Electronic Supplemental Information (ESI):

Combination of inverse electron-demand Diels-Alder reaction with highly efficient oxime ligation expands the toolbox of site-selective peptide conjugations
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1 Experimental
1.1 General

1.1.1 Reagents
All reagents were used as supplied by Sigma Aldrich, Alfa Aesar, or Acros Organics without further purification. Amino acids and resins for solid-phase peptide synthesis (SPPS) were purchased from Novabiochem (brand of Merck KGaA, Darmstadt, Germany), CEM (Kamp-Lintfort, Germany) or Iris Biotech (Marktredwitz, Germany).

1.1.2 Mass spectrometry
Electrospray ionization mass spectrometry (ESI-MS) spectra were obtained using a Shimadzu LCMS-2020 mass spectrometer equipped with a Phenomenex Jupiter 5u C4 LC column (50x1 mm, 5 μm, 300 Å). The eluent system consisted of 0.1% (v/v) aq. formic acid (LC-MS grade, Sigma-Aldrich (St. Louis, USA)) (eluent A) and acetonitrile containing 0.1% (v/v) formic acid (LC-MS grade, Karl Roth GmbH, Karlsruhe, Germany) (eluent B).

ESI-HRMS measurements were performed on a maXis Q-TOF mass spectrometer (Bruker Daltonik) equipped with an electrospray ion source. 2 μL solution of the dissolved analytes were injected (autoinjector G1313A, Agilent) and transferred into the ion source together with methanol (flow 0.2 mL/min) provided by a pump (G1311A, Agilent). Electrospray mass spectra were measured in the positive ion mode. Mass calibration using a mixture of calibrants (Agilent) was applied. Compound 31: ESI(+)-HRMS was carried on a FTICR mass spectrometer (APEX IV, Bruker Daltonik). Analyte solution was introduced into the electrospray ion source via a syringe pump. MALDI mass spectra were acquired applying a MALDI-TOF instrument Autoflex (Bruker). 2,5-dihydroxy benzoic acid (DHB) was used as a matrix.
1.1.3 Liquid chromatography

Analytical reversed-phase high performance liquid chromatography (RP-HPLC) was performed on a Varian 920 LC equipped with a Phenomenex Luna Hypersil 5u BDS C₁₈ LC column (5 μ, 130 Å, 150×4.60 mm, 5 μm) at a flow rate of 1 mL/min. For isolation of peptides by semi-preparative RP-HPLC a Varian 940 LC equipped with a preparative C₁₈ column (Phenomenex Luna 5u C₁₈ (250×20 mm; S-4 μm, 8 nm)) was used. At a flow rate of 18 mL/min, 5 min of isocratic flow (starting concentration of eluent B) was followed by 20 min of gradient flow. Eluent A: with 0.1% (v/v) aq. trifluoroacetic acid (TFA), eluent B: 90% (v/v) aq. MeCN with 0.1% (v/v) TFA. Absorption was measured by an UV/VIS detector at 220 nm and 280 nm or 220 nm and 534 nm.

1.2 Synthetic procedures:

1.2.1 Synthesis of compound 1

\[
\begin{align*}
\text{H}_2\text{N} & \quad \text{O} & \quad \text{O} & \quad \text{NH}_2 \quad \overset{10 \text{ eq.}}{\rightarrow} \quad \text{CHCl}_3 \\
\text{O} & \quad \text{O} & \quad \text{O} & \quad \text{O} & \quad \text{O} & \quad \text{O} & \quad \text{H}_2\text{N} & \quad \text{O} & \quad \text{O} & \quad \text{NH}
\end{align*}
\]

To a cooled solution (0°C) of 30.0 g (202.5 mmol) 2,2-(ethylenedioxy)bis(ethylamine) in 200 mL chloroform, a solution of 4.425 g (20.3 mmol) di-tert.-butyl dicarbonate in 200 mL chloroform was drop-wise added under inert atmosphere (N₂). After complete addition, the mixture was allowed to warm to ambient temperature and then stirred overnight. The organic solvent was removed and the colorless liquid was dissolved in 300 mL deionized water. It was extracted three times with each 100 mL methylene chloride. The combined organic phases were washed with 100 mL brine and dried over MgSO₄. The solvent was removed resulting in 4.74 g of turbid, colorless oil (yield: 94.1%).

\(^1\text{H-NMR}\) (300 MHz, DMSO-\(\text{d}_6\)) δ: 1.37 (s, 9H), 2.63 (t, \(J=5.8\) Hz, 2H), 3.05 (q, \(J=6.1\) Hz, 2H), 3.35 (t, \(J=5.7\) Hz, 2H), 3.38 (t, \(J=6.1\) Hz, 2H), 3.49 (s, 4H).

1.2.2 Synthesis of compound 2

Reppe anhydride 2 was synthesized as described in reference 1. 5 g (48 mmol) cyclooctatetraene and 4.708 g (48 mmol) maleic anhydride were converted in 1,2-dichlorobenzene. The crude product had a melting point of 162°C (lit. 165-168°C). It was purified by sublimation on high vacuum resulting in 6.123 g of colorless crystals with a melting point of 167°C (yield: 63%).

\(^1\text{H-NMR}\) (500 MHz, DMSO-\(\text{d}_6\)) δ: 2.81 (s, 2H), 3.04 (s, 2H), 3.25 (s, 2H), 5.89 (s, 2H), 5.97 (dd, \(J=4.6\) Hz, 2H).

\(^{13}\text{C-NMR}\) (125 MHz, DMSO-\(\text{d}_6\)) δ: 36.42, 42.68, 43.71, 128.90, 138.10, 173.72.
1.2.3 Synthesis of compound 3

![Chemical structure of compound 3]

To obtain compound 3, 0.870 g (1.1 eq., 3.54 mmol) Boc-PEG 1 were reacted with 0.651 g (1 eq., 3.22 mmol) Reppe anhydride 2 in 80 mL MeOH. The reaction mixture was heated for 3 h at 80°C under reflux. As the TLC analysis verified the completion of the reaction, the solution was cooled down and methanol was evaporated under reduced pressure. The crude product was dissolved in water and extracted twice with each 30 mL diethyl ether. The organic phase was dried over MgSO₄ and the solvent was removed. 1.1 g of compound 3 were obtained (yield: 82%).

