Chemoselective Removal of Protecting Groups from *O*-Glycosyl Amino Acid and Peptide (Methoxyethoxy)ethyl Esters Using Lipases and Papain

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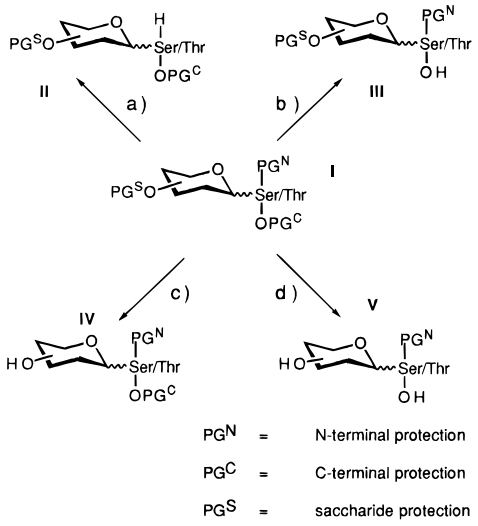
The selective C-terminal deprotection of *O*-glycopeptide (methoxyethoxy)ethyl esters is achieved under mild conditions (pH 6.6, 37 °C) by enzymatic hydrolysis using papain or lipase M from *Mucor javanicus* to give building blocks useful for chain-extending glycopeptide synthesis. On the other hand, the selective removal of acetyl protecting groups from the saccharide portion of glycopeptides is accomplished by alternative enzymatic hydrolysis with lipase WG from wheat germ to furnish model substrates for enzymatic glycosyl transfer reactions in order to extend the carbohydrate side chain of these conjugates.

Introduction

Glycoproteins exhibit important biological functions. For example, glycoproteins are involved in cell−cell communication during cell growth and have also been identified as tumor-associated antigens.[1] Glycopeptides representing partial structures of glycoproteins are receiving increasing interest as model compounds for biological recognition processes. In contrast to peptides and proteins, they are not easily accessible in pure form from biological sources or gene-technological procedures. For their chemical synthesis, the glycosidic bonds have to be formed stereoselectively, and numerous functional groups have to be protected and deprotected chemoselectively. Additional difficulties arise in the preparation of glycopeptides containing an *O*-glycosidic bond between serine or threonine and the carbohydrate. Under acidic conditions anomerization or even cleavage of the glycosidic bond may occur.[2] On the other hand, at pH values > 11 the entire carbohydrate part may be lost through β-elimination.[3] Therefore, the synthesis of these complex molecules requires protecting groups that can be selectively removed under mild conditions.[4] Enzymatic hydrolysis offers an advantageous alternative to chemical deprotection methods.[5] The mild and neutral reaction conditions typical for the enzymatic removal of protecting groups afford reliable access to selectively deblocked products. Cleavage of the carboxy protecting group leads to compounds which can be used for peptide chain extension (Scheme 1, paths b and d). Removal of the protecting groups of the saccharide portion yields substrates for enzymatic glycosylations using glycosyltransferases or glycosidases (Scheme 1, paths c and d).

Scheme 1



In this paper we report on the realization of these alternative deprotections of glycopeptides: The polar solubilizing (methoxyethoxy)ethyl (MEE) esters[6] of *O*-glycosyl amino acids and *O*-glycopeptides are hydrolyzed using papain or lipase M from *Mucor javanicus*, respectively, as the catalyst. Alternatively, the *O*-acetyl groups of the saccharide portions of glycopeptides are selectively hydrolyzed by application of lipase WG from wheat germ.

Results and Discussion

Synthesis of the Serine and Threonine Glycosides. The following model compounds for enzymatic hydrolysis were synthesized: β-D-xylosylserine derivatives, which represent linkage structures of proteoglycans, β-D-galactosylserine and -threonine conjugates being substructures of collagen-type *O*-glycoproteins, and α-(*N*-acetylgalactosaminyl)serine and -threonine derivatives, representing partial structures of mucin-type glycoproteins and tumor-associated T_N-antigen structures.[4,7]

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(1) For a recent review, see: Lis, H.; Sharon, N. *Eur. J. Biochem.* **1993**, *218*, 1.

(2) Garg, H. G.; Jeanloz, R. W. *Carbohydr. Res.* **1976**, *49*, 482.

(3) (a) Vercellotti, J. R.; Fernandez, R.; Chang, C. J. *Carbohydr. Res.* **1967**, *5*, 97. (b) Wakabayashi, K.; Pigman, W. *Ibid.* **1974**, *35*, 3. (c) Marshall, R. D.; Neuberger A. *Adv. Carbohydr. Chem. Biochem.* **1970**, *25*, 407.

(4) (a) Kunz, H. *Angew. Chem.* **1987**, *99*, 297; *Angew. Chem., Int. Ed. Engl.* **1987**, *26*, 294. (b) Garg, H. G.; von dem Bruch, K.; Kunz, H. *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 277.

(5) (a) Reidel, A.; Waldmann, H. *J. Prakt. Chem.* **1993**, *335*, 109. (b) Glass, J. D. In *The Peptides*; Undenfried, S., Meienhofer, J., Eds.; Academic Press: San Diego, 1987; Vol 9, p 167. (c) Herrmann, P. *Wiss. Z. Univ. Halle* **1987**, *36*, 17. (d) Review: Whitesides, G. M.; Wong, C.-H. *Angew. Chem., Int. Ed. Engl.* **1985**, *24*, 617.

(6) Kunz, H.; Kowalczyk, D.; Braun, P.; Braum G. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 336.

(7) Springer, G. F. *Science* **1984**, *224*, 1198.

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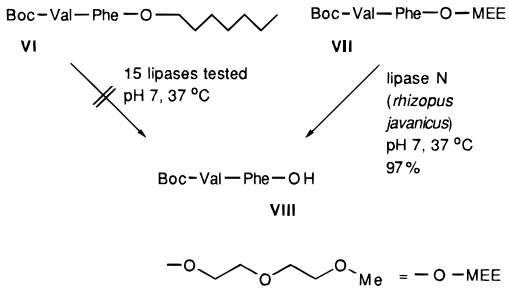
Scheme 2. Peptide MEE Ester Are Superior Substrates to Lipases Compared to the Corresponding Heptyl Esters



N-Protected serine or threonine (methoxyethoxy)ethyl esters (MEE) **3** were used as glycosyl acceptors. The hydrophilic MEE esters show a higher rate of hydrolysis and are superior substrates for lipases in comparison to the hydrophobic heptyl esters.[8] These advantages are illustrated by comparing the N-protected dipeptides **VI** and **VII**. While the dipeptide heptyl ester **VI** is not hydrolyzed by any out of the 15 lipases tested,[8] the corresponding MEE ester **VII** is readily cleaved under identical conditions to give the selectively deprotected dipeptide **VIII** in almost quantitative yield[6] (Scheme 2). The superior reactivity of MEE esters is generally observed, but their improved lipase-catalyzed hydrolysis is, in particular, found for esters of hydrophobic peptides. In addition, MEE esters of peptides with C-terminal proline can be hydrolyzed using lipases (lipase CE from *Humicola lanuginosa*),[6] whereas heptyl esters of proline peptides were not accepted as substrates.[8] Presumably, the advantageous properties of the MEE esters are mainly caused by their polar nature increasing the solubility or, at least, wettability in aqueous solutions. Due to the hydrophilic nature of the MEE esters, the structure of the peptide derivative obviously has less influence on the lipase-catalyzed hydrolysis. This is also demonstrated by comparing the *N*-acetylgalactosylamine threonine conjugates **IX** and **X**. Whereas the heptyl ester **IX** was not hydrolyzed by any of the 15 lipases investigated, the analogous MEE ester **X** was converted, if even slowly, into the carboxy-deblocked galactosaminyl threonine synthon **XI** (Scheme 3). Analogous behavior was also found for corresponding serine conjugates,[6] and again underlines the superior substrate properties of the polar MEE esters. MEE esters of amino acids are readily accessible by azeotropic esterification of serine or threonine with diethylene glycol monomethyl ether. Subsequent acylation of the amino group with allyl chloroformate (Aloc-Cl),[9] benzyl chloroformate (Z-Cl), or 9-fluorenylmethyl chloroformate (Fmoc-Cl),[10] respectively, gave the required glycosyl acceptors **3** (Table 1).

Since the Aloc group is sensitive to soft electrophiles,[11] the glycosylation of the Aloc-protected serine ester **3e** was carried out by silver ion-promoted activation of the 2-azidogalactosyl bromide.[12] The glycosylation of Fmoc-Ser-OMEE (**3a**), Fmoc-Thr-OMEE (**3b**), and Z-Thr-OMEE (**3d**) was achieved by activation of the thioglycoside **5** with dimethylthio(methylthio)sulfonium triflate (DMTST) (Table 2). In both glycosylation reactions a mixture of anomers was formed, from which the desired α-anomer was separated by flash chromatography.

Scheme 3. In Contrast to Heptyl Esters, MEE Esters of *N*-Acetylgalatoamine Amino Acid Conjugates Can Be Hydrolyzed Using Lipases

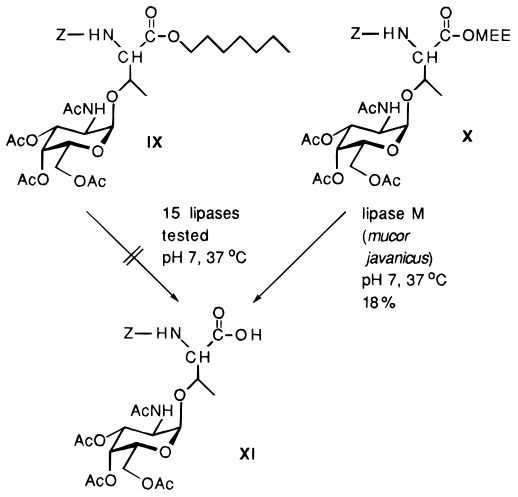
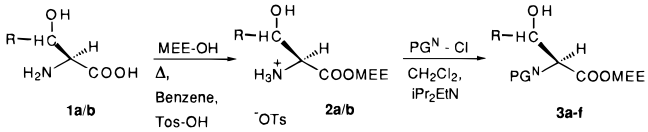


Table 1. Synthesis of N-Protected Serine and Threonine MEE Esters



1a R = H
1b R = CH_3

2a R = H
2b R = CH_3

	R	PG^N	yield (%)
3a	H	Fmoc	61
3b	CH_3	Fmoc	64
3c	H	Z	60
3d	CH_3	Z	60
3e	H	Aloc	79
3f	CH_3	Aloc	77

The azido galactosides **6a**−**d** were transformed to the *N*-acetylgalactosamine conjugates **7a**−**d** (T_N-antigen) by applying thioacetic acid[13] (Table 3). Model compounds carrying *β*-galactose, *β*-glucose, and *β*-xylose as the carbohydrate part were synthesized by reacting the per-*O*-acetylated glycosides **8**−**10** with *N*-Aloc-protected glycosyl acceptors **3e/f** and trimethylsilyl trifluoromethanesulfonate (TMS-triflate) as the promotor[14] (Table 4). Exclusively *β*-configurated products **11a**−**f** were obtained.

Selective Removal of the MEE Ester Group by Hydrolysis with Papain. Ishii et al.[15a] described that papain can be used to selectively cleave the methyl ester of N-glycosylated asparagine methyl esters. In 1991, Cantacuzène et al.[15b] reported the use of papain for

(8) (a) Braun, P.; Waldmann, H.; Vogt, W.; Kunz, H. *Liebigs Ann. Chem.* **1991**, 165. (b) Braun, P.; Waldmann, H.; Vogt, W.; Kunz, H. *Synlett* **1990**, 105. (c) Braun, P.; Waldmann, H.; Kunz, H. *Synlett* **1992**, 39. (d) Braun, P.; Waldmann, H.; Kunz, H. *BioMed. Chem. Lett.* **1993**, *1*, 197.

(9) Kunz, H.; Unverzagt, C. *Angew. Chem., Int. Ed. Engl.* **1984**, *23*, 436.

(10) Carpino, L. A.; Han, G. Y. *J. Am. Chem. Soc.* **1970**, *92*, 5748.

(11) (a) Kunz, H.; Wernig, P.; Schultz, M. *Synlett* **1990**, 631. (b) Braun, P. Dissertation, Universität Mainz, Germany, 1993, 37.

