

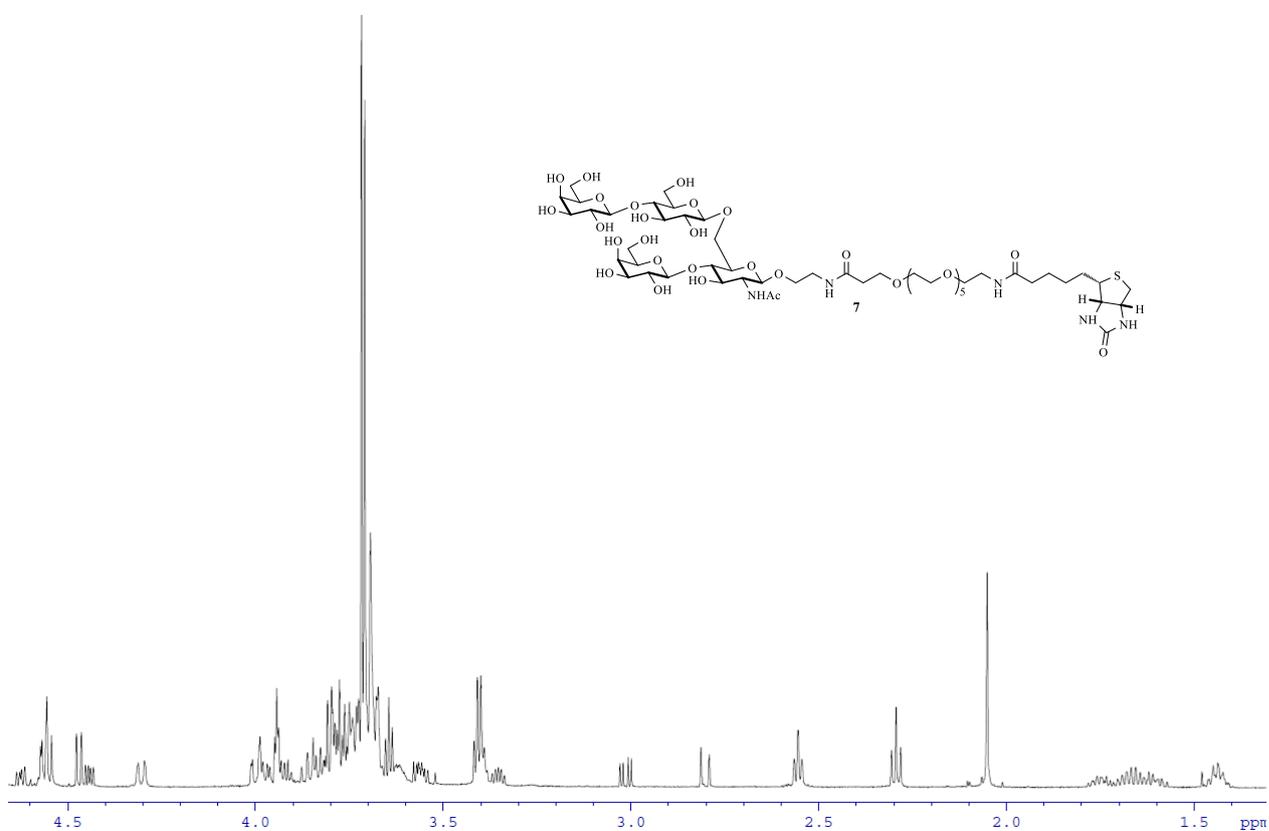
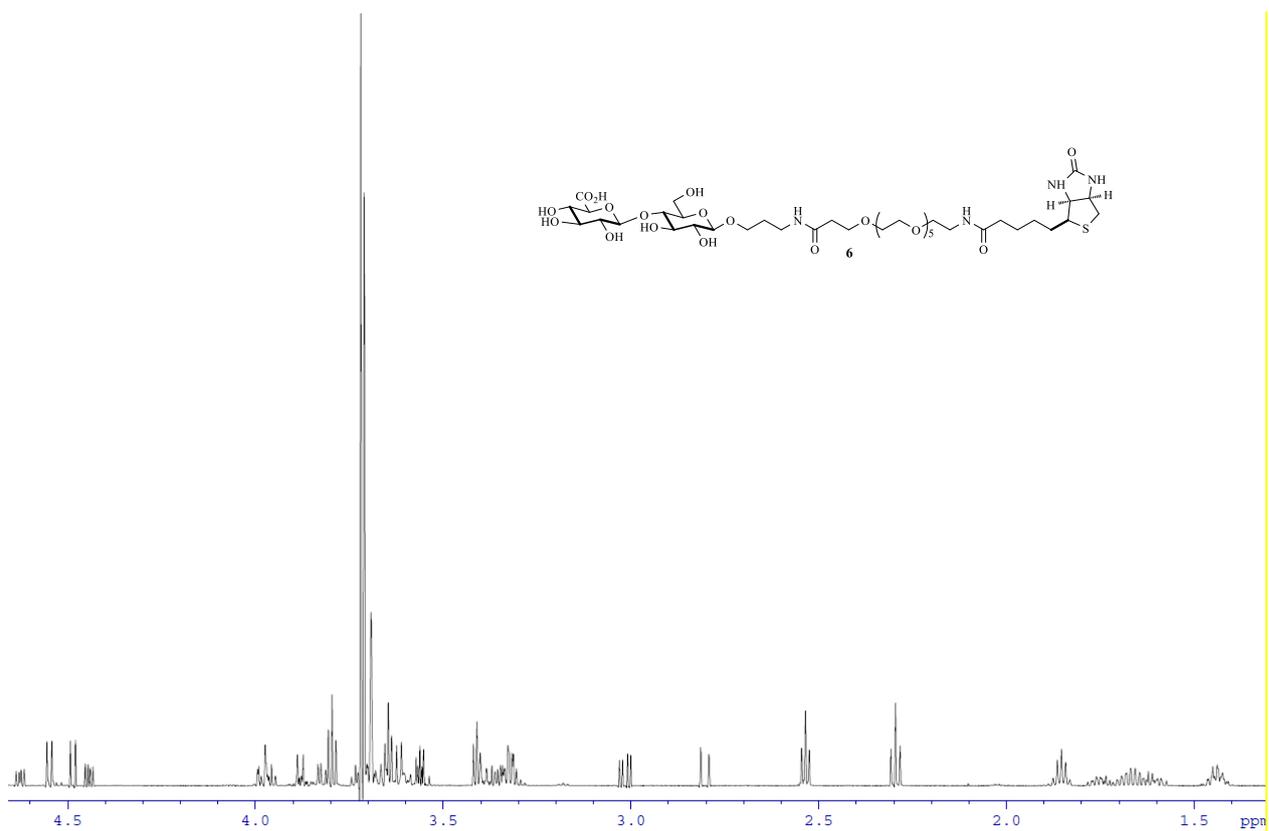
High-resolution mass spectra of biotinylated, HEG-spacered molecular probes with carbohydrate fragments of the capsular polysaccharides from *Streptococcus pneumoniae*

