Finite Element Model to Study Two Dimensional Unsteady State Cytosolic Calcium Diffusion in Presence of Excess Buffers

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Abstract—Calcium, a vital second messenger for signal transduction in neurons, plays an important role in almost every organ of our human body. Thus modeling of Calcium signaling mechanism can help us understand this mechanism in a better way. Here, a finite element mathematical model has been developed to study the flow of calcium in two dimensions with time. This model assumes EBA (Excess Buffering Approximation), incorporating all the important parameters like time, association rate, influx, buffer concentration, diffusion coefficient etc. Finite element method is used to obtain calcium concentration in two dimensions and numerical integration is used to compute the effect of time over 2-D Calcium profile. Comparative study of calcium signaling in two dimensions with time is done with important physiological parameters, like buffer concentration, buffer association rate. A program has been developed for the entire problem and simulated on an AMD-Turion 64X2 machine to compute the numerical results.

Index Terms— FEM, EBA, MATLAB, Ca$^{2+}$ influx, Ca$^{2+}$ profile.

I. INTRODUCTION

Neurons communicate to each other through two types of junctions or synapses, namely, i) Electrical synapse and ii) Chemical synapse. The communication through electrical synapse is fast while the communication through chemical synapse is slow. However, chemical synapses are considered to be more significant than electrical synapses as they come into play when the distance between the neurons is more than 4–5 nm [1]. When the gap in between neurons is more, i.e., of the order of 20–50 nm, then signaling in between neurons cannot take place through electrical synapses. In such cases, electrical signal is converted into a chemical signal so that the message can be transmitted through a chemical synapse. This process of conversion of an electrical signal into a chemical signal is known as the process of signal transduction. Calcium acts as a switch in this process of signal transduction. It is also present near the VDCC [5]. Near to these VDCC’s there are neurotransmitters filled synaptic vesicles to which cytosolic Ca$^{2+}$ gets bound to initiate the process of exocytosis [6]. Since, exocytosis occurs in the immediate vicinity of VDCC’s therefore, [Ca$^{2+}$], transients cannot be measured in situ due to the spatiotemporal limitations of the [Ca$^{2+}$], measuring technologies [7]. Mathematical and Computational simulation of Ca$^{2+}$ kinetics provides a beautiful alternative to study the effect of several parameters over [Ca$^{2+}$], transients [8], [5], [9].

In this article, Ca$^{2+}$ dynamics are studied by developing a Finite Element Model for two-dimensional unsteady state Ca$^{2+}$ diffusion under EBA. A computer program has been developed in MATLAB for the whole approach and simulated on an AMD Turion 64X2 machine with 1.6 GHz processing speed and 2.5 GB memory. The numerical results are used to demonstrate the two-dimensional Ca$^{2+}$ profile in x and y directions. Also, numerical results are used to study the interrelationship between [Ca$^{2+}$], and other parameters viz., buffer specie, buffer concentration, association rate etc.

II. MATHEMATICAL FORMULATION

Calcium kinetics in neurons is governed by a set of reaction-diffusion equations which can be framed assuming the following bimolecular reaction between Ca$^{2+}$ and buffer species:

\[
[Ca^{2+}] + [B_j] \xrightarrow{k^+} [CaB_j]
\]

where \([B_j]\) and \([CaB_j]\) are free and bound buffer respectively, and \(j\) is an index over buffer species. It is conventional to assume isotropy, homogeneity and Fickian diffusion. With these assumptions, Ca$^{2+}$ dynamics can be represented with the help of the following system of partial differential equations [5], [8], [9]:

\[
\frac{\partial [Ca^{2+}]}{\partial t} = D_{Ca}\left(\frac{\partial^2 [Ca^{2+}]}{\partial x^2} + \frac{\partial^2 [Ca^{2+}]}{\partial y^2}\right) + \sum_j R_j
\]

(2)

\[
+ \sigma \delta (r)
\]

where, \(R_j = -k^+_j [Ca^{2+}] + k^-_j [CaB_j]\)

