Supporting Information for

Domain Stability in Biomimetic Membranes Driven by Lipid Tail Unsaturation

Xubo Lin[a], Joseph H. Lorent[a], Allison D. Skinkle[a], Kandice R. Levental[a], M. Neal Waxham[b], Alemayehu A Gorfe[a,]*, and Ilya Levental[a,]*

a. Department of Integrative Biology and Pharmacology, McGovern Medical School, The University of Texas Health Science Center at Houston, Texas 77030, USA

b. Department of Neurobiology and Anatomy, McGovern Medical School, The University of Texas Health Science Center at Houston, Texas 77030, USA

E-mail: ilya.levental@uth.tmc.edu (IL), alemayehu.g.abebe@uth.tmc.edu (AAG).

Tel: +1-713-500-5566 (IL), +1-713-500-7538 (AAG)

Methods for Additional Analysis:

Radial Pair Distribution Function. The 2D radial pair distribution function $g_{2D}(r)$ is defined as

$$g_{2D}(r) = \lim_{dr \to 0} \frac{p(r)}{\frac{N}{A} 2\pi r dr}$$

where r is the (in-plane) distance between centers of mass of two lipids, $p(r)$ is the average number of pairs found at a distance between r and $r + dr$, A is the area of the system in the xy-plane, and N is the number of unique pairs: $N = n \cdot (n-1)$ for n identical particles, and $N = n1 \cdot n2$ for $n1$ type 1 lipids and $n2$ type 2 lipids.[1]

Diffusion Coefficient. Diffusion coefficient D is calculated based on the Einstein relation:

$$MSD(t) = \langle \vec{r_i}(t) - \vec{r_i}(0) \rangle = nDt$$

where $\vec{r_i}(t)$ is position of molecule i at time t, $\langle \vec{r_i}(t) - \vec{r_i}(0) \rangle$ is the mean square displacement (MSD), and n is the numerical constant which depends on dimensionality: $n = 2, 4, 6$, for 1, 2, or 3 dimensional diffusion. The diffusion coefficient D is calculated by least squares fitting a straight line through the $MSD(t)$. In our simulations, ten 400 ns blocks of the last 4 µs trajectories of 20 µs simulations were used for calculating diffusion coefficient. For the fluid phase, lipid diffusion coefficient is expected to be larger than $10^{-8} cm^2/s$.[2]

Domain Thickness. PO4-type beads (colored in tan, Fig. S1) of saturated/unsaturated lipids are used for bilayer thickness calculation. In order to avoid the effects of membrane undulation on the bilayer thickness calculation, we divided the whole membrane into 14×14 small patches ($\sim 1.3nm \times 1.3nm$), calculated the local thickness of each patch, and then averaged over all patches. For certain system, there is no obvious domain formation. Hence, we re-phrase this thickness as "*head-to-head distance along z axis*" in **Fig. S2** and **Fig. S3**.