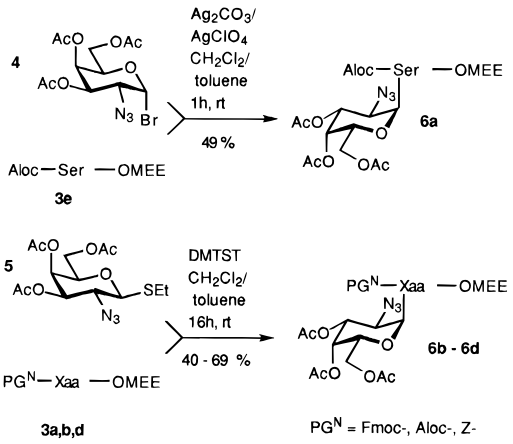
(12) (a) Wulff, G.; Röhle, G. *Angew. Chem., Int. Ed. Engl.* **1974**, *13*, 157. (b) Paulsen, H. *Chem. Soc. Rev.* **1984**, *13*, 15. (c) Paulsen, H; Hölck J.-P. *Carbohydr. Res.* **1982**, *109*, 89.

(13) Rosen, T.; Lico, I. M.; Chu, D. T. W. *J. Org. Chem.* **1988**, *53*, 1580.

(14) Ogawa, T.; Beppu, K.; Nakabayashi, S. *Carbohydr. Res.* **1981**, *93*, C6.

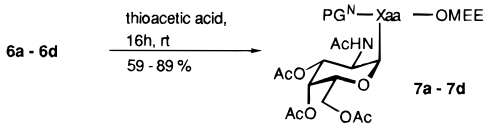
(15) (a) Ishii, H; Funabashi, K.; Mimura, Y.; Inoue, Y. *Bull. Chem. Soc. Jpn.* **1990**, *63*, 3042. (b) Cantacuzène, D.; Attal, S.; Bay, S. *BioMed. Chem. Lett.* **1991**, 197. (c) Attal, S.; Bay, S.; Cantacuzène, D. *Tetrahedron* **1992**, *48(42)*, 9251.

Table 2. Synthesis of the Azidogalactosyl Compounds



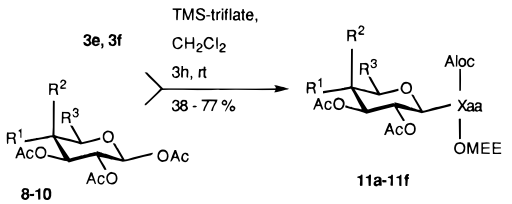
	Xaa	PG^N	yield (%)
6b	Ser	Fmoc	69
6c	Thr	Z	45
6d	Thr	Fmoc	40

Table 3. Synthesis of the *N*-Acetylgalactosamine Conjugates 7a–d



	PG^N	Xaa	yield (%)
7a	Aloc	Ser	59
7b	Fmoc	Ser	77
7c	Z	Thr	89
7d	Fmoc	Thr	75

Table 4. Synthesis of the Saccharide Amino Acid Conjugates 11a–f



8 : Ac_5Glc

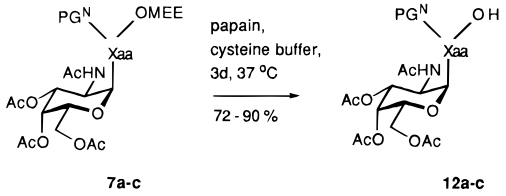
9 : Ac_5Gal

10 : Ac_4Xyl

	Xaa	R^1	R^2	R^3	yield (%)
11a	Ser	OAc	H	CH_2OAc	38
11b	Thr	OAc	H	CH_2OAc	44
11c	Ser	H	OAc	CH_2OAc	52
11d	Thr	H	OAc	CH_2OAc	69
11e	Ser	OAc	H	H	77
11f	Thr	OAc	H	H	73

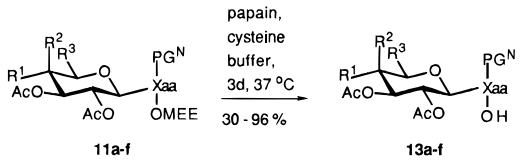
C-terminal deprotection of *O*-galactosylated Aloc-Ser-OMe. In a series of experiments, we could demonstrate that glycosylated amino acid MEE esters **7a–c** and **11a–f** are hydrolyzed by papain under mild reaction conditions (pH = 6.6, T = 37 °C) with complete selectivity (Tables 5 and 6). Neither the *O*-acetyl groups of the carbohydrate portions nor the amino protecting groups or the α- (**7a–c**) or β-glycosidic bonds (**11a–f**) were affected. As a rule, the selectively deblocked products **12a–c** or **13a–f**, respectively, were isolated in high yield.

Table 5. Removal of the MEE Ester Protecting Group from α-Glycosidic Conjugates with Papain



	Xaa	PG^N	yield (%)
12a	Ser	Aloc	75
12b	Ser	Fmoc	90
12c	Thr	Z	72

Table 6. Removal of the MEE Ester Protecting Group from β-Glycosidic Conjugates with Papain



	Xaa	PG^N	R^1	R^2	R^3	yield (%)
13a	Ser	Aloc	OAc	H	CH_2OAc	96
13b	Thr	Aloc	OAc	H	CH_2OAc	30[a]
13c	Ser	Aloc	H	OAc	CH_2OAc	82
13d	Thr	Aloc	H	OAc	CH_2OAc	87
13e	Ser	Aloc	OAc	H	H	90
13f	Thr	Aloc	OAc	H	H	90

[a] Difficult chromatography.

These results show that the enzymatic cleavage of glycosyl amino acid esters using papain tolerates quite different carbohydrate residues in such glycoconjugates and should therefore be generally applicable for the synthesis of N-protected glycosyl amino acid building blocks valuable for versatile sythesis of glycopeptides either in solution or on solid phase.

Selective Removal of the MEE Ester Group from Glycopeptides with Lipase M. Because papain cleaves peptide bonds, the enzymatic hydrolysis of glycopeptide MEE esters requires a biocatalyst which does not exhibit proteolytic activity. The deprotection of glycopeptide MEE esters was efficiently achieved with lipase M (Amano). To obtain a model substrate, dipeptide **14** was deprotected at the N-terminus using HCl/ether and, subsequently, condensed with the *O*-glycosyl amino acid **12a** using *O*-[(cyano(ethoxycarbonyl)methylene)amino]-*N*,*N*,*N*′,*N*′-tetramethyluronium tetrafluoroborate (TOTU) as the coupling agent[16] (Scheme 4). Cleavage of the MEE ester in compound **15** with lipase M (Amano) was carried out in phosphate buffer at pH = 7.0 and 37 °C. Product **16** was isolated in pure form (Scheme 4). The other protecting groups, the glycosidic bond, and the peptide bonds of **16** remained unaffected.

Selective Removal of the *O*-Acetyl Protecting Groups from the Carbohydrate with Lipase WG from Wheat Germ. For the selective removal of the protecting groups of the saccharide portions of glycopeptides 15 lipases were tested. Lipase WG (Sigma) from wheat germ accepted glycosyl amino acid esters **7a,b/11a–f** as substrates and was able to cleave the *O*-acetyl bonds in the saccharide domain (**17a,b**, **18a–f**). After

(16) Breipohl, G.; König, W. (Höchst AG); European Patent, EP-A 0460446, A1, 23 May 1990.

Scheme 4

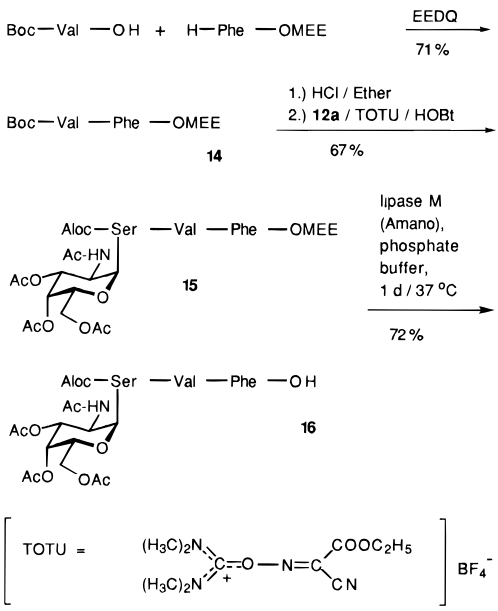
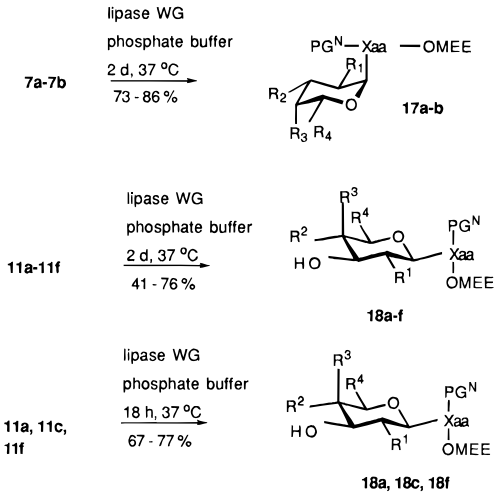


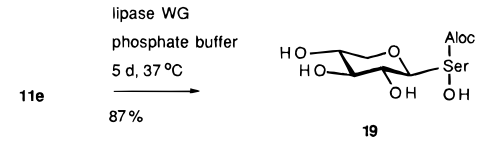
Table 7. Removal of the Saccharide Protecting Groups



	Xaa	PG^N	R^1	R^2	R^3	R^4	yield (%) reaction time 2 d	18 h
17a	Ser	Aloc	NHAc	OH	OH	CH_2OH	73	
17b	Ser	Fmoc	NHAc	OH	OH	CH_2OH	86	
18a	Ser	Aloc	OH	OH	H	CH_2OH	48	77
18b	Thr	Aloc	OH	OH	H	CH_2OH	76	
18c	Ser	Aloc	OH	H	OH	CH_2OH	41	69
18d	Thr	Aloc	OH	H	OH	CH_2OH	56	
18e	Ser	Aloc	OH	OH	H	H	51	
18f	Thr	Aloc	OH	OH	H	H	61	67

being shaken in phosphate buffer for 2 d, substrate **7b** carrying N-terminal Fmoc protection gave product **17b** in a yield of 86%. The analogous *N*-Aloc-protected substrate **7a** which is more soluble than **7b** was hydrolyzed with lipase WG within 2 d to give the deacetylated compound **17a** in a yield of 73%. The *O*-acetyl bonds of glycopeptides **11a**−**f** were hydrolyzed within 2 d to give the deacetylated compounds **18a**−**f** in yields of 41−76%, together with a more hydrophilic byproduct, detected by TLC. Therefore, the reaction time was shortened to 18 h (Table 7) and the yields increased to 67−77%. In order to identify the hydrophilic byproduct, the reaction time for compound **11e** was extended from 2 to 5 d (Scheme 5). Under these conditions, the deacetylated and carboxy-deblocked compound **19** was obtained in high yield. The amino protecting group remained intact. No elemental analysis could be obtained for hygroscopic compounds **18a**−**f** and **19**. They were characterized by NMR and MS. The mild reaction conditions (pH = 7.0; 37 °C) did not affect the glycosidic linkages of the *O*-glycosyl amino acid derivatives. As was found for the papain-catalyzed cleavage of the MEE esters (*vide supra*), the selective enzymatic hydrolysis of the carbohydrate acetates using lipase WG can be applied to carbohydrate amino acid conjugates of quite different structure.

Scheme 5



In conclusion, the presented results demonstrate that the application of papain- and lipase-catalyzed removal of MEE esters and *O*-acetyl protecting groups provides preparative access to glycosyl amino acid and glycopeptide building blocks selectively deprotected either in the peptide or in the carbohydrate portion. We have extended the selective cleavage of glycopeptide MEE esters using lipases and papain, previously reported only for compound **15**,[8] to a wide variety of structurally different and biologically relevant substrates. This methodology is ideally supplemented by the efficient use of lipase WG for the removal of *O*-acetyl groups from the carbohydrate portions. The resulting products can be applied advantageously for the synthesis of *O*-glycopeptides. The biocatalysts tolerate a wide variety of structures, N-terminal protecting groups (Z, Fmoc, Aloc), saccharide portions (glucose, galactose, galactosamine, and xylose), and anomeric configurations. In all cases, the enzyme-mediated transformation is completely selective, and the reaction conditions are so mild that none of the various other functionalities present are affected.