(5)

\[
D_{Ca}, D_{Bj}, D_{CaBj} \text{ are diffusion coefficient of free calcium, free buffer, and bound buffer, respectively; } k^+_j \text{ and } k^-_j \text{ are}
\]

(Advance online publication: 19 August 2010)
is the standard Dirac delta function.

\[ u(x,y) = \begin{cases} 1 & \text{for } x = 0, y = 0, \\ 0 & \text{otherwise} \end{cases} \]

The discretized variational form of (6 – 8) can be written as:

\[ I^{(e)} = \frac{1}{2} \int_A \left[ \phi^{(e)}_i \phi^{(e)}_j + u^{(e)}_i u^{(e)}_j + \frac{2 u^{(e)}_i \phi^{(e)}_j + 2 u^{(e)}_j \phi^{(e)}_i}{\lambda^2} \right] dA + \int_A \left[ \frac{2 u^{(e)}_i \phi^{(e)}_j D_{Ca}}{\lambda^2} \frac{\partial}{\partial t} u^{(e)} dA - \mu^{(e)} \int_A \frac{\sigma}{\lambda^2} u^{(e)} dA \right] \]

Fig. 1 Finite Element discretization of the cytosol, small black circle at element number ‘30’ and ‘33’ represents point source of calcium.

Further, the cytosol is divided into 60 linear triangular elements of different sizes (see Fig. 1).

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where, \[ B \] and \[ C \] are element nodal \( \sigma \delta \) and \( \sigma \) respectively. Assembling (13) for \( e = 30, 33 \) and (14) for rest of the elements.

Here, we have used ‘\( u \)’ in lieu of \( [\text{Ca}^{2+}] \) for notational convenience, \( e = 1 \) to 60, \( \lambda \) is the characteristic length and is equal to \( \frac{D_{Ca}}{k^m_{m}[B]_{m}} \), the subscripts ‘\( x \)’ and ‘\( y \)’ denote derivatives of ‘\( u \)’ in respective directions. Also the second term \( (\mu^{(e)} = 1) \) for \( e = 30, 33 \) and \( (\mu^{(e)} = 0) \) for rest of the elements.

The source of calcium.

\[ \text{Assume a single mobile buffer specie, i.e. } [\text{Ca}^{2+}] \text{ is the characteristic length and is known as the reaction term and the third term is the area of the element and } \sigma \text{ is the area of the element.} \]

In this article, we have considered cytosol to be a circle of radius 5 \( \mu \) m. The centre of the circle is supposed to be situated at origin (i.e. \( x = 0, y = 0 \)). We have assumed that there is a point source of calcium situated at \( x = -5, y = 0 \). An appropriate flux condition for it can be framed as [5], [9],

\[ \lim_{x \rightarrow -5, y \rightarrow 0} -D_{Ca} \frac{\partial \text{[Ca}^{2+}]}{\partial x} = \sigma \]

For other boundary condition and initial condition, it is assumed that \( \text{[Ca}^{2+}] \) attains its steady state concentration of 0.1 \( \mu \) M as it goes far away from the source i.e.

\[ \lim_{x \rightarrow -5, y \rightarrow 0} \text{[Ca}^{2+}] = \text{[Ca}^{2+}]_\infty \]

Now, using (10 – 12) in (9) and extremizing (9) with respect to nodal concentration we have,

\[ \frac{\partial I^{(e)}}{\partial u_i} = \int_A \left[ N_i^{(e)} \left[ N_i^{(e)} \right]^T + N_j^{(e)} \left[ N_j^{(e)} \right]^T dA \right] \mu^{(e)} dA + \int_A \frac{1}{\lambda^2} \left[ N_i^{(e)} \left[ N_i^{(e)} \right]^T \right] \frac{\partial u_i}{\partial t} dA \]

