MODELING IP_3 RECEPTOR FUNCTION USING STOCHASTIC APPROACHES

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ABSTRACT

The time evolution of chemical systems is traditionally modeled using deterministic ordinary differential equations. Chemical reactions, however, are random in nature, and the deterministic approach is valid only for a restricted class of systems. Stochastic models take random fluctuations into account and are thus more realistic. In this work, we simulate an inositol trisphosphate receptor model using ordinary differential equations, stochastic differential equations, and the Gillespie stochastic simulation algorithm. The main goal of this work is to study the applicability of these methods for a system containing small numbers of molecules and ions. We concentrate especially on the SDE approach and investigate how well it models systems with small numbers of chemical species.

1. INTRODUCTION

Biochemical reactions can be modeled stochastically using numerous different methods [1, 2]. An ideal model would have the following three important properties. First, the model should be as realistic as possible, second, the mathematical method should be easily implementable as a computer algorithm, and third, the algorithm should be computationally effective. Some realistic modeling approaches can be derived directly from chemical kinetics without making any approximations. Such approaches are called exact. A good example of an exact modeling approach is the stochastic simulation algorithm (SSA) developed by Gillespie [3, 4]. The SSA is applicable when the molecular populations in the system are small, but it becomes computationally inefficient when the numbers of molecules increase [4].

In order to construct stochastic models that can be effectively simulated, new mathematical approaches have to be explored. As an approximate method also stochastic differential equations (SDEs) have been considered a promising way to model biochemical reactions stochastically [5]. The SDE approach is attractive especially if we consider a system for which the SSA is computationally inefficient and the traditional deterministic ordinary differential equation (ODE) approach cannot be used as a good approximation.

In this study, we simulate the inositol trisphosphate receptor (IP_3R) model containing small numbers of chemical species. The SSA is evidently the most efficient modeling approach in this case. However, our goal is rather to study the typical characteristics of different approaches. This kind of knowledge is extremely valuable when the modeling approaches are applied for larger systems.

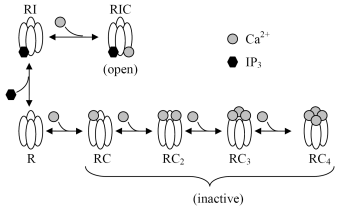


Figure 1. States and transitions of the IP_3R model.

2. SYSTEM AND METHODS

Several models have been proposed for the IP_3 receptor (for a review, see, e.g. [6]). In this study, we use the model of Doi et al. [7] which was originally published as a part of a larger model for calcium ion (Ca^{2+}) dynamics in the cerebellar Purkinje cell spine. The graphical illustration of the model is given in Figure 1. The transitions between the states are described by reversible chemical reactions of the form

$$\mathrm{A} + \mathrm{B} \underset{k_b}{\overset{k_f}{\rightleftharpoons}} \mathrm{C}, \tag{1}$$

where A, B, and C are chemical species, and k_f and k_b are *rate constants* for forward and backward reactions, respectively. The reactions of the model are given in Table 1. The rate constants of these reactions have been determined from experimental data [7]. The used volume of cytosol is $0.1\ \mu\mathrm{m}^3$. In the following, [X] denotes the concentration of species X.

The IP_3R model involves one open state (i.e. RIC). Once the IP_3R channel structure is open, Ca^{2+} flux from the endoplasmic reticulum (ER) to the cytosol starts. In this study, we model the Ca^{2+} flux using the differential equation

$$\begin{aligned}\frac{d[\mathrm{Ca}^{2+}]_{\mathrm{cyt}}}{dt} &= -\frac{d[\mathrm{Ca}^{2+}]_{\mathrm{ER}}}{dt} \\ &= k[\mathrm{RIC}]([\mathrm{Ca}^{2+}]_{\mathrm{ER}} - [\mathrm{Ca}^{2+}]_{\mathrm{cyt}}), \\ &\quad \text{when } [\mathrm{Ca}^{2+}]_{\mathrm{ER}} - [\mathrm{Ca}^{2+}]_{\mathrm{cyt}} > 0, \text{otherwise } 0,\end{aligned} \tag{2}$$

Table 1. Reversible reactions, reaction rates, and rate constants for the IP_3R model of Doi et al. [7]