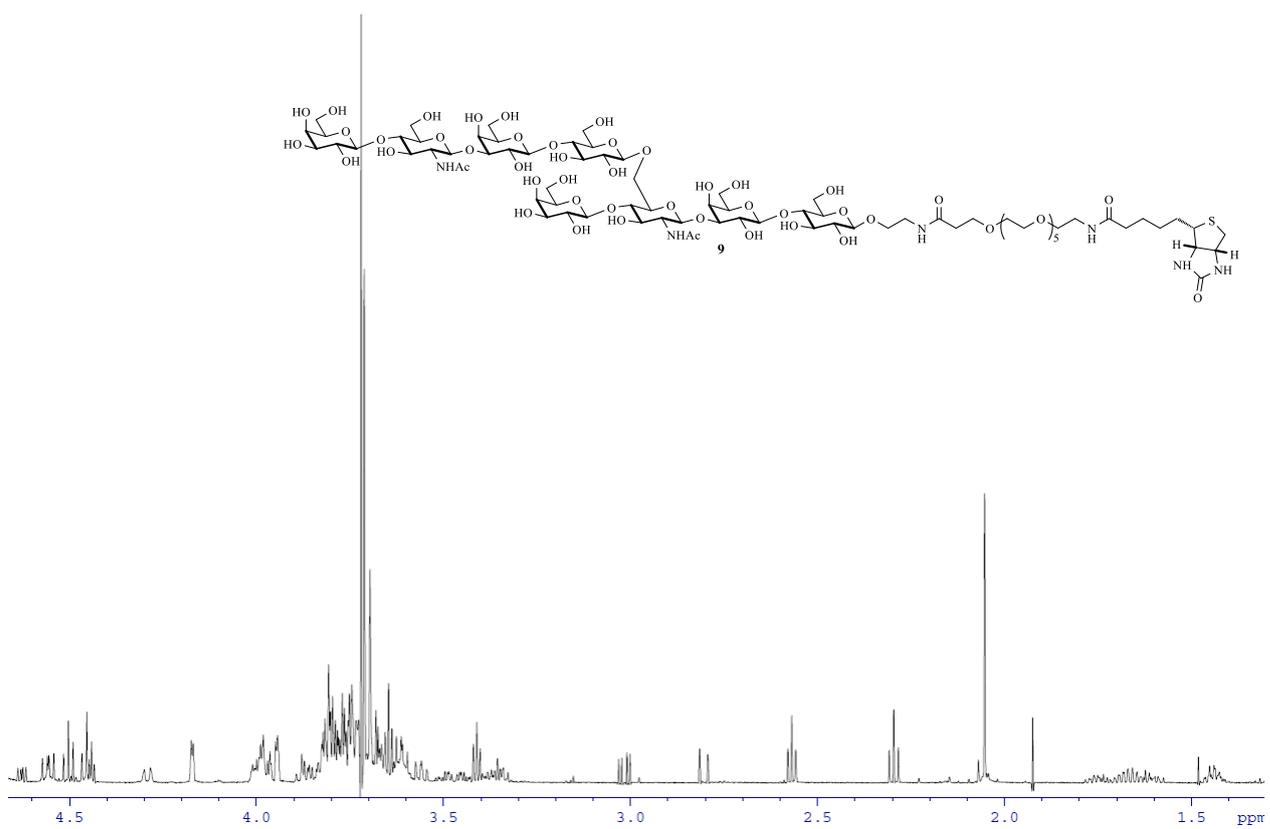
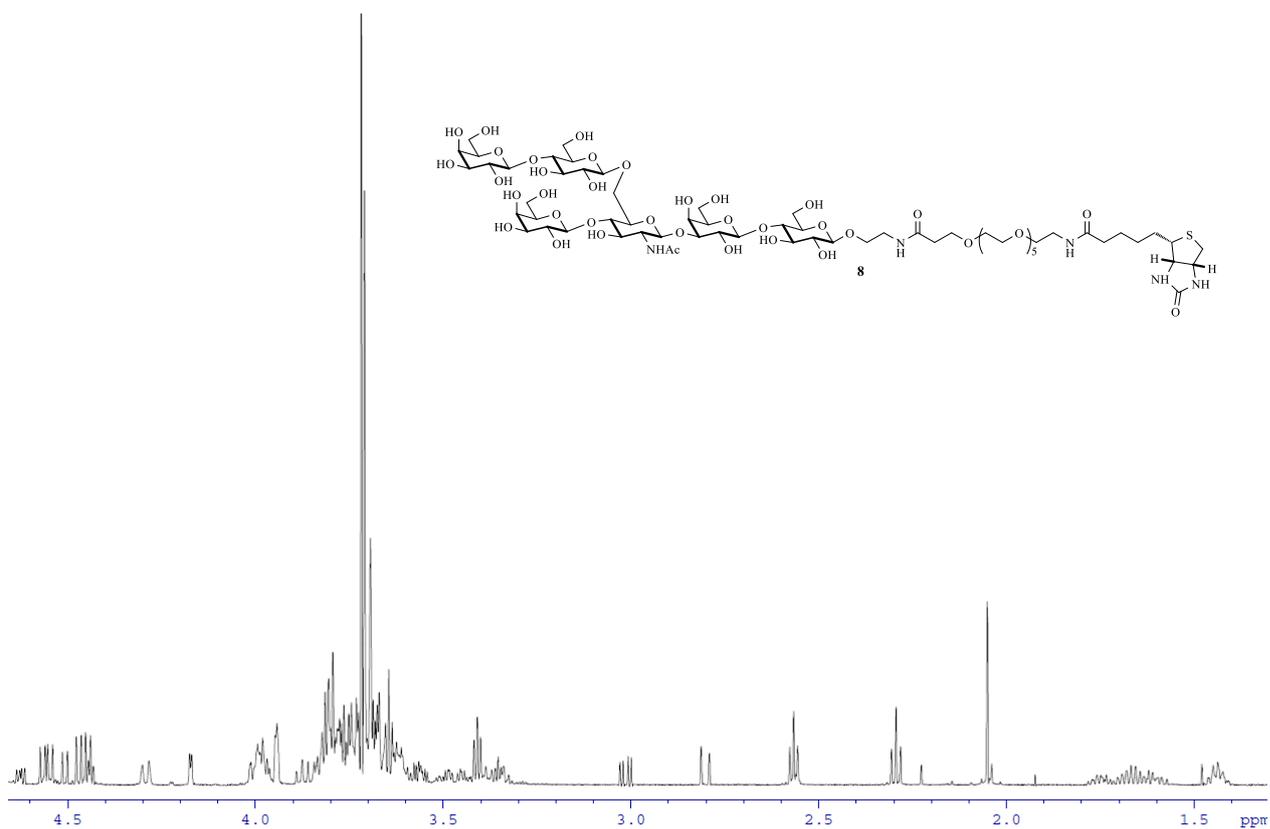
Alexander O. Chizhov, Elena V. Sukhova, Elena A. Khatuntseva, Yury E. Tsvetkov and Nikolay E. Nifantiev

Synthesis of compounds 6-9. General procedure.

An aminoalkyl glycoside (1.4-4.0 μmol) was dissolved in DMF (0.1 ml), then a solution of pentafluorophenyl ester **5** (1.7-4.9 μmol , 1.2 equiv.) in DMF (20-60 μl) and triethylamine (4 μl /1 μmol of **5**) were added. The resulting mixture was stirred at room temperature and monitored by TLC (chloroform – methanol – water 5:5:1). When the reaction was complete (usually after 16-18 h), the solvent was removed in vacuum of an oil pump, and the residue was subjected to gel-chromatography on a Sephadex G-15 column in water. Isolated products were lyophilized to provide biotinylated probes **6-9** (58-81%) as fluffy amorphous solids.

^1H NMR spectra of compounds **6-9** (600 MHz, D_2O).