Here, \( u^{(e)} = \left[ u_i, u_j, u_k \right]^T \). Assembling (13) for \( e = 1 \) to 60, we have,

\[ \frac{\partial I^{(e)}}{\partial u_i} = \sum_{e=1}^{60} \frac{\partial I^{(e)}}{\partial u_i} = 0 \]

where, \( i = 1, 2, \ldots, 37 \). Rearranging (14) and writing in matrix form, we have a system of ordinary differential equations (see Appendix for details).
\[
[K]_{37,37} \begin{bmatrix} \bar{u} \\ \end{bmatrix}_{37,1} + [M]_{37,37} \begin{bmatrix} \partial \bar{u} \\ \end{bmatrix}_{37,1} = [F]_{37,1} \quad (15)
\]

Here, \( \bar{u} = u_1, u_2, ..., u_{37} \), \([K]\) and \([M]\) are system matrices, and \([F]\) is the system vector. For the solution of (15), we have developed a computer program in MATLAB that uses numerical integration to approximate the solution at discrete time steps \([11]\). The time taken for simulating the mathematical model for 1 sec, while taking \( \Delta t = 0.0001 \) sec, is nearly 2 minutes on the aforesaid computer.

### III. NUMERICAL RESULTS AND DISCUSSION

In this section, numerical results are shown in the form of figures explaining the relationship observed between the physiological parameters. All the investigations were done assuming that cytosolic Ca\(^{2+}\) is buffered using 50 µM EGTA, Table I List of physiological parameters used for numerical results \([12]\), \([5]\), \([13]\),

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>(D_{Ca})</td>
<td>Diffusion coefficient</td>
<td>250 µm(^2).s(^{-1})</td>
</tr>
<tr>
<td>(k_m^{\text{(EGTA)}})</td>
<td>Buffer association rate</td>
<td>1.5 µM(^{-1}).s(^{-1})</td>
</tr>
<tr>
<td>(k_m^{\text{(BAPTA)}})</td>
<td>Buffer association rate</td>
<td>600 µM(^{-1}).s(^{-1})</td>
</tr>
<tr>
<td>(k_m^{\text{(Troponin-C)}})</td>
<td>Buffer association rate</td>
<td>90 µM(^{-1}).s(^{-1})</td>
</tr>
<tr>
<td>(k_m^{\text{(Calmodulin-D28K)}})</td>
<td>Buffer association rate</td>
<td>250 µM(^{-1}).s(^{-1})</td>
</tr>
<tr>
<td>([B_{\infty}^{\text{m}}}])</td>
<td>Buffer concentration</td>
<td>50 µM</td>
</tr>
<tr>
<td>([Ca^{2+}]_{\infty})</td>
<td>Background Ca(^{2+}) Concentration</td>
<td>0.1 µM</td>
</tr>
<tr>
<td>(\sigma)</td>
<td>Source amplitude</td>
<td>1 pA</td>
</tr>
<tr>
<td>F</td>
<td>Faraday’s Constant</td>
<td>96487 C/moles</td>
</tr>
<tr>
<td>V</td>
<td>Volume of the cytosol</td>
<td>523.6 µm(^3)</td>
</tr>
</tbody>
</table>

*All parameter values are taken as per Table I unless otherwise stated.

Here source amplitude is converted into µM.s\(^{-1}\) by using Faradays constant and using the fact that 1 L = 10\(^{15}\) µm\(^3\) to compute the results.

In Fig. 2, cytosolic diffusion is shown in x and y directions for time \(t = 100\) ms. As proposed, Ca\(^{2+}\) attains its background concentration of 0.1 µM as it goes far away from the Ca\(^{2+}\) channel. Since source amplitude is taken to be 1 pA therefore the highest Ca\(^{2+}\) concentration observed is 1176 µM. Thus, if an electrical signal arrives at the mouth of a VDCC, it can increase the intracellular Ca\(^{2+}\) concentration to an extent of 1176 µM. As observed by Brose et al. \([14]\), synaptotagmin is activated at high cytosolic [Ca\(^{2+}\)] \(\sim 10\) µM and not at lower Ca\(^{2+}\) concentrations. An ample amount of neurotransmitters are supposed be released and thus signal transduction can take place at this point of time. In Fig. 3, the effect of time over two-dimensional calcium profile is shown. In this figure changes with respect to time are observed for whole of the cytosol. It is apparent from the figure that calcium begins to rise slowly as time elapses and attains a steady state calcium concentration of 1176 µM. It was also observed that there is no change in calcium profile after 100 ms (not shown in this article) which means that calcium attains steady state after 100 ms.
time $t = 100$ ms. Fig. 4(A), 4(B), 4(C) and 4(D) are for buffer concentration taken to be 50 $\mu$M, 100 $\mu$M, 150 $\mu$M, and 200 $\mu$M, respectively. In all the four cases buffer specie is taken to be Ethylene Glycol - bis(beta – aminoethyl - ether)-N,N,N',N'-TetraAcetate(EGTA). As expected the increase in buffer concentration increases the decay of calcium which is evident in both the directions. In other words, it can be said that increase in buffer concentration alters the time required to achieve the steady state.

