Diary of ZHAO Zhilei

As the team leader of Peking iGEM 2012, my main duty is not cloning or characterizing our parts, on which my talented team members had done very good job. Instead, I focused on devices and experiments design and testing the possibility of novel ideas. I successfully made an incubator shaker for testing whether our Luminesensor was sensitive to natural light. I anticipated in the design of our 2D and 3D printing program and the magic iPrinting program! I also designed the experiments and devices for light-communication with other team members. The feeling was pretty wonderful when realized that we were the first team to achieve light-communication in the world!

7.1~7.7

Plasmid Backbones Plan:

1. Transform pSB1A3 plasmids and pSB3T5 plasmids to Trans5-alpha competent cells.

2. Miniprep.

- 3. Measure the DNA concentration of miniprep products.
- 4. Stock the products in -20° C.

7.8~7.14

Growth curve measurement with LU Tian:

- Pick colonies of Trans5-alpha, BL21(DE3), Luxbrick, △ cheZ strain, △ LexA strain and shake overnight at 37 °C.
- 2. For every 5 mL medium in the test tube, 10 μ L cultures were inoculated for each strain.
- Shake at 30 °C. Take samples off the test tube and measure the OD₆₀₀ every 30 minutes.
- 4. Draw the growth curve of each strain.

7.15~7.21

Test the property of Luminesensor with ZHANG Zidong:

- Pick colonies of Luminesensor+colE-mCherry and shake overnight at 30 °C under light.
- 2. For 30 mL medium in the conical flask, 300 μ L cultures were inoculated.
- 3. Shake at 30 $\,^{\circ}C$ under LED arrays.
- For every 2 hours, take 3 mL sample off the conical flask into test tube under dark, still shake at 30 °C.
- 5. After 20 hours, centrifuge and resuspend all samples obtained into 1.5mL EP tubes.
- 6. Take photos of all the samples under the same light environment with the same camera parameters sets.

7.22~7.28

Make incubator shaker for testing whether Luminesensor is sensitive to natural light.

- 1. Draw the design sketch of incubator shaker.
- 2. Make the incubator shaker with electrical air fan and foam.
- 3. Test the shaking efficiency of the incubator shaker.
- 4. Put the incubator shaker on the balcony under natural light.
- 5. Ready for work!

7.29~8.5

First try for light-communication.

- 1. Design the devices for light-communication.
- 2. Test the efficiency of the air pumping.
- 3. Pick colonies of Luxbrick and shake overnight at 30 $\,^{\circ}C$ under light.
- 4. For 100mL medium in the conical flask, 1 mL cultures were inoculated.
 3mL 0.1M arabinose and 2 mL 0.01M ATP solution are added.
 Control test without the cultures of Luxbrick.
- Shake the mix at 30 °C with the pumping of air for 10 hours.
 Pick colonies of Luminesensor+sulA-GFP and shake overnight at 30 °C under light.
- 6. For 3mL medium, 30 μL cultures were inoculated in the test tube.Put the test tube into the conical flask.

The whole device was packed with three layers of aluminum foil to ensure a dark environment.

Shake at 30 ℃.

- After 10 hours, centrifuge and resuspend all the samples obtained into 1.5mL EP tubes.
- 8. Take photos of all the samples under the same light environment with the same camera parameters sets.
- 9. Take 500 μ L of all the samples into a black Elisa plate and a transparent Elisa plate.
- 10. Measure the OD_{600} and GFP expression of all the samples.

8.6~8.12

Optimization for 2D printing program with LI Hanxi and SUN Sibai:

- 1. Design the light-emitting device for 2D printing program.
- 2. Test whether the light output of the light-emitting device is parallel.
- 3. Measure the light intensity of the light output at the 10 cm distance.
- 4. Make the mask for printing with black plastic paper.
- 5. Mix the Luminesensor+sulA-GFP plates.
- 6. Cover the mask on the top of the plate. Put the plate under the light emitting device.
- 7. The whole device was packed with three layers of aluminum foil to ensure a dark environment.
- 8. Put the whole device at 30 $^{\circ}$ C.

9. After 10 hours, put the plates under the blue light for GFP excitation. Take photos of the plates.

8.13~8.19

iPrinting program with LI Hanxi and SUN Sibai:

- 1. Design the device for iPrinting program.
- 2. Calculate and measure the focal distance of the optical lens group.
- 3. Make the device based on the focal distance.
- 4. Place the iPad, on which a picture of "apple" was displayed, on one side of the device and the plate mixed with Luminesensor+sulA-GFP on the other side.The whole device was packed by aluminum foil to ensure a dark environment.
- 5. Put the whole device at 30 $^{\circ}$ C.
- 6. After 10 hours, put the plates under the blue light for GFP excitation.Take photos of the plates.

8.20~8.26

Video for light-communication:

- 1. Pick colonies of Luxbrick and shake overnight at 30 $\,^{\circ}C$ under light.
- For 100mL medium in the conical flask, 1 mL cultures were inoculated.
 3mL 0.1M arabinose and 2 mL 0.01M ATP solution are added.

Control test without the cultures of Luxbrick.

- 3. Shake the mix at 30 $^{\circ}$ C with the pumping of air for 10 hours.
- 4. Pick colonies of Luminesensor+sulA-GFP and shake overnight at 30 °C under light.
- 5. For every 5 mL medium in test tube, 50 μ L cultures were inoculated.
- 6. Put the test tube into the conical flask.
- 7. The whole device was packed with three layers of aluminum foil to ensure a dark environment.
- 8. Shake at 30 $^{\circ}$ C.
- After 8 hours, take 300 μL samples off the test tube for every 15 minutes. Centrifuge and resuspend all the samples obtained in 1.5mL EP tubes.
- 10. Put the samples under the blue light for GFP excitation. Take photos of all the samples under the same light environment with the same camera parameters sets.Take photos of the conical flask with the Luxbrick under dark environment at the same time.

8.27~9.5

- 3D printing program with LI Hanxi and YAN Jiawei:
- 1. Design the device for 3D printing program.
- 2. Calculate and measure the focal distance of the optical lens group.
- 3. Make the device based on the focal distance.

Put the beaker with solid cultures of Luminesensor+sulA-GFP under the light of the device.

The whole device was packed to ensure a dark environment.

- 5. Put the whole device at 30 $^{\circ}$ C.
- 6. After 10 hours, put the beaker under the blue light for GFP excitation.

Take photos of the beaker.