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Hexafluoroacetone as Protecting and Activating Reagent. A New Approach to *N*-Glycosides

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Summary. *N*-Glycosides of *Asn* and homologues have been synthesized starting from hexafluoroacetone-protected ω-activated *Asp*, *Glu*, and *Aad* derivatives and glycosylamines. The synthetic value of the new building blocks was demonstrated by the concise incorporation of *N*-glycosylated *Asn*, *Gln*, and *Aad* ω-amides into glycopeptides.

Keywords. Hexafluoroacetone; *N*-Glycosylation; *N*-Glycosylamino acids; *N*-Glycopeptides; Dielectrophiles.

Introduction

O- and *N*-Glycosylation are important co- and post-translational modifications of proteins [1–3]. Glycosylation influences properties of peptides and proteins such as solubility, thermal and proteolytical stability [4], as well as conformation and folding [5]. In addition, it can also affect biological functions of proteins in events of molecular recognition, like cell to cell communication and adhesion of bacteria or of viruses to cell-surface proteins [6]. This suggests an enormous potential of glycoconjugates in drug design [7], particularly because glycocluster exhibit enhanced binding properties [8].

Glycoconjugates isolated from biological sources or produced gentechnologically are microheterogeneous [9]. The different properties exhibited by each component within these microheterogenous mixtures create problems in determining the exact function in structure/activity relationships [10, 11]. Consequently, new methodologies for the assembly of homogeneous and more robust glycopeptides and glycopeptide mimetics have to be developed [12], applying the chemoenzymatic

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[13], the convergent, and the building block approach [14]. Among these strategies the building block approach is particulary attractive due to the ease of incorporating the desired glycosylated amino acid *via* solid-phase peptide synthesis (SPPS) [14, 15].

The remarkable feature of the *N*-glycosylation is that the generally unreactive carboxamide nitrogen of asparagine reacts as nucleophile in an enzyme mediated reaction. Direct chemical glycosylation of asparagine has not been described so far [16]. There are no confirmed reports on the glycosylation of glutamine [1].

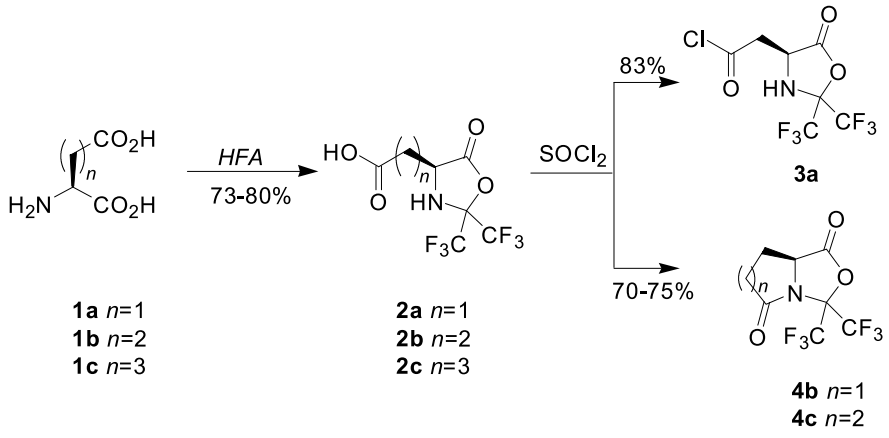
The preferred synthetic approach to *N*-glycosyl asparagine is the coupling reaction of glycosylamines [17] with β-activated α-protected aspartic acid derivatives [18]. *Via* this route the first examples of *N*-glycosylated α-trifluoromethyl asparagine derivatives have been prepared [19]. However, this strategy is often plagued by ready anomerization of the glycosylamine moiety, to give anomeric mixtures of glycopeptides [20, 21]. Furthermore, the glycosylamine approach may also be complicated by intramolecular aspartimide formation [10a]. An alternative approach is the chemical ligation of an electrophilic carbohydrate unit with a nucleophilic amino acid ω-amide derivative. In this context, a series of new nucleophilic asparagine surrogates have been introduced, *i.a.* alanine-β-hydrazide [22, 23], asparagine hydroxamate [24], alanine-β-hydroxylamine [22], to serve as glycosyl acceptors.

Results and Discussion

We now report on a new protection/activation strategy which offers some significant advantages: The introduction and the cleavage of the protecting group occur under mild conditions. Since amino group protection and carboxy group activation and on the other hand peptide bond formation and deblocking of the amino group occur in one step, the new strategy offers the possibility of a shorter reaction sequence. Moreover, the reaction can be monitored easily and quickly by ^{19}F NMR spectroscopy without any loss of material or even can be run in a NMR tube.

ω-Carboxy-α-amino acids (aspartic, glutamic, and α-amino adipic acid) react with hexafluoroacetone (*HFA*) in *DMSO* or *DMF* at room temperature to give exclusively five-membered lactones **2** in very good yields [25]. In only one step, protection of the α-amino and the adjacent carboxy group is achieved. Concomitantly, the α-carboxy group is selectively activated toward nucleophiles [26]. The ω-carboxy group remains unaffected and can be derivatized separately after selective activation (**2a** $\rightarrow$ **3a**, Scheme 1).

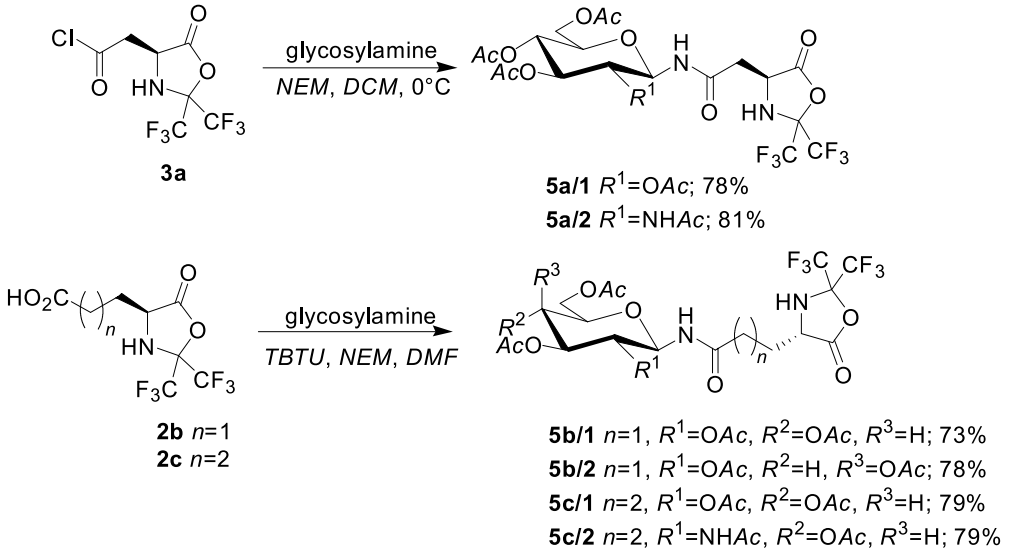
The β-carboxy group of hexafluoroacetone-protected aspartic acid can be regioselectively activated by transformation into the corresponding acid chloride on treatment with thionyl chloride (**2a** $\rightarrow$ **3**). In the case of *HFA*-protectected glutamic and α-aminoadipic acid, the acid chlorides formed, spontaneously undergo intramolecular ring closure to give bicyclic lactam derivatives **4b** and **4c** [27]. However, activation of the ω-carboxy group of *Glu* and *Aad* can be achieved *via* acid fluorides [28, 29], mixed anhydrides, and on treatment with *TBTU* without competing lactam formation. The ω-activated compound **3a** represents a dielectrophile. The two electrophilic centers exhibit different reactivities. Consequently, **3a** is capable for two consecutive acylation steps. Especially on reaction with weak



