Distribution Agreement

In presenting this thesis or dissertation as a partial fulfillment of the requirements for an advanced degree from Emory University, I hereby grant to Emory University and its agents the non-exclusive license to archive, make accessible, and display my thesis or dissertation in whole or in part in all forms of media, now or hereafter known, including display on the world wide web. I understand that I may select some access restrictions as part of the online submission of this thesis or dissertation. I retain all ownership rights to the copyright of the thesis or dissertation. I also retain the right to use in future works (such as articles or books) all or part of this thesis or dissertation.

Signature:

_____ _____

Claney L. Pereira Date

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of Methodology: Synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of Natural and Unnatural oligomers

By

Claney L. Pereira
Doctor of Philosophy

Chemistry

_____

Frank E. McDonald, Ph.D.
Advisor

_____

Dennis C. Liotta, Ph.D.
Committee Member

_____

Simon Blakey, Ph.D.
Committee Member

Accepted:

_____

Lisa A. Tedesco, Ph.D.
Dean of the James T. Laney School of Graduate Studies

_____

Date

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of Methodology: Synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of Natural and Unnatural oligomers

By

Claney L. Pereira
B.S., St. Aloysius College, Mangalore University, 1999
M.S., Mangalore University, 2001

Advisor: Frank E. McDonald, Ph.D.

An abstract of
A dissertation submitted to the Faculty of the
James T. Laney School of Graduate Studies of Emory University
in partial fulfillment of the requirements for the degree of
Doctor of Philosophy
in Chemistry
2010

Abstract

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of Methodology: Synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of Natural and Unnatural oligomers

By Claney L. Pereira

Chapter 1. Total synthesis of Fumonisin B_1: The first total synthesis of fumonisin B_1, a sphingolipid biosynthesis inhibitor was achieved. The synthesis used the 2-oxonia-[3, 3]-sigmatropic rearrangement methodology developed in the Mc-Donald laboratory, thereby demonstrating the utility of these rearrangements in total synthesis. The synthesis also confirmed the assigned stereochemistry and the biological profile of the natural product.

Chapter 2. Synthesis of Unnatural Sphingolipids, 1-deoxy-5-hydroxysphinganine and its diastereomers: Utilizing 2-oxonia-[3, 3]-sigmatropic rearrangements, we synthesized *cis* and *trans* homoallylic alcohols which were converted to the epoxides. Epoxide opening using an azide nucleophile and azide reduction gave the respective 1-deoxy-5-hydroxysphinganine sphingolipid compounds.

Chapter 3. Development of Methodology, Synthesis of *cis*- and *trans*- homoallylic alcohols: Diastereo- and enantioselective synthesis of branched homoallylic synthons were achieved. These were then used to synthesize the linear *cis*- and *trans*- homoallylic alcohols through a 2-oxonia-[3, 3]-sigmatropic rearrangement.

Chapter 4. Human Milk Oligosaccharides (HMO), Synthesis of Natural and Unnatural oligomers: Natural HMO's 2'-fucosyllactose, 3-fucosyllactose and unnatural HMO 2'-glucoseptanosyllactose were synthesized using a lactose derived acceptor and a fucosyl or septanosyl derived donor. The synthesis utilised a greener approach of using non-toxic reagents and milder reaction conditions for the preparation of mono- and disaccharide synthons.

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of Methodology: Synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of Natural and Unnatural oligomers

By

Claney L. Pereira
B.S., St. Aloysius College, Mangalore University, 1999
M.S., Mangalore University, 2001

Advisor: Frank E. McDonald, Ph.D.

A dissertation submitted to the Faculty of the
James T. Laney School of Graduate Studies of Emory University
in partial fulfillment of the requirements for the degree of
Doctor of Philosophy
in Chemistry
2010

Acknowledgments

I would like to express my deepest gratitude to everyone who has helped, encouraged and supported through my five years of graduate school. The main architect who made all this possible is my advisor Dr. Frank E. McDonald. His intellectual guidance, mentorship, enthusiasm and passion for the subject, has played a key role for me to being a good scientist. The immense freedom to think and work creatively, a word of encouragement in times of frustration has made the past five years in the laboratory worthwhile.

I extend my appreciation to my committee members, Dr. Dennis C. Liotta and Dr. Simon B. Blakey. They have also helped me grow scientifically from year to year with their constructive suggestions and motivation.

I would like to extend my gratitude to Dr. Shaoxiong Wu and Dr. Bing Wang for there support in the NMR facility. Being a student instructor under them has helped me immensely both in my research and personal development. Thanks are also due for Dr. Kenneth Hardcastle in the X-ray crystallography center and to Dr. Fred Strobel in the mass spectrometry facility.

Patti Barnett, Sarah Keller and Steve Krebs from the stockroom, Ann Dasher our graduate coordinator have played a key role to ensure that my research work goes well. They are the people behind the scene and I owe them a huge gratitude.

The McDonald laboratory has been a wonderful place to work because of the diversed group of people. Dr. Bonsuk Koo, Dr. Hi-Hung Chen, Dr. Ann Dougherty, Dr. Brad Balthaser, Dr. ZhongBo Fei and Dr. Rongbiao Tong were

great colleagues. Dr. Omar Robles, was a great roommate for 4 years, Mary Smart was a source of encouragement when I began working in the first few years.

My friends at Emory University and around United States like Nadege, Armin, Mark Baillie, Tiffany, Savita Sharma, Reggie, Alex Wein, Amnider, Lokesh, Pradeep and my classmates have helped me build up a whole new atmosphere.

I also made a friend for life, Dr. Matthew Allen Boone. As avid aviation enthusiasts we have spent quiet sometime plane spotting in Atlanta. He has taught me a lot about United States, fishing and tennis. His parents David and Debbie Boone have become my family far away from home.

My family has been riding along with me in this journey every step of my way. My sister Clara and my brother-in-law Alwyn have been providing the well needed boost. My niece Calvina and my nephew Ashton have filled me with joy. My friends back in India, Safia, Shekar, Umesh and my relatives have been a constant source of encouragement. My girlfriend Sharavathi has been in every step of my graduate school, filling me with love, confidence, and assurance.

This thesis is dedicated to my parents Valerian and Lethysia Pereira. My Dad and Mom’s love and support has been unwavering as always.

Table of Contents

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

List of Figures

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of methodology: synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of natural and unnatural oligomers

List of Schemes

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of methodology: synthesis of *cis*- and *trans*-homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of natural and unnatural oligomers

List of Tables

Chapter 1. Total synthesis of Fumonisin B_1

Chapter 2. Synthesis of Unnatural Sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

Chapter 3. Development of Methodology: Synthesis of *cis*- and *trans*- homoallylic alcohols

Chapter 4. Human Milk Oligosaccharides: Synthesis of Natural and Unnatural oligomers

Abbreviations

Ac	acetate
appd	apparent doublet
appt	apparent triplet
Aq	aqueous
BBN	9-Borabicyclo (3.3.1) noname
BINAP	2,2'-bis(diphenylphosphino)-1,1'-binaphthyl
BINOL	1,1'-Bi-2-naphthol
Bn	benzyl
Boc	Di-*tert*-butyl dicarbonate
Bz	benzoyl
CBZ	Carbobenzyloxy
CDI	*N,N'*-Carbonyldiimidazole
COD	cyclooctadiene
CSA	camphorsulphonicacid
d	doublet
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DCC	*N,N'*-Dicyclohexylcarbodiimide
DIAD	diisopropyl azodicarboxylate
DIBAL	diisobutylaluminum hydride
DIPT	diisopropyl tartrate
DMAP	*N, N*-dimethylaminopyridine
DMF	dimethylformamide
DMP	Dess-Martin periodinane
DMSO	dimethylsulphoxide
DTBAD	Di-tert-butyl azodicarboxylate
EC50	half maximal effective concentration
EDC	1-ethyl-3-(3-dimethylaminopropyl) carbodiimide)
Eng	enigmol
EtOAc	ethylacetate
IBX	iodoxybenzoicacid
LDA	lithiumdiisopropylamine
LiHMDS	lithium bis(trimethylsilyl)amide
m	multiplet
m-CPBA	metachloro perbenzoicacid
mL	milliliter
MS	molecular seives
MW	microwave
n-BuLi	*n*-butyllithium

NIS	N-iodosuccinimide
Ph	phenyl
$PhCF_3$	trifluorotoluene
Piv	pivaloate
PMB	para-methoxylbenzyl
p-TSA	para-toluenesulfonic acid
Py	pyridine
q	quartet
RT	room temperature
s	singlet
So	Sphingosine
t	triplet
TBAF	tetrabutylammonium fluoride
TBHP	tetrabutyl hydrogenperoxide
Tf	triflic
TFA	trifluoroaceticacid
THF	tetrahydrofuran
TIPS	triisopropylsilyl
TMS	trimethylsilyl

Chapter 1

Chapter 1

Total synthesis of Fumonisin B_1

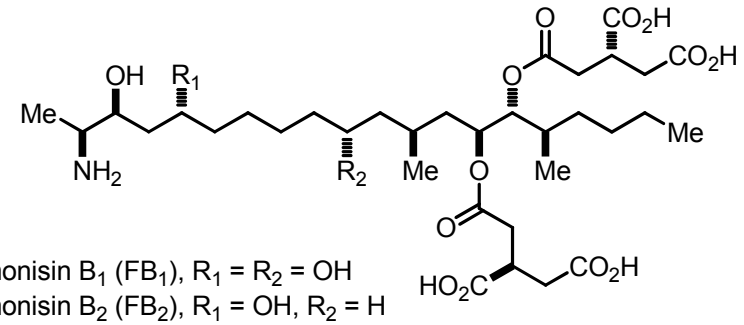
1.1. Introduction and background

Corn is the main food source for both people and animal throughout the world. It could be considered a miracle crop because of its vast applications. On the other hand the contaminants of corn, namely fungi are causing grave concerns throughout the world because of their ability to cause diseases.

Fusarium verticillioides Nirenberg (= *Fusarium moniliforme* Sheldon) is one of the most prevalent of the fungal contaminants of corn.[1] The first problems associated with these causative agents were equine leukoencephalomalacia (ELEM) reported from South Africa in 1970.[2] It was in 1988 that the mycotoxins named fumonisins were first isolated and their chemical structures determined (figure 1).[3]

Since their discovery 28 different fumonisins have been characterized from 15 *Fusarium* species and are grouped into four main categories, fumonisins A, B, C and P.[1] The most abundant of the series is B and the predominant mycotoxin is fumonisin B_1 (FB_1) **(1)**, which accounts for 70-80% (figure 1). The highest yield of FB_1 reported for a *Fusarium* species was obtained from *F. proliferatum* from Spain, from cultured maize (31,000 mg/Kg).[1]

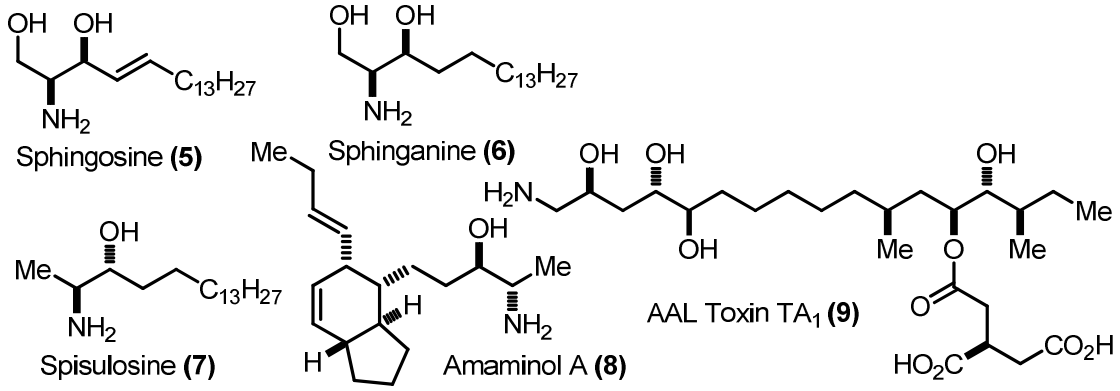
Figure 1. Structure of fumonisin-B series



1. fumonisin B_1 (FB_1), $R_1 = R_2 = OH$
2. fumonisin B_2 (FB_2), $R_1 = OH$, $R_2 = H$
3. fumonisin B_3 (FB_3), $R_1 = H$, $R_2 = OH$
4. fumonisin B_4 (FB_4), $R_1 = R_2 = H$

Structurally, FB_1 **(1)** contains 10 stereocenters, two tricarballylic acid fragments and an amino diol head group (figure 1).[3] They are also structurally similar to the AAL phytotoxins **(9)** produced by *Alternaria alternate* sp*. lycopersici*, a tomato pathogen (figure 2).[4] The relative and absolute stereochemistry of fumonisin B_1 was established using a combination of synthetic derivatization and NMR methods.[5] The aminodiol head is structurally similar to sphingoid bases found in sphingolipids like sphingosine **(5)** and related compounds, which also explains fumonisin's biological role as a sphingolipid biosynthesis inhibitor (figure 2).[6]

Figure 2. Structurally similar-sphingoid based compounds



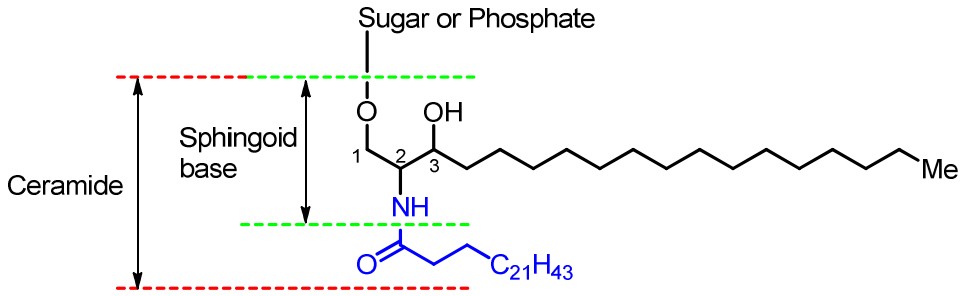
1.1.1. Biological significance of fumonisins

Fumonisin classes of natural products are associated with biological effects that occur at the molecular, cellular and organism levels. The free amino group is suspected to play a specific role in this biological activity both in terms of toxicity and inhibition.[7] The biological role of fumonisins is outlined below.

a) Inhibition of sphingolipid biosynthesis

The structural similarity of fumonisins and the sphingolipid, sphingosine (**5**, figure 2) indicates that they may be biosynthetically related and have thus generated interests in their mode of action (figure 3).[8] Sphingolipids are found in all eukaryotic cells and as regulators of cell functions. Over 300 sphingolipids have been identified and they have a common long chain sphingoid base backbone (figure 3, details in the next chapter).

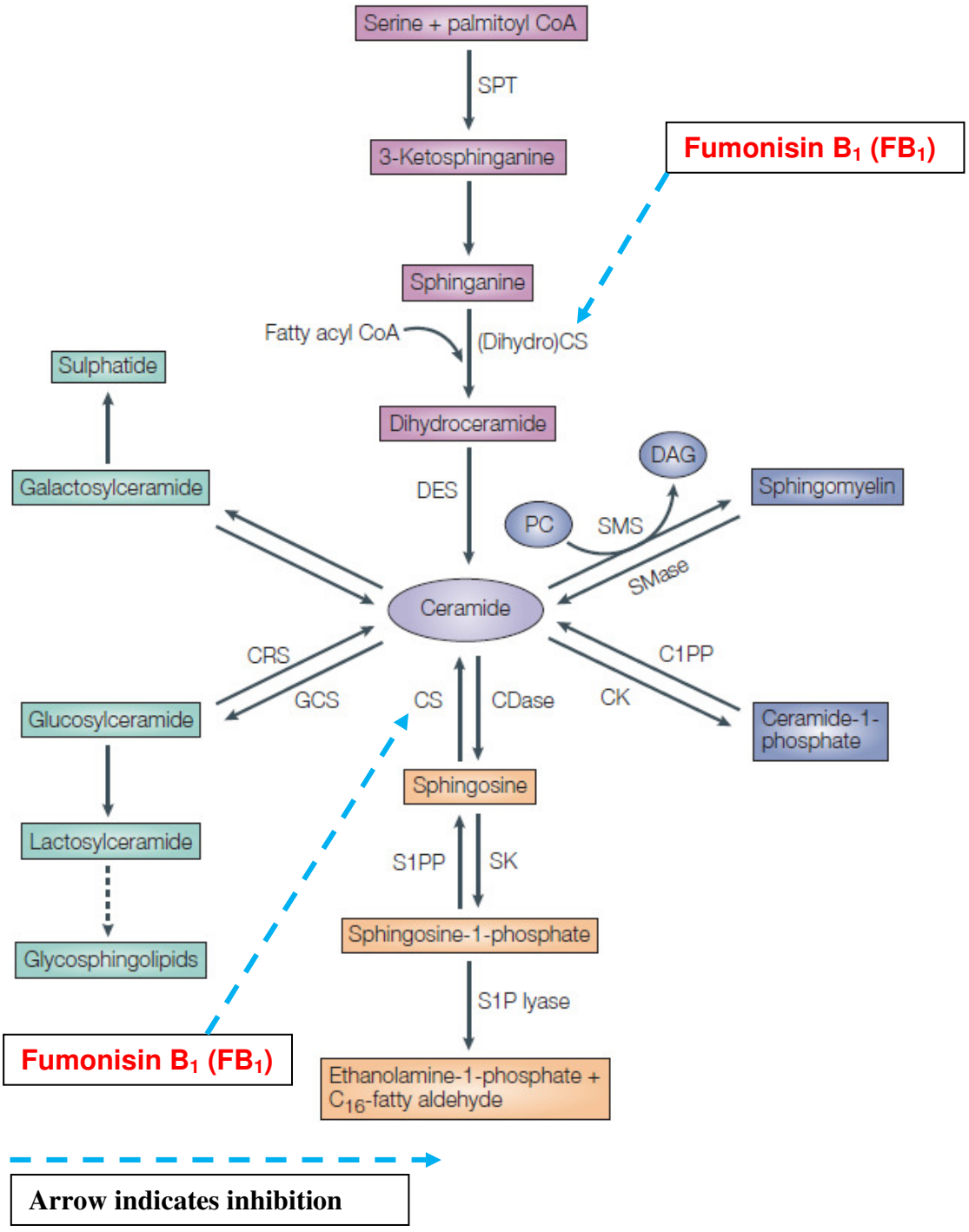
Figure 3. Representative structure of sphingolipid



Sphingolipids are synthesized via a *de novo* pathway at the cytosolic face of the endoplasmic reticulum. The enzyme serine palmitoyltranferase (SPT) catalyzes the condensation of L-serine and palmitoyl-CoA to produce ketosphinganine which is reduced by ketosphinganine reductase (KR) to sphinganine (figure 4).[9] This is rapidly converted to dihydroceramide (*N*-

acylsphinganine) by a family of sphinganine and sphingosine *N*-acyltransferase enzyme collectively called ceramide synthase (DCS). The *E* double bond is then introduced by a dehydrogenation process catalysed by desaturase (DES).

Figure 4. *de novo* sphingolipid synthesis in mammalian cells

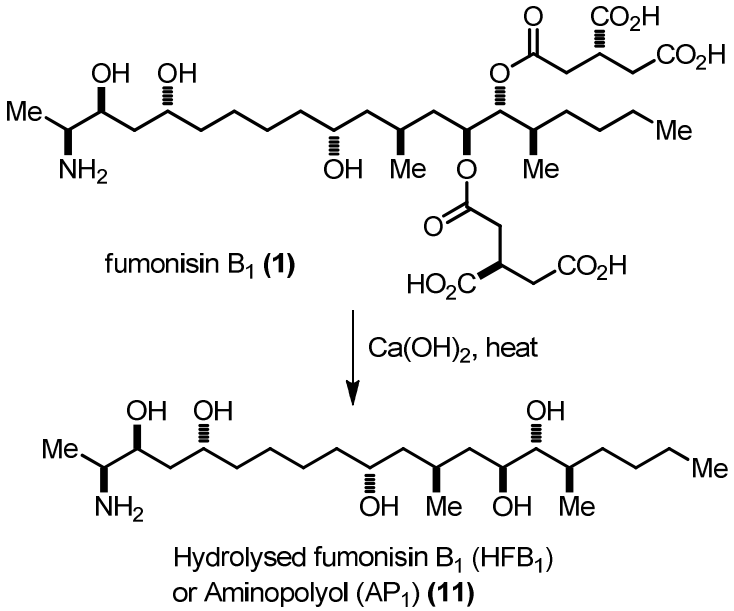


Ceramide then becomes a major substrate and a branching point for the synthesis of more complex sphingolipids. Ceramide can be metabolized to sphingosine by ceramidases (Cdase), which in turn are phosphorylated by sphingosine kinase (SK) to form sphingosine-1-phosphate (S1P). This can be synthesized back to sphingosine by phosphatase (S1PP) or can be cleaved by lyase (S1P lyase) into ethanolamine-1-phosphate and C_{16}-fatty aldehyde (figure 4). This process takes place in the endoplasmic reticulum and the Golgi apparatus. The degradation of complex sphingolipids takes place in lysosomes, endosomes and plasma membrane.

FB_1 is a potent inhibitor of ceramide synthase (CS) (figure 4).[6] This results in the accumulation of sphinganine, a cytotoxic and to some extent sphingosine and they are converted to sphinganine-1-phosphate and sphingosine-1-phosphate respectively. This process also decreases the amount of dihydroceramide, ceramide and other complex sphingolipids. This disruption in the sphingolipids metabolism is an important part in the cascade of events leading to altered cell growth, differentiation and has been proposed as the main cause for toxicity and possible carcinogenicity of FB_1.[6]

FB_1 **(1)** is relatively heat stable and thus persists in food. By a process called nixtamalization (scheme 1), treating corn with aqueous calcium hydroxide and heat, the tricarballylic acid (TCA) can be removed to produce hydrolyzed fumonisin (HFB_1) or aminopolyols (AP_1) **(11)**.[10] This molecule is 10 fold less potent than the parent molecule in inhibiting ceramide synthase.[10]

Scheme 1. Process of nixtamalization



b). Toxic effects

Effects in humans: Fumonsins are associated with esophageal cancer in humans. Studies carried out in China and South Africa showed that higher levels of *F. verticillioides* and higher concentrations of FB_1 **(1)** and FB_2 **(2)** in corn growing areas with high incidence of esophageal cancer.[11] The disruption of sphingolipid metabolism by FB_1 may affect folate uptake, deficiency of which according to clinical trials, may be a major risk factor for neural tube defects.[12] A consumption of sorghum contaminated with *Fusarium* and *Aspergillus* fungi containing high amounts of FB_1 was associated with acute mycotoxicosis, characterized with acute abdominal pain and diarrhea.[13] The international agency for research on cancer has categorized FB_1 as possibly carcinogenic to humans (group 2B).[14]

Effects in animals: The main target organs of fumonisin B_1 in animals are liver, kidney and brain. In horses FB_1 causes equine leukoencephalomalacia (ELEM),

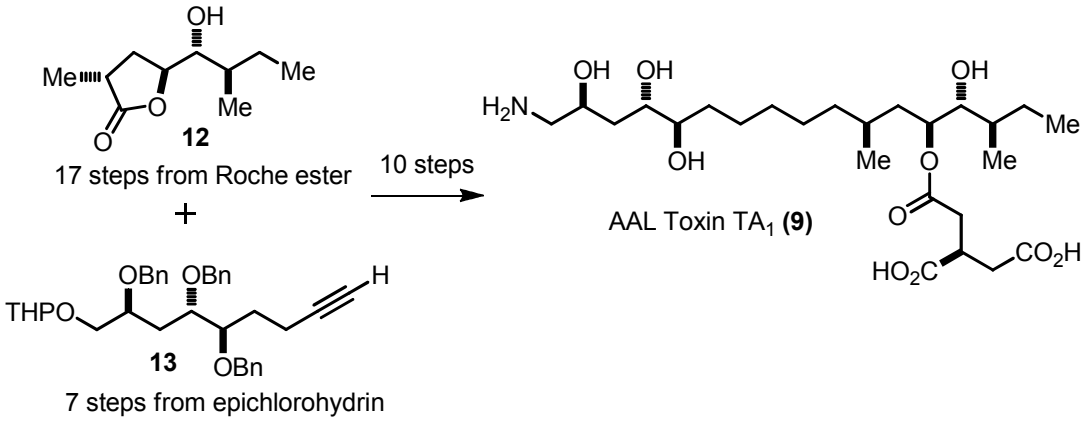
a neurotoxic disease.[15, 3c] Studies carried out by administrating FB_1 orally or intravenously led to ELEM, resulting in brain lesions and histopathological abnormalities.[16] In pigs FB_1 is associated with porcine pulmonary edema, wherein FB_1 reduces the mechanical efficiency of the left ventricle.[17] This is hypothesized because of the accumulation of sphinganine and sphingosine. Studies carried out on rats and mice have indicated that FB_1 is carcinogenic. It resulted in the formation of cholangio and hepatocellular carcinoma.[18] As with other effects carcinogenicity of FB_1 is associated with disruption of sphingolipid metabolism. The reported IC_{50} value for inhibition of 50% cell proliferation in H4TG (rat hepatoma) and MDCK (dog kidney) cell lines ranged from 1.7 to 56 μM.[18b, 3b]

1.2. Synthetic studies on fumonisins

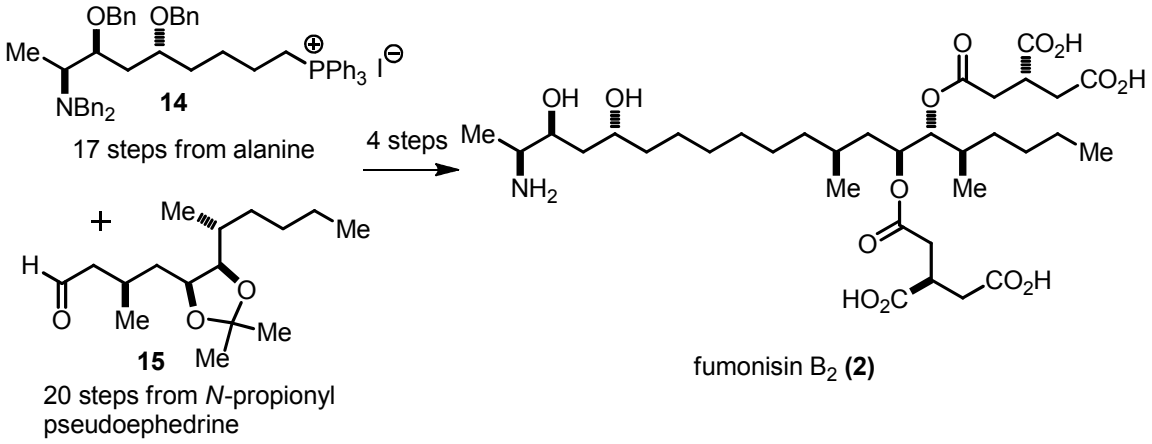
Extensive studies have been carried out to establish the absolute stereochemistry of FB_1 by various groups but very little synthetic progress has been observed in this family of natural products.[5] Though not belonging to fumonisin, but structurally similar and also an inhibitor of sphingolipid biosynthesis is AAL toxin TA_1 **(9)**.[4] The first synthesis of this molecule was reported by Oikawa *et al.* (scheme 2a).[19] The first total synthesis of fumonisins was of FB_2 **(2)**, reported by Kishi using a highly stereoselective and convergent approach (scheme 2b).[20] The two complex fragments were put together by a Wittig homologation and their protecting group pattern allowed for the installation of the tricarballylic acid part thus completing the synthesis.

Scheme 2. Synthesis of AAL-toxin TA_1 **(9)** and fumonisin B_2 **(2)**

a. Oikawa synthesis



b. Kishi synthesis

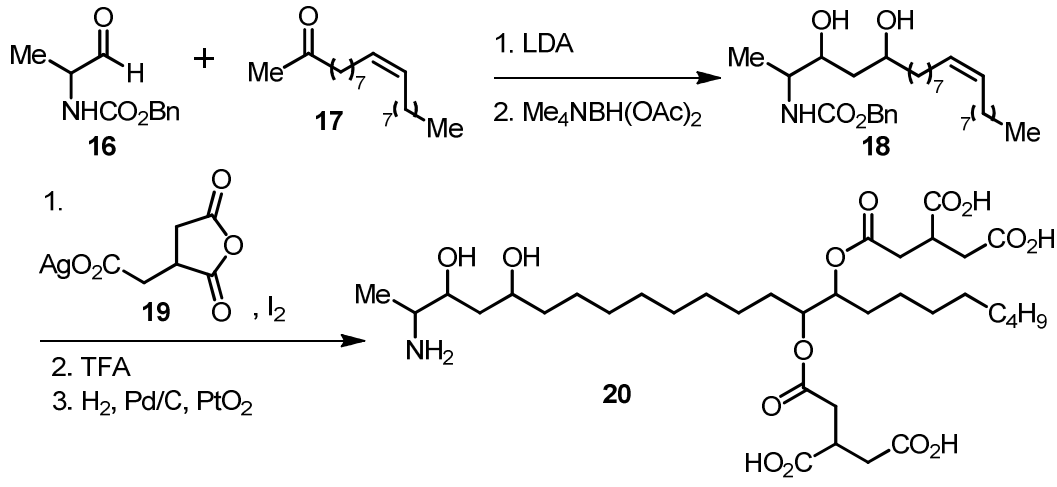


The first attempted synthesis of FB_1 **(1)** was the construction of an analog by Kraus *et al*. using aldol condensation (scheme 3a).[21] During their synthesis the stereochemistry of FB_1 **(1)** was unknown. The laboratory of Gurjar reported the synthesis of fumonisin B_1 aminopentol as the hexaacetate derivative **24**.[22] Their synthesis used carbohydrates as precursors and their protecting group strategy did not allow them to add the tricarballylic esters (scheme 3b). Overall

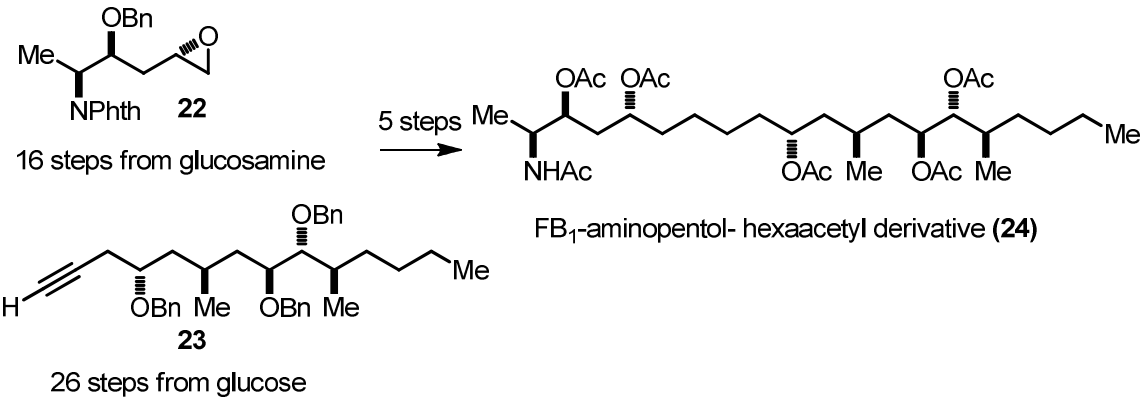
these syntheses have been lengthy and any new approach towards this family of natural products had to take this into account.

