Synthetic Biology = design and engineering of biological systems that aren't found in nature

Why would we want to do this?

- Want to understand natural systems. One of the best ways to understand a system is to change it or make new, related ones

- To fully "understand" a system, we should be able to predict the outcome when we change the system

- For molecular biology, this means:
  - designing new gene circuits and networks
  - modeling the designed systems & predicting their properties
  - making & testing the designs
  - updating our understanding from the model/test agreement

Engineers often look at biological systems & think that the systems are equivalent to electronic circuits

e.g,

fluorescent proteins	light bulbs or LEDs
transcription factors	transistors or logic gates
repressors	NOT gates
activators	OR/AND gates
polymerases (transcriptional machinery)	batteries

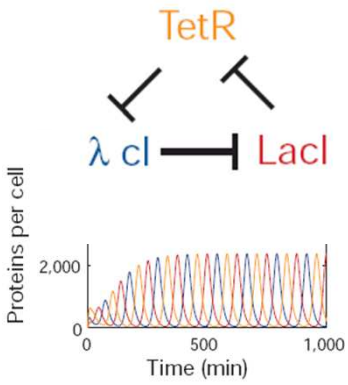
and so on...

Are they right?

→ raises the possibility that biological parts (genes, proteins, etc.) could be combined using the rules established for analog/digital circuits

The Repressilator = engineered genetic circuit designed to make bacteria glow in a oscillatory fashion = "repressor" + "oscillator"

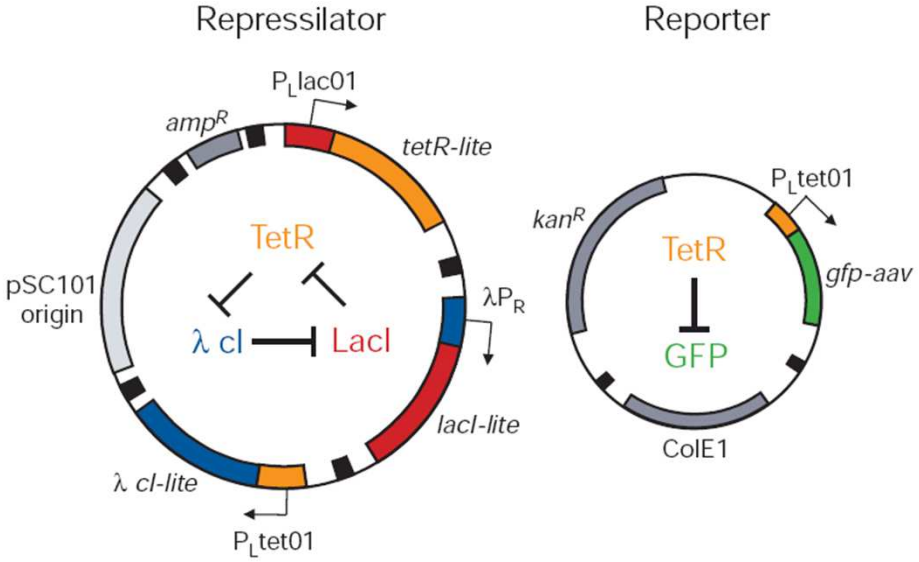
Transcriptional repressors



Green fluorescent protein

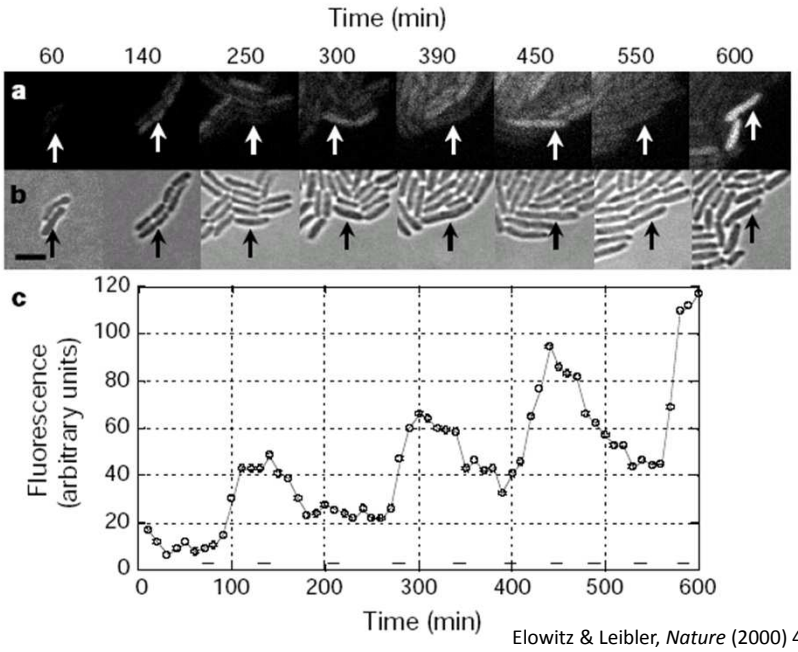
Elowitz & Leibler, *Nature* (2000) 403:335-8

