Phenolic constituents of Chinese Quince (*Chaenomeles sinensis* Koehne) and their anti-neuroinflammatory, neurotrophic, and cytotoxic activities

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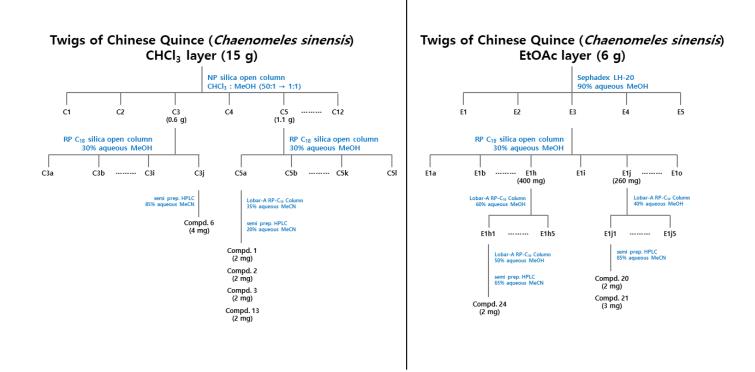
Contents

General Experimental Procedures. (Detailed isolation process)1
Scheme S1. Fractionation and isolation process of compounds 1–24 from <i>C. sinensis</i> twigs2
Figure S1. HRESIMS spectrum of 10
Figure S2. ECD spectrum of 11
Figure S3. ¹ H NMR spectrum of 1 in chloroform- <i>d</i> (700 MHz)2
Figure S4. ¹³ C NMR spectrum of 1 in chloroform- <i>d</i> (175 MHz)
Figure S5. ¹ H- ¹ H COSY spectrum of 1 in chloroform- <i>d</i>
Figure S6. HSQC spectrum of 1 in chloroform-d
Figure S7. HMBC spectrum of 1 in chloroform- <i>d</i>
Figure S8. HRFABMS spectrum of 97
Figure S9. ECD spectra of 9 (top) and 25 (bottom, adopted from the original paper)
Figure S10. ¹ H NMR spectrum of 9 in methanol- <i>d</i> ₄ (700 MHz)9
Figure S11. ¹³ C NMR spectrum of 9 in methanol- d_4 (175 MHz)10
Figure S12. ¹ H- ¹ H COSY spectrum of 9 in methanol- d_4
Figure S13. HSQC spectrum of 9 in methanol- d_4
Figure S14. HMBC spectrum of 9 in methanol-d ₄
Figure S15. Extracted ion chromatograms (EICs, m/z 431.1311) of chiral derivatized L- and D-
rhamnopyranose purchased or obtained by hydrolysis of 914
Figure S16. HRESIMS spectrum of 11
Figure S17. ¹ H NMR spectrum of 11 in methanol- <i>d</i> ₄ (700 MHz)
Figure S18. ¹³ C NMR spectrum of 11 in methanol- d_4 (175 MHz)
Figure S19. ¹ H- ¹ H COSY spectrum of 11 in methanol- d_4
Figure S20. HSQC spectrum of 11 in methanol- d_4 19
Figure S21. HMBC spectrum of 11 in methanol-d ₄
Figure S22. Extracted ion chromatograms (EICs, m/z 447.1260) of chiral derivatized D- and L-
glucopyranose purchased or obtained by hydrolysis of 1121
Figure S23. ¹ H NMR spectrum of 2 in chloroform- <i>d</i> (700 MHz)
Figure S24. ¹³ C NMR spectrum of 2 in chloroform- <i>d</i> (700 MHz)
Figure S25. ¹ H NMR spectrum of 3 in chloroform- <i>d</i> (700 MHz)24
Figure S26. ¹³ C NMR spectrum of 3 in chloroform- <i>d</i> (700 MHz)25

General Experimental Procedures. (Detailed isolation process)

The CHCl₃ soluble layer (15 g) was applied to silica gel column chromatography (CHCl₃-MeOH, 50:1 \rightarrow 1:1) to yield 12 fractions (C1-C12). Fraction C3 (600 mg) was subjected to RP-C₁₈ silica gel column (60% aqueous MeOH) to give ten subfractions (C3a-C3j). Compound 6 (4 mg) was obtained by purification of fraction C3j using semipreparative HPLC (85% aqueous MeCN). Fraction C5 (1.1 g) separated with RP-C₁₈ silica gel column with solvent system 70% aqueous MeOH to afford 12 subfractions (C5a-C5l). Subfraction C5a was fractionated over LiChroprep Lobar-A RP-C₁₈ column with 35% aqueous MeCN and further purified by semipreparative HPLC (20% aqueous MeCN) to yield compounds 1 (2 mg), 2 (2 mg), 3 (2 mg) and 13 (2 mg). The EtOAc soluble layer (3 g) was subjected to a Sephadex LH-20 column (90% aqueous MeOH) to give five fractions (E1-E5). Fraction E1 (3.8 g) was applied to silica gel column chromatography (CHCl₃-MeOH, 10:1) to yield 15 subfractions (E1a-E1o). E1h (400 mg) was chromatographed over Lichroprep Lobar-A RP-C₁₈ (60% aqueous MeOH) to give five subfractions (E1h1-E1h5). E1h1 was fractionated over Lichroprep Lopbar-A RP-C₁₈ column (50% aqueous MeOH) followed by semipreparative HPLC (65% aqueous MeCN) to yield compound 24 (2 mg). Subfraction E1j (260 mg) was subjected to LiChroprep Lobar-A RP-C₁₈ column with 40% aqueous MeOH to give five subfractions (E1j1-E1j5). Subfraction E1j1 was purified by semipreparative HPLC (15% aqueous MeCN) to yield compounds 20 (2 mg) and 21 (3 mg). n-BuOH soluble layer was applied to silica gel column chromatography eluted with CHCl₃-MeOH-H₂O (3:1:0.1) yielding 10 fractions (B1-B10). Fraction B3 (60 mg) was separated with RP-C₁₈ Sep-pak (45% aqueous MeOH) and further purified by semipreparative HPLC (35% MeOH) to yield compound 7 (2 mg). Fraction B4 (1.9 g) was fractionated by RP-C18 silica gel column (40% aqueous MeOH) to give three subfractions (B4a-B4c). Subfraction B4a (1.5 g) was further purified by semipreparative HPLC (35% aqueous MeCN) to yield compound 18 (70 mg). Further isolation of fraction B5 (500 mg) by RP-C18 silica gel column using 40% aqueous MeOH, yielded nine subfractions (B5a-B5i). Subfraction B5a was purified by semipreparative HPLC (15% aqueous MeCN) to obtain compounds 12 (5 mg) and 17 (17 mg). Subfraction B5c was purified by semipreparative HPLC (17% aqueous MeCN) to yield compounds 5 (2 mg), 9 (2 mg) and 10 (2 mg). Subfraction B5d was purified by semipreparative HPLC (17% aqueous MeCN) to give compounds 4 (5 mg) and 8 (3 mg). Subfraction B5i was purified by semipreparative HPLC (50% aqueous MeOH) to yield compounds 14 (3 mg) and 16 (4 mg). Fraction B6 (700 mg) was subjected to a RP-C₁₈ silica gel chromatography eluted with 40% aqueous MeOH to give eight subfractions (B6a-B6h). Subfraction B6a (150 mg) was purified by semipreparative HPLC (7% aqueous MeCN) to obtain compounds 22 (4 mg). Subfraction B6c (120 mg) was purified by semipreparative HPLC with solvent system 25% aqueous MeOH to give compounds 19 (34 mg). Subfraction B6d (100 mg) was chromatographed over Lichroprep Lobar-A (EtOAc-MeOH-H₂O, 5:1:0.1) and further purified by semipreparative HPLC (25% aqueous MeOH) to obtain compounds 11 (5 mg). Subfraction B6e (40 mg) was purified by semipreparative HPLC with solvent system 30% aqueous MeOH to yield compounds 23 (3 mg). Subfraction B6h was purified by semipreparative HPLC (40% aqueous MeOH) to give compounds 15 (7 mg).

