Synthesis of β-analogues of *C*-mannosyltryptophan, a novel *C*-glycosylamino acid found in proteins†

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α-*C*-Mannosyltryptophan (α-C-Man-Trp) has been found to be a novel post-translational modification of tryptophan found from some biologically important glycoproteins. In order to analyze the biological functions of α-C-Man-Trp, we have developed an efficient synthetic strategy for α-C-Man-Trp and its glucose and galactose analogues, starting from α-*C*-glycosidation of the corresponding hexapyranoside derivatives with tinacetylene. According to the synthetic routes, we describe here syntheses of β-anomers of C-Man-Trp, and its glucose and galactose analogues from the corresponding β-*C*-glycosylacetylenes. During this study, we have developed a highly stereocontrolled synthesis of β-*C*-mannosylacetylene that is required for the synthesis of β-C-Man-Trp, while the precedented method gave an anomeric mixture of the *C*-mannosylacetylene. The synthetic C-Man-Trp and its analogues were analyzed by HPLC.

Introduction

α-*C*-Mannosyltryptophan (α-C-Man-Trp (**1**) in Fig. 1) was first found as a novel linkage "*C*-glycoside" between a protein and carbohydrate from human ribonuclease 2 (RNase 2).[1] Interestingly, the mannose moiety of α-C-Man-Trp adopts a $^1C_4$ conformation as a major conformer due to a bulky tryptophan moiety occupying the α-position of the anomeric center and a lack of anomeric effect. Since its discovery, C-Man-Trp has been identified in several biologically important proteins such as interleukin-12,[2] the terminal four components of a complement system (C6, C7, C8α, β and C9),[3] properdin,[4] thrombospondin,[5] F-spondin,[6] mucins (MUC5AC and MUC5B),[7] and erythropoietin receptors.[8] Extensive studies on the biosynthetic pathway have revealed that "*C*-mannosyltransferase," still not identified as an enzyme for installation of mannose to tryptophan, recognizes the amino acid sequence W–X–X–W to modify the first Trp of this motif.[9] Since the recognition sequence is included in conserved sequences such as TSP-1 (W–X–X–W–X–X–W–X–X–C) and the WS motif (W–S–X–W–S) in thrombospondin type 1 repeats (TSRs),[10] it is likely that other proteins with these motifs may be modified by *C*-mannosylation. On the other hand, the biological functions of α-C-Man-Trp have not been clarified, although several possibilities have been studied.[11] In order to identify α-C-Man-Trp from proteins, peptide sequencing utilizing Edman degradation, the MS/MS method,[12] and specific antibodies against α-C-Man-Trp[13–15] have been employed; however, such methods have not been able to exclude the possibility of other stereoisomers having a different carbohydrate moiety with the same molecular weight as that of α-C-Man-Trp. Although NMR spectroscopy is the only way to discriminate these isomers, a sufficient amount of the sample containing the modification is difficult to obtain. Thus, a new analytical method to differentiate the isomers of α-C-Man-Trp has been highly desired.[3,6,8] To supply the α-C-Man-Trp for analyzing its biological functions, we have previously synthesized α-C-Man-Trp, and its glucose and galactose analogues,[16,17] and its probes for use in biochemical studies.[18] The corresponding β-isomers, however, have been reported to be obtained in only minute amounts by heating L-tryptophan with D-mannose, galactose, and glucose in the presence of acid.[19] We therefore describe herein stereocontrolled syntheses of the β series of C-Man-Trp and its glucose and galactose analogues, which can be employed as authentic samples for their trace analyses.

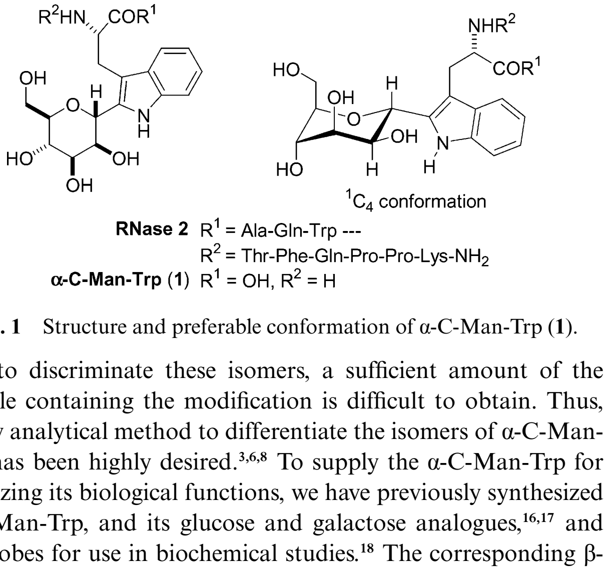

Fig. 1 Structure and preferable conformation of α-C-Man-Trp (**1**).

Synthetic plan for β-*C*-glycosyltryptophan

In our previous papers, we established the synthesis of α-C-Man-Trp and its glucose and galactose analogues as outlined in Scheme 1. Thus, *C*-glycosidation of glycosyl-1-acetate **2** with tinacetylene in the presence of TMSOTf as a Lewis acid exclusively gave the corresponding α-*C*-glycosylacetylene **3**,[20] which was coupled with *N*-Ts-*o*-iodoanilide **4** by palladium catalyst to *o*-ethylaniline derivatives **5**. Copper(I)-mediated indole formation was followed by deprotection of the Ts group to give α-*C*-glycosylindole **6**, which reacted with L-serine-derived aziridine carboxylate **7** in the presence of $Sc(ClO_4)_3$ as a Lewis acid[21] to afford a fully protected α-C-Gly-Trp **8**. Finally, two-step

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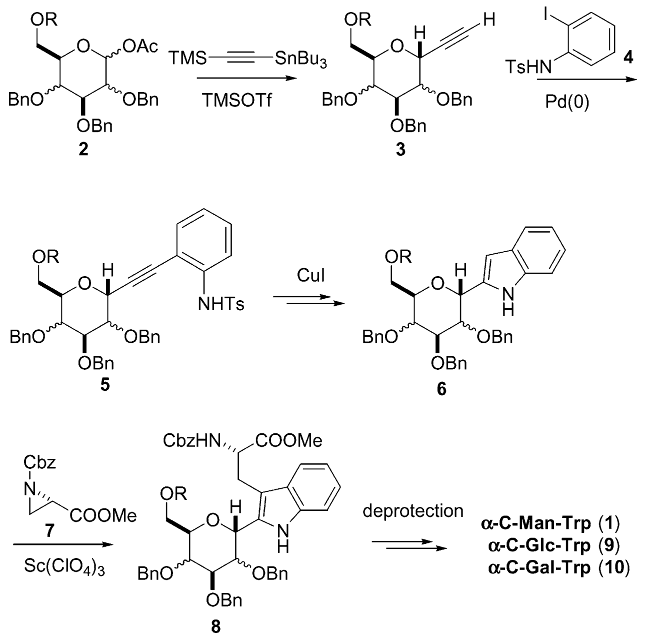


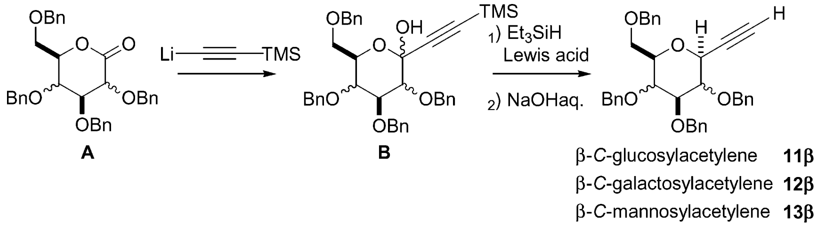
Scheme 1 Synthesis of α-C-Man-Trp and its analogues.

deprotection furnished α-*C*-mannosyltryptophan (**1**, α-C-Man-Trp), α-*C*-glucosyltryptophan (**9**, α-C-Glc-Trp), and α-*C*-galactosyltryptophan (**10**, α-C-Gal-Trp). Based on this synthesis, we planned to synthesize the β-series of the analogues starting from β-*C*-glycosylacetylenes.

Highly stereoselective synthesis of β-*C*-mannosylacetylene

The β-*C*-glycosylacetylenes, including glucose, galactose, and mannose, were required as the starting materials. Meldal and Vasella *et al.*[22] have reported the syntheses of these β-sugar acetylenes as tetrabenzyl ether according to Kishi's protocol,[23] a widely employed and reliable synthetic method for β-*C*-hexapyranosides (Scheme 2); addition of lithium acetylide to the corresponding sugar lactones **A** followed by reduction of the resulting ketal **B** with triethylsilane in the presence of a Lewis acid.[24] The synthetic sequence enabled us to synthesize β-*C*-glucosylacetylene **11β** and β-*C*-galactosylacetylene **12β** in highly stereoselective manner.[22] However, when the same procedure was applied to D-mannopyranolactone, reduction of the corresponding ketal proceeded in a low stereoselective manner, giving a 1 : 2 mixture of α- and β-*C*-mannosylacetylenes **13** (see Table 1). Kishi has also reported that the reduction of an allyl-substituted ketal of mannose gave a 1 : 1 mixture of the anomeric isomer. Many efforts have been reported to achieve stereoselective synthesis of β-*C*-mannosides,[25,26] however, no stereocontrolled synthesis of β-*C*-mannosylacetylene has been reported to date.[27] We thus initiated our studies from the perspective of improving the stereoselectivity in the synthesis of β-*C*-mannosylacetylene **13β**, a starting material for β-C-Man-Trp.

