Synthesis of a Model Linear Mannoheptaose for the Backbone Structure of Fruit Body Polysaccharide of *Tremella fuciformis* and *Dictyophora indusiata* FISCH[†]

Tomoya OGAWA* and Hisao YAMAMOTO**

RIKEN (The Institute of Physical and Chemical Research),
Wako, Saitama 351, Japan

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Synthesis of linear D-manno-oligosaccharides, $O\text{-}\alpha\text{-D-Man-}\{(1\rightarrow3)\text{-}O\text{-}\alpha\text{-D-Man}\}_n\text{-(1}\rightarrow3)\text{-}O\text{-}\alpha\text{-D-Man}$ ($n=0\sim4$), which correspond to part of the structure of the cell-wall polysaccharide of *Tremella fuciformis* and *Dictyophora indusiata* FISCH, is described.

Recently, the structure of the repeating unit of an acidic heteropolysaccharide isolated from fruit bodies of *Tremella fuciformis* BERK was proposed to be **1** (ref. 2), which contains a linear $(1\rightarrow3)\text{-}\alpha\text{-D-mannan}$ chain as the backbone structure. A linear $\alpha(1\rightarrow3)\text{-D-mannan}$ chain **2** carrying *O*-acetyl groups was also proposed for the polysaccharide isolated from fruit bodies of *Dictyophora indusiata* FISCH.³⁾

As part of a project on the synthesis of microbial cell surface glycans, we describe here regio- and stereo-controlled synthesis of D-mannoheptaose **3** which corresponds to part of the structures of glycans **1** and **2**.

RESULTS AND DISCUSSION

Based on retrosynthetic considerations, two monosaccharide synthons **4** and **5**, were designed for a stepwise approach to the synthesis of target mannoheptaose **3**. Since a synthetic approach to only **4** has been reported,^{1,4)} we describe first a synthetic approach to glycosyl acceptor **5** and then stepwise elongation of the glycan chain by employing both **4** and **5**.

Tritylation of benzyl $\alpha\text{-D-mannopyranoside}$ **6** to **7**, and stannylation⁵⁾ with $(\text{Bu}_3\text{Sn})_2\text{O}$ followed by allylation in the presence of Bu_4NBr ⁶⁾ afforded an 81% yield of a mixture of 3-*O*-allyl **8** and 2,3-di-*O*-allyl derivative **9** in

1

2

3

M= $\alpha\text{-D-mannopyranosyl}$; GA= $\beta\text{-D-glucuronic acid}$
X= $\beta\text{-D-xylopyranosyl}$

4

5

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* To whom enquiries should be addressed.

** Present address: Research and Development Laboratories, Sapporo Breweries Ltd., 10 Okatohme, Yaizu-shi, Shizuoka 425, Japan.

SCHEME 1

SCHEME 2

a ratio of 7:1. Solvolysis of **8** and **9** in aq. AcOH afforded **10** and **11**, respectively, of which the structures were determined from the ^{13}C -NMR data as follows. The ^{13}C -NMR spectrum of **10** contains a deshielded signal for C-3 at δ 79.34 in agreement with the 3-*O*-allyl structure, while the spectrum of **11** contains two deshielded signals for C-2 and C-3 at δ 74.13 and 79.15, respectively, in addition to the shielded signal for C-1 at δ 97.67 in accordance with the 2,3-di-*O*-allyl structure. Major-product **11** was benzylated to **12**, which was deallylated with PdCl_2 ⁷⁾ in MeOH to give glycosyl acceptor **5** in a 65% overall

yield from **11**.

