

Stochastic Modeling

Stochasticity is a key player in regulation of gene expression especially when number of molecular species involved is small; therefore, genetic circuits such as ceRNA networks are usually embedded in more complex networks so that induced interactions might be of regulatory value.^[3] Due to potential crosstalk between miRNA targets is quite effective right in proximity to the threshold^[4], where free molecular species are present in small numbers, it is necessary a stochastic analysis of the ceRNA system.

ceRNA stochastic Modeling

Here, we are trying to represent our Model to simply describe a two-states where a miRNA bind to the circular RNA representing a miRNA sponge.

- Let λ_0 be the flow rate from miRNA to circular RNA sponge which is parameterized to ceRNA.miRNA complex association rate
- λ_1 be the flow rate from Sponge to miRNA which is parameterized to ceRNA.miRNA complex dissociation rate.
- Model was represented by a continuous time markov chain as shown in figure-1.

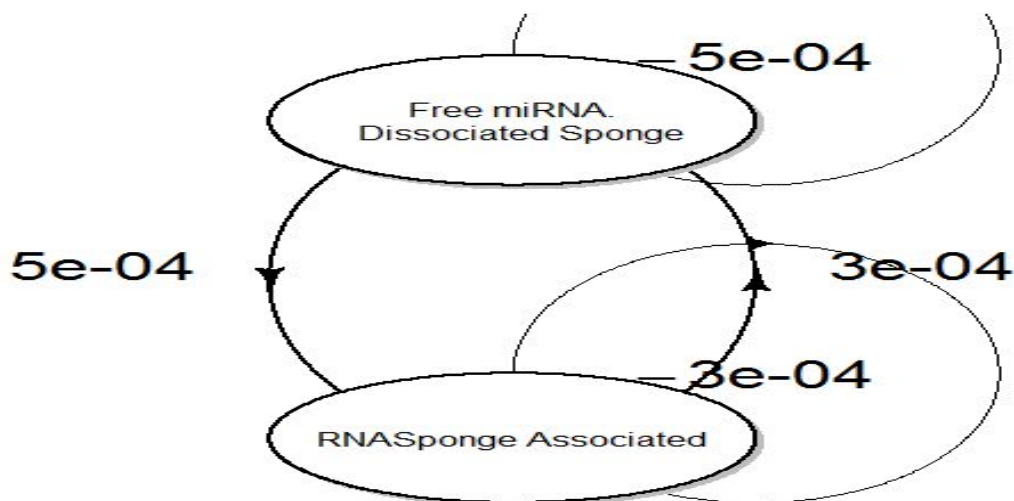


Figure-1 Continuous time markov chain CTMC representing association and dissociation of ceRNA sponge

Continuous time markov chain

A simple CTMC inspired by this lecture ^[2] is the chain with two states dissociated circRNA-miRNA and associated RNA sponge. Let λ_0 be the flow rate from dissociated circRNA-miRNA to associated RNA sponge and λ_1 be the flow rate from associated RNA sponge to dissociated circRNA-miRNA. The transition rate matrix is given by

$$Q = \begin{bmatrix} -\lambda_0 & \lambda_0 \\ \lambda_1 & -\lambda_1 \end{bmatrix}.$$

The probability vector is given by

$$\frac{df(t)}{dt} = f(t)Q$$

This gives the two balance equations

$$\frac{df_0(t)}{dt} = -\lambda_0 f_0(t) + \lambda_1 f_1(t)$$

$$\frac{df_1(t)}{dt} = -\lambda_1 f_1(t) + \lambda_0 f_0(t)$$

Now, since there are only two states, at any t Since

$$f_1(t) + f_0(t) = 1$$

Therefore the first equation becomes

$$\frac{df_0(t)}{dt} = \lambda_1 - (\lambda_0 + \lambda_1)f_0(t)$$

A solution then is suspected of the form

$$f_0(t) = K + Ce^{-(\lambda_0 + \lambda_1)t}$$

Differentiating and substituting into (1) gives

$$C(\lambda_0 + \lambda_1)e^{-(\lambda_0 + \lambda_1)t} = \lambda_1 - (\lambda_0 + \lambda_1)k - C(\lambda_0 + \lambda_1)e^{-(\lambda_0 + \lambda_1)t}$$

$$\therefore K = \frac{\lambda_1}{(\lambda_0 + \lambda_1)}$$

To determine the boundary condition then

$$\text{assume } f_0(0) = F_0$$

Substitute then Let now the final equation be

$$f_0(t) = \frac{\lambda_1}{\lambda_0 + \lambda_1} + (f_0 - \frac{\lambda_1}{\lambda_0 + \lambda_1})e^{-(\lambda_0 + \lambda_1)t}$$

