Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

Supporting Information

Synthesis and *in vitro* preliminary evaluation of prostate-specific membrane antigen targeted upconversion nanoparticles as a step towards radio/fluorescence-guided surgery of prostate cancer

Axel Cordonnier[a,b], Damien Boyer[b], Sophie Besse[a], Rodolphe Valleix[b], Rachid Mahiou[b], Mercedes Quintana[a], Arnaud Briat[a], Mhammed Benbakkar[d], Frédérique Penault-Llorca[a,c], Aurélie Maisonial-Besset[a], Benoit Maunit[a], Sébastien Tarrit[a], Magali Vivier[a], Tiffany Witkowski[a], Leslie Mazuel[a], Françoise Degoul[a], Elisabeth Miot-Noirault[a], Jean-Michel Chezal[a,*]

[a]Université Clermont Auvergne, Inserm, Imagerie Moléculaire et Stratégies Théranostiques, UMR 1240, F-63000 Clermont-Ferrand, France

[b]Université Clermont Auvergne, Clermont Auvergne INP, CNRS, F-63000 Clermont-Ferrand, France

[c]Department of Pathology and Biopathology, Jean Perrin Comprehensive Cancer Centre, Clermont-Ferrand, France

[d]Université Clermont Auvergne, CNRS, Laboratoire Magmas et Volcans, UMR 6524, F-63000 Clermont-Ferrand, France

*Corresponding author.

E-mail address: j-michel.chezal@uca.fr

Table of contents

1. Synthetic procedures

1.1. Materials and methods

All commercially available reagents and solvents were purchased at the following commercial suppliers: Sigma Aldrich, Alpha Aesar, ABX, Acros Organics, Fisher Scientific or Carlo Erba Reagents. Tetrahydrofuran was dried over a Pure Solv™ Micro Solvent Purification System (Sigma-Aldrich) and whenever necessary, other solvents were dried using common techniques[1]. Polyethylene glycol 2000 (average M_n 1900-2200) was purchased from Sigma while PEG methyl ethers 1000 (average M_n 950-1050) and 2000 (average M_n 2000) were obtained from TCI and Aldrich, respectively. Analytical thin layer chromatography (TLC) was performed on precoated silica gel 60 F_{254} or neutral aluminium oxide 60 F_{254} plates (Merck or Macherey-Nagel) and visualized under UV light (254 nm) and/or developed with phosphomolybdic acid (8 wt%) in ethanol and/or Dragendorff's reagent for pegylated derivatives. Flash column chromatography was performed on silica gel 60A normal phase, 35-70 μm (Merck or SDS) or neutral aluminium oxide 90 standardized, 63-200 μm (Merck). Uncorrected melting points (mp) were recorded on an electrothermal capillary Digital Melting Point Apparatus IA9100 (Bibby Scientific). NMR spectra (200.13 or 500.13 MHz for ^{1}H and 50.32 or 125.76 MHz for ^{13}C) were recorded on Bruker Avance 200 or 500 instrument with chemical shift values (δ) expressed in parts per million (ppm) relative to residual solvent as standard. Coupling constants (J) are given in Hz; ^{31}P NMR spectra (202.6 MHz) were recorded on a Bruker Avance 500 apparatus using a solution of phosphoric acid (85% wt) in D_2O (1/99; v/v) as a reference standard (0 ppm) in a coaxial insert. Infrared spectra (IR) were recorded in the range 4000-440 cm^{-1} on a Nicolet IS10 (Fisher Scientific) with attenuated total reflectance (ATR) accessory. Low molecular weight organic compounds were analysed by High-Resolution Mass Spectrometry (HRMS) in positive mode (Waters® Micromass® Q-Tof micro™ Mass Spectrometer) and/or by Electrospray Ionization Mass Spectrometry (ESI-MS) recorded on a Esquire-LC ion trap mass spectrometer (Bruker Daltonics). For ESI-MS, the analysis of samples was performed at a final concentration between 1 and 10 pmol/μL. Each ESI-MS spectrum was recorded by averaging of 10 spectra. Pegylated derivatives were characterized by Matrix Assisted Laser Desorption Ionization Time-of-Flight (MALDI-TOF) mass spectrometry, carried out on a MALDI-TOF/TOF Autoflex Speed (Bruker Daltonics). PEGs stock solutions of 50 mg/mL were prepared in methanol. Three PEGs working solutions at 5, 1 and 0.15 mg/mL were subsequently prepared in methanol-water (4/1, v/v). The compound 2,5-dihydroxybenzoic acid (DHB, >99%, Sigma) served as matrix at a concentration of 10 mg/mL in water. The DHB matrix solution was subsequently mixed in a ratio of 9:1 (v/v) with the aqueous solution of the

cationizing agent sodium iodide at 20 mg/mL. Three samples deposits were realized on the MALDI-MS target adding 1 µL of PEG working solution followed by 0.5 µL of the matrix and cationizing agent mixture solution. Spectra acquisition was realized in the positive ion reflectron mode with an ion source voltage of 19.00 kV, a laser power of 75% and smart-beam set at 3_medium 1000 Hz. A number of 4000 shots were averaged for each spectrum, in the mass-range between 400 and 3500 *m*/*z*. Calibration was performed in a close external mode using standards of PEG 1000, PEG 2000 or their monomethyl ether derivative solutions. Data analyses were conducted with Bruker's flex Analysis software.

Preparative RP-HPLC purifications were carried out on a CombiFlash EZ prep system (Teledyne Isco) equipped with a UV-visible detector. Separation was performed on a C18 column (Teledyne, Redisep Prep C18, 20 mm x 250 mm, 100 Å pore size, 5 μm) at room temperature using the following solvent systems: Conditions A: 0.1% trifluoroacetic acid in water (solvent A) and 0.1% trifluoroacetic acid in acetonitrile (solvent B); 0-2.5 min: isocratic elution 90% A; 2.5-35 min: gradient elution 90%-0% A; Conditions B: 0.01% trifluoroacetic acid in water (solvent A) and 0.01% trifluoroacetic acid in acetonitrile (solvent B); 0-3 min: isocratic elution 95% A; 3-25 min: gradient elution 95%-0% A; Conditions C: 0.01% trifluoroacetic acid in water (solvent A) and 0.01% trifluoroacetic acid in acetonitrile (solvent B); 0-3 min: isocratic elution 90% A; 3-25 min: gradient elution 90%-0% A; Conditions D: 0.1% trifluoroacetic acid in water (solvent A) and 0.1% trifluoroacetic acid in acetonitrile (solvent B); 0-3 min: isocratic elution 95% A; 3-25 min: gradient elution 95%-0% A. The mobile phase flow rate was maintained at 15 mL.min^{-1} and eluents were monitored at 214 and 254 nm.

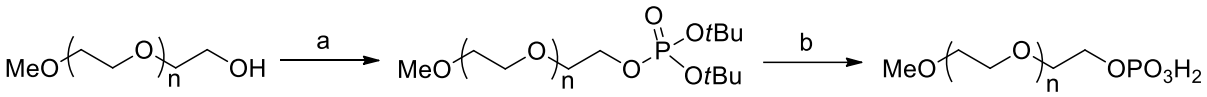
1.2. Two-step synthesis of 980-excited core/shell UCNP

The core/shell UCNP were synthesized via a co-precipitation method using minor modifications of the procedure previously reported by Francolon, N. et al.[2] To a 100 mL three-neck round-bottom flask containing a mixture of oleic acid (6 mL) and octadecene (15 mL) were successively added under stirring yttrium(III) chloride (152.3 mg, 800 μmol), ytterbium(III) chloride (55.9 mg, 200 μmol) and thulium(III) chloride (5.5 mg, 20 μmol). The solution was heated under stirring at 150 °C for 30 min to form a homogenous solution. Freshly prepared methanolic solutions of sodium hydroxide (100 mg, 2.5 mmol, 10 mL) and ammonium fluoride (148 mg, 4.00 mmol, 10 mL) were sonicated for 5 min, pooled and added to the reaction mixture cooled to 60 °C. The resulting solution was heated under stirring at 110 °C for 20 min then under vacuum for 10 min to remove volatile solvent. The flask was then filled with argon and heated under stirring at 302 °C for 1 h (heating rate 10 °C/min). After cooling down to room temperature, the reaction mixture was transferred into a PTFE centrifuge tube and the

UCNP were purified by three sedimentation-redispersion cycles involving precipitation by the addition of ethanol (25 mL), centrifugation (6666 g, 10 min, 22 °C, Sigma 3-18K centrifuge), discarding of the supernatant and redispersion in cyclohexane (20 mL). Finally, the resulting solution was centrifuged (999 g, 5 min, 22 °C, Sigma 3-18K centrifuge) and the supernatant containing core UCNP-OA (180-200 mg per batch) in cyclohexane was collected.

To a 100 mL three-neck round-bottom flask containing a mixture of oleic acid (6 mL), octadecene (15 mL) and YCl_3 (48,8 mg, 250 μmol), previously heated under stirring at 150 °C for 30 min, was added at room temperature a solution of core UCNP-OA (90 mg) in cyclohexane (15 mL). The reaction mixture was heated under stirring at 110 °C for 10 min. Freshly prepared methanolic solutions of sodium hydroxide (25 mg, 625 μmol, 10 mL) and ammonium fluoride (37 mg, 1,00 mmol, 10 mL) were sonicated for 5 min, pooled and added to the reaction mixture cooled to 40 °C. The resulting solution was heated under stirring at 110 °C for 20 min then under vacuum for 10 min to remove volatile solvent. The flask was then filled with argon and heated under stirring at 302 °C for 1.5 h (heating rate 10 °C/min). After cooling down to room temperature, the core/shell UCNP-OA were purified as described above for core UCNP-OA and stored for further use at room temperature at a final concentration of ca. 5 mg/mL in cyclohexane.

1.3. Synthesis of mono and bifunctional PEGylated ligands



S1a, n = 21
S1b, n = 44

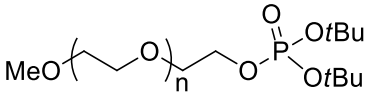
S2a, n = 21
S2b, n = 44

L1, n = 21
L2, n = 44

Reaction conditions: a) (i) $(iPr)_2NP(OtBu)_2$, tetrazole, CH_2Cl_2, RT; (ii) mCPBA, RT; b) HCl 2N, Et_2O, 0 °C then RT.

Scheme S1: Synthesis of m-PEGylated phosphoric acids **L1** and **L2**

α-Methyl-ω-(di-*tert*-butoxyphosphoryl)oxy-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S2a)

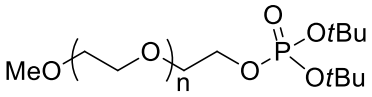


S2a, n = 21

To a solution of α-methyl-ω-hydroxy-poly(oxyethane-1,2-diyl)$_{(1000)}$ (**S1a**) (712 mg, 710 μmol) in anhydrous toluene (10 mL) was added under argon a 0.45 M solution of tetrazole in anhydrous acetonitrile (6.33 mL, 2.85 mmol). The solution was evaporated under reduced pressure and the residue was diluted in anhydrous toluene (10 mL). After evaporation under reduced pressure the residue was diluted under argon in anhydrous dichloromethane (10 mL). The solution was cooled down to -5 °C and di-*tert*-butyl *N,N*-diisopropylphosphoramidite (450 μL, 1.42 mmol) was then added. After warming to room temperature, the solution was stirred for 3 h. After cooling down to -40 °C, a solution of meta-chloroperbenzoic acid (246 mg, 1.43 mmol) in dichloromethane (10 mL) was added. The reaction mixture was warmed to room temperature and stirred for 1 h. The reaction was then quenched by the addition of 10% aqueous solution of sodium sulfite (5 mL). After decantation, the organic layer was washed successively with saturated aqueous solution of sodium hydrogen carbonate (2 x 10 mL) and brine (10 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 98/2, v/v) to afford compound **S2a** as an off-white wax (746 mg, 625 μmol). Yield: 88%. R_f = 0.23 (Al_2O_3, dichloromethane/ethanol, 98/2, v/v); IR (ATR accessory) ν 3000-2700, 1466, 1359, 1279, 1241, 1145, 1105, 1061, 1039, 992, 960 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.39 (s, 18H), 3.28 (s, 3H), 3.45 (m, 2H), 3.50-3.65 (m, ca. 82H), 3.60 (t, 2H, *J* = 5.3 Hz), 3.98 (m, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 29.7 (d, 6C, $^3J_{C-P}$ = 4.3 Hz), 58.9,

65.6 (d, 1C, $^{3}J_{C\text{-}P}$ = 6.4 Hz), 70.0 (d, 1C, $^{2}J_{C\text{-}P}$ = 8.1 Hz), 70.4-70.6 (m, ca. 41C), 71.8, 82.1 (d, 2C, $^{2}J_{C\text{-}P}$ = 7.4 Hz); ^{31}P NMR (202.6 MHz, $CDCl_3$) δ -9.7.

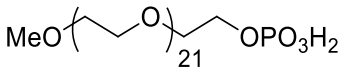
α-Methyl-ω-(di-*tert*-butoxyphosphoryl)oxy-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S2b)



S2b, n = 44

Compound **S2b** (210 mg, 95.2 μmol) was obtained as a yellow wax according to the procedure described for **S2a** starting from α-methyl-ω-hydroxy-poly(oxyethane-1,2-diyl)$_{(2000)}$ (**S1b**) (503 mg, 250 μmol); the crude was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 99/1, v/v). Yield: 38%. R_f = 0.05 (Al_2O_3, dichloromethane/methanol, 99/1, v/v); IR (ATR accessory) ν 3000-2700, 1466, 1341, 1279, 1241, 1147, 1101, 1060, 1040, 1000, 959 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.46 (s, 18H), 3.36 (s, 3H), 3.45-3.80 (m, ca. 178H), 3.90-4.15 (m, 2H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 29.8 (d, 6C, $^{3}J_{C\text{-}P}$ = 4.1 Hz), 58.9, 65.5 (d, 1C, $^{3}J_{C\text{-}P}$ = 6.5 Hz), 69.4-71.5 (m, ca. 88C), 71.8, 82.1 (d, 2C, $^{2}J_{C\text{-}P}$ = 7.3 Hz); ^{31}P NMR (202.6 MHz, $CDCl_3$) δ -9.7.

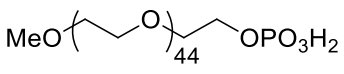
α-Methyl-ω-phosphonooxy-poly(oxyethane-1,2-diyl)$_{(1000)}$ (L1)



L1

Compound **S2a** (1.00 g, 0.871 mmol) was slowly diluted at 0 °C and under argon with a 3M anhydrous hydrogen chloride solution in diethyl ether (10 mL). After stirring for 10 min at 0 °C, the reaction mixture was warmed to room temperature and stirred for 30 min. The solution was then evaporated under reduced pressure to afford compound **L1** as an off-white wax (923 mg, 0.853 mmol). Yield: 98%. IR (ATR accessory) ν 3000-2700, 1466, 1343, 1279, 1241, 1101, 947 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.33 (s, 3H), 3.50 (m, 2H), 3.50-3.70 (m, ca. 84H), 4.12 (m, 2H), 9.02 (brs, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 59.0, 66.0 (d, 1C, $^{3}J_{C\text{-}P}$ = 4.4 Hz), 70.2-70.6 (m, ca. 42C), 72.0; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 1.1; MALDI-TOF *m/z* calculated for $C_{45}H_{93}KO_{26}P$ $[M+K]^+$: 1119.533, found: 1119.539.

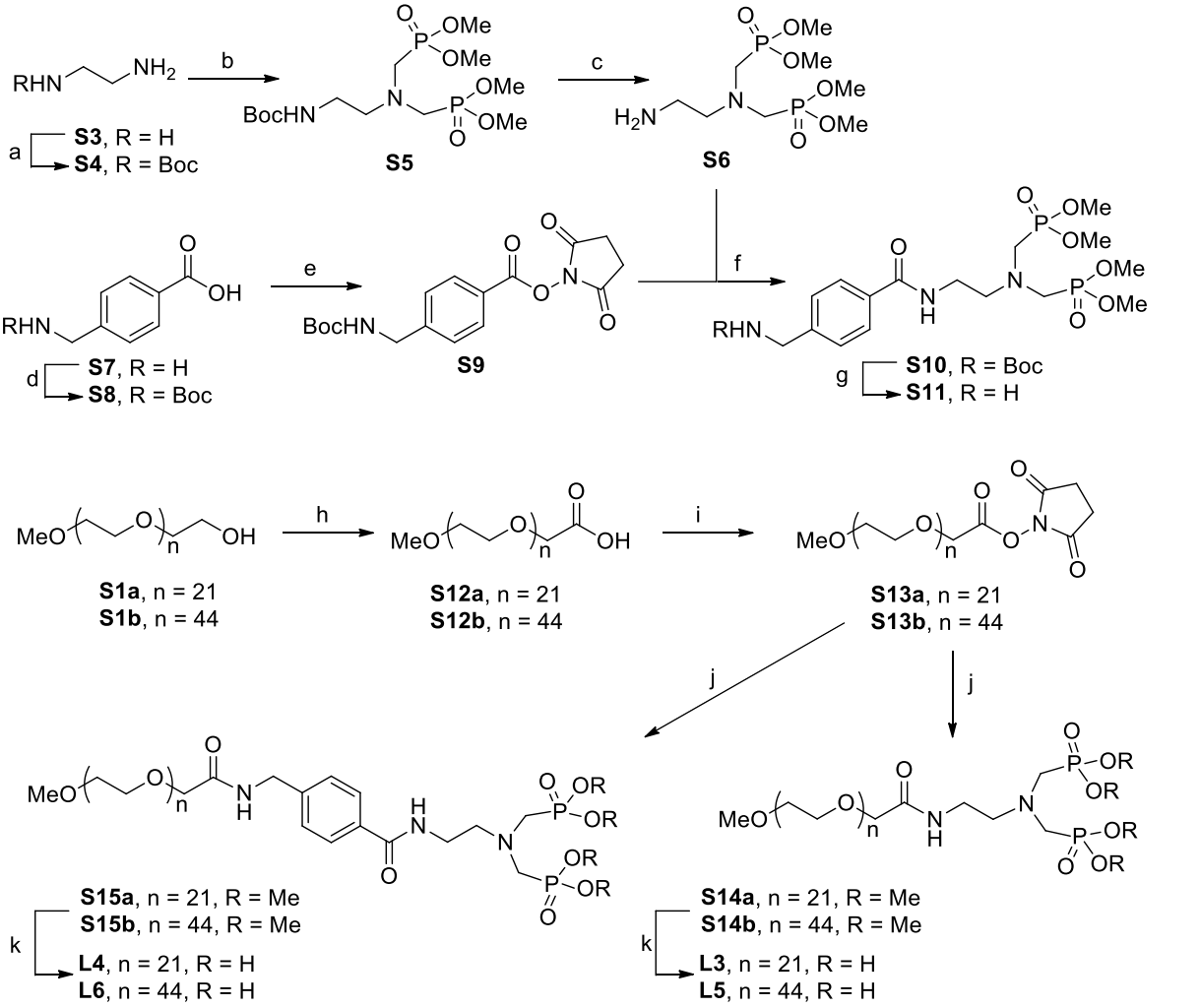
α-Methyl-ω-phosphonooxy-poly(oxyethane-1,2-diyl)$_{(2000)}$ (L2)



L2

Compound **S2b** (2.27 g, 1.03 mmol) was diluted at 0 °C and under argon with a 1.43 M anhydrous hydrogen chloride solution in diethyl ether (25 mL). After stirring for 10

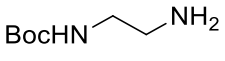
min at 0 °C, the reaction mixture was warmed to room temperature and stirred for 30 min. The solution was then evaporated under reduced pressure to afford compound **L2** as a yellow wax (2.05 g, 0.98 mmol). Yield: 95%. IR (ATR accessory) ν 3000-2700, 1467, 1342, 1279, 1241, 1147, 1109, 1061, 963 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.25 (s, 3H), 3.42 (m, 2H), 3.45-3.65 (m, ca. 176H), 4.04 (m, 2H), 9.72 (m, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 58.8, 65.6 (d, 1C, $^{3}J_{C\text{-}P}$ = 4.8 Hz), 69.9 (d, 1C, $^{2}J_{C\text{-}P}$ = 6.1 Hz), 70.0-70.6 (m, ca. 87C), 71.7; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 1.2; MALDI-TOF *m/z* calculated for $C_{91}H_{186}O_{49}P$ $[M+H]^+$: 2094.180, found: 2094.339.



Reaction conditions: a) $(Boc)_2O$, CH_2Cl_2, RT; b) (i) 35% aq CH_2O, THF, RT; (ii) HPO_3Me_2, reflux; c) TFA, CH_2Cl_2, 0 °C; d) $(Boc)_2O$, NaOH, acetone, H_2O, RT; e) NHS, THF, DCC, CH_2Cl_2, RT; f) DIPEA, DMF, RT; g) TFA, CH_2Cl_2, 0 °C; h) Jones reagent, acetone, 0 °C then RT; i) NHS, EDC.HCl, CH_2Cl_2, RT; j) DIPEA, DMF, appropriate amine (**S6** or **S11**), RT; k) (i) TMSBr, CH_2Cl_2, 0 °C then RT; (ii) MeOH, RT.

Scheme S2: Synthesis of m-PEGylated bisphosphonic acids **L3-L6**

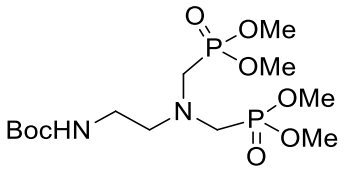
tert-Butyl 2-aminoethylcarbamate (S4)[3]



S4

To a solution of ethylenediamine (**S3**) (27.0 mL, 404 mmol) in dichloromethane (300 mL) was added dropwise over 2.5 h a solution of di-*tert*-butyl dicarbonate (8.86 g, 40.6 mmol) in dichloromethane (200 mL). After stirring at room temperature for 24 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in saturated aqueous sodium hydrogen carbonate solution (100 mL) and extracted with dichloromethane (3 x 100 mL). The combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S4** as a yellow oil (6.09 g, 38.0 mmol) which was stored under inert atmosphere at 4 °C and used in the next step without further purification. Yield: 94%. IR (ATR accessory) ν 3358, 2976, 2933, 2880, 1683, 1525, 1455, 1391, 1365, 1271, 1249, 1165 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.21 (s, 2H), 1.38 (s, 9H), 2.73 (t, 2H, *J* = 5.9 Hz), 3.11 (q, 2H, *J* = 5.9 Hz), 5.09 (brs, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.4 (3C), 41.9, 43.4, 79.1, 156.3; HRMS *m*/*z* calculated for $C_7H_{17}N_2O_2$ $[M+H]^+$: 161.1285, found: 161.1285.

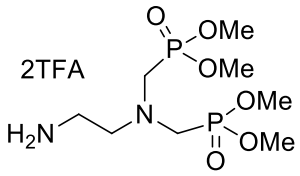
***tert*-Butyl (2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl)carbamate (S5)**



S5

To a solution of **S4** (1.46 g, 9.11 mmol) in tetrahydrofuran (75 mL) was added a 35% aqueous formaldehyde solution (1.81 mL, 22.8 mmol). The reaction mixture was stirred at room temperature for 20 min. Dimethyl phosphite (4.18 mL, 45.6 mmol) was then added and the solution was stirred under reflux for 16 h. After cooling down to room temperature, the reaction mixture was evaporated under reduced pressure and the residue was purified by column chromatography (SiO_2, ethyl acetate/ethanol, 85/15, v/v) to give compound **S5** as a colourless oil (1.33 g, 3.29 mmol). Yield: 36% (>85% purity as estimated by ^{31}P NMR). R_f = 0.28 (SiO_2, ethyl acetate/ethanol, 85/15, v/v); IR (ATR accessory) ν 3400-3150, 3150-2750, 1686, 1634, 1532, 1270, 1250, 1140, 1020, 976 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.40 (s, 9H), 2.88 (t, 2H, *J* = 5.5 Hz), 3.16 (d, 4H, $^{2}J_{H-P}$ = 8.3 Hz), 3.20 (m, 2H), 3.76 (d, 12H, $^{3}J_{H-P}$ = 10.5 Hz), 5.28 (s, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.5 (3C), 38.6, 49.5 (dd, 2C, $^{1}J_{C-P}$ = 158.0 Hz, $^{3}J_{C-P}$ = 6.3 Hz), 52.9 (m, 4C), 56.0 (t, 1C, $^{3}J_{H-P}$ = 6.6 Hz), 79.0, 156.3; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 26.9; HRMS *m*/*z* calculated for $C_{13}H_{31}N_2O_8P_2$ $[M+H]^+$: 405.1550, found: 405.1553.

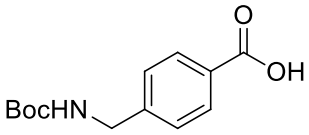
Tetramethyl (((2-aminoethyl)azanediyl)bis(methylene))bis(phosphonate) trifluoroacetic acid salt (S6)



S6

To a solution of **S5** (170 mg, 0.42 mmol) in anhydrous dichloromethane (1 mL) cooled at 0 °C was added under argon trifluoroacetic acid (500 μL). After stirring at 0 °C for 4.5 h, the reaction mixture was evaporated under reduced pressure. The residue was co-evaporated with dichloromethane (2 x 5 mL) to remove residual traces of trifluoroacetic acid to afford compound **S6** as a colourless oil (163 mg, 0.31 mmol). Yield: 73% (>85% purity as estimated by ^{31}P NMR). IR (ATR accessory) ν 3200-2750, 1772, 1672, 1457, 1146, 1028 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 3.13 (brs, 4H), 3.24 (d, 4H, $^{2}J_{H\text{-}P}$ = 7.7 Hz), 4.79 (d, 12H, $^{3}J_{H\text{-}P}$ = 10.8 Hz), 7.48 (brs, 3H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 38.2, 49.3 (dd, 2C, $^{1}J_{C\text{-}P}$ = 159.5 Hz, $^{3}J_{C\text{-}P}$ = 5.0 Hz), 53.6 (m, 4C), 53.8, 115.3 (q, 2C, $^{1}J_{C\text{-}F}$ = 286.4 Hz), 160.3 (q, 2C, $^{2}J_{C\text{-}F}$ = 40.0 Hz); ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 27.5; HRMS *m*/*z* calculated for $C_8H_{23}N_2O_6P_2$ $[M+H]^+$: 305.1026, found: 305.1026.

4-(((*tert*-Butoxycarbonyl)amino)methyl)benzoic acid (S8)[3]

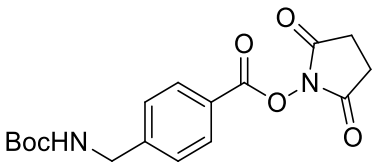


S8

To a solution of 4-(aminomethyl)benzoic acid (**S7**) (5.00 g, 33.1 mmol) in a mixture of acetone (100 mL) and deionized water (100 mL) were successively added di-*tert*-butyl dicarbonate (7.20 g, 33.0 mmol) and sodium hydroxide (1.32 g, 33.0 mmol). After stirring at room temperature for 30 h, acetone was evaporated under reduced pressure and the remaining aqueous solution was washed with diethyl ether (2 x 50 mL). Ethyl acetate (100 mL) was then added and the mixture was stirred vigorously while the pH was adjusted to 3-4 by slow addition of 10% aqueous citric acid solution. The mixture was decanted and the aqueous layer was extracted with ethyl acetate (50 mL). The combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S8** as a white solid (8.11 g, 32.3 mmol). Yield: 98%. mp 167-169 °C (Lit.[4]: 167-168 °C); IR (ATR accessory) ν 3354, 3000-2750, 2700-2300, 1682, 1506, 1429, 1321, 1292, 1242, 1165, 1121, 1051, 942 cm^{-1}; ^{1}H NMR

(200.13 MHz, $CDCl_3$) δ 1.47 (s, 9H), 4.40 (d, 2H, *J* = 5.7 Hz), 4.95 (m, 1H), 7.38 (d, 2H, *J* = 8.3 Hz), 8.07 (d, 2H, *J* = 8.3 Hz); ^{13}C NMR (50.32 MHz, DMSO-d_6) δ 28.3 (3C), 43.3, 78.1, 127.0 (2C), 129.2, 129.4 (2C), 145.4, 155.9, 167.3; HRMS *m*/*z* calculated for $C_{13}H_{17}NNaO_4$ $[M+Na]^+$: 274.1050, found: 274.1054.

