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Synthesis and characterization of cyclomaltoheptaose-based metal chelants as probes for intestinal permeability

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Abstract

The syntheses of two cyclomaltoheptaose-based metal chelants, cyclomaltoheptaose-ethylenediaminetetraacetate (CD-EDTA) and cyclomaltoheptaose-diamide-disulfur (CD-DADS), are described. The chelant moieties are attached to the 6-position of a single pyranose in the cyclomaltoheptaose via a short diamine spacer chain. Characterization of these novel chelants has been achieved using NMR and MS techniques. The peculiar fluxional properties of the CD-EDTA molecules is also discussed.

Keywords: Cyclomaltoheptaose; β-Cyclodextrin; Monosubstituted derivatives; Ethylenediaminetetraacetate; Cyclic disulfide

1. Introduction

Studies concerning intestinal permeability in health and disease have generally focused on the relationship between the observed urinary excretion of orally administered probes and the molecular dimension and charge of these molecules. It has been suggested that the absorption of probes of different sizes proceeds via disparate permeation pathways with smaller molecules crossing the epithelial barrier via transcellular routes, whereas larger probes cross via intercellular avenues [1]. Clinical measurements have been carried out using probes in the 100–500 dalton range [rhamnose [2], mannitol [3], lactulose [4], ethylenediaminetetraacetate [5], and polyethylene glycol (PEG 400) [6]] or on larger molecules with molecular weights greater than 10 000 daltons [polyvinylpyrrolidine (PVP)

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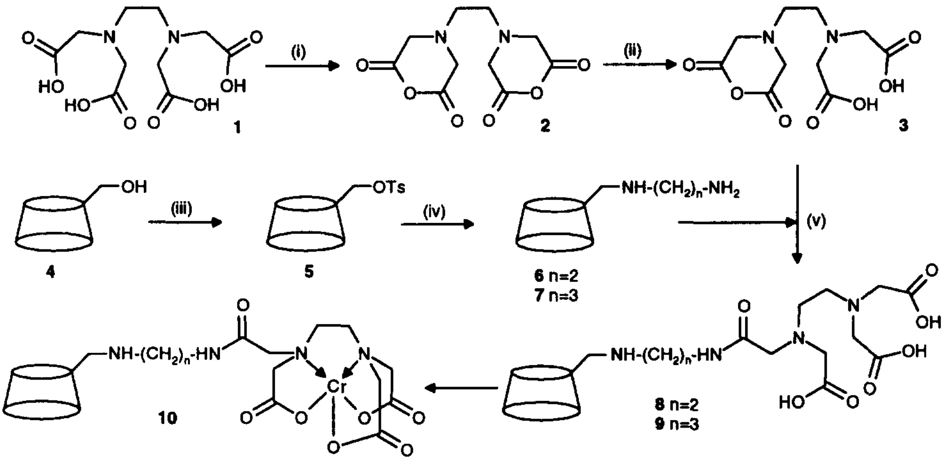

Scheme 1. Reagents and conditions: (i) Ac_2O, pyridine, 65°C, 24 h; (ii) 1 equiv H_2O, DMF, 80°C, 3 h; (iii) TsCl, pyridine, 0°C, 24 h; (iv) NH_2-$(CH_2)_n$-NH_2 (excess), 70°C, 24 h; (v) pyridine, room temperature, 24 h.

[7] and albumin [8]]. The desire for a probe intermediate in size to those studied thus far has lead to the synthesis of cyclomaltoheptaose-based probes.

The cyclomaltoses (cyclodextrins) are a family of toroidal shaped molecules having molecular weights of ca. 1300 daltons [9]. The unusual size and shape of these dextrins make them unique candidates for intestinal permeability probes. Efforts have been directed towards the syntheses of cyclomaltoheptaoses (the cyclomaltose containing seven cyclically linked D-glucose units) equipped with a metal chelating moiety, and the present work describes the preparation of two such cyclomaltoheptaose derivatives.

2. Results and discussion

Cyclomaltoheptaose-EDTA Complexes.—Chromium-51 labelled ethylenediaminetetraacetate (EDTA) has been used diagnostically to detect alterations in the intestinal permeability of patients afflicted with inflammatory colonic disease [5]. The successful application of such complexes has led us to develop the syntheses of cyclomaltoheptaose-EDTA derivatives (**8** and **9**, see Scheme 1). The EDTA fragment of **8** or **9** will serve as the chelator for chromium-51, effectively labelling the probe **10**, and allowing its progress through the intestine to be monitored.

The key intermediate in the syntheses of the desired cyclomaltoheptaose-EDTA targets is the monosulfonylated cyclomaltoheptaose **5** (Scheme 1). Since all the hydroxyls of cyclomaltoheptaose (shown in caricature as **4** in Scheme 1) can potentially undergo modification (the primary sites to a greater extent than the secondary ones), the desired monosubstituted product is often contaminated with polysulfonylated cyclomaltoses. Accounts in the literature [10] describing tosylation reactions using an excess of cyclomaltose also report the formation of polytosylated byproducts. Purification of these mixtures involved repeated recrystallizations and, ultimately, resulted in less than impressive yields.

A more efficient preparation of **5** was developed involving the reaction of one equivalent of *p*-toluenesulfonyl (tosyl) chloride with one equivalent of cyclomaltoheptaose and purification of the mixture using liquid chromatography under the conditions outlined by Fujita et al. [11]. This procedure resulted in a 43% overall yield of the monotosylated product as established by ^{1}H NMR spectroscopy [1] and by fast-atom bombardment mass spectrometry (FABMS).

In order to permit attachment of the EDTA moiety, transformation of the tosylated cyclomaltoheptaose into an amino-cyclomaltoheptaose was then carried out. The amino-cyclomaltoheptaose would ultimately be reacted with an activated EDTA allowing the two fragments to be linked together through an amide bond. The most obvious route to an amino-cyclomaltoheptaose would involve the displacement of the tosylate of **5** with azide and subsequent reduction to provide an amine (for an example of this approach see [12]). The approach taken in the present work, though, involved the preparation of diamino-cyclomaltoheptaoses (see Scheme 1, compounds **6** or **7**). Diamino-cyclomaltoheptaoses were found to be easily prepared and avoided the cumbersome purification of the two-step, azide-amine route. Furthermore, separating the cyclomaltoheptaose and the metal chelating moiety with an aliphatic spacer chain will ensure that permeation through the intestine will be a function of the cyclomaltoheptaose size only.

Both 6-aminoethylamino-6-deoxycyclomaltoheptaose **6** [10] and 6-aminopropylamino-6-deoxycyclomaltoheptaose **7** (Scheme 1) were prepared by reacting the monotosylated cyclomaltoheptaose **5** with an excess of the appropriate diamine. An excess of diamine precluded the formation of a bis-cyclomaltoheptaose product wherein the diamine would be substituted at both ends. Isolation of the desired diamino-cyclomaltoheptaose involved removal of the unreacted diamine by evaporation and adding the residue to 2-propanol. This allowed for the precipitation of an inclusion complex that was collected and dried under high vacuum to remove the 2-propanol and afford pure cyclomaltoheptaose-diamine.

FAB mass spectrometry demonstrated the success of the tosylate displacement reactions showing an $[M+1]$ ion at m/z 1178 and 1192 for **6** and **7**, respectively. The ^{1}H NMR spectra for the diamines were found to be quite congested, difficult to interpret and, ultimately, failed to provide definitive evidence for diamine formation or for purity. On the other hand, the ^{13}C NMR spectra clearly affirmed amination showing a shift of the substituted cyclomaltoheptaose C-6′ carbon from 69.74 ppm in the tosylate to 50.02 ppm in **6** and 50.53 ppm in **7**. The disappearance of the tosylate signals and the appearance of the aliphatic spacer chain signals at 40.40 and 50.22 ppm in **6** and 29.53, 39.33, and 47.47 ppm in **7** provided additional proof of diamine formation.

