



An efficient reduced-lattice model of IP₃R for probing Ca²⁺ dynamics

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ABSTRACT

Numerous cellular processes are regulated by Ca²⁺ signals, and the endoplasmic reticulum (ER) membrane's inositol triphosphate receptor (IP₃R) is critical for modulating intracellular Ca²⁺ dynamics. The IP₃Rs are seen to be clustered in a variety of cell types. The combination of IP₃Rs clustering and IP₃Rs-mediated Ca²⁺-induced Ca²⁺ release results in the hierarchical organization of the Ca²⁺ signals, which challenges the numerical simulation given the multiple spatial and temporal scales that must be covered. The previous methods rather ignore the spatial feature of IP₃Rs or fail to coordinate the conflicts between the real biological relevance and the computational cost. In this work, a general and efficient reduced-lattice model is presented for the simulation of IP₃Rs-mediated multiscale Ca²⁺ dynamics. The model highlights biological details that make up the majority of the calcium events, including IP₃Rs clustering and calcium domains, and it reduces the complexity by approximating the minor details. The model's extensibility provides fresh insights into the function of IP₃Rs in producing global Ca²⁺ events and supports the research under more physiological circumstances. Our work contributes to a novel toolkit for modeling multiscale Ca²⁺ dynamics and advances knowledge of Ca²⁺ signals.

1. Introduction

Calcium ions Ca²⁺ is a universal second messenger which plays a vital role in the regulation of a large number of cellular processes [1]. Although cytosolic Ca²⁺ concentrations in cells typically remain low, stimulation can cause brief increases in [Ca²⁺]. The inositol 1,4,5-trisphosphate (IP₃) receptor (IP₃R), which is found on the surface of the endoplasmic reticulum (ER), is a vital part of this signaling pathway in numerous cell types [2,3]. In addition to releasing Ca²⁺, IP₃R channels are also controlled by the surrounding calcium concentration. This results in a Ca²⁺-induced Ca²⁺ release (CICR) mechanism in which the IP₃Rs are activated at low [Ca²⁺] but blocked at high [Ca²⁺].

IP₃Rs typically gather to form clusters of 300–800 nm in width [4,5]. Each cluster has a few tens of channels (around 3–30) [6] and is separated from the next by a distance ranging from 1 to 7 μm [7]. As a result of this spatial organization and CICR, Ca²⁺ signals display a hierarchical spatiotemporal organization. “Blips” [8] are the tiny Ca²⁺ releases that occur through a single opening IP₃R. The localized [Ca²⁺] burst raises

the possibility that an IP₃R will open nearby and causes a coordinated opening of multiple IP₃Rs in a cluster, known as “puffs” [8,9]. Both are localized signals. At the cell level, the coupling of clusters can result in global [Ca²⁺] emergence called “spikes”, which invade the whole cell and last at least a few seconds [10,11]. This type of signal generates periodically to form Ca²⁺ oscillations and propagates globally to form Ca²⁺ waves [12,13]. Studying the processes underlying such complicated dynamic behaviors has thus become a popular topic [14–17].

The investigation of complex systems has benefited greatly from the use of mathematical models. Numerical analysis has been made more difficult over time [18] due to IP₃R-mediated spatiotemporal dynamics of Ca²⁺ that span multiple scales. A description of each IP₃R's dynamics is necessary for the simulation of localized signals since it allows for nanoscale distance resolution [5,19]. Studies had previously modeled IP₃Rs calcium release and calcium signaling in a spatiotemporal manner [20]. However, it is computationally expensive to build a detailed continuous-spatial model with this refined resolution and detailed IP₃Rs simulation to describe global Ca²⁺ dynamics that last for hundreds of

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seconds at the millimeter scale [19,20]. Although simplified approaches to discretizing the modeled system or simply ignoring the spatial heterogeneity have been proposed, they cannot have both detailed descriptions of biological reality and low complexity simultaneously.

The lattice model is an alternative method to the spatial models, which use a coarser grid to simplify the Ca^{2+} diffusion [7,21,22] but are still computationally expensive and the lattice filled in the system restricts the modifiability of the model. The mean-field models rather than spatial models are more preferred due to their high time efficiency, which assumes the system to be homogeneous, i.e. the receptor channels and reactants are considered to be well-mixed within the system, and the effects of morphological or geometric volume are ignored. However, the mean-field assumption for reactants assumes that the localized $[\text{Ca}^{2+}]$ changes caused by the uptake or release of reactants from channels are mixed in the global system without delay, ignoring the spatial pattern of the heterogeneous system resulting from reactant diffusion, for example, the Ca^{2+} domains occurring after the activity of an individual or clustering IP₃Rs. These Ca^{2+} domains constitute an important part of the IP₃R's feedback in the calcium events, ignoring them will cause inaccurate modeling results [23,24]. Moreover, the mean-field assumption for channels often adjusts permeability or conductance to describe the ability of a certain kind of channel [25], but has no means to describe the spatial organization of IP₃Rs while the IP₃R clustering is inseparable from modulating the cell-level calcium signals such as oscillations and waves [26,27]. Thus, it is a challenge to develop a model which is not only detailed enough to account for multiscale Ca^{2+} dynamics and special distribution of IP₃Rs, but also simple enough to have acceptable computational efficiency for numerical simulation [6].

To resolve the inconsistencies in efficiency and accuracy in other methods, this work provides a reduced-lattice method for mimicking IP₃Rs-mediated multiscale Ca^{2+} signals from localized puff to global spike. To calculate efficient reaction kinetics, this technique maintains the independent Ca^{2+} concentration within IP₃Rs clusters and represents the localized calcium gradients using a modified calcium flux equation. To lessen the complexity of the model, the Ca^{2+} diffusion out of the domains, which has little impact on the channel's feedback, is simplified. A comparison with earlier conventional models has been done and the biological agreement of the new model has been assessed. We also gain several fresh insights under the physiological conditions that were difficult to describe by previous models.

2. Method

A two-dimensional quadrate system with periodic boundaries was built to represent a layer of cytosol above the ER in a universal non-excitable cell with the presence of IP₃Rs clusters (Fig. 1a). Most of the data and cell morphology comes from *Xenopus laevis* oocytes [6,7,22,28]. Several common Ca^{2+} fluxes through ER considered in the model are shown in Fig. 1(b), in which the (sarco)endoplasmic reticulum Ca^{2+} /ATPase (SERCA) pumps and Ca^{2+} leakage channels are regarded as homogeneous distribution in the system.

