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"Click Chemistry" Inspired Synthesis of *pseudo*-Oligosaccharides and Amino Acid Glycoconjugates

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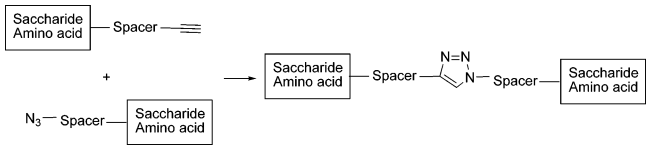

Various *pseudo*-oligosacchardies and amino acid glycoconjugates were synthesized via an intermolecular 1,3-dipolar cycloaddition ("click") reaction using easily accessible carbohydrate and amino acid derived azides and alkynes as building blocks. It is pertinent to mention that the conjugation reaction is highly regioselective and high yielding and can be carried out under mild reaction conditions.

Oligosaccharides and glycopeptides play crucial roles in various cellular recognition events including signal transduction.[1] A recent survey identified that synthetic oligosaccharides covalently attached to proteins facilitate the development of vaccines against *Haemophilus influenzae* type b, *Streptococcus pneumoniae*, and *Neisseria meningitides* group C.[2] Lack of sufficient quantities of oligosaccharides and peptides often limit the efficient conjugation of oligosaccharides to oligosaccharides/peptides. It is anticipated that such homo and hetero dimeric glycoconjugates will be excellent probes that can act as potent reversible cross-linking reagents and also to measure the distances between carbohydrate binding sites in polyvalent recognition sites.[1b] Thus there exists a demand to develop efficient strategies for the conjugation of oligosaccharides to oligosaccharides/peptides.

Many methods are available in the current literature to prepare homo and hetero dimers of oligosaccharides and glycoconjugates. Some of the important methods that give access to these interesting compounds are (i) olefin metathesis of alkenyl glycosides;[3a] (ii) intermolecular enyne metathesis of alkynyl and alkenyl glycosides;[3b] (iii) native chemical ligation of oligosaccharides to peptides;[3c] (iv) glycosylation of diols;[3d] (v) the cycloaddition of azide and alkyne under modified Huisgen ("click") conditions;[3e,f] and (vi) coupling of alkynyl glycosides.[3g] Many times, it is desirable to conduct the conjugation in aqueous solution, near physiological pH, at ambient temperature, and in short times in order to prevent the denaturation of the protein.

One of the reactions that can fulfill all of the above conditions is Huisgen's 1,3-dipolar cycloaddition reaction between terminal alkynes and azides resulting in the formation of 1,2,3-triazoles.[4] Recently, Cu(I) species was found to mediate the formation of 1,2,3-triazoles efficiently, and consequently regioselective synthesis of 1,2,3-triazoles through Huisgen's (3 + 2) cycloaddition between a terminal alkyne and an azide is currently referred to as a click reaction.[5] Application of this protocol to carbohydrate substrates has recently begun, and most of the reports are concerned with the use of anomeric azides.[3,6] However, introduction of a spacer between the sugar moiety and the protein is required, and currently available methods will not enable us to do so. Also it would be advantageous if the linker were at the terminus of the saccharide chain, and the conjugation reaction must proceed at equimolar ratio, should not yield offensive byproducts, and should enable recovery or removal of the unreacted conjugation partners. Recently, it was found that inhibition of hemagglutination by dimeric saccharides synthesized via cross metathesis reaction was superior compared to their corresponding monomeric glycosides.[3g] Apart from the synthetic promise, triazole moieties are also interesting conjugation entities as they are proven to be relatively stable to metabolic degradation and the triazole ring also can participate in the hydrogen bonding, which can be excellent in the context of biomolecular targets and solubility.[7] In view of the above facts, fascinating biological significance and continued interest[6a] in the application of click chemistry to carbohydrate substrates

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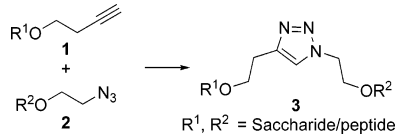
SCHEME 1. Click Chemistry Inspired Conjugation of Oligosaccharides to Oligosaccharides/Peptides



prompted us to develop an efficient procedure for the conjugation of oligosaccharides to oligosaccharides/amino acids (Scheme 1).

