ChemE 5440/7770: Problem Set 2

Sneha Kabaria

February 19, 2019

Problem 1

Sensitivity Analysis was performed using the Julia file (found in github repository). Based on graphed results during sensitivity analysis. The table below describes the behavior of the mRNA concentration as a function of [I] at different values of [I] due to an increase in the parameter of the first column. Explanations of the significance of changers are summarized below the graph.

Parameter Increased	Low [I]	Mid [I]	High [I]
Lj (length mRNA)	started higher	steeper slope	shift up
kI	started higher	shift left/steeper slope	shift up
Kej	no change	no change	no change
Tau	shift down	shift right/less steep slope	shift down
kej	shift up	shift left/steeper slope	shift up
RNAP total	shift up	shift left/steeper slope	shift up
Gj	negligible change	negligible change	negligible change
Km	negligible change	shifted right/same slope	negligible change
k1	started higher	shifted left/same slope	negligible change
k2	negligible effect	slope shifted left/same slope	negligible change
k_d	shift down	shift right/less steep slope	shift down
mu	negligible change	negligible change	shift up

Lj = length of mRNA

kI = transcription initiation rate

Kej = saturation transcription

tau = time constant

kej = transcription rate

RNAP total = total RNAP in the cell

Gj = copy in the cell

Km = affinity of the inducer

k1 = basal transcription level (leaky expression)

k2 = weight of the inducer effect

kd = mRNA degradation rate

mu = dilution rate

The Appendix contains the graphs used to make all of the conclusions. Half and double the original amount were used in all cases to look at the difference of changing the parameters.

The mRNA concentration at "low [I]" starting higher means that the initial mRNA concentration before the inducer had significant effect increasing the basal level of mRNA production (leaky expression).

For "mid [I]" -The slope refers to the critical range of [I] needed for the drastic increases in mRNA concentration – meaning a steep slope indicates a narrower range for I that causes a dramatic increases (in others words, more sensitivity.) In the [I] range around the slope, a steep slopes indicates that mRNA concentration is very sensitive to small changes in [I].

For "mid [I]" If the line is shifted left, it is good, because it means that a lower concentration of I is needed for a signification induction of mRNA transcription.

A higher final mRNA (shift up) at the range "high [I]" means that the maximum equilibrium concentration of mRNA, and hence maximum velocity of mRNA production was higher due to an increase in the parameter in the first column. For "high [I]" if there is little difference, it means that there is negligible effect of the increase of the parameter on the final maximum mRNA equilibrium concentration.

Problem 2

IMPORTANT NOTE:

Code contains values of all parameters and full equations. This is the high level summary.

Assumptions:

–Promoter control models Moon/Voigt formulation

–translation operates a the kinetic limit

–RNAP and Ribosome levels are constant

Knowns:

doubling time = 30 min

mass of cell water = 70 %

plasmid copies = 200

concentration basis = umol gDW-1

L_X = 1000 nt (characteristic length)

L_T = 333 AA (characteristic length)

$L_{X,1}$ = 1200 nt

$L_{X,2}$ = 2400 nt

$L_{X,3}$ = 600 nt

$L_{L,i} = (1/3)L_{X,i}$

Equations for Mass Balances:

$$\frac{dm_i}{dt} = TX - k_deg * m_i - \mu_dilution * m_i \tag{1}$$

$$\frac{dp_i}{dt} = TL - k_deg * p_i - \mu_dilution * p_i \tag{2}$$

where mi is the mRNA, and pi is the protein, and i = 1,2,3 for both mRNAs and proteins. Transcription rate is:

$$TX = k_(e, j) * \frac{G_j * RNAP_T}{\tau_x * K_x, j + (\tau_x + 1) * G_j} * u \tag{3}$$

and translations rate is:

$$TL = k_e, L * ribosomes_T * \frac{m_i}{]tau_L * K_L + (\tau_L + 1) * m_i} * u \tag{4}$$

Everything before u in both TX and TL is the maximum rate. u is the control function. For mRNA, the u function is not equal to one (see julia code for 2b "srkps2.jl"):

$$u! = 1 \tag{5}$$

For mRNA, the u function is equal to one, because it is at the kinetic limit (see julia code for 2b "srkps2.jl"):

$$u = 1 \tag{6}$$

FULL FORM OF EQUATIONS ARE IN THE CODES.

