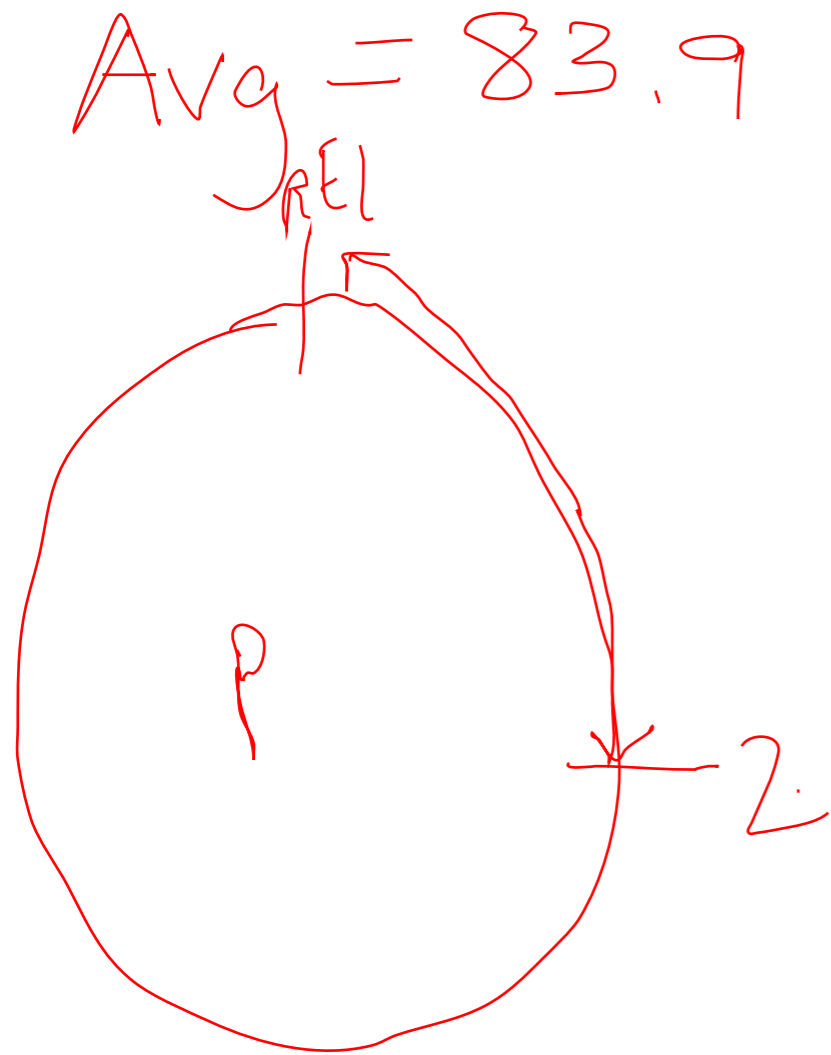


A little about the lab certification...



No DNA \equiv No Ab

Unknown DNA \equiv other cells

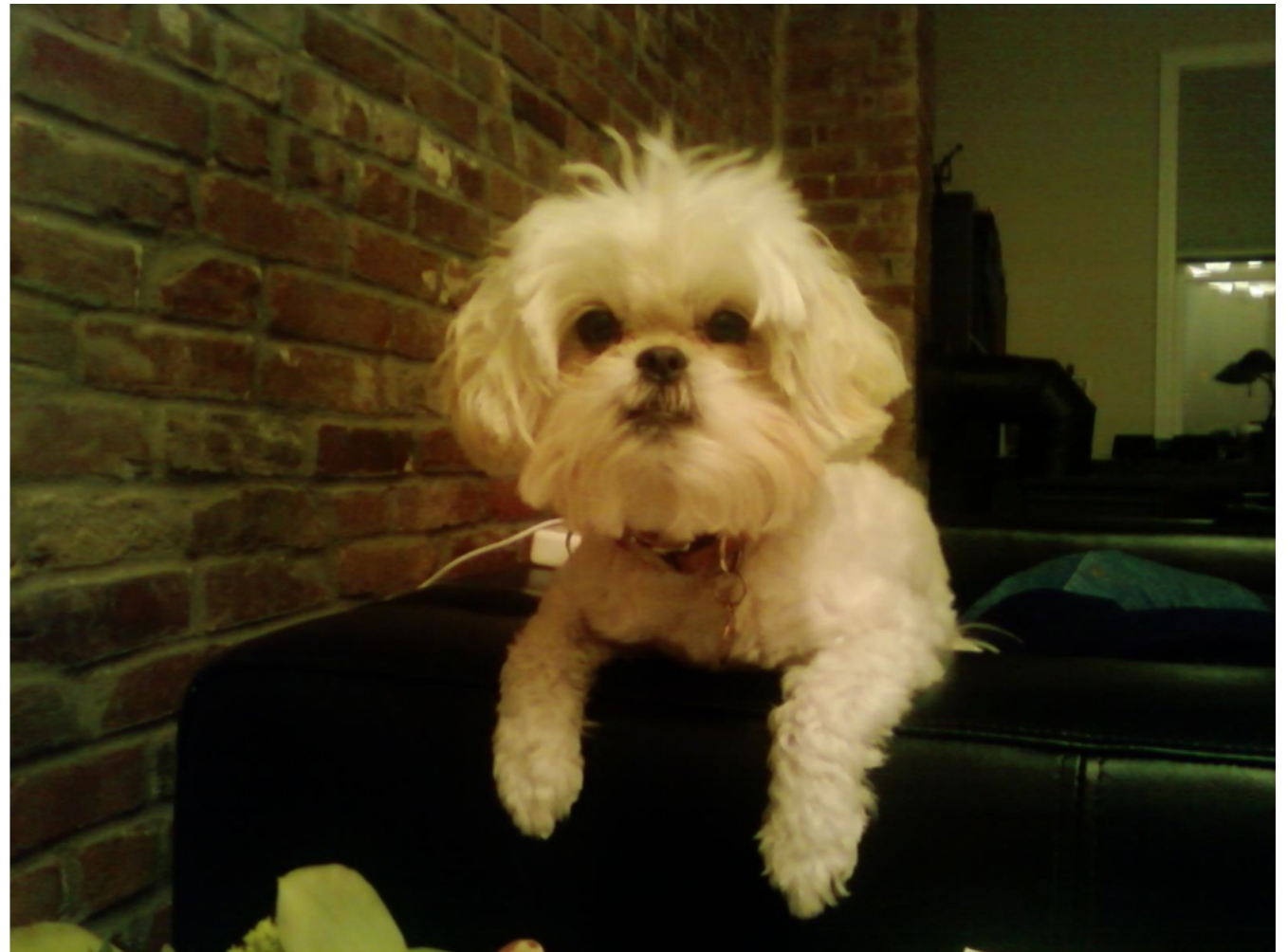
M2D I: Testing an engineered biological system