In our approach, we considered performing the Huisgen (click) reaction[5] between alkynyl (**1**) and azidoethyl (**2**) glycosides in the presence of CuI to obtain 1,2,3-triazole bridged homo and hetero dimeric oligosaccharides (**3**) (Scheme 1). Accordingly, for a pilot study, we synthesized 3-butynyl 2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranoside (**4**)[8] as an alkyne component using a $BF_3{\cdot}Et_2O$-mediated glycosylation. The other coupling partner was achieved in two steps from 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose. First, per-*O*-acetylated 2-chloroethyl-β-D-glucoside was synthesized[8] and subsequently converted to the azido derivative (**5**) using NaN_3 in anhydrous DMF at 80 °C for 12 h. In the ^{13}C NMR spectrum of compound **5**, the anomeric carbon was identified at δ 100.4 ppm and the azido methylene group was noticed at δ 50.3 ppm along with other resonances in accordance with the assigned structure.[9] The presence of the azido group was also evident from the IR spectrum wherein transmittance due to the azido group was observed at 2102 cm^{-1}.

The crucial 1,3-dipolar cycloaddition reaction in acetonitrile was successfully carried out between **4** and **5** using CuI in the presence of *N*,*N*-diisopropylethylamine (Scheme 2).[10] It is interesting to note that the Huisgen reaction was found to be highly regioselective, yielding 1,4-disubstituted 1,2,3-triazole containing *pseudo*-oligosaccharide (**6**) in 96% yield.[9] The ^1H NMR spectrum of conjugated compound **6** showed resonances corresponding to the acetyl group between δ 1.95 and 2.10 ppm as singlets, allylic methylene resonances were observed at δ 3.00 ppm (t, 2 H, J = 6.53 Hz), and the olefinic proton associated with the 1,2,3-triazole moiety was identified δ 7.43 ppm as a singlet along with other resonances in accordance with the assigned structure.[9] The ^{13}C NMR spectrum of compound **6** confirmed the presence of conjugated product as the two anomeric carbons were identified at δ 100.4 and 100.2 ppm, and olefinic carbons were noticed at δ 123.2 and 143.8 ppm. The DEPT spectrum revealed that the resonance at δ 143.8 was absent (a quarternary olefinic carbon) along with six inversely phased signals attributable to six -CH_2 groups(δ 26.0, 49.5, 61.4, 61.6, 67.5, and 68.4), thereby confirming the compound **6**.[9]

Furthermore, the IR spectrum of compound **6** showed transmittance due to a carbonyl group at 1753 cm^{-1}. In addition, the conjugated product gave satisfactory elemental analysis.

Having identified a practical procedure for the chemical ligation of oligosaccharides, we did an initial substrate compatibility survey using various alkynyl (**7**) and azidoalkyl (**8** and **9**[6a]) saccharides. As depicted in Scheme 2, we could extrapolate our efforts to conjugate *gluco*-, *lacto*-, and *xylo*-derived alkynes and azides to 1,2,3-triazole conjugated oligosaccharides (**10**, **11**, **12**, and **13**) successfully (Scheme 2).

Our next endeavors were devoted toward ligating oligosaccharides to amino acid derived azides and alkynes. Toward this, we have synthesized various alkynyl and azide containing amino acids using the traditional *t*-Boc chemistry and utilized the click chemistry guided conjugation reaction successfully (Scheme 3).

The reactive site (azide or alkyne) was separated from the cysteine or phenyl alanine by a 6-aminocaproic acid linker. Subsequently, azidoethyl lactoside (**8**) was reacted with amino acid anchored alkyne (**14**) in the presence of CuI to obtain the ligated glycoconjugate **15** in 91% yield. In the ^1H NMR spectrum of compound **15**, resonances corresponding to the seven acetyl groups were noticed between δ 1.96 and 2.15 ppm as singlets and that of the *tert*-butoxy carbonyl group was at δ 1.41 ppm as a sharp singlet. Also, the olefinic proton of 1,2,3-triazole was identified at δ 7.63 ppm as a singlet with other resonances in accordance with the assigned structure.