Having prepared glycosyl acceptor **5**, we performed stepwise chain elongation of **5** with glycosyl donor **4** as follows. Glycosidation of **5** with **4** in the presence of $\text{AgOSO}_2\text{CF}_3$ and powdered molecular sieves 4A afforded protected mannoside **13**, which was deacetylated to give **14** in an 87% overall yield from **5**. The stereochemistry of the glycosidation was assigned as α -D from the ^{13}C -NMR data which included two signals for C-1a and C-1b at δ 96.69 and 98.98, respectively, with a $^1J_{\text{CH}}$ value of about 170 Hz.⁸⁾ Hydrogenolysis of **14** with 10% Pd-C in AcOH at 50° af-

TABLE I. ¹³C-CHEMICAL SHIFTS OF D-MANNO-OLIGOSACCHARIDES

Structure	Each carbon atom at the reducing end (α-D)						Each carbon atom at the reducing end (β-D)					
	1	2	3	4	5	6	1	2	3	4	5	6
α-M(ref. 9)	95.20	71.85	71.35	68.05	73.35	62.15						
β-M(ref. 9)							94.85	72.35	74.10	67.75	77.30	62.15
M-3M(ref. 10) 15	94.79	71.15	78.66	67.01	73.35	61.75	93.50	71.69	81.14	67.55	76.70	61.75
	(170.9)						(161.1)					
M-3M-3M 18	94.79	71.15	78.85	66.91	73.35	61.70	94.29	71.85	81.24	67.64	76.76	61.70
	(169.7)						(162.4)					
M-(3M) ₂ -3M 21	94.92	71.10	78.80	66.77	73.29	61.80				67.55		61.80
	(170.1)											
M-(3M) ₃ -3M 24	94.87	71.25	79.00	67.07	73.45	61.87			81.26	67.78		61.80
M-(3M) ₄ -3M 3	94.87	71.21	79.00	66.90		61.80				67.61		61.80

Structure	Each carbon atom of the internal residues						Each carbon atom at the non-reducing end					
	1	2	3	4	5	6	1	2	3	4	5	6
M-3M 15							103.12	71.15	70.81	67.55	74.08	61.75
							(172.1)					
M-3M-3M 18	103.12	70.47	78.85	66.91	74.22	61.70	103.12	71.15	70.81	67.64	74.22	61.70
	(170.1)						(172.1)					
M-(3M) ₂ -3M 21	{102.98	70.40	78.80	66.77	74.17	61.80}	102.98	71.10	70.70	67.55	74.17	61.80
M-(3M) ₃ -3M 24	{102.94	70.54	79.00	67.07	74.39	61.87}	102.94	71.25	70.92	67.78	74.39	61.87
M-(3M) ₄ -3M 3	{103.00	70.50	79.00	66.90	74.20	61.80}	103.00	71.21	70.83	67.61	74.20	61.80

The values of δ_C are expressed in ppm as in the case of Fig. 1. Values in parentheses correspond to $^1J_{CH}$ (in Hz).

forded free mannobiose **15**. Glycosidation of mannobiosyl glycosyl acceptor **14** with glycosyl donor **4** again with use of $\text{AgOSO}_2\text{CF}_3$ gave protected mannotrioxide **16** in a 91% yield, which was deacetylated to give mannotrioxyl glycosyl acceptor **17**. A third glycosidation of acceptor **17** with donor **4** to give **19** and subsequent deacetylation of **19** afforded mannotetraosyl glycosyl acceptor **20** in an 88% overall yield from **17**. A fourth glycosidation of **20** to **22** (96%), deacetylation of **22** to **23** (87%), and a fifth glycosidation of **23** afforded **25** (73%). Deacetylation of **25** to **26** and subsequent hydrogenolysis of **26** finally afforded target mannoheptaose **3**. The stereochemistry of **3** and the synthetic intermediates were deduced from the synthetic sequences and confirmed by the ¹³C and ¹H-NMR data for the free mannooligosaccharides which were obtained by hydrogenolysis of the glycosyl acceptors, as shown in Table I and Figs. 1 and 2.