The aforementioned results imply that the β-benzyloxy substituent at the C-2 position of the mannose derivative induces a different conformation of the oxocarbenium cation from those of the corresponding glucose and galactose derivatives. This assumption might be supported by a recent study of Shuto and co-workers,[28] from which it was reported that conformationally restricted ketal derived from a 4,6-benzylidene acetal-protected mannopyranolactone undertook a highly stereoselective reduction with Et_3SiH and TMSOTf to exclusively afford β-*C*-mannoside. We supposed that even if the transition state conformation is different from those of the corresponding glucose and galactose intermediates, the α-face of the oxocarbenium cation should be less hindered and therefore sterically hindered silanes would improve the stereoselectivity of the reduction. We thus examined the influence of sterically hindered silanes in the reduction of ketal **16**, which was prepared from 2,3,4,6-tetra-*O*-benzylmannose (**14**) *via* the corresponding lactone **15** (Scheme 3 and Table 1).[29] To our delight, tris(trimethylsilyl)silane $(TMS)_3SiH$, one of the most hindered silanes, reduced the ketal **16** to afford β-*C*-mannosylacetylene **13β** as a single isomer (entry 2). Further examination led us to find that triisopropylsilane (i-Pr_3SiH) exhibits a high stereoselectivity, giving **13β** in 67% yield along with a trace amount of **13α**.[30] Since the coupling constant between

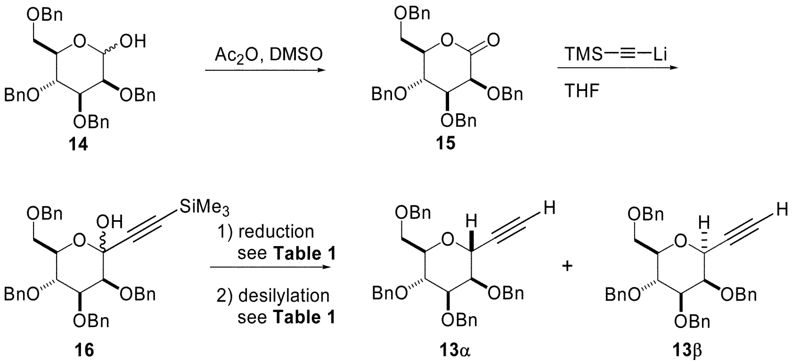


Scheme 2 General synthetic route for β-*C*-glycosylacetylenes.

Table 1 Stereoselectivities of the reduction of ketal **16** with silanes

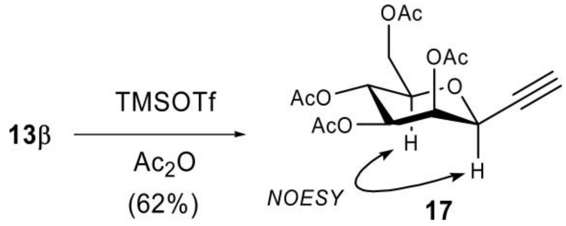
	Conditions for 1)[a]			Products	
Entry	Silane	Temp.	Conditions for 2)	Yield[b]	**13α** : **13β**[c]
1	Et_3SiH	−40 °C	TBAF	56% (from **14**)	1 : 2
2	$(TMS)_3SiH$	0 °C	TBAF	76 (from **16**)	0 : 100
3	i-Pr_3SiH	0 °C	K_2CO_3, MeOH	67 (from **16**)	1 : 99

[a] All reactions were performed with $BF_3 \cdot OEt_2$ as a Lewis acid in CH_3CN–CH_2Cl_2 (85 : 15). [b] The yields were isolated yields. [c] The ratios were determined by ^1H NMR.



Scheme 3 Synthesis β-*C*-mannosylacetylene **13β**.

the protons at the C-1 and C-2 positions of **13α** and **13β** are very close ($J = 2.0$ Hz for **13α**, $J = 1.0$ Hz for **13β**), the α-configuration of **13β** was confirmed by the NOESY spectrum of tetraacetate **17** obtained by acetolysis of **13β**[22] (Scheme 4).



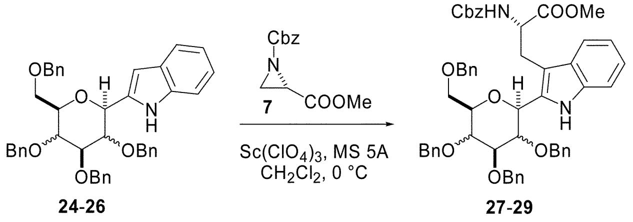
Scheme 4 Proof of the configuration of **13β**.

Synthesis of fully protected β-*C*-glycosyltryptophan

With three β-*C*-glycosylacetylenes (**11β**, **12β** and **13β**) from glucose, galactose, and mannose in hand, we next transformed these products to β-*C*-glycosylindoles, as shown in Table 2, according to our previous studies. The details are including in the supporting information†. It is worth noting that in the case of mannosylacetylene **13β**, the cross coupling between **13β** and **4** gave a mixture of **20** and **23** in a *ca.* 3.5 : 1 ratio,[31] which could be directly transformed into **26** by treatment with TBAF in refluxing THF.[32]

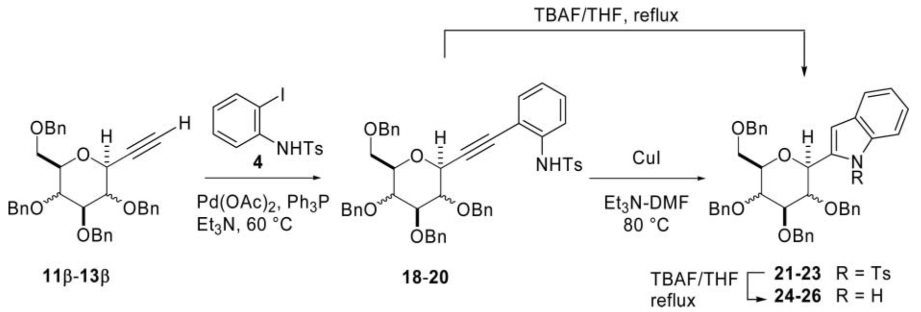
Coupling of β-*C*-glycosylindoles **24–26** with aziridine carboxylate **7** was then carried out as shown in Table 3 to afford β-C-Man-Trp and its glucose and galactose analogues in moderate yields.

Table 3 Sc(ClO$_4$)$_3$-promoted coupling of glycosylindole (**24–26**) with the aziridine **7**



Entry	Substrate		Product	Yield (%)
1	Glucose	**24**	**27**	62
2	Galactose	**25**	**28**	39
3	Mannose	**26**	**29**	49

Table 2 Pd-catalyzed coupling between glycosylacetylenes (**11β–13β**) and *N*-Ts-*o*-iodoanilide **4**, and synthesis of β-*C*-glycosylindoles (**24–26**)



			Pd coupling with **4**		Synthesis of indole		Deprotection of Ts group	
Entry	Glycosylacetylene		Products	Yield (%)	Products	Yield (%)	Products	Yield (%)
1	**11β**	Glucose	**18**	86	**21**	91	**24**	94
2	**12β**	Galactose	**19**	93	**22**	90	**25**	92
3	**13β**	Mannose	**20**	67[a]	**23**	87	**26**	Quant.

[a] The indole **23** was isolated in 19% as a by-product.

Deprotection

Deprotection of the methyl ester and benzyl groups of **27**, **28** and **29** would then be carried out according to the synthesis of analogues of α-C-Man-Trp (**1**). However, many modifications of the conditions were necessary for satisfactory yields of the final products **33**, **34** and **35**.

First, methyl esters of the fully protected C–Gly–Trp (**27–29**) were hydrolyzed with aqueous lithium hydroxide (Table 4). In the syntheses of analogues of α-C-Man-Trp (**1**), 2-propanol was employed as a solvent for the hydrolysis, however, the corresponding β-analogues **27–29** were found to be not sufficiently soluble in 2-propanol. After some experiments, we found a 1 : 1 mixture of acetonitrile and methanol to be a suitable solvent.[33] Under the improved conditions, the esters of **29**, **28**, and **27** were hydrolyzed to give **30**, **31**, and **32** in good to moderate yields (entries 1, 2, and 4).

Benzyl groups of tetrabenzyl β-C-Man-Trp **30** were then removed under hydrogenolytic conditions with 5% Pd–C in methanol (Table 4). Because of our initial concern regarding a reductive opening of the pyranose ring that we encountered in the synthesis of α-C-Man-Trp, we were reluctant to add hydrochloric acid. However, debenzylation of **30** was very sluggish in the absence of the acid. Fortunately, the debenzylation proceeded in the presence of concentrated hydrochloric acid to give β-C-Man-Trp (**33**) in moderate yield (entry 1). Under these conditions, the ring-opening by-product was not observed. When deprotection of tetrabenzyl C-Gal-Trp (**31**) was conducted with 5% Pd–C in methanol in the presence of 1 N HCl for 42 hours, β-C-Gal-Trp (**34**) was obtained in 54% yield along with the corresponding *N*-methyl C-Gal-Trp in 9% yield (entry 2). However, we found that a shorter time (18.5 hours) suppressed the side reaction to give a 69% yield of the desired product **34** with a trace amount of the *N*-methyl product (entry 3). In the deprotection of tetrabenzyl β-C-Glc-Trp (**32**), we found that both the substrate **32** and the product **35** were not sufficiently soluble in methanol, resulting in lower yields. A mixture of dioxane and water was found to be a better solvent,[19] and β-C-Glc-Trp **35** was thus obtained in good yield (entry 4). The NMR spectra of these synthetic materials are in good agreement with those of the literature.[18]

HPLC analysis of the synthetic analogues of C-Man-Trp

With all six possible isomers (**1**, **9**, **10**, **33**, **34**, and **35**) of C-Man-Trp in hand, we next analyzed the synthetic compounds by HPLC with an ODS column attached to UV and fluorescence detectors. Fortunately, all six isomers were clearly separated under conventional conditions as shown in Fig. 2. Interestingly, only β-C-Man-Trp (**33**) was eluted at a completely different retention time from those of other analogues.