In the ^{13}C NMR spectrum of compound **15**, the anomeric carbons were noticed at δ 100.8 and 100.2 ppm, and aromatic carbons were observed from δ 126.7 to 129.4 ppm, along with 11 -CH_2 groups at 24.2, 26.0, 28.8, 33.7, 33.8, 39.0, 49.9, 57.2, 60.6, 61.5, 67.6 ppm, thereby confirming the ligated product. Furthermore, olefin carbons of the 1,2,3-triazole ring of compound **15** were observed at δ 124.7 and 142.5 (quarternary) ppm along with all other resonances in accordance with the assigned structure.[9] IR spectral analysis of **15** showed carbonyl stretches at 1751 cm^{-1} and those of the amide at 1670 cm^{-1}. In addition, compound **15** gave satisfactory elemental analysis. We then extrapolated the ligation protocol successfully to other amino acid derived azides (**16** and **17**) and synthesized various glycoconjugates (**18**, **19**, **20**, and **21**) (Scheme 3).

In summary, we have developed a practical procedure for the ligation of oligosaccharides to oligosaccharides/peptides under neutral reaction conditions. We anticipate that the 1,2,3-triazole containing dimeric saccharides and amino acid glycoconjugates may show a variety of bioactivities as several compounds containing 1,2,3-triazoles display broad spectra of biological activities including antibacterial,[11a] herbicidal and fungicidal,[11b] antiallergic,[11c] and anti-HIV.[11d] A combination of different monosaccharide and peptide building blocks has been showed and should potentially give rise to a large number of *pseudo*-oligosaccharides and amino acid glycoconjugates.

Experimental Section

General Experimental Procedure for the Conjugation of Oligosaccharides to Oligosaccharides/Peptides. To a solution of alkyne (1 mmol) and azide (1 mmol) in 10 mL of anhydrous acetonitrile were added CuI (2 mmol) and *N*,*N*-diisopropylethylamine (3 mmol) at room temperature, and the mixture was stirred for the specified time. At the end of the reaction as judged by TLC analysis, the reaction mixture was diluted using 25 mL of water and 10 mL of NH_4Cl, the aqueous layer was extracted with ethyl acetate (3 × 50 mL), and the combined organic layer was washed

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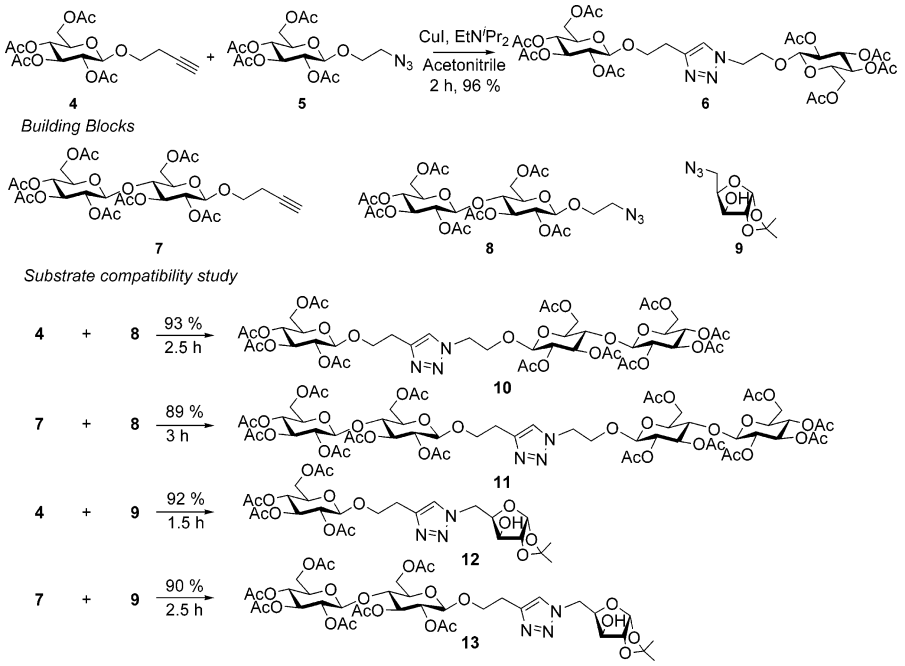
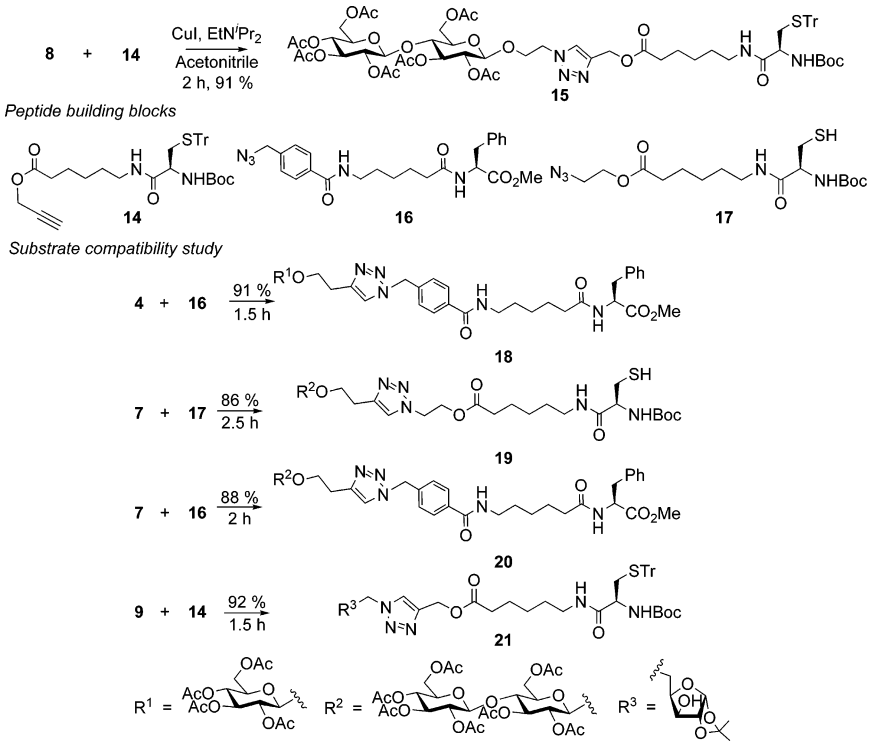
SCHEME 2. Click Chemistry Guided Synthesis of *pseudo*-Oligosaccharides



