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Synthesis of the N-oxyamide-linked glycolipids and glycopeptides

Na Chen

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Na Chen. Synthesis of the N-oxyamide-linked glycolipids and glycopeptides. Other. Université Paris Saclay (COMUE), 2015. English. NNT : 2015SACLN017 . tel-01245320

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NNT : 2005SACLN017

**THESE DE DOCTORAT
DE L'UNIVERSITE PARIS-SACLAY
Préparée à l'École Normale Supérieure de Cachan**

ÉCOLE DOCTORALE N°571
Sciences chimiques : molécules, matériaux, instrumentation et biosystèmes
Spécialité de doctorat : Chimie

Par

Mme Na CHEN

Synthesis of *N*-oxyamide-linked glycolipids and glycopeptides

Thèse présentée et soutenue à Cachan, le 9 décembre 2015 :

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Acknowledgement

This dissertation is accomplished in the “Laboratoire de Photophysique et Photochimie Supramoléculaires et Macromoléculaires (PPSM)” in École Normale Supérieure de Cachan (ENS Cachan). Here I must express my sincere gratitude to all the people for giving me the help to make this dissertation possible and particularly the directors of PPSM Prof. Keitaro Nakatani, Prof. Fabien Miomandre and Dr. Isabelle Leray.

First and foremost I wish to thank my advisor, Prof. Joanne Xie, for her help both in work and in life. She offered me the opportunity to work in ENS Cachan and gave me very professional guidance in this dissertation. I am deeply grateful of her help in the completion of this manuscript. Her gamut of knowledge, infectious enthusiasm and high efficient in the work will always influence me. In daily life, she is so kind to me and invited me to her house for lunch or dinner several times.

For this dissertation I would like to thank the committee members: Dr. Christine Gravier-Pelletier, Prof. Arnaud Haudrechy and Prof. Olivier Martin for their time, interest, and comments.

A special acknowledgement goes to Prof. Yunbao Jiang in Xiamen University, who is my science mentor in China. His insightful knowledge, confident prediction and unselfish supporting inspire me move on during these years. Thanks for his support and help in the application of the scholarship, the funding that made my Ph.D. work possible.

I am indebted to all the kind and lovely members in Xie’s research group, and all of them make the lab like a family. The group has been a source of friendships as well as good advice and collaborations. Thanks very much for the humor of Dr. Nicolas Bogliotti, the hearty laughter of Claire DEO, the warmth of Dr. Nhi-Ha Nguyen, the kindness of Dr. Olivier Noël, Dr. Laura Nodin, and Chaoqi Lin. Special thanks go to Stéphane Maisonneuve, my best friend in the lab. We shared all the laboratory equipment and supplies and he taught me how to measure the NMR, melting point, optical rotation, etc. He also provided insightful discussions and suggestions about the research. Moreover, he is also a guarantor for me in Crous.

I would like to thank all the faculty members and staff in PPSM: Dr. Rémi Métivier, Prof. Pierre Audebert, Dr. Robert Pansu, Dr. Valérie Alain-Rizzo, Dr. Clémence Allain, Dr. Gilles Clavier, Dr. Cécile Dumas-Verdes, Dr. Valérie Génot, Dr. Carine Julien-Rabant, Prof. Rachel Méallet-Renault, Dr. Laurent Galmiche, Andrée Husson, Christian Jean-Baptiste, Arnaud Brosseau. Thanks to Jacky Formont for handling all the problems of my computer.

I would also like to acknowledge the members of PPSM: Dr. Haitao Zhang, Dr. Yuanyuan Liao, Dr. Eva Julien, Dr. Alex Depauw, Dr. Jia Su, Meriem Stamboul, Dr. Yang Si, Yuan Li, Paul Rouschmeyer, Charlotte Remy, Clarisse Tourbillon, Corentin Pavageau, Marine Louis, Dr. Naresh Kumar, Yayang Tian, Qui Pham and Etienne Barrez. Thanks for your kind-

ness to offer a very harmonious study environment.

Words alone cannot express my thanks to Dr. Yanhua Yu, who treats me like a sister. She has been a source of love and energy ever since. Many thanks to Dr. Yibin Ruan, he is my labmate both in Xiamen University and ENS Cachan. He helped me a lot in work and daily life, such as picking me up at the airport, offering me a lot of stuffs. Thanks again to Yuan Li for the help and accompany, which make me feel I am not alone in France. Thanks again to Yayang Tian for her help in daily life. I also would like to thank all my friends in ENS Cachan.

I am forever grateful to the care and support from my close friend, Su Chen. We worked on our manuscript during the same time, so she gave me a lot of understanding, accompanies, and encouragement. It is a precious memory for me forever. Thanks to Yuan Yang for all the help in daily life. I am so lucky to be as your neighbor. Thanks to all my friends in France and in China. A faithful friend is a source of strength, and all of you give me the strength.

Finally, my deepest gratitude goes to my family for their unflagging love and encouragement. Thanks to my parents who raised me with a love of science and supported me in all my pursuits, and thanks to my lovely brother.

Na CHEN

ENS Cachan

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Abbreviations

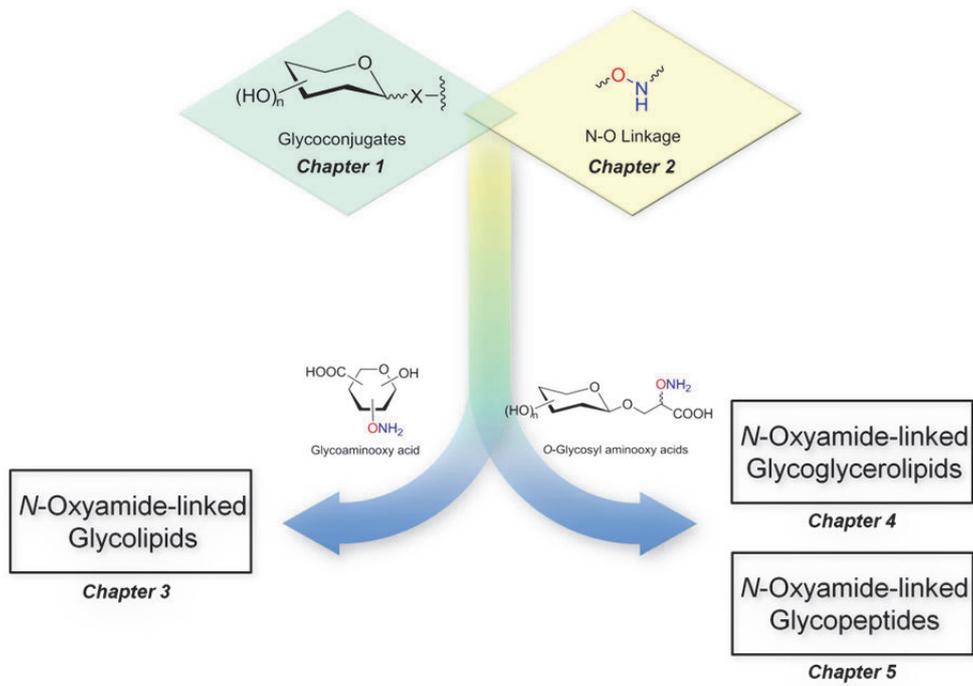
Ac	Acetyl
AGLs	Agelasphins
AIBN	azobisisobutyronitrile
Ala	Alanine
Akt	Protein kinase B
Ar	aromatic
Asn	Asparagine
Asp	Aspartic acid
ATPase	Adenosine 5'-triphosphatase
BA	Betulinic acid
Bn	Benzyl
bNAbs	Broadly neutralizing antibodies
Boc	<i>tert</i> -Butoxycarbonyl
Bu	Butyl
CAN	Ceric ammonium nitrate
CD	Circular dichroism
Cer	Ceramide
COSY	Collision induced dissociation
C-SA	<i>South African</i> subtype <i>C</i>
CSA	Camphor sulfonic acid
Cys	Cysteine
d	Doublet
DBU	1,8-Diazabicycloundec-7-ene
DCC	<i>N,N'</i> -Dicyclohexylcarbodiimide
dd	Doublet and Doublet
DDQ	2,3-Dichloro-5,6-dicyano-1,4-benzoquinone
DEAD	Diethyl azodicarboxylate
Dept	Distortionless Enhancement by Polarization Transfer
DGDG	Digalactosyl diacylglycerol
DIAD	Diisopropyl azodicarboxylate
DMAP	4-Dimethylaminopyridine

DMF	<i>N,N</i> -Dimethylformamide
DNA	Deoxyribonucleic acid
EDC·HCl	<i>N</i> -Ethyl- <i>N'</i> -(3-dimethylaminopropyl)carbodiimide HCl
EPL	Expressed protein ligation
eq	Equivalent
Et	Ethyl
Fc	Crystallizable fragment
Fmoc	<i>N</i> -9-Fluorenylmethyloxycarbonyl
Fmoc-OSu	<i>N</i> -(9-Fluorenylmethoxycarbonyloxy)succinimide
FT-IR	Fourier transform infrared spectroscopy
g	Gram
Gal	Galactosyl
GalCer	Galactosyl ceramide
GalNAc	<i>N</i> -acetylgalactosamine
GGLs	Glycoglycerolipids
Glc	Glucosyl
GlcADG	Glucuronosyldiacylglycerol
Gly	Glycine
GSLs	Glycosphingolipids
h	hour
HETCOR	Heteronuclear Correlation
HIV	Human immunodeficiency virus
HMQC	Heteronuclear Multiple Quantum Coherence
HOBt	Hydroxybenzotriazole
HPLC	High-performance liquid chromatography
HRMS	High-resolution mass spectrometry
<i>i</i>-Bu	Isobutyl
IC50	50% Inhibition concentration
IFN-γ	Interferon- γ
IgG1	Immunoglobulin G1
IL-4	Interleukin 4
<i>i</i>NKT	Invariant natural Killer T
<i>i</i>Pr	Isopropyl
IVA	Influenza A virus
<i>J</i>	Coupling constant

L	Liter
LacCer	Lactosylceramide
LG	Leaving group
M	Molar
m	Multiplet
MD	Molecular dynamics
MGDG	Monogalactosyl diacylglycerol
Min	Minute
mL	milliliter
mM	Millimolar
mmol	Millimole
Myt1	Myelin transcription factor 1
NCL	Native chemical ligation
NCS	<i>N</i> -chlorosuccinimide
NHS or HOSu	<i>N</i> -hydroxysuccinimide
NMR	Nuclear magnetic resonance
NOE	Nuclear Overhauser Effect
NOESY	Nuclear Overhauser Effect Spectroscopy
NPs	Nanoparticles
NuH	Nucleophile
OVA	Ovalbumin
PCC	Pyridinium chlorochromate
PG	Protecting group
Ph	Phenyl
Phth	Phthaloyl
PhthN-OH	<i>N</i> -hydroxyphthalimide
PI3P	3-Phosphorylated phosphatidylinositol
PMB	<i>p</i> -Methoxybenzyl
ppm	Parts-per-million (10^{-6})
PTC	Phase transfer catalyzed
Pyr	Pyridine
q	Quartet
s	singlet
Ser	Serine
S_N	Nucleophilic substitution

SPPS	Solid phase peptide synthesis
SQDG	Sulfoquinovosyl diacylglycerol
t	Triplet
TBABr	Tetra- <i>n</i> -butylammonium bromide
TBAF	Tetra- <i>n</i> -butylammonium fluoride
TBAHS	Tetra- <i>n</i> -butylammonium hydrogen sulfate
TBAI	Tetra- <i>n</i> -butylammonium iodide
TBS	<i>tert</i> -Butyldimethylsilyl
<i>t</i>-Bu	<i>Tert</i> -butyl
TFA	Trifluoroacetic acid
Tf₂O	Trifluoromethanesulfonic anhydride
Th1	T helper 1
THF	Tetrahydrofuran
TLC	Thin layer chromatography
TMSOTf	Trimethylsilyl trifluoromethanesulfonate
Tn	GalNAc- <i>O</i> -
Tr	Triphenylmethyl
RNA	Ribonucleic acid
Thr	Threonine
Trp	Tryptophan
Tyr	Tyrosine
μM	Micromolar
UV	Ultraviolet
Val	Valine
VRE	Vancomycin-resistant <i>Enterococci</i>
δ	Chemical shift expressed in ppm

General Introduction



As part of glycoconjugate family, glycolipids and glycopeptides are involved in a variety of important biological, physiological and pathological processes, such as cell-cell interactions, viral and bacterial infections, immune response, cancer progression, etc. Synthesis of glycoconjugate mimics has attracted intensive research interest for biological and pharmaceutical applications, which has heightened the need for developing new types of glycoconjugates.

Aminoxy acids, a class of unnatural amino acids in which the amine group is replaced by an aminoxy function, have emerged as very attractive scaffolds, since the peptides containing aminoxy acids can easily organize into turns and helices through intramolecular hydrogen bond formation. Furthermore, the *N*-oxypeptides are easily formed and resistant to chemical and enzymatic hydrolysis. These unique properties make *N*-oxyamide linkage attractive for the modification of biomolecules.

Recently, our group is interested in synthesizing *N*-oxyamide-containing compounds. As a continuing research program, we try to introduce the *N*-oxyamide function into glycolipids and glycopeptides to prepare novel glycoconjugates for studying their potential biological applications. This thesis is structured as follows:

The first chapter introduces the structures and functions of glycoconjugates. After presenting the synthetic strategy and stereochemistry of glycosidic bond, the synthesis and biological applications of glycopeptides/glycoproteins will be described. We will also introduce the classifications, structures, modifications and biological functions of glycolipids.

The second chapter attempts to review the N-O linkage in carbohydrate-based compounds. After introduction of aminoxy acids, the formation and applications of *N*-glycosyl hydroxylamines, *O*-glycosyl hydroxylamines and *O*-amino sugar derivatives will be presented.

The third chapter describes the synthesis of *N*-oxyamide-linked glycolipids. The glycoaminoxy ester, with both the carboxylic acid and aminoxy functions attached on the sugar frame, has been prepared as a multifunctional building block. Further derivatization of this glycoaminoxy ester leads to the formation of *N*-oxyamide-linked glycolipids.

The fourth chapter presents the synthesis of *N*-oxyamide-linked glycolipids with one or two lipids chains as novel mimics of glycolipids and glycosphingolipids.

In the last chapter, we focus on the stereoselective synthesis of *O*-glycosyl aminoxy esters and their use for the construction of *N*-oxyamide-linked glycopeptides.

Chapter 1

Structures and Functions of Glycoconjugates

I Introduction

Carbohydrates are implicated in a variety of important biological and physiological processes, such as biological transport, cell-cell recognition, activation of growth factors, modulation of the immune system, etc.¹ Carbohydrate-based compounds have been successfully applied in the biopharmaceutical and medical fields, especially in diagnostics, vaccines and therapeutics.²

There are three major classes of carbohydrates, which can be defined as monosaccharides, oligosaccharides and polysaccharides. Compounds with mono- or oligosaccharides covalently attached to a non-sugar moiety such as lipids, peptides and proteins through glycoside bond, are called glycoconjugates. The major glycoconjugates are glycopeptides, glycoproteins, glycolipids, and proteoglycans.

Many reports supported that the glycans, including oligosaccharides, polysaccharides, and glycoconjugates, can play a very important role in biological recognition processes.³ Now they could be reasonably added to the ‘central dogma molecules’ (nucleic acids, proteins), by playing a distinct role in information transfer (Figure 1).^{3a,4}

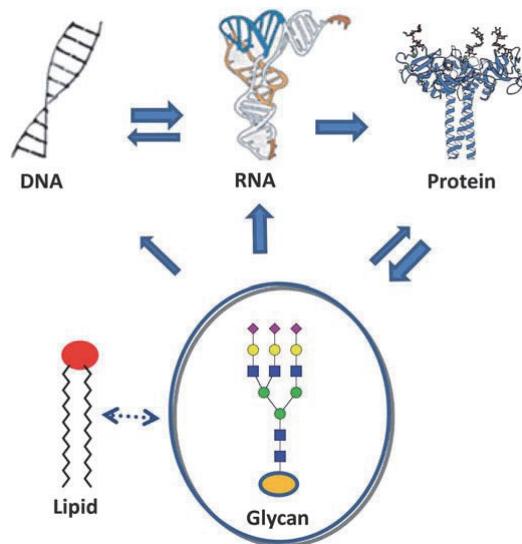


Figure 1: A modified central dogma of biological information flow.^{3a}

¹ (a) Wolfert, M. A.; Boons, G.-J. *Nat. Chem. Biol.* **2013**, *9*, 776-784; (b) Kolarich, D.; Lepenies, B.; Seeberger, P. H. *Curr. Opin. Chem. Biol.* **2012**, *16*, 214-220; (c) Rudd, P. M.; Elliott, T.; Cresswell, P.; Wilson, I. A.; Dwek, R. A. *Science* **2001**, *291*, 2370-2376; (d) Dwek, R. A. *Chem. Rev.* **1996**, *96*, 683-720.

² (a) Fernandez-Tejada, A.; Canada, F. J.; Jimenez-Barbero, J. *Chem. Eur. J.* **2015**, *21*, 10616-10628; (b) Fernandez-Tejada, A.; Canada, F. J.; Jimenez-Barbero, J. *ChemMedChem* **2015**, *10*, 1291-1295; (c) Hudak, J. E.; Bertozzi, C. R. *Chem. Biol.* **2014**, *21*, 16-37; (d) Galan, M. C.; Benito-Alifonso, D.; Watt, G. M. *Org. Biomol. Chem.* **2011**, *9*, 3598-3610; (e) Seeberger, P. H.; Werz, D. B. *Nature* **2007**, *446*, 1046-1051; (f) Freeze, H. H.; Westphal, V. *Biochimie* **2001**, *83*, 791-799; (g) Butters, T. D.; Dwek, R. A.; Platt, F. M. *Chem. Rev.* **2000**, *100*, 4683-4696.

³ (a) Wang, L. X.; Davis, B. G. *Chem. Sci.* **2013**, *4*, 3381-3394; (b) Cecioni, S.; Imberty, A.; Vidal, S. *Chem. Rev.* **2014**, *115*, 525-561; (c) Bertozzi, C. R.; Kiessling, L. L. *Science* **2001**, *291*, 2357-2364; (d) Herzner, H.; Reipen, T.; Schultz, M.; Kunz, H. *Chem. Rev.* **2000**, *100*, 4495-4538; (e) Davis, B. G. *J. Chem. Soc.; Perkin Trans. 1.* **1999**, 3215-3237.

⁴ Crick, F. *Nature*, **1970**, *227*, 561-563.

The branching structures and the stereochemistry of the glycosidic linkage result in the complexity of glycoconjugates.⁵ The stereocontrol in the glycoside formation is one of the most important aspects of oligosaccharides and glycoconjugates synthesis. In this chapter, we will focus on the glycosidic bond formation (glycosylation), as well as the structures, functions and synthesis of glycopeptides and glycolipids.

II Formation of glycosidic bonds

The chemistry of glycosylation reactions, which is a common and essential process in all organisms, has been developed most intensively in the past few decades.⁶ The glycosylation usually involves the coupling of a glycosyl donor and a suitable protected glycosyl acceptor via initiation using an activator under suitable reaction conditions (Figure 2). Examples of the products of glycosylation reactions are glycopeptides, glycolipids, glycosaminoglycans, oligosaccharides, and polysaccharides.

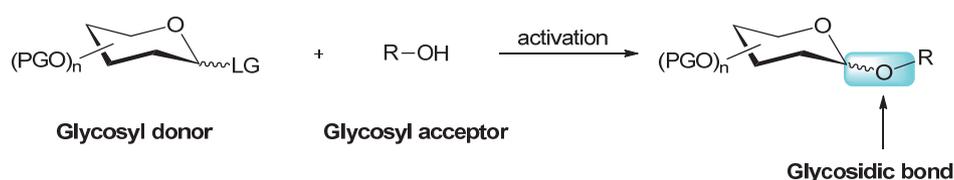


Figure 2: General chemical glycosylation. PG = protecting group, LG = leaving group.

II.1 Glycosylation strategies

The first glycosylation reaction was reported by Arthur Michael in 1879, which proceeded by the nucleophilic displacement of an anomeric leaving group (Scheme 1a).⁷ In 1893, Emil Fischer reported a new approach for the glycosylation reaction, in which an aldose or ketose was coupled with an alcohol or phenol catalyzed by an acid (Scheme 1b).⁸ The Koenigs-Knorr glycosylation, a substitution reaction by reacting a glycosyl halide with a conventional alcohol using the halophilic promoter (silver salts, mercury salts, or Lewis acid) to give a glycoside, is one of the oldest and most famous glycosylation reactions (Scheme 1c).⁹ Since then, a very large number of glycosylation methods,⁶ and the mechanism of gly-

⁵ Seeberger, P. H. *Nat. Chem. Biol.* **2009**, *5*, 368-372.

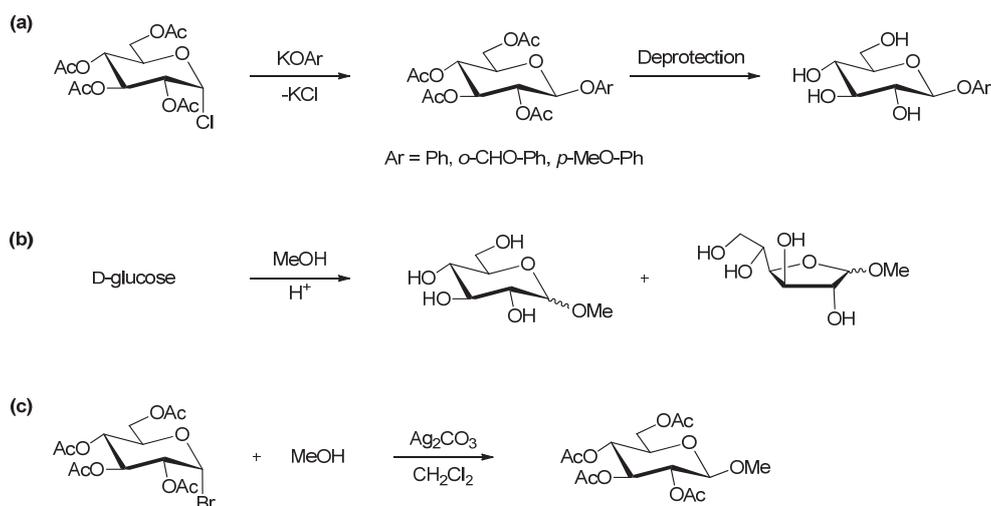
⁶ (a) Satoh, H.; Manabe, S. *Chem. Soc. Rev.* **2013**, *42*, 4297-4309; (b) Ishiwata, A.; Lee, Y. J.; Ito, Y. *Org. Biomol. Chem.* **2010**, *8*, 3596-3608; (c) Zhu, X.; Schmidt, R. R. *Angew. Chem. Int. Ed.* **2009**, *48*, 1900-1934; (d) Demchenko, A. V. *Handbook of Chemical Glycosylation*, Wiley-VCH, Weinheim, **2008**; (e) Galonic, D. P.; Gin, D. Y. *Nature* **2007**, *446*, 1000-1007; (f) Toshima, K. *Carbohydr. Res.* **2006**, *341*, 1282-1297; (g) Demchenko, A. V. *Synlett* **2003**, 1225-1240; (h) Schmidt, R. R.; Kinzy, W. *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 21-123.

⁷ Michael, A. *Am. Chem. J.* **1879**, *1*, 305-312.

⁸ Fischer, E. *Ber. Dtsch. Chem. Ges.* **1893**, *26*, 2400-2412.

⁹ Koenigs, W.; Knorr, E. *Ber. Dtsch. Chem. Ges.* **1901**, *34*, 957-981.

cosylation¹⁰ have been developed and reviewed.



Scheme 1: (a) Michael, (b) Fisher and (c) Koenigs-Knorr glycosylation reactions.^{10b}

Most of the glycosylation methods involving different glycosyl donors, could be named by the functionalities of the glycosyl donors, such as trichloroacetimidate method, phosphite method, thioglycoside, etc. Galonic *et al.* have summarized the glycosylation strategies.^{6e} The general glycosylation approach usually involves the coupling of a selectively protected glycosyl donor with an anomeric leaving group (LG) (**1**, Figure 3a) and a suitable protected glycosyl acceptor via initiation using a suitable electrophilic activator (EI⁺) under suitable reaction conditions to generate the glycoconjugate (**3**).

The glycosyl donors and the corresponding electrophilic activator for the acetal exchange reactions were listed in Figure 3b. The glycosyl donors mainly contain: anomeric hydroxyl group (**4**); glycosyl halides including fluorides, chlorides, bromides and iodides (**5**); trichloroacetimidates (**6**); phosphates/phosphites (**7**); esters/carbonates/thiocarbonates (**8**); aryloxy groups (**9**); 4-pentenyl glycosides (**10**); glycosyl sulphides and sulphoxides (**11**).

The glycal assembly approach has been developed extensively in the synthesis of complex carbohydrates. The 1,2-alkene functionality enables various electrophilic oxidants (EI⁺) reacting with enol ether nucleophiles (**12**) to generate a three-membered ring, which is an activated glycosyl donor (**13**). Opening of the epoxide ring by an appropriate nucleophilic glycosyl acceptor (Nu-H) furnished the corresponding glycoconjugate (**14**), along with the introduction of functionalities 'Z' at the C2-position (Figure 3c). Various electrophilic oxidants and the correspondingly generated glycosides are listed in Figure 3d.

¹⁰ (a) Frihed, T. G.; Bols, M.; Pedersen, C. M. *Chem. Rev.* **2015**, *115*, 4963-5013; (b) Mydock, L. K.; Demchenko, A. V. *Org. Biomol. Chem.* **2010**, *8*, 497-510.

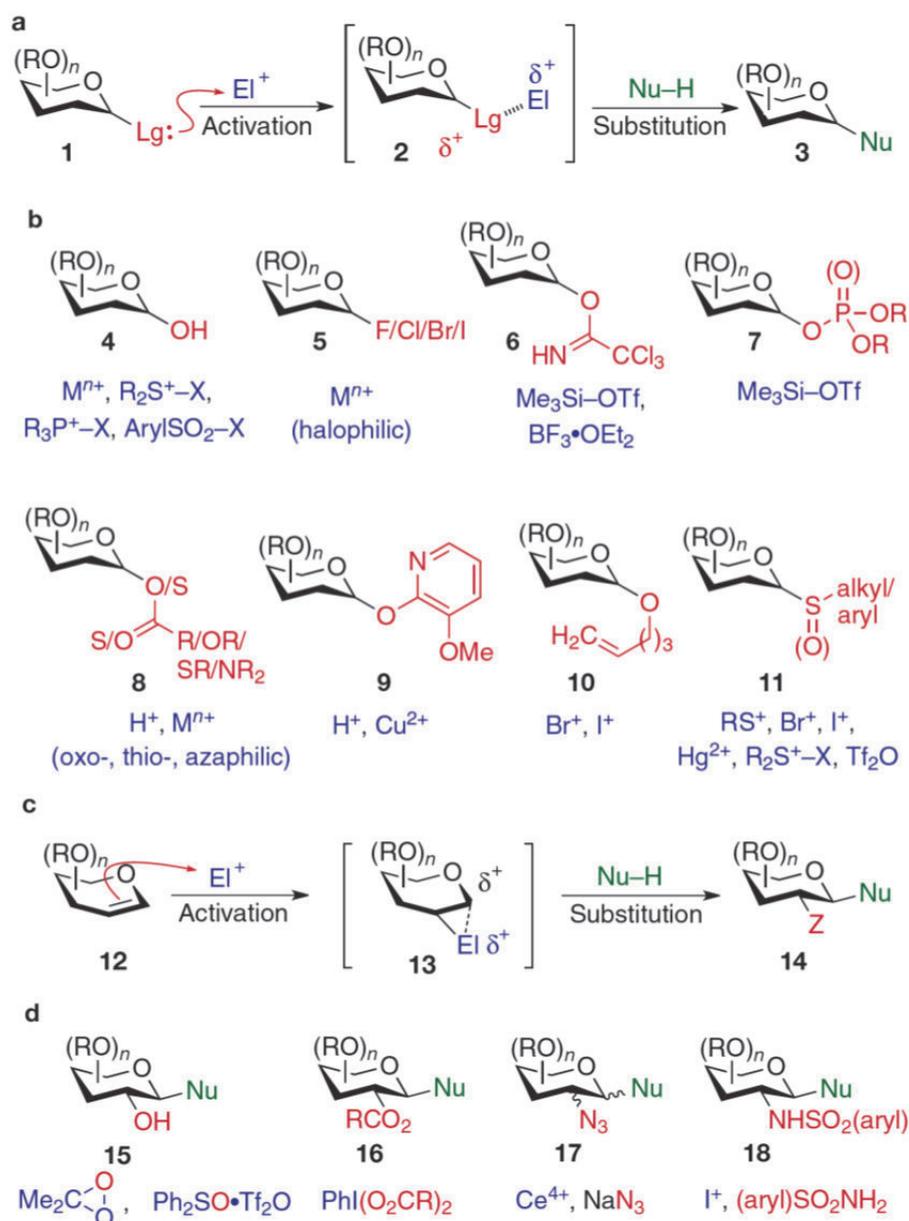


Figure 3: Glycosylation methods reviewed by Galonic *et al.* (a), (b): Glycosylation of acetal-derived glycosyl donors. (c), (d): Glycosylation with glycal donors. Lg = leaving group; EI^+ = electrophilic promoter (blue); Nu-H = the acceptor (green); M = metal; R = various substituents; X = various leaving groups; Z = various functionalities.^{6e}

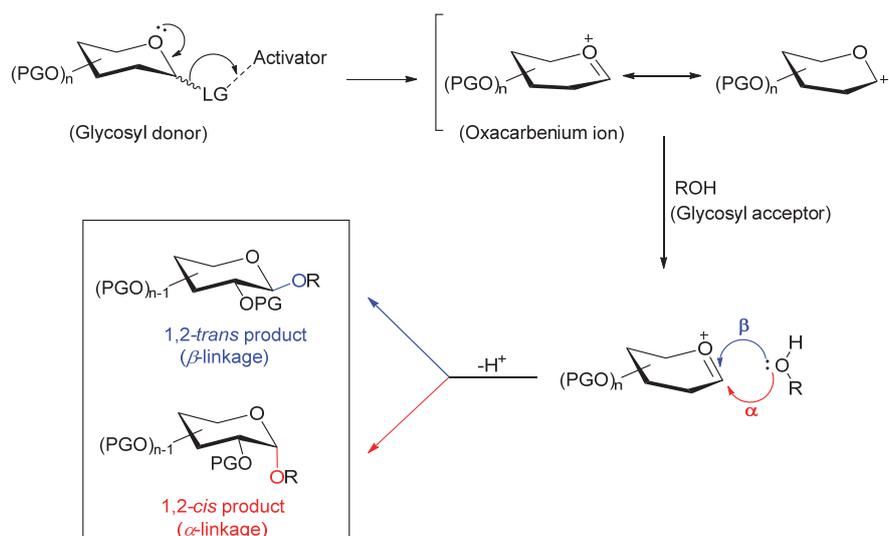
Other methods were also reported for the glycoside bond formation, such as one-pot and automated glycosylation,¹¹ solid-phase oligosaccharide synthesis,¹² intramolecular glycoside formation *etc.*^{6b,13}

¹¹ (a) Wang, Y.; Ye, X.-S.; Zhang, L.-H. *Org. Biomol. Chem.* **2007**, *5*, 2189-2200; (b) Koeller, K. M.; Wong, C.-H. *Chem. Rev.* **2000**, *100*, 4465-4493.

¹² (a) Seeberger, P. H.; Haase, W.-C. *Chem. Rev.* **2000**, *100*, 4349-4394; (b) Plante, O. J.; Palmacci, E. R.; Seeberger, P. H. *Science* **2001**, *291*, 1523-1527; (c) Larsen, K.; Thygesen, M. B.; Guillaumie, F.; Willats, W. G. T.; Jensen, K. J. *Carbohydr. Res.* **2006**, *341*, 1209-1234.

II.2 Stereochemistry

The formation of a glycoside linkage results in the creation of a new stereogenic center, which often leads to a mixture of two anomeric stereoisomers. There are two types of *O*-glycosides formed through a sp^2 -hybridized oxonium intermediate. The linkages formed are defined as 1,2-*cis* and 1,2-*trans* glycosides (Scheme 2). The stereoselective introduction of glycosidic linkages during glycosylation reactions is one of the most challenging aspects that have yet to be completely addressed by the synthetic chemist.^{6c,10b,14}



Scheme 2: Generic mechanism of the stereoselective formation of *O*-glycosidic bonds.^{10b}

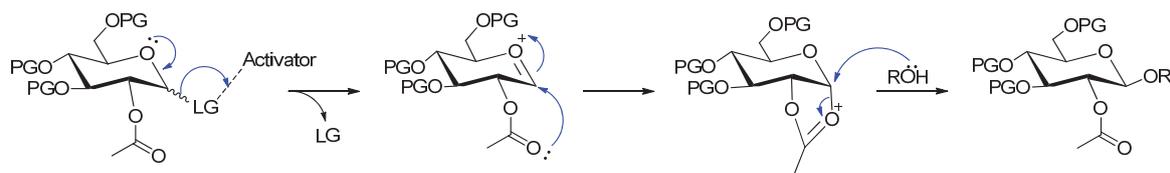
The nature of the protecting group at 2-position of a glycosyl donor is a major determinant of the anomeric selectivity.¹⁵ A protecting group at C-2, which can perform neighboring group participation during a glycosylation, leads reliably to the formation of 1,2-*trans* glycosides by a S_N2 mechanism. As shown in Scheme 3, a promoter activates an anomeric leaving group in its departure and the formation of an oxocarbenium ion. Subsequent neighboring group participation by the 2-*O*-acyl protecting group will give a more stable five-membered cyclic acetoxonium ion intermediate, which can only be formed as a 1,2-*cis* fused ring system. An alcohol can then attack the anomeric center of the acetoxonium ion intermediate from only one face providing a 1,2-*trans*-glycoside. Thus, in the case of glucopyranosyl-type donors, β-anomers will be obtained whereas mannopyranosides will give the

¹³ (a) Cumpstey, I. *Carbohydr. Res.* **2008**, *343*, 1553-1573; (b) Jung, K.-H.; Müller, M.; Schmidt, R. R. *Chem. Rev.* **2000**, *100*, 4423-4442.

¹⁴ (a) Nigudkar, S. S.; Demchenko, A. V. *Chem. Sci.* **2015**, *6*, 2687-2704; (b) Cox, D. J.; Singh, G. P.; Watson, A. J. A.; Fairbanks, A. J. *Eur. J. Org. Chem.* **2014**, 4624-4642.

¹⁵ (a) Wulff, G.; Röhle, G. *Angew. Chem. Int. Ed. Engl.* **1974**, *13*, 157-170; (b) Wulff, G.; Röhle, G. *Angew. Chem.* **1974**, *86*, 173-187.

corresponding α -anomers.¹⁶



Scheme 3: Generic mechanism of neighboring group participation in glycosylation reaction.

The presence of a sterically non-assisting, non-participating functionality at the 2-position is often used for the synthesis of 1,2-*cis*-glycosides. However, the stereocontrolled synthesis of 1,2-*cis*-glycosidic linkages is considerably more difficult.^{6b,17} In this type of glycosylation reaction, the anomeric selectivity will be affected by many factors such as the activation condition, the constitution of the glycosyl donor and acceptor (for example, type of saccharide, leaving group at the anomeric center, protection and substitution pattern), as well as the solvent, temperature, concentration, and even the sequence of addition of the reactants.¹⁸

III Structures and functions of glycopeptides and glycoproteins

III.1 Types of glycosidic linkages

The types of glycosidic bonds are classified according to the identity of the atom of the glycosyl acceptor which binds to the carbohydrate chain, i.e. *N*-glycoside, *O*-glycoside, *C*-glycoside or *S*-glycoside.

Two main forms of the naturally occurring protein glycosylations are *N*- and *O*-glycosides. *N*-glycoside, the most common type of glycosidic linkage in glycoproteins, is the attachment of a sugar molecule to the nitrogen atom of asparagine (Asn) side chains. In *O*-glycoside, the saccharide is linked to the hydroxyl group of serine (Ser), threonine (Thr), or tyrosine (Tyr). The majority of motifs are β -*N*-GlcNAc-Asn, α -*O*-GalNAc-Ser/Thr and β -*O*-GlcNAc-Ser/Thr. Other unusual linkage forms have also been reported, such as α -*N*-GlcNAc-Asn, Glc-*N*-Asn, GalNAc-*N*-Asn, α -*O*-Fuc-Ser/Thr and *O*-glycosides of hydroxylysine and hydroxyproline. Structures of some representative *N*- and *O*-glycosides are listed in Figure 4.^{1a,19}

¹⁶ Boltje, T. J.; Buskas, T.; Boons, G.-J. *Nature Chem.* **2009**, *1*, 611-622.

¹⁷ (a) Demchenko, A. V. *Curr. Org. Chem.* **2003**, *7*, 35-79; (b) Fairbanks, A. J. *Synlett* **2003**, 1945-1958.

¹⁸ Boons, G.-J. *Contemp. Org. Synth.* **1996**, *3*, 173-200.

¹⁹ Gamblin, D. P.; Scanlan, E. M.; Davis, B. G. *Chem. Rev.* **2009**, *109*, 131-163.

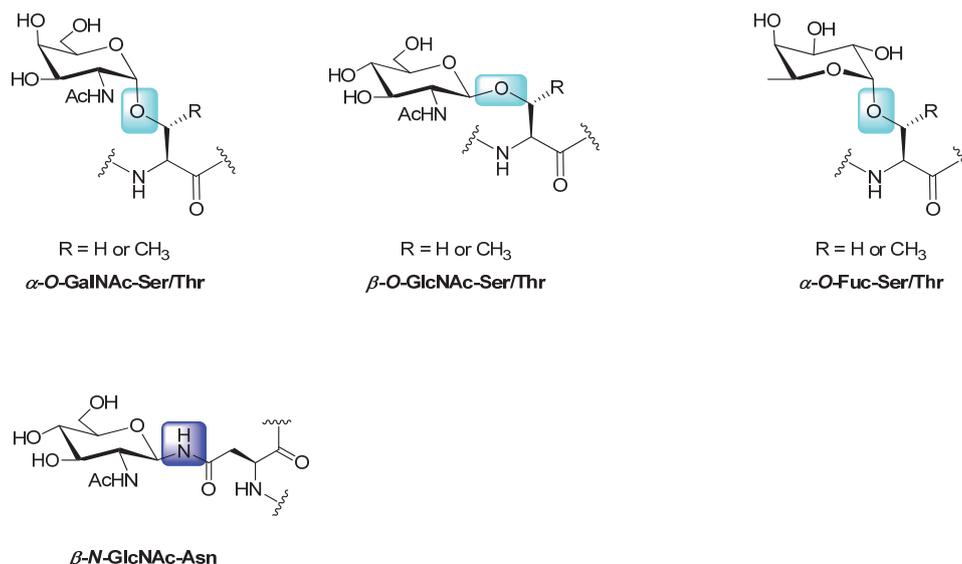


Figure 4: Structure of *O*-linked glycosyl serine and threonine, and *N*-linked glycosyl asparagine in glycopeptides.^{1a}

Natural *C*-glycopeptides are also existing. For example, a mannose is attached to a tryptophan (Trp) residue within an extracellular protein. The extremely rare *S*-glycopeptides, in which saccharides are covalently linked to the sulfur atoms of cysteine (Cys) residue, have been found in human and bacterial peptides.²⁰ Structures of *C*-linked and *S*-linked glycopeptides are presented in Figure 5.

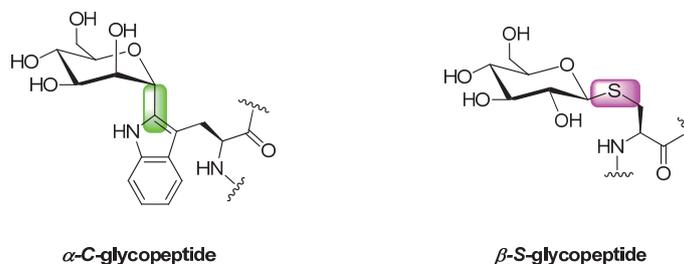


Figure 5: Structure of *C*- and *S*-linked glycopeptides.

III.2 Biological functions of glycopeptides and glycoproteins

Nowadays glycopeptides and glycoproteins are mainly recognized for their applications in various cellular activities such as cell-surface binding, including growth, differentiation, proliferation, adhesion or fertilization and immune responses. Furthermore, they also play a

²⁰ (a) Oman, T. J.; Boettcher, J. M.; Wang, H.; Okalibe, X. N.; van der Donk, W. A. *Nat. Chem. Biol.* **2011**, *7*, 78-80; (b) Stepper, J.; Shastri, S.; Loo, T. S.; Preston, J. C.; Novak, P.; Man, P.; Moore, C. H.; Havlicek, V.; Patchett, M. L.; Norris, G. E. *Febs. Lett.* **2011**, *585*, 645-650.

important role in pathogenic processes such as viral and bacterial infections, tumor growth and metastasis, autoimmune disorders and chronic inflammations.²¹

Structurally defined glycopeptides and glycoproteins are useful tools for functional biological studies, because they contain precise informations about the carbohydrate structures and glycosylation sites. Synthetic glycopeptides have also been used as vaccines to induce specific immune responses or for the inhibition of protein-binding events (for example, lectin recognition). In a review paper, Westerlind *et al.* described the development of glycopeptide anti-tumor vaccines. He also gave a summary on the synthesis of homogenous glycoproteins and glycopeptide dendrimers for the inhibition of microbe binding events.²¹ Besides, Ashford *et al.* have reviewed the recent advance in the synthesis of new glycopeptide antibiotics.²²

Thanks to the important biological functions of glycopeptides and glycoproteins, the development of glycopeptide and glycoprotein mimics attracted a lot of interest.²³

Aussedat *et al.* designed and chemically synthesized several homogeneous glycoprotein 120 (gp120) V1V2 domain containing two closely spaced high-mannose glycans at Asn¹⁶⁰ and Asn¹⁵⁶ as potential human immunodeficiency virus (HIV)-1-directed vaccines (Figure 6).^{2a,24} The synthetic glycopeptides were found to have high affinity to the broadly neutralizing antibodies (bNAbs) PG9 which can effectively neutralize diverse strains of HIV-1, while the oligosaccharides alone were not active. The characterization of the antigenic properties of these glycopeptides and the evaluation of their immunogenicity in animal models are in progress.

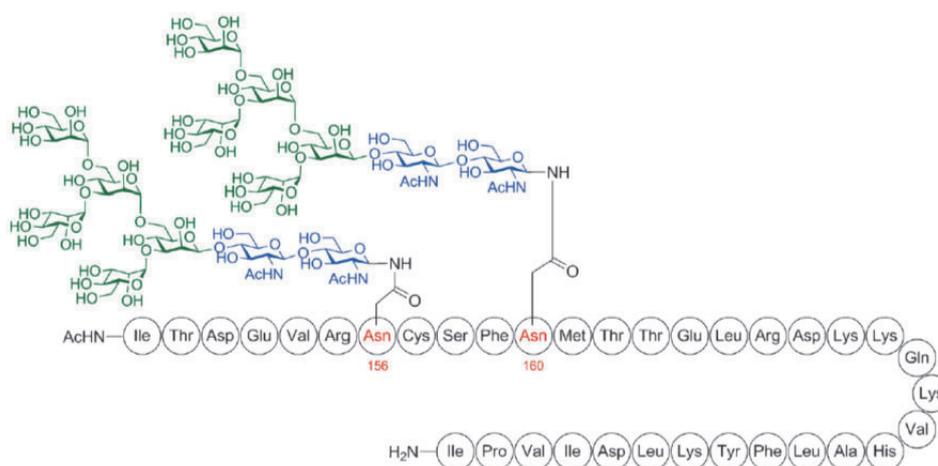


Figure 6: Synthetic glycopeptide antigens as PG9 epitope mimics for HIV-1-directed vaccines.^{2a,24}

²¹ Westerlind, U. *Beilstein J. Org. Chem.* **2012**, *8*, 804-818.

²² Ashford, P. A.; Bew, S. P. *Chem. Soc. Rev.* **2012**, *41*, 957-978.

²³ Pratt, M. R.; Bertozzi, C. R. *Chem. Soc. Rev.* **2005**, *34*, 58-68.

²⁴ (a) Aussedat, B.; Vohra, Y.; Park, P. K.; Fernández-Tejada, A.; Alam, S. M.; Dennison, S. M.; Jaeger, F. H.; Anasti, K.; Stewart, S.; Blinn, J. H.; Liao, H.-X.; Sodroski, J. G.; Haynes, B. F.; Danishefsky, S. J. *J. Am. Chem. Soc.* **2013**, *135*, 13113-13120; (b) Alam, S. M.; Dennison, S. M.; Aussedat, B.; Vohra, Y.; Park, P. K.; Fernández-Tejada, A.; Stewart, S.; Jaeger, F. H.; Anasti, K.; Blinn, J. H. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 18214-18219.

Glycopeptoids, in which one or several carbohydrate moieties were attached to *N*-substituted glycine or β -alanine oligomer backbone, have been developed as proteolytically stable glycopeptide mimics (Figure 7).²⁵ This type of glycoconjugates has greater conformational flexibility relative to glycopeptides and could be used as multivalent glycocluster constructs for targeting carbohydrate-lectin interactions or RNA biomolecules.²⁶ Different synthetic methods and applications of glycopeptoids have been reviewed by Szekely *et al.*²⁷

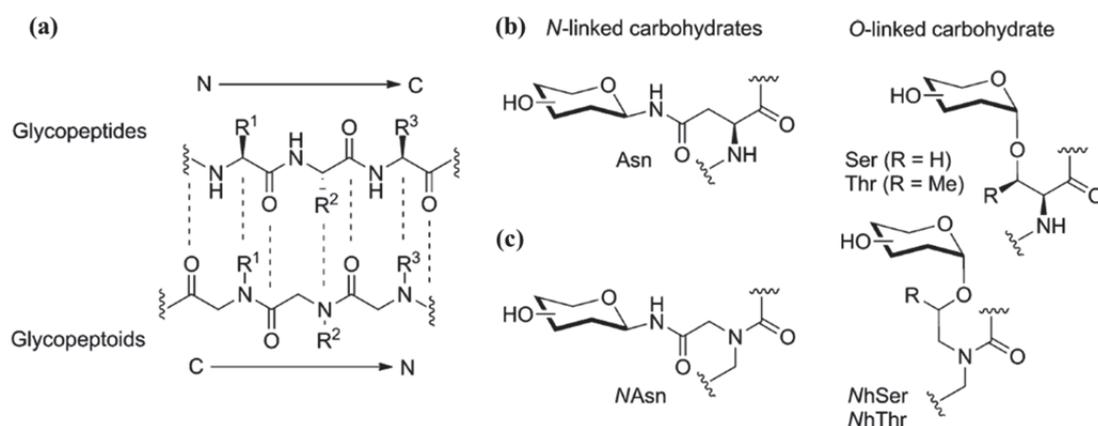


Figure 7: (a) Similarities between *N*-to-*C* glycopeptides and *C*-to-*N* glycopeptoids; (b) typical naturally occurring *N*- and *O*-linked glycosylated amino acids in peptides and (c) their peptoid analogues.²⁷

Galan *et al* have reviewed the recent advances in the preparation of linear and cyclic glycopeptides, and their applications in glycobiology, such as antitumoral vaccines, inhibitors against pathogens and ligands for carbohydrate-binding proteins.²⁸ Moreover, cyclopeptides could be developed as non-immunogenic carriers with improved resistance against proteolytic degradation. Recently, Pawar *et al.* reported the chemical synthesis of a series of linear and cyclic glycopeptides (Figure 8) which showed the inhibition activities against wild type HIV-1 C-SA protease.²⁹

²⁵ (a) Miller, S. M.; Simon, R. J.; Ng, S.; Zuckermann, R. N.; Kerr, J. M.; Moos, W. H. *Bioorg. Med. Chem. Lett* **1994**, *4*, 2657-2662; (b) Saha, U. K.; Roy, R. *Tetrahedron Lett.* **1995**, *36*, 3635-3638.

²⁶ Seo, J.; Michaelian, N.; Owens, S. C.; Dashner, S. T.; Wong, A. J.; Barron, A. E.; Carrasco, M. R. *Org. Lett.* **2009**, *11*, 5210-5213.

²⁷ Szekely, T.; Roy, O.; Faure, S.; Taillefumier, C. *Eur. J. Org. Chem.* **2014**, 5641-5657.

²⁸ Galan, M. C.; Dumy, P.; Renaudet, O. *Chem. Soc. Rev.* **2013**, *42*, 4599-4612.

²⁹ Pawar, S. A.; Jabgunde, A. M.; Maguire, G. E.; Kruger, H. G.; Sayed, Y.; Soliman, M. E.; Dhavale, D. D.; Govender, T. *Eur. J. Med. Chem.* **2013**, *60*, 144-154.

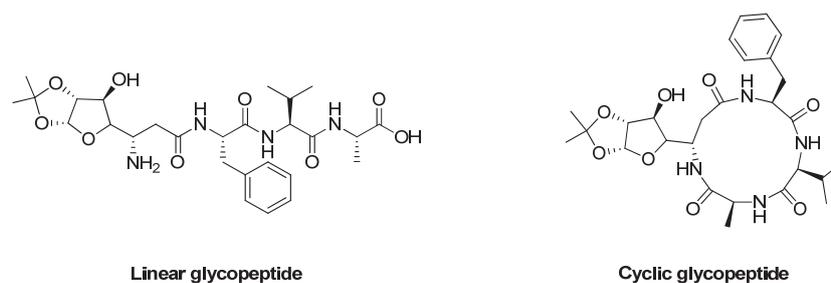


Figure 8: Synthetic linear and cyclic glycopeptides reported by Pawar *et al.*²⁹

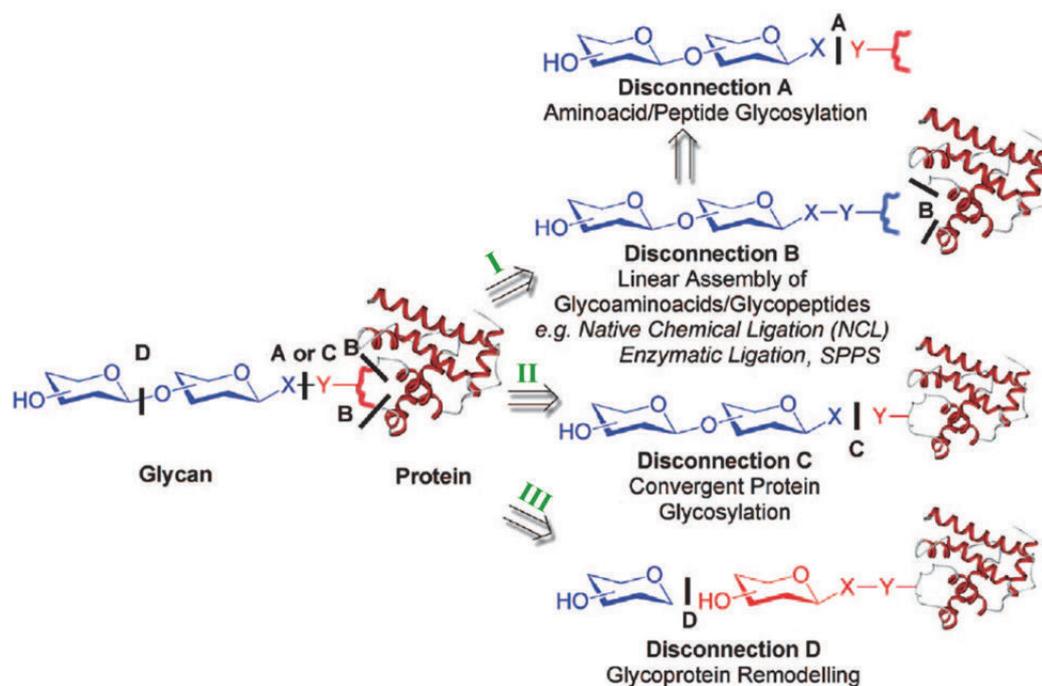
III.3 Chemical synthesis of glycopeptides and glycoproteins

Structurally defined glycopeptides and glycoproteins have been proved to play an important role in studying their diverse biological phenomena. Various methods, such as chemical or chemoenzymatic synthesis, native chemical ligation (NCL), expressed protein ligation (EPL), as well as several other assembly strategies and enzyme-based routes, have been developed for the preparation of functional glycopeptides and glycoproteins. A number of reviews on the synthesis of glycopeptides and glycoproteins can be found in the literature.³⁰

III.3.a Retrosynthetic analysis of glycoproteins

Glycoproteins could be considered as polyamino acids-poly/oligosaccharide conjugates. A generalized disconnective analysis of glycoprotein synthesis reported by Gamblin *et al.* is shown in Scheme 4, in which glycoproteins may be prepared through three strategies.^{30c,30e} Method **I** displayed that the glycoproteins could be prepared by linear assembly of preformed glycoamino acids/glycopeptides via native chemical ligation (NCL), enzymatic ligation, solid-phase peptide synthesis (SPPS), etc (disconnection **B**). The glycoamino acids/glycopeptides could be obtained by amino acids/peptides glycosylation (disconnection **A**). Convergent protein glycosylation with prepared glycan array represents the disconnection **C** (**II**, Scheme 4). Another approach is glycoprotein remodeling, in which the glycoprotein with monosaccharide moiety is synthesized firstly, and then elongation with endoglycosidases or recombinant glycosyltransferases to rebuild the new glycoprotein (**III**, Scheme 4).

³⁰ (a) Crucho, C. I.; Correia-da-Silva, P.; Petrova, K. T.; Barros, M. T. *Carbohydr. Res.* **2015**, *402*, 124-132; (b) Wu, C. Y.; Wong, C. H. *Chem. Commun.* **2011**, *47*, 6201-6207; (c) Gamblin, D. P.; Scanlan, E. M.; Davis, B. G. *Chem. Rev.* **2009**, *109*, 131-163; (d) Liu, L.; Bennett, C. S.; Wong, C. H. *Chem. Commun.* **2006**, 21-33; (e) Davis, B. G. *Chem. Rev.* **2002**, *102*, 579-602; (f) Sears, P.; Wong, C.-H. *Science* **2001**, *291*, 2344-2350; (g) Seitz, O. *ChemBioChem.* **2000**, *1*, 214-246; (h) St Hilaire, P. M.; Meldal, M. *Angew. Chem. Int. Ed.* **2000**, *39*, 1162-1179.



Scheme 4: Glycoprotein retrosynthetic analysis reported by Gamblin *et al.*^{30c}

III.3.b Formation of the glycosidic linkages

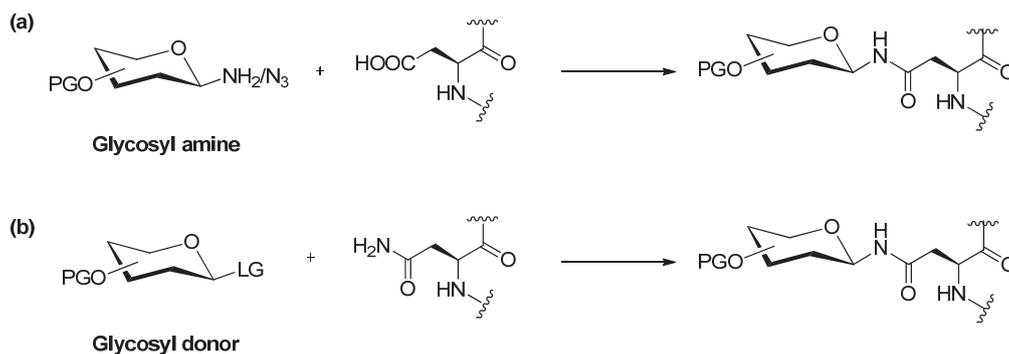
The key step of all glycopeptide synthetic strategies is the formation of glycan-amino acid bond (glycosidic linkage). Several reviews have already covered the chemical synthesis of glycosidic linkages.^{30c,30e,31} Herein, we list some common methods for the formation of *N*- and *O*-glycosidic linkages.

III.3.b (1) Formation of *N*-glycopeptides

The most common approach for the formation of *N*-glycosides is via reaction of a protected glycosyl amine (by using a coupling reagent) or glycosyl azide (by Staudinger reaction in the presence of triphenylphosphine (Ph_3P), by Staudinger ligation with an Asp-derived phosphinothioester, or by phototransamidation strategy³²) with a suitably protected Asp derivative (Scheme 5a) to generate an amide bond. Another strategy is the direct glycosylation of the amide side chain of Asn using a glycosyl donor (Scheme 5b).³¹

³¹ (a) Taylor, C. M. *Tetrahedron* **1998**, *54*, 11317-11362; (b) Arsequell, G.; Valencia, G. *Tetrahedron: Asymmetry* **1999**, *10*, 3045-3094.

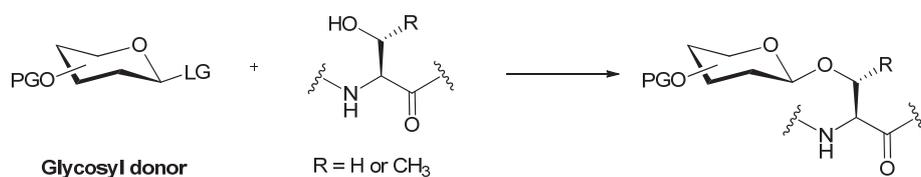
³² Kaneshiro, C. M.; Michael, K. *Angew. Chem. Int. Ed.* **2006**, *45*, 1077-1081.



Scheme 5: Methods for preparation of *N*-linked glycopeptides. (a) Amide bond formation; (b) Direct glycosylation of an Asn residue.³¹

III.3.b (2) Formation of *O*-glycopeptides

Direct glycosylation of the hydroxyl group of a suitably protected Ser or Thr using a standard glycosyl donor like trichloroacetimidate, thioglycoside or *n*-pentenyl glycosides has been reported as a commonly used route for the formation of *O*-glycosidic bond in glycopeptide (Scheme 6).^{30c,31a}



Scheme 6: Method for the preparation of *O*-linked glycopeptides.^{30c,31a}

III Structures and functions of glycolipids

III.1 Classification of glycolipids

As an important part of the glycoconjugate family, glycolipids are comprised of lipids covalently attached to one or several monosaccharides. The hydrophilic polar sugar head group and the hydrophobic lipid moiety give their amphiphilic character, which make the glycolipids able to self-insert in cell membranes.³³ Nowadays, glycolipids are well known to play an important role in a variety of important biological phenomena such as cell-cell interactions, viral and bacterial infections, activation and modulation of immune system, signal transduction, cell proliferation, etc.³⁴

Based on their lipid moieties, glycolipids can be mainly classified as glyco glycerolipids (GGLs), glycosphingolipids (GSLs) and other lipids.³⁵ The classification and some representative structures of glycolipids are shown in Figure 9.

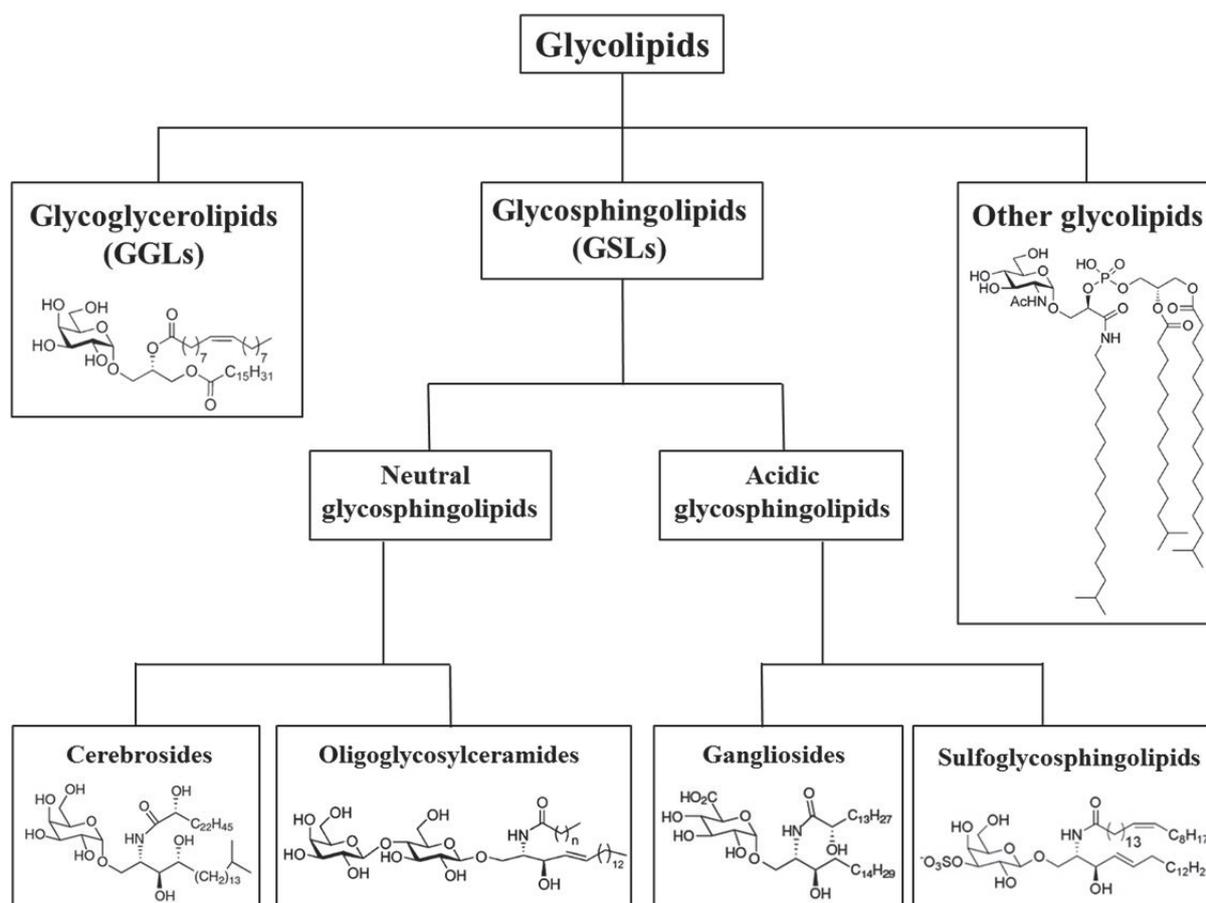


Figure 9: Classification and representative structures of glycolipids.

³³ Hashim, R.; Sugimura, A.; Minamikawa, H.; Heidelberg, T. *Liq. Cryst.* **2012**, *39*, 1-17.

³⁴ Brandenburg, K.; Holst, O. **2015**, Glycolipids: Distribution and Biological Function. eLS. 1–10.

³⁵ IUPAC-IUB, Joint Commission on Biochemical Nomenclature (JCBN). *Carbohydr. Res.* **1998**, *312*, 167-175.

GGLs are glycolipids containing mono- or oligosaccharide moieties linked to the C-3 hydroxyl group of glycerol which may be attached with one or two fatty acid chains.

GSLs are comprised of a ceramide backbone covalently linked to one or several saccharide residues. GSLs can be subdivided as neutral GSLs, and acidic GSLs. In neutral GSLs family, cerebrosides are monoglycosylceramides in which the sugar residue is attached by *O*-ether linkage to the primary alcohol, while oligoglycosylceramides contain more than one sugar moiety. The acidic GSLs were further divided into sulfoglycosphingolipids and gangliosides containing respectively sulfonic or carboxylic acid functions.³⁶

III.2 Glycoglycerolipids (GGLs)

III.2.a Structures and functions of natural glycoglycerolipids (GGLs)

GGLs mainly exist in the thylakoid membranes in chloroplasts of higher plants, eukaryotic algae and cyanobacteria. Lesser amounts of GGLs are also found in animals. They demonstrated various biological activities, such as anti-tumor, anti-viral, and anti-inflammatory activities.

The common structures of GGLs extracted from natural products are mainly comprised of three types: monogalactosyl diacylglycerol (MGDG), digalactosyl diacylglycerol (DGDG), and sulfoquinovosyl diacylglycerol (SQDG) (Figure 10). For example, the chemical structures of three major glycolipids extracted from spinach were determined as MGDG, DGDG and SQDG by Mizushina's group.³⁷ It has been reported that both the MGDG and its monoacyl derivative (MGMG) obtained by hydrolyzing MGDG with a pancreatic lipase, can inhibit the DNA polymerase activities and the growth of NUGC-3 human gastric cancer cells, with a stronger inhibition by MGMG (Figure 10).³⁸ Further studies showed that the SQDG from spinach can inhibit mammalian DNA polymerase activity, human cultured cell growth and *in vivo* solid tumor proliferation. Both of the sulfo group and fatty acid moiety potently affected the DNA polymerase inhibition of the SQDG.³⁹

³⁶ Malhotra, R. *Biochem. Anal. Biochem.* **2012**, *1*, 108.

³⁷ Maeda, N.; Matsubara, K.; Yoshida, H.; Mizushina, Y. *Mini-Rev. Med. Chem.* **2011**, *11*, 32-38.

³⁸ Murakami, C.; Kumagai, T.; Hada, T.; Kanekazu, U.; Nakazawa, S.; Kamisuki, S.; Maeda, N.; Xu, X.; Yoshida, H.; Sugawara, F.; Sakaguchi, K.; Mizushina, Y. *Biochem. Pharmacol.* **2003**, *65*, 259-267.

³⁹ (a) Maeda, N.; Hada, T.; Yoshida, H.; Mizushina, Y. *Curr. Med. Chem.* **2007**, *14*, 955-967; (b) Maeda, N.; Kokai, Y.; Ohtani, S.; Sahara, H.; Hada, T.; Ishimaru, C.; Kuriyama, I.; Yonezawa, Y.; Iijima, H.; Yoshida, H. *Nutr. Cancer* **2007**, *57*, 216-223.

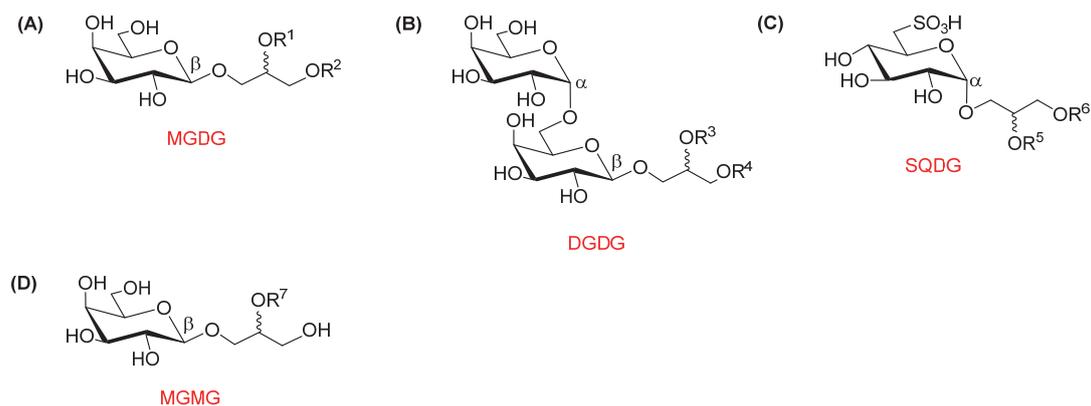


Figure 10: Chemical structure of MGDG, DGDG, SQDG and MGMG. R^1 to R^7 = acyl chains.³⁷

The natural aminoglycoglycerolipids have also shown good biological activities. From the rhizomes of an herbaceous plant *serratula strangulate*, Dai *et al* isolated the aminoglycoglycerolipid **a** (Figure 11) and other two GGLs. These compounds were reported to exhibit significant antibacterial and antitumor activities.⁴⁰ Another two aminoglycoglycerolipids (**b** and **c**, Figure 11) were isolated from marine alga by Zhou *et al.* and exhibited good inhibitory activity against myelin transcription factor 1 (Myt1)-kinase with an IC_{50} value of 0.12 $\mu\text{g/mL}$.⁴¹

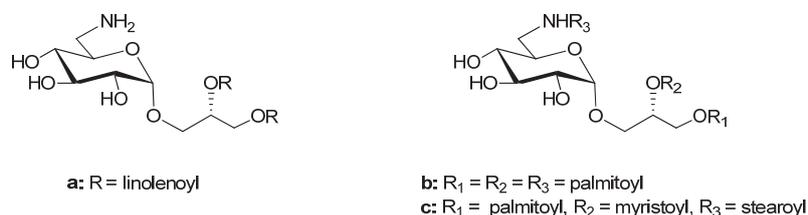


Figure 11: Chemical structure of the natural 6-amino-6-deoxy- α -glycoglycerolipids.^{40,41}

Another class of 6-*O*-ether-linked GGLs, nigricanosides **A**, **B** and their respective dimethyl ester derivatives **A-1**, **B-1** (Figure 12) were extracted from the green alga *Avrain Villeda nigricans* by Williams *et al*, which showed potent antimitotic activity and the ability to promote tubulin polymerization.⁴²

⁴⁰ Dai, J. Q.; Zhu, Q. X.; Zhao, C. Y.; Yang, L.; Li, Y. *Phytochemistry* **2001**, 58, 1305-1309.

⁴¹ Zhou, B.-N.; Tang, S.; Johnson, R. K.; Mattern, M. P.; Lazo, J. S.; Sharlow, E. R.; Harich, K.; Kingston, D. G. I. *Tetrahedron* **2005**, 61, 883-887.

⁴² Williams, D. E.; Sturgeon, C. M.; Roberge, M.; Andersen, R. J. *J. Am. Chem. Soc.* **2007**, 129, 5822-5823.

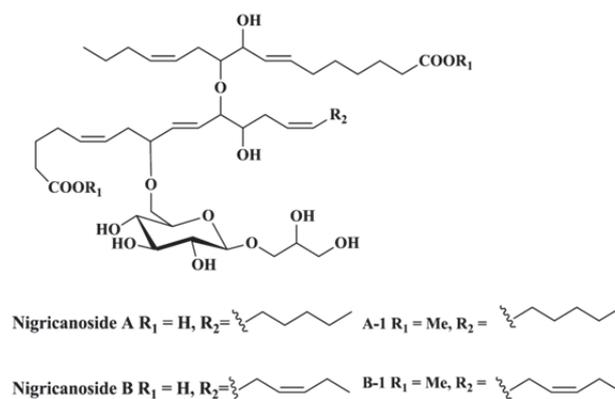


Figure 12: Chemical structure of ether-linked GGLs reported by Williams *et al.*⁴²

There are a number of naturally existing GGLs which have shown interesting biological activities. In a review paper, Holzl *et al.* have described the structures and functions of GGLs in plants and bacteria.⁴³ Very recently, Li and co-workers reviewed the structures and activities of GGLs from marine organisms.⁴⁴

III.2.b Modifications of glycolycerolipids (GGLs)

Various GGLs have been extracted from the natural products and shown potent biological activities. Studies focused on the structure-activity relationships suggested that the biological activities of the glycolipids were influenced by their chemical structures.⁴⁴ Consequently, in order to obtain GGLs with the similar or better biological activities, different strategies have been developed for the modification of GGLs and synthesis of their analogues.

Modifications are mainly based on four types:

- (a) Substitution and variation of the sugar moiety;
- (b) Nature of the glycoside bond and anomeric configuration;
- (c) Position of the glycerol linkage to the sugar;
- (d) Variation on the length and location of the lipid chains.

A number of synthetic GGL mimics have been designed and synthesized. For example, with the purpose to obtain better Myt1-kinase inhibitors, Li *et al.* synthesized series of 6-amino-6-deoxy-glycolycerolipid analogues with different glycosyl (D-gluco-,⁴⁵ D-galacto-,⁴⁶ D-manno-⁴⁷) moieties and lipid chains (Figure 13). Evaluation of the inhibition activity against Myt1-kinase of compounds **A1-A8**, and **B1-B7** showed that the activities of

⁴³ Holzl, G.; Dormann, P. *Prog. Lipid. Res.* **2007**, *46*, 225-243.

⁴⁴ Zhang, J.; Li, C.; Yu, G.; Guan, H. *Marine Drugs* **2014**, *12*, 3634-3659.

⁴⁵ Sun, Y.; Zhang, J.; Li, C.; Guan, H.; Yu, G. *Carbohydr. Res.* **2012**, *355*, 6-12.

⁴⁶ Li, C.; Sun, Y.; Zhang, J.; Zhao, Z.; Yu, G.; Guan, H. *Carbohydr. Res.* **2013**, *376*, 15-23.

⁴⁷ Zhang, J.; Sun, Y.; Wang, W.; Zhang, X.; Li, C.; Guan, H. *Carbohydr. Res.* **2013**, *381*, 74-82.

the galactose series were generally better than the corresponding glucose derivatives; and with the same glycosyl moiety, the increasing length of the lipid chains led to the enhancement of the activities.⁴⁶ Importantly, among these compounds, the aminogalactoglycerolipids **B7** with a branched acyl chain exhibited the best inhibitory activity, and in their following research on the mannose analogues, the **C7** with a branched acyl chain showed effective anti-influenza A virus (IAV) activity.⁴⁷

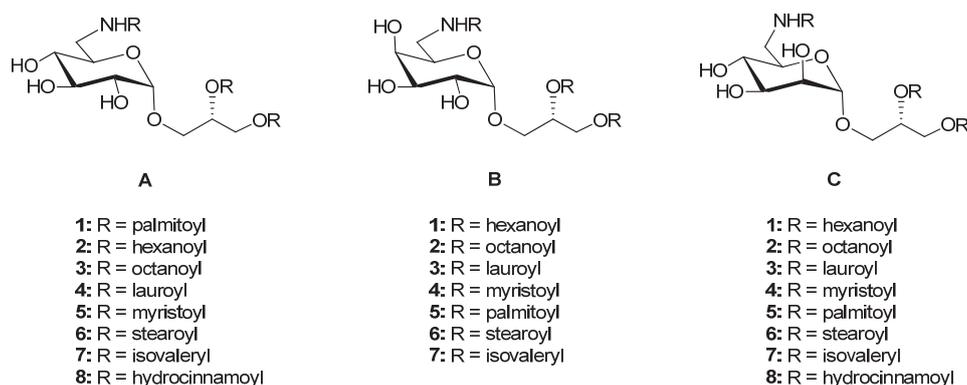


Figure 13: Structures of the natural aminoglycolipids **A1** and glucose analogues **A2-A8**,⁴⁵ galactose analogues **B1-B7**,⁴⁶ mannose analogues **C1-C8**.⁴⁷

Vetro *et al.* reported the synthesis of new glucuronosyldiacylglycerol (GlcADG) analogues as 3-phosphorylated phosphatidylinositol (PI3P) analogues (Figure 14) which could target the protein kinase B (also known as Akt).⁴⁸ In an *in vitro* kinase assay, compounds **Ia** and **Iib** showed the Akt inhibitory effect, and antiproliferative activity in human ovarian carcinoma cells.

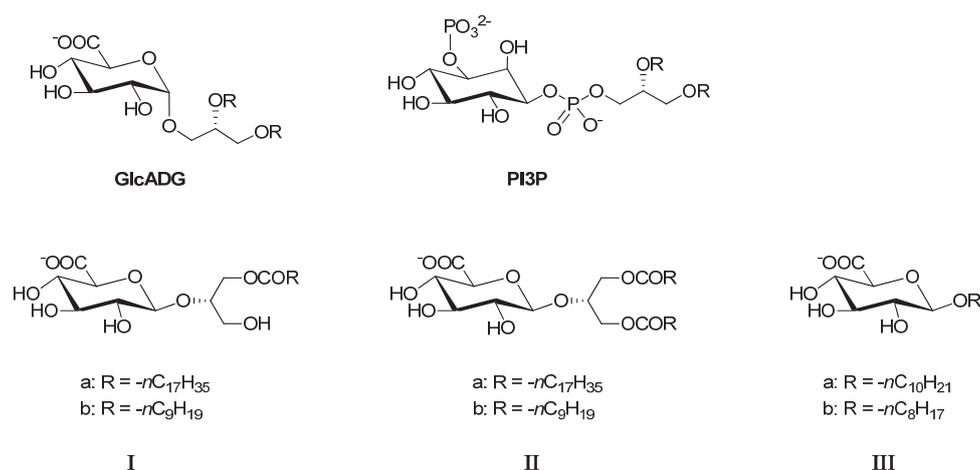


Figure 14: Structure of natural GlcADG, PI3P and their analogues.⁴⁸

⁴⁸ Vetro, M.; Costa, B.; Donvito, G.; Arrighetti, N.; Cipolla, L.; Perego, P.; Compostella, F.; Ronchetti, F.; Colombo, D. *Org. Biomol. Chem.* **2015**, *13*, 1091-1099.

III.3 Glycosphingolipids (GSLs)

III.3.a Structures and functions of natural glycosphingolipids (GSLs)

As a most abundant and diverse class of glycolipids in animals, GSLs are found in the plasma membrane of cells and play key roles in cell-cell communication and the modulation of membrane-protein function.⁴⁹ They also could be found in fungi, plants, and invertebrates. The most widely known structures of GSLs are α - or β -galactosyl ceramide (α -/ β -GalCer), in which a galactosyl moiety is linked to an *N*-acylated sphingosine backbone (ceramide).

A series of glycolipids termed agelasphins (AGLs) was isolated from the marine sponge *Agelas mauritianus* by Natori *et al.*, which was known as α -GalCer.⁵⁰ The diversity of the compositions and lengths on the lipid chains lead to different agelasphins, and some structures are present in Figure 15. The **AGLs-9b** showed potent *in vivo* anti-tumor activity against the murine B16 mouse melanoma.



Figure 15: Chemical structure of the natural AGLs from marine sponge.⁵⁰

In 1991, Harouse *et al.* have reported that the antibodies against the β -GalCer **1** (Figure 16) can inhibit the HIV-1 infection of CD4-negative cell lines in nerve system.⁵¹

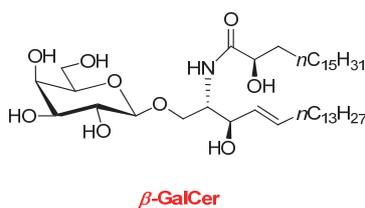


Figure 16: Chemical structure of the natural β -GalCer in nerve system.⁵¹

⁴⁹ (a) D'Angelo, G.; Capasso, S.; Sticco, L.; Russo, D. *FEBS J.* **2013**, *280*, 6338-6353; (b) Wu, D.; Fujio, M.; Wong, C.-H. *Bioorg. Med. Chem.* **2008**, *16*, 1073-1083.

⁵⁰ Natori, T.; Koezuka, Y.; Higa, T. *Tetrahedron Lett.* **1993**, *34*, 5591-5592.

⁵¹ Harouse, J. M.; Bhat, S.; Spitalnik, S. L.; Laughlin, M.; Stefano, K.; Silberberg, D. H.; Gonzalez-Scarano, F. *Science* **1991**, *253*, 320-323.

III.3.b Modifications of glycosphingolipids (GSLs)

As GGLs, modifications of GSLs have attracted intensive research interest, in particular, since the discovery of KRN 7000 (Figure 17), a synthetic analogue of **ASLs-9b**. The KRN 7000 was found to possess the similar anti-tumor activity as ASLs-9b.⁵² More importantly, it was reported to have potential activities against cancer, malaria, type 1 diabetes, and multiple sclerosis. Its ability to bind to CD1d and to activate invariant natural Killer T (*i*NKT) cells was also observed.

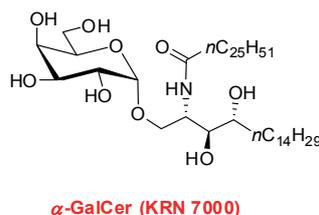


Figure 17: Chemical structure of the synthetic α -GalCer analogue KRN 7000.⁵²

Various efforts were made on the modification of KRN 7000, and have been summarized by Haudrechy and co-workers in 2011.⁵³ The four types of modifications described in this review paper are listed in Figure 18.

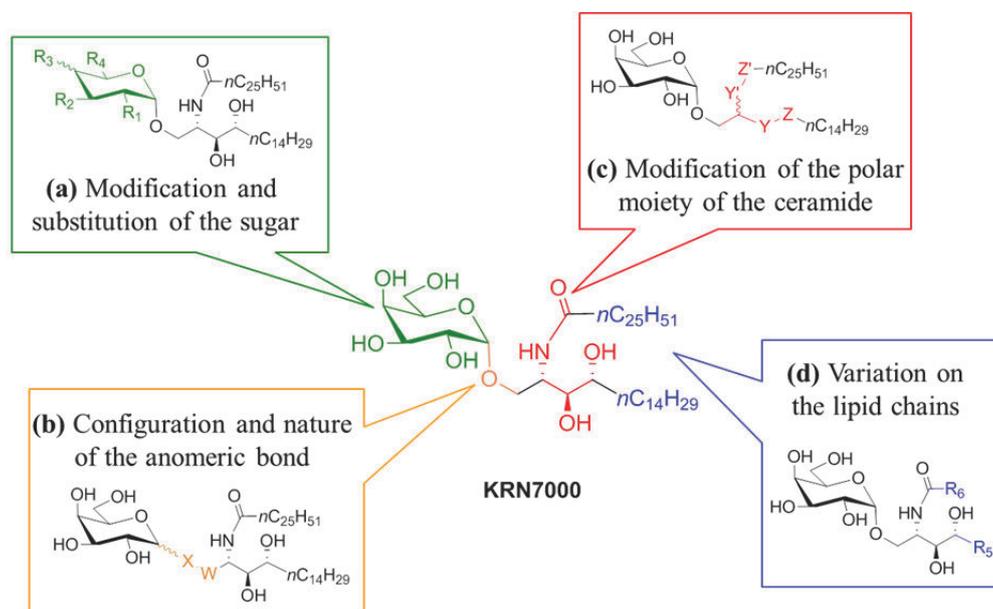


Figure 18: Four types of modifications to access KRN 7000 analogues reviewed by Haudrechy and co-workers.⁵³ (a) $R_1, R_2, R_3 = \text{OH}$ or substituent group, $R_4 = \text{CH}_2\text{OH}$ or substituent group; (b) $X = \text{CH}_2, \text{O}, \text{NH}, \text{S}$, etc; $W = \text{CH}_2, =\text{CH}-$, etc; (c) $Y, Z = \text{CH}_2, \text{CH-OH}, \text{CH-NH}_2$, etc; (d) $R_5, R_6 = \text{lipid chains}$.

⁵² Morita, M.; Motoki, K.; Akimoto, K.; Natori, T.; Sakai, T.; Sawa, E.; Yamaji, K.; Koezuka, Y.; Kobayashi, E.; Fukushima, H. *J. Med. Chem.* **1995**, *38*, 2176-2187.

⁵³ Banchet-Cadeddu, A.; Henon, E.; Dauchez, M.; Renault, J. H.; Monneaux, F.; Haudrechy, A. *Org. Biomol. Chem.* **2011**, *9*, 3080-3104.

Very recently, by replacing the ring oxygen of the galactopyranose residue with a sulfur atom, a new KRN 7000 structural analogue (**a**, Figure 19) was designed by Bi and co-workers.⁵⁴ Another two 5-thio-analogues **b** and **c**, in which the modifications were taken place on both the sugar ring and the lipid chain have also been prepared. *In vitro* tests of the analogue **b** showed that it can selectively compress the production of interleukin 4 (IL-4) in the presence of relatively high interferon- γ (IFN- γ) production of NKT cells, which may be useful for developing a new potent immunostimulating agent for specific inducing *i*NKT cells to produce T helper 1 (Th1) cytokines.

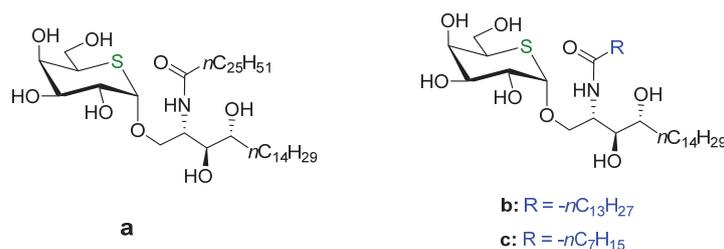


Figure 19: Structure of the 5-S-substituted KRN 7000 analogues.⁵⁴

Guillaume *et al.* designed and synthesized an aromatic α -GalCer, the 6'-naphthylurea derivative of KRN 7000 (**A**, Figure 20), which showed a potent Th1 response in both mice and men.⁵⁵ Based on this previous research, another two series of α -GalCers analogues **B** and **C** with modification on the 6'-position of the galactose were prepared in order to evaluate their biological activities as *i*NKT cell agonists.⁵⁶ Structure of the 6'-naphthylurea derivative **A**, 6'-carbamate derivative **B** and 5'-uronamide derivative **C** is shown in Figure 20.

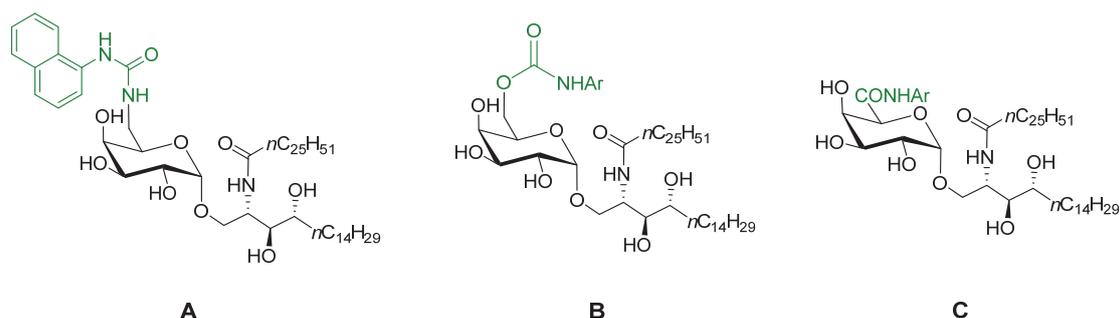


Figure 20: Structure of the 6'-naphthylurea (**A**), 6'-carbamate (**B**) and 5'-uronamide derivatives (**C**) of α -GalCer.⁵⁶

⁵⁴ Bi, J.; Wang, J.; Zhou, K.; Wang, Y.; Fang, M.; Du, Y. *ACS Med. Chem. Lett.* **2015**, *6*, 476-480.

⁵⁵ Aspeslagh, S.; Li, Y.; Yu, E. D.; Pauwels, N.; Trappeniers, M.; Girardi, E.; Decrui, T.; Van Beneden, K.; Venken, K.; Drennan, M.; Leybaert, L.; Wang, J.; Franck, R. W.; VanCalenbergh, S.; Zajonc, D. M.; Elewaut, D. *EMBO J.* **2011**, *30*, 2294-2305.

⁵⁶ Guillaume, J.; Pauwels, N.; Aspeslagh, S.; Zajonc, D. M.; Elewaut, D.; Van Calenbergh, S. *Bioorg. Med. Chem.* **2015**, *23*, 3175-3182.

Various analogues of β -GalCer were also synthesized and showed inhibition activity to HIV infection. For example, two analogues (*C*-glycoside **1** and aza-*C*-glycoside **2**, Figure 21) were synthesized by Thota *et al.* It has been shown that the aza-*C*-glycoside exhibited a significantly higher affinity to gp120 than GalCer, whereas the *C*-glycoside was as active as GalCer.⁵⁷

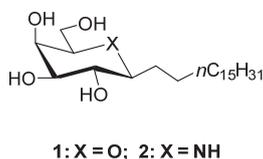


Figure 21: Structure of the *C*-glycosidic analogues of β -GalCer.⁵⁷

Thota *et al.* synthesized another *C*-glycosidic analogue of β -GalCer as a potential HIV inhibitor, in which the structural modifications were also taken place on the polar moiety of the ceramide and the length of lipid chains (Figure 22).⁵⁸

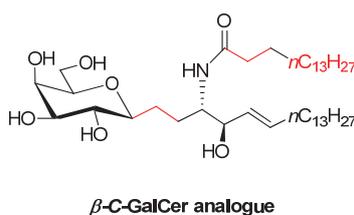


Figure 22: Structure of β -*C*-GalCer analogue synthesized by Thota *et al.*⁵⁸

Thanks to their potent biological activities, such as anti-tumor and anti-HIV, the design and synthesis of glycolipid mimics has become a useful strategy in pharmaceutical and medical research.⁵⁹

IV Conclusion

In this chapter, we have given a short literature overview on the structures and functions of glycopeptides/glycoproteins and glycolipids. Many efforts have been made for the synthesis of functional glycoconjugates. Both of the synthetic glycopeptides and glycolipids have shown interesting biological activities.

Our group has been interested in synthesizing *N*-oxyamide-linked compounds in the past

⁵⁷ Garg, H.; Francella, N.; Tony, K. A.; Augustine, L. A.; Barchi, J. J.; Fantini, J.; Puri, A.; Mootoo, D. R.; Blumenthal, R. *Antivir. Res.* **2008**, *80*, 54-61.

⁵⁸ Thota, V. N.; Brahmaiah, M.; Kulkarni, S. S. *J. Org. Chem.* **2013**, *78*, 12082-12089.

⁵⁹ (a) Daniotti, J. L.; Vilcaes, A. A.; Demichelis, V. T.; Ruggiero, F. M.; Rodriguez-Walker, M. *Front. Oncol.* **2013**, *3*, 306; (b) Wennekes, T.; van den Berg, R. J.; Boot, R. G.; van der Marel, G. A.; Overkleeft, H. S.; Aerts, J. M. *Angew. Chem. Int. Ed.* **2009**, *48*, 8848-8869; (c) McReynolds, K. D.; Gervay-Hague, J. *Chem. Rev.* **2007**, *107*, 1533-1552.

few years. In this thesis, we were interested in preparing novel glycolipids and glycopeptides analogues containing the *N*-oxyamide linkage. In the next chapter, we will present the properties of the N-O linkage and the functions of the N-O linked compounds.

