

Electronic Supplementary Information

2'-O-(1,3,6,9,12-Pentaoxacyclopentadecan-2-ylmethyl)-aminocarbonyl-3',5'-O-(tetraisopropylidisiloxan-1,3-diyl)-uridine (3e).

Method B. To the solution of **2**, prepared from 974 mg (2.0 mmol) of **1** and CDI (325 mg, 2.0 mmol) in DCM (5 cm³) 2-aminomethyl-15-crown-5 (514 mg, 2.0 mmol) was added under nitrogen. The mixture was allowed to react for 3 months at ambient temperature, then diluted with CHCl₃ (100 cm³), washed with water (100 cm³), 5% citric acid (100 cm³), and water (100 cm³), dried (Na₂SO₄), evaporated, and the residue was chromatographed on silica gel (stepwise gradient of 2→3→4→6→8→10% MeOH in CHCl₃, v/v). Yield 679 mg (44.5%).

N α -[3',5'-O-(Tetraisopropylidisiloxan-1,3-diyl)uridin-2'-O-ylcarbonyl]-L-leucyl-L-phenylalaninamide (3f).

Method B. To the solution of **2** (2.0 mmol), prepared in DCM (5 cm³), H-Leu-Phe-NH₂ hydrochloride (942 mg, 3.0 mmol) and DIEA (0.610 cm³, 3.5 mmol) were added under nitrogen. The mixture was allowed to react for 3 months at ambient temperature, then diluted with CHCl₃ (100 cm³), washed with water (100 cm³), 5% citric acid (100 cm³), and water (100 cm³), dried (Na₂SO₄), evaporated, and the residue was chromatographed on silica gel (stepwise gradient of 33→50% EtOAc in CHCl₃, then 33→50% acetone in CHCl₃-EtOAc (1:1), v/v). Yield 364 mg (23.0%).

2'-O-[4,9-bis(Trifluoroacetyl)-12-(trifluoroacetylamino)-4,9-diazadodecan-1-ylaminocarbonyl]-3',5'-O-(tetraisopropylidisiloxan-1,3-diyl)uridine (3p).

Method B. The solution of **2** (2.904 g, 5.0 mmol) in dry DCM (50 cm³) was added dropwise to the stirred, ice-cooled solution of spermine (1.012 g, 5.0 mmol) in DCM (100 cm³) within 30 min. After 1 h the mixture was allowed to warm to 20°C, evaporated to dryness, coevaporated with THF (30 cm³), dissolved in dry THF (20 cm³), and *S*-ethyl trifluoroacetate (5.0 cm³, 39 mmol) was added in one portion. The mixture was kept overnight, evaporated to dryness (*STENCH!*) and the residue was chromatographed on silica gel (stepwise gradient of 0→5→10→15→20% acetone in CHCl₃-EtOAc, 1:1, v/v/v). Compound **1** (526 mg, 21.6%) was eluted in the first fractions. The desired **3p** (white foam; 2.208 g, 44.0%) was eluted with 5→10% acetone (in CHCl₃-EtOAc, 1:1 v/v). Further increase of the eluent polarity (15→20% acetone) gave the minor product, **1,12-bis[3',5'-O-(tetraisopropylidisiloxan-1,3-diyl)uridin-2'-O-ylcarbonylamino]-4,9-bis(trifluoroacetyl)-4,9-diazadodecane (3r)** as a white foam (1.147 g, 32.3%). *R*_f 0.53 (EtOAc). MALDI-TOF (2,5-DHBA): [M+H]⁺ calc. 1420.77, found 1421.18, [M+Na]⁺ calc. 1442.75, found 1442.93, [M+K]⁺ calc. 1458.86, found 1458.56. ¹H-NMR: 11.41 (s, 2H, *H*-3), 7.69 (d, 2H, *J*_{5,6} = 8.0 Hz, *H*-6), 7.50, 7.44 (2 br. t, 2H, OCONH, rotamers), 5.65 (s, 2H, *H*-1'), 5.59 (d, 2H, *J*_{5,6} = 8.0 Hz, *H*-5), 5.31 (m, 2H, *H*-2'), 4.49 (m, 2H, *H*-3'), 4.12–3.88 (m, 4H, *H*-5'), 3.83 (m, 2H, *H*-4'), 3.42–3.26 (m, 8H[#], CH₂NCH₂), 2.98 (m, 4H, NHCH₂), 1.85–1.60 (m, 4H, CH₂CH₂NH), 1.51 (m, 4H, CH₂CH₂CH₂CH₂), 1.08–0.80 (m, 56H, Pr^r).