Conclusion

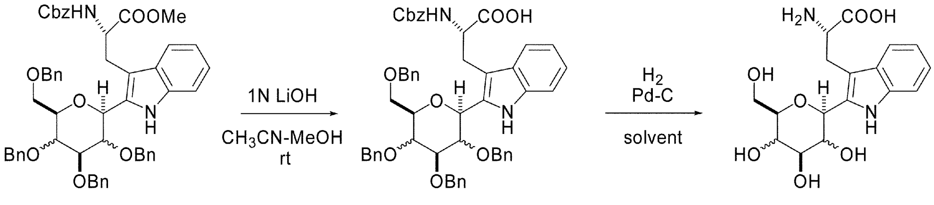
The synthesis of β-C-Man-Trp and its Glc, Gal analogues was carried out in a highly stereoselective manner. The synthesized materials should be useful as authentic samples for developing new analytical methods.

Experimental

General

Optical rotations were measured on a JASCO DIP-370 digital polarimeter. Infrared spectra (IR) were recorded on a JASCO FT/IR-8300 spectrophotometer and are reported in wavenumbers (cm^{-1}). Proton nuclear magnetic resonance (^1H NMR) spectra were recorded on a Bruker AMX-600 (600 MHz), a Bruker ARX-400 (400 MHz), a Bruker AVANCE-400 (400 MHz) or a Varian Gemini-2000 (300 MHz) spectrometer. Data are reported as follows; chemical shift, integration, multiplicity (s = singlet, d = doublet, t = triplet, br = broadened, m = multiplet), coupling constant and assignment. Carbon nuclear magnetic resonance (^{13}C NMR) spectra were recorded on a Bruker AMX-600 (150 MHz), a Bruker ARX-400 (100 MHz), a Bruker AVANCE-400 (100 MHz) or a Varian Gemini-2000 (75 MHz) spectrometer. High resolution mass spectra (HRMS) were recorded on a JEOL JMS-700 or a LC-MATE spectrometer and are reported in *m/z*. Elemental analyses were performed by the Analytical Laboratory at the Graduate School of Bioagricultural Sciences, Nagoya University. Reactions were monitored by thin-layer chromatography (TLC) on 0.25 mm silica gel coated glass plates 60 F_{254} (Merck. #1.05715).

Table 4 Deprotection of esters and benzyl groups



27-29 **30-32** **33-35**

			Deprotection of ester		Deprotection of benzyl groups					
Entry	Substrate		Product	Yield	Cat.	Additive	Solvent	Time/h	Product	Yield (%)
1	**29**	Mannose	**30**	86%	5% Pd–C	12 N HCl	MeOH	25	**33**	43
2	**28**	Galactose	**31**	64	5% Pd–C	1 N HCl	MeOH	42	**34**[a]	54
3	—	—	—	—	5% Pd–C	1 N HCl	MeOH	18.5	**34**	69
4	**27**	Glucose	**32**	64	10% Pd–C	1 N HCl	Dioxane–H_2O	7	**35**	Quant.

[a] *N*-Methyl-C-Gal-Trp was obtained as a by-product in 9% yield.

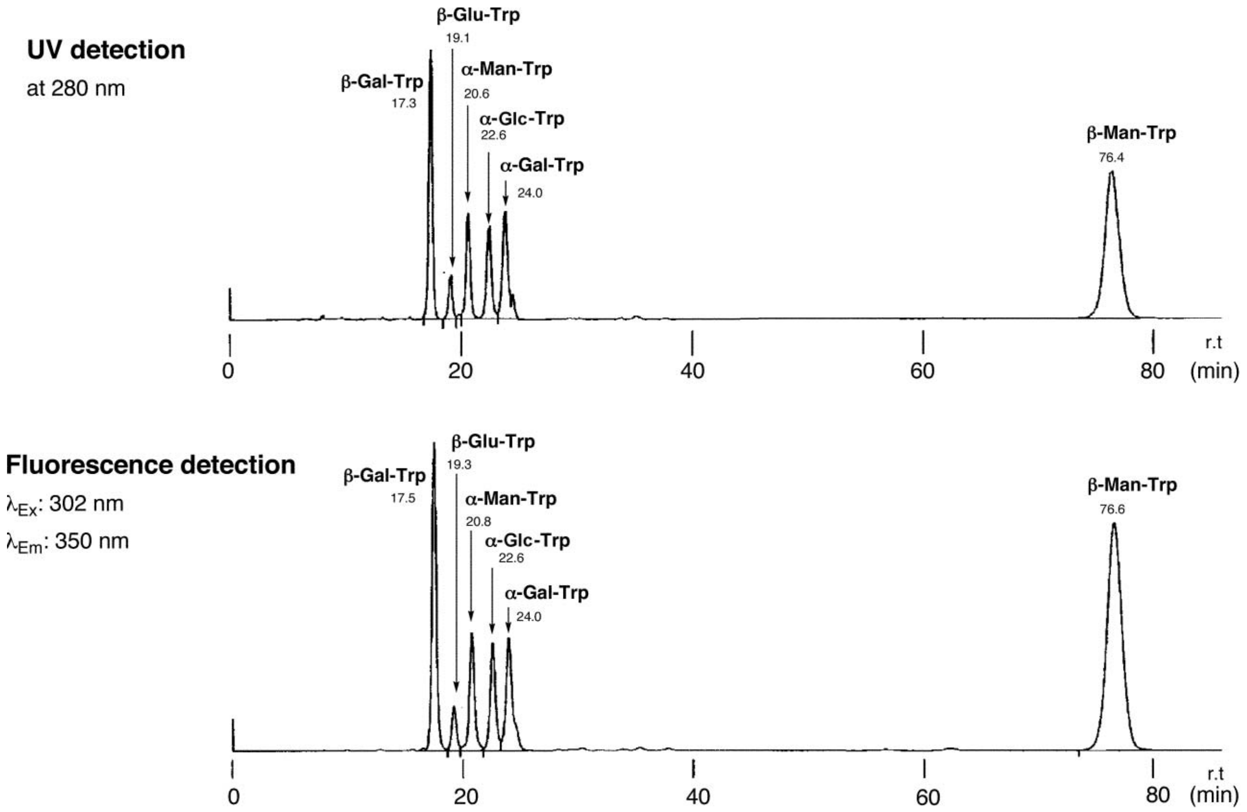


Fig. 2 HPLC chromatogram of synthetic α-C-Man-Trp and its analogues. Conditions: column ODS-5 Develosil, size; 4.6 × 250 mm, mobile phase: 10% MeOH–H_2O, 0.1% TFA, flow rate: 0.5 ml min^{-1}.

Cica-reagent silica gel 60 (particle size 0.063–0.2 mm ASTM) and Silica Gel 60 N (spherical, neutral) were used for open-column chromatography. Preparative thin-layer chromatographic separations were carried out on 0.5 mm silica gel plates 60 F_{254} (Merck. #1.05774). Unless otherwise noted, non-aqueous reactions were carried out in oven-dried (120 °C) or flame-dried glassware under a nitrogen atmosphere. Dry THF was purchased from Wako Pure Chemical Industries, Ltd. Dry CH_2Cl_2, CH_3CN and Et_3N were distilled from CaH_2 under a nitrogen atmosphere. $Sc(ClO_4)_3$ was prepared according to the literature. All other commercially available reagents were used as received.

1-Trimethylsilylethynyl-2,3,4,6-tetra-*O*-benzyl-D-mannopyranose (16). (1) A solution of mannolactol **14** (6.29 g, 11.6 mmol) in DMSO (36 ml) and Ac_2O (24 ml) was stirred at rt for 7 h, and the reaction mixture was quenched with H_2O at 0 °C. The resulting mixture was extracted with AcOEt (×2). The combined organic extracts were washed with water (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was dissolved in Et_2O, and passed through a short column packed with neutral silica gel, and the eluate was concentrated to afford lactone **15**. This material **15** (5.82 g) was used for the next step without further purification. (2) Trimethylsilylethyne (3.05 ml, 21.6 mmol) was dissolved in dry THF (60 ml) and the solution was cooled to −78 °C, and stirred at −78 °C for 20 min. To this solution was added *n*-BuLi (10.3 ml, 16.2 mmol, 1.57 M in hexane). After stirring at −78 °C for 40 min, the solution was warmed to 0 °C and stirred at 0 °C for 40 min. This solution was again cooled to −78 °C and stirred at −78 °C for 20 min. To this solution was added a THF (56 ml) solution of the lactone **15** (5.82 g, 10.8 mmol, dried azeotropically with toluene). After stirring for 3.5 h, the reaction was quenched with saturated NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue (6.87 g) was purified by silica gel column chromatography (150 g, AcOEt–hexane = 1 : 6) to afford ketal **16** (5.52 g, 75% in 2 steps).