Chapter 2

N-O Linkage in Carbohydrates

I Introduction

As described in Chapter 1, glycopeptides and glycolipids play very important biological functions in living organisms. Various methodologies have been developed for the construction of glycoconjugate mimetics which have shown interesting properties. Design and synthesis of new carbohydrate derivatives with various functions are attracting more and more research efforts.

One of the recent research subjects in our group is the synthesis of *N*-oxyamide-containing biomolecules mimetics. Studies on aminoxy acids by Yang's group showed that aminoxy acid derived peptides can easily organize into turns and helices structures through intramolecular hydrogen bond formation.⁶⁰ This unique property makes *N*-oxyamide linkage attractive for the modification of biomolecules. Furthermore, the *N*-oxyamide linkage is resistant to chemical and enzymatic hydrolysis,⁶¹ and *N*-oxy peptides could be readily prepared using classical peptide coupling methods.

There are a large number of compounds containing an oxygen-nitrogen bond, such as hydroxylamine, hydroxamic acid, oxime, nitron, and *N*-oxyamide compounds, etc. (Figure 23). In this chapter, we will present the aminoxy acids and N-O linkage in carbohydrate derivatives.

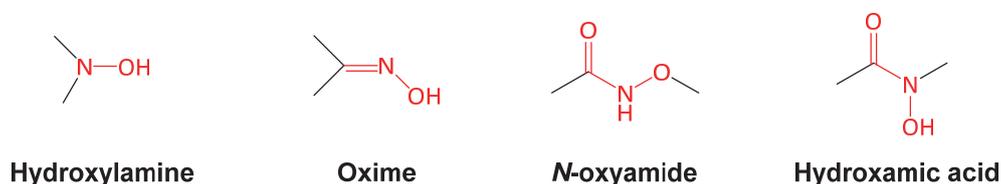


Figure 23: Examples of compounds containing N-O linkage.

II Aminoxy acids

II.1 Structure of aminoxy acids

Aminoxy acids are a class of unnatural amino acids where an aminoxy function has been introduced in place of the amine group (Figure 24).

⁶⁰ Li, X.; Wu, Y.-D.; Yang, D. *Accounts. Chem. Res.* **2008**, *41*, 1428-1438.

⁶¹ Chen, F.; Ma, B.; Yang, Z.-C.; Lin, G.; Yang, D. *Amino Acids* **2012**, *43*, 499-503.

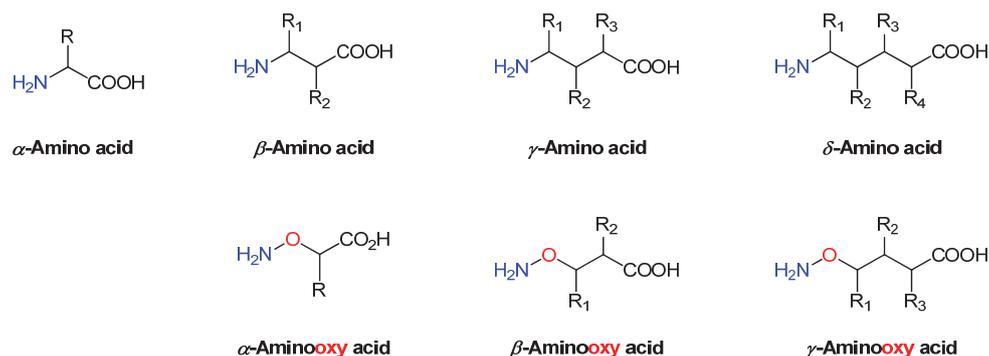


Figure 24: Chemical structure of amino acids and aminoxy acids.

L-Canavanine, the first amino acid with a substituted hydroxylamine moiety, was isolated from the non-protein fraction of Jack bean *Canvalia ensiformis* by Kitagawa and co-workers in 1929 (**1**, Figure 25).⁶² L-Canaline, which contains a free aminoxy group, was obtained by hydrolysis of the guanidinoxy group in L-canavanine by a liver ferment (**2**, Figure 25).⁶³ The L-canaline was reported to possess the inhibition activities against ornithine-dependent enzyme, and function as a lysine antagonist. It has also shown significant antineoplastic activity *in vitro* against human pancreatic cancer cells.⁶⁴

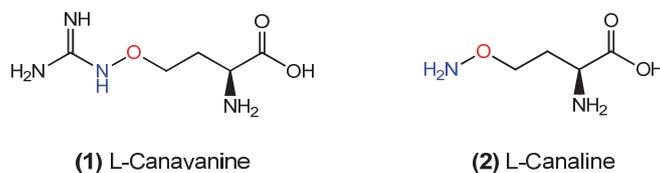


Figure 25: Chemical structure of L-canavanine and L-canaline.^{62,63}

Another natural aminoxy acid derivative isolated by Kuehl Jr *et al* was D-cycloserine, a cyclic product of 2-amino-3-aminoxypropionic acid, which showed a broad spectrum of antibacterial activity (Figure 26).⁶⁵

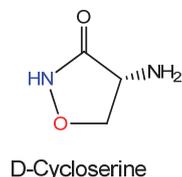


Figure 26: Chemical structure of D-cycloserine.⁶⁵

⁶² Kitagawa, M.; Tomiyama, T. *J. Biochem.* **1929**, *11*, 265-271.

⁶³ (a) Kitagawa, M.; Yamada, H. *J. Biochem.* **1932**, *16*, 339-350; (b) Kitagawa, M.; Monobe, S.-I. *J. Biochem.* **1933**, *18*, 333-343.

⁶⁴ Rosenthal, G. A. *Life Sci.* **1997**, *60*, 1635-1641.

⁶⁵ Kuehl Jr, F. A.; Wolf, F. J.; Trenner, N. R.; Peck, R. L.; Buhs, R. P.; Howe, E.; Putter, I.; Hunnewell, B. D.; Ormond, R.; Downing, G. *J. Am. Chem. Soc.* **1955**, *77*, 2344-2345.

Two aminoxy analogues of peptides were synthesized by Briggs and Morley (Figure 27).⁶⁶ Compared to aspartame, its aminoxy analogue was not sweet. Unlike gastrine, its aminoxy analogue presented no stimulation of gastric acid secretion, however resistant to enzymatic hydrolysis.



Figure 27: Aminoxy analogues of aspartame and gastrine.

II.2 Methods for the introduction of oxyamine function

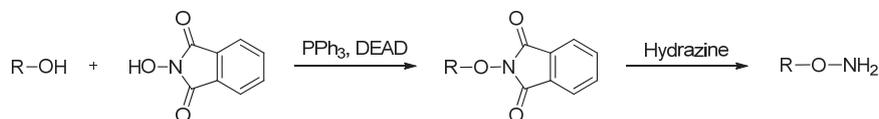
The reported methods for introducing oxyamine function are mainly divided into two approaches: (i) nucleophilic substitution reaction; (ii) amination of alcohols

II.2.a Nucleophilic substitution reaction

II.2.a (1) Alcohol to oxyamine transformation

This transformation is generally performed using *N*-hydroxyphthalimide (PhthN-OH) under Mitsunobu-like conditions or using *N*-protected hydroxylamine derivatives under nucleophilic substitution condition, followed by deprotection to provide the oxyamine product.

The synthesis of free *O*-substituted hydroxylamine using Mitsunobu reaction was firstly reported by Grochowski and Jurczak.⁶⁷ Treatment of a primary or secondary alcohol with PhthN-OH, PPh₃ and diethyl azodicarboxylate (DEAD) furnished the *O*-alkyl phthalimide. Subsequently, the phthaloyl group was removed by hydrazinolysis to give the free oxyamine (Scheme 7).⁶⁸



Scheme 7: Synthesis of oxyamine compounds using Mitsunobu reaction followed by hydrazinolysis.

In the case of second alcohols, the Mitsunobu reaction leads usually to the inversion of

⁶⁶ Briggs, M. T.; Morley, J. S. *J. Chem. Soc., Perkin Trans I.* **1979**, 2138-2143.

⁶⁷ Grochowski, E.; Jurczak, J. *Synthesis* **1976**, 682-684.

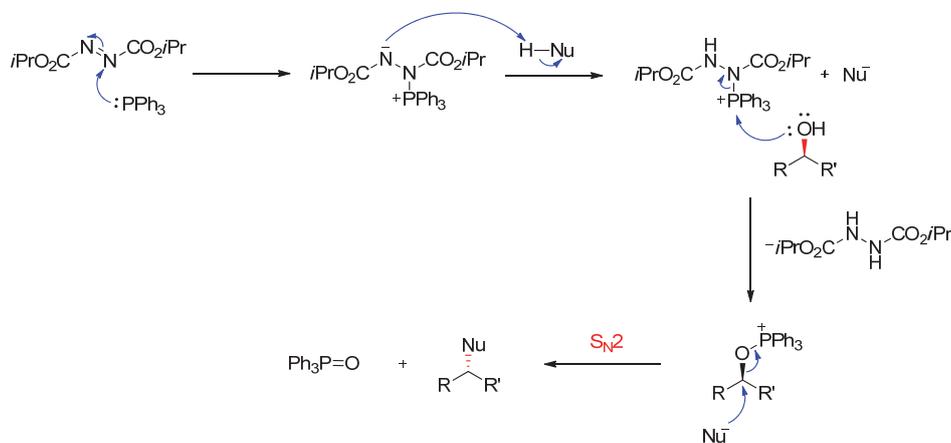
⁶⁸ Chemla, P. *Tetrahedron Lett.* **1993**, 34, 7391-7394.

configuration (Scheme 8).⁶⁹



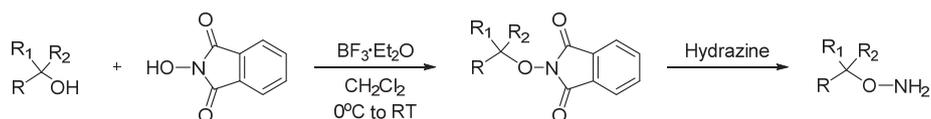
Scheme 8: Mitsunobu reaction with inversion of configuration.

The proposed mechanism of the reaction between a nucleophile (NuH) and a chiral secondary alcohol in the presence of DIAD and PPh₃ is shown in Scheme 9. Reaction of PPh₃ and DIAD quickly generates a zwitterionic intermediate which is able to deprotonate the nucleophile to form the anionic nucleophile and the phosphonium ion. The alcohol then attacks the phosphonium ion to form an oxyphosphonium salt, which was attacked by the deprotonated nucleophile in an S_N2 manner to yield the final product with inversion of configuration.



Scheme 9: Proposed mechanism of Mitsunobu reaction with the inversion of configuration.^{69a}

Different from the S_N2 mechanism of the Mitsunobu reaction, the preparation of tertiary alkoxyamine reported by Palandoken *et al.* involves the displacement of a hydroxyl group via S_N1 mechanism.⁷⁰ The tertiary alcohol was treated with stoichiometric amount of BF₃·Et₂O and PhthN-OH in CH₂Cl₂ to give the *O*-alkyl phthalimide. However, the S_N1 mechanism led to a loss of stereochemistry, and this method is not applicable to secondary alcohol. Hydrazinolysis of the phthalimide group afforded the (*tert*-alkoxy)amine product in good yield (Scheme 10).



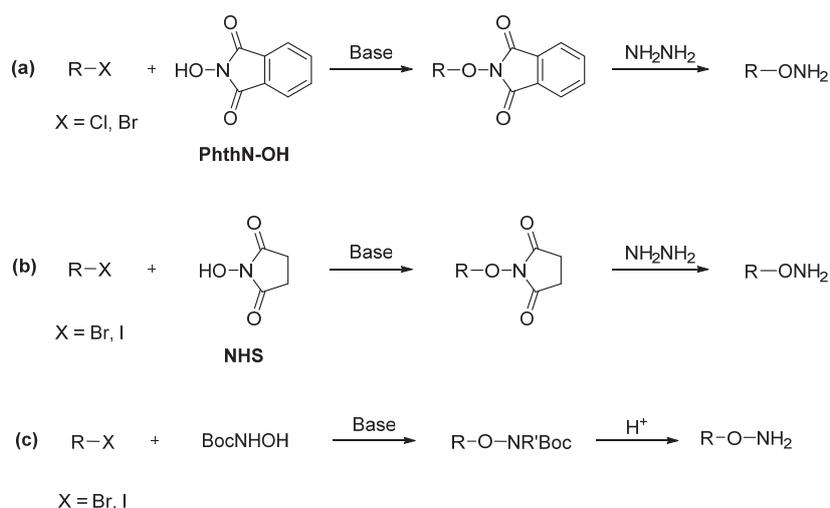
Scheme 10: Preparation of tertiary alkoxyamine reported by Palandoken *et al.*⁷⁰

⁶⁹ (a) Swamy, K. C.; Kumar, N. N.; Balaraman, E.; Kumar, K. V. *Chem. Rev.* **2009**, *109*, 2551-2651; (b) Shin, I.; Lee, M.-r.; Lee, J.; Jung, M.; Lee, W.; Yoon, J. *J. Org. Chem.* **2000**, *65*, 7667-7675. (c) Mitsunobu, O.; Yamada, M.; Mukaiyama, T. *B. Chem. Soc. Jpn.* **1967**, *40*, 935-939; (c) Mitsunobu, O.; Eguchi, M. *B. Chem. Soc. Jpn.* **1971**, *44*, 3427-3430.

⁷⁰ Palandoken, H.; Bocian, C. M.; McCombs, M. R.; Nantz, M. H. *Tetrahedron Lett.* **2005**, *46*, 6667-6669.

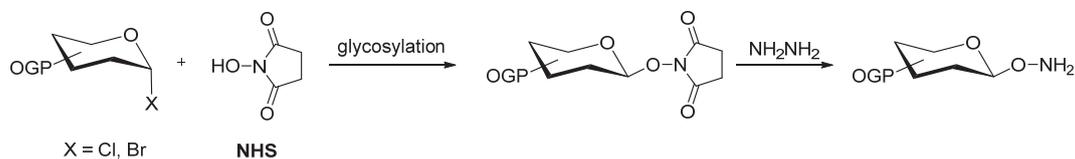
II.2.a (2) Halogen to oxyamine transformation

The S_N2 displacement of halides can also be applied for the introduction of aminoxyyl functions, by using *N*-hydroxyphthalimide,⁷¹ *N*-hydroxysuccinimide (NHS),⁷² or other *N*-protected hydroxylamine derivatives⁷³ as nucleophile reagents (Scheme 11). For example, Jones *et al.* reported that the alkyl iodide and bromides can be easily converted to *N*-protected aminoxy compounds by treatment with commercially available *N*-(*tert*-butyloxycarbonyl)hydroxylamine (BocNHOH) and 1,8-diazabicycloundec-7-ene (DBU).^{73c} Removal of the Boc group gave the free aminoxy (Scheme 11c).



Scheme 11: Displacement of halogens to prepare the aminoxy derivatives.^{71,72,73}

Successful stereoselective synthesis of glycosyloxysuccinimides via glycosylation of NHS with glycosyl halides was reported by Cao *et al.* Treatment of the obtained glycosyloxy succinimides with an excess of hydrazine hydrate gave the corresponding *O*-glycosyl hydroxylamine (Scheme 12).^{72a}



Scheme 12: Synthesis of *O*-glycosyl hydroxylamine by glycosylation and hydrazinolysis.

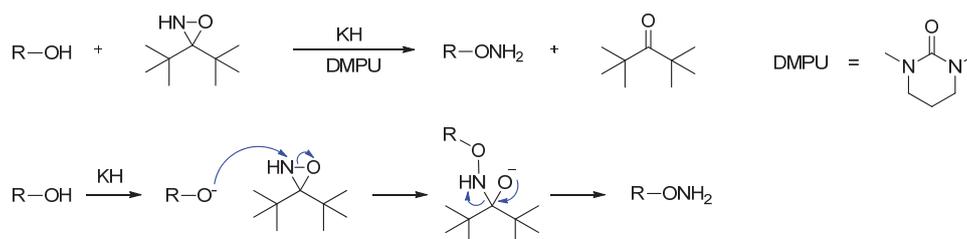
⁷¹ (a) Weller, C. E.; Huang, W.; Chatterjee, C. *ChemBioChem* **2014**, *15*, 1263-1267; (b) Sharma, C.; Thadhaney, B.; Pemawat, G.; Talesara, G. *Indian J. Chem.* **2008**, *47*, 1892-1897; (c) Canne, L. E.; Bark, S. J.; Kent, S. B. *J. Am. Chem. Soc.* **1996**, *118*, 5891-5896.

⁷² (a) Cao, S.; Tropper, F. D.; Roy, R. *Tetrahedron* **1995**, *51*, 6679-6686; (b) Andersson, M.; Oscarson, S. *Glycoconjugate. J.* **1992**, *9*, 122-125.

⁷³ (a) Carrasco, M. R.; Alvarado, C. I.; Dashner, S. T.; Wong, A. J.; Wong, M. A. *J. Org. Chem.* **2010**, *75*, 5757-5759; (b) Jones, D. S.; Hammaker, J. R.; Tedder, M. E. *Tetrahedron Lett.* **2000**, *41*, 1531-1533; (c) Porter, N. A.; Caldwell, S. E.; Lowe, J. R. *J. Org. Chem.* **1998**, *63*, 5547-5554.

II.2.b Amination of alcohols

The direct electrophilic amination of alcohols, with the advantage of retention of alcohol stereochemistry, provides another approach to prepare the aminoxy product. For example, Choong *et al.* reported that a substituted oxaziridine can be used as an electrophilic amination reagent for the transformation of an alkoxide nucleophile to the corresponding alkoxyamine (Scheme 13).⁷⁴



Scheme 13: Amination of alcohols to alkoxyamines.⁷⁴

II.2 α -, β - and γ -aminoxy acids

Compared to amine function ($\text{p}K_a \approx 10$), the oxyamine is a weaker base ($\text{p}K_a \approx 4.6$) due to lone pair repulsion between adjacent nitrogen and oxygen. Likewise, the *N*-oxy amide bond ($\text{p}K_a$ 10-15) is more acidic than a regular amide bond ($\text{p}K_a \approx 20$ -25) (Table 1).⁷⁵ This fact makes NH group in *N*-oxyamide a better hydrogen bond donor and the carbonyl group good hydrogen bond acceptor.

Amine/amide	Oxyamine/ <i>N</i> -oxyamide
$\text{R-NH}_3^+ \rightleftharpoons \text{R-NH}_2$ <p style="text-align: center;">pKa \approx 10</p>	$\text{R-ONH}_3^+ \rightleftharpoons \text{R-ONH}_2$ <p style="text-align: center;">pKa \approx 4.6</p>
$\text{R-C(=O)-NH-R}' \rightleftharpoons \text{R-C(=O)-N}^-\text{R}'$ <p style="text-align: center;">pKa \approx 20-25</p>	$\text{R-C(=O)-N(OR)R}' \rightleftharpoons \text{R-C(=O)-N}^-\text{OR}'$ <p style="text-align: center;">pKa \approx 10-15</p>

Table 1: The $\text{p}K_a$ value of amine, oxyamine, amide, *N*-oxyamide reported in literatures.⁷⁵

⁷⁴ Choong, I. C.; Ellman, J. A. *J. Org. Chem.* **1999**, *64*, 6528-6529.

⁷⁵ (a) Bissot, T. C.; Parry, R. W.; Campbell, D. H. *J. Am. Chem. Soc.* **1957**, *79*, 796-800; (b) Bordwell, F. G. *Acc. Chem. Res.* **1988**, *21*, 456-463; (c) Bordwell, F. G.; Fried, H. E.; Hughes, D. L.; Lynch, T. Y.; Satish, A. V.; Whang, Y. E. *J. Org. Chem.* **1990**, *55*, 3330-3336.

A few family of foldamers has been developed from α -, β - and γ -aminoxy acids. It has been reported that peptides constructed from aminoxy acids can fold into turn (α N-O turns, β N-O turns, γ N-O turns and reverse turns) or helix structure through intramolecular hydrogen bond. These properties are very important for the specific biological functions of biomolecules which are closely related to their secondary structures. The current methodology to analyse the secondary structures mainly includes NMR spectroscopy, circular dichroism (CD) spectroscopy, X-ray crystallography and theoretical calculation, etc.

II.2.a α -Aminoxy acids

Yang and co-workers have reported that α -aminoxy acids induced a strong eight-membered-ring intramolecular hydrogen bond between adjacent residues (called α N-O turn) when it was incorporated into a peptide backbone (Figure 28). The D- α -aminoxy acids induce a right-handed α N-O turn while the L-enantiomers induce a left-handed N-O turn.⁶⁰ They have also shown that the homochiral oligomers of D- α -aminoxy acids adopted a right-handed 1.8₈ helix consisting of consecutive N-O turn.⁷⁶

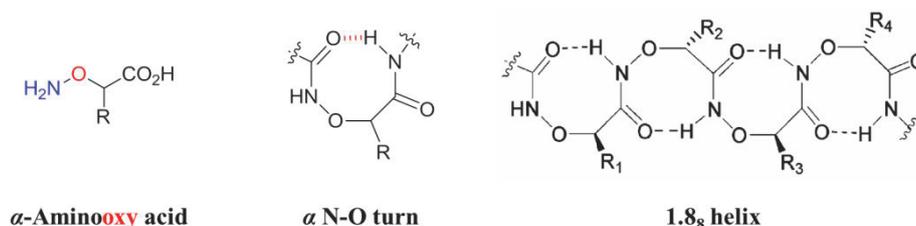


Figure 28: Structure of α -aminoxy acid, α N-O turn and 1.8₈ helix.⁷⁶

In order to study the secondary structures of *N*-oxy peptides, Yang *et al.* examined the ¹H NMR and IR spectroscopies of three diamide compounds with one or two *N*-oxyamide units (A, B, C, Figure 29). All of three compounds have the general formula *t*-Bu-CO-NH-O-CH₂-CO-NRR'.⁷⁷

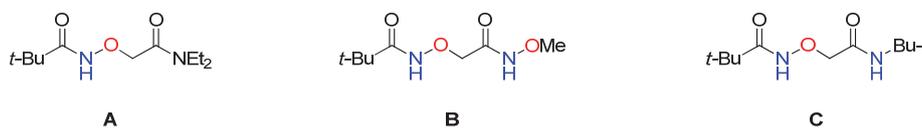


Figure 29: Structures of three diamides compounds for NMR and IR study.⁷⁷

The NH stretch region FT-IR spectra of these compounds in CH₂Cl₂ at room temperature

⁷⁶ (a) Peter, C.; Daura, X.; van Gunsteren, W. F. *J. Am. Chem. Soc.* **2000**, *122*, 7461-7466; (b) Wu, Y.-D.; Wang, D.-P.; Chan, K. W.; Yang, D. *J. Am. Chem. Soc.* **1999**, *121*, 11189-11196; (c) Yang, D.; Qu, J.; Li, B.; Ng, F.-F.; Wang, X.-C.; Cheung, K.-K.; Wang, D.-P.; Wu, Y.-D. *J. Am. Chem. Soc.* **1999**, *121*, 589-590.

⁷⁷ Yang, D.; Ng, F.-F.; Li, Z.-J.; Wu, Y.-D.; Chan, K. W.; Wang, D.-P. *J. Am. Chem. Soc.* **1996**, *118*, 9794-9795.

were shown in Figure 30, with the subtraction of the spectrum of pure CH_2Cl_2 . The spectra of compound **A** at the concentration of 5 mM showed a single peak at 3400 cm^{-1} , corresponding to the stretching frequency of the non-hydrogen-bonded NH of an *N*-oxyamide (**A**, Figure 30). Two peaks at 3386 and 3198 cm^{-1} , respectively assigned to the non-hydrogen-bonded amide NH (at the *N*-terminus) and an intramolecular hydrogen-bonded amide NH (at the *C*-terminus), were observed from the IR spectra of compound **B** at 0.5 mM (**B**, Figure 30). For the IR spectra of compound **C**, there are two major peaks at 3384 and 3294 cm^{-1} , and a small shoulder peak at 3427 cm^{-1} , which were assigned to be the stretching frequency of non-hydrogen-bonded *N*-oxyamide NH, hydrogen-bonded *N*-isobutyl amide NH and the non-hydrogen-bonded isobutyl amide NH, respectively (**C**, Figure 30).

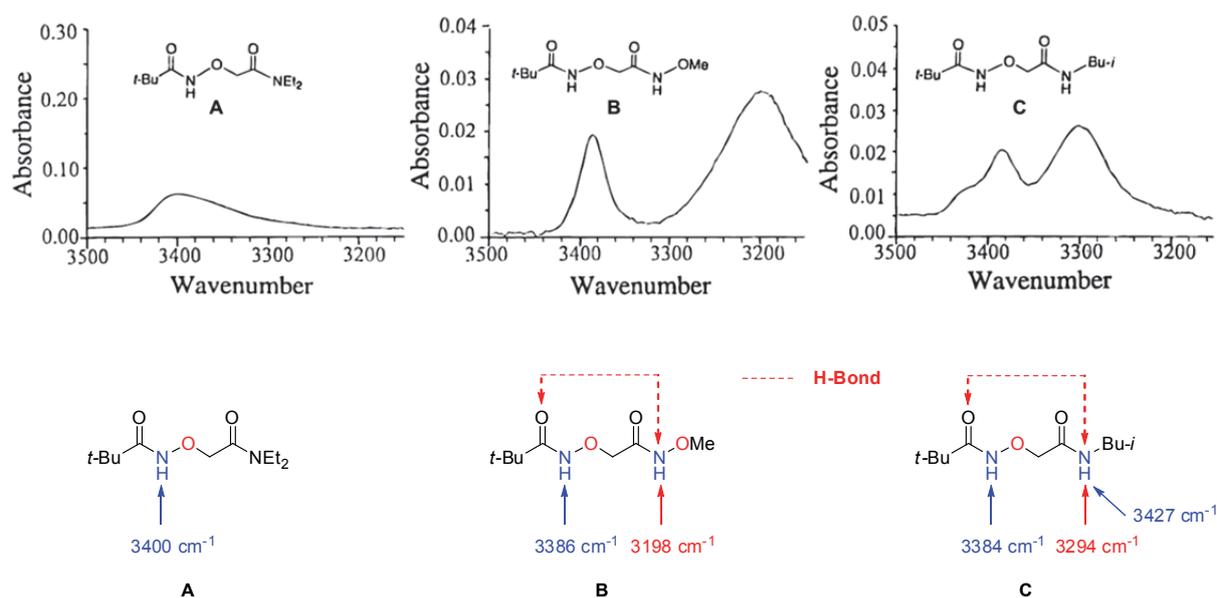


Figure 30: (a) N-H stretch region FT-IR data of diamide in CH_2Cl_2 at room temperature after subtraction of the spectrum of pure CH_2Cl_2 . From left to right: **A** (5 mM), maximum at 3400 cm^{-1} ; **B** (0.5 mM), maxima at 3386 and 3198 cm^{-1} ; **C** (0.5 mM), maxima at 3427 (shoulder), 3384 and 3294 cm^{-1} .⁷⁷

The ^1H NMR chemical shifts of the amide protons for compounds **A**, **B** and **C** tested in CD_2Cl_2 were listed in Table 2. For compound **A**, the chemical shift of *N*-oxy amide NH appeared at 9.74 ppm at 5 mM and without change in diluted concentration. For compound **B** at or below 1 mM, two singlets at 11.41 and 8.71 ppm were assigned to an intramolecular hydrogen-bonded NH, and a non-hydrogen-bonded NH, respectively. The chemical shifts of the two NH protons in compound **C** appeared at 8.63 ppm (singlet) and 8.28 ppm (triplet), which corresponded to the *N*-oxy amide NH and the isobutyl amide NH, respectively. The unusually downfield chemical shift at 8.28 ppm suggested that the *C*-terminus amide NH was involved in an intramolecular hydrogen bonding.⁷⁷

Compounds	A	B	C
δ NH-CO (ppm)	9.74 (s)	11.41 (s)	8.63 (s)
		8.71 (s)	8.28 (t)

Table 2: Chemical shifts of amide protons for compounds **A**, **B** and **C** in CD_2Cl_2 at 25 °C. **A** at 5 mM, **B** and **C** at 0.25 mM.⁷⁷

From the ^1H NMR and IR data of the NH in compounds **A**, **B**, **C**, we can suggest the general chemical shifts and FT-IR stretch regions of the hydrogen-bonded and non-hydrogen-bonded NH in amide and *N*-oxyamide in dichloromethane (Table 3).

		NMR (ppm)	IR (cm^{-1})
Amide	Non-hydrogen-bonded NH	6-7	~3400
	Hydrogen-bonded NH	>8	~3300
<i>N</i> -Oxyamide	Non-hydrogen-bonded NH	8-9	~3400
	Hydrogen-bonded NH	>10	~3200

Table 3: The general chemical shifts and FT-IR stretch regions of the hydrogen-bonded and non-hydrogen-bonded NH in amide and *N*-oxyamide.

Three amide NH signals at 9.25, 8.28, and 5.23 ppm were observed in the ^1H NMR spectra of the tripeptide **D** at or below 1 mM, which were assigned to *N*-oxy amide NH, alanine NH, and valine carbamate NH, respectively. The α N-O turn (the eight-membered-ring intramolecular hydrogen bond Val-C=O...HN-Ala) in tripeptide **D** was evidenced from its unusually downfield δ (Ala-NH) of 8.28 ppm (Figure 31).⁷⁷

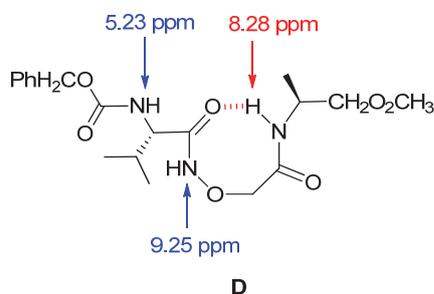


Figure 31: Structure of the tripeptide **D**.⁷⁷

Because of the acidity of their *N*-oxy amide protons, peptides constructed from α -aminooxy acids can also act as good receptor for anion, like carboxylate and chloride

through intermolecular hydrogen bond.⁷⁸ For example, Yang and co-workers have developed two cyclic hexapeptides (**A** and **B** in Figure 32) as chloride ion receptors through intermolecular hydrogen bond. The cyclic hexapeptide **A**, which was comprised of alternating D,L- α -aminoxy acids, showed affinities for halides with higher selectivity for Cl⁻ ion than F⁻ and Br⁻ in nonpolar solvent (Figure 32a).^{78a} Another cyclic hexapeptide **B** consisting of alternating D- α -amino and D- α -aminoxy acids functioned as an efficient chloride ion receptor (Figure 32b).^{78b}

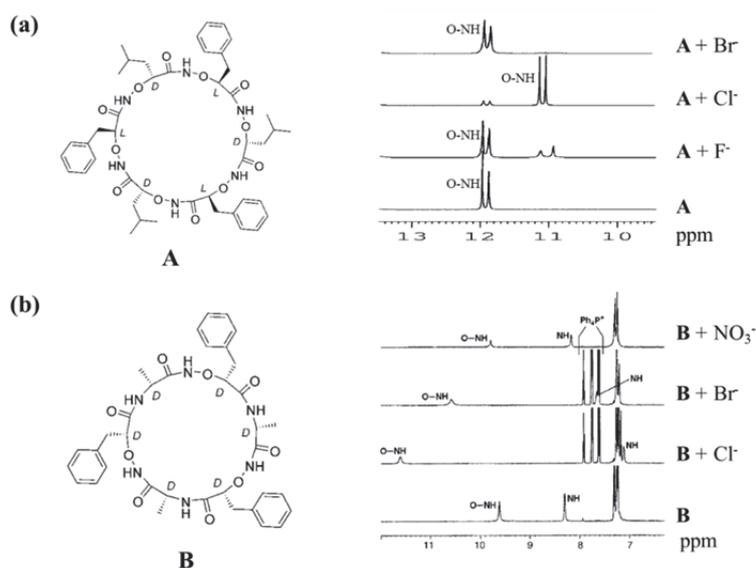


Figure 32: (a) Structure of cyclic hexapeptide **A** and the amide NH region of overlaid ¹H NMR spectra of free **A** and the 1:1 mixture of **A** with (*n*-Bu)₄NF, Ph₄PCl, and Ph₄PBr, respectively (3 mM in CDCl₃ at -30 °C);^{78a} (b) Structure of cyclic hexapeptide **B** and the amide NH region of overlaid ¹H NMR spectra of free **B** and 1:1 mixtures of **B** with Ph₄PCl, Ph₄PBr, or Bu₄NNO₃ (4 mM in CD₂Cl₂ at 298 K).^{78b}

A cyclic tetrapeptide **C** was prepared by alternating C-linked carbo- β -amino acid and (*R*)-aminoxy acid.⁷⁹ NMR and mass spectral studies of this cyclic peptide showed selective and appreciable binding affinity with the chloride ion, meaning that it could be used as a selective chloride ion receptor (Figure 33).

⁷⁸ (a) Yang, D.; Li, X.; Sha, Y.; Wu, Y. D. *Chem. Eur. J.* **2005**, *11*, 3005-3009. (b) Yang, D.; Qu, J.; Li, W.; Zhang, Y.-H.; Ren, Y.; Wang, D.-P.; Wu, Y.-D. *J. Am. Chem. Soc.* **2002**, *124*, 12410-12411; (c) Yang, D.; Li, X.; Fan, Y.-F.; Zhang, D.-W. *J. Am. Chem. Soc.* **2005**, *127*, 7996-7997.

⁷⁹ Sharma, G. V.; Manohar, V.; Dutta, S. K.; Sridhar, B.; Ramesh, V.; Srinivas, R.; Kunwar, A. C. *J. Org. Chem.* **2010**, *75*, 1087-1094.

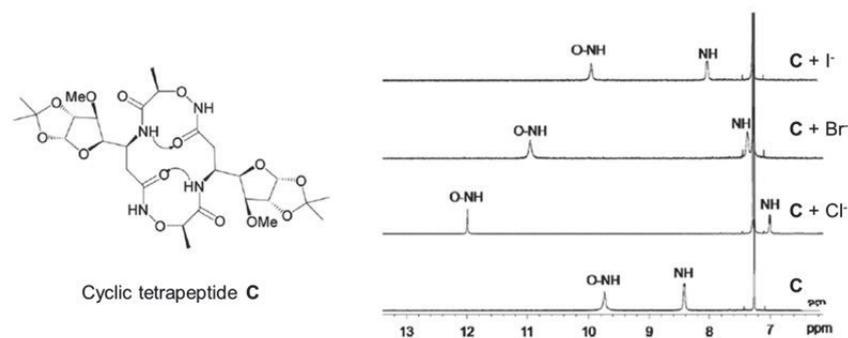


Figure 33: Structures of cyclic tetrapeptide **C** and the chemical shift of amide protons of free **C** and a mixtures of **C** with anions (Cl^- , Br^- , I^-).⁷⁹

II.2.b β -Aminoxy acids

β -Aminoxy acids are analogues of γ -amino acids in which the γ -carbon atom is replaced by an oxygen atom. Thanks to the extra carbon atom in their backbone (compared with α -aminoxy acids), this type of aminoxy acids can provide more substitution patterns, such as β^2 -, β^3 -, $\beta^{2,2}$ -, $\beta^{2,3}$ -, and $\beta^{3,3}$ -aminoxy acids (Figure 34). Most of them incorporated into peptide backbones induce a rigid nine membered-ring hydrogen bond between adjacent residues, which is called β N-O turn (Figure 34).⁸⁰

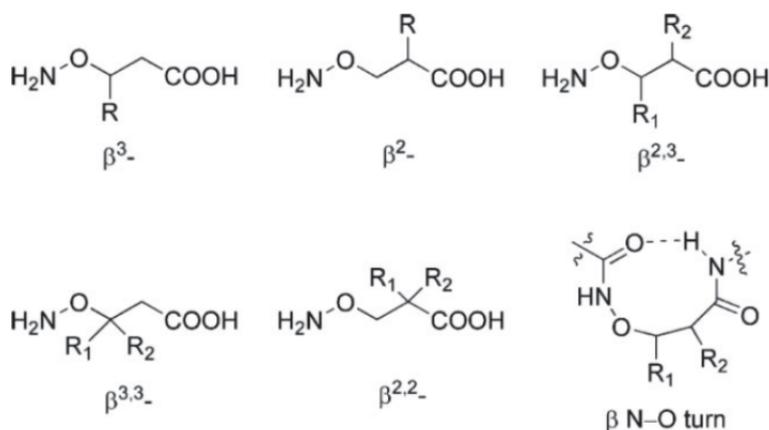


Figure 34: Structures of various substitution patterns of β -aminoxy acids and β N-O turn.⁸⁰

For example, the structural properties of the diamide **A** (Figure 35a) and triamide **B** (Figure 35d) were studied by X-ray crystallography and 2D NMR spectroscopy.^{80b} The crystal structures of **A** and **B** are shown in Figure 35b and Figure 35e, respectively. In the crystal structure of diamide **A**, a β N-O turn between the $\text{C}=\text{O}_i$ and NH_{i+2} groups was observed. Another six-membered-ring hydrogen bond between the NH_{i+2} and NO_{i+1} groups further stabi-

⁸⁰ (a) Li, X.; Yang, D. *Chem. Commun.* **2006**, 3367-3379; (b) Yang, D.; Zhang, Y.-H.; Zhu, N.-Y. *J. Am. Chem. Soc.* **2002**, *124*, 9966-9967.

lized this structure. A well-defined helical structure of triamide **B** was shown in its X-ray structure. Nuclear overhauser effects (NOEs) in 2D NOESY spectra in CDCl_3 at 5 mM were summarized in Figure 35c and Figure 35f, which showed that the β N-O turn structures of **A** and **B** also existed in CDCl_3 .

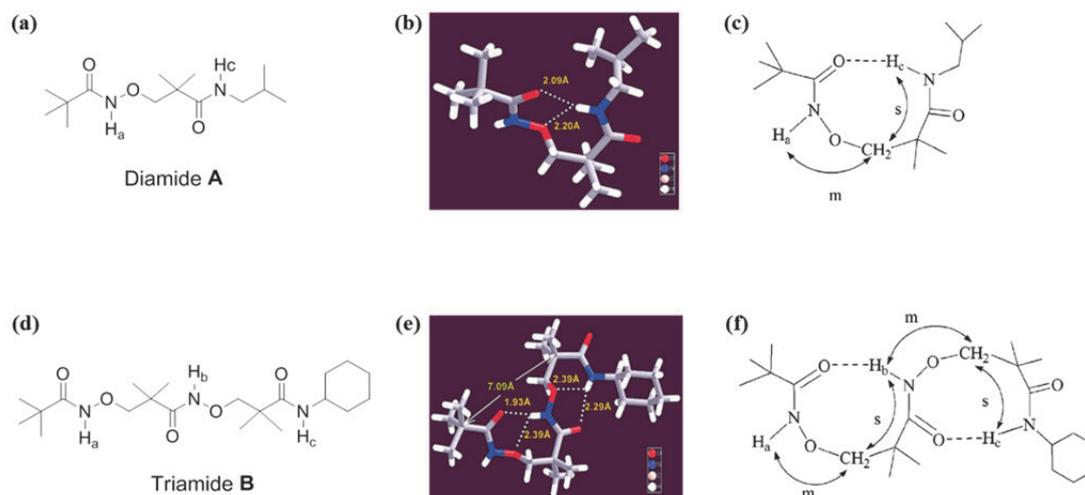
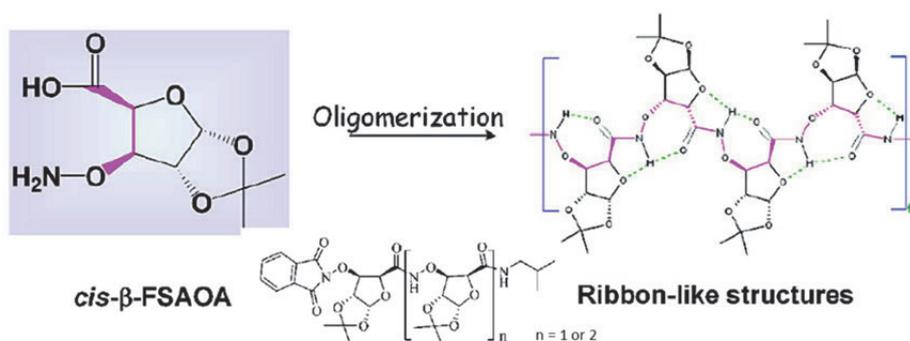


Figure 35: (a) and (d): Structures of diamide **A** and triamide **B**; (b) and (e): X-Ray structures of **A** and **B**; (c) and (f): NOEs of **A** and **B** observed in CDCl_3 at 5 mM (s, strong NOE; M, medium NOE).^{80b}

A *cis*- β -^{2,3}-furanoid sugar aminoxy acid and its short oligomers were designed, synthesized and characterized by Chandrasekhar *et al.*⁸¹ A series of structure studies by DFT calculation, NMR spectroscopies and MD simulation techniques suggested that the short homo-oligomers building block exhibited rigid ribbon-like secondary structures involving 5/7 bifurcated intramolecular hydrogen-bonded rings (Scheme 14).



Scheme 14: Oligomerization of the *cis*- β -^{2,3}-furanoid sugar aminoxy acid.⁸¹

⁸¹ Chandrasekhar, S.; Rao, C. L.; Reddy, M. S.; Sharma, G. D.; Kiran, M. U.; Naresh, P.; Chaitanya, G. K.; Bhanuprakash, K.; Jagadeesh, B. *J. Org. Chem.* **2008**, *73*, 9443-9446.

II.2.c γ -Aminoxy acids

Like the construction of α and β N-O turns, γ N-O turns are the 10-membered-ring structures which are formed through intramolecular hydrogen bonds in peptides consisting of γ -aminoxy acids. It has been reported that γ -aminoxy peptides can form turn and helix structures incorporating γ N-O turns.⁸²

For example, the secondary structures of γ^4 -Ph aminoxy diamide (Figure 36a) have been investigated by using FT-IR, NMR spectroscopy, and X-ray crystallography.^{82a} In the FT-IR spectrum, three peaks were observed at 3428 cm^{-1} , 3388 cm^{-1} , 3324 cm^{-1} , which were respectively assigned to the stretching frequencies of the non-hydrogen-bonded amide NH_b , non-hydrogen-bonded N -oxyamide NH_a and hydrogen-bonded amide NH_b (Figure 36b). The stronger NH_b peak at 3324 cm^{-1} suggested NH_b was involved in a 10-membered-ring intramolecular hydrogen bond. A strong NOE signal was found between H_b and the γ -proton in 2D-NOESY studies (Figure 36c). X-Ray crystallographic structural analysis showed the presence of γ N-O turn, involving a 10-membered-ring intramolecular hydrogen bond between the CO_i and NH_{i+2} in this γ^4 -aminoxy peptides (Figure 36d).

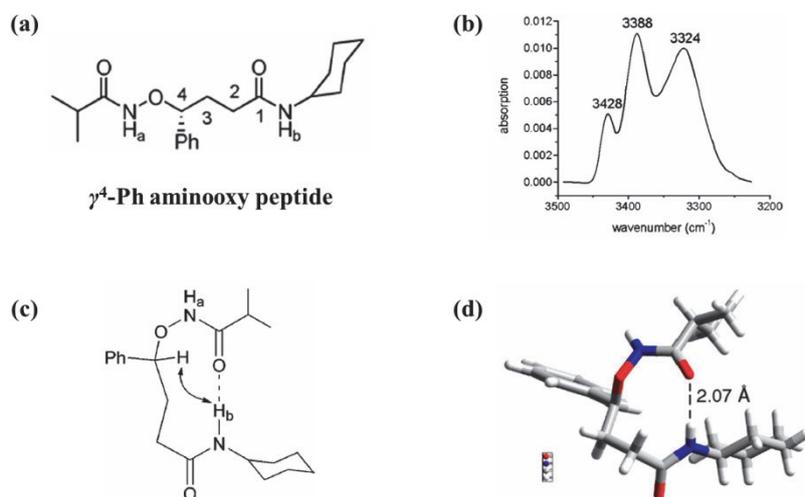


Figure 36: (a) Chemical structure of γ^4 -Ph aminoxy peptide; (b) N-H stretch region FT-IR data of γ^4 -Ph aminoxy peptide in 2 mM CH_2Cl_2 ; (c) NOE signal observed in CDCl_3 at 5 mM; (d) X-Ray structure.^{82a}

⁸² (a) Chen, F.; Zhu, N.-Y.; Yang, D. *J. Am. Chem. Soc.* **2004**, *126*, 15980-15981; (b) Chen, F.; Song, K.-S.; Wu, Y.-D.; Yang, D. *J. Am. Chem. Soc.* **2008**, *130*, 743-755.

III N-O linkage in sugar chemistry

For the development of glycoconjugates, various strategies have been exploited, by direct conjugation to carbohydrates or by linking the carbohydrate to a bifunctional linker which can be further functionalized. Among different approaches, use of oxyamine allows easy conjugation with carbonyl or carboxylic compounds through oxime, *N*-glycoside or *N*-oxy amide bonds. Moreover, the oxyamine function can be easily introduced and oxyamine-derived compounds are quite stable to hydrolytic cleavage.⁸³

The stability of hydrazine and hydroxylamine-based glycoconjugates in aqueous solution has been studied by Gudmundsdottir *et al.*⁸⁴ It has been proved that the hydrolysis rates accelerated under increasing acidic conditions and all the conjugates became increasingly stable as the pH approaches neutrality, with the half-life varied from 1 to 300 days. Besides, the hydrolysis rates of the glycoconjugates formed from electron-rich monosaccharides were significantly more rapid than those from electron-poor monosaccharides (xylose (fastest) > glucose > *N*-acetylglucosamine (slowest)) (Figure 37 and Table 4).

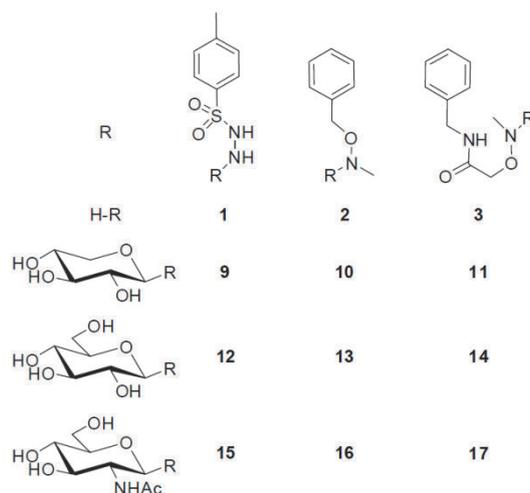


Figure 37: Nine glycoconjugates synthesized and subjected to hydrolysis experiments.⁸⁴

⁸³ Munneke, S.; Prevost, J. R.; Painter, G. F.; Stocker, B. L.; Timmer, M. S. *Org. Lett.* **2015**, *17*, 624-627.

⁸⁴ Gudmundsdottir, A. V.; Paul, C. E.; Nitz, M. *Carbohydr. Res.* **2009**, *344*, 278-284.

Compound	pH 4.0 $t_{1/2}$ (h)	pH 5.0 $t_{1/2}$ (h)	pH 6.0 $t_{1/2}$ (h)
9	2.9	13	29
10	7.8	26	59
11	13	140	540
12	19	48	71
13	11	210	890
14	100	990	5100
15	72	230	1100
16	76	660	2600
17	350	3100	7500

Table 4: Half-lives in aqueous solution of glycoconjugates **9-17**. 2 mM sample in 20 mM NaOAc (pH 4.0, or pH 5.0), or in 20 mM Na₂HPO₄ (pH 6.0) was incubated at 37 °C. 200 μL samples were taken out at different time intervals and quenched at 4 °C with the addition of 400 μL of 200 mM Na₂HPO₄ buffer at pH 7.0 and immediately analyzed by HPLC.⁸⁴

Based on the position of the N-O linkage, the N-O linked carbohydrates are mainly divided into three types, such as *N*-glycosyl hydroxylamine, *O*-glycosyl hydroxylamine and *O*-amino sugar derivatives (Figure 38).

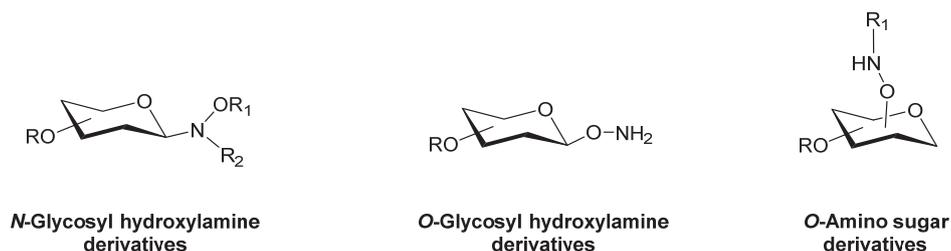


Figure 38: Chemical structure of *N*-glycosyl hydroxylamine, *O*-glycosyl hydroxylamine and *O*-amino sugar derivatives.

III.1 *N*-Glycosyl hydroxylamine derivatives

III.1.a Formation of *N*-glycosyl hydroxylamines

III.1.a (1) Neoglycosylation

N-Glucuronosyl-*N*-hydroxyl aromatic amine (Figure 39) was first synthesized by Radomski and co-workers to study the metabolic reaction of carcinogenic *N*-hydroxyl aromatic amines in the late 1970s.⁸⁵

⁸⁵ (a) Moreno, H. R.; Radomski, J. L. *Cancer Lett.* **1978**, *4*, 85-88; (b) Poupko, J. M.; Hearn, W. L.; Radomski, J. L. *Toxicol. Appl. Pharm.* **1979**, *50*, 479-484.

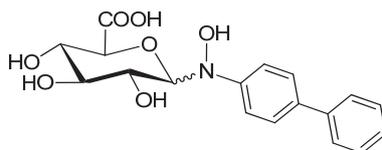
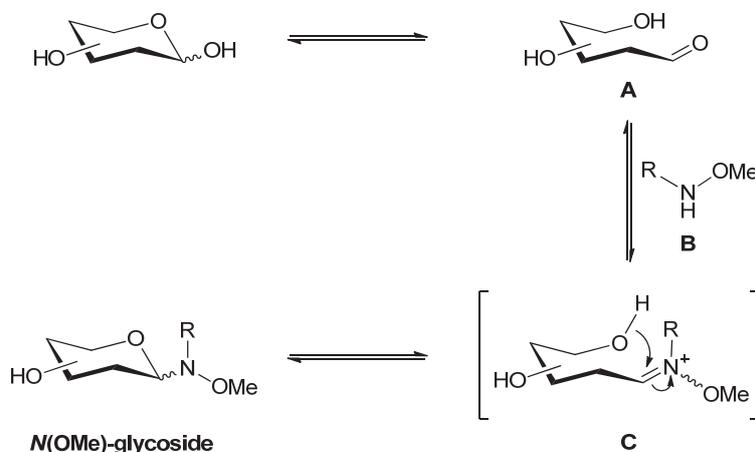


Figure 39: Chemical structure of *N*-(4-biphenyl)-*N*-hydroxyl-D-glucuronosylamine.^{85a}

Twenty years later, Peri *et al.* developed a general method for the glycosylation of oxyamine with reducing sugars where the oxyamines were used as bifunctional linker for the preparation of neoglycopeptides and neoglycolipids.⁸⁶ Thanks to the uniquely high chemical reactivity and specificity of *N,O*-disubstituted hydroxylamine, the reaction can be performed with unprotected and non-activated reducing sugars like glucose, galactose, mannose, *N*-acetylglucosamine and lactose in aqueous solution (in buffer at pH 4) or in polar solvent (generally a mixture of acetic acid and DMF).⁸⁶ The cyclic nature of the saccharide reducing unit is preserved, and this reaction proceeds chemoselectively and with high β -anomeric selectivity without requirement of activation at the anomeric center and protection of functional groups. A proposed mechanism is shown in Scheme 15. The aminooxyl group **B** selectively reacts with the aldehyde of the reducing sugar **A** to give an oxy-iminium intermediate **C**. Subsequent ring closure results in the formation of the *N*-(*O*-methoxy)-glycoside.

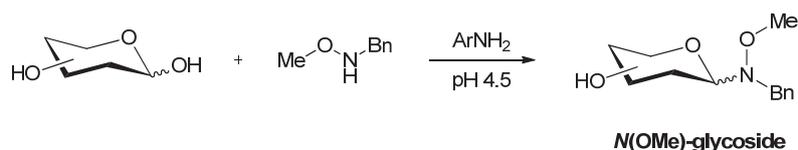


Scheme 15: Mechanism of *N*(OMe)glycoside formation via reaction between reducing sugars and *O,N*-disubstituted hydroxylamines.⁸⁶

Loskot *et al.* have reported that the aniline derivatives can enhance the rate of acid catalyzed neoglycosylation for a range of sugar substrates up to a factor of 32 relative to the uncatalyzed reaction (Scheme 16).⁸⁷

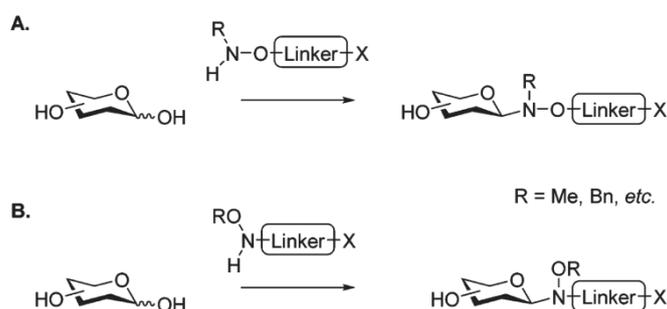
⁸⁶ Peri, F.; Dumy, P.; Mutter, M. *Tetrahedron* **1998**, *54*, 12269-12278.

⁸⁷ Loskot, S. A.; Zhang, J.; Langenhan, J. M. *J. Org. Chem.* **2013**, *78*, 12189-12193.



Scheme 16: Nucleophilic catalysis of *N(OMe)*-neoglycoside formation by aniline derivatives.⁸⁷

In the past few years, this chemoselective ligation method has emerged as a unique and simple strategy to prepare various N-O linked glycoconjugates (Scheme 17). The development, the scope, and the utility of the neoglycosylation reaction were reviewed by Goff and Thorson.⁸⁸



Scheme 17: Two general strategies for the conjugation of carbohydrates to oxyamine linkers.⁸⁸

The relative stability of *O*-linked and *N(OMe)*-linked glycosides to glycosidase was compared by Iqbal and co-workers.⁸⁹ From their experimental data, they suggested that degradation of aminoxy glycoconjugates *in vivo* is more likely to arise from acid-catalyzed, rather than glycosidase-catalyzed hydrolysis.

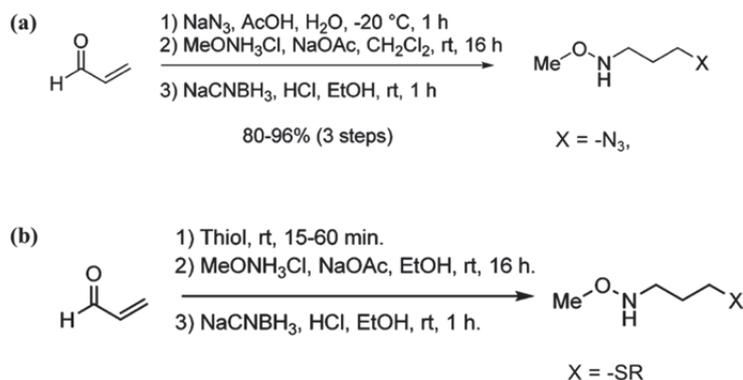
III.1.a (2) Preparation of oxyamine linkers

Much efforts have been devoted for the development of oxyamine linkers.^{83,90} For example, Munneke *et al.* have synthesized a number of *N*-alkylmethoxyamine bifunctional linkers, which contain a *N*-methoxyamine functional group for conjugation to carbohydrates and a terminal group (i.e. amine, azide, thiol, or carboxylic acid) for conjugation to the probe of choice. The reported strategies are rapid and efficient for the synthesis of linkers, which can be conjugated to glycans in excellent yield and showed excellent stability towards hydrolytic cleavage (Scheme 18).⁸³

⁸⁸ Goff, R. D.; Thorson, J. S. *Med. Chem. Comm.* **2014**, *5*, 1036-1047.

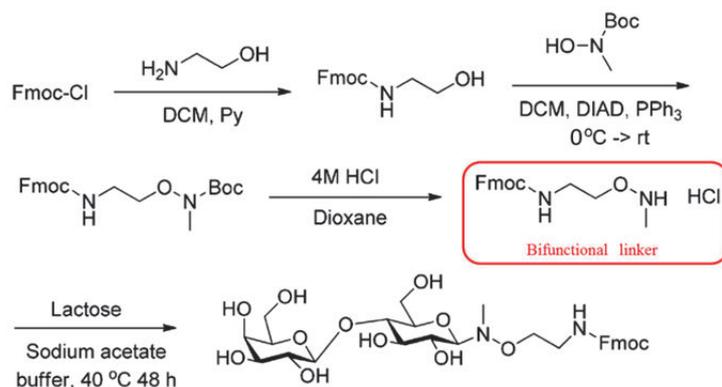
⁸⁹ Iqbal, A.; Chibli, H.; Hamilton, C. J. *Carbohydr. Res.* **2013**, *377*, 1-3.

⁹⁰ (a) Prudden, A. R.; Chinoy, Z. S.; Wolfert, M. A.; Boons, G. J. *Chem. Commun.* **2014**, *50*, 7132-7135; (b) McReynolds, K. D.; Dimas, D.; Le, H. *Tetrahedron Lett.* **2014**, *55*, 2270-2273.



Scheme 18: Synthesis of azide-, amine-, thiol- and carboxylic acid-functionalized oxyamine linkers.⁸³

A multi-functional linker that can readily be attached to the anomeric center of reducing sugars was developed by Prudden *et al.* (Scheme 19).^{90a} Due to the lipophilicity of the Fmoc group, the Fmoc-protected linker can be used to purify glycans prepared by chemoenzymatic approaches using reverse phase column chromatography and monitor progress of enzymatic glycosylations by HPLC. Moreover, the free amine, which is helpful for glycan immobilization and array development, can be easily obtained by removing the Fmoc protecting group under mild basic conditions.



Scheme 19: Synthesis of bifunctional linker and lactose conjugation reported by Prudden *et al.*^{90a}

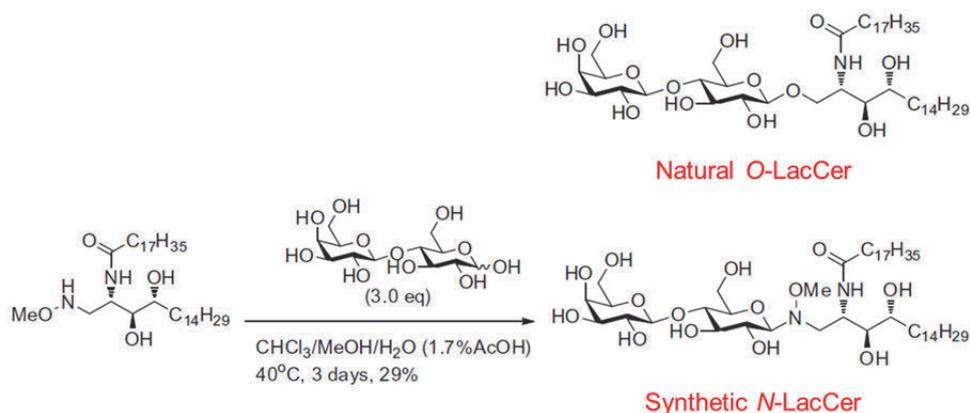
III.1.b Applications of neoglycosylation

Thanks to the simplicity and efficiency, the chemoselective neoglycosylation has been demonstrated as an excellent method for the formation of oligosaccharide analogues, glycoconjugates such as glycolipid and glycopeptide, and glycoproteins, as well as glycoarrays.⁹¹

III.1.b (1) Synthesis of neoglycolipids

⁹¹ Peri, F.; Nicotra, F. *Chem. Commun.* **2004**, 623-627.

A neoglycosphingolipid (*N*-LacCer) was prepared as a natural *O*-lactosylceramide (*O*-LacCer) mimic, in which the carbohydrate moiety was linked to a *N*-methoxyamino-functionalized ceramide (Scheme 20).⁹² The synthetic neoglycolipid showed potent inhibition against hydrolysis of natural *O*-LacCer by a recombinant endoglycoceramidase II from *Rhodococcus* sp.



Scheme 20: Structure of natural lactosylceramide and synthesis of non-natural *N*-LacCer by one step glycosylation reaction.⁹²

III.1.b (2) Synthesis of neoglycopeptides/glycopeptoids

A series of liponeoglycopeptide variants were prepared by neoglycosylation of a *N*-methoxyamine-appended vancomycin aglycon with 2-,3-,4- or 6-*N'*-decanoyl or biphenoyl D-glucopyranose (Figure 40).⁹³ This method led to good yield and excellent stereoselectivity. The activity test against vancomycin-resistant *Enterococci* (VRE) revealed the 3- or 4-acylated glucose was the optimal one for neoglycopeptide lipidation, while the natural ones favored the attachment of lipid on the 2-position (like teicoplanin). These results demonstrate that the natural *O*-glycosidic linkage in glycopeptide can be replaced with the neoglycoside *N*-glycosidic linkage with enhanced bioactivity.

⁹² Ishida, J.; Hinou, H.; Naruchi, K.; Nishimura, S. *Bioorg. Med. Chem. Lett.* **2014**, *24*, 1197-1200.

⁹³ Griffith, B. R.; Krepel, C.; Fu, X.; Blanchard, S.; Ahmed, A.; Edmiston, C. E.; Thorson, J. S. *J. Am. Chem. Soc.* **2007**, *129*, 8150-8155.

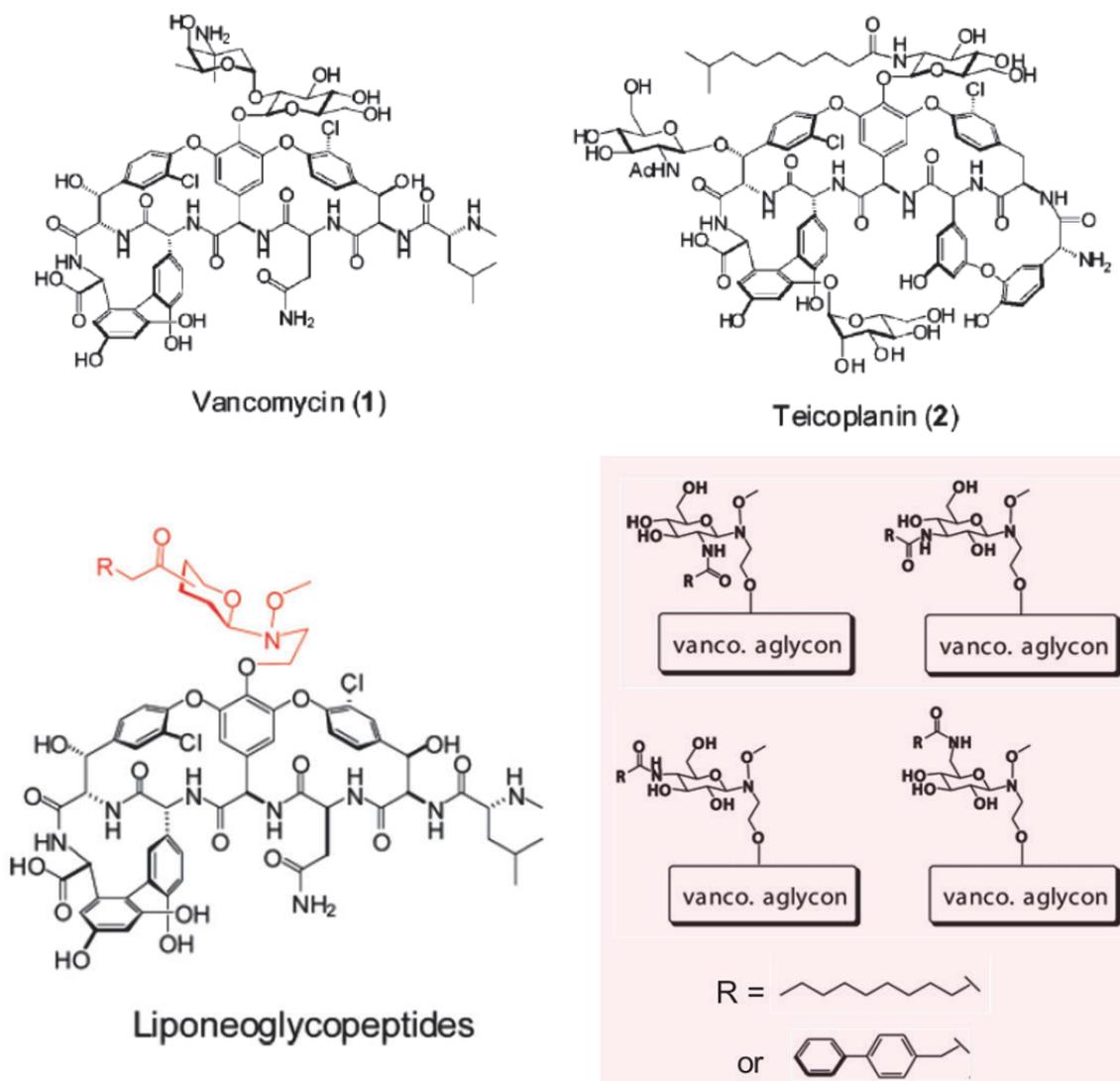
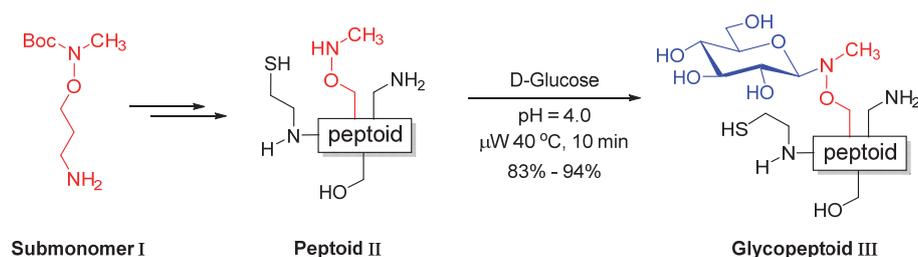


Figure 40: Chemical structure of natural glycopeptide antibiotics vancomycin and teicoplanin, and synthetically modified liponeoglycopeptides.⁹³

Seo and co-workers reported an efficient synthesis of glycopeptoids via microwave assisted chemoselective glycosylation of *N*-alkyl oxyamine containing peptoid with unprotected reducing sugars (Scheme 21).⁹⁴ The protected *N*-alkyl aminoxy amine **I** was synthesized as a bifunctional linker, and then incorporated into oligopeptoids using standard solid phase peptoid synthesis procedures. Oligopeptoids **II** was then neoglycosylated with unprotected reducing sugars to produce the glycopeptoids **III**. The increasing conversion and short reaction time of the glycosylation were achieved by employing microwave irradiation. Reducing sugars like D-glucose, D-maltose, D-melibiose, and maltotriose, ligated smoothly to the peptoid with β -stereoselectivity in good yield.

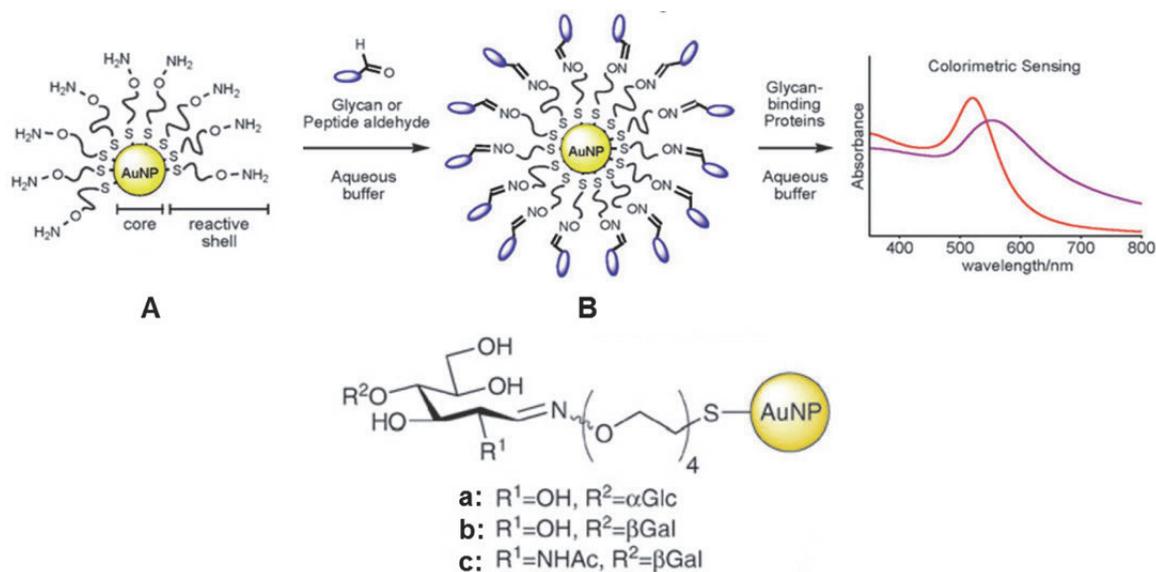
⁹⁴ Seo, J.; Michaelian, N.; Owens, S. C.; Dashner, S. T.; Wong, A. J.; Barron, A. E.; Carrasco, M. R. *Org. Lett.* **2009**, *11*, 5210-5213.



Scheme 21: Synthesis of glycopeptoids by microwave-assisted chemoselective glycosylation.⁹⁴

III.1.b (3) Synthesis of gold glyconanoparticles

The chemoselective oxime coupling was used for the synthesis of gold glyconanoparticles (glyco-AuNPs) by Thygesen *et al* (Scheme 22).⁹⁵ The core-shell AuNPs **A** carrying reactive aminoxy groups on the organic shell was prepared firstly. Subsequently, the unprotected reducing glycans and glycopeptide aldehydes were readily anchored through a single chemoselective reaction onto the core-shell AuNPs **A** to generate the glyco-AuNPs **B**. This methodology avoids the protection and deprotection of glycans and allows direct evaluation of glycan-binding proteins interactions (colorimetric detection) with high specificity.



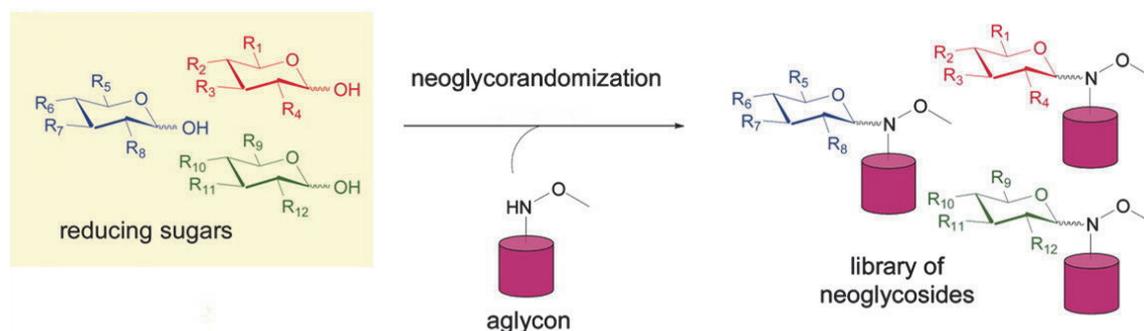
Scheme 22: Strategy for nanoparticle functionalization and detection of protein-glycan interactions.⁹⁵

III.1.b (4) Study of structure-biological activity relationship

Neoglycorandomization method, based upon the chemoselective formation of glycosidic bonds between reducing sugars and a secondary alkoxyamine-containing aglycon to generate a large combinatorial neoglycoside-based libraries, is a powerful tool in glycobiology and

⁹⁵ Thygesen, M. B.; Sorensen, K. K.; Clo, E.; Jensen, K. J. *Chem. Commun.* **2009**, 6367-6369.

drug discovery to understand the relationship between attached sugars and biological activities in the glycosylated products (Scheme 23).⁹⁶



Scheme 23: Principle of neoglycorandomisation.⁹⁶

Thorson and co-workers synthesized a number of digitoxin (also known as cardiac glycoside) analogs with different sugar moieties (Figure 41). Some analogs display significantly enhanced Na^+/K^+ -ATPase-inhibitory potency and tumor specific cytotoxicity.⁹⁶

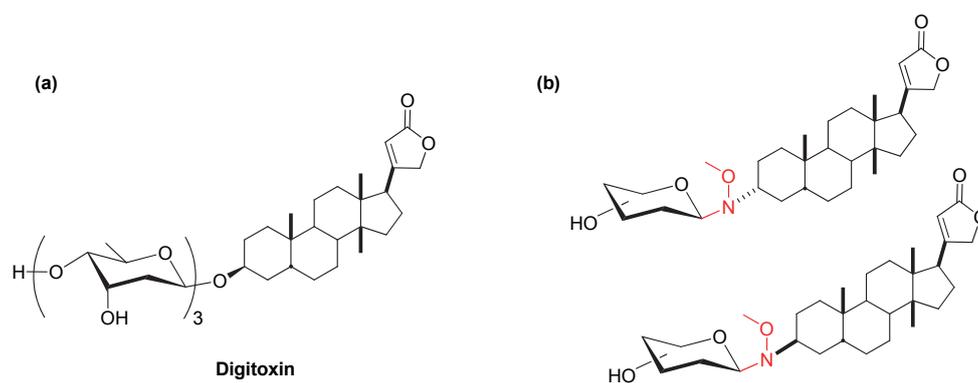
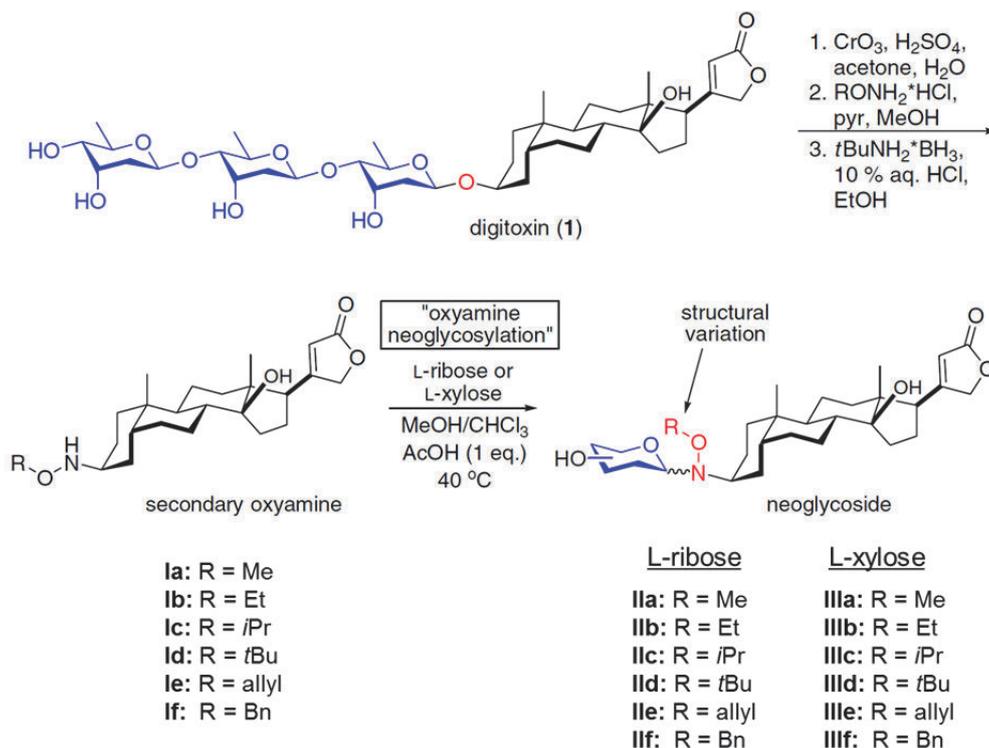


Figure 41: Structure of digitoxin (a) and its neoglycoside analogs (b).

By using the same method, Langenhan *et al* have prepared a new class of digitoxin analogs by varying the nature of *N*-alkoxy group (Scheme 24).⁹⁷ It has been observed that neoglycosides **IIa–c** and **IIIa–c** were more potent cytotoxins than the corresponding aglycons (**Ia–c**), confirming the importance of sugar attachments to the cytotoxicity of cardiac neoglycosides.

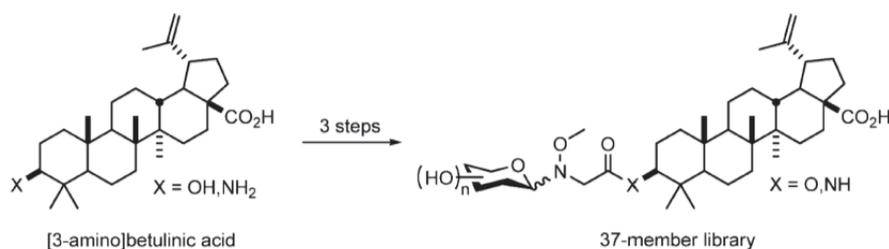
⁹⁶ Langenhan, J. M.; Peters, N. R.; Guzei, I. A.; Hoffmann, F. M.; Thorson, J. S. *Proc. Natl. Acad. Sci U S A* **2005**, *102*, 12305-12310.

⁹⁷ Langenhan, J. M.; Engle, J. M.; Slevin, L. K.; Fay, L. R.; Lucker, R. W.; Smith, K. R.; Endo, M. M. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 670-673.



Scheme 24: Synthesis of digitoxin analogs by oxyamine neoglycosylation.⁹⁷

A library of 37 differentially glycosylated triterpenoid betulinic acids (BA) variants were synthesized using neoglycosylation and their biological activities of anticancer and anti-HIV were examined by Goff and Thorson (Scheme 25).⁹⁸ The biological test revealed that the desired bioactivities of glycosylated BAs were predominately dictated by the appended sugars, and they were also influenced by the nature of alkoxyamine handle connection to the scaffold (i.e., ester versus amide).

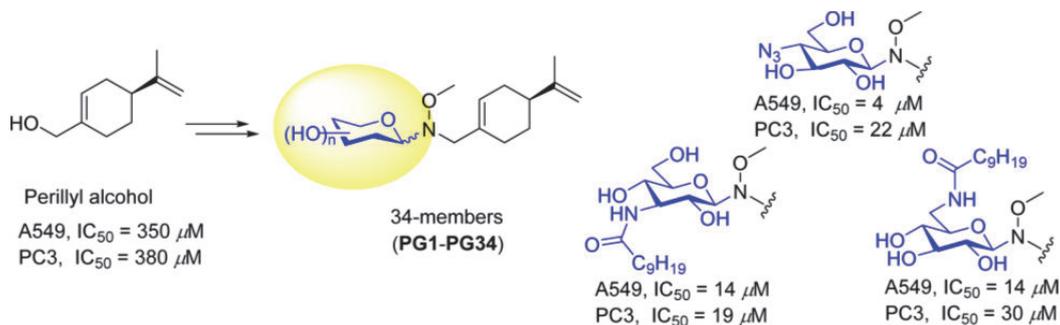


Scheme 25: Synthesis of betulinic acid neoglycosides.⁹⁸

Recently, the same group reported the facile synthesis of a library of 34 differentially perillyl alcohol glycosides (PG1-PG34) by neoglycosylation of the perillyl aglycon with dif-

⁹⁸ Goff, R. D.; Thorson, J. S. *Org. Lett.* **2009**, *11*, 461-464.

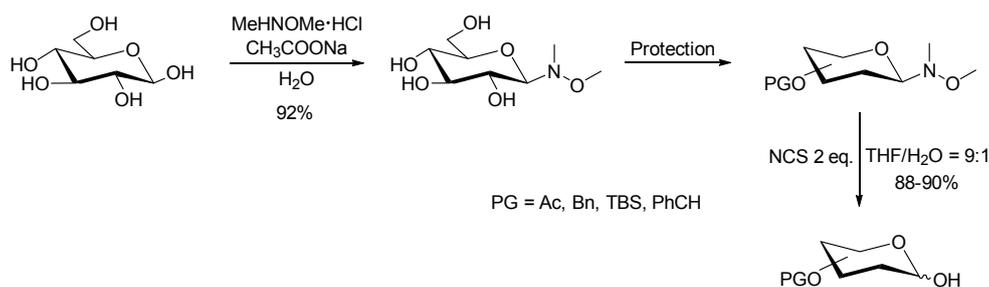
ferent sugars (Scheme 26).⁹⁹ *In vitro* studies show that the antiproliferative activity was dependent on the sugar moiety.



Scheme 26: Synthesis of perillyl alcohol glycosides and their IC₅₀ value against cell viability of two human cancer cell lines (non-small-cell lung A549 and prostate PC3).⁹⁹

III.1.b (5) N-O linkage as an anomeric protecting group

Dasgupta and Nitz reported that the *N,O*-dimethylhydroxylamine could be used as an anomeric protecting group in carbohydrate synthesis, as the *N*-methoxy-*N*-methyl-*N*-glycosides are tolerant of various protecting group manipulations, such as acylation, alkylation, silylation, acetal formation, and stable under glycosylation conditions.¹⁰⁰ The *N*-methoxy-*N*-methyl-*N*-glycosides were prepared under mildly acidic conditions by reacting with *N,O*-dimethylhydroxylamine hydrochloride, and then the glycoside was protected with desired protecting groups. Finally, the oxyamino group can be selectively removed with *N*-chlorosuccinimide (NCS) in more than 88% yield (Scheme 27).



Scheme 27: Carbohydrate synthesis using *N,O*-dimethylhydroxylamine as an anomeric protecting group.¹⁰⁰

⁹⁹ Nandurkar, N. S.; Zhang, J.; Ye, Q.; Ponomareva, L. V.; She, Q. B.; Thorson, J. S. *J. Med. Chem.* **2014**, *57*, 7478-7484.

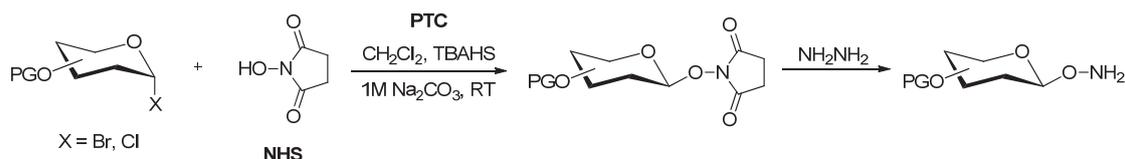
¹⁰⁰ Dasgupta, S.; Nitz, M. *J. Org. Chem.* **2011**, *76*, 1918-1921.

III.2 *O*-Glycosyl hydroxylamine derivatives

III.2.a Preparation of *O*-glycosyl hydroxylamines

The *O*-glycosyl hydroxylamines, widely used as efficient intermediates for the preparation of glycoconjugates, were firstly reported as a new class of monosaccharides by Nashed *et al.*¹⁰¹ The Mitsunobu reaction of PhthN-OH with reducing sugars, followed by hydrazinolysis, has been reported for the synthesis of *O*-glycosyl hydroxylamines.¹⁰² *O*-Glycopyranosyl-*N*-hydroxysuccinimides of glucose and lactose have been synthesized, by using peracetylated sugars, peracetylated glycosyl bromides, or benzoylated thioethyl glycosides as glycosyl donors.¹⁰³

The phase transfer catalyzed (PTC) condition was also reported and applied for the synthesis of *O*-glycosyl hydroxylamine with high stereoselectivity (Scheme 28).¹⁰⁴ In this reaction, NHS is glycosylated with protected sugars using tetra-*n*-butylammonium hydrogen sulfate (TBAHS) as catalyst in a CH₂Cl₂/1 M Na₂CO₃ biphasic system at room temperature.



Scheme 28: Phase transfer catalyzed (PTC) synthesis of *O*-glycosyl hydroxylamine.¹⁰⁴

The glycosylation of PhthN-OH using glycosyl fluoride as a donor and BF₃·Et₂O as a promoter was described by Dumy and co-workers.¹⁰⁵ Chen *et al.* reported that both of the α - and β -galactosyl hydroxylamine can be synthesized from the same precursor using different promoters (Scheme 29).¹⁰⁶ The α -glycoside was prepared by treating the peracetylated galactose with trimethylsilyl trifluoromethanesulfonate (TMSOTf) in acetonitrile at low temperature, while the β -glycoside using BF₃·Et₂O in CH₂Cl₂. The NHS group was cleaved by hydrazinolysis to produce the corresponding *O*-galactosyl hydroxylamine.

¹⁰¹ Nashed, E. M.; Grochowski, E.; Czyzewska, E. *Carbohydr. Res.* **1990**, *196*, 184-190.

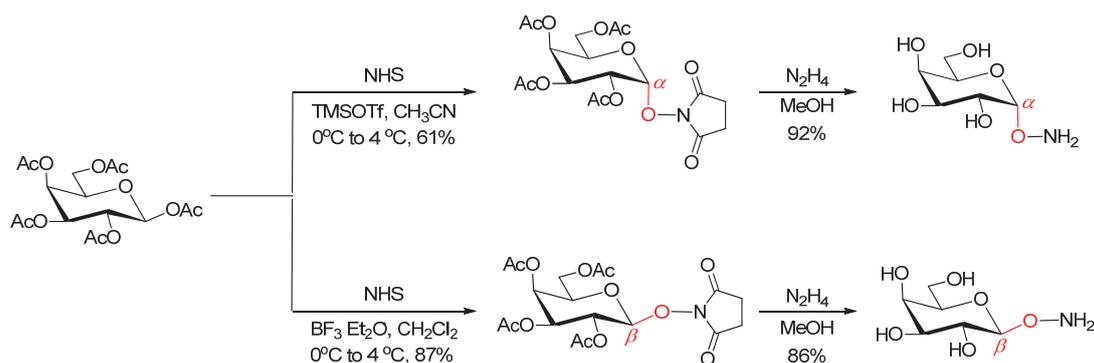
¹⁰² (a) Yang, D.; Kim, S. H.; Kahne, D. *J. Am. Chem. Soc.* **1991**, *113*, 4715-4716; (b) Grochowski, E.; Jurczak, J. *Carbohydr. Res.* **1976**, *50*, C15-C16.

¹⁰³ Andersson, M.; Oscarson, S. *Glycoconjugate. J.* **1992**, *9*, 122-125.

¹⁰⁴ (a) Rodriguez, E. C.; Winans, K. A.; King, D. S.; Bertozzi, C. R. *J. Am. Chem. Soc.* **1997**, *119*, 9905-9906; (b) Rodriguez, E. C.; Marcaurette, L. A.; Bertozzi, C. R. *J. Org. Chem.* **1998**, *63*, 7134-7135; (c) Cao, S.; Tropper, F. D.; Roy, R. *Tetrahedron* **1995**, *51*, 6679-6686.

¹⁰⁵ (a) Dulery, V.; Renaudet, O.; Philouze, C.; Dumy, P. *Carbohydr. Res.* **2007**, *342*, 894-900; (b) Renaudet, O.; Dumy, P. *Tetrahedron Lett.* **2001**, *42*, 7575-7578

¹⁰⁶ Chen, W.; Xia, C.; Cai, L.; Wang, P. G. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 3859-3862.



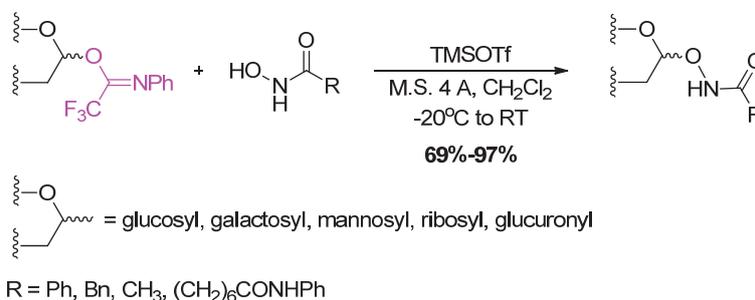
Scheme 29: Synthesis of α - and β -*O*-galactosyl hydroxylamine.¹⁰⁶

Reddy *et al.* described the Ferrier glycosylation of hydroxylamine derivatives promoted by various Lewis acids for the preparation of aminoxy pseudoglycols.¹⁰⁷ For example, the glycosylation of PhthN-OH or NHS with tri-*O*-protected D-glucal catalyzed by zinc(II) chloride gave the corresponding aminoxy pseudoglycol with high α -selectivity in good yields (Scheme 30).



Scheme 30: Synthesis of aminoxy pseudoglycol.¹⁰⁷

The first efficient *O*-glycosylation of hydroxamic acids to prepare glycosyl hydroxamates (i.e. glucosyl, galactosyl, mannosyl, glucuronyl, and ribosyl hydroxamates) in good yield was reported by Thoma *et al* (Scheme 31).¹⁰⁸ In this reaction, the glycosyl *N*-phenyl trifluoroacetimidates were used as glycosyl donors and TMSOTf as promoter in CH_2Cl_2 .

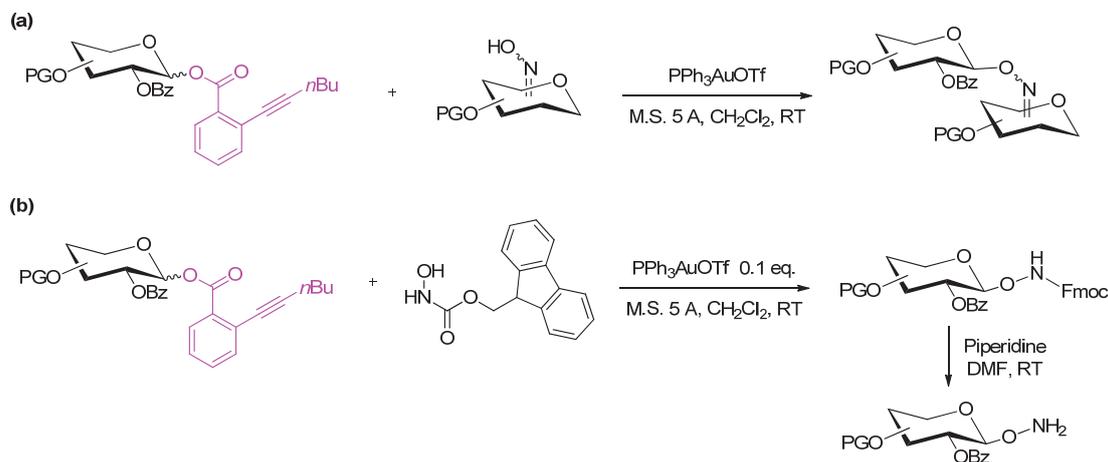


Scheme 31: *O*-glycosylation of hydroxamic acids.¹⁰⁸

¹⁰⁷ Reddy, C.; Rao, Y.; Kumar, T.; Reddy, K.; Chandrasekhar, S. *Synthesis* **2008**, 122-126.

