Setting Standards in Synthetic Biology

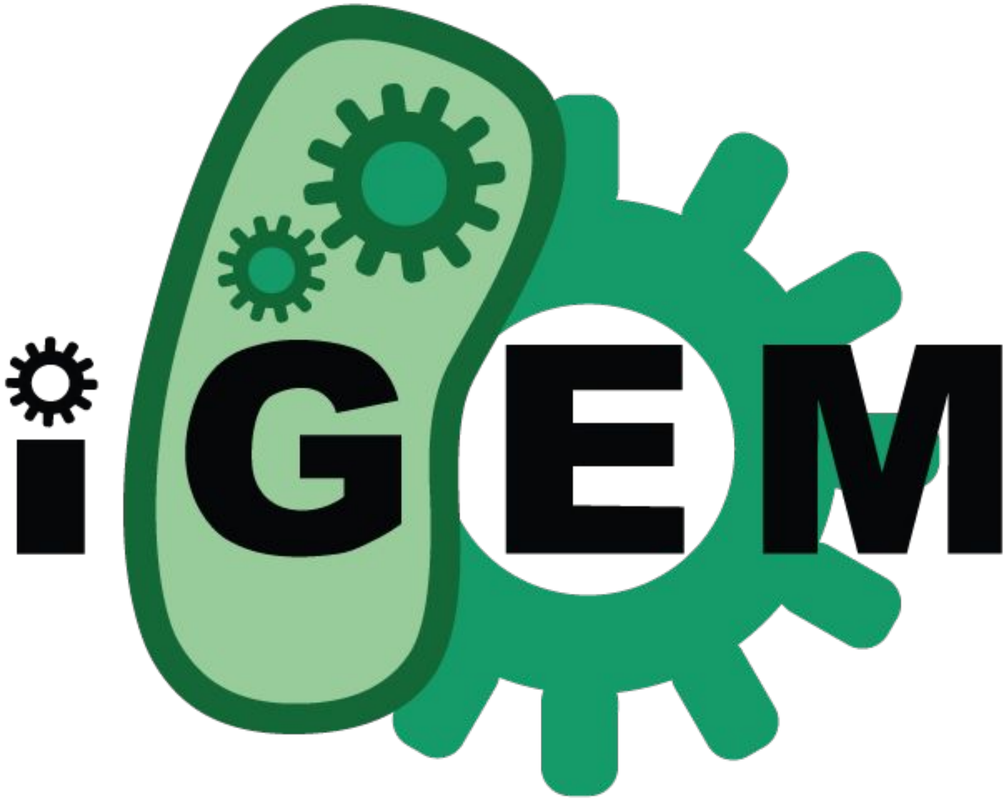
Traci Haddock-Angelli, Ph.D.

Director of the Competition

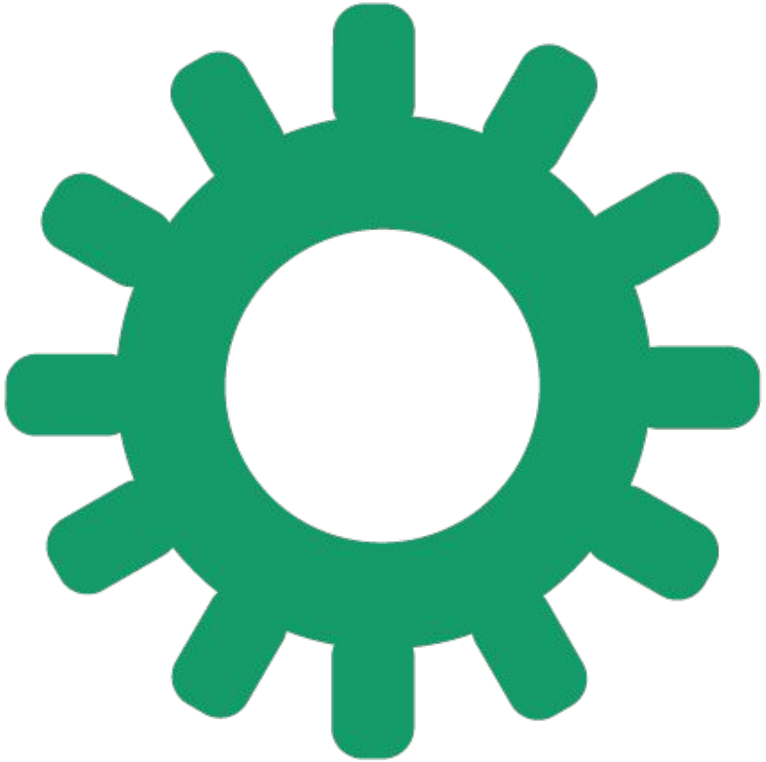
iGEM Foundation

2018.igem.org

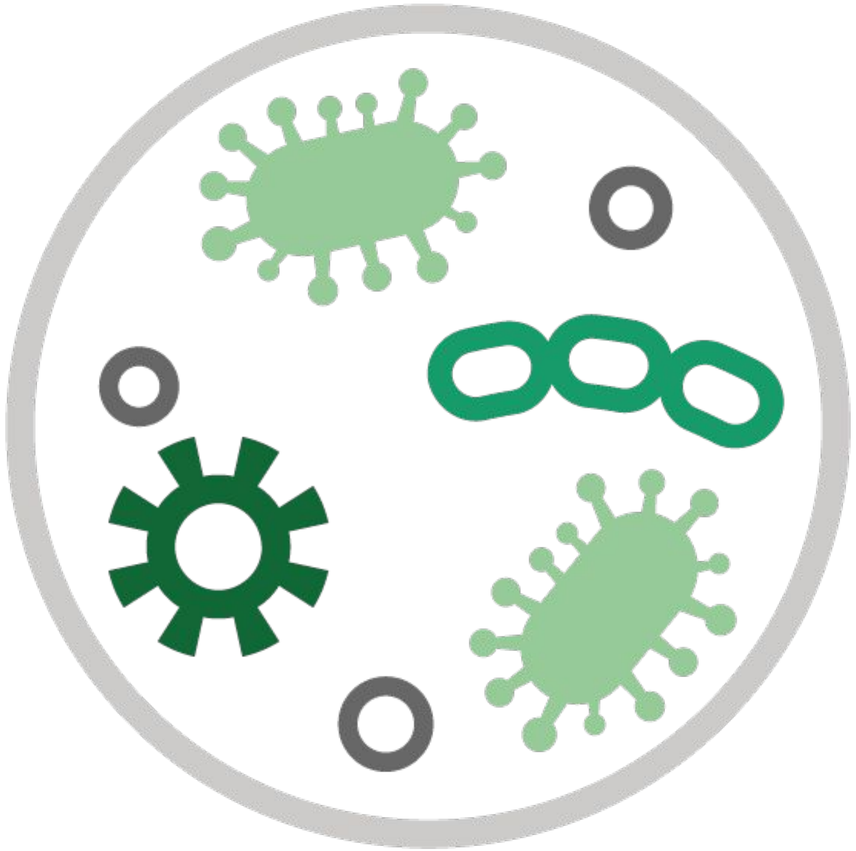
traci@igem.org



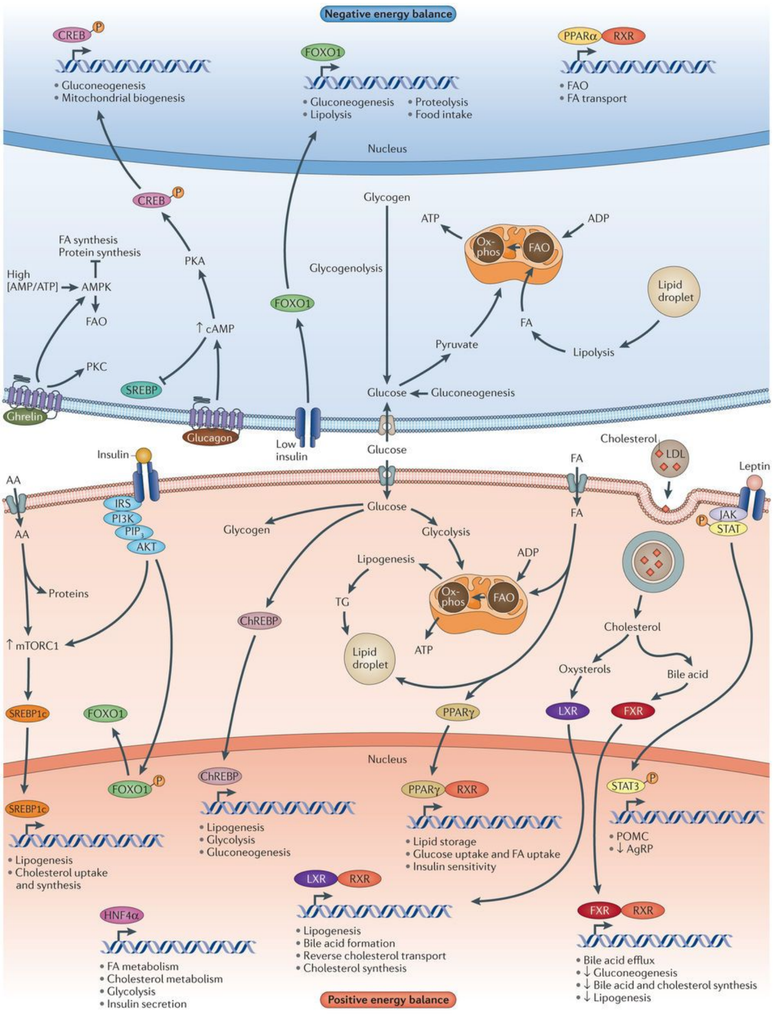
What is iGEM?

