Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supporting Information

Voltage-dependent structural changes of the membrane-bound anion channel hVDAC1 probed by SEIRA and electrochemical impedance spectroscopy

Jacek Kozuch[a*], Conrad Weichbrodt[b], Diego Millo[c], Karin Giller[d], Stefan Becker[d], Peter Hildebrandt[a*], Claudia Steinem[b]

[a] Technische Universität Berlin, Institut für Chemie, Straße des 17. Juni 135, D-10623 Berlin, Germany. [b] Georg-August-Universität Göttingen, Institut für Organische und Biomolekulare Chemie, Tammannstrasse 2, 37077 Göttingen, Germany. [c] Vrije Universiteit Amsterdam, Biomolecular Spectroscopy/LaserLaB Amsterdam, De Boelelaan 1083, 1081 HV Amsterdam, The Netherlands. [d] Max-Planck Institut für Biophysikalische Chemie, Am Faßberg 11, D-37077 Göttingen, Germany.

* correspondence address.

Content:

1. Equivalent Circuit

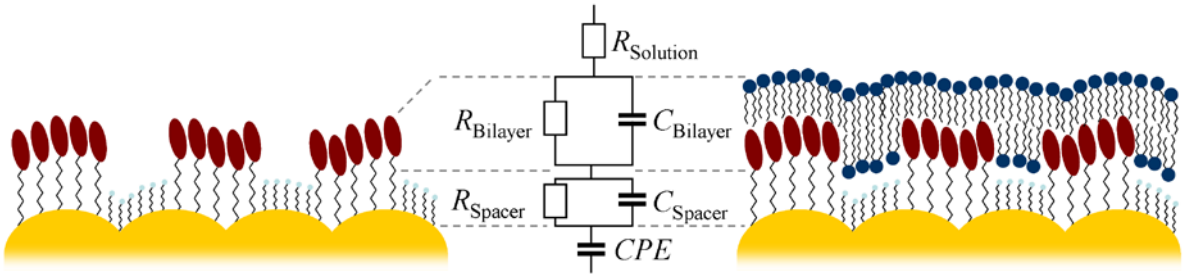


Figure S1. Equivalent circuit used for the analysis of the impedance data. The $R_{Spacer}C_{Spacer}$-element describes the electric properties of the „spacer“ molecules, i.e. the 6MH and the triethyleneglycol-chain of CPEO3. The cholesteryl-headgroup region and the POPC-bilayer region is described by the $R_{Bilayer}C_{Bilayer}$-element. The constant-phase element (CPE) accounts for the non-ideal electrical behaviour of the system visible as a frequency-dependent tail at low frequencies in the impedance spectra. The CPE used in this work is defined as follows:

$$Z_{CPE} = \frac{1}{Q(i\omega)^{\alpha}}$$

2. Single-channel recordings in free-standing bilayers

To prove the functionality of the recombinant hVDAC1 preparations, electrophysiological experiments were carried out using the Port-a-Patch® technique. hVDAC1 was reconstituted into giant unilamellar vesicles composed of DPhPC/cholesterol (9:1) and spread on a small aperture in glass. Electrical recordings were performed using the Axopatch 200B integrating patch clamp amplifier equipped with a CV 203BU head stage. Resulting signals were recorded with sampling rates of 10 or 50 kHz, pre-filtered with a 1 kHz 4-pole-Bessel filter (-80 dB/decade), digitized *via* a 1322A Digidata 16-bit digitizer and evaluated using the pCLAMP 9 software package (Axon Instrumens, Union City, CA, USA).

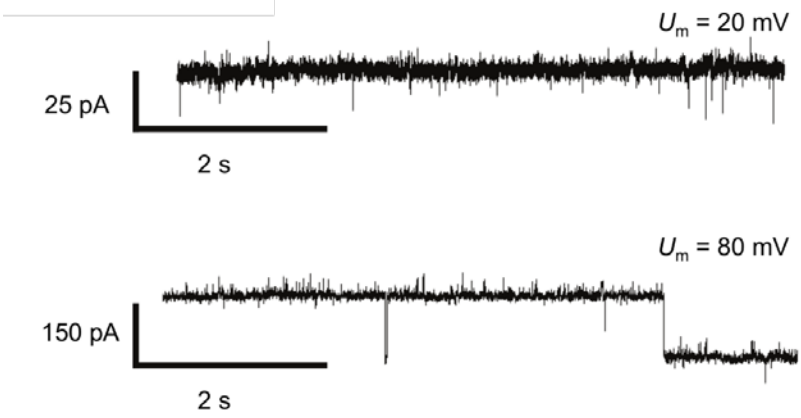


Figure S2. Representative current traces of a single hVDAC1 molecule reconstituted into a solvent-free membrane composed of DPhPC/cholesterol (9:1) bathed in 1 M KCl, 1 mM $CaCl_2$, 5 mM HEPES, pH 7.4 and recorded at the transmembrane potentials as indicated. The drop in current at U_m = + 80 mV corresponds to a decrease in conductance of 2.0 nS.

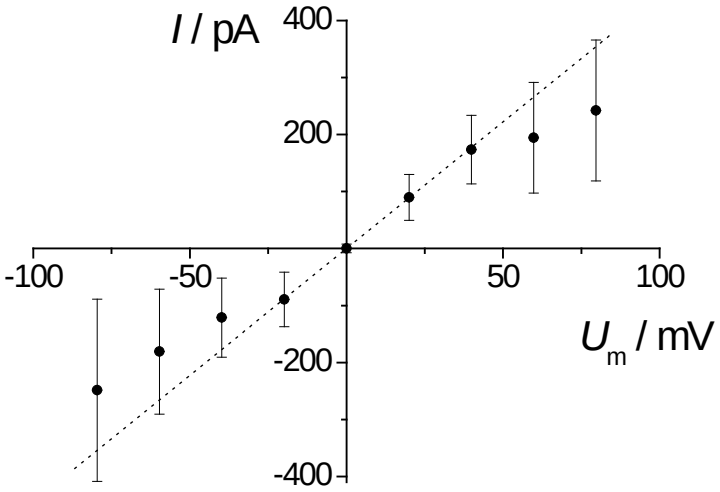


Figure S3. Current-voltage relationship of a single hVDAC1 molecule. For each potential, the mean value of the recorded current obtained from a membrane with a single inserted hVDAC1 molecule averaged over 15 experiments is plotted with its standard deviation of the mean. The dotted line marks the conductivity of the fully open state of hVDAC1 with G_o = 4 nS.

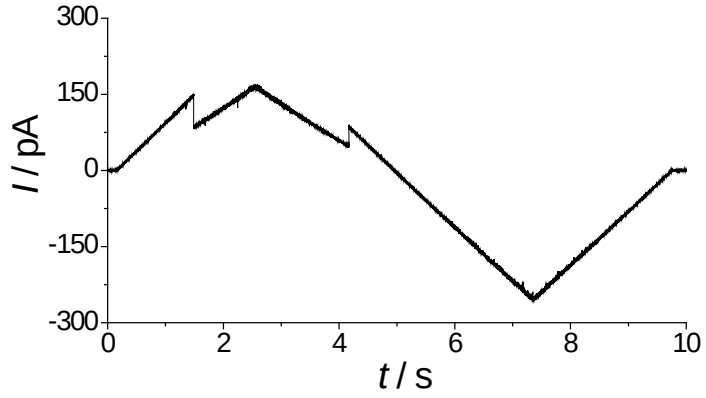


Figure S4. Representative current trace of a single hVDAC1 channel reconstituted into a solvent-free membrane composed of DPhPC/cholesterol (9:1) bathed in 1 M KCl, 1 mM $CaCl_2$, 5 mM HEPES, pH 7.4. A 100 mHz voltage wave with maximum amplitude of U_m = ±60 mV was applied leading to closing events at elevated potentials. The difference between the open state conductivity of G_o = 4.5 nS and a closed state conductivity of G_c = 2.6 nS appears as a sharp drop in the current signal at elevated positive potentials.