For the calcium dynamic in the system, the affected blip event durations due to the steep Ca^{2+} gradients around channels have been reported by literature [23,24]. Furthermore, the cluster scale calcium domains are also found during puff events and were determined to constitute an important part of IP₃R's feedback [23]. The reduced-lattice model retains this process by individually modeling the $[\text{Ca}^{2+}]$ of quadrate compartments that contain clusters of IP₃Rs. Meanwhile, referring to the work from Rüdiger et al. [24], the steep $[\text{Ca}^{2+}]$ gradient of the domain will only maintain during the puff event and within the cluster area, and the $[\text{Ca}^{2+}]$ evolution in the space without IP₃Rs is rather stationary [Figs. S1(a) and (b)]. This fact is simplified to a public Ca^{2+} pool [Fig. 1(d)]. The compartments do not have spatial relations with each other and can only communicate with the Ca^{2+} pool. The $[\text{Ca}^{2+}]$ in the compartments and the pool are both considered to be homogeneous.

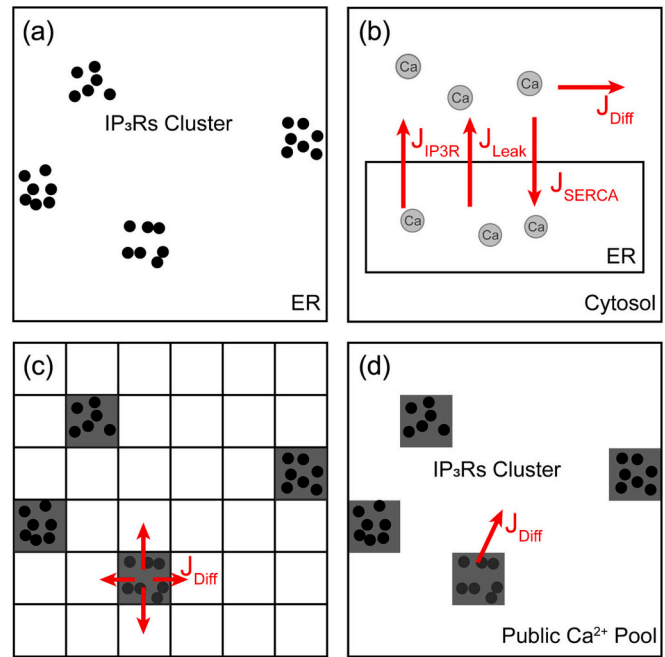


Fig. 1. Schematic drawing of the IP₃R-mediated Ca^{2+} system and model. (a) IP₃R distribution on the surface of ER. (b) Ca^{2+} fluxes involved in the model. J_{IP3R} represents the Ca^{2+} -induced Ca^{2+} release via IP₃Rs. J_{SERCA} is the SERCA pump Ca^{2+} flux and J_{Leak} is the leakage Ca^{2+} flux from ER into the cytosol. J_{Diff} describe the Ca^{2+} diffusion. (c) A sketch of the lattice model, which is a kind of spatial model, representing the IP₃R distribution in (a). Clusters correspond to the dark compartments. (d) The sketch of the reduced-lattice model representing the same IP₃R distribution. The system is separated into cluster compartments and a public Ca^{2+} pool.

The stochastic activity in the model was based on the work of Voorsluijs et al. [7]. Rather than simulating individual IP₃Rs, the model bases on a phenomenological cluster model developed by Calabrese et al. [22] and took the cluster as a unit in simulation for the efficiency of large-scale modeling and to enhance the clustering distribution of IP₃Rs. Each cluster as a whole can be in four different states: an open state *O*, in which Ca^{2+} release occurs, a closed state *C* and two inhibited states *I*₁ and *I*₂. The transition rate between each state is listed in Table 1, in which $C \rightarrow O$ and $O \rightarrow I_1$ are related to the Ca^{2+} concentration. The parameter k_{co} is between 5 and 20 $\mu\text{M}^{-1} \text{s}^{-1}$ determined by IP₃ stimulation and unless otherwise specified, it is taken as 10 $\mu\text{M}^{-1} \text{s}^{-1}$ in this work. The state transitions of each cluster are independent and described by the Markov process. A particular scheme of the Gillespie algorithm developed by Shuai and Peter [29] was used to describe this

Table 1
Parameters for the simulation.

I_{cha}	[0.1–0.5] pA	Channel Ca^{2+} current	[28]
N_{cha}	3–30	Channel number	[6]
b	98 %	Buffering	[7]
th	0.4 μm	System thickness	–
S_{cha}	0.025 μm^2	Area per channel	–
F	96,485 C mol ^{−1}	Faraday constant	–
v_p	0.8 $\mu\text{M s}^{-1}$	Maximal rate of SERCA	[7]
K_p	0.1 μM	SERCA binding constant	[7]
$[\text{Ca}^{2+}]_b$	0.04 μM	Basal $[\text{Ca}^{2+}]$	[7]
c	4	Number of directions	–
D	40 $\mu\text{M}^2 \text{s}^{-1}$	Ca^{2+} diffusion coefficient	[7]
L	[1–7] μm	Cluster separated distance	[7]
k_{co}	[5–20] $\mu\text{M}^{-1} \text{s}^{-1}$	Rate/ratio of $C \rightarrow O$	[22]
k_{oi1}	0.05 $\mu\text{M}^{-3} \text{s}^{-1}$	Rate/ratio of $O \rightarrow I_1$	[22]
k_{oi2}	40 s^{-1}	Rate/ratio of $O \rightarrow I_2$	[22]
k_{i1c}	0.005 s^{-1}	Rate/ratio of $I_1 \rightarrow C$	[22]
k_{i2c}	2 s^{-1}	Rate/ratio of $I_2 \rightarrow C$	[22]

Markov process. In detail, the states of each cluster are updated for every time step δt . If a cluster is in state A at time t and its transition rate to state B is α , then the probability that it remains A at time $t + \delta t$ is $\exp(-\alpha \bullet \delta t)$. Random numbers are drawn to determine the state transition of cluster.