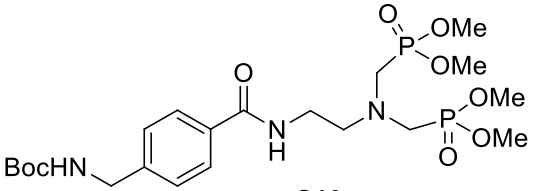
Succinimidyl 4-(((*tert*-butoxycarbonyl)amino)methyl)benzoate (S9)



S9

To a solution of **S8** (10.5 g, 41.9 mmol) in anhydrous tetrahydrofuran (200 mL) were successively added under argon *N*-hydroxysuccinimide (5.40 g, 46.9 mmol) and a solution of *N*,*N*'-dicyclohexylcarbodiimide (9.55 g, 46.3 mmol) in anhydrous dichloromethane (30 mL). After stirring at room temperature for 22 h, the formed solid was filtered off and the filtrate was evaporated under reduced pressure. The residue was purified by column chromatography (SiO_2, ethyl acetate/cyclohexane, 60/40→75/25, v/v) to give compound **S9** as an off-white solid (13.7 g, 39.3 mmol). Yield: 94%. mp 154-155 °C; R_f = 0.72 (SiO_2, ethyl acetate/cyclohexane, 70/30, v/v); IR (ATR accessory) ν 3449, 2985, 2936, 1759, 1735, 1705, 1497, 1236, 1214, 1184, 1166, 1072, 995 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 1.45 (s, 9H), 2.89 (s, 4H), 4.38 (d, 2H, *J* = 5.6 Hz), 5.06 (m, 1H), 7.40 (d, 2H, *J* = 8.3 Hz), 8.07 (d, 2H, *J* = 8.3 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 25.8 (2C), 28.4 (3C), 44.3, 80.0, 124.0, 127.6 (2C), 131.0 (2C), 146.9, 156.0, 161.7, 169.4 (2C); HRMS *m*/*z* calculated for $C_{17}H_{20}N_2NaO_6$ $[M+Na]^+$: 371.1214, found: 371.1217.

***tert*-Butyl 4-((2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl)carbamoyl) benzyl carbamate (S10)**

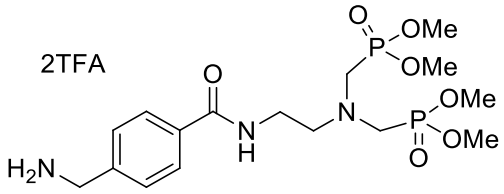


S10

To a solution of **S6** (1.26 g, 2.37 mmol) in anhydrous *N*,*N*-dimethylformamide (5 mL) were successively added under argon *N*,*N*-diisopropylethylamine (2.90 mL, 16.6 mmol) and activated ester **S9** (832 mg, 2.39 mmol). After stirring at room temperature

for 17 h, dichloromethane (30 mL) and deionized water (30 mL) were added. After decantation, the aqueous layer was extracted with dichloromethane (30 mL) and the combined organic layers were washed with brine (2 x 30 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (SiO_2, dichloromethane/methanol, 100/0→95/5, v/v) to afford compound **S10** as a colourless oil (675 mg, 1.26 mmol). Yield: 53%. R*f* = 0.45 (Al_2O_3, dichloromethane/ethanol, 90/10, v/v); IR (ATR accessory) ν 3460-3200, 3200-2700, 1648, 1536, 1505, 1454, 1392, 1366, 1218, 1165, 1024 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 1.38 (s, 9H), 2.98 (brs, 2H), 3.12 (d, 4H, $^2J_{H-P}$ = 8.8 Hz), 3.44 (m, 2H), 3.66 (d, 12H, $^3J_{H-P}$ = 10.3 Hz), 4.26 (brs, 2H), 7.26 (d, 2H, *J* = 7.9 Hz), 7.82 (d, 2H, *J* = 7.9 Hz), 8.01 (m, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.3 (3C), 38.2, 44.2, 49.5 (d, 2C, $^1J_{C-P}$ = 160.0 Hz), 52.7 (4C), 55.5, 79.5, 127.1 (2C), 127.5 (2C), 133.0, 142.5, 156.0, 167.1; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 27.1; HRMS *m/z* calculated for $C_{21}H_{38}N_3O_9P_2$ $[M+H]^+$: 538.2078, found: 538.2079.

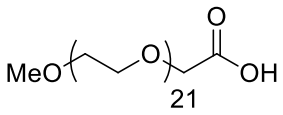
Tetramethyl (((2-(4-(aminomethyl)benzamido)ethyl)azanediyl)bis(methylene))bis (phosphonate) trifluoroacetic acid salt (S11)



S11

To a solution of **S10** (0.96 g, 1.79 mmol) in anhydrous dichloromethane (5 mL) cooled at 0 °C was added under argon trifluoroacetic acid (2.5 mL). After stirring at 0 °C for 2.5 h, the reaction mixture was evaporated under reduced pressure and the residue was co-evaporated with dichloromethane (2 x 10 mL) to remove residual traces of trifluoroacetic acid to afford compound **S11** as a colourless oil (1.18 g, 1.77 mmol). Yield: 99%. IR (ATR accessory) ν 3200-2750, 1776, 1633, 1549, 1509, 1454, 1317, 1143, 1030 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 3.08 (brs, 2H), 3.26 (d, 4H, $^2J_{H-P}$ = 9.3 Hz), 3.53 (m, 2H), 3.73 (d, 12H, $^3J_{H-P}$ = 10.8 Hz), 4.17 (brs, 2H), 7.43 (d, 2H, *J* = 7.0 Hz), 7.75 (d, 2H, *J* = 7.0 Hz), 8.04 (brs, 3H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 38.3, 43.6, 49.6 (dd, 2C, $^1J_{C-P}$ = 161.4 Hz, $^3J_{C-P}$ = 7.7 Hz), 53.6 (d, 4C, $^2J_{C-P}$ = 7.7 Hz), 55.7 (m, 1C), 115.2 (q, 2C, $^1J_{C-F}$ = 285.9 Hz), 128.0 (2C), 129.4 (2C), 133.9, 136.3, 160.1 (q, 2C, $^2J_{C-F}$ = 40.4 Hz), 168.8; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 26.5; HRMS *m/z* calculated for $C_{16}H_{30}N_3O_7P_2$ $[M+H]^+$: 438.1553, found: 438.1550.

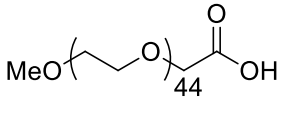
α-Methyl-ω-(carboxymethyl)oxy-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S12a)



S12a

Preparation of the 1.25 M Jones' reagent solution: to a solution of chromium trioxide (28.0 g, 0.28 mol) in deionized water (200 mL) was slowly added at 4 °C concentrated sulfuric acid (24.4 mL). The reaction mixture was stirred for 10 min and stored at 4 °C. A solution of α-methoxypoly(ethylene glycol)$_{(1000)}$ (**S1a**) (10.00 g, 10 mmol) in acetone (250 mL) was stirred for 10 min before adding dropwise over 20 min and at 0 °C a 1.25 M Jones' reagent solution (32 mL, 40 mmol). After warming to room temperature, the reaction mixture was stirred for 3 h. The progress of the reaction was monitored by TLC (SiO_2, dichloromethane/ethanol 60/40, v/v, R_f = 0.08). After completion, propan-2-ol (15 mL) was added dropwise and the solution was stirred at room temperature for 4 h. The mixture was decanted and the greenish solid was washed with acetone (2 x 40 mL). The liquid layers were pooled and concentrated under reduced pressure to remove acetone. Dichloromethane (150 mL) was added and the solution was decanted. The organic layer was successively washed with 1 N hydrochloric acid solution (3 x 40 mL) and brine (40 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was stored overnight in a vacuum desiccator to afford compound **S12a** as an off-white wax (8.77 g, 8.64 mmol) which was used in the next step without further purification. Yield: 86%. IR (ATR accessory) ν 2869, 1743, 1466, 1344, 1281, 1242, 1097, 947, 842 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 3.36 (s, 3H), 3.52 (m, 2H), 3.56-3.66 (m, ca. 80H), 3.66-3.70 (m, 2H), 4.05 (s, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 59.1, 69.0, 70.5, 70.6-70.8 (m, ca. 39C), 71.3, 72.0, 171.8; MALDI-TOF m/z calculated for $C_{45}H_{90}KO_{24}$ $[M+K]^+$: 1053.546, found: 1053.620.

α-Methyl-ω-(carboxymethyl)oxy-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S12b)

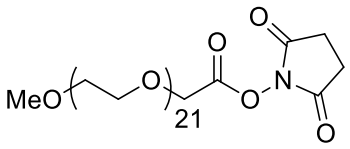


S12b

A solution of α-methoxypoly(ethylene glycol)$_{(2000)}$ (**S1b**) (20.00 g, 10 mmol) in acetone (250 mL) was stirred for 10 min before adding dropwise over 20 min and at 0 °C a 1.25 M Jones' reagent solution (32 mL, 40 mmol). After warming to room temperature, the reaction mixture was stirred for 3.5 h. The progress of the reaction was monitored by TLC (SiO_2, dichloromethane/ethanol/TFA, 50/50/1, v/v/v, R_f = 0.40). After completion, propan-2-ol (25 mL) was added dropwise and the solution was stirring at room

temperature for 15 min. The mixture was decanted and the greenish solid was washed with acetone (2 x 60 mL). The liquid layers were pooled and concentrated under reduced pressure to remove acetone. Dichloromethane (150 mL) was added and the solution was decanted. The organic layer was successively washed with 1 N hydrochloric acid solution (3 x 60 mL) and brine (60 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was stored overnight in a vacuum desiccator to afford compound **S12b** as an off-white wax (17.20 g, 8.48 mmol) which was used in the next step without further purification. Yield: 85%. IR (ATR accessory) v 2883, 1748, 1466, 1359, 1341, 1279, 1240, 1146, 1104, 1060, 947 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.36 (s, 3H), 3.53 (m, 2H), 3.55-3.70 (m, ca. 174H), 3.73 (m, 2H), 4.14 (s, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 59.1, 69.0, 70.4-70.8 (m, ca. 86C), 71.4, 72.0, 171.7; MALDI-TOF *m/z* calculated for $C_{91}H_{182}NaO_{47}$ $[M+Na]^+$: 2050.175, found: 2050.470.

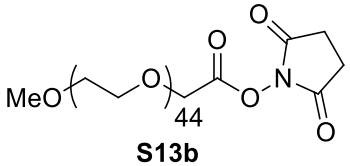
α-Methyl-ω-(2-((succinimidyl)oxy)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S13a)



S13a

Compound **S12a** (529 mg, 521 μmol) was co-evaporated under reduced pressure with anhydrous toluene (10 mL) to remove traces of water. The residue was diluted in anhydrous dichloromethane (10 mL) before addition, under argon and stirring, of *N*-hydroxysuccinimide (90 mg, 782 μmol) and *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide, hydrochloride salt (152 mg, 793 μmol) successively. After stirring at room temperature for 27 h, the solution was diluted with dichloromethane (10 mL) and extracted successively with 5% aqueous citric acid solution (2 x 20 mL), 5% aqueous sodium hydrogen carbonate (2 x 20 mL) and brine (2 x 20 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure to afford compound **S13a** as a white wax (486 mg, 437 μmol) which was used in the next step without further purification. Yield: 84%. ^{1}H NMR (500 MHz, $CDCl_3$) δ 2.84 (s, 4H), 3.36 (s, 3H), 3.55-3.75 (m, ca. 84H), 4.51 (s, 2H).

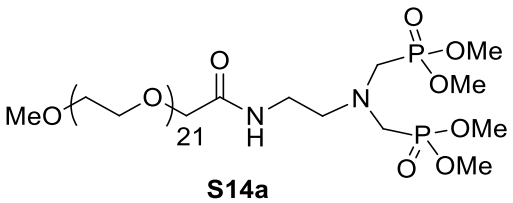
α-Methyl-ω-(2-(succinimidyl)oxy)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S13b)



S13b

Compound **S13b** (984 mg, 463 μmol) was obtained as a white wax according to the procedure described for **S13a** starting from **S12b** (983 mg, 485 μmol); reaction time at room temperature of 19 h. Yield: 95%. ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.85 (s, 4H), 3.37 (s, 3H), 3.50-3.75 (m, ca. 174H), 3.78 (m, 2H), 4.51 (s, 2H). This compound was used in the next step without further purification.

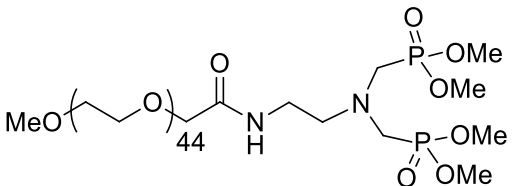
α-Methyl-ω-(2-((2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl)amino)-2-oxo ethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S14a)



S14a

To a solution of **S13a** (700 mg, 629 μmol) was added under argon a solution of **S6** (335 mg, 667 μmol) and *N*,*N*-diisopropylethylamine (548 μL, 3.15 mmol) in anhydrous *N*,*N*-dimethylformamide (1 mL). After stirring at room temperature for 22 h, dichloromethane (15 mL) and deionized water (15 mL) were added. After decantation, the aqueous layer was extracted with dichloromethane (15 mL). The combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, ethyl acetate/ethanol, 90/10, v/v) to give compound **S14a** as a colourless oil (562 mg, 432 μmol). Yield: 69%. R_f = 0.26 (Al_2O_3, ethyl acetate/ethanol, 90/10, v/v); IR (ATR accessory) ν 3324, 2870, 1668, 1537, 1449, 1348, 1242, 1087, 1042 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.93 (t, 2H, *J* = 6.0 Hz), 3.17 (d, 4H, $^{2}J_{H\text{-}P}$ = 8.4 Hz), 3.35 (s, 3H), 3.36 (q, 2H, *J* = 6.0 Hz), 3.52 (m, 2H), 3.57-3.64 (m, 80H), 3.65 (m, 2H), 3.75 (d, 12H, $^{3}J_{H\text{-}P}$ = 10.6 Hz), 3.97 (s, 2H), 7.34 (m, 1H);^{13}C NMR (50.32 MHz, $CDCl_3$) δ 36.6, 49.3 (dd, 2C, $^{1}J_{C\text{-}P}$ = 156.5 Hz, $^{3}J_{C\text{-}P}$ = 5.2 Hz), 52.5 (m, 4C), 55.6 (t, 1C, $^{3}J_{H\text{-}P}$ = 7.6 Hz), 58.9, 70.4 (m, ca. 41C), 70.6, 71.7, 169.9; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 29.4; MALDI-TOF *m*/*z* calculated for $C_{53}H_{110}N_2NaO_{29}P_2$ $[M+Na]^+$: 1323.657, found: 1323.945.

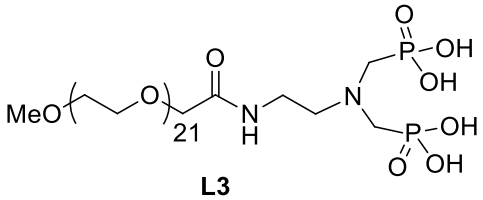
α-Methyl-ω-(2-((2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl)amino)-2-oxo ethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S14b)



S14b

To a solution of **S6** (710 mg, 1.33 mmol) in anhydrous *N,N*-dimethylformamide (2 mL) were successively added under argon *N,N*-diisopropylethylamine (1.75 mL, 10.0 mmol) and a solution of **S13b** (1.93 g, 908 μmol) in anhydrous *N,N*-dimethylformamide (5 mL). After stirring at room temperature for 96 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (120 mL) and the resulting solution was successively washed with deionized water (2 x 80 mL) and brine (2 x 80 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 95/5, v/v) to afford compound **S14b** as an orange wax (1.13 g, 489 μmol). Yield: 54%. R_f = 0.12 (Al_2O_3, dichloromethane /ethanol, 95/5, v/v); IR (ATR accessory) ν 2884, 1672, 1466, 1359, 1341, 1279, 1240, 1146, 1103, 1060, 1010, 957 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 2.84 (t, 2H, *J* = 5.3 Hz), 3.08 (d, 4H, $^2J_{H-P}$ = 8.7 Hz), 3.15-3.35 (m, 5H), 3.35-3.80 (m, ca. 176H), 3.65 (d, 12H, $^3J_{H-P}$ = 10.6 Hz), 3.89 (s, 2H), 7.27 (m, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 49.3 (dd, 2C, $^1J_{C-P}$ = 157.5 Hz, $^3J_{C-P}$ = 6.1 Hz), 52.5 (m, 4C), 55.6 (t, 1C, $^3J_{C-P}$ = 6.5 Hz), 58.9, 69.5-71.5 (m, ca. 89C), 71.8, 169.9; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 29.1; MALDI-TOF *m*/*z* calculated for $C_{99}H_{202}N_2NaO_{52}P_2$ $[M+Na]^+$: 2336.260, found: 2336.676.

α-Methyl-ω-(2-((2-(bis((phosphonato)methyl)amino)ethyl)amino)-2-oxoethoxy)-poly (oxyethane-1,2-diyl)$_{(1000)}$ (L3)

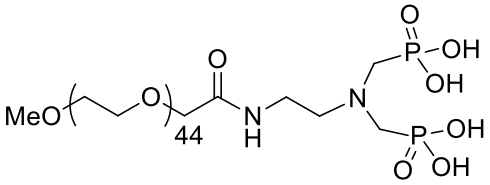


L3

To a solution of **S14a** (800 mg, 615 μmol) in anhydrous dichloromethane (35 mL) was added, at 0 °C and under argon, bromotrimethylsilane (650 μL, 4.93 mmol). The reaction mixture was warmed to room temperature and stirred for 24 h. After evaporation under reduced pressure, the residue was diluted with methanol (20 mL) and stirred for 8 h at room temperature. After evaporation under reduced pressure, the residue was triturated with anhydrous diethyl ether (3 x 10 mL) and dried under reduced pressure to afford compound **L3** as a white wax (729 mg, 585 μmol). Yield: 95%. IR (ATR accessory) ν 3323, 3000-2750, 1624, 1567, 1449, 1344, 1242, 1087 cm^1; ^1H NMR (500.13 MHz, D_2O) δ 3.39 (s, 3H), 3.63 (m, 2H), 3.65-3.80 (m, ca. 90H), 4.15 (s, 2H); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 35.8, 53.1 (d, 2C, $^1J_{C-P}$ = 136.5 Hz), 58.3, 59.4, 61.8, 69.0, 70.6-71.3 (m, ca. 37C), 71.5, 71.7, 72.4, 73.1,

175.4; ^{31}P NMR (202.6 MHz, D_2O) δ 7.0; MALDI-TOF *m/z* calculated for $C_{49}H_{102}N_2NaO_{29}P_2$ $[M+Na]^+$: 1267.594, found: 1267.819.

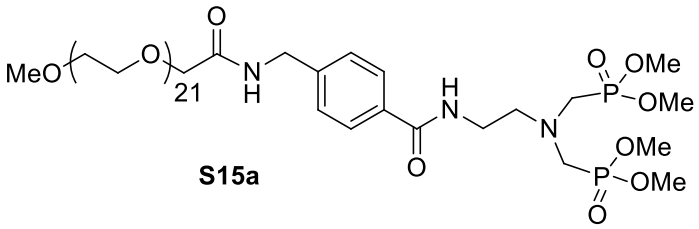
α-Methyl-ω-(2-((2-(bis((phosphono)methyl)amino)ethyl)amino)-2-oxoethoxy)-poly (oxyethane-1,2-diyl)$_{(2000)}$ (L5)



L5

Compound **L5** (312 mg, 138 μmol) was obtained as a brown solid according to the procedure described for **L3** starting from **S14b** (319 mg, 138 μmol); reaction time with bromotrimethylsilane of 18 h; reaction time with methanol of 2 h. Yield: quant. IR (ATR accessory) ν 2882, 1466, 1342, 1279, 1241, 1146, 1101, 1060, 946 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 3.28 (s, 3H), 3.40-3.80 (m, ca. 184H), 4.04 (s, 2H); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 35.8 (2C), 53.0 (d, 2C, $^1J_{C\text{-}P}$= 134.5 Hz, $^3J_{C\text{-}P}$ = 3.7 Hz), 58.0, 59.4, 61.8, 69.0, 70.5-71.3 (m, ca. 82C), 71.5, 71.8, 72.4, 73.1, 175.3; ^{31}P NMR (202.6 MHz, D_2O) δ 7.1; MALDI-TOF *m/z* calculated for $C_{95}H_{194}N_2NaO_{52}P_2$ $[M+Na]^+$: 2280.197, found: 2280.221.

α-Methyl-ω-(2-((4-((2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl) carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S15a)

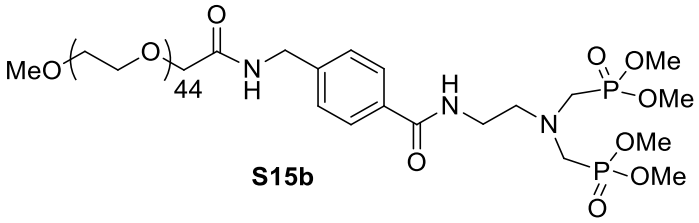


S15a

To a solution of **S13a** (780 mg, 1.17 mmol) in anhydrous *N,N*-dimethylformamide (2 mL) was added under argon a solution of **S11** (1.03 g, 926 μmol) and *N,N*-diisopropylethylamine (1.55 mL, 8.90 mmol) in anhydrous *N,N*-dimethylformamide (5 mL). After stirring at room temperature for 96 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (60 mL) and the resulting solution was successively washed with deionized water (2 x 40 mL) and brine (2 x 40 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 100/0→95/5, v/v) to afford compound **S15a** as a white wax (709 mg, 494 μmol). Yield: 53%. R_f = 0.17 (Al_2O_3, dichloromethane/ethanol, 95/5, v/v); IR (ATR accessory) ν 3323, 3000-2750, 1625,

1539, 1449, 1344, 1308, 1279, 1241, 1108, 1045 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.03 (t, 2H, *J* = 5.1 Hz), 3.16 (d, 4H, *J* = 8.3 Hz), 3.36 (s, 3H), 3.53 (m, 8H), 3.58-3.65 (m, ca. 78H), 3.72 (d, 12H, *J* = 10.6 Hz), 4.05 (s, 2H), 4.51 (d, 2H, *J* = 6.2 Hz), 7.34 (d, 2H, $^{2}J_{H\text{-}P}$ = 8.3 Hz), 7.52 (m, 1H), 7.94 (d, 2H, *J* = 8.3 Hz), 7.99 (m, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 38.5, 42.4, 49.8 (dd, 2C, $^{1}J_{C\text{-}P}$ = 160.0 Hz, $^{3}J_{C\text{-}P}$ = 7.3 Hz), 52.9 (m, 4C), 55.7 (t, 1C, $^{3}J_{C\text{-}P}$ = 6.0 Hz), 59.1, 70.3, 70.5-70.8 (m, ca. 40C), 71.2, 72.0, 127.6 (2C), 127.7 (2C), 133.4, 141.8, 167.0, 170.3; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 29.6; MALDI-TOF *m/z* calculated for $C_{61}H_{117}N_3NaO_{30}P_2$ $[M+Na]^+$: 1456.709, found: 1456.922.

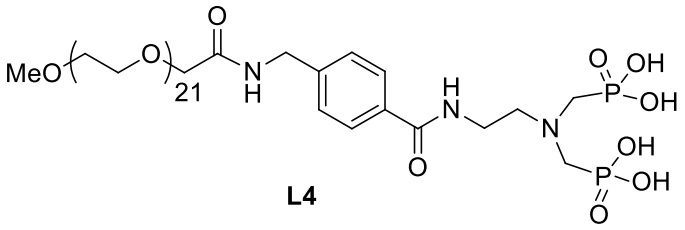
α-Methyl-ω-(2-((4-((2-(bis((dimethoxyphosphoryl)methyl)amino)ethyl) carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S15b)



S15b

To a solution of **S11** (780 mg, 1.17 mmol) in anhydrous *N*,*N*-dimethylformamide (2 mL) were successively added under argon *N*,*N*-diisopropylethylamine (1.55 mL, 8.90 mmol) and a solution of **S13b** (1.93 g, 908 μmol) in anhydrous *N*,*N*-dimethylformamide (5 mL). After stirring at room temperature for 96 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (120 mL) and the resulting solution was successively washed with deionized water (2 x 80 mL) and brine (2 x 80 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 100/0→95/5, v/v) to afford compound **S15b** as a yellowish wax (1.22 g, 498 μmol). Yield: 55%. R_f = 0.14 (Al_2O_3, dichloromethane/ethanol, 95/5, v/v); IR (ATR accessory) ν 2884, 1656, 1538, 1466, 1359, 1341, 1279, 1240, 1146, 1104, 1060, 1028, 959 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.01 (m, 2H), 3.14 (d, 4H, $^{2}J_{H\text{-}P}$ = 8.4 Hz), 3.34 (s, 3H), 3.40-3.80 (m, ca. 178H), 3.70 (d, 12H, $^{3}J_{H\text{-}P}$ = 10.6 Hz), 4.03 (s, 2H), 4.49 (d, 2H, *J* = 6.1 Hz), 7.32 (d, 2H, *J* = 8.1 Hz), 7.56 (m, 1H), 7.91 (d, 2H, *J* = 8.1 Hz), 7.97 (m, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 38.0, 41.9, 49.4 (dd, 2C, $^{1}J_{C\text{-}P}$ = 160.1 Hz, $^{3}J_{C\text{-}P}$ = 7.6 Hz), 52.5 (m, 4C), 55.3 (t, 1C, $^{3}J_{C\text{-}P}$ = 6.2 Hz), 58.7, 69.8, 69.9-70.4 (m, ca. 86C), 70.7, 71.6, 127.1 (2C), 127.2 (2C), 132.9, 141.5, 166.6, 169.9; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 27.0; MALDI-TOF *m/z* calculated for $C_{107}H_{209}N_3NaO_{53}P_2$ $[M+Na]^+$: 2469.312, found: 2469.612.

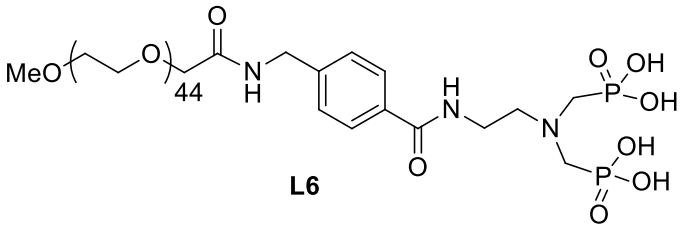
α-Methyl-ω- (2-((4-((2-(bis((phosphono)methyl)amino)ethyl)carbamoyl)benzyl) amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (L4)



L4

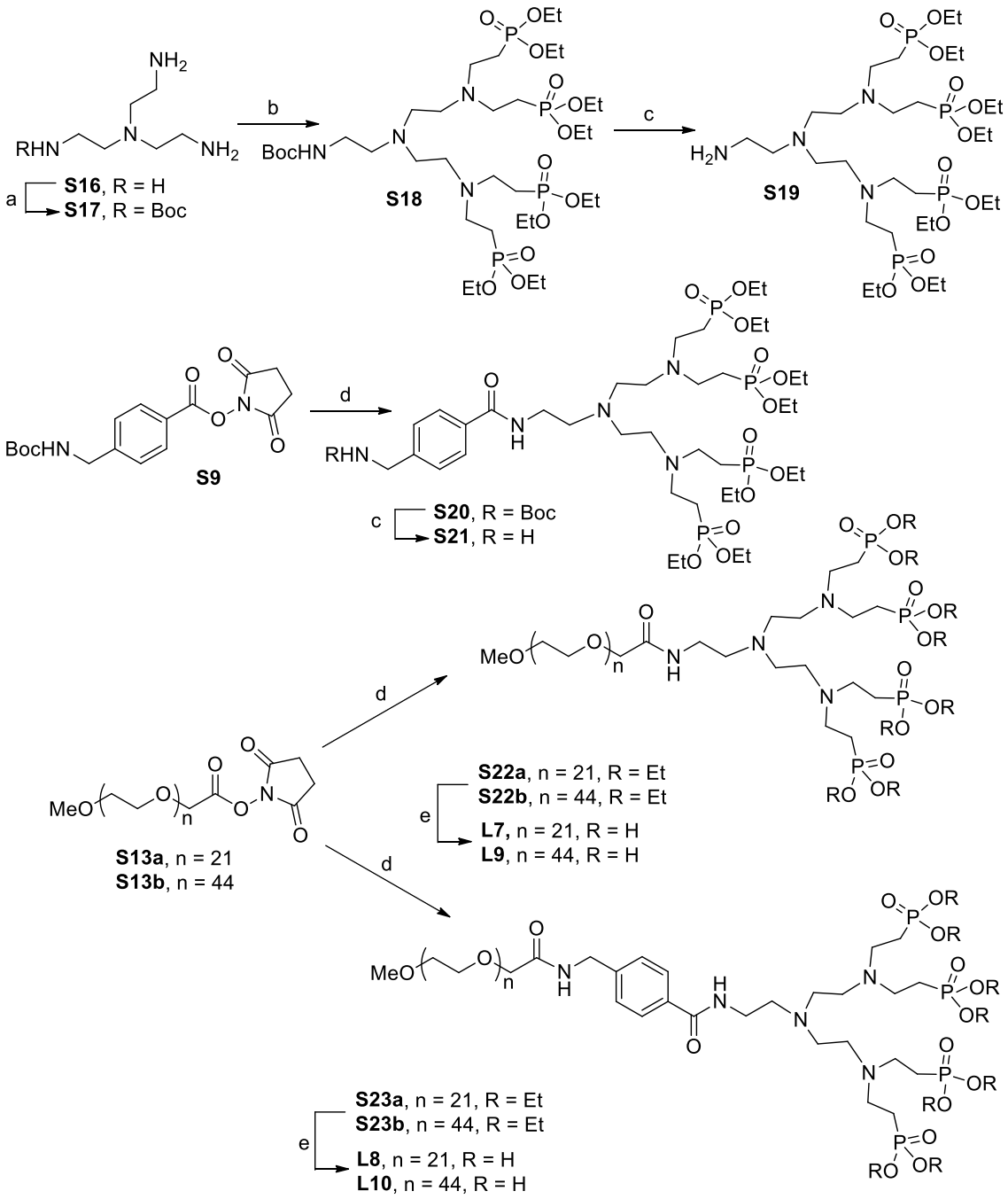
Compound **L4** (678 mg, 492 μmol) was obtained as a white wax according to the procedure described for **L3** starting from **S15a** (709 mg, 494 μmol); reaction time with bromotrimethylsilane of 18 h; reaction time with methanol of 1 h. Yield: 99%. IR (ATR accessory) ν 3323, 2877, 1627, 1549, 1448, 1401, 1343, 1279, 1242, 1201, 1106, 946 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 3.36 (s, 3H), 3.55-3.72 (m, ca. 86H), 3.76 (m, 2H), 3.83 (m, 4H), 4.14 (s, 2H), 4.51 (s, 2H), 7.43 (d, 2H, *J* = 8.2 Hz), 7.79 (d, 2H, *J* = 8.2 Hz); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 36.7, 43.4, 53.0 (dd, 2C, $^{1}J_{C\text{-}P}$= 138.1 Hz, $^{3}J_{C\text{-}P}$ = 2.5 Hz), 58.5, 59.4, 70.5-71.5 (m, ca. 41C), 71.8, 72.4, 128.8 (2C), 129.1 (2C), 132.9, 143.9, 172.1, 173.8; ^{31}P NMR (202.6 MHz, D_2O) δ 7.2; MALDI-TOF *m/z* calculated for $C_{57}H_{111}N_3O_{30}P_2$ $[M+H]^+$: 1378.664, found: 1378.830.