(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)ethyne (13β). *Reduction with $(TMS)_3SiH$ (entry 2 in Table 1).* (1) The ketal **16** (3.25 g, 5.11 mmol) was dried azeotropically from toluene, and dissolved in dry CH_3CN (83 ml) and CH_2Cl_2 (15 ml). To this solution cooled at 0 °C were added $(TMS)_3SiH$ (7.90 ml, 25.6 mmol) and $BF_3{\cdot}Et_2O$ (1.94 μl, 15.3 mmol). After 1.5 h, the reaction was quenched with saturated $NaHCO_3$ solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue (8.39 g) was purified by silica gel column chromatography (150 g, AcOEt–hexane = 1 : 15 → 1 : 10 → 1 : 7) to afford β-*C*-trimethylsilylmannosylacetylene (2.58 g, 81%). (2) The β-*C*-trimethylsilylmannosylacetylene (2.58 g, 4.16 mmol) was dissolved in THF (74 ml)–H_2O (4 ml), and TBAF (4.16 ml, 4.16 mmol, 1 M in THF) was added. After stirring at rt for 1.5 h, the reaction was quenched with aqueous NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was purified by silica gel column chromatography (120 g, AcOEt–hexane = 1 : 6) to afford β-*C*-mannosylacetylene **13β** (2.13 mg, 93%) as a white solid: $[\alpha]_D^{30}$ −31.4 (*c* 1.05, $CHCl_3$); IR (KBr) ν_{max} 3284, 3032, 2866, 2127, 1954, 1877, 1811, 1606 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 2.47 (1H, d, J = 2 Hz, C≡C*H*), 3.47 (1H, ddd, J = 9.5, 5, 2 Hz, H-5), 3.55 (1H, dd, J = 9.5, 3 Hz, H-3), 3.68–3.80 (2H, m, H-6), 3.92 (1H, t, J = 9.5 Hz, H-4), 3.97 (1H, dd, J = 3, 1 Hz, H-2), 4.14 (1H, dd, J = 2, 1 Hz, H-1), 4.53 (1H, d, J = 11 Hz, CH_AH_BPh), 4.56 (1H, d, J = 13 Hz, CH_CH_DPh), 4.60 (1H, d, J = 11.5 Hz, CH_EH_FPh), 4.63 (1H, d, J = 13 Hz, CH_CH_DPh), 4.65 (1H, d, J = 11.5 Hz, CH_EH_FPh), 4.86 (1H, d, J = 11 Hz, CH_AH_BPh), 4.98 (2H, s, CH_2Ph), 7.12–7.50 (20H, m, aromatic); ^{13}C NMR (75 MHz, $CDCl_3$) δ 69.0, 69.3, 72.0, 73.4, 74.3, 74.5, 74.6, 75.2, 75.8, 79.9, 80.1, 83.4, 127.5, 127.6, 127.7, 127.7, 128.0, 128.1, 128.1, 128.2, 128.3, 128.4, 128.5, 138.1, 138.2, 138.3, 138.5; Anal. Calcd for $C_{36}H_{36}O_5$: C, 78.81; H, 6.61. Found: C, 78.81; H, 6.70.

Reduction with $(i\text{-}Pr)_3SiH$ (entry 3 in Table 1). The ketal **16** (55 mg, 0.086 mmol) was dried azeotropically with toluene and dissolved in dry CH_3CN–CH_2Cl_2 (1.40 ml–0.25 ml). To this solution was added $(i\text{-}Pr)_3SiH$ (88 μl, 0.43 mmol). After the solution was stirring at 0 °C for 25 min, $BF_3{\cdot}OEt_2$ (33 μl, 0.26 mmol) was added. After 15 min, the reaction was quenched with saturated $NaHCO_3$ solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue (52 mg) was dissolved in MeOH (1.0 ml), and K_2CO_3 (52 mg, 0.38 mmol) was added. After stirring at room temperature for 20 min, the reaction was quenched with saturated NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was purified by column chromatography (silica gel 1.6 g, AcOEt–hexane = 1 : 7 → 1 : 5) to afford β-*C*-mannosylacetylene **13β** (32 mg, 67% in 2 steps).

(2,3,4,6-Tetra-*O*-acetyl-β-D-mannopyranosyl)ethyne (17). To an ice-cold solution of β-*C*-mannosylacetylene **13β** (54 mg, 0.098 mmol) in Ac_2O (1.6 ml) was added TMSOTf (0.13 ml, 0.63 mmol). After stirring at rt for 38 h 45 min, the reaction was quenched with saturated $NaHCO_3$ solution and extracted with AcOEt (×3). The combined organic extracts were washed with saturated $NaHCO_3$ solution (×2), H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue (98 mg) was purified by silica gel column chromatography (10 g, Et_2O–hexane = 1 : 2 → 1 : 1 → 2 : 1) to afford β-*C*-tetraacetylmannosylacetylene **17** (22 mg, 62%) as a yellow oil. The NMR spectra were identical to those of the literature, diagnostic NOESY correlations were observed between H-1 (δ 4.46, dd, J = 2, 1 Hz) and H-3 (δ 5.06, dd, J = 10, 3.5 Hz) as well as H-1 and H-5 (δ 3.67, 1H, ddd, J = 10, 5.5, 2 Hz).

1-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-2-*o*-(*p*-toluensulfoamidyl)phenylethyne (18). A two-necked round-bottomed flask was charged with β-*C*-glucosylacetylene **11β** (278 mg, 0.507 mmol), *N*-Ts-*o*-iodoanilide **4** (376 mg, 1.01 mmol) and PPh_3 (13.3 mg, 0.0507 mmol), and connected to a vacuum/argon line. The flask was evacuated and then filled with argon. This evacuation–filling cycle was repeated three times. Et_3N (8.3 ml) was added and then the mixture was heated to 60 °C. After these reagents were completely dissolved, $Pd(OAc)_2$ (5.6 mg, 0.025 mmol) was added. After stirring at 60 °C for 2 h 45 min, the mixture was cooled to room temperature. The reaction was then quenched with saturated NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with saturated NH_4Cl solution (×2), H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was purified by silica gel column chromatography (30 g, CH_2Cl_2 → Et_2O–hexane = 1 : 1) to afford glucosyl-β-1-ethynylaniline **18** (346 mg, 86%) as yellow oil: $[\alpha]_D^{27}$ +20 (*c* 0.98, $CHCl_3$); IR (KBr) ν_{max} 3031, 2868, 2343, 1495, 1454, 1400, 1342, 1167, 1091, 1028 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 2.26 (3H, s, CH_3 of Ts), 3.50–3.83 (6H, m, H-2, H-3, H-4, H-5, H-6), 4.24 (1H, d, J = 9 Hz, H-1), 4.57 (1H, d, J = 12 Hz, CH_AH_BPh), 4.59 (1H, d, J = 11 Hz, CH_CH_DPh), 4.65 (1H, d, J = 12 Hz, CH_AH_BPh), 4.80 (1H, d, J = 11 Hz, CH_EH_FPh), 4.83 (1H, d, J = 14 Hz, CH_GH_HPh), 4.85 (1H, d, J = 11 Hz, CH_CH_DPh), 4.90 (1H, d, J = 14 Hz, CH_GH_HPh), 4.92 (1H, d, J = 11 Hz, CH_EH_FPh), 6.98 (1H, t, J = 7 Hz, aromatic), 7.11 (2H, d, J = 8 Hz, H-3″ of Ts), 7.15–7.38 (23H, m, aromatic, N*H*), 7.59 (1H, d, J = 8 Hz, aromatic), 7.68 (2H, d, J = 8 Hz, H-2″ of Ts); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 21.4, 68.9, 70.2, 73.6, 75.1, 75.5, 75.7, 77.6, 79.2, 80.7, 82.2, 86.1, 93.5, 113.2, 119.8, 124.2, 127.5, 127.7, 127.7, 127.8, 127.9, 127.9, 127.9, 128.0, 128.4, 129.6, 129.9, 132.1, 136.1, 137.7, 137.9, 138.0, 138.2, 138.4, 143.9; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.05; H, 6.14; N, 1.62.

1-(2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl)-2-*o*-(*p*-toluensulfoamidyl)phenylethyne (19). Following the procedure for **18**, **19** (2.50 g, 93%) was obtained as a yellow oil from β-*C*-galactosylacetylene **12β** (1.85 g, 3.38 mmol) and *N*-Ts-*o*-iodoanilide **4** (2.51 g, 6.76 mmol) after column chromatography (silica gel 120 g, CH_2Cl_2 → AcOEt–hexane = 1 : 3): $[\alpha]_D^{27}$ −9.0 (*c* 1.5, $CHCl_3$); IR (KBr) ν_{max} 3032, 2868, 1496, 1456, 1341, 1167, 1093, cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 2.22 (3H, s, CH_3 of Ts), 3.57 (1H, dd, J = 9.5, 3 Hz, H-3), 3.61–3.71 (3H, m, H-5, H-6), 4.00 (1H, t, J = 9.5 Hz, H-2), 4.02 (1H, dd, J = 3, 0.5 Hz, H-4), 4.19 (1H, d, J = 9.5 Hz, H-1), 4.46 (1H, d, J = 12 Hz, CH_AH_BPh), 4.54 (1H, d, J = 12 Hz, CH_AH_BPh), 4.65 (1H, d, J = 11.5 Hz, CH_CH_DPh), 4.73 (1H, d, J = 12 Hz, CH_EH_FPh), 4.78 (1H, d, J = 12 Hz, CH_EH_FPh), 4.79 (1H, d, J = 11 Hz, CH_GH_HPh), 4.86 (1H, d, J = 11 Hz, CH_GH_HPh), 4.98 (1H, d, J = 11.5 Hz, CH_CH_DPh), 6.98 (1H, dt, J = 7.5 1 Hz, aromatic), 7.06 (2H, br d, J = 8 Hz, aromatic), 7.17–7.41 (23H, m, aromatic, N*H*), 7.59 (1H, br d, J = 8 Hz, aromatic), 7.64 (2H, br d, J = 8 Hz, aromatic); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 21.4, 68.7, 70.6, 72.7, 73.7, 74.0, 75.0, 75.7, 77.5, 78.8, 80.1, 83.5, 93.7, 120.3, 124.3, 127.5, 127.6, 127.8, 127.9, 128.1, 128.1, 128.3, 128.4, 128.5, 129.6, 129.8, 132.1, 138.1, 138.1; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.12; H, 5.79; N, 1.66.