In Fig. 5, the effect of two diverse calcium chelators on cytosolic calcium profile is shown. These chelators are used to increase or decrease the time required by calcium to attain steady state.

![EGTA](image1)

Fig. 5 Change in calcium profile for two calcium chelators namely EGTA and BAPTA.

We used two exogenous buffers i) BAPTA (1,2-bis(o-minophenoxy)ethane-N,N,N',N'-tetraacetic acid) which is a very fast calcium chelator and ii) EGTA which is a very slow calcium chelator. It is observed from the figure that the highest calcium concentration is only 46.37 $\mu$M (see Fig. 5(B)) when cytosol is introduced to BAPTA while the same is about 1176 $\mu$M (see Fig. 5(A)) when cytosol is introduced to EGTA. It is so because when we introduce BAPTA inside the cytosol it binds calcium faster as compared to EGTA and reduces the free calcium concentration faster than EGTA.

![BAPTA](image2)

In Fig. 6 Ca$^{2+}$ diffusion in x-direction is shown, for different buffer types, just as if we are studying Ca$^{2+}$ diffusion in one-dimension. Because of the presence of different buffer types there is a variation in cytosolic Ca$^{2+}$ profile. Thus, before plotting the curves the [Ca$^{2+}$] values were normalized to make them comparable. To be specific, Ca$^{2+}$ diffusion was studied for three different buffer types namely, i) EGTA, ii) Troponin-C and iii) Calmodulin-D$_{28K}$. The solid curve is for Ca$^{2+}$ diffusion in the presence of 50 $\mu$M EGTA, the ‘*’ curve is for Ca$^{2+}$ diffusion in the presence of 50 $\mu$M Troponin-C and the ‘o’ curve is for Ca$^{2+}$ diffusion in the presence of 50 $\mu$M Calmodulin-D$_{28K}$. These results further validate our previous hypothesis, as for the given three buffers $\lambda$ values are decreasing and hence the time to achieve steady state is also decreasing. It can also be concluded from the figure that the time to achieve steady state is directly proportional to characteristic length constant ‘$\lambda$’. As, this characteristic length constant ‘$\lambda$’ depends upon association rate and buffer concentration.

![Ca$^{2+}$ diffusion with buffers](image3)

Fig. 6 Effect of different buffer types over 1–D Ca$^{2+}$ profile.

IV. CONCLUSION

The results shown in this paper are primarily for Ca$^{2+}$ diffusion in 2-D with relevance to buffers following EBA. Further, the results obtained in this paper are in agreement with the physiological facts. Some of the results obtained have also been observed by previous researchers but they were all for one–dimensional case. The finite element model developed is quite versatile and flexible as we are able to incorporate the minute details of processes involved and study the effect of excess buffers on calcium diffusion in cytosol. There is a significant variation in calcium profiles due to various excess buffers used in the present problem. Further, the results obtained can be of great use to biomedical scientists for development of new protocols for treatment and diagnosis of neuronal diseases.

