C.i.mager
C.imager
C.imager

- Color mimic
- Motion control

*Caulobacter crescentus*
Color mimic
C. imager

Color mimic

Motion control

Caulobacter crescentus
Motion control
C. imager

- Color mimic
- Motion control

Caulobacter crescentus
Light-inducible circuits

GREEN

RED

BLUE

Evan J Olson, Nature Methods 11, 2014
Results---- pigment

Florescent proteins produced directly without sensing lights
Results---- Blue circuits

[Images of experiment results showing blue circuits under different conditions.]
Results---- Green Circuits
Results---- projecting imaging and presenting image
Problem:
Bacteria may not present the image we project!
Desired Image → RGB Components → Protein output → Light Input
Desired Image → RGB Components → Protein output → Light Input
\[ P = b + \frac{aI^n}{I^n + k^n} \]

\[ I = k \left( \frac{a}{P - b} - 1 \right)^{-\frac{1}{n}} \]

P-protein output
I-light intensity

Evan J Olson, Nature Methods 11, 2014
C. imager

- Color mimic
- Motion control

Caulobacter crescentus
Caulobacter Crescentus
An amazing vector for bacterial photography
Adhesive ability of *Caulobacter crescentus*

<table>
<thead>
<tr>
<th>Name</th>
<th>Shear Strength/$N \cdot mm^{-1}$</th>
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<tbody>
<tr>
<td>SBS Glue</td>
<td>0.8</td>
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<tr>
<td>Rubber with Metal</td>
<td>1.028</td>
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<tr>
<td>AB Glue</td>
<td>8.5</td>
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<tr>
<td>Holdfast of <em>C. crescentus</em></td>
<td>68</td>
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</tbody>
</table>
Regulation: More stalked cell

Special lifecycle

Pamela J B Brown, Advances in microbial physiology. 2009
Validation of adhesive ability of *C. crescentus*
Validation of adhesive ability of \textit{C. crescentus}

\begin{itemize}
\item \textit{C. crescentus}, 1 day growth, 2 min water flow, 400X
\item \textit{E. coli}, 1 day growth, 2 min water flow, 400X
\end{itemize}

Validation of adhesive ability of \textit{C. crescentus}
Motion Control - Flagellum Rotation
Motion Control - Holdfast Biosynthesis

HfiA → HfsJ → Holdfast
Design of Blue *C. imager*
C. Imager simulation

E. coli, 48h

C. crescentus, 48h
Conjugation
Conjugation

F Plasmid (With OriT)

Initial Concentration

Resulting Bacteria

• Fang Teng; Barbara E Murray; George M Weinstock, Plasmid. 1998. 10.1006/plas.1998.1336
ODE Model

\[
\begin{align*}
\frac{dn_1}{dt} &= r_1 \left(1 - \frac{n_1 + n_2 + n_3}{K}\right) - d_1 n_1 \\
\frac{dn_2}{dt} &= r_2 \left(1 - \frac{n_1 + n_2 + n_3}{K}\right) - d_2 n_2 - d_3 n_1 - k n_1 n_2 \\
\frac{dn_3}{dt} &= r_2 \left(1 - \frac{n_1 + n_2 + n_3}{K}\right) - d_2 n_3 + k n_1 n_2
\end{align*}
\]

Concentration

- \( n_1(t) \): E. coli (With Plasmids)
- \( n_2(t) \): C. C. (No Plasmids)
- \( n_3(t) \): C. C. (With Plasmids)

Birth, Competition, Death, Conjugation
Parameters From Experiments

\[ r_1, r_2, d_1, d_2, d_3, k, K \]

- \( r \) – birth
- \( d \) – death
- \( K \) – capacity
- \( k \) – conjugation

<table>
<thead>
<tr>
<th></th>
<th>( r_1 )</th>
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<th>( d_1 )</th>
<th>( d_2 )</th>
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<table>
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<th>( k )</th>
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<td>0.5</td>
<td>1.5</td>
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</table>
Results

\[ n \]

\[ \frac{nt}{h} \]

\[ 100 \]

\[ 60 \]

\[ 24 \]

\[ 60 \]

\[ 100 \]

\[ t/h \]
Results in Phase Space
1. **Whatever** the initial concentration, C.c. (With plasmids) will dominate the system finally.
2. Putting more C.c. than E. coli initially will accelerate the dominance.
Results of Conjugation

Red *C.imager*  

Blue *C.imager*
RNA Logic Gates

To make parts work faster and more accurate
Hammerhead Ribozyme

Advantages

Fast
Efficient
Less Cross Talk

Robert Penchovsky. Computational design and biosensor applications of small molecule-sensing allosteric ribozymes. Biomacromolecules. 2013. 10.1021/bm400299a
Safety

Kill Switch
Safety Design: Kill Switch

LPS

LALF

Nutrition
IPTG $\rightarrow$ LALF(Lac I Anti-LPS Factor)
Result

Amount of *E. coli* decreases
Concentration of IPTG increases

Growth of *E. coli* in gradient IPTG medium
Summary

The work we have done
Summary

Color Mimic

- Constructed RGB light inducible circuits.
- Tested the photographic system with projector.
- Calibrated chromatic aberration by modeling.