Scheme S1. Fractionation and isolation process of compounds 1–24 from *C. sinensis* twigs.



Twigs of Chinese Quince (*Chaenomeles sinensis*) *n*-BuOH layer (30 g)

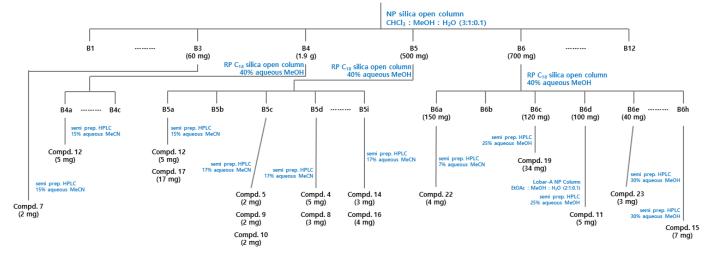
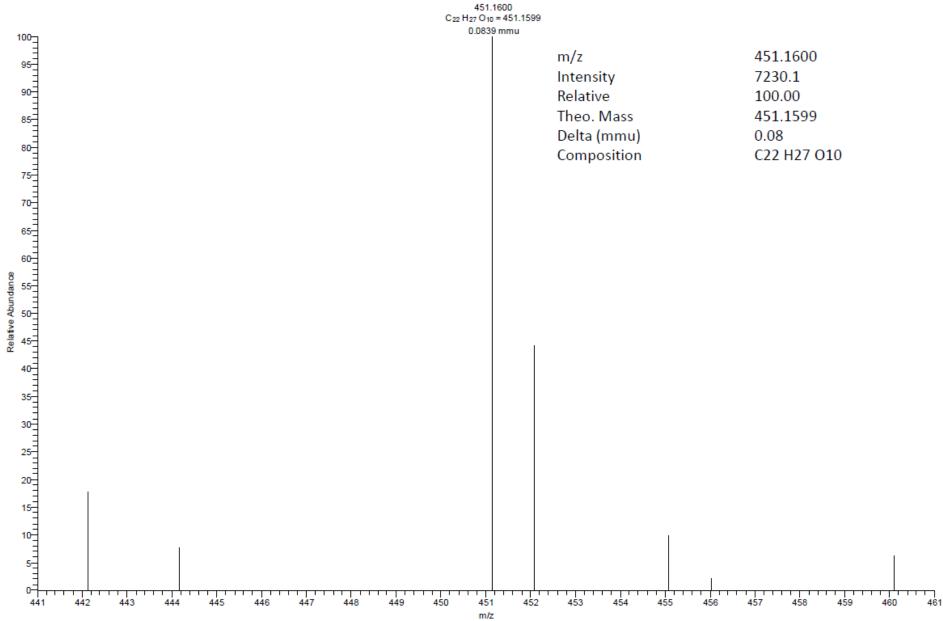


Figure S1. HRESIMS spectrum of 1

140709_MGCC7_004-c2 #26-34 RT: 0.53-0.69 AV: 9 SB: 2 0.38-0.40 NL: 7.23E3 T: + c FAB Full ms [995:601-600.50]



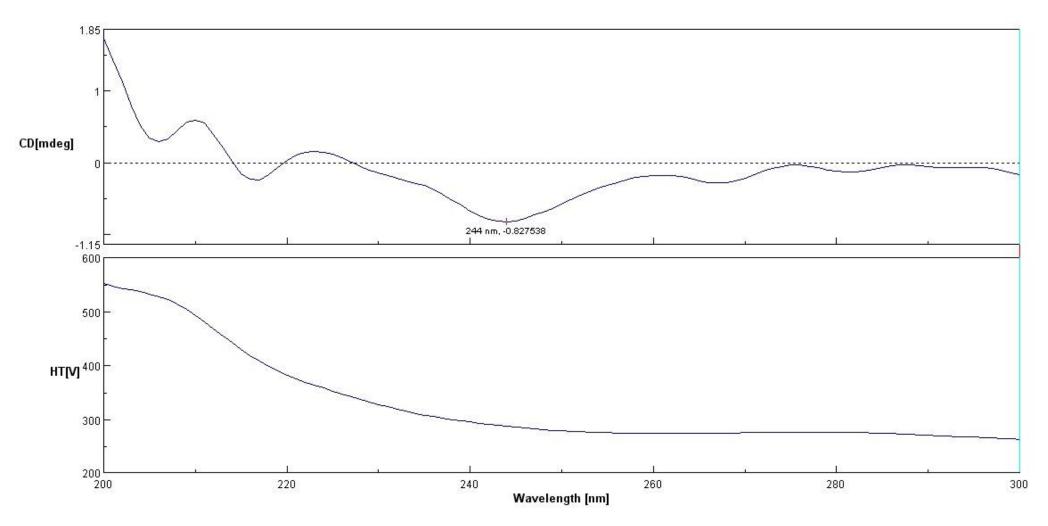


Figure S3. ¹H NMR spectrum of 1 in chloroform-*d* (700 MHz)

