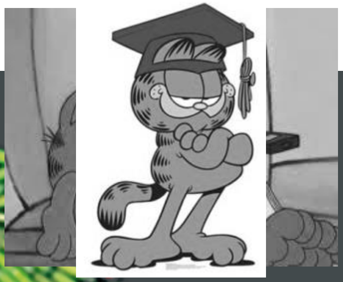
Information processing by bacterial quorum sensing systems

Ilka B. Bischofs

µCATs-Lab @ BioQuant, Heidelberg



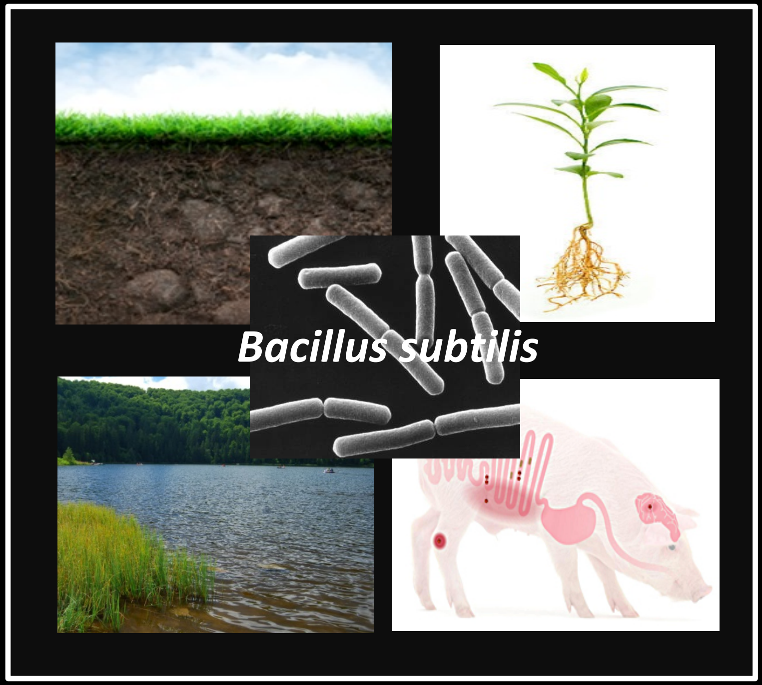
Understand

Control

Engineer

Adaptation

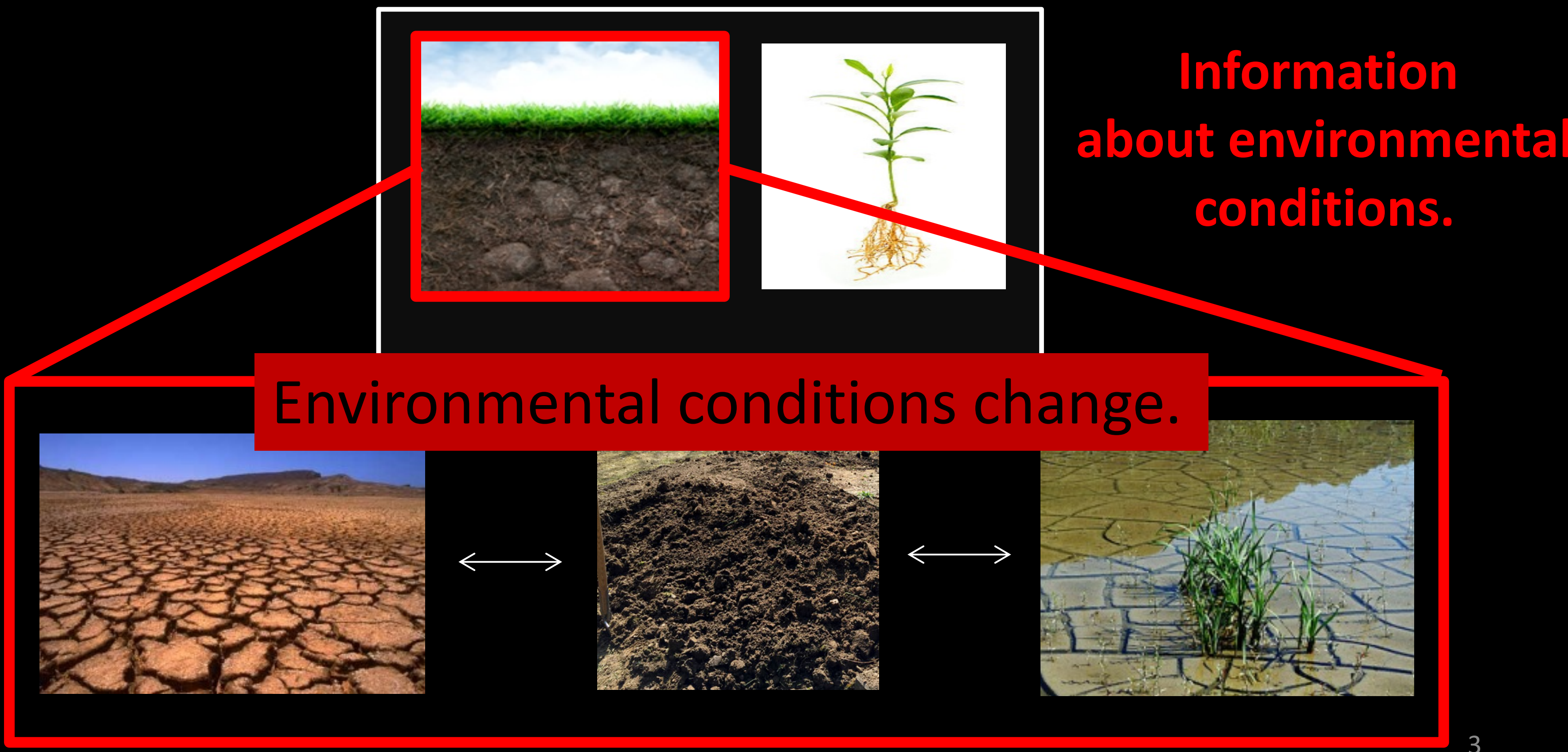
„The only constant in life is change.“



Information about the niche.

Adaptation

„The only constant in life is change.“

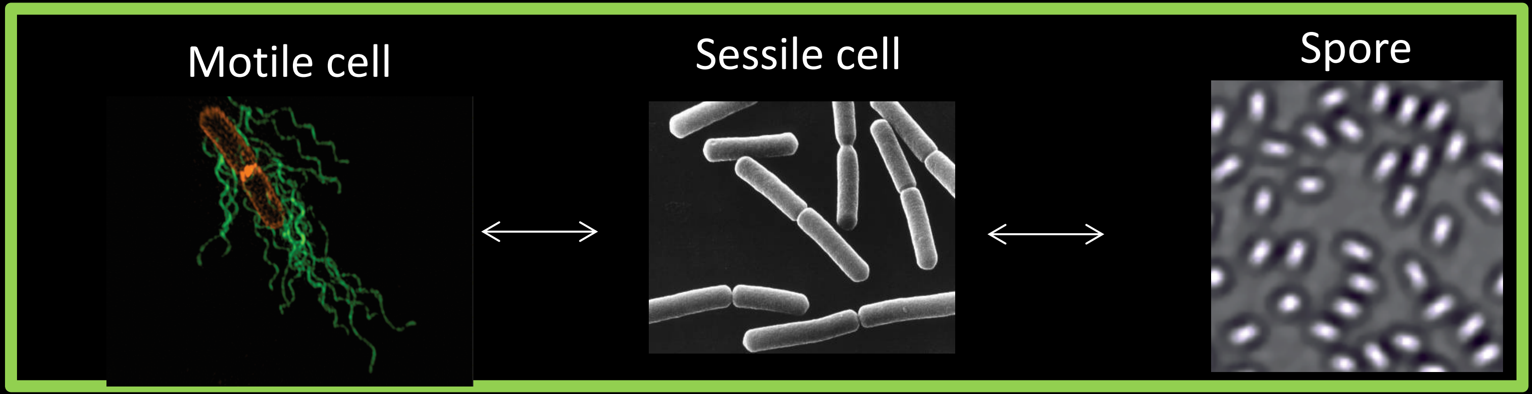


Adaptation

Environment → **Cellular state**

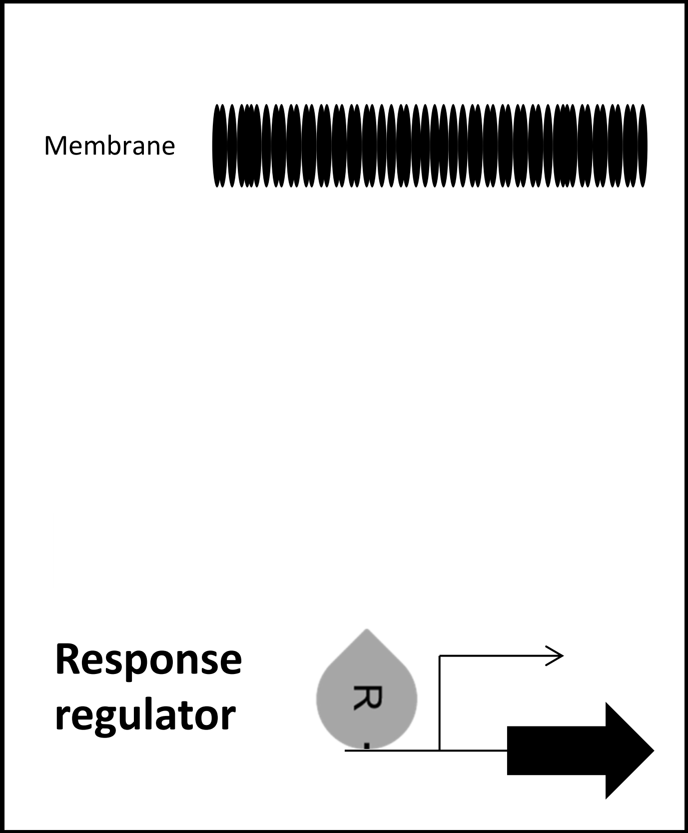
Gene expression

Cell differentiation



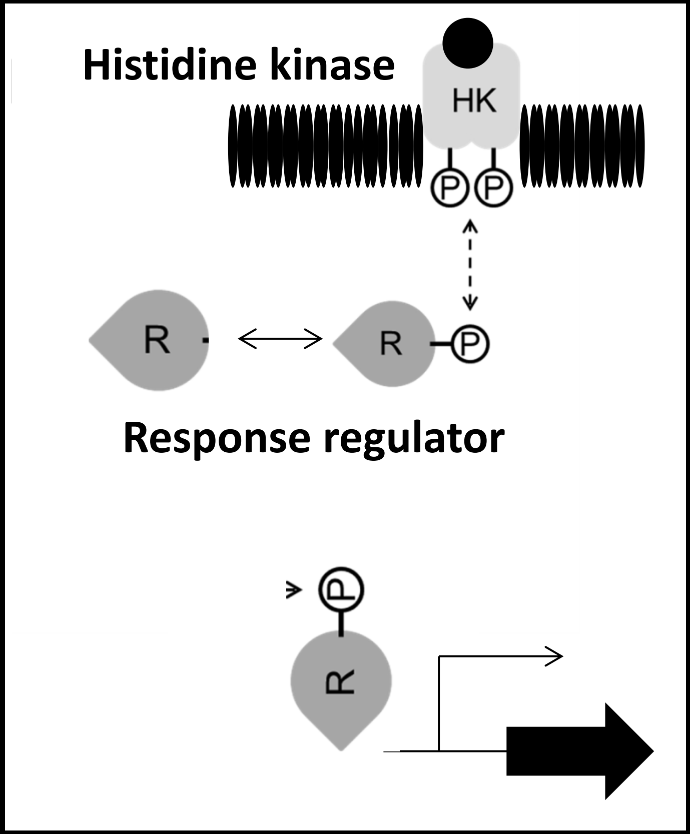
Signal Transduction in Bacteria

One component systems



e.g. *Lac repressor*

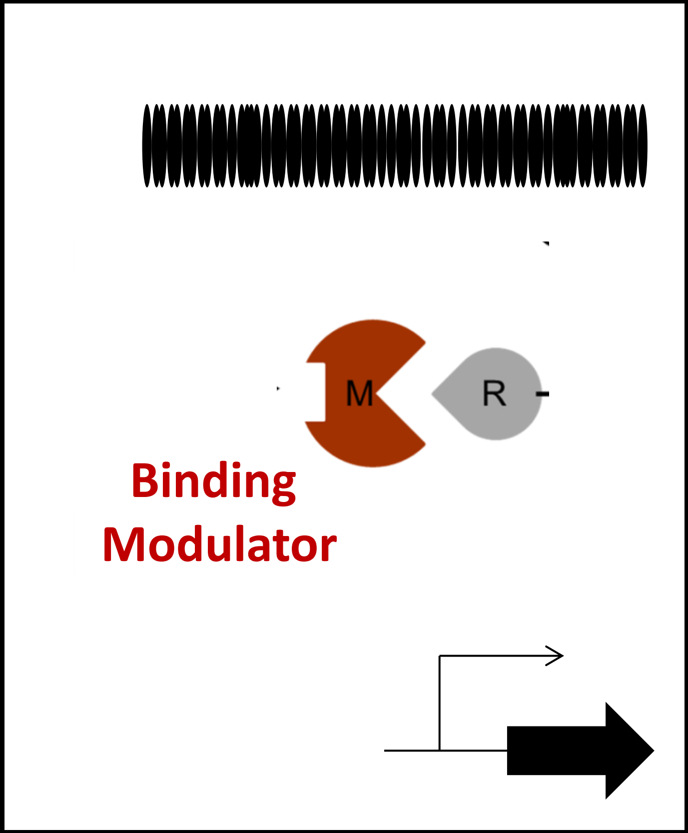
Two component systems



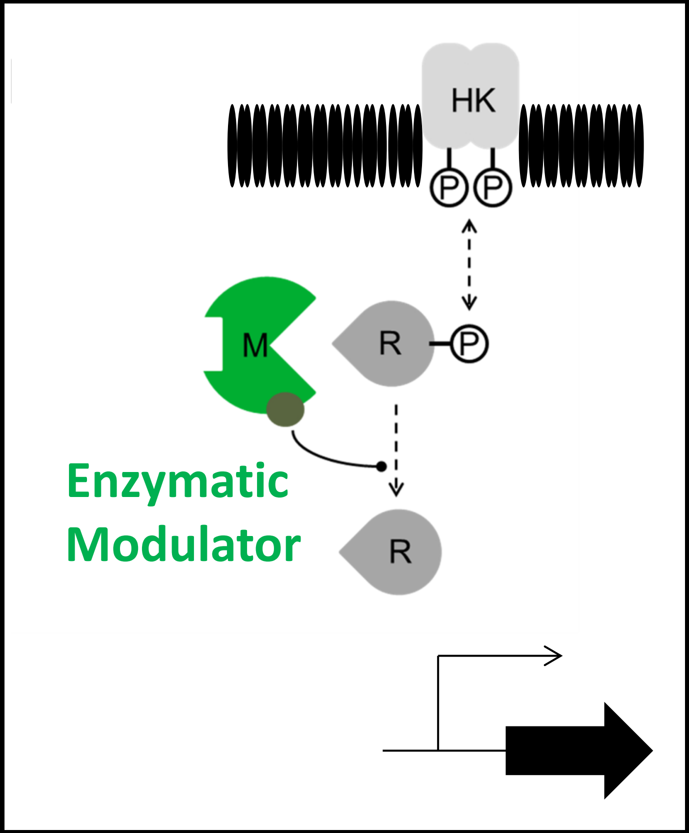
e.g. *chemotaxis CheA-CheY, EnvZ/OmpR*

Signal Transduction in Bacteria

One component systems



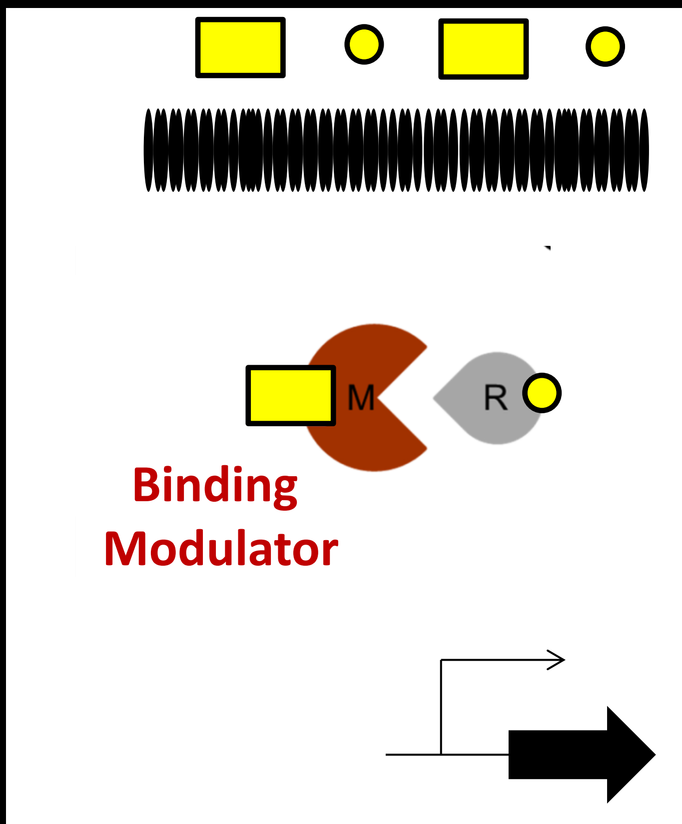
Two component systems



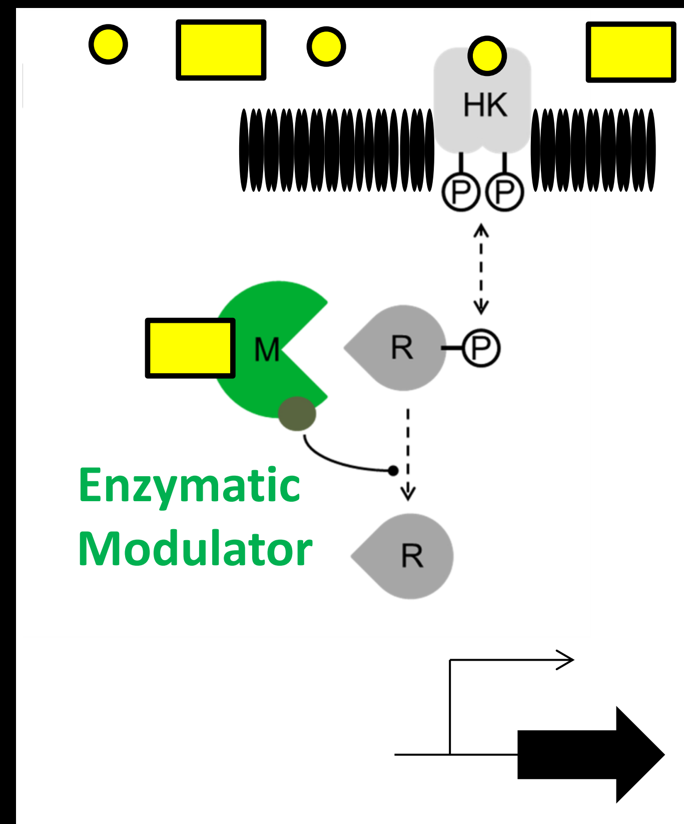
Auto-inducer signaling (Quorum sensing)

