Contra-Hofmeister Anion Extraction by Cyclosteroidal Receptors

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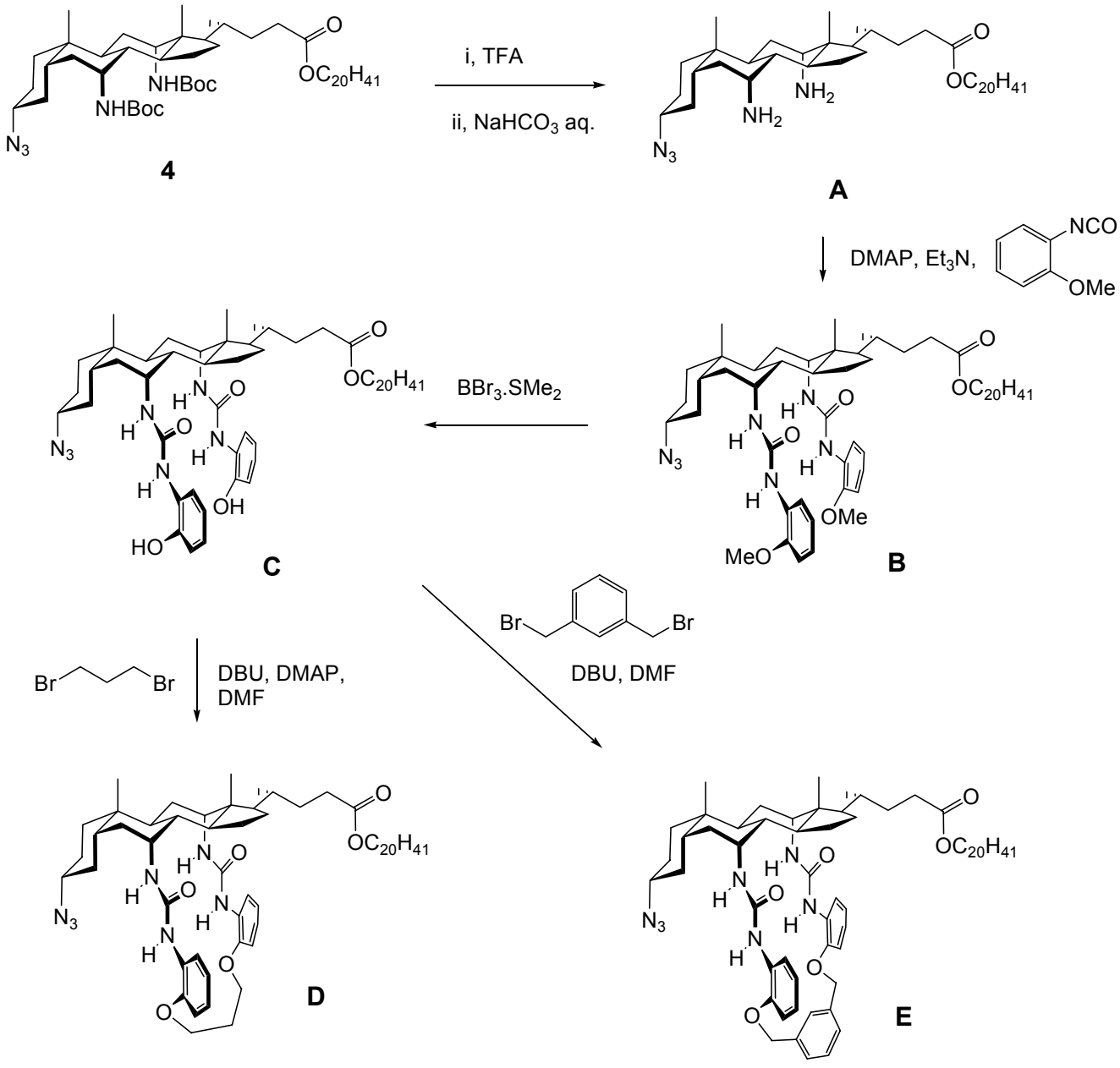
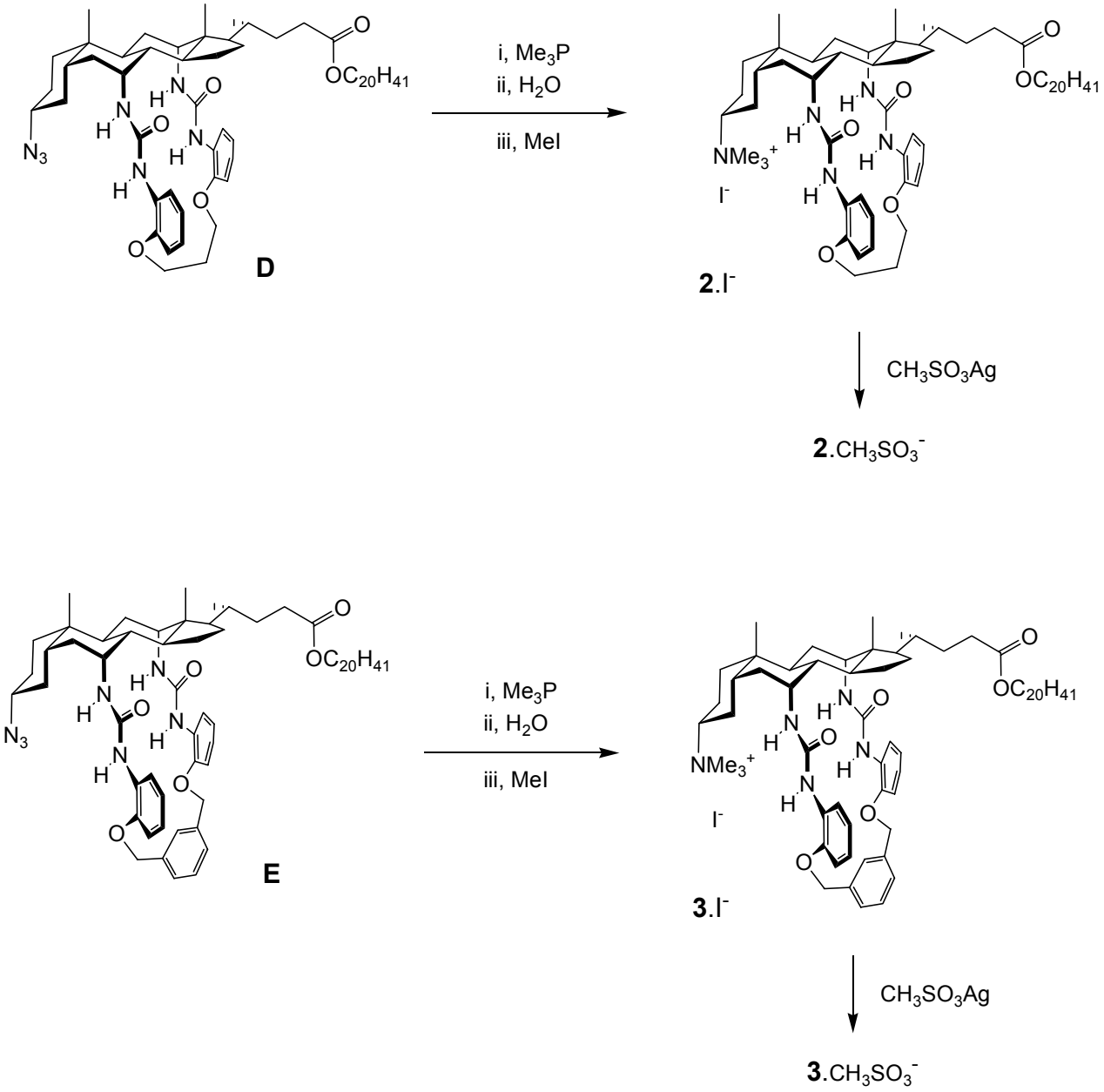
Supplementary Information