¹⁰⁸ Thomas, M.; Gesson, J.-P.; Papot, S. *J. Org. Chem.* **2007**, *72*, 4262-4264.

Yu's group reported a direct glycosylation of sugar oximes and HONHFmoc using glycosyl *ortho*-hexynylbenzoates as glycosyl donors catalyzed by PPh_3AuOTf for the efficient preparation of N-O linked disaccharides and *O*-glycosyl hydroxylamines (Scheme 32).¹⁰⁹ Furthermore, the N-O linked saccharides could be obtained stereoselectively by reduction of the resulting oxime with $\text{NaBH}_3\text{CN}/\text{BF}_3\cdot\text{Et}_2\text{O}$.



Scheme 32: Preparation of N-O linked saccharides (a) and *O*-glycosyl hydroxylamines (b).¹⁰⁹

III.2.b Oxime-linked glycoconjugates

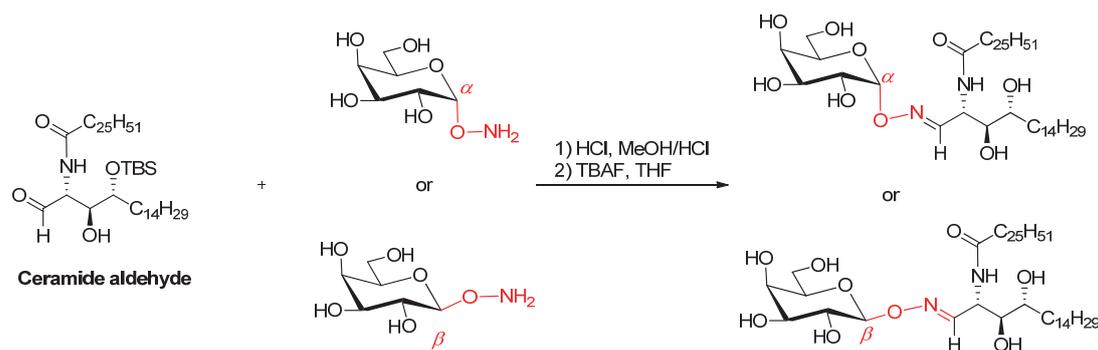
Thanks to the ready formation¹¹⁰ and the stability under physiological conditions of the oxime bond, oxime ligation constitutes an attractive route for the conjugation of bioorganic molecules and macromolecules, such as peptide dendrimers, protein-polymer conjugates, bi-functional chelating agents, glycoconjugates, etc.

III.2.b (1) Oxime-linked glycolipids

Two stereospecific oxime-linked galactosylceramide analogues were successfully synthesized by Wang and co-workers (Scheme 33).¹⁰⁶ In this synthesis, both α - and β -linkages were pre-installed, and then the glycolipids were prepared by oxime ligation of the α - and β -*O*-glycosyl hydroxylamines with ceramide aldehyde. Their bioactivity for activating iNKT cells has not been reported.

¹⁰⁹ Yu, J.; Sun, J.; Yu, B. *Org. Lett.* **2012**, *14*, 4022-4025.

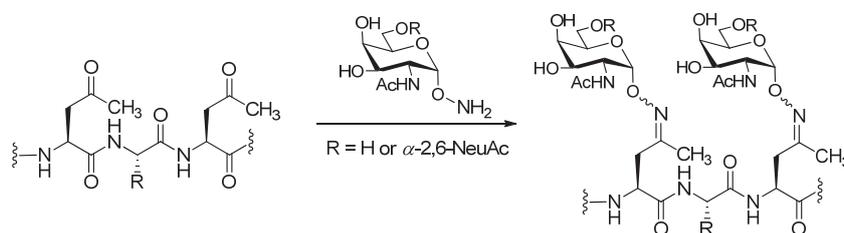
¹¹⁰ Forget, D.; Renaudet, O.; Defranq, E.; Dumy, P. *Tetrahedron Lett.* **2001**, *42*, 7829-7832.



Scheme 33: Synthesis of oxime galactosylceramide analogues.¹⁰⁶

III.2.b (2) Oxime-linked glycopeptides/glycoproteins

The oxime-based strategy provides an efficient way for the preparation of complex glycosyl amino acids for peptide synthesis, and the incorporation of glycopeptides into larger, full-length proteins. Marcaurrelle *et al* reported the synthesis of oxime-linked mucin mimics containing the tumor-related Tn and sialyl Tn antigens (Scheme 34).¹¹¹ The multiple clustered ketone residues were incorporated into a peptide firstly, and then connected to aminoxy sugars by oxime ligation.

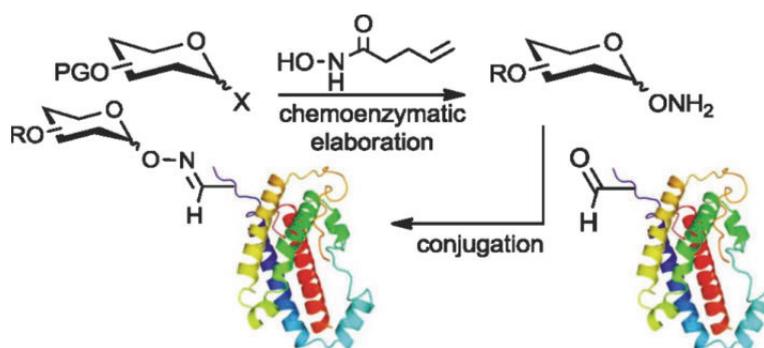


Scheme 34: Synthesis of oxime-linked mucin mimics by Marcaurrelle *et al*.¹¹¹

The oxime chemical ligation has also been used for the preparation of homogeneously glycosylated proteins, which are important targets for fundamental research and biopharmaceutical development. Hudak *et al* described the chemical ligation of aminoxy glycans to recombinantly produced proteins via oxime linkages to construct site-specifically glycosylated variants of the human growth hormone (Scheme 35).¹¹² The aldehyde tag method was used to obtain a recombinant protein with the aldehyde-bearing formylglycine residue at a specific site, and the aminoxy glycans were prepared by glycosylation with *N*-pentenoyl hydroxamate as key intermediate followed by appropriate chemoenzymatic glycosylations.

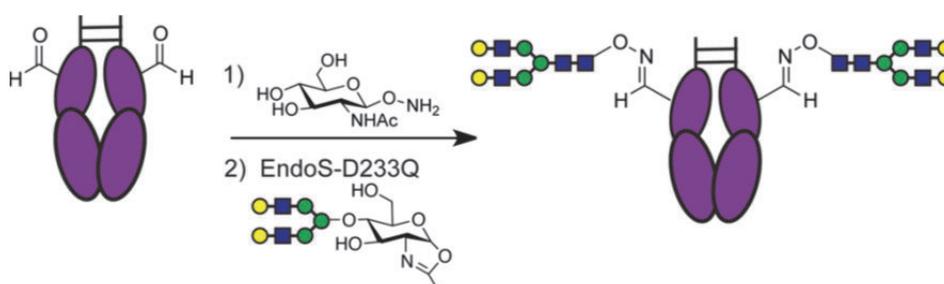
¹¹¹ Marcaurrelle, L. A.; Shin, Y.; Goon, S.; Bertozzi, C. R. *Org. Lett.* **2001**, *3*, 3691-3694.

¹¹² Hudak, J. E.; Yu, H. H.; Bertozzi, C. R. *J. Am. Chem. Soc.* **2011**, *133*, 16127-16135.



Scheme 35: Synthesis of homogeneously glycosylated proteins.¹¹²

A tailored glycosylation method of proteins by a combination of genetic, chemical, and enzymatic methods has been reported by Smith *et al* (Scheme 36).¹¹³ A genetically encoded aldehyde tag has been installed at the internal glycosylation site of the crystallizable fragment (Fc) of immunoglobulin G1 (IgG1) without perturbing protein structure. The aldehyde groups were then conjugated with aminoxy *N*-acetylglucosamine by oxime linkages to generate the modified Fc. Subsequent enzymatic transfer of complex *N*-glycans from the corresponding glycan oxazolines by an EndoS-derived glycosynthase afforded the oxime-linked glycoconjugates.

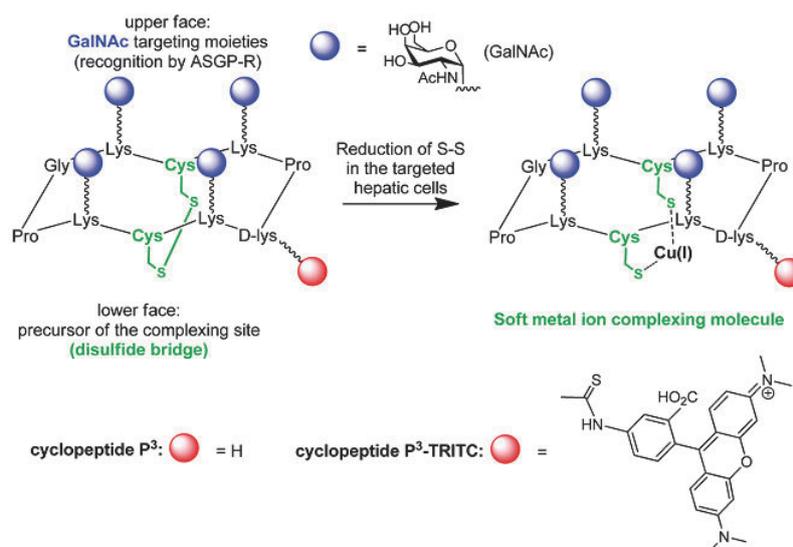


Scheme 36: Synthesis of oxime-linked glycoconjugates.¹¹³

Dumy, Renaudet and co-workers reported the preparation of a number of oxime-linked glycocyclopeptides.¹¹⁴ These neoglycocyclopeptides have been developed for various biological applications. For example, disulfide bridged compound P³ could be used as a good candidate to fight copper overload in the liver (Scheme 37),^{114a} and neoglycocyclopeptide in Figure 42 for protein-glycan interaction study.^{114b,c,g} These multivalent neoglycopeptides have also been studied as cancer related T-antigen.^{114f}

¹¹³ Smith, E. L.; Giddens, J. P.; Iavarone, A. T.; Godula, K.; Wang, L. X.; Bertozzi, C. R. *Bioconjugate Chem.* **2014**, *25*, 788-795.

¹¹⁴ (a) Pujol, A. s. M.; Cuillel, M.; Renaudet, O.; Lebrun, C.; Charbonnier, P.; Cassio, D.; Gateau, C.; Dumy, P.; Mintz, E.; Delangle, P. *J. Am. Chem. Soc.* **2011**, *133*, 286-296; (b) Bossu, I.; Sulc, M.; Krenek, K.; Dufour, E.; Garcia, J.; Berthet, N.; Dumy, P.; Kren, V.; Renaudet, O. *Org. Biomol. Chem.* **2011**, *9*, 1948-1959; (c) Andre, S.; Renaudet, O.; Bossu, I.; Dumy, P.; Gabius, H. J. *J. Pept. Sci.* **2011**, *17*, 427-437; (d) Renaudet, O.; Dumy, P. *Org. Biomol. Chem.* **2006**, *4*, 2628-2636; (e) Singh, Y.; Renaudet, O.; Defrancq, E.; Dumy, P. *Org. Lett.* **2005**, *7*, 1359-1362; (f) Renaudet, O.; Dumy, P. *Tetrahedron Lett.* **2004**, *45*, 65-68; (g) Renaudet, O.; Dumy, P. *Org. Lett.* **2003**, *5*, 243-246.



Scheme 37: General structure of Cu(I) chelators designed to target hepatocytes.^{114a}

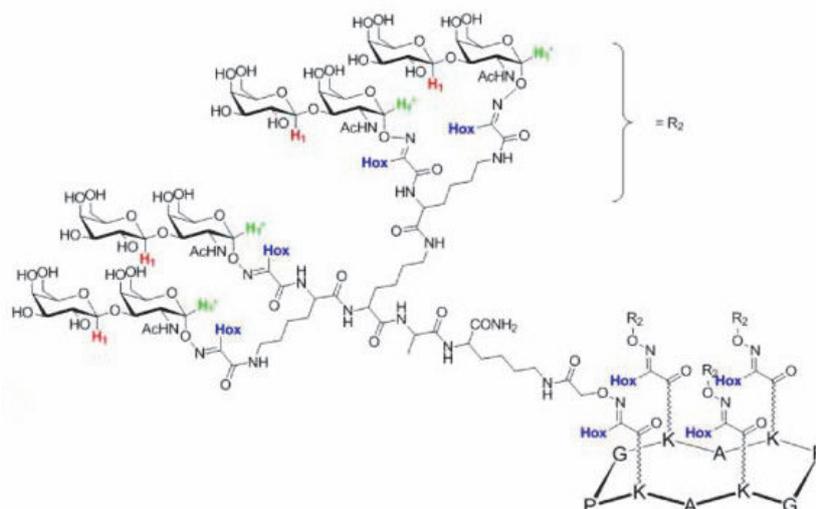


Figure 42: Oxime-linked neoglycyclopeptides reported by Dumy, Renaudet and co-workers.^{114c}

Oxime-ligation has also been employed to conjugate an oxyamine functionalized peptide with glycosyl aldehyde/ketone.

A C-glycoside analogue of the GalNAc (Tn epitope) was synthesized as a non hydrolyzable anticancer vaccine by Peri *et al.* (Figure 43).¹¹⁵ Chemoselective convergent synthetic approach was applied for the covalent conjugation of B-epitope with a T cell epitope via an oxime bond.

¹¹⁵ Peri, F.; Cipolla, L.; Rescigno, M.; La Ferla, B.; Nicotra, F. *Bioconjugate Chem.* **2001**, *12*, 325-328.

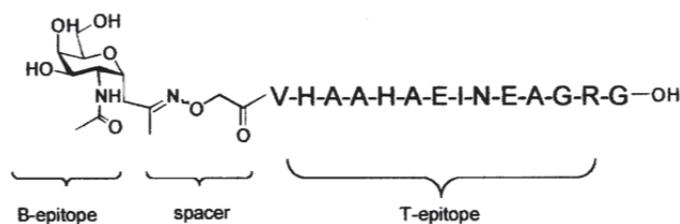


Figure 43: Chemical structure of the synthetic vaccine.¹¹⁵

Based on their previous work, Peri's group reported another three neoglycopeptides, in which the full unprotected α -C-glycosyl analogue of *N*-acetylgalactosamine was conjugated to the segment peptides ³²⁸⁻³⁴⁰OVA (Ovalbumin) and ³²⁷⁻³³⁹OVA by an oxime bond (Figure 44).¹¹⁶ Compounds **A-C** could be presented to and recognized by the T cell receptor, with the neoglycopeptide **C** containing two B-epitopes presenting higher efficiency than **B**.

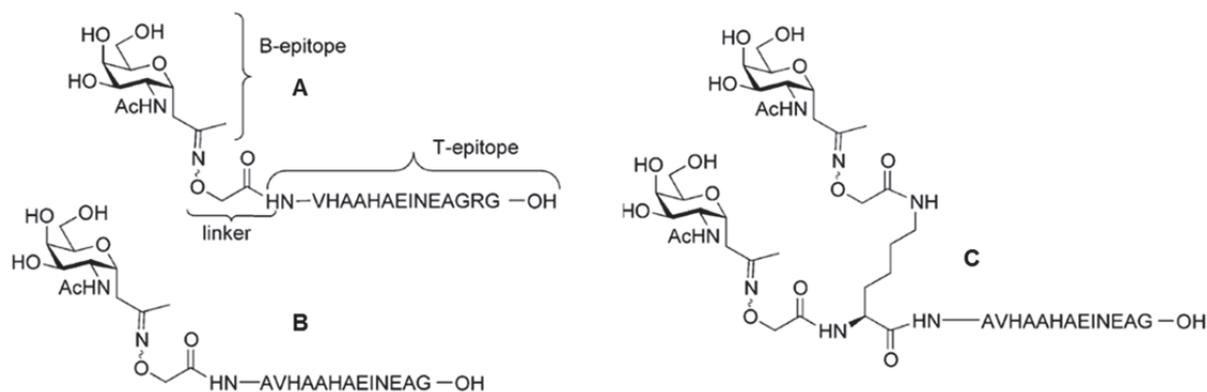


Figure 44: Chemical structure of neoglycopeptides as potential anticancer vaccines.¹¹⁶

III.3 *O*-Amino sugar derivatives

Carbohydrates and glycoconjugates bearing an aminoxy function not directly linked to the anomeric carbon have been reported as multifunctional building blocks.

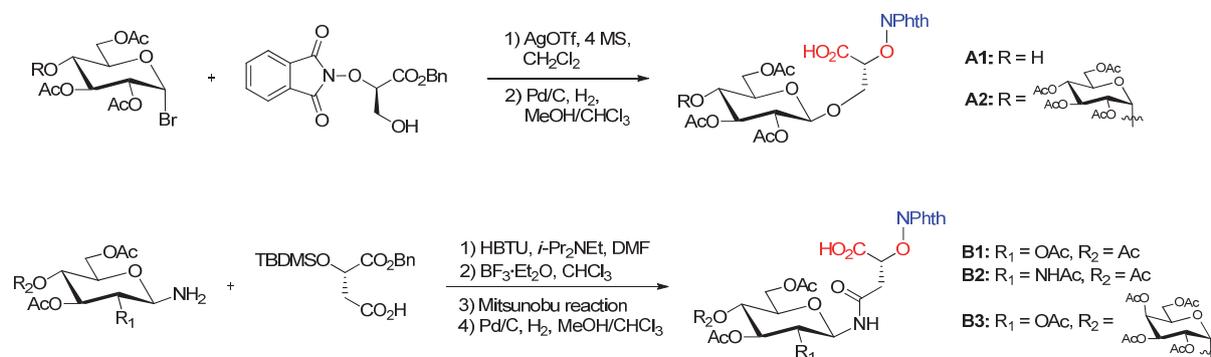
III.3.a Glycosyl aminoxy acids

Synthesis of diversely functionalized carbohydrate building blocks has attracted a great attention for the generation of new carbohydrate mimetics. Glycosyl aminoxy acids, containing an aminoxy function instead of amino one, are useful building blocks for the synthesis of novel glycopeptides and glycoproteins mimetics.

Two series of *O*- and *N*-glycosylated α -aminoxy acids have been prepared by Shin and

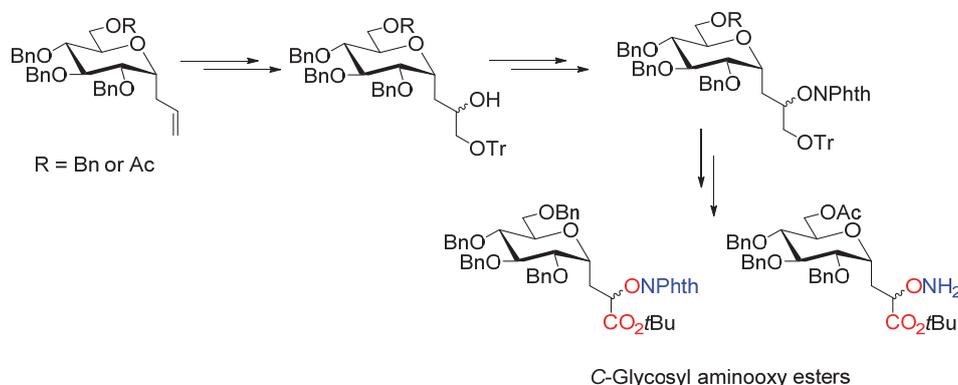
¹¹⁶ Cipolla, L.; Rescigno, M.; Leone, A.; Peri, F.; La Ferla, B.; Nicotra, F. *Bioorg. Med. Chem.* **2002**, *10*, 1639-1646.

Lee (Scheme 38).¹¹⁷ The *O*-glycosylated α -aminoxy acids were obtained by glycosylation of α -aminoxy acid derivative with glycosyl bromide, while the *N*-glycosylated one by coupling of glycosyl amine with (*S*)-malic acid derivative, followed by introduction of phthalimidoxyl function using Mitsunobu reaction.



Scheme 38: Synthesis of *N*- and *O*-glycosylated α -aminoxy acids.¹¹⁷

Recently, Peyrat *et al.* in our group have synthesized *C*-glycosyl (*R*) and (*S*)-aminoxy acid derivatives as novel glycosyl amino acid building blocks from *C*-allyl glycoside, through bishydroxylation, selective protection, Mitsunobu reaction with PhthN-OH and oxidation reactions (Scheme 39).¹¹⁸



Scheme 39: Synthesis of *C*-glycosyl aminoxy esters.¹¹⁸

III.3.b *O*-Amino sugars and glycoaminoxy acids

The first *O*-phthalimido sugar derivatives were synthesized by Grochowski *et al.* in 1976, by using Mitsunobu reaction (Figure 45).¹¹⁹ Subsequent hydrazinolysis led to the corresponding *O*-amino sugars which were then used to prepare various oxime derivatives or reduced as *O*-glycosyl hydroxylamines.¹²⁰ 5-*O*-Amino-D-psicofuranose has been prepared as a

¹¹⁷ Shin, I.; Lee, J. *Synlett* **2000**, 1297-1299.

¹¹⁸ Peyrat, S.; Cheng, K.; Xie, J. *Synthesis* **2013**, 45, 2737-2744.

¹¹⁹ Grochowski, E.; Jurczak, J. *Carbohydr. Res.* **1976**, 50, C15-C16.

¹²⁰ Tronchet, J. M.; Zosimo-Landolfo, G.; Galland-Barrera, G.; Dolatshahi, N. *Carbohydr. Res.* **1990**, 204, 145-156.

synthetic precursor for the synthesis of (+)-hydantocidin (Scheme 40).¹²¹

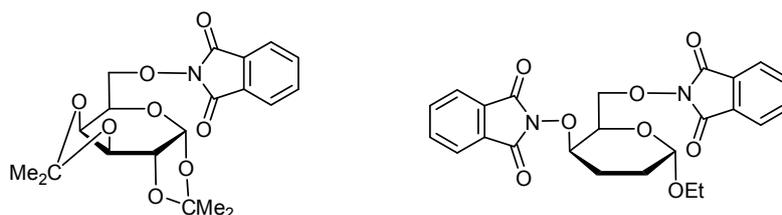
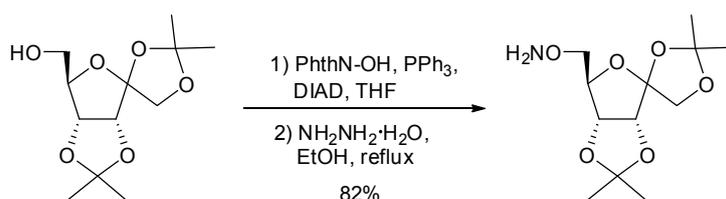
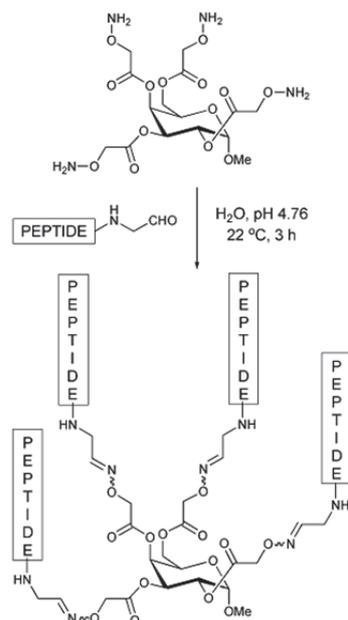


Figure 45: Structure of the first *O*-phthalimido sugar derivatives.¹¹⁹



Scheme 40: Synthesis of 5-*O*-amino-D-psicofuranose.¹²¹

Jensen and coll. have developed aminoxy-functionalized D-galactose template for chemoselective ligation with aldehyde-functionalized peptides (Scheme 41).¹²² The glycoproteins carrying imidazolyl and pyridyl moieties have been investigated for metal ion-binding studies and for kinetic investigation of esterase activity.¹²³



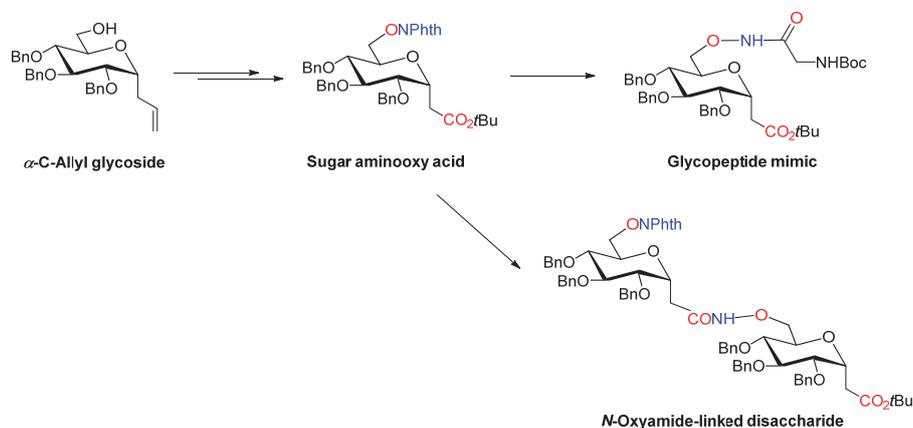
Scheme 41: Chemoselective ligation of aminoxy-functionalized D-galactose template with aldehyde-functionalized peptides.¹²²

¹²¹ Chemla, P. *Tetrahedron Lett.* **1993**, *34*, 7391-7394.

¹²² (a) Brask, J.; Jensen, K. J. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 697-700; (b) Brask, J.; Jensen, K. J. *J. Pept. Sci.* **2000**, *6*, 290-299.

¹²³ Pernille Tofteng, A.; Hansen, T. H.; Brask, J.; Nielsen, J.; Thulstrup, P. W.; Jensen, K. J. *Org. Biomol. Chem.* **2007**, *5*, 2225-2233.

Glycoaminoxy acids, with both aminoxy and carboxyl groups on the sugar frame were first reported by our group. From the readily available α -C-allyl glycoside, Malapelle *et al.* have prepared the glucopyranoid aminoxy acid as a new type of sugar building block.¹²⁴ This glycoaminoxy acid has been successfully converted into an *N*-oxyamide-linked disaccharide and glycopeptide mimetic (Scheme 42).



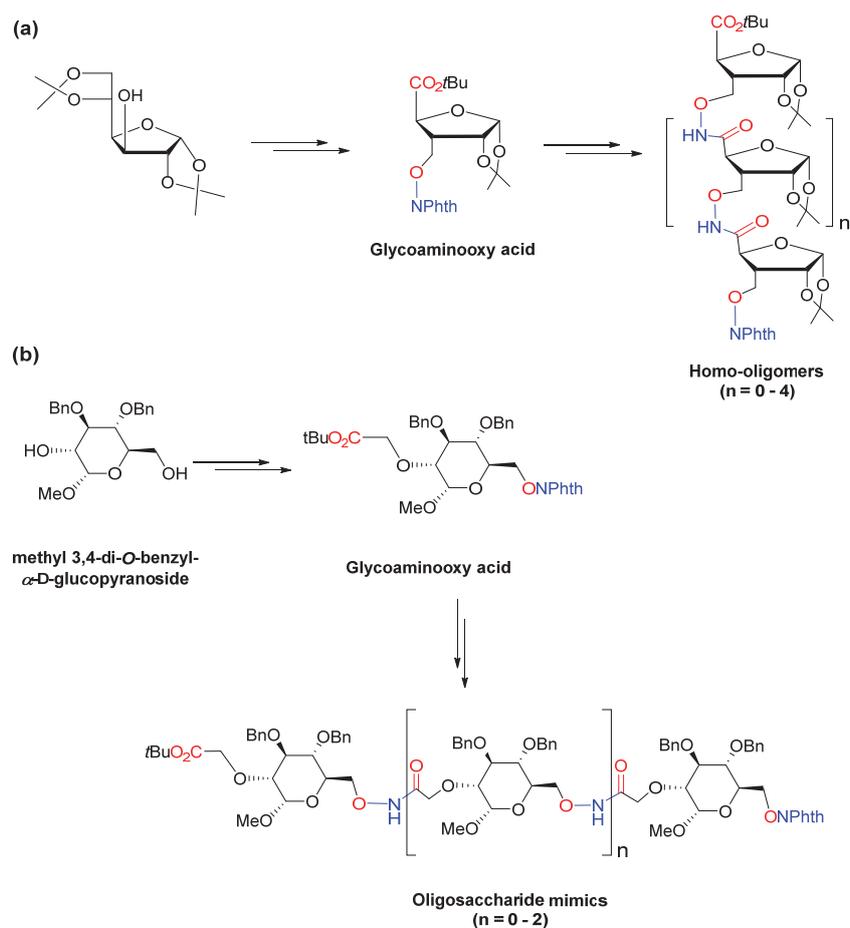
Scheme 42: Synthesis of pyranoid glycoaminoxy acid derivatives.¹²⁴

As a continuing research program in our group, Gong *et al.* synthesized D-ribofuranoid glycoaminoxy acid derivatives from 1,2:5,6-di-*O*-isopropylidene-D-glucose by introducing aminoxy and carboxylic acid functions onto the 3- and 5-positions of the furanose ring.¹²⁵ The obtained glycoaminoxy acid has been successfully used as a building block to synthesize homo-oligomers (dimer to hexamer) as a novel type of oligosaccharide mimetics (Scheme 43a). Furthermore, Song *et al.* also synthesized a pyranoid glycoaminoxy acid with carboxylic and aminoxy functions onto its 2,6-positions from methyl 3,4-di-*O*-benzyl- α -D-glucopyranoside.¹²⁶ The obtained glycoaminoxy acid building unit was successfully used to construct the *N*-oxy amide-linked oligosaccharide mimetics via *N*-acylation (Scheme 43b).

¹²⁴ Malapelle, A.; Ramozzi, R.; Xie, J. *Synthesis* **2009**, 888-890.

¹²⁵ Gong, Y.; Sun, H.; Xie, J. *Eur. J. Org. Chem.* **2009**, 6027-6033.

¹²⁶ Song, Z.; He, X.-P.; Chen, G.-R.; Xie, J. *Synthesis* **2011**, 2761-2766.



Scheme 43: Synthesis of glycoaminoxy acids and their oligomers.^{125,126}

V Conclusion

Aminoxy acids have emerged as very attractive scaffolds since the discovery of turns and helices structures in peptides containing aminoxy acids through intramolecular hydrogen bonds formation. Improved hydrolytic and metabolic stability have also been demonstrated in *N*-oxyamide-containing compounds. Furthermore, thanks to its high nucleophilicity, oxyamine can react readily with carbonyl and carboxylic compounds. These interesting properties make oxyamine attractive for the modification of biomolecules.

Carbohydrates bearing oxyamine group can be found in *N*- or *O*-glycosyl hydroxylamine and *O*-amino sugar derivatives. Neoglycosylation and oxime ligation have been proven very efficient for the preparation of various N-O linked glycoconjugates, with interesting biological activities. Glycosyl aminoxy acids and glycoaminoxy acids have been developed as multifunctional building blocks for the construction of novel glycoconjugate mimics. For the PhD work, we were interested in synthesizing orthogonally protected glyco(syl)aminoxy acids for the development of *N*-oxyamide-linked glycolipids and glycopeptides. Three main

parts were included in this thesis. In the Chapter 3, we will focus on the preparation of glycoaminoxy acids and the *N*-oxyamide-linked glycolipids with lipid chains attached on the sugar frame (Figure 46).

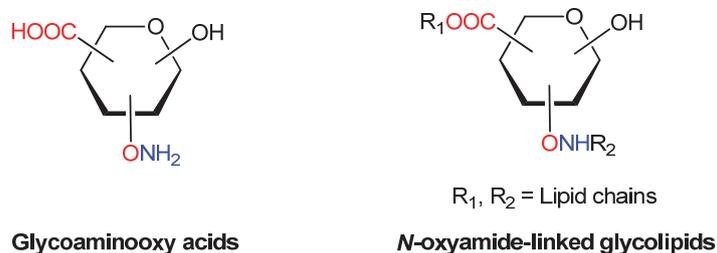


Figure 46: General structure of glycoaminoxy acids and *N*-oxyamide-linked glycolipids.

In the Chapter 4, a series of the *N*-oxyamide-linked β -*O*-glycosyl glycerolipids were synthesized as novel mimics of glyco glycerolipids and glycosphingolipids (Figure 47). The preparation of glycosyl aminoxy acid and *N*-oxyamide-linked glycopeptides was described in Chapter 5 (Figure 47).

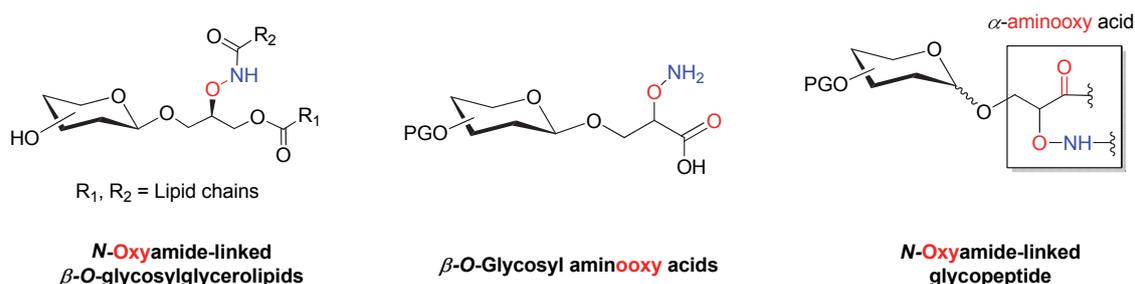
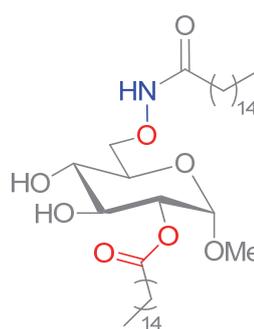
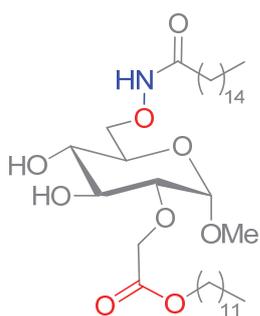
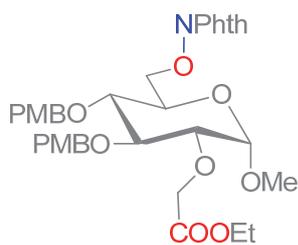


Figure 47: General structure of *N*-oxyamide-linked glyco glycerolipids, glycosyl aminoxy acids and *N*-oxyamide-linked glycopeptides.

Chapter 3

Synthesis of Glycoaminoxy Acid and *N*-Oxyamide-linked Glycolipids



I Introduction

Carbohydrates exist in all the living systems and play key roles in various biological and physiological processes. The rapidly growing importance of carbohydrate-based compounds in biological systems and their potential applications in the biopharmaceutical and medical fields, have attracted intensive research interest in the design and synthesis of new carbohydrate derivatives.

Sugar or glyco-aminoxy acids are sugar amino acid analogues with both a carboxylic acid and aminoxy function connected to the sugar scaffold (Figure 48). Due to the easily hydrogen bond formation, and the resistance to chemical and enzymatic hydrolysis of the *N*-oxyamide bond (Chapter 2), this kind of compounds have been developed as multifunctional building blocks for the construction of novel glycoconjugate mimics through *N*-oxyamide or oxime bond, providing a potent tool for the design of novel functional carbohydrate mimetics.

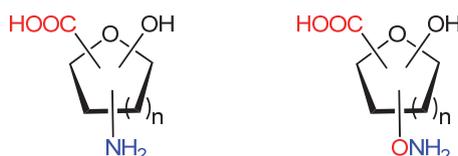


Figure 48: Structure of sugar amino acids and glycoaminoxy acids.

In the previous work of our group, Song *et al.* have synthesized a pyranoid glycoaminoxy acid with carboxylic and aminoxy functions onto its 2,6-positions (Figure 49) which was successfully used to construct the *N*-oxyamide-linked oligosaccharide mimetics via *N*-acylation (Chapter 2. III.3.b).¹²⁶

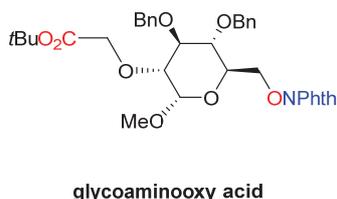


Figure 49: Chemical structure of the developed glycoaminoxy acid.¹²⁶

In this work, the benzyl ether was chosen as a protecting group because it is robust and stable to a wide range of acidic and basic conditions. However, the removal of the benzyl ether usually requires radical conditions (catalytic hydrogenolysis or single-electron transfer) or strong acids, which renders it incompatible with O-N and *O*-glycoside bond. Consequently, it was impossible to remove the benzyl groups of these pyranoid glycoaminoxy acid derivatives.

The substituted benzyl ether protecting group, *p*-methoxybenzyl ether (PMB), is one of the widely used hydroxyl protecting groups (Figure 50). The electron-rich PMB ether is much less stable to acidic condition than benzyl ether and can be selectively removed by aqueous mineral acid, camphor sulfonic acid (CSA) in methanol, or trifluoroacetic acid (TFA) in CH₂Cl₂/water. Nevertheless, their prime virtue is that they can be cleaved oxidatively with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in CH₂Cl₂/water or with ceric ammonium nitrate (CAN) in acetonitrile/water, in which conditions many other protecting groups such as acetals, esters, benzyl ethers, allyl ethers and TBS ethers are stable. Therefore the PMB ether can be used in the synthesis of functionally complex molecules.¹²⁷

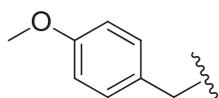
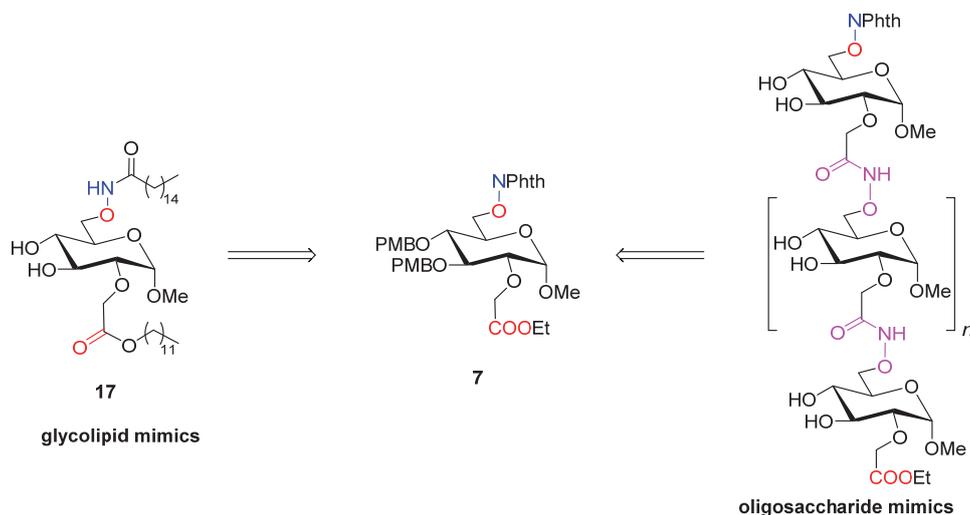


Figure 50: Structure of PMB.

We decided to synthesize the glycoaminoxy acid **7**, in which the 3,4-hydroxyl groups were protected by PMB group, and both aminoxy and carboxyl group attached on the sugar scaffold. From this multifunctional glycoaminoxy ester **7**, we decided to introduce lipid chains to prepare new glycolipid mimics for biological applications. The *N*-oxyamide linked oligosaccharide mimics could be also constructed for the conformational studies (Scheme 44). In this chapter, we will present the synthesis of glycoaminoxy acid derivatives and *N*-oxyamide-linked glycolipids.



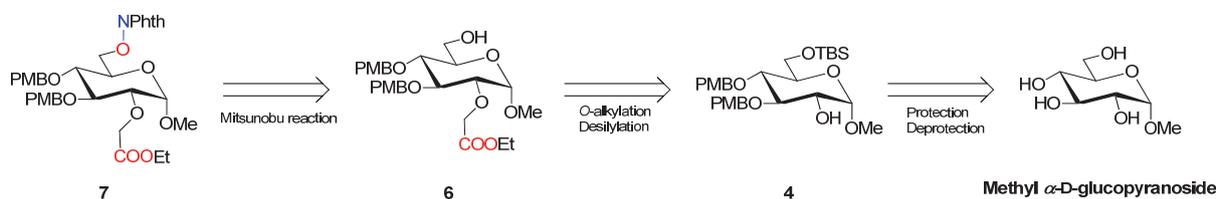
Scheme 44: Structures of the designed glycoaminoxy acid derivatives and novel glycoconjugates.

¹²⁷ Kocienski, P. J. *Protecting Groups*, 3rd edition; Thieme: Stuttgart **2005**.

II Synthesis of glycoaminoxy ester

II.1 Retrosynthesis

Our synthetic strategy for the preparation of pyranoid glycoaminoxy ester **7**, starting from the commercially availability methyl α -D-glucopyranoside, is shown in Scheme 45. Pyranoid glycoaminoxy ester **7** could be synthesized by introducing aminoxy and carboxylic functions onto the 2,6-positions of the pyranoid ring. The aminoxy function could be introduced through Mitsunobu reaction onto the 6-position of **6**, which in turn can be prepared through *O*-alkylation with ethyl bromoacetate and desilylation from the TBS ether **4**. The compound **4** could be synthesized by selective protection and deprotection from methyl α -D-glucopyranoside.



Scheme 45: Retrosynthesis of pyranoid glycoaminoxy ester **7**.

II.2 Synthesis of pyranoid glycoaminoxy ester

II.2.a Synthesis of the orthogonally protected glucopyranoside **4**

With our synthetic strategy described above, the first goal was the preparation of orthogonally protected glucopyranoside **4**. Methyl α -D-glucopyranoside was chosen as the starting material because of its commercial availability and low cost (1kg, 250 euros). The 2- and 6-hydroxy groups were firstly regioselectively protected.

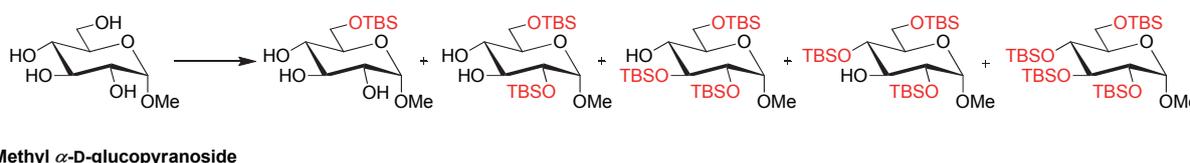
The *tert*-butyldimethylsilyl (TBS) group is useful for temporary protection of hydroxyl groups since it can be easily introduced and readily removed under mild conditions (by reaction with fluoride ion or by treatment with acid),¹²⁸ and the TBS ether is stable under a wide range of conditions. The TBS group has been used for the selective protection of the primary hydroxyl groups in diols¹²⁹, nucleosides¹³⁰, and carbohydrate derivatives. Accordingly, TBS was chosen as the protecting group to selectively protect the 2- and 6-positions of the methyl α -D-glucopyranoside.

¹²⁸ Corey, E. J.; Venkateswarlu, A. *J. Am. Chem. Soc.* **1972**, *94*, 6190-6191.

¹²⁹ Chaudhary, S. K.; Hernandez, O. *Tetrahedron Lett.* **1979**, *20*, 99-102.

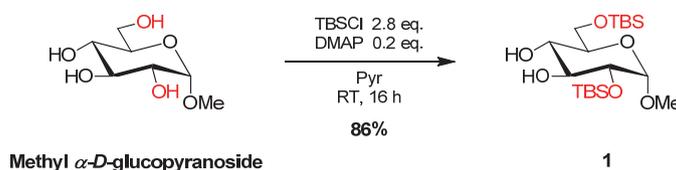
¹³⁰ Ogilvie, K. K.; Iwacha, D. J. *Tetrahedron Lett.* **1973**, *14*, 317-319.

Halmos *et al.* have reported the selective protection of methyl α/β -D-gluco/galactopyranoside, and methyl α -D-mannopyranoside, using TBSCl in the presence of imidazole, or Et₃N and DMAP in DMF.¹³¹ They found that the methyl glycosides examined showed good selectivity in their reactions towards TBSCl depending on the reaction conditions. For example, treatment of methyl α -D-glucopyranoside with 2.2 equivalents of TBSCl in the presence of imidazole (2 eq.) in DMF at room temperature for 24 h, gave 6-*O*-TBS ether (2%), 2,6- and 3,6-di-*O*-TBS ether (70% and 11%, respectively), 2,3,6- or 2,4,6-tri-*O*-TBS ether (9%). When the imidazole (2 eq.) was changed to Et₃N (2.4 eq.) and DMAP (0.08 eq.), the reaction was much less selective, giving 6-*O*-TBS ether (2%), 2,6- and 3,6-di-*O*-TBS ether (38% for each), and tri-*O*-TBS ether (12%) (Scheme 46).



Scheme 46: Various products of the silylation of methyl α -D-glucopyranoside.¹³¹

In our case, treatment of methyl α -D-glucopyranoside with 2.8 equivalents of TBSCl in the presence of 0.2 equivalent of DMAP in pyridine at room temperature for 16 h, followed by column chromatography gave the 2,6-*O*-disilylated compound **1** in 87% yield (Scheme 47).



Scheme 47: Synthesis of the 2,6-*O*-disilylated compound **1**.

Then, for the protection of 3- and 4-hydroxyl groups of compound **1**, PMB was chosen as the protecting group. When the compound **1** was treated with 6 equivalents of sodium hydride (NaH) in anhydrous DMF at 0 °C for 20 min, followed by dropwise addition of 4 equivalents of PMBCl,¹³² TLC showed the low conversion rate and the presence of several products which were difficult to separate (Table 5, Entry 1).

As the sequence of addition of reactants may affect the result of the reaction, different methods were attempted to prepare the targeted product. When NaH and PMBCl were added alternately over 40 min (following the method B) to a solution of compound **1** in dry DMF at

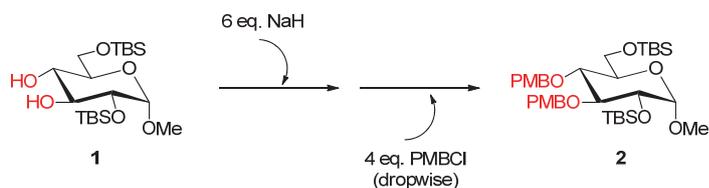
¹³¹ Halmos, T.; Montserret, R.; Filippi, J.; Antonakis, K. *Carbohydr. Res.* **1987**, *170*, 57-69.

¹³² Morales, J. C.; Zurita, D.; Penadés, S. *J. Org. Chem.* **1998**, *63*, 9212-9222.

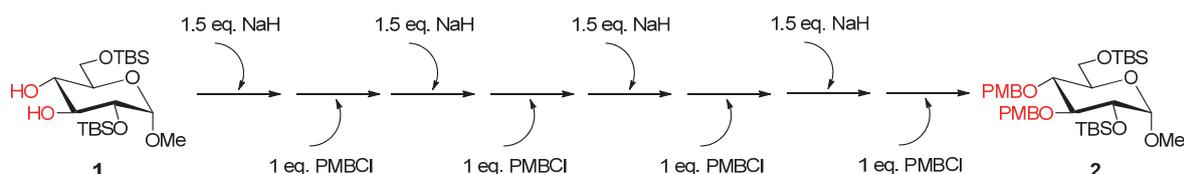
0 °C, the compound **2** was obtained in 50% yield after stirring at room temperature for 16 h (Entry 2). A better yield of 62% was obtained with the same reactants using method C, in which NaH and PMBCl were added in four portions over 40 min (NaH and PMBCl were added at the same time) (Entry 3).

Hopefully, the best yield of 80% was obtained by using method C, by adding 6 equivalents of NaH and 4 equivalents of PMBCl (1.5 equivalents of NaH and 1 equivalent of PMBCl every 20 min) to a solution of compound **1** in dry DMF under vigorous stirring at 0 °C under argon, followed by stirring at room temperature for 16 h (Entry 4).

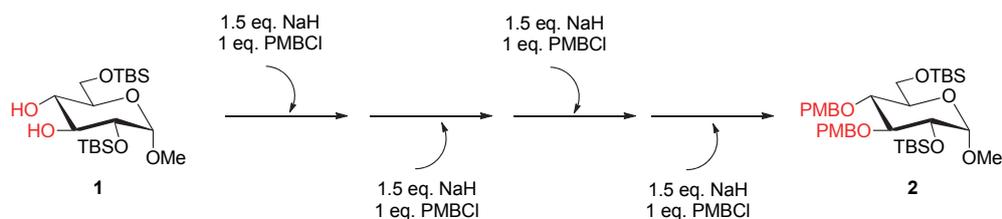
A)



B)



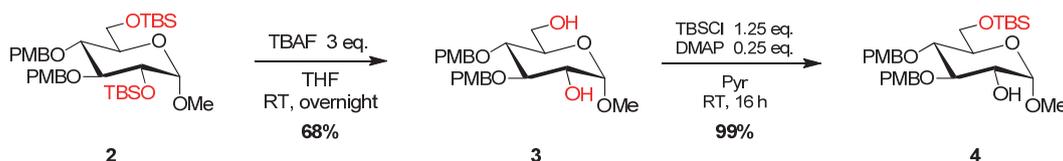
C)



Entry	Reactants Addition Sequence	Results
1	Method A	Several products
2	Method B (during 40 min)	50% yield
3	Method C (during 40 min)	62% yield
4	Method C (during 80 min)	80% yield

Table 5: Essays for the synthesis of compound **2**. General condition: NaH (6 eq.) and PMBCl (4 eq.) in DMF, 0 °C to RT, 16 h.

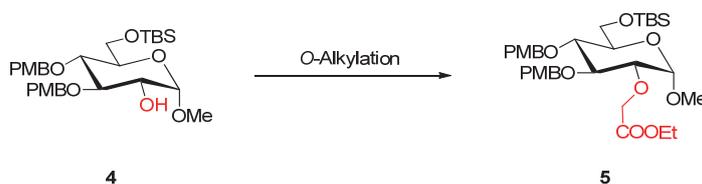
Subsequently, desilylation of compound **2** using 3 equivalents of tetra-*n*-butylammonium fluoride (TBAF) in THF at room temperature, afforded compound **3** with two free hydroxyl groups in 90 % yield (Scheme 48). In order to introduce carboxyl function onto the 2-position, the 6-hydroxyl group was reprotected by TBS. Treatment of **3** with 1.2 equivalents of TBSCl in pyridine at 0 °C in the presence of 0.25 equivalent of DMAP, afforded the TBS ether **4** in excellent yield (99%) after the resulting mixture being stirred at room temperature for 16 h (Scheme 48).



Scheme 48: Synthesis of compound **4**.

II.2.b Introduction of the carboxylic function through *O*-alkylation

O-Alkylation reaction has been successfully used for introducing the *tert*-butoxy carbonylmethyl group onto the 2-position of benzyl analogue of **4**.¹²⁶ We then tried the similar condition to introduce the ethoxycarbonylmethyl group onto the 2-position of compound **4** (Scheme 49). Compound **4** (50 mg) was treated with NaH in dry DMF (1 mL) at 0 °C, followed by addition of ethyl bromoacetate. However, as shown in Table 6, no *O*-alkylation of **4** was observed with small amount of NaH (1.1 eq. to 5 eq.) and ethyl bromoacetate (1.1 eq. to 3 eq.) (Entry 1-3). Hopefully, increasing amount of NaH (10 eq.) and ethyl bromoacetate (10 eq.) resulted in 20% conversion (Entry 4). Following the report of Tomaszewski *et al.*,¹³³ addition of catalytic imidazole (0.2 eq.) and tetrabutylammonium bromide (TBABr) (0.2 eq.) improved the conversion to around 30% (Entry 5). The use of TBAI (3 eq.) instead of imidazole and TBABr, followed by dropwise addition of ethyl bromoacetate, afforded a better conversion of 40% (Entry 6).



Scheme 49: Synthesis of **5** through *O*-alkylation.

In order to improving the conversion, the reactions of *O*-alkylation were performed under various reaction conditions, with a fixed reactants of NaH (10 eq.), BrCH₂COOEt (10 eq.) and TBAI (3 eq.). As shown in Table 6, the conversion ratio increased to 50% when the BrCH₂COOEt was added 2 h later than NaH and TBAI (Entry 7), which suggests that the deprotonation of the 2-hydroxyl group of **4** is very slow. The longer deprotonation time (5 h)

¹³³ Tomaszewski, M. J.; Holme, K. R.; Perlin, A. S. *Carbohydr. Res.* **1991**, *217*, 237-244.

led to the degradation of starting material (Entry 8). Interestingly, when the BrCH₂COOEt was added dropwise more slowly (during 30 min), the conversion increased to 64% which afforded a satisfactory corrected yield of 93% (Entry 9). These results indicated that controlling the reaction time (both deprotonation and dropping time) is crucial for this reaction.

Entry	Reactants	Conversion
1	4, NaH (1.1 eq.) stirring at 0 °C for 30 min, BrCH ₂ COOEt (1.1 eq.) was added.	0
2	4, NaH (3 eq.) stirring at 0 °C for 30 min, BrCH ₂ COOEt (3 eq.) was added.	0
3	4, NaH (5 eq.) stirring at 0 °C for 30 min, BrCH ₂ COOEt (5 eq.) was added.	0
4	4, NaH (10 eq.) stirring at 0 °C for 30 min, BrCH ₂ COOEt (10 eq.) was added.	~ 20%
5	4, NaH (10 eq.), Imidazole (0.2 eq.), TBABr (0.2 eq.) stirring at 0 °C for 30 min, Br ₂ CH ₂ COOEt (10 eq.) was added.	~ 30%
6	4, NaH (10 eq.) and TBAI (3 eq.) stirring at 0 °C for 30 min, BrCH ₂ COOEt (10 eq.) was added dropwise during 10 min.	~ 40%
7	4, NaH (10 eq.) and TBAI (3 eq.) stirring at 0 °C for 2 h, BrCH ₂ COOEt (10 eq.) was added dropwise during 10 min.	~ 50%
8	4, NaH (10 eq.) and TBAI (3 eq.) stirring at 0 °C for 5 h, BrCH ₂ COOEt (10 eq.) was added dropwise during 10 min.	Degration
9	4, NaH (10 eq.) and TBAI (3 eq.) stirring at 0 °C for 2 h, BrCH ₂ COOEt (10 eq.) was added dropwise during 30 min.	64%* (Corrected yield: 93%)
10	4, NaH (5 eq.) and TBAI (3 eq.) stirring at 0 °C for 2 h, BrCH ₂ COOEt (10 eq.) was added dropwise during 30 min.	~ 40%
11	4, NaH (2 eq.) and TBAI (3 eq.) stirring at 0 °C for 2 h, BrCH ₂ COOEt (10 eq.) was added dropwise during 30 min.	~ 30%

Table 6: Attempted conditions for *O*-alkylation of 4. All the reactions were performed with 50 mg of 4 in 1 mL DMF, from 0 °C (30 min) to room temperature (15 h). The conversion was determined by TLC except for Entry 9 (*The conversion ratio and corrected yield were calculated after column chromatography).

As described above, the most efficient conversion of 64% was observed when the compound **4** was treated with 10 equivalents of NaH and 3 equivalents of TBAI in dry DMF at 0 °C for 2 h, followed by slow, dropwise addition of 3 equivalents of ethyl bromoacetate. By investigating various amounts of reactants (2 eq., 5 eq., 10 eq.), we found that the effective conversion decreased with decreasing reactants in a fixed reaction time (Entry 9, 10, 11), which suggests that a large excess of NaH and ethyl bromoacetate is necessary for this reaction.

It is worth noting that, when this reaction was performed in small scale (less than 200 mg), TLC showed the presence of starting material and the target product (identified by NMR). However, two new spots along with the target product (44% isolated yield) appeared when this reaction was performed in larger scale (1 g), which means that part of the starting material was converted to side-products (Figure 51). The *O*-alkylation of **4** was then performed in small scale.

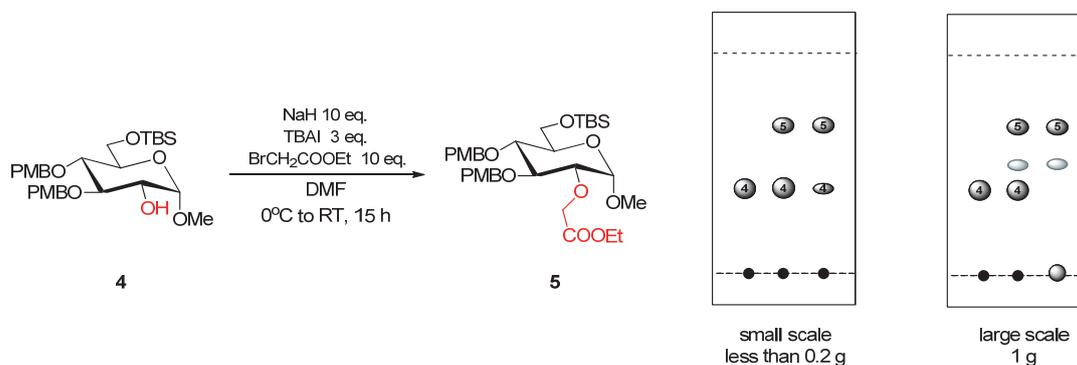
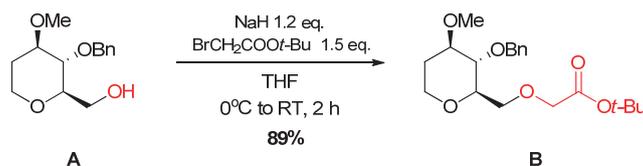


Figure 51: The TLC plates of the *O*-alkylation of **4**.

Compound **4** was also treated with BrCH₂COOEt (5 eq.) in THF under microwave heating, in the presence of NaH (5 eq.) and TBAI (3 eq.). After 45 min at 150 °C, TLC showed 20% conversion (Microwave condition: T = 150 °C; P = 2.1-2.3 bars; Stirring: 1000 rpm/min; Vol = 1mL/10 mL; t = 45 min).

We also tried the *O*-alkylation of **4**, following a reported procedure by Feher-Voelger *et al.*,¹³⁴ which showed an excellent yield of 89% for the *O*-alkylation reaction (Scheme 50). Compound **A** in THF at 0 °C was first treated with NaH (1.2 eq.) for 5 min. Then the mixture was added in portion to a solution of *tert*-butyl bromoacetate (1.5 eq.) in THF. The compound **B** was obtained in 89 % yield after 2 h stirring at room temperature. In our case, treatment of **4** with NaH (1.2 eq.), then ethyl bromoacetate (1.5 eq.) in THF led to no conversion.

¹³⁴ Feher-Voelger, A.; Borges-Gonzalez, J.; Carrillo, R.; Morales, E. Q.; Gonzalez-Platas, J.; Martin, T. *Chem. Eur. J.* **2014**, *20*, 4007-4022.



Scheme 50: *O*-Alkylation of compound **A**.¹³⁴

Compared to the easy introduction of carboxylic function to the 2-position of compound **C** in our group,¹²⁶ perhaps the incomplete conversion of **4** was due to the increased steric hindrance from the PMB groups (Figure 52).

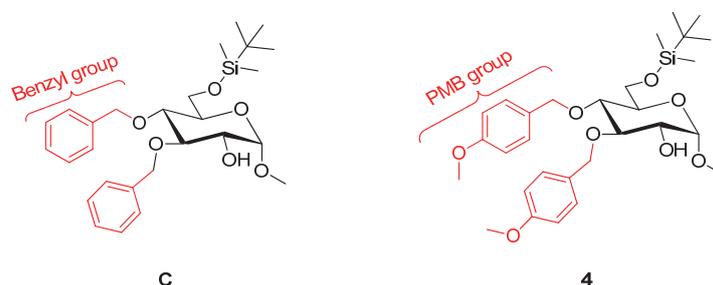
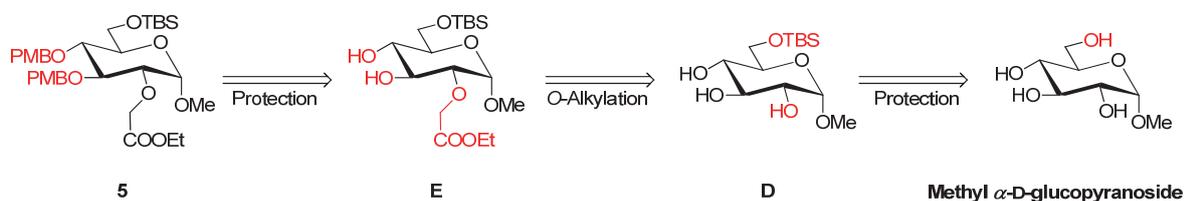


Figure 52: Structure of compounds **A** and **4**.

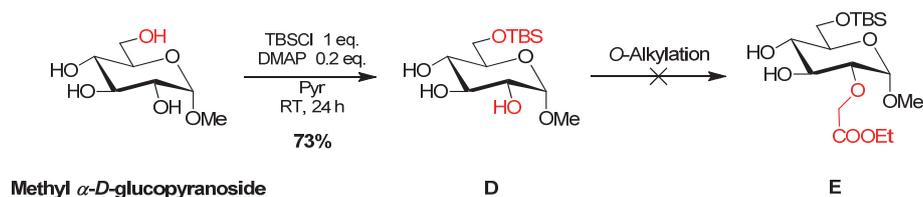
Considering that the difficulty of *O*-alkylation of **4** may be due to the presence of PMB groups, we designed a novel synthetic route for preparing compound **5** from methyl α -D-glucopyranoside (Scheme 51). The target compound **5** could be prepared by using PMBCl to protect the 3,4-positions of compound **E**, which could be synthesized by 2-*O*-alkylation of compound **D**. The TBS ether **D** can be obtained through regioselective silylation from methyl α -D-glucopyranoside.



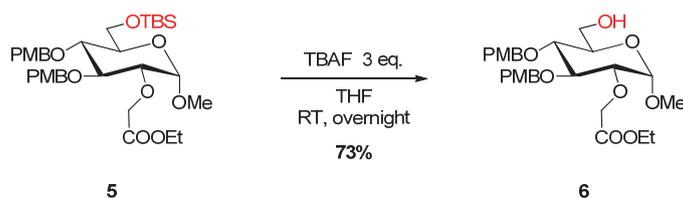
Scheme 51: Retrosynthesis of compound **5**.

The 6-*O*-silylated compound **D** was easily prepared by treatment of methyl α -D-glucopyranoside with equimolar amount of TBSCl in pyridine in the presence of 0.2 equivalent of DMAP at room temperature for 24 h (Scheme 52). Unfortunately, 2-*O*-alkylation of compound **D** using NaH (1.1 eq.) and BrCH₂COOEt (3 eq.) in DMF failed to give **E**, since this reaction gave very low conversion and a number of by-products which were difficult to be separated

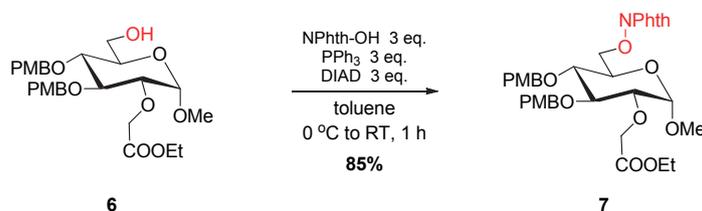
(Scheme 52).

**Scheme 52:** Attempted method for synthesizing **E**.**II.2.c Synthesis of the glycoaminoxy ester 7**

With the carboxylic acid derivative **5** in hand, the glycoaminoxy ester was prepared by introducing aminoxy function to the 6-position of the pyranose ring. Removal of the TBS group was realized by treatment of compound **5** with 3 equivalents of TBAF in THF at room temperature, affording the compound **6** in 73% yield (Scheme 53).

**Scheme 53:** Desilylation of **5** to synthesize **6**.

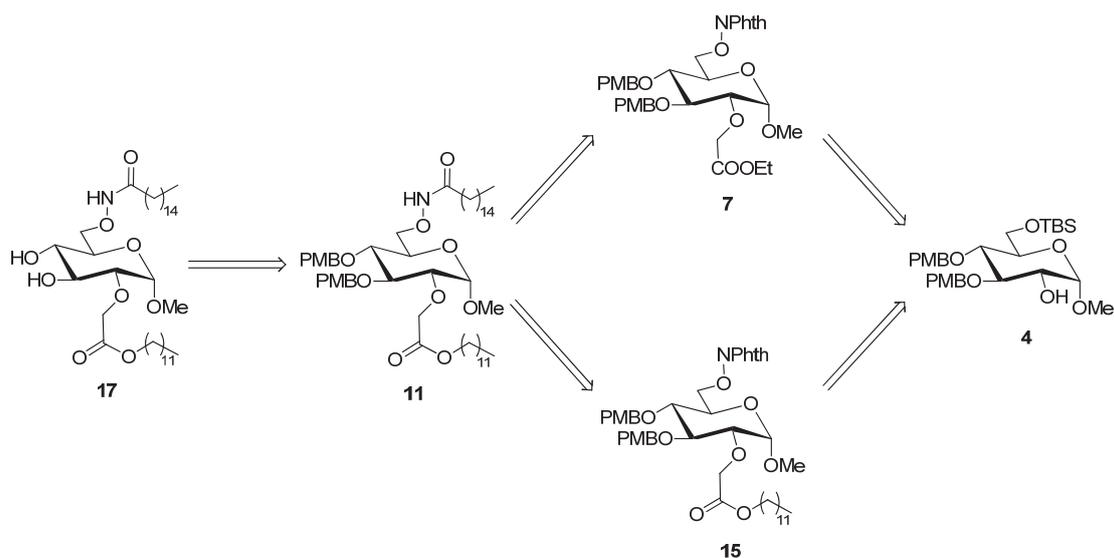
The aminoxy function was introduced to the 6-position of compound **6** via Mitsunobu reaction. Compound **6** was treated with 3 equivalents of PhthN-OH in toluene in the presence of 3 equivalents of PPh₃ at 0 °C, followed by slow dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds). The appearance of a dark red coloring means the success of this Mitsunobu reaction. The reaction finished after 1 h stirring at room temperature to give the target glycoaminoxy ester **7** in its fully protected form in 85% yield (Scheme 54).

**Scheme 54:** Synthesis of glycoaminoxy ester **7** via Mitsunobu reaction.

III Synthesis of *N*-oxyamide-linked glycolipid 17

III.1 Retrosynthesis

The target *N*-oxyamide-linked glycolipid **17** could be synthesized by deprotecting the PMB group of glycolipid **11**, which could be obtained by introducing lipid chains to the 2- and 6-positions of glycoaminoxy ester **7**, or through the intermediate **15** by introducing directly the first lipid chain to the 2-position of compound **4** (Scheme 55).

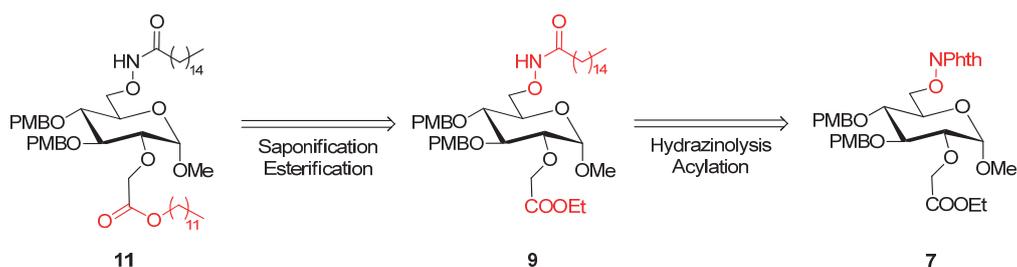


Scheme 55: Retrosynthesis of *N*-oxyamide-linked glycolipid **17**.

III.2 Synthesis of *N*-oxyamide-linked glycolipid 17

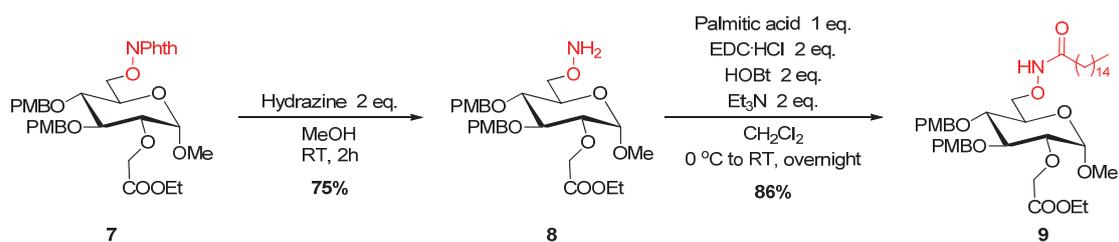
III.2.a Synthesis of the glycolipid **11** from **7**

From the obtained glycoaminoxy ester **7**, the glycolipid **11** could be obtained by introducing acyl chains onto the 2- and 6-positions. Our synthetic strategy is outlined in Scheme 56. The target product could be synthesized by saponification and esterification from compound **9**, which could be obtained by hydrazinolysis of **7** followed by acylation.



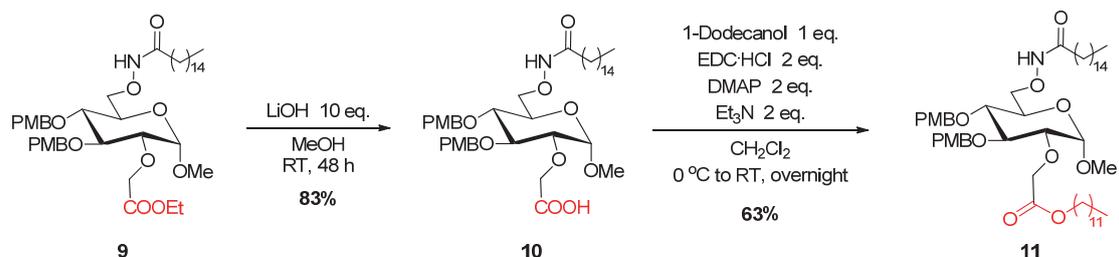
Scheme 56: Retrosynthesis of glycolipid **11** from **7**.

The phthaloyl group of **7** was first removed by hydrazinolysis. Compound **7** was treated with 2 equivalents of hydrazine in MeOH at room temperature for 2 h to give the oxyamine derivative **8** in 75% yield (Scheme 57). Coupling of the obtained oxyamine **8** with equimolar quantity of palmitic acid in CH₂Cl₂ in the presence of 2 equivalents each of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC·HCl), hydroxybenzotriazole (HOBt) and Et₃N at room temperature, afforded the glucolipid **9** in 86% yield (Scheme 57). The proton of the *N*-oxyamide bond appeared at 8.04 ppm in its ¹H NMR spectrum in CDCl₃ solution.



Scheme 57: Synthesis of compound **9**.

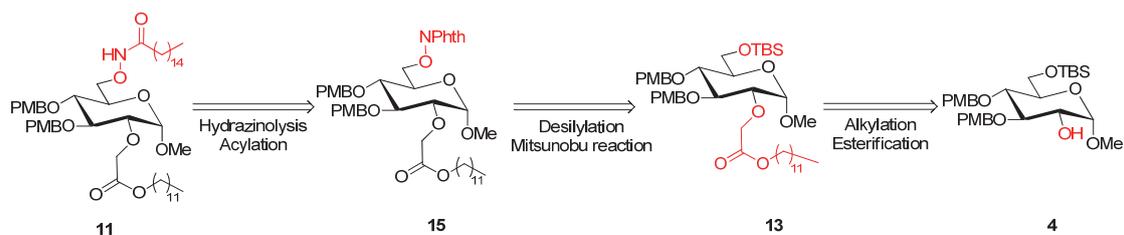
Saponification of compound **9** with 10 equivalents of LiOH at room temperature for 48 h afforded the carboxylic acid derivative **10** in 83% yield (Scheme 58). Subsequently, the glycolipid **11** was obtained by esterification. Coupling of the obtained carboxylic acid derivative **10** with equimolar quantity of 1-dodecanol in CH₂Cl₂ in the presence of EDC·HCl/DMAP/Et₃N, afforded **11** in 63% yield (Scheme 58).



Scheme 58: Synthesis of compound **11**.

III.2.b Synthesis of glycolipid **11** from **4**

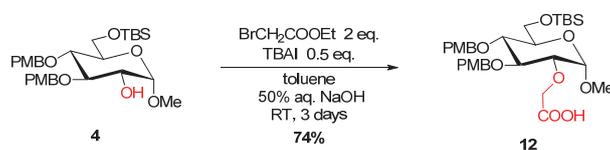
Another synthetic strategy for the preparation of glycolipid **11**, starting from the compound **4**, is shown in Scheme 59. Compound **11** could be synthesized by deprotection and acylation of the *O*-phthalimido derivative **15**, which could be prepared from **13**, an esterified product of compound **4**.



Scheme 59: Retrosynthesis of glycolipid **11** from **4**.

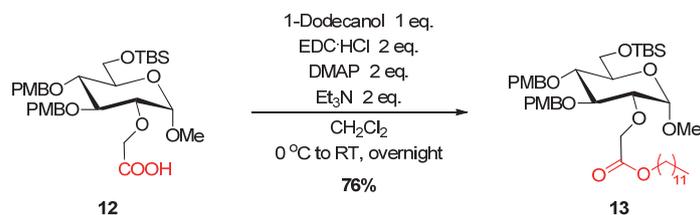
The carboxyl function was first introduced onto the 2-position through *O*-alkylation with ethyl bromoacetate under phase transfer conditions.¹³⁵ Compound **4** was treated with 0.5 equivalent of TBAI in a mixture of 50% aqueous NaOH and toluene (1/1, V/V), followed by dropwise addition of 2 equivalents of ethyl bromoacetate. After the resulting mixture was stirred at room temperature for 48 h, neutralized with acid and purified by column chromatography, a pure product was obtained in 74% corrected yield (conversion: 67%).

Based on the HRMS and NMR spectra (¹H NMR, ¹³C NMR, Dept135, COSY, HMQC), the obtained molecule was identified to be the compound **12** (Scheme 60). This result suggested that the saponification was performed simultaneously in this basic condition, which removed the ethyl group to afford the carboxylic acid derivative **12**.



Scheme 60: Synthesis of compound **12**.

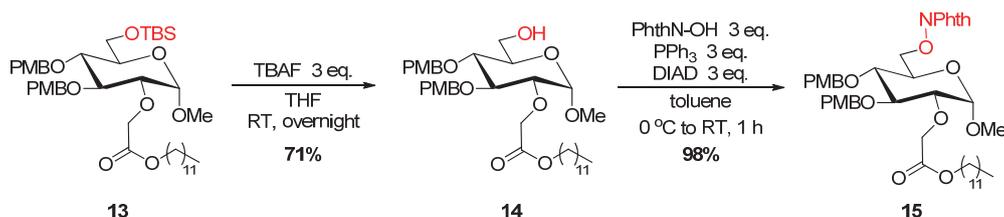
The glycolipid **13** was then prepared by esterification. Coupling of the obtained carboxylic acid derivative **12** with equimolar quantity of 1-dodecanol in CH₂Cl₂ in the presence of EDC·HCl/DMAP/Et₃N, afforded the glycolipid **13** in 76% yield (Scheme 61).



Scheme 61: Esterification of **12** to synthesize compound **13**.

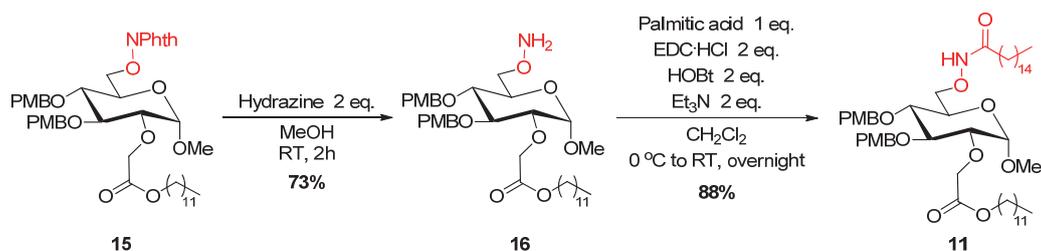
¹³⁵ a) Salman, A. A.; Tabandeh, M.; Heidelberg, T.; Hussien, R. S. D. *Carbohydr. Res.* **2015**, *406*, 41-45; b) Karst, N.; Jacquinet, J.-C. *J. Chem. Soc., Perkin Trans I*, **2000**, 2709-2717.

In order to introduce the aminoxy function to the 6-position of the sugar template, the TBS group of compound **13** was removed by 3 equivalents of TBAF in THF at room temperature to give compound **14** in 71% yield (Scheme 62). Subsequently, the aminoxy function was introduced through Mitsunobu reaction. Compound **14** was reacted with PhthN-OH (3 eq.) in toluene in the presence of PPh₃ (3 eq.) at 0 °C, followed by slow dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds). The appearance of a dark red coloring means the success of this Mitsunobu reaction. The reaction finished after 1 h stirring at room temperature to give the *O*-phthalimido derivative **15** in excellent yield of 98% (Scheme 62).



Scheme 62: Synthesis of *O*-phthalimido derivative **15**.

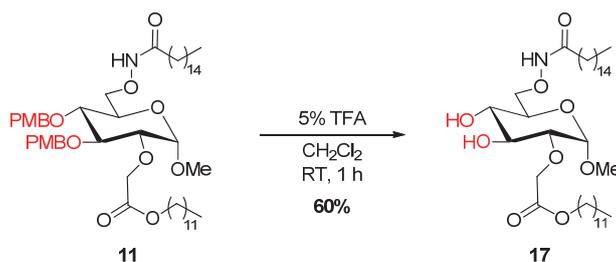
Finally, hydrazinolysis of the *O*-phthalimido derivative **15** with 2 equivalents of hydrazine in MeOH at room temperature for 2 h, afforded the oxyamine **16** in 73% yield (Scheme 63). Coupling of the obtained oxyamine **16** with equimolar quantity of palmitic acid in CH₂Cl₂ in the presence of EDC·HCl/DMAP/Et₃N at room temperature, afforded the glycolipid **11** in 88% yield (Scheme 63). The proton of the *N*-oxyamide bond appeared at 8.01 ppm in its ¹H NMR spectrum in CDCl₃ solution.



Scheme 63: Synthesis of glucolipid **11**.

III.2.c Deprotection of **11**

The deprotection of PMB ethers was carried out using 5% TFA in CH₂Cl₂ at room temperature for 1 h. The target glycolipid **17** was obtained in 60% yield after purification by column chromatography (Scheme 64). The signal of NH in the *N*-oxyamide bond of **17** appeared at 8.74 ppm in its ¹H NMR spectrum in CDCl₃ solution. All structures of the synthetic compounds have been fully characterized by ¹H, ¹³C NMR and HRMS.

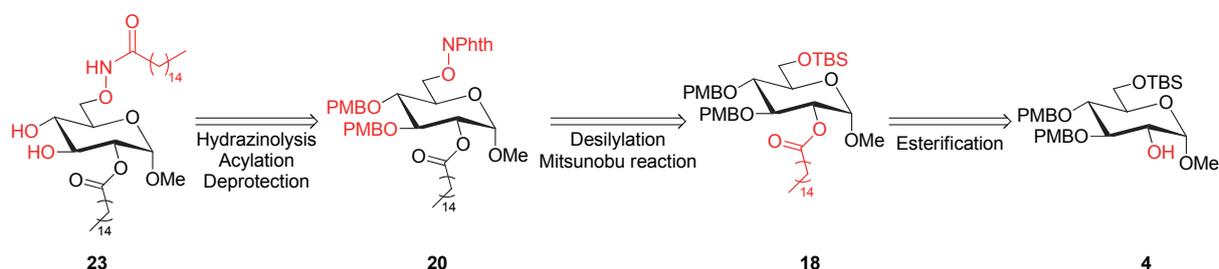


Scheme 64: Deprotection of compound **11** to synthesize glycolipid **17**.

IV Synthesis of glycolipid **23**

IV.1 Retrosynthesis

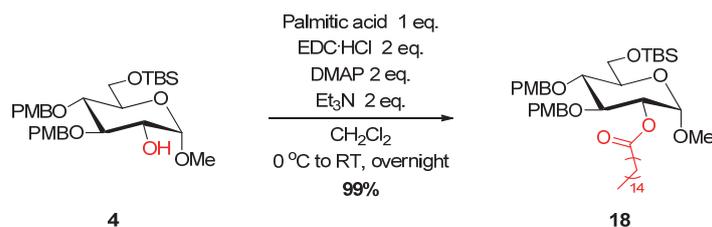
The retrosynthetic analysis for the preparation of glycolipid **23** is depicted in Scheme 65. We chose to prepare **23** from the *O*-phthalimido derivative **20**, on which the acyl chain could be readily ligated at the 6-position through hydrazinolysis and acylation reactions. Compound **20** is easily accessible from **18** which could be prepared by esterification of palmitic acid with **4** bearing a free hydroxyl group at the 2-position on the sugar template.



Scheme 65: Retrosynthesis of glycolipid **23**.

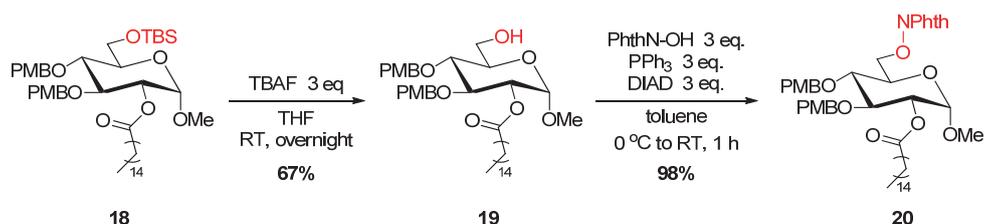
IV.2 Synthesis of glycolipid **23**

The initial effort was to synthesize the compound **18** as shown in Scheme 66. Introduction of the first acyl chain on the 2-position of **4** by condensation with equimolar quantity of palmitic acid in CH_2Cl_2 in the presence of EDC·HCl/DMAP/ Et_3N at room temperature, produced the glucolipid **18** in excellent yield of 99%.



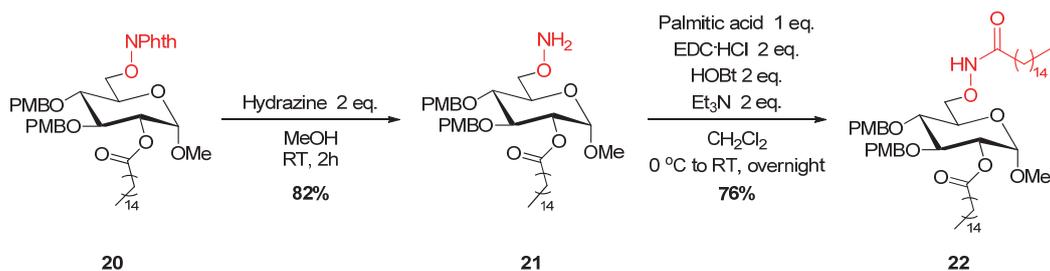
Scheme 66: Esterification of **4** to obtain the glucolipid **18**.

Removal of the TBS group of compound **18** using 3 equivalents of TBAF in THF at room temperature, gave the compound **19** containing a free 6-hydroxyl group in 67% yield (Scheme 67). Then the aminoxyyl function was introduced to the 6-position via Mitsunobu reaction. When compound **19**, PhthN-OH (3 eq.) and PPh₃ (3 eq.) were dissolved in toluene at 0 °C, followed by dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds) with the appearance of a dark red coloring, the *O*-phthalimido derivative **20** was produced in 98% yield after 1 h stirring at room temperature (Scheme 67).



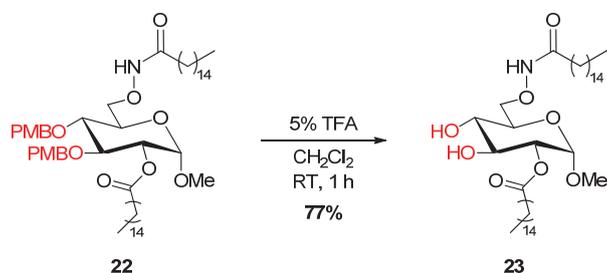
Scheme 67: Synthesis of *O*-phthalimido derivative **20**.

Subsequently, the phthaloyl group of compound **20** was removed by hydrazinolysis using 2 equivalents of hydrazine in MeOH, affording the oxyamine **21** in 82% yield after 2 h stirring at room temperature (Scheme 68). Introduction of the second acyl chain on the oxyamine of compound **21** by condensation with equimolar quantity of palmitic acid in CH₂Cl₂ in the presence of EDC·HCl/DMAP/Et₃N, produced the glucolipid **22** in 76% yield (Scheme 68). The ¹H NMR spectrum of **22** in CDCl₃ solution showed the NH signal appearing at 8.03 ppm, indicating the formation of *N*-oxyamide bond.



Scheme 68: Synthesis of glucolipid **22**.

Finally, removal of the PMB group using 5% TFA in CH_2Cl_2 at room temperature for 1 h, generated the target compound **23** in 77 % yield after purification by column chromatography (Scheme 69). The signal of NH in the *N*-oxyamide bond of **23** appeared at 8.55 ppm in its ^1H NMR spectrum in CDCl_3 solution.



Scheme 69: Deprotection of compound **22** to synthesize glycolipid **23**.

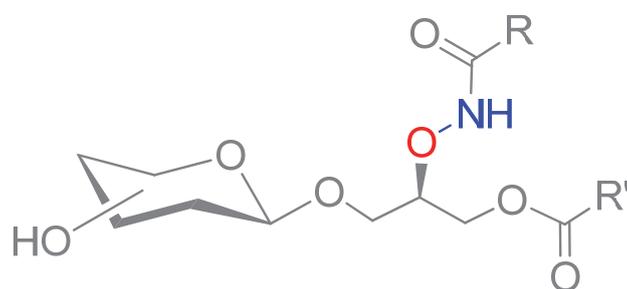
V Conclusion

From the commercially available methyl α -D-glucopyranoside, we have successfully synthesized glycoaminoxy ester **7**, in which the carboxylic acid function was introduced onto the 2-position through *O*-alkylation and the aminoxy function was introduced onto the 6-position via Mitsunobu reaction (Scheme 70). It is worth mentioning that the 3- and 4-hydroxyl groups were protected by PMB ether, which can be readily removed without affecting other functional groups.

The orthogonally protected glycoaminoxy ester **7** constitutes a useful multifunctional building block for further derivatization. We have succeeded in the use of glycoaminoxy ester **7** for the efficient synthesis of new glycolipid **17**, in which two lipid chains were attached on the 2- and 6-positions of pyranose ring through *N*-oxyamide and ester bond. This glycolipid **17** can be also prepared by introducing two lipid chains to the intermediate compound **4**, from which another type of glycolipid **23** has been easily prepared (Scheme 70). We have also tried to prepare the *N*-oxyamide-linked oligosaccharide mimics. However, the corresponding carboxylic acid of **7** needed for coupling with the oxyamine derivative **8** was not obtained due to the ring opening side reaction of the phthaloyl group during the saponification.¹²⁵ Consequently, a carboxyl protecting group compatible with phthaloyl group should be introduced to accomplish the synthesis of *N*-oxyamide-linked oligosaccharides.

Chapter 4

Synthesis of *N*-Oxyamide-linked Glycoglycerolipids



I Introduction

As described in Chapter 1.III, glycolipids are composed of one or several monosaccharide residues bound by a glycosidic linkage to a hydrophobic moiety like an acylglycerol (termed glycoglycerolipids, GGLs) or a ceramide (glycosphingolipids, GSLs). As part of the glycoconjugate family, they play central roles in a variety of important biological processes, such as cell-cell interactions, viral and bacterial infections, immune response, signal transduction, cell proliferation, etc. The design of glycolipid mimics has become a useful strategy in drug discovery.

The synthetic glycolipids have shown interesting biological activities. Modifications have been made on the sugar part, on the configuration and nature of the anomeric bond (*C*-, *S*-glycoside), on the polar moiety of the ceramide or glycerol, or on the lipid chains. Reports showed that these minor structural changes affected biological activities of glycolipids. The ability to undergo extensive interlipid hydrogen bonding has been considered to be fundamental for the functions of glycolipids in membranes, through imparting structural integrity to the membranes of the organisms.³⁶

The *N*-oxyamide-containing compounds have shown improved metabolic stability, and interesting secondary structures thanks to the easy H-bond formation property of *N*-oxyamide compounds (Chapter 2). With a continuing interest on *N*-oxyamide-containing biomolecules, we designed the *N*-oxyamide-linked glycoglycerolipids, by replacing the ester function in GGLs by a *N*-oxyamide (Figure 53). This kind of glycolipids, never reported in the literature, could be considered as analogues of both GGL and GSL.

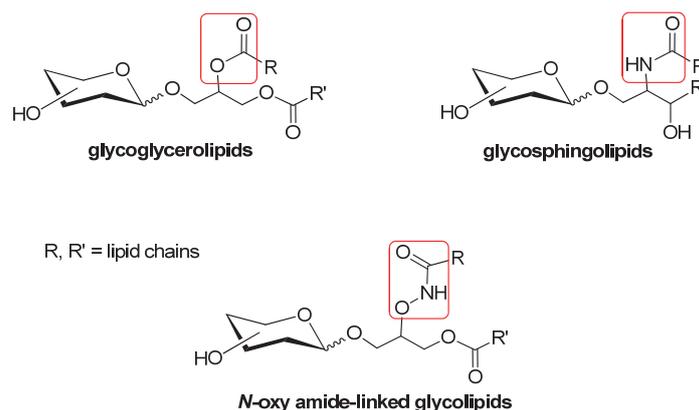


Figure 53: Structure of GGLs, GSLs and *N*-oxyamide-linked glycolipids.