In conclusion, linear α(1→3)-D-mannoheptaose **3** was synthesized by a stepwise strategy employing mannosyl donor **4** and mannosyl acceptor **5**. The ¹³C-NMR data for the synthetic **3** were found to be in good agreement with those for the natural α(1→3)-D-mannan from *Dictyophora indusiata* FISCH reported by Ukai and coworkers.³⁾

EXPERIMENTAL

General. Melting points were determined with a Yanagimoto micro melting-point apparatus and are uncorrected. Optical rotations were determined with a Perkin-Elmer Model 241 MC polarimeter, for solutions in CHCl_3 at 25°C, unless noted otherwise. Column chromatography was performed on columns of Silica Gel Merck (70~230 mesh; E. Merck, Darmstadt, Germany). Flash chromatography was performed on columns of Wako gel C-300 (200~300 mesh; Wako Pure Chemicals, Osaka, Japan). Thin-layer chromatography (TLC) was performed on plates (layer thickness, 0.25 mm) precoated with Silica Gel 60 F₂₅₄ (E. Merck, Darmstadt, Germany). High performance thin layer chromatography (HPTLC) was

FIG. 1. 22.5 MHz ^{13}C -NMR Spectra (in D_2O at 20°) of (a) $\text{M} \rightarrow 3\text{M}$ **15**, (b) $\text{M} \rightarrow 3\text{M} \rightarrow 3\text{M}$ **18**, (c) $\text{M} \rightarrow (3\text{M})_2 \rightarrow 3\text{M}$ **21**, (d) $\text{M} \rightarrow (3\text{M})_3 \rightarrow 3\text{M}$ **24**, and (e) $\text{M} \rightarrow (3\text{M})_4 \rightarrow 3\text{M}$ **3**.

The values of δ_c are expressed in ppm downward from tetramethylsilane, with indirect reference to an internal standard of 1,4-dioxane (δ 67.40).

FIG. 2. 400 MHz ^1H -NMR Spectra (in D_2O at 63°) of (a) $\text{M} \rightarrow (3\text{M})_4 \rightarrow 3\text{M}$ **3**, (b) $\text{M} \rightarrow (3\text{M})_3 \rightarrow 3\text{M}$ **24**, (c) $\text{M} \rightarrow (3\text{M})_2 \rightarrow 3\text{M}$ **21**, (d) $\text{M} \rightarrow 3\text{M} \rightarrow 3\text{M}$ **18**, and (e) $\text{M} \rightarrow 3\text{M}$ **15**.

The values of δ_{H} are expressed in ppm downward from the internal standard, sodium 2,2,3,3-tetradeuterio-4,4-dimethyl-4-silapentanoate. The values in parentheses correspond to $^3J_{\text{HH}}$ (in Hz).

performed on plates (layer thickness, 0.20 mm) precoated with Silica Gel 60 F₂₅₄ (E. Merck, Darmstadt, Germany). IR spectra were recorded with an EPI-G2 Hitachi spectrophotometer, using KBr pellets for the crystalline samples, and neat films for the liquid samples. ¹H-NMR spectra were recorded with either a JNM-GX400 or a JNM-FX90Q NMR spectrometer, using tetramethylsilane as the internal standard. ¹³C-NMR spectra were recorded with a JNM-FX 100FT NMR spectrometer operated at 25.05 MHz. The values of δ_C and δ_H are expressed in ppm downwards from the internal standard, for solutions in CDCl₃, unless noted otherwise.

Benzyl 6-O-trityl- α -D-mannopyranoside 7. To a solution of benzyl α -D-mannopyranoside **6** (20 g, 74 mmol) in pyridine (200 ml) was added trityl chloride (25 g, 90 mmol) and the mixture was stirred for 24 hr at 25°C. Processing and chromatography over SiO₂ C-300 (500 g) in 3:1 CHCl₃-MeOH gave **7** (24.5 g, 62%), $[\alpha]_D^{25} + 19.3^\circ$ ($c = 1.0$), *Rf* 0.59 in 2:1 CHCl₃-MeOH, δ_H : 4.82 (1H, s, H-1), 4.76 and 4.46 (2H, AB, $J = 12$ Hz, -CH₂-Ph).

