Quantitative quality of lipid-cholesterol interactions in atomistic resolution molecular dynamics simulations

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The quantitative quality of lipid-cholesterol interactions in atomistic resolution models will be determined against NMR and scattering data.

INTRODUCTION

Details of intermolecular interactions determine the phase behaviour and details lateral organization of lipid and bilayers containing cholesterol [**?**]. Formation lateral heterogeneities [**?**], lipid rafts [**?**] and superlattices [**?**] in cellular membranes have been suggested to be driven by interactions between cholesterol and lipids. While detailed experimental information of these interactions is relatively sparse, atomistic resolution molecular dynamics simulations have been widely applied to give detailed explanations of lipid bilayer lateral organization and lipid-cholesterol interactions [**?**].

Simulations qualitatively reproduce the cholesterol condensation effect, but quantitatve comparison to robust experimental data has not been typically done. This is partly due to the lack of availability of systematic experimental data sets for the comparison and partly due to the lack of protocols for validating intermolecular interactions in lipid bilayer simulations against experiments. Scattering experiments are more difficult to interpret for mixed lipid bilayers than for single component bilayers [**?**], thus area per molecule values, one of the main quantity used to compare simulations to experiments, have not been available for lipid cholesterol mixtures. Systematic experimental data set for lipid C-H bond order parameters with different cholesterol concentrations in POPC bilayer has been published only relatively recently [1].

In this work we present also the experimental scattering form factor data for POPC-cholesterol mixtures by systematically increasing the cholesterol concentration. Our goal is to show that the combination of systematically measured C-H bond order parameter and scattering form factor data can be used to validate the quality of lipid-cholesterol intermolecular interactions MD simulations. MD simulations can be also potentially used to give structural interpretation for form factor measured from mixed systems, which is a major challenge for current methods [**?**]. The approach should be also applicable for mixed lipid bilayers with other than lipid-cholesterol mixtures.

METHODS

X-ray scattering experiments

SAXS data on POPC multilamellar vesicles (MLVs) at various cholesterol concentrations has been measured. Data have been obtained at the EMBL BioSAXS beamline (Hamburg) using 20 keV photons, T = 27C. Data were analyzed in terms of the SDP-GAP model described in Heftberger et al., J. Appl. Cryst. 2013 and Heftberger et al. Biophys. J. 2015. Data from MLVs are a convolute of structure factor (the crystalline lattice) and form factor. By fitting the scattered intensity data we obtain both contributions. Here we posted only form factors (ASCII format). For information on the quality of the fit we also give plots of the fitted intensity data. The electron density profile has been modelled in terms of the SDP model (see papers by Kucerka and coworkers), that is volume distribution functions are modelled by individual Gaussians or error functions. Cholesterol is also accounted for by two Gaussians. This model has been proposed by Jianjun Pan (USF, Tampa, FL), but is to the best of our knowledge not published (see also PhD Thesis by Peter Heftberger). Additional figures show the volume distribution functions and the resulting electron density profiles.

Authors to consult and potentially include in publications using this data: Peter Heftberger (peter.heftberger@gmx.at), Georg Pabst (georg.pabst@uni-graz.at)

Molecular dynamics simulations

COMPARISON OF ACYL CHAIN ORDER PARAMETERS BETWEEN EXPERIMENTS AND SIMULATIONS

Order parameters from simulations and experiments for acyl chains of 1-palmitoyl-2-oleoylphosphatidylcholine (POPC) are shown in Fig. 1.

The order parameter changes as a function of cholesterol for each segment are shown in Fig. 2 (currently only sn-1).

