRICK FUSIONS



In the field of synthetic biology, a biological part is defined as an engineered, well-characterized piece of DNA with defined structure and function. While these parts are assumed to be functionally independent when used together, Yao et al. (DOI: 10.1021/sb300114d) now call into question this notion of contextual independence of functions in standardized biological parts. The authors describe the unintended creation of a relatively strong constitutive promoter in a commonly used *E. coli* BioBrick (Bba_C0051), through the addition of DNA encoding an epitope tag, a standardized double stop codon, and a DNA bar code to the gene encoding the cI repressor. The study also demonstrates how easily such "errors" in single parts can propagate through a parts registry when parts are commonly used and reused in composite devices. These results highlight the informational plasticity of DNA and how it can complicate the construction of synthetic devices by standardized biological parts.

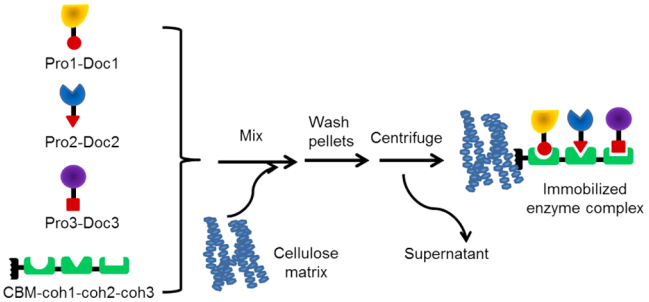
ANALYSIS OF ENGINEERED *SHEWANELLA ONEIDENSIS* TO BIND GOLD ELECTRODES



Gold electrodes are commonly used in electrochemical interrogation of proteins and bacteria that have the ability to respire insoluble substrates. *Geobacter* and *Shewanella* are the two best-studied bacteria that can respire electrodes and are themselves interesting chassis for future synthetic biology efforts. While *Geobacter* happily forms robust biofilms on gold surfaces, *Shewanella* cannot. Here, Kane et al. (DOI: 10.1021/sb300042w) took a synthetic biology approach to remedy this deficiency.

Using an outer membrane protein from *E. coli* into which the authors had engineered a 5x repeat of a Gold Binding Protein, they dramatically increased the ability of the cell to adhere to gold surfaces. However, they also report the displacement of many of the native proteins that are known to be essential to move electrons to insoluble substrates, highlighting the challenges in merging synthetic biology approaches, native cellular pathways, and cell surface charge.

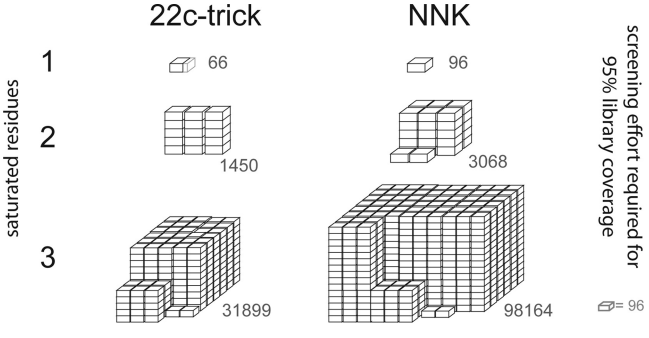
SELF-ASSEMBLY OF SYNTHETIC METABOLONS



In vitro synthetic biology is an important direction of synthetic biology. Compared to *in vivo* synthetic biology, cell-free biosystems are open to easy access and control and have a number of applications including biofuel production. However, the preparation of a number of *in vitro* building blocks is labor intensive and expensive. Here, You and Zhang (DOI: 10.1021/sb300068g) describe the development of a general one-step mechanism to purify multienzyme complexes from a mixture of cell extracts, using a low-cost cellulosic material.

The authors assembled three-enzyme complexes, called synthetic metabolons, using the high-affinity interaction between the docker in each enzyme and three cohesins in the synthetic scaffolds. These immobilized or free metabolons exhibited a significant increase in initial reaction rates compared to that of the noncomplexed three enzyme mixture at the same enzyme loading, suggesting that the use of synthetic metabolons not only decreases protein purification labor and cost but also accelerates reaction rates by 1 order of magnitude. Synthetic metabolons thus have the potential to be an important biocatalytic module for *in vitro* and *in vivo* synthetic biology projects.

REDUCING CODON REDUNDANCY AND SCREENING EFFORTS



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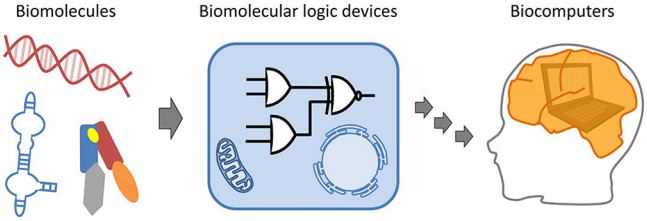
Any type of genetic, metabolic, or genomic engineering requires a fundamental understanding of proteins and of the proper use of available knowledge to evolve these proteins for desired traits. Saturation mutagenesis has now become an essential tool in protein−protein interaction studies, protein engineering, and directed evolution. Thus, any strategy that significantly reduces the waste of resources and the likelihood of misinterpreting results, while increasing the probability of efficiently exploring the sequence space of proteins, must be seriously considered. Here, Kille et al. (DOI: 10.1021/sb300037w) demonstrate the striking effect of codon redundancy removal on screening efforts.

The authors introduce a new degeneracy for saturation mutagenesis that encodes all 20 canonical amino acids in 22 codons. This new degeneracy, named 22c-trick, was achieved by mixing nonredundant degeneracies of NDT (12 codons) and VHG (9 codons) with a TGG codon. This manuscript discusses the impact of the described strategy on the probabilistic nature of the screening process and the importance of introducing a "Quick Quality Control" before screening, to assess the success of the expected degeneracy.

SYNTHESIZING BIOMOLECULE-BASED BOOLEAN LOGIC GATES



Synthetic biology intersects engineering and biology and has now become a highly powerful tool in making use of biomolecules, cells and even organisms for the betterment of human health and society. In this Review, Miyamoto et al. (DOI: 10.1021/sb3001112) thoroughly recapitulate the recent advancement in the creation of biocomputers with a particular focus on logic gates, the minimal computational unit in computers. The information presented in this review promises to be of considerable significance to engineers and biologists and both clinicians and industrial experts.

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