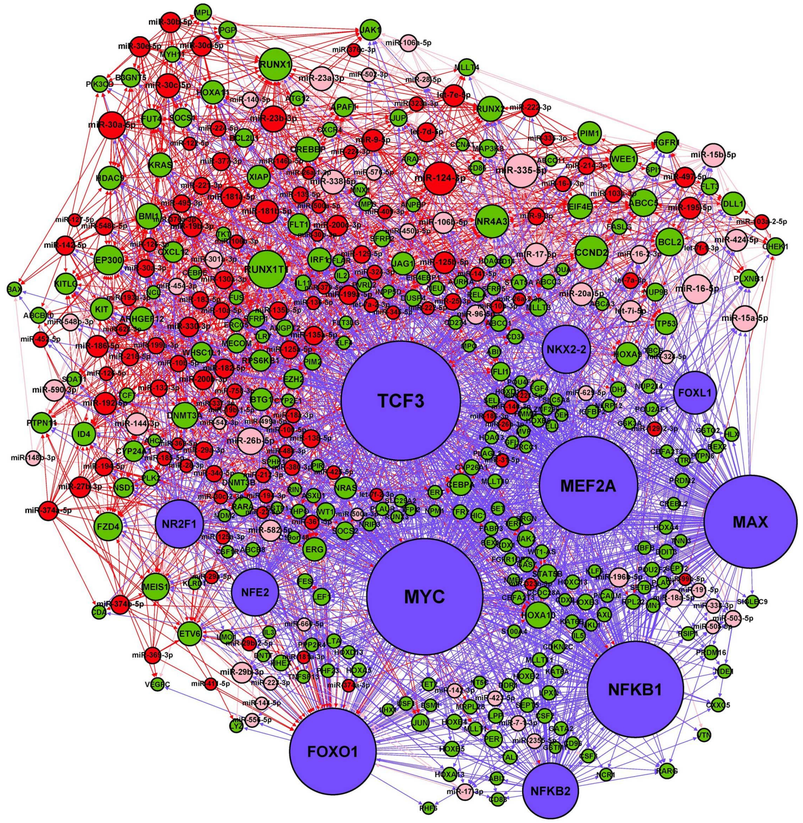


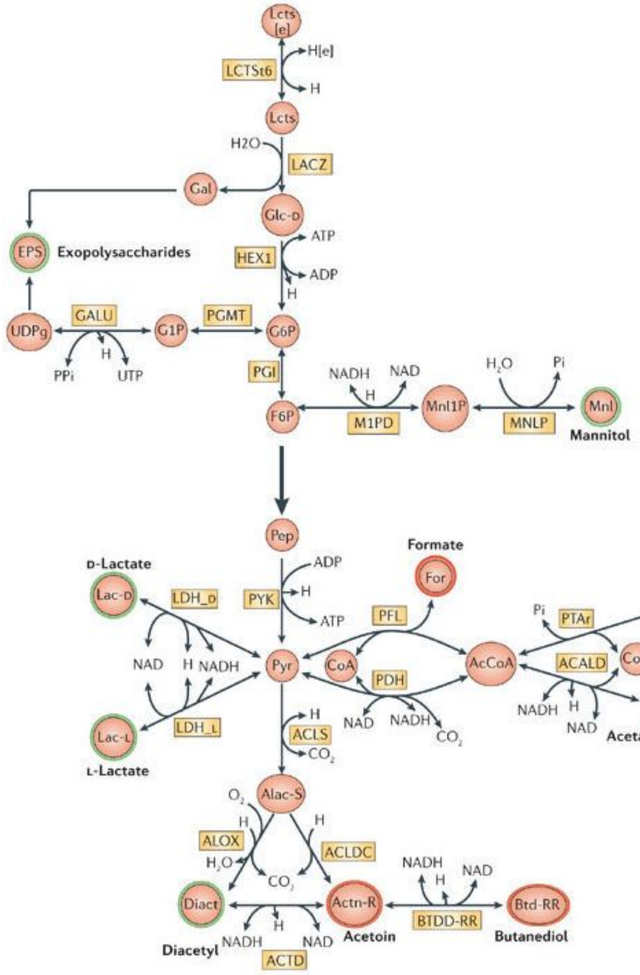
INTERNATIONAL GENETICALLY ENGINEERED MACHINE



WHAT IS SYNTHETIC BIOLOGY?