Scheme 3. Synthesis of fumonisin B_1 related analogs

a. Kraus synthesis



b. Gurjar synthesis

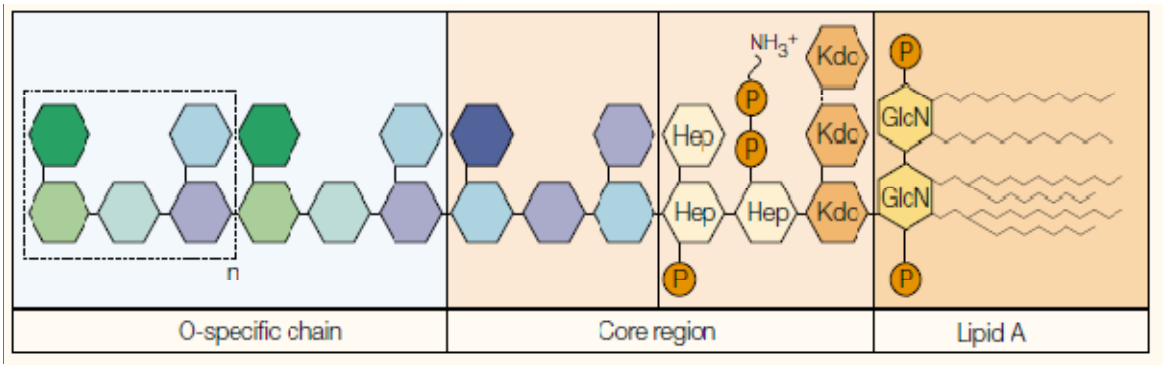


1.3. Why a synthetic endeavour for fumonisin B_1?

The 20 carbon FB_1 **(1)** mycotoxin with 10 stereocentres and two carballylic acid fragments is a complex synthetic target.[3] Isolating these hydrophilic molecules from their natural source is difficult because they are not soluble in organic solvents. The most satisfactory solvent combination for extraction thus

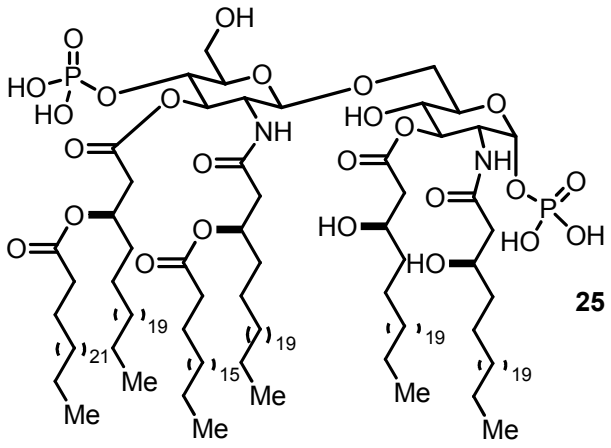
far has been a combination of water with methanol or acetonitrile.[23] The problem with such a combination is that many polar impurities like salts, sugars and peptides are co-extracted. The similarity in structures between fumonisins makes it difficult to be purified from one another.[23] The other issue with the natural product isolation is the presence of endotoxins.[24] Endotoxins, also called lipopolysaccharides (LPS) are an important structural component of the outer membrane of gram negative bacteria.[25] They consist of a lipid component, Lipid A, a core oligosaccharide region and a long heteropolysaccharide O-specific chain (figure 5).[26]

Figure 5. Representative endotoxin (LPS) from *Escherichia coli*



Lipid A **(25)** is a member of a family of glycophospholipids having unique structural features. A representative example from *Escherichia coli* is shown in figure 6. This is a important disease causing toxin of Gram-negative bacteria. TNF-α (Tumor necrosis factor) is considered as a possible endogenous mediator of endotoxin and has been a way to monitor the effects of LPS.[27]

Figure 6. Representative Lipid A from *Escherichia coli*



25

In humans and animals endotoxins have a very strong biological response with symptoms ranging from fever, to hypotension, adult respiratory distress syndrome, disseminated intravascular coagulation and endotoxin shock.[25a] Natural FB_1 that is obtained from Sigma Aldrich shows a considerable amount of endotoxin, whereas that from Promec (South Africa) does not show any.[28] It thus becomes very important to ascertain whether the biological profile shown, is by the mycotoxin FB_1, or the endotoxin. Thus a synthesis of this natural product becomes befitting.

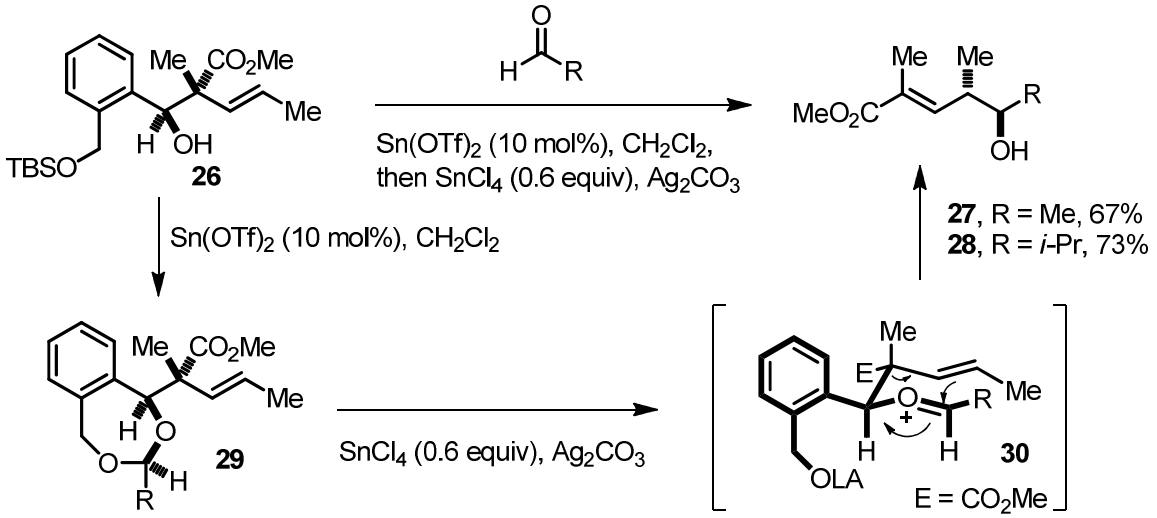
1.4. Synthetic strategy towards the total synthesis of FB_1 (1)

1.4.1. Initial approach

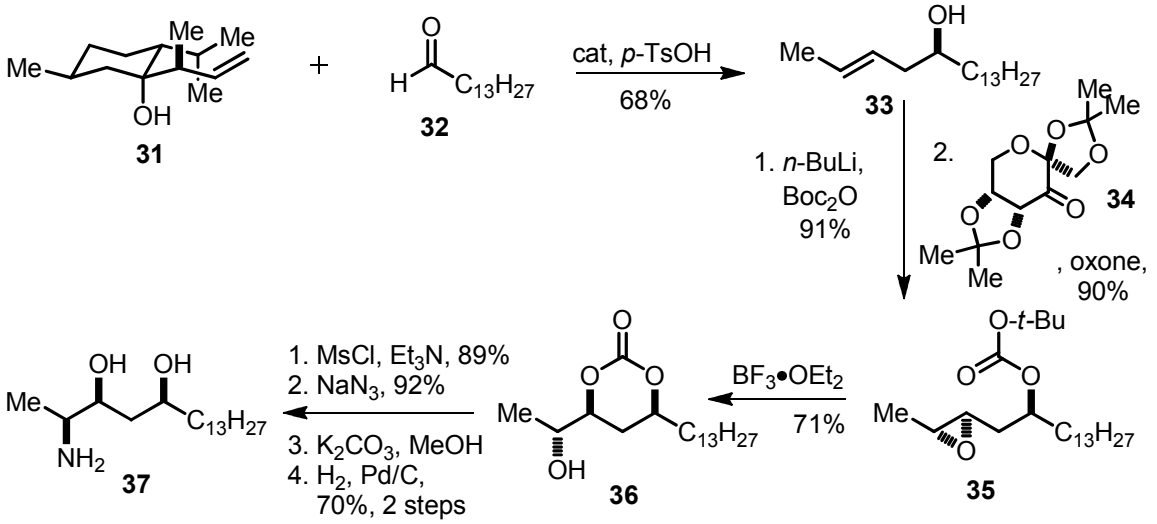
Our initial work towards the total synthesis of FB_1 was carried out by Dr. Yi-Hung Chen in the McDonald lab.[29] The basis for this approach was the development of an allylic rearrangement using synthon **26** (Details about these allylic rearrangements will be addressed in depth in chapter 3 of this thesis) in the lab by the same (scheme 4),[30] and a stereoselective synthesis of unnatural

sphingolipid enigmol **37**, the sphingolipid that has the same sphingoid head group as FB_1 **(1)** by John Wiseman in the McDonald lab utilizing an allylic rearrangement, epoxidation and oxacyclization as the key step (scheme 5).[31]

Scheme 4. Allylic rearrangement with synthon **26** by Dr. Yi-Hung Chen



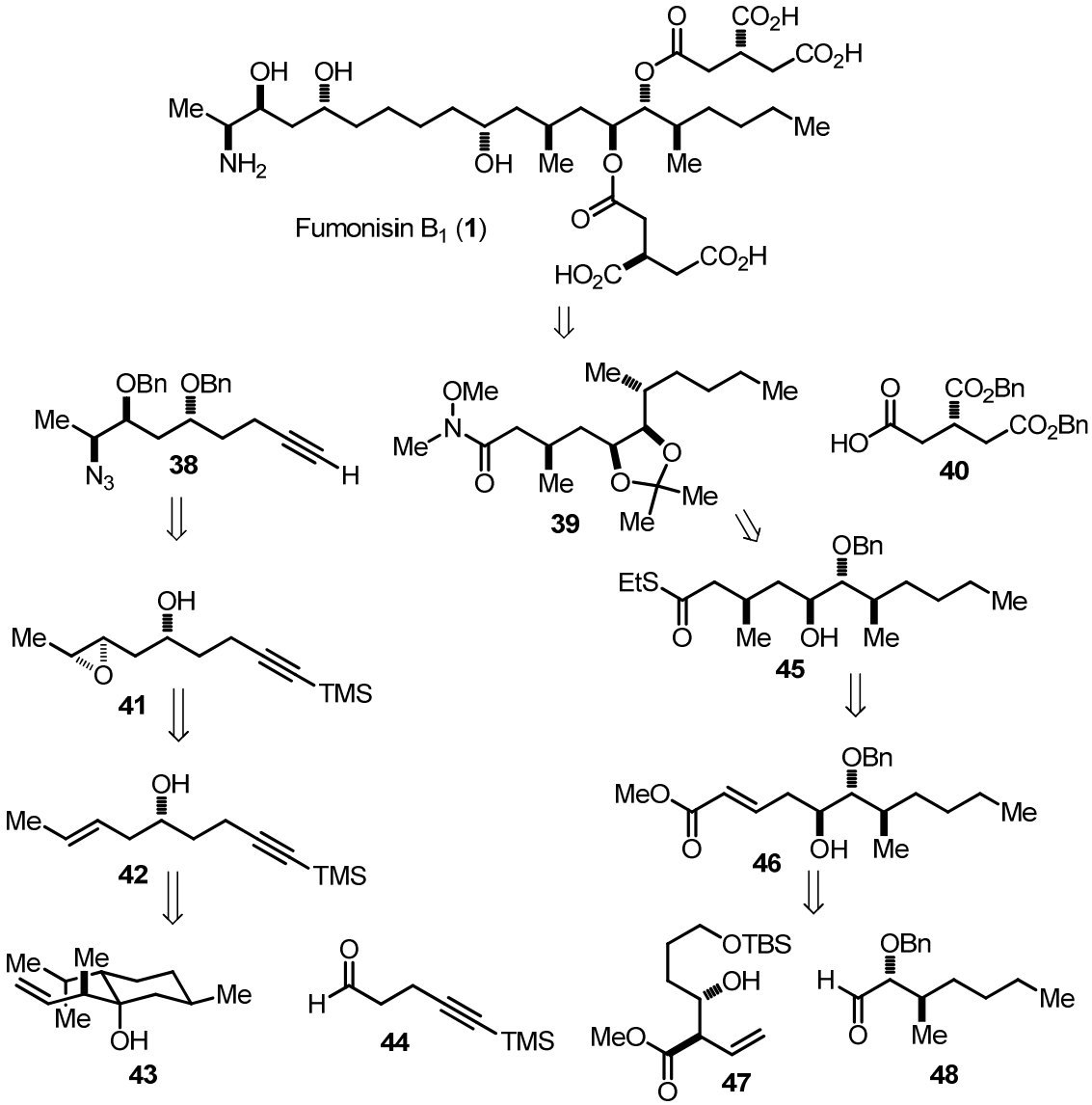
Scheme 5. Enigmol **37** synthesis by John Wiseman



The initial approach of Dr Yi-Hung Chen towards the synthesis of FB_1 **(1)** is outlined in the retrosynthetic scheme 6. The complete carbon skeleton of FB_1

(1) would be put together from intermediates **38**, **39** and tricarballylic acid fragment **40** in the last stage. The C-20 carbon framework would be put together from alkyne **38** and Weinreb amide **39**.[32] The final stereocenter at C10 would be set using various methods available in literature for alkynone reduction.[33]

Scheme 6. Proposed retrosynthesis of fumonisin B_1 **(1)**



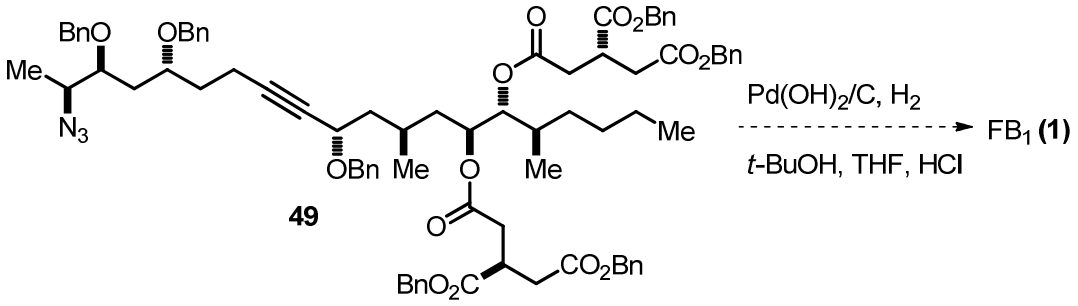
The left hand fragment **38** would be obtained starting from aldehyde **44** through an allylic rearrangement using the synthon **43** obtained from (-)-

menthone.[34] Adopting the strategy used for the synthesis of enigmol **(37)**, [31] epoxidation followed by oxacyclization and azide substitution would give **38**. Right hand fragment **39** would be put together utilizing the allylic rearrangement to obtain the homoallylic alcohol **46**, [30] which would be converted to a thioester **45** setting the stage for a Michael addition to introduce the methyl group.[35] Overall the intermediate **46** would form the linchpin for the synthesis, which would also establish to demonstrate the application of the 2-oxonia-[3, 3]-Cope rearrangement in total synthesis.

1.4.2. The unexpected rearrangement

Dr Yi-Hung Chen synthesized the assigned left hand fragment **38** and right hand fragment **39** and put together the entire carbon frame work **49** with tricarballylic acid fragment **40** but was not able to deprotect the benzyl ether and ester groups (scheme 7).[29]

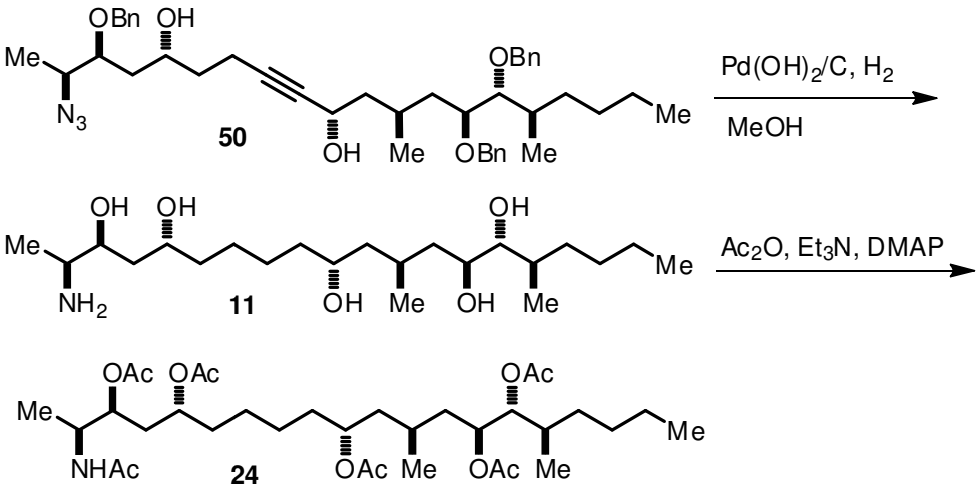
Scheme 7. Proposed and attempted synthesis of FB_1 **(1)** by Dr. Yi-Hung Chen



49

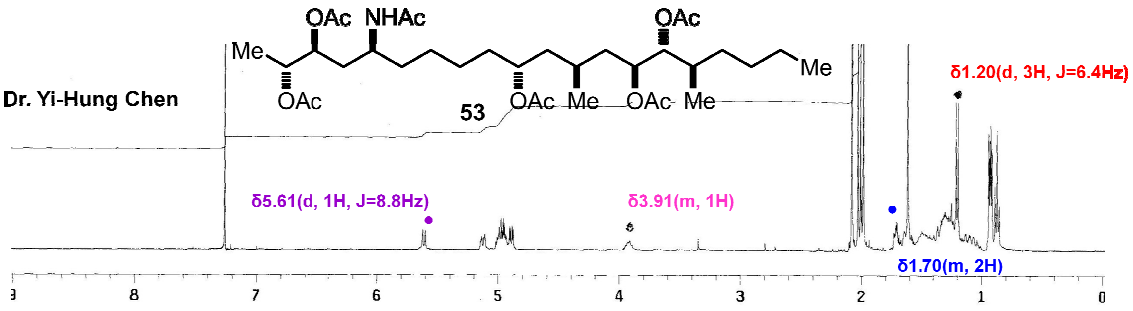
In an effort to push forward in the synthesis, an attempt was made to synthesize the FB_1-hexaacetate derivative **24**. In this case the deprotection of the benzyl ether from intermediate **50** was successful, peracylation gave a compound assigned as structure **24** (scheme 8).[29]

Scheme 8. Proposed and attempted synthesis of FB_1-hexaacetate **24** by Dr. Yi-Hung Chen



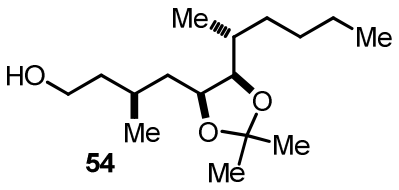
A comparison of the ^{1}H NMR data of **24** with that reported by Gurjar did not match.[22] [Gurjar, (400 MHz, $CDCl_3$): δ 0.87 (t, *J* = 6.8 Hz, 3H), 0.93 (d, *J* = 6.8 Hz, 6H), **1.10 (d, *J* = 6.7 Hz, 3H)**, 1.2-1.65 (m, 20H), **1.76 (m, 2H)**, 1.99 (s, 6H), 2.01, 2.02, 2.07, 2.09 (4s, 12H), **4.17 (m,** 1H), 4.8-5.02 (m, 4H), 5.17 (dt, *J* = 2.5, 10.1 Hz, 1H), **5.58 (d, *J* = 9.3 Hz, 1H)**]. (The proton signals that did not match with Dr. Yi-Hung's are highlighted in bold. The ^{1}H spectra is shown in figure 7).

Figure 7. ^{1}H NMR data for proposed FB_1-hexacaetate **24** synthesis by Dr. Yi-Hung Chen with the correct structure **53** shown



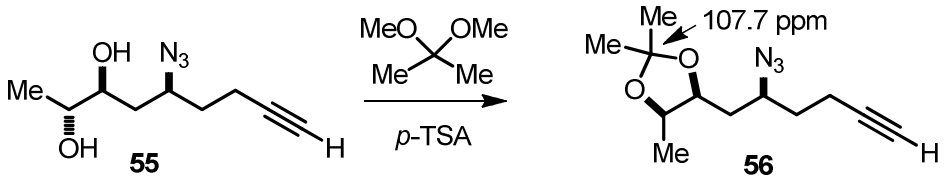
Yi-Hung had established the stereochemistry of the right hand fragment **39** by comparing the ^{1}H NMR data of intermediate **54**, to that reported by Kishi (figure 8).[20]

Figure 8. Kishi intermediate **54** during the synthesis of fumonisin B_2



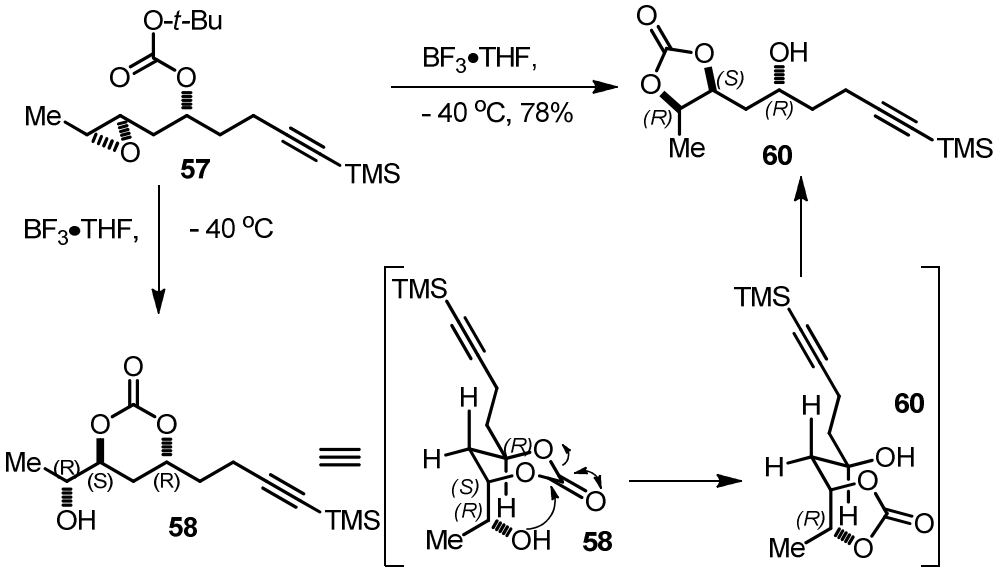
So from this it was concluded that there might have been a regiochemical issue with the left hand fragment **38** of the molecule (scheme 6). Further evaluation of one of the intermediate **55** from the left hand fragment **38** as its acetonide **56** substantiated this finding (scheme 9). From literature it is well known that a 6-membered ring acetonide with *syn*-stereochemistry has the quaternary carbon appear at lower than ≤100 ppm and the two methyls at 30 and 19 ppm in the ^{13}C NMR.[36] In the case of *anti*-stereochemistry this is observed at ≥100 ppm for the quaternary and 24 ppm for the two methyls. The ^{13}C data for compound **56** showed peaks at 107.7, 28.4, and 25.7 ppm. A literature search for ^{13}C data for 5-membered acetonide indicated that the quaternary carbon appears between 108-110 ppm.[37]

Scheme 9. Synthesis of acetonide **56** to establish relative stereochemistry at C3 and C5 position



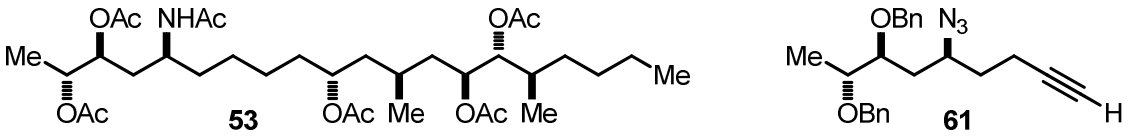
Shier *et al.*, during their study to determine the absolute configuration of fumonisin B_1 **(1)** had also shown that the ketal carbon of the 6-membered acetonide appeared at 100.4 ppm in the ^{13}C NMR for the *anti*-stereochemistry of the 3, 5-diol.[5e] Based on these evidences the unexpected outcome was explained as a rearrangement of the 6-membered cyclic carbonate **58** to the 5-membered cyclic carbonate compound **60** (scheme 10).

Scheme 10. Proposed mechanism for the unexpected rearrangement



Based on these finding the correct structures for intermediates **24** (now **53**) and **38** (now **61**) were proposed. This finding also warranted that the results published from the McDonald group on the synthesis of isoenigmol needed to be revisited (details in chapter 3 of this thesis).[31]

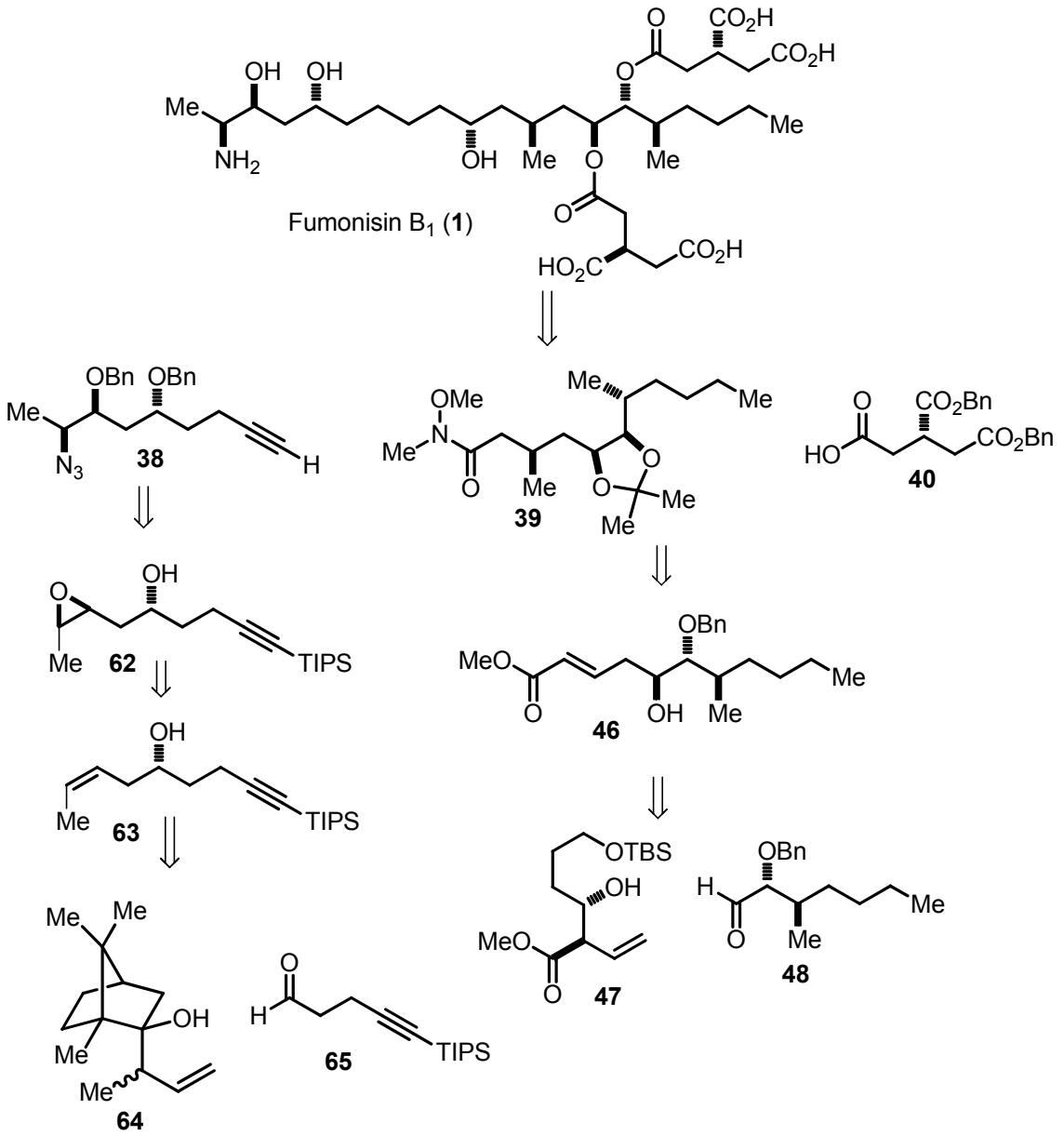
Figure 9. Corrected structures **53** and **61** in the synthesis of FB_1 **(1)** by Dr. Yi-Hung Chen



1.4.3. Total synthesis of FB_1 (1), the new approach

With the problems identified, a new approach to the synthesis of left hand fragment **38** was envisioned which eliminated some steps to shorten the synthesis. The new retrosynthetic scheme is outlined in scheme 11.

Scheme 11. New retrosynthetic approach towards fumonisin B_1 **(1)**



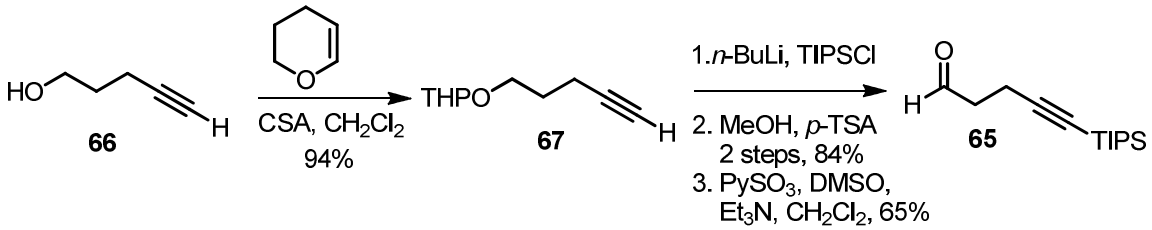
For the right hand fragment **39** rather than using the thioester for the Michael addition of the methyl group, a methyl ester **46** would be used thus saving some steps.[38] The left hand fragment **38** would be obtained through an allylic rearrangement as before, but starting from intermediate **63** with a *cis*-geometry.[39] Epoxidation followed by azide substitution without oxacyclization would give **38**. Tricarballylic acid fragment **40** was previously synthesized using Kishi's method and now a new method was envisioned which would reduce the steps by 4.[40] Finally modifiying the final approach of the synthesis by eliminating the diimide reduction step to carry out one global reduction would give FB_1 **(1)**.

1.5. Results and Discussions

1.5.1. Synthesis of left hand alkyne fragment 38

The synthesis began from the commercial alkynol **66** (scheme 12).[41] Protection of the alcohol and alkyne followed by removal of THP group and the oxidation of the primary alcohol gave aldehyde **65** required for the rearrangement in 4 steps.

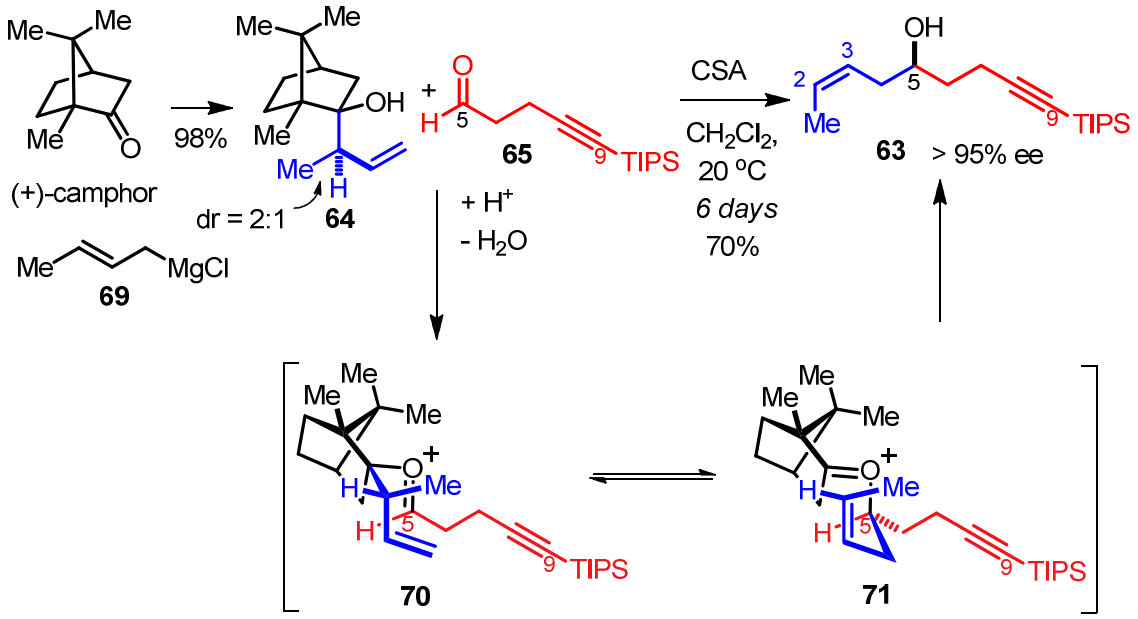
Scheme 12. Synthesis of aldehyde **65**



Reaction of aldehyde **65** with the allyl donor reagent **64**[42] obtained from (+)-camphor in a single step gave the *cis*-homoallylic alcohol **63** through a 2-oxonia-[3, 3]-Cope rearrangement (scheme 13).[39] Currently this method developed by Loh is the only literature method that produces the *cis*-homoallylic alcohol

through allyl transfer in a stereoselective manner, but with caveat that, it is a very slow reaction (6 days). This drawback became the driving force for developing a new methodology in the McDonald lab, which will be addressed in depth in chapter 3 of this thesis.

