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Reaction of *N*-Fmoc aspartic anhydride with glycosylamines: a simple entry to *N*-glycosyl asparagines

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ABSTRACT

The reaction of *N*-Fmoc-aspartic anhydride with glycosyl amines in DMSO selectively leads to the formation of β-substituted products, thus providing a simple and efficient route to *N*-glycosyl asparagine derivatives, the building blocks for glycopeptide synthesis.

The growing attention paid to glycoproteins is due to the numerous roles they play in biological processes.[1] As small analogs of glycoproteins, glycopeptides are convenient models for investigating the biological and physiological functions of glycoconjugates in biological systems.[2] Glycopeptides have been used for the preparation of synthetic vaccines[3] and for therapeutic applications.[2b,4] Moreover, the ability of glycosylated peptides to penetrate the blood–brain barrier[5] may allow their use in the treatment of brain diseases.[6]

The utilization of *N*-glycosyl asparagine derivatives as building blocks for the synthesis of *N*-glycopeptides remains the most versatile approach[7] because it is suitable for solid-phase synthesis.[8]

Most of the known procedures for the preparation of *N*-glycosyl asparagine derivatives involve coupling of glycosylamine[9] or its equivalent[10] with a suitably protected and activated aspartic acid derivative, the synthesis of which is a laborious multistep procedure. Five steps are necessary for the preparation of 1-*tert*-butyl *N*-Fmoc-L-aspartate, the typical precursor of *N*-glycosyl asparagines.[11] Another two steps are required for the activation of the β-carboxylic function and for the deprotection of the α-carboxylic group of the resulting *N*-glycosyl asparagine ester.

In contrast, N-protected cyclic aspartic anhydride derivatives can be easily prepared from aspartic acid in two steps. These compounds are powerful acylation agents that react readily with amines. No additional protection-deprotection steps are required. When the reaction is regioselective, it leads to the formation of the desired asparagine derivative with a free α-carboxylic group ready for peptide chain elongation.

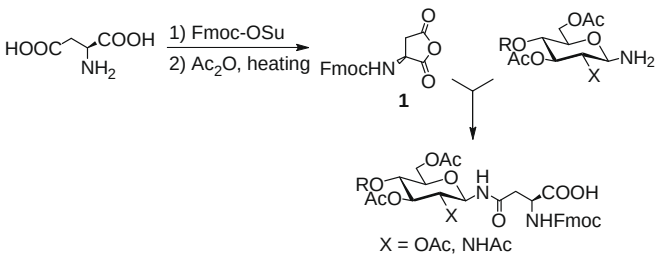
Aspartic anhydride derivatives are well known and are widely used in the synthesis[12] of various N-substituted asparagine derivatives. Moreover, some aspartic anhydride derivatives have been used for the preparation of N-glycosylated asparagines and peptides.[13] However, the regioselectivity was poor, leading to almost equal amounts of asparagine and isoasparagine derivatives.

Recently, *N*-Fmoc-aspartic anhydride has been applied for the preparation of various N-substituted asparagine derivatives. In DMSO, the reaction with amines leads almost exclusively to the formation of the desired β-N-substituted products with good regioselectivity.[14]

Here we present an efficient method for the preparation of N-glycosylated asparagine derivatives based upon the reaction of *N*-Fmoc-aspartic anhydride with per-O-acetylated glycosyl amines (Scheme 1).

N-Fmoc-aspartic anhydride (**1**) was prepared from aspartic acid in two steps in high yield. The product precipitated from the reaction mixture as colorless needles and did not require any purification. It is stable and can be stored for long periods at room temperature.

We found that the regioselectivity of *N*-Fmoc-aspartic anhydride aminolysis with per-O-acetylated glycosyl amines varied greatly, depending on the polarity of the reaction media. In less polar solvents, the isoasparagine derivative was the main product, whereas more polar solvents increased the yields of the desired glycosylated asparagines. The highest yields of the target products



Scheme 1. Synthesis of *N*-glycosyl asparagines.