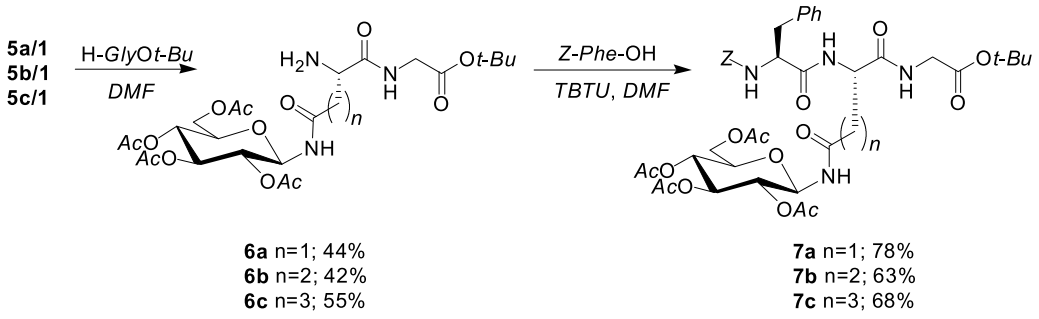
Scheme 1

nucleophiles, like glycosylamines, excellent regioselectivities are obtained. ω-Glycosylation in the presence of bases like *N*-ethyl morpholine (*NEM*) furnishes *N*-glycosylated products **5**, while the lactone moiety functions in this reaction step as protective group. The yields after purification by flash chromatography or recrystallization are high (70–80%, Scheme 2). ^{1}H NMR and ^{19}F NMR spectra taken from the crude products, give no evidence for the formation of α-anomers as by-products. TLC analysis confirms these findings.

Then, in a second step under appropriate conditions the lactone moiety reacts as an activated ester, to give *N*-glycosylated dipeptides on reaction with amino acid esters (**5** → **6**). Peptide bond formation *via* cleavage of the lactone ring is always coupled with the deprotection of the *N*-terminal amino group. Therefore, elongation of the peptide backbone can be continued in *N*-terminal position without the



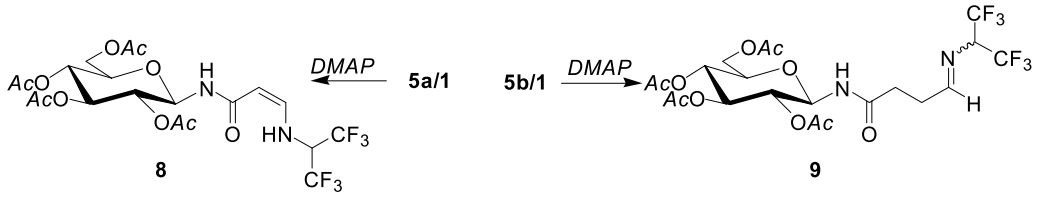
Scheme 2



6a n=1; 44%
6b n=2; 42%
6c n=3; 55%

7a n=1; 78%
7b n=2; 63%
7c n=3; 68%

Scheme 3



Scheme 4

need of a separate deprotection step (**6** → **7**). The reaction sequence is perfectly suited for the generation of libraries of *N*-glycosylated peptides (Scheme 3).

When 2,2-bis(trifluoromethyl)-1,3-oxazolidin-5-ones **5** are treated with dimethylaminopyridine (*DMAP*) in dichloromethane (*DCM*) at room temperature a fragmentation reaction was initiated starting with a β-elimination step, (**5a**/**1** → **8**; **5b**/**1** → **9**) mechanistically similar to those described recently for non-glycosylated species [30] (Scheme 4).

Conclusion

In summary, the "hexafluoroacetone route" provides an efficient access to *N*-glycosylated asparagine and its homologues which represent valuable building blocks for the synthesis of therapeutic targets in solution or *via* SPPS. The new protection/activation strategy offers significant advantages: Introduction and cleavage of the protecting group occurs under mild conditions. No anomerization of the glycosylamine moiety can be detected. Furthermore, the new strategy saves steps, since protection and activation and on the other hand peptide bond formation and deprotection of the α-amino group are performed in only one step. Moreover, the progress of the reaction can be monitored easily and quickly by ^{19}F NMR spectroscopy.

Experimental

Solvents were purified and dried prior to use. Reagents were used as purchased. TLC was performed on Silica Gel 60 F_{254} (Merck) with detection by UV light or phosphomolybdic acid/ceric sulphate in 5% aqueous sulfuric acid followed by heating. Flash column chromatography was performed using silica gel (32–63 µm) with solvent systems given in the text. Melting points (uncorrected) were determined on a *Boetius* heating table. Optical rotation indices were measured using a Schmidt &

Haensch Polartronic-D polarimeter in a 5 cm cell. ^{1}H (200 MHz, 300 MHz), ^{13}C (50 MHz, 75 MHz), and ^{19}F (188 MHz, 282 MHz) NMR spectra were recorded on Varian Gemini 200 or Varian Gemini 300 spectrometers. *TMS* was used as reference for ^{1}H and ^{13}C NMR spectra (internal), and CF_3COOH for ^{19}F NMR spectra (external). IR spectra were obtained with a FTIR spectrometer (Genesis ATI Mattson/Unicam). Mass spectra were recorded on a Bruker Daltonics APEX II ESI-FT-ICR spectrometer or on a Finnigan ZAB-HSQ spectrometer (FAB-matrix: 3-*NBA*).