Synthesis of receptors 2 and 3

The receptors were prepared from intermediate **4**[1] as indicated in the following schemes.





Eicosyl 3α-azido-7α,12α-bis[(o-methoxyphenylaminocarbonyl)amino]-5β-cholan-24-oate B. Bis-carbamate **4**[1] (0.71 g, 0.78 mmol) was dissolved in dry dichloromethane (15 mL) and placed in an ice bath. To the stirred solution was added trifluoroacetic acid (7.5 mL) dropwise. After 1 h the reaction mixture was allowed to warm to r.t. and stirred for a further 2 h The solvent was evaporated under reduced pressure, dissolved in dichloromethane, washed with sat. aq. $NaHCO_3$, dried ($MgSO_4$) and evaporated under reduced pressure. The residual bis-amino compound **A** was dissolved in dry THF (15 mL) with DMAP (0.105 g, 0.86

mmol), triethylamine (272 μL, 0.197 g, 1.95 mmol) and o-methoxyphenylisocyanate (180 μL, 159 mg, 1.60 mmol) and the solution was stirred at 50 ^{o}C for 24 h. The reaction solution was evaporated under reduced pressure. The dry residue was purified by flash column chromatography (eluent DCM/methanolic ammonia 99:1 to 98:2) to give product bis-urea **B** (0.69 g, 88 %) as a white solid: R_f = 0.3 (DCM/methanolic ammonia 99:1); ν_{max} = 3395, 2922, 2853, 2090, 1692, 1521, 1457, 1248, 1119, 1027, 743 cm^{-1}; ^{1}H NMR (400 MHz, $(CD_3)_2CO$, 25 ^{o}C, TMS): δ = 0.85 – 0.92 (m, 9 H; 18-H_3 + 20'-H_3 + 21-H_3), 1.05 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(CH_2)_{17}CH_3$), 3.31 – 3.42 (m, 1 H; 3β-H), 3.87 (s, 3 H; OCH_3), 3.89 (s, 3 H; OCH_3), 3.99 (t, ^{3}J(H,H) = 6.6 Hz, 2 H; 1'-H_2), 4.06 (bs, 1 H; 7β-H), 4.17 – 4.23 (m, 1 H; 12β-H), 6.24 – 6.36 (m, 2 H; 2 x CH-N**H**CO), 6.83 – 6.98 (m, 6 H), 7.69 (bs, 1 H; Ar-NHCO), 7.88 (bs, 1 H; Ar-NHCO), 8.34 - 8.38 (m, 2 H); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 ^{o}C, TMS): δ = 14.2 (18-CH_3), 14.4 (20'-CH_3), 17.7 (21-CH_3), 23.4 (19'-CH_2), 23.5 (19-CH_3), 24.2 (CH_2), 26.7 (18'-CH_2), 27.8 (CH_2), 27.8 (CH_2), 28.1 (17'-CH_2), 29.5 (CH_2), 29.8 (CH), 30.0 (16'-CH_2), 30.1 (15'-CH_2), 30.3 (14'-CH_2), 30.3 (13'-CH_2), 30.4 (12'-CH_2), 30.4 ($COOCH_2CH_2(CH_2)_9(CH_2)_8CH_3$), 31.7 (2'-$CH_2$), 31.8 ($CH_2$), 32.7 ($CH_2$), 33.6 ($CH_2$), 35.6 (CH), 35.8 ($CH_2$), 35.9 (10-C), 36.1 ($CH_2$), 38.3 (CH), 42.8 (CH), 45.4 (CH), 45.8 (13-C), 47.0 (CH), 48.9 (7-CH), 53.8 (12-CH), 56.1 (2 x OCH_3), 62.7 (3-CH), 64.6 (1'-CH_2), 111.0 (Ar-CH), 111.0 (Ar-CH), 119.4 (Ar-CH), 119.6 (Ar-CH), 121.6 (2 x Ar-CH), 121.8 (Ar-CH), 121.8 (Ar-CH), 131.0 (Ar-C), 131.1 (Ar-C), 148.4 (2 x Ar-C), 155.3 (NHCONH), 155.4 (NHCONH), 174.0 (**C**$OOCH_2(CH_2)_{18}CH_3$); MS (ES+): m/*z* (%): 1011 (100) $[M + H]^+$, 1032 (20) $[M + Na]^+$; HRMS (ES+): m/*z* calculated for $[M + H]^+$ = 1010.7417, found 1010.7426; elemental analysis calculated (%) for $C_{60}H_{95}N_7O_6$ + 1.0 mol. H_2O: C 70.07, H 9.51, N 9.53; found C 70.24, H 9.45, N 9.29.

Eicosyl 3α-azido-7α,12α-bis[(o-hydroxyphenylaminocarbonyl)amino]-5β-cholan-24-oate C. To a solution of boron tribromide-methyl sulfide complex (0.28 g, 0.91 mmol) in dry DCM (8 mL) was added **B** (0.42 g, 0.41 mmol). The reaction mixture was stirred overnight after which time 0.5 N $HCl_{(aq.)}$ (8 mL) was