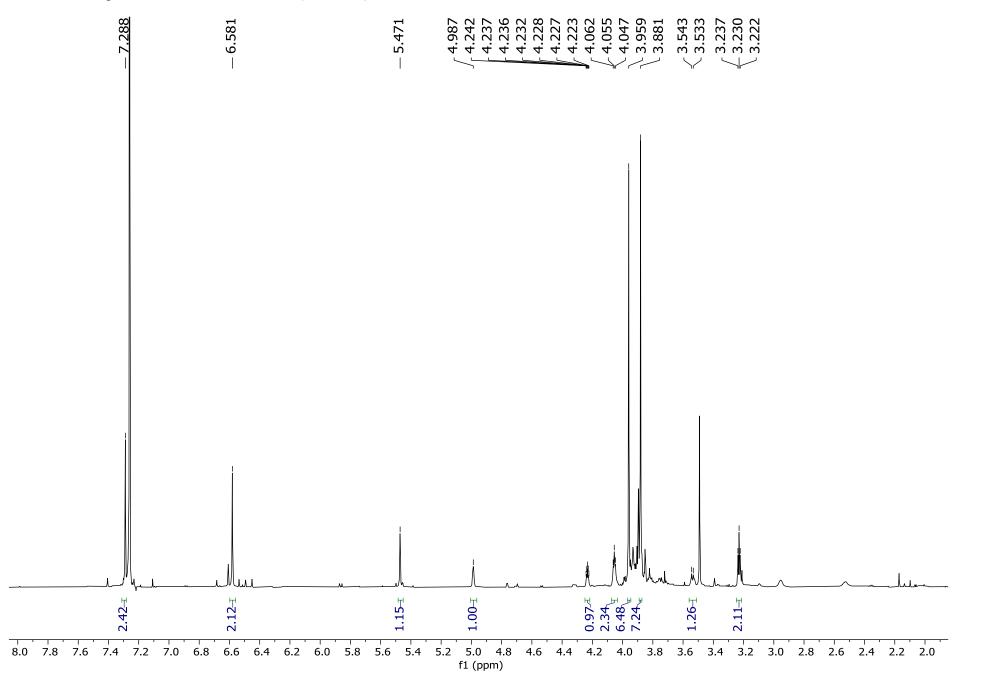


Figure S4. ¹³ C NMR	spectrum of 1 in chloroform	n- <i>d</i> (175 MHz)
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— 199.09			- 153.57	- 139. - 139.	\int 134.21 \sim 133.08 \sim 130.32		~ 105.59 ~ 102.69	- 87.50	— 73.09	60.69 58.28 56.61 56.52	40.44		

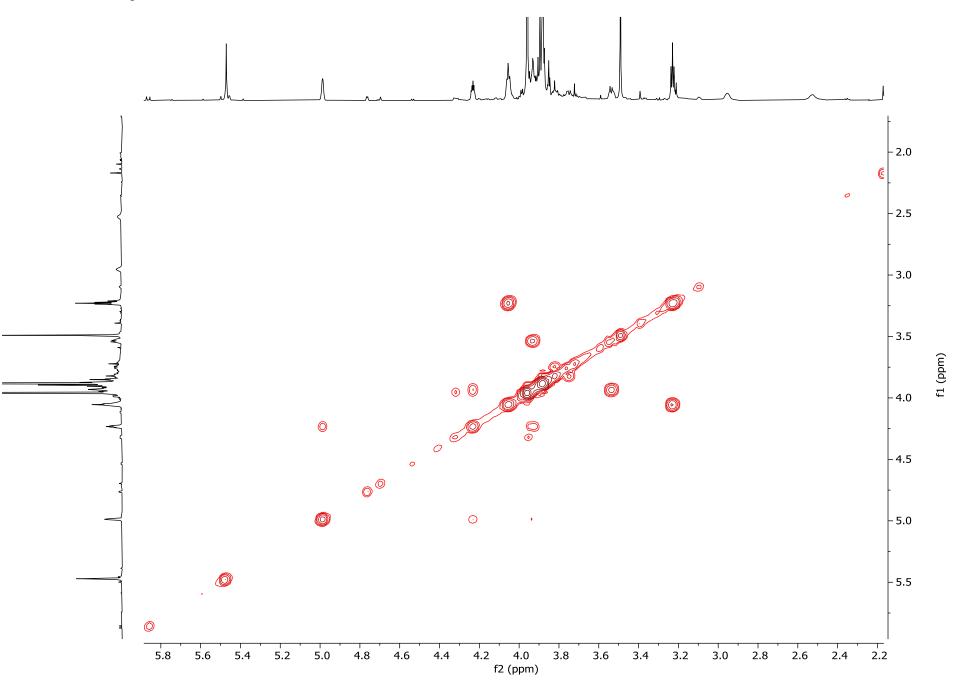
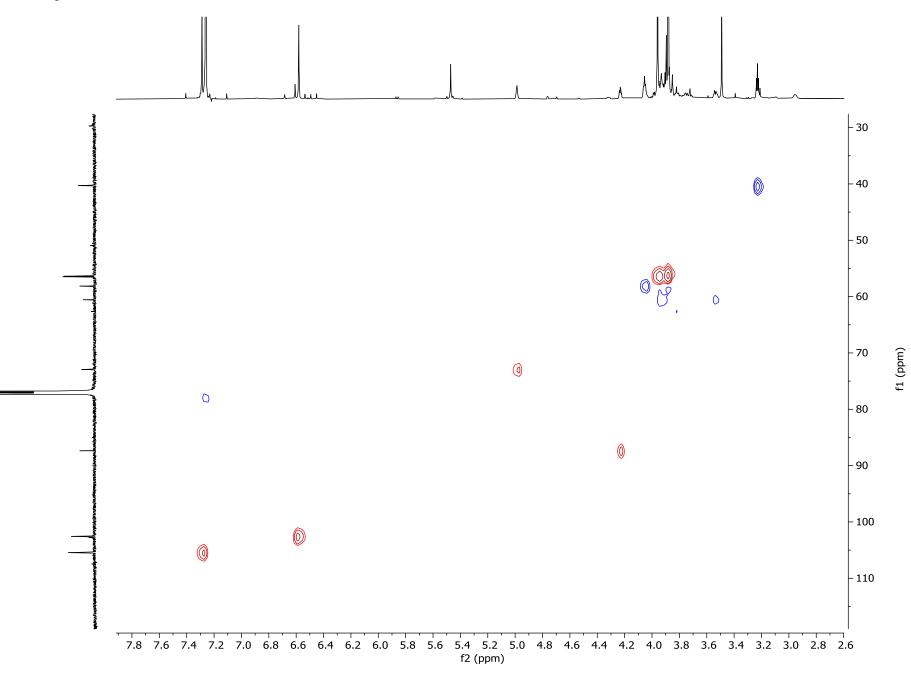