II Synthesis of *N*-oxyamide-linked glucoglycerolipids

II.1 Retrosynthesis

GGLs begin with either glucose or galactose attached in α - or β -linkage to the 1-hydroxyl of mono-/di-acylglycerol or ceramide. As β -linked glucose unit is widely existing in natural glycolipids, we investigated first the synthesis of *O*-amino β -glucoglycerol and *N*-oxyamide linked glucoglycerolipids. Since the modification of lipid chains have shown very interesting results (Chapter 1), we proposed to introduce different fatty chain in the same position to synthesize a series of compounds to study their biological activities. The *N*-oxyamide linked glycoglycerolipids with just one lipid chain were also targeted (Figure 54).

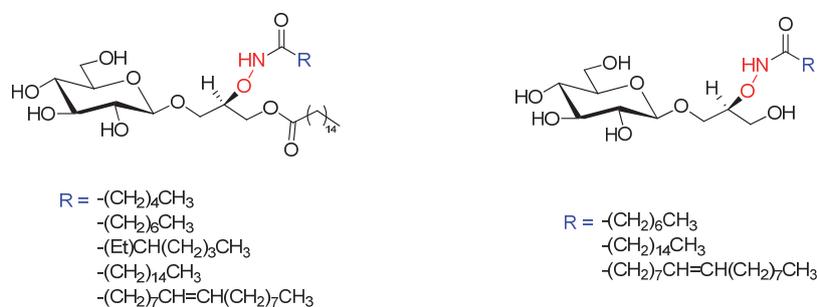


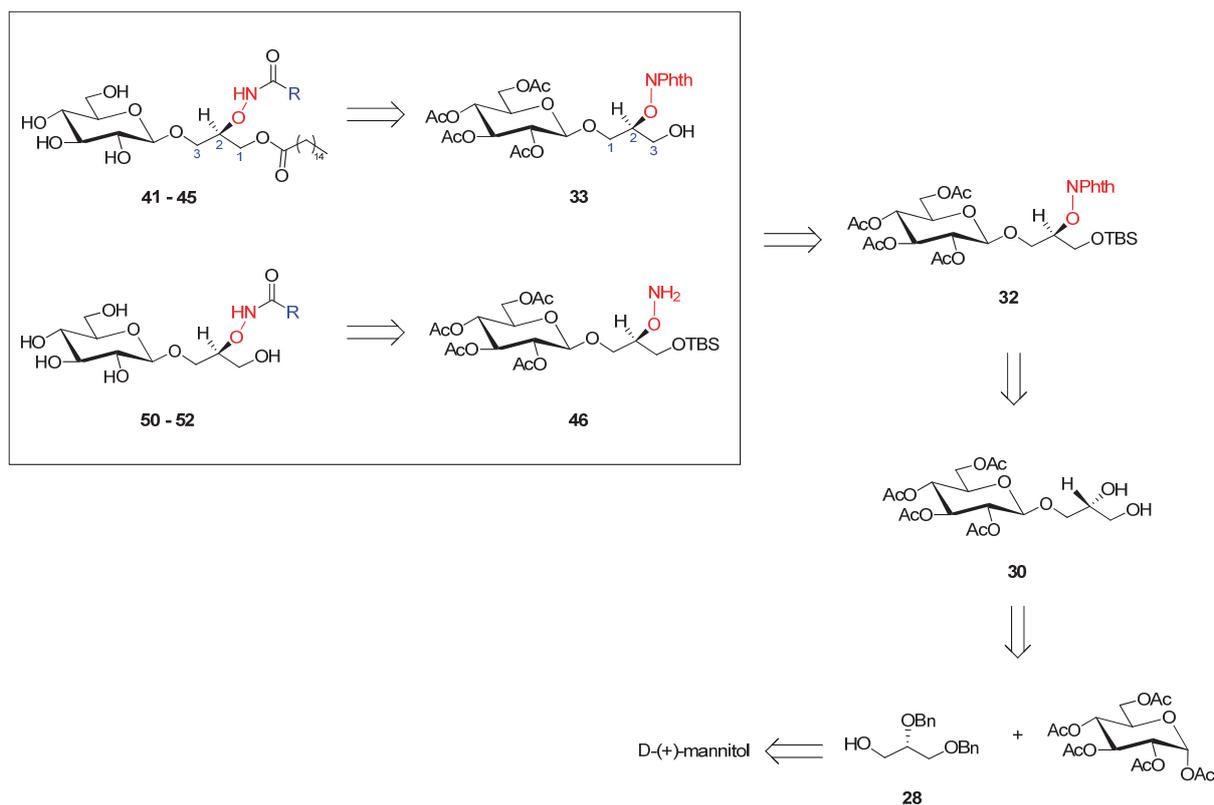
Figure 54: Structure of *N*-oxyamide-linked glucoglycerolipids.

The synthetic strategy toward *N*-oxyamide containing glucolipids **41-45** is presented in Scheme 71. It is noteworthy that glucolipids **41-45** have four common structural features, which contain two lipid chains at C1 and C2 positions, *N*-oxyamide-linkage, β -glycosidic bond and the chiral center. The two lipid chains could be ligated at C2 and C3 positions of *O*-amino β -glucoglycerol **33**, which can be obtained via desilylation of compound **32**. The aminoxy function of **32** was introduced through Mitsunobu reaction on the glucosylglycerol **30**, which was envisaged as a key intermediate. We assumed that the glycosyl donor α -*D*-glucose pentaacetate with a participating acetyl group at 2'-position could couple with (*S*)-1,2-di-*O*-benzyl-glycerol **28** to construct the β -glycosidic bond. From D-(+)-mannitol as the chiral source, glycerol **28** could be prepared easily using reported conditions.^{136,137}

The *N*-oxyamide-linked glucoglycerolipids with just one lipid chain **50-52** could be synthesized by acylation of the oxyamine derivative **46** with different fatty acids. The oxyamine derivative **46** could be obtained by hydrazinolysis of **32** (Scheme 71).

¹³⁶ van Boeckel, C. A. A.; Visser, G. M.; van Boom, J. H. *Tetrahedron* **1985**, *41*, 4557-4565.

¹³⁷ Mannock, D. A.; Lewis, R. N.; McElhaneay, R. N. *Chem. Phys. Lipids*. **1987**, *43*, 113-127.



Scheme 71: Retrosynthesis of *N*-oxyamide-linked glucoglycerolipids.

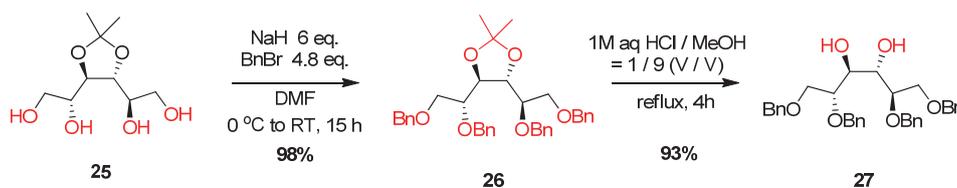
II.2 Synthesis of *N*-oxyamide-linked glucoglycerolipids

II.2.a Synthesis of (*S*)-1,2-di-*O*-benzyl-glycerol

In order to introduce our required functions, (*S*)-1,2-di-*O*-benzyl-glycerol **28** was chosen as the glycosyl acceptor. The benzyl protecting group could be selectively removed in the presence of ester group which was used to protect hydroxyl functions of the glycosyl donor.

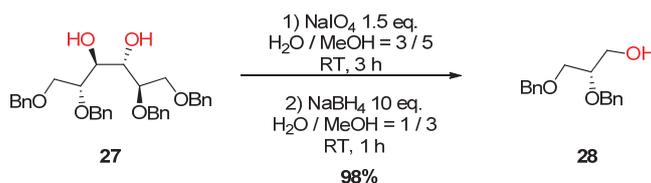
The synthesis of (*S*)-1,2-di-*O*-benzyl-glycerol **28** was performed following a reported procedure by van Boeckel *et al.*¹³⁶ The commercial D-(+)-mannitol was chosen as the starting material. The conversion of D-(+)-mannitol to 1,2:3,4:5,6-tri-*O*-isopropylidene-D-mannitol **24** was realized in 90% yield using a procedure developed by Wiggins,¹³⁸ by shaking mannitol and equimolar quantity of sulphuric acid in dry acetone for 6 h at room temperature (Scheme 72).

¹³⁸ Wiggins, L. F. *J. Chem. Soc.* **1946**, 13-14.



Scheme 73: Synthesis of 1,2,5,6-tetra-*O*-benzyl-D-mannitol **27**.

Subsequently, oxidative cleavage of the compound **27** with 1.5 equivalents of sodium periodate (NaIO_4) in a mixture of H_2O and MeOH (3/5) at room temperature for 3 h gave 2,3-di-*O*-benzyl-D-glyceraldehyde, which was sufficiently pure to be used without any further purification. Finally, the desired (*S*)-1,2-di-*O*-benzyl-glycerol **28** was obtained by reduction of the obtained aldehyde with 10 equivalents of sodium borohydride (NaBH_4) in a mixture of H_2O and MeOH (1/3) at room temperature for 1 h. After being neutralized with acetic acid and purification by column chromatography, the pure product was produced in excellent yield (98% for two steps) (Scheme 74).¹³⁷

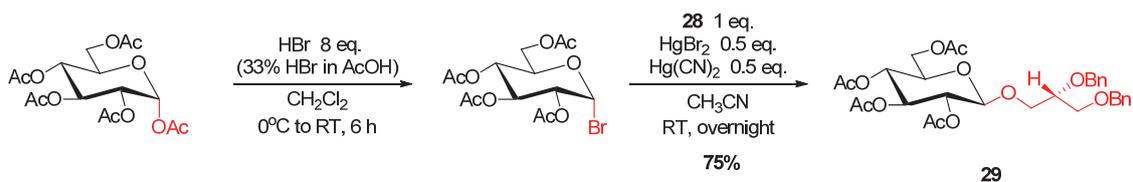


Scheme 74: Synthesis of (*S*)-1,2-di-*O*-benzyl-glycerol **28**.

II.3.2 Synthesis of 2-*O*-amino- β -glucoglycerol

In order to have high stereoselectivity and efficiency of glycosylation, we decided to use the Koenigs-Knorr procedure which usually worked well for the synthesis of β -glucoside with good yield. With the presence of a participating group at the 2'-hydroxyl function, the per-*O*-acetyl- α -D-glucopyranosyl bromide was chosen as the glycosyl donor.

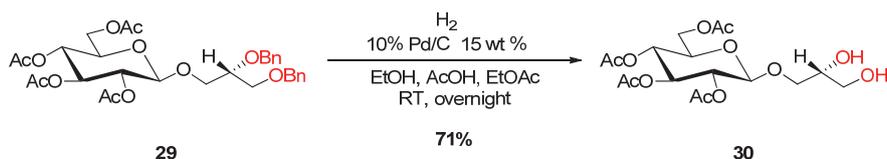
Treatment of α -D-Glucose pentaacetate with 8 equivalents of HBr (33% HBr in AcOH) in anhydrous CH_2Cl_2 at room temperature for 6 h gave the per-*O*-acetyl- α -D-glucopyranosyl bromide, which was used directly for the glycosylation without further purification (Scheme 75). Condensation of equimolar quantity of (*S*)-1,2-di-*O*-benzyl-glycerol **28** with the glucosyl bromide in dry acetonitrile, in the presence of HgBr_2 (0.5 eq.) and $\text{Hg}(\text{CN})_2$ (0.5 eq.), afforded β -glucoglycerol derivative **29** after stirring at room temperature overnight. Dropwise addition of glycosyl bromide over a period of 30 min is crucial for the good yield (75% for two steps) (Scheme 75).¹³⁶



Scheme 75: Synthesis of β -glucoglycerol derivative **29**.

The ^1H and ^{13}C -NMR values for all the protons and carbons were assigned on the basis of Dept-135, COSY, and HMQC spectra. The large coupling constant observed for the anomeric proton of **29** appeared at δ 4.55 (d, $J_{1,2} = 8.2$ Hz), which suggested the stereochemistry of the new glycoside bond being in the β -position (the coupling constants for the anomeric proton of α -anomer is about 3 Hz).¹³⁹

Compound **29** was then debenzylated by catalytic hydrogenolysis with 10% palladium on carbon (15 wt %) in ethyl acetate containing 5% each of EtOH and AcOH at room temperature, affording **30** in 71% yield (Scheme 76).

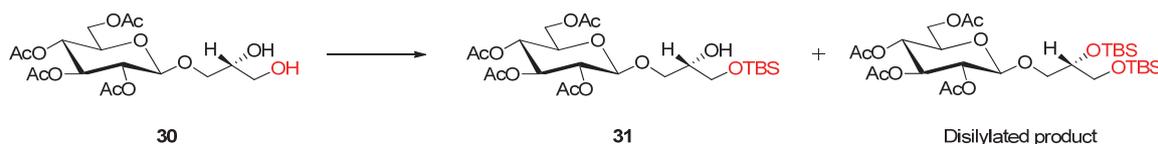


Scheme 76: Debenzylation of **29** affording β -glucoglycerol **30**.

In order to introduce the oxyamine group on the expected position, TBS was chosen as the protecting group for the regioselective protection of **30**. Compound **30** was treated with 1.4 equivalents of TBSCl in pyridine in the presence of catalytic DMAP (0.2 eq.), but the conversion is only about 10% after stirring at room temperature overnight. When additional 1.4 equivalents of TBSCl and 0.2 equivalent of DMAP was added to the reaction mixture for stirring another 24 h, the conversion is still about 10% (Entry 1, Table 8). A better conversion of 50% was obtained with the condition of TBSCl (1.2 eq.) and imidazole (2.5 eq.) in DMF at room temperature overnight (Entry 2).¹⁴⁰ Increasing amount of TBSCl (2.5 eq.) and imidazole (5.0 eq.) led to 100% conversion, but unfortunately, the obtained compound was exclusively disilylated product (Entry 3).

¹³⁹ Prakash, I.; Chaturvedula, V. S. *Molecules* **2014**, *19*, 20280-20294.

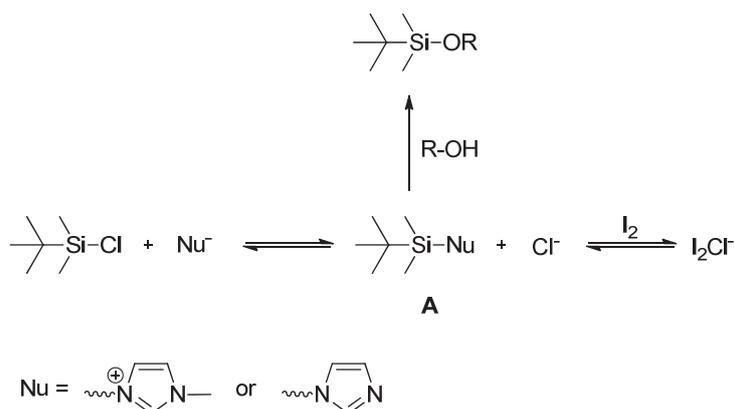
¹⁴⁰ Wang, X.; Porco, J. A. *J. Am. Chem. Soc.* **2003**, *125*, 6040-6041.



Entry	Conditions	Results
1	1.4 eq TBSCl, 0.2 eq. DMAP in pyridine, then 1.4 eq TBSCl, 0.2 eq. DMAP	~90% SM 30 + 10% 31
2	1.2 eq TBSCl, 2.5 eq. imidazole in DMF	~50% SM 30 + 50% 31
3	2.5 eq TBSCl, 5 eq. imidazole in DMF	100% disilylated product

Table 8: Different conditions for the synthesis of **31**.

Bartoszewicz *et al.* reported an efficient silylating system for primary, secondary and tertiary alcohols, consisting of a silyl chloride/*N*-methylimidazole/iodine.¹⁴¹ They speculated a mechanism for this system. Firstly, the reaction of a silyl chloride with a nucleophile catalyst (such as imidazole, *N*-methylimidazole) forms a reactive intermediate **A**. Then, the expected silylated product was formed from the reaction of **A** with an alcohol (R-OH). In the presence of iodine, which was assumed to have a propensity to the formation of polyhalide anions (I₂Cl⁻), the concentration of chloride anions could be depleted due to formation of I₂Cl⁻. Thus, the equilibrium between a silyl chloride and a nucleophile catalyst could be shifted toward the reactive intermediate **A**, which is crucial for the formation of silylated product (Scheme 77).

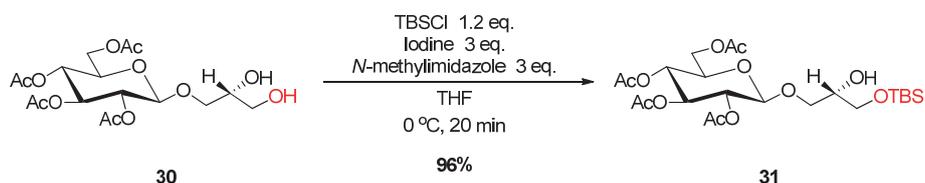


Scheme 77: A putative mechanism of silylation of alcohols in the presence of iodine and a nucleophile catalyst.¹⁴¹

The silyl chloride/*N*-methylimidazole/iodine system was attempted for the regioselective silylation of compound **30**. Fortunately, the best yield of 96% was obtained by using 1.2

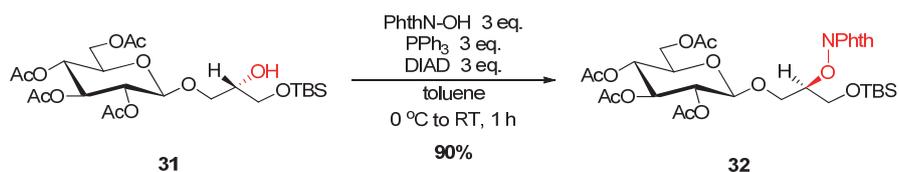
¹⁴¹ Bartoszewicz, A.; Kalek, M.; Stawinski, J. *Tetrahedron* **2008**, *64*, 8843-8850.

equivalents of TBSCl in THF, promoted by *N*-methylimidazole (3 eq.) and iodine (3 eq.), at 0 °C for 20 min (Scheme 78).¹⁴²



Scheme 78: Regioselective silylation of compound **30**.

The oxyamine function was introduced via Mitsunobu reaction. Compound **31** was reacted with PhthN-OH (3 eq.) in the presence of PPh₃ (3 eq.) at 0 °C, followed by slow drop-wise addition of 3 equivalents of DIAD (1 drop every 5 seconds). The appearance of a dark red coloring means the formation of compound **32** which is accompanied by an inversion of configuration.^{143,144} The excellent yield of 90% was obtained after 1 h stirring at room temperature (Scheme 79).



Scheme 79: Synthesis of **32** via Mitsunobu reaction.

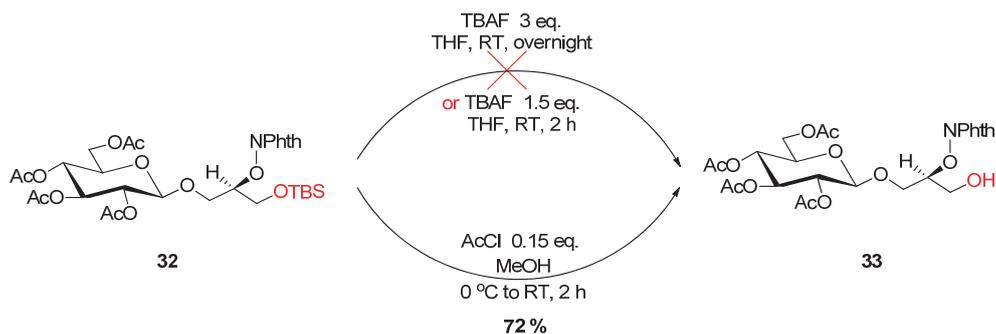
To remove the TBS group, compound **32** was firstly treated with 3 equivalents of TBAF in THF at room temperature overnight, but this condition deprotected also acetyl groups leading to a mixture of products. The milder condition (1.5 equivalents of TBAF in THF at room temperature for 2 h) also led to a mixture of products (Scheme 80). We then decided to realize the desilylation under acidic condition (AcCl in MeOH).¹⁴⁵ Catalytic AcCl (0.15 eq.) was added to a stirred solution of **32** in dry MeOH at ice-bath temperature, following by stirring at room temperature for 2 h. 5% Aqueous NaHCO₃ was added to neutralize the reaction mixture after completion of the reaction (monitored by TLC). Hopefully, this condition is efficient for removing only the TBS group, leading to the target compound **33** in 72% yield (Scheme 80).

¹⁴² Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

¹⁴³ Schenk, S.; Weston, J.; Anders, E. *J. Am. Chem. Soc.* **2005**, *127*, 12566-12576.

¹⁴⁴ But, T. Y. S.; Toy, P. H. *Chem. Asian J.* **2007**, *2*, 1340-1355

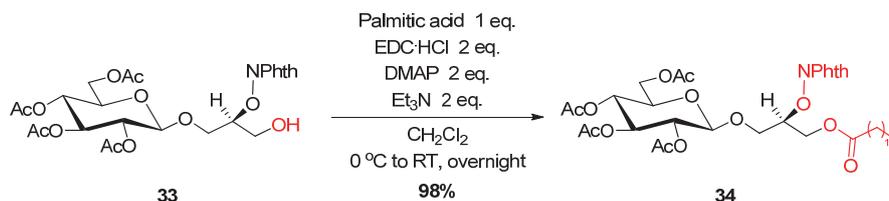
¹⁴⁵ Lin, L.; Shen, Q.; Chen, G. R.; Xie, J. *Bioorg. Med. Chem.* **2008**, *16*, 9757-9763.



Scheme 80: Desilylation of compound **32** using TBAF or AcCl.

II.2.c Synthesis of glucoglycerolipids

From 2-*O*-phthaloylamino- β -glucoglycerol **33**, *N*-oxyamide-linked glucoglycerolipids could be prepared by introduction of lipid chains and final deprotection. Firstly, the chosen palmitoyl chain was introduced by esterification. Coupling equimolar quantity of palmitic acid with *O*-phthaloylamino- β -glucoglycerol **33** in the presence of EDC·HCl/DMAP/Et₃N, afforded the glucolipid **34** in 98% yield (Scheme 81).



Scheme 81: Synthesis of **34** by esterification.

The oxyamine group was deprotected by hydrazinolysis. The reported methods were used to remove the phthaloyl group (Table 9).^{146,147} As shown in Table 9, treatment of **33** with 3 equivalents of methylhydrazine in a mixture of CHCl₃/MeOH (10/1) after 2 h at room temperature gave two spots on the TLC (Entry 1). Unfortunately, it was very difficult to separate the two products, and NMR of the mixture was complicated. The same two spots were observed on the TLC when we changed the solvent to a mixture of CH₂Cl₂/MeOH (1/1, V/V), or CH₂Cl₂/EtOH (1/3, V/V) (Entry 2, 3). When the compound **34** was treated with 2 equivalents of hydrazine in MeOH at room temperature for 3 h, a main spot and three other spots were observed on the TLC (Entry 4). After workup, NMR of the mixture showed the missing of acetyl group. As the acetyl group could also be removed by hydrazine¹⁴⁸, mild conditions (1.1

¹⁴⁶ Johnson, S. M.; Petrassi, H. M.; Palaninathan, S. K.; Mohamedmohaideen, N. N.; Purkey, H. E.; Nichols, C.; Chiang, K. P.; Walkup, T.; Sacchettini, J. C.; Sharpless, K. B. *J. Med. Chem.* **2005**, *48*, 1576-1587.

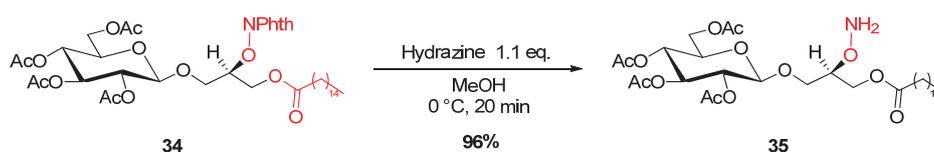
¹⁴⁷ Leduc, A. B.; Kerr, M. A. *Angew. Chem. Int. Ed. Engl.* **2008**, *47*, 7945-7948.

¹⁴⁸ Janwitayanuchit, W.; Suwanborirux, K.; Patarapanich, C.; Pummangura, S.; Lipipun, V.; Vilaivan, T. *Phytochemistry* **2003**, *64*, 1253-1264.

equivalent of hydrazine, 20 min at 0 °C) were used to deprotect selectively the phthaloyl group to obtain oxyamine **35** in 96% yield (Entry 5) (Scheme 82).

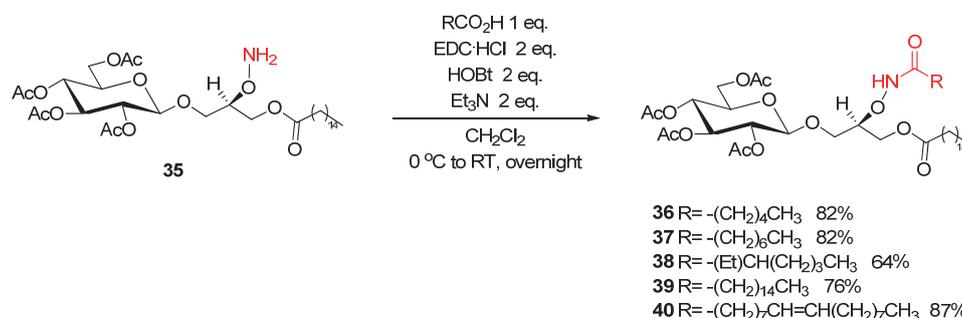
Entry	Conditions	Results
1	MeNHNH ₂ (3 eq.), CHCl ₃ /MeOH 10/1, RT, 2 h	Two products (difficult to separate)
2	MeNHNH ₂ (1.5 eq.), CH ₂ Cl ₂ /MeOH 1/1, RT, 2h	Two products (difficult to separate)
3	MeNHNH ₂ (1.5 eq.), CH ₂ Cl ₂ /EtOH 1/3, RT, 2 h	Two products (difficult to separate)
4	NH ₂ NH ₂ (2 eq.), MeOH, RT, 3 h	Targeted product and deacetylated products
5	NH ₂ NH ₂ (1.1 eq.), MeOH, 0 °C, 20 min	Target product (96% yield)

Table 9: Attempted conditions for the phthaloyl deprotection of **34**.



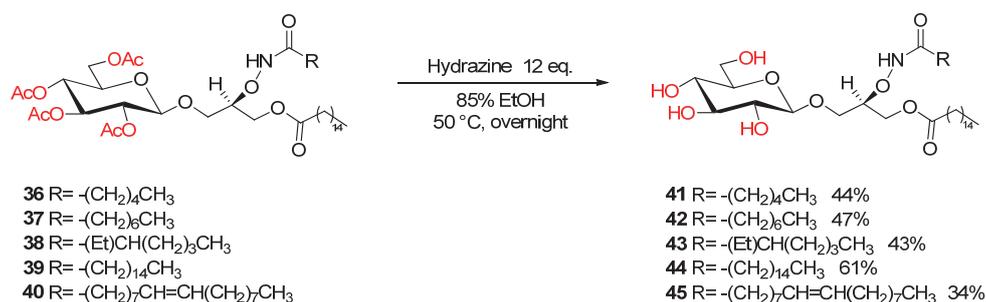
Scheme 82: Deprotection of **34**.

Subsequently, acylation of the obtained oxyamine **35** with equimolar quantity of different fatty acids under usual conditions led successfully to the *N*-oxyamide-linked glucoglycerolipids **36-40** in good yields [**36** (82%), **37** (82%), **38** (64%), **39** (76%), **40** (87%)] (Scheme 83). The ¹H NMR spectra of **36-40** in CDCl₃ showed the NH signals appearing in the range of 8.81-8.87 ppm, indicating the formation of *N*-oxyamide bond.



Scheme 83: Synthesis of *N*-oxyamide-linked glucoglycerolipids **36-40**.

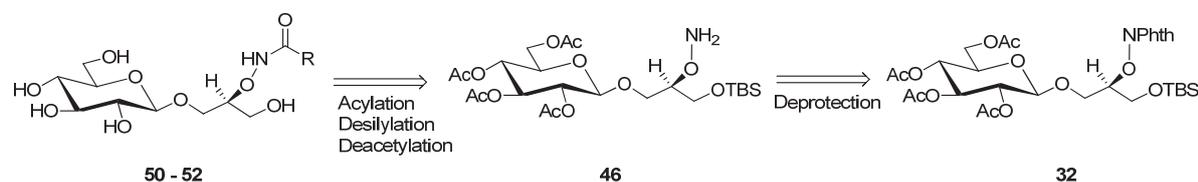
The most delicate step in this synthesis was the selective removal of the acetate groups of **36-40** in the presence of the fatty ester groups on the glycerol moiety. Hopefully, deacetylation of **36-40** was carried out with 12 equivalents of hydrazine (3 eq. per acetate group) at 50 °C in 85% aqueous ethanol overnight to give the corresponding glucoglycerolipids **41-45** (Scheme 84). Due to their amphiphilic nature, some glycolipids were lost during the workup, leading to low yields [**41** (44%), **42** (47%), **43** (43%), **44** (61%), **45** (34%)]. All of the final *N*-oxyamide-linked glucoglycerolipids **41-45** have been fully characterized by ¹H, ¹³C NMR and HRMS. Regretfully, the signal of NH in the *N*-oxyamide bond could not be observed on the ¹H NMR in CD₃OD.



Scheme 84: Deacetylation of **36-40** to obtain final *N*-oxyamide-linked glucoglycerolipids **41-45**.

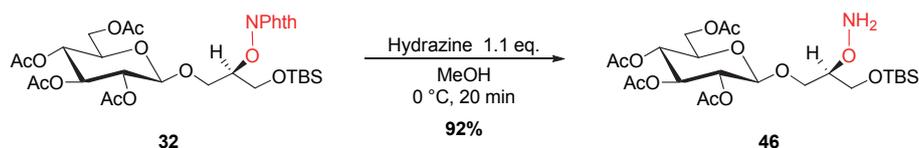
II.3 Synthesis of *N*-oxyamide-linked glucoglycerolipids with one lipid chain

From intermediate *O*-phthaloylamino β -glucoglycerol **32**, *N*-oxyamide-linked glucoglycerolipids with just one lipid chain (**50-52**) could be prepared by deprotection of phthaloyl group and introduction of lipid chains, followed by desilylation and final deacetylation (Scheme 85).



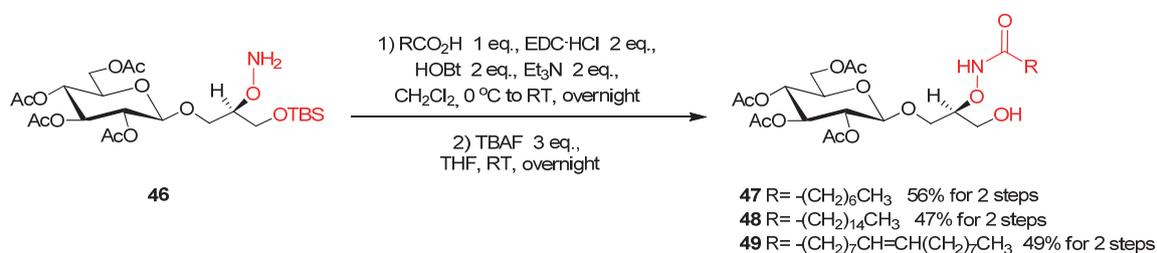
Scheme 85: Retrosynthesis of *N*-oxyamide-linked glucoglycerolipids with one lipid chain.

Firstly, the selective deprotection of phthaloyl group was realized following the procedure which was used for synthesis of **35**. Hydrazinolysis under mild conditions (1.1 equivalents of hydrazine, 20 min at 0 °C) deprotected selectively the phthaloyl group of **32** affording the oxyamine derivative **46** in 92% yield (Scheme 86).



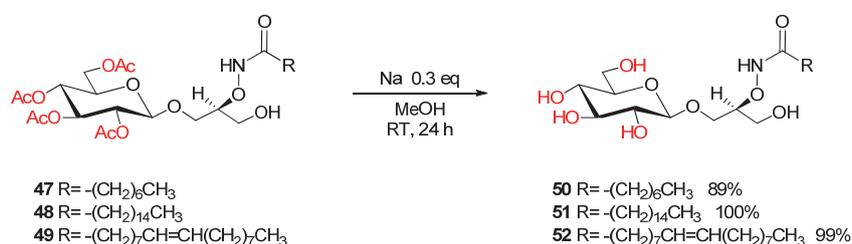
Scheme 86: Deprotection of the oxyamine of **32** to obtain **46**.

Subsequent coupling of the oxyamine **46** with equimolar quantity of different fatty acids in CH_2Cl_2 in the presence of EDC·HCl/DMAP/ Et_3N , followed by desilylation with 3 equivalents of TBAF in THF at room temperature, led to the corresponding *N*-oxyamide-linked glucolipids with one lipid chain (**47-49**) in 47-56% yields for two steps (Scheme 87). The ^1H NMR spectra of **47-49** in CDCl_3 showed the NH signals appearing in the range of 9.19-9.27 ppm, indicating the formation of *N*-oxyamide bond.



Scheme 87: Synthesis of glucoglycerolipids **47-49**.

Finally, acetyl groups of **47-49** were removed by using catalytic Na (0.3 eq.) in methanol at room temperature for 24 h to give the corresponding (2*S*)-1-*O*- β -D-glucopyranosyl-2-*O*-acylaminoglycerol **50-52** in excellent yields [**50** (89%), **51** (100%), **52** (99%)] (Scheme 88). All of the final *N*-oxyamide-linked glucoglycerolipids **50-52** have been fully characterized by ^1H , ^{13}C NMR and HRMS. Once again, the signal of NH in the *N*-oxyamide bond could not be observed on the ^1H NMR in CD_3OD .

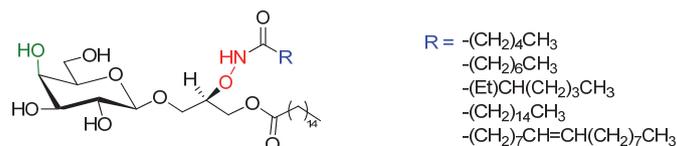


Scheme 88: Deacetylation of **47-49** to obtain **50-52**.

III Synthesis of *N*-oxyamide-linked galactoglycerolipids

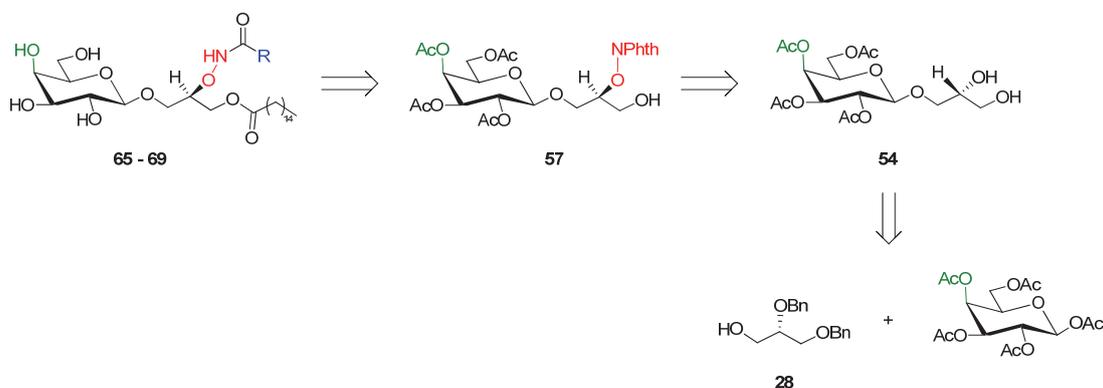
III.1 Retrosynthesis

In the previous section, we have successfully prepared the *N*-oxy amide-linked glucoglycerolipids. In order to compare the effect of sugar modification, we have decided to synthesize *N*-oxyamide-linked galactoglycerolipids (Scheme 89).



Scheme 89: Structures of *N*-oxyamide-linked galactoglycerolipids.

Similar to the synthetic route for *N*-oxyamide-linked glucoglycerolipids, the *N*-oxyamide-linked galactoglycerolipids could be prepared easily. The desired galactolipids **65-69** would be obtained by introducing two lipid chains on the 2-*O*-amino- β -galactoglycerol **57** which could be prepared through Mitsunobu reaction on galactosylglycerol **54**. The galactosylglycerol **54** can be obtained via glycosylation from galactose pentaacetate and dibenzylated glycerol **28** (Scheme 90).



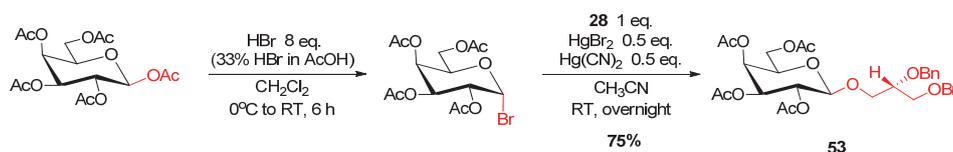
Scheme 90: Retrosynthesis of *N*-oxyamide-linked galactoglycerolipids.

III.2 Synthesis of *N*-oxyamide-linked galactoglycerolipids

III.2.a Synthesis of 2-*O*-amino- β -galactoglycerol

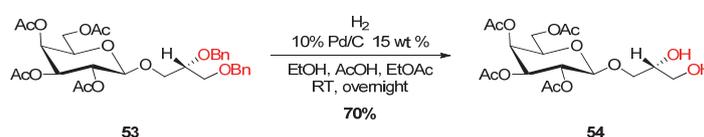
For the synthesis of 2-*O*-amino- β -galactoglycerol, the β -galactoglycerol derivative was firstly synthesized by glycosylation following the Koenings-Knorr method. β -D-Galactose

pentaacetate was converted to galactosyl bromide in about 100% yield by means of 8 equivalents of HBr (33% HBr in AcOH) in anhyd CH₂Cl₂ at room temperature for 6 h (Scheme 91). Glycosylation of the freshly prepared per-*O*-acetyl- α -D-galactopyranosyl bromide with equimolar quantity of (*S*)-1,2-di-*O*-benzyl-glycerol **28** in dry acetonitrile, in the presence of HgBr₂ (0.5 eq.) and Hg(CN)₂ (0.5 eq.), gave β -galactoglycerol derivative **53** after stirring at room temperature for 15 h. The galactosyl bromide was added drop by drop under vigorous stirring over a period of 30 min, leading to good yield (75% for two steps) (Scheme 90). The large coupling constant observed for the anomeric proton of **53** appeared at δ 4.54 (d, $J_{1,2} = 7.9$ Hz), which suggested the stereochemistry of the new glycoside bond being in the β -orientation.



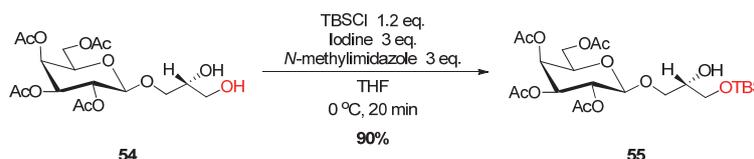
Scheme 91: Synthesis of β -galactoglycerol derivative **53**.

Debenzylation of **53** by catalytic hydrogenolysis with 10% palladium on carbon (0.15 eq.) in ethyl acetate containing 5% each of EtOH and AcOH at room temperature afforded β -galactoglycerol **54** in 70% yield (Scheme 92).



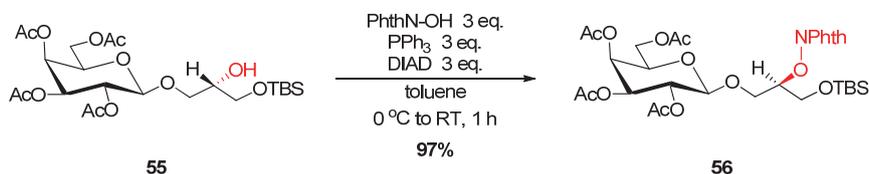
Scheme 92: Debenzylation of **53** affording β -galactoglycerol **54**.

After debenzylation, regioselective silylation of **54** was realized using the efficient silylating system reported by Bartoszewicz *et al*, in which the presence of *N*-methylimidazole and iodine was required as promoter to ensure good reactivity and regioselectivity.¹⁴¹ The expected compound **55** was obtained rapidly in 90% yield by condensation of β -galactoglycerol **54** with 1.2 equivalents of TBSCl in THF at 0 °C for 20 min, promoted by *N*-methylimidazole (3 eq.) and iodine (3 eq.) (Scheme 93).



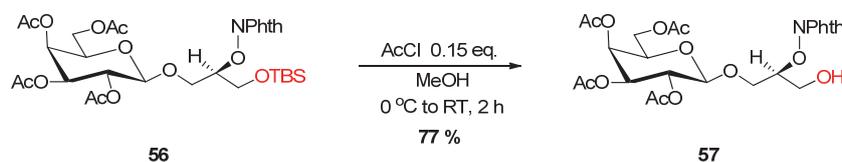
Scheme 93: Regioselective silylation of **54** to obtain **55**.

The introduction of the aminoxy function was realized via Mitsunobu reaction. Treatment of compound **55** with PhthNOH (3 eq.) in the presence of PPh₃ (3 eq.) at 0 °C, followed by slow dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds), led successfully to the compound **56** in 97% yield (Scheme 94).^{143,144}



Scheme 94: Synthesis of **56** via Mitsunobu reaction.

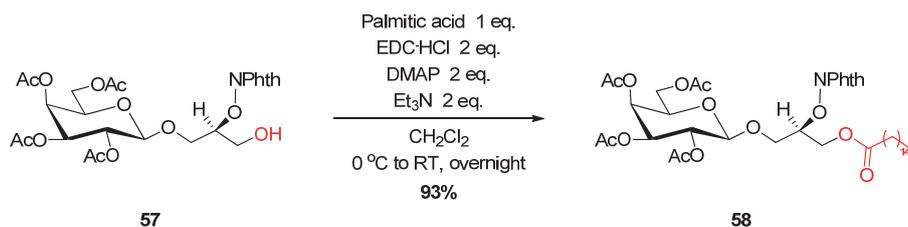
Removal of TBS group was achieved by adding catalytic AcCl (0.15 eq.) to a stirred solution of **56** in MeOH at 0 °C, followed by stirring at room temperature for 2 h. 5% Aqueous NaHCO₃ was added to neutralize the reaction mixture after completion of the reaction (monitored by TLC). This condition was efficient for preparing the 2-*O*-phthaloylamino- β -galactoglycerol **57** in 77% yield, instead of TBAF which deprotected also acetyl groups, leading to a mixture of products (Scheme 95).



Scheme 95: Desilylation of **56** affording 2-*O*-phthaloylamino- β -galactoglycerol **57**.

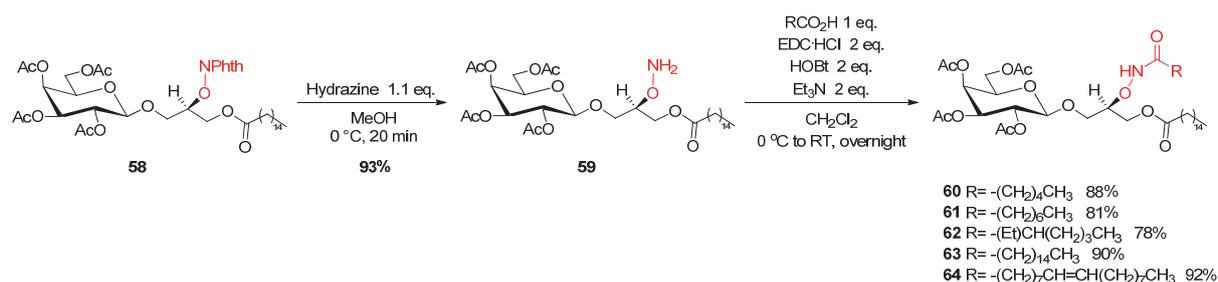
III.2.b Synthesis of galactoglycerolipids

The target *N*-oxyamide-linked galactoglycerolipids could be synthesized by introduction of lipid chains to *O*-phthaloylamino- β -galactoglycerol **57** and final deacetylation. Firstly, esterification of the *O*-phthaloylamino β -galactoglycerol **57** with equimolar quantity of palmitic acid in the presence of EDC·HCl/DMAP/Et₃N, furnished the galactolipid **58** in 93% yield (Scheme 96).



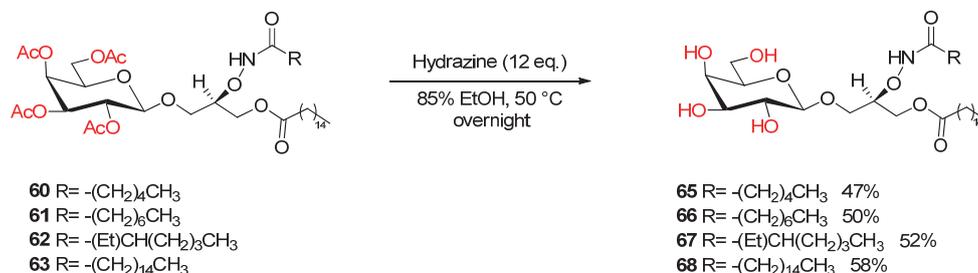
Scheme 96: Synthesis of **58** by esterification.

Since the acetyl group could also be removed by hydrazine, mild conditions (1.1 eq. hydrazine, 20 min, at 0 °C) was used to deprotect selectively the phthaloyl group to obtain oxyamine compound **59** (93% yield). Then, coupling of the oxyamine **59** with equimolar quantity of different fatty acids in the presence of EDC·HCl (2 eq.), HOBt (2 eq.) and Et₃N (2 eq.) at room temperature, led successfully to the *N*-oxyamide-linked galactoglycerolipids **60-64** in good yields [**60** (88%), **61** (81%), **62** (78%), **63** (90%), **64** (92%)] (Scheme 97). On the ¹H NMR spectra in CDCl₃, the NH signals in compounds **60-64** appeared in the range of 8.81-8.87 ppm indicating the formation of *N*-oxyamide bond.



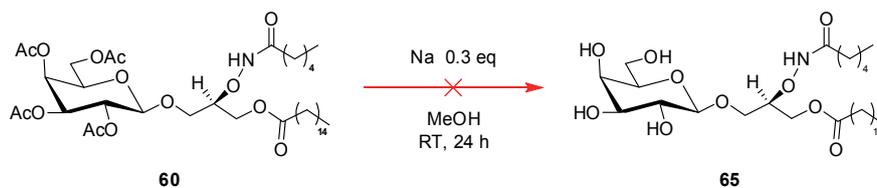
Scheme 97: Synthesis of *N*-oxyamide-linked galactoglycerolipids **60-64**.

Finally, deacetylation of **60-63** was realized with 12 equivalents of hydrazine (3 eq. per acetate group) at 50 °C in 85% aqueous ethanol to give the corresponding galactoglycerolipids **65-68** [**65** (47%), **66** (50%), **67** (52%), **68** (58%)] (Scheme 98). The signal of NH in the *N*-oxyamide bond could not be observed on the ¹H NMR in CD₃OD.



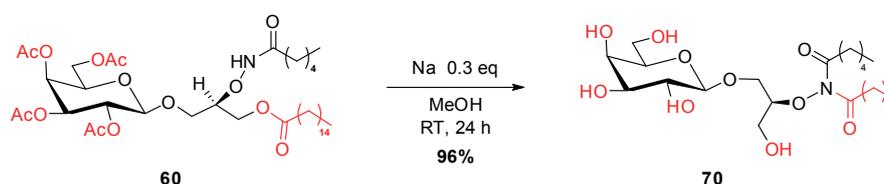
Scheme 98: Deacetylation of **60-63** to obtain final *N*-oxyamide-linked galactoglycerolipids **65-68**.

For the deprotection of glucoglycerolipids with one lipid chain (Chapter 4.II.3), we used 0.3 equivalent of sodium in MeOH at room temperature for 24 h to remove the acetyl group, and the final *N*-oxyamide-linked glucoglycerolipids **50-52** were obtained in excellent yield. The same condition (0.3 eq. Na in MeOH, at room temperature for 24 h) was used to remove the acetyl group of compound **60**. A new deacetylated product was obtained, but unfortunately, it was not the expected product **65** after comparing NMR spectra (Scheme 99 and Figure 55, Figure 56).



Scheme 99: Deacetylation of **60** using Na/MeOH.

Based on the HRMS and NMR spectra (^1H NMR, ^{13}C NMR, Dept135, COSY, HMQC), the obtained molecule was identified to be the compound **70** (Scheme 100). The proof of NMR and reaction mechanism will be discussed.



Scheme 100: Formation of compound **70**.

The ^1H and ^{13}C NMR spectra of **65** and **70** (Figure 55 and Figure 56) were compared below, and all of them were obtained in CD_3OD . As we can observe from the Figure 55, the chemical shift of H^{I} moved from δ 4.37-4.23 ppm in **65** to δ 3.71-3.61 ppm in **70**, and the H^{II} also moved to higher field, from δ 4.21-4.15 ppm in **65** to δ 4.01-3.95 ppm in **70**. The variation was also observed in the chemical shifts of C^{I} and C^{II} on the ^{13}C NMR spectra of **65** and **70** (Figure 56). The C^{I} moved to higher field from δ 63.3 ppm in **65** to δ 61.0 ppm in **70**, while the C^{II} moved to lower field, from δ 83.7 ppm in **65** to δ 87.5 ppm in **70**. The signal of NH in the *N*-oxyamide bond could not be observed in CD_3OD . According to these changes, we speculated that the palmitoyl lipid chain may transfer to oxyamine to give the structure **70**.

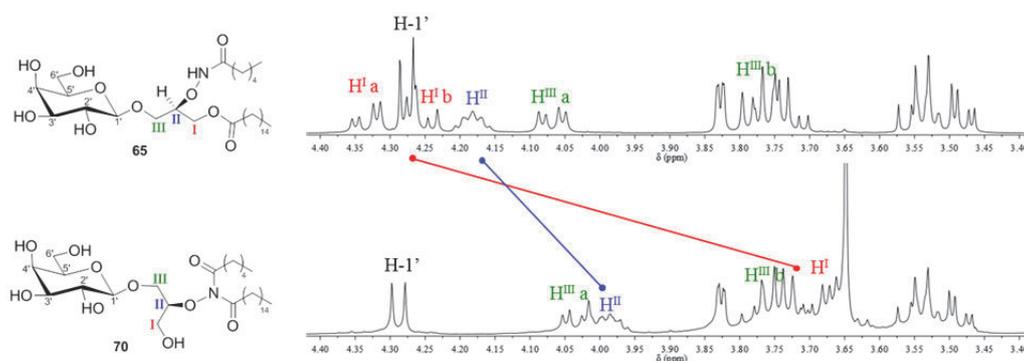


Figure 55: ^1H NMR spectra of compounds **65** and **70** in CD_3OD .

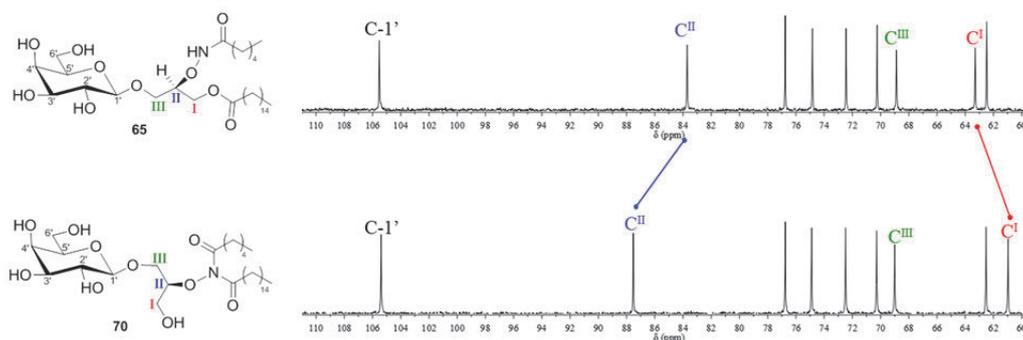
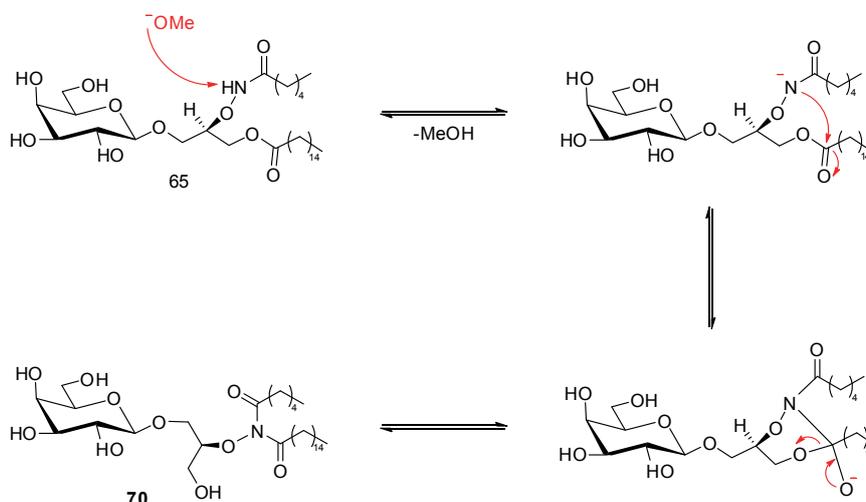


Figure 56: ^{13}C NMR spectra of compounds **65** and **70** in CD_3OD .

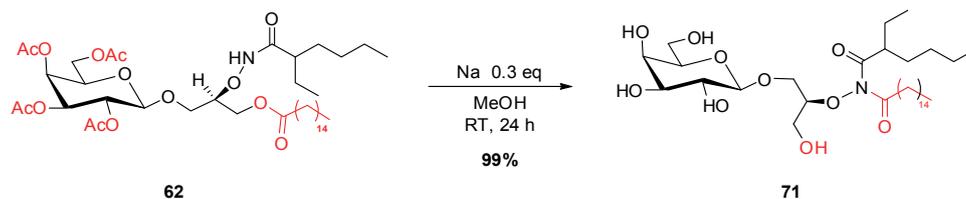
We assumed that compound **60** was deacetylated by Na/MeOH to afford compound **65** firstly, then the ester bond was cleaved by transacylation reaction. Because of the electronegativity of the adjacent oxygen, the pK_a of the oxyamide is about 10-15. The proton of $-\text{NH}$ could be removed by (NaOMe firstly, then the nucleophilic N^- attacked the electrophilic C of the ester $\text{C}=\text{O}$, breaking the π bond and creating the tetrahedral intermediate. The intermediate collapses, reforming the $\text{C}=\text{O}$, resulting in the transfer of acyl chain and the formation of alcohol, leading to the structure **70** (Scheme 101).



Scheme 101: A proposed mechanism for the formation of compound **70**.

Similarly, treatment of compound **62** with 0.3 equivalent of Na in MeOH at room temperature for 24 h led to the deacetylated and transacylated compound **71** (Scheme 102). Comparison of the ^1H NMR spectra of compounds **67** and **71** in CD_3OD (Figure 57) revealed the chemical shift of H^{I} and H^{II} moved to higher field (H^{I} : from δ 4.37-4.23 ppm in **67** to δ 3.71-3.62 ppm in **71**; H^{II} : from δ 4.21-4.15 ppm in **67** to δ 4.01-3.95 ppm in **71**). As it was shown in Figure 58, the chemical shifts of C^{I} and C^{II} also moved (C^{I} : from δ 63.2 ppm in **67**

to δ 60.9 ppm in **71**; C^{II}: from δ 83.9 ppm in **67** to δ 87.6 ppm in **71**).



Scheme 102: Formation of compound **71**.

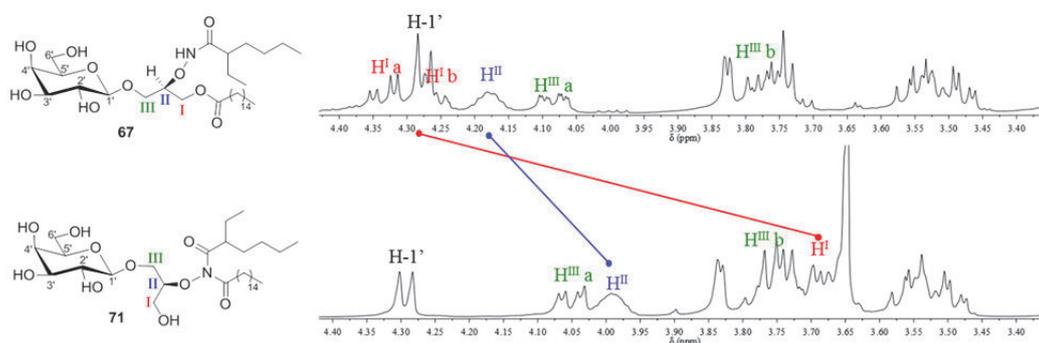


Figure 57: ¹H NMR spectra of compounds **67** and **71** in CD₃OD.

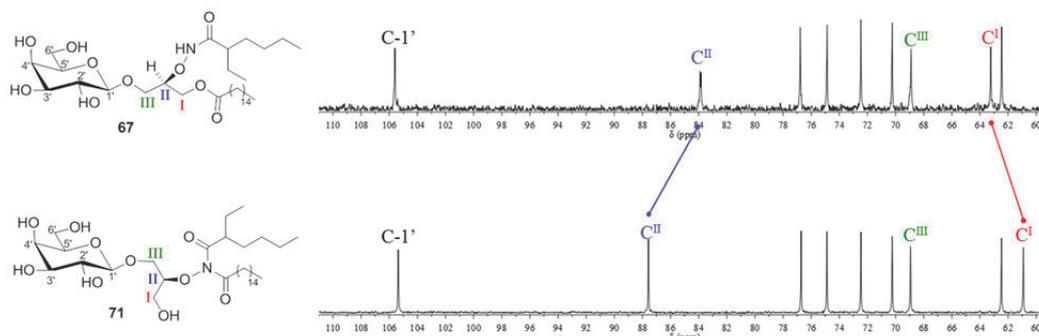
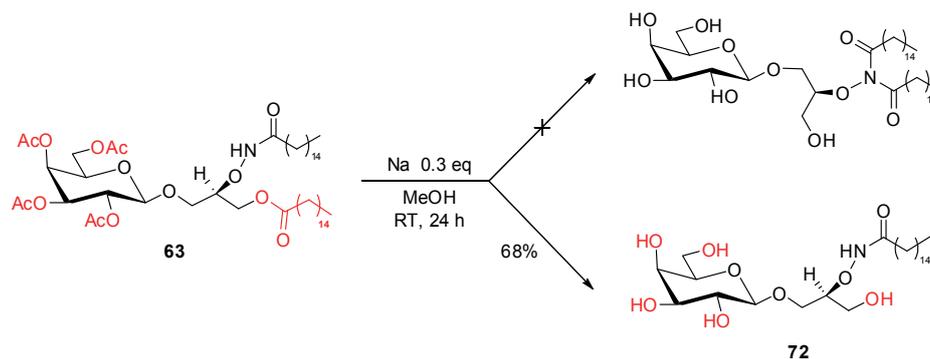


Figure 58: ¹³C NMR spectra of compounds **67** and **71** in CD₃OD.

In conclusion, both conditions (3 eq. hydrazine per OAc or 0.3 eq. Na/MeOH) could remove acetyl groups, but the transesterification condition with MeONa can lead to the intramolecular transacylation (Scheme 100 and Scheme 102).

When the compound **63** was treated with 0.3 equivalent of Na in MeOH at room temperature for 24 h, the transacylated product was not obtained. Comparison of the ¹H NMR spectra of obtained product with compound **68** shows that one lipid chain was missing. Based on the HRMS and NMR spectra, the obtained molecule was identified to be compound **72** which

has just one lipid chain attached (Scheme 103).



Scheme 103: Deprotection of compound **63** with Na/MeOH.

Comparison of the ^1H NMR spectra of compounds **68** and **72** in CD_3OD (Scheme 70) reveals that the chemical shift of H^{I} and H^{II} moved to higher field (H^{I} : from δ 4.37-4.23 ppm in **68** to δ 3.71-3.62 ppm in **72**; H^{II} : from δ 4.22-4.15 ppm in **68** to δ 4.01-3.96 ppm in **72**). Once again, comparison of the ^{13}C NMR spectra revealed that the C^{I} moved to higher field from δ 63.4 ppm in **68** to δ 60.9 ppm in **72**, while the C^{II} moved to lower field, from δ 83.8 ppm in **68** to δ 87.4 ppm in **72** (Figure 60).

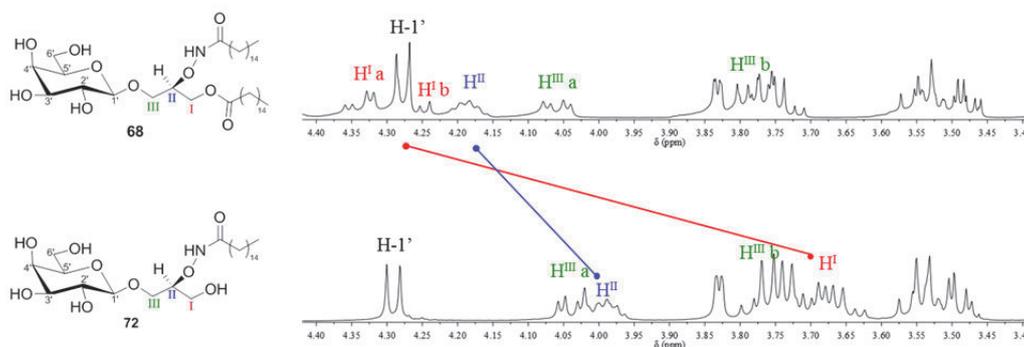


Figure 59: ^1H NMR spectra of compounds **68** and **72** in CD_3OD .

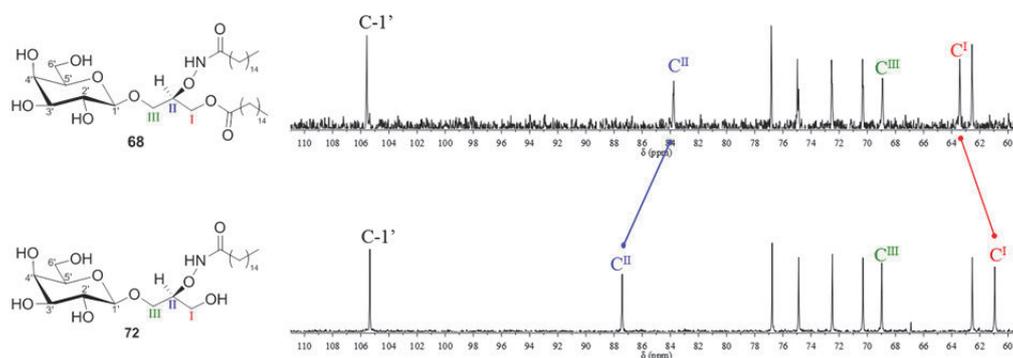


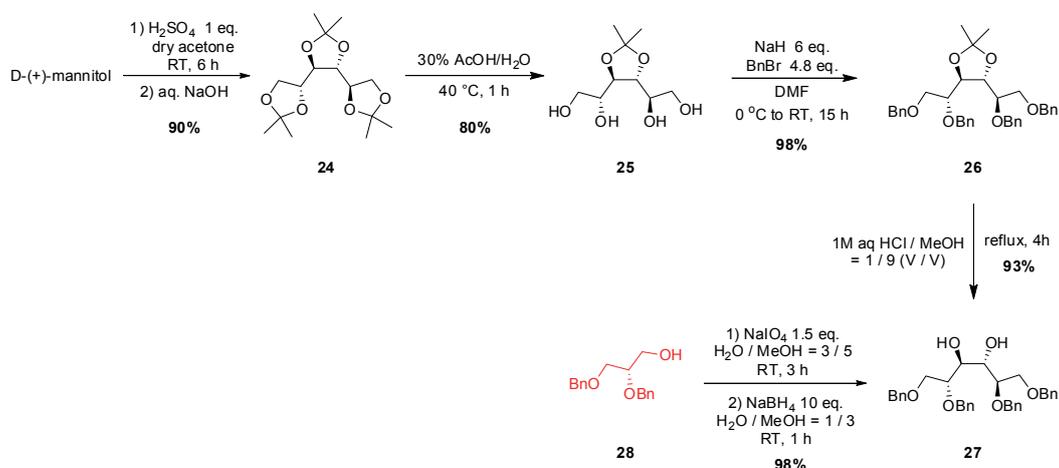
Figure 60: ^{13}C NMR spectra of compounds **68** and **72** in CD_3OD .

IV Conclusion

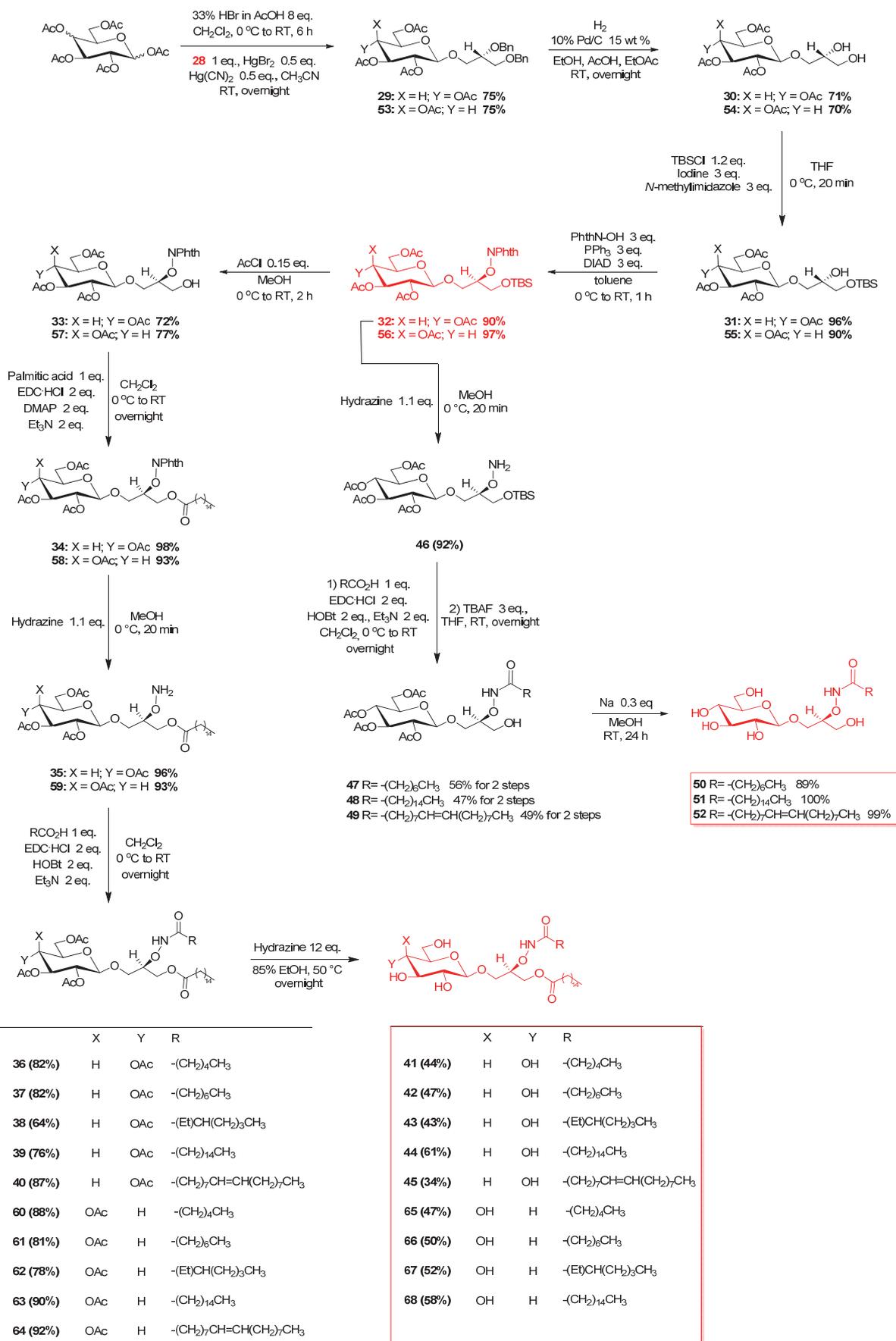
In this chapter, we have successfully achieved the first synthesis of *N*-oxyamide-linked β -glycolipids as novel mimics of glycoglycerolipids and glycosphingolipids.

From readily available D-glucose/galactose pentaacetate and D-mannitol, the β -glycosyl-glycerols **30/54** were synthesized by glycosylation. Through Mitsunobu reactions, the aminoxy functions were successfully introduced to afford the 2-*O*-amino β -glycoglycerols **33/57**. Different lipid chains were easily introduced on hydroxyl and aminoxy functions to access to a variety of glycolipid structures (Scheme 105). Synthesis of (*S*)-1,2-di-*O*-benzyl-glycerol **28** has also been optimised (Scheme 104).

The cytotoxicity of the synthesized glycoglycerolipids against the cancer cell lines H929 and Hela have been evaluated in collaboration with Dr Xiaopeng He at East China University of Science and Technology, showing no significant activity.

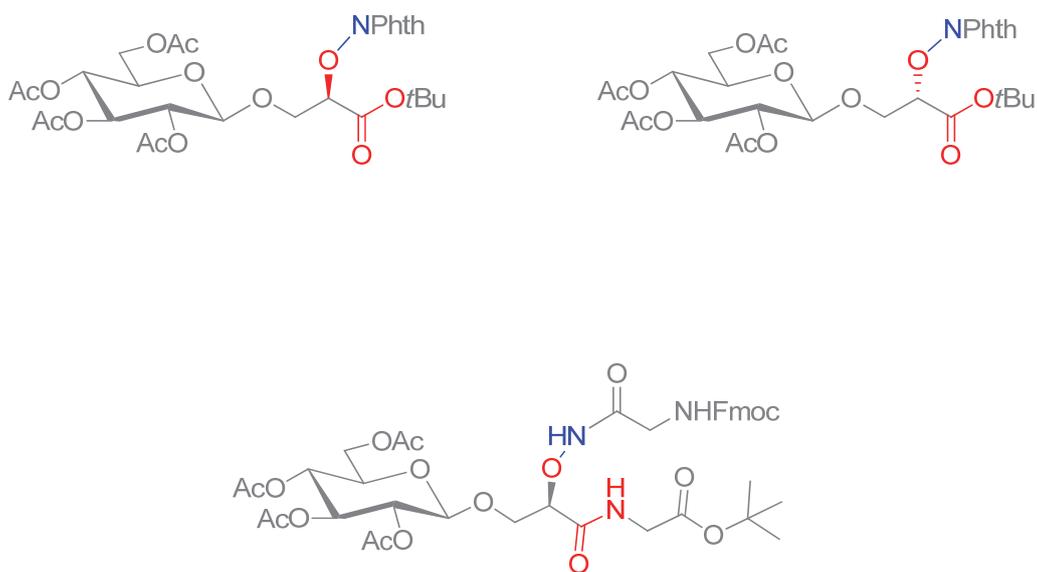


Scheme 104: Synthesis of (*S*)-1,2-di-*O*-benzyl-glycerol **28**.

Scheme 105: Synthesis of *N*-oxyamide-linked glycoglycerolipids.

Chapter 5

Synthesis of *O*-Glycosyl Aminoxy Esters and Glycopeptides



I Introduction

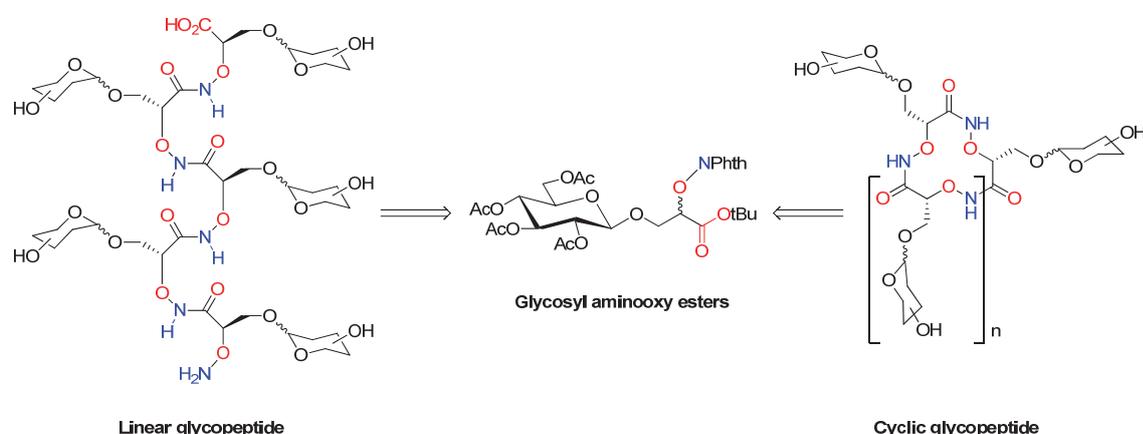
As described in Chapter 1, glycopeptides and glycoproteins play an important role in various cellular activities and pathogenic processes. The development of glycopeptide and glycoprotein mimics is of great interest. Most of the interesting functions performed by proteins in biological systems, such as molecular recognition, electron transfer and catalysis, are closely related to their unique secondary and tertiary structures. Understanding the secondary structures of the designed molecule is also required for the theoretical and pharmaceutical significance.

Studies on aminoxy-containing peptides showed that they can easily form intramolecular hydrogen bonds with turns and helices structures, and the *N*-oxyamide linkage is stable to chemical and enzymatic hydrolysis (Chapter 2). We are interested in the synthesis of glycosyl aminoxy acids and peptides, where an aminoxy function has been introduced in place of the amine group in natural ones (Figure 61).



Figure 61: Structure of *O*-glycopeptides and its aminoxy analogues.

In this chapter, we decided to synthesize the β -*O*-glucosyl 2(*R*)- and 2(*S*)-aminoxy esters as novel glycopeptide mimics, in which the aminoxy acid was linked to the glucose unit through *O*-glycosidic bond. From the multifunctional glycosyl aminoxy esters, we proposed to prepare the *N*-oxyamide-linked linear as well as the cyclic glycopeptides (Scheme 106).

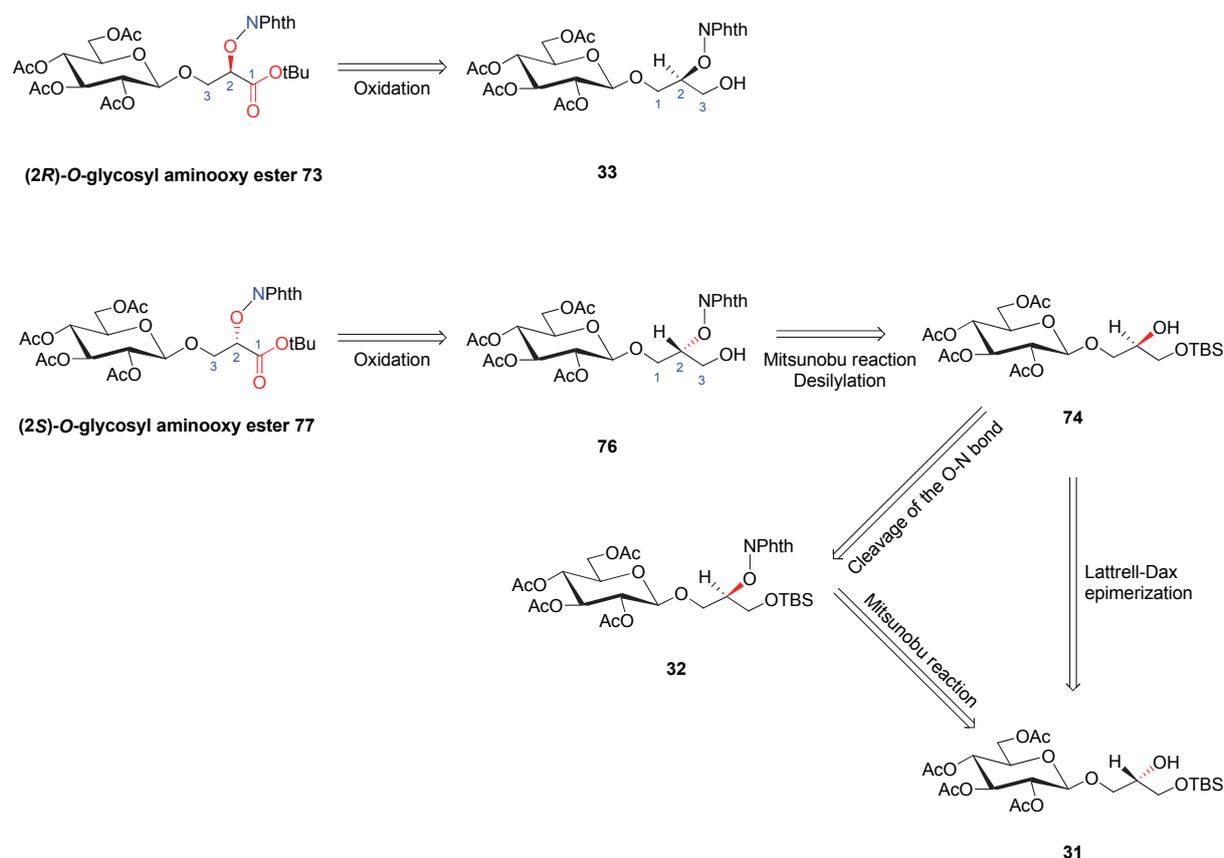


Scheme 106: Structures of the designed β -*O*-glucosyl aminoxy esters and novel glycopeptides.

II Synthesis of the β -O-glycosyl aminoxy esters

I.1 Retrosynthesis

Our retrosynthetic analysis for the preparation of β -O-glycosyl aminoxy ester **73** and **77** is shown in Scheme 107. The targeted (2*R*)-3-O-glycosyl aminoxy ester **73** could be easily synthesized by oxidation of the (2*S*)-1-O-glycosyl phthalimidooxy alcohol **33**, which has been prepared in Chapter 4.II.2.b. Likewise, the target (2*S*)-3-O-glycosyl aminoxy ester **77** could be synthesized by oxidation of the (2*R*)-1-O-glycosyl phthalimidooxy alcohol **76**, which could be obtained through Mitsunobu reaction and desilylation from the compound **74**. The key intermediate **74** could be prepared by C-2 Lattrell-Dax epimerization from its diastereomer **31**, or by cleaving the O-N bond of compound **32**. The synthesis of compounds **31** and **32** has been presented in Chapter 4.II.2.b.

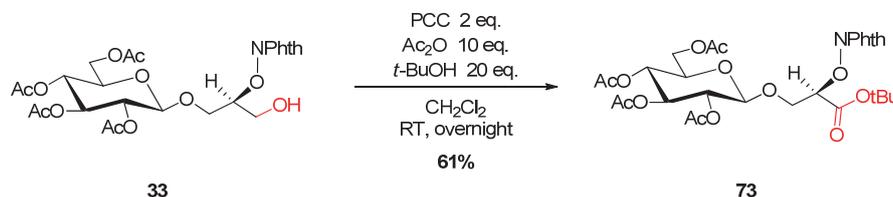


Scheme 107: Retrosynthesis of the O-glycosyl aminoxy esters.

I.2 Synthesis of (2*R*)-3-O-glycosyl aminoxy ester **73**

With the (2*S*)-1-O-glycosyl phthalimidooxy alcohol **33** in hand, the (2*R*)-3-O-glycosyl aminoxy ester **73** was prepared by oxidation. Treatment of the primary alcohol **33** with 2

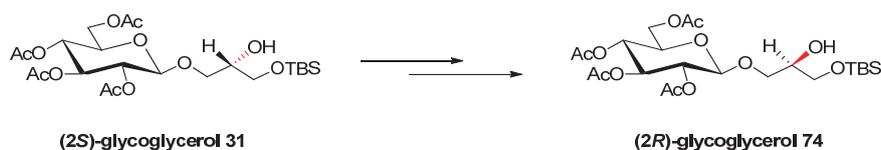
equivalents of pyridinium chlorochromate (PCC) in the presence of 10 equivalents of acetic anhydride (Ac_2O) and 20 equivalents of *t*-BuOH in CH_2Cl_2 at room temperature for one night, led successfully to the compound **73** in 61% yield (Scheme 108).



Scheme 108: Synthesis of (2*R*)-3-*O*-glycosyl aminoxy ester **73**.

I.3 Synthesis of (2*S*)-3-*O*-glycosyl aminoxy ester **77**

In order to prepare the (2*S*)-3-*O*-glycosyl aminoxy ester **77** from compound **31**, an epimerization on the C-2 position of (2*S*)-glycoglycerol **31** to generate the corresponding diastereomer (2*R*)-glycoglycerol **74** was proposed (Scheme 109).



Scheme 109: Epimerization of **31** to synthesize **74**.

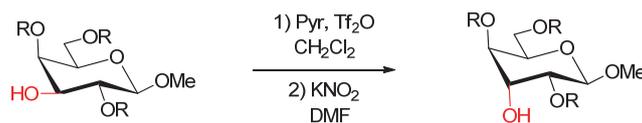
There are several different synthetic methods for the epimerization, including protocols based on the Mitsunobu reaction, sequential oxidation/reduction routes, and enzymatic methods. In our case, for the preparation of the (2*S*)-3-*O*-glycosyl aminoxy ester **77** from compound **31**, two methods were used. One was based on the Lattrell-Dax epimerization, and the other was based on the Mitsunobu reaction.

I.3.a Epimerization based on Lattrell-Dax method

The Lattrell-Dax method, in which the free hydroxyl group of the carbohydrate compound was firstly converted into a triflate, followed by a nitrite-mediated substitution, is a very efficient and convenient method to generate structures with the inversion of configuration (Scheme 110).^{149,150}

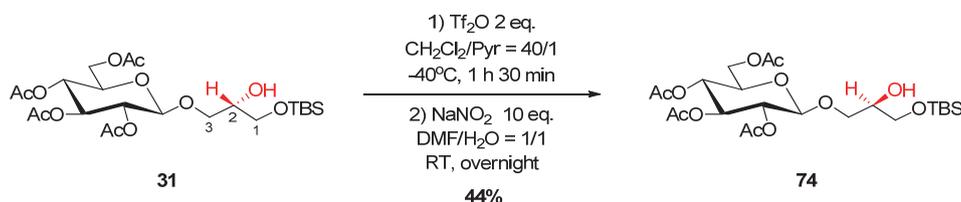
¹⁴⁹ Lattrell, R.; Lohaus, G. *Justus Liebigs Ann. Chem.* **1974**, 901-920.

¹⁵⁰ Albert, R.; Dax, K.; Link, R. W.; Stütz, A. E. *Carbohydr. Res.* **1983**, *118*, C5-C6.



Scheme 110: Lattrell-Dax epimerization.

Epimerization of the 2-hydroxyl group of **31** was accomplished through a two-step procedure to afford the desired diastereomer **74**. The 2-OH of **31** was first converted to a good leaving group 2-*O*-triflate. Treatment of **31** with 2 equivalents of Tf₂O in a mixture of CH₂Cl₂/pyridine (40: 1, V/V) at -40 °C for 1 h 30 min, gave the corresponding 2-*O*-triflate intermediate. After work up, the freshly prepared triflate derivative was treated with 10 equivalents of sodium nitrite in DMF/H₂O (1/1, V/V), after stirring the mixture at room temperature for one night, affording the corresponding diastereomer **74** in 44% yield (Scheme 111).



Scheme 111: Synthesis of **74** through Lattrell-Dax epimerization.

In order to confirm the configuration of compound **74**, the ¹H and ¹³C NMR spectra of **31** and **74** were compared in Figure 62. It was shown that the H^{II} in both of compounds **31** and **74** appeared as a multiple due to the coupling with two vicinal CH₂, while its chemical shift moved to lower δ value, from δ 3.83-3.77 ppm in **31** to δ 3.79-3.73 ppm in **74**. Moreover, the chemical shift difference between the diastereotopic protons (H^{III}a: δ 3.84 ppm, *J* = 10.1, 4.6 Hz; H^{III}b: δ 3.66 ppm, *J* = 10.1, 5.5 Hz) in compound **74** was consistently larger compared to **31** (H^{III}: δ 3.77-3.72 ppm). A very small variation was observed in the chemical shift of C^{III} on ¹³C NMR spectra of **31** and **74**. The C^{III} moved slightly to upfield from δ 71.7 ppm in **31** to δ 71.4 ppm in **74**.

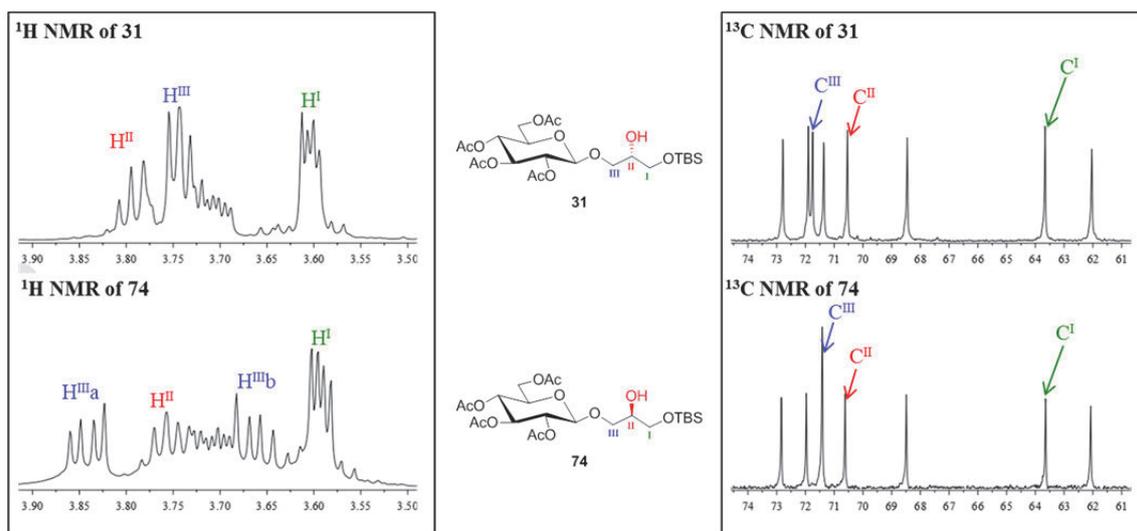
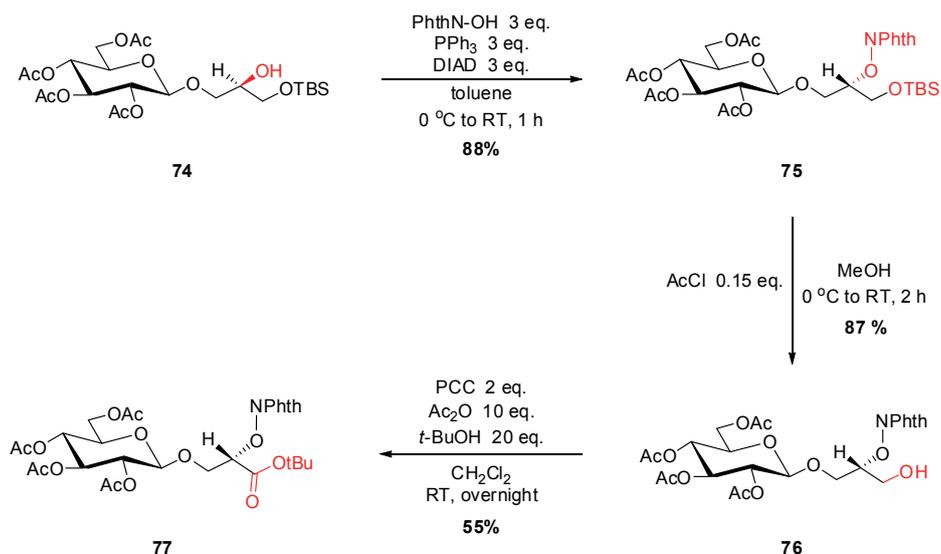


Figure 62: ^1H and ^{13}C NMR spectra of compounds **31** and **74** in CDCl_3 .

From the obtained diastereomer **74**, the synthesis of (2*S*)-3-*O*-glycosyl aminoxy ester **77** was carried out in three steps (Scheme 112). The aminoxy function was first introduced into the C-2 position through Mitsunobu reaction. Treatment of compound **74** with PhthN-OH (3 eq.) in toluene in the presence of PPh_3 (3 eq.) at 0°C , followed by slow dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds) with the appearance of a dark red coloring, led successfully to the 2-*O*-phthalimidooxy derivative **75** with inversion of configuration in 88% yield (Scheme 112).

Subsequently, to remove efficiently the TBS group without deprotecting acetyl groups, compound **75** was treated with catalytic AcCl (0.15 eq.) in MeOH for 2 h (monitored by TLC) and then neutralized with 5% NaHCO_3 to afford the corresponding alcohol **76** in 87% yield after purification (Scheme 112).

Finally, oxidation of the primary alcohol with PCC (2 eq.) in the presence of Ac_2O (10 eq.) and *t*-BuOH (20 eq.) in CH_2Cl_2 at room temperature for one night, led successfully to the (2*S*)-3-*O*-glycosyl aminoxy ester **77** in 55% yield (Scheme 112).



Scheme 112: Synthesis of (2*S*)-3-*O*-glycosyl aminoxy ester **77**.