¹H-NMR (300 MHz, CDCl₃) δ: 1.376 (s, 9H), 2.715 (s, 2H), 2.744 (s, 2H), 3.092 (m, 2H), 3.242 (t, J=5.6 Hz, 2H), 3.443 (m, 2H), 3.489 (m, 2H), 3.549 (m, 2H), 5.804 (m, 4H), 5.818 (s, 2H).

ESI-MS calc. for C₂₃H₃₂N₂O₆ 432.23; meas.: 433.4 [M+H]⁺.

1.2.4 Synthesis of compound 4

![Chemical structure of compound 4]

Synthesis of compound 4 was performed as described in reference 2. 15 g (80.7 mmol) iodoacetic acid were dissolved in 30 mL deionized water and 5 mL aq. NaOH (40% (w/w)) were slowly added at 0°C. The solution was allowed to warm to ambient temperature. 9.56 g (92.8 mmol) ethyl-acetohydroxamate were added drop-wise to the stirring solution followed by the addition of 7.5 mL aq. NaOH (40% (w/w)) and 20 mL deionized water. The mixture was heated to 80°C for four hours. During this time, the pH was retained above pH 12 using aq. NaOH (40% (w/w)). The yellow solution was cooled down to ambient temperature and 100 mL deionized water were added. The solution was washed twice with each 50 mL methylene chloride. The pH of the solution was adjusted to pH 2.0 using 1 M aq. hydrochloric acid. The solution was extracted four times with each 50 mL methylene chloride. Between the extraction steps, the pH was adjusted to pH 2.0 using 1 M aq. hydrochloric acid. The combined organic phases were washed with 50 mL brine and dried over CaSO₄. Evaporation of the solvent resulted in 6.98 g colorless oil (yield: 53.8%). This compound is the precursor for compound 4.

¹H-NMR (300 MHz, DMSO-d₆) δ: 1.26 (t, J=6.8 Hz, 3H), 2.01 (s, 3H), 4.00 (q, J=7.0 Hz, 2H), 4.49 (s, 2H).

¹³C-NMR (75 MHz, DMSO-d₆) δ: 13.95, 14.21, 62.68, 70.11, 164.73, 175.45.
The colorless oil (6.98 g, 43.2 mmol) and 4.973 g (43.2 mmol) N-hydroxysuccinimide were dissolved in a mixture containing 100 mL dioxane and 120 mL AcOEt. The solution was cooled to 0°C and 6.69 mL (5.452 g, 43.2 mmol) N,N′-diisopropylcarbodiimide (DIC) were added drop-wise. After complete addition the cooled mixture was stirred for additional 30 min and was afterwards allowed to warm to ambient temperature and stirred overnight. The white precipitate was separated by filtration and the solvent was reduced to a volume of 50 mL in vacuo. The colorless precipitate was removed by filtration and the remaining solvent was removed. The colorless oil was dissolved in 200 mL chloroform and washed three times with each 40 mL aq. NaHCO₃ (5% (w/w)) and three times with each 40 mL brine. The organic phase was dried over MgSO₄. After removing the solvent a colorless oil containing a white precipitate was obtained. The precipitate was removed by filtration using a syringe equipped with a Teflon filter. 8.421 g colorless oil were obtained which crystallized quickly upon freezing at -20°C (yield: 75.5%). Compound 4 should be stored sealed below -20°C.

1H-NMR (300 MHz, DMSO-d₆) δ: 1.26 (t, J=7.0 Hz, 3H), 2.01 (s, 3H), 2.84 (s, 4H), 4.02 (q, J=7.1 Hz, 2H), 4.78 (s, 2H).

13C-NMR (75 MHz, DMSO-d₆) δ: 13.89, 14.23, 25.57, 62.76, 68.51, 164.77, 165.70, 168.74.

ESI-MS calc. for C₁₀H₁₄N₂O₆ m/z: 258.09 meas. 259.05 [M+H]+.

1.2.5 Synthesis of compound 5

![Diagram of synthetic route]

The deprotection of the Boc group was conducted in 20% TFA in chloroform for 1 h at ambient temperature. As LC-MS analysis confirmed the complete Boc elimination, TFA was removed under reduced pressure. 0.8 g (1.0 eq., 1.85 mmol) of the deprotected Reppe-PEG 3 were reacted with 1.01 g (2.0 eq., 3.7 mmol) NHS activated aminooxy building block 4 in 30 mL dichloromethane for 16 h at ambient temperature. After complete reaction dichloromethane was removed under reduced pressure and the crude product was dissolved in deionized water. The aqueous solution was extracted thrice with chloroform. The organic phase was dried over MgSO₄ and the solvent evaporated in vacuo. Final purification was done by flash column chromatography, applying an isocratic DCM:MeOH (9.5:0.5) mixture giving 0.167 g light yellow oil (yield: 19%).

1H-NMR (300 MHz, CDCl₃) δ: 1.196 (t, J=7.1 Hz, 3H), 1.918 (s, 3H), 2.713 (s, 2H), 2.745 (s, 2H), 3.087 (s, 2H), 3.484 (m, 12H), 3.912 (q, J=7.0 Hz, 2H), 4.299 (s, 2H), 5.799 (q, J=4.6 Hz 2 H), 5.819 (s, 2 H).

ESI-MS calc. for C₂₄H₃₃N₃O₇ m/z: 475.24 meas. 476.45 [M+H]+.
1.2.6 Synthesis of compound 6

![Synthesis reaction diagram]

Compound 6 was synthesized according to reference 3. 5 g (47.57 mmol) 2-pyrimidinecarbonitrile and 7 g (47.57 mmol) 4-cyanobenzoic acid were employed as described in reference 3. 2.915 g of a yellow solid were obtained (yield: 21.7%). This substance is the precursor for compound 6.

$^1$H-NMR (300 MHz, DMSO-$d_6$) δ: 7.61 (t, $J$=4.9 Hz, 1H), 7.93 (d, $J$=9.6 Hz, 2H), 8.00 (d, $J$=8.4 Hz, 2H), 8.83 (s, 1H), 8.92 (d, $J$=4.8 Hz, 2H), 9.52 (s, 1H).

$^{13}$C-NMR (75 MHz, DMSO-$d_6$) δ: 122.09, 126.27, 129.41, 132.20, 133.65, 145.62, 146.64, 155.89, 157.67, 166.72.

2.8 g (9.9 mmol) of the yellow solid were oxidized as described in reference 3. 2.37 g of purple solid 6 were obtained (yield: 85.5%).