Anal. Calcd. for C₃₃H₃₂O₆: C, 74.98; H, 6.29. Found: C, 74.59; H, 6.47%.

Benzyl 3-O-allyl-6-O-trityl- α -D-mannopyranoside 8 and benzyl 2,3-di-O-allyl-6-O-trityl- α -D-mannopyranoside 9. A mixture of **7** (5.0 g) and (Bu₃Sn)₂O (4.0 g) in toluene (100 ml) was stirred under reflux for 17 hr with azeotropic removal of water and finally concentrated to the volume of 30 ml. To this solution was added allyl bromide (4 ml) and Bu₄NBr (0.5 g). The mixture was stirred for 16 hr at 100°C, and concentrated *in vacuo*. A solution of the residue in EtOAc (150 ml) was stirred with satd. aq. KF (50 ml) and precipitated Bu₃SnF was filtered off. The organic layer was processed and the product was chromatographed on SiO₂ C-300 (300 g) in 5:1 toluene-EtOAc to give **8** (3.79 g, 71.0%) *Rf* 0.40 and **9** (600 mg, 10.0%) *Rf* 0.73 in 5:1 toluene-EtOAc. Compound **9** was used for the next step without further characterization. **8**: $[\alpha]_D^{25} + 15.6^\circ$ ($c = 1.50$). δ_H : 7.6~7.1 (20H, m, aromatic), 6.1~5.7 (1H, m, -CH=CH₂), 5.36~5.10 (2H, m, -CH=CH₂), 4.92 (1H, d, $J = 2$ Hz, H-1), 4.74 and 4.50 (2H, AB, $J = 12$ Hz, CH₂-Ph).

Anal. Calcd. for C₃₅H₃₆O₆: C, 76.06; H, 6.57. Found: C, 76.18; H, 6.68%.

Benzyl 3-O-allyl- α -D-mannopyranoside 10. A solution of **8** (3.7 g) in 80% aq. AcOH (100 ml) was stirred for 20 min at 80°C. Evaporation *in vacuo* and chromatography on SiO₂ C-300 (60 g) in toluene and then 1:3 toluene-EtOAc gave **10** (2.05 g, 99.0%) $[\alpha]_D^{25} + 40.9^\circ$ ($c = 0.35$), *Rf* 0.35 in 1:3 toluene-EtOAc. δ_H : 7.32 (5H, s, aromatic), 6.10~5.76 (1H, m, -CH=CH₂), 5.40~5.12 (2H, m, -CH=CH₂), 4.95 (1H, s, H-1), 4.70 and 4.46 (2H, AB, $J = 12$ Hz, CH₂Ph). δ_C : 99.13 (¹J_{CH} = 169.7 Hz, C-1), 79.34 (C-3), 72.81 (C-5), 71.11 (C₃-O-CH₂-CH=), 69.21 (C₁-O-CH₂-Ph), 68.23 (C-2), 64.97 (C-4), 61.07 (C-6).

Anal. Calcd. for C₁₆H₂₂O₆: C, 61.92; H, 7.15. Found: C,

61.61; H, 7.08%.

Benzyl 2,3-di-O-allyl- α -D-mannopyranoside 11. **9** (600 mg) was detritylated and processed as above to give **11**. δ_C : 97.67 (¹J_{CH} = 168.5 Hz, C-1), 79.15 (C-3), 74.13 (C-2), 72.71 (C-5), 72.23 (C₂-O-CH₂-CH=), 70.77 (C₃-O-CH₂-CH=), 69.01 (C₁-O-CH₂-Ph), 66.77 (C-4), 62.33 (C-6).

