

Computer Simulation Studies of Model Biological Membranes

LEONOR SAIZ* AND MICHAEL L. KLEIN

*Center for Molecular Modeling and Chemistry Department,
University of Pennsylvania, 231 South 34th Street,
Philadelphia, Pennsylvania 19104-6323*

Received January 23, 2002

ABSTRACT

This Account is focused on computer simulation studies of model biological membrane systems with potential applications in biomedical research. In the past decade, classical molecular dynamics has provided novel insights into the properties of model biomembrane systems, including the nature of the DNA–lipid interactions, the effect of pore-forming transmembrane peptides on the lipid environment, and the partitioning of volatile anesthetic molecules. Such simulations, employing full atomic detail, are typically restricted to systems of dimensions less than ~ 10 nm. Simplified models of the coarse-grain type have been intended to bridge the gap between full atomistic detail and the mesoscopic (micron) regime. The use of such models is illustrated with the example of anesthetics in a phospholipid bilayer.

Introduction

Biological membranes are composed of a complex (organized) mixture of lipids, proteins, and carbohydrates. These sheetlike structures form the different compartments within the cell and the boundary that separates living cells from their environment.¹ In the past decade, the usual view of lipids as a mere fluid medium for membrane-bound proteins² is fading out, and a new picture in which lipids have a functional role is emerging.^{3,4} From (complex) lipid “rafts”⁴ in plasma membranes

to (mere) large scale phase separation⁵ in lipid mixtures, this new view is in large part based on the notion of microdomain formation and lateral organization of membrane heterogeneities. Cholesterol is an essential component of biomembranes and plays an important role in the formation of microdomains.⁵ Cholesterol seems to be crucial in the sorting of membrane heterogeneities and specially in the formation of the so-called lipid rafts⁴ in plasma membranes, which are rich in cholesterol and sphingolipids and are thought to act as platforms for adhesion and signaling. Glycolipids, whose headgroups are attached to oligosaccharides, form an important class of lipids that play a fundamental role in cell–cell and cell–matrix interactions. They not only function as specific recognition sites but also form a soft cushion between cells and tissues as a result of their unique swelling behavior, preventing nonspecific adhesion. This indicates a complex interplay between weak generic forces and strong specific forces.⁶ Recent experiments⁶ of synthetic glycolipids with headgroups composed of lactose oligomers at interfaces have shown that these represent quite realistic models for the glycocalyx. Lipid–peptide and peptide–peptide interactions in membrane environments play a fundamental role not only in the folding of natural proteins or association of synthetic peptides but also in the mechanisms of action of toxins and antimicrobial peptides or other peptides with lytic activity, which are lethal to the cells.⁷ Because of the different scales of time and length involved in the processes of insertion and assembly, peptide aggregation in membranes and the formation of pores is an important and challenging problem.

The development of novel algorithms⁸ and the revolutionary advances in computer hardware technology that took place during the last two decades have permitted computer simulations of complex biological membranes to advance at a similar pace as and in connection to experiment in biomedical applications.⁹ Computer simulations provide a unique tool to analyze biomembrane properties from an atomic perspective with a level of detail missing in any other technique. The excellent agreement with experiment obtained in classical molecular dynamics (MD) simulation studies^{10–18} on simple model membranes consisting of lipid bilayers in the biologically relevant fluid lamellar phase have permitted new applications to membrane systems of a considerable degree of complexity. These more complex systems usually consist of the introduction of additives or foreign (bio- or nonbio-) molecules, such as small amphipathic molecules or other solutes, membrane peptides or proteins, or other biopolymers, into a simple model lipid bilayer. Such studies have provided invaluable information about the nature of lipid–lipid,¹⁵ DNA–lipid,¹⁹ peptide–lipid,^{9,20} and peptide–peptide^{1,9} interactions. It would be of great interest in future studies to investigate at a microscopic level the kinds of fundamental problems described in the previous paragraph, in which very different types of forces are

Leonor Saiz was born in Barcelona, Spain in 1969. She received a B.S. degree (1992) in physics from the University of Barcelona (UB). During the period 1992–1993, she was a research assistant at the physics department of the UB and SOCIMAG S. A. focusing on magnetic recording. In 1998, she received a Ph.D. in Physics from the UB, where she worked with Joan A. Padró on computer simulation studies of hydrogen-bonded liquids. She then moved to Manchester, U.K., where she was a postdoctoral fellow at UMIST studying crystal nucleation in liquid alkanes and polymers. Since 1999, she has been a postdoctoral fellow at the University of Pennsylvania, working on computer simulation studies of model biological membranes with Michael L. Klein.

Michael L. Klein was born in London, U.K., on March 13th, 1940. He attended Bristol University, where he received a B.S. in 1961 and a Ph.D. in 1964. He was a CIBA-GEIGY Fellow at the Instituto di Fisica, University of Genoa, Italy from 1964 to 1965 and an ICI Fellow in the Department of Theoretical Chemistry at Bristol University from 1965 to 1967. In 1967, he moved to the United States to accept a position as a research associate in the Department of Physics at Rutgers University. In 1968, he moved to the Chemistry Division of the NRCC in Ottawa, Canada, where he remained for 19 years, rising from Associate Research Officer to Principal Research Officer. In 1987, he returned to the United States to accept his current position as Professor of Chemistry at the University of Pennsylvania, where he was appointed William Smith Professor of Chemistry from 1991 to 1993. Since 1993, he has been Hepburn Professor of Physical Sciences. In addition, Professor Klein has been the Director of the Laboratory for Research on the Structure of Matter since 1993 and Director of the Penn Center for Molecular Modeling since 1994. His current research involves structure and dynamics of disordered molecular systems using computer simulation techniques; systems of interest include aqueous solutions, model membranes, and membrane-bound proteins.

* Electronic mail: leonor@cmm.chem.upenn.edu.

present or diverse lengths and temporal scales are involved. Simplified models of the coarse grain type could be useful for this kind of application and have been successfully applied earlier to polymers and more recently to surfactants.²¹

In coarse grain models, solute and solvent sites are usually represented as a group of atoms that interact in an effective way, reducing in this way the computer time needed for the calculations. Coarse grain models can be adjusted to mimic a specific system instead of a general phenomenon.²² They have been shown to successfully describe collective phenomena occurring at mesoscopic scales,^{22–25} which have only been reached very recently by atomistic simulations^{26,27} after sacrificing an accurate treatment of the long range (electrostatic) forces.