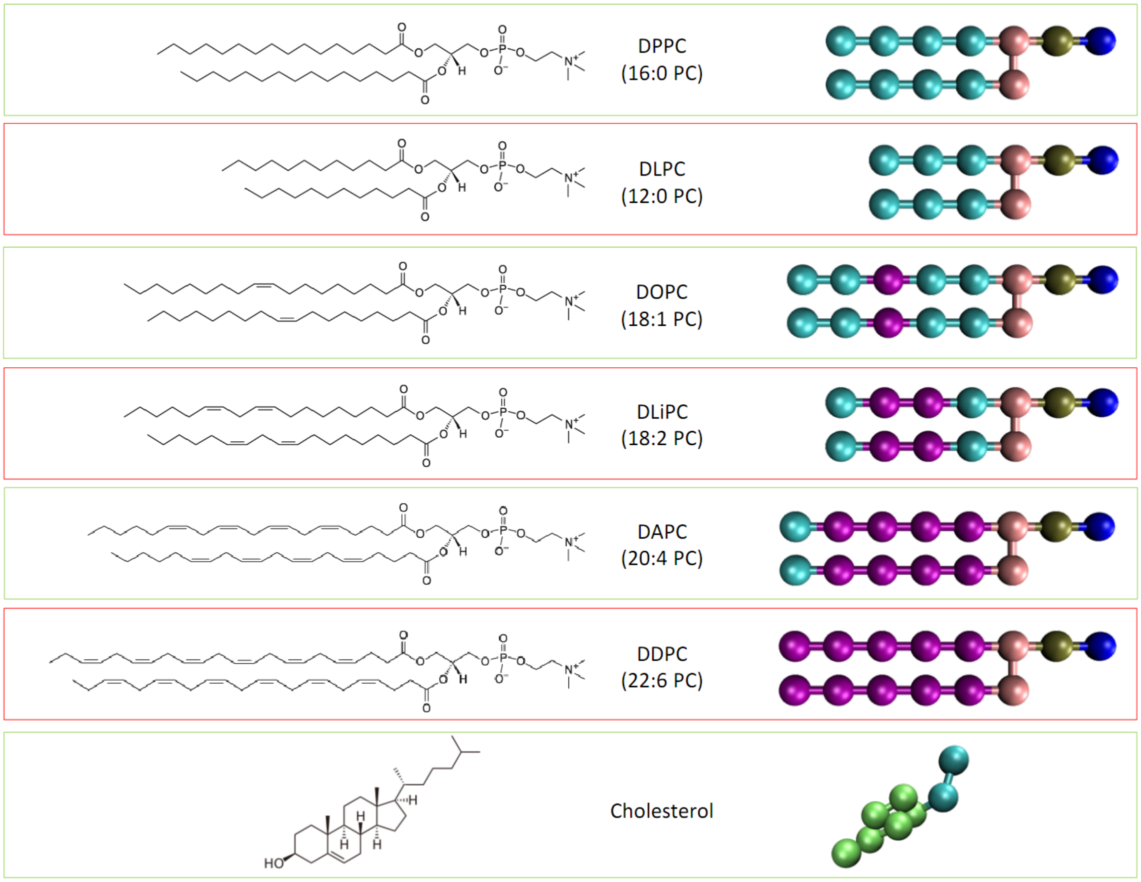

Figure S1. Chemical structure (left) and Martini-based configuration (right) of lipids studied in the current work.

Table S1. Detailed synaptic membrane lipidome

Lipid Sub-species	Headgroup Class	Length	Double Bonds	Mean ± s.d. (mol%)
Cer-36:1;2	Cer	36	1	0.314±0.044
Cer-36:2;2	Cer	36	2	0.071±0.006
Cer-37:2;2	Cer	37	2	0.003±0.001
Cer-38:1;2	Cer	38	1	0.032±0.006
Cer-38:2;2	Cer	38	2	0.005±0.000
Cer-39:1;2	Cer	39	1	0.001±0.001
Cer-40:1;2	Cer	40	1	0.003±0.001
Cer-41:1;2	Cer	41	1	0.003±0.001
Cer-42:1;2	Cer	42	1	0.002±0.001
Cer-42:2;2	Cer	42	2	0.003±0.000
chol	ST	0	0	33.181±0.959
CL-68:2;0(16:0;0-18:1;0-16:0;0-18:1;0)	CL	68	2	0.004±0.005
CL-68:4;0(16:1;0-18:1;0-16:1;0-18:1;0)	CL	68	4	0.020±0.028
CL-70:3;0(16:1;0-18:0;0-18:2;0-18:0;0)	CL	70	3	0.002±0.003

CL-70:4;0(16:1;0-18:1;0-18:1;0-18:1;0)	CL	70	4	0.008±0.011
CL-70:5;0(16:1;0-18:1;0-18:2;0-18:1;0)	CL	70	5	0.008±0.011
CL-72:4;0(18:1;0-18:1;0-18:1;0-18:1;0)	CL	72	4	0.017±0.024
CL-72:6;0(18:2;0-18:2;0-18:2;0-18:0;0)	CL	72	6	0.016±0.022
CL-72:7;0(18:2;0-18:2;0-18:2;0-18:1;0)	CL	72	7	0.019±0.027
CL-72:8;0(16:1;0-20:4;0-18:2;0-18:1;0)	CL	72	8	0.011±0.016
CL-74:10;0(16:1;0-20:4;0-20:4;0-18:1;0)	CL	74	10	0.011±0.015
CL-74:11;0(16:1;0-20:4;0-18:2;0-20:4;0)	CL	74	11	0.008±0.011
CL-74:8;0(16:0;0-20:4;0-20:4;0-18:0;0)	CL	74	8	0.035±0.049
CL-74:9;0(18:2;0-20:4;0-18:2;0-18:1;0)	CL	74	9	0.018±0.025
CL-76:10;0(18:2;0-20:4;0-20:4;0-18:0;0)	CL	76	10	0.052±0.030
CL-76:11;0(18:2;0-20:4;0-20:4;0-18:1;0)	CL	76	11	0.056±0.005
CL-76:9;0(16:0;0-20:3;0-18:2;0-22:4;0)	CL	76	9	0.017±0.024
CL-78:13;0(20:4;0-20:4;0-20:4;0-18:1;0)	CL	78	13	0.013±0.018
CL-78:14;0(18:2;0-20:4;0-20:4;0-20:4;0)	CL	78	14	0.007±0.010
CL-80:15;0(18:2;0-22:6;0-22:6;0-18:1;0)	CL	80	15	0.009±0.012
DAG-33:0;0(17:0;0-16:0;0)	DAG	33	0	0.015±0.022
DAG-34:1;0(18:1;0-16:0;0)	DAG	34	1	0.070±0.008
DAG-35:0;0(18:0;0-17:0;0)	DAG	35	0	0.008±0.012
DAG-36:1;0(18:0;0-18:1;0)	DAG	36	1	0.021±0.004
DAG-36:2;0(18:0;0-18:2;0)	DAG	36	2	0.001±0.001
DAG-36:2;0(18:1;0-18:1;0)	DAG	36	2	0.006±0.008
DAG-36:4;0(20:4;0-16:0;0)	DAG	36	4	0.065±0.004
DAG-38:4;0(18:0;0-20:4;0)	DAG	38	4	0.380±0.023
DAG-38:5;0(18:1;0-20:4;0)	DAG	38	5	0.014±0.020
DAG-38:6;0(22:6;0-16:0;0)	DAG	38	6	0.011±0.015
DAG-40:6;0(18:0;0-22:6;0)	DAG	40	6	0.010±0.014
HexCer-36:1;2	HexCer	36	1	0.018±0.001
HexCer-40:1;2	HexCer	40	1	0.005±0.007
HexCer-40:1;3	HexCer	40	1	0.046±0.005
HexCer-41:1;3	HexCer	41	1	0.021±0.003
HexCer-42:1;2	HexCer	42	1	0.011±0.015
HexCer-42:1;3	HexCer	42	1	0.117±0.012
HexCer-42:2;2	HexCer	42	2	0.030±0.001
HexCer-42:2;3	HexCer	42	2	0.055±0.001
LPA-16:0;0	LPA	16	0	0.001±0.002
LPC O--12:1;0(12:1;0)	LPC O-	12	1	0.018±0.007
LPC O--14:2;0(14:2;0)	LPC O-	14	2	0.072±0.028
LPC O--16:2;0(16:2;0)	LPC O-	16	2	0.006±0.002