Coupling of the cyclomaltoheptaose-diamine to an EDTA fragment was achieved using the activated EDTA synthon **3**. Following the method described by Geigy [13], treatment of EDTA **1** with acetic anhydride in pyridine resulted in the formation of EDTA dianhydride (**2**). Hydrolytic opening of only one of the cyclic anhydrides in **2** allowed for the preparation of EDTA monoanhydride (**3**). The hydrolysis reaction, developed by Takeshita et al. [14], takes advantage of the differing solubilities of the monoanhydride and dianhydride in *N,N*-dimethylformamide (Me_2NCHO) at elevated and room temperatures. Each EDTA derivative has been characterized [14].

Dropwise addition of the EDTA monoanhydride in pyridine to a solution of cyclomaltoheptaose-diamine led to the formation of the cyclomaltoheptaose-EDTA derivatives (**8**

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[1] By comparing the relative area of integration for the tosylate protons and the signal for the anomeric protons on the cyclomaltoheptaose in the ^{1}H NMR spectrum of **5**, monofunctionalization was confirmed.

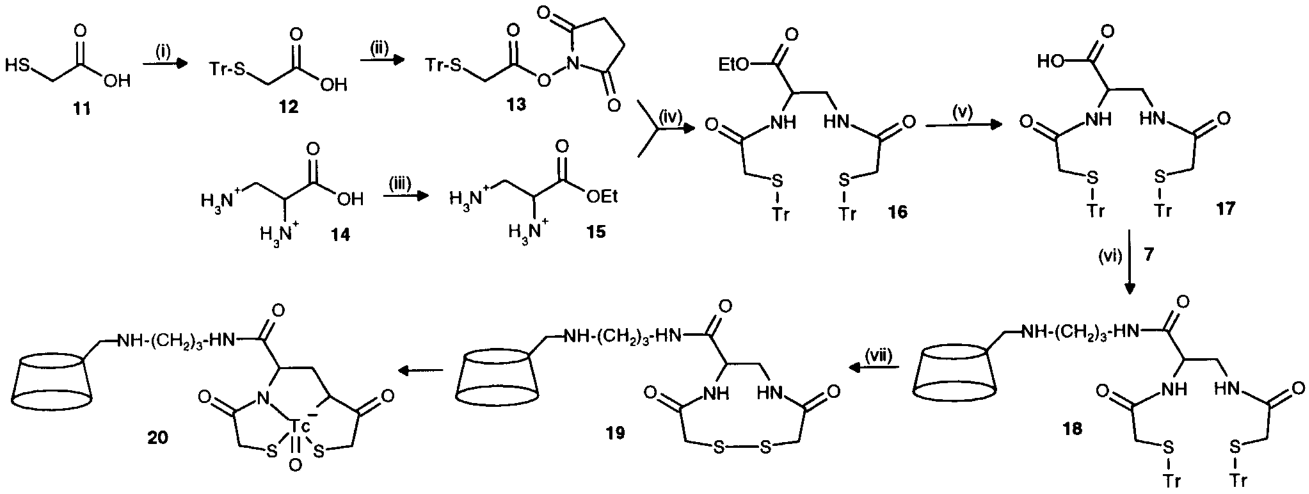
and **9**, Scheme 1). The increased steric congestion about the 2° amine site in comparison to the 1° amine site and the greater nucleophilicity of the 1° amine over the cyclomaltoheptaose hydroxyl groups in **6** and **7** ensured that **8** and **9**, respectively, were the only products from the reaction. Purification was facilitated by size-exclusion filtration using Amicon^{TM} filters. By choosing a filter with a molecular weight cut-off of ca. 1000 daltons, the cyclomaltoheptaose-EDTA adduct could be separated from any unreacted diamine, pyridine, or EDTA produced from adventitious hydrolysis.

The purified cyclomaltoheptaose-EDTA molecules gave variable C and H analyses, largely because residual water could not be removed without inducing some decomposition. They were characterized by TLC and FABMS which showed molecular ions at m/z 1451 and 1465 for **8** and **9**, respectively, while ^{13}C NMR spectroscopy showed the newly formed NH*C*O signal at 177.95 ppm and 179.59 for **8** and **9**, respectively. Curiously, the ^{13}C NMR spectroscopy carried out on the cyclomaltoheptaose-EDTA adducts revealed the presence of additional resonances which were unexpected given the apparent single identity of the compounds as established by TLC and FAB mass spectrometry. Careful inspection of the ^{13}C NMR spectra of **8** and **9** showed that for both of these compounds there were two distinguishable conformations present, of approximately equal energy, the interconversion of which was slow on the NMR time scale at room temperature. Normally secondary amides such as **8** or **9** exist exclusively in the *Z* (or *s-cis*) conformation with no contribution from the *E* (or *s-trans*) form. The presence of an open-chain (and presumably charged) form of the EDTA fragment appeared necessary to stabilize the second *E* conformer because the addition of one equivalent of Ca^{2+} to a sample of **9** resulted in a ^{13}C NMR spectrum wherein the additional signals disappeared. The chelation of Ca^{2+} by the EDTA moiety effectively removed any possibility of interaction between a mobile carboxylate ion and either the protonated secondary amine adjacent to the cyclomaltoheptaose ring or possibly the amide function and thus destroyed stabilization of the second conformer. Additionally, it was noted that the ^{13}C NMR spectrum of the cyclomaltoheptaose-DADS intermediate, **18**, (*vide infra*) showed the presence of only one conformation, further implicating the EDTA fragment as the source of the conformational differences.

Cyclomaltoheptaose-DADS complexes.—The desire to image the intestinal areas through which the cyclomaltoheptaose probe permeates, has led us to develop a technetium-chelating cyclomaltoheptaose. Technetium-99m is the radionuclide of choice for diagnostic imaging [15] and has been used previously in the study of intestinal diseases [16]. The tetradentate diamide-disulfur (DADS) ligand [17] has been shown to form highly stable, well-characterized complexes with Tc and is, therefore, an attractive chelant for the development of Tc based radiopharmaceuticals. Consequently, the Tc-99m DADS complex was chosen as a radiolabel carrier for cyclomaltoheptaose. The synthesis of the cyclomaltoheptaose-DADS derivative **19** is shown in Scheme 2.

The metal-chelating moiety is derived from two molecules of mercaptoacetic acid (**11**) and one molecule of (±)-2,3-diaminopropanoic acid (**14**). Protection of the thiol in **11** was deemed necessary since the free functionality would interfere with the planned carboxyl activation necessary for the attachment to cyclomaltoheptaose-diamine. A variety of protecting groups were considered, and the triphenylmethyl group was chosen.

Tritylation of mercaptoacetic acid under Lewis acid catalysis readily afforded the *S*-protected acid **12**, which was then treated with *N*-hydroxysuccinimide and 1,3-dicyclo-



Scheme 2. Reagents and conditions: (i) Ph_3COH, CH_3COOH, BF_3 · etherate, room temperature, 1 h; (ii) *N*-hydroxysuccinimide, DCC, DME, 5°C, 18 h; (iii) HCl, EtOH, 4 days; (iv) Et_3N, CH_3CN, room temperature, 36 h; (v) Amberlyst A-26 (OH^-); Aliquat 336, CH_2Cl_2, EtOH, room temperature, 48 h; (vi) EDC, DMF, Et_3N, room temperature, 48 h; (vii) I_2, EtOH, room temperature, 18 h.

hexylcarbodiimide (DCC) to give the activated ester **13**. Concurrently, ethyl (±)-2,3-diaminopropanoate dihydrochloride (**15**) was prepared employing standard Fischer esterification conditions. Treatment of two equivalents of the activated *N*-hydroxysuccinimidoyl ester **13** with the ethyl ester **15** in acetonitrile allowed for the acylation of the amino groups and the formation of the ethyl ester-diamide **16**.