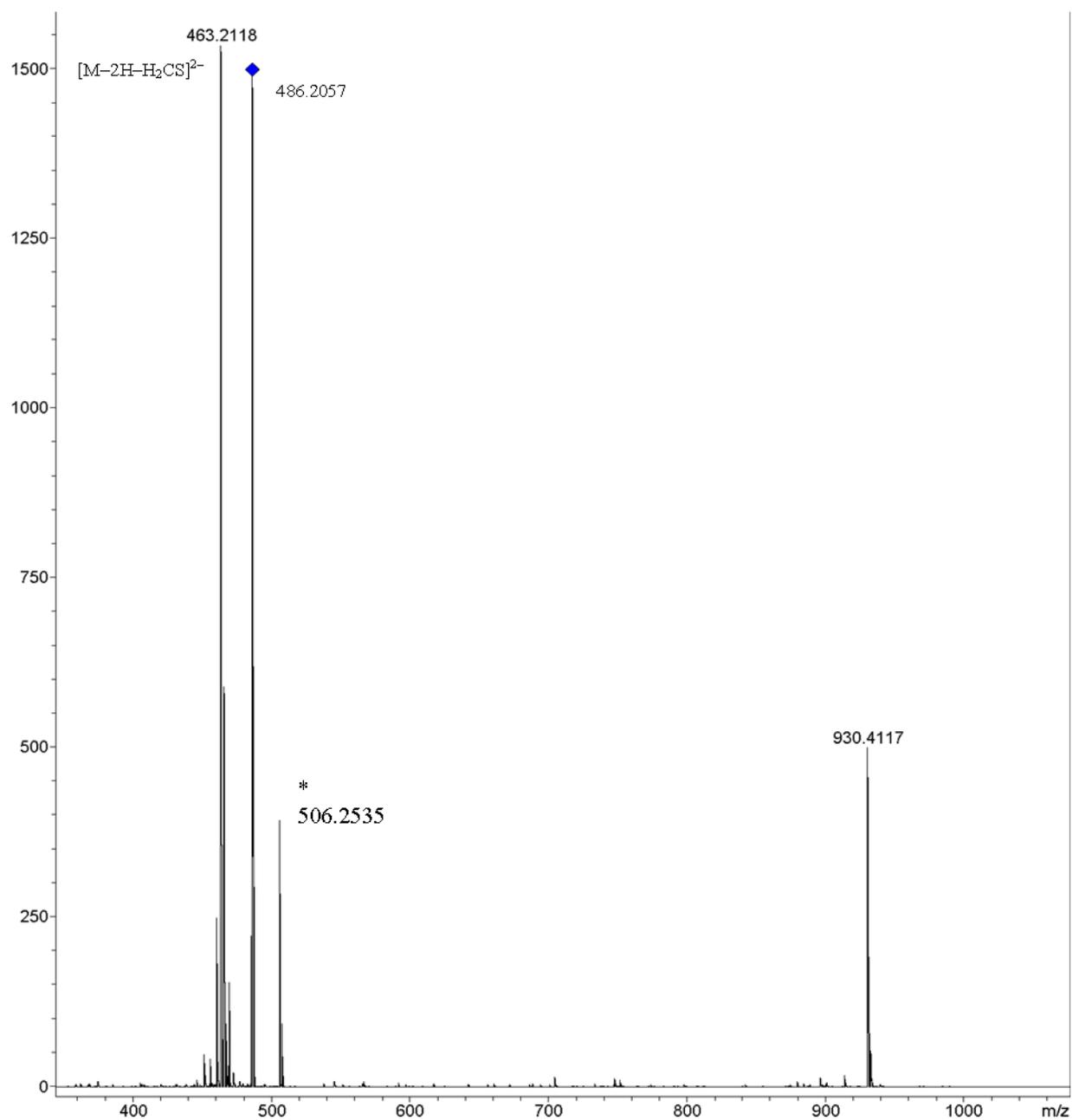


Fig. S1. High-resolution ESI MS/MS of an $[M-2H]^{2-}$ ion of compound **6** (activation energy 20 eV).

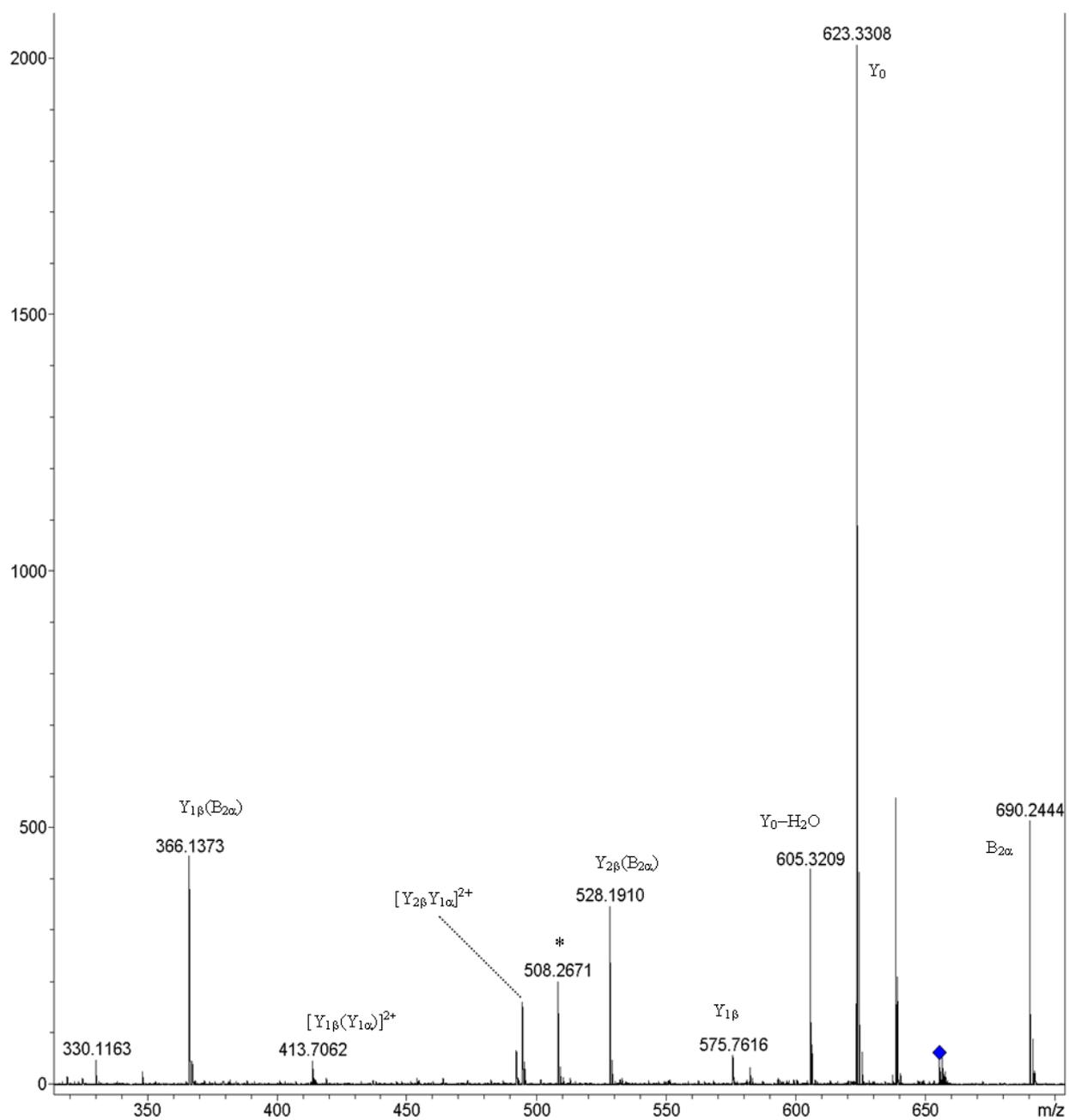


Fig. S2. High-resolution ESI MS/MS of an $[M+2H]^{2+}$ ion of compound **7** (activation energy 20 eV).

