

# Minutes of the third iGEM meeting

04/09/2010

During the 3rd iGEM meeting the group discussed and developed potential ideas for a project. Apart from this, Charanya who could not attend the last meeting where all others presented their favourite project from last year's iGEM competition, presented her favourite ideas and Lukas informed us about new developments concerning the group wiki.

## 1 Group wiki and Google Wave

Lucas created a wiki-page for our iGEM team using free space on his server. The link is: <http://praxis-ergologo.de/igem/>. We need to register and Lucas confirms the registration. This will be a temporal server until we have the official one up.

Jon was wondering if we should adopt Google Wave as an alternative to the email and the wiki for collaborating and discussing in real-time. For those of you who don't know what Google Wave is or what it offers; it is basically email - in real time. Similar to instant messaging but with a few twists. Attached is a website that will hopefully help with the explanation: <http://mashable.com/2009/05/28/google-wave-guide/>.

## 2 Charanya's presentation

### 2.1 JHU - Artificial chromosomes

By using overlap assembly PCR, followed by Uracil Specific Excision Reaction (USER), and finally, multiple rounds of homologous recombination, the JHU have created pieces of chromosomes and finally full *Saccharomyces cerevisiae* Sc2.0 chromosomes. They have used software tools that have also been entirely been created and written by them. The synthetic yeast redesign carries several useful applications.

### 2.2 K.U.Leuven - Vanillin producing bacterium

Essencia coli is a vanillin producing bacterium equipped with a control system that keeps the concentration of vanillin at a constant level and requires

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blue light irradiation. The K.U.Leuven team has produced a wonderful feedback mechanism using the vanillin receptor, blue light receptor and the key-antikey (which are basically complementary RNA strands that compare the signals from blue light and vanillin receptor to keep control of vanillin synthesis).

### **2.3 Imperial College London - Safe inanimate pill**

Team Imperial College London has attempted various steps in the production of a safe inanimate pill. This sequential process involves drug production, protective encapsulation and genome deletion by means of chemo, auto and thermo induction. This methodology avoids the need for expensive storage, packaging and purification processes.

### **2.4 PN UNAM Mexico - Spatio-temporal structures**

This team's goal is to show that spatio-temporal structures can be generated by the behavior of a genetic regulatory network. The action of the morphogenes as originally proposed by Turing is equivalent to the effect of diffusion of chemicals interacting with the synthetic network. This they show by means of a model with a self-activatory and inhibitory network (lac and lux operons). The biobricks included various promoters, plasmids and inverters which they have made really good use of.

## **3 Potential projects**

The following proposed projects are sorted after the time we spent discussing them.

### **3.1 Sequencing bacteria**

The idea is design bacteria that are able to sequence a strand of DNA. This approach should lead to a DNA sequencing process that is on the one hand faster than current sequencing techniques and on the other hand cheaper. We first thought about designing four different bacteria, one specific for each nucleotide. Since we will probably encounter a size problem because the bacteria are much bigger than the DNA, we thought about some adaptor between DNA and the bacteria. We had the idea to use the codon-anticodon system of tRNA. The output system could be the expression of a certain fluorescent protein. Everyone was really excited about this idea and we have split up groups to brainstorm on this.

The second idea for the sequencing bacteria idea was just to label a DNA sequence with one specific fluorescent nucleotide by PCR (4 mixes for each nucleotide) and to use exonucleases in the bacteria to cut one base

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after another in the four mixes. The fluorescent signal in the DNA would be quenched by the other nucleotide not fluorescently marked (for example some aminoacid). All bacteria would start at the same time to cut a nucleotide and digest it. The reaction would be repeated for a few hundred times on a well plate for every type of nucleotide to get plenty of data. From the fluorescence signal output we could calculate the sequence itself. At one end of the DNA should be a non-digestable nucleotide to prevent two exonucleases from working on each end. The problem would be to get the DNA in the bacteria without much work (some simple transformation procedure), to cut the DNA and digest the fluorescent marker in a two-step-system and to start all bacteria simultaneously (maybe by light of a defined wavelength).

## **3.2 Biosensor to distinguish between viral and bacterial infection**

### **3.2.1 General idea**

Keeping in mind that Procalcitonin is a biomarker for Sepsis, it was initially suggested that we develop a biosensor system in order to rapidly distinguish between a viral and bacterial infection. With Procalcitonin serving as a positive marker for endotoxin/ bacterial infection.

### **3.2.2 Theoretical background**

The hormone Calcitonin is assembled from Procalcitonin (PCT). Normally PCT is present in minute concentrations ( $< 0.05 \text{ ng/mL}$ ) in the blood of healthy patients. However, during the onset of Sepsis (see definition below) the PCT-level can reach very high levels ( $1000 \text{ ng/mL}$ ).

Sepsis is the result of excessive stimulation of the host immune cells, most notably monocytes and macrophages, by bacterial lipopolysaccharide. At the fundamental level, the symptoms of septic shock syndrome (SSS) are caused by excessive release of two pro-inflammatory cytokines, Tumor necrosis factor- $\alpha$  and Interleukine- $1\beta$ . It is the excessive amounts of these two cytokines that make SSS so lethal.

### **3.2.3 Problems**

Later we found out that there is already a procalcitonin test on the market that has been FDA approved. (Information: <http://www.procalcitonin.com/>) They use a two antibody system with different epitopes on the peptide and they quench a linked fluorescent marker from one to the other antibody. Since antibodies are in general expensive, our chance would be to design a system that is less pricey.

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### **3.3 'Poo be gone' system**

We thought about engineering a spray containing enzymes or bacteria that can be sprayed on dog poo in the streets. The spray should lead to a faster degradation of the poo and maybe thereby produce a nice smell. As a matter of fact for any waste treatment. We can also think of degrading plastics form by enzymes or bacteria.

## **4 Next meeting**

The next meeting will take place on friday, april 16 at 4pm. Again it's only the students who meet to further discuss the ideas. We would like to meet with the 'doctors' on april 19 or 20 when all students are back from holiday.