APPENDIX

When we extremize (9) we have,

$$\frac{\partial I^{(e)}}{\partial u} = \int_{\lambda} \left[ \frac{\partial u^{(e)}}{\partial x} \frac{\partial u^{(e)}}{\partial x} \right] dA$$

(16)

Also from (10) we have,

$$\frac{\partial u^{(e)}}{\partial x} = \left[ \frac{\partial N_i}{\partial x} \frac{\partial N_j}{\partial x} \frac{\partial N_k}{\partial x} \right] u^{(e)}$$

$$\frac{\partial}{\partial u_i} \left( \frac{\partial u^{(e)}}{\partial x} \right) = \frac{\partial N_j}{\partial x}$$

(17)
\[
\frac{\partial u^{(e)}}{\partial t} = \left[N\begin{bmatrix} \frac{\partial u_i}{\partial t} & \frac{\partial u_j}{\partial t} & \frac{\partial u_k}{\partial t} \end{bmatrix}\right]^T
\]

(18)

where, \( [N] = \begin{bmatrix} N_i & N_j & N_k \end{bmatrix} \). Thus, (16) can be written as,

\[
\frac{\partial F^{(e)}}{\partial u_i} = K^{(e)} u^{(e)} + M^{(e)} u^{(e)} - F^{(e)} = 0
\]

(19)

where \( \dot{u} \) represents time derivative of \( u \) and \( K^{(e)}, M^{(e)}, F^{(e)} \) are given by,

\[
K^{(e)} = \int_A \left( \frac{\partial N^{(e)} \partial N^{(e)T}}{\partial x} + \frac{1}{\lambda^2} \frac{\partial N^{(e)} \partial N^{(e)T}}{\partial y} \right) dA
\]

(20)

\[
M^{(e)} = \frac{1}{D_C} \int_A N^{(e)} dA
\]

\[
F^{(e)} = \mu_0 \int_A N^{(e)} dA + \mu_0 \int_A \sigma \frac{\partial N^{(e)}}{\partial t} dA
\]

(21)

Further from (11) we have,

\[
\frac{\partial N_{\alpha}}{\partial x} = b_\alpha
\]

\[
\frac{\partial N_{\alpha}}{\partial y} = c_\alpha
\]

(22)

where, \( \alpha = i, j, k \). Also, since our triangle is linear by using factorial formula we have [16],

\[
\int_A \frac{\partial N^{(e)} \partial N^{(e)T}}{\partial x} dA = \frac{1}{4A^{(e)}} \begin{bmatrix} b_j^2 & b_j b_k & b_k^2 \\ b_j b_k & b_j^2 & b_k^2 \\ b_k^2 & b_k^2 & b_k^2 \end{bmatrix}
\]

\[
\int_A \frac{\partial N^{(e)} \partial N^{(e)T}}{\partial y} dA = \frac{1}{4A^{(e)}} \begin{bmatrix} c_i^2 & c_i c_j & c_i c_k \\ c_j c_i & c_j^2 & c_j c_k \\ c_k c_i & c_k c_j & c_k^2 \end{bmatrix}
\]

(23)

\[
\int_A N^{(e)} N^{(e)T} dA = A^{(e)} \begin{bmatrix} 2 & 1 & 1 \\ 1 & 2 & 1 \\ 1 & 1 & 2 \end{bmatrix}
\]

\[
\int_A N^{(e)} dA = \frac{A^{(e)}}{3} \begin{bmatrix} 1 \\ 1 \\ 1 \end{bmatrix}
\]

For the assembly of all the elements, we write,

\[
K = \sum_{e=1}^{36} D^{(e)} K^{(e)} D^{(e)T}
\]

\[
M = \sum_{e=1}^{36} D^{(e)} M^{(e)} D^{(e)T}
\]

(24)

\[
F = \sum_{e=1}^{36} D^{(e)} F^{(e)}
\]

Thus for the whole system we have,

\[
[K]_{37 \times 37} [u]_{37 \times 1} + [M]_{37 \times 37} \left[ \frac{\partial u}{\partial t} \right]_{37 \times 1} = [F]_{37 \times 1}
\]

ACKNOWLEDGMENT

The authors are highly grateful to Department of Biotechnology, New Delhi, India for providing support in the form of Bioinformatics Infrastructure Facility for carrying out this work.

REFERENCES