TABLE I: Simulated lipid bilayers containing cholesterol. The simulation file data sets marked with * include also part of the trajectory. [a] The number of lipid molecules [b] The number of cholesterol molecules [c] Cholesterol concentration (mol%) [d] The number of water molecules [e] Simulation temperature [f] The total simulation time [g] Time frames used in the analysis [h] Reference link for the downloadable simulation files [i] Reference for the full simulation details

Force field	lipid	[a]N_l	[b]N_{chol}	[c]C_{CHOL}	[d]N_w	[e]T (K)	[f]t_{sim}(ns)	[g]t_{anal} (ns)	[h]Files	[i]Details
Berger-POPC-07 [2]	POPC	128	0	0%	7290	298	270	240	[3]*	[4]
/Höltje-CHOL-13 [1, 5]										
	POPC	120	8	6%	7290	298	100	80	[6]*	[1]
	POPC	110	18	14%	8481	298	100	80	[7]*	[1]
	POPC	84	44	34%	6794	298	100	80	[8]*	[1]
	POPC	64	64	50%	10314	298	100	80	[9]*	[1]
	POPC	50	78	61%	5782	298	100	80	[10]*	[1]
CHARMM36[11, 12]	POPC	200	0	0%	9000	310	?	100	[13]*	SI
	POPC	200	22	10%	9000	310	?	100	[13]*	SI
	POPC	200	50	20%	9000	310	?	100	[13]*	SI
	POPC	200	86	30%	9000	310	?	100	[13]*	SI
	POPC	200	134	40%	15030	310	109	100	[14]*	SI
	POPC	200	200	50%	18000	310	109	100	[14]*	SI
Slipids[15–17]	POPC	200	0	0%	?	310	?	100	[18]*	SI
	POPC	512	0	0%	23943	310	170	100	[19]*	SI
	POPC	200	22	10%	?	310	?	100	[18]*	SI
	POPC	200	50	20%	?	310	?	100	[18]*	SI
	POPC	200	86	30%	?	310	?	100	[18]*	SI
	POPC	358	154	30%	21183	298	170	100	[20]*	SI
	POPC	200	134	40%	?	310	109	100	[21]*	SI
	POPC	200	200	50%	?	310	109	100	[21]*	SI
	POPC	256	256	50%	20334	298	170	100	[22]*	SI
MacRog[23]	POPC	128	0	0%	6400	310	400	200	[24]*	[25]
	POPC	114	14	11%	6400	310	400	200	[24]*	[25]
	POPC	72	56	44%	6400	310	400	200	[24]*	[25]
	POPC	64	64	50%	6400	310	400	200	[24]*	[25]
	POPC	56	72	56%	6400	310	400	200	[24]*	[25]

COMPARISON OF FORM FACTORS BETWEEN EXPERIMENTS AND SIMULATIONS

The form factors calculated from different simulations with different cholesterol content are shown in Fig. 3.

CONCLUSIONS

SUPPLEMENTARY INFORMATION

CHARMM36 results from different simulation packages

The results from CHARMM36 model for lipid bilayers from different simulation packages have been reported to give different results in the literature [26, 27]. The results are mainly dependent on different Lennart-Jones cut–off settings, but all the details are not quite understood. In this work we use the results from Gromacs 5 with settings suggested to be optimal by Gromacs webpage. We also compared the results from Gromacs 5 with these settings to the results simulated with NAMD, OpenMM and literature values. The comparison is shown in Fig. 4

