SUPPLEMENTARY INFORMATION

Acid-labile mPEG-Vinyl Ether-1,2-Dioleylglycerol Lipids with Tunable pH Sensitivity: Synthesis and Structural Effects on Hydrolysis Rates, DOPE Liposome Release Performance and Pharmacokinetics

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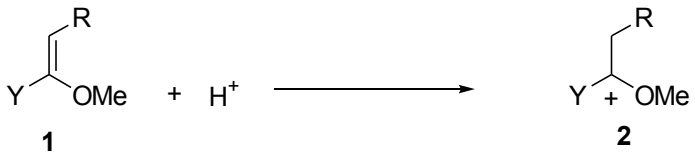
1. Details of DFT Computations

Density functional theory (DFT) calculations were used to determine the proton affinity (PA) values of a series of vinyl ethers using Gaussian 03.[1] The DFT method was employed using the B3LYP hybrid functional.[2,3] Neutral and protonated structures were optimized with 6-31G** basis set. Single point energies of the optimized structures were obtained with the cc-pVTZ basis set.[4]

The PA is the negative of the standard enthalpy variation (ΔrH°) for the reaction,



1 **2**

where PA(M) = $-\Delta rH_{298K.}$

Table S1. PA values of a series of methyl vinyl ethers **1**.

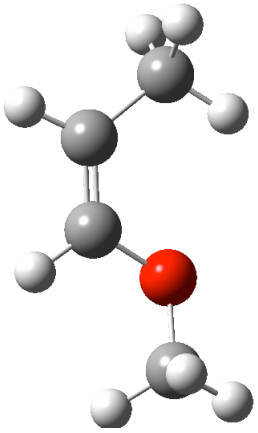
CURVE	R	Y	Neutral (hartree)	Protonated (hartree)	PA (kcal/mol)
1a	Me	H	-232.5224815	-232.8665094	-215.88
1b	H	Me	-232.5219559	-232.8838625	-227.10
1c	H	Ph	-424.3222993	-424.6918630	-231.90
1d	H	C(O)OMe	-421.152028864	-421.487347205	-210.42
1e	H	OMe	-307.7636225	-308.1461169	-240.02
1f	Ph	OMe	-538.8910531	-539.2711618	-238.52
1g	OMe	OMe	-422.3154414	-422.698326	-240.26
1h	H	C(O)NHMe			-207.51
1i	H	$CH_2OC(O)NHMe$			-237.32

2. Optimized Structures and Energies of CURVE

1a (neutral)

SCF Done: E(RB+HF-LYP) = -232.522481542 A.U. after 7 cycles

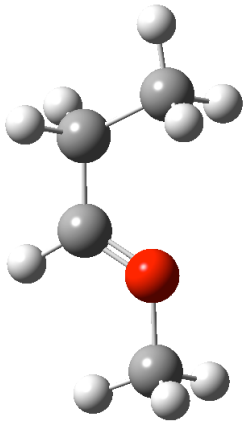
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.060027	0.717308	-0.061671
2	6	0	1.269857	0.649651	0.037431
3	1	0	1.800768	1.595990	0.099833
4	8	0	-0.847327	-0.392121	-0.213003
5	6	0	2.074311	-0.617231	0.048552
6	1	0	1.424148	-1.494570	0.051990
7	1	0	2.728865	-0.683900	-0.830063
8	1	0	2.724834	-0.666242	0.930694
9	6	0	-2.213206	-0.209000	0.124714
10	1	0	-2.344552	-0.026996	1.199780
11	1	0	-2.656698	0.628309	-0.432402
12	1	0	-2.732874	-1.129470	-0.147514
13	1	0	-0.591488	1.669478	-0.062445



2a (protonated)

SCF Done: E(RB+HF-LYP) = -232.866509399 A.U. after 7 cycles

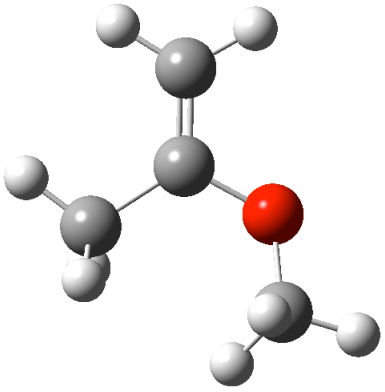
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	0.112576	0.733999	-0.000144
2	6	0	-1.347533	0.643966	0.000028
3	1	0	-1.671553	1.259463	-0.859908
4	8	0	0.827792	-0.302836	-0.000187
5	6	0	-1.964236	-0.752971	0.000074
6	1	0	-1.669022	-1.318610	-0.886639
7	1	0	-1.669036	-1.318550	0.886834
8	1	0	-3.051627	-0.663848	0.000071
9	6	0	2.304226	-0.247824	0.000105
10	1	0	2.616060	-0.779838	-0.897515
11	1	0	2.636211	0.791247	0.000421
12	1	0	2.615752	-0.780288	0.897564
13	1	0	0.612398	1.710424	0.000589
14	1	0	-1.671710	1.259666	0.859702



1b (neutral)

SCF Done: E(RB+HF-LYP) = -232.521955932 A.U. after 6 cycles

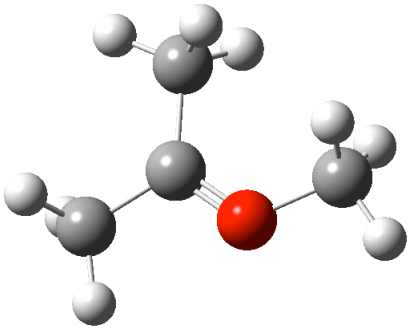
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	0.555853	-0.088060	-0.040812
2	6	0	1.623729	-0.873185	0.130449
3	1	0	2.610492	-0.446022	0.258729
4	8	0	-0.652657	-0.690063	-0.315010
5	6	0	-1.835040	-0.074398	0.176131
6	1	0	-1.824869	0.014022	1.270857
7	1	0	-2.005516	0.917886	-0.259438
8	1	0	-2.658801	-0.728462	-0.115971
9	1	0	1.525188	-1.952275	0.115788
10	6	0	0.606557	1.416443	-0.037579
11	1	0	0.022833	1.850887	0.782185
12	1	0	1.638248	1.754861	0.072479
13	1	0	0.207080	1.824805	-0.973689



2b (protonated)

SCF Done: E(RB+HF-LYP) = -232.883862491 A.U. after 6 cycles

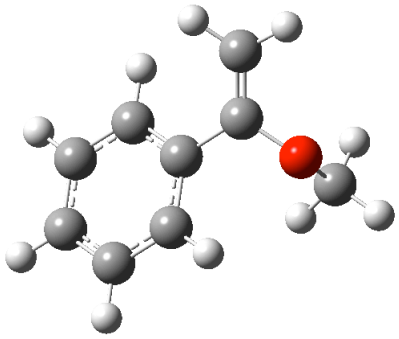
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.467231	-0.003916	-0.000275
2	6	0	-1.754276	-0.727065	0.000092
3	1	0	-2.338406	-0.411857	0.875500
4	8	0	0.574323	-0.736848	-0.000068
5	6	0	1.947293	-0.203526	0.000074
6	1	0	2.098873	0.388338	-0.903185
7	1	0	2.098434	0.389130	0.902884
8	1	0	2.578372	-1.088149	0.000608
9	1	0	-2.338520	-0.412369	-0.875428
10	6	0	-0.394045	1.477359	0.000007
11	1	0	0.160588	1.828827	-0.880157
12	1	0	-1.387878	1.922980	-0.000221
13	1	0	0.159913	1.828430	0.880776
14	1	0	-1.616406	-1.807655	0.000382



1c (neutral)

SCF Done: E(RB+HF-LYP) = -424.322299341 A.U. after 7 cycles

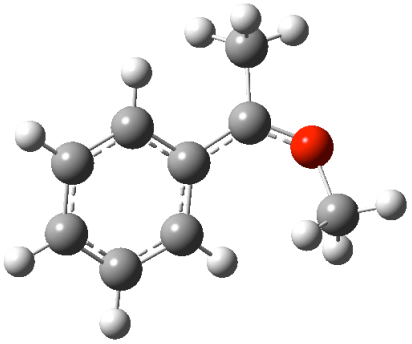
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-1.369855	0.529381	-0.296574
2	6	0	-1.886078	1.741172	-0.535789
3	8	0	-2.219425	-0.561820	-0.287358
4	6	0	-2.293860	-1.269440	0.950727
5	1	0	-1.314511	-1.642717	1.271504
6	1	0	-2.707874	-0.629803	1.740727
7	1	0	-2.964452	-2.114355	0.782524
8	1	0	-2.957518	1.868049	-0.636928
9	1	0	-1.248479	2.605014	-0.678350
10	6	0	0.079197	0.244392	-0.139696
11	6	0	0.945965	1.174667	0.455418
12	6	0	0.611915	-0.967207	-0.610117
13	6	0	2.310431	0.910431	0.557206
14	1	0	0.540573	2.099486	0.854310
15	6	0	1.976011	-1.231786	-0.503689
16	1	0	-0.053441	-1.688295	-1.073935
17	6	0	2.830847	-0.293563	0.078133
18	1	0	2.966862	1.640331	1.022241
19	1	0	2.373325	-2.170233	-0.879817
20	1	0	3.893481	-0.501199	0.162883



2c (protonated)

