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Validating a Coarse-Grained Voltage Activation Model by Comparing Its Performance to the Results of Monte Carlo Simulations

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Abstract

Simulating the nature of voltage-activated systems is a problem of major current interest, ranging from the action of voltage-gated ion channels to energy storage batteries. However, fully microscopic converging molecular simulations of external voltage effects present a major challenge, and macroscopic models are associated with major uncertainties about the dielectric treatment and the underlying physical basis. Recently we developed a coarse-grained (CG) model that represents explicitly the electrodes, the electrolytes, and the membrane/protein system. The CG model provides a semimacroscopic way of capturing the microscopic physics of voltage-activated systems. Our method was originally validated by reproducing macroscopic and analytical results for key test cases and then used in modeling voltage-activated ion channels and related problems. In this work, we further establish the reliability of the CG voltage model by comparing it to the results of Monte Carlo (MC) simulations with a microscopic electrolyte model. The comparison explores different aspects of membrane, electrolyte, and electrode systems ranging from the Gouy-Chapman model to the determination of the electrolyte charge distribution in the solution between two electrodes (without and with a separating membrane), as well as the evaluation of gating charges. Overall the agreement is very impressive. This provides confidence in the CG model and also shows that the MC model can be used in realistic simulation of voltage activation of membrane proteins with sufficient computer time.

Graphical Abstract

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Notes

The authors declare no competing financial interest.

Supporting Information

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Additional details of the protocols and models used, justification of procedures used, minimum image convention, estimation of the long-range electrostatic interactions, and energy plots (PDF)

1. INTRODUCTION

Recent years have involved great progress in structural and biophysical studies of voltage-activated ion channels (e.g., refs 1–5). Unfortunately, we still do not have a full detailed physical understanding of the relevant structure-function correlation. Furthermore, despite significant progress in computational modeling of ion channels and ion selectivity,^{6–10} as well as voltage activation^{11–13} of such systems, the coupling to the external voltage is far from being understood. Using fully microscopic atomistic simulations for advancing our understanding of voltage activation may suffer from serious convergence problems. In fact, even simulations with massive computer power (e.g., ref 14) have probably not reached convergence, for example, the electrolyte environment needed unrealistically large potentials for activation and could not produce relevant free energies.¹⁴ Thus, one needs to explore less rigorous approaches.

One may consider continuum models, but unfortunately, despite interesting Poisson-Boltzmann (PB) macroscopic studies that evaluated gating charges,¹⁵ the corresponding perspective may cause very serious problems as it did in early nonmicroscopic studies of electrostatic interactions in proteins (see a review in ref 16).

Another option is to use semimacroscopic models with realistic electrostatic treatment such as the PDL/D-S-LRA¹⁶ that has provided a practical and realistic strategy for modeling the multi-ion current^{6,17} and yielded a very physical, rational nature of the actual selectivity (relating mainly to the change in the effective ion-ion dielectric and the corresponding repulsion). However, such a model does not consider the protein conformational energy and has not been extended to describe external voltage effects.

In order to reduce the problems mentioned above, we have developed a powerful coarse-grained (CG) model for describing protein/membrane systems that are subjected to external potential (e.g., refs 18 and 19). This model focuses on efficient, consistent, and reliable treatment of the description of the electrode potential, using semimacroscopic representation of the electrolytes in the solution separating the electrodes and the membrane, as well as consistent representation of the combined potential from the electrolytes and electrodes and determination of the protein ionization states. Our CG model goes very significant way beyond the PB bulk model in providing much clearer physical insight on the response to the

voltage and charge distribution of electrolytes in membrane protein systems, including near the electrodes and the membrane.

Nevertheless, although the CG model appeared to be very powerful, it has been validated only by comparing to PB and analytical results, rather than to microscopic results that, of course, face major convergence problems. In this work, we attempt to push further the validation process by comparing the results of the CG model to that of Monte Carlo (MC) simulations.

The MC simulations are based on powerful implementation that allows us to explore membrane, electrolyte, electrode systems in a reasonable computer time. Remarkably, the results of the MC and CG models appear to be very similar, further validating the CG modeling.