LPC-14:1;0(14:1;0)	LPC	14	1	0.054±0.023
LPC-14:2;0(14:2;0)	LPC	14	2	0.007±0.001
LPC-15:0;0(15:0;0)	LPC	15	0	0.007±0.002
LPC-15:2;0(15:2;0)	LPC	15	2	0.004±0.005
LPC-15:3;0(15:3;0)	LPC	15	3	0.002±0.002
LPC-16:0;0(16:0;0)	LPC	16	0	0.388±0.063
LPC-16:1;0(16:1;0)	LPC	16	1	0.002±0.003
LPC-17:6;0(17:6;0)	LPC	17	6	0.009±0.004
LPC-18:0;0(18:0;0)	LPC	18	0	0.128±0.027
LPC-18:1;0(18:1;0)	LPC	18	1	0.152±0.038
LPC-18:2;0(18:2;0)	LPC	18	2	0.008±0.003
LPC-19:2;0(19:2;0)	LPC	19	2	0.001±0.002
LPC-20:1;0(20:1;0)	LPC	20	1	0.006±0.001
LPC-20:3;0(20:3;0)	LPC	20	3	0.001±0.002
LPC-20:4;0(20:4;0)	LPC	20	4	0.037±0.008
LPC-22:4;0(22:4;0)	LPC	22	4	0.007±0.002
LPC-22:6;0(22:6;0)	LPC	22	6	0.031±0.005
LPE O--16:0;0(16:0;0)	LPE O-	16	0	0.001±0.001
LPE O--16:1;0(16:1;0)	LPE O-	16	1	0.021±0.005
LPE O--18:0;0(18:0;0)	LPE O-	18	0	0.001±0.001
LPE O--18:1;0(18:1;0)	LPE O-	18	1	0.037±0.008
LPE O--18:2;0(18:2;0)	LPE O-	18	2	0.011±0.002
LPE-14:1;0(14:1;0)	LPE	14	1	0.014±0.003
LPE-16:0;0(16:0;0)	LPE	16	0	0.041±0.009
LPE-18:0;0(18:0;0)	LPE	18	0	0.164±0.041
LPE-18:1;0(18:1;0)	LPE	18	1	0.040±0.009
LPE-18:2;0(18:2;0)	LPE	18	2	0.001±0.001
LPE-20:1;0(20:1;0)	LPE	20	1	0.000±0.001
LPE-20:4;0(20:4;0)	LPE	20	4	0.040±0.010
LPE-22:3;0(22:3;0)	LPE	22	3	0.002±0.001
LPE-22:4;0(22:4;0)	LPE	22	4	0.012±0.004
LPE-22:5;0(22:5;0)	LPE	22	5	0.005±0.002
LPE-22:6;0(22:6;0)	LPE	22	6	0.081±0.018
LPI-12:4;0	LPI	12	4	0.011±0.013
LPI-13:4;0	LPI	13	4	0.008±0.012
LPI-16:1;0	LPI	16	1	0.001±0.002
LPI-18:0;0	LPI	18	0	0.024±0.010
LPI-18:1;0	LPI	18	1	0.001±0.001
LPI-20:4;0	LPI	20	4	0.003±0.005

