20.109
Synthetic Biology Module
Lecture #1

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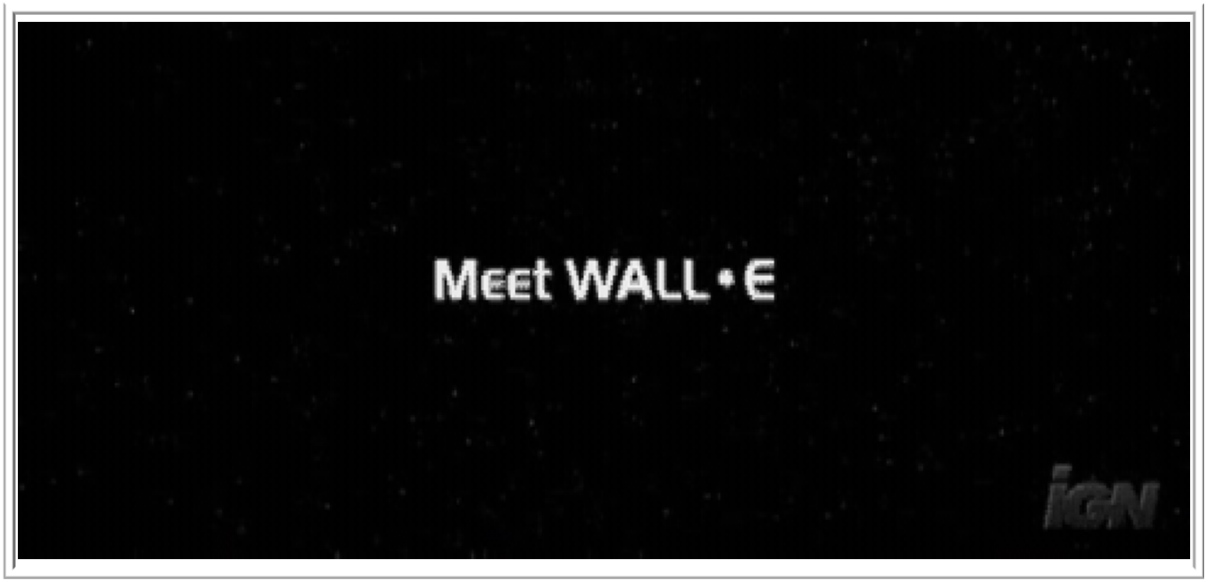
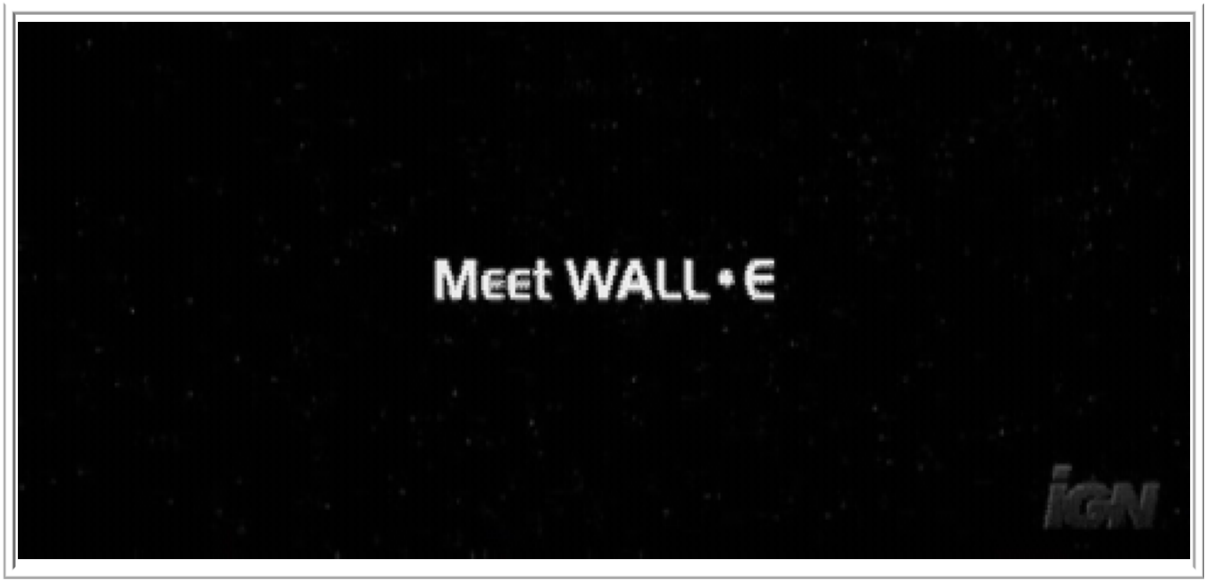
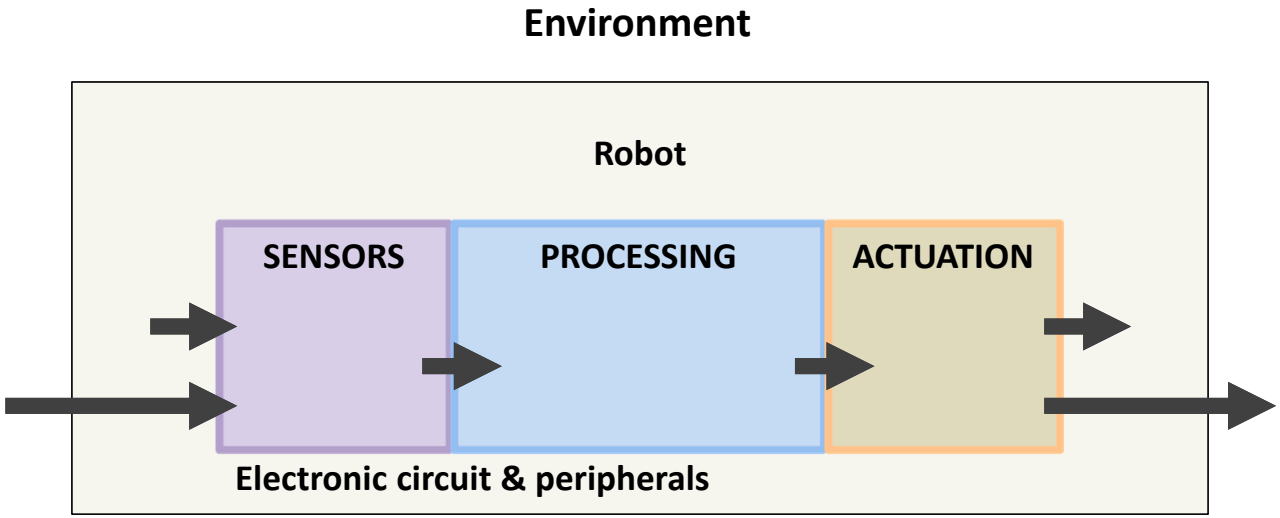
CSAIL