Scheme 13. Synthesis of the *cis*-homoallylic alcohol **63**

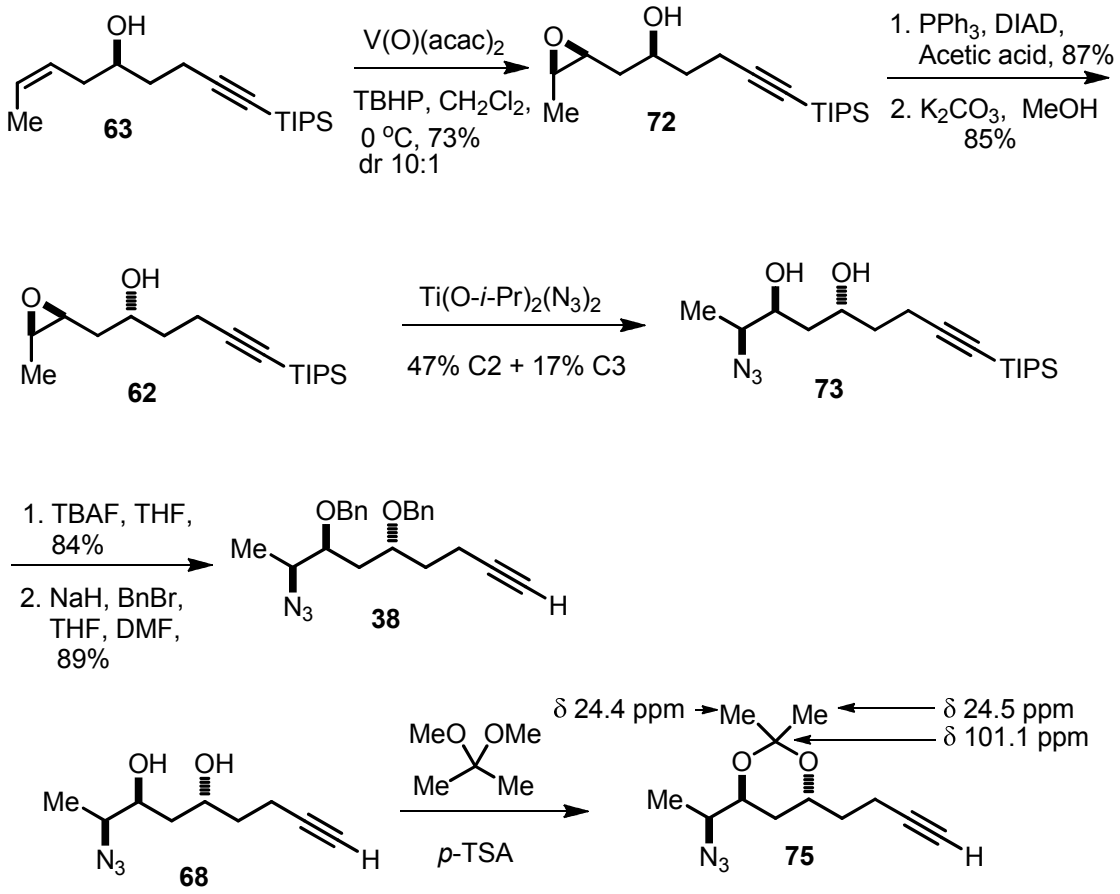


Vanadium catalyzed hydroxyl directed epoxidation[43] of **63** was followed by a Mitsunobu inversion to obtain compound **62** with the correct stereocenter at C5 (scheme 14).[44] The azide was then introduced with modest selectivity at the C2 position using the chelating reagent $Ti(O\text{-}i\text{-}Pr)_2(N_3)_2$.[45] The unwanted C3 regioisomer was removed by column chromatography at this stage. Many other methods were attempted to introduce the azide regioselectively, especially using chiral ligands, but all of these methods either failed to give good yield or regioselectivity.[46]

Removal of the TIPS group followed by benzyl protection of the diol gave left hand alkyne fragment **38**. To confirm the relative stereochemistry of the 3, 5-

diol, intermediate **68** was converted to acetonide **75** and the ^{13}C data for a *trans* diol agreed for such diols reported in the literature (scheme 14).[36]

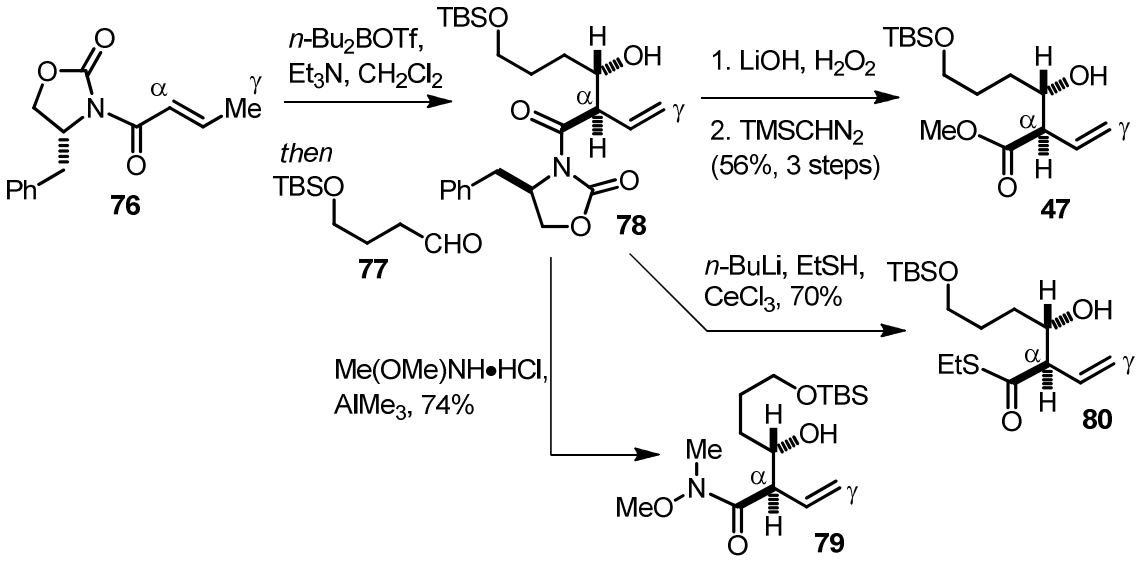
Scheme 14. Synthesis of left hand alkyne fragment **38**



1.5.2. Synthesis of synthon 47 for 2-oxonia-[3, 3]-Cope rearrangement

Based on the success of bispropionate transfer through the oxonia-Cope rearrangement, [30] a new synthon **47** was developed to carry out similar transformations (scheme 15). Starting from the chiral crotylimide **76**,[47] an aldol reaction of dibutylboryl enolate with aldehyde **77**[48] provided only α-adduct **78**. Intermediate **78** was then converted to either the methylester **47**,[49] thioester **80**,[50] and the Weinreb amide **79** (scheme 15).[51]

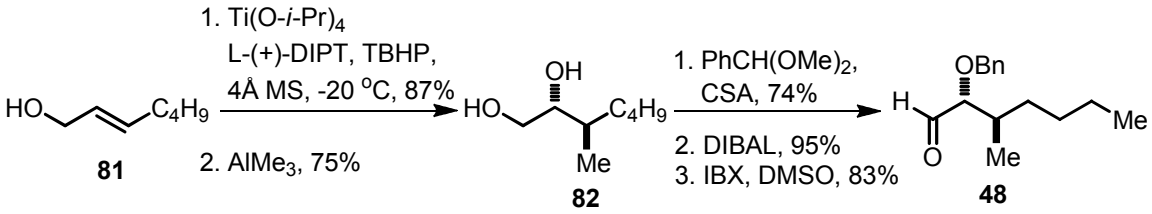
Scheme 15. Synthesis of synthon **47** required for 2-oxonia-[3, 3]-Cope rearrangement



1.5.3. Synthesis of aldehyde 48 for 2-oxonia-[3, 3]-Cope rearrangement

Sharpless asymmetric epoxidation (SAE) of *trans*-2-hepten-1-ol **(81)** using (+) diisopropyl tartrate (DIPT) gave the epoxide,[52] which was opened regio- and stereoselectively using $AlMe_3$ to give the desired diol **82** with the required stereochemistry at C3.[53] The diol **82** was then converted to the benzylidene acetal, which underwent regioselective reductive cleavage with DIBAL to give the primary alcohol, oxidation of which using IBX gave the desired aldehyde **48** without any observable epimerization product.

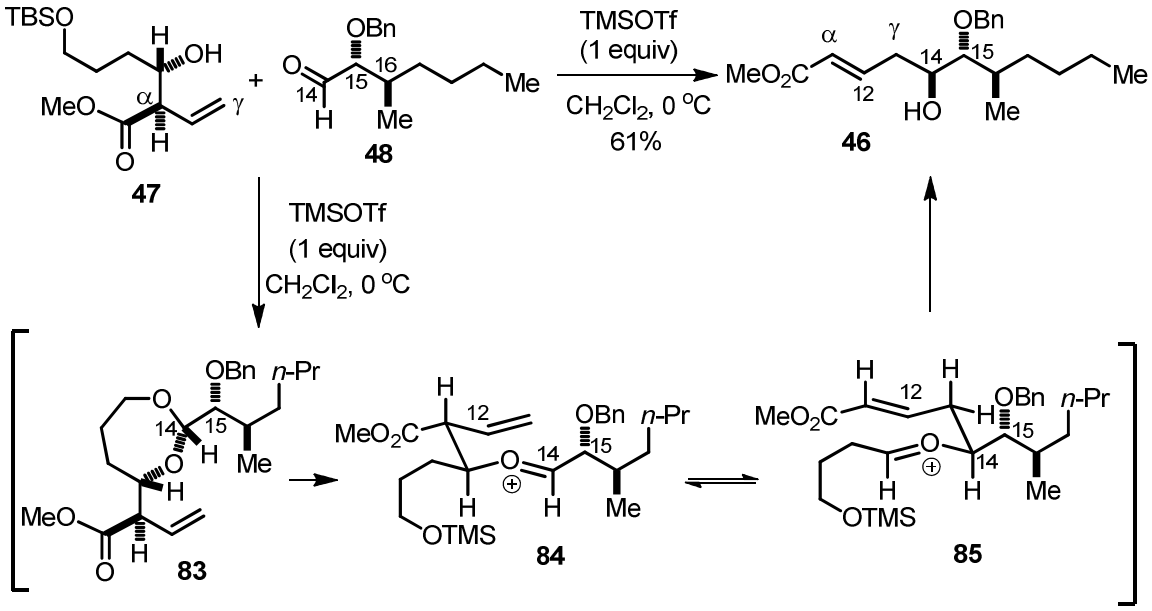
Scheme 16. Synthesis of the chiral aldehyde **48** from *trans*-2-hepten-1-ol



1.5.4. 2-oxonia-[3, 3]-Cope rearrangement studies

During the synthesis of *trans*-homoallylic alcohol **46**, Dr. Chen had established a protocol of mixing together the aldehyde **48** and synthon **47** using catalytic amount of TMSOTf to form the 7-membered ring acetal **83** followed by further addition of TMSOTf to give the rearranged product **46** (scheme 17).[29] This protocol proved inefficient and resulted in irreproducible yields. The procedure was therefore modified by carrying out the addition of 1 equiv of the Lewis acid in one go to obtain compound **46** in reproducible yields.

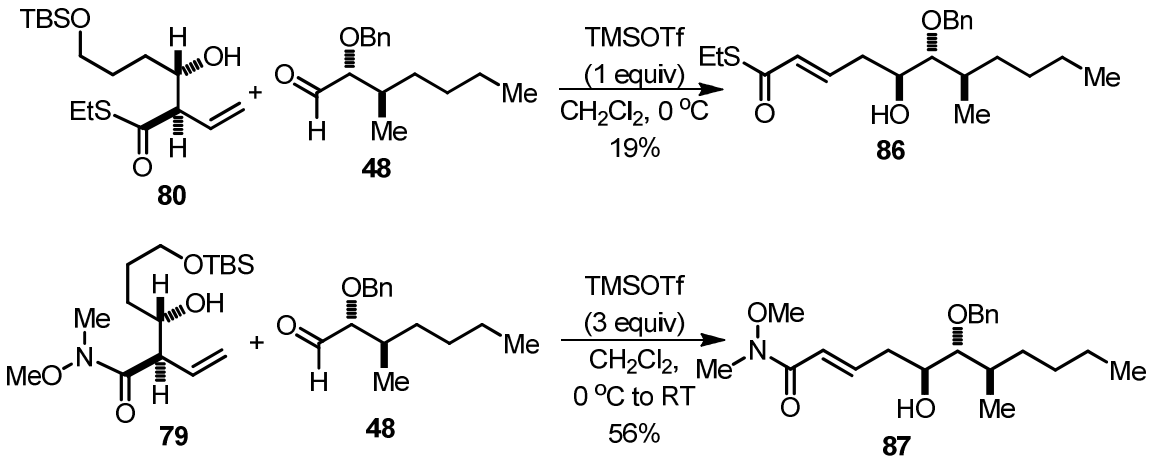
Scheme 17. Synthesis of *trans*-homoallylic alcohol **46** through 2-oxonia-[3, 3]-Cope rearrangement



The 2-oxonia-[3, 3]-Cope rearrangement also gave homoallylic alcohol **86** and **87** starting from synthon **80** and **79** respectively (scheme 18). The homoallylic alcohol **86** was made with an intention of carrying out the Michael addition without having to convert the methyl ester to the thioester as demonstrated by Dr. Chen.[29] However the difficultly of making the thioester

synthon **80** and the irreproducible rearrangement made us to abandon this approach. The homoallylic alcohol **87** was never carried further because of few literature precedent to carry out conjugate addition with Weinreb amides,[54] but nevertheless proved to be a good result for the rearrangement.

Scheme 18. *trans*-homoallylic alcohol **86** and **87** using 2-oxonia-[3, 3]-Cope rearrangement



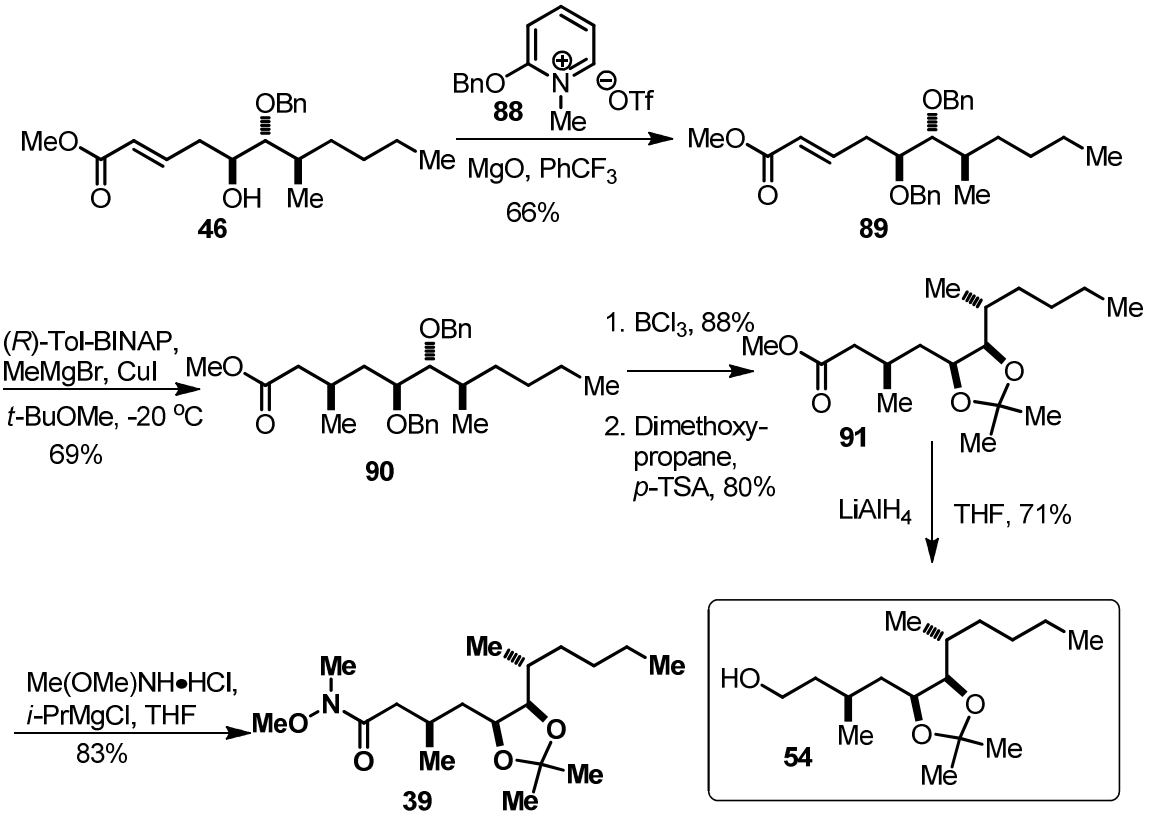
1.5.5. Synthesis of right hand fragment 39, the Weinreb amide

Attempted benzyl ether protection of homoallylic alcohol **46** using the method adopted by Dr. Chen using Tf_2O and BnOH proved problematic because of the observed epimerization by NMR (Dr. Chen had observed that the NMR spectra of the compound **89** were never clean after the benzyl protecting step).[29]

Using a recently developed mild and neutral method for benzylation by Dudley, [55] intermediate **89** was obtained without any epimerization (scheme 19). An asymmetric conjugate addition using methylmagnesium bromide following Loh's method gave methyl ester **90**, [38] thereby avoiding the need to convert the methyl ester to the thioester as demonstrated by Dr. Chen.[29] From here on the synthesis was similar to Dr. Chen's with benzyl deprotection of intermediate **90**

followed by acetonide protection to differentiate the two hydroxyls for the late stage coupling with tricarballylic acid fragment **40**. The methyl ester intermediate **91** was finally converted to Weinreb amide fragment **39** and to the primary alcohol **54**, which provided a spectroscopic correlation with an intermediate from Kishi's synthesis of fumonisin B_2 (scheme 19).[20]

Scheme 19. Synthesis of the Weinreb amide fragment **39**

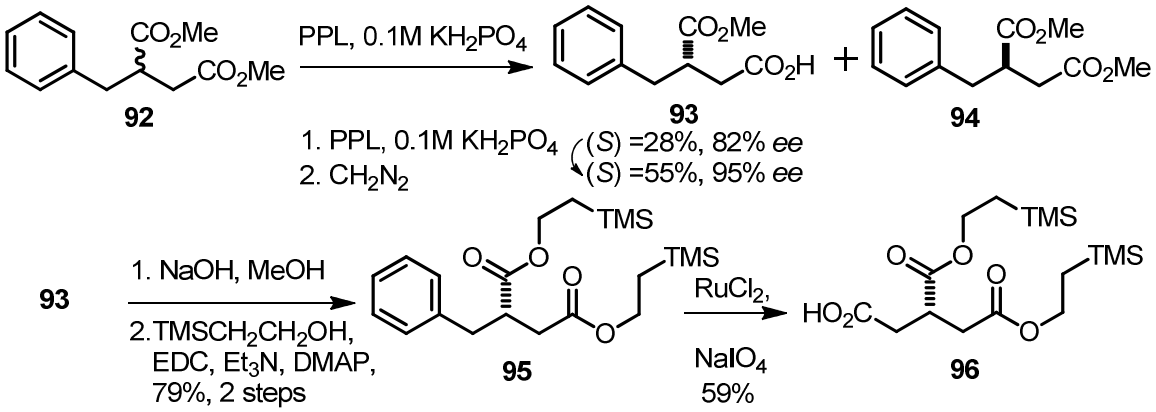


1.5.6. A new approach towards tricarballylic acid (TCA) fragment 40

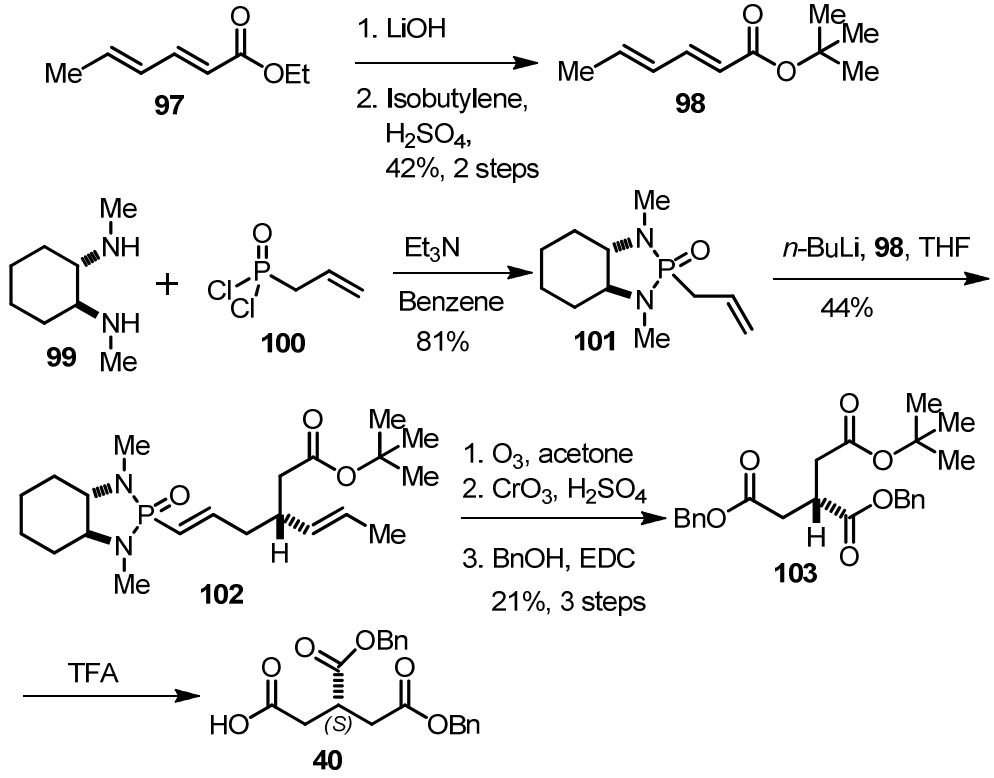
The TCA[56] fragment for Oikawa's AAL toxin TA_1 **(9)** synthesis was synthesized using kinetic resolution and multiple steps (scheme 20).[57] The *ee* of the obtained compound was 82% and so the process had to be repeated twice to obtain TCA fragment in 95% *ee*. For our initial synthesis we adopted the method

used by Kishi towards the synthesis of FB_2 (scheme 21).[20] This route was not efficient enough and involved multiple steps.

Scheme 20. TCA synthesis by Oikawa for AAL Toxin TA_1

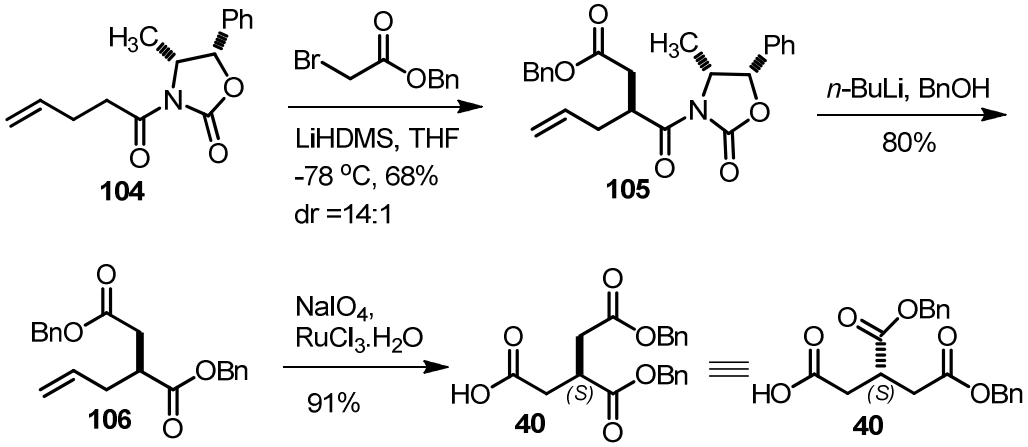


Scheme 21. Initial synthesis of TCA fragment **40** based on Kishi's approach



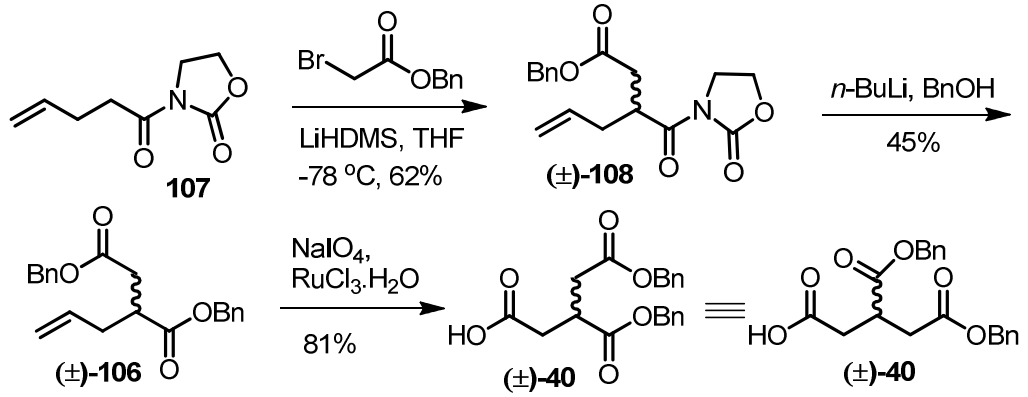
In this regard we opted for a more concise approach based on stereoselective enolate alkylation and alkene oxidation to carboxylic acid.[40, 56] From the chiral but-3-enoyloxazolidinone **104**, [57] intermediate **105** was obtained through an alkylation using the benzyl bromoacetate as the electrophile (scheme 22).[58] Removal of the auxillary and formation of the benzyl ester **106** in one pot followed by alkene oxidation using $RuCl_3/NaIO_4$ gave the TCA fragment **40**.[40]

Scheme 22. Stereoselective synthesis of TCA fragment **40**



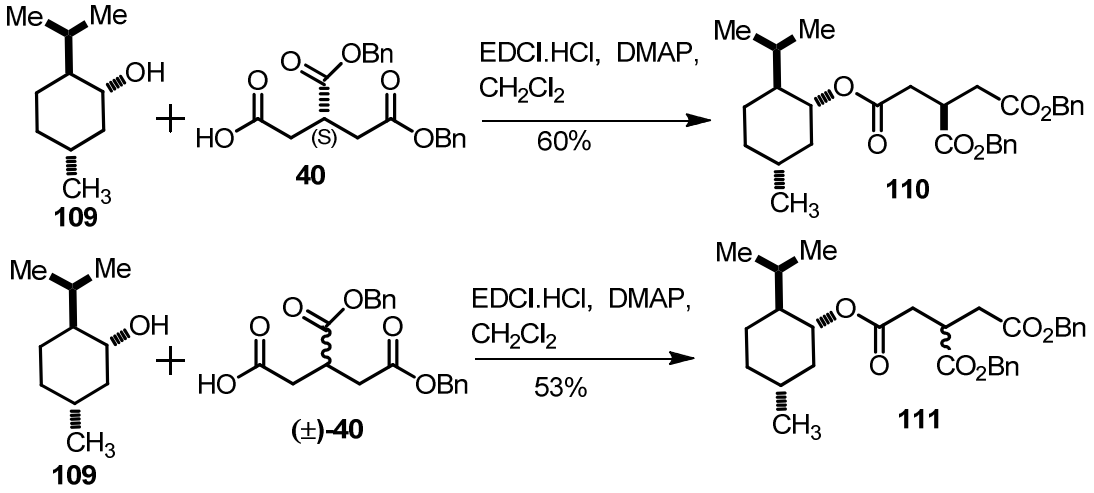
The ***rac*-40**, required for determining the *ee* of the asymmetric version was synthesized in a similar manner but starting from the achiral *N*-acyloxazolidinone **107** (scheme 23).