The resultant ethyl ester-diamide **16** was hydrolyzed catalytically to the free acid **17** in 92% yield using a macrorecticular anion-exchange resin (in the hydroxide form) and an alkylammonium phase-transfer reagent. This novel method of hydrolysis doubled as an effective purification scheme for the removal of non-anionic impurities, particularly the troublesome dicyclohexylurea byproduct of the earlier DCC-mediated coupling reaction. The contaminants and the phase transfer catalyst were easily separated by washing the resin-bound carboxylate with ethanol and acetone. Elution of the acid from the resin was accomplished with 0.08 M hydrochloric acid affording **17** in 92% yield.

Coupling of the cyclomaltoheptaose-diaminopropane **7** with the *S*-trityl ligand **17** via 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide (EDC) reaction in DMF gave the amide adduct **18** in 53% yield. The ^{1}H NMR spectrum of **18** displayed a shift to lower field for the NHCOC*H* signal relative to the acid (δ 4.02 in **17** from δ 3.89 in **16**), while the ^{13}C NMR spectrum exhibited a third resonance in the carbonyl region at 167 ppm corresponding to the formation of an additional amide bond. The IR spectrum also showed no $C=O$ carboxylic acid stretching frequency in the range 1725–1740 cm^{-1}, providing further confirmation for amide construction. Compound **18** will exist as two diastereomers as the acid **17** is racemic and diamine **7** is a single enantiomer. However, no separation of the two diastereomers could be effected in a variety of TLC systems and there was no evidence from the ^{13}C NMR spectrum that the two species had differences in any of their chemical shifts. For practical purposes **18** was treated as a single entity. A similar situation was found with compound **19** (see below).

Early experiments attempting to couple the cyclomaltoheptaoses with the deprotected and cyclized disulfide from **17**, which is the more convergent approach from the synthetic viewpoint, gave poor and irreproducible yields, largely it appears because the cyclic disulfide has astonishingly low solubility in all solvents tried. The deprotection and cyclization step was therefore left to the final step in the synthesis.

The choice of the *S*-protecting group for the mercaptoacetic acid fragment may now be appreciated. Its selection was based upon the reagents and conditions required for the deprotection step converting **18** to **19**, and particularly the preservation the glycosidic linkages of the cyclomaltoheptaose and the avoidance of side reactions [18]. We noted that iodine has been used in the removal of trityl groups from sulfur with facile, in situ oxidation to disulfides. Moreover, disulfides are stable and easy to cleave with mild reductants (for example, $SnCl_2$) and as such represent attractive precursors to Tc chelants, particularly in a clinical environment.

Initial attempts to detritylate the cyclomaltoheptaose adduct **18** under standard deprotection conditions (employing 1.1 equiv of I_2) resulted in the recovery of ca. 35% of **18** after work-up. This unexpected result was attributed to the formation of a 1:1 guest–host complex between iodine and the cyclomaltoheptaose. A crystal structure of the iodine complex of cyclomaltohexaose has been reported and added support to this I_2 guest–host explanation [19]. As a result, deprotection of **18** was repeated using 2.1 equiv of I_2 in 20:80 dichloromethane–ethanol under conditions which strictly excluded oxygen. By maintaining high-dilution conditions to preclude intermolecular disulfide formation, in situ intramolecular oxidative cyclization prevailed and afforded the disulfide **19** in 34% yield.

Positive-ion FABMS analysis indicated a [M + 1] peak at m/z 1423 for **19**. Furthermore, the ^{1}H and ^{13}C NMR spectra confirmed the absence of the trityl group. Cyclization was also inferred from the marked signal broadening of all ^{13}C resonances in the 10-membered ligand heterocycle. This broadening phenomenon is characteristic of these cyclic systems and is indicative of an intermediate rate of exchange between different conformational isomers of the ring [20]. An alternative explanation is that, in addition to the conformational interconversions, the disulfide ring may be undergoing internal inclusion complex formation with the cyclomaltoheptaose ring. Molecular models do indeed suggest that the diamido-disulfide ring may insert into a cyclomaltoheptaose ring, but we have carried out no experiments to explore this intriguing possibility.

3. Conclusions

The preparative routes for cyclomaltoheptaose-ethylenediaminetetraacetate and cyclomaltoheptaose-diamide-disulfur complexes described above can be used to furnish substantial amounts of the dextrin-based chelants economically.

While biological testing of the cyclomaltoheptaose-DADS technetium complex **20** has yet to be carried out, preliminary studies using the cyclomaltoheptaose-EDTA chromium complex **10** have proved interesting. The complete results of the bio-testing will be published elsewhere.

4. Experimental

General methods.—Melting points were recorded on a Gallenkamp capillary tube melting point apparatus and are uncorrected. Proton magnetic resonance (^{1}H NMR) spectra were recorded on a Varian EM-390 (at 90 MHz), a Bruker AC 200 (at 200.13 MHz), or a Bruker AM 500 (at 500.13 MHz) spectrometer. The natural-abundance carbon-13 magnetic resonances (^{13}C NMR) were recorded on a Bruker AM 500 (at 125.776 MHz) or a Bruker AC 200 (at 50.324 MHz) instrument. All ^{13}C spectra were recorded with broad-band proton decoupling. Low-resolution mass spectra (MS) were obtained on VG ZAB-E mass spectrometer with samples being introduced through a direct inlet system. Ions were generated using fast-atom bombardment (FAB) (5% trifluoroacetic acid in thioglycerol matrix), electron impact (EI) or through desorption chemical ionization (DCI) with NH_3 as the reactant gas. Infrared spectra (IR) were recorded on a Bio-Rad FTS-40 or a Perkin–Elmer 283 spectrometer. *N,N*-Dimethylformamide (Me_2NCHO) was refluxed over and subsequently distilled from BaO. Acetonitrile was distilled from P_2O_5, followed by refluxing of the distillate from CaH_2, discarding the first 5% and the last 10%. Pyridine was refluxed over and subsequently distilled from CaH_2. Ethanol was dried and purified by distillation from magnesium turnings and stored over 4A molecular seives. Amberlyst A-26 anion-exchange resin (Cl^-) was prewashed and converted to the OH^- form just prior to use.

Commercial grade resin was washed successively with M HCl, deionized H_2O, 1:1 H_2O–acetone, EtOH, M NaOH, deionized H_2O (until the eluent tested neutral), and EtOH. The washed resin was stored in EtOH. The NMR solvents, chloroform-*d* ($CDCl_3$) and dimethyl sulfoxide-d_6 (Me_2SO-d_6), were stored over 4A molecular sieves prior to use. Silica Gel 60 F_{254} (E. Merck Co.) plates of 0.2-mm thickness were used for analytical thin-layer chromatography (TLC). Plates were developed in either solvent *A*, consisting of 7:7:5 EtOAc–2-propanol–H_2O; solvent *B*, composed of 7:7:5:4 EtOAc–2-propanol–ammonium hydroxide–H_2O; solvent *C*, composed of 1:9 MeOH–CH_2Cl_2; solvent *D*, composed of 2:98 MeOH–CH_2Cl_2, or solvent E, composed of 70:28:2 propanol–H_2O–formic acid. Visualization was achieved using a UV lamp at 254 nm (detection method A) or via treatment of the TLC plate with either a molybdic acid spray (20 g of molybdic acid and 15 g of ceric sulfate dissolved in 1 L of 10% H_2SO_4) (detection method B), a ninhydrin spray (0.2% ninhydrin in EtOH), followed by heating (detection method C) or a carbohydrate spray (9.2 mL of *m*-anisaldehyde in 338 mL of 95% EtOH, 3.75 mL of glacial AcOH, and 12.5 mL of concd H_2SO_4) (detection method D).

