Diary of ZHANG Zidong

June

6.20-6.25

Preparation work for our lab:

1 I cleaned the lab thoroughly

2 I prepared some LB plates and liquid LB medium

3 I prepared different kinds of backbones which might be used in the future. (backbones are from distribution kits. They were amplified in cells and double digested by EcoRIhf and PstI)

6.26-6.30

We decided to choose luxbrick from partregistry (Cambridge 2010 iGEM) as the light emitting cell for our light communication part.

I transformed the part BBa_K325909 into different kinds of cells (DH5alpha, BMTOP10) and induced them with L-arabinose but the cells didn’t emit light as expected

7.1-7.7

From literature I found that I grew the cells in wrong temperature and I

moved the cells from 37℃ to 30℃. The luxbrick harbored TOP10 cell finally emit blue light 6hrs after L-ara induction.

7.8-7.14

1 I chose some strong constitutive promoters and intended to put the luxbrick under those promoters to increase the intensity of light.

2 I PCR the RBS and coding sequence of biopart luxbrick (BBa_K325909) using phusion polymerase.

3 I transformed the LexA-VVD plasmid and ColE-mcherry plasmid from prof.Yang into BL21(delta LexA)strain from Prof.Yang.

7.15-7.21

1 I finally chose T7 promoter (BBa_I712074) and ligate it with luxbrick PCR product. Then a new part T7-Llux was successfully constructed.

2 I did some preliminary experiments to test Prof.Yang's strain and plasmid. The strain containing LexA-VVD and ColE-mcherry worked well at 30℃ (the bacteria were red in dark and white under light)

3 I sent my request for some delta-hns strains to CGSC (coli genetic stock center) but I didn't receive the strain. I guess the strain was stuck in the customs.

7.22-7.28

1 I began to characterize Prof.Yang's LexA-VVD and ColE-mcherry in a more detailed way, which is the time curve.

2 the induction of T7-lux contained BL21 with IPTG failed.

7.29-8.4

1 the induction of T7-luxbrick succeed but the intensity of light emitted is very low. Myelin gave me a plasmid containing a T7-polymerase under a constitutive promoter. After I changed its backbone and transform it into T7-lux harbored cell, I was informed that the plasmid containing T7 polymerase was wrong.

2 I got some data, especially some great photos of the characterization experiment.

3 I sent my request for delta-hns strain to Dr. Summers in Cambridge but I didn't received the strain either. I guess the strain was unfortunately stuck in the customs again.

8.4-8.9

I went back home for several days

8.10-8.14

I repeated the previous characterization experiment in a different way, and received better data.

8.15-8.29

We three freshmen went to military training

8.31-9.5

1 I continued Zhilei’s work on red luciferase, which might be the reporter of our light-on system.

2 I helped Zhang Hong on his characterization work.

3 I began to write our wiki on bio-light communication