Often, the substrate specificity of enzymes limits their application in synthetic organic chemistry. The presented examples prove that this is not the case for the enzymatic deprotection procedures providing glycosyl amino acid and glycopeptide synthons selectively deblocked either at the carboxylic function or in the carbohydrate part.

Experimental Section

Materials and Methods. Optical rotations were measured with a Perkin-Elmer-241 polarimeter. ^{1}H-NMR (90 MHz) were recorded on a Bruker-WH 90 spectrometer. ^{1}H-NMR (200 MHz) spectra and ^{13}C-NMR (50.3 MHz) spectra were recorded on a Bruker-AC-200 and ^{1}H-NMR (400 MHz) and ^{13}C-NMR (100.6 MHz) spectra on a Bruker-AM 400. Melting points are uncorrected. Analytical TLC plates (silica gel 60-F_{254}) were purchased from E. Merck, Darmstadt, Germany. Flash chromatography was carried out on silica gel 30−60 μm purchased from J. T. Baker, Gross Gerau, Germany. Visualization by spraying with a 0.3% solution of ninhydrine in methanol/acetic acid, 93:3 (v/v), or a solution of 0.2% *p*-methoxyphenol in ethanol/2 N H_2SO_4 (1/1, v/v) and heating. Exclusively L-amino acids were used. Dichloromethane was distilled from Na/Pb alloy. The phosphate buffer was prepared from a solution of disodium hydrogen phosphate (36.5 g, 0.2 mol) in 1 L water which was adjusted to pH 7.0 with H_3PO_4. The cysteine buffer is instable and was freshly prepared from cystein hydrochloride (787 mg, 5 mmol) in 100 mL of water and adjusted to pH

6.6 by NaOH. The NMR signals of the Fmoc-, Z-, Aloc-, Ac-, and MEE-protecting groups were described for only one example.

General Procedure for the Preparation of N-Protected Serine/Threonine (Methoxyethoxy)ethyl Esters 3. The amino acid **1a/b** (0.3 mol) and *p*-toluenesulfonic acid (77.6 g, 0.4 mol) were dissolved in diethylene glycol monomethyl ether (188 mL, 1.6 mol) and benzene (300 mL, 3.4 mol). The solution was refluxed for 10 h. The diethylene glycol monomethyl ether and benzene were evaporated in vacuo. The crude products **2a/b** were identified by 90-MHz ^{1}H-NMR spectra and converted directly to the N-terminal-protected derivatives without further purification.

To a solution of amino acid (methoxyethoxy)ethyl ester hydro-*p*-toluenesulfonate **2a/b** (0.05 mol) and *N*-ethyldiisopropylamine (20.4 mL, 0.12 mol) in 500 mL of dry dichloromethane was added the corresponding chloroformate in portions at 0 °C within 30 min. After being stirred at 0 °C for 1 h and a further 14 h at rt, the mixture was washed three times with 125 mL of 0.2 mol HCl, 125 mL of saturated $NaHCO_3$, and 100 mL of water. The organic layer was dried with $MgSO_4$ and concentrated in vacuo. The product was purified by flash chromatography.

***N*-(9-Fluorenylmethyloxycarbonyl)-L-serine (methoxyethoxy)ethyl ester (3a):** eluent of flash chromatography, petroleum ether/ethyl acetate (3:1, v/v); yield 13.3 g (61%); colorless crystals; mp 65−67 °C; $[\alpha]^{22}_D = -4.6$ ($c = 1.1$, $CHCl_3$); $R_f = 0.28$ (petroleum ether/ethyl acetate = 1:2); 200-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 7.90-7.30$ (m, 9H, arom H, NH), 5.0 (s, 1H, OH), 4.31−4.12 (m, 6H, α-CH Ser, $COOCH_2$ MEE, OCH_2 Fmoc, H-9 Fmoc), 3.68−3.38 (m, 8H, 3 OCH_2 MEE, β-CH_2 Ser), 3.20 (s, 3H, OCH_3 MEE). Anal. Calcd for $C_{23}H_{27}NO_7$: C, 64.32; H, 6.34; N, 3.26. Found: C, 64.41; H, 6.49; N, 3.27.

***N*-(9-Fluorenylmethyloxycarbonyl)-L-threonine (methoxyethoxy)ethyl ester (3b):** eluent of flash chromatography, petroleum ether/ethyl acetate (3:1, v/v); yield 14.2 g (64%); colorless crystals; mp 60−62 °C; $[\alpha]^{22}_D = -22.6$ ($c = 1.1$, $CHCl_3$); $R_f = 0.33$ (petroleum ether/ethyl acetate = 1:2); 200-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 7.93-7.34$ (m, 9H, arom H, NH), 4.33−4.06 (m, 7H, α-CH Thr, β-CH Thr, $COOCH_2$ MEE, OCH_2 Fmoc, 9-H Fmoc), 3.68−3.38 (m, 3 OCH_2 MEE, H_2O), 3.38 (s, 3H, OCH_3 MEE), 1.12 (d, $J = 5.9$ Hz, 3H, CH_3 Thr). Anal. Calcd for $C_{24}H_{29}NO_7$: C, 65.00; H, 6.59; N, 3.16. Found: C, 64.82; H, 6.62; N, 3.02.

***N*-(Benzyloxycarbonyl)-L-serine (methoxyethoxy)ethyl ester (3c):** eluent of flash chromatography, petroleum ether/ethyl acetate (3:1, v/v); yield 10.2 g (60%); colorless oil; $[\alpha]^{22}_D = -6.9$ ($c = 1.1$, $CHCl_3$); $R_f = 0.28$ (petroleum ether/ethyl acetate = 1:2); 200-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 7.33$ (m, 5H, arom H), 5.92 (d, 1H, NH), 5.10 (s, 2H, $OCH_2C_6H_5$), 4.54−4.41 (m, 2H, α-CH Ser, $COOCH_{2a}$ MEE), 4.22−4.12 (m, 1H, $COOCH_{2b}$ MEE), 4.04 (dd, $J_1 = 2.6$ Hz, $J_2 = 11.4$ Hz, 1H, β-CH_{2a} Ser), 3.75 (dd, $J_1 = 3.0$ Hz, $J_2 = 11.4$ Hz, 1H, β-CH_{2b} Ser), 3.68−3.46 (m, 6H, 3 OCH_2 MEE), 3.33 (s, 4H, OCH_3 MEE, OH). Anal. Calcd for $C_{16}H_{23}NO_7$: C, 56.30; H, 6.79; N, 4.10. Found: C, 56.05; H, 7.00; N, 4.24.

***N*-(Benzyloxycarbonyl)-L-threonine (methoxyethoxy)ethyl ester (3d):** eluent of flash chromatography, petroleum ether/ethyl acetate (3:1, v/v); yield 10.7 g (60%); colorless oil; $[\alpha]^{22}_D = -23.0$ ($c = 1.1$, $CHCl_3$); $R_f = 0.37$ (petroleum ether/ethyl acetate = 1:5); 200-MHz ^{1}H-NMR (DMSO-d_6) $\delta = 7.26$ (s, 5H, arom H), 7.05 (d, 1H, NH), 5.07 (s, 2H, $OCH_2C_6H_5$), 4.62 (d, 1H, OH), 4.10−3.70 (m, 4H, α-CH Thr, β-CH Thr, $COOCH_2$), 3.53−3.26 (m, 6H, 3 OCH_2 MEE), 3.04 (s, 3H, OCH_3 MEE), 0.93 (d, 3H, CH_3 Thr). Anal. Calcd for $C_{17}H_{25}NO_7$: C, 57.45; H, 7.09; N, 3.94. Found: C, 56.99; H, 7.29; N, 3.76.

***N*-(Allyloxycarbonyl)-L-serine (methoxyethoxy)ethyl ester (3e):** eluent of flash chromatography, petroleum ether/ethyl acetate (1:2, v/v); yield 11.5 g (79%); colorless oil; $[\alpha]^{22}_D = -72.1$ ($c = 1.0$, $CHCl_3$); $R_f = 0.31$ (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.86$ (m, 1H, =CH−), 5.80 (d, $J = 8.1$ Hz, 1 H, NH), 5.28 (m, $J_1 = 16.1$ Hz, $J_2 = 1.0$ Hz, 1H, CH_2=CH− trans), 5.17 (dd, $J_1 = 10.5$ Hz, $J_2 = 1.2$ Hz, 1H, CH_2=CH− cis), 4.55−4.51 (m, 3H, OCH_2 Aloc, $COOCH_{2a}$ MEE), 4.40 (m, 1H, α-CH Ser), 4.16 (m, 1H, $COOCH_{2b}$ MEE), 4.04 (d, $J = 11.0$ Hz, 1H, β-CH_{2a} Ser), 3.79 (d, $J = 10.9$ Hz, 1H, β-CH_{2b} Ser), 3.67, 3.57, 3.50 (3m, 6H, 3 OCH_2 MEE), 3.33 (s, 3H, OCH_3 MEE); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 170.56$ (C=O), 156.03 (C=O, urethane), 132.59 (=CH−), 117.70 (CH_2=CH−), 71.77, 70.08, 68.71, 65.84 (4 OCH_2 MEE), 63.79, 63.17 (OCH_2 Aloc, β-CH_2 Ser), 58.87 (OCH_3 MEE), 56.26 (α-CH Ser). Anal. Calcd for $C_{12}H_{21}NO_7$: C, 49.48; H, 7.27; N, 4.81. Found: C, 49.71; H, 7.34; N, 4.60.

***N*-(Allyloxycarbonyl)-L-threonine (methoxyethoxy)ethyl ester (3f):** eluent of flash chromatography, petroleum ether/ethyl acetate (1:2, v/v); yield 11.8 g (77%); colorless oil; $[\alpha]^{22}_D = -266.4$ ($c = 1.0$, $CHCl_3$); $R_f = 0.37$ (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 4.35$ (d, 1H, β-CH Thr), 4.31 (m, 1H, α-CH Thr), 1.20 (d, 3H, CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 67.64$ (β-CH Thr), 59.43 (α-CH Thr), 19.70 (γ-CH_3 Thr). Anal. Calcd for $C_{13}H_{23}NO_7$: C, 51.14; H, 7.59; N, 4.59. Found: C, 50.80; H, 7.43; N, 4.60.

***N*-(Allyloxycarbonyl)-*O*-(3,4,6-tri-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranosyl)-L-serine (Methoxyethoxy)ethyl Ester (6a).** To a solution of **3e** (3.3 g, 11 mmol) in dry CH_2Cl_2/toluene (150 mL, v/v = 1/1) was added 5 g of molecular sieves (4 Å, powder), and the mixture was stirred for 1 h. The solution was cooled to 0 °C, and silver carbonate (4.5 g, 16 mmol) and silver perchlorate[12c] (300 mg, 1.4 mmol) were added. The mixture was stirred for 1 h in the dark. A solution of the glycosyl bromide **4** in dry CH_2Cl_2/toluene (150 mL, v/v) was added dropwise at 0 °C within 30 min. The mixture was stirred for a further 15 h at rt, filtered off, and concentrated in vacuo and the residue purified by flash chromatography (petroleum ether/ethyl acetate 4:1, v/v); yield 3.33 g (49%); colorless oil; $[\alpha]^{22}_D = +85.4$ ($c = 1.0$, $CHCl_3$); $R_f = 0.53$ (petroleum ether/ethyl acetate = 1:3); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.36$ (d, 1H, 4-H), 5.20 (dd, $J_{2,3} = 11.2$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 4.95 (d, $J_{1,2} = 3.4$ Hz, 1H, 1-H), 4.52 (m, 3H, α-CH Ser, OCH_2 Aloc), 4.30−3.91 (m, 7H, $COOCH_2$ MEE, β-CH_2 Ser, 6-Ha/b, 5-H), 3.57−3.52 (m, 3H, OCH_2 MEE, 2-H); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 155.62$ (C=O, urethane), 98.97 (C-1), 71.72, 70.29, 69.34, 68.62, 65.79, 64.80 (OCH_2 Aloc, 4 OCH_2 MEE, β-CH_2 Ser), 67.76, 67.36, 67.01 (C-3, C-4, C-5); 61.50 (C-6); 58.77, 57.26 (OCH_3 MEE, C-2), 54.25 (α-CH Ser). Anal. Calcd for $C_{24}H_{36}N_4O_{14}$: C, 47.68; H, 6.00; N, 9.27. Found: C, 47.68; H, 5.91; N, 9.26.