SCF Done: E(RB+HF-LYP) = -424.691862966 A.U. after 8 cycles

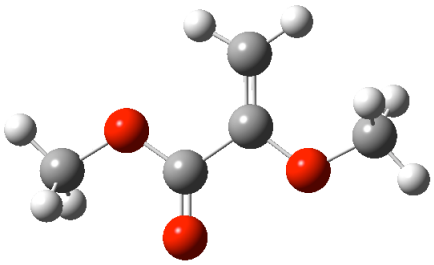
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.601343	-1.085219	-0.405770
2	6	0	-0.088144	0.190903	-0.056198
3	6	0	-0.996130	1.216424	0.315983
4	6	0	-2.355836	0.956990	0.386491
5	6	0	-2.843744	-0.308919	0.041677
6	6	0	-1.967697	-1.320194	-0.369108
7	1	0	0.053372	-1.866189	-0.769089
8	1	0	-0.628790	2.201053	0.581247
9	1	0	-3.039353	1.738541	0.699550
10	1	0	-3.910636	-0.504666	0.081796
11	1	0	-2.355094	-2.288815	-0.665534
12	6	0	1.314612	0.519913	-0.129886
13	6	0	1.797734	1.913579	-0.370434
14	1	0	1.180148	2.433743	-1.103838
15	8	0	2.294844	-0.309986	0.003350
16	6	0	2.238520	-1.684834	0.495014
17	1	0	1.384368	-1.821528	1.157148
18	1	0	2.198324	-2.356078	-0.363012
19	1	0	3.179496	-1.815794	1.026321
20	1	0	1.751684	2.475664	0.572362
21	1	0	2.839890	1.892103	-0.690354



1d (neutral)

SCF Done: E(RB+HF-LYP) = -421.152028864 A.U. after 8 cycles

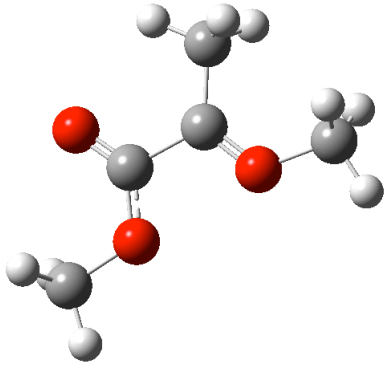
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.689357	0.299235	-0.000021
2	6	0	-0.898907	1.624571	-0.000028
3	1	0	-0.051403	2.294388	-0.000053
4	8	0	-1.629627	-0.677483	0.000010
5	6	0	0.682042	-0.315243	-0.000045
6	8	0	0.895272	-1.506683	-0.000039
7	1	0	-1.889724	2.059842	-0.000010
8	8	0	1.655603	0.623173	0.000006
9	6	0	2.994160	0.103514	0.000052
10	1	0	3.170012	-0.508317	-0.888165
11	1	0	3.647410	0.975701	0.000090
12	1	0	3.169917	-0.508415	0.888220
13	6	0	-2.986294	-0.263345	0.000036
14	1	0	-3.581872	-1.176663	0.000058
15	1	0	-3.222076	0.329513	0.893273
16	1	0	-3.222112	0.329501	-0.893187



2d (protonated)

SCF Done: E(RB+HF-LYP) = -421.487347205 A.U. after 8 cycles

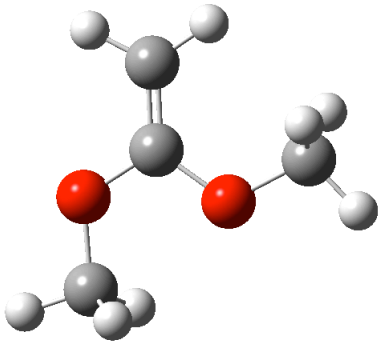
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	0.720395	0.311619	0.000021
2	8	0	1.200907	-0.856823	-0.000268
3	6	0	2.651333	-1.140141	-0.000214
4	1	0	3.089470	-0.711092	-0.902068
5	1	0	3.087895	-0.716787	0.905112
6	1	0	2.711858	-2.224870	-0.003492
7	6	0	-0.815028	0.401108	0.000029
8	8	0	-1.335405	1.491973	-0.000680
9	8	0	-1.370480	-0.789142	0.000415
10	6	0	-2.832882	-0.817389	0.000117
11	1	0	-3.091483	-1.873003	0.000583
12	1	0	-3.203963	-0.314832	0.894355
13	1	0	-3.203585	-0.315713	-0.894772
14	6	0	1.516531	1.549254	0.000582
15	1	0	2.176432	1.571286	0.879570
16	1	0	2.172721	1.573980	-0.881178
17	1	0	0.858374	2.416273	0.002944



1e (neutral)

SCF Done: E(RB+HF-LYP) = -307.763622465 A.U. after 7 cycles

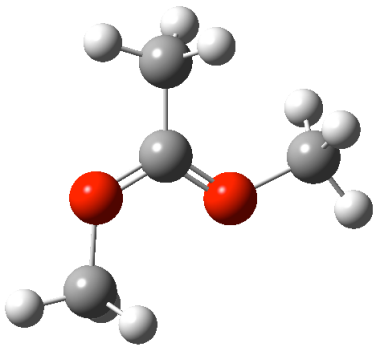
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.035618	0.512160	-0.000167
2	6	0	-0.738429	1.654528	0.000308
3	1	0	-1.817295	1.667662	0.000382
4	8	0	1.319799	0.533666	-0.000598
5	6	0	2.029796	-0.704187	0.000432
6	1	0	1.806714	-1.299863	0.891091
7	1	0	1.806554	-1.301427	-0.889127
8	1	0	3.085023	-0.427292	0.000100
9	1	0	-0.205510	2.594605	0.000514
10	8	0	-0.527347	-0.754098	-0.000373
11	6	0	-1.941386	-0.898660	0.000113
12	1	0	-2.137149	-1.971553	0.000008
13	1	0	-2.381743	-0.441002	0.893494
14	1	0	-2.382388	-0.440724	-0.892806



2e (protonated)

SCF Done: E(RB+HF-LYP) = -308.146116926 A.U. after 7 cycles

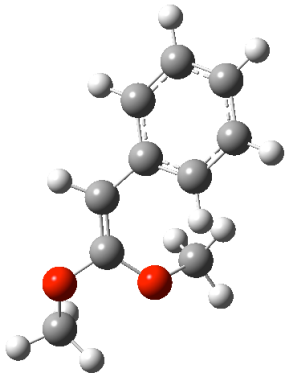
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.002278	0.408812	0.000205
2	6	0	0.795354	1.662067	-0.000077
3	8	0	-1.276370	0.519921	0.000126
4	1	0	0.127823	2.522286	0.002677
5	8	0	0.492456	-0.779217	0.000503
6	6	0	1.941832	-1.026474	-0.000241
7	1	0	2.028921	-2.109870	-0.004068
8	1	0	2.380874	-0.598752	-0.901721
9	1	0	2.380264	-0.605226	0.904605
10	1	0	1.439107	1.696263	-0.885953
11	1	0	1.443683	1.693918	0.882505
12	6	0	-2.147667	-0.670778	-0.000263
13	1	0	-1.945777	-1.255624	-0.897120
14	1	0	-1.953529	-1.249822	0.902090
15	1	0	-3.153494	-0.260573	-0.005795



1f (neutral)

SCF Done: E(RB+HF-LYP) = -538.891053113 A.U. after 8 cycles

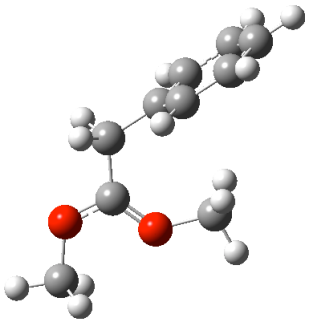
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-1.599255	-0.284304	-0.217192
2	6	0	-0.459030	-0.998700	-0.107160
3	8	0	-2.753380	-0.930533	-0.523175
4	6	0	-3.972690	-0.402943	0.008514
5	1	0	-3.962270	-0.404962	1.105633
6	1	0	-4.162783	0.611117	-0.349671
7	1	0	-4.757634	-1.073359	-0.343759
8	1	0	-0.602160	-2.074564	-0.088689
9	8	0	-1.752453	1.064996	-0.114188
10	6	0	-1.212406	1.714356	1.046241
11	1	0	-1.771110	2.645718	1.157587
12	1	0	-1.346525	1.090181	1.934813
13	1	0	-0.149436	1.935109	0.922147
14	6	0	0.924688	-0.516462	-0.088664
15	6	0	1.922892	-1.318119	0.498755
16	6	0	1.328458	0.692248	-0.691386
17	6	0	3.257395	-0.920187	0.509428
18	1	0	1.638466	-2.263638	0.953406
19	6	0	2.663875	1.090631	-0.676826
20	1	0	0.588430	1.304130	-1.197701
21	6	0	3.636472	0.291068	-0.073164
22	1	0	4.004428	-1.558282	0.973529
23	1	0	2.948077	2.024967	-1.153418
24	1	0	4.676795	0.602361	-0.066257



2f (protonated)

SCF Done: E(RB+HF-LYP) = -539.271161795 A.U. after 8 cycles

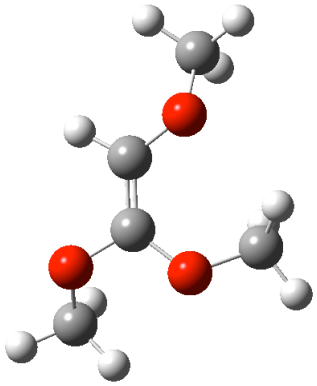
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	1.688294	-0.235539	-0.000428
2	6	0	0.530602	-1.203385	-0.001655
3	8	0	2.839298	-0.813732	0.000042
4	6	0	4.085195	-0.034065	0.001025
5	1	0	4.122618	0.582317	-0.896882
6	1	0	4.121896	0.581154	0.899760
7	1	0	4.869030	-0.786502	0.000861
8	1	0	0.716931	-1.851041	0.865500
9	8	0	1.703572	1.045698	0.000161
10	6	0	0.557651	1.976970	-0.000706
11	1	0	1.034187	2.954244	-0.000854
12	1	0	-0.034279	1.816625	-0.898179
13	1	0	-0.035002	1.817254	0.896410
14	6	0	-0.887759	-0.681167	-0.000671
15	6	0	-1.557616	-0.456379	-1.210353
16	6	0	-1.556145	-0.457085	1.209969
17	6	0	-2.870449	0.017091	-1.208517
18	1	0	-1.060659	-0.662295	-2.155456
19	6	0	-2.868967	0.016407	1.210014
20	1	0	-1.058047	-0.663581	2.154346
21	6	0	-3.524478	0.259823	0.001215
22	1	0	-3.384738	0.184002	-2.149425
23	1	0	-3.382109	0.182783	2.151643
24	1	0	-4.547413	0.621955	0.001946
25	1	0	0.716659	-1.848658	-0.870656