1-(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)-2-*o*-(*p*-toluensulfoamidyl)phenylethyne (20). Following the procedure for **18**, **20** (2.05 g, 67%) was obtained as a yellow oil along with mannosyl-β-1-tosylindole **23** (579 mg, 19%) from β-*C*-mannosylacetylene **13β** (2.13 g, 3.89 mmol) and *N*–Ts-*o*-iodoanilide **4** (2.89 g, 7.77 mmol) after column chromatography (silica gel 120 g, CH_2Cl_2 → AcOEt–hexane = 1 : 4 → 1 : 2): $[\alpha]_D^{30}$ −19.1 (*c* 0.24, $CHCl_3$); IR (KBr) ν_{max} 3063, 3032, 2865, 1954, 1811, 1653, 1598, 1496, 1093 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 2.26 (3H, s, CH_3 of Ts), 3.55 (1H, ddd, J = 9, 5 Hz, H-5), 3.64 (1H, dd, J = 9, 3 Hz, H-3), 3.73–3.85 (2H, m, H-6 × 2), 3.99 (1H, t, $J = 9$ Hz, H-4), 4.01 (1H, dd, $J = 3, 1$ Hz, H-2), 4.36 (1H, d, $J = 1$ Hz, H-1), 4.57 (1H, d, $J = 10.5$ Hz, CH_AH_BPh), 4.58 (1H, d, $J = 12$ Hz, CH_CH_DPh), 4.65 (1H, d, $J = 12$ Hz, CH_CH_DPh), 4.68 (1H, d, $J = 12$ Hz, CH_EH_FPh), 4.72 (1H, d, $J = 12$ Hz, CH_EH_FPh), 4.90 (1H, d, $J = 10.5$ Hz, CH_AH_BPh), 4.91 (1H, d, $J = 12$ Hz, CH_GH_HPh), 5.01 (1H, d, $J = 12$ Hz, CH_GH_HPh), 6.98 (1H, td, $J = 3, 1$ Hz, aromatic), 7.10–7.42 (26H, m, aromatic), 7.54 (1H, dd, $J = 8, 1$ Hz, aromatic), 7.68 (1H, dt, $J = 8.5, 2$ Hz, aromatic);^{13}C NMR ($CDCl_3$, 100 MHz) δ 21.4, 69.5, 69.8, 72.4, 73.5, 74.6, 74.6, 75.3, 76.0, 79.9, 80.7, 83.6, 92.8, 113.2, 119.6, 124.1, 127.5, 127.5, 127.5, 127.6, 127.7, 127.8, 128.0, 128.1, 128.1, 128.3, 128.4, 128.5, 129.6, 129.8, 132.1, 136.1, 138.0, 138.2, 138.3, 143.8; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.11; H, 6.05; N, 1.53.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-1-(*p*-toluenesulfonyl)indole (21). Glucosyl-β-1-ethynylaniline **18** (493 mg, 0.623 mmol) was dissolved in Et_3N (9.9 ml) and DMF (4.9 ml), and CuI (23 mg, 0.13 mmol) was added. This solution was stirred at 80 °C for 2.5 h. The reaction was quenched with saturated NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with saturated NH_4Cl solution (×2), H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was purified by silica gel column chromatography (15 g, AcOEt–hexane = 1 : 5) to afford glucosyl-β-1-tosylindole **21** (447 mg, 91%) as a yellow oil: $[\alpha]_D^{27}$ −46.5 (*c* 1.03, $CHCl_3$); IR (KBr) ν_{max} 3031, 2865, 1598, 1497, 1453, 1368, 1176, 1092 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 2.20 (3H, s, CH_3 of Ts), 3.68–4.04 (6H, m, H-2, H-3, H-4, H-5, H-6), 4.47 (1H, d, $J = 12$ Hz, CH_AH_BPh), 4.52 (1H, d, $J = 11$ Hz, CH_CH_DPh), 4.58 (1H, d, $J = 12$ Hz, CH_AH_BPh), 4.64 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.77 (1H, d, $J = 11$ Hz, CH_CH_DPh), 4.88 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.94 (1H, d, $J = 11.5$ Hz, CH_GH_HPh), 4.98 (1H, d, $J = 11.5$ Hz, CH_GH_HPh), 5.39 (1H, br d, $J = 9$ Hz, H-1), 6.68 (1H, s, H-3′), 6.92 (2H, br d, $J = 8$ Hz, aromatic), 6.97 (2H, br dd, $J = 8, 1$ Hz, aromatic), 7.07–7.36 (20H, m, aromatic) 7.39 (1H, br d, $J = 7.5$ Hz, aromatic), 7.78 (2H, d, $J = 8$ Hz aromatic), 8.08 (1H, d, $J = 9$ Hz, aromatic); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 21.4, 69.1, 73.3, 74.3, 74.9, 75.7, 78.3, 79.0, 81.2, 87.6, 115.2, 121.3, 123.6, 125.0, 127.0, 127.5, 127.6, 127.7, 127.8, 127.9, 128.1, 128.3, 128.5, 128.5, 129.1, 129.5, 135.6, 137.0, 137.9, 138.1, 138.3, 138.6, 144.4; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.07; H, 6.00; N, 1.64.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl)-1-(*p*-toluenesulfonyl)indole (22). Following the procedure for **21**, **22** (2.24 g, 90%) was obtained as a yellow oil from galactosyl-β-1-ethynylaniline **19** (2.50 g, 3.16 mmol) after column chromatography (120 g, AcOEt–hexane = 1 : 5): $[\alpha]_D^{27}$ −49.7 (*c* 0.35, $CHCl_3$); IR (KBr) ν_{max} 3031, 2869, 1598, 1497, 1497, 1454, 1367, 1092 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 2.18 (3H, s, CH_3 of Ts), 3.54–4.14 (5H, m, H-3, H-4, H-5, H-6), 4.32 (1H, t, $J = 9.5$ Hz, H-2), 4.42 (1H, d, $J = 12$ Hz, CH_AH_BPh), 4.46 (1H, d, $J = 12$ Hz, CH_AH_BPh), 4.58 (1H, d, $J = 11$ Hz, CH_CH_DPh), 4.68 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.74 (1H, d, $J = 11.5$ Hz, CH_GH_HPh), 4.82 (1H, d, $J = 11.5$ Hz, CH_GH_HPh), 4.89 (1H, d, $J = 11$ Hz, CH_CH_DPh), 5.00 (1H, d, $J = 11$ Hz, CH_EH_FPh), 5.41 (1H, d, $J = 9.5$ Hz, H-1), 6.71 (1H, s, H-3′), 6.88 (2H, br d, $J = 8.5$ Hz, aromatic), 7.00 (2H, dd, $J = 8, 1$ Hz, aromatic), 7.07–7.42 (21H, m, aromatic), 7.73 (2H, d, $J = 8.5$ Hz, aromatic), 8.06 (1H, d, $J = 8.5$ Hz, aromatic); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 21.4, 68.6, 72.5, 73.2, 73.4, 73.9, 74.7, 77.1, 77.2, 77.7, 85.5, 111.0, 115.1, 121.3, 123.5, 124.8, 126.9, 127.4, 127.5, 127.7, 127.9, 128.1, 128.2, 128.4, 128.5, 129.2, 129.3, 135.6, 136.9, 137.9, 138.2, 138.3, 138.9, 139.0, 144.2; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.13; H, 6.06; N, 1.78.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)-1-(*p*-toluenesulfonyl)indole (23). Following the procedure for **21**, **23** (17.2 mg, 87%) was obtained as a yellow oil from mannosyl-β-1-ethynylaniline **20** (19.8 mg, 0.025 mmol) after column chromatography (5 g, AcOEt–hexane = 1 : 4): $[\alpha]_D^{30}$ −94.6 (*c* 0.76, $CHCl_3$); IR (KBr) ν_{max} 3309, 3064, 3031, 2864, 1952, 1597, 1497, 1454 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 2.28 (3H, s, CH_3 of Ts), 3.68–3.74 (1H, m, H-5), 3.79–3.86 (2H, m, H-6), 3.88 (1H, dd, $J = 9.5, 2.5$ Hz, H-3), 4.01 (1H, t, $J = 9.5$ Hz, H-4), 4.25 (1H, d, $J = 11.5$ Hz, CH_AH_BPh), 4.50 (1H, d, $J = 11.5$ Hz, CH_AH_BPh), 4.53 (1H, br d, $J = 2.5$ Hz, H-2), 4.60 (1H, d, $J = 12$ Hz, CH_CH_DPh), 4.61 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.65 (1H, d, $J = 12$ Hz, CH_GH_HPh), 4.70 (1H, d, $J = 12$ Hz, CH_CH_DPh), 4.75 (1H, d, $J = 12$ Hz, CH_GH_HPh), 4.95 (1H, d, $J = 11$ Hz, CH_EH_FPh), 5.13 (1H, s, H-1), 6.89–7.45 (26H, m, aromatic, N*H*), 7.55 (2H, br d, $J = 8$ Hz, aromatic), 8.08 (1H, d, $J = 8$ Hz, aromatic); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 21.5, 69.9, 71.8, 73.4, 74.3, 74.9, 75.1, 75.6, 77.2, 80.3, 84.3, 112.4, 115.1, 120.9, 122.4, 123.9, 124.4, 126.3, 126.8, 127.3, 127.4, 127.5, 127.5, 127.6, 127.8, 127.9, 128.0, 128.0, 128.1, 128.3, 128.3, 128.4, 129.5, 129.6, 129.8, 130.1, 135.1, 137.1, 137.9, 138.1, 138.2, 138.5, 138.5, 139.1, 144.9; Anal. Calcd for $C_{49}H_{47}NO_7S$: C, 74.12; H, 5.97; N, 1.76. Found: C, 74.13; H, 6.06; N, 1.83.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-1*H*-indole (24). To a solution of glucosyl-β-1-tosylindole **21** (377 mg, 0.476 mmol) in THF (11 ml) was added TBAF (2.3 ml, 2.4 mmol, 1 M in THF). This solution was heated at reflux temperature with stirring for 2 h. The reaction was quenched with saturated NH_4Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with H_2O (×2) and brine (×2), dried over anhydrous Na_2SO_4, and concentrated. The residue was purified by silica gel column chromatography (5 g, CH_2Cl_2) to afford glucosyl-β-1-indole **24** (287 mg, 94%) as a yellow oil: $[\alpha]_D^{27}$ −14.8 (*c* 1.01, $CHCl_3$); IR (KBr) ν_{max} 3406, 3032, 2902, 2865, 1455, 1359, 1135, 1062 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 3.55–3.85 (6H, m, H-2, H-3, H-4, H-5, H-6), 4.02 (1H, s, $J = 10.5$ Hz, H-1) 4.50 (1H, d, $J = 10$ Hz, CH_AH_BPh), 4.56 (1H, d, $J = 11.5$ Hz, CH_CH_DPh), 4.59 (1H, d, $J = 10$ Hz, CH_AH_BPh), 4.62 (1H, d, $J = 11.5$ Hz, CH_CH_DPh), 4.63 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.88 (1H, d, $J = 11$ Hz, CH_EH_FPh), 4.91 (1H, d, $J = 10.5$ Hz, CH_GH_HPh), 4.98 (1H, d, $J = 10.5$ Hz, CH_GH_HPh), 6.58 (1H, s, indole), 7.00–7.40 (23H, m, aromatic), 7.60 (1H, s, aromatic), 8.48 (1H, s, N*H*); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 68.9, 73.5, 75.0, 75.1, 75.7, 75.7, 77.9, 79.1, 82.5, 86.5, 101.6, 111.1, 119.8, 120.6, 121.9, 127.7, 127.8, 127.9, 127.9, 128.0, 128.2, 128.4, 128.4, 128.5, 135.7, 136.1, 137.5, 138.0, 138.1, 138.6; Anal. Calcd for $C_{42}H_{41}NO_5$: C, 78.85; H, 6.46; N, 2.19. Found: C, 78.86; H, 6.61; N, 2.19.