Motion Control

- Validated the adhesiveness of *C. crescentus*.
- Extracted motion control parts from chromosomal DNA: DgrA/DgrB and HfiA.
- Constructed standard parts for motion control (Flagellum Control & holdfast biosynthesis).
Summary

Conjugation

- Found optimal initial concentrations in conjugation by modeling.
- Developed the protocol of conjugation between *C. crescentus* and *S17-1 E.coli*.
- Constructed parts for conjugation.
- Validated the effectiveness of adhesiveness in raising the resolution of images by modeling.

Safety

- Constructed kill-switch parts and validated its functionality.
Parts Construction

Blue and red light sensing-imaging system
- BBa_K1363400
- BBa_K1363401

Motion control
- BBa_K1363000
- BBa_K1363002
- BBa_K1363001
- BBa_K1363005

Conjugation parts from *E.coli* S17-1 to other bacteria
- BBa_K1363003
- BBa_K1363004
- BBa_K1363006

Kill switch based on LALF regulation
- BBa_K1363200
- BBa_K1363201

RNA Logic Gates

- BBa_K1363603
- BBa_K1363604
- BBa_K1363605
- BBa_K1363601
- BBa_K1363600
- BBa_K1363500
- BBa_K1363610
- BBa_K1363611
- BBa_K1363606
- BBa_K1363607
- BBa_K1363609
- BBa_K1363610
- BBa_K1363602
Future Work

Accomplishment of *C.imager*

- Light-induced color mixture tests
- Light-induced chromatic pattern tests

RNA logic circuit improvement

- Introducing RNA logic gates in the imaging system to improve its accuracy and efficiency.
Policy & Practice

C. imager, not only in lab, but in public.
Making ambers & Rubbing plants
Caulobacter Crescentus
Multiplex Color Display

2014 USTC-China

Our team, founded in 2007, joined the iGEM competition as one of the earliest teams in China and we got the 2nd runner up and fundamental advance project that year.

Nowadays, we still spare no time trying to make our project come true and propagate the biological knowledge to public.

Our Project
- Figure out the optimum conditions of C. Crescentus development, transformation and expression.
- Develop the holdfast in C. Crescentus.
- Extract the gene regulating the expression of holdfast in C. Crescentus and build parts with the regulating system.
- Construct the light sensing-response system.
- Guide the accomplished system into E. coli and C. Crescentus.
- Pre-test the function and efficiency of ribozymes.
- Plan the industrialization and further development in biological imaging industry.
- Design the projector specialized for biological imaging.

Questions it raises:
- Do you wanna try photographic film produced by bacteria?
- What do you care most when using this product?
- How do you like the development of other field’s technology into biological system?
- Will you believe the future when everything we use and require is produced by organism?

stark@mail.ustc.edu.cn

This year, our team attempt to combine the photographic film with biological system triggered by much careful investigation of bacteria imaging study and light inducible response both in recent research and iGEM team work.

We would like to call the system as Fast and Accurate Biological Chromatic Sensing-Imaging System (FABCSIS) by respectively joint red-green-blue (RGB) light sensing proteins with RGB fluorescent proteins into a new pedestal Caulobacter Crescentus, a kind of bacterium possessing adhesive holdfast which we can utilize for more stable expression in distinct surroundings like the water flow. And we are determinant to improve the system with ribozymes regulation to enhance the accuracy and speed of perception and response. At last, we will construct a customized projector to measure the efficiency of system and make a plan for the possibility of industrialization in the future. The FABCSIS, as we wish, will realize the chromatic imaging system firstly held in the media, regarding each individual as a pixel just like photographing. We believe our work will drive the further study and development of imaging system in other living organisms and one day biological photographic films will appear in everybody’s daily life to change the world for its lowcost and high resolution.

This year, we attempt to improve the technology in biological system and hope our job could influence the development and study of biological imaging chromatically. Besides, we seized many opportunities to introducing genetic engineering and synthetic biology to students who are intrigued in science, parents and people who are worried about GMO without any biological conception. We hope one day in China, everyone could make a decision scientifically with knowledge and judgement with our tiny efforts.
Outreach

Nanjing

Taiwan

Peking

China
Lab open day
Team
Jiong Hong  Haiyan Liu  Zhi Liang

Advisors
Sponsors
C.imager, view colorfully

Thanks for Listening 😊