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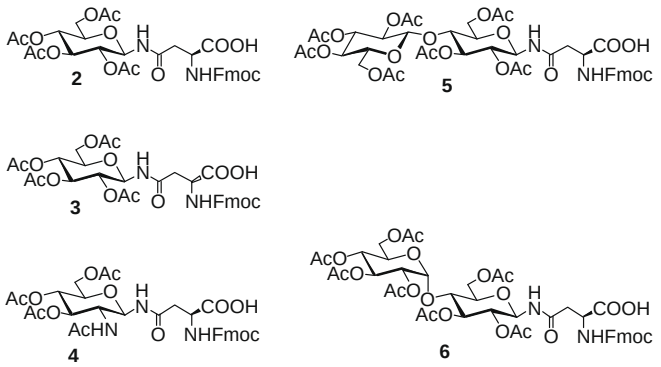

Figure 1. Synthesized *N*-glycosyl asparagines.

were obtained using DMSO as the reaction solvent. The reaction proceeds smoothly and usually takes about 1 h, affording the desired products in high yield after a simple work-up.

Typical procedures are as follows. *N*-Fmoc-aspartic anhydride: A solution of Fmoc-OSu (5.1 g, 15 mmol) in THF (30–50 mL) was added with stirring to a solution of aspartic acid (2.0 g, 15 mmol) in H_2O (30 mL) containing Na_2CO_3 (3.5 g, 33 mmol). After stirring at room temperature for 20 h, the mixture was washed with ether (2×20 mL). The aqueous phase was acidified to pH 2 using HCl, and was extracted with EtOAc (2×30 mL). The organic phase was dried ($MgSO_4$) and evaporated. The residue was dissolved in Ac_2O (15 mL) with rapid heating and shaking. When the starting material had dissolved (typically in 1 min), the reaction mixture was quickly cooled to room temperature. The precipitate was filtered off, washed with dry ether and dried in a desiccator. The overall yield of *N*-Fmoc-aspartic anhydride was 79%. Per-O-acetylated glycosyl amines were synthesized from the corresponding glycosyl azides, prepared by the published methods,[15] using palladium-on-charcoal catalyzed hydrogenolysis in EtOAc. Glycosylated *N*-Fmoc asparagine derivatives: to a stirred solution of acetylated glycosyl amine (5 mmol) in DMSO (3 mL), *N*-Fmoc-aspartic anhydride (5.0 mmol) was added. The reaction mixture was kept at ambient temperature for one hour and then diluted with methanol. The precipitate was filtered and recrystallized from methanol. In the case of disaccharide derivatives, the product was isolated by dilution of the reaction mixture with water, followed by extraction with $CHCl_3$. Evaporation and recrystallization from methanol afforded the pure product.

In order to demonstrate the practical utility of the approach, *N*-Fmoc-asparagine derivatives of glucose **2**, *N*-acetylglucosamine **4**, cellobiose **5**, and maltose **6** were prepared (Fig. 1). The obtained products were characterized by NMR and HRMS.[16]

Possible racemization during the preparation of *N*-Fmoc-L-aspartic anhydride[17] was evaluated by ^{1}H NMR, using as a standard, the acetylated glucosyl D-asparagine derivative (**3**), prepared under the same reaction conditions as the L-isomer (**2**).

The ^{1}H NMR spectrum (Fig. 2) of the racemic mixture (spectrum DL) contains two pairs of doublets due to the amide protons (at δ 8.75 and 7.56 for the L-isomer and at δ 8.78 and 7.62 for the D-isomer). The signal due to one of the methylene protons of the D-isomer at 2.44 was also well resolved. Therefore, the amount of the D derivative could be estimated by integration of these signals. *N*-Fmoc-aspartic anhydride and per-*O*-acetyl β-glucosylamine were dissolved in DMSO-d_6 and the ^{1}H NMR spectrum of the reaction mixture was recorded after 1 h, when the reaction was complete (rm). This spectrum contains two weak signals (about 0.2–0.3% intensity, visible at high magnification) at δ 8.78 and 2.44, possibly arising from the D-isomer. From this observation it can be concluded that racemization of the *N*-Fmoc aspartic anhydride was less than 0.3%.

