Synthetic Biology Applications for Space Exploration

Brown-Stanford iGEM 2011
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Energy Production
Harvest methane from the Martian surface, alcohol fermentation from photosynthetically produced sugars, and microbial hydrogen production.

Cellular Factories
Produce a wide range of biological products—such as drugs, nutrients, enzymes, and reagents—from a DNA database and an interchangeable cellular chassis.

In situ resource use for colony assembly
Minimize upmass of construction materials by mineralizing existing regolith.
• **Goal**
  Develop a universal, sustainable energy source to power biological tools

• **Application**
  Feed the biological tools that will transform raw materials into fuel, food, drugs, and other products useful to settlers
The Premise

• All organisms need a set of essential elements (CHONPS)
• Our goal:
Anabaena PCC7120

- Photosynthetic
- Freshwater
  - Sucrose as osmotic buffer
  - Inducible sucrose production with salt stress
- Nitrogen fixing
  - Products diffuse into other cells

N₂ → Nitrogenous products → Other cells
Sucrose secretion

- We focused on sugar secretion
- CscB sucrose permease
- Why sucrose?
  - Un-metabolized

Salt induced sucrose production
Sucrose utilization

• Can other organisms survive on generated sucrose?

• Testing growth with *E. coli* W on minimal media + sucrose
Construct Design

- CscB under control of psaC promoter
  - psaC is protein in photosystem I
  - isolated upstream region (~400bp)
  - expresses CscB only in cells that photosynthesize
- GFP reporter to verify localization

- Full PowerCell construct (BBa_K656012)
DNA Transformation

- Restriction enzymes digest foreign DNA taken up by cell
- We need to protect our plasmid...
Tri-Parental Mating

- Cargo plasmid pRL25
- \textit{E. coli} DH5aMCR with helper plasmid pRL623
- \textit{E. coli} ED8654 with conjugation plasmid pRL443
- \textit{Anabaena} target cell
Results of triparental mating
Selection Cultures for Anabaena

Day 3

Day 7

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RegoBricks

Goal:
Modularize the well studied process of biocementation.

Application:
Reliable, protective habitats for any extraterrestrial settlement, which are not limited by payload capabilities

Why *Sporosarcina pasteurii*?
• precipitates calcite by hydrolyzing urea, which is currently a waste product in space missions
• a spore forming bacterium, allowing populations to survive extreme environmental conditions
Biocementation

- Urease enzyme cleaves urea into ammonia and carbon dioxide
- Ammonia raises the pH causing the formation of divalent carbonate anions
- These anions meet with calcium cations that are in solution to form calcium carbonate crystals
Protocol - Urease BioBrick

Isolated from pBU11- recombinant *E. coli* created containing urease genes from *S. pasteurii*

- PCR amplification
- Restriction digest
- Ligation into pSB1C3
Results & Characterization

- Urease operon successfully transformed into *E. coli*, submitted as part BBa_K656013 in the registry
- Verification of part functionality on urease test plates; phenol red signal indicates increases in pH as a result of ureolysis
- Future directions
**Going to (near) Space**

**Goal:**
Send balloon into near-space; Stratosphere (24km-34km) mimics conditions found on Mars

Temperature
Pressure
UV radiation

**Application:**
Gain insight into our organisms’ potential for space utilization
Lab Recap

- Generated DNA and submitted to Registry of Standard Biological Parts:
  - *Anabaena* pSac vegetative promoter (BBa_656010)
  - CscB sucrose permease from *E. coli* W (BBa_656011)
  - Full *Anabaena* PowerCell construct (BBa_656012)
  - *Sporosarcina* Urease Operon (BBa_656013)

- Characterized 3 of the newly submitted parts

- Environmental tolerance testing in upper atmosphere
Human Practices

• What is the motive to explore and colonize space?

• What can engineered biology do for space exploration?

• How should we go about expanding onto other planets?

• What is the value of alien life?
Outreach
Outreach

Mission Statement

The mission statement of this website can be described as a simple, concise tale of a journey. It is a tale of a journey that involves the exploration of new horizons, the pursuit of knowledge, and the sharing of experiences. It is a tale of a journey that is open to all, regardless of age, background, or experience. It is a tale of a journey that is meant to inspire and to challenge. It is a tale of a journey that is meant to be shared.