The normal numbering system for pyranose rings is employed in ^1H and ^{13}C NMR spectral assignments. In the cyclomaltoses, substituted ring atoms are indicated by a prime and adjacent rings by double primes.

Ethylenediaminetetraacetic acid dianhydride (*EDTA dianhydride*) (**2**).—The synthesis was performed according to the method described by Geigy [13]. To a suspension of ethylenediaminetetraacetic acid (EDTA, 18.0 g, 60.0 mmol) in pyridine (31 mL) under N_2 was added Ac_2O (24 mL). The mixture was stirred at 65°C for 24 h at which time the mixture was cooled and filtered under a steady stream of N_2. The white solid collected was washed with Ac_2O (200 mL) and diethyl ether (200 mL) and dried under high vacuum. The yield was 94% (14.4 g, 56.4 mmol); mp 194–195°C (dec); IR (ν_{max}, cm^{-1}): 1770 (s, anhydrides), 2900 (w); ^1H NMR (Me_2SO-d_6, 500 MHz): δ 2.66 [s, 4 H, $N(CH_2)_2N$], 3.69 (s, 8 H, $O{=}CCH_2N$); ^{13}C NMR (Me_2SO-d_6, 125 MHz): δ 51.15 [$N(CH_2)_2N$], 52.23 ($O{=}CCH_2N$), 165.71 ($C{=}O$); EIMS [*m/z* (RI%)]: 256 (7) $[M]^+$, 229 (6), 184 (4), 141 (21), 128 (100) [symmetrical cleavage], 56 (60).

Ethylenediaminetetraacetic acid monoanhydride (*EDTA monoanhydride*) (**3**).—The synthesis was performed according to the method described by Takeshita et al. [14]. EDTA dianhydride **2** (5.0 g, 19.5 mmol) was dissolved in dry Me_2NCHO (30 mL) at 80°C under an inert N_2 atmosphere. Water (350 μL, 19.5 mmol) was added dropwise by means of a microsyringe. The solution was stirred at this temperature for 3 h, over which time the EDTA monoanhydride precipitated. The turbid mixture was cooled to room temperature, filtered under a steady stream of N_2, and washed with Me_2NCHO (25 mL) and diethyl ether (50 mL). The colorless solid collected was dried under a high vacuum. The yield was 81% (4.3 g, 15.8 mmol); mp 188–190°C (dec); IR (ν_{max}, cm^{-1}): 1640 (m, carboxyl), 1770 (s, anhydride), 2950 (w); ^1H NMR (Me_2SO-d_6, 500 MHz): δ 2.58 [t, *J* 5.8 Hz, 2 H, $CH_2NCH_2(CO)$(carboxylate)], 2.79 [t, *J* 5.6 Hz, 2 H, $CH_2NCH_2(CO)$(anhydride)], 3.42 [s, 4 H, ($NCH_2(CO)$(carboxylate)], 3.71 [s, 4 H, $NCH_2(CO)$(anhydride)]; ^{13}C NMR (Me_2SO-d_6, 125 MHz): δ 50.60 [$CH_2NCH_2(CO)$(anhydride)], 52.10 [$CH_2NCH_2(CO)$(carboxylate)], 52.56 [$NCH_2(CO)$(anhydride)], 54.68 [NCH_2 (CO)(carboxylate)], 165.74 (C = O, anhydride), 172.35 (C = O, carboxylate); EIMS [*m*/

z (RI%)]: 274 (5) $[M]^+$, 256 (3) $[M-H_2O]^+$, 216 (4) $[M-CH_2COO]^+$, 187 (12), 171 (98), 146 (21), 128 (100), 143 (46), 56 (83).

Mono-6-O-tolylsulfonylcyclomaltoheptaose (**5**).—Cyclomaltoheptaose (6.0 g, 5.29 mmol) was dried at 110°C under high vacuum for 10 h. The dextrin was then dissolved in pyridine (60 mL), cooled in a ice bath, and treated dropwise with a solution of *p*-toluenesulfonyl chloride (1 g, 5.25 mmol) in pyridine (6 mL). The mixture was stirred at room temperature under N_2 for 18 h at which time H_2O (5 mL) was added to the mixture. The pyridine was evaporated under a reduced pressure so as to concentrate the mixture to a small volume (ca. 3–5 mL). Chromatography of the crude product on a reversed-phase column (Lobar column LiChroprep RP 8, Merck Ltd.) and elution with 1:9 H_2O–EtOH as described by Fujita et al. [11] allowed for isolation of pure monotosylated cyclomaltoheptaose. The yield was 43% (2.91 g, 2.27 mmol); mp 161–162°C (dec) (lit. [11], 160–162°C dec); TLC: R_f 0.76 (solvent *A*, detection method A and B); ^{1}H NMR (Me_2SO-d_6, 200 MHz): δ 2.41 (s, 3 H, CH_3), 3.25–3.75 (m, 42 H, cyclomaltoheptaose Hs), 4.83 (m, 7 H, H-1), 7.41 (d, *J* 8.1 Hz, 2 H, *m*-Ts), 7.73 (d, *J* 8.1 Hz, 2 H, *o*-Ts); ^{13}C NMR (Me_2SO-d_6; 125 MHz): δ 21.23 (CH_3), 59.35 (C-6″), 59.97 (C-6), 68.96 (C-5′) [2], 69.74 (C-6′), 72.08 (C-5), 72.46 (C-2), 72.75 (C-2′ or C-3′), 73.10 (C-3), 80.24 (C-4′), 81.24 (C-4″), 81.58 (C-4), 101.97 (C-1), 102.23 (C-1′), 127.60 (*o*-Ts), 129.92 (*m*-Ts), 132.72 (*p*-Ts), 144.85 (S-C); FABMS [*m/z* (RI%)]: 1290 $[M+1]^+$ (100).

Mono-6-(2-aminoethyl)amino-6-deoxycyclomaltoheptaose (**6**).—Mono-6-*O*-tolylsulfonylcyclomaltoheptaose (1 g, 0.78 mmol) was dissolved in 1,2-diaminoethane (25 mL), and the solution was stirred at 70°C for 24 h. The excess diaminoethane was evaporated under reduced pressure and the residue redissolved in M ammonium hydroxide solution (100 mL). The residual diaminoethane was extracted using CH_2Cl_2 (3 × 100 mL), and the aqueous layer was then concentrated (to 2 mL) by evaporation under reduced pressure. The solution was added dropwise to 2-propanol (50 mL), resulting in the immediate formation of a colourless precipitate. The product was collected by filtration and dried under a high vacuum. The yield was 90% (0.83 g, 0.70 mmol); mp 268–271°C (dec); TLC: R_f 0.19 (solvent *B*, detection method D); IR (ν_{max}, cm^{-1}): 3364 (br, N-H and O-H), 2928 (m, C-H), 1576 (w), 1156, 1081, 1034 (br, O-C); ^{1}H NMR (D_2O, 500 MHz): δ 2.54–2.78 (m, $HNCH_2CH_2NH_2$), 2.85–3.0 (m, C-6′ CH_2), 3.28–3.87 (m, cyclomaltoheptaose Hs), 4.92 (s, br, anomeric Hs); ^{13}C NMR (D_2O, 125 MHz): δ 40.40 (C-6′$NHCH_2CH_2NH_2$), 50.08 (C-6′), 50.22 (C-6′$NHCH_2CH_2NH_2$), 61.34 (C-6), 71.53 (C-5′), 72.89 (C-5), 73.15 (C-2), 74.10 (C-3′), 74.16 (C-3), 82.03 (C-4′), 82.19 (C-4), 84.53 (C-4′), 102.70 (C-1′), 102.98 (C-1); FABMS [*m/z* (RI%)]: 1178, $[M+1]^+$ (100). Compound **6** has been previously prepared [10,21], but no physical data were reported.