SCHEME 3. Conjugation of Oligosaccharides to Amino Acid Derived Azides and Alkynes



with brine solution, dried over anhydrous sodium sulfate, and concentrated in vacuo to obtain a crude residue that was purified by silica gel column chromatography using a gradient of ethyl acetate and petroleum ether (60−80 °C) to obtain the desired 1,4-disubstited 1,2,3-triazole as a white solid.

Characterization data of compound **6**: mp = 115−118 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.1) = −15.30. IR (cm^{-1}): 1753. ^{1}H NMR ($CDCl_3$, 200 MHz): δ 1.95, 2.00, 2.02, 2.09 (4s, 24 H), 3.00 (t, 2 H, J = 6.53 Hz), 3.72 (m, 2 H), 3.89 (m, 2 H), 4.07−4.33 (m, 6 H), 4.45−4.58 (m, 4 H), 4.90−5.25 (m, 6 H), 7.43 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.2−20.5, 26.0, 49.5, 61.4, 61.6, 67.5, 67.9, 68.0, 68.5, 70.6, 70.9, 71.4, 71.6, 72.1, 72.4, 100.2, 100.4, 123.2, 143.8, 168.9, 169.0, 169.1, 169.2, 169.8, 169.9, 170.2, 170.3. Anal. Calcd for $C_{34}H_{47}N_3O_{20}$: C, 49.94; H, 5.79; N, 5.14. Found: C, 50.13; H, 5.49; N, 4.92. MALDI-TOF: mol wt calcd 817.75, found 840.83 (M + 23 for Na).

Characterization data of compound **10**: mp = 158−162 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.0) = −4.58. IR (cm^{-1}): 1753. ^{1}H NMR ($CDCl_3$, 200 MHz): δ 1.94, 1.95, 1.97, 2.00, 2.03, 2.04, 2.05, 2.06, 2.09, 2.13, 2.15 (11s, 33 H), 2.99 (t, 3 H, J = 6.48 Hz), 3.45−3.98 (m, 5 H), 4.00−4.36 (m, 8 H), 4.51 (m, 5 H), 4.81−5.27 (m, 7 H), 5.35 (d, 1 H, J = 2.92 Hz), 7.42 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ

20.4−20.7, 26.1, 49.6, 60.6, 61.6, 61.7, 66.4, 67.7, 68.2, 68.7, 68.9, 70.5, 70.8, 71.0, 71.1, 71.6, 72.3, 72.6, 72.6, 75.9, 100.2, 100.6, 100.9, 123.3, 143.9, 168.9, 169.1, 169.3, 169.3, 169.5, 169.9, 170.0, 170.1, 170.2, 170.2, 170.5. Anal. Calcd for $C_{46}H_{63}N_3O_{28}$: C, 49.95; H, 5.74; N, 3.80. Found: C, 50.01; H, 5.86; N, 3.76.