3. Potential-dependent current traces

Potential-dependent current traces were recorded, controlled by the GPES software and carried out under constant purging with Argon, by applying potentials ranging from –80 to +80 mV (with respect to the open circuit potential E_{OCP}) in 20 mV-steps and a 5 min residence time at each potential.

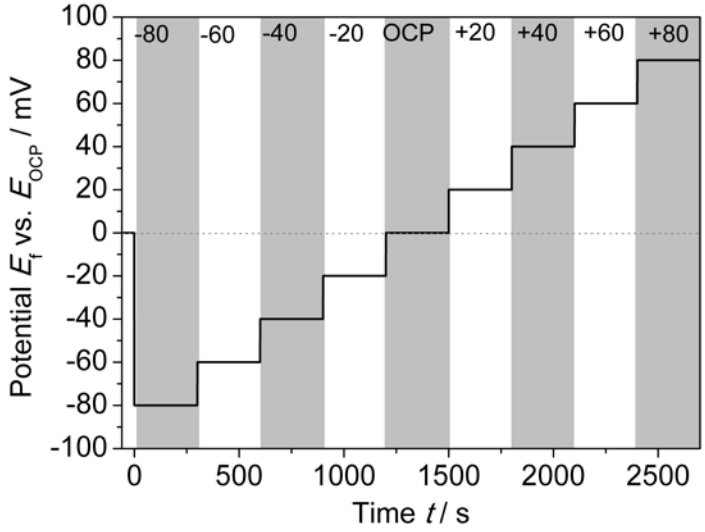


Figure S5. Shape of the voltage steps during the recording of potential-dependent currents.

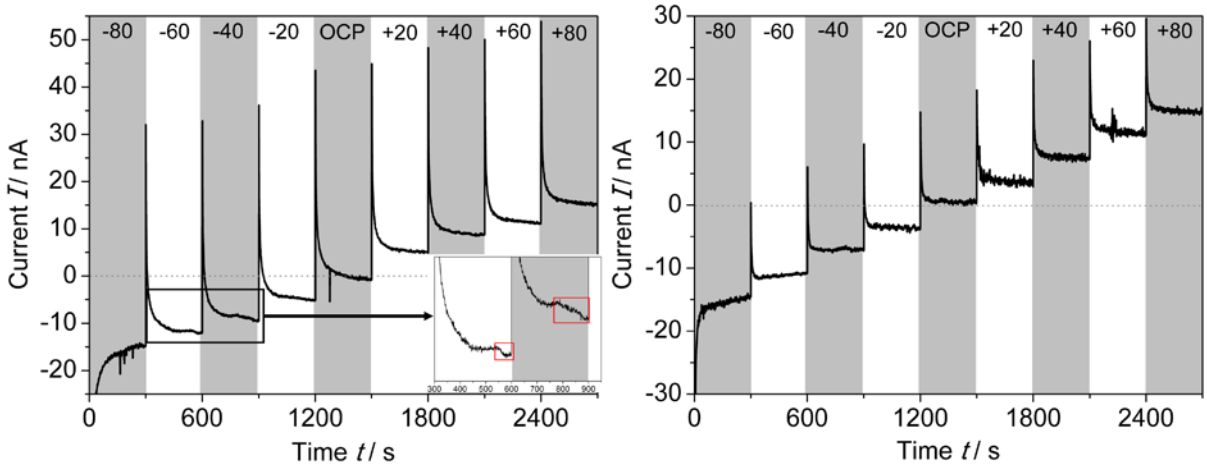


Figure S6. Potential-dependent current traces of hVDAC1-tBLMs (left) and pure tBLMs (right), following a sequence of potential steps from the E_{OCP} to a final potential E_f (vs. E_{OCP}). The top row shows the final potentials applied during the marked interval in respect to the E_{OCP}. The inset in the left plot highlights the small deviations from the quasi-stationary currents.

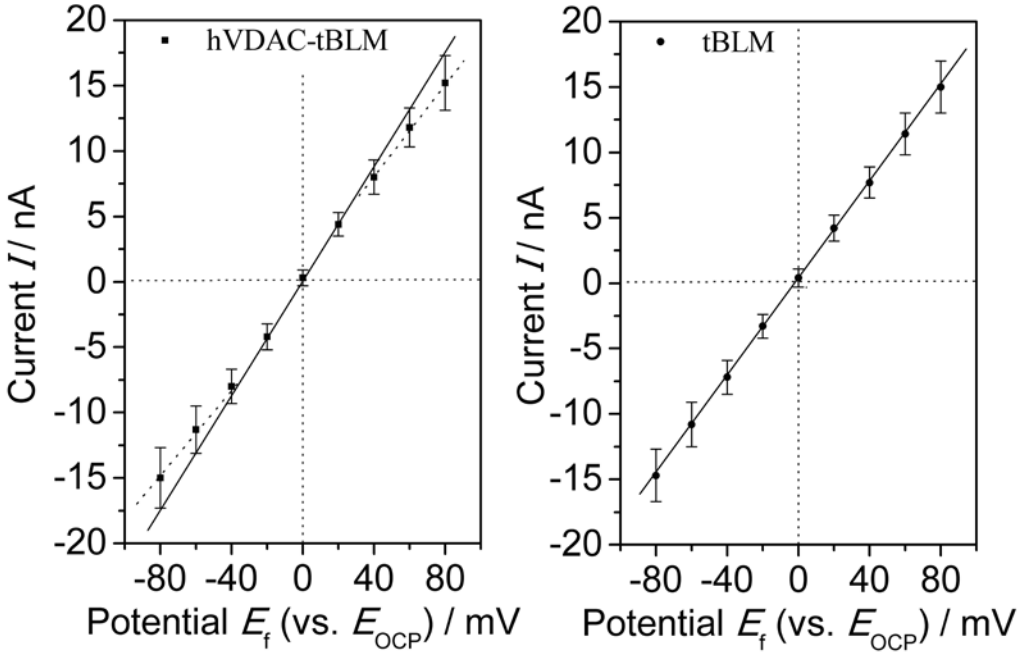


Figure S7. Currents of the hVDAC1-tBLM (left) and the pure tBLM (right) systems extracted from potential-dependent current traces in Figure S6. The data represent averages over three measurements. The solid lines represent linear fits to the data in the range between –20 and +20 mV (open state of hVDAC1). The dashed lines indicate the trend at potentials above |±20 mV|, where the channel tends to "close.

Each potential step caused an instantaneous response (see Figure S6) of the current that decayed biexponentially with time constants of ca. 2 and 30 s. Subsequently, the current slowly decayed against a current of zero on the long minute time-scale. The decay was not completed prior to the next potential step, but may be considered as quasi-stationary with respect to the fast decay components. These quasi-stationary currents increase after each potential step, superimposing the rapid current changes by a linear slope.

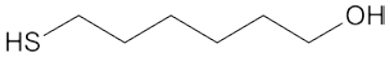
In the case of hVDAC-tBLMs (Figure S6, left), qualitatively similar current traces are observed as for pure tBLMs (Figure S6, right) albeit with higher maximum currents and faster decays. However, a small but clearly detectable deviation from the quasi-stationary current was observed at negative potentials (in particular at -60 mV and -40 mV; see inset in Figure S6, left), decreasing the current slightly just prior to the next potential step. This was only observed in the presence of hVDAC in the tBLM system. At positive E_f (vs. E_{OCP}) such an additional current decay was not observed during the residence time at the final potential.

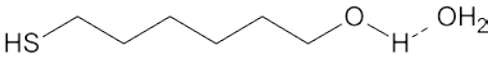
The quasi-stationary currents are plotted against the applied potential in Figure S7. In the absence of hVDAC, the plot shows a linear behaviour corresponding to one resistance over the potential range of –80 to +80 mV (Figure S7, right). However, when hVDAC was present in the tBLM a slight deviation from that linearity is observed (Figure S7, left).