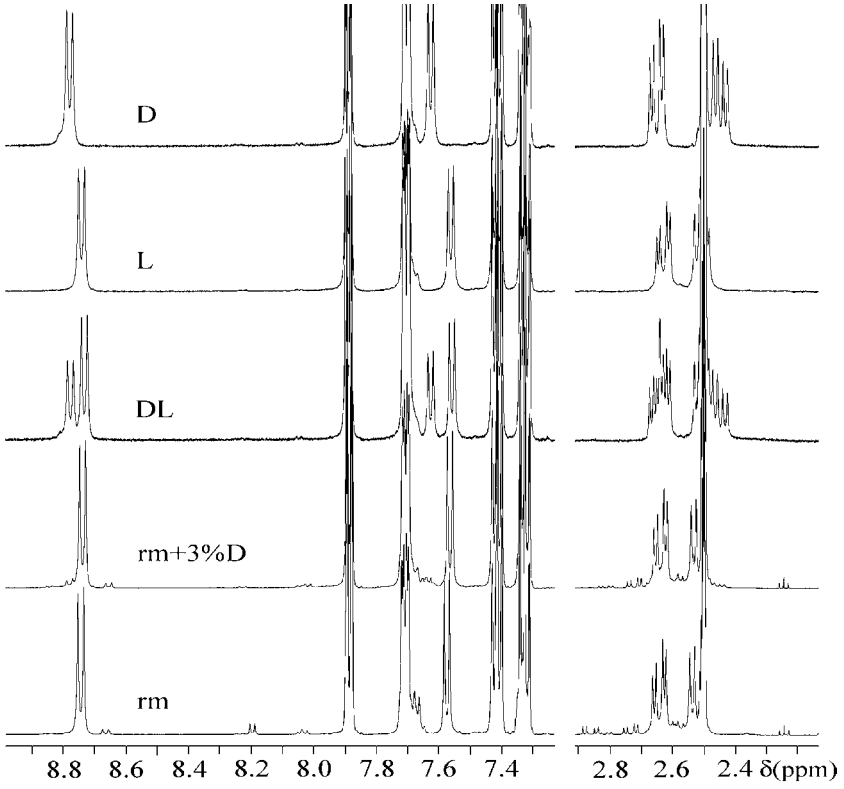

Figure 2. ^{1}H NMR spectra. D and L, pure D- and L-isomers, respectively; (DL) mixture of both isomers; rm, reaction mixture; rm + 3% D, reaction mixture containing 3% of the D-isomer.

The spectrum (rm + 3% D), corresponding to the reaction mixture with 97% ee, was measured after the addition of about 3% per-O-acetylated glucosyl *N*-Fmoc-D-asparagine to the reaction mixture.

In summary, a simple and efficient one-step method for the preparation of *N*-glycosyl asparagine derivatives, based on the reaction of glycosyl amines with *N*-Fmoc-aspartic anhydride in DMSO, has been developed. The starting anhydride can easily be prepared in two steps from aspartic acid. The method provides an efficient and simple route for the preparation of various *N*-glycosyl asparagine derivatives, the building blocks for liquid- and solid-phase syntheses of glycopeptides.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.tetlet.2009.08.106.