added. After vigorous stirring for 1 h the two phase mixture was separated. The organic phase was washed with water, dried ($MgSO_4$) and evaporated under reduced pressure. Purification by flash column chromatography (eluent DCM/methanolic ammonia 99:1 to 98:2) gave product **C** (0.33 g, 81 %) as a brown solid: R_f = 0.4 (DCM/methanolic ammonia 98:2); ν_{max} = 3357, 2922, 2853, 2091, 1658, 1540, 1498, 1246, 745 cm^{-1}; ^1H NMR (400 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 0.84 – 0.94 (m, 9 H; 18-H_3 + 20'-H_3 + 21-H_3), 1.05 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(CH_2)_{17}CH_3$), 3.34 – 3.44 (m, 1 H; 3β-H), 3.99 (t, ^3J(H,H) = 6.6 Hz, 2 H; 1'-H_2), 4.05 (bs, 1 H; 7β-H), 4.16 – 4.22 (m, 1 H; 12β-H), 6.29 – 6.45 (m, 2 H; 2 x CH-N**H**CO), 6.71 – 6.80 (m, 2 H), 6.80 – 6.89 (m, 4 H), 7.41 – 7.53 (m, 2 H), 7.92 (s, 1 H; Ar-NHCO), 8.09 (s, 1 H; Ar-NHCO); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 14.2 (18-CH_3), 14.4 (20'-CH_3), 17.6 (21-CH_3), 23.4 (19'-CH_2), 23.5 (19-CH_3), 24.1 (CH_2), 26.7 (18'-CH_2), 27.6 (CH_2), 27.8 (CH_2), 28.0 (17'-CH_2), 29.5 (CH_2), 29.8 (CH), 30.0 (16'-CH_2), 30.1 (15'-CH_2), 30.3 (14'-CH_2), 30.3 (13'-CH_2), 30.4 (12'-CH_2), 30.4 ($COOCH_2CH_2(\mathbf{C}H_2)_9(CH_2)_8CH_3$), 31.7 (2'-$CH_2$), 31.8 ($CH_2$), 32.7 ($CH_2$), 33.5 ($CH_2$), 35.5 (CH), 35.9 (10-C), 35.9 (CH_2), 36.0 (CH_2), 38.3 (CH), 42.6 (CH), 45.4 (CH), 45.8 (13-C), 47.5 (CH), 49.1 (7-CH), 54.3 (12-CH), 62.6 (3-CH), 64.7 (1'-CH_2), 118.1 (Ar-CH), 118.3 (Ar-CH), 120.6 (Ar-CH), 120.6 (Ar-CH), 121.2 (2 x Ar-CH), 124.2 (Ar-CH), 124.3 (Ar-CH), 129.0 (Ar-C), 129.0 (Ar-C), 148.4 (2 x Ar-C), 155.5 (2 x NHCONH), 174.0 ($\mathbf{C}OOCH_2(CH_2)_{18}CH_3$); MS (ES+): m/*z* (%): 983 (20) $[M + H]^+$, 1005 (100) $[M + Na]^+$, MS (ES-): m/*z* (%): 981 (100) $[M - H]^-$; HRMS (ES+): m/*z* calculated for $[M + H]^+$ = 982.7104, found 982.7101

Propylene linked macrocycle D. To a stirred solution of **C** (0.60 g, 0.61 mmmol) and DMAP[2] (0.15 g, 1.2 mmol) in DMF (6.1 mL) was added DBU (180 µL, 0.19 g, 1.2 mmol) followed by dibromopropane (63 µL, 0.124 g, 0.61 mmol). After stirring for 7 days the solvent was removed azeotropically with toluene/DCM under reduced pressure. The residue was dissolved in DCM and washed with 1 N $HCl_{(aq.)}$ followed by 4 % $NaHCO_{3(aq.)}$, dried ($MgSO_4$) and evaporated under reduced pressure. Purification by flash column

chromatography (eluent EtOAc/hexane 20:80 to 30:70) gave product **D** (0.100 g, 16 %) as a white solid: R_f = 0.4 (EtOAc/hexane 30:70); ν_{max} = 3404, 2922, 2853, 2091, 1700, 1530, 1448, 1249, 1195, 744 cm^{-1}; ^1H NMR (400 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 0.84 – 0.90 (m, 6 H; 18-H_3 + 20'-H_3), 0.98 (d, $^3J(H,H)$ = 6.2 Hz, 3 H; 21-H_3), 1.06 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(C\mathbf{H}_2)_{17}CH_3$), 3.41 – 3.50 (m, 1 H; 3β-H), 3.96 (t, $^3J(H,H)$ = 6.6 Hz, 2 H; 1'-H_2), 4.02 – 4.30 (m, 6 H, 7β-H + 12β-H + 2 x Ar-OCH_2), 5.92 (d, $^3J(H,H)$ = 8.4 Hz, 1 H; CH-N**H**CO), 6.13 (d, $^3J(H,H)$ = 9.5 Hz, 1 H; CH-N**H**CO), 6.82 – 6.94 (m, 2 H), 6.98 – 7.03 (m, 2 H), 7.59 (s, 1 H; Ar-NHCO), 7.87 (s, 1 H; Ar-NHCO), 8.36 - 8.41 (m, 2 H); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 13.9 (18-CH_3), 14.4 (20'-CH_3), 18.5 (21-CH_3), 23.0 (19-CH_3), 23.3 (19'-CH_2), 24.9 (CH_2), 26.7 (18'-CH_2), 26.8 (CH_2), 27.7 (CH_2), 28.1 (17'-CH_2), 28.9 (CH), 29.5 (CH_2), 30.0 (16'-CH_2), 30.1 (15'-CH_2), 30.3 (14'-CH_2), 30.3 (13'-CH_2), 30.4 (12'-CH_2), 30.4 ($COOCH_2CH_2(\mathbf{C}H_2)_9(CH_2)_8CH_3$), 31.02 ($OCH_2\mathbf{C}H_2CH_2O$), 31.6 (2'-$CH_2$), 32.0 ($CH_2$), 32.7 ($CH_2$), 33.9 ($CH_2$), 35.7 (CH), 36.1 ($CH_2$), 36.1 (10-C), 36.3 ($CH_2$), 38.9 (CH), 42.3 (CH), 44.1 (CH), 46.4 (13-C), 47.5 (CH), 49.0 (7-CH), 52.2 (12-CH), 62.8 (3-CH), 64.6 (1'-CH_2), 67.4 (Ar-OCH_2), 68.1 (Ar-OCH_2), 114.6 (Ar-CH), 115.0 (Ar-CH), 118.4 (Ar-CH), 118.8 (Ar-CH), 121.7 (Ar-CH), 121.8 (Ar-CH), 122.7 (Ar-CH), 122.8 (Ar-CH), 132.3 (Ar-C), 132.4 (Ar-C), 147.0 (Ar-C), 147.5 (Ar-C), 154.7 (NHCONH), 155.6 (NHCONH), 174.0 ($\mathbf{C}OOCH_2(CH_2)_{18}CH_3$); MS (ES+): m/*z* (%): 1023 (100) $[M + H]^+$.