2. METHODS SECTION

2.1. The MC Model.

To simplify the structure of the electrolyte solution, the molecules of the solvent are excluded from the simulated system. The implicit effect of the solvent is represented by an effective Coulomb's law dielectric constant between the ions in the system. The ions are introduced in the system as hard spheres with a certain diameter, where the charge of each ion is located at the center of the sphere. Such a simplification of the electrolyte solution is basically the primitive electrolyte model (PM).²⁰ Following further the PM, a pair of electrodes, whose role is to confine the system in the z direction and create a potential, is introduced as nonmaterial infinite parallel sheets with a certain surface charge density. These simplifications are the basis to draw the simulation box (unit cell) adopted by the MC model of the simulation implemented here.

Figure 1a schematically represents the size, shape, and content of the employed unit cell with orthogonal unit vectors, where the lower left vortex of the box coincides with the center of the coordinate system (0,0,0). Its volume is $W \times W \times L$, where L is the length and W is the thickness. The implicit solvent charge-charge dielectric constant is set to $\epsilon^{\text{wat}} = 80$. In total, the simulation box contains $N = N_{\text{Na}} + N_{\text{Cl}}$ ions, where $N_{\text{Na}} = N_{\text{Cl}} = N/2$ are the corresponding total numbers of Na^+ and Cl^- and $N_p = N(N-1)/2$ is the number of ion pairs. Each pair is denoted as $\{q_p, q_q\} \kappa$, where $p = 1, \dots, N-1$ and $q = p+1, \dots, N$ are the unique indexes of the p th and q th ions, k is the unique index of the pair computed as $k = N(p-1) - [p(p-1)/2] + q$, and q_p and q_q are the relative ion charges (initially +1 for Na^+ and -1 for Cl^-). The distance between the ions engaged in the k th pair, r_k , is taken by means of periodic boundary conditions (PBCs) in the x and y directions. When the simulation requires the presence of a membrane, it is implicitly introduced as a rectangular void with volume of $W \times W \times L_M$, where $L_M = L_R - L_L$ is the membrane thickness, as shown in Figure 1b, where L_L and L_R are the positions of the left and right ends of the membrane in the z direction. The presence of the membrane changes the initial setup of the unit cell, which divides the cell volume into two equal subvolumes, each one accommodating half of the total number of Na^+ and Cl^- ions. If the membrane is permeable, the ions could freely jump between the subvolumes. This process requires no special protocol for traversing the ions

through the membrane because we do not study the dynamics of the process. The distance between the i th ion and the closest surface of the membrane is defined as $z_{i,m} = |z_m - z_i|$, where z_m is either L_R or L_L , depending on the subvolume where the i th ion is located.

The implicit electrodes confining the system as parallel planes at $z_l = 0$ and $z_r = L$ have been assigned charge densities $q_{s,l}/W^2$ (on the left plane) and $q_{s,r}/W^2$ (on the right plane), where $q_{s,l}$ and $q_{s,r}$ are the corresponding relative charges (see Figure 2a). The electrostatic interactions between the i th ion ($i = 1, \dots, N$) located at $z = z_i$ and the left and right electrodes depend on only the shortest z distances between that ion and the planes, $z_{i,l} = |z_i - z_l|$ and $z_{i,r} = |z_i - z_r|$.

To run the MC simulation, the initial positions of the ions inside of the unit cell should be generated free of collisions. Here, a collision is any distance of direct contact between a pair of ions, which is below the given critical minimum value, d ("exclusion diameter"). Unlike most classical studies of PM MC simulations, where the spheres of both Na^+ and Cl^- ions have an exclusion diameter $d_{\text{Na}} = d_{\text{Cl}} = d = 4.25 \text{ \AA}$,²¹ this study adopts a slightly smaller one: $d = 2.5 \text{ \AA}$. Such value of d allows one to implement the 12–6 Lennard-Jones (LJ) potential²² as an instrument to treat most of the collisions. Because the new value of d mostly affects the ion-pairing process in the bulk, we keep the thickness of the exclusion layer at the electrodes to its classical value of $d^* = 4.25/2 = 2.175 \text{ \AA}$. Section S1 in the SI explains how the ions are settled at their initial positions free of collisions and the reason for adopting smaller d in a combination with the LJ potential. Because the electrodes are not explicitly introduced as atomic structure, it is not possible to implement the LJ potential to treat the collisions that might occur between the ions and the planes. In this particular case, we follow the classical way of avoiding the collisions by rejecting any direct contacts of the ion-electrode, $z_{i,l}$, $z_{i,r}$, which are below d^* (see Figure S1 in the SI).²¹ As for the corresponding lower limit of the direct contact between any ion and the closest plane of the membrane, $z_{i,m}$, it is set to $5 \text{ \AA}$, based on the current CG setup structural parameters.¹⁸