10/11/12 13:14:15 pm

Mod2: System Engineering Bacterial Photography

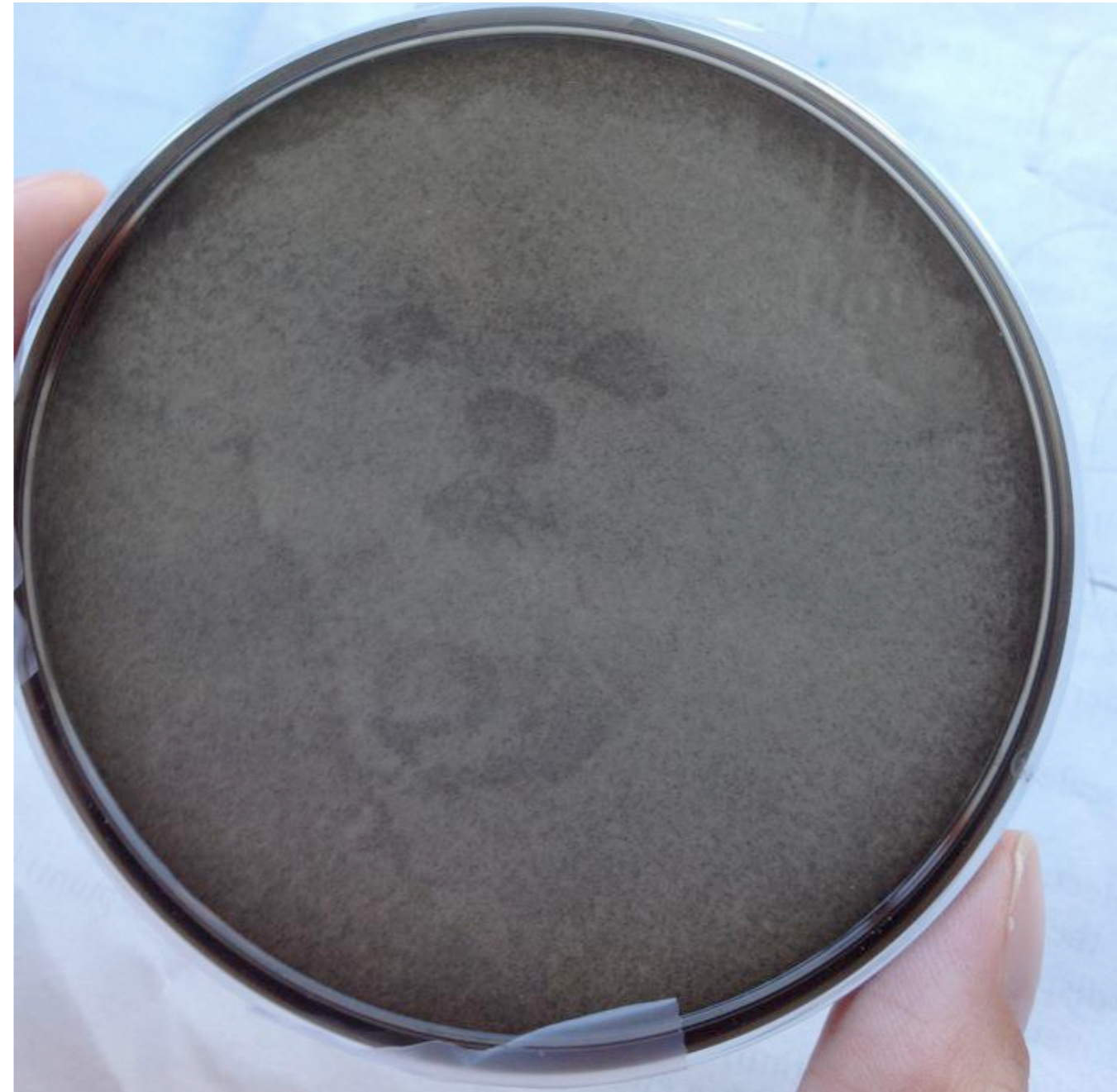
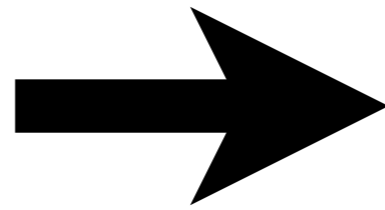
56-389