5'-O-(4,4'-Dimethoxytrityl)-2'-O-(1,3,6,9,12-pentaoxa-cyclopentadecan-2-ylmethylaminocarbonyl)-uridine (5e). The fast eluting compound, **3',5'-O,O'-bis(4,4'-dimethoxytrityl)-2'-O-(1,3,6,9,12-pentaoxacyclopentadecan-2-yl)methylaminocarbonyluridine** was isolated (293 mg, 13.0%); white amorphous solid, *R*_f 0.36 (CHCl₃-MeOH, 17:3). MALDI-TOF (2,4,6-THAP): [M+Na]⁺ calc. 1147.22, found 1147.84, [M+K]⁺ calc. 1163.33, found 1163.93. ¹H-NMR: 11.41 (s, 1H, *H*-3), 7.65 (m, 1H, OCONH), 7.50 (d, 1H, *J*_{5,6} = 8.1 Hz, *H*-6), 7.29–7.10 (m, 18H, ArH), 6.86–6.72 (m, 8H, ArH), 6.08 (d, 1H, *J*_{1',2'} = 6.1 Hz, *H*-1'), 5.35 (d, 1H, *J*_{5,6} = 8.1 Hz, *H*-5), 5.02 (m, 1H, *H*-2'), 4.24 (m, 1H, *H*-4'), 3.71 (m, 12H, CH₃), 3.67–3.39 (m, 21H[#], *H*-5', CH(CH₂OCH₂)₄CH₂), 3.05 (m, 2H, CH₂N), 2.87 (m, 1H, *H*-3').

N α -[5'-O-(4,4'-Dimethoxytrityl)uridin-2'-O-ylcarbonyl]-L-leucyl-L-phenylalaninamide (5f). Compound **3f** (593 mg, 0.75 mmol) was desilylated and tritylated according to general procedures. The desired product was purified by column chromatography on silica gel (stepwise gradient of 3→5→10% MeOH in CHCl₃-acetone (2:1) + 0.5% Et₃N (v/v/v)). Yield 485 mg (76.0%), white foam. *R*_f 0.30 (acetone). MALDI-TOF MS (2,4,6-THAP): [M+Na]⁺ calc. 872.35, found 872.62, [M+K]⁺ calc. 888.32, found 888.62. ¹H-NMR: 11.45 (s, 1H, *H*-3), 7.90 (d, 1H, *J* = 8.4 Hz, NHCHCH₂Ph), 7.72 (d, 1H, *J*_{5,6} = 8.1 Hz, *H*-6), 7.48–7.10 (m, 17H, ArH, OCONH, NH₂), 6.89 (d, 4H, *J* = 8.8 Hz, ArH), 5.90 (d, 1H, *J*_{1',2'} = 4.4 Hz, *H*-1'), 5.52 (d, 1H, *J* = 5.9 Hz, 3'-OH), 5.43 (d, 1H, *J*_{5,6} = 8.1 Hz, *H*-5), 5.13 (m, 1H, *H*-2'), 4.47 (m, 1H, CHCH₂Ph), 4.37 (m, 1H, *H*-3'), 4.17 (m, 1H, CHBu^t), 3.98 (m, 1H, *H*-4'), 3.73 (s, 6H, OCH₃), 3.52–3.10 (m, 2H, *H*-5'), 3.05–2.70 (m, 2H, CH₂Ph), 1.57–1.22 (m, 3H, CH₂CHMe₂), 0.86–0.69 (m, 6H, CH₂CH(CH₃)₂).

