

The Scattering Density Profile Model of POPG Bilayers as Determined by Molecular Dynamics Simulations, and Small-Angle Neutron and X-ray Scattering Experiments

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The complex dynamics exhibited by biological membranes are closely correlated to the membrane's structure. Accurate structural data regarding the various membrane components are therefore important in determining specific biomembrane functions. The binding free energy, for example, of Lactoferricin B to mammalian-like membranes (i.e., no net charge) and bacterial-like membranes (i.e., net negative charge) has been predicted from molecular dynamics (MD) simulations. However, for the simulation to make any kind of prediction, an accurate structure of the membrane lipids is needed. Area per lipid is often used as the key parameter when assessing the validity of MD simulations. On the other hand, lipid areas obtained from experiment have used models and are thus model dependent. It has therefore been proposed that a better test for validating MD simulations is to compare them to "raw" experimental data (e.g., in form of scattering form factors). Experimentally obtained scattering form factors then become the basis for the synergy between experiment and simulation, whereby the simulation results guide the development of more realistic models, and in turn, experimental data aid in the development of more accurate MD force fields.

X-ray and neutron scattering are arguably two of the most powerful experimental techniques when it comes to elucidating the structure of biomembranes. At the same time, the two techniques complement each other as they are differentially sensitive to different parts of the lipid bilayer. For example, in the case of X-rays the electron-dense phosphate groups contrast very well with the lower electron density hydrocarbon region. Thus, X-ray data are well suited for the refinement of lipid headgroups and hydrocarbon chains. On the other

hand, the high neutron scattering length density (NSLD) of D₂O (often used in neutron experiments instead of H₂O) permits neutron scattering experiments to accurately determine the total bilayer thickness and, consequently, lipid area when volumetric information is available. In order to address this complementarity, we recently developed a model for calculating scattering density profiles (SDP) whereby the data sets from the two techniques are jointly refined. Thus, by appropriately parsing a lipid molecule and simultaneously analyzing the different experimentally obtained "contrast" data, a more precise structure of the bilayer can be determined.

We combine MD simulations and experiment, both small-angle neutron (SANS) and small-angle X-ray scattering (SAXS), to determine the precise structure of bilayers comprised of 1-palmitoyl-2-oleoyl-*sn*-glycero-3-phosphatidylglycerol (POPG), a lipid commonly encountered in bacterial membranes. Experiment and simulation are used to develop a one-dimensional SDP model suitable for the analysis of experimental data. The joint refinement of such data (i.e. SANS and SAXS) results in the area per lipid that is then used in the fixed-area simulations. In the final step, the direct comparison of simulated-to-experimental data gives rise to the detailed structure of POPG bilayers. From these studies we conclude that POPG's molecular area is $66.0 \pm 1.3 \text{ \AA}^2$, its overall bilayer thickness is $36.7 \pm 0.7 \text{ \AA}$, and its hydrocarbon region thickness is $27.9 \pm 0.6 \text{ \AA}$ - assuming a simulated value of 1203 \AA^3 for the total lipid volume.

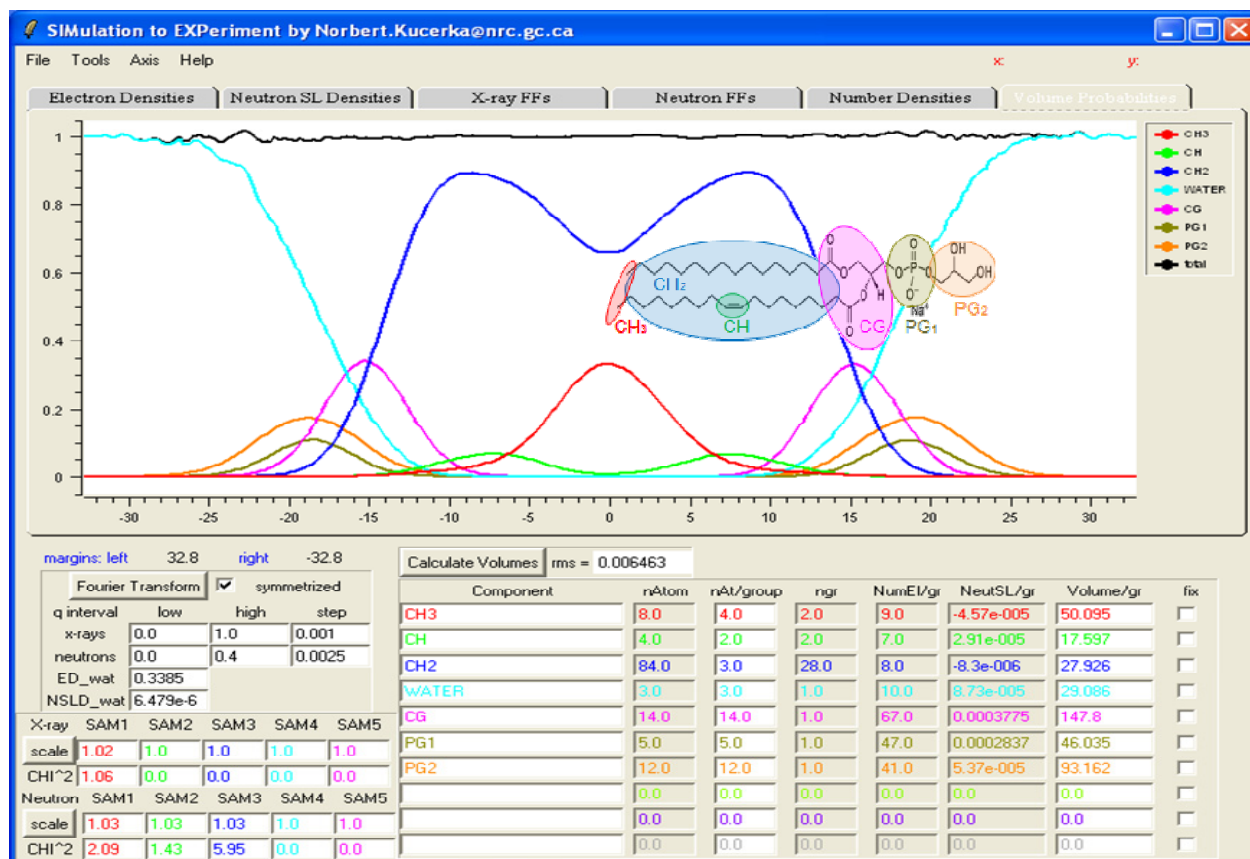


Fig. 1 SIMtoEXP software showing the SDP model parsing of a POPG lipid molecule. Lipid chains are divided into terminal methyl (CH₃), methine (CH) and methylene (CH₂) groups, while the PG headgroup is parsed in carbonyl-glycerol (CG), phosphate (PG1) and glycerol (PG2) moieties.