Auto-inducer

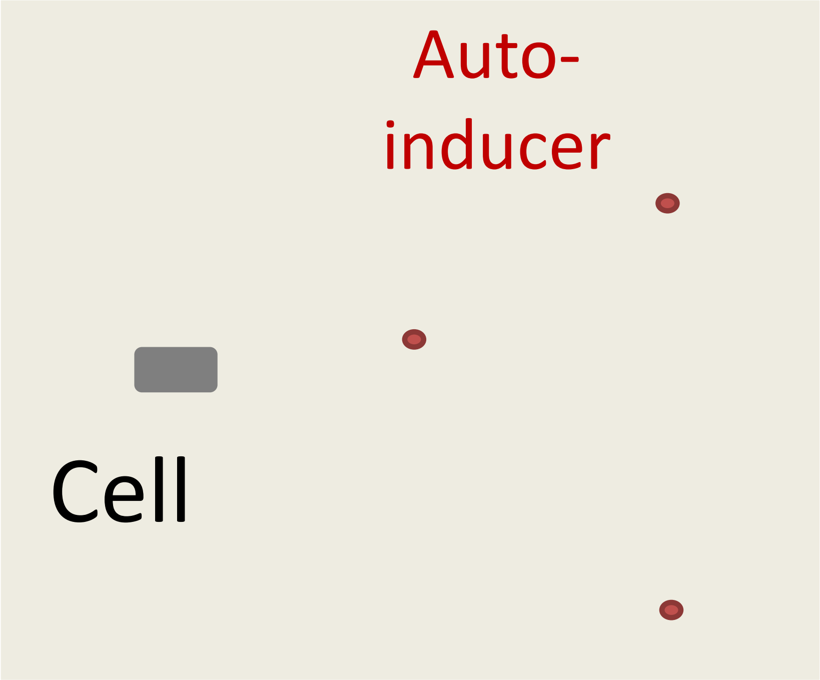
One component systems



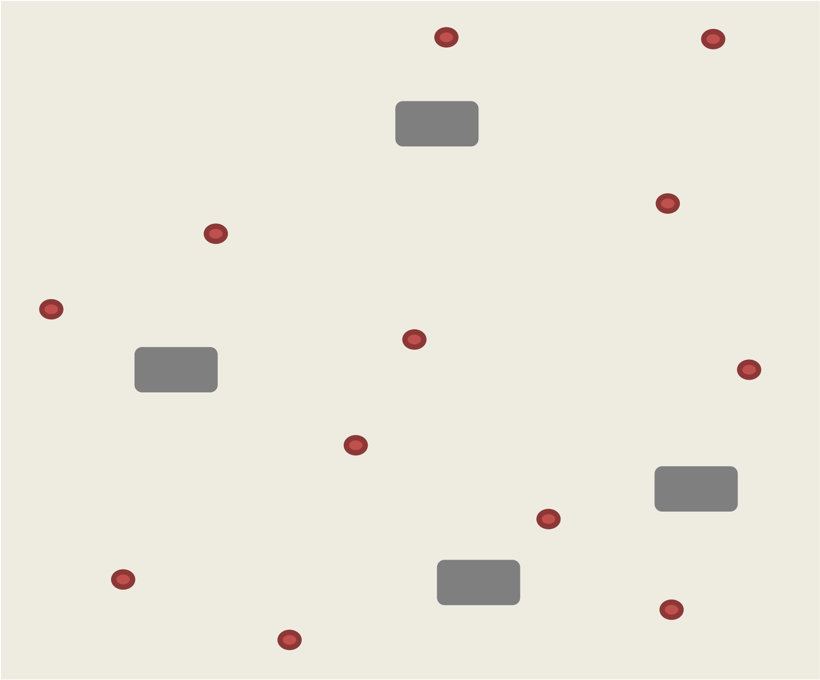
Two component systems



Quorum Sensing

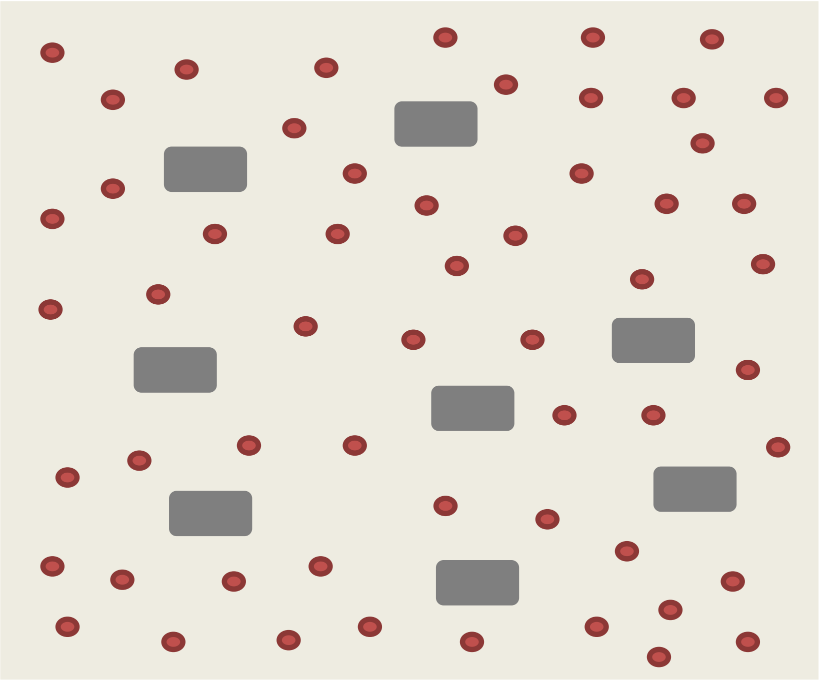


Quorum Sensing



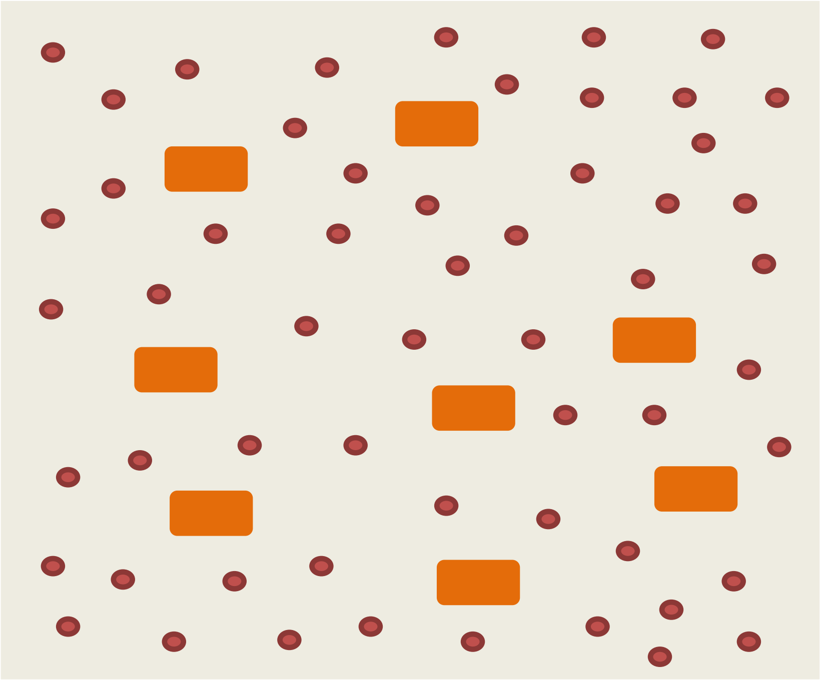
more cells – more signal

Quorum Sensing



„The quorum“ –
a minimal behavioral unit

Quorum Sensing

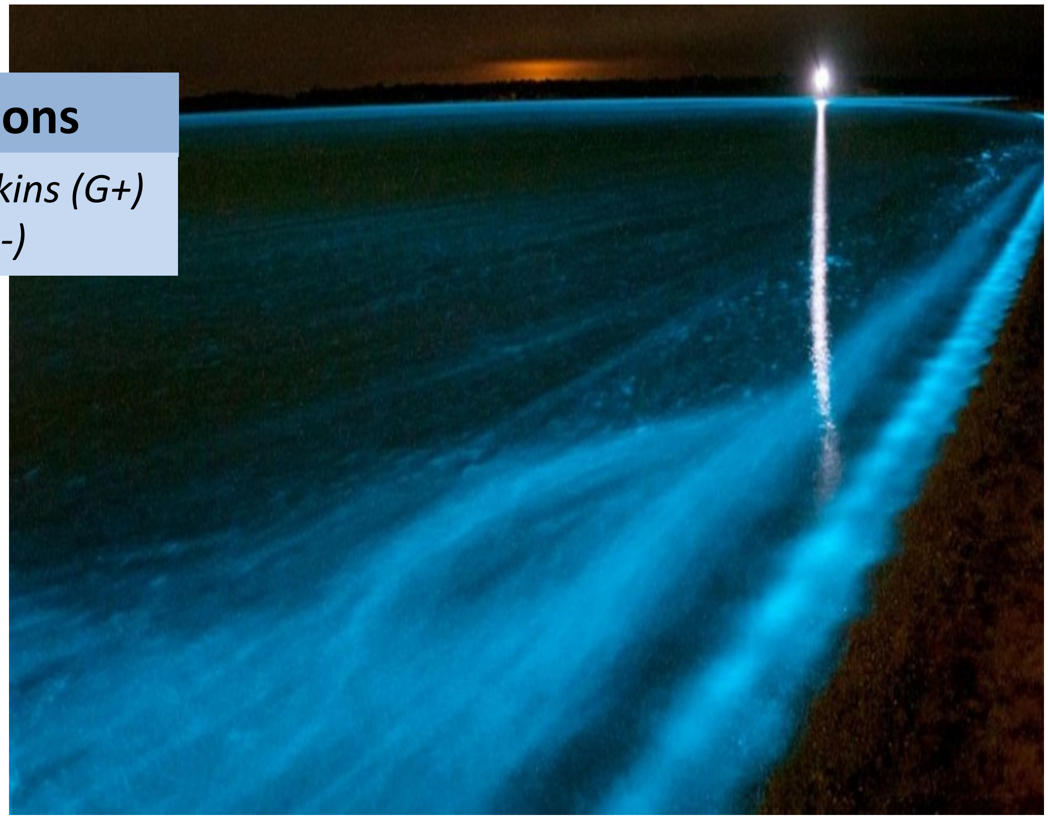


„The quorum response"

(Almost) 50 Years of Quorum Sensing

First observations

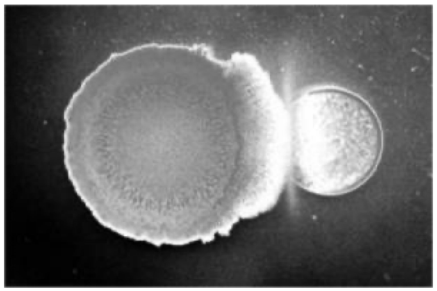
1964 *Tomasz & Hotchkins (G+)*
1970 *Neaslon et al. (G-)*



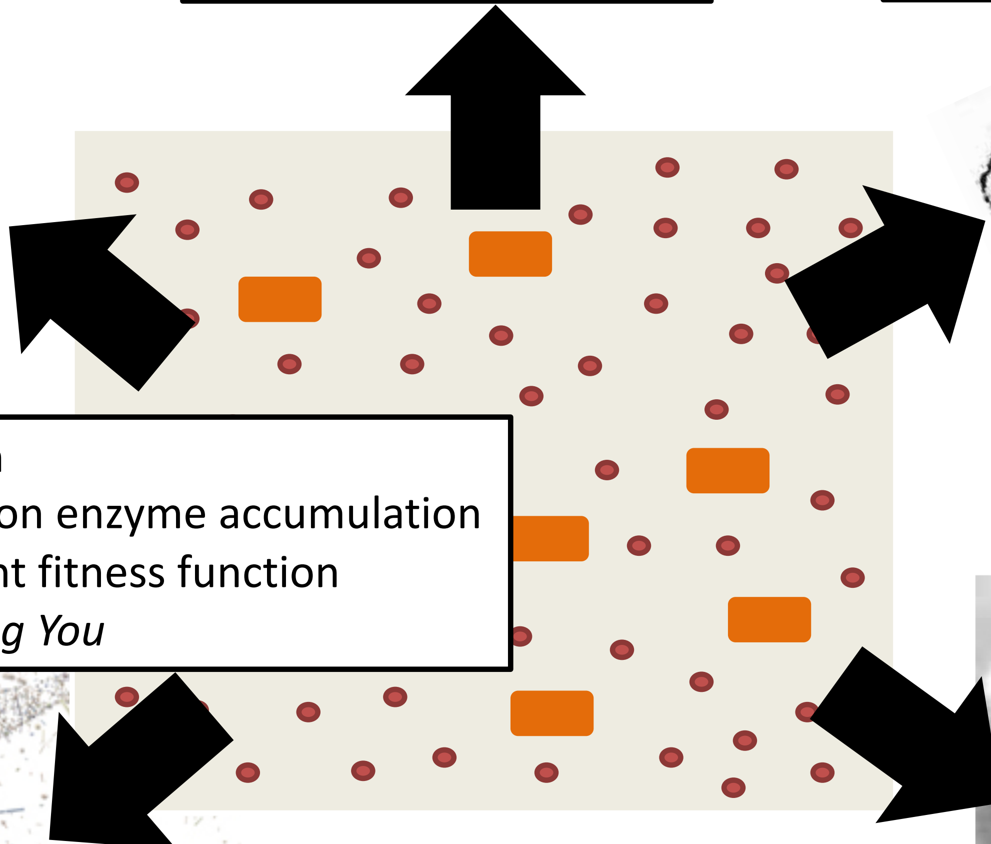
Bioluminescence is regulated by cell density.