Scheme 23. Racemic synthesis of TCA fragment **40** from achiral material



The *ee* was determined by comparison of the menthyl ester **110** with an authentic sample of **111** using ^{1}H NMR and was found to be 13:1 in dr (scheme 24).[20]

Scheme 24. Determination of *ee* of TCA fragment **40** using (-)-menthol

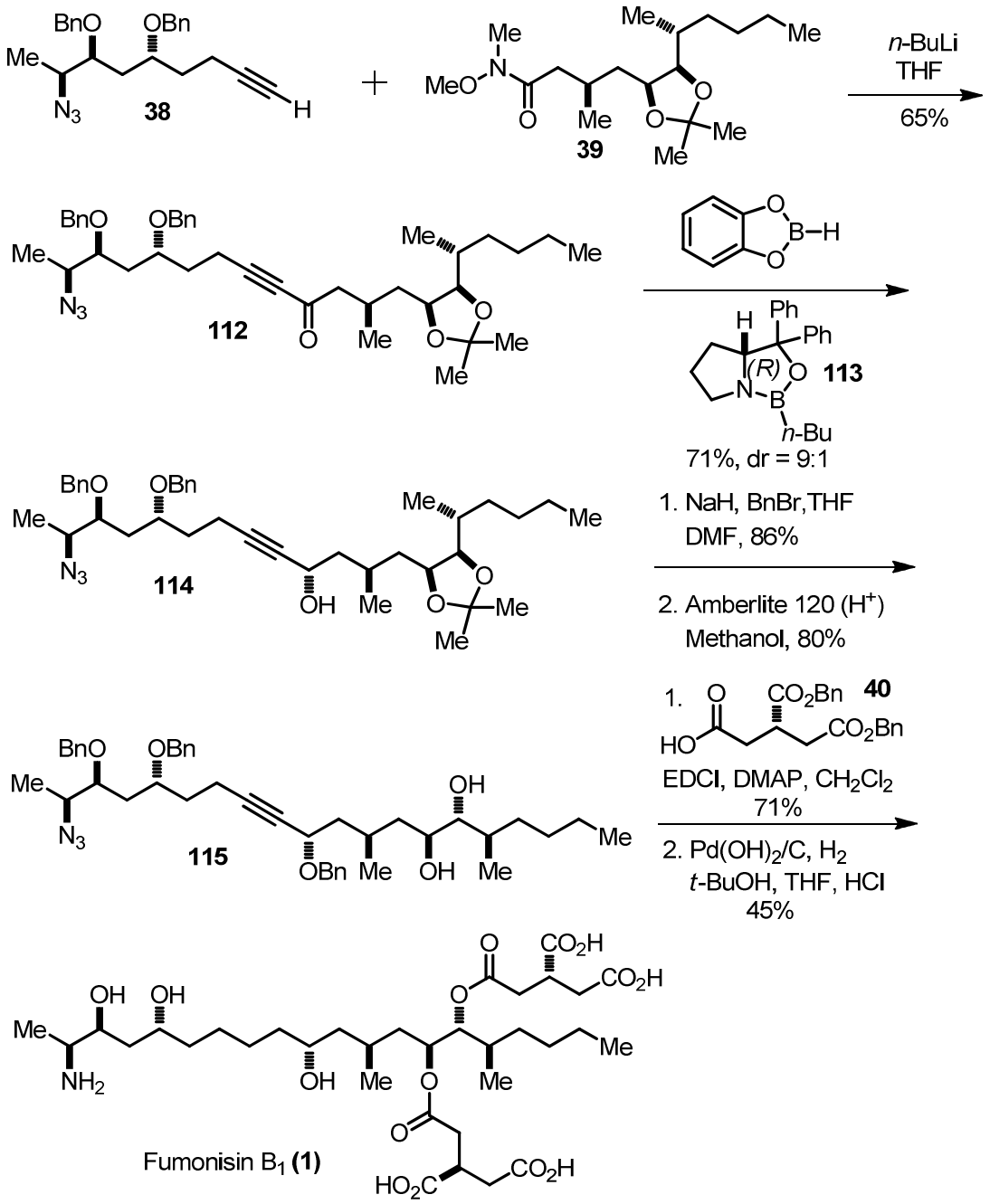


1.5.7. Coupling and completion of the total synthesis of fumonisin B_1 (1)

The 20 carbon chain framework of fumonisin B_1 was assembled from the lithium acetylide of left hand fragment **38** and Weinreb amide **39** (scheme 25). The alkynone **112** was then diastereoselectively reduced using the CBS method.[33c] (Mosher ester analysis of compound **50**, scheme 26 confirmed the absolute stereochemistry of the last set chiral center.) Benzyl protection followed by acid catalyzed acetonide cleavage afforded the diol **115**. Esterification of the two hydroxyl group with tricarballylic acid dibenzyl ester **40** using EDCI gave the fully protected fumonisin B_1.[20] A global hydrogenation of the azide, the alkyne, and the benzyl ethers and esters afforded fumonisin B_1 **(1)**.[59] The purification of this material proved challenging because of the issue of solubility. After many failed attempts of purification based on the procedures of isolation and

extraction,[60] purification on a C18 reverse phase column afforded the pure natural product after lyophilization.[61]

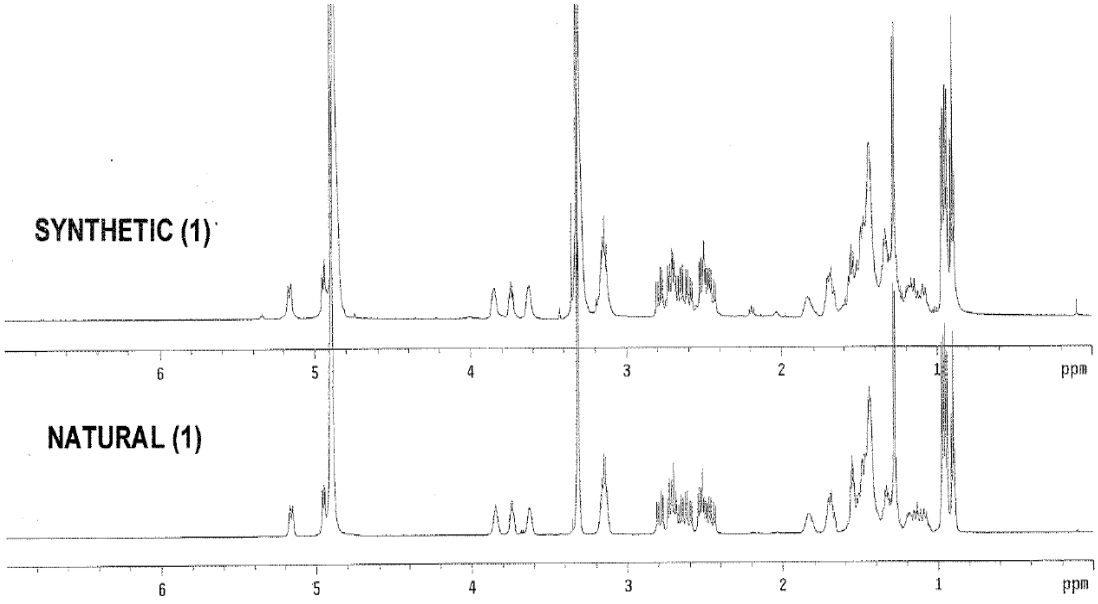
Scheme 25. Completion of the total synthesis of fumonisin B_1 **(1)**



The spectroscopic data of the synthetic FB_1 **(1)** was compared with the natural, obtained from Sigma Aldrich and was found to be a perfect match in

terms of ^{1}H, ^{13}C, mass, and optical rotation (figure 10). However the ^{1}H and ^{13}C were concentration dependent and this phenomenon has previously been studied and reported for fumonisin B_1.[23]

Figure 10. ^{1}H NMR comparison between natural and synthetic FB_1 **(1)**

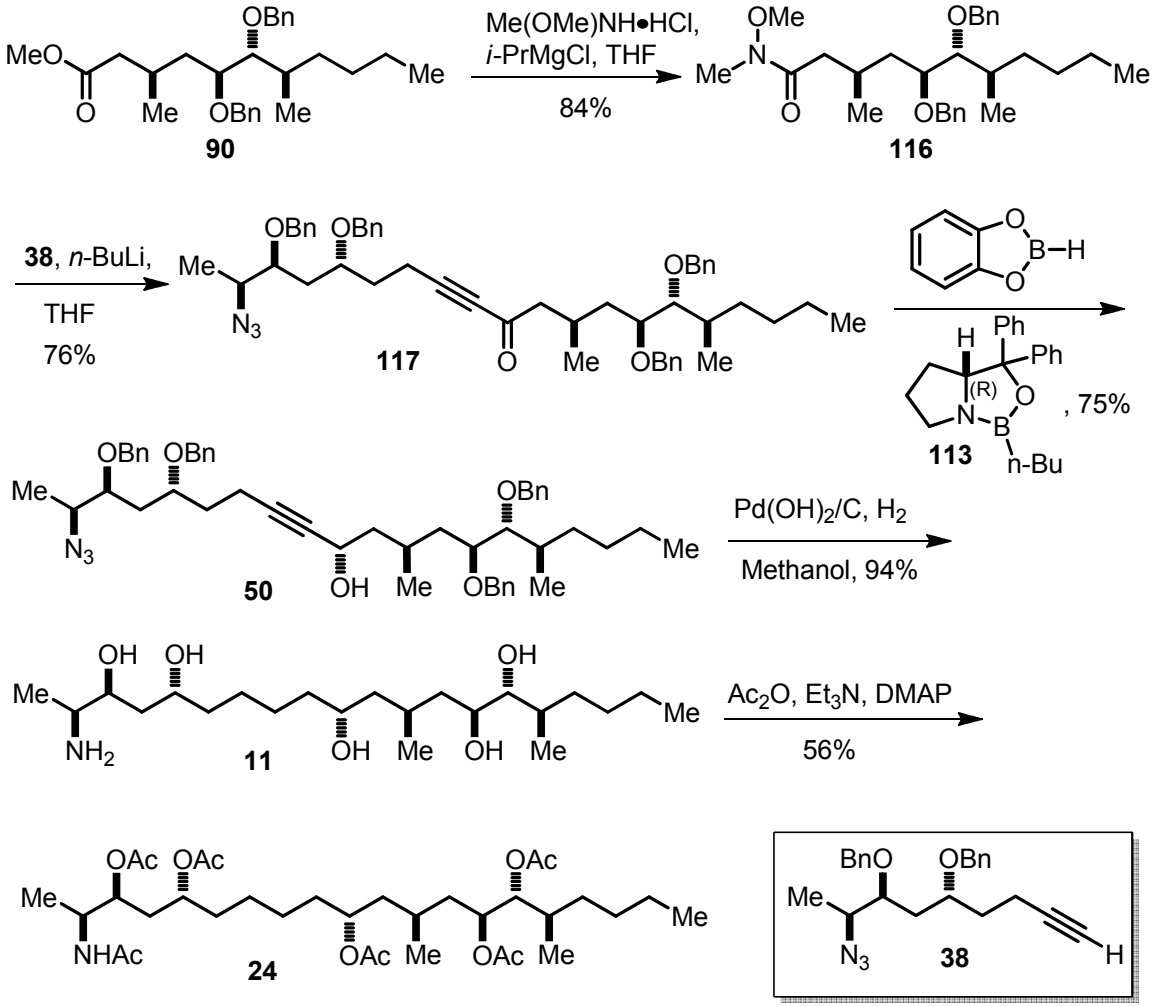


1.5.8. Towards the synthesis of FB_1-aminopentol (HFB_1) 11, and FB_1-hexaacetate 24

As mentioned previously the process of nixtamalization generated the hydrolyzed form of fumonisin B_1 (HFB_1) **(11)**.[10] Though the potency of inhibition of ceramide synthase was reduced, **11** was found to cause hepatic and renal lesion in feeding experiments in rats.[62] In studies of a possible mechanism to understand the toxicity of hydrolyzed fumonisins, HFB_1 **(11)** was found to be acylated by ceramide synthase to form the metabolite *N*-palmitoyl-aminopolyol, which was found to be highly toxic for HT29 cells.[62] In order to better understand the mechanism of action a pure synthetic HFB_1 **(11)** would be desirable and our synthetic strategy does allow access to this material.

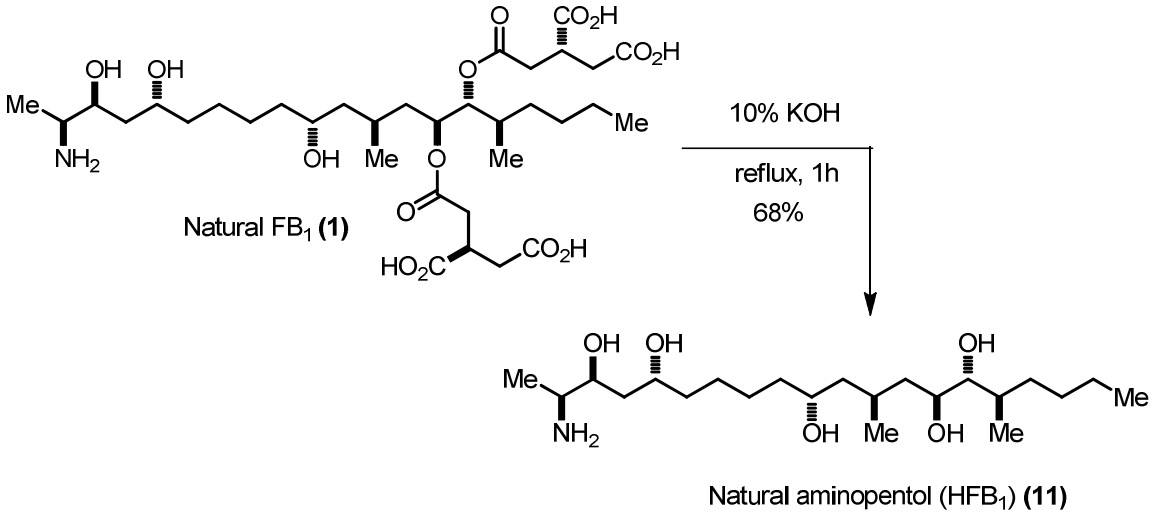
In this regard Weinreb amide **116** was synthesized from **90** (scheme 26). The 20 carbon framework of HFB_1 was then coupled from the lithium acetylide **38** and Weinreb amide **116**. Enantioselective alkynone reduction using (*R*)-CBS, followed by global hydrogenation provided HFB_1 **(11)**.[63] To make sure that the right CBS catalyst was used, a reduction carried out using the (*S*)-CBS-**113** catalyst followed by global hydrogenation and peracylation gave aminopentol with opposite stereochemistry at C10, which did not match spectroscopically to that reported in literature.[22]

Scheme 26. Synthesis of hydrolyzed FB_1 **(11)** and FB_1-hexaacetate **(24)**



HFB_1 **(11)** was then peracylated for further comparison with the known hexaacetyl derivative **24**.[22] The natural aminopentol **11** was obtained by the hydrolysis of natural FB_1 **(1)** obtained from Aldrich (scheme 27). All the spectroscopic data of both compounds match well with that obtained from the natural source. [63]

Scheme 27. Hydrolysis of natural FB_1 **(1)** to aminopentol HFB_1 **(11)**



1.6. Biological evaluation of synthetic fumonisin B_1 (1)

The synthetic fumonisin B_1 was sent for biological analysis to Prof. Alfred H. Merrill, Jr., one of the pioneers in the field of sphingolipid biology. There were two things to be established from these studies. 1) The synthetic FB_1 is a sphingolipid biosynthesis inhibitor, and 2) The absence of endotoxin in the synthetic material.

The biological data for endotoxin study using the synthetic FB_1 is given in table 1. As mentioned before TNF is considered to be a way in which endotoxins can be detected and measured.[27] It was found that the synthetic FB_1 **(1)** showed

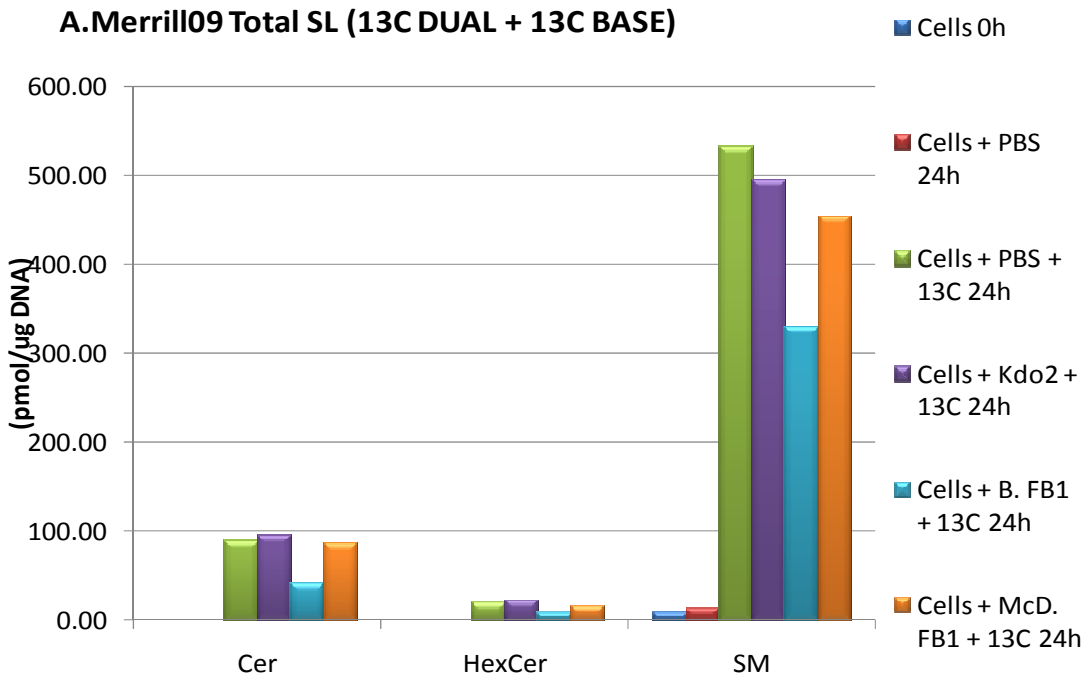
the same TNF values as the gold standard Promec sample of FB_1, indicating the complete absence of endotoxin in our synthetic sample.[64]

Table 1. Biological data for endotoxin study with synthetic FB_1 **(1)**

Cells incubated with	mTNF pg/mL
PBS only (control)	78
LM Kdo2 Lipid A (endotoxin)	19659
Promec FB_1	78
McDonald FB_1 (synthetic)	80

On the other hand the synthetic FB_1 showed only 50% inhibition of ^{13}C-palmitate incorporation to the sphingolipid base backbone of ceramide in RAW 264.7 cells when added at 0.1μM which is at the K_i for ceramide synthase (figure 11).[64]

Figure 11. Biological data of synthetic fumonisin B_1 **(1)**



This according to Merrill might be because of the problem associated with weighing of small amount of the synthetic material, (The amount a chemist indicates may not be accurate because of the probable contamination of the surface of the container with oil and dust) and thus affecting the concentration of the solution prepared and thus the results.

1.7. Conclusions

The first total synthesis of fumonisin B_1, the sphingolipid biosynthesis inhibitor was achieved. The synthesis demonstrated the role of 2-oxonia-[3, 3]-sigmatropic rearrangement in total synthesis. The synthesis also proved the assigned stereochemistry and the biological profile of the natural product.

1.8. Experimental details

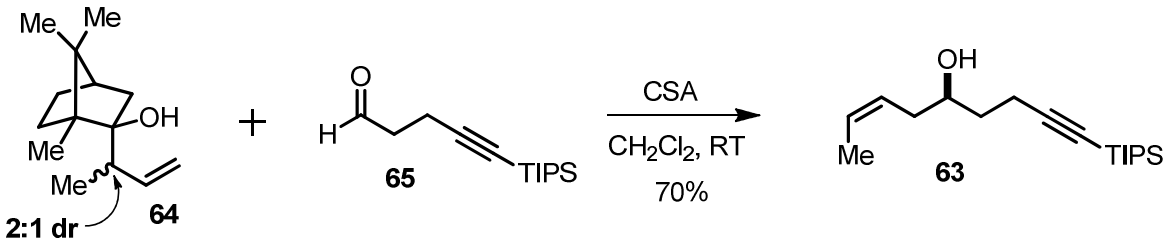
General information: ^{1}H NMR and ^{13}C NMR spectra were recorded on Varian INOVA 600, Unity 600 and INOVA 400 spectrometers. NMR spectra were recorded in solutions of deuterated chloroform ($CDCl_3$) with the residual chloroform (7.27 ppm for ^{1}H NMR and 77.23 ppm for ^{13}C NMR) taken as the internal standard, deuterated methanol (CD_3OD) with residual methanol (3.31 ppm for ^{1}H NMR and 49.3 ppm for ^{13}C NMR) taken as the internal standard, or deuterated benzene with residual benzene (7.16 ppm for ^{1}H NMR and 128.23 ppm for ^{13}C NMR) taken as the internal standard, and were reported in parts per million (ppm). Abbreviations for signal coupling are as follows: s, singlet; d, doublet; t, triplet; q, quartet; dd, doublet of doublet; ddd, doublet of doublet of doublet; dt, doublet of triplet; m, multiplet.

IR spectra were collected on a Mattson Genesis II FT-IR spectrometer as neat films on sodium chloride discs. Mass spectra (high resolution ESI and APCI) were recorded on a Finnigan LTQ FTMS Mass spectrometer. Optical rotations were measured using a Perkin-Elmer 341 polarimeter (concentration in g/100mL). Thin Layer Chromatography (TLC) was performed on precoated glass backed plates purchased from Whatman (silica gel $60F_{254}$; 0.25mm thickness). Flash column chromatography was carried out with silica gel 60 (230-400 mesh ASTM) from EM Science.

All reactions were carried out with anhydrous solvents in oven dried or flame dried and argon-charged glassware. All anhydrous solvents were dried with 4Å molecular sieves purchased from Sigma-Aldrich and tested for trace

water content with Coulometric KF titrator from Denver instruments. All solvents used in extraction procedures and chromatography were used as received from commercial suppliers without prior purification.

Synthesis of C1- C9 left hand fragment 38

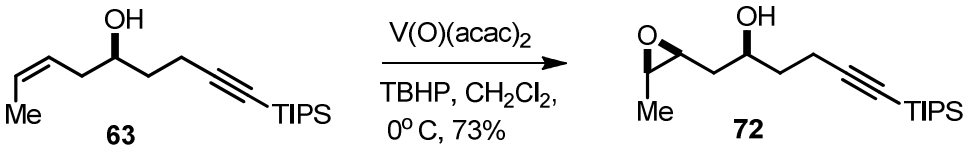


Synthesis of homoallylic alcohol 63: The camphor derivative **64** (8.6 g, 41.5 mmol) and aldehyde **65** (3.3 g, 13.8 mmol) were dissolved in CH_2Cl_2 (2.3 mL). CSA (0.32 g, 1.37 mmol) was then added and the reaction mixture was stirred at room temperature for 5-6 days. The reaction mixture was diluted with CH_2Cl_2, and the organic layer was dried over $MgSO_4$, filtered and concentrated to obtain colorless oil. The crude product was purified by flash chromatography using 6% ethyl acetate in hexanes as eluent to provide compound **63** as a colorless oil (2.83 g, 70%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.68-5.62 (m, 1H), 5.46-5.41 (m, 1H), 3.85-3.80 (m, 1H), 2.45-2.36 (m, 2H), 2.30-2.22 (m, 2H), 1.92 (d, 1H, *J* = 6.0 Hz), 1.76-1.66 (m, 2H), 1.64 (d, 3H, *J* = 6.6 Hz), 1.06-1.05 (m, 21H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 127.6, 126.0, 108.7, 81.1, 70.9, 35.7, 18.8, 16.8, 13.2, 11.4; **HRMS** (APCI): *m/z* calcd. for $C_{18}H_{35}OSi$ ($M+H^+$) 295.2451, found 295.2452; **FT-IR**: 3365, 3018, 2942, 2865, 2171, 1463, 1064, 883 cm^{-1}; **$[\alpha]_D^{25}$** = -3.8 (c = 0.745, $CHCl_3$). The enantioselectivity was determined to be > 95 : 5 er, by *in situ* formation of the Mosher esters of compound **63**. Specifically, an NMR tube

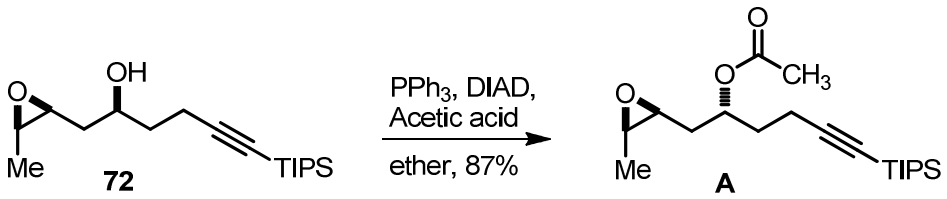
containing the alcohol **63** (ca.10 mg) and pyridine-d_5 (2 - 3 drops) was dissolved in $CDCl_3$ (ca. 0.5 mL), and 2 – 3 drops of (*S*)- or (*R*)-methoxy(trifluoromethyl)-phenylacetyl chloride (MTPA-Cl) were added. The tube was gently shaken and then allowed to stand overnight, to afford a solution of the (*R*)- or (*S*)-MTPA ester, respectively.

NMR data in $CDCl_3$ (400 MHz):

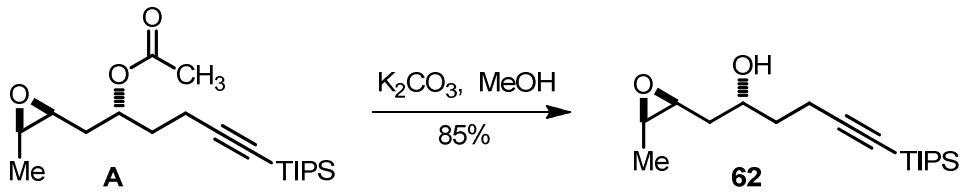
	Compound **63**	***R*-Mosher ester**	***S*-Mosher ester**
H1	1.64 (d)	1.53	1.57
H2	5.65 (app ddq)	5.52	5.58
H3	5.44 (app tq)	5.25	5.35
H4	2.41 (app t, 2H)	2.45	2.50
H4'		2.28	2.36
H5	3.83 (dddd)	5.25	5.29
H6	1.73 (m)	2.28 (2H)	2.14
H6'	1.69 (app q)		1.99
H7, H7'	2.26 (ddd, 2H)	1.81 (2H)	1.76 (2H)



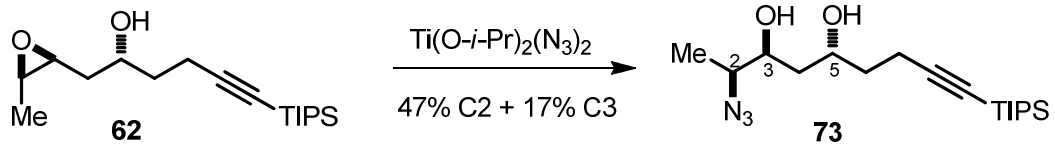
Synthesis of epoxide 72: Homoallylic alcohol **63** (2.8 g, 9.5 mmol) and $V(O)(acac)_2$ (0.12 g, 0.45 mmol) were dissolved in CH_2Cl_2 (100 mL) and the resulting solution was cooled to 0 ^oC. *tert*-Butyl hydroperoxide (TBHP, 5.5 M in decane, 2.6 mL, 14.2 mmol) was then added dropwise, and the reaction mixture was stirred at 0 ^oC for 3 h and then allowed to stir for another 24 h at room temperature. The reaction mixture was diluted with 10% $Na_2S_2O_3$ (10 mL). The organic layer was separated and washed with brine. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain brown oil. This crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **72** (2.16 g, 73%) as a colorless oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.08-4.04 (m, 1H), 3.09-3.06 (m, 1H), 3.05-3.0 (m, 1H), 2.52 (s, 1H), 2.42-2.34 (m, 2H), 1.78 (dt, 1H, *J* = 4.8, 14.8 Hz), 1.74-1.67 (m, 2H), 1.55 (dt, 1H, *J* = 14.4, 8.4 Hz), 1.24 (d, 3H, *J* = 6.0 Hz), 1.02-0.97 (m, 21H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 108.4, 81.3, 70.0, 55.2, 52.0, 36.2, 34.6, 18.7, 16.5, 13.5, 11.4; **HRMS** (APCI): *m/z* calcd. for $C_{18}H_{35}O_2Si$ ($M+H^+$) 311.2401, found 311.2402; **FT-IR**: 3430, 2942, 2865, 2171, 1463, 1074, 883 cm^{-1}; **$[\alpha]^{25}_D$** = -3.5 (c = 1.215, $CHCl_3$).



Synthesis of epoxyacetate A: Epoxy alcohol **72** (1.3 g, 4.18 mmol) was dissolved in ether (71 mL). Triphenylphosphine (2.19 g, 8.3 mmol) and acetic acid (0.47 mL, 8.3 mmol) were then added, and the reaction mixture was cooled to 0 ^{o}C. Diisopropyl azodicarboxylate (DIAD, 1.69 mL, 8.3 mmol) was then added, and the reaction mixture was stirred at 0 ^{o}C for 2 h. The reaction mixture was diluted with water (50 mL), and the aqueous layer was extracted with ether. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain yellow oil. This crude product was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to provide compound **A** (1.28 g, 87%) as a colorless oil. **^{1}H NMR** (400 MHz, $CDCl_3$) δ 5.17-5.13 (m, 1H), 3.06-3.02 (m, 1H), 2.99-2.95 (m, 1H), 2.38-2.28 (m, 2H), 2.06 (s, 3H), 1.93-1.87 (m, 2H), 1.86-1.78 (m, 2H), 1.26 (d, 3H, *J* = 6.0 Hz), 1.07-1.02 (m, 21H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.5, 107.4, 81.2, 71.3, 53.9, 52.5, 33.7, 32.6, 21.3 18.8, 16.3, 13.5, 11.4; **HRMS** (APCI): *m/z* calcd. for $C_{20}H_{37}O_3Si$ ($M+H^+$) 353.2506, found 353.2508; **FT-IR**: 2942, 2865, 2173, 1743, 1463, 1373, 1238, 1070, 1020, 883 cm^{-1}; **$[\alpha]_D^{25}$** = +23.0 (c = 0.96, $CHCl_3$).

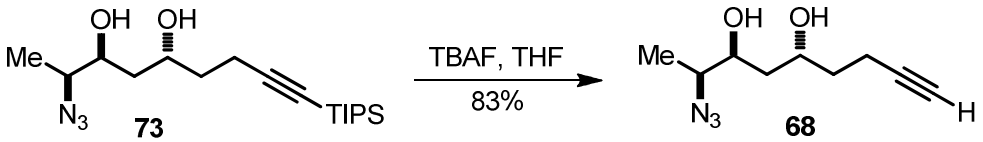


Synthesis of epoxyalcohol 62: Compound **A** (1.2 g, 3.68 mmol) was dissolved in methanol (12 mL). Potassium carbonate (0.25 g, 1.84 mmol) was then added and the reaction mixture was stirred at room temperature for 3 h. Methanol was removed in vacuum and the crude product was dissolved in water. The aqueous layer was then extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain oil. The crude product was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to provide compound **62** (0.85 g, 85%) as a colorless oil. **^{1}H NMR** (400 MHz, $CDCl_3$) δ 4.07-4.02 (m, 1H), 3.13-3.07 (m, 2H), 2.47-2.39 (m, 2H), 2.35 (d, 1H, *J* = 3.6 Hz), 1.84-1.70 (m, 3H), 1.61 (ddd, 1H, *J* = 2.8, 4.8, 9.4 Hz), 1.28 (d, 3H, *J* = 4.0 Hz), 1.08-1.00 (m, 21H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 108.4, 81.6, 69.7, 54.5, 52.8, 36.2, 34.9, 18.8, 16.7, 13.6, 11.4; **HRMS** (APCI): *m/z* calcd. for $C_{18}H_{35}O_2Si$ ($M+H^+$) 311.2400, found 311.2400; **FT-IR**: 3432, 2942, 2865, 2171, 1463, 1072, 883 cm^{-1}; **$[\alpha]^{25}_D$** = -6.0 (c = 1.40, $CHCl_3$).



Synthesis of azidodiol 73: Trimethylsilylazide (0.74 mL, 5.66 mmol) was added to a solution of titanium isopropoxide (0.83 mL, 2.83 mmol) in benzene (20 mL), and the solution was heated to 80 ^oC for 5 h to generate a solution of Ti(O-*i*-

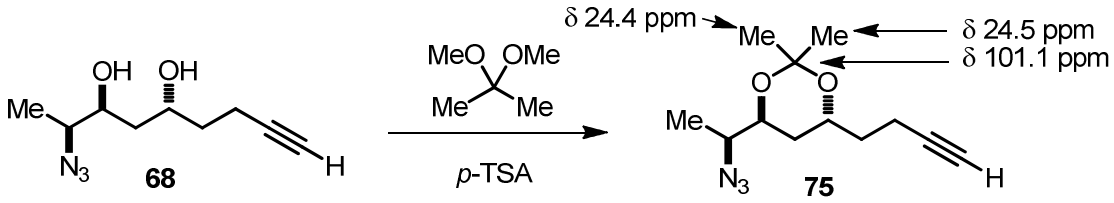
$Pr)_2(N_3)_2$. A solution of epoxyalcohol **62** (0.8 g, 2.57 mmol) in benzene (8 mL) was then added, and the reaction was stirred for 15 min before cooling to room temperature. Benzene was removed under vacuum and the crude product was diluted with ether. A solution of 5% H_2SO_4 was then added and the solution was stirred for 1 h at room temperature. The organic layer was separated and the aqueous layer was extracted with ether. The combined organic layers were dried over $MgSO_4$, filtered and concentrated to obtain brown oil. This crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **73** as oil (0.43 g, 47%). Also, 17% of the C-3 azide regioisomer was obtained. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.11-4.08 (m, 1H), 3.76-3.72 (m, 1H), 3.45-3.40 (m, 1H) 2.72-2.71 (m, 2H), 2.42-2.33 (m, 2H), 1.75-1.62 (m, 3H), 1.56 (ddd, 1H, *J* = 2.4, 8.7, 12.9 Hz), 1.27 (d, 3H, *J* = 7.2 Hz) 1.03-0.99 (m, 21H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 108.3, 81.8, 72.1, 68.7, 62.5, 39.6, 36.0, 18.8, 16.8, 15.8, 11.4; **HRMS** (APCI): *m/z* calcd. for $C_{18}H_{36}N_3O_2Si$ ($M+H^+$) 354.2571, found 354.2572; **FT-IR**: 3367, 2942, 2865, 2171, 2111, 1463, 1257, 1070, 883 cm^{-1}; **$[\alpha]_D^{25}$** = +16.5 (c = 1.6, $CHCl_3$).