Matrices - Answer to 2a

In matrix form the final equations are:

$$\frac{d\mathbf{x}}{dt} = \mathbf{A}\mathbf{x} + \mathbf{S}\mathbf{r} \tag{7}$$

where

$$\mathbf{A} = \begin{pmatrix} -\mu - kd_m & 0 & 0 & 0 & 0 & 0 \\ 0 & -\mu - kd_m & 0 & 0 & 0 & 0 \\ 0 & 0 & -\mu - kd_m & 0 & 0 & 0 \\ 0 & 0 & 0 & -\mu - kd_p & 0 & 0 \\ 0 & 0 & 0 & 0 & -\mu - kd_p & 0 \\ 0 & 0 & 0 & 0 & 0 & -\mu - kd_p \end{pmatrix}. \tag{8}$$

(kdm is degradation rate of mRNA, kdp is degradation rate of protein, mu is dilution rate.)
(m's are the mRNA, and p's are the proteins) and,

$$\mathbf{x} = \begin{pmatrix} m_1 \\ m_2 \\ m_3 \\ p_1 \\ p_2 \\ p_3 \end{pmatrix}. \tag{9}$$

and

$$\mathbf{S} = \begin{pmatrix} 1 & 0 & 0 & 0 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 0 & 0 & 1 \end{pmatrix}. \tag{10}$$

$$\mathbf{r} = \begin{pmatrix} TX_1 \\ TX_2 \\ TX_3 \\ TL_1 \\ TL_2 \\ TL_3 \end{pmatrix}. \tag{11}$$

where TX is the transcription rate, and TL is the translation rate.

Graphs - Answer to 2b

Full code is in both "srkps2.jl" which runs the odes encoded "srkps2balances.jl"

Images are uploaded and labeled.

p1 = black

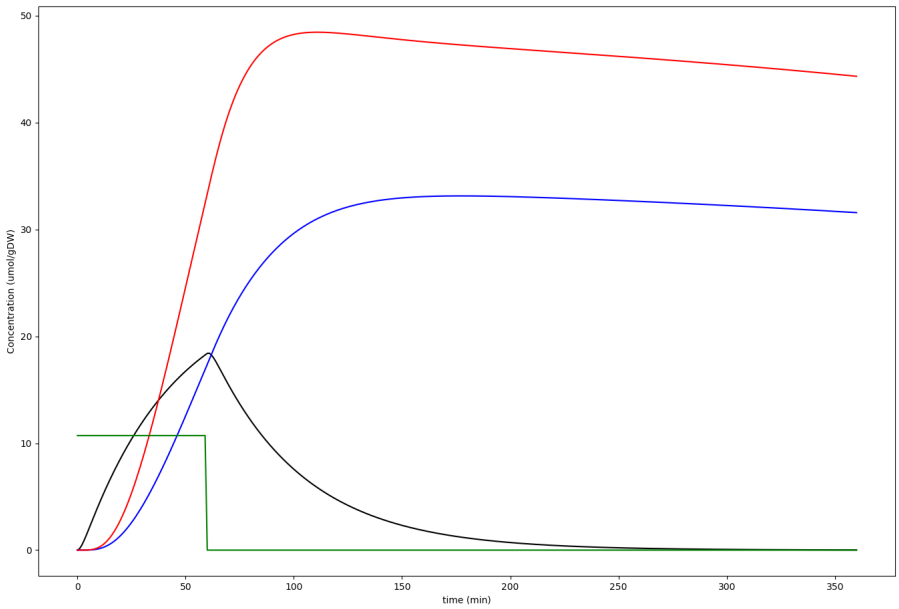
p2 = blue

p3 = red

I = green (scaled)

y-axis = umol/gDW ; x-axis = time (min)