LPS-16:0;0	LPS	16	0	0.002±0.002
LPS-18:0;0	LPS	18	0	0.175±0.037
LPS-18:1;0	LPS	18	1	0.023±0.005
LPS-19:4;0	LPS	19	4	0.001±0.001
LPS-20:4;0	LPS	20	4	0.002±0.002
LPS-22:4;0	LPS	22	4	0.002±0.003
LPS-22:5;0	LPS	22	5	0.002±0.003
PA-32:0;0(16:0;0-16:0;0)	PA	32	0	0.004±0.006
PA-34:0;0(18:0;0-16:0;0)	PA	34	0	0.080±0.114
PA-34:1;0(18:1;0-16:0;0)	PA	34	1	0.011±0.016
PA-36:1;0(18:0;0-18:1;0)	PA	36	1	0.006±0.009
PA-36:2;0(18:0;0-18:2;0)	PA	36	2	0.002±0.003
PA-36:2;0(18:1;0-18:1;0)	PA	36	2	0.002±0.003
PA-36:4;0(18:2;0-18:2;0)	PA	36	4	0.001±0.002
PA-36:4;0(20:4;0-16:0;0)	PA	36	4	0.002±0.003
PA-38:4;0(18:0;0-20:4;0)	PA	38	4	0.011±0.015
PA-38:5;0(18:1;0-20:4;0)	PA	38	5	0.002±0.003
PA-40:6;0(18:0;0-22:6;0)	PA	40	6	0.007±0.010
PC O--32:0;0(16:0;0-16:0;0)	PC O-	32	0	0.076±0.031
PC O--32:1;0(14:1;0-18:0;0)	PC O-	32	1	0.001±0.002
PC O--32:1;0(16:0;0-16:1;0)	PC O-	32	1	0.014±0.002
PC O--32:1;0(16:1;0-16:0;0)	PC O-	32	1	0.036±0.007
PC O--34:1;0(16:0;0-18:1;0)	PC O-	34	1	0.017±0.024
PC O--34:1;0(16:1;0-18:0;0)	PC O-	34	1	0.002±0.002
PC O--34:1;0(18:1;0-16:0;0)	PC O-	34	1	0.076±0.034
PC O--34:2;0(18:1;0-16:1;0)	PC O-	34	2	0.006±0.008
PC-30:0;0(12:0;0-18:0;0)	PC	30	0	0.002±0.003
PC-30:0;0(14:0;0-16:0;0)	PC	30	0	0.075±0.014
PC-31:0;0(14:0;0-17:0;0)	PC	31	0	0.003±0.002
PC-31:0;0(15:0;0-16:0;0)	PC	31	0	0.037±0.002
PC-32:0;0(14:0;0-18:0;0)	PC	32	0	0.282±0.050
PC-32:0;0(15:0;0-17:0;0)	PC	32	0	0.013±0.018
PC-32:0;0(16:0;0-16:0;0)	PC	32	0	6.665±0.159
PC-32:1;0(14:0;0-18:1;0)	PC	32	1	0.022±0.003
PC-32:1;0(16:1;0-16:0;0)	PC	32	1	0.163±0.001
PC-32:2;0(16:1;0-16:1;0)	PC	32	2	0.251±0.355
PC-32:2;0(16:2;0-16:0;0)	PC	32	2	0.147±0.208
PC-32:3;0(14:3;0-18:0;0)	PC	32	3	0.695±0.052
PC-32:4;0(15:4;0-17:0;0)	PC	32	4	0.006±0.008

PC-33:0;0(15:0;0-18:0;0)	PC	33	0	0.010±0.003
PC-33:1;0(15:0;0-18:1;0)	PC	33	1	0.108±0.019
PC-33:1;0(16:0;0-17:1;0)	PC	33	1	0.165±0.028
PC-33:4;0(15:4;0-18:0;0)	PC	33	4	0.014±0.005
PC-34:0;0(16:0;0-18:0;0)	PC	34	0	1.773±0.137
PC-34:1;0(16:0;0-18:1;0)	PC	34	1	9.675±0.239
PC-34:1;0(16:1;0-18:0;0)	PC	34	1	0.062±0.006
PC-34:2;0(16:0;0-18:2;0)	PC	34	2	0.241±0.001
PC-34:2;0(16:1;0-18:1;0)	PC	34	2	0.055±0.002
PC-34:3;0(16:1;0-18:2;0)	PC	34	3	0.003±0.004
PC-34:3;0(18:3;0-16:0;0)	PC	34	3	0.019±0.004
PC-34:4;0(12:0;0-22:4;0)	PC	34	4	0.027±0.038
PC-35:0;0(17:0;0-18:0;0)	PC	35	0	0.011±0.015
PC-35:1;0(16:0;0-19:1;0)	PC	35	1	0.023±0.009
PC-35:1;0(17:1;0-18:0;0)	PC	35	1	0.016±0.004
PC-35:2;0(17:1;0-18:1;0)	PC	35	2	0.070±0.099
PC-36:1;0(16:0;0-20:1;0)	PC	36	1	0.252±0.002
PC-36:1;0(18:1;0-18:0;0)	PC	36	1	1.646±0.029
PC-36:2;0(16:0;0-20:2;0)	PC	36	2	0.084±0.007
PC-36:2;0(18:1;0-18:1;0)	PC	36	2	0.379±0.025
PC-36:2;0(18:2;0-18:0;0)	PC	36	2	0.070±0.001
PC-36:3;0(16:0;0-20:3;0)	PC	36	3	0.061±0.005
PC-36:3;0(18:2;0-18:1;0)	PC	36	3	0.024±0.002
PC-36:4;0(16:0;0-20:4;0)	PC	36	4	1.658±0.033
PC-36:5;0(16:1;0-20:4;0)	PC	36	5	0.465±0.657
PC-37:4;0(20:4;0-17:0;0)	PC	37	4	0.053±0.004
PC-38:1;0(16:0;0-22:1;0)	PC	38	1	0.009±0.002
PC-38:1;0(18:0;0-20:1;0)	PC	38	1	0.022±0.006
PC-38:1;0(18:1;0-20:0;0)	PC	38	1	0.003±0.005
PC-38:2;0(16:0;0-22:2;0)	PC	38	2	0.002±0.003
PC-38:2;0(18:0;0-20:2;0)	PC	38	2	0.028±0.004
PC-38:2;0(18:1;0-20:1;0)	PC	38	2	0.027±0.001
PC-38:3;0(18:1;0-20:2;0)	PC	38	3	0.001±0.002
PC-38:3;0(20:3;0-18:0;0)	PC	38	3	0.005±0.007
PC-38:4;0(16:0;0-22:4;0)	PC	38	4	0.276±0.022
PC-38:4;0(18:1;0-20:3;0)	PC	38	4	0.035±0.004
PC-38:4;0(20:4;0-18:0;0)	PC	38	4	1.149±0.003
PC-38:5;0(16:0;0-22:5;0)	PC	38	5	0.123±0.012
PC-38:5;0(20:4;0-18:1;0)	PC	38	5	0.246±0.030

