A Convenient Synthesis of *S*-(α-D- and *S*-(β-D)-Glucopyranosyl-6-thiomaltodextrins

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(2,3-Di-*O*-acetyl-6-deoxy-6-iodo)-hexakis(2,3,6-tri-*O*-acetyl)cyclomaltoheptaose **(9)** was prepared by reaction of cyclomaltoheptaose **(5)** with trityl chloride followed by acetylation, *O*-detritylation, mesylation, and displacement of the mesyloxy group by iodine using sodium iodide. This cyclomaltoheptaose derivative was *S*-glycosylated in high yield with either of the anomers of 2,3,4,6-tetra-*O*-acetyl-*S*-acetyl-1-thioglucopyranose (**1α** or **1β**). Deacetylation afforded *S*-(α-D- and *S*-(β-D-glucopyranosyl)-6-thiocyclomaltoheptaose (**10b**) and **11b**, respectively. Procedures arising from model experiments also provided a new syntheses of *S*-(α-D-glucopyranosyl)-6-thiomaltose (**3b**) and of its β-isomer (**4b**).

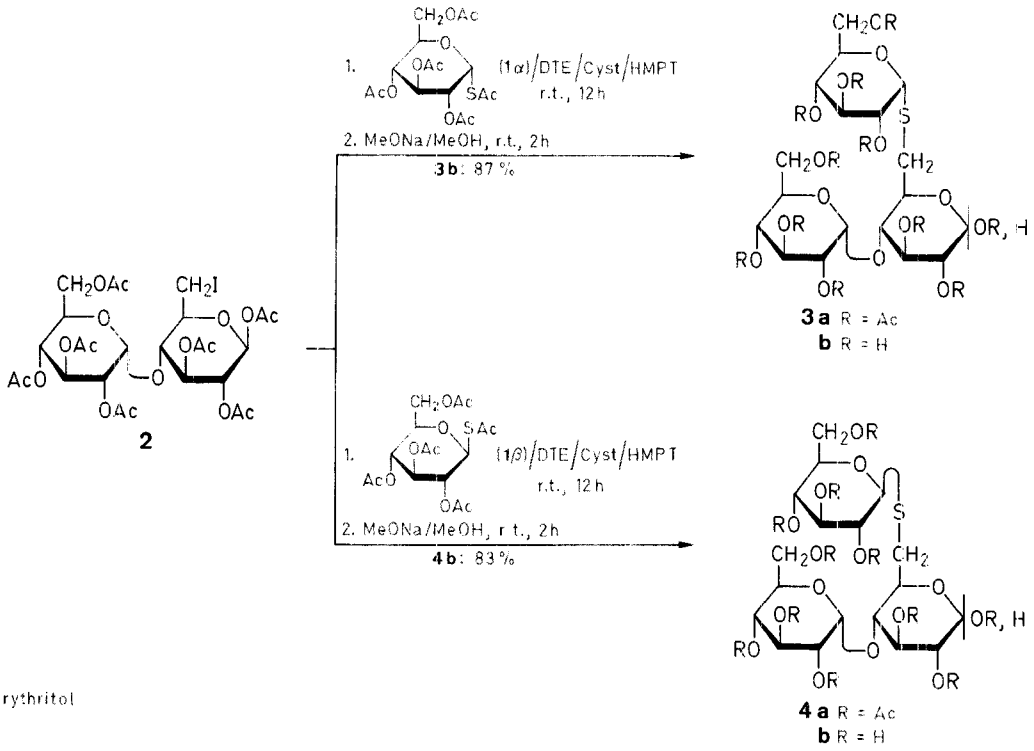
Cyclodextrins branched at the 6-position(s) with α-D-glucopyranosyl unit(s) were first discovered by French et al.[1] and have recently been obtained enzymatically[2] and chemically.[3,4] However, there is no report of branched non-natural products. The presence of a glycosylthio moiety on these cyclodextrins was of interest in our studies of amylose-degrading enzyme by the substrate–analog approach. Compounds **10b** and **11b** were obtained in good overall yield by application of a recently developed *S*-glycosylation method[5] and the synthesis of partially protected cyclomaltoheptaoses **6–9**.