Figure S6. HSQC spectrum of 1 in chloroform-*d*



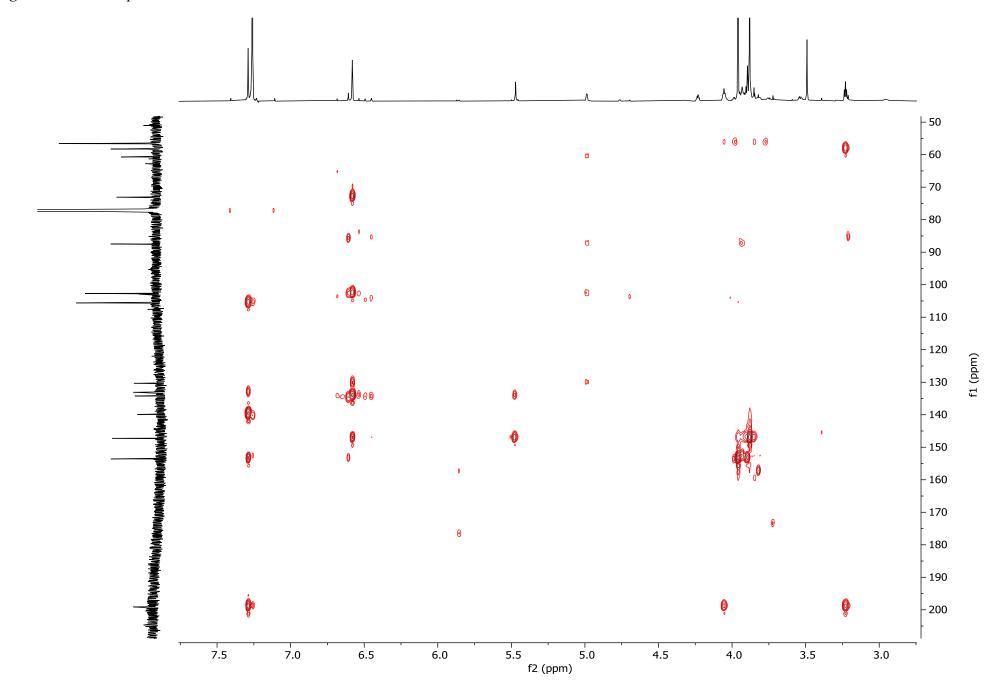


Figure S7. HMBC spectrum of 1 in chloroform-*d*

Figure S8. HRFABMS spectrum of 9

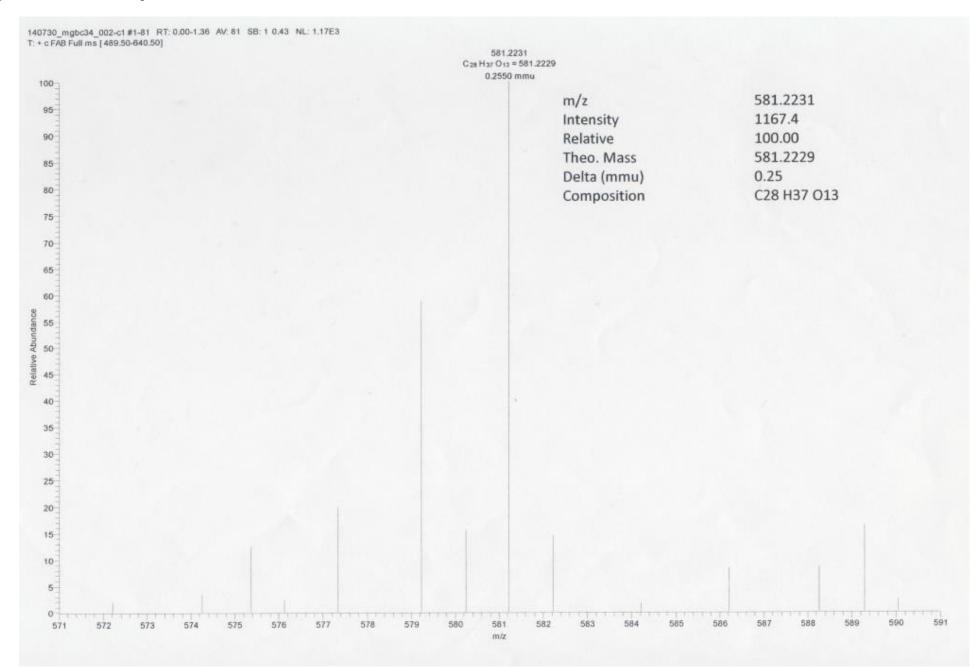
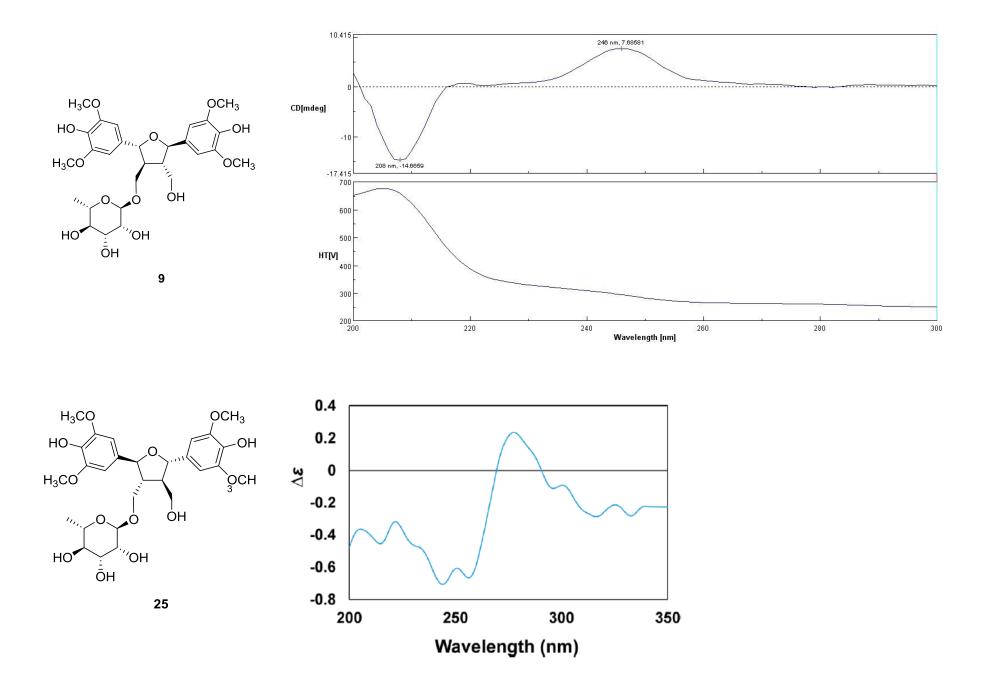


