sb2cl Lab
INSTITUTO DE AUTOMÁTICA E INFORMÁTICA INDUSTRIAL
UNIVERSIDAD POLITECNICA DE VALENCIA
Synthetic Biology and Biosystems Control Lab
Valencia UPV

iGEM 2020 Measurement

Modeling: Modeling circuits with ODEs and experimental data

Section 2: Relating parameters and data

by Alejandro Vignoni (alvig2@upv.es)

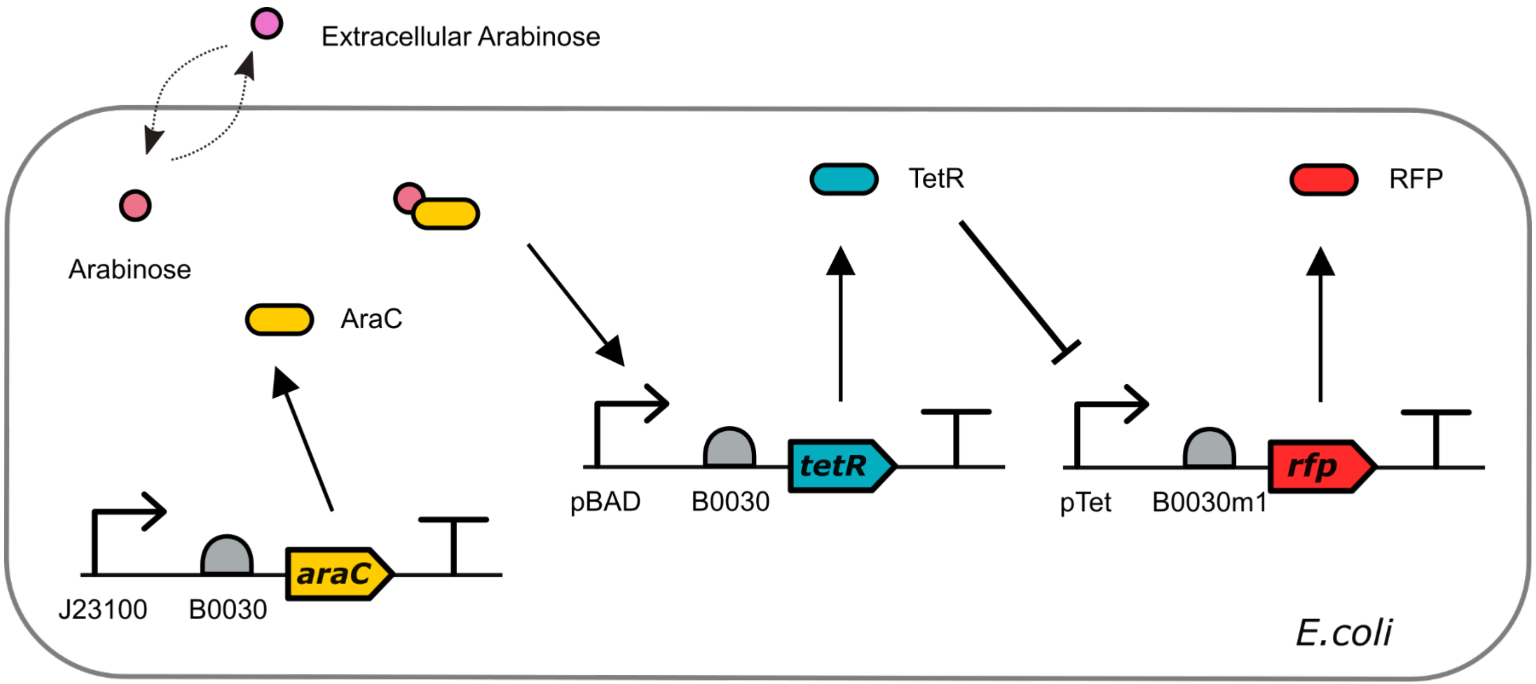
An iGEM Measurement Committee Webinar
Week 3a, June 30th, 2020

Today Webinar’s Topics

- Section 1: Composing circuit models from Hill functions (15 min)
- Section 2: Relating parameters and data (15 min)
- Section 3: Example: Incoherent feed-forward loop (model & data) (15 min)
- Q&A – (at the end of each 15 minutes block, total 15 min)

Relating model parameters and data

Example Sense-Compute-Act

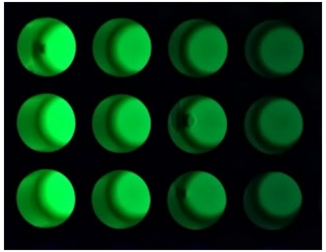


But first we need to get experimental data:

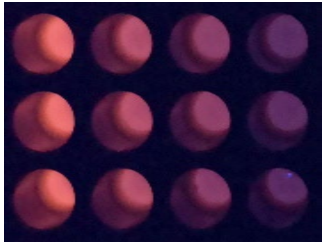
Measurement -> Calibrated measurement

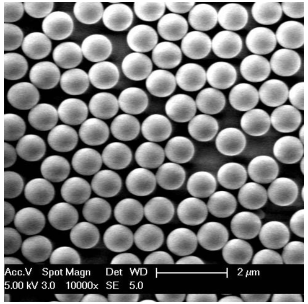
Plate Reader

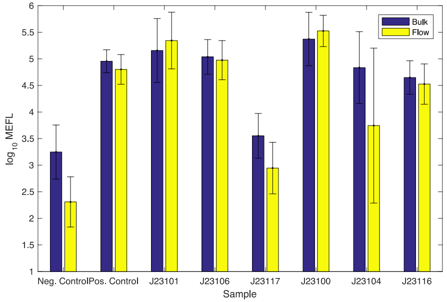
Fluorescein



Texas Red

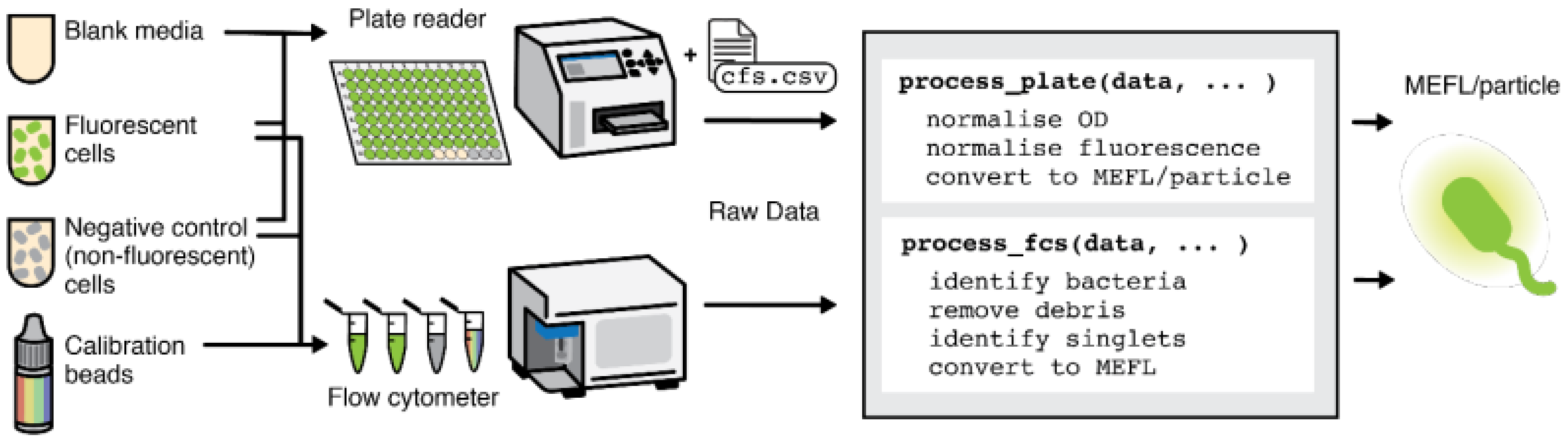






https://2020.igem.org/Measurement/Protocols#validation

Measurement Calibration

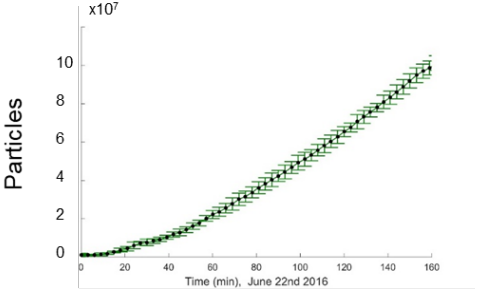


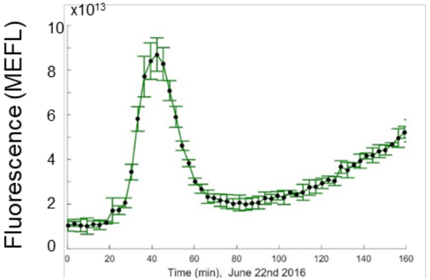
Stay in tune for Measurement Committee Webinars about Calibration:

Week 5 - Tuesday July 14th - 7am EDT - Quantifying fluorescence and cell count with plate readers
Week 6 - Tuesday July 23rd - 7am EDT - Quantifying fluorescence and cell phenotypes with flow cytometry

(Robinson et al, 2020, DOI: 10.1101/2020.06.01.127084)

Why? Because it is exactly what we get from the model



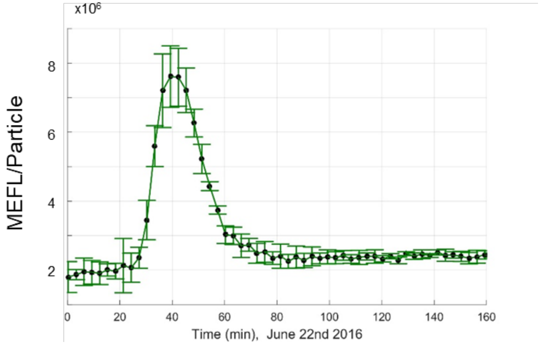


$$\frac{d[R]}{dt} = \frac{\mathrm{p_R C_N k_R}}{\mathrm{dm_R} + \mu} - (\mathrm{d_R} + \mu)\,[R]$$

$$\frac{d[cI]}{dt} = \frac{\mathrm{p_{cI} C_N k_{cI}}}{\mathrm{dm_{cI}} + \mu}\left(\alpha + (1-\alpha)\,\frac{\frac{1}{\mathrm{k_{dlux}}}\left(\frac{[R][A]}{\mathrm{k_{d2} C_N}}\right)^2}{1 + \frac{1}{\mathrm{k_{dlux}}}\left(\frac{[R][A]}{\mathrm{k_{d2} C_N}}\right)^2}\right) - (\mathrm{d_{cI}} + \mu)\,[cI]$$

$$\frac{d[GFP]}{dt} = \frac{\mathrm{p_G C_N k_G}}{\mathrm{dm_G} + \mu}\left(\alpha + (1-\alpha)\,\frac{\frac{1}{\mathrm{k_{dlux}}}\left(\frac{[R][A]}{\mathrm{k_{d2} C_N}}\right)^2}{1 + \frac{1}{\mathrm{k_{dlux}}}\left(\frac{[R][A]}{\mathrm{k_{d2} C_N}}\right)^2}\,\frac{1}{1 + \frac{[cI]^2}{\mathrm{k_{dcI} C_N}}}\right) - (\mathrm{d_G} + \mu)\,[G]$$