The change of Ca^{2+} concentration in cluster i compartment is governed by the three Ca^{2+} fluxes through channels and a diffusion flux as

$$\frac{d[\text{Ca}^{2+}]_{clu}^i}{dt} = \text{open}^i \bullet J_{IP3R}^i - J_{SERCA}^i + J_{Leak}^i - J_{Diff}^i \quad (1)$$

where $[\text{Ca}^{2+}]_{clu}^i$ is the Ca^{2+} concentration in the cluster i compartments. J_{IP3R}^i is the IP_3R flux via the IP_3Rs cluster. The parameter open^i can only take 1 or 0, corresponding to the open state O with Ca^{2+} release and the other states without Ca^{2+} release of cluster i respectively. J_{SERCA}^i represents the SERCA pump refilling the ER by pumping Ca^{2+} back from the cytosol and J_{Leak}^i is the leakage flux from the ER into the cytosol. J_{Diff}^i is the diffusion flux from the cluster i compartments into the Ca^{2+} pool.

The IP_3R flux is at a constant rate [30], which is modified from the previous model [7] as

$$J_{IP3R}^i = \frac{I_{cha} N_{cha} (1 - b) \times 10^9 \times 50\%}{2V_{clu} F} \quad (2)$$

where the channel Ca^{2+} current I_{cha} is between 0.1 and 0.5 pA as reported [28] and is generally considered 0.1 pA in this work. N_{cha} is channel number in a cluster. The buffering effect is approximately measured by b . The factor 10^9 ensures that the flux is in μMs^{-1} . We assumed half of the channels are open in cluster activation on average and used 50 % to limit this flux. F is the Faraday constant. The volume of the cluster compartments V_{clu} is calculated as $V_{clu} = th \bullet S_{clu}$ where th is the thickness of the layer system. S_{clu} is the area of the IP_3Rs cluster, which can be calculated as $S_{clu} = S_{cha} \bullet N_{cha}$. S_{cha} is the average area per channel in the cluster derived from the area of the cluster and the number of channels within it, which is set to $0.025 \mu\text{m}^2$. Since Each cluster contains around 3–30 channels [6], the side length of a cluster compartment calculated on the basis of the above assumptions is about 274–866 nm, which is corresponding to the cluster width recorded in experiments (300–800 nm) [4,5].

The SERCA and leak fluxes are described as in the previous model [7], which assumes that ER $[\text{Ca}^{2+}]$ is always at a stable high level and ignores the influence of ER $[\text{Ca}^{2+}]$ change on the flux:

$$J_{SERCA}^i = \frac{\nu_p [\text{Ca}^{2+}]_{clu}^i}{K_p^2 + [\text{Ca}^{2+}]_{clu}^i} \quad (3)$$

$$J_{Leak}^i = \frac{\nu_p [\text{Ca}^{2+}]_b}{K_p^2 + [\text{Ca}^{2+}]_b} \quad (4)$$

where the associated parameters are given in Table 1.

To represent the diffusion flux, we modified the discrete Laplace operator and the diffusion equation between compartments given in the lattice model [7] and considered the flux as

$$J_{Diff}^i = \frac{cD([\text{Ca}^{2+}]_{clu}^i - [\text{Ca}^{2+}]_{pool})}{S_{clu}} \quad (5)$$

where c is the number of diffusion directions, which is 4 in the two-dimensional discrete system. D is the macroscopic diffusion coefficient. $[\text{Ca}^{2+}]_{pool}$ is the Ca^{2+} concentration in the public Ca^{2+} pool. The diffusion rate is directly proportional to the differential concentration between the compartment and the pool. During the puff event, $[\text{Ca}^{2+}]$ within the cluster has a sudden burst while the diffusion flux ability is limited. As a result, the calcium ions with a high concentration in the

cluster are not able to be transported out through diffusive Ca^{2+} flux in time and are maintained in the cluster compartments which eventually form the localized Ca^{2+} domain.

Evolution for $[\text{Ca}^{2+}]$ in the public Ca^{2+} pool is described by the following fluxes unrelated to IP_3Rs :

$$\frac{d[\text{Ca}^{2+}]_{pool}}{dt} = -J_{SERCA} + J_{Leak} + \sum_i \frac{S_{clu}}{S_{pool}} J_{Diff}^i \quad (6)$$

and

$$J_{SERCA} = \frac{\nu_p [\text{Ca}^{2+}]_{pool}^2}{K_p^2 + [\text{Ca}^{2+}]_{pool}^2} \quad (7)$$

where S_{pool} is the area of the pool which can be calculated as $S_{pool} = \text{Width}^2 - N_{clu} \bullet S_{clu}$, where Width is the width of the system and N_{clu} is the cluster number. Ca^{2+} diffusing out of the compartments is well-mixed in the Ca^{2+} pool according to the area ratio and causes stationary elevations of calcium concentrations in the system.

3. Result

3.1. Validation of the reduced-lattice model

To validate the biological agreement of the reduced-lattice model, we first checked the performance of the model in puff and spike conditions and compared these two kinds of calcium dynamics to experiment records. The EGTA is an exogenous slow buffer to prevent diffusion-mediated coupling between clusters and is commonly used for puff recording [11]. To mimic EGTA conditions, we referred to Voorsluijs et al. [7] and considered a single cluster located in a $5 \times 5 \mu\text{m}^2$ system. The isolated single cluster in the system thus can only exhibit puff pattern. The puff sequence shows the small amplitude bursts of the average $[\text{Ca}^{2+}]$ in the reduced-lattice system which is similar to the experimental observation [Fig. 2(a)]. The Ca^{2+} spike sequence is also recorded in the multicluster system [Fig. 2(b)]. Unless otherwise stated, the channel number in the cluster is set to 10 in this work, corresponding to a $0.5 \mu\text{m}$ cluster width. In the multi-cluster experiment, the clusters are considered to be distributed on ER by a cluster density of $\rho_c = 0.44 \mu\text{m}^{-2}$, which corresponds to a $1 \mu\text{m}$ average cluster interval and is consistent with experiment reports [7]. The MATLAB code showing the results in Fig. 2(a) and (b) is provided in the Supplemental Material. As shown in Table 2, the mean inter-puff time interval (IPI) and mean interspike time interval (ISI) obtained from the above two conditions are consistent with experiments reported [11,31]. The intensity of IP_3 stimulations varies in different experiments, we set k_{co} as $20 \mu\text{M}^{-1} \text{s}^{-1}$ to fit the experimental records.