The Quorum Response Spectrum

Production of exofactors



Horizontal gene transfer



Cell differentiation

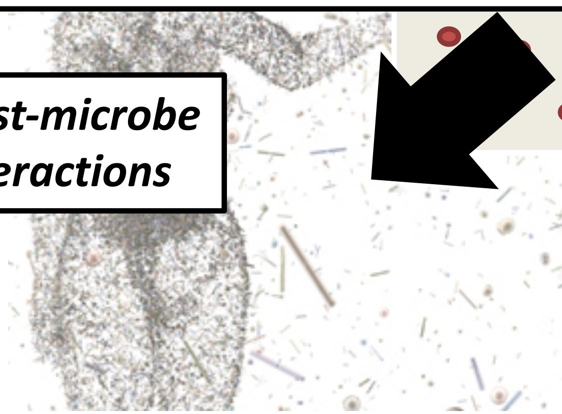
- Cost of production
- Benefit depedent on enzyme accumulation

=> Density-dependent fitness function

See work by Lingchong You

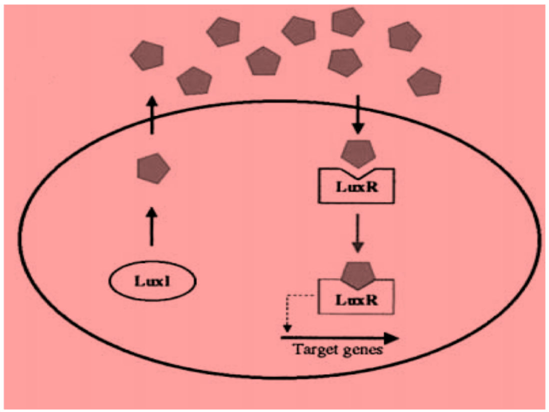
Biofilms

Host-microbe interactions



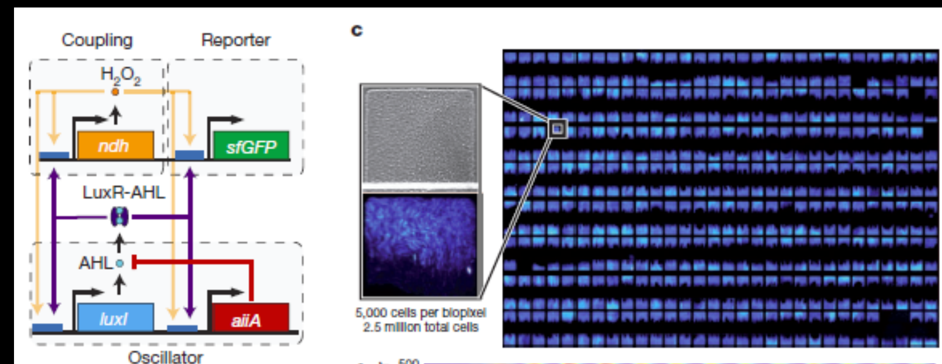
Quorum Sensing in Synthetic Biology Applications

LuxIR-type of QS Circuitry



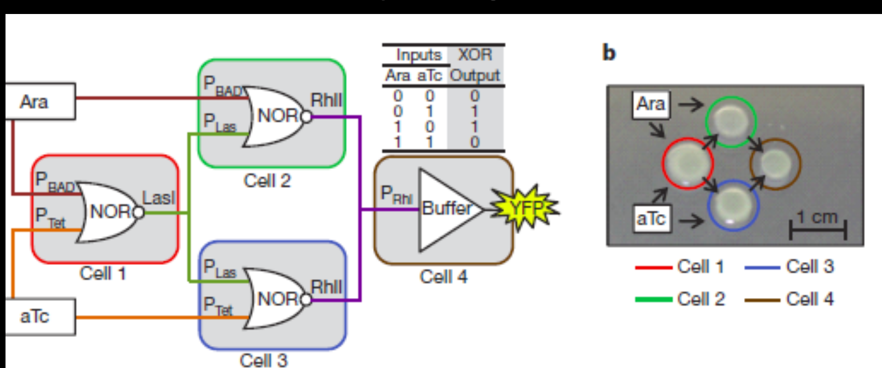
V.fischeri (Bassler, 2002)

Advanced Biosensors



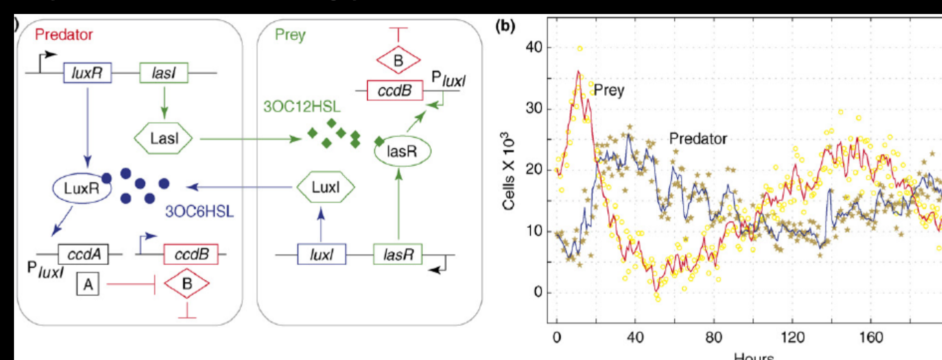
Prindle et al., Nature 2012

Multi-cellular Computing



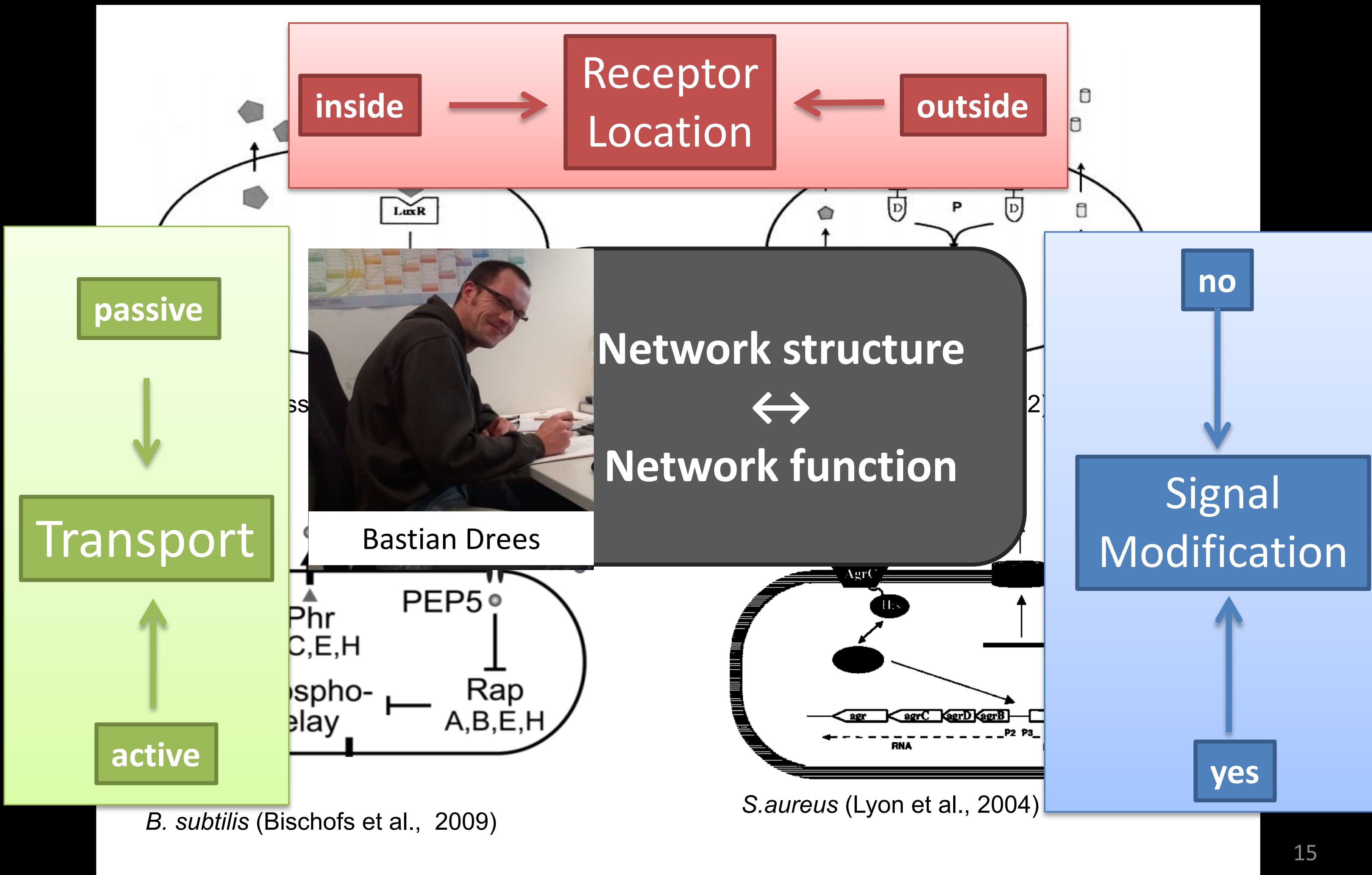
Tamsir et al., Nature 2011

Synthetic Ecology



Brenner et al., Trends in Biotechnology, 2008

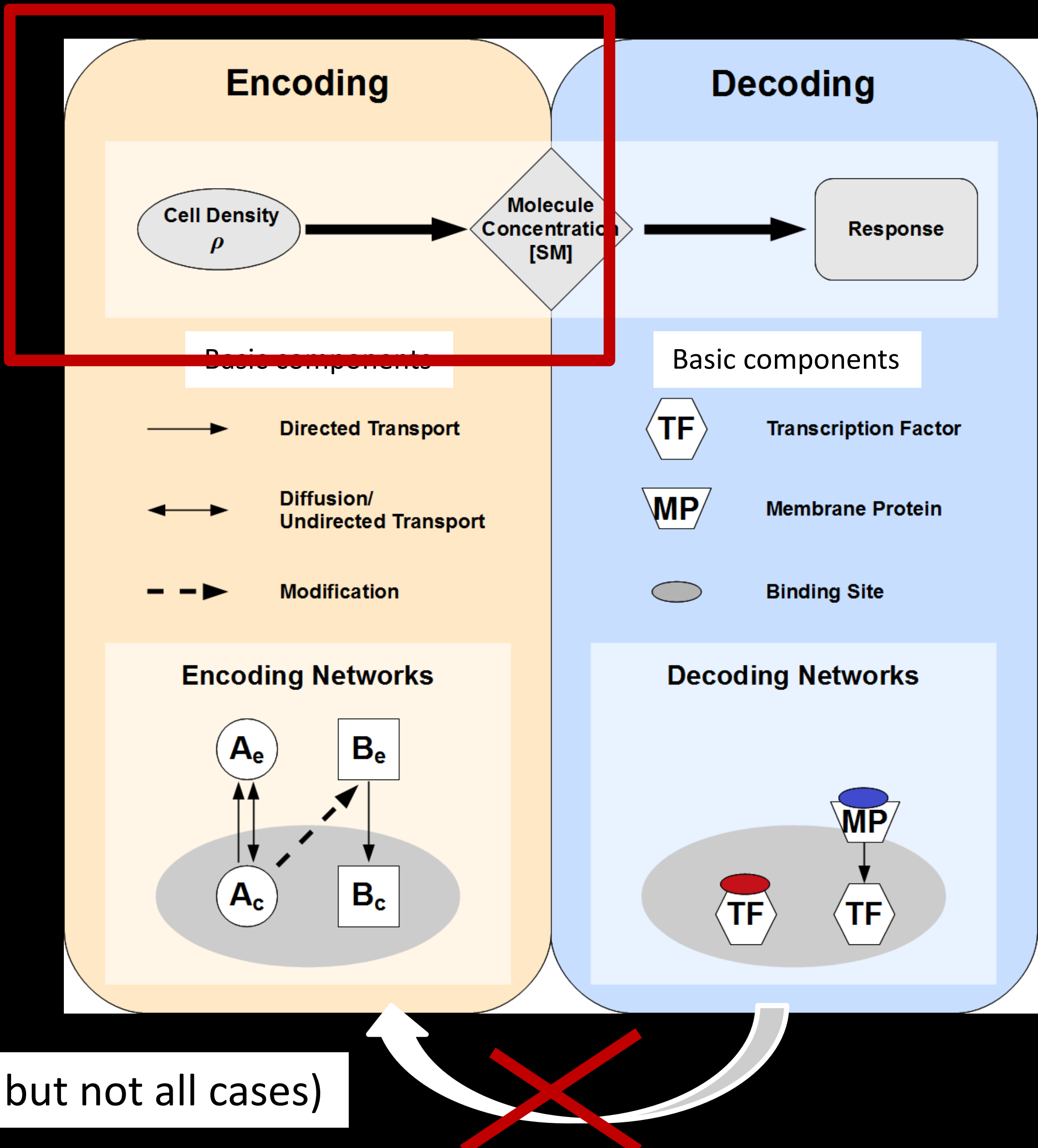
Diversity of Quorum Sensing Architectures



B. subtilis (Bischofs et al., 2009)

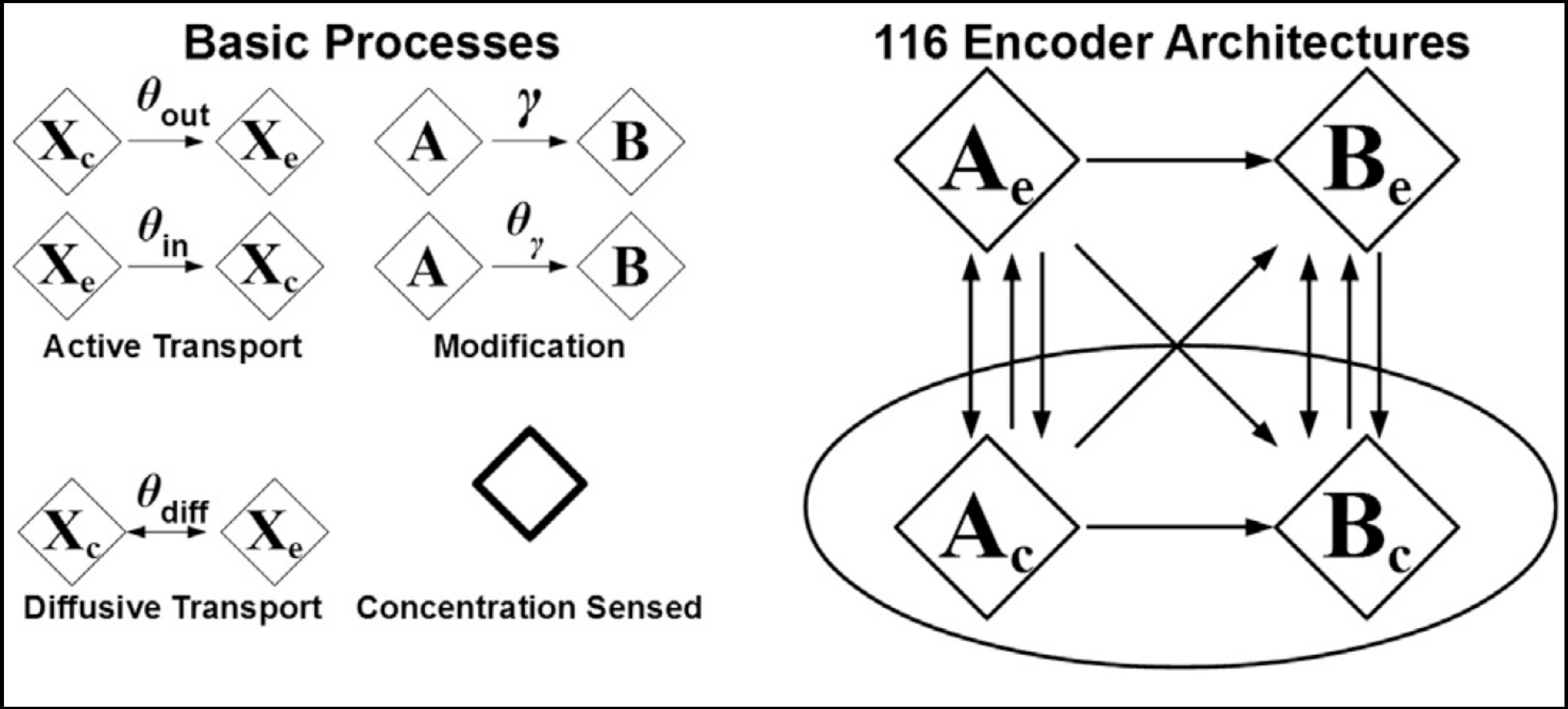
S.aureus (Lyon et al., 2004)

Adopting a modular view on QSS



Feedback (often but not all cases)