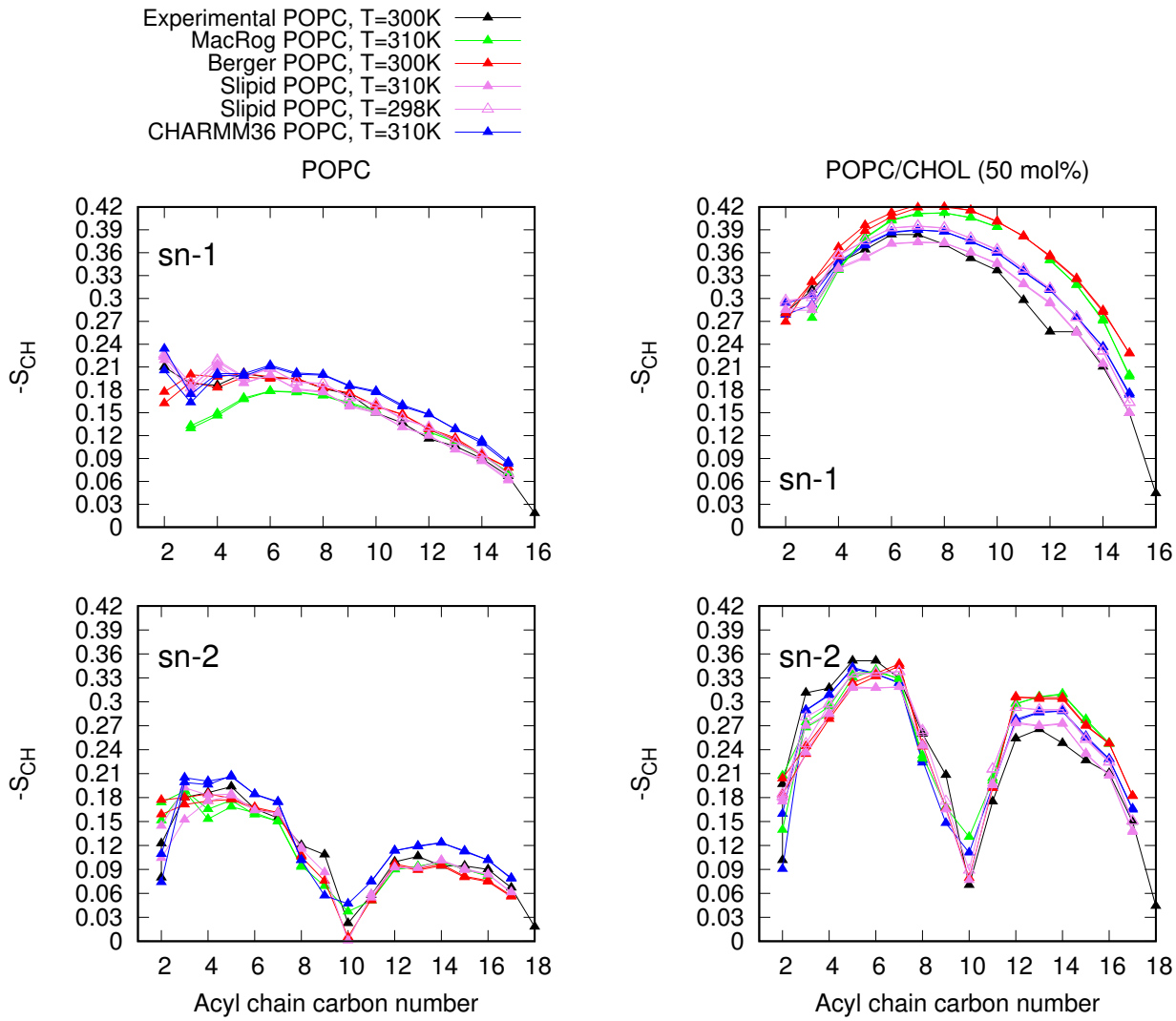

FIG. 1: Order parameters from simulations and experiments for acyl chains of 1-palmitoyl-2-oleoylphosphatidylcholine (POPC).

1.Why are the order parameters of CHARMM36 too large compared to simulations even without cholesterol? Discussion in https://github.com/NMRLipids/NmrLipidsCholXray/issues/4

2.Why there is decrease in order parameters towards beginning of acyl chain in MacRog model without cholesterol?

3.Do the results suggests that condensation effect is too strong in MacRog and Berger models? Discussion in https://github.com/NMRLipids/NmrLipidsCholXray/issues/5

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[1] T. M. Ferreira, F. Coreta-Gomes, O. H. S. Ollila, M. J. Moreno, W. L. C. Vaz, and D. Topgaard, Phys. Chem. Chem. Phys. **15**, 1976 (2013).

[2] S. Ollila, M. T. Hyvönen, and I. Vattulainen, J. Phys. Chem. B **111**, 3139 (2007).

[3] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC bilayer (Berger model delivered by Tieleman, Gromacs 4.5)* (2014), URL `http://dx.doi.org/10.5281/zenodo.13279`.

[4] T. M. Ferreira, O. H. S. Ollila, R. Pigliapochi, A. P. Dabkowska, and D. Topgaard, J. Chem. Phys. **142**, 044905 (2015).