Flushing the column (eluent EtOAc/hexane 1:1) allowed the recovery of starting material (0.30 g, 50 %)

m-Xylylene linked macrocycle E. To a stirred solution of **C** (1.32 g, 1.34 mmmol) in DMF (13.5 mL) was added DBU (400 μL, 0.41 g, 2.7 mmol) followed by α,α'-dibromo-m-xylene (0.36 g, 1.34 mmol). After stirring for 12 days the solvent was removed azeotropically with toluene/DCM under reduced pressure. The resultant residue was dissolved in DCM and washed with 3 portions of 4 % $KHSO_{4(aq.)}$, dried ($MgSO_4$) and evaporated under reduced pressure. Purification by flash column chromatography (eluent EtOAc/ hexane 10:90 to 30:70) gave product **E** (0.30 g, 21 %) as a white solid: R_f = 0.6 (EtOAc/hexane 30:70); ν_{max} = 3402,

2922, 2852, 2090, 1695, 1521, 1447, 1220, 1193, 742 cm^{-1}; ^1H NMR (400 MHz, $(CD_3)_2CO$, 25 °C, TMS): δ = 0.84 (s, 3 H; 18-H_3) 0.88 (t, $^3J(H,H)$ = 7.0 Hz, 3 H 20'-H_3), 0.91 (d, $^3J(H,H)$ = 6.2 Hz, 3 H; 21-H_3), 1.01 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(C\mathbf{H}_2)_{17}CH_3$), 3.31 – 3.41 (m, 1 H; 3β-H), 3.95 (bs, 1 H; 7β-H), 3.99 (t, $^3J(H,H)$ = 6.6 Hz, 2 H; 1'-H_2), 4.24 – 4.29 (m, 1 H; 12β-H), 5.06 – 5.16 (m, 4 H; 2 x CCH_2O), 5.97 – 6.02 (m, 2 H; 2 x CH-N**H**CO), 6.89 – 6.95 (m, 4 H), 7.19 – 7.25 (m, 2 H), 7.51 (s, 1 H; Ar-NHCO), 7.56 – 7.64 (m, 4 H), 7.67 (s, 1 H; Ar-NHCO), 8.50 – 8.55 (m, 2 H); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 °C, TMS): δ = 14.0 (18-CH_3), 14.4 (20'-CH_3), 18.1 (21-CH_3), 23.2 (19'-CH_2), 23.4 (19-CH_3), 24.3 (CH_2), 26.7 (18'-CH_2), 27.4 (CH_2), 27.6 (2 x CH_2), 28.0 (17'-CH_2), 29.3 (CH), 30.0 (16'-CH_2), 30.1 (15'-CH_2), 30.3 (14'-CH_2), 30.3 (13'-CH_2), 30.4 (12'-CH_2), 30.4 ($COOCH_2CH_2(\mathbf{C}H_2)_9(CH_2)_8CH_3$), 31.9 (2'-$CH_2$), 32.1 ($CH_2$), 32.7 ($CH_2$), 33.2 ($CH_2$), 35.6 (CH), 35.9 ($CH_2$), 36.0 ($CH_2$), 38.2 (CH), 39.7 (10-C), 42.6 (CH), 44.7 (CH), 45.7 (13-C), 47.6 (CH), 49.3 (7-CH), 52.8 (12-CH), 62.8 (3-CH), 64.7 (1'-CH_2), 72.0 ($C\mathbf{C}H_2O$), 72.1 ($C\mathbf{C}H_2O$), 112.5 (Ar-CH), 112.7 (Ar-CH), 118.9 (Ar-CH), 119.2 (Ar-CH), 121.7 (Ar-CH), 121.8 (Ar-CH), 122.1 (2 x Ar-CH), 130.0 (Ar-CH), 131.0 (Ar-CH), 131.4 (2 x Ar-C), 131.4 (Ar-CH), 131.5 (Ar-CH), 138.0 (Ar-C), 138.1 (Ar-C), 147.5 (Ar-C), 147.5 (Ar-C), 154.9 (NHCONH), 155.3 (NHCONH), 174.0 ($\mathbf{C}OOCH_2(CH_2)_{18}CH_3$); MS (ES+): m/*z* (%): 1086 (100) $[M + H]^+$.

Flushing the column (eluent EtOAc/hexane 1:1) allowed the recovery of starting material (0.45 g, 34 %).

Propylene linked macrocyclic methanesulfonate 2.$MeSO_3^-$ via iodide salt 2.I^-. To a solution of azido compound **D** (0.081 g, 0.079 mmol) in dry THF (0.8 mL) was added trimethylphosphine (0.158 mL as a 1 M solution in THF, 0.158 mmol). The reaction solution was stirred for 3 h before water (15 μL, 0.015 g, 0.8 mmol) was added and the solution was stirred for a further 48 h. Solvent was removed in vacuo and the residue dried azeotropically with toluene/DCM. To the dried crude amino product was added potassium hydrogenphosphate (0.029 g, 0.166 mmol), dry acetonitrile (1.6 mL), dry THF (0.8 mL) and iodomethane (0.16 mL). The resultant mixture was stirred for 48 h before solvent was removed in vacuo. The crude product was then re-dissolved in DCM, washed with water and the solvent removed in vacuo. Subsequent

purification by flash column chromatography (eluent DCM/methanolic ammonia 97:3 to 96:4) gave quaternary ammonium iodide **2.I**$^-$ (0.060 g, 66 %) as a white solid: R_f = 0.3 (DCM/methanolic ammonia 95:5); ^{1}H NMR (400 MHz, $(CD_3)_2CO$, 25 ^{o}C, TMS): δ = 0.82 – 0.90 (m, 9 H; 18-H_3 + 20'-H_3 + 21-H_3), 1.07 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(C\mathbf{H}_2)_{17}CH_3$), 2.59 – 2.70 (m, 1 H), 2.72 – 2.82 (m, 1 H), 2.80 – 2.88 (m, 2 H; $OCH_2C\mathbf{H}_2CH_2O$), 3.05 – 3.16 (m, 1 H; 3β-H), 3.32 (s, 9 H; $(CH_3)N^+$), 3.97 (t, ^3J(H,H) = 6.6 Hz, 2 H; 1'-H_2), 4.09 – 4.52 (m, 6 H, 7β-H + 12β-H + 2 x Ar-OCH_2), 6.78 – 6.92 (m, 4 H), 6.97 – 7.01 (m, 2 H), 7.23 – 7.29 (m, 2 H; 2 x CH-N**H**CO), 8.20 – 8.31 (m, 2 H), 8.49 (s, 1 H; Ar-NHCO), 8.60 (s, 1 H; Ar-NHCO)