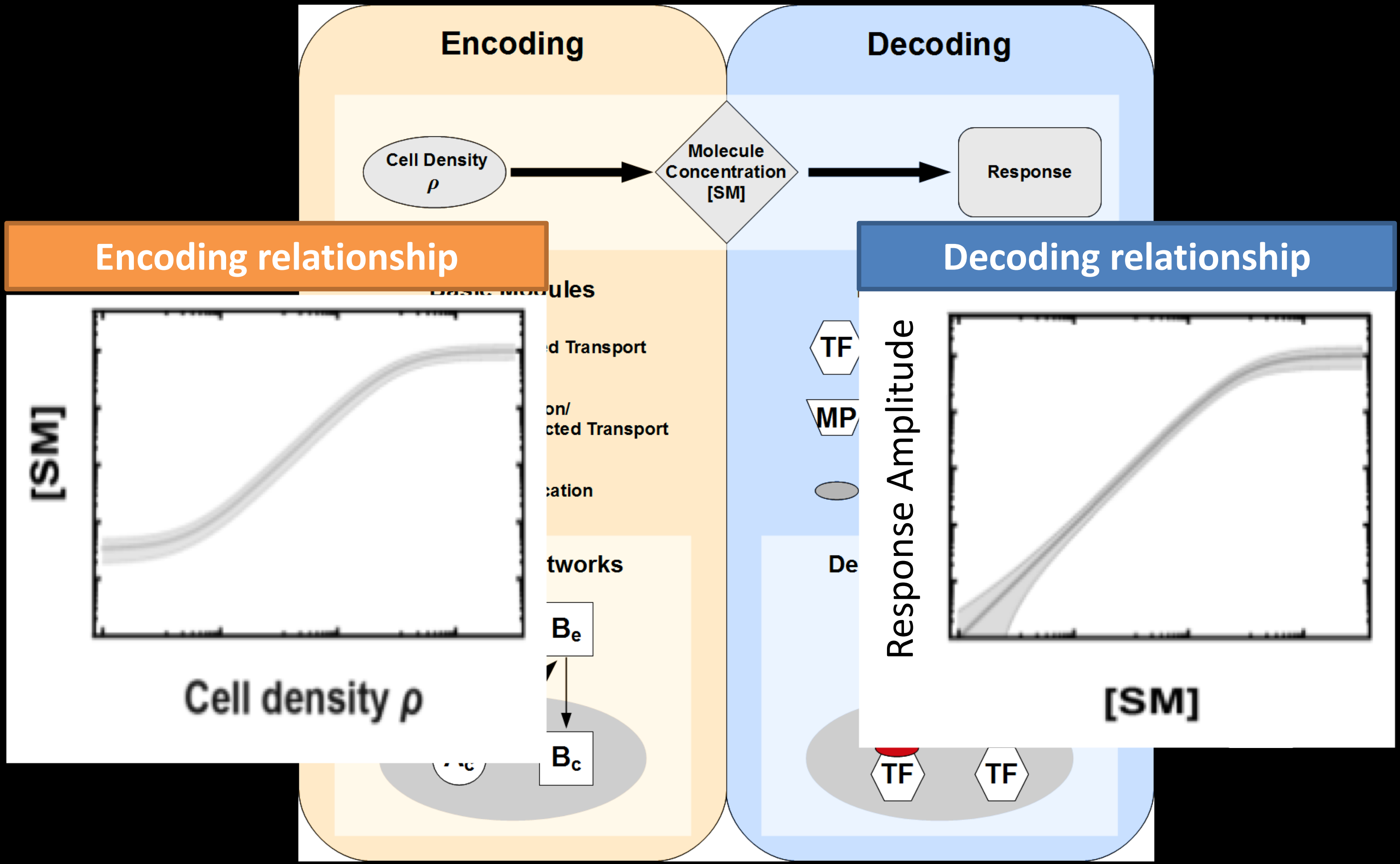
Diversity of Encoding Architectures



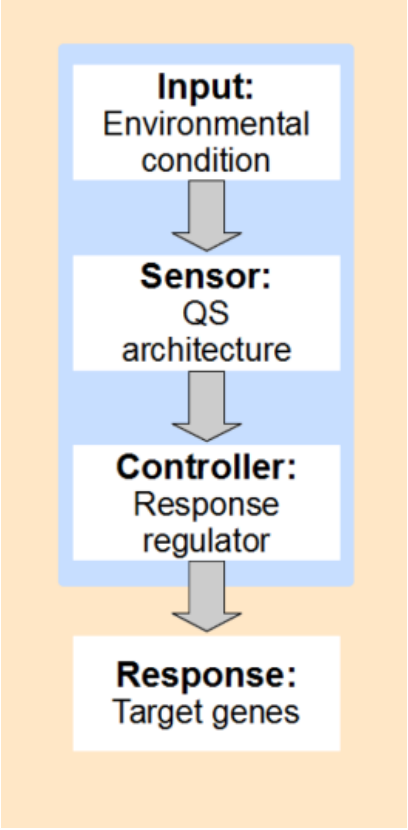
99 networks (85%) are capable
of encoding information about cell density into SM concentration.

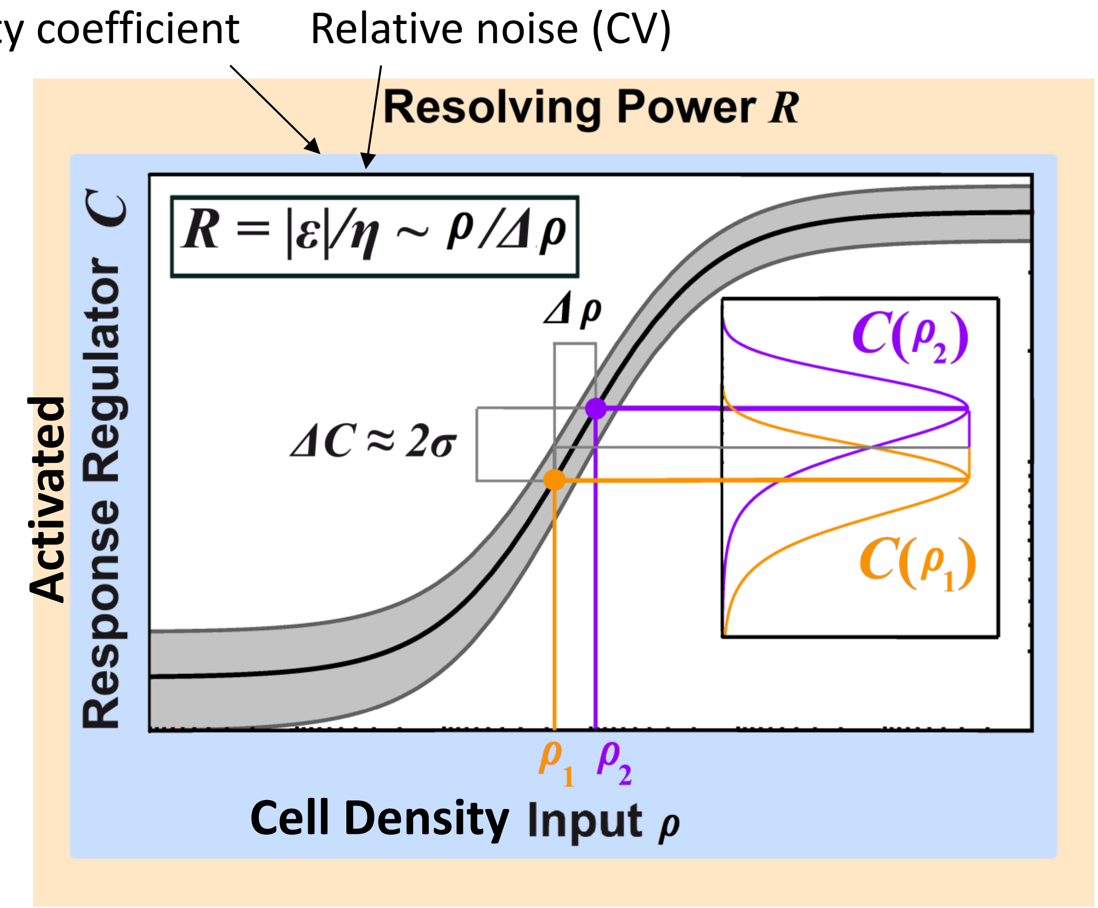
Drees at al., BPJ 2014

Adopting a Modular View on QSS



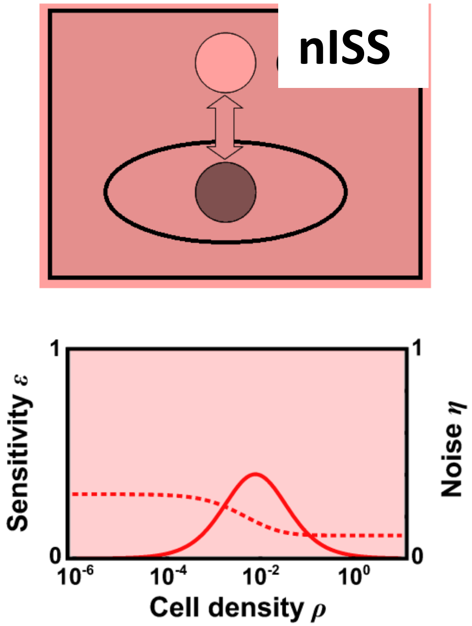
Defining a Performance Criterion

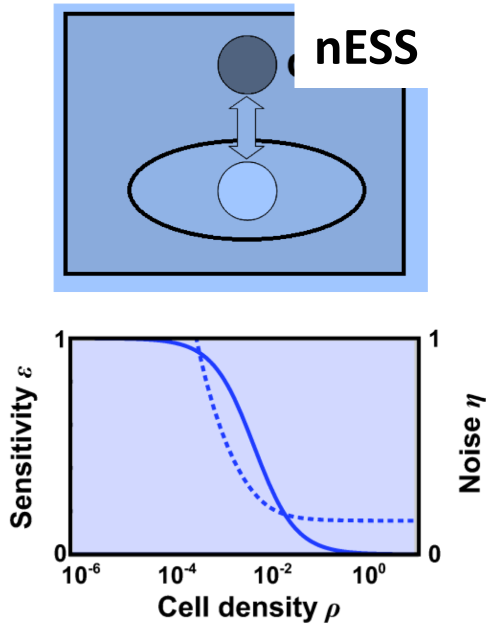


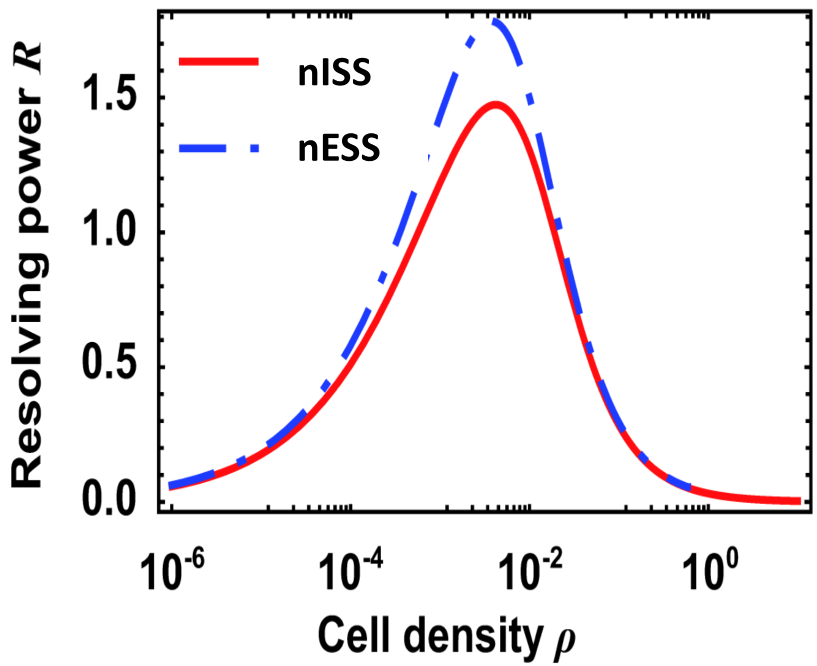


Relation to mutual information: $M_{max} = \log_2\left(\frac{1}{\sqrt{2\pi e}}\int\frac{R(I)}{I}dI\right)$

Quantifying Performance



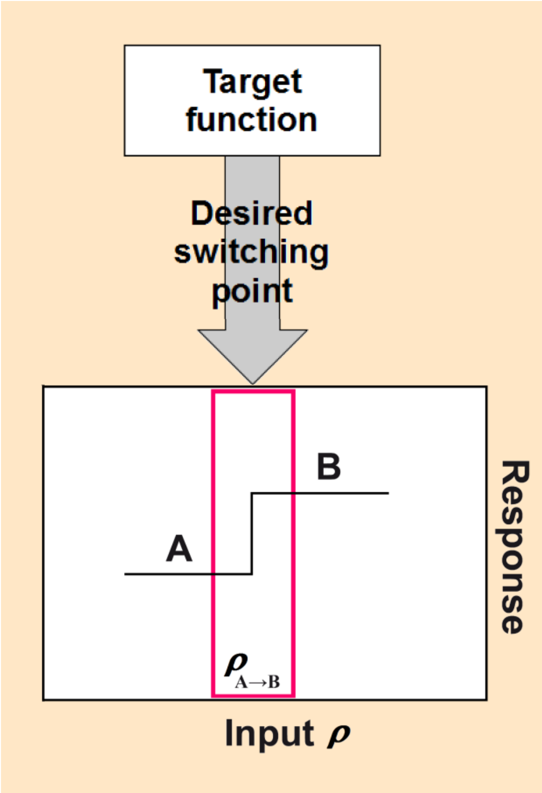


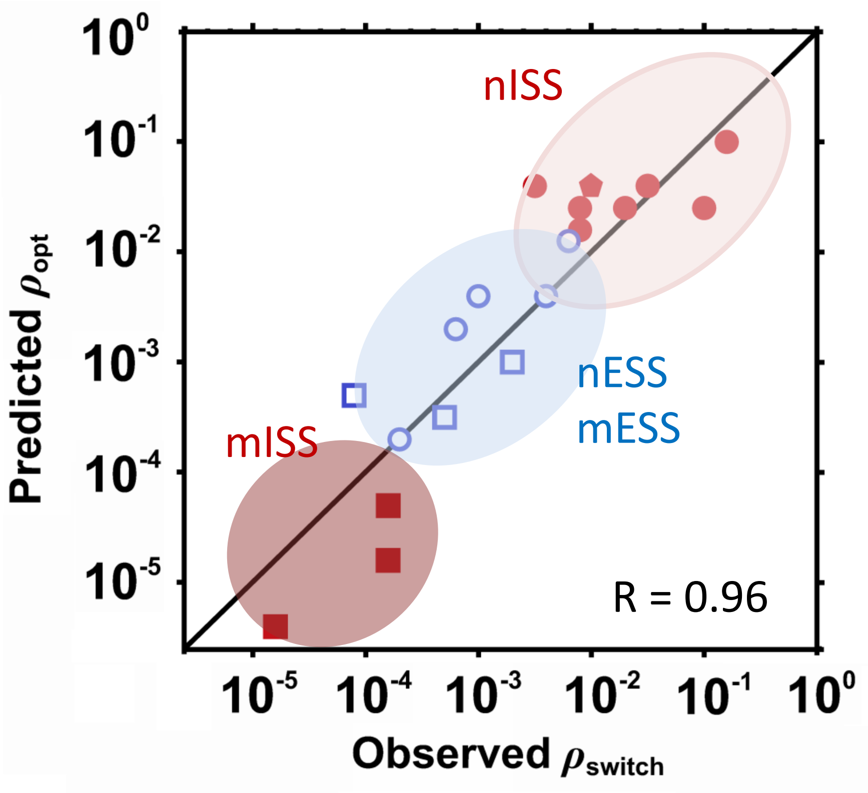


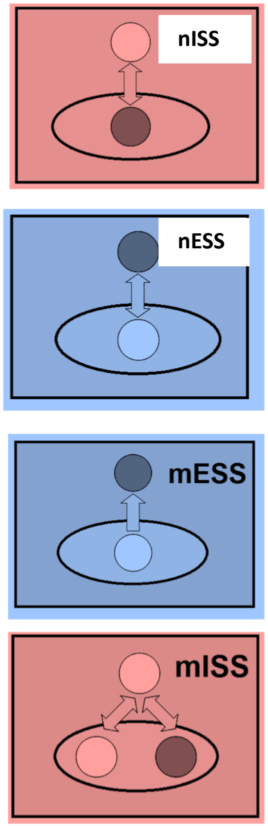
QS architectures have distinct sensitivity and noise charachteristics.

QSS achieve optimal performance at a certain cell density input.

The optimal sensory performance is matched to the „quorum“







Performance Trade-offs

Encoder with fixed parameters

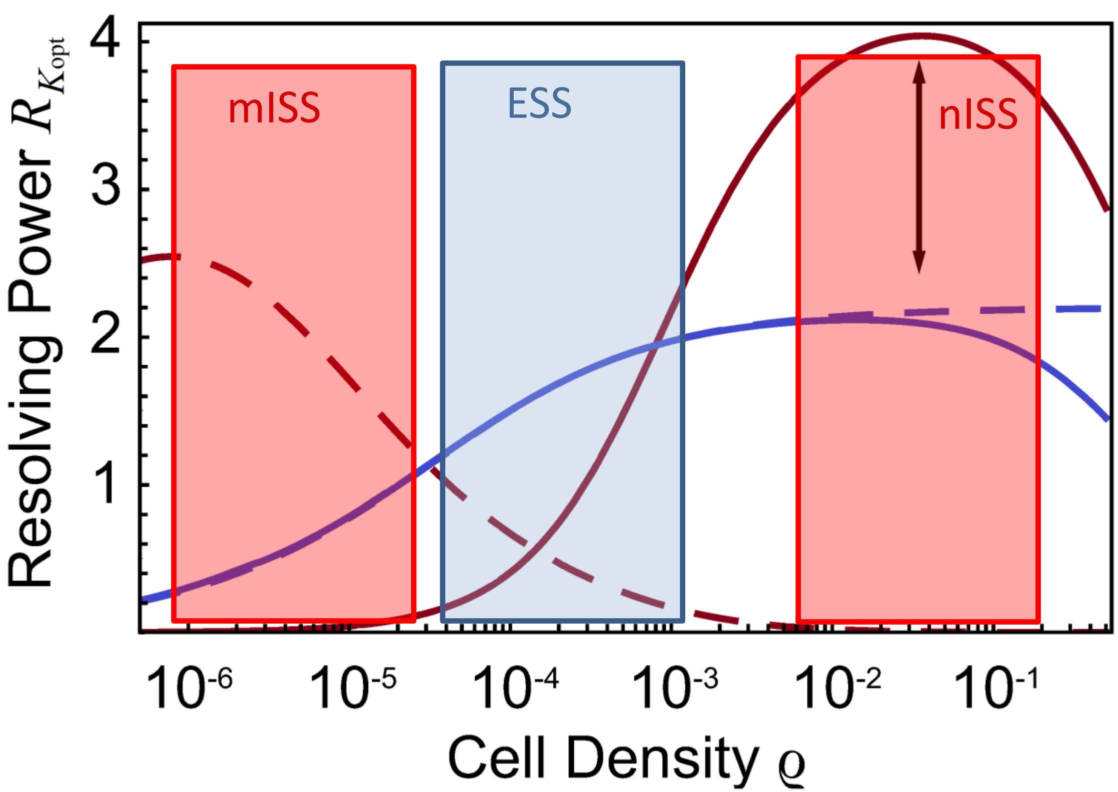
↓

Decoder – Encoder Matching (optimize receptor affinities Ks)

↓

Maximal Resolving Power

Resolving power with a „matched" decoder.