Characterization data of compound **11**: mp = 122−125 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.2) = + 1.14. IR (cm^{-1}): 1751. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.93−2.17 (14s, 42 H), 2.98 (t, 2 H, *J* = 6.42 Hz), 3.59 (m, 2 H), 3.72−3.91 (m, 6 H), 4.00−4.20 (m, 8 H), 4.32−4.61 (m, 7 H), 4.81−5.23 (m, 9 H), 5.34 (dd, 2 H, *J* = 0.61, 3.02 Hz), 7.41 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.3−20.7, 26.1, 49.7, 60.6, 60.6, 61.6, 61.8, 66.4, 66.4, 67.7, 68.6, 68.9, 68.9, 70.4, 70.5, 70.6, 70.8, 70.9, 71.1, 71.4, 72.3, 72.5, 72.5, 75.8, 76.0, 100.2, 100.4, 100.8, 100.9, 123.3, 143.9, 168.8−170.5. Anal. Calcd for $C_{58}H_{79}N_3O_{36}$: C, 49.96; H, 5.71; N, 3.01. Found: C, 50.20; H, 5.49; N, 3.07.

Characterization data of compound **12**: mp = 138−142 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.1) = −26.34. IR (cm^{-1}): 1753. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.31, 1.46 (2s, 6 H), 1.97, 2.00, 2.03, 2.07 (4s, 12 H), 3.01 (t, 2 H, *J* = 6.19 Hz), 3.65−3.78 (m, 1 H), 3.81 (m, 1 H), 3.92 (d, 1 H, *J* = 5.05 Hz), 4.07−4.20 (m, 3 H), 4.28 (dd,1 H, *J* = 4.81, 12.39 Hz), 4.45−4.63 (m, 4 H), 4.73 (dd, 1 H, *J* = 7.70, 13.43 Hz), 5.01 (dd, 1 H, *J* = 9.13, 17.37 Hz), 5.12 (d, 1 H, *J* = 16.30 Hz), 5.18 (ABq, 1 H, *J* = 9.31 Hz), 5.99 (d, 1 H, *J* = 3.41 Hz), 7.56 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.3, 20.4, 20.5, 20.6, 26.0, 26.2, 26.6, 48.4, 61.7, 68.2, 68.5, 71.2, 71.7, 72.5, 74.1, 79.1, 85.2, 100.6, 105.0, 111.8, 123.6, 144.3, 169.3, 169.5, 170.1, 170.6. Anal. Calcd for $C_{26}H_{37}N_3O_{14}$: C, 50.73; H, 6.06; N, 6.83. Found: C, 50.44; H, 6.47; N, 6.67.

Characterization data of compound **13**: mp = 145−150 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.0) = − 7.53. IR (cm^{-1}): 1753. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.31, 1.45 (2s, 6 H), 1.97, 1.98, 2.05, 2.06, 2.07, 2.12, 2.16 (7s, 21 H), 2.99 (t, 2 H, *J* = 5.92 Hz), 3.65 (m, 1 H), 3.70−3.95 (m, 4 H), 4.01−4.19 (m, 5 H), 4.40−4.65 (m, 6 H), 4.71 (dd, 1 H, *J* = 7.70, 13.38 Hz), 4.78−5.25 (m, 4 H), 5.35 (d, 1 H, *J* = 2.84 Hz), 5.99 (d, 1 H, *J* = 3.40 Hz), 7.54 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.4−20.9, 26.1, 26.3, 26.7, 48.1, 60.7, 61.8, 66.5, 68.5, 69.0, 70.6, 70.9, 71.6, 72.5, 72.6, 74.2, 76.1, 79.1, 85.2, 100.5, 101.0, 105.1, 111.9, 123.6, 144.5, 169.0, 169.7, 169.9, 170.0, 170.1, 170.3, 170.4. Anal. Calcd for $C_{38}H_{53}N_3O_{22}$: C, 50.50; H, 5.90; N, 4.65. Found: C, 50.55; H, 5.67; N, 4.78.