$$\frac{dN}{dt} = \mu N\left(1 - \frac{N}{\mathrm{N_{max}}}\right)$$



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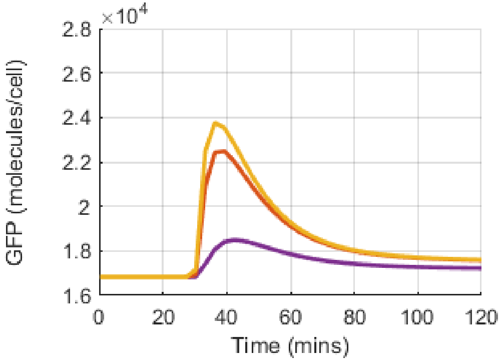
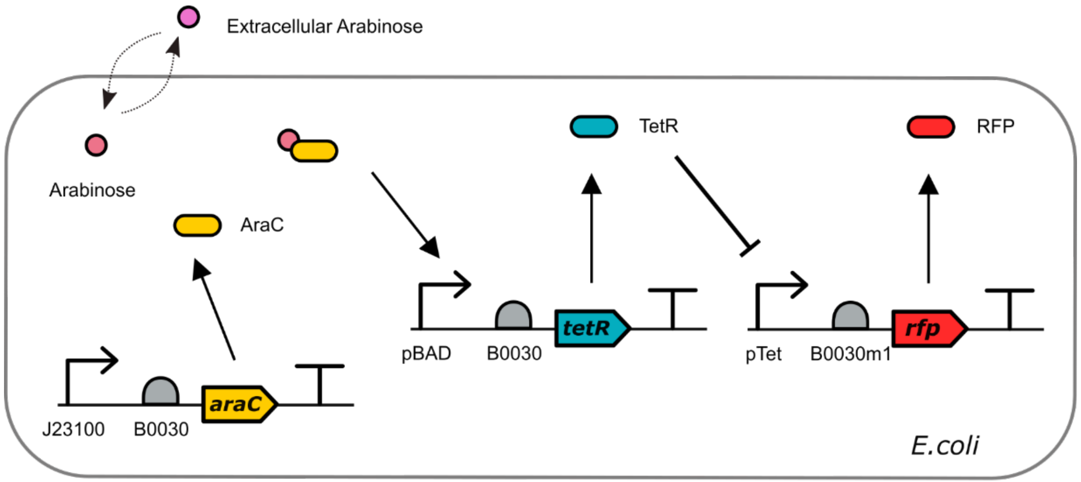


MEFL/Particle unit is equivalent to number of molecules/cell from the mathematical model

Measuring a genetic circuit Example Sense-Compute-Act

SENSE - COMPUTE - ACT

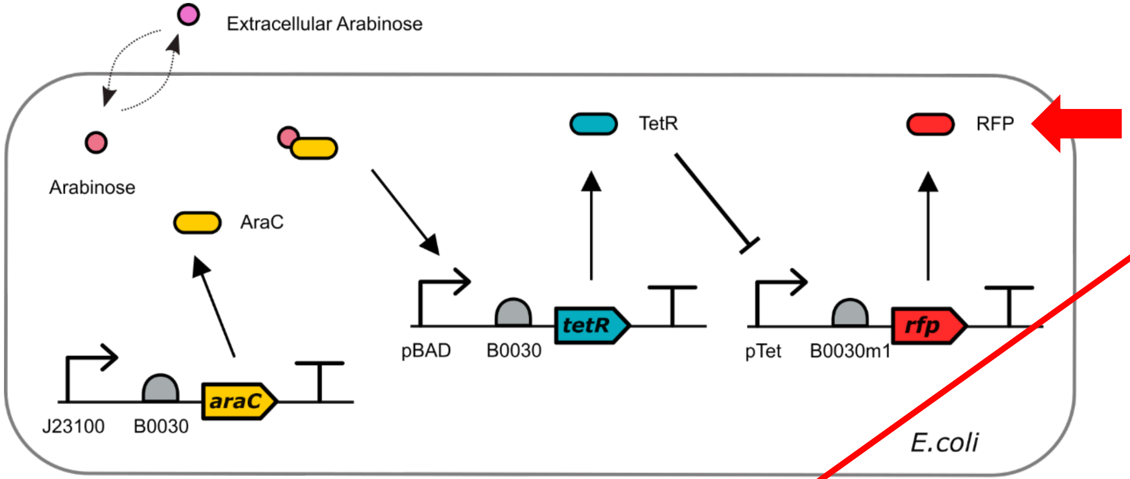


$$[\mathrm{TetR}] = \frac{\alpha_{pBAD}}{d_{\mathrm{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\mathrm{Arab}]^{n_a}}{\left(K_{d_{\mathrm{pBAD}}}\right)^{n_a} + [\mathrm{Arab}]^{n_a}}\right)$$

$$[\mathrm{RFP}] = \frac{\alpha_{\mathrm{pTet}}}{d_{\mathrm{RFP}}}\left(\beta_{o_{\mathrm{pTet}}} + \frac{\left(1-\beta_{o_{\mathrm{pTet}}}\right)[\mathrm{TetR}]^{n_t}}{\left(K_{d_{\mathrm{pTet}}}\right)^{n_t} + [\mathrm{TetR}]^{n_t}}\right)$$