In this Account, we focus on multicomponent membranes with potential applications in biomedical research in which computer simulation, classical MD in particular, has been applied successfully to get new insights into their behavior. Specifically, we will discuss the nature of DNA–lipid complexes that may be relevant in gene therapy, the effect of a pore-forming bundle of peptides on the lipid environment, and the partitioning of anesthetic molecules within model membranes. The possible utility of a simplified model of the coarse grain type to study the effects of anesthetics in phospholipid bilayers is also illustrated. Some other aspects of lipid bilayer simulations are described in the Account by Pastor et al. (this issue).

DNA–Lipid Complexes

DNA–lipid complexes are important biomedical materials because of their potential use as vectors in gene therapy.²⁸ These complexes are effective carriers and deliver DNA to the nucleus of living cells, where damaged or missing genes are replaced. Currently, the most common method of gene delivery is by means of viral carriers of DNA.²⁹ However, the risk involved in the clinical applications of the latter points to lipid–DNA complexes as a potential alternative. Recent developments in the field have selected a few cases (binary mixtures of suitable cationic and neutral lipids) with the adequate properties.³⁰ For instance, mixtures containing the unsaturated neutral DOPC and the cationic DMTAP lipid formed a novel multilayer structure with alternating lipid bilayers and DNA monolayers.^{31,32} There, the hydrated DNA molecules are intercalated between each pair of lipid bilayers in the water region. In each layer, the DNA strands are parallel to one another, forming a two-dimensional smectic phase. Similar structures have been also observed in binary mixtures of the saturated neutral lipid DMPC and the cationic DMTAP with DNA.^{33,34} When one of the lipids has the tendency to adopt a negative curvature at the lipid–water interface, there is a second kind of liquid crystal structure, which consists of an inverted hexagonal phase that performs better for gene therapy purposes than the lamellar case.³⁰

Recent all-atom MD simulations of lipid–DNA complexes containing DMPC and DMTAP lipids¹⁹ have ob-

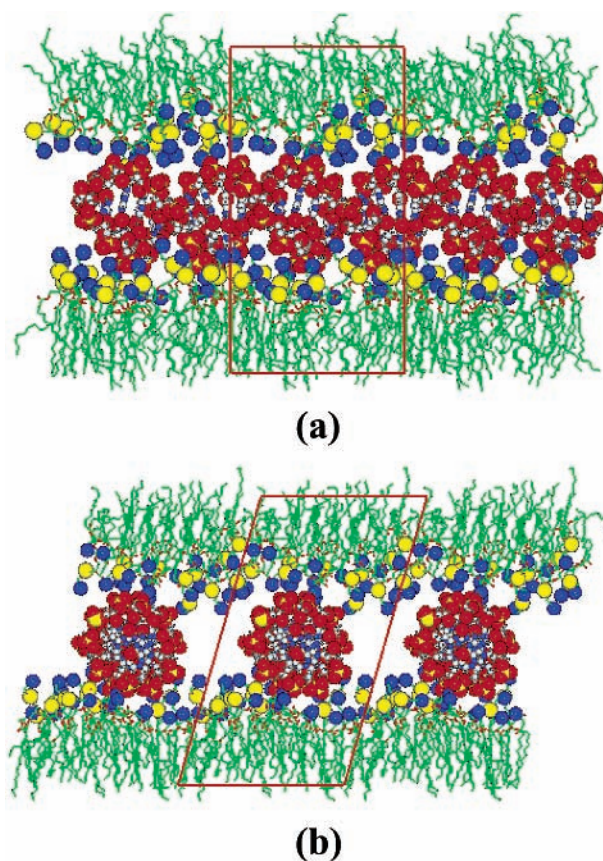


FIGURE 1. Configuration of the DMPC/DMTAP–DNA complex taken from an MD simulation after 5.5 ns.¹⁹ The details of the simulation can be found in ref 19. Two different views are shown: (a) perpendicular to, and (b) along, the DNA axis. The DNA and the lipid headgroup P and N atoms are drawn as spheres, whereas the lipid chain atoms are drawn as sticks. The atom coloring scheme is N, blue; O, red; P, yellow; C (DNA), gray; C (lipid), green; and H, dark gray. For clarity, the water molecules and the lipid H atoms are not shown. The (central) simulation cell is shown in red.

tained interesting insights into the nature of the lipid–DNA interactions and the role of the lipid interface in the formation of these complexes. This simulation was carried out in the fluid lamellar phase of the complex ($T = 50\text{ }^{\circ}\text{C}$) at the isoelectric point (the mixing ratio for which the DNA and lipid charges cancel each other) and contained 24 DMPC, 20 DMTAP, 1003 water molecules, and a DNA duplex $d(\text{CCAACGTTGG})_2$.¹⁹ Unless otherwise stated, all of the atomistic simulations reported herein were performed using the PINY_MD program^{8,35} with the latest all-atom CHARMM force field^{13,36} for the different components of the biomembranes and a rigid TIP3P water model.³⁷ Figure 1 shows the configuration of this system after 5.5 ns. The presence of the DNA intercalated between the two lipid–water interfaces, which were initially nearly flat, produces significant undulations of the membrane surfaces. This indicates a strong interaction between the lipid interface and the DNA. In particular, these calculations revealed that the phosphate groups of the DNA can interact directly or via bridging water molecules, with either the choline group of the neutral DMPC lipids or the cationic lipids with equal probability. The negative charges of the DNA phosphate groups are, thus, screened

by both lipid species. This behavior is due to the electrostatic interactions taking place at the lipid bilayer surface. At the interface, three different kinds of interactions were observed,¹⁹ namely: PC group–PC group via the formation of $P^- \cdots N^+ \cdots P^- \cdots N^+$ charge pairs linking the negatively charged phosphate groups (P^-) and the positively charged choline groups (N^+), typical of pure (PC) lipid bilayers;^{15,38} P^- groups of the DMPC molecules and the N^+ of the TAP groups ($N^+ \cdots P^- \cdots N^+$ charge pairs); and N^+ of both the PC and the TAP groups of the lipids with the anionic DNA phosphates. The interactions between the P^- groups of the DMPC molecules and the N^+ of the TAP groups cause a reorientation of the PC headgroup dipole moments (denoted by $P^- \rightarrow N^+$) away from the lipid–water interface, in agreement with NMR experiments on similar systems,³⁹ thereby pushing the covalently bonded choline groups out of the interface, which then become available to interact with the anionic DNA phosphates.