Synthetic biology analogy: bio-bots

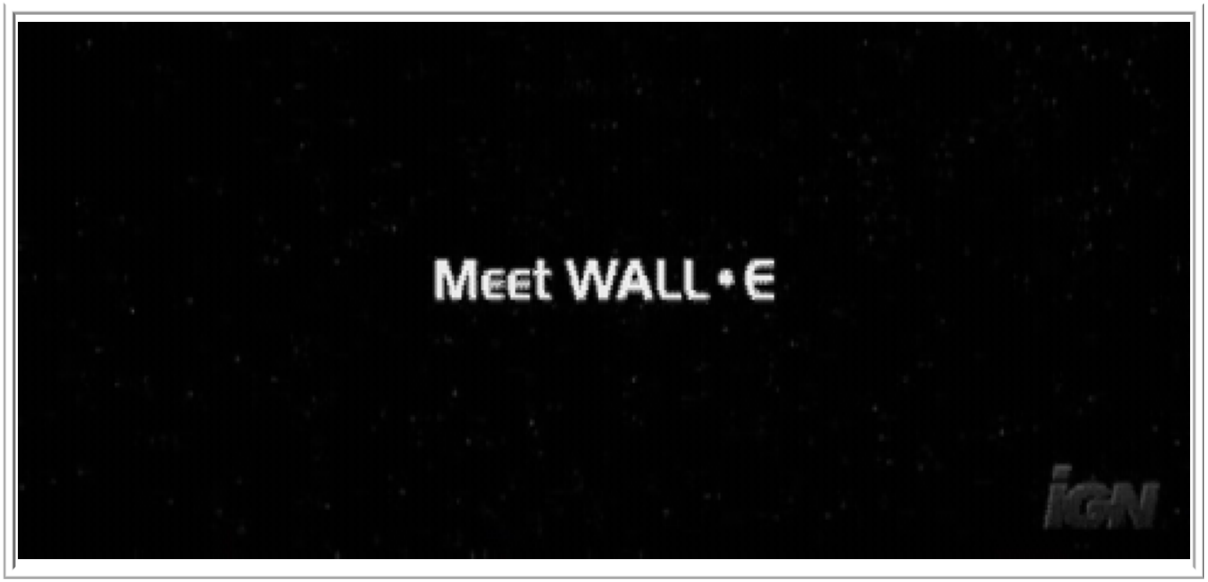


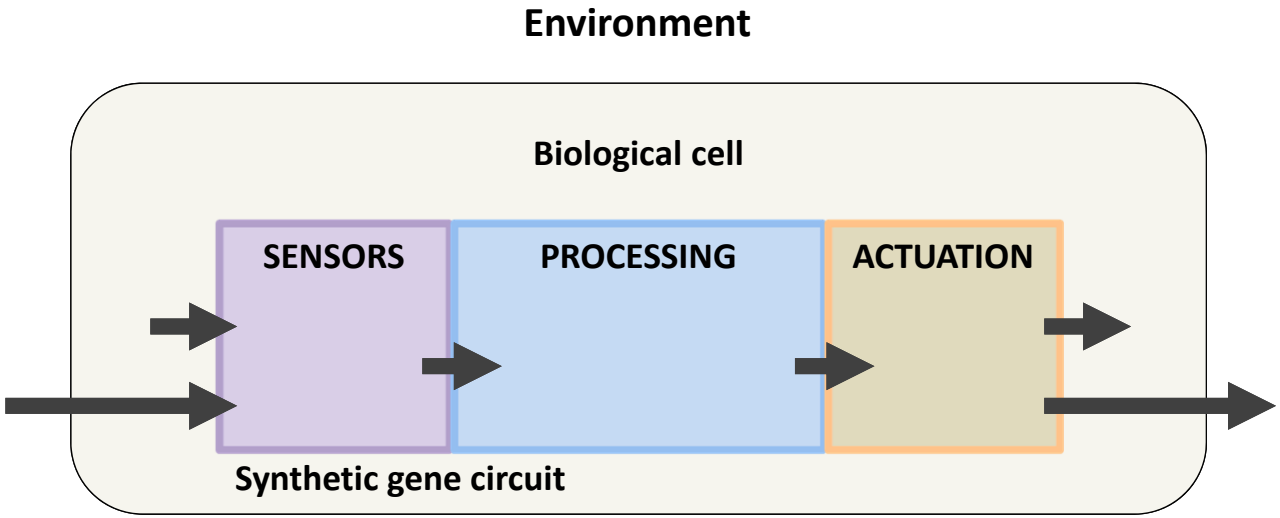
Synthetic biology analogy: bio-bots



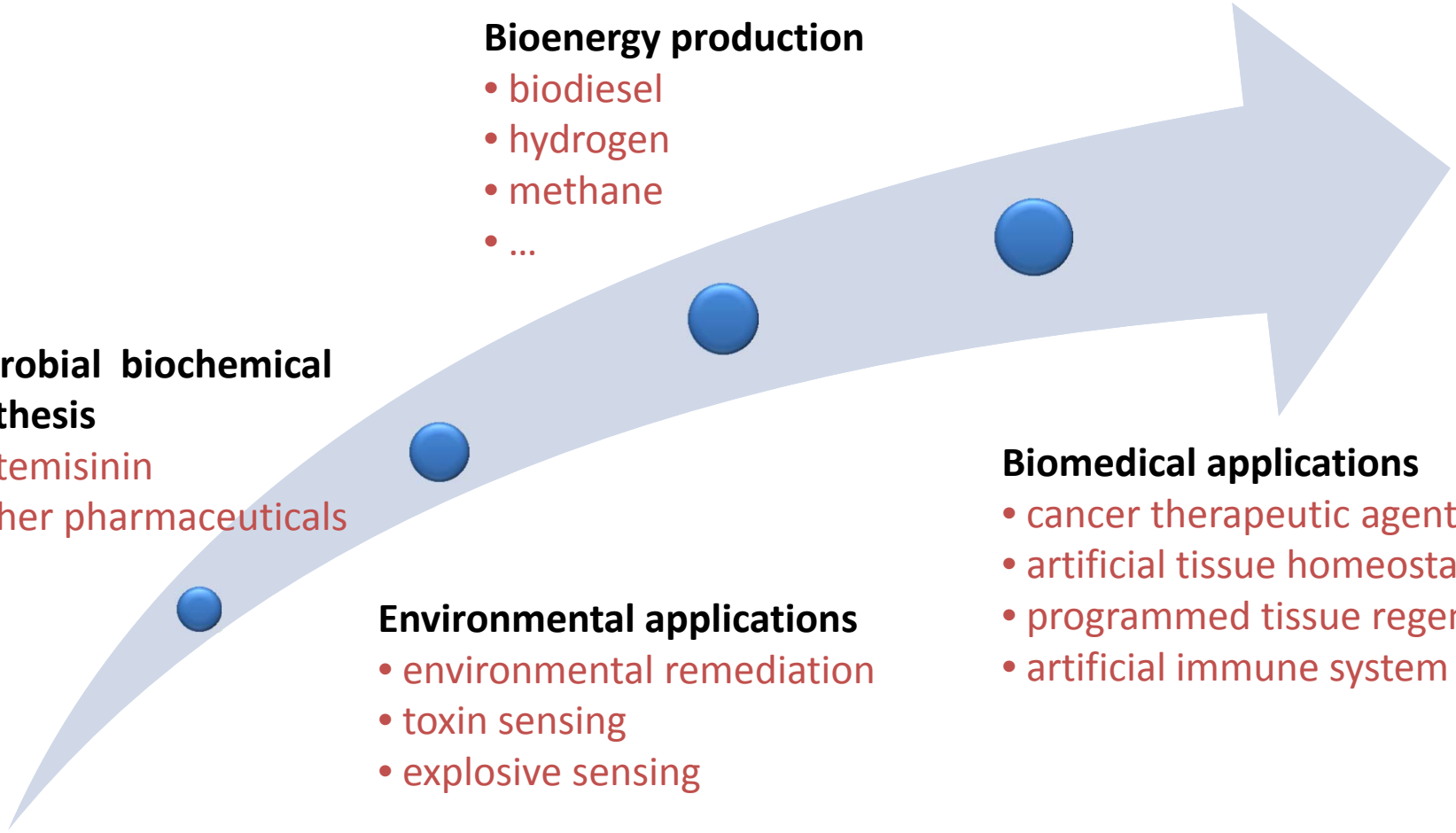


Synthetic biology analogy: bio-bots





Synthetic biology applications



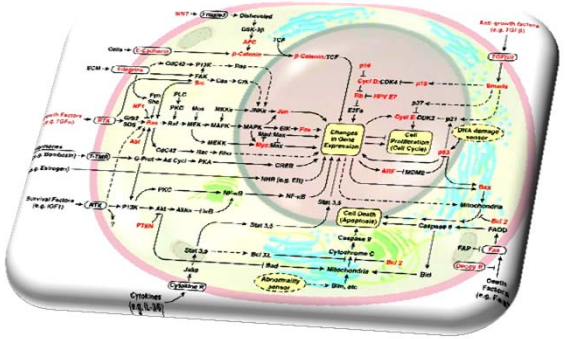
Microbial biochemical synthesis
- artemisinin
- other pharmaceuticals

Bioenergy production
- biodiesel
- hydrogen
- methane
- …

Environmental applications
- environmental remediation
- toxin sensing
- explosive sensing

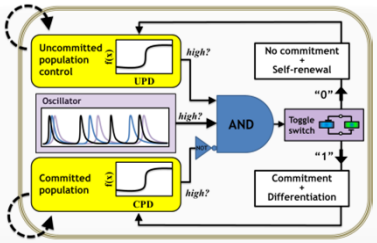
Biomedical applications
- cancer therapeutic agents
- artificial tissue homeostasis
- programmed tissue regeneration
- artificial immune system