α-Methyl-ω- (2-((4-((2-(bis((phosphono)methyl)amino)ethyl)carbamoyl)benzyl) amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (L6)



L6

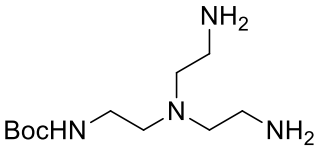
Compound **L6** (1.17 g, 489 μmol) was obtained as a brown solid according to the procedure described for **L3** starting from **S15b** (1.22 g, 498 μmol); reaction time with bromotrimethylsilane of 18 h; reaction time with methanol of 1 h. Yield: 98%. Yield: 87%. IR (ATR accessory) v 2881, 1680-1610, 1548, 1466, 1342, 1279, 1241, 1145, 1100, 1060, 959 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 3.36 (s, 3H), 3.55-3.75 (m, ca. 184H), 3.76 (m, 2H), 3.84 (m, 4H), 4.14 (s, 2H), 4.51 (s, 2H), 7.42 (d, 2H, *J* = 8.0 Hz), 7.79 (d, 2H, *J* = 8.0 Hz); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 36.6, 43.5, 52.8 (d, 2C, $^{1}J_{C\text{-}P}$ = 136.8 Hz), 58.0, 59.4, 69.3-69.9 (m, ca. 86C), 71.9, 72.4, 73.2, 128.8 (2C), 129.2 (2C), 133.1, 143.9, 172.2, 173.9; ^{31}P NMR (202.6 MHz, D_2O) δ 7.3; MALDI-TOF *m/z* calculated for $C_{103}H_{201}N_3NaO_{53}P_2$ $[M+Na]^+$: 2413.250, found: 2413.808.



Reaction conditions: a) Boc_2O, $CHCl_3$, 0 °C then RT; b) diethyl vinylphosphonate, H_2O, RT; c) TFA, CH_2Cl_2, 0 °C; d) appropriate amine (**S19** or **S21**), DIPEA, DMF, RT; e) (i) TMSBr, 0 °C then RT; (ii) MeOH, RT.

Scheme S3: Synthesis of m-PEGylated tetraphosphonic acids **L7-L10**

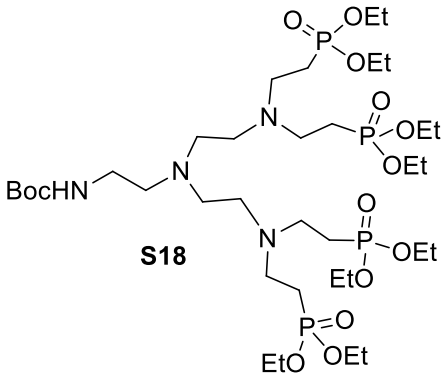
tert-Butyl (2-(bis(2-aminoethyl)amino)ethyl)carbamate (S17)[5]



S17

To a solution of *N*,*N*-bis(2-aminoethyl)ethylene-1,2-diamine (**S16**) (13.40 g, 91.6 mmol) in chloroform (600 mL) was added dropwise over 2 h at 0 °C, a solution of di-*tert*-butyl dicarbonate (2.00 g, 9.16 mmol) in chloroform (100 mL). The reaction mixture was warmed to room temperature and stirred for 20 h. Deionized water (30 mL) was then added and the solution was stirred for 15 min. After decantation, the aqueous layer was extracted with chloroform (2 x 60 mL) and the combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol/ammonia, 95/5/0.2, v/v/v) to give compound **S17** as a colourless oil (2.11 g, 8.57 mmol). Yield: 93%. R_f = 0.11 (Al_2O_3, dichloromethane/ethanol/ammonia, 95/5/0.2, v/v/v); IR (ATR accessory) ν 3500-3150, 3150-2650, 1682, 1519, 1455, 1364, 1278, 1249, 1168 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.26 (s, 9H), 2.35 (t, 4H, *J* = 6.0 Hz), 2.38 (t, 2H, *J* = 6.1 Hz), 2.57 (t, 4H, *J* = 6.0 Hz), 3.00 (m, 2H), 5.51 (brs, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 28.2 (3C), 38.6, 39.6 (2C), 53.8, 57.2 (2C), 78.6, 156.0; HRMS *m*/*z* calculated for $C_{11}H_{27}N_4O_2$ $[M+H]^+$: 247.2129, found: 247.2134.

***tert*-Butyl (2-(bis(2-(bis(2-(diethoxyphosphoryl)ethyl)amino)ethyl)amino)ethyl) carbamate (S18)**

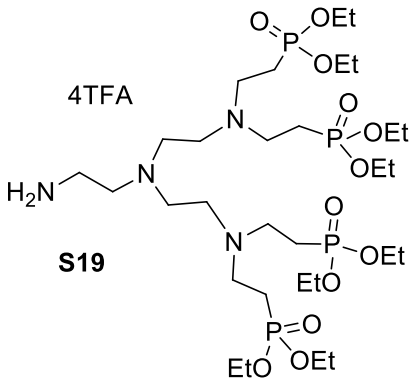


S18

To a solution of **S17** (378 mg, 1.53 mmol) in deionized water (3 mL) was added diethyl vinylphosphonate (0.91 mL, 6.11 mmol). After stirring at room temperature for 11 d, the reaction mixture was evaporated under reduced pressure and the residue was purified by column chromatography (SiO_2, dichloromethane/ethanol, 95/5→80/20, v/v then dichloromethane/ethanol/triethylamine, 80/20/0.1, v/v/v) to give compound **S18** as a yellow oil (0.87 g, 0.96 mmol). Yield: 63%. R_f = 0.10 (SiO_2, dichloromethane/ethanol/triethylamine, 80/20/0.1, v/v/v); IR (ATR accessory) ν 3600-3100, 3050-2700, 1706, 1521, 1445, 1391, 1365, 1232, 1164, 1097, 1022, 955 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.30 (t, 24H, *J* = 7.1 Hz), 1.42 (s, 9H), 1.90 (m, 8H), 2.45 (t, 4H, *J* = 6.9 Hz), 2.54 (m, 6H), 2.74 (m, 8H), 3.07 (m, 2H), 4.07 (m, 16H), 6.29 (brs, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.6 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 22.6 (d, 4C,

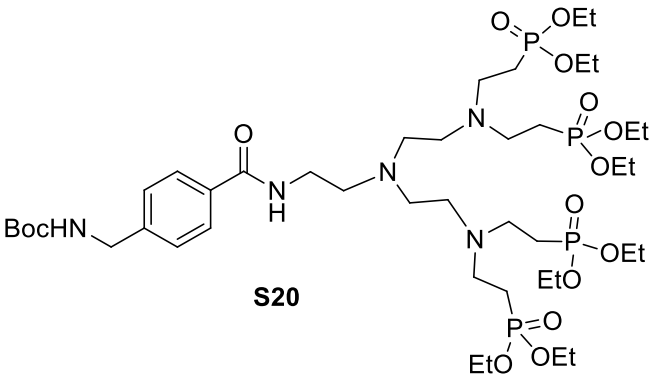
$^{1}J_{C\text{-}P}$ = 137.5 Hz), 28.6 (3C), 39.2, 46.6 (4C), 51.2 (2C), 52.4 (2C), 52.5, 61.6 (d, 8C, $^{2}J_{C\text{-}P}$ = 6.4 Hz), 78.8, 156.3; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.3; HRMS *m*/*z* calculated for $C_{35}H_{79}N_4O_{14}P_4$ $[M+H]^+$: 903.4543, found: 903.4550.

Octaethyl (((((2-aminoethyl)azanediyl)bis(ethane-2,1-diyl))bis(azanetriyl)) tetrakis(ethane-2,1-diyl))tetrakis(phosphonate) trifluoroacetic acid salt (S19)



To a solution of **S18** (1.01 g, 1.12 mmol) in anhydrous dichloromethane (5 mL) was added at 0 °C and under argon trifluoroacetic acid (2.5 mL). The solution was stirred at 0 °C for 3 h. After warming up to room temperature, the solvent was evaporated under reduced pressure to afford compound **S19** as a yellow oil (1.34 g, 1.07 mmol). Yield: 96%. IR (ATR accessory) ν 2990, 1773, 1672, 1396, 1157, 1022, 973 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.33 (t, 24H, *J* = 7.1 Hz), 2.35 (m, 8H), 2.97 (m, 2H), 3.06 (m, 4H), 3.26 (brs, 2H), 3.47 (m, 12H), 4.15 (td, 16H, $^{2}J_{H\text{-}P}$ = 15.4 Hz, *J* = 7.1 Hz), 7.53 (m, 3H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.0 (d, 8C, $^{3}J_{C\text{-}P}$ = 6.0 Hz), 20.1 (d, 4C, $^{1}J_{C\text{-}P}$ = 143.0 Hz), 37.4, 47.5 (4C), 47.8 (2C), 50.3 (2C), 51.0, 64.2 (d, 8C, $^{2}J_{C\text{-}P}$ = 6.9 Hz), 115.3 (q, $^{1}J_{C\text{-}F}$ = 287.0 Hz), 160.7 (q, $^{2}J_{C\text{-}F}$ = 40.1 Hz); ^{31}P NMR (202.76 MHz, $CDCl_3$) δ 25.0; HRMS *m*/*z* calculated for $C_{30}H_{71}N_4O_{12}P_4$ $[M+H]^+$: 803.4014, found: 803.4006.

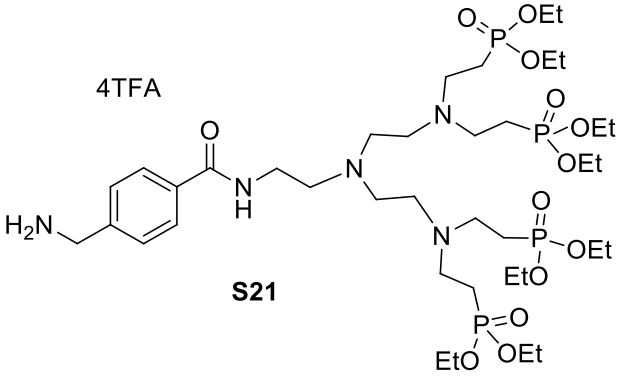
***tert*-Butyl (4-((2-(bis(2-(bis(2-(diethoxyphosphoryl)ethyl)amino)ethyl)amino) ethyl)carbamoyl)benzyl)carbamate (S20)**



To a solution of **S19** (4.22 g, 3.35 mmol) in anhydrous *N*,*N*-dimethylformamide (2 mL) were successively added, under argon, *N*,*N*-diisopropylethylamine (3.90 mL, 22.4

mmol) and a solution of activated ester **S9** (790.0 mg, 2.27 mmol) in anhydrous *N*,*N*-dimethylformamide (5 mL). The reaction mixture was stirred at room temperature for 20 h before the addition of dichloromethane (30 mL) and deionized water (30 mL). After decantation, the aqueous layer was extracted with dichloromethane (30 mL). The combined organic layers were washed with brine (2 x 30 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol/ammonia, 95/5/0.2, v/v) to give compound **S20** as a yellow oil (2.07 g, 2.00 mmol). Yield: 88%. R_f = 0.39 (dichloromethane/ethanol/ammonia (95/5/0.2, v/v/v); IR (ATR accessory) ν 3296, 2979, 1709, 1649, 1537, 1391, 1365, 1227, 1164, 1097, 1021, 954 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.29 (t, 24H, *J* = 7.1 Hz), 1.44 (s, 9H), 1.78 (m, 8H), 2.46 (t, 4H, *J* = 6.3 Hz), 2.58 (t, 4H, *J* = 6.3 Hz), 2.66 (t, 2H, *J* = 5.2 Hz), 2.72 (m, 8H), 3.44 (m, 2H), 3.95-4.15 (m, 16H), 4.33 (d, 2H, *J* = 5.5 Hz), 5.59 (brs, 1H), 7.32 (d, 2H, *J* = 8.0 Hz), 7.55 (brs, 1H), 7.75 (d, 2H, *J* = 8.0 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.6 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 23.0 (d, 4C, $^1J_{C\text{-}P}$ = 137.5 Hz), 28.5 (3C), 38.1, 44.6, 46.7 (4C), 51.1 (2C), 52.7 (2C), 52.9, 61.7 (d, 8C, $^2J_{C\text{-}P}$ = 6.5 Hz), 79.4, 127.4 (2C), 127.8 (2C), 134.2, 142.5, 156.2, 167.4; ^{31}P NMR (202.76 MHz, $CDCl_3$) δ 30.2; HRMS *m/z* calculated for $C_{43}H_{86}N_5O_{15}P_4$ $[M+H]^+$: 1036.5065, found: 1036.5059.

Octaethyl (((((2-(4-(aminomethyl)benzamido)ethyl)azanediyl)bis(ethane-2,1-diyl))bis(azanetriyl))tetrakis(ethane-2,1-diyl))tetrakis(phosphonate) trifluoroacetic acid salt (S21)

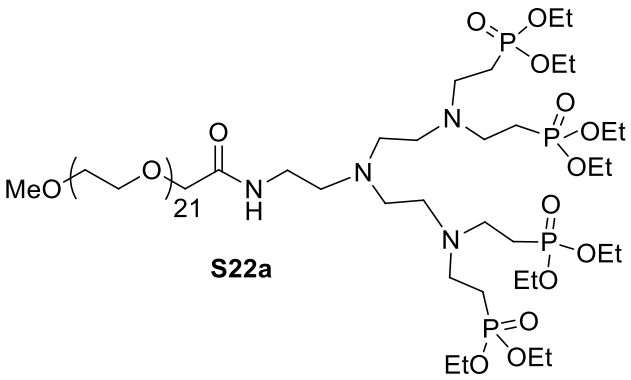


S21

To a solution of **S20** (541 mg, 522 μmol) in anhydrous dichloromethane (5 mL) was added, at 0 °C and under argon trifluoroacetic acid (2.5 mL). The solution was stirred at 0 °C for 3 h. After warming up to room temperature, the solvent was evaporated under reduced pressure to afford compound **S21** as a yellow oil (726 mg, 522 μmol). Yield: quant. IR (ATR accessory) ν 2990, 1771, 1668, 1557, 1395, 1147, 1019, 972 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.31 (t, 24H, *J* = 7.1 Hz), 2.31 (m, 8H), 3.38 (m, 2H), 3.50 (m, 8H), 3.69 (m, 4H), 3.74 (m, 6H), 4.13 (quint, 16H, *J* = 7.1 Hz), 4.20 (m, 2H), 7.43 (d, 2H, *J* = 8.2 Hz), 7.75 (d, 2H, *J* = 8.2 Hz), 7.83 (brs, 3H), 8.25 (m, 1H); ^{13}C

NMR (125.76 MHz, $CDCl_3$) δ 15.9 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 20.2 (d, 4C, $^1J_{C\text{-}P}$ =142.9 Hz), 36.1, 43.8, 47.8 (4C), 48.5 (2C), 48.9 (2C), 54.3, 64.2 (d, 8C, $^2J_{C\text{-}P}$ = 6.9 Hz), 115.3 (q, $^1J_{C\text{-}F}$ = 287.0 Hz), 128.2 (2C), 129.5 (2C), 133.0, 136.6, 160.7 (q, $^2J_{C\text{-}F}$ = 40.2 Hz), 170.1; ^{31}P NMR (202.76 MHz, $CDCl_3$) δ 24.7; HRMS *m*/*z* calculated for $C_{38}H_{78}N_5O_{13}P_4$ $[M+H]^+$: 936.4541, found: 936.4548.

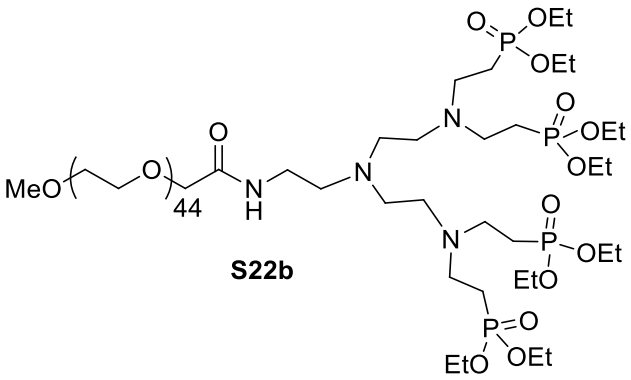
α-Methyl-ω-(2-((bis(2-(bis((diethoxyphosphoryl)ethyl)amino)ethyl)amino)ethyl) amino-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S22a)



S22a

To a solution of **S19** (1.08 g, 858 μmol) in anhydrous *N*,*N*-dimethylformamide (2 mL) were successively added under argon *N*,*N*-diisopropylethylamine (1.15 mL, 6.60 mmol) and a solution of activated ester **S13a** (540 mg, 486 μmol) in anhydrous *N*,*N*-dimethylformamide (5 mL). After stirring at room temperature for 16 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (60 mL) and the resulting solution was successively washed with deionized water (2 x 40 mL) and brine (2 x 40 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 95/5, v/v) to afford compound **S22a** as a yellow wax (639 mg, 355 μmol). Yield: 73%. R_f = 0.17 (Al_2O_3, dichloromethane/methanol, 95/5, v/v); IR (ATR accessory) ν 2950-2700, 1675, 1529, 1448, 1390, 1348, 1241, 1098, 1051, 1024, 951 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.29 (t, 24H, *J* = 7.1 Hz), 1.86 (m, 8H), 2.45 (m, 4H), 2.55 (m, 6H), 2.73 (m, 8H), 3.27 (q, 2H, *J* = 6.0 Hz), 3.34 (s, 3H), 3.51 (m, 2H), 3.55-3.70 (m, ca. 82H), 3.96 (s, 2H), 4.07 (m, 16H), 7.05 (brs, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.6 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 23.3 (d, 4C, $^1J_{C\text{-}P}$ = 137.5 Hz), 36.9, 46.9 (4C), 51.4 (2C), 53.0 (2C), 53.7, 59.1, 61.6 (d, 8C, $^2J_{C\text{-}P}$ = 6.4 Hz), 70.5, 70.6 (m, ca. 40C), 70.9, 72.0, 169.8; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.3; MALDI-TOF *m/z* calculated for $C_{75}H_{158}N_4NaO_{35}P_4$ $[M+Na]^+$: 1821.956, found: 1821.746.

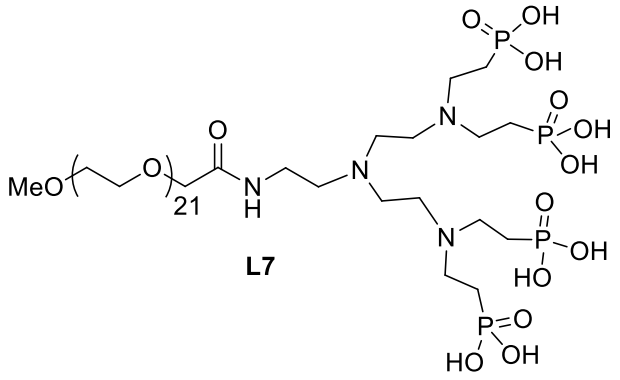
α-Methyl-ω-(2-((bis(2-(bis((diethoxyphosphoryl)ethyl)amino)ethyl)amino)ethyl) amino-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S22b)



S22b

To a solution of **S19** (960 mg, 763 μmol) in anhydrous *N*,*N*-dimethylformamide (1 mL) were successively added, under argon, *N,N*-diisopropylethylamine (1.0 mL, 5.74 mmol) and a solution activated ester **S13b** (477 mg, 224 μmol) in anhydrous *N,N*-dimethylformamide (3 mL). After stirring at room temperature for 20 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (40 mL) and successively washed with deionized water (2 x 40 mL) and brine (2 x 40 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 98/2, v/v) to afford compound **S22b** as a white wax (442 mg, 157 μmol). Yield: 70%. R_f = 0.14 (Al_2O_3, dichloromethane/methanol, 98/2, v/v); IR (ATR accessory) ν 2883, 1671, 1533, 1466, 1359, 1342, 1279, 1241, 1144, 1106, 1059, 1027, 957 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.31 (t, 24H, *J* = 7.1 Hz), 1.80-1.95 (m, 8H), 2.47 (m, 4H), 2.57 (m, 6H), 2.74 (m, 8H), 3.29 (q, 2H, *J* = 6.2 Hz), 3.36 (s, 3H), 3.53 (m, 2H), 3.55-3.74 (m, ca. 174H), 3.98 (s, 2H), 4.00-4.15 (m, 16H), 7.07 (t, 1H, *J* = 5.3 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.6 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 23.3 (d, 4C, $^1J_{C\text{-}P}$ = 137.5 Hz), 37.0, 46.9 (4C), 51.5 (2C), 53.0 (2C), 53.7, 59.1, 61.7 (d, 8C, $^2J_{C\text{-}P}$ = 6.4 Hz), 70.5-71.0 (m, ca. 88C), 72.0, 169.8; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.4; MALDI-TOF $C_{121}H_{250}N_4NaO_{58}P_4$ $[M+Na]^+$: 2834.558, found: 2834.793.

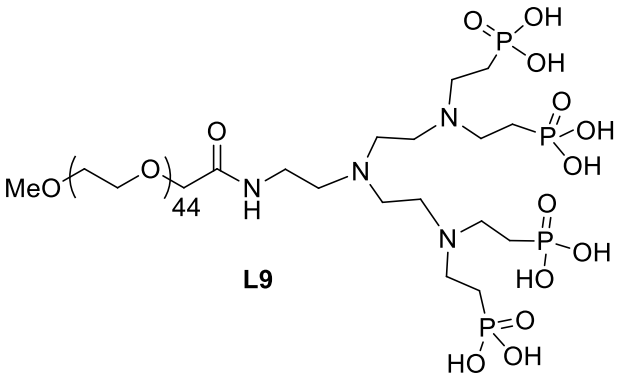
α-Methyl-ω-(2-((bis(2-(bis((phosphono)ethyl)amino)ethyl)amino)ethyl)amino-2-oxoethoxy)-poly(oxyethane-1,2-diyl)(1000) (L7)



L7

To a solution of **S22a** (862 mg, 479 μmol) in anhydrous dichloromethane (5 mL) was added, at 0 °C and under argon, bromotrimethylsilane (1.01 mL, 7.65 mmol). The reaction mixture was warmed to room temperature and stirred for 17 h. The solution was evaporated under reduced pressure and the residue was diluted in methanol (5 mL). After stirring at room temperature for 1 h 20, the solution was evaporated under reduced pressure. The residue was triturated with anhydrous diethyl ether (2 x 5 mL) and dried under reduced pressure to afford compound **L7** as a yellow solid (754 mg, 476 μmol). Yield: quant. IR (ATR accessory) ν 2866, 1668, 1456, 1346, 1245, 1093, 927 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 2.10-2.20 (m, 8H), 2.97 (t, 2H, *J* = 6.6 Hz), 3.25 (t, 4H, *J* = 6.2 Hz), 3.33 (s, 3H), 3.40-3.55 (m, 14H), 3.58 (m, 2H), 3.60-3.75 (m, ca. 82H), 4.08 (s, 2H); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 23.3 (d, 4C, $^{1}J_{C-P}$ = 133.1 Hz), 35.9, 49.7 (2C), 49.9 (4C), 50.4 (2C), 53.2, 59.4, 70.8-71.1 (m, ca. 41C), 71.7, 72.4, 174.6; ^{31}P NMR (202.6 MHz, D_2O) δ 19.0; MALDI-TOF *m/z* calculated for $C_{59}H_{127}N_4O_{35}P_4$ $[M+H]^+$: 1575.723, found: 1575.658.

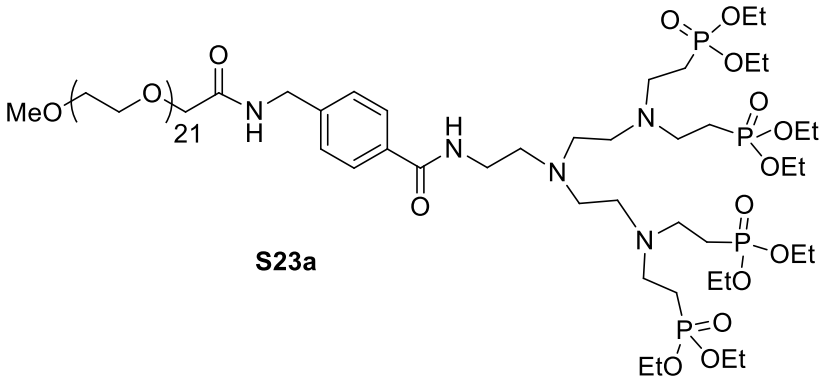
α-Methyl-ω-(2-((bis(2-(bis((phosphono)ethyl)amino)ethyl)amino)ethyl)amino-2-oxo ethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (L9)



L9

Compound **L9** (598 mg, 231 μmol) was obtained as a yellowish solid according to the procedure described for **L7** starting from **S22b** (661 mg, 235 μmol); reaction time with bromotrimethylsilane of 24 h; reaction time with methanol of 1 h. Yield: 98%. IR (ATR accessory) ν 2881, 1627, 1466, 1359, 1342, 1279, 1241, 1146, 1099, 1060, 959 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 2,14 (m, 8H), 2.95 (t, 2H, *J* = 6.7 Hz), 3.23 (t, 4H, *J* = 6.3 Hz), 3.35 (s, 3H), 3.38-3.50 (m, 14H), 3.59 (m, 2H), 3.63-3.80 (m, ca. 174H), 4.09 (s, 2H); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 23.4 (d, 4C, $^{1}J_{C-P}$ = 132.3 Hz), 36.0, 49.7 (2C), 50.0 (4C), 50.8 (2C), 52.7, 59.4, 70.7-71.2 (m, ca. 87C), 71.7, 72.4, 174.5; ^{31}P NMR (202.6 MHz, D_2O) δ 19.3; MALDI-TOF *m/z* calculated for $C_{105}H_{219}N_4O_{58}P_4$ $[M+H]^+$: 2588.326, found: 2588.084.