PC-38:6;0(16:0;0-22:6;0)	PC	38	6	1.138±0.057
PC-38:6;0(18:2;0-20:4;0)	PC	38	6	0.017±0.000
PC-38:7;0(16:1;0-22:6;0)	PC	38	7	0.015±0.022
PC-39:5;0(17:0;0-22:5;0)	PC	39	5	0.041±0.004
PC-39:6;0(22:6;0-17:0;0)	PC	39	6	0.023±0.007
PC-40:4;0(18:0;0-22:4;0)	PC	40	4	0.139±0.002
PC-40:5;0(18:1;0-22:4;0)	PC	40	5	0.018±0.003
PC-40:5;0(20:4;0-20:1;0)	PC	40	5	0.013±0.001
PC-40:5;0(22:5;0-18:0;0)	PC	40	5	0.035±0.004
PC-40:6;0(18:1;0-22:5;0)	PC	40	6	0.024±0.034
PC-40:6;0(20:4;0-20:2;0)	PC	40	6	0.009±0.001
PC-40:6;0(22:6;0-18:0;0)	PC	40	6	0.548±0.010
PC-40:7;0(22:6;0-18:1;0)	PC	40	7	0.178±0.010
PC-40:8;0(20:4;0-20:4;0)	PC	40	8	0.020±0.002
PE O--32:1;0(16:1;0-16:0;0)	PE O-	32	1	0.013±0.018
PE O--34:1;0(16:0;0-18:1;0)	PE O-	34	1	0.010±0.014
PE O--34:1;0(18:1;0-16:0;0)	PE O-	34	1	0.067±0.010
PE O--34:2;0(16:1;0-18:1;0)	PE O-	34	2	0.111±0.021
PE O--34:2;0(18:2;0-16:0;0)	PE O-	34	2	0.071±0.006
PE O--36:1;0(18:0;0-18:1;0)	PE O-	36	1	0.013±0.003
PE O--36:2;0(18:1;0-18:1;0)	PE O-	36	2	0.183±0.020
PE O--36:3;0(18:2;0-18:1;0)	PE O-	36	3	0.188±0.033
PE O--36:4;0(16:0;0-20:4;0)	PE O-	36	4	0.047±0.014
PE O--36:4;0(16:1;0-20:3;0)	PE O-	36	4	0.012±0.017
PE O--36:5;0(16:1;0-20:4;0)	PE O-	36	5	0.678±0.083
PE O--37:5;0(17:1;0-20:4;0)	PE O-	37	5	0.010±0.013
PE O--38:2;0(18:1;0-20:1;0)	PE O-	38	2	0.010±0.014
PE O--38:4;0(16:0;0-22:4;0)	PE O-	38	4	0.047±0.000
PE O--38:4;0(18:0;0-20:4;0)	PE O-	38	4	0.034±0.003
PE O--38:4;0(18:1;0-20:3;0)	PE O-	38	4	0.021±0.003
PE O--38:5;0(16:0;0-22:5;0)	PE O-	38	5	0.003±0.004
PE O--38:5;0(16:1;0-22:4;0)	PE O-	38	5	0.418±0.170
PE O--38:5;0(18:1;0-20:4;0)	PE O-	38	5	0.672±0.313
PE O--38:5;0(21:5;0-17:0;0)	PE O-	38	5	0.672±0.147
PE O--38:6;0(16:0;0-22:6;0)	PE O-	38	6	0.070±0.005
PE O--38:6;0(16:1;0-22:5;0)	PE O-	38	6	0.068±0.001
PE O--38:6;0(18:2;0-20:4;0)	PE O-	38	6	0.368±0.023
PE O--38:7;0(16:1;0-22:6;0)	PE O-	38	7	1.397±0.117
PE O--39:7;0(17:1;0-22:6;0)	PE O-	39	7	0.020±0.028