The Repressilator = engineered genetic circuit designed to make bacteria glow in a oscillatory fashion



Elowitz & Leibler, *Nature* (2000) 403:335-8

The repressilator in action...



Elowitz & Leibler, *Nature* (2000) 403:335-8

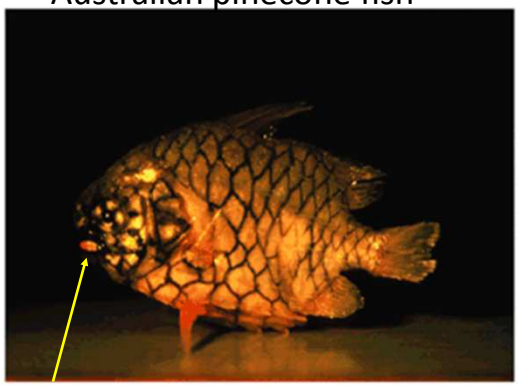
What other kinds of circuits can be built? First, we need some more parts!

Some of the other parts available include:

- various sensors
  - light, dark, heat, cold
- more switches, logic gates
  - more repressors, activators
- parts for intracellular communication
  - helpful if cells could tell each what condition they're in
    → **quorum sensing**
- parts for signaling the output of circuits
  - fluorescent & luminescent proteins

Bioluminescence – occurs when bacteria are at high density
→ bacteria communicate in order to establish their density

Australian pinecone fish



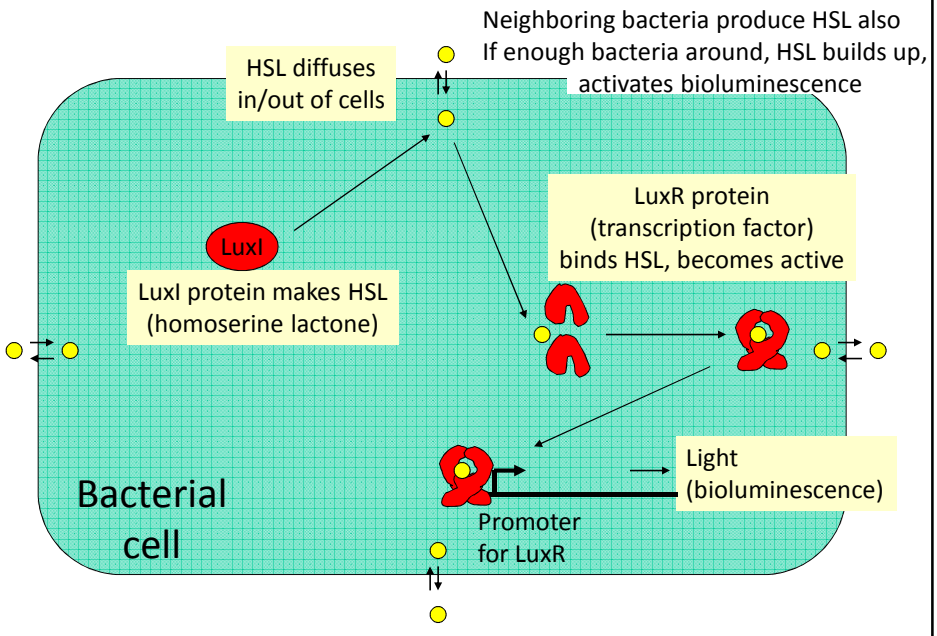
~10^{10} Vibrio bacteria/ml fluid
Fish uses to hunt for prey

Hawaiian bobtail squid

~10^{11} Vibrio bacteria/ml fluid in light organ in squid mantle
Squid uses for disguise (light shines downward, looks like moonlight)

