TEAM
BACKGROUND

Heart attack is one of the top five most expensive conditions (cost of about $11.5 billion for 612,000 hospital stays.)

A heart attack occurs every 20 seconds which results in a death approximately every minute.

Major causes of heart disease are excessive eating and hypertension sourced by salt consumption.
Survey amongst iGEMers reveals the excessive calorie intake
Average salt consumption of iGEMers exceeds by 3 grams
The more we eat, the more we are going to eat = Viscous circle
Excessive eating causes plaque formation!
Plaque rupture causes blood clotting!
Blood clot eventually blocks the bloodstream, causing hypoxia!
Possible oxidative stress also damages our cells!
1. Our cells must sense hypoxia

2. Hypoxia causing effects must be removed.

3. Cells must be prepared for a possible oxidative burst.

4. Our hypoxia sensing system must be compensated.
SENSING AT GENE LEVEL

BBa_K1456004
SENSING AT GENE LEVEL

BBa_K1456004
SENSING AT GENE LEVEL

Luciferase Conc. / Time Graph

- **BBa_K1456004**
SENSING AT GENE LEVEL

pHRE-Luc - HEK 293T/HEPG2

Fold activation

0 uM CoCl2/HEK 293T  100 uM CoCl2/HEK 293T  0 uM CoCl2/HEPG2  100 uM CoCl2/HEPG2

BBa_K1456004
SENSING AT PROTEIN LEVEL

BBa_K1456005
SENSING AT PROTEIN LEVEL

Normoxia

Ubiquitination and degradation of hydroxylated ODD in proteasome

no transcription

Gene of interest
Luciferase

BBa_K1456005
SENSING AT PROTEIN LEVEL

Hypoxia

Gene of interest
Luciferase

BBa_K1456005
SENSING AT PROTEIN LEVEL

BBa_K1456005
We designed two novel sensing mechanisms in mammalian cells.

Results of both sensing mechanisms show that we successfully sense hypoxia.

These results are compatible with the mathematical modelling we conducted.
THERAPY
THERAPY
THERAPY

75 KDa
50 KDa
25 KDa

BBa_K1456001
THERAPY

![Graph showing tPA Activity - HEK 293T/HEPG2](chart)

- **tPA Activity - HEK 293T/HEPG2**

<table>
<thead>
<tr>
<th>Condition</th>
<th>Fold Activity</th>
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</thead>
<tbody>
<tr>
<td>Ptre/HEK 293T</td>
<td>1</td>
</tr>
<tr>
<td>pTRE-tPA/HEK 293T</td>
<td>50,85398837</td>
</tr>
<tr>
<td>Ptre-HEPG2</td>
<td>1</td>
</tr>
<tr>
<td>Ptre/HEK 293T</td>
<td>44,85334361</td>
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<tr>
<td>Ptre-tPA/HEPG2</td>
<td>1</td>
</tr>
<tr>
<td>Ptre-tPA/HEK 293T</td>
<td>20,7602104</td>
</tr>
</tbody>
</table>

HEK 293T Lysate (intracellular)

HEK293T-HEPG2 Medium (extracellular)
THERAPY

SOD

BBa_K1456003
THERAPY

Blank

Negative Control

Our Sample

SOD1 Assay

Fold Activation

pTRE

pTRE-SOD1

BBa_K1456003
THERAPY

BBa_K1456002
THERAPY

BBa_K1456002
THERAPY

GPX1 Assay

<table>
<thead>
<tr>
<th>Condition</th>
<th>Fold Activation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1000 nM NaSeO3</td>
<td>1.0 (pTRE)</td>
</tr>
<tr>
<td></td>
<td>1.5 (pTRE-GPX1)</td>
</tr>
<tr>
<td>500 nM NaSeO3</td>
<td>2.0 (pTRE-GPX1)</td>
</tr>
</tbody>
</table>

BBa_K1456002
Aprotinin Activity

Serine Protease Activity

- pTRE
- pTRE-Aprotinin
- Negative control
- Bovine Aprotinin

THERAPY
SENSING

BBa_K1456015
SENSING

ρκBRE-Luc

Fold activation

Control  50 μM H2O2  20ng/ml hTNFα

BBa_K1456015
THERAPY ACHIEVEMENTS

✅ We successfully designed and produced four new therapeutic proteins.

✅ According to our specific assays all of these proteins were functional and working at precise levels.
P&P M2: CLARIFICATION
P&P M3: SPECIALIZATION
FUTURE PLANS
FUTURE PLANS

IMPLEMENTATION
FUTURE PLANS

SIDE EFFECTS
FUTURE PLANS

TERMINATION OF TREATMENT
SAFETY EXPERIMENT

![Graph showing the effect of Tetracycline concentration on Firefly/Renilla Luciferase activity. The graph plots % inhibition against Tetracycline concentration (0, 0.001, 0.01, 0.1, 1, 2, 0.25, 0.5). There is a significant increase in inhibition at 0.001 concentration compared to higher concentrations.](image-url)
## P&P M4: APPLICATION

<table>
<thead>
<tr>
<th><strong>Cost</strong></th>
<th><strong>Frequency of repeat</strong></th>
<th><strong>Area of influence</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart Attack (treatment) $3935* per attack</td>
<td>Heart Attack (treatment) 95% the risk of heart attack in 1 year</td>
<td>TIA (treatment) Heart Attack (treatment) Systemic</td>
</tr>
<tr>
<td>TIA (treatment) $6049* per attack</td>
<td>TIA (treatment) The risk of recurrence within 1 year: 14% The risk of recurrence within 5 years: 40%</td>
<td>TIA (treatment) Systemic</td>
</tr>
<tr>
<td>Gene therapy $25000</td>
<td>Gene therapy Don’t Repeat</td>
<td>Gene therapy Local</td>
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</table>
ACHIEVEMENTS OVERALL

✓ We have created a system which aims to treat ischemia: the number one cause of deaths worldwide.

✓ We designed 7 new BioBricks for mammalian cells which are all functional and have been submitted to the Registry.

✓ We evaluated several mathematical modelings of our sensing devices which are compatible with our wetlab results.

✓ We questioned the future plans of our project coming up with the design of safety mechanism working significantly according to our model and experiments.

✓ We have separated our P&P into four modules where extensive research and evaluation to find alternative ways of raising peoples’ awareness.

✓ We collaborated with METU-Turkey and Paris Bettencourt 2013 iGEM teams.
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- **Assist. Prof. Dr. Muradiye Acar** TOU Department of Medical Genetics
- **Assist. Prof. Dr. Sadik Cigdem** TOU Department of Medical Genetics
THANK YOU
FOR YOUR PATIENCE