$^1$H-NMR (300 MHz, DMSO-$d_6$) δ: 7.83 (t, $J$=4.4 Hz, 1H), 8.24 (d, $J$=8.5 Hz, 2H), 8.68 (d, $J$=8.5 Hz, 2H), 9.17 (d, $J$=4.8 Hz, 2H)

$^{13}$C-NMR (75 MHz, DMSO-$d_6$) δ: 123.00, 128.31, 130.23, 134.63, 135.25, 158.50, 159.01, 162.89, 163.16, 166.68.

1.2.7 Synthesis of compound 7

![Synthesis reaction diagram]

0.62 g (2.08 mmol) of compound 6 were suspended in 10 mL thionyl chloride and 10 drops of DMF were added. The setup was equipped with a dry pipe filled with MgSO$_4$. The mixture was stirred for three hours at 90°C. Afterwards the mixture was allowed to cool to ambient temperature and the excess of thionyl chloride was removed in vacuo.

The residue was dissolved in 100 mL chloroform and 1.445 mL (1.072 g, 8.3 mmol) $N,N'$-diisopropylethylamine (DIEA) were added. The mixture was cooled to 0°C. 1.031 g
(4.16 mmol) of compound 1 were dissolved in 10 mL chloroform and the solution was added drop-wise. The setup was equipped with a dry pipe filled with MgSO₄ and the mixture was stirred overnight at ambient temperature. The reaction mixture was washed five times with each 30 mL deionized water and the organic phase was dried over MgSO₄. After removing the solvent the purple residue was purified by flash column chromatography using the solvent mixture EtOH:DCM 1:9 (v:v) (Rf=0.44). 0.452 g purple solid were obtained (yield: 42.6%).

1H-NMR (500 MHz, CDCl₃) δ: 1.35 (s, 9H), 3.25 (d, J=4.2 Hz, 2H), 3.50 (s, 2H), 3.59 (s, 2H), 3.60 (s, 2H), 3.66 (s, 4H), 4.94 (s, 1H), 6.94 (s, 1H), 7.54 (t, J=4.9 Hz, 1H), 7.99 (d, J=9.4 Hz, 2H), 8.73 (d, J=5.0 Hz, 2H), 9.07 (d, J=4.8 Hz, 2H).

13C-NMR (125 MHz, CDCl₃) δ: 28.40, 39.97, 40.28, 70.34, 122.58, 128.05, 128.92, 133.88, 138.79, 158.45, 159.46, 163.18, 164.04, 166.51.

ESI-MS calc. for C₂₄H₃₀N₆O₅ m/z: 510.24 meas. 511.45 [M+H]+.

1.2.8 Synthesis of compound 8

0.452 g (0.885 mmol) of compound 7 were dissolved in 5 mL 20% TFA (v:v) in chloroform. After two hours at ambient temperature, complete deprotection of compound 7 was confirmed by thin-layer chromatography (TLC). The solvent was removed and a purple oil containing residual TFA was obtained. To remove remaining TFA, the oil was dissolved in a mixture of methanol and cyclohexane and the solvent was removed in vacuo. After several steps, the remaining oil was dissolved in water and dried by lyophilization. The purple solid was dissolved in 60 mL DMF and 0.462 mL (0.343 g, 2.66 mmol) DIEA were added. 0.457 g (1.77 mmol) of compound 4 were dissolved in 10 mL DMF and were added drop-wise to the mixture. After complete addition, the mixture was stirred for additional three hours at ambient temperature. The solvent was removed in high vacuum and the excess of DIEA was removed thoroughly. The remaining purple oil was dissolved in 70 mL deionized water and the aqueous phase was extracted twice with each 30 mL chloroform. The solvent was removed and 0.584 g of a purple solid were obtained which was purified by flash column chromatography using the solvent mixture DCM:MeOH (9:5:1 (v:v)) (Rf=0.59). 0.302 g of yellow solid were obtained (yield: 61.2%).

1H-NMR (500 MHz, CDCl₃) δ: 1.18 (t, J=7.2 Hz, 3H), 1.90 (s, 3H), 3.44 (q, J=5.0 Hz, 2H), 3.53 (t, J=5.0 Hz, 2H), 3.59 (m, 4H), 3.65 (m, 4H), 3.89 (q, J=7.2 Hz, 2H), 4.28 (s, 2H), 6.64 (s, 1H), 6.99 (s, 1H), 7.54 (t, J=4.8 Hz, 1H), 8.00 (d, J=8.5 Hz, 2H), 8.74 (d, J=8.5 Hz, 2H), 9.08 (d, J=4.8 Hz, 2H)
13C-NMR (125 MHz, CDCl₃) δ: 13.85, 14.28, 38.59, 39.97, 62.69, 69.80, 69.91, 70.24, 70.36, 72.85, 122.60, 128.07, 128.91, 133.88, 138.74, 158.45, 159.44, 163.19, 164.02, 166.51, 170.50.

RP-HPLC, 10→100% B, tᵣ=12.62 min.

ESI-MS calc. for C₂₅H₃₁N₉O₆ m/z: 553.25 meas. 554.46 [M+H]+.

Deprotection of compound 8 towards 8a:

For deprotection of compound 8 it was dissolved in 20% (v:v) TFA in chloroform and stirred for 120 min at ambient temperature (the deprotected compound forms a second phase). The solvent was removed and residual TFA removed by dissolving the purple oil in water followed by lyophilization.

RP-HPLC, 10→100% B, tᵣ=9.11 min.

ESI-MS calc. for C₂₁H₂₅N₉O₅ m/z: 483.20 meas. 484.45 [M+H]+.

1.2.9 Microwave-assisted solid-phase peptide synthesis (SPPS) of peptides 9-14

Microwave-assisted Fmoc-SPPS was performed on a CEM liberty® peptide synthesizer equipped with a CEM discover® SPS microwave (CEM GmbH) using DMF as solvent. As solid supports 2-chlorotrityl resin from Iris Biotech GmbH (peptides 9, 10, 12, and 13) and AmphiSphere™ 40 RAM resin from Varian Inc. (peptides 11 and 14) were used. Triple coupling of the amino acids and double deprotection was performed upon microwave assistance. For all amino acids except arginine, coupling was performed at 30 W and 50°C for 15 min. Coupling of arginine was performed at ambient temperature for 60 min. Deprotection was performed at 30 W at 50°C for 5 min). Each amino acid was attached using 4 eq. of the respective Fmoc-protected amino acid, 3.9 eq. of O-(benzotriazol-1-yl)-N,N,N′,N′-tetramethyluronium hexafluorophosphate (HBTU) (Iris Biotech GmbH), and 8 eq. base (DIEA for all amino acids except cysteine; for coupling of Fmoc-S-trityl-L-cysteine, collidine (Sigma-Aldrich) was used).