The ^1H and ^{13}C NMR values for all the protons and carbons of compounds **73** and **77** were assigned on the basis of Dept-135, COSY, and HETCOR spectra. Comparison of the ^1H NMR spectra of compounds **73** and **77** in CDCl_3 revealed the chemical shift of H-2 moved to higher field, from δ 4.93 ppm in **73** to δ 4.77 ppm in **77**, while the anomeric proton H-1' moved to lower field, from δ 4.63 ppm in **73** to δ 5.00-4.97 ppm in **77** (Figure 63). Moreover, there were also different degrees of downfield shifts of the signals of H-3', 4', 5' on the sugar frame (H-3': from δ 5.21 ppm in **73** to δ 5.29-5.20 ppm in **77**; H-4': from δ 5.07 ppm in **73** to δ 5.10 ppm in **77**; H-5': from δ 3.74 ppm in **73** to δ 3.83 ppm in **77**). It was noted that the H-3' of **73** appeared as a *t* ($J = 9.6$ Hz) at 5.21 ppm, while the H-3' of **77** appeared as a multiple. From the compared ^{13}C NMR spectra of **73** and **77**, we can observe that the chemical shifts of C-1', 2, 5' in **73** and **77** moved very slightly (Figure 64).

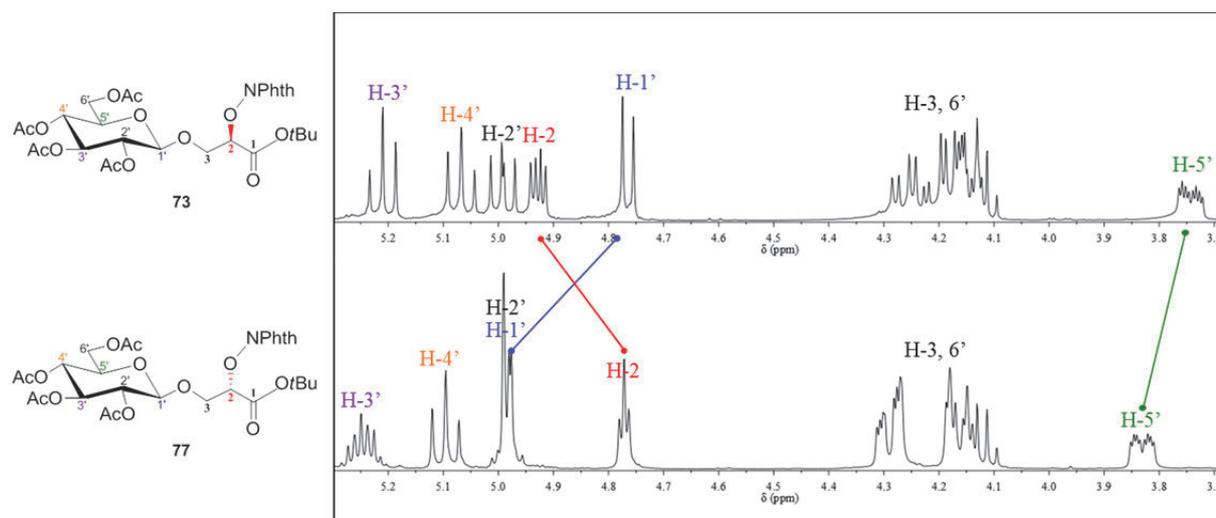


Figure 63: ^1H NMR spectra of compounds **73** and **77** in CDCl_3 .

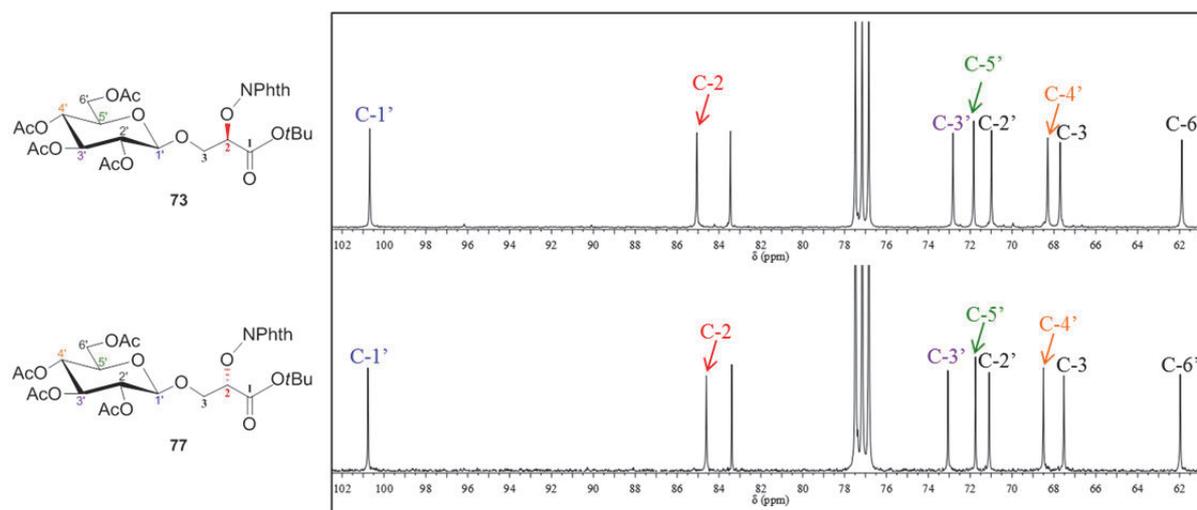


Figure 64: ^{13}C NMR spectra of compounds **73** and **77** in CDCl_3 .

In a previous work of our group,¹¹⁸ we have synthesized the (2*S*)- and (2*R*)-*C*-glucosyl aminoxy ester as novel glycosyl amino acid building blocks from *C*-allyl glycoside. Based on ^1H NMR spectroscopic data of these two compounds, we had observed that the CH-2 of the (2*S*)-*C*-glucosyl aminoxy ester appeared as a *t* ($J = 6.0$ Hz) at 4.13 ppm, while the CH-2 of (2*R*)-*C*-glucosyl aminoxy ester appeared as a *dd* ($J = 2.8$ and 11.0 Hz) at 4.16 ppm (Figure 65).

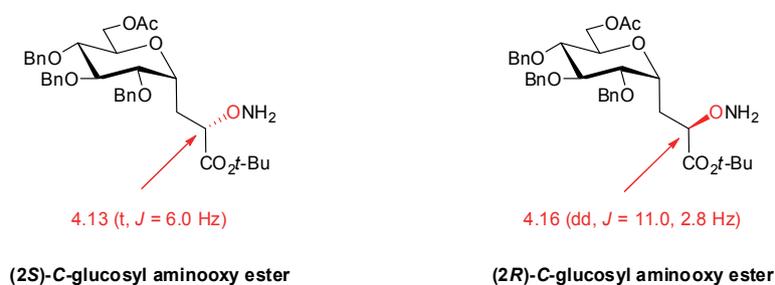


Figure 65: Structures of *C*-glucosyl aminoxy esters.¹¹⁸

In our case, from the ^1H NMR spectra of compounds **73** and **77**, we observed the similar coupling multiplets of the CH-2. The CH-2 of the (2*S*)-3-*O*-glucosyl aminoxy ester **77** appeared as a *t* ($J = 3.4$) at 4.77 ppm, while the CH-2 of the (2*R*)-3-*O*-glucosyl aminoxy ester **73** appeared as a *dd* ($J = 3.5$ and 7.0 Hz) at 4.93 ppm (Figure 66). These results help us to confirm the configuration of *C*-2 stereogenic center of the obtained *O*-glucosyl aminoxy esters, and may be useful for suggesting the configuration of this type of compounds.

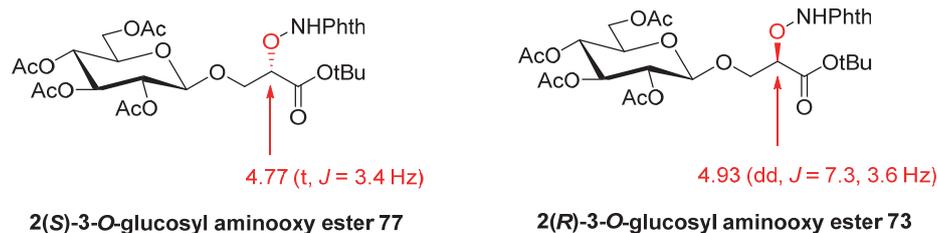
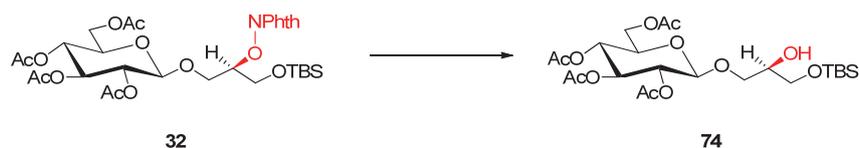


Figure 66: Structures of (2*S*)- and (2*R*)-3-*O*-glucosyl aminoxy esters.

I.3.b Epimerization based on Mitsunobu reaction

The Mitsunobu reaction was often used for the inversion of configuration in secondary alcohols using DEAD or DIAD and PPh₃, which has been described in Chapter 2.⁶⁹ The reaction proceeds with clean stereochemical inversion, and the reaction conditions are compatible with many functional groups, which make it a popular method for the inversion of stereogenic center.

In Chapter 4, the (2*S*)-2-*O*-phthalimidooxy derivative **32** has been prepared. We tried to cleave the O-N bond to obtain the compound **74** (Scheme 113). Compound **32** was first treated with 0.03 equivalent of CuSO₄·5H₂O in MeOH,¹⁵¹ but no reaction was observed on the TLC after stirring the mixture at room temperature overnight (Entry 1, Table 10). Treatment of **32** with 0.15 equivalent of azobisisobutyronitrile (AIBN) and 1.1 equivalents of tri-*n*-butyltin hydride (*n*-Bu₃SnH) in benzene under reflux for 2 h led to the degradation of **32** (Entry 2).¹⁵² Finally, hydrogenolysis of **32** (35 mg) with 10% palladium on carbon (35 wt %) in MeOH^{125,153} at room temperature afforded **74** in 54% yield (Entry 3).



Scheme 113: Conversion of compound **32** to **74**.

Entry	Conditions	Results
1	CuSO ₄ ·5H ₂ O (0.03 eq.) in MeOH, RT, overnight	No reaction
2	AIBN (0.15 eq.) and <i>n</i> -Bu ₃ SnH (1.1 eq.) in benzene, Reflux, 2 h	Degradation
3	10% Pd/C (35 wt %) and H ₂ in MeOH, RT, overnight	54% of 74

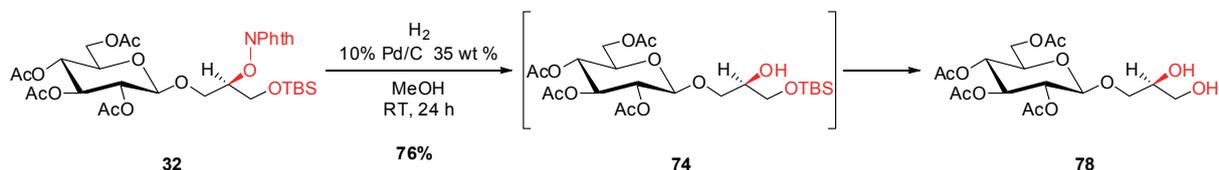
Table 10: Attempted conditions for the synthesis of compound **74**.

¹⁵¹ Momiyama, N.; Yamamoto, H. *J. Am. Chem. Soc.* **2003**, *125*, 6038-6039.

¹⁵² Kim, S.; Lee, T. A.; Song, Y. *Synlett* **1998**, 471-472.

¹⁵³ Zhang, P.; Li, C.; Yang, H.; Wei, H.; Xia, Z.; Ma, D.; Chen, H.; Wang, K.; Li, X. *Carbohydr. Res.* **2014**, *398*, 36-39.

Compound **74** obtained from **32** showed exactly the same spectroscopic data as the compound obtained using the Lattrell-Dax method. When we enlarged the scale of this reaction to 1.4 g, the TLC showed the presence of compound **74**. However, the TBS group was lost during the work up and column chromatography to give the compound **78** in 76% yield (**Scheme 114**).



Scheme 114: Hydrogenolysis of compound **32** in large scale.

The compound **78** has been fully characterized by ^1H , ^{13}C NMR and HRMS. The (2*S*)-configuration of **78** was confirmed by comparing the NMR spectra with compound **30** (Figure 67). It was clearly observed from ^1H NMR spectra that the chemical shift differences between H^{IIIa} and H^{IIIb} in compound **78** is much larger than that of **30**. No big difference was observed in the chemical shifts of H^{I} and H^{II} between compounds **78** and **30**. As for the *O*-TBS derivatives **31** and **74**, there is also a slight upfield shift of the signal of C^{III} , from δ 72.6 ppm in **30** to δ 71.9 ppm in **78**.

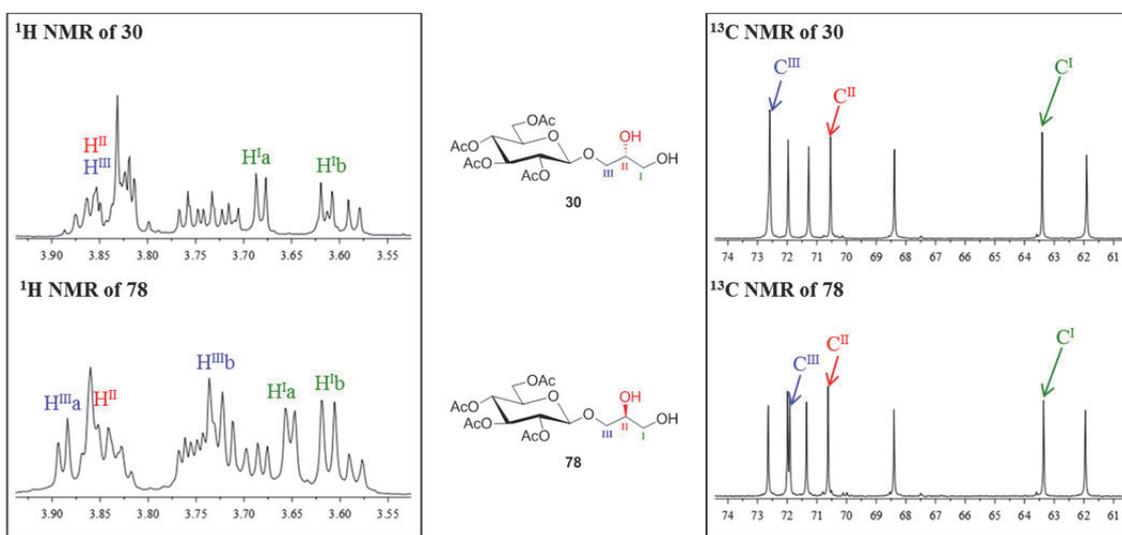
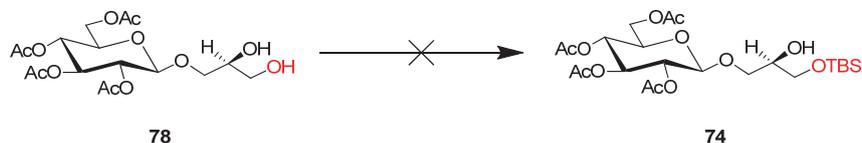


Figure 67: ^1H and ^{13}C NMR spectra of compounds **30** and **78** in CDCl_3 .

In Chapter 4, the regioselective silylation of compound (2*R*)-glucoglycerol **30** was realized by using 1.2 equivalents of TBSCl in THF, promoted by *N*-methylimidazole (3 eq.) and iodine (3 eq.), at 0 °C for 20 min. However, no reaction was observed when the isomer **78** was

treated in the same conditions. Addition of more TBSCl (1.5 eq.) was not helpful for this reaction (Entry 1, Table 11). Treatment of **78** with TBSCl (1.2 eq.) and DMAP (0.2 eq.) in pyridine at room temperature for 16 h failed again to give compound **74** (Entry 2).



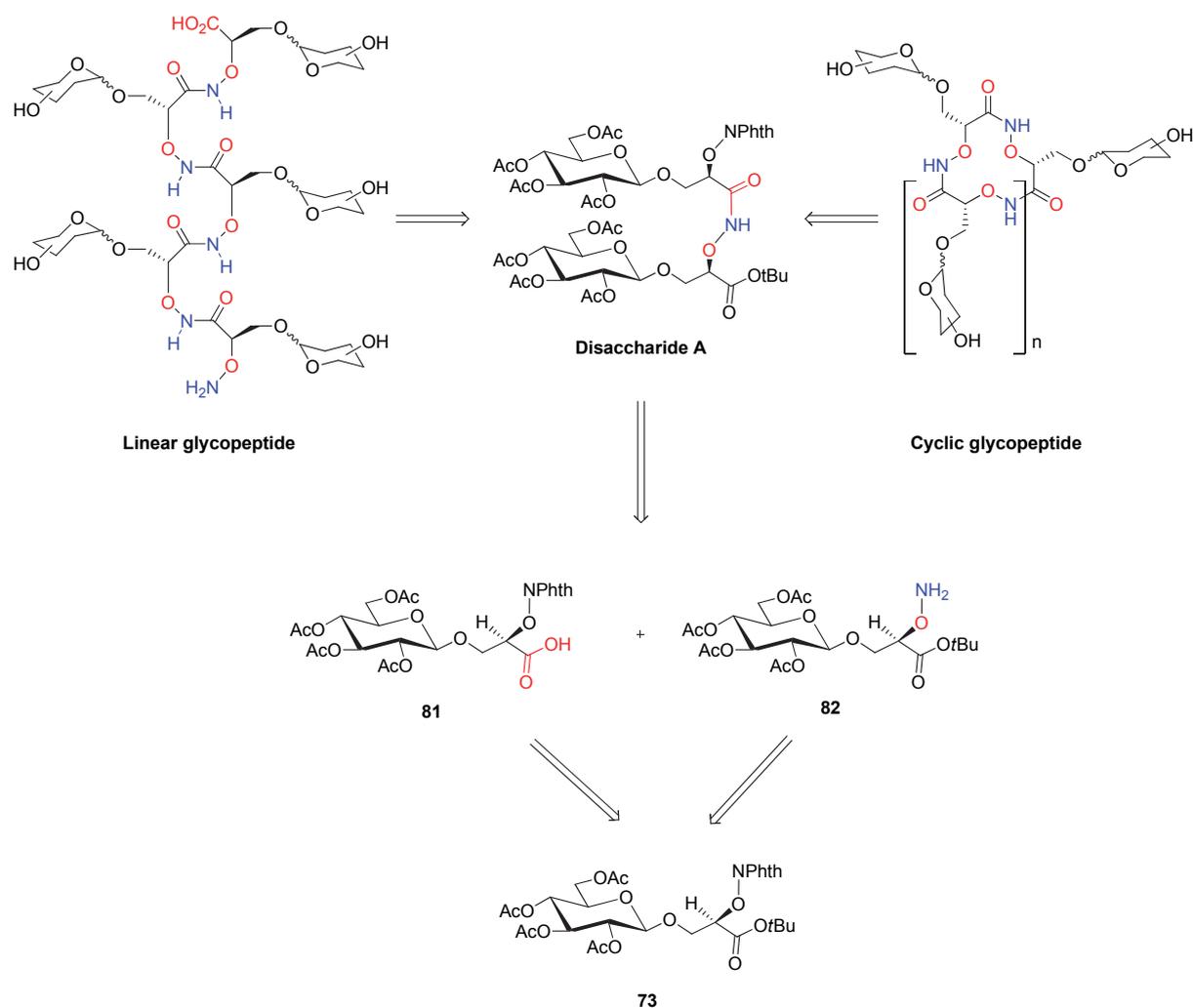
Entry	Conditions	Results
1	TBSCl (1.2 eq.), <i>N</i> -methylimidazole (3 eq.) and iodine (3 eq.) in THF, 0 °C for 20 min, then TBSCl (1.5 eq.) was added, RT, 2 days	No reaction
2	TBSCl (1.2 eq.) and DMAP (0.2 eq.) in pyridine, RT, 16 h	No reaction

Table 11: Attempted regioselective silylation of compound **78**.

Due to the failed protection of compound **78** using TBS group, we tried to protect the primary alcohol of **78** with triphenylmethyl (Tr) group. Hopefully, treatment of **78** with 1.2 equivalents of TrCl in pyridine under reflux for 24 h led to the compound **79** in a corrected yield of 92% (Conversion: 62%) (Scheme 115).

The regioselectively tritylated compound **79** allowed the introduction of aminoxy group onto the C-2 position through Mitsunobu reaction. Treatment of compound **79** with PhthNOH (3 eq.) in toluene in the presence of PPh₃ (3 eq.) at 0 °C, followed by slow dropwise addition of 3 equivalents of DIAD (1 drop every 5 seconds) with the appearance of a dark red coloring, led successfully to the 2-*O*-phthalimidooxy derivative **80** with inversion of configuration in 97% yield (Scheme 115).

Finally, detritylation of compound **80** on treatment with catalytic AcCl (0.15 eq.) in MeOH at 0 °C to room temperature for 5 h furnished the corresponding alcohol **76** in 71% yield (Scheme 115).

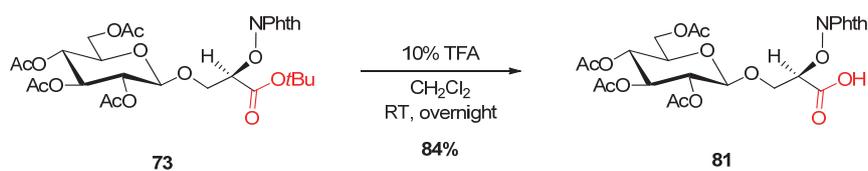


Scheme 116: Retrosynthesis of the *N*-oxyamide-linked linear and cyclic glycopeptides.

III.2 Synthesis of *N*-oxyamide-linked glycopeptides

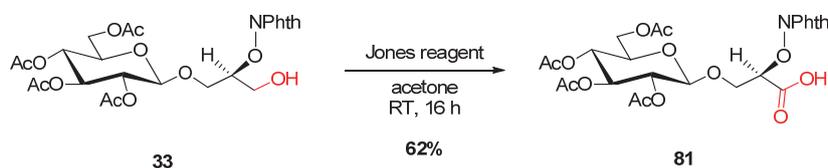
III.2.a Attempted synthesis of *N*-oxyamide-linked disaccharide A

In order to be used in the following coupling reactions, the protecting group of compound **73** was selectively removed. Firstly, removal of the *t*-Bu group of compound **73** with 10% TFA in CH_2Cl_2 at room temperature gave the carboxylic acid derivative **81** in 84% yield (Scheme 117).



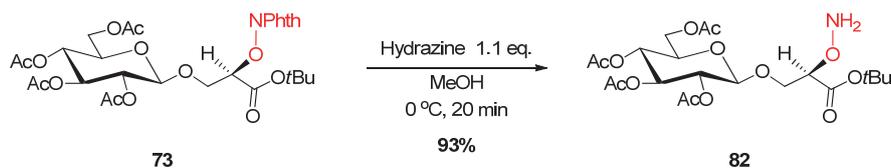
Scheme 117: Hydrolysis of **73** to prepare **81**.

The carboxylic acid derivative **81** could also be synthesized by oxidation of the (2*S*)-1-*O*-glycosyl phthalimidooxy alcohol **33** with Jones reagent.¹⁵⁴ Compound **33** was treated with Jones reagent (3 eq.) at room temperature for 16 h, affording compound **81** in 62% yield (Scheme 118).



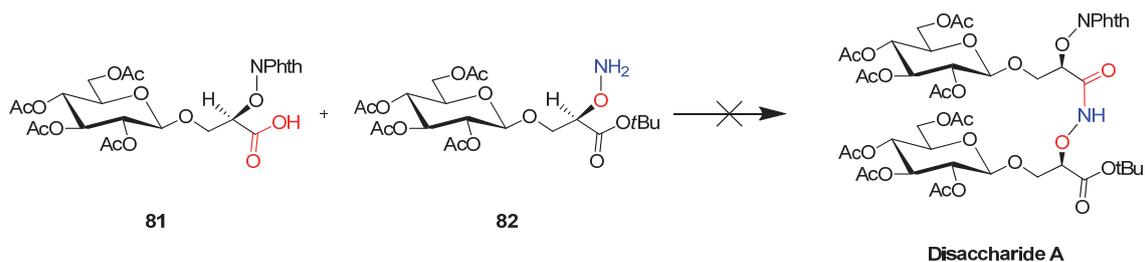
Scheme 118: Oxidation of **33** to prepare **81**.

Hydrazinolysis of **80** with hydrazine (1.1 eq.) under mild conditions (0 °C, 20 min) deprotected selectively the phthaloyl group to give the oxyamine derivative **82** in 93% yield (Scheme 119). It is worthy noted that the purification of **82** by chromatography may lead its partial decomposition due to the instability of O-NH₂ function in silica. A fast column chromatography was necessary to obtain the pure product.



Scheme 119: Deprotection of the oxyamine of **73**.

With the two monomers in hand, we were interested in the preparation of disaccharide **A** by coupling the carboxylic acid **81** with oxyamine **82** (Scheme 120). The carboxylic acid **81** was reacted with equimolar quantity of the oxyamine **82** in a mixture of DMF/CH₂Cl₂ (1/2, V/V) at 0 °C in the presence of EDC·HCl (1.8 eq.) and HOBT (1.8 eq.). After stirring the mixture at room temperature for 24 h, no reaction was observed on the TLC (Entry 1, Table 12). Addition of more coupling reagent failed again (Entry 2). Under the coupling conditions of DCC (2 eq.), HOBT (2 eq.) and Et₃N (2 eq.) in CH₂Cl₂ at 0 °C to room temperature for 24 h, the reaction led to the degradation of starting materials (Entry 3).



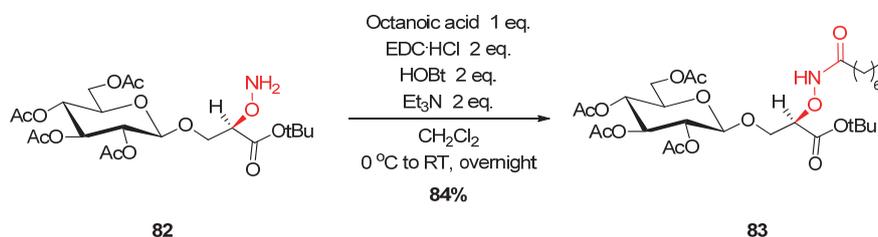
¹⁵⁴ Harding, K. E.; May, L. M.; Dick, K. F. *J. Org. Chem.* **1975**, *40*, 1664-1665.

Scheme 120: Attempted synthesis of the disaccharide A.

Entry	Coupling conditions	Results
1	EDC·HCl (1.8 eq.), HOBT (1.8 eq.) in DMF/CH ₂ Cl ₂ (1/2, V/V), 0 °C to RT, 24 h	No reaction
2	EDC·HCl (2 eq.), HOBT (2 eq.) and Et ₃ N (2 eq.) in CH ₂ Cl ₂ , 0 °C to RT, 24 h, then EDC·HCl (2 eq.), HOBT (2 eq.), Et ₃ N (2 eq.), reflux, 24 h	No reaction
3	DCC (1.2 eq.), HOBT (1.2 eq.) and Et ₃ N (2 eq.) in CH ₂ Cl ₂ , 0 °C to RT, 24 h	Degradation

Table 12: Coupling conditions for preparing the disaccharide A.

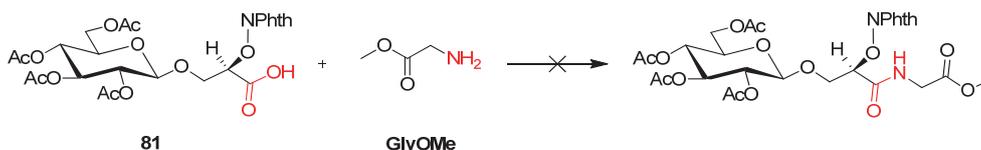
In order to understand the reason of the failure of coupling reaction, we firstly tried to couple the oxyamine **82** with a simple carboxylic acid. Coupling of the equimolar quantity of octanoic acid with the oxyamine **82** in the presence of EDC·HCl/HOBT/Et₃N in CH₂Cl₂ at 0 °C to room temperature overnight, led successfully to the glycolipid **83** in 84% yield (Scheme 121). The ¹H NMR spectrum of **83** in CDCl₃ showed the NH signal appearing at 8.88 ppm, indicating the formation of *N*-oxyamine bond.

**Scheme 121:** Coupling reaction of **82** with octanoic acid.

Then, the carboxylic acid **81** was coupled with equimolar quantity of glycine methyl ester (GlyOMe) (Scheme 122). As shown in Table 13, reaction of **81** and GlyOMe·HCl in DMF/CH₂Cl₂ (1/2, V/V) in the presence of EDC·HCl (1.8 eq.), HOBT (1.8 eq.) and Et₃N (2.8 eq.) in CH₂Cl₂ at 0 °C to room temperature for 2 days, led to several products which are difficult to be separated (Entry 1). When compound **81** was treated with 1.1 equivalents of isobutylchloroformate (*i*-BuOCOCl) and 3 equivalents of Et₃N in anhydrous CH₂Cl₂ at -10 °C for 10 min,¹⁵⁵ followed by addition of a solution of GlyOMe·HCl in DMF/CH₂Cl₂, a new product was obtained after 18 h stirring at room temperature (Entry 2). The NMR spectrum of the new product showed the absence of phthaloyl group, which mean that the coupling reaction

¹⁵⁵ Xie, J. *Eur. J. Org. Chem.* **2002**, 3411-3418.

failed to give the expected glycopeptide again.



Scheme 122: Coupling reaction of **81** with GlyOMe.

Entry	Conditions	Results
1	EDC·HCl (1.8 eq.), HOBT (1.8 eq.) and Et ₃ N (2.8 eq.) in DMF/CH ₂ Cl ₂ (1/2, V/V), 0 °C to RT, 2 days	Several products
2	<i>i</i> -BuOCOCl (1.1 eq.), Et ₃ N (3 eq.) in CH ₂ Cl ₂ , -10 °C to RT, 18 h ¹⁵⁵	Undesired product

Table 13: Attempted conditions for coupling **81** with GlyOMe.

From these results, we speculated that the failure of the coupling reaction of **81** with **82** or GlyOMe may be attributed to the presence of the phthaloyl group which was sterically bulky and might hide the carboxylic acid function. We decided to replace the phthaloyl group with other protecting groups to attempt the synthesis of disaccharide.

III.2.b Replacement of the Phth protecting group with Fmoc

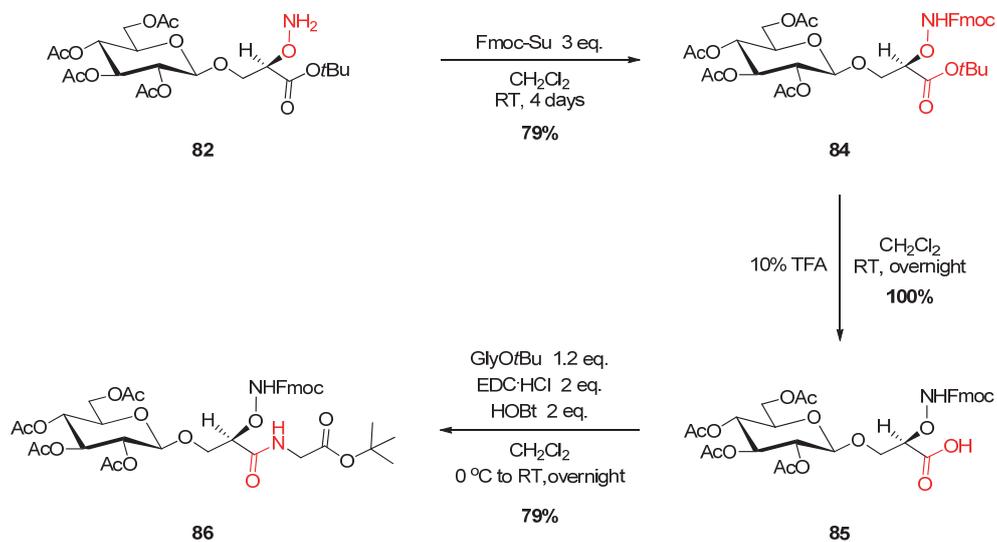
The fluorenylmethoxy carbonyl (Fmoc) protecting group is one of the widely used amino-protecting groups in peptide synthetic processes. It can be readily introduced by reacting the amine with an active 9-fluorenylmethyl carbonate, such as Fmoc-Cl, Fmoc-OSu, or Fmoc-OBt.¹⁵⁶ Most importantly, the Fmoc group has been shown to have an excellent acid stability, which allows the selective removal of other protection groups (e.g. Boc, *O**t*-Bu) in its presence. It can be easily removed under very mild basic conditions, such as 20% piperidine or 50% morpholine in DMF.¹⁵⁷

Treatment of the oxyamine **82** with 3 equivalents of Fmoc-OSu in CH₂Cl₂ at room temperature for 4 days gave the Fmoc protected compound **84** in 79% yield (Scheme 123). Subsequent hydrolysis of **84** with 10% TFA at room temperature overnight afforded the carboxylic acid **85** in 100% yield. Finally, reaction of the obtained carboxylic acid **85** with 1.2 equivalents of glycine *t*-butyl ester (GlyOtBu) in the presence of EDC·HCl (2 eq.) and HOBT (2 eq.) in CH₂Cl₂ at 0 °C to room temperature overnight, led successfully to the glycopeptide **86** in 79% yield (Scheme 123). In the ¹H NMR spectrum of **86** in CDCl₃, there were two new

¹⁵⁶ Chinchilla, R.; Dodsworth, D. J.; Nájera, C.; Soriano, J. M. *Tetrahedron Lett.* **2001**, *42*, 7579-7581.

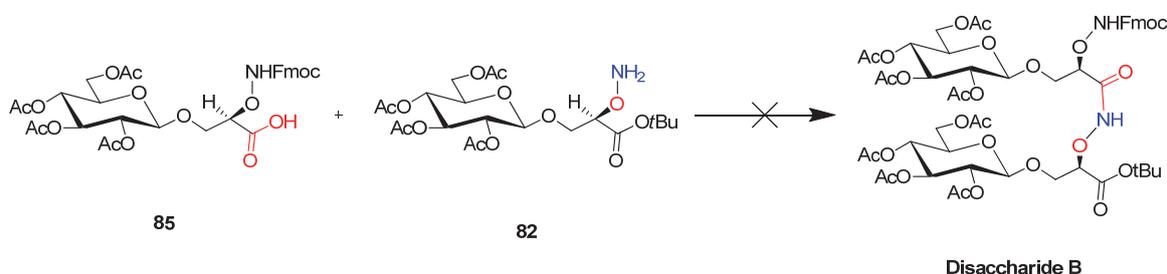
¹⁵⁷ Greene, T. W.; Wuts, P. G. M. *Protective Groups in Organic Synthesis*, 4th edition. **2006**, Chapter 7.

NH signals appearing at 8.75 and 8.32 ppm, which demonstrated the formation of the amide bond.



Scheme 123: Synthesis of the glycopeptide **86**.

Unfortunately, by using the same coupling conditions, reaction of the carboxylic acid **85** and oxycarbonyl oxycarbonyl ester **82** failed to give the desired disaccharide **B** (Scheme 124).



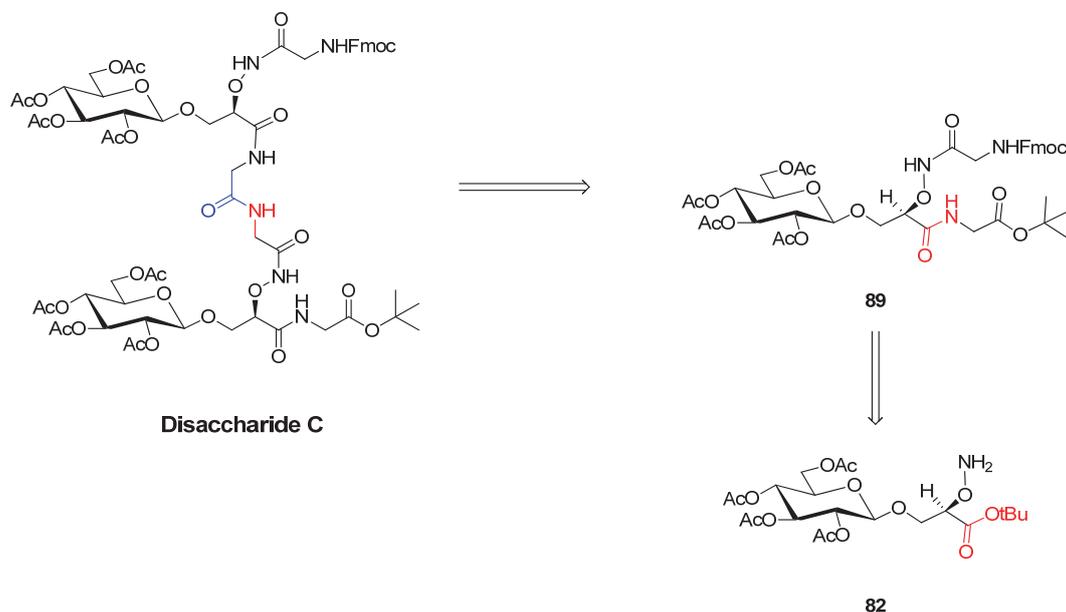
Scheme 124: Attempted synthesis of the disaccharide **B**.

In summary, the glycosyl carboxylic acid **85** can be coupled with GlyOtBu. However its reaction with the glycosyl oxycarbonyl ester **82** to disaccharide **B** was unsuccessful.

III.2.c Attempted synthesis of the *N*-oxyamide-linked disaccharide **C**

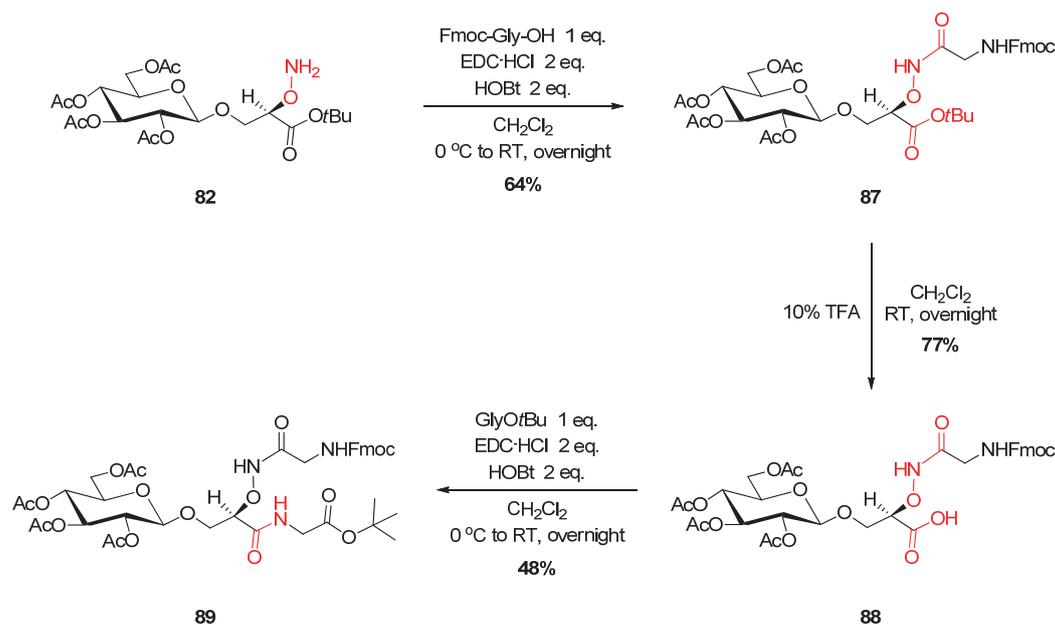
We then changed our mind to synthesize the glycosyl aminoxy ester derivative **89**, in which both of the carboxylic acid and oxycarbonyl functions were attached with an amino acid. In this structure, both of the functional carboxylic acid and amine were not adjacent to the C-2, which can decrease the steric hindrance of the coupling reaction for the synthesis of disaccharide **C** (Scheme 125). The compound **89** could be synthesized by attaching the amino acids

Fmoc-Gly-OH and GlyOtBu to the oxyamine and carboxylic acid functions of compound **82**, respectively.



Scheme 125: Retrosynthesis of the disaccharide **C**.

For the preparation of glycosyl aminoxy ester derivative **89**, the oxyamine **82** was first treated with equimolar quantity of Fmoc-Gly-OH in CH_2Cl_2 in the presence of EDC·HCl (2 eq.) and HOBT (2 eq.) at 0°C . After reaction at room temperature overnight, the compound **87** was obtained in 64% yield (Scheme 126). Subsequent hydrolysis of **87** with 10% TFA at room temperature overnight afforded the carboxylic acid **88** in 77% yield. It is interesting to notice that the oxyamide NH signal appeared at 10.5 ppm in the ^1H NMR spectrum of compound **88**, suggesting that the oxyamide NH might be involved in an intramolecular hydrogen bonding. Further studies by FT-IR and NMR titration are needed to investigate its secondary structure. Finally, coupling of the carboxylic acid **88** with equimolar quantity of GlyOtBu in the presence of EDC·HCl (2 eq.) and HOBT (2 eq.) in CH_2Cl_2 at 0°C to room temperature, led successfully to the glycopeptide **89** in 48% yield (Scheme 126). In the ^1H NMR spectrum of **89** in CDCl_3 , there were three NH signals appearing at 9.99, 8.78 and 5.56 ppm, which demonstrated the success of the coupling reaction.



Scheme 126: Synthesis of *N*-oxyamide-linked glycopeptide **89**.

Le Grel *et al.* have reported that both of the hydrazide (CO-NH-N) and *N*-oxy amide linkages (CO-NH-O) displayed the *Z/E* isomerism, and the chemical shifts of the *Z*-isomer and *E*-isomer were different.¹⁵⁸ In the ¹H NMR spectrum of Ac-NH-OMe, two signals of NH were observed, which reflected the presence of *Z*- and *E*- forms of the *N*-oxyamide bond (Figure 68).

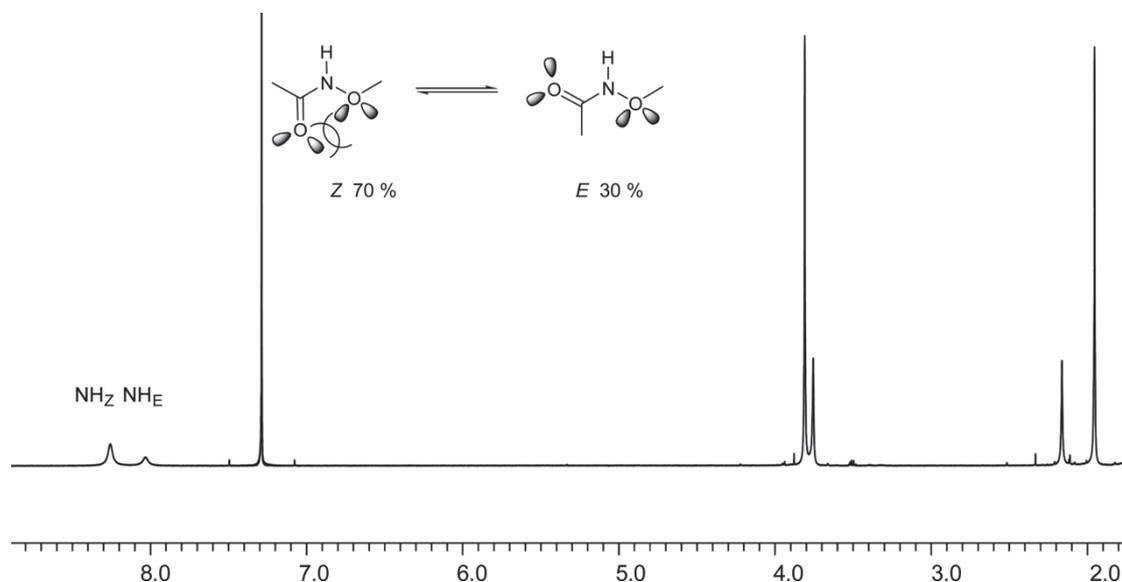


Figure 68: ¹H NMR spectrum of Ac-NH-OMe in CDCl₃.

¹⁵⁸ Le Grel, P.; Salaun, A.; Mocquet, C.; Le Grel, B.; Roisnel, T.; Potel, M. *J. Org. Chem.* **2011**, *76*, 8756-8767.

In our work, the ^1H NMR spectrum of **87** in CDCl_3 showed a similar feature. We observed two resolved O-NH-CO signals at 9.50 and 8.73 ppm, which suggest the presence of *Z/E* isomerism of compound **87** (Figure 69).

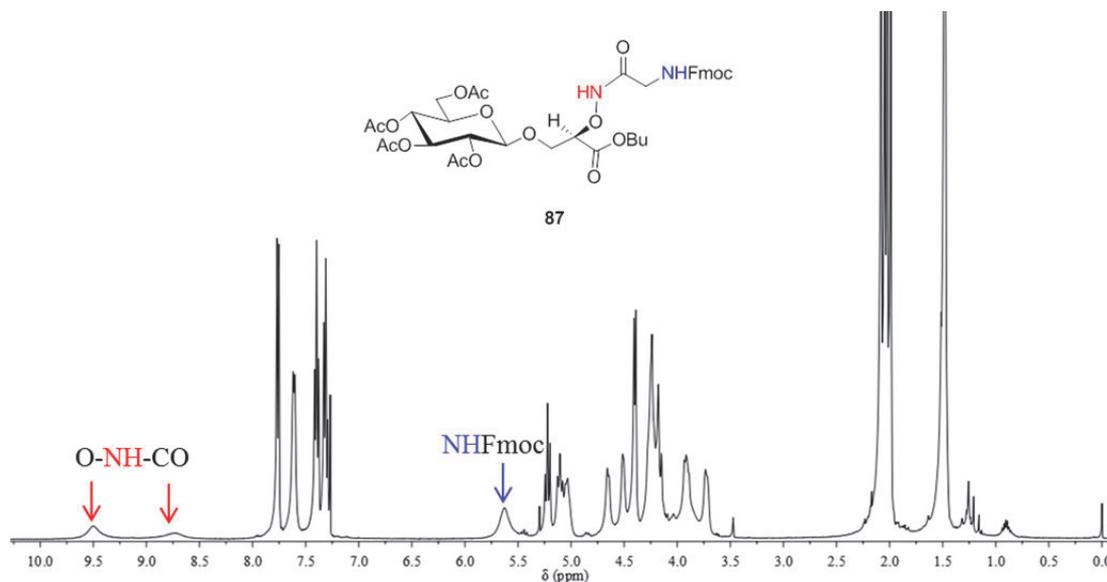
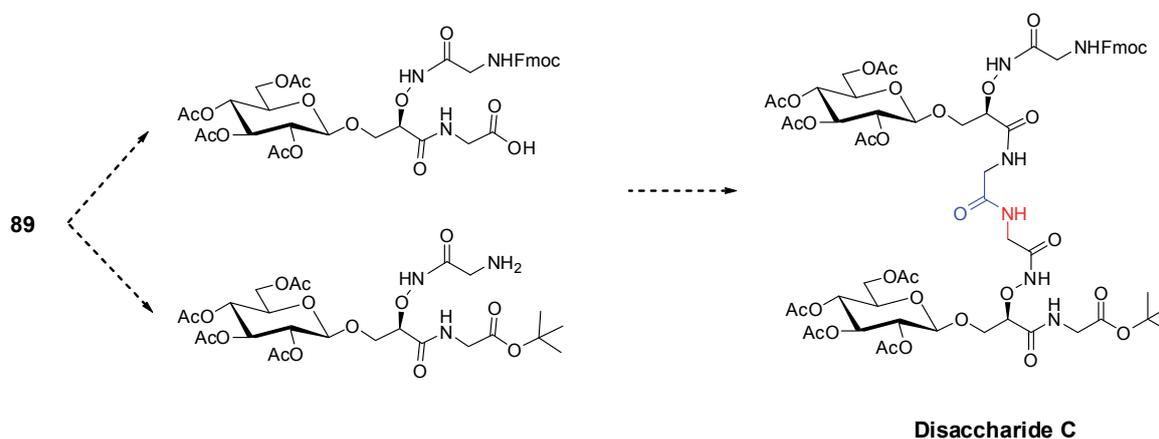


Figure 69: ^1H NMR spectrum of compound **87** in CDCl_3 .

Further transformation of **89** could lead to the disaccharide **C** (Scheme 127). This part of work is still in progress.



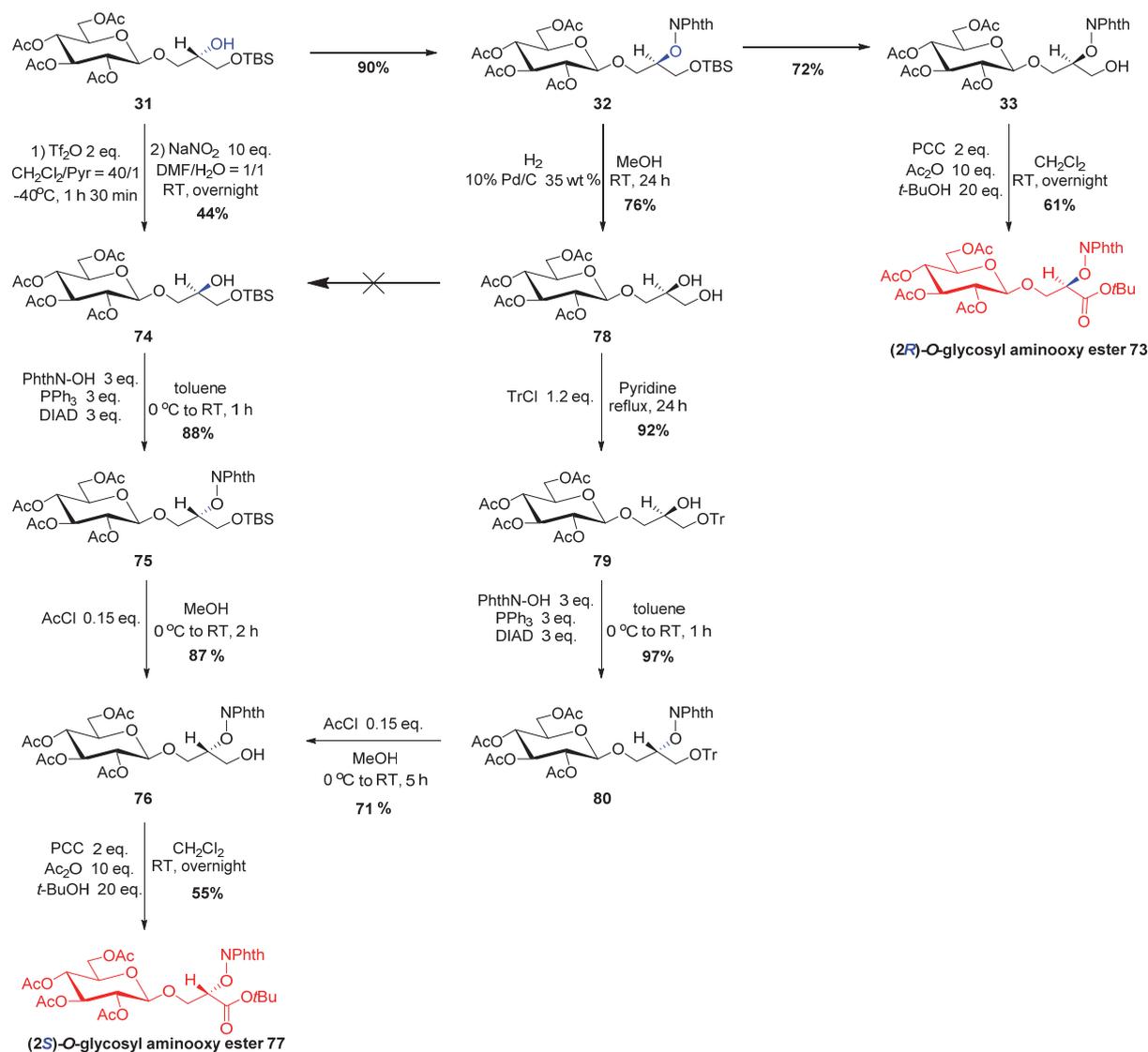
Scheme 127: Synthetic route to the disaccharide **C**.

IV Conclusion

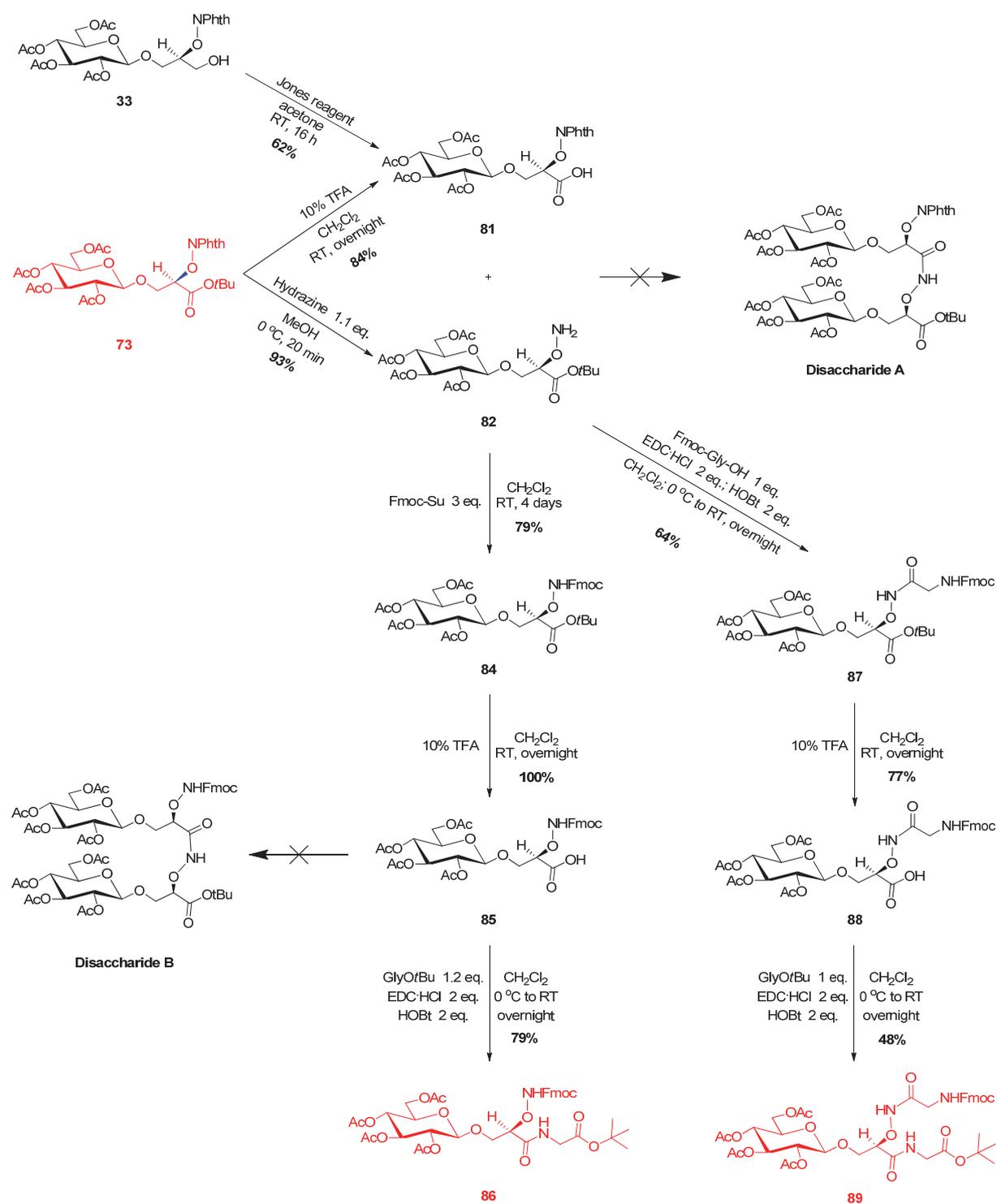
In this chapter, we have successfully prepared the (2*R*)-3- β -*O*-glycosyl aminoxy ester **73** by oxidation of the (2*S*)-1-*O*-glycosyl phthalimidooxy alcohol **33**. Synthesis of (2*S*)-3- β -*O*-glycosyl aminoxy ester **77** has been developed in two synthetic routes, which

were based on the Lattrell-Dax epimerization and Mitsunobu reactions from the same glyco-glycerol derivative **31** (Scheme 128).

As a new class of glycosyl amino acid building blocks, these multifunctionalized glyco-syl aminoxy esters could be useful for the preparation of novel glycopeptide mimics through oxime, amide, and *N*-oxy amide linkage. From the glycosyl aminoxy ester **73**, we have succeeded in the synthesis of *N*-oxyamide-linked glycopeptides **86** and **89** (Scheme 129).



Scheme 128: Synthesis of (2*R*)- and (2*S*)- β -*O*-glycosyl aminoxy esters.

Scheme 129: Synthesis of *N*-oxyamide-linked glycopeptides.

General Conclusion and Perspectives

The work reported in this dissertation is focused on the synthesis of orthogonally protected sugar and glycosyl aminoxy acids for the development of *N*-oxyamide-linked glycolipids and glycopeptides.

The first Chapter overviewed the structures and functions of glycopeptides/glycoproteins and glycolipids. Due to their potent biological implications, many efforts have been made for the development of functional glycoconjugates. The interesting properties of *N*-oxyamide linked peptides motivate us to prepare novel *N*-oxyamide-linked glycoconjugate mimics. The synthesis and properties of N-O linked carbohydrate-based compounds are reviewed in the second Chapter.

In the third Chapter, the 2,6-*O*-functionalized pyranoid glycoaminoxy ester **7** has been designed as a multifunctional building block for further derivatization to new *N*-oxyamide-linked glycolipids and oligosaccharide mimics. For the preparation of the orthogonally protected glycoaminoxy ester **7**, the carboxylic acid function was introduced onto the 2-position of the methyl α -D-glucopyranoside through the *O*-alkylation reaction and the aminoxy function onto the 6-position via the Mitsunobu reaction. The 3,4-hydroxyl groups were protected by PMB ether, as it can be readily removed without affecting other functional groups. Two lipid chains were attached on the 2- and 6-positions of **7** through *N*-oxyamide and ester bond, followed by removing the PMB group with TFA to produce the deprotected *N*-oxyamide-linked glycolipid **17**. The glycolipid **17** has also been prepared from the methyl 3,4-di-*O*-(4-methoxybenzyl)-6-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside **4**, from which another glycolipid **23** has been easily synthesized.

With a continuing interest in aminoxy glycolipids, the *N*-oxyamide-linked β -gluco-/galactoglycerolipids bearing one or two lipid chains have been prepared as novel mimics of glyco-glycerolipids and glycosphingolipids in Chapter 4. The β -glycosylglycerols **30/54** were synthesized by glycosylation of (*S*)-1,2-di-*O*-benzyl-glycerol **28** with D-glucose/D-galactose pentaacetate. The aminoxy functions were introduced through Mitsunobu reactions with the conversion of configuration to afford the 2-*O*-amino β -glyco-glycerols **33/57** in excellent yield. Introduction of lipid chains by coupling reactions and deprotection of the acetyl groups using excess amount of hydrazine, afforded the gluco- and galactoglycerolipids. The acetyl groups can also be removed by catalytic Na/MeOH, but this condition led to a transacylation reaction, producing new glycolipids. About 15 new glyco-glycerolipids have been synthesized. Their activity towards iNKT cells will be tested.

As perspectives, the sulfo-glyco-glycerolipids could be obtained by sulfation of obtained glyco-glycerolipids. The α -glyco-glycerolipids could be also prepared by using our developed methodology.

The (*2R*) and (*2S*)-3- β -*O*-glycosyl aminoxy ester **73/77** have been successfully prepared

as a new class of glycosyl amino acid building blocks, as described in Chapter 5. The (2*R*)-3- β -*O*-glycosyl aminoxy ester **73** was easily prepared by oxidation of the (2*S*)-1-*O*-glycosyl phthalimidooxy alcohol **33**. Synthesis of (2*S*)-3- β -*O*-glycosyl aminoxy ester **77** has been developed in two synthetic routes, which were based on the Lattrell-Dax epimerization and the Mitsunobu reactions from the same glyco-glycerol derivative **31**. These multifunctionalized glycosyl aminoxy esters could be useful for the preparation of novel glycopeptide mimics through oxime, amide, and *N*-oxy amide linkage. From the (2*R*)-3- β -*O*-glycosyl aminoxy ester **73**, *N*-oxyamide-linked glycopeptides **86** and **89** have been successfully synthesized. These oxyamine-functionalized glycopeptides offer possibilities for the development of novel glycopeptides mimics through *N*-oxy amide bond, oxime ligation or neoglycosylation.

References

- [1] Wolfert, M. A.; Boons, G.-J. *Nat. Chem. Boil.* **2013**, *9*, 776-784; (b) Kolarich, D.; Lepenies, B.; Seeberger, P. H. *Curr. Opin. Chem. Biol.* **2012**, *16*, 214-220; (c) Rudd, P. M.; Elliott, T.; Cresswell, P.; Wilson, I. A.; Dwek, R. A. *Science* **2001**, *291*, 2370-2376; (d) Dwek, R. A. *Chem. Rev.* **1996**, *96*, 683-720.
- [2] (a) Fernandez-Tejada, A.; Canada, F. J.; Jimenez-Barbero, J. *Chem. Eur. J.* **2015**, *21*, 10616-10628; (b) Fernandez-Tejada, A.; Canada, F. J.; Jimenez-Barbero, J. *ChemMedChem* **2015**, *10*, 1291-1295; (c) Hudak, J. E.; Bertozzi, C. R. *Chem. Biol.* **2014**, *21*, 16-37; (d) Galan, M. C.; Benito-Alifonso, D.; Watt, G. M. *Org. Biomol. Chem.* **2011**, *9*, 3598-3610; (e) Seeberger, P. H.; Werz, D. B. *Nature* **2007**, *446*, 1046-1051; (f) Freeze, H. H.; Westphal, V. *Biochimie* **2001**, *83*, 791-799; (g) Butters, T. D.; Dwek, R. A.; Platt, F. M. *Chem. Rev.* **2000**, *100*, 4683-4696.
- [3] (a) Wang, L. X.; Davis, B. G. *Chem. Sci.* **2013**, *4*, 3381-3394; (b) Cecioni, S.; Imberty, A.; Vidal, S. *Chem. Rev.* **2014**, *115*, 525-561; (c) Bertozzi, C. R.; Kiessling, L. L. *Science* **2001**, *291*, 2357-2364; (d) Herzner, H.; Reipen, T.; Schultz, M.; Kunz, H. *Chem. Rev.* **2000**, *100*, 4495-4538; (e) Davis, B. G. *J. Chem. Soc.; Perkin Trans. 1.* **1999**, 3215-3237.
- [4] Crick, F. *Nature*, **1970**, *227*, 561-563.
- [5] Seeberger, P. H. *Nat. Chem. Biol.* **2009**, *5*, 368-372.
- [6] (a) Satoh, H.; Manabe, S. *Chem. Soc. Rev.* **2013**, *42*, 4297-4309; (b) Ishiwata, A.; Lee, Y. J.; Ito, Y. *Org. Biomol. Chem.* **2010**, *8*, 3596-3608; (c) Zhu, X.; Schmidt, R. R. *Angew. Chem. Int. Ed.* **2009**, *48*, 1900-1934; (d) Demchenko, A. V. *Handbook of Chemical Glycosylation*, Wiley-VCH, Weinheim, **2008**; (e) Galonic, D. P.; Gin, D. Y. *Nature* **2007**, *446*, 1000-1007; (f) Toshima, K. *Carbohydr. Res.* **2006**, *341*, 1282-1297; (g) Demchenko, A. V. *Synlett* **2003**, 1225-1240; (h) Schmidt, R. R.; Kinzy, W. *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 21-123.
- [7] Michael, A. *Am. Chem. J.* **1879**, *1*, 305-312.
- [8] Fischer, E. *Ber. Dtsch. Chem. Ges.* **1893**, *26*, 2400-2412.
- [9] Koenigs, W.; Knorr, E. *Ber. Dtsch. Chem. Ges.* **1901**, *34*, 957-981.
- [10] (a) Frihed, T. G.; Bols, M.; Pedersen, C. M. *Chem. Rev.* **2015**, *115*, 4963-5013; (b) Mydock, L. K.; Demchenko, A. V. *Org. Biomol. Chem.* **2010**, *8*, 497-510.
- [11] (a) Wang, Y.; Ye, X.-S.; Zhang, L.-H. *Org. Biomol. Chem.* **2007**, *5*, 2189-2200; (b) Koeller, K. M.; Wong, C.-H. *Chem. Rev.* **2000**, *100*, 4465-4493.
- [12] (a) Seeberger, P. H.; Haase, W.-C. *Chem. Rev.* **2000**, *100*, 4349-4394; (b) Plante, O. J.; Palmacci, E. R.; Seeberger, P. H. *Science* **2001**, *291*, 1523-1527; (c) Larsen, K.; Thygesen, M. B.; Guillaumie, F.; Wil-lats, W. G. T.; Jensen, K. J. *Carbohydr. Res.* **2006**, *341*, 1209-1234.
- [13] (a) Cumpstey, I. *Carbohydr. Res.* **2008**, *343*, 1553-1573; (b) Jung, K.-H.; Müller, M.; Schmidt, R. R. *Chem. Rev.* **2000**, *100*, 4423-4442.
- [14] (a) Nigudkar, S. S.; Demchenko, A. V. *Chem. Sci.* **2015**, *6*, 2687-2704; (b) Cox, D. J.; Singh, G. P.; Watson, A. J. A.; Fairbanks, A. J. *Eur. J. Org. Chem.* **2014**, 4624-4642.

- [15] (a) Wulff, G.; Röhle, G. *Angew. Chem. Int. Ed. Engl.* **1974**, *13*, 157-170; (b) Wulff, G.; Röhle, G. *Angew. Chem.* **1974**, *86*, 173-187.
- [16] Boltje, T. J.; Buskas, T.; Boons, G.-J. *Nature Chem.* **2009**, *1*, 611-622.
- [17] (a) Demchenko, A. V. *Curr. Org. Chem.* **2003**, *7*, 35-79; (b) Fairbanks, A. J. *Synlett* **2003**, 1945-1958.
- [18] Boons, G.-J. *Contemp. Org. Synth.* **1996**, *3*, 173-200.
- [19] Gamblin, D. P.; Scanlan, E. M.; Davis, B. G. *Chem. Rev.* **2009**, *109*, 131-163.
- [20] (a) Oman, T. J.; Boettcher, J. M.; Wang, H.; Okalibe, X. N.; van der Donk, W. A. *Nat. Chem. Biol.* **2011**, *7*, 78-80; (b) Stepper, J.; Shastri, S.; Loo, T. S.; Preston, J. C.; Novak, P.; Man, P.; Moore, C. H.; Havlicek, V.; Patchett, M. L.; Norris, G. E. *Febs. Lett.* **2011**, *585*, 645-650.
- [21] Westerlind, U. *Beilstein J. Org. Chem.* **2012**, *8*, 804-818.
- [22] Ashford, P. A.; Bew, S. P. *Chem. Soc. Rev.* **2012**, *41*, 957-978.
- [23] Pratt, M. R.; Bertozzi, C. R. *Chem. Soc. Rev.* **2005**, *34*, 58-68.
- [24] (a) Aussedat, B.; Vohra, Y.; Park, P. K.; Fernández-Tejada, A.; Alam, S. M.; Dennison, S. M.; Jaeger, F. H.; Anasti, K.; Stewart, S.; Blinn, J. H.; Liao, H.-X.; Sodroski, J. G.; Haynes, B. F.; Danishefsky, S. J. *J. Am. Chem. Soc.* **2013**, *135*, 13113-13120; (b) Alam, S. M.; Dennison, S. M.; Aussedat, B.; Vohra, Y.; Park, P. K.; Fernández-Tejada, A.; Stewart, S.; Jaeger, F. H.; Anasti, K.; Blinn, J. H. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 18214-18219.
- [25] (a) Miller, S. M.; Simon, R. J.; Ng, S.; Zuckermann, R. N.; Kerr, J. M.; Moos, W. H. *Bioorg. Med. Chem. Lett* **1994**, *4*, 2657-2662; (b) Saha, U. K.; Roy, R. *Tetrahedron Lett.* **1995**, *36*, 3635-3638.
- [26] Seo, J.; Michaelian, N.; Owens, S. C.; Dashner, S. T.; Wong, A. J.; Barron, A. E.; Carrasco, M. R. *Org. Lett.* **2009**, *11*, 5210-5213.
- [27] Szekely, T.; Roy, O.; Faure, S.; Taillefumier, C. *Eur. J. Org. Chem.* **2014**, 5641-5657.
- [28] Galan, M. C.; Dumy, P.; Renaudet, O. *Chem. Soc. Rev.* **2013**, *42*, 4599-4612.
- [29] Pawar, S. A.; Jabgunde, A. M.; Maguire, G. E.; Kruger, H. G.; Sayed, Y.; Soliman, M. E.; Dhavale, D. D.; Govender, T. *Eur. J. Med. Chem.* **2013**, *60*, 144-154.
- [30] (a) Crucho, C. I.; Correia-da-Silva, P.; Petrova, K. T.; Barros, M. T. *Carbohydr. Res.* **2015**, *402*, 124-132; (b) Wu, C. Y.; Wong, C. H. *Chem. Commun.* **2011**, *47*, 6201-6207; (c) Gamblin, D. P.; Scanlan, E. M.; Davis, B. G. *Chem. Rev.* **2009**, *109*, 131-163; (d) Liu, L.; Bennett, C. S.; Wong, C. H. *Chem. Commun.* **2006**, 21-33; (e) Davis, B. G. *Chem. Rev.* **2002**, *102*, 579-602; (f) Sears, P.; Wong, C.-H. *Science* **2001**, *291*, 2344-2350; (g) Seitz, O. *Chem. Bio. Chem.* **2000**, *1*, 214-246; (h) St Hilaire, P. M.; Meldal, M. *Angew. Chem. Int. Ed.* **2000**, *39*, 1162-1179.
- [31] (a) Taylor, C. M. *Tetrahedron* **1998**, *54*, 11317-11362; (b) Arsequell, G.; Valencia, G. *Tetrahedron: Asymmetry* **1999**, *10*, 3045-3094.
- [32] Kaneshiro, C. M.; Michael, K. *Angew. Chem. Int. Ed.* **2006**, *45*, 1077-1081.
- [33] Hashim, R.; Sugimura, A.; Minamikawa, H.; Heidelberg, T. *Liq. Cryst.* **2012**, *39*, 1-17.
- [34] Brandenburg, K.; Holst, O. **2015**, Glycolipids: Distribution and Biological Function. eLS. 1-10.
- [35] IUPAC-IUB, Joint Commission on Biochemical Nomenclature (JCBN). *Carbohydr. Res.* **1998**, *312*, 167-175.
- [36] Malhotra, R. *Biochem. Anal. Biochem.* **2012**, *1*, 108.

- [37] Maeda, N.; Matsubara, K.; Yoshida, H.; Mizushina, Y. *Mini-Rev. Med. Chem.* **2011**, *11*, 32-38.
- [38] Murakami, C.; Kumagai, T.; Hada, T.; Kanekazu, U.; Nakazawa, S.; Kamisuki, S.; Maeda, N.; Xu, X.; Yoshida, H.; Sugawara, F.; Sakaguchi, K.; Mizushina, Y. *Biochem. Pharmacol.* **2003**, *65*, 259-267.
- [39] (a) Maeda, N.; Hada, T.; Yoshida, H.; Mizushina, Y. *Curr. Med. Chem.* **2007**, *14*, 955-967; (b) Maeda, N.; Kokai, Y.; Ohtani, S.; Sahara, H.; Hada, T.; Ishimaru, C.; Kuriyama, I.; Yonezawa, Y.; Iijima, H.; Yoshida, H. *Nutr. Cancer* **2007**, *57*, 216-223.
- [40] Dai, J. Q.; Zhu, Q. X.; Zhao, C. Y.; Yang, L.; Li, Y. *Phytochemistry* **2001**, *58*, 1305-1309.
- [41] Zhou, B.-N.; Tang, S.; Johnson, R. K.; Mattern, M. P.; Lazo, J. S.; Sharlow, E. R.; Harich, K.; Kingston, D. G. I. *Tetrahedron* **2005**, *61*, 883-887.
- [42] Williams, D. E.; Sturgeon, C. M.; Roberge, M.; Andersen, R. J. *J. Am. Chem. Soc.* **2007**, *129*, 5822-5823.
- [43] Holzl, G.; Dormann, P. *Prog. Lipid. Res.* **2007**, *46*, 225-243.
- [44] Zhang, J.; Li, C.; Yu, G.; Guan, H. *Marine Drugs* **2014**, *12*, 3634-3659.
- [45] Sun, Y.; Zhang, J.; Li, C.; Guan, H.; Yu, G. *Carbohydr. Res.* **2012**, *355*, 6-12.
- [46] Li, C.; Sun, Y.; Zhang, J.; Zhao, Z.; Yu, G.; Guan, H. *Carbohydr. Res.* **2013**, *376*, 15-23.
- [47] Zhang, J.; Sun, Y.; Wang, W.; Zhang, X.; Li, C.; Guan, H. *Carbohydr. Res.* **2013**, *381*, 74-82.
- [48] Vetro, M.; Costa, B.; Donvito, G.; Arrighetti, N.; Cipolla, L.; Perego, P.; Compostella, F.; Ronchetti, F.; Colombo, D. *Org. Biomol. Chem.* **2015**, *13*, 1091-1099.
- [49] (a) D'Angelo, G.; Capasso, S.; Sticco, L.; Russo, D. *FEBS J.* **2013**, *280*, 6338-6353; (b) Wu, D.; Fujio, M.; Wong, C.-H. *Bioorg. Med. Chem.* **2008**, *16*, 1073-1083.
- [50] Natori, T.; Koezuka, Y.; Higa, T. *Tetrahedron Lett.* **1993**, *34*, 5591-5592.
- [51] Harouse, J. M.; Bhat, S.; Spitalnik, S. L.; Laughlin, M.; Stefano, K.; Silberberg, D. H.; Gonzalez-Scarano, F. *Science* **1991**, *253*, 320-323.
- [52] Morita, M.; Motoki, K.; Akimoto, K.; Natori, T.; Sakai, T.; Sawa, E.; Yamaji, K.; Koezuka, Y.; Kobayashi, E.; Fukushima, H. *J. Med. Chem.* **1995**, *38*, 2176-2187.
- [53] Banchet-Cadeddu, A.; Henon, E.; Dauchez, M.; Renault, J. H.; Monneaux, F.; Haudrechy, A. *Org. Biomol. Chem.* **2011**, *9*, 3080-3104.
- [54] Bi, J.; Wang, J.; Zhou, K.; Wang, Y.; Fang, M.; Du, Y. *ACS Med. Chem. Lett.* **2015**, *6*, 476-480.
- [55] Aspeslagh, S.; Li, Y.; Yu, E. D.; Pauwels, N.; Trappeniers, M.; Girardi, E.; Decruy, T.; Van Beneden, K.; Venken, K.; Drennan, M.; Leybaert, L.; Wang, J.; Franck, R. W.; VanCalenbergh, S.; Zajonc, D. M.; Elewaut, D. *EMBO J.* **2011**, *30*, 2294-2305.
- [56] Guillaume, J.; Pauwels, N.; Aspeslagh, S.; Zajonc, D. M.; Elewaut, D.; Van Calenbergh, S. *Bioorg. Med. Chem.* **2015**, *23*, 3175-3182.
- [57] Garg, H.; Francella, N.; Tony, K. A.; Augustine, L. A.; Barchi, J. J.; Fantini, J.; Puri, A.; Mootoo, D. R.; Blumenthal, R. *Antivir. Res.* **2008**, *80*, 54-61.
- [58] Thota, V. N.; Brahmaiah, M.; Kulkarni, S. S. *J. Org. Chem.* **2013**, *78*, 12082-12089.
- [59] (a) Daniotti, J. L.; Vilcaes, A. A.; Demichelis, V. T.; Ruggiero, F. M.; Rodriguez-Walker, M. *Front. Oncolo.* **2013**, *3*, 306; (b) Wennekes, T.; van den Berg, R. J.; Boot, R. G.; van der Marel, G. A.; Overkleeft, H. S.; Aerts, J. M. *Angew. Chem. Int. Ed.* **2009**, *48*, 8848-8869; (c) McReynolds, K. D.;

- Gervay-Hague, J. *Chem. Rev.* **2007**, *107*, 1533-1552.
- [60] Li, X.; Wu, Y.-D.; Yang, D. *Accounts. Chem. Res.* **2008**, *41*, 1428-1438.
- [61] Chen, F.; Ma, B.; Yang, Z.-C.; Lin, G.; Yang, D. *Amino Acids* **2012**, *43*, 499-503.
- [62] Kitagawa, M.; Tomiyama, T. *J. Biochem.* **1929**, *11*, 265-271.
- [63] (a) Kitagawa, M.; Yamada, H. *J. Biochem.* **1932**, *16*, 339-350; (b) Kitagawa, M.; Monobe, S.-I. *J. Biochem.* **1933**, *18*, 333-343.
- [64] Rosenthal, G. A. *Life Sci.* **1997**, *60*, 1635-1641.
- [65] Kuehl Jr, F. A.; Wolf, F. J.; Trenner, N. R.; Peck, R. L.; Buhs, R. P.; Howe, E.; Putter, I.; Hunnewell, B. D.; Ormond, R.; Downing, G. *J. Am. Chem. Soc.* **1955**, *77*, 2344-2345.
- [66] Briggs, M. T.; Morley, J. S. *J. Chem. Soc., Perkin Trans I.* **1979**, 2138-2143.
- [67] Grochowski, E.; Jurczak, J. *Synthesis* **1976**, *10*, 682-684.
- [68] Chemla, P. *Tetrahedron Lett.* **1993**, *34*, 7391-7394.
- [69] (a) Swamy, K. C.; Kumar, N. N.; Balaraman, E.; Kumar, K. V. *Chem. Rev.* **2009**, *109*, 2551-2651; (b) Shin, I.; Lee, M.-r.; Lee, J.; Jung, M.; Lee, W.; Yoon, J. *J. Org. Chem.* **2000**, *65*, 7667-7675. (c) Mitsunobu, O.; Yamada, M.; Mukaiyama, T. *B. Chem. Soc. Jpn.* **1967**, *40*, 935-939; (c) Mitsunobu, O.; Eguchi, M. *B. Chem. Soc. Jpn.* **1971**, *44*, 3427-3430.
- [70] Palandoken, H.; Bocian, C. M.; McCombs, M. R.; Nantz, M. H. *Tetrahedron Lett.* **2005**, *46*, 6667-6669.
- [71] (a) Weller, C. E.; Huang, W.; Chatterjee, C. *ChemBioChem* **2014**, *15*, 1263-1267; (b) Sharma, C.; Thadhaney, B.; Pemawat, G.; Talesara, G. *Indian J. Chem.* **2008**, *47*, 1892-1897; (c) Canne, L. E.; Bark, S. J.; Kent, S. B. *J. Am. Chem. Soc.* **1996**, *118*, 5891-5896.
- [72] (a) Cao, S.; Tropper, F. D.; Roy, R. *Tetrahedron* **1995**, *51*, 6679-6686; (b) Andersson, M.; Oscarson, S. *Glycoconjugate. J.* **1992**, *9*, 122-125.
- [73] (a) Carrasco, M. R.; Alvarado, C. I.; Dashner, S. T.; Wong, A. J.; Wong, M. A. *J. Org. Chem.* **2010**, *75*, 5757-5759; (b) Jones, D. S.; Hammaker, J. R.; Tedder, M. E. *Tetrahedron Lett.* **2000**, *41*, 1531-1533; (c) Porter, N. A.; Caldwell, S. E.; Lowe, J. R. *J. Org. Chem.* **1998**, *63*, 5547-5554.
- [74] Choong, I. C.; Ellman, J. A. *J. Org. Chem.* **1999**, *64*, 6528-6529.
- [75] (a) Bissot, T. C.; Parry, R. W.; Campbell, D. H. *J. Am. Chem. Soc.* **1957**, *79*, 796-800; (b) Bordwell, F. G. *Accounts. Chem. Res.* **1988**, *21*, 456-463; (c) Bordwell, F. G.; Fried, H. E.; Hughes, D. L.; Lynch, T. Y.; Satish, A. V.; Whang, Y. E. *J. Org. Chem.* **1990**, *55*, 3330-3336.
- [76] (a) Peter, C.; Daura, X.; van Gunsteren, W. F. *J. Am. Chem. Soc.* **2000**, *122*, 7461-7466; (b) Wu, Y.-D.; Wang, D.-P.; Chan, K. W.; Yang, D. *J. Am. Chem. Soc.* **1999**, *121*, 11189-11196; (c) Yang, D.; Qu, J.; Li, B.; Ng, F.-F.; Wang, X.-C.; Cheung, K.-K.; Wang, D.-P.; Wu, Y.-D. *J. Am. Chem. Soc.* **1999**, *121*, 589-590.
- [77] Yang, D.; Ng, F.-F.; Li, Z.-J.; Wu, Y.-D.; Chan, K. W.; Wang, D.-P. *J. Am. Chem. Soc.* **1996**, *118*, 9794-9795.
- [78] (a) Yang, D.; Li, X.; Sha, Y.; Wu, Y. D. *Chem. Eur. J.* **2005**, *11*, 3005-3009. (b) Yang, D.; Qu, J.; Li, W.; Zhang, Y.-H.; Ren, Y.; Wang, D.-P.; Wu, Y.-D. *J. Am. Chem. Soc.* **2002**, *124*, 12410-12411; (c) Yang, D.; Li, X.; Fan, Y.-F.; Zhang, D.-W. *J. Am. Chem. Soc.* **2005**, *127*, 7996-7997.

- [79] Sharma, G. V.; Manohar, V.; Dutta, S. K.; Sridhar, B.; Ramesh, V.; Srinivas, R.; Kunwar, A. C. *J. Org. Chem.* **2010**, *75*, 1087-1094.
- [80] (a) Li, X.; Yang, D. *Chem. Commun.* **2006**, 3367-3379; (b) Yang, D.; Zhang, Y.-H.; Zhu, N.-Y. *J. Am. Chem. Soc.* **2002**, *124*, 9966-9967.
- [81] Chandrasekhar, S.; Rao, C. L.; Reddy, M. S.; Sharma, G. D.; Kiran, M. U.; Naresh, P.; Chaitanya, G. K.; Bhanuprakash, K.; Jagadeesh, B. *J. Org. Chem.* **2008**, *73*, 9443-9446.
- [82] (a) Chen, F.; Zhu, N.-Y.; Yang, D. *J. Am. Chem. Soc.* **2004**, *126*, 15980-15981; (b) Chen, F.; Song, K.-S.; Wu, Y.-D.; Yang, D. *J. Am. Chem. Soc.* **2008**, *130*, 743-755.
- [83] Munneke, S.; Prevost, J. R.; Painter, G. F.; Stocker, B. L.; Timmer, M. S. *Org. Lett.* **2015**, *17*, 624-627.
- [84] Gudmundsdottir, A. V.; Paul, C. E.; Nitz, M. *Carbohydr. Res.* **2009**, *344*, 278-284.
- [85] (a) Moreno, H. R.; Radomski, J. L. *Cancer Lett.* **1978**, *4*, 85-88; (b) Poupko, J. M.; Hearn, W. L.; Radomski, J. L. *Toxicol. Appl. Pharm.* **1979**, *50*, 479-484.
- [86] Peri, F.; Dumy, P.; Mutter, M. *Tetrahedron* **1998**, *54*, 12269-12278.
- [87] Loskot, S. A.; Zhang, J.; Langenhan, J. M. *J. Org. Chem.* **2013**, *78*, 12189-12193.
- [88] Goff, R. D.; Thorson, J. S. *Med. Chem. Comm.* **2014**, *5*, 1036-1047.
- [89] Iqbal, A.; Chibli, H.; Hamilton, C. J. *Carbohydr. Res.* **2013**, *377*, 1-3.
- [90] (a) Prudden, A. R.; Chinoy, Z. S.; Wolfert, M. A.; Boons, G. J. *Chem. Commun.* **2014**, *50*, 7132-7135; (b) McReynolds, K. D.; Dimas, D.; Le, H. *Tetrahedron Lett.* **2014**, *55*, 2270-2273.
- [91] Peri, F.; Nicotra, F. *Chem. Commun.* **2004**, 623-627.
- [92] Ishida, J.; Hinou, H.; Naruchi, K.; Nishimura, S. *Bioorg. Med. Chem. Lett.* **2014**, *24*, 1197-1200.
- [93] Griffith, B. R.; Krepel, C.; Fu, X.; Blanchard, S.; Ahmed, A.; Edmiston, C. E.; Thorson, J. S. *J. Am. Chem. Soc.* **2007**, *129*, 8150-8155.
- [94] Seo, J.; Michaelian, N.; Owens, S. C.; Dashner, S. T.; Wong, A. J.; Barron, A. E.; Carrasco, M. R. *Org. Lett.* **2009**, *11*, 5210-5213.
- [95] Thygesen, M. B.; Sorensen, K. K.; Clo, E.; Jensen, K. J. *Chem. Commun.* **2009**, 6367-6369.
- [96] Langenhan, J. M.; Peters, N. R.; Guzei, I. A.; Hoffmann, F. M.; Thorson, J. S. *Proc. Natl. Acad. Sci U S A* **2005**, *102*, 12305-12310.
- [97] Langenhan, J. M.; Engle, J. M.; Slevin, L. K.; Fay, L. R.; Lucker, R. W.; Smith, K. R.; Endo, M. M. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 670-673.
- [98] Goff, R. D.; Thorson, J. S. *Org. Lett.* **2009**, *11*, 461-464.
- [99] Nandurkar, N. S.; Zhang, J.; Ye, Q.; Ponomareva, L. V.; She, Q. B.; Thorson, J. S. *J. Med. Chem.* **2014**, *57*, 7478-7484.
- [100] Dasgupta, S.; Nitz, M. *J. Org. Chem.* **2011**, *76*, 1918-1921.
- [101] Nashed, E. M.; Grochowski, E.; Czyzewska, E. *Carbohydr. Res.* **1990**, *196*, 184-190.
- [102] (a) Yang, D.; Kim, S. H.; Kahne, D. *J. Am. Chem. Soc.* **1991**, *113*, 4715-4716; (b) Grochowski, E.; Jurczak, J. *Carbohydr. Res.* **1976**, *50*, C15-C16.
- [103] Andersson, M.; Oscarson, S. *Glycoconjugate. J.* **1992**, *9*, 122-125.
- [104] (a) Rodriguez, E. C.; Winans, K. A.; King, D. S.; Bertozzi, C. R. *J. Am. Chem. Soc.* **1997**, *119*, 9905-9906; (b) Rodriguez, E. C.; Marcaurelle, L. A.; Bertozzi, C. R. *J. Org. Chem.* **1998**, *63*,

- 7134-7135; (c) Cao, S.; Tropper, F. D.; Roy, R. *Tetrahedron* **1995**, *51*, 6679-6686.
- [105] (a) Dulery, V.; Renaudet, O.; Philouze, C.; Dumy, P. *Carbohydr. Res.* **2007**, *342*, 894-900; (b) Renaudet, O.; Dumy, P. *Tetrahedron Lett.* **2001**, *42*, 7575-7578
- [106] Chen, W.; Xia, C.; Cai, L.; Wang, P. G. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 3859-3862.
- [107] Reddy, C.; Rao, Y.; Kumar, T.; Reddy, K.; Chandrasekhar, S. *Synthesis* **2008**, 122-126.
- [108] Thomas, M.; Gesson, J.-P.; Papot, S. *J. Org. Chem.* **2007**, *72*, 4262-4264.
- [109] Yu, J.; Sun, J.; Yu, B. *Org. Lett.* **2012**, *14*, 4022-4025.
- [110] Forget, D.; Renaudet, O.; Defrancq, E.; Dumy, P. *Tetrahedron Lett.* **2001**, *42*, 7829-7832.
- [111] Marcaurelle, L. A.; Shin, Y.; Goon, S.; Bertozzi, C. R. *Org. Lett.* **2001**, *3*, 3691-3694.
- [112] Hudak, J. E.; Yu, H. H.; Bertozzi, C. R. *J. Am. Chem. Soc.* **2011**, *133*, 16127-16135.
- [113] Smith, E. L.; Giddens, J. P.; Iavarone, A. T.; Godula, K.; Wang, L. X.; Bertozzi, C. R. *Bioconjugate Chem.* **2014**, *25*, 788-795.
- [114] (a) Pujol, A. s. M.; Cuillel, M.; Renaudet, O.; Lebrun, C.; Charbonnier, P.; Cassio, D.; Gateau, C.; Dumy, P.; Mintz, E.; Delangle, P. *J. Am. Chem. Soc.* **2011** *133*, 286-296; (b) Bossu, I.; Sulc, M.; Krenek, K.; Dufour, E.; Garcia, J.; Berthet, N.; Dumy, P.; Kren, V.; Renaudet, O. *Org. Biomol. Chem.* **2011**, *9*, 1948-1959; (c) Andre, S.; Renaudet, O.; Bossu, I.; Dumy, P.; Gabius, H. J. *J. Pept. Sci.* **2011**, *17*, 427-437; (d) Renaudet, O.; Dumy, P. *Org. Biomol. Chem.* **2006**, *4*, 2628-2636; (e) Singh, Y.; Renaudet, O.; Defrancq, E.; Dumy, P. *Org. Lett.* **2005**, *7*, 1359-1362; (f) Renaudet, O.; Dumy, P. *Tetrahedron Lett.* **2004**, *45*, 65-68; (g) Renaudet, O.; Dumy, P. *Org. Lett.* **2003**, *5*, 243-246.
- [115] Peri, F.; Cipolla, L.; Rescigno, M.; La Ferla, B.; Nicotra, F. *Bioconjugate Chem.* **2001**, *12*, 325-328.
- [116] Cipolla, L.; Rescigno, M.; Leone, A.; Peri, F.; La Ferla, B.; Nicotra, F. *Bioorg. Med. Chem.* **2002**, *10*, 1639-1646.
- [117] Shin, I.; Lee, J. *Synlett* **2000**, 1297-1299.
- [118] Peyrat, S.; Cheng, K.; Xie, J. *Synthesis* **2013**, *45*, 2737-2744.
- [119] Grochowski, E.; Jurczak, J. *Carbohydr. Res.* **1976**, *50*, C15-C16.
- [120] Tronchet, J. M.; Zosimo-Landolfo, G.; Galland-Barrera, G.; Dolatshahi, N. *Carbohydr. Res.* **1990**, *204*, 145-156.
- [121] Chemla, P. *Tetrahedron Lett.* **1993**, *34*, 7391-7394.
- [122] (a) Brask, J.; Jensen, K. J. *Bioorg. Med. Chem. Lett* **2001**, *11*, 697-700; (b) Brask, J.; Jensen, K. J. *J. Pept. Sci.* **2000**, *6*, 290-299.
- [123] Pernille Tofteng, A.; Hansen, T. H.; Brask, J.; Nielsen, J.; Thulstrup, P. W.; Jensen, K. J. *Org. Biomol. Chem.* **2007**, *5*, 2225-2233.
- [124] Malapelle, A.; Ramozzi, R.; Xie, J. *Synthesis* **2009**, 888-890.
- [125] Gong, Y.; Sun, H.; Xie, J. *Eur. J. Org. Chem.* **2009**, 6027-6033.
- [126] Song, Z.; He, X.-P.; Chen, G.-R.; Xie, J. *Synthesis* **2011**, 2761-2766.
- [127] Kocienski, P. J. *Protecting Groups*, 3rd edition; Thieme: Stuttgart **2005**.
- [128] Corey, E. J.; Venkateswarlu, A. *J. Am. Chem. Soc.* **1972**, *94*, 6190-6191.
- [129] Chaudhary, S. K.; Hernandez, O. *Tetrahedron Lett.* **1979**, *20*, 99-102.
- [130] Ogilvie, K. K.; Iwacha, D. J. *Tetrahedron Lett.* **1973**, *14*, 317-319.