When performing the MC simulation, every newly derived set of coordinates (proposal) is obtained based on the previous one by executing the displacement protocol described in section S3 in the SI. The method of generating proposals guarantees them to exclude collision events. In addition, to ensure the spatial randomness of the newly derived ion positions, the Mersenne Twister random number generator is employed.²³

Every proposed ion coordinate set, which is generated free of collisions, should be sent to a rejection sampling subroutine, which will decide whether to accept or reject the proposal, based only on the total potential energy of the system. The one employed here is based on the Metropolis-Hastings method.²⁴ According to the method, a proposal is acceptable only if

$$\min\{1, \exp(-\Delta U/RT)\} > \rho \quad (1)$$

where $U = U_2 - U_1$ is the difference between the potential energies of the proposal under examination, U_2 , and the previously accepted one, U_1 , $T = 298.15 \text{ K}$, and ρ is a random

number uniformly distributed in $[0,1)$. If there is no point-charge accommodated inside of the membrane, the total potential energy of the system is defined as

$$U \equiv U_{\text{SR}} + U_{\text{LR}} + U_{\text{IE}} + U_{\text{LJ}} \quad (2)$$

where U_{SR} is the potential of the short-range electrostatic interactions between the ions, U_{LR} takes into account the longrange electrostatic interactions,²¹ U_{IE} is the potential of the interactions between the ions and electrodes,²¹ and U_{LJ} is the 12–6 LJ potential between the ions. In explicit form, eq 2 expands to

$$U = \frac{f}{\epsilon^{\text{wat}}} \sum_{k=1}^{N_p} \frac{Q_k}{r_k} - \frac{f}{\epsilon^{\text{wat}} W^2} \sum_{k=1}^{N_p} Q_k \{ 2\pi \Delta z_k + \phi(\Delta z_k, W) \} \quad (3)$$

$$- \frac{2\pi f}{\epsilon^{\text{wat}} L^2} \sum_{i=1}^N q_i (\Delta z_{i,l} q_{s,l} + \Delta z_{i,r} q_{s,r}) + 4 \sum_{k=1}^{N_p} \epsilon_k \left\{ \left(\frac{\sigma_k}{r_k} \right)^{12} - \left(\frac{\sigma_k}{r_k} \right)^6 \right\}$$

Here, f is the electric conversion factor of 332 kcal/mol/Å, $Q_k = q_p q_p$ is the product of the relative atomic charges of the k th ion pair, r_k is the distance between the ions of the k th pair, $z_k = |z_p - z_q|$ is the z -component of r_k , the supplementary function $\phi(\Delta z_k, W)$ is defined as²¹

$$\phi(\Delta z_k, W) \equiv 4W \ln \left(\frac{0.5 + r_1}{r_2} \right) - \Delta z_k \left\{ 2\pi - 4 \arctan \left(\frac{4r_1 \Delta z_k}{W} \right) \right\} \quad (4)$$

with the help of the notations

$$r_1 = \sqrt{0.5 + (\Delta z_k / W)^2} \quad r_2 = \sqrt{0.25 + (\Delta z_k / W)^2} \quad (5)$$

and the Lennard-Jones parameters for the k th pair

$$\epsilon_k \equiv \sqrt{\epsilon_p \epsilon_q} \quad \sigma_k \equiv \frac{1}{2}(\sigma_p + \sigma_q) \quad (6)$$

are computed using the values of ϵ_p , ϵ_q , σ_p , and σ_q given in Table 1. Because r_k is part of the expressions defining U_{SR} and U_{LJ} , the minimum image approach, which is described in section S4 in the SI, is applied only to the first and last terms in eq 3.