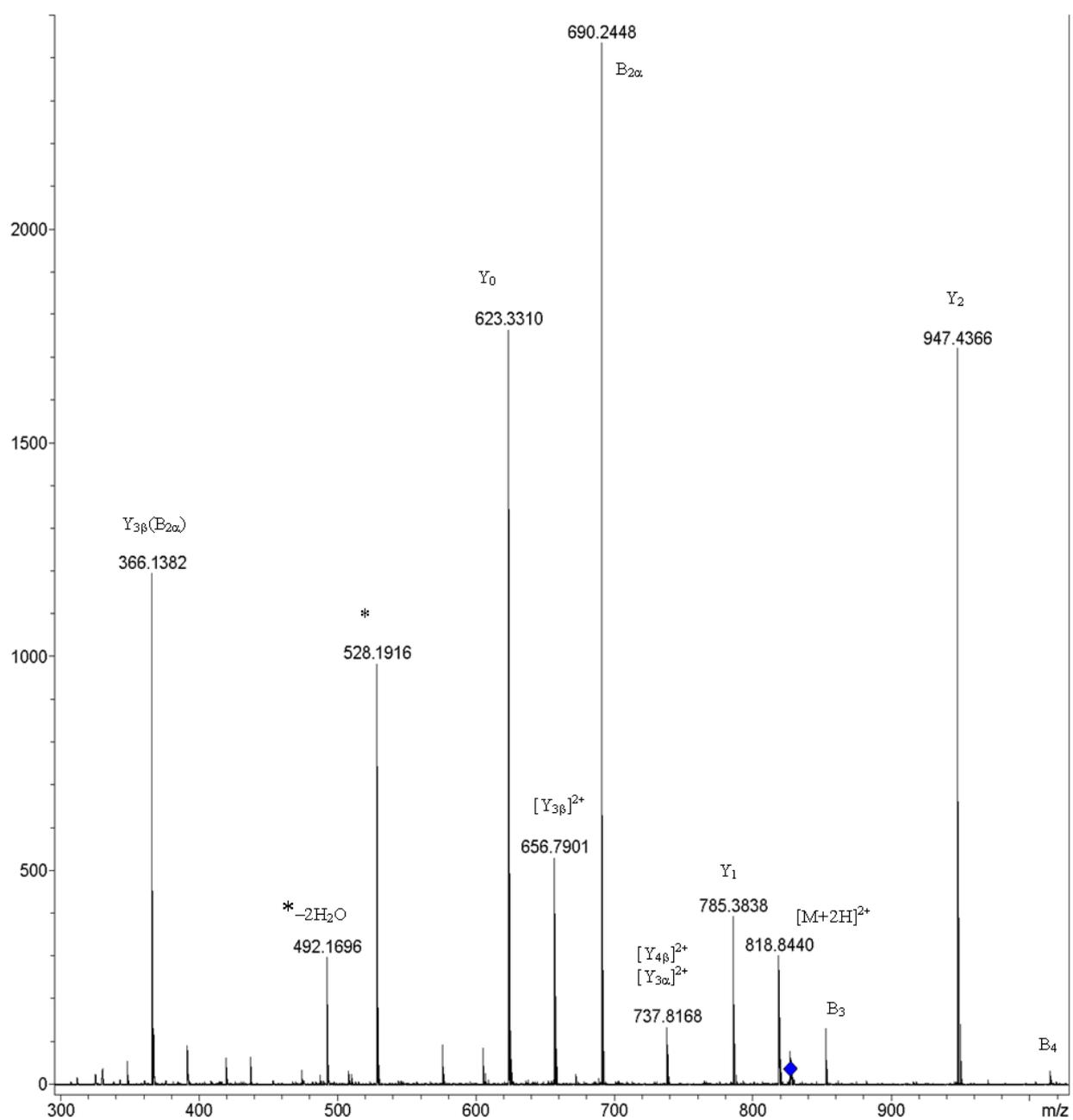


Fig. S3. High-resolution ESI MS/MS of an $[M+H+NH_4]^{2+}$ ion of compound **8** (activation energy 20 eV).

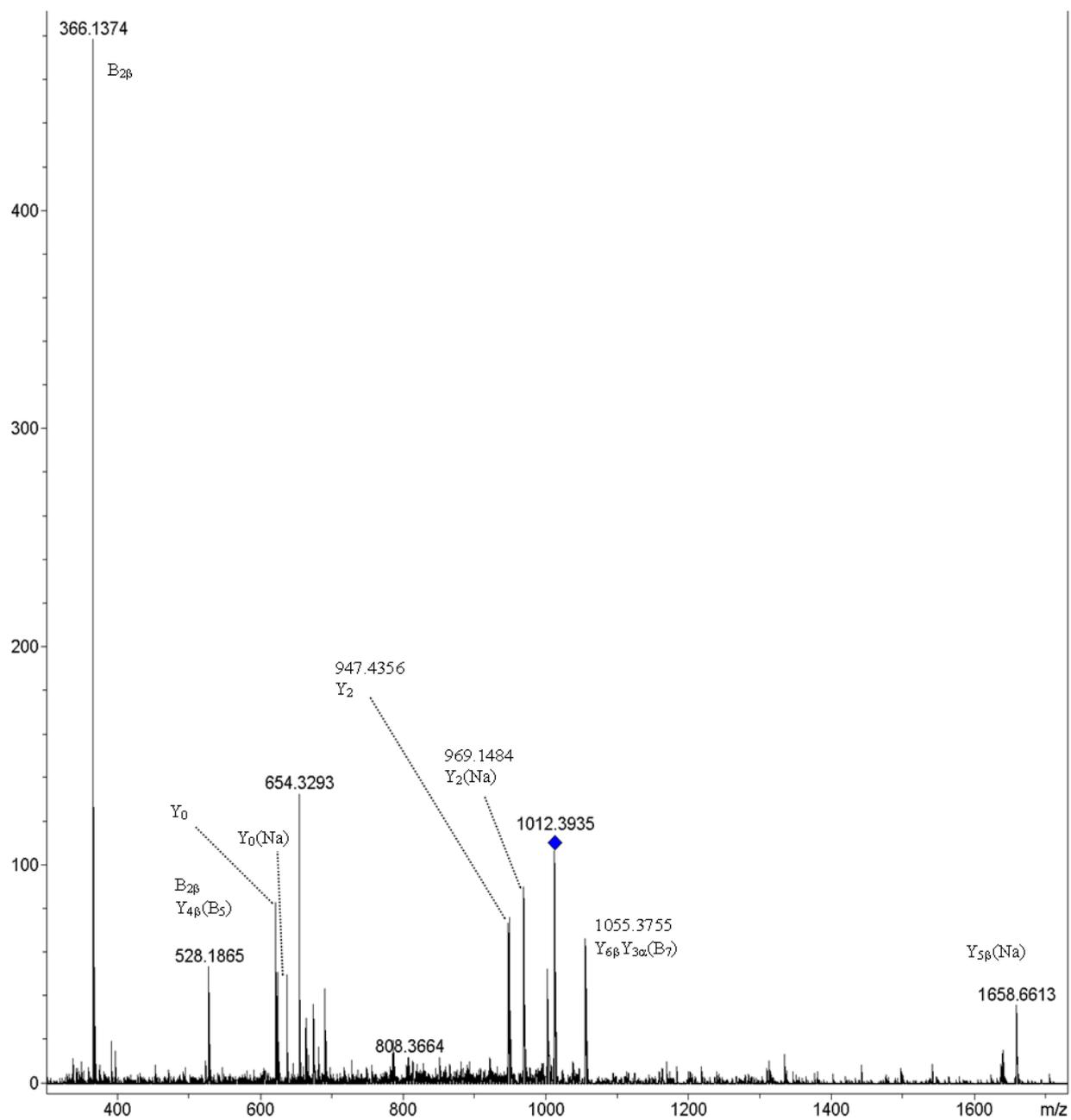


Fig. S4. High-resolution ESI MS/MS of an $[M+H+Na]^{2+}$ ion of compound **9** (activation energy 20 eV).

Table S1.