Mono-6-(3-aminopropyl)amino-6-deoxycyclomaltoheptaose (**7**).—The procedure used for the preparation of mono-6-aminoethylamino-6-deoxycyclomaltoheptaose was employed with 25 mL of 1,3-diaminopropane substituted for the 1,2-diaminoethane. The yield was 97.9% (0.91 g, 0.76 mmol); mp 250–254°C (dec); TLC: R_f 0.18 (solvent *B*, detection method D); IR (ν_{max}, cm^{-1}): 3362 (br, N-H and O-H), 2928 (m, C-H), 1575

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[2] Note that primes, as in C-5′, refer to the substituted pyranose residue. Double primes, as in C-6″, refer to an adjacent pyranose residue. Such signals were readily distinguishable by their relative intensities.

(m), 1157, 1082, 1050 (br, O-C); ^{1}H NMR (D_2O, 500 MHz): δ 1.55 (qt, $NHCH_2C\mathit{H}_2CH_2N$), 2.45 (t, br, $NHCH_2CH_2C\mathit{H}_2NH_2$), 2.7 (t, br, $CH_2NHC\mathit{H}_2CH_2$), 2.9 (m, C-6′ CH_2), 3.2–3.8 (m, cyclomaltoheptaose Hs), 4.85 (s, br, anomeric Hs); ^{13}C NMR (D_2O, 125 MHz): δ 29.53 (C-6′$NHCH_2\mathit{C}H_2CH_2NH_2$), 39.33 (C-6′$NHCH_2CH_2\mathit{C}H_2NH_2$), 47.47 (C-6′$NH\mathit{C}H_2CH_2CH_2NH_2$), 50.53 (C-6′), 61.33 (C-6′), 61.47 (C-6), 71.57 (C-5′), 72.98 (C-5), 73.21 (C-2), 74.16 (3′), 74.23 (C-3), 82.03 (C-4″), 82.27 (C-4), 84.78 (C-4′), 102.69 (C-1′), 102.98 (C-1); FABMS [*m/z* (RI%)]: 1192 $[M+1]^+$ (100).

Mono-6-[2-(ethylenediaminetetracetyl)aminoethyl]amino-6-deoxycyclomaltoheptaose (**8**).—To a solution of the cyclomaltoheptaose diaminoethane (**6**; 412 mg, 0.35 mmol) in pyridine (20 mL) was added dropwise a solution of EDTA monoanhydride (95 mg, 0.35 mmol) in pyridine (20 mL). The mixture was stirred at room temperature for 24 h, after which time the pyridine was removed under reduced pressure, and the crude mixture was evaporated to dryness. The sample was then redissolved in H_2O (10 mL), prefiltered using microporous filter paper (Millipore, filter type GS; pore size, 0.22 μm) to remove any suspended materials, and then purified by Amicon filtration (YM1, Diaflo ultrafiltration membranes, MW cutoff of 1000 daltons). The yield was 71% (361 mg, 0.25 mmol); mp 268–272°C (dec); TLC: R_f 0.19 (solvent *B*, detection method D); IR (ν_{max}, cm^{-1}): 3385 (br, N-H and O-H), 2927 (m, C-H), 1636 (s, C = O carboxylate and amide I), 1401, 1157, 1082, 1030 (s, O-C); ^{13}C NMR (D_2O, 125 MHz): δ 36.54 (conformation 1: $NHCH_2\mathit{C}H_2NHCO$), 36.80 (conformation 2: $NHCH_2\mathit{C}H_2NHCO$), 46.08 (conformation 1: $NH\mathit{C}H_2CH_2NHCO$), 49.28 (conformation 2: $NH\mathit{C}H_2CH_2NHCO$), 49.35 (conformation 1: C-6′), 50.04 (conformation 2: C-6′), 50.74 ($NHCOCH_2N\mathit{C}H_2CH_2N$), 54.14 ($NHCOCH_2NCH_2\mathit{C}H_2N$), 58.00 ($N(\mathit{C}H_2COOH)_2$), 58.11 ($NHCO\mathit{C}H_2N$), 58.66 ($NCO\mathit{C}H_2COOH$), 61.45 (C-6), 61.69 (C-6′), 68.54 (conformation 2: C-5′), 68.86 (conformation 1: C-5′), 72.98 (C-5), 73.22 (C-2), 73.45 (C-2′), 74.09 (C-3′), 74.26 (C-3), 81.60 (C-4″), 82.26 (C-4), 82.50 (C-4‴), 84.31 (C-4′), 102.41 (C-1′), 103.00 (C-1), 171.48 ($N(CH_2\mathit{C}OOH)_2$), 175.45 ($\mathit{C}OOH$), 177.95 ($NH\mathit{C}O$); FABMS [*m/z* (RI%)]: 1452 $[M+1]^+$ (70), 1178 $[M-EDTA]^+$ (100).

Mono-6-[3-(ethylenediaminetetracetyl)aminopropyl]amino-6-deoxycyclomaltoheptaose (**9**).—The procedure employed was that used in the synthesis of mono-6-[2-(ethylenediaminetetracetyl)aminoethyl}amino-6-deoxycyclomaltoheptaose with the cyclomaltoheptaose-diaminopropane adduct **7** (418 mg, 0.35 mmol) used in place of the cyclomaltoheptaose-diaminoethane adduct **6**. The yield was 76.8% (0.395 g, 0.27 mmol); mp 275–280°C (dec); TLC: R_f 0.16 (solvent *B*, detection method D); IR (ν_{max}, cm^{-1}): 3379 (br, N-H and O-H), 2928 (m, C-H), 1730 (w, C=O), 1634 (s, C=O and amide I), 1399 (w), 1156, 1083, 1038 (br, O-C); ^{13}C NMR (D_2O, 125 MHz): δ 24.86 (conformation 1: $NHCH_2\mathit{C}H_2CH_2NHCO$), 26.39 (conformation 2: $NHCH_2\mathit{C}H_2CH_2NHCO$), 37.01 (conformation 1: $NHCH_2CH_2\mathit{C}H_2NHCO$), 37.77 (conformation 2: $NHCH_2CH_2\mathit{C}H_2NHCO$), 46.45 (conformation 1: $NH\mathit{C}H_2CH_2CH_2NHCO$), 46.88 (conformation 2: $NH\mathit{C}H_2CH_2CH_2NHCO$), 49.17 (conformation 2: C-6′), 49.49 (conformation 1: C-6′), 51.24 ($NHCOCH_2N\mathit{C}H_2CH_2N$), 53.91 ($NHCOCH_2NCH_2\mathit{C}H_2N$), 57.40 [$N(\mathit{C}H_2COOH)_2$], 59.10 (conformation 2: $NHCO\mathit{C}H_2N$), 59.64 ($N\mathit{C}H_2COOH$), 60.53 (conformation 1: $NHCO\mathit{C}H_2N$), 61.29 (C-6″), 61.49 (C-6), 61.73 (C-6‴), 68.68 (conformation 2: C-5′), 68.81 (conformation 1: C-5′), 72.97 (C-5), 73.23 (C-2), 73.52 (C-2′), 74.05 (C-3′), 74.26 (C-3), 81.66 (C-4″), 82.26 (C-4), 82.55 (C-4‴), 84.18 (C-4′), 102.38 (C-

1′), 102.98 (C-1), 171.64 $N(CH_2\mathit{C}OOH)_2$), 175.25 (*C*OOH), 179.59 (NH*C*O); FABMS [*m*/*z* (RI%)]: 1483 $[M+H_2O]^+$ (100), 1466 $[M+1]^+$ (22).