When an ionized residue, with a relative net atomic charge q_r , is accommodated inside of the membrane of the CG model, the MC method follows it by introducing a fixed point-charge with a relative atomic charge q_r at position $(W/2, W/2, z_r^*)$. Here z_r^* , $L_L < z_r^* < L_R$, is the z position of the charge introduced in CG. Subsequently, eq 2 needs to be extended into

$$U^* \equiv U + U_{\text{SR}}^* + U_{\text{LR}}^* \quad (7)$$

where U is the potential energy given by eq 2 and U_{SR}^* and U_{LR}^* are the short- and long-range electrostatic interaction potentials between the fixed point-charge in the membrane and the mobile ions located inside of the subvolumes. Due to the positioning of the fixed point-charge at the geometrical center of the intersection of the box in xy , the minimum image approach is not required for computing U_{SR}^* . Also, the 12–6 LJ interactions are excluded from eq 7 because the fixed point-charge and any of the ions are always kept at least $\sim 2.5 \sigma$ apart, based on the defined gaps.

In more explicit form, eq 7 reads

$$U^* \equiv U + f \sum_{i=1}^N \frac{Q_i^*}{\epsilon_{s,i}^* r_i^*} - \frac{f}{W^2} \sum_{i=1}^N \frac{Q_i^*}{\epsilon_{s,i}^*} \{2\pi \Delta z_i^* + \phi(\Delta z_i^*, W)\} \quad (8)$$

where Q_i^* is the atomic charge product $q_i q_j$, r_i^* is the distance between the fixed ion and i th mobile ion, Δz_i^* is the z component of r_i^* , and $\epsilon_{s,i}^*$ is the reduced distance-dependent dielectric constant, defined as¹⁶

$$\epsilon_{s,i}^* = 1 + 60 \{1 - \exp(-0.1 r_i^*)\} \quad (9)$$

Two important clarifications regarding the computation of U are required to be given at this point. First, there is an alternative method for taking the long-range electrostatic potential in confined electrolyte solutions.²⁵ However, despite its solid theoretical background,²⁶ it is not employed here due to its high computational cost caused by multiple calls of the modified Bessel function of the second kind, K_0 . Second, the use of the 12–6 LJ potential is not a typical part of most of the published PM MC protocols. The goal of having it is to support fast equilibration and reconfiguration of the system by increasing the probability predicted by eq 1 for some of the proposals.

When simulating a system without a membrane or without a fixed point-charge inside of the membrane, only one simulation per content is required to obtain the charge distribution profiles. It was found that 10 000 000 is the optimal number of simulation steps that reduces the data noise in the profiles, providing each one out of 1000 frames is appended to the simulation trajectory.

The MC procedure is also used for calculating gating charges. The CG calculation of the gating charge started with an initial equilibration where the electrolytes were allowed to move across the membrane and then (after movement of the permanent charge) allowed to equilibrate, without the possibility of moving across the membrane (more discussion on the nature of the gating charge is given in ref 18).

The MC gating charge calculations require accommodation of a fixed point-charge inside of the membrane and two-stage simulation. Each stage should perform at least 30 000 000 steps to guarantee a low noise level in the results. Reducing the data noise requires one to perform more steps, compared to the system with no membrane. Before running Stage 1 simulation, the fixed point-charge is introduced at $z_r^* = z_{r,1}^*$, and the membrane is set to be permeable. This allows establishment of total charge excess on both sides of the membrane during the simulation. To evaluate the total charge excess, the average number of ions of a given type, located inside of the respective subvolume of the simulation box, $\langle N_{I,J} \rangle$, should be computed based on the frames captured in the trajectory

$$\langle N_{I,J} \rangle = \frac{1}{N_f} \sum_{i=1}^{N_f} N_{I,J,i} \quad (10)$$

Here, N_f is the total number of processed frames (30 000 000 as recommended), and $N_{I,J,i}$ is the total number of ions of sort I ($I = \text{Na}^+, \text{Cl}^-$) located inside of the subvolume J ($J = \text{L}(\text{eft}), \text{R}(\text{ight})$), found when processing the i th frame. Once $\langle N_{I,J} \rangle$ is known, the total charge excess, Q_J^e , is calculated as the ensemble average

$$Q_J^e \equiv \langle N_{\text{Na},J} \rangle q_{\text{Na}} + \langle N_{\text{Cl},J} \rangle q_{\text{Cl}} \quad (11)$$

The numerical values of $\langle N_{I,J} \rangle$ and Q_J^e obtained based on Stage 1 simulations results are given in Table 2, and Figure 7 illustrates the averaging process.