The first part of this (the constant) is the equilibrium solution and the second part is a damped term which shows how the probabilities approach the equilibrium. In this case the balance equations become

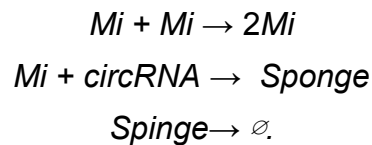
$$-\lambda_0\pi_0 + \lambda_1\pi_1 = 0$$

$$\lambda_0\pi_0 - \lambda_1\pi_1 = 0$$

Model Implementation of CTMC using Gillespie SSA Algorithm

Let's implement our previous model in a Gillespie Algorithm where the Circular RNA Sponge acts as a predator for a miRNA which is a prey.

First, let Mi represents $[miRNA]$ and the RNA Sponge represents $[Circular RNA]$ as a stochastic network as



The first "reaction" represents miRNA Transcription, the second miRNA Sponge interaction and the third represents Sponge Degradation. This was encoded in R as a stochastic Petri net (SPN) using Simulation to a realisation of the process on a regular time grid over 10 time units the interval $[0, 100]$ in steps of 0.1 time units.

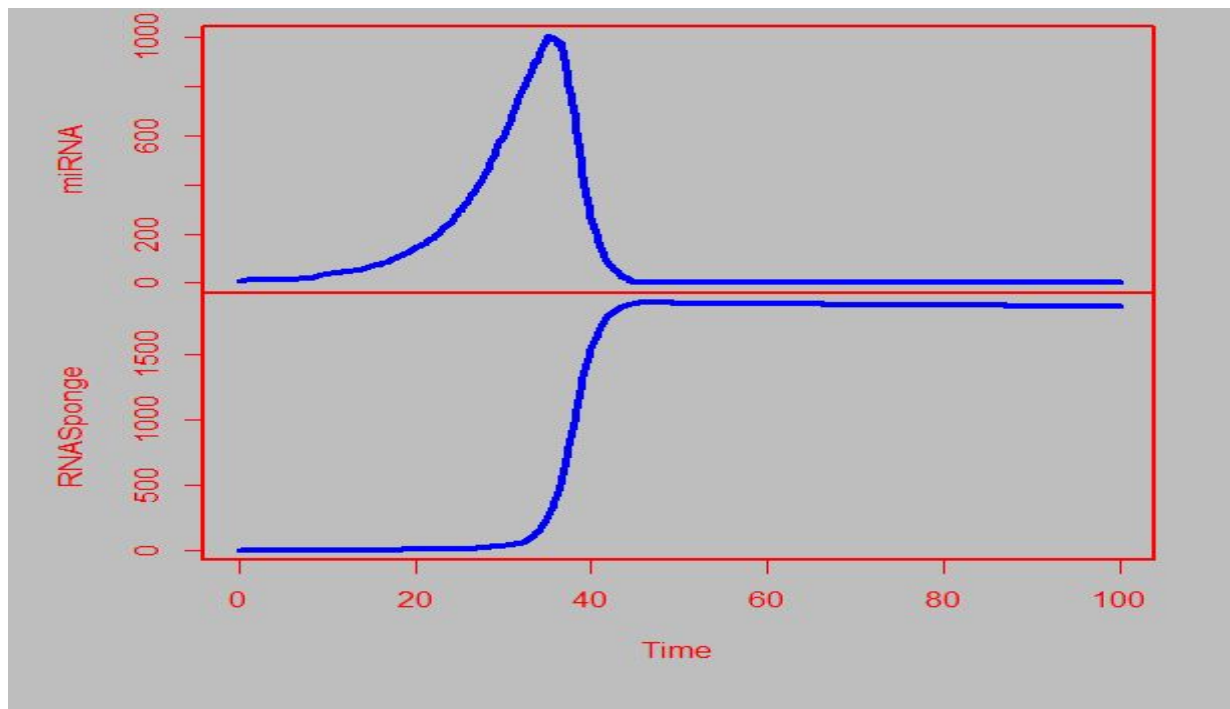


Figure-2 stochastic implementation of Gillespie to ceRNA inhibition to miRNA a general network of miRNAs. This simulation shows the circRNA sponge targeting action to free miRNAs which is down regulated along the transcription of circRNA to be acting as an