N α -[3'-O-(*N,N*-Diisopropylamino-2-cyanoethoxyphosphinyl)-5'-O-4,4'-dimethoxytrityluridin-2'-O-ylcarbonyl]-L-leucyl-L-phenylalaninamide (6f). Chromatography: 50→100% acetone in CHCl₃-EtOAc (1:1) + 1% Et₃N (v/v/v). Yield 271 mg (51.6%), *R*_f 0.13, 0.20. MALDI-TOF (2,6-DHAP-citrate): [M+H]⁺ calc. 1051.15, found 1053.41, [M+Na]⁺ calc. 1072.46, found 1071.67. ¹H-NMR: 11.47 (s, 1H, *H*-3), 8.08 (d, 2H, *J* = 11.7 Hz, NH₂), 7.83 (d, 1H, *J* = 8.6 Hz, NHCHCH₂Ph), 7.70 (d, 1H, *J*_{5,6} = 8.1 Hz, *H*-6), 7.43–7.07 (m, 15H, ArH, OCONH), 6.88 (d, 4H, *J* = 8.8 Hz, ArH), 5.86 (d, 1H, *J*_{1',2'} = 4.8 Hz, *H*-1'), 5.46 (d, 1H, *J*_{5,6} = 7.8 Hz, *H*-5), 5.32 (t, 1H, *J* = 5.3 Hz, *H*-2'), 4.64 (m, 1H, CHCH₂Ph), 4.46 (m, 1H, *H*-3'), 4.17 (m, 1H, CHBu^t), 3.95 (m, 1H, *H*-4'), 3.73 (s, 6H, OCH₃), 3.58–3.46 (m, 4H, POCH₂, CHN), 3.04–2.99 (m, 2H, *H*-5'), 2.87–2.81 (m, 2H, CH₂Ph), 2.59 (m, 2H, CH₂CN), 1.23–0.98 (m, 12H, NCHCH₃), 0.83–0.72 (m, 3H, CH₂CHMe₂), 0.65–0.59 (m, 6H, CH₂CH(CH₃)₂). ³¹P (DMSO-*d*₆): 148.926.

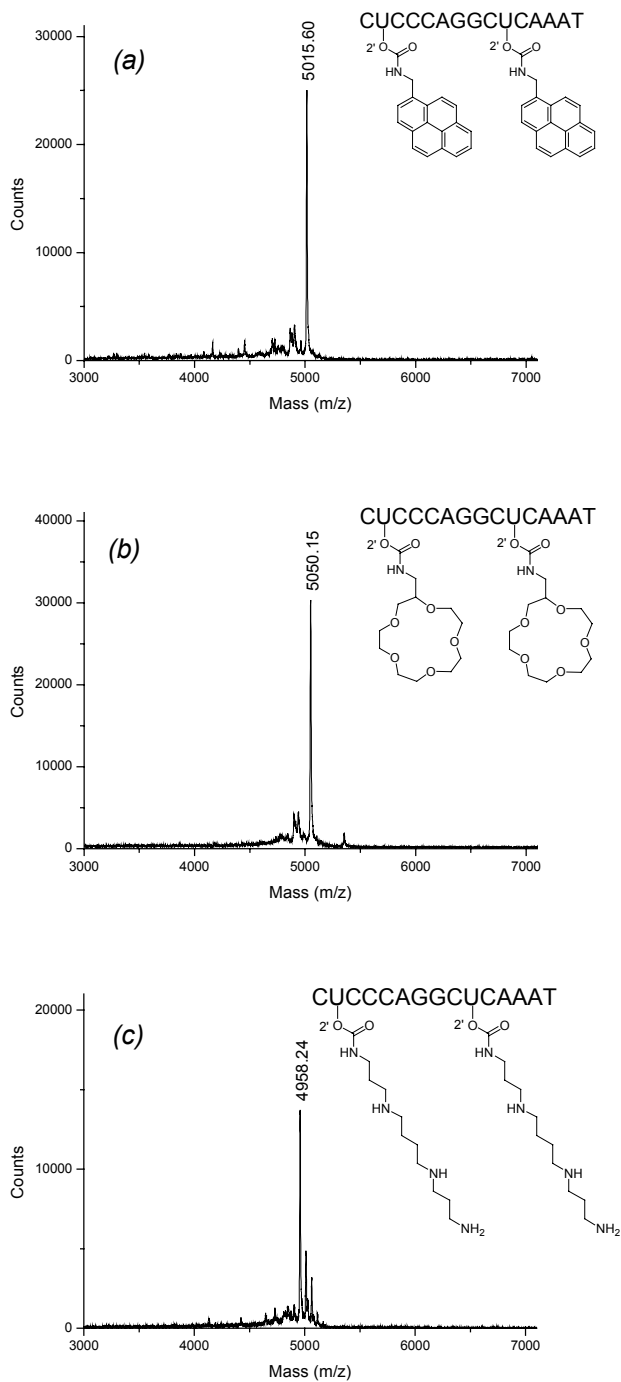


Fig. 1 Examples of MALDI-TOF mass spectra of 2'-carbamate-modified oligonucleotides: *a*) bis-pyrene (ON26), *b*) bis-15-crown-5 (ON27), and *c*) bis-spermine (ON31).