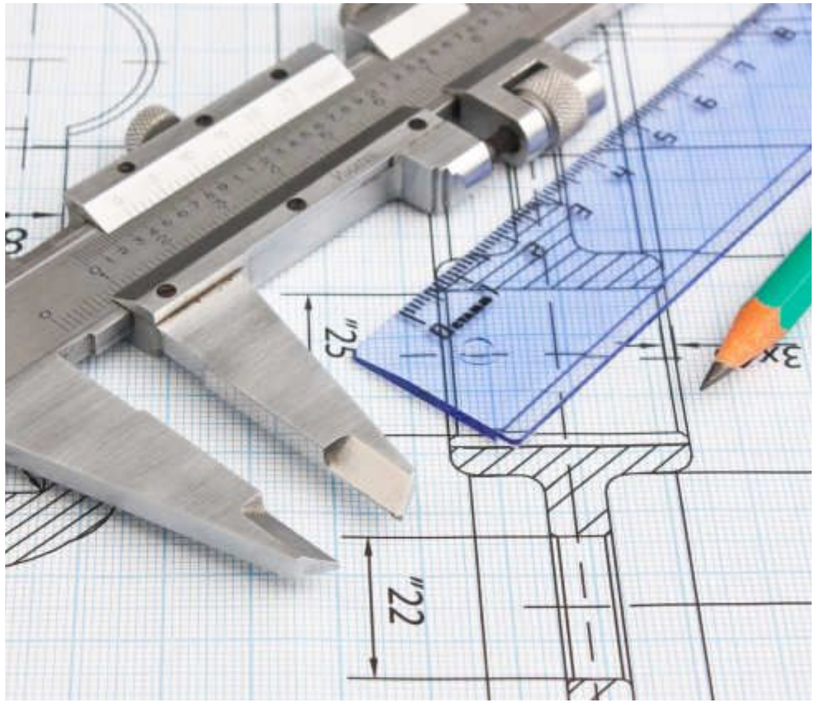


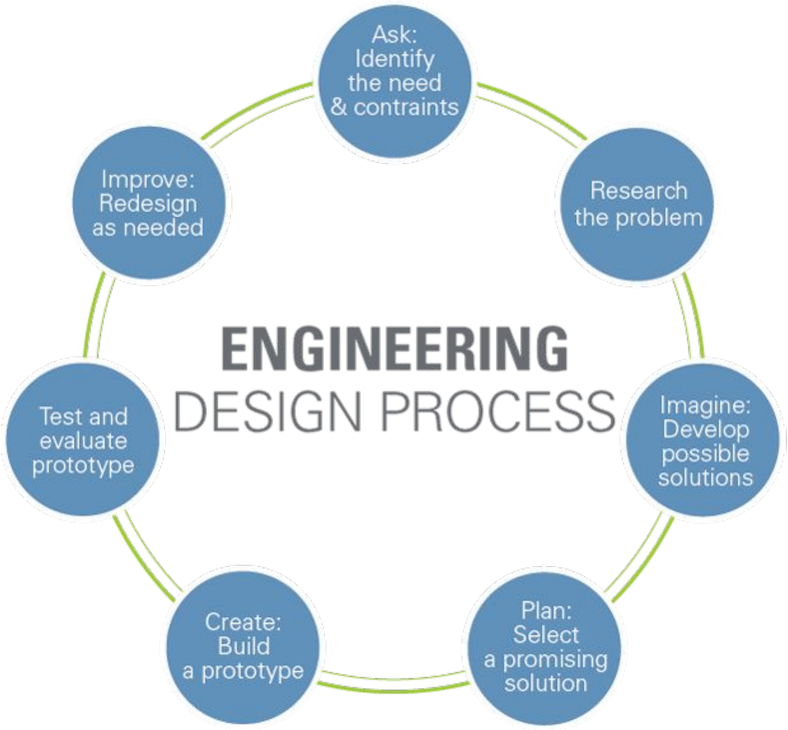


Nature Reviews | Microbiology

BIOLOGY

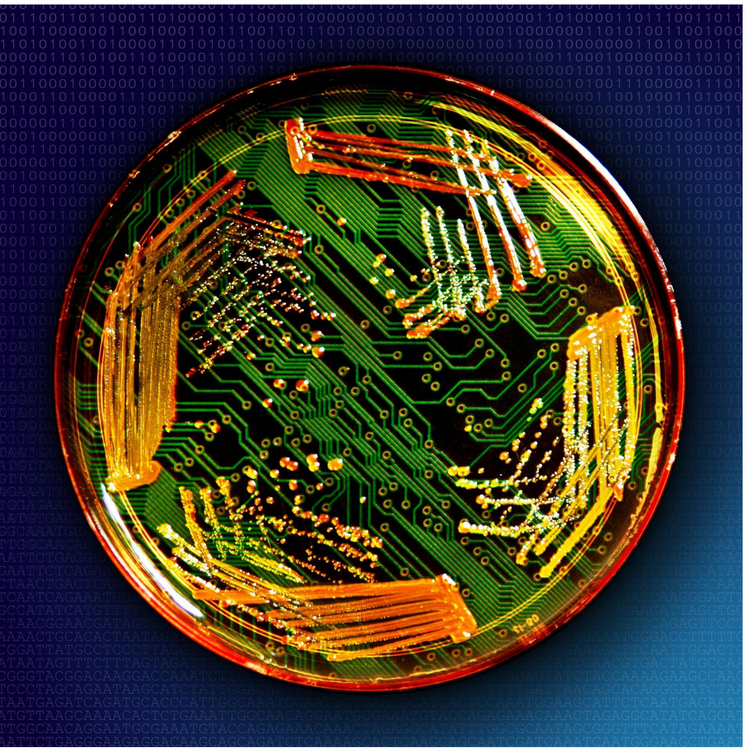
IS VERY COMPLEX!