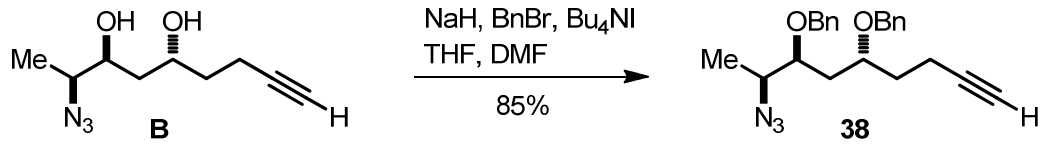
Synthesis of azidodiol - alkyne 68: Compound **73** (0.35 g, 0.98 mmol) was dissolved in THF (1.5 mL) and cooled to 0 °C. TBAF (1.29 g, 4.9 mmol, 1.0 M in THF) was then added and the reaction mixture was stirred at room temperature for 6 h. The reaction mixture was diluted with water and the aqueous layer was

extracted with ethyl acetate. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain oil. The crude product was purified by flash chromatography using 25% ethyl acetate in hexanes as eluent to provide compound **68** (0.16 g, 84%) as oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.12-4.08 (m, 1H), 3.8-3.5 (m, 1H), 3.49-3.45 (m, 1H), 2.80 (s, 1H), 2.73 (s, 1H), 2.35 (dt, 2H, *J* = 2.4, 6.9, 13.8 Hz), 2.0 (t, 1H, *J* = 1.8, 4.8 Hz), 1.77-1.65 (m, 3H), 1.59 (ddd, 1H, *J* = 2.4, 8.7, 13.8 Hz), 1.31 (d, 3H, *J* = 6.0 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 84.0, 72.0, 69.3, 67.9, 62.5, 39.6, 35.7, 15.8, 15.2; **HRMS** (APCI): *m/z* calcd. for $C_9H_{16}N_3O_2$ ($M+H^+$) 198.1237, found 198.1235; **FT-IR**: 3390, 2948, 2919, 2117, 1444, 1263, 1068 cm^{-1}; **$[\alpha]_D^{25}$** = +36.6 (c = 0.89, $CHCl_3$).

The relative stereochemistry of the diol **68** was confirmed by formation of the acetonide **75** and evaluation of the ^{13}C NMR chemical shifts, specifically the two methyls at 25 ± 1 ppm (24.5, 24.4) and the ketal carbon at ≥ 100 ppm (101.1):

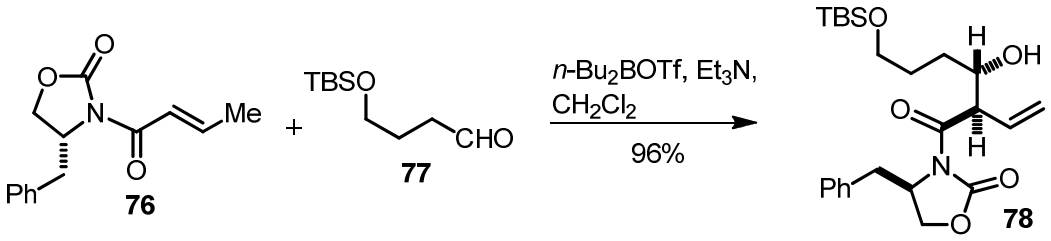


Acetonide 75: ^{1}H NMR (400 MHz, $CDCl_3$) δ 4.00-3.93 (m, 1H), 3.76 (ddd, 1H, *J* = 6.8, 9.2 Hz), 3.38 (q, 1H, *J* = 6.8 Hz), 2.30 (ddd, 2H, *J* = 2.8, 6.8 Hz), 1.94 (t, 1H, *J* = 2.8 Hz), 1.79-1.72 (m, 1H), 1.71-1.65 (m, 2H), 1.59-1.47 (m, 1H), 1.38 (s, 6H), 1.17 (d, 3H, *J* = 6.8 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 101.1, 83.9, 70.8, 68.7, 65.3, 60.1, 35.6, 34.3, 24.5, 24.4, 15.3, 14.8; **FT-IR**: 3303, 2987, 2939, 2111, 1380, 1224 cm^{-1}; **$[\alpha]_D^{25}$** = -9.1 (c = 0.815, $CHCl_3$).



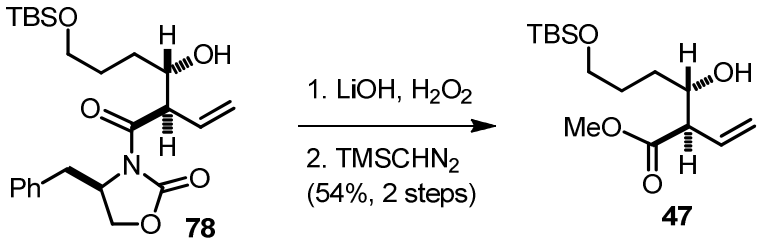
Synthesis of dibenzyl ether - alkyne 38: Azidodiol - alkyne **B** (0.14 g, 0.7 mmol) was dissolved in a mixture of THF (1.5 mL) and DMF (0.15 mL). Tetrabutylammonium iodide (0.01 g, 0.027 mmol) was then added and the reaction mixture was cooled to 0 ^{o}C. NaH (0.085 g, 2.12 mmol, 60% in mineral oil) was then added and the reaction mixture was stirred at room temperature for 30 min. Benzyl bromide (0.36 g, 2.12 mmol) was then added and the reaction mixture was stirred at room temperature for 15 h. The reaction mixture was quenched with water, and the aqueous layer was extracted with ether. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain crude **38** as oil. The crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **38** as oil (0.22 g, 85%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.35-7.29 (m, 10H), 4.60 (d, 1H, *J* = 8.4 Hz), 4.58 (d, 1H, *J* = 9.6 Hz), 4.40 (d, 1H, *J* = 11.4 Hz), 4.36 (d, 1H, *J* = 11.4 Hz), 3.79-3.75 (m, 1H), 3.61-3.57 (m, 2H), 2.37-2.26 (m, 2H), 1.99 (t, 1H, *J* = 3.0 Hz), 1.90-1.80 (m, 2H), 1.78-1.65 (m, 2H), 1.28 (d, 3H, *J* = 6.6); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.6, 138.3, 128.6, 128.5, 128.0, 127.9, 127.9, 127.8, 84.3, 78.7, 74.3, 73.3, 70.9, 68.8, 59.8, 36.2, 33.0, 15.0, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{23}H_{28}N_3O_2$ ($M+H^+$) 378.2176, found 378.2176; **FT-IR**: 3301, 3031, 2929, 2871, 2107, 1454, 1257, 1064, 736 cm^{-1}; **$[\alpha]_D^{25}$** = -36.2 (c = 0.63, $CHCl_3$).

Synthesis of C10-C20 right hand fragment



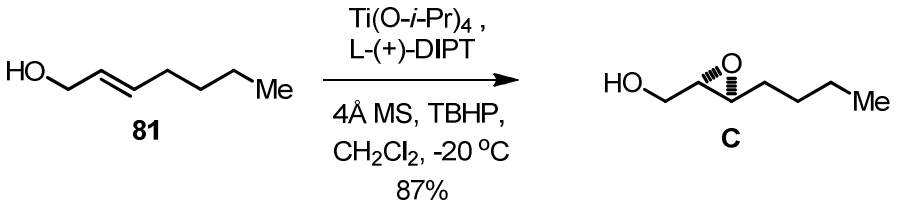
Synthesis of aldol product 78: To a stirred solution of imide **76** (7.6 g, 37.5 mmol) in CH_2Cl_2 (110 mL) was added Bu_2BOTf (27.8 mL, 1.0 M solution in CH_2Cl_2, 27.7 mmol) at -78 ^oC. The mixture was stirred for 5 min and then treated with freshly distilled triethylamine (4.9 mL, 35.3 mmol). After 1 h at -78 ^oC and 15 min at 0 ^oC, the solution was recooled to -78 ^oC and treated with freshly prepared aldehyde **77** (6.2 g, 25.2 mmol). The solution was kept for 2 h at -78 ^oC and 1.5 h at 0 ^oC, and then partitioned between NH_4Cl and a 1 : 1 mixture of ethyl acetate and hexanes. The organic phase was washed with brine and concentrated to an oil, which was dissolved in ether (50 mL) and cooled to 0 ^oC. The resulting solution was treated with pH 7 buffer (10 mL) and 30% H_2O_2 (10 mL) at 0 ^oC. After stirring rapidly for 1 h the mixture was partitioned between water and a 1 : 1 mixture of ethyl acetate and hexanes. The organic phase was washed with $NaHCO_3$ and brine, dried over $MgSO_4$ and concentrated to afford the crude product as yellow oil. Purification by flash column chromatography using 15% ethyl acetate in hexanes as eluent gave alcohol **78** (10.5 g, 96%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.34-7.28 (m, 3H), 7.22-7.19 (m, 2H), 6.05 (ddd, 1H, *J* = 9.6, 17.4 Hz), 5.40-5.37 (m, 2H), 4.73-4.70 (m, 1H), 4.55 (dd, 1H, *J* = 3.6, 8.4 Hz), 4.22-4.16 (m, 2H), 4.03-4.02 (m, 1H), 3.65 (t, 2H, *J* = 6.0 Hz), 3.36 (d, 1H, *J* = 2.4 Hz), 3.26 (dd, 1H, *J* = 3.0, 13.2 Hz), 2.76 (dd, 1H, *J* = 10.2, 16.5 Hz), 1.72-1.54 (m,

4H), 0.89 (s, 9H), 0.05 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 174.1, 153.1, 135.2, 131.7, 129.6, 129.1, 127.6, 121.3, 71.8, 66.1, 63.3, 55.3, 52.7, 37.7, 31.3, 29.2, 26.1, 18.5, -5.1; **HRMS** (APCI): *m/z* calcd. for $C_{24}H_{38}O_5NSi$ (M^+) 448.2513, found 448.2520; **FT-IR**: 3521, 3029, 2952, 2929, 2858, 1781, 1697, 1386, 1359, 1209, 1101, 836 cm^{-1}; **$[\alpha]_D^{25}$** = -10.4 (c = 1.37, $CHCl_3$).



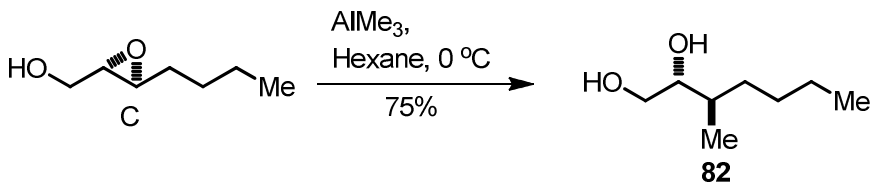
Synthesis of homoallylic alcohol synthon 47: A solution of imide **78** (7.50 g, 16.8 mmol) in dioxane (148 mL) was cooled to 0 °C, and H_2O_2 (23.6 mL, 30% in water, 201 mmol) and LiOH (1.41 g, 58.6 mmol) were added at 0 °C. The resulting solution was stirred at 0 °C for 1 h and at room temperature for 1.5 h, and then partitioned between saturated NH_4Cl and CH_2Cl_2. The aqueous layer was acidified to pH = 3 and then extracted with ethyl acetate. The organic layer was dried over $MgSO_4$, filtered, and concentrated. The crude acid was then dissolved in benzene (180 mL) and methanol (45 mL), and (trimethylsilyl)-diazomethane (20.9 mL, 2 M in diethyl ether, 42 mmol) was added *via* syringe at room temperature. The resulting solution was stirred at room temperature for 2 h, and concentrated to afford the crude product as yellow oil. Purification by flash column chromatography using 10% ethyl acetate in hexanes as eluent provided compound **47** (2.7 g, 54%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 5.96 (ddd, 1H, *J* = 9.2, 10.2, 17.2 Hz), 5.31-5.20 (m, 2H), 3.98-3.93 (m, 1H), 3.72 (s, 3H), 3.65 (t, 2H, *J* = 6.0 Hz), 3.13 (d, 1H, *J* = 3.6 Hz), 3.09 (dd, 1H, *J* = 4.8, 9.2 Hz),

1.72-1.45 (m, 4H), 0.89 (s, 9H), 0.05 (s, 6H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 173.8, 132.2, 120.1, 71.5, 63.3, 56.1, 52.1, 31.4, 29.1, 26.0, 18.4, -5.2; **HRMS** (APCI): *m/z* calcd. for $C_{15}H_{31}O_4Si$ ($M+H^+$) 303.1986, found 303.1990; **FT-IR**: 3455, 3081, 2952, 2858, 1737, 1471, 1255, 1099 cm^{-1}; **$[\alpha]_D^{25}$** = +29.9 (c = 1.28, $CHCl_3$).



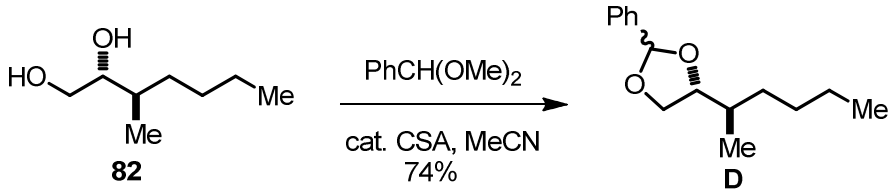
Synthesis of epoxyalcohol C: L-(+)-Diisopropyl tartrate (1.2 g, 5.25 mmol) and titanium tetraisopropoxide (1.2 g, 4.37 mmol) were added to a suspension of 4 Å molecular sieves (12.5 g) in CH_2Cl_2 (150 mL) at -20 °C, and the solution was stirred for 15 min. The resulting solution was treated with *tert*-Butyl hydroperoxide (15.9 mL, 5.5 M in decane, 87.6 mmol) at -20 °C and stirred for another 30 min before the addition of *trans*-2-heptene-1-ol (**81**, 5.0 g, 43.7 mmol). The reaction mixture was stirred at -20 °C for 12 h, and then quenched with citric acid. The mixture was diluted with ether and acetone, stirred at 0 °C for 30 min, and filtered through celite. The crude product was purified by flash chromatography using 15% ethyl acetate in hexanes as eluent to provide compound **C** as oil (5.0 g, 87%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.87 (ddd, 1H, *J* = 3.0, 5.7, 12.9 Hz), 3.59 (ddd, 1H, *J* = 4.8, 7.2, 12.3 Hz), 2.92 (dt, 1H, *J* = 2.4, 5.7), 2.89-2.88 (m, 1H), 1.77 (t, 1H, *J* = 6.6 Hz), 1.56-1.52 (m, 2H), 1.45-1.29 (m, 4H), 0.92 (t, 3H, *J* = 6.6 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 61.8, 58.6, 56.1, 31.4, 28.2, 22.6, 14.1; **HRMS**

(APCI): *m/z* calcd. for $C_7H_{15}O_2$ ($M+H^+$) 131.1066, found 131.1066; **FT-IR**: 3413, 2958, 2931, 2861, 1467, 1022 cm^{-1}; $[\alpha]_D^{25}$ = -36.0 (c = 0.29, $CHCl_3$).

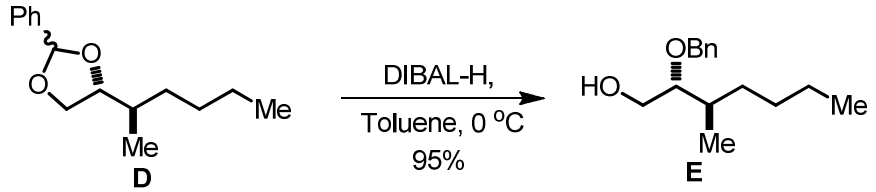


82

Synthesis of diol 82: Epoxy alcohol **C** (5.50 g, 42.2 mmol) in anhydrous hexanes (54 mL) was added slowly to a solution of trimethylaluminum (54 mL, 1.0 M in hexanes, 107 mmol) at 0 °C. After stirring for 1 h at 0 °C, the resulting mixture was diluted with CH_2Cl_2, treated with NaF (47 g) and water. Vigorous stirring of the resulting suspension was continued at room temperature for 30 min. The semi-solid was filtered through celite. Extractions were performed with ethyl acetate, and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **82** as oil (4.7 g, 75%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 3.74-3.67 (m, 1H), 3.54-3.47 (m, 2H), 2.41 (bs, 1H), 2.29 (bs, 1H), 1.60-1.49 (m, 2H), 1.42-1.10 (m, 5H), 0.90 (t, 3H, *J* = 7.2 Hz), 0.88 (d, 3H, *J* = 6.8 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 76.4, 64.8, 36.3, 32.3, 29.3, 23.1, 15.3, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_8H_{19}O_2$ ($M+H^+$) 147.1379, found 147.1378; **FT-IR**: 3390, 2958, 2929, 2873, 1463, 1072 cm^{-1}; $[\alpha]_D^{25}$ = +10.7 (c = 0.985, $CHCl_3$).

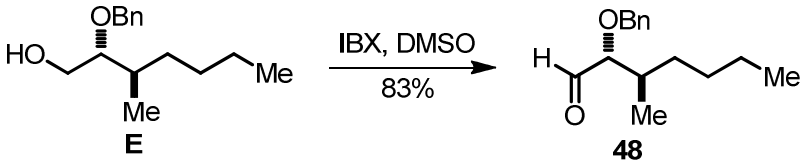


Synthesis of benzylidene acetal D: The diol **82** (4.7 g, 32.1 mmol) was dissolved in acetonitrile (94 mL). Benzaldehyde dimethyl acetal (7.3 g, 48.2 mmol) and CSA (0.38 g, 1.6 mmol) were added at 0 ^{o}C. The reaction mixture was stirred at 0 ^{o}C for 30 min followed by room temperature for 1 h. The reaction was quenched with water and the extractions were performed with ethyl acetate. The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by flash chromatography using 2% ethyl acetate in hexanes as eluent to provide compound **D** as oil (5.5 g, 74%). ^{1}H NMR analysis of the product revealed ~1 : 1 dr at the acetal center.



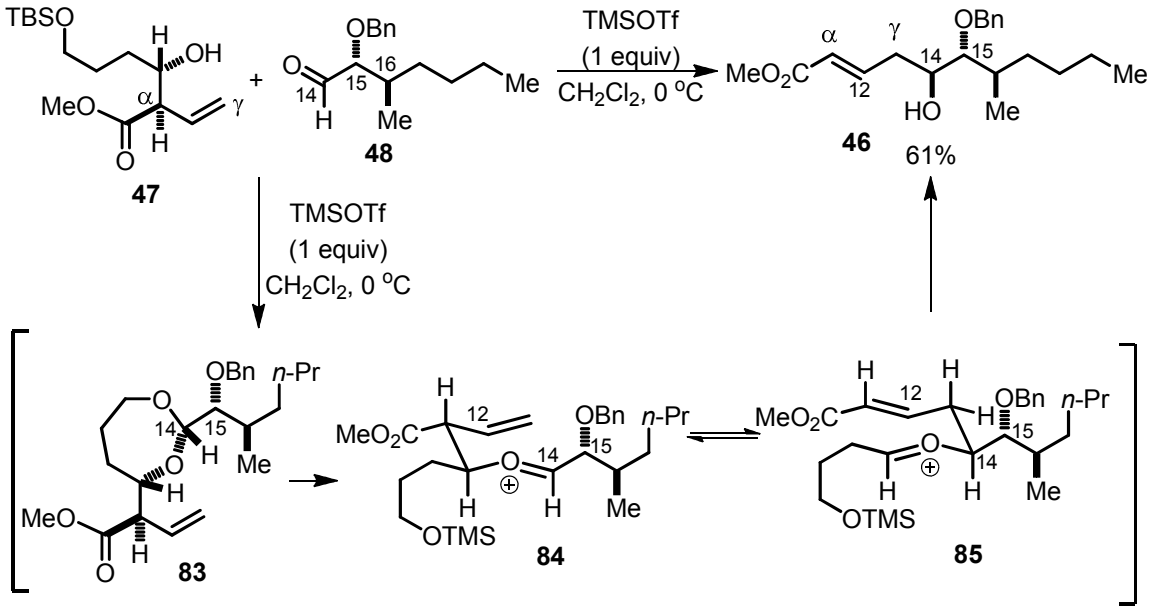
Synthesis of primary alcohol E: The benzylidene acetal **D** (5.5 g, 23.0 mmol) was dissolved in toluene (130 mL) and cooled to 0 ^{o}C. DIBAL-H (117.0 mL, 1.0 M in CH_2Cl_2, 117 mmol) was added *via* syringe pump over 1 h at 0 ^{o}C. The resulting solution was further stirred at 0 ^{o}C for 2 h, and then quenched with ethyl acetate and saturated aqueous Rochelle's salt. Vigorous stirring of the resulting mixture was continued at room temperature for 20 min. Extractions were performed with ethyl acetate, and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by flash

chromatography using 5% ethyl acetate in hexanes as eluent to provide compound **E** as oil (5.27 g, 95%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.37-7.29 (m, 5H), 4.64 (d, 1H, *J* = 12.0 Hz), 4.53 (d, 1H, *J* = 11.4 Hz), 3.69 (d, 1H, *J* = 9.0 Hz), 3.62 (dd, 1H, *J* = 6.6, 11.7 Hz), 3.38 (ddd, 1H, *J* = 3.0, 6.9 Hz), 2.01 (bs, 1H), 1.91-1.85 (m, 1H), 1.50-1.45 (m, 1H), 1.40-1.22 (m, 4H), 1.20-1.13 (m, 1H), 0.92 (d, 3H, *J* = 7.2 Hz), 0.91 (t, 3H, *J* = 6.6, 7.2 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.7, 128.6, 128.0, 127.9, 84.0, 72.1, 61.6, 33.8, 32.7, 29.7, 23.1, 14.9, 14.2; **HRMS** (APCI): *m/z* calcd. for $C_{15}H_{25}O_2$ ($M+H^+$) 237.1849, found 237.1849; **FT-IR**: 3426, 3064, 3031, 2956, 2929, 2871, 1454, 1378, 1091 cm^{-1}; **$[\alpha]_D^{25}$** = -11.0 (c = 1.17, $CHCl_3$).



Synthesis of aldehyde 48: IBX (3.6 g, 12.9 mmol) was dissolved in DMSO (12 mL). A solution of alcohol **E** (1.8 g, 7.61 mmol) in DMSO (6.0 mL) was then added at room temperature. After stirring for 2 h at room temperature, the reaction was quenched with water. The precipitate was filtered off and the residue stirred with ether. Additional precipitate was filtered, and the organic layer was dried over $MgSO_4$, filtered, and concentrated. The aldehyde **48** was used for the next step without further purification (1.47 g, 83%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 9.68 (d, 1H, *J* = 2.8 Hz), 7.39-7.29 (m, 5H), 4.69 (d, 1H, *J* = 11.6 Hz), 4.50 (d, 1H, *J* = 12.0 Hz), 3.55 (dd, 1H, *J* = 3.2, 5.6 Hz), 2.01-1.92 (m, 1H), 1.56-1.47 (m, 1H), 1.33-1.17 (m, 5H), 0.97 (d, 3H, *J* = 7.2 Hz), 0.89 (t, 3H, *J* = 6.8 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 204.8, 137.6, 128.6, 128.1, 87.6, 72.9, 35.2,

31.6, 29.3, 22.9, 15.7, 14.2; **HRMS** (APCI): *m/z* calcd. for $C_{15}H_{23}O_2$ ($M+H^+$) 235.1692, found 235.1691; **FT-IR**: 3033, 2958, 2933, 2873, 1706, 1456, 1286 cm^{-1}.



Synthesis of hydroxy alkenyl ester 46: Alkenyl alcohol **47** (1.17 g, 3.88 mmol) and aldehyde **48** (0.7 g, 2.98 mmol) were dissolved in CH_2Cl_2 (28 mL), and cooled to 0 °C. TMSOTf (0.7 g, 3.28 mmol) was added dropwise. The resulting solution was stirred at 0 °C for 1 h before quenching with saturated aqueous $NaHCO_3$ at 0 °C. The resulting solution was stirred at room temperature for another 15 min. Extractions were performed with CH_2Cl_2. The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by flash chromatography using 15% ethyl acetate in hexanes as eluent to provide compound **46** as oil (0.61 g, 61%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.38-7.34 (m, 4H), 7.33-7.29 (m, 1H), 7.05 (ddd, 1H, *J* = 7.5, 15.6 Hz), 5.93 (d, 1H, *J* = 17.4 Hz), 4.69 (d, 1H, *J* = 11.4 Hz), 4.62 (d, 1H, *J* = 11.4 Hz), 3.91-3.90 (m, 1H), 3.73 (s, 3H), 3.29 (dd, 1H, *J* = 4.8, 5.4 Hz), 2.54-2.50 (m, 1H), 2.44-2.38

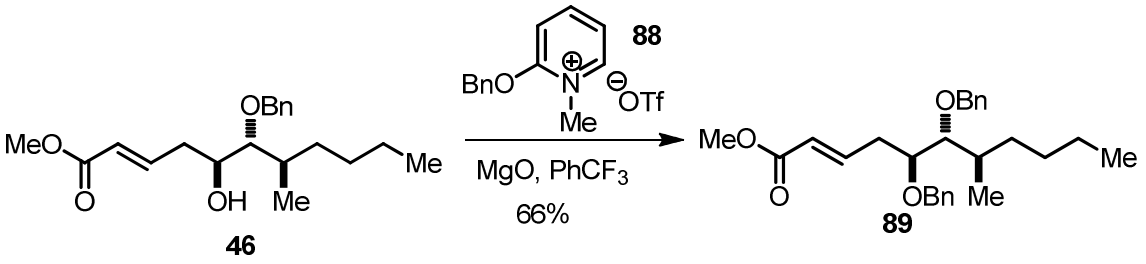
(m, 1H), 1.84 (bs, 1H), 1.78-1.73 (m, 1H), 1.68-1.64 (m, 1H), 1.42-1.18 (m, 5H), 0.96 (d, 3H, *J* = 6.6 Hz), 0.90 (t, 3H, *J* = 7.2 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 166.9, 146.5, 128.6, 127.9, 127.8, 123.5, 87.0, 74.9, 71.2, 51.6, 35.6, 35.1, 32.0, 29.6, 23.2, 16.4, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{20}H_{31}O_4$ ($M+H^+$) 335.2216, found 335.2217; **FT-IR**: 3444, 3064, 3031, 2952, 2929, 2871, 1724, 1658, 1454, 1324, 1070 cm^{-1}; **$[\alpha]_D^{25}$** = -21.9 (c = 1.16, $CHCl_3$).

For the seven-membered ring acetal intermediate, **83**: (dr 3.4 : 1 at acetal) **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.38-7.31 (m, 4H), 7.28-7.24 (m, 1H), 6.03-5.94 (m, 1H), 5.25- 5.18 (m, 1H), 4.83 (d, 1H, *J* = 11.6 Hz), 4.75 (d, 1H, *J* = 6.4 Hz),4.47 (d, 1H, *J* = 11.6 Hz), 4.16-4.11 (m, 1H), 3.91-3.85 (m, 1H), 3.74-3.71 (m, 1H), 3.62 (s, 3H), 3.23-3.16 (m, 2H), 1.81-1.62 (m, 4H), 1.51-1.43 (m, 1H), 1.36-1.10 (m, 6H), 0.96 (m, 6H).

Compound **46** was produced with complete *trans*-alkene selectivity and > 95:5 er, determined from the ^{1}H and ^{13}C NMR spectra. Mosher ester analysis as described earlier confirmed the stereochemical assignment at C14.

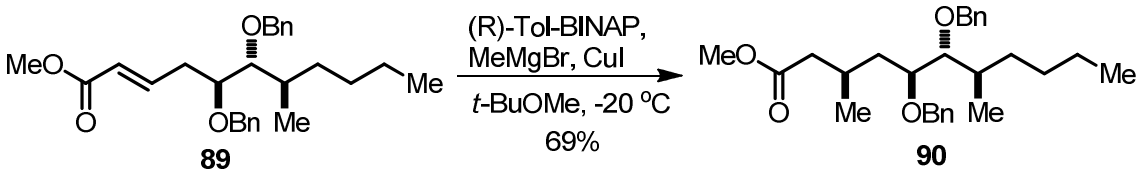
NMR ($CDCl_3$, 400 MHz):

	Compound **46**	***R*-Mosher ester**	***S*-Mosher ester**
H11	5.93 (d)	5.85	5.75
H12	7.05 (ddd)	6.90	6.81
H13	2.52 (ddd)	2.56	2.56
H13'	2.41 (ddd)	2.66	2.57
H14	3.91 (br dd)	5.38	5.43
H15	3.29 (dd)	3.22	3.33
OCH_2Ph	4.69, 4.62	4.24, 4.08	4.54, 4.36
	(AB dd, 2H)	(2H)	(2H)
H22	0.96 (d)	0.930	0.937



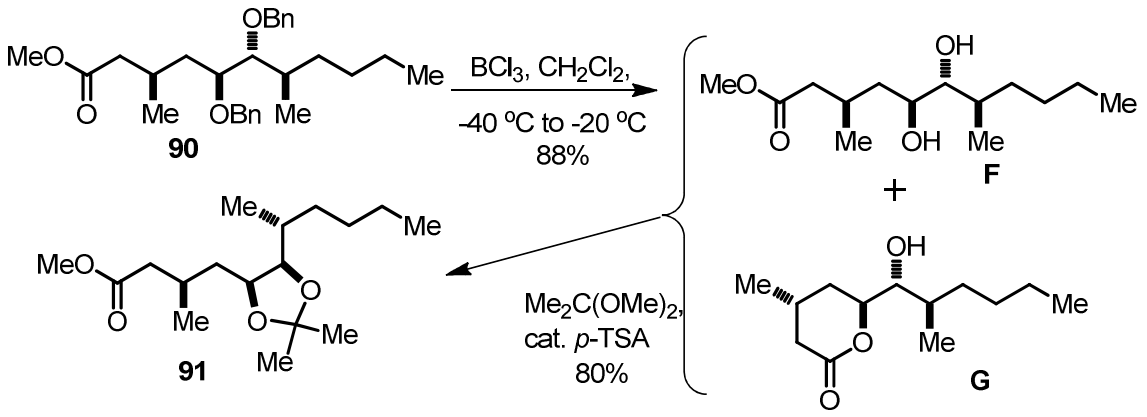
Synthesis of benzyl ether 89: The *E*-homoallylic alcohol **46** (1.4 g, 4.33 mmol), 2-benzyloxy-1-methylpyridinium triflate (3.0 g, 8.67 mmol) and MgO (0.35 g, 8.67 mmol, vacuum dried at 150 ^oC for 6 h) were dissolved in trifluorotoluene (9.0 mL), and the reaction mixture was heated at 85 ^oC for 24 h. After cooling, the reaction mixture was diluted with dichloromethane, and the reaction mixture was filtered through celite. The solvent was removed under vacuum to obtain the crude product as brown oil. The crude product was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to provide compound **89** as oil

(1.13 g, 66%). 150 mg of compound **46** was recovered. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.39-7.28 (m, 10H), 7.10 (ddd, 1H, *J* = 7.2, 15.6 Hz), 5.91 (d, 1H, *J* = 18.0 Hz), 4.77 (d, 1H, *J* = 11.4 Hz), 4.60 (d, 1H, *J* = 10.8 Hz), 4.57-4.52 (m, 2H), 3.73 (s, 3H), 3.70-3.68 (m, 1H), 3.41 (dd, 1H, *J* = 4.2, 6.0 Hz), 2.66-2.61 (m, 1H), 2.57-2.53 (m, 1H), 1.73-1.70 (m, 1H), 1.62-1.56 (m, 1H), 1.37-1.15 (m, 5H), 0.92 (d, 3H, *J* = 7.2 Hz), 0.90 (t, 3H, *J* = 7.2 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 167.0, 147.2, 138.9, 138.2, 128.5, 128.4, 128.1, 128.0, 127.9, 127.6, 122.8, 84.0, 79.4, 74.4, 71.9, 51.6, 35.2, 33.2, 32.1, 29.4, 23.2, 16.7, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{27}H_{37}O_4$ ($M+H^+$) 425.2686, found 425.2680; **FT-IR**: 3087, 3029, 2952, 2927, 2871, 1724, 1658, 1454, 1070 cm^{-1}; **$[\alpha]_D^{25}$** = -13.7 (c = 1.115, $CHCl_3$).