A sample of **9** in D_2O was treated with 0.5 equiv of Ca^{2+} and the ^{13}C NMR spectrum was recorded. Some of the signals broadened and decreased in intensity. An additional 0.5 equiv of Ca^{2+} was added, and the ^{13}C NMR spectrum was recorded showing: δ 26.41 ($NHCH_2\mathit{C}H_2CH_2NHCO$), 37.73 ($NHCH_2CH_2\mathit{C}H_2NHCO$), 46.42 ($COCH_2N\mathit{C}H_2CH_2N$), 46.90 ($NH\mathit{C}H_2CH_2CH_2NH$), 49.20 (C-6′), 49.34 $[NCH_2\mathit{C}H_2N(CH_2COO^-)_2]$, 57.9 [v br, $N(\mathit{C}H_2COO^-)_2$], 61.39 (C-6), 61.70 (C-6″), 68.68 (C-5′), 72.79 (C-5), 73.01 (C-5″), 73.16 (C-2), 73.59 (C-2′), 74.02 (C-3′), 74.27 (C-3), 81.39 (C-4″), 82.11 (C-4), 82.44 (C-4‴), 84.24 (C-4′), 102.21 (C-1′), 102.95 (C-1). Signals for carbons at $NHCO\mathit{C}H_2N$, $N\mathit{C}H_2COO^-$, and the three carboxyls and single amide were all very broad and not reliably distinguishable from background noise.

2-(Triphenylmethylthio)acetic acid (**12**).—This synthesis was performed according to the method of Brenner et al. [22]. A solution of triphenylmethanol (48.0 g, 184.0 mmol), mercaptoacetic acid (12.2 mL, 184.0 mmol), and glacial AcOH (160 mL) was heated to 70°C. Boron trifluoride etherate (32.0 mL) was added, and the resulting brown solution was stirred at room temperature for 50 min. The mixture was poured into ice-cold H_2O, depositing a clear oil which solidified when left at 5°C for 18 h. The solid was washed with distilled H_2O (100 mL) and diethyl ether (100 mL), and recrystallized from toluene to give **12** as a colourless solid. The yield was 72% (44.3 g, 132.5 mmol); mp 155.0–157.0°C (lit. [22], 158.5–160.0°C); TLC: R_f 0.33 (solvent *C*, detection method B); IR (ν_{max}, cm^{-1}): 3000 (s) (O-H), 1700 (s) (C=O, acid), 1600, 1490, 1445 (m) (Ar-C=C); ^1H NMR (acetone-d_6, 90 MHz): δ 2.90 (s, 2 H, SCH_2), 7.25 (m, 15 H, Ar), 10.20 (s, 1 H, COOH); ^{13}C NMR ($CDCl_3$, 50 MHz): δ 34.47 ($S\mathit{C}H_2$), 67.24 ($\mathit{C}Ph_3$), 127.02 (*para* C), 127.83 (*meta* C), 128.13 (*ortho* C), 143.90 (subst. C), 175.57 (COOH); FABMS [*m*/*z* (RI%)]: 333 (22) $[M-1]^-$, 441 (11) $[M+TG-1]^-$, 243 (73) $[Tr]^-$.

N-*Succinimido-2-(triphenylmethylthio)acetate* (**13**).—This synthesis was performed according to the method of Brenner et al. [22]. To a solution of 2-(triphenylmethylthio)acetic acid (5.01 g, 15.00 mmol) and *N*-hydroxysuccinimide (1.73 g, 15.00 mmol) in dimethoxyethane (38 mL) cooled at 0°C was added 1,3-dicyclohexylcarbodiimide (DCC; 3.20 g, 15.5 mmol). The resulting mixture was then left at 5°C for 18 h during which time crystalline 1,3-dicyclohexylurea precipitated. The dicyclohexylurea was filtered, washed well with cold CH_2Cl_2, and discarded. The filtrate and CH_2Cl_2 washings were combined and concentrated in vacuo to afford a colourless solid. Residual amounts of dicyclohexylurea were removed via recrystallization from EtOAc to give **13** as large, opaque prisms. The yield was 84% (5.43 g, 12.60 mmol); mp 178.5–180.0°C (lit. [22], 178.5–179.5°C); TLC: R_f 0.46 (solvent *D*, detection method B); IR (ν_{max}, cm^{-1}): 3070 (w) (Ar-C=C-H), 2940, 2860 (w) (aliphatic C-H), 1820, 1790 (s) (N-C=O), 1745 (s) (C=O, ester), 1630 (w), 1580 (w), 1495 (m), 1445 (m) (Ar-C=C), 735, 690 (s) (monosubstituted Ar); ^1H NMR ($CDCl_3$, 500 MHz): δ 2.67 [s, 4 H, $(CH_2C=O)_2$], 3.08 (s, 2 H, SCH_2), 7.17 (m, 18 H, Ar); ^{13}C NMR ($CDCl_3$, 125 MHz): δ 25.71 ($(\mathit{C}H_2C=O)_2$), 31.60 (SCH_2), 127.32 (*para* C), 128.13 (*meta* C), 129.94 (*ortho* C), 143.73 (subst. C), 165.24 (*C*OON), 168.76 ($CH_2\mathit{C}=O$); DCIMS [*m*/*z* (RI%)]: 449(7) $[M+NH_4]^+$, 243 (100) $[Tr]^+$.

Ethyl 2,3-diaminopropanoate, hydrochloride salt (**15**).—This synthesis was performed according to the method of Fritzberg et al. [17]. Hydrogen chloride was bubbled for 30 min into a stirred suspension of the hydrochloride salt of (±)-2,3-diaminopropanoic acid (504 mg, 3.58 mmol) in abs EtOH (84 mL) and cooled using an ice bath. The suspension was then allowed to reflux for 4 days. The resulting clear solution was filtered and concentrated in vacuo to afford **15** as an amorphous, colourless solid. The yield was 98% (704 mg, 3.51 mmol); mp 164–166°C; TLC: R_f 0.27 (solvent E, detection method C); IR ($\nu_{\max}$, cm^{-1}): 2900 (s) (N-H stretch of salt), 1745 (s) (C=O, ester), 1580, 1505 (m) (N-H bend of salt), 1245 (s) (O-C); ^{1}H NMR (Me_2SO-d_6, 200 MHz): δ 1.25 (t, J 7.1 Hz, 3 H, OCH_2CH_3), 3.31–3.34 (m, 2 H, diastereotopic H-3), 4.22 (m, 2 H, diastereotopic OCH_2CH_3), 4.40 (m, 1 H, $CH_2CHCOOEt$), 8.82 (br s, 6 H, NH_3+); ^{13}C NMR (Me_2SO-d_6; 125 MHz): δ 13.75 (OCH_2CH_3), 38.50 (NCH_2CH), 50.10 ($CH_2CHCOOEt$), 62.65 (OCH_2CH_3), 166.44 ($COOCH_2$); FABMS [m/z (RI%)]: 133 (100) $[(M-2HCl)+1]^+$, 265 (10) $[2(M-2HCl)+1]^+$.