General Procedure for the Glycosylation of Fmoc- and Z-Protected Amino Acid Esters 6b−d. A mixture of thioglycoside **5** (2.25 g, 6.0 mmol), protected amino acid **3a,b,d** (6.0 mmol), and 4.0 g of molecular sieves (4 Å, powder) in 20 mL of dry CH_2Cl_2/toluene (1:1) was stirred for 1 h at rt. After addition of dimethyl(methylthio)sulfonium trifluoromethanesulfonate[17] (DMTST, 3.87 g, 9 mmol) the suspension was stirred for another 16 h at rt. The mixture was neutralized by addition of *N*-ethyldiisopropylamine (1.94 g, 9 mmol), filtered, and dried in vacuo. The two anomers were separated by flash chromatography (petroleum ether/ethyl acetate).

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(3,4,6-tri-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranosyl)-L-serine (methoxyethoxy)ethyl ester (6b):** yield 3.76 g (84.4%), α:β = 4:1, 3.08 g (69%, pure α); colorless oil; $[\alpha]^{22}_D = +88.4$ ($c = 1.1$, $CHCl_3$); $R_f = 0.5$ (petroleum ether/ethyl acetate = 1:2); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 6.02$ (d, $J = 8.0$ Hz, 1H, NH), 5.42 (d, 1H, 4-H), 5.29 (dd, $J_{2,3} = 11.2$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 4.97 (d, $J_{1,2} = 3.5$ Hz, 1H, 1-H), 4.58 (m, 1H, α-CH Ser), 4.39−4.34 (m, 3H), 4.23 (t, $J = 7.2$ Hz, 1H), 4.17 (t, $J = 6.5$ Hz, 1H), 4.10 (dd, $J_1 = 3.1$ Hz, $J_2 = 10.7$ Hz, 1H, β-CH_{2a} Ser), 4.01−3.98 (m, 3H, 6-Ha/b, β-CH_{2b}-Ser), 3.61−3.58 (m, 3H, OCH_2 MEE, 2-H); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 155.82$ (C=O, urethane), 99.15 (C-1), 71.82, 70.41, 69.42, 68.73, 67.28, 64.95 (OCH_2 Fmoc, 4 OCH_2 MEE, β-CH_2 Ser), 67.87, 67.47, 67.60 (C-3, C-4, C-5), 61.60 (C-6), 58.89, 57.40 (OCH_3 MEE, C-2), 54.43 (α-CH Ser), 47.05 (C-9 Fmoc). Anal. Calcd for $C_{35}H_{42}N_4O_{14}\cdot H_2O$: C, 55.26; H, 5.83; N, 7.36. Found: C, 55.75; H, 5.75; N, 7.07.

***N*-(Benzyloxycarbonyl)-*O*-(3,4,6-tri-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (6c):** yield 2.97 g (74%), α:β = 3:2, 1.81 g

(17) Fügedi, P.; Garegg, P. J. *Carbohydr. Res.* **1986**, *149*, C9.

(45%, pure α); colorless wax; $[\alpha]^{22}_D = +69.5$ ($c = 1.0$, $CHCl_3$); $R_f = 0.8$ (ethyl acetate/ethanol = 10:1); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.54$ (d, $J = 9.4$ Hz, 1H, NH), 5.40 (d, 1H, 4-H), 5.18 (dd, $J_{2,3} = 11.2$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 5.08 (d, $J_{1,2} = 3.7$ Hz, 1H, 1-H), 4.45−4.36 (m, 3H, α-CH Thr, β-CH Thr, $COOCH_{2a}$ MEE), 4.20 (m, 1H, 5-H), 4.03 (d, $J = 6.5$ Hz, 2H, 6-Ha/b), 3.62 (dd, $J_{1,2} = 3.7$ Hz, $J_{2,3} = 11.2$ Hz, 1H, 2-H), 1.30 (d, $J = 6.3$ Hz, 3H, CH_3 Thr); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 156.75$ (C=O, urethane), 99.50 (C-1), 76.8 (β-CH Thr), 68.24, 67.50, 66.87, 61.75 (C-3, C-4, C-5), 61.75 (C-6), 58.88, 58.69, 57.71 (OCH_3 MEE, α-CH Thr, C-2), 18.50 (CH_3 Thr). Anal. Calcd for $C_{29}H_{40}N_4O_{14}\cdot 1.5H_2O$: C, 50.07; H, 6.23; N, 8.05. Found: C, 50.14; H, 5.89; N, 7.66.

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(3,4,6-tri-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranosyl)-L-threonine (Methoxyethoxy)ethyl Ester (6d).** Complete characterization as compound **7d**: yield 2.76 g (62%), α:β = 3:2, 1.78 g (40%, pure α); colorless oil; $R_f = 0.65$ (petroleum ether/ethyl acetate = 1:3); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.68$ (d, $J = 9.5$ Hz, 1H, NH), 5.44 (d, 1H, 4-H), 5.27 (dd, $J_{2,3} = 11.2$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 5.13 (d, $J_{1,2} = 3.7$ Hz, 1H, 1-H), 4.49−4.23 (m, 8H, α-CH Thr, β-CH Thr, OCH_2 Fmoc, 9-H Fmoc, $COOCH_2$ MEE, 5-H), 4.06 (d, $J = 6.5$ Hz, 2H, 6-Ha/b), 3.65 (dd, $J_{1,2} = 3.7$ Hz, $J_{2,3} = 11.2$ Hz, 1H, 2-H), 1.33 (d, $J = 6.4$ Hz, CH_3 Thr); 50.3-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 156.66$ (C=O, urethane), 99.32 (C-1), 76.69 (β-CH Thr), 68.08, 67.44, 66.82 (C-3, C-4, C-5), 61.65 (C-6), 58.69, 57.67 (OCH_3 MEE, α-CH Thr), 18.44 (CH_3 Thr), $C_{36}H_{44}N_4O_{14}$.

General Procedure for the Transformation of Azidogalactosyl to Acetamidogalactosyl Amino Acid Esters 7. A solution of 2-azidogalactosyl amino acid ester **6a**−**d** (1 mmol) in thioacetic acid (10 mL) was stirred for 16 h at rt under nitrogen atmosphere. The solvent was evaporated in vacuo and the crude product purified by flash chromatography (petroleum ether/ethyl acetate 4:1).

***N*-(Allyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serine (methoxyethoxyethyl ester (7a):** yield 366 mg (59%); colorless amorphous solid; $[\alpha]^{22}_D = +68.1$ ($c = 1.1$, $CHCl_3$); $R_f = 0.1$ (petroleum ether/ethyl acetate = 1:3); 400-MHz ^{1}H-NMR ($CDCl_3$) δ = 5.97−5.84 (m, 3H, 2 NH, =CH−), 5.33 (d, 1H, 4-H), 5.03 (dd, $J_{2,3} = 11.4$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 4.83 (d, $J_{1,2} = 3.7$ Hz, 1H, 1-H), 4.56−4.49 (m, 3H, α-CH Ser, OCH_2 Aloc), 4.51 (m, 1H, 2-H), 4.14−4.05 (m, 2H, 5-H, 6-H_a), 4.01 (dd, $J_{5,6b} = 6.6$ Hz, $J_{6a,6b} = 10.6$ Hz, 1H, 6-H_b), 3.92 (m, β-CH_2 Ser), 2.11, 2.01, 1.94, 1.92 (4s, 12H, 4 CH_3CO); 100.6-MHz ^{13}C-NMR ($CDCl_3$) δ = 170.22, 170.34, 170.21, 170.03 (C=O), 155.67 (C=O, urethane), 98.91 (C-1), 71.76, 70.21, 69.31, 68.58, 66.04, 64.59 (OCH_2 Aloc, 4 OCH_2 MEE, β-CH_2 Ser), 68.29, 67.20, 67.15 (C-3, C-4, C-5), 61.85 (C-6), 54.36 (α-CH Ser), 47.43 (C-2), 22.96 (CH_3CON). Anal. Calcd for $C_{26}H_{40}N_2O_{15}$: C, 50.32; H, 6.50; N, 4.51. Found: C, 50.41; H, 6.43; N, 4.41.

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serine (methoxyethoxy)ethyl ester (7b):** yield 683 mg (90%); colorless crystals; mp 45−49 °C; $[\alpha]^{22}_D = +60.6$ (c = 0.5, $CHCl_3$); $R_f = 0.76$ (petroleum ether/ethyl acetate = 2:1); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 6.04$ (d, $J = 8.2$ Hz, 1H, NH), 5.99 (d, $J = 9.6$ Hz, 1H, NH), 5.34 (d, 1H, 4-H), 5.06 (dd, $J_{2,3} = 11.4$ Hz, $J_{3,4} = 2.85$ Hz, 1H, 3-H), 4.83 (d, $J_{1,2} = 3.3$ Hz, 1H, 1-H), 4.57 (m, 2H), 4.45−4.40 (m, 2H), 4.34−4.18 (m, 3H), 4.11 (t, $J = 6.3$ Hz, 1H), 4.05 (dd, $J_1 = 5.8$ Hz, $J_2 = 11.2$ Hz, 1H), 4.00 (dd, $J_1 = 7.3$ Hz, $J_2 = 11.2$ Hz, 1H), 3.93 (s, 2H), 3.72−3.57 (m, 4H), 3.46 (m, 2H); 100.6-MHz ^{13}C-NMR ($CDCl_3$) δ = 155.83 (C=O, urethane), 98.96 (C-1), 71.68, 70.16, 69.33, 68.51, 67.14, 64.52 (OCH_2 Fmoc, 4 OCH_2 MEE, β-CH_2 Ser), 68.27, 67.18, 67.10 (C-3, C-4, C-5), 61.84 (C-6), 54.39 (α-CH Ser), 47.38, 47.05 (C-2, C-9 Fmoc). Anal. Calcd for $C_{37}H_{46}N_2O_{15}$: C, 58.56; H, 6.11; N, 3.70. Found: C, 58.55; H, 6.02; N, 3.71.

***N*-(Benzyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (7c):** yield 609 mg (89%); colorless oil; $[\alpha]^{22}_D = +52.2$ ($c = 1.0$, $CHCl_3$); $R_f = 0.32$ (ethyl acetate/ethanol = 10:1); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.88$ (d, J = 9.7 Hz, 1H, NH), 5.70 (d, $J = 9.6$ Hz, 1H, NH), 5.33 (d, 1H, 4-H), 5.01 (dd, $J_{2,3} = 11.4$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 4.93 (d, $J_{1,2} = 3.6$ Hz, 1H, 1-H), 4.50 (m, 1H, 2-H), 4.42 (dd, $J_1 = 1.7$ Hz, $J_2 = 9.6$ Hz, 1H, α-CH Thr), 4.29−4.20 (m, 3H, β-CH Thr, $COOCH_2$ MME), 4.17 (m, 1H, 5-H), 4.04 (m, 2H, 6-Ha/b), 1.30 (d, $J = 6.2$ Hz, 3H, CH_3 Thr); 100.6-MHz ^{13}C-NMR ($CDCl_3$) δ = 156.40 (C=O, urethane), 99.94 (C-1), 77.12 (β-CH Thr), 68.44, 67.34, 67.08 (C-3, C-4, C-5), 62.04 (C-6), 58.86, 58.47 (α-CH Thr, OCH_3 MEE), 47.36 (C-2), 18.16 (CH_3 Thr). Anal. Calcd for $C_{31}H_{44}N_2O_{15}$: C, 54.38; H, 6.48; N, 4.10. Found: C, 54.97; H, 6.67; N, 3.88.