	Reaction	Reaction rate	k_f	k_b
R_1	RI + $Ca^{2+} \underset{k_b^{R_1}}{\overset{k_f^{R_1}}{\rightleftharpoons}}$ RIC	$v_{R1} = k_f^{R_1}[RI][Ca^{2+}]_{cyt} - k_b^{R_1}[RIC]$	$8 \times 10^9 \frac{1}{Ms}$	$2000 \frac{1}{s}$
R_2	R + $IP_3 \underset{k_b^{R_2}}{\overset{k_f^{R_2}}{\rightleftharpoons}}$ RI	$v_{R_2} = k_f^{R_2}[R][IP_3] - k_b^{R_2}[RI]$	$10^9 \frac{1}{Ms}$	$25800 \frac{1}{s}$
R_3	R + $Ca^{2+} \underset{k_b^{R_3}}{\overset{k_f^{R_3}}{\rightleftharpoons}}$ RC	$v_{R_3} = k_f^{R_3}[R][Ca^{2+}]_{cyt} - k_b^{R_3}[RC]$	$8.889 \times 10^6 \frac{1}{Ms}$	$5 \frac{1}{s}$
R_4	RC + $Ca^{2+} \underset{k_b^{R_4}}{\overset{k_f^{R_4}}{\rightleftharpoons}}$ RC_2	$v_{R_4} = k_f^{R_4}[RC][Ca^{2+}]_{cyt} - k_b^{R_4}[RC_2]$	$2 \times 10^7 \frac{1}{Ms}$	$10 \frac{1}{s}$
R_5	RC_2 + $Ca^{2+} \underset{k_b^{R_5}}{\overset{k_f^{R_5}}{\rightleftharpoons}}$ RC_3	$v_{R_5} = k_f^{R_5}[RC_2][Ca^{2+}]_{cyt} - k_b^{R_5}[RC_3]$	$4 \times 10^7 \frac{1}{Ms}$	$15 \frac{1}{s}$
R_6	RC_3 + $Ca^{2+} \underset{k_b^{R_6}}{\overset{k_f^{R_6}}{\rightleftharpoons}}$ RC_4	$v_{R_6} = k_f^{R_6}[RC_3][Ca^{2+}]_{cyt} - k_b^{R_6}[RC_4]$	$6 \times 10^7 \frac{1}{Ms}$	$20 \frac{1}{s}$

where k is rate parameter, [RIC] is the concentration of open channels, and Ca^{2+} denotes calcium ions passing through the open channel. For k, we use the value $5.8 \times 10^8 \frac{1}{Ms}$, and the initial value for $[Ca^{2+}]_{ER}$ is 150 μM (cf. [8]).

2.1. Ordinary differential equation modeling

A set of chemical reactions can be modeled deterministically using the law of mass action and ODEs. According to the law of mass action, we can determine the *reaction rate* v of the reaction in Equation 1 by means of the equation

$$v = -\frac{d[A]}{dt} = -\frac{d[B]}{dt} = \frac{d[C]}{dt} = k_f[A][B] - k_b[C]. \quad (3)$$

If we consider a system of n species X_i, $i = 1, \ldots, n$, and m reactions R_j, $j = 1, \ldots, m$, the time evolution of the ith species is described by the equation

$$\frac{d[X_i]}{dt} = \sum_{j=1}^{m} s_{ij} v_j, \quad (4)$$

where s_{ij} is the stoichiometric coefficient and v_j is the reaction rate of the jth reaction. The stoichiometric coefficient $s_{ij} \in \mathbb{Z}$ describes how many molecules of a certain kind are involved in a certain reaction. It is positive if the amount of the molecule is increasing, negative if the amount is decreasing, and 0, if the amount is not changing in the reaction.

We now have a set of coupled ordinary differential equations that can be written in the form

$$\frac{d\mathbf{X}(t)}{dt} = \mathbf{S}\mathbf{v}(\mathbf{K}, \mathbf{X}(t)), \quad (5)$$

where $\mathbf{X}(t) : [0, \infty) \longrightarrow \mathbb{R}^n$ consists of the concentrations of the chemical species $X_i, i = 1, \ldots, n$, $\mathbf{v}(\mathbf{K}, \mathbf{X}) : \mathbb{R}^n \longrightarrow \mathbb{R}^m$ describes the reaction rates, $\mathbf{S} \in \mathbb{R}^{n \times m}$ is the stoichiometric matrix including the stoichiometric constants, and $\mathbf{K}$ is a vector including the rate constants.

2.2. Stochastic differential equation modeling

SDE modeling is based on the theory of stochastic integration. If we consider the n-dimensional deterministic ODE model introduced in Subsection 2.1, we can obtain an SDE model by incorporating an Itô integrable stochastic term in Equation 5. As a result, we have the equation

$$d\mathbf{X}(t) = \mathbf{S}\mathbf{v}(\mathbf{K}, \mathbf{X}(t))dt + \mathbf{S}\mathbf{P}\mathbf{V}(\mathbf{X}(t))d\mathbf{B}(t), \quad (6)$$

where $\mathbf{B}(t) \sim N(\mathbf{0}, t\mathbf{I})$ is the m-dimensional Brownian motion, $\mathbf{P} \in \mathbb{R}^{m \times m}$ is a diagonal matrix describing the parameters, $\mathbf{V} : \mathbb{R}^n \longrightarrow \mathbb{R}^{m \times m}$ is a diagonal matrix including reaction rates without rate constants, and $\mathbf{X}, \mathbf{S}$, and $\mathbf{v}$ are as in the ODE model described by Equation 5 [5]. If we want to incorporate randomness in each reaction rate constant separately, we just consider one reversible reaction as two separate non-reversible reactions and use the same technique as described above.