Then, the approximately linear relation $\sigma_{ISI} = \alpha(T_{av} - T_{min})$ reported in the lattice model was tested [7,10], where σ_{ISI} is the standard deviation of the ISI, T_{av} is the average of ISI and T_{min} is the absolute refractory period. The $\sigma_{ISI} - T_{av}$ diagram shown in Fig. 2(c) suggests this kind of linear relationship between the standard deviation and the average ISI is also exhibited in the reduced-lattice model.

3.2. Comparison of the Ca^{2+} dynamics models

Since the simulation of Ca^{2+} dynamics spanning scales of spatial and temporal with detailed biologically realistic features such as IP_3R clustering and individual channel dynamics is very hard and costly [19,20], approximations are unavoidable to reduce the computational cost, including the use of lattice models and mean-field assumptions to simplify reactants diffusion and the heterogeneous system respectively. However, existing methods are unable to have both biological details and low complexity at the same time, which leads to a conflict between efficiency and accuracy. We then compared the reduced-lattice model

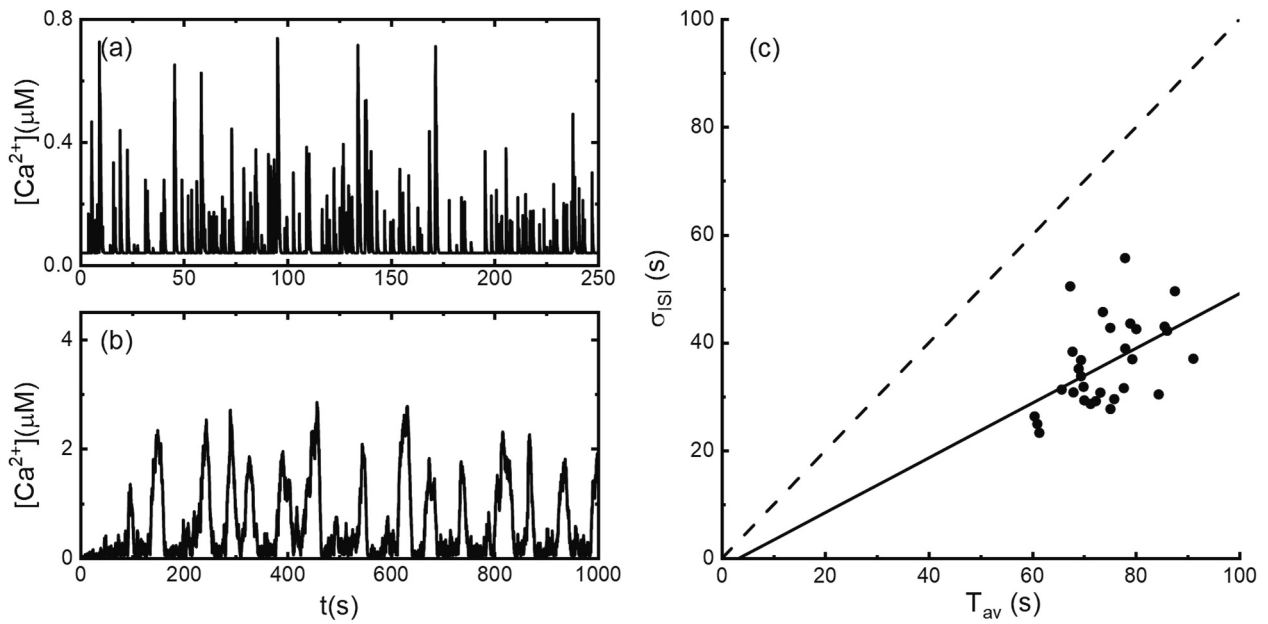


Fig. 2. (a) Puff sequence showing the local $[Ca^{2+}]$ in 1 fL around an isolated cluster in a $5 \mu m \times 5 \mu m$ system. The width of the cluster is $0.5 \mu m$, containing 10 channels. $k_{co} = 20 \mu M^{-1} s^{-1}$ (b) Ca^{2+} oscillation generated by a $10 \mu m \times 10 \mu m$ system, $\rho_c = 0.44 \mu m^{-2}$, $k_{co} = 20 \mu M^{-1} s^{-1}$. (c) $\sigma_{ISI} - T_{av}$ plot obtained by numerical simulations in a $10 \mu m \times 10 \mu m$ system, $\rho_c = 0.44 \mu m^{-2}$, $k_{co} = 10 \mu M^{-1} s^{-1}$. Each dot corresponds to a spike sequence of 20 spikes. The solid line is the linear fit through the dots and the dashed line corresponds to $\alpha = 1$. For the solid line, the minimal $T_{av} = 3.31$ s.

Table 2

Comparison of the results from the reduced-lattice model and real experiments.

	Reduced-lattice model	Experiments	Ref.
IPI (s)	1.57 ± 1.40	1.50 ± 1.37	[31]
ISI (s)	61.2 ± 22.3	10–100	[11]

k_{co} takes $20 \mu M^{-1} s^{-1}$.

with existing models and emphasized its advantages in reconciling efficiency and accuracy.

We intentionally built a lattice model and a mean-field model as representative of the spatial model and point model respectively in the Supplemental Material and simulated the same biological system with these three models, which is a $10 \mu m \times 10 \mu m$ system with $\rho_c = 0.44 \mu m^{-2}$. The lattice model needs 5000 s to simulate 100 s real time while the reduced-lattice model needs <1 s and the mean-field model needs only about 0.1 s. All simulations were performed with an Intel i7-9700 processor with 3.00-GHz processing speed and 32.0-GB RAM. In this case, the reduced-lattice model is >5000 times faster than the lattice model, and it has a friendly efficiency close to the mean-field model.