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (7d):** yield 564 mg (73%); colorless crystals; mp 55−60 °C; $[\alpha]^{22}_D = +38.7$ ($c = 0.84$, $CHCl_3$); $R_f = 0.18$ (ethyl acetate); 400-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 6.18$ (d, $J = 11.4$ Hz, 1H, NH), 6.15 (d, $J = 10.0$ Hz, 1H, NH), 5.33 (d, 1H, 4-H), 5.03 (dd, $J_{2,3} = 11.4$ Hz, $J_{3,4} = 3.1$ Hz, 1H, 3-H), 4.90 (d, $J_{1,2} = 3.6$ Hz, 1H, 1-H), 4.51−4.38 (m, 4H), 4.31−4.29 (m, 1H), 4.22−4.15 (m, 4H), 4.08−3.99 (m, 2H), 3.67−3.52 (m, 4H), 3.45−3.31 (m, 2H), 1.25 (d, $J = 6.3$ Hz, CH_3 Thr); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 156.60$ (C=O, urethane), 99.82 (C-1), 76.85 (β-CH Thr), 68.35, 67.32, 66.97 (C-3, C-4, C-5), 62.02 (C-6), 58.75, 58.50 (OCH_3 MEE, α-CH Thr), 47.30, 47.12 (C-9 Fmoc, C-2), 18.15 (CH_3 Thr). Anal. Calcd for $C_{38}H_{48}N_2O_{15}\cdot 0.5H_2O$: C, 58.38; H, 6.32; N, 3.58. Found: C, 58.28; H, 6.30; N, 3.70.

General Procedure for the Synthesis of β-Glycosyl Amino Acid Esters 11. A solution of per-*O*-acetylated monosaccharide **8**−**10** (1mmol) and protected amino acid ester **3e**−**f** (1mmol) in dry CH_2Cl_2 (10 mL) was stirred at rt for 1 h with molecular sieves (4 Å, spherical). In a second flask a solution of trimethylsilyl trifluoromethanesulfonate (TMS-triflate, 0.64 mL) and molecular sieves (4 Å, spherical) in CH_2Cl_2 was stirred under argon for 15 min. TMS-triflate was then added dropwise under argon atmosphere to the solution of saccharide and amino acid ester. After 2−4 h, triethylamine (5 mL) was added and the reaction mixture was diluted with CH_2Cl_2 (100 mL), washed with H_2O, dried with $MgSO_4$, and concentrated in vacuo. The crude product was purified by flash chromatography (petroleum ether/ethyl acetate 1:1).

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-L-serine (methoxyethoxy)ethyl ester (11a):** yield 236 mg (38%); colorless oil; $[\alpha]^{22}_D = +57.2$ (c = 1.0, $CHCl_3$); $R_f = 0.42$ (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.55$ (d, $J = 7.7$ Hz, 1H, NH), 5.14, 5.01, (2t, 2H, 3-H, 4-H), 4.90 (dd, $J_{2,3} = 9.5$ Hz, $J_{2,1} = 8.0$ Hz, 1H, 2-H), 4.49 (d, $J_{1,2} = 8.0$ Hz, 1H, 1-H), 4.34−4.20 (m, 2H, α-CH Ser, $COOCH_{2b}$ MEE), 4.26 (dd, $J_{6a,6b} = 14.6$ Hz, $J_{6a,5}$ = 2.5 Hz, 1H, 6-Ha), 4.28 (dd, $J_{6b,6a} = 11.9$ Hz, $J_{6b,5} = 2.5$ Hz, 1H, 6-Hb), 4.07 (dd, $J_1 = 2.9$ Hz, $J_2 = 11.0$ Hz, 1H, β-CH_{2a} Ser), 3.85 (dd, $J_1 = 3.2$ Hz, $J_2 = 10.7$ Hz, 1H, β-CH_{2b} Ser), 3.63 (m, 1H, 5-H); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 101.09$ (C-1), 72.59, 71.90, 71.07, 68.25 (C-2, C-3, C-4, C-5), 69.49 (β-CH_2 Ser), 61.78 (C-6), 54.33 (α-CH Ser). Anal. Calcd for $C_{26}H_{39}NO_{16}$: C, 50.24; H, 6.32; N, 2.25. Found: C, 50.21; H, 6.10; N, 2.43.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (11b):** yield 280 mg (44%); colorless oil; $[\alpha]^{22}_D = -83.7$ (c = 1.0, $CHCl_3$); $R_f = 0.48$ (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.55$ (d, $J = 8.9$ Hz, 1H, NH), 5.14, 5.05 (2t, 2H, 3-H, 4-H), 4.86 (dd, $J_{2,3} = 9.6$ Hz, $J_{2,1} = 8.0$ Hz, 1H, 2-H), 4.48 (d, $J_{1,2} = 7.9$ Hz, 1H, 1-H), 4.37−4.20 (m, 4H, β-CH Thr, $COOCH_{2b}$ MEE, 6-Ha, 6-Hb), 4.04 (dd, J_1 = 2.5 Hz, $J_2 = 8.0$ Hz, 1H, α-CH Thr), 3.64 (m, 1H, 5-H), 1.18 (d, $J = 6.6$ Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 99.04$ (C-1), 75.39 (β-CH Thr), 72.47, 71.56, 71.14, 68.04 (C-2, C-3, C-4, C-5), 61.50 (C-6), 58.33 (α-CH Thr), 17.16 (γ-CH_3 Thr). Anal. Calcd for $C_{27}H_{41}NO_{16}$: C, 51.02; H, 6.50; N, 2.21. Found: C, 51.12; H, 6.78; N, 2.08.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-L-serine (methoxyethoxy)ethyl ester (11c):** yield 323 mg (52%); colorless oil; $[\alpha]^{22}_D = +152.8$ (c = 1.0, $CHCl_3$); $R_f = 0.47$ (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.54$ (d, $J = 7.8$ Hz, 1H, NH), 5.34 (d, $J_{4,5} = 2.7$ Hz, 1H, 4-H), 5.12 (dd, $J_{2,3} = 10.4$ Hz, $J_{2,1}$ =

7.9 Hz, 1H, 2-H), 5.01, (dd, $J_{3,2}$ = 10.4 Hz, $J_{3,4}$ = 3.4 Hz, 1H, 3-H), 4.46 (d, $J_{1,2}$ = 7.8 Hz, 1H, 1-H), 4.36−4.23 (m, 3H, α-CH Ser, $COOCH_2$ MEE), 4.18−4.05 (m, 3H, 6-Ha, 6-Hb, β-CH_{2a} Ser), 4.00−3.81 (m, 2H, β-CH_{2b} Ser, 5-H); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 101.66 (C-1), 70.82, 70.71, 68.62, 66.93 (C-2, C-3, C-4, C-5), 69.45 (β-CH_2 Ser), 61.10 (C-6), 54.35 (α-CH Ser). Anal. Calcd for $C_{26}H_{39}NO_{16}$: C, 50.24; H, 6.32; N, 2.25. Found: C 49.72, H 5.94, N 1.89.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (11d):** yield 439 mg (69%); colorless oil; $[\alpha]^{22}_D$ = −66.4 (*c* = 1.0, $CHCl_3$); R_f = 0.42 (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.55 (d, *J* = 9.1 Hz, 1H, NH), 5.32 (d, $J_{4,5}$ = 3.3 Hz, 1H, 4-H), 5.05 (dd, $J_{2,3}$ = 10.4 Hz, $J_{2,1}$ = 7.8 Hz, 1H, 2-H), 4.95 (dd, $J_{3,4}$ = 3.3 Hz, $J_{3,2}$ = 10.5 Hz, 1H, 3-H), 4.44 (d, $J_{1,2}$ = 7.8 Hz, 1H, 1-H), 4.39−4.15 (m, 4H, β-CH Thr, α-CH Thr, H-6a, H-6b), 3.85 (m, 1H, 5-H), 1.21 (d, *J* = 8.8 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 100.05 (C-1), 75.85 (β-CH Thr), 70.71, 70.54, 68.91, 66.84 (C-2, C-3, C-4, C-5), 60.85 (C-6), 58.94 (α-CH Thr), 17.63 (γ-CH_3 Thr). Anal. Calcd for $C_{27}H_{41}NO_{16}$: C, 51.02; H, 6.50; N, 2.21. Found: C, 50.84; H, 6.50; N, 2.22.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4-tri-*O*-acetyl-β-D-xylopyranosyl)-L-serine (methoxyethoxy)ethyl ester (11e):** yield 423 mg (77%); colorless oil; $[\alpha]^{22}_D$ = −45.1 (*c* = 1.0, $CHCl_3$); R_f = 0.46 (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.53 (d, *J* = 8.3 Hz, 1H, NH), 5.08, (t, $J_{3,4}$ = 7.96 Hz, $J_{3,2}$ = 7.9 Hz, 1H, 3-H), 4.85 (dd, $J_{2,3}$ = 7.9 Hz, $J_{2,1}$ = 4.8 Hz, 1H, 4-H), 4.81 (dd, $J_{2,3}$ = 8.0 Hz, $J_{2,1}$ = 6.1 Hz, 1H, 2-H), 4.47 (d, $J_{1,2}$ = 6.1 Hz, 1H, 1-H), 4.34−4.25 (m, 3H, α-CH Ser, $COOCH_2$ MEE), 4.21 (dd, J_1 = 10.8 Hz, J_2 = 3.2 Hz, 1H, β-CH_{2a} Ser), 4.02 (dd, $J_{5a,5b}$ = 12.9 Hz, $J_{5a,4}$ = 4.7 Hz, 1H, 5-Ha), 3.77 (m, J_1 = 2.9 Hz, J_2 = 10.3 Hz, 1H, β-CH_{2b} Ser), 3.33 (m, $J_{5b,5a}$ = 12.0 Hz, $J_{5b,4}$ = 8.0 Hz, 1H, 5-Hb); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 100.59 (C-1), 70.67, 70.20, 68.48 (C-2, C-3, C-4), 69.10 (β-CH_2 Ser), 61.57 (C-5), 54.19 (α-CH Ser). Anal. Calcd for $C_{23}H_{35}NO_{14}$: C, 50.27; H, 6.24; N, 2.55. Found: C, 49.68; H, 6.00; N, 2.23.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4-tri-*O*-acetyl-β-D-xylopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (11f):** yield 411 mg (73%); colorless oil; $[\alpha]^{22}_D$ = −361.2 (*c* = 1.0, $CHCl_3$); R_f = 0.41 (petroleum ether/ethyl acetate = 1:4); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.40 (d, *J* = 7.7 Hz, 1H, NH), 5.08, 4.84, (t, m, 2H, 3-H, 4-H), 4.78 (dd, $J_{2,3}$ = 8.3 Hz, $J_{2,1}$ = 6.6 Hz, 1H, 2-H), 4.47 (d, $J_{1,2}$ = 8.0 Hz, 1H, 1-H), 4.39 (m, 1H, β-CH Thr), 4.33 (m, 1H, α-CH Thr), 3.97 (dd, $J_{5a,5b}$ = 11.8 Hz, $J_{5a,4}$ = 4.9 Hz, 1H, 5-Ha), 3.28 (dd, $J_{5b,5a}$ = 11.8 Hz, $J_{5b,4}$ = 8.4 Hz, 1H, 5-Hb), 1.18 (d, *J* = 6.3 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 98.79 (C-1), 74.69 (β-CH Thr), 70.97, 70.61, 68.61 (C-2, C-3, C-4), 61.67 (C-5), 58.41 (α-CH Thr), 17.09 (γ-CH_3 Thr). Anal. Calcd for $C_{24}H_{37}NO_{14}$: C, 51.15; H, 6.62; N, 2.49. Found: C, 50.76; H, 6.20; N, 2.61.

General Procedure for the Hydrolysis of *O*-Glycosyl Amino Acid (Methoxyethoxy)ethyl Esters with Papain 12/13. The glycosylated amino acid MEE ester **7a−c/11a−f** (2mmol) was dissolved in 1.5 mL of acetone and added to a solution of 300 mg of papain in cystein buffer (36 mL). The solution was shaken for 3 d at 37 °C, saturated with NaCl, and extracted five times with ethyl acetate (25 mL). The organic layer was dried with $MgSO_4$ and evaporated and the remaining product purified by flash chromatography (ethyl acetate/petroleum ether = 4:1 → ethyl acetate → ethyl acetate/ethanol = 1:4).