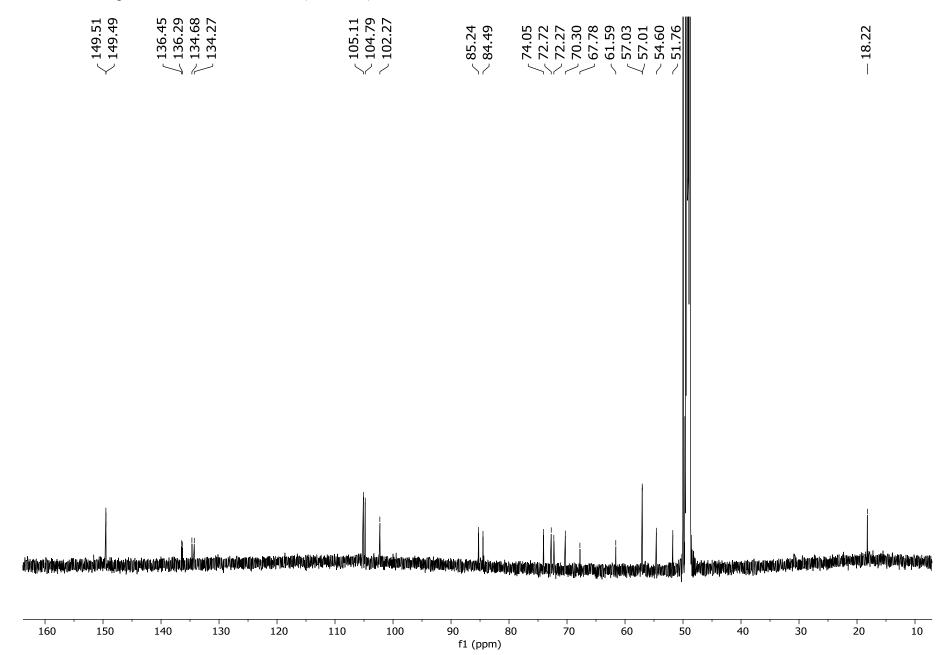
Figure S9. ECD spectra of 9 (top) and 25 (bottom, adopted from the original paper).



6.762 6.754 5.50705.0705.0735.0795.0735.0793.8153.8153.8153.8153.8153.8153.8153.8153.8133.8133.8133.8133.8133.8133.8133.8253.6503.6503.6553.6503.6553.6553.6503.6553.6503.6553.6553.6503.6553.6553.6553.6553.6553.6553.6553.6553.6553.6553.5Ü, 0.94 1.00 ≩ ዞ 번 번 ቸ 붜 ыння 12.52 0.99 1.06 1.11 1.02 1.06 3.28 3.92 ∞ 29 2 -// 7.5 5.0 7.0 2.5 1.5 6.5 6.0 5.5 4.5 4.0 3.5 3.0 2.0 1.0 0.5 f1 (ppm)

Figure S10. ¹H NMR spectrum of **9** in methanol-*d*₄ (700 MHz)

Figure S11. ¹³C NMR spectrum of 9 in methanol-*d*₄ (175 MHz)