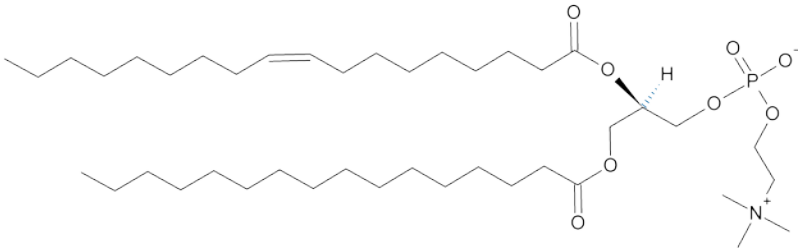
4. Computational calculations

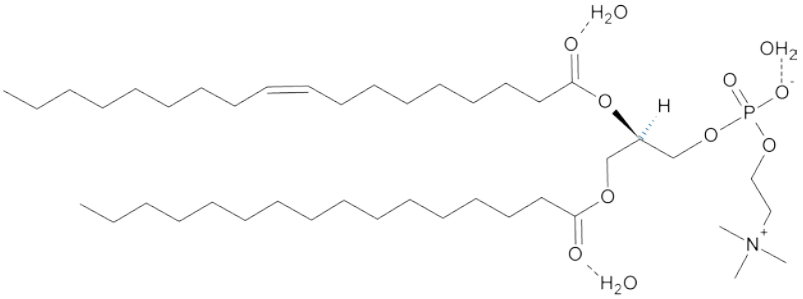
For a better understanding of the spectra of pure tBLMs, IR spectra of CPEO3, 6MH, and POPC were calculated using density functional theory. Geometry optimization and vibrational analysis were performed on the BP86[1,2] level of theory using Gaussian 09. For C, H, N, and O atoms the 6-31g* basis set, for the heavier atoms S, and P the TZVP basis set was employed.[3,4] For POPC, the D2 version of Grimme's dispersion model[5] was introduced to prevent the acyl chains to drift apart and thus to obtain a more relevant conformation.

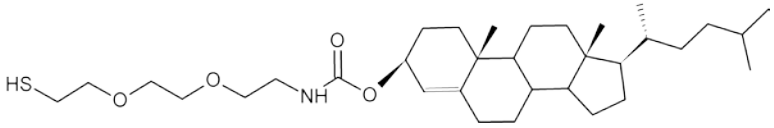
In addition, for all three molecules H_2O-hydrogen-bonded structures were calculated to approximate the effect of hydrogen bonding. In total, the IR spectra of the following structures were calculated:

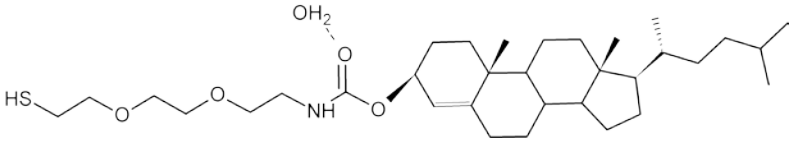
a) 6MH: 

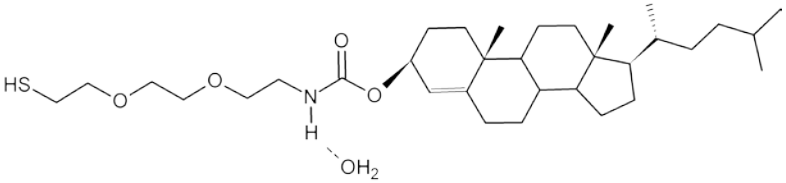
b) 6MH-H_2O: 

c) POPC: 

d) POPC-H_2O: 

e) CPEO3: 

f) CPEO3-H_2O (CO): 

g) CPEO3-H_2O (NH): 

Due to the flat potential energy surface, resulting from the high flexibility of the acyl chains and the triethoxyl chain, for some of the POPC and CPEO3 structures imaginary frequencies in the range of 0 to -20 cm^{-1} could not be avoided. Section 2 provides a summary of the results of the calculations and a comparison with one of the potential dependent difference spectra of the pure tBLMs. In section 3, the Cartesian coordinates, the total charge, and the spin state of all optimized geometries are given.

[1] J. P. Perdew, *Phys. Rev. B* **1986**, *33*, 8822–8824.

[2] A. D. Becke, *Phys. Rev. A* **1988**, *38*, 3098–3100.

[3] F. Weigend, R. Ahlrichs, *Phys. Chem. Chem. Phys.* **2005**, *7*, 3297–3305.

[4] Y. Rippers, T. Utesch, P. Hildebrandt, I. Zebger, M. A. Mroginski, *Phys. Chem. Chem. Phys.* **2011**, *13*, 16146–16149.

[5] S. Grimme, *J. Comput. Chem.* **2006**, *27*, 1787–1799.

5. Calculated IR spectra and normal modes

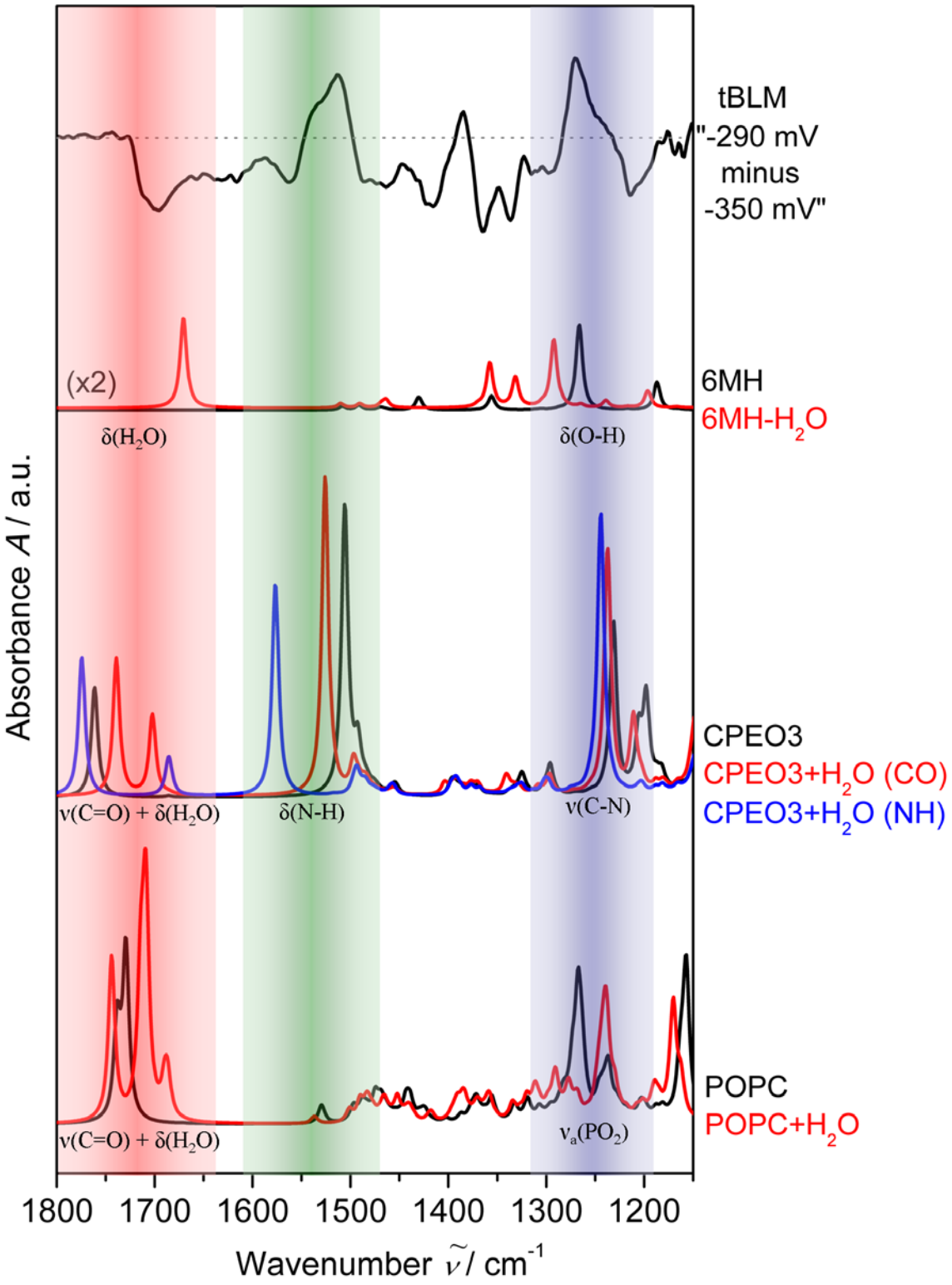


Figure S8. Comparison of the potential-dependent tBLM spectrum (-290 mV minus -350 mV) with the calculated spectra of 6MH, CPEO3, and POPC (HWHM = 4 cm^{-1}). The spectral regions discussed in the manuscript are marked in red, green, and blue and assigned to the main vibrations of the components. The assignment of the most intense bands are listed in tables S1-S3 (*vide infra*).