The general strategy for the synthesis of oligosaccharides containing an *S*-atom as the interglycosidic linkage is as follows. A good leaving group, generally a triflate, on a glycosyl residue was replaced by a 1-glycosylthio group via an S_N2-type mechanism. This reaction was diastereospecific, the anomeric orientation was preserved, and inversion occurred at the interglycosidic carbon. The thiol can then be activated for condensation by total deacetylation of the molecule with sodium methoxide[6] or by treatment of β-D-isomer of the free thiol with sodium hydride.[7] We have developed a new and efficient procedure for this type of condensation in which a selective *in situ* *S*-deacetylation and activation occured by treatment of the fully acetylated thio compound with cysteamine (cyst.) in hexamethylphosphortriamide (HMPT) in the presence of 1,4-dithioerythritol (DTE).[5] However, the yield obtained from this condensation may be influenced by, the counterion of the thiolate, the leaving group, and the solvent. For these reasons, the condensation of fully acetylated 1-thioglucose (**1α** or **1β**) with maltose derivatives substituted at C-6 with various leaving groups was investigated. When acetylated 6-iodomaltose **(2)** was used under the above conditions, the expected products **3a** or **4a** were obtained in high yields (87 and 83%, respectively) (Scheme **A**). Compounds **3b** and **4b** were then obtained by catalytic deacetylation. These results suggested that this approach could be successfully applied to the synthesis of branched cyclomaltoheptaose derivatives.

The readily available cyclomaltoheptaose derivative **6** was treated with aqueous tetrafluoroboric acid to give the required partially protected cyclomaltoheptaose derivative **7** having only one unprotected hydroxy group (Scheme **B**). The overall yield from the parent compound **5** was 37%, this protection–deprotection sequence being more efficient than the best method hitherto described for this compound.[3] The iodo compound **9** was synthesized from **7** in higher yield by a two step procedure than by direct substitution of the hydroxy group.[8] Treatment of

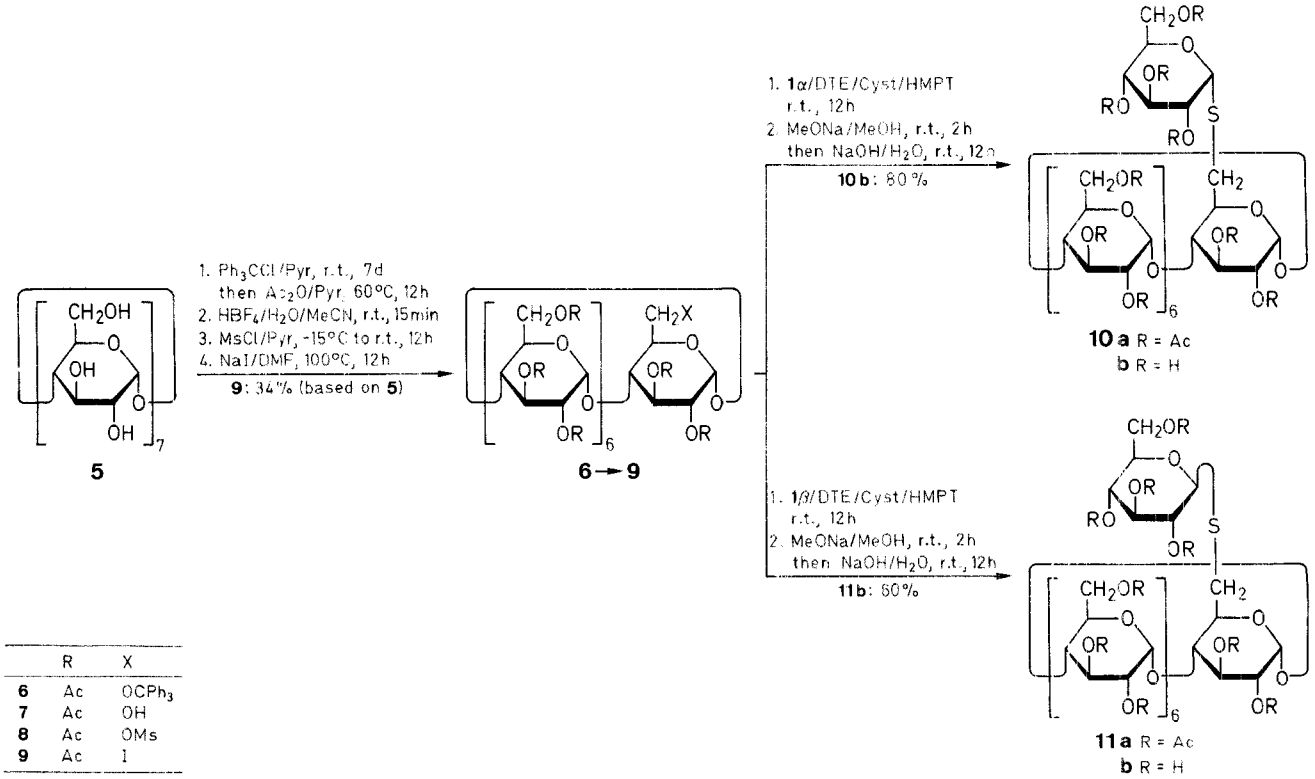
compound **7** with mesyl chloride led to the *O*-mesyl derivative **8** in 92% yield. Displacement of the mesyloxy group by iodine using sodium iodide in dimethylformamide gave the iodo compound **9** in 87% yield. Under the described conditions, compound **9** reacted with peracetylated 1-thio-α-D-glucopyranose (**1α**) or 1-thio-β-D-glucopyranose (**1β**) at room temperature (2 h) to give the expected compounds **10a** or **11a** in 82 and 63% yields, respectively. Deacetylation then led to the free compounds **10b** and **11b**.