1g (neutral)

SCF Done: E(RB+HF-LYP) = -422.315441378 A.U. after 7 cycles

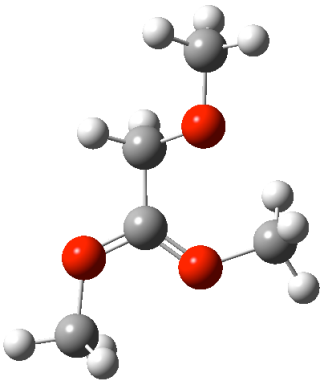
Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.533740	-0.201858	-0.289463
2	6	0	0.650387	-0.831128	-0.221651
3	8	0	-1.664193	-0.956460	-0.366087
4	6	0	-2.713811	-0.589319	0.535365
5	1	0	-2.379071	-0.665770	1.578224
6	1	0	-3.069457	0.426086	0.342477
7	1	0	-3.521407	-1.301965	0.361196
8	1	0	0.669496	-1.911496	-0.102836
9	8	0	-0.788433	1.130164	-0.382340
10	6	0	0.141312	2.042836	0.203703
11	1	0	-0.365403	3.009683	0.225460
12	1	0	0.393948	1.736707	1.226144
13	1	0	1.058890	2.110703	-0.383396
14	8	0	1.855277	-0.194604	-0.434487
15	6	0	2.876516	-0.605616	0.462854
16	1	0	3.796983	-0.109604	0.147515
17	1	0	2.644454	-0.322433	1.499108
18	1	0	3.026371	-1.694203	0.424569



2g (protonated)

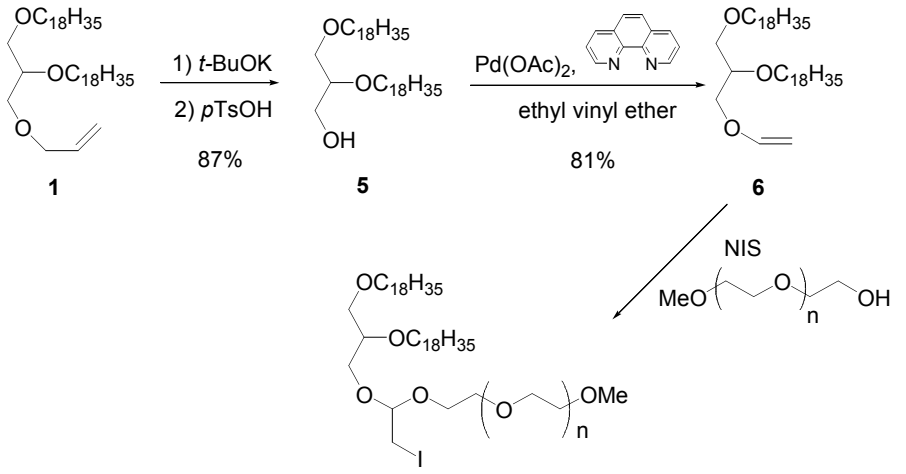
SCF Done: E(RB+HF-LYP) = -422.698326045 A.U. after 7 cycles

Center Number	Atomic Number	Atomic Type	Coordinates (Angstroms) X	Y	Z
1	6	0	-0.588808	-0.201177	0.096984
2	6	0	0.764670	-0.806126	0.380787
3	8	0	-1.555565	-1.038495	0.025318
4	6	0	-2.943576	-0.581390	-0.171526
5	1	0	-3.214743	0.091965	0.641124
6	1	0	-3.014005	-0.082892	-1.137820
7	1	0	-3.528731	-1.496319	-0.148627
8	1	0	0.716494	-1.862174	0.078814
9	8	0	-0.856506	1.039468	-0.043693
10	6	0	0.113860	2.151230	0.091415
11	1	0	-0.522042	3.026500	0.196559
12	1	0	0.731263	1.991519	0.972178
13	1	0	0.715059	2.184764	-0.812829
14	8	0	1.736416	-0.074059	-0.299999
15	6	0	3.059021	-0.609510	-0.133530
16	1	0	3.721975	0.034559	-0.709839
17	1	0	3.356735	-0.597091	0.922112
18	1	0	3.114987	-1.633697	-0.520294
19	1	0	0.897253	-0.790609	1.480828

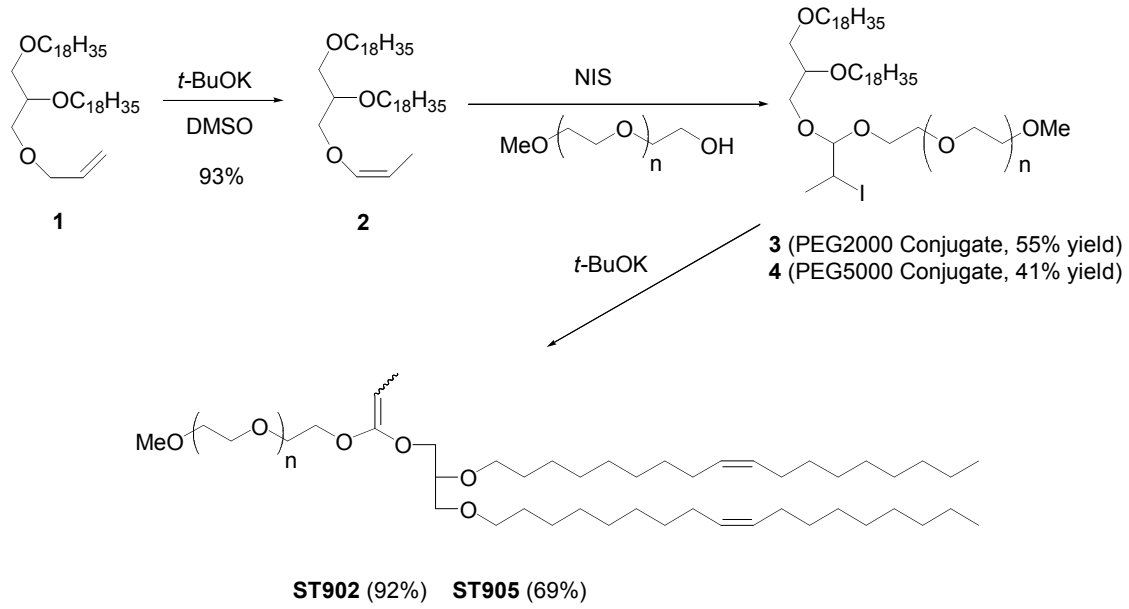


3. Synthesis of mPEG-Vinyl Ether Lipids

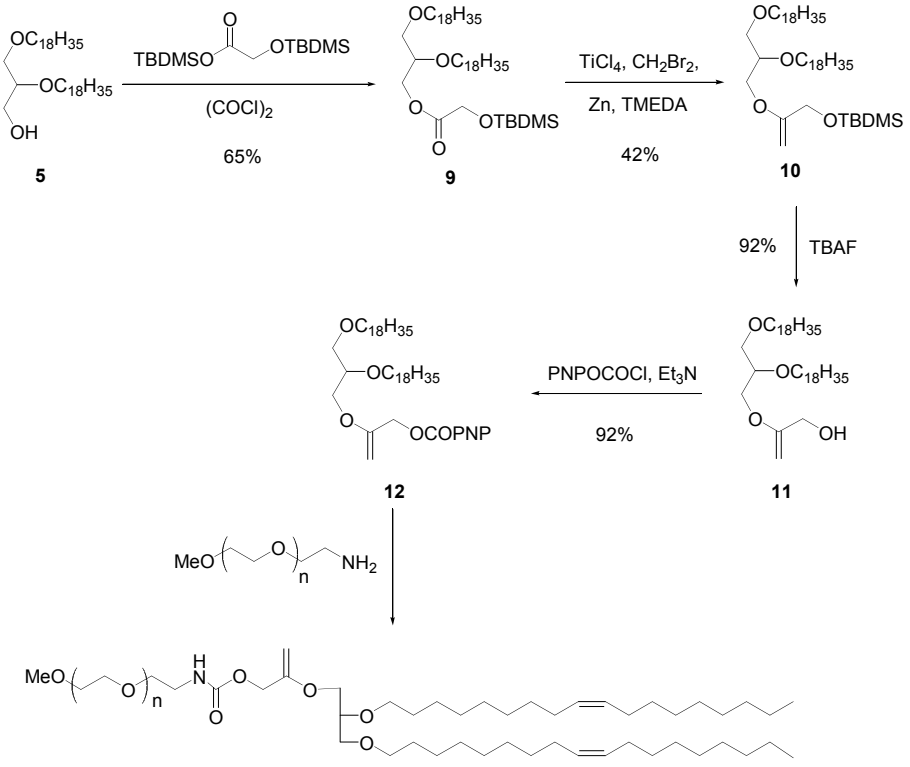
3a. Synthesis route for MeO-PEG-1'-oxyethene-1'-O-1,2-dioleylglycerol (ST912 & 915).



3b. Synthesis route for MeO-PEG-1'-oxy-1'-propene-1'-O-1,2-dioleylglycerol (ST902 & 905).