Measuring a genetic circuit Example Sense-Compute-Act

SENSE - COMPUTE - ACT



We can measure only RFP

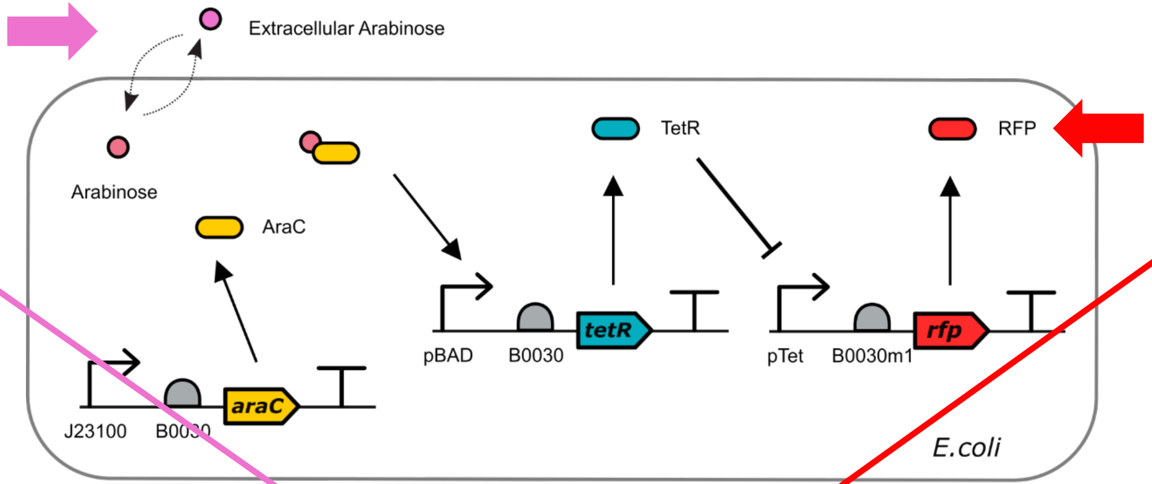
$$[\mathrm{TetR}] = \frac{\alpha_{pBAD}}{d_{\mathrm{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\mathrm{Arab}]^{n_a}}{\left(K_{d_{\mathrm{pBAD}}}\right)^{n_a} + [\mathrm{Arab}]^{n_a}}\right)$$

$$[\mathrm{RFP}] = \frac{\alpha_{\mathrm{pTet}}}{d_{\mathrm{RFP}}}\left(\beta_{o_{\mathrm{pTet}}} + \frac{\left(1-\beta_{o_{\mathrm{pTet}}}\right)[\mathrm{TetR}]^{n_t}}{\left(K_{d_{\mathrm{pTet}}}\right)^{n_t} + [\mathrm{TetR}]^{n_t}}\right)$$

Measuring a genetic circuit Example Sense-Compute-Act

SENSE - COMPUTE - ACT

We can only change the amount of Arabinose



We can measure only RFP

$$[\mathrm{TetR}] = \frac{\alpha_{pBAD}}{d_{\mathrm{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\mathrm{Arab}]^{n_a}}{\left(K_{d_{\mathrm{pBAD}}}\right)^{n_a} + [\mathrm{Arab}]^{n_a}}\right)$$

$$[\mathrm{RFP}] = \frac{\alpha_{\mathrm{pTet}}}{d_{\mathrm{RFP}}}\left(\beta_{o_{\mathrm{pTet}}} + \frac{\left(1-\beta_{o_{\mathrm{pTet}}}\right)[\mathrm{TetR}]^{n_t}}{\left(K_{d_{\mathrm{pTet}}}\right)^{n_t} + [\mathrm{TetR}]^{n_t}}\right)$$

Measuring a genetic circuit Example Sense-Compute-Act

SENSE - COMPUTE - ACT

We can only change the amount of Arabinose

We need more!!

What can we do?

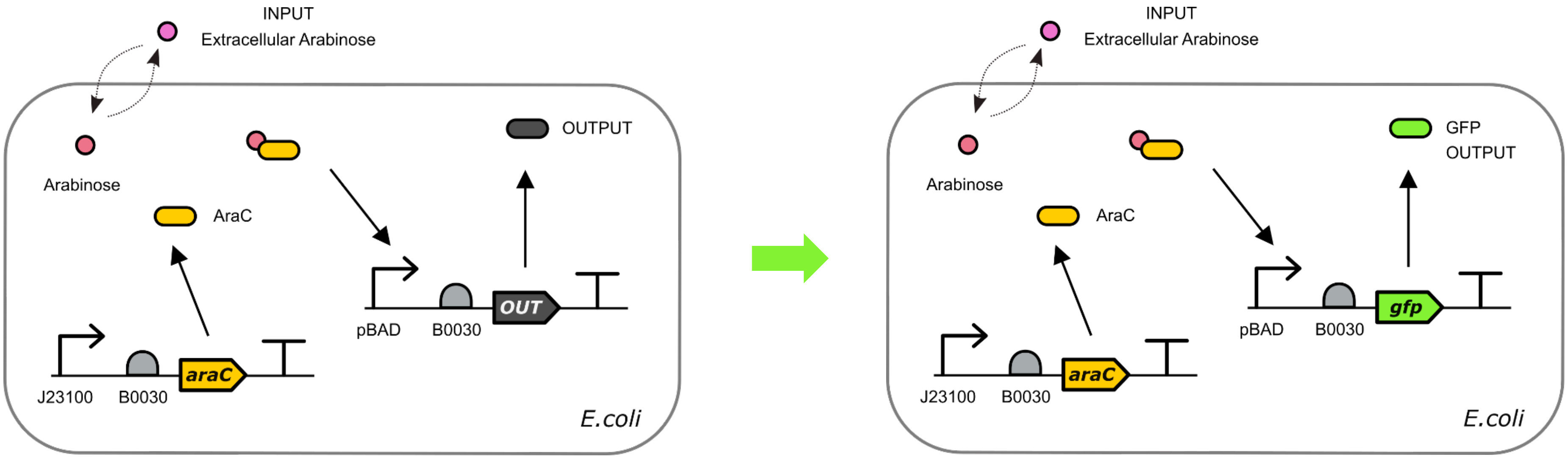
We can measure only RFP

$$[\mathrm{TetR}] = \frac{\alpha_{pBAD}}{d_{\mathrm{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\mathrm{Arab}]^{n_a}}{\left(K_{d_{\mathrm{pBAD}}}\right)^{n_a} + [\mathrm{Arab}]^{n_a}}\right)$$