The ^{13}C-NMR spectra of compounds **3a, b, 4a, b, 10a, b**, and **11a, b** are in good accord with the assigned structures. However, certain aspects are worthy of special comment. As seen in the Table, a difference in the chemical shift occurs between the *O*- and *S*-series, but also in the same series between the linear and the cyclic compounds. The introduction of an α- or β-glucopyranosylthio group at the 6-position of a cyclodextrin causes deshielding of the intersugar carbon of the ring (C-4) by $\delta = 2.4$ and 3.2, respectively. These assignments are substantially lower than those found in the *O*-series (about $\delta = 2$-3). The



DTE = 1,4-dithioerythritol

Scheme A



	R	X
6	Ac	$OCPh_3$
7	Ac	OH
8	Ac	OMs
9	Ac	I

Scheme B

same effect is observed for the trisaccharides listed in the Table; it can plausibly be rationalized as arising from a γ-shift due to the S-atom at C-6 of a glycosyl residue.

Table. ^{13}C-NMR Data for 6-Substituted Maltose and Cyclomaltoheptaose in the *S*- and *O*-Series; δ

Product	3b	4b	10b	11b	12[a]	13[b]	14[c]	15[d]
C-4	81.2[e]	81.1[e]	84.4	85.0	78.7	79.4[e]	82.0	81.8
C-1''[e]	85.5[e]	86.2	85.9	87.1	99.2	103.4[e]	99.6	103.3

[a] **12:** Methyl 6-*O*-α-D-glucopyranosyl-β-maltoside, data obtained from Ref. 9.

[b] **13:** 6-*O*-(β-D-Glucopyranosyl)maltose obtained by the Koenigs-Knorr method in 90% yield using acetobromoglucose and 1,2,2',3,3',4',6-hepta-*O*-acetyl-β-maltose.[10] The physical constants are identical with those reported.[11]

[c] **14:** 6-*O*-(α-D-Glucopyranosyl)cyclomaltoheptaose, data obtained from Ref. 3.

[d] **15:** 6-*O*-(β-D-Glucopyranosyl)cyclomaltoheptaose obtained in low yield by the Koenigs-Knorr method using acetobromoglucose and compound **7**. An improved synthesis will be reported elsewhere.

[e] Carbons of the α-anomer of the reducing unit.

Commercial compounds were used without purification, except where otherwise stated. Cyclomaltoheptaose and dithioerythritol were purchased from Sigma; cysteamine was purchased from Fluka Chemical Co.

During work-up, the aqueous-washed organic phase is dried (Na_2SO_4) and evaporated under reduced pressure at temperature below 45°C, and the aqueous washings are then back-extracted with $CHCl_3$. TLC is performed on silica gel (Merck F 254, Merck, Darmstadt, Germany) with detection by UV light and/or by charring with H_2SO_4/MeOH/H_2O. For acetylated compounds, chromatographic purifications are performed under pressure on dry Kieselgel 60 (230–400 Mesh) columns. The following eluent systems are used (v/v): (A) EtOAc/hexane (1:1); (B) acetone/hexane (1:1.5), (C) acetone/hexane (1:1); (D) acetone/hexane (1.5:1).

