Background
Background

Individual vs. Collective

ARS/USDA
Background

**INDIVIDUAL**

**COLLECTIVE**
Background

*Betzig 2014*
GOAL

Create a synthetic cellular framework to model collective behaviors emergent from individual autonomous rules.
WHAT DOES THE SIMPLEST SYNTHETIC COLLECTIVE LOOK LIKE?

Input

individual cell response
VARIABLE

Communication circuit

community response
COORDINATED
WHAT DOES THE SIMPLEST SYNTHETIC COLLECTIVE LOOK LIKE?

individual cell response

community response

VARIABLE

COORDINATED
Individual Response

Doxycycline

rtTA

pTET

GFP
INDIVIDUAL RESPONSE

rtTA

pTET

GFP

COMMUNITY RESPONSE

rtTA

pTET

MFα

RFP

other cells
ENGINEERING CONTROLLED VARIABILITY

variable cell response

# cells
INDIVIDUAL RESPONSE

rtTA → rTAT

pTET → rtTA

GFP

COMMUNITY RESPONSE

pTET → MFα

RFP

Community Response

other cells
PROMOTER STRENGTH ALTERS INDIVIDUAL RESPONSE

Individual Response

TEF1 promoter mutants
Parts BBa_K1346000-4
Nevoigt 2006

DOXYCYCLINE

pTEF1
rtTA

mRNA

pTET
GFP

Fluorescence (A.U.)

Individual Response

GFP Fluorescence (A.U.)

H
L

0 0.03 0.06 0.09 0.3 0.6 0.9 3 6 9
doxycycline concentration (ng/μl)

LOW
HIGH

No Circuit Control
pTEF1
pTEF1(m3)
pTEF1(m6)
pTEF1(m7)
pTEF1(m10)
INDIVIDUAL RESPONSE

rtTA → rtTA

pTET → GFP

Community Response

ALPHA FACTOR

other cells

INDIVIDUAL RESPONSE

rtTA → rtTA

pTET → GFP

pTET → MFα

RFP

Individual response

Individual response

Individual response
**ALPHA FACTOR RESPONSIVE PROMOTER DRIVES RFP OUTPUT**

Parts: BBa_K1346004 - BBa_K1346013

![Graph showing mean GFP (AU) vs. [alpha factor] (nM)](image)

- pAGA1
circuit results in *individual response* (GFP) and *community response* (RFP) with *stimulus*

**INDIVIDUAL RESPONSE**

**COMMUNITY RESPONSE**

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<th>-dox</th>
<th>+dox</th>
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HOW DO WE OBSERVE COLLECTIVE BEHAVIOR?

1. Mix strains with different individual responses (GFP)
HOW DO WE OBSERVE COLLECTIVE BEHAVIOR?

1. Mix strains with different individual responses (GFP)

2. Co-culture and allow for communication

3. Measure community response level (RFP)

[Diagram showing different levels of response labeled as LOW and HIGH.]
HOW DO WE OBSERVE COLLECTIVE BEHAVIOR?

1. Mix strains with different Individual responses (GFP)

2. Co-culture and allow for communication

3. Measure community response level (RFP)
COMMUNITY CONSENSUS
COMMUNITY CONSENSUS

Individual Response

Community Response

GFP Fluorescence (AU)

RFP Fluorescence (AU)
We have built a **synthetic cellular community** of yeast that communicates and exhibits **convergent collective behaviors**.
FUTURE WORK: EXPLORING COMMUNICATION PARAMETERS

What happens if we alter the range of communication?

Youk and Lim, 2014
ALTERING RANGE OF COMMUNICATION

Dispersed

Local
ALTERING COMMUNICATION

Bar1 = a protease that degrades extracellular alpha factor

MODELING
Can reduce noise and limit range of communication
Positive feedback can be engineered by either inducing more alpha factor secretion or increased receptors.
ALTERING COMMUNICATION

Positive feedback can be engineered by either inducing more alpha factor secretion or increased receptors!

This can amplify the signal locally and contribute to population divergence.
SUMMARY

- CHARACTERIZED 15 DIFFERENT PROMOTERS
- BUILT SENSE & SECRETE CIRCUIT MEASURES INDIVIDUAL AND COLLECTIVE RESPONSES
- DEMONSTRATE COORDINATED COLLECTIVES
WE ARE MODELING COLLECTIVE CELL BEHAVIORS WITH SIMPLE ROBOTS

Kilobots – swarm robots developed by the Self-Organizing Systems Research Group at Harvard

Can send and receive signals and flash an LED to output state
WE ARE MODELING COLLECTIVE CELL BEHAVIORS WITH SIMPLE ROBOTS

GOALS:

what parameters are important for each behavior?

can we see different and new behaviors?

See wiki for more info!
We have built the library of available parts for work in yeast systems

BBa_K1346000-BBa_K1346013
PARTS REGISTRY

BIORICKS SUBMITTED

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Promoter Activity By Alpha Factor Dose

Pheromone Response Factors: 2
Biobrick Specification

ASG7: Protein that regulates signaling from a G protein beta subunit Ste4p and its relocalization within the cell, specific to mating type α-cells and induced by alpha-factor
HUMAN PRACTICES

Engaged the public at Exploratorium science museum event with 3,000 people.

Made synthetic biology and our project approachable and transparent.
HUMAN PRACTICES

Engaged the public at Exploratorium science museum event with 3,000 people

Made synthetic biology and our project approachable and transparent

Developed this into a curriculum for distribution to middle/high schools
THE UCSF & UCB TEAM

5 Abraham Lincoln High School graduates
2 UCSF iGEM alumni
2 UCB students
1 Peking iGEM alumna
• 8th year of partnership
• ~47 students
• 2 Year Biotechnology Pathway
• Mr. George Cachianes and Ms. Julie Reis
THE UCSF&UCB TEAM

- Abraham Lincoln High School graduates: 5
- UCSF iGEM alumni: 2
- UCB students: 2
- Peking iGEM alumna: 1
ACKNOWLEDGEMENTS

MENTORS

ADVISORS

Not pictured: Ben Heineike, Alain Bonny, Jonathan Asfaha, PJ Buske, Justin McLaurin
ACKNOWLEDGEMENTS
Questions?