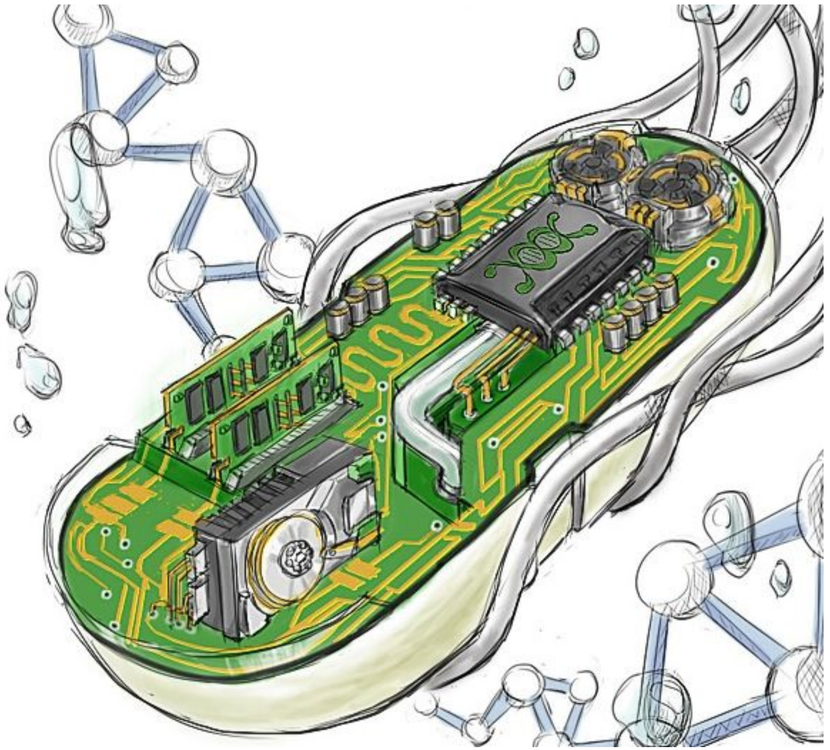


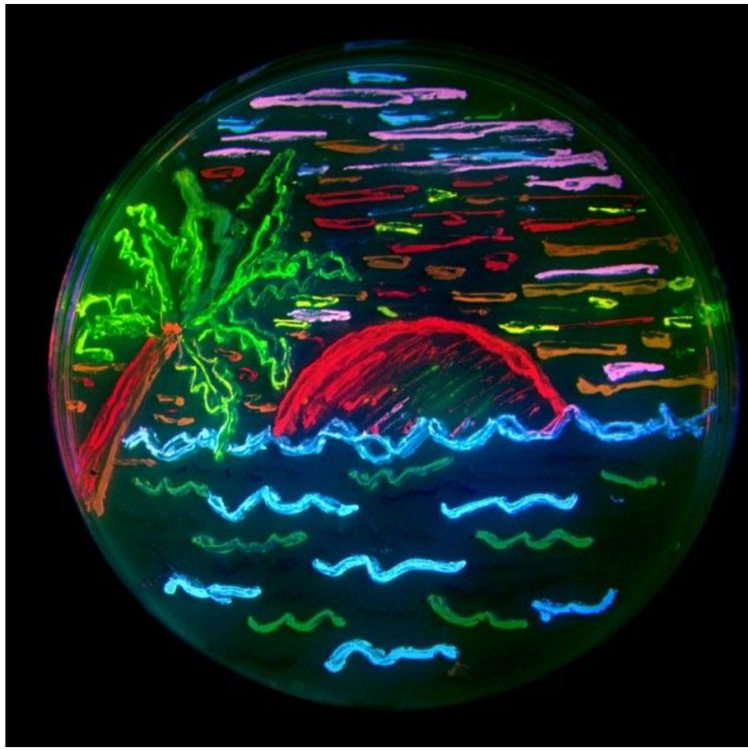


ENGINEERING

SIMPLIFY AND BUILD

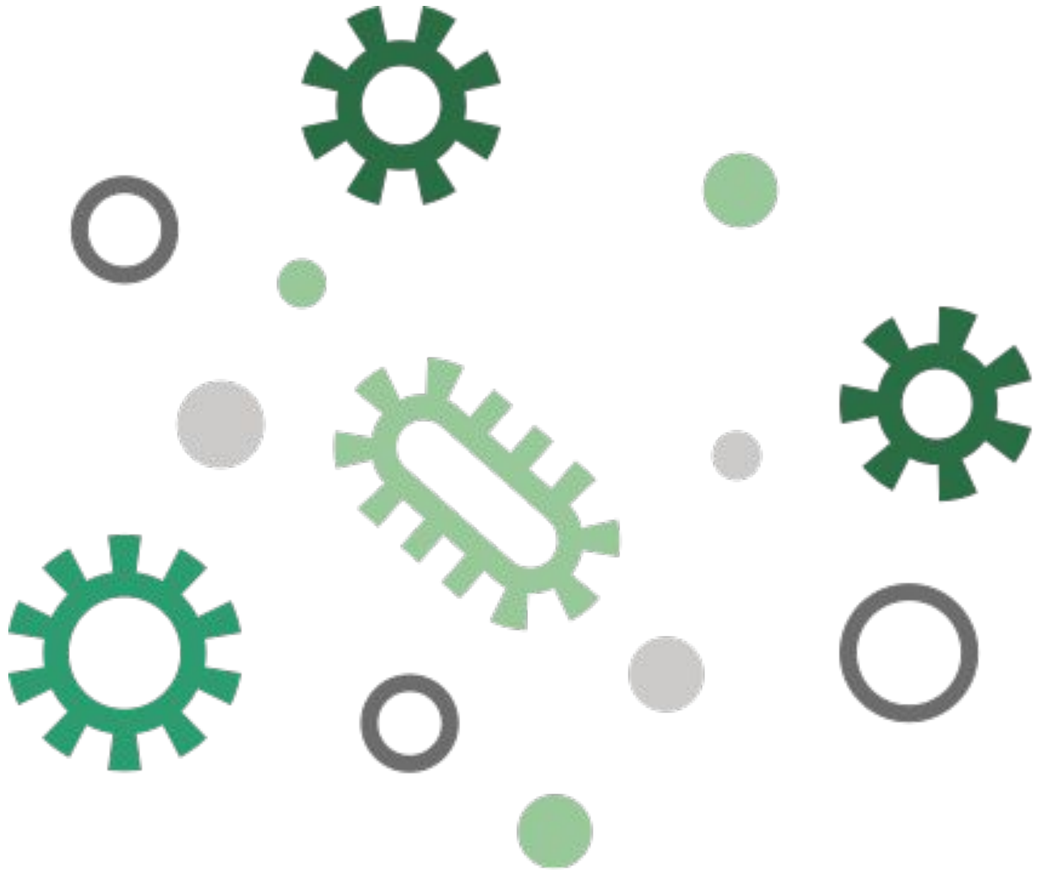






ENGINEERING + BIOLOGY

SYNTHETIC BIOLOGY

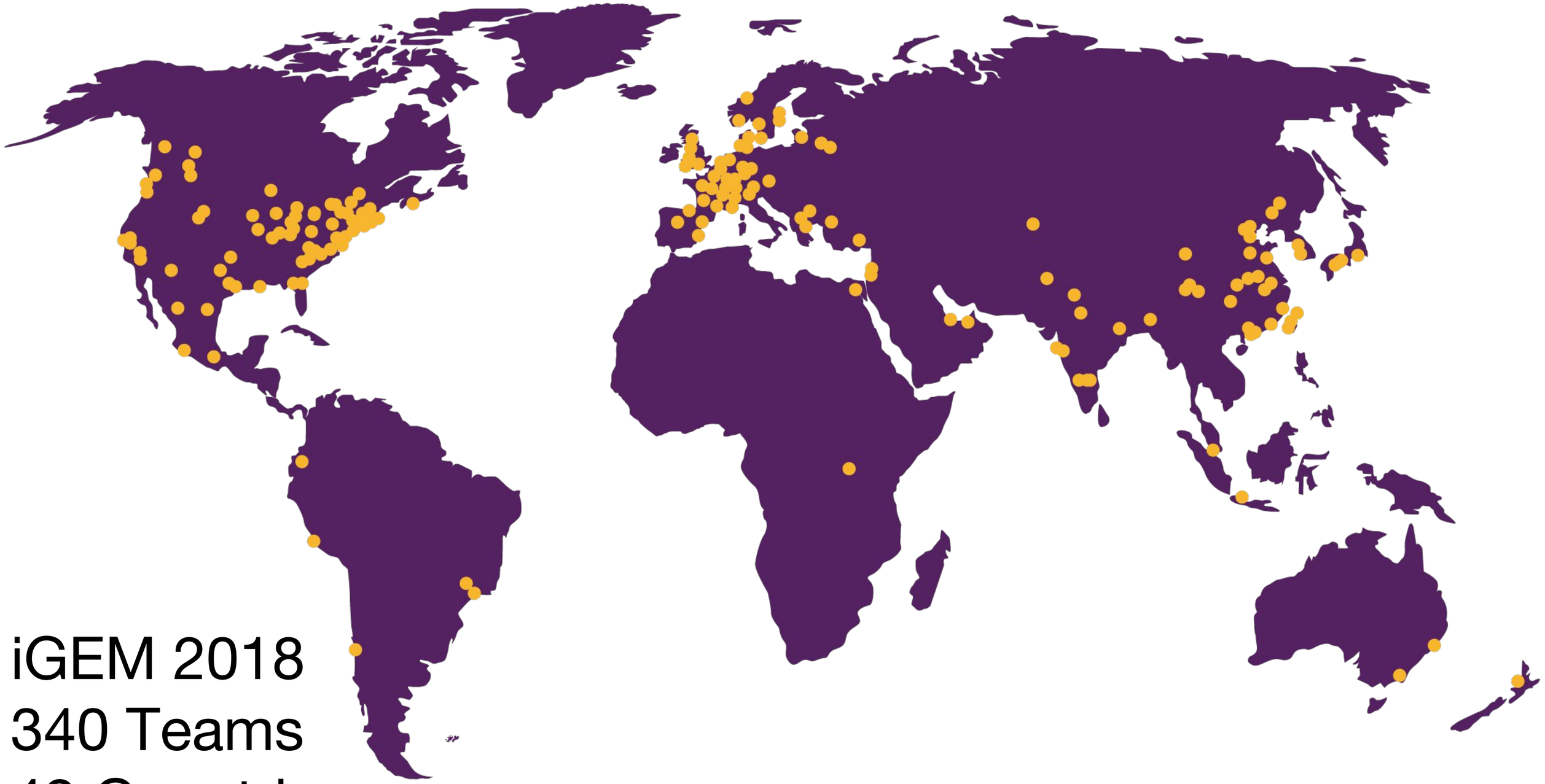


USING DNA
TO PROGRAM CELLS

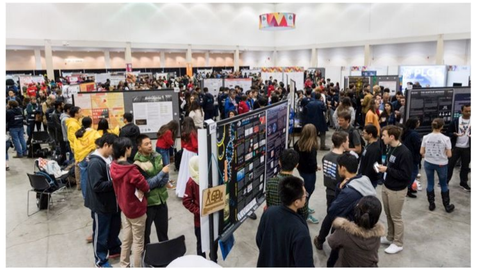

MIT
MIT

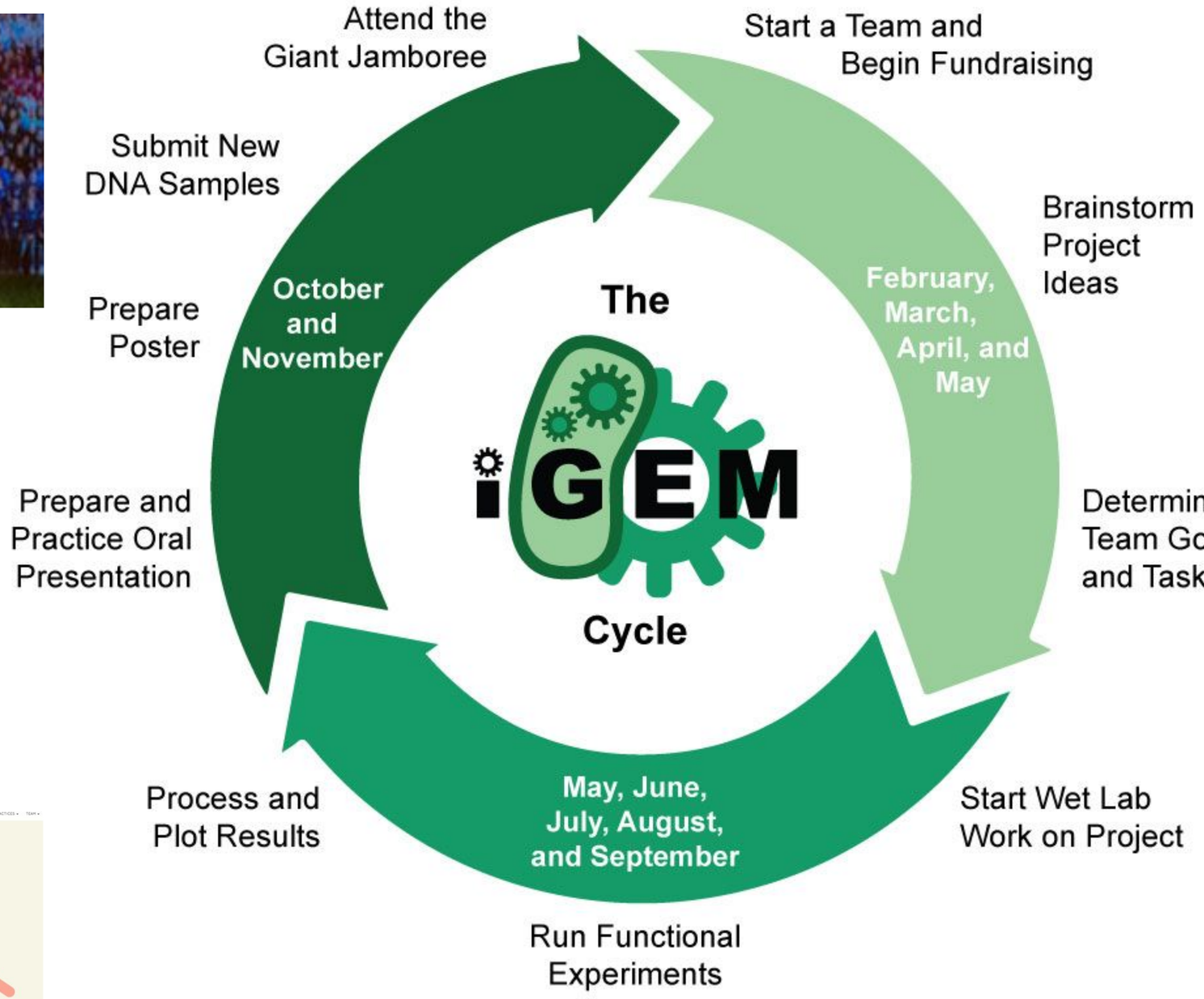
iGEM 2004
5 teams
1 country

iGEM 2017
310 teams
44 countries


iGEM 2018
340 Teams
42 Countries
2018.igem.org

Giant Jamboree



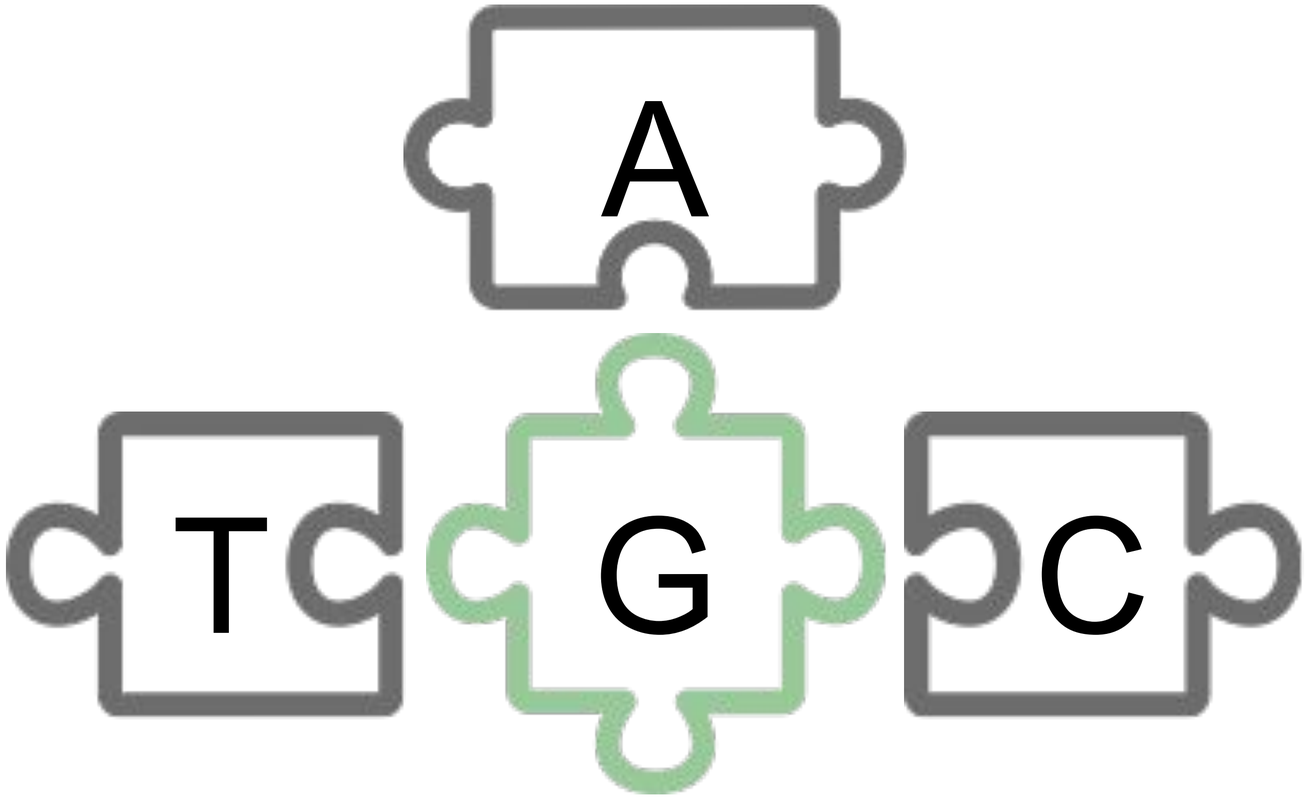


Teams receive DNA Distribution Kits

Work in the Lab


Meagan Lizarazo
iGEM Foundation
1 Kendall Square - Suite B6104
Cambridge, MA 02139
USA
Linearized Plasmid Backbones
iGEM
STOP!
DO NOT FREEZE
Storage instructions inside
iGEM 2017
DNA Distribution
Kit Plate 1
Kit Plate 2
Kit Plate 3
Kit Plate 4
Kit Plate 5
Kit Plate 6
Kit Plate 7
Competent Cell Test Kit
Measurement Kit
WELCOME !
iGEM
2017

Technology



BIOBRICKS

STANDARDIZED DNA SEQUENCES

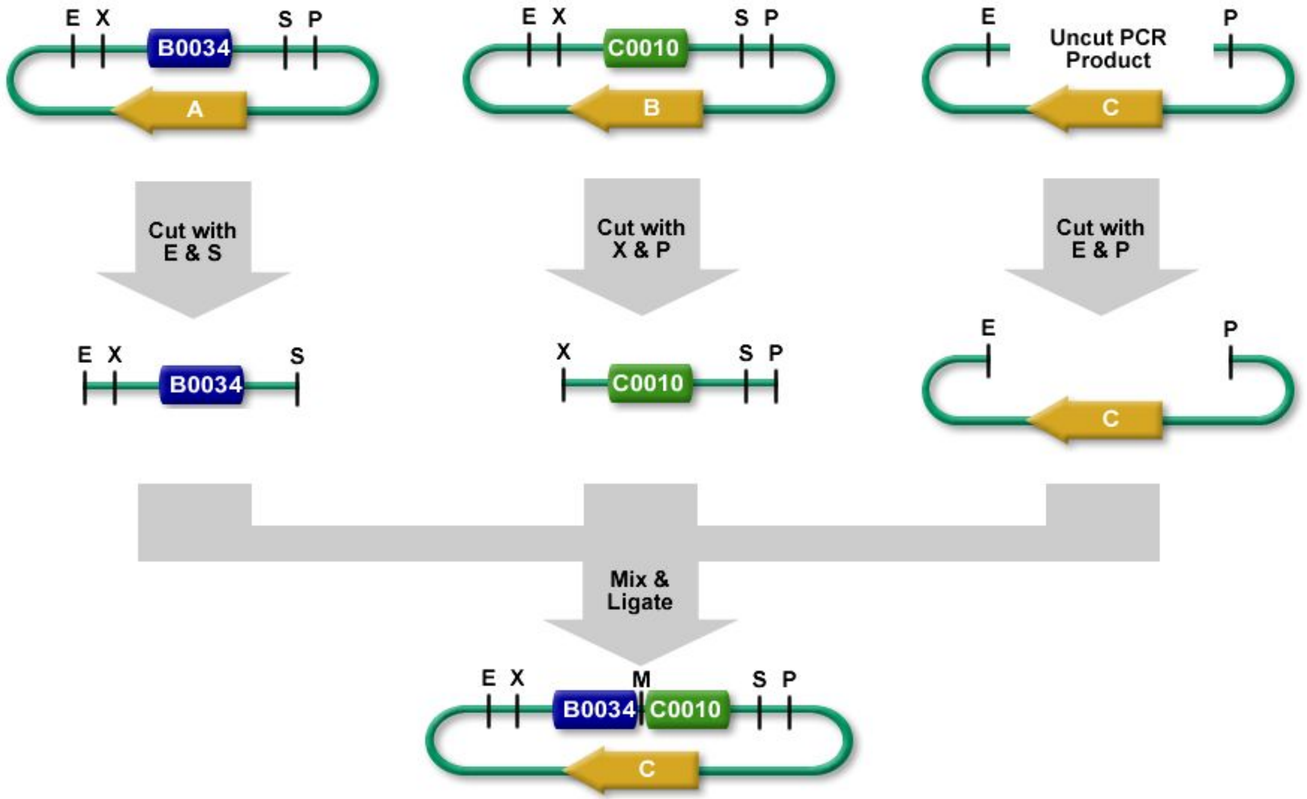
BioBrick Standard

Developed by Tom Knight in 2003

Very easy to use

Restriction based assembly

Four enzymes in the method



Registry of Standard Biological Parts

parts.igem.org

tools catalog repository assembly protocols help search BBa_

Add and Document Parts

Start adding and documenting your parts now! Your parts should be well characterized and measured, and follow the Registry's requirements.

Sample Submission

iGEM Teams must complete a submission form and ship their part samples by **October 10, 11:59PM EDT**. Follow the Registry's requirements for part submission, and don't forget to include a tracking number!

Registry Updates

The Registry will be undergoing updates (some major, some minor) over the next few months. If you notice any issues with functionality, please let us know at **hq (at) igem (dot) org**.

Request for Comments (RFC)

– Physical assembly standard
– Reference standard
– Functional composition standard
– Data exchange formats

The BioBricks Foundation:RFC

RFCs for Physical Assembly Standards

10, 21, 23, 24, 25, 26, 28, 37, 39, 45,
47, 53, 54, 61, 65, 69, 75, 81, 88, 92,
94, 98, 104, 106, 109, 110, 111, 113
…and counting!