***N*-(Allyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serine (12a):** yield 778 mg (75%); colorless amorphous solid; $[\alpha]^{22}_D$ = +113.3 (*c* = 1.0, $CHCl_3$) (lit.[18] $[\alpha]^{22}_D$ = +117.5 (*c* = 1.0, $CHCl_3$)); R_f = 0.13 (ethyl acetate/methanol = 2:1); 400-MHz ^{1}H-NMR (DMSO-d_6) δ = 7.87 (d, *J* = 8.4 Hz, 1H, NH), 7.14 (d, *J* = 7.8 Hz, 1H, NH Ser), 5.27 (d, 1H, 4-H), 5.00 (dd, $J_{2,3}$ = 11.7 Hz, $J_{3,4}$ = 3.1 Hz, 1H, 3-H), 4.82 (d, $J_{1,2}$ = 3.4 Hz, 1H, 1-H), 4.24 (t, *J* = 6.3 Hz, 1H, α-CH Ser), 4.17 (ddd, $J_{1,2}$ = 3.4 Hz, $J_{2,3}$ = 11.6 Hz, $J_{2\text{-H,NH}}$ = 8.4 Hz, 1H, 2-H), 4.03 (m, 2H, 6-Ha, 5-H), 4.01 (dd, $J_{5,6b}$ = 6.6 Hz, $J_{6a,6b}$ = 10.6 Hz, 1H, 6-Hb), 3.78 (dd, J_1 = 3.9 Hz, J_2 = 10.6 Hz, 1H, β-CH_{2a} Ser), 3.78 (dd, J_1 = 5.1, J_2 = 10.6 Hz, 1H, β-CH_{2b} Ser); 100.6-MHz ^{13}C-NMR ($CDCl_3$) δ = 97.97 (C-1), 68.83, 64.42 (OCH_2 Aloc, β-CH_2 Ser), 67.75, 67.14, 66.22 (C-3, C-4, C-5) 61.54 (C-6), 55.07 (α-CH Ser), 47.04 (C-2); $C_{21}H_{30}N_2O_{13}$.

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serine (12b):** yield 1.18 g (90%); colorless amorphous solid; $[\alpha]^{22}_D$ = +90.7 (*c* = 1.0, $CHCl_3$) (lit.[19] $[\alpha]^{22}_D$ = +89.9 (*c* = 1.0, $CHCl_3$)); R_f = 0.38 (ethyl acetate/methanol = 2:1); $C_{32}H_{36}N_2O_{13}$.

***N*-(Benzyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-threonine (12c):** yield 839 mg (72%); colorless crystals; mp 94−98 °C; $[\alpha]^{22}_D$ = +94.2 (*c* = 0.55, CH_3OH); R_f = 0.33 (ethyl acetate/methanol = 2:1); 400-MHz ^{1}H-NMR (CD_3OD) δ = 5.42 (d, 1H, 4-H), 5.13 (dd, $J_{2,3}$ = 11.6 Hz, $J_{3,4}$ = 3.1 Hz, 1H, 3-H), 5.00 (d, $J_{1,2}$ = 3.8 Hz, 1H, 1-H), 4.47 (dd, J_1 = 1.7 Hz, J_2 = 6.5 Hz, 1H, β-CH Thr), 4.42 (dd, $J_{1,2}$ = 3.8 Hz, $J_{2,3}$ = 11.5 Hz, 1H, 2-H), 4.36 (t, $J_{5,6}$ = 6.5 Hz, 1H, 5-H), 4.32 (d, *J* = 1.2 Hz, 1H, α-CH Thr), 4.14−4.13 (m, 2H, 6-Ha/b), 1.35 (d, *J* = 6.5 Hz, 3H, CH_3 Thr); 100.6-MHz ^{13}C-NMR (CD_3OD) δ = 100.59 (C-1), 77.79 (β-CH Thr), 69.82, 68.86, 68.17 (C-3, C-4, C-5), 63.32 (C-6), 60.36 (α-CH Thr), 48.79 (C-2), 19.14 (γ-CH_3 Thr). Anal. Calcd for $C_{26}H_{34}N_2O_{13}\cdot H_2O$: C, 52.00; H, 6.04; N, 4.66. Found: C, 52.26; H, 5.78; N, 4.57.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-L-serine (13a):** yield 997 mg (96%); colorless oil; $[\alpha]^{22}_D$ = +57.2 (*c* = 1.0, CH_3OH); R_f = 0.25 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.64 (d, *J* = 8.0 Hz, 1H, NH), 5.15 (t, $J_{3,4}$ = 9.3 Hz, $J_{3,2}$ = 9.4 Hz, 1H, 3-H), 5.04 (t, $J_{4,5}$ = 9.7 Hz, $J_{4,3}$ = 9.5 Hz, 1H, 4-H), 4.93 (dd, $J_{2,3}$ = 9.2 Hz, $J_{2,1}$ = 8.0 Hz, 1H, 2-H), 4.51 (d, $J_{1,2}$ = 7.9 Hz, 1H, 1-H), 4.46 (m, *J* = 4.0 Hz, 1H, α-CH Ser), 4.24 (dd, $J_{6a/6b}$ = 10.7 Hz, $J_{6a/5}$ = 2.8 Hz, 1H, 6-Ha), 4.18 (dd, J_1 = 4.4 Hz, J_2 = 12.4 Hz, 1H, β-CH_{2a} Ser), 4.12 (dd, $J_{6b,6a}$ = 12.3 Hz, $J_{6b,5}$ = 2.4 Hz, 1H, 6-Hb), 4.07 (dd, J_1 = 3.4 Hz, J_2 = 10.6 Hz, 1H, β-CH_{2b} Ser), 3.67 (m, 1H, 5-H); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 100.97 (C-1), 72.62, 71.86, 71.08, 68.30 (C-2, C-3, C-4, C-5), 69.42 (β-CH_2 Ser), 61.82 (C-6), 53.99 (α-CH Ser). Anal. Calcd for $C_{21}H_{29}NO_{14}$: C, 48.54; H, 5.63; N, 2.70. Found: C, 48.57; H, 5.67; N, 2.73.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)-L-threonine (13b).** Difficult separation of product and byproduct by flash chromatography: yield 320 mg (30%); colorless oil; $[\alpha]^{22}_D$ = −83.7 (*c* = 1.0, CH_3OH); R_f = 0.25 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.51 (d, *J* = 9.1 Hz, 1H, NH), 5.14 (t, $J_{3,4}$ = 9.4 Hz, $J_{3,2}$ = 9.4 Hz, 1H, 3-H), 5.05, (t, $J_{4,3}$ = 9.7 Hz, $J_{4,5}$ = 9.5 Hz, 1H, 4-H), 4.88 (dd, $J_{2,3}$ = 9.2 Hz, $J_{2,1}$ = 7.9 Hz, 1H, 2-H), 4.50 (d, $J_{1,2}$ = 7.9 Hz, 1H, 1-H), 4.37 (dd, $J_{6a,6b}$ = 11.9 Hz, $J_{6a,5}$ = 2.5 Hz, 1H, 6-Ha), 4.34−4.24 (m, 2H, β-CH Thr, α-CH Thr), 4.04 (dd, $J_{6b,6a}$ = 12.3 Hz, $J_{6b,5}$ = 3.8 Hz, 1H, 6-Hb), 3.64 (m, $J_{5,6a}$ = 2.9 Hz, $J_{5,6b}$ = 3.2 Hz, $J_{5,4}$ = 9.8 Hz, 1H, 5-H), 1.19 (d, *J* = 6.3 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 99.79 (C-1), 76.29 (β-CH Thr), 72.56, 71.62, 71.06, 68.26 (C-2, C-3, C-4, C-5), 61.34 (C-6), 58.07 (α-CH Thr), 17.71 (γ-CH_3 Thr). Anal. Calcd for $C_{22}H_{31}NO_{14}$: C, 49.53; H, 5.86; N, 2.63. Found: C, 49.10; H, 6.25; N, 2.94.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-L-serine (13c):** yield 852 mg (82%); colorless oil; $[\alpha]^{22}_D$ = +152.8 (*c* = 1.0, CH_3OH); R_f = 0.33 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) δ = 5.61 (d, *J* = 7.7 Hz, 1H, NH), 5.34 (d, $J_{4,5}$ = 3.2 Hz, 1H, 4-H), 5.12 (dd, $J_{2,3}$ = 10.3 Hz, $J_{2,1}$ = 7.9 Hz, 1H, 2-H), 4.97, (dd, $J_{3,2}$ = 10.4 Hz, $J_{3,4}$ = 3.36 Hz, 1H, 3-H), 4.47 (d, $J_{1,2}$ = 7.8 Hz, 1H, 1-H), 4.56−4.46 (m, 1H, α-CH Ser), 4.36 (m, 1H, β-CH_{2a} Ser), 4.13 (dd, $J_{6a,6b}$ = 11.2 Hz, $J_{6a,5}$ = 6.4 Hz, 1H, 6-Ha), 4.06 (dd, $J_{6b,6a}$ = 11.1 Hz, $J_{6a,5}$ = 6.8 Hz, 1H, 6-Hb), 4.03−3.86 (m, 2H, β-CH_{2b} Ser, 5-H); 100.6 MHz ^{13}C-NMR ($CDCl_3$) δ = 101.58 (C-1), 70.84, 70.69, 68.64, 66.95 (C-2, C-3, C-4, C-5), 69.34 (β-CH_2 Ser), 61.16 (C-6), 54.87 (α-CH Ser). Anal. Calcd for $C_{21}H_{29}NO_{14}$: C, 48.54; H, 5.63; N, 2.70. Found: C, 48.45; H, 5.83; N, 2.70.

(18) (a) Paulsen, H.; Merz, G.; Brockhausen, I. *Liebigs Ann. Chem.* **1990**, 719. (b) Paulsen, H.; Merz, G.; Brockhausen, I. *Liebigs Ann. Chem.* **1990**, 1272.

(19) Paulsen, H.; Adermann, K. *Liebigs Ann. Chem.* **1989**, 751.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4,6-tetra-*O*-acetyl-*β*-D-galactopyranosyl)-L-threonine (13d):** yield 928 mg (87%); colorless oil; $[\alpha]^{22}_D = -66.4$ ($c = 1.0$, CH_3OH); $R_f = 0.26$ (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.55$ (d, $J = 8.9$ Hz, 1H, NH), 5.33 (d, $J_{4,5} = 3.4$ Hz, 1H, 4-H), 5.07 (dd, $J_{2,3} = 10.4$ Hz, $J_{2,1} = 7.9$ Hz, 1H, 2-H), 4.95 (dd, $J_{3,4} = 3.3$ Hz, $J_{3,2} = 10.4$ Hz, 1H, 3-H), 4.47 (d, $J_{1,2} = 7.90$ Hz, 1H, 1-H), 4.40 (d, $J = 5.9$ Hz, 1H, β-CH Thr), 4.33 (d, $J = 8.5$ Hz, 1H, α-CH Thr), 4.18 (dd, $J_{6a,6b} = 11.1$ Hz, $J_{6a,5} = 5.8$ Hz, 1H, 6-Ha), 3.99 (dd, $J_{6b,6a} = 11.0$ Hz, $J_{6b,5} = 7.1$ Hz, 1H, 6-Hb), 3.82 (m, 1H, 5-H), 1.20 (d, $J = 6.2$ Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 100.62$ (C-1), 76.26 (β-CH Thr), 70.63, 70.60, 68.67, 66.92 (C-2, C-3, C-4, C-5), 61.01 (C-6), 58.86 (α-CH Thr), 17.82 (γ-CH_3 Thr). Anal. Calcd for $C_{22}H_{31}NO_{14}$: C, 49.53; H, 5.86; N, 2.63. Found: C, 49.53; H, 5.88; N, 2.56.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4-tri-*O*-acetyl-*β*-D-xylopyranosyl)-L-serine (13e):** yield 805 mg (90%); colorless oil; $[\alpha]^{22}_D = -45.1$ ($c = 1.0$, CH_3OH); $R_f = 0.24$ (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.61$ (d, $J = 8.1$ Hz, 1H, NH), 5.10 (t, $J_{3,4} = 8.0$ Hz, $J_{3,2} = 8.0$ Hz, 1H, 1-H), 4.91−4.84 (m, 2H, 4-H, 2-H), 4.50 (m, $J_{1,2} = 6.1$ Hz, 2H, α-CH Ser, 3-H), 4.25 (dd, $J_1 = 10.2$ Hz, $J_2 = 2.4$ Hz, 1H, β-CH_{2a} Ser), 4.06 (dd, $J_{5a,5b} = 12.0$ Hz, $J_{5a,4} = 4.7$ Hz, 1H, 5-Ha), 3.79 (dd, $J_1 = 10.2$ Hz, $J_2 = 3.2$ Hz, 1H, β-CH_{2b} Ser), 3.35 (m, $J_{5b,5a} = 11.5$ Hz, $J_{5b,4} = 8.3$ Hz, 1H, 5-Hb); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 100.55$ (C-1), 70.67, 70.33, 68.48 (C-2, C-3, C-4), 68.86 (β-CH_2 Ser), 61.60 (C-5), 53.92 (α-CH Ser). Anal. Calcd for $C_{18}H_{25}NO_{12}$: C, 48.32; H, 5.63; N, 3.13. Found: C, 48.20; H, 5.62; N, 3.17.