$$[\mathrm{RFP}] = \frac{\alpha_{\mathrm{pTet}}}{d_{\mathrm{RFP}}}\left(\beta_{o_{\mathrm{pTet}}} + \frac{\left(1-\beta_{o_{\mathrm{pTet}}}\right)[\mathrm{TetR}]^{n_t}}{\left(K_{d_{\mathrm{pTet}}}\right)^{n_t} + [\mathrm{TetR}]^{n_t}}\right)$$

Measuring a genetic circuit Example Sense-Compute-Act

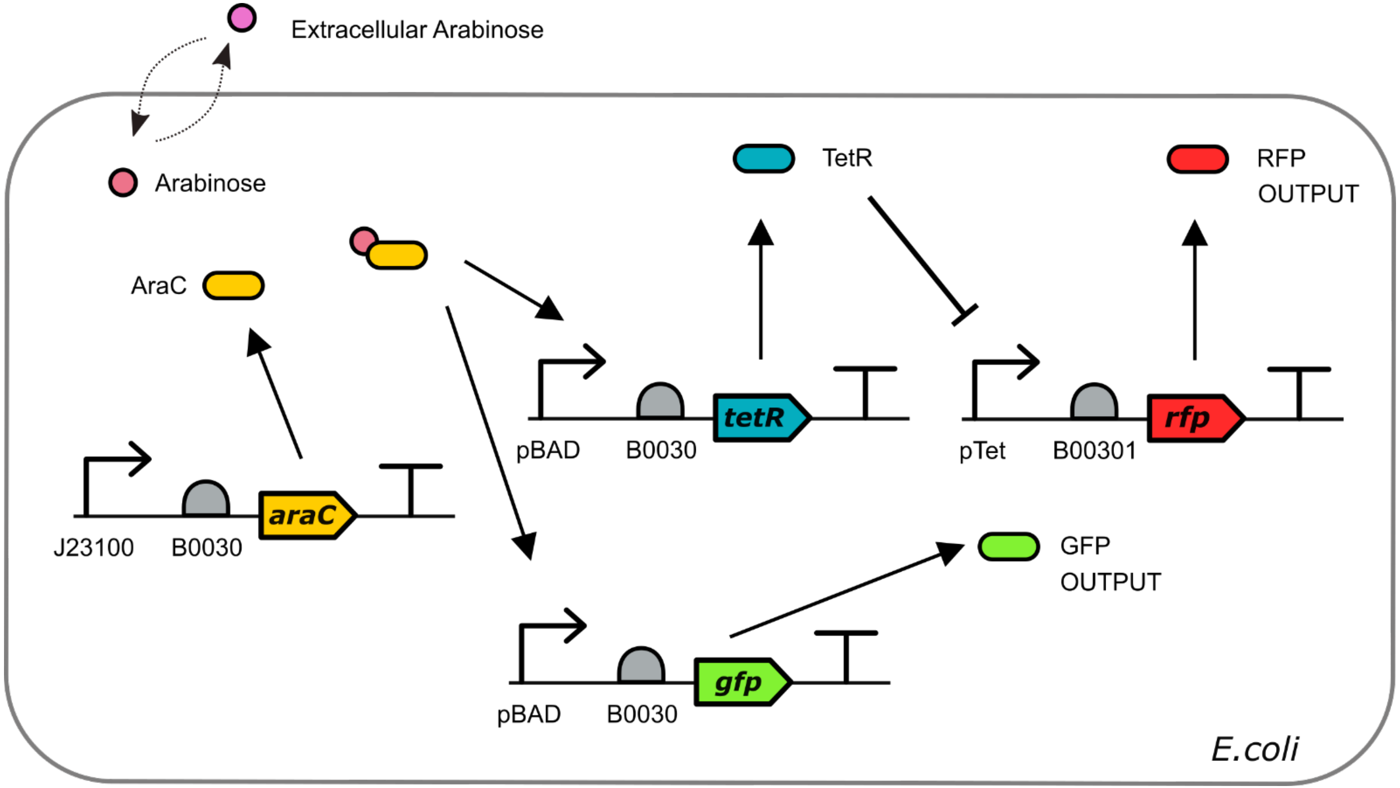
SENSE



We can make another construct, with GFP as OUTPUT.