Synthesis of N-Glycosides of Asparagine and Homologues and Their Incorporation into Peptides

Protocol 1: A solution of acid chloride **3a** (1 equiv.) in *DCM* (5 cm^3 per 1 mmol) was cooled to 0°C. Over a period of 30 min a freshly prepared solution of the corresponding *O*-peracetylated *β-D*-glycosylamine (1 equiv.) and *NEM* (1 equiv.) in *DCM* (5 cm^3 per 1 mmol) were added. After vigorous stirring for 1 h at 0°C, *DCM* (20 cm^3 per 1 mmol) was added. The organic phase was extracted with diluted citric acid (10%) and sat. NaCl solution (10 cm^3 per 1 mmol). After drying with $MgSO_4$, the organic solvent was evaporated and the residue purified by flash chromatography or recrystallization.

Protocol 2: A solution of hexafluoroacetone-protected amino acid **2** (1 equiv.) in *DMF* (2.5 mol per 1 mmol) was stirred with 1-[bis(dimethylamino)methylene]-1H-benzotriazolium-tetrafluoroborate-3-oxide (*TBTU*) (1 equiv.) and *NEM* (2 equiv.) for 5 min at room temperature. Then the corresponding *β-D*-glycosylamine was added. The mixture was stirred for another 2 h at room temperature. *DMF* was evaporated *in vacuo* and the residue was redissolved in $CHCl_3$ (50 cm^3 per 1 mmol). The organic phase was washed with citric acid (10%), sat. $NaHCO_3$ solution and sat. NaCl solution (10 cm^3 per 1 mmol). After drying of the organic layer with $MgSO_4$ the solvent was evaporated and compounds **5** were purified by flash chromatography.

Protocol 3: Equimolar amounts of **5** and *DMAP* were dissolved in *DCM* (10 cm^3 per 1 mmol) and stirred at room temperature. The reaction mixture chanced color to brown within a few minutes. The reaction mixture was stirred until TLC indicated complete consumption of the starting material. *DCM* was added (100 cm^3 per 1 mmol), the organic layer was extracted with citric acid (10%, 25 cm^3 per 1 mmol), dried with $MgSO_4$, and evaporated to dryness.

Protocol 4: A suspension of the amino acid *tert*-butylester hydrochloride (1.2 equiv.) in *DMF* (5 cm^3 per 1 mmol) was stirred with 1.2 equiv. *NEM* at room temperature for 5 min. Then 1.0 equiv. of **5** was added at room temperature with stirring. As soon as TLC indicated complete consumption of the starting materials (2–3 d), the volatiles were evaporated *in vacuo*. Then the residue was purified by flash chromatography.

Protocol 5: A solution of the *N*-terminal protected amino acid (1.2 equiv.) in *DMF* (5 cm^3 per 1 mmol) was treated with *TBTU* (1.2 equiv.) and diisopropylethylamine (*DIEA*) (2.4 equiv.). After stirring the reaction mixture for 3 min, a solution of **6** (1.0 equiv.) in *DMF* (2.5 cm^3 per 1 mmol) was added. Afterwards the reaction mixture was stirred for 4 h at room temperature. Work-up for compounds **7** see protocol 2.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-2-[(4S)-5-oxo-2,2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]acetamide (**5a**/**1**, $C_{21}H_{24}F_6N_2O_{12}$)

Ac_4-β-D-Glc-NH_2 (1.48 g, 4.26 mmol) and acid chloride **3** (1.28 g, 4.26 mmol) were reacted due to protocol 1. Purification by flash chromatography (ethyl acetate:petroleum ether = 1:1, $R_f = 0.40$). Yield 2.04 g (78%), colorless needles, mp 155–156°C (*DCM*/petroleum ether); $[\alpha]_D = -2^\circ\ cm^3\ g^{-1}\ dm^{-1}$ ($c = 2.0$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 2.01$, 2.03, 2.04, 2.08 (4s, 4OA*c*), 2.46 (dd, 1H, $J = 9.9$, 16.2 Hz, CH_2^{Asn}), 2.90 (dd, 1H, $J = 2.7$, 16.2 Hz, CH_2^{Asn}), 3.83 (m, 5-H), 3.95

(d, $J = 7.5$ Hz, NH^{HFO}), 4.09 (dd, $J = 2.1$, 12.3 Hz, 6-H_a), 4.29 (dd, $J = 4.8$, 12.3 Hz, 6-H_b), 4.38 (m, α-CH^{Asn}), 4.92 (dd, $J = 9.3$, 9.9 Hz, 2-H), 4.97 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.24 (dd, $J = 9.3$, 9.0 Hz, 1-H), 5.31 (dd, $J = 9.9$, 9.3 Hz, 3-H), 6.65 (d, $J = 9.0$ Hz, 1-NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = -2.76$ (q, 3F, $J = 9.0$ Hz), -2.17 (q, 3F, $J = 9.0$ Hz) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz, HETCOR): $\delta = 20.77$, 20.89, 20.90, 21.06 (4O*Ac*), 39.12 (CH_2^{Asn}), 51.96 (CH^{Asn}), 62.05 (6-CH_2), 68.53 (4-CH), 71.14 (2-CH), 72.86 (3-CH), 74.22 (5-CH), 78.64 (1-CH), 89.01 (sept, $J = 34$ Hz), 120.57 (q, $J = 285$ Hz), 121.57 (q, $J = 290$ Hz), 169.91, 170.14, 170.48, 170.81, 171.21, 171.84 ppm; IR (KBr): $\bar{\nu} = 3446$, 1832, 1751, 1695, 1524 cm^{-1}; MS (FAB): $m/z = 611.1$ $[M + H]^+$.

N-(3,4,6-Tri-O-acetyl-2-acetamido-2-deoxy-β-D-glucopyranosyl)-2-[(4S)-5-oxo-2, 2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]acetamide (**5a**/**2**, $C_{21}H_{25}F_6N_3O_{11}$)