Then we compared the performance of these three models in simulating Ca^{2+} dynamics. Take the lattice model as the standard because of its fine spatial resolution in simulating the Ca^{2+} diffusion and the Ca^{2+} gradients around the clusters [see Fig. S1 in Supplemental Material], which is important in guaranteeing the accuracy of the simulation results [23,24]. As shown in Fig. 3(a), the ISI distributions of the three models are relatively close, while the mean ISI in the mean-field model is slightly smaller. The distributions of the peak values of Ca^{2+} spikes shown in Fig. 3(b) suggest the generally over-large signal intensity in the mean-field model. We then borrowed the Kolmogorov-Smirnov test to evaluate the difference between these data distributions quantitatively. Between the mean-field model and the lattice model, the ISI distributions show no significant difference ($p = 0.19$) while there is a significant difference in the distributions of the peak values, $p < 0.001$. When it comes to the comparison of the reduced-lattice model and the lattice

model, there are no significant differences in both the distribution of ISIs and peak values ($p = 0.79$ and $p = 0.34$ respectively) and the larger p -values here indicate the less difference between the results from these two models. The results show that the reduced-lattice model has a good accuracy close to the lattice model. Since there is no relative position relationship between each IP₃R cluster in the reduced-lattice system, we in addition checked the Ca^{2+} spike sequence in the lattice model with different cluster distributions in the Supplemental Material. As shown in Fig. S2, a few significant differences between the distributions of Ca^{2+} signals features generated by different cluster distributions are evident.

3.3. Extensibility of the reduced-lattice model

The IP₃R clustering is regulated by many mechanisms and is important in the modulation of global Ca^{2+} dynamics [26,27]. The mean-field description for the IP₃R channels, although widely used for its high time efficiency, it lacks accuracy because of holding a hypothesis of IP₃R homogeneous distribution rather than clustering and rescaling the ability of IP₃R Ca^{2+} flux by the scale factor. Meanwhile, although various spatial models can describe the special distribution of IP₃R, their complexity is unfriendly for the simulation at a larger scale, so they are commonly used to study localized signals. The lattice model is a useful alternative but still costly in global simulations and the channel distributions are restricted by grid structure. Thus, the previous models are deficient in the extensibility to adapt to more physiological conditions, especially short of considering the channel distribution in global scale simulations. The reduced-lattice model, however, provides a potential solution for modeling multiscale Ca^{2+} dynamics with detailed IP₃R spatial features. We then used this model to give some repeatable predictions about the role of IP₃R on global Ca^{2+} dynamics.

Our initial focus was on determining how the clustering level of IP₃R influences the generation of calcium events. We investigated the effect of channel clustering on calcium signaling while maintaining a constant overall IP₃R expression. As shown in Fig. 4, we examined the average ISI with varying numbers of channels in the cluster in a $10 \mu m \times 10 \mu m$ system. The total number of IP₃R channels in the system was kept to be 480, which is close to the expression in a typical $10 \mu m \times 10 \mu m$ system with a density of $\rho_c = 0.44 \mu m^{-2}$ (10 channels). We found that

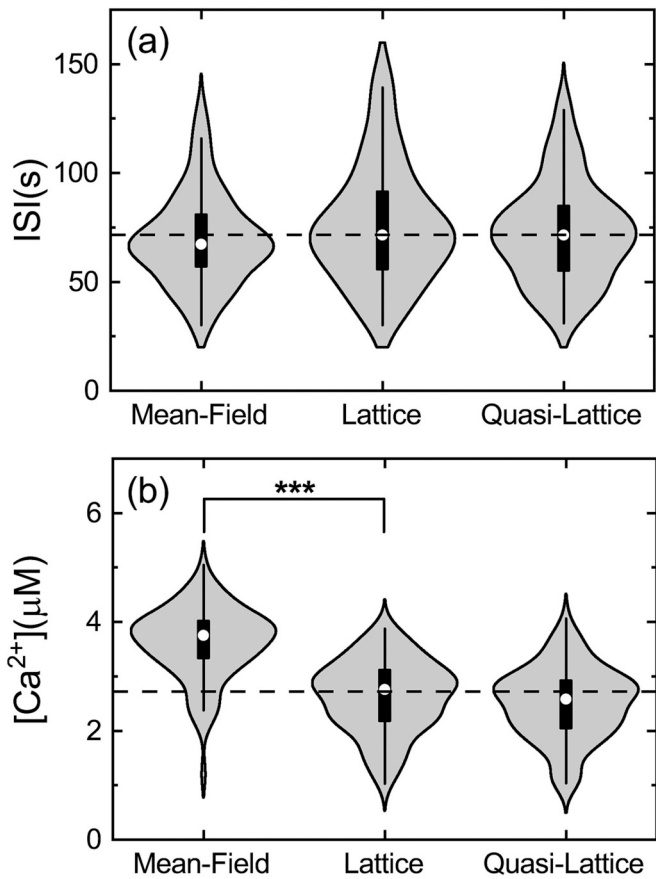


Fig. 3. (a) Violin and box plots showing the ISI distributions of Ca^{2+} spikes generated by the model with the mean-field assumption for reactant, lattice model and reduced-lattice model. (b) the peak value distributions of Ca^{2+} spikes. (a), (b) 100 data points are involved in each distribution. White points: mean values of distributions; edges of boxes: 25th and 75th percentiles; whiskers: most extreme data points not considered outliers (beyond 1.5 times the interquartile range); dashed lines: mean values from the results of the lattice model. The whole system size is $10 \mu m \times 10 \mu m$. The cluster size is $0.5 \mu m \times 0.5 \mu m$ with 10 channels. $\rho_c = 0.44 \mu m^{-2}$. The width of the quadrature compartments in the lattice model is $0.1 \mu m$ which provides a sufficient spatial resolution to simulate the Ca^{2+} diffusion accurately [see Fig. S1(c) in Supplemental Material].

the mean ISI was lowest for clusters containing approximately 12 channels, which corresponds to the range reported in the literature for clusters containing 3–30 channels [6]. Few or more channels will both lead to an increase in the spike generation interval.