***N*-(Allyloxycarbonyl)-*O*-(2,3,4-tri-*O*-acetyl-*β*-D-xylopyranosyl)-L-threonine (13f):** yield 831 mg (90%); colorless oil; $[\alpha]^{22}_D = -361.2$ ($c = 1.0$, CH_3OH); $R_f = 0.30$ (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR ($CDCl_3$) $\delta = 5.51$ (d, $J = 9.1$ Hz, 1H, NH), 5.09 (t, $J_{3,4} = 8.2$ Hz, $J_{3,2} = 8.2$ Hz, 1H, 3-H), 4.87 (m, $J_{4,3} = 8.3$ Hz, $J_{4,5a} = 4.8$ Hz, $J_{4,5b} = 8.3$ Hz, 1H, 4-H), 4.80 (dd, $J_{2,3} = 8.2$ Hz, $J_{2,1} = 6.5$ Hz, 1H, 2-H), 4.51 (d, $J_{1,2} = 6.4$ Hz, 1H, 1-H), 4.41 (d, $J = 6.1$ Hz, 1H, β-CH Thr), 4.33 (m, $J = 8.8$ Hz, 1H, α-CH Thr), 4.01 (dd, $J_{5a,5b} = 11.8$ Hz, $J_{5a,4} = 4.7$ Hz, 1H, 5-Ha), 3.28 (dd, $J_{5b,5a} = 11.7$ Hz, $J_{5b,4} = 8.5$ Hz, 1H, 5b), 1.20 (d, $J = 6.3$ Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR ($CDCl_3$) $\delta = 98.66$ (C-1), 74.70 (β-CH Thr), 70.96, 70.67, 68.61 (C-2, C-3, C-4), 61.55 (C-5), 58.94 (α-CH Thr), 16.85 (γ-CH_3 Thr). Anal. Calcd for $C_{19}H_{27}NO_{12}$: C, 49.46; H, 5.90; N, 3.04. Found: C, 49.79; H, 6.11; N, 3.08.

***N*-(*tert*-Butyloxycarbonyl)-L-valyl-L-phenylalanine (Methoxyethoxy)ethyl Ester (14).** A solution of the phenylalanine MEE ester (10 mmol) and *N*-ethyldiisopropylamine (10 mmol) in dry CH_2Cl_2 (20 mL) was added to Boc valine (10 mmol) and 1-(ethoxycarbonyl)-2-ethoxy-1,2-dihydroquinoline[20] (EEDQ, 12 mmol) in dry CH_2Cl_2 (20 mL). The solution was stirred for 12 h at rt and washed three times with 20 mL of 0.5 N HCl, 0.5 N $NaHCO_3$, and water. The organic layer was dried with $MgSO_4$ and evaporated in vacuo. The crude product was purified by flash chromatography (petroleum ether/ethyl acetate = 2:1): yield 3.33 g (71%); colorless oil; $[\alpha]^{22}_D = -23.3$ ($c = 1.0$, $CHCl_3$); 200-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 6.41$ (d, $J = 7.7$ Hz, 1H, NH Phe), 5.07 (d, $J = 8.8$ Hz, 1H, NH Val), 4.87 (m, 1H, α-CH), 3.90 (m, 1H, α-CH), 3.10 (d, $J = 5.8$ Hz, 2H, β-CH_2 Phe), 2.13−2.00 (m, 1H, β-CH Val), 1.41 (s, 9H, $C(CH_3)_3$), 0.89 (d, $J = 6.9$ Hz, 3H, CH_3 Val), 0.83 (d, $J = 6.9$ Hz, 3H, CH_3 Val). Anal. Calcd for $C_{24}H_{38}N_2O_7$: C, 61.78; H, 8.21; N, 6.00. Found: C, 61.69; H, 8.29; N, 6.11.

***N*-(Allyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serylvalylphenylalanine (Methoxyethoxy)ethyl Ester (15). (a) N-Terminal Deprotection of the Dipeptide 14.** To a solution of **14** (933 mg, 2 mmol) in dry ether (10 mL) was added a dry saturated solution of HCl in ether (15 mL). The mixture was stirred for 4 h at rt and evaporated in vacuo, and the residue was extracted three times with diethyl ether, dried in vacuo, and used for the subsequent coupling without purification. Yield: 703 mg (87%).

(b) Coupling. To a solution of the *O*-glycosyl amino acid **12a** (340 mg, 655 μmol) in 5 mL of dry DMF at 0 °C were added *O*-[(cyano(ethoxycarbonyl)methylidene)amino]-1,1,3,3-tetramethyluroniumtetrafluoroborate[16] (TOTU, 215 mg, 655 μmol), 1-hydroxybenzotriazole (210 mg, 1.55 mmol), and *N*-ethyldiisopropylamine (85 mg, 656 mmol). The solution was added to a stirred solution of the N-terminal-deprotected dipeptide **14** and *N*-ethyldiisopropylamine (85 mg, 656 μmol) in dry DMF (5 mL), stirred for 1 h at rt, diluted with CH_2Cl_2 (50 mL), was washed with 0.1 N HCl (20 mL), saturated $NaHCO_3$ (20 mL), and water. The organic layer was dried with $MgSO_4$, filtered, and evaporated in vacuo. The crude product was purified by flash chromatography (petroleum ether/ethyl acetate = 1:10): yield 380 mg (67%); colorless amorphous solid; $[\alpha]^{22}_D = +65.2$ ($c = 0.8$, $CHCl_3$); $R_f = 0.64$ (ethyl acetate/methanol = 2:1); 400-MHz ^{1}H-NMR [^{1}H−^{1}H-COSY] ($CDCl_3$) $\delta = 7.01$ (d, $J = 9.2$ Hz, 1H, NH-Ac), 6.95 (d, $J = 8.3$ Hz, 1H, NH Val), 6.76 (d, $J = 7.6$ Hz, 1H, NH Phe), 5.98 (d, $J = 7.6$ Hz, 1H, NH Ser), 5.32 (d, 1H, 4-H), 5.02 (dd, $J_{2,3} = 11.5$ Hz, $J_{3,4} = 3.2$ Hz, 1H, 3-H), 4.91 (d, $J_{1,2} = 3.6$ Hz, 1H, 1-H), 4.79 (m, 1H, α-CH Phe), 4.38 (m, 1H, α-CH Ser), 4.30 (dd, $J_1 = 6.4$ Hz, $J_2 = 8.3$ Hz, 1H, α-CH Val), 3.97−4.09 (m, 3H, 5-H, 6-Ha/b), 3.79 (dd, $J_1 = 4.8$ Hz, $J_2 = 10.6$ Hz, 1H, β-CH_{2a} Ser), 3.65 (dd, $J_1 = 7.2$ Hz, $J_2 = 10.6$ Hz, 1H, β-CH_{2b} Ser), 3.12 (dd, $J_1 = 6.0$ Hz, $J_2 = 13.8$ Hz, 1H, β-CH_{2a} Phe), 3.06 (dd, $J_1 = 6.6$ Hz, $J_2 = 13.8$ Hz, 1H, β-CH_{2b} Phe), 2.08−1.92 (m, 1H, β-CH Val), 0.88 (d, $J = 6.7$ Hz, 3H, CH_3 Val), 0.85 (d, $J = 6.8$ Hz, 3H, CH_3 Val); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 98.48$ (C-1), 71.80, 70.35, 68.68, 67.72, 66.05, 64.33 (OCH_2 Aloc, 4 OCH_2 MEE, β-C Ser), 68.00, 67.17, 67.02 (C-3, C-4, C-5), 61.83 (C-6), 58.92, 58.28, 53.92, 53.29, 47.44 (OCH_3 MEE, α-C Phe, α-C Val, α-C Ser, C-2), 37.58 (β-C Phe), 31.73 (β-C Val), 18.89, 17.91 (CH_3 Val). Anal. Calcd for $C_{40}H_{58}N_4O_{17}$: C, 55.42; H, 6.74; N, 6.46. Found: C, 55.27; H, 6.84; N, 6.30.

***N*-(Allyloxycarbonyl)-*O*-(2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl)-L-serylvalylphenylalanine (16).** A solution of compound **15** (60 mg, 69.2 μmol) in 0.5 mL of acetone was added to a solution of lipase M (*M. javanicus*, Amano) in phosphate buffer (10 mL) and shaken for 16 h at 37 °C. The purification was carried out as described for **12/13**: yield 38 mg (72%); colorless amorphous solid; $[\alpha]^{22}_D = +71.3$ ($c = 1.2$, $CHCl_3$); $R_f = 0.33$ (ethyl acetate/methanol = 2:1); 200-MHz ^{1}H-NMR ($CDCl_3$) $\delta = 7.43-7.03$ (m, 8H, arom H, 3 NH), 6.00−5.81 (m, 2H, NH, =CH−), 5.04 (dd, $J_{2,3} = 11.5$ Hz, $J_{3,4} = 2.9$ Hz, 1H, 3-H), 4.80 (m, 1H, α-CH Phe), 4.72 (d, $J_{1,2} = 3.2$ Hz, 1H, 1-H), 4.50−4.35 (m, 2H, α-CH Ser, α-CH Val), 4.07 (m, 3H, 5-H, 6-Ha/b), 3.70−3.5 (m, 2H, β-CH_2 Ser), 3.10−2.85 (m, 2H, β-CH_2 Phe), 0.82 (t, $J = 6$ Hz, 6H, 2 CH_3 Val); FAB-MS $(M - H)^-$ 763.3. Anal. Calcd for $C_{35}H_{48}N_4O_{15}\cdot 1.5H_2O$ (764.8): C, 53.09; H, 6.49; N, 7.08. Found: C, 52.95; H, 6.59; N, 7.24.

General Procedure for the Removal of the *O*-Acetyl Groups from *O*-Glycosyl Amino Acid MEE Esters with Lipase WG from Wheat Germ 17−19. Glycosyl amino acid MEE ester **7/11** (2 mmol) was dissolved in acetone (1.5 mL), added to a solution of 300 mg of lipase WG (Sigma) in phosphate buffer (36 mL), shaken for 2 d or 18 h at 37 °C, and then lyophilized. The product was extracted three times with 30 mL of ethanol. After drying with $MgSO_4$ and evaporation, the residue was purified by flash chromatography (ethyl acetate/petroleum ether = 3:1 → ethyl acetate → ethyl acetate/ethanol = 1:10).

***N*-(Allyloxycarbonyl)-*O*-(2-acetamido-2-deoxy-α-D-galactopyranosyl)-L-serine (methoxyethoxy)ethyl ester (17a):** yield 722 mg (73%, reaction time 2 d); colorless amorphous solid; $[\alpha]^{22}_D = +83.7$ ($c = 1.0$, $CHCl_3$); $R_f = 0.71$ (ethyl acetate/methanol = 2:1); 400-MHz ^{1}H-NMR (DMSO-d_6) $\delta = 7.69$ (d, $J = 8.7$ Hz, 1H, NH), 7.00 (d, $J = 8.9$ Hz, 1H, NH Ser), 4.58 (d, $J_{1,2} = 3.7$ Hz, 1H, 1-H), 4.56 (m, 2H, 2 OH), 4.44 (d, $J = 6.5$ Hz, 1H, OH), 4.33 (m, 1H, α-CH), 4.00−3.96 (m, 1H, 2-H), 3.71−3.40 (m, 13H, 5-H, 4-H, 3-H, 6-Ha/b, β-CH_2 Ser, 3 OCH_2 MEE); 100.6-MHz ^{13}C-NMR ($CDCl_3$) $\delta = 99.97$ (C-1), 72.88, 71.26, 69.81, 69.01, 66.79, 65.70 (OCH_2 Aloc, 4 OCH_2 MEE, β-CH_2 Ser), 72.80, 70.25, 69.57 (C-3, C-4, C-5), 62.74 (C-6), 55.95 (α-CH Ser), 51.36 (C-2). Anal. Calcd for $C_{20}H_{34}N_2O_{12}\cdot 0.5H_2O$: C, 47.71; H, 7.01; N, 5.56. Found: C, 47.69; H, 7.12; N, 5.87.