Ac$_3$-*β-D-Glc*N*Ac*-NH_2 (816 mg, 2.36 mmol) and **3** (707 mg, 2.36 mmol) were reacted due to protocol 1. Purification by flash chromatography (ethyl acetate, $R_f = 0.41$). Yield 1.16 g (81%), crystalline solid, mp 196–197°C; $[\alpha]_D = -14°$ cm^3 g^{-1} dm^{-1} ($c = 1.0$, *DCM*); ^{1}H-NMR (*DMSO*-d_6, 300 MHz, COSY): $\delta = 1.75$ (s, 3H, NH*Ac*), 1.92, 1.98, 2.01 (3s, 3O*Ac*), 2.55 (dd, 1H, $J = 5.1$, 16.0 Hz, CH_2^{Asn}), 2.69 (dd, 1H, $J = 3.3$, 16.0 Hz, CH_2^{Asn}), 3.83–3.97 (m, 2,5-H, 6-H_a), 4.18 (dd, $J = 4.2$, 12.6 Hz, 6-H_b), 4.32 (m, α-CH^{Asn}), 4.82 (dd, $J = 9.3$, 9.6 Hz, 4-H), 5.13 (dd, $J = 9.3$, 9.6, 3-H), 5.15 (dd, $J = 9.3$, 9.3 Hz, 1-H), 5.93 (d, $J = 6.6$ Hz, NH^{HFO}), 7.91 (d, 1H, $J = 9.3$ Hz, NH*Ac*), 8.73 (d, $J = 9.3$ Hz, 1-NH) ppm; ^{19}F NMR (*DMSO*-d_6, 188 MHz): $\delta = -4.08$ (q, 3F, $J = 9.0$ Hz), -3.46 (q, 3F, $J = 9.0$ Hz) ppm; ^{13}C NMR (*DMSO*-d_6, 75 MHz, HETCOR): $\delta = 20.38$. 20.42, 20.47 (3O*Ac*), 22.52 (NH*Ac*), 37.04 (CH_2^{Asn}), 50.97 (α-CH^{Asn}), 52.32 (2-CH), 61.96 (6-CH_2), 68.45 (4-CH), 72.36 (5-CH), 73.30 (3-CH), 78.16 (1-CH), 88.09 (sept, $J = 34$ Hz), 120.23 (q, $J = 287$ Hz), 121.41 (q, $J = 291$ Hz), 168.29, 169.34, 169.56, 169.59, 170.07, 171.74 ppm; IR (KBr): $\bar{\nu} = 3500$–3350, 1830, 1745, 1668, 1544 cm^{-1}; MS (FAB): $m/z = 610.1$ $[M + H]^+$.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-3-[(4S)-5-oxo-2,2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]propanoylamide (**5b**/**1**, $C_{22}H_{26}F_6N_2O_{12}$)

Compound **2b** (1.20 g, 4.07 mmol) and *Ac*$_4$-*β-D-Glc*-NH_2 (1.41 g, 4.07 mmol) were reacted due to protocol 2. Purification by flash chromatography (ethyl acetate/petroleum ether, $R_f = 0.31$). Yield 1.85 g (73%), foam; $[\alpha]_D = +6°$ cm^3 g^{-1} dm^{-1} ($c = 2.4$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz): $\delta = 2.02$, 2.03, 2.04, 2.08 (4s, 4O*Ac*), 2.04–2.43 (m, β,γ-CH_2^{Gln}), 3.82 (ddd, $J = 2.1$, 4.5, 9.9 Hz, 5-H), 3.99 (m, α-CH^{Gln}), 4.08 (dd, $J = 2.1$, 12.3 Hz, 6-H_a), 4.15 (d, $J = 6.6$ Hz, NH^{HFO}), 4.30 (dd, $J = 4.5$, 12.3 Hz, 6-H_b), 4.89 (dd, 1H, $J = 9.6$, 9.6 Hz), 5.05 (dd, 1H, $J = 9.9$, 9.6 Hz), 5.21 (dd, 1H, $J = 9.0$, 9.6 Hz), 5.32 (dd, 1H, $J = 9.3$, 9.6 Hz) (1,2,3,4-H), 6.43 (d, $J = 9.0$ Hz, 1-NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = -2.32$ (q, 3F, $J = 9.0$ Hz), -2.87 (q, 3F, $J = 9.0$ Hz) ppm. ^{13}C NMR ($CDCl_3$, 50 MHz): $\delta = 20.63$ (2O*Ac*), 20.75 (2O*Ac*), 26.96 (β-CH_2^{Gln}), 31.63 (γ-CH_2^{Gln}), 54.45 (α-CH^{Gln}), 61.77 (6-CH_2), 68.27, 70.91, 72.72, 73.73 (2,3,4,5-CH), 78.34 (1-CH), 88.75 (sept, $J = 34$ Hz), 120.31 (q, $J = 287$ Hz), 121.2 (q, $J = 290$ Hz), 169.69, 169.93, 170.73, 171.22, 171.44, 172.59 ppm; IR (KBr): $\bar{\nu} = 3360$, 1828, 1749, 1685, 1539 cm^{-1}; MS (FAB): $m/z = 625.1$ $[M + H]^+$.

N-(2,3,4,6-Tetra-O-acetyl-β-D-galactopyranosyl)-3-[(4S)-5-oxo-2,2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]propanoylamide (**5b**/**2**, $C_{22}H_{26}F_6N_2O_{12}$)

Compound **2b** (2.74 g, 9.28 mmol) and *Ac*$_4$-*β-D-Gal*-NH_2 (3.22 g, 9.28 mmol) were reacted due to protocol 2. Purification by flash chromatography (ethyl acetate:petroleum ether = 2:1, $R_f = 0.41$). Yield 4.51 g (78%), foamy solid; $[\alpha]_D = +16°$ cm^3 g^{-1} dm^{-1} ($c = 2.2$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 1.99$, 2.03, 2.05, 2.14 (4s, 4O*Ac*), 1.95–2.05 (m, 1H, β-CH_2^{Gln}), 2.19–2.28 (m, 1H, β-CH_2^{Gln}), 2.38–2.44 (m, γ-CH_2^{Gln}), 3.97–4.05 (m, α-CH^{Gln}, 5-H), 4.06–4.12 (m, 6-H_2), 4.22 (d,

$J = 6.6$ Hz, NH^{HFO}), 5.05 (dd, $J = 9.0$, 10.2 Hz, 2-H), 5.13 (dd, $J = 3.3$, 10.2 Hz, 3-H), 5.19 (dd, $J = 9.0$, 9.0 Hz, 1-H), 5.43 (dd, $J = 1.2$, 3.3 Hz, 4-H), 6.45 (d, $J = 9.0$ Hz, 1-NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = -2.37$ (q, 3F, $J = 9.0$ Hz), -2.84 (q, 3F, $J = 9.0$ Hz) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz, HETCOR): $\delta = 20.59$ (2O*Ac*), 20.71 (2*Ac*), 27.04 (β-CH_2^{Gln}), 31.72 (γ-CH_2^{Gln}), 54.44 (α-CH^{Gln}), 61.26 (6-CH_2), 67.25 (4-CH), 68.66 (2-CH), 70.83 (3-CH), 72.52 (5-CH), 78.68 (1-CH), 88.75 (sept, $J = 34$ Hz), 120.38 (q, $J = 286$ Hz), 121.24 (q, $J = 289$ Hz), 170.02, 170.24, 170.69, 171.57, 171.76, 172.66 ppm; IR (KBr): $\bar{\nu} = 3427$, 1830, 1749, 1687, 1537 cm^{-1}; MS (FAB): $m/z = 625.1$ $[M + H]^+$.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-4-[(4S)-5-oxo-2,2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]butanoylamide (**5c**/**1**, $C_{23}H_{28}F_6N_2O_{12}$)