Synthesis of ester 90: Copper iodide (0.005 g, 0.026 mmol) and (*R*)–Tol-BINAP (0.03 g, 0.044 mmol) were dissolved in dichloromethane (5.5 mL) and stirred at room temperature for 20 min. The solvent was removed under vacuum and the resulting yellow solid was redissolved in methyl *t*-butyl ether (MTBE, 5.5 mL). The reaction mixture was cooled to -20 °C, and MeMgBr (2.3 ml, 6.94 mmol, 3.0 M in ether) was added dropwise. The reaction mixture was stirred for 15 min at -20 °C. A solution of the compound **89** (0.59 g, 1.38 mmol) in MTBE (1.6 mL) was then added over a period of 2 h at -20 °C using a syringe pump. The reaction mixture was stirred for an additional 45 min after addition, before quenching with methanol (5 mL) and saturated NH_4Cl solution (5 mL). The biphasic solution was stirred for 15 min at room temperature before extracting

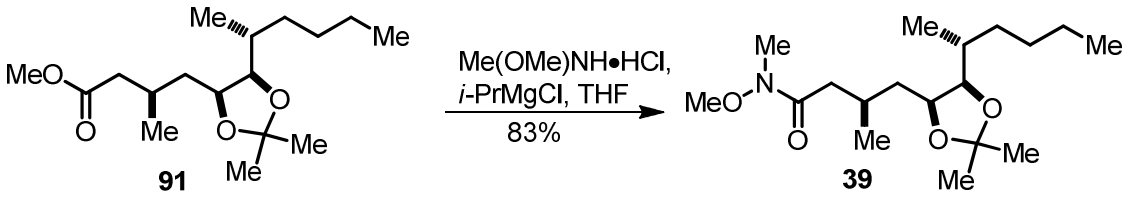
with ether. The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by flash chromatography using 15% ethyl acetate in hexanes as eluent to provide compound **90** as an oil (0.42 g, 69%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.39-7.27 (m, 10H), 4.87 (d, 1H, *J* = 11.4 Hz), 4.69 (d, 1H, *J* = 11.4 Hz), 4.56 (d, 1H, *J* = 11.4 Hz), 4.48 (d, 1H, *J* = 11.4 Hz), 3.65 (s, 3H), 3.64-3.61 (m, 1H), 3.45 (dd, 1H, *J* = 1.8, 7.8 Hz), 2.30-2.23 (m, 2H), 2.0 (ddd, 1H, *J* = 6.0, 6.9, 7.8 Hz), 1.77 (ddd, 1H, *J* = 4.8, 9.9, 14.2 Hz), 1.72–1.67 (m, 1H), 1.65-1.60 (m, 1H), 1.46 (ddd, 1H, *J* = 2.4, 9.0, 14.2 Hz), 1.37-1.24 (m, 3H), 1.22-1.14 (m, 2H), 1.01 (d, 3H, *J* = 6.6 Hz), 0.91-0.88 (m, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 173.7, 139.3, 138.6, 128.5, 128.4, 128.2, 128.1, 127.8, 127.5, 83.5, 78.8, 74.3, 71.5, 51.5, 40.7, 36.7, 35.3, 32.8, 29.2, 27.5, 23.3, 21.3, 16.6, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{28}H_{41}O_4$ ($M+H^+$) 441.2999, found 441.2999; **FT-IR**: 3029, 2954, 2871, 1739, 1454, 1070, 734, 698 cm^{-1}; **$[\alpha]_D^{25}$** = -8.1 (c = 0.965, $CHCl_3$).



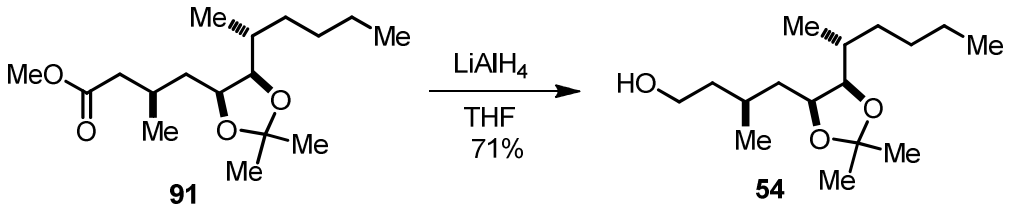
Synthesis of acetonide 91: Boron trichloride (3.8 mL, 1.0 M in heptane, 3.8 mmol) was added to a solution of compound **90** (0.42 g, 0.95 mmol) in CH_2Cl_2 (15 mL) at -45 °C. The resulting solution was stirred between -45 °C to -20 °C for

2 h and quenched with methanol (4 mL) after cooling to -78 ^{o}C, followed by warming to room temperature. The extractions were performed with CH_2Cl_2, and the organic layer was dried over $MgSO_4$, filtered and concentrated to obtain an oil. The crude product was purified by flash chromatography using 35% ethyl acetate in hexanes as eluent to provide a mixture of compounds **F** and **G** as an oil (0.218 g, 88%).

To a mixture of compound **F** and **G** (0.218 g, 0.83 mmol) were added 2, 2-dimethoxypropane (0.87 g, 8.37 mmol) and *p*-TSA (0.019 g, 0.1 mmol) at room temperature. The resulting solution was stirred at room temperature for 1 h and quenched with saturated $NaHCO_3$. The extractions were performed with CH_2Cl_2 and organic layer was dried over $MgSO_4$, filtered and concentrated to obtain crude product as oil. The crude product was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to provide compound **91** as an oil (0.20 g, 80%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.09 (ddd, 1H, *J* = 2.7, 3.0, 11.4 Hz), 3.72 (dd, 1H, *J* = 4.8, 9.6 Hz), 3.65 (s, 3H), 2.45 (dd, 1H, *J* = 4.2, 13.5 Hz), 2.26-2.17 (m, 2H), 1.71-1.67 (m, 1H), 1.62-1.59 (m, 1H), 1.50-1.46 (m, 1H), 1.42 (s, 3H), 1.39-1.25 (m, 7H), 1.24-1.18 (m, 1H), 1.15-1.09 (m, 1H), 1.02 (d, 3H, *J* = 6.6 Hz), 0.90 (t, 3H, *J* = 7.2 Hz), 0.82 (d, 3H, *J* = 6.6 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 173.6, 107.7, 82.8, 75.6, 51.5, 39.9, 35.8, 33.7, 32.3, 28.9, 28.7, 27.1, 26.4, 23.2, 21.2, 15.8, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{17}H_{33}O_4$ ($M+H^+$) 301.2373, found 301.2373; **FT-IR**: 2956, 2935, 2859, 1739, 1459, 1378, 1247, 1218, 1164, 1058, 877 cm^{-1}; $\mathbf{[\alpha]_D^{25}}$ = -46.2 (c = 1.74, $CHCl_3$).



Synthesis of Weinreb amide 39: Me(MeO)NH•HCl (0.089 g, 0.91 mmol) was dissolved in THF (1.8 mL) and cooled to -25 °C. *i*-PrMgCl (0.9 mL, 2.0 M in THF, 1.83 mmol) was added dropwise, and the reaction mixture stirred at -25 °C for 40 min. A solution of compound **91** (0.11 g, 0.36 mmol) in THF (0.7 mL) was then added, and the reaction mixture stirred for 45 min at -25 °C. The reaction mixture was quenched with saturated NH_4Cl solution, and the aqueous layer was extracted with ether. The organic layer was dried over Na_2SO_4, filtered and concentrated to obtain the crude product as oil. The crude product was purified by flash chromatography using 25% ethyl acetate in hexanes as eluent to provide compound **39** as oil (0.10 g, 83%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.11 (ddd, 1H, *J* = 2.4, 5.1, 10.5 Hz), 3.71 (dd, 1H, *J* = 5.4, 9.9 Hz), 3.68 (s, 3H), 3.17 (s, 3H), 2.55 (d, 1H, *J* = 13.8 Hz), 2.32-2.26 (m, 2H), 1.71-1.66 (m, 1H), 1.64-1.60 (m, 1H), 1.50-1.45 (m, 1H), 1.42 (s, 3H), 1.39-1.24 (m, 7H), 1.21-1.15 (m, 1H), 1.14-1.09 (m, 1H), 1.04 (d, 3H, *J* = 6.6 Hz), 0.90 (t, 3H, *J* = 6.6 Hz), 0.83 (d, 3H, *J* = 6.6 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 174.2, 107.6, 82.9, 76.0, 61.3, 37.8, 36.1, 33.7, 32.3, 28.8, 28.7, 27.1, 26.4, 23.2, 21.3, 16.0, 14.4; **HRMS** (APCI): *m/z* calcd. for $C_{18}H_{36}NO_4$ ($M+H^+$) 330.2638, found 330.2643; **FT-IR**: 2956, 2935, 2871, 1666, 1461, 1378, 1218, 1056 cm^{-1}; **$[\alpha]_D^{25}$** = -45.2 (c = 0.87, $CHCl_3$).



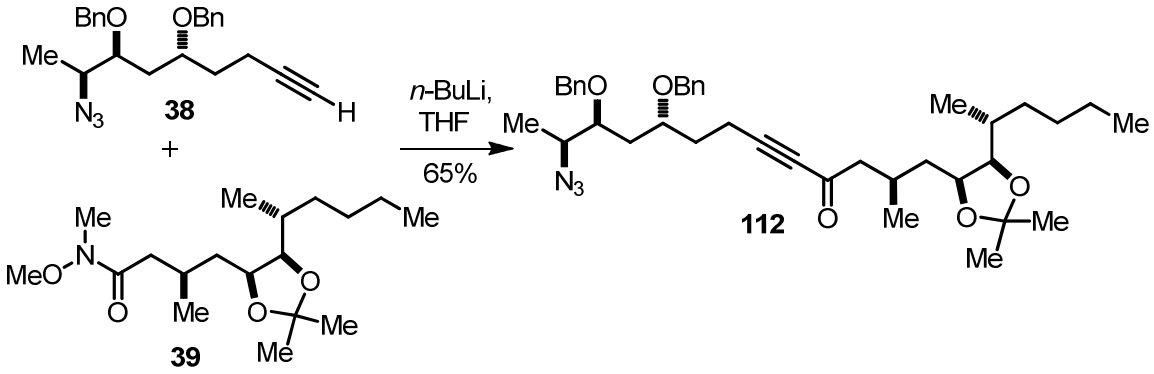
Synthesis of alcohol 54: $LiAlH_4$ (0.0083 g, 0.31 mmol, 1.0 M in THF) was suspended in THF (0.3 mL) and cooled to 0 ^oC. Compound **91** (0.027 g, 0.08 mmol) in THF (0.2 mL) was then added dropwise, and the reaction mixture was stirred for 15 min. The reaction was quenched with NH_4Cl, and the aqueous layer was extracted with ether. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the compound **54** (0.017 g, 71%) as oil, $\mathbf{[\alpha]_D^{25}}$ = -48.3 (c = 0.85, $CHCl_3$). The crude was analyzed without furthur purification for comparison with the reported spectroscopic data.[20]

FT-IR (cm^{-1})	
This work	Ref. 20
3417	3409
2931	2959, 2946
2871	2869
1461	
1378	
1245	
1060	

Comparison of spectroscopic data for compound **54** (this work) with ref. 20:

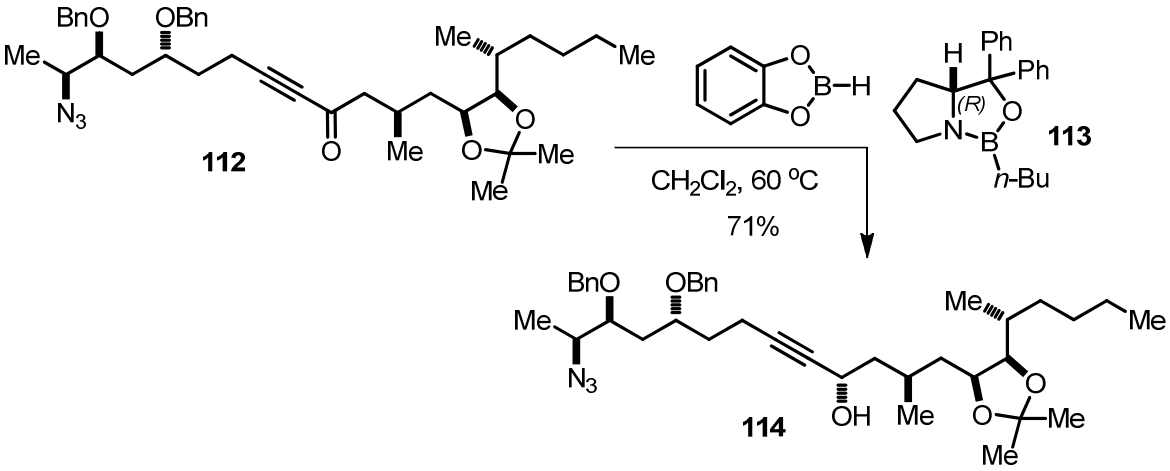
^{1}H NMR (400 MHz, $CDCl_3$, δ)		^{13}C NMR (100 MHz, $CDCl_3$, δ)	
This work	Ref. 20	This work	Ref. 20
4.13 (ddd, 1H, *J* = 2.8, 5.0, 11.6 Hz)	4.14 - 4.10 (m, 1H)	107.6	107.52
3.76-3.70 (m, 2H)	3.75 - 3.69 (m, 2H)	82.9	82.80
3.68-3.63 (m, 1H)	3.68 - 3.64 (m, 1H)	75.7	75.63
1.90-1.83 (m, 1H)	1.90 - 1.83 (m, 1H)	61.4	61.31
1.80-1.57 (m, 4H)	1.79 - 1.73 (m, 1H)	38.6	38.53
	1.70 - 1.66 (m, 1H)	37.1	36.99
	1.63 - 1.59 (m, 1H)	33.7	33.58
	1.57 - 1.53 (m, 1H)	32.3	32.18
1.52-1.44 (m, 1H)	1.51 - 1.45 (m, 1H)	28.9	28.75
1.42 (s, 3H)	1.42 (s, 3H)	28.7	28.62
1.40-1.27 (m, 3H)	1.40 - 1.25 (m, 3H)	26.4	26.28
1.26-1.18 (m, 2H)	1.24 - 1.20 (m, 2H)	26.3	26.25
1.32 (s, 3H)	1.32 (s, 3H)	23.2	23.11
1.16-1.08 (m, 1H)	1.16 - 1.11 (m, 1H)	21.5	21.35
0.98 (d, 3H, *J* = 6.4 Hz)	0.97 (d, 3H, *J* = 6.8 Hz)	16.0	15.89
0.90 (t, 3H, *J* = 7.2 Hz)	0.90 (t, 3H, *J* = 7.2 Hz)	14.4	14.24
0.82 (d, 3H, *J* = 6.8 Hz)	0.82 (d, 3H, *J* = 6.6 Hz)		

Cross-Coupling of fragment 38 and 39



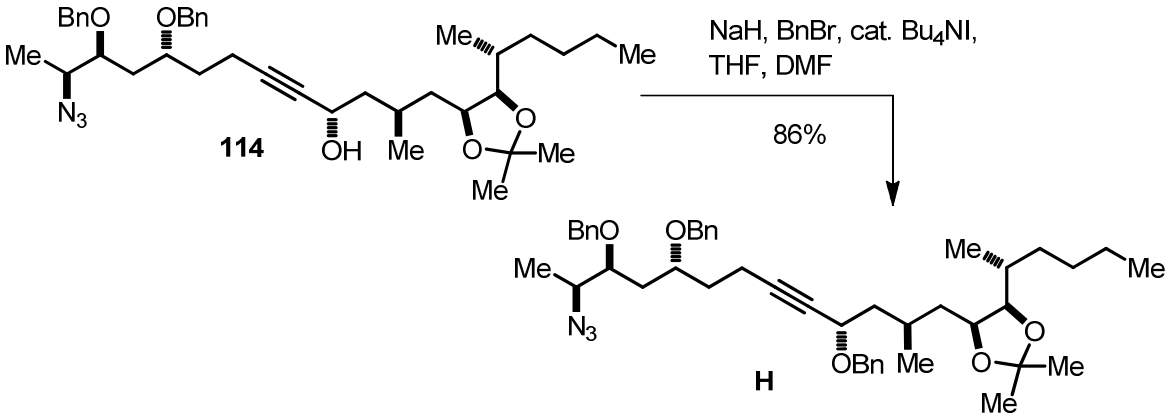
Synthesis of alkynone 112: A solution of alkyne **38** (0.17 g, 0.45 mmol) in THF (4 mL) was cooled to -78 °C under argon, and *n*-BuLi (0.17 ml, 2.5 M in hexanes, 0.42 mmol) was added. After 5 min at -78 °C, the reaction mixture was warmed to 0 °C and stirred for 30 min. The mixture was then cooled to -78 °C, and a solution of amide **39** (0.1 g, 0.3 mmol) in THF (1.0 mL) was added slowly. The resulting solution was stirred at -78 °C for 5 min and 0 °C for 1.5 h, and then quenched with saturated NH_4Cl. The extractions were performed with ether, and the organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude product as oil. The crude product was purified by flash chromatography using 7% ethyl acetate in hexanes as eluent to provide compound **112** (0.12 g, 65%) and recovered alkyne **38** (0.033 g). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.30-7.20 (m, 10H), 4.58 (d, 1H, *J* = 12.0 Hz), 4.53 (d, 1H, *J* = 11.4 Hz), 4.53 (d, 1H, *J* = 11.4 Hz), 4.37 (d, 1H, *J* = 11.4 Hz), 4.33 (d, 1H, *J* = 12.0 Hz), 4.07-4.05 (m, 1H), 3.73-3.68 (m, 2H), 3.61-3.57 (m, 2H), 2.67 (q, 1H, *J* = 8.4, 18.9 Hz), 2.49-2.33 (m, 4H), 1.93-1.81 (m, 2H), 1.77-1.67 (m, 2H), 1.65-1.60 (m, 2H), 1.50-1.45 (m, 1H), 1.42 (s, 3H), 1.40-1.18 (m, 12H), 1.59-1.09 (m, 1H), 1.13 (d, 3H, *J* = 6.0 Hz),

0.91 (t, 3H, *J* = 7.2 Hz), 0.82 (d, 3H, *J* = 7.2 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 188.0, 138.4, 138.2, 128.7, 128.6, 128.0, 128.0, 127.8, 127.9, 107.7, 93.4, 82.9, 81.6, 78.5, 75.5, 74.2, 73.4, 70.9, 59.5, 51.5, 36.0, 33.7, 32.3, 32.1, 28.7, 26.8, 26.4, 23.2, 21.1, 15.9, 14.9, 14.6, 14.4; **HRMS** (APCI): *m/z* calcd. for $C_{39}H_{56}O_5N_3$ ($M+H^+$) 646.4214, found 646.4227; **FT-IR**: 3031, 2929, 2871, 2211, 2105, 1670, 1456, 1378, 1249, 1060, 738 cm^{-1}; **$[\alpha]_D^{25}$** = -34.8 (c = 0.6, $CHCl_3$).



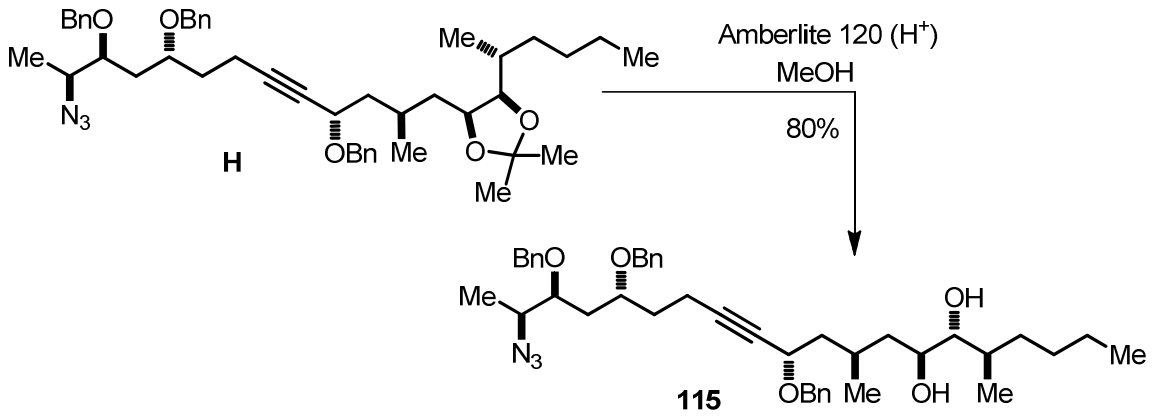
Synthesis of propargylic alcohol O: Alkynone **112** (0.06 g, 0.092 mmol) was treated with freshly prepared (*R*)-*B*-butyldiphenyloxazaborolidine **113** (0.13 mL, 0.5 M in toluene, 0.065 mmol). Toluene was removed *in vacuo*, CH_2Cl_2 (0.5 mL) was then added, and the solution was cooled to -78 °C. A solution of catecholborane (0.11 mL, 1.0 M in THF, 0.11 mmol) was then added dropwise. After stirring for 2.5 h at -78 °C and another 2.5 h between -65 to -55 °C, the reaction was quenched with methanol (0.3 mL) and the solution was warmed to room temperature. The reaction mixture was diluted with ether and washed with a mixture of 1 N NaOH : saturated $NaHCO_3$ (2 : 1) solution. The combined organic layers were washed with brine, dried over $MgSO_4$, filtered, and

concentrated. The crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **114** (0.043 g, 71%). ^{1}H NMR analysis of the crude product revealed ~ 9 : 1 dr. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.36-7.27 (m, 10H), 4.58-4.55 (m, 2H), 4.39-4.37 (m, 2H), 4.33 (d, 1H, *J* = 11.4 Hz), 4.13-4.11 (m, 1H), 3.73-3.69 (m, 2H), 3.61-3.55 (m, 2H), 2.35-2.24 (m, 2H), 2.07 (d, 1H, *J* = 5.4 Hz), 1.91-1.86 (m, 1H), 1.84-1.78 (m, 1H), 1.76-1.65 (m, 3H), 1.64-1.60 (m, 1H), 1.50-1.45 (m, 1H), 1.43 (s, 3H), 1.41-1.35 (m, 3H), 1.34-1.29 (m, 5H), 1.27 (d, 3H, *J* = 6.0 Hz), 1.24-1.10 (m, 4H), 1.10 (d, 3H, *J* = 6.6 Hz), 0.91 (t, 3H, *J* = 7.2 Hz), 0.82 (d, 3H, *J* = 6.6 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.6, 138.3, 128.6, 128.0, 127.9, 127.8, 107.6, 84.4, 82.8, 82.6, 78.7, 75.5, 74.4, 73.4, 70.7, 60.8, 59.7, 44.5, 36.9, 36.2, 33.6, 32.9, 32.3, 28.8, 28.7, 26.4, 26.2, 23.2, 21.1, 16.0, 15.0, 14.6, 14.4; **HRMS** (APCI): *m/z* calcd. for $C_{39}H_{58}O_5N_3$ (M^+) 648.4371, found 648.4375; **FT-IR**: 3428, 3031, 2933, 2871, 2107, 1454, 1378, 1247, 1060, 736 cm^{-1}; **$[\alpha]_D^{25}$** = -42.5 (c = 0.725, $CHCl_3$).



Synthesis of benzyl ether H: Sodium hydride (0.0037 g, 60% dispersion in mineral oil, 0.092 mmol) was added to a solution of propargylic alcohol **114** (0.04

g, 0.061 mmol) and tetrabutylammonium iodide (catalytic) in THF (0.2 mL) and 5 drops of DMF under argon at 0 °C. The resulting solution was stirred at 0 °C for 10 min and room temperature for 30 min before adding benzyl bromide (0.015 g, 0.092 mmol) at room temperature. The resulting solution was stirred at room temperature for 2 h and quenched with saturated NH_4Cl. The extractions were performed with ether, and the organic layer was dried over $MgSO_4$ followed by concentration to afford the crude product, which was purified by flash chromatography using 15% ethyl acetate in hexanes as eluent to provide compound **H** (0.039 g, 86%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.38-7.27 (m, 15H), 4.77 (d, 1H, *J* = 12.0 Hz), 4.59 (d, 1H, *J* = 8.4 Hz), 4.57 (d, 1H, *J* = 9.0 Hz), 4.47 (d, 1H, *J* = 11.4 Hz), 4.41 (d, 1H, *J* = 11.4 Hz), 4.36 (d, 1H, *J* = 11.4 Hz), 4.17-4.16 (m, 1H), 4.11-4.09 (m, 1H), 3.75-3.69 (m, 2H), 3.62-3.55 (m, 2H), 2.41-2.29 (m, 2H), 2.02-1.98 (m, 2H), 1.89-1.84 (m, 2H), 1.79-1.75 (m, 1H), 1.71-1.67 (m, 1H), 1.62-1.58 (m, 1H), 1.46-1.41 (m, 4H), 1.40-1.35 (m, 2H), 1.35-1.26 (m, 8H), 1.24-1.09 (m, 3H), 0.96 (d, 3H, *J* = 6.0 Hz), 0.91 (t, 3H, *J* = 7.8, 6.6 Hz), 0.80 (d, 3H, *J* = 6.6 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.6, 138.4, 138.3, 128.6, 128.6, 128.4, 128.1, 128.0, 127.9, 127.8, 127.6, 107.5, 84.8, 82.9, 80.1, 78.7, 76.0, 74.6, 73.3, 70.8, 70.1, 67.0, 59.7, 42.7, 37.0, 36.2, 33.6, 33.3, 32.4, 28.8, 28.7, 26.5, 26.4, 23.2, 20.9, 16.1, 15.0, 14.6, 14.4; **HRMS** (APCI): *m/z* calcd. for $C_{46}H_{64}O_5N_3$ ($M+H^+$) 738.4840, found 738.4842; **FT-IR**: 3031, 2929, 2869, 2107, 1454, 1378, 1064, 736 cm^{-1}; **$[\alpha]_D^{25}$** = -65.4 (c = 1.7, $CHCl_3$).

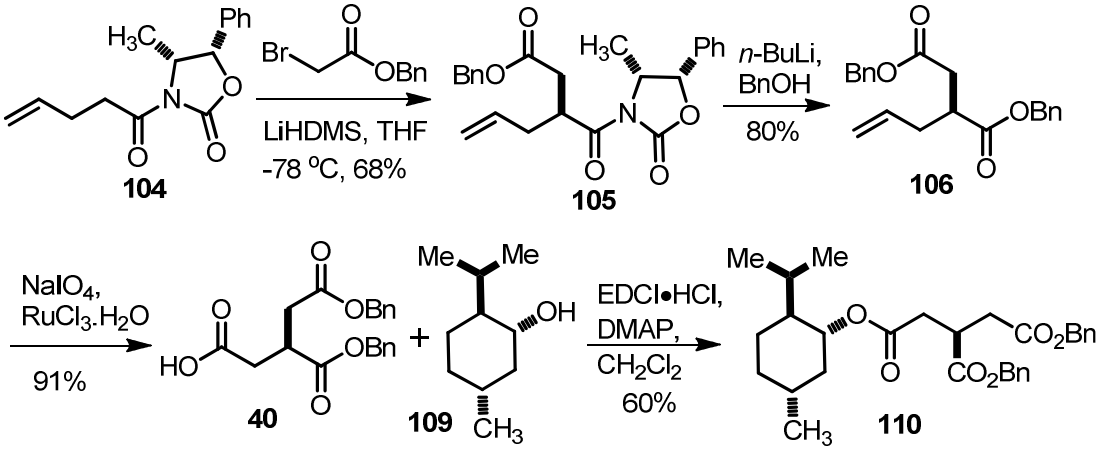


Synthesis of diol 115: The acetonide compound **H** (0.032 g, 0.043 mmol) was dissolved in methanol (0.5 mL), and preactivated Amberlite-120 acidic resin (0.5 g) was added at room temperature. The reaction mixture was stirred for 24 h, after which the resin was filtered off and the solvent was removed under vacuum. The crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **115** (0.024 g, 80%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.36-7.27 (m, 15H), 4.79 (d, 1H, *J* = 8.0 Hz), 4.59 (d, 1H, *J* = 4.0 Hz), 4.57 (d, 1H, *J* = 3.6 Hz), 4.47 (d, 1H, *J* = 7.6Hz), 4.40 (d, 1H, *J* = 7.6 Hz), 4.36 (d, 1H, *J* = 7.6 Hz), 4.15-4.11 (m, 1H), 3.79-3.72 (m, 2H), 3.62-3.55 (m, 2H), 3.35-3.33 (m, 1H), 2.41-2.30 (m, 3H), 2.05 (d, 1H, *J* = 1.60 Hz), 1.98-1.89 (m, 2H), 1.86-1.81 (m, 2H), 1.77 (ddd, 1H, *J* = 1.6, 6.0, 9.4 Hz), 1.69 (ddd, 2H, *J* = 2.4, 6.6, 10.0 Hz), 1.51-1.47 (m, 2H), 1.44-1.30 (m, 4H), 1.30-1.26 (m, 4H), 1.23-1.08 (m, 2H), 0.94 (d, 3H, *J* = 4.4 Hz), 0.91 (t, 3H, *J* = 4.8 Hz), 0.81 (d, 3H, *J* = 4.8 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 138.6, 138.3, 137.9, 128.6, 128.6, 128.6, 128.3, 128.0, 127.9, 127.9, 127.9, 86.0, 79.8, 79.1, 78.6, 74.5, 73.3, 70.8, 70.7, 69.5, 67.4, 59.7, 42.2, 36.9, 36.1, 34.9, 33.2, 32.4, 29.0, 26.2, 23.2, 21.5, 15.5, 15.0, 14.6, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{43}H_{60}O_5N_3$ ($M+H^+$) 698.4527,

found 698.4519; **FT-IR**: 3436, 3031, 2929, 2867, 2105, 1454, 1066, 1027, 736 cm^{-1}; $[\alpha]_D^{25}$ = -61.9 (c = 1.05, $CHCl_3$).

A new synthesis of (*S*)-Tricarballylic aciddibenzyl ester (40)

Although compound **40** is known from Kishi's synthesis of fumonisin B_2, we opted to develop a more concise synthesis based on stereoselective enolate alkylation and alkene oxidation to carboxylic acid **40**, analogous to the synthesis of a fully orthogonally protected tricarballylic acid diester reported by Harmat *et al.* The enantiomeric purity of **40** was confirmed by comparison of the menthyl ester **110** with an authentic sample of *rac*-**40**, prepared from the achiral *N*-acyloxazolidinone analog of **107**.