PE O--40:4;0(18:0;0-22:4;0)	PE O-	40	4	0.059±0.004
PE O--40:5;0(18:0;0-22:5;0)	PE O-	40	5	0.015±0.002
PE O--40:5;0(18:1;0-22:4;0)	PE O-	40	5	0.593±0.040
PE O--40:6;0(18:0;0-22:6;0)	PE O-	40	6	0.068±0.004
PE O--40:6;0(18:1;0-22:5;0)	PE O-	40	6	0.082±0.002
PE O--40:6;0(18:2;0-22:4;0)	PE O-	40	6	0.135±0.024
PE O--40:7;0(18:1;0-22:6;0)	PE O-	40	7	2.569±0.212
PE O--40:8;0(18:2;0-22:6;0)	PE O-	40	8	0.440±0.049
PE O--42:7;0(20:1;0-22:6;0)	PE O-	42	7	0.019±0.026
PE-32:0;0(16:0;0-16:0;0)	PE	32	0	0.025±0.001
PE-34:0;0(16:0;0-18:0;0)	PE	34	0	0.089±0.009
PE-34:1;0(16:0;0-18:1;0)	PE	34	1	0.371±0.001
PE-34:1;0(16:1;0-18:0;0)	PE	34	1	0.012±0.001
PE-34:2;0(16:0;0-18:2;0)	PE	34	2	0.008±0.001
PE-34:2;0(16:1;0-18:1;0)	PE	34	2	0.010±0.001
PE-35:2;0(17:1;0-18:1;0)	PE	35	2	0.003±0.004
PE-36:1;0(16:0;0-20:1;0)	PE	36	1	0.012±0.001
PE-36:1;0(18:1;0-18:0;0)	PE	36	1	0.337±0.007
PE-36:2;0(16:0;0-20:2;0)	PE	36	2	0.004±0.001
PE-36:2;0(17:1;0-19:1;0)	PE	36	2	0.000±0.001
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PE-36:4;0(16:0;0-20:4;0)	PE	36	4	0.344±0.000
PE-36:4;0(16:1;0-20:3;0)	PE	36	4	0.001±0.001
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PE-38:1;0(18:0;0-20:1;0)	PE	38	1	0.005±0.007
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PE-38:5;0(20:5;0-18:0;0)	PE	38	5	0.003±0.004
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PE-38:6;0(18:2;0-20:4;0)	PE	38	6	0.027±0.006

PE-38:7;0(16:1;0-22:6;0)	PE	38	7	0.008±0.000
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PE-40:8;0(20:4;0-20:4;0)	PE	40	8	0.029±0.000
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PG-34:1;0(16:1;0-18:0;0)	PG	34	1	0.003±0.000
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PI-36:1;0(18:0;0-18:1;0)	PI	36	1	0.002±0.002
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PS-36:2;0(18:1;0-18:1;0)	PS	36	2	0.346±0.016
PS-36:2;0(18:2;0-18:0;0)	PS	36	2	0.022±0.015
PS-36:3;0(18:2;0-18:1;0)	PS	36	3	0.005±0.007
PS-36:4;0(16:0;0-20:4;0)	PS	36	4	0.009±0.012
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PS-38:1;0(18:1;0-20:0;0)	PS	38	1	0.001±0.002
PS-38:2;0(18:0;0-20:2;0)	PS	38	2	0.003±0.004
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PS-38:2;0(19:1;0-19:1;0)	PS	38	2	0.000±0.000
PS-38:3;0(20:3;0-18:0;0)	PS	38	3	0.011±0.015
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PS-38:4;0(20:4;0-18:0;0)	PS	38	4	0.004±0.006
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PS-40:7;0(22:6;0-18:1;0)	PS	40	7	0.047±0.066
PS-42:4;0(18:0;0-24:4;0)	PS	42	4	0.005±0.007
PS-44:10;0(22:5;0-22:5;0)	PS	44	10	0.001±0.001
PS-44:10;0(22:6;0-22:4;0)	PS	44	10	0.027±0.038
PS-44:11;0(22:6;0-22:5;0)	PS	44	11	0.005±0.007
PS-44:12;0(22:6;0-22:6;0)	PS	44	12	0.013±0.018
SM-33:1;2	SM	33	1	0.008±0.002
SM-34:1;2	SM	34	1	0.043±0.006
SM-35:1;2	SM	35	1	0.034±0.002

SM-35:4;2	SM	35	4	0.045±0.010
SM-36:1;2	SM	36	1	1.792±0.147
SM-36:1;3	SM	36	1	0.014±0.001
SM-36:2;2	SM	36	2	0.113±0.007
SM-37:2;2	SM	37	2	0.030±0.008
SM-37:4;2	SM	37	4	0.014±0.001
SM-38:1;2	SM	38	1	0.184±0.017
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SM-42:2;2	SM	42	2	0.019±0.001