Equation 6, describing a stochastic process, can also be written in the form

$$\mathbf{X}(t) = \mathbf{X}_0 + \int_0^t \mathbf{S}\mathbf{v}ds + \int_0^t \mathbf{S}\mathbf{P}\mathbf{V}d\mathbf{B}(s), \quad (7)$$

where $\mathbf{X}_0$ is the initial state, the first integral is the Riemann integral, and the second integral is the Itô integral [9]. The expected value and the variance of this process are usually difficult to solve. Simulation studies are thus needed. Parameters included in $\mathbf{P}$ should be estimated using some estimation algorithm.

2.3. Stochastic simulation algorithm

The stochastic simulation algorithm (SSA) is a Monte Carlo procedure, which is used to generate numerically the time evolution of a chemically reacting system [3]. It treats chemical species discretely and simulates every reaction one at a time [3, 4]. In the following, the basic idea of the SSA is presented.

Let us consider the system of n species and m reactions introduced earlier in Subsections 2.1 and 2.2, and let $\mathbf{X}(t) : [0, \infty) \longrightarrow \mathbb{Z}^n$ be a vector containing the numbers of molecules of each species at time t. Each reaction R_j, $j = 1, \ldots, m$, in the system can be characterized by a *propensity function* $a_j(\mathbf{X})$ which depends on the current state of the system. A *state change vector* $\mathbf{v}_j \in \mathbb{Z}^n$ describes the stoichiometry of the reaction R_j. In the simulation algorithm, the propensity functions are used for determining the distributions of the next reaction to happen (j) and the time to the next reaction (τ). These distributions are then sampled and the state of the system is updated by state change vector. The SSA consists of the following steps:

1. Initialize the time $t = t_0$ and the state of the system $\mathbf{X}(t) = \mathbf{X}_0$.
2. Evaluate $a_j(\mathbf{X}(t)), j = 1, \ldots, m$, and $a_0(\mathbf{X}(t)) = \sum_{k=1}^{m} a_k(\mathbf{X}(t))$.
3. Generate two uniformly distributed random variables r_1 and r_2 and take $\tau = (1/a_0(\mathbf{X}(t))) \ln(1/r_1)$ and j such that $\sum_{k=1}^{j-1} a_k(\mathbf{X}(t)) < r_2 a_0(\mathbf{X}(t)) \leq \sum_{k=1}^{j} a_k(\mathbf{X}(t))$.
4. Replace $\mathbf{X}(t + \tau) = \mathbf{X}(t) + \mathbf{v}_j$ and $t = t + \tau$.
5. Return to step 2 or end the simulation.

3. RESULTS

We simulate the IP_3R model using ODEs, SDEs, and the SSA. All simulatios are run in MATLAB®. The Ca^{2+} flux described by Equation 2 is modeled simply as a part of the set of differential equations in the ODE and SDE implementations. In the SSA simulations, the flux is described as a forward reaction for which the propensity function is determined by the number of open channels and by the number of Ca^{2+} ions in the cytosol and ER.

3.1. ODE and SSA

When modeling biochemical systems, the selection of the model plays an important role. The model should describe the natural phenomenon as rigorously as possible, but ignore the details that are not essential for system level behavior. After a proper model has been selected, the next step is to choose the formalism to describe the model and find out how to implement the model as an algorithm.

Previous computational studies considering the IP_3R model show that the traditional ODE approach provides us with a satisfactory approximation only in the case in which the concentrations are relatively large (see e.g. [8]). When the numbers of chemical species are small, the relative amount of random fluctuations in the system is greater. In this case, we have to use modeling methods that are capable of taking these fluctuations into account. In the following, we concentrate on the cases in which stochastic methods are needed.

When the IP_3R model is simulated stochastically using the SSA, the results differ notably from the results of the ODE simulations (Figure 2(a)). The main reason for this is that the SSA simulation quite often leads to a closed receptor state. This means that there is no open channel for Ca^{2+} flux from the ER and thus the number of Ca^{2+} ions in the cytosol does not increase. The SSA simulations also support the intuitive assumption that the two reactions leading to the open state of the receptor are the most essential when the stochastic nature of the model is concerned.

It is clear that the SSA is the most efficient approach when it comes to computational time if the numbers of chemical species are small. However, it is also useful to study approximative methods in order to learn about their properties and behavior. It is clear that many continuous time approximations of the SSA cannot be applied. For example, the use of the chemical Langevin equation (CLE) requires certain conditions to be fulfilled [4]. First, several reactions must occur during one time step, and second, the time step should be small enough. When we take a closer look at our SSA simulations, we observe that both of these conditions cannot be satisfied at the same time.