For the hydroxy-free compounds chromatographic filtrations are achieved on reversed-phase silica (C-18) under pressure (3 bar) with 50% aqueous MeOH. Optical rotations were determined with a Perkin Elmer polarimeter. Mass spectra were recorded on a Nermag R 10-10-C using the positive fast-atom bombardment (FAB+) technique. ^{13}C-NMR spectra were recorded at 25.18 MHz on a Bruker WP 100 and at 75.412 MHz on a Bruker AM 300 for the glycosylated cyclodextrins. Chemical shifts are given in δ from acetone as internal reference for D_2O solution or TMS for $CDCl_3$ solution. Primed numbers refer to the carbon of the glucosyl α (1 to 4) linked and double-primed numbers of the glucosyl α or β (1 to 6) linked (side chain). The numbers of the glucosyl bearing the side chain are not primed.

1,2,3-Tri-*O*-acetyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-6-*S*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-6-thio-β-D-glucopyranose (3a):

Cysteamine (20 mg) is added to a stirred mixture of 6-iodomaltose[12] (**2**; 106 mg, 0.14 mmol), tetra-*O*-acetyl-*S*-acetyl-1-thio-α-D-glucose[13] (**1α**; 75 mg, 0.10 mmol), and dithioerytritol (30 mg) in HMPT (3 mL). After 12 h (room temperature), ice-water (10 mL), is added to the mixture. The precipitate is isolated by filtration through a bed of celite and washed with H_2O. Column chromatography (eluent A) affords the branched trisaccharide **3a**; yield: 180 mg (87%); $[\alpha]_D^{25}$ + 132° (c = 2.5, $CHCl_3$).

$C_{40}H_{54}O_{26}S$	calc.	C 48.87	H 5.53	S 3.26
(982.9)	found	48.34	5.55	3.53

^{13}C-NMR ($CDCl_3$/TMS): δ = 30.3 (C-6); 61.5, 61.7 (C-6', C-6''); 68.0–77.4 (C-2, C-3, C-4, C-5, C-2', C-3', C-4', C-5', C-2'', C-3'', C-4'', C-5''); 83.2 (C-1''); 91.1 (C-1); 95.6 (C-1').

1,2,3-Tri-*O*-acetyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-6-*S*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-6-thio-β-D-glucopyranose (4a):

Under the experimental conditions given above but starting from tetra-*O*-acetyl-*S*-acetyl-1-thio-β-D-glucose[14] (**1β**; 90 mg, 0.22 mmol), compound **4a** is obtained; yield: 115 mg (83%); $[\alpha]_D^{25}$ + 22° (c = 1.5, $CHCl_3$).

$C_{40}H_{54}O_{26}S$	calc.	C 48.87	H 5.53	S 3.26
(982.9)	found	48.77	5.77	3.23

^{13}C-NMR ($CDCl_3$/TMS): δ = 31.3 (C-6); 61.7 (C-6', C-6''); 68.1–76.1 (C-2, C-3, C-4, C-5, C-2', C-3', C-4', C-5', C-2'', C-3'', C-4'', C-5''); 83.0 (C-1''); 91.2 (C-1); 95.7 (C-1').

4-*O*-α-D-Glucopyranosyl-6-*S*-α-D-glucopyranosyl-6-thio-D-glucopyranose (3b):

A solution of compound **3a** (90 mg, 0.09 mmol) in MeOH (10 mL) containing NaOMe in MeOH (1 M, 100 µL) is stirred for 2 h at r.t., then neutralized with Amberlite IR 77-N (H^+), filtered, and concentrated to dryness. Freeze-drying of an aqueous solution of the residue affords **3b** as a foam; yield: 47 mg (100%); $[\alpha]_D^{25}$ + 211° (c = 1, H_2O).

$C_{18}H_{32}O_{15}S.H_2O$	calc.	C 40.15	H 6.36	S 5.95
(538.5)	found	40.05	6.35	5.85

^{13}C-NMR (D_2O/acetone): δ = 31.8 (C-6β); 32.0 (C-6α); 61.6 (C-6', C-6''); 69.9–76.1 (C-2, C-3, C-5, C-2', C-3', C-4', C-5', C-2'', C-3'', C-4'', C-5''); 80.7 (C-4β); 81.2 (C-4α); 85.5 (C-1''α); 85.6 (C-1''β); 92.0 (C-1α); 96.0 (C-1β); 100.8 (C-1').

4-*O*-α-D-Glucopyranosyl-6-*S*-β-D-glucopyranosyl-6-thio-D-glucopyranose (4b):

Compound **4a** (90 mg, 0.09 mmol) is deacetylated as above to afford **4b**: yield: 47 mg (100%); $[\alpha]_D^{25}$ + 79° (c = 0.8, H_2O).