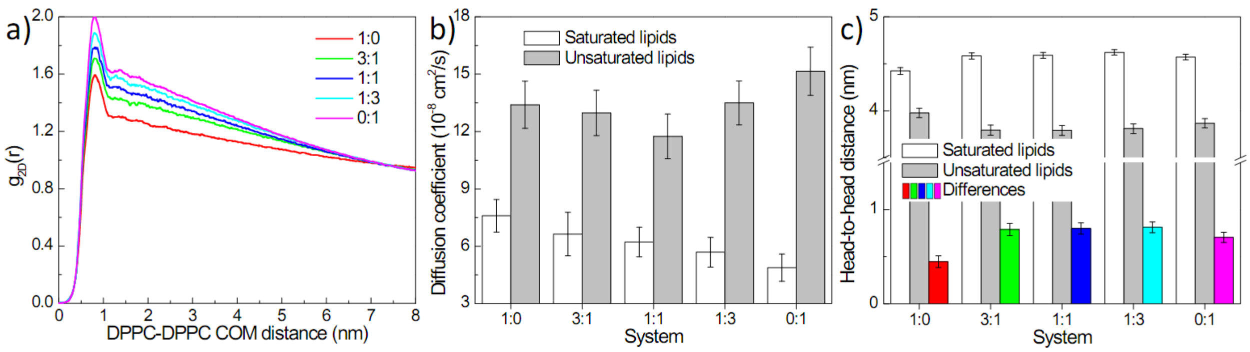

Figure S2. Four-component lipid systems with increasing DDPC molar ratio (ratios shown are DLiPC:DDPC): **a)** Two-dimensional (2D) radial pair distribution function of DPPC molecules; **b)** Diffusion coefficients of saturated lipids and unsaturated lipids indicate that all lipids are in the fluid phase; **c)** Head-to-head (phosphate-to-phosphate) distance along z axis of saturated lipids, unsaturated lipids and their differences.

Table S2. Dataset for **Fig. 2 c-e**

System	**Saturated lipids**	**Unsaturated lipids**	**Differences**
Order parameter, S_z			
1:0	0.674±0.014	0.320±0.013	0.354±0.018
3:1	0.694±0.013	0.284±0.012	0.410±0.017
1:1	0.700±0.013	0.259±0.011	0.441±0.016
1:3	0.717±0.013	0.235±0.012	0.482±0.016
0:1	0.730±0.014	0.212±0.012	0.519±0.017
Lipid height, h (nm)			
1:0	1.893±0.013	1.468±0.016	0.425±0.019
3:1	1.911±0.012	1.437±0.017	0.473±0.019
1:1	1.914±0.011	1.420±0.017	0.493±0.020
1:3	1.925±0.012	1.408±0.019	0.518±0.021
0:1	1.930±0.012	1.395±0.020	0.535±0.022
Cholesterol preference*, χ			
1:0	0.847±0.015	0.153±0.015	0.693±0.015
3:1	0.891±0.010	0.109±0.010	0.782±0.020
1:1	0.914±0.009	0.086±0.009	0.827±0.018
1:3	0.941±0.008	0.059±0.008	0.882±0.016
0:1	0.965±0.006	0.035±0.006	0.930±0.006

*Since sum of the cholesterol preferences for saturated and unsaturated lipids equals 1, they share the same standard deviation.

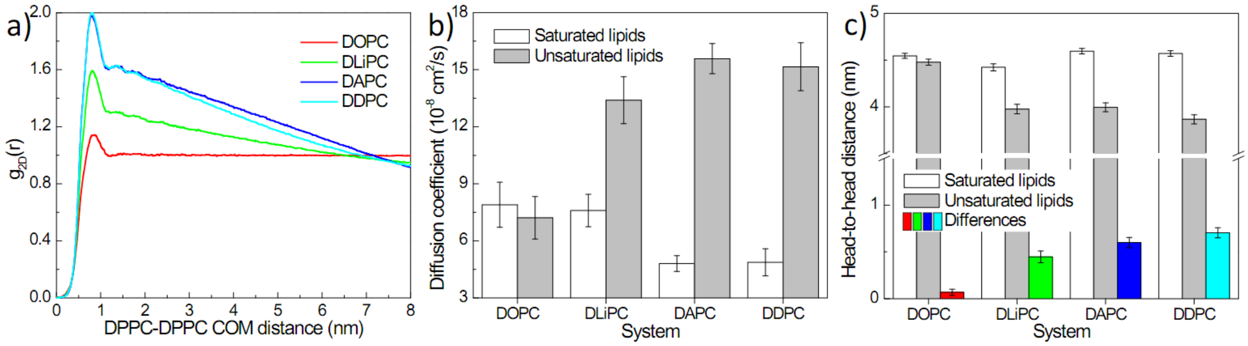

Figure S3. Three-component lipid systems with increasing unsaturation of unsaturated lipids: **a)** Two-dimensional (2D) radial pair distribution function of DPPC molecules; **b)** Diffusion coefficients of saturated lipids and unsaturated lipids indicate that all lipids are in the fluid phase; **c)** Head-to-head (phosphate-to-phosphate) distance along z axis of saturated lipids, unsaturated lipids and their differences.