Butterstick



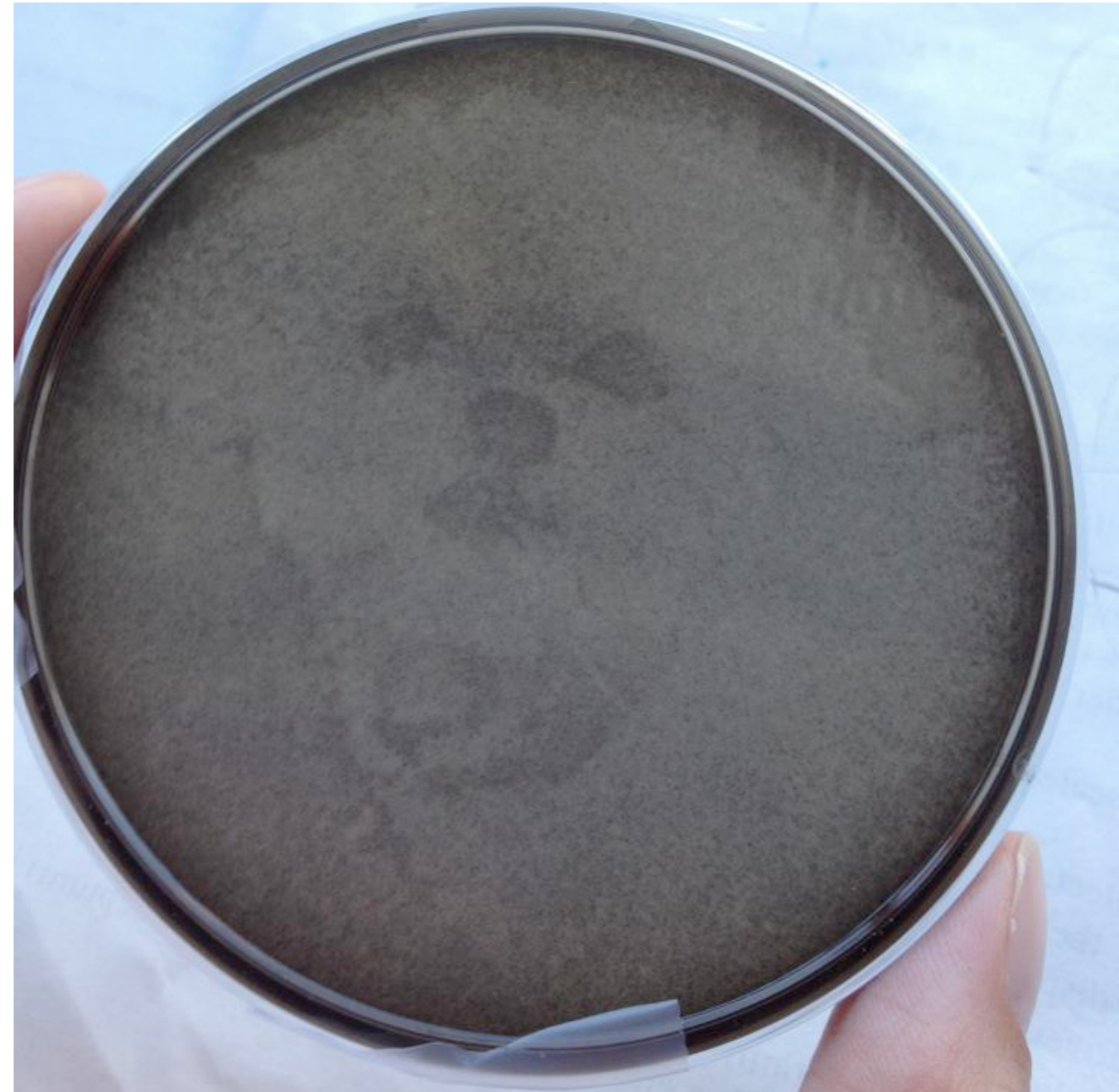
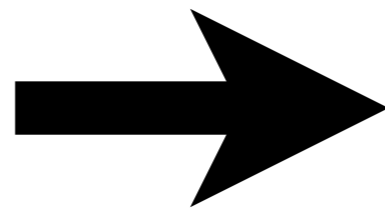
Mod2: System Engineering Bacterial Photography

Butterstick



Mod2: System Engineering Bacterial Photography

Butterstick



“What manner of sorcery is this?”