1.2.10 Peptide cleavage and workup

All peptides were cleaved from the resin using 92% (v:v) trifluoroacetic acid (TFA), 2% (v:v) H₂O, 4% (v:v) triethylsilane (TES), and 2% (v:v) anisole. In case of cysteine-containing
peptides, a spatula tip of dithiothreitol (DTT) was added to the cleavage cocktail. After shaking 2 h at room temperature, the solution was filtered and injected into ice-cold diethyl ether. The mixture was cooled down to -20°C for one hour and the precipitate was separated by centrifugation. The supernatant was discharged and the precipitate resuspended in ice-cold diethyl ether. The procedure was repeated additionally two times and the precipitate was dried in a desiccator. The obtained solid was dissolved in 10% (v:v) aqueous MeCN and the solvent was removed by lyophilization resulting in a fluffy consistency of the solid.

1.2.11 Oxidation of N-terminal serine residues

Cysteine-free crude products were oxidized using 10 eq. of sodium periodate in PBS buffer for 10 min. The reaction product was isolated by RP-HPLC using a Varian 940-LC equipped with a YMC Europe GmbH C\textsubscript{18} column (250 × 20mm; S- 4 µm, 8 nm). Thereby, aldehyde-bearing peptides 9, 12, and 13 were obtained. For the synthesis of aldehyde-bearing folded peptides 10, 11 and 14, oxidative folding was followed by the above-described periodate oxidation procedure and by RP-HPLC purification.\textsuperscript{[4,5]} The structures of all synthesized peptides are given in scheme ESI-2.

![Scheme ESI-1: Mechanism of periodate oxidation of N-terminal serine residues.](image)

**Scheme ESI-1:** Mechanism of periodate oxidation of N-terminal serine residues.

<table>
<thead>
<tr>
<th>Compound</th>
<th>HPLC gradient</th>
<th>( t_R ) [min]</th>
<th>Formula and calculated molar mass</th>
<th>ESI-MS measured</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>9</td>
<td>10→100 B</td>
<td>13.22</td>
<td>( \text{C}<em>{48}\text{H}</em>{76}\text{N}<em>{18}\text{O}</em>{15} ) 1116.57</td>
<td>1117.8 [M+H]\textsuperscript{+}, 559.5 [M+2H]\textsuperscript{2+}, 373.2 [M+3H]\textsuperscript{3+}.</td>
<td>43%</td>
</tr>
</tbody>
</table>

**Scheme ESI-2:** Bioactive peptides 9-14 equipped with aldehyde functionality.

**Table ESI-1:** Analytical data of compounds 9-14.
1.2.12 General procedure for oxime ligation of inverse electron-demand Diels-Alder building blocks 5 and 8 to peptides 9-14

Each 1 eq. of the aldehyde-bearing peptides 9-14 and varying equivalents of the aminooxy-bearing inverse electron-demand Diels-Alder building blocks 5 and 8 were mixed in 50% (v:v) aq. TFA and the mixture was shaken overnight at ambient temperature. The TFA was removed under reduced pressure and the aqueous solution was purified by semi-preparative HPLC. Reaction details are listed in table ESI-1 and table ESI-2. The structures of peptides 15-20 equipped with inverse electron-demand Diels-Alder functional groups are shown in scheme ESI-3.

**Table ESI-2**: reaction details of the synthesis of bioactive peptides 15-17 equipped with inverse electron-demand Diels-Alder functionality.

<table>
<thead>
<tr>
<th>Synthesis of</th>
<th>Peptide no.</th>
<th># mg peptide</th>
<th># mmol peptide</th>
<th># equiv. of 8</th>
<th># mg of 8</th>
<th># mmol of 8</th>
<th>Yield [mg]</th>
<th>Yield [%]</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>9</td>
<td>2.0</td>
<td>0.0018</td>
<td>5.0</td>
<td>5.0</td>
<td>0.0090</td>
<td>1.4</td>
<td>49.4</td>
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<tr>
<td>16</td>
<td>10</td>
<td>10.0</td>
<td>0.0059</td>
<td>5.0</td>
<td>16.4</td>
<td>0.0030</td>
<td>1.7</td>
<td>13.3</td>
</tr>
<tr>
<td>17</td>
<td>11</td>
<td>13.0</td>
<td>0.0040</td>
<td>5.0</td>
<td>10.9</td>
<td>0.0198</td>
<td>2.0</td>
<td>13.6</td>
</tr>
</tbody>
</table>

**Table ESI-3**: reaction details of the synthesis of bioactive peptides 18-20 equipped with inverse-electron-demand Diels-Alder functionality.

<table>
<thead>
<tr>
<th>Synthesis of</th>
<th>Peptide no.</th>
<th># mg peptide</th>
<th># mmol peptide</th>
<th># equiv. of 5</th>
<th># mg of 5</th>
<th># mmol of 5</th>
<th>Yield [mg]</th>
<th>Yield [%]</th>
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<tbody>
<tr>
<td>18</td>
<td>12</td>
<td>19.0</td>
<td>0.0278</td>
<td>1.2</td>
<td>15.4</td>
<td>0.0334</td>
<td>3.0</td>
<td>10.1</td>
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<td>19</td>
<td>13</td>
<td>10.0</td>
<td>0.0092</td>
<td>1.2</td>
<td>5.1</td>
<td>0.0110</td>
<td>1.4</td>
<td>10.3</td>
</tr>
<tr>
<td>20</td>
<td>14</td>
<td>8.0</td>
<td>0.0024</td>
<td>1.5</td>
<td>1.7</td>
<td>0.0036</td>
<td>0.8</td>
<td>8.9</td>
</tr>
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Scheme ESI-3: Inverse electron-demand Diels-Alder building blocks grafted to diverse bioactive peptides.

Table ESI-4: Analytical data of compounds 15-20.