Pore-Forming Peptides

The incorporation of relatively large membrane proteins, such as those forming pores that function as ion channels, in membranes is expected to modify the physical properties of the lipid bilayer. However, because of the fundamental importance of these proteins, most of the effort of the experimental and theoretical works has been devoted to the study of the effects of the lipid environment on the structure, dynamics, and function of membrane proteins. For instance, recent notable successes of computer simulation in the area of transmembrane channels include the KcsA K^+ channel⁴⁰ and the *Escherichia coli* aquaglyceroporin GlpF,⁴¹ shown at full atomic detail. The difference between the hydrophobic length along the membrane normal of the lipid bilayer and the protein (hydrophobic mismatch) and the membrane curvature have been identified as the key factors for this interplay between structure and function.^{1,9,42}

Recent experiments have been focused on the study of the influence of simple membrane peptides on the membrane properties in an effort to get more insights into the structure and dynamics of model membranes. The effect of incorporating small membrane proteins or model transmembrane peptides into lipid bilayers changes the phase behavior of the water–lipid system^{43,44} and can even promote nonbilayer phases.⁴³ For instance, single transmembrane peptides can decrease the temperature of the phase transition and increase the orientational order of the lipid chains.⁴⁵ Other NMR experiments performed on a mostly hydrophobic cationic peptide showed that the order parameters of the hydrophobic region were significantly modified neither by the peptide in DMPC lipid bilayers nor in DMPC/DMPS (5:1) but that the peptide affects the membrane interface by rotating the headgroup dipoles in a direction away from the membrane plane as a result of the (sign of the) “bound” surface charges.⁴⁶ In these experiments, even though a major influence is expected to occur on the lipids located closer to the peptide, the formation of lipid–protein complexes was

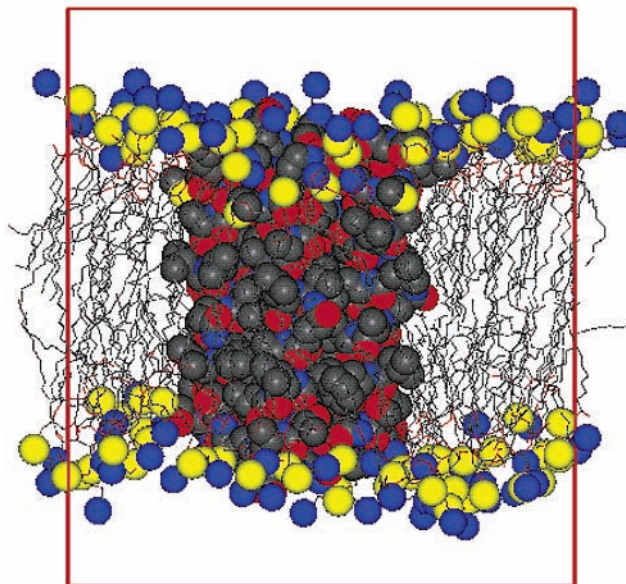


FIGURE 2. Configuration of the 5×M2–DMPC system taken from an MD simulation after 2 ns.⁴⁷ The details of the simulation can be found in ref 47. The lipid molecules are shown as balls and sticks except the N and P atoms of the headgroups, which are displayed as blue and yellow spheres, respectively. For clarity, the water molecules and hydrogen atoms are not shown. The coloring scheme for the M2 helices is N, blue; O, red; C, gray; and S, yellow. The radii of the spheres correspond to the atomic van der Waals radii of the different species. The C-terminus (synaptic or extracellular⁴⁸) is located at the bottom of the bundle.

ruled out at least in the time scale of the experiments ($\approx 10^{-5}$ s).⁴⁶ Systematic NMR and ESR spectroscopy studies using hydrophobic polypeptides of different lengths,⁴³ however, indicate that peptides can induce a hydrophobic mismatch that perturbs the bilayer in a systematic manner. Differences obtained for peptides with different shapes indicate also that the topology of the peptide surface modulates the effect of hydrophobic mismatch.^{43,45}

In Figure 2, we show a snapshot of a recently performed atomistic MD simulation⁴⁷ of the transmembrane homopentameric bundle of the α -helical M2 segments that are thought to form the pore region of the nicotinic acetylcholine receptor (nAChR) ion channel.⁴⁸ The nAChR is the neurotransmitter-gated ion channel responsible for the rapid propagation of electrical signals between cells at the nerve–muscle synapse.⁴⁹ Previous simulation studies on the nAChR were focused exclusively in peptide–peptide interactions of simplified models or the effects of the α -helices environment on the secondary structure.⁵⁰ To investigate the effect of the presence of pore-forming peptides on the properties of the lipid environment, the peptide bundle was embedded in a fully hydrated DMPC lipid bilayer in the fluid lamellar phase, L_{α} , at ambient conditions ($T = 303$ K and $p = 1$ atm) with a lipid/peptide molar ratio of 19:1.⁴⁷ The M2 segments are characterized by the sequence GSEKMSTAISVLLAQAVFLLLSQR and correspond to the δ subunit of the native nAChR ion channel of the *Rattus norvegicus*.⁴⁸ Preliminary results⁵¹ indicate that the main effect of the incorporation of the pentameric bundle into the DMPC lipid bilayer consists

of an increase of the bilayer thickness and the orientational order of the DMPC lipid acyl chains. The orientational distribution of the lipid dipole moments with the membrane normal, which is broader and has a smaller average orientation than that corresponding to the pure DMPC lipid bilayer, indicates a perturbation of the $P^- \rightarrow N^+$ dipoles at the membrane interface. The presence of the peptide bundle renders the otherwise symmetric lipid bilayer asymmetric. Consistently, the behavior of lipids located in the two different leaflets differed. The calculation of the different properties for the lipids as a function of their distance from the bundle center of mass indicated that, on average, the effect is stronger for those lipids located closer to the peptides. For these lipids, for instance, the orientation order parameters are higher. Interestingly, specific lipid–peptide interactions and the formation of lipid–peptide complexes mainly between the Lys⁺ (K) residues located at the external surface of the bundle and the negatively charged lipid phosphate groups have been identified and seem to be responsible for the inhomogeneous effect observed as a function of the distance to the peptides, as compared to the homogeneous effect observed in the other monolayer.

