USC iGEM 2012

An original composition by:
Megan Bernstein, Rebecca Gao, Stephan Genyk, Rachel Kohan, Ellen Park, Luke Quinto, and Eric Siryj

Conducted by: Sean Curran and Percy Genyk
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M 2012

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E. musici
E. musici
Facilitating communication between bacteria and researchers through song
Communication
Every living thing communicates!
Even bacteria!
E. coli tumble response
University of Southern California
iGEM 2012
E. musici
Flagella
E. Coli Forward Run

Counterclockwise

and clockwise turning leads to tumbling
Tumbles and Runs
cheA
BBa_K842006

MCP
MCP
cheW
cheW
adapter

cheR
cheY

run

cheB

P

P

P
Conductor

Experimental Values
- Rotations per Second
- Bacteria Length

Standardized Values
- Flagella Wavelength
- Number of Flagella
- Scaling Factor

More Accurate Results
- 1) Decrease number of standardized values
- 2) Improve experimental analysis
Tumbling
2-D Rotational Velocity in Bacterial Cells

Measured: \( \frac{\text{rotations}}{\text{second}} \quad \frac{\text{distance}}{\text{rotation}} = \text{circumference} = \pi d \)

rotational velocity = \( \frac{\text{rotations}}{\text{second}} \times \frac{\text{distance}}{\text{rotation}} = \frac{\text{distance}}{\text{second}} \)
2-D Rotational Velocity in Bacterial Cells

\[
\text{Measured: } \frac{\text{rotations}}{\text{second}} \quad \frac{\text{distance}}{\text{rotation}} = \text{circumference} = \pi d
\]

\[
\text{rotational velocity} = \frac{\text{rotations}}{\text{second}} \times \frac{\text{distance}}{\text{rotation}} = \frac{\text{distance}}{\text{second}}
\]

**Experimental Values**
- Rotations per Second
- Bacteria Length

**Standardized Values**
- Flagella Wavelength
- Number of Flagella
- Scaling Factor
>> rotate
rotations per second = [.57 .6 .68 .78 .74]
average length of bacteria (um) = 2
frequencies of the individual e.coli flagellum (Hz)
358.1416 376.9911 427.2566 490.0885 464.9557

average frequency of e.coli flagella movement (Hz)
423.4867

note = G#
standard deviation of frequencies
56.1282
More Accurate Results

Needs
1) Decrease number of standardized values
2) Improve experimental analysis

Improvements
Flagella Staining
1) Experimental wavelength
2) Increased tumbling visualization

fliC gene → GFP gene

fliC-GFP
BBa_K842012
Experimental Design

Western Blots

Time (h): 0 1 2 4 16

No IPTG

+ IPTG

We will test many conditions:
1. NaCl concentration
2. pH levels
3. Temperature

DNA Damage Repair

Can we use genetic strategies to prevent DNA damage?
- Conclusive evidence
- Specific targets
- Environmental
Can we use synthetic biology to override the endogenous tumbling response?
- Constitutive promoters
- Inducible
- Environmental
**Western Blots**

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>4</th>
<th>16</th>
</tr>
</thead>
<tbody>
<tr>
<td>IPTG</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
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</tbody>
</table>

- 6XHIS-CheZ
- 6XHIS-CheY
No IPTG
We will test many conditions
1. NaCl concentration
2. pH levels
3. Temperature
<table>
<thead>
<tr>
<th></th>
<th>0.0 M NaCl</th>
<th>0.5 M NaCl</th>
<th>0.9 M NaCl</th>
</tr>
</thead>
<tbody>
<tr>
<td>RPS (rotations/sec)</td>
<td>0.21</td>
<td>0.80</td>
<td>0.21</td>
</tr>
<tr>
<td>Frequency (Hz)</td>
<td>132</td>
<td>503</td>
<td>132</td>
</tr>
<tr>
<td></td>
<td>0.17</td>
<td>0.83</td>
<td>0.55</td>
</tr>
<tr>
<td></td>
<td>0.48</td>
<td>0.72</td>
<td>0.71</td>
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<tr>
<td></td>
<td>0.71</td>
<td>0.71</td>
<td>0.40</td>
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<td>0.45</td>
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<td>0.54</td>
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<td>302</td>
<td>446</td>
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<td></td>
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<td>339</td>
</tr>
<tr>
<td></td>
<td></td>
<td>446</td>
<td>346</td>
</tr>
<tr>
<td></td>
<td></td>
<td>446</td>
<td>251</td>
</tr>
</tbody>
</table>
CheY
0.0 M NaCl

<table>
<thead>
<tr>
<th>RPS (rotations/sec)</th>
<th>0.57</th>
<th>0.60</th>
<th>0.68</th>
<th>0.78</th>
<th>0.74</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency (Hz)</td>
<td>358</td>
<td>377</td>
<td>427</td>
<td>490</td>
<td>465</td>
</tr>
</tbody>
</table>
0.5 M NaCl

<table>
<thead>
<tr>
<th>RPS (rotations/sec)</th>
<th>0.32</th>
<th>0.40</th>
<th>0.43</th>
<th>0.25</th>
<th>0.35</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency (Hz)</td>
<td>201</td>
<td>251</td>
<td>270</td>
<td>157</td>
<td>220</td>
</tr>
</tbody>
</table>
### 0.9 M NaCl

<table>
<thead>
<tr>
<th>RPS (rotations/sec)</th>
<th>0.31</th>
<th>0.32</th>
<th>0.21</th>
<th>0.25</th>
<th>0.28</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frequency (Hz)</td>
<td>195</td>
<td>201</td>
<td>132</td>
<td>157</td>
<td>176</td>
</tr>
</tbody>
</table>

![Graph showing frequency response in 0.9 M NaCl](image-url)
Music
cheY + cheZ
B = ~247 Hertz
A# = ~233 Hertz
A = ~220 Hertz
Ab = ~208 Hertz
G = ~196 Hertz
Human Practices

"Man on the street"
What is E. coli?
It's a food borne illness, like on TV in vegetables?
-Alex

What is synthetic biology?
Artificial limbs? Regenerative tissue?
-Tony

What is a BioBrick?
It's when you are studying for biology and you are stuck... It's a BioBrick!
-Fay Fay

1. Music Video
2. Man on the Street
3. Documentary Mash-up

Online Comments

"Feels very cool to see a little bit of what goes on in a biology laboratory for research" - Illuminair

'Great work! Thanks for all the efforts to educate people to understand more about synthetic biology. " - jwa wang

"I'm not a science major so this was really informative" - kattx3ie
1. Music Video
2. Man on the Street
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"Man on the street"

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"Great work! Thanks for all the efforts to educate people to understand more about synthetic biology. " - jwa wang

"I'm not a science major so this was really informative" - kattx3le
"I still don't like the idea of "playing God" as they said. It's arrogant for people to think that just because they developed a new technology they can change life however they wish. Humans are just too selfish." - Paulina Le

"I feel as though this project is making light of a potentially dangerous field. There is so much potential for abuse, we need to regulate this." - ukrainianak47
What now?

Flagella!

The World!
Research!
Flagella!
The World!
Special Thanks!

Advisors: Dr. Sean Curran
Percy Genyk
Advisors:
Dr. Sean Curran
Percy Genyk
Our private sponsors
USC Davis School of Gerontology
USC Viterbi School of Engineering
USC Dornsife College of Letters, Arts and Sciences
Questions?