To a solution of quaternary ammonium iodide **2.I**$^-$ (0.06 g, 0.05 mmol) in THF (1 mL), was added a solution of silver methanesulfonate (0.0116 g, 0.057 mmol) in water (0.2 mL). The resultant solution was stirred for 2 h during which period a precipitate of silver iodide formed. The solvent was removed in vacuo and further dried under high vacuum. The mixture was suspended in DCM and filtered through a cotton wool plug to remove inorganics. Removal of the solvent in vacuo gave methanesulfonate **2.MeSO$_3$**$^-$ as a white solid (0.055 g, quantitative): ν_{max} = 3346, 2922, 2853, 1694, 1532, 1448, 1216, 1165, 745 cm^{-1}; ^{1}H NMR (400 MHz, $(CD_3)_2CO$, 25 ^{o}C, TMS): δ = 0.79 – 0.91 (m, 9 H; 18-H_3 + 20'-H_3 + 21-H_3), 1.06 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(C\mathbf{H}_2)_{17}CH_3$), 2.70 (s, 3 H; $CH_3SO_3^-$), 3.12 – 3.25 (m, 1 H; 3β-H), 3.22 (s, 9 H; $(CH_3)N^+$), 3.95 (t, ^3J(H,H) = 6.8 Hz, 2 H; 1'-H_2), 4.05 – 4.10 (m, 1 H; 7β-H), 4.11 – 4.76 (m, 5 H, 12β-H + 2 x Ar-OCH_2), 6.72 – 6.79 (m, 1 H; CH-N**H**CO), 6.82 – 6.92 (m, 4 H), 6.93 – 7.04 (m, 2 H), 7.28 – 7.32 (m, 1 H; CH-N**H**CO), 8.04 – 8.14 (m, 2 H), 8.45 (s, 1 H; Ar-NHCO), 8.51 (s, 1 H; Ar-NHCO); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 ^{o}C, TMS): δ = 14.2 (18-CH_3), 14.4 (20'-CH_3), 19.4 (21-CH_3), 22.4 (CH_2), 23.3 (19-CH_3), 23.3 (19'-CH_2), 25.0 (CH_2), 26.3 (18'-CH_2), 26.7 (CH_2), 27.4 (17'-CH_2), 29.5 (CH_2), 30.0 (16'-CH_2), 30.2 (CH), 30.2 (15'-CH_2), 30.3 (14'-CH_2), 30.3 (13'-CH_2), 30.4 (12'-CH_2), 30.4 ($COOCH_2CH_2(\mathbf{C}H_2)_9(CH_2)_8CH_3$), 31.1 ($OCH_2\mathbf{C}H_2CH_2O$), 31.3 ($CH_2$), 31.8 (2'-$CH_2$), 32.6 (2 x CH_2), 34.5 (CH), 35.0 (CH_2), 36.5 (10-C), 36.9 (CH_2), 39.4 (CH), 39.9 ($CH_3SO_3^-$), 43.6 (CH), 44.0 (CH), 47.4 (CH),

47.7 (13-C), 48.4 (7-CH), 51.9 (12-CH), 52.3 $(CH_3)_3N^+$, 64.5 (1'-CH_2), 67.0 (Ar-OCH_2), 68.3 (Ar-OCH_2), 77.9 (3-CH), 114.6 (Ar-CH), 116.4 (Ar-CH), 121.1 (Ar-CH), 121.8 (Ar-CH), 121.8 (Ar-CH), 122.1 (Ar-CH), 122.1 (Ar-CH), 122.4(Ar-CH), 133.0 (Ar-C), 133.4 (Ar-C), 149.0 (Ar-C), 149.7 (Ar-C), 156.6 (NHCONH), 157.6 (NHCONH), 174.1 (**C**$OOCH_2(CH_2)_{18}CH_3$); MS (ES+): m/*z* (%): 1039 (100) $[M]^+$, MS (ES-): m/*z* (%): 80 (100) $[SO_3]^-$, 95 (70) $[EtSO_3]^-$; HRMS (ES+): m/*z* calculated for $[M]^+$ = 1038.7981, found 1038.8029.

m-Xylylene linked macrocyclic methanesulfonate 3.$MeSO_3^-$ via iodide salt 3.I^-. To a solution of azide **E** (0.10 g, 0.092 mmol) in dry THF (1 mL) was added trimethylphosphine (0.184 mL as a 1 M solution in THF, 0.184 mmol). The reaction solution was stirred for 3 h before water (17 μL, 0.017 g, 0.92 mmol) was added and the solution was stirred for a further 48 h. Solvent was removed in vacuo and the residue dried azeotropically with toluene/DCM. To the dried crude amino product was added potassium hydrogen phosphate (0.034 g, 0.193 mmol), dry acetonitrile (2 mL), dry THF (1 mL) and iodomethane (0.2 mL). The resultant mixture was stirred overnight before solvent was removed in vacuo. The crude product was then re-dissolved in DCM, washed with water and the solvent removed in vacuo. Subsequent purification by flash column chromatography (eluent DCM/methanolic ammonia 95:5 to 94:6) gave product ammonium iodide **3.I^-** (0.074 g, 75 %) as a white solid: R_f = 0.25 (DCM/methanolic ammonia 95:5); ^{1}H NMR (400 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 0.81 (s, 3 H; 18-H_3) 0.85 – 0.90 (m, 6 H; 20'-H_3 + 21-H_3), 1.00 (s, 3 H; 19-H_3), 1.29 (s, 34 H; $COOCH_2CH_2(CH_2)_{17}CH_3$), 2.36 – 2.46 (m, 1 H), 2.59 – 2.71 (m, 1 H), 3.08 (s, 9 H; $(CH_3)_3N^+$), 3.17 – 3.28 (m, 1 H; 3β-H), 3.90 (bs, 1 H; 7β-H), 3.96 (t, ^{3}J(H,H) = 6.4 Hz, 2 H; 1'-H_2), 4.26 – 4.34 (m, 1 H; 12β-H), 5.06 – 5.26 (m, 4 H; 2 x CCH_2O), 6.85 – 6.95 (m, 4 H), 7.13 – 7.54 (m, 6 H), 8.22 – 8.30 (m, 2 H), 8.41 (s, 1 H; Ar-NHCO), 8.62 (s, 1 H; Ar-NHCO)

To a solution of ammonium iodide **3.I^-** (0.049 g, 0.042 mmol) in THF (0.5 mL), was added a solution of silver methanesulfonate (0.010 g, 0.047 mmol) in water (0.1 mL). The resultant solution was stirred for 2 h during which period a precipitate of silver iodide formed. The solvent was removed in vacuo and further