Table S3. Dataset for **Fig. 3 c-e**

System	**Saturated lipids**	**Unsaturated lipids**	**Differences**
Order parameter, S_z			
DOPC	0.619±0.011	0.497±0.012	0.122±0.013
DLiPC	0.674±0.014	0.320±0.013	0.354±0.018
DAPC	0.737±0.014	0.236±0.011	0.501±0.017
DDPC	0.730±0.014	0.211±0.012	0.519±0.017
Lipid height, h (nm)			
DOPC	1.852±0.011	2.024±0.017	-0.172±0.017
DLiPC	1.893±0.013	1.468±0.016	0.425±0.019
DAPC	1.939±0.012	1.536±0.019	0.403±0.022
DDPC	1.930±0.012	1.395±0.020	0.535±0.022
Cholesterol preference*, χ			
DOPC	0.642±0.012	0.358±0.012	0.285±0.012
DLiPC	0.847±0.015	0.153±0.015	0.693±0.015
DAPC	0.939±0.009	0.061±0.009	0.878±0.009
DDPC	0.965±0.006	0.035±0.006	0.930±0.006

*Since sum of the cholesterol preferences for saturated and unsaturated lipids equals 1, they share the same standard deviation.

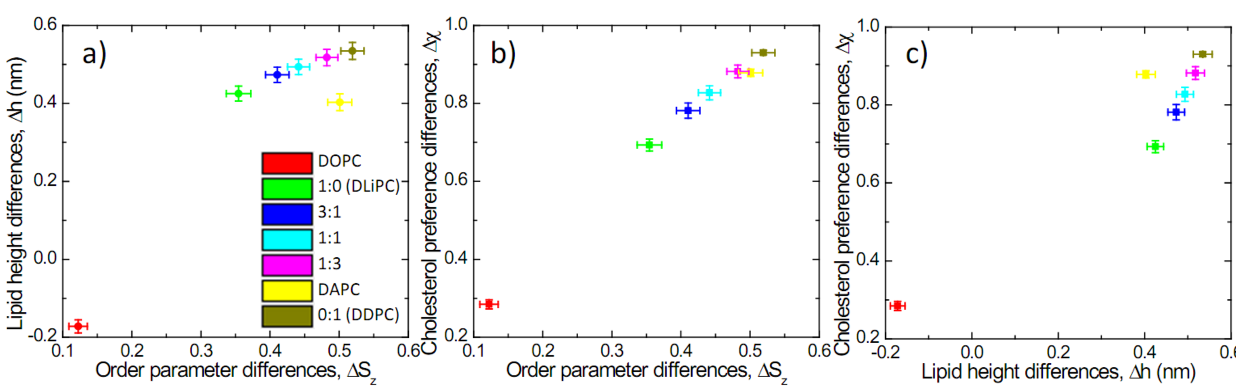

Figure S4. Correlation among the differences of the lipid chain order parameter S_z, lipid height h and cholesterol preference χ between L_o and L_d domains. Results in **(a-c)** are obtained via analyzing the last 4 μs trajectories of 20 μs simulations, and error represents the standard deviation. Order parameter differences correlate best with cholesterol preference differences.

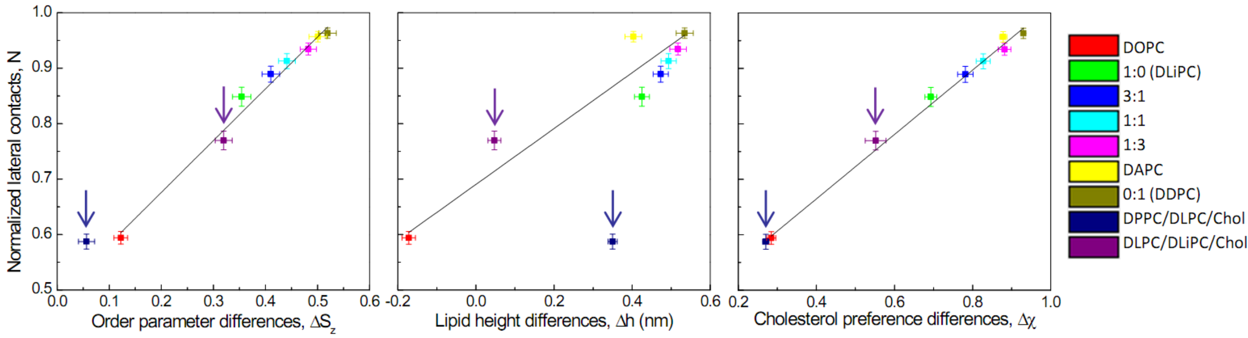

Figure S5. Figure 4 of the main text, including data from the DPPC/DLPC/Chol and DLPC/DLiPC/Chol systems, showing that differences in lipid chain order and cholesterol preference show improved correlation with normalized lateral contacts of unsaturated lipids that measure domain size / stability.

Supporting references:

1. Hong, C., D.P. Tieleman, and Y. Wang, *Microsecond molecular dynamics simulations of lipid mixing. Langmuir*, **2014**, 30, 11993-12001.
2. Dick, R.A. and V.M. Vogt, *Membrane interaction of retroviral Gag proteins. Front. Microbiol.*, **2014**, 5, 187.