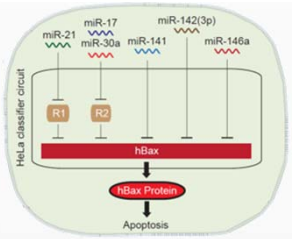
From idea to implementation



Hierarchical organization

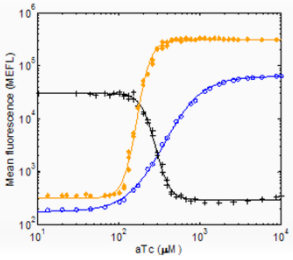
Applications

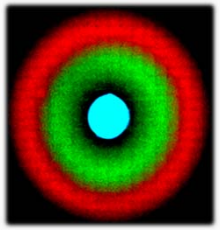




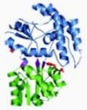
System integration

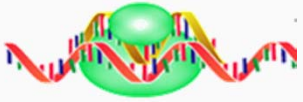
Modules



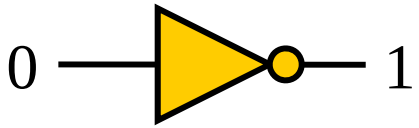


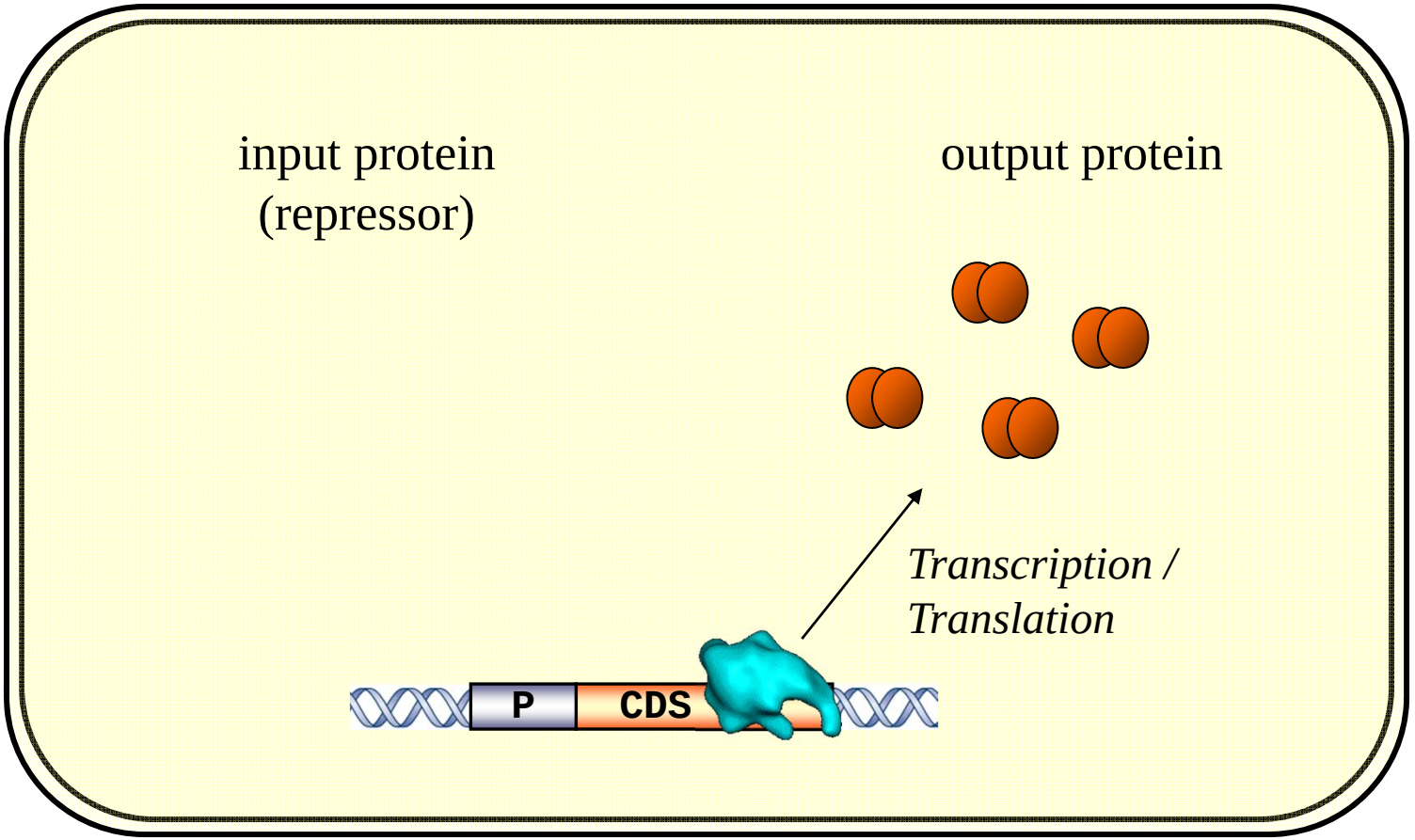
Genetic parts



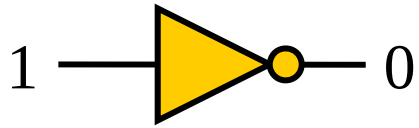


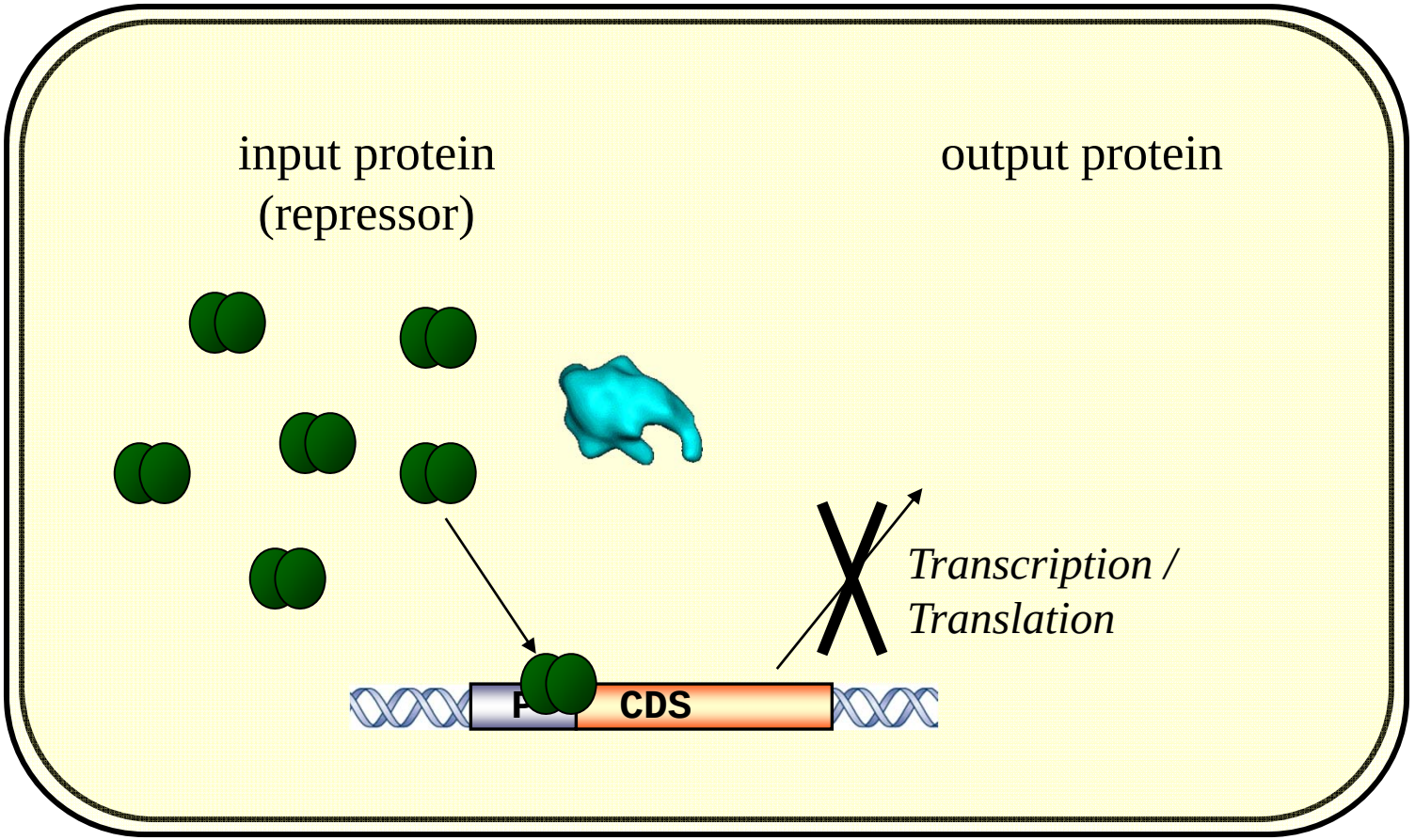
Genetic Building Block – Digital Inverter



