Easily made BioBricks

Cloning

Or NOT...

Our Approach

- Rapid
- Reliable
- In Vitro
- Easy to troubleshoot

BrickMason Assembly Protocol: An UnStandard
Easily made BioBricks

Our Approach

- Biblical
- Seabirds
- 18th floor
- Find 'em out in biolevel

Characterization of gene-regulatory functions

A modern educational tool

Mass education

Real education

Real education

Intellectual challenge

Real education
Characterization of gene-regulatory functions

Production of gene-regulatory function

Dual colour detection
Easily made BioBricks

Characterization of gene-regulatory functions

A modern educational tool

Our Approach
- Result
- Feedback
- Support
- Teamwork

Workshop annually funded by Sloan Foundation
A modern educational tool

Mass education

Far reach

Intellectual challenge
The BrickMason Assembly

Step 1: Monomer Amplification

Step 2: Digestion

Step 3: Ligate

Step 4: Digestion

Step 5: Heterodimer amplification

Step 6: Mix and amplify

Step 7: Gel extract
The BrickMason Assembly

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Step 5: Heterodimer
Step 5: Heterodimer amplification

Step 6: Mix and amplify

Step 7: Gel extract
Step 6: Mix and amplify

Step 7: Gel extract
Target construct

Ade2 up  KanMX6  TDH3  Ura3  GFP  Ade2 down
Parallel Assembly

1. Monomers

2. Heterodimers

Final Construct

NEB
2-Log ladder

6 pieces, 4216 bp
1. Monomers
2. Heterodimers
Final Construct

6 pieces, 4216 bp
Transformation into the ADE2 locus

GFP expression in 6 colonies

- ~40,000 nucleotides
- 7 definitive mutations
- 4 at Ura3-GFP junction
- 0.000175 errors / nucleotide
31 red colonies out of 55
to the ADE2 locus

Ade2 up  KanMX6  TDH3  Ura3  GFP  Ade2 down
GFP expression in 6 colonies

- ~40,000 nucleotides
- 7 definitive mutations
- 4 at Ura3-GFP junction
- 0.000175 errors / nucleotide
BrickMason 2.0
The Illegal Construct

Ade2 up → KanMX6 → TDH3 → Trp → GFP → Ade2 down
GFP expression from 16 YBA3m transformants

Fluorescence (AU)

Colony
Transformation into the ADE2 locus

GFP expression in 6 colonies

- ~40,000 nucleotides
- 7 definitive mutations
- 4 at Ura3-GFP junction
- 0.000175 errors / nucleotide
Building and characterizing gene-regulatory functions

A.

B.

Ade2 up  KanMX6  pGal 1/10  Repressor  BFP  Ade2 down

Ade4 up  NatMX  Distal  Proximal  GFP  Ade4 down

Ade2 up  KanMX6  pGal 1/10  Repressor  BFP  VP16  Ade2 down

Ade4 up  NatMX  Distal  Proximal  GFP  Ade4 down
BFP fluorescence for tagged TFs

<table>
<thead>
<tr>
<th>Sample</th>
<th>BFP Fluorescence (AU)</th>
</tr>
</thead>
<tbody>
<tr>
<td>WT</td>
<td>Uninduced</td>
</tr>
<tr>
<td>TetR BFP</td>
<td>2% Gal</td>
</tr>
<tr>
<td>TetR BFP VP16</td>
<td>Uninduced</td>
</tr>
</tbody>
</table>

Legend:
- **Uninduced**
- **2% Gal**
Chimeric promoter: Adh1 distal-Gal1/10 proximal

BBa_K642007

Wildtype

Experimental
Human Practices:
Gears of Evolution
Community page

Team uOttawa - Gears of Evolution game

October 20, 2011

"Team uOttawa has posted a game called Gears of Evolution. It was designed as a fun educational tool to inform high school students and those with a general interest in the sciences about synthetic biology. We also plan to port the game to the iPhone shortly, and possibly incorporate a scoring system based on time. Please give us feedback on what you think of the game!"

uOttawa wiki

Game: Gears of Evolution

Community Bricks

1. Open App Store
2. Search for "Gears of Evolution"
3. Download the app
4. Learn about synthetic biology
5. Play and explore
6. Share with friends
7. Feedback requested
In summary

• Terrific new assembly method

• Dual colour system for characterizing transcription factors and their cognate promoters

• Super awesome fun educational video game
In summary

- Terrific new assembly method
- Dual colour system for characterizing transcription factors and their cognate promoters
• Terrific new assembly method

• Dual colour system for characterizing transcription factors and their cognate promoters

• Super awesome fun educational video game
characterizing transcripts and their cognate promoters

- Super awesome fun educational video game
In summary

• Terrific new assembly method

• Dual colour system for characterizing transcription factors and their cognate promoters

• Super awesome fun educational video game
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uOttawa team