[5] M. Höltje, T. Förster, B. Brandt, T. Engels, W. von Rybinski, and H.-D. Höltje, Biochim. Biophys. Acta **1511**, 156 (2001).

[6] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC/cholesterol (7 mol%) bilayer (Berger model delivered by Tieleman, modified Höltje, Gromacs 4.5)* (2014), URL `{http://dx.doi.org/10.5281/zenodo.13282}`.

[7] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC/cholesterol (15 mol%) bilayer (Berger model delivered by Tieleman, modified Höltje, Gromacs 4.5)* (2014), URL `{http://dx.doi.org/10.5281/zenodo.13281}`.

[8] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC/cholesterol (34 mol%) bi-*

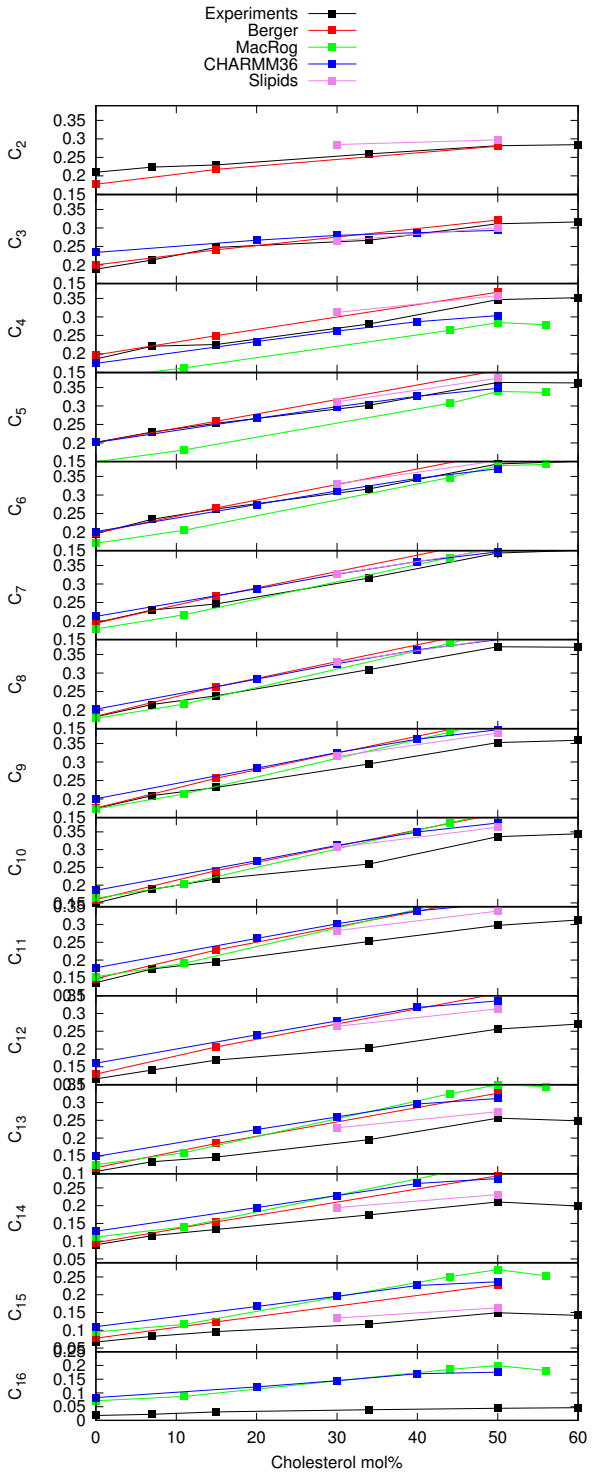

FIG. 2: Order parameter changes from simulations and experiments for each segment in sn-1 chain of 1-palmitoyl-2-oleoylphosphatidylcholine (POPC) as a function of cholesterol concentration.