Ethyl 2,3-bis(triphenylmethylthioacetylamino)propanoate (**16**).—To a stirred solution of the succinimidoyl ester **13** (3.81 g, 8.82 mmol) in dry MeCN (160 mL) under N_2 was added the diamino ester **15** (905 mg, 4.41 mmol) in dry MeCN (40 mL) and Et_3N (3 mL). After 36 h, the mixture was concentrated using a rotary evaporator, and the residual white paste was dissolved in 0.5 M HCl (100 mL) and extracted into CH_2Cl_2 (2 × 100 mL). The combined CH_2Cl_2 layers were washed with 0.2 M $NaHCO_3$ (200 mL) and satd brine (200 mL), and dried over $MgSO_4$. Filtration and evaporation of the solvent under reduced pressure afforded **16** as a colourless solid. The yield was 97% (3.28 g, 4.28 mmol); mp 63–66°C; TLC: R_f 0.50 (solvent C, detection method B); IR ($\nu_{\max}$, cm^{-1}): 3300 (s) (N-H bonded), 3060 (m) (Ar-C=C-H), 2980, 2940 (s) (aliphatic C-H), 1740 (s) (C=O, ester), 1660(s) (C=O, amide I), 1520 (s) (N-H, amide II), 1490, 1450 (s) (Ar-C=C), 1200 (s) (C-O, ester), 740, 695 (s) (monosubstituted Ar-C-H); ^{1}H NMR ($CDCl_3$, 200 MHz): δ 1.27 (t, J 6.7 Hz, 3 H, OCH_2CH_3), 3.05 (br s, 4 H, SCH_2), 3.21 (m, 2 H, NCH_2), 4.15 (m, 3 H, overlapping $CHCOOEt$ and OCH_2CH_3), 6.26 (t, J 6.4 Hz, 1 H, $CONHCH_2$), 6.82 (d, J 6.8 Hz, 1 H, $CONHCH$), 7.19–7.47 (m, 31 H, Ar); ^{13}C NMR ($CDCl_3$, 125 MHz): δ 13.93 (OCH_2CH_3), 35.70 (SCH_2), 35.88 (SCH_2), 41.59 (NCH_2CH), 52.68 ($CHCOOEt$), 61.84 (OCH_2CH_3), 67.57 (CPh_3), 67.62 (CPh_3), 126.88 (*para* C), 128.17 (*meta* C), 129.43 (*ortho* C), 143.74 (subst. C), 143.79 (subst. C), 168.48 (NHCO), 168.69 (NHCO), 169.46 (COOEt).

2,3-Bis(triphenylmethylthioacetylamino)propanoic acid (**17**).—To ester **16** (1.23 g, 1.61 mmol) dissolved in solution of 33% CH_2Cl_2 in EtOH (120 mL) was added Amberlyst A-26 (OH^-) ion-exchange resin (100 mL, 100 mequiv pre-swollen in EtOH), Aliquat 336 (0.25 mL), and deionized H_2O (10 mL). After 48 h at ambient temperature, the mixture was filtered, and the collected resin was washed with EtOH (2 × 100 mL) and acetone (2 × 100 mL). The desired acid **17** was then eluted from the resin using a solution consisting of 1:1 0.08 M HCl–acetone until the eluent had a $pH < 2$, at which point the resin was washed with $CHCl_3$ (2 × 100 mL). The aqueous and organic eluents were combined, and the $CHCl_3$ was evaporated under reduced pressure. The colourless precipitate which formed was extracted into $CHCl_3$ (200 mL) and washed with 0.5 M HCl (200 mL). The $CHCl_3$ layer was dried over $MgSO_4$ and evaporated under reduced pressure. The remaining yellow oil was triturated with diethyl ether (100 mL), and the resultant suspension was kept at

− 15°C for 18 h and filtered, and the residue was washed with cold ether. The solid **17** was collected, dried under high vacuum, and recrystallized from 2-propanol. The yield was 92% (1.09 g, 1.48 mmol); mp 206.0–207.5°C; TLC: R_f 0.22 (solvent *C*, detection method B); IR (ν_{max}, cm^{-1}): 3000 (s) (COOH), 1725 (s) (C=O, acid), 1665 (s) (C=O, amide I), 1600, 1490, 1440 (m) (Ar-C=C); ^1H NMR (Me_2SO-d_6, 500 MHz): δ 2.75 (s, 2 H, $SCH_2CONHCH_2$), 2.79 (AB q, *J* − 13.9 Hz, 2 H, $SCH_2CONHCH$), 3.16 and 3.21 (m, 2 H, diastereotopic NCH_2), 3.89 (m, 1 H, *CH*COOH), 7.21–7.33 (m, 30 H, Ar), 7.61 (d, *J* 7.2 Hz, 1 H, CON*H*CH), 7.75 (t, *J* 5.2 Hz, 1 H, $CONHCH_2$); ^{13}C NMR ($CDCl_3$, 125 MHz): δ 35.66 (SCH_2), 35.84 (SCH_2), 41.59 (NCH_2), 55.24 (N*C*HCOOH), 67.72 (*C*Ph_3), 127.08 (*para* C), 128.17 (*meta* C), 129.66 (*ortho* C), 143.77 (subst. C), 169.97 (NH*C*O), 170.99 (*C*OOH); FABMS [*m/z* (RI%)]: 737 (19) $[M+1]^+$, 493 (31), 391 (100) $[M+1-C_{22}H_{20}OSN]^+$, 243 (71) $[Tr]^+$.

Mono-6-[2,3-bis(triphenylmethylthioacetylamino)propanoyl]-6-deoxycyclomaltoheptaose (**18**).—Acid **17** (13 mg, 0.02 mmol) was dissolved in Me_2NCHO (1 mL) and treated with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC, 6 mg, 0.03 mmol) under N_2. After 10 min, a mixture of **7** (20 mg, 0.02 mmol) and Et_3N (0.05 mL) in Me_2NCHO (1 mL) was added dropwise to the activated acid, and the solution was stirred at room temperature for 48 h. The clear solution was concentrated under reduced pressure to a viscous, clear oil. Treatment with distilled H_2O (10 mL) allowed for precipitation of **18**, which was filtered and washed with H_2O (20 mL) and $CHCl_3$ (20 mL). The product was recrystallized from EtOH to yield **18** (ditrityl-DADS) as a colourless, amorphous solid. The yield was 53% (21 mg, 0.01 mmol); TLC: R_f 0.85 (solvent *A*, detection method D); IR (ν_{max}, cm^{-1}): 3410 (s) (N-H, O-H bonded), 3060 (m) (Ar-C=C-H), 2930 (s) (aliphatic C-H), 1655 (s) (C=O, amide I), 1520 (m) (N-H, amide II), 1490, 1440 (m) (Ar-C=C), 1150 (w), 1075, 1030 (m) (C-O), 735, 695 (s) (monosubstituted Ar-C-H); ^1H NMR (Me_2SO-d_6, 500 MHz): δ 1.7 (m, $NCH_2CH_2CH_2N$), 2.8–3.25 (m, SCH_2s and NCH_2s), 3.3–3.8 (m, cyclomaltoheptaose Hs), 4.18–4.5 (br, OHs), 4.85 (br, anomeric Hs), 5.6--6.0 (br, OHs), 7.15–7.4 (m, ArHs), 7.8, 7.91, 7.94 (br, three CON*H*s); ^{13}C NMR (Me_2SO-d_6, 125 MHz): δ 26.28 ($NHCH_2CH_2CH_2NHCO$), 35.88 (SCH_2), 35.98 (SCH_2), 36.67 ($NHCH_2CH_2CH_2NHCO$), 40.52 ($NHCH_2CHCONH$), 43.17 ($NHCH_2CH_2CH_2NHCO$), 52.83 (C-6′), 55.32 (NHCO*C*HN), 59.88 (C-6), 65.85 (*C*Ph_3), 70.0 (C-5′), 72.00 (br C-5), 72.39 (br C-2), 73.00 (br C-3), 81.51 (br C-4), 101.89 (br C-1), 126.76 (*para* C), 128.03 (*meta* C), 129.02 (*ortho* C), 143.98 (subst. C), 167.33 (NH*C*O), 167.43 (NH*C*O), 167.71 (NH*C*O). (Note that some of the substituted pyranose ring signals in this sample were difficult to distinguish reliably from baseline noise.)