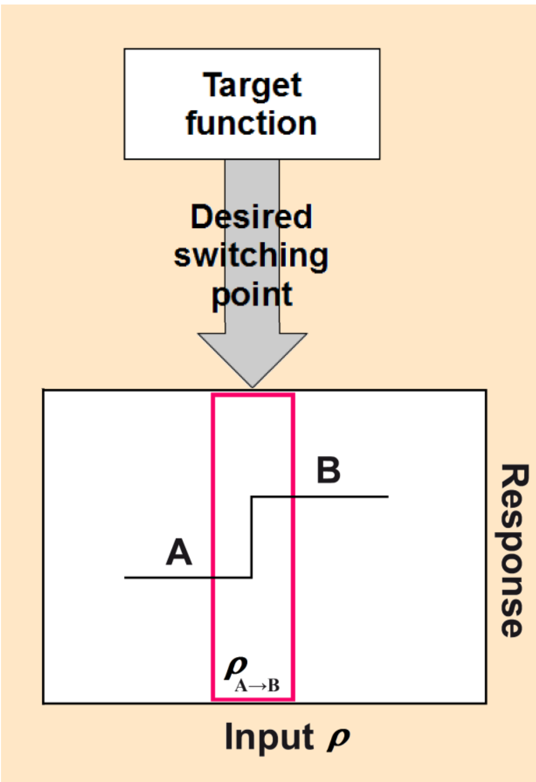
Trade-off

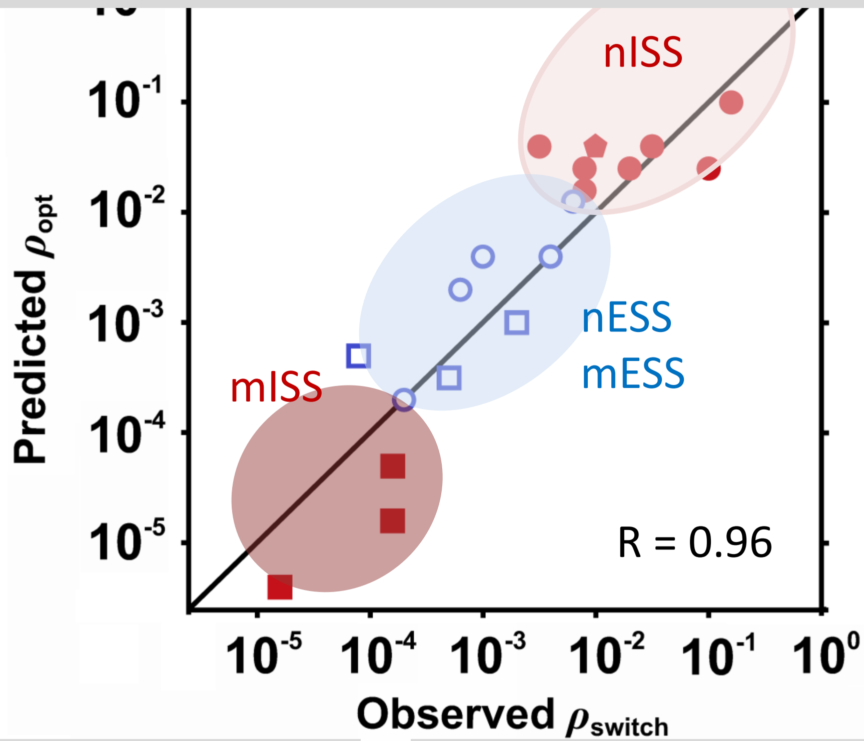
Architectural Complexity
(better sensitivity characteristics)
nESS

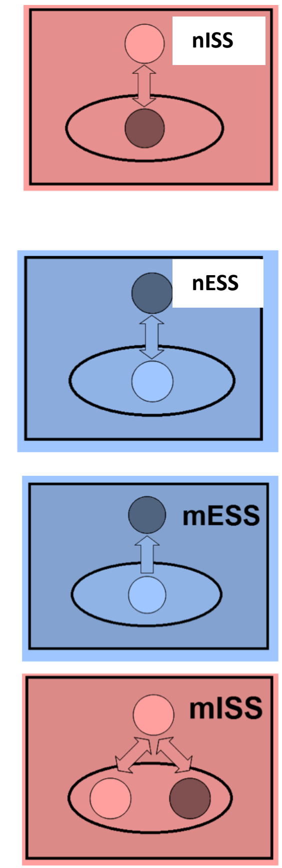
Architectural Simplicity
(better noise characteristics)
nISS

Conclusions

<u>Design Principle for Synthetic Biology</u>
Low cell density sensing may require other QSS architectures than the simple LuxIR-type system.







<u>Evolution of QSS target genes:</u>
Does the QSS architecture constrain the evolution of QSS target genes?

<u>It's really hard to find parameters to model QSS ...</u>
Calculations based on guestimates .....

Traditional Quorum Sensing Research

The QS Paradigm

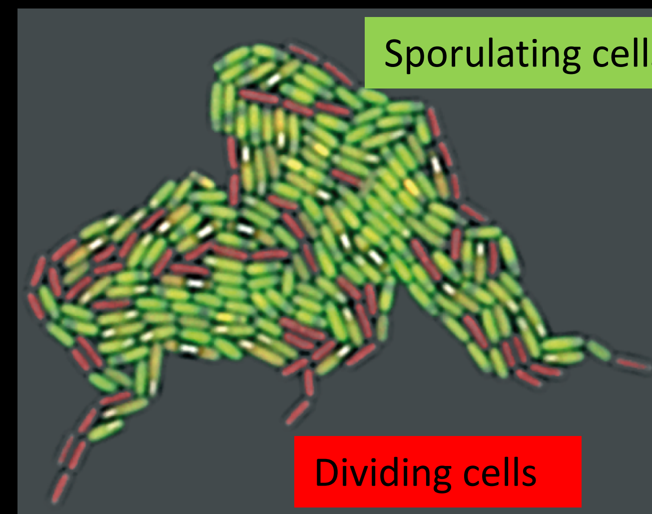
Homogeneous population

Cell density sensing

Synchronized response

Complex adaptive traits

Heterogeneous population



Distribution of different phenotypes

Division of labor

J. van Gestel et al.,
Plos Biology, 2015

Bet hedging

Reviewed in: Veening et al.,
Annu. Rev. Microbiol. 2008

Population-based <u>quantitative</u> trait

Ackermann, Nature Reviews Microbiology (2015)

Our mission

Molecular Determinants & Mechanisms

understand

Complex Adaptive Traits (CATs)

Organizing Principles

Molecular Level

Population Level

engineer

control

Fluorescence timelapse microscopy

Stephanie Trauth

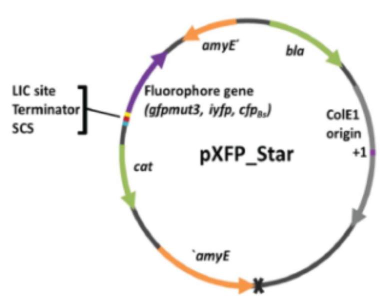
Marika Ziesack

Charlotte Kaspar

Svenja Schwindt

Group of K. Rohr

Genetic engineering



Fluorescence reporter

Trauth & Bischofs, 2014

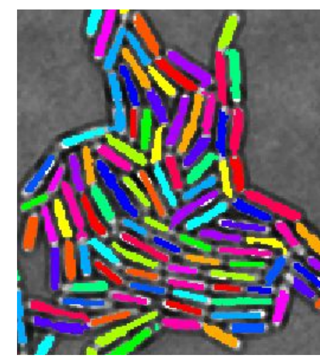
Timelapse microscopy

Hydrogels

Microfluidics

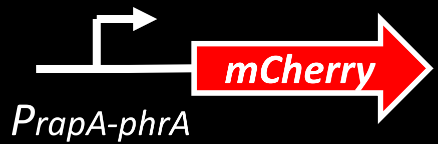
Group of D. Kohlheyer

Image Processing

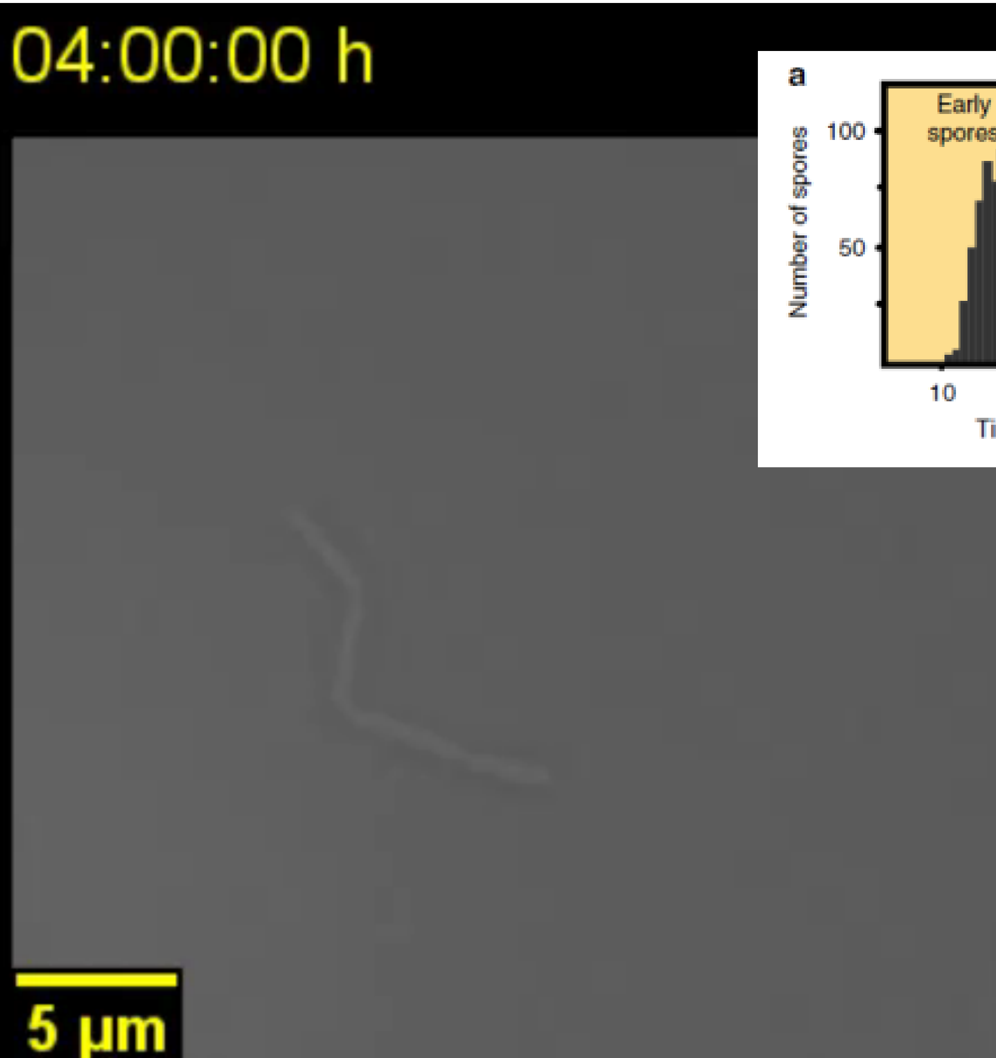


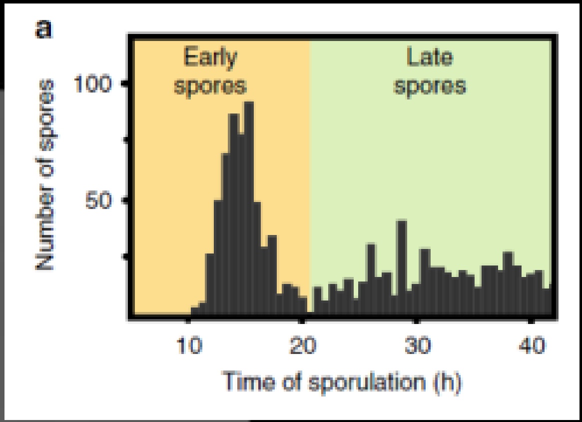
Liluashvili V. et al., 2015

Heterochronic population response



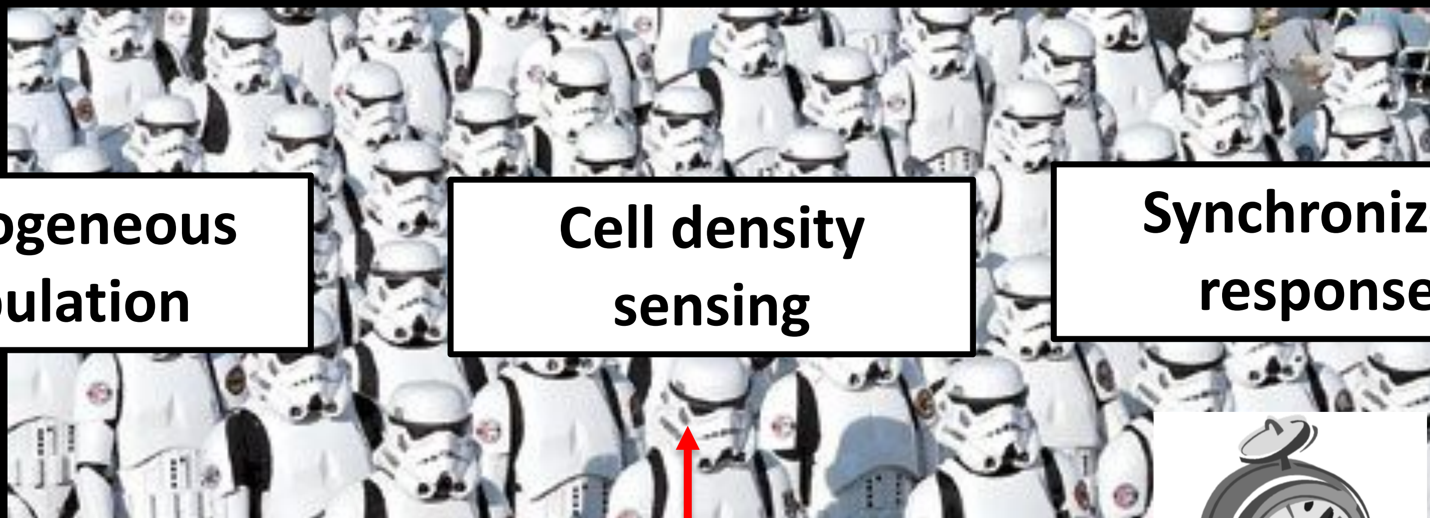
Alper Mutlu





Mutlu et al., Nat. Commun. (2018)   Mutlu et al., ISME J (2020)

Going beyond the QS paradigm



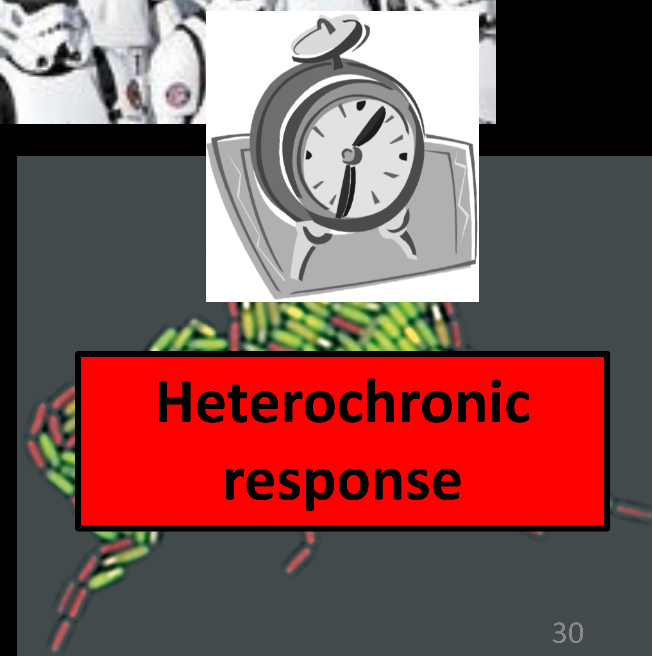
Homogeneous population

Cell density sensing

Synchronized response

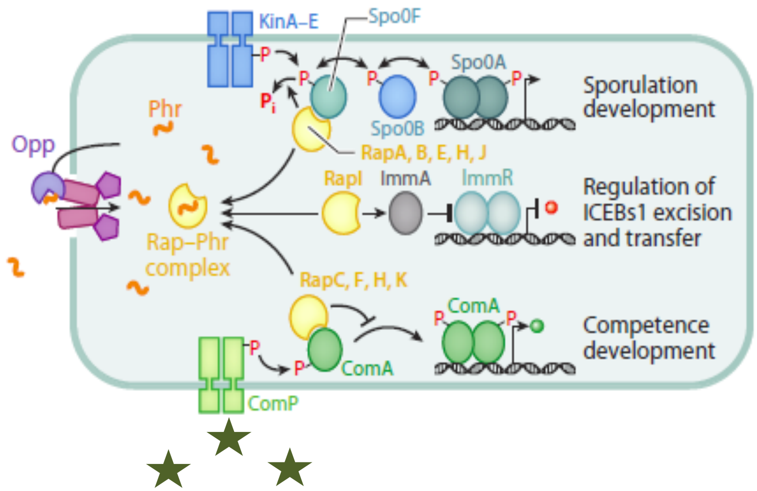
Heterogeneous population

?