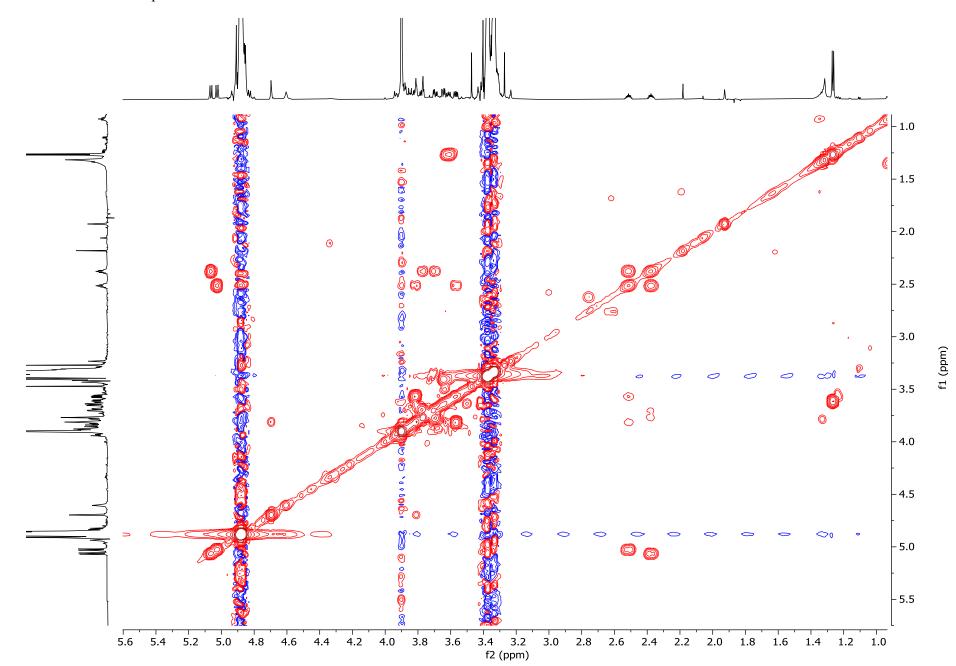


Figure S12. ¹H-¹H COSY spectrum of **9** in methanol-*d*₄

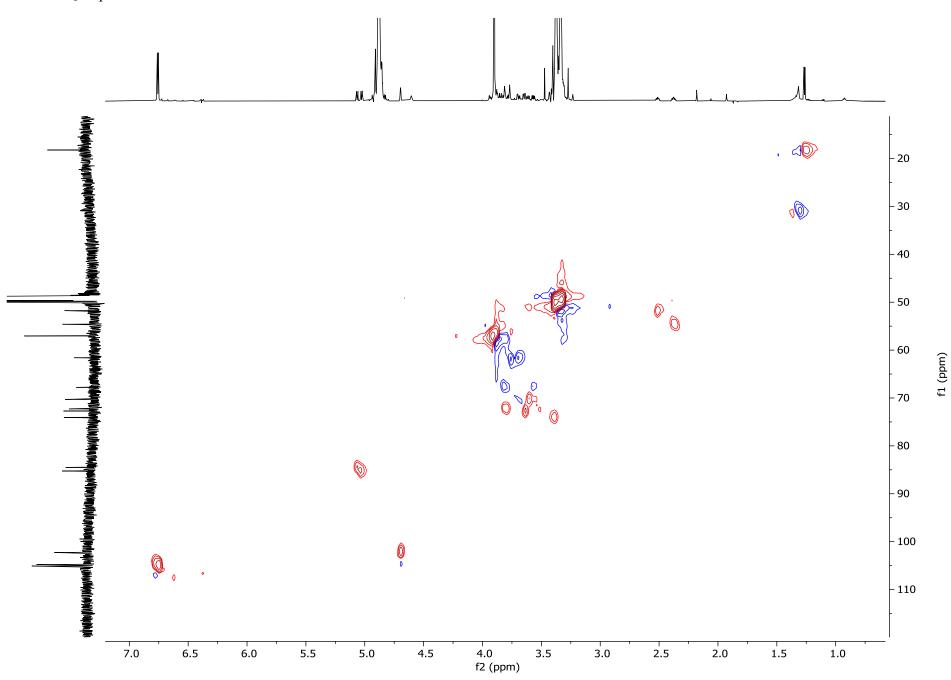


Figure S13. HSQC spectrum of 9 in methanol-*d*₄

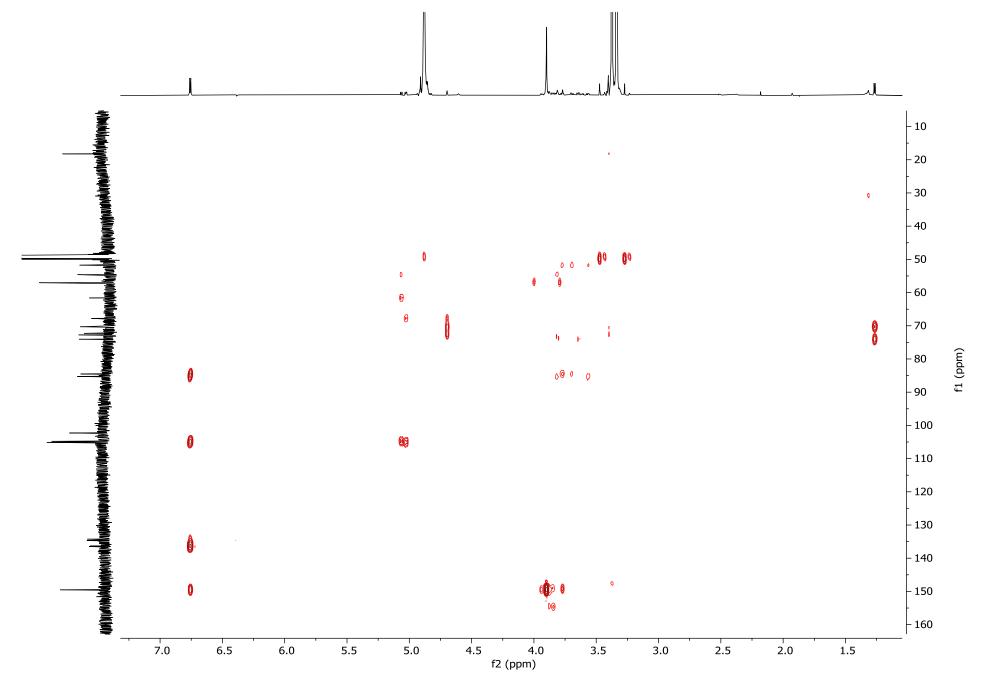


Figure S14. HMBC spectrum of 9 in methanol-d₄

Figure S15. Extracted ion chromatograms (EICs, *m/z* 431.1311) of chiral derivatized L- and D-rhamnopyranose purchased or obtained by hydrolysis of 9.

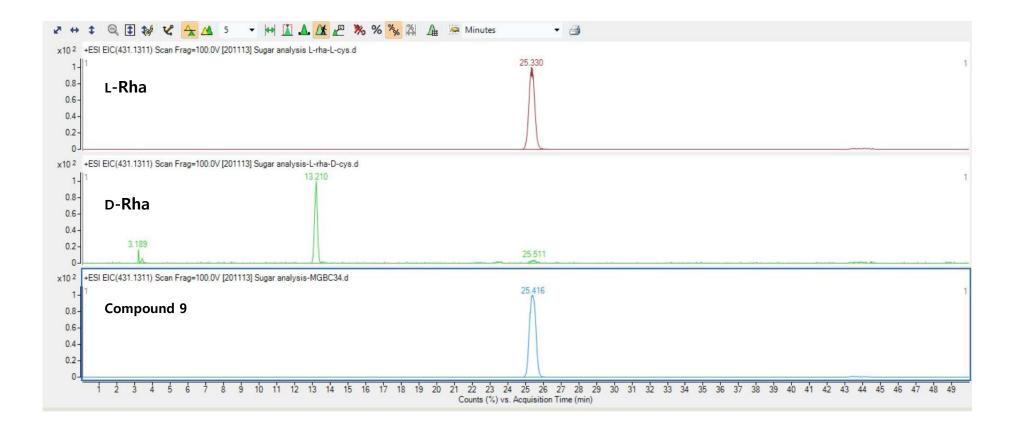
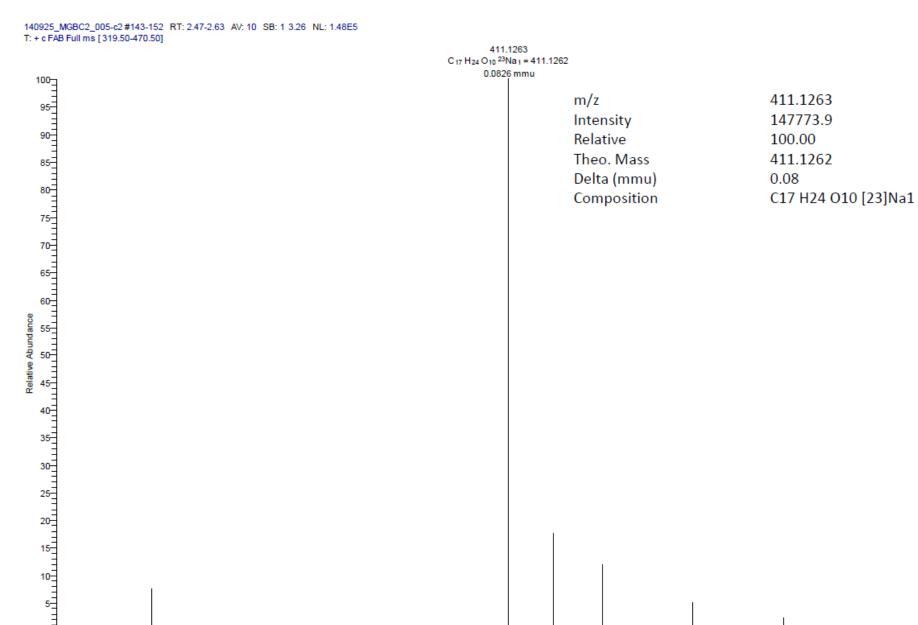