BioBrick Assembly is RFC10

https://openwetware.org/wiki/The_BioBricks_Foundation:RFC

Part:BBa_B0034

RBS

Designed by: Vinay S Mahajan, Voichita D. Marinescu, Brian Chow, Alexander D Wissner-Gross and Peter Carr IAP, 2003. Group: Antiquity (2003-01-31)

Released HQ 2013
Sample In stock
1 Registry Star
4572 Uses
10 Twins
Get This Part

RBS (Elowitz 1999) -- defines RBS efficiency

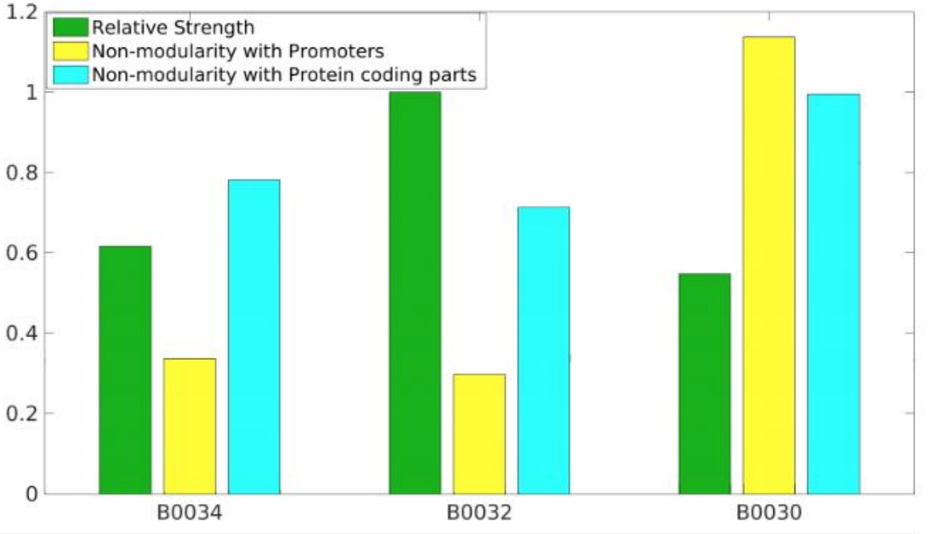
RBS based on Elowitz repressilator.

Usage and Biology [edit]

IIT Madras 2016's Characterization [edit]

Modelling [edit]

Global non-modularity towards promoters & protein coding parts and relative strength was estimated for RBSs B0030, B0032, B0034 in our modelling

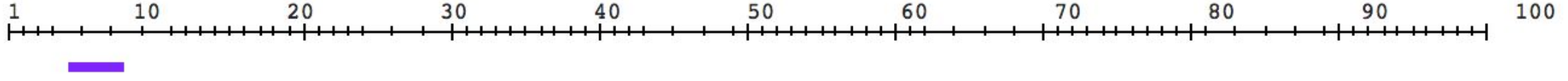


Experimentation [edit]

Biobrick RBSs B0030, B0031, B0032, B0034 were used in our 'Noise in Device' experiment to understand the role of RBS parts in giving rise to noise.

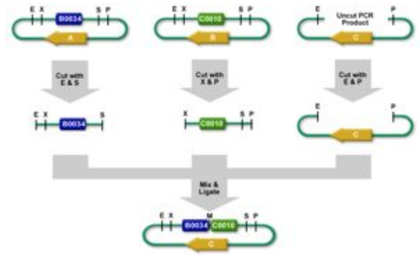
Sequence and Features

Subparts | **Ruler** | SS | DS **Length: 12 bp** View plasmid Get part sequence.

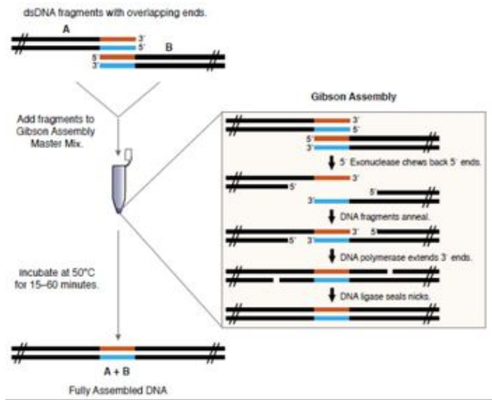


Assembly Compatibility: 10 12 21 23 25 1000

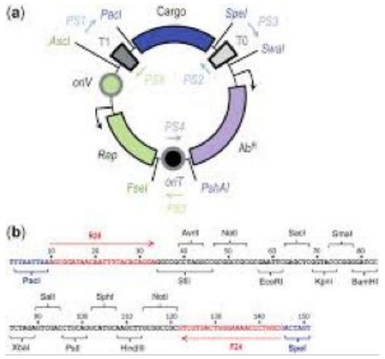
Snapshot of DNA Assembly



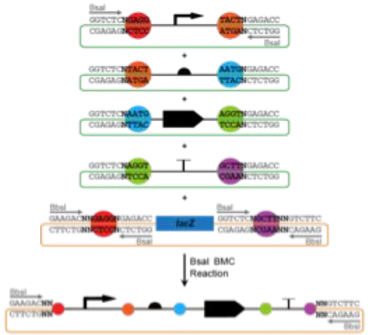
3A BioBrick



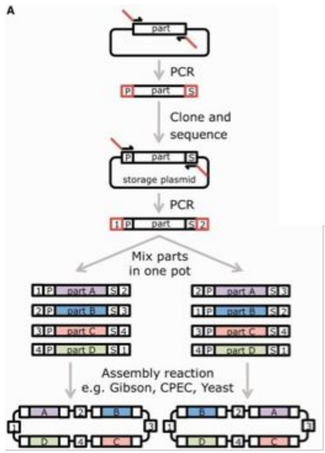
Gibson



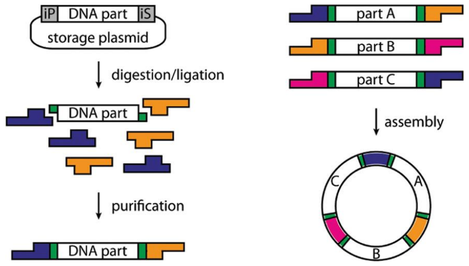
SEVA



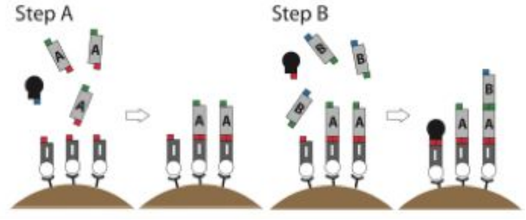
MoClo



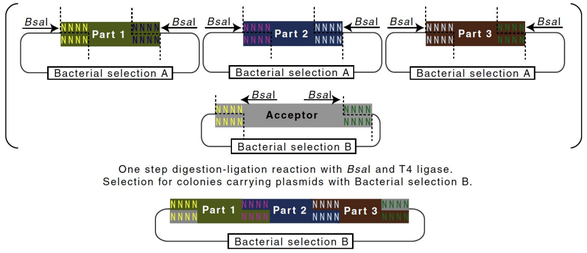
MODAL



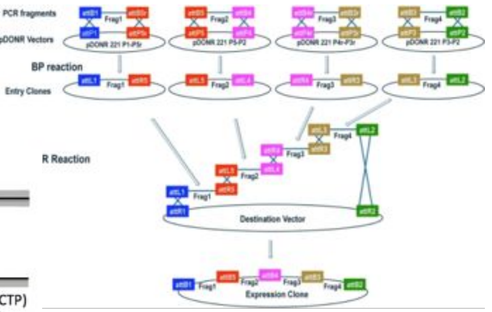
BASIC



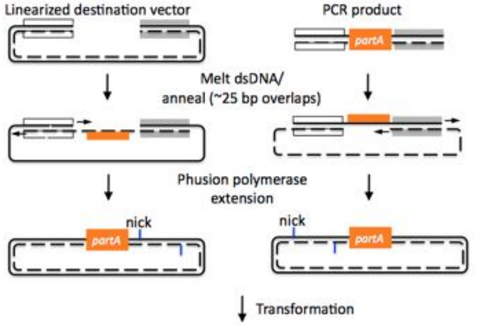
ICA



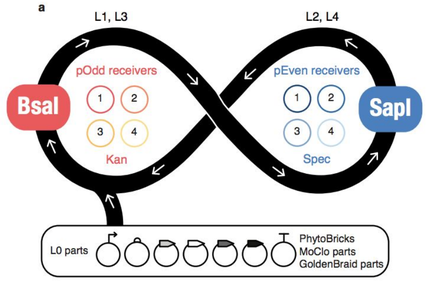
PhytoBricks



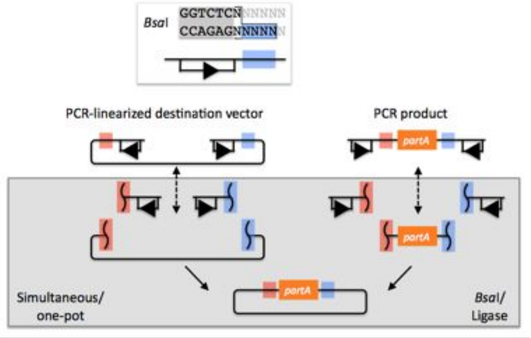
Gateway



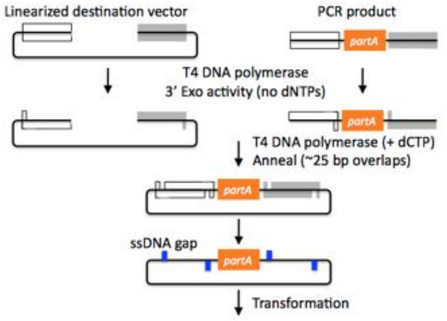
CPEC



Loop



Golden Gate



SLIC

Adopting a New Standard in iGEM

- Can all teams easily use the assembly method?
- Is the system reliable? Does it “just work”?
- Will parts be assembled in order?
- Can multiple parts be assembled at the same time?
- Will it work in multiple chassis?
- Has the system been widely adopted?