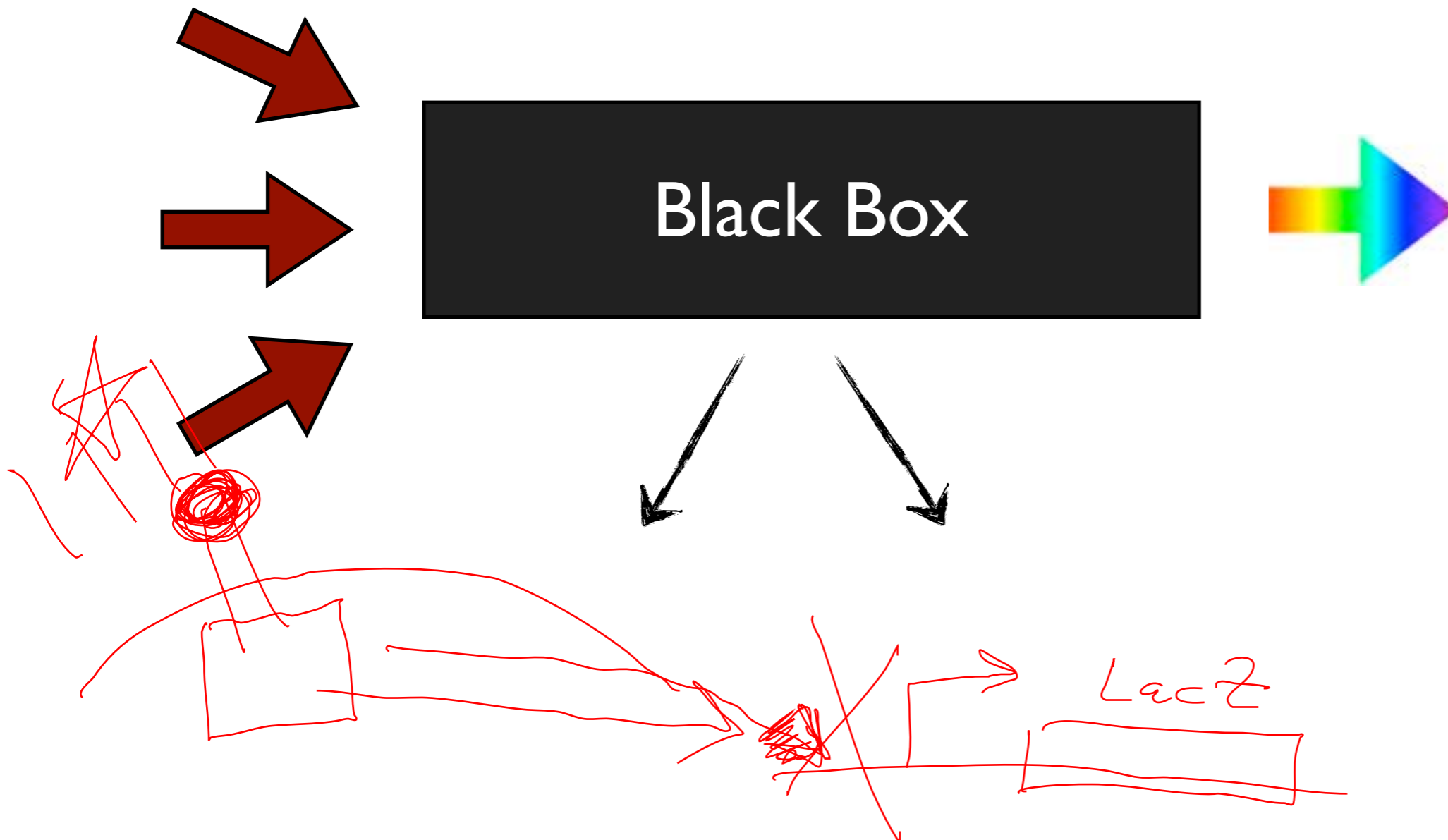
“wow! there is just so much
awesome all over this!”

What *is* the manner of sorcery involved?