Next, in Stage 2 simulations, the membrane is not permeable anymore, the fixed point-charge is moved to $z_r^* = z_{r,2}^*$ (close to the opposite side of the membrane), and the total charge excess previously established is kept the same. Before running Stage 2 simulations, the atomic charges of Na^+ and Cl^- need to be recomputed to keep the charge excess obtained during Stage 1 the same. This step is required because the excess is a floating-point number, but the membrane is no more permeable, and the number of ions inside of each subvolume is an integer. The new atomic charges, q_{Na}^* and q_{Cl}^* , are the solution of the following system of equations

$$\begin{cases} N_{\text{Na},\text{L}} q_{\text{Na}}^* + N_{\text{Cl},\text{L}} q_{\text{Cl}}^* = \langle N_{\text{Na},\text{L}} \rangle q_{\text{Na}} + \langle N_{\text{Cl},\text{L}} \rangle q_{\text{Cl}} \\ N_{\text{Na},\text{R}} q_{\text{Na}}^* + N_{\text{Cl},\text{R}} q_{\text{Cl}}^* = \langle N_{\text{Na},\text{R}} \rangle q_{\text{Na}} + \langle N_{\text{Cl},\text{R}} \rangle q_{\text{Cl}} \end{cases} \quad (12)$$

where $N_{\text{Na},\text{L}}$, $N_{\text{Cl},\text{L}}$, $N_{\text{Na},\text{R}}$, and $N_{\text{Cl},\text{R}}$ are estimated by rounding $\langle N_{\text{Na},\text{L}} \rangle$, $\langle N_{\text{Cl},\text{L}} \rangle$, $\langle N_{\text{Na},\text{R}} \rangle$, and $\langle N_{\text{Cl},\text{R}} \rangle$ to the closest integer. Thus, the obtained $N_{\text{Na},\text{L}}$, $N_{\text{Cl},\text{L}}$, $N_{\text{Na},\text{R}}$, and $N_{\text{Cl},\text{R}}$ are introduced into the subvolumes of the simulation box by means of the protocol described in section S1 in the SI. Now, the MC simulations take 30 000 000 steps. Similar to

Stage 1, the atomic coordinates derived after extracting each one out of 1000 steps are saved as a frame to the trajectory to support the data analysis later.

Once Stage 2 simulations are completed, the accumulation of the atomic charges based on the difference between q_{Na} and q_{Cl} alongside the z axis of the simulation box gives the charge accumulation, Q_z

$$Q_z^{(j)} = \sum_{i=1}^{N_z} \langle N_{\text{Na}} \rangle_i |q_{\text{Na}}| - \langle N_{\text{Cl}} \rangle_i |q_{\text{Cl}}| \quad (13)$$

where $j = 1, 2$ is the identifier of the stage of the simulation, N_z is the number of bins defined along the z axis of the simulation box, and $\langle N_{\text{Na}} \rangle_i$, $\langle N_{\text{Cl}} \rangle_i$ are the average numbers of Na^+ and Cl^- inside of the i th bin, respectively. Finally, the gating charge, Q_g , is estimated as the difference between the accumulated charge from Stage 1, $Q_z^{(1)}$, and Stage 2, $Q_z^{(2)}$

$$Q_g = |Q_z^{(2)} - Q_z^{(1)}| \quad (14)$$

2.2. The CG Model.

Our CG model has been described elsewhere (e.g., refs 18 and 19, and thus, we do not give here the details of the model. Overall the model is based on representing the electrolytes by a grid of induced charges, a neutral membrane (with possible permanent charges), and two electrodes, as shown in Figure 2b. Each grid point is assigned to a volume element ($\tau = \text{\AA}^3$), and then, we place at the center of the i th grid site a residual charge, q_i^g , defined by