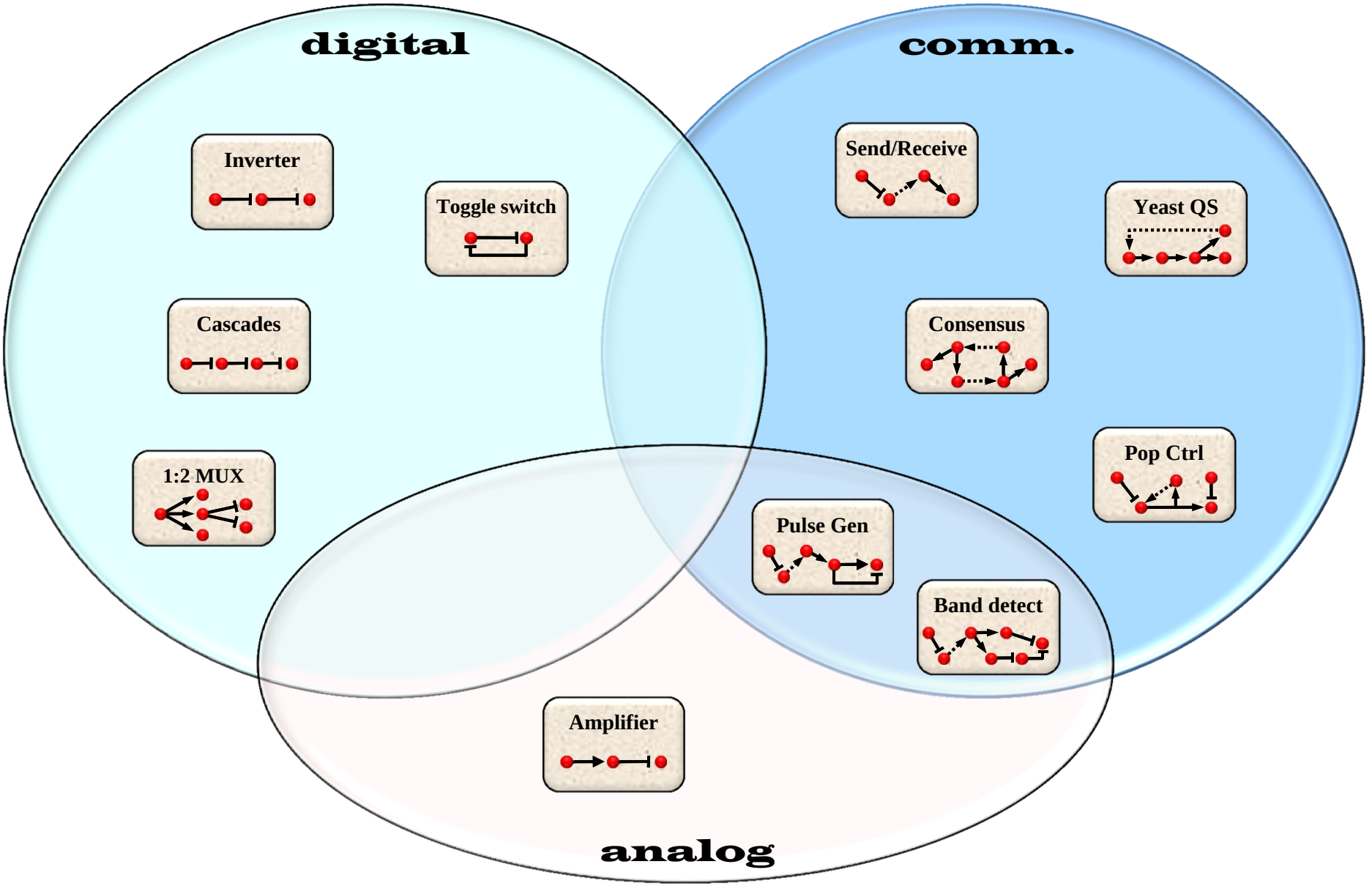


Genetic Building Block – Digital Inverter



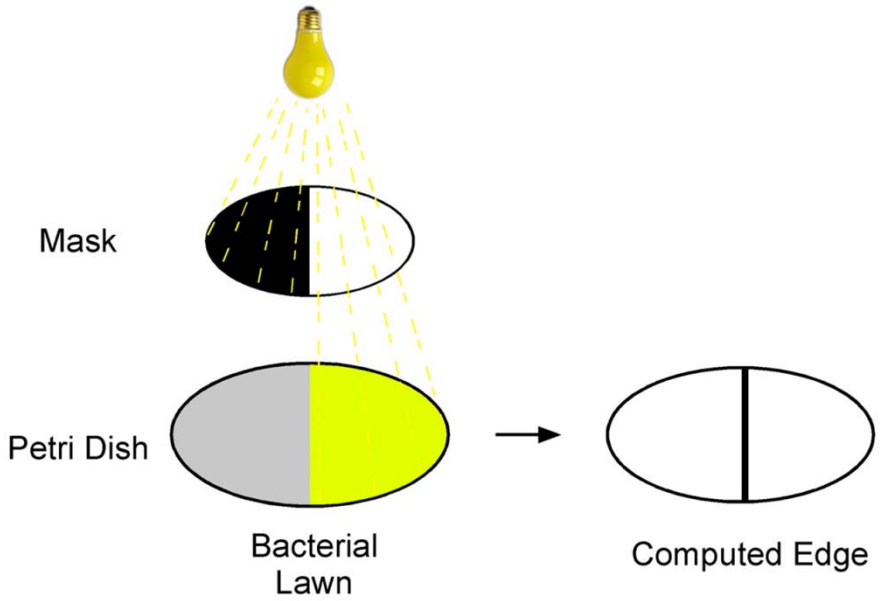


Modules implemented in Weiss lab



Genetic Edge Detector

Goal: Implement a parallel edge detection algorithm wherein each bacterium within a population functions as an independent signal processor, and the population cooperates to find the edges.

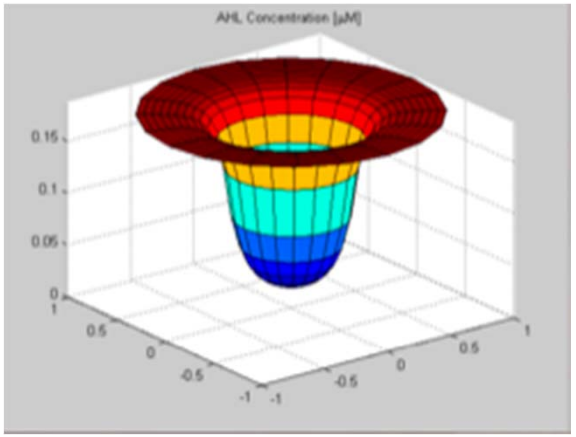


A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1273. Figure 1A

20.109 / System Engineering Module

<u>Our goal:</u> Understand, analyze, and improve edge detector

<u>Specifically:</u> One technical aspect of the presentation today is incorrect (from paper). The goal of our 20.109 module is to fix this problem!



Experimental Goals

Design: DNA modification

- Sub-clone new DNA
- Express in bacteria
- Characterize new system

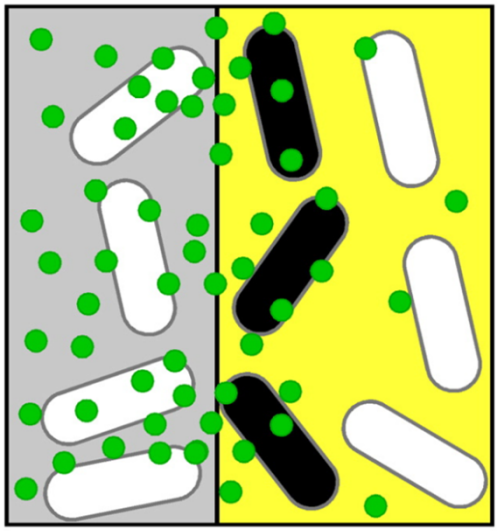
Lab+Analytical Skills

- Culture bacteria
- Make and analyze DNA
- Measure enzyme levels
- Model/make predictions
- Explore modular composition

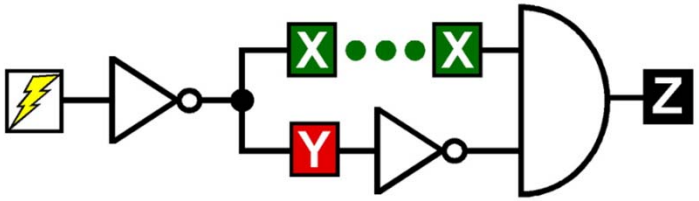
Genetic Edge Detection Algorithm

- Pseudocode:

```
IF NOT (light)
    produce signal
IF signal AND NOT (NOT light)
    produce pigment
```