(20) Belleau, B.; Malek, G. *J. Am. Chem. Soc.* **1968**, *90*, 1651.

***N*-(9-Fluorenylmethyloxycarbonyl)-*O*-(2-acetamido-2-deoxy-α-D-galactopyranosyl)-L-serine (methoxyethoxy)-ethyl ester (17b):** yield 1.09 g (86%, reaction time 2 d); colorless crystals; mp 158−160 °C; $[\alpha]^{22}_D = +83.0$ (c = 1.0, $CHCl_3$); R_f = 0.55 (ethyl acetate/methanol = 2:1); 400-MHz ^{1}H-NMR (DMSO-d_6), characteristic signals after D_2O-exchange δ = 4.60 (d, $J_{1,2}$ = 3.6 Hz, 1H, 1-H), 4.33 (m, 3H, OCH_2 Fmoc, α-CH Ser), 4.24 (t, 1H, Fmoc-CH), 4.11 (m, 1H, 2-H); 100.6-MHz ^{13}C-NMR (CD_3OD) δ = 99.94 (C-1), 72.78, 71.18, 69.73, 68.93, 67.95, 65.65 (OCH_2 Fmoc, 4 OCH_2 MEE, β-CH_2 Ser), 72.75, 70.20, 69.69 (C-3, C-4, C-5), 62.73 (C-6), 55.96 (α-CH Ser), 51.32 (C-2). Anal. Calcd for $C_{31}H_{40}N_2O_{12}\cdot 0.5H_2O$: C, 58.03; H, 6.48; N, 4.37. Found: C, 58.23; H, 6.48; N, 4.05.

***N*-(Allyloxycarbonyl)-*O*-(β-D-glucopyranosyl)-L-serine (methoxyethoxy)ethyl ester (18a):** yield 434 mg (48%, reaction time 2d); 696 mg (77%, reaction time 18 h); colorless oil; $[\alpha]^{22}_D = -14.1$ (c = 2.6, CH_3OH); R_f = 0.50 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.41 (m, J_1 = 3.7 Hz, J_2 = 3.7 Hz, 1H, α-CH Ser), 4.41 (dd, J_1 = 4.1 Hz, J_2 = 10.0 Hz, 1H, β-CH_{2a} Ser), 4.30 (d, $J_{1,2}$ = 7.7 Hz, 1H, 1-H), 3.90 (d, $J_{6a,6b}$ = 11.6 Hz, 1H, 6-Ha), 3.81 (dd, J_1 = 3.5 Hz, J_2 = 10.0 Hz, 1H, β-CH_{2b} Ser), 3.71 (m, 1H, 5-H), 3.42−3.37 (m, 1H, 6-Hb), 3.31−3.28 (m, 2H, 3-H, 4-H), 3.21 (dd, $J_{2,1}$ = 7.8 Hz, $J_{2,3}$ = 8.9 Hz, 1H, 2-H); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 104.55 (C-1), 78.03, 77.82, 74.99, 71.50 (C-2, C-3, C-4, C-5), 70.50 (β-CH_2 Ser), 62.69 (C-6), 55.84 (α-CH Ser); $C_{18}H_{31}NO_{12}$ (453.3), FAB-MS (M − H)$^-$ 452.2.

***N*-(Allyloxycarbonyl)-*O*-(β-D-glucopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (18b):** yield 710 mg (76%, reaction time 2 d); colorless oil; $[\alpha]^{22}_D = -10.1$ (c = 2.7, CH_3OH); R_f = 0.55 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.60 (m, 2H, α-CH Thr, β-CH Thr), 4.36 (d, $J_{1,2}$ = 7.6 Hz, 1H, 1-H), 3.89 (d, $J_{6a,6b}$ = 11.5 Hz, 1H, 6-Ha), 3.70 (m, 1H, 5-H), 3.39−3.35 (m, 1H, 6-Hb), 3.35−3.29 (m, 2H, 3-H, 4-H), 3.15 (dd, $J_{2,1}$ = 7.8 Hz, $J_{2,3}$ = 9.0 Hz, 1H, 2-H), 1.30 (d, J = 6.3 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 101.02 (C-1), 77.89, 77.73, 74.90, 71.56 (C-2, C-3, C-4, C-5), 73.99 (β-CH Thr), 62.75 (C-6), 60.44 (α-CH Thr), 16.63 (γ-CH_3 Thr); $C_{19}H_{33}NO_{12}$ (467.3); FAB-MS (M − H)$^-$ 466.1.

***N*-(Allyloxycarbonyl)-*O*-(β-D-galactopyranosyl)-L-serine (methoxyethoxy)ethyl ester (18c):** yield 372 mg (41%, reaction time 2 d); 626 mg (69%, reaction time 18 h); colorless oil; $[\alpha]^{22}_D = -2.7$ (c = 1.9, CH_3OH); R_f = 0.62 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.51 (m, J_1 = 3.7 Hz, J_2 = 3.6 Hz, 1H, α-CH Ser), 4.41 (dd, J_1 = 4.0 Hz, J_2 = 10.0 Hz, 1H, β-CH_{2a} Ser), 4.26 (d, $J_{1,2}$ = 7.0 Hz, 1H, 1-H), 3.81 (dd, J_1 = 3.5 Hz, J_2 = 10.0 Hz, 1H, β-CH_{2b} Ser), 3.86−3.28 (m, 6H, 2-H, 3-H, 4-H, 5-H, 6-Ha, 6-Hb); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 105.23 (C-1), 76.84, 74.83, 72.49, 70.25 (C-2, C-3, C-4, C-5), 70.50 (β-CH_2 Ser), 62.48 (C-6), 55.91 (α-CH Ser); $C_{18}H_{31}NO_{12}$ (453.2); FAB-MS (M + H)$^+$ 454.0.

***N*-(Allyloxycarbonyl)-*O*-(β-D-galactopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (18d):** yield 523 mg (56%, reaction time 2 d); colorless oil; $[\alpha]^{22}_D = -20.5$ (c = 2.1, CH_3OH); R_f = 0.64 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.60 (m, 1H, β-CH Thr), 4.33 (m, 1H, α-CH Thr), 4.31 (d, $J_{1,2}$ = 6.5 Hz, 1H, 1-H), 3.86−3.59 (m, 6H, 2-H, 3-H, 4-H, 5-H, 6-Ha, 6-Hb), 1.30 (d, J = 6.3 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 101.70 (C-1), 76.49, 74.54, 72.21, 69.99 (C-2, C-3, C-4, C-5), 73.94 (β-CH Thr), 62.15 (C-6), 60.28 (α-CH Thr), 16.65 (γ-CH_3 Thr); $C_{19}H_{33}NO_{12}$ (467.3); FAB-MS (M − H)$^-$ 466.1.

***N*-(Allyloxycarbonyl)-*O*-(β-D-xylopyranosyl)-L-serine (methoxyethoxy)ethyl ester (18e):** yield 432 mg (51%, reaction time 2 d); colorless oil; $[\alpha]^{22}_D = -5.5$ (c = 2.1, CH_3OH); R_f = 0.63 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.51 (m, J_1 = 3.5 Hz, J_2 = 3.5 Hz, 1H, α-CH Ser), 4.36 (dd, J_1 = 3.6 Hz, J_2 = 10.7 Hz, 1H, β-CH_{2a} Ser), 4.23 (d, $J_{1,2}$ = 7.5 Hz, 1H, 1-H), 3.87 (d, $J_{5a,5b}$ = 11.3 Hz, $J_{5a,4}$ = 5.3 Hz, 1H, 5-Ha), 3.81 (dd, J_1 = 3.4 Hz, J_2 = 9.9 Hz, 1H, β-CH_{2b} Ser), 3.48 (dd, $J_{4,5}$ = 5.3 Hz, $J_{4,3}$ = 8.8 Hz, 1H, 4-H), 3.34 (m, 1H, 5-Hb), 3.25−3.18 (m, $J_{2,1}$ = 7.7 Hz, $J_{3,4}$ = 10.3 Hz, 2H, 3-H, 2-H); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 104.33 (C-1); 76.78, 74.00, 70.24 (C-2, C-3, C-4), 70.34 (β-CH_2 Ser), 64.99 (C-5), 55.83 (α-CH Ser); $C_{17}H_{29}NO_{11}$ (423.4); FAB-MS (M + H)$^+$ 424.4.

***N*-(Allyloxycarbonyl)-*O*-(β-D-xylopyranosyl)-L-threonine (methoxyethoxy)ethyl ester (18f):** yield 537 mg (61%, reaction time 2 d); 590 mg (67%, reaction time 18 h); colorless oil; $[\alpha]^{22}_D = -5.4$ (c = 1.8, CH_3OH); R_f = 0.52 (ethyl acetate/ethanol = 2:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.54 (m, J_1 = 2.4 Hz, J_2 = 6.3 Hz, 2H, α-CH Thr, β-CH Thr), 4.28 (d, $J_{1,2}$ = 7.8 Hz, 1H, 1-H), 3.84 (dd, $J_{5a,5b}$ = 11.3 Hz, $J_{5a,4}$ = 5.3 Hz, 1H, 5-Ha), 3.49 (m, $J_{4,5a}$ = 5.1 Hz, $J_{4,5b}$ = 5.3 Hz, $J_{4,3}$ = 10.1 Hz, 1H, 4-H), 3.36−3.34 (m, 1H, 5-Hb), 3.22−3.12 (m, $J_{3,4}$ = 11.4 Hz, $J_{2,1}$ = 7.6 Hz, 2H, 3-H, 2-H), 1.29 (d, J = 6.3 Hz, 3H, γ-CH_3 Thr); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 101.66 (C-1), 77.39, 74.53, 71.00 (C-2, C-3, C-4), 73.87 (β-CH Thr), 66.60 (C-5), 60.15 (α-CH Thr), 16.46 (γ-CH_3 Thr); $C_{18}H_{31}NO_{11}$ (437.4); FAB-MS (M − H)$^-$ 436.1.

Cleavage of the *O*-Acetyl Groups and the (Methoxyethoxy)ethyl Ester from *O*-Glycosyl Amino Acid MEE Esters with Lipase WG from Wheat Germ 19. The reaction was carried out as described for the preparation of **17/18**, but the reaction time was prolonged to 5 d.

***N*-(Allyloxycarbonyl)-*O*-(β-D-xylopyranosyl)-L-serine (19):** yield 559 mg (87%); colorless oil; $[\alpha]^{22}_D = -3.8$ (c = 1.0, CH_3OH); R_f = 0.55 (propanol/AcOH/H_2O = 30:4:1); 400 MHz ^{1}H-NMR (CD_3OD) δ = 4.27 (m, J = 4.3 Hz, 1H, α-CH Ser), 4.28−4.24 (m, 1H, β-CH_{2a} Ser), 4.24 (d, $J_{1,2}$ = 7.3 Hz, 1H, 1-H), 3.88 (d, $J_{5a,5b}$ = 11.4 Hz, $J_{5a,4}$ = 5.2 Hz, 1H, 5-Ha), 3.74 (m, 1H, β-CH_{2b} Ser), 3.50 (dd, $J_{4,5}$ = 5.3 Hz, $J_{4,3}$ = 8.8 Hz, 1H, 4-H), 3.37−3.34 (m, 1H, 5-Hb), 3.25−3.19 (m, 2H, 3-H, 2-H); 100.6 MHz ^{13}C-NMR (CD_3OD) δ = 105.34 (C-1), 77.35, 74.66, 71.13 (C-2, C-3, C-4), 71.92 (β-CH_2 Ser), 66.85 (C-5), 49.84 (α-CH Ser); $C_{12}H_{19}NO_9$ (321.2); FAB-MS (M + H)$^+$ 322.0.

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