RNA sponge. This simulation run was plotted using Stochastic Modelling for Systems Biology smfsb R package^[2].

Stochastic Modeling using R

We have used the function solveStoch of sysBio R package which allowed us to simulate the model using Gillespie stochastic simulation algorithm through the "ssa" function of the GillespieSSA package.

- A vector was created for modeling parameters.
- Species , rates and parameters of each model were checked.
- The list of reactions for stochastic simulation was prepared as a propensity function list.
- Stochastic matrix was generated where each column represents a reaction while rows represents reaction species.
- A vector of propensity functions was created.
- Finally, models were solved stochastically.

Simulations were plotted (figure-2, figure-3) to visualize species evolution overtime. These plots were used to assess the accuracy of stochastic modelling using ssa function by comparing stochastic models to deterministic modeling simulations.

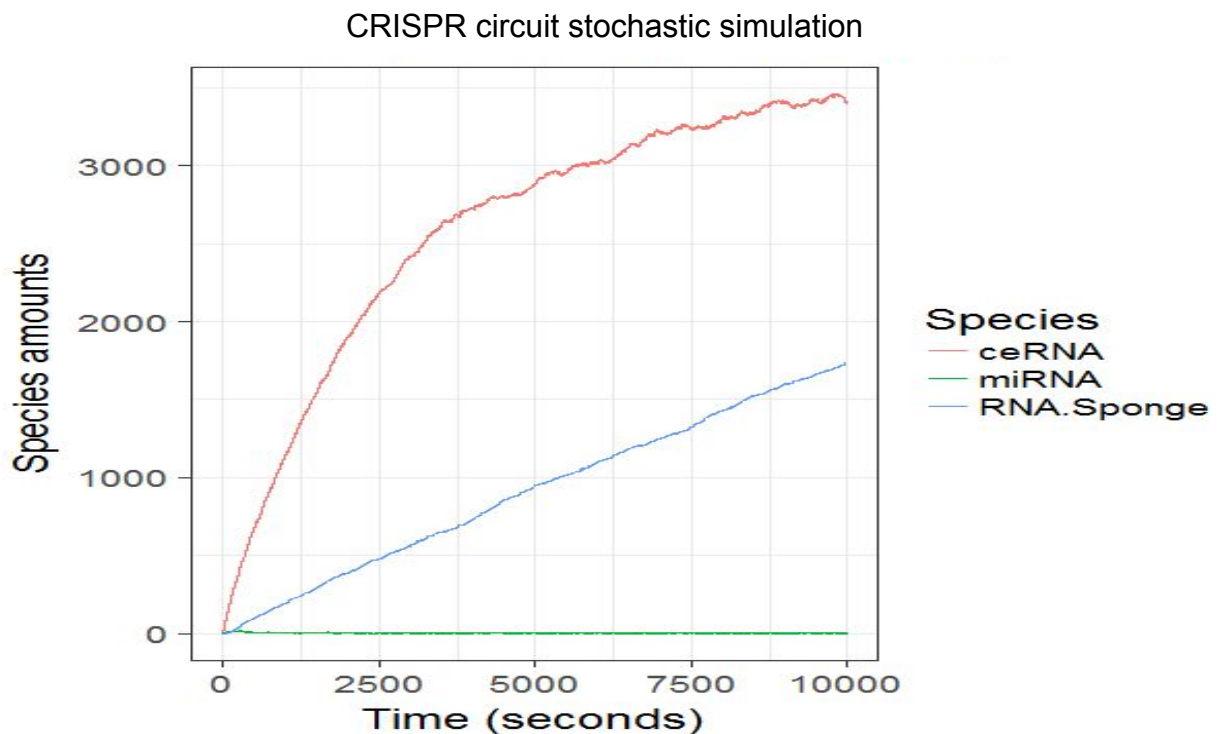


Figure-2 Stochastic Simulation run for for ceRNA network model using GillespieSSA^[6] R package. This simulation was run using sysBio^[5] R package describing stochastic effects on ceRNA network Sponge production and species levels in the network.