dried under high vacuum. The mixture was suspended in DCM and filtered through a cotton wool plug to remove inorganics. Removal of the solvent in vacuo gave methanesulfonate **3.MeSO$_3^-$** as a white solid (0.045 g, quantitative): ν_{max} = 3336, 2922, 2853, 1685, 1531, 1447, 1217, 1172, 745 cm^{-1}; ^{1}H NMR (400 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 0.84 – 0.90 (m, 9 H; 18-H$_3$ + 20'-H$_3$ + 21-H$_3$), 1.02 (s, 3 H; 19-H$_3$), 1.29 (s, 34 H; COOCH$_2$CH$_2$(C**H**$_2$)$_{17}$CH$_3$), 2.42 – 2.54 (m, 1 H), 2.90 (s, 3 H; $CH_3SO_3^-$), 3.08 (s, 9 H; $(CH_3)_3N^+$), 3.12 – 3.22 (m, 1 H; 3β-H), 3.87 (t, ^{3}J(H,H) = 6.6 Hz, 2 H; 1'-H$_2$), 4.06 (bs, 1 H; 7β-H), 4.21 – 4.28 (m, 1 H; 12β-H), 4.79 – 5.44 (m, 4 H; 2 x CCH$_2$O), 6.89 – 7.20 (m, 6 H), 7.15 – 7.30 (m, 2 H), 7.45 – 7.49 (m, 2 H), 8.13 – 8.20 (m, 1 H), 8.35 – 8.46 (m, 1 H), 8.45 (s, 1 H; Ar-NHCO), 8.48 (s, 1 H; Ar-NHCO); ^{13}C NMR (100 MHz, $(CD_3)_2CO$, 25 ^oC, TMS): δ = 14.2 (18-CH$_3$), 14.4 (20'-CH$_3$), 18.5 (21-CH$_3$), 22.2 (CH$_2$), 23.4 (19'-CH$_2$), 23.5 (19-CH$_3$), 24.4 (CH$_2$), 26.7 (18'-CH$_2$), 28.1 (17'-CH$_2$), 28.6 (2 x CH$_2$), 30.0 (16'-CH$_2$), 30.2 (15'-CH$_2$), 30.3 (14'-CH$_2$), 30.3 (13'-CH$_2$), 30.4 (12'-CH$_2$), 30.4 (COOCH$_2$CH$_2$(**C**H$_2$)$_9$(CH$_2$)$_8$CH$_3$), 30.8 (CH), 31.4 (CH$_2$), 31.8 (2'-CH$_2$), 32.7 (2 x CH$_2$), 35.5 (CH), 35.7 (CH$_2$), 36.7 (CH$_2$), 38.6 (CH), 39.3 (10-C + $CH_3SO_3^-$), 44.1 (CH), 45.4 (CH), 46.4 (CH), 46.4 (13-C), 48.7 (7-CH), 52.2 (12-CH), 52.5 (3-CH + $(CH_3)_3N^+$), 64.5 (1'-CH$_2$), 73.0 (2 x C**C**H$_2$O), 113.4 (2 x Ar-CH), 120.5 (2 x Ar-CH), 121.4 (2 x Ar-CH), 122.2 (Ar-CH), 122.4 (Ar-CH), 122.6 (Ar-CH), 128.9 (2 x Ar-CH), 130.1 (Ar-CH), 131.7 (2 x Ar-C), 139.0 (2 x Ar-C), 148.5 (2 x Ar-C), 156.0 (NHCONH), 156.4 (NHCONH), 174.0 (**C**OOCH$_2$(CH$_2$)$_{18}$CH$_3$); MS (ES+): m/*z* (%): 1101 (100) $[M]^+$, MS (ES-): m/*z* (%): 80 (100) $[SO_3]^-$, 95 (20) $[EtSO_3]^-$; elemental analysis calculated (%) for $C_{70}H_{109}N_5O_9S$: C 70.26, H 9.18, N 5.85; found C 70.24, H 9.50, N 5.54

Measurement of K for eqn (1) - general procdeure

A solution of quaternary ammonium salt[3] (0.001 M, 2 mL) in chloroform was added to a sample tube (30 mL). To this was added an aqueous solution of $NaEtSO_3$ (0.05 M, 5 mL) and an aqueous solution of NaX (5 mL; X^- = test substrate, concentrations varied such that eqn (1) is roughly in balance). A small magnetic stirring bar was then added to the tube and the lower half of the tube was immersed in a water bath at 303 K. The tube was stoppered and the contents stirred vigorously. After 30 minutes stirring was stopped and the two phases were allowed to separate. The organic phase was taken up with a pipette and filtered through a Whatman 1PS hydrophobic filter into a round bottomed flask. Solvent was then evaporated under reduced pressure. The resultant solid was dissolved in deuterated solvent (DMSO-d6, CD_3OD, or MeCN-d3; chosen to maximise peak separation), and a solution of tetraphenylphosphonium bromide (45 μL, 0.1 m) in the deuterated solvent was added.[4] The ^{1}H-NMR spectrum was recorded, and the ethylene peaks of ethanesulfonate were integrated relative to the steroid 1'-H_2 signal. The result was used to calculate the position of the anion exchange equilibrium (eqn (1)), assuming that cation in excess over ethanesulfonate must be accompanied by X^-.

Molecular modelling

Modelling was performed using Macromodel 7.1, accessed via Maestro 3.0, employing the MMFFS force field and GBSA chloroform solvation. All receptors were modelled as their methyl ester analogues. Selected results are illustrated in Figures S1-S4 below.