High-resolution ESI mass spectra of molecular probes **6–9**. Calculated m/z values are given in parentheses.

| Ion | Compound | | | |
|-------------------|--------------------------|--------------------------|--------------------------|--------------------------|
| | 6 | 7 | 8 | 9 |
| $[M+H]^+$ | 975.4320 (975.4326) | 1312.5679 (1312.5699) | — | — |
| $[M+NH_4]^+$ | 992.4583 (992.4592) | 1329.5959 (1329.5964) | — | — |
| $[M+Na]^+$ | 997.4136 (997.4145) | 1334.5504 (1334.5518) | — | — |
| $[M+K]^+$ | 1013.3852 (1013.3885) | 1350.5234 (1350.5258) | — | — |
| $[M+2H]^{2+}$ | 488.2211 (488.2199) | 656.7899 (656.7886) | 818.8427 (818.8414) | 1001.4057 (1001.4075) |
| $[M+H+NH_4]^{2+}$ | 496.7308 (496.7332) | 665.3014 (665.3019) | 827.3549 (827.3547) | 1009.9194 (1009.9208) |
| $[M+H+Na]^{2+}$ | 499.2102 (499.2109) | 667.7803 (667.7796) | 829.8335 (829.8324) | 1012.3975 (1012.3985) |
| $[M+H+K]^{2+}$ | 507.1954 (507.1949) | 675.7670 (686.7575) | 837.8202 (837.8193) | 1020.3860 (1020.3854) |
| $[M+2Na]^{2+}$ | 510.2014 (510.2019) | 678.7717 (678.7705) | 840.8274 (840.8233) | 1023.3894 (1023.3894) |
| $[M+Na+K]^{2+}$ | 518.1868 (518.1889) | 686.7581 (686.7575) | 848.8121 (848.81030) | 1031.3795 (1031.3764) |
| $[M+2K]^{2+}$ | 526.1727 (526.1758) | 694.7451 (694.7445) | 856.7987 (856.7973) | 1039.3702 (1039.3634) |
| $[M-H]^-$ | 973.4177 (973.4180) | 1310.5544 (1310.5553) | 1634.6611 (1634.6610) | 1999.7925 (1999.7932) |
| $[M-2H]^{2-}$ | 486.2050 (486.2054) | 654.7748 (654.7740) | 816.8275 (816.8269) | 999.3931 (999.3929) |
| $[M-3H]^{3-}$ | — | 436.1795 (436.1803) | 544.2155 (544.2155) | 685.9267 (685.9262) |

Table S2.

High-resolution ESI MS/MS of an $[M+NH_4]^+$ ion of compound **6** (activation energy 50 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------|--|-------------------------|
| 270.1303 | $C_{12}H_{20}N_3O_2S^+$ | Biot(CH ₂) ₂ O ⁺ | 270.1271 |
| 402.2056 | $C_{18}H_{32}N_3O_5S^+$ | | 402.2057 |
| 508.2684 | $C_{22}H_{42}N_3O_8S^+$ | * | 508.2687 |
| 619.3368 | $C_{28}H_{51}N_4O_9S^+$ | Y ₀ -H ₂ O | 619.3371 |
| 637.3472 | $C_{28}H_{53}N_4O_{10}S^+$ | Y ₀ | 637.3477 |

* The unusual cleavage of the HEG chain is designated as an asterisk (this table and below)

Table S3.

High-resolution ESI MS/MS of an $[M+Na]^+$ ion of compound **6** (activation energy 60 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------|---------------------|-------------------------|
| 530.2420 | $C_{22}H_{41}N_3O_8SNa^+$ | *(Na) | 530.2507 |
| 659.3261 | $C_{28}H_{52}N_4O_{10}SNa^+$ | Y ₀ (Na) | 659.3296 |
| 821.3821 | $C_{34}H_{62}N_4O_{15}SNa^+$ | Y ₁ (Na) | 821.3825 |
| 997.4132 | $C_{40}H_{70}N_4O_{21}SNa^+$ | residual peak | 997.4145 |

Table S4.

High-resolution ESI MS/MS of an $[M+K]^+$ ion of compound **6** (activation energy 60 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------|--------------------|-------------------------|
| 546.2165 | $C_{22}H_{41}N_3O_8SK^+$ | * | 546.2246 |
| 675.2994 | $C_{28}H_{52}N_4O_{10}SK^+$ | Y ₀ (K) | 675.3036 |
| 837.3564 | $C_{34}H_{62}N_4O_{15}SK^+$ | Y ₁ (K) | 837.3564 |
| 1013.3849 | $C_{40}H_{70}N_4O_{21}SK^+$ | residual peak | 1013.3885 |

Table S5.

High-resolution ESI MS/MS of an $[M+2H]^{2+}$ ion of compound **6** (activation energy 60 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------|--|-------------------------|
| 270.1258 | $C_{12}H_{20}N_3O_2S^+$ | Biot(CH ₂) ₂ O ⁺ | 270.1271 |
| 310.1716 | | unk. | |

| | | | |
|----------|-------------------------------|------------------------------------|----------|
| 319.1767 | $C_{28}H_{54}N_4O_{10}S^{2+}$ | $[Y_0+2H]^{2+}$ | 319.1775 |
| 350.2157 | | unk. | |
| 368.2265 | | unk. | |
| 376.1862 | $C_{16}H_{30}N_3O_5S^+$ | *-3C ₂ H ₄ O | 376.1901 |
| 420.2162 | $C_{18}H_{34}N_3O_6S^+$ | *-2C ₂ H ₄ O | 420.2163 |
| 464.2416 | $C_{20}H_{38}N_3O_7S^+$ | *-C ₂ H ₄ O | 464.2425 |
| 508.2685 | $C_{22}H_{42}N_3O_8S^+$ | * | 508.2687 |
| 619.3359 | $C_{28}H_{51}N_4O_9S^+$ | Y ₀ +H-H ₂ O | 619.3371 |
| 637.3471 | $C_{28}H_{53}N_4O_{10}S^+$ | Y ₀ +H | 637.3477 |

Table S6.

High-resolution ESI MS/MS of an $[M+2Na]^{2+}$ ion of compound **6** (activation energy 25 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|-----------------------------------|---------------------|-------------------------|
| 340.1497 | | unk. | |
| 422.1856 | $C_{34}H_{62}N_4O_{15}SNa^{2+}$ | $[Y_1(2Na)]^{2+}$ | 422.1858 |
| 510.2020 | $C_{40}H_{70}N_4O_{21}SNa_2^{2+}$ | residual peak | 510.2019 |
| 659.3299 | $C_{28}H_{52}N_4O_{10}SNa^+$ | Y ₀ (Na) | 659.3296 |
| 821.3808 | $C_{34}H_{62}N_4O_{15}SNa^+$ | Y ₁ (Na) | 821.3825 |
| 997.4155 | $C_{40}H_{70}N_4O_{21}SNa^+$ | $[M+Na]^+$ | 997.4145 |

Table S7.

High-resolution ESI MS/MS of an $[M-H]^-$ ion of compound **6** (activation energy 60 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------|---------------------|-------------------------|
| 460.2646 | $C_{21}H_{38}N_3O_8^-$ | *-H ₂ CS | 460.2664 |
| 466.1558 | $C_{22}H_{28}NO_8S^-?$ | unk. | 466.1541 |
| 472.2653 | $C_{22}H_{38}N_3O_8^-$ | *-H ₂ S | 472.2664 |
| 506.2537 | $C_{22}H_{40}N_3O_8S^-$ | * | 506.2542 |
| 973.4175 | $C_{40}H_{69}N_4O_{21}S^-$ | residual peak | 973.4180 |

Table S8.