Table S1. Calculated vibrational frequencies of 6MH in the region (1800-1100 cm^{-1}) and assignment the respective vibrational modes. Frequencies are given in cm^{-1}; intensities are listed in brackets and are given km/mol. Blue frequencies refer to the regions highlighted in Figure S8.

	6MH	**6MH + H_2O**
δ(OH) + δ(CH_2) wagg.	1430 (11)	1464 (6)
δ(CH_2) wagg.	1356 (12)	1358 (37)
δ(OH) + δ(CH_2) wagg.	-	1332 (25)
δ(OH) + δ(CH_2) wagg.	**1266** (69)	**1292** (56)
δ(OH) + δ(CH_2) wagg.	1187 (23)	1196 (14)

Table S2. Calculated vibrational frequencies of POPC in the region (1800-1000 cm^{-1}) and assignment the respective vibrational modes. Frequencies are given in cm^{-1}; intensities are listed in brackets and are given km/mol. Blue and red frequencies refer to the regions highlighted in Figure S8. For a detailed listing of the vibrations of the acyl chain in particular, see S. Krishnamurty, M. Stefanov, T. Mineva, S. Bégu, J. M. Devoisselle, A. Goursot, R. Zhu, D. R. Salahub, *J. Phys. Chem. B* 2008, *112*, 13433–42.

	POPC	**POPC + 3 H_2O**
Head group region		
ν(C=O)	**1738** (155)	**1714** (214)
ν(C=O)	**1729** (287)	**1709** (368)
$\delta_a(CH_3)$ head + $\delta_a(CH_2)$ head	1530 (30), 1497 (22)	1537 (12), 1501 (20)
$\delta_a(CH_3)$ head	1475 (39), 1468 (21)	1482 (22), 1466 (23)
$\delta_a(CH_3)$ head + $\delta_a(CH_2)$ head + $\delta_a(CH_2)$ chains	1443 (24), 1439 (29)	1452 (29), 1439 (18)
δ(CH_3) head wagg. + $\nu_a(PO_2)$	**1273** (53)	**1269** (22)
$\nu_a(PO_2)$	**1267** (184)	**1238** (118)
ν(CO) chain	1163 (82), 1157 (254), 1093 (51)	1170 (183), 1162 (59), 1092 (41)
δ(CH_3) head wagg.	1134 (14)	1095 (53)

Table S3. Calculated vibrational frequencies of CPEO3 in the region (1800-1000 cm^{-1}) and assignment the respective vibrational modes. Frequencies are given in cm^{-1}; intensities are listed in brackets and are given km/mol. Blue, green, and red frequencies refer to the regions highlighted in Figure S8.

	CPEO3	**CPEO3 + H_2O (CO)**	**CPEO3 + H_2O (NH)**
Linker region			
ν(C=O)	**1761** (183)	**1739** (228)	**1774** (231)
δ(NH) + ν(C-N)	**1506** (480)	**1526** (529)	**1577** (354)
δ(CH_2)	1492 (50)	1497 (30), 1491 (8)	1494 (25)
δ(CH_2) wagg.	1397 (11), 1325 (34)	1404 (19), 1341 (25)	1397 (8), 1325 (12)
δ(CH_2) twist	1296 (50)	1296 (33)	1302 (18), 1298 (14)
ν(CN) + δ(NH) + δ(CH_2) twist	**1231** (286), **1206** (83)	**1238** (316), **1236** (107), 1203 (13)	**1244** (453), 1242 (20), 1203 (10)
ν(CN) + δ(NH)	**1198** (150)	**1211** (85), 1210 (36)	
ν(CO) + ν(CN) + δ(NH)	1149 (24), 1140 (71), 1129 (543), 1122 (109), 1113 (39)	1149 (57), 1145 (57), 1131 (351), 1123 (177), 1114 (12)	1150 (15), 1141 (184), 1122 (196), 1116 (47), 1111 (205)
δ(CH_2) rock. + δ(NH)	1120 (63)	1125 (37),	1128 (99)
Cholesteryl-head region			
$\delta_a(CH_3)$ + $\delta_a(CH_2)$	1493 (21), 1483 (9), 1394 (14)	1497 (18), 1493 (10), 1483 (10), 1391 (14)	1496 (13), 1492 (11), 1484 (13), 1392 (22)
δ(CH_2) + δ(CH) wagg.	1377 (10), 1370 (10)	1377 (11), 1370 (15)	1376 (9),
δ(CH_2) twist	1206 (11), 1187 (11), 1182 (28)	1207 (9), 1188 (11), 1181 (14)	1234 (9), 1181 (11)
δ(CH_3) wagg. + δ(CH_2) twist	1165 (10), 1135 (11), 1132 (17), 1122 (12)	1153 (12), 1135 (99), 1133 (80), 1127 (16)	1166 (9), 1135 (9), 1122(8), 1117 (24)
δ(CH_2) twist + ν(CC) + linker vib.	1108 (29)	1108 (31)	1106 (108), 1104 (104)

6. Optimized geometries

6MH

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
C	-0.83060500	2.81350000	-0.05887800
H	-0.41307800	3.34425100	0.81417700
H	-1.93191400	2.86991700	-0.01208000
C	-0.36721000	1.35059000	-0.07092400
H	0.73676900	1.31564300	-0.15343200
H	-0.76708900	0.84609200	-0.97224700
C	-0.81533600	0.58717000	1.18996000
H	-0.41895600	1.09763200	2.09104600
H	-1.92031400	0.62844300	1.27340400
C	-0.35967400	-0.88345400	1.19728400
H	0.74557100	-0.92249300	1.11158500
H	-0.75541300	-1.39163300	0.29419300
C	-0.80650400	-1.64643200	2.45574100
H	-1.90996000	-1.62338800	2.54465900
H	-0.40715000	-1.15338400	3.36319700
C	-0.35271600	-3.10638500	2.46113100
H	0.75807700	-3.15197800	2.38929000
H	-0.75871900	-3.62656800	1.56354300
S	-0.24887900	3.65321500	-1.61020200
H	-0.77966700	4.88083000	-1.33745900
O	-0.82369300	-3.70759900	3.67177400
H	-0.52826900	-4.64049200	3.66522600

6MH-H_2O

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
C	-0.83775400	2.93399100	-0.04828600
H	-0.26911900	3.42577200	0.75985900
H	-1.91575100	3.04741000	0.15991400
C	-0.46184200	1.44929200	-0.14366100
H	0.61461800	1.35776600	-0.38793100
H	-1.01610200	0.98294400	-0.98159200
C	-0.76185800	0.68748100	1.16147300
H	-0.21071000	1.16053400	1.99956900
H	-1.83886600	0.78623100	1.40691900
C	-0.39215300	-0.80527600	1.08845900
H	0.68482700	-0.90154900	0.84033400
H	-0.94260700	-1.27568600	0.24795300
C	-0.69079300	-1.56745100	2.39003400
H	-1.76652800	-1.48626000	2.64136000
H	-0.13710200	-1.11201300	3.23440100
C	-0.32373000	-3.05178000	2.31795900
H	0.76068400	-3.14998300	2.07365900
H	-0.88103000	-3.52691400	1.47595700
S	-0.44374300	3.77280700	-1.65850000
H	-0.86185200	5.02091800	-1.29679800
O	-0.64029100	-3.65429800	3.56733500
H	-0.39872200	-4.61366400	3.50522600
O	-0.09655600	-6.42520100	3.78451100
H	-0.99420200	-6.67355600	4.08812500
H	0.41848500	-6.35942900	4.61518800