Anesthetic Partitioning into Phospholipid Bilayers

The interaction of small molecules with biological membranes is a basic issue in the understanding of the mechanisms of general anesthesia and, specifically, the search for the site of anesthetic action (lipid bilayer membrane or membrane proteins?), which is still unresolved.^{52,53} Recent computer simulations⁵⁴ on inhaled (volatile) anesthetics (halothane, $CF_3CHBrCl$) at concentrations higher (mol fraction of 50%) than those clinically used to study their effects on the (pure) lipid bilayer membranes have shed some light onto this problem. The most relevant observation is a nonuniform distribution of the anesthetic molecules (initially located in the lipid bilayer hydrophobic region) along the axis perpendicular to the lipid–water interface. This indicates that the halothane molecules are located preferentially in the upper region of the lipid acyl chains, below the carbonyl groups. This has been observed to affect the membrane interior by reducing the order of the lower half of the hydrocarbon chains and increasing the number of gauche defects and the mobility of the lipid chains. The presence of the halothane molecules modified not only the hydrocarbon region of the system, as compared with the pure lipid bilayer membrane, but also the interface by inducing a reorientation of the lipid headgroup $P^- \rightarrow N^+$ dipole moments. In this calculation,⁵⁴ the $P^- \rightarrow N^+$ orientational distribution changed from being almost uniform to a distribution with a most probable orientation of 120° with the membrane normal, -30° with the membrane surface pointing toward the lipid bilayer interior and away from the water phase. This change may alter the electrostatic properties of the membrane interface, such as the membrane potential (the electrostatic potential across the

membrane interface) of the mixed system. Similarly, the area available per lipid molecule increases, and this is accompanied by a concomitant reduction of the spacing between the lamellae. In contrast, when a similar molecule, C_2F_6 , is introduced in a pure lipid bilayer membrane,⁵⁵ the mixed system exhibits a remarkable perturbation of neither the membrane interior nor the interface. In this case, the C_2F_6 molecules are evenly distributed along the hydrocarbon region with a higher probability at the bilayer center. The C_2F_6 compound, which is known to produce two of the key effects of anesthesia (amnesia and analgesia) but it is not an immobilizer, is more hydrophobic and lacks the small dipole moment of halothane.

To illustrate the differences existing between the distributions of both types of molecules in a model membrane, we depict in Figure 3 the configurations of two very recent simulations⁵⁶ of a polyunsaturated SDPC lipid bilayer¹⁸ with halothane and with the nonimmobilizer C_2F_6 after 2 ns. The simulations were carried out at $T = 30^\circ C$ and $p = 1$ atm, where the pure SDPC lipid bilayer is in the fluid lamellar phase,¹⁸ with 16 halothane/ C_2F_6 molecules and 64 SDPC molecules (mol fraction of 25%).⁵⁶ Figure 3a indicates that the halothane molecules in the SDPC lipid bilayer have the tendency to move toward the membrane interface, below the carbonyl region, although there are also some molecules remaining at the membrane center (methyl trough). Conversely, the distribution emerging from Figure 3b shows that the C_2F_6 molecules are more probably located at the membrane interior and closer to the membrane center.

The study of the effect of anesthetics and nonimmobilizer in highly unsaturated lipid bilayers is particularly relevant in biology as a result of the essential role of fatty acids with cis double bonds. Recent simulations on the fully hydrated mixed (saturated/polyunsaturated) chain SDPC lipid bilayer with the highly unsaturated docosahexaenoic fatty acid (DHA) in the biologically relevant fluid lamellar phase¹⁸ provided interesting insights into the origin of the special properties of these complex lipid systems. In these systems, the presence of the polyunsaturated chains, which have an enhanced tendency to visit the lipid–water interface, increases the disorder of the membrane.⁵⁷ This is due to the different intramolecular conformation and dynamics^{18,38,57} of the polyunsaturated chains compared to those of the better-studied saturated chains. In fact, the distinct conformations of the DHA chain have a different (and inhomogeneous) effect on the order parameters of the saturated chains. The most linear conformations of DHA increased the order, whereas the nonlinear conformations decreased the order of the bottom part of the saturated chains.⁵⁷

Coarse Grain Models of Phospholipids

The utility of coarse grain (CG)-type models to study collective phenomena in model membranes is well-established.^{24,25} In early studies, however, the parameters of the model were not fitted to specific systems. Here, we will describe a simplified model of the coarse grain type,²²