Figure S16. HRESIMS spectrum of 11



m/z

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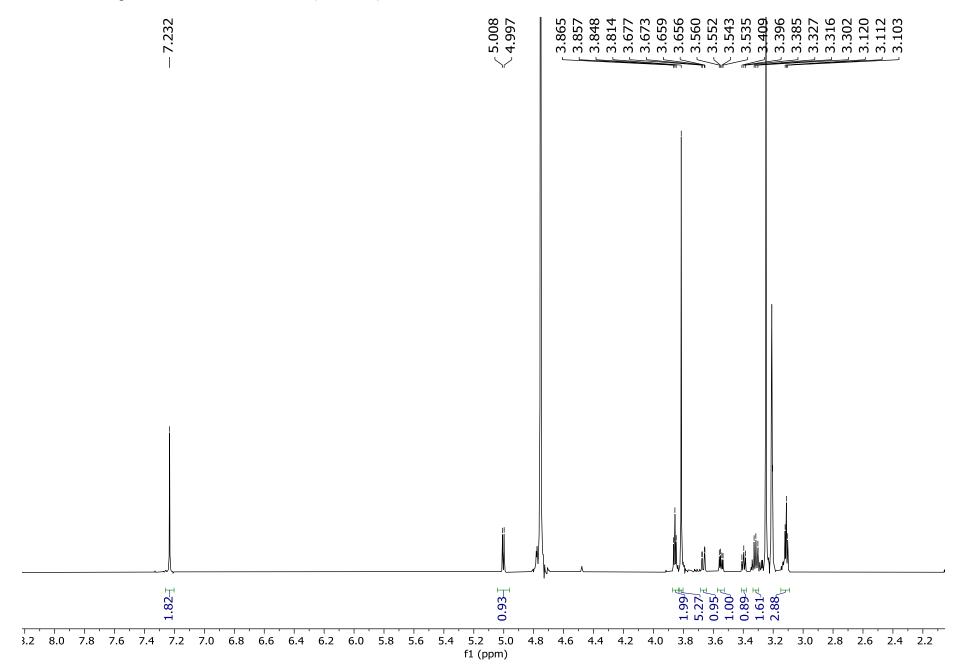
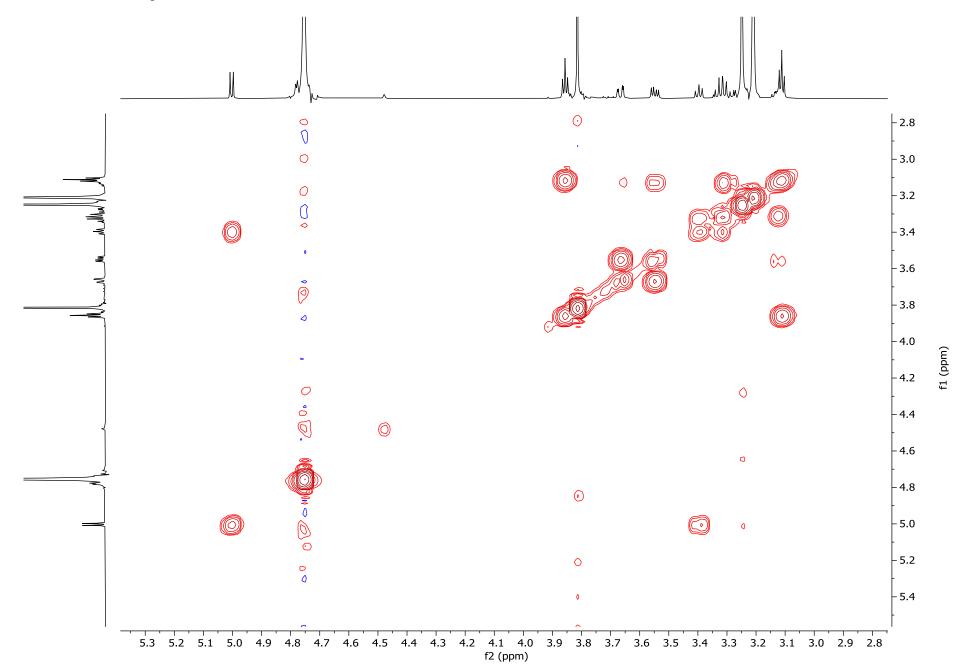
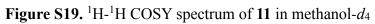
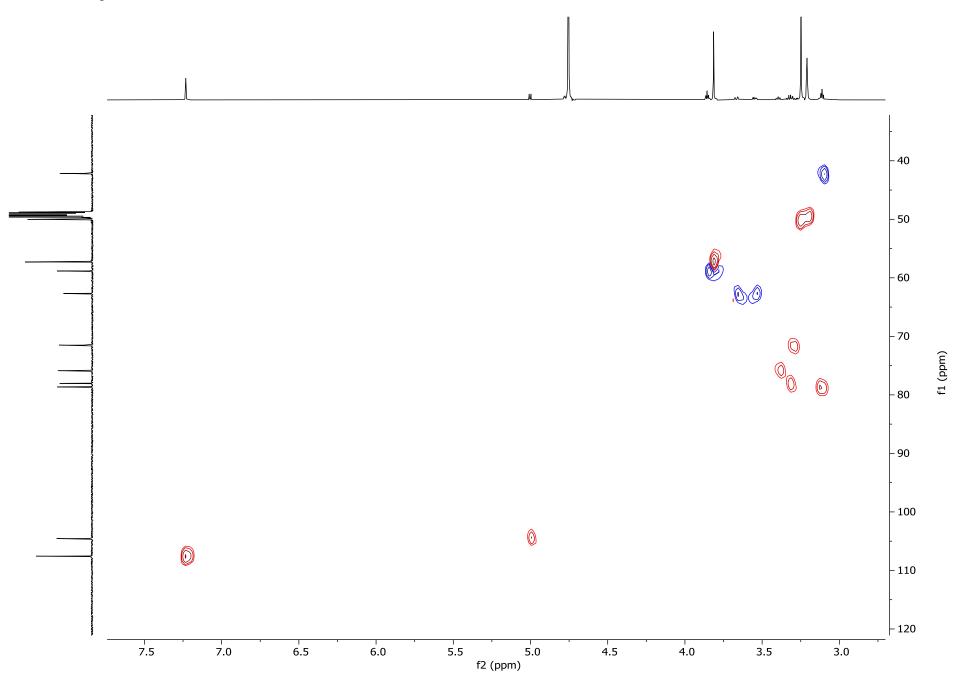


Figure S18. ¹³C NMR spectrum of **11** in methanol- d_4 (175 MHz)

- 199.96	— 154.44	— 140.70 — 134.57	- 107.56 104.56	∑ 78.63 ∑ 78.03 ∑ 75.87 ∑ 71.51	 7 58.83 57.28 42.15
210 200 190 1	80 170 160 150	140 130 f1 (pp	L20 110 100 90 m)	80 70	60 50 40 30