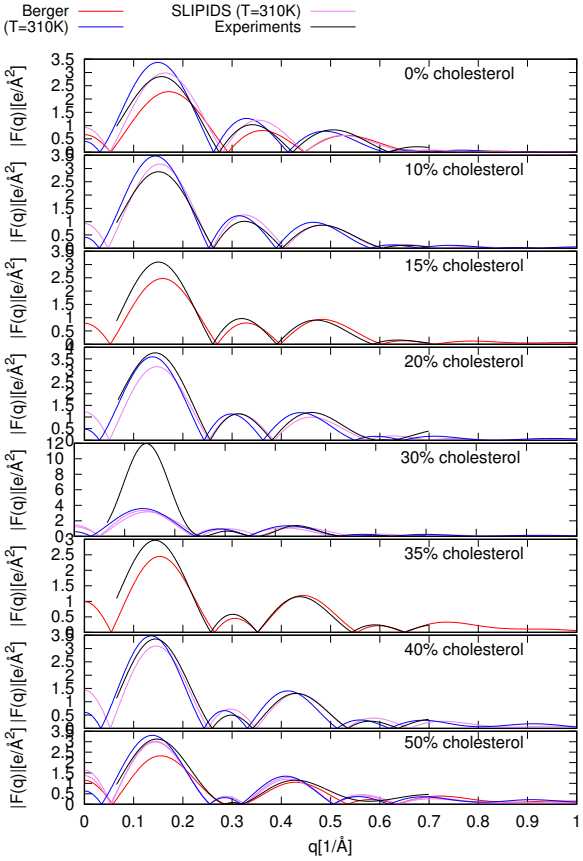

FIG. 3: Form factors from simulations and experiments.

4.Form factor calculation method should be double checked.

5.Experimental form factor amplitudes are not scaled to match with simulations, as done usually

6.Not all experimental and simulation data is here.

layer (Berger model delivered by Tieleman, modified Höltje, Gromacs 4.5) (2014), URL {http://dx.doi.org/10.5281/zenodo.13283}.

[9] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC/cholesterol (50 mol%) bilayer (Berger model delivered by Tieleman, modified Höltje, Gromacs 4.5)* (2014), URL {http://dx.doi.org/10.5281/zenodo.13285}.

[10] O. H. S. Ollila, T. Ferreira, and D. Topgaard, *MD simulation trajectory and related files for POPC/cholesterol (60 mol%) bilayer (Berger model delivered by Tieleman, modified Höltje, Gromacs 4.5)* (2014), URL {http://dx.doi.org/10.5281/zenodo.13286}.

[11] J. B. Klauda, R. M. Venable, J. A. Freites, J. W. O'Connor, D. J. Tobias, C. Mondragon-Ramirez, I. Vorobyov, A. D. MacKerell Jr, and R. W. Pastor, J. Phys. Chem. B **114**, 7830 (2010).

[12] J. B. Lim, B. Rogaski, and J. B. Klauda, J. Phys. Chem. B **116**, 203 (2012).