- Produce signal – generate diffusible communication signal
- Produce pigment – produce black pigment

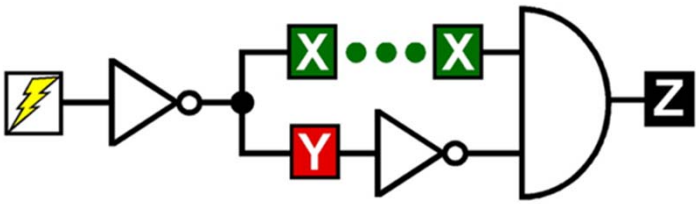


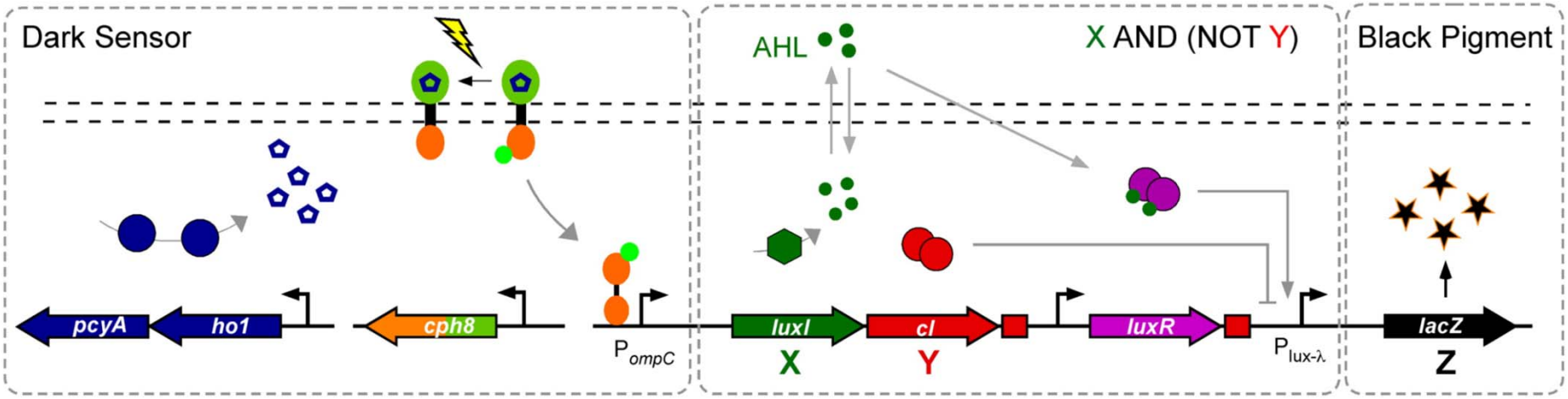
X	Y	Z
0	0	0
1	0	1
0	1	0
1	1	0

A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1273. Figure 1B, 1C

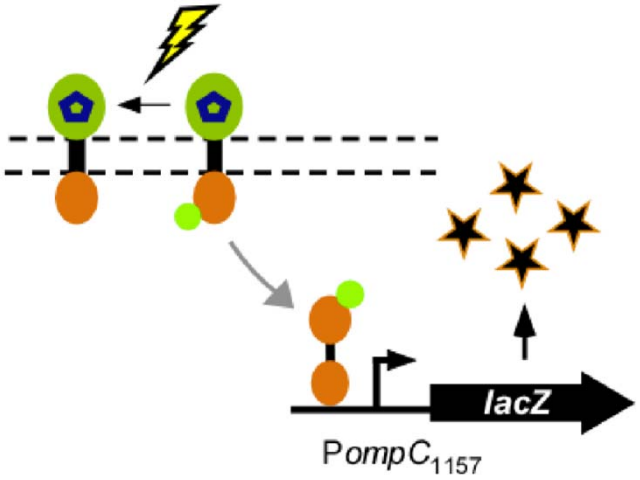
Genetic Edge Detection Algorithm

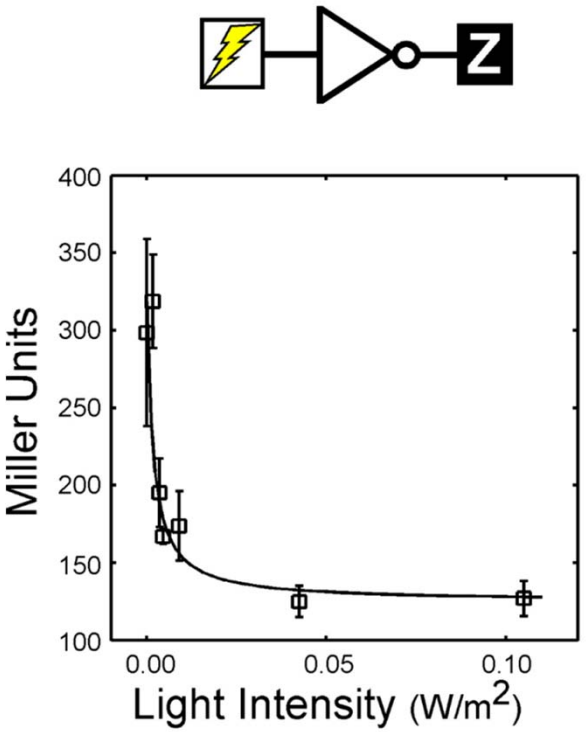
I. Photographic Bacteria using Dark Sensor
II. Cell-Cell Communication
III. Inverter
IV. Full Logic Function

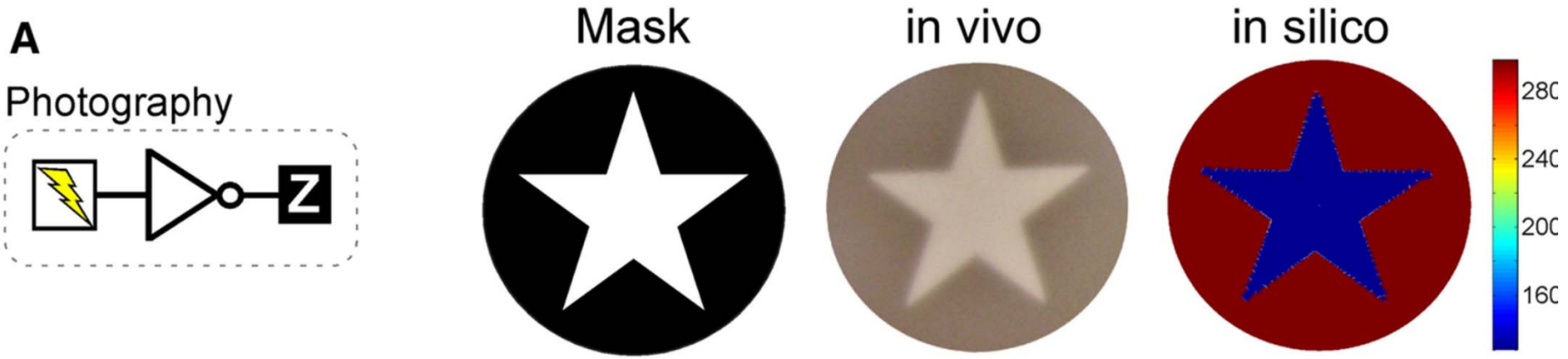




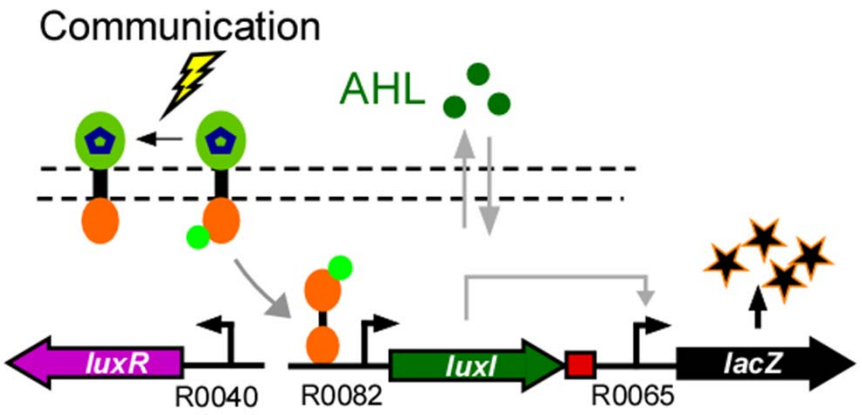
Dark Sensor

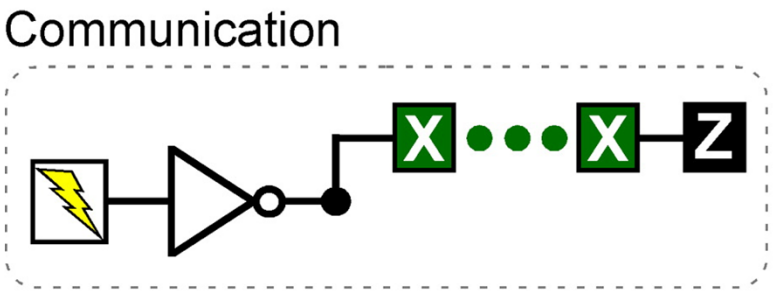

lacZ
PompC1157


Z
400
350
300
250
200
150
100
Miller Units
0.00
0.05
0.10
Light Intensity (W/m2)