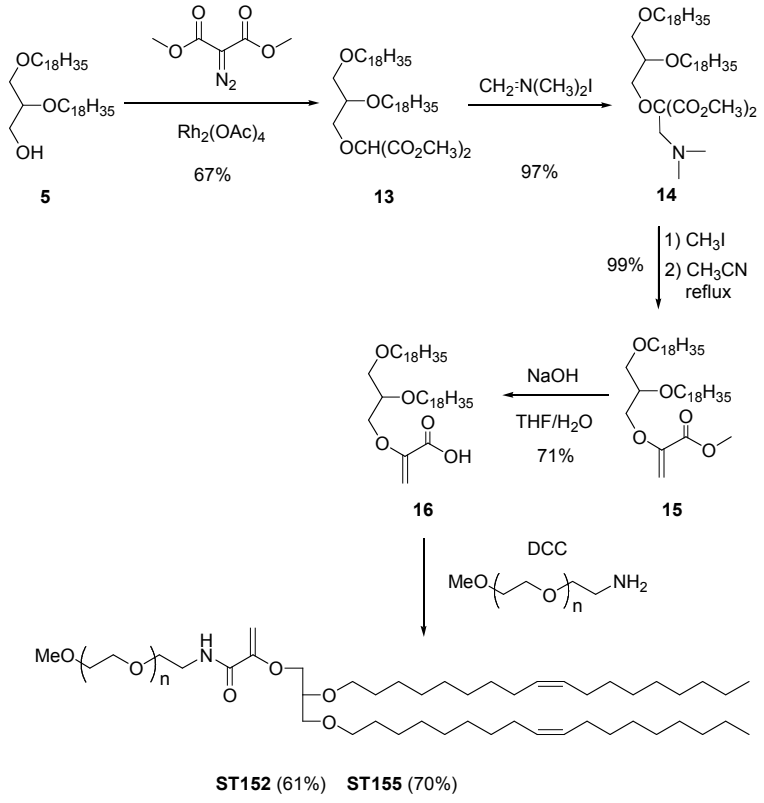
3c. Synthesis route for MeO-PEG-1'-oxy-2'-propene-2'-O-1,2-dioleylglycerol (ST502 & 505).



5 9 10 11 12

ST502 (83%) **ST505** (71%)

3d. Synthesis route for MeO-PEG-1'-amido-2'-propene-2'-O-1,2-dioleylglycerol (ST152 & 155).



5 13 14 15 16

ST152 (61%) **ST155** (70%)

4. Detailed Description of MeO-PEG-VE-DOG Lipid Syntheses

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)propene (2). t-BuOK (552 mg, 4.92 mmol) was added to a flask containing **1** (3.114 g, 4.92 mmol) in DMSO (50 mL). The mixture was stirred at 100 °C for 3 h and then diluted with diethyl ether (200 mL) before washing with water (2 x 50 mL). Silica gel column chromatographic purification (10:1 hexane:Et_2O as eluent) of the organic residue gave **2** as an oil (2.881 g, 4.55 mmol, 93% yield). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H), 1.26 (m, 44H), 1.54 (m, 7H), 1.98 (m, 8H), 3.39-3.85 (m, 9H), 4.35 (quintet, 1H, J = 6 Hz), 5.32 (m, 4H), 5.94 (d, 1H, J = 6 Hz); ^{13}C NMR ($CDCl_3$): δ 9.4, 14.3, 22.9, 26.3, 26.4, 27.5, 29.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 30.3, 32.2, 32.8, 70.5, 71.0, 71.9, 72.4, 78.0, 101.1, 130.1, 130.2, 146.3; CI $(M + H)^+$ calc'd 633, found 633.

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)-1-(ω-methoxy-polyethylene glycol[2000]oxy)-2-iodopropane (3). N-Iodosuccinimide (246 mg, 1.10 mmol) was added to a flask containing **2** (750 mg, 1.18 mmol) and mPEG2000 (1.82 g, 0.911 mmol) in CH_2Cl_2 (20 mL) at 0 °C and the mixture stirred at 23 °C for 3 h. The reaction mixture then was directly loaded onto a silica gel column and purified via step gradient elution with 20:1, then 10:1 CH_2Cl_2:MeOH to give **3** (1.385 g, 5.02 mmol, 55%). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.52 (m, 4H), 1.83 (d, 3H, J = 7 Hz), 1.98 (m, 8H), 3.3-3.9 (m, ca. 200H), 4.16 (m, 1H), 4.45 (m, 1H), 5.32 (m, 4H).

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)-1-(ω-methoxy-polyethylene glycol[5000]oxy)-2-iodopropane (4). Compound **4** was prepared as described above in 41% yield using mPEG5000. ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.52 (m, 4H), 1.83 (d, 3H, J = 7 Hz), 1.98 (m, 8H), 3.3-3.9 (m, ca. 500H), 4.16 (m, 1H), 4.45 (m, 1H), 5.32 (m, 4H).

ST902. t-BuOK (117 mg, 1.04 mmol) was added to 30 mL of THF containing **3** (1.308g, 0.474 mmol) and the mixture stirred at 23 °C for 4 h. The reaction mixture then was diluted with CH_2Cl_2 (50 mL) and washed with 0.05 M of Na_2CO_3 solution (10 mL). The organic layer was dried over anhydrous

Na_2CO_3 and concentrated to give a solid. The resulting solid was dissolved in benzene and lyophilized to give **ST902** (1.148 g, 0.436 mmol, 92%). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.51 (d, 3H, J = 7 Hz), 1.52 (m, 4H), 1.98 (m, 8H), 3.3-4.0 (m, ca. 200H), 5.32 (m, 4H).

ST905. ST905 was prepared from **4** in 69% yield as described above for **ST902**. ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.51 (d, 3H, J = 7 Hz), 1.52 (m, 4H), 1.98 (m, 8H), 3.3-4.0 (m, ca. 500H), 5.32 (m, 4H).

1,2-Di-O-oleyl-*rac*-glycerol (5). t-BuOK (1.514 g, 13.5 mmol) was added to a flask containing **1** (8.541 g, 13.5 mmol) in DMSO (100 mL). The mixture was stirred at 100 °C overnight, then diluted with hexane (300 mL) and washed with water (100 mL). The organic layer was dried over anhydrous $MgSO_4$, concentrated under reduced pressure, and the resulting residue dissolved in a mixed solvent of MeOH:CH_2Cl_2 (50 mL:20 mL). p-TsOH (300 mg, 1.58 mmol) was added and the mixture stirred at 23 °C for 1 h before concentrating it to 20 mL and then adding 200 mL CH_2Cl_2. The organic solution was washed with 0.05 M Na_2CO_3 solution (100 mL) and water (100 mL). Silica gel column chromatographic purification (2:1 hexane:Et_2O as eluent) of the organic residue gave **5** as an oil (6.952 g, 11.7 mmol, 87% yield). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H), 1.26 (m, 44H), 1.54 (m, 4H), 1.98 (m, 8H), 2.1 (s, 1H), 3.39-3.71 (m, 9H), 5.32 (m, 4H); ^{13}C NMR ($CDCl_3$): δ 14.3, 22.9, 26.3, 27.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 30.3, 32.2, 32.8, 63.3, 70.6, 71.2, 72.1, 78.5, 130.1, 130.2; CI $(M + H)^+$ calc'd 593, found 593.

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)ethene (6). $Pd(OAc)_2$ (217 mg, 0.97 mmol) and 1,10-phenanthroline (174 mg, 0.97 mmol) were added to a dry 100 mL Schlenk flask containing **5** (5.743 g, 9.68 mmol) in a mixture of ethyl vinyl ether (30 mL) and CH_2Cl_2 (15mL) under an atmosphere of Ar. The mixture was stirred at 70 °C for 20 h, prior to cooling to 0 °C and exposure to air. The mixture was filtered using a short alumina column and the filtrate concentrated. Silica gel column chromatographic purification (8:1 hexane:Et_2O as eluent) gave **6** as an oil (4.826 g, 8.80 mmol, 81%

yield) and recovered starting material **5** (1.098 g, 1.85 mmol, 19%). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H), 1.26 (m, 44H), 1.54 (m, 4H), 1.98 (m, 8H), 3.39-3.81 (m, 9H), 3.97 (dd, 1H, J =2, 7 Hz), 4.17 (dd, 1H, J = 2, 14 Hz), 5.32 (m, 4H), 6.45 (dd, 1H, J = 7, 14 Hz); ^{13}C NMR ($CDCl_3$): δ 14.3, 22.9, 26.3, 26.4, 27.4, 29.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 30.3, 32.2, 32.8, 68.3, 70.5, 70.9, 71.9, 77.0, 86.7, 130.0, 130.1, 152.2; CI $(M + H)^+$ calc'd 619, found 619.

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)-1-(ω-methoxy-polyethylene glycol[2000]oxy)-2-iodoethane (7). Compound **7** was prepared in 44% yield starting from **6** and mPEG2000 using the same procedure described for the synthesis of **3**. ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.51 (m, 4H), 1.97 (m, 8H), 3.21 (d, 2H, J = 5 Hz), 3.3-3.9 (m, ca. 200H), 4.68 (m, 1H), 5.32 (m, 4H).

1-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)-1-(ω-methoxy-polyethylene glycol[5000]oxy)-2-iodoethane (8). Compound **8** was prepared in 52% yield starting from **6** and mPEG5000 using same procedure described for the synthesis of **3**. ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.51 (m, 4H), 1.97 (m, 8H), 3.21 (d, 2H, J = 5 Hz), 3.3-3.9 (m, ca. 500H), 4.68 (m, 1H), 5.32 (m, 4H).

ST912. ST912 was prepared in 75% yield starting from **7** using the same procedure described for the synthesis of **ST902**. ^{1}H NMR ($CDCl_3$): δ 0.84 (t, 6H, J = 6 Hz), 1.24 (m, 44H), 1.51 (m, 4H), 1.97 (m, 8H), 3.12 (s, 2H), 3.3-3.9 (m, ca. 200H), 5.32 (m, 4H).