$$q_i^g = q_i^+ + q_i^- \quad (15)$$

where

$$q_i^\pm = \frac{\alpha^\pm (N_{\text{box}}^\pm + N_{\text{box}}^\pm e^{\mp\beta\phi_i})}{(\sum_{i \in \text{box}} e^{\mp\beta\phi_i} + N_{\text{bulk}}^{\text{grid}} e^{\mp\beta\phi_{\text{bulk}}})} \quad (16)$$

where q_i^+ and q_i^- are the positive and negative residual charges, respectively, that are placed on the i th grid point. $\alpha^\pm$ is the total charge, in atomic units, of the electrolyte ions, $N_{\text{box}}^\pm$ is the total number of cations/anions in the simulation volume, $Q_{\text{box}}^\pm$ (the total charge in the simulation box) is defined by $Q_{\text{box}}^\pm = \alpha^\pm N_{\text{box}}^\pm$. ϕ_i is the potential (times a unit charge) at the i th grid site, and $\beta = (k_B T)^{-1}$. $N_{\text{bulk}}^{\text{grid}}$ is the number of grid points in the bulk region, while ϕ_{bulk} is the potential at the bulk grid points. ϕ_i can be expressed as

$$\phi_i = 332 \sum_j \frac{q_j^p}{\epsilon_{\text{eff}}^{\text{gp}} r_{ij}} + 332 \sum_{k \neq i} \frac{q_k^g}{\epsilon^{\text{wat}} r_{ik}} + V_i^{\text{ext}} \quad (17)$$

where V_i^{ext} is the external potential (multiplied by a unit charge) at the i th grid site. q_j^p is the charge of the j th protein residue, and q_k^g is the point-charge at the k th grid site. The final set of grid charges, q^g , is determined iteratively.

The long-range electrostatic interaction is treated by a partial periodic treatment, applying a periodic model with three additional layers in each $+x$, $-x$, $+y$, and $-y$ direction (this leads to sufficient charge convergence). The eight nearest neighbor images surrounding the unit cell in the xy plane include the explicit grid image nodes, whereas the averaged charges are used beyond the eight nearest images to simplify the periodic replicas (see details in refs 18 and 19).

3. RESULTS AND DISCUSSION

To validate our CG model, we have examined it on several levels, as described below.

3.1. Gouy-Chapman Potential.

We started our analysis by verifying the ability of our CG model to predict the potential and the charge distribution of the electrolytes near a charged surface at a given ionic strength. The resulting distribution, frequently determined by the Gouy-Chapman (GC) approximation,²⁷ has been studied extensively by PB, MC, and MD simulations.^{20,21,28–36} Hence, it presents a useful test for the agreement between the CG and the MC models. To conduct a CG simulation, we introduced a system containing 0.01 M electrolyte (Debye length is ~ 30 Å) in a unit cell with dimensions $200 \times 200 \times 200$ Å, with no membrane inside. The charges of the ions were spread on the grid nodes, and an external potential of 0.1 V was created by a pair of explicit electrodes. Then, the charge distribution profiles were established by letting any of the charges located on a grid node interact with the rest of the charges and with the charges on the electrodes.

The corresponding MC simulation was conducted inside of a system of the same size and electrolyte concentration, but the Na^+ and Cl^- ions of the electrolyte were accommodated as point-charges, following the primitive model. The external potential was created based on the implicit electrodes with charges density $10 |e|/200/200 \text{ C}/\text{\AA}^2$ (see also section 2.1). Then, the charge distribution was established by randomly displacing the ions and accept those of the displacements matching the Metropolis- Hastings method criteria. Finally, the charge distribution profiles were processed by means of eq 18 to compute the potential.²¹

$$\Psi(z) = 4\pi \int_z^H [q_+ n_+(z') + q_- n_-(z')] (z - z') dz' \quad (18)$$

where $n_+(z')$ and $n_-(z')$ are the density distributions of the cation and anion, respectively. In addition, q_+ and q_- are the charges of the cation and anion. This is plotted in Figure 3 along with the one provided by the CG model.