i. not broken circuit

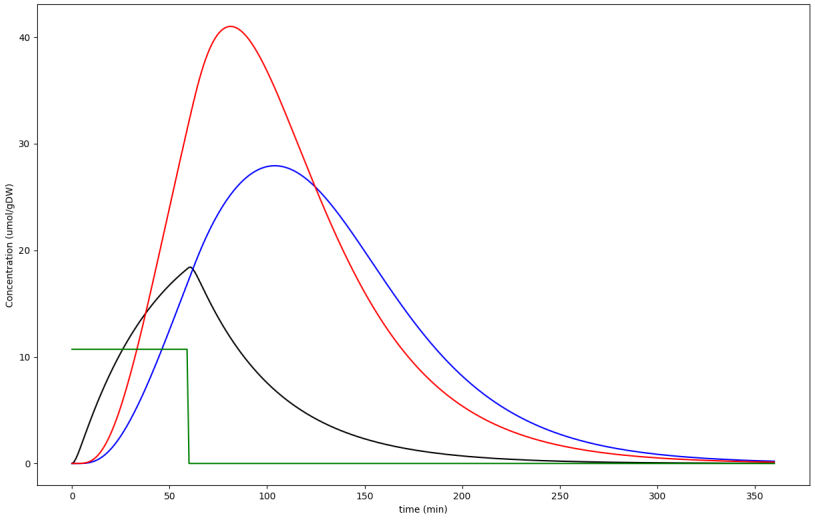


]

Since p1 was only induced by I, removing I causes p1 to drop significantly right away. Since p2 and p3 induce each other, they only decrease slightly after taking away p1, and reach a steady equilibrium.

Drops are due to degradation and dilution of the proteins.

ii. broken circuit



]

Since p1 was only induced by I, removing I causes p1 to drop significantly right away. p2 no longer induces p3, so p3 descreases next when p1 is gone. Since p3 does induce p2, it takes some time before the decrease in p2 is enough to decrease induction of p3. p3 eventually falls as well.

Drops are due to degradation and dilution of the proteins.

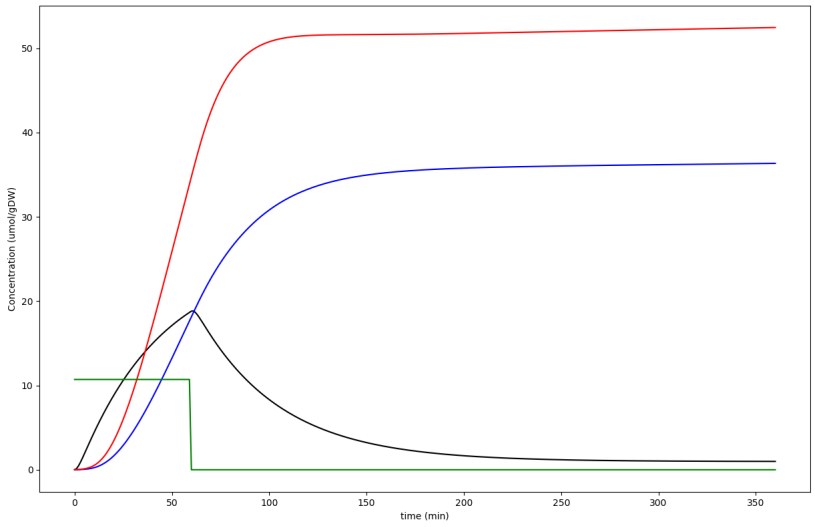
iii. Why does it go to zero?

Basal transcription level (not induced) was set really low (weight = .008), which is why the protein 1 concentration drops to nearly 0 and is not readable on the graph.

Essentially, you assume there is negligible basal transcription.

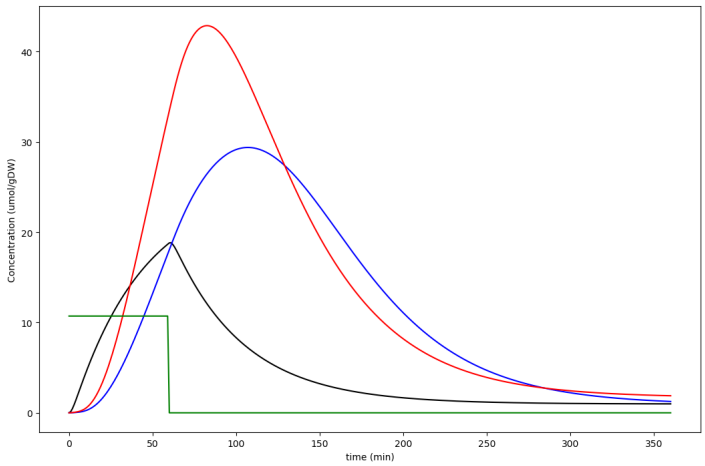
If basal transcription weight were set higher, protein concentrations would have reached equilibrium as some amount a bit more greater than 0.

– i, basal weight = 0.01, not broken circuit



basal.PNG]

– ii, basal weight = 0.01, broken circuit


Concentration (umol/gDW)
time (min)
0
10
20
30
40
0
50
100
150
200
250
300
350

]

Graphs - Answer to 2c

Full code is in both "srkps2-2c.jl" which runs and graphs the "supercool" math. "srkps2.jl" is used to overlay with the answers to 2b.

