***Modelling Gene Expression 1***

**INVESTIGATION USING A DETERMINISTIC MODEL**

In the following exercise we will construct and simulate a deterministic model of gene expression, and explore the effect of adding negative feedback into the system.

The first step will be to construct our model of gene expression. To do this we model the rates and reactions that occur in the following processes:

|  |  |  |
| --- | --- | --- |
| Process | Description | Rate |
|  | Gene in active state switches to gene in inactive state. |  |
|  | Gene in inactive state switches to gene in active state. |  |
|  | RNAP binds to active gene and transcribes the gene to mRNA |  |
|  | Ribosome binds to mRNA and translates the mRNA to proteins |  |
|  | Degradation/dilution of mRNA |  |
|  | Degradation/dilution of protein |  |

Initially let’s assume we are using the model to represent the expression rates in a large population of identical cells each containing a single copy of the gene.

In this case the variables in the model can be described in the following way:

the fraction of cells in which the gene is in the active state

the fraction of cells in which the gene is in the inactive state

the average copy number of mRNA transcripts from the gene in each cell

P the average copy number of the translated protein P in the cell

In this model we do not explicitly model the effect of cell division, but instead include its effect on protein and mRNA numbers through modelling a degradation/dilution process.

***Construction of the model***

*1) Use the rate expressions to complete the table below:*

|  |  |
| --- | --- |
| Species | Rate equation |
|  |  |
|  |  |
|  |  |
|  |  |

***Analysis of steady states***

*2) Analyse the equations you found to identify the steady state conditions for:*

*a) the concentration of mRNA in the system [mRNA]*

*b) the concentration of protein in the system [P]*

In this model the gene either exists in a fully-active or fully-inactive state. This leads to the following conservation law:

*3a) Use this equation to rewrite the rate equation for in terms of only.*

*b) Analyse the resulting expression to find the steady state value for , equivalent to the fraction of genes in the active state.*

***Initial Conditions and Parameters***

We will use the following parameters as our initial parameters and rate constants:

|  |  |
| --- | --- |
| Model parameter | Value (seconds-1) |
|  |  |
|  |  |
|  |  |
|  |  |
|  |  |
|  |  |

|  |  |
| --- | --- |
| Species | Initial condition |
|  | 0 |
|  | 1 |
|  | 0 |
|  | 0 |

***Analysis of model timescales***

In the model we have only assumed processes can be modelled using 1st order mass action dynamics. The rate of each process is defined by its “*k*” parameter or *rate constant*.

We can use standard mathematical relations for these to extract biologically relevant information about our system from these values.

Firstly we note that for a population that is declining with rate constant *k* the half-life can be calculated as:

half-life = ln(2)/*k*

*4) Use the parameter values provided to calculate the half-lives (in minutes) associated with:*

*a) protein degradation/dilution*

*b) mRNA degradation/dilution*

*c) Comment on the timescales that you found with respect to the likely biological processes that dominate the rate in each case.*

If the gene in its active state switches to its inactive state with rate *koff* the average time interval over which it remains active can be shown to be 1/*koff*.

*5 a) Use this relationship to calculate the average time interval that the gene will spend in the active state.*

*b) A similar argument can be made for the time for the inactive gene to switch on. Use this to find an estimate for the average time interval that the gene spends in its inactive state.*

*c) Compare your answers to a) and b). Comment on how they relate to the steady state value for gon,*

***Simulating the gene expression model***

*6) Simulate the model using the template file* gene\_exp1.py

*(If you want additional practise you could alternatively write this yourself from scratch.)*

*a) Create separate plots showing the behaviour of gon, mRNA, P, and extend the simulation time until you see the whole system reach steady state.*

*Comment on the behaviour you observe, and whether it is biologically sensible.*

*b) Examine the figures produced to read off the steady states reached by the system, and compare these values to the predictions made from your analysis of the model equations.*

***Modifying the model to include negative feedback***

Suppose we want to model a situation where the protein produced acts cooperatively as a repressor for its own expression.

This can be done by introducing a Hill function term into the transcription rate expression:

|  |  |  |
| --- | --- | --- |
|  | RNAP binds to active gene and transcribes the gene to mRNA, with repression from protein P |  |

*7a) Explain how the action of this term is to model protein P as a repressor for the transcription process. e.g. explain the rate behaviour for different P levels.*

*b) Suggest a mechanism by which P may repress transcription.*

*c) Make a copy of the rate function (e.g. call it* sdot\_with\_repression*) and edit it to include the repressor term into your model.*

*Simulate the behaviour using* n=2 *and* K=4000*.*

*Comment on how has the steady-state behaviour changed and whether this is in line with your expectations*

***Measuring the reaction times of the system***

An important feature of gene expression networks is how quickly they react to a signal.

Suppose we assume that in addition to the processes modelled the system requires the presence of an activating signal molecule to induce transcription.

We will assume that when the signal is not present transcription cannot occur and *ktranscription* = 0.

To do this we will assume the following situation:

0 < t < 5000 signal off *ktranscription* = 0

5000 < t < 20000 signal on *ktranscription* = 1/20

t > 20000 signal off *ktranscription* = 0

*8a) Edit the code in the rate functions so that the value of ktranscription is dependant on the current time elapsed.*

*Hint. You can implement this by adding the following lines of code into the rate equations:*

if t<5000 or t>20000:

k\_t=0

*b) Use the original expression model (without repression term). Simulate how the gene expression system responds to the signal and examine the resulting plot of behaviour.*

The reaction time for a gene circuit measures the time taken for the system to move to its new steady-state, this is usually calculated as the time taken for the system to shift halfway to its new steady state.

*Inspect the figure to estimate the reaction time taken for the protein levels to respond to:*

*i) the signal switch on*

*ii) the signal switch off*

*b) Repeat the analysis for the model that includes the negative feedback via repression and note the system reaction time for:*

*i) the signal switch on*

*ii) the signal switch off*

*c) Compare the reaction times for the two models and discuss how/why the effect could be experimentally useful to discriminate between the two types of system.*

**Additional advanced analysis tasks (optional)**

**(Easy-ish)**

In your analysis of the system without negative feedback you identified steady state conditions for variables mRNA and protein P.

You should be able to combine these to write an expression for the steady state of P that is only in terms of parameters:

Check that this value is in line with the simulation output.

**(Harder)**

You should be able to repeat this analysis for the model equations that include the negative feedback term.

In this case the steady state condition for P now involves an additional term involving parameter *K* and [P] itself!

Check that the behaviour of the system (i.e. final steady state of [P] reached) is in line with this expression.

**(Even Harder)**

The above equation is difficult to solve analytically.

Write Python code to solve the equation iteratively.