Characterization data of compound **15**: mp = 98−102 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.1) = +10.36. IR (cm^{-1}): 1751, 1670. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.26 (m, 3 H), 1.41 (s, 9 H), 1.59 (dd, 3 H, *J* = 7.36, 15.13 Hz), 1.96, 1.97, 2.03, 2.04, 2.06, 2.12, 2.15 (7s, 21 H), 2.29 (t, 2 H, *J* = 7.20 Hz), 2.50 (dd, 1 H, *J* = 5.44, 12.75 Hz), 2.70 (dd, 1 H, *J* = 6.93, 12.81 Hz), 3.17 (q, 2 H, *J* = 6.66, 12.97 Hz), 3.61 (m, 1 H), 3.72−3.97 (m, 4 H), 4.01−4.26 (m, 4 H), 4.40−4.63 (m, 5 H), 4.80−5.25 (m, 7 H), 5.35 (d, 1 H, *J* = 3.09 Hz), 6.08 (t, 1 H, *J* = 5.47 Hz), 7.15−7.45 (m, 15 H), 7.63 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.3−20.7, 24.2, 26.0, 28.1, 28.8, 33.7, 33.8, 39.0, 49.9, 53.4, 57.2, 60.6, 61.5, 66.4, 66.9, 67.6, 68.9, 70.5, 70.7, 71.1, 72.3, 72.6, 75.8, 80.0, 100.2, 100.8, 124.7, 126.7−129.4, 142.5, 144.2, 155.1, 168.9, 169.4, 169.5, 169.9, 170.0, 170.1, 170.1, 170.2, 173.1. Anal. Calcd for $C_{64}H_{81}N_5O_{23}S$: C, 58.20; H, 6.18; N, 5.30; S, 2.43. Found: C, 57.95; H, 6.52; N, 6.62; S, 2.20.

Characterization data of compound **18**: mp = 128−132 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.0) = +6.65. IR (cm^{-1}): 1747, 1651. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.35 (m, 2 H), 1.64 (m, 4 H), 1.89, 1.99, 2.02, 2.05 (4s, 12 H), 2.19 (t, 2 H, *J* = 6.99 Hz), 2.99 (t, 2 H, *J* = 6.33 Hz), 3.09 (t, 2 H, *J* = 6.00 Hz), 3.42 (dd, 2 H, *J* = 6.45, 12.37 Hz), 3.66 (m, 1 H), 3.71 (s, 3 H), 3.85 (m, 1 H), 4.08 (m, 2 H), 4.23 (dd, 1 H, *J* = 4.77, 12.22 Hz), 4.50 (d, 1 H, *J* = 7.94 Hz), 4.80−5.25 (m, 4 H), 5.54 (bs, 2 H), 6.01 (d, 1 H, *J* = 7.31 Hz), 6.62 (m, 1 H), 7.08 (m, 2 H), 7.26 (m, 3 H), 7.30 (d, 2 H, *J* = 8.28 H), 7.37 (s, 1 H), 7.80 (d, 2 H, *J* = 8.27 Hz). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.4−20.6, 24.6, 26.1, 26.3, 28.8, 35.8, 37.6, 38.5, 39.5, 52.2, 52.9, 53.3, 61.8, 68.2, 68.5, 71.1, 71.6, 72.6, 100.6, 123.3, 126.8−129.1, 134.9, 135.8, 137.9, 144.9, 166.7, 169.3, 169.4, 170.1, 170.5, 172.1, 172.5. Anal. Calcd for $C_{42}H_{53}N_5O_{14}$: C, 59.21; H, 6.27; N, 8.22. Found: C, 59.43; H, 6.35; N, 8.25.

Characterization data of compound **19**: mp = 78−82 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.0) = +6.48. IR (cm^{-1}): 1749, 1666. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.27 (m, 2 H), 1.46 (s, 9 H), 1.58 (m, 4 H), 1.96, 1.97, 2.04, 2.05, 2.06, 2.12, 2.15 (7s, 21 H), 2.31 (t, 2 H, *J* = 7.42 Hz), 3.00 (m, 4 H), 3.22 (m, 2 H), 3.48−3.90 (m, 5 H), 4.00−4.30 (m, 5 H), 4.32−4.63 (m, 7 H), 4.65−5.25 (m, 4 H), 5.35 (d, 1 H, *J* = 2.75 Hz), 5.64 (d,1 H, *J* = 9.11 Hz), 7.45 (s, 1 H), 7.72 (bs, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.3−20.8, 24.3, 26.3, 28.3−28.4, 29.3, 33.6, 33.7, 39.3, 46.8, 49.0, 54.7, 60.7, 62.1, 66.6, 68.7, 69.1, 70.6, 70.9, 71.6, 72.6, 72.7, 76.2, 80.0, 100.6, 101.0, 122.6, 144.6, 155.8, 169.0, 169.6, 170.0−170.3, 172.8. Anal. Calcd for $C_{46}H_{69}N_5O_{23}S$: C, 50.59; H, 6.37; N, 6.41; S, 2.94. Found: C, 50.24; H, 6.23; N, 6.25; S, 2.79.