Images are uploaded and labeled.

p1 = purple

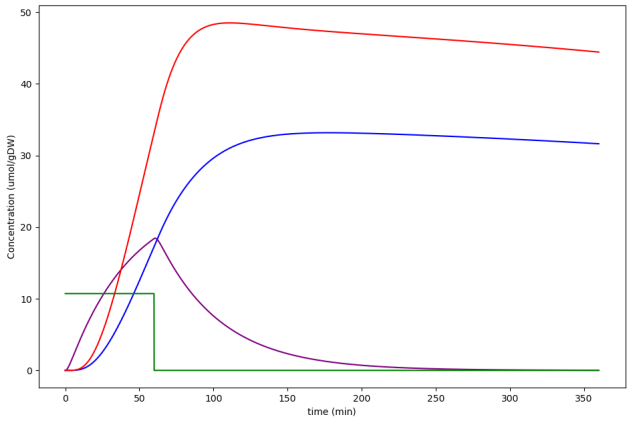
p2 = blue

p3 = red

I = green (scaled)

y-axis = umol/gDW ; x-axis = time (min)

i. not broken circuit

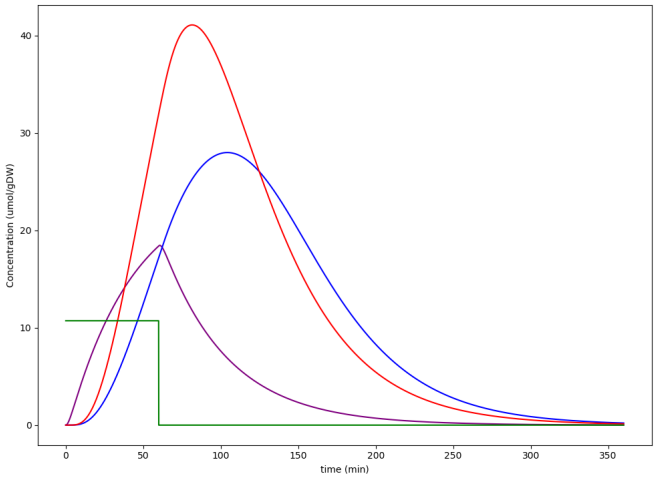


]

Since p1 was only induced by I, removing I causes p1 to drop significantly right away. Since p2 and p3 induce each other, they only decrease slightly after taking away p1, and reach a steady equilibrium.

Drops are due to degradation and dilution of the proteins.

ii. broken circuit



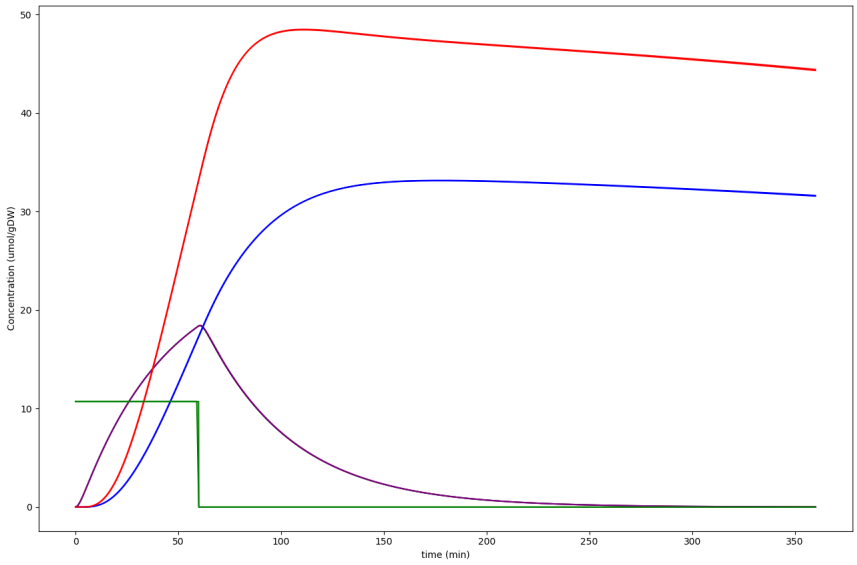
]

Since p1 was only induced by I, removing I causes p1 to drop significantly right away. p2 no longer induces p3, so p3 descreases next when p1 is gone. Since p3 does induce p2, it takes some time before the decrease in p2 is enough to decrease induction of p3. p3 eventually falls as well.