Compound **2c** (1.43 g, 4.62 mmol) and *Ac*$_4$-*β-D-Glc*-NH_2 (1.72 g, 4.62 mmol) were reacted due to protocol 2. Purification by flash chromatography (ethyl acetate:petroleum ether = 2:1, $R_f = 0.41$). Yield 2.33 g (79%), foamy solid; $[\alpha]_D = +10°$ $cm^3\,g^{-1}\,dm^{-1}$ ($c = 2.0$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 1.66$–1.95 (m, β,γ-CH_2^{Aad}), 2.01, 2.02, 2.04, 2.06 (4s, 4O*Ac*), 2.27 (t, $J = 6.3$ Hz, δ-CH_2^{Aad}), 3.74 (m, α-CH^{Aad}), 3.81 (ddd, $J = 2.1$, 4.5, 9.9 Hz, 5-H), 3.91 (br.d, NH^{HFO}), 4.07 (dd, $J = 2.1$, 12.6 Hz, 6-H_a), 4.29 (dd, $J = 4.5$, 12.6 Hz, 6-H_b), 4.89 (dd, $J = 9.6$, 9.6 Hz, 2-H), 5.05 (dd, $J = 9.3$, 9.9 Hz, 4-H), 5.24 (dd, $J = 9.3$, 9.3 Hz, 1-H), 5.31 (dd, $J = 9.3$, 9.6 Hz, 3-H), 6.35 (d, $J = 9.3$ Hz, 1-NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = -2.71$–-2.58 (m, 6F) ppm; ^{13}C NMR ($CDCl_3$, 50 MHz): $\delta = 20.46$ (3O*Ac*), 20.61 (γ-CH_2^{Aad}, O*Ac*), 32.06 (β-CH_2^{Aad}), 35.18 (δ-CH_2^{Aad}), 54.38 (α-CH^{Aad}), 61.88 (6-CH_2), 68.46, 70.91, 72.88, 73.67 (2,3,4,5-CH), 78.15 (1-CH), 88.58 (sept, $J = 34$ Hz), 120.32 (q, $J = 286$ Hz), 121.36 (q, $J = 288$ Hz), 169.65, 169.92, 170.68, 170.93, 171.34, 172.81 ppm; IR (KBr): $\bar{\nu} = 3432$, 1830, 1751, 1684, 1535 cm^{-1}; MS (FAB): $m/z = 639.2$ $[M + H]^+$.

N-(3,4,6-Tri-O-acetyl-2-acetamido-2-deoxy-β-D-glucopyranosyl)-4-[(4S)-5-oxo-2,2-bis(trifluoromethyl)-1,3-oxazolidin-4-yl]butanoylamide (**5c**/**2**, $C_{23}H_{29}F_6N_3O_{11}$)

Compound **2c** (869 mg, 2.81 mmol) and *Ac*$_3$-*β-D-Glc*N*Ac*-NH_2 (973 mg, 2.81 mmol) were reacted due to protocol 2. Purification by flash chromatography (ethyl acetate, $R_f = 0.23$). Yield 1.43 g (79%), crystalline solid, mp 166–167°C; $[\alpha]_D = -5°$ $cm^3\,g^{-1}\,dm^{-1}$ ($c = 1.0$, *DCM*); ^{1}H NMR (*DMSO*-d_6, 300 MHz, COSY): $\delta = 1.53$–1.77 (m, β,γ-CH_2^{Aad}), 1.75 (s, NH*Ac*), 1.92, 1.98, 2.01 (3s, 3O*Ac*), 2.16 (t, $J = 6.7$ Hz, δ-CH_2^{Aad}), 3.81 (m, 5-H), 3.86–3.97 (m, 2-H, 6-H_a), 4.13 (m, α-CH^{Aad}), 4.17 (dd, $J = 4.2$, 12.6 Hz, 6-H_b), 4.81 (dd, $J = 9.6$, 9.9 Hz, 4-H), 5.10 (dd, $J = 9.9$, 9.9 Hz, 3-H), 5.17 (dd, $J = 9.9$, 9.3 Hz, 1-H), 6.07 (d, $J = 6.6$ Hz, NH^{HFO}), 7.93 (d, $J = 9.3$ Hz, NH*Ac*), 8.47 (d, $J = 9.3$ Hz, 1-NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = -0.68$ (q, 3F, $J = 9.0$ Hz), -1.98 (q, 3F, $J = 9.0$ Hz) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz, HETCOR): $\delta = 20.35$, 20.38, 20.49 (3O*Ac*), 20.79 (γ-CH_2^{Aad}), 22.46 (NH*Ac*), 32.00 (β-CH_2^{Aad}), 34.55 (δ-CH_2^{Aad}), 52.19 (2-CH), 53.62 (α-CH^{Aad}), 61.91 (6-CH_2), 68.49 (4-CH), 72.32 (5-CH), 73.37 (3-CH), 78.03 (1-CH), 88.23 (sept, $J = 34$ Hz), 120.28 (q, $J = 285$ Hz), 121.15 (q, $J = 289$ Hz), 169.34, 169.50, 169.56, 170.04, 172.04, 172.36 ppm; IR (KBr): $\bar{\nu} = 3305$, 1830, 1747, 1670, 1541 cm^{-1}; MS (FAB): $m/z = 638.2$ $[M + H]^+$.

tert-Butyl N′-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)asparaginylglycinate (H-*Asn*(*Ac*$_4$-*β-D-Glc*)-*Gly*-O^t*Bu*, **6a**, $C_{24}H_{37}N_3O_{13}$)

Compound **5a**/**1** (305 mg, 0.50 mmol) and HCl∗H-*Gly*-O^t*Bu* (101 mg, 0.60 mmol) were reacted due to protocol 4. Reaction time: 2 d. Purification by flash chromatography ($CHCl_3$:*Me*OH = 15:1, $R_f = 0.25$). Yield 126 mg (44%), colorless foam; ^{1}H NMR ($CDCl_3$, 200 MHz): $\delta = 1.46$ (s, 9H, CH_3^{tBu}), 2.00, 2.02, 2.04, 2.06 (4s, 4O*Ac*), 2.49–2.76 (m, CH_2^{Asn}), 3.66 (m, α-CH^{Asn}), 3.79 (m, 5-H), 3.90 (d, $J = 5.4$ Hz, CH_2^{Gly}), 4.06 (dd, $J = 1.8$, 12.3 Hz, 6-H_a), 4.27 (dd, $J = 4.5$, 12.3 Hz, 6-H_b), 4.93 (dd, 1H, $J = 9.6$,