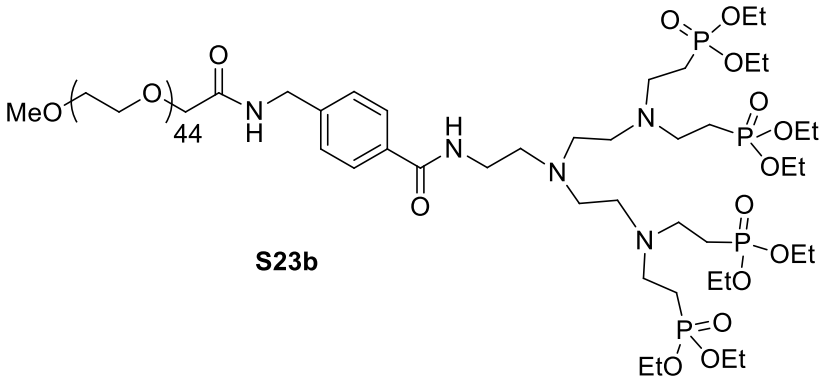
α-Methyl-ω-((2-((4-((bis(2-(bis((diethoxyphosphoryl)ethyl)amino)ethyl)amino) ethyl)carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (S23a)



S23a

To a solution of **S21** (1.36 g, 905 μmol) in anhydrous *N*,*N*-dimethylformamide (2 mL) were successively added under argon *N*,*N*-diisopropylethylamine (1.50 mL, 8.60 mmol) and a solution of activated ester **S13a** (832 mg, 748 μmol) in anhydrous *N*,*N*-dimethylformamide (5 mL). The reaction mixture was stirred at room temperature for 16 h before the addition of dichloromethane (20 mL) and deionized water (20 mL). After decantation, the aqueous layer was extracted with dichloromethane (20 mL) and the combined organic layers were washed with brine (2 x 20 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 95/5, v/v) to afford compound **S23a** as a yellow wax (656 mg, 339 μmol). Yield: 45%. R_f = 0.23 (Al_2O_3, dichloromethane/ethanol/ammonia, 95/5, v/v); IR (ATR accessory) ν 2876, 1656, 1537, 1466, 1342, 1279, 1240, 1144, 1104, 1055, 1024, 954 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.29 (t, 24H, *J* = 7.1 Hz), 1.83 (m, 8H), 2.48 (t, 4H, *J* = 7.3 Hz), 2.59 (t, 4H, *J* = 6.3 Hz), 2.68 (t, 2H, *J* = 6.1 Hz), 2.74 (m, 8H), 3.35 (s, 3H), 3.44 (m, 2H), 3.50-3.70 (m, ca. 84H), 4.05 (m, 18H), 4.50 (d, 2H, *J* = 6.1 Hz), 7.32 (d, 2H, *J* = 8.4 Hz), 7.47 (t, 1H, *J* = 5.0 Hz), 7.52 (t, 1H, *J* = 6.0 Hz), 7.76 (d, 2H, *J* = 8.4 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 16.2 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 22.6 (d, 4C, $^1J_{C\text{-}P}$ = 137.6), 37.6, 42.1, 46.3 (4C), 50.7 (2C), 52.3 (2C), 52.8, 58.7, 61.4 (d, 8C, $^2J_{C\text{-}P}$ = 6.5 Hz), 69.8-70.6 (m, ca. 40C), 70.8, 71.6, 72.4, 127.2 (2C), 127.3 (2C), 133.6, 141.6, 166.9, 169.9; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.3; MALDI-TOF *m/z* calculated for $C_{83}H_{166}N_5O_{36}P_4$ $[M+H]^+$: 1933.026, found: 1932.891.

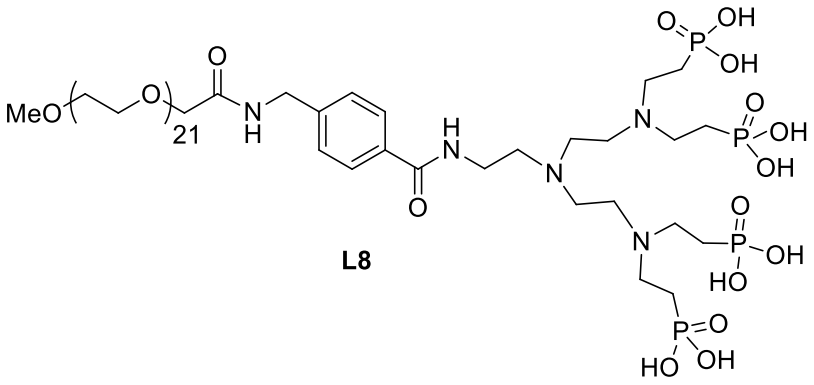
α-Methyl-ω-((2-((4-((bis(2-(bis((diethoxyphosphoryl)ethyl)amino)ethyl)amino) ethyl)carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S23b)



S23b

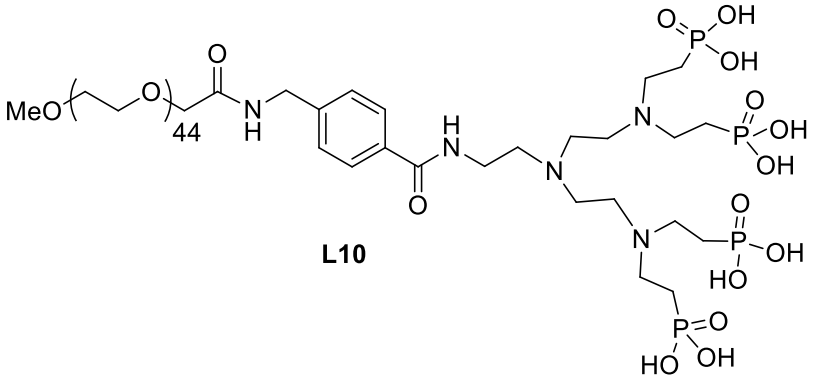
To a solution of **S21** (1.33 g, 880 μmol) in anhydrous *N*,*N*-dimethylformamide (1 mL) were successively added under argon *N*,*N*-diisopropylethylamine (1.15 mL, 6.60 mmol) and a solution of activated ester **S13b** (477 mg, 224 μmol) in anhydrous *N*,*N*-dimethylformamide (2 mL). After stirring at room temperature for 19 h, the reaction mixture was evaporated under reduced pressure and the residue was diluted with dichloromethane (40 mL). The resulting solution was successively washed with deionized water (2 x 40 mL) and brine (2 x 40 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 98/2, v/v) to afford compound **S23b** as an off-white wax (410 mg, 139 μmol). Yield: 62%. R*f* = 0.25 (Al_2O_3, dichloromethane/ethanol, 98/2, v/v); IR (ATR accessory) ν 2885, 1656, 1538, 1466, 1359, 1341, 1279, 1240, 1147, 1104, 1059, 1026, 957 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.29 (t, 24H, *J* = 7.1 Hz), 1.78-1.88 (m, 8H), 2.48 (t, 4H, *J* = 6.6 Hz), 2.59 (t, 4H, *J* = 6.6 Hz), 2.68 (t, 2H, *J* = 5.9 Hz), 2.74 (q, 8H, *J* = 8.1 Hz), 3.36 (s, 3H), 3.46 (m, 2H), 3.50-3.72 (m, ca. 176H), 3.99-4.12 (m, 18H), 4.50 (d, 2H, *J* = 6.1 Hz), 7.33 (d, 2H, *J* = 8.2 Hz), 7.50 (t, 1H, *J* = 5.0 Hz), 7.57 (t, 1H *J* = 6.0 Hz), 7.77 (d, 2H, *J* = 8.2 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.6 (d, 8C, $^3J_{C\text{-}P}$ = 6.0 Hz), 23.1 (d, 4C, $^1J_{C\text{-}P}$ = 137.6 Hz), 38.0, 42.4, 46.7 (4C), 51.1 (2C), 52.7 (2C), 53.2, 59.1, 61.7 (d, 8C, $^2J_{C\text{-}P}$ = 6.5 Hz), 70.3, 70.50-70.75 (m, ca. 86C), 71.2, 72.0, 127.5 (2C), 127.7 (2C), 134.0, 141.9, 167.2, 170.2; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.2; MALDI-TOF *m*/*z* calculated for $C_{129}H_{257}N_5NaO_{59}P_4$ $[M+Na]^+$: 2967.611, found: 2967.464.

α-Methyl-ω-((2-((4-((bis(2-(bis((phosphono)ethyl)amino)ethyl)amino)ethyl) carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(1000)}$ (L8)



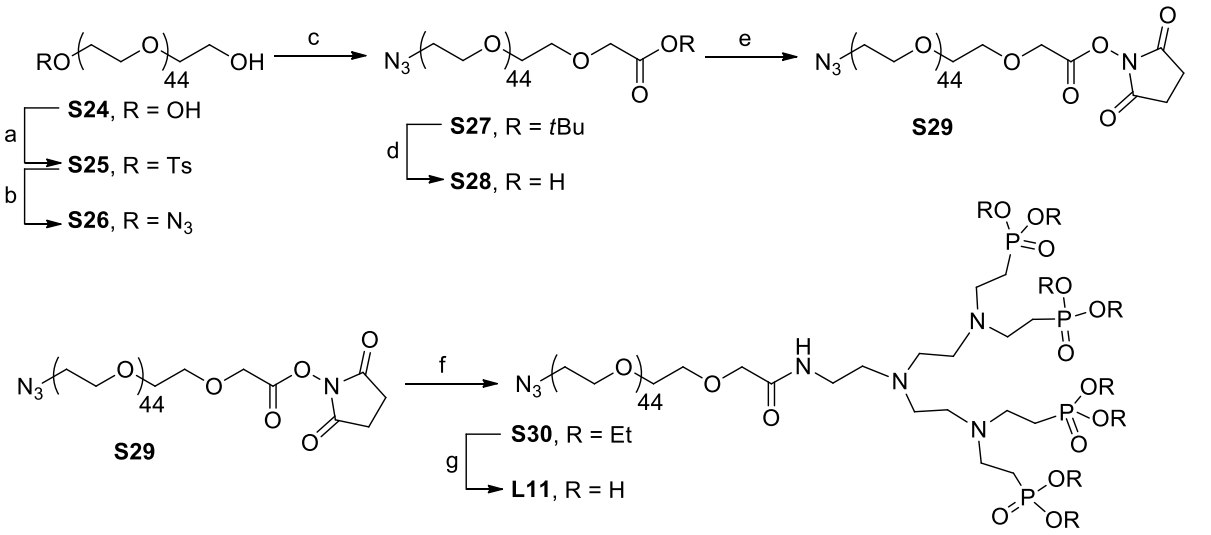
Compound **L8** (410 mg, 240 μmol) was obtained as an off-white solid according to the procedure described for **L7** starting from **S23a** (473 mg, 245 μmol); reaction time with bromotrimethylsilane of 19 h; reaction time with methanol of 1 h; Yield: 98%. IR (ATR accessory) ν 3050-2750, 1674, 1530, 1466, 1343, 1240, 1101, 1024, 953 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 2.05-2.15 (m, 8H), 3.02 (t, 2H, *J* = 6.4 Hz), 3.23 (t, 4H, *J* = 6.2 Hz), 3.35 (s, 3H), 3.35-3.50 (m, 12H), 3.55-3.73 (m, ca. 84H), 3.75 (m, 2H), 4.14 (s, 2H), 4.50 (s, 2H), 7.43 (d, 2H, *J* = 8.3 Hz), 7.78 (d, 2H, *J* = 8.3 Hz); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 23.3 (d, 4C, $^{1}J_{C\text{-}P}$ = 133.3 Hz), 37.0, 43.4, 49.8 (4C), 49.9 (2C), 50.4 (2C), 53.7, 59.4, 70.7-71.1 (m, ca. 41C), 71.8, 72.3, 128.9 (2C), 129.0 (2C), 133.0, 143.9, 171.4, 173.8; ^{31}P NMR (202.6 MHz, D_2O) δ 19.2; MALDI-TOF *m/z* calculated for $C_{67}H_{134}N_5O_{36}P_4$ $[M+H]^+$: 1708.776, found: 1708.514.

α-Methyl-ω-((2-((4-((bis(2-(bis((phosphono)ethyl)amino)ethyl)amino)ethyl) carbamoyl)benzyl)amino)-2-oxoethoxy)-poly(oxyethane-1,2-diyl)$_{(2000)}$ (L10)



Compound **L10** (404 mg, 146 μmol) was obtained as an off-white solid according to the procedure described for **L7** starting from **S23b** (436 mg, 148 μmol); reaction time

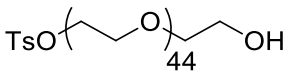
with bromotrimethylsilane of 24 h; reaction time with methanol of 1 h; Yield: 99%. IR (ATR accessory) ν 3000-2750, 1644, 1545, 1466, 1359, 1341, 1279, 1241, 1146, 1101, 1060, 946 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O) δ 2.10-2.25 (m, 8H), 3.09 (t, 2H, *J* = 6.2 Hz), 3.30 (t, 4H, *J* = 5.8 Hz), 3.43 (s, 3H), 3.45-3.57 (m, 12H), 3.60-3.90 (m, ca. 178H), 4.22 (s, 2H), 4.59 (s, 2H), 7.51 (d, 2H, *J* = 8.3 Hz), 7.86 (d, 2H, *J* = 8.3 Hz); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 23.3 (d, 4C, $^{1}J_{C\text{-}P}$ = 133.0 Hz), 37.1, 43.4, 49.8 (4C), 49.9 (2C), 50.7 (2C), 53.5, 59.4, 70.5-71.5 (m, ca. 87C), 71.8, 72.3, 128.9 (2C), 129.0 (2C), 133.1, 143.9, 171.5, 173.9; ^{31}P NMR (202.6 MHz, D_2O) δ 19.2; MALDI-TOF *m/z* calculated for $C_{113}H_{225}N_5NaO_{59}P_4$ $[M+Na]^+$: 2743.361, found: 2743.337.



Reaction conditions: a) TsCl, TEA, CH_2Cl_2, 0 °C; b) NaN_3, DMF, 90 °C; c) (i) NaH, THF, 0 °C; (ii) $BrCH_2CO_2tBu$, 50 °C; d) aq. HCl 2N, reflux; e) NHS, DCC, CH_2Cl_2, 0 °C then RT; f) **S19**, DIPEA, DMF, RT; g) (i) TMSBr, 0 °C then RT; (ii) MeOH, RT.

Scheme S4: Synthesis of N_3-PEGylated tetraphosphonic acid **L11**

α-Tosyl-ω-hydroxy-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S25)

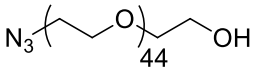


S25

To a solution of *p*-toluene sulfonyl chloride (5.73 g, 30.0 mmol) in dichloromethane (100 mL) were successively added, at 0 °C, $PEG_{(2000)}$ (**S24**) (20.0 g, 10.0 mmol) and a solution of triethylamine (4.20 mL, 30.0 mmol) in dichloromethane (100 mL). The solution was stirred at 0 °C for 2 h. After warming to room temperature, the reaction mixture was evaporated under reduced pressure. The residue was dissolved in dichloromethane (500 mL) and successively washed with deionized water (2 x 250 mL) and brine (2 x 250 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 99/1, v/v) to afford compound

S25 as a white wax (6.72 g, 3.12 mmol). Yield: 31%. R_f = 0.06 (Al_2O_3, dichloromethane/methanol, 99/1, v/v); IR (ATR accessory) ν 2883, 1466, 1359, 1341, 1279, 1240, 1147, 1101, 1060, 957 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 2.25 (s, 3H), 3.15-3.70 (m, ca. 178H), 3.94 (m, 2H), 7.15 (d, 2H, *J* = 8.2 Hz), 7.57 (d, 2H, *J* = 8.2 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 21.8, 61.8, 68.8, 69.4, 70.4, 70.5-70.8 (m, ca. 84C), 70.9, 72.7, 128.1 (2C), 129.9 (2C), 133.1, 144.9; MALDI-TOF *m*/*z* calculated for $C_{97}H_{188}KO_{48}S$ $[M+K]^+$: 2192.2, found: 2193.5.

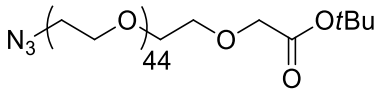
α-Hydro-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S26)



S26

To a solution of **S25** (1.53 g, 710 μmol) in anhydrous *N*,*N*-dimethylformamide (17 mL) was added under argon sodium azide (231 mg, 3.55 mmol). The suspension was heated at 90 °C for 18 h. After cooling down to room temperature, the reaction mixture was filtered and the filtrate was evaporated under reduced pressure. The residue was diluted in dichloromethane (100 mL) and washed successively with deionized water (2 x 50 mL) and brine (2 x 50 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was diluted in dichloromethane (1.5 mL) and slowly poured into cold diethyl ether (100 mL). The formed precipitate was filtered off and dried under reduced pressure to afford compound **S26** as a yellow wax (1.22 g, 602 μmol). Yield: 85%. IR (ATR accessory) ν 2883, 2098, 1466, 1359, 1341, 1279, 1241, 1147, 1098, 1060, 959 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 3.38 (m, 2H), 3.50-3.90 (m, ca. 178H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 50.3, 61.2, 69.5-72.0 (m, ca. 87C), 72.2.

α-(*tert*-Butyloxycarbonyl)methyl-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S27)

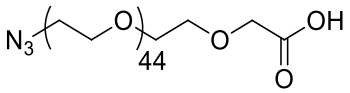


S27

A solution of **S26** (505 mg, 250 μmol) in anhydrous toluene (15 mL) was evaporated under reduced pressure and the residue was diluted in anhydrous tetrahydrofuran (15 mL). After cooling down to 0 °C, sodium hydride (60% in mineral oil, 21.7 mg, 543 μmol) was added under argon and the reaction mixture was stirred for 3 h while maintaining the temperature at 0 °C. *tert*-Butyl bromoacetate (80 µL, 545 µmol) was then added and the solution was warmed to 50 °C and stirred for 24 h. After cooling down to room temperature, the reaction mixture was filtered and the residue was washed with tetrahydrofuran (3 x 10 mL). The filtrate was evaporated under reduced

pressure, diluted with dichloromethane (20 mL) and washed with deionized water (10 mL). The aqueous layer was extracted with dichloromethane (2 x 20 mL) and the combined organic layers were washed with brine (30 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 99/1, v/v) to afford compound **S27** as a yellow wax (395 mg, 185 μmol). Yield: 74%. R_f = 0.01 (Al_2O_3, dichloromethane/ethanol, 99/1, v/v); IR (ATR accessory) ν 2884, 2101, 1752, 1466, 1340, 1279, 1240, 1147, 1105, 1060, 957 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.47 (s, 9H), 3.39 (t, 2H, *J* = 5.1 Hz), 3.55-3.75 (m, ca. 178H), 4.02 (s, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 28.1 (3C), 50.7, 69.0, 70.0, 70.2-71.0 (m, ca. 88C), 81.5, 169.7; MALDI-TOF *m/z* calculated for $C_{96}H_{191}N_3NaO_{47}$ $[M+Na]^+$: 2161.255, found: 2161.552.

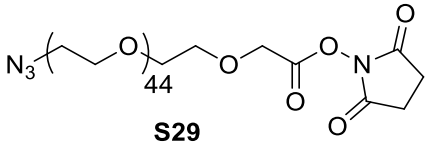
α-Carboxymethyl-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S28)



S28

A solution of **S27** (348 mg, 163 μmol) in 2N hydrochloric acid solution (10 mL) was stirred at reflux for 1 h 15. After cooling down to room temperature, the reaction mixture was diluted with deionized water (10 mL) and extracted with dichloromethane (5 x 20 mL). The combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure to afford compound **S28** as a yellow wax (340 mg, 163 μmol) which was used in the next step without further purification. Yield: quant. R_f = 0.44 (SiO_2, chloroform/methanol, 85/15, v/v); IR (ATR accessory) ν 2883, 2101, 1743, 1466, 1359, 1340, 1279, 1240, 1147, 1104, 1060, 947 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 3.36 (t, 2H, *J* = 5.1 Hz), 3.55-3.70 (m, ca. 176H), 3.73 (m, 2H), 4.13 (s, 2H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 50.8, 69.0, 70.1, 70.3-71.0 (m, ca. 87C), 71.3, 171.6; MALDI-TOF *m/z* calculated for $C_{92}H_{183}N_3NaO_{47}$ $[M+Na]^+$: 2105.192, found: 2105.590.

α-((Succinimidyloxy)carbonyl)methyl-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S29)

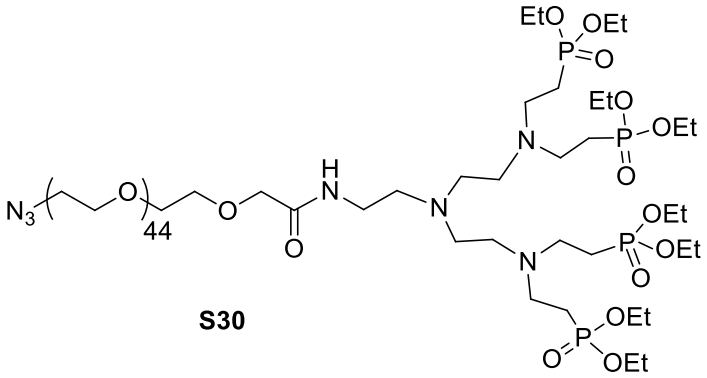


S29

A solution of **S28** (528 mg, 253 μmol) was co-evaporated under reduced pressure with anhydrous toluene (10 mL) to remove traces of water. The residue was diluted in anhydrous dichloromethane (10 mL) and *N*-hydroxysuccinimide (44 mg, 382 μmol) and *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide, hydrochloride salt (732 mg, 381

μmol) were successively added under argon. After stirring at room temperature for 18 h, the solution was diluted with dichloromethane (40 mL) and washed successively with 5% aqueous citric acid solution (2 x 50 mL), 5% aqueous sodium hydrogen carbonate (2 x 50 mL) and brine (2 x 50 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure to afford compound **S29** as a yellow wax (526 mg, 241 μmol) which was used in the next step without further purification. Yield: 95%. ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.82 (s, 4H), 3.34 (t, 2H, *J* = 5.1 Hz), 3.50-3.70 (m, ca. 176H), 3.75 (m, 2H), 4.48 (s, 2H).

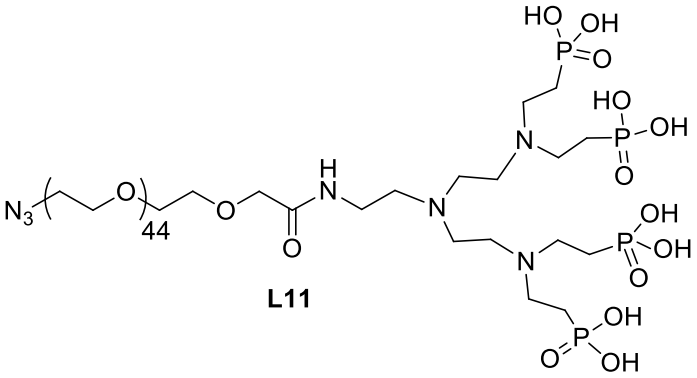
α-(2-((Bis(2-(bis((diethoxyphosphoryl)ethyl)amino)ethyl)amino)ethyl)amino-2-oxoethyl)-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (S30)



S30

To a solution of **S19** (527 mg, 419 μmol) in anhydrous *N*,*N*-dimethylformamide (0.8 mL) were successively added under argon *N*,*N*-diisopropylethylamine (550 μL, 3.16 mmol) and a solution of activated ester **S29** (526 mg, 241 μmol) in anhydrous *N*,*N*-dimethylformamide (2 mL). After stirring at room temperature for 16 h, the reaction mixture was evaporated under reduced pressure. The residue was diluted with dichloromethane (50 mL) and washed successively with deionized water (2 x 30 mL) and with brine (2 x 30 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (Al_2O_3, dichloromethane/methanol, 98/2, v/v) to afford compound **S30** as an off-white wax (517 mg, 180 μmol). Yield: 75%. R*f* = 0.20 (Al_2O_3, dichloromethane/methanol, 98/2, v/v); IR (ATR accessory) ν 2884, 2100, 1673, 1466, 1359, 1342, 1279, 1240, 1147, 1105, 1059, 1026, 957 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.27 (t, 24H, *J* = 7.1 Hz), 1.79-1.89 (m, 8H), 2.43 (m, 4H), 2.53 (m, 8H), 2.71 (m, 8H), 3.26 (q, 2H, *J* = 6.1 Hz), 3.33 (t, 2H, *J* = 5.1 Hz), 3.50-3.70 (m, ca. 176H), 3.94 (s, 2H), 3.96-4.10 (m, 16H), 7.06 (t, 1H, *J* = 5.3 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 16.5 (d, 8C, $^{3}J_{C\text{-}P}$ = 6.0 Hz), 23.2 (d, 4C, $^{1}J_{C\text{-}P}$ = 137.5 Hz), 36.8, 46.8 (4C), 50.7 (2C), 51.3 (2C), 52.9 (2C), 53.6, 61.6 (d, 8C, $^{2}J_{C\text{-}P}$ = 6.5 Hz), 70.0, 70.4-70.9 (m, ca. 88C), 169.7; ^{31}P NMR (202.6 MHz, $CDCl_3$) δ 30.3; MALDI-TOF *m/z* calculated for $C_{122}H_{252}N_7O_{58}P_4$ $[M+H]^+$: 2867.594, found: 2867.716.

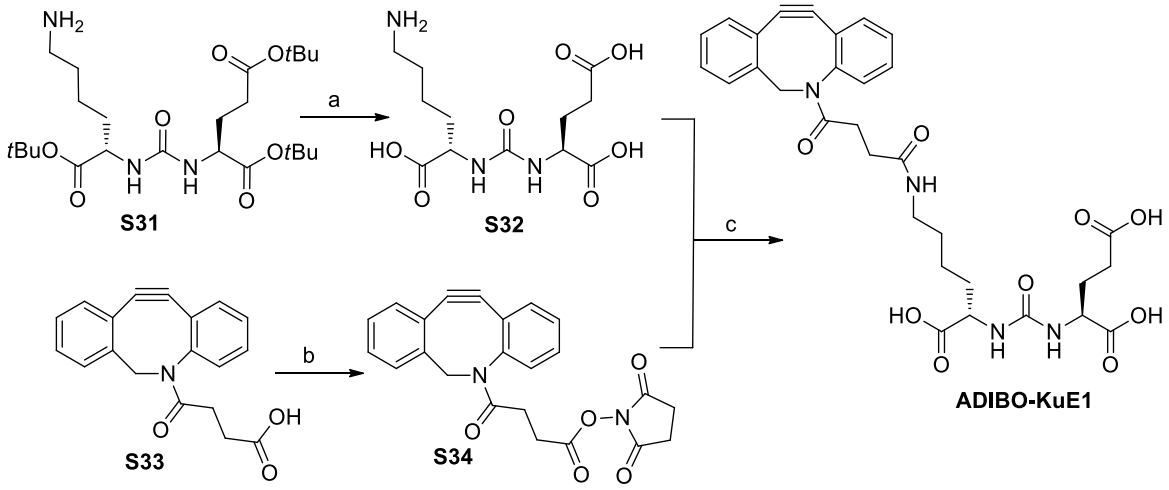
α-(2-((Bis(2-(bis((phosphono)ethyl)amino)ethyl)amino)ethyl)amino-2-oxo ethoxy)-ω-azido-poly(oxyethane-1,2-diyl)$_{(2000)}$ (L11)



L11

Compound **L11** (147 mg, 56 μmol) was obtained as an off-white solid according to the procedure described for **L7** starting from **S30** (161 mg, 56 μmol); reaction time with bromotrimethylsilane of 16 h; reaction time with methanol of 1 h. Yield: 99%. IR (ATR accessory) ν 2882, 2101, 1646, 1466, 1359, 1342, 1279, 1241, 1146, 1102, 1060, 945 cm^{-1}; ^{1}H NMR (500.13 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 2.10-2.28 (m, 8H), 3.00 (t, 2H, J = 6.1 Hz), 3.29 (m, 4H), 3.40-3.55 (m, 16H), 3.55-3.75 (m, ca. 178H), 4.08 (s, 2H); ^{13}C NMR (125.76 MHz, D_2O/acetone-d_6, 95/5, v/v) δ 23.3 (d, 4C, $^{1}J_{C\text{-}P}$ = 132.8 Hz), 36.0, 49.7 (2C), 49.9 (4C), 50.8 (2C), 51.5 (4C), 52.8, 70.6, 70.8-71.1 (m, ca. 85C), 71.7, 174.5; ^{31}P NMR (202.6 MHz, D_2O) δ 20.0; MALDI-TOF *m/z* calculated for $C_{106}H_{220}N_7O_{58}P_4$ $[M+H]^+$: 2643.343, found: 2644.041.

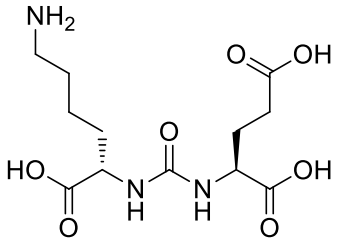
1.4. Synthesis of PSMA targeting ligands



Reaction conditions: a) TFA, CH_2Cl_2, RT; b) NHS, EDC.HCl, CH_2Cl_2, RT; c) DIPEA, DMF, RT.