Drops are due to degradation and dilution of the proteins.

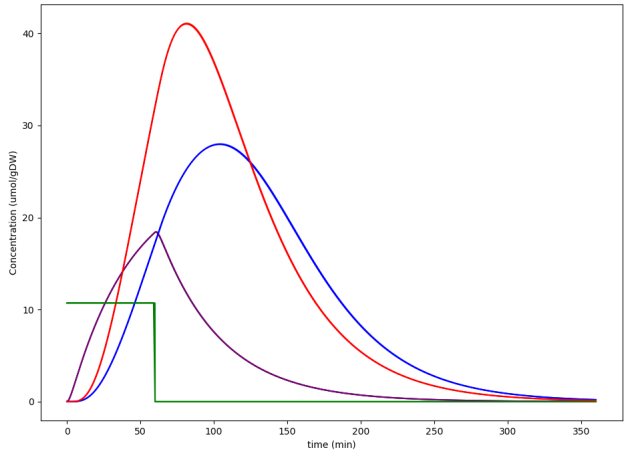
Graphs with the method from 2c overlaid almost perfectly with those from 2a. This shows that the supercool math trick works well.

i. not broken circuit overlay



]

ii. broken circuit overlay



]

iii. Why does it go to zero?

As with 2b, in 2c the basal weight was set really low in order to really observe the effect of the inducer and p1 (weight = 0.008). Essentially, you assume there is negligible basal transcription.

Appendix: Graphs for Problem 1

For all graphs:

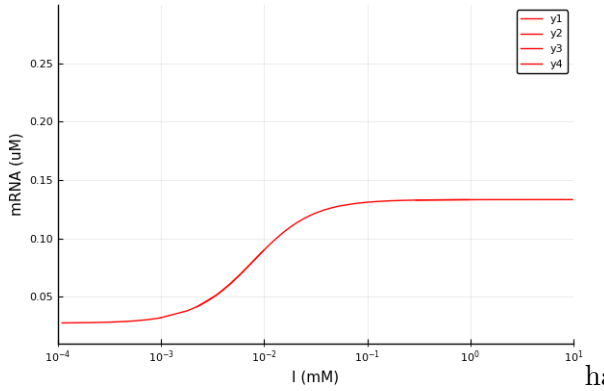
Green means parameter was doubled.

Red means parameter was halved.

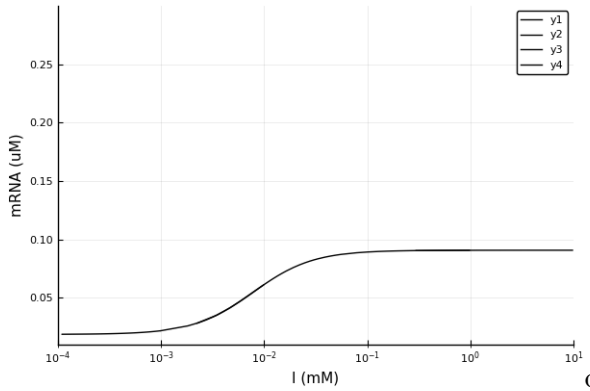
Black means the original parameter values.

All parameter values are shown in the code "jv-ps1-solution.jl"