<table>
<thead>
<tr>
<th>Compound</th>
<th>HPLC gradient</th>
<th>t_R [min]</th>
<th>Formula and mass calculated</th>
<th>MS type</th>
<th>MS measured</th>
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<td>15</td>
<td>10→100 B</td>
<td>12.98</td>
<td>C_{69}H_{99}N_{25}O_{19} 1581.7549</td>
<td>ESI-HR-MS</td>
<td>791.8851 [M+2H]^{2+}, 528.2598 [M+3H]^{3+}.</td>
</tr>
<tr>
<td>16</td>
<td>10→100 B</td>
<td>12.20</td>
<td>C_{91}H_{137}N_{33}O_{25}S_{2} 2155.9905</td>
<td>ESI-HR-MS</td>
<td>1079.0016 [M+2H]^{2+}, 719.6713 [M+3H]^{3+}, 540.0067 [M+4H]^{4+}.</td>
</tr>
<tr>
<td>17</td>
<td>10→100 B</td>
<td>14.67</td>
<td>C_{155}H_{229}N_{53}O_{44}S_{6} 3728.5635</td>
<td>ESI-HR-MS</td>
<td>1243.8574 [M+3H]^{3+}, 933.1459 [M+4H]^{4+}.</td>
</tr>
<tr>
<td>19</td>
<td>10→100 B</td>
<td>16.72</td>
<td>C_{71}H_{103}N_{15}O_{16} 1453.7606</td>
<td>ESI-HR-MS</td>
<td>1454.7668 [M+H]^{+}, 727.8883 [M+2H]^{2+}.</td>
</tr>
<tr>
<td>20</td>
<td>10→100 B</td>
<td>15.07</td>
<td>C_{157}H_{238}N_{28}O_{47}S_{6} 3737.5877</td>
<td>ESI-HR-MS</td>
<td>1246.8713 [M+3H]^{3+}, 935.4055 [M+4H]^{4+}.</td>
</tr>
</tbody>
</table>

1.2.13 Synthesis of compound 30

0.51 g (4.94 mmol) 4-aminobutyric acid and 0.5 g (2.47 mmol) of compound 2 were suspended in 10 mL methanol. The mixture was stirred for two hours at 100°C. After cooling down to ambient temperature, the solvent was removed in vacuo. The colorless solid was
purified by flash column chromatography by using a solvent mixture of toluene and methanol in a ratio of 9:1 (v:v) ($R_f=0.43$). 472.1 mg of a colorless solid were obtained (yield: 66.6%).

$^1$H-NMR (300 MHz, DMSO-$d_6$) δ: 1.60 (q, $J=7.1$ Hz, 2H), 2.13 (t, $J=7.5$ Hz, 2H), 2.80 (s, 2H), 2.85 (s, 2H), 2.98 (s, 2H), 3.31 (t, $J=6.7$ Hz, 2H), 5.80 (d, $J=4.5$ Hz, 2H), 5.88 (d, $J=4.5$ Hz, 2H).

$^{13}$C-NMR (75 MHz, DMSO-$d_6$) δ: 22.61, 30.80, 36.21, 37.06, 42.62, 43.58, 128.03, 137.93, 173.73, 178.50.

1.2.14 Synthesis of compound 31

![Synthesis of compound 31](image)

0.1 g (0.348 mmol) of compound 30 were dissolved in 5 mL dry acetonitrile and the solution was cooled to 0°C. 0.121 mL (0.09 g, 0.696 mmol) DIEA were added to the cooled solution. 0.093 g (0.452 mmol) dicyclohexylcarbodiimide (DCC) and 0.06 g (0.522 mmol) $N$-hydroxysuccinimide (NHS) were dissolved in 3 mL acetonitrile and were added drop-wise to the cooled solution. After complete addition, the mixture was stirred for one hour on ice and afterwards stirred at ambient temperature overnight. The colorless precipitate was separated by filtration. The solvent was removed in vacuo and the colorless solid dissolved in 40 mL dry DMSO. 1.225 g (1.044 mmol) of octammonium POSS (Hybrid Plastics) were dissolved in 10 mL dry DMSO. To this vigorously stirred solution, activated compound 30 was added drop-wise at ambient temperature and after complete addition, the mixture was stirred overnight. The solvent was removed by lyophilization and the colorless oil was purified by semi-preparative RP-HPLC (gradient: 10→100). The solvent was removed by lyophilization and 0.113 g of a white solid were obtained (yield: 28.3%).

RP-HPLC, 10→100% B, $t_R=12.64$ min.

ESI-HR-MS calc. for $C_{40}H_{79}N_9O_{15}Si_8$ 1149.3850 meas. 575.7000 [M+3H]$^3^+$, 384.1356 [M+4H]$^4^+$.

1.2.15 General procedure for inverse electron-demand Diels-Alder reactions of peptides 15-20

Inverse electron-demand Diels-Alder functional groups grafted peptides 15 to 20 were dissolved in 10% aq. acetonitrile containing 0.1% TFA at a concentration of 10 mg/mL (w:v). Equimolar amounts were mixed according to table ESI-5 and were shaken at ambient temperature overnight. The reaction is schematically shown in scheme ESI-4. The reaction mixtures were analyzed by RP-HPLC and the turnover was determined by comparing the area of the absorption at 220 nm corresponding to the peptides 15 to 20 before and after the reaction. The HPLC-traces of the reactions are shown in figure ESI-1 and ESI-2. The
reaction products of the Diels-Alder reaction with inverse electron-demand (DAR$_{inv}$) reactions are shown in scheme ESI-5.

<table>
<thead>
<tr>
<th>Peptides</th>
<th>15</th>
<th>16</th>
<th>17</th>
</tr>
</thead>
<tbody>
<tr>
<td>18</td>
<td>21</td>
<td>24</td>
<td>27</td>
</tr>
<tr>
<td>19</td>
<td>22</td>
<td>25</td>
<td>28</td>
</tr>
<tr>
<td>20</td>
<td>23</td>
<td>26</td>
<td>29</td>
</tr>
</tbody>
</table>

*Table ESI-5*: Overview of the DAR$_{inv}$ building block bearing bioactive peptides 15-20 and the formed reaction products 21-29.

*Scheme ESI-4*: DAR$_{inv}$ reaction for the synthesis of heterodimers of bioactive peptides.