Benzyl 3-O-allyl-2,4,6-tri-O-benzyl- α -D-mannopyranoside 12. To a solution of **10** (1.4 g) in DMF (20 ml) was added NaH (50%, 0.5 g) and the mixture was stirred for 1 hr at 20°C. To this mixture was added dropwise benzyl bromide (3 ml) at -5°C and the mixture was stirred for 2 hr at 15~20°C. Excess NaH was decomposed by adding MeOH (5 ml) at 0°C. The reaction mixture was concentrated *in vacuo*, and then partitioned between water and EtOAc. The organic layer was washed with water, satd. saline, dried (MgSO₄) and concentrated *in vacuo*. The residue was chromatographed on SiO₂ C-300 (70 g) in 10:1, toluene-EtOAc to give **12** (1.8 g, 74.0%), $[\alpha]_D^{25} + 38.6^\circ$ ($c = 0.78$), *Rf* 0.61 in 10:1 toluene-EtOAc.

Anal. Calcd. for C₃₇H₄₀O₆: C, 76.52; H, 6.94. Found: C, 76.23; H, 6.71%.

Benzyl 2,4,6-tri-O-benzyl- α -D-mannopyranoside 5. A mixture of **12** (1.6 g) and PdCl₂ (100 mg) in MeOH (20 ml) was stirred for 2 hr at 20°C and filtered through celite. The filtrate was evaporated and the residue was partitioned between H₂O and EtOAc. The organic layer was washed with water, satd. saline, dried (MgSO₄), and concentrated *in vacuo*. The residue was chromatographed on SiO₂ C-300 (50 g) in 10:1 toluene-EtOAc to give **5** (1.3 g, 87%), $[\alpha]_D^{25} + 20.0^\circ$ ($c = 0.78$), *Rf* 0.41 in 10:1 toluene-EtOAc. δ_H : 7.5~7.1 (20H, m, aromatic), 5.01 (1H, d, $J = 2$ Hz, H-1). δ_C : 96.11 (¹J_{CH} = 167.2 Hz, C-1), 78.32 (C-4), 76.62 (C-2), 74.86 (C₄-O-CH₂-Ph), 73.40 (C₆-O-CH₂-Ph), 72.81 (C₂-O-CH₂-Ph), 71.84 (C-5), 71.16 (C-3), 69.01 (C-6 and C₁-O-CH₂-Ph).

Anal. Calcd. for C₃₄H₃₆O₆: C, 75.53; H, 6.71. Found: C, 75.18; H, 6.58%.

Benzyl O-(3-O-acetyl-2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 13. To powdered molecular sieves **4A** (3 g) was added a solution of AgOSO₂CF₃ (500 mg) in toluene (10 ml), and the toluene was evaporated *in vacuo* at 50°C. To the remaining mixture was added dropwise a solution of **5** (850 mg, 1.57 mmol) in Cl(CH₂)₂Cl (20 ml) and then a solution of **4** (950 mg, 1.78 mmol) in Cl(CH₂)₂Cl (3 ml) at -15°C. The mixture was stirred for 24 hr at 20°C under argon and then further **4** (70 mg) in Cl(CH₂)₂Cl (1 ml) was added dropwise at -15°C. The mixture was stirred for 24 hr and processed. Chromatography on SiO₂ C-300 (10 g) in 10:1 toluene-EtOAc afforded crude **13** (2.1 g). An analytical sample of **13** was obtained by rechromatography of a small portion

of crude **13**. $[\alpha]_D + 14.1^\circ$ ($c=0.60$), R_f 0.67 in 5:1 toluene–EtOAc. δ_H : 5.34 (1H, q, $J=3$ Hz, 9 Hz, H-3b), 5.25 (1H, d, $J=2$ Hz, H-1b), 4.97 (1H, d, $J=2$ Hz, H-1a), 1.94 (3H, s, Ac).

Anal. Calcd. for $C_{63}H_{66}O_{12}$: C, 74.54; H, 6.55. Found: C, 74.10; H, 6.53%.