Scheme S5: Synthesis of **ADIBO-KuE1** conjugate

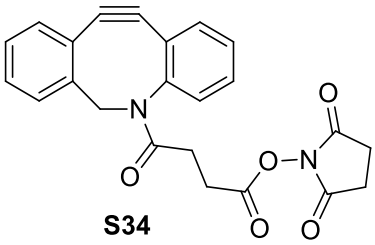
(2*S*)-2-(3-((*S*)-5-Amino-1-carboxypentyl)ureido)pentanedioic acid (S32)[6]



S32

To a solution of di-*tert*-butyl (2*S*)-2-(3-((*S*)-6-amino-1-(*tert*-butoxy)-1-oxohexan-2-yl)ureido)pentanedioate (**S31**)[6] (90 mg, 185 μmol) in anhydrous dichloromethane was added under argon trifluoroacetic acid (115 μL). The reaction mixture was stirred at room temperature for 20 h. The solvent was evaporated under reduced pressure and residual traces of trifluoroacetic acid were removed by co-evaporation under reduced pressure with dichloromethane (2 x 5 mL). The residue was taken up in a mixture of water/acetonitrile (10/2, v/v, 1.2 mL) and purified by preparative RP-HPLC (Combiflash system, conditions D) (t_R = 8.4 min). The collected fractions were pooled to give after freeze drying compound **S32** as an off-white solid (41 mg, 94.6 μmol). Yield: 51%. mp 65-68 °C; IR (ATR accessory) δ 3400-3200, 2945, 2700-2400, 1714, 1662, 1559, 1398, 1186, 1138 cm^{-1}; ^{1}H NMR (200.13 MHz, CD_3OD) δ 1.50 (q, 2H, *J* = 6.8 Hz), 1.58-1.80 (m, 3H), 1.80-2.00 (m, 2H), 2.00-2.25 (m, 1H), 2.42 (t, 2H, *J* = 7.0 Hz), 2.92 (t, 2H, *J* = 6.9 Hz), 4.29 (m, 2H); ^{13}C NMR (50.32 MHz, CD_3OD) δ 23.5, 27.9, 28.7, 31.1, 33.0, 40.5, 53.5, 53.6, 160.2, 176.0, 176.1, 176.5; HRMS *m*/*z* calculated for $C_{12}H_{22}N_3O_7$ $[M+H]^+$: 320.1452, found: 320.1444.

Succinimidyl 4-(11,12-didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-4-oxobutanoate (S34)[7]

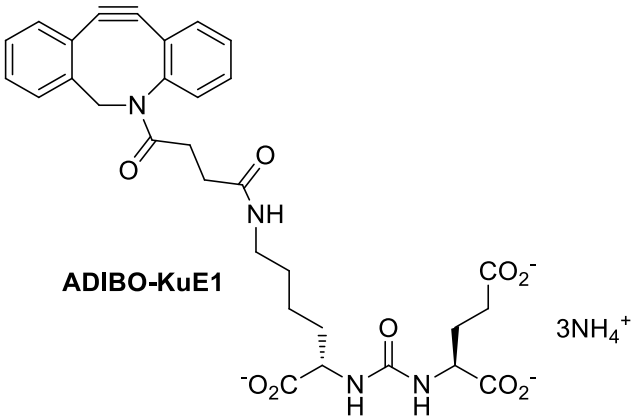


S34

To a solution of 5-(11,12-didehydrodibenzo[*b*,*f*]azocin-5(6*H*)-yl)-4-oxobutanoic acid (**S33**)[8] (1.00 g, 3.28 mmol) in anhydrous dichloromethane (50 mL) were added, under argon, *N*-hydroxysuccinimide (565 mg, 4.91 mmol) and *N*-ethyl-*N'*-(3-dimethylaminopropyl)carbodiimide, hydrochloride salt (942 mg, 4.91 mmol). The reaction mixture was stirred at room temperature for 3.5 h. The solution was then successively washed with 5% aqueous citric acid solution (2 x 50 mL), 5% aqueous sodium hydrogen carbonate solution (2 x 50 mL) and brine (50 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure.

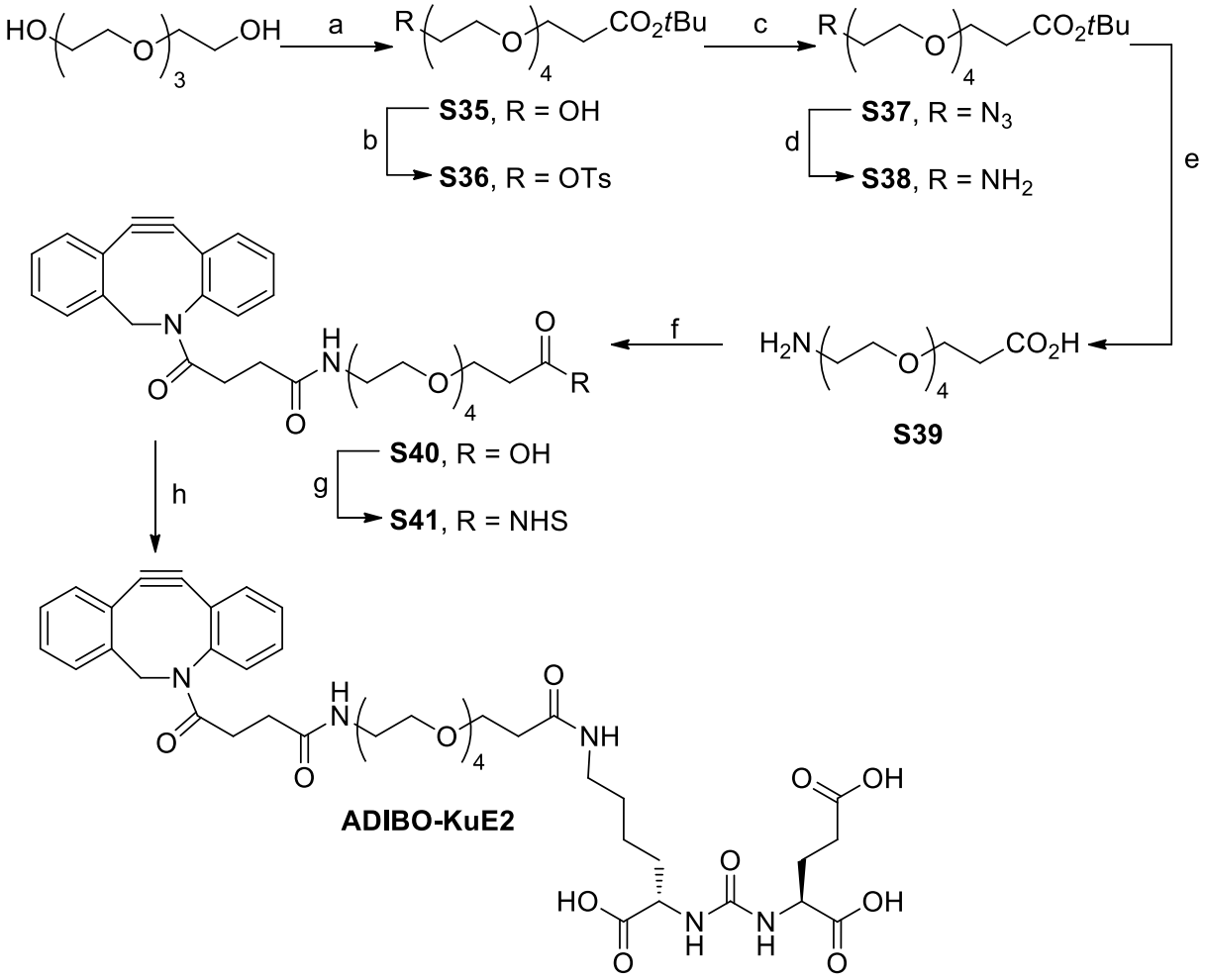
The residue was purified by column chromatography (SiO_2, ethyl acetate/cyclohexane, 50/50, v/v) to give compound **S34** as an off-white solid (1.22 g, 3.03 mmol). Yield: 93%. mp 162-164 °C; R_f = 0.32 (SiO_2, ethyl acetate/cyclohexane, 50/50, v/v); IR (ATR accessory) ν 1734, 1656, 1204, 1069 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 2.08 (m, 1H), 2.55-3.10 (m, 7H), 3.68 (d, 1H, *J* = 13.8 Hz), 5.18 (d, 1H, *J* = 13.8 Hz), 7.20-7.50 (m, 7H), 7.69 (d, 1H, *J* = 7.6 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 25.6 (2C), 26.5, 29.3, 55.7, 107.7, 115.1, 122.8, 123.1, 125.6, 127.3, 127.9, 128.5 (2C), 128.7, 129.2, 132.4, 147.9, 151.1, 168.4, 169.0 (2C), 170.4; ESI-MS *m/z* calculated for $C_{23}H_{18}N_2NaO_5$ $[M+Na]^+$: 425.11, found: 425.09.

(*S*)-2-(3-((*S*)-1-Carboxy-5-(4-oxo-4-(11,12-didehydrodibenzo[b,f]azocin-5-(6*H*)-yl) butanamido)pentyl)ureido)pentanedioic acid ammonium salt (ADIBO-KuE1)



To a solution of **S32** (70 mg, 168 µmol) in anhydrous *N*,*N*-dimethylformamide (200 µL) were successively added, under argon, *N*,*N*-diisopropylethylamine (275 µL, 1.6 mmol) and after 5 min a solution of activated ester **S34** (85 mg, 211 µmol) in anhydrous *N*,*N*-dimethylformamide (500 µL). After stirring at room temperature for 6 h, the resulting solution was diluted with deionized water (1 mL) and purified by preparative RP-HPLC (Combiflash system, conditions B) (t_R = 20.2 min). The collected fractions were pooled, neutralized by addition of small amounts of ammonia to give, after freeze drying, compound **ADIBO-KuE1** as a white solid (64 mg, 106 µmol). Yield: 65%. IR (ATR accessory) ν 3450-3300, 3300-2650, 2650-2300, 1720, 1621, 1558, 1447, 1407, 1159 cm^{-1}; ^1H NMR (500.13 MHz, CD_3OD) (mixture of cis-trans amide bond rotamers) δ 1.28-1.45 (m, 4H), 1.55-1.70 (m, 1H), 1.70-1.83 (m, 1H), 1.85-2.05 (m, 2H), 2.05-2.20 (m, 2H), 2.25-2.42 (m, 3H), 2.60-2.75 (m, 1H), 2.95-3.10 (m, 2H), 3.68 (d, 1H, *J* = 14.0 Hz), 4.05-4.25 (m, 2H), 5.12 and 5.14 (d, 1H, *J* = 14.0 Hz), 7.23 (d, 1H, *J* = 7.4 Hz), 7.30 (td, 1H, *J* = 7.6, 1.3 Hz), 7.35 (td, 1H, *J* = 7.5, 1.4 Hz), 7.40-7.50 (m, 3H), 7.55-7.61 (m, 1H), 7.64 and 7.67 (d, 1H, *J* = 7.2 Hz); ^{13}C NMR (125.76 MHz, CD_3OD) (mixture of cis-trans amide bond rotamers) δ 23.80 and 23.97, 29.77 and 29.84, 29.98 and 30.04, 31.33 and 31.46, 31.92 and 31.97, 32.19 and 32.23, 33.59 and 33.67, 40.15 and 40.23, 55.17 and 55.22, 55.44 and 55.49, 56.69, 108.73 and 108.77, 115.58 and

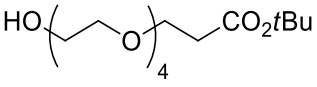
115.59, 123.69 and 123.71, 124.33 and 124.35, 126.44, 128.06, 128.86, 129.18 and 129.20, 129.61, 129.97, 130.55 and 130.57, 133.42 and 133.46, 149.42, 152.64, 160.10, 174.11 and 174.15, 174.34 and 174.39, 178.02 and 178.08, 178.21 and 178.25, 178.75 and 178.79; HRMS *m/z* calculated for $C_{31}H_{35}N_4O_9$ $[M+H]^+$: 607.2399, found: 607.2404.



Reaction conditions: a) NaH (60 wt% in mineral oil), *tert*-butyl acrylate, THF, RT; b) TsCl, pyridine, RT; c) NaN_3, DMF, RT then 55 °C; d) PPh_3, Et_2O, H_2O, RT; e) TFA, anisole, CH_2Cl_2, 0 °C then RT; f) **S34**, DIPEA, DMF, RT; g) NHS, EDC.HCl, CH_2Cl_2, RT; h) **S32**, DIPEA, DMF, RT

Scheme S6: Synthesis of **ADIBO-KuE2** conjugate

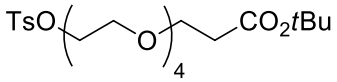
tert-Butyl 1-hydroxy-3,6,9,12-tetraoxapentadecan-15-oate (S35)



S35

Tetraethylene glycol (80.0 g, 0.41 mol) was dried by co-evaporation under reduced pressure with anhydrous toluene (2 x 20 mL) and dissolved under argon in anhydrous tetrahydrofuran (200 mL). Sodium hydride (60 wt% in mineral oil, 290 mg, 7.25 mmol) was then added portionwise. After gas evolution stopped (10 min), a solution of *tert*-butyl acrylate (20.1 mL, 138 mmol) in anhydrous tetrahydrofuran (60 mL) was added dropwise over 1 h. After stirring at room temperature for 20 h, the reaction mixture was neutralized by addition of acetic acid (140 µL) and deionised water (690 µL). After stirring at room temperature for 30 min, brine (150 mL) was added and the resulting solution was extracted with ethyl acetate (3 x 150 mL). The combined organic layers were washed with brine (3 x 150 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S35** as a colourless oil (32.05 g, 99.4 mmol). Yield: 73%; IR (ATR accessory) ν 3700-3300, 3100-2700, 1727, 1366, 1157, 1100 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.40 (s, 9H), 2.46 (t, 2H, J = 6.5 Hz), 2.98 (brs, 1H), 3.50-3.70 (m, 18 H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.1 (3C), 36.2, 61.7, 66.9, 70.4, 70.6 (5C), 72.6, 80.6, 171.0; HRMS *m*/*z* calculated for $C_{15}H_{30}NaO_7$ $[M+Na]^+$: 345.1884, found: 345.1885.

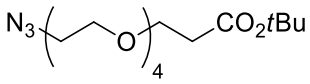
***tert*-Butyl 1-tosyloxy-3,6,9,12-tetraoxapentadecan-15-oate (S36)**



S36

To a solution of **S35** (17.78 g, 55.2 mmol) in anhydrous pyridine (23 mL) was added portionwise, under argon and at 0 °C, tosyl chloride (11.60 g, 60.8 mmol). The reaction was stirred at 0 °C for 2 h then at 4 °C for 60 h. The resulting solution was poured into a mixture of ice/water (300 mL) and extracted with dichloromethane (4 x 100 mL). The combined organic layers were washed successively with a 1N hydrochloric acid solution (2 x 100 mL) and brine (100 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by chromatography (SiO_2, ethyl acetate/cyclohexane 80/20, v/v) to give compound **S36** as a colourless oil (24.06 g, 50.5 mmol). Yield: 91%. R_f = 0.60 (SiO_2, ethyl acetate/cyclohexane, 80/20, v/v); IR (ATR accessory) ν 3050-2850, 1732, 1352, 1174, 1095, 917 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.40 (s, 9H), 2.41 (s, 3H), 2.45 (t, 2H, J = 6.5 Hz), 3.50-3.60 (m, 14H), 3.66 (t, 2H, J = 6.5 Hz), 4.12 (t, 2H, J = 4.7 Hz), 7.31 (d, 2H, J = 8.3 Hz), 7.75 (d, 2H, J = 8.3 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 21.5, 27.9 (3C), 36.1, 66.7, 68.4, 69.2, 70.2, 70.3 (2C), 70.4 (2C), 70.5, 80.2, 127.8 (2C), 129.7 (2C), 132.8, 144.7, 170.7; HRMS *m*/*z* calculated for $C_{22}H_{36}NaO_9S$ $[M+Na]^+$: 499.1972, found: 499.1971.

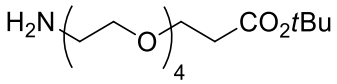
***tert*-Butyl 1-azido-3,6,9,12-tetraoxapentadecan-15-oate (S37)**



S37

To a solution of **S36** (35.20 g, 73.9 mmol) in anhydrous *N*,*N*-dimethylformamide (110 mL) was added under argon sodium azide (16.81 g, 259 mmol). The reaction mixture was stirred at room temperature for 24 h then at 55 °C (external temperature) for 1.5 h. After cooling to room temperature, the reaction was diluted with deionized water (650 mL) and extracted with ethyl acetate (3 x 250 mL). The combined organic layers were washed successively with deionized water (3 x 250 mL) and brine (300 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S37** as a pale yellow oil (25.52 g, 73.5 mmol) which was used in the next step without further purification. Yield: 99%. IR (ATR accessory) ν 2950-2800, 2101, 1727, 1366, 1281, 1253, 1106 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.38 (s, 9H), 2.43 (t, 2H, *J* = 6.5 Hz), 3.33 (t, 2H, *J* = 5.00 Hz), 3.50-3.75 (m, 16H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.0 (3C), 36.2, 50.6, 66.8, 70.0, 70.3, 70.4, 70.6 (4C), 80.4, 170.8; ESI-MS *m/z* calculated for $C_{15}H_{29}N_3NaO_6$ $[M+Na]^+$: 370.20, found: 370.21.

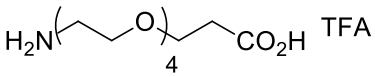
***tert*-Butyl 1-amino-3,6,9,12-tetraoxapentadecan-15-oate (S38)**



S38

To a solution of **S37** (532 mg, 1.53 mmol) in diethyl ether (5 mL) was added deionized water (30 µL, 1.67 mmol) and triphenylphosphine (410 mg, 1.56 mmol). The reaction mixture was stirred at room temperature for 13 h. The solution was filtered and the solid was washed with diethyl ether (2 x 5 mL). The filtrate was evaporated under reduced pressure and the residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol/ammonia, 90/10/0.5, v/v/v) to give compound **S38** as a colourless oil (0.39 g, 1.21 mmol). Yield: 79%. R_f = 0.67 (Al_2O_3, dichloromethane/ethanol/ammonia, 90/10/0.5, v/v/v); IR (ATR accessory) ν 2869, 1727, 1366, 1251, 1104 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.43 (s, 9H), 1.70 (brs, 2H), 2.49 (t, 2H, *J* = 6.6 Hz), 2.85 (t, 2H, *J* = 5.2 Hz), 3.50 (t, 2H, *J* = 5.2 Hz), 3.55-3.60 (m, 12H), 3.69 (t, 2H, *J* = 6.6 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 28.0 (3C), 36.1, 41.5, 66.7, 70.1, 70.2, 70.4 (4C), 73.0, 80.4, 170.8; HRMS *m/z* calculated for $C_{15}H_{32}NO_6$ $[M+H]^+$: 322.2224, found: 322.2212.

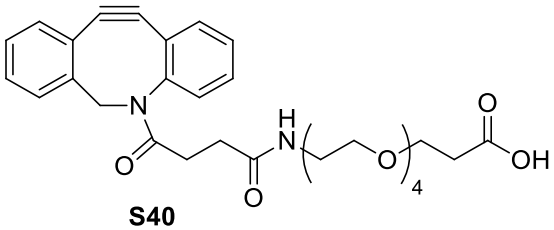
1-Amino-3,6,9,12-tetraoxapentadecan-15-oic acid trifluoroacetic acid salt (S39)



S39

To a solution of **S38** (181 mg, 563 μmol) in anhydrous dichloromethane (1 mL) was successively added, at 0 °C and under argon, anisole (170 μL) and trifluoroacetic acid (500 μL). The reaction mixture was warmed to room temperature and stirred for 24 h. The volatiles were evaporated under reduced pressure and residual traces of trifluoroacetic acid were removed by co-evaporation under reduced pressure with dichloromethane (2 x 5 mL) to give compound **S39** as a yellow oil (213 mg, 562 μmol) which was used in the next step without further purification. Yield: 99%. IR (ATR accessory) ν 3300-3000, 3000-2700, 1723, 1668, 1250-950 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 2.57 (t, 2H, *J* = 5.3 Hz), 3.19 (m, 2H), 3.45-3.85 (m, 16H), 7.30-7.65 (m, 3H), 11.03 (brs, 1H); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 34.7, 40.1, 66.9, 67.0, 69.9, 70.0, 70.1 (2C), 70.2, 70.3, 115.8 (q, $^{1}J_{C\text{-}F}$ = 288.8 Hz), 161.0 (q, $^{2}J_{C\text{-}F}$ = 38.4 Hz), 175.2; HRMS *m/z* calculated for $C_{11}H_{24}NO_6$ $[M+H]^+$: 266.1598, found: 266.1600.

20-(11,12-Didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-17,20-dioxo-4,7,10,13-tetraoxa-16-azaicosan-1-oic acid (S40)

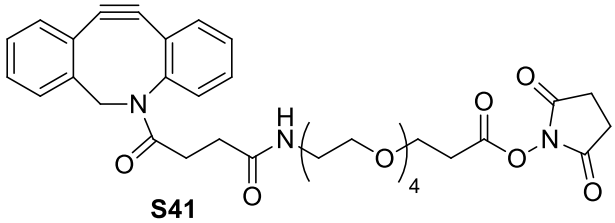


S40

To a solution of **S39** (1.24 g, 3.27 mmol) in anhydrous *N*,*N*-dimethylformamide (20 mL) were successively added, under argon, *N*,*N*-diisopropylethylamine (1.67 mL, 9.82 mmol) and activated ester **S34** (1.10 g, 2.73 mmol). The reaction mixture was stirred at room temperature for 24 h. A 1N hydrochloric acid solution (100 mL) was then added and the resulting solution was extracted with dichloromethane (3 x 100 mL). The organic layers were pooled, washed with brine (3 x 100 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (SiO_2, dichloromethane/ethanol, 90/10, v/v) to give compound **S40** as a colourless oil (1.35 g, 2.44 mmol). Yield: 89%. R_f = 0.14 (SiO_2, dichloromethane/ethanol, 90/10, v/v); IR (ATR accessory) ν 3400-3200, 3000-2700, 1729, 1652, 1435, 1397, 1253, 1184, 1094 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.97 (td, 1H, *J* = 16.5, 6.1 Hz), 2.19 (td, 1H, *J* = 15.1, 6.1 Hz), 2.45 (m, 1H), 2.55 (t, 2H, *J* = 6.1 Hz), 2.84 (m, 1H), 3.35 (m, 2H), 3.48 (m, 2H), 3.55-3.80 (m, 15H), 5.15 (d, 1H, *J* =

13.8 Hz), 6.73 (m, 1H), 7.20-7.45 (m, 6H), 7.54 (m, 1H), 7.66 (m, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 30.2, 31.1, 34.9, 39.3, 55.6, 66.6, 69.9, 70.2, 70.3, 70.4 (2C), 70.6 (2C), 107.9, 114.7, 122.4, 123.2, 125.5, 127.1, 127.8, 128.2, 128.3, 128.8, 129.4, 132.3, 148.0, 151.3, 172.5, 172.8, 174.1; HRMS *m*/*z* calculated for $C_{30}H_{37}N_2O_8$ $[M+H]^+$: 553.2544, found: 553.2544.

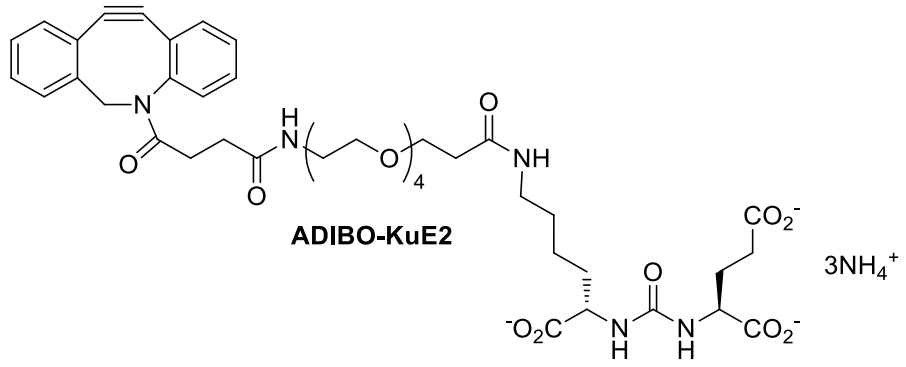
Succinimidyl 20-(11,12-didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-17,20-dioxo-4,7,10,13-tetraoxa-16-azaicosan-1-oate (S41)



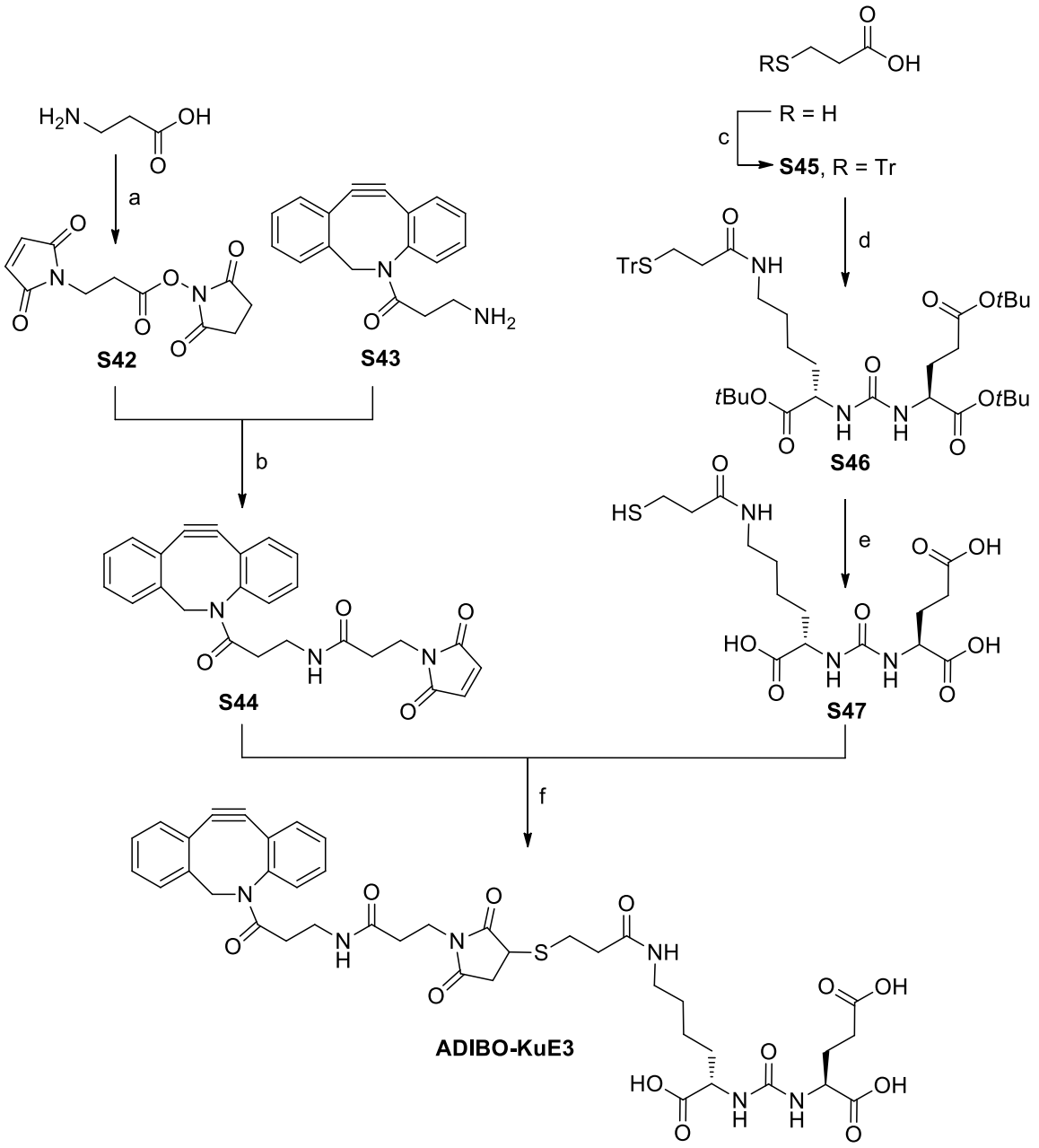
S41

To a solution of **S40** (1.35 g, 2.44 mmol) in anhydrous dichloromethane (50 mL) were successively added, under argon, *N*-hydroxysuccinimide (422 mg, 3.67 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride salt (702 mg, 3.66 mmol). After stirring at room temperature for 22 h, the reaction solution was washed successively with a 5% aqueous citric acid solution (2 x 25 mL), 5% aqueous sodium hydrogen carbonate solution (2 x 25 mL) and brine (2 x 25 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S41** as a yellow oil (1.57 g, 2.31 mmol) which was used in the next step without further purification. Yield: 99%. IR (ATR accessory) ν 3400-3200, 3050-2800, 1814, 1782, 1735, 1655, 1538, 1431, 1396, 1351, 1252, 1203, 1093, 1066 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 1.93 (td, 1H, *J* = 16.3, 6.1 Hz), 2.16 (td, 1H, *J* = 15.1, 6.2 Hz), 2.46 (m, 1H), 2.81 (s, 4H), 2.85 (m, 3H), 3.34 (m, 2H), 3.46 (m, 2H), 3.55-3.75 (m, 13H), 3.82 (t, 2H, *J* = 6.3 Hz), 5.15 (d, 1H, *J* = 13.8 Hz), 6.26 (t, 1H, *J* = 5.2 Hz), 7.20-7.48 (m, 6H), 7.53 (m, 1H), 7.67 (d, 1H, *J* = 6.8 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 25.5 (2C), 30.1, 31.0, 32.1, 39.1, 55.4, 65.6, 69.6, 70.1, 70.4 (3C), 70.5, 70.6, 107.9, 114.6, 122.3, 123.1, 125.4, 127.0, 127.6, 128.1 (2C), 128.6, 129.4, 132.2, 148.1, 151.4, 166.7, 169.0 (2C), 172.0, 172.1; HRMS *m*/*z* calculated for $C_{34}H_{40}N_3O_{10}$ $[M+H]^+$: 650.2708, found: 650.2720.