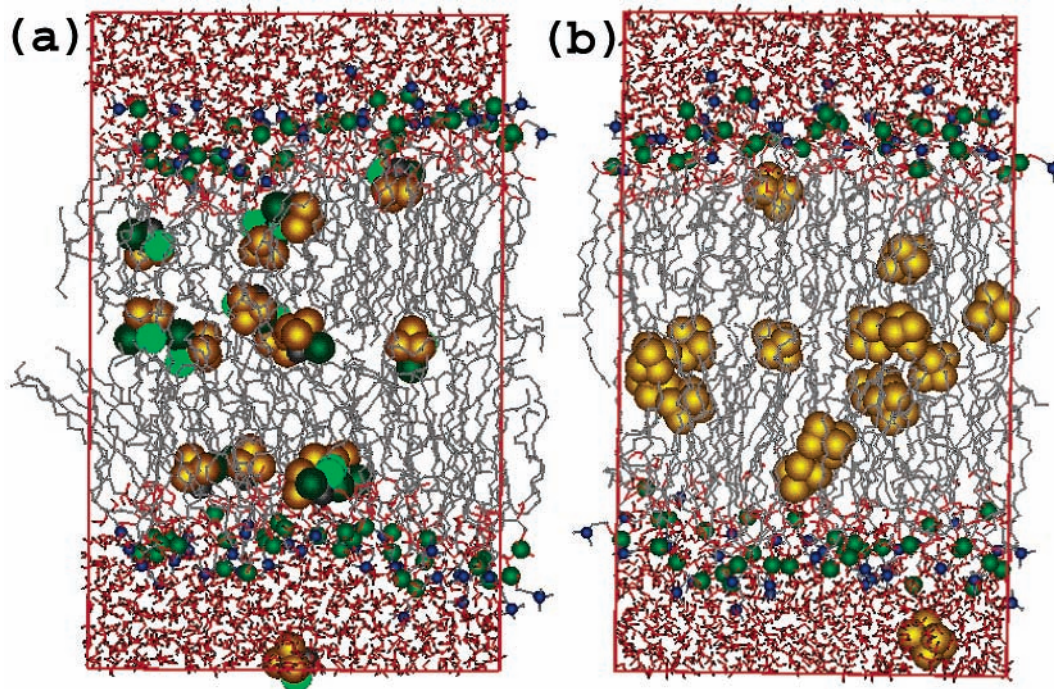


FIGURE 3. Configurations of the halothane—SDPC (a) and C_2F_6 —SDPC (b) systems taken from MD simulations after 2 ns.⁵⁶ The details of the simulations can be found in ref 56. The water and SDPC lipid molecules are shown as balls and sticks except the N and P atoms of the lipid headgroups, which are displayed as blue and green spheres, respectively, to highlight the interface. For clarity, the hydrogen atoms of the lipids are not shown. Halothane and C_2F_6 molecules are depicted with the atomic van der Waals radii (F, yellowish; Cl, light green; Br, dark green; H, gray).

which has been constructed to mimic specific phospholipids and has been successfully applied to model a few complex systems.^{22,23} In this approach, all of the molecular components, including the solvent, are considered explicitly. In the model, the CG waters, represent clusters of three water molecules and are spherically symmetric and interact via a Lennard—Jones potential with the parameters chosen to match a number of the properties of water (i.e., CG water should adopt the correct density, remain liquid within the desired interval, and be consistent with hydrodynamics). The CG model for the DMPC lipids is as follows (see Figure 4). The lipid chains are represented by one site for each triplet of methylene or methyl atoms (SM and ST). Harmonic bond length and bond angle potentials are used to link sites together forming the hydrocarbon chain and to maintain the stiffness and tail length, respectively, and Lennard—Jones interactions are used for the nonbonded interactions. The hydrophilic region of the lipids is built in a similar way, and the main feature is that the beads representing the choline (CH) and the phosphate (PH) groups have a charge of $+e$ and $-e$, respectively, and a dielectric constant of 78 is used for the electrostatic interactions. A single CG site (GL) represents the glycerol backbone (CH_2CHCH_2) that links the two chains and the headgroup together. The number of interaction sites for the CG model of DMPC is thus considerably reduced (by 1 order of magnitude; from 118 atoms to 13 interaction sites), and the number of charged sites in the CG model is two per lipid molecule instead of the 118 charged atoms of the all-atom DMPC. This, in turn, reduces considerably the computer time

consumed in the calculation of the long-range interactions. For all of the components, the different interactions were parametrized on the basis of the corresponding radial distribution functions (RDFs), $g_{\alpha\beta}(r)$, obtained from atomistic simulations. These RDFs were used as the starting point. The method to reconstruct the CG Hamiltonian from the RDFs^{22,58} uses the potential of mean force [$V_{\alpha\beta} \propto -k_B T \ln(g_{\alpha\beta}(r))$] as the initial choice for the effective interaction potentials between the different CG sites. The parameters are then adjusted iteratively until the RDFs or other desired quantities are appropriately matched within the pertinent accuracy. The model constructed in this way has been shown to reproduce semiquantitatively the density profiles of the different components/sites in the direction normal to the interface of a preassembled DMPC lipid bilayer at room temperature, even though it produces pressures that are somewhat too high.²² This CG model also self-assembles into a lipid bilayer system from a random initial configuration.^{22,23,59} The coarse grain simulations were performed using the CM³D program.⁶⁰

To investigate whether this type of model can be used to study more complex biological membranes, the CG model for the DMPC lipid bilayer was generalized to include interactions with a foreign molecule,²³ for example, the anesthetic halothane, at conditions similar to those of the fully atomistic simulations⁵⁴ described in the previous section. The results obtained showed that a careful parametrization and, consequently, a necessity of gathering the adequate data for the full parametrization is vital in order for the CG model to be predictive. The refining of the parameters (basically, the depth of the well

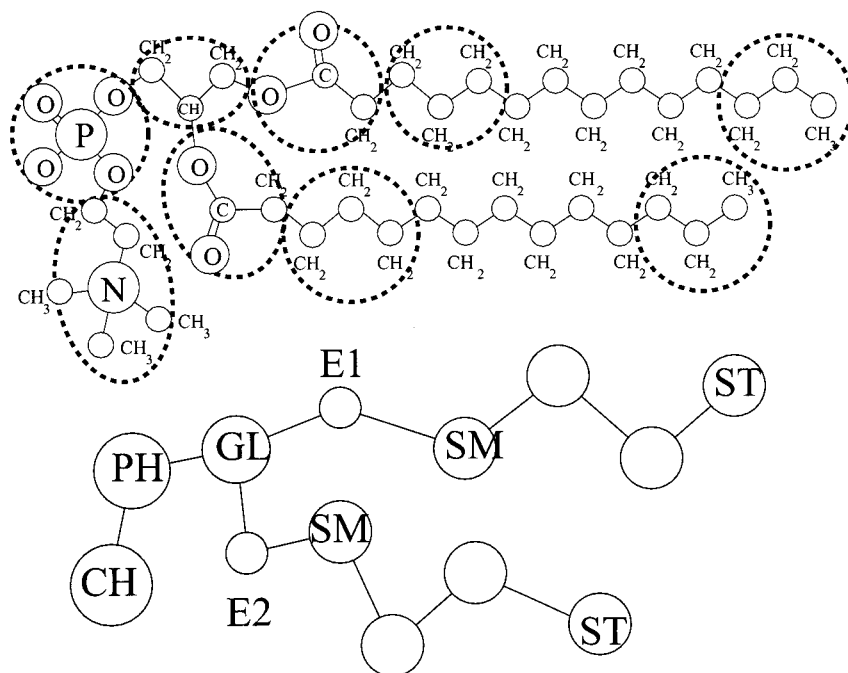


FIGURE 4. Atomistic (top) and coarse grain (bottom) schematic representations of the molecular structure of DMPC. For the sake of clarity, the hydrogen atoms are not explicitly shown in the sketch of the atomistic model.