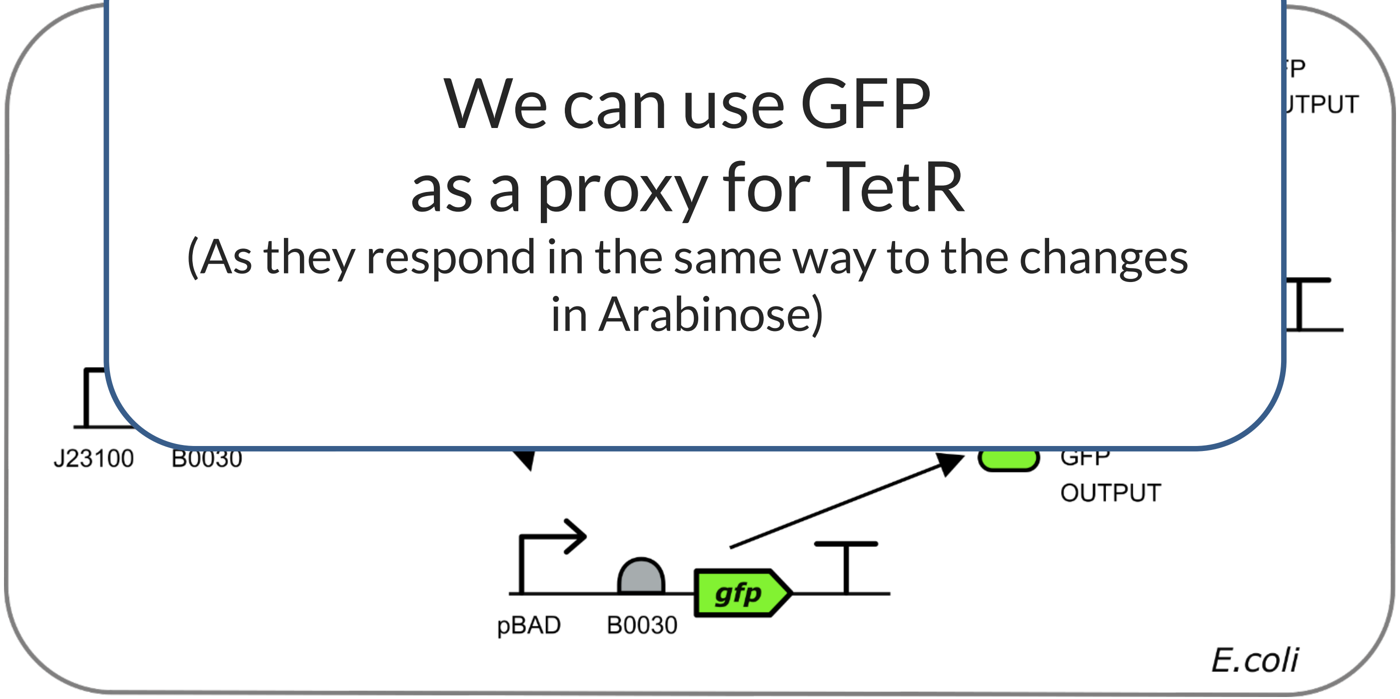
Measuring a genetic circuit Example Sense-Compute-Act

SENSE - COMPUTE - ACT for measurement



Measuring a genetic circuit Example Sense-Compute-Act

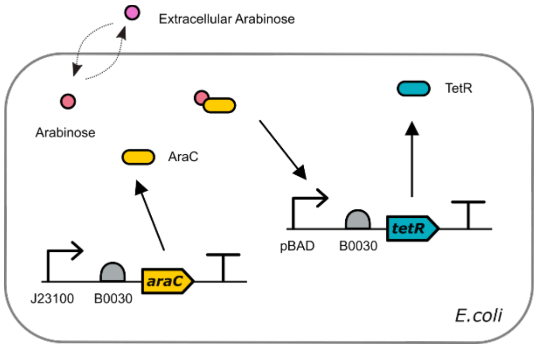
SENSE - COMPUTE - ACT for measurement



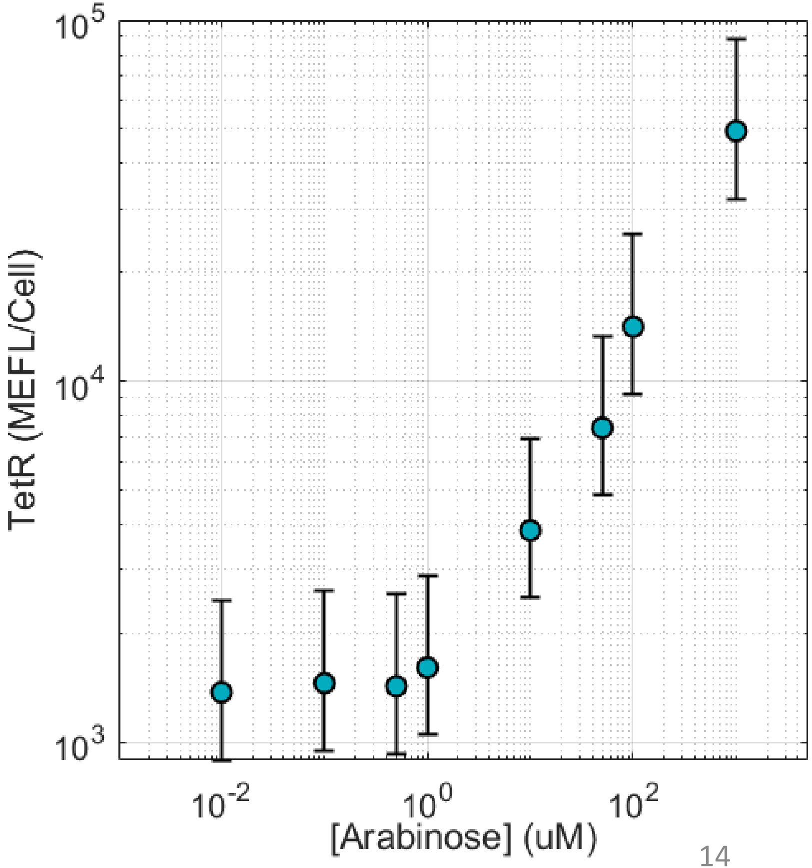
We can use GFP
as a proxy for TetR
(As they respond in the same way to the changes in Arabinose)

Modeling a genetic circuit Example Sense-Compute-Act

SENSE

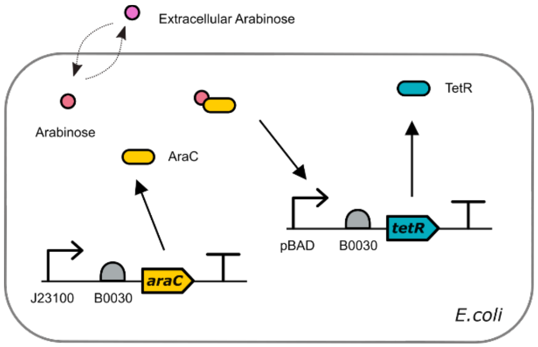


We make experiments with 8 different levels of Arabinose induction, measure GFP (TetR) in a flowcytometer and calibrate the mesurement.