(3*S*,7*S*)-32-(11,12-Didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-5,13,29,32-tetraoxo-16,19,22,25-tetraoxa-4,6,12,28-tetraazadotriacontane-1,3,7-tricarboxylic acid ammonium salt (ADIBO-KuE2)



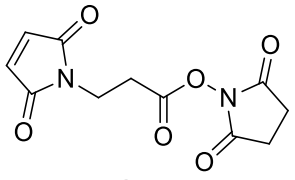
To a solution of **S32** (110 mg, 264 µmol) in anhydrous *N*,*N*-dimethylformamide (200 µL) were successively added, under argon, *N*,*N*-diisopropylethylamine (432 µL, 2.54 mmol) and after 5 min a solution of activated ester **S41** (208 mg, 320 µmol) in anhydrous *N*,*N*-dimethylformamide (300 µL). After stirring at room temperature for 22 h, the reaction mixture was diluted with deionized water (1 mL) and purified by preparative RP-HPLC (Combiflash system, condition B) (t_R = 20.5 min). The collected fractions were pooled, neutralized by addition of small amounts of ammonia to afford, after freeze drying, compound **ADIBO-KuE2** (139 mg, 163 µmol) as an off-white solid. Yield: 62%. IR (ATR accessory) ν 3450-3150, 3150-3000, 3000-2750, 1715, 1633, 1549, 1435, 1398, 1348, 1240, 1201, 1092 cm^{-1}; ^1H NMR (200.13 MHz, CD_3OD) (mixture of cis-trans amide bond rotamers) δ 1.36-1.45 (m, 2H), 1.45-1.57 (m, 2H), 1.58-1.70 (m, 1H), 1.75-1.85 (m, 1H), 1.85-2.02 (m, 2H), 2.05-2.13 (m, 1H), 2.13-2.22 (m, 1H), 2.30-2.40 (m, 3H), 2.42 (t, 2H, , *J* = 6.1 Hz), 2.70 (tdd, 1H, *J* = 2.7 , 7.5, 16.5 Hz), 3.16 (t, 2H, *J* = 6.7 Hz), 3.24 (t, 2H, *J* = 5.6 Hz), 3.43 (sex, 2H, *J* = 5.6 Hz), 3.53-3.63 (m, 11H), 3.66-3.73 (m, 3H), 4.16 (dd, 1H, *J* = 5.0, 6.9 Hz), 4.20 (dd, 1H, *J* = 5.2, 7.7 Hz), 5.12 and 5.13 (d, 1H, *J* = 14.0 Hz), 7.24 (dd, 1H, *J* = 7.3, 1.5 Hz), 7.32 (td, 1H, *J* = 7.5, 1.3 Hz), 7.36 (td, 1H, *J* = 7.5, 1.5 Hz), 7.42-7.49 (m, 3H), 7.57-7.62 (m, 1H), 7.62-7.67 (m, 1H); ^{13}C NMR (50.32 MHz, CD_3OD) (mixture of cis-trans amide bond rotamers) δ 24.01, 29.99, 30.03, 31.37, 31.89, 32.07, 33.78, 37.57, 40.24, 40.33, 55.06, 55.34, 56.68, 68.31, 70.45, 71.19, 71.24, 71.36, 71.41 (2C), 71.43, 108.80, 115.57, 123.67, 124.36, 126.45, 128.10, 128.87, 129.17, 129.64, 130.00, 130.58, 133.43 and 133.44, 149.46, 152.65, 160.03, 173.81, 173.99 and 174.00, 174.61, 177.84, 177.98, 178.51; HRMS *m*/*z* calculated for $C_{42}H_{56}N_5O_{14}$ $[M+H]^+$: 854.3818, found: 854.3834.



Reaction conditions: a) (i) maleic anhydride, DMF, 60 °C; (ii) NHS, DCC, 0 °C then RT; b) CH_3CN, DIPEA, RT; c) TrCl, CH_2Cl_2, RT; d) **S31**, HOBt, DIPEA, EDC.HCl, DMF, -10 °C to RT; e) triisopropylsilane, CH_2Cl_2, TFA, 0 °C then RT; f) *N*-methylmorpholine, DMF, RT.

Scheme S7: Synthesis of **ADIBO-KuE3** conjugate

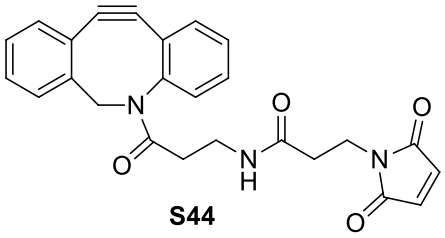
N-Succinimidyl 3-maleimidopropanoate (S42)[9]



S42

To a solution of maleic anhydride (4.30 g, 43.9 mmol) in anhydrous *N*,*N*-dimethylformamide (50 mL) was added under argon 3-aminopropanoic acid (3.91 g, 43.9 mmol). The solution was heated at 60 °C for 2 h. After cooling down to 0 °C, *N*-hydroxysuccinimide (6.31 g, 54.8 mmol) was added, followed by *N*,*N'*-dicyclohexylcarbodiimide over 30 min in several portions. The reaction mixture was warmed to room temperature and stirred for 18 h. The precipitate obtained was filtered and washed successively with deionized water (50 mL) and dichloromethane (50 mL). The filtrate was decanted and the aqueous layer was extracted with dichloromethane (2 x 20 mL). The organic layers were combined, washed successively with deionized water (25 mL) and saturated aqueous sodium hydrogen carbonate solution (2 x 20 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was taken up with ethanol (30 mL) and the resulting solution was stirred for 2 h. The precipitate obtained was filtered, washed with ethanol (2 x 15 mL) and dried overnight in a vacuum desiccator to give compound **S42** as an off-white solid (5.84 g, 21.9 mmol). Yield: 50%. mp 162-164 °C (Lit.[9]: 169-171 °C); IR (ATR accessory) ν 1698, 1380, 1209, 1149, 1068 cm^{-1}; ^{1}H NMR (200.13 MHz, DMSO-d_6) δ 2.79 (s, 4H), 3.04 (t, 2H, J = 6.8 Hz), 3.74 (t, 2H, J = 6.8 Hz), 7.03 (s, 2H); ^{13}C NMR (50.32 MHz, DMSO-d_6) δ 25.5 (2C), 29.1, 32.7, 134.7 (2C), 166.8, 170.1 (2C), 170.6 (2C); HRMS *m/z* calculated for $C_{11}H_{11}N_2O_6$ $[M+H]^+$: 267.0612, found: 267.0612.

N-(3-(11,12-Didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-3-oxopropyl)-3-maleimido propanamide (S44)

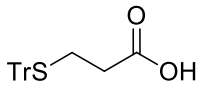


S44

To a solution of *N*-(3-aminopropionyl)-5,6-dihydro-11,12-didehydro-dibenzo[*b*,*f*]azocine (**S43**)[10] (298 mg, 1.08 mmol) in anhydrous acetonitrile (10 mL) were successively added, under argon, activated ester **S42** (431 mg, 1.62 mmol) and *N,N*-diisopropylethylamine (280 μL, 1.6 mmol). The reaction mixture was stirred at room temperature for 1.5 h. The solution was then evaporated under reduced pressure and the residue was purified by column chromatography (Al_2O_3, dichloromethane/ethanol, 98/2, v/v) to give compound **S44** as a white solid (280 mg, 655 μmol). Yield: 61%. mp 79-81 °C; R_f = 0.43 (Al_2O_3, dichloromethane/ethanol, 98/2, v/v); IR (ATR accessory) ν 3400-3150, 2923, 1703, 1645, 1445, 1404, 1230, 1205 cm^{-1}; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.92 (ddd, 1H, J = 3.6, 7.2, 16.7 Hz), 2.32 (m, 2H), 2.47 (ddd, 1H, J = 3.8, 8.0, 16.7 Hz), 3.16 (m, 1H), 3.34 (m, 1H), 3.70 (d, 1H, J = 13.9

Hz), 3.74 (t, 2H, *J* = 7.2 Hz), 5.12 (d, 1H, *J* = 13.9 Hz), 6.08 (m, 1H), 6.66 (s, 2H), 7.27 (dd, 1H, *J* = 1.2, 7.5 Hz), 7.31 (t, 1H, *J* = 7.4 Hz), 7.34-7.43 (m, 5H), 7.66 (d, 1H, *J* = 7.6 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 34.4, 34.7, 34.8, 35.4, 55.7, 107.9, 114.9, 122.7, 123.1, 125.8, 127.4, 128.0, 128.5, 128.6, 128.8, 129.3, 132.2, 134.3 (2C), 148.1, 151.2, 169.5 (2C), 170.5, 172.4; HRMS *m/z* calculated for $C_{25}H_{22}N_3O_4$ $[M+H]^+$: 428.1605, found: 428.1609.

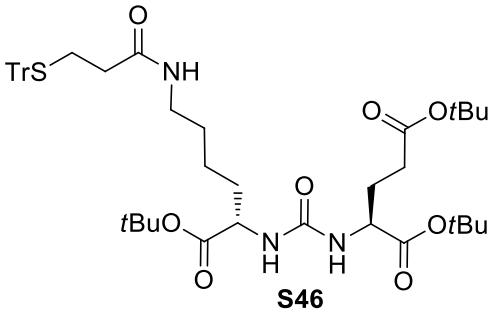
3-(Tritylthio)propanoic acid (S45)[11]



S45

To a solution of 3-mercaptopropionic acid (1.00 mL, 11.5 mmol) in anhydrous dichloromethane (10 mL) was added, over 1 h and under argon, a solution of trityl chloride (3.49 g, 12.5 mmol) in anhydrous dichloromethane (10 mL). The reaction mixture was stirred for 15 h at room temperature. The precipitate obtained was filtered, washed with diethyl ether (3 x 10 mL) and dried overnight in a vacuum desiccator at 35 °C to afford compound **S45** as a white solid (4.00 g, 11.5 mmol). Yield: quant. mp 217-219 °C (lit.[12]: 203-204 °C); IR (ATR accessory) ν 3200-2800, 2571, 1700, 1487, 1430, 1408, 1252 cm^{-1}; ^{1}H NMR (200.13 MHz, DMSO-d_6) δ 2.16 (m, 2H), 2.27 (m, 2H), 7.10-7.45 (m, 15H); ^{13}C NMR (50.32 MHz, DMSO-d_6) δ 27.1, 33.3, 66.6, 127.3 (3C), 128.5 (6C), 129.5 (6C), 144.7 (3C), 173.3; HRMS *m/z* calculated for $C_{22}H_{20}NaO_2S$ $[M+Na]^+$: 371.1076, found: 371.1067.

Di-*tert*-butyl (2*S*)-2-(3-((*S*)-1-*tert*-butoxy-1-oxo-6-(3-(tritylthio)propanamido) hexan-2-yl)ureido)pentanedioate (S46)

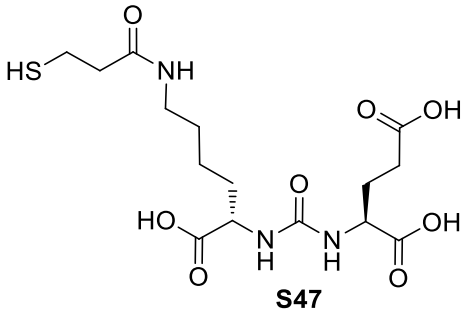


S46

To a solution of **S45** (170 mg, 488 μmol) in anhydrous *N,N*-dimethylformamide (5 mL) were successively added 1-hydroxybenzotriazole (78 mg, 580 μmol) and *N,N*-diisopropylethylamine (195 μL, 1.13 mmol). The solution was cooled down to -10 °C and *N*-ethyl-*N'*-(3-dimethylaminopropyl)carbodiimide hydrochloride salt (121 mg, 631 μmol) and a solution of di-*tert*-butyl (2*S*)-2-(3-((*S*)-6-amino-1-(*tert*-butoxy)-1-oxohexan-2-yl)ureido)pentanedioate (**S31**)[6] (219 mg, 449 μmol) in anhydrous *N,N*-

dimethylformamide (5 mL) were successively added. The solution was warmed up to room temperature and stirred for 22 h. The reaction mixture was evaporated under reduced pressure and the residue was diluted in ethyl acetate (30 mL). The solution was washed with deionized water (2 x 40 mL) and the combined aqueous layers were extracted with ethyl acetate (2 x 80 mL). The combined organic layers were dried over magnesium sulfate, filtered and evaporated under reduced pressure. The residue was purified by column chromatography (SiO_2, cyclohexane/ethyl acetate, 70/30, v/v) to give compound **S46** as a white solid (320 mg, 391 μmol). Yield: 87%. mp 72-74 °C; R_f = 0.14 (SiO_2, cyclohexane/ethyl acetate, 70/30, v/v); IR (ATR accessory) ν 3450-3150, 2977, 2931, 1727, 1643, 1549, 1445, 1367, 1253, 1150 cm^{-1}; ^1H NMR (500.13 MHz, $CDCl_3$) δ 1.25-1.45 (m, 4H), 1.43 (s, 9H), 1.44 (m, 18H), 1.50-1.62 (m, 1H), 1.70-1.80 (m, 1H), 1.80-1.90 (m, 1H), 2.01-2.08 (m, 1H), 2.09 (t, 2H, *J* = 7.3 Hz), 2.20-2.35 (m, 2H), 2.43-2.53 (m, 2H), 3.05-3.15 (m, 1H), 3.15-3.25 (m, 1H), 4.23-4.33 (m, 2H), 5.22 (brs, 2H), 5.87 (m, 1H), 7.20 (t, 3H, *J* = 7.3 Hz), 7.27 (t, 6H, *J* = 7.3 Hz), 7.40 (d, 6H, *J* = 7.3 Hz); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 22.5, 28.0, 28.2 (6C), 28.3 (3C), 28.3, 28.9, 31.8, 32.6, 35.7, 39.2, 53.4, 53.5, 67.0, 80.7, 81.9, 82.3, 126.8 (3C), 128.1 (6C), 129.8 (6C), 144.9 (3C), 157.2, 171.5, 172.5 (2C), 172.5; ESI-MS *m/z* calculated for $C_{46}H_{64}N_3O_8S$ $[M+H]^+$: 818.44, found: 818.46.

(*S*)-2-(3-((*S*)-1-Carboxy-5-(3-mercaptopropanamido)pentyl)ureido)pentanedioic acid (S47)

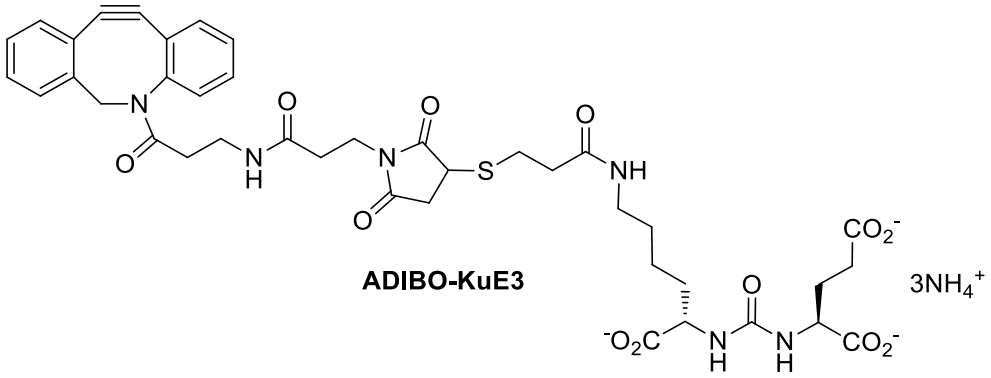


S47

To a solution of **S46** (423 mg, 517 μmol) in anhydrous dichloromethane (5 mL) were successively added, under argon and at 0 °C, trifluoroacetic acid (1.20 mL) and triisopropylsilane (530 μL, 2.59 mmol). The solution was warmed up to room temperature and stirred for 19 h. The volatiles were removed by evaporation under reduced pressure and then by co-evaporation with dichloromethane (5 mL). The residue was diluted with a mixture of deionized water (600 μL) and acetonitrile (1800 μL) and purified by preparative RP-HPLC (Combiflash system, conditions B) (t_R = 14.8 min). The collected fractions were combined, neutralized by addition of small amounts of ammonia to afford, after freeze drying, compound **S47** as a white solid (83.8 mg, 206 μmol). Yield: 40%. IR (ATR accessory) ν 3450-3200, 3200-2700, 2600-2200,

1713, 1621, 1553, 1430, 1413, 1365, 1177 cm^{-1}; ^{1}H NMR (500.13 MHz, CD_3OD) δ 1.40-1.49 (m, 2H), 1.49-1.58 (m, 2H), 1.62-1.71 (m, 1H), 1.78-1.94 (m, 2H), 2.10-2.19 (m, 1H), 2.38-2.45 (m, 2H), 2.48 (t, 2H, *J* = 6.9 Hz), 2.73 (t, 2H, *J* = 6.9 Hz), 3.19 (td, 2H, *J* = 1.6, 7.0 Hz), 4.26 (dd, 1H, *J* = 4.9, 8.6 Hz), 4.31 (dd, 1H, *J* = 4.3, 8.4 Hz); ^{13}C NMR (125.76 MHz, CD_3OD) δ 21.1, 23.9, 28.9, 29.9, 31.1, 33.2, 40.1, 41.0, 53.5, 53.9, 160.1, 173.8, 175.9, 176.4, 176.5; HRMS *m/z* calculated for $C_{15}H_{26}N_3O_8S$ $[M+H]^+$: 408.1435, found: 408.1442.

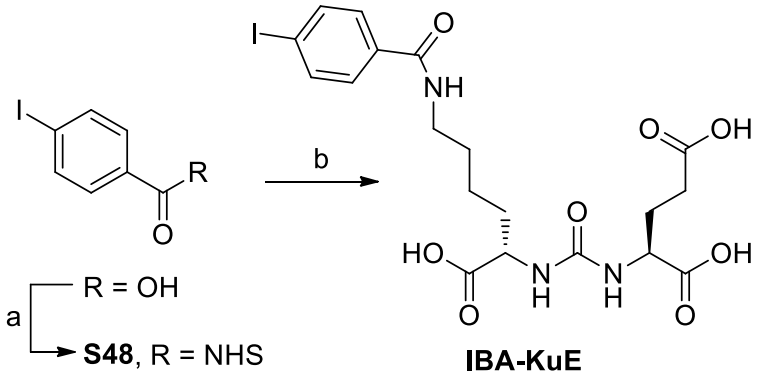
(2*S*)-2-(3-((1*S*)-1-Carboxy-5-(3-((1-(3-((3-(11,12-didehydrodibenzo[b,f]azocin-5(6*H*)-yl)-3-oxopropyl)amino)-3-oxopropyl)-2,5-dioxopyrrolidin-3-yl)thio) propanamido)pentyl)ureido)pentanedioic acid, ammonium salt (ADIBO-KuE3)



To a solution of **S47** (114 mg, 280 μmol) in anhydrous *N*,*N*-dimethylformamide (500 μL) were successively added, under argon, *N*-methylmorpholine (150 μL, 1.36 mmol) and a solution of maleimide **S44** (129 mg, 302 μmol) in anhydrous *N*,*N*-dimethylformamide (500 μL). After stirring at room temperature for 1 h, the resulting solution was diluted with deionized water (1 mL) and purified by preparative RP-HPLC (Combiflash system, conditions C) (t_R = 19.0 min). The collected fractions were combined, neutralized by addition of small amounts of ammonia to afford, after freeze drying, compound **ADIBO-KuE3** as a white solid (66 mg, 79 μmol). Yield: 28%. IR (ATR accessory) ν 3700-3200, 3100-2700, 2700-2300, 1702, 1633, 1558, 1442, 1402, 1206, 1163, 1080, 1045 cm^{-1}; ^{1}H NMR (500.13 MHz, CD_3OD) (mixture of diastereoisomers and/or cis-trans amide bond rotamers) δ 1.36-1.48 (m, 2H), 1.48-1.58 (m, 2H), 1.60-1.70 (m, 1H), 1.76-1.86 (m, 1H), 1.86-1.96 (m, 1H), 1.98-2.07 (m, 1H), 2.07-2.15 (m, 1H), 2.30 (t, 2H, *J* = 7.0 Hz), 2.38 (m, 2H), 2.41-2.49 (m, 2H), 2.50-2.56 (m, 3H), 2.90-3.00 (m, 1H), 3.05-3.25 (m, 7H), 3.65 (t, 2H, *J* = 7.1 Hz), 3.71 (d, 1H, *J* = 14.1 Hz), 3.88 (m, 1H), 4.21 (m, 2H), 5.15 (d, 1H, *J* = 14.1 Hz), *7*.27 (m, 1H), 7.33 (t, 1H, *J* = 7.4 Hz), 7.37 (td, 1H, *J* = 7.4, 1.5 Hz), 7.42-7.50 (m, 3H), 7.53 (m, 1H), 7.66 (d, 1H, *J* = 7.4 Hz); ^{13}C NMR (125.76 MHz, CD_3OD)(mixture of diastereoisomers and/or cis-trans amide bond rotamers) δ 23.99 and 24.01, 28.51 and 28.55, 29.84, 29.93 and 29.94, 31.86, 33.71, 34.70, 35.22 and 35.25, 36.49, 36.77 and 36.80, 36.80

and 36.83, 37.03, 40.31 and 40.32, 40.73-40.81, 54.81, 55.08, 56.63, 108.85, 115.63, 123.68, 124.33, 126.58, 128.16, 128.95, 129.22, 129.69, 130.06, 130.53, 133.42, 149.49, 152.58, 160.07, 172.64, 173.30 and 173.32, 173.61, 176.58 and 176.60, 177.54, 177.58 and 177.59, 178.07, 178.36 and 178.38; HRMS *m/z* calculated for $C_{40}H_{46}N_6NaO_{12}S$ $[M+Na]^+$: 857.2792, found: 857.2782.

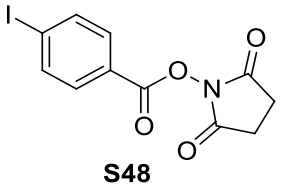
1.5. Synthesis of radiolabelling precursors and references



Reaction conditions: a) NHS, EDC.HCl, CH_2Cl_2, RT; b) **S32**, DIPEA, DMF, RT.

Scheme S8: Synthesis of **IBA-KuE** conjugate

Succinimidyl 4-iodobenzoate (S48)

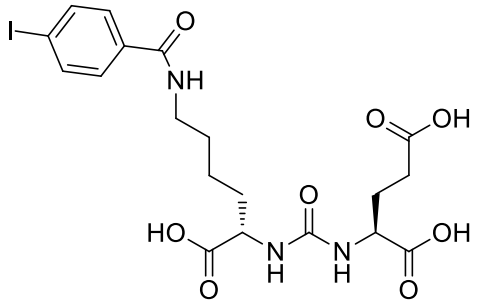


S48

To a solution of 4-iodobenzoic acid (1.00 g, 4.03 mmol) in anhydrous dichloromethane (100 mL) were successively added, under argon, *N*-hydroxysuccinimide (464 mg, 4.03 mmol) and *N*-ethyl-*N'*-(3-dimethylaminopropyl)carbodiimide hydrochloride salt (773 mg, 4.03 mmol). After stirring at room temperature for 2.5 h, the reaction mixture was washed successively with 5% aqueous citric acid solution (30 mL), 10% sodium hydrogen carbonate solution (2 x 30 mL) and deionized water (3 x 30 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S48** as a white solid (1.08 g, 3.13 mmol). Yield: 78%. mp 230-231 °C (Lit.[13]: 212 °C); IR (ATR accessory) ν 1768, 1721, 1584, 1205, 1075, 1049, 987 cm^{-1}; ^1H NMR (200.13 MHz, DMSO-d_6) δ 2.89 (m, 4H), 7.82 (d, 2H, J = 8.3 Hz), 8.05 (d, 2H, J = 8.3 Hz); ^{13}C NMR (50.32 MHz, DMSO-d_6) δ 25.7 (2C), 105.1, 124.0,

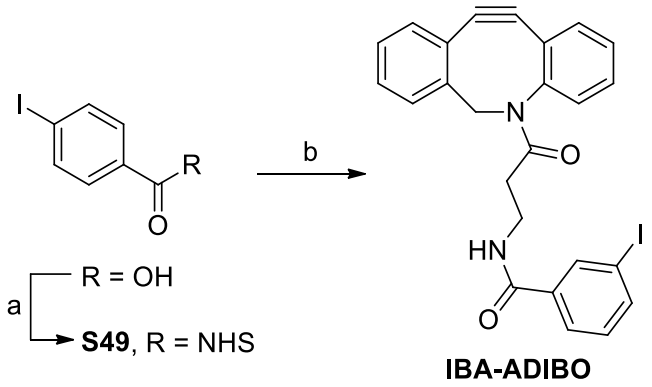
131.5 (2C), 138.8 (2C), 161.8, 170.4 (2C); HRMS *m*/*z* calculated for $C_{11}H_8INNaO_4$ $[M+Na]^+$: 367.9390, found: 367.9392.

(*S*)-2-(3-((*S*)-1-Carboxy-5-(4-iodobenzamido)pentyl)ureido)pentanedioic acid (IBA-KuE)[14]



IBA-KuE

To a solution of **S32** (188 mg, 0.45 mmol) in anhydrous *N*,*N*-dimethylformamide (1 mL) were successively added, under argon, *N*,*N*-diisopropylethylamine (370 µL, 2.17 mmol) and after 5 min activated ester **S48** (150 mg, 0.43 mmol). After stirring at room temperature for 25 h, the reaction mixture was diluted with deionized water (1 mL) and purified by preparative RP-HPLC (Combiflash system, conditions A) (t_R = 21 min) to afford, after freeze drying, compound **IBA-KuE** as an off-white solid (125 mg, 0.23 mmol). Yield: 52**%**. IR (ATR accessory) ν 3450-3200, 3200-2700, 2700-2300, 1706, 1629, 1588, 1545, 1410, 1304, 1211, 1175, 1115 cm^{-1}; ^1H NMR (200.13 MHz, CD_3OD) δ 1.30-2.00 (m, 7H), 2.20-2.30 (m, 1H), 2.41 (m, 2H), 3.33 (t, 2H, *J* = 6.6 Hz), 4.30 (m, 2H), 7.55 (d, 2H, *J* = 8.5 Hz), 7.82 (d, 2H, *J* = 8.5 Hz); ^{13}C NMR (50.32 MHz, CD_3OD) δ 24.0, 28.8, 29.9, 31.1, 33.1, 40.8, 50.3, 53.5, 99.0, 130.0 (2C), 135.2, 138.8 (2C), 160.1, 169.4, 175.9, 176.5 (2C); HRMS *m*/*z* calculated for $C_{19}H_{25}IN_3O_8$ $[M+H]^+$: 550.0681, found: 550.0685.

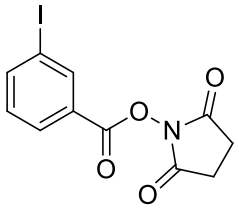


IBA-ADIBO

Reaction conditions: a) DSC, pyridine, CH_3CN, RT;
b) **S43**, DIPEA, DMF, RT.

