

Supplementary Material

Total Synthesis of the Chlorinated Indigo-*N*-Glycosides Akashin A, B and C

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General methods:

Unless otherwise noted, chemicals were obtained from commercial suppliers and used without further purification. Methanol was dried with Mg-turnings (5 g / L) and distilled under argon. Dichloromethane was dried over phosphorous pentoxide, distilled and kept over 3 Å molecular sieves. Dimethylformamide and pyridine were p.a. quality, acetonitrile was HPLC grade (>99.9%).

TLC was performed on precoated silica gel plates (Macherey-Nagel, Alugram Sil G/UV254, silica layer thickness: 0.25 mm). Fluorescent compounds were detected using a UV illuminator (254 nm). Glycosylated compounds were detected using a 1:1 solution of 0.2% of resorcinol monomethyl ether (3-methoxyphenol) and 2 M sulfuric acid in ethanol followed by heating.

Flash chromatography was carried out on Merck silica gel (0.040-0.063 mm).

For solid phase extraction Waters Sep-Pak Vac C18 cartridges were used.

Optical rotations were determined in 1 dm cells (1 mL) in a Perkin-Elmer 241 polarimeter at 589 nm.

UV measurements were performed with a Specord 200 Jena Analytik spectrophotometer.

ESI-mass spectra were recorded on a Micromass LCT ESI-TOF mass spectrometer coupled to an Agilent HP 1100 HPLC with a photodiode array UV detector. The following reversed phase columns were used for LC-MS: YMC-Pack Pro C8, S-3 µm (50 x 2.1 mm), YMC-Pack Pro C18, S-3 µm (50 x 2.1 mm). Eluent A, water (0.1% formic acid); eluent B, acetonitrile (0.1% formic acid).

El-mass spectra were recorded on a Finnigan MS 8500 mass spectrometer. ¹H and ¹³C NMR spectra were recorded on Jeol JNM-EX-270, Bruker Avance-360 and on Bruker Avance DRX-500 spectrometers. [D₆]-DMSO (δ (¹H) = 2.49 ppm, δ (¹³C) = 39.5 ppm), CD₃OD (δ (¹H) = 3.35 ppm, δ (¹³C) = 49.3 ppm) and D₂O were used as solvents, the internal standard was [D₆]-DMSO. Chemical shifts are given in ppm relative to SiMe₄ (δ (¹H) = 0 ppm), coupling constants are given in Hertz. ¹H, Jmod, HH-COSY, HMQC-COSY, NOESY and HMBC experiments were used for assignment.

Proton and carbon atoms of the indigo glycosides are assigned as shown below:

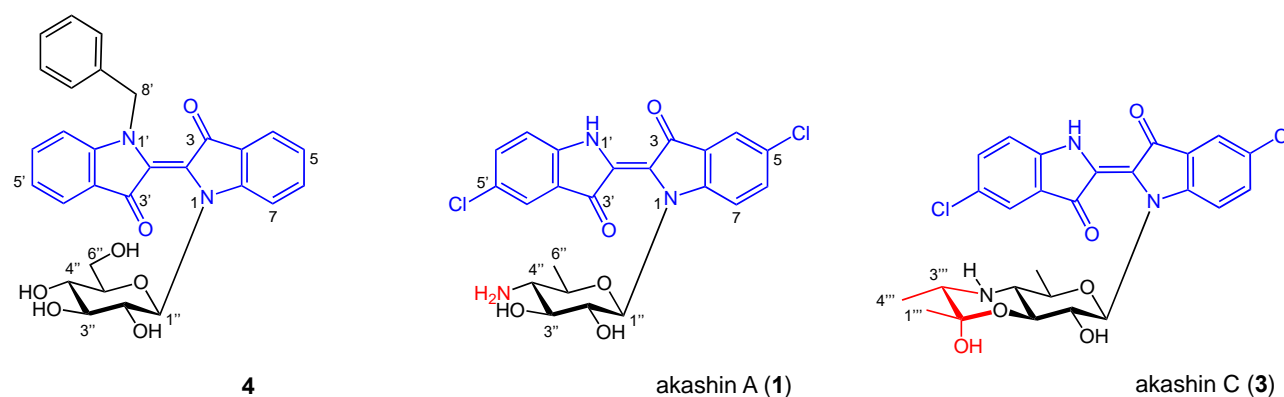


Figure S1: NMR assignment of indigo glycosides

3,4,6-Tri-O-acetyl-2-O-benzoyl- α -D-glucopyranosyl-trichloroacetimidate (5)

20 g (57.5 mmol, 1.0 eq) of 1,3,4,6-tetraacetylglucose **7** were dissolved in 200 mL of dichloromethane and 24 mL of pyridine. The solution was cooled to 0 °C followed by dropwise addition of 17 mL (146 mmol) of

benzoylchloride and subsequent stirring for 2½ h at room temperature. The reaction was stopped by adding 100 mL of saturated potassium hydrogen carbonate and 350 g of crushed ice. The mixture was stirred for further 1½ h. The aqueous phase was extracted with 200 mL of dichloromethane (2 x) and the combined organic layers were extracted with 200 mL of 2 N hydrochloric acid (2 x) and 200 mL of saturated potassium hydrogen carbonate. The organic phase was dried over MgSO₄, concentrated in vacuo and purified by flash chromatography (7 x 18 cm, eluent: cyclohexane/ethyl acetate 9:1 → 5:1 → 2:1 → 1:1).

yield of 1,3,4,6-tetra-*O*-acetyl-2-*O*-benzoyl- α -D-glucopyranoside **7a**:

25.9 g (57.4 mmol, 99 %, α/β 9:1)

R_f = 0.49 (cyclohexane/ethyl acetate 2:1)

$[\alpha]_D^{22} = +113.5^\circ$ (c = 1.02, dichloromethane)

C₁₉H₂₄O₁₁ (452.41)

ESI/MS: M_{cal.} = 452.13

M_{found} = 475.24 [M+Na]⁺, 927.51 [2M+Na]⁺

α -anomer:

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.86 (d, 2H, *J* = 7.6 Hz, H_o-Ar), 7.69 (dd, 1H, *J* = 7.6 Hz, H_p-Ar), 7.54 (dd, 2H, *J* = 7.6 Hz, H_m-Ar), 6.35 (d, 1H, *J*_{1,2} = 3.6 Hz, H-1), 5.53 (dd, 1H, *J*_{2,3} = *J*_{3,4} = 9.9 Hz, H-3), 5.28 (dd, 1H, *J*_{1,2} = 3.6 Hz, *J*_{2,3} = 9.9 Hz, H-2), 5.20 (dd, 1H, *J*_{3,4} = *J*_{4,5} = 9.9 Hz, H-4), 4.25-4.18 (m, 2H, H-5, H-6a), 4.07-4.01 (m, 1H, H-6b), 2.17 (s, 3H, Ac), 2.03 (s, 6H, 2Ac), 1.93 (s, 3H, Ac).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 170.6, 170.3, 169.7, 169.5 (CO-Ac), 165.2 (CO-Bz), 134.5 (C_p-Ar), 129.8 (C_m-Ar), 129.5 (C_o-Ar), 128.9 (C_i-Ar), 88.9 (C-1), 70.3 (C-5), 69.9 (C-2), 69.8 (C-3), 67.7 (C-4), 61.9 (C-6), 21.1, 20.9, 20.8 (Ac).

To a stirred solution of 8.70 g (19.4 mmol, 1.0 eq) of tetraacetyl-2-benzoyl- α -D-glucose **7a** in 25 mL of *N,N*-dimethylformamide were added 2.67 g (29 mmol, 1.5 eq) of hydrazine acetate over 15 minutes. After 90 minutes the reaction was stopped by addition of 12 mL of acetone and stirred until the excess of hydrazine acetate was dissolved. The solution was diluted with 400 mL of dichloromethane and extracted with water and saturated potassium hydrogen carbonate. The organic layer was dried over MgSO₄, concentrated in vacuo and purified by flash chromatography (5 x 18 cm, eluent: cyclohexane/acetone 3:1 → 2:1).

yield of 3,4,6-Tri-*O*-acetyl-2-*O*-benzoyl- α -D-glucopyranoside (**7b**):

6.22 g (15.2 mmol, 80 %, α/β 9:1)

R_f = 0.40 (cyclohexane/acetone 2:1)

$[\alpha]_D^{26} = +115.3^\circ$ (c = 0.52, dichloromethane)

C₁₉H₂₂O₁₀ (410.37)

ESI/MS: M_{cal.} = 410.12

M_{found} = 433.20 [M+Na]⁺, 843.33 [2M+Na]⁺

α -anomer:

¹H NMR (270 MHz, [D₆]-DMSO): δ = 7.90 (d, 2H, *J* = 7.6 Hz, H_o-Ar), 7.69 (dd, 1H, *J* = 7.6 Hz, H_p-Ar), 7.54 (dd, 2H, *J* = 7.6 Hz, H_m-Ar), 7.38 (d, 1H, *J*_{OH,1} = 4.9 Hz, OH), 5.57 (dd, 1H, *J*_{2,3} = *J*_{3,4} = 9.9 Hz, H-3), 5.43-5.39 (m, 1H, H-1), 5.02 (dd, 1H, *J*_{3,4} = *J*_{4,5} = 9.9 Hz, H-4), 4.93 (dd, 1H, *J*_{1,2} = 4.9 Hz, *J*_{2,3} = 9.9 Hz, H-2), 4.25-4.11 (m, 2H, H-5, H-6a), 4.10-4.00 (m, 1H, H-6b), 2.02 (s, 6H, 2Ac), 1.91 (s, 3H, Ac).

^{13}C NMR (68 MHz, $[\text{D}_6]$ -DMSO): δ = 170.7, 170.3, 169.9 (CO-Ac), 165.5 (CO-Bz), 134.4 (C_p -Ar), 129.8 (C_m -Ar), 129.4 (C_o -Ar), 129.4 (C_i -Ar), 88.6 (C-1), 72.5 (C-2), 69.9 (C-3), 68.8 (C-4), 67.0 (C-5), 62.7 (C-6), 21.1, 21.0, 20.9 (Ac).

11.65 g (28.4 mmol, 1.0 eq) of hemiacetal **7b** was dissolved in 160 mL of absolute dichloromethane and cooled to 0 °C. 28.5 mL (283.9 mmol, 10 eq) of trichloroacetonitrile were added followed by 0.85 mL (5.68 mmol, 0.2 eq) of DBU. After 60 minutes the solvent was concentrated in vacuo (20 °C) and the residue was purified by flash chromatography (7.5 x 11 cm, eluent: cyclohexane/acetone 5:1).

yield of imidate **7**: 14.20 g (25.6 mmol, 90.3 %),

R_f = 0.52 (cyclohexane/ethyl acetate 2:1)

$[\alpha]_D^{28}$ = - 5.5 ° (c = 0.53, dichloromethane)

$\text{C}_{21}\text{H}_{22}\text{Cl}_3\text{NO}_{10}$ (553.76)

ESI/MS: $M_{\text{cal.}}$ = 553.03

M_{found} = 576.14 [^{35}Cl]M+Na $^+$, 578.14 [^{37}Cl]M+Na $^+$

^1H -NMR (270 MHz, $[\text{D}_6]$ -DMSO): δ = 9.96 (s, 1H, NH), 7.88 (d, 2H, J = 7.4 Hz, H_o -Ar), 7.67 (dd, 1H, J = 7.4 Hz, H_p -Ar), 7.52 (dd, 2H, J = 7.4 Hz, H_m -Ar), 6.62 (bs, 1H, H-1), 5.62 (dd, 1H, $J_{2,3}$ = $J_{3,4}$ = 9.9 Hz, H-3), 5.40 (dd, 1H, $J_{1,2}$ = 2.3 Hz, $J_{2,3}$ = 9.9 Hz, H-2), 5.26 (dd, 1H, $J_{3,4}$ = $J_{4,5}$ = 9.9 Hz, H-4), 4.27-4.08 (m, 3H, H-5, H-6a, H-6b), 2.03 (s, 6H, 2xAc), 1.93 (s, 3H, Ac).

^{13}C -NMR (68 MHz, $[\text{D}_6]$ -DMSO): δ = 170.5, 170.2, 169.8 (CO-Ac), 165.1 (CO-Bz), 134.6 (C_p -Ar), 129.8 (C_m -Ar), 129.5 (C_o -Ar), 128.8 (C_i -Ar), 92.5 (C-1), 70.5 (C-2, C-5), 69.7 (C-3), 67.7 (C-4), 61.9 (C-6), 21.1, 20.8 (Ac).

N-Benzylindigo (**6**)

1 g (3.81 mmol, 1.0 eq) of indigo **8** was suspended in 25 mL of absolute *N,N*-dimethylformamide, 100 mg (2.5 mmol, 0.66 eq) of NaH (60 % in oil suspension) were added and the suspension stirred at room temperature. After 30 minutes further 100 mg (2.5 mmol, 0.66 eq) NaH (60 % in oil suspension) were added. After 2 h 540 μL (4.55 mmol, 1.2 eq) of benzylbromide were added dropwise, and the reaction was stirred for 1 h at room temperature. Subsequently, the reaction mixture was concentrated in vacuo (60 °C), the residue was dissolved in 200 mL of dichloromethane and extracted with water (3 x). The organic layer was dried over MgSO_4 , concentrated in vacuo and purified by flash chromatography (3.5 cm x 28 cm, eluent: toluene).

Yield of **6**: 641 mg (3.6 mmol, 48 %), blue solid

R_f = 0.57 (toluene/ethyl acetate 15:1)

UV/VIS: λ_{max} ($\lg \epsilon$) = 626 nm (3.89) in dichloromethane

$\text{C}_{23}\text{H}_{16}\text{N}_2\text{O}_2$ (352.39)

EI/MS (70 eV): m/z (%) = 352 (54) [M] $^+$, 261 (31) [M-Benzyl] $^+$, 91 (92) [Benzyl] $^+$

^1H -NMR (270 MHz, $[\text{D}_6]$ -DMSO): δ = 7.68 (d, 1H, $J_{4,5}$ = 7.7 Hz, H-4), 7.59 (dd, 1H, $J_{5,6}$ = $J_{6,7}$ = 7.7 Hz, H-6), 7.55 (d, 1H, $J_{4',5'}$ = 7.7 Hz, H-4'), 7.50 (dd, 1H, $J_{5',6'}$ = $J_{6',7'}$ = 7.7 Hz, H-6'), 7.37 (d, 1H, $J_{6,7}$ = 7.7 Hz, H-7), 7.35 (d, 1H, $J_{6',7'}$ = 7.7 Hz, H-7'), 7.26-7.16 (m, 3H, H_m -Ar, H_p -Ar), 7.12-7.07 (m, 2H, H_o -Ar), 7.06 (dd, 1H, $J_{4,5}$ = 7.7 Hz, H-5), 6.94 (dd, 1H, $J_{4',5'}$ = 7.7 Hz, H-5'), 5.78 (s, 2H, H-8').

^{13}C -NMR (68 MHz, $[\text{D}_6]$ -DMSO): δ = 188.8 (C_q -3'), 187.2 (C_q -3), 153.3, 152.5 (C_q -7a, C_q -7a'), 138.3 (C_i -Ar), 136.7, 136.5 (C-6, C-6'), 129.1, 127.7, 127.3 (C-Ar), 125.0 (C_q -2'), 124.4, 124.0 (C-4, C-4'), 123.3 (C_q -2), 121.7, 121.0 (C-5, C-5'), 121.2 (C_q -3a'), 119.6 (C_q -3a), 113.8 (C-7'), 112.7 (C-7), 50.3 (CH_2 -8').

N-[(2,3,4-Tri-*O*-acetyl-2-*O*-benzoyl- β -*D*-glucopyranosyl)-*N*'-benzyl]-indigo (**9**)

Under an inert gas, 764 mg (1.38 mmol, 1.0 eq) of imidate **5**, 500 mg (1.42 mmol, 1.03 eq) of *N*-benzylindigo **6** and 1.0 g of molecular sieves 4 Å were suspended in 10 mL of absolute dichloromethane, stirred for 30 minutes at room temperature and cooled to -18 °C. The reaction was started by adding of 100 μ L (0.55 mmol, 0.4 eq) of TMSOTf. After 3 h at -18 °C the suspension was slowly warmed to room temperature over 12 h. The reaction was quenched by adding 10 mL of saturated potassium hydrogen carbonate. After 10 minutes the mixture was filtered over Celite and washed with dichloromethane. The organic filtrate was extracted with water, saturated potassium hydrogen carbonate and brine. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (5 x 20 cm, solvent: toluene/ethyl acetate 15:1 → 12:1 → 9:1). Unreacted *N*-benzylindigo **6** was recovered 124 mg (0.35 mmol, 25 %)

Yield of **9**: 232 mg (0.31 mmol, 22.6 %), blue solid

R_f = 0.27 (cyclohexane/ethyl acetate 1.5:1)

UV/VIS: λ_{\max} (lg ϵ) = 630 nm (3.80) in dichloromethane

$[\alpha]_D^{23} = +5428.6^\circ$ (c = 7.0 μ g/mL, dichloromethane)

C₄₂H₃₆N₂O₁₁ (744.74)

ESI/MS: M_{cal.} = 744.23

M_{found} = 393.39 [C₁₉H₂₁O₉]⁺, 745.76 [M+H]⁺, 767.76 [M+Na]⁺, 783.75 [M+K]⁺, 1512.62 [2M+Na]⁺

¹H-NMR (360 MHz, [D₆]-DMSO): δ = 7.73 (d, 1H, $J_{6,7} = 7.9$ Hz, H-7), 7.69 (d, 1H, $J_{4',5'} = 7.6$ Hz, H-4'), 7.69 (dd, 2H, $J = 7.9$ Hz, H-6, H_p-Ar), 7.54 (dd, 2H, $J = 8.0$ Hz, H_m-Ar), 7.47 (d, 1H, $J_{4,5} = 7.3$ Hz, H-4), 7.40 (dd, 2H, $J = 8.0$ Hz, H-6', H_p-Ar), 7.35-7.32 (m, 4H, H_o-Ar), 7.11 (dd, 1H, $J_{4,5} = 7.3$ Hz, $J_{5,6} = 7.9$ Hz, H-5), 7.03 (dd, 1H, $J_{4',5'} = 7.6$ Hz, $J_{5',6'} = 8.0$ Hz, H-5'), 6.92 (dd, 2H, $J = 7.7$ Hz, H_m-Ar), 6.66 (d, 1H, $J_{6',7'} = 8.0$ Hz, H-7'), 6.01 (d, 1H, $J_{1'',2''} = 8.7$ Hz, H-1''), 5.74-5.68 (m, 2H, H-2'', H-3''), 5.33 (dd, 1H, $J_{3'',4''} = J_{4'',5''} = 9.4$ Hz, H-4''), 5.02 (d, 1H, $J_{\text{gem}} = 16.8$ Hz, H-8'), 4.71 (d, 1H, $J_{\text{gem}} = 16.8$ Hz, H-8''), 4.31-4.26 (m, 1H, H-5''), 4.23-4.14 (m, 1H, H-6a''), 4.08-4.02 (m, 1H, H-6b''), 2.22 (s, 3H, Ac), 2.03 (s, 3H, Ac), 1.74 (s, 3H, Ac).

¹³C-NMR (90 MHz, [D₆]-DMSO): δ = 185.1 (C_q-3'), 183.3 (C_q-3), 170.3, 169.9, 169.4 (CO-Ac), 163.9 (CO-Bz), 151.8 (C_q-7a'), 149.5 (C_q-7a), 135.1 (C-6'), 135.0 (C-6), 133.6, 133.4 (C_p-Ar), 128.5, 128.1 (C_m-Ar), 128.0 (C_o-Ar), 127.8 (C_r-Ar), 123.7 (C-4), 123.4 (C_q-2'), 123.1 (C-4'), 122.2 (C-5), 121.7 (C_q-2), 121.4 (C-5'), 119.0 (C-3a, C-3a'), 115.7 (C-7), 112.7 (C-7'), 87.5 (C-1''), 74.1 (C-5''), 73.1 (C-3''), 68.1 (C-2''), 68.6 (C-4''), 61.4 (C-6''), 53.5 (CH₂-8'), 20.6, 20.4, 20.2 (Ac).

N-(2,3,4-Tri-*O*-acetyl-2-*O*-benzoyl- β -*D*-glucopyranosyl)-indigo (**9b**)

320 mg (0.43 mmol, 1.0 eq) of glycoside **9** were dissolved in 8 mL of toluene/acetic acid (1:1) and stirred in an open flask for 28 h at 50 °C. The solution was diluted with 150 mL of dichloromethane, extracted with water and saturated potassium hydrogen carbonate. The organic layer was dried over MgSO₄, concentrated in vacuo and purified by flash chromatography (5 x 18 cm, eluent: toluene/ethyl acetate 15:1 → 12:1).

Yield: 173 mg (0.26 mmol, 61.4 %), blue solid

R_f = 0.41 (toluene/ethyl acetate 5:1)

UV/VIS: λ_{\max} (lg ϵ) = 600 nm (4.04) in dichloromethane

$[\alpha]_D^{23} = +2328.2^\circ$ (c = 3.9 μ g/mL, dichloromethane)

C₃₅H₃₀N₂O₁₁ (654.62)

ESI/MS: M_{cal.} = 654.18

$$M_{\text{found}} = 393.39 [\text{C}_{19}\text{H}_{21}\text{O}_9]^+, 655.30 [\text{M}+\text{H}]^+, 677.21 [\text{M}+\text{Na}]^+, 694.13 [\text{M}+\text{K}]^+$$

$^1\text{H-NMR}$ (360 MHz, $[\text{D}_6]$ -DMSO): δ = 10.75 (s, 1H, N'H), 7.76 (d, 1H, $J_{6,7}$ = 7.5 Hz, H-7), 7.68 (dd, 1H, $J_{5,6}$ = $J_{6,7}$ = 7.5 Hz, H-6), 7.67 (d, 1H, $J_{4',5'}$ = 7.5 Hz, H-4'), 7.57-7.50 (m, 5H, H-4, H-6', H_o-Ar, H_p-Ar), 7.34 (d, 1H, $J_{6',7'}$ = 7.6 Hz, H-7'), 7.30 (dd, 1H, J = 7.6 Hz, H_m-Ar), 7.14 (dd, 1H, $J_{4,5}$ = $J_{5,6}$ = 7.5 Hz, H-5), 7.00 (dd, 1H, $J_{4',5'}$ = $J_{5',6'}$ = 7.6 Hz, H-5'), 6.56 (d, 1H, $J_{1'',2''}$ = 9.1 Hz, H-1''), 5.60 (dd, 1H, $J_{1'',2''}$ = $J_{2'',3''}$ = 9.1 Hz, H-2''), 5.52 (dd, 1H, $J_{2'',3''}$ = $J_{3'',4''}$ = 9.1 Hz, H-3''), 5.36 (dd, 1H, $J_{3'',4''}$ = $J_{4'',5''}$ = 9.1 Hz, H-4''), 4.34-4.28 (m, 2H, H-6a'', H-6b''), 4.23-4.17 (m, 1H, H-5''), 2.13 (s, 3H, Ac), 2.04 (s, 3H, Ac), 1.78 (s, 3H, Ac).

$^{13}\text{C-NMR}$ (90 MHz, $[\text{D}_6]$ -DMSO): δ = 186.9 (C_q-3'), 186.8 (C_q-3), 170.1, 169.4 (CO-Ac), 163.9 (CO-Bz), 152.4 (C_q-7a'), 149.8 (C_q-7a), 136.5 (C-6'), 135.7 (C-6), 133.6 (C_p-Ar), 129.0 (C_m-Ar), 128.6 (C_o-Ar), 127.9 (C_i-Ar), 124.6 (C_q-2'), 124.2 (C-4'), 123.3 (C-4), 122.6 (C-5), 122.1 (C_q-2), 120.8 (C-5'), 119.0 (C-3a, C-3a'), 116.2 (C-7), 113.3 (C-7'), 85.1 (C-1''), 74.2 (C-5''), 73.0 (C-3''), 68.7 (C-2''), 68.0 (C-4''), 61.9 (C-6''), 20.6, 20.4, 20.2 (Ac).

N-β-D-Glucopyranosyl)-indigo **4**

Under an argon atmosphere 179 mg (0.27 mmol, 1.0 eq) of indigo **9b** were dissolved in 9 mL of absolute methanol. 9 mL of NaOMe (c = 0.01 mol/l) in MeOH were added and the reaction was stirred for 11 h at room temperature. The reaction was stopped by neutralization with Amberlyst 15 (H⁺). The ion exchange resin was removed by filtration over Celite and washed with methanol. The filtrate was concentrated and purified in three aliquots by solid phase extraction (SepPak[®]-C₁₈, 2 g) with a gradient of 5-15 % acetonitrile in water in 1 %-steps. The product eluted between 12-15 % acetonitrile and was lyophilized.

yield: 58 mg (0.14 mmol, 50.7 %), blue solid

R_f: 0.25 (dichloromethane/methanol 10:1)

UV/VIS: λ_{max} (lgε) = 606 nm (4.08) in methanol

$[\alpha]_{\text{D}}^{27} = +2258^\circ$ (c = 6.2 μg/mL, methanol)

C₂₂H₂₀N₂O₇ (424.40)

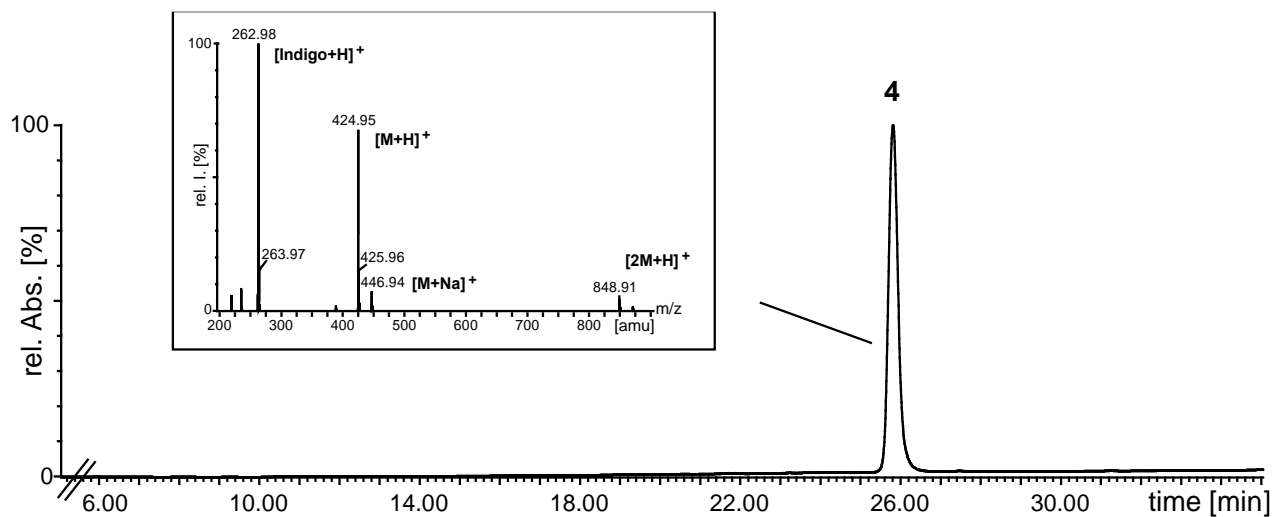
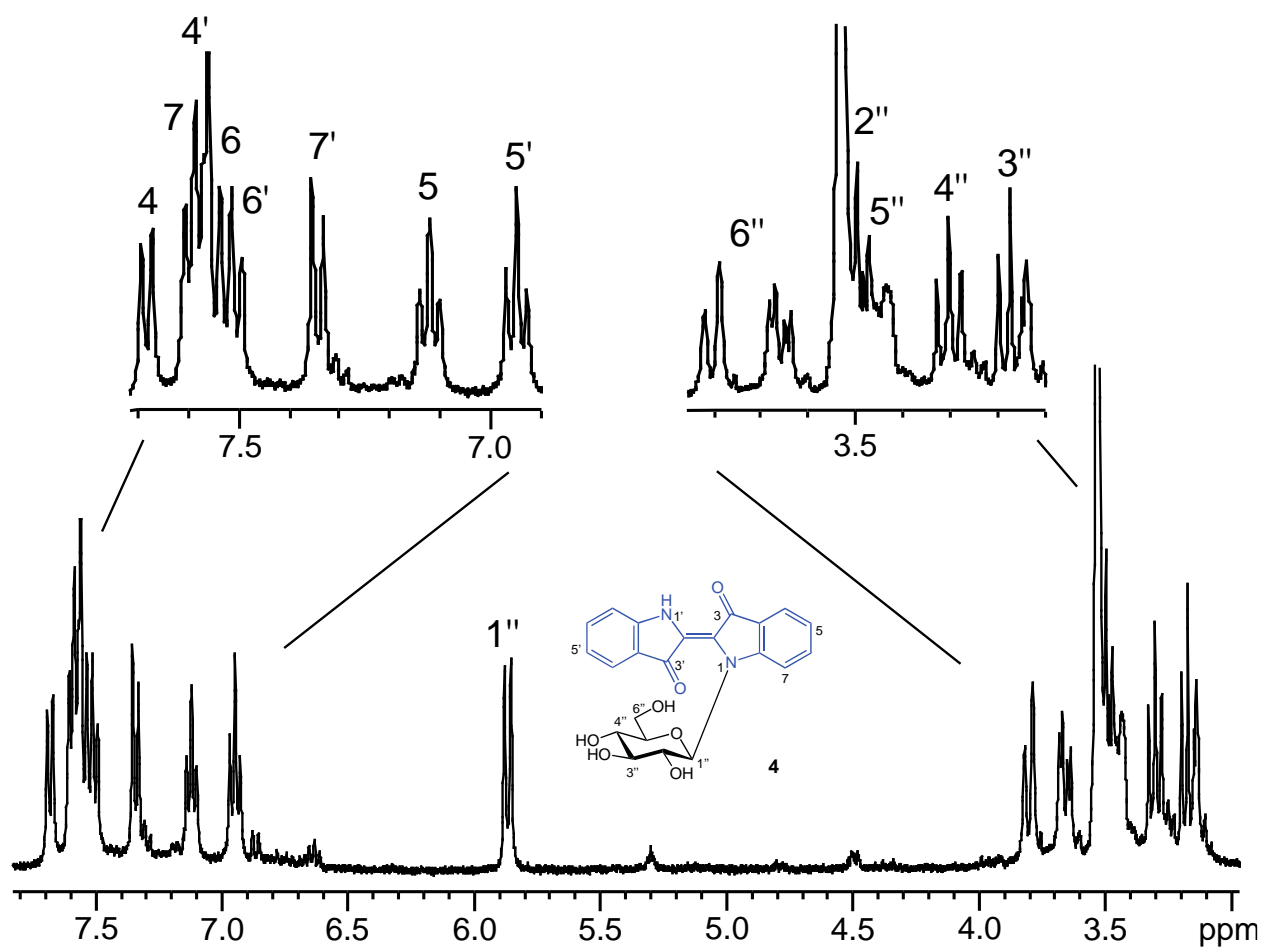
ESI/MS: M_{cal.} = 424.13

$$M_{\text{found}} = 425.07 [\text{M}+\text{H}]^+, 447.09 [\text{M}+\text{Na}]^+, 871.18 [2\text{M}+\text{Na}]^+$$

$^1\text{H-NMR}$ (500 MHz, $[\text{D}_6]$ -DMSO with D₂O): δ = 7.68 (d, 1H, $J_{4,5}$ = 7.5 Hz, H-4), 7.59 (d, 1H, $J_{6,7}$ = 7.5 Hz, H-7), 7.56 (d, 1H, $J_{4',5'}$ = 7.7 Hz, H-4'), 7.55 (dd, 1H, $J_{5,6}$ = $J_{6,7}$ = 7.5 Hz, H-6), 7.51 (d, 1H, $J_{5',6'}$ = $J_{6',7'}$ = 7.7 Hz, H-6'), 7.33 (d, 1H, $J_{6',7'}$ = 7.7 Hz, H-7'), 7.12 (dd, 1H, $J_{4,5}$ = $J_{5,6}$ = 7.5 Hz, H-5), 6.95 (dd, 1H, $J_{4',5'}$ = $J_{5',6'}$ = 7.7 Hz, H-5'), 5.87 (d, 1H, $J_{1'',2''}$ = 9.1 Hz, H-1''), 3.80 (dd, 1H, $J_{5'',6a''}$ < 1.0 Hz, $J_{6a'',6b''}$ = 11.3 Hz, H-6a''), 3.66 (dd, 1H, $J_{5'',6b''}$ = 4.9 Hz, $J_{6a'',6b''}$ = 11.3 Hz, H-6b''), 3.51 (dd, 1H, $J_{1'',2''}$ = $J_{2'',3''}$ = 9.1 Hz, H-2''), 3.47-3.39 (m, 1H, H-5''), 3.31 (dd, 1H, $J_{3'',4''}$ = $J_{4'',5''}$ = 9.1 Hz, H-4''), 3.19 (dd, 1H, $J_{2'',3''}$ = $J_{3'',4''}$ = 9.1 Hz, H-3'').

$^1\text{H-NMR}$ (360 MHz, $[\text{D}_6]$ -DMSO): δ = 10.85 (bs, 1H, N'H), 4.98 (d, 1H, $J_{\text{OH-}3'',3''}$ = 5.8 Hz, OH-3''), 4.96 (d, 1H, $J_{\text{OH-}4'',4''}$ = 5.2 Hz, OH-4''), 4.82 (d, 1H, $J_{\text{OH-}2'',2''}$ = 6.8 Hz, OH-2''), 4.68 (t, 1H, $J_{\text{OH-}6'',6''}$ = 5.7 Hz, OH-6'').

$^{13}\text{C-NMR}$ (126 MHz, $[\text{D}_6]$ -DMSO mit D₂O): δ = 188.2 (C_q-3'), 186.5 (C_q-3), 152.1 (C_q-7a'), 150.8 (C_q-7a), 136.0 (C-6'), 135.1 (C-6), 124.0 (C-4'), 123.9 (C_q-2), 123.6 (C_q-2'), 123.2 (C-4), 122.7 (C_q-3a), 121.7 (C-5), 120.3 (C-5'), 119.1 (C_q-3a'), 116.6 (C-7), 113.1 (C-7'), 88.5 (C-1''), 80.7 (C-5''), 78.0 (C-3''), 70.2, 70.1 (C-2'', C-4''), 61.4 (C-6'').

a) LC-MS of **4**:b) assigned ¹H-NMR of **4** (500 MHz):Figure S2: LC-MS (a) and NMR (b) of **4**.

1,2,3-Tri-O-benzoyl-4,6-O-benzylidene-D-galactopyranose (12)

The synthesis of **12** was carried out following the procedure of Gros and Deulofeu. The anomers obtained after benzylation of 10g of 4,6-benzylidene-D-galactose were separated by flash chromatography (7.5 x 12 cm, eluent: cyclohexane/ethyl acetate 9:1 → 6:1 → 5:1 → 3:1 → 2:1 → 1:1).

***α*-anomer 12 α :**

yield: 18.88 g (32.5 mmol, 87 %)

R_f = 0.62 (dichloromethane/methanol 2:1)

[α]_D²⁴ = - 79.2 ° (c = 0.50, dichloromethane)

C₃₄H₂₈O₉ (580.58)

ESI/MS: M_{cal.} = 580.17

M_{found} = 603.21 [M+Na]⁺

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 8.14 (d, 2H, *J* = 7.3 Hz, H-Ar), 7.88 (d, 2H, *J* = 7.3 Hz, H-Ar), 7.74 (d, 2H, *J* = 7.3 Hz, H-Ar), 7.62 (dd, 3H, *J* = 7.7 Hz, H-Ar), 7.57-7.33 (m, 10H, H-Ar), 6.79 (d, 1H, *J*_{1,2} = 3.4 Hz, H-1), 6.01 (dd, 1H, *J*_{2,3} = 10.9 Hz, *J*_{3,4} = 3.1 Hz, H-3), 5.80 (dd, 1H, *J*_{1,2} = 3.4 Hz, *J*_{2,3} = 10.9 Hz, H-2), 5.78 (s, 1H, H-7), 4.87 (dd, 1H, *J*_{3,4} = *J*_{4,5} = 3.1 Hz, H-4), 4.41-4.39 (m, 1H, H-5), 4.22-4.14 (m, 2H, H-6a, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.6, 165.4, 164.9 (CO-Bz), 138.6 (C_i-benzylidene), 134.7, 134.4, 133.8 (C_p-Ar), 130.2, 129.8, 129.7, 129.6, 129.5 (C-Ar), 129.4 (C_i-Ar), 129.3 (C-Ar), 129.2 (C_i-Ar), 129.1 (C-Ar), 129.0 (C_i-Ar), 128.8, 126.6 (C-Ar), 100.1 (C-7), 91.4 (C-1), 74.0 (C-4), 69.3 (C-3), 68.8 (C-6), 68.0 (C-2), 65.5 (C-5).

***β*-anomer 12 β :**

yield: 2.72 g (4.69 mmol, 13 %)

R_f = 0.49 (dichloromethane/methanol 2:1)

[α]_D²⁴ = - 14.2 ° (c = 0.50, dichloromethane)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 8.00-7.80 (m, 6H, H-Ar), 7.70-7.32 (m, 14H, H-Ar), 6.45 (d, 1H, *J*_{1,2} = 7.5 Hz, H-1), 5.92 (dd, 1H, *J*_{3,2} = 10.5 Hz, *J*_{3,4} = 2.9 Hz, H-3), 5.84 (dd, 1H, *J*_{2,1} = 7.5 Hz, *J*_{2,3} = 10.5 Hz, H-2), 5.76 (s, 1H, H-7), 4.79 (dd, 1H, *J*_{4,3} = *J*_{4,5} = 2.9 Hz, H-4), 4.27-4.18 (m, 3H, H-5, H-6a, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.6, 165.5, 164.7 (CO-Bz), 138.6 (C_i-benzylidene), 134.8, 134.4, 133.4 (C_p-Ar), 129.9, 129.8, 129.7, 129.6, 129.4, (C-Ar), 129.3 (C_i-Ar), 129.2 (C-Ar), 129.1 (C_i-Ar), 128.7 (C-Ar), 128.6 (C_i-Ar), 126.5 (C-Ar), 100.1 (C-7), 92.9 (C-1), 73.6 (C-4), 72.1 (C-3), 69.3 (C-2), 68.5 (C-6), 67.6 (C-5).

1,2,3-Tri-O-benzoyl-6-deoxy-6-iodo- α -D-galactopyranose (13 α)

16.45 g (28.4 mmol, 1.0 eq) of tribenzoyl-4,6-O-benzylidene- β -D-galactose **12 α** were dissolved in 165 mL of dry acetonitrile und 107 mL of absolute methanol. 3.34 g (17.6 mmol, 0.62 eq) of *p*-toluenesulfonic acid monohydrate in 34 mL of absolute methanol were added over 10 minutes. After 3 h of stirring 1.67 g (8.8 mmol, 0.31 eq) of *p*-toluenesulfonic acid monohydrate in 17 mL absolute methanol were added. After 21 h the reaction was quenched by adding 4.2 mL of triethylamine. The reaction mixture was concentrated in vacuo and the residue was purified by flash chromatography (7.5 x 16 cm, eluent: cyclohexane/ethyl acetate 6:1 → 5:1 → 3:1 → 1:1 → 1:2).

yield of 1,2,3-tri-O-benzoyl- α -D-galactopyranose (**12 α 1**):

10.35 g (21.0 mmol, 74.1 %)

R_f = 0.24 (cyclohexane/ethyl acetate 1.5:1)

$[\alpha]_D^{26} = +277^\circ$ (c = 0.50, dichloromethane)

C₂₇H₂₄O₉ (492.47)

ESI/MS: M_{cal.} = 492.14

M_{found} = 515.29 [M+Na]⁺, 1007.53 [2M+Na]⁺

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 8.09 (d, 2H, J = 7.9 Hz, H-Ar), 8.01 (d, 2H, J = 7.9 Hz, H-Ar), 7.77-7.69 (m, 3H, H-Ar), 7.66-7.43 (m, 6H, H-Ar), 7.37 (dd, 2H, J = 7.7 Hz, H-Ar), 6.65 (d, 1H, J_{1,2} = 3.4 Hz, H-1), 5.81 (dd, 1H, J_{1,2} = 3.4 Hz, J_{2,3} = 10.5 Hz, H-2), 5.70-5.73 (m, 1H, OH-4), 5.68 (dd, 1H, J_{2,3} = 10.5 Hz, J_{3,4} = 2.4 Hz, H-3), 4.81 (t, 1H, J_{OH,6,6} = 5.6 Hz, OH-6), 4.44-4.33 (m, 1H, H-4), 4.21 (dd, 1H, J_{4,5} = J_{5,6} = 6.0 Hz, H-5), 3.69-3.52 (m, 2H, H-6a, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.8, 165.6, 164.9 (CO-Bz), 134.7, 134.3, 134.1 (C_p-Ar), 130.1 (C-Ar), 130.0 (C_r-Ar), 129.9, 129.6, 129.5 (C-Ar), 129.4 (C_r-Ar), 129.3, 129.2 (C-Ar), 129.2 (C_r-Ar), 91.1 (C-1), 74.8 (C-5), 71.9 (C-3), 68.5 (C-2), 66.5 (C-4), 60.4 (C-6).

3.0 g (6.09 mmol, 1.0 eq) of tribenzoyl-α-D-galactose (**12α-1**) and 4.11 g (18.27 mmol, 3.0 eq) of *N*-iodosuccinimide were dissolved in 60 mL of absolute *N,N*-dimethylformamide and cooled to 0 °C. 6.39 g (24.36 mmol, 4.0 eq) of triphenylphosphine were added over 20 minutes, the reaction mixture was heated to 55 °C. After stirring for 24 h the reaction was stopped by adding of 15 mL of methanol. The organic layer was concentrated in vacuo (60 °C), diluted with dichloromethane, washed with water, dried (MgSO₄) and filtered. The filtrate was concentrated and the residue purified by flash chromatography (5 x 20 cm, eluent: cyclohexane/ethyl acetate 9:1 → 6:1).

yield of **13α**: 2.69 g (4.46 mmol, 73.3 %)

R_f = 0.40 (cyclohexane/ethyl acetate 3:1)

$[\alpha]_D^{24} = -142.9^\circ$ (c = 0.50, dichloromethane)

C₂₇H₂₃IO₈ (602.37)

ESI/MS: M_{cal.} = 602.04

M_{found} = 602.79 [M+H]⁺, 1227.57 [2M+Na]⁺

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 8.11 (d, 2H, J = 7.3 Hz, H-Ar), 7.95 (d, 2H, J = 7.3 Hz, H-Ar), 7.79-7.71 (m, 3H, H-Ar), 7.60 (dd, 3H, J = 7.5 Hz, H-Ar), 7.58-7.52 (m, 1H, H-Ar), 7.47 (dd, 2H, J = 7.5 Hz, H-Ar), 7.37 (dd, 2H, J = 7.7 Hz, H-Ar), 6.64 (d, 1H, J_{1,2} = 3.4 Hz, H-1), 5.97 (d, 1H, J_{OH,4,4} = 5.6 Hz, OH-4), 5.80 (dd, 1H, J_{1,2} = 3.4 Hz, J_{2,3} = 10.6 Hz, H-2), 5.73 (dd, 1H, J_{2,3} = 10.6 Hz, J_{3,4} = 1.9 Hz, H-3), 4.53 (ddd, 1H, J_{3,4} = 1.9 Hz, J_{4,5} = J_{OH,4,4} = 5.5 Hz, H-4), 4.45-4.41 (m, 1H, H-5), 3.43 (dd, 1H, J_{5,6a} = 6.1 Hz, J_{6a,6b} = 10.0 Hz, H-6a), 3.32 (dd, 1H, J_{5,6b} = 7.7 Hz, J_{6a,6b} = 10.0 Hz, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.8, 165.5, 164.8 (CO-Bz), 134.7, 134.3, 134.1 (C_p-Ar), 130.2, 129.9 (C-Ar), 129.8 (C_r-Ar), 129.6, 129.5, 129.3 (C-Ar), 129.2 (C_r-Ar), 90.8 (C-1), 74.7 (C-5), 71.6 (C-3), 68.0 (C-2), 67.5 (C-4), 3.8 (C-6).

1,2,3-Tri-O-benzoyl-6-deoxy-6-iodo-β-D-galactopyranose (**13β**)

5.0 g (8.6 mmol, 1.0 eq) of tribenzoyl-4,6-O-benzylidene-β-D-galactose **12β** were dissolved in 50 mL of acetonitrile and 50 mL of methanol and heated to 80 °C. 5.0 mL of hydrochloric acid (2 N) were added dropwise and the reaction was stirred under reflux for 2 h. The reaction was stopped by adding 4.2 mL of triethylamine. The organic layer was concentrated in vacuo, diluted with dichloromethane, washed with

water, dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo und the residue purified by flash chromatography (5 x 25 cm, eluent: cyclohexane/ethyl acetate 6:1 → 1:1).

yield of 1,2,3-tri-*O*-benzoyl-β-D-galactopyranose (**12β-1**):

3.44 g (6.99 mmol, 81.2 %)

R_f = 0.57 (cyclohexane/acetone 1:1)

[α]_D²⁶ = -247.2 ° (c = 0.54, dichloromethane)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.91-7.82 (m, 6H, H-Ar), 7.70-7.38 (m, 9H, H-Ar), 6.28 (d, 1H, J_{1,2} = 8.4 Hz, H-1), 5.84 (dd, 1H, J_{1,2} = 8.4 Hz, J_{2,3} = 10.2 Hz, H-2), 5.59 (d, 1H, J_{OH-4,4} = 5.5 Hz, OH-4), 5.53 (dd, 1H, J_{2,3} = 10.2 Hz, J_{3,4} = 3.0 Hz, H-3), 4.88 (t, 1H, J_{OH-6,6} = 5.7 Hz, OH-6), 4.27 (dd, 1H, J_{OH-4,4} = 5.5 Hz, J_{3,4} = 3.0 Hz, H-4), 4.05 (dd, 1H, J_{5,6} = 8.6 Hz, H-5), 3.70-3.52 (m, 2H, H-6a, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.7, 164.6 (CO-Bz), 134.7, 134.3, 134.1 (C_p-Ar), 129.9, 129.8, 129.6, 129.4 (C-Ar), 129.3 (C_r-Ar), 129.2 (C-Ar), 128.8 (C_r-Ar), 93.2 (C-1), 76.7 (C-5), 74.8 (C-3), 69.9 (C-2), 65.9 (C-4), 60.1 (C-6).

6.88 g (14.0 mmol, 1.0 eq) of tribenzoyl-β-D-galactose (**12β-1**) and 9.53 g (42.3 mmol, 3.0 eq) *N*-iodosuccinimide were dissolved in 140 mL of absolute *N,N*-dimethylformamide. and cooled to 0 °C. 11.0 g (41.9 mmol, 3.0 eq) of triphenylphosphine were added over 20 minutes and the reaction mixture was heated to 65 °C. After stirring for 18h the reaction was stopped by adding 15 mL of methanol. The organic layer was concentrated in vacuo (60 °C), the residue diluted with dichloromethane, washed with water, dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo und the residue purified by flash chromatography (7.5 x 18 cm, eluent: cyclohexane/ethyl acetate 9:1 → 3:1).

yield of **13β**: 7.12 g (11.8 mmol, 85 %)

R_f = 0.50 (cyclohexane/ethyl acetate 3:1)

[α]_D²⁸ = -238.3 ° (c = 0.50, dichloromethane)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.93-7.80 (m, 6H, H-Ar), 7.70-7.39 (m, 9H, H-Ar), 6.31 (d, 1H, J_{1,2} = 8.2 Hz, H-1), 5.88 (d, 1H, J_{OH-4,4} = 5.5 Hz, OH-4), 5.81 (dd, 1H, J_{1,2} = 8.2 Hz, J_{2,3} = 10.3 Hz, H-2), 5.57 (dd, 1H, J_{2,3} = 10.3 Hz, J_{3,4} = 2.8 Hz, H-3), 4.41 (ddd, 1H, J_{3,4} = 2.8 Hz, J_{OH-4,4} = J_{4,5} = 5.5 Hz, H-4), 4.35 (m, 1H, H-5), 3.47 (dd, 1H, J_{5,6a} = 6.6 Hz, J_{6a,6b} = 10.1 Hz, H-6a), 3.30 (dd, 1H, J_{5,6b} = 7.3 Hz, J_{6a,6b} = 10.1 Hz, H-6b).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.7, 164.5 (CO-Bz), 134.8, 134.3, 134.1 (C_p-Ar), 129.9, 129.8 (C-Ar), 129.7 (C_r-Ar), 129.6, 129.4, 129.2 (C-Ar), 129.2 (C_r-Ar), 92.9 (C-1), 76.4 (C-5), 74.5 (C-3), 69.5 (C-2), 66.8 (C-4), 3.5 (C-6).

1,2,3-Tri-O-benzoyl-6-deoxy-α-D-galactopyranose (**14α**)

8.73 g (8.22 mmol, 1.5 eq) of Pd/C (10 % Pd) were suspended in 150 mL of absolute methanol containing 17 mL of acetic acid (98 %) and stirred for 2 h under a hydrogen atmosphere. 3.42 g (5.68 mmol, 1.0 eq) of iodide **13α** dissolved in 20 mL of absolute methanol were added and the reaction mixture was stirred for 2 d under a hydrogen atmosphere. Subsequently, the catalyst was removed by filtration over Celite and washed extensively with dichloromethane. The combined organic layers were extracted with saturated potassium hydrogen carbonate and water, dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo und the

residue purified by flash chromatography (5 x 18 cm, eluent: cyclohexane/ethyl acetate 8:1 → 7:1 → 6:1 → 4:1). 1.02 g (1.69 mmol, 29.8 %) of the starting material **13** α were recovered.

yield: 1.51 g (3.17 mmol, 55.9 %)

R_f = 0.22 (cyclohexane/ethyl acetate 4:1)

[α]_D²⁴ = + 155.0 ° (c = 0.50, dichloromethane)

C₂₂H₂₄O₈ (476.47)

ESI/MS: M_{cal.} = 476.15

M_{found} = 477.12 [M+H]⁺, 499.09 [M+Na]⁺

¹H-NMR (360 MHz, [D₆]-DMSO): δ = 8.07 (d, 2H, *J* = 7.5 Hz, H-Ar), 7.93 (d, 2H, *J* = 7.5 Hz, H-Ar), 7.75-7.70 (m, 3H, H-Ar), 7.65-7.57 (m, 3H, H-Ar), 7.53 (dd, 2H, *J* = 7.5 Hz, H-Ar), 7.47 (dd, 2H, *J* = 7.9 Hz, H-Ar), 7.36 (dd, 2H, *J* = 7.9 Hz, H-Ar), 6.58 (d, 1H, *J*_{1,2} = 3.2 Hz, H-1), 5.78-5.70 (m, 3H, H-2, H-3, OH-4), 4.43 (dq, 1H, *J*_{4,5} < 1.0 Hz, *J*_{5,6} = 6.2 Hz, H-5), 4.15-4.12 (m, 1H, H-4), 1.22 (d, 3H, *J*_{5,6} = 6.2 Hz, H-6).

¹³C-NMR (90 MHz, [D₆]-DMSO): δ = 165.3, 165.0, 164.4 (CO-Bz), 134.0, 133.7, 133.5 (C_p-Ar), 129.5 (C-Ar), 129.3 (C_r-Ar), 129.0 (C-Ar), 129.0 (C_r-Ar), 128.7 (C-Ar), 128.6 (C_r-Ar), 90.7 (C-1), 71.2 (C-3), 69.5 (C-5), 68.7 (C-4), 67.7 (C-2), 16.2 (C-6).

1,2,3-Tri-O-benzoyl-6-deoxy- β -D-galactopyranose (**14** β)

11.2 g (10.6 mmol, 2.06 eq) of Pd/C-catalyst (10 % Pd) were suspended in 155 mL of absolute methanol and 16 mL of acetic acid (98 %) and stirred for 2 h under a hydrogen atmosphere. A solution of 3.1 g (5.15 mmol, 1.0 eq) of iodide **13** β in 20 mL of absolute methanol was added. After 2 d the catalyst was removed by filtration over Celite and repeatedly washed with dichloromethane. The combined organic layer was extracted with saturated potassium hydrogen carbonate and water, dried (MgSO₄) and concentrated in vacuo. The residue was purified by flash chromatography (5 x 20 cm, eluent: cyclohexane/ethyl acetate 8:1 → 5:1). 1.9 g (3.15 mmol, 61.3 %) of the starting material **13** β were recovered.

yield: 867 mg (1.82 mmol, 35.3 %)

R_f = 0.34 (cyclohexane/ethyl acetate 4:1)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.91-7.82 (m, 5H, H-Ar), 7.70-7.35 (m, 10H, H-Ar), 6.22 (d, 1H, *J*_{1,2} = 8.1 Hz, H-1), 5.79 (dd, 1H, *J*_{1,2} = 8.1 Hz, *J*_{2,3} = 10.3 Hz, H-2), 5.62 (d, 1H, *J*_{OH-4,4} = 5.8 Hz, OH-4), 5.53 (dd, 1H, *J*_{2,3} = 10.3 Hz, *J*_{3,4} = 3.0 Hz, H-3), 4.21 (dq, 1H, *J*_{4,5} < 1.0 Hz, *J*_{5,6} = 6.2 Hz, H-5), 4.03-4.00 (m, 1H, H-4), 1.25 (d, 3H, *J*_{5,6} = 6.2 Hz, H-6).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.7, 165.6, 164.7 (CO-Bz), 134.7, 134.3, 134.1 (C_p-Ar), 129.9, 129.8 (C-Ar), 129.6 (C_r-Ar), 129.4 (C-Ar), 129.0 (C_r-Ar), 93.2 (C-1), 74.8 (C-3), 71.8 (C-5), 69.6 (C-2), 68.8 (C-4), 16.7 (C-6).

4-Azido-1,2,3-tri-O-benzoyl-4,6-dideoxy- α -D-glucopyranose (**15** α)

7.3 g (15.3 mmol, 1.0 eq) of 6-deoxysugar **14** α were dissolved in 112 mL of absolute dichloromethane and 19 mL (236 mmol) of absolute pyridine and cooled to -18 °C. 6.2 mL (37.6 mmol, 2.4 eq) of trifluoromethane sulfonic acid anhydride was added. After stirring for 110 minutes the solution was extracted with water, 2 x saturated potassium hydrogen carbonate and 2 N hydrochloric acid. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated and dried in high vacuum. Subsequently, 9.0 g (138 mmol, 8.5 eq) of sodium azide was added to the crude triflate. The mixture was suspended in 240 mL of absolute *N,N*-

dimethylformamide and stirred for 30 minutes at room temperature. The organic layer was concentrated in vacuo (60 °C) and the residue was diluted with 300mL of dichloromethane. The solution was extracted with water, dried over MgSO₄ and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (7.5 x 17 cm, eluent: cyclohexane/ethyl acetate 12:1 → 9:1).

yield: 7.05 g (14.1 mmol, 92 % over 2 steps)

R_f = 0.47 (cyclohexane/acetone 3:1)

[α]_D²⁵ = + 213.2 ° (c = 0.55, dichloromethane)

C₂₇H₂₃N₃O₇ (501.49)

ESI/MS: M_{cal.} = 501.15

M_{found} = 518.78 [M+H₂O]⁺, 523.73 [M+Na]⁺

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 8.06 (d, 2H, J = 7.1 Hz, H-Ar), 7.97 (d, 2H, J = 7.3 Hz, H-Ar), 7.79-7.49 (m, 9H, H-Ar), 7.39 (dd, 2H, J = 7.7 Hz, H-Ar), 6.64 (d, 1H, J_{1,2} = 3.6 Hz, H-1), 5.87 (dd, 1H, J_{2,3} = J_{3,4} = 10.1 Hz, H-3), 5.54 (dd, 1H, J_{1,2} = 3.6 Hz, J_{2,3} = 10.1 Hz, H-2), 4.27 (dd, 1H, J_{3,4} = J_{4,5} = 10.1 Hz, H-4), 4.14-4.06 (m, 1H, H-5), 1.38 (d, 3H, J_{5,6} = 6.0 Hz, H-6).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.7, 165.2, 164.2 (CO-Bz), 134.8, 134.6, 134.5 (C_p-Ar), 130.1, 129.9, 129.7, 129.6, 129.5, 129.4 (C-Ar), 129.1, 129.0, 128.7 (C_i-Ar), 90.3 (C-1), 71.5 (C-3), 71.0 (C-2), 69.5 (C-5), 64.4 (C-4), 18.5 (C-6).

4-Azido-1,2,3-tri-O-benzoyl-4,6-dideoxy-β-D-glucopyranose (**15β**)

Under an inert gas, 3.86 g (8.1 mmol, 1.0 eq) of 6-deoxysugar **14β** were dissolved in 60 mL of absolute dichloromethane and 10 mL (124 mmol) of absolute pyridine and cooled to - 18 °C. 3.0 mL (24.3 mmol, 2.2 eq) of trifluoromethane sulfonic acid anhydride were added. After stirring for 60 minutes the solution was extracted with water, 2 x saturated potassium hydrogen carbonate and 2 N hydrochloric acid. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated and dried in high vacuum. Subsequently, 5.26 g (81 mmol, 10 eq) of sodium azide were added to the crude triflate. The mixture was suspended in 116 mL of absolute *N,N*-dimethylformamide and stirred for 30 minutes at room temperature. The organic layer was concentrated in vacuo (60 °C) and the residue was diluted with 300mL of dichloromethane. The solution was extracted with water, dried over MgSO₄ and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (5 x 16 cm, eluent: cyclohexane/ethyl acetate 12:1 → 9:1).

Yield: 3.41 g (6.8 mmol, 84 % over 2 steps)

R_f = 0.34 (cyclohexane/acetone 3:1)

[α]_D²⁶ = - 27.4 ° (c = 0.48, dichloromethane)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.96-7.87 (m, 4H, H-Ar), 7.79 (d, 2H, J = 7.3 Hz, H-Ar), 7.70-7.40 (m, 9H, H-Ar), 6.37 (d, 1H, J_{1,2} = 8.1 Hz, H-1), 5.94 (dd, 1H, J_{2,3} = J_{3,4} = 9.5 Hz, H-3), 5.54 (dd, 1H, J_{2,3} = 9.5 Hz, J_{1,2} = 8.1 Hz, H-2), 4.10-4.05 (m, 2H, H-4, H-5), 1.36 (d, 3H, J_{5,6} = 5.1 Hz, H-6).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.6, 165.4, 164.5 (CO-Bz), 134.5, 134.4, 134.3 (C_p-Ar), 129.9, 129.8, 129.6, 129.5, 129.4 (C-Ar), 129.1, 128.9, 128.6 (C_i-Ar), 92.5 (C-1), 73.4 (C-3), 72.0 (C-2), 71.3 (C-5), 64.9 (C-4), 18.4 (C-6).

4-Azido-2,3-di-O-benzoyl-4,6-dideoxy-D-glucopyranose (**10a**)

and 4-azido-2,3-di-O-benzoyl-4,6-dideoxy- α -D-glucopyranosyl-trichloroacetimidate (**10**)

from **15 α** :

1.32 g (2.63 mmol, 1.0 eq) of azide **15 α** were dissolved in 12 mL of absolute *N,N*-dimethylformamide. 2.42 g (26.3 mmol, 10 eq) of hydrazine acetate were added and the suspension was stirred at room temperature. After 5.5 h the reaction was stopped by adding 10 mL of acetone and stirring was continued until the remaining solid hydrazine acetate was completely dissolved. The solution was diluted with 200 mL of dichloromethane and extracted with water and saturated potassium hydrogen carbonate. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo and purified by flash chromatography (3.5 x 28 cm, eluent: cyclohexane/acetone 10:1).

yield of hemiacetal **10a**: 886 mg (2.23 mmol, 84.8 %, α/β = 2:1)

R_f = 0.26 β -anomer, 0.34 α -anomer (cyclohexane/acetone 2:1)

from **15 β** :

3.40 g (6.8 mmol, 1.0 eq) of azide **15 β** were dissolved in 18 mL of absolute *N,N*-dimethylformamide. 4.70 g (50.9 mmol, 7.5 eq) of hydrazine acetate were added and the suspension was stirred at room temperature. After 3.5 h the reaction was stopped by adding 20 mL of acetone and stirring was continued until the remaining solid hydrazine acetate was completely dissolved. The solution was diluted in 400 mL of dichloromethane and extracted with water and saturated potassium hydrogen carbonate. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo and purified by flash chromatography (5 x 20 cm, eluent: cyclohexane/acetone 10:1).

yield of hemiacetal **10a**: 2.38 g (6.0 mmol, 88.6 %, α/β 2:1)

¹H-NMR (270 MHz, [D₆]-DMSO): δ = 7.96 (d, 2H, *J* = 7.1 Hz, H-Ar), 7.90 (d, 1H, *J* = 7.1 Hz, H-Ar), 7.83 (d, 3H, *J* = 7.1 Hz, H-Ar), 7.66-7.57 (m, 3H, H-Ar), 7.51-7.43 (m, 6H, H-Ar), 7.28 (d, 1H, *J*_{OH-1 β ,1} = 6.2 Hz, OH-1 β), 7.24 (d, 1H, *J*_{OH-1 α ,1} = 3.9 Hz, OH-1 α), 5.75 (dd, 1H, *J*_{2 α ,3 α} = *J*_{3 α ,4 α} = 9.6 Hz, H-3 α), 5.61 (dd, 1H, *J*_{2 β ,3 β} = *J*_{3 β ,4 β} = 9.5 Hz, H-3 β), 5.39 (dd, 1H, *J*_{OH-1 α ,1} = *J*_{1,2} = 3.6 Hz, H-1 α), 5.11 (dd, 1H, *J*_{1,2} = 3.6 Hz, *J*_{2,3} = 9.6 Hz, H-2 α), 5.12 (dd, 1H, *J*_{1,2} = 7.6 Hz, *J*_{2,3} = 9.6 Hz, H-2 β), 5.02 (dd, 1H, *J*_{1,2} = 7.6 Hz, *J*_{OH-1 β ,1} = 6.2 Hz, H-1 β), 4.01-3.83 (m, 3H, H-5 α , H-4 α , H-4 β), 3.80-3.70 (m, 1H, H-5 β), 1.32 (d, 3H, *J*_{5,6} = 6.0 Hz, H-6 β), 1.31 (d, 3H, *J*_{5,6} = 5.6 Hz, H-6 α).

¹³C-NMR (68 MHz, [D₆]-DMSO): δ = 165.9, 165.6 (CO-Bz α), 165.7, 165.3 (CO-Bz β), 134.3 (C_p-Ar α), 134.1 (C_p-Ar β), 129.8, 129.7, 129.4 (C-Ar α), 129.6, 129.3, 129.2 (C-Ar β), 94.3 (C-1 β), 89.7 (C-1 α), 74.5 (C-2 β), 74.3 (C-3 β), 73.0 (C-2 α), 71.5 (C-3 α), 69.8 (C-5 β), 65.5 (C-4 α , C-5 α), 65.3 (C-4 β), 18.5 (C-6 α , C-6 β).

1.08 g (2.7 mmol, 1.0 eq) of hemiacetal **10a** was dissolved in 25 mL of absolute dichloromethane and cooled to 0 °C. 2.8 mL (27.9 mmol, 10.3 eq) of trichloroacetonitrile were added and the reaction was started by addition of 110 μ L (0.74 mmol, 0.35 eq) of DBU. After 2 h the solvent was concentrated in vacuo (20 °C) and the residue purified by flash chromatography (3 x 20 cm, eluent: cyclohexane/acetone 15:1).

yield of imidate **10**: 1.38 g (2.55 mmol, 94.7 %)

R_f = 0.47 (cyclohexane/acetone 3:1)

$[\alpha]_D^{26} = +55.7^\circ$ (c = 0.52, dichloromethane)

C₂₂H₁₉Cl₃N₄O₆ (541.77)

ESI/MS: M_{cal.} = 540.04

M_{found} = 562.92 [M+Na]⁺

$^1\text{H-NMR}$ (270 MHz, $[\text{D}_6]$ -DMSO): δ = 9.91 (s, 1H, NH), 7.96 (d, 2H, J = 7.5 Hz, H-Ar), 7.82 (d, 2H, J = 7.5 Hz, H-Ar), 7.70-7.57 (m, 2H, H-Ar), 7.55-7.42 (m, 4H, H-Ar), 6.60 (bs, 1H, H-1), 5.81 (dd, 1H, $J_{2,3} = J_{3,4} = 10.0$ Hz, H-3), 5.51 (dd, 1H, $J_{1,2} = 3.0$ Hz, $J_{2,3} = 10.0$ Hz, H-2), 4.23 (dd, 1H, $J_{3,4} = J_{4,5} = 10.0$ Hz, H-4), 3.94 (dq, 1H, $J_{4,5} = 10.0$ Hz, $J_{5,6} = 6.2$ Hz, H-5), 1.38 (d, 3H, $J_{5,6} = 6.2$ Hz, H-6).

$^{13}\text{C-NMR}$ (68 MHz, $[\text{D}_6]$ -DMSO): δ = 165.6, 165.2 (CO-Bz), 134.5, 134.3 (C_p -Ar), 129.9, 129.8, 129.4 (C-Ar), 129.0, 128.7 (C_o -Ar), 94.3 (C_q - CCl_3), 92.9 (C-1), 71.2 (C-3), 70.9 (C-2), 69.4 (C-5), 64.3 (C-4), 18.2 (C-6).

5-Chloro-2-carboxymethylamino-benzoic acid (**17**)

10.0 g (58.3 mmol, 1.0 eq) of 4-chloroanthranilic acid **16** were dissolved in 50 mL of 1 N sodium carbonate and the pH of the solution was adjusted to 8.5 with 1 N sodium carbonate. 7.2 g (76.2 mmol, 1.3 eq) of chloroacetic acid were dissolved in 15 mL of 1 N sodium carbonate and adjusted to pH 3 with 1 N sodium carbonate. Both solutions were united and adjusted to pH 9 with 1 N sodium carbonate. The reaction was stirred under reflux and a pH of 9 was kept by periodical addition of 1 N sodium carbonate. After 6 h the reaction was cooled to room temperature and the solution was acidified to pH 4 with 2N hydrochloric acid. The resulting precipitate was left for 12 h at 3 °C and filtered. The precipitate was briefly washed with a small amount of water and dried by lyophilization. The product was used for the next reaction without any purification. An aliquot was purified by flash chromatography for analytical purposes (eluent: toluene/ethyl acetate containing 1 % HOAc 7:1 \rightarrow 5:1).

yield of **17**: 8.77 g (38.2 mmol, 65.5 %)

R_f = 0.34 (cyclohexane/ethyl acetate 1:1 with 1.0 % HOAc (98 %))

$\text{C}_9\text{H}_8\text{ClNO}_4$ (229.62)

EI/MS (70 eV): m/z (%) = 231 (9) [^{37}Cl]M $^+$, 229 (27) [^{35}Cl]M $^+$, 186 (12) [^{37}Cl]M-COOH $^+$, 184 (45) [^{35}Cl]M-COOH $^+$

$^1\text{H-NMR}$ (270 MHz, $[\text{D}_6]$ -DMSO): δ = 13.06 (bs, 2H, COOH), 8.12 (bs, 1H, NH), 7.73 (d, 1H, $J_{4,6} = 2.6$ Hz, H-6), 7.38 (dd, 1H, $J_{3,4} = 9.0$ Hz, $J_{4,6} = 2.6$ Hz, H-4), 6.64 (d, 1H, $J_{3,4} = 9.0$ Hz, H-3), 4.00 (s, 2H, H-7).

$^{13}\text{C-NMR}$ (68 MHz, $[\text{D}_6]$ -DMSO): δ = 172.0, 169.1 (COOH), 149.3 (C_q -NH), 134.5 (C-4), 131.0 (C-6), 118.6 (C_q -Cl), 114.2 (C-3), 112.1 (C_q -COOH), 44.8 (C-7).

3-Acetoxy-5-chloroindol (**18**) via 1,3-diacetyl-5-chloroindol (**17a**)

8.77 g (38.2 mmol, 1.0 eq) of **17** were stirred in 88 mL (0.9 mmol) of acetic anhydride with 9.39 g (0.1 mmol) of sodium acetate at 85 °C. After 3h the reaction was cooled to room temperature and the precipitate was removed by filtration. The precipitate was washed with 300 mL of ethyl acetate and the resulting filtrate was extracted with water (3 x) and saturated potassium hydrogen carbonate (2 x). The organic layer was dried (MgSO_4) and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (5 x 20 cm, eluent: cyclohexane/ethyl acetate 7:1 \rightarrow 5:1).

yield of 1,3-diacetyl-5-chloroindol (**17a**):

6.09 g (24.2 mmol, 63 %)

R_f = 0.64 (toluene/ethyl acetate 5:1 with 1.0 % HOAc (98 %))

$\text{C}_{12}\text{H}_{10}\text{ClNO}_3$ (251.67)

EI/MS (70 eV): m/z (%) = 253 (7) [^{37}Cl]M $^+$, 251 (20) [^{35}Cl]M $^+$, 211 (12) [^{37}Cl]M-Ac $^+$, 209 (39) [^{35}Cl]M-Ac $^+$

$^1\text{H-NMR}$ (360 MHz, $[\text{D}_6]$ -DMSO): δ = 8.33 (d, 1H, $J_{6,7} = 8.5$ Hz, H-7), 7.96 (s, 1H, H-2), 7.63 (d, 1H, $J_{4,6} = 2.1$ Hz, H-4), 7.40 (dd, 1H, $J_{4,6} = 2.1$ Hz, $J_{6,7} = 8.5$ Hz, H-6), 2.60 (s, 3H, NAc), 2.37 (s, 3H, OAc).

^{13}C -NMR (90 MHz, $[\text{D}_6]$ -DMSO): δ = 169.5, 168.4 (CO-Ac), 132.6 ($\text{C}_{\text{q}}\text{-7a}$), 131.1 ($\text{C}_{\text{q}}\text{-3}$), 128.0 ($\text{C}_{\text{q}}\text{-5}$), 125.5 (C-7), 125.2 ($\text{C}_{\text{q}}\text{-3a}$), 117.6, 117.5 (C-2, C-4, C-6), 23.6 (NAc), 20.5 (OAc).

Under an inert gas, 1.0 g (4.0 mmol, 1.0 eq) of 1,3-diacetyl-5-chloroindole (**17a**) was dissolved in 50 mL of sodium hydroxide (5 %) and stirred under reflux. After 30 minutes the reaction was cooled to 0 °C and 7 mL (74 mmol, 18.5 eq) of acetic anhydride were added dropwise. The reaction was stirred for 30 minutes. The resulting white-blue precipitate was filtered and washed with water. The precipitate was dissolved in dichloromethane and the insoluble blue material was removed by filtration. The filtrate was concentrated and dried in high vacuo.

yield of **18**: 0.73 g (3.48 mmol, 87.1 %)

R_f = 0.40 (toluene/ethyl acetate 7:1)

$\text{C}_{10}\text{H}_8\text{ClNO}_2$ (209.63)

EI/MS (70 eV): m/z (%) = 211 (7) [^{37}Cl]M $^+$, 209 (22) [^{35}Cl]M $^+$

^1H -NMR (270 MHz, $[\text{D}_6]$ -DMSO): δ = 11.18 (bs, 1H, NH), 7.44 (d, 1H, $J_{4,6}$ = 1.7 Hz, H-4), 7.41 (s, 1H, H-2), 7.38 (d, 1H, $J_{6,7}$ = 8.8 Hz, H-7), 7.11 (dd, 1H, $J_{4,6}$ = 1.7 Hz, $J_{6,7}$ = 8.8 Hz, H-6), 2.31 (s, 3H, Ac).

^{13}C -NMR (68 MHz, $[\text{D}_6]$ -DMSO): δ = 169.3 (CO-Ac), 132.3 ($\text{C}_{\text{q}}\text{-7a}$), 129.1 ($\text{C}_{\text{q}}\text{-3}$), 124.2 ($\text{C}_{\text{q}}\text{-5}$), 122.3 (C-6), 121.3 ($\text{C}_{\text{q}}\text{-3a}$), 117.1, 116.9 (C-2, C-4), 114.1 (C-7), 21.1 (Ac).

5,5'-Dichloroindigo (**19**)

294 mg (1.40 mmol, 0.5 eq) **17** was dissolved in 15 mL of methanol and 1 mL of 1N sodium hydroxide was added. Immediately, compressed air was bubbled through the solution. After 15 minutes the blue precipitate was filtered, washed with 250 mL of water, 200 mL of methanol and 200 mL of diethyl ether. The residue was dried in a compartment dryer at 70 °C.

yield: 168 mg (0.51 mmol, 72.9 %), blue solid

$\text{C}_{16}\text{H}_8\text{Cl}_2\text{N}_2\text{O}_2$ (331.15)

EI/MS (70 eV): m/z (%) = 332 (67) [^{37}Cl]M $^+$, 330 (100) [^{35}Cl]M $^+$

For solution NMR 5,5'-dichloroindigo **19** was converted to its vat indigo form **19v** by heating a mixture of **19**, sodium dithionite and potassium hydroxide in D_2O .

NMR analysis of vat indigo **19v**:

^1H -NMR (270 MHz, D_2O): δ = 7.53 (d, 2H, $J_{4,6}$ = 1.9 Hz, H-4), 7.26 (d, 2H, $J_{7,6}$ = 8.6 Hz, H-7), 6.99 (dd, 2H, $J_{4,6}$ = 1.9 Hz, $J_{6,7}$ = 8.6 Hz, H-6).

^{13}C -NMR (68 MHz, D_2O): δ = 138.9 ($\text{C}_{\text{q}}\text{-7a}$), 132.7 ($\text{C}_{\text{q}}\text{-3a}$), 124.3 ($\text{C}_{\text{q}}\text{-2}$), 122.8 ($\text{C}_{\text{q}}\text{-5}$), 120.3 (C-6), 116.5 (C-4), 116.3 ($\text{C}_{\text{q}}\text{-3}$), 112.1 (C-7).

N-Benzyl-5,5'-dichloroindigo (**11**)

2.06 g (6.21 mmol, 1.0 eq) of 5,5'-dichloroindigo **19** were suspended in 50 mL of absolute *N,N*-dimethylformamide, 273 mg (6.83 mmol, 1.1 eq) of NaH (60 % in oil suspension) were added and the suspension was stirred at room temperature. After 30 minutes 273 mg (6.83 mmol, 1.1 eq) of NaH (60 % in oil suspension) were added. After 2 h 0.9 mL (7.57 mmol, 1.2 eq) of benzylbromide were added dropwise. The reaction was stirred for 1 h at room temperature and was subsequently concentrated in vacuo (60 °C). The oily

residue was dissolved in 150 mL of dichloromethane and extracted with water (3 x). The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (6 x 16 cm, eluent: toluene).

Yield: 1.53 g (3.6 mmol, 59 %), blue solid

R_f = 0.68 (toluene/ethyl acetate 15:1)

UV/VIS: λ_{max} (lgε) = 642 nm (4.11) in dichloromethane

C₂₃H₁₄Cl₂N₂O₂ (421.27)

EI/MS (70 eV): m/z (%) = 422 (67) [(³⁷Cl)M]⁺, 420 (100) [(³⁵Cl)M]⁺, 331 (40) [(³⁷Cl)M-Benzyl]⁺, 329 (58) [(³⁵Cl)M-Benzyl]⁺, 91 (92) [Benzyl]⁺

¹H-NMR (360 MHz, [D₆]-DMSO): δ = 7.69 (d, 1H, J_{4,6} = 2.0 Hz, H-4), 7.62 (dd, 1H, J_{4,6} = 2.0 Hz, J_{6,7} = 8.7 Hz, H-6), 7.55 (d, 1H, J_{4',6'} = 2.0 Hz, H-4'), 7.54 (d, 1H, J_{4',6'} = 2.0 Hz, J_{6',7'} = 8.7 Hz, H-6'), 7.39 (d, 2H, J = 8.7 Hz, H-7, H-7'), 7.22-7.18 (m, 3H, H_p-Ar, H_m-Ar), 7.08 (d, 2H, J = 7.2 Hz, H_o-Ar), 5.74 (s, 2H, CH₂).

¹³C-NMR (90 MHz, [D₆]-DMSO): δ = 186.8 (C_q-3), 184.4 (C_q-3'), 151.1 (C_q-7a'), 150.4 (C_q-7a), 137.3 (C_i-Ar), 135.6, 135.4 (C-6, C-6'), 128.6 (C_o-Ar), 127.3 (C_p-Ar), 126.7 (C_m-Ar), 125.6 (C_q-5), 124.7 (C_q-5'), 124.6 (C_q-2), 123.3 (C_q-2'), 123.1 (C-4'), 122.7 (C-4), 121.7 (C_q-3a'), 120.1 (C_q-3a), 115.1 (C-7), 114.1 (C-7'), 50.1 (CH₂).

N-[(4''-Azido-2'',3''-di-*O*-benzoyl-4'',6''-dideoxy-β-*D*-glucopyranosyl)-*N'*-benzyl]-5,5'-dichloroindigo (**20**)

213 mg (0.39 mmol, 1.0 eq) of imidate **10**, 165 mg (0.39 mmol, 1.0 eq) of *N*-benzyl-5,5'-dichloroindigo **11** and 400 mg molecular sieves 4 Å were suspended in 22 mL of absolute dichloromethane, stirred for 30 minutes at room temperature followed by cooling to -18 °C. The reaction was started by adding 23 μL (0.127 mmol, 0.33 eq) of TMSOTf. The reaction was stirred for 2 h at -18 °C and slowly warmed to room temperature over 12 h. The reaction was stopped by adding 10 mL of saturated potassium hydrogen carbonate. After 10 minutes the suspension was filtered through Celite and washed with dichloromethane. The filtrate was extracted with water, saturated potassium hydrogen carbonate and brine. The organic layer was dried (MgSO₄) and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (5 x 18 cm, eluent: cyclohexane/ethyl acetate 30:1 → 20:1 → 15:1 → 10:1). 89 mg (0.19 mmol, 54 %) of *N*-benzyl-5,5'-dichloroindigo **11** were recovered.

yield: 139 mg **20** (0.17 mmol, 44.6 %), turquoise solid

R_f = 0.33 (cyclohexane/ethyl acetate 4:1)

UV/VIS: λ_{max} (lgε) = 646 nm (4.04) in dichloromethane

[α]_D²³ = + 5378.2 ° (c = 5.8 μg/mL, dichloromethane)

C₄₃H₃₁Cl₂N₅O₇ (800.64)

ESI/MS: M_{cal.} = 799.16

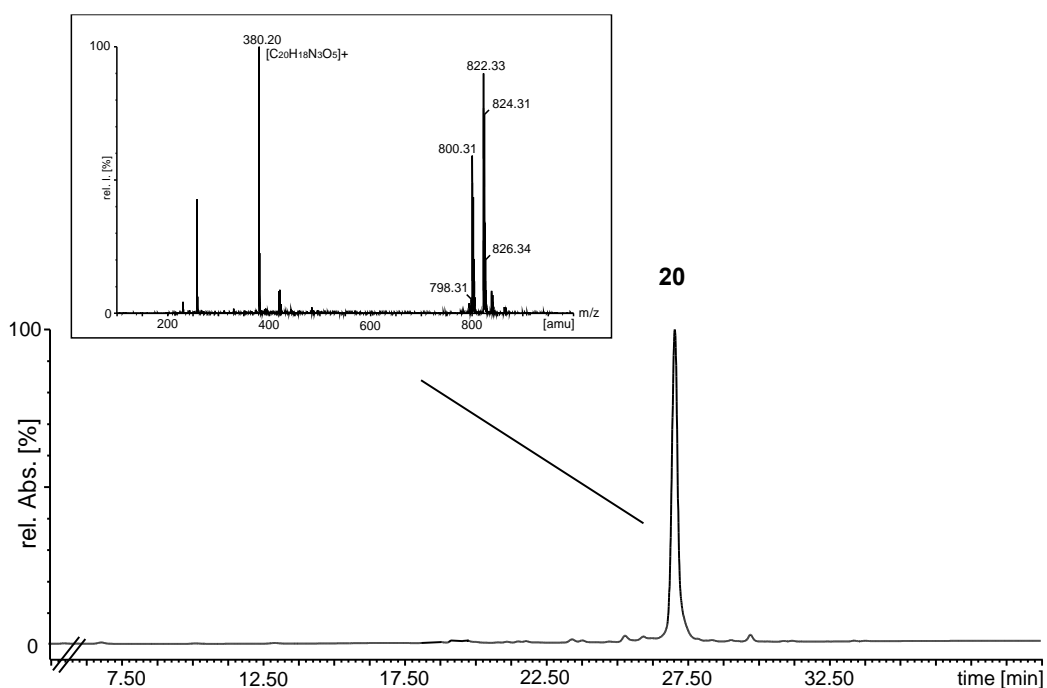
M_{found} = 800.39 [(^{35/35}Cl)M+H]⁺, 802.39 [(^{35/37}Cl)M+H]⁺, 804.39 [(^{37/37}Cl)M+H]⁺, 822.34 [(^{35/35}Cl)M+Na]⁺, 824.37 [(^{35/37}Cl)M+Na]⁺, 826.35 [(^{37/37}Cl)M+Na]⁺, 1621.63 [2M+Na]⁺, 1623.73 [2M+Na]⁺, 1625.63 [2M+Na]⁺, 1627.73 [2M+Na]⁺

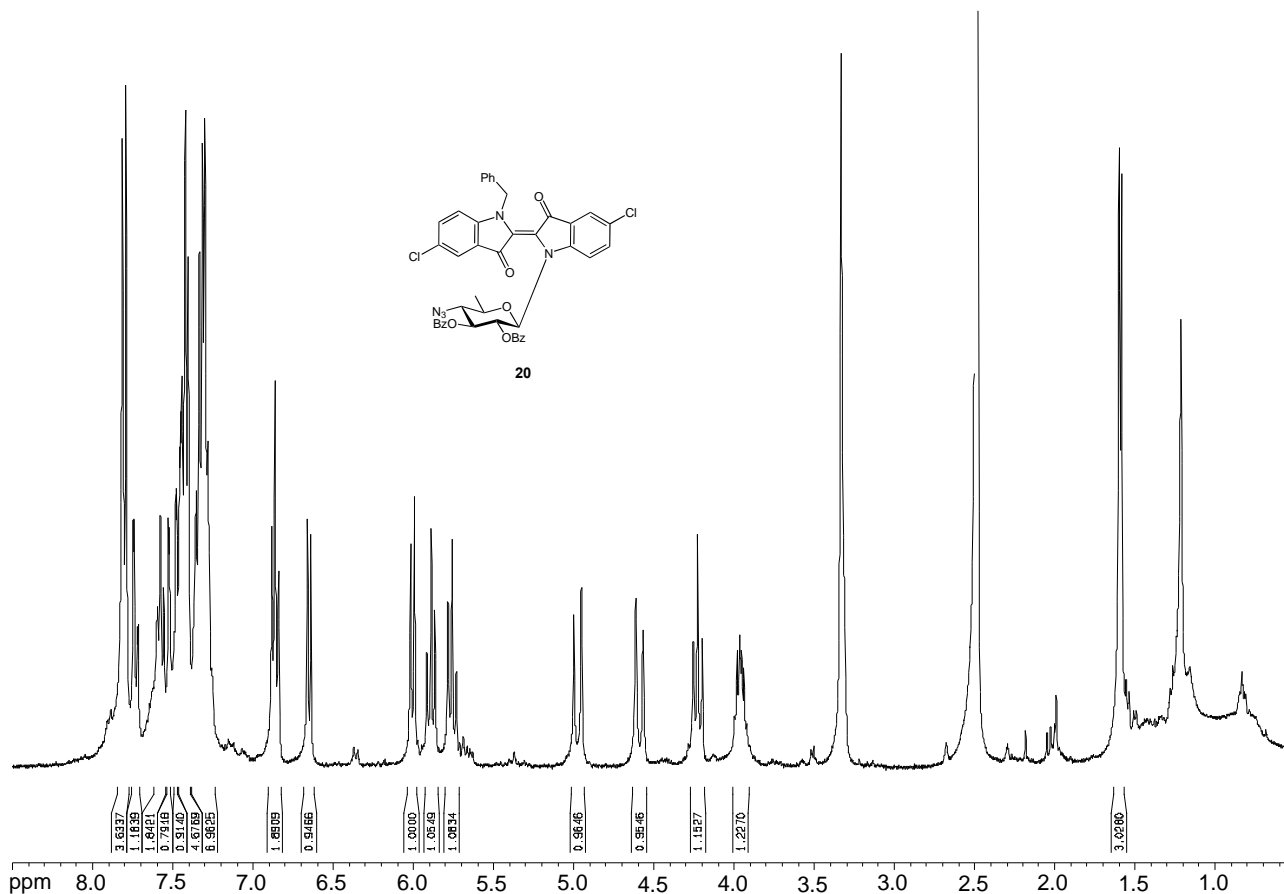
¹H-NMR (360 MHz, [D₆]-DMSO): δ = 7.82-7.78 (m, 3H, H-7, H-4, H-Ar), 7.73 (dd, 1H, J_{4,6} = 2.1 Hz, J_{6,7} = 8.7 Hz, H-6), 7.57 (dd, 1H, J = 7.5 Hz, H-Ar), 7.52 (d, 1H, J_{4',6'} = 2.1 Hz, H-4'), 7.47 (dd, 1H, J_{4',6'} = 2.1 Hz, J_{6',7'} = 8.6 Hz, H-6'), 7.45-7.40 (m, 5H, H-Ar), 7.38-7.27 (m, 6H, H-Ar), 6.86 (dd, 2H, J = 7.8 Hz, H-Ar), 6.64 (d, 1H, J_{6',7'} = 8.6 Hz, H-7'), 6.00 (d, 1H, J_{1'',2''} = 9.1 Hz, H-1''), 5.89 (dd, 1H, J_{2'',3''} = J_{3'',4''} = 9.3 Hz, H-3''), 5.76 (dd, 1H, J_{1'',2''} = J_{2'',3''} = 9.3 Hz, H-

2''), 4.97 (d, 1H, $J_{\text{gem}} = 16.9$ Hz, H-8'), 4.59 (d, 1H, $J_{\text{gem}} = 16.9$ Hz, H-8'), 4.23 (dd, 1H, $J_{3'',4''} = J_{4'',5''} = 9.2$ Hz, H-4''), 4.05-3.90 (m, 1H, H-5''), 1.59 (d, 3H, $J_{5'',6''} = 6.0$ Hz, H-6'').

^{13}C -NMR (90 MHz, $[\text{D}_6]$ -DMSO): $\delta = 185.6$ (C_q -3), 185.3 (C_q -3'), 164.9, 164.0 (CO-Bz), 150.9 (C_q -7a'), 148.5 (C_q -7a), 136.2 (C_r -9'), 135.8 (C-6'), 135.0 (C-6), 134.6, 133.7, 133.8 (C_p -Ar), 129.5, 129.1, 128.8 (C-Ar), 128.6, 128.4 (C_r -Ar), 128.3 (C-Ar), 127.6 (C_q -5), 127.1 (C_q -5'), 127.0, 126.3 (C-Ar), 125.1 (C_q -2), 124.0 (C_q -2'), 123.6 (C-4), 122.6 (C-4'), 122.7 (C_q -3a), 120.2 (C_q -3a'), 117.9 (C-7), 115.2 (C-7'), 85.3 (C-1''), 74.4 (C-3''), 73.4 (C-5''), 69.1 (C-3''), 65.3 (C-4''), 54.5 (CH_2 -8'), 18.2 (C-6'').

a) LC-MS of **20**:



b) $^1\text{H-NMR}$ of **20** (360 MHz):Figure S3. LC-MS (a) and NMR (b) of **20**.*N*-(4''-Azido-2'',3''-di-*O*-benzoyl-4'',6''-dideoxy- β -*D*-glucopyranosyl)-5,5'-dichloroindigo (**21**)

184 mg (0.23 mmol, 1.0 eq) of **20** were dissolved in 52 mL of toluene/acetic acid (1:1) and stirred in an open flask for 28 h at 50 °C. The solution was diluted with 200 mL of dichloromethane, extracted with water and saturated potassium hydrogen carbonate. The organic layer was dried (MgSO_4) and filtered. The filtrate was concentrated in vacuo and the residue purified by flash chromatography (5 x 18 cm, eluent: cyclohexane/ethyl acetate 20:1 \rightarrow 15:1).

yield: 98 mg (0.14 mmol, 60 %), blue solid

R_f = 0.53 (toluene)

UV/VIS: λ_{max} ($\lg \epsilon$) = 618 nm (3.35) in dichloromethane

$[\alpha]_D^{23} = +4929.0^\circ$ ($c = 5.1 \mu\text{g/mL}$, dichloromethane)

$\text{C}_{36}\text{H}_{25}\text{Cl}_2\text{N}_5\text{O}_7$ (710.52)

ESI/MS: $M_{\text{cal.}} = 709.11$

$M_{\text{found}} = 709.99$ [$(^{35}/^{35}\text{Cl})\text{M}+\text{H}$] $^+$, 712.01 [$(^{35}/^{37}\text{Cl})\text{M}+\text{H}$] $^+$, 713.99 [$(^{37}/^{37}\text{Cl})\text{M}+\text{H}$] $^+$, 731.99 [$(^{35}/^{35}\text{Cl})\text{M}+\text{Na}$] $^+$, 733.95 [$(^{35}/^{37}\text{Cl})\text{M}+\text{Na}$] $^+$, 735.96 [$(^{37}/^{37}\text{Cl})\text{M}+\text{Na}$] $^+$, 1440.97 [2M+Na] $^+$, 1443.01 [2M+Na] $^+$, 1445.06 [2M+Na] $^+$, 1447.09 [2M+Na] $^+$

$^1\text{H NMR}$ (270 MHz, $[\text{D}_6]$ -DMSO): $\delta = 10.91$ (s, 1H, NH), 7.84-7.80 (m, 3H, H-7, H_o-Ar), 7.77-7.71 (m, 2H, H-4, H-6), 7.64-7.56 (m, 3H, H-4', H-6', H_p-Ar), 7.53-7.43 (m, 5H, H_o-Ar, H_p-Ar), 7.40 (d, 2H, $J = 8.6$ Hz, H-7'), 7.29 (dd,

^1H , $J = 7.7$ Hz, $H_{m\text{-Ar}}$), 6.38 (d, 1H, $J_{1'',2''} = 8.6$ Hz, H-1''), 5.75-5.65 (m, 2H, H-2'', H-3''), 4.28 (dd, 1H, $J_{3'',4''} = J_{4'',5''} = 9.6$ Hz, H-4''), 4.02-3.90 (m, 1H, H-5''), 1.55 (d, 3H, $J_{5'',6''} = 6.0$ Hz, H-6'').

^{13}C NMR (90 MHz, $[\text{D}_6]\text{-DMSO}$): $\delta = 186.1$ ($\text{C}_{\text{q-3}}$), 185.8 ($\text{C}_{\text{q-3}'}$), 165.5, 164.6 (CO-Bz), 151.4 ($\text{C}_{\text{q-7a}'}$), 149.1 ($\text{C}_{\text{q-7a}}$), 136.4 ($\text{C-6}'$), 135.6 (C-6), 134.4, 134.3 ($\text{C}_{\text{p-Ar}}$), 129.8, 129.6, 129.5, 129.4, 129.2 (C-Ar), 129.0 ($\text{C}_i\text{-Ar}$), 128.2 ($\text{C}_{\text{q-5}}$), 127.7 ($\text{C}_{\text{q-5}'}$), 125.9 ($\text{C}_{\text{q-2}}$), 125.6 ($\text{C}_{\text{q-2}'}$), 124.6 ($\text{C}_{\text{q-3a}}$), 124.2 (C-4), 123.2 ($\text{C-4}'$), 120.7 ($\text{C}_{\text{q-3a}'}$), 118.5 (C-7), 115.8 ($\text{C-7}'$), 86.1 (C-1''), 75.0, 69.7 (C-2'' , C-3''), 74.4 (C-5''), 64.9 (C-4''), 18.7 (C-6'').

O'-[(4''-Azido-2'',3''-di-*O*-benzoyl-4'',6''-dideoxy- β -*D*-glucopyranosyl)-*N'*-benzyl]-5,5'-dichlorindigo (**20a**) and *O'*-[(4''-azido-2'',3''-di-*O*-benzoyl-4'',6''-dideoxy- α -*D*-glucopyranosyl)-*N'*-benzyl]-5,5'-dichlorindigo (**20b**)

100 mg (0.185 mmol, 1.0 eq) of imidate **10**, 78 mg (0.185 mmol, 1.0 eq) of *N*-benzyl-5,5'-dichloroindigo **11** and 200 mg of molecular sieves 4 Å were suspended in 11 mL of absolute dichloromethane, stirred for 30 minutes at room temperature and subsequently cooled to -18 °C. The reaction was started by the addition of 11 μL (0.062 mmol, 0.33 eq) of TMSOTf. After stirring for 2 h at -18 °C the reaction was quenched by adding 200 μL of 0.5 % Et_3N in dichloromethane. On TLC in cyclohexane/ethyl acetate 4:1 three colored spots were obtained: $R_f = 0.64$ (blue), $R_f = 0.51$ (green), $R_f = 0.44$ (red). The reaction mixture was filtered over Celite and the filtrate was extracted with saturated sodium hydrogen carbonate solution. The organic layer was dried (MgSO_4) and filtered. After adding 7 mL of 0.5 % Et_3N in dichloromethane the filtrate was concentrated in vacuo and the residue purified by flash chromatography (2.5 x 14 cm, eluent: cyclohexane/ethyl acetate 30:1 containing 0.5 % Et_3N). Four fractions were obtained:

Fraction 1: indigo **11** (blue) $R_f = 0.64$

Fraction 2: *N*-glycoside **20** (green) $R_f = 0.51$

Fraction 3: *N*-glycoside **20** and α -*O*-glycoside **20b** (red) $R_f = 0.48$

Fraction 4: β -*O*-glycoside **20a** (red) $R_f = 0.44$

R_f values in cyclohexane/ethyl acetate 4:1

β -*O*-glycoside (**20a**)

yield: 6.6 mg (4.5 %)

UV/VIS: λ_{max} ($\lg \epsilon$) = 488 nm (2.41) in dichloromethane

$[\alpha]_{\text{D}}^{23} = +1579$ ° ($c = 9.5$ $\mu\text{g/mL}$, dichloromethane)

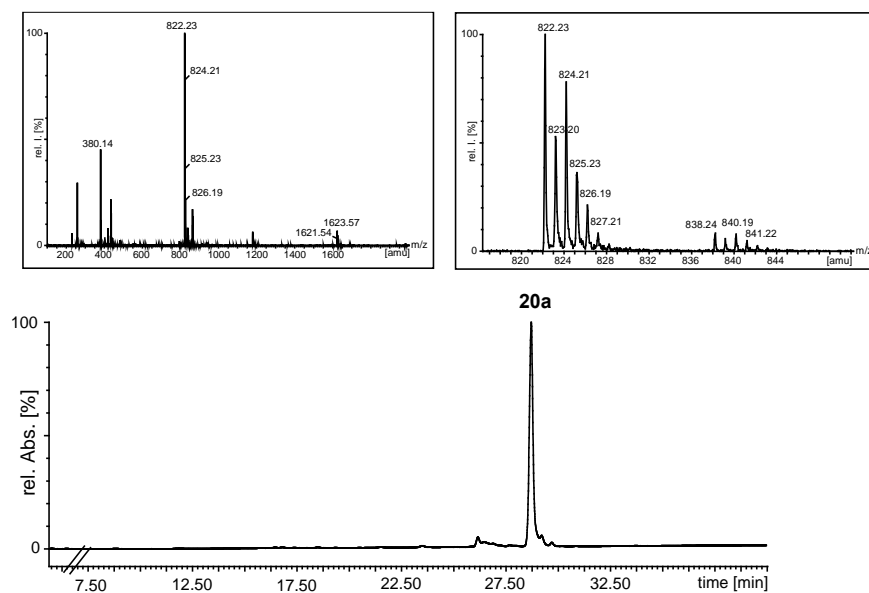
$\text{C}_{43}\text{H}_{31}\text{Cl}_2\text{N}_5\text{O}_7$ (800.64)

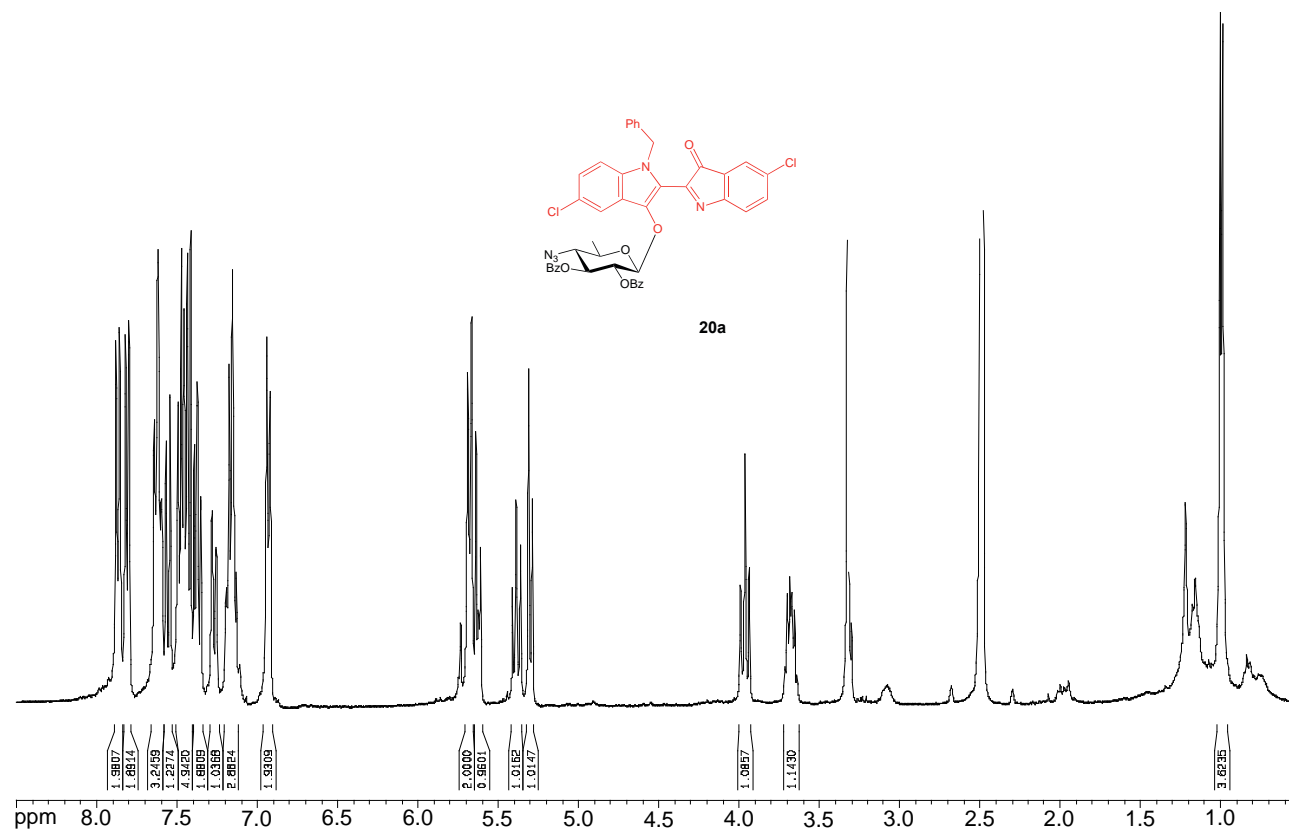
ESI/MS: $M_{\text{cal.}} = 799.16$

$M_{\text{found}} = 822.37$ [$^{35/35}\text{Cl}$]M+Na] $^+$, 824.36 [$^{35/37}\text{Cl}$]M+Na] $^+$, 826.34 [$^{37/37}\text{Cl}$]M+Na] $^+$, 838.34 [$^{35/35}\text{Cl}$]M+K] $^+$, 840.34 [$^{35/37}\text{Cl}$]M+K] $^+$, 842.39 [$^{37/37}\text{Cl}$]M+K] $^+$

^1H -NMR (360 MHz, $[\text{D}_6]\text{-DMSO}$): $\delta = 7.86$ (d, 2H, $J = 7.4$ Hz, H-Ar), 7.80 (d, 2H, $J = 7.5$ Hz, H-Ar), 7.63-7.59 (m, 3 H, H-6, H-Ar), 7.55 (d, 1H, $J_{6',7'} = 9.0$ Hz, H-7'), 7.49-7.42 (m, 4H, H-Ar), 7.41 (bs, 1H, H-4'), 7.38 (d, 1H, $J_{6,7} = 7.1$ Hz, H-7), 7.35 (d, 1H, $J_{4,6} = 1.7$ Hz, H-4), 7.26 (dd, 1H, $J_{6',7'} = 8.9$ Hz, $J_{4',6'} = 1.7$ Hz, H-6'), 7.19-7.11 (m, 3H, H-benzyl), 6.93 (d, 2H, $J = 7.0$ Hz, H-benzyl), 5.71 (d, 1H, $J_{\text{gem}} = 16.2$ Hz, H-8'a), 5.64 (d, 1H, $J_{\text{gem}} = 16.2$ Hz, H-8'b), 5.62 (dd, 1H, $J_{2'',3''} = J_{3'',4''} = 9.3$ Hz, H-3''), 5.38 (dd, 1H, $J_{1'',2''} = J_{2'',3''} = 9.3$ Hz, H-2''), 5.29 (d, 1H, $J_{1'',2''} = 8.0$ Hz, H-1''), 3.96 (dd, 1H, $J_{3'',4''} = J_{4'',5''} = 9.3$ Hz, H-4''), 3.67 (td, 1H, $J_{4'',5''} = 9.3$, $J_{5'',6''} = 6.1$ Hz, H-5''), 0.99 (d, 3H, $J_{5'',6''} = 6.1$ Hz, H-6'').

^{13}C -NMR (90 MHz, $[\text{D}_6]$ -DMSO): δ = 187.8 (C_q -3), 164.9, 164.4 (CO -Bz), 157.8 (C_q -3a), 157.4 (C_q -7a), 139.3 (C_q -3'), 137.7 (C_ipso -9'), 135.7 (C-6), 135.3 (C_q -7a'), 133.9, 133.8 (C_p -Ar), 133.1 (C_q -2), 129.3, 129.0, 128.9 (C-Ar), 128.5 (C_m -benzyl), 128.4, 128.3 (C_q -Ar), 127.2 (C_p -benzyl), 126.4 (C_o -benzyl), 125.6 (C-6'), 125.3 (C_q -3a'), 124.5 (C-4), 123.3 (C-7), 123.0 (C_q -5), 120.9 (C_q -2'), 120.5 (C_q -5'), 117.8 (C-4'), 113.4 (C-7'), 101.4 (C-1''), 73.4 (C-3''), 72.0 (C-2''), 69.7 (C-5''), 63.9 (C-4''), 47.2 (CH_2 -8'), 17.2 (C-6'').

a) LC-MS of **20a**:

b) $^1\text{H-NMR}$ of **20a** (360 MHz):Figure S4: LC-MS (a) and NMR (b) of **20a**. *α -O-glycoside (20b)*

The α -O-glycoside **20b** was isolated from fraction 3 by flash chromatography (5 x 17 cm, eluent: cyclohexane/ethyl acetate 30:1+0.5 % $\text{NEt}_3 \rightarrow 25:1+0.5$ % NEt_3).

yield: 2.4 mg (1.6 %)

$\text{C}_{43}\text{H}_{31}\text{Cl}_2\text{N}_5\text{O}_7$ (800.64)

ESI/MS: $M_{\text{cal.}} = 799.16$

$M_{\text{found}} = 800.93$ $[(^{35}/^{35}\text{Cl})\text{M}+\text{H}]^+$, 802.93 $[(^{35}/^{37}\text{Cl})\text{M}+\text{H}]^+$, 804.93 $[(^{37}/^{37}\text{Cl})\text{M}+\text{H}]^+$, 822.95 $[(^{35}/^{35}\text{Cl})\text{M}+\text{Na}]^+$, 824.94 $[(^{35}/^{37}\text{Cl})\text{M}+\text{Na}]^+$, 826.97 $[(^{37}/^{37}\text{Cl})\text{M}+\text{Na}]^+$, 838.34 $[(^{35}/^{35}\text{Cl})\text{M}+\text{K}]^+$, 840.34 $[(^{35}/^{37}\text{Cl})\text{M}+\text{K}]^+$, 842.39 $[(^{37}/^{37}\text{Cl})\text{M}+\text{K}]^+$

$^1\text{H NMR}$ (360 MHz, $[\text{D}_6]\text{-DMSO}$): $\delta = 7.93$ (d, 2H, $J = 7.3$ Hz, H-Ar), 7.71 (dd, 1H, $J_{4,6} = 1.9$ Hz, $J_{6,7} = 8.2$ Hz, H-6), 7.67 (d, 2H, $J = 7.4$ Hz, H-Ar), 7.61 (d, 1H, $J_{4',6'} = 2.0$ Hz, H-4'), 7.59 (d, 1H, $^4J_{4,6} = 1.9$ Hz, H-4), 7.56-7.46 (m, 4H, H-7', H-Ar), 7.44 (d, 1H, dd, 1H, $J_{6,7} = 8.1$ Hz, H-7), 7.28 (dd, 2H, $J = 7.7$ Hz, H-Ar), 7.25 (dd, 1H, $J_{6',7'} = 2.0$ Hz, $J = 6.6$ Hz H-6'), 7.20-7.13 (m, 3H, H-benzyl), 6.92 (d, 2H, $J = 7.3$ Hz, H_o-benzyl), 5.82 (dd, 1H, $J_{2'',3''} = J_{3'',4''} = 9.9$ Hz, H-3''), 5.81 (d, 1H, $J_{1'',2''} = 3.4$ Hz, H-1''), 5.66 (d, 1H, $J_{\text{gem}} = 16.5$ Hz, H-8'), 5.58 (d, 1H, $J_{\text{gem}} = 16.5$ Hz, H-8'), 5.35 (dd, 1H, $J_{1'',2''} = 3.4$ Hz, $J_{2'',3''} = 9.9$ Hz, H-2''), 4.13 (dd, 1H, $J_{3'',4''} = J_{4'',5''} = 9.9$ Hz, H-4''), 4.05 (td, 1H, $J_{4'',5''} = 9.9$ Hz, $J_{5'',6''} = 5.9$ Hz, H-5''), 1.19 (d, 3H, $J_{5'',6''} = 5.9$ Hz, H-6'').

$^{13}\text{C NMR}$ (90 MHz, $[\text{D}_6]\text{-DMSO}$): $\delta = 188.7$ (C_q -3), 165.1, 164.9 (CO-Bz), 157.5 (C_q -3a), 157.0 (C_q -7a), 140.2 (C_q -3'), 137.7 (C_i -9'), 136.1 (C-6), 135.4 (C_q -7a'), 133.9, 133.8 (C_p -Ar), 133.2 (C_q -2), 129.3, 129.1, 128.9, 128.7 (C-Ar), 128.6 (C_q Ar), 128.4 (C_m -benzyl), 128.1 (C_q -Ar), 127.3 (C_p -benzyl), 126.5 (C_o -benzyl), 125.7 (C-6'), 125.3 (C_q 3a'),

Akashin A (1)

Under an argon atmosphere **21** was dissolved in thoroughly degassed and dried methanol (0.8 mL/mg starting material) and 30 eq of propanedithiol and 15 eq of diisopropylethylamine were added. Upon addition of the amine the color of the solution changed from blue to pale yellow. After for 3-4 d at room temperature under strict exclusion of light the reaction was transferred into dichloromethane/acetic acid 99:1 (2 mL/mg starting material) and stirred in an open flask for 60-90 minutes. Within this time the yellowish vat indigo was reoxidised to the desired blue indigo. The organic phase was extracted with 25 mM hydrochloric acid. The blue aqueous phase was submitted to solid phase extraction using SepPak[®]-C₁₈ material. Elution was carried out with acetonitrile in water (1 % acetic acid) in steps of 2.5 % from 0-30 % acetonitrile. The product eluted from 17.5-25 % acetonitrile and was lyophilized. For removal of the acetic acid the product was chromatographed by a Sephadex LH 20 column using dichloromethane/methanol (6:4) for elution.

yield: 39-73 % of **1**, blue solid

R_f = 0.10 (dichloromethane/methanol 15:1)

UV/VIS: λ_{max} (lgε) = 620 nm (4.15) in methanol

[α]_D²⁴ = + 2591.0 ° (c = 6.9 μg/mL, methanol)

C₂₂H₁₉Cl₂N₃O₅ (476.31)

ESI/MS: M_{cal.} = 475.07

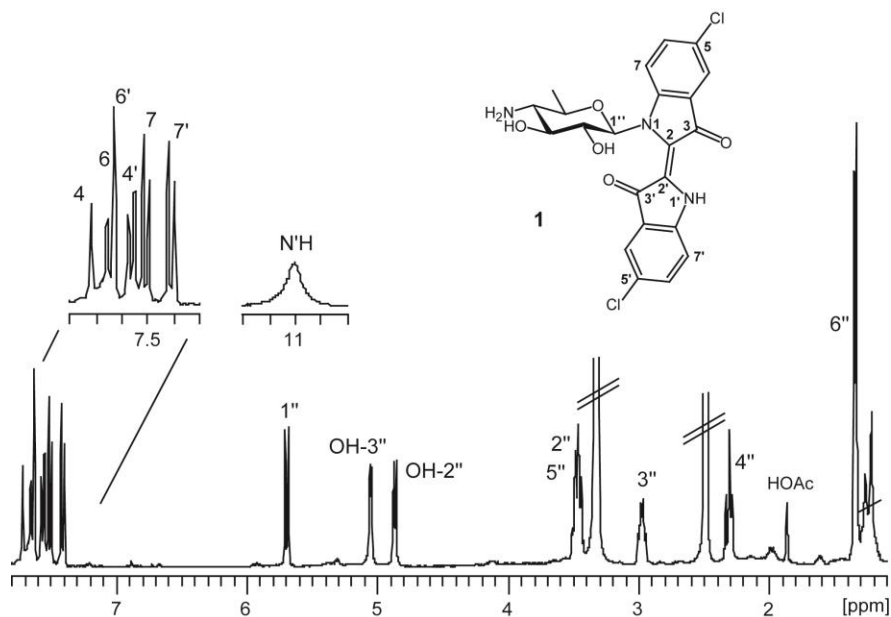
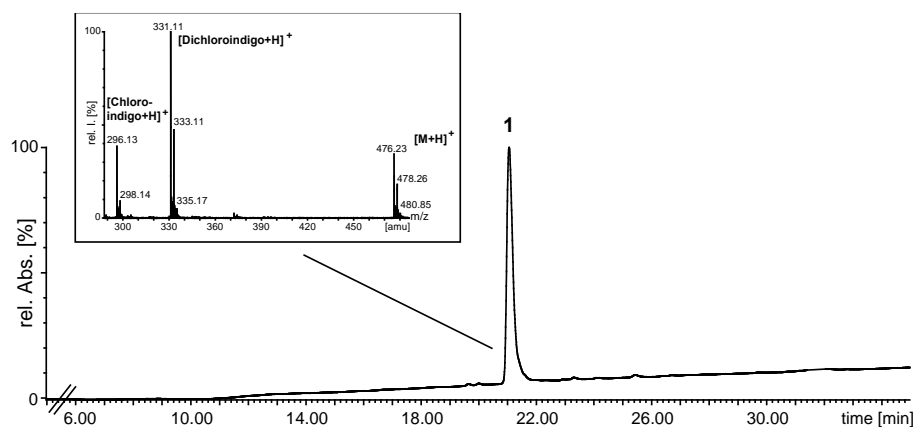
M_{found.} = 296.05 [(³⁵Cl)monochloroindigo+H]⁺, 298.05 [(³⁷Cl)monochloro-indigo+H]⁺, 331.02 [(^{35/35}Cl)dichloroindigo+H]⁺, 333.05 [(^{35/37}Cl)dichloroindigo+H]⁺, 335.05 [(^{37/37}Cl)dichloroindigo+H]⁺, 476.12 [(^{35/35}Cl)M+H]⁺, 478.14 [(^{35/37}Cl)M+H]⁺, 480.13 [(^{37/37}Cl)M+H]⁺, 498.10 [(^{35/35}Cl)M+Na]⁺, 951.24 [2M+H]⁺, 953.21 [2M+H]⁺, 955.24 [2M+H]⁺, 957.22 [2M+H]⁺, 973.22 [2M+Na]⁺, 975.21 [2M+Na]⁺, 977.21 [2M+Na]⁺, 979.21 [2M+Na]⁺

¹H-NMR (360 MHz, [D₆]-DMSO): δ = 11.00 (bs, 1H, N'H), 7.71 (d, 1H, J_{4,6} = 1.4 Hz, H-4), 7.64 (dd, 1H, J_{4,6} = 1.4 Hz, J_{6,7} = 9.0 Hz, H-6), 7.63 (d, 1H, J_{4',6'} = 1.6 Hz, H-4'), 7.55 (dd, 1H, J_{4',6'} = 1.6 Hz, J_{6',7'} = 8.6 Hz, H-6'), 7.50 (d, 1H, J_{6,7} = 9.0 Hz, H-7), 7.41 (d, 1H, J_{6',7'} = 8.6 Hz, H-7'), 5.69 (d, 1H, J_{1'',2''} = 9.1 Hz, H-1''), 5.05 (d, 1H, J_{OH-3'',3''} = 5.0 Hz, OH-3''), 4.87 (d, 1H, J_{OH-2'',2''} = 6.9 Hz, OH-2''), 3.52-3.42 (m, 2H, H-2'', H-5''), 2.95 (ddd, 1H, J_{2'',3''} = J_{3'',4''} = 9.0 Hz, J_{OH-3'',3''} = 5.0 Hz, H-3''), 2.31 (dd, 1H, J_{3'',4''} = J_{4'',5''} = 9.0 Hz, H-4''), 1.34 (d, 3H, J_{5'',6''} = 5.8 Hz, H-6'').

¹³C-NMR (90 MHz, [D₆]-DMSO): δ = 185.2 (C_q-3'), 184.1 (C_q-3), 150.7 (C_q-7a'), 149.2 (C_q-7a), 135.6 (C-6'), 134.7 (C-6), 126.6 (C_q-5), 124.6 (C_q-5'), 124.2 (C_q-2), 124.0 (C_q-2'), 123.9 (C_q-3a), 123.2 (C-4'), 122.6 (C-4), 120.2 (C_q-3a'), 117.9 (C-7), 115.1 (C-7'), 88.0 (C-1''), 77.2 (C-3''), 76.3 (C-5''), 70.0 (C-2''), 59.2 (C-4''), 18.5 (C-6'').

¹H NMR (360 MHz, [D₆]-DMSO acidified with HCl): δ = 8.46-8.38 (bs, 1H, N'H₃⁺), 7.71 (bs, 1H, H-4), 7.61 (m, 1H, H-6), 7.61 (m, 1H, H-4'), 7.56 (dd, 1H, J_{4',6'} = 1.7 Hz, J_{6',7'} = 8.1 Hz, H-6'), 7.57-7.50 (m, 1H, H-7), 7.39 (d, 1H, J_{6',7'} = 8.5 Hz, H-7'), 5.74-5.69 (m, 1H, H-1''), 3.61 (dd, 1H, J_{1'',2''} = J_{2'',3''} = 9.0 Hz, H-2''), 2.95 (dd, 1H, J_{2'',3''} = J_{3'',4''} = 9.0 Hz, H-3''), 3.00-2.86 (m, 1H, H-4''), 3.96 (qd, 1H, J_{5'',4''} = 9.9 Hz, J_{5'',6''} = 6.0 Hz, H-5''), 1.43 (d, 3H, J_{5'',6''} = 6.0 Hz, H-6'').

¹³C NMR (90 MHz, [D₆]-DMSO acidified with HCl; ¹³C-signals taken from HMQC-COSY): δ = 135.5 (C-6'), 134.0 (C-6), 123.4 (C-4'), 122.1 (C-4), 117.1 (C-7), 114.5 (C-7'), 87.1 (C-1''), 72.6 (C-3''), 71.5 (C-5''), 69.4 (C-2''), 57.0 (C-4''), 18.2 (C-6'').

a) LC-MS of **1**:b) assigned $^1\text{H-NMR}$ of **1** (360 MHz, $[\text{D}_6]$ -DMSO):**Figure S6:** LC-MS (a) and NMR (b) of **1**.**Akashin B (2):**

9.6 mg (20 μmol , 1.0 eq) of akashin A (**1**) was dissolved in 9.6 mL of methanol. 192 μL (2.0 mmol, 100 eq) of acetic anhydride and 288 μL (16 mol, 800 eq) of water were added and mixture was stirred for 30 minutes at room temperature. The crude product was concentrated in vacuo after addition of toluene. The remainder was purified by solid phase extraction over SepPak[®]-C₁₈ material using a step gradient of acetonitrile in water (20-100 % in 10 %-steps). The product eluted between 40-70 % acetonitrile and was lyophilized.

yield: 10.3 mg **2** (19.9 μmol , 99.4 %), blue solid

R_f = 0.24 (dichloromethane/methanol 15:1)

UV/VIS: λ_{max} ($\text{lg}\epsilon$) = 620 nm (4.18) in methanol

$[\alpha]_D^{27} = +4137.9^\circ$ ($c = 5.8 \mu\text{g/mL}$, methanol)

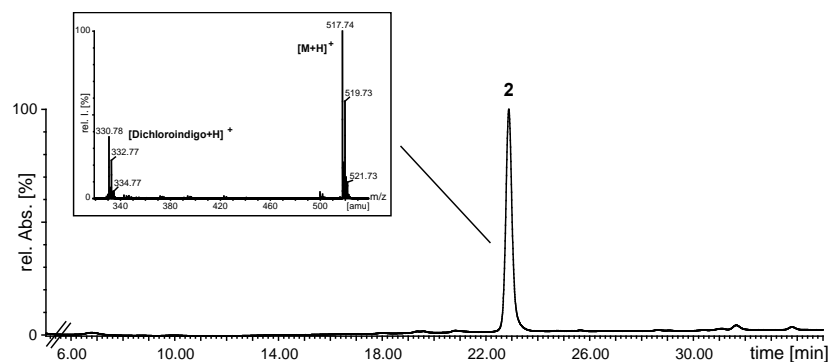
$\text{C}_{24}\text{H}_{21}\text{Cl}_2\text{N}_3\text{O}_6$ (518.35)

ESI/MS: $M_{\text{cal.}} = 517.08$

$M_{\text{found}} = 295.73$ [^{35}Cl]monochloroindigo+H $^+$, 297.73 [^{37}Cl]monochloro-indigo+H $^+$, 330.66 [$^{35/35}\text{Cl}$]dichloroindigo+H $^+$, 332.66 [$^{35/37}\text{Cl}$]dichloroindigo+H $^+$, 334.65 [$^{37/37}\text{Cl}$]dichloroindigo+H $^+$, 518.20 [$^{35/35}\text{Cl}$]M+H $^+$, 520.19 [$^{35/37}\text{Cl}$]M+H $^+$, 522.19 [$^{37/37}\text{Cl}$]M+H $^+$, 540.18 [$^{35/35}\text{Cl}$]M+Na $^+$, 542.18 [$^{35/37}\text{Cl}$]M+Na $^+$, 544.18 [$^{37/37}\text{Cl}$]M+Na $^+$, 1035.34 [2M+H] $^+$, 1037.34 [2M+H] $^+$, 1039.33 [2M+H] $^+$, 1057.33 [2M+Na] $^+$, 1059.35 [2M+Na] $^+$, 1061.32 [2M+Na] $^+$, 1063.35 [2M+Na] $^+$

$^1\text{H-NMR}$ (360 MHz, $[\text{D}_6]$ -DMSO): $\delta = 11.01$ (s, 1H, NH'), 7.77 (d, 1H, $J_{\text{NH}''',4''} = 8.9$ Hz, NH''), 7.72 (s, 1H, H-4), 7.67 (d, 1H, $J_{6,7} = 8.2$ Hz, H-6), 7.62 (s, 1H, H-4'), 7.60 - 7.54 (m, 2H, H-6', H-7), 7.41 (d, 1H, $J_{6',7'} = 8.2$ Hz, H-7'), 5.71 (d, 1H, $J_{1'',2''} = 9.0$ Hz, H-1''), 5.0 - 4.95 (m, 2H, OH-2'', OH-3''), 3.68 - 3.59 (m, 1H, H-5''), 3.60 - 3.49 (m, 2H, H-2'', H-4''), 3.24 (ddd, 1H, $J_{2'',3''} = J_{3'',4''} = 9.1$ Hz, $J_{\text{OH-3}'',3''} = 5.9$ Hz, H-3''), 1.84 (s, 3H, NH-Ac), 1.23 (d, 3H, $J_{5'',6''} = 5.1$ Hz, H-6'').

$^{13}\text{C-NMR}$ (90 MHz, $[\text{D}_6]$ -DMSO): $\delta = 187.4$ (C_q -3), 186.0 (C_q -3'), 170.1 (CO-Ac), 151.5 (C_q -7a'), 149.9 (C_q -7a), 136.4 (C-6'), 135.6 (C-6), 127.1 (C_q -5), 125.4 (C_q -5'), 125.0 (C_q -2'), 124.8 (C_q -2), 124.7 (C_q -3a), 120.9 (C_q -3a'), 123.9 (C-4'), 123.4 (C-4), 118.7 (C-7), 115.9 (C-7'), 88.6 (C-1''), 75.1 (C-3''), 71.0 (C-2''), 59.2 (C-4''), 23.8 (NAc), 19.1 (C-6'').

a) LC-MS of **2**:

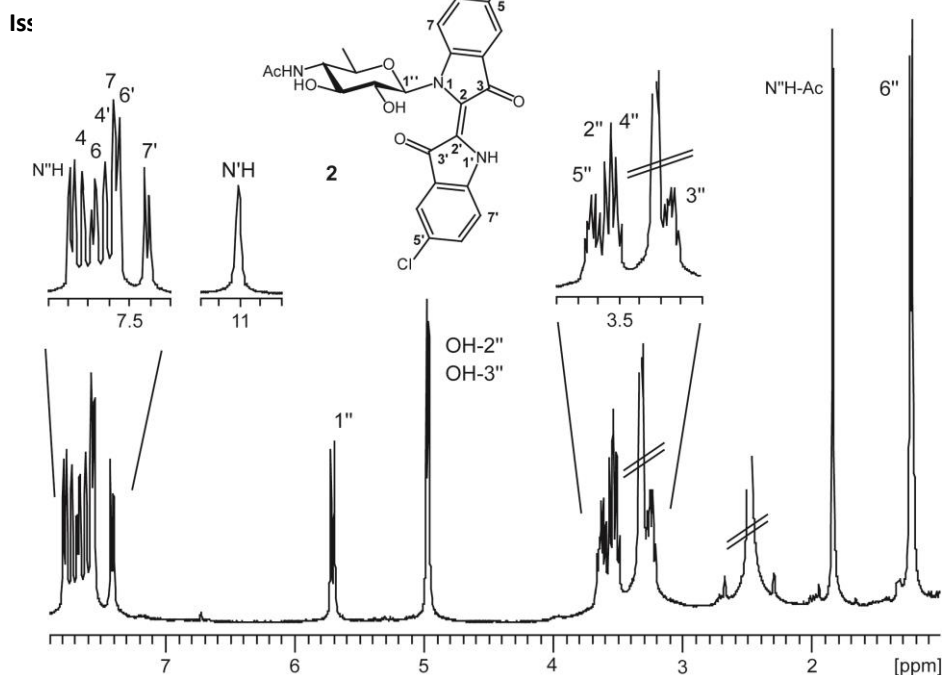


Figure S7: LC-MS (a) and NMR (b) of **2**.

Akashin C (**3**)

4 mg (8.4 μmol , 1.0 eq) of akashin A (**1**) were dissolved in 4.3 mL of absolute methanol. 4.4 μL (50.3 μmol , 6.0 eq) of diacetyl in 200 μL of absolute methanol, 12 μL (110 μmol , 13 eq) of trimethylorthoformate in 200 μL of absolute methanol and 0.39 mg (1.7 μmol , 0.2 eq) of camphorsulfonic acid in 200 μL of absolute methanol were added. After stirring for 2-3 h at room temperature 5.3 mg (84 μmol , 10 eq) of sodium cyanoborhydride in 200 μL of absolute methanol) were added. After 25 minutes the reaction was concentrated in vacuo and purified by flash chromatography (\varnothing 2 cm, \downarrow 10 cm, solvent: dichloromethane/methanol 40:1 \rightarrow 30:1 \rightarrow 25:1 containing 0.1 % formic acid).

yield: 2.7 mg **3** (4.9 μmol , 59 %), blue solid

R_f = 0.54 (dichloromethane/methanol 15:1)

UV/VIS: λ_{max} ($\text{lg}\epsilon$) = 618 nm (3.71) in methanol

$[\alpha]_D^{27} = +1143^\circ$ ($c = 14 \mu\text{g/mL}$, methanol)

$\text{C}_{26}\text{H}_{25}\text{Cl}_2\text{N}_3\text{O}_6$ (546.40)

ESI/MS: $M_{\text{cal.}} = 545.11$

$M_{\text{found}} = 296.36$ [^{35}Cl]monochloroindigo+H] $^+$, 298.36 [^{37}Cl]monochloro-indigo+H] $^+$, 331.36 [$^{35/35}\text{Cl}$]dichloroindigo+H] $^+$, 333.56 [$^{35/37}\text{Cl}$]dichloroindigo+H] $^+$, 335.36 [$^{37/37}\text{Cl}$]dichloroindigo+H] $^+$, 546.75 [$^{35/35}\text{Cl}$]M+H] $^+$, 548.73 [$^{35/37}\text{Cl}$]M+H] $^+$, 550.74 [$^{37/37}\text{Cl}$]M+H] $^+$, 568.74 [$^{35/35}\text{Cl}$]M+Na] $^+$, 570.74 [$^{35/37}\text{Cl}$]M+Na] $^+$, 1092.46 [2M+H] $^+$, 1094.47 [2M+H] $^+$, 1096.47 [2M+H] $^+$, 1098.48 [2M+H] $^+$, 1114.50 [2M+Na] $^+$, 1116.47 [2M+Na] $^+$, 1118.49 [2M+Na] $^+$, 1120.46 [2M+Na] $^+$

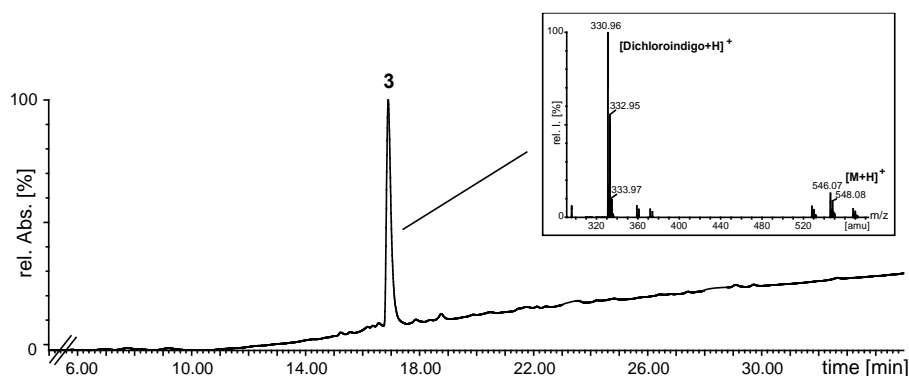
$^1\text{H-NMR}$ (360 MHz, CD_3OD): $\delta = 7.72$ (s, 1H, H-4), 7.62-7.58 (m, 3H, H-4', H-6, H-7), 7.50 (d, 1H, $J_{6',7'} = 8.7$ Hz, H-6'), 7.22 (d, 1H, $J_{6',7'} = 8.7$ Hz, H-7'), 6.05 (d, 1H, $J_{1'',2''} = 9.3$ Hz, H-1''), 3.91-3.82 (m, 2H, H-2'', H-5''), 3.77 (dd, 1H, $J_{2'',3''} = J_{3'',4''} = 9.3$ Hz, H-3''), 2.81 (q, 1H, $J_{3''',4'''} = 6.3$ Hz, H-3'''), 2.54 (dd, 1H, $J_{3'',4''} = 9.3$ Hz, H-4''), 1.45 (d, 3H, $J_{5'',6''} = 6.0$ Hz, H-6''), 1.30 (s, 3H, Me-1'''), 1.13 (d, 3H, $J_{3''',4'''} = 6.3$ Hz, Me-4''').

^{13}C NMR (90 MHz, CD_3OD ; ^{13}C -signals taken from HMQC-COSY): δ = 135.0 (C-6'), 133.9 (C-6), 122.7 (C-4'), 121.9 (C-4), 116.9 (C-7), 113.3 (C-7'), 87.7 (C-1''), 73.7 (C-3'', C-5''), 67.3 (C-2''), 61.6 (C-4''), 57.1 (C-3'''), 23.8 (C-1'''), 15.9 (C-6''), 14.7 (C-4''').

^1H -NMR (360 MHz, $[\text{D}_6]$ -DMSO): δ = 11.00 (s, 1H, NH'), 7.72 (s, 1H, H-4), 7.65-7.60 (m, 2H, H-4', H-6'), 7.59-7.51 (m, 2H, H-6, H-7), 7.40 (d, 1H, $J_{6',7'} = 8.5$ Hz, H-7'), 5.80 (d, 1H, $J_{1'',2''} = 8.4$ Hz, H-1''), 5.67 (s, 1H, OH-2''), 4.88 (d, 1H, $J_{\text{OH-}2'',2''} = 5.0$ Hz, OH-2''), 3.73-3.46 (m, 3H, H-2'', H-4'', H-5''), 2.65-2.57 (m, 1H, H-3'''), 2.45-2.40 (m, 1H, H-3''), 1.32 (d, 3H, $J_{5'',6''} = 5.2$ Hz, H-6''), 1.14 (s, 3H, Me-1'''), 0.95 (d, 3H, $J_{3''',4'''} = 6.1$ Hz, Me-4''').

^{13}C -NMR (90 MHz, $[\text{D}_6]$ -DMSO; ^{13}C -signals taken from HMQC-COSY): δ = 135.5 (C-6'), 134.5 (C-6), 122.9 (C-4'), 122.5 (C-4), 117.5 (C-7), 114.9 (C-7'), 88.1 (C-1''), 74.1 (C-2''), 73.8 (C-5''), 67.1 (C-4''), 61.9 (C-3''), 57.5 (C-3'''), 25.3 (C-1'''), 17.8 (C-6''), 16.6 (C-4''').

a) LC-MS of **3**:



b) assigned ^1H -NMR of **3** (360 MHz, CD_3OD):

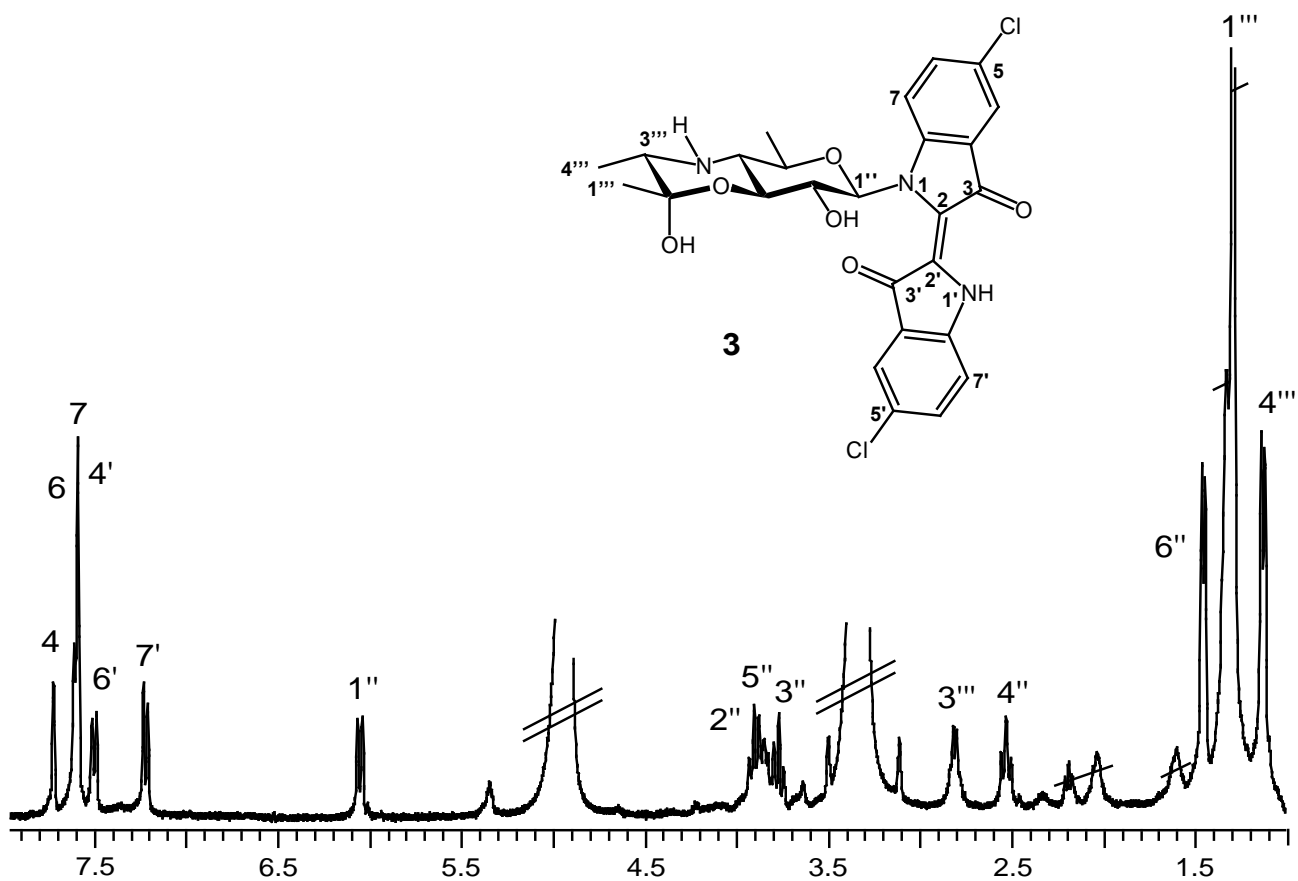


Figure S8: LC-MS (a) and NMR (b) of **3**.