9.6 Hz), 5.03 (dd, 1H, $J = 9.9$, 9.6 Hz), 5.26 (dd, 2H, $J = 9.3$, 9.3 Hz) (1,2,3,4-H), 7.26 (d, $J = 9.0$ Hz, 1-NH), 7.67 (t, $J = 5.4$ Hz, NH^{Gly}) ppm; ^{13}C NMR ($CDCl_3$, 50 MHz): $\delta = 20.70$ (2O*Ac*), 20.80 (2O*Ac*), 28.19 ($CH_3{}^{tBu}$), 40.92 ($CH_2{}^{Asn}$), 42.03 ($CH_2{}^{Gly}$), 52.21 (α-CH^{Asn}), 61.86 (6-CH_2), 68.27, 70.71, 73.11, 73.74 (2,3,4,5-CH) 78.02 (1-CH), 82.45 (C^{tBu}), 169.20, 169.67, 170.08, 170.79, 170.95, 171.80, 173.94 ppm; HRMS (ESI): calcd. for $C_{24}H_{38}N_3O_{13}$ ($M + H^+$) 576.23991, found 576.23922.

tert-Butyl N′-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)glutaminylglycinate
(H-*Gln*(Ac_4-*β-D-Glc*)-*Gly*-O*ᵗBu*, **6b**, $C_{25}H_{39}N_3O_{13}$)

Compound **5b**/**1** (312 mg, 0.50 mmol) and HCl∗H-*Gly*-O*ᵗBu* (101 mg, 0.60 mmol) were reacted due to protocol 4. Reaction time: 2 d. Purification by flash chromatography ($CHCl_3$:*Me*OH = 10:1, $R_f = 0.28$). Yield 123 mg (42%), colorless foam; ^1H NMR ($CDCl_3$, 300 MHz): $\delta = 1.50$ (s, 9H, $CH_3{}^{tBu}$), 2.00, 2.02, 2.03, 2.06 (4s, 4O*Ac*), 2.00–2.35 (m, β,γ-$CH_2{}^{Gln}$), 3.51 (m, α-CH^{Gln}), 3.79 (m, 5-H), 3.88 (dd, 1H, $J = 5.1$, 18.0 Hz, $CH_2{}^{Gly}$), 4.05–4.14 (m, 2H, 6-H_a, $CH_2{}^{Gly}$), 4.27 (dd, $J = 4.5$, 12.3 Hz, 6-H_b), 4.91 (dd, 1H, $J = 9.6$, 9.3 Hz), 4.99 (dd, 1H, $J = 9.9$, 9.6 Hz), 5.27 (dd, 2H, $J = 9.3$, 9.3 Hz), 5.32 (dd, 1H, $J = 9.3$, 9.6 Hz) (1,2,3,4-H), 7.54 (br.t, NH^{Gly}), 7.59 (d, $J = 9.3$ Hz, 1-NH) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz): $\delta = 20.65$ (2O*Ac*), 20.81 (2O*Ac*), 28.15 ($CH_3{}^{tBu}$), 30.54, 32.60 (β,γ-$CH_2{}^{Gln}$), 41.42 ($CH_2{}^{Gly}$), 54.39 (α-CH^{Gln}), 61.78 (6-CH_2), 68.12, 70.67, 73.20, 73.59 (2,3,4,5-CH), 77.82 (1-CH), 82.75 (C^{tBu}), 169.69, 170.00, 170.31, 170.75, 170.85, 174.26, 174.67 ppm; HRMS (ESI): calcd. for $C_{25}H_{40}N_3O_{13}$ ($M + H^+$) 590.25556, found 590.25574.

tert-Butyl N′-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)homoglutaminylglycinate
(H-*hGln*(Ac_4-*β-D-Glc*)-*Gly*-O*ᵗBu*, **6c**, $C_{26}H_{41}N_3O_{13}$)

Compound **5c**/**1** (319 mg, 0.50 mmol) and HCl∗H-*Gly*-O*ᵗBu* (101 mg, 0.60 mmol) were reacted due to protocol 4. Reaction time: 2 d. Purification by flash chromatography ($CHCl_3$:*Me*OH = 20:3, $R_f = 0.25$). Yield 165 mg (55%), colorless foam; ^1H NMR ($CDCl_3$, 300 MHz): $\delta = 1.48$ (s, 9H, $CH_3{}^{tBu}$), 1.52–1.90 (m, γ-$CH_2{}^{Aad}$, β-$CH_2{}^{Aad}$), 2.00, 2.01, 2.03, 2.06 (4s, 4O*Ac*), 2.17–2.26 (m, δ-$CH_2{}^{Aad}$), 3.42 (m, α-CH^{Aad}), 3.79 (m, 5-H), 3.92 (d, $J = 5.7$ Hz, $CH_2{}^{Gly}$), 4.06 (dd, $J = 1.8$, 12.3 Hz, 6-H_a), 4.28 (dd, $J = 4.8$, 12.3 Hz, 6-H_b), 4.92 (dd, 1H, $J = 9.6$, 9.3 Hz), 5.03 (dd, 1H, $J = 9.9$, 9.6 Hz), 5.24 (dd, 1H, $J = 9.3$, 9.3 Hz), 5.28 (dd, 1H, $J = 9.6$, 9.3 Hz) (1,2,3,4-H), 6.69 (d, $J = 9.3$ Hz, 1-NH), 7.71 (t, $J = 5.7$ Hz, NH^{Gly}) ppm; ^{13}C NMR ($CDCl_3$, 50 MHz): $\delta = 20.52$ (2O*Ac*), 20.66 (2O*Ac*), 21.08 (γ-$CH_2{}^{Aad}$), 28.08 ($CH_3{}^{tBu}$), 34.15, 35.85 (β,δ-$CH_2{}^{Aad}$), 41.73 ($CH_2{}^{Gly}$), 54.58 (α-CH^{Aad}), 61.86 (6-CH_2), 68.35, 70.74, 73.10, 73.60 (2,3,4,5-CH), 78.07 (1-CH), 82.13 (C^{tBu}), 169.17, 169.52, 169.79, 170.52, 170.61, 173.24, 175.00 ppm; HRMS (ESI): calcd. for $C_{26}H_{42}N_3O_{13}$ ($M + H^+$) 604.27121, found 604.27053.

tert-Butyl N-(Benzyloxycarbonyl)phenylalanyl-N′-
(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl) asparaginylglycinate
(*Z-Phe-Asn*(Ac_4-*β-D-Glc*)-*Gly*-O*ᵗBu*, **7a**, $C_{41}H_{52}N_4O_{16}$)