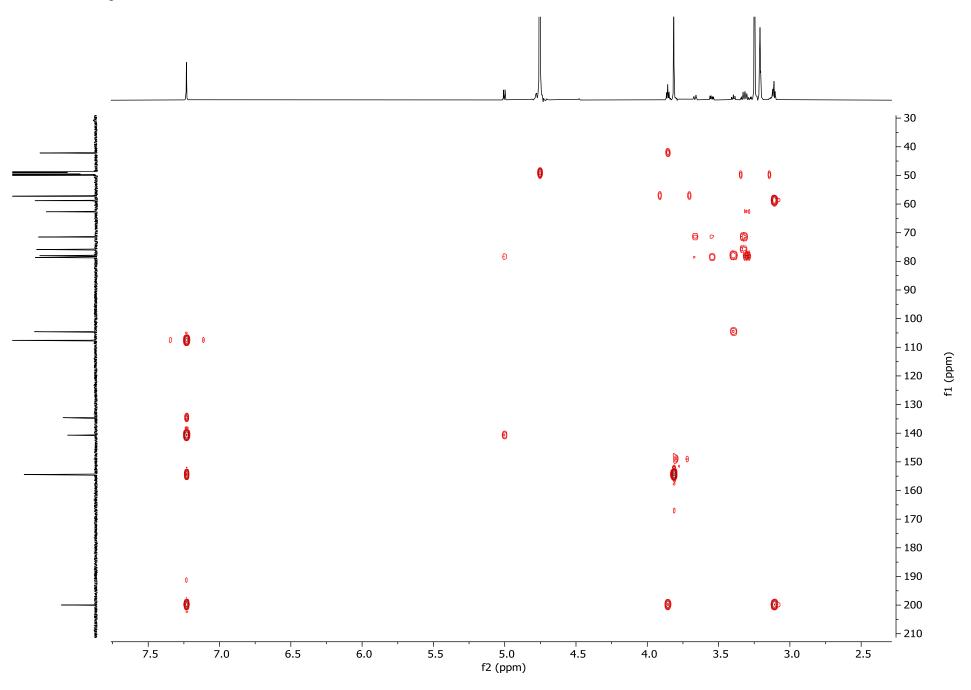
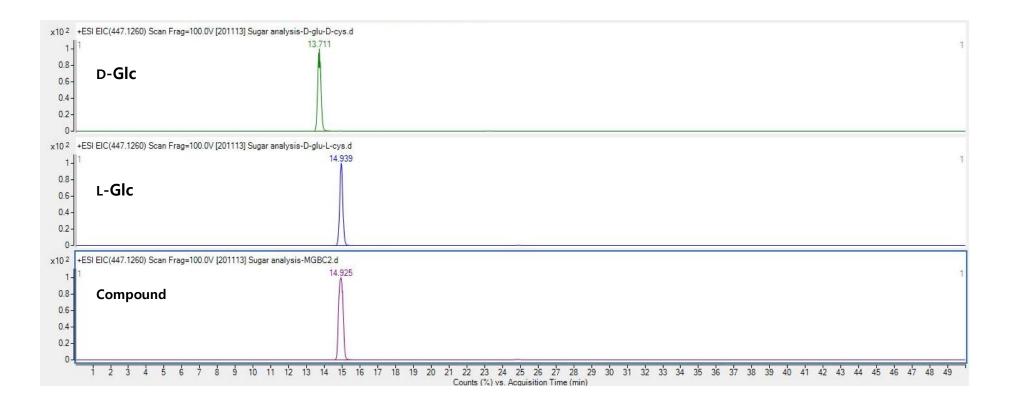


Figure S22. Extracted ion chromatograms (EICs, *m/z* 447.1260) of chiral derivatized D- and L-glucopyranose purchased or obtained by hydrolysis of 11.



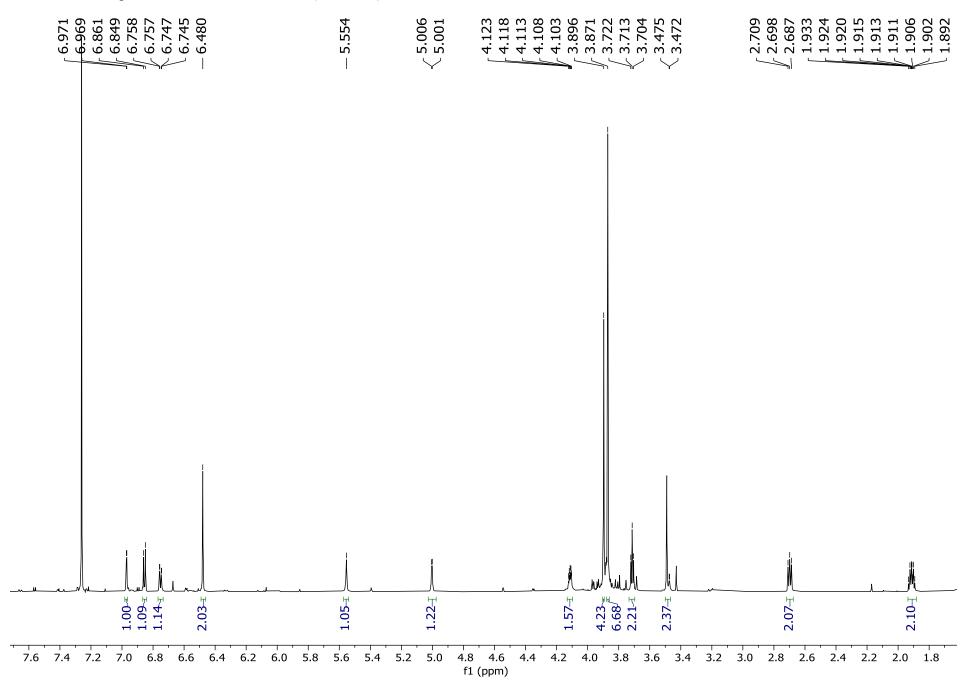


Figure S23. ¹H NMR spectrum of **2** in chloroform-*d* (700 MHz)

Figure S24. ¹³C NMR spectrum of **2** in chloroform-*d* (700 MHz)

- 153.28	— 138.79 — 133.06	— 105.47	- 87.19	- 72.62	 62.34 60.77 56.14 	~ 34.37 ~ 32.75
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165 160 155 150 1	L45 140 135 1	30 125 120 115 110 105	100 95 90 85 f1 (ppm)	80 75 70	65 60 55 50	45 40 35 30

4.110 4.106 4.100 4.095 4.090 3.878 3.878 3.719 3.719 3.710 3.455 3.455 - 4.994 - 4.990 6.587 6.492 2.718 2.707 2.696 $\begin{array}{c} 1.939\\ 1.930\\ 1.921\\ 1.919\\ 1.917\\ 1.912\\ 1.912\\ 1.908\\ 1.899\end{array}$ 5.454 11 14.23∃ ㅠㅜ Ħ ተ Ч Ч Ч Ч Ψ 1.07 2.09 1.98 1.00 1.15 1.33 1.901.98 2.12 7.4 7.2 7.0 6.8 6.6 6.4 6.2 6.0 5.8 5.6 5.4 5.2 5.0 4.8 4.6 4.4 4.2 4.0 3.8 3.6 3.4 3.2 3.0 2.8 2.6 2.4 2.2 2.0 1.8 f1 (ppm)

Figure S25. ¹H NMR spectrum of **3** in chloroform-*d* (700 MHz)

Figure S26. ¹³C NMR spectrum of **3** in chloroform-*d* (700 MHz)

	- 153.31	— 147.19	-138.87 $\int 134.00$ $\int 133.08$	130.61		105 46	— 102.63 — 102.63		- 87.24		72.74	62.33	- 60.77 56.50 56.38				~ 34.36 ~ 32.76	
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