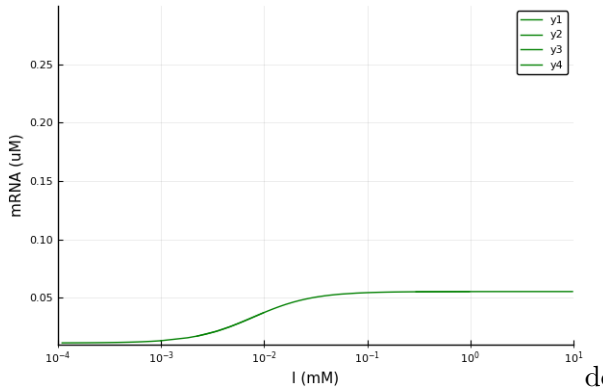
Lj = length of mRNA



half



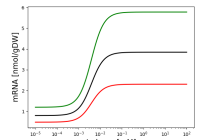
original



double

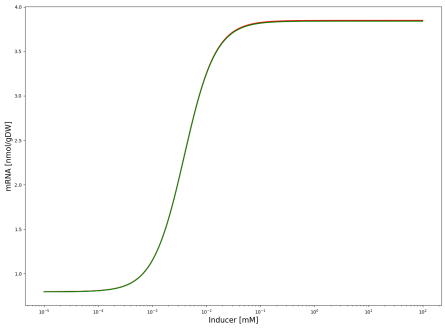
---

kI = transcription initiation rate



change.PNG

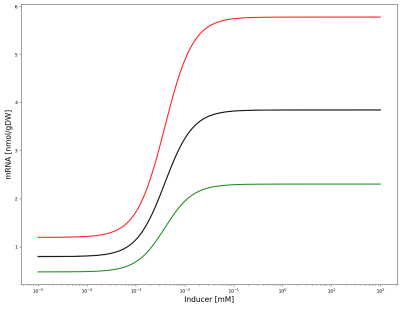
Kej = saturation transcription



big change.PNG

---

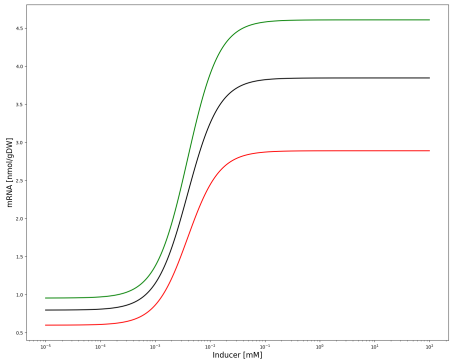
tau = time constant



tau.PNG

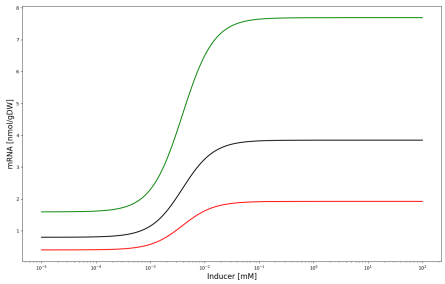
---

kej = transcription rate



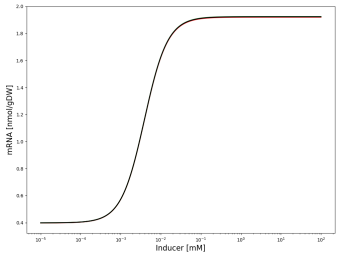
transcription rate change.PNG

RNAP total = total RNAP in the cell



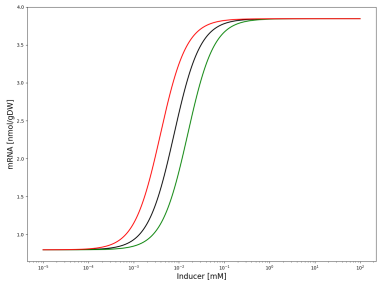
total change.PNG

Gj = copy in the cell



number change.PNG

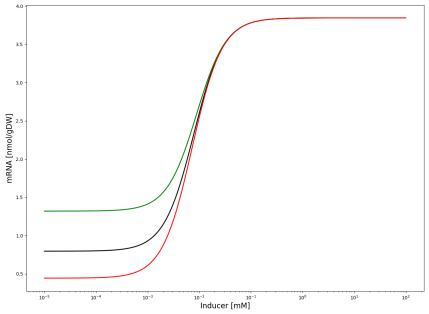
Km = affinity of the inducer



Km - binding affinity constant.PNG

---

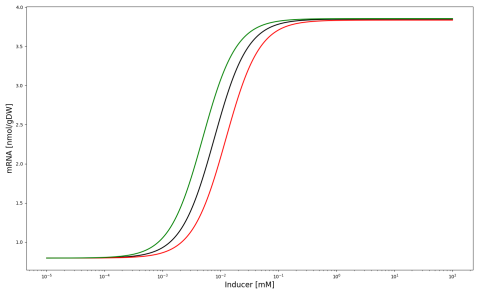
k1 = basal transcription level (leaky expression)



k1 - basal transcription rate.PNG

---

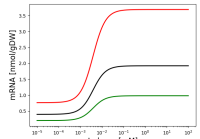
k2 = weight of the inducer effect



k2 - weight of inducer.PNG

---

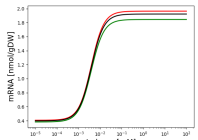
kd = mRNA degradation rate



change.PNG

---

mu = dilution rate



dilution rate change.PNG