Benzyl O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 14. A solution of crude **13** (2.0 g) in THF (20 ml) and 0.14 N NaOMe–MeOH (35 ml) was stirred for 4 hr at 25°C. Neutralization with Amberlist A-15 and chromatography on SiO_2 C-300 (20 g) in 10:1 toluene–EtOAc gave **14** (1.49 g, 87% from **5**), $[\alpha]_D + 16.0^\circ$ ($c=0.60$), R_f 0.59 in 5:1 toluene–EtOAc. δ_H : 5.32 (1H, d, $J=2$ Hz, H-1b), 4.98 (1H, d, $J=2$ Hz, H-1a). δ_C : 98.98 ($^1J_{CH}=169.7$ Hz, C-1b), 96.69 ($^1J_{CH}=168.5$ Hz, C-1a).

Anal. Calcd. for $C_{61}H_{65}O_{11}$: C, 75.21; H, 6.73. Found: C, 75.09; H, 6.63%.

O- α -D-mannopyranosyl-(1→3)- α -D-mannopyranose 15. A mixture of **14** (100 mg) and 10% Pd-C (100 mg) in AcOH (2 ml) was stirred under H_2 for 3 hr at 50°C. Processing and gel filtration on Sephadex G-25 (10 g) in H_2O gave **15** (30 mg, 89%), R_f 0.32 in 2:1:1:1 BuOH–AcOH– H_2O .

Anal. Calcd. for $C_{12}H_{22}O_{11}$: C, 42.10; H, 6.48. Found: C, 42.41; H, 6.53%.

Benzyl O-(3-O-acetyl-2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 16. To a mixture of molecular sieves 4A (3.0 g) and $AgOSO_2CF_3$ (750 mg) was added dropwise a solution of **14** (1.65 g, 1.70 mmol) in $Cl(CH_2)_2Cl$ (20 ml) and then a solution of **4** (970 mg, 1.81 mmol) in $Cl(CH_2)_2Cl$ (5 ml). The reaction mixture was stirred under argon for 2 days at 20–25°C. Processing and chromatography on SiO_2 C-300 (30 g) in 10:1 toluene–EtOAc gave **16** (2.12 g, 91%), $[\alpha]_D + 14.0^\circ$ ($c=0.30$), R_f 0.66 in 5:1 toluene–EtOAc. δ_H : 5.24 and 5.20 (two 1H, d, $J=2$ Hz, H-1b and H-1c), 4.96 (1H, d, $J=2$ Hz, H-1a), 1.92 (1H, s, Ac).

Anal. Calcd. for $C_{90}H_{94}O_{17}$: C, 74.67; H, 6.54. Found: C, 74.64; H, 6.47%.

Benzyl O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 17. A solution of **16** (1.6 g) in THF (20 ml) and 0.2 N NaOMe–MeOH (25 ml) was stirred for 3 hr at 20–25°C. Processing and chromatography on SiO_2 C-300 (20 g) in 10:1 toluene–EtOAc gave **17** (1.40 g, 90%). $[\alpha]_D + 14.3^\circ$, R_f 0.58 in 5:1 toluene–EtOAc.

Anal. Calcd. for $C_{88}H_{92}O_{16}$: C, 75.12; H, 6.60. Found: C, 75.07; H, 6.53%.

O- α -D-mannopyranosyl-(1→3)-O- α -D-mannopyranosyl-(1→3)- α -D-mannopyranose 18. A mixture of **18** (92 mg) and

10% Pd-C (60 mg) in dioxane (1 ml) and MeOH (5 ml) was stirred under H_2 for 16 hr at 25°C. Processing and trituration with H_2O –EtOH gave **18** (33 mg, quantitative) as an amorphous powder. $[\alpha]_D + 19.3^\circ$ ($c=0.30$, H_2O). R_f 0.17 in 2:1:1:1 BuOH–AcOH– H_2O .

Anal. Calcd. for $C_{18}H_{32}O_{16} \cdot 0.75 H_2O$: C, 41.74; H, 6.52. Found: C, 41.60; H, 6.61%.