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16. The NMR data are presented using the convention followed in Carbohydrate Research (see instructions to Authors).*N*-Fmoc-L-aspartic anhydride (**1**). ^{1}H NMR (DMSO-d_6, 500 MHz): δ (ppm) 8.21 (d, 1H, J = 7.6 Hz, NH-Asp), 7.88 (d, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 7.68 (d, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 7.42 (t, 2H, J = 7.4, Hz, H_{Ar}-Fmoc), 7.34 (t, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 4.70 (dq, 1H, $J_{,1}$ = 10.1 Hz, $J_{,2}$ = 6.2 Hz, α-H-Asp), 4.44 (dd, 1H, $J_{2a,2b}$ = 10.4 Hz, a-CH_2-Fmoc), 4.41 (dd, 1H, b-CH_2-Fmoc), 4.26 (dd, 1H, $J_{a,9}$ = 6.7 Hz, $J_{b,9}$ = 6.3 Hz, H-9 Fmoc), 3.27 (dd, 1H, $J_{1,2}$ = 18.4 Hz, β1-H-Asp), 2.89 (dd, 1H, β2-H-Asp). ^{13}C NMR (DMSO-d_6, 125 MHz): δ (ppm) 172.2, 169.9 (CO-Asp), 156.0 (CO-Fmoc), 143.7, 143.6, 140.9, 127.8, 127.2, 127.1, 125.2, 125.1, 120.3, 120.2 (Ar-Fmoc), 66.2 (CH_2-Fmoc), 50.5 (CH-Asp), 46.7 (CH-Fmoc), 34.8 (CH_2-Asp).2,3,4,6-tetra-*O*-acetyl-*N*-[*N*-Fmoc-L-aspart-4-oyl]-β-D-glucopyranosylamine (**2**). Yield 82%. ^{1}H NMR (DMSO-d_6, 500 MHz): δ (ppm) 8.75 d 1H J_{1NH} = 9.4 Hz, NH-Glc), 7.88 (d, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.70 (dd, 2H, J = 7.4 Hz, J = 7.4 Hz, H_{Ar}-Fmoc), 7.57 (d, 1H, J = 8.4 Hz, NH-Asn), 7.40 (t, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.32 (t, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 5.41 (dd, 1H, $J_{1,2}$ = 9.3 Hz, H-1), 5.33 (dd, 1H, $J_{3,4}$ = 9.6 Hz, H-3), 4.91 (dd, 1H, $J_{4,5}$ = 9.7 Hz, H-4), 4.82 (dd, 1H, $J_{2,3}$ = 9.4 Hz, H-2), 4.38 (dt, 1H, $J_{,1}$ = 6.1 Hz, $J_{,2}$ = 7.4 Hz, α-CH-Asn), 4.27 (d, 2H, a,b-CH_2-Fmoc), 4.20 (dd, 1H, J = 6.6 Hz, H-9 Fmoc), 4.16 (dd, 1H, $J_{6a,6b}$ = 12.3 Hz, H-6a), 4.09 (dq, 1H, $J_{5,6a}$ = 4.4 Hz, $J_{5,6b}$ = 2.2, H-5), 3.97 (dd, 1H, H-6b), 2.64 (dd, 1H, $J_{1,2}$ = 15.8 Hz, β1-H-Asn), 2.52 (m, 1H, β2-H-Asn), 1.99, 1.98, 1.92, 1.89 (4s, each 3H, 4 × CH_3CO). ^{13}C NMR (DMSO-d_6, 125 MHz): δ (ppm) 172.81 (COOH), 169.90 (CO-NH), 169.63, 169.39, 169.20, 169.00 (CO-Ac), 155.73 (CO-Fmoc), 143.70, 143.64, 140.59, 127.53, 126.98, 125.16, 120.00 (Ar-Fmoc), 76.70 (C-1), 72.77 (C-3), 71.99 (C-5), 70.45 (C-2), 67.69 (C-4), 65.60 (CH_2-Fmoc), 61.62 (C-6), 50.12 (CH-Asn), 46.49 (CH-Fmoc), 20.41, 20.27, 20.21, 20.18 (CH_3-Ac). ESI FT-ICR HRMS (*m*/*z*): 707.2066 (707.2064 calculated for $C_{33}H_{36}N_2NaO_{14}$ [M+Na]$^+$).2,3,4,6-tetra-*O*-acetyl-*N*-[*N*-Fmoc-D-aspart-4-oyl]-β-D-glucopyranosylamine (**3**). Yield 51%. ^{1}H NMR (DMSO-d_6, 500 MHz): δ (ppm) 8.83 d 1H J_{1NH} = 9.4 Hz, NH-Glc), 7.89 (d, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.70 (dd, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 7.59 (d, 1H, J = 7.9 Hz, NH-Asn), 7.41 (dt, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.33 (2dt, 2H, J = 7.5 Hz, J = 7.4 Hz, H_{Ar}-Fmoc), 5.41 (dd, 1H, $J_{1,2}$ = 9.4 Hz, H-1), 5.34 (dd, 1H, $J_{3,4}$ = 9.6 Hz, H-3), 4.88 (dd, 1H, $J_{4,5}$ = 9.7 Hz, H-4), 4.81 (dd, 1H, $J_{2,3}$ = 9.4 Hz, H-2), 4.36 (dt, 1H, $J_{,1}$ = 6.2 Hz, $J_{,2}$ = 7.3 Hz, α-CH-Asn), 4.29 (d, 2H, a,b-CH_2-Fmoc), 4.22 (m, 1H, H-9 Fmoc), 4.13 (dd, 1H, $J_{6a,6b}$ = 12.3 Hz, H-6a), 4.08 (ddd, 1H, $J_{5,6a}$ = 4.4 Hz, $J_{5,6b}$ = 2.2, H-5), 3.95 (dd, 1H, H-6b), 2.64 (dd, 1H, $J_{1,2}$ = 15.7 Hz, β1-H-Asn), 2.44 (dd, 1H, β2-H-Asn), 1.983, 1.978, 1.93, 1.92 (4s, each 3H, 4 × CH_3CO).^{13}C NMR (DMSO-d_6, 125 MHz): δ (ppm) 172.79 (COOH), 169.89 (CO-NH), 169.64, 169.40, 169.21, 169.08 (CO-Ac), 155.71 (CO-Fmoc), 143.69, 140.60, 127.53, 127.00, 125.15, 120.02 (Ar-Fmoc), 76.69 (C-1), 72.75 (C-3), 71.99 (C-5), 70.53 (C-2), 67.70 (C-4), 65.58 (CH_2-Fmoc), 61.61 (C-6), 50.10 (CH-Asn), 46.1 (CH-Fmoc), 20.41, 20.28, 20.22, 20.20 (CH_3-Ac). ESI FT-ICR HRMS (*m*/*z*): 707.2066 (707.2064 calculated for $C_{33}H_{36}N_2NaO_{14}$ [M+Na]$^+$).2-acetamido-2-deoxy-3,4,6-tri-*O*-acetyl-*N*-[*N*-Fmoc-L-aspart-4-oyl]-β-D-glucopyranosylamine (**4**). Yield 85%. ^{1}H NMR (DMSO-d_6, 500 MHz): δ (ppm) 8.61 d 1H J_{1NH} = 9.0 Hz, NH-Glc), 7.91 (d, 1H, J = 9.0 Hz, NH-Ac), 7.88 (d, 2H, J = 7.6 Hz, H_{Ar}-Fmoc), 7.70 (dd, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.52 (d, 1H, J = 8.6 Hz, α-NH-Asn), 7.41 (t, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.32 (t, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 5.19 (dd, 1H, $J_{1,2}$ = 9.5 Hz, H-1), 5.11 (dd, 1H, $J_{3,4}$ = 9.8 Hz, H-3), 4.83 (dd, 1H, $J_{4,5}$ = 9.8 Hz, H-4), 4.40 (dt, 1H, $J_{,1}$ = 5.5 Hz, $J_{,2}$ = 8.3 Hz, α-CH-Asn), 4.36 (dd, 1H, $J_{a,b}$ = 10.3 Hz, $J_{a,9}$ = 7.0 Hz, a-CH_2 Fmoc), 4.25 (dd, 1H, $J_{b,9}$ = 6.9 Hz, b-CH_2 Fmoc), 4.21 (m, 1H, H-9 Fmoc), 4.19 (dd, 1H, $J_{6a,6b}$ = 12.4 Hz, H-6a), 3.95 (m, 1H, H-6b), 