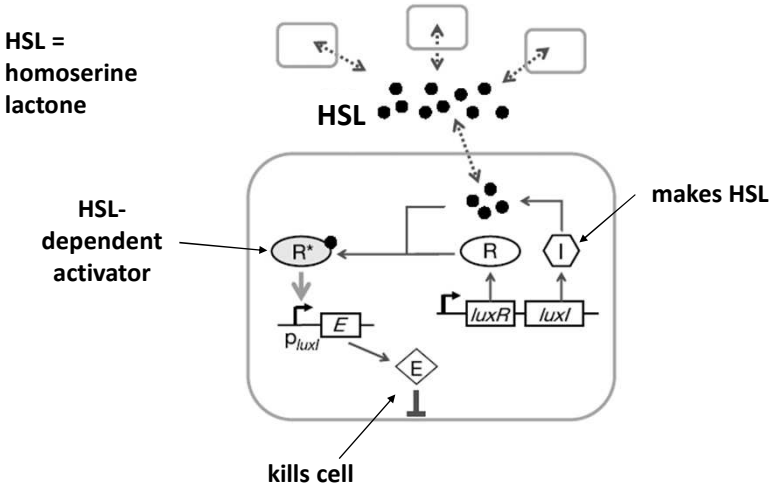
Nature Reviews Molecular Cell Biology **3**; 685-695 (2002)

Quorum sensing: chemical-based bacterial communication



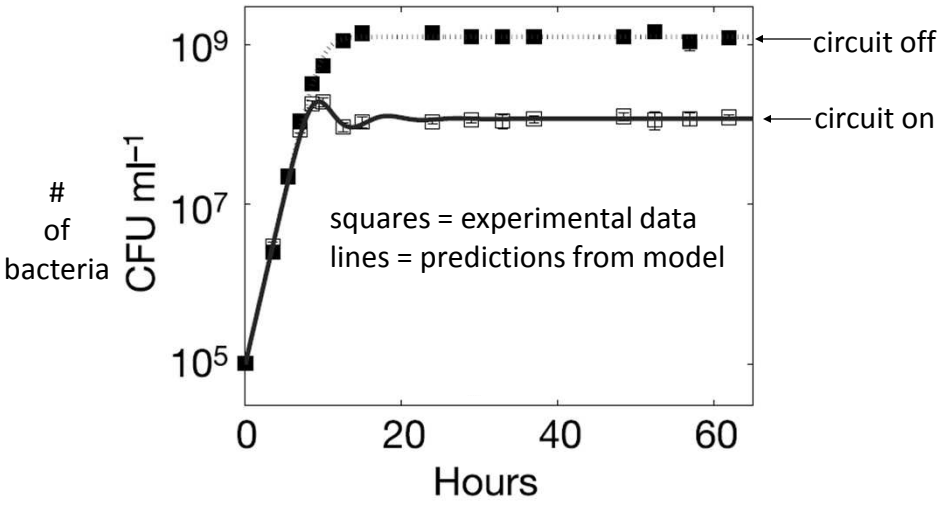
An application of quorum sensing

Programming population control into bacteria with a simple designed circuit



You, Cox, Weiss, Arnold, *Nature* (2004)

& the engineered circuit works ...



You, Cox, Weiss, Arnold, *Nature* (2004)