ST915. ST915 was prepared in 94% yield starting from **8** using the same procedure described for the synthesis of **ST902**. ^{1}H NMR ($CDCl_3$): δ 0.84 (t, 6H, J = 6 Hz), 1.24 (m, 44H), 1.51 (m, 4H), 1.97 (m, 8H), 3.12 (s, 2H), 3.3-3.9 (m, ca. 500H), 5.32 (m, 4H).

1,2-Dioleyl-*rac*-3-(2'-*t*-butyldimethylsilyloxy)acetate (9). Oxalyl chloride (1.02 mL, 11.7 mmol) was added to a flask containing bis(*t*-butyldimethylsilyloxy)glycolate (2.965g, 9.74 mmol) and 2

drops of DMF in CH_2Cl_2 (15 mL) at 0 °C. The mixture was stirred at 0 °C for 10 min and then at 23 °C for 2 h before concentrating the reaction mixture under reduced pressure and then dissolving the resulting residue with CH_2Cl_2 (20 mL). Compound **5** (4.813 g, 8.12 mmol) in pyridine was added at 0 °C and the reaction mixture stirred at 23 °C for 2 h. The mixture then was diluted with CH_2Cl_2 (50 mL) and washed with water (50 mL). Silica gel column chromatographic purification (8:1 hexane:Et_2O as eluent) of the organic residue gave **9** as an oil (4.021 g, 5.25 mmol, 65% yield). ^{1}H NMR ($CDCl_3$): δ 0.08 (s, 6H), 0.87 (m, 15H), 1.24 (m, 44H), 1.53 (m, 4H), 1.98 (m, 8H), 3.38-3.62 (m, 7H), 4.10-4.30 (m, 4H), 5.32 (m, 4H); ^{13}C NMR ($CDCl_3$): δ -5.2, 14.3, 18.6, 22.9, 26.0, 26.3, 27.4, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 30.3, 32.2, 32.8, 61.9, 64.5,70.4, 70.9, 72.0, 76.7, 130.0, 130.1, 171.8; ESI $(M + H)^+$ calc'd 765, found 765.

1,2-Di-O-oleyl-*rac*-glyceryloxy-3-(1'-*t*-butyldimethylsilyloxy-2'-propene) (10). $TiCl_4$ (2.14 mL, 19.5 mmol) was slowly added to THF (100 mL) at 0 °C and the solution became yellow. TMEDA (5.9 mL, 39 mmol) was added at 0 °C and the yellow solution became brown. Zn (2.868 mg, 43.8 mmol) containing 3% Pb was added and the solution stirred at 23 °C for 30 min (the brown solution became green). A mixture of **9** (3.731, 4.875 mmol) and CH_2Br_2 in THF (10 mL) was added at 0 °C and the mixture stirred at 23 °C for 1 d. Saturated K_2CO_3 (10 mL) was added at 0 °C and the mixture stirred at 0 °C for 15 min before filtering the reaction mixture to remove the hydroxy metalate salts and concentrating the filtrate under reduced pressure. Alumina column chromatographic purification (2:1 hexane:CH_2Cl_2 as eluent) of the organic residue gave **10** as an oil (1.543 g, 2.02 mmol, 42% yield). ^{1}H NMR (C_6D_6): δ 0.04 (s, 6H), 0.87 (m, 15H), 1.24 (m, 44H), 1.55 (m, 4H), 2.06 (m, 8H), 3.33 (t, 2H, J = 7 Hz), 3.49-3.56 (m, 4H), 3.72-3.89 (m, 3H), 4.08 (s, 1H), 4.12 (s, 2H), 4.40 (s, 1H), 5.46 (m, 4H); ^{13}C NMR (C_6D_6): δ -5.2, 14.3, 18.5, 23.1, 26.1, 26.6, 27.7, 29.6, 29.7, 29.8, 29.9, 30.0, 30.1, 30.2, 30.7, 32.3, 33.1, 63.6, 68.2, 70.8, 71.3, 71.8, 77.6, 81.2, 130.2, 162.4; ESI $(M + H)^+$ calc'd 763, found 763.

1,2-Di-O-oleyl-*rac*-glyceryloxy-2'-propen-1'-ol (11). TBAF (3.0 mL, 1.0 M in THF) was added to a flask containing **10** (763 mg, 1.00 mmol) in THF (10 mL). The mixture was stirred at 23 °C for 2 h and filtered using a short alumina column (Et_2O eluent). The solution was concentrated and the residue purified via silica gel column chromatography (1:1 hexane:Et_2O as eluent) to give **11** as an oil (594 mg, 0.92 mmol, 92% yield). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 7 Hz), 1.25 (m, 44H), 1.52 (m, 4H), 1.98 (m, 8H), 2.17 (s, 1H), 3.41 (t, 2H, J = 7 Hz), 3.49 (d, 2H, J = 7 Hz), 3.55 (t, 2H, J = 7 Hz), 3.65-3.83 (m, 3H), 4.01 (m, 3H), 4.14 (s, 1H), 5.32 (m, 4H); ^{13}C NMR ($CDCl_3$): δ 14.1, 22.6, 26.0, 26.1, 27.2, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 31.9, 32.6, 63.6, 67.6,70.4, 70.7, 71.7, 76.8, 82.3, 129.8, 129.9, 161.3; CI $(M + H)^+$ calc'd 649, found 649.

1,2-Di-O-oleyl-*rac*-glyceryl-3-(2'-propenyl-1'-(4''-nitrophenyl)carbonate) (12). Triethylamine (413 mg, 4.1 mmol) was added to **11** (531 mg, 0.818 mmol) in THF (5 mL). 4-Nitrophenyl chloroformate (247 mg, 1.23 mmol) was added and the mixture stirred for 1 h. The reaction mixture was concentrated and the resulting residue purified by silica gel column chromatography (4:1 hexane:Et_2O as eluent) to give **12** as an oil (612 mg, 0.752 mmol, 92% yield). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H, J = 6 Hz), 1.24 (m, 44H), 1.53 (m, 4H), 1.97 (m, 8H), 3.43 (t, 2H, J = 7 Hz), 3.49 (d, 2H, J = 7 Hz), 3.56 (t, 2H, J = 7 Hz), 3.68-3.88 (m, 3H), 4.21 (d, 1H, J = 3 Hz), 4.30 (d, 1H, J = 3 Hz), 4.67 (s, 2H), 5.31 (m, 4H), 7.37 (d, 2H, J = 9 Hz), 8.24 (d, 2H, J = 9 Hz); ^{13}C NMR ($CDCl_3$): δ 14.0, 22.6, 26.0, 27.2, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 30.0, 31.9, 32.5, 68.0, 68.9, 70.2, 70.8, 71.7, 76.7, 86.3, 121.7, 125.2, 129.7, 129.9, 145.4, 152.2, 155.5, 155.8; ESI calc'd $(M + Na)^+$ 836, found 836.

ST502. Triethylamine (230 mg, 2.3 mmol), **12** (380 mg, 0.467 mmol), and mPEGamine2000 (623 mg, 0.311 mmol) were combined in DMF (10 mL) and stirred at 23 °C for 12 h. The reaction mixture was concentrated and purified by silica gel column chromatography (10:1 CH_2Cl_2:MeOH as eluent) to give **ST502** (692 mg, 0.258 mmol, 83% yield). ^{1}H NMR ($CDCl_3$): δ 0.80 (t, 6H, J = 6 Hz), 1.20

(m, 44H), 1.48 (m, 4H), 1.93 (m, 8H), 3.2-3.8 (m, ca. 200H), 4.02 (d, 1H, J = 2 Hz), 4.10 (d, 1H, J = 2 Hz), 4.41 (s, 2H), 5.31 (m, 5H).

ST505. ST505 was prepared as described above in 71% yield using mPEGamine5000. ^{1}H NMR ($CDCl_3$): δ 0.82 (t, 6H, J = 6 Hz), 1.23 (m, 44H), 1.50 (m, 4H), 1.95 (m, 8H), 3.2-3.8 (m, ca. 500H), 4.05 (d, 1H, J = 2 Hz), 4.13 (d, 1H, J = 2 Hz), 4.43 (s, 2H), 5.34 (m, 5H).

Dimethyl 2-(1',2'-di-O-oleyl-*rac*-glyceryloxy)malonate (13). $Rh_2(OAc)_4$ (17 mg, 0.038 mmol) was added to a flask containing **5** (2.234 g, 3.77 mmol) and dimethyl diazomalonate in benzene (50 mL). The mixture was heated at reflux for 3 h before cooling, concentrating the reaction mixture, and purifying the residue by silica gel column chromatography (1:1 hexane:Et_2O as eluent) to give **13** as an oil (1.832 g, 2.53 mmol, 67% yield). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 7 Hz), 1.24 (m, 44H), 1.51 (m, 4H), 1.97 (m, 8H), 3.4-3.8 (m, 9H), 3.78 (s, 6H), 4.65 (s, 1H), 5.31 (m, 4H); ^{13}C NMR ($CDCl_3$): δ 14.0, 22.6, 26.0, 26.1, 27.2, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 29.8, 29.9, 30.0, 31.5, 31.9, 32.6, 52.7, 70.0, 70.6, 71.5, 71.7, 77.9, 79.5, 129.8, 129.9, 166.9; ESI $(M + H)^+$ calc'd 723, found 723.