Benzyl O-(3-O-acetyl-2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1→3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₂-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 19. To a mixture of molecular sieves 4A (1.5 g) and $AgOSO_2CF_3$ (200 mg) was added dropwise a solution of **17** (250 mg, 0.18 mmol) in $Cl(CH_2)_2Cl$ (8 ml) and then a solution of **4** (80 mg, 0.17 mmol) in $Cl(CH_2)_2Cl$ (2 ml) at 20°C. The mixture was stirred for 16 hr at 20°C and then further **4** (20 mg, 0.04 mmol) in $Cl(CH_2)_2Cl$ (1 ml) was added. After stirring for a further 2 hr, the usual processing and chromatography on SiO_2 C-300 (15 g) in 10:1 toluene–EtOAc gave **19** (294 mg, 88%), $[\alpha]_D + 22.4^\circ$ ($c=0.60$). R_f 0.39 in 10:1 toluene–EtOAc.

Anal. Calcd. for $C_{117}H_{122}O_{22}$: C, 74.74; H, 6.54. Found: C, 74.87; H, 6.49%.

Benzyl O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1→3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₂-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 20. A solution of **19** (294 mg) in THF (5 ml) and 0.06 N NaOMe–MeOH (5 ml) was stirred for 16 hr at 20°C. Processing and chromatography on SiO_2 C-300 (10 g) in 10:1 toluene–EtOAc gave **20** (260 mg, 90%). $[\alpha]_D + 14.1^\circ$ ($c=0.70$), R_f 0.29 in 10:1 toluene–EtOAc.

Anal. Calcd. for $C_{115}H_{120}O_{21}$: C, 75.23; H, 6.58. Found: C, 75.09; H, 6.69.

O- α -D-Mannopyranosyl-[(1→3)-O- α -D-mannopyranosyl]₂-(1→3)- α -D-mannopyranose 21. A mixture of **20** (87 mg) and 10% Pd-C (50 mg) in THF (1 ml) and MeOH (5 ml) was stirred under H_2 for 3 days at 25°C. Processing and trituration of the residue with EtOH– H_2O gave **21** (30 mg, quantitative) as an amorphous solid. $[\alpha]_D + 25.4^\circ$ ($c=0.30$, H_2O), R_f 0.12 in 2:1:1:1 BuOH–AcOH– H_2O .

Anal. Calcd. for $C_{24}H_{42}O_{21} \cdot 1.5 H_2O$: C, 41.56; H, 6.54. Found: C, 41.50; H, 6.53%.

Benzyl O-(3-O-acetyl-2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1→3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₃-(1→3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside 22. To a mixture of molecular sieves 4A (1.5 g) and $AgOSO_2CF_3$ (200 mg) was added a solution of **20** (250 mg, 0.135 mmol) in $Cl(CH_2)_2Cl$ (5 ml) and then a solution of **4** (60 mg, 0.118 mmol) in $Cl(CH_2)_2Cl$ (1 ml) at 20°C. The mixture was stirred under argon for 16 hr at 20°C. To this mixture was added a solution of $AgOSO_2CF_3$ (100 mg) in toluene (5 ml) and a solution of **4** (20 mg, 0.039 mmol) in $Cl(CH_2)_2Cl$ (1 ml). The reaction mixture

was further stirred for 5 hr. Processing and chromatography on SiO₂ C-300 (20 g) in 10:1 toluene-EtOAc gave **22** (320 mg, 96%), $[\alpha]_D + 12.2^\circ$ ($c=0.68$), R_f 0.40 in 10:1 toluene-EtOAc. δ_H : 1.93 (3H, s, Ac).

Anal. Calcd. for C₁₄₄H₁₅₀O₂₇: C, 74.78; H, 6.54. Found: C, 74.41; H, 6.56%.

*Benzyl O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1 $\rightarrow$ 3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₃-(1 $\rightarrow$ 3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside **23**.* A solution of **22** (200 mg) in THF (10 ml) and 0.1 N NaOMe-MeOH (10 ml) was stirred for 3 hr at 20°C. The usual processing and chromatography on SiO₂ C-300 (10 g) in 10:1 toluene-EtOAc gave **23** (170 mg, 87%), $[\alpha]_D + 14.4^\circ$ ($c=0.50$), R_f 0.32 in 10:1 toluene-EtOAc.

