Bac INVADER

Hphl   HpyF10VI
Tasl   Sdul   Alu
aaa  ttg  tgt  ggg  ctc  gtt  ctg  ctt  gga  ctg  aag  cta  tgt  aaa

IGEM WARS AW TEAM

NOT SO LONG AGO, IN A
LAB FAR, FAR AWAY...
A bacterium that will kill the CANCER cells
...and briefly how it works

Invasion

Escape from the endosome

Protein secretion

Apoptosis
Entering the cell using invasin...
...and escapes from endosome using listeriolyisin...
...finally, Bax and p53 expression starts.
Cytoplasm - overview

Induction of apoptosis

secretion

p53

entering mitochondrion
Whole system design

Java-based system Simulation
Gene network

We successfully described the both bistable switches using the set of ordinary differential equations which prove that it works.

\[
\begin{align*}
\frac{\partial [\text{mRNAcl}]}{\partial t} &= pcl \text{max transcription rate} \times K_{m\text{cl}}^{n} \left( \frac{K_{m\text{cl}}^{n}}{K_{m\text{cl}}^{n} + [\text{LacProtein}]^{n}} - \text{degradation rate}[\text{clmRNA}] \right) \\
\frac{\partial [\text{Proteincl}]}{\partial t} &= \text{max translation rate} [\text{clmRNA}] - \text{degradation function} \\
\text{degradation function}[\text{clProtein}] &= \begin{cases} \text{degradation rate}[\text{clProtein}] & \text{if } T < 40^\circ C \\ \text{current cl protein concentration} & \text{if } T \geq 42^\circ C \end{cases}
\end{align*}
\]

When LacI dependent promoter is active:

\[
\begin{align*}
\frac{\partial [\text{mRNAclacI}]}{\partial t} &= pLac \text{ max transcription rate} pLac \times K_{m\text{cl}}^{n} \left( \frac{K_{m\text{cl}}^{n}}{K_{m\text{cl}}^{n} + [\text{clProtein}]^{n}} - \text{degradation rate}[\text{lacImRNA}] \right) \\
\frac{\partial [\text{LacIProtein}]}{\partial t} &= \text{max translation rate} [\text{lacImRNA}] - \text{degradation rate}[\text{LacIProtein}] \\
\frac{\partial [\text{IPTG}]}{\partial t} &= -kb[\text{LacI}][\text{IPTG}] + kd[\text{IPTGLacIcomplex}] \\
\frac{\partial [\text{IPTGLacIcomplex}]}{\partial t} &= kb[\text{LacI}][\text{IPTG}] - kd[\text{IPTGLacIcomplex}] 
\end{align*}
\]
Protein modelling

Several computational structure prediction methods were used to find out if our fusion proteins are functional.

Structural alignment of two different models of secretion domain

Correctness of all generated models was evaluated using various bioinformatics tools.
Results

18 BioBricks

Bax K177027
Cro binding box (OR1) K177002
cro K177009
clt S K177050

LacI R0010 B0030 E0040 B0010 B0012
GFP E0040 B0010 B0012
AraC R0060 B0030
AraC O2 R0061
Cro binding box (OR3) K177047
Lysteriolysin O K177026

p53 K177016
ECFP B0032 E0022
cl lam B0032 C0051
lacl B0032 C0012 B0034 E1010 B0010 B0012
We improved cl BioBrick

cl protein is thermosensitive now

Primary version - BBa_C0051:

BioBrick received from MIT

Improved version - BBa_K177050:

1. Removal of STOP codon
   GGTAGTCAGGC -> GGTAGCCAGGC
2. Thermosensitivity
   TTTTGCAAGCAA -> TTTTGTAAAGCAA
We created bistable switch
A device to control biohazard genes.

GREEN colonies

RED colonies
...and it works!

30°C/IPTG

42°C

37°C

(stable for up to 9 h)
Human practices

1. Founding the first students’ society of synthetic biology in Poland

2. Participating in sociological research project regarding young Polish scientists

3. Short survey about Synthetic Biology made during the Science Festival in Warsaw
Human practices

How do people feel about synthetic biology?

140 persons were interviewed

<table>
<thead>
<tr>
<th>Benefit</th>
<th>Harm</th>
</tr>
</thead>
<tbody>
<tr>
<td>80.00%</td>
<td>20.00%</td>
</tr>
</tbody>
</table>

What will the development of synthetic biology bring us?
Human practices

Do they think it should be controlled?

140 persons were interviewed

<table>
<thead>
<tr>
<th>Yes</th>
<th>79.41%</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>20.59%</td>
</tr>
</tbody>
</table>
Human practices

By whom should the synthetic biology be controlled?

After the synthetic biology’s legal status is defined (by parliament) who should control the conducted experiments?

A. Government agencies
B. Scientific agencies
C. International agencies
D. Church authorities
E. Nobody

140 persons were interviewed
Powerful tool

New system for delivering various proteins to the eukaryotic cells

Cancer treatment

Medicine

Vaccines

Basic research

Nanotechnology
Final conclusions

• We improved already existing BioBrick – the cl protein is thermosensitive now.
• We created bistable switch and proved it works.
• We created the majority of parts needed to assemble the whole biological system.
Future plans

- Targeting the tumor cells via recognition the specific receptors on the cell surface
- Error proofing and disposing of the run down bacteria
- Mitochondrial transformation
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Yes! We’ve done it!