I. TEAM: SHENZHEN

Shenzhen is a special consortium. Team members are not from one school, but eight. Students came to Shenzhen and joined a co-cultured program, Genomics Innovation Classes, at BGI College. After lectures introducing synthetic biology and iGEM given by our instructor IC, 22 students from eight schools, including HUST, ZJUT, JESTC, WuH, SNU, SEU, and GuGk, have decided to set up a unified team for iGEM 2012.

II. PROJECT: YAO #1.0

This year, our team focuses on one of the major problems we faced in synthetic biology, interference. When a synthetic pathway is introduced into the cell, it may somehow disturb the original system. On the other hand, the original system may prevent the synthetic pathway from working as expected. To solve this problem, our idea is to construct a real semiautonomous organelle from the natural one, yeast mitochondrion, we call it Yeast Artificial Organelle and YAO for short.

When YAO works as an organelle factory, it inherits some key features from mitochondrion, as membrane structure and semi-self-replication. Its genome and transport systems have to be engineered into artificial ones to meet our needs, as called YAO Genome and YAO Channel.

As Moreover, we also want our YAO to have new properties, YAO Senser, to sense its status, send signals out and respond to signals; YAO Suicider, to trigger suicide pathway for bio-safety purpose.

And finally we want to combine all those components into a YAO Factory to prove that YAO is more safe, stable and effective in bio synthesis.

III. YAO.GENOME

Introduction

YAO Genome is the core part of YAO which is similar to genomes of mitochondria and chloroplasts but ready to be embedded any new system.

Stages to YAO.Genome

1. Homologous recombination
2. Shuttle plasmid
3. Design of YAO.Genome

Result

1. Homologous recombination, positive clones;
2. Homologous recombination, control clones;
3. Shuttle plasmid experiment, positive clones;
4. Shuttle plasmid experiment, control clones.

Top: bright field;
Bottom: under UV.

IV. YAO.CHANNEL

Stages to New Transport Machinery

1. Construction of signal peptide system
2. Installation of new transport machinery
3. Transportation assay

Design

To install TOC on outer membrane & TIC on inner membrane complexes from chloroplast onto mitochondrial membranes, with signal peptides to mitochondrial membranes. Verify signal peptides and TIC & TOC with YFP fusion protein.

Troubles

A frameshift mutation was detected when we sequenced the synthesized composite BioBricks. Due to inaccurate information in PartsRegistry.

V. YAO_SENSOR

Design

In YAO sensor project, we designed a sensor system to monitor NADH/NAD ratio in Yeast mitochondrion. REX protein, which can competitively combine with NADH and NADH are used in our project. When combined with NADH, REX can bind to a specific DNA sequence, known as REX operator, or ROP for short.

While NADH ratio is low, ROP is unbound and GFP can express. While NADH ratio is high, REX binds with ROP, preventing GFP's expression.

Conclusions

1. Design

2. Amazing Features

3. Advantages

4. Simulation

5. Production of IPR at different concentrations of NADPH.

6. Production of IPR at different concentrations of enzymes.

7. Production of IPR.

8. Mevalonate pathway, which can produce precursors for many terpenoids and terpene-based products such as Artemisin.

VIII. CONCLUSION

Project YAO could been seen as pioneering work in iGEM and synthetic biology. Here are some highlights:

1. When we added some new categories into PartsRegistry, organelles as mitochondria, have opened their access to iGEMers and synthetic biologists.

2. We have taken the initial success, on sending signals in and out organelles, and engineering transcriptional regulation in organelle for the first time.

3. We have been trying to engineer, to measure and to theoretically model a series of signal peptides, related with mitochondria and chloroplasts.

4. We have carried out lots of mathematical simulations, illustrating an exciting blueprint of YAO in bio-synthesis.

IX. SPECIAL THANKS

华大基因学院
华大基因研究院

VII. YAO.FACTORY

Introduction

Our YAO system can be used as a relatively independent compartment for biosynthesis in a yeast cell.

Advantages

1. Less harm to host;
2. Enzymes clustered;
3. Self-replication

Result

1. GAL1 promoter mutation and HLDN modification