Heterochronic response

Communication in *B. subtilis*

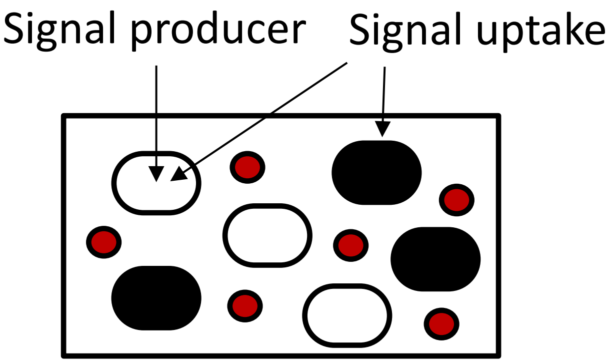


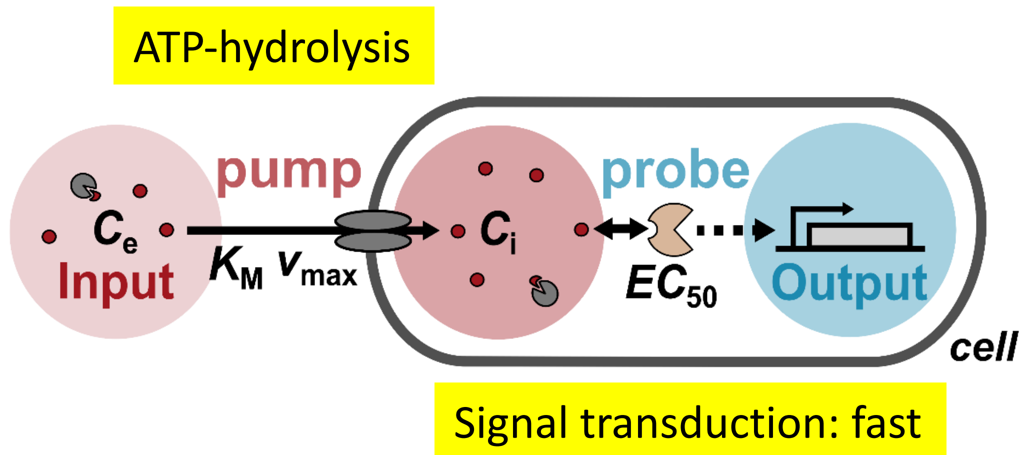
Neiditch et al.,
Ann. Rev. Genetics (2017)

Rap-Phr-Systems are commonly referred as **quorum sensing** systems.
However, there is little experimental evidence for a cell-density dependent type of regulation.

The Pump-Probe Model

Phenomenological (ODE-type) model





Assumptions

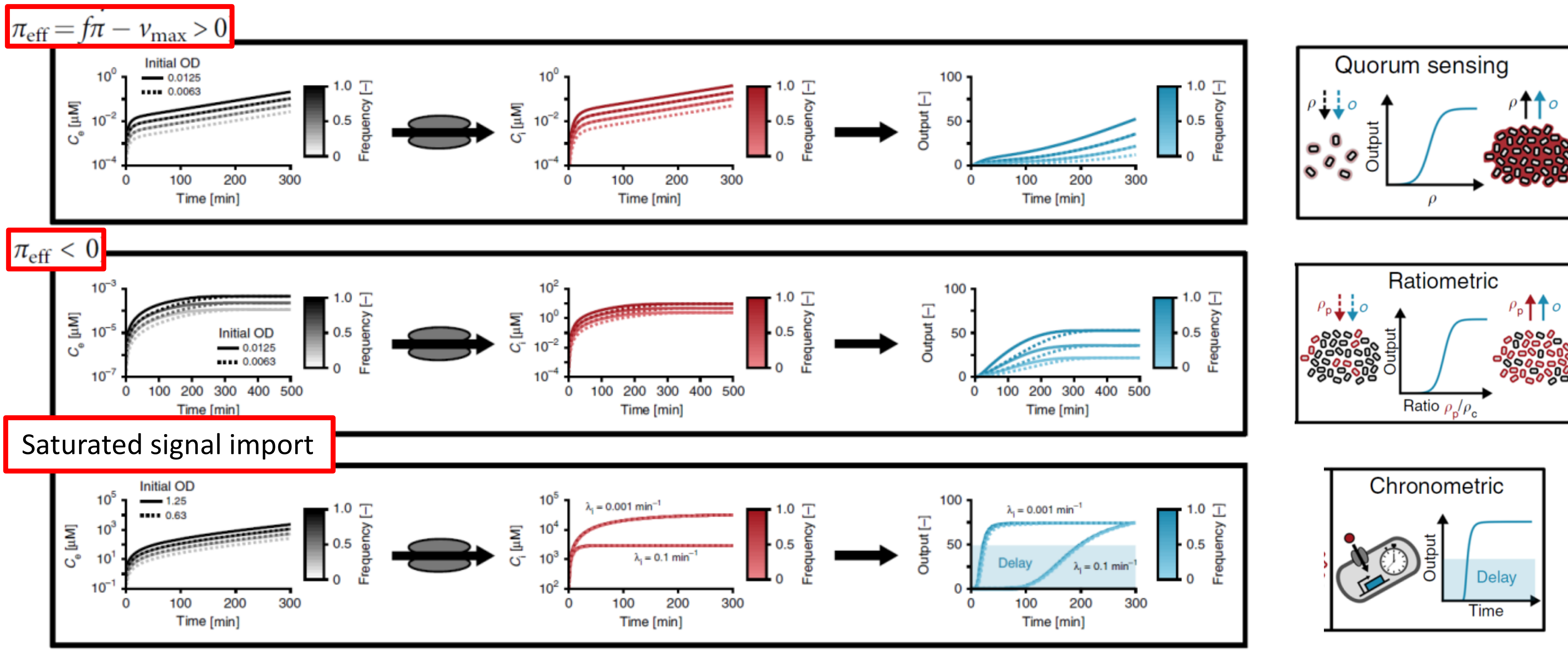
Population model

- Heterogeneous population:
  Signal produced by a subpopulation of cells present at frequency *f*
- Signal uptake by all cells.
- Exponentially growing population
  (same growth rate & well-mixed)
- Different starting cell densities (OD)

Signal transduction model

- Signal uptake with Michaelis-Menten pump kinetics
- Signal degradation (and dilution by cell growth)
- Output is a function of the intracellular signal concentration using a Hill function

One network architecture – different control functions

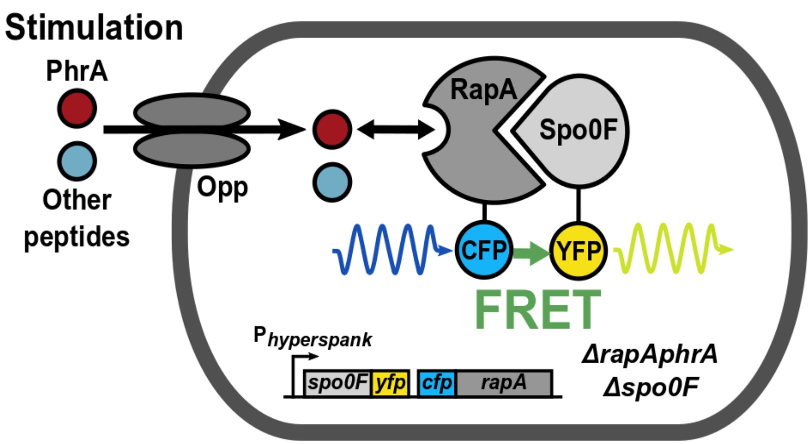


Control functions depend on network parameters and operating conditions

not (well) defined

Fluorescence (Förster) resonance energy transfer (FRET)

A new tool for quantitative studies of PhrA-signaling in bacterial cells



Tool for studying
protein-protein interactions
in vivo

(relatively) **short-ranged** interaction

Förster radius:
R_0 ~ nm

Energy transfer rate:

$$k_T = 1/\tau_D * \left(\frac{R_0}{R}\right)^6$$

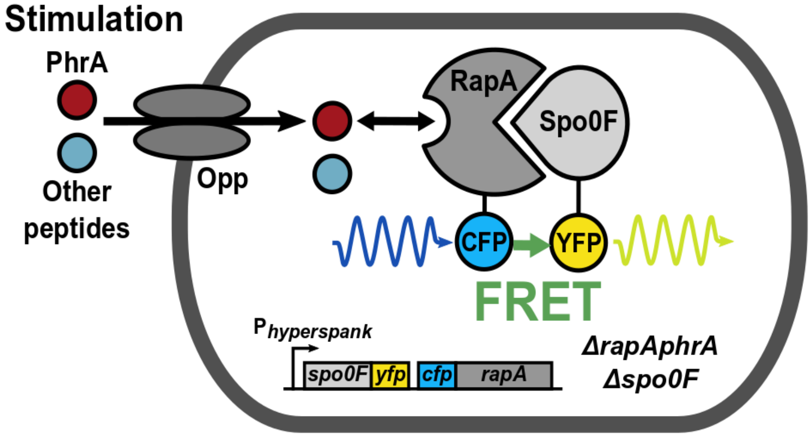
τ_D: Fluorescence lifetime of the donor

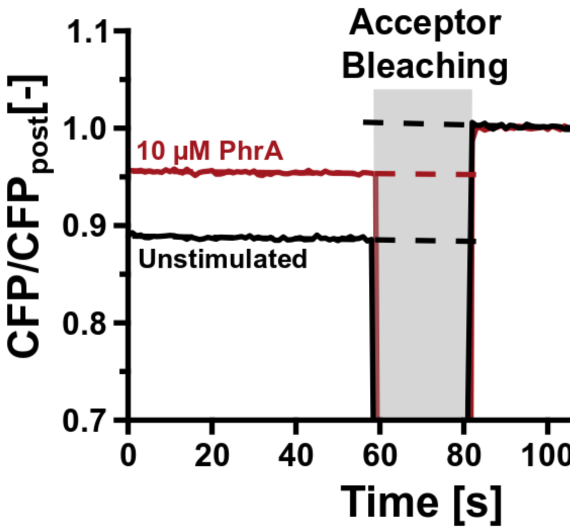
R: Distance between chromophores

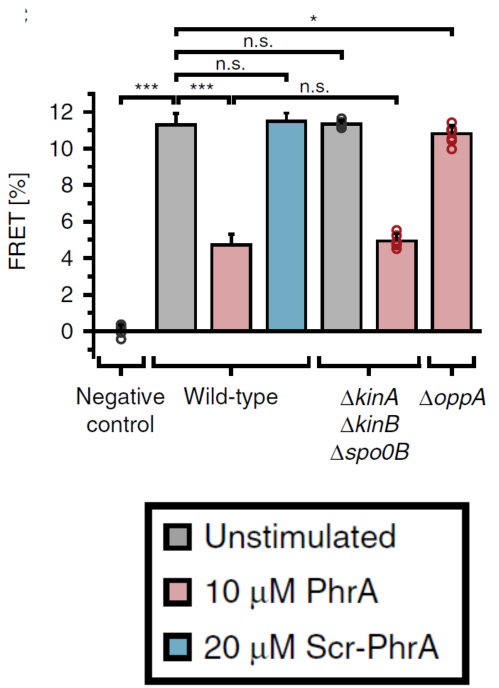
Babel et al., Nat. Commun. 2020

Acceptor-photobleaching experiments

FRET efficiency: $$\text{FRET} = \frac{\text{CFP}_{\text{post}} - \text{CFP}_{\text{pre}}}{\text{CFP}_{\text{post}}} \cdot 100\%$$





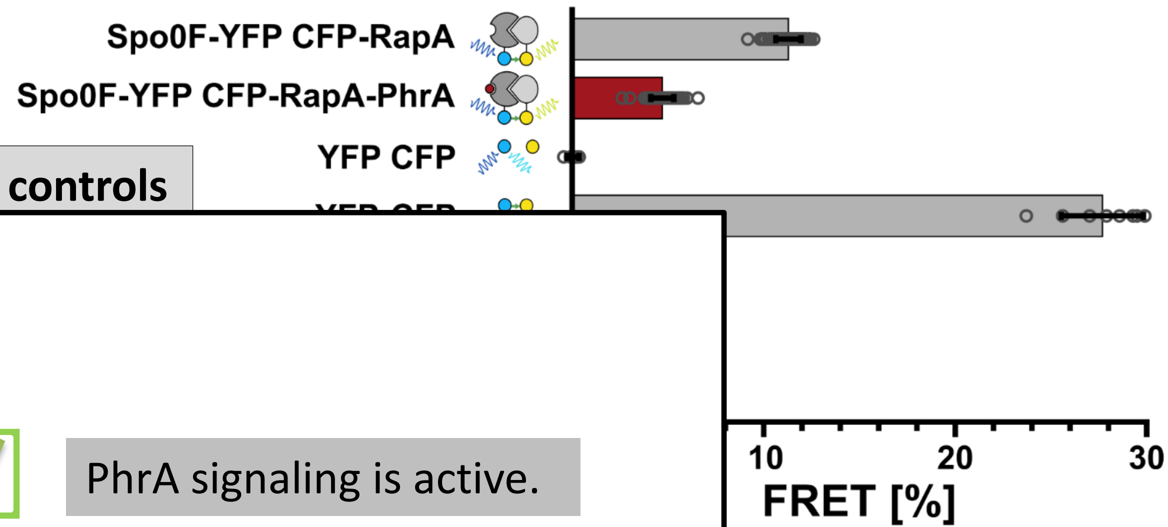


Population-average measurement

Babel et al., Nat. Commun. 2020

FRET specifically reports on PhrA

FRET controls



Molecular crowding controls

Bleed

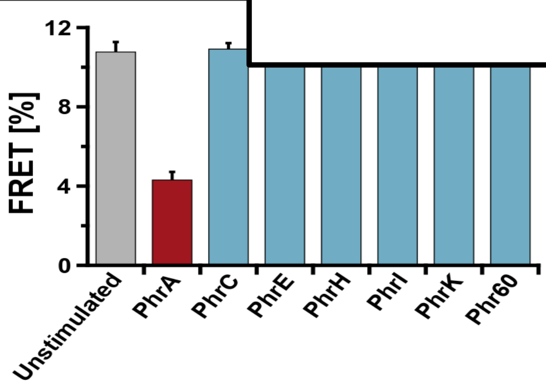
Valida

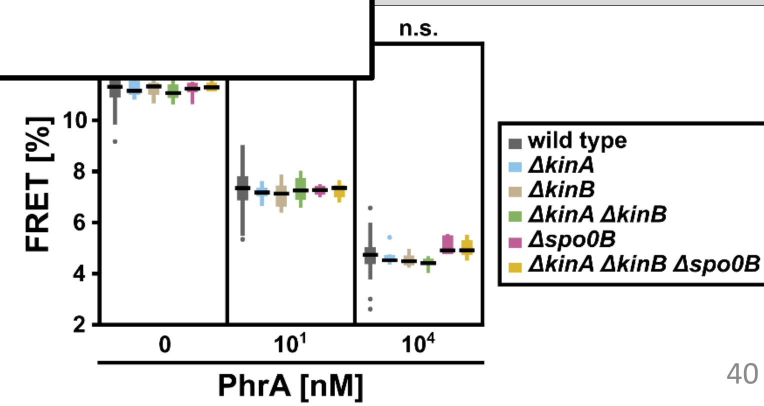
PhrA signaling is active.