The behaviour can be predicted with a simple model

rate of cell growth

$$\frac{dN}{dt} = kN(1 - N/N_m) - dEN$$

cell growth rate

cell death rate

amount of killer protein

rate of killer protein production

$$\frac{dE}{dt} = k_E A - d_E E$$

amount of HSL

killer protein synthesis rate

killer protein degradation rate

rate of HSL production

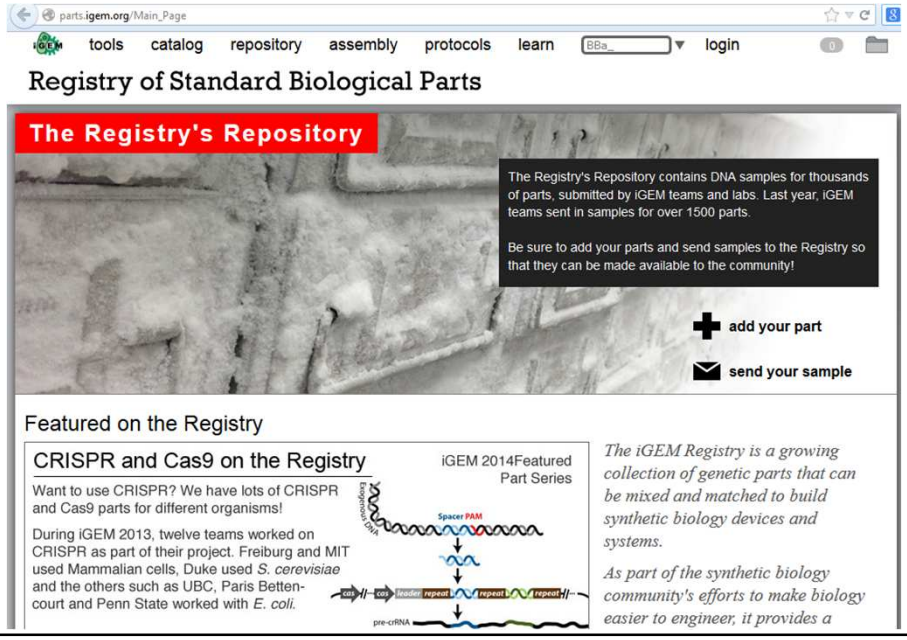
$$\frac{dA}{dt} = v_A N - d_A A$$

HSL synthesis rate

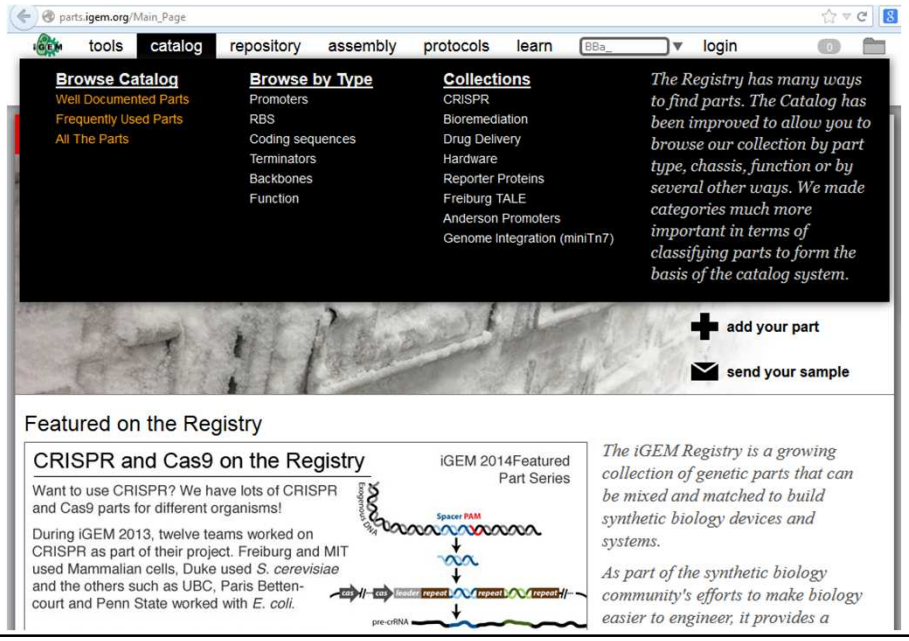
HSL degradation rate

You, Cox, Weiss, Arnold, *Nature* (2004)

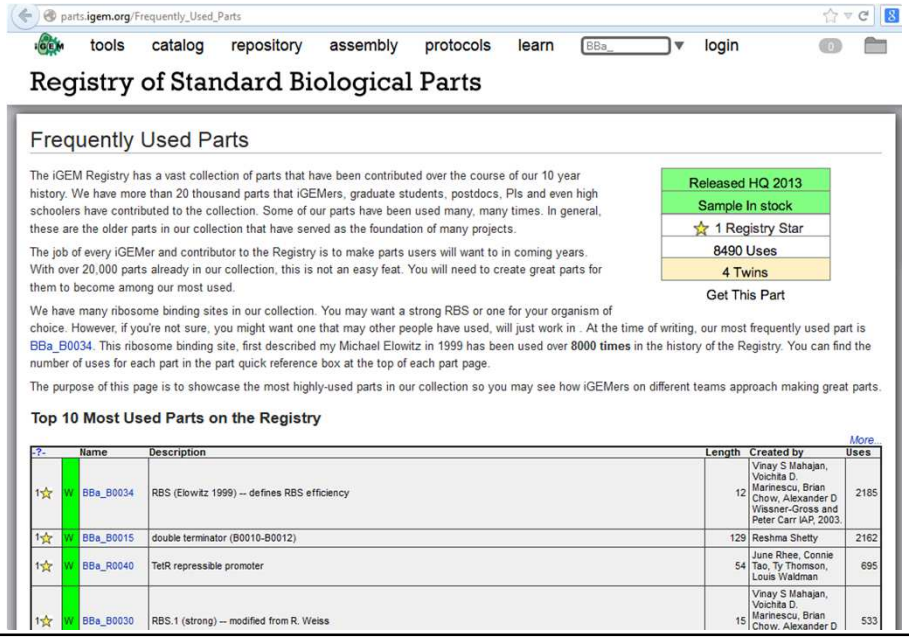
Standardization of parts: the iGEM "BioBricks" project



Standardization of parts: the iGEM “BioBricks” project



Standardization of parts: the iGEM “BioBricks” project



iGEM: A synthetic biology contest

(from iGEM's web site)

Can simple biological systems be built from standard, interchangeable parts and operated in living cells? Or is biology simply too complicated to be engineered in this way?