A
Photography
Z
Mask
in vivo
in silico
280
240
200
160

II. Cell-cell Communication

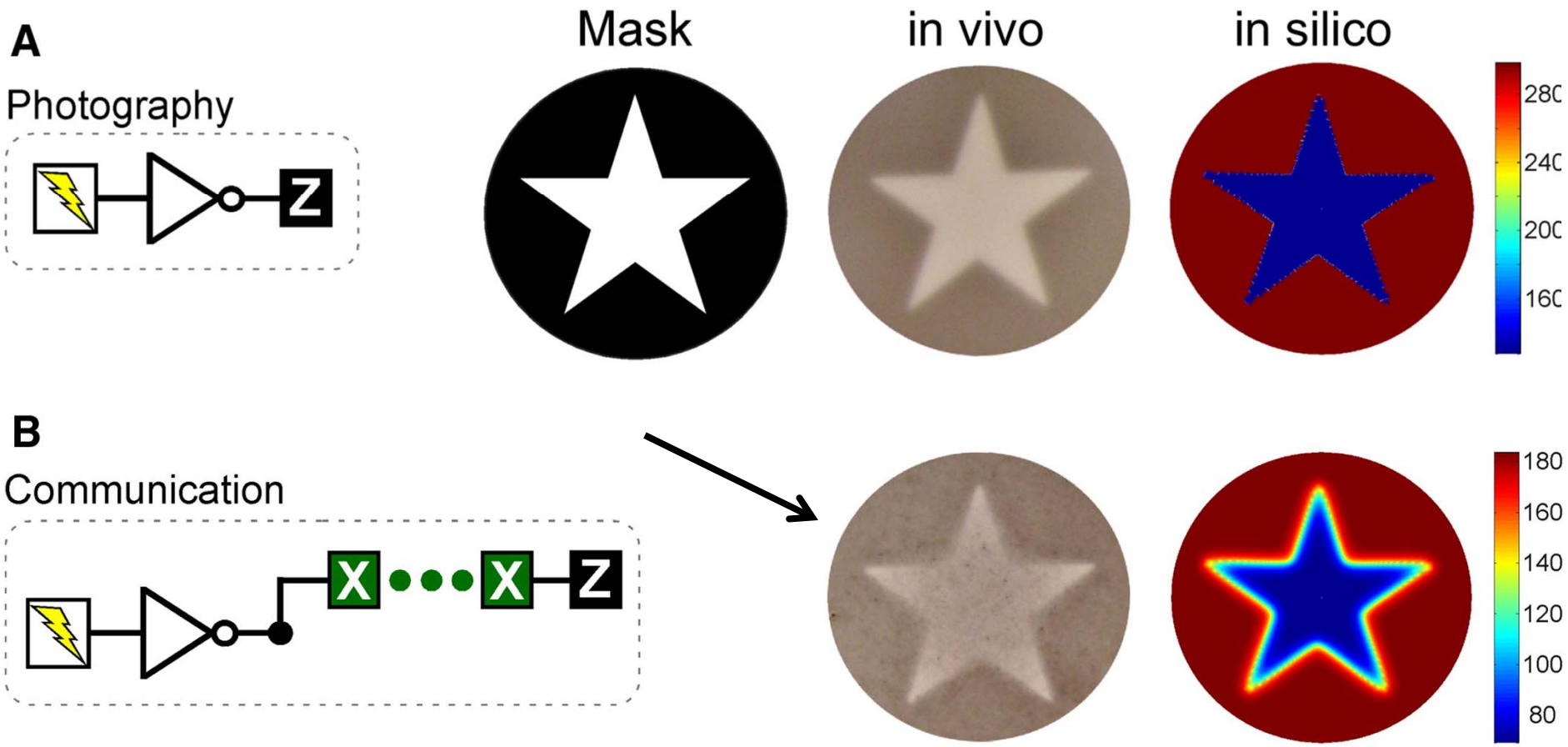




- Communicate using *V. fischeri* quorum sensing system
- Dark
  → transcription of *luxI*
  → membrane diffusible compound 3-oxohexanoyl-homoserine lactone (AHL)
- AHL binds constitutively expressed LuxR
  →β-galactosidase
- Additional blurring component

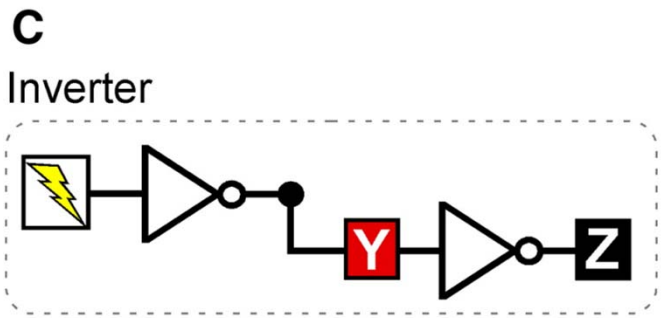
A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3B