POPC

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
N	-46.36043700	-41.48957500	-13.60945000
C	-45.31796500	-41.86495800	-12.55028400
C	-46.10562700	-42.22029000	-14.91049700
C	-46.33784700	-39.99732800	-13.86589800
C	-47.70389500	-41.88596900	-13.06936100
H	-45.32314300	-42.96795900	-12.50679900
H	-45.68918600	-41.43248300	-11.60485800
H	-45.18896600	-41.79021700	-15.35882200
H	-46.00514900	-43.29339000	-14.68372800
H	-46.97627600	-42.03882500	-15.56295400
H	-45.42155400	-39.78410200	-14.45761400
H	-47.24052700	-39.75170600	-14.44959500
H	-46.34266600	-39.48914900	-12.88733500
H	-48.46673200	-41.65865500	-13.83122000
H	-47.68999600	-42.96695700	-12.85409100
H	-47.89528200	-41.31239800	-12.14813200
C	-43.91808600	-41.31096500	-12.76717200
H	-43.97035700	-40.21650000	-12.87039600
H	-43.39682600	-41.48734900	-11.80270100
P	-42.67402100	-40.86974700	-15.08638200
O	-43.94499700	-40.28308800	-15.70992200
O	-41.57716800	-41.60243100	-15.80659600
O	-43.18968200	-41.92083200	-13.81697600
O	-42.07870600	-39.56415300	-14.16319100
C	-41.10523100	-39.85635500	-13.16453800
H	-41.43883600	-40.68555900	-12.51138800
H	-40.13781400	-40.14560000	-13.61814800
C	-40.93716100	-38.59794400	-12.28291000
H	-40.21875400	-37.87809900	-12.71616200
O	-40.39592200	-39.11316900	-11.02966900
C	-40.01932400	-38.22762300	-10.06525200
O	-40.03415600	-37.00712200	-10.19334400
C	-39.62750400	-38.97841400	-8.80422800
H	-40.56831200	-39.16731100	-8.24663900
H	-39.22795400	-39.97151400	-9.07975800
C	-42.24425300	-37.84095900	-12.05705100
H	-42.05216800	-36.93513300	-11.46971000
H	-42.67285000	-37.59382500	-13.04303800
O	-43.15263100	-38.74100000	-11.36797100
C	-44.31660800	-38.29962000	-10.81994100
O	-45.21806100	-39.11749700	-10.64559000
C	-44.41494700	-36.84196700	-10.38560300
H	-44.28851900	-36.18308800	-11.26716900
H	-45.44373900	-36.71244000	-10.01072900
C	-38.64733100	-38.21337000	-7.90503100
H	-37.72009500	-37.98988100	-8.46790900
H	-39.08810600	-37.23752000	-7.62760900
C	-38.31282700	-39.02148300	-6.64347100
H	-39.25206600	-39.25569500	-6.10307300
H	-37.86928900	-39.99547400	-6.93652200
C	-37.35366500	-38.29347600	-5.69218600
H	-36.42452700	-38.02410600	-6.23537900

H	-37.81303700	-37.33713500	-5.36704000
C	-36.99952000	-39.12947000	-4.45375500
H	-37.93299500	-39.41961600	-3.93103600
H	-36.51108100	-40.07329000	-4.77293200
C	-36.08423800	-38.38361100	-3.47571600
H	-35.16197100	-38.06166200	-4.00174600
H	-36.58795500	-37.45493000	-3.13677600
C	-35.69954100	-39.21742300	-2.24392500
H	-36.62209700	-39.55853600	-1.72966600
C	-34.82458600	-38.44574600	-1.27786000
H	-34.51060200	-37.44127900	-1.59821200
C	-34.42125100	-38.90474000	-0.07709300
H	-33.79870200	-38.25820500	0.55596500
C	-34.79240600	-40.25996100	0.48008800
H	-35.81398700	-40.19928500	0.91200300
H	-34.85395900	-41.00018400	-0.34287400
C	-33.85180900	-40.76651300	1.59327500
H	-34.24379000	-41.73358500	1.96760000
H	-32.84258300	-40.97007800	1.18386700
C	-33.75287100	-39.77645600	2.76984500
H	-33.05940900	-38.94983300	2.51733800
H	-34.74410100	-39.30865200	2.92572500
C	-33.31305400	-40.42318400	4.09054000
H	-34.05715900	-41.19861800	4.37036000
H	-32.34795600	-40.95396400	3.95782100
C	-33.19590200	-39.39883000	5.23017000
H	-32.33889200	-38.72212700	5.03446700
H	-34.10234800	-38.76076700	5.23341300
C	-33.04302500	-40.04538000	6.61367800
H	-33.90504700	-40.72211600	6.79050100
H	-32.13511400	-40.68355000	6.63243100
C	-32.97093700	-39.01495500	7.75117700
H	-32.11564100	-38.33237000	7.57148000
H	-33.88383500	-38.38518000	7.72172100
C	-32.83474200	-39.67125000	9.13411800
H	-31.91550900	-40.28584100	9.18683500
H	-32.78866200	-38.91766400	9.94226200
H	-33.69489100	-40.33808000	9.33662600
C	-43.38753000	-36.43973900	-9.29136600
H	-42.39887000	-36.22338200	-9.73380800
H	-43.73827400	-35.49008800	-8.84288400
C	-43.18119000	-37.48076900	-8.17905900
H	-44.14218900	-37.70545700	-7.67364800
H	-42.82670400	-38.43188500	-8.62726000
C	-42.14615000	-36.97604700	-7.16235500
H	-41.19492700	-36.78383900	-7.69833600
H	-42.48082900	-35.99618800	-6.76259100
C	-41.89504600	-37.92932300	-5.98558300
H	-42.85441000	-38.15242800	-5.47457200
H	-41.51136000	-38.90146400	-6.36040400
C	-40.89871100	-37.34446400	-4.97317900
H	-39.94836400	-37.11187900	-5.49384600
H	-41.29178900	-36.37768600	-4.59615400
C	-40.61255800	-38.27207900	-3.78435000
H	-41.56819500	-38.54566000	-3.29060000
H	-40.17207500	-39.22168400	-4.15363500
C	-39.66802900	-37.64446100	-2.74797800

H	-38.72048000	-37.35540800	-3.24426500
H	-40.11937900	-36.70679700	-2.36294700
C	-39.36893100	-38.58501000	-1.57208100
H	-40.32864800	-38.92317200	-1.12766800
H	-38.86737300	-39.49993800	-1.95188700
C	-38.50537800	-37.95348200	-0.46938200
H	-37.51282600	-37.67293500	-0.87554500
H	-38.98447900	-37.01411300	-0.12325800
C	-38.32011500	-38.89822100	0.72797900
H	-39.31888400	-39.21308200	1.09725400
H	-37.81390300	-39.82443700	0.38478500
C	-37.53109900	-38.27598700	1.89006200
H	-36.51172200	-38.01073800	1.54208700
H	-38.02192000	-37.32677000	2.19061100
C	-37.43624900	-39.20442100	3.11207300
H	-38.44347900	-39.60437600	3.35202000
H	-36.80854000	-40.08458900	2.85823300
C	-36.86219100	-38.51429200	4.36071200
H	-35.90460800	-38.01890300	4.10109800
H	-37.55219700	-37.70559800	4.67507200
C	-36.63497600	-39.49806200	5.51891900
H	-35.91952300	-40.28742200	5.21906300
H	-36.22196800	-38.99329600	6.41241300
H	-37.58288900	-39.99120700	5.80812100
H	-35.18225500	-40.14495600	-2.57309000