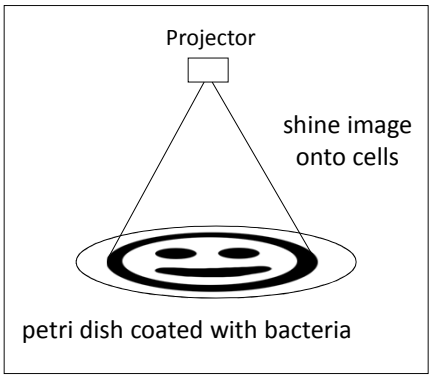
iGEM's broader goals include:

- To enable systematic engineering of biology
- To promote open & transparent development of tools for engineering biology
- To help construct a society that can productively apply biological technology

2004: MIT, UT, Princeton, Boston University, Cornell
2005: 13 teams (the above + UK, Germany, more...)
2006: 32 teams, incl. Japan/Latin America/Korea/India/more Europe

54 teams in 2007, 84 teams in 2008, 112 teams in 2009, 130 teams in 2010, 165 teams in 2011, and 245 teams in 2012 and 2013…

UT's 2004/2005 iGEM project – build bacterial edge detector



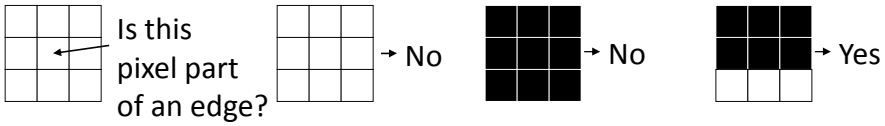
Original image

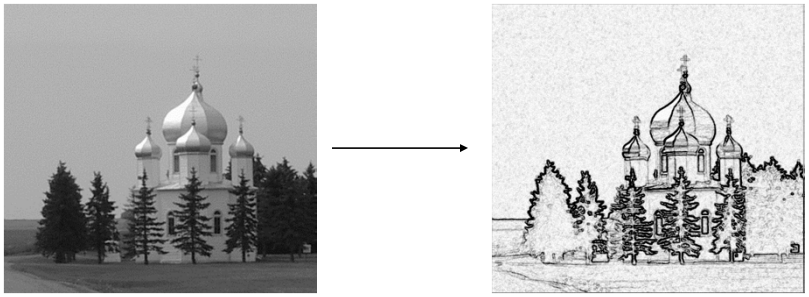
Cells luminesce along the light/dark boundaries

Adapted from Zack Simpson

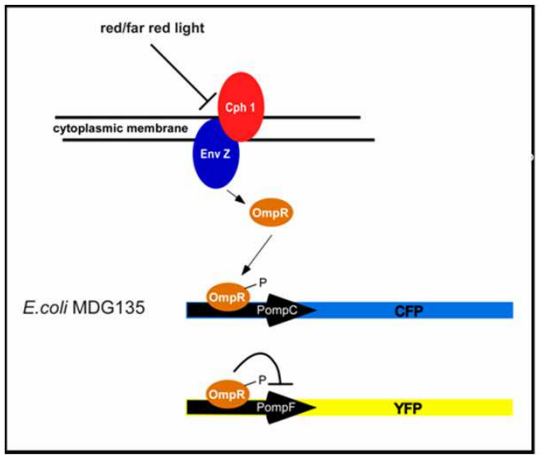
How does edge detection work in principle?

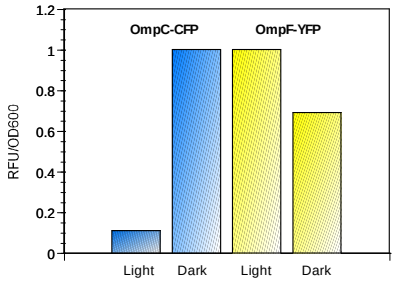
A computer might visit each pixel in turn, and check to see if it is bordered by both black & white pixels. If yes, highlight the pixel.