In the frame of our proof-of-concept study, the products 21-29 were HPLC purified in the amounts required to perform the HR-MS analysis. As we did not plan further activity assays, the fractions from the analytical purification runs were collected and analyzed. Therefore no isolated yields but HPLC conversions are given (Fig. 4B).
**Figure ESI-1**: HPLC traces of DAR<sub>inv</sub> reactions for generation of heterodimers comprising different bioactive peptides. The reaction mixtures were investigated at the beginning and after a reaction time of 12 hours. Absorbance at 220 nm. Reaction of: a) 15 and 18; b) 15 and 19; c) 15 and 20; d) 16 and 18; e) 16 and 19; f) 16 and 20.
Figure ESI-2: HPLC traces of DAR_{inv} reactions for generation of heterodimers comprising different bioactive peptides. The reaction mixtures were investigated at the beginning and after a reaction time of 12 hours. Absorbance at 220 nm, gradient 10→100. Reaction of: g) 18 and 17; h) 17 and 19; i) 17 and 20.

Scheme ESI-5: Structures of DAR_{inv} conjugates 21-29.
### Table ESI-6: Mass spectrometric data of DAR<sub>inv</sub> conjugates 21-29 and 32.

<table>
<thead>
<tr>
<th>compound</th>
<th>Formula and calculated molar mass</th>
<th>MS method</th>
<th>MS measured</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>C&lt;sub&gt;117&lt;/sub&gt;H&lt;sub&gt;167&lt;/sub&gt;N&lt;sub&gt;35&lt;/sub&gt;O&lt;sub&gt;35&lt;/sub&gt; 2622.2364</td>
<td>HR ESI</td>
<td>875.0857 [M+3H]&lt;sup&gt;3+&lt;/sup&gt;, 656.5662 [M+3H]&lt;sup&gt;3+&lt;/sup&gt;</td>
</tr>
<tr>
<td>22</td>
<td>C&lt;sub&gt;140&lt;/sub&gt;H&lt;sub&gt;208&lt;/sub&gt;N&lt;sub&gt;38&lt;/sub&gt;O&lt;sub&gt;37&lt;/sub&gt; 3005.4937</td>
<td>HR ESI</td>
<td>1002.8383 [M+3H]&lt;sup&gt;3+&lt;/sup&gt;, 752.3804 [M+4H]&lt;sup&gt;4+&lt;/sup&gt;, 602.1064 [M+5H]&lt;sup&gt;5+&lt;/sup&gt;</td>
</tr>
<tr>
<td>23</td>
<td>C&lt;sub&gt;226&lt;/sub&gt;H&lt;sub&gt;333&lt;/sub&gt;N&lt;sub&gt;71&lt;/sub&gt;O&lt;sub&gt;66&lt;/sub&gt;S&lt;sub&gt;6&lt;/sub&gt; 5289.3208</td>
<td>HR ESI</td>
<td>1058.8703 [M+5H]&lt;sup&gt;5+&lt;/sup&gt;, 882.5639 [M+6H]&lt;sup&gt;6+&lt;/sup&gt;</td>
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<td>24</td>
<td>C&lt;sub&gt;139&lt;/sub&gt;H&lt;sub&gt;205&lt;/sub&gt;N&lt;sub&gt;43&lt;/sub&gt;O&lt;sub&gt;41&lt;/sub&gt;S&lt;sub&gt;2&lt;/sub&gt; 3196.4720</td>
<td>HR ESI</td>
<td>800.1251 [M+4H]&lt;sup&gt;4+&lt;/sup&gt;, 640.2998 [M+5H]&lt;sup&gt;5+&lt;/sup&gt;</td>
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<td>C&lt;sub&gt;162&lt;/sub&gt;H&lt;sub&gt;236&lt;/sub&gt;N&lt;sub&gt;46&lt;/sub&gt;O&lt;sub&gt;43&lt;/sub&gt;S&lt;sub&gt;2&lt;/sub&gt; 3579.7292</td>
<td>HR ESI</td>
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<td>C&lt;sub&gt;248&lt;/sub&gt;H&lt;sub&gt;371&lt;/sub&gt;N&lt;sub&gt;79&lt;/sub&gt;O&lt;sub&gt;72&lt;/sub&gt;S&lt;sub&gt;8&lt;/sub&gt; 5863.5564</td>
<td>HR ESI</td>
<td>978.2661 [M+6H]&lt;sup&gt;6+&lt;/sup&gt;, 838.6411 [M+7H]&lt;sup&gt;7+&lt;/sup&gt;, 733.9730 [M+8H]&lt;sup&gt;8+&lt;/sup&gt;</td>
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<tr>
<td>27</td>
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<td>MALDI</td>
<td>4775.3 [M+H]&lt;sup&gt;+&lt;/sup&gt;</td>
</tr>
<tr>
<td>28</td>
<td>C&lt;sub&gt;228&lt;/sub&gt;H&lt;sub&gt;332&lt;/sub&gt;N&lt;sub&gt;66&lt;/sub&gt;O&lt;sub&gt;62&lt;/sub&gt;S&lt;sub&gt;6&lt;/sub&gt; 5154.3179</td>
<td>HR ESI</td>
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<td>MALDI</td>
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<td>32</td>
<td>C&lt;sub&gt;131&lt;/sub&gt;H&lt;sub&gt;214&lt;/sub&gt;N&lt;sub&gt;40&lt;/sub&gt;O&lt;sub&gt;40&lt;/sub&gt;S&lt;sub&gt;2&lt;/sub&gt;Si&lt;sub&gt;8&lt;/sub&gt; 3275.3537</td>
<td>HR ESI</td>
<td>819.8464 [M+4H]&lt;sup&gt;4+&lt;/sup&gt;, 656.0783 [M+5H]&lt;sup&gt;5+&lt;/sup&gt;, 546.8998 [M+6H]&lt;sup&gt;6+&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

#### 1.2.16 Inverse electron-demand Diels-Alder reaction for the synthesis of compound 32

Compound 32 was synthesized using 1.0 mg of 31 (1 eq., 0.87 µmol) and 2.3 mg of 16 (1.2 eq., 1.044 µmol). Both compounds were dissolved in dry DMSO and reacted overnight at ambient temperature. The reaction completion was confirmed via LC-MS.

**ESI-HR-MS** calc. for C<sub>131</sub>H<sub>214</sub>N<sub>40</sub>O<sub>40</sub>S<sub>2</sub>Si<sub>8</sub> 3275.3537 meas. 819.8464 [M+4H]<sup>4+</sup>, 656.0783 [M+5H]<sup>5+</sup>, 546.8998 [M+6H]<sup>6+</sup>.
1.2 Analytical data

1.2.1 Compound 1

Figure ESI-3: $^1$H-NMR spectrum of compound 1.