$C_{18}H_{32}O_{15}S.5/2\,H_2O$	calc.	C 38.22	H 6.59	S 5.66
(565.5)	found	38.02	6.33	5.15

^{13}C-NMR (D_2O/acetone): δ = 32.0 (C-6β); 32.2 (C-6α); 60.7–61.0 (C-6', C-6''); 69.5–76.0 (C-2, C-3, C-5, C-2', C-3', C-4', C-5', C-2'', C-4''); 77.3 (C-3''); 79.9 (C-5''); 80.9 (C-4β); 81.1 (C-4α; 86.2 (C-1''); 93.3 (C-1α); 96.2 (C-1β); 100.0 (C-1').

(2,3-Di-*O*-acetyl-6-*O*-trityl)hexakis(2,3,6-tri-*O*-acetyl)cyclomaltoheptaose (6):

Freeze-dried cyclomaltoheptaose (**5**; 9.25 g, 8.16 mmol) is dissolved in dry pyridine (450 mL). To remove the trapped H_2O and ensure dryness, the solution is concentrated (to 400 mL) by distillation at atmospheric pressure. It is then cooled to r.t., trityl chloride (1.58 g, 5.71 mmol) is added, and the solution is stirred for 5 days; then, further trityl chloride (0.68 g, 2.45 mmol) is added and the solution is stirred for 2 days. After concentration (to 70 mL) under reduced pressure, the mixture is acetylated (60°C, 12 h) with Ac_2O (30 mL) in the presence of DMAP (100 mg). The product is precipitated with ice (300 g), isolated by suction, dissolved in $CHCl_3$ (300 mL), washed with ice-cold H_2O (500 mL), and purified by chromatography using eluent B to give product **6** as a solid foam; yield: 8.84 g (49%); $[\alpha]_D^{25}$ + 110° (c = 0.65, $CHCl_3$).

$C_{101}H_{124}O_{55}$	calc.	C 54.49	H 5.63
(2218.1)	found	54.31	5.57

(2,3-Di-*O*-acetyl)hexakis(2,3,6-tri-*O*-acetyl)cyclomaltoheptaose (7):

Tetrafluoroboric acid (35% in H_2O; 0.5 mL, 2.5 mmol) is added to a stirred solution of compound **6** (2.3 g, 1 mmol) in MeCN (50 mL) and stirring is continued for 15 min at r.t. (quantitative transformation). Then, Et_3N (0.5 mL) is added, and the solvent is evaporated. The product is purified by column chromatography using eluent C to give **7** as a solid foam; yield: 1.5 g (76%). Physical data are as described in Lit.[3]: $[\alpha]_D^{25}$ + 116° (c = 1, $CHCl_3$); Lit.[3] $[\alpha]_D^{25}$ + 107° (c = 0.5, $CHCl_3$).

(2,3-Di-*O*-acetyl-6-*O*-mesyl)hexakis(2,3,6-tri-*O*-acetyl)cyclomaltoheptaose (8):

To a stirred solution of compound **7** (980 mg, 0.5 mmol) in pyridine (15 mL) at −15°C is added mesyl chloride (170 µL, 1 mmol). The temperature is slowly raised to r.t. After 12 h, ice (50 g) is added to the mixture and the precipitated product is isolated by suction and washed with H_2O. Column chromatography using eluent C gives the mesyl derivative **8**; yield: 940 mg (92%); $[\alpha]_D^{25}$ + 114° (c = 1, $CHCl_3$).

$C_{83}H_{112}O_{57}S$	calc.	C 48.53	H 5.49	S 1.56
(2053.8)	found	48.62	5.30	1.72

(2,3-Di-*O*-acetyl-6-deoxy-6-iodo)hexakis(2,3,6-tri-*O*-acetyl)cyclomaltoheptaose (9):
Compound **8** (465 mg, 0.23 mmol) is dissolved in DMF (10 mL) and NaI (150 mg) is added. After 12 h at 100°C, mixture is evaporated under reduced pressure. The residue is dissolved in $CHCl_3$ (50 mL), and washed with H_2O; and purified by column chromatography (eluent C) to give **9** as a solid foam; yield: 410 mg (87%); $[\alpha]_D^{25}$ + 109° (*c* = 1, $CHCl_3$).

$C_{82}H_{109}IO_{54}$	calc.	C 47.22	H 5.26	I 6.08
(2085.6)	found	47.75	5.48	4.74

MS (FAB): *m/z* = 2086 (M + 1).