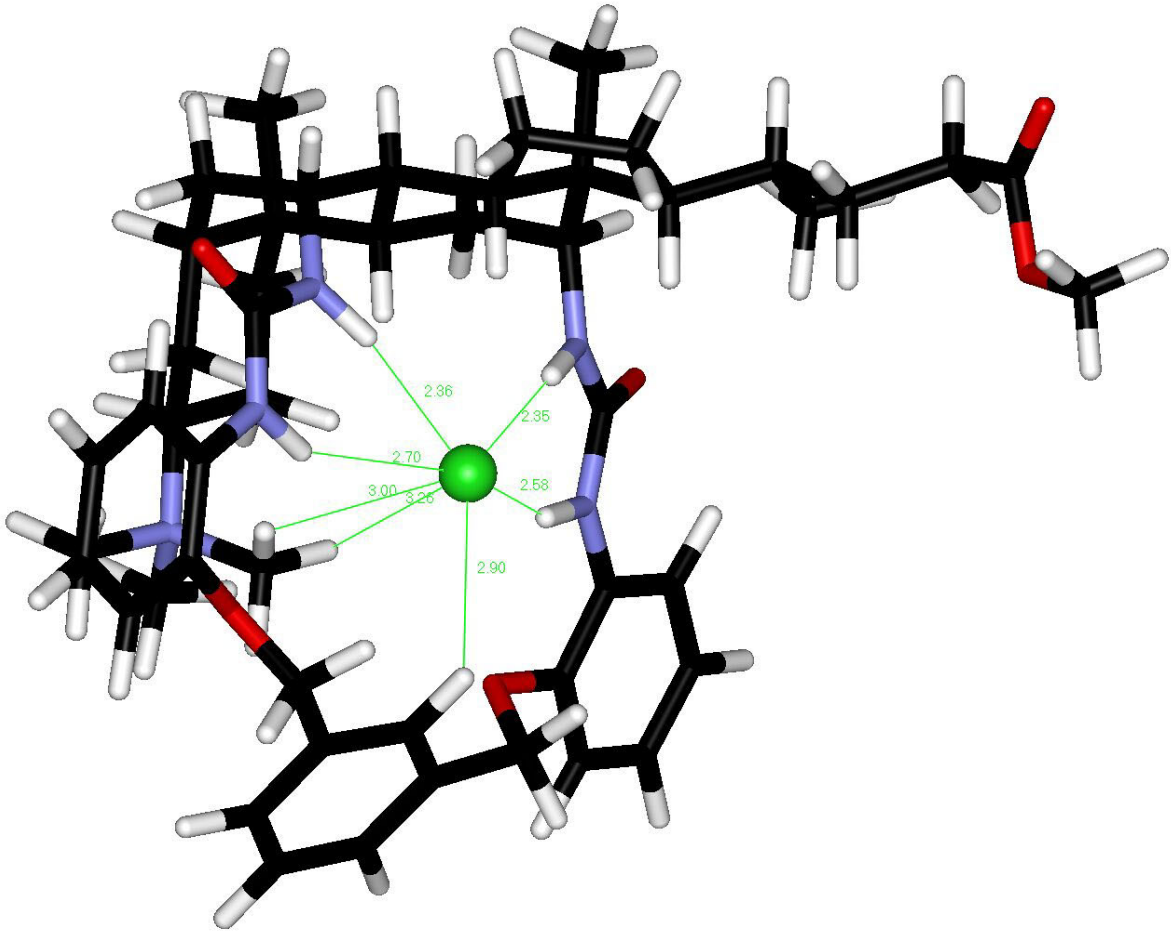


Figure S1. Model of **3**.Cl^-. The structure, which is the same as that shown in Figure 3, was derived from a Monte Carlo Molecular Mechanics conformational search. This image shows the NH···Cl^- bond lengths (2.35-2.70 Å) and selected CH···Cl^- contacts (2.90-3.26 Å).

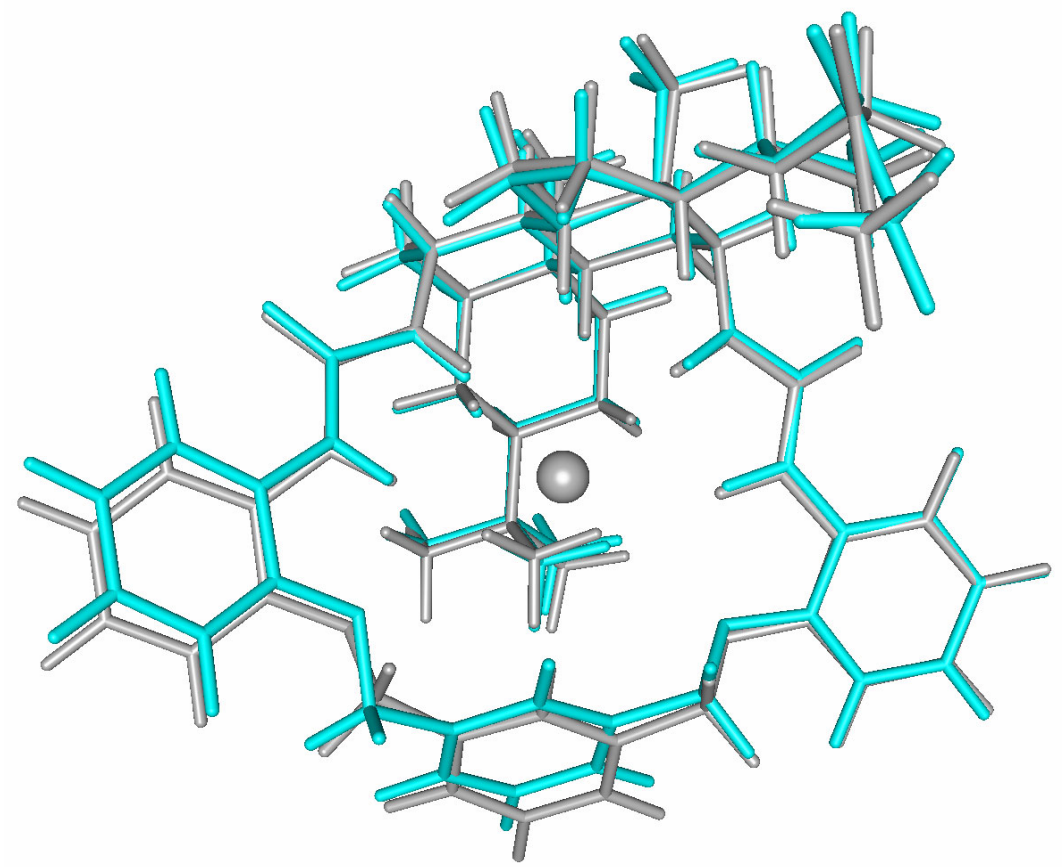


Figure S2. Superposition of **3**.Cl^- and **3**. The structures are viewed down the long axis of the steroid (methyl esters in foreground). Shown in silver is the model of **3**.Cl^- from Figure S1. Shown in turquoise is the structure obtained by removing the chloride and reminimising. The two structures are very similar, implying that the introduction of Cl^- imposes little or no strain on the receptor.