Characterization data of compound **20**: mp = 135−140 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.1) = +10.12. IR (cm^{-1}): 1751, 1654. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.36 (m, 2 H), 1.62 (m, 4 H), 1.90, 1.96, 2.03, 2.04, 2.06, 2.08, 2.15 (7s, 21 H), 2.19 (t, 2 H, *J* = 7.40 Hz), 2.98 (t, 2 H, *J* = 6.19 Hz), 3.09 (t, 2 H, *J* = 6.19 Hz), 3.43 (q, 2 H, *J* = 6.61, 12.81 Hz), 3.58 (m, 1 H), 3.70 (s, 3 H), 3.74−3.94 (m, 3 H), 4.01−4.15 (m, 4 H), 4.48 (t, 3 H, *J* = 6.19 Hz), 4.76−5.04 (m, 3 H), 5.04−5.22 (m, 2 H), 5.35 (d, 1 H, *J* = 2.99 Hz), 5.53 (bs, 2 H), 6.02 (d, 1 H, *J* = 7.26 Hz), 6.63 (d, 1 H, *J* = 5.00 Hz), 7.07 (m, 2 H), 7.27 (m, 3 H), 7.29 (d, 2 H, *J* = 8.31 Hz), 7.35 (s, 1 H), 7.80 (d, 2 H, *J* = 8.30 Hz). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 20.2−20.6, 24.5, 25.9, 26.1, 28.7, 35.6, 37.5, 38.3, 39.4, 52.0, 52.9, 53.2, 60.6, 61.7, 66.4, 68.4, 68.9, 70.3, 70.7, 71.3, 72.4, 75.9, 100.2, 100.7, 122.2, 126.7−129.0, 134.7, 135.8, 137.9, 144.8, 166.6, 168.9, 169.5, 169.6, 169.8, 169.9, 170.1, 170.2, 172.0, 172.6. Anal. Calcd for $C_{54}H_{69}N_5O_{22}$: C, 56.89; H, 6.10; N, 6.14. Found: C, 56.64; H, 6.47; N, 5.99.

Characterization data of compound **21**: mp = 78−84 °C; $[\alpha]_D$ ($CHCl_3$, *c* 1.1) = +2.1. IR (cm^{-1}): 1710, 1670. ^1H NMR ($CDCl_3$, 200 MHz): δ 1.26 (m, 2 H), 1.28, 1.44 (2s, 6 H), 1.40 (s, 9 H), 1.60 (m, 4 H), 2.30 (t, 2 H, *J* = 7.08 Hz), 2.50 (dd, 1 H, *J* = 5.37, 12.59 Hz), 2.68 (dd, 1 H, *J* = 7.04, 12.96 Hz), 3.18 (q, 2 H, *J* = 6.22, 12.57 Hz), 3.87 (q, 1 H, *J* = 6.74, 12.78 Hz), 4.07 (m, 2 H), 4.45−4.88 (m, 4 H), 4.96 (q, 1 H, *J* = 7.05, 14.61 Hz), 5.20 (dd, 2 H, *J* = 12.89, 17.81 Hz), 5.96 (d, 1 H, *J* = 3.58 Hz), 6.21 (m, 1 H), 7.15−7.46 (m, 15 H), 7.74 (s, 1 H). ^{13}C NMR ($CDCl_3$, 50 MHz): δ 24.2, 25.9, 26.1, 26.7, 28.1−28.2, 28.8, 33.7, 33.8, 39.1, 48.6, 53.5, 57.3, 67.0, 74.2, 79.1, 80.3, 85.3, 105.0, 111.8, 125.0, 126.7−129.5, 142.8, 144.2−144.3, 155.29, 170.6, 173.2. Anal. Calcd for $C_{44}H_{55}N_5O_9S$: C, 63.67; H, 6.68; N, 8.44; S, 3.86. Found: C, 63.98; H, 6.95; N, 8.28; S, 3.63.

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Supporting Information Available: ^1H, ^{13}C, and DEPT NMR spectral charts of compounds **4**−**8** and **10**−**21**. This material is available free of charge via the Internet at http://pubs.acs.org.

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