Dimethyl 2-(1',2'-di-O-oleyl-*rac*-glyceryloxy)-2-dimethylaminomethyl malonate (14). Eschenmoser's salt (1.454 g, 7.86 mmol) was added to a flask containing **13** (2.841 g, 3.93 mmol) and triethylamine (1.1 mL, 7.86 mmol) in CH_2Cl_2 (50 mL). The mixture was stirred at 23 °C for 12 h before washing the reaction mixture with water (2 x 20 mL) and concentration under reduced pressure. The resulting residue was purified by silica gel column chromatography (1:1 hexane:Et_2O as eluent) to give **14** as an oil (2.970 g, 3.81 mmol, 97% yield). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H, J = 7 Hz), 1.25 (m, 44H), 1.52 (m, 4H), 1.98 (m, 8H), 2.28 (s, 6H), 2.91 (s, 2H), 3.35-3.65 (m, 9H), 3.75 (s, 6H), 5.32 (m, 4H); ^{13}C NMR ($CDCl_3$): δ 14.0, 22.6, 26.0, 26.1, 27.2, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 30.0, 31.9, 32.5, 47.2, 52.3, 62.1, 66.7, 70.6, 70.9, 71.5, 77.7, 86.5, 129.8, 129.9, 168.3; ESI $(M + H)^+$ calc'd 780, found 780.

Methyl 2-(1',2'-di-O-oleyl-*rac*-glyceryloxy)acrylate (15). MeI (1.36 mL, 21.8 mmol) was added to a flask containing **14** (3.01 g, 3.86 mmol) in CH_2Cl_2 (50 mL). The mixture was heated at reflux for 12 h before cooling, concentration under reduced pressure and addition of CH_3CN (50 mL). Then, the reaction mixture was heated at reflux for 1 d before concentration under reduced pressure, and purification of the organic residue by silica gel column chromatography (1:1 hexane:Et_2O as eluent) to give **15** as an oil (2.012 g, 2.97 mmol, 99% yield). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H), 1.24 (m, 44H), 1.52 (m, 4H), 1.98 (m, 8H), 3.35-3.90 (m, 12H), 4.62 (d, 1H, J = 2 Hz), 5.32 (m, 5H); ^{13}C NMR ($CDCl_3$): δ 14.0, 22.6, 26.0, 27.1, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 30.0, 31.8, 32.5, 52.1, 68.8, 70.2, 70.9, 71.7, 76.5, 94.3, 129.8, 129.9, 151.2, 163.4; ESI $(M + H)^+$ calc'd 677, found 677.

2-(1',2'-Di-O-oleyl-*rac*-glyceryloxy)acrylic acid (16). NaOH solution (2.5 mL at 2.0 M) was added to a flask containing **15** (1.107 g, 1.635 mmol) in a mixed solvent of THF (20 mL) and H_2O (10 mL). The reaction mixture was heated at reflux for 3 h and concentrated under reduced pressure. H_2O (20 mL) and Et_2O (20 mL) were added and the pH of the aqueous layer was adjusted to around 4 by slow addition of 1.0 M HCl. The organic layer was concentrated and the resulting residue purified by silica gel column chromatography (Et_2O as eluent) to give **16** as an oil (770 mg, 1.16 mmol, 71% yield). ^{1}H NMR ($CDCl_3$): δ 0.85 (t, 6H, J = 7 Hz), 1.25 (m, 44H), 1.54 (m, 4H), 1.98 (m, 8H), 3.35-3.90 (m, 9H), 4.71 (d, 1H, J = 2 Hz), 5.31 (m, 4H), 5.46 (d, 1H, J = 2 Hz); ^{13}C NMR ($CDCl_3$): δ 14.0, 22.6, 26.0, 26.1, 27.2, 29.1, 29.2, 29.3, 29.4, 29.5, 29.6, 29.7, 30.0, 31.9, 32.5, 69.0, 70.0, 70.9, 71.8, 76.4, 76.0, 129.8, 129.9, 150.4, 166.2; ESI $(M + H)^+$ calc'd 663, found 663.

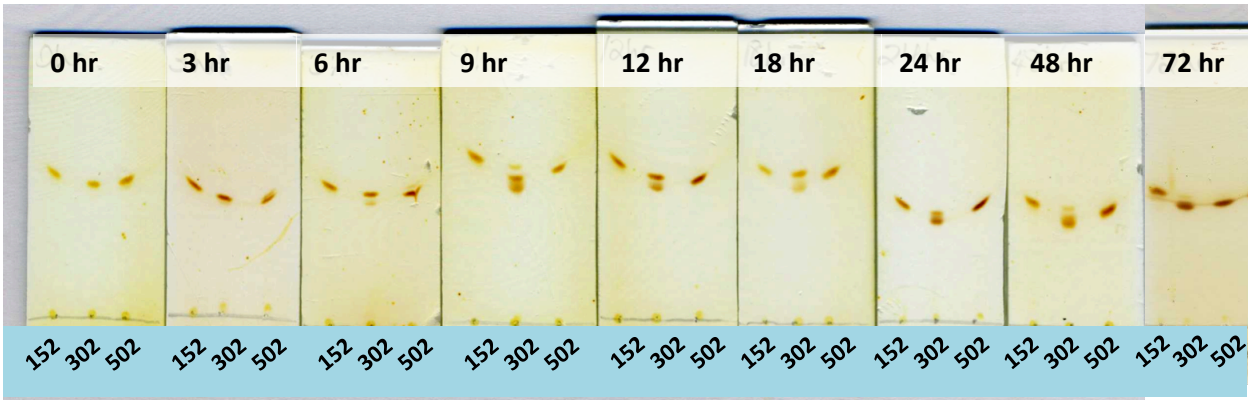
ST152. DCC (115 mg, 0.558 mmol), **16** (493 mg, 0.744 mmol), mPEGamine2000 (743 mg, 0.372 mmol), and DMAP (2 mg) were combined in CH_2Cl_2 (30 mL) and stirred at 23 °C for 12 h. The reaction mixture was filtered to remove the urea byproduct, the filtrate concentrated under reduced pressure, and the organic residue purified by silica gel column chromatography (10:1 CH_2Cl_2:MeOH as

eluent) to give **ST152** (603 mg, 0.227 mmol, 61% yield). ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.54 (m, 4H), 1.98 (m, 8H), 3.3-3.9 (m, ca. 200H), 4.44 (d, 1H, J = 2 Hz), 5.32 (m, 4H), 5.37 (d, 1H, J = 2 Hz), 6.94 (t, 1H, J = 5 Hz).

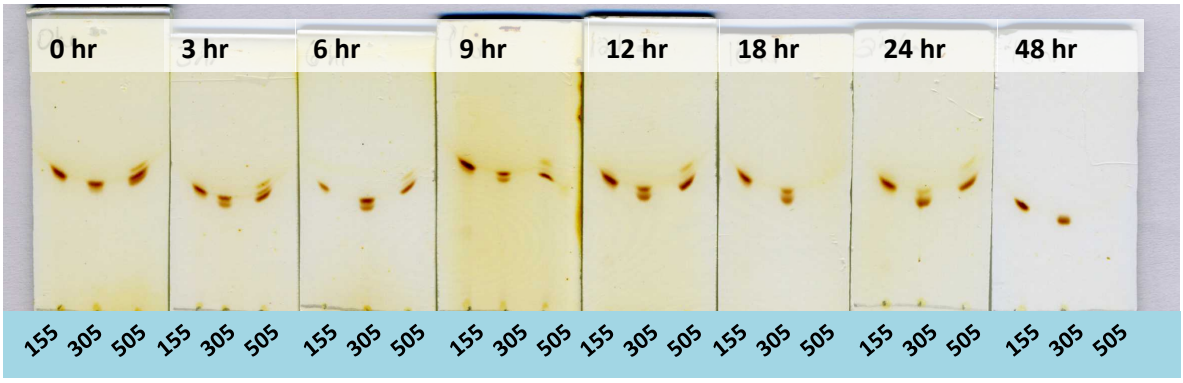
ST155. **ST155** was prepared as described above in 70% yield using mPEGamine5000. ^{1}H NMR ($CDCl_3$): δ 0.86 (t, 6H, J = 6 Hz), 1.25 (m, 44H), 1.54 (m, 4H), 1.98 (m, 8H), 3.3-3.9 (m, ca. 500H), 4.44 (d, 1H, J = 2 Hz), 5.32 (m, 4H), 5.37 (d, 1H, J = 2 Hz), 6.94 (t, 1H, J = 5 Hz).