- [131] Halmos, T.; Montserret, R.; Filippi, J.; Antonakis, K. *Carbohydr. Res.* **1987**, *170*, 57-69.
- [132] Morales, J. C.; Zurita, D.; Penadés, S. *J. Org. Chem.* **1998**, *63*, 9212-9222.
- [133] Tomaszewski, M. J.; Holme, K. R.; Perlin, A. S. *Carbohydr. Res.* **1991**, *217*, 237-244.
- [134] Feher-Voelger, A.; Borges-Gonzalez, J.; Carrillo, R.; Morales, E. Q.; Gonzalez-Platas, J.; Martin, T. *Chem. Eur. J.* **2014**, *20*, 4007-4022.
- [135] (a) Salman, A. A.; Tabandeh, M.; Heidelberg, T.; Hussen, R. S. D. *Carbohydr. Res.* **2015**, *406*, 41-45;
(b) Karst, N.; Jacquinet, J.-C. *J. Chem. Soc., Perkin Trans 1*, **2000**, 2709-2717.
- [136] van Boeckel, C. A. A.; Visser, G. M.; van Boom, J. H. *Tetrahedron* **1985**, *41*, 4557-4565.
- [137] Mannock, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids.* **1987**, *43*, 113-127.
- [138] Wiggins, L. F. *J. Chem. Soc.* **1946**, 13-14.
- [139] Prakash, I.; Chaturvedula, V. S. *Molecules* **2014**, *19*, 20280-20294.
- [140] Wang, X.; Porco, J. A. *J. Am. Chem. Soc.* **2003**, *125*, 6040-6041.
- [141] Bartoszewicz, A.; Kalek, M.; Stawinski, J. *Tetrahedron* **2008**, *64*, 8843-8850.
- [142] Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.
- [143] Schenk, S.; Weston, J.; Anders, E. *J. Am. Chem. Soc.* **2005**, *127*, 12566-12576.
- [144] But, T. Y. S.; Toy, P. H. *Chem. Asian J.* **2007**, *2*, 1340-1355
- [145] Lin, L.; Shen, Q.; Chen, G. R.; Xie, J. *Bioorg. Med. Chem.* **2008**, *16*, 9757-9763.
- [146] Johnson, S. M.; Petrassi, H. M.; Palaninathan, S. K.; Mohamedmohaideen, N. N.; Purkey, H. E.; Nichols, C.; Chiang, K. P.; Walkup, T.; Sacchettini, J. C.; Sharpless, K. B. *J. Med. Chem.* **2005**, *48*, 1576-1587.
- [147] Leduc, A. B.; Kerr, M. A. *Angew. Chem. Int. Ed. Engl.* **2008**, *47*, 7945-7948.
- [148] Janwitayanuchit, W.; Suwanborirux, K.; Patarapanich, C.; Pummangura, S.; Lipipun, V.; Vilaivan, T. *Phytochemistry* **2003**, *64*, 1253-1264.
- [149] Lattrell, R.; Lohaus, G. *Justus Liebigs Ann. Chem.* **1974**, 901-920.
- [150] Albert, R.; Dax, K.; Link, R. W.; Stütz, A. E. *Carbohydr. Res.* **1983**, *118*, C5-C6.
- [151] Momiyama, N.; Yamamoto, H. *J. Am. Chem. Soc.* **2003**, *125*, 6038-6039.
- [152] Kim, S.; Lee, T. A.; Song, Y. *Synlett* **1998**, 471-472.
- [153] Zhang, P.; Li, C.; Yang, H.; Wei, H.; Xia, Z.; Ma, D.; Chen, H.; Wang, K.; Li, X. *Carbohydr. Res.* **2014**, *398*, 36-39.
- [154] Harding, K. E.; May, L. M.; Dick, K. F. *J. Org. Chem.* **1975**, *40*, 1664-1665.
- [155] Xie, J. *Eur. J. Org. Chem.* **2002**, 3411-3418.
- [156] Chinchilla, R.; Dodsworth, D. J.; Nájera, C.; Soriano, J. M. *Tetrahedron Lett.* **2001**, *42*, 7579-7581.
- [157] Greene, T. W.; Wuts, P. G. M. *Protective Groups in Organic Synthesis, 4th edition.* **2006**, Chapter 7.
- [158] Le Grel, P.; Salaun, A.; Mocquet, C.; Le Grel, B.; Roisnel, T.; Potel, M. *J. Org. Chem.* **2011**, *76*, 8756-8767.

Experimental Section

I. General

All commercial available reagents purchased from Sigma-Aldrich, TCI Chemical and Carlo-Erba Reagents, etc. were used without further purification. All solvents for column chromatography were used without purification. The reactions carried out under anhydrous conditions are performed under argon in glassware previously dried in an oven. Methanol is dried over molecular sieve 3Å. Tetrahydrofuran (THF), dimethylformamide (DMF), dichloromethane, acetonitrile and toluene previously were dried through alumina cartridge using a solvent purificator MBRAUN SPS-800. Petroleum ether used refers to the fraction boiling in the range 60-90 °C.

All reactions were monitored by TLC on Silica Gel 60F-254 plates with detection by UV (254 nm or 365 nm) or by spraying with 10% H₂SO₄ in EtOH and heating about 30 s at 400-600 °C.

All Column chromatography was performed on Silica gel 60 (40-60 μm).

Melting points were determined with a Kofler melting point apparatus.

Optical rotations were measured using a Jasco P-2000 polarimeter at room temperature in a 10 cm, 1 mL cell.

NMR spectra were recorded on a JOEL ESC-400 spectrometer in CDCl₃ or CD₃OD solution. Chemical shift was given in units of parts per million related to TMS or solvent proton (¹H: CHCl₃ in CDCl₃, 7.27 ppm; CH₃OH in CD₃OD, 3.31 ppm; ¹³C: CHCl₃ in CDCl₃, 77.16 ppm; CH₃OH in CD₃OD, 49.00 ppm) as internal reference.

High-resolution mass Spectra (HRMS) were recorded on a Q-TOF MaXis using standard conditions or Bruker Microflex™ MALDI-TOF mass spectrometry.

II. General Procedures

Desilylation:

Procedure A: To a solution of silylated compound (1 eq.) in anhydrous THF (15 mL/mmol) under argon, was added TBAF (3 eq.). After stirring at room temperature overnight, the solvent was removed under vacuum. The residue was diluted with EtOAc (25 mL/mmol), washed with saturated aq NH₄Cl (10 mL/mmol), H₂O (10 mL/mmol) and brine (10 mL/mmol), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel to afford the silylated compound.

Procedure B: To a solution of silylated compound (1 eq.) in anhydrous MeOH (20

mL/mmol), was added AcCl (0.15 eq.) at 0 °C. The resulting mixture was stirred at room temperature for 2 h under argon. After completion of the reaction, 5% NaHCO₃ was added to neutralize the reaction mixture, and then the solvent was removed under vacuum. The residue was taken up in H₂O-CH₂Cl₂ (1/1, 30 mL/mmol) and the aqueous layer was extracted with CH₂Cl₂ (3×20 mL/mmol). The combined organic layers were washed with brine (20 mL/mmol), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel to afford the desilylated compound.

Mitsunobu Reaction:

Procedure C: To a solution of alcohol (1 eq.), Ph₃P (3 eq.) and PhthNOH (3 eq.) in toluene (20 mL/mmol) at 0 °C under argon, was added DIAD (3 eq.) dropwise. The resulting mixture was stirred at room temperature for 1 h and then extracted with EtOAc (3×30 mL/mmol). The combined organic layers were washed with saturated aq NaHCO₃ (3×30 mL/mmol), H₂O (30 mL/mmol) and brine (30 mL/mmol), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel to afford the *O*-phthalimido compound.

Hydrazinolysis:

Procedure D: To a solution of *O*-phthalimido compound (1 eq.) in MeOH (20 mL/mmol), was added N₂H₄·H₂O (2 eq.). The mixture was stirred at room temperature for 2 h and then extracted with CH₂Cl₂ (3×50 mL/mmol). The combined organic layers were washed with saturated aq NaHCO₃ (50 mL/mmol), H₂O (50 mL/mmol) and brine (50 mL/mmol), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel to afford the *O*-amino compound.

Procedure E: To a solution of *O*-phthalimido compound (1 eq.) in anhydrous MeOH (20 mL/mmol), was added N₂H₄·H₂O (1.1 eq.). The resulting mixture was stirred at 0 °C for 20 min under argon and then extracted with CH₂Cl₂ (3×50 mL/mmol). The combined organic layers were washed with saturated aq NaHCO₃ (50 mL/mmol), H₂O (50 mL/mmol) and brine (50 mL/mmol), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel to afford the oxyamine compound.

Coupling Reaction for N-oxyamide Formation:

Procedure F: To a solution of carboxylic acid (1 eq.) in anhydrous CH₂Cl₂ (15 mL/mmol), was added HOBt (2 eq.), EDC·HCl (2 eq.) and Et₃N (2 eq.) under argon at 0 °C. After stirring for 20 min, the oxyamine derivative (1 eq.) was added. The resulting mixture was stirred at room temperature overnight. The solution was diluted with EtOAc (100 mL/mmol), washed

with aq HCl (1N, 2×40 mL/mmol), saturated aq NaHCO₃ (2×40 mL/mmol) and brine (40 mL/mmol), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel to give the *N*-oxyamide.

Esterification:

Procedure G: To a solution of carboxylic acid (1 eq.) in anhydrous CH₂Cl₂ (15 mL/mmol), were added EDC·HCl (2 eq.), DMAP (2 eq.) and Et₃N (2 eq.) under argon at 0 °C. After the mixture being stirred for 20 min, the alcohol (1 eq.) was added. The resulting mixture was stirred at room temperature overnight. The solution was diluted with CH₂Cl₂ (50 mL/mmol), washed with aq HCl (1N, 2×40 mL/mmol), saturated aq NaHCO₃ (2×40 mL/mmol) and brine (40 mL/mmol), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel to give the ester.

Deacetylation:

Procedure H: To a solution of acetylated compound (1 eq.) in 85% EtOH (50 mL/mmol) was added N₂H₄·H₂O (12 eq.). The mixture was stirred at 50 °C overnight. The solution was poured into ice-cold brine and extracted with CHCl₃ (3×100 mL/mmol). The combined CHCl₃ layers were dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel to give the deacetylated compound.

Procedure I: To a solution of acetylated compound (1eq.) in anhydrous MeOH (20 mL/mmol), was added sodium methoxide (0.3 eq.). After completion of the reaction, cation exchange resin was added to neutralize the solution. The resin was filtered and the filtrate was concentrated to afford the deacetylated compound.

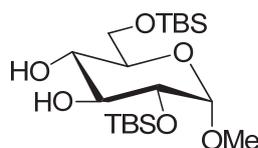
III. Synthesis and Characterization

Methyl 2,6-di- <i>O</i> - <i>tert</i> -butyldimethylsilyl- α -D-glucopyranoside (1)	166
Methyl 3,4-di- <i>O</i> -(4-methoxybenzyl)-2,6-di- <i>O</i> - <i>tert</i> -butyldimethylsilyl- α -D-glucopyranoside (2)	167
Methyl 3,4-di- <i>O</i> -(4-methoxybenzyl)- α -D-glucopyranoside (3)	168
Methyl 3,4-di- <i>O</i> -(4-methoxybenzyl)-6- <i>O</i> - <i>tert</i> -butyldimethylsilyl- α -D-glucopyranoside (4)	169
Methyl 2- <i>O</i> -ethoxycarbonylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)-6- <i>O</i> - <i>tert</i> -butyldimethylsilyl- α -D-glucopyranoside (5)	170
Methyl 2- <i>O</i> -ethoxycarbonylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)- α -D-glucopyranoside (6)	171
Methyl 2- <i>O</i> -ethoxycarbonylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)-6- <i>O</i> -phthalimido- α -D-glucopyranoside (7)	172
Methyl 6- <i>O</i> -amino-2- <i>O</i> -ethoxycarbonylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)- α -D-glucopyranoside (8)	173
Methyl 2- <i>O</i> -ethoxycarbonylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)-6- <i>O</i> -palmitoylamino- α -D-glucopyranoside (9)	174
Methyl 2- <i>O</i> -carboxylmethyl-3,4-di- <i>O</i> -(4-methoxybenzyl)-6- <i>O</i> -palmitoylamino- α -D-glucopyranoside (10)	175
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Methyl 2,6-di-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (1)Chemical Formula: C₁₉H₄₂O₆Si₂

Exact Mass: 422.2520 g/mol

To a solution of methyl α -D-glucopyranoside (0.97 g, 5 mmol) in pyridine (10 mL) under argon at 0 °C, were added TBSCl (2.1 g, 14 mmol) and DMAP (0.12 g, 1 mmol). After stirring at room temperature for 16 h, the solvent was removed under vacuum, and the concentrated mixture was partitioned between CH₂Cl₂ (50 mL) and H₂O (50 mL). The aqueous layer was extracted with CH₂Cl₂ (2×50 mL). The combined organic layers were dried over MgSO₄, filtered, concentrated, and purified by column chromatography over silica gel (petroleum ether/EtOAc = 10/1) to afford compound **1** (1.82 g, 86.3%) as a colourless paste.

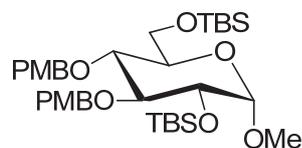
R_f = 0.66 (petroleum ether/EtOAc = 5/1).

¹H NMR (400 MHz, CDCl₃): δ 4.60 (d, *J* = 3.7 Hz, 1H, H-1), 3.87-3.74 (m, 3H, H-3,6), 3.63-3.57 (m, 1H, H-5), 3.55-3.45 (m, 2H, H-2,4), 3.37 (s, 3H, OCH₃), 2.78 (s, 2H, 2×OH), 0.89, 0.89 (2×s, 18H, 2×*t*-Bu), 0.12-0.04 (m, 12H, 2×Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 100.0 (C-1), 74.1 (C-3), 73.6 (C-2), 71.8 (C-4), 70.8 (C-5), 64.1 (CH₂), 55.3 (OCH₃), 26.0, 25.9 (CH₃, *t*-Bu); 18.5, 18.3 (C_q, *t*-Bu); -4.4, -4.5, -5.3, -5.3 (CH₃, Si(CH₃)₂).

Ref.: Halmos, T.; Montserret, R.; Filippi, J.; Antonakis, K. *Carbohydr. Res.* **1987**, *170*, 57-69. (Known structure)

Methyl 3,4-di-*O*-(4-methoxybenzyl)-2,6-di-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (2)

Chemical Formula: C₃₅H₅₈O₈Si₂

Exact Mass: 662.3670 g/mol

To a solution of compound **1** (4.5 g, 10.6 mmol) in anhydrous DMF (50 ml) under argon at 0 °C, was added NaH (60%, 2.55 g, 63.8 mmol) and PMBCl (5.8 ml, 42.4 mmol) in portions over 1 h. After addition, the solution was stirred at room temperature for 16 h. Ice was added to destroy the excess NaH, and then the solvent was evaporated. The residue was diluted with EtOAc (200 mL), washed with saturated aq NH₄Cl (100 mL), H₂O (100 mL) and brine (100 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc = 20/1) to afford compound **2** (5.64 g, 80%) as a colourless paste.

(NaH and PMBCl was added at the same time, 1/4 equivalent every 20 min.)

R_f = 0.48 (petroleum ether/EtOAc = 10/1).

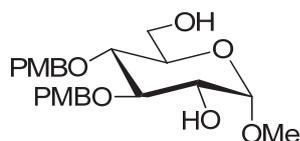
[α]_D = +36.3 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.33-7.20 (m, 4H, PMB), 6.90-6.82 (m, 4H, PMB), 4.79-4.40 (m, 4H, 2 \times OCH₂), 4.33 (d, *J* = 3.6 Hz, 1H, H-1), 3.96 (t, *J* = 8.9 Hz, 1H, H-3), 3.80, 3.78 (2 \times s, 6H, 2 \times OCH₃), 3.72-3.59 (m, 2H, H-6), 3.53-3.46 (m, 1H, H-5), 3.31-3.19 (m, 5H, H-2,4, OCH₃), 0.94, 0.85 (2 \times s, 18H, 2 \times *t*-Bu), 0.13-0.02 (m, 12H, 2 \times Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 159.4, 159.1, 130.8, 130.6 (C_q, PMB); 130.0, 129.3, 113.8 (CH, PMB); 98.0 (C-1), 80.1 (C-2), 79.2 (C-4), 74.6 (CH₂), 74.1 (C-3), 73.3 (CH₂), 71.4 (C-5), 62.6 (CH₂), 55.4, 54.8 (OCH₃); 26.0, 26.0 (CH₃, *t*-Bu); 18.4, 18.3 (C_q, *t*-Bu); -3.7, -4.1, -5.0, -5.3 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₅H₅₈NaO₈Si₂ 685.3568; Found 685.3562.

Methyl 3,4-di-*O*-(4-methoxybenzyl)- α -D-glucoopyranoside (3)

Chemical Formula: C₂₃H₃₀O₈Exact Mass: 434.1941 g/mol

Desilylation of compound **2** (5.64 g, 8.52 mmol) using TBAF according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **3** (2.53 g, 68.4%) as a white solid.

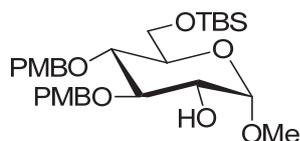
R_f = 0.23 (petroleum ether/EtOAc = 1/1).

¹H NMR (400 MHz, CDCl₃): δ 7.33-7.25 (m, 4H, PMB), 6.92-6.85 (m, 4H, PMB), 4.82 (d, J = 11.0 Hz, 1H, OCH), 4.67-4.57 (m, 3H, OCH, OCH₂), 4.54 (d, J = 3.7 Hz, 1H, H-1), 4.05 (t, J = 9.2 Hz, 1H, H-3), 3.81, 3.80 (2×s, 6H, 2×OCH₃), 3.79-3.67 (m, 2H, H-6), 3.62-3.57 (m, 1H, H-5), 3.42 (t, J = 9.4 Hz, 1H, H-4), 3.36-3.27 (m, 4H, H-2, OCH₃), 2.43 (s, 1H, OH), 1.67 (s, 1H, OH).

¹³C NMR (100 MHz, CDCl₃): δ 159.7, 159.5, 130.5, 130.1 (C_q, PMB); 129.9, 129.9, 114.1, 114.0 (CH, PMB); 97.7 (C-1), 79.4 (C-2), 77.0 (C-4), 74.3 (CH₂), 73.5 (C-3), 72.9 (CH₂), 70.3 (C-5), 62.1 (CH₂), 55.4, 55.3 (OCH₃).

Ref.: Gathirwa, J. W.; Maki, T. *Tetrahedron* **2012**, *68*, 370-375. (Known structure)

Methyl 3,4-di-*O*-(4-methoxybenzyl)-6-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (4)

Chemical Formula: C₂₉H₄₄O₈Si

Exact Mass: 548.2805 g/mol

To a solution of compound **3** (2.53 g, 5.82 mmol) in pyridine (40 mL) under argon at 0 °C, were added TBSCl (1.09 g, 7.27 mmol) and DMAP (0.18 g, 1.46 mmol). After stirring at room temperature for 16 h, the solvent was removed under vacuum, and the concentrated mixture was partitioned between CH₂Cl₂ (100 mL) and H₂O (50 mL). The aqueous layer was extracted with CH₂Cl₂ (2×50 mL). The combined organic layers were dried over MgSO₄, filtered, concentrated, and purified by column chromatography over silica gel (petroleum ether/EtOAc = 4/1) to afford compound **4** (3.15 g, 98.7%) as a colourless paste.

R_f = 0.55 (petroleum ether/EtOAc = 2/1).

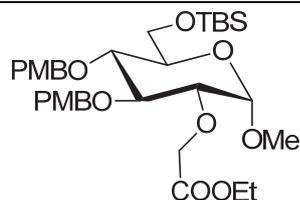
$[\alpha]_D = +58.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.32-7.24 (m, 4H, PMB), 6.92-6.84 (m, 4H, PMB), 4.80 (d, J = 11.0 Hz, 1H, OCH), 4.66-4.56 (m, 4H, OCH, OCH₂, H-1), 4.04 (t, J = 9.2 Hz, 1H, H-3), 3.84-3.73 (m, 8H, 2×OCH₃, H-6), 3.55 (ddd, J = 9.9, 4.3, 2.1 Hz, 1H, H-5), 3.41 (t, J = 9.4 Hz, 1H, H-4), 3.36-3.27 (m, 4H, H-2, OCH₃), 2.40 (broad s, 1H, OH), 0.90 (s, 9H, *t*-Bu), 0.65-0.45 (m, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 159.6, 159.4, 130.9, 130.3 (C_q, PMB); 129.8, 129.7, 114.1, 114.0 (CH, PMB); 97.4 (C-1), 79.6 (C-2), 77.4 (C-4), 74.3 (CH₂), 73.7 (C-3), 72.8 (CH₂), 71.2 (C-5), 62.5 (CH₂), 55.4, 55.0 (OCH₃); 26.1 (CH₃, *t*-Bu), 18.5 (C_q, *t*-Bu), -5.0, -5.2 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₂₉H₄₄NaO₈Si 571.2703; Found 571.2698.

Methyl 2-*O*-ethoxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (5)



Chemical Formula: C₃₃H₅₀O₁₀Si

Exact Mass: 634.3173 g/mol

To a solution of compound **4** (200 mg, 0.36 mmol) in DMF (5 mL) under argon at 0 °C, was added NaH (60%, 144 mg, 3.6 mmol) and TBAI (386 mg, 1.1 mmol). After stirring at 0 °C for 2 h, ethyl bromoacetate (0.4 mL, 3.6 mmol) was added drop by drop during 30 min, and the resulting mixture was stirred at room temperature for 15 h. The reaction was quenched with ice, and the mixture was extracted with EtOAc (3× 20 mL). The combined organic layers were washed with saturated aq NH₄Cl (20 mL) and brine (20 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc = 4/1) to afford compound **4** (72 mg, conversion: 64%) and compound **5** (135 mg, corrected yield: 92.5%) as a colourless paste.

R_f = 0.55 (petroleum/EtOAc = 3/1).

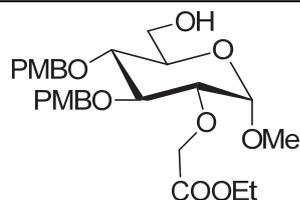
[α]_D = +27.3 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.35-7.23 (m, 4H, PMB), 6.94-6.83 (m, 4H, PMB), 4.86 (d, *J* = 10.5 Hz, 1H, OCH), 4.71-4.47 (m, 6H, OCH, 2×OCH₂, H-1), 4.22 (q, *J* = 7.0 Hz, 2H, OCH₂), 3.88-3.72 (m, 9H, 2×OCH₃, H-3,6), 3.59-3.47 (m, 3H, H-2,4,5), 3.29 (s, 3H, OCH₃), 1.27 (t, *J* = 7.1 Hz, 3H, CH₃), 0.96-0.86 (m, 9H, *t*-Bu), 0.09-0.02 (m, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.4 (C=O), 159.5, 159.3, 130.9, 130.4 (C_q, PMB); 129.8, 114.0, 113.9 (CH, PMB); 97.8 (C-1), 83.7, 79.8, 77.4 (CH); 74.8, 73.0 (CH₂); 71.4 (CH), 71.1, 62.4, 60.8 (CH₂); 55.4, 54.9 (OCH₃); 26.0 (CH₃, *t*-Bu), 18.4 (C_q, *t*-Bu), 14.4 (CH₃), -5.0, -5.2 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₃H₅₀NaO₁₀Si 657.3071; Found 657.3065.

Methyl 2-*O*-ethoxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)- α -D-glucopyranoside (6)

Chemical Formula: C₂₇H₃₆O₁₀

Exact Mass: 520.2308 g/mol

Desilylation of compound **5** (0.50 g, 0.79 mmol) according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **6** (0.3 g, 73%) as a colourless paste.

R_f = 0.48 (petroleum/EtOAc = 1/2).

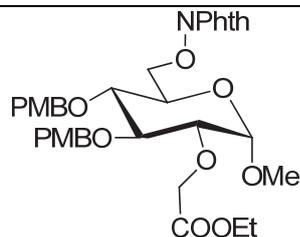
$[\alpha]_D = +34.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.39-7.25 (m, 4H, PMB), 6.95-6.84 (m, 4H, PMB), 4.88 (d, J = 11.0 Hz, 1H, OCH), 4.74-4.36 (m, 6H, OCH, 2×OCH₂, H-1), 4.21 (q, J = 7.0 Hz, 2H, OCH₂), 3.92-3.77 (m, 7H, 2×OCH₃, H-3), 3.76-3.62 (m, 2H, H-6), 3.60-3.46 (m, 3H, H-2,4,5), 3.29 (s, 3H, OCH₃), 1.27 (t, J = 7.1 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.3 (C=O), 159.6, 159.5, 130.5, 130.2 (C_q, PMB); 130.0, 129.8, 114.0 (CH, PMB); 98.1 (C-1), 83.4, 79.7, 76.5 (CH); 74.7, 73.0, 70.9 (CH₂); 70.5 (CH), 62.0, 60.9 (CH₂); 55.4, 55.2 (OCH₃); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₂₇H₃₆NaO₁₀ 543.2206; Found 543.2201.

Methyl 2-*O*-ethoxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-phthalimido- α -D-glucopyranoside (7)



Chemical Formula: C₃₅H₃₉NO₁₂

Exact Mass: 665.2472 g/mol

The phthalimido group was introduced at the 6-position of compound **6** (0.3 g, 0.58 mmol) using Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **7** (0.33 g, 85%) as a colourless paste.

$R_f = 0.47$ (petroleum ether/EtOAc: 1/1).

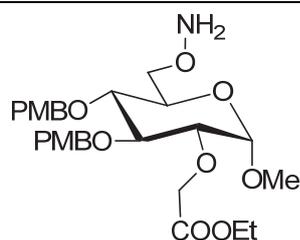
$[\alpha]_D = +47.3$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.86-7.72 (m, 4H, Phth), 7.35-7.26 (m, 4H, PMB), 6.90-6.82 (m, 4H, PMB), 4.98 (d, $J = 10.1$ Hz, 1H, OCH), 4.81 (d, $J = 10.5$ Hz, 1H, OCH), 4.70 (d, $J = 11.9$ Hz, 1H, OCH), 4.56 (d, $J = 9.6$ Hz, 1H, OCH), 4.55-4.35 (m, 5H, OCH₂, H-1,6), 4.21 (q, $J = 7.0$ Hz, 2H, OCH₂), 3.87-3.72 (m, 9H, 2×OCH₃, H-3,4,5), 3.57 (dd, $J = 9.3, 3.5$ Hz, 1H, H-2), 3.35 (s, 3H, OCH₃), 1.27 (t, $J = 7.1$ Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.2 (C=O), 163.3 (C=O, Phth); 159.5, 159.4 (C_q); 134.5 (CH, Phth), 130.6, 130.2 (C_q); 130.1, 129.9 (CH, PMB); 129.0 (C_q), 123.6 (CH, Phth), 114.0, 113.9 (CH, PMB); 98.2 (C-1), 83.4, 79.2 (CH); 76.5 (CH₂), 76.1 (CH), 74.9, 73.1, 71.0 (CH₂); 69.2 (CH), 60.8 (CH₂), 55.6, 55.3 (OCH₃); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₅H₃₉NaO₁₂ 688.2370; Found 688.2374.

Methyl 6-*O*-amino-2-*O*-ethoxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)- α -D-glucopyranoside (8)

Chemical Formula: C₂₇H₃₇NO₁₀

Exact Mass: 535.2417 g/mol

Hydrazinolysis of compound **7** (67 mg, 0.1 mmol) according to the general **procedure D** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **8** (40 mg, 75%) as a colourless paste.

R_f = 0.25 (petroleum ether/EtOAc: 1/1).

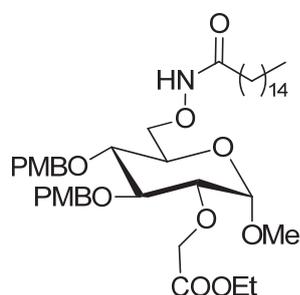
[α]_D = +30.3 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.38-7.22 (m, 4H, PMB), 6.97-6.82 (m, 4H, PMB), 4.91-4.35 (m, 7H, 3 \times OCH₂, H-1), 4.20 (q, *J* = 7.0 Hz, 2H, OCH₂), 3.93-3.72 (m, 9H, 2 \times OCH₃, H-3,6), 3.72-3.65 (m, 1H, H-5), 3.57-3.45 (m, 2H, H-2,4), 3.31 (s, 3H, OCH₃), 1.26 (t, *J* = 7.1 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.3 (C=O), 159.5, 159.4, 130.6, 130.2 (C_q, PMB); 130.0, 129.9, 114.0, 113.9 (CH, PMB); 98.0 (C-1), 83.5, 79.5, 76.8 (CH); 74.8, 74.4, 73.0, 70.9 (CH₂); 69.5 (CH), 60.8 (CH₂), 55.4, 55.3 (OCH₃); 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₂₇H₃₇KNO₁₀ 574.2055; Found 574.2057.

Methyl 2-*O*-ethoxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-palmitoylamino- α -*D*-glucopyranoside (9**)**



Chemical Formula: C₄₃H₆₇NO₁₁

Exact Mass: 773.4714 g/mol

Compound **9** was prepared from compound **8** (70 mg, 0.13 mmol) and palmitic acid according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/2) afforded compound **9** (86 mg, 86%) as a white solid.

R_f = 0.48 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = +39.7$ (c 0.1, CHCl₃).

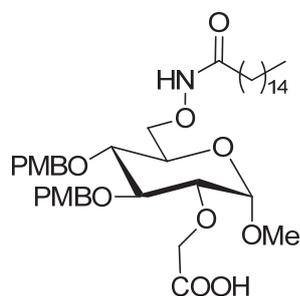
M.p. 75 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.04 (s, 1H, NH), 7.39-7.21 (m, 4H, PMB), 6.97-6.81 (m, 4H, PMB), 4.95-4.35 (m, 7H, 3×OCH₂, H-1), 4.20 (q, J = 7.0 Hz, 2H, OCH₂), 4.11-3.99 (m, 2H, H-6), 3.88-3.68 (m, 8H, 2×OCH₃, H-3,5), 3.61-3.46 (m, 2H, H-2,4), 3.31 (s, 3H, OCH₃), 2.43-1.92 (m, 2H, CH₂), 1.66-1.54 (m, 2H, CH₂), 1.45-1.17 (m, 27H, 12×CH₂, CH₃), 0.88 (t, J = 6.4 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.7, 170.2 (C=O); 159.6, 159.4, 130.3, 130.2 (C_q, PMB); 129.8, 114.0, 113.9 (CH, PMB); 98.1 (C-1), 83.4, 79.5, 76.3 (CH); 74.7, 73.0, 70.9 (CH₂); 69.5 (CH), 60.9 (CH₂), 55.5, 55.4 (OCH₃); 33.4, 32.0, 29.8, 29.6, 29.5, 25.5, 22.8 (CH₂); 14.3, 14.2 (CH₃).

HRMS (ESI) m/z : [M+K]⁺ Calcd for C₄₃H₆₇KNO₁₁ 812.4351; Found 812.4353.

Methyl 2-*O*-carboxymethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-palmitoylamino- α -D-glucopyranoside (10)

Chemical Formula: C₄₁H₆₃NO₁₁

Exact Mass: 745.440 g/mol

To a solution of compound **9** (74 mg, 0.096 mmol) in MeOH (2 mL), was added LiOH (23 mg, 0.96 mmol). The mixture was stirred at room temperature for 48 h and then extracted with CH₂Cl₂ (3×10 mL). The combined organic layers were washed with aq HCl (1N, 10 mL) and brine (10 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (CH₂Cl₂/ MeOH: 25/1) to afford compound **10** (59 mg, 83%) as a white solid.

R_f = 0.33 (CH₂Cl₂/ MeOH: 25/1).[α]_D = +62.0 (*c* 0.1, CHCl₃).

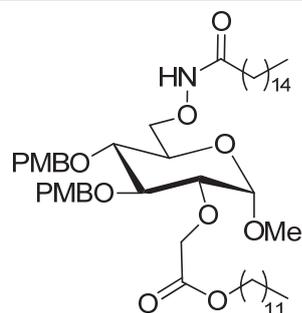
M.p. 115 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.14 (s, 1H, NH), 7.35-7.15 (m, 4H, PMB), 6.95-6.77 (m, 4H, PMB), 4.82-4.52 (m, 5H, 2×OCH₂, H-1), 4.44 (d, *J* = 17.4, OCH), 4.36 (d, *J* = 17.4, OCH), 4.20-3.93 (m, 2H, H-6), 3.81, 3.79 (2×s, 6H, 2×OCH₃), 3.76-3.57 (m, 3H, H-3,4,5), 3.49 (dd, *J* = 9.5, 3.1 Hz, 1H, H-2), 3.30 (s, 3H, OCH₃), 2.43-1.95 (m, 2H, CH₂), 1.68-1.52 (m, 2H, CH₂), 1.36-1.06 (m, 24H, 12×CH₂), 0.88 (t, *J* = 6.4 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.1, 171.0 (C=O); 160.0, 159.7 (C_q, PMB); 130.4, 130.2 (CH, PMB); 129.5, 128.5 (C_q, PMB); 114.2, 114.1 (CH, PMB); 97.2 (C-1), 82.2, 78.0, 77.4 (CH); 74.9, 74.3, 72.6, 70.5 (CH₂); 69.5 (CH), 55.6, 55.4 (OCH₃); 33.4, 32.0, 29.8, 29.6, 29.5, 25.4, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₄₁H₆₃NaNO₁₁ 768.4299; Found 768.4297.

Methyl 2-*O*-dodecyloxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-palmitoylamino- α -D-glucopyranoside (11**)**



Chemical Formula: C₅₃H₈₇NO₁₁

Exact Mass: 913.6279 g/mol

- (a) Esterification of compound **10** (52 mg, 0.07 mmol) with 1-dodecanol according to the general **procedure G** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/1) afforded compound **11** (40 mg, 63%) as a white solid.
- (b) Compound **11** could also be prepared from compound **16** (20 mg, 0.03 mmol) and palmitic acid according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **11** (24 mg, 88%) as a paste.

$R_f = 0.56$ (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = +27.3$ (c 0.1, CHCl₃).

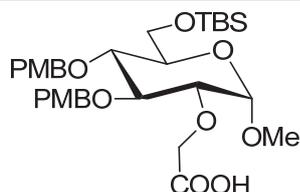
M.p. 70 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.01 (s, 1H, NH), 7.37-7.20 (m, 4H, PMB), 6.96-6.82 (m, 4H, PMB), 4.95-4.36 (m, 7H, 3×OCH₂, H-1), 4.13 (t, $J = 6.8$ Hz, 2H, OCH₂), 4.09-3.89 (m, 2H, H-6), 3.86-3.75 (m, 7H, 2×OCH₃, H-3), 3.74-3.67 (m, 1H, H-5), 3.60-3.44 (m, 2H, H-2,4), 3.30 (s, 3H, OCH₃), 2.41-1.93 (m, 2H, CH₂), 1.69-1.53 (m, 4H, 2×CH₂), 1.40-1.19 (m, 42H, 21×CH₂), 0.93-0.83 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.6, 170.3 (C=O); 159.5, 159.4, 130.5, 130.1 (C_q, PMB); 130.4, 129.8, 114.0, 113.9 (CH, PMB); 98.1 (C-1), 83.4, 79.4, 76.2 (CH); 74.7, 73.1, 70.8 (CH₂); 69.5 (CH), 65.1 (CH₂), 55.5, 55.4 (OCH₃); 33.4, 32.0, 29.8, 29.6, 29.5, 29.4, 28.7, 26.0, 25.5, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₅₃H₈₇NaNO₁₁ 936.6177; Found 936.6179.

Methyl 2-*O*-carboxymethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-*tert*-butyldimethylsilyl- α -*D*-glucopyranoside (12)



Chemical Formula: C₃₁H₄₆O₁₀Si

Exact Mass: 606.2860 g/mol

To a mixture of compound **4** (0.31 g, 0.57 mmol), aqueous NaOH (5 mL, 50%) and TBAI (104 mg, 0.28 mmol) in toluene (10 mL), was added ethyl bromoacetate (0.13 mL, 1.14 mmol) dropwise. After stirring at room temperature for 3 days, the solution was concentrated under vacuum. The residue was diluted with EtOAc (50 mL), washed with saturated aq HCl (1N, 3×20 mL), H₂O (20 mL) and brine (20 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (CH₂Cl₂/ MeOH: 100/3) to afford compound **4** (103 mg, conversion: 67%) and compound **12** (170 mg, corrected yield: 74%) as a white powder.

R_f = 0.25 (CH₂Cl₂/ MeOH: 100/3).

[α]_D = +64.0 (*c* 0.1, CHCl₃).

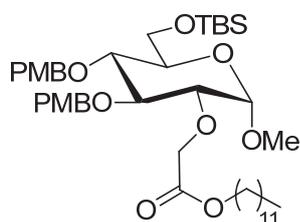
M.p. 138 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.38-7.20 (m, 4H, PMB), 6.99-6.82 (m, 4H, PMB), 4.79-4.46 (m, 5H, 2×OCH₂, H-1), 4.40 (d, *J* = 17.4 Hz, OCH), 4.32 (d, *J* = 18.4 Hz, OCH), 4.02-3.75 (m, 8H, 2×OCH₃, H-6), 3.71 (t, *J* = 8.9 Hz, H-3), 3.61-3.43 (m, 3H, H-2,4,5), 3.30 (s, 3H, OCH₃), 0.91 (s, 9H, *t*-Bu), 0.16-0.02 (m, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 172.1 (C=O), 159.9, 159.7 (C_q, PMB); 130.2, 130.0 (CH, PMB); 129.5, 128.6 (C_q, PMB); 114.2, 114.1 (CH, PMB); 96.8 (C-1), 82.0, 78.5, 77.6 (CH); 74.8, 72.4 (CH₂); 71.5 (CH), 70.5, 61.9 (CH₂); 55.4, 55.1 (OCH₃); 26.0 (CH₃, *t*-Bu), 18.4 (C_q, *t*-Bu), -5.0, -5.2 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₁H₄₆NaO₁₀Si 629.2758; Found 629.2759.

Methyl 2-*O*-dodecyloxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (13)

Chemical Formula: C₄₃H₇₀O₁₀Si

Exact Mass: 774.4738 g/mol

Esterification of compound **12** (148 mg, 0.24 mmol) with 1-dodecanol according to the general **procedure G** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 5/1) afforded compound **13** (144 mg, 76%) as a white paste.

R_f = 0.63 (petroleum ether/EtOAc: 4/1).

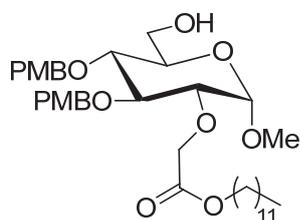
$[\alpha]_D = +23.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.50-7.19 (m, 4H, PMB), 7.05-6.80 (m, 4H, PMB), 5.00-4.32 (m, 7H, 3×OCH₂, H-1), 4.10 (t, J = 6.7 Hz, 2H, OCH₂), 4.04-3.68 (m, 9H, 2×OCH₃, H-3,6), 3.64-3.44 (m, 3H, H-2,4,5), 3.26 (s, 3H, OCH₃), 1.70-1.54 (m, 2H, CH₂), 1.52-1.17 (m, 18H, 9×CH₂), 1.02-0.82 (m, 12H, *t*-Bu, CH₃), 0.16-0.02 (m, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.4 (C=O), 159.4, 159.3, 130.7, 130.3 (C_q, PMB); 129.8, 113.8 (CH, PMB); 97.7 (C-1), 83.6, 79.7, 76.8 (CH); 74.7, 73.0 (CH₂); 71.3 (CH), 70.9, 65.0, 62.3 (CH₂); 55.3, 54.9 (OCH₃); 32.0, 29.7, 29.6, 29.4, 29.3, 28.7 (CH₂); 26.0 (CH₃, *t*-Bu), 25.9, 22.8 (CH₂); 18.4 (C_q, *t*-Bu), 14.2 (CH₃), -5.1, -5.3 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₄₃H₇₀NaO₁₀Si 797.4636; Found 797.4639.

Methyl 2-*O*-dodecyloxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)- α -D-glucopyranoside (14)

Chemical Formula: C₃₇H₅₆O₁₀

Exact Mass: 660.3873 g/mol

Desilylation of compound **13** (74 mg, 0.096 mmol) using TBAF according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **14** (45 mg, 71%) as a colourless paste.

R_f = 0.10 (petroleum ether/EtOAc = 3/1).

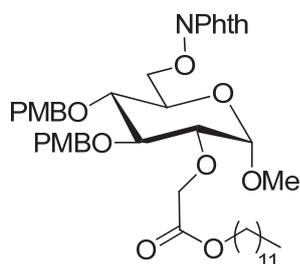
[α]_D = +56.7 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.40-7.22 (m, 4H, PMB), 7.00-6.81 (m, 4H, PMB), 4.91-4.35 (m, 7H, 3 \times OCH₂, H-1), 4.10 (t, *J* = 6.8 Hz, 2H, OCH₂), 3.91-3.73 (m, 7H, 2 \times OCH₃, H-3'), 3.73 (dd, *J* = 11.9, 2.8 Hz, 1H, H-6a), 3.65 (dd, *J* = 11.9, 3.2 Hz, 1H, H-6b), 3.57-3.43 (m, 3H, H-2,4,5), 3.26 (s, 3H, OCH₃), 1.79 (s, 1H, OH), 1.65-1.55 (m, 2H, CH₂), 1.40-1.18 (m, 18H, 9 \times CH₂), 0.85 (t, *J* = 6.8 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.4 (C=O), 159.5, 159.4, 130.4, 130.1 (C_q, PMB); 130.0, 129.8, 113.9 (CH, PMB); 98.0 (C-1), 83.3, 79.6, 76.4 (CH); 74.7, 73.0, 70.8 (CH₂); 70.5 (CH), 65.0, 61.9 (CH₂); 55.3, 55.2 (OCH₃); 32.0, 29.7, 29.6, 29.4, 29.3, 28.7, 25.9, 22.8 (CH₂); 14.2 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₇H₅₆NaO₁₀ 683.3771; Found 683.3775.

Methyl 2-*O*-dodecyloxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)-6-*O*-phthalimido- α -D-glucopyranoside (15)

Chemical Formula: C₄₅H₅₉NO₁₂

Exact Mass: 805.4037 g/mol

The phthalimido group was introduced at the 6-position of compound **14** (55 mg, 0.083 mmol) using Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **15** (66 mg, 98%) as a colourless paste.

$R_f = 0.77$ (petroleum ether/EtOAc: 1/1).

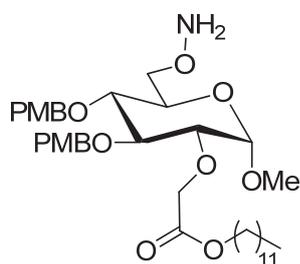
$[\alpha]_D = +65.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.88-7.72 (m, 4H, Phth), 7.36-7.26 (m, 4H, PMB), 6.91-6.82 (m, 4H, PMB), 4.98 (d, $J = 10.1$ Hz, 1H, OCH), 4.82 (d, $J = 10.1$ Hz, 1H, OCH), 4.75-4.33 (m, 7H, 2×OCH₂, H-1,6), 4.15 (t, $J = 6.9$ Hz, 2H, OCH₂), 3.91-3.72 (m, 9H, 2×OCH₃, H-3,4,5), 3.58 (dd, $J = 9.1, 3.4$ Hz, 1H, H-2), 3.35 (s, 3H, OCH₃), 1.70-1.57 (m, 2H, CH₂), 1.43-1.19 (m, 18H, 9×CH₂), 0.88 (t, $J = 6.8$ Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.3 (C=O), 163.3 (C=O, Phth); 159.5, 159.3 (C_q); 134.6 (CH, Phth), 130.6, 130.2 (C_q); 130.1, 129.9 (CH, PMB); 129.0 (C_q), 123.6 (CH, Phth), 113.93, 113.88 (CH, PMB); 98.2 (C-1), 83.4, 79.1 (CH); 76.4 (CH₂), 76.0 (CH), 74.9, 73.1, 70.9 (CH₂); 69.1 (CH), 65.1 (CH₂), 55.6, 55.3 (OCH₃); 32.0, 29.8, 29.7, 29.6, 29.5, 29.4, 28.7, 26.0, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : $[M+K]^+$ Calcd for C₄₅H₅₉KNO₁₂ 844.3674; Found 844.3678.

Methyl 6-*O*-amino-2-*O*-dodecyloxycarbonylmethyl-3,4-di-*O*-(4-methoxybenzyl)- α -D-glucopyranoside (16)

Chemical Formula: C₃₇H₅₇NO₁₀

Exact Mass: 675.3982 g/mol

Hydrazinolysis of compound **15** (58 mg, 0.071 mmol) according to the general **procedure D** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **16** (35 mg, 73%) as a yellowish paste.

R_f = 0.39 (petroleum ether/EtOAc: 1/1).

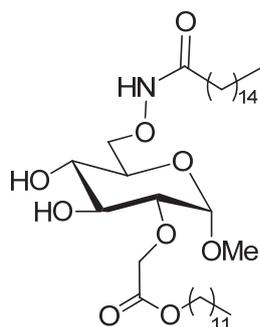
$[\alpha]_D = +45.3$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.34-7.24 (m, 4H, PMB), 6.93-6.84 (m, 4H, PMB), 4.93-4.34 (m, 7H, 3×OCH₂, H-1), 4.13 (t, J = 6.8 Hz, 2H, OCH₂), 3.91-3.74 (m, 9H, 2×OCH₃, H-3,6), 3.72-3.65 (m, 1H, H-5), 3.56-3.45 (m, 2H, H-2,4), 3.30 (s, 3H, OCH₃), 1.69-1.57 (m, 2H, CH₂), 1.39-1.17 (m, 18H, 9×CH₂), 0.88 (t, J = 6.8 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.4 (C=O), 159.5, 159.4, 130.5, 130.2 (C_q); 130.1, 129.9, 114.0 113.9 (CH, PMB); 98.1 (C-1), 83.5, 79.4, 76.7 (CH); 74.8, 74.3, 73.1, 70.9 (CH₂); 69.4 (CH), 65.0 (CH₂), 55.4, 55.3 (OCH₃); 32.0, 29.8, 29.7, 29.6, 29.5, 29.4, 28.7, 26.0, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₇H₅₇NaNO₁₀ 698.3880; Found 698.3884.

Methyl 2-*O*-dodecyloxycarbonylmethyl-6-*O*-palmitoylamino- α -D-glucopyranoside (17)

Chemical Formula: C₃₇H₇₁NO₉

Exact Mass: 673.5129 g/mol

To a solution of compound **11** (29 mg, 0.03 mmol) in CH₂Cl₂ (1 mL), was added TFA (0.05 mL, 5% in CH₂Cl₂). The mixture was stirred at room temperature for 1 h under argon and then extracted with CH₂Cl₂ (3×10 mL). The combined organic layers were washed with brine, dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (petroleum ether/EtOAc: 1/1) to afford compound **17** (12 mg, 60%) as a white solid.

R_f = 0.44 (petroleum ether/EtOAc: 1/1).

[α]_D = +80.0 (*c* 0.1, CHCl₃).

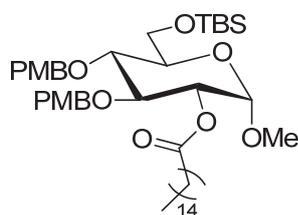
M.p. 70 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.74 (s, 1H, NH), 4.80 (d, *J* = 3.2 Hz, 1H, H-1), 4.48 (s, 2H, OCH₂), 4.21-4.08 (m, 4H, OCH₂, H-6), 3.88 (t, *J* = 9.3 Hz, 1H, H-4), 3.76-3.63 (m, 2H, H-2,5), 3.51 (t, *J* = 9.0 Hz, 1H, H-3), 3.42 (s, 3H, OCH₃), 2.09 (t, *J* = 7.4 Hz, 2H, CH₂), 1.70-1.56 (m, 4H, 2×CH₂), 1.38-1.19 (m, 42H, 21×CH₂), 0.92-0.82 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 173.8, 171.8 (C=O); 99.7 (C-1), 83.8 (CH), 75.9 (CH₂), 71.8, 70.3 (CH); 69.3, 65.9 (CH₂); 55.6 (OCH₃), 33.3, 32.1, 29.8, 29.7, 29.6, 29.5, 29.4, 29.3, 28.6, 25.9, 25.4, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₇H₇₁NaNO₉ 696.5027; Found 696.5025.

Methyl 3,4-di-*O*-(4-methoxybenzyl)-2-*O*-palmitoyl-6-*O*-*tert*-butyldimethylsilyl- α -D-glucopyranoside (18)



Chemical Formula: C₄₅H₇₄O₉Si

Exact Mass: 786.5102 g/mol

Esterification of compound **4** (0.62 g, 1.13 mmol) with palmitic acid according to the general **procedure G** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 8/1) afforded compound **18** (0.88 g, 99%) as a colourless paste.

R_f = 0.78 (petroleum ether/EtOAc: 3/1).

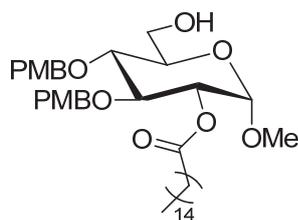
$[\alpha]_D = +26.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.25-7.14 (m, 4H, PMB), 6.94-6.81 (m, 4H, PMB), 5.53 (t, J = 9.6 Hz, 1H, H-3), 4.61-4.47 (m, 5H, 2×OCH₂, H-1), 3.90-3.74 (m, 8H, 2×OCH₃, H-6), 3.68-3.62 (m, 1H, H-5), 3.52 (t, J = 9.6 Hz, 1H, H-4), 3.43-3.36 (m, 1H, H-2), 3.33 (s, 3H, OCH₃), 2.26-2.18 (m, 2H, CH₂), 1.68-1.55 (m, 2H, CH₂), 1.50-1.16 (m, 24H, 12×CH₂), 0.98-0.82 (m, 12H, *t*-Bu, CH₃), 0.11-0.05 (m, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 172.7 (C=O), 159.4, 159.3, 130.4, 130.3 (C_q, PMB); 129.5, 129.4, 113.9 (CH, PMB); 97.8 (C-1), 77.7, 76.0 (CH); 73.8 (CH₂), 73.5 (CH), 72.6 (CH₂), 71.2 (CH), 62.2 (CH₂), 55.4, 55.0 (OCH₃); 34.7, 32.1, 29.8, 29.6, 29.5, 29.4 (CH₂); 26.1 (CH₃, *t*-Bu), 25.1, 22.8 (CH₂); 18.5 (C_q, *t*-Bu), 14.3 (CH₃), -5.0, -5.2 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₄₅H₇₄NaO₉Si 809.5000; Found 809.5005.

Methyl 3,4-di-*O*-(4-methoxybenzyl)-2-*O*-palmitoyl- α -D-glucopyranoside (19)

Chemical Formula: C₃₉H₆₀O₉

Exact Mass: 672.4237 g/mol

Desilylation of compound **18** (0.82 g, 1.04 mmol) according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **19** (0.48 g, 67%) as a white paste.

R_f = 0.13 (petroleum ether/EtOAc = 3/1).

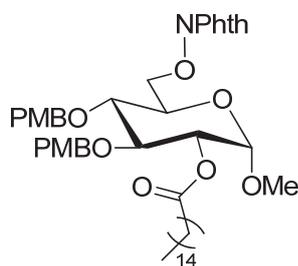
$[\alpha]_D = +23.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.26-7.17 (m, 4H, PMB), 6.91-6.82 (m, 4H, PMB), 5.54 (t, J = 9.6 Hz, 1H, H-3), 4.62-4.45 (m, 5H, 2×OCH₂, H-1), 3.81, 3.79 (2×s, 6H, 2×OCH₃), 3.77-3.65 (m, 3H, H-5,6), 3.56 (t, J = 9.5 Hz, 1H, H-4), 3.40 (dd, J = 10.0, 3.5 Hz, 1H, H-2), 3.33 (s, 3H, OCH₃), 2.29-2.21 (m, 2H, CH₂), 1.67-1.56 (m, 2H, CH₂), 1.39-1.18 (m, 24H, 12×CH₂), 0.88 (t, J = 6.8 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.7 (C=O), 159.5, 130.2, 130.1 (C_q, PMB); 129.7, 129.6, 113.9 (CH, PMB); 98.1 (C-1), 77.4, 75.7 (CH); 74.0 (CH₂), 73.4 (CH), 72.7 (CH₂), 70.4 (CH), 61.8 (CH₂), 55.4 (OCH₃), 34.7, 32.1, 29.8, 29.7, 29.5, 29.4, 25.1, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₉H₆₀NaO₉ 695.4135; Found 695.4133.

Methyl 3,4-di-*O*-(4-methoxybenzyl)-2-*O*-palmitoyl-6-*O*-phthalimido- α -D-glucopyranoside (20)

Chemical Formula: C₄₇H₆₃NO₁₁

Exact Mass: 817.4401 g/mol

The phthalimido group was introduced at the 6-position of compound **19** (0.43 g, 0.64 mmol) using Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **20** (0.51 g, 98%) as a colourless paste.

R_f = 0.62 (petroleum ether/EtOAc: 1/1).

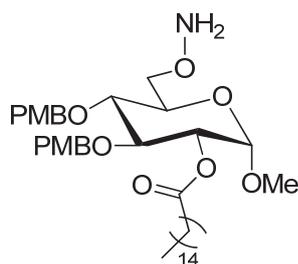
$[\alpha]_D = +48.3$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.93-7.73 (m, 4H, Phth), 7.35-7.21 (m, 4H, PMB), 6.94-6.81 (m, 4H, PMB), 5.57 (t, J = 9.6 Hz, 1H, H-3), 4.83-4.38 (m, 7H, 2×OCH₂, H-1,6), 3.99 (t, J = 9.4 Hz, H-4), 3.93-3.84 (m, 1H, H-5), 3.80, 3.77 (2×s, 6H, 2×OCH₃), 3.52 (dd, J = 10.1, 3.4 Hz, 1H, H-2), 3.34 (s, 3H, OCH₃), 2.27 (t, J = 7.7 Hz, 2H, CH₂), 1.72-1.56 (m, 2H, CH₂), 1.49-1.17 (m, 24H, 12×CH₂), 0.88 (t, J = 6.8 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.7 (C=O), 163.3 (C=O, Phth); 159.5, 159.4 (C_q); 134.6 (CH, Phth), 130.3 (C_q); 129.8, 129.6 (CH, PMB); 129.0 (C_q), 123.7 (CH, Phth), 113.9 (CH, PMB), 98.3 (C-1), 77.0 (CH), 76.0 (CH₂), 75.2 (CH), 74.2 (CH₂), 73.5 (CH), 72.8 (CH₂), 69.4 (CH), 55.7, 55.4 (OCH₃); 34.7, 32.1, 29.8, 29.7, 29.5, 29.4, 25.1, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₄₇H₆₃NaNO₁₁ 840.4299; Found 840.4296.

Methyl 6-*O*-amino-3,4-di-*O*-(4-methoxybenzyl)-2-*O*-palmitoyl- α -D-glucopyranoside (21**)**

Chemical Formula: C₃₉H₆₁NO₉

Exact Mass: 687.4346 g/mol

Hydrazinolysis of compound **20** (0.41 g, 0.50 mmol) according to the general **procedure D** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **21** (0.28 g, 82%) as a white paste.

R_f = 0.25 (petroleum ether/EtOAc: 1/1).

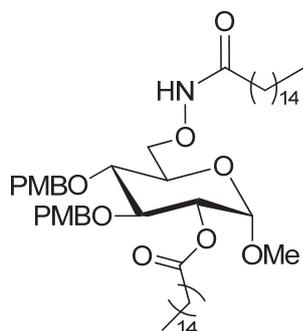
$[\alpha]_D = +31.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.26-7.17 (m, 4H, PMB), 6.94-6.82 (m, 4H, PMB), 5.52 (t, J = 9.6 Hz, 1H, H-3), 4.62-4.45 (m, 5H, 2×OCH₂, H-1), 3.94-3.76 (m, 9H, 2×OCH₃, H-5,6), 3.56-3.47 (m, 1H, H-4), 3.42 (dd, J = 10.0, 3.4 Hz, 1H, H-2), 3.34 (s, 3H, OCH₃), 2.30-2.21 (m, 2H, CH₂), 1.69-1.56 (m, 2H, CH₂), 1.42-1.17 (m, 24H, 12×CH₂), 0.88 (t, J = 6.8 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.6 (C=O), 159.5, 159.4, 130.1 (C_q); 129.7, 129.6, 113.9 (CH, PMB); 98.0 (C-1), 77.4, 76.0 (CH); 74.3, 73.9 (CH₂); 73.3 (CH), 72.7 (CH₂), 69.3 (CH), 55.4 (OCH₃), 34.7, 32.0, 29.8, 29.6, 29.5, 29.4, 25.1, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : [M+K]⁺ Calcd for C₃₉H₆₁KNO₉ 726.3983; Found 726.3936.

Methyl 3,4-di-*O*-(4-methoxybenzyl)-2-*O*-palmitoyl-6-*O*-palmitoylamino- α -D-glucopyranoside (22**)**

Chemical Formula: C₅₅H₉₁NO₁₀

Exact Mass: 925.6643 g/mol

Compound **22** was prepared from compound **21** (0.34 g, 0.50 mmol) and palmitic acid according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **22** (0.35 g, 76%) as a white solid.

R_f = 0.56 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = +32.7$ (c 0.1, CHCl₃).

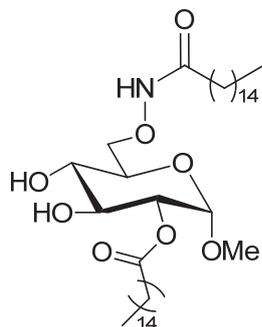
M.p. 90 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.03 (s, 1H, NH), 7.35-7.12 (m, 4H, PMB), 6.95-6.82 (m, 4H, PMB), 5.52 (t, J = 9.6 Hz, 1H, H-3), 4.68-4.45 (m, 5H, 2×OCH₂, H-1), 4.20-3.96 (m, 2H, H-6), 3.90-3.75 (m, 7H, 2×OCH₃, H-5), 3.70-3.58 (m, 1H, H-4), 3.45-3.40 (m, 1H, H-2), 3.34 (s, 3H, OCH₃), 2.30-1.94 (m, 4H, 2×CH₂), 1.70-1.54 (m, 4H, 2×CH₂), 1.50-1.15 (m, 48H, 24×CH₂), 0.95-0.84 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.7 (C=O), 159.6, 130.1, 130.0 (C_q); 129.6, 114.0 (CH, PMB); 98.1 (C-1), 77.4, 75.8 (CH); 74.8, 74.0 (CH₂); 73.4 (CH), 72.8 (CH₂), 69.4 (CH), 55.6, 55.4 (OCH₃); 34.7, 33.5, 32.1, 29.8, 29.7, 29.5, 29.4, 25.1, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₅₅H₉₁NaNO₁₀ 948.6541; Found 948.6542.

Methyl 2-*O*-palmitoyl-6-*O*-palmitoylamino- α -D-glucopyranoside (23**)**

Chemical Formula: C₃₉H₇₅NO₈

Exact Mass: 685.5493

To a solution of compound **22** (150 mg, 0.16 mmol) in CH₂Cl₂ (4 mL), was added TFA (2 mL, 5% in CH₂Cl₂). The mixture was stirred at room temperature for 1 h under argon and then extracted with CH₂Cl₂ (3×20 mL). The combined organic layers were washed with brine, dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/3) to afford compound **23** (85 mg, 77%) as a white powder.

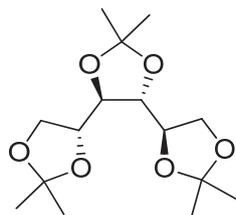
R_f = 0.28 (CH₂Cl₂/MeOH: 100/3).[α]_D = +84.7 (*c* 0.1, CHCl₃).

M.p. 96 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.55 (s, 1H, NH), 5.14 (t, *J* = 9.7 Hz, 1H, H-3'), 4.77 (d, 1H, *J* = 3.6 Hz, 1H, H-1'), 4.25-4.13 (m, 2H, H-6), 3.96 (t, *J* = 9.6 Hz, 1H, H-4), 3.83-3.73 (m, 1H, H-5), 3.67-3.56 (m, 1H, H-2), 3.43 (s, 3H, OCH₃), 2.41 (t, *J* = 7.6 Hz, 2H, CH₂), 2.09 (t, *J* = 7.3 Hz, 2H, CH₂), 1.73-1.57 (m, 4H, 2×CH₂), 1.45-1.19 (m, 48H, 24×CH₂), 0.94-0.84 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 175.0, 171.9 (C=O); 100.0 (C-1), 75.5 (CH₂), 75.0, 71.4, 70.7, 68.1 (CH); 55.7 (OCH₃), 34.6, 33.2, 32.1, 29.8, 29.6, 29.5, 29.4, 29.2, 25.3, 25.1, 22.8 (CH₂); 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₉H₇₅NaNO₈ 708.5390; Found 708.5392.

1,2:3,4:5,6-Tri-*O*-isopropylidene-D-mannitol (24)Chemical Formula: C₁₅H₂₆O₆

Exact Mass: 302.1729 g/mol

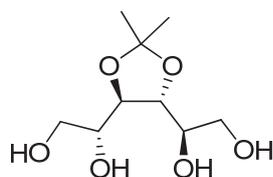
To a suspension of D-mannitol (20 g, 109.8 mmol) in dry acetone (600 mL) was added conc. H₂SO₄ (98%, 6 mL) and stirred for 6 h at room temperature. Reaction was neutralized by saturated aq NaOH (100 mL) and the solvent was removed in vacuo. The residue was diluted with EtOAc (150 mL) and saturated with NaCl (10 g). The organic layer was separated and the remaining aqueous layer was extracted with EtOAc (3×50 mL). The combined organic solution was washed with sat.aq. NaOH (1×25 mL), H₂O (1×25 mL) and brine (1×25 mL) and dried. Removal of the solvent gave the crude product which was purified by column chromatography (petroleum ether/EtOAc: 20/1) to afford compound **24** (29.80 g, 90%) as a white solid.

R_f = 0.40 (petroleum ether/EtOAc: 10/1).

¹H NMR (400 MHz, CDCl₃): δ (ppm) 4.26-4.15 (m, 2H, 2×CH), 4.07 (dd, *J* = 8.5, 6.4 Hz, 2H, 2×CH), 4.01-3.91 (m, 4H, 2×CH₂), 1.41 (s, 6H, 2×CH₃), 1.38 (s, 6H, 2×CH₃), 1.34 (s, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ (ppm) 110.3, 109.7 (C_q); 79.5, 76.5 (CH); 66.4 (CH₂); 27.6, 26.7, 25.5 (CH₃).

Ref.: (a) Yadav, V. K.; Agrawal, D. *Chem. Commun.* **2007**, 5232. (b) Mannoek, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (Known structure)

3,4-O-Isopropylidene-D-mannitol (25)Chemical Formula: C₉H₁₈O₆

Exact Mass: 222.1103 g/mol

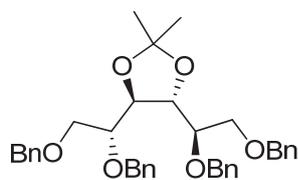
To a stirred solution of 30% aq AcOH (200 mL), was added D-mannitol triacetone **24** (10 g, 33.1 mmol). The mixture was stirred at 40 °C. After 1 h, the solution was evaporated and residual AcOH removed by repeated co-evaporation with toluene. Dry acetone was added and the mixture was thoroughly stirred with excess anhydrous K₂CO₃ and then filtered. The insoluble material (remaining mannitol) was washed with acetone and the combined filtrates evaporated. The residue was then dissolved in a small volume of EtOAc and allowed to crystallize at room temperature to give pure product **25** (5.89 g, 80.1%) as a crystal.

R_f = 0.43 (CH₂Cl₂/MeOH: 10/1).

¹H NMR (400 MHz, acetone-*d*₆): δ 4.73 (s, 2H, 2×OH), 3.96-3.90 (m, 2H, 2×CH), 3.77-3.54 (m, 8H, 2×CH, 2×CH₂, 2×OH), 1.32 (s, 6H, 2×CH₃).

¹³C NMR (100 MHz, acetone-*d*₆): δ 109.8 (C_q), 80.8, 74.2 (CH); 64.6 (CH₂), 27.3 (CH₃).

Ref.: van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannoek, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (Known structure)

1,2,5,6-Tetra-*O*-benzyl-3,4-*O*-isopropylidene-D-mannitol (26)Chemical Formula: C₃₇H₄₂O₆

Exact Mass: 582.2981 g/mol

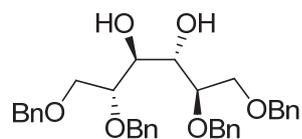
NaH (60%, 6.36 g, 159 mmol) was added to a stirred solution of compound **25** (5.88 g, 26.49 mmol) in dry DMF (200 mL). The suspension was stirred for 0.5 h at 0 °C in an ice bath. BnBr (15.11 mL, 127 mmol) was then added and the stirring continued at room temperature for 15 h. Ice was added to destroy the excess NaH, the solution was then concentrated, diluted with saturated aq NH₄Cl and the product extracted with EtOAc (200 mL). The extract was washed with water, brine, dried over MgSO₄ and evaporated. Purification by column chromatography (petroleum ether/EtOAc: 15/1) afforded compound **26** (15.10 g, 98%) as a colorless syrup.

R_f = 0.31 (petroleum ether/EtOAc: 10/1).

¹H NMR (400 MHz, CDCl₃): δ 7.41-7.17 (m, 20H, H-Ph), 4.73 (d, *J* = 11.8 Hz, 2H, 2×CH), 4.57 (d, *J* = 11.7 Hz, 2H, 2×CH), 4.48-4.46 (m, 4H, 2×CH₂), 4.24-4.15 (m, 2H, 2×CH), 3.81-3.55 (m, 6H, 2×CH₂, 2×CH), 1.35 (s, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 138.6, 138.5 (C_q); 128.5, 128.4, 128.0, 127.7, 127.6 (CH-Ph); 109.9 (C_q); 79.4, 78.6 (CH); 73.4, 72.9, 70.7 (CH₂); 27.3 (CH₃).

Ref.: (a) van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannoek, D. A.; Lewis, R. N.; McElhane, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (Known structure)

1,2,5,6-Tetra-*O*-benzyl-D-mannitol (27)Chemical Formula: C₃₄H₃₈O₆

Exact Mass: 542.2668 g/mol

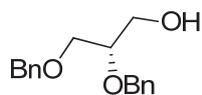
Compound **26** (15.10 g, 25.96 mmol) was treated with 1M aq HCl/MeOH (1:9, v/v, 250 mL) under reflux for 4 h, when TLC showed a new single spot. Excess solid NaHCO₃ was added and the solution evaporated to give an oil which was dissolved in EtOAc (300 mL), dried over MgSO₄ and evaporated. Purification by column chromatography (petroleum ether/EtOAc: 5/1) afforded compound **27** (13.08 g, 93%) as a colorless syrup.

R_f = 0.12 (petroleum ether/EtOAc: 5/1).

¹H NMR (400 MHz, CDCl₃): δ 7.34-7.26 (m, 20H, H-Ph), 4.72 (d, *J* = 11.5 Hz, 2H, 2×CH), 4.59 (d, *J* = 11.5 Hz, 2H, 2×CH), 4.54 (s, 4H, 2×CH₂), 3.96 (m, 2H, 2×CH), 3.80-3.65 (m, 6H, 2×CH₂, 2×CH), 3.04 (s, 1H, OH), 3.02 (s, 1H, OH).

¹³C NMR (100 MHz, CDCl₃): δ 138.3, 138.1 (C_q); 128.5, 128.1, 127.9, 127.8 (CH-Ph); 79.3 (CH); 73.6, 73.2, 70.3 (CH₂); 70.1 (CH).

Ref.: van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannoek, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (Known structure)

(2S)-1,2-Di-O-benzyl-glycerol (28)Chemical Formula: C₁₇H₂₀O₃

Exact Mass: 272.1412 g/mol

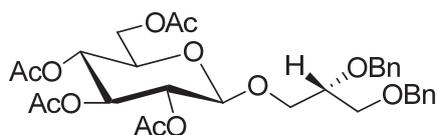
To a solution of compound **27** (14.10 g, 26 mmol) in MeOH (250 mL), was added an aq solution of NaIO₄ (8.56 g, 40 mmol in 150 mL water). The reaction mixture was stirred for 3 h at room temperature, after which time TLC analysis revealed the oxidation to be complete ($R_f = 0.52$, petroleum ether/EtOAc: 3/1). The reaction mixture was diluted with MeOH (200 mL) and cooled. The precipitate was filtered off and to the filtrate was added NaBH₄ (9.84 g, 260 mmol). After 1 h, the reaction mixture was treated with AcOH and the solution evaporated to a small volume which was diluted in CH₂Cl₂ (300 mL), dried over MgSO₄ and concentrated. Purification by column chromatography (petroleum ether/EtOAc: 3/1) afforded compound **28** (13.87 g, 98%) as a colorless syrup.

$R_f = 0.24$ (petroleum ether/EtOAc: 3/1).

¹H NMR (400 MHz, CDCl₃): δ 7.38-7.26 (m, 10H, H-Ph), 4.74-4.53 (m, 4H, 2×CH₂), 3.79-3.57 (m, 5H, 2×CH₂, CH), 2.05 (s, 1H, OH).

¹³C NMR (100 MHz, CDCl₃): δ 138.4, 138.1 (C_q); 128.6, 128.6, 128.0, 127.9, 127.8 (CH-Ph); 78.1 (CH); 73.7, 72.3, 70.3, 63.0 (CH₂).

Ref.: van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannoek, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (Known structure)

(2R)-1,2-Di-O-benzyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (29)Chemical Formula: C₃₁H₃₈O₁₂

Exact Mass: 602.2363 g/mol

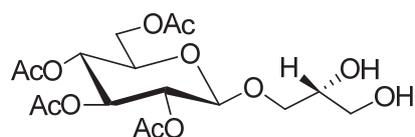
To a solution of α-D-glucose pentaacetate (2 g, 5.12 mmol) in anhydrous CH₂Cl₂ (15 mL) was added HBr (33% in AcOH, 7 mL, 41 mmol) at 0 °C. The mixture was stirred at room temperature under argon for 6 h, and then the solution neutralized with aq NaHCO₃. The resulting 2,3,4,6-tetra-O-acetyl-α-D-glucopyranosyl bromide was extracted with CH₂Cl₂ (100 mL), dried over MgSO₄ and evaporated to give a syrup. To a solution of (2S)-1,2-di-O-benzyl-glycerol (**28**) (1.41 g, 5.2 mmol), HgBr₂ (0.94 g, 2.6 mmol) and Hg(CN)₂ (0.66 g, 2.6 mmol) in dry acetonitrile (20 mL) was added dropwise over a period of 1 h, a solution of glycosyl bromide in acetonitrile (10 mL). The reaction solution was stirred at room temperature overnight, and then concentrated to an oil which was dissolved in CH₂Cl₂ (100 mL) and washed with saturated aq NaHCO₃ (2×30 mL) and water (30 mL). The dried (MgSO₄) organic layer was concentrated. Purification by column chromatography (petroleum ether/EtOAc: 3/1) afforded compound **29** (2.30 g, 75%) as a colorless paste.

R_f = 0.39 (petroleum ether/EtOAc: 2/1).

¹H NMR (400 MHz, CDCl₃): δ 7.37-7.28 (m, 10H, H-Ph), 5.18 (t, *J* = 9.4 Hz, 1H, H-3'), 5.09 (t, *J* = 9.7 Hz, 1H, H-4'), 5.00 (dd, *J* = 9.5, 8.2 Hz, 1H, H-2'), 4.65 (s, 2H, CH₂), 4.57-4.50 (m, 3H, H-1', CH₂), 4.28-4.09 (m, 2H, H-6'), 3.99-3.69 (m, 3H, H-2,3), 3.65 (ddd, *J* = 10.0, 4.5, 2.3 Hz, 1H, H-5'), 3.59-3.57 (m, 2H, H-1), 2.07, 2.04, 2.03, 2.01 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.5, 169.6, 169.5 (C=O); 138.5, 138.3 (C_q); 128.6, 128.5 (CH-Ph); 101.2 (C-1'), 73.6 (CH₂), 72.9 (C-3'), 72.3 (CH₂), 71.9 (C-5'), 71.4 (C-2'), 69.8, 69.4 (CH₂); 68.5 (C-4'), 62.0 (C-6'), 20.9, 20.8 (OAc).

Ref.: van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannock, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (c) Janwitayanuchit, W.; Suwanborirux, K.; Patarapanich, C.; Pummangura, S.; Lipipun, V.; Vilaivan, T. *Phytochemistry* **2003**, *64*, 1253-1264. (Known structure)

(2R)-1-O-(2',3',4',6'-Tetra-O-acetyl-β-D-glucopyranosyl)glycerol (30)Chemical Formula: C₁₇H₂₆O₁₂

Exact Mass: 422.1424 g/mol

A mixture of compound **29** (12 g, 20 mmol) and 10% Pd/C (1.8 g) in EtOAc (100 mL) containing 5 mL each of EtOH and AcOH was vigorously shaken under H₂ at room temperature overnight. The catalyst was filtered off, the filtrate evaporated. Purification by column chromatography (CH₂Cl₂/MeOH: 25/1) afforded compound **30** (6 g, 71%) as a white solid.

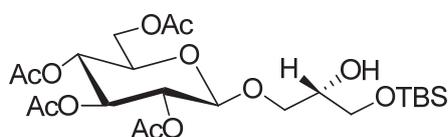
R_f = 0.12 (CH₂Cl₂/MeOH: 25/1).

¹H NMR (400 MHz, CDCl₃): δ 5.22 (t, *J* = 9.6 Hz, 1H, H-3'), 5.11-5.04 (m, 1H, H-4'), 5.04-4.97 (m, 1H, H-2'), 4.55 (d, *J* = 7.8 Hz, 1H, H-1'), 4.25-4.16 (m, 2H, H-6'), 3.89-3.79 (m, 3H, H-2,3), 3.77-3.57 (m, 3H, H-1,5'), 2.32 (s, 2H, 2×OH), 2.11, 2.07, 2.04, 2.01 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.7, 169.5 (C=O); 101.4 (C-1'), 72.6 (C-3'), 72.0 (C-5'), 71.3 (C-2'), 70.5 (CH), 68.4 (C-4'), 63.4, 61.9 (CH₂); 20.8, 20.7 (OAc).

Ref.: van Boeckel, C.; Visser, G.; van Boom, J. *Tetrahedron* **1985**, *41*, 4557-4565. (b) Mannock, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113-127. (c) Janwitayanuchit, W.; Suwanborirux, K.; Patarapanich, C.; Pummangura, S.; Lipipun, V.; Vilaivan, T. *Phytochemistry* **2003**, *64*, 1253-1264. (Known structure)

(2*S*)-1-*O*-*tert*-Butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (31)



Chemical Formula: C₂₃H₄₀O₁₂Si

Exact Mass: 536.2289 g/mol

To a solution of compound **30** (2 g, 4.74 mmol), *N*-methylimidazole (1.14 mL, 14.22 mmol) and iodine (3.61 g, 14.22 mmol) in anhydrous THF (20 mL) under argon at 0 °C, was added TBSCl (0.86 g, 5.69 mmol). The reaction mixture was stirred for 20 min at 0 °C, then quenched with saturated aq Na₂S₂O₃ and diluted with EtOAc (50 mL). The aqueous layers were extracted with EtOAc (2×50 mL). The combined organic layers were dried over MgSO₄, filtered, concentrated, and purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/1) to afford compound **31** (2.43 g, 95.7%) as a yellowish paste.

R_f = 0.74 (CH₂Cl₂/MeOH: 100/3).

[α]_D = -9.7 (*c* = 0.1, CHCl₃).

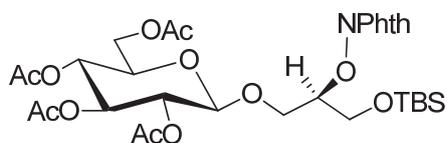
¹H NMR (400 MHz, CDCl₃): δ 5.20 (dd, *J* = 12.0, 6.9 Hz, 1H, H-3'), 5.05 (t, *J* = 9.7 Hz, 1H, H-4'), 4.98 (dd, *J* = 9.6, 8.1 Hz, 1H, H-2'), 4.54 (d, *J* = 7.9 Hz, 1H, H-1'), 4.25-4.11 (m, 2H, H-6'), 3.82-3.68 (m, 4H, H-2,3,5'), 3.62-3.58 (m, 2H, H-1), 2.08, 2.04, 2.01, 1.99 (4×s, 12H, 4×OAc), 0.88 (s, 9H, *t*-Bu), 0.05 (s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.7, 170.3, 169.5, 169.4 (C=O); 101.4 (C-1'), 72.8 (C-3'), 71.9 (C-5'), 71.7 (CH₂), 71.4 (C-2'), 70.5 (CH), 68.5 (C-4'), 63.7, 62.0 (CH₂); 25.9, 20.8, 20.7 (CH₃); 18.3 (C_q, *t*-Bu), -5.4 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₂₃H₄₁O₁₂Si 537.2367; Found 537.2362

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-Phthalimido-1-*O*-*tert*-butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (32**)**



Chemical Formula: C₃₁H₄₃NO₁₄Si

Exact Mass: 681.2453 g/mol

The phthalimido group was introduced to compound **31** (2.10 g, 3.91 mmol) via Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **32** (2.39 g, 90%) as a colourless paste.

$R_f = 0.53$ (petroleum ether/EtOAc: 1/1).

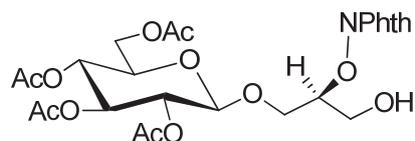
$[\alpha]_D = -13.3$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.83-7.70 (m, 4H, Phth), 5.16 (t, $J = 9.6$ Hz, 1H, H-3'), 5.02 (t, $J = 9.8$ Hz, 1H, H-4'), 4.92 (dd, $J = 9.4, 8.1$ Hz, 1H, H-2'), 4.66 (d, $J = 7.8$ Hz, 1H, H-1'), 4.39-4.41 (m, 1H, H-2), 4.21 (dd, $J = 12.3, 4.8$ Hz, 1H, H-6'a), 4.15-4.06 (m, 2H, H-3a,6'b), 3.93 (dd, $J = 11.7, 6.2$ Hz, 1H, H-3b), 3.89 (d, $J = 4.7$ Hz, 2H, H-1), 3.69 (ddd, $J = 10.0, 4.7, 2.2$ Hz, 1H, H-5'), 2.06, 2.01, 1.99, 1.96 (4×s, 12H, 4×OAc), 0.77 (s, 9H, *t*-Bu), -0.02, -0.03 (2×s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.4, 169.6, 169.6 (C=O), 163.7 (C=O, Phth); 134.5, 129.0 (C_q), 123.6 (Phth), 100.8 (C-1'), 87.0 (C-2), 73.0 (C-3'), 71.8 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 68.0, 62.0, 61.9 (CH₂); 25.8, 20.8, 20.8, 20.7 (CH₃); 18.2 (C_q, *t*-Bu), -5.6 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+H]⁺ Calcd for C₃₁H₄₄NO₁₄Si 682.2531; Found 682.2526.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2S)-2-O-Phthalimido-1-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (33)Chemical Formula: C₂₅H₂₉NO₁₄

Exact Mass: 567.1588 g/mol

Desilylation of compound **32** (1.0 g, 1.47 mmol) using AcCl according to the general **procedure B** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) gave compound **33** (0.59 g, 72%) as a white powder.

$R_f = 0.23$ (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -17.3$ (c 0.1, CHCl₃).

M.p. 72 °C.

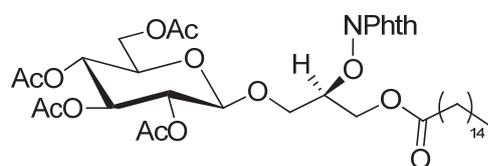
¹H NMR (400 MHz, CDCl₃): δ 7.87-7.74 (m, 4H, Phth), 5.22-5.15 (m, 1H, H-3'), 5.03 (t, $J = 10.1$ Hz, 1H, H-4'), 4.97 (m, 1H, H-2'), 4.64 (d, $J = 7.8$ Hz, 1H, H-1'), 4.32-4.26 (m, 1H, H-2), 4.22 (dd, $J = 12.3, 4.9$ Hz, 1H, H-6'a), 4.14-4.06 (m, 2H, H-3a,6'b), 4.01 (dd, $J = 11.6, 6.3$ Hz, 1H, H-3b), 3.83-3.66 (m, 3H, H-1,5'), 2.07, 2.04, 1.99, 1.97 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.6, 169.5 (C=O), 164.6 (C=O, Phth); 135.0, 128.8 (C_q); 124.0 (Phth), 100.9 (C-1'), 88.1 (CH), 72.8 (C-3'), 71.9 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.8, 61.9, 60.4 (CH₂); 20.8, 20.7, 20.7 (OAc).

HRMS (ESI) m/z : $[M+H]^+$ Calcd for C₂₅H₃₀NO₁₄ 568.1666; Found 568.1661.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-1-O-Palmitoyl-2-O-phthalimido-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (34)



Chemical Formula: C₄₁H₅₉NO₁₅

Exact Mass: 805.3885 g/mol

Esterification of compound **33** (0.58 g, 1.02 mmol) and palmitic acid according to the general **procedure G** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/1) afforded compound **34** (1.05 g, 98%) as a white solid.

R_f = 0.54 (petroleum ether/EtOAc: 1/1).

[α]_D = -11.0 (c 0.1, CHCl₃).

M.p. 65 °C.

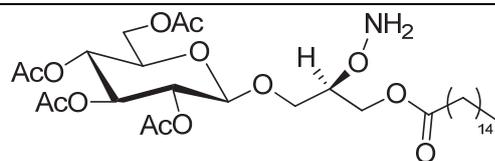
¹H NMR (400 MHz, CDCl₃): δ 7.87-7.74 (m, 4H, Phth), 5.20 (t, *J* = 9.5 Hz, 1H, H-3'), 5.06 (t, *J* = 9.7 Hz, 1H, H-4'), 4.96 (dd, *J* = 9.2, 7.8 Hz, 1H, H-2'), 4.68 (d, *J* = 8.2 Hz, 1H, H-1'), 4.62-4.53 (m, 1H, H-2), 4.44-4.31 (m, 2H, H-1), 4.25 (dd, *J* = 12.3, 4.8 Hz, 1H, H-6'a), 4.18-4.07 (m, 2H, H-3a,6'b), 4.01 (dd, *J* = 11.8, 5.8 Hz, 1H, H-3b), 3.73 (ddd, *J* = 9.9, 4.7, 2.4 Hz, 1H, H-5'), 2.38-2.24 (m, 2H, CH₂), 2.10, 2.05, 2.02, 1.99 (4×s, 12H, 4×OAc), 1.65-1.52 (m, 2H, CH₂), 1.35-1.20 (m, 24H, 12×CH₂), 0.88 (t, *J* = 6.9 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 173.5, 170.8, 170.4, 169.6, 169.5, 163.6 (C=O); 134.8, 128.9 (C_q); 123.8 (CH), 100.9 (C-1'), 84.5 (CH), 72.9 (C-3'), 72.0 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.8, 62.4, 62.0, 34.1, 32.1, 29.8, 29.8, 29.6, 29.5, 29.4, 29.3, 24.9, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₄₁H₆₀NO₁₅ 806.3963; Found 806.3957.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-2-O-Amino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (35)



Chemical Formula: C₃₃H₅₇NO₁₃

Exact Mass: 675.3830 g/mol

Hydrazinolysis of compound **34** (0.38 g, 0.47 mmol) by hydrazine under mild conditions according to the general **procedure E** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 3/2) afforded compound **35** (0.31 g, 96%) as a paste.

R_f = 0.40 (petroleum ether/EtOAc: 1/1).

[α]_D = -46.7 (c 0.1, CHCl₃).

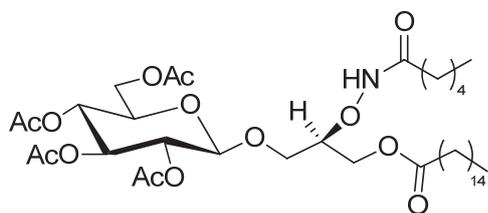
¹H NMR (400 MHz, CDCl₃): δ 5.22 (t, *J* = 8.7 Hz, 1H, H-3'), 5.06 (t, *J* = 9.6 Hz, 1H, H-4'), 4.96 (dd, *J* = 12.7, 4.9 Hz, 1H, H-2'), 4.57 (d, *J* = 7.8 Hz, 1H, H-1'), 4.39-4.06 (m, 4H, H-1,6'), 4.04-3.90 (m, 2H, H-3a,2), 3.79-3.65 (m, 2H, H-3b,5'), 2.32 (t, *J* = 7.5 Hz, 2H, CH₂), 2.09, 2.07, 2.03, 2.01 (4×s, 12H, 4×OAc), 1.68-1.57 (m, 2H, CH₂), 1.39-1.20 (m, 24H, 12×CH₂), 0.88 (t, *J* = 6.7 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 173.9, 170.8, 170.4, 169.5 (C=O); 101.2 (C-1'), 80.1 (CH), 72.8 (C-3'), 72.0 (C-5'), 71.3 (C-2'), 68.5 (C-4'), 68.0, 62.0, 61.9, 34.3, 32.1, 29.8, 29.6, 29.5, 29.4, 29.3, 25.1, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.2 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₃₃H₅₈NO₁₃ 676.3908; Found 676.3903.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-Hexanoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (36)



Chemical Formula: C₃₉H₆₇NO₁₄

Exact Mass: 773.4562 g/mol

Compound **36** was prepared from compound **35** (153 mg, 0.23 mmol) and hexanoic acid (29 μ L, 0.23 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **36** (145 mg, 82%) as a paste.

R_f = 0.60 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -35.7$ (c 0.1, CHCl₃).

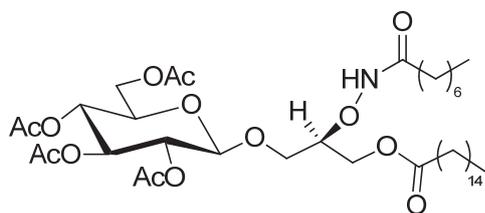
¹H NMR (400 MHz, CDCl₃): δ 8.83 (s, 1H, NH), 5.24 (t, J = 9.5 Hz, 1H, H-3'), 5.09 (t, J = 9.7 Hz, 1H, H-4'), 5.02 (dd, J = 9.6, 7.8 Hz, 1H, H-2'), 4.57 (d, J = 8.0 Hz, 1H, H-1'), 4.42 (dd, J = 12.2, 3.2 Hz, 1H, H-1a), 4.32-4.09 (m, 4H, H-2,1b,6'), 4.03-3.96 (m, 1H, H-3a), 3.81-3.68 (m, 2H, H-3b,5'), 2.39-2.33 (m, 2H, CH₂), 2.17-2.01 (m, 14H, 4 \times OAc, CH₂), 1.73-1.57 (m, 4H, 2 \times CH₂), 1.39-1.21 (m, 28H, 14 \times CH₂), 0.96-0.86 (m, 6H, 2 \times CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.4, 170.8, 170.3, 169.6 (C=O); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.5 (C-2'), 68.4 (C-4'), 67.9, 61.8, 61.4, 34.2, 33.2, 32.1, 31.5, 29.8, 29.8, 29.6, 29.5, 29.4, 29.2, 25.2, 25.0, 22.8, 22.5 (CH₂); 20.9, 20.7, 14.3, 14.1 (CH₃).