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl)-1*H*-indole (25). Following the procedure for **24**, **25** (1.67 g, 92%) was obtained as a yellow solid from galactosyl-β-1-tosylindole **22** (2.24 g, 2.83 mmol) after column chromatography (silica gel 90 g, AcOEt–hexane = 1 : 4): $[\alpha]_D^{22}$ −20.8 (*c* 1.05, $CHCl_3$); IR (KBr) ν_{max} 3423, 3032, 2869, 1497, 1455, 1364, 1294, 1101 cm^{-1}; ^{1}H NMR (CDCl$_3$, 300 MHz) δ 3.64 (2H, dd, J = 6.5, 1.5 Hz, H-6), 3.68–3.78 (2H, m, H-3, H-5), 3.98 (1H, t, J = 9.5 Hz, H-2), 4.06–4.10 (1H, m, H-4), 4.09 (1H, d, J = 10 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BPh}$), 4.45 (1H, d, J = 11.5 Hz, $\mathrm{C}H_\mathrm{C}\mathrm{H_DPh}$), 4.47 (1H, d, J = 9.5 Hz, H-1), 4.50 (1H, d, J = 11.5 Hz, $\mathrm{CH_C}H_\mathrm{D}\mathrm{Ph}$), 4.56 (1H, d, J = 10 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{Ph}$), 4.64 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{E}\mathrm{H_FPh}$), 4.79 (2H, s, $\mathrm{C}H_2\mathrm{Ph}$), 5.00 (1H, d, J = 11 Hz, $\mathrm{CH_E}H_\mathrm{F}\mathrm{Ph}$), 6.55 (1H, br d, J = 1.5 Hz, H-3′), 7.02–7.42 (23H, m, aromatic), 7.59 (1H, br d, J = 8 Hz, aromatic), 8.59 (1H, s, NH); ^{13}C NMR (CDCl$_3$, 75 MHz) δ 68.7, 72.7, 73.5, 74.3, 74.9, 75.2, 76.1, 77.4, 78.9, 84.0, 101.6, 111.1, 119.6, 120.6, 121.8, 127.6, 127.7, 127.8, 127.9, 128.0, 128.1, 128.3, 128.3, 128.4, 128.5, 135.9, 136.0, 137.9, 138.4, 138.7; Anal. Calcd for $C_{42}H_{41}NO_5$: C, 78.85; H, 6.46; N, 2.19. Found: C, 78.86; H, 6.35; N, 2.23.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)-1*H*-indole (26) from 23. Following the procedure for **24**, **26** (10 mg, quant.) was obtained as a yellow solid from mannosyl-β-1-tosylindole **23** (12 mg, 0.015 mmol) after column chromatography (silica gel 5 g, AcOEt–hexane = 1 : 4): $[\alpha]_\mathrm{D}^{30}$ −7.8 (c 1.30, CHCl$_3$); IR (KBr) ν_max 3440, 3062, 2863, 1952, 1586, 1455, 1101 cm^{-1}; ^{1}H NMR (CDCl$_3$, 300 MHz) δ 3.61 (1H, m, H-5), 3.76–3.81 (2H, m, H-6), 3.80 (1H, dd, J = 9.5, 2.5 Hz, H-3), 4.02 (1H, dd, J = 2.5, 1 Hz, H-2), 4.10 (1H, t, J = 9.5 Hz, H-4), 4.23 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BPh}$), 4.54 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{C}\mathrm{H_DPh}$), 4.63 (1H, d, J = 10.5 Hz, $\mathrm{C}H_\mathrm{E}\mathrm{H_FPh}$), 4.64 (1H, d, J = 12 Hz, $\mathrm{CH_C}H_\mathrm{D}\mathrm{Ph}$), 4.68 (1H, d, J = 1 Hz, H-1), 4.73 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{G}\mathrm{H_HPh}$), 4.77 (1H, d, J = 12 Hz, $\mathrm{CH_G}H_\mathrm{H}\mathrm{Ph}$), 4.78 (1H, d, J = 11 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{Ph}$), 4.94 (1H, d, J = 10.5 Hz, $\mathrm{CH_E}H_\mathrm{F}\mathrm{Ph}$), 6.34 (1H, d, J = 1.5 Hz, indole), 7.02–7.40 (23H, m, aromatic), 7.57 (1H, br d, J = 8 Hz, aromatic), 8.84–8.90 (1H, br, NH); ^{13}C NMR (CDCl$_3$, 100 MHz) δ 69.4, 72.4, 73.5, 74.6, 74.8, 75.0, 75.3, 78.5, 79.7, 84.4, 99.5, 111.0, 119.5, 120.4, 121.7, 127.4, 127.5, 127.6, 127.7, 127.7, 127.8, 128.1, 128.2, 128.3, 128.3, 128.5, 129.6, 135.6, 135.8, 138.0, 138.2, 138.3, 138.3; Anal. Calcd for $C_{42}H_{41}NO_5$: C, 78.85; H, 6.46; N, 2.19. Found: C, 78.83; H, 6.61; N, 2.15.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)-1*H*-indole (26) from 23. A mixture of β-1-ethynylmannose **20** (2.05 g, 2.59 mmol) and mannosyl-β-1-tosylindole **23** (579 mg, 0.73 mmol) was dissolved in THF (79 ml), and TBAF (10 ml, 10 mmol, 1 M in THF) was added. This solution was stirred at reflux temperature for 6 h. The reaction was quenched with saturated NH$_4$Cl solution and extracted with AcOEt (×3). The combined organic extracts were washed with H$_2$O (×2) and brine (×2), dried over anhydrous Na$_2$SO$_4$, and concentrated. The residue was purified by column chromatography (silica gel 110 g, AcOEt–hexane = 1 : 4) to afford mannosyl-β-1-indole **26** (1.57 g, 74%) as a yellow solid.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-L-(*N*-carbobenzyloxyl)-tryptophan methyl ester (27). Sc(ClO$_4$)$_3$ (221 mg, 0.645 mmol) placed in a reaction vessel was freeze-dried with benzene for 1 h, then cooled to 0 °C. To this flask was added MS 5A (300 mg) and the vessel was connected to a vacuum/argon line. The flask was evacuated and then filled with argon. This evacuation–filling cycle was repeated three times. In a separate flask, glucosylindole **24** (206 mg, 0.322 mmol) and aziridine **7** (152 mg, 0.645 mmol) were dried azeotropically with benzene, and dissolved in dry CH$_2$Cl$_2$ (6 ml). The solution was added to the reaction vessel *via* cannular tubing. The reaction mixture was stirred at the same temperature for 3 h, and directly subjected to silica gel column chromatography (15 g, AcOEt–hexane = 1 : 4 → 1 : 3) to give **27** (176 mg, 62%) as a brown oil: $[\alpha]_\mathrm{D}^{27}$ +8.4 (c 0.22, CHCl$_3$); IR (KBr) ν_max 3311, 3032, 2922, 1954, 1718, 1455, 1210, 1062 cm^{-1}; ^{1}H NMR (CDCl$_3$, 400 MHz) δ 3.19 (1H, dd, J = 15, 4 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BCHCOOMe}$), 3.53 (1H, dd, J = 15, 6.5 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{CHCOOMe}$), 3.53–3.58 (1H, m, H-5), 3.60 (3H, s, $\mathrm{COOC}H_3$), 3.62 (1H, t, J = 9 Hz, H-2), 3.69 (1H, dd, J = 10.5, 2 Hz, H-6), 3.76 (1H, dd, J = 10.5, 3 Hz, H-6), 3.75–3.85 (2H, m, H-3, H-4), 4.15 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BPh}$), 4.43 (1H, d, J = 11.5 Hz, $\mathrm{C}H_\mathrm{C}\mathrm{H_DPh}$), 4.45 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{E}\mathrm{H_FPh}$), 4.52 (1H, d, J = 11 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{Ph}$), 4.56 (1H, d, J = 9 Hz, H-1), 4.58 (1H, d, J = 11.5 Hz, $\mathrm{CH_C}H_\mathrm{D}\mathrm{Ph}$), 4.65–4.72 (1H, m, $\mathrm{C}H\mathrm{COOCH_3}$), 4.74 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{G}\mathrm{H_HPh}$), 4.76 (1H, d, J = 11 Hz, $\mathrm{CH_E}H_\mathrm{F}\mathrm{Ph}$), 4.80 (1H, d, J = 11 Hz, $\mathrm{CH_G}H_\mathrm{H}\mathrm{Ph}$), 5.08 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{I}\mathrm{H_JPh}$), 5.12 (1H, d, J = 12 Hz, $\mathrm{CH_I}H_\mathrm{J}\mathrm{Ph}$), 6.64 (1H, d, J = 8.5 Hz, aromatic), 6.90 (2H, d, J = 7 Hz, aromatic), 7.06 (2H, br t, J = 7.5 Hz, aromatic), 7.09–7.39 (24H, m, aromatic), 7.53 (1H, d, J = 8 Hz aromatic), 8.41 (1H, s, NH of indole); ^{13}C NMR (100 MHz, CDCl$_3$) δ 26.9, 52.2, 54.9, 66.9, 68.4, 73.5, 74.2, 74.5, 75.0, 75.5, 77.6, 79.0, 81.0, 86.9, 108.9, 111.2, 118.8, 119.5, 122.4, 127.6, 127.7, 127.8, 128.0, 128.1, 128.2, 128.3, 128.4, 128.4, 128.5, 132.5, 135.6, 137.0, 137.7, 138.2, 138.5, 156.3, 172.6; HRMS (FAB) Calcd for $C_{54}H_{55}N_2O_9$ (M + H): 875.3908, Found: 875.3887.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl)-L-(*N*-carbobenzyloxyl)-tryptophan methyl ester (28). Following the procedure for **27**, **28** (60 mg, 39%) was obtained as a yellow oil from galactosylindole **25** (112 mg, 0.175 mmol) and aziridine **7** (82 mg, 0.35 mmol) after column chromatography (12 g, AcOEt–hexane = 1 : 4): $[\alpha]_\mathrm{D}^{27}$ +10.1 (c 0.36, CHCl$_3$); IR (KBr) ν_max 3298, 3032, 2871, 1721, 1455, 1212, 1071 cm^{-1}; ^{1}H NMR (CDCl$_3$, 400 MHz) δ 3.19 (1H, dd, J = 15, 4 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BCHCOOMe}$), 3.53 (1H, dd, J = 15, 6.5 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{CHCOOMe}$), 3.53–3.58 (1H, m, H-5), 3.60 (3H, s, $\mathrm{COOC}H_3$), 3.62 (1H, t, J = 9 Hz, H-2), 3.69 (1H, dd, J = 10.5, 2 Hz, H-6), 3.76 (1H, dd, J = 10.5, 3 Hz, H-6), 3.75–3.85 (2H, m, H-3, H-4), 4.15 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_BPh}$), 4.43 (1H, d, J = 11.5 Hz, $\mathrm{C}H_\mathrm{C}\mathrm{H_DPh}$), 4.45 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{E}\mathrm{H_FPh}$), 4.52 (1H, d, J = 11 Hz, $\mathrm{CH_A}H_\mathrm{B}\mathrm{Ph}$), 4.56 (1H, d, J = 9 Hz, H-1), 4.58 (1H, d, J = 11.5 Hz, $\mathrm{CH_C}H_\mathrm{D}\mathrm{Ph}$), 4.65–4.72 (1H, m, $\mathrm{C}H\mathrm{COOCH_3}$), 4.74 (1H, d, J = 11 Hz, $\mathrm{C}H_\mathrm{G}\mathrm{H_HPh}$), 4.76 (1H, d, J = 11 Hz, $\mathrm{CH_E}H_\mathrm{F}\mathrm{Ph}$), 4.80 (1H, d, J = 11 Hz, $\mathrm{CH_G}H_\mathrm{H}\mathrm{Ph}$), 5.08 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{I}\mathrm{H_JPh}$), 5.12 (1H, d, J = 12 Hz, $\mathrm{CH_I}H_\mathrm{J}\mathrm{Ph}$), 6.64 (1H, d, J = 8.5 Hz, aromatic), 6.90 (2H, d, J = 7 Hz, aromatic), 7.06 (2H, br t, J = 7.5 Hz, aromatic), 7.09–7.39 (24H, m, aromatic), 7.53 (1H, d, J = 8 Hz aromatic), 8.41 (1H, s, NH of indole); ^{13}C NMR (100 MHz, CDCl$_3$) δ 26.9, 52.1, 54.7, 66.7, 68.5, 73.5, 74.3, 74.4, 74.9, 77.2, 77.5, 84.7, 108.8, 111.2, 118.9, 119.5, 122.3, 127.5, 127.7, 127.7, 127.8, 127.9, 128.1, 128.2, 128.2, 128.3, 128.4, 128.4, 128.7, 128.8, 132.9, 135.5, 136.6, 137.1, 137.8, 138.6, 172.5; HRMS (FAB) Calcd for $C_{54}H_{55}N_2O_9$ (M + H): 875.3908, Found: 875.3887.