1.2.2 Compound 2

Figure ESI-4: $^1$H-NMR spectrum of compound 2.
Figure ESI-5: $^{13}$C-NMR spectrum of compound 2.

1.2.3 Compound 3

Figure ESI-6: $^1$H-NMR spectrum of compound 3.
1.2.4 Compound 4

Figure ESI-7: $^1$H-NMR spectrum of the precursor compound 4.

Figure ESI-8: $^{13}$C-NMR spectrum of the precursor of compound 4.
Figure ESI-9: $^1$H-NMR spectrum of compound 4.

Figure ESI-10: $^{13}$C-NMR spectrum of compound 4.
**Figure ESI-11:** ESI-MS spectrum of compound 4; calc. for $C_{10}H_{14}N_{2}O_{6}$ $M$: 258.09 m/z meas.: 259.05 [M+H]$^+$. 

**1.2.5 Compound 5**

**Figure ESI-12:** $^1$H-NMR spectrum of compound 5.
**Figure ESI-13:** $^{13}$C-NMR spectrum of compound 5.

**Figure ESI-14:** ESI-MS spectrum of compound 5; calc. for $C_{24}H_{33}N_3O_7$ M: 475.24, m/z meas.: 476.45 [M+H]$^+$. 
1.2.6 Compound 6

Figure ESI-15: $^1$H-NMR spectrum of the precursor of compound 6.

Figure ESI-16: $^{13}$C-NMR spectrum of the precursor of compound 6.
Figure ESI-17: $^1$H-NMR spectrum of compound 6.

Figure ESI-18: $^{13}$C-NMR spectrum of compound 6.
1.2.7 Compound 7

Figure ESI-19: $^1$H-NMR spectrum of compound 7.

Figure ESI-20: $^{13}$C-NMR spectrum of compound 7.
Figure ESI-21: ESI-MS spectrum of compound 7; calc. for C_{24}H_{30}N_{8}O_{5} M: 510.24, m/z meas.: 511.45 [M+H]^+.

1.2.8 Compound 8

Figure ESI-22: $^1$H-NMR spectrum of compound 8.
Figure ESI-23: $^{13}$C-NMR spectrum of compound 8.

Figure ESI-24: HPLC trace of compound 8. Absorbance at 220 nm; gradient 10→100 B; $t_R$=12.62 min.

Figure ESI-25: HPLC trace of compound 8a. Absorbance at 220 nm; gradient 10→100 B; $t_R$=9.11 min.
1.2.9 Compound 9

**Figure ESI-28:** HPLC trace of compound 9. Absorbance at 220 nm; gradient 10→100 B; t_R=13.22 min.

**Figure ESI-29:** ESI-MS spectrum of compound 9; calc. for C_{48}H_{76}N_{16}O_{16} M: 1116.57, m/z meas.: 1117.8 [M+H]^+, 559.62 meas. 559.5 [M+2H]^2+, 373.41 meas. 373.2 [M+3H]^3+. 

**Figure ESI-26:** ESI-MS spectrum of compound 8; calc. for C_{25}H_{31}N_{9}O_{6} M: 553.25, m/z meas.: 554.46 [M+H]^+.

**Figure ESI-27:** ESI-MS spectrum of compound 8a; calc. for C_{21}H_{25}N_{9}O_{5} M: 483.20, m/z meas.: 484.45 [M+H]^+. 

**Figure ESI-29:** ESI-MS spectrum of compound 9; calc. for C_{48}H_{76}N_{16}O_{16} M: 1116.57, m/z meas.: 1117.8 [M+H]^+, 559.62 meas. 559.5 [M+2H]^2+, 373.41 meas. 373.2 [M+3H]^3+. 

**Figure ESI-29:** ESI-MS spectrum of compound 9; calc. for C_{48}H_{76}N_{16}O_{16} M: 1116.57, m/z meas.: 1117.8 [M+H]^+, 559.62 meas. 559.5 [M+2H]^2+, 373.41 meas. 373.2 [M+3H]^3+. 

**Figure ESI-28:** HPLC trace of compound 9. Absorbance at 220 nm; gradient 10→100 B; t_R=13.22 min.
1.2.10 Compound 10

Figure ESI-30: HPLC trace of compound 10. Left: absorbance at 220 nm; right: absorbance at 220 nm; gradient 10→100 B; t<sub>R</sub>=11.54 min.

Figure ESI-31: ESI-MS spectrum of compound 10; calc. for C<sub>70</sub>H<sub>114</sub>N<sub>24</sub>O<sub>21</sub>S<sub>2</sub> M: 846.98, m/z meas.: 846.9 [M+2H]<sup>2+</sup>, calc. 564.98 meas. 571.0 [M+3H+H<sub>2</sub>O]<sup>3+</sup>, calc. 423.99 meas. 428.5 [M+4H+H<sub>2</sub>O]<sup>4+</sup>.

1.2.11 Compound 11

Figure ESI-32: HPLC trace of compound 11. Left: absorbance at 220 nm; right: absorbance at 280 nm; gradient 10→100 B; t<sub>R</sub>=14.27 min.

Figure ESI-33: ESI-MS spectrum of compound 11; calc. for C<sub>134</sub>H<sub>206</sub>N<sub>44</sub>O<sub>40</sub>S<sub>6</sub> M: 1633.88, m/z meas.: 1642.89 [M+2H+H<sub>2</sub>O]<sup>2+</sup>, calc. 1095.59 meas. 1095.57 [M+3H+H<sub>2</sub>O]<sup>3+</sup>, calc. 821.95 meas. 821.86 [M+4H+H<sub>2</sub>O]<sup>4+</sup>.
1.2.12 Compound 12

**Figure ESI-34**: HPLC trace of compound 12. Left: absorbance at 220 nm; right: absorbance at 280 nm; gradient 10→100 B; \( t_R = 10.76 \text{ min} \).

**Figure ESI-35**: ESI-MS spectrum of compound 12; calc. for \( \text{C}_{28}\text{H}_{45}\text{N}_9\text{O}_{11} \); \( M: 683.32 \), m/z meas.: 684.4 \([\text{M+H}]^+\), 702.4 \([\text{M+H+H}_2\text{O}]^+\).

1.2.13 Compound 13

**Figure ESI-36**: HPLC trace of compound 13. Absorbance at 220 nm; gradient 10→100 B; \( t_R = 15.06 \text{ min} \).
Figure ESI-37: ESI-MS spectrum of compound 13; calc. for C$_{51}$H$_{78}$N$_{12}$O$_{13}$ M: 1067.26, m/z meas.: 1085.87 [M+H+H$_2$O]$^+$, 534.63 meas. 543.55 [M+2H+H$_2$O]$^{2+}$.