In addition, many models typically regulate the Ca^{2+} flux of IP_3Rs through gating, but rarely consider the effect of IP_3R expression. Some stochastic models describe the number of IP_3Rs in the system, but only change the stochasticity [29]. The flux is adjusted by the opening ratio, but the total capacity of this pathway remains unchanged. A similar situation arises in research on the disruption of calcium homeostasis in Alzheimer's disease (AD). The effect of AD is indirectly described by altering the IP_3 level when the IP_3R flux is involved [32]. However, experiments found AD can double the expression of IP_3Rs in astrocytes, which is a kind of non-excitable cell with IP_3Rs on ER, within some particular brain regions [33]. Few studies have investigated the influence of this effect on Ca^{2+} dynamics. As shown in Fig. 5(a), we increased the expression of IP_3R by altering the number of channels in the cluster or the number of clusters in the system and examined the average ISI under different conditions. The results showed that both pathways started to produce Ca^{2+} spikes when the expression reached a certain level, and the ISI continued to shorten with the expression increase. The

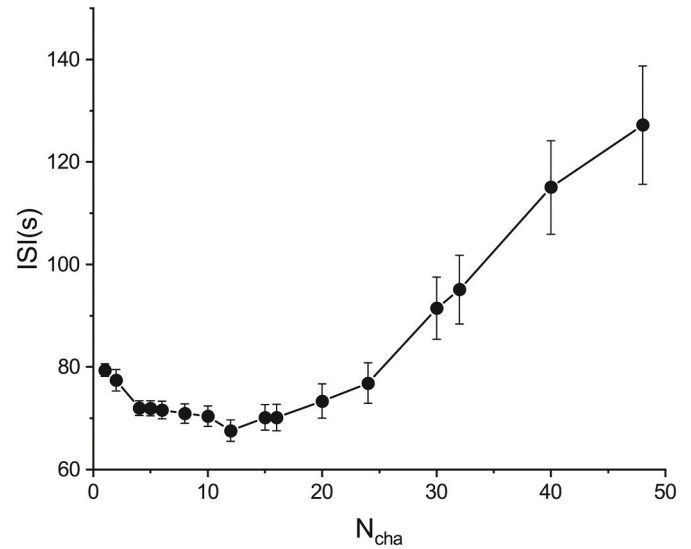


Fig. 4. Mean ISIs and 95 % confidence intervals of Ca^{2+} spikes under different channel numbers within a cluster. The total IP_3Rs expression is fixed by keeping the total 480 of IP_3Rs in the system. The whole system size is $10 \mu m \times 10 \mu m$. >100 data are involved in each data point.

black line shows the condition with 320 IP_3Rs in the $10 \mu m \times 10 \mu m$ system to form clusters in a density of more than $\rho_c = 0.44 \mu m^{-2}$, which can be confirmed with regular conditions [4–7,34]. The red line presents the conditions with a doubled expression of IP_3Rs in the system. The result is consistent with the previous conclusion that the frequency of calcium oscillations in astrocytes will significantly increase in the AD environment [32]. In Fig. 5(b), we further check the effect of IP_3 in the system. The open receptors proportion in the cluster and transition rate k_{co} both increase with $[IP_3]$, while they both result in shortened ISI, which suggests the promotion of IP_3 in calcium spike, corresponding with previous reports [1].

Furthermore, previous literature has demonstrated that increasing the number of stochastic units, including IP_3R channels and clusters, in a fixed system leads to simulation outcomes that approach the deterministic model [7,29]. Given that the reduced-lattice model enables easy regulation of the size parameters of the simulated system, we validated the influence of increasing IP_3Rs by changing the system size while maintaining the same IP_3R distribution feature. For the Ca^{2+} spike sequences in $5 \mu m \times 5 \mu m$ and $15 \mu m \times 15 \mu m$ systems shown in Fig. 6(a) and (b) respectively, the larger system produces more regular and periodic Ca^{2+} spikes. The quantitative descriptions of this phenomenon are illustrated in Fig. 6(c–e). The dispersion of the peak values and ISIs both decrease with larger system sizes, while their means exhibit little change. The results suggest that a larger system with more stochastic IP_3R units in the same physiological conditions can enhance the determinacy of the signals while maintaining their mean features.

4. Discussion

Numerous studies have been conducted to simulate the spatially and temporally complex patterns of IP_3R -mediated cytosolic Ca^{2+} signals. However, balancing the consideration of detailed biological relevance and computational cost has hindered numerical analysis. Selectively simulating biological details based on their importance in the targeted physiological process may be a rational solution and could help to improve our understanding of these complex Ca^{2+} dynamics.

The liberation of Ca^{2+} sequestered in the ER through IP_3R is a major mechanism of calcium signaling in various cell types [2,3]. The clustering distribution of IP_3Rs and their feedback processes in CICR are the most important biological factors contributing to the generation of