3.2. Two Electrodes Case.

The performance of our approach was explored next by considering a system composed of two electrodes (with a 0.1 V potential difference) and electrolytes. In the CG model, we considered two different approaches for incorporating the electric field in the simulation system. The first approach employs a displacement vector, and the second one introduces explicit charges on the electrodes (both approaches are described in ref 18). The CG charge distributions along the z axis, obtained by these two different approaches, are given in Figure 4 where they are compared to the distribution obtained by the explicit MC model (with the same parameters of the simulation reported in section 3.1). As seen from the figure, the CG and MC models produce very similar results.

3.3. Neutral Membrane.

Next, we examined the performance of our models for a system with a 0.1 V external potential (introduced as a displacement vector), electrolyte solution, and neutral membrane. The calculations were done inside of a simulation box of a $200 \times 200 \times 200$ Å dimension, a membrane with a $L_M = 40$ Å thickness, and an electrolyte concentration of 0.1 M, while forcing both sides of the membrane to stay electroneutral. This is equivalent to equilibration of the electrolytes while not allowing charge exchange through the membrane.

The MC simulation was performed by following the protocol given in section 2.1 and using the parameters reported in section 3.1. Figure 5 compares the results obtained by both methods, which are practically the same. The plotted charge distribution profiles establish that the electrolytes lead to an almost complete screening of the external potential and start to change only near the membrane. Such a trend is similar to that found in macroscopic treatments, but it involves much more challenging explicit treatment of the electrolytes.

3.4. Gating Charge.

As a final test, we considered the effect of moving an ARG residue with a net charge of +16 $|e|$ located on the $C\beta$ atom, across the membrane. Within the MC approach, that atom is handled as a fixed point-charge (see section 2.1). The membrane is $L_M = 40$ Å thick, with its geometrical center at $z = 200$ Å, accommodated inside of a unit cell with $80 \times 80 \times 400$ Å dimensions, immersed in 0.1 M 1:1 electrolyte solution. The CG model represents a potential of 0.1 V by means of a displacement vector, while the MC approach sets a charge density of $0.85 |e| / 80/80 \text{ C}/\text{\AA}^2$ on the implicit electrodes, which also corresponds to a voltage of ~ 0.1 V. Initially, the $C\beta$ atom was positioned in the left part of the (permeable) membrane at $(0, 0, 190)$ Å. The MC method implementation of this part is described as Stage 1 simulations in section 2.1. After establishing the equilibrium charge distribution, the $C\beta$ atom was moved to $(0, 0, 210)$ Å and the membrane was changed from permeable to nonpermeable. Then, a new stage of the simulation was executed (Stage 2 in the case of the MC method; see section 2.1) to establish a new charge distribution. The corresponding

simulation model is in some respect a simplified way of generating the trend in the Kv1.2 voltage-activated channel.^{18,19}

The estimation of the gating charge, based on the difference between the accumulated charge profiles,^{18,37} is shown in Figure 6, where the CG model predicts 1.323 |e| while the MC method gives 1.409 |e|. It should be noticed that the CG and MC methods adopt different ways of keeping the charge excess accumulated on both sides of the membrane during the first simulation (when the membrane is permeable). The CG model distributes the charge of the ions in the electrolyte as partial charges spread on grid nodes, while the MC approach keeps the charge excess by correcting the atomic charge of the ions. Figure 7 represents the accumulation of the charge excess observed during Stage 1 simulation. More discussion on the nature of the gating charge is given in refs 18 and 37. It is important to note that the results of this figure are not supposed to be equal to that of Figure 9 in ref 18 because in that work we did not use periodic replicas of the charges in the membrane. However, in view of the difficulty of using non periodic treatment for the membrane charges in the MC implementation, we used here a periodic treatment of the membrane charges, in both the MC and CG studies. At any rate, the main point here is that MC and CG give the same results for the same system.