Scheme S9: Synthesis of **IBA-ADIBO** conjugate

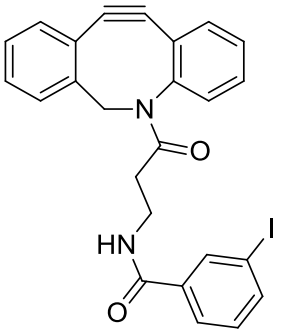
Succinimidyl 3-iodobenzoate (S49)



S49

To a solution of 3-iodobenzoic acid (1.09 g, 4.39 mmol) in anhydrous acetonitrile (60 mL) were successively added, under argon, anhydrous pyridine (427 µL, 5.28 mmol) and *N*,*N*'-disuccinimidyl carbonate (1.35 g, 5.27 mmol). After stirring at room temperature for 6 h, the reaction mixture was evaporated under reduced pressure. The residue was taken up in dichloromethane (100 mL) and washed successively with 5% aqueous citric acid solution (30 mL), 5% sodium hydrogen carbonate solution (2 x 30 mL) and deionized water (3 x 30 mL). The organic layer was dried over magnesium sulfate, filtered and evaporated under reduced pressure to give compound **S49** as a white solid (1.14 g, 3.30 mmol). Yield: 75%. mp 143-145 °C (Lit.[15]: 147-148 °C); IR (ATR accessory) ν 1773, 1726, 1228, 1203, 1066, 1047, 1005, 991 cm^{-1}; ^{1}H NMR (200.13 MHz, $CDCl_3$) δ 2.90 (s, 4H), 7.26 (t, 1H, *J* = 7.7 Hz), 8.00 (d, 1H, *J* = 7.7 Hz), 8.09 (d, 1H, *J* = 7.7 Hz), 8.44 (s, 1H); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 25.7 (2C), 94.1, 126.9, 129.6, 130.5, 139.0, 143.7, 160.5, 169.2 (2C); HRMS *m*/*z* calculated for $C_{11}H_8INNaO_4$ $[M+Na]^+$: 367.9390, found: 367.9394.

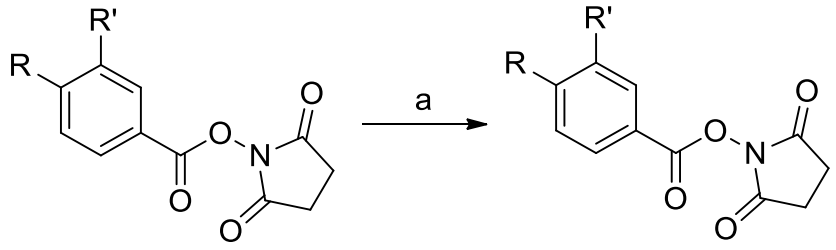
***N*-(3-(3-Iodobenzamido)propionyl)-5,6-dihydro-11,12-didehydrodibenzo[b,f]azocine (IBA-ADIBO)**



IBA-ADIBO

To a solution of *N*-(3-aminopropionyl)-5,6-dihydro-11,12-didehydro-dibenzo[*b*,*f*]azocine (**S43**)[10] (122 mg, 0.44 mmol) in anhydrous *N*,*N*-dimethylformamide (1 mL) were successively added, under argon, *N*,*N*-diisopropylethylamine (150 µL, 0.88 mmol) and after 5 min activated ester **S49** (152 mg, 0.44 mmol). The reaction mixture was stirred at room temperature for 24 h. The resulting solution was evaporated under reduced pressure and the residue purified

by column chromatography (SiO_2, ethyl acetate/cyclohexane, 30/70→50/50, v/v) to give compound **IBA-ADIBO** as an off-white solid (137 mg, 0.27 mmol). Yield: 61%. mp 76-78 °C; R_f = 0.08 (SiO_2, cyclohexane/ethyl acetate, 30/70, v/v); IR (ATR accessory) ν 3450-3200, 2924, 1640, 1558, 1523, 1480, 1466, 1432, 1396, 1285, 1228, 1204 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 2.06 (ddd, 1H, *J* = 16.6, 7.3, 4.1 Hz), 2.53 (ddd, 1H, *J* = 16.6, 6.8, 4.3 Hz), 3.30-3.60 (m, 2H), 3.67 (d, 1H, *J* = 13.8 Hz), 5.12 (d, 1H, *J* = 13.8 Hz), 6.78 (t, 1H, 5.7 Hz), 7.06 (t, 1H, *J* = 8.0 Hz), 7.12 (d, 1H, *J* = 7.3 Hz), 7.20-7.50 (m, 7H), 7.66 (d, 1H, *J* = 7.1 Hz), 7.76 (d, 1H, *J* = 7.9 Hz), 7.95 (t, 1H, *J* = 1.5 Hz); ^{13}C NMR (50.32 MHz, $CDCl_3$) δ 34.7, 35.9, 55.6, 94.2, 107.7, 114.8, 122.5, 122.8, 125.7, 126.1, 127.3, 127.9, 128.3, 128.5, 128.6, 129.0, 130.1, 132.1, 136.1, 136.4, 140.1, 147.9, 150.9, 165.6, 172.3; HRMS *m/z* calculated for $C_{25}H_{20}IN_2O_2$ $[M+H]^+$: 507.0564, found: 507.0572.



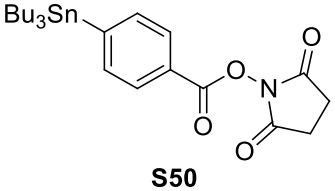
S48, R = I, R' = H
S49, R = H, R' = I

S50, R = $SnBu_3$, R' = H
S51, R = H, R' = $SnBu_3$

Reaction conditions: a) Sn_2Bu_6, $Pd(PPh_3)_4$, toluene, reflux.

Scheme S10: Synthesis of stannylated precursors **S50** and **S51**

Succinimidyl 4-(tributylstannyl)benzoate (S50)

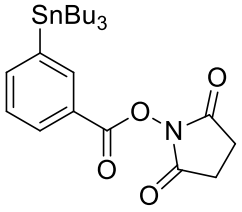


S50

To a solution of iodinated compound **S48** (300 mg, 0.87 mmol) in anhydrous toluene (10 mL) was added hexabutylditin (0.88 mL, 1.74 mmol). The solution was degassed, under argon, for 10 min before addition of tetrakis(triphenylphosphine)palladium(0) (20 mg, 17 µmol). The resulting solution was refluxed for 5 h under argon. After cooling to room temperature, the reaction mixture was evaporated under reduced pressure and the residue was purified by column chromatography (SiO_2, dichloromethane) to give

compound **S50** as a colourless oil (387 mg, 0.76 mmol). Yield: 88%; R_f = 0.23 (SiO_2, dichloromethane); IR (ATR accessory) ν 2955, 2922, 2870, 2851, 1770, 1739, 1254, 1233, 1201, 1068, 994 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 0.88 (t, 9H, J = 7.2 Hz), 1.09 (m, 6H), 1.20-1.70 (m, 12H), 2.89 (s, 4H), 7.62 (d, 2H, J = 8.2 Hz, $J_{H-}{}^{119}_{Sn}$ = 35.6 Hz, $J_{H-}{}^{117}_{Sn}$ = 34.1 Hz), 8.03 (d, 2H, J = 8.2 Hz); NMR ^{13}C (50.32 MHz, $CDCl_3$) δ 9.9 (3C, $J_{C-}{}^{119}_{Sn}$= 343 Hz, $J_{C-}{}^{117}_{Sn}$ = 328 Hz), 13.8 (3C), 25.8 (2C), 27.4 (3C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 56 Hz), 29.1 (3C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 21 Hz), 124.5, 129.2 (2C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 38 Hz), 136.9 (2C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 30 Hz), 153.3, 162.4, 169.5 (2C); HRMS *m*/*z* calculated for $C_{23}H_{35}NO_4Sn$ $[M+H]^+$: 510.1661, found: 510.1666.

Succinimidyl 3-(tributylstannyl)benzoate (S51)



S51

Compound **S51** (304 mg, 0.60 mmol) was obtained as a colourless oil according to the procedure described for **S50** starting from **S49** (325 mg, 0.94 mmol); reaction time at reflux of 6.5 h; the crude was purified by column chromatography (SiO_2, cyclohexane/ethyl acetate, 20/80, v/v). Yield 64%; R_f = 0.31 (SiO_2, cyclohexane/ethyl acetate, 20/80, v/v); IR (ATR accessory) ν 2955, 2923, 2871, 2851, 1770, 1739, 1231, 12011067, 1006 cm^{-1}; ^1H NMR (200.13 MHz, $CDCl_3$) δ 0.88 (t, 9H, J = 7.1 Hz), 1.09 (m, 6H), 1.20-70 (m, 12H), 2.89 (s, 4H), 7.44 (t, 1H, J = 7.5 Hz), 7.76 (d, 1H, J = 7.2 Hz, $J_{H-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 34.8 Hz), 8.05 (d, 1H, J = 7.9 Hz), 8.19 (s, 1H, $J_{H-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 40.8 Hz); NMR ^{13}C (50.32 MHz, $CDCl_3$) δ 9.7 (3C, $J_{C-}{}^{119}_{Sn}$ = 347 Hz, $J_{C-}{}^{117}_{Sn}$ = 332 Hz), 13.7 (3C), 25.8 (2C), 27.4 (3C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 55.7 Hz), 29.0 (3C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 20.4 Hz), 124.5, 128.1 (1C, $J_{C-}{}^{117}_{Sn/}{}^{119}_{Sn}$ = 35.4 Hz), 130.1, 138.2, 143.0, 143.6, 162.5, 169.5 (2C); HRMS *m*/*z* calculated for $C_{23}H_{36}NO_4Sn$ $[M+H]^+$: 510.1661, found: 510.1665.

2. Radiochemistry

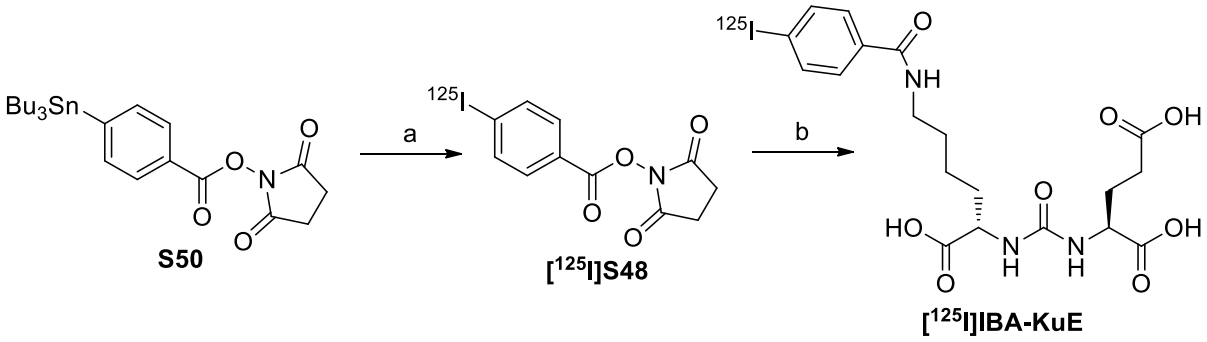
2.1. Materials and methods

$[^{125}I]$NaI (3.70 GBq/mL, 643.8 MBq/mg) was purchased from PerkinElmer, Inc. as a no-carrier-added solution in reductant free 1.0 × 10^{-5} M aqueous sodium hydroxide solution (pH 8-11). Sep-Pack light C18 and Chromafix dry (Na_2SO_4) cartridges were

purchased from Waters and Macherey-Nagel, respectively. Size exclusion chromatographies were performed using prepacked PD-10 column (Sephadex^{TM} G-25M, GE Healthcare Life Sciences). Radio-instant thin-layer chromatography (radio-ITLC) analyses were measured on a miniGITA Dual radio-TLC system (Elysia-Raytest) using silica gel-impregnated chromatography paper (Varian inc.). Analytical RP-HPLC measurements were performed on a system consisting of a HP1100 (Hewlett Packard, Les Ulis, France) and a Flow one A_{500} Radiomatic detector (Packard, Canberra, Australia). The separation was carried out on a C_{18} column (zorbax 80 Å, 4.6 × 150 mm, 5 µm, Agilent). Semi-preparative RP-HPLC purifications of radiotracers **[^{125}I]S48**, **[^{125}I]IBA-KuE**, **[^{125}I]S49** and **[^{125}I]IBA-ADIBO** were performed on a Perkin Elmer system consisting of a Flexar LC autosampler and PDA detector, a Series 200 pump, a Peltier column oven and vacuum degasser, and a GabiStar Raytest detector. The separation was carried out on a C_{18} column (zorbax 80 Å, 4.6 × 150 mm, 5 µm, Agilent). All radiotracers were shown by radio-RP-HPLC to be identical to the authentic non-radioactive material and to be free of significant chemical and radiochemical impurities.

Molar activities were determined based on a calibration curve carried out under analytical HPLC conditions at an appropriate maximum of UV absorbance (λ = 268 nm for **[^{125}I]S48**, λ = 250 nm for **^{125}I]IBA-KuE**, λ = 232 nm for **[^{125}I]S49**, λ = 240 nm for **[^{125}I]IBA-ADIBO**.

2.2. Preparation of 2-[3-[1-carboxy-5-(4-[^{125}I]iodobenzoylamino)pentyl]-ureido]pentanedioic acid ([^{125}I]IBA-KuE)[14]



Reaction conditions: a) [^{125}I]NaI, CAT, AcOH, MeOH, RT; b) **S32**, DIPEA, DMSO, RT.

Scheme S11: Radiosynthesis of **[^{125}I]IBA-KuE**

To a methanolic solution of organotin precursor **S50** (50 µL, 1 mg.mL^{-1}) were successively added [^{125}I]NaI (16 µL, 48.84 MBq), acetic acid (2.5 µL) and a methanolic solution of Chloramine-T (5 µL, 1 mg.mL^{-1}). The mixture was stirred at room temperature for 15 min. The reaction was quenched by addition of an aqueous sodium metabisulfite solution (5 µL, 1 mg.mL^{-1}) and purified by semi-preparative RP-HPLC (acetonitrile/water (0.1% TFA), 45/55, v/v, isocratic elution in 20 min; flow rate: 1.0 mL/min, t_R = 7.4 min). The fraction containing the radiolabelled product was collected, diluted with deionized water (20 mL) and loaded onto a SepPack® light C18 cartridge (previously washed successively with deionized water (5 mL), methanol (5 mL) and deionised water (5 mL)). Air was passed through the cartridge before connection of a Chromafix® Dry (Na_2SO_4) cartridge to its bottom. The columns were eluted with acetonitrile (2 mL) and the eluate was evaporated to dryness at 40 °C using an argon stream (15-20 min) to afford **[^{125}I]S48** (44.8 MBq, radiochemical yield: 92%, radiochemical purity: >98%). Analytical RP-HPLC conditions: acetonitrile/water (0.1% TFA), 45/55, v/v, isocratic elution for 10 min then linear gradient 45/55 → 100/0, v/v, linear gradient in 5 min; flow rate: 1.0 mL/min, t_R = 7.0 min.

To the activated ester **[^{125}I]S48** (44.4 MBq) were successively added a solution of (*S*)-2-(3-((*S*)-5-amino-1-carboxypentyl)ureido)pentanedioic acid (**S32**) (250 µL, 10 mg.mL^{-1} in anhydrous dimethyl sulfoxide) and *N*,*N*-diisopropylethylamine (5 µL). The reaction mixture was vortexed every 10 min for 45 min, quenched by addition of trifluoroacetic acid (3 µL) and purified by semi-preparative RP-HPLC (acetonitrile/water (0.1% TFA), , linear gradient 20/80 → 70/30, v/v in 20 min; flow rate: 1.0 mL/min, t_R = 8.8 min). The fraction containing the radiolabelled product was collected, diluted with deionized water (20 mL) and loaded onto a SepPack® light C18 cartridge (previously washed successively with deionized water (5 mL), methanol (5 mL) and deionized water (5 mL)). Air was passed through the cartridge before elution with ethanol (2 mL). The eluate was evaporated to dryness at 40 °C using an argon stream (15-20 min) and the product was diluted with dimethyl sulfoxide to afford **[^{125}I]IBA-KuE** (17.8 MBq) (radiochemical yield: 40%, overall radiochemical yield: 37%, radiochemical purity: >99%, molar activity (over 5 experiments): 2.2-21.7 GBq.µmol^{-1}). Analytical RP-HPLC conditions: acetonitrile:water (0.1% TFA), linear gradient 20/80 → 100/0, v/v, in 20 min; flow rate: 1.0 mL/min, t_R = 6.3 min. Stored at -80 °C, the solution of **[^{125}I]IBA-KuE** in dimethyl sulfoxide was stable over 2 weeks.

2.3. Analytical (radio/UV)-HPLC chromatograms of S48, [^{125}I]S48, IBA-KuE and [^{125}I]IBA-KuE

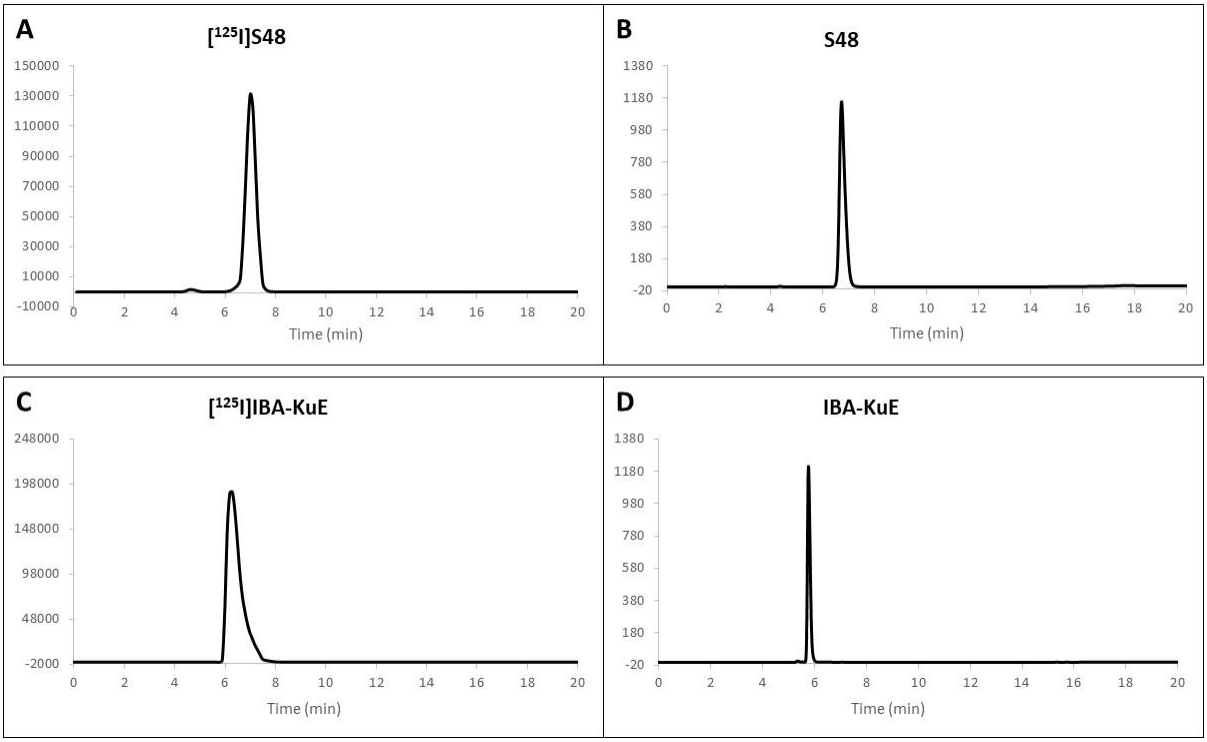
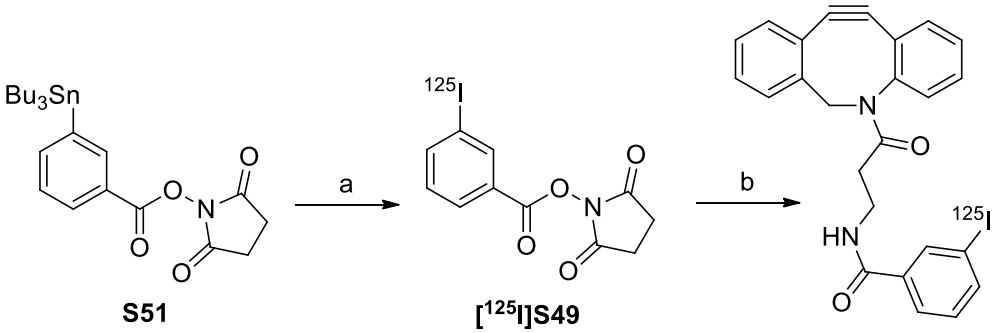


Figure S1. Analytical radio-HPLC chromatograms of purified **[^{125}I]S48** (**A**) and **[^{125}I]IBA-KuE** (**C**) compared to analytical UV-HPLC chromatograms of non radioactive references **S48** (**B**) and **IBA-KuE** (**D**). Radio-HPLC detector was connected in series after the UV detector accounting for the slight difference of retention times (≈ 0.3 min) observed between ^{125}I and ^{127}I products.

2.4. Preparation of *N*-(3-(3-[^{125}I]iodobenzamido)propionyl)-5,6-dihydro-11,12-didehydrodibenzo[*b*,*f*]azocine ([^{125}I]IBA-ADIBO)



S51 **[^{125}I]S49** **[^{125}I]IBA-ADIBO**

Reaction conditions: a) [^{125}I]NaI, CAT, AcOH, MeOH, RT; b) **S43**, DIPEA, DMSO, RT.

Scheme S12: Radiosynthesis of **[^{125}I]IBA-ADIBO**

To a methanolic solution of organotin precursor **S51** (50 μL, 1 mg.mL^{-1}) were successively added [^{125}I]NaI (5 μL, 28.9 MBq), acetic acid (2.5 μL) and a methanolic solution of Chloramine-T (5 μL, 1 mg.mL^{-1}). The mixture was stirred at room temperature for 15 min. The reaction was quenched by addition of an aqueous sodium metabisulfite solution (5 μL, 1 mg.mL^{-1}) and purified by semi-preparative RP-HPLC (acetonitrile/water (0.1% TFA), 45/55, v/v, isocratic elution for 20 min; flow rate: 1.0 mL/min, t_R = 7.5 min). The fraction containing the radiolabelled product was collected, diluted with deionized water (20 mL) and loaded onto a SepPack® light C18 cartridge (previously washed successively with deionized water (5 mL), methanol (5 mL), and deionized water (5 mL)). Air was passed through the cartridge before connection of a Chromafix® Dry (Na_2SO_4) cartridge to its bottom. The columns were eluted with acetonitrile (1.5 mL) and the eluate was evaporated to dryness using an argon stream at 40 °C (15-20 min) to afford **[^{125}I]S49** (20.4 MBq) (radiochemical yield: 71%, radiochemical purity: >99%). Analytical RP-HPLC conditions: acetonitrile/water (0.1% TFA), 45:55 isocratic elution in 10 min then linear gradient 45/55 → 100/0, v/v, in 5 min; flow rate: 1.0 mL/min, t_R = 6.5 min.

To **[^{125}I]S49** (20.4 MBq) were successively added a solution of amine **S43** (20 μL, 10 mg.mL^{-1} in anhydrous dimethyl sulfoxide), anhydrous dimethyl sulfoxide (40 μL) and *N*,*N*-diisopropylethylamine (3 μL). The reaction mixture was vortexed every 10 min for 1.5 h, diluted with a solution of acetonitrile containing 0.1% TFA (100 µL) and purified by semi-preparative RP-HPLC (acetonitrile/water (0.1% TFA), linear gradient 30/70 → 100/0, v/v in 25 min; flow rate: 1.0 mL/min, t_R = 19.2 min). The fraction containing the radiolabelled product was collected, diluted with deionized water (20 mL) and loaded onto a SepPack® light C18 cartridge (previously washed successively with deionized water (5 mL), methanol (5 mL) and deionized water (5 mL)). Air was passed through the cartridge before connection of a Chromafix® Dry (Na_2SO_4) cartridge to its bottom. The columns were eluted with acetonitrile (2 mL) and the eluate was evaporated to dryness using an argon stream at 40 °C (15-20 min). The crude was diluted with dimethyl sulfoxide (200 µL) to afford **[^{125}I]IBA-ADIBO** (11.3 MBq) (radiochemical yield: 55%, overall radiochemical yield: 39%, radiochemical purity: >99%, molar activity (over 8 experiments): 2.39-56.2 GBq. μmol^{-1}). Analytical RP-HPLC conditions: acetonitrile/water (0.1% TFA), linear gradient 70/30 → 100/0, v/v in 15 min then isocratic elution 100/0, v/v for 5 min; flow rate: 1.0 mL/min, t_R = 8.1 min. The solution of **[^{125}I]IBA-ADIBO** in dimethyl sulfoxide was stable over 5 h at room temperature.

2.5. Analytical (radio/UV)-HPLC chromatograms of S49, [^{125}I]S49, IBA-ADIBO and [^{125}I]IBA-ADIBO

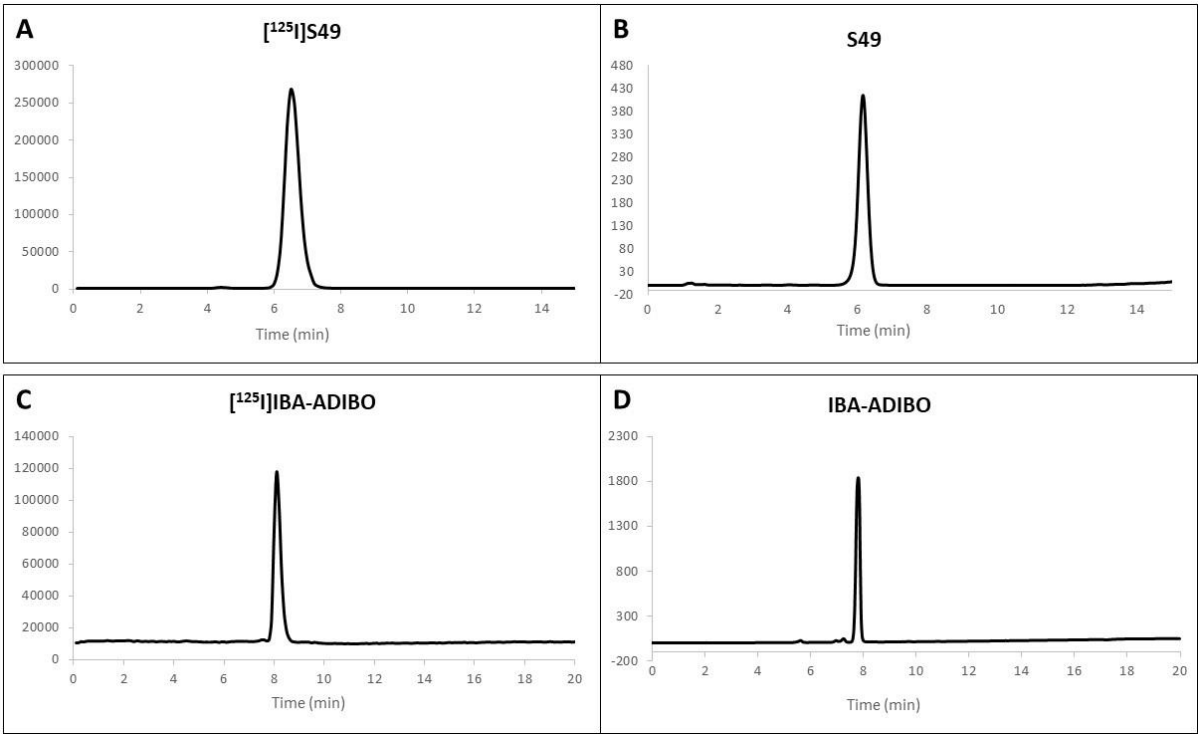


Figure S2. Analytical radio-HPLC chromatograms of purified **[^{125}I]S49** (**A**) and **[^{125}I] IBA-ADIBO** (**C**) compared to analytical UV-HPLC chromatograms of non radioactive references **S49** (**B**) and **IBA-ADIBO** (**D**). Radio-HPLC detector was connected in series after the UV detector accounting for the slight difference of retention times (≈ 0.3 min) observed between ^{125}I and ^{127}I products.