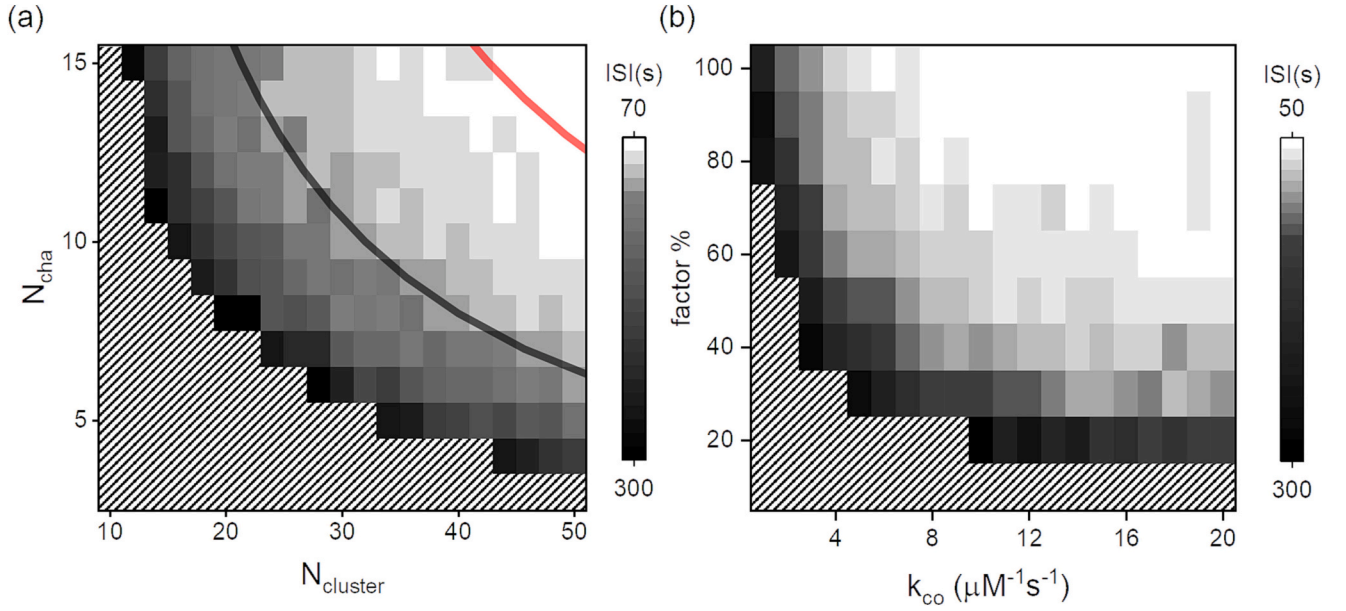


Fig. 5. (a) Mean ISIs of Ca^{2+} spikes under different channel numbers within a cluster and different cluster numbers. The black and red lines represent the total 320 and 640 IP_3R expressions in the whole system respectively. (b) Mean ISIs of Ca^{2+} spikes under different open receptors proportion in the cluster and transition rate k_{co} . $\rho_c = 0.44 \mu\text{m}^{-2}$. The physiological conditions in the shaded area cannot generate Ca^{2+} spikes or the intervals of the spikes are too large (> 300 s). Ca^{2+} signals have a higher frequency in the brighter blocks. The whole system size is $10 \mu\text{m} \times 10 \mu\text{m}$.

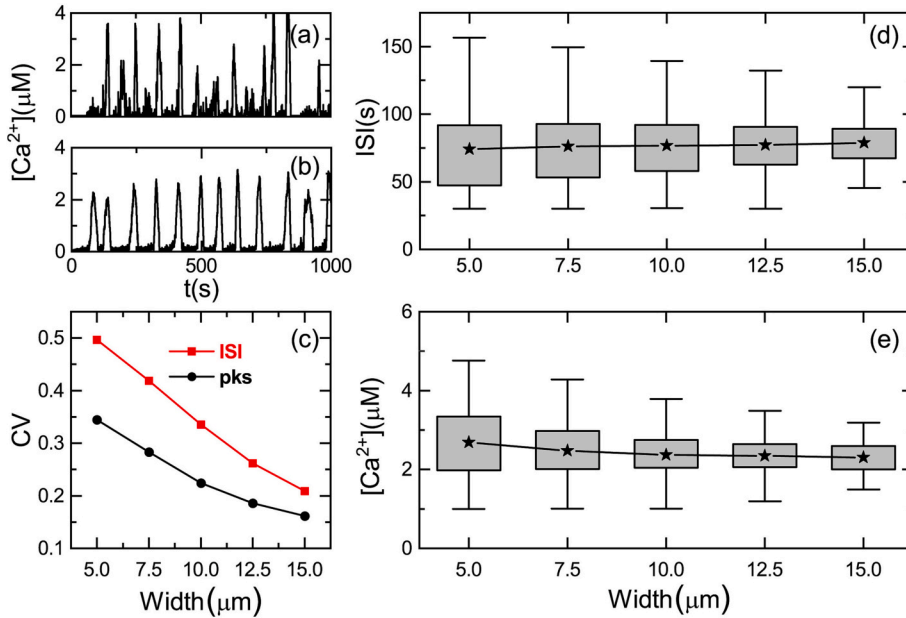


Fig. 6. (a) The Ca^{2+} spike sequence in a $5 \mu\text{m} \times 5 \mu\text{m}$ system. (b) The Ca^{2+} spike sequence in a $15 \mu\text{m} \times 15 \mu\text{m}$ system. (c) The coefficient of variation (CV) of the ISI distributions and the peak value (pks) distributions in systems with different widths. > 500 data points are involved in each distribution. (d-e) The box plot of the ISI and the peak value distributions in (c). Star points: mean values of each distribution; edges of boxes: 25th and 75th percentiles; whiskers: most extreme data points not considered outliers (beyond 1.5 times the interquartile range); (a-e) Cluster size is $0.5 \mu\text{m} \times 0.5 \mu\text{m}$ with 10 channel. $\rho_c = 0.44 \mu\text{m}^{-2}$.

multiscale Ca^{2+} signals. Due to its ease and efficiency in modeling, the mean-field assumption for reactants and channels is widely used in Ca^{2+} dynamic simulations. The mean-field assumption adjusts the channel ability by the total permeability or conductance of a certain type of channel [25], but it cannot describe their spatial organization while the clustering of IP_3R is inseparable from modulating the global Ca^{2+} dynamics [26,27]. In this study, we proposed a reduced-lattice model with an independent simulation of each IP_3R cluster, which allows the reproduction of the independent puff event within a single cluster and the coupling phenomenon of clusters in larger spike events. Meanwhile, the reduced-lattice model can simulate puff and spike events with experimental consistent IPI and ISI and report similar linear relations between CV and ISI with the lattice model.

Moreover, studies show that the mean-field assumption for Ca^{2+} does not hold because of the existence of calcium domains in different scales which causes an inaccurate description of the IP_3R 's feedback process, and eventually leads to inaccuracy of the Ca^{2+} signals simulation [23,24]. The localized gradient during the puff event keeps a high Ca^{2+} level within the IP_3R s cluster [Figs. S1(a) and S1(b)], which promotes the CICR process and results in shortening the transition time from the open state to inhibited state and increasing the possibility of transitioning to I_1 state with a longer recovery time which promotes the inhibitory effect of high $[\text{Ca}^{2+}]$ in the IP_3R s feedback mechanism. However, the absence of this phenomenon in the mean-field model leads to a harder transition between the open state and I_1 state and generally causes more Ca^{2+} release and less recovery time, which is a possible

explanation for the high signal intensity in the mean-field model [Fig. 3 (b)]. In this study, the reduced-lattice model uses an approximate description of the Ca^{2+} diffusion between the cluster and Ca^{2+} pool shown in Eq. (5) to reach a similar simulation result to the spatial models, which avoids the inaccuracy caused by the mean-field assumption for reactants. The consideration of important biological details by the reduced-lattice model supports its good biological agreement.