HRMS (ESI) m/z : $[M+H]^+$ Calcd for C₃₉H₆₈NO₁₄ 774.4640; Found 774.4634.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-Octanoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (37)



Chemical Formula: C₄₁H₇₁NO₁₄

Exact Mass: 801.4875 g/mol

Compound **37** was prepared from compound **35** (151 mg, 0.22 mmol) and octanoic acid (35 μ L, 0.22 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **37** (147 mg, 82%) as a paste.

R_f = 0.62 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -49.3$ (c 0.1, CHCl₃).

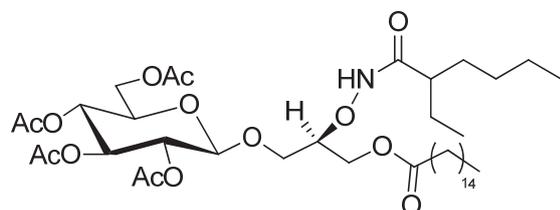
¹H NMR (400 MHz, CDCl₃): δ 8.81 (s, 1H, NH), 5.24 (t, J = 9.4 Hz, 1H, H-3'), 5.09 (t, J = 9.6 Hz, 1H, H-4'), 5.02 (t, J = 8.8 Hz, 1H, H-2'), 4.57 (d, J = 7.8 Hz, 1H, H-1'), 4.42 (d, J = 10.0 Hz, 1H, H-1a), 4.31-4.09 (m, 4H, H-2,1b,6'), 4.03-3.96 (m, 1H, H-3a), 3.79-3.68 (m, 2H, H-3b,5'), 2.36 (t, J = 7.2 Hz, 2H, CH₂), 2.16-2.00 (m, 14H, 4 \times OAc, CH₂), 1.68-1.58 (m, 4H, 2 \times CH₂), 1.40-1.20 (m, 32H, 16 \times CH₂), 0.93-0.84 (m, 6H, 2 \times CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.4, 170.8, 170.4, 170.3, 169.6 (C=O); 100.7 (C-1'), 82.5 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.5 (C-2'), 68.4 (C-4'), 67.9, 61.8, 61.4, 34.2, 33.2, 32.1, 31.8, 29.8, 29.6, 29.5, 29.4, 29.2, 29.1, 25.5, 25.0, 22.8, 22.7 (CH₂); 20.9, 20.7, 14.3 (CH₃).

HRMS (ESI) m/z : [M+H]⁺ Calcd for C₄₁H₇₂NO₁₄ 802.4953; Found 802.4947.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-(2-Ethylhexanoyl)amino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (38**)**



Chemical Formula: C₄₁H₇₁NO₁₄

Exact Mass: 801.4875 g/mol

Compound **38** was prepared from compound **35** (151 mg, 0.22 mmol) and 2-ethylhexanoic acid (35 μ L, 0.22 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **38** (114 mg, 64%) as a paste.

R_f = 0.47 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -38.0$ (c 0.1, CHCl₃).

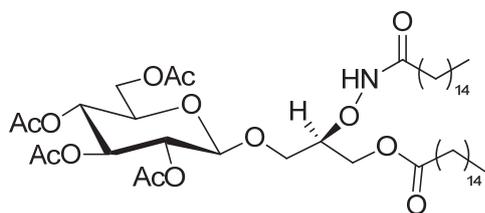
¹H NMR (400 MHz, CDCl₃): δ 8.87 (s, 1H, NH), 5.25 (t, J = 9.4 Hz, 1H, H-3'), 5.09 (t, J = 9.7 Hz, 1H, H-4'), 5.01 (t, J = 8.7 Hz, 1H, H-2'), 4.57 (d, J = 7.7 Hz, 1H, H-1'), 4.47-4.39 (m, 1H, H-1a), 4.31-4.10 (m, 4H, H-2,1b,6'), 4.03-3.96 (m, 1H, H-3a), 3.78-3.69 (m, 2H, H-3b,5'), 2.36 (t, J = 7.2 Hz, 2H, CH₂), 2.10, 2.09, 2.04, 2.02 (4 \times s, 12H, 4 \times OAc), 1.95-1.85 (m, 1H, CH), 1.69-1.57 (m, 4H, 2 \times CH₂), 1.52-1.39 (m, 2H, CH₂), 1.37-1.20 (m, 28H, 14 \times CH₂), 0.95-0.84 (m, 9H, 3 \times CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.4, 173.7, 170.8, 170.4, 170.2, 169.6 (C=O); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.0 (C-5'), 71.5 (C-2'), 68.3 (C-4'), 67.9, 61.8, 61.3 (CH₂); 45.7 (CH), 34.3, 34.2, 32.2, 32.0, 29.8, 29.6, 29.5, 29.4, 29.2, 25.9, 24.9, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.2, 14.1, 12.1 (CH₃).

HRMS (ESI) m/z : $[M+H]^+$ Calcd for C₄₁H₇₂NO₁₄ 802.4953; Found: 802.4947.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-1-*O*-Palmitoyl-2-*O*-palmitoylamino-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (39**)**



Chemical Formula: C₄₉H₈₇NO₁₄

Exact Mass: 913.6127 g/mol

Compound **39** was prepared from compound **35** (189 mg, 0.28 mmol) and palmitic acid (72 mg, 0.28 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **39** (192 mg, 76%) as a white solid.

R_f = 0.57 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -42.3$ (c 0.1, CHCl₃).

M.p. 74 °C.

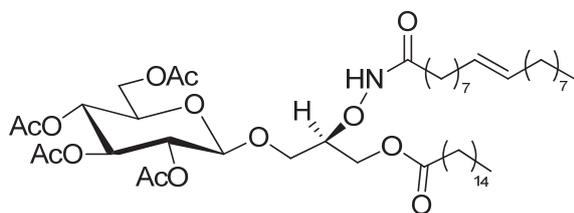
¹H NMR (400 MHz, CDCl₃): δ 8.86 (s, 1H, NH), 5.24 (t, J = 9.5 Hz, 1H, H-3'), 5.09 (t, J = 9.7 Hz, 1H, H-4'), 5.02 (t, J = 9.6, 7.8 Hz, 1H, H-2'), 4.57 (d, J = 7.9 Hz, 1H, H-1'), 4.42 (dd, J = 12.2, 3.3 Hz, 1H, H-1a), 4.32-4.08 (m, 4H, H-2,1b,6'), 4.03-3.96 (m, 1H, H-3a), 3.78-3.69 (m, 2H, H-3b,5'), 2.36 (t, J = 7.5 Hz, 2H, CH₂), 2.18-2.01 (m, 14H, 4×OAc, CH₂), 1.68-1.58 (m, 4H, 2×CH₂), 1.40-1.22 (m, 48H, 24×CH₂), 0.93-0.81 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.4, 171.2, 170.8, 170.2, 169.5 (C=O); 100.7 (C-1'), 82.1 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.4 (C-2'), 68.4 (C-4'), 67.8, 61.8, 61.3, 34.2, 33.3, 32.0, 29.8, 29.6, 29.5, 29.4, 29.2, 25.5, 24.9, 22.8 (CH₂); 20.8, 20.7, 14.2 (CH₃).

HRMS (ESI) m/z : [M+H]⁺ Calcd for C₄₉H₈₈NO₁₄ 914.6205; Found 914.6199.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-Oleoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl) glycerol (40**)**



Chemical Formula: C₅₁H₈₉NO₁₄

Exact Mass: 939.6283 g/mol

Compound **40** was prepared from compound **35** (151 mg, 0.22 mmol) and oleic acid (62 mg, 0.22 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **40** (180 mg, 87%) as a white solid.

R_f = 0.52 (petroleum ether/EtOAc: 1/1).

[α]_D = -35.3 (*c* 0.1, CHCl₃).

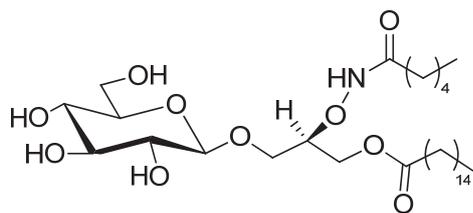
M.p. 57 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.83 (s, 1H, NH), 5.43-5.32 (m, 2H, CH=CH), 5.24 (t, *J* = 9.5 Hz, 1H, H-3'), 5.09 (t, *J* = 9.7 Hz, 1H, H-4'), 5.02 (dd, *J* = 9.6, 8.0 Hz, 1H, H-2'), 4.57 (d, *J* = 7.9 Hz, 1H, H-1'), 4.42 (dd, *J* = 12.2, 3.3 Hz, 1H, H-1a), 4.32-4.08 (m, 4H, H-2,1b,6'), 4.03-3.96 (m, 1H, H-3a), 3.78-3.69 (m, 2H, H-3b,5'), 2.36 (t, *J* = 7.5 Hz, 2H, CH₂), 2.20-1.98 (m, 16H, 4×OAc, 2×CH₂), 1.71-1.57 (m, 4H, 2×CH₂), 1.39-1.21 (m, 46H, 23×CH₂), 0.95-0.82 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.5, 170.8, 170.4, 170.3, 169.6 (C=O); 130.1, 129.8 (CH=CH); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.4 (C-2'), 68.4 (C-4'), 67.8, 61.8, 61.3, 34.2, 33.3, 32.0, 29.9, 29.8, 29.6, 29.5, 29.4, 29.2, 27.3, 25.5, 25.0, 22.8 (CH₂); 20.8, 20.7, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₅₁H₉₀NO₁₄ 940.6361; Found 940.6356.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-3-O-β-D-Glucopyranosyl-2-O-hexanoylamino-1-O-palmitoylglycerol (41)Chemical Formula: C₃₁H₅₉NO₁₀

Exact Mass: 605.4139 g/mol

Deacetylation of compound **36** (105 mg, 0.14 mmol) with hydrazine according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **41** (30 mg, 44%) as a white solid.

R_f = 0.43 (CH₂Cl₂/MeOH: 8/1).

[α]_D = -32.0 (c 0.1, MeOH).

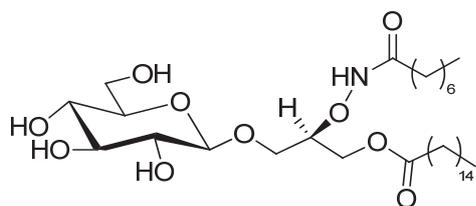
M.p. 94 °C.

¹H NMR (400 MHz, CD₃OD): δ 4.35-4.20 (m, 3H, H-1',1), 4.19-4.12 (m, 1H, H-2), 4.04 (dd, *J* = 11.4, 3.9 Hz, 1H, H-3a), 3.86 (d, *J* = 11.7 Hz, 1H, H-6'a), 3.75 (dd, *J* = 11.4, 6.1 Hz, 1H, H-3b), 3.67-3.61 (m, 1H, H-6'b), 3.40-3.24 (m, 3H, H-3',4',5'), 3.22-3.15 (m, 1H, H-2'), 2.34 (t, *J* = 7.4 Hz, 2H, CH₂), 2.07 (t, *J* = 7.4 Hz, 2H, CH₂), 1.70-1.55 (m, 4H, 2×CH₂), 1.45-1.22 (m, 28H, 14×CH₂), 0.97-0.85 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.2, 173.2 (C=O); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.3, 62.7, 34.9, 33.7, 33.1, 32.4, 30.8, 30.6, 30.5, 30.2, 26.3, 25.9, 23.7, 23.4 (CH₂); 14.5, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₃₁H₆₀NO₁₀ 606.4217; Found: 606.4212.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-3-O-β-D-Glucopyranosyl-2-O-octanoylamino-1-O-palmitoylglycerol (42)Chemical Formula: C₃₃H₆₃NO₁₀

Exact Mass: 633.4452 g/mol

Deacetylation of compound **37** (95 mg, 0.12 mmol) with hydrazine according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **42** (35 mg, 47%) as a white solid.

R_f = 0.26 (CH₂Cl₂/MeOH: 10/1).

[α]_D = -21.7 (c 0.1, MeOH).

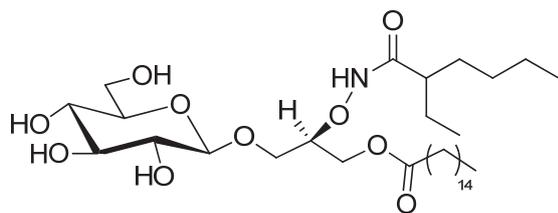
M.p. 96 °C

¹H NMR (400 MHz, CD₃OD): δ 4.37-4.19 (m, 3H, H-1',1), 4.19-4.12 (m, 1H, H-2), 4.04 (dd, *J* = 11.4, 3.8 Hz, 1H, H-3a), 3.86 (d, *J* = 11.6 Hz, 1H, H-6'a), 3.75 (dd, *J* = 11.4, 6.2 Hz, 1H, H-3b), 3.63 (dd, *J* = 9.8, 7.2 Hz, 1H, H-6'b), 3.40-3.24 (m, 3H, H-3',4',5'), 3.22-3.16 (m, 1H, H-2'), 2.34 (t, *J* = 7.4 Hz, 2H, CH₂), 2.07 (t, *J* = 7.4 Hz, 2H, CH₂), 1.69-1.54 (m, 4H, 2×CH₂), 1.48-1.22 (m, 32H, 16×CH₂), 1.01-0.81 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.2, 173.2 (C=O); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.3, 62.7, 34.9, 33.8, 33.1, 32.9, 30.8, 30.6, 30.5, 30.2, 30.2, 26.6, 25.9, 23.8, 23.7 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₃₃H₆₄NO₁₀ 634.4530; Found 634.4525.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-2-O-(2-Ethylhexanoyl)amino-3-O-β-D-glucopyranosyl-1-O-palmitoylglycerol (43)Chemical Formula: C₃₃H₆₃NO₁₀

Exact Mass: 633.4452 g/mol

Deacetylation of compound **38** (85 mg, 0.11 mmol) with hydrazine according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **43** (29 mg, 43%) as a paste.

R_f = 0.39 (CH₂Cl₂/MeOH: 10/1).

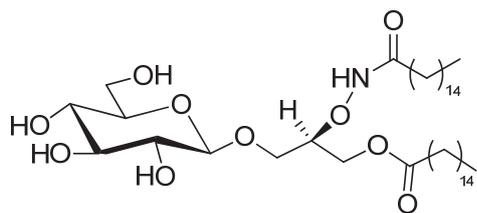
[α]_D = -29.7 (c 0.1, MeOH).

¹H NMR (400 MHz, CD₃OD): δ 4.37-4.21 (m, 3H, H-1',1), 4.20-4.12 (m, 1H, H-2), 4.05 (dd, J = 11.1, 3.9 Hz, 1H, H-3a), 3.85 (d, J = 11.7 Hz, 1H, H-6'a), 3.75 (dd, J = 11.3, 6.2 Hz, 1H, H-3b), 3.71-3.60 (m, 1H, H-6'b), 3.44-3.24 (m, 3H, H-3',4',5'), 3.22-3.15 (m, 1H, H-2'), 2.34 (t, J = 7.3 Hz, 2H, CH₂), 2.03-1.90 (m, 1H, CH), 1.70-1.50 (m, 4H, 2×CH₂), 1.49-1.17 (m, 30H, 15×CH₂), 1.06-0.81 (m, 9H, 3×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.7, 175.2 (C=O); 104.9 (C-1'), 83.9, 78.0, 77.9, 75.0, 71.5 (CH); 69.0, 63.2, 62.7 (CH₂); 46.6 (CH), 34.8, 33.3, 33.1, 30.8, 30.6, 30.5, 30.2, 26.9, 25.9, 23.7, 23.7 (CH₂); 14.5, 14.4, 12.4 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₃₃H₆₄NO₁₀ 634.4530; Found 634.4525.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-3-O-β-D-Glucopyranosyl-1-O-palmitoyl-2-O-palmitoylaminoglycerol (44)Chemical Formula: C₄₁H₇₉NO₁₀

Exact Mass: 745.5704 g/mol

Deacetylation of compound **39** (62 mg, 0.068 mmol) with hydrazine according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **44** (31 mg, 61%) as a white solid.

R_f = 0.24 (CH₂Cl₂/MeOH: 20/1).

[α]_D = -14.0 (c 0.1, MeOH).

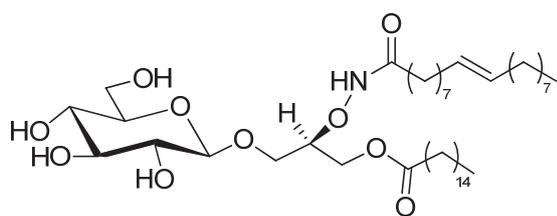
M.p. 177 °C.

¹H NMR (400 MHz, CD₃OD): δ 4.39-4.22 (m, 3H, H-1',1), 4.22-4.16 (m, 1H, H-2), 4.08 (dd, *J* = 11.6, 3.5 Hz, 1H, H-3a), 3.90 (d, *J* = 12.0 Hz, 1H, H-6'a), 3.78 (dd, *J* = 11.6, 6.3 Hz, 1H, H-3b), 3.69 (dd, *J* = 11.8, 5.2 Hz, 1H, H-6'b), 3.43-3.23 (m, 4H, H-2',3',4',5'), 2.37 (t, *J* = 7.5 Hz, 2H, CH₂), 2.10 (t, *J* = 7.4 Hz, 2H, CH₂), 1.69-1.58 (m, 4H, 2×CH₂), 1.49-1.10 (m, 48H, 24×CH₂), 0.96-0.86 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.1, 172.9 (C=O); 104.6 (C-1'), 83.4, 77.5, 74.6, 71.2 (CH); 68.9, 63.0, 62.5, 34.7, 33.6, 32.8, 30.5, 30.3, 30.2, 30.0, 26.3, 25.6, 23.4 (CH₂); 14.4 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₄₁H₈₀NO₁₀ 746.5782; Found: 746.5777.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2R)-3-O-β-D-Glucopyranosyl-2-O-oleoylamino-1-O-palmitoylglycerol (45)Chemical Formula: C₄₃H₈₁NO₁₀

Exact Mass: 771.5860 g/mol

Deacetylation of compound **40** (125 mg, 0.13 mmol) with hydrazine according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 25/1) led to compound **45** (35 mg, 34%) as a white solid.

R_f = 0.32 (CH₂Cl₂/MeOH: 10/1).

[α]_D = -18.0 (c 0.1, MeOH).

M.p. 162 °C.

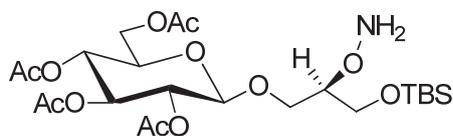
¹H NMR (400 MHz, CD₃OD): δ 5.32 (s, 2H, CH=CH), 4.37-4.20 (m, 3H, H-1',1), 4.19-4.11 (m, 1H, H-2), 4.04 (d, *J* = 10.0 Hz, 1H, H-3a), 3.85 (d, *J* = 11.3 Hz, 1H, H-6'a), 3.79-3.71 (m, 1H, H-3b), 3.68-3.58 (m, 1H, H-6'b), 3.40-3.22 (m, 3H, H-3',4',5'), 3.19 (d, *J* = 7.9 Hz, 1H, H-2'), 2.40-2.28 (m, 2H, CH₂), 2.14-1.94 (m, 4H, 2×CH₂), 1.67-1.52 (m, 4H, 2×CH₂), 1.48-1.10 (m, 46H, 23×CH₂), 0.97-0.80 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.2, 173.2 (C=O); 130.9, 130.8 (CH=CH); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.4, 62.7, 34.9, 33.8, 33.1, 30.8, 30.7, 30.5, 30.4, 30.2, 28.2, 26.6, 26.0, 23.8 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₄₃H₈₂NO₁₀ 772.5939; Found 772.5933.

Ref.: Chen, N.; Xie, J. *J. Org. Chem.* **2014**, *79*, 10716-10721.

(2*R*)-2-*O*-Amino-1-*O*-*tert*-butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (46)



Chemical Formula: C₂₃H₄₁NO₁₂Si

Exact Mass: 551.2398 g/mol

Hydrazinolysis of compound **32** (123 mg, 0.18 mmol) by hydrazine under mild conditions according to the general **procedure E** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/1) afforded compound **46** (91 mg, 92%) as a colourless paste.

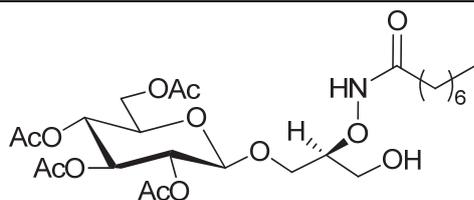
R_f = 0.39 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -52.3$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.20 (t, J = 9.4 Hz, 1H, H-3'), 5.07 (t, J = 9.9 Hz, 1H, H-4'), 5.00 (dd, J = 9.4, 8.0 Hz, 1H, H-2'), 4.58 (d, J = 7.8 Hz, 1H, H-1'), 4.29-4.08 (m, 2H, H-6'), 3.98 (dd, J = 10.6, 3.3 Hz, 1H, H-3a), 3.78-3.62 (m, 5H, H-1,2,3b,5'), 2.07, 2.05, 2.02, 2.00 (4×s, 12H, 4×OAc), 0.88 (s, 9H, *t*-Bu), 0.05 (s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.4, 169.5 (C=O); 101.2 (C-1'), 83.8 (CH), 72.9 (C-3'), 71.9 (C-5'), 71.4 (C-2'), 68.5 (C-4'), 68.3, 62.0, 61.5 (CH₂); 25.9, 20.8, 20.7 (CH₃); 18.4 (C_q, *t*-Bu), -5.3 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+K]⁺ Calcd for C₂₃H₄₁KNO₁₂Si 590.2035; Found 590.2037.

(2S)-2-O-Octanoylamino-1-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (47)Chemical Formula: C₂₅H₄₁NO₁₃

Exact Mass: 563.2578 g/mol

Coupling of oxyamine compound **46** (102 mg, 0.186 mmol) with octanoic acid according to the general **procedure F** led to the *N*-oxyamide compound which was used directly for the next step without purification. Desilylation of this crude compound using TBAF according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/2) afforded compound **47** (58 mg, 56%, two steps) as a white solid.

$R_f = 0.12$ (petroleum ether/EtOAc = 1/1).

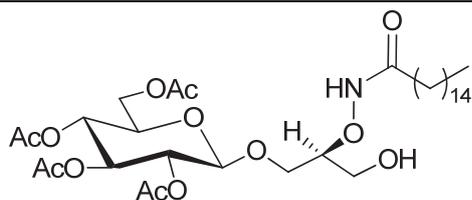
$[\alpha]_D = -103.7$ (c 0.1, CHCl₃).

M.p. 108 °C.

¹H NMR (400 MHz, CDCl₃): δ 9.27 (s, 1H, NH), 5.23 (t, $J = 9.6$ Hz, 1H, H-3'), 5.05 (t, $J = 9.9$ Hz, 1H, H-4'), 4.99 (dd, $J = 9.7, 7.9$ Hz, 1H, H-2'), 4.48 (d, $J = 8.2$ Hz, 1H, H-1'), 4.24 (dd, $J = 12.4, 4.6$ Hz, 1H, H-6'a), 4.14 (dd, $J = 12.4, 2.3$ Hz, 1H, H-6'b), 4.04-3.90 (m, 1H, H-1a,2), 3.77-3.67 (m, 1H, H-5'), 3.63-3.47 (m, 3H, H-1b,3), 2.24-1.96 (m, 14H, 4×OAc, CH₂), 1.69-1.57 (m, 2H, CH₂), 1.41-1.19 (m, 8H, 4×CH₂), 0.85 (t, $J = 6.5$ Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.6, 171.1, 170.8, 170.2, 169.6 (C=O); 101.1 (C-1'), 88.8 (CH), 72.1 (C-3'), 72.0 (C-5'), 71.8 (C-2'), 69.4 (CH₂), 68.3 (C-4'), 61.8, 60.4, 33.0, 31.7, 29.1, 29.0, 25.3, 22.7 (CH₂); 21.0, 20.8, 20.7, 14.1 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₂₅H₄₁NNaO₁₃ 586.2476; Found 586.2474.

(2*S*)-2-*O*-Palmitoylamino-1-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol(48)Chemical Formula: C₃₃H₅₇NO₁₃

Exact Mass: 675.3830 g/mol

Coupling of oxyamine compound **46** (135 mg, 0.245 mmol) with palmitic acid according to the general **procedure F** led to the *N*-oxyamide compound which was used directly for the next step without purification. Desilylation of this crude compound using TBAF according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **48** (78 mg, 47%, two steps) as a white solid.

R_f = 0.14 (petroleum ether/EtOAc = 1/1).

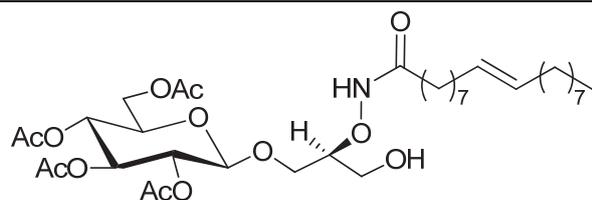
$[\alpha]_D = -66.7$ (c 0.1, CHCl₃).

M.p. 117 °C.

¹H NMR (400 MHz, CDCl₃): δ 9.19 (s, 1H, NH), 5.24 (t, J = 9.6 Hz, 1H, H-3'), 5.06 (t, J = 9.8 Hz, 1H, H-4'), 5.00 (dd, J = 9.6, 7.8 Hz, 1H, H-2'), 4.48 (d, J = 7.8 Hz, 1H, H-1'), 4.25 (dd, J = 12.8, 4.6 Hz, 1H, H-6'a), 4.14 (dd, J = 12.4, 1.8 Hz, 1H, H-6'b), 4.01-3.91 (m, 2H, H-1a, 2), 3.74-3.67 (m, 1H, H-5'), 3.61-3.48 (m, 3H, H-1b,3), 2.16-1.96 (m, 14H, 4×OAc, CH₂), 1.68-1.56 (m, 2H, CH₂), 1.34-1.17 (m, 24H, 12×CH₂), 0.85 (t, J = 6.9 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.6, 171.1, 170.8, 170.2, 169.6 (C=O); 101.2 (C-1'), 89.0 (CH), 72.2 (C-3'), 72.0 (C-5'), 71.8 (C-2'), 69.4 (CH₂), 68.3 (C-4'), 61.8, 60.4, 33.0, 32.0, 29.7, 29.6, 29.4, 29.3, 25.4, 22.8 (CH₂); 21.1, 20.9, 20.7, 14.2 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₃H₅₇NNaO₁₃ 698.3728; Found 698.3734.

(2S)-2-O-Oleoylamino-1-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (49)Chemical Formula: C₃₅H₅₉NO₁₃

Exact Mass: 701.3986 g/mol

Coupling of oxyamine compound **46** (110 mg, 0.20 mmol) with oleic acid according to the general **procedure F** led to the *N*-oxyamide compound which was used directly for the next step without purification. Desilylation of this crude compound using TBAF according to the general **procedure A** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **49** (69 mg, 49%, two steps) as a white solid.

$R_f = 0.16$ (petroleum ether/EtOAc = 1/1).

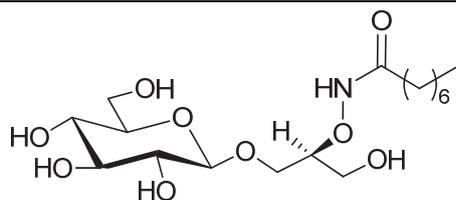
$[\alpha]_D = -72.7$ (c 0.1, CHCl₃).

M.p. 85 °C.

¹H NMR (400 MHz, CDCl₃): δ 9.21 (s, 1H, NH), 5.42-5.28 (m, 2H, CH=CH), 5.24 (t, $J = 9.6$ Hz, 1H, H-3'), 5.10-4.96 (m, 2H, H-2',4'), 4.48 (d, $J = 7.8$ Hz, 1H, H-1'), 4.30-4.11 (m, 2H, H-6'), 4.03-3.91 (m, 2H, H-1a,2), 3.76-3.67 (m, 1H, H-5'), 3.63-3.48 (m, 3H, H-1b,3), 2.24-1.93 (m, 16H, 4×OAc, 2×CH₂), 1.69-1.57 (m, 2H, CH₂), 1.46-1.12 (m, 22H, 11×CH₂), 0.85 (t, $J = 6.6$ Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 172.6, 171.1, 170.8, 170.1, 169.5 (C=O); 130.1, 129.8 (CH=CH); 101.1 (C-1'), 88.9 (CH), 72.1 (C-3'), 72.0 (C-5'), 71.8 (C-2'), 69.4 (CH₂), 68.3 (C-4'), 61.8, 60.4, 33.0, 32.0, 29.8, 29.6, 29.4, 29.2, 29.0, 27.3, 25.3, 22.7 (CH₂); 21.0, 20.8, 20.7, 14.2 (CH₃).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₃₅H₅₉NNaO₁₃ 724.3884; Found 724.3878.

(2S)-1-O-β-D-Glucopyranosyl-2-O-octanoylaminoglycerol (50)Chemical Formula: C₁₇H₃₃NO₉

Exact Mass: 395.2155 g/mol

Deacetylation of compound **47** (28 mg, 0.05 mmol) with Na/MeOH according to the general **procedure I** led to compound **50** (17 mg, 89%) as a yellowish paste.

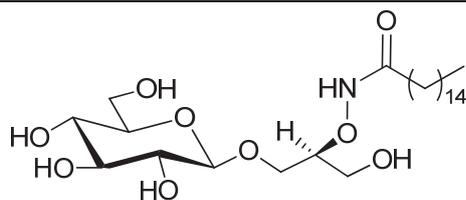
$R_f = 0.44$ (CH₂Cl₂/MeOH: 5/1).

$[\alpha]_D = -6.33$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CD₃OD): δ 4.32 (d, $J = 7.8$ Hz, 1H, H-1'), 4.03 (dd, $J = 11.0, 3.8$ Hz, 1H, H-1a), 4.00-3.93 (m, 1H, H-2), 3.87 (d, $J = 11.6$ Hz, 1H, H-6'a), 3.79-3.60 (m, 4H, H-1b,3,6'b), 3.41-3.17 (m, 4H, H-2',3',4',5'), 2.11 (t, $J = 7.3$ Hz, 2H, CH₂), 1.68-1.54 (m, 2H, CH₂), 1.41-1.20 (m, 8H, 4×CH₂), 0.90 (t, $J = 6.4$ Hz, 3H, CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 174.0 (C=O), 104.7 (C-1'), 87.1, 78.0, 75.0, 71.6 (CH); 69.0, 62.7, 61.0, 33.7, 32.9, 30.8, 30.2, 30.1, 26.6, 23.7 (CH₂); 14.4 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₁₇H₃₃NNaO₉ 418.2053; Found 418.2054.

(2S)-3-O-β-D-Glucopyranosyl-2-O-palmitoylaminoglycerol (51)Chemical Formula: C₂₅H₄₉NO₉

Exact Mass: 507.3407 g/mol

Deacetylation of compound **48** (48 mg, 0.071 mmol) with Na/MeOH according to the general **procedure I** led to compound **51** (36 mg, 100%) as a yellowish paste.

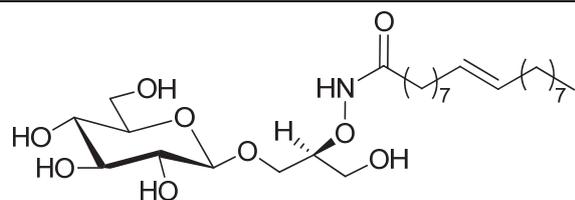
R_f = 0.62 (CH₂Cl₂/MeOH: 5/1).

[α]_D = +45.0 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CD₃OD): δ 4.33 (d, *J* = 7.8 Hz, 1H, H-1'), 4.04 (dd, *J* = 11.0, 4.1 Hz, 1H, H-1a), 4.01-3.94 (m, 1H, H-2), 3.91-3.84 (m, 1H, H-6'a), 3.78-3.60 (m, 4H, H-1b,3,6'b), 3.40-3.24 (m, 3H, H-3',4',5'), 3.21 (dd, *J* = 9.1, 7.9 Hz, 1H, H-2'), 2.12 (t, *J* = 7.3 Hz, 2H, CH₂), 1.66-1.55 (m, 2H, CH₂), 1.38-1.21 (m, 24H, 12×CH₂), 0.90 (t, *J* = 6.9 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 173.9 (C=O), 104.7 (C-1'), 87.4, 78.0, 75.0, 71.6 (CH); 69.0, 62.7, 60.9, 33.7, 33.1, 30.8, 30.6, 30.5, 30.4, 30.2, 26.6, 23.7 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₅H₄₉NNaO₉ 530.3305; Found 530.3306.

(2S)-3-O-β-D-Glucopyranosyl-2-O-oleoylamino-glycerol (52)Chemical Formula: C₂₇H₅₁NO₉

Exact Mass: 533.3564 g/mol

Deacetylation of compound **49** (49 mg, 0.07 mmol) with Na/MeOH according to the general **procedure I** led to compound **52** (37 mg, 99%) as a yellowish paste.

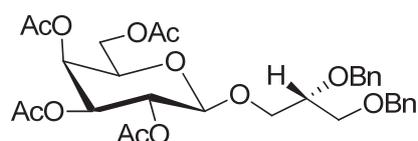
R_f = 0.51 (CH₂Cl₂/MeOH: 5/1).

[α]_D = -22.3 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CD₃OD): δ 5.40-5.29 (m, 2H, CH=CH), 4.33 (d, *J* = 7.8 Hz, 1H, H-1'), 4.04 (dd, *J* = 11.1, 4.1 Hz, 1H, H-1a), 4.01-3.94 (m, 1H, H-2), 3.91-3.83 (m, 1H, H-6'a), 3.78-3.60 (m, 4H, H-1b,3,6'b), 3.39-3.24 (m, 3H, H-3',4',5'), 3.21 (dd, *J* = 9.1, 7.8 Hz, 1H, H-2'), 2.12 (t, *J* = 7.4 Hz, 2H, CH₂), 2.07-1.97 (m, 4H, 2×CH₂), 1.66-1.55 (m, 2H, CH₂), 1.41-1.19 (m, 20H, 10×CH₂), 0.90 (t, *J* = 6.6 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 173.9 (C=O), 130.9, 130.8 (CH=CH); 104.7 (C-1'), 87.3, 78.0, 75.0, 71.6 (CH); 69.0, 62.7, 60.9, 33.7, 33.1, 30.8, 30.6, 30.5, 30.3, 30.2, 28.1, 26.6, 23.7 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₇H₅₁NNaO₉ 556.3462; Found 556.3463.

(2R)-1,2-Di-O-benzyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-galactopyranosyl)glycerol (53)Chemical Formula: C₃₁H₃₈O₁₂

Exact Mass: 602.2363 g/mol

To a solution of β-D-galactose pentaacetate (2 g, 5.12 mmol) in CH₂Cl₂ (15 mL) was added HBr (33% in AcOH, 3.52 mL, 20.48 mmol) at 0 °C. The mixture was stirred at room temperature under argon for 6 h, and then the solution neutralized with aq NaHCO₃ in an ice bath. The resulting 2,3,4,6-tetra-O-acetyl-α-D-galactopyranosyl bromide was extracted with CH₂Cl₂ (100 mL), dried over MgSO₄, filtered and evaporated to give a syrup. To a solution of (2S)-1,2-di-O-benzyl-glycerol **28** (1.39 g, 5.12 mmol), HgBr₂ (0.92 g, 2.56 mmol) and Hg(CN)₂ (0.65 g, 2.56 mmol) in dry acetonitrile (10 mL) was added dropwise over a period of 1 h, a solution of glycosyl bromide in acetonitrile (5 mL). The reaction solution was stirred at room temperature for 15 h, and then concentrated to an oil which was dissolved in CH₂Cl₂ (100 mL) and washed with saturated aq NaHCO₃ (2×30 mL) and water (30 mL). The dried (MgSO₄) organic layer was concentrated. Purification by column chromatography (petroleum ether/EtOAc: 3/1) afforded compound **53** (2.30 g, 75%) as a colorless paste.

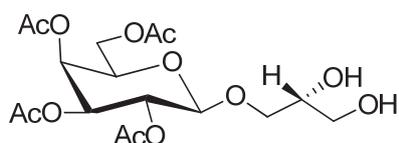
R_f = 0.20 (petroleum ether/EtOAc: 3/1).

$[\alpha]_D = -15.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.38-7.27 (m, 10H, H-Ph), 5.40-5.34 (m, H-4'), 5.20 (dd, J = 10.5, 7.9 Hz, 1H, H-2'), 4.99 (t, J = 10.5, 3.4 Hz, 1H, H-3'), 4.67-4.62 (m, 2H, OCH₂), 4.56-4.46 (m, 3H, H-1', OCH₂), 4.19-4.08 (m, 2H, H-6'), 3.98 (dd, J = 9.8, 4.2 Hz, 1H, H-3a), 3.86 (td, J = 6.5, 0.8 Hz, 1H, H-5'), 3.78-3.68 (m, 2H, H-2,3b), 3.63-3.55 (m, 2H, H-1), 2.15, 2.04, 1.98, 1.95 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.5, 170.4, 170.3, 169.5 (C=O); 138.6, 138.3 (C_q); 128.5, 127.8 (CH-Ph); 101.2 (C-1'), 77.0 (CH), 73.6, 72.3 (CH₂); 71.0 (C-3'), 70.8 (C-5'), 69.9, 69.3 (CH₂); 69.0 (C-2'), 67.2 (C-4'), 61.4 (C-6'), 20.8, 20.7 (OAc).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₁H₃₈NaO₁₂ 625.2261; Found 625.2261.

(2R)-1-O-(2',3',4',6'-Tetra-O-acetyl-β-D-galactopyranosyl)glycerol (54)Chemical Formula: C₁₇H₂₆O₁₂

Exact Mass: 422.1424 g/mol

A mixture of compound **53** (7 g, 11.63 mmol) and 10% Pd/C (1 g) in EtOAc (100 mL) containing 10 mL each of EtOH and AcOH was vigorously shaken under H₂ at room temperature overnight. The catalyst was filtered off, the filtrate evaporated. Purification by column chromatography (CH₂Cl₂/MeOH: 100/3) afforded compound **54** (3.45 g, 70%) as a colourless paste.

R_f = 0.22 (CH₂Cl₂/MeOH: 100/3).

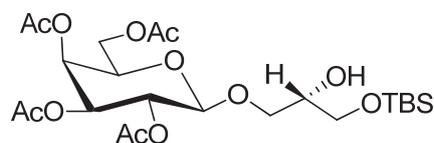
[α]_D = +3.0 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.40 (dd, *J* = 3.7, 0.9 Hz, 1H, H-4'), 5.20 (dd, *J* = 10.5, 7.8 Hz, 1H, H-2'), 5.03 (dd, *J* = 10.5, 3.2 Hz, 1H, H-3'), 4.51 (d, *J* = 8.2 Hz, 1H, H-1'), 4.21-4.10 (m, 2H, H-6'), 3.99-3.92 (m, 1H, H-5'), 3.90-3.75 (m, 3H, H-2,3), 3.70 (dd, *J* = 11.4, 3.7 Hz, 1H, H-1a), 3.61 (dd, *J* = 11.4, 4.6 Hz, 1H, H-1b), 2.99 (s, 2H, 2×OH), 2.17, 2.08, 2.07, 2.00 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.6, 170.4, 170.3, 170.0 (C=O); 102.0 (C-1'), 72.8 (CH₂), 71.0 (C-5'), 70.7 (C-3'), 70.5 (CH), 68.9 (C-2'), 67.1 (C-4'), 63.5, 61.5 (CH₂), 20.9, 20.8, 20.7 (OAc).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₁₇H₂₆NaO₁₂ 445.1322; Found 445.1321.

(2*S*)-1-*O*-*tert*-Butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (55)



Chemical Formula: C₂₃H₄₀O₁₂Si

Exact Mass: 536.2289 g/mol

To a solution of compound **54** (2.8 g, 6.64 mmol), *N*-methylimidazole (1.59 mL, 19.92 mmol) and iodine (5.05 g, 19.92 mmol) in anhydrous THF (30 mL) under argon at 0 °C, was added TBSCl (1.1 g, 7.30 mmol). The reaction mixture was stirred for 20 min at 0 °C, then quenched with saturated aq Na₂S₂O₃ and the solution was concentrated. The residue was diluted with EtOAc (200 mL), washed with saturated aq Na₂S₂O₃ (2×50 mL), H₂O (50 mL) and brine (50 mL), dried over MgSO₄, filtered, concentrated, and purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/1) to afford compound **55** (3.20 g, 90%) as a colourless paste.

R_f = 0.73 (CH₂Cl₂/MeOH: 100/3).

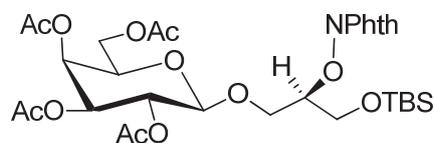
[α]_D = -7.3 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.34 (d, *J* = 3.5 Hz, 1H, H-4'), 5.16 (dd, *J* = 10.5, 8.0 Hz, 1H, H-2'), 4.97 (dd, *J* = 10.5, 3.3 Hz, 1H, H-3'), 4.47 (d, *J* = 7.8 Hz, 1H, H-1'), 4.13-4.05 (m, 2H, H-6'), 3.93-3.85 (m, 1H, H-5'), 3.80-3.65 (m, 3H, H-2,3), 3.61-3.53 (m, 2H, H-1), 2.54 (s, 1H, OH), 2.11, 2.01, 2.01, 1.93 (4×s, 12H, 4×OAc), 0.85 (s, 9H, *t*-Bu), 0.02 (s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.4, 170.2, 170.1, 169.5 (C=O); 101.8 (C-1'), 71.6 (CH₂), 70.9, 70.8, 70.5, 68.9, 67.1 (CH); 63.7, 61.4 (CH₂); 25.9, 20.8, 20.7, 20.6 (CH₃); 18.3 (C_q, *t*-Bu), -5.4 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₃H₄₀NaO₁₂Si 559.2187; Found 559.2188.

(2R)-2-O-Phthalimido-1-O-tert-butyltrimethylsilyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-galactopyranosyl)glycerol (56)



Chemical Formula: C₃₁H₄₃NO₁₄Si

Exact Mass: 681.2453 g/mol

The phthalimido group was introduced to compound **55** (1.68 g, 3.13 mmol) via Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/0.5) afforded compound **56** (2.06 g, 97%) as a yellowish paste.

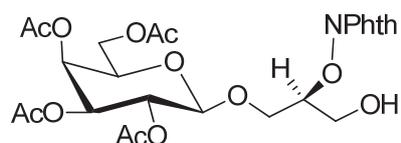
R_f = 0.60 (CH₂Cl₂/MeOH: 100/1).

[α]_D = +4.0 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.88-7.71 (m, 4H, Phth), 5.36 (dd, *J* = 3.4, 1.0 Hz, 1H, H-4'), 5.16 (dd, *J* = 10.6, 7.8 Hz, 1H, H-2'), 5.00 (dd, *J* = 10.6, 3.6 Hz, 1H, H-3'), 4.65 (d, *J* = 8.2 Hz, 1H, H-1'), 4.46-4.39 (m, 1H, H-2), 4.18-4.08 (m, 3H, H-3a,6'), 3.98-3.88 (m, 4H, H-1,3b,5'), 2.12, 2.05, 2.04, 1.96 (4×s, 12H, 4×OAc), 0.79 (s, 9H, *t*-Bu), 0.00, -0.01 (2×s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.5, 170.4, 170.3, 169.7 (C=O), 163.8 (C=O, Phth); 134.5 (CH, Phth), 129.1 (C_q), 123.6 (CH, Phth), 101.3 (C-1'), 87.0 (CH), 71.1 (C-3'), 70.7 (C-5'), 68.8 (C-2'), 67.9 (CH₂), 67.1 (C-4'), 62.0, 61.4 (CH₂), 25.8, 20.9, 20.8 (CH₃); 18.3 (C_q, *t*-Bu), -5.6 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₁H₄₃NNaO₁₄Si 704.2351; Found 704.2346.

(2S)-2-O-Phthalimido-1-O-(2',3',4',6'-tetra-O-acetyl-β-D-galactopyranosyl)glycerol (57)Chemical Formula: C₂₅H₂₉NO₁₄

Exact Mass: 567.1588 g/mol

Desilylation of compound **56** (1.45 g, 2.13 mmol) using AcCl according to the general **procedure B** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/3) gave compound **57** (0.93 g, 77%) as a white solid.

$R_f = 0.39$ (petroleum ether/EtOAc: 1/2).

$[\alpha]_D = -17.7$ (c 0.1, CHCl₃).

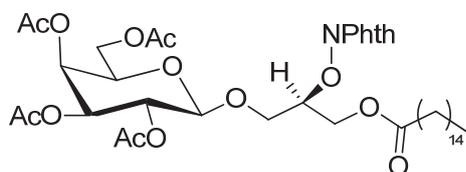
M.p. 66 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.97-7.78 (m, 4H, Phth), 5.40 (d, $J = 3.2$ Hz, 1H, H-4'), 5.23 (dd, $J = 10.6, 8.2$ Hz, 1H, H-2'), 5.03 (dd, $J = 10.6, 3.2$ Hz, 1H, H-3'), 4.65 (d, $J = 7.8$ Hz, 1H, H-1'), 4.44-4.30 (m, 1H, H-2), 4.28-4.11 (m, 3H, H-3a,6'), 4.05 (dd, $J = 11.6, 6.3$ Hz, 1H, H-3b), 4.01-3.92 (m, 1H, H-5'), 3.84 (dd, $J = 13.8, 2.8$ Hz, 1H, H-1a), 3.73 (dd, $J = 13.8, 4.1$ Hz, 1H, H-1b), 2.16, 2.10, 2.07, 1.99 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.6, 170.4, 170.3, 169.8 (C=O), 164.7 (C=O, Phth); 135.1 (CH, Phth), 128.8 (C_q), 124.1 (CH, Phth), 101.4 (C-1'), 88.2 (CH), 71.0, 70.9, 68.7 (CH); 67.7 (CH₂), 67.2 (CH), 61.4, 60.5 (CH₂); 20.9, 20.8, 20.7 (OAc).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₂₅H₂₉NNaO₁₄ 590.1486; Found 590.1483.

(2*R*)-1-*O*-Palmitoyl-2-*O*-phthalimido-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (58**)**



Chemical Formula: C₄₁H₅₉NO₁₅

Exact Mass: 805.3885 g/mol

Esterification of compound **57** (0.93 g, 1.64 mmol) with palmitic acid according to the general **procedure G** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 3/2) afforded compound **58** (1.23 g, 93%) as a colourless paste.

R_f = 0.60 (petroleum ether/EtOAc: 1/1).

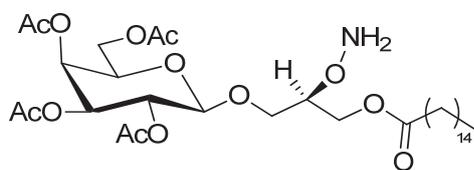
[α]_D = -18.7 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.93-7.75 (m, 4H, Phth), 5.38 (d, *J* = 3.6 Hz, 1H, H-4'), 5.19 (dd, *J* = 10.5, 8.2 Hz, 1H, H-2'), 5.02 (dd, *J* = 10.5, 3.2 Hz, 1H, H-3'), 4.64 (d, *J* = 8.2 Hz, 1H, H-1'), 4.62-4.55 (m, 1H, H-2), 4.42 (dd, *J* = 12.8, 3.7 Hz, 1H, H-1a), 4.34 (dd, *J* = 12.8, 5.9 Hz, 1H, H-1b), 4.23-4.09 (m, 3H, H-3a,6'), 4.01 (dd, *J* = 11.7, 5.8 Hz, 1H, H-3b), 3.98-3.91 (m, 1H, H-5'), 2.31 (t, *J* = 7.6 Hz, 1H, CH₂), 2.15, 2.08, 2.06, 1.98 (4×s, 12H, 4×OAc), 1.67-1.54 (m, 2H, CH₂), 1.45-1.21 (m, 24H, 12×CH₂), 0.88 (t, *J* = 6.9 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 173.5, 170.6, 170.4, 170.3, 169.7 (C=O), 163.6 (C=O, Phth); 134.7 (CH, Phth), 128.9 (C_q), 123.8 (CH, Phth), 101.3 (C-1'), 84.5 (CH), 71.0 (C-3'), 70.8 (C-5'), 68.6 (C-2'), 67.6 (CH₂), 67.1 (C-4'), 62.4, 61.3, 34.1, 32.0, 29.8, 29.8, 29.7, 29.6, 29.5, 29.4, 29.2, 24.8, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.2 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₄₁H₅₉NNaO₁₅ 828.3782; Found 828.3788.

(2R)-2-O-Amino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-galactopyranosyl)glycerol (59)

Chemical Formula: C₃₃H₅₇NO₁₃

Exact Mass: 675.3830 g/mol

Hydrazinolysis of compound **58** (50 mg, 0.062 mmol) by hydrazine under mild conditions according to the general **procedure E** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/1) afforded compound **59** (39 mg, 93%) as a white paste.

R_f = 0.44 (petroleum ether/EtOAc: 1/1).

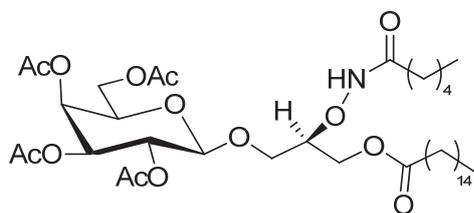
[α]_D = -53.0 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): 5.39 (d, *J* = 3.2 Hz, 1H, H-4'), 5.21 (dd, *J* = 10.5, 7.8 Hz, 1H, H-2'), 5.00 (dd, *J* = 10.5, 3.2 Hz, 1H, H-3'), 4.53 (d, *J* = 8.2 Hz, 1H, H-1'), 4.41-4.33 (m, 1H, H-2), 4.28-4.09 (m, 4H, H-1,6'), 3.97 (dd, *J* = 11.4, 5.0 Hz, 1H, H-3a), 3.93-3.87 (m, 1H, H-5'), 3.77 (dd, *J* = 11.2, 6.2 Hz, 1H, H-3b), 2.31 (t, *J* = 7.6 Hz, 1H, CH₂), 2.16, 2.06, 2.04, 1.99 (4×s, 12H, 4×OAc), 1.71-1.57 (m, 2H, CH₂), 1.35-1.22 (m, 24H, 12×CH₂), 0.88 (t, *J* = 6.9 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 173.7, 170.6, 170.4, 170.4, 169.6 (C=O); 101.8 (C-1'), 79.1 (CH), 71.0 (C-3'), 70.7 (C-5'), 68.9 (C-2'), 68.4 (CH₂), 67.1 (C-4'), 62.8, 61.3, 34.3, 32.1, 29.8, 29.6, 29.5, 29.4, 29.3, 25.1, 22.8 (CH₂); 20.8, 20.8, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₃H₅₇NNaO₁₃ 698.3782; Found 698.3715.

(2*R*)-2-*O*-Hexanoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (60**)**



Chemical Formula: C₃₉H₆₇NO₁₄

Exact Mass: 773.4562 g/mol

Compound **60** was prepared from compound **59** (115 mg, 0.17 mmol) and hexanoic acid (21 μ L, 0.17 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **60** (115 mg, 88%) as a colourless paste.

R_f = 0.55 (petroleum ether/EtOAc: 1/1).

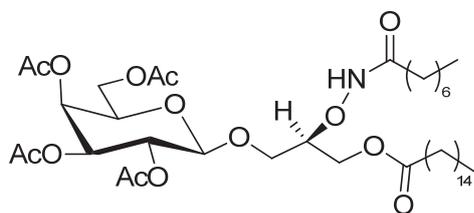
$[\alpha]_D = -62.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 8.86 (s, 1H, NH), 5.37 (t, J = 3.2 Hz, 1H, H-4'), 5.16 (dd, J = 10.3, 8.6 Hz, 1H, H-2'), 5.01 (dd, J = 10.5, 3.2 Hz, 1H, H-3'), 4.50 (d, J = 7.8 Hz, 1H, H-1'), 4.36 (dd, J = 12.2, 3.5 Hz, 1H, H-1a), 4.24-4.04 (m, 4H, H-2,1b,6'), 4.02-3.95 (m, 1H, H-3a), 3.93-3.87 (m, 1H, H-5'), 3.67 (dd, J = 10.1, 7.2 Hz, 1H, H-3b), 2.31 (t, J = 7.6 Hz, 2H, CH₂), 2.15-1.93 (m, 14H, 4 \times OAc, CH₂), 1.65-1.53 (m, 4H, 2 \times CH₂), 1.35-1.11 (m, 28H, 14 \times CH₂), 0.91-0.79 (m, 6H, 2 \times CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.2, 170.8, 170.4, 170.2, 170.1 (C=O); 101.2 (C-1'), 82.3 (CH), 70.8 (C-5'), 70.6 (C-3'), 69.1 (C-2'), 68.2 (CH₂), 66.9 (C-4'), 61.4, 61.2, 34.1, 33.1, 31.9, 31.4, 29.7, 29.6, 29.5, 29.4, 29.3, 29.1, 25.1, 24.9, 23.9, 22.7, 22.4 (CH₂); 21.0, 20.9, 20.7, 20.6, 14.2, 14.0 (CH₃).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₃₉H₆₇NNaO₁₄ 796.4459; Found 796.4456.

(2*R*)-2-*O*-Octanoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (61)



Chemical Formula: C₄₁H₇₁NO₁₄

Exact Mass: 801.4875 g/mol

Compound **61** was prepared from compound **59** (135 mg, 0.20 mmol) and octanoic acid (32 μ L, 0.20 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **61** (127 mg, 81%) as a yellowish paste.

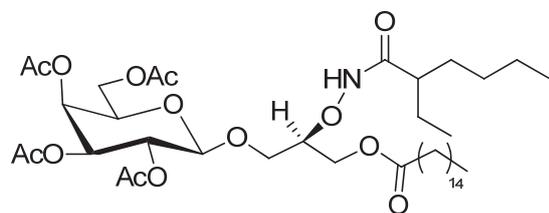
R_f = 0.56 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -52.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 8.89 (s, 1H, NH), 5.41 (t, J = 3.2 Hz, 1H, H-4'), 5.21 (dd, J = 10.1, 8.3 Hz, 1H, H-2'), 5.05 (dd, J = 10.5, 3.2 Hz, 1H, H-3'), 4.54 (d, J = 7.8 Hz, 1H, H-1'), 4.42 (dd, J = 12.4, 3.2 Hz, 1H, H-1a), 4.26-4.09 (m, 4H, H-2,1b,6'), 4.06-3.98 (m, 1H, H-3a), 3.97-3.90 (m, 1H, H-5'), 3.71 (dd, J = 9.8, 7.4 Hz, 1H, H-3b), 2.36 (t, J = 7.3 Hz, 2H, CH₂), 2.19-1.97 (m, 14H, 4 \times OAc, CH₂), 1.69-1.57 (m, 4H, 2 \times CH₂), 1.38-1.20 (m, 32H, 16 \times CH₂), 0.91-0.84 (m, 6H, 2 \times CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.3, 170.8, 170.5, 170.3, 170.1 (C=O); 101.2 (C-1'), 82.4 (CH), 70.9 (C-5'), 70.6 (C-3'), 69.1 (C-2'), 68.2 (CH₂), 67.0 (C-4'), 61.4, 61.3, 34.2, 33.2, 32.0, 31.8, 29.8, 29.7, 29.6, 29.5, 29.4, 29.3, 29.2, 29.0, 25.5, 24.9, 22.8, 22.7 (CH₂); 21.0, 20.8, 20.7, 14.2 (CH₃).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₄₁H₇₁NNaO₁₄ 824.4772; Found 824.4769.

(2R)-2-O-(2-Ethylhexanoyl)amino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-galactopyranosyl)glycerol (62)Chemical Formula: C₄₁H₇₁NO₁₄

Exact Mass: 801.4875 g/mol

Compound **62** was prepared from compound **59** (184 mg, 0.23 mmol) and 2-ethylhexanoic acid (36 μL, 0.23 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **62** (143 mg, 78%) as a colourless paste.

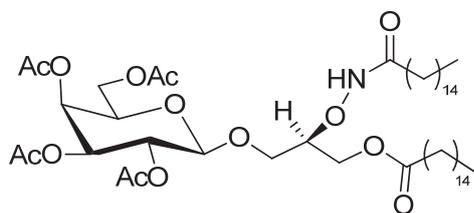
R_f = 0.45 (petroleum ether/EtOAc: 1/1).

[α]_D = -54.3 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 8.89 (s, 1H, NH), 5.39 (t, *J* = 2.8 Hz, 1H, H-4'), 5.18 (dd, *J* = 10.2, 8.0 Hz, 1H, H-2'), 5.03 (dd, *J* = 10.5, 3.2 Hz, 1H, H-3'), 4.51 (d, *J* = 7.8 Hz, 1H, H-1'), 4.46-4.37 (m, 1H, H-1a), 4.26-4.08 (m, 4H, H-2,1b,6'), 3.99 (dd, *J* = 10.0, 4.4 Hz, 1H, H-3a), 3.95-3.89 (m, 1H, H-5'), 3.70 (dd, *J* = 10.0, 7.3 Hz, 1H, H-3b), 2.34 (t, *J* = 7.5 Hz, 2H, CH₂), 2.13, 2.08, 2.03, 1.97 (4×s, 12H, 4×OAc), 1.94-1.84 (m, 1H, CH), 1.69-1.55 (m, 4H, 2×CH₂), 1.52-1.36 (m, 2H, CH₂), 1.36-1.18 (m, 28H, 14×CH₂), 0.95-0.82 (m, 9H, 3×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.2, 173.4, 170.7, 170.4, 170.2, 170.0 (C=O); 101.1 (C-1'), 82.4 (CH), 70.8 (C-5'), 70.5 (C-3'), 69.1 (C-2'), 68.0 (CH₂), 66.9 (C-4'), 61.4, 61.2 (CH₂); 45.8 (CH), 34.1, 32.1, 31.9, 29.7, 29.5, 29.4, 29.1, 25.8, 24.8, 22.7 (CH₂); 20.9, 20.6, 14.1, 14.0, 12.0 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₄₁H₇₁NNaO₁₄ 824.4772; Found 824.4767.

(2*R*)-1-*O*-Palmitoyl-2-*O*-palmitoylamino-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (63)Chemical Formula: C₄₉H₈₇NO₁₄

Exact Mass: 913.6127 g/mol

Compound **63** was prepared from compound **59** (170 mg, 0.25 mmol) and palmitic acid (64 mg, 0.25 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **63** (204 mg, 90%) as a white solid.

R_f = 0.29 (petroleum ether/EtOAc: 2/1).

$[\alpha]_D = -38.0$ (c 0.1, CHCl₃).

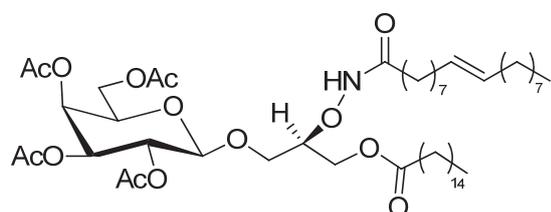
M.p. 45 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.86 (s, 1H, NH), 5.41 (t, J = 2.8 Hz, 1H, H-4'), 5.21 (dd, J = 10.4, 8.3 Hz, 1H, H-2'), 5.05 (dd, J = 10.5, 3.2 Hz, 1H, H-3'), 4.53 (d, J = 7.8 Hz, 1H, H-1'), 4.42 (dd, J = 12.4, 3.2 Hz, 1H, H-1a), 4.26-4.08 (m, 4H, H-2,1b,6'), 4.05-3.98 (m, 1H, H-3a), 3.96-3.89 (m, 1H, H-5'), 3.71 (dd, J = 10.1, 7.3 Hz, 1H, H-3b), 2.36 (t, J = 7.6 Hz, 2H, CH₂), 2.18-1.98 (m, 14H, 4×OAc, CH₂), 1.68-1.57 (m, 4H, 2×CH₂), 1.36-1.17 (m, 48H, 24×CH₂), 0.91-0.83 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.3, 170.8, 170.5, 170.3, 170.2 (C=O); 101.3 (C-1'), 82.4 (CH), 70.9 (C-5'), 70.6 (C-3'), 69.1 (C-2'), 68.2 (CH₂), 67.0 (C-4'), 61.4, 61.3, 34.2, 33.3, 32.0, 29.8, 29.6, 29.5, 29.4, 29.2, 25.5, 25.0, 22.8 (CH₂); 21.1, 20.8, 20.7, 14.3 (CH₃).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₄₉H₈₇NNaO₁₄ 936.6024; Found 936.6024.

(2*R*)-2-*O*-Oleoylamino-1-*O*-palmitoyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-galactopyranosyl)glycerol (64)



Chemical Formula: C₅₁H₈₉NO₁₄

Exact Mass: 939.6283 g/mol

Compound **64** was prepared from compound **59** (115 mg, 0.17 mmol) and oleic acid (48 mg, 0.217 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **64** (147 mg, 92%) as colourless paste.

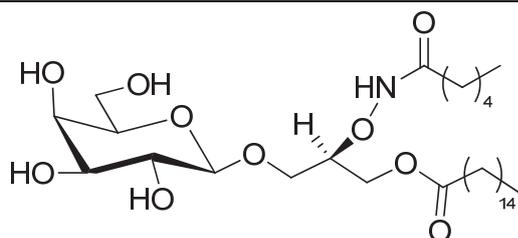
R_f = 0.56 (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -42.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 8.84 (s, 1H, NH), 5.41 (t, J = 3.2 Hz, 1H, H-4'), 5.39-5.29 (m, 2H, CH=CH), 5.21 (dd, J = 10.6, 7.8 Hz, 1H, H-2'), 5.05 (dd, J = 10.6, 3.6 Hz, 1H, H-3'), 4.53 (d, J = 7.8 Hz, 1H, H-1'), 4.42 (dd, J = 12.4, 3.2 Hz, 1H, H-1a), 4.26-4.09 (m, 4H, H-2, 1b, 6'), 4.05-3.98 (m, 1H, H-3a), 3.96-3.89 (m, 1H, H-5'), 3.71 (dd, J = 10.5, 7.3 Hz, 1H, H-3b), 2.36 (t, J = 7.6 Hz, 2H, CH₂), 2.18-1.96 (m, 16H, 4×OAc, 2×CH₂), 1.68-1.57 (m, 4H, 2×CH₂), 1.38-1.17 (m, 46H, 23×CH₂), 0.91-0.84 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 174.3, 170.8, 170.5, 170.3, 170.1 (C=O); 130.1, 129.8 (CH=CH); 101.2 (C-1'), 82.4 (CH), 70.9 (C-5'), 70.6 (C-3'), 69.1 (C-2'), 68.2 (CH₂), 67.0 (C-4'), 61.4, 61.2, 34.2, 33.2, 32.0, 29.8, 29.6, 29.4, 29.2, 27.3, 25.5, 24.9, 22.8 (CH₂); 21.0, 20.8, 20.6, 14.2 (CH₃).

HRMS (ESI) m/z : $[M+Na]^+$ Calcd for C₅₁H₈₉NNaO₁₄ 962.6181; Found 962.6176.

(2R)-3-O-β-D-Galactopyranosyl-2-O-hexanoylamino-1-O-palmitoylglycerol (65)Chemical Formula: C₃₁H₅₉NO₁₀

Exact Mass: 605.4139 g/mol

Deacetylation of **60** (115 mg, 0.15 mmol) according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **65** (42 mg, 47%) as a yellowish solid.

R_f = 0.17 (CH₂Cl₂/MeOH: 100/8).

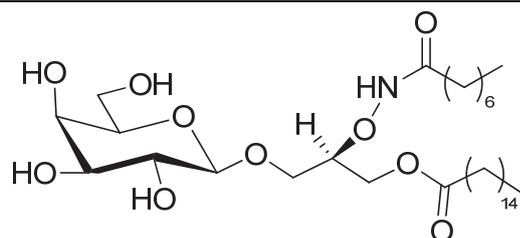
[α]_D = -17.3 (c 0.1, MeOH).

M.p. 65 °C.

¹H NMR (400 MHz, CD₃OD): δ 4.33 (dd, *J* = 12.1, 4.0 Hz, 1H, H-1a), 4.30-4.22 (m, 2H, H-1', 1b), 4.21-4.15 (m, 1H, H-2), 4.07 (dd, *J* = 11.4, 4.1 Hz, H-3a), 3.83 (dd, *J* = 3.2, 0.6 Hz, 1H, H-4'), 3.81-3.69 (m, 3H, H-3b, 6'), 3.58-3.46 (m, 3H, H-2', 3', 5'), 2.36 (t, *J* = 7.4 Hz, 2H, CH₂), 2.10 (t, *J* = 7.6 Hz, 2H, CH₂), 1.67-1.56 (m, 4H, 2×CH₂), 1.39-1.24 (m, 28H, 14×CH₂), 0.95-0.87 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.2, 173.2 (C=O); 105.5 (C-1'), 83.7, 76.8, 74.8, 72.4, 70.2 (CH); 68.9, 63.3, 62.5, 34.9, 33.7, 33.1, 32.4, 30.8, 30.6, 30.5, 30.2, 26.3, 25.9, 23.7, 23.4 (CH₂); 14.5, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₃₁H₅₉KNO₁₀ 644.3776; Found 644.3778.

(2R)-3-O-β-D-Galactopyranosyl-2-O-octanoylamino-1-O-palmitoylglycerol (66)Chemical Formula: C₃₃H₆₃NO₁₀

Exact Mass: 633.4452 g/mol

Deacetylation of **61** (81 mg, 0.10 mmol) according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **66** (32 mg, 50%) as a yellowish paste.

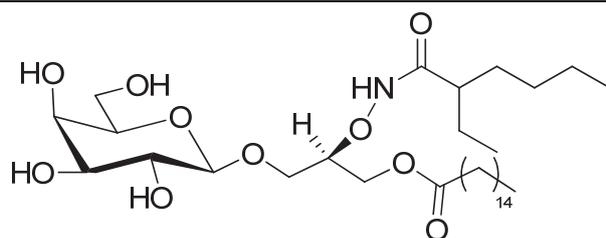
R_f = 0.18 (CH₂Cl₂/MeOH: 100/8).

[α]_D = -17.3 (c 0.1, MeOH).

¹H NMR (400 MHz, CD₃OD): δ 4.33 (dd, *J* = 12.1, 4.0 Hz, 1H, H-1a), 4.30-4.22 (m, 2H, H-1', 1b), 4.21-4.14 (m, 1H, H-2), 4.06 (dd, *J* = 11.4, 4.1 Hz, H-3a), 3.82 (d, *J* = 2.8 Hz, 1H, H-4'), 3.81-3.69 (m, 3H, H-3b, 6'), 3.59-3.44 (m, 3H, H-2', 3', 5'), 2.36 (t, *J* = 7.5 Hz, 2H, CH₂), 2.10 (t, *J* = 7.4 Hz, 2H, CH₂), 1.67-1.55 (m, 4H, 2×CH₂), 1.39-1.23 (m, 32H, 16×CH₂), 0.95-0.84 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.3, 173.2 (C=O); 105.5 (C-1'), 83.7, 76.8, 74.8, 72.5, 70.3 (CH); 68.9, 63.3, 62.5, 34.9, 33.8, 33.1, 32.9, 30.8, 30.6, 30.5, 30.2, 30.1, 26.6, 25.9, 23.8, 23.7 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₃H₆₃NNaO₁₀ 656.4350; Found 656.4346.

(2R)-2-O-(2-Ethylhexanoyl)amino-3-O-β-D-galactopyranosyl-1-O-palmitoylglycerol (67)Chemical Formula: C₃₃H₆₃NO₁₀

Exact Mass: 633.4452 g/mol

Deacetylation of **62** (64 mg, 0.08 mmol) according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **67** (26 mg, 52%) as a yellowish paste.

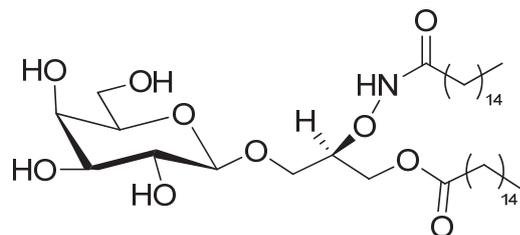
R_f = 0.25 (CH₂Cl₂/MeOH: 100/8).

[α]_D = -15.7 (c 0.1, MeOH).

¹H NMR (400 MHz, CD₃OD): δ 4.34 (dd, *J* = 12.2, 4.2 Hz, 1H, H-1a), 4.30-4.23 (m, 2H, H-1',1b), 4.22-4.14 (m, 1H, H-2), 4.12-4.05 (m, H-3a), 3.83 (d, *J* = 3.2 Hz, 1H, H-4'), 3.81-3.70 (m, 3H, H-3b,6'), 3.60-3.45 (m, 3H, H-2',3',5'), 2.37 (t, *J* = 7.5 Hz, 2H, CH₂), 2.02-1.92 (m, 1H, CH), 1.69-1.54 (m, 4H, 2×CH₂), 1.52-1.22 (m, 30H, 15×CH₂), 0.97-0.87 (m, 9H, 3×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.7, 175.2 (C=O); 105.6 (C-1'), 83.9, 76.8, 74.9, 72.5, 70.2 (CH); 68.9, 63.2, 62.5 (CH₂); 46.6 (CH), 34.8, 33.3, 33.1, 30.8, 30.6, 30.5, 30.2, 26.9, 25.9, 23.7, 23.7 (CH₂); 14.5, 14.3, 12.4 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₃H₆₃NNaO₁₀ 656.4350; Found 656.4353.

(2R)-3-O-β-D-Galactopyranosyl-1-O-palmitoyl-2-O-palmitoylaminoglycerol (68)Chemical Formula: C₄₁H₇₉NO₁₀

Exact Mass: 745.5704 g/mol

Deacetylation of **63** (55 mg, 0.06 mmol) according to the general **procedure H** and purification by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) led to compound **68** (26 mg, 58%) as a white powder.

R_f = 0.38 (CH₂Cl₂/MeOH: 10/1).

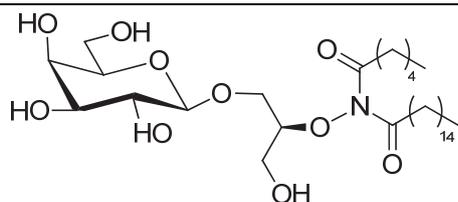
[α]_D = -13.3 (c 0.1, MeOH).

M.p. 117 °C.

¹H NMR (400 MHz, CD₃OD): δ 4.34 (dd, *J* = 12.1, 4.0 Hz, 1H, H-3a), 4.31-4.23 (m, 2H, H-1', 3b), 4.22-4.15 (m, 1H, H-2), 4.06 (dd, *J* = 11.4, 4.1 Hz, H-1a), 3.87-3.82 (m, 1H, H-4'), 3.82-3.70 (m, 3H, H-1b, 6'), 3.61-3.45 (m, 3H, H-2', 3', 5'), 2.36 (t, *J* = 7.4 Hz, 2H, CH₂), 2.10 (t, *J* = 7.3 Hz, 2H, CH₂), 1.71-1.57 (m, 4H, 2×CH₂), 1.42-1.24 (m, 48H, 24×CH₂), 0.97-0.87 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.2, 173.2 (C=O); 105.6 (C-1'), 83.8, 76.8, 75.0, 72.5, 70.3 (CH); 68.9, 63.4, 62.6, 34.9, 33.8, 33.1, 33.0, 30.8, 30.6, 30.5, 30.4, 30.2, 26.6, 25.9, 23.8, 23.7 (CH₂); 14.5, 14.4 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₄₁H₇₉NaNO₁₀ 768.5602; Found 768.5606.

(2S)-1-O-β-D-Galactopyranosyl-2-O-(N-hexanoyl-N'-palmitoyl)aminoglycerol (70)Chemical Formula: C₃₁H₅₉NO₁₀

Exact Mass: 605.4139 g/mol

Deacetylation of compound **60** (94 mg, 0.12 mmol) with Na/MeOH according to the general **procedure I** led to compound **70** (70 mg, 96%) as a yellowish paste.

R_f = 0.27 (CH₂Cl₂/MeOH: 5/1).

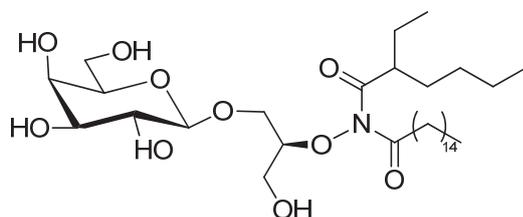
[α]_D = -23.3 (c 0.1, MeOH).

¹H NMR (400 MHz, CD₃OD): δ 4.29 (d, 1H, d, *J* = 7.8 Hz, H-1'), 4.03 (dd, *J* = 11.0, 4.0 Hz, 1H, H-1a), 4.01-3.95 (m, 1H, H-2), 3.85-3.81 (m, 1H, H-4'), 3.80-3.71 (m, 3H, H-1b,6'), 3.71-3.61 (m, 2H, H-3), 3.58-3.46 (m, 3H, H-2',3',5'), 2.31 (t, *J* = 7.5 Hz, 2H, CH₂), 2.12 (t, *J* = 7.4 Hz, 2H, CH₂), 1.68-1.55 (m, 4H, 2×CH₂), 1.43-1.23 (m, 28H, 14×CH₂), 0.97-0.86 (m, 6H, 2×CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 175.9, 173.8 (C=O); 105.4 (C-1'), 87.5, 76.8, 74.9, 72.5, 70.3 (CH); 69.0, 62.5, 61.0, 34.8, 33.6, 33.1, 32.4, 30.8, 30.7, 30.6, 30.5, 30.4, 30.2, 26.3, 26.0, 23.7, 23.4 (CH₂); 14.5, 14.3 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₁H₅₉NaNO₁₀ 628.4037; Found 628.4034.

**(2*S*)-1-*O*- β -D-Galactopyranosyl-2-*O*-{*N*-(2-ethylhexanoyl)-*N'*-palmitoyl}aminoglycerol
(71)**



Chemical Formula: C₃₃H₆₃NO₁₀

Exact Mass: 633.4452 g/mol

Deacetylation of compound **62** (92 mg, 0.115 mmol) with Na/MeOH according to the general **procedure I** led to compound **71** (72 mg, 99%) as a yellowish paste.

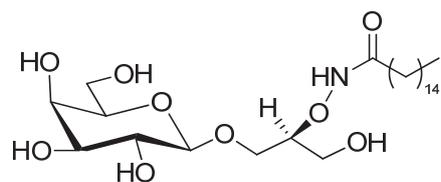
R_f = 0.33 (CH₂Cl₂/MeOH: 5/1).

[α]_D = -17.3 (*c* 0.1, MeOH).

¹H NMR (400 MHz, CD₃OD): δ 4.29 (d, 1H, *d*, *J* = 7.8 Hz, H-1'), 4.05 (dd, *J* = 11.0, 3.7 Hz, 1H, H-1a), 4.02-3.95 (m, 1H, H-2), 3.83 (d, *J* = 3.2 Hz, 1H, H-4'), 3.81-3.71 (m, 3H, H-1b,6'), 3.71-3.62 (m, 2H, H-3), 3.60-3.47 (m, 3H, H-2',3',5'), 2.31 (t, *J* = 7.4 Hz, 2H, CH₂), 2.04-1.94 (m, 1H, CH), 1.67-1.54 (m, 4H, 2 \times CH₂), 1.52-1.39 (m, 2H, CH₂), 1.39-1.21 (m, 28H, 14 \times CH₂), 0.98-0.86 (m, 9H, 3 \times CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 176.3, 175.8 (C=O); 105.4 (C-1'), 87.6, 76.7, 74.9, 72.5, 70.2 (CH); 68.9, 62.5, 60.9 (CH₂); 46.6 (CH), 34.8, 33.2, 33.1, 30.8, 30.7, 30.6, 30.5, 30.4, 30.2, 26.9, 26.0, 23.7, 23.6 (CH₂); 14.5, 14.3, 12.3 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₃₃H₆₃KNO₁₀ 672.4089; Found 672.4088.

(2S)-1-O-β-D-Galactopyranosyl-2-O-palmitoylaminoglycerol (72)Chemical Formula: C₂₅H₄₉NO₉

Exact Mass: 507.3407 g/mol

Deacetylation of compound **63** (50 mg, 0.055 mmol) with Na/MeOH according to the general **procedure I** led to compound **72** (25 mg, 68%) as a yellowish solid.

R_f = 0.44 (CH₂Cl₂/MeOH: 5/1).

[α]_D = -25.7 (c 0.1, MeOH).

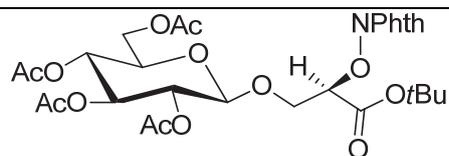
M.p. 91 °C.

¹H NMR (400 MHz, CD₃OD): δ 4.29 (d, 1H, d, *J* = 7.4 Hz, H-1'), 4.04 (dd, *J* = 11.0, 4.0 Hz, 1H, H-1a), 4.01-3.96 (m, 1H, H-2), 3.83 (d, *J* = 2.8 Hz, 1H, H-4'), 3.81-3.71 (m, 3H, H-1b,6'), 3.71-3.62 (m, 2H, H-3), 3.59-3.46 (m, 3H, H-2',3',5'), 2.13 (t, *J* = 7.4 Hz, 2H, CH₂), 1.68-1.56 (m, 2H, CH₂), 1.42-1.22 (m, 24H, 12×CH₂), 0.95-0.85 (m, 3H, CH₃).

¹³C NMR (100 MHz, CD₃OD): δ 173.9 (C=O), 105.4 (C-1'), 87.4, 76.8, 74.9, 72.5, 70.3 (CH); 69.0, 62.5, 60.9, 33.7, 33.1, 30.8, 30.6, 30.5, 30.4, 30.2, 26.6, 23.7 (CH₂); 14.5 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₅H₄₉NNaO₉, 530.3305; Found 530.3311.

***tert*-Butyl (2*R*)-2-phthalimidooxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]-propanoate (73)**



Chemical Formula: C₂₉H₃₅NO₁₅

Exact Mass: 637.2007 g/mol

To a solution of compound **33** (0.83 g, 1.46 mmol) in CH₂Cl₂ (30 mL), were added PCC (98%, 0.63 mg, 2.92 mmol), Ac₂O (1.38 ml, 14.63 mmol) and *t*-BuOH (2.80 mL, 29.27 mmol). The resulting mixture was stirred at room temperature and then extracted with CH₂Cl₂ (3×30 mL). The combined organic layers were washed with brine (30 mL), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) to give compound **73** (0.57 g, 61%) as a white solid.

R_f = 0.44 (petroleum ether/EtOAc: 1/1).

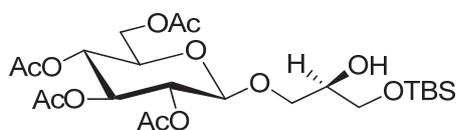
[α]_D = +2.0 (*c* 0.1, CHCl₃).

M.p. 64 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.91-7.76 (m, 4H, Phth), 5.21 (t, *J* = 9.6 Hz, 1H, H-3'), 5.07 (t, *J* = 9.6 Hz, 1H, H-4'), 4.99 (dd, *J* = 9.6, 7.8 Hz, 1H, H-2'), 4.93 (dd, *J* = 7.3, 3.6 Hz, 1H, H-2), 4.63 (d, *J* = 7.8 Hz, 1H, H-1'), 4.30-4.09 (m, 4H, H-3,6'), 3.74 (ddd, *J* = 10.0, 4.6, 2.2 Hz, 1H, H-5'), 2.11, 2.08, 2.03, 2.00 (4×s, 12H, 4×OAc), 1.47 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.7, 170.2, 169.6, 169.5, 165.8 (C=O), 162.9 (C=O, Phth); 134.7 (CH, Phth), 128.8 (C_q), 123.7 (CH, Phth), 100.7 (C-1'), 85.1 (CH), 83.5 (C_q, *t*-Bu), 72.8 (C-3'), 71.8 (C-5'), 71.0 (C-2'), 68.3 (C-4'), 67.7, 61.9 (CH₂); 27.8, 20.8, 20.7, 20.6 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₉H₃₅NNaO₁₅ 660.1904; Found 660.1899.

(2R)-1-O-tert-Butyldimethylsilyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-glycerol (74)Chemical Formula: C₂₃H₄₀O₁₂Si

Exact Mass: 536.2289 g/mol

To a solution of compound **31** (54 mg, 0.1 mmol) in CH₂Cl₂ (2 mL) and pyridine (0.05 mL) at -40 °C under argon, was added dropwise trifluoromethanesulfonic anhydrous (34 μL, 0.2 mmol). After 1 h 30 min, the mixture was diluted with CH₂Cl₂ (20 mL), washed with aq HCl (1N, 10 mL) and brine (20 mL), dried over MgSO₄, filtered, evaporated. The residue containing triflate derivative was treated with NaNO₂ (69 mg, 1 mmol) in DMF/H₂O (2 mL, 1/1). After stirring at room temperature overnight, the mixture was diluted with H₂O (10 mL), and the product was extracted with CH₂Cl₂ (3×15 mL). The combined organic layers were washed with brine (15 mL), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/0.5) to give compound **74** (24 mg, 44%) as a yellowish paste.

R_f = 0.35 (CH₂Cl₂/MeOH: 100/1).

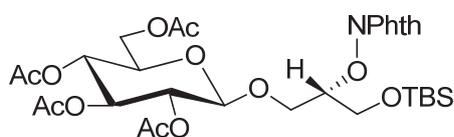
[α]_D = +7.7 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.20 (t, *J* = 9.6 Hz, 1H, H-3'), 5.06 (t, *J* = 9.7 Hz, 1H, H-4'), 4.99 (dd, *J* = 9.6, 8.1 Hz, 1H, H-2'), 4.55 (d, *J* = 8.2 Hz, 1H, H-1'), 4.23 (dd, *J* = 12.4, 5.0 Hz, 1H, H-6'a), 4.13 (dd, *J* = 12.4, 2.3 Hz, 1H, H-6'b), 3.84 (dd, *J* = 10.1, 4.5 Hz, 1H, H-3a), 3.79-3.69 (m, 2H, H-2,5'), 3.66 (dd, *J* = 10.1, 5.5 Hz, 1H, H-3b), 3.63-3.55 (m, 2H, H-1), 2.08, 2.04, 2.02, 2.00 (4×s, 12H, 4×OAc), 0.88 (s, 9H, *t*-Bu), 0.05 (s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.4, 169.6, 169.5 (C=O); 101.3 (C-1'), 72.8 (C-3'), 72.0 (C-5'), 71.4 (CH₂), 71.4 (C-2'), 70.6 (CH), 68.5 (C-4'), 63.6, 62.1 (CH₂); 26.0, 20.8, 20.7 (CH₃); 18.4 (C_q, *t*-Bu), -5.3 (CH₃, Si(CH₃)₂).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₂₃H₄₀NaO₁₂Si 559.2187; Found 559.2182.

(2*S*)-2-*O*-Phthalimido-1-*O*-*tert*-butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)glycerol (75)



Chemical Formula: C₃₁H₄₃NO₁₄Si

Exact Mass: 681.2453 g/mol

The phthalimido group was introduced to compound **74** (43 mg, 0.08 mmol) via Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **75** (48 mg, 88%) as a yellowish paste.

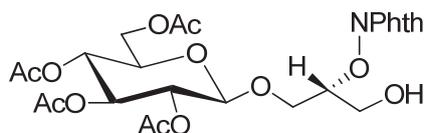
$R_f = 0.63$ (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = +13.7$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.94-7.72 (m, 4H, Phth), 5.21 (t, $J = 9.6$ Hz, 1H, H-3'), 5.04 (t, $J = 9.8$ Hz, 1H, H-4'), 4.95 (dd, $J = 9.6, 7.9$ Hz, 1H, H-2'), 4.70 (d, $J = 7.8$ Hz, 1H, H-1'), 4.43-4.32 (m, 1H, H-2), 4.25 (dd, $J = 11.9, 4.6$ Hz, 1H, H-6'a), 4.19-4.08 (m, 2H, H-3a,6'b), 4.04-3.89 (m, 3H, H-1,3b), 3.78-3.70 (m, 1H, H-5'), 2.08, 2.07, 2.01, 1.99 (4×s, 12H, 4×OAc), 0.84 (s, 9H, *t*-Bu), 0.02, 0.01 (2×s, 6H, Si(CH₃)₂).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.4, 169.6 (C=O), 163.8 (C=O, Phth); 134.6 (CH, Phth), 129.1 (C_q), 123.6 (CH, Phth), 101.2 (C-1'), 87.0 (C-2), 72.9 (C-3'), 71.8 (C-5'), 71.3 (C-2'), 68.5 (C-4'), 67.2, 62.0, 61.3 (CH₂); 25.8, 20.9, 20.7 (CH₃); 18.3 (C_q, *t*-Bu), -5.5 (CH₃, Si(CH₃)₂).

HRMS (ESI) m/z : [M+Na]⁺ Calcd for C₃₁H₄₃NaNO₁₄Si 704.2351; Found 704.2353.

(2R)-2-O-Phthalimido-1-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucofuranosyl)glycerol (76)Chemical Formula: C₂₅H₂₉NO₁₄

Exact Mass: 567.1588 g/mol

- (a) To a solution of compound **80** (0.43 g, 0.53 mmol) in MeOH (10 mL), was added AcCl (5.8 μL, 0.08 mmol) at 0 °C. The resulting mixture was stirred at room temperature for 5 h under argon. After completion of the reaction, 5% NaHCO₃ was added to neutralize the reaction mixture, and then the solvent was removed under vacuum. The residue was taken up in H₂O-CH₂Cl₂ (1/1, 50 mL) and the aqueous layer was extracted with CH₂Cl₂ (3×20 mL). The combined organic layers were washed with brine (30 mL), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (petroleum ether/EtOAc: 2/3) to give compound **76** (0.21 g, 71%) as a white solid.
- (b) Compound **76** also could be obtained from desilylation of compound **75** (27 mg, 0.04 mmol) using AcCl according to the general **procedure B**. Purification by column chromatography over silica gel (petroleum ether/EtOAc = 2/3) gave compound **76** (20 mg, 87%) as a white solid.

$R_f = 0.49$ (petroleum ether/EtOAc: 1/2).

$[\alpha]_D = +1.7$ (c 0.1, CHCl₃).

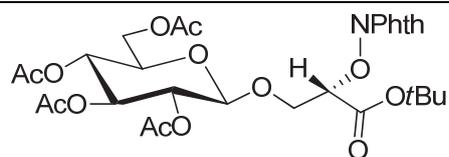
M.p. 156 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.96-7.76 (m, 4H, Phth), 5.23 (t, $J = 9.4$ Hz, 1H, H-3'), 5.08 (t, $J = 9.6$ Hz, 1H, H-4'), 4.86 (dd, $J = 9.6, 7.8$ Hz, 1H, H-2'), 4.72 (d, $J = 8.2$ Hz, 1H, H-1'), 4.35-4.09 (m, 4H, H-3a, 2, 6'), 4.03 (dd, $J = 11.0, 6.1$ Hz, 1H, H-3b), 3.87-3.75 (m, 3H, H-1, 5'), 2.10, 2.05, 2.03, 2.00 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.5 (C=O), 164.5 (C=O, Phth); 135.0 (CH, Phth), 128.8 (C_q), 124.0 (CH, Phth), 101.2 (C-1'), 88.2 (CH), 72.8 (C-3'), 71.9 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 67.3, 61.9, 60.3 (CH₂); 20.8, 20.7 (OAc).

HRMS (ESI) m/z : $[M+K]^+$ Calcd for C₂₅H₂₉KNO₁₄ 606.1225; Found 606.1223.

***tert*-Butyl (2*S*)-2-phthalimodooxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy] propanoate (77)**



Chemical Formula: C₂₉H₃₅NO₁₅

Exact Mass: 637.2007 g/mol

To a solution of compound **76** (193 mg, 0.34 mmol) in CH₂Cl₂ (10 mL), were added PCC (98%, 147 mg, 0.68 mmol), Ac₂O (0.32 ml, 3.4 mmol) and *t*-BuOH (0.64 mL, 6.8 mmol). The resulting mixture was stirred at room temperature and then extracted with CH₂Cl₂ (3×20 mL). The combined organic layers were washed with brine (20 mL), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) to give compound **77** (120 mg, 55%) as a colourless paste.

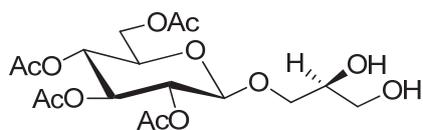
R_f = 0.53 (petroleum ether/EtOAc: 1/1).

[α]_D = +16.0 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 7.88-7.75 (m, 4H, Phth), 5.29-5.20 (m, 1H, H-3'), 5.10 (t, *J* = 9.9 Hz, 1H, H-4'), 5.02-4.95 (m, 2H, H-1',2'), 4.77 (t, *J* = 3.4 Hz, 1H, H-2), 4.33-4.09 (m, 4H, H-3,6'), 3.83 (ddd, *J* = 10.0, 4.1, 2.2 Hz, 1H, H-5'), 2.12, 2.09, 2.03, 2.00 (4×s, 12H, 4×OAc), 1.48 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.3, 169.8, 169.6, 165.2 (C=O), 163.3 (C=O, Phth); 134.9 (CH, Phth), 128.9 (C_q), 123.8 (CH, Phth), 100.8 (C-1'), 84.6 (CH), 83.4 (C_q, *t*-Bu), 73.1 (C-3'), 71.7 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.5, 62.0 (CH₂); 28.0, 20.8, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₂₉H₃₅KNO₁₅ 676.1644; Found 676.1646.

(2S)-1-O-(2',3',4',6'-Tetra-O-acetyl-β-D-glucopyranosyl)glycerol (78)Chemical Formula: C₁₇H₂₆O₁₂

Exact Mass: 422.1424 g/mol

Compound **32** (1.41 g, 2.07 mmol) was hydrogenated in MeOH (50 mL), in the presence of 10% Pd/C (0.5 g) at room temperature. After completion of the reaction, the catalyst was filtered off, the filtrate evaporated. Purification by column chromatography (CH₂Cl₂/MeOH: 100/3) afforded compound **78** (0.66 g, 76%) as a colourless paste.