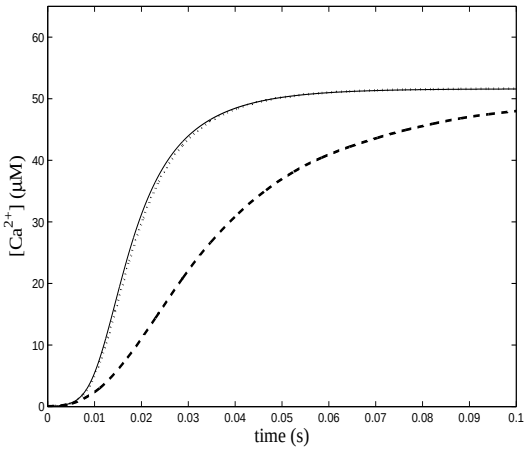
3.2. SDE

In biological systems, the concentrations of chemical species are often very small and the SDE modeling is thus challenging. The possibility of negative concentrations and the risk of an unstable model are always present. This means that although the model would be mathematically correct, it might not be biologically realistic. Therefore, the type of the SDE model, the model parameters, and the numerical method for solving the SDE have to be chosen carefully.

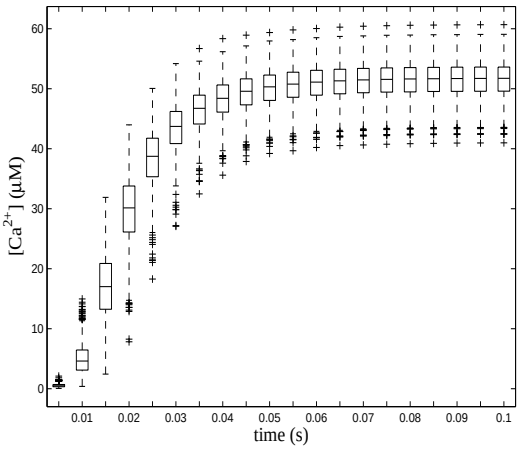
The SDE models tested in this study are built on the basis of the results obtained from the SSA simulations. As mentioned already in Subsection 3.1, the two reactions leading to the open state of the IP_3 receptor (R_1 and R_2 in Table 1) are the most significant when we study the Ca^{2+} levels in the system. When the SDE model is tuned so that randomness is incorporated only in these two reactions, the model is incabable of producing similar results as the SSA. The problem is that in order to avoid negative concentrations, we have to adjust the model parameters and the time step so that variance in the rate constants is very small. Thus, the system is always driven towards the open state and consequently the Ca^{2+} concentration in the cytosol increases. The same result is obtained if randomness is incorporated in all rate constants.

In addition to the two reactions leading to the open state, also the Ca^{2+} flux has an essential role in the model. When the whole model is constructed using the SDE, we are able to allow a greater variance of the fluctuations in the rate parameter of the flux. The drawback of this approach is that random fluctuations in the flux overpower the fluctuations in the other rate constants. This shows that the same results can be obtained using an SDE model in which randomness is incorporated only in the flux.

In order to illustrate the results, we show in Figure 2(a) the sample mean of $[Ca^{2+}]$ from thousand SSA and SDE model (randomness only in the flux) runs, and the deterministic ODE model response. In the simulations, the initial concentrations for Ca^{2+}, IP_3, and R were 0.05



(a) Sample mean of Ca^{2+} concentration.



(b) Boxplot illustration of the distribution of SDE paths.

Figure 2. (a) Sample mean of Ca^{2+} concentration in IP_3R model simulated with SDE ($\cdots$) and SSA ($--$), and deterministic response of the ODE (—). (b) Boxplot illustration of the distribution of SDE paths.

μM, 0.2 μM, and 0.2657 μM, respectively. Other initial concentrations were equal to zero. We see clearly that the SSA differs from the deterministic response, whereas the SDE model converges to it. Figure 2(b) illustrates the distribution of the solution of the SDE model. Similar analysis for the SSA reveals the great variance of the SSA paths (not shown). The deterministic response is solved numerically using the Euler method with time step 2×10^{-6} s and the SDE model is simulated using the Euler-Maruyama method with the same time step.

4. CONCLUSION

In this study, three approaches to the modeling of chemically reacting systems are introduced. The modeling approaches, namely the deterministic differential equation modeling, stochastic differential equation modeling, and the stochastic simulation algorithm, are then applied in the modeling of an IP_3 receptor model. The simulations show that when the numbers of molecules in the system are small, realistic results can be obtained only using stochastic modeling approaches. In addition, it is concluded that stochastic differential equation modeling might lead to an unstable model when the numbers of molecules are small.

5. ACKNOWLEDGMENTS

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