Synthesis of compound 105: Imide **104** (2.0 g, 7.77 mmol) was dissolved in THF (44 mL) and cooled to -78 °C. LiHMDS (1.56 g, 9.3 mmol, 1.0 M in THF) was then added and the reaction mixture was stirred at 0 °C for 20 min. The temperature was then lowered to -78 °C, and a solution of benzyl bromoacetate (2.31 g, 10.0 mmol) in THF (32 mL) was added over a period of 1.5 h using a

syringe pump. The reaction mixture was stirred at 0 °C for 30 min, after which the reaction was quenched with saturated NH_4Cl. The aqueous layer was extracted with ethyl acetate, the organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude product as a brown oil. This product was purified by flash chromatography with 10% ethyl acetate in hexanes as eluent to provide compound **105** as a colorless oil (2.13 g, 68%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.45-7.26 (m, 10H), 5.86-5.75 (m, 1H), 5.64 (d, 1H, *J* = 7.2 Hz), 5.19-5.02 (m, 4H), 4.76-4.69 (m, 1H), 4.42-4.34 (m, 1H), 2.95 (dd, 1H, *J* = 10.8, 17.0 Hz), 2.58 (dd, 1H, *J* = 4.4, 17.2 Hz), 2.45 (ddd, 1H, *J* = 6.8, 13.6 Hz), 2.25 (ddd, 1H, *J* = 7.6, 13.6 Hz), 0.79 (d, 3H, *J* = 6.4 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 174.9, 171.8, 152.8, 135.8, 134.4, 133.4, 128.9, 128.8, 128.7, 128.5, 128.4, 118.3, 79.0, 66.7, 55.1, 39.1, 36.6, 35.6, 14.1; **HRMS** (APCI): *m/z* calcd. for $C_{25}H_{26}O_4N$ ($M+H^+$) 408.1805, found 408.1803; **FT-IR**: 3066, 3033, 2981, 1779, 1733, 1697, 1346, 1195 cm^{-1}; **$[\alpha]_D^{25}$** = +12.9 (c = 1.405, $CHCl_3$).

Synthesis of compound 106: Benzyl alcohol (0.048 g, 0.44 mmol) was dissolved in THF (1.5 mL) and cooled to 0 °C. *n*-BuLi (0.18 mL g, 0.44 mmol, 2.5 M in hexanes) was then added and the reaction mixture was stirred for 15 min. A solution of compound **105** (0.15 g, 0.36 mmol) in THF (0.2 mL) was then added, and the reaction mixture was stirred at 0 °C for 30 min. The reaction was quenched with saturated NH_4Cl solution, and the aqueous layer was extracted with ether. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain colorless oil. This crude product was purified by flash chromatography using 10% ether in pentane as eluent to provide compound **106** as colorless oil

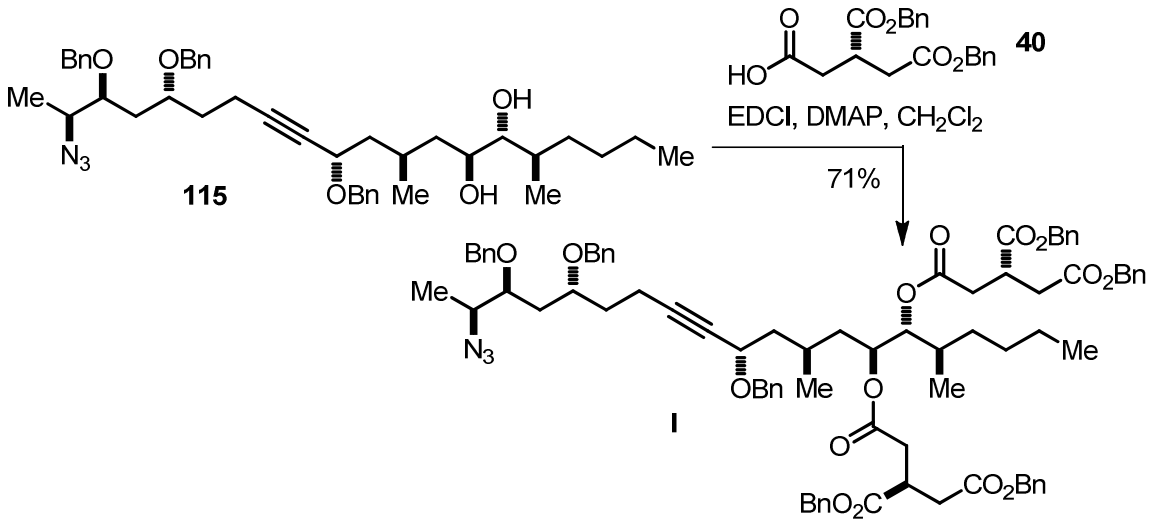
(0.1 g, 80%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.37-7.32 (m, 10H), 5.72-5.65 (m, 1H), 5.15-5.02 (m, 6H), 3.05-3.00 (m, 1H), 2.79 (dd, 1H, *J* = 9.0, 16.2 Hz), 2.54 (dd, 1H, *J* = 4.8, 17.1 Hz), 2.46 (ddd, 1H, *J* = 6.9, 13.8 Hz), 2.31 (ddd, 1H, *J* = 7.5, 14.4 Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 174.1, 171.8, 136.0, 135.9, 134.4, 128.7, 128.7, 128.4, 128.4, 119.2, 118.2, 66.7, 66.6, 41.0, 36.1, 35.3; **HRMS** (APCI): *m/z* calcd. for $C_{21}H_{23}O_4$ ($M+H^+$) 339.1590, found 339.1589; **FT-IR**: 3066, 3033, 2952, 1731, 1456, 1259, 1160 cm^{-1}; $[\alpha]_D^{25}$ = +9.2 (c = 1.045, $CHCl_3$).

Synthesis of tricarballylic acid synthon 40: Compound **105** (0.025 g, 0.073 mmol) was dissolved in a 2 : 2 : 3 mixture of CCl_4 : CH_3CN : H_2O (0.18 mL: 0.18 mL: 0.25 mL). Sodium periodate (0.063 g, 0.029 mmol) was then added followed by ruthenium trichloride (3.6 µg), and the reaction mixture was stirred for 4 h. The reaction mixture was diluted with water, and filtered through celite. The aqueous layer was then extracted with ether, and the organic layer was dried over $MgSO_4$, filtered and concentrated to afford compound **40** as an oil (0.024 g, 91%), which was used without further purification. **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.36-7.29 (m, 10H), 5.10 (d, 4H, *J* = 10.8 Hz), 3.33 (ddd, 1H, *J* = 6.8, 12.8 Hz), 2.88 (dd, 1H, *J* = 2.4, 6.8 Hz), 2.84 (dd, 1H, *J* = 2.8, 7.0 Hz), 2.72 (dd, 1H, *J* = 2.0, 6.6 Hz), 2.67 (dd, 1H, *J* = 2.4, 6.2 Hz); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 177.2, 172.8, 171.2, 135.6, 135.6, 128.7, 128.7, 128.5, 128.5, 128.3, 67.2, 66.9, 37.3, 35.3, 35.0; **FT-IR**: 3245, 3035 2956, 1737, 1714, 1164 cm^{-1}.

Synthesis of menthyl ester 110: (-)- Menthol, **109** (0.013 g, 0.08 mmol) and tricarballylic acid **40** (0.045 g, 0.12 mmol) were dissolved in CH_2Cl_2 (1.0 mL). DMAP (0.03 g, 0.24 mmol) and EDCI•HCl (0.048 g, 0.24 mmol) were added

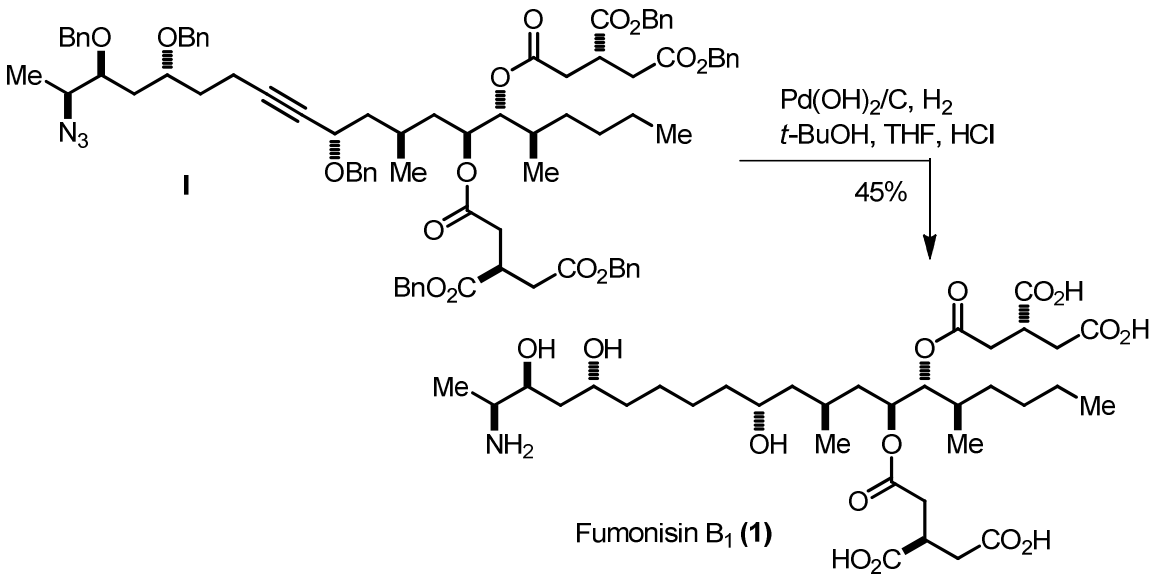
sequentially. The reaction mixture was stirred for 24 h The reaction was quenched with water, and the aqueous layer was extracted with dichloromethane. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain colorless oil. This crude product was purified by flash chromatography using 2% ethyl acetate in pentane as eluent to provide compound **110** as colorless oil (0.024 g, 60%). The diastereomer ratio was determined by ^{1}H NMR analysis in comparison with that for the racemic mixture, and was found to be 13:1.

Completion of the synthesis of Fumonisin B_1



Synthesis of fully protected fumonisin B_1 (compound I): The diol **115** (0.011 g, 0.015 mmol) and the tricarballylic acid **40** (0.017 g, 0.047 mmol) were dissolved in CH_2Cl_2 (0.6 mL). DMAP (0.011 g, 0.094 mmol) and EDCI-HCl (0.018 g, 0.094 mmol) were added sequentially. The reaction mixture was stirred for 20 h, and was then loaded on a column that was packed with silica gel and 2% Et_3N in hexanes. Elution with 10 to 20% ethyl acetate and 2% Et_3N in hexanes provided compound **I** (0.015 g, 71%). **^{1}H NMR** (600 MHz, C_6D_6) δ 7.43

(d, 2H, *J* = 7.2 Hz), 7.29 (dd, 4H, *J* = 7.2, 13.2 Hz), 7.22-7.13 (m, 14H), 7.10-7.01 (m, 15H), 5.48-5.45 (m, 1H), 5.28 (dd, 1H, *J* = 3.6, 8.7 Hz), 5.06-4.99 (m, 4H), 4.97-4.89 (m, 5H), 4.55 (d, 1H, *J* = 11.4 Hz), 4.42 (d, 1H, *J* = 11.4 Hz), 4.35 (d, 1H, *J* = 12.0 Hz), 4.33-4.30 (m, 1H), 4.26 (d, 1H, *J* = 11.4 Hz), 4.22 (d, 1H, *J* = 11.4 Hz), 3.72-3.68 (m, 1H), 3.54-3.47 (m, 1H), 3.40-3.36 (m, 1H), 3.21-3.17 (m, 1H), 2.96 (d, 1H, *J* = 6.0, 16.8 Hz), 2.89-2.71 (m, 4H), 2.57 (d, 1H, *J* = 6.0, 16.5 Hz), 2.53 (d, 1H, *J* = 6.0, 16.8 Hz), 2.32-2.27 (m, 5H), 2.25-2.19 (m, 2H), 2.14-2.08 (m, 1H), 1.82-1.67 (m, 4H), 1.61-1.56 (m, 3H), 1.54-1.16 (m, 8H), 1.09-0.99 (m, 1H), 0.97 (d, 3H, *J* = 7.2 Hz), 0.95 (d, 3H, *J* = 6.6 Hz), 0.85 (t, 3H, *J* = 6.9 Hz), 0.79 (d, 3H, *J* = 7.2 Hz); **^{13}C NMR** (150 MHz, C_6D_6) δ 173.3, 173.0, 171.68, 171.62, 171.3, 139.7, 139.3, 139.3, 136.9, 136.7, 129.0, 128.9, 128.8, 128.5, 128.3, 128.2, 86.7, 80.7, 79.0, 78.2, 75.1, 73.3, 72.3, 71.0, 70.8, 67.5, 67.3, 67.1, 66.8, 66.7, 59.7, 43.3, 38.2, 38.1, 35.8, 35.5, 34.4, 33.7, 32.6, 30.5, 29.2, 27.0, 23.5, 20.9, 15.8, 15.1, 14.8, 14.6; **HRMS** (ESI): *m/z* calcd. for $C_{83}H_{96}O_{15}N_3$ ($M+H^+$) 1374.6836, found 1374.6863; **FT-IR**: 3033, 2931, 2871, 2107, 1754, 1743, 1725, 1496, 1068, 750 cm^{-1}; **$[\alpha]_D^{25}$** = -46.5 (c = 0.59, C_6D_6).



Synthesis of fumonisin B_1 (1): Compound **I** (0.015 g, 0.0109 mmol) was dissolved in 8 drops of THF and 2 mL solution of *t*-BuOH-THF-HCl (0.2 mL of 1.0 N HCl in 15 mL of *t*-BuOH and 5 mL of THF). Pearlman catalyst (0.012 g) was then added, and the reaction mixture was evacuated under vacuum for 5 min before backfilling with hydrogen. The reaction mixture was stirred at room temperature for 18 h before filtering through a small bed of celite. The solvent was removed under vacuum to obtain crude product as colorless oil. The crude product was redissolved in a mixture of 1 : 1 MeOH : H_2O and loaded onto a pipette column containing C18 reverse phase silica gel (Bondesil, 40 µ, Varian Inc). The column was eluted with 1 : 1 MeOH : H_2O (6 mL), 3 : 1 MeOH : H_2O (4 mL), and finally with pure MeOH (10 mL). The solvents were removed under high vacuum (without warming) to provide fumonisin B_1 **(1)** (3.5 mg, 45%) as a white solid. $[\alpha]^{25}_{D}$ = -17 (c = 0.25, MeOH); Natural $[\alpha]^{25}_{D}$ = -11 (c = 0.1, MeOH); **FT-IR**: 3399, 2931, 2858, 1731, 1560, 1396, 1189, 1054 cm^{-1}; Natural 3397, 2931, 2859, 1729, 1565, 1405, 1162, 1064 cm^{-1}; **HRMS (ESI):** *m/z* calcd. for $C_{34}H_{59}O_{15}N$ ($M+H^+$) 722.3957, found 722.3977.

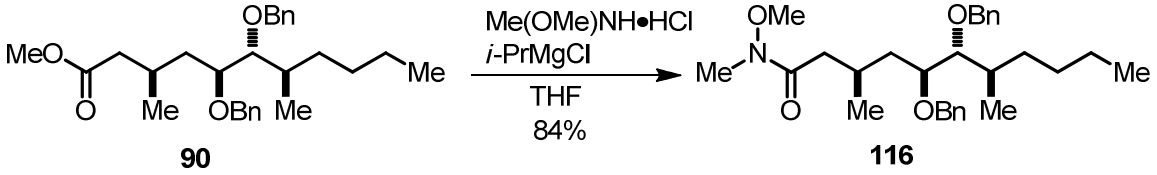
Spectroscopic comparison of natural fumonisin B_1 (purchased from Sigma, lot 028K4037) with synthetic **1** (this work)

^{13}C NMR (150 MHz, CD_3OD)		^{1}H NMR (600 MHz, CD_3OD)	
Natural	**Synthetic**	**Natural**	**Synthetic**
178.74	178.78	5.15 (ddd, 1H, *J* = 2.7, 11.4 Hz)	5.16 (ddd, 1H, *J* = 3.6, 10.2 Hz)
178.19	178.23	4.95 (dd, 1H, *J* = 3.6, 8.1 Hz)	4.94 (dd, 1H, *J* = 4.2, 8.1 Hz)
177.25	177.31	3.88-3.82 (m, 1H)	3.88-3.82 (m, 1H)
176.73	176.82	3.75-3.72 (m, 1H)	3.75-3.72 (m, 1H)
173.59	173.61	3.66-3.60 (m, 1H)	3.66-3.60 (m, 1H)
173.35	173.35	3.18-3.11 (m, 3H)	3.18-3.11 (m, 3H)
78.89	78.87	2.79 (dd, 1H, *J* = 7.8, 16.8 Hz)	2.78 (dd, 1H, *J* = 7.8, 16.8 Hz)
72.89	72.85	2.74-2.68 (m, 2H)	2.73-2.68 (m, 2H)
70.52	70.51	2.67-2.58 (m, 2H)	2.68-2.57 (m, 2H)
70.11	70.10	2.54-2.48 (m, 2H)	2.52-2.46 (m, 2H)
68.57	68.54	2.45 (dd, 1H, *J* = 6.0, 16.8 Hz)	2.44 (dd, 1H, *J* = 6.6, 16.8 Hz)
53.91	53.89	1.82 (bs, 1H)	1.81 (bs, 1H)
44.67	44.66	1.71-1.66 (m, 2H)	1.71-1.66 (m, 2H)
41.99	41.99	1.58-1.37 (m, 13H)	1.57-1.37 (m, 13H)
39.74	39.75	1.36-1.23 (m, 6H)	1.36-1.23 (m, 6H)
39.64	39.64	1.22-1.05 (m, 3H)	1.22-1.05 (m, 3H)
39.16	39.15	0.96 (d, 3H, *J* = 6.6 Hz)	0.96 (d, 3H, *J* = 6.6 Hz)
37.95	37.99	0.94 (d, 3H, *J* = 6.6 Hz)	0.94 (d, 3H, *J* = 7.2 Hz)
37.66	37.69	0.90 (t, 3H, *J* = 7.2 Hz)	0.90 (t, 3H, *J* = 7.2 Hz)
37.32	37.32		
37.04	37.00		
36.99			
35.03	35.03		
33.12	33.11		
29.89	29.90		
27.04	27.03		
26.81			
26.76	26.75		
24.05	24.07		
20.96	20.97		
16.16	16.19		
14.58	14.58		

Concentration dependence of ^{13}C NMR spectra of fumonisin B_1 (1)

Natural (from Sigma) 5 mm probe, 600 MHz 5 mg in 0.5 mL CD_3OD (0.013 M)	Synthetic (this work) 3 mm probe, 600 MHz 2 mg in 0.1 mL CD_3OD (0.027 M)	Synthetic (this work) 5 mm probe, 600 MHz 3.5 mg in 0.5 mL CD_3OD (0.0096 M)
178.74	179.34	178.78
178.19	178.95	178.23
177.25	177.91	177.31
176.73	177.46	176.82
173.59	173.70	173.61
173.35	173.61	173.35
78.89	78.83	78.87
72.89	72.90	72.85
70.52	70.51	70.51
70.11	70.05	70.10
68.57	68.55	68.54
53.91	53.93	53.89
44.67	44.72	44.66
41.99	42.02	41.99
39.74	40.07	39.75
39.64	39.26	39.64
39.16	39.21	39.15
37.95	38.44	37.99
37.66	38.32	37.69
37.32	37.47	37.32
37.04	37.22	37.00
36.99	36.92	-
35.03	35.05	35.03
33.12	33.18	33.11
29.89	29.87	29.90
27.04	27.04	27.03
26.81	26.89	-
26.76	26.81	26.75
24.05	24.07	24.07
20.96	20.92	20.97
16.16	16.17	16.19
14.58	14.59	14.58

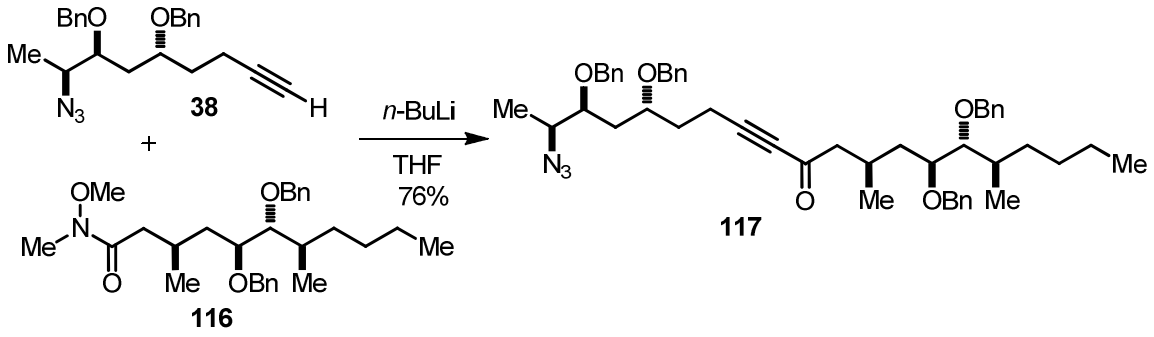
Synthesis of Fumonisin B_1 aminopentol (11) and hexaacetyl derivative (24)



90 **116**

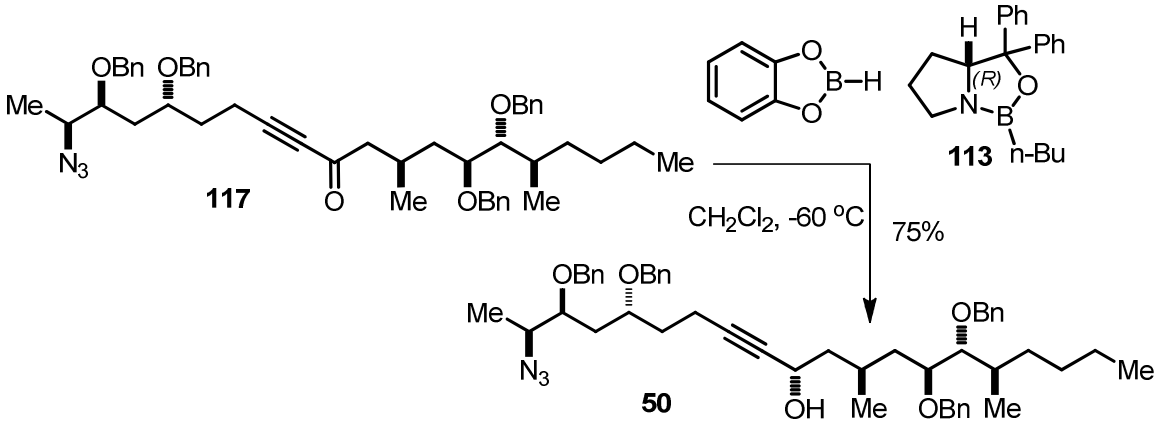
Synthesis of Weinreb amide 116: Me(MeO)NH•HCl (0.13 g, 1.34 mmol) was dissolved in THF (3.0 mL) under argon and cooled to -25 ^oC. *i*-PrMgCl (1.34 mL, 2.0 M in THF, 2.68 mmol) was added dropwise and the reaction mixture was stirred at -25 ^oC for 40 min. A solution of compound **90** (0.15 g, 0.31 mmol) in THF (1.0 mL) was then added, and the reaction mixture was stirred for 45 min at -25 ^oC. The reaction mixture was quenched with saturated NH_4Cl solution, and the aqueous layer was extracted with ether. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude product as oil. The crude product was purified by flash chromatography using 25% ethyl acetate in hexanes as eluent to provide compound **116** as oil (0.12 g, 84%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.39-7.28 (m, 10H), 4.86 (d, 1H, *J* = 11.4 Hz), 4.67 (d, 1H, *J* = 11.4 Hz), 4.56 (d, 1H, *J* = 11.4 Hz), 4.51 (d, 1H, *J* = 10.8 Hz), 3.68-3.66 (m, 1H), 3.59 (s, 3H), 3.43 (dd, 1H, *J* = 1.8, 7.5 Hz), 3.16 (s, 3H), 2.42-2.40 (m, 1H), 2.38-2.32 (m, 1H), 2.27-2.18 (m, 1H), 1.74 (ddd, 1H, *J* = 5.4, 9.3, 14.7 Hz), 1.70-1.64 (m, 2H), 1.55 (ddd, 1H, *J* = 2.4, 8.4, 14.7 Hz), 1.34-1.24 (m, 3H), 1.20-1.16 (m, 2H), 1.02 (d, 3H, *J* = 7.2 Hz), 0.91- 0.87 (m, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 174.2, 139.3, 138.8, 128.5, 128.4, 128.1, 127.6, 127.5, 83.9, 79.5, 74.3, 71.6, 61.2, 38.7, 37.4, 35.3, 32.7, 32.2, 29.2, 27.4, 23.3, 21.4, 16.7, 14.3; **HRMS**

(APCI): *m/z* calcd. for $C_{29}H_{44}O_4N_1$ ($M+H^+$) 470.3264, found 470.3269; **FT-IR**: 3064, 3029, 2956, 2871, 1666, 1454, 1095 cm^{-1}; $[\alpha]_D^{25}$ = -9.2 (c = 1.135, $CHCl_3$).



Synthesis of alkynone 117: Alkyne **38** (0.15 g, 0.375 mmol) was dissolved in THF (5.4 mL) and cooled to -78 °C, and *n*-BuLi (0.15 ml, 2.5 M in hexanes, 0.42 mmol) was added. After 5 min at -78 °C, the mixture was warmed to 0 °C and stirred for 30 min. The reaction mixture was then cooled to -78 °C, and a solution of amide **116** (0.12 g, 0.26 mmol) in THF (1.2 mL) was added slowly. The resulting solution was stirred at -78 °C for 5 min and 0 °C for 1.5 h and then quenched with saturated NH_4Cl. The extractions were performed with ether, and the organic layer was dried over $MgSO_4$ followed by concentration to afford the crude product. This material was purified by flash chromatography using 8% ethyl acetate in hexanes as eluent to provide compound **117** (0.16 g, 76%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 7.39-7.25 (m, 20H), 4.87 (d, 1H, *J* = 11.2 Hz), 4.69 (d, 1H, *J* = 11.2 Hz), 4.57 (d, 1H, *J* = 11.2 Hz), 4.56 (d, 1H, *J* = 11.2 Hz), 4.51 (d, 1H, *J* = 11.6 Hz), 4.48 (d, 1H, *J* = 11.2 Hz), 4.36 (d, 1H, *J* = 10.8 Hz), 4.31 (d, 1H, *J* = 11.2 Hz), 3.70-3.62 (m, 1H), 3.62-3.55 (m, 3H), 3.44 (dd, 1H, *J* = 2.0, 8.0 Hz), 2.51-2.30 (m, 4H), 2.21 (dd, 1H, *J* = 8.4, 14.8 Hz), 1.91-1.66 (m, 5H), 1.63-1.56

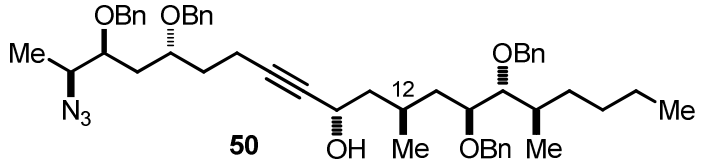
(m, 2H), 1.43 (ddd, 1H, *J* = 2.0, 8.6, 14.4 Hz), 1.33-1.24 (m, 6H), 1.23-1.14 (m, 2H), 1.00 (d, 3H, *J* = 6.4 Hz), 0.91-0.87 (m, 6H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 188.1, 139.2, 138.6, 138.3, 138.2, 128.6, 128.5, 128.4, 128.09, 128.06, 128.02, 127.9, 127.8, 127.5, 83.4, 81.4, 78.8, 74.3, 74.1, 73.3, 71.4, 70.8, 59.5, 52.1, 36.9, 35.9, 35.3, 32.7, 32.1, 29.1, 26.9, 23.2, 21.3, 16.6, 14.8, 14.5, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{50}H_{64}O_5N_3$ ($M+H^+$) 786.4853, found 786.4852; **FT-IR**: 3029, 2929, 2871, 2210, 2107, 1668, 1454, 1066, 736 cm^{-1}; **$[\alpha]_D^{25}$** = -10.5 (c = 0.955, $CHCl_3$).



Synthesis of propargylic alcohol 50: Alkynone **117** (0.06 g, 0.076 mmol) was treated with freshly prepared oxazaborolidine (0.09 mL, 0.5 M in toluene, 0.045 mmol). Toluene was removed *in vacuo*, CH_2Cl_2 (0.5 mL) was added, and the solution was cooled to -78 °C. A solution of catecholborane (0.09 mL, 1.0 M in THF, 0.09 mmol) was then added dropwise. After stirring for 2.5 h at -78 °C and 2.5 h between -65 to -55 °C, the reaction was quenched with methanol (0.3 mL) and the solution was warmed to room temperature. The solution was diluted with ether and washed with a mixture of 1 N NaOH-saturated $NaHCO_3$ (2 : 1) solution. The combined organic layers were washed with brine, dried over $MgSO_4$, filtered,

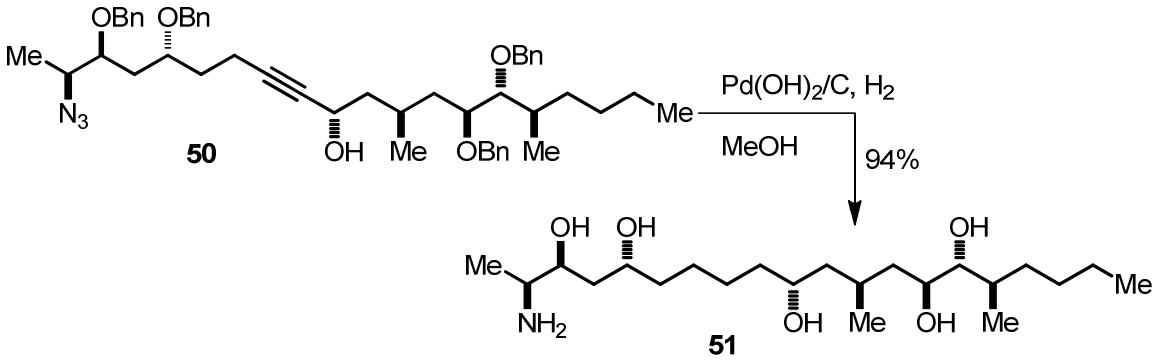
and concentrated. The crude product was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to provide compound **50** (0.045 g, 75%). 7 mg of the alkynone **117** was also recovered. ^{1}H NMR analysis of the crude product **50** revealed ~9 : 1 dr. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.39-7.27 (m, 20H), 4.86 (d, 1H, *J* = 11.4 Hz), 4.66 (d, 1H, *J* = 11.4 Hz), 4.58-4.54 (m, 3H), 4.49 (d, 1H, *J* = 11.4 Hz), 4.37 (d, 1H, *J* = 11.4 Hz), 4.36-4.31 (m, 2H), 3.71-3.68 (m, 2H), 3.60-3.54 (m, 2H), 3.42 (d, 1H, *J* = 7.8 Hz), 2.34-2.20 (m, 2H), 1.96 (bs, 1H), 1.85-1.56 (m, 9H), 1.45-1.39 (m, 1H), 1.35-1.22 (m, 7H), 1.20-1.14 (m, 2H), 0.98 (d, 3H, *J* = 6.6 Hz), 0.90 (d, 3H, *J* = 7.2 Hz), 0.89 (t, 3H, *J* = 7.2Hz); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 139.3, 138.8, 138.6, 138.3, 128.6, 128.5, 128.4, 128.2, 128.1, 128.0, 127.9, 127.8, 127.8, 127.5, 84.5, 84.0, 82.5, 79.0, 78.7, 74.4, 74.3, 73.4, 71.6, 70.7, 60.8, 59.7, 45.0, 37.5, 36.2, 35.3, 32.9, 32.7, 29.2, 26.5, 23.3, 20.9, 16.8, 15.0, 14.6, 14.3; **HRMS** (APCI): *m/z* calcd. for $C_{50}H_{66}O_5N_3$ ($M+H^+$) 788.5002, found 788.5001; **FT-IR**: 3436, 3087, 3064, 2929, 2871, 2360, 2105, 1454, 1257, 1066, 748 cm^{-1}; **$[\alpha]_D^{25}$** = -23.9 (c = 1.065, $CHCl_3$).