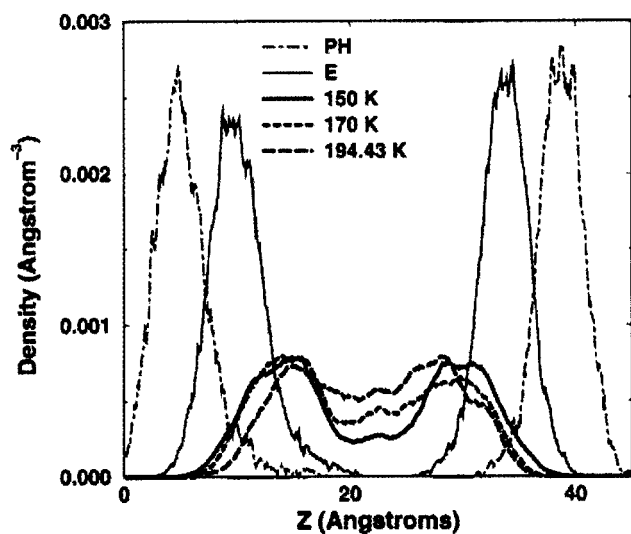


FIGURE 5. Density profiles along the membrane normal for the coarse grain simulation containing halothane.²³ The details of the simulation can be found in ref 23. Curves are given for the phosphate groups (PH), the ester groups (E), and three choices of the H–S interaction parameter ϵ_{SH} ($\epsilon_{SH} = 150, 170, \text{ and } 194.43 \text{ K}$).

of the Lennard–Jones interactions of halothane with the lipid chain groups, ϵ_{SH} , and with the choline group) leads to reasonable agreement with the existing atomistic data, especially concerning the density profiles of the halothane distribution along the membrane normal.²³ In Figure 5, the density profiles for the ester groups (E, O_2CCH_2) and the phosphate groups of the DMPC lipid and that corresponding to the halothane molecules for three values for the ϵ_{SH} parameter are shown for the CG model.²³ The differences obtained in the distributions corresponding to the three values of ϵ_{SH} illustrate the sensitivity of the model to the parameters. The nonuniform distribution for

the halothane molecules obtained in the atomistic study⁵⁴ resembles that of the CG model with the shallowest halothane tail (H–S)–CG interaction potential. It is interesting to point out that by fine-tuning a single parameter of all of the CG interactions of the solute (solute–hydrocarbon chain in this case), one can go from a behavior typical of an anesthetic ($\epsilon_{SH} = 150 \text{ K}$) to that typical of a nonanesthetic molecule ($\epsilon_{SH} = 170, \text{ and } 194.43 \text{ K}$).

Concluding Remarks

We have considered in detail three examples that illustrate the current ability of fully atomistic computer simulations to describe model membranes with potential applications in biomedical research. Here, computer simulation has been successfully applied to get new insights into the properties of multicomponent systems with a considerable degree of complexity. The nature of DNA–lipid interactions, the interplay between functional membrane peptides and their lipid environment, and the effect of foreign molecules on model membranes are timely topics in membrane science. These and related studies have provided invaluable information about the nature of lipid–lipid, peptide–lipid, and peptide–peptide interactions, which have been possible as a result of the full atomistic detail available from this kind of computer simulation.^{40,41}

In most of the cases of biological interest in which particular molecular interactions play a fundamental role, however, very different types of forces are present, or diverse lengths and temporal scales are involved, thus making necessary a complete description covering full atomistic detail up to mesoscopic scales. This is the case, for instance, in processes such as peptide insertion and assembly, peptide aggregation in membranes and pore-

formation, microdomain formation and lateral organization of membrane heterogeneities, and in glycolipid systems, in which weak generic forces and strong specific forces are responsible for the formation of a soft cushion between cells and for recognition, respectively. Even though the revolutionary advances in computer hardware technology and state-of-the-art simulation methodologies of the past decade indicate that longer time and length scales will be progressively achieved, there is still a need for less refined models of biomembrane systems. Simplified models of the coarse grain type, which have been successfully applied to polymers and surfactants, could perhaps be useful for this kind of application. As an example, to illustrate the possible utility of coarse-grain models, we have described the partitioning of the anesthetic halothane in a model phospholipid bilayer. The successes achieved to date suggest that in the next few years, it will be useful to continue to pursue both fully atomistic and coarse-grain approaches to membrane systems.

This work was supported by generous grants from the National Institutes of Health and the National Science Foundation. Computer resources were provided by NPACI centers NCSA, SDSC, and PSC. We thank Alex MacKerell for providing us with the latest versions of the all-atom CHARMM force field. Sanjoy Bandyopadhyay, Laure Koubi, Daphna Scharf, John Shelley, and Mounir Tarek generously shared with us the results of their research on membrane systems.

Note Added after ASAP Posting

The final sentence was inadvertently omitted from the Acknowledgment published with this article in the ASAP version posted on the Web 05/03/02. The corrected version was posted 5/30/02.