POPC-H_2O

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
N	-46.98673800	-40.98569600	-13.81775600
C	-46.13403800	-41.13772900	-12.55521500
C	-46.81625800	-42.16344400	-14.74844500
C	-46.63660200	-39.71439800	-14.55686400
C	-48.41911900	-40.90507400	-13.36531500
H	-46.35848100	-42.14692800	-12.16828300
H	-46.50519500	-40.36509200	-11.85978800
H	-45.80342600	-42.17898800	-15.17481500
H	-47.01116500	-43.08286500	-14.17400500
H	-47.55338700	-42.05247400	-15.56025000
H	-45.61602500	-39.83640800	-14.97481300
H	-47.37232300	-39.60346700	-15.37090700
H	-46.72563800	-38.88123200	-13.84114400
H	-49.05844700	-40.82566400	-14.25932200
H	-48.65913700	-41.82515700	-12.80716200
H	-48.51398800	-40.01354300	-12.72411500
C	-44.63612700	-40.94273400	-12.72486100
H	-44.42843900	-39.95809100	-13.18034400
H	-44.23034900	-40.89443200	-11.69693100
P	-43.07685300	-41.54549700	-14.78941300
O	-43.93594000	-40.63317800	-15.67109900
O	-42.40401500	-42.81223800	-15.26344700
O	-44.01480900	-42.00532700	-13.43864300
O	-41.99478300	-40.47368700	-14.04759800
C	-41.29171700	-40.94574400	-12.89895100
H	-41.98688000	-41.38973700	-12.16094700
H	-40.53370200	-41.70490800	-13.17085200
C	-40.62124100	-39.71488400	-12.26540000
H	-39.72286600	-39.39881300	-12.82384000
O	-40.23048800	-40.14174100	-10.92170400
C	-39.42275100	-39.31137100	-10.21995100
O	-38.91164100	-38.29924700	-10.70518400
C	-39.28248000	-39.78497100	-8.78776700
H	-40.26830900	-39.62755800	-8.30333800
H	-39.12683400	-40.88099500	-8.79594300
C	-41.55757300	-38.51301800	-12.17635000
H	-41.05533700	-37.67677900	-11.67135200
H	-41.86937800	-38.23286400	-13.19623300
O	-42.72621400	-38.95026100	-11.43230900
C	-43.64822500	-38.07394600	-10.98227400
O	-44.72300500	-38.54233100	-10.59361200
C	-43.29805600	-36.59806700	-10.88824700
H	-42.93052900	-36.22662300	-11.86371000
H	-44.24381900	-36.08068800	-10.65849400
C	-38.18647900	-39.06846300	-7.98930800
H	-37.21648700	-39.17680200	-8.51380800
H	-38.41200200	-37.98558200	-7.95379200
C	-38.07858300	-39.64639400	-6.57138400
H	-39.05845200	-39.55044700	-6.06296400
H	-37.86303000	-40.73334900	-6.63522500
C	-37.00064500	-38.96952900	-5.71374700
H	-36.03034400	-38.98748500	-6.25208200

H	-37.26057100	-37.90061900	-5.56886000
C	-36.83618400	-39.64230100	-4.34301600
H	-37.81584700	-39.65863200	-3.82504900
H	-36.53982300	-40.70171900	-4.48849500
C	-35.80680300	-38.94103000	-3.44925500
H	-34.83191500	-38.88351400	-3.97574200
H	-36.12657200	-37.89336600	-3.27241400
C	-35.61239200	-39.63715100	-2.09338800
H	-36.59711100	-39.73745500	-1.59108700
C	-34.64739300	-38.89394500	-1.19327900
H	-34.18967700	-37.98469200	-1.61022300
C	-34.33036100	-39.27025200	0.06109400
H	-33.63012300	-38.65284300	0.63971700
C	-34.90230300	-40.48858900	0.74870000
H	-35.91161000	-40.23585900	1.13775700
H	-35.05715200	-41.29904500	0.00809200
C	-34.06368400	-40.99746200	1.93944700
H	-34.59568100	-41.85435300	2.39954000
H	-33.08624200	-41.38258700	1.58741600
C	-33.84694300	-39.90980800	3.00900900
H	-33.04289700	-39.21604700	2.69240900
H	-34.76679700	-39.29870500	3.08644100
C	-33.51956800	-40.46581500	4.40177200
H	-34.36571100	-41.10216500	4.73697600
H	-32.63276300	-41.13062400	4.35315600
C	-33.28410800	-39.35184500	5.43390700
H	-32.34171600	-38.81741400	5.19519600
H	-34.09744700	-38.60385000	5.34610500
C	-33.23983300	-39.86421300	6.87997700
H	-34.18658600	-40.40069500	7.09942100
H	-32.42487800	-40.60957300	6.99035400
C	-33.04982700	-38.73827800	7.90809000
H	-32.10863900	-38.19577900	7.68535500
H	-33.87035100	-38.00138400	7.78724900
C	-33.02439600	-39.25777500	9.35421600
H	-32.19601200	-39.97814600	9.49679200
H	-32.89155200	-38.43593300	10.08224200
H	-33.96872000	-39.78157700	9.59794800
C	-42.24322500	-36.30012900	-9.77610400
H	-41.21332400	-36.36224600	-10.17198900
H	-42.37902900	-35.24570700	-9.47014600
C	-42.34255000	-37.20940000	-8.53837600
H	-43.38457800	-37.23131400	-8.15968100
H	-42.09846500	-38.25154100	-8.83496800
C	-41.37599400	-36.74498000	-7.43935100
H	-40.36226400	-36.62836500	-7.87272400
H	-41.67680700	-35.73070100	-7.10390000
C	-41.32003100	-37.67435500	-6.21845700
H	-42.31946700	-37.73988400	-5.74031600
H	-41.06377200	-38.70586900	-6.54113500
C	-40.28370300	-37.19310200	-5.19212000
H	-39.28475400	-37.18070300	-5.67344900
H	-40.50777500	-36.14127900	-4.91851300
C	-40.22233400	-38.04081600	-3.91427500
H	-41.22854600	-38.08168200	-3.44794500
H	-39.95791600	-39.08764300	-4.17190500
C	-39.21082900	-37.49668600	-2.89397600

H	-38.21031700	-37.43886900	-3.36756500
H	-39.48954700	-36.45694100	-2.62476600
C	-39.12683800	-38.34603400	-1.61813100
H	-40.14513400	-38.46084800	-1.19091900
H	-38.78634800	-39.37014600	-1.87955500
C	-38.19840200	-37.75909900	-0.54391600
H	-37.16479100	-37.67813800	-0.93649300
H	-38.52758600	-36.72699400	-0.30284200
C	-38.19181100	-38.60131200	0.74096000
H	-39.23574600	-38.72447400	1.09870500
H	-37.82543400	-39.62208800	0.50474000
C	-37.34450600	-37.99377100	1.86932400
H	-36.28922300	-37.91682300	1.53584400
H	-37.69320000	-36.95773600	2.06239400
C	-37.41615200	-38.80201900	3.17536000
H	-38.47640900	-39.02704200	3.41441500
H	-36.91882100	-39.78405600	3.02881200
C	-36.77526500	-38.08098500	4.37309000
H	-35.75251000	-37.75057600	4.09997900
H	-37.35013400	-37.15769600	4.58745300
C	-36.71148200	-38.96917000	5.62516900
H	-36.10718800	-39.87448900	5.42494200
H	-36.25032000	-38.44100400	6.48077100
H	-37.72461500	-39.29722800	5.92728700
H	-35.25788600	-40.67765800	-2.25928700
H	-44.04970100	-43.41086500	-16.57194700
O	-44.85344700	-43.02827900	-16.99626700
H	-44.61316800	-42.07117700	-16.94607300
H	-38.60483700	-36.76770800	-9.75289600
O	-38.62398200	-35.93639400	-9.21626200
H	-37.73428000	-35.90521200	-8.81242600
H	-46.37715900	-38.24227300	-11.23093800
O	-47.29975100	-38.24599600	-11.59702800
H	-47.85855700	-38.09727000	-10.80861100