Reporter controls

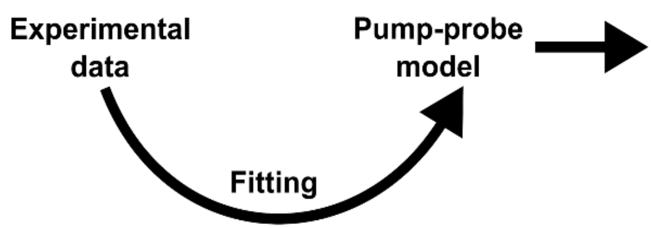
No cross-tal

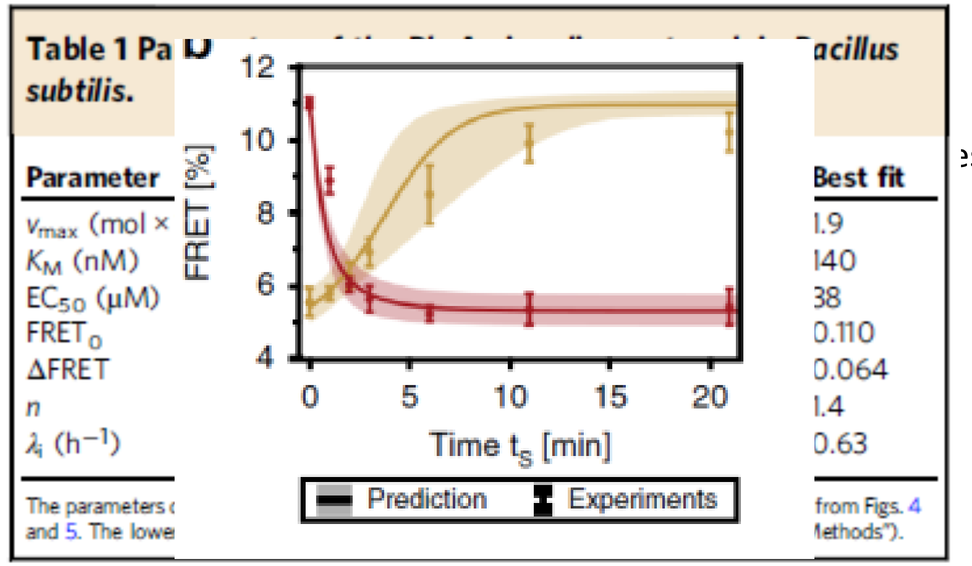
osphorelay signaling

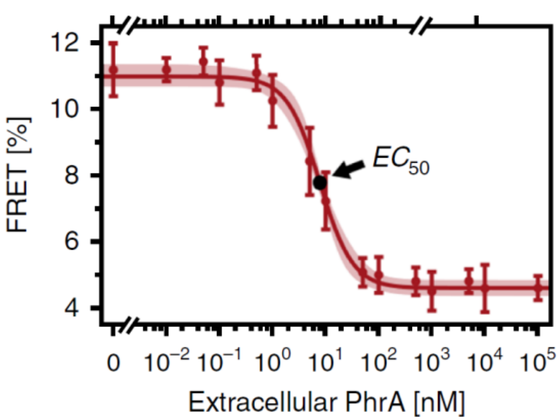


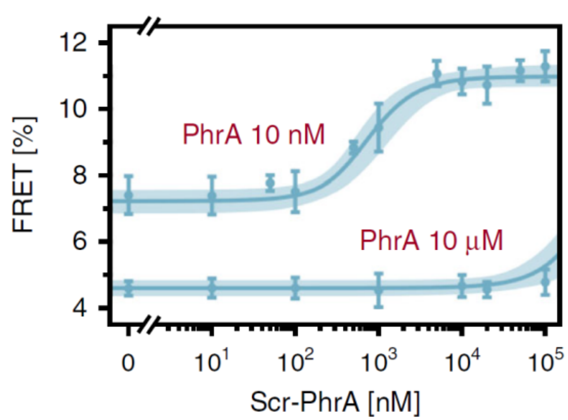


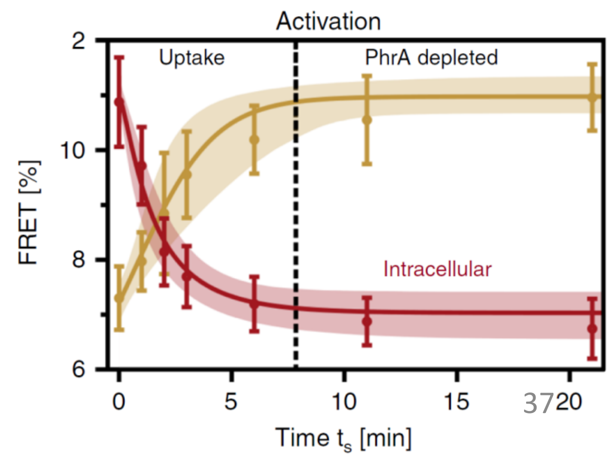
PhrA processing is well described by the pump-probe model











Cells accumulate PhrA and trigger a response at µM-levels

PhrA is „integrated“ over a relatively long time.

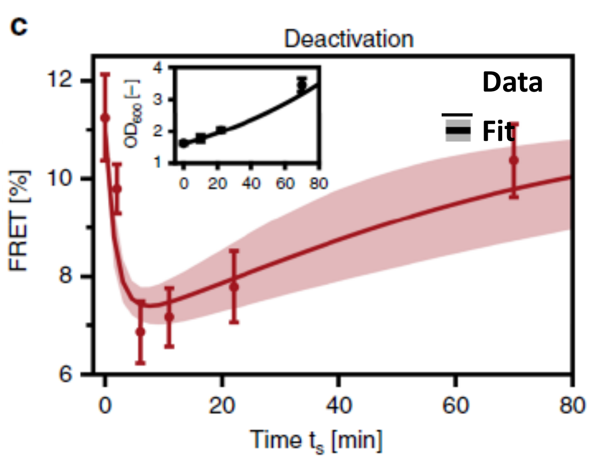
$$\tau = 1/(\mu + \lambda_i) \sim 50 \text{ min}$$

Model: Large signal amplification upon extra- to intracellular conversion.

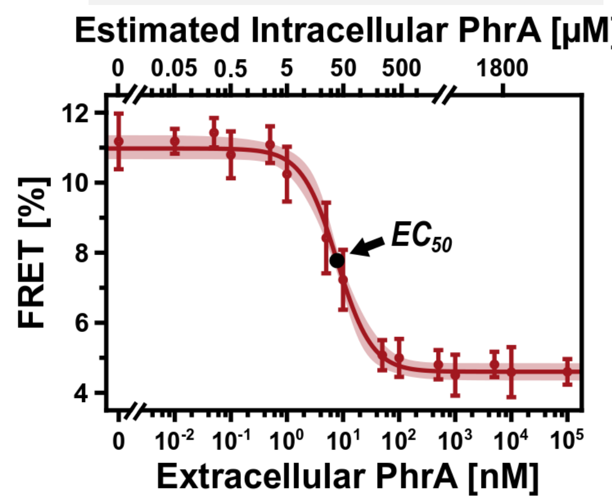
Intracellular FRET response is triggered in the **µM**-regime.