1.2.14 Compound 14

Figure ESI-38: HPLC trace of compound 14. Left: absorbance at 220 nm; right: absorbance at 280 nm; gradient 10→100 B; t$_R$=13.56 min.

Figure ESI-39: ESI-MS spectrum of compound 14; calc. for C$_{137}$H$_{211}$N$_{45}$O$_{42}$S$_6$ M: 1677.42, m/z meas.: 1686.39 [M+2H+H$_2$O]$^{2+}$, 1118.61 meas. 1124.67 [M+3H+H$_2$O]$^{3+}$, 839.21 meas. 843.76 [M+4H+H$_2$O]$^{4+}$. 
1.2.15 Compound 15

Figure ESI-40: HPLC trace of compound 15. Absorbance at 220 nm; gradient 10→100 B; $t_R=12.98$ min.

Figure ESI-41: Section of HR ESI-MS spectrum of compound 15. Upper panel: measured isotopic pattern $[M+2H]^2^+$. Lower panel: simulated isotopic pattern $[M+2H]^2^+$. 
1.2.16 Compound 16

**Figure ESI-42:** HPLC trace of compound 16. Absorbance at 220 nm; gradient 10→100 B; \( t_R = 12.20 \text{ min} \).

**Figure ESI-43:** Section of HR ESI-MS spectrum of compound 16. Upper panel: measured isotopic pattern \([\text{M}+3\text{H}]^{3+}\). Lower panel: simulated isotopic pattern \([\text{M}+3\text{H}]^{3+}\).
**1.2.17 Compound 17**

**Figure ESI-44:** HPLC trace of compound 17. Absorbance at 220 nm; gradient 10→100 B; t\(_R\)=14.67 min.

**Figure ESI-45:** Section of HR ESI-MS spectrum of compound 17. Upper panel: measured isotopic pattern [M+3H]\(^{3+}\). Lower panel: simulated isotopic pattern [M+3H]\(^{3+}\).
**1.2.18 Compound 18**

*Figure ESI-46:* HPLC trace of compound 18. Absorbance at 220 nm; gradient 10→100 B; $t_R=14.78$ min.

*Figure ESI-47:* Section of HR ESI-MS spectrum of compound 18. Upper panel: measured isotopic pattern $[M+H]^+$. Lower panel: simulated isotopic pattern $[M+H]^+$. 
1.2.19 Compound 19

**Figure ESI-48**: HPLC trace of compound 19. Absorbance at 220 nm; gradient 10→100 B; $t_R=16.72$ min.

**Figure ESI-49**: Section of HR ESI-MS spectrum of compound 19. Upper panel: measured isotopic pattern [M+2H]$^{2+}$. Lower panel: simulated isotopic pattern [M+2H]$^{2+}$. 
1.2.20 Compound 20

Figure ESI-50: HPLC trace of compound 20. Absorbance at 220 nm; gradient 10→100 B; $t_R=15.07$ min.

Figure ESI-51: Section of HR ESI-MS spectrum of compound 20. Upper panel: measured isotopic pattern [M+3H]$^{3+}$. Lower panel: simulated isotopic pattern [M+3H]$^{3+}$.
1.2.21 Compound 21


1.2.22 Compound 22

Figure ESI-53: Section of HR ESI-MS spectrum of compound 22. Upper panel: measured isotopic pattern [M+3H]3+. Lower panel: simulated isotopic pattern [M+3H]3+. 
1.2.23 Compound 23

Figure ESI-54: Section of HR ESI-MS spectrum of compound 23. Upper panel: measured isotopic pattern [M+6H]$^{6+}$. Lower panel: simulated isotopic pattern [M+6H]$^{6+}$.

1.2.24 Compound 24

Figure ESI-55: Section of HR ESI-MS spectrum of compound 24. Upper panel: measured isotopic pattern [M+4H]$^{4+}$. Lower panel: simulated isotopic pattern [M+4H]$^{4+}$. 
1.2.25 Compound 25

Figure ESI-56: Section of HR ESI-MS spectrum of compound 25. Upper panel: measured isotopic pattern [M+3H]^3+. Lower panel: simulated isotopic pattern [M+3H]^3+.

1.2.26 Compound 26

Figure ESI-57: Section of HR ESI-MS spectrum of compound 26. Upper panel: measured isotopic pattern [M+7H]^7+. Lower panel: simulated isotopic pattern [M+7H]^7+.
1.2.27 Compound 27

**Figure ESI-58:** Section of deconvoluted HR ESI-MS spectrum of compound 27. Upper panel: deconvoluted isotopic pattern measured. Lower panel: calculated isotopic pattern.

**Figure ESI-59:** Fragment of a MALDI-MS spectrum of compound 27. Calc. for $C_{203}H_{299}N_{63}O_{60}S_6$ 4774.4 meas. 4775.3 [M+H]$^+$.
1.2.28 Compound 28

**Figure ESI-60:** Section of HR ESI-MS spectrum of compound 28. Upper panel: measured isotopic pattern [M+5H]^{5+}. Lower panel: simulated isotopic pattern [M+5H]^{5+}.

1.2.29 Compound 29 possessed extremely poor ionization upon mass-spectrometric analysis, combined with strong aggregation tendency. Therefore, although mass peaks corresponding to [M+K^{+}+H^{+}] species were found in MALDI-MS spectrum, it was not used for characterization, and analysis relied on chromatographic methods (see Fig. ESI-2).

1.2.30 Compound 30

**Figure ESI-62:** $^1$H-NMR spectrum of compound 30.
Figure ESI-63: $^{13}$C-NMR spectrum of compound 30.

1.2.31 Compound 31

Figure ESI-64: HPLC trace of compound 31. Absorbance at 220 nm; gradient 10→100 B; $t_R=12.27$ min.
Figure ESI-65: Section of ESI-HR-MS spectrum of compound 31. Left: measured isotopic pattern \([M+3H]^3+\); right: calculated isotopic pattern \([M+3H]^3+\).

1.2.32 Compound 32

Figure ESI-66: Section of HR ESI-MS spectrum of compound 32. Upper panel: measured isotopic pattern \([M+5H]^5+\). Lower panel: simulated isotopic pattern \([M+5H]^5+\).

2 References