5. Hydrolysis Studies by TLC

TLC Analysis of mPEG2000-VE-Lipid Hydrolysis



TLC Analysis of mPEG5000-VE-Lipid Hydrolysis



6. Hydrolysis Studies by HPLC-CAD

Figure S6. HPLC Analysis of ST912 Hydrolysis

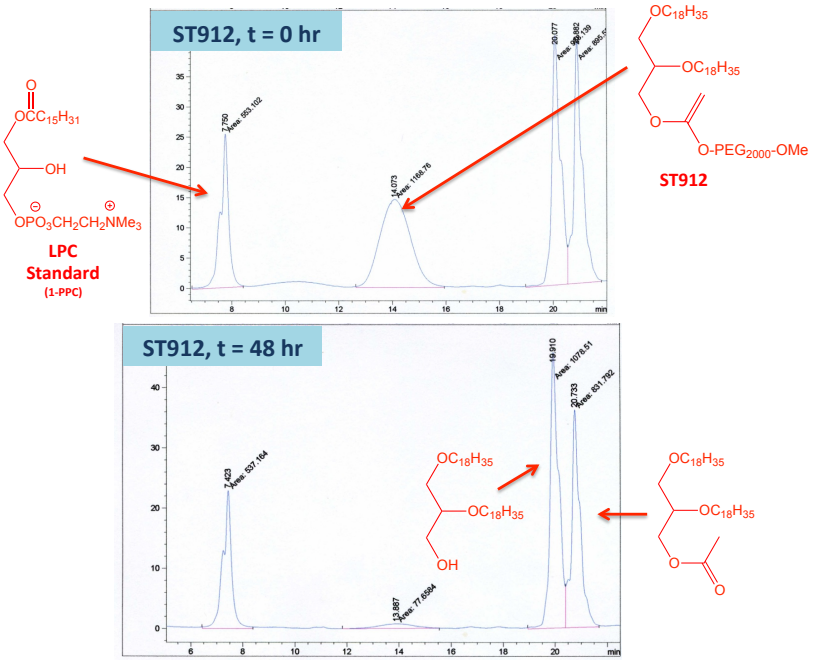


Figure S7. HPLC Analysis of ST502 Hydrolysis

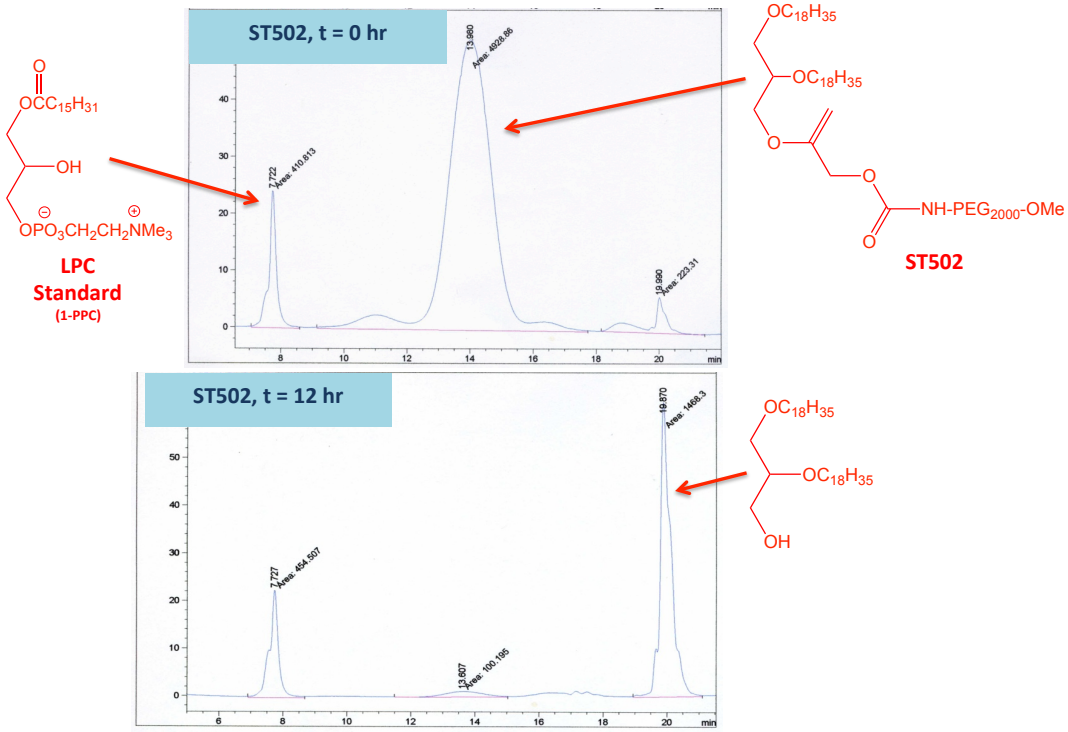


Figure S8. **HPLC Analysis of ST302 Hydrolysis**

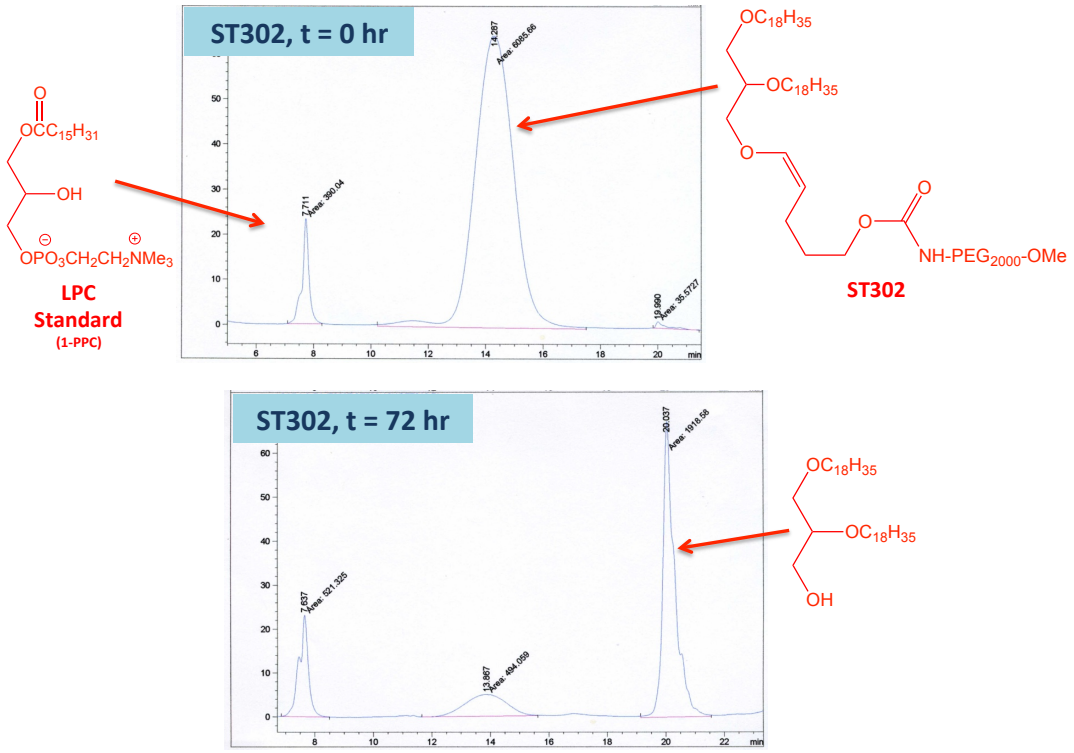


Figure S9. **HPLC Analysis of ST152 Hydrolysis**

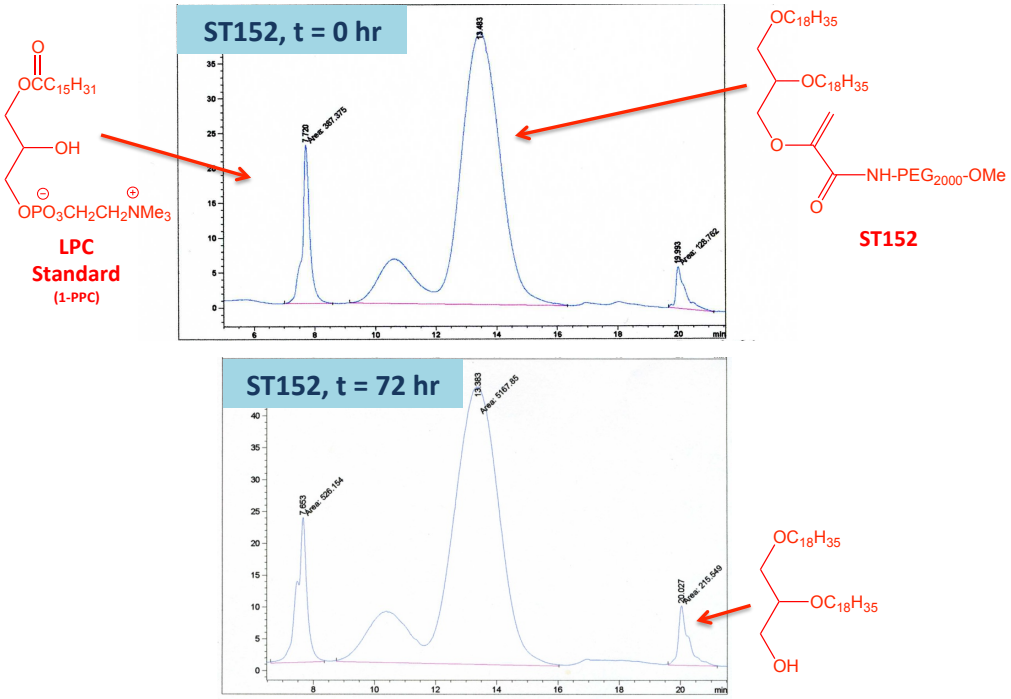
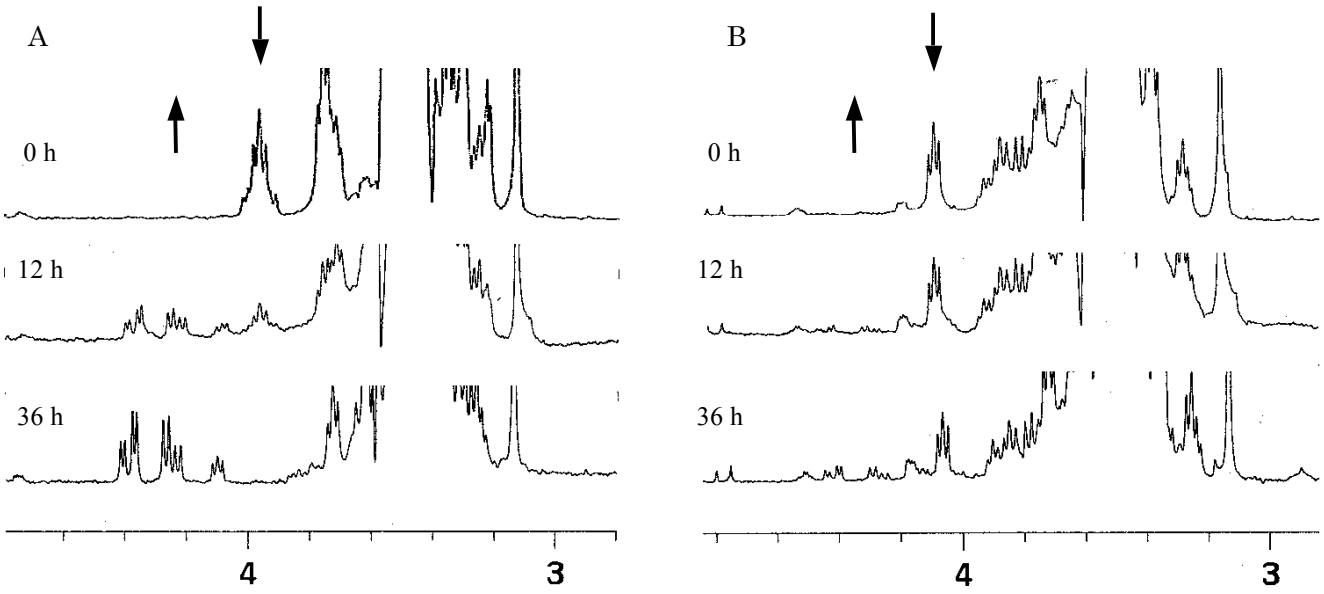