Mono-6-[3-(5,8-diaza-4,9-dioxo-1,2-dithiadodec-6-anoyl)aminopropyl]amino-6-deoxycyclomaltoheptaose (**19**).—This synthesis was a modification of the method of Kamber [23]. Resublimed I_2 (31 mg, 2.1 equiv, 0.12 mmol) was added to a solution of the *S*-tritylated cyclomaltoheptaose **18** (65 mg, 0.06 mmol) in 20:80 CH_2Cl_2–abs EtOH (degassed using aspirator vacuum, 50 mL). The amber-brown solution was degassed for 30 min, vented to dry N_2, and stirred at room temperature. After 12 h, the suspension was cooled to − 15°C for 18 h and filtered with cold EtOH. The residue was dissolved in warm, deionized H_2O (100 mL), washed with $CHCl_3$ (2×50 mL), filtered through glass wool, and evaporated under reduced pressure to give **19** (CD-DADS) as a pale-yellow solid. The yield was 34% (29 mg, 0.02 mmol); mp 195–200°C (dec); TLC: R_f 0.05 (solvent *A*, detection method C); IR ($\nu_{\max}$, cm^{-1}): 3444 (s) (O-H, N-H bonded), 2924, 2853 (s) (aliphatic C-H), 1652 (s) (C=O, amide I), 1558, 1535 (m) (N-H, amide II), 1156, 1079, 1028 (s) (C-O); ^{13}C NMR (acetone-d_6, 125 MHz): δ 25.83 ($NHCH_2\mathit{C}H_2CH_2NHCO$), 36.66 [conformation 1 of disulfide ring (major) $NHCH_2CH_2\mathit{C}H_2NHCO$], 37.19 [conformation 2 (minor) $NHCH_2CH_2\mathit{C}H_2NHCO$], 38.98 (conformation 2, $S\mathit{C}H_2$), 40.91 (br, conformation 1, $S\mathit{C}H_2$), 41.47 (br, conformation 1, $OCNH\mathit{C}H_2CH$), 42.57 (conformation 2, $OCNH\mathit{C}H_2$-CH), 46.18 ($NH\mathit{C}H_2CH_2CH_2NHCO$), 48.58 (C-6′), 53.69 (conformation 2, $NHCO\mathit{C}HN$), 54.55 (br, conformation 1, $NHCO\mathit{C}HN$), 60.72 (C-6), 61.09 (C-6″), 67.94 (C-5′), 72.21 (C-5), 72.47 (C-2), 73.34 (C-3′), 73.53 (C-3), 80.85 (C-4″), 81.53 (C-4), 81.80 (C-4‴), 83.56 (C-4′), 101.59 (C-1′), 102.28 (C-1), 172.25 (br, 3 NHCO); FABMS [*m*/*z* (RI%)]: 1423 (20) $[M+1]^+$, 1216 (100) $[(M+1)-C_6H_{11}N_2O_2S_2]^+$.

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References

[1] I.S. Menzies, in E. Skadhauge and K. Heintze (Eds.), *Intestinal Absorption and Secretion, Falk Symposium* 36, MTP Press, Lancaster, 1983, p 527.

[2] I.S. Menzies, M.F. Laker, R. Pounder, J. Bull, S. Heyer, P.G. Wheeler, and B. Creamer, *Lancet*, II (1979) 1107–1109.

[3] I. Cobden, R.J. Dickinson, J. Rothwell, and A.T.R. Axon, *Brit. Med. J.*, 2 (1978) 1060.

[4] S.O. Ukabam, J.R. Clamp, and B.T. Cooper, *Digestion*, 27 (1983) 70–74.

[5] I. Bjarnason, C. O'Morain, A.J. Levi, and T.J. Peters, *Gastroenterology*, 85 (1983) 318–322.

[6] (a) V.S. Chadwick, S.F. Phillips, and A.F. Hofmann, *Gastroenterology*, 73 (1977) 241–246; (b) V.S. Chadwick, S.F. Phillips, and A.F. Hofmann, *ibid.*, 73 (1977) 247–251; (c) R.T. Jenkins, R.L. Goodacre, P.J. Rooney, J. Bienenstock, T. Sivakumaran, and W.H.C. Walker, *Clin. Biochem.*, 19 (1986) 298–302.

[7] R. Ecknauer, B. Buck, and D. Breitig, *Digestion*, 26 (1983) 24–32.

[8] T.A. Waldmann, *Lancet*, II (1961) 121–123.

[9] (a) M.L. Bender and M. Komiyama, *Cyclodextrin Chemistry*, Springer-Verlag, Berlin, 1978; (b) W. Saenger, *Angew. Chem. Int. Ed. Engl.*, 19 (1980) 344–362; (c) J.S. Pagington, *Chem. Br.*, (1987) 445.

[10] (a) Y. Matsui, T. Yokoi, and K. Mochida, *Chem. Lett.*, (1976) 1037–1040; (b) K. Tsujihara, H. Kurita, and M. Kawazu. *Bull. Chem. Soc. Jpn.*, 50 (1977) 1567–1571; Y. Matsui and A. Okimoto, *ibid.*, 51 (1978) 3030–3034.

[11] K. Fujita, A. Matsunaga, and T. Imoto, *Tetrahedron Lett.*, 25 (1984) 5533–5536.

[12] R.C. Petter, J.S. Salek, C.T. Sikorski, G. Kumaravel, and F.-T. Lin, *J. Am. Chem. Soc.*, 112 (1990) 3860–3868.

[13] J.R.A. Geigy, Fr. Pat., 1 548 888 (1969).

[14] T. Takeshita, T.-A. Shimohara, and S. Maeda, *J. Am. Oil Chem. Soc.*, 59 (1982) 104–107.

[15] G. Mariam, C. Rosa, M. Raciti, C. Fatigante, E. Consoli, and G. Parenti, in *Technetium in Chemistry and Nuclear Medicine*, Vol. 2, Raven Press, New York, 1986, p 305.

[16] G.D. Zanelli, I. Bjarnason, T. Smith, J.C.W. Crawley, A.J. Levi, and R.I. Copeland, *Nucl. Med. Commun.*, 7 (1986) 17–24.

[17] A.R. Fritzberg, C.C. Kuni, W.C. Klingensmith, J. Stevens, and W.P. Whitnet, *J. Nucl. Med.*, 23 (1982) 592–598.

[18] R.H. Mach, H.F. Kung, P. Jungwiwattanaporn, and Y.-Z. Guo, *Tetrahedron Lett.*, 30 (1989) 4069–4072.

[19] W.J. James and D. French, *Proc. Iowa Acad. Sci.*, 59 (1952) 197–201.

[20] R.B. Maharajh, J.F. Britten and R.A. Bell, *Can. J. Chem.*, submitted.

[21] Y. Chao, Ph.D. Thesis, Columbia University, 1972; I. Tabushi, Y. Kuroda, and K. Shimokawa, *J. Am. Chem. Soc.*, 101 (1979) 4759–4760.

[22] D. Brenner, A. Davison, J. Lister-Jones, and A.G. Jones, *Inorg. Chem.*, 23 (1984) 3793–3977.

[23] B. Kamber, *Helv. Chim. Acta*, 54 (1971) 398–422.