3.88 (dd, 1H, $J_{2,3}$ = 9.8 Hz, $J_{2,NH}$ = 9.8 Hz, H-2), 3.82 (dq, 1H, $J_{5,6a}$ = 4.1 Hz, $J_{5,6b}$ = 1.9 Hz, H-5), 2.67 (dd, 1H, $J_{a,b}$ = 16.1 Hz, β1-H-Asn), 2.51 (m, 1H, β2-H-Asn), 1.99, 1.96, 1.91 (3s, each 3H, 3 × CH_3CO), 1.73 (s, 3H, CH_3CON). ^{13}C NMR (DMSO-d_6, 125 MHz): δ (ppm) 172.85 (COOH), 169.92 (CO-NH Asn), 169.70 (CO-AcNH), 169.39, 169.37, 169.20 (CO-Ac), 155.72 (CO-Fmoc), 143.68, 140.59, 127.53, 126.98, 125.15, 120.00 (Ar-Fmoc), 77.98 (C-1), 73.26 (C-3), 72.18 (C-5), 68.28 (C-4), 65.60 (CH_2-Fmoc), 61.73 (C-6), 52.01 (C-2), 49.91 (CH-Asn), 46.50 (CH-Fmoc), 36.77 (CH_3-NAc), 20.42, 20.30, 20.27 (CH_3-Ac). ESI FT-ICR HRMS (*m*/*z*): 706.2225 (706.2224 calculated for $C_{33}H_{37}N_3NaO_{13}$ [M+Na]$^+$).4-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-2,3,6-tri-*O*-acetyl-*N*-[*N*-Fmoc-L-aspart-4-oyl]-β-D-glucopyranosylamine (**5**). Yield 73%. ^{1}H NMR ($CDCl_3$, 500 MHz): δ (ppm) 7.75 (d, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.58 (dd, 2H, J = 7.5 Hz, H_{Ar}-Fmoc), 7.39 (t, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 7.30 (t, 2H, J = 7.4 Hz, H_{Ar}-Fmoc), 6.68 d 1H J_{1NH} = 9.0 Hz, NH-Glc), 6.19 (d, 1H, J = 8.1 Hz, NH-Asn), 5.30 (dd, 1H, $J_{3,4}$ = 8.2 Hz, H-3), 5.23 (dd, 1H, $J_{1,2}$ = 9.4 Hz, H-1), 5.14 (dd, 1H, $J_{3',4'}$ = 9.3 Hz, H-3′), 5.07 (dd, 1H, $J_{4',5'}$ = 9.7 Hz, H-4′), 4.92 (dd, 1H, $J_{2',3'}$ = 9.0 Hz, H-2′) 4.84 (dd, 1H, $J_{2,3}$ = 9.5 Hz, H-2), 4.57 (m, 1H, $J_{,1}$ = 4.6 Hz, $J_{,2}$ = 5.0 Hz, H-α-Asn), 4.50 (d, 1H, $J_{1',2'}$ = 8.0 Hz, H-1), 4.46 (m, 1H, $J_{6a,6b}$ = 11.9 Hz, H-6a), 4.39 (m, 1H, $J_{a,b}$ = 10.8 Hz, a-CH_2-Fmoc), 4.36 (m, 1H, $J_{6'a,6'b}$ = 12.7 Hz, H-6′a), 4.34 (m, 1H, b-CH_2-Fmoc), 4.21 (dd, 1H, J = 7.3 Hz, H-9-Fmoc), 4.14 (m, 1H, H-6b), 4.03 (dd, 1H, H-6′b), 3.75 (m, 2H, H-4, H-5), 3.65 (dq, 1H, $J_{5',6'a}$ = 3.5 Hz, $J_{5',6'b}$ = 2.4 Hz, H-5′), 2.89 (dd, 1H, $J_{1,2}$ = 16.2 Hz, H-β1-Asn), 2.75 (dd, 1H, H-β2-Asn), 2.09, 2.08, 2.024, 2.02, 2.01, 1.98 (6s, 21H, 7 × CH_3CO). ^{13}C NMR ($CDCl_3$, 125 MHz): δ (ppm) 172.61 (COOH), 171.59, 171.47, 170.51, 170.36, 170.24, 169.65, 169.30, 169.06 (CO-NH, CO-Ac), 156.64 (CO-Fmoc), 143.69, 143.61, 141.25, 127.76, 127.10, 125.12, 120.00 (Ar-Fmoc), 100.63 (C-1′), 77.89 (C-1), 76.09, 74.72, 72.87, 72.15, 71.93, 71.52, 70.75, 67.79 (C-2, C-2′, C-3, C-3′, C-4, C-4′, C-5, C-5′),