Figure S10. Table of Intrinsic Second Order Rate Constants and Proton Affinities Calculated by DFT

mPEG-VE-Lipid	Pseudo-First Order Rate Constant (s-1)	Intrinsic Second Order Rate Constant (M-1*s-1)	log k (M^{-1} s^{-1})
ST912	5.2×10^{7}	5.2×10^{4}	4.71
ST502	7.3×10^{-1}	730	2.86
ST505	2.4×10^{-1}	240	2.38
ST302	1.4×10^{-5}	1.4×10^{-2}	-1.85
ST305	8.3×10^{-6}	8.3×10^{-3}	-2.08
ST152	5×10^{-9}	5×10^{-6}	-5.30
ST155	2.5×10^{-8}	2.5×10^{-5}	-4.60

Figure S11. ^{1}H NMR of ST912 and ST902 hydrolysis proceeding in d_6-benzene with <5% water added (pH 7).



7. Calcein Release Rates for Various mPEG-VE-lipid Loadings in DOPE Liposomes

Figure S12. Release rates from DOPE:ST152, DOPE:ST155 and DOPE:ST305 liposomes as a function of pH.

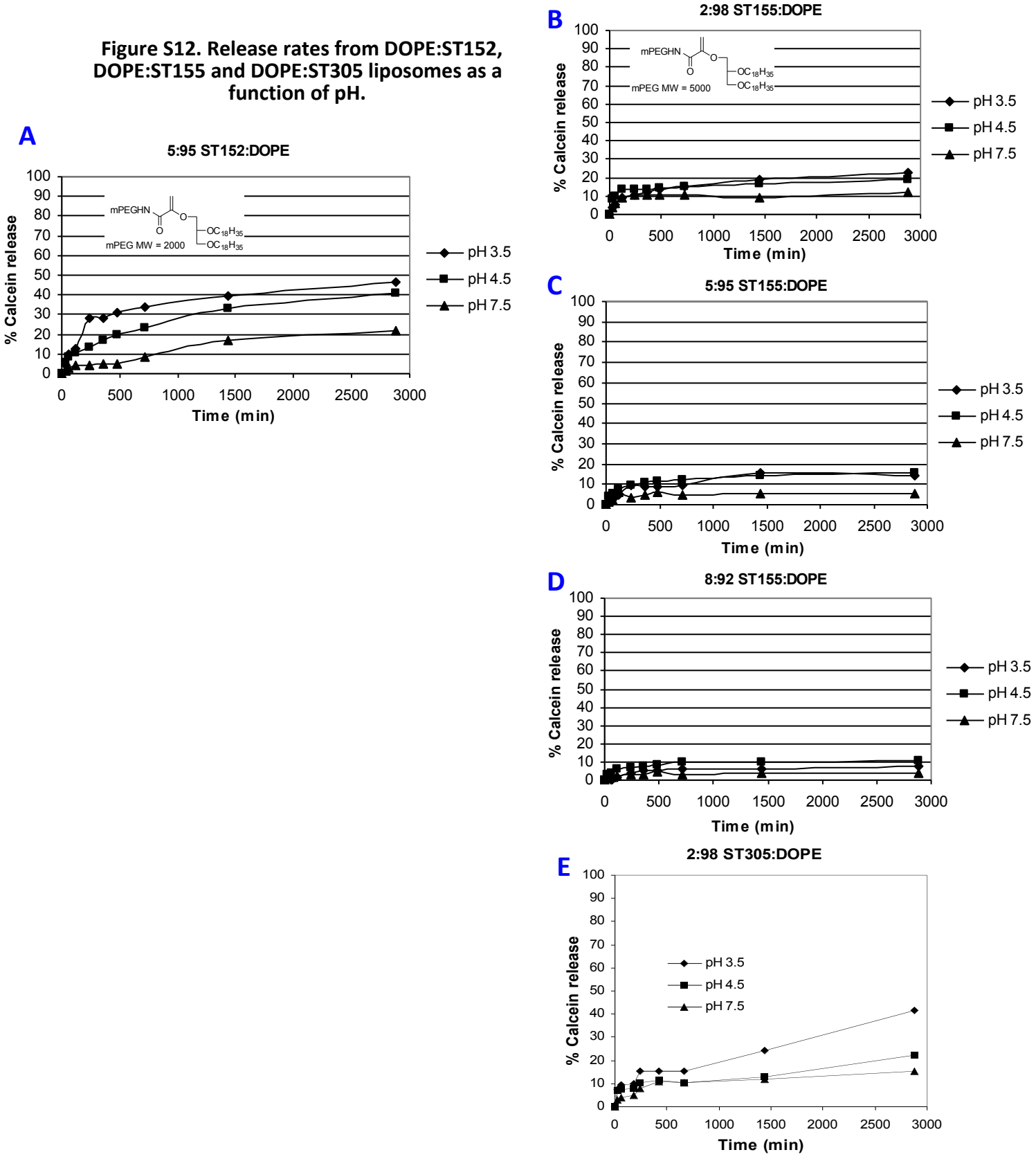
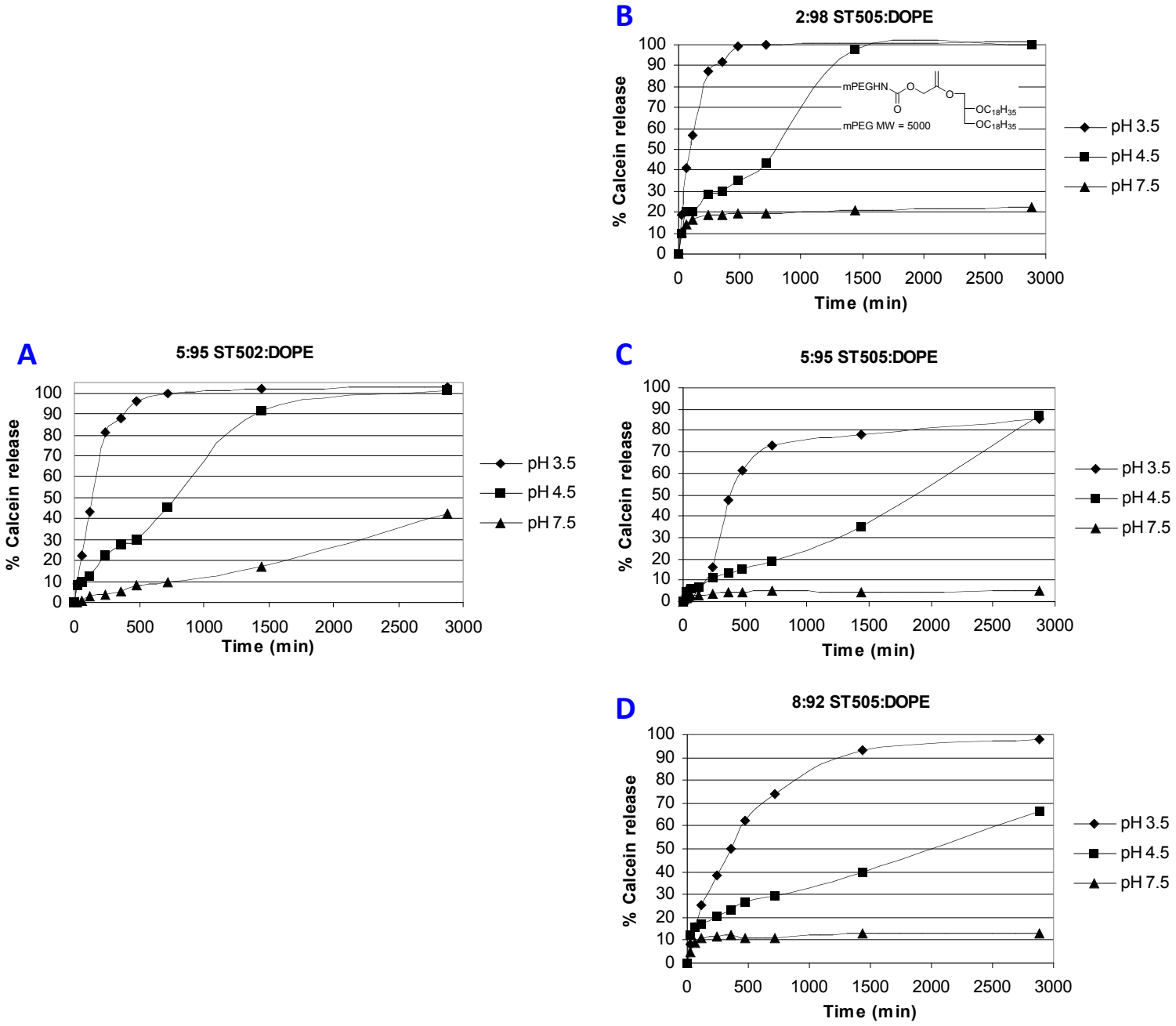


Figure S13. Calcein release rates from DOPE:ST502 and DOPE:ST505 liposomes as a function of pH.



VI. References

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