3. Supplementary figures

3.1. Characterization of UCNP-OA obtained from the two steps procedure

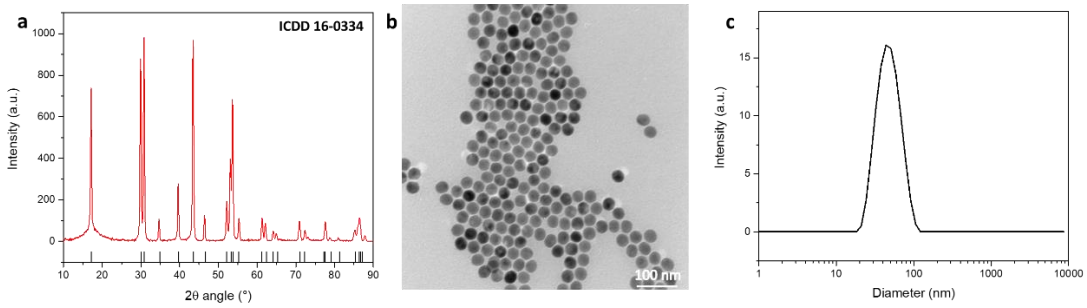


Figure S3: (**a**) XRD pattern, (**b**) TEM image, and (**c**) DLS distribution in cyclohexane of core/shell UCNP-OA obtained from the two steps synthesis.

3.2. ATR-FTIR spectra of UCNP-PEG

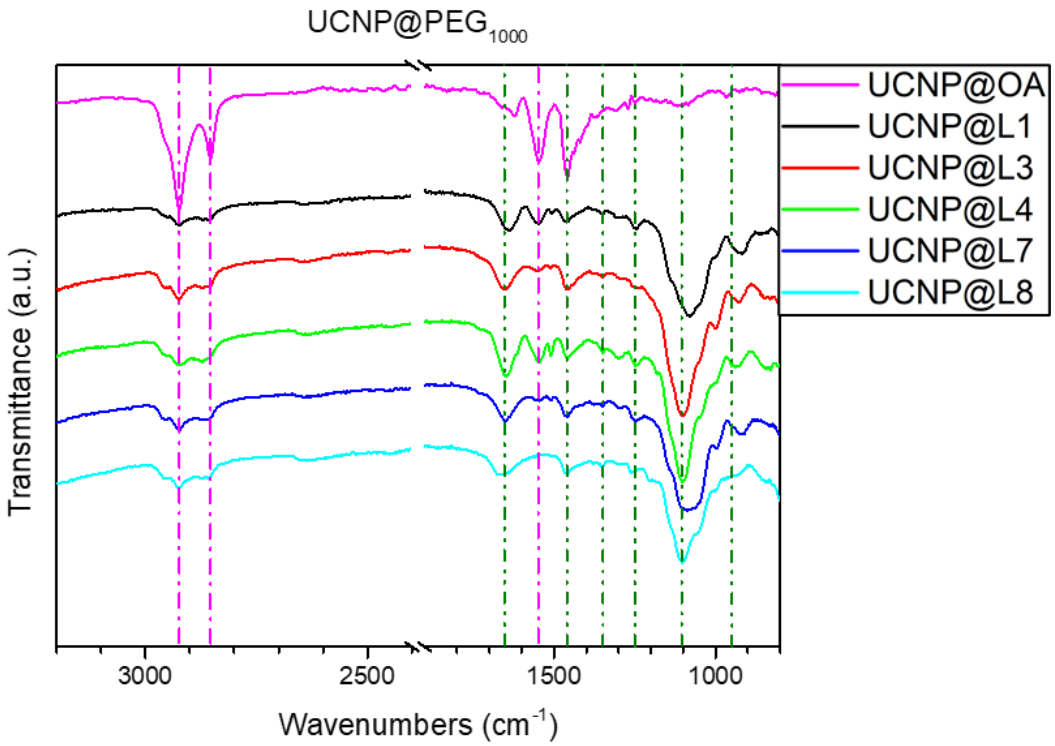


Figure S4: ATR-FTIR spectra of UCNP@OA and UCNP after ligand exchange with $PEG_{(1000)}$ ligands (UCNP@L1, L3, L4, L7 and L8). Magenta dashed lines highlight OA =C-H and C=O bands while green dashed lines highlight PEG backbone bands.

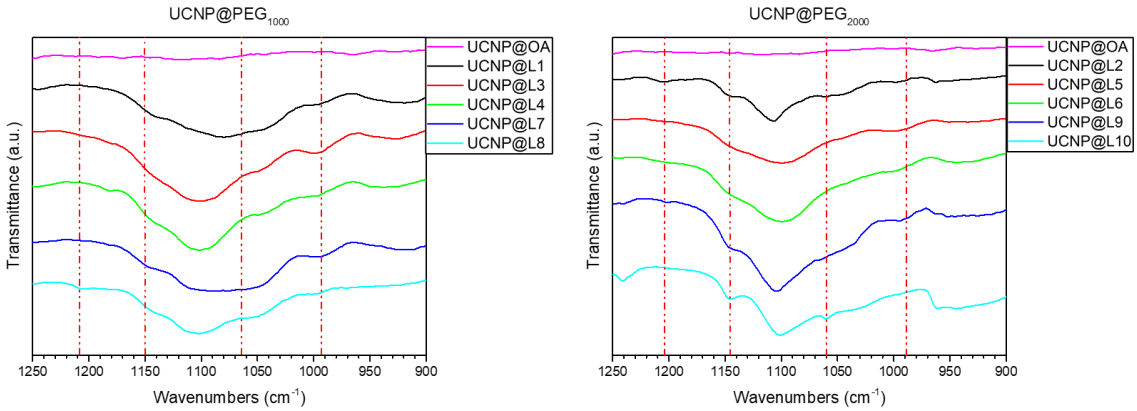


Figure S5: ATR-FTIR spectra of UCNP@OA and UCNP@L(1-10), zoom in the range of 900-1250 cm^{-1}. Red dashed lines highlight P=O and P-O bands not or faintly visible in the spectra.

3.3. TGA analysis

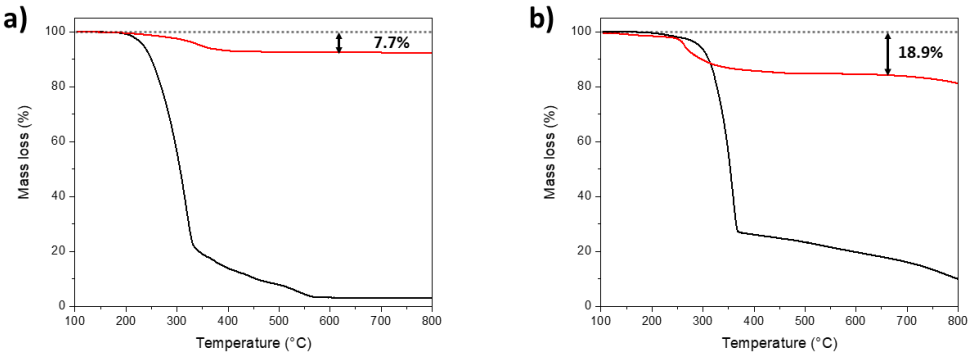


Figure S6. (**a**) TGA profile of UCNP@OA and (**b**) UCNP@L9. Red and black lines show the degradation profile of UCNP and organic ligands, respectively.

3.4. UCL spectra of UCNP@OA and UCNP@L9

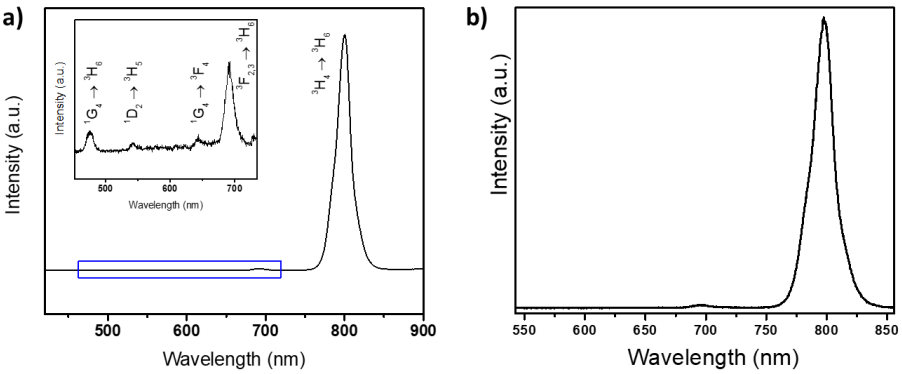
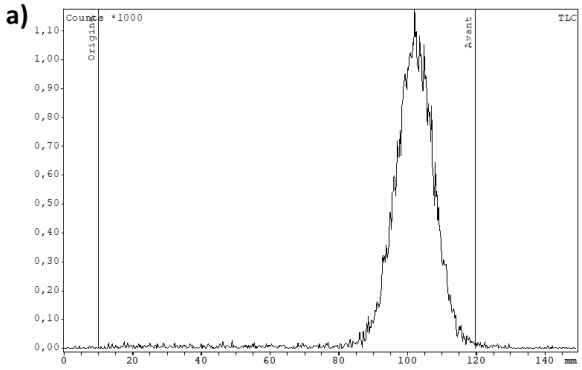


Figure S7. (**a**) Full UCL spectrum recorded upon 980 nm excitation from one-pot synthesized UCNPs, upper insert shows all the Tm^{3+} transitions in the 450-750 nm range exhibiting a weak intensity in comparison with that of the $^3H_4 \rightarrow {}^3H_6$ transition at 800 nm; (**b**) UCL spectrum recorded from UCNP@L9 in deionized water upon excitation at 980 nm.

3.5. ITLC radio-chromatograms of radioiodinated IBA-ADIBO and UCNP



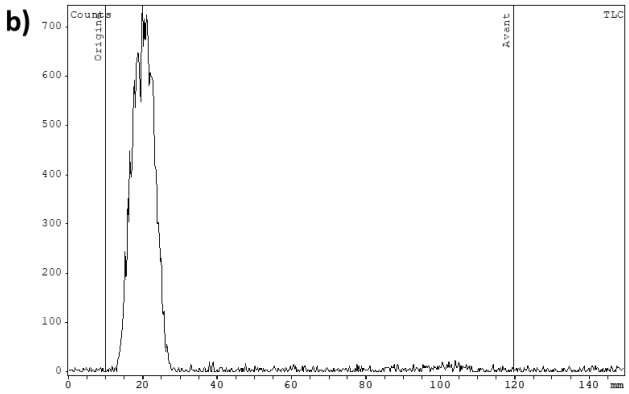


Figure S8. Examples of ITLC radio-chromatograms obtained after elution with acetone of: (**A**) **[^{125}I]IBA-ADIBO**; (**B**) UCNP@N_3(10%) radiolabelled with **[^{125}I]IBA-ADIBO** (reaction time: 30 min at 37 °C).

3.6. Radioactive-based quantification of UCNP azide functions with [^{125}I]IBA-ADIBO after conjugation with increasing concentrations of ADIBO-CO_2H S33

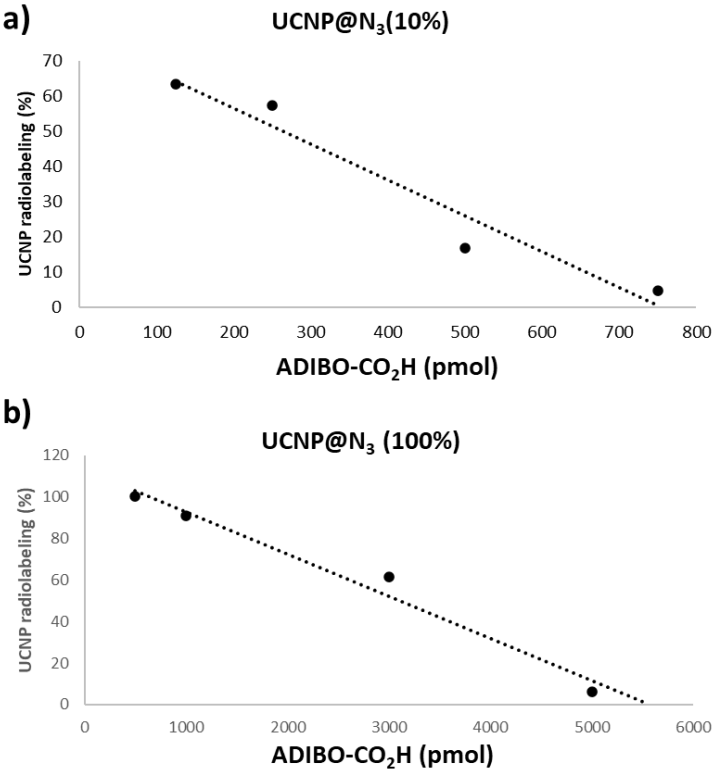


Figure S9. Calibration curves obtained after UCNP@$N_{3(10\%)}$ (**a**) and UCNP@$N_{3(100\%)}$ (**b**) (100 μg) conjugation with increasing concentration of ADIBO-CO_2H **S33** (125-750 and 500-5000 pmol, respectively) followed by radiolabelling with a fixed amount of **[^{125}I]IBA-ADIBO** (74 kBq) and ITLC radio-chromatogram counting.

3.7. DLS spectra of UCNP@N_3, UCNP@CO_2H and UCNP@KuE(1-3)

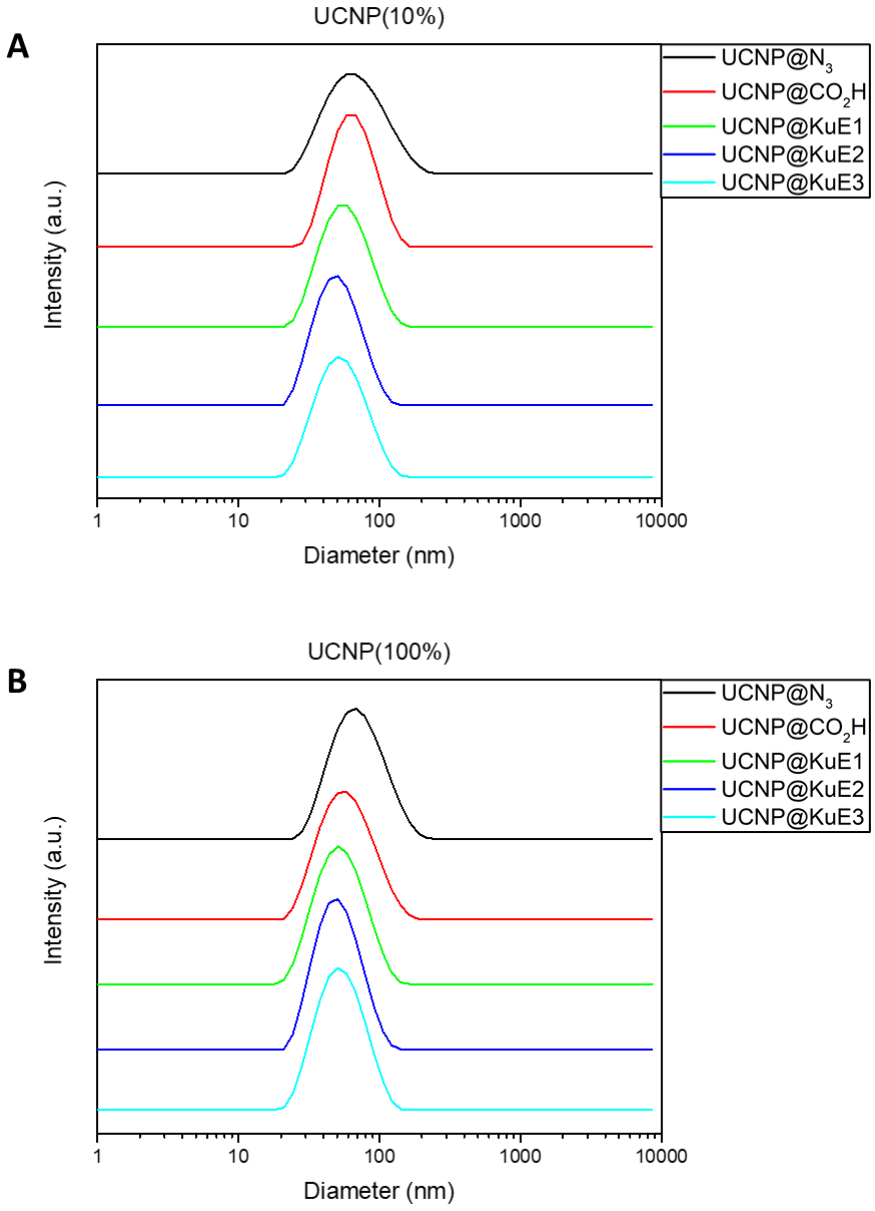


Figure S10. DLS analysis of particle-size distribution of UCNP@$N_{3(10\%)}$ (**A**) and UCNP@$N_{3(100\%)}$ (**B**) before and after SPAAC reaction with **ADIBO-KuE(1-3)** and ADIBO-CO_2H **S33** derivatives. Particle size was analysed using a diluted suspension of UCNP (1 mg/mL) in deionized water.

3.8. Cell survival rates of LNCaP-Luc, PC3-Luc and fibroblast cells after 24 h incubation with UCNP@N_3, UCNP@CO_2H and UCNP@KuE

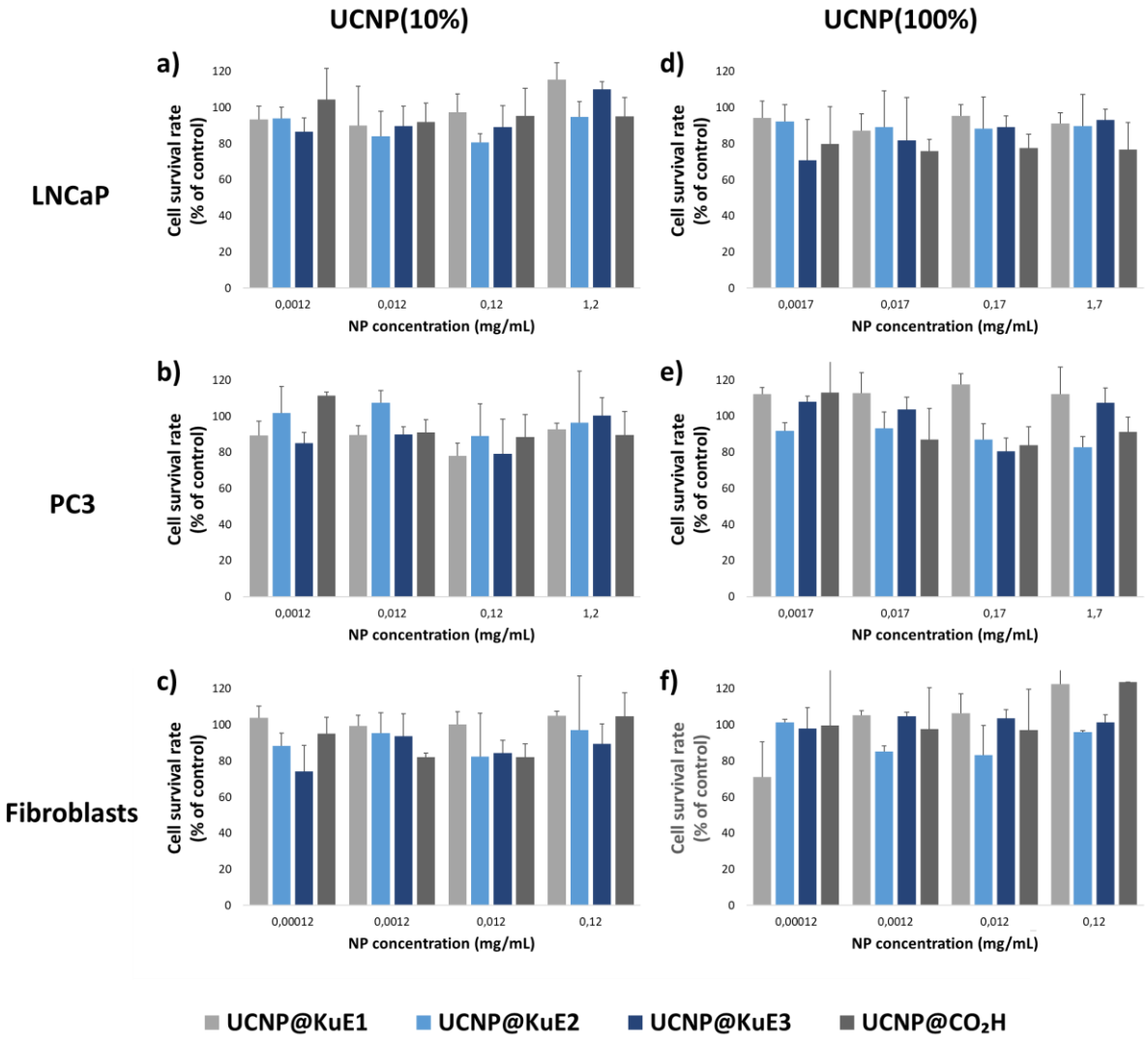


Figure S11. Cell survival rates determined by resazurin assay after 24 h incubation at 37 °C of LNCaP-Luc, PC3-Luc and fibroblast cells in the presence of increasing concentrations of UCNP@KuE(1-3)$_{(10\%)}$ or UCNP@CO2H$_{(10\%)}$ (**a**, **b** and **c**) and UCNP@KuE(1-3)$_{(100\%)}$ or UCNP@CO2H$_{(100\%)}$ (**d**, **e** and **f**). Each experiment was performed in triplicate.

4. Supplementary table

Table S1. Polydispersity index of DLS data in deionized water given in Figure 2.

UCNP@PEG	t = 0	t = 7 d
UCNP@L1	0.134	0.207
UCNP@L2	0.136	0.133
UCNP@L3	0.207	0.309
UCNP@L4	0.226	0.406
UCNP@L5	0.141	0.162
UCNP@L6	1.000	1.000
UCNP@L7	0.169	0.137
UCNP@L8	0.215	0.590
UCNP@L8	0.131	0.086
UCNP@L8	0.172	0.355

Table S2. Biodistribution in LNCaP-Luc xenografted nude mice of [^{125}I]UCNP@$CO_2H_{(10\%)}$ after i.v. injection in the tail vein.

organs	5 min	1 h	3 h	24 h	48 h
Tumour	0.95 ± 0.71	0.60 ± 0.43	0.48 ± 0.34	0.35 ± 0.15	0.49 ± 0.19
Lungs	8.63 ± 1.50	2.72 ± 2.27	0.83 ± 0.53	0.32 ± 0.02	0.44 ± 0.23
Spleen	5.12 ± 0.10	12.8 ± 10.6	13.3 ± 16.4	16.0 ± 0.47	19.9 ± 10.6
Kidneys	5.45 ± 0.97	1.41 ± 1.68	0.98 ± 0.63	0.84 ± 0.11	0.70 ± 0.18
Stomach	1.15 ± 0.51	1.69 ± 1.22	0.46 ± 0.48	0.30 ± 0.13	0.28 ± 0.08
Liver	8.01 ± 1.78	15.2 ± 13.8	12.0 ± 13.3	21.6 ± 2.06	14.5 ± 1.96
Heart	3.92 ± 0.84	1.84 ± 1.32	0.54 ± 0.23	0.19 ± 0.03	0.17 ± 0.05
Small intestin	1.01 ± 0.17	0.93 ± 0.82	0.69 ± 0.68	0.74 ± 0.17	0.66 ± 0.23

Brain	0.79 ± 0.30	0.27 ± 0.23	0.06 ± 0.04	0.02 ± 0.03	0.006 ± 0.001
Thyroid	0.12 ± 0.01	0.11 ± 0.07	0.03 ± 0.02	0.06 ± 0.02	0.06 ± 0.01
Salivary glands	2.01 ± 0.08	0.92 ± 0.66	0.35 ± 0.19	0.21 ± 0.06	0.14 ± 0.01
Muscle	0.38 ± 0.03	0.31 ± 0.23	0.12 ± 0.04	0.10 ± 0.04	0.17 ± 0.18
Prostate	1.56 ± 1.42	1.23 ± 0.65	0.45 ± 0.32	0.19 ± 0.15	0.08 ± 0.07
Bone	1.10 ± 0.23	1.37 ± 1.09	1.08 ± 1.06	1.09 ± 0.30	1.05 ± 0.73
Bladder	1.12 ± 0.51	4.80 ± 4.24	4.92 ± 7.21	1.06 ± 0.62	1.07 ± 0.06
Blood	17.1 ± 1.12	8.42 ± 7.19	1.38 ± 0.70	0.04 ± 0.03	0.04 ± 0.01
Tail	2.19 ± 1.02	11.8 ± 17.1	16.5 ± 13.3	0.97 ± 0.70	1.15 ± 1.42

The mean of all values is given in %ID/g organ except for thyroid (%ID) (n = 3 in one experimentation, mean±SD).

5. References

[1] W. L. F. Armarego and C. L. L. Chai, Purification of Laboratory Chemicals (6th Edition), Elsevier, Oxford, 2009.

[2] N. Francolon, D. Boyer, F. Leccia, E. Jouberton, A. Walter, C. Bordeniau, A. Parat, D. Felder-Flesh, S. Begin-Colin, E. Miot-Noirault, J.-M. Chezal and R. Mahiou, Preparation of core/shell $NaYF_4$:Yb,Tm@dendrons nanoparticles with enhanced upconversion luminescence for in vivo imaging, *Nanomedicine* 2016, **12**(7), 2107-2113.

[3] J. W. Grate, K. F. Mo and M. D. Daily, Triazine-based sequence-defined polymers with side-chain diversity and backbone-backbone interaction motifs, *Angew. Chem. Int. Ed. Engl.*, 2016, **55**(12), 3925-3930.

[4] M. Erdelyi, M. Varedian, C. Skoeld, I. B. Niklasson, J. Nurbo, A. Persson, J. Bergquist and A. Gogoll, Chemistry and folding of photomodulable peptides - stilbene and thioaurone-type candidates for conformational switches, *Org. Biomol. Chem.*, 2008, **6**(23), 4356-4373.

[5] W. Tai, R. Mo, Y. Lu, T. Jiang and Z. Gu, Folding graft polymer with pendant drug segments for co-delivery of anticancer drugs, *Biomaterials*, 2014, **35**(25), 7194-7203.

[6] K. P. Maresca, S. M. Hillier, F. J. Femia, D. Keith, C. Barone, J. L. Joyal, C. N. Zimmerman, A. P. Kozikowski, J. A. Barrett, W. C. Eckelman and J. W. Babich, A series of halogenated heterodimeric inhibitors of prostate specific membrane antigen (PSMA) as radiolabeled probes for targeting prostate cancer, *J. Med. Chem.*, 2009, **52**(2), 347-357.

[7] L. S. Campbell-Verduyn, L. Mirfeizi, A. K. Schoonen, R. A. Dierckx, P. H. Elsinga and B. L. Feringa, Strain promoted copper-free "Click" chemisty for ^{18}F-radiolabeling of bombesin, *Angew. Chem. Int. Ed.*, 2011, **50**(47), 11117-11120.

[8] R. C. Chadwick, S. Van Gyzen, S. Lioger and A. Adronov, Scalable synthesis of strained cyclooctyne derivatives, *Synthesis*, 2014, **46**(5), 669-677.

[9] J. Magano, B. G. Conway, D. Farrand, M. Lovdahl, M. T. Maloney, M. J. Pozzo, J. J. Teixeira, J. Rizzo and D. Tumelty, Scalable and cost-effective synthesis of a linker for bioconjugation with a peptide and a monoclonal antibody, *Synthesis*, 2014, **46**(10), 1399-1406.

[10] A. Kuzmin, A. Poloukhine, M. A. Wolfert and V. V. Popik, Surface functionalization using catalyst-free azide-alkyne cycloaddition, *Bioconjugate Chem.*, 2010, **21**(11), 2076-2085.

[11] C. Benjamin, K. Wright, S. Hyun, K. Krynksi and G. Yu, Non-fouling NTA-PEG-based TEM grid coatings for selective capture of histidine-tagged protein targets from cell lysates, *Langmuir*, 2016, **32**(2), 551-559.

[12] A. Polidori, N. Michel, A. S. Fabiano and B. Pucci, Exotic aqueous behavior of synthetic lipids: formation of vesicular nanotubes. *Chem. Phys. Lipids*, 2005, **136**(1), 23-46.

[13] A. Khalaj, D. Beiki, H. Rafiee and R. Najafi, A new and simple synthesis of *N*-succinimidyl-4-[$^{127/125}$I]iodoenzoate involving a microwave-accelerated iodination step, *J. Labelled Cpd. Radiopharm.*, 2001, **44**(3), 235-240.

[14] Y. Chen, C. A. Foss, Y. Byun, S. Nimmagadda, M. Pullambhatla, J. J. Fox, M. Castanares, S. E. Lupold, J. W. Babich, R. C. Mease and M. G. Pomper, Radiohalogenated prostate-specific membrane Antigen (PSMA)-based ureas as imaging agents for prostate cancer, *J. Med. Chem.*, 2008, **51**(24), 7933–7943.

[15] M. S. Bhojani, R. Ranga, G. D. Luker, A. Rehemtulla, B. D. Ross and M. E. Van Dort, Synthesis and investigation of radioiodinated F3 peptide analog as a SPECT tumor imaging radioligand, *PLoS One*, 2011, **6**(7), e22418.