67.48 (CH_2-Fmoc), 61.74, 61.60 (C-6, C-6′), 50.30 (CH-Asn), 47.00 (CH-Fmoc), 37.63 (CH_2-Asn), 20.83, 20.64, 20.57, 20.52, 20.49 (CH_3-Ac). ESI Q-TOF HRMS (*m*/*z*): 995.2939 (995.2909 calculated for $C_{45}H_{52}N_2NaO_{22}$ $[M+Na]^+$).4-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)-2,3,6-tri-*O*-acetyl-*N*-[*N*-Fmoc-L-aspart-4-oyl]-β-D-glucopyranosylamine (**6**). Yield 67%. ^{1}H NMR ($CDCl_3$, 500 MHz): δ (ppm) 7.74 (d, 2H, *J* = 7.6 Hz, H_{Ar}-Fmoc), 7.60 (dd, 2H, *J* = 7.5 Hz, H_{Ar}-Fmoc), 7.41 (t, 2H, *J* = 7.5 Hz, H_{Ar}-Fmoc), 7.32 (t, 2H, *J* = 7.6 Hz, H_{Ar}-Fmoc), 6.80 d 1H J_{1NH} = 9.1 Hz, NH-Glc), 6.24 (d, 1H, *J* = 7.9 Hz, NH-Asn), 5.39 (d, 1H, $J_{1',2'}$ = 4.1 Hz, H-1′), 5.38 (dd, 1H, $J_{3,4}$ = 9.4 Hz, H-3), 5.35 (dd, 1H, $J_{3',4'}$ = 10.1 Hz, H-3′), 5.29 (dd, 1H, $J_{1,2}$ = 9.3 Hz, H-1), 5.06 (dd, 1H, $J_{4',5'}$ = 9.9 Hz, H-4′), 4.85 (dd, 1H, $J_{2',3'}$ = 10.6 Hz, H-2′), 4.79 (dd, 1H, $J_{2,3}$ = 9.4 Hz, H-2), 4.55 (m, 1H, $J_{,1}$ = 4.0 Hz, $J_{,2}$ = 4.0 Hz, H-α-Asn), 4.42 (dd, 1H, $J_{6a,6b}$ = 12.3 Hz, H-6a), 4.35 (ddd, 2H, $J_{a,b}$ = 10.8 Hz, a,b-CH_2-Fmoc), 4.23 (dd, 1H, $J_{6'a,6'b}$ = 12.6 Hz, H-6′a), 4.21 (dd, 1H, *J* = 7.7 Hz, H-9-Fmoc), 4.20 (m, 1H, H-6b), 4.05 (dd, 1H, H-6′b), 3.96 (dd, 1H, $J_{4,5}$ = 9.3 Hz, H-4), 3.93 (m, 1H, $J_{5',6'a}$ = 2.8 Hz, $J_{5',6'b}$ = 2.8 Hz, H-5′), 3.78 (m, 1H, $J_{5,6a}$ = 3.4 Hz, $J_{5,6b}$ = 2.9 Hz, H-5), 2.87 (dd, 1H, $J_{1,2}$ = 16.5 Hz, H-β1-Asn), 2.74 (dd, 1H, H-β2-Asn), 2.09, 2.04, 2.02, 2.01, 2.01, 2.00 (5s, 21H, 7 × CH_3CO). ^{13}C NMR ($CDCl_3$, 125 MHz): δ (ppm) 173.16 (COOH), 171.37, 171.33, 170.60, 170.57, 170.54, 169.99, 169.85, 169.43 (CO-NH, CO-Ac), 156.40 (CO-Fmoc), 143.68, 143.63, 141.23, 127.75, 127.09, 125.13, 120.00 (Ar-Fmoc), 95.60 (C-1′), 77.52 (C-1), 75.08, 74.11, 72.46, 71.25, 69.98, 69.24, 68.55, 67.89 (C-2, C-2′, C-3, C-3′, C-4, C-4′, C-5, C-5′), 67.45 (CH_2-Fmoc), 62.63, 61.36 (C-6, C-6′), 50.51 (CH-Asn), 46.98 (CH-Fmoc), 37.63 (CH_2-Asn), 20.88, 20.77, 20.65, 20.58, 20.56, 20.54, 20.51 (CH_3-Ac). ESI Q-TOF HRMS (*m*/*z*): 995.2921 (995.2909 calculated for $C_{45}H_{52}N_2NaO_{22}$ $[M+Na]^+$).

17. Buron, F.; Deguest, G.; Bischoff, L.; Fruit, C.; Marsais, F. *Tetrahedron: Asymmetry* **2007**, *18*, 1625–1627.