THE PROBE

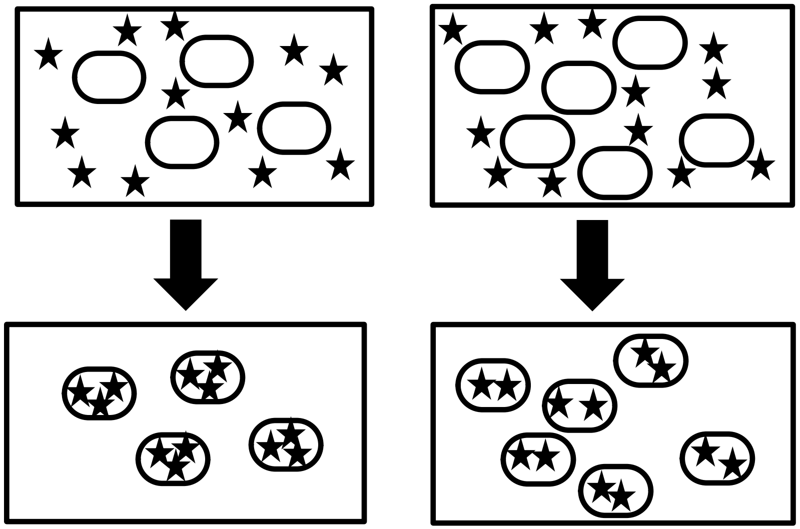
Dynamics of recovery

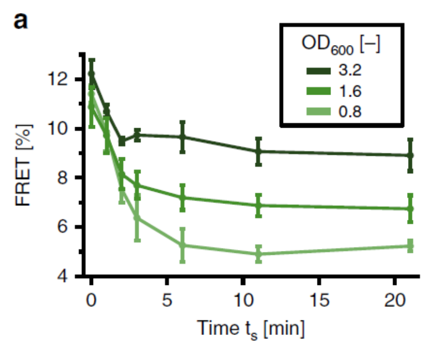


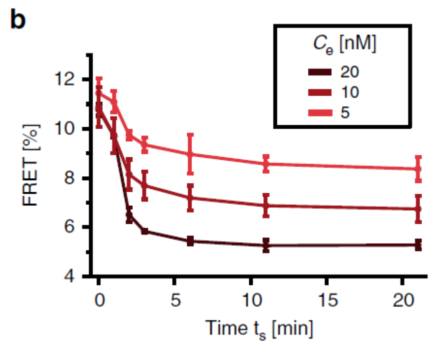
Intracellular FRET-response



Dose-dependent signal processing



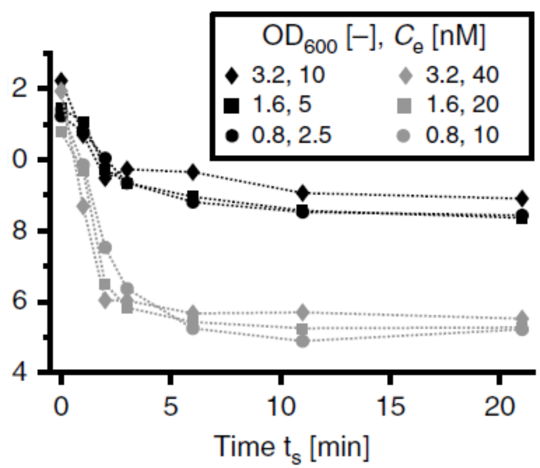


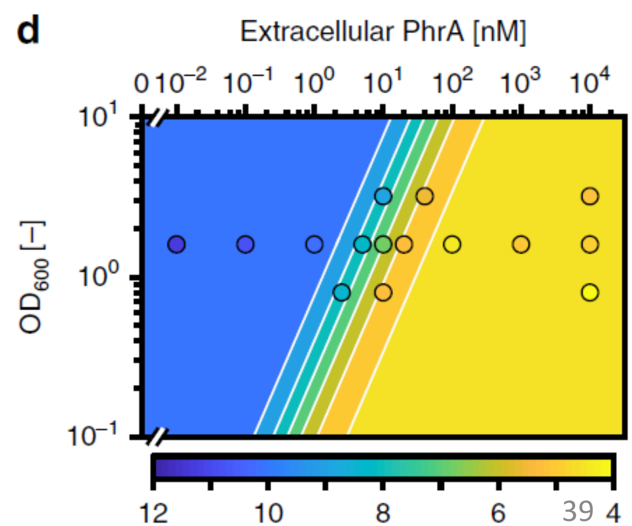


CELL DENSITY EFFECT

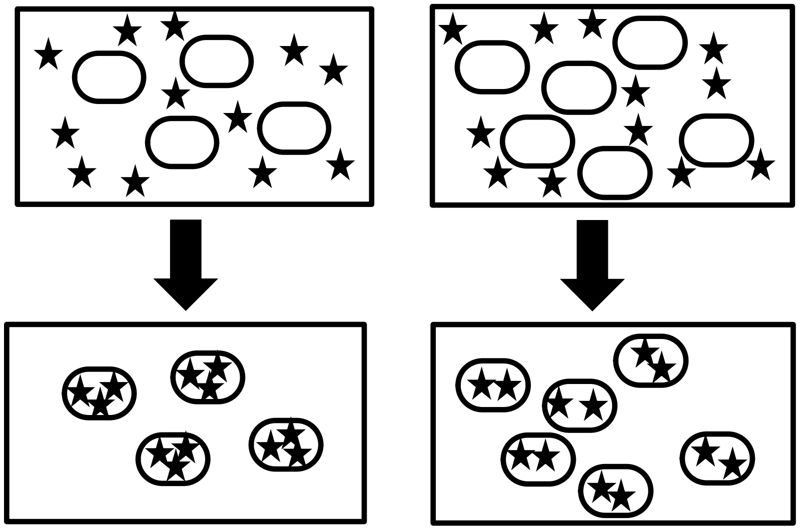
CONCENTRATION EFFECT

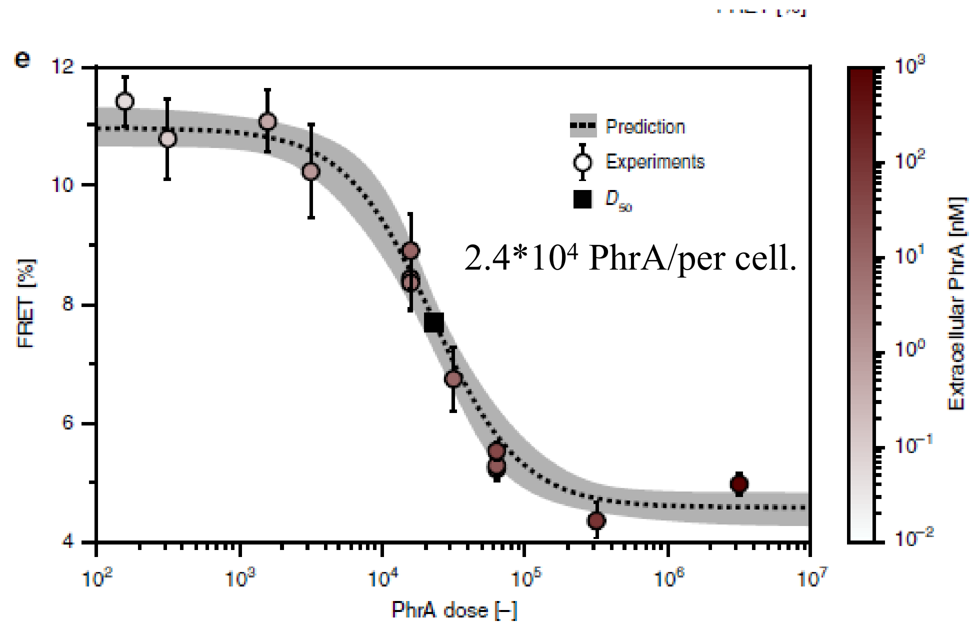
DOSE-DEPENDENT EFFECT



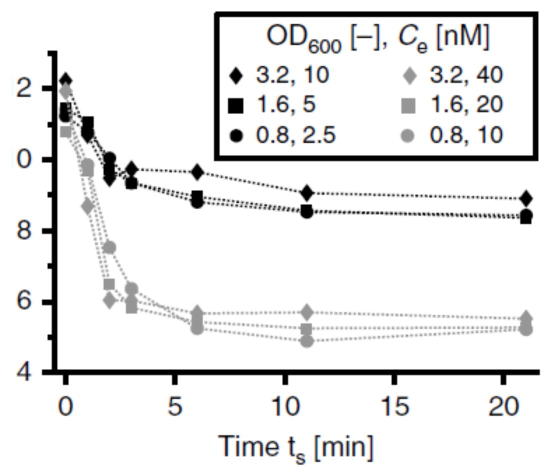


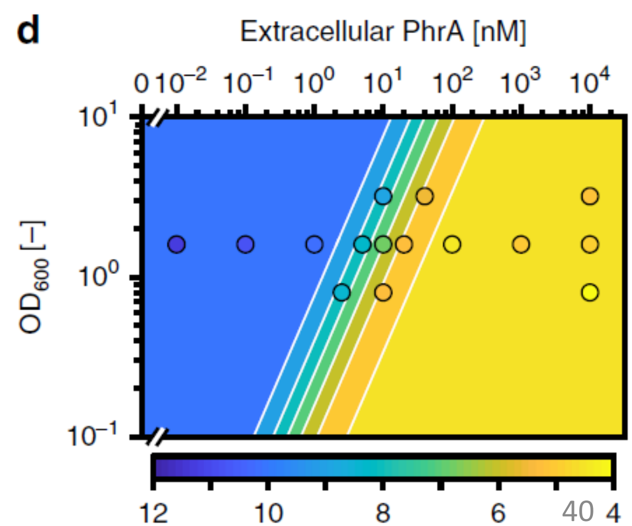
Dose-dependent signal processing



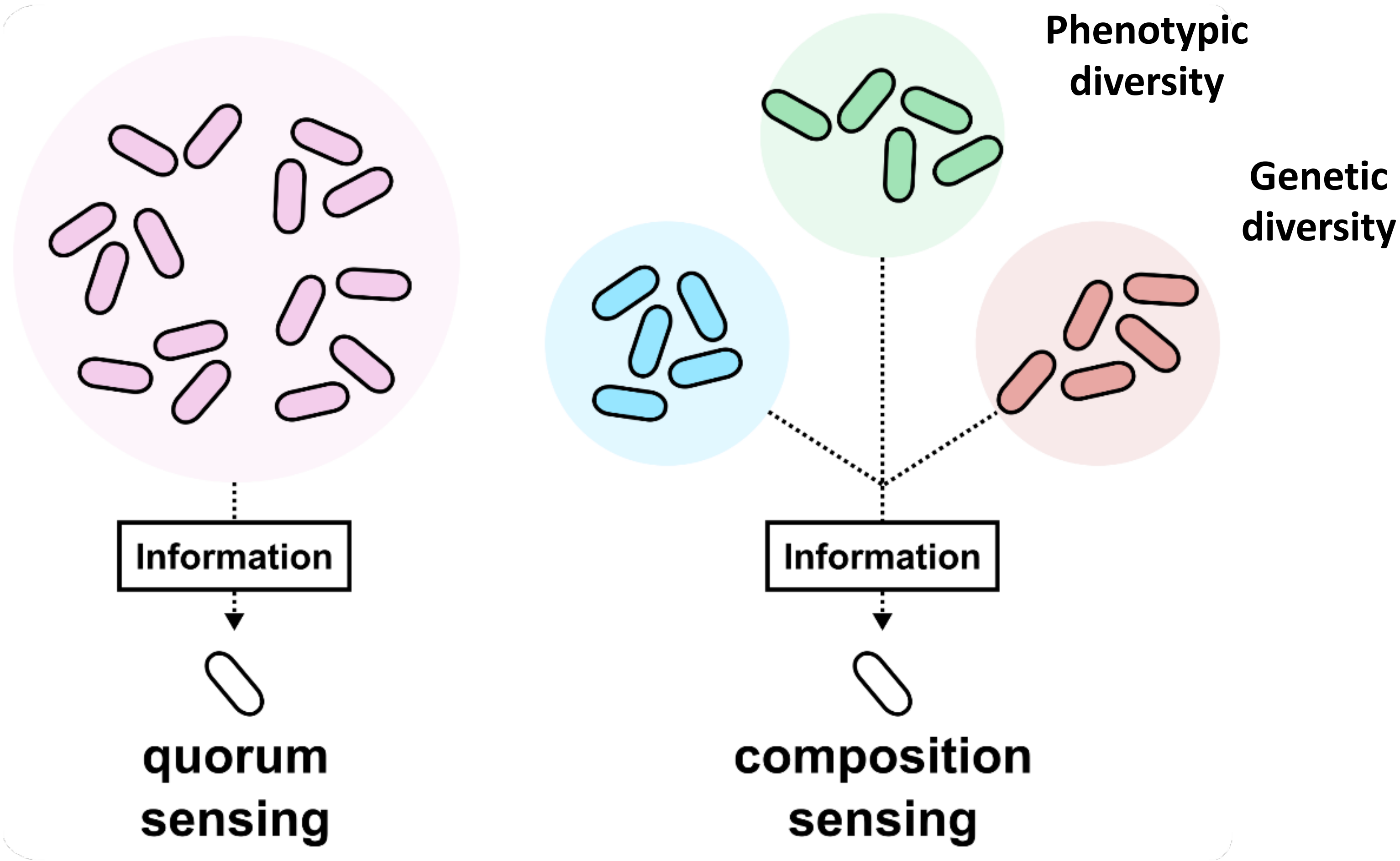


DOSE-DEPENDENT EFFECT



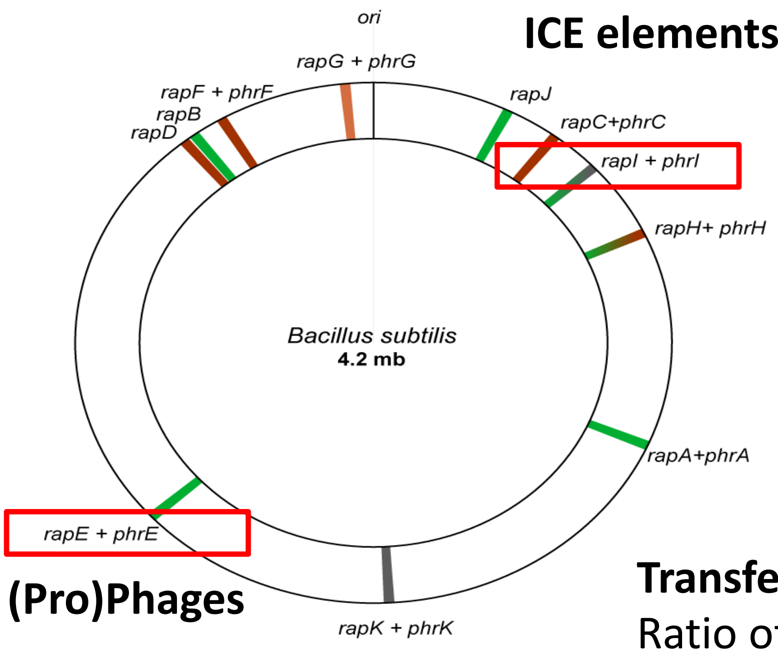


Composition Sensing



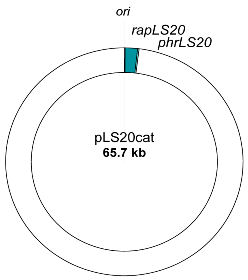
PP Systems: Frequency-dependent regulation

ICE elements



(Pro)Phages

Conguative Plasmids



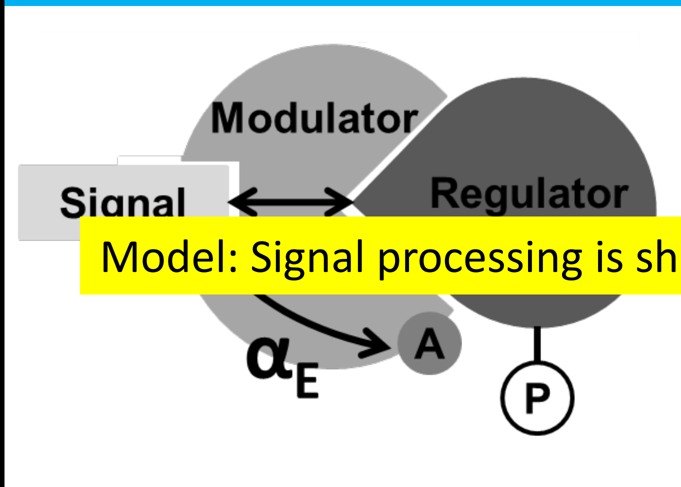
Transfer of genetic material
Ratio of donor and recipient.

Frequency dependent investments into mating in yeast (Banderas et al., 2016)
Conjugation in Enterococcus faecalis, Banderas et al. , BioArXiv 2019.
See also upcoming work by Avigdor Eldar on phages (Tel Aviv University).

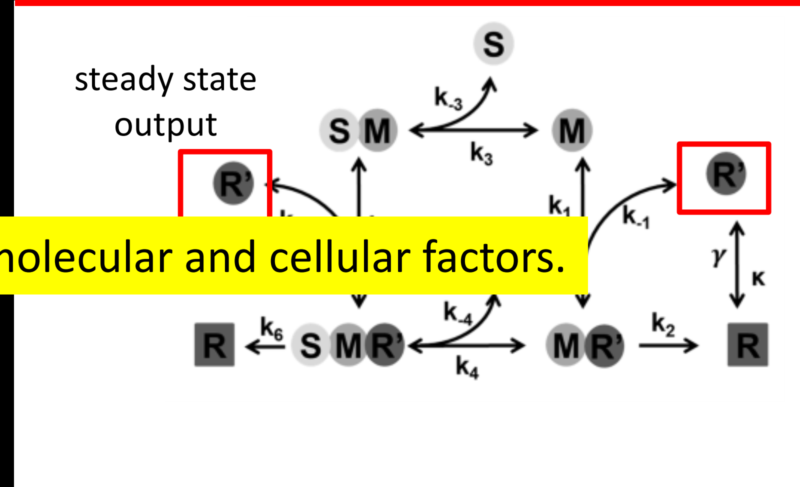
Future work

The Enzymatic Modulator Model

Molecular view



Network motif



Model: Signal processing is shaped by molecular and cellular factors.

Molecular Parameters

Binding Allostery	Enzymatic Allostery	Relative affinity
$\alpha_B = \frac{K'_M}{K_M}$	$\alpha_E = \frac{k_6}{k_2}$	$\beta_E = \frac{K_3}{K_M} = \frac{K_4}{K'_M}$

Cellular Parameters („Context“)

$$\mu = \frac{[M_T]}{[R_T]}$$

$$\kappa_\gamma = \frac{\kappa}{\gamma + \kappa}$$

$$\iota = \frac{k_2}{\gamma + \kappa}$$

$$K_r = \frac{K_M}{[R_T]}$$

Babel & Bischofs, BMC Systems Biology (2016)

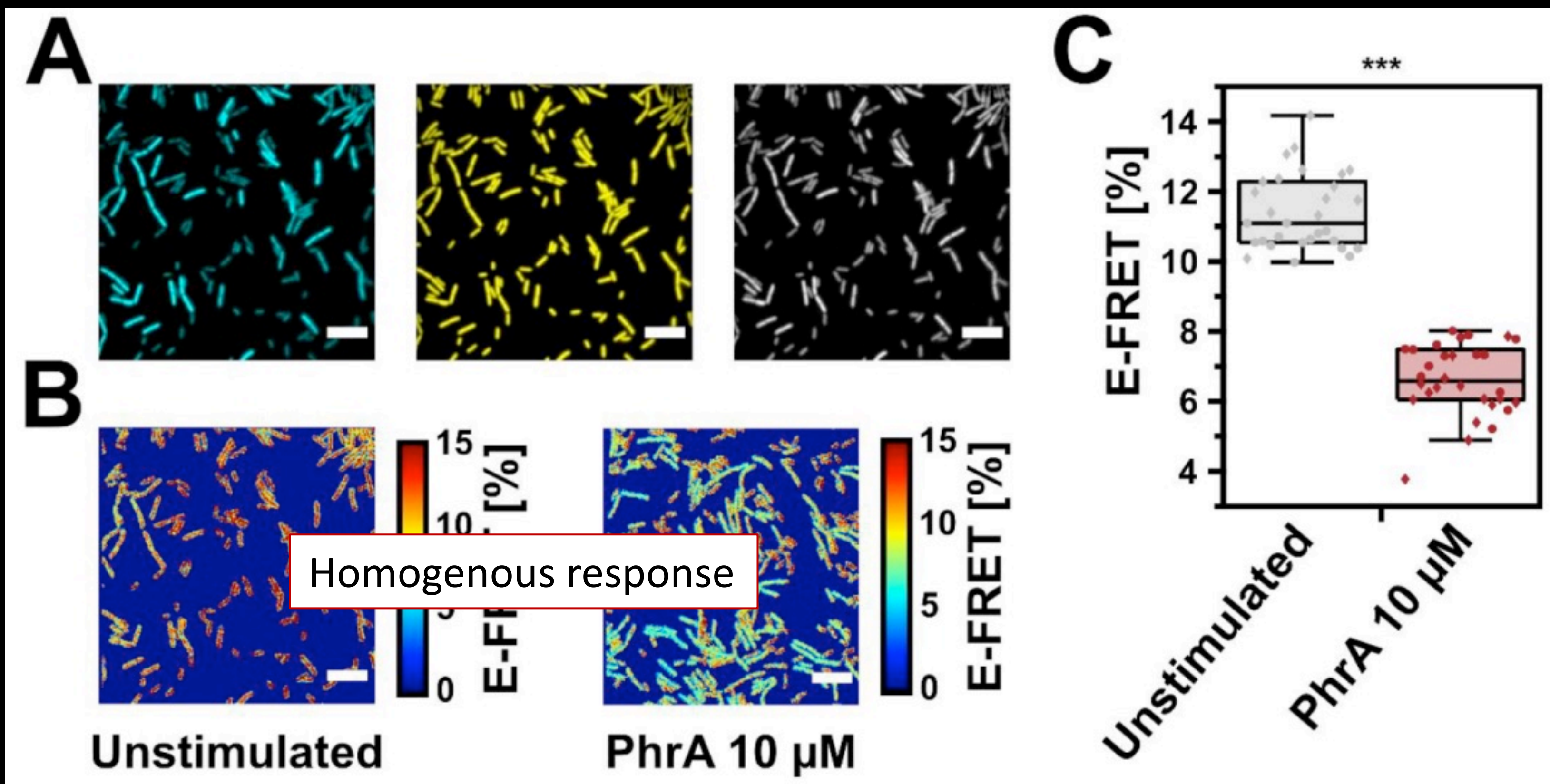
Signal processing by single cells

E-FRET Imaging (Zal & Gascoigne, 2004)

$$E_{app} = \frac{I_{DA} - aI_{AA} - dI_{DD}}{I_{DA} - aI_{AA} + (G - d)I_{DD}}.$$

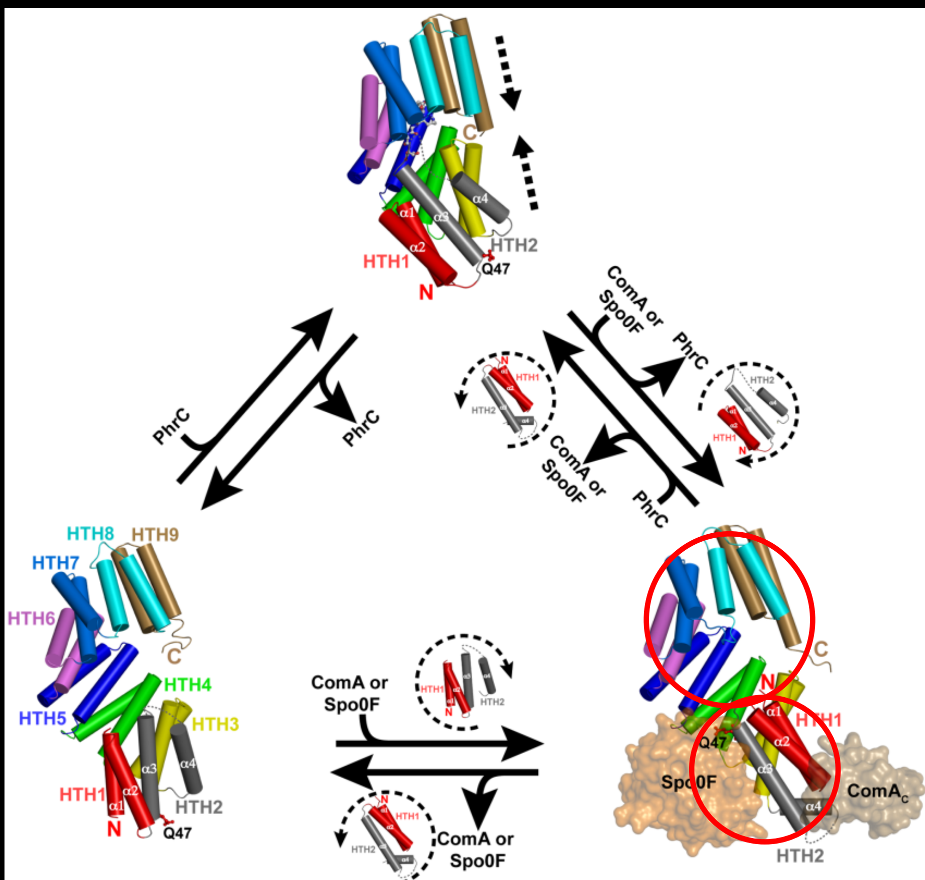
abcd: correction factors

G: calibration factor



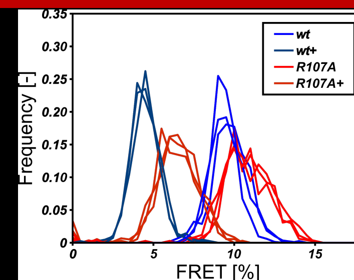
Homogenous response

Signal processing by engineered receptors

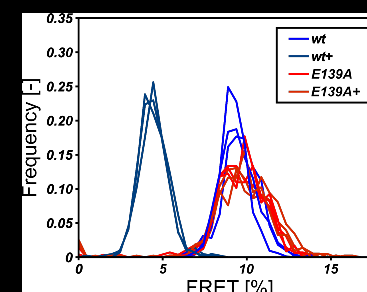


Baker et al., 2011
Parashar et al., 2011
Gallego et al., 2013
Parashar et al., 2013

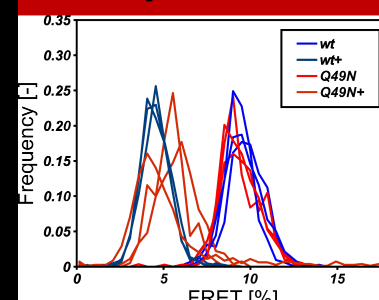
Allosteric residue



Allosteric residue



Catalytic residue



Summary

One sensory function – different architectures

The architecture of QSS could constrain the operating regime for cell density sensing.

One architecture – different sensory functions

Pump-probe networks could implement versatile control functions, including the ability for composition sensing and frequency-dependent regulation.

FRET is a powerful way to quantitatively interrogate signal processing in bacteria by montoring protein-protein interactions in the cell.

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Bastian Drees
Alper Mutlu
Heiko Babel
Sonja Schulmeister
Stephanie Trauth

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