Baking a Community Cake

According to social media trends, the recipe for making a successful cake involves the following steps:

1. Add social media to the cake.
2. Bake it in a community oven.
3. Serve it to everyone.

HOW TO JOIN AlumniIGEM

1) Sign up for the mailing list: AlumniIGEM@igem.org!
2) Find us on Facebook.
Outreach

CommunityBricks is an attempt to change the way that we do outreach and address human practices in iGEM. By creating an open source community of lesson plans and activity protocols, we hope to stimulate deeper, more engaging experiences for both sides of the exchange. Used in this way, you will find tools to present synthetic biology to those who have never heard of it, plans and presentations to give motivating recruitment talks at high schools, and thoughtful considerations about ethics and the rules in should play a role here.

We are tired of lying to mediocre learners and calling it "Outreach". We hope that you try, too. Let's use a new mindset for how we do outreach, something that we will be proud of. This is a lot of fun, so we're creating an account at good prices for educators and learners, let us seek to match that same quality in every field we do.

Granted, building a legacy site or organizing an activity is more work than just tying up a, but harnessing community support for synthetic biology may actually make our less success in the long run. So, if you start thinking about your results, outreach plans, talk to the members to evaluate the growing content on this site to say whether you're really doing. Try it out, and tell everyone else how it went. And if you have ideas of your own, post your stellar outreach plans and share them with everyone!
Outreach

(Allumr GEM) → Community Bricks → Maker Faire → BBC

Brown-Stanford iGEM
Acknowledgements

**PowerCell**
- Dr. Daniel Ducat, Dr. Jeffrey Way, and Dr. Pamela Silver (Harvard University) for initial guidance on our work with sugar secretion and inspiring our use of cscB
- Dr. Manuel Varela (Eastern New Mexico University) for cscB gene, Dr. James Golden (UC San Diego) for cyanobacterial strains, and Dr. Peter Wolk (Michigan State University) for helper plasmid pRL623
- Dr. Jeffrey Elhai (Virginia Commonwealth University) for guidance with transformation of Anabaena

**RegoBricks**
- Dr. Sookie Bang (South Dakota School of Mining and Technology) for advice in working with S. pasteurii
- Ginger Dosier for assistance with biocementation
- Dr. Christopher Mason (Cornell University) for assistance with sequencing of the urease cassette
- Yale 2011 iGEM Team for BBa_6562001, BBa_6562001

**Flight experiments**
- Jack Cackler (King Abdullah University of Science and Technology) for assistance with balloon launches

For a complete list of acknowledgements, please see our team wiki!
Acknowledgements

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André Burnier¹, Evan Clark², Julius Ho¹, Ryan Kent², Lei Ma¹, Eli Moss¹, Jesse Palmer², Max Song¹, Jovian Yu¹
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Advisors:
Lynn J. Rothschild (Adjunct Professor, Molecular Biology, Cell Biology, and Biochemistry at Brown University; Consulting Professor, Human Biology at Stanford University)
Gary Wessel (Molecular Biology, Cell Biology, and Biochemistry at Brown University)

Special thanks to our mentors Norman Wang (University of Hawaii), Kosuke Fujishima (NASA Ames Research Center), John Cumbers (NASA Ames Research Center), Mike Grace (NASA Ames Research Center), Thomas Beer (University of Halle, Germany)

Urease test plates
Martian conditions

<table>
<thead>
<tr>
<th>Mars</th>
<th>Earth</th>
</tr>
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<tbody>
<tr>
<td>6.36 mb pressure</td>
<td>1014 mb pressure</td>
</tr>
<tr>
<td>Carbon dioxide 95.32%</td>
<td>Nitrogen 78.08%</td>
</tr>
<tr>
<td>Nitrogen 2.70%</td>
<td>Oxygen 20.95%</td>
</tr>
<tr>
<td>Argon 1.60%</td>
<td>Argon 9340 ppm</td>
</tr>
<tr>
<td>Oxygen 0.13%</td>
<td>Carbon dioxide 380 ppm</td>
</tr>
<tr>
<td>Carbon monoxide 0.08%</td>
<td>Neon 18.18 ppm</td>
</tr>
</tbody>
</table>

Daytime Temperature

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Martian dust

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PowerCell design

Niederholtmeyer et al. 2010

Argueta et al. 2004
Brickmaking with pasteurii