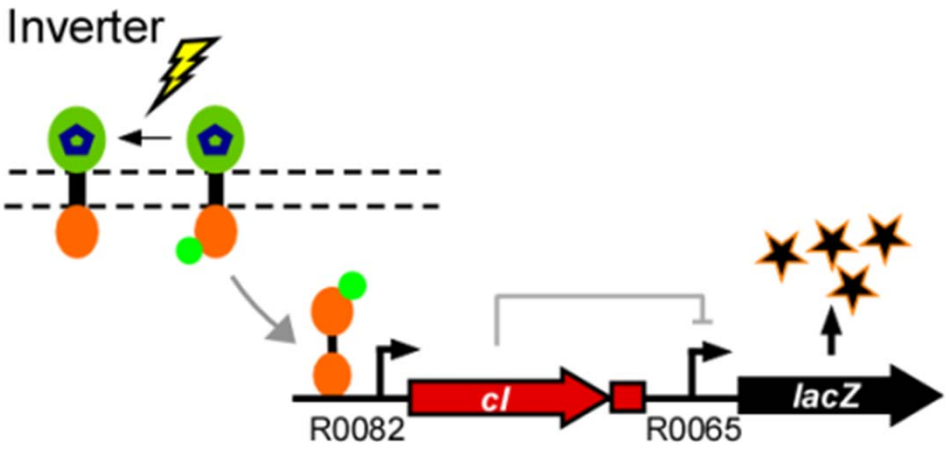
II. Cell-cell Communication



A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3B

III. Inverter

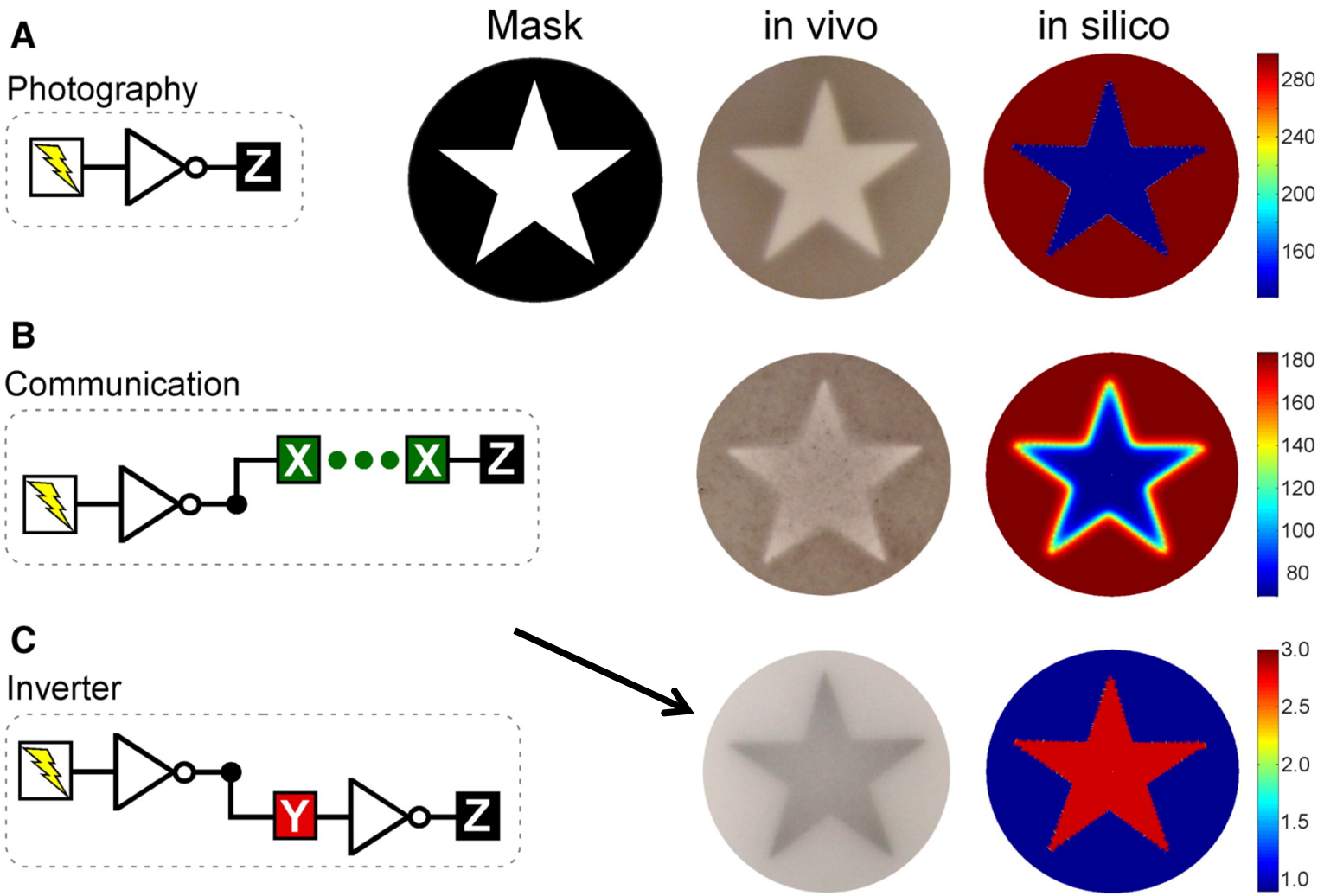




- B-gal expressed only where
  AHL AND light
  or more accurately
  AHL AND NOT (NOT light)
- NOT function = genetic inverter
- *cI* gene from phage λ → dimeric transcriptional repressor turns OFF the output promoter when the input promoter is ON
- Negative bacterial photograph

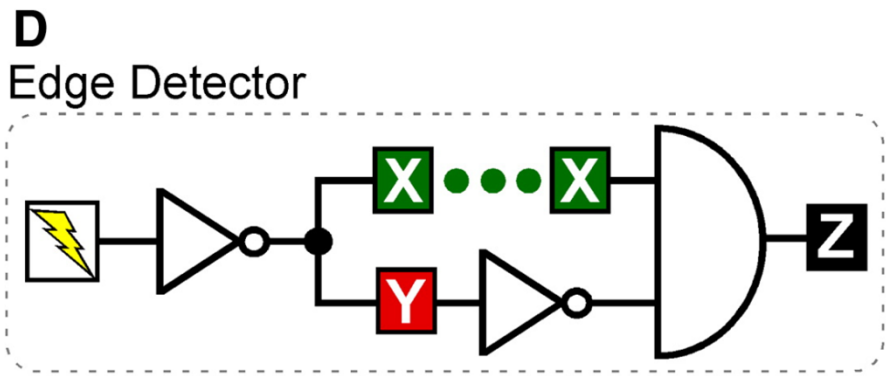
A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3C

III. Inverter

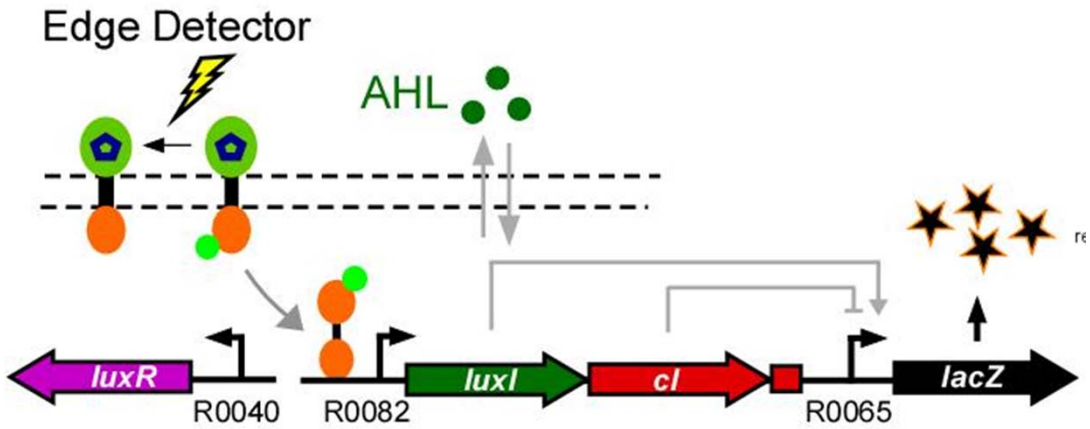


A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3C

IV. Full Logic Function

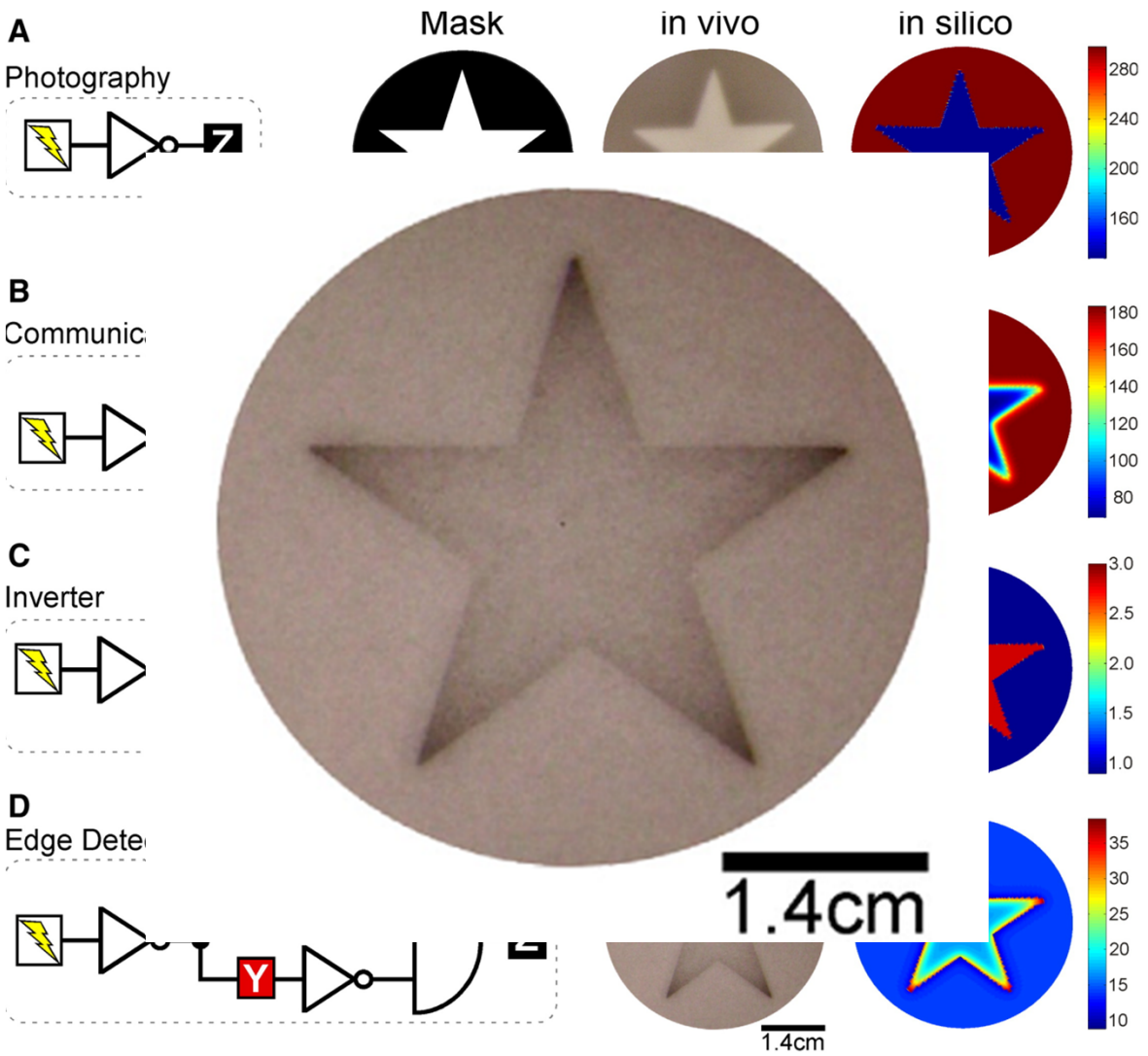


- AHL AND NOT (NOT light)
- Two-input promoter $P_{lux\text{-}\lambda}$
  - activated by AHL/LuxR
  - dominantly repressed by CI