Compound **6a** (113 mg, 0.20 mmol) and *Z-Phe*-OH (71 mg, 0.24 mmol) were reacted due to protocol 5. Purification by flash chromatography (ethyl acetate, $R_f = 0.35$). Yield 134 mg (78%), foam; $[\alpha]_D = +10^\circ\,cm^3\,g^{-1}\,dm^{-1}$ ($c = 1.3$, *DCM*); ^1H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 1.45$ (s, 9H, $CH_3{}^{tBu}$), 2.01, 2.02, 2.06, 2.11 (4s, 4O*Ac*), 2.43 (m, 1H, $CH_2{}^{Asn}$), 2.74 (m, 1H, $CH_2{}^{Asn}$), 3.02–3.16 (m, $CH_2{}^{Phe}$), 3.68–3.81 (m, $CH_2{}^{Gly}$, 5-H), 4.06 (dd, $J = 1.8$, 12.3 Hz, 6-H_a), 4.27 (dd, $J = 4.5$, 12.3 Hz, 6-H_b), 4.43 (m, α-CH^{Phe}), 4.76 (m, α-CH^{Asn}), 4.95 (dd, $J = 9.6$, 9.6 Hz, 2-H), 5.06 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.08 (s, $CH_2{}^{Z}$), 5.20 (dd, $J = 9.3$, 9.3 Hz, 1-H), 5.28 (dd, $J = 9.3$, 9.6 Hz, 3-H), 5.31 (d, $J = 6.9$ Hz, NH^{Phe}), 6.78 (d, $J = 9.0$ Hz, 1-NH), 6.77 (br.t, NH^{Gly}), 7.17–7.38 (m, 10H, arom), 7.67 (d, $J = 8.4$ Hz, NH^{Asn}) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz, APT): $\delta = 20.65$ (2O*Ac*), 20.78 (2O*Ac*), 28.09 ($CH_3{}^{tBu}$), 36.58 ($CH_2{}^{Asn}$), 38.23 ($CH_2{}^{Phe}$), 42.14 ($CH_2{}^{Gly}$), 49.50 (α-CH^{Asn}), 56.53 (α-CH^{Phe}), 61.69 (6-CH_2),

67.21 (CH_2^Z), 68.14 (4-CH), 70.29 (2-CH), 72.80 (3-CH), 73.66 (5-CH), 78.00 (1-CH), 82.12 (C^{tBu}), 127.16, 128.10, 128.26, 128.60, 128.85, 129.33, 136.18, 136.22, 156.21, 168.34, 169.59, 169.99, 170.15, 170.71, 171.31, 171.46, 172.13 ppm; IR (KBr): $\bar{\nu} = 1744$, 1680, 1538, 1512 cm^{-1}; HRMS (ESI): calcd. for $C_{41}H_{53}N_4O_{16}$ ($M + H^+$) 857.34511, found 857.34626.

tert-Butyl N-(Benzyloxycarbonyl)phenylalanyl-N′-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl) glutaminylglycinate (*Z-Phe-Gln*(Ac_4-*β-D-Glc*)-*Gly*-O*t Bu*, **7b**, $C_{42}H_{54}N_4O_{16}$)

Compound **6b** (92 mg, 0.16 mmol) and *Z-Phe*-OH (56 mg, 0.19 mmol) were reacted due to protocol 5. Purification by flash chromatography (ethyl acetate, $R_f = 0.30$). Yield 86 mg (63%), colorless foam; $[\alpha]_D = -2^\circ\ cm^3\ g^{-1}\ dm^{-1}$ ($c = 1.3$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 1.48$ (s, 9H, CH_3^{tBu}), 2.01, 2.03, 2.04, 2.07 (4s, 4O*Ac*), 1.95–2.18 (m, β,γ-CH_2^{Gln}), 3.05–3.09 (m, CH_2^{Phe}), 3.76 (m, 5-H), 3.78 (dd, 1H, $J = 5.1$, 18.0 Hz, CH_2^{Gly}), 3.99 (dd, 1H, $J = 6.0$, 18.0 Hz, CH_2^{Gly}), 4.08 (dd, $J = 1.8$, 12.3 Hz, 6-H_a), 4.27 (dd, $J = 4.8$, 12.3 Hz, 6-H_b), 4.36–4.46 (m, α-CH^{Gln}, α-CH^{Phe}), 4.95 (dd, $J = 9.6$, 9.3 Hz, 2-H), 5.03 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.07 (s, CH_2^Z), 5.26 (dd, $J = 9.3$, 9.3 Hz, 1,3-H), 5.38 (d, $J = 7.2$ Hz, NH^{Phe}), 6.85 (d, $J = 7.2$ Hz, NH^{Gln}), 7.04 (br.t, NH^{Gly}), 7.14–7.36 (m, 11H, 1-NH, arom) ppm; ^{13}C NMR ($CDCl_3$, 50 MHz, APT, HETCOR): $\delta = 20.69$ (2O*Ac*), 20.82 (2O*Ac*), 28.21 (CH_3^{tBu}), 28.53 (β-CH_2^{Gln}), 32.41 (γ-CH_2^{Gln}), 38.49 (CH_2^{Phe}), 42.08 (CH_2^{Gly}), 52.29 (α-CH^{Gln}), 56.42 (α-CH^{Phe}), 61.83 (6-CH_2), 67.34 (CH_2^Z), 68.28 (4-CH), 71.09 (2-CH), 73.09 (3-CH), 73.85 (5-CH), 78.43 (1-CH), 82.53 (C^{tBu}), 127.15, 128.23, 128.38, 128.68, 128.82, 129.49, 136.31, 156.25, 169.01, 169.62, 169.93, 170.71, 170.86, 171.16, 171.43, 173.41 ppm; IR (KBr): $\bar{\nu} = 1749$, 1672, 1536, 1512 cm^{-1}; HRMS (ESI): calcd. for $C_{42}H_{55}N_4O_{16}$ ($M + H^+$) 871.36075, found 871.36021.

tert-Butyl N-(Benzyloxycarbonyl)phenylalanyl-N′-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl) homoglutaminylglycinate (*Z-Phe-hGln*(Ac_4-*β-D-Glc*)-*Gly*-O*t Bu*, **7c**, $C_{43}H_{56}N_4O_{16}$)