R_f = 0.30 (CH₂Cl₂/MeOH: 25/1).

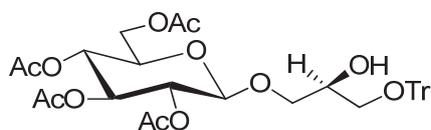
[α]_D = +4.3 (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.22 (t, *J* = 9.6 Hz, 1H, H-3'), 5.07 (t, *J* = 10.1 Hz, 1H, H-4'), 5.00 (dd, *J* = 9.6, 8.0 Hz, 1H, H-2'), 4.56 (d, *J* = 7.8 Hz, 1H, H-1'), 4.23 (dd, *J* = 12.4, 5.0 Hz, 1H, H-6'a), 4.18 (dd, *J* = 12.4, 2.8 Hz, 1H, H-6'b), 3.91-3.81 (m, 2H, H-2,3a), 3.78-3.69 (m, 2H, H-3b,5'), 3.67 (dd, *J* = 11.5, 4.1 Hz, 1H, H-1a), 3.60 (dd, *J* = 11.4, 5.5 Hz, 1H, H-1b), 2.56 (s, 2H, 2×OH), 2.11, 2.07, 2.04, 2.02 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.8, 169.5 (C=O); 101.3 (C-1'), 72.6 (C-3'), 72.0 (C-5'), 71.9 (CH₂), 71.3 (C-2'), 70.6 (CH), 68.4 (C-4'), 63.3, 61.9 (CH₂); 20.7, 20.6 (OAc).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₁₇H₂₆NaO₁₂ 445.1322; Found 445.1323.

(2*R*)-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)-1-*O*-triphenylmethylglycerol (79)



Chemical Formula: C₃₆H₄₀O₁₂

Exact Mass: 664.2520 g/mol

To a solution of compound **78** (0.45 g, 1.07 mmol) in pyridine (10 mL), was added TrCl (0.36 g, 1.28 mmol). The mixture was refluxed for 24 h and then cooled to room temperature. The solution was concentrated under vacuum to a syrup. The residue was taken up in H₂O-CH₂Cl₂ (1/1, 100 mL) and the aqueous layer was extracted with CH₂Cl₂ (3×30 mL). The combined organic layers were washed with brine (50 mL), dried over MgSO₄, filtered, evaporated and purified by column chromatography over silica gel (CH₂Cl₂/ MeOH: 100/1) to afford compound **78** (0.17 g, conversion: 62%) and compound **79** (0.4 g, corrected yield: 92%) as a white powder.

$R_f = 0.79$ (CH₂Cl₂/ MeOH: 100/3).

$[\alpha]_D = -1.0$ (c 0.1, CHCl₃).

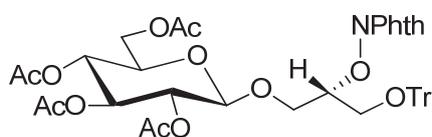
M.p. 63 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.47-7.21 (m, 15H, Tr), 5.20 (t, $J = 9.6$ Hz, 1H, H-3'), 5.06 (t, $J = 9.6$ Hz, 1H, H-4'), 4.98 (dd, $J = 9.6, 8.0$ Hz, 1H, H-2'), 4.54 (d, $J = 7.8$ Hz, 1H, H-1'), 4.22 (dd, $J = 12.4, 5.0$ Hz, 1H, H-6'a), 4.13 (dd, $J = 12.4, 2.3$ Hz, 1H, H-6'b), 3.96-3.87 (m, 2H, H-2,3a), 3.82-3.74 (m, 1H, H-3b), 3.71 (ddd, $J = 10.0, 5.1, 2.4$ Hz, 1H, H-5'), 3.20 (dd, $J = 9.2, 5.5$ Hz, 1H, H-1a), 3.14 (dd, $J = 9.2, 5.5$ Hz, 1H, H-1b), 2.65 (s, 1H, OH), 2.03, 2.02, 2.00, 1.97 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.7, 170.3, 169.5 (C=O); 143.8 (C_q), 128.7, 127.9, 127.2 (CH, Tr); 101.3 (C-1'), 86.7 (C_q, Tr), 72.7 (C-3'), 72.3 (CH₂), 71.8 (C-5'), 71.4 (C-2'), 69.8 (CH), 68.4 (C-4'), 64.1, 62.0 (CH₂); 20.7, 20.7 (OAc).

HRMS (ESI) m/z : $[M+K]^+$ Calcd for C₃₆H₄₀KO₁₂ 703.2157; Found 703.2159.

(2*S*)-2-*O*-Phthalimido-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)-1-*O*-triphenyl methylglycerol (80**)**



Chemical Formula: C₄₄H₄₃NO₁₄

Exact Mass: 809.2684 g/mol

The phthalimido group was introduced to compound **79** (0.37 g, 0.56 mmol) via Mitsunobu reaction according to the general **procedure C**. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 3/1) afforded compound **80** (0.44 g, 97%) as a white solid.

$R_f = 0.83$ (petroleum ether/EtOAc: 1/1).

$[\alpha]_D = -3.3$ (c 0.1, CHCl₃).

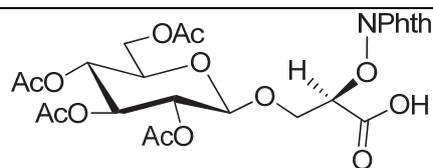
M.p. 115 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.84-7.72 (m, 4H, Phth), 7.42-7.13 (m, 15H, Tr), 5.17 (t, $J = 9.4$ Hz, 1H, H-3'), 5.03 (t, $J = 9.6$ Hz, 1H, H-4'), 4.86 (dd, $J = 9.6, 8.0$ Hz, 1H, H-2'), 4.68 (d, $J = 8.2$ Hz, 1H, H-1'), 4.38-4.30 (m, 1H, H-2), 4.25 (dd, $J = 12.4, 4.6$ Hz, 1H, H-6'a), 4.11 (dd, $J = 12.4, 2.3$ Hz, 1H, H-6'b), 4.06 (dd, $J = 12.4, 3.6$ Hz, 1H, H-3a), 3.96 (dd, $J = 12.4, 4.1$ Hz, 1H, H-3b), 3.76-3.66 (m, 1H, H-5'), 3.58 (dd, $J = 11.4, 4.1$ Hz, 1H, H-1a), 3.48 (dd, $J = 11.4, 6.0$ Hz, 1H, H-1b), 2.05, 2.01, 1.98, 1.85 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.4, 169.6, 169.5 (C=O), 163.6 (C=O, Phth); 143.7 (C_q), 134.6 (CH, Phth), 129.1 (C_q), 128.7, 127.9, 127.2 (CH, Tr); 123.6 (CH, Phth), 101.0 (C-1'), 87.3 (C_q, Tr), 86.2 (CH), 72.9 (C-3'), 71.8 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.8, 62.5, 62.0 (CH₂); 20.9, 20.7, 20.6 (OAc).

HRMS (ESI) m/z : $[M+K]^+$ Calcd for C₄₄H₄₃KNO₁₄ 848.2321; Found 848.2323.

(2R)-2-Phthalimidooxy-3-[(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)oxy]propanoic acid (81**)**



Chemical Formula: C₂₅H₂₇NO₁₅

Exact Mass: 581.1381 g/mol

- (a) To a solution of compound **80** (190 mg, 0.3 mmol) in CH₂Cl₂ (6 mL), was added TFA (0.6 mL). The resulting mixture was stirred at room temperature overnight. After the reaction finished, most of the solvent was evaporated, and the residue was dried under high vacuum (oil pump) until no acidic smell could be detected. The residue was purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 50/1) to give compound **81** (146 mg, 84%) as a white solid.
- (b) The carboxylic acid derivative **81** also could be synthesized by oxidation of the (2*S*)-1-*O*-glycosyl phthalimidooxy alcohol **33** with Jones reagent. The Jones reagent was prepared by dissolving 2.67 g of chromium trioxide (CrO₃) in 2.3 mL of concentrated sulfuric acid, and then the mixture was diluted to 10 mL with water. Compound **33** was treated with Jones reagent (3 eq.) at room temperature for 16 h, affording compound **81** in 62% yield.

R_f = 0.14 (CH₂Cl₂/MeOH: 20/1).

[α]_D = -45.3 (*c* 0.1, CHCl₃).

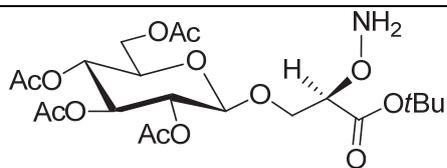
M.p. 218 °C.

¹H NMR (400 MHz, CDCl₃): δ 7.96-7.81 (m, 4H, Phth), 5.17 (t, *J* = 9.4 Hz, 1H, H-3'), 5.01-4.88 (m, 3H, H-2,2',4'), 4.58 (d, *J* = 7.8 Hz, 1H, H-1'), 4.52 (d, *J* = 12.4 Hz, 1H, H-3a), 4.23-4.06 (m, 3H, H-3b,6'), 3.70 (ddd, *J* = 10.0, 4.7, 2.6 Hz, 1H, H-5'), 2.10, 2.09, 2.01, 1.99 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.3, 169.8, 169.6, 168.4 (C=O), 164.2 (C=O, Phth); 135.5 (CH, Phth), 128.5 (C_q), 124.5 (CH, Phth), 101.4 (C-1'), 85.9 (CH), 72.7 (C-3'), 72.0 (C-5'), 70.9 (C-2'), 68.9 (CH₂), 68.4 (C-4'), 62.0 (CH₂), 20.9, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₂₅H₂₈NO₁₅ 582.1459; Found 582.1453.

***tert*-Butyl (2*R*)-2-aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propionate (**82**)**

Chemical Formula: C₂₁H₃₃NO₁₃Exact Mass: 507.1952 g/mol

Hydrazinolysis of compound **80** (100 mg, 0.16 mmol) by hydrazine under mild conditions according to the general **procedure E** and purification by column chromatography over silica gel (petroleum ether/EtOAc = 1/1) afforded compound **82** (75 mg, 93%) as a white paste.

R_f = 0.26 (petroleum ether/EtOAc: 1/1).

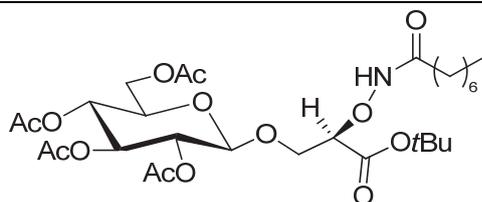
[α]_D = -31.0 (*c* 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 5.21 (t, *J* = 9.6 Hz, 1H, H-3'), 5.09 (t, *J* = 9.6 Hz, 1H, H-4'), 5.00 (dd, *J* = 9.6, 8.0 Hz, 1H, H-2'), 4.63 (d, *J* = 7.8 Hz, 1H, H-1'), 4.33 (dd, *J* = 7.8, 2.8 Hz, 1H, H-2), 4.26 (dd, *J* = 12.4, 5.0 Hz, 1H, H-6'a), 4.18-4.06 (m, 2H, H-3a,6'b), 3.83 (dd, *J* = 11.6, 7.6 Hz, 1H, H-3b), 3.70 (ddd, *J* = 9.9, 4.6, 2.3 Hz, 1H, H-5'), 2.09, 2.07, 2.03, 2.01 (4 \times s, 12H, 4 \times OAc), 1.48 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.4, 169.5, 168.9 (C=O); 101.1 (C-1'), 83.3 (CH), 82.3 (C_q, *t*-Bu), 72.8 (C-3'), 71.9 (C-5'), 71.3 (C-2'), 68.4 (C-4'), 68.3, 62.0 (CH₂); 28.1, 20.8, 20.8, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+H]⁺ Calcd for C₂₁H₃₄NO₁₃ 508.2030; Found 508.2033.

***tert*-Butyl (2*R*)-*O*-octanoylamino-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]-propanoate (**83**)**

Chemical Formula: C₂₉H₄₇NO₁₄

Exact Mass: 633.2997 g/mol

Compound **83** was prepared from compound **82** (80 mg, 0.16 mmol) and octanoic acid (25 μ L, 0.16 mmol) according to the general **procedure F** for *N*-oxyamide formation. Purification by column chromatography over silica gel (petroleum ether/EtOAc: 2/1) afforded compound **83** (85 mg, 84%) as a paste.

R_f = 0.41 (petroleum ether/EtOAc: 1/1).

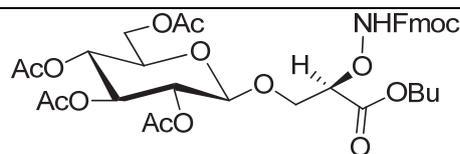
$[\alpha]_D = -23.0$ (c 0.1, CHCl₃).

¹H NMR (400 MHz, CDCl₃): δ 8.92 (s, 1H, NH), 5.22 (t, J = 9.6 Hz, 1H, H-3'), 5.09 (t, J = 9.6 Hz, 1H, H-4'), 5.01 (t, J = 9.2 Hz, 1H, H-2'), 4.85-4.65 (m, 1H, H-1'), 4.60-4.45 (m, 1H, H-2), 4.27 (dd, J = 12.4, 4.6 Hz, 1H, H-6'a), 4.24-4.13 (m, 2H, H-3a,6'b), 3.97-3.86 (m, 1H, H-3b), 3.81-3.71 (m, 1H, H-5'), 2.20-2.01 (m, 14H, 4 \times OAc, CH₂), 1.68-1.59 (m, 2H, CH₂), 1.49 (s, 9H, *t*-Bu), 1.38-1.25 (m, 8H, 4 \times CH₂), 0.88 (t, J = 6.4 Hz, 3H, CH₃).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.8, 169.5, 167.8 (C=O); 100.4 (C-1'), 83.2 (CH), 83.0 (C_q, *t*-Bu) 72.8 (C-3'), 71.9 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 67.7, 61.8, 33.2, 31.7, 29.2 (CH₂); 28.1 (CH₃), 25.4, 22.7 (CH₂), 20.8, 20.7, 14.1 (CH₃).

HRMS (ESI) m/z : [M+H]⁺ Calcd for C₂₉H₄₈NO₁₄ 634.3075; Found 634.3069.

***tert*-Butyl (2*R*)-2-*N*-(9-fluorenylmethoxycarbonyl)aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoate (**84**)**



Chemical Formula: C₃₆H₄₃NO₁₅

Exact Mass: 729.2633 g/mol

To a solution of the oxyamine derivative **82** in anhydrous CH₂Cl₂ (5 mL), was added Fmoc-OSu (0.48 g, 1.41 mmol). The resulting mixture was stirred at room temperature for 4 days. After the completion of the reaction, the solution was concentrated to a syrup which was purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/0.5) to give compound **84** (0.27 g, 79%) as a white powder.

R_f = 0.50 (CH₂Cl₂/MeOH: 100/1).

[α]_D = +4.7 (*c* 0.1, CHCl₃).

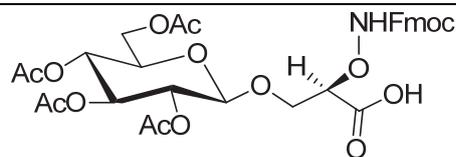
M.p. 76 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.06 (s, 1H, NH), 7.82-7.29 (m, 8H, Fmoc), 5.07 (t, *J* = 9.4 Hz, 1H, H-3'), 5.09 (t, *J* = 9.8 Hz, 1H, H-4'), 5.01 (dd, *J* = 9.4, 8.0 Hz, 1H, H-2'), 4.70 (d, *J* = 7.8 Hz, 1H, H-1'), 4.47 (d, *J* = 7.4 Hz, 2H, CH₂-Fmoc), 4.44-4.39 (m, 1H, H-2), 4.31-4.22 (m, 2H, CH-Fmoc, H-6'a), 4.19-4.10 (m, 2H, H-3a,6'b), 3.89 (dd, *J* = 11.6, 6.0 Hz, 1H, H-3b), 3.71 (ddd, *J* = 9.9, 4.5, 2.4 Hz, 1H, H-5'), 2.07, 2.04, 2.02, 2.00 (4×s, 12H, 4×OAc), 1.49 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.6, 169.5, 167.6, 156.8 (C=O); 143.5, 143.5, 141.4 (C_q); 127.9, 127.2, 125.2, 120.1 (CH, Fmoc); 100.4 (C-1'), 83.7 (CH), 83.1 (C_q, *t*-Bu), 72.9 (C-3'), 71.9 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 67.6, 67.4, 61.9 (CH₂); 47.0 (CH), 28.1, 20.8, 20.8, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₃₆H₄₃KNO₁₅ 768.2270; Found 768.2273.

(2*R*)-2-*N*-(9-Fluorenylmethoxycarbonyl)aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoic acid (85**)**



Chemical Formula: C₃₂H₃₅NO₁₅

Exact Mass: 673.2007 g/mol

To a solution of compound **84** (124 mg, 0.17 mmol) in CH₂Cl₂ (3 mL), was added TFA (0.3 mL). The resulting mixture was stirred at room temperature overnight. After the reaction finished, most of the solvent was evaporated, and the residue was dried under high vacuum (oil pump) until no acidic smell could be detected. The residue was purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 20/1) to give compound **85** (114 mg, 100%) as a white powder.

R_f = 0.39 (CH₂Cl₂/MeOH: 20/1).

[α]_D = -12.0 (*c* 0.1, CHCl₃).

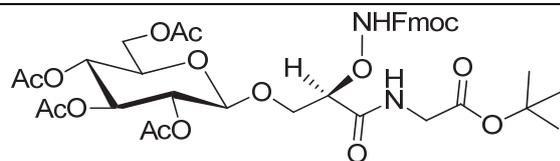
M.p. 90 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.06 (s, 1H, NH), 7.87-7.30 (m, 8H, Fmoc), 5.25 (t, *J* = 9.4 Hz, 1H, H-3'), 5.08 (t, *J* = 9.9 Hz, 1H, H-4'), 5.02 (m, 1H, H-2'), 4.66-4.50 (m, 4H, H-1', 2, CH₂-Fmoc), 4.39-4.14 (m, 4H, H-3a,6', CH-Fmoc), 3.89-3.81 (m, 1H, H-3b), 3.78-3.70 (m, 1H, H-5'), 2.08, 2.06, 2.04, 2.03 (4×s, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.8, 170.3, 169.6, 169.1, 159.8 (C=O); 143.0, 141.5 (C_q); 128.2, 127.4, 125.1, 125.0, 120.3 (CH, Fmoc); 101.4 (C-1'), 86.4 (CH), 72.3 (C-3'), 72.1 (C-5'), 71.7 (C-2'), 68.9 (CH₂), 68.3 (C-4'), 61.8 (CH₂), 46.9 (CH), 21.0, 20.9, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₂H₃₅NNaO₁₅ 696.1904; Found 696.1906.

***N*-{(2*R*)-2-*N*-(9-Fluorenylmethoxycarbonyl)aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoyl}-glycine *tert*-butyl ester (**86**)**



Chemical Formula: C₃₈H₄₆N₂O₁₆

Exact Mass: 786.2847 g/mol

To a solution of compound **85** (121 mg, 0.18 mmol) in anhydrous CH₂Cl₂ (3 mL), were added EDC·HCl (69 mg, 0.36 mmol) and HOBT (49 mg, 0.36 mmol) under argon at 0 °C. After being stirred for 20 min, GlyOtBu (28 mg, 0.22 mmol) was added. The resulting mixture was stirred at room temperature overnight. After the completion of the reaction, the solution was concentrated to a syrup which was purified by column chromatography over silica gel (petroleum ether/EtOAc: 3/2) to afford compound **86** (111 g, 79%) as a white powder.

R_f = 0.31 (petroleum ether/EtOAc: 1/1).

[α]_D = -9.3 (*c* 0.1, CHCl₃).

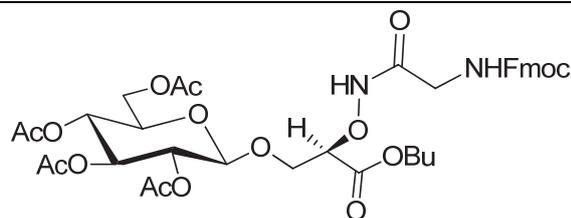
M.p. 76 °C.

¹H NMR (400 MHz, CDCl₃): δ 8.75 (s, 1H, NH), 8.32 (s, 1H, NH), 7.80-7.28 (m, 8H, Fmoc), 5.26 (t, *J* = 9.6 Hz, 1H, H-3'), 5.12-5.02 (m, 2H, H-2',4'), 4.57-4.46 (m, 4H, H-1',2, CH₂), 4.39 (dd, *J* = 11.0, 3.2 Hz, CH₂_{sn}a), 4.29-4.15 (m, 3H, H-6', CH), 3.94 (t, *J* = 5.3 Hz, 2H, CH₂), 3.82 (dd, *J* = 10.9, 9.0 Hz, 1H, CH₂_{sn}b), 3.73 (ddd, *J* = 10.0, 4.7, 2.3 Hz, 1H, H-5'), 2.09, 2.07, 2.04, 2.03 (4×s, 12H, 4×OAc), 1.45 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.9, 170.6, 170.2, 169.5, 168.4, 167.9, 158.0 (C=O); 143.4, 143.3, 141.4 (C_q); 128.0, 127.3, 127.2, 125.2, 125.1, 120.2 (CH, Fmoc); 101.2 (C-1'), 86.3 (CH), 82.2 (C_q), 72.3 (C-3'), 72.1 (C-5'), 71.7 (C-2'), 69.5 (CH₂), 68.3 (C-4'), 68.1, 61.8 (CH₂), 46.9 (CH), 41.8 (CH₂), 28.1, 21.0, 20.8, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+Na]⁺ Calcd for C₃₈H₄₆N₂NaO₁₆ 809.2745; Found 809.2747.

tert-Butyl (2*R*)-2-*N*-(9-fluorenylmethoxycarbonylamino)methylcarbonyl)aminooxy-3[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoate (**87**)



Chemical Formula: C₃₈H₄₆N₂O₁₆

Exact Mass: 786.2847 g/mol

To a solution of compound **82** (0.48 g, 0.94 mmol) in anhydrous CH₂Cl₂ (5 mL), were added EDC·HCl (0.28 g, 1.88 mmol) and HOBT (0.25 g, 1.88 mmol) under argon at 0 °C. After being stirred for 20 min, Fmoc-Gly-OH (0.28 g, 0.94 mmol) was added. The resulting mixture was stirred at room temperature overnight. After the completion of the reaction, the solution was concentrated to a syrup which was purified by column chromatography over silica gel (petroleum ether/EtOAc: 1/2) to afford compound **87** (0.47 g, 64%) as a white powder.

R_f = 0.50 (petroleum ether/EtOAc: 1/3).

[α]_D = +7.7 (*c* 0.1, CHCl₃).

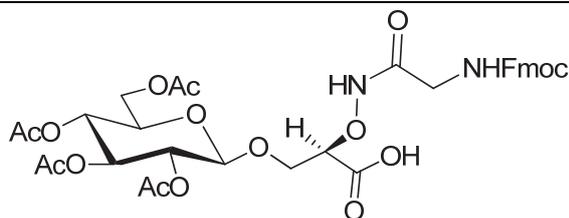
M.p. 77 °C.

¹H NMR (400 MHz, CDCl₃): δ 9.50 and 8.73 (2×s, 1H, NH), 7.84-7.28 (m, 8H, Fmoc), 5.63 (s, 1H, NH-Fmoc) 5.22 (t, *J* = 9.6 Hz, 1H, H-3'), 5.17-4.98 (m, 2H, H-2',4'), 4.65 (d, *J* = 6.0 Hz, 1H, H-1') 4.55-4.48 (m, 1H, H-2), 4.40 (d, *J* = 6.8 Hz, CH₂), 4.34-4.11 (m, 5H, H-6', CH₂Fmoc, CH_{Fmoc}), 3.96-3.87 (m, 2H, CH₂), 3.77-3.69 (m, 1H, H-5'), 2.08, 2.05, 2.02, 1.99 (4×s, 12H, 4×OAc), 1.48 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 170.8, 170.3, 169.9, 169.8, 169.5, 167.2, 156.6 (C=O); 143.9, 141.4 (C_q); 127.8, 127.2, 125.2, 120.1 (CH, Fmoc); 100.6 (C-1'), 83.6 (CH), 77.4 (C_q), 72.7 (C-3'), 72.0 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 67.8, 67.4, 61.8 (CH₂), 47.2 (CH), 42.6 (CH₂), 28.1, 20.8, 20.7 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₃₈H₄₆KN₂O₁₆ 825.2484; Found 825.2486.

(2*R*)-2-*N*-(9-Fluorenylmethoxycarbonylamino)methylcarbonyl)aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoic acid (88**)**



Chemical Formula: C₃₄H₃₈N₂O₁₆

Exact Mass: 730.2221 g/mol

To a solution of compound **87** (246 mg, 0.3 mmol) in CH₂Cl₂ (4 mL), was added TFA (0.4 mL). The resulting mixture was stirred at room temperature overnight. After the reaction finished, most of the solvent was evaporated, and the residue was dried under high vacuum (oil pump) until no acidic smell could be detected. The residue was purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 25/1) to give compound **88** (175 mg, 77%) as a white powder.

R_f = 0.35 (CH₂Cl₂/MeOH: 20/1).

[α]_D = +4.0 (*c* 0.1, CHCl₃).

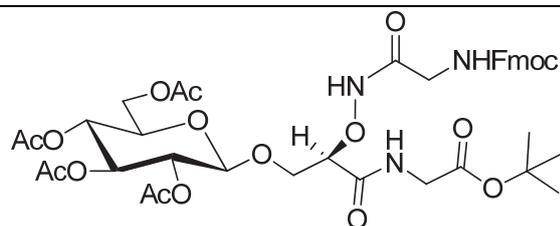
M.p. 166 °C.

¹H NMR (400 MHz, CDCl₃): δ 10.5 (s, 1H, NH), 8.80 (broad s, 1H, COOH), 7.81-7.20 (m, 8H, Fmoc), 5.63 (s, 1H, NH-Fmoc), 5.23 (t, *J* = 9.6 Hz, 1H, H-3'), 5.12-4.90 (m, 2H, H-2',4'), 4.63-4.53 (m, 1H, H-2), 4.49 (d, *J* = 6.4 Hz, 1H, H-1'), 4.46-4.30 (m, 3H, H-3a, CH₂), 4.28-4.08 (m, 3H, H-6', CH_{Fmoc}), 3.99-3.89 (m, 2H, CH_{2Fmoc}), 3.89-3.78 (m, 1H, H-3b), 3.73-3.65 (m, 1H, H-5'), 2.12-1.91 (m, 12H, 4×OAc).

¹³C NMR (100 MHz, CDCl₃): δ 171.5, 171.0, 170.2, 169.8, 169.6, 157.0 (C=O); 143.7, 141.3, (C_q); 127.9, 127.2, 125.2, 120.1 (CH, Fmoc); 101.2 (C-1'), 85.5 (CH), 72.0 (C-3',5'), 71.6 (C-2'), 68.9 (CH₂), 68.3 (C-4'), 67.6, 61.8 (CH₂); 47.0 (CH), 42.0 (CH₂), 20.9, 20.8, 20.7, 20.6 (CH₃).

HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₃₄H₃₈N₂NaO₁₆ 753.2119; Found 753.2115.

***N*-{(2*R*)-2-*N*-(9-Fluorenylmethoxycarbonylamino)methylcarbonyl)aminoxy-3-[(2',3',4',6'-tetra-*O*-acetyl- β -D-glucopyranosyl)oxy]propanoyl}-glycine *tert*-butyl ester (**89**)**



Chemical Formula: C₄₀H₄₉N₃O₁₇

Exact Mass: 843.3062 g/mol

To a solution of compound **88** (183 mg, 0.25 mmol) in anhydrous CH₂Cl₂ (5 mL), were added EDC·HCl (96 mg, 0.5 mmol) and HOBT (68 mg, 0.5 mmol) under argon at 0 °C. After being stirred for 20 min, GlyOtBu (42 mg, 0.25 mmol) was added. The resulting mixture was stirred at room temperature overnight. After the completion of the reaction, the solution was concentrated to a syrup which was purified by column chromatography over silica gel (CH₂Cl₂/MeOH: 100/3) to afford compound **89** (100 g, 48%) as a white powder.

R_f = 0.25 (petroleum ether/EtOAc: 1/3).

[α]_D = -9.0 (*c* 0.1, CHCl₃).

M.p. 90 °C.

¹H NMR (400 MHz, CDCl₃): δ 9.99 (s, 1H, NH), 8.78 (s, 1H, NH), 7.84-7.29 (m, 8H, Fmoc), 5.57 (t, *J* = 5.1 Hz, 1H, NH-Fmoc), 5.27 (t, *J* = 9.6 Hz, 1H, H-3'), 5.13-4.97 (m, 2H, H-2', 4'), 4.60-4.38 (m, 5H, H-1', 2 \times CH, CH₂Fmoc), 4.30-4.15 (m, 3H, H-6', CH_{Fmoc}), 4.02-3.90 (m, 4H, 2 \times CH₂), 3.84-3.76 (m, 1H, CH), 3.75-3.69 (m, 1H, H-5'), 2.10, 2.07, 2.04, 1.97 (4 \times s, 12H, 4 \times OAc), 1.47 (s, 9H, *t*-Bu).

¹³C NMR (100 MHz, CDCl₃): δ 171.7, 171.0, 170.2, 169.6, 168.6, 168.0, 167.6, 156.5 (C=O); 143.8, 141.4 (C_q); 127.9, 127.2, 125.2, 120.1 (CH, Fmoc); 101.1 (C-1'), 86.2 (CH), 82.3 (C_q), 72.1, 72.0 (CH); 69.8 (CH₂), 68.3 (CH), 67.4, 61.7 (CH₂), 47.1 (CH), 42.1, 41.8 (CH₂), 28.1, 21.1, 20.9, 20.7 (CH₃).

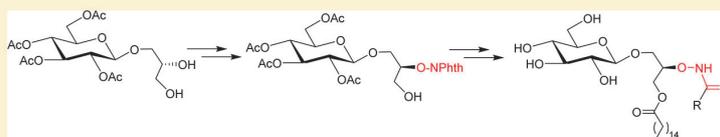
HRMS (ESI) *m/z*: [M+K]⁺ Calcd for C₄₀H₄₉N₃NaO₁₇ 866.2960; Found 866.2962.

Synthesis of *N*-Oxyamide-Linked Neoglycolipids

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S Supporting Information



ABSTRACT: *N*-Oxyamide-containing compounds have shown improved metabolic stability and interesting secondary structures due to the good hydrogen bond-donating property of *N*-oxyamide. β -Glucolipids linked by the *N*-oxyamide bond have been successfully synthesized as novel mimics of glycosphingolipids and glycosphingolipids.

As part of the glycoconjugate family, glycolipids are implicated in a variety of important biological phenomena such as cell–cell interactions, viral and bacterial infections, immune response, signal transduction, cell proliferation, etc. Glycolipids are composed of one or several monosaccharide residues bound by a glycosidic linkage to a hydrophobic moiety like an acylglycerol (termed glycosphingolipids, GGLs) or a ceramide (glycosphingolipids, GSLs). GGLs and mammalian GSLs begin with either glucose or galactose attached in α - or β -linkage to the 1-hydroxyl of mono/diacylglycerol or ceramide. Ceramides consist of fatty-amide-linked sphingosine (a long-chain amino alcohol). GGLs mainly exist in plants, algae, and bacteria. They have shown interesting anti-tumor-promoting activities.¹ Inhibitory effects on DNA polymerase, human Myt1-kinase, human lanosterol synthase, and antitumor activities have also been reported.² Bacterial glycosphingolipid BbGL2 has been shown to induce natural killer T cell (NKT cell) proliferation and cytokine production,³ an important property known for GSL agelashins.⁴ As a most abundant and diverse class of glycolipids in animals, GSLs are found in the plasma membrane of cells and play key roles in cell–cell interactions and protein activities.⁵ As immunostimulating agents,⁶ these glycolipids have attracted intensive research interest, in particular, since the discovery of a potent NKT immune cell activation effect of synthetic α -galactosyl ceramide (α -GalCer or KRN7000).⁷ The design of glycolipid mimics has become a useful strategy in drug discovery. Modifications have been made on the sugar part, on the configuration and nature of the anomeric bond (*C*-, *S*-glycoside), on the polar moiety of the ceramide or glycerol, or on the lipid chains.⁷ Triazole-containing glycolipids have also been reported, showing a comparable stimulatory effect on cytokine production as α -GalCer.⁸ The ability to undergo extensive interlipid hydrogen bonding has been considered to be fundamental for the functions of glycolipids in membranes, through imparting structural integrity to the membranes of the organisms.⁹ *N*-Oxyamide bonds in peptide¹⁰ and sugar derivatives¹¹ have been found to be good hydrogen-bond-donating groups due to lone pair repulsion between adjacent nitrogen and oxygen. They can

easily organize into turns and helices through intramolecular hydrogen bond formation. This unique property makes *N*-oxyamide linkage attractive for the modification of biomolecules. Furthermore, *N*-oxyamides are resistant to chemical and enzymatic hydrolysis.¹² With a continuing interest in *N*-oxyamide-containing biomolecules,¹³ we designed the *N*-oxyamide-linked glycolipids, by replacing the ester function in GGLs by a *N*-oxyamide (Figure 1). These kinds of glycolipids,

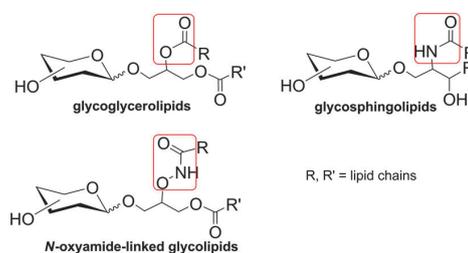


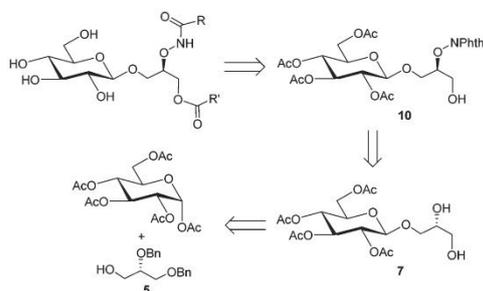
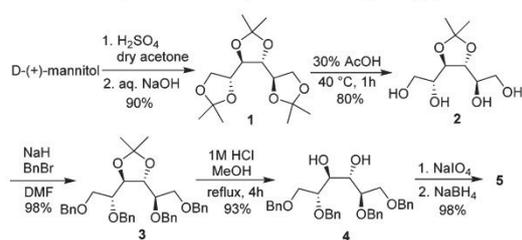
Figure 1. Structure of GGLs, GSLs, and *N*-oxyamide linked glycolipids.

never reported in the literature, could be considered as analogues of both GGL and GSL (Figure 1). Only one *N*(OMe)-glycosphingolipid has very recently been reported showing potent inhibitory activity against recombinant endoglycosphingolipidase II.¹⁴ Because the β -linked glucose unit widely exists in natural glycolipids, we investigated first the synthesis of *O*-amino β -glucoglycerol and its application to *N*-oxyamide-containing glycolipids.

The synthetic strategy toward *N*-oxyamide-containing glycolipids would be the stereoselective synthesis of *O*-amino β -glucoglycerol **10** through Mitsunobu reaction on glycosylglycerol **7**, which can be obtained via glycosylation with

Received: September 16, 2014

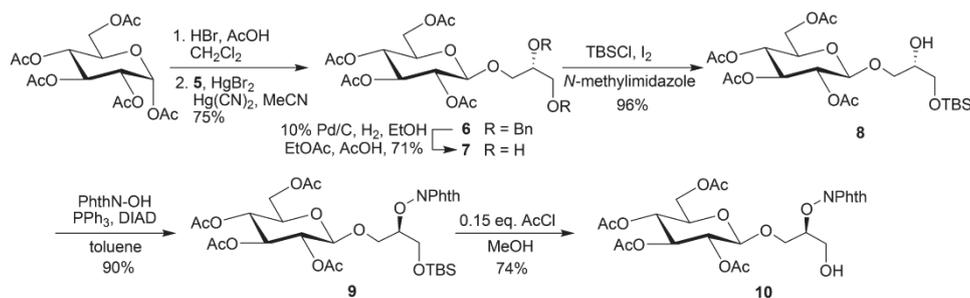
Published: October 15, 2014

Scheme 1. Retrosynthesis of *N*-Oxyamide-Linked GlycolipidsScheme 2. Preparation of 1,2-Di-*O*-benzyl-*sn*-glycerol 5

dibenzylated glycerol **5**, followed by introduction of lipid chains and final deprotection (Scheme 1).

The required glycerol **5**¹⁵ was prepared in 64% overall yield from *D*-(+)-mannitol, as outlined in Scheme 2. It is important to control the reaction conditions to convert triacetone **1**¹⁶ to 3,4-isopropylidene-*D*-mannitol **2**.¹⁵ Deprotection with 70% AcOH at 40 °C¹⁵ gave 50% yield of **2**. The best result (80%) was obtained using 30% AcOH¹⁷ at 40 °C during 1 h.

Glycosylation of glycerol **5** was realized with per-*O*-acetyl- α -*D*-glucopyranosyl bromide generated from *D*-glucose pentaacetate (Scheme 3). Dropwise addition of glycosyl bromide in dry acetonitrile into the solution containing compound **5**, HgBr₂, and Hg(CN)₂ is crucial to prepare β -glucoglycerol **6**¹⁵ in good yield (75% for two steps). After debenzylation, regioselective silylation of **7**¹⁸ required the presence of *N*-methylimidazole and iodine¹⁹ as promoter to ensure good reactivity and regioselectivity. The oxyamine function was then introduced via Mitsunobu reaction with PhthNOH to afford compound **9** with inversion of configuration.²⁰ Removal of the TBS group was achieved with catalytic AcCl in MeOH in 72% yield, instead of TBAF which deprotected acetyl groups, leading to a mixture of products.

Scheme 3. Synthesis of *O*-Amino β -Glucoglycerol **10**

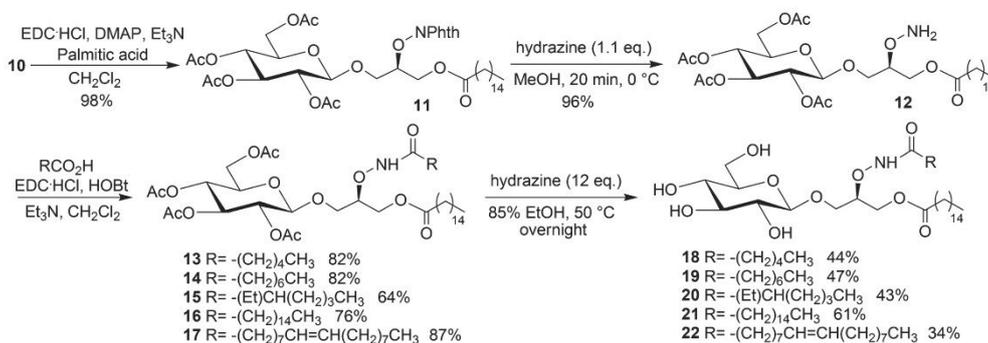
Next, esterification of the *O*-phthaloylamino β -glucoglycerol **10** with palmitic acid using 1-ethyl-3-(3-(dimethylamino)propyl)carbodiimide (EDC) furnished the glucolipid **11** in excellent yield (98%) (Scheme 4). Hydrazinolysis under mild conditions (1.1 equiv, 20 min at 0 °C) deprotected selectively the phthaloyl group. Coupling of the resulting oxyamine **12** with different fatty acids led successfully to the neoglycolipids **13–17** in 64–87% yield. The NH of the *N*-oxyamide bond appeared between 8.81 and 8.87 ppm on the ¹H NMR in CDCl₃. Final deacetylation was realized with hydrazine at 50 °C in EtOH to give the target compounds **18–22**. Due to their amphiphilic nature, some glycolipids are lost during the workup, leading to low yields (34–61%).

In conclusion, we have achieved the first synthesis of *N*-oxyamide-linked β -glucolipids from readily available *D*-glucose pentaacetate and *D*-mannitol via the *O*-amino β -glucoglycerol **10** as a versatile intermediate. Different lipid chains could be easily introduced on hydroxyl and oxyamine functions to access a variety of glycolipid structures. Synthesis of 1,2-di-*O*-benzyl-*sn*-glycerol **5** has also been optimized.

EXPERIMENTAL SECTION

3,4-O-Isopropylidene-*D*-mannitol (2). To a stirred solution of 30% aq AcOH (200 mL) was added *D*-mannitol triacetone **1** (10 g, 33.1 mmol). The mixture was stirred at 40 °C. After 1 h, the solution was evaporated and residual AcOH removed by repeated coevaporation with toluene. Dry acetone was added, and the mixture was thoroughly stirred with excess anhyd K₂CO₃ and then filtered. The insoluble material (remaining mannitol) was washed with acetone, and the combined filtrates were evaporated. The residue was then dissolved in a small volume of EtOAc and allowed to crystallize at rt to give pure product (5.89 g, 80.1%): *R*_f = 0.43 (CH₂Cl₂/MeOH 10/1); ¹H NMR (400 MHz, acetone-*d*₆) δ 4.73 (s, 2H, 2 \times OH), 3.96–3.90 (m, 2H, 2 \times CH), 3.77–3.54 (m, 8H, 2 \times CH, 2 \times CH₂, 2 \times OH), 1.32 (s, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, acetone-*d*₆) δ 109.8 (C_q), 80.8, 74.2 (CH); 64.6 (CH₂), 27.3 (CH₃).

1,2,5,6-Tetra-*O*-benzyl-3,4-*O*-isopropylidene-*D*-mannitol (3). NaH (60%, 6.36 g, 159 mmol) was added to a stirred solution of **2** (5.88 g, 26.49 mmol) in dry DMF (200 mL). The suspension was stirred for 0.5 h at 0 °C in an ice bath. BnBr (15.11 mL, 127 mmol) was then added, and the stirring continued at rt overnight. Ice was added to destroy the excess NaH, and the solution was then concentrated, diluted with saturated aq NH₄Cl, and the product extracted with EtOAc (200 mL). The extract was washed with water and brine, dried over MgSO₄, and evaporated. Purification by column chromatography (petroleum ether/EtOAc 15/1) afforded compound **3** as a colorless syrup (15.10 g, 98%): *R*_f = 0.31 (petroleum ether/EtOAc 10/1); ¹H NMR (400 MHz, CDCl₃) δ 7.41–7.17 (m, 20H, H-Ph), 4.73 (d, *J* = 11.8 Hz, 2H, 2 \times CH), 4.57 (d, *J* = 11.7 Hz, 2H, 2 \times CH), 4.48–4.46 (m, 4H, 2 \times CH₂), 4.24–4.15 (m, 2H, 2 \times CH), 3.81–3.55 (m, 6H, 2 \times CH₂, 2 \times CH), 1.35 (s, 6H, 2 \times CH₃); ¹³C

Scheme 4. Synthesis of *N*-Oxamide-Linked Neoglycolipids

NMR (100 MHz, CDCl_3) δ 138.6, 138.5 (C_q); 128.5, 128.4, 128.0, 127.7, 127.6 (CH-Ph); 109.9 (C_q); 79.4, 78.6 (CH); 73.4, 72.9, 70.7 (CH_2); 27.3 (CH_3).

1,2,5,6-Tetra-*O*-benzyl-*D*-mannitol (4). Compound 3 (15.10 g, 25.96 mmol) was treated with 1 M aq HCl/MeOH (1:9, v/v, 250 mL) under reflux for 4 h, when TLC showed a new single spot. Excess solid NaHCO_3 was added, and the solution evaporated to give an oil which was dissolved in EtOAc (300 mL), dried over MgSO_4 , and evaporated. Purification by column chromatography (petroleum ether/EtOAc 5/1) afforded compound 4 (13.08 g, 93%) as a colorless syrup: $R_f = 0.12$ (petroleum ether/EtOAc 5/1); ^1H NMR (400 MHz, CDCl_3) δ 7.34–7.26 (m, 20H, H-Ph), 4.72 (d, $J = 11.5$ Hz, 2H, 2 \times CH), 4.59 (d, $J = 11.5$ Hz, 2H, 2 \times CH), 4.54 (s, 4H, 2 \times CH_2), 3.96 (m, 2H, 2 \times CH), 3.80–3.65 (m, 6H, 2 \times CH_2 , 2 \times CH), 3.04 (s, 1H, OH), 3.02 (s, 1H, OH); ^{13}C NMR (100 MHz, CDCl_3) δ 138.3, 138.1 (C_q); 128.5, 128.1, 127.9, 127.8 (CH-Ph); 79.3 (CH); 73.6, 73.2, 70.3 (CH_2); 70.1 (CH).

1,2-Di-*O*-benzyl-*sn*-glycerol (5). To a solution of 4 (14.10 g, 26 mmol) in MeOH (250 mL) was added an aq solution of NaIO_4 (8.56 g, 40 mmol in 150 mL of water). The reaction mixture was stirred for 3 h at rt, after which time TLC analysis revealed the oxidation to be complete ($R_f = 0.52$, petroleum ether/EtOAc 3/1). The reaction mixture was diluted with MeOH (200 mL) and cooled. The precipitate was filtered off, and to the filtrate was added NaBH_4 (9.84 g, 260 mmol). After 1 h, the reaction mixture was treated with AcOH, and the solution evaporated to a small volume which was diluted in CH_2Cl_2 (300 mL), dried over MgSO_4 , and concentrated. Purification by column chromatography (petroleum ether/EtOAc 3/1) afforded compound 5 (13.87 g, 98%) as a colorless syrup: $R_f = 0.24$ (petroleum ether/EtOAc 3/1); ^1H NMR (400 MHz, CDCl_3) δ 7.38–7.26 (m, 10H, H-Ph), 4.74–4.53 (m, 4H, 2 \times CH_2), 3.79–3.57 (m, 5H, 2 \times CH_2 , CH), 2.05 (s, 1H, OH); ^{13}C NMR (100 MHz, CDCl_3) δ 138.4, 138.1 (C_q); 128.6, 128.6, 128.0, 127.9, 127.8 (CH-Ph); 78.1 (CH); 73.7, 72.3, 70.3, 63.0 (CH_2).

(2*R*)-1,2-Di-*O*-benzyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -*D*-glucopyranosyl)glycerol (6). To a solution of α -*D*-glucose pentaacetate (2 g, 5.12 mmol) in CH_2Cl_2 (15 mL) was added HBr (33% in AcOH, 7 mL, 41 mmol) at 0 °C. The mixture was stirred at rt under Ar for 6 h, and then the solution was neutralized with aq NaHCO_3 . The resulting 2,3,4,6-tetra-*O*-acetyl- α -*D*-glucopyranosyl bromide was extracted with CH_2Cl_2 (100 mL), dried over MgSO_4 , and evaporated to give a syrup. To a solution of 1,2-di-*O*-benzyl-*sn*-glycerol (5) (1.41 g, 5.2 mmol), HgBr_2 (0.94 g, 2.6 mmol), and $\text{Hg}(\text{CN})_2$ (0.66 g, 2.6 mmol) in dry MeCN (20 mL) was added dropwise over a period of 1 h a solution of glycosyl bromide in MeCN (10 mL). The reaction solution was stirred at rt overnight and then concentrated to an oil which was dissolved in CH_2Cl_2 (100 mL) and washed with saturated aq KBr (2 \times 30 mL) and water (30 mL). The dried (MgSO_4) organic layer was concentrated. Purification by column chromatography (petroleum ether/EtOAc 3/1) afforded compound 6 (2.30 g, 75%) as a colorless paste: $R_f = 0.39$ (petroleum ether/EtOAc 2/1); ^1H NMR (400 MHz, CDCl_3) δ 7.37–7.28 (m, 10H, H-Ph), 5.18 (t, $J = 9.4$ Hz, 1H, H-3'), 5.09 (t, $J = 9.7$ Hz, 1H, H-4'), 5.00 (dd, $J = 9.5, 8.2$ Hz, 1H, H-2'), 4.65 (s, 2H, CH_2), 4.57–4.50 (m, 3H, H-1', CH_2), 4.28–4.09

(m, 2H, H-6'), 3.99–3.69 (m, 3H, H-2,3), 3.65 (ddd, $J = 10.0, 4.5, 2.3$ Hz, 1H, H-5'), 3.59–3.57 (m, 2H, H-1), 2.07, 2.04, 2.03, 2.01 (4 \times s, 12H, 4 \times OAc); ^{13}C NMR (100 MHz, CDCl_3) δ 170.9, 170.5, 169.6, 169.5 (C=O); 138.5, 138.3 (C_q); 128.6, 128.5 (CH-Ph); 101.2 (C-1'), 76.8 (CH), 73.6 (CH_2), 72.9 (C-3'), 72.3 (CH_2), 71.9 (C-5'), 71.4 (C-2'), 69.8, 69.4 (CH_2); 68.5 (C-4'), 62.0 (C-6'), 20.9, 20.8 (OAc).

(2*R*)-1-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -*D*-glucopyranosyl)-glycerol (7). A mixture of 6 (12 g, 20 mmol) and 10% Pd/C (1.8 g) in EtOAc (100 mL) containing 5 mL each of EtOH and AcOH was vigorously shaken under H_2 at rt overnight. The catalyst was filtered off, and the filtrate evaporated. Purification by column chromatography ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ 25/1) afforded compound 7 (6 g, 71%) as a white solid: $R_f = 0.12$ ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ 25/1); ^1H NMR (400 MHz, CDCl_3) δ 5.22 (t, $J = 9.6$ Hz, 1H, H-3'), 5.11–5.04 (m, 1H, H-4'), 5.04–4.97 (m, 1H, H-2'), 4.55 (d, $J = 7.8$ Hz, 1H, H-1'), 4.25–4.16 (m, 2H, H-6'), 3.89–3.79 (m, 3H, H-2,3), 3.77–3.57 (m, 3H, H-1,5'), 2.32 (s, 2H, 2 \times OH), 2.11, 2.07, 2.04, 2.01 (4 \times s, 12H, 4 \times OAc); ^{13}C NMR (100 MHz, CDCl_3) δ 170.8, 170.3, 169.7, 169.5 (C=O); 101.4 (C-1'), 72.6 (C-3'), 72.0 (C-5'), 71.3 (C-2'), 70.5 (CH), 68.4 (C-4'), 63.4, 61.9 (CH_2); 20.8, 20.7 (OAc).

1-*O*-*tert*-Butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -*D*-glucopyranosyl)-*sn*-glycerol (8). To a solution of 7 (2 g, 4.74 mmol), *N*-methylimidazole (1.14 mL, 14.22 mmol), and iodine (3.61 g, 14.22 mmol) in anhyd THF (20 mL) under Ar at 0 °C was added TBSCl (0.86 g, 5.69 mmol). The reaction mixture was stirred for 20 min at 0 °C and then quenched with saturated aq $\text{Na}_2\text{S}_2\text{O}_3$ and diluted with EtOAc (50 mL). The aqueous layers were extracted with EtOAc (2 \times 50 mL). The combined organic layers were dried over MgSO_4 , filtered, concentrated, and purified by column chromatography over silica gel ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ 100/1) to afford 8 (2.43 g, 95.7%) as a yellowish paste: $R_f = 0.74$ ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ 100/3); $[\alpha]_D -9.7$ (c 0.1, CHCl_3); ^1H NMR (400 MHz, CDCl_3) δ 5.20 (dd, $J = 12.0, 6.9$ Hz, 1H, H-3'), 5.05 (t, $J = 9.7$ Hz, 1H, H-4'), 4.98 (dd, $J = 9.6, 8.1$ Hz, 1H, H-2'), 4.54 (d, $J = 7.9$ Hz, 1H, H-1'), 4.25–4.11 (m, 2H, H-6'), 3.82–3.68 (m, 4H, H-2,3,5'), 3.62–3.58 (m, 2H, H-1), 2.08, 2.04, 2.01, 1.99 (4 \times s, 12H, 4 \times OAc), 0.88 (s, 9H, *t*-Bu), 0.05 (s, 6H, Si(CH_3)₂); ^{13}C NMR (100 MHz, CDCl_3) δ 170.7, 170.3, 169.5, 169.4 (C=O); 101.4 (C-1'), 72.8 (C-3'), 71.9 (C-5'), 71.7 (CH_2), 71.4 (C-2'), 70.5 (CH), 68.5 (C-4'), 63.7, 62.0 (CH_2); 25.9, 20.8, 20.7 (CH_3); 18.3 (C_q , *t*-Bu), -5.4 (CH_3 , Si(CH_3)₂); HRMS (ESI) m/z [M + H]⁺ calcd for $\text{C}_{23}\text{H}_{41}\text{O}_{12}\text{Si}$ 537.2367; found 537.2362 (all HRMS spectra were recorded on a Q-TOF MaXis using standard conditions).

(2*R*)-2-*O*-Phthalimido-1-*O*-*tert*-butyldimethylsilyl-3-*O*-(2',3',4',6'-tetra-*O*-acetyl- β -*D*-glucopyranosyl)glycerol (9). To a solution of 8 (2.10 g, 3.91 mmol) in toluene (60 mL) were added Ph_3P (3.08 g, 11.7 mmol), PhthNOH (1.91 g, 11.7 mmol), and DIAD (2.30 mL, 11.7 mmol) at 0 °C. The resulting mixture was stirred at rt for 2 h under Ar and then extracted with EtOAc (3 \times 50 mL). The combined organic layers were washed with brine (50 mL), dried over MgSO_4 , filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc 3/1) to afford 9 (2.39 g, 90%) as a paste: $R_f = 0.53$ (petroleum ether/EtOAc 1/1); $[\alpha]_D -13.3$ (c 0.1,

CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 7.83–7.70 (m, 4H, Phth), 5.16 (t, *J* = 9.6 Hz, 1H, H-3'), 5.02 (t, *J* = 9.8 Hz, 1H, H-4'), 4.92 (dd, *J* = 9.4, 8.1 Hz, 1H, H-2'), 4.66 (d, *J* = 7.8 Hz, 1H, H-1'), 4.39–4.41 (m, 1H, H-2), 4.21 (dd, *J* = 12.3, 4.8 Hz, 1H, H-6'a), 4.15–4.06 (m, 2H, H-3a,6'b), 3.93 (dd, *J* = 11.7, 6.2 Hz, 1H, H-3b), 3.89 (d, *J* = 4.7 Hz, 2H, H-1), 3.69 (ddd, *J* = 10.0, 4.7, 2.2 Hz, 1H, H-5'), 2.06, 2.01, 1.99, 1.96 (4 × s, 12H, 4 × OAc), 0.77 (s, 9H, *t*-Bu), –0.02, –0.03 (2 × s, 6H, Si(CH₃)₂); ¹³C NMR (100 MHz, CDCl₃) δ 170.8, 170.4, 169.6, 169.6 (C=O), 163.7 (C=O, Phth); 134.5, 129.0 (C_q), 123.6 (Phth), 100.8 (C-1'), 87.0 (C-2), 73.0 (C-3'), 71.8 (C-5'), 71.2 (C-2'), 68.4 (C-4'), 68.0, 62.0, 61.9 (CH₂); 25.8, 20.8, 20.8, 20.7 (CH₃); 18.2 (C_q, *t*-Bu), –5.6 (CH₃, Si(CH₃)₂); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₃₁H₄₄NO₁₄Si 682.2531; found 682.2526.

2-O-Phthalimido-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-sn-glycerol (10). To a solution of **9** (1.0 g, 1.47 mmol) in MeOH (30 mL) was added AcCl (16 μL, 0.22 mmol) at 0 °C. The resulting mixture was stirred at rt for 2 h under Ar and then extracted with CH₂Cl₂ (3 × 30 mL). The combined organic layers were washed with brine, dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc 1/1) to give **10** (0.59 g, 72%) as a white powder. mp 72 °C; *R*_f = 0.23 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –17.3 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 7.87–7.74 (m, 4H, Phth), 5.22–5.15 (m, 1H, H-3'), 5.03 (t, *J* = 10.1 Hz, 1H, H-4'), 4.97 (m, 1H, H-2'), 4.64 (d, *J* = 7.8 Hz, 1H, H-1'), 4.32–4.26 (m, 1H, H-2), 4.22 (dd, *J* = 12.3, 4.9 Hz, 1H, H-6'a), 4.14–4.06 (m, 2H, H-3a,6'b), 4.01 (dd, *J* = 11.6, 6.3 Hz, 1H, H-3b), 3.83–3.66 (m, 3H, H-1,5'), 2.07, 2.04, 1.99, 1.97 (4 × s, 12H, 4 × OAc); ¹³C NMR (100 MHz, CDCl₃) δ 170.8, 170.3, 169.6, 169.5 (C=O), 164.6 (C=O, Phth); 135.0, 128.8 (C_q); 124.0 (Phth), 100.9 (C-1'), 88.1 (CH), 72.8 (C-3'), 71.9 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.8, 61.9, 60.4 (CH₂); 20.8, 20.7, 20.7 (OAc); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₂₈H₃₀NO₁₄ 568.1666; found 568.1661.

(2R)-1-O-Palmitoyl-2-O-phthalimido-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (11). To a solution of palmitic acid (0.26 g, 1.02 mmol) in anhyd CH₂Cl₂ (10 mL) were added DMAP (0.25 g, 2.04 mmol), EDC-HCl (0.39 g, 2.04 mmol), and Et₃N (0.28 mL, 2.04 mmol) under Ar at 0 °C. After the mixture was stirred for 20 min, compound **10** (0.58 g, 1.02 mmol) was added. The resulting mixture was stirred at rt overnight. The solution was diluted with EtOAc (30 mL), washed with aq HCl (1 N, 2 × 15 mL), saturated aq NaHCO₃ (2 × 15 mL), and brine (15 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc 2/1) to afford **11** (1.05 g, 98.6%) as a white solid: mp 65 °C; *R*_f = 0.54 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –11.0 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 7.87–7.74 (m, 4H, Phth), 5.20 (t, *J* = 9.5 Hz, 1H, H-3'), 5.06 (t, *J* = 9.7 Hz, 1H, H-4'), 4.96 (dd, *J* = 9.2, 7.8 Hz, 1H, H-2'), 4.68 (d, *J* = 8.2 Hz, 1H, H-1'), 4.62–4.53 (m, 1H, H-2), 4.44–4.31 (m, 2H, H-3), 4.25 (dd, *J* = 12.3, 4.8 Hz, 1H, H-6'a), 4.18–4.07 (m, 2H, H-1a,6'b), 4.01 (dd, *J* = 11.8, 5.8 Hz, 1H, H-1b), 3.73 (ddd, *J* = 9.9, 4.7, 2.4 Hz, 1H, H-5'), 2.38–2.24 (m, 2H, CH₂), 2.10, 2.05, 2.02, 1.99 (4 × s, 12H, 4 × OAc), 1.65–1.52 (m, 2H, CH₂), 1.35–1.20 (m, 24H, 12 × CH₂), 0.88 (t, *J* = 6.9 Hz, 3H, CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 173.5, 170.8, 170.4, 169.6, 169.5, 163.6 (C=O); 134.8, 128.9 (C_q); 123.8 (CH), 100.9 (C-1'), 84.5 (CH), 72.9 (C-3'), 72.0 (C-5'), 71.1 (C-2'), 68.5 (C-4'), 67.8, 62.4, 62.0, 34.1, 32.1, 29.8, 29.8, 29.6, 29.5, 29.4, 29.3, 24.9, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.3 (CH₃); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₄₁H₆₀NO₁₅ 806.3963; found 806.3957.

(2R)-2-O-Amino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (12). To a solution of **11** (0.38 g, 0.47 mmol) in MeOH (3 mL) was added N₂H₄·H₂O (25.2 μL, 0.52 mmol) at 0 °C. The resulting mixture was stirred at 0 °C for 20 min under Ar and then extracted with CH₂Cl₂ (3 × 20 mL). The combined organic layers were washed with brine (20 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc 3/2) to afford **12** (0.31 g, 96%) as a paste: *R*_f = 0.40 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –46.7 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 5.22 (t, *J* = 8.7 Hz, 1H, H-3'), 5.06 (t, *J* = 9.6 Hz, 1H, H-4'), 4.96 (dd, *J* = 9.6, 7.8 Hz, 1H, H-2'), 4.57 (d, *J* = 7.8 Hz, 1H, H-1'), 4.39–4.06 (m, 4H, H-3,6'), 4.04–3.90

(m, 2H, H-1a,2), 3.79–3.65 (m, 2H, H-1b,5'), 2.32 (t, *J* = 7.5 Hz, 2H, CH₂), 2.09, 2.07, 2.03, 2.01 (4 × s, 12H, 4 × OAc), 1.68–1.57 (m, 2H, CH₂), 1.39–1.20 (m, 24H, 12 × CH₂), 0.88 (t, *J* = 6.7 Hz, 3H, CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 173.9, 170.8, 170.4, 169.5 (C=O); 101.2 (C-1'), 80.1 (CH), 72.8 (C-3'), 72.0 (C-5'), 71.3 (C-2'), 68.5 (C-4'), 68.0, 62.0, 61.9, 34.3, 32.1, 29.8, 29.6, 29.5, 29.4, 29.3, 25.1, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.2 (CH₃); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₃₃H₅₈NO₁₃ 676.3908; found 676.3903.

General Procedure A for *N*-Oxamide Formation. Synthesis of (2R)-2-O-Acylamino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (13–17). To a solution of carboxylic acid (0.25 mmol) in anhyd CH₂Cl₂ (4 mL) were added HOBt (0.5 mmol), EDC-HCl (0.5 mmol), and Et₃N (0.5 mmol) under Ar at 0 °C. After being stirred for 20 min, the oxamide **12** (0.25 mmol) was added. The resulting mixture was stirred at rt overnight. The solution was diluted with EtOAc (25 mL), washed with aq HCl (1 N, 2 × 10 mL), saturated aq NaHCO₃ (2 × 10 mL), and brine (10 mL), dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (petroleum ether/EtOAc 2/1).

(2R)-2-O-Hexanoylamino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (13). From **12** (153 mg, 0.23 mmol) and hexanoic acid, compound **13** was obtained as a paste (145 mg, 82%); *R*_f = 0.60 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –35.7 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 8.83 (s, 1H, NH), 5.24 (t, *J* = 9.5 Hz, 1H, H-3'), 5.09 (t, *J* = 9.7 Hz, 1H, H-4'), 5.02 (dd, *J* = 9.6, 7.8 Hz, 1H, H-2'), 4.57 (d, *J* = 8.0 Hz, 1H, H-1'), 4.42 (dd, *J* = 12.2, 3.2 Hz, 1H, H-3a), 4.32–4.09 (m, 4H, H-2,3b,6'), 4.03–3.96 (m, 1H, H-1a), 3.81–3.68 (m, 2H, H-1b,5'), 2.39–2.33 (m, 2H, CH₂), 2.17–2.01 (m, 14H, 4 × OAc, CH₂), 1.73–1.57 (m, 4H, 2 × CH₂), 1.39–1.21 (m, 28H, 14 × CH₂), 0.96–0.86 (m, 6H, 2 × CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 174.4, 170.8, 170.3, 169.6 (C=O); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.5 (C-2'), 68.4 (C-4'), 67.9, 61.8, 61.4, 34.2, 33.2, 32.1, 31.5, 29.8, 29.8, 29.6, 29.5, 29.4, 29.2, 25.2, 25.0, 22.8, 22.5 (CH₂); 20.9, 20.7, 14.3, 14.1 (CH₃); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₃₉H₆₈NO₁₄ 774.4640; found 774.4634.

(2R)-2-O-Octanoylamino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (14). From **12** (151 mg, 0.22 mmol) and octanoic acid, compound **14** was obtained as a paste (147 mg, 82%); *R*_f = 0.62 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –49.3 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 8.81 (s, 1H, NH), 5.24 (t, *J* = 9.4 Hz, 1H, H-3'), 5.09 (t, *J* = 9.6 Hz, 1H, H-4'), 5.02 (t, *J* = 8.8 Hz, 1H, H-2'), 4.57 (d, *J* = 7.8 Hz, 1H, H-1'), 4.42 (d, *J* = 10.0 Hz, 1H, H-3a), 4.31–4.09 (m, 4H, H-2,3b,6'), 4.03–3.96 (m, 1H, H-1a), 3.79–3.68 (m, 2H, H-1b,5'), 2.36 (t, *J* = 7.2 Hz, 2H, CH₂), 2.16–2.00 (m, 14H, 4 × OAc, CH₂), 1.68–1.58 (m, 4H, 2 × CH₂), 1.40–1.20 (m, 32H, 16 × CH₂), 0.93–0.84 (m, 6H, 2 × CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 174.4, 170.8, 170.4, 170.3, 169.6 (C=O); 100.7 (C-1'), 82.5 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.5 (C-2'), 68.4 (C-4'), 67.9, 61.8, 61.4, 34.2, 33.2, 32.1, 31.8, 29.8, 29.6, 29.5, 29.4, 29.2, 29.1, 25.5, 25.0, 22.8, 22.7 (CH₂); 20.9, 20.7, 14.3 (CH₃); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₄₁H₇₂NO₁₄ 802.4953; found 802.4947.

(2R)-2-O-(2-Ethylhexanoyl)amino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)glycerol (15). From **12** (151 mg, 0.22 mmol) and 2-ethylhexanoic acid, compound **15** was obtained as a paste (114 mg, 64%); *R*_f = 0.47 (petroleum ether/EtOAc 1/1); [α]_D²⁰ –38.0 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 8.87 (s, 1H, NH), 5.25 (t, *J* = 9.4 Hz, 1H, H-3'), 5.09 (t, *J* = 9.7 Hz, 1H, H-4'), 5.01 (t, *J* = 8.7 Hz, 1H, H-2'), 4.57 (d, *J* = 7.7 Hz, 1H, H-1'), 4.47–4.39 (m, 1H, H-3a), 4.31–4.10 (m, 4H, H-2,3b,6'), 4.03–3.96 (m, 1H, H-1a), 3.78–3.69 (m, 2H, H-1b,5'), 2.36 (t, *J* = 7.2 Hz, 2H, CH₂), 2.10, 2.09, 2.04, 2.02 (4 × s, 12H, 4 × OAc), 1.95–1.85 (m, 1H, CH), 1.69–1.57 (m, 4H, 2 × CH₂), 1.52–1.39 (m, 2H, CH₂), 1.37–1.20 (m, 28H, 14 × CH₂), 0.95–0.84 (m, 9H, 3 × CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 174.4, 173.7, 170.8, 170.4, 170.2, 169.6 (C=O); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.0 (C-5'), 71.5 (C-2'), 68.3 (C-4'), 67.9, 61.8, 61.3 (CH₂); 45.7 (CH), 34.3, 34.2, 32.2, 32.0, 29.8, 29.6, 29.5, 29.4, 29.2, 25.9, 24.9, 22.8 (CH₂); 20.9, 20.8, 20.7, 14.2, 14.1, 12.1 (CH₃); HRMS (ESI) *m/z* [M + H]⁺ calcd for C₄₁H₇₂NO₁₄ 802.4953; found 802.4947.

(2R)-1-O-Palmitoyl-2-O-palmitoylamino-3-O-(2',3',4',6'-tetra-O-acetyl- β -D-glucopyranosyl)glycerol (16). From 12 (189 mg, 0.28 mmol) and palmitic acid, compound 15 was obtained as a white solid (192 mg, 76%): mp 74 °C; R_f = 0.57 (petroleum ether/EtOAc 1/1); $[\alpha]_D^{25}$ -42.3 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 8.86 (s, 1H, NH), 5.24 (t, J = 9.5 Hz, 1H, H-3'), 5.09 (t, J = 9.7 Hz, 1H, H-4'), 5.02 (t, J = 9.6, 7.8 Hz, 1H, H-2'), 4.57 (d, J = 7.9 Hz, 1H, H-1'), 4.42 (dd, J = 12.2, 3.3 Hz, 1H, H-3a), 4.32–4.08 (m, 4H, H-2,3b,6'), 4.03–3.96 (m, 1H, H-1a), 3.78–3.69 (m, 2H, H-1b,5'), 2.36 (t, J = 7.5 Hz, 2H, CH₂), 2.18–2.01 (m, 14H, 4 \times OAc, CH₂), 1.68–1.58 (m, 4H, 2 \times CH₂), 1.40–1.22 (m, 48H, 24 \times CH₂), 0.93–0.81 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 174.4, 171.2, 170.8, 170.2, 169.5 (C=O); 100.7 (C-1'), 82.1 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.4 (C-2'), 68.4 (C-4'), 67.8, 61.8, 61.3, 34.2, 33.3, 32.0, 29.8, 29.6, 29.5, 29.4, 29.2, 25.5, 24.9, 22.8 (CH₂); 20.8, 20.7, 14.2 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₃₉H₈₈NO₁₄ 914.6205; found 914.6199.

(2R)-2-O-Oleoylamino-1-O-palmitoyl-3-O-(2',3',4',6'-tetra-O-acetyl- β -D-glucopyranosyl)glycerol (17). From 12 (151 mg, 0.22 mmol) and oleic acid, compound 17 was obtained as a white solid (206 mg, 87%): mp 57 °C; R_f = 0.52 (petroleum ether/EtOAc 1/1); $[\alpha]_D^{25}$ -35.3 (c 0.1, CHCl₃); ¹H NMR (400 MHz, CDCl₃) δ 8.83 (s, 1H, NH), 5.43–5.32 (m, 2H, CH=CH), 5.24 (t, J = 9.5 Hz, 1H, H-3'), 5.09 (t, J = 9.7 Hz, 1H, H-4'), 5.02 (dd, J = 9.6, 8.0 Hz, 1H, H-2'), 4.57 (d, J = 7.9 Hz, 1H, H-1'), 4.42 (dd, J = 12.2, 3.3 Hz, 1H, H-3a), 4.32–4.08 (m, 4H, H-2,3b,6'), 4.03–3.96 (m, 1H, H-1a), 3.78–3.69 (m, 2H, H-1b,5'), 2.36 (t, J = 7.5 Hz, 2H, CH₂), 2.20–1.98 (m, 16H, 4 \times OAc, 2 \times CH₂), 1.71–1.57 (m, 4H, 2 \times CH₂), 1.39–1.21 (m, 46H, 23 \times CH₂), 0.95–0.82 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CDCl₃) δ 174.5, 170.8, 170.4, 170.3, 169.6 (C=O); 130.1, 129.8 (CH=CH); 100.7 (C-1'), 82.2 (CH), 72.5 (C-3'), 72.1 (C-5'), 71.4 (C-2'), 68.4 (C-4'), 67.8, 61.8, 61.3, 34.2, 33.3, 32.0, 29.9, 29.8, 29.6, 29.5, 29.4, 29.2, 27.3, 25.5, 25.0, 22.8 (CH₂); 20.8, 20.7, 14.3 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₅₁H₉₀NO₁₄ 940.6361; found 940.6356.

General Procedure B for Deacetylation. Synthesis of (2R)-2-O-acylamino-3-O- β -D-glucopyranosyl-1-O-palmitoylglycerol (18–22). To a solution of acetylated compound (0.1 mmol) in 85% EtOH (5 mL) was added N₂H₄·H₂O (1.2 mmol). The mixture was stirred at 50 °C overnight. The solution was poured into ice-cold brine and extracted with CHCl₃ (3 \times 20 mL). The combined CHCl₃ layers were dried over MgSO₄, filtered, evaporated, and purified by column chromatography over silica gel (CH₂Cl₂/MeOH 30/1 to 20/1).

(2R)-3-O- β -D-Glucopyranosyl-2-O-hexanoylamino-1-O-palmitoylglycerol (18). Deacetylation of 13 (105 mg, 0.14 mmol) led to 18 as a white solid (30 mg, 44% yield): mp 94 °C; R_f = 0.43 (CH₂Cl₂/MeOH 8/1); $[\alpha]_D^{25}$ -32.0 (c 0.1, MeOH); ¹H NMR (400 MHz, CD₃OD) δ 4.35–4.20 (m, 3H, H-1',3'), 4.19–4.12 (m, 1H, H-2), 4.04 (dd, J = 11.4, 3.9 Hz, 1H, H-1a), 3.86 (d, J = 11.7 Hz, 1H, H-6'a), 3.75 (dd, J = 11.4, 6.1 Hz, 1H, H-1b), 3.67–3.61 (m, 1H, H-6'b), 3.40–3.24 (m, 3H, H-3',4',5'), 3.22–3.15 (m, 1H, H-2'), 2.34 (t, J = 7.4 Hz, 2H, CH₂), 2.07 (t, J = 7.4 Hz, 2H, CH₂), 1.70–1.55 (m, 4H, 2 \times CH₂), 1.45–1.22 (m, 28H, 14 \times CH₂), 0.97–0.85 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CD₃OD) δ 175.2, 173.2 (C=O); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.3, 62.7, 34.9, 33.7, 33.1, 32.4, 30.8, 30.6, 30.5, 30.2, 26.3, 25.9, 23.7, 23.4 (CH₂); 14.5, 14.3 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₃₁H₆₀NO₁₀ 606.4217; found 606.4212.

(2R)-3-O- β -D-Glucopyranosyl-2-O-octanoylamino-1-O-palmitoylglycerol (19). Deacetylation of 14 (95 mg, 0.12 mmol) led to 19 as a white solid (35 mg, 47% yield): mp 96 °C; R_f = 0.26 (CH₂Cl₂/MeOH 10/1); $[\alpha]_D^{25}$ -21.7 (c 0.1, MeOH); ¹H NMR (400 MHz, CD₃OD) δ 4.37–4.19 (m, 3H, H-1',3'), 4.19–4.12 (m, 1H, H-2), 4.04 (dd, J = 11.4, 3.8 Hz, 1H, H-1a), 3.86 (d, J = 11.6 Hz, 1H, H-6'a), 3.75 (dd, J = 11.4, 6.2 Hz, 1H, H-1b), 3.63 (dd, J = 9.8, 7.2 Hz, 1H, H-6'b), 3.40–3.24 (m, 3H, H-3',4',5'), 3.22–3.16 (m, 1H, H-2'), 2.34 (t, J = 7.4 Hz, 2H, CH₂), 2.07 (t, J = 7.4 Hz, 2H, CH₂), 1.69–1.54 (m, 4H, 2 \times CH₂), 1.48–1.22 (m, 32H, 16 \times CH₂), 1.01–0.81 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CD₃OD) δ 175.2, 173.2 (C=O); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.3, 62.7,

34.9, 33.8, 33.1, 32.9, 30.8, 30.6, 30.5, 30.2, 30.2, 26.6, 25.9, 23.8, 23.7 (CH₂); 14.5 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₃₃H₆₄NO₁₀ 634.4530; found 634.4525.

(2R)-2-O-(2-Ethylhexanoylamino-3-O- β -D-glucopyranosyl-1-O-palmitoylglycerol (20). Deacetylation of 15 (85 mg, 0.106 mmol) led to 20 as a paste (29 mg, 43% yield): R_f = 0.39 (CH₂Cl₂/MeOH 10/1); $[\alpha]_D^{25}$ -29.7 (c 0.1, MeOH); ¹H NMR (400 MHz, CD₃OD) δ 4.37–4.21 (m, 3H, H-1',3'), 4.20–4.12 (m, 1H, H-2), 4.05 (dd, J = 11.1, 3.9 Hz, 1H, H-1a), 3.85 (d, J = 11.7 Hz, 1H, H-6'a), 3.75 (dd, J = 11.3, 6.2 Hz, 1H, H-1b), 3.71–3.60 (m, 1H, H-6'b), 3.44–3.24 (m, 3H, H-3',4',5'), 3.22–3.15 (m, 1H, H-2'), 2.34 (t, J = 7.3 Hz, 2H, CH₂), 2.03–1.90 (m, 1H, CH), 1.70–1.50 (m, 4H, 2 \times CH₂), 1.49–1.17 (m, 30H, 15 \times CH₂), 1.06–0.81 (m, 9H, 3 \times CH₃); ¹³C NMR (100 MHz, CD₃OD) δ 175.7, 175.2 (C=O); 104.9 (C-1'), 83.9, 78.0, 77.9, 75.0, 71.5 (CH); 69.0, 63.2, 62.7 (CH₂); 46.6 (CH), 34.8, 33.3, 33.1, 30.8, 30.6, 30.5, 30.2, 26.9, 25.9, 23.7, 23.7 (CH₂); 14.5, 14.4, 12.4 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₃₃H₆₄NO₁₀ 634.4530; found 634.4525.

(2R)-3-O- β -D-Glucopyranosyl-1-O-palmitoyl-2-O-palmitoylamino-glycerol (21). Deacetylation of 16 (62 mg, 0.068 mmol) led to 21 as a white solid (31 mg, 61% yield): mp 177 °C; R_f = 0.24 (CH₂Cl₂/MeOH 20/1); $[\alpha]_D^{25}$ -14.0 (c 0.1, MeOH); ¹H NMR (400 MHz, CD₃OD) δ 4.39–4.22 (m, 3H, H-1',3'), 4.22–4.16 (m, 1H, H-2), 4.08 (dd, J = 11.6, 3.5 Hz, 1H, H-1a), 3.90 (d, J = 12.0 Hz, 1H, H-6'a), 3.78 (dd, J = 11.6, 6.3 Hz, 1H, H-1b), 3.69 (dd, J = 11.8, 5.2 Hz, 1H, H-6'b), 3.43–3.23 (m, 4H, H-2',3',4',5'), 2.37 (t, J = 7.5 Hz, 2H, CH₂), 2.10 (t, J = 7.4 Hz, 2H, CH₂), 1.69–1.58 (m, 4H, 2 \times CH₂), 1.49–1.10 (m, 48H, 24 \times CH₂), 0.96–0.86 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CD₃OD) δ 175.1, 172.9 (C=O); 104.6 (C-1'), 83.4, 77.5, 74.6, 71.2 (CH); 68.9, 63.0, 62.5, 34.7, 33.6, 32.8, 30.5, 30.3, 30.2, 30.0, 26.3, 25.6, 23.4 (CH₂); 14.4 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₄₁H₈₀NO₁₀ 746.5782; found 746.5777.

(2R)-3-O- β -D-Glucopyranosyl-2-O-oleoylamino-1-O-palmitoylglycerol (22). Deacetylation of 17 (125 mg, 0.133 mmol) led to 22 as a white solid (35 mg, 34% yield): mp 162 °C; R_f = 0.32 (CH₂Cl₂/MeOH 10/1); $[\alpha]_D^{25}$ -18.0 (c 0.1, MeOH); ¹H NMR (400 MHz, CD₃OD) δ 5.32 (s, 2H, CH=CH), 4.37–4.20 (m, 3H, H-1',3'), 4.19–4.11 (m, 1H, H-2), 4.04 (d, J = 10.0 Hz, 1H, H-1a), 3.85 (d, J = 11.3 Hz, 1H, H-6'a), 3.79–3.71 (m, 1H, H-1b), 3.68–3.58 (m, 1H, H-6'b), 3.40–3.22 (m, 3H, H-3',4',5'), 3.19 (t, J = 7.9 Hz, 1H, H-2'), 2.40–2.28 (m, 2H, CH₂), 2.14–1.94 (m, 4H, 2 \times CH₂), 1.67–1.52 (m, 4H, 2 \times CH₂), 1.48–1.10 (m, 46H, 23 \times CH₂), 0.97–0.80 (m, 6H, 2 \times CH₃); ¹³C NMR (100 MHz, CD₃OD) δ 175.2, 173.2 (C=O); 130.9, 130.8 (CH=CH); 104.9 (C-1'), 83.7, 78.0, 77.9, 75.0, 71.6 (CH); 69.0, 63.4, 62.7, 34.9, 33.8, 33.1, 30.8, 30.7, 30.5, 30.4, 30.2, 28.2, 26.6, 26.0, 23.8 (CH₂); 14.5 (CH₃); HRMS (ESI) m/z [M + H]⁺ calcd for C₄₃H₈₂NO₁₀ 772.5939; found 772.5933.

■ ASSOCIATED CONTENT

Supporting Information

Copies of ¹H, ¹³C, Dept-135, COSY, and HMQC NMR spectra of all described compounds. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

N.C. gratefully acknowledges China Scholarship Council (CSC) for a doctoral scholarship.

REFERENCES

- (1) Shirahashi, H.; Murakami, N.; Watanabe, M.; Nagatsu, A.; Sakakibara, J.; Tokuda, H.; Nishino, H.; Iwashima, A. *Chem. Pharm. Bull.* **1993**, *41*, 1664–1666.
- (2) (a) Murakami, C.; Kumagai, T.; Hada, T.; Kanekazu, U.; Nakazawa, S.; Kamisuki, S.; Maeda, N.; Xu, X.; Yoshida, H.; Sugawara, F.; Sakaguchi, K.; Mizushina, Y. *Biochem. Pharmacol.* **2003**, *65*, 259–267. (b) Maeda, N.; Hada, T.; Murakami-Nakai, C.; Kuriyama, I.; Ichikawa, H.; Fukumori, Y.; Hiratsuka, J.; Yoshida, H.; Sakaguchi, K.; Mizushina, Y. *J. Nutr. Biochem.* **2005**, *16*, 121–128. (c) Maeda, N.; Matsubara, K.; Yoshida, H.; Mizushina, Y. *Mini-Rev. Med. Chem.* **2011**, *11*, 32–38. (d) Colombo, D.; Scala, A.; Taino, I. M.; Toma, L.; Ronchetti, F.; Tokuda, H.; Nishino, H.; Nagatsu, A.; Sakakibara, J. *Cancer Lett.* **1998**, *123*, 83–86. (e) Sun, Y.; Zhang, J.; Li, C.; Guan, H.; Yu, G. *Carbohydr. Res.* **2012**, *355*, 6–12. (f) Tanaka, R.; Sakano, Y.; Nagatsu, A.; Shibuya, M.; Ebizuka, Y.; Goda, Y. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 159–162.
- (3) Kinjo, Y.; Tupin, E.; Wu, D.; Fujio, M.; Garcia-Navarro, R.; Benhnia, M. R.; Zajonc, D. M.; Ben-Menachem, G.; Ainge, G. D.; Painter, G. F.; Khurana, A.; Hoebe, K.; Behar, S. M.; Beutler, B.; Wilson, I. A.; Tsuji, M.; Sellati, T. J.; Wong, C.-H.; Kronenberg, M. *Nat. Immunol.* **2006**, *7*, 978–986.
- (4) (a) Natori, T.; Morita, M.; Akimoto, K.; Koezuka, Y. *Tetrahedron* **1994**, *50*, 2771–2784. (b) Morita, M.; Motoki, K.; Akimoto, K.; Natori, T.; Sakai, T.; Sawa, E.; Yamaji, K.; Koezuka, Y.; Kobayashi, E.; Fukushima, H. *J. Med. Chem.* **1995**, *38*, 2176–2187.
- (5) (a) Schnaar, R. L.; Suzuki, A.; Stanley, P. In *Essentials of Glycobiology*, 2nd ed.; Varki, A., Cummings, R. D., Esko, J. D., Freeze, H. H., Stanley, P., Bertozzi, C. R., Hart, G. W., Etzler, M. E., Eds.; Cold Spring Harbor Laboratory Press: Cold Spring Harbor, NY, 2009; Chapter 10. (b) Schnaar, R. L. *Arch. Biochem. Biophys.* **2004**, *426*, 163–172.
- (6) (a) Long, X.; Deng, S.; Mattner, J.; Zang, Z.; Zhou, D.; McNary, N.; Goff, R. D.; Teyton, L.; Bendelac, A.; Savage, P. B. *Nat. Chem. Biol.* **2007**, *3*, 559–564. (b) Wu, D.; Fujio, M.; Wong, C.-H. *Bioorg. Med. Chem.* **2008**, *16*, 1073–1083.
- (7) Banchet-Cadeddu, A.; Hénon, E.; Dauchez, M.; Renault, J.-H.; Monneaux, F.; Haudrechy, A. *Org. Biomol. Chem.* **2011**, *9*, 3080–3104 and references therein.
- (8) Lee, T.; Cho, M.; Ko, S.-Y.; Youn, H.-J.; Baek, D. J.; Cho, W.-J.; Kang, C.-Y.; Kim, S. *J. Med. Chem.* **2007**, *50*, 585–589.
- (9) Malhotra, R. *Biochem. Anal. Biochem.* **2012**, *1*, 108.
- (10) Li, X.; Wu, Y.-D.; Yang, D. *Acc. Chem. Res.* **2008**, *41*, 1428–1438.
- (11) Chandrasekhar, S.; Rao, C. L.; Reddy, M. S.; Sharma, G. D.; Kiran, M. U.; Naresh, P.; Chaitanya, G. K.; Bhanuprakash, K.; Jagadeesh, B. *J. Org. Chem.* **2008**, *73*, 9443–9446.
- (12) Chen, F.; Ma, B.; Yang, Z.-C.; Lin, G.; Yang, D. *Amino Acids* **2012**, *43*, 499–503.
- (13) (a) Malapelle, A.; Ramozzi, R.; Xie, J. *Synthesis* **2009**, 888–890. (b) Gong, Y. C.; Sun, H. B.; Xie, J. *Eur. J. Org. Chem.* **2009**, 6027–6033. (c) Gong, Y.; Peyrat, S.; Sun, H.; Xie, J. *Tetrahedron* **2011**, *67*, 7114–7120. (d) Song, Z.; He, X.-P.; Chen, G.-R.; Xie, J. *Synthesis* **2011**, 2761–2766. (e) Peyrat, S.; Xie, J. *Synthesis* **2012**, *44*, 1718–1724. (f) Noel, O.; Xie, J. *Synthesis* **2013**, *45*, 134–140. (g) Peyrat, S.; Cheng, K.; Xie, J. *Synthesis* **2013**, *45*, 2737–2744. (h) Zhang, H.-L.; Zang, Y.; Xie, J.; Chen, G.-R.; He, X.-P.; Tian, H. *Sci. Rep.* **2014**, *4*, 5513.
- (14) Ishida, J.; Hinou, H.; Naruchi, K.; Nishimura, S.-I. *Bioorg. Med. Chem. Lett.* **2014**, *24*, 1197–1200.
- (15) van Boeckel, C. A. A.; Visser, G. M.; van Boom, J. H. *Tetrahedron* **1985**, *41*, 4557–4565.
- (16) Yadav, V. K.; Agrawal, D. *Chem. Commun.* **2007**, 5232–5234.
- (17) Mannock, D. A.; Lewis, R. N.; McElhaney, R. N. *Chem. Phys. Lipids* **1987**, *43*, 113–127.
- (18) Janwitayanuchit, W.; Suwanborirux, K.; Patarapanich, C.; Pummangura, S.; Lipipun, V.; Vilaivan, T. *Phytochemistry* **2003**, *64*, 1253–1264.
- (19) Bartoszewicz, A.; Kalek, M.; Stawinski, J. *Tetrahedron* **2008**, *64*, 8843–8850.
- (20) (a) Schenk, S.; Weston, J.; Anders, E. *J. Am. Chem. Soc.* **2005**, *127*, 12566–12576. (b) But, T. Y. S.; Toy, P. H. *Chem.—Asian J.* **2007**, *2*, 1340–1355.

SUMMARY

As part of glycoconjugate family, glycolipids and glycopeptides are involved in a variety of important biological, physiological and pathological processes, such as cell-cell interactions, viral and bacterial infections, immune response, cancer progression, etc. Synthesis of glycoconjugate mimics has attracted intensive research interest for biological and pharmaceutical applications. This thesis was devoted to the synthesis of *N*-oxyamide-linked glycolipids and glycopeptides, since *N*-oxyamide-containing compounds have shown improved metabolic stability, and interesting secondary structures due to the easy H-bond formation property of *N*-oxyamide compounds.

From methyl α -D-glucopyranoside, the 2,6-functionalized pyranoid glycoaminoxy acid derivatives have been successfully prepared as a multifunctional building block for further derivatization to new *N*-oxyamide glycolipids. From glucose or galactose pentaacetate and (*S*)-1,2-di-*O*-benzyl-glycerol, we have successfully achieved the first synthesis of *N*-oxyamide-linked β -glycolipids with one or two lipids chains, as novel mimics of glyco glycerolipids and glycosphingolipids.

In addition, the (*2R*) and (*2S*)-3- β -*O*-glycosyl aminoxy esters have been stereoselectively synthesized from (*2R*)- β -glycoglycerol, with Mitsunobu reaction and Lattrell-Dax epimerization as key steps. *N*-Oxyamide-linked glycopeptides have been prepared from the orthogonally protected glycosyl aminoxy esters.

RESUME

Les glycoconjugués comme les glycolipides et les glycopeptides sont impliqués dans de nombreux processus biologique, physiologique et pathologique, tels que les interactions cellule-cellule, les infections virales et bactériales, les réponses immunitaires, le cancer, etc. Ces propriétés ont suscité de recherche intensive pour la synthèse de mimes de glycoconjugués pour des applications en biologie et en pharmacie. Cette thèse est consacrée à la synthèse de glycolipides et de glycopeptides liés par la liaison *N*-oxy amide qui possède une meilleure stabilité métabolique et une facilité de formation de liaison H conduisant à des structures secondaires très intéressantes.

Les dérivés de pyranoid glycoaminoxy acides fonctionnés en positions 2 et 6 du sucre sont synthétisés à partir du methyl α -D-glucopyranoside, puis transformés en *N*-oxyamide glycolipides. En tant que nouveaux mimes de glyco glycérolipides et glycosphingolipides, les *N*-oxyamide β -glycolipides possédant une ou deux chaînes lipidiques sont préparés à partir du glucose ou du galactose penta-acétate et de (*S*)-1,2-di-*O*-benzyl-glycérol.

De plus, une synthèse stéréosélective de (*2R*) et de (*2S*)-3- β -*O*-glycosyl aminoxy esters a été réalisée à partir du (*2R*)- β -glycoglycérol, avec la réaction de Mitsunobu et l'épimérisation de Lattrell-Dax comme étapes clés. Les *N*-oxyamide glycopeptides ont pu être préparés à partir de glycosyl aminoxy esters orthogonalement protégés.