References

- Lipowsky, R.; Sackmann, E., Eds. *Structure and dynamics of membranes*. Elsevier: Amsterdam, 1995; Handbook of Biological Physics, Vols. 1A and 1B.
- Singer, S.; Nicholson, G. L. The fluid mosaic model of fluid membranes. *Science* **1972**, *177*, 720.
- See the two recent special issues of *Science* on Lipid Biology and Carbohydrates and glycobiology by various authors. *Science* **2001**, *294*, 1861–1885 and **2001**, *291*, 2337–2376.
- Simons, K.; Ikonen, E. Functional rafts in cell membranes. *Nature* **1997**, *387*, 569–572.
- Brown, D. A.; London, E. Structure and origin of ordered lipid domains in biological membranes. *J. Membr. Biol.* **1998**, *164*, 103–114.
- Schneider, M. F.; Mathe, G.; Tanaka, M.; Gege, C.; Schmidt, R. R. Thermodynamic properties and swelling behavior of glycolipid monolayers at interfaces. *J. Phys. Chem. B* **2001**, *105*, 5178–5185.
- White, S. H.; Wimley, W. C. Membrane protein folding and stability: Physical principles. *Annu. Rev. Biophys. Biomol. Struct.* **1999**, *28*, 319–365.
- Martyna, G. J.; Tuckerman, M. E.; Tobias, D. J.; Klein, M. L. Explicit reversible integrators for extended system dynamics. *Mol. Phys.* **1996**, *87*, 1117–1157.
- Merz, K. M., Jr.; Roux, B., Eds. *Biological membranes: A molecular perspective from computation and experiment*; Birkhauser: Boston, 1996.
- Damodaran, K. V.; Merz, K. M. A comparison of DMPC-based and DLPE-based lipid bilayers. *Biophys. J.* **1994**, *66*, 1076–1087.
- Tu, K.; Tobias, D. J.; Klein, M. L. Constant pressure and temperature molecular dynamics simulation of a fully hydrated liquid crystal phase dipalmitoylphosphatidylcholine bilayer. *Biophys. J.* **1995**, *69*, 2558–2562.
- Tieleman, D. P.; Berendsen, H. J. C. Molecular dynamics simulations of a fully hydrated dipalmitoyl phosphatidylcholine bilayer with different macroscopic boundary conditions and parameters. *J. Chem. Phys.* **1996**, *105*, 4871–4880.
- Feller, S. E.; Yin, D.; Pastor, R. W.; MacKerell, A. D., Jr. Molecular dynamics simulation of unsaturated lipids at low hydration: Parametrization and comparison with diffraction studies. *Biophys. J.* **1997**, *73*, 2269–2279.
- Essman, U.; Berkowitz, M. L. Dynamical properties of phospholipid bilayers from computer simulation. *Biophys. J.* **1999**, *76*, 2081–2089.
- Pasenkiewicz-Gierula, M.; et al. Charge pairing of headgroups in phosphatidylcholine membranes: A molecular dynamics simulation study. *Biophys. J.* **1999**, *76*, 1228–1240.
- Chiu, S. W.; Jakobsson, E.; Subramaniam, S.; Scott, H. L. Combined Monte Carlo and molecular dynamics simulation of fully hydrated dioleoyl and palmitoyl-oleoyl phosphatidylcholine lipid bilayers. *Biophys. J.* **1999**, *77*, 2462–2469.
- Forrest, L. R.; Sansom, M. S. P. Membrane simulations: bigger and better? *Curr. Opin. Struct. Biol.* **2000**, *10*, 174–181.
- Saiz, L.; Klein, M. L. Structural properties of a highly polyunsaturated lipid bilayer from molecular dynamics simulations. *Biophys. J.* **2001**, *81*, 204–216.
- Bandyopadhyay, S.; Tarek, M.; Klein, M. L. Molecular dynamics study of a lipid–DNA complex. *J. Phys. Chem. B* **1999**, *103*, 10075–10080.
- Tieleman, D. P.; Forrest, L. R.; Sansom, M. S. P.; Berendsen, H. J. C. Lipid properties and orientation of aromatic residues in OmpF, influenza M2, and alamethicin systems: Molecular dynamics simulations. *Biochemistry* **1998**, *37*, 17554–17561.
- Shelley, J. C.; Shelley, M. Y. Computer simulation of surfactant solutions. *Curr. Opin. Colloid Interface Sci.* **2000**, *5*, 101–110.
- Shelley, J. C.; et al. A coarse grain model for phospholipid simulations. *J. Phys. Chem. B* **2001**, *105*, 4464–4470.
- Shelley, J. C.; et al. Simulations of phospholipids using a coarse grain model. *J. Phys. Chem. B* **2001**, *105*, 9785–9792.
- Goetz, R.; Gompper, G.; Lipowsky, R. Mobility and elasticity of self-assembled membranes. *Phys. Rev. Lett.* **1999**, *82*, 221–224.
- Venturoli, M.; Smit, B. Simulating the self-assembly of model membranes. *Phys. Chem. Commun.* **1999**, *10*.
- Marrink, S. J.; Lindahl, E.; Edholm, O.; Mark, A. E. Simulation of the spontaneous aggregation of phospholipids into bilayers. *J. Am. Chem. Soc.* **2001**, *123*, 8638–8639.
- Lindahl, E.; Edholm, O. Mesoscopic undulations and thickness fluctuations in lipid bilayers from molecular dynamics simulations. *Biophys. J.* **2000**, *79*, 426–433.
- Felgner, P. L., et al. Lipofection: a highly efficient, lipid-mediated DNA-transfection procedure. *Proc. Natl. Acad. Sci. U.S.A.* **1987**, *84*, 7413–7417.
- Crystal, R. G. Transfer of genes to humans: early lessons and obstacles to success. *Science* **1995**, *270*, 404–410.
- For a review on DNA electrostatics, see: Gelbart, W. M.; Bruinsma, R. F.; Pincus, P. A.; Parsegian, V. A. DNA-Inspired electrostatics. *Phys. Today* **2000**, *53*, 38.
- Rädler, J. O.; Koltover, I.; Salditt, T.; Safinya, C. R. Structure of DNA–cationic liposome complexes: DNA intercalation in multilamellar membranes in distinct interhelical packing regimes. *Science* **1997**, *275*, 810–814.
- Salditt, T.; Koltover, I.; Rädler, J. O.; Safinya, C. R. Two-dimensional smectic ordering of linear DNA chains in self-assembled DNA–cationic liposome mixtures. *Phys. Rev. Lett.* **1997**, *79*, 2582–2585.
- Artzner, F.; Zantl, R.; Rapp, G.; Rädler, J. O. Observation of a rectangular columnar phase in condensed lamellar cationic lipid–DNA complexes. *Phys. Rev. Lett.* **1998**, *81*, 5015–5018.
- Zantl, R.; Artzner, F.; Rapp, G.; Rädler, J. O. Thermotropic structural changes of saturated-cationic-lipid–DNA complexes. *Europhys. Lett.* **1999**, *45*, 90–96.
- Tuckerman, M. E.; et al. Exploiting multiple levels of parallelism in Molecular Dynamics based calculations via modern techniques and software paradigms on distributed memory computers. *Comput. Phys. Commun.* **2000**, *128*, 333–376.
- Schlenkerich, M.; Brickmann, J.; MacKerell, A. D., Jr.; Karplus, M. An empirical potential energy function for phospholipids: Criteria for parameter optimization and applications. In Merz, K. M., Jr., Roux, B., Eds. *Biological membranes: A molecular perspective from computation and experiment*; Birkhauser: Boston, 1996, pp 31–82. Feller, S. E.; MacKerell, A. D., Jr. An improved empirical potential energy function for molecular simulations of phospholipids. *J. Phys. Chem. B* **2000**, *104*, 7510–7515. Foloppe, N.; MacKerell, A. D., Jr. All-atom empirical force field for nucleic acids: 1) Parameter optimization based on small molecule and