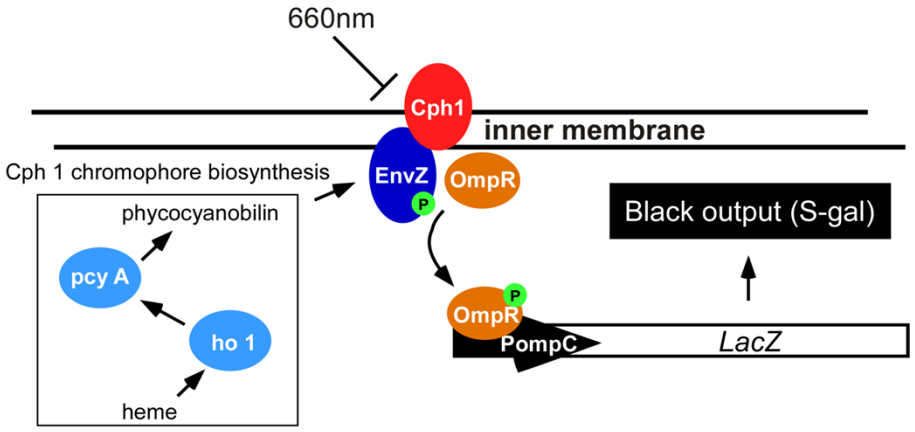
Light-dependent gene expression



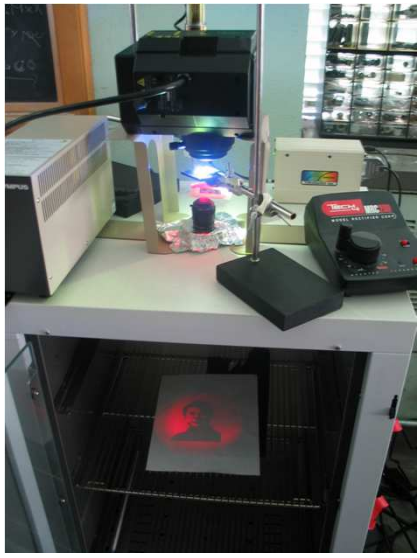


Levskaya *et al. Nature*, 438(7067):441-2 (2005)

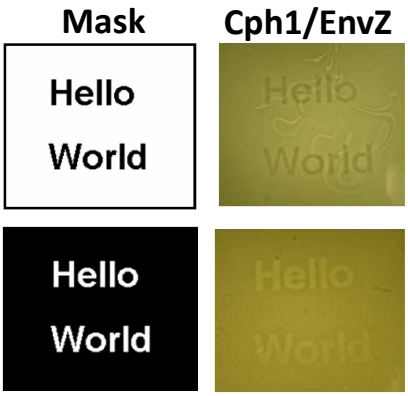
Bacterial photography



Levskaya *et al. Nature*, 438(7067):441-2 (2005)



"Light cannon" developed by Aaron Chevalier, UT undergraduate



Levskaya *et al. Nature*, 438(7067):441-2 (2005)

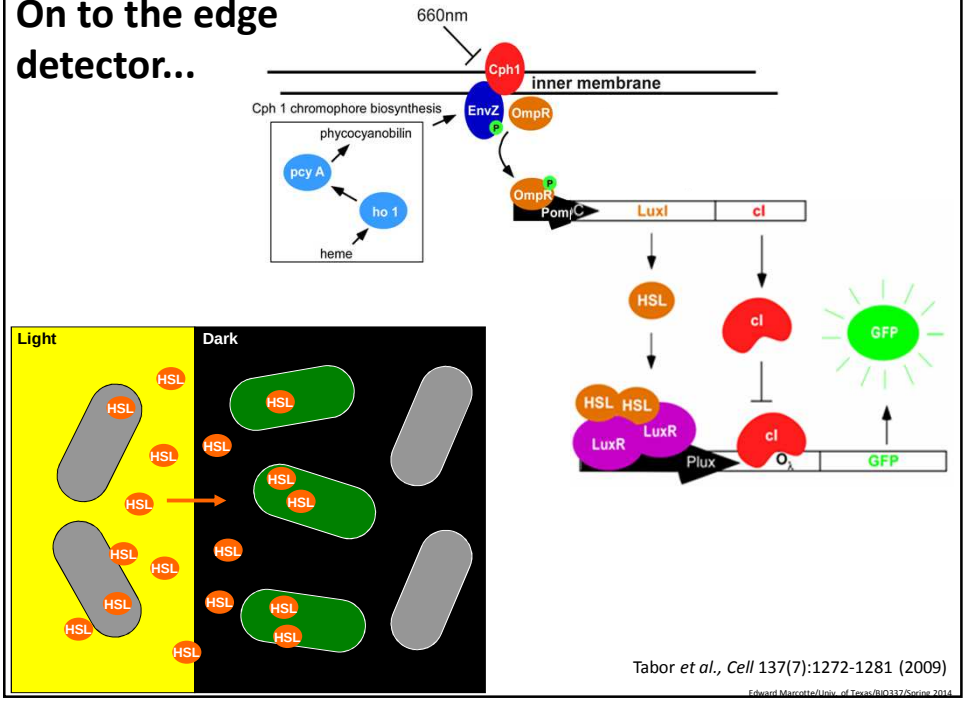
The first bacterial photograph (coliroid?)...