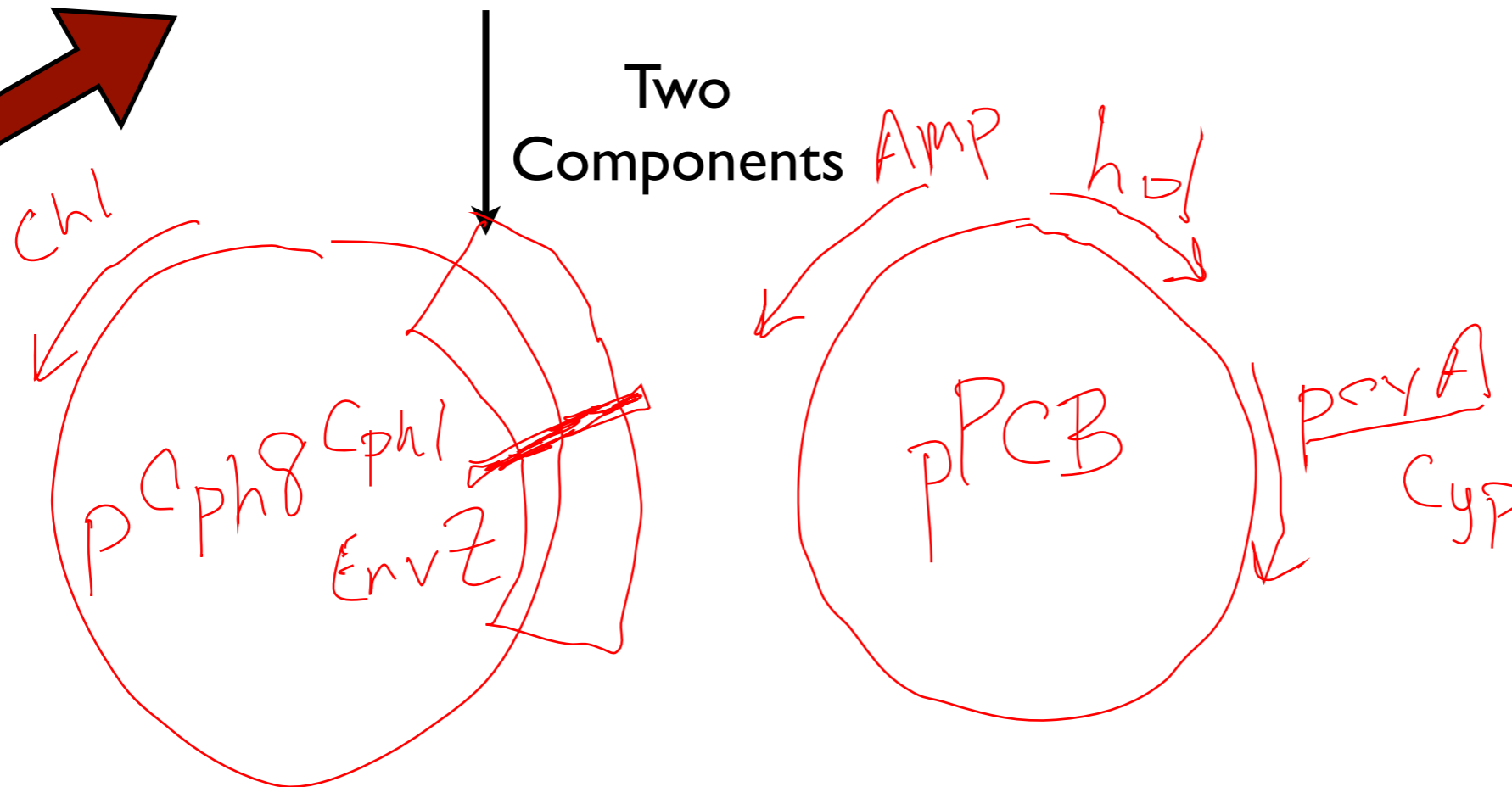
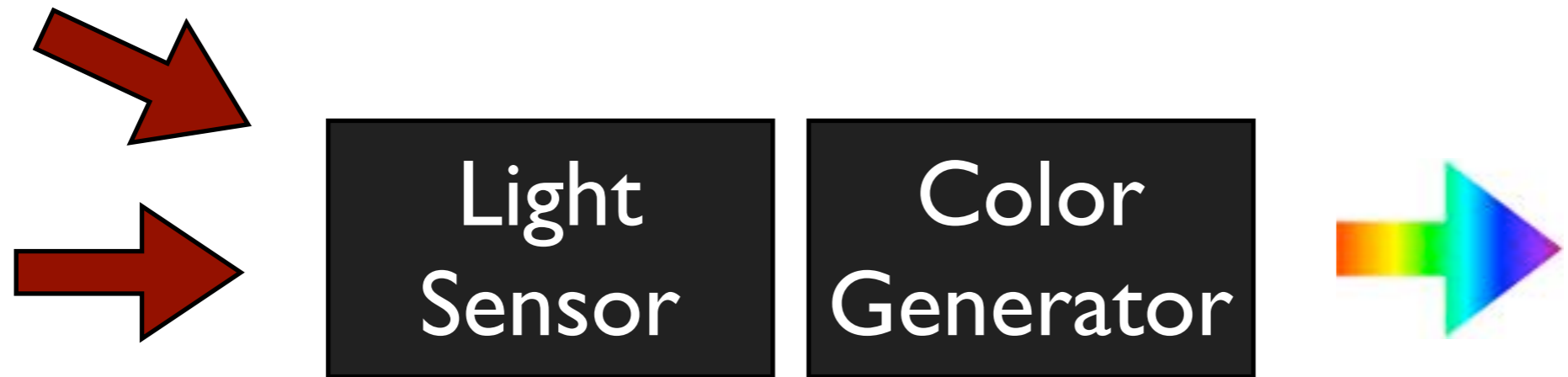
An engineered biological light sensing device.

Input: *Light*

Output: *Color change*



A little deeper into our black box:



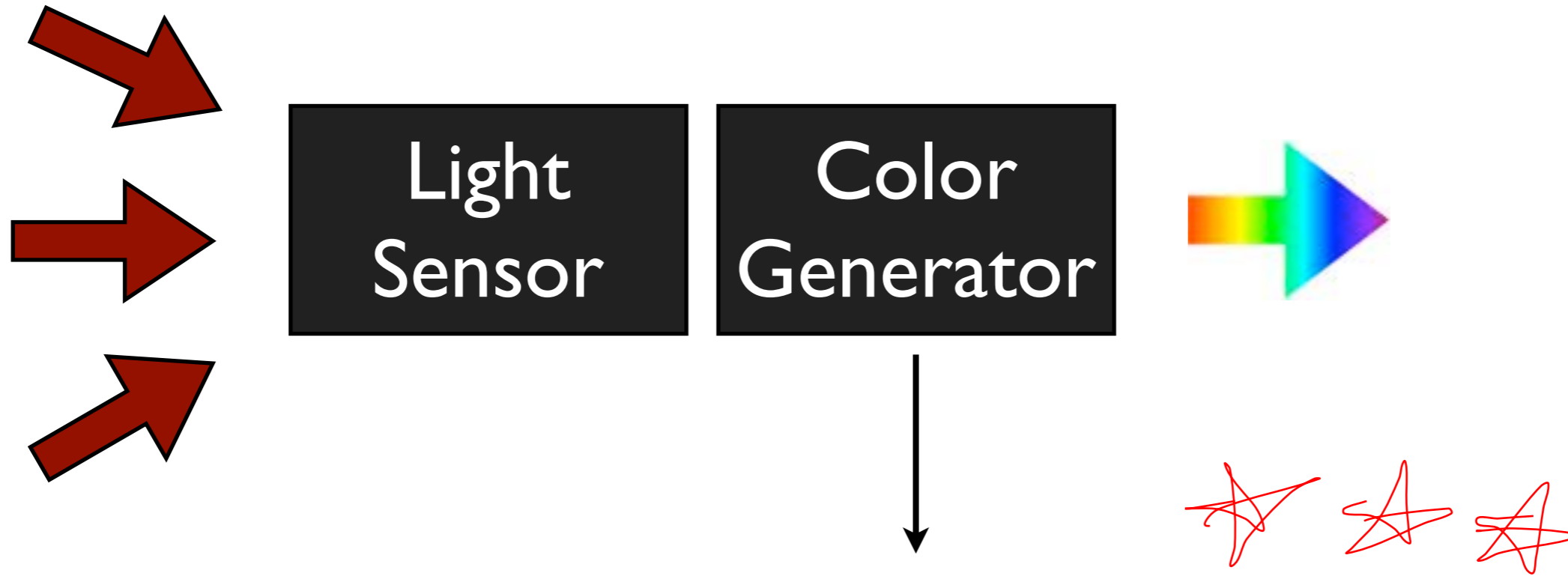
CphI = bacterial photoreceptor
EnvZ = cytoplasmic responder (gene regulation!)

PCB = photoresponsive protein from *Synechocystis*

Yuri Gorby, J.C. Venter Institute



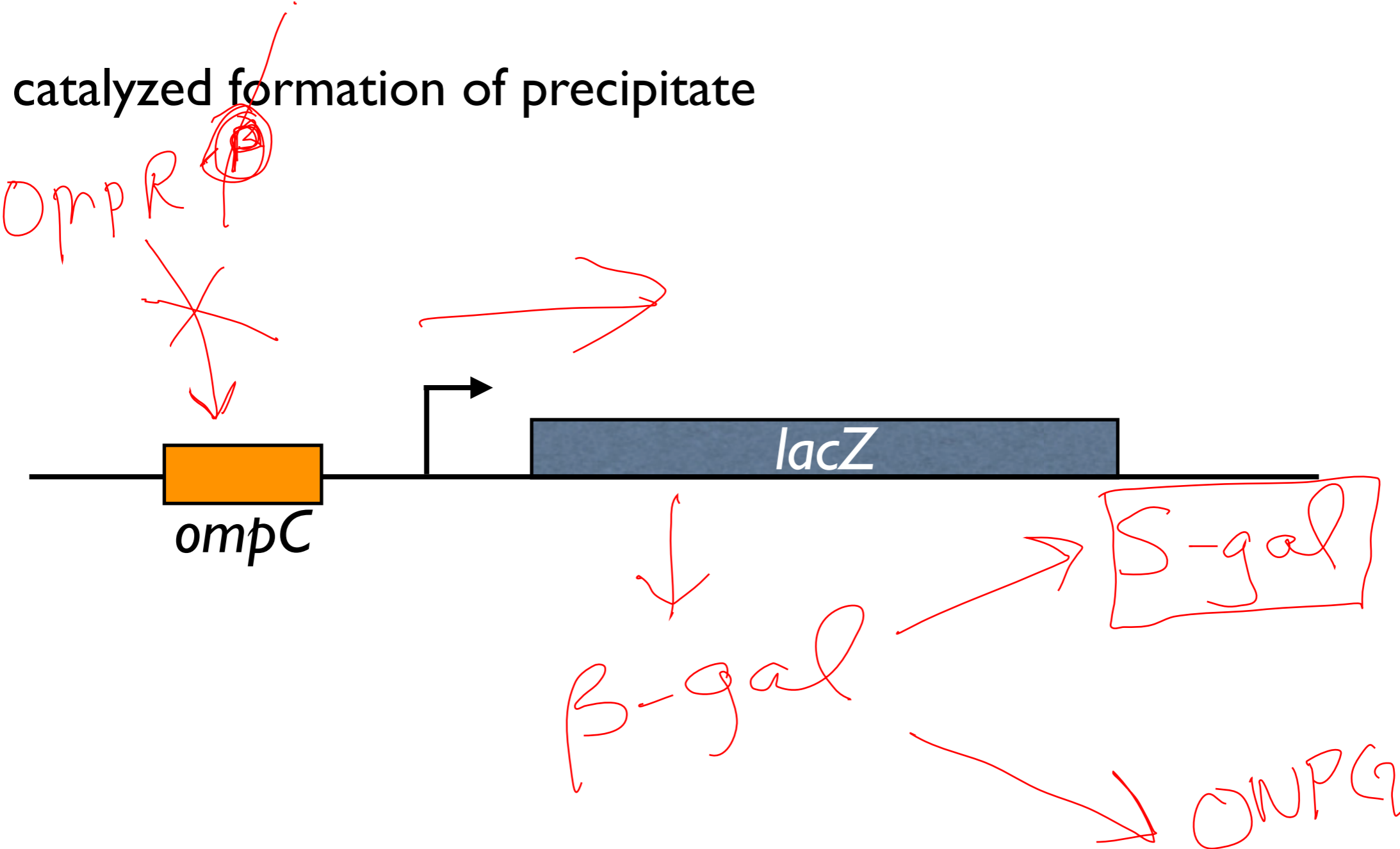
A little deeper into our black box:



	"Wild Type"	20.109
Sensor <i>HK</i>	<i>EnvZ</i>	<i>Cph8</i>
Responder	<i>OmpR</i>	<i>OmpR</i>
	<i>Osmolarity</i>	<i>Light</i>

How do we measure the efficiency of our engineered system?

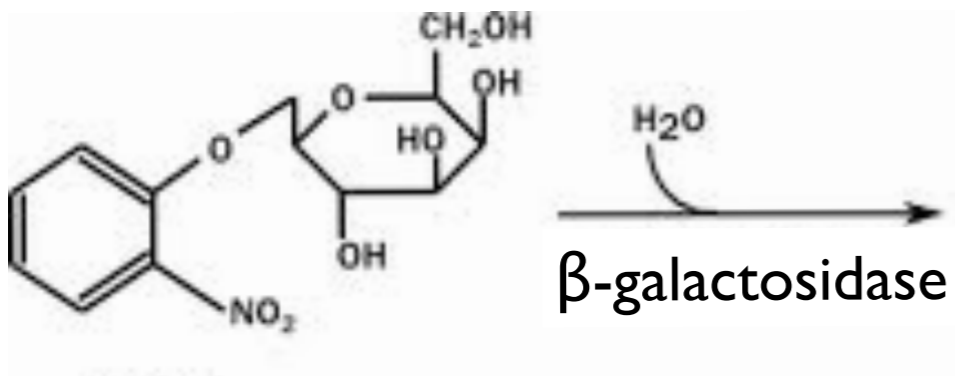
LacZ catalyzed formation of precipitate



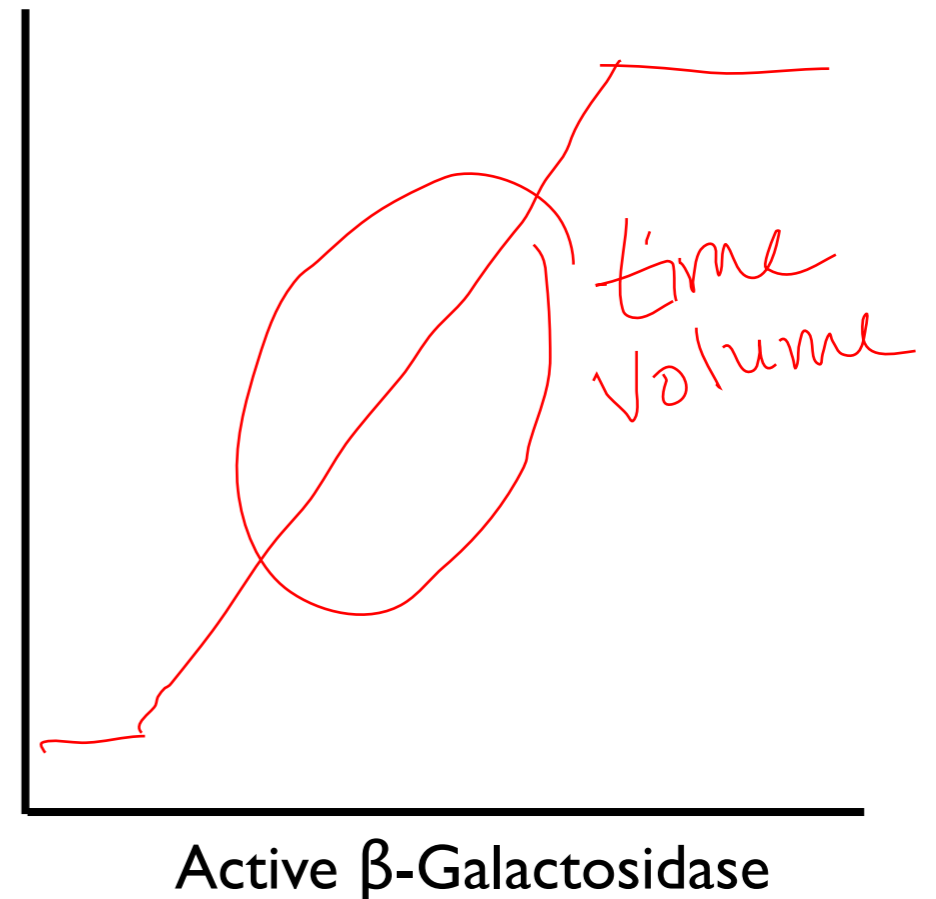
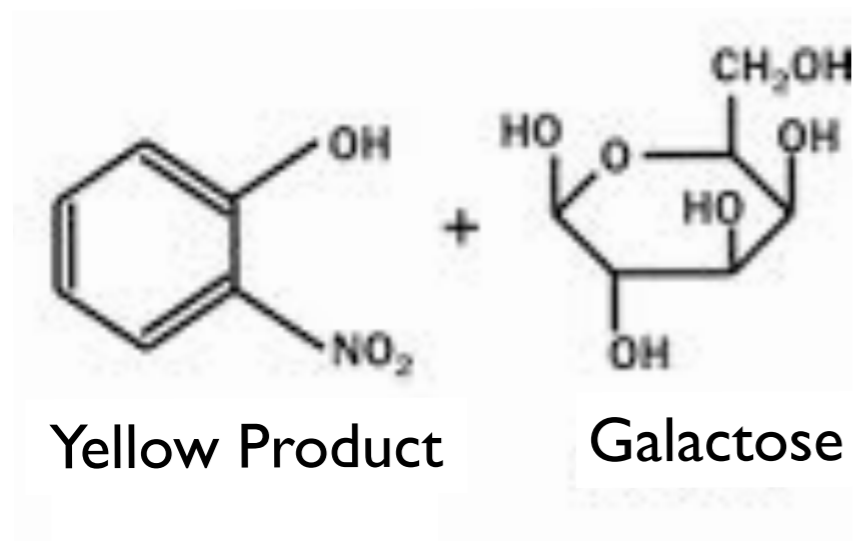
How do we measure the efficiency of our engineered system?