High-resolution ESI MS/MS of an $[M-2H]^{2-}$ ion of compound **6** (activation energy 20 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|--------------------------------|---|-------------------------|
| 451.6941 | $C_{37}H_{65}N_3O_{20}S^{2-}?$ | unk. | 451.6947 |
| 456.1873 | unk./2- | unk. | |
| 460.2659 | $C_{21}H_{38}N_3O_8^-$ | *-H ₂ CS | 460.2664 |
| 463.2118 | $C_{39}H_{66}N_4O_{21}^{2-}$ | $[M-2H-H_2CS]^{2-}$ | 463.2115 |
| 466.1559 | $C_{18}H_{28}NO_{13}^-$ | GlcAGlcO(CH ₂) ₂ NHCO(CH ₂) ₂ | 466.1566 |
| 469.2125 | $C_{40}H_{66}N_4O_{21}^{2-}$ | $[M-2H-H_2S]^{2-}$ | 469.2115 |
| 486.2057 | $C_{40}H_{68}N_4O_{21}S^{2-}$ | residual peak | 486.2054 |
| 506.2535 | $C_{22}H_{40}N_3O_8S^-$ | * | 506.2542 |
| 930.4117 | $C_{39}H_{68}N_3O_{20}S^-$ | $[M-2H-CNO]^-$ | 930.4122 |

Table S9.High-resolution ESI MS/MS of an $[M+2H]^{2+}$ ion of compound **7** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|-------------------------------|---|-------------------------|
| 330.1163 | $C_{14}H_{20}NO_8^+$ | $Y_{1\beta}(B_{2\alpha})-2H_2O$ | 330.1183 |
| 366.1373 | $C_{14}H_{24}NO_{10}^+$ | $Y_{1\beta}(B_{2\alpha})$ | 366.1395 |
| 413.7062 | $C_{35}H_{65}N_5O_{15}S^{2+}$ | $[Y_{1\beta}(Y_{1\alpha})]^{2+}$ | 413.7093 |
| 492.1700 | $C_{20}H_{30}NO_{13}^{+?}$ | $Y_{2\beta}(B_{2\alpha})-2H_2O$ and $Y_{1\alpha}(B_{2\alpha})-2H_2O$ | 492.1712 |
| 494.7368 | $C_{41}H_{75}N_5O_{20}S^{2+}$ | $[Y_{2\beta}Y_{1\alpha}]^{2+}$ | 494.7358 |
| 508.2671 | $C_{22}H_{42}N_3O_8S^+$ | * | 508.2687 |
| 528.1910 | $C_{20}H_{34}NO_{15}^+$ | $Y_{2\beta}(B_{2\alpha})$ and $Y_{1\alpha}(B_{2\alpha})$ | 528.1923 |
| 575.7616 | $C_{47}H_{85}N_5O_{25}S^{2+}$ | $Y_{1\beta}$ and $Y_{2\beta}(Y_{1\alpha})$ | 575.7622 |
| 605.3209 | $C_{27}H_{49}N_4O_9S^+$ | Y_0-H_2O | 605.3215 |
| 623.3308 | $C_{27}H_{51}N_4O_{10}S^+$ | Y_0 | 623.3320 |
| 638.3078 | $C_{31}H_{48}N_3O_9S^{+?}$ | unk. | 638.3106 |
| 690.2444 | $C_{26}H_{44}NO_{20}^+$ | $B_{2\alpha}$ | 690.2451 |

Table S10.High-resolution ESI MS/MS of an $[M+H+Na]^{2+}$ ion of compound **7** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|---------------------------------|---|-------------------------|
| 330.1171 | $C_{14}H_{20}NO_8^+$ | $Y_{1\beta}(B_{2\alpha})-2H_2O$ | 330.1183 |
| 366.1374 | $C_{14}H_{24}NO_{10}^+$ | $Y_{1\beta}(B_{2\alpha})$ | 366.1395 |
| 424.6995 | unk./2+ | | |
| 492.1696 | $C_{20}H_{30}NO_3^+$ | $Y_{2\beta}(B_{2\alpha})-2H_2O$ and $Y_{1\alpha}(B_{2\alpha})-2H_2O$ | 492.1712 |
| 505.7262 | $C_{41}H_{74}N_5O_{20}SNa^{2+}$ | $[Y_{2\beta}(Y_{1\alpha})(Na)]^{2+}$ | 505.7267 |
| 508.2675 | $C_{22}H_{42}N_3O_8S^+$ | * | 508.2687 |
| 528.1914 | $C_{20}H_{34}NO_{15}^+$ | $Y_{2\beta}(B_{2\alpha})$ and $Y_{1\alpha}(B_{2\alpha})$ | 528.1923 |
| 586.7532 | $C_{47}H_{84}N_5O_{25}SNa^{2+}$ | $[Y_{1\alpha}(Na)]^{2+}$ and $[Y_{2\beta}(Na)]^{2+}$ | 586.7531 |
| 605.3205 | $C_{27}H_{49}N_4O_9S^+$ | Y_0-H_2O | 605.3215 |
| 623.3313 | $C_{27}H_{51}N_4O_{10}S^+$ | Y_0 | 623.3320 |
| 645.3133 | $C_{27}H_{50}N_4O_{10}SNa^+$ | $Y_0(Na)$ | 645.3140 |
| 657.7908 | unk./2+ | | |
| 667.7739 | $C_{53}H_{94}N_5O_{30}SNa^{2+}$ | residual peak | 667.7796 |
| 690.2445 | $C_{26}H_{44}NO_{20}^+$ | $B_{2\alpha}$ | 690.2451 |
| 848.3935 | $C_{35}H_{63}N_5O_{15}SNa^+$ | $Y_{1\beta}(Y_{1\alpha})(Na)$ | 848.3934 |
| 1010.4464 | $C_{41}H_{73}N_5O_{20}SNa^+$ | $Y_{2\beta}(Y_{1\alpha})(Na)$ | 1010.4462 |

Table S11.High-resolution ESI MS/MS of an $[M+2Na]^{2+}$ ion of compound **7** (activation energy 50 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|---|---|-------------------------|
| 530.2510 | C ₂₂ H ₄₁ N ₃ O ₈ SNa ⁺ | *(Na) | 530.2507 |
| 597.7432 | C ₄₇ H ₈₃ N ₅ O ₂₅ SNa ₂ ²⁺ | [Y _{2β} (2Na)] ²⁺ and [Y _{1α} (2Na)] ²⁺ | 597.7441 |
| 627.3035 | C ₂₇ H ₄₈ N ₄ O ₉ SNa ⁺ | Y ₀ (Na)–H ₂ O | 627.3034 |
| 645.3130 | C ₂₇ H ₅₀ N ₄ O ₁₀ SNa ⁺ | Y ₀ (Na) | 645.3140 |
| 678.7712 | C ₅₃ H ₉₃ N ₅ O ₃₀ SNa ₂ ²⁺ | residual peak | 678.7705 |
| 848.3884 | C ₃₅ H ₆₃ N ₅ O ₁₅ SNa ⁺ | Y _{1β} (Y _{1α})(Na) | 848.3934 |
| 1010.4424 | C ₄₁ H ₇₃ N ₅ O ₂₀ SNa ⁺ | Y _{2β} (Y _{1α})(Na) | 1010.4462 |
| 1172.4953 | C ₄₇ H ₈₃ N ₅ O ₂₅ SNa ⁺ | Y _{2β} (Na) and Y _{1α} (Na) | 1172.4990 |
| 1334.5499 | C ₅₃ H ₉₃ N ₅ O ₃₀ SNa ⁺ | [M+Na] ⁺ | 1334.5518 |

Table S12.High-resolution ESI MS/MS of an $[M+2H]^{2+}$ ion of compound **8** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|--|---|-------------------------|
| 366.1334 | C ₁₄ H ₂₄ NO ₁₀ ⁺ | B _{2β} | 366.1395 |
| 429.2354 | C ₁₆ H ₃₇ N ₄ O ₇ S ⁺ ? | unk. | 429.2317 |
| 528.1925 | C ₂₀ H ₃₄ NO ₅ ⁺ | Y _{4β} (B _{2α}) and Y _{3α} (B _{2α}) | 528.1923 |
| 623.3300 | C ₂₇ H ₅₁ N ₄ O ₁₀ S ⁺ | Y ₀ | 623.3320 |
| 690.2453 | C ₂₆ H ₄₄ NO ₂₀ ⁺ | B _{2α} | 690.2451 |
| 947.4390 | C ₃₉ H ₇₀ N ₄ O ₂₀ S ⁺ | Y ₂ | 947.4377 |