Levskaya *et al. Nature*, 438(7067):441-2 (2005)

Escherichia darwinia

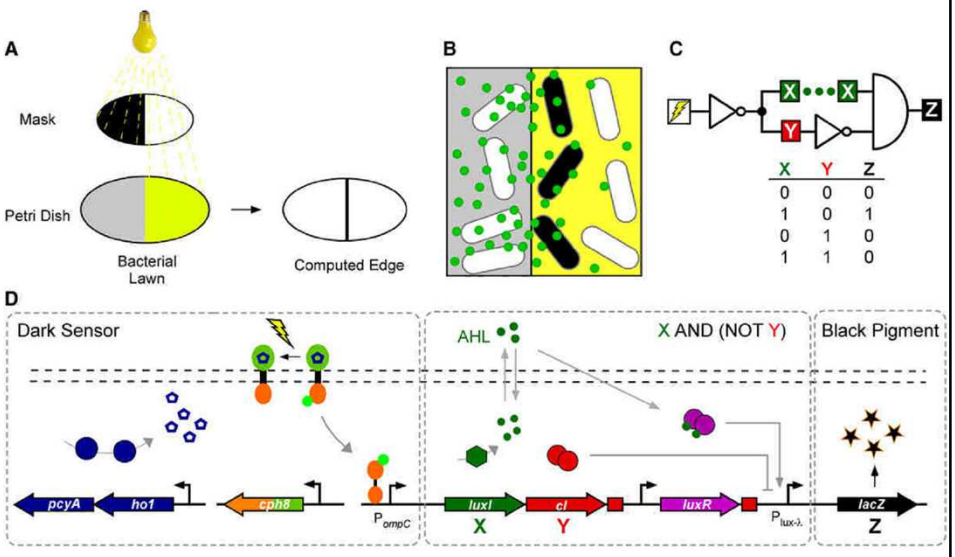
Image: Aaron Chevalier

On to the edge detector...



Tabor *et al.*, *Cell* 137(7):1272-1281 (2009)

The edge detector circuit in more detail



Tabor *et al.*, *Cell* 137(7):1272-1281 (2009)

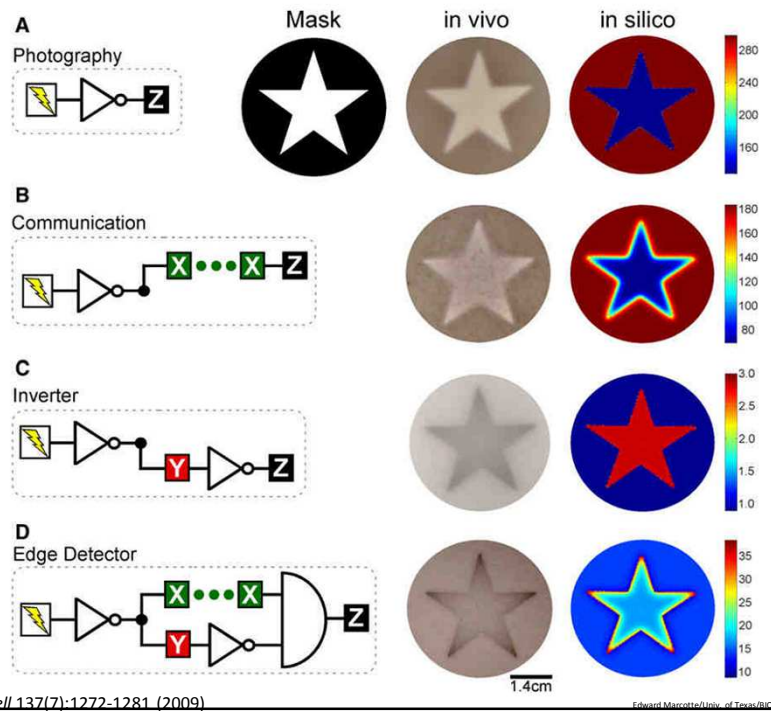
It works!