Limitations of our equipment and our assay:

ONPG



Yellow Color
Abs = 420 nM



How do we measure the efficiency of our engineered system?

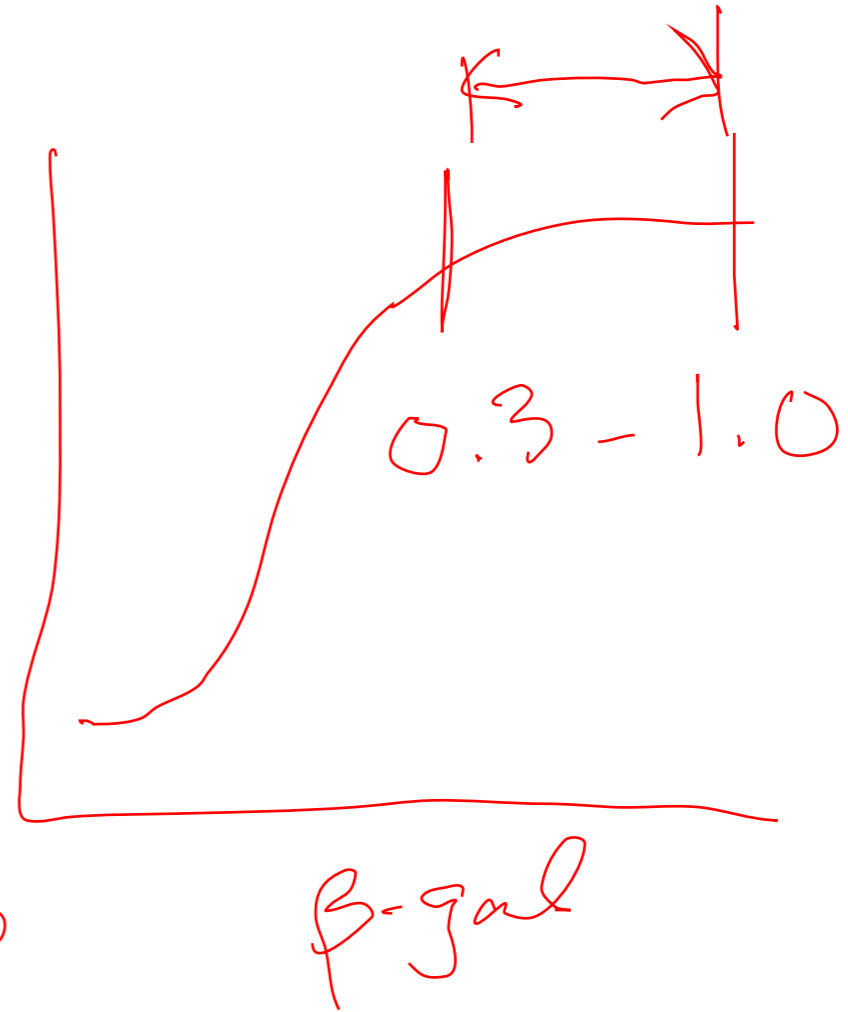
β -Gal Assay:

1. Measure [cell]
2. Lyse cells
3. Start Reaction
4. Stop Reaction
5. Get rid of debris
6. Measure yellow product
7. Calculate activity

NBS
~~10/15~~
Abs600

Abs550

420



$$1 \text{ Miller Unit} = 1000 * \frac{(\text{Abs}420 - (1.75 * \text{Abs}550))}{(t * v * \text{Abs}600)}$$

Plan for today & Mod2 Assignment #1

- ① Small liquid cultures of NB466 - quantification
- ② plates - visual ↖ 3Ab
- ③ β -gal - NB5

Pick your paper!