2-(2,3,4,6-Tetra-*O*-benzyl-β-D-mannopyranosyl)-L-(*N*-carbobenzyloxyl)-tryptophan methyl ester (29). Following the procedure for **27**, **29** (140 mg, 49%) was obtained as a yellow oil from mannosylindole **26** (210 mg, 0.329 mmol) and aziridine **7** (154 mg, 0.657 mmol) after column chromatography (15 g, AcOEt–hexane = 1 : 4 → 1 : 3): $[\alpha]_\mathrm{D}^{27}$ +5.7 (c 0.35, CHCl$_3$); IR (KBr)

$\nu_{\max}$ 3429, 3032, 2868, 1721, 1497, 1455, 1215, 1098 cm^{-1}; ^{1}H NMR (CDCl$_3$, 400 MHz) δ 3.18 (1H, dd, J = 15, 6.5 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_B}$CHCOOMe), 3.25 (1H, dd, J = 15, 5.5 Hz, $\mathrm{CH_A}H_\mathrm{B}$CHCOOMe), 3.61 (3H, s, COOCH_3), 3.59–3.66 (1H, m, H-5), 3.69–3.80 (2H, m, H-6), 3.82 (1H, m, H-3), 4.05 (1H, m, H-2), 4.13 (1H, t, J = 9.5 Hz, H-4), 4.23 (1H, d, J = 10.5 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_B}$Ph), 4.42 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{C}\mathrm{H_D}$Ph), 4.53–4.62 (1H, m, C$H$COOCH$_3$), 4.56 (1H, d, J = 12 Hz, $\mathrm{CH_C}H_\mathrm{D}$Ph), 4.61 (1H, d, J = 10.5 Hz, $\mathrm{C}H_\mathrm{E}\mathrm{H_F}$Ph), 4.68–4.78 (3H, m, H-1, CH_2Ph), 4.86 (1H, d, J = 10.5 Hz, $\mathrm{CH_A}H_\mathrm{B}$Ph), 4.93 (1H, d, J = 10.5 Hz, $\mathrm{CH_E}H_\mathrm{F}$Ph), 5.02 (1H, d, J = 12 Hz, $\mathrm{C}H_\mathrm{I}\mathrm{H_J}$Ph), 5.06 (1H, d, J = 12 Hz, $\mathrm{CH_I}H_\mathrm{J}$Ph), 5.44 (1H, d, J = 7 Hz, NH–Cbz), 7.03–7.39 (28H, m, aromatic), 7.49 (1H, d, J = 7.5 Hz aromatic), 8.96 (1H, s, NH of indole); ^{13}C NMR (100 MHz, CDCl$_3$) δ 27.1, 52.4, 54.9, 66.9, 69.3, 72.0, 72.5, 73.4, 74.7, 75.1, 75.2, 77.2, 77.7, 79.8, 84.7, 106.4, 111.1, 118.7, 119.4, 122.2, 127.4, 127.6, 127.6, 127.7, 127.9, 128.0, 128.0, 128.1, 128.2, 128.3, 128.4, 128.4, 128.5, 133.0, 135.3, 136.2, 138.0, 138.2, 138.4, 155.8, 172.4; HRMS (FAB) Calcd for C$_{54}$H$_{55}$N$_2$O$_9$ (M + H): 875.3908, Found: 875.3887.