Table S13.High-resolution ESI MS/MS of an $[M+H+NH_4]^{2+}$ ion of compound **8** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|---|--|-------------------------|
| 366.1382 | C ₁₄ H ₂₄ NO ₁₀ ⁺ | Y _{3β} (B _{2α}), etc. | 366.1395 |

| | | | |
|-----------|--------------------------------|--|-----------|
| 492.1696 | $C_{20}H_{30}NO_{13}^+$ | *-2H ₂ O | 492.1712 |
| 508.2692 | $C_{22}H_{42}N_3O_8S^+$ | * | 508.2687 |
| 510.1819 | $C_{20}H_{32}NO_{14}^+$ | Y _{4β} (B _{2α})-H ₂ O and Y _{3α} (B _{2α})-H ₂ O | 510.1817 |
| 528.1916 | $C_{20}H_{34}NO_{15}^+$ | Y _{4β} (B _{2α}) and Y _{3α} (B _{2α}) | 528.1923 |
| 575.7639 | $C_{47}H_{85}N_5O_{25}S^{2+}$ | [Y _{3β} (Y _{3α})] ²⁺ and [Y _{3α} (Y _{3β})] ²⁺ | 575.7622 |
| 605.3208 | $C_{27}H_{49}N_4O_9S^+$ | Y ₀ -H ₂ O | 605.3215 |
| 623.3310 | $C_{27}H_{51}N_4O_{10}S^+$ | Y ₀ | 623.3320 |
| 656.7901 | $C_{53}H_{95}N_5O_{30}S^{2+}$ | [Y _{3β}] ²⁺ and [Y _{4β} (Y _{3α})] ²⁺ | 656.7886 |
| 690.2448 | $C_{26}H_{44}NO_{20}^+$ | B _{2α} | 690.2451 |
| 737.8168 | $C_{59}H_{105}N_5O_{35}S^{2+}$ | [Y _{4β}] ²⁺ and [Y _{3α}] ²⁺ | 737.8150 |
| 785.3838 | $C_{33}H_{61}N_4O_{15}S^+$ | Y ₁ | 785.3838 |
| 818.8440 | $C_{65}H_{115}N_5O_{40}S^{2+}$ | [M+2H] ²⁺ | 818.8414 |
| 852.2962 | $C_{32}H_{54}NO_{25}^+$ | B ₃ | 852.2979 |
| 947.4366 | $C_{39}H_{71}N_4O_{20}S^+$ | Y ₂ | 947.4377 |
| 1014.3499 | $C_{38}H_{64}NO_{30}^+$ | B ₄ | 1014.3508 |

Table S14.High-resolution ESI MS/MS of an $[M+H+Na]^{2+}$ ion of compound **8** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|----------------------------------|---|-------------------------|
| 366.1391 | $C_{14}H_{24}NO_{10}^+$ | $Y_{3\beta}(B_{2\alpha})$, etc. | 366.1395 |
| 492.1699 | $C_{20}H_{30}NO_{13}^+$ | *-2H ₂ O | 492.1712 |
| 528.1912 | $C_{20}H_{34}NO_{15}^+$ | $Y_{4\beta}(B_{2\alpha})$ and $Y_{3\alpha}(B_{2\alpha})$ | 528.1923 |
| 623.3324 | $C_{27}H_{51}N_4O_{10}S^+$ | Y_0 | 623.3320 |
| 645.3140 | $C_{27}H_{50}N_4O_{10}SNa^+$ | $Y_0(Na)$ | 645.3140 |
| 667.7734 | $C_{53}H_{93}N_5O_{30}SNa^{2+}$ | $[Y_{3\beta}(Na)]^{2+}$ and $[Y_{4\beta}Y_{3\alpha}(Na)]^{2+}$ | 667.7796 |
| 690.2447 | $C_{26}H_{44}NO_{20}^+$ | $B_{2\alpha}$ | 690.2451 |
| 712.2260 | $C_{26}H_{43}NO_{20}Na^+$ | $Y_1(Na)$ | 712.2271 |
| 748.8065 | $C_{59}H_{104}N_5O_{35}SNa^{2+}$ | $[Y_{4\beta}(Na)]^{2+}$ and $[Y_{3\alpha}(Na)]^{2+}$ | 748.8060 |
| 785.3855 | $C_{33}H_{61}N_4O_{15}S^+$ | Y_1 | 785.3849 |
| 807.3685 | $C_{33}H_{60}N_4O_{15}SNa^+$ | $Y_1(Na)$ | 807.3668 |
| 829.8300 | $C_{65}H_{114}N_5O_{40}SNa^{2+}$ | residual peak | 829.8324 |
| 947.4493 | $C_{39}H_{71}N_4O_{20}S^+$ | Y_2 | 947.4377 |
| 969.4231 | $C_{39}H_{70}N_4O_{20}SNa^+$ | $Y_2(Na)$ | 969.4196 |
| 1131.4819 | $C_{45}H_{80}N_4O_{25}SNa^+$ | # | 1131.4725 |
| 1293.5224 | $C_{51}H_{90}N_4O_{30}SNa^+$ | # | 1293.5253 |

#These small peaks arise apparently due to rearrangements, see M. Wuhrer, A.M. Deelder, and Y.E.M. van der Burgt, *Mass Spectrom. Rev.*, 2011, **30**, 664.

Table S15.

High-resolution MS/MS of an $[M-2H]^{2-}$ ion of compound **8** (activation energy 20 eV).

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|--------------------------------|------------------|-------------------------|
| 506.2538 | $C_{22}H_{40}N_3O_8S^-$ | * | 506.2542 |
| 563.1948 | $C_{43}H_{70}N_2O_{32}^{2-}$ | ** | 563.2542 |
| 816.8273 | $C_{65}H_{111}N_5O_{40}S^{2-}$ | residual peak | 816.8269 |
| 1127.3974 | $C_{43}H_{71}N_2O_{32}^-$ | ** | 1127.3995 |

Table S16.High-resolution ESI MS/MS of an $[M+H+NH_4]^{2+}$ ion of compound **9** (activation energy 20 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|--------------------------------|--|-------------------------|
| 366.1376 | $C_{14}H_{24}NO_{10}^+$ | $B_{2\beta}$, etc. | 366.1395 |
| 528.1909 | $C_{20}H_{34}NO_{15}^+$ | $B_{2\beta}$, $Y_{4\beta}(B_5)$, etc. | 528.1923 |
| 623.3384 | $C_{27}H_{51}N_4O_{10}S^+$ | Y_0 | 623.3320 |
| 690.2435 | $C_{26}H_{44}NO_{20}^+$ | $B_{3\beta}$, etc. | 690.2451 |
| 785.3810 | $C_{33}H_{61}N_4O_{15}S^+$ | Y_1 | 785.3849 |
| 818.8410 | $C_{65}H_{115}N_5O_{40}S^{2+}$ | $[Y_{5\beta}]^{2+}$ | 818.8414 |
| 852.2914 | $C_{32}H_{54}NO_{25}^+$ | $Y_{5\beta}(B_7)$ | 852.2979 |
| 893.3203 | $C_{34}H_{57}N_2O_{25}^+$ | $Y_{6\beta}(B_7)$ | 893.3245 |
| 920.3774 | $C_{73}H_{128}N_6O_{45}S^{2+}$ | $[Y_{6\beta}]^{2+}$ and $[Y_{3\alpha}]^{2+}$ | 920.3811 |
| 947.4336 | $C_{39}H_{71}N_4O_{20}S^+$ | Y_2 | 947.4377 |
| 1001.4096 | $C_{79}H_{138}N_6O_{50}S^{2+}$ | $[M+2H]^{2+}$ | 1001.4075 |
| 1055.3726 | $C_{40}H_{67}N_2O_{30}^+$ | $Y_{6\beta}Y_{3\alpha}(B_7)$ | 1055.3773 |
| 1109.4785 | $C_{45}H_{81}N_4O_{25}S^{+?}$ | unk. | 1109.4905 |
| 1217.4253 | $C_{46}H_{77}N_2O_{35}^+$ | $Y_{6\beta}(B_7)$ and $Y_{3\alpha}(B_7)$ | 1271.5433 |
| 1271.5467 | $C_{51}H_{91}N_4O_{30}S^{+?}$ | unk. | 1271.5433 |
| 1312.5606 | $C_{53}H_{94}N_5O_{30}S^+$ | $Y_{3\beta}$ | 1312.5699 |
| 1379.4763 | $C_{52}H_{87}N_2O_{40}^+$ | B_7 | 1379.4830 |
| 1474.6259 | $C_{59}H_{104}N_5O_{35}S^+$ | $Y_{4\beta}$ | 1474.6227 |
| 1636.6749 | $C_{65}H_{114}N_5O_{40}S^+$ | $Y_{5\beta}$ | 1636.6755 |