CPEO3

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
C	-0.28072500	2.67630200	4.13562000
C	-1.64066800	1.97423800	4.50196600
C	-2.18669100	1.20410700	3.28747800
C	-1.44371400	0.93663600	2.19261400
C	0.00675400	1.30038400	2.02369300
C	0.67914200	1.74019700	3.33947700
H	-0.54441600	3.49477700	3.42839400
H	-1.91869000	0.40828500	1.35224600
H	0.55712200	0.44433200	1.58423400
H	0.09675400	2.11685700	1.27196800
H	0.88236500	0.83045100	3.94102200
C	-3.63433400	0.74186700	3.35939900
C	-2.68223000	3.07821000	4.88274500
C	-4.13232400	2.57566800	5.01912300
C	-4.57922500	1.89434100	3.72371300
H	-3.93822900	0.29077200	2.39741500
H	-3.75678600	-0.04589000	4.13195700
H	-4.62394700	2.63883500	2.90874900
H	-2.65551900	3.86486100	4.10177600
H	-2.37736100	3.56408800	5.82806100
H	-4.22595300	1.85452300	5.85409600
H	-4.80626200	3.42273800	5.23928200
C	0.41031800	3.34377800	5.35658500
H	-0.26125700	4.10913300	5.78848000
H	0.56444100	2.59362300	6.15385200
C	1.99854400	2.48138800	3.06542100
C	1.77219500	4.00092400	5.03031300
H	1.61483900	4.86679600	4.35455200
H	2.21940300	4.40360100	5.96112100
C	2.72258100	3.00015500	4.34109100
H	1.71269000	3.39751000	2.49617600
C	-1.47268300	0.98407100	5.68893800
H	-1.26159100	1.51558200	6.63398800
H	-2.38600000	0.38654300	5.85064200
H	-0.64915600	0.27263100	5.50073900
C	3.11151900	1.86785000	5.32391700
H	3.61199400	2.30244700	6.20988300
H	2.24406800	1.28941500	5.68494700
H	3.81887300	1.15264600	4.87043200
C	4.02469400	3.61109200	3.73706800
H	4.87575800	3.50652900	4.43528100
H	3.89726400	4.69579200	3.56230300
C	3.12673600	1.82000900	2.25322500
C	4.27511400	2.86803800	2.36939500
H	4.09343200	3.60783600	1.56340400
H	2.85679200	1.62869600	1.19895400
H	3.40905900	0.84560200	2.69623400
C	5.71220600	2.33028400	2.12030000
H	5.68170000	1.81777100	1.13382600
C	6.18330800	1.29347100	3.15872500
H	5.48012400	0.44550000	3.23991700
H	7.16525600	0.87139300	2.88224800

H	6.28860700	1.74263700	4.16386200
C	6.71168400	3.50750600	1.98861200
H	6.27738900	4.25929700	1.29722800
H	6.80668300	4.01778300	2.96970600
C	8.11507300	3.12897200	1.47769000
H	8.59868600	2.43452400	2.19035100
H	8.01756100	2.57307600	0.52154000
C	9.01869200	4.35714800	1.25914900
H	8.49514900	5.06597700	0.58479200
H	9.14795500	4.89499500	2.22239500
C	10.41488500	4.05555500	0.66503900
H	10.26239400	3.48645700	-0.27691600
C	11.14387100	5.36631100	0.31157400
H	12.12838000	5.16792900	-0.15047400
H	10.55472600	5.97885700	-0.39553300
H	11.31799100	5.97672300	1.21853000
C	11.28159600	3.19421000	1.60404800
H	11.44252800	3.70960600	2.57075800
H	10.81807400	2.21508200	1.81838500
H	12.27541700	3.00120900	1.16006600
O	-5.91020500	1.31565900	3.87447100
C	-6.95627500	2.17211900	3.62160900
O	-6.85167500	3.34624500	3.27026400
N	-8.14427800	1.51344900	3.84255800
C	-9.42230900	2.12493000	3.51058600
H	-9.32678200	3.20959700	3.67822900
H	-10.19989000	1.73756000	4.19255600
C	-9.84014900	1.87051700	2.05373900
H	-9.08677800	2.31812300	1.36936900
H	-9.87511600	0.77558700	1.84594200
C	-11.58988300	2.32278300	0.52956000
H	-11.69048900	1.25147100	0.24619100
H	-10.89002700	2.79588700	-0.19369500
C	-12.95432900	3.00686000	0.45206500
H	-12.85265200	4.07974600	0.72724100
H	-13.65188100	2.53885500	1.18116000
C	-14.70182100	3.47148700	-1.07221500
H	-15.46523200	3.02830200	-0.39596700
H	-14.65126700	4.56082400	-0.84435200
C	-15.10192100	3.25209900	-2.53155800
H	-14.34577700	3.67312800	-3.21402500
H	-15.17839100	2.17201000	-2.74256900
O	-11.12050600	2.46047800	1.86518500
O	-13.42923000	2.85993200	-0.88226900
S	-16.76816700	3.93819700	-2.92702800
H	-8.09291900	0.51100700	4.01741500
H	-16.43395200	5.25434800	-2.78393800

CPEO3+H_2O (CO)

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
C	-6.31299700	2.89047400	-2.26693300
C	-6.15004700	2.80710100	-0.70382400
C	-4.92182100	1.95497900	-0.34162500
C	-3.97747900	1.60664200	-1.24113000
C	-3.96215000	2.03228700	-2.68437600
C	-4.96110200	3.16506100	-2.99371900
H	-6.61932200	1.87105100	-2.59334100
H	-3.14561900	0.96769700	-0.90839600
H	-2.93694600	2.34171800	-2.97053300
H	-4.18819100	1.15346600	-3.32954000
H	-4.53059100	4.11460900	-2.61482700
C	-4.80058400	1.50162200	1.10565200
C	-7.41667200	2.10669000	-0.10959500
C	-7.30000000	1.71847900	1.37715700
C	-6.08352700	0.81546400	1.59132700
H	-3.93811600	0.82053100	1.22086000
H	-4.61279000	2.36782900	1.77393900
H	-6.23579500	-0.14652600	1.07070000
H	-7.61418700	1.18785300	-0.69785900
H	-8.29836400	2.76019800	-0.24333500
H	-7.19475300	2.61773800	2.01448700
H	-8.21371300	1.18965000	1.70175700
C	-7.44067000	3.86207100	-2.71262200
H	-8.40592700	3.52695500	-2.28922500
H	-7.25713100	4.86599500	-2.28760200
C	-5.21306300	3.28245800	-4.50625500
C	-7.58821000	3.99731900	-4.24669600
H	-7.93454800	3.03342800	-4.67333100
H	-8.37344600	4.74500700	-4.47650900
C	-6.24510300	4.37692800	-4.90372200
H	-5.67981900	2.31459200	-4.80682400
C	-5.98722600	4.21700300	-0.06866900
H	-6.91993800	4.80435900	-0.14024000
H	-5.72665700	4.15263300	1.00159100
H	-5.18109200	4.78640400	-0.56420400
C	-5.82037300	5.80161300	-4.46890800
H	-6.59746800	6.52830300	-4.77262200
H	-5.68374300	5.90053400	-3.37845200
H	-4.87778100	6.11542200	-4.94902100
C	-6.20698700	4.29431300	-6.46113800
H	-6.38663500	5.28527800	-6.91763200
H	-7.00994400	3.63061100	-6.83270400
C	-4.04981900	3.50855800	-5.48890200
C	-4.79595800	3.70416400	-6.84370100
H	-4.97133400	2.68464700	-7.24337700
H	-3.34058300	2.66247400	-5.53444400
H	-3.46578400	4.40483800	-5.20454100
C	-4.00416700	4.45256600	-7.95253400
H	-3.03777000	3.91247500	-8.05772800
C	-3.67295200	5.91758200	-7.60706100
H	-3.13338800	6.00103100	-6.64683500
H	-3.02745400	6.37194900	-8.37885100