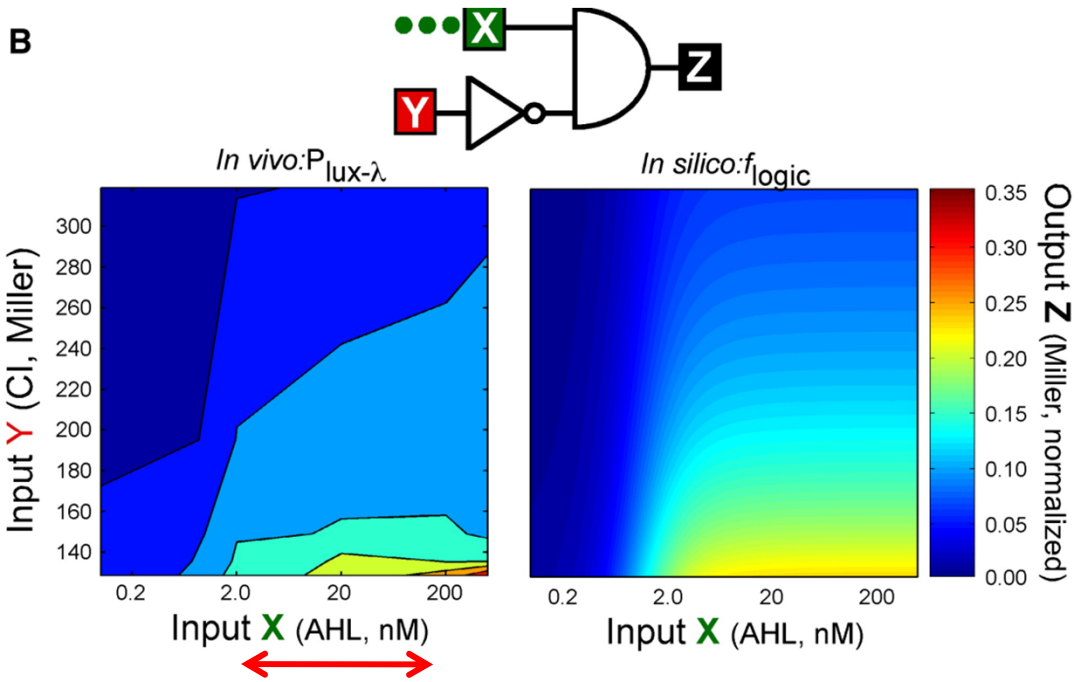
A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3D

IV. Full Logic Function



A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1275. Figure 3

Logic Transfer Function X AND (NOT Y)



$$f_{\text{logic}}(u_1, u_2) = \frac{(c_0 + c_1 f_{\text{Lux}})}{1 + c_0 + c_1 f_{\text{Lux}} + c_2 f_{\text{CI}}^n + c_1 c_2 f_{\text{Lux}} f_{\text{CI}}^n}$$

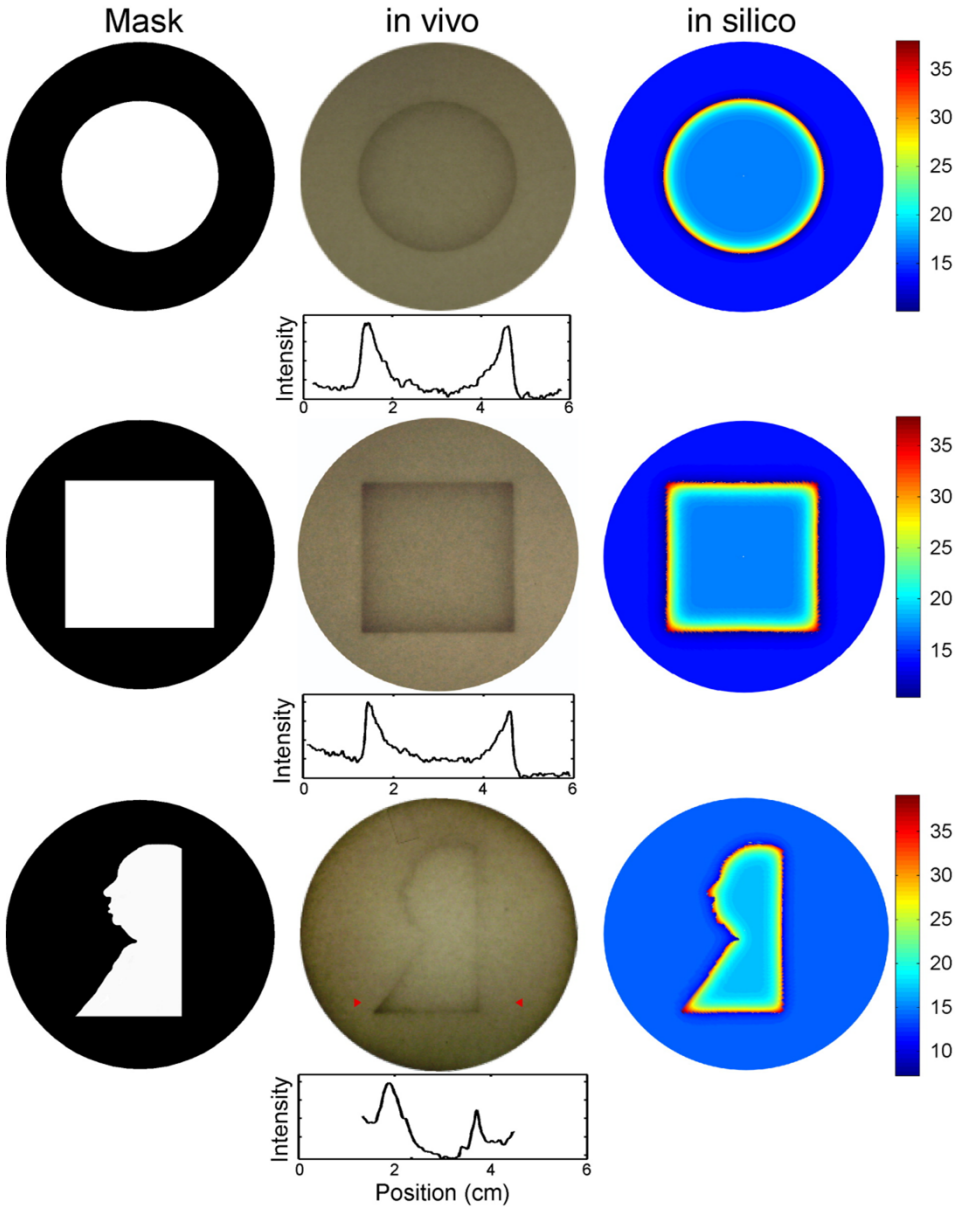
- Constitutively expressed *luxR* gene
- Exogenously vary AHL and light
- Measure β-gal activity

→two-dimensional transfer function of $P_{lux\text{-}\lambda}$

– as a function of [AHL] (u_1) and [CI] (u_2)

A synthetic genetic edge detection program. Tabor JJ, Salis HM, Simpson ZB, Chevalier AA, Levskaya A, Marcotte EM, Voigt CA, Ellington AD. **Cell.** 2009 Jun 26;137(7):1274 Figure 2B

Edge Detection of Complex Patterns



Next

- Lab exercises
  - Understand problem with edge detector
  - Design a fix by the second lab session