Bacteria: *E. coli* and *Bacillus subtilis*

Plants: *Marchantia polymorpha* and *Nicotiana benthamiana*

Yeast: *Saccharomyces cerevisiae*

Mammalian: HEK293, HeLa, and CHO

Type IIS Assemblies

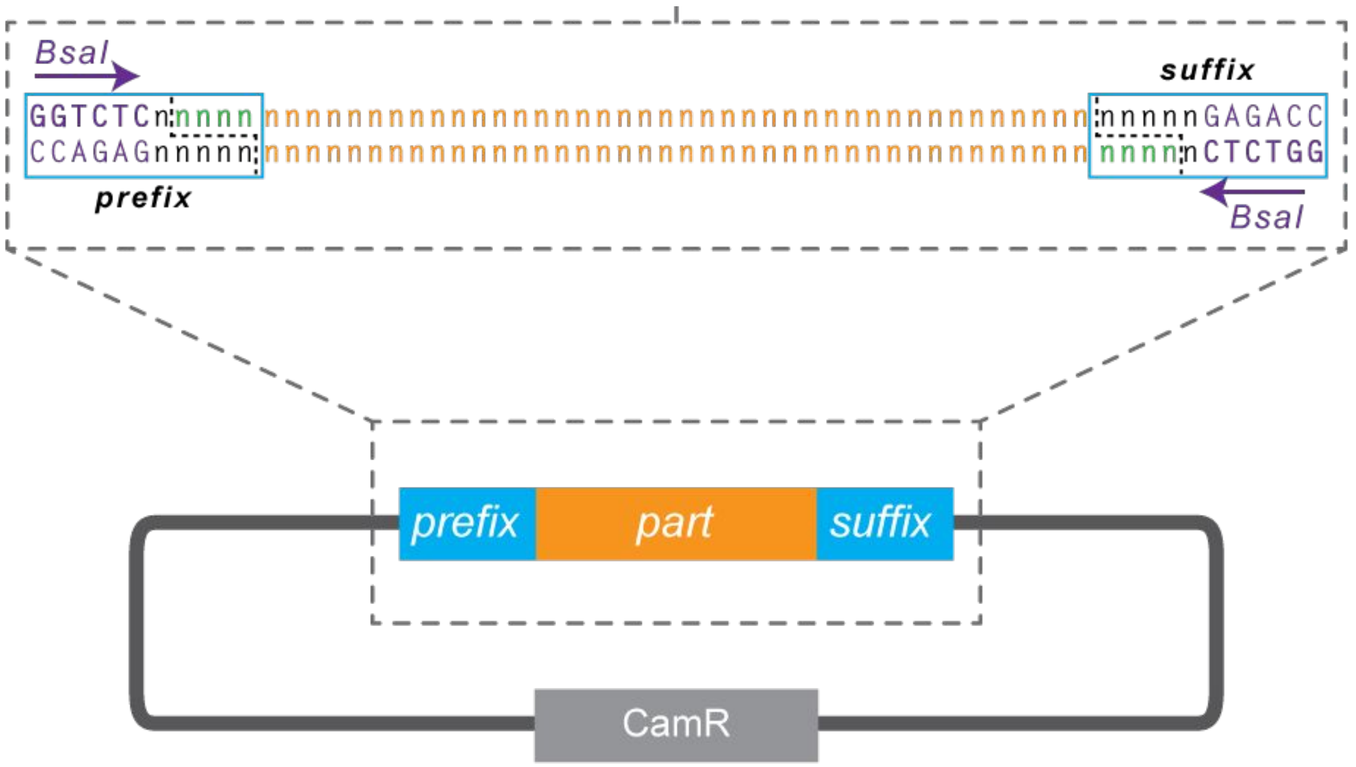
Type IIS enzymes cut outside of their recognition sites