Compound **6c** (152 mg, 0.25 mmol) and *Z-Phe*-OH (90 mg, 0.30 mmol) were reacted due to protocol 5. Purification by flash chromatography (ethyl acetate, $R_f = 0.32$). Yield 149 mg (68%), colorless foam; $[\alpha]_D = -6^\circ\ cm^3\ g^{-1}\ dm^{-1}$ ($c = 1.3$, *DCM*); ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 1.51$ (s, 9H, CH_3^{tBu}), 1.51–1.58 (m, 3H, γ-CH_2^{Aad}, β-CH_2^{Aad}), 1.85–1.97 (m, β-CH_2^{Aad}), 2.03, 2.04 (2s, 2O*Ac*), 2.07 (s, 2O*Ac*), 2.15–2.20 (m, δ-CH_2^{Aad}), 3.10–3.14 (m, CH_2^{Phe}), 3.79 (m, 5-H), 3.88 (d, $J = 5.1$ Hz, CH_2^{Gly}), 4.04 (dd, $J = 1.8$, 12.3 Hz, 6-H_a), 4.36 (dd, $J = 4.8$, 12.3 Hz, 6-H_b), 4.47–4.53 (m, α-CH^{Aad}, α-CH^{Phe}), 4.98 (dd, $J = 9.6$, 9.3 Hz, 2-H), 5.06 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.12 (s, CH_2^Z), 5.28 (dd, $J = 9.3$, 9.3 Hz, 1-H), 5.31 (dd, $J = 9.6$, 9.3 Hz, 3-H), 5.57 (d, $J = 7.2$ Hz, NH^{Phe}), 6.74 (t, $J = 5.1$ Hz, NH^{Gly}), 6.83 (d, $J = 9.0$ Hz, 1-NH), 6.95 (d, $J = 7.2$ Hz, NH^{Aad}), 7.21–7.38 (m, 10H, arom) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz, HETCOR): $\delta = 20.67$ (2O*Ac*), 20.73 (2O*Ac*), 20.80 (γ-CH_2^{Aad}), 28.12 (CH_3^{tBu}), 31.29 (β-CH_2^{Aad}), 35.51 (δ-CH_2^{Aad}), 38.42 (CH_2^{Phe}), 42.04 (CH_2^{Gly}), 52.57 (α-CH^{Aad}), 56.28 (α-CH^{Phe}), 61.80 (6-CH_2), 67.18 (CH_2^Z), 68.27 (4-CH), 70.70 (2-CH), 73.09 (3-CH), 73.73 (5-CH), 78.10 (1-CH), 82.37 (C^{tBu}), 127.05, 128.14, 128.28, 128.61, 128.74, 129.41, 136.35, 156.30, 168.89, 169.67, 169.99, 170.84 (2C), 171.14, 171.50, 173.48 ppm; IR (KBr): $\bar{\nu} = 1741$, 1665, 1653, 1535 cm^{-1}; HRMS (ESI): calcd for $C_{43}H_{57}N_4O_{16}$ ($M + H^+$) 885.37641, found 885.37816.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-3-(2,2,2-trifluoro-1-trifluoromethylethylamino) acrylamide (**8**, $C_{20}H_{24}F_6N_2O_{10}$)

Compound **5a**/**1** (254 mg, 0.42 mmol) and *DMAP* (51 mg, 0.42 mmol) were reacted due to protocol 3. Reaction time: 1 h. Yield 130 mg (55%), foamy solid, 90% purity by NMR and DC analysis (ethyl acetate:petroleum ether = 2:3, $R_f = 0.28$). Decomposition during flash column chromatography on

silica gel. The spectroscopic data were taken from the crude product. ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 2.02$, 2.03, 2.05, 2.07 (4s, 4O*Ac*), 3.83 (m, 5-H), 3.99 (m, $CH(CF_3)_2$), 4.08 (dd, $J = 2.1$, 12.6 Hz, 6-H_a), 4.33 (dd, $J = 4.2$, 12.6 Hz, 6-H_b), 4.63 (d, $J = 8.4$ Hz, =CH), 4.94 (dd, $J = 9.3$, 9.6 Hz, 2-H), 5.04 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.24 (dd, $J = 9.3$, 9.3 Hz, 1-H), 5.31 (dd, $J = 9.3$, 9.6 Hz, 3-H), 6.03 (d, $J = 9.3$ Hz, 1-NH), 6.45 (dd, $J = 8.4$, 11.1 Hz, =CH), 8.90 (t, $J = 11.1$ Hz, NH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = 5.22$ (m, 6F) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz): $\delta = 20.60$, 20.62, 20.69, 20.74 (4O*Ac*), 61.65 (6-CH_2), 61.65 (sept, $J = 32.0$ Hz, $CH(CF_3)_2$), 68.18, 70.63, 72.90, 73.44 (2,3,4,5-CH), 78.22 (1-CH), 91.12 (=CH), 121.59 (q, $J = 284$ Hz, CF_3), 148.34 (=CH), 169.72, 169.80, 170.03, 170.88, 171.24 ppm; IR (KBr): $\bar{\nu} = 3450$, 1730, 1659, 1623, 1540 cm^{-1}; MS (FAB): $m/z = 567.1$ $[M + H]^+$.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-4-(2,2,2-trifluoro-1-trifluoromethylethylimino) butyramide (**9**, $C_{21}H_{26}F_6N_2O_{10}$)

Compound **5b**/**1** (312 mg, 0.5 mmol) and *DMAP* (61 mg, 0.5 mmol) were reacted due to protocol 3. Purification by flash chromatography (ethyl acetate:petroleum ether = 3:2, $R_f = 0.25$). Yield 110 mg (38%), foamy solid; ^{1}H NMR ($CDCl_3$, 300 MHz, COSY): $\delta = 2.02$, 2.03, 2.04, 2.07 (4s, 4O*Ac*), 2.57 (t, $J = 6.6$ Hz, β-CH_2), 2.73–2.80 (m, α-CH_2), 3.81 (m, 5-H), 4.00–4.09 (m, $CH(CF_3)_2$, 6-H_a), 4.33 (dd, $J = 4.2$, 12.6 Hz, 6-H_b), 4.90 (dd, $J = 9.6$, 9.6 Hz, 2-H), 5.06 (dd, $J = 9.9$, 9.6 Hz, 4-H), 5.21 (dd, $J =$ 9.3, 9.3 Hz, 1-H), 5.30 (dd, $J = 9.6$, 9.6 Hz, 3-H), 6.40 (d, $J = 9.3$ Hz, 1-NH), 8.05 (s, 1H, γ-CH) ppm; ^{19}F NMR ($CDCl_3$, 282 MHz): $\delta = 6.55$ (d, 6F, $J = 6.0$ Hz) ppm; ^{13}C NMR ($CDCl_3$, 75 MHz): $\delta = 20.65$ (2O*Ac*), 20.74, 20.80 (2O*Ac*), 28.47, 30.90 (α,β-CH_2), 61.73 (6-CH_2), 71.66 (sept, $J = 30.0$ Hz, $CH(CF_3)_2$), 68.19, 70.55, 72.74, 73.60 (2,3,4,5-CH), 78.28 (1-CH), 121.10 (q, $J = 282$ Hz, CF_3), 169.57, 169.88, 170.62, 171.04, 171.75 (C=O), 175.84 (CH=N) ppm; IR (KBr): $\bar{\nu} = 3400$, 1755, 1700–1669, 1535 cm^{-1}; MS (FAB): $m/z = 581.2$ $[M + H]^+$.

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