Modeling a genetic circuit Example Sense-Compute-Act

SENSE

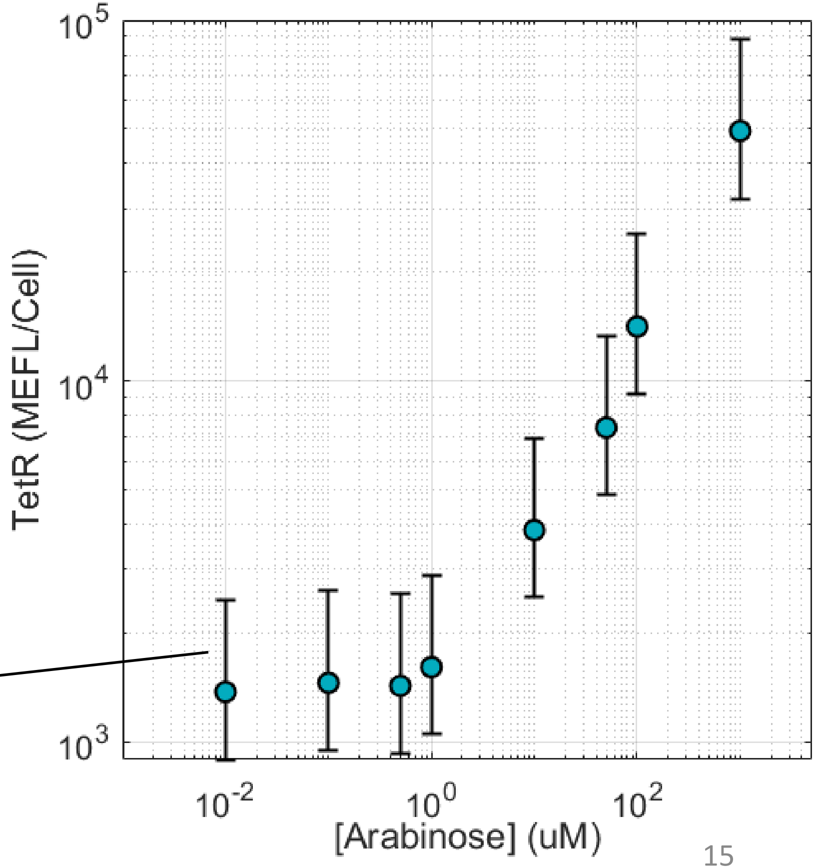


$$[\mathrm{TetR}] = \frac{\alpha_{pBAD}}{d_{\mathrm{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\mathrm{Arab}]^{n_a}}{\left(K_{d_{\mathrm{pBAD}}}\right)^{n_a} + [\mathrm{Arab}]^{n_a}}\right)$$

$$\mathrm{Error} = \frac{1}{m}\sum_{i}^{m}\left([TetR]_{model,\,i} - [TerR]_{measured,\,i}\right)^2$$

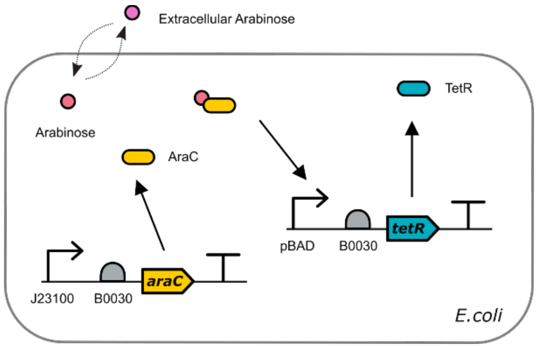
For the m different concentrations of Arabinose.