[2,3-Di-*O*-acetyl-6-*S*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)]hexakis (2,3,6-tri-*O*-acetyl)-6-thiocyclomaltoheptaose (10a):
Cysteamine (15 mg) is added to a stirred mixture of the iodocyclodextrin **9** (220 mg, 0.1 mmol), tetra-*O*-acetyl-*S*-acetyl-1-thio-α-D-glucose (**1α**; 100 mg, 0.24 mmol) and dithioerytritol (30 mg) in HMPA (1 mL). Work-up as described for **3a** and column chromatography (eluent D) affords the branched cyclodextrin **10a**; yield: 190 mg (82%); $[\alpha]_D^{25}$ + 135° (*c* = 1, $CHCl_3$).

$C_{96}H_{128}O_{63}S$	calc.	C 49.65	H 5.55	S 1.38
(2322.1)	found	49.50	5.80	1.48

^{13}C-NMR ($CDCl_3$/TMS): δ = 29.6 (C-6); 61.5 (C-6″); 62.4, 62.7 (C-6′); 83.2 (C-1″); 96.5–97.1 (C-1, C-1′).

[2,3-Di-*O*-acetyl-*S*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)]hexakis(2,3,6-tri-*O*-acetyl)-6-thiocyclomaltoheptaose (11a):
Under the experimental conditions given above but starting from tetra-*O*-acetyl-*S*-acetyl-1-thio-β-D-glucose (**1β**; 100 mg 0.24 mmol), compound **11a** is obtained; yield: 175 mg (63%); $[\alpha]_D^{23}$ + 94° (*c* = 1, $CHCl_3$).

$C_{96}H_{128}O_{63}S$	calc.	C 49.65	H 5.55	S 1.38
(2322.1)	found	49.56	5.46	1.41

^{13}C-NMR ($CDCl_3$/TMS): δ = 29.7 (C-6); 61.5 (C-6″); 62.2–62.4 (C-6′); 83.0 (C-1″); 96.4–97.0 (C-1, C-1′).

***S*-(α-D-Glucopyranosyl)-6-thiocyclomaltoheptaose (10b):**
The acetylated compound **10a** (90 mg, 39 μmol) is dissolved in MeOH (10 mL) and partially deacetylated by stirring with MeONa in MeOH (100 μL, 1 M) at r.t. After 2 h, the deposited solid is dissolved by the addition of H_2O (5 mL), and aqueous NaOH (1 M, 1 mL) is added. After 12 h at r.t., the mixture is deionized with Amberlite IRN-77 (H^+) resin, filtered, concentrated, and freeze-dried to give **10b**; yield: 50 mg (98%); $[\alpha]_D^{23}$ + 153° (*c* = 1, H_2O)

$C_{48}H_{80}O_{39}S.7H_2O$	calc.	C 40.05	H 6.58	S 2.22
(1439.3)	found	40.01	6.19	2.14

MS (FAB): *m/z* = 1314 (M + 1).

^{13}C-NMR (D_2O/acetone): δ = 30.9 (C-6); 60.6–60.9 (C-6′, C-6″); 66.3 (C-4″); 69.9–73.9 (C-2, C-3, C-5, C-2′, C-3′, C-5′, C-2″, C-3″, C-5″); 81.5–81.6 (C-4′); 84.4 (C-4); 85.9 (C-1″); 102.0–102.3 (C-1, C-1′).

***S*-(β-D-Glucopyranosyl)-6-thiocyclomaltoheptaose (11b):**
Compound **11a** (85 mg, 36 μmol) is treated as above to afford **11b**; yield: 45 mg (95%); $[\alpha]_D^{25}$ + 110° (*c* = 1, H_2O).

$C_{48}H_{80}O_{39}S.9H_2O$	calc.	C 39.07	H 6.69	S 2.17
(1475.3)	found	39.04	6.14	1.82

MS (FAB): *m/z* = 1314 (M + 1).

^{13}C-NMR (D_2O/acetone): δ = 32.7 (C-6); 60.6–61.2 (C-6′, C-6″); 69.8 (C-4″); 72.0–73.5 (C-2, C-3, C-5, C-2′, C-3′, C-5′, C-2″); 77.6 (C-3″); 80.1 (C-5″); 81.5–81.7 (C-4′); 85.0 (C-4); 87.1 (C-1″); 102.0–102.3 (C-1, C-1′).

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