Projected Mask

Photo strain

Edge detector strain

Tabor *et al., Cell* 137(7):1272-1281 (2009)



Tabor *et al., Cell* 137(7):1272-1281 (2009)

UT's 2012 iGEM project – build caffeine biosensor

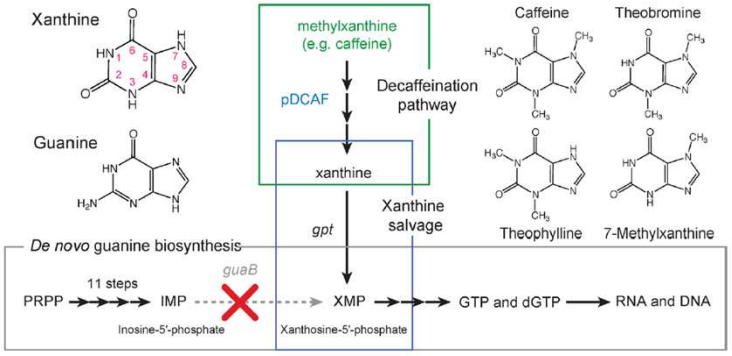
ACS SyntheticBiology — Letter — pubs.acs.org/synthbio

Decaffeination and Measurement of Caffeine Content by Addicted *Escherichia coli* with a Refactored *N*-Demethylation Operon from *Pseudomonas putida* CBB5

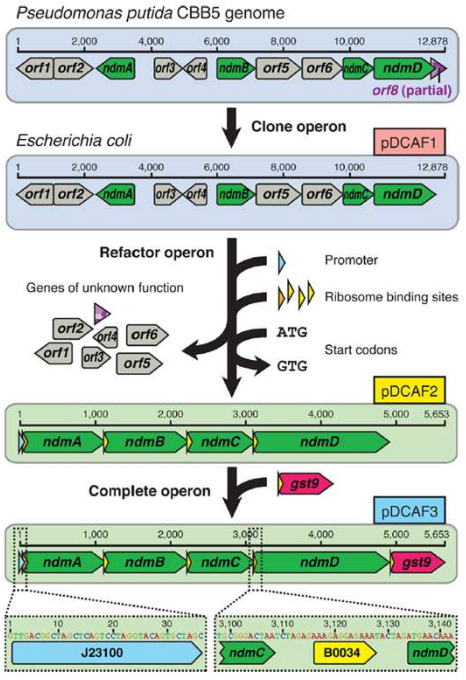
Erik M. Quandt,[†] Michael J. Hammerling,[†] Ryan M. Summers,[‡] Peter B. Otoupal,[†] Ben Slater,[†] Razan N. Alnahhas,[†] Aurko Dasgupta,[†] James L. Bachman,[†] Mani V. Subramanian,[‡] and Jeffrey E. Barrick*[,†]

Basic idea

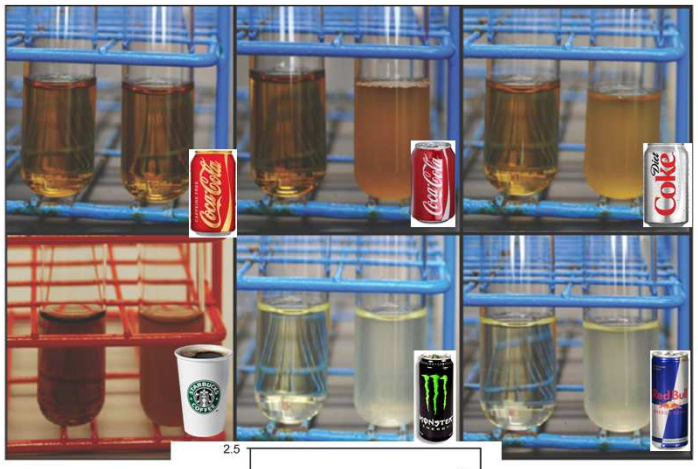
Block *de novo* guanine synthesis
Convert caffeine to xanthine
Addict *E. coli* bacteria to caffeine



ACS Synth. Biol. 2013, 2, 301–307



ACS Synth. Biol. 2013, 2, 301–307



ACS Synth. Biol. 2013, 2, 301−307