Then we minimize the error…



Modeling a genetic circuit Example Sense-Compute-Act

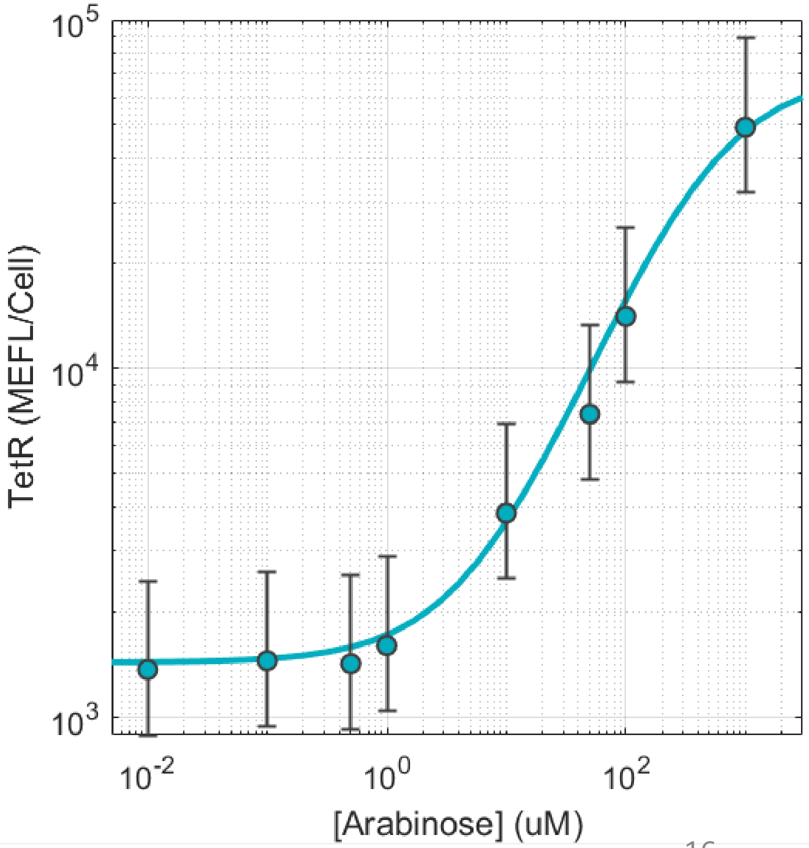
SENSE



$$[\text{TetR}] = \frac{\alpha_{pBAD}}{d_{\text{TetR}}}\left(\beta_{o_{pBAD}} + \frac{\left(1-\beta_{o_{pBAD}}\right)[\text{Arab}]^{n_a}}{\left(K_{d_{\text{pBAD}}}\right)^{n_a} + [\text{Arab}]^{n_a}}\right)$$

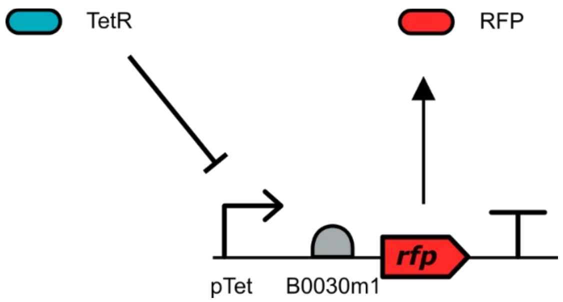
$$\frac{\alpha_{pBAD}}{d_{\text{TetR}}} = 7.056 \times 10^4 \text{molecules}$$

$K_{d_{pBAD}} = 444.5\mu M$ $\beta_{o_{pBAD}} = 0.02$ $n_a = 1$



Modeling a genetic circuit Example Sense-Compute-Act

COMPUTE - ACT



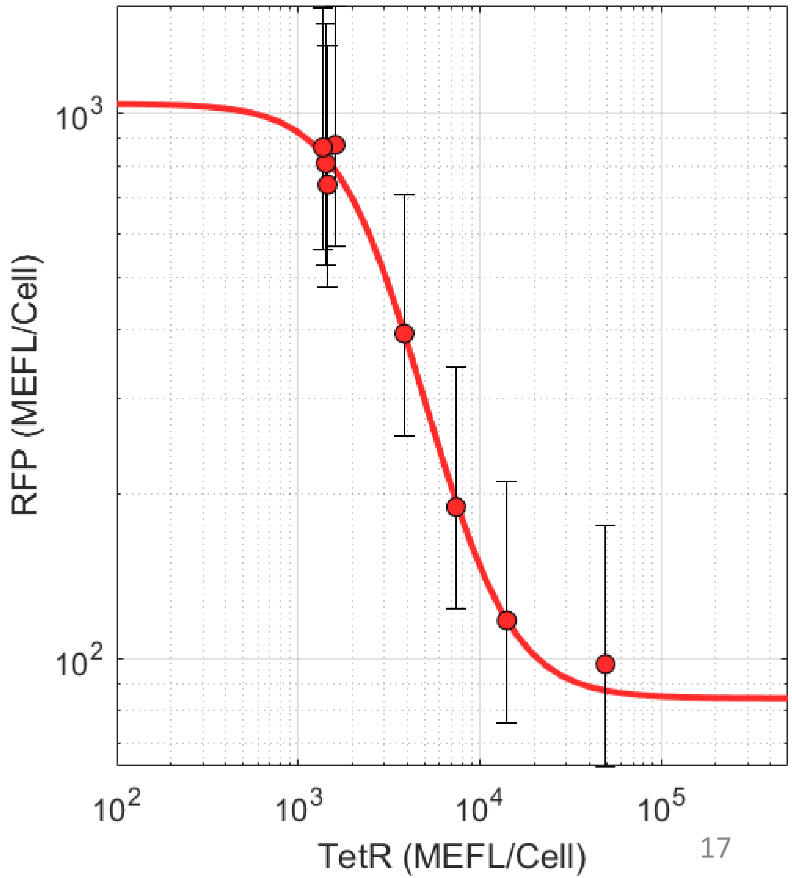
$$[\mathrm{RFP}] = \frac{\alpha_{\mathrm{pTet}}}{d_{\mathrm{RFP}}}\left(\beta_{o_{\mathrm{pTet}}} + \frac{\left(1-\beta_{o_{\mathrm{pTet}}}\right)[\mathrm{TetR}]^{n_t}}{\left(K_{d_{\mathrm{pTet}}}\right)^{n_t} + [\mathrm{TetR}]^{n_t}}\right)$$

$$\frac{\alpha_{pTet}}{d_{\mathrm{RFP}}} = 1039 \text{ molecules}$$

$$\beta_{o_{pBAD}} = 0.08$$

$$K_{d_{pTet}} = 2668 \text{ molecules}$$

$$n_t = 2$$



Questions?
Ask writing in the chat or contact me by email (alvig2 [at] upv [dot] es)

Stay tuned, next Section 3:

Example: Incoherent feed-forward loop (model & data)