Table S17.High-resolution ESI MS/MS of an $[M+H+Na]^{2+}$ ion of compound **9** (activation energy 20 eV)

| m/z , experiment [#] | composition/charge of an ion | origin of an ion | m/z , calculated |
|---------------------------------|----------------------------------|---|--------------------|
| 366.1376 | $C_{14}H_{24}NO_{10}^+$ | $B_{2\beta}$, etc. | 366.1395 |
| 528.1874 | $C_{20}H_{34}NO_{15}^+$ | $B_{2\beta}$, $Y_{4\beta}(B_5)$, etc. | 528.1923 |
| 623.3313 | $C_{27}H_{51}N_4O_{10}S^+$ | Y_0 | 623.3320 |
| 645.3135 | $C_{27}H_{50}N_4O_{10}SNa^+$ | $Y_0(Na)$ | 645.3140 |
| 690.2411 | $C_{26}H_{44}NO_{20}^+$ | $B_{3\beta}$, etc | 690.2451 |
| 785.3780 | $C_{33}H_{61}N_4O_{15}S^+$ | Y_1 | 785.3849 |
| 807.3669 | $C_{33}H_{60}N_4O_{15}SNa^+$ | $Y_1(Na)$ | 807.3668 |
| 852.2887 | $C_{32}H_{54}NO_{25}^+$ | $Y_{5\beta}(B_7)$ | 852.2979 |
| 893.3258 | $C_{34}H_{57}N_2O_{25}^+$ | $Y_{6\beta}(B_7)$ | 893.3245 |
| 947.4336 | $C_{39}H_{71}N_4O_{20}S^+$ | Y_2 | 947.4377 |
| 969.4184 | $C_{39}H_{70}N_4O_{20}SNa^+$ | $Y_2(Na)$ | 969.4196 |
| 1012.3933 | $C_{79}H_{137}N_6O_{50}SNa^{2+}$ | residual peak | 1012.3985 |
| 1055.3755 | $C_{40}H_{67}N_2O_{30}^+$ | $Y_{6\beta} Y_{3\alpha}(B_7)$ | 1055.3773 |
| 1334.5576 | $C_{53}H_{93}N_5O_{30}SNa^+$ | $Y_{3\beta}(Na)$ | 1334.5518 |
| 1496.6018 | $C_{59}H_{103}N_5O_{35}SNa^+$ | $Y_{4\beta}(Na)$ | 1496.6047 |
| 1658.6549 | $C_{65}H_{113}N_5O_{40}SNa^+$ | $Y_{5\beta}(Na)$ | 1658.6575 |

[#]No peaks at m/z ca. 508 or 530 were registered.

Table S18.High-resolution ESI MS/MS of an $[M+2Na]^{2+}$ ion of compound **9** (activation energy 60 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|------------------------------------|--------------------------|-------------------------|
| 366.1396 | $C_{14}H_{24}NO_{10}^+$ | $B_{2\beta}$, etc. | 366.1395 |
| 388.1178 | $C_{14}H_{23}NO_{10}Na^+$ | $B_{2\beta}(Na)$, etc. | 388.1214 |
| 406.1289 | unk. | unk. | |
| 508.2660 | $C_{22}H_{42}N_3O_8S^+$ | *(H) | 508.2687 |
| 530.2493 | $C_{22}H_{41}N_3O_8SNa^+$ | *(Na) | 530.2507 |
| 645.3456 | $C_{27}H_{50}N_4O_{10}SNa^+$ | $Y_0(Na)?$ | 645.3140 |
| 759.7953 | $C_{59}H_{102}N_5O_{35}SNa^{2+}$ | $[Y_{4\beta}(2Na)]^{2+}$ | 759.2930 |
| 807.3619 | $C_{33}H_{60}N_4O_{15}SNa^+$ | Y_1 | 807.3668 |
| 840.8258 | $C_{65}H_{112}N_5O_{40}SNa_2^{2+}$ | $[Y_{5\beta}(2Na)]^{2+}$ | 840.3194 |
| 874.2822 | unk. | unk. | |
| 942.3700 | $C_{73}H_{126}N_6O_{45}SNa_2^{2+}$ | $[Y_{6\beta}(2Na)]^{2+}$ | 942.3630 |
| 969.4164 | $C_{39}H_{70}N_4O_{20}SNa^+$ | $Y_2(Na)$ | 969.4196 |
| 1023.3904 | $C_{79}H_{136}N_6O_{50}SNa_2^{2+}$ | residual peak | 1023.3894 |
| 1077.3517 | $C_{40}H_{66}N_2O_{30}SNa^+$ | $B_5(Na)$ | 1077.3593 |
| 1334.5282 | $C_{53}H_{93}N_5O_{30}SNa^+$ | $Y_{3\beta}(Na)$ | 1334.5518 |
| 1496.6031 | $C_{59}H_{103}N_5O_{35}SNa^+$ | $Y_{4\beta}(Na)$ | 1496.6047 |
| 1658.6730 | $C_{65}H_{113}N_5O_{40}SNa^+$ | $Y_{5\beta}(Na)$ | 1658.6575 |

Table S19.High-resolution ESI MS/MS of an $[M+Na+K]^{2+}$ ion of compound **9** (activation energy 60 eV)

| <i>m/z</i> , experiment | composition/charge of an ion | origin of an ion | <i>m/z</i> , calculated |
|-------------------------|-----------------------------------|--|-------------------------|
| 270.1272 | $C_{12}H_{20}N_3O_2S^+$ | Biot(CH ₂) ₂ O ⁺ | 270.1271 |
| 366.1376 | $C_{14}H_{24}NO_{10}^+$ | B _{2β} , etc. | 366.1395 |
| 508.2660 | $C_{22}H_{42}N_3O_8S^+$ | *(H) | 508.2687 |
| 530.2478 | $C_{22}H_{41}N_3O_8SNa^+$ | *(Na) | 530.2507 |
| 546.2289 | $C_{22}H_{41}N_3O_8SK^+$ | *(K) | 546.2246 |
| 807.3593 | $C_{33}H_{60}N_4O_{15}SNa^+$ | Y ₁ | 807.3668 |
| 969.4127 | $C_{39}H_{70}N_4O_{20}S^+$ | Y ₂ (Na) | 969.4196 |
| 1031.3804 | $C_{79}H_{136}N_6O_{50}SNaK^{2+}$ | residual peak | 1031.3764 |
| 1658.6729 | $C_{65}H_{113}N_5O_{40}SNa^+$ | Y _{5β} (Na) | 1658.6575 |
| 2023.8059 | $C_{79}H_{136}N_6O_{50}SNa^+$ | $[M+Na]^+$ | 2023.7897 [#] |

[#]The peak of $[M+K]^+$ (calcd. *m/z* 2039.7636) was not observed.