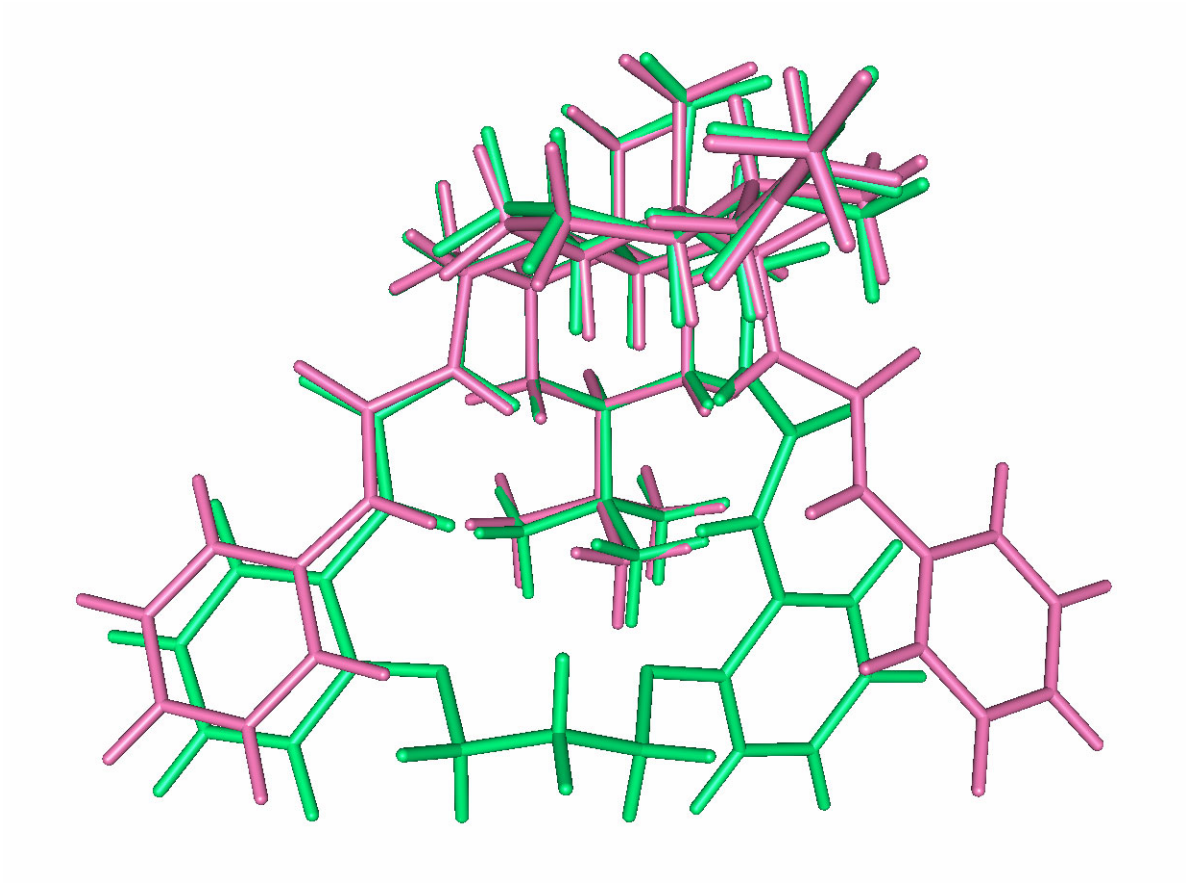


Figure S3. Superposition of acyclic receptor **1** (pink) and $(CH_2)_3$-bridged receptor **2** (green) viewed down the long axis of the steroid (methyl esters in foreground). **1** was minimised from a conformation based on a crystal structure.[5] **2** was subjected to a Monte Carlo Molecular Mechanics conformational search. The binding site of **2** is considerably smaller than that of **1**, the ureas being pulled together by the propylene bridge.

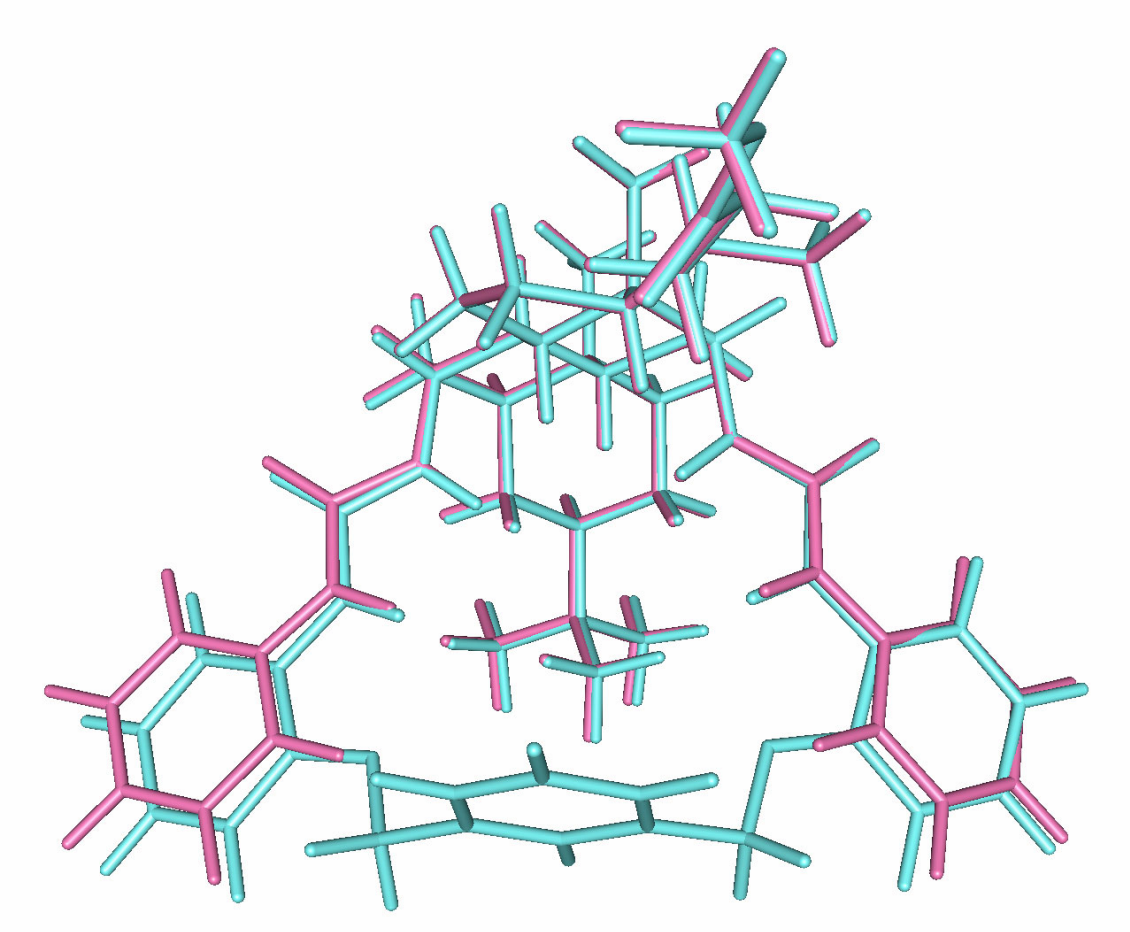


Figure S4. Superposition of methyl ester analogues of acyclic receptor **1** (pink) and xylyl-bridged receptor **3** (turquoise) viewed down the long axis of the steroid (methyl esters in foreground). **1** was minimised from a conformation based on a crystal structure.[5] **3** was subjected to a Monte Carlo Molecular Mechanics conformational search. The positions of the quaternary ammonium and urea NH groups are closely similar.

References and Notes

1. A. J. Ayling, M. N. Pérez-Payán and A. P. Davis, *J. Am. Chem. Soc.*, 2001, **123**, 12716.
2. This cyclisation proceeded more cleanly when both DBU and DMAP were present than when DBU was used alone. The role of the DMAP is uncertain, but it seemed to inhibit the formation of one or more side-products containing olefinic protons.
3. Receptors **1**-**3** were used as methanesulfonate salts.
4. Addition of the bromide salt improved the resolution of the spectrum, allowing more accurate integration.

5. A. L. Sisson, J. P. Clare, L. H. Taylor, J. P. H. Charmant and A. P. Davis, *Chem. Commun.*, 2003, 2246.