2-β-D-Mannopyranosyl-L-tryptophan (33). (1) To a solution of **29** (130 mg, 0.149 mmol) in CH$_3$CN–MeOH (3 ml/3 ml) was added 1 N LiOH solution (0.30 ml, 0.30 mmol). After stirring at rt for 14 h, sat. NH$_4$Cl solution was added. The pH of the mixture was adjusted to 2 with 1 N HCl and then extracted with AcOEt (×3). The combined organic layer was washed with water and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The residue was purified by column chromatography (silica gel 10 g, MeOH–CH$_2$Cl$_2$ = 1 : 20) to give **30** (110 mg, 86%). (2) A two-necked flask was charged with 5% Pd–C (31 mg) and connected to an inlet adaptor. The flask was evacuated and then filled with nitrogen. A solution of **30** (31 mg, 0.037 mmol) in MeOH (0.94 ml) and 1 N HCl (14 μl) were added. The flask was then evacuated and then filled with hydrogen. After vigorous stirring for 25 h, the mixture was filtered through a pad of Hyflo Super-Cel, and the precipitate was washed with MeOH and H$_2$O. The combined filtrate was concentrated. The residue (15.2 mg) was purified by preparative TLC (CHCl$_3$–MeOH–H$_2$O = 65 : 65 : 15) to give **33**, which was further purified by reversed phase column chromatography (Cosmosil 75C$_{18}$, H$_2$O as an eluate) to give **33** (5.8 mg, 43%) as a white solid: $[\alpha]_\mathrm{D}^{22}$ +14.1 (c 0.17, H$_2$O); ^{1}H NMR (D$_2$O, 600 MHz) δ 3.26 (1H, dd, J = 15, 9.5 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_B}$CHCOOH), 3.61 (1H, dd, J = 15, 4.5 Hz, $\mathrm{CH_A}H_\mathrm{B}$CHCOOH), 3.66 (1H, ddd, J = 9.5, 6.5, 2 Hz, H-5), 3.76 (1H, t, J = 9.5 Hz, H-4), 3.81 (1H, dd, J = 12, 6.5 Hz, H-6), 3.88 (1H, dd, J = 9.5, 3 Hz, H-3), 4.01 (1H, dd, J = 9.5, 4.5 Hz, CHCOOH), 4.04 (1H, dd, J = 12, 2 Hz, H-6), 4.29 (1H, d, J = 3 Hz, H-2), 5.08 (1H, s, H-1), 7.23 (1H, dd, J = 8, 7, Hz, indole), 7.31 (1H, dd, J = 8, 7, Hz, indole), 7.53 (1H, d, J = 8 Hz, indole), 7.75 (1H, d, J = 8 Hz, indole); ^{13}C NMR (120 MHz, D$_2$O) δ 28.6, 58.0, 63.9, 69.7, 74.3, 76.5, 76.6, 82.9, 109.3, 114.6, 121.2, 122.5, 125.3, 129.8, 136.1, 138.3, 177.3; HRMS (FAB) Calcd for C$_{17}$H$_{23}$N$_2$O$_7$ (M + H): 367.1505, Found: 367.1500.

2-β-D-Galactopyranosyl-L-tryptophan (34). (1) To a solution of **28** (60 mg, 0.069 mmol) in CH$_3$CN–MeOH (2.7 ml/2.7 ml) was added 1 N LiOH solution (0.14 ml, 0.14 mmol). After stirring at rt for 6 h 40 min, sat. NH$_4$Cl solution was added. The mixture was adjusted to pH 2 with 1 N HCl and then extracted with AcOEt (×3). The combined organic layer was washed with water and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The residue was purified by preparative TLC (10% MeOH–CH$_2$Cl$_2$) to give **31** (38 mg, 64%). (2) A two-necked flask was charged with 5% Pd–C (11.6 mg) and connected to an inlet adaptor. The flask was evacuated and then filled with nitrogen. A solution of **31** (11.6 mg, 0.013 mmol) in MeOH (0.34 ml) and 1 N HCl (4 μl) were added. The flask was then evacuated and then filled with hydrogen. After vigorous stirring for 18.5 h, the mixture was filtered through a pad of Hyflo Super-Cel, and the precipitate was washed with MeOH and H$_2$O. The combined filtrate was concentrated. The residue (4 mg) was purified by preparative TLC (CHCl$_3$–MeOH–H$_2$O = 65 : 65 : 15) to give **35**, which was further purified by reversed phase column chromatography (Cosmosil 75C$_{18}$, H$_2$O as an eluate) to give **34** (3.3 mg, 69%) as a white solid: $[\alpha]_\mathrm{D}^{21}$ +3.0 (c 0.17, H$_2$O); ^{1}H NMR (D$_2$O, 600 MHz) δ 3.30 (1H, dd, J = 15, 9.5 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_B}$CHCOOH), 3.60 (1H, dd, J = 15, 4.5 Hz, $\mathrm{CH_A}H_\mathrm{B}$CHCOOH), 3.80 (1H, dd, J = 14, 12 Hz, H-6), 3.81 (1H, s, H-6), 3.85 (1H, dd, J = 9.5, 3 Hz, H-3), 3.94–3.97 (1H, m, H-5), 4.09 (1H, dd, J = 9.5, 4.5 Hz, CHCOOH), 4.11 (1H, t, J = 9.5 Hz, H-2), 4.11 (1H, br s, H-4), 4.68 (1H, d, J = 9.5 Hz, H-1), 7.23 (1H, ddd, J = 8, 7, 1 Hz, indole), 7.33 (1H, ddd, J = 8, 7, 1 Hz, indole), 7.55 (1H, d, J = 8 Hz, indole), 7.77 (1H, d, J = 8 Hz, indole); ^{13}C NMR (120 MHz, D$_2$O) δ 28.4, 58.0, 64.2, 72.0, 72.8, 76.6, 76.8, 81.8, 111.0, 114.7, 121.6, 122.5, 125.8, 129.6, 136.2, 138.8, 177.1; HRMS (FAB) Calcd for C$_{17}$H$_{23}$N$_2$O$_7$ (M + H): 367.1505, Found: 367.1532.

2-β-D-Glucopyranosyl-L-tryptophan (35). (1) To a solution of **27** (172 mg, 0.197 mmol) in CH$_3$CN–MeOH (7.9 ml/7.9 ml) was added 1 N LiOH solution (0.39 ml, 0.39 mmol). After stirring at rt for 15 h, sat. NH$_4$Cl solution was added. The pH of the mixture was adjusted to 2 with 1 N HCl and then extracted with AcOEt (×3). The combined organic layer was washed with water and brine, dried over anhydrous Na$_2$SO$_4$, and concentrated. The residue was purified by silica gel column chromatography (15 g, MeOH–CH$_2$Cl$_2$ = 1 : 20) to give **32** (109 mg, 64%). (2) A two-necked flask was charged with 10%Pd–C (6.4 mg) and connected to an inlet adaptor. The flask was evacuated and then filled with nitrogen. A solution of **32** (6.4 mg, 0.0074 mmol) in dioxane–H$_2$O (0.19 ml : 0.032 ml) was added. The flask was then evacuated and then filled with hydrogen. After vigorous stirring for 36 h, 1 N HCl (2 μl) was added and stirring was continued for 7 h. The mixture was filtered through a pad of Hyflo Super-Cel, and the precipitate was rinsed with MeOH–CH$_3$Cl–H$_2$O (65 : 65 : 15). The combined filtrate was concentrated. The residue was washed with MeOH and the precipitate was further washed with CHCl$_3$–MeOH–H$_2$O (65 : 65 : 15) to give **35** (3.1 mg, quant.) as a white solid: $[\alpha]_\mathrm{D}^{27}$ +2.6 (c 0.16, H$_2$O); ^{1}H NMR (D$_2$O, 600 MHz) δ 2.88 (1H, dd, J = 15, 9 Hz, $\mathrm{C}H_\mathrm{A}\mathrm{H_B}$CHCOOH), 3.15 (1H, dd, J = 15, 4.5 Hz, $\mathrm{CH_A}H_\mathrm{B}$CHCOOH), 3.34 (1H, t, J = 9.5 Hz, H-4), 3.41 (1H, t, J = 9.5 Hz, H-3), 3.41 (1H, ddd, J = 9.5, 5, 2 Hz, H-5), 3.53 (1H, dd, J = 12.5, 5 Hz H-6), 3.54 (1H, t, J = 9.5 Hz, H-2), 3.53–3.57 (1H, m, CHCOOH), 3.64 (1H, dd, J = 12.5, 2 Hz, H-6), 4.47 (1H, d, J = 9.5 Hz, H-1), 6.92 (1H, t, J = 7.5 Hz, indole), 7.02 (1H, t, J = 7.5 Hz, indole), 7.23 (1H, d, J = 8 Hz, indole), 7.48 (1H, d, J = 8 Hz, indole); ^{13}C NMR (120 MHz, CDCl$_3$) δ 29.8, 58.6, 63.2, 72.1, 75.4, 76.1, 79.6, 82.3, 112.2, 114.2, 121.5, 122.1, 125.4, 129.6, 135.2, 138.6; HRMS (FAB) Calcd for C$_{17}$H$_{23}$N$_2$O$_7$ (M + H): 367.1505, Found: 367.1524.

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