Methods of Evaluating Gene Expression Levels

Technology #15194

Applications

The ability to build a higher level representation of a novel biological design from known parts, with flexible protocol automation and DNA expression characterization, is useful in any industrial or academic setting where accurate predictions of the protein expression output of a genetic circuit is relevant to the project design.

Problem Addressed

The ability to accurately predict the expression levels of biological circuits is built on an understanding of the behavior of the individual elements involved in the biological circuit. Previous prediction methods relied heavily on human experience to supplement insufficient data. This design process is often inconsistent and difficult to replicate. A new system of tools that uses modular data on the input and response dynamics of individual circuit elements to accurately and consistently predict circuit behavior would greatly advance the field of knowledge and provide researchers with a systematic approach to the design and implementation of circuits.

Technology

This technology includes a pipeline to characterize the expression levels of test proteins whose expressions are regulated by test regulatory elements. These test proteins are actively transcribed when a constitutively expressed effector-regulated protein is activated by an effector molecule and binds to the test regulatory element controlling the expression of the test proteins. Input reporter proteins controlled by the same test regulatory elements as the genetic elements are used to quantify the level of effector protein activity, while output reporter proteins controlled by the proteins encoded by the genetic elements are used to quantify the level of expression of the genetic element itself. The process for collecting data on this circuit includes first transfecting a population of cells with plasmids that contain separate elements of the genetic circuit, followed by the transfecting cells with a plasmid containing all the elements of the genetic circuit, then by integrating the entire circuit within the cells' chromosomes, and finally by transfecting cells with a non-replicable single-copy plasmid containing all the elements of the genetic circuit. At each step of the way, the cells are screened by fluorescence activated cell sorting (FACS) to measure the expression levels of input and output reporter proteins in order to determine the activities of the effector-regulated protein and test protein. The data, which can be collected as early as two weeks into the characterization process, are parsed and analyzed by the BioCompiler software system to determine the expression curve of the test proteins. A separate MatchMaker software system can use this data to predict the signal compatibility of test proteins when building a circuit containing multiple test protein elements, and the Puppeteer software system then automatically generates a protocol for a liquid-handling robot to implement the circuit designs in the laboratory.

Advantages

255 Main Street, room NE 18-501
Cambridge, MA 02142-1601
Phone: 617-253-6966 Fax: 617-258-6790
http://tlo.mit.edu
Contact the Technology Manager: tlo-inquiries@mit.edu

- High-level representation of biological designs with BioCompiler
- Automated DNA part assignment with MatchMaker
- Flexible protocol automation with Puppeteer
- High-throughput DNA element characterization method with multiple data collection points and fast turnover time

Categories For This Invention:

Life Sciences
Biotechnology
Research Tools
DNA
Other (Research Tools)
Screening Assays
Transgenic
Vector & Plasmid
Synthetic Biology
Bacterial
Mammalian

Intellectual Property:

Methods of evaluating gene expression levels
Issued US Patent
8,809,057

Inventors:

Ron Weiss
Noah Davidsohn

Publications:

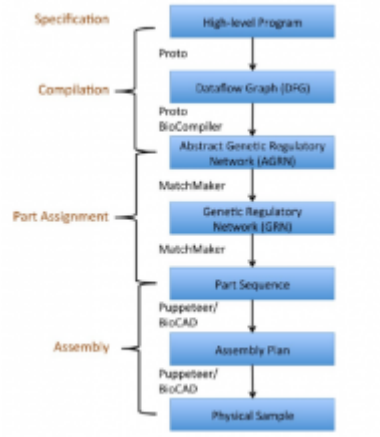
An End-to-End Workflow for Engineering of Biological Networks from High-Level Specifications
ACS Synthetic Biology
July 10, 2012, pp. 317-331

External Links:

Weiss Lab
http://groups.csail.mit.edu/synbio/

Image Gallery:

255 Main Street, room NE 18-501
Cambridge, MA 02142-1601
Phone: 617-253-6966 Fax: 617-258-6790
http://tlo.mit.edu
Contact the Technology Manager: tlo-inquiries@mit.edu



255 Main Street, room NE 18-501
Cambridge, MA 02142-1601
Phone: 617-253-6966 Fax: 617-258-6790
http://tlo.mit.edu
Contact the Technology Manager: tlo-inquiries@mit.edu