

May --

01/05

- First lab visit of the team and instructions on how to handle the instruments and safety guidelines were presented

02/05

- Practicing inoculation, streaking and spreading of bacterial cultures of a biobrick used in previous year
- LA plates and LB media preparation

03/05

- Practicing plasmid isolation and transformation with the part inoculated yesterday
- Inoculation of following parts from iGEM kit plate:-
  1. RBS + GFP + T(without deg tag)
  2. RBS + TetR + T
  3. SYFP2
  4. J23119 promoter
  5. J23107 promoter
- Plasmid isolation of :-
  1. RBS + GFP + T(without deg tag)
  2. RBS + TetR + T
  3. SYFP2
  4. J23119 promoter
  5. J23107 promoter
- Transformation of plasmids mentioned previously following iGEM protocol

04/05

- Discussion on toggles switches and oscillators in relation to iGEM IITD project - 2016. More detailed discussion on light activated self-repression and its effect on oscillator frequency.
- Discussion on IITD 2015 project
- Discussion on some iGEM projects of other countries.
- Received the colonies transformed yesterday

06/05

- Further discussion on the research papers and different aspects of project

09/05

- Formation of a marketing and human practices team and discussion on marketing strategies used last year

11/05

- Video and content for crowd-funding initiative
- Mails and calls to potential sponsors

13/05

- Discussion on gene regulation in prokaryotes and eukaryotes with special emphasis on prokaryotes.
- Discussion on how we can modify the Lac system by keeping its switching on properties intact and replacing the gene part with, suppose, insulin such that presence Lactose molecules trigger the activation of insulin gene
- Discussion on the different class of promoters

15/05

- Discussion on PCR
- Sample PCR product formed and ran on gel

17/05

- 1. Logic Gates:
  - a) AND GATE:

- discussion on simple AND gate
- AND gate with modified tRNA (Discussion on tRNA that would read TAG as Serine)
- b) OR GATE
- c) NOR GATE
- d) XOR GATE: Using combination of different gates i.e. OR , AND , NOR, NAND, NOT and further discussion on other ways of XOR gate formation.
- 2. Steady state: Rate of accumulation=0
- 3. Discussion on different equations for rate of change of protein and rate of change of RNA (First and second order reactions) by discussing the journey from promoter to mRNA to protein to nothingness.
- 4. Enzyme Kinetics
- $S + E \rightleftharpoons ES \rightarrow P + E$
- Differential equations of rate of change of ES and P discussed with equations of conservation of mass for the same.
- Graphical analysis of ES/E0 vs S/S0 with discussion on **quasi steady state**.
- Graphical discussion on Concentration vs Time of the reaction.
- 5. Michaelis menten equation
- 6. Discussion on Degradation Tag and how it works in a bacterium where translation and transcription happen in the same compartment and there are no stop codons.
- 7. Discussion on Cancer Cell about how it was thought to find a cure of cancer using these logic gates with inputs as miRNA(regulate genes) and output as apoptosis.
- 8. Example of virus that can be used as vectors for human cells. e.g. *Vaccinia* virus

18/05

- Discussion on ways to generate different signal responses in bacterial gene regulatory circuits

22/05

- Discussion on :-

- 1) Chemotactic response in bacteria
- 2) Temporal and spatial response in chemotaxis
- 3) Mechanism of chemotaxis in bacteria flagellum
- 4) Electrical model of chemotaxis
- 5) Adaptation time of chemotaxis
- 6) Finding electrical analogue of the biological system by varying frequency of the electrical circuit made
- 7) Finding optimal frequency
- 8) Biological Robustness and causes of it in a biological system
- 9) System controls, bistability, modularity, buffering, bow-tie framework, decoupling
- 10) Robustness in signal transduction pathways and insect's segmental development
- 11) Robustness trade-offs

24/05

- Discussion on :-

- 1) Design principles of biochemical oscillators
- 2) Time delay's relevance in oscillation
- 3) Introduction of time delay with Intermediates
- 4) Limit cycles, conservative cycles and circadian rhythms
- 5) Time delay by positive feedback
- 6) Different classes of feedback
- 7) Linear and hyperbolic response
- 8) Goldbeter Koshel Function
- 9) Sniffers
- 10) Toggle Switch

## 11) Mutual inhibition and mutual activation

26/05

- Discussion on :-

- 1) A brief review of Sniffers, Buzzers and Toggle switch
- 2) Negative feedback oscillations
- 3) Positive feedback oscillations
- 4) Neutral feedback oscillations
- 5) Sub-critical and Super-critical hop bit
- 6) Study of mitosis

27/05

- Prepared lab supplies like LA, LB etc. to be used in the lab

29/05

- 1) Preparation of LuxI and PLux double digest using restriction enzymes EcoRI and PstI
- 2) Preparation of buffer and electrophoresis gel
- 3) Loading ladder, control and digest in the wells in gel
- 4) Electrophoresis and subsequent observation of gel to ensure digestion of plasmids

31/05

Discussion on:-

- 1) Plasmid isolation
- 2) Lysis solutions and their composition
- 3) Function of each component in the solutions