Mosher ester analysis and chemical shifts of methyl substituent at C12:



50

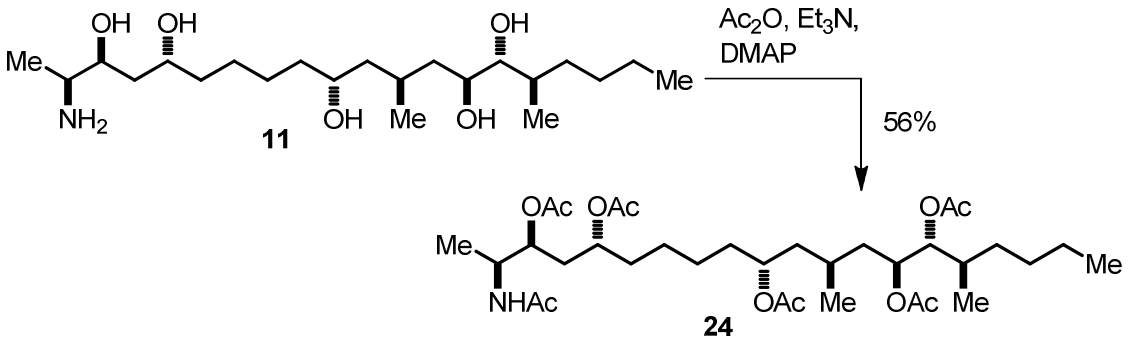
Compound 50 ($CDCl_3$)	**(*S*)-Mosher Ester ($CDCl_3$)**	**(*R*)-Mosher Ester ($CDCl_3$)**
0.98 (d)	**0.88 (d)**	**0.95 (d)**



Synthesis of fumonisin B_1 aminopentol (11): Compound **50** (0.022 g, 0.028 mmol) was dissolved in methanol (1 mL) and purged with argon for 5 min. Pearlman catalyst (0.02 g) was then added and the reaction mixture was purged with argon for another 5 min. The reaction mixture was then backfilled with hydrogen and stirred at room temperature for 14 h. The product mixture was filtered through celite, and the celite was washed with methanol. The solvent was removed under vacuo to obtain a white solid, which was washed with hexanes and filtered to provide aminopentol **11** as white solid (0.0108 g, 94%). $\mathbf{[\alpha]_D^{25}}$ = -20.3 (c = 0.36, MeOH); comparison with a sample from saponification of commercial fumonisin B_1, $\mathbf{[\alpha]_D^{25}}$ = -15 (c = 0.1, MeOH); **HRMS** (ESI): *m/z* calcd. for $C_{22}H_{47}O_5N$ ($M+H^+$) 406.3527, found 406.3528; **FT-IR**: 3376, 2929, 2858, 1610, 1461, 1282, 1056 cm^{-1}.

Spectroscopic comparison of "natural" aminopentol (**11**) obtained from hydrolysis of commercial fumonisin B_1, with synthetic **11**

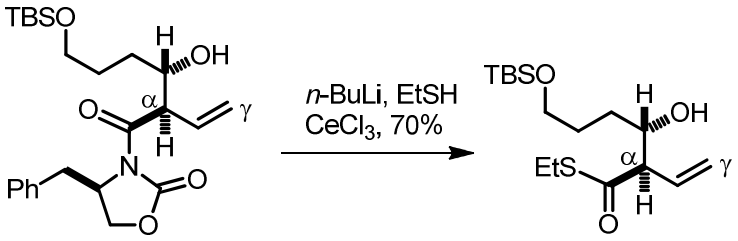
^{13}C NMR (150 MHz, CD_3OD)		^{1}H NMR (600 MHz, CD_3OD)	
Natural	**Synthetic**	**Natural**	**Synthetic**
80.92	80.91	3.86-3.78 (m, 1H)	3.86-3.78 (m, 1H)
70.54	70.53	3.75-3.71 (m, 1H)	3.76-3.70 (m, 1H)
	70.49	3.69-3.62 (m, 2H)	3.70-3.60 (m, 2H)
70.15	70.15	3.20 (t, 1H, *J* = 5.4, 9.6 Hz)	3.19 (t, 1H, *J* = 5.4, 9.6 Hz)
68.68	68.68	3.13- 3.09 (m,1H)	3.12 (t, 1H, *J* = 6.4 Hz)
53.96	53.98	2.00-1.90 (m, 1H)	2.00-1.90 (m, 1H)
44.81	44.79	1.74-1.51 (m, 4H)	1.74-1.52 (m, 4H)
42.08	42.07	1.50-1.31 (m, 13H)	1.49-1.31 (m, 13H)
41.83	41.81	1.30-1.06 (m, 7H)	1.29-1.04 (m, 7H)
39.79	39.78	0.98 (d, 3H, *J* = 6.6 Hz)	0.98 (d, 3H, *J* = 6.4 Hz)
39.46	39.44	0.93-0.91 (m, 6H)	0.94-0.90 (m, 6H)
36.05	36.04		
31.79	31.80		
30.84	30.83		
27.07	27.07		
	26.96		
24.38	24.37		
21.54	21.54		
16.97	16.97		
16.16	16.12		
14.68	14.68		



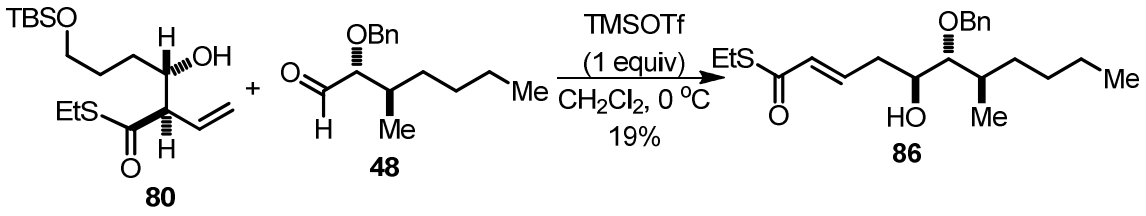
Synthesis of hexaacetyl derivative 24: Compound **11** (0.0024 mg, 0.0059 mmol) was dissolved in dichloromethane (0.2 mL). DMAP (one crystal) and Et_3N (5 drops) were then added at room temperature, followed by acetic anhydride (5 drops). The reaction temperature was stirred for 45 min and then diluted with water. The aqueous layer was extracted with dichloromethane. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude product was purified by flash chromatography using 2% ethyl acetate and hexanes to obtain compound **24** (0.0022 g, 56%) as oil.

Synthetic 24 (this work) **(400 MHz, CDCl3)**	**Reported by Gurjar**[22]
5.55 (d, 1H, *J* = 8.0 Hz)	5.58 (d, 1H, *J* = 9.3 Hz)
5.13 (dt, 1H, *J* = 2.8, 11.2 Hz)	5.17 (dt, 1H, *J* = 2.5, 10.1 Hz)
5.00- 4.82 (m, 4H)	5.02- 4.80 (m, 4H)
4.20- 4.11 (m, 1H)	4.17 (m, 1H)
2.09 (s, 3H)	2.09 (s, 3H)
2.08 (s, 3H)	2.07 (s, 3H)
2.03 (s, 3H)	2.02 (s, 3H)
2.01 (s, 3H)	2.01 (s, 3H)
1.999 (s, 3H)	1.99 (s, 6H)
1.996 (s, 3H)	
1.77 (m, 2H)	1.76 (m, 2H)
1.68-1.22 (m, 20H)	1.65-1.20 (m, 20H)
1.10 (d, 3H, *J* = 6.8 Hz)	1.10 (d, 3H, *J* = 6.7 Hz)
0.94 (d, 6H, *J* = 6.8 Hz)	0.93 (d, 6H, *J* = 6.8 Hz)
0.88 (d, 3H, *J* = 7.0 Hz)	0.87 (d, 3H, *J* = 6.8 Hz)

Synthesis of the thioester synthon 80:



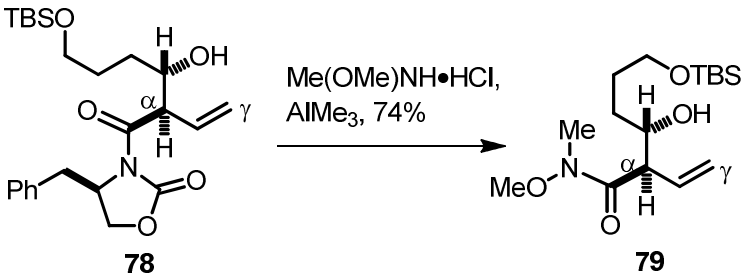
A solution of activated $CeCl_3$ (2.6 g, 10.6 mmol) in THF (75 mL) was cooled to -78 ^oC. EtSH (0.6 g, 9.78 mmol) was added followed by *n*-BuLi (0.56 g, 8.8 mmol) and stirred the reaction at 0 ^oC for 30 min before cooling to -25 ^oC. A solution of compound **78** (2.5 g, 4.4 mmol) in THF (25 mL) was added and stirred for 30 min. The reaction was quenched with saturated NH_4Cl at -25 ^oC and extracted the aqueous with ether, dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 2% ether in hexanes as eluent to provide compound **80** as oil (1.38 g, 70%). Recovered also 0.25 g of **78**. **^{1}H NMR** (400 MHz, $CDCl_3$) δ 6.01 (ddd, 1H, *J* = 9.6, 17.6 Hz), 5.32 (dd, *J* = 1.6, 10.0 Hz, 2H), 5.21 (dd, *J* = 1.6, 16.8 Hz, 1H), 4.21-4.17 (m, 1H), 3.59-3.57 (m, 2H), 3.18 (dd, *J* = 4.4, 9.4 Hz, 1H), 2.85 (dq, *J* = 1.6, 7.4 Hz, 2H), 1.52-1.49 (m, 4H), 1.23 (t, *J* = 7.6 Hz, 3H), 0.88 (s, 9H), 0.04 (s, 6H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 200.4, 132.6, 121.0, 73.0, 63.9, 63.2, 31.8, 28.5, 26.1, 26.0, 18.5, 14.6, -5.2.



For procedure refer compound **46**.

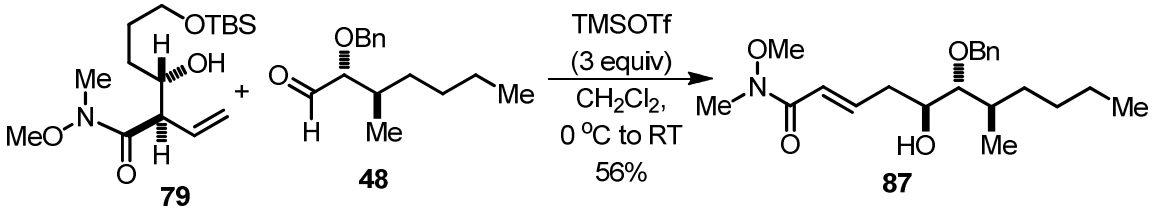
Compound **86**:**^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.38-7.30 (m, 5H), 6.96 (ddd, *J* = 6.8, 15.6 Hz, 1H), 6.19 (d, *J* = 15.6 Hz, 1H), 4.69 (d, *J* = 11.6 Hz, 1H), 4.61 (d, *J* = 10.8 Hz, 1H), 3.94-3.88 (m, 1H), 3.29 (app t, *J* = 5.2 Hz, 1H), 2.95 (q, *J* = 7.2 Hz, 2H), 2.53-2.48 (m, 1H), 2.44-2.35 (m, 1H), 1.81 (d, *J* = 3.2 Hz, 1H), 1.79-1.73 (m, 1H), 1.69-1.61 (m, 1H), 1.40-1.21 (m, 5H), 1.28 (t, *J* = 7.6 Hz, 3H), 0.95 (d, *J* = 6.8 Hz, 3H), 0.90 (t, *J* = 7.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 190.0, 142.1, 138.6, 131.1, 128.6, 127.9, 127.8, 87.0, 74.9, 71.2, 35.5, 35.1, 32.0, 29.6, 23.2 (2C), 16.4, 15.0, 14.3.

Synthesis of Weinreb synthon 79:



To a 0 °C cooled solution of *N*, *O*-dimethylhydroxyl amine•HCl (0.46 g, 4.6 mmol) in THF (7.8 mL) was added $AlMe_3$ (0.34 g, 4.71 mmol) dropwise and the reaction stirred for 30 min. A solution of the compound **78** (1.0 g, 2.23 mmol) in THF (3.3 mL) was then added dropwise and stirred for 30 min. The reaction was quenched by dropwise addition of 1.0 M HCl (1.3 mL) and the aqueous layer was then extracted with ether. The organic layer was dried over $MgSO_4$, filtered and

concentrated to obtain oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to provide compound **79** as an oil (0.57 g, 74%). **^{1}H NMR** (400 MHz, $CDCl_3$) δ 5.98 (ddd, *J* = 9.6, 17.2 Hz, 1H), 5.31 (dd, *J* = 1.6, 10.4 Hz, 1H), 5.22 (dd, *J* = 17.2 Hz, 1H), 4.03 (bs, 1H), 3.90 (bs, 1H), 3.69 (s, 3H), 3.67-3.60 (m, 2H), 3. 58-3.55 (m, 1H), 3.19 (s, 3H), 1.74-1.64 (m, 1H). 1.62-1.56 (m, 1H), 1.55-1.47 (m, 2H), 0.88 (s, 9H), 0.04 (s, 6H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 174.9, 132.7, 120.1, 71.6, 63.3, 61.7, 50.4, 32.0, 30.8, 29.2, 26.1, 18.5, -5.2.



For procedure refer to compound **46**.

Compound **87**: **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.36-7.35 (m, 4H), 7.30 (dd, *J* = 4.0, 8.6 Hz, 1H), 7.03 (ddd, *J* = 6.8, 15.6 Hz, 1H), 6.51 (d, *J* = 15.6 Hz, 1H), 4.68 (d, *J* = 11.2 Hz, 1H), 4.65 (d, *J* = 11.2 Hz, 1H), 3.96-3.90 (m, 1H), 3.69 (s, 3H), 3.30 (dd, *J* = 4.8 Hz, 1H), 3.25 (s, 3H), 2.62-2.56 (m, 1H), 2.49-2.41 (m, 1H), 1.84 (d, *J* = 6.0 Hz, 1H), 1.79-1.74 (m, 1H), 1.40-1.19 (m, 6H), 0.96 (d, *J* = 6.8 Hz, 3H), 0.90 (t, *J* = 6.8 Hz, 3H).

Chapter 2

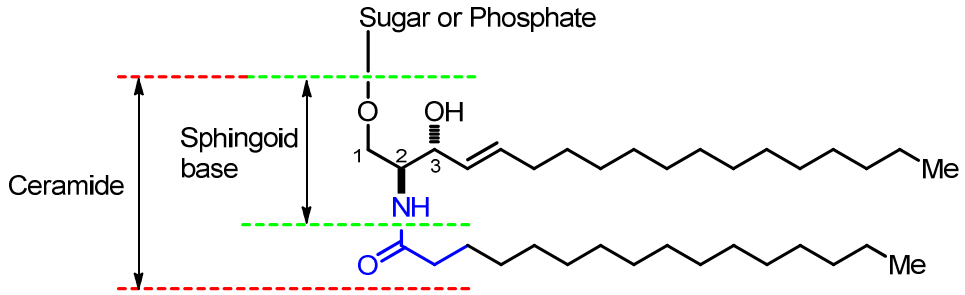
Chapter 2

Synthesis of unnatural sphingolipids: 1-deoxy-5-hydroxysphinganine and its diastereomers

2.1. Introduction and Background

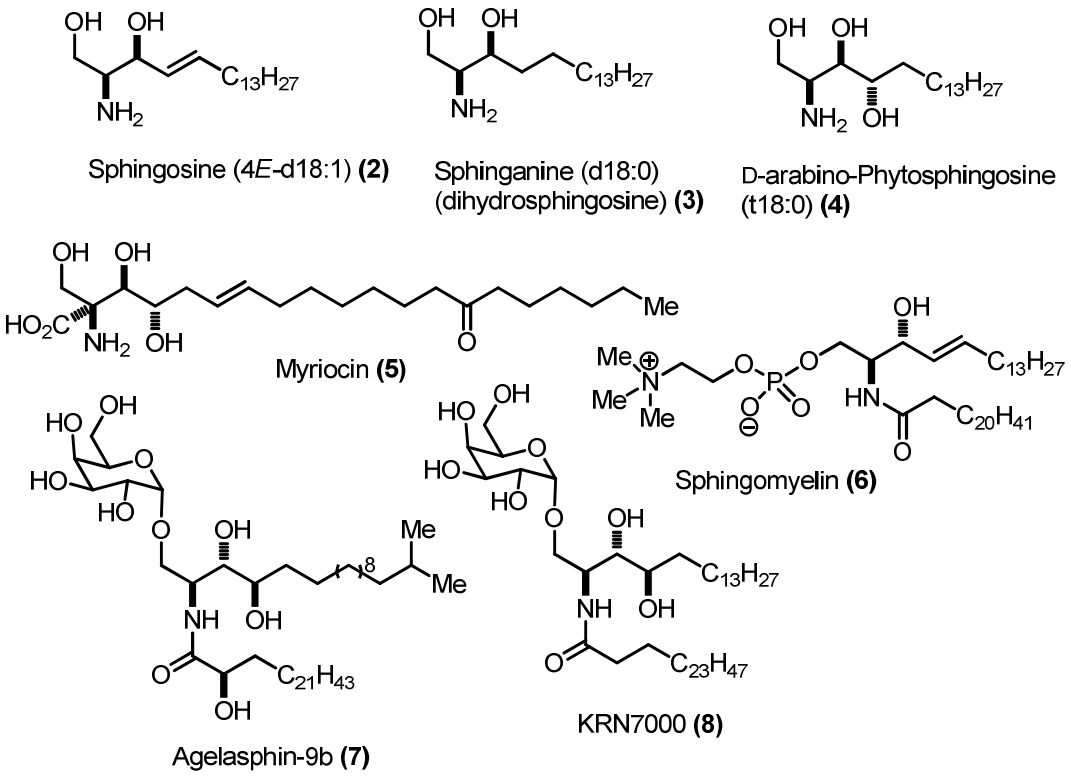
Sphingolipids are a diverse class of compounds that are found in all eukaryotic and some prokaryotic organisms.[65] They play a key role in signal transduction pathways that mediate cell growth, differentiation, cell functions and cell death.[66] They were first characterized in 1884 by J. L. W. Thudichum from the brain.[66] The term sphingo- was first coined by him because the enigmatic nature of the molecules reminded him of the riddle of the sphinx. The term "sphingolipide" was introduced by Herbert E. Carter, who discovered sphingosine and dihydrosphingosine.[67] More than 300 sphingolipids have been identified so far and the number is rising.[65] Though they are diverse they contain a common structural framework called the sphingoid base. This sphingoid base consists of a 2-amino-1,3-diol head group and an aliphatic chain that may be branched, saturated, or contain one or more double bonds (figure 1).[67]

Figure 1. Representative sphingolipid framework



The structural complexity and diversity of these molecules arises because of the various substituents attached to the 2-amino-1, 3-diol head group. The substituents can be fatty acids, sugars, proteins and zwitterionic species.[67] When a fatty acid is linked to the sphingoid base by an amide bond, the compounds are called ceramides (figure 1 and 2). From here the sphingolipids fall into two major classes called sphingomyelins **(6)** and glycosphingolipids **(7)** and **(8)** (figure 2).[68] Sphingomyelins contain a phosphocholine group and the glycosphingolipids contain various carbohydrates.

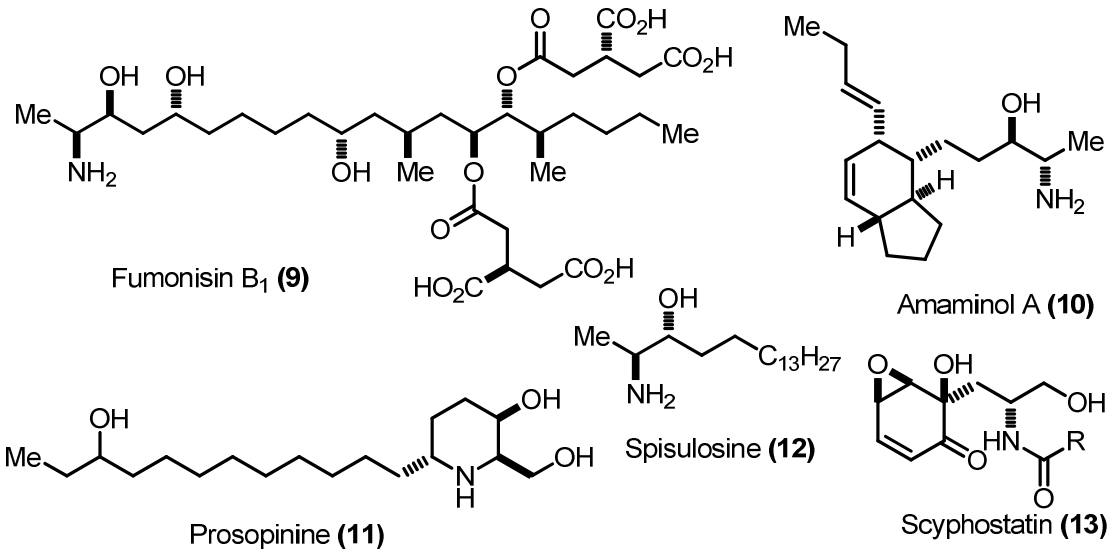
Figure 2. Some representative examples of various classes of sphingolipids



There are also natural compounds which are not sphingolipids but do possess a similar framework with a sphingoid base. Compounds such as

fumonisin B_1 **(9)**, spisulosine **(12)** and others represent this growing category of molecules with varying biological activity (figure 3).[65, 67]

Figure 3. Some representative sphingoid baselike compounds

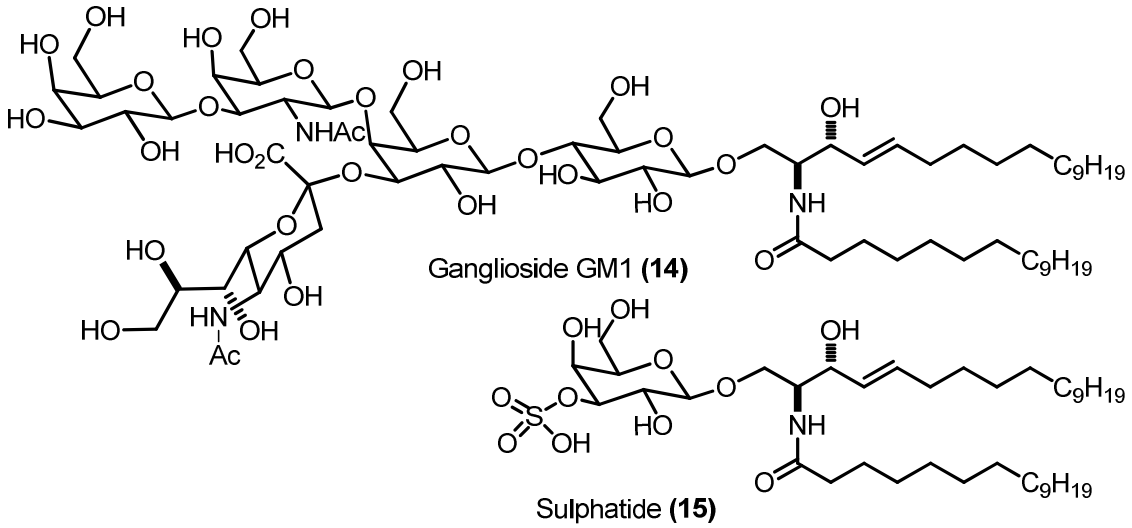


Sphingomyelins **(6)** are located in the plasma membrane as well as in intracellular organelles. The metabolites of these compounds generate anticancer signals that may inhibit cell proliferation, induce differentiation or promote apotosis.[68] Glycosphingolipids are also membrane components and are the major glycans of vertebrate brain. They are classified as neutral (gangliosides **(14)**) or acidic (sulfatides **(15)**) (figure 4). Their role is in cell-cell interaction and modulating protein activities.[68]

Since these sphingolipids are complex, a shorthand nomenclature of naming is followed similar to that used for fatty acids.[69] The number of hydroxyl groups are abbreviated as " d " or " t " for di- and tri-hydroxyl group followed by the number of carbon atoms and double bonds. Sphingosine (**2**, figure 2) would

thus be d18:1 (d = dihydroxyl, 18 = C18-carbon chain, 1 = one double bond), similarly phytosphingosine (**4**, figure 2) would be t18:0. If one wants to indicate the position and geometry of the double bond then a superscript would be added. Sphingosine **(2)** would thus become 4*E*-d18:1 or d18:1$^{\Delta 4}$.[69]

Figure 4. Neutral and acidic glycosphingolipids

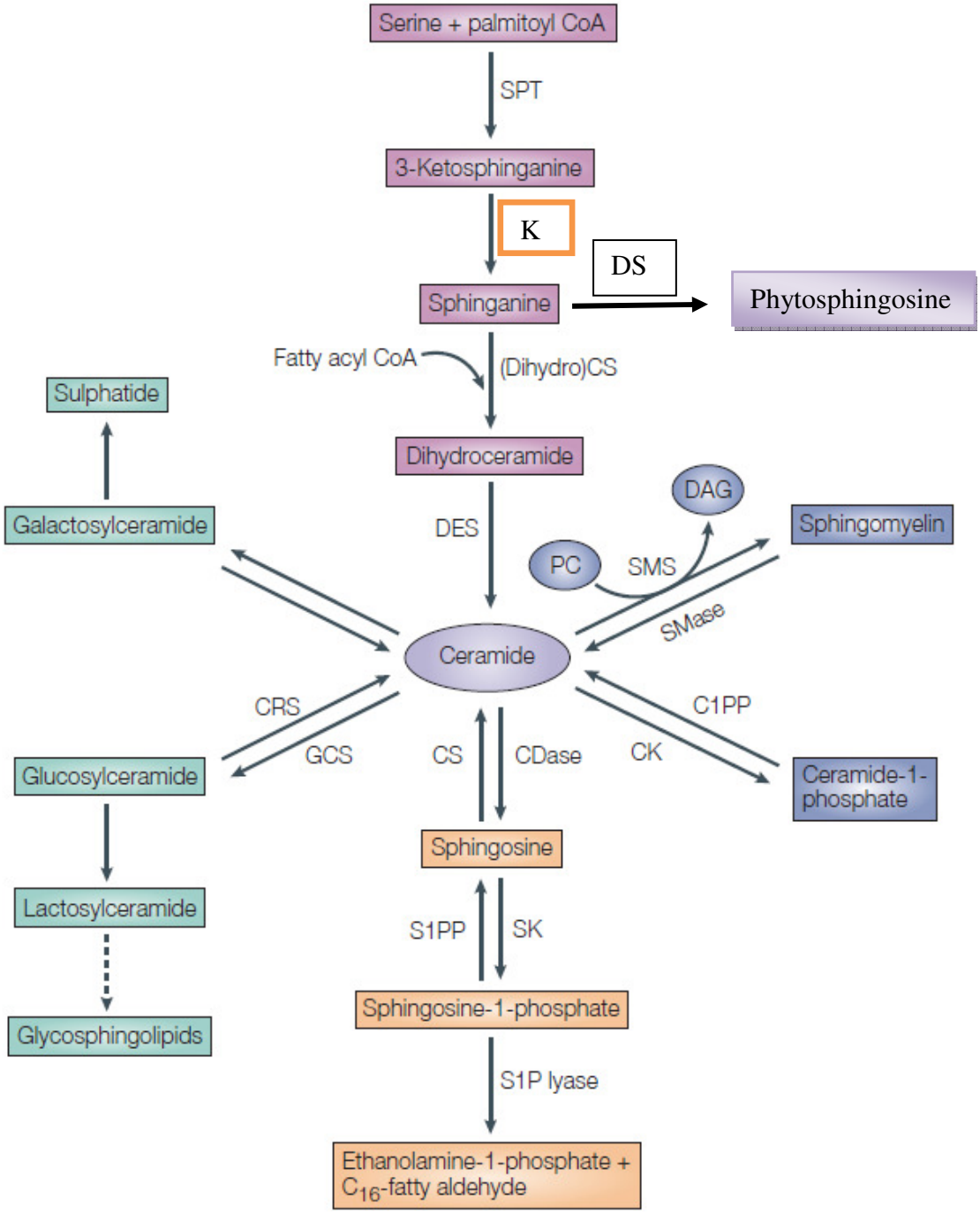


2.2. *De novo* synthesis of sphingolipids

Before going into the details of the sphingolipids biological role, it is important to know how these compounds are synthesized and metabolized endogenously. In the endoplasmic reticulum the enzyme serine palmitoyltranferase (SPT) catalyses the condensation of L-serine and palmitoyl-CoA to produce ketosphinganine which is reduced by ketosphinganine reductase (KR) to sphinganine (figure 5).[9] This is rapidly converted to dihydroceramide (*N*-acylsphinganine) by a family of sphinganine and sphingosine *N*-acyltransferase

enzyme collectively called ceramide synthase (DCS). The *E* double bond is then introduced by a dehydrogenation process catalysed by desaturase (DES).

Figure 5. Sphingolipid de novo synthesis and metabolism



Ceramide then becomes a major substrate and a branching point for the synthesis of more complex sphingolipids. Ceramide can be metabolized to sphingosine by ceramidases (Cdase), which in turn are phosphorylated by sphingosine kinase (SK) to form sphingosine-1-phosphate (S1P).[9] This can be synthesized back to sphingosine by phosphotase (S1PP) or can be cleaved by lyase (S1P lyase) into ethanolamine-1-phosphate and C_{16}-fatty aldehyde. Glycosphingolipids are generated by the sequential addition of monosaccharides like glucose, galactose, or sialic acid by glycosyltransferaces and sialyltransferases. The breakdown of complex sphingolipids like glycosphingolipids and sphingomyelin by glucosidases and sphingomyelinases also results in the formation of ceramide thus resulting in sphigolipid turnover.[9]

2.3. Biological significance of sphingolipids generated from biosynthesis

Ceramides: These mediate the regulation of growth, differentiation, senescense, apoptosis and protein secretion.[66] They also play a role in mediating the action of vitamin D3, TNF-α, and FAS ligand.[68, 70]

Sphingosine and sphinganine: These mediate the regulation of growth, differentiation, senescense, and apoptosis.[66] At lower concentrations these are growth stimulators, but at higher concentrations they are cytotoxic to normal cells. They are also inhibitors of protein kinase C (PKC).[68, 70]

Sphingosine-1-phosphate and sphinganine-1-phosphate: These are regulators of growth, cell mobility and intracellular calcium homeostasis.[66] They also mediate the action of platelet derived growth factor.[68, 71]

Sphingomyelin, glycosphingolipid and other complex sphingolipids: They regulate cell growth, protein trafficking and sorting, cell-cell communication and adhesion.[66, 68] They also are recognition sites for microorgansims and toxins.

2.4. Role of Sphingolipids in regulating cancer