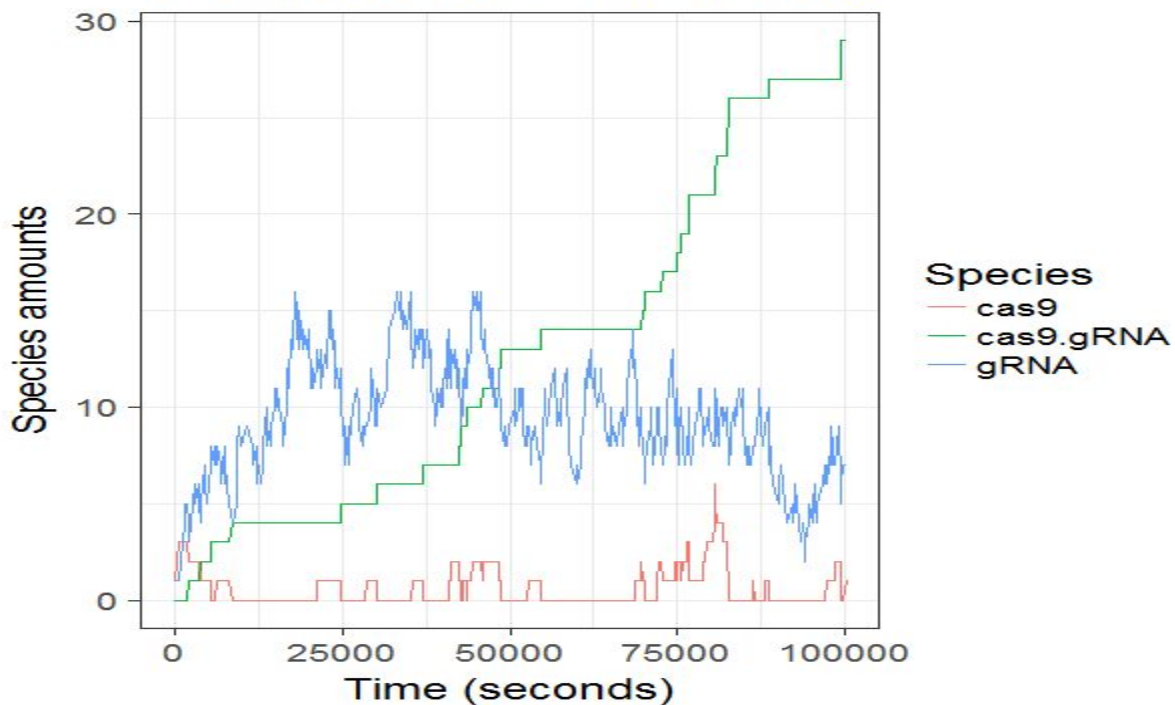


Figure-3 Stochastic Simulation run for for cas9 network model model using GillespieSSA^[6] R package. This simulation was run using sysBio^[5] R package describing stochastic effects of gRNA.cas9 association to form their intermediate complex.

References :

- 1- www.richardclegg.org/networks2/Lecture7_06.pdf
- 2- <https://cran.r-project.org/web/packages/smfsb/vignettes/smfsb.pdf>
- 3- Introduction to Stochastic Processes with Applications in the Biosciences (2013), David F. Anderson, University of Wisconsin at Madison
- 4- van Kampen N (2007) Stochastic processes in physics and chemistry. North holland.
- 5- <https://github.com/Vessy/sysBio/commits/master>
- 6- Pineda-Krch M. 2008. Implementing the stochastic simulation algorithm in R. Journal of Statistical Software 25(12): 1-18