4. CONCLUDING REMARKS

Elucidating the microscopic nature of voltage-activated biological systems is important for both fundamental and practical interest. However, most current models are based on macroscopic concepts^{38–41} that may not represent correctly the underlying microscopic reality. For example, while continuum studies of the potential and electrolyte distribution near membranes have been useful, the description of the electrolytes' behavior at a distance from the membrane and in proximity to the electrodes has not been explicitly considered. On the other hand, one expects major convergence problems in the all-atom MD description of electrolyte equilibration.

In view of the above difficulties, we developed our CG model that treats the electrolyte explicitly but in a simplified way. This model has been found to be very powerful but has not been validated in a rigorous way. Thus, we look for a way to obtain a microscopic validation.

In trying to validate the CG electrolyte model, we note that MC models have been very instrumental in simulations of electrolyte behaviors in simple cases,^{20,42} although such studies have not been extended (to the best of our knowledge) to studies of the membrane potential and the modeling of voltage-activated channels.

Here, we decided to use MC simulations in validating the CG model. Encouragingly, we found out that the CG approach reproduced the more rigorous MC results. This provides evidence that the CG model is a very powerful tool for modeling voltage activation processes.

Interestingly, although MC is significantly more expensive than the CG model, we found out that our version of the MC model should be capable describing complex systems including voltage-activated biological systems and electric cells.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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ABBREVIATIONS

CG	Coarse-grained
MC	Monte Carlo
PM	Primitive electrolyte model
LJ	Lennard-Jones

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Figure 1. Schematic representation of the shape, dimensions, and content of the simulation box (unit cell) confined by implicit electrodes (without a membrane (a) and with a membrane (b)), employed here by the MC simulation protocol. The box unit vectors form an orthogonal basis. See section 2.1 for more information regarding parametrization of the system.

Figure 2.

Schematic comparison between the approaches to the spatial charge distribution provided by (a) the employed MC simulation protocol and (b) the CG model. Despite sharing the same membrane abstraction, they implement a different way of changing the charge spatial distribution during the simulation. The MC approach follows the PM, where any mobile point representing an ion in the solution is assigned a fixed charge, while the CG model deals with induced partial charges assigned to fixed grid nodes.

Figure 3. Comparison of the GC potential, obtained by MC and CG models. The blue line is the result of the MC simulation, and the red one is the prediction given by the CG model.

Figure 4.

Plots of the total charge distribution obtained within a solution of 0.1 M 1:1 electrolyte under external potential, by means of MC and CG models. Those correspond to the CG compare the two available methods for introducing the potential: a displacement vector¹⁸ or explicit electrode.¹⁸ The MC plot corresponds to the adopted system confined by implicit electrodes (see Figure 1 and section 2.1).

Figure 5. Charge distribution obtained by MC and CG models across a system divided by a nonpermeable membrane, under an external potential. Inside of the subvolumes defined by the membrane, 0.1 M 1:1 electrolyte is introduced. See section 3.3 for more details and discussion.

Figure 6.

Gating charge estimated as the maximum difference between the profiles of the cumulative charge obtained for a system with a fixed point-charge of +16 e, accommodated inside of the membrane. The gating charge obtained by (a) MC and (b) CG approaches is compared. Note that the results of this figure are not supposed to be equal to that of Figure 9 of ref 18 because in that work we did not use periodic replicas of the charges in the membrane (see section 3.4).

Figure 7. Charge excess collected by analyzing the frames of Stage 1 MC simulations trajectory (see section 2.1). The dashed lines visualize the (ensemble) average charge excess, computed by eq 11 for each subvolume.

Table 1.

12–6 Lennard-Jones Parameters for the Interaction between Na^+ and Cl^- , Taken from the Parameter Library Supporting the AMBER99 Force Field⁴³

ion	ϵ [kcal/mol]	σ [Å]
Na^+	0.0028	3.328
Cl^-	0.1000	4.401

Table 2.

Average Number of Na^+ and Cl^- Ions, $\langle N_{\text{I},J} \rangle$, and the Total Charge Excess, Q_J^e , Obtained for Each of the Subvolumes during the Stage 1 Simulation

subvolume	$\langle N_{\text{Na},L} \rangle$	$\langle N_{\text{Cl},L} \rangle$	Q_J^e
left ($J=1$)	80	82	-1.7749
right ($J=2$)	80	78	1.7749