However, redundant details in modeling will have expensive computational costs but rarely increase the accuracy. In the simulation of cell-level signals, the spatial model with the detailed resolution is computationally expensive. The lattice model is a less computationally intensive alternative [7,21], but it is still costly in global signals simulation and the grid structure in the system restricts the modifiability of the model. However, the feedback events are confined to the clusters and no biological process is regulated by Ca^{2+} concentration in the spaces without IP_3Rs . Thus, the detailed Ca^{2+} diffusion in those spaces is less important and causes unnecessary computational consumption in simulation. The reduced-lattice model simplifies the diffusion through the concept of the public Ca^{2+} pool. In the spatial model, the evolution for $[\text{Ca}^{2+}]$ in the spaces without IP_3Rs is completely calculated while those $[\text{Ca}^{2+}]$ change in the reduced-lattice model is only calculated once in the Ca^{2+} pool, which significantly reduces the model complexity. Since the reduced-lattice model cannot consider the relative positions of the clusters compared to the traditional spatial model, we confirmed that different clusters distribution has little effect on the calcium dynamic [see Fig. S2 in Supplemental Material]. This suggests the rationality of considering the reduced-lattice system, in which the clusters have no concept of the locations but only the interaction with the public Ca^{2+} pool. Furthermore, the similarity of the results between the reduced-lattice model and the lattice model indicates the little effect of the public Ca^{2+} pool simplification on model accuracy [Fig. 3].

The real biological relevance and extensibility of the reduced-lattice model can support the study of more physiological conditions. We especially focused on the influences of the special IP_3Rs distribution on global Ca^{2+} dynamics, which is missing in previous modeling studies. We first investigated the effect of IP_3Rs clustering. One interesting finding is that when the expression of IP_3Rs remains unchanged, the aggregation of about 12 channels in a cluster can minimize the spikes interval, which corresponds to the number of channels clustered in the biological reality [6], indicating that the clustering of IP_3Rs in a certain range can better mediate the generation of Ca^{2+} spikes. This finding may help reveal the significance of the clustering distribution of IP_3Rs . We then investigated the relationship between IP_3Rs expression and the Ca^{2+} dynamics. The important finding is that the increase in IP_3Rs expression, whether reflected in the number of clusters or the channel number within a cluster, will lead to an abnormal increase in the production frequency of calcium spikes. Since the AD environment can promote both IP_3 stimulation and IP_3Rs expression, this result appears to reveal a new pathway, through which AD affects intracellular calcium homeostasis. In addition, previous works showed the deterministic feature of the modeling results and attributed this phenomenon to the increased number of stochastic units like IP_3R channels and clusters in a fixed system [7,29]. We also found that the increase of IP_3Rs with the same spatial distribution as the increase in system size can also lead to signals with more deterministic features while maintaining their average value of amplitude and frequency. These findings represent an interesting and counterintuitive phenomenon in IP_3Rs -mediated calcium signaling, where more stochastic units counterintuitively produce more deterministic results.

It is challenging to model IP_3Rs -mediated hierarchical Ca^{2+} signals. We here focused on the biological relevance and try to provide a new useful modeling framework but still have many restrictions. First, blip events are a kind of inseparable Ca^{2+} localized events that constitute the multiscale Ca^{2+} signals. However, the cluster-based reduced-lattice model is unable to consider this scale of Ca^{2+} event in simulation so fails

to integrate all scales signaling. Second, by approximation, our model becomes a spatially zero-dimensional model, in which the spatial position and heterogeneity in ion concentration are also ignored in the public Ca^{2+} pool. It is thus unable to simulate the communication between individual clusters and the propagation of Ca^{2+} waves independently. Moreover, the dynamics of a cluster in this model were developed based on puff information [22], which might affect the simulation when the cluster only contains a few numbers of channels.

In this study, we aim to develop a simplified model retaining only the most basic parts with preferable computational cost but inevitably losing some details. Some studies revealed that most IP_3R clusters are mobile, however, Ca^{2+} signals are generated by a small population of immobile IP_3Rs [35]. These small number of immobile IP_3R clusters reside alongside ER and plasma membrane junctions where they initiate Ca^{2+} signals by responding to IP_3 delivered from PLC and regulate SOCE [35]. STM1 , STM2 , and TRPC channels have long been recognized to serve as polymodal sensors in Ca^{2+} handling within such ER-PM junctions [35–37]. These components are pivotal in the generation of spatiotemporal Ca^{2+} signaling patterns. Therefore, we can improve the model by adding these pathways in the follow-up work.

One of the hotspots in neuroscience is modeling the neuronal processes underlying many kinds of memory. The astrocyte, which is a kind of non-excitatory cell, exhibits calcium transients and participates in the formation of the neuron-astrocyte network that is adapted to accommodate memory processes. Recently, it was shown that multiscale astrocyte calcium events can significantly improve the efficiency of the neuron-astrocyte network as an AI system [38]. The proposed reduced-lattice model here can help to model these multiscale events and is able to model AI features as local integration of information processing that is important for the functioning of the neuron-astrocyte network as an artificial intelligence system, which will be our future work [39].

5. Conclusion

In summary, we developed a novel modeling framework for simulating IP_3R -mediated hierarchical signals that tends to incorporate existing approaches by carefully taking into account actual biological relevance. With efficiency and precision, the proposed reduced-lattice model can mimic Ca^{2+} dynamics from the puff size to the global level. With its strong extensibility, this model can also make up for the shortcomings of earlier models and offer several novel and intriguing insights on the function of IP_3Rs in producing global Ca^{2+} events. This model can also serve as a foundation for studies on Ca^{2+} dynamics across a wider range. This approach can be applied to a more thorough analysis of complicated AI scenarios in our future work. We expect that by bringing these scale-spanning studies together, this modeling approach will make it easier to do future research on multiscale Ca^{2+} dynamics.

CRedit authorship contribution statement

H.G. S.C. and A.Z. designed the research; H.G., L.L. and S.C. and performed experiments; H.G. and L.L. contributed computational model; H.G., L.L. and S.C. analyzed data; H.G., L.L., A.Z. and S.C. wrote the manuscript; S.C. supervised the research. All authors approved the submission.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.bbamem.2023.184195>.

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