User-defined scars

Sequential ordering of parts

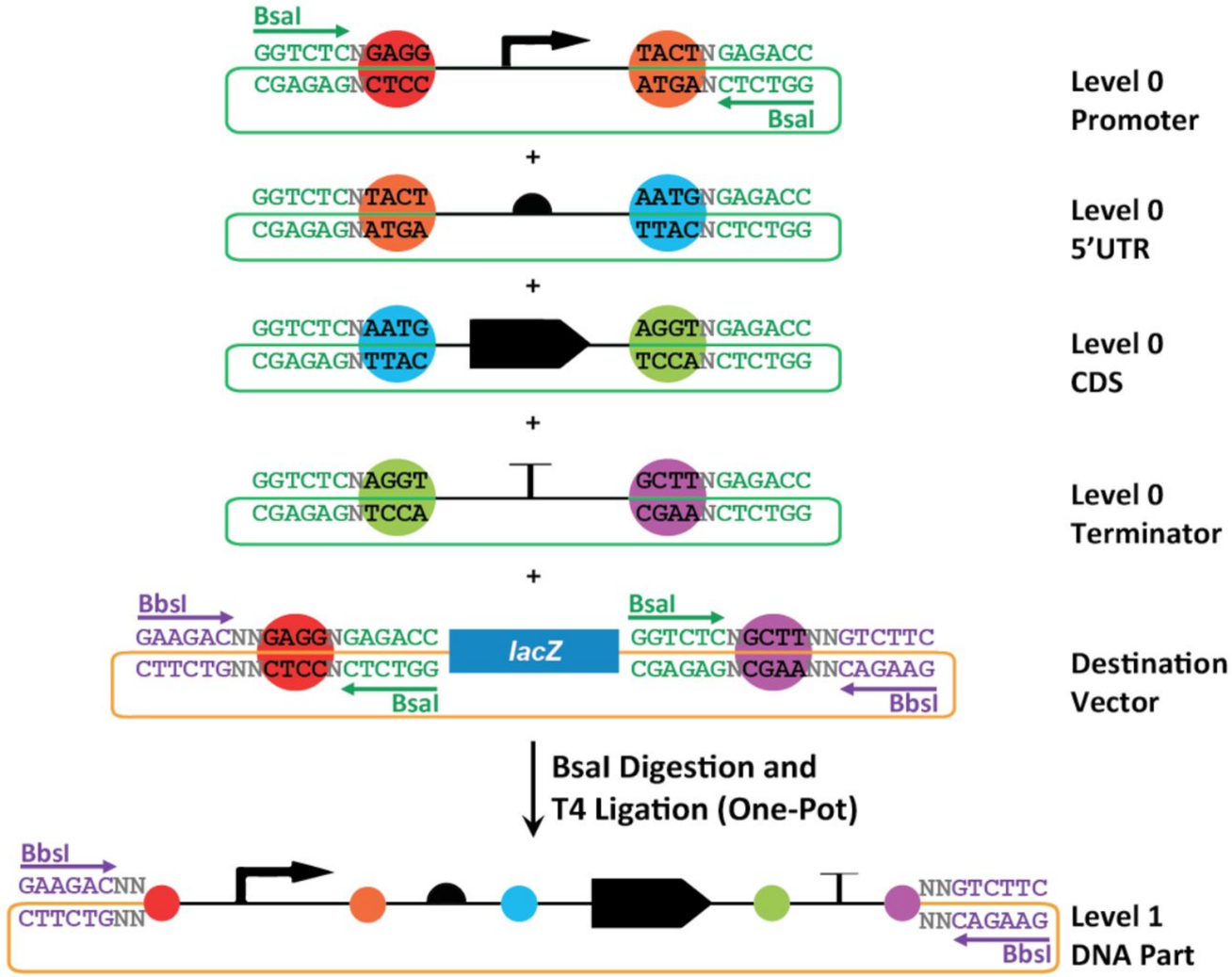
Fewer enzymes

Library generation is easier



RFC94: MoClo

2012-2014 Boston University iGEM teams developed MoClo library for *E. coli*



RFC106: PhytoBricks

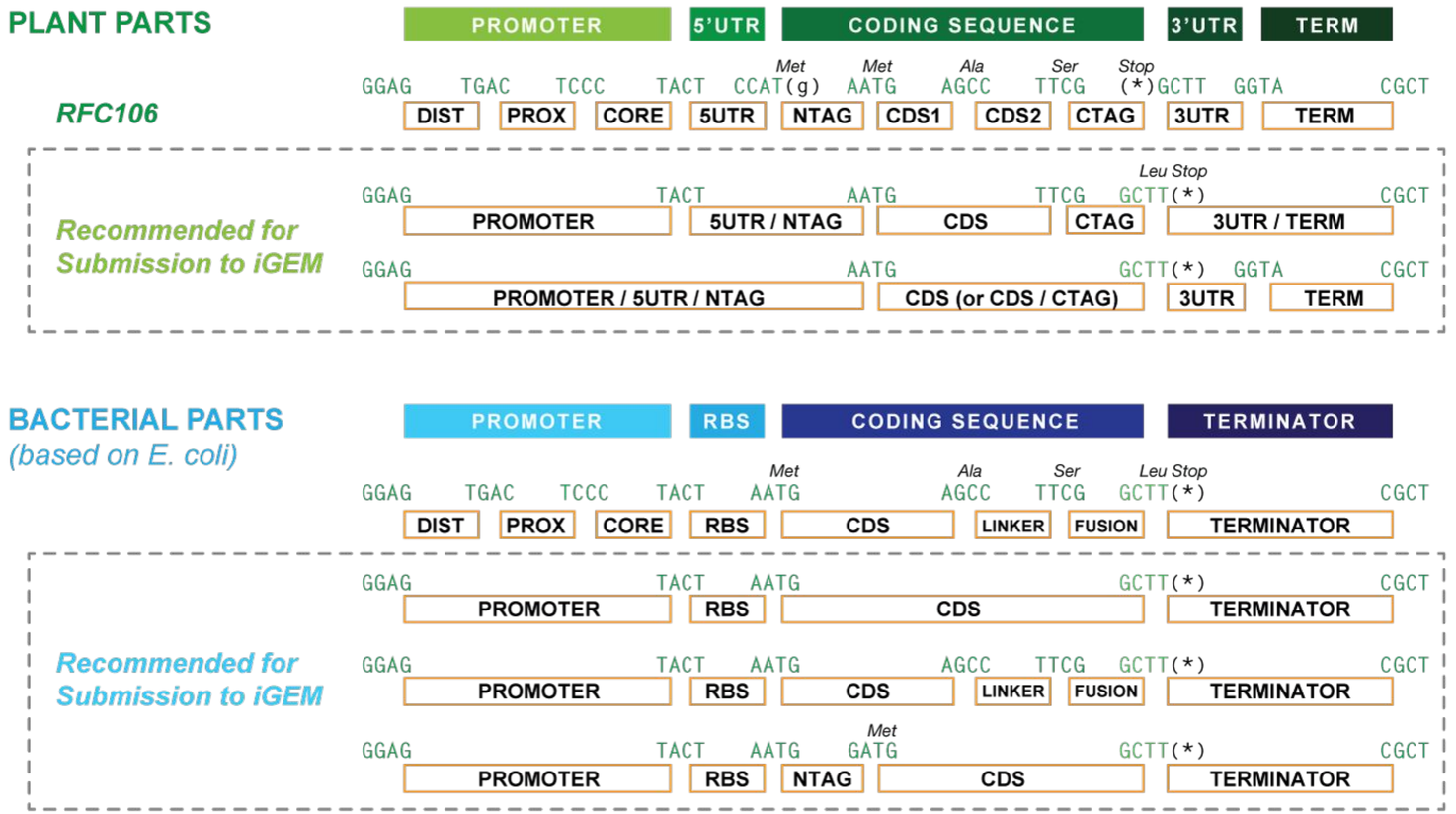
Combined effort from three 2014 iGEM teams (Cambridge-JIC, Valencia UPV, and NRP-UEA)

Widely adopted by plant syn bio community

Multi-part assembly in one-pot - easy to use

Compatible with other Type IIS systems

Began accepting RFC106 parts from iGEM teams in 2015



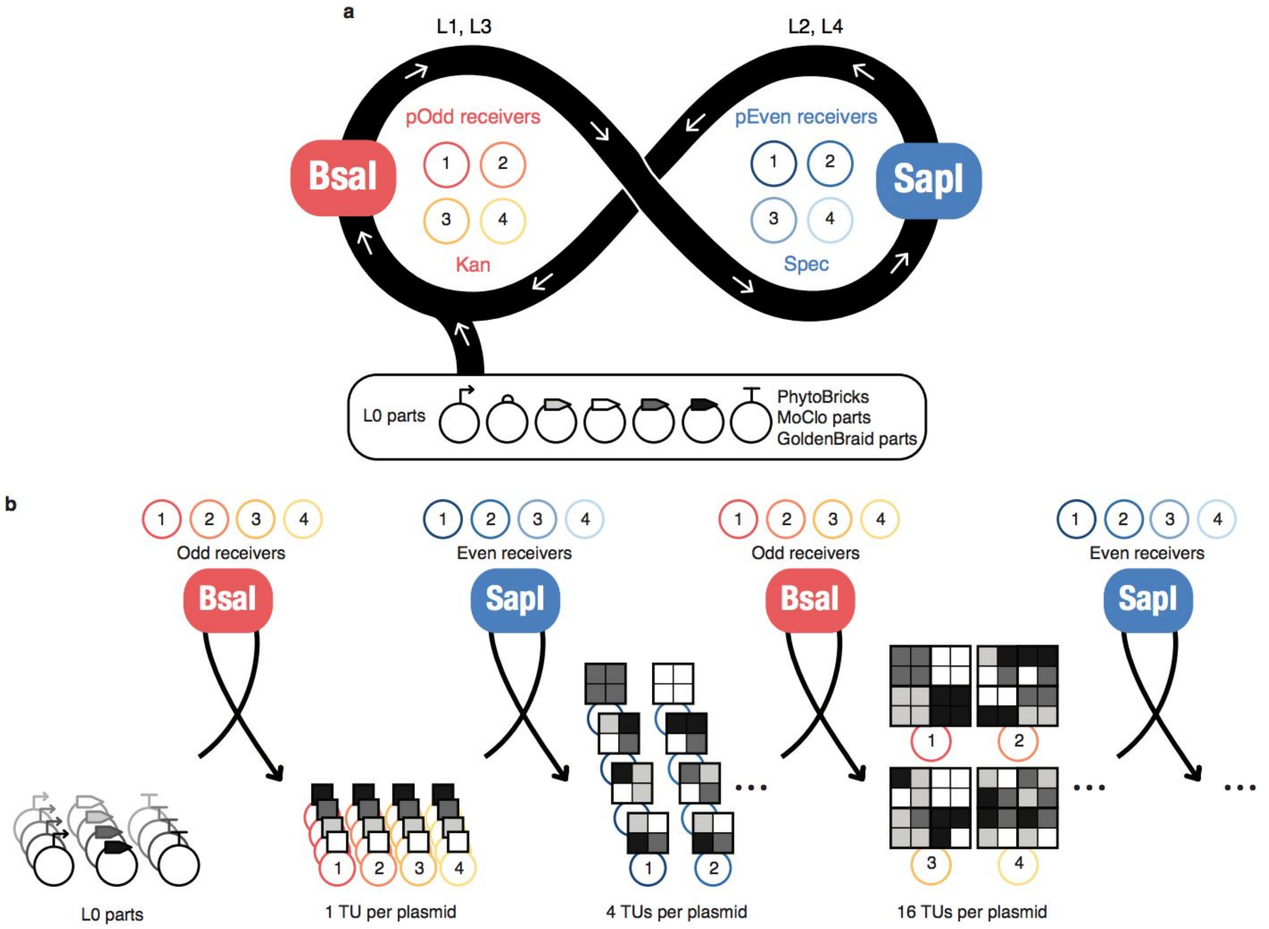
Loop Assembly

Two enzyme system, built from PhytoBricks

Compatible with other Type IIS systems (MoClo, PhytoBricks, GoldenBraid)

Reliable assembly of multiple parts in one reaction

Open source



Pollak et al., 2018; bioRxiv 247593; doi: https://doi.org/10.1101/247593

Measurement in iGEM

InterLab Study
- Development of a standard measurement protocol to measure green fluorescent protein (GFP) in *E. coli* using plate readers

Measurement Committee
- 10 volunteer members
- Develop InterLab
- Support teams
- Write publication

Measurement Hub:
http://2018.igem.org/Measurement

Dr. Jacob Beal
BBN Technologies
Committee Chair

Prof. Natalie Farny
WPI
Committee Co-Chair

Brief History of the iGEM InterLab Study

36 teams — *67 teams* — *92 teams* — *210 teams* — *279 teams*

2014: Year 1 study

Teams were asked to build and measure 3 specific devices

No protocol instruction provided (teams were asked to measure the devices to the best of their ability)

2015: Year 2 study

Teams were asked to build and measure 3 specific devices (2 new, 1 repeat)

Control devices were recommended but not required

Basic protocol is provided

2016: Year 3 study

Teams provided 5 specific devices (same as 2015, plus specific controls)

Detailed protocol is provided

Standard curve material provided

2017: Year 4 study

Teams provided 8 specific devices (same as 2016 , plus 3 new)

Detailed protocol and standard curve material is provided

Specific *E. coli* strain required

2018: Year 5 study

Teams provided 8 specific devices (same as 2016 , plus 3 new)

Detailed protocol and standard curve material is provided

Specific *E. coli* strain required

CFU protocol introduced

2018

GIANT JAMBOREE

Hynes Convention Center
October 24 - 28*
Boston

Questions?

traci@igem.org
@Traci_H_Angelli