- condensed phase macromolecular target data. *J. Comput. Chem.* **2000**, *21*, 86–104. MacKerell, A. D., Jr.; et al. All-atom empirical potential for molecular modeling and dynamics studies of proteins. *J. Phys. Chem. B* **1998**, *102*, 3586–3616.
- (37) Jorgensen, W. L.; et al. Comparison of simple potential functions for simulating liquid water. *J. Chem. Phys.* **1983**, *79*, 926–935.
- (38) Saiz, L.; Klein, M. L. Electrostatic interactions in a neutral model phospholipid bilayer by molecular dynamics simulations. *J. Chem. Phys.* **2002**, *116*, 3052–3057.
- (39) Scherer, P. G.; Seelig, J. Electric charge effects on phospholipid headgroups—Phosphatidylcholine in mixtures with cationic and anionic amphiphiles. *Biochemistry* **1989**, *28*, 7720–7728.
- (40) Berneche, S.; Roux, B. Energetics of ion conduction through the K⁺ channel. *Nature* **2001**, *414*, 73–77.
- (41) Tajkhorshid, E.; et al. Control of the selectivity of the Aquaporin water channel family by Global orientation tuning. *Science* **2002**, *296*, 525–530.
- (42) Andersen, O. S.; et al. Gramicidin channels: Molecular force transducers in lipid bilayers. *Biol. Skr. Dan. Vid. Selsk.* **1998**, *49*, 75–82.
- (43) de Planque, M. R. R.; et al. Influence of lipid/peptide hydrophobic mismatch on the thickness of diacylphosphatidylcholine bilayers. A ²H NMR and ESR study using designed transmembrane α -helical peptides and gramicidin A. *Biochemistry* **1998**, *37*, 9333–9345.
- (44) Zein, M.; Winter, R. Effect of temperature, pressure, and lipid acyl chain length on the structure and phase behaviour of phospholipid–gramicidin bilayers. *Phys. Chem. Chem. Phys.* **2000**, *2*, 4545–4551.
- (45) Paré, C.; et al. Differential scanning calorimetry and ²H nuclear magnetic resonance and Fourier transform infrared spectroscopy studies of the effects of transmembrane α -helical peptides on the organization of phosphocholine bilayers. *Biochim. Biophys. Acta* **2001**, *1511*, 60–73.
- (46) Roux, M.; et al. Conformational changes of phospholipid headgroups induced by a cationic integral membrane peptide as seen by deuterium magnetic resonance. *Biochemistry* **1989**, *28*, 2313–2321.
- (47) Saiz, L.; Klein, M. L. Structure of the pore region of the nicotinic acetylcholine receptor ion channel: A molecular dynamics simulation study (preprint). **2002**.
- (48) Opella, S. J.; et al. Structures of the M2 channel-lining segments from nicotinic acetylcholine and NMDA receptors by NMR spectroscopy. *Nature Struct. Biol.* **1999**, *6*, 374–379.
- (49) Hille, B. *Ionic Channels of Excitable Membranes*; Sinauer: Sunderland, MA, 1992.
- (50) Law, R. J.; et al. Structure and dynamics of the pore-lining helix of the nicotinic receptor: MD simulations in water, lipid bilayers, and transbilayer bundles. *Proteins* **2000**, *39*, 47–55.
- (51) Saiz, L.; Bandyopadhyay, S.; Klein, M. L. Effect of a pore-forming bundle of transmembrane peptides on the physical properties of a lipid bilayer (preprint). **2002**.
- (52) Franks, N. P.; Lieb, W. R. Molecular and cellular mechanisms of general anesthesia. *Nature* **1994**, *367*, 607–614.
- (53) Koblin, D. D.; Chortkoff, B. S.; Laster, M. J.; Eger, E. I.; Halsey, M. J.; Ionescu, P. Polyhalogenated and perfluorinated compounds that disobey the Meyer–Overton hypothesis. *Anesth. Analg.* **1994**, *79*, 1043–1048.
- (54) Koubi, L.; Tarek, M.; Klein, M. L.; Scharf, D. Distribution of Halothane in a Dipalmitoylphosphatidylcholine Bilayer from Molecular Dynamics Calculations. *Biophys. J.* **2000**, *78*, 800–811.
- (55) Koubi, L.; Tarek, M.; Bandyopadhyay, S.; Klein, M. L.; Scharf, D. Membrane structural perturbations caused by anesthetics and nonimmobilizers: A molecular dynamics investigation. *Biophys. J.* **2001**, *81*, 3339–3345.
- (56) Koubi, L.; Tarek, M.; Saiz, L.; Klein, M. L.; Scharf, D. Molecular dynamics simulations of halothane and its nonimmobilizer analogue in a polyunsaturated lipid bilayer (preprint). **2002**.
- (57) Saiz, L.; Klein, M. L. Influence of highly polyunsaturated lipid acyl chains of biomembranes on the NMR order parameters. *J. Am. Chem. Soc.* **2001**, *123*, 7381–7387.
- (58) Lyabartsev, A. P.; Laaksonen, A. Calculation of effective interaction potentials from radial distribution functions: A reverse Monte Carlo approach. *Phys. Rev. E* **1995**, *52*, 3730–3737.
- (59) Lopez, C.; Moore, P. B.; Shelley, J. C.; Shelley, M. Y.; Klein, M. L. Computer simulation studies of biomembranes using a coarse grain model. *Comput. Phys. Comm.* **2002**, in press.
- (60) Moore, P. B.; Klein, M. L. Implementation of a general integration for extended system molecular dynamics. *Technology Report, University of Pennsylvania* **1997**.

AR010167C