H	-4.58710300	6.53599800	-7.53604600
C	-4.74011900	4.33448300	-9.31137800
H	-5.02887300	3.27346700	-9.46191000
H	-5.69202400	4.90306200	-9.26167300
C	-3.93708500	4.79999600	-10.54111400
H	-3.69808600	5.87609300	-10.44720500
H	-2.96323000	4.26727100	-10.56412100
C	-4.68215300	4.55015700	-11.86597300
H	-4.96223500	3.47769400	-11.91763000
H	-5.63952600	5.11336000	-11.86154700
C	-3.89531800	4.91371100	-13.14737300
H	-2.92832200	4.36768400	-13.11290800
C	-4.65895900	4.44203200	-14.39989600
H	-4.09129500	4.65900500	-15.32334400
H	-4.85356200	3.35415100	-14.36926300
H	-5.63669600	4.95477600	-14.48122400
C	-3.58565900	6.42039300	-13.24156300
H	-4.52134100	7.01234400	-13.25405300
H	-2.97514500	6.77470700	-12.39231200
H	-3.03155200	6.65381800	-14.16919900
O	-5.89179800	0.53067300	3.01279400
C	-6.60288400	-0.52726400	3.51420900
O	-7.37820000	-1.22266700	2.84530800
N	-6.29723500	-0.69580400	4.83733200
C	-6.98358100	-1.62916100	5.72547200
H	-6.24830300	-2.05766100	6.43046100
H	-7.40384300	-2.45539400	5.12416800
C	-8.10380600	-0.94391500	6.52465300
H	-7.71909700	-0.02233000	7.02326300
H	-8.92008800	-0.63169900	5.83633800
C	-9.68018000	-1.38354300	8.22937700
H	-10.52834800	-1.12230900	7.55960900
H	-9.40797700	-0.46728200	8.79965300
C	-10.10636400	-2.48605400	9.19804200
H	-9.25028400	-2.76211100	9.85282300
H	-10.39932500	-3.39423200	8.62791500
C	-11.68976400	-2.93961300	10.89521100
H	-12.04064700	-3.85950200	10.37868100
H	-10.88704900	-3.24516800	11.60507600
C	-12.84729900	-2.28730800	11.65193000
H	-12.51748000	-1.36273300	12.15320700
H	-13.65236300	-2.01711000	10.94780100
O	-8.57225700	-1.87706800	7.48685300
O	-11.19694900	-1.98188500	9.96308300
S	-13.63695100	-3.42467500	12.87111200
H	-12.57193000	-3.50631800	13.72180800
H	-5.68438600	0.01115300	5.24306900
H	-8.20586000	-2.85407100	3.23396000
O	-8.43648200	-3.74973200	3.58592300
H	-7.66902000	-4.28911500	3.30835700

CPEO3+H_2O (NH)

S = 0 total charge = 0

Atom Coordinates (Ångstroms)

	X	Y	Z
C	-4.00465400	-4.90273100	3.12631400
C	-4.16150000	-3.43589800	3.67486500
C	-4.73885200	-3.45913000	5.10038300
C	-4.80410000	-4.57968100	5.85011400
C	-4.28870400	-5.92927100	5.42925900
C	-3.38680800	-5.87163400	4.18032400
H	-5.04051500	-5.27467800	2.95857500
H	-5.26157200	-4.51757900	6.84889200
H	-3.74124900	-6.39791000	6.27142800
H	-5.14997800	-6.60697400	5.23301100
H	-2.39458200	-5.48573900	4.49229100
C	-5.25393800	-2.13921700	5.65576200
C	-5.16979700	-2.66810600	2.75711500
C	-5.62872400	-1.30271800	3.30478500
C	-6.24627200	-1.47312700	4.69429500
H	-5.72732300	-2.29911500	6.64128300
H	-4.41199000	-1.43201800	5.81696800
H	-7.17527300	-2.06444300	4.61861700
H	-6.06502500	-3.30625100	2.61439500
H	-4.72363800	-2.52457400	1.75571000
H	-4.77735400	-0.59717400	3.37200700
H	-6.37471100	-0.85441500	2.62472300
C	-3.27710700	-4.96713100	1.75485200
H	-3.84616700	-4.38405200	1.00678000
H	-2.29008300	-4.47556500	1.83342800
C	-3.22224800	-7.26558100	3.55196800
C	-3.06955600	-6.40274400	1.21698400
H	-4.05347900	-6.85521700	0.97579400
H	-2.49927900	-6.36128800	0.26749300
C	-2.35731800	-7.29011500	2.25878800
H	-4.24578300	-7.56894300	3.22746200
C	-2.80020300	-2.68486100	3.69777000
H	-2.43574300	-2.47439500	2.67654900
H	-2.87997800	-1.71560300	4.21914700
H	-2.02922300	-3.27341600	4.22582500
C	-0.90832100	-6.79189800	2.48504300
H	-0.35163700	-6.82666700	1.52946400
H	-0.85882200	-5.75484400	2.85857900
H	-0.36092900	-7.42699100	3.20228200
C	-2.31958900	-8.81562600	1.93457600
H	-1.36328200	-9.09827800	1.45674000
H	-3.11670500	-9.07716500	1.21380400
C	-2.67749000	-8.44723600	4.37453600
C	-2.55467700	-9.56938100	3.29917600
H	-3.55619400	-10.04162600	3.23751200
H	-3.34227800	-8.75039100	5.20341100
H	-1.69745800	-8.19420100	4.82266700
C	-1.57213400	-10.72593700	3.63614600
H	-1.87486700	-11.10405500	4.63717700
C	-0.09844100	-10.28707200	3.74073500
H	0.03494100	-9.46684400	4.46845200
H	0.54209400	-11.12134400	4.07609500

H	0.29372800	-9.94147100	2.76611400
C	-1.75461500	-11.89565300	2.63592900
H	-2.84005000	-12.10530500	2.53746000
H	-1.41632200	-11.57232300	1.62949400
C	-1.03780100	-13.20614400	3.01360500
H	0.05554900	-13.04005300	3.04476200
H	-1.33552800	-13.50164100	4.04160000
C	-1.35746600	-14.35888900	2.04318000
H	-2.45929500	-14.47576200	1.98330800
H	-1.02631600	-14.08112700	1.01995200
C	-0.73867500	-15.72749700	2.41367700
H	-1.05368500	-15.96750500	3.45174200
C	-1.28315000	-16.83267200	1.48816800
H	-0.88094200	-17.82444400	1.76511000
H	-2.38637200	-16.88989700	1.53207700
H	-0.99962800	-16.63985700	0.43557900
C	0.80184500	-15.70687200	2.37880400
H	1.16711300	-15.44658300	1.36643400
H	1.22539500	-14.97364800	3.08766500
H	1.21735900	-16.69789000	2.63815000
O	-6.59953100	-0.17717600	5.26945800
C	-7.87325300	0.29774600	4.93964700
O	-8.66203700	-0.30849700	4.21766000
N	-8.08170200	1.50273900	5.54466200
C	-9.34575800	2.20821700	5.39021800
H	-9.56111800	2.76514500	6.31888700
H	-10.14754100	1.46693500	5.23793900
C	-9.32698300	3.18128400	4.20143400
H	-8.49159300	3.91049400	4.31199100
H	-9.15624000	2.61348300	3.26071900
C	-10.69313100	4.75355400	3.08237900
H	-10.57333900	4.22645100	2.11032400
H	-9.91252300	5.54517100	3.13200800
C	-12.07971900	5.39198800	3.15695000
H	-12.20230400	5.91045600	4.13357400
H	-12.86075200	4.60275200	3.09656700
C	-13.45412000	6.95804000	2.03856300
H	-14.28128800	6.22173200	1.93850500
H	-13.62872600	7.52459700	2.98195500
C	-13.44941600	7.90903900	0.84127400
H	-12.62326800	8.63451500	0.91885500
H	-13.30680800	7.33832500	-0.09194600
O	-10.58221900	3.85085200	4.17505900
O	-12.18730000	6.30755500	2.07137800
S	-15.05389600	8.79316100	0.62331500
H	-14.97246500	9.54789400	1.75833900
H	-7.32386800	1.88559000	6.13494500
O	-5.57667500	1.86771300	6.93572700
H	-5.01730000	2.37335900	6.31117000
H	-5.62170700	0.97084900	6.52230400