AN *IN VITRO* AUTOGENE

Technological advances have allowed synthetic biologists to better mimic cellular environments in a cell-free manner. This enables the development of proto-self-replicating systems and encourages efforts toward creation of minimal cells from a "bottom-up" approach. Here, Davidson et al. (DOI: 10.1021/sb3000113) detail the development of a cell-free, self-amplifying genetic system capable of propagation through multiple cycles of evolution.
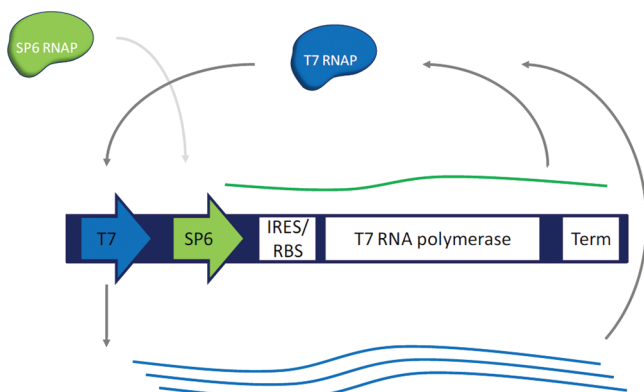
The authors describe a highly mutagenic system composed of a RNA polymerase autogene in a compartmentalized transcription and translation reaction. The highly mutagenic nature of the system rapidly leads to population diversity from a single starting sequence. The *in vitro* technology limitations and evolutionary implications of such a system are discussed.

GENETIC CONTROL OF METABOLIC FLOW

Manipulation of metabolic pathways is a mechanism often used for the production of chemicals in transgenic microorganisms. In this process, exogenous genes required for the production of target chemicals are expressed under strong promoters, while endogenous genes are suppressed. These manipulations often result in adverse effects on the host, slowing down growth and thus reducing efficiency. However, Yamanishi and Matsuyama (DOI: 10.1021/sb200017p) now describe the use of a genetic switch as a novel mechanism to overcome these obstacles.
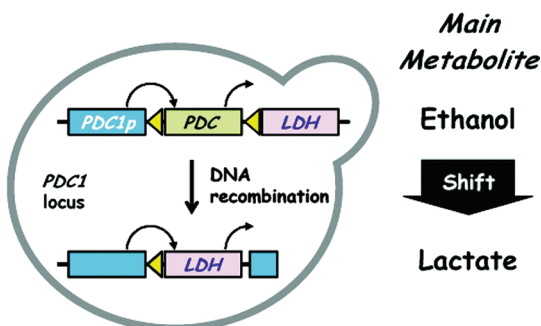

The authors describe the use of a Cre-*lox* genetic switch in lactate-producing yeast *Saccharomyces cereviesiae* to drive the production of the target chemical, lactate. This method enables transgenic yeast to produce lactate from up to 85.4% of its glucose substrate and may prove to be a useful tool in the improvement of biofuel and Bioplastic production.

SMALL RNAS THAT ACTIVATE BACTERIAL TRANSCRIPTION

There is increasing awareness of the role of RNA in cellular processes in both eukaryotes and prokaryotes; control of gene expression by small, non-coding RNA (sRNA) has been shown to play a role in genetic regulation. To date, several sRNAs that function at the transcriptional level and a few that can inhibit transcription have been identified. Here, Goodson et al. (DOI: 10.1021/sb2000275) investigate whether sRNAs in bacteria can activate transcription.
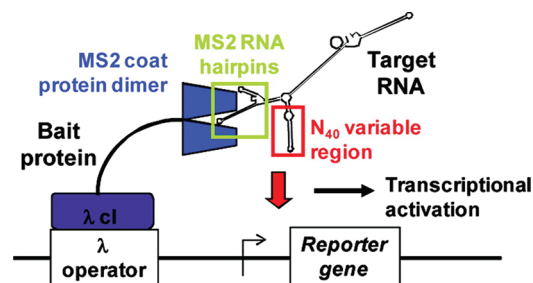
An *in vivo* selection procedure utilizing a random RNA library screen was used by the authors to identify sRNAs that activate transcription. Mutation of the strongest activator sRNA identified by this screen resulted in activators with a range of activator potentials, the best exhibiting an 8-fold increase in transcriptional activation compared to the unmutated parent sequence. This is the first description of RNA-based transcriptional activation in bacteria through the recruitment of transcriptional machinery and adds a new, important component to the tool kit of synthetic biology.

TETRACYCLINE-REGULATED GENE EXPRESSION SYSTEMS

Ectopic gene expression in mammalian cells has long been controlled by tetracycline-regulated expression systems. However, background expression often limits the use of these systems in applications that require low level control of expression. Here, Peacock et al. (DOI: 10.1021/sb200029a) describe the development of a new tetracycline-regulated expression system using a single tetracycline-responsive genetic element.

Using a system that repressed the expression of both the target gene of interest and the transcriptional activator, the authors were able to achieve increased, inducible range of control with decreased background expression. This work promises to have a significant impact on the field on synthetic biology by allowing scientists to take advantage of long- and short-range capabilities of synthetic transcription factors.
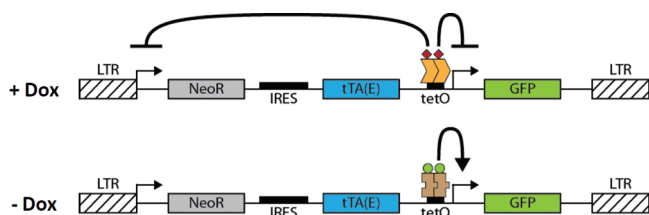
■ SYNTHETIC CONTROL OF GENE CIRCUIT MODULAR DESIGN

While the use of engineering principles to design gene circuits is one of the trademarks of synthetic biology, the increasing complexity of these gene-circuits demands a more rational approach to network design. The "bottom-up" approach describes a method of assembling complex devices from well-characterized elementary components. However, it is unknown whether the behavior of the final device can be predicted simply from that of its modular parts. Here, Ceroni et al. (DOI 10.1021/sb200021s) test this "bottom-up" approach to gene circuit design and highlight its limitations using quantitative tests.
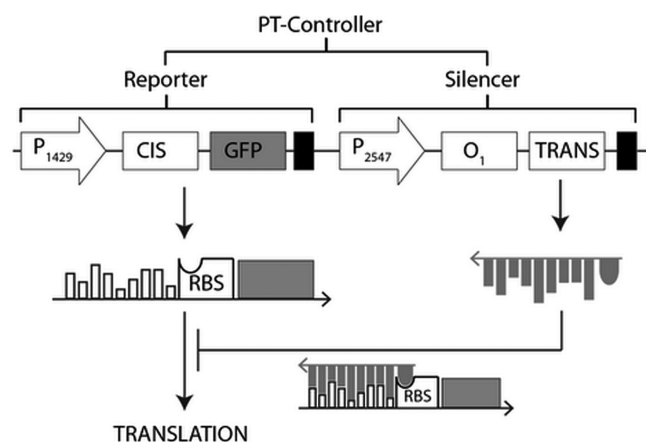
The authors describe a device for post-transcriptional control of gene expression, assembled from previously characterized modular parts. The behavior of the complete gene network was simulated using mathematical modeling, while the modularity of the device was evaluated by comparing experimental data and model results. The authors show that modular parts may be unpredictable when assembled into more complex circuits and stress the need for new strategies for the rational design of synthetic devices.