Anal. Calcd. for C₁₄₂H₁₄₈O₂₆: C, 75.11; H, 6.57. Found: C, 75.21; H, 6.91%.

*O- α -D-Mannopyranosyl-[(1 $\rightarrow$ 3)-O- α -D-mannopyranosyl]₃-(1 $\rightarrow$ 3)- α -D-mannopyranose **24**.* A mixture of **23** (100 mg) and 10% Pd-C (100 mg) in THF (1 ml) and MeOH (5 ml) was stirred under H₂ for 3 days at 20°C. After addition of H₂O (2 ml), the mixture was further stirred under H₂ for 2 days at 20°C. Processing and trituration of the residue with EtOH-H₂O gave **24** (26 mg, 73%) as an amorphous solid. $[\alpha]_D + 25.6^\circ$ ($c=0.20$, H₂O), R_f 0.11 in 2:1:1 BuOH-AcOH-H₂O.

Anal. Calcd. for C₃₀H₅₂O₂₆·3H₂O: C, 40.82; H, 6.62. Found: C, 40.96; H, 6.71%.

*Benzyl O-(3-O-acetyl-2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1 $\rightarrow$ 3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₄-(1 $\rightarrow$ 3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside **25**.* To a mixture of molecular sieves 4A (1 g) and AgOSO₂CF₃ (200 mg) was added dropwise a solution of **23** (100 mg, 0.043 mmol) in Cl(CH₂)₂Cl (10 ml) and then a solution of **4** (30 mg, 0.056 mmol) in Cl(CH₂)₂Cl (1 ml). The mixture was stirred under argon for 5 hr and processed. Chromatography on SiO₂ C-300 (20 g) in 3:1 hexane-THF afforded **25** (88 mg, 72%). $[\alpha]_D + 11.0^\circ$ ($c=0.50$), R_f 0.45 in 10:1 toluene-EtOAc. δ_H : 1.90 (3H, s, Ac).

Anal. Calcd. for C₁₇₁H₁₇₈O₃₂: C, 74.82; H, 6.54. Found: C, 74.73; H, 6.66%.

*Benzyl O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)-[(1 $\rightarrow$ 3)-O-(2,4,6-tri-O-benzyl- α -D-mannopyranosyl)]₄-(1 $\rightarrow$ 3)-2,4,6-tri-O-benzyl- α -D-mannopyranoside **26**.* A solution of **25** (78 mg) in THF (2 ml) and 0.1 N NaOMe-MeOH (2 ml) was stirred for 3 hr at 20°C. Processing and chromatography on SiO₂ C-300 (5 g) in 10:1 toluene-EtOAc gave **26** (66 mg, 86%), $[\alpha]_D + 13.1^\circ$ ($c=0.50$), R_f

0.34 in 10:1 toluene-EtOAc.

Anal. Calcd. for C₁₆₉H₁₇₆O₃₁·0.5 C₆H₅CH₃: C, 75.36; H, 6.60. Found: C, 75.53; H, 6.61%.

*α -D-Mannopyranosyl-[(1 $\rightarrow$ 3)- α -D-mannopyranosyl]₄-(1 $\rightarrow$ 3)- α -D-mannopyranose **3**.* A mixture of **26** (60 mg) and 10% Pd-C (50 mg) in THF (1 ml) and MeOH (5 ml) was stirred under H₂ for 2 days at 20°C. After further addition of 10% Pd-C (50 mg) and MeOH (5 ml), the mixture was stirred for 3 days under H₂. Processing and trituration with EtOH-H₂O gave **3** (20 mg, 96%) as an amorphous solid. $[\alpha]_D + 26.8^\circ$ ($c=0.20$, H₂O), R_f 0.08 in 2:1:1 BuOH-AcOH-H₂O.

Anal. Calcd. for C₃₆H₆₄O₃₂·2H₂O: C, 41.38; H, 6.51. Found: C, 41.09; H, 6.61%.

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