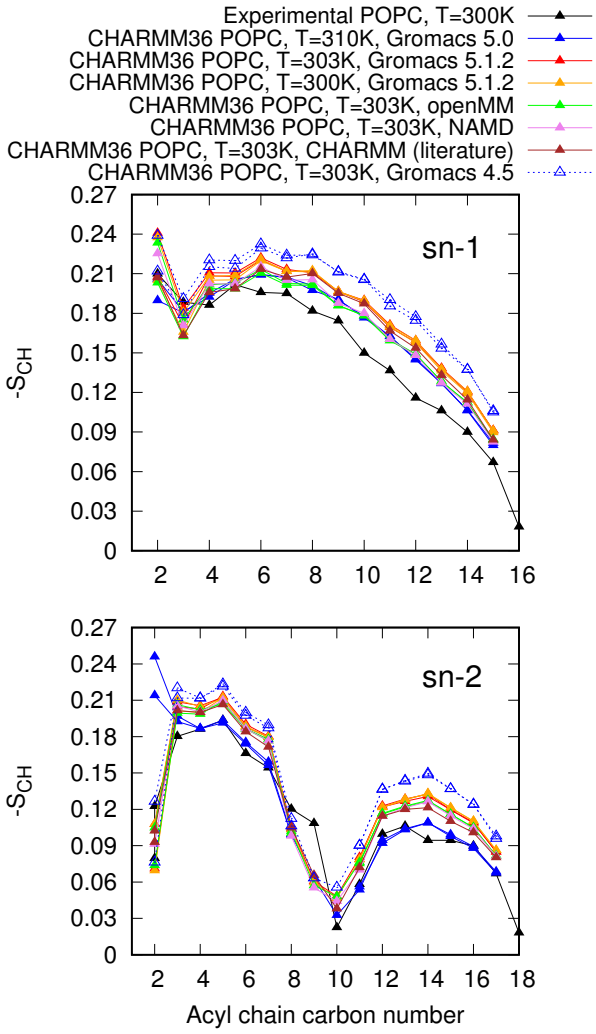

FIG. 4: Results for CHARMM36 model [11] from different simulation packages. Discussion going on at *https://github.com/NMRLipids/NmrLipidsCholXray/issues/4*.

[13] M. Javanainen, *POPC with 0, 10, 20, and 30 mol-310 K. Charmm36 force field.* (2016), URL `https://doi.org/10.5281/zenodo.159759`.

[14] M. Javanainen, *POPC with 40 and 50 mol-Slipids force field.* (2016), URL `https://doi.org/10.5281/zenodo.154346`.

[15] J. P. M. Jämbeck and A. P. Lyubartsev, J. Chem. Theory Comput. **8**, 2938 (2012).

[16] J. P. M. Jämbeck and A. P. Lyubartsev, J. Phys. Chem. B **116**, 3164 (2012).

[17] J. P. M. Jmbeck and A. P. Lyubartsev, Journal of Chemical Theory and Computation **9**, 774 (2013).

[18] M. Javanainen, *POPC with 0, 10, 20, and 30 mol-310 K. Slipids force field.* (2016), URL `https://doi.org/10.5281/zenodo.60607`.

[19] F. Favela-Rosales, *MD simulation trajectory of a lipid bilayer: Pure POPC in water. SLIPIDS, Gromacs 4.6.3. 2016.* (2016), URL `https://doi.org/10.5281/zenodo.166034`.

[20] F. Favela-Rosales, *MD simulation trajectory of a lipid bilayer: 70/30 mol% POPC/Cholesterol . SLIPIDS, Gromacs 4.6.3. 2016.* (2016), URL `https://doi.org/10.5281/zenodo.62026`.

[21] M. Javanainen, *POPC with 40 and 50 mol-Slipids force field.* (2016), URL `https://doi.org/10.5281/zenodo.154346`.

[22] F. Favela-Rosales, *MD simulation trajectory of a lipid bilayer: 50/50 mol% POPC/Cholesterol . SLIPIDS, Gromacs 4.6.3. 2016.* (2016), URL `https://doi.org/10.5281/zenodo.159434`.

[23] W. Kulig, M. Pasenkiewicz-Gierula, and T. Róg, Chem. Phys. Lipids **195**, 12 (2016).

[24] M. Javanainen, *POPC/Cholesterol @ 310K. 0, 10, 40, 50 and 60 mol-cholesterol. Model by Maciejewski and Róg* (2015), URL `{http://dx.doi.org/10.5281/zenodo.13877}`.

[25] A. Botan, F. Favela-Rosales, P. F. J. Fuchs, M. Javanainen, M. Kanduč, W. Kulig, A. Lamberg, C. Loison, A. Lyubartsev, M. S. Miettinen, et al., J. Phys. Chem. B **119**, 15075 (2015).

[26] T. J. Piggot, Á. Piñeiro, and S. Khalid, J. Chem. Theory Comput. **8**, 4593 (2012).

[27] J. Lee, X. Cheng, J. M. Swails, M. S. Yeom, P. K. Eastman, J. A. Lemkul, S. Wei, J. Buckner, J. C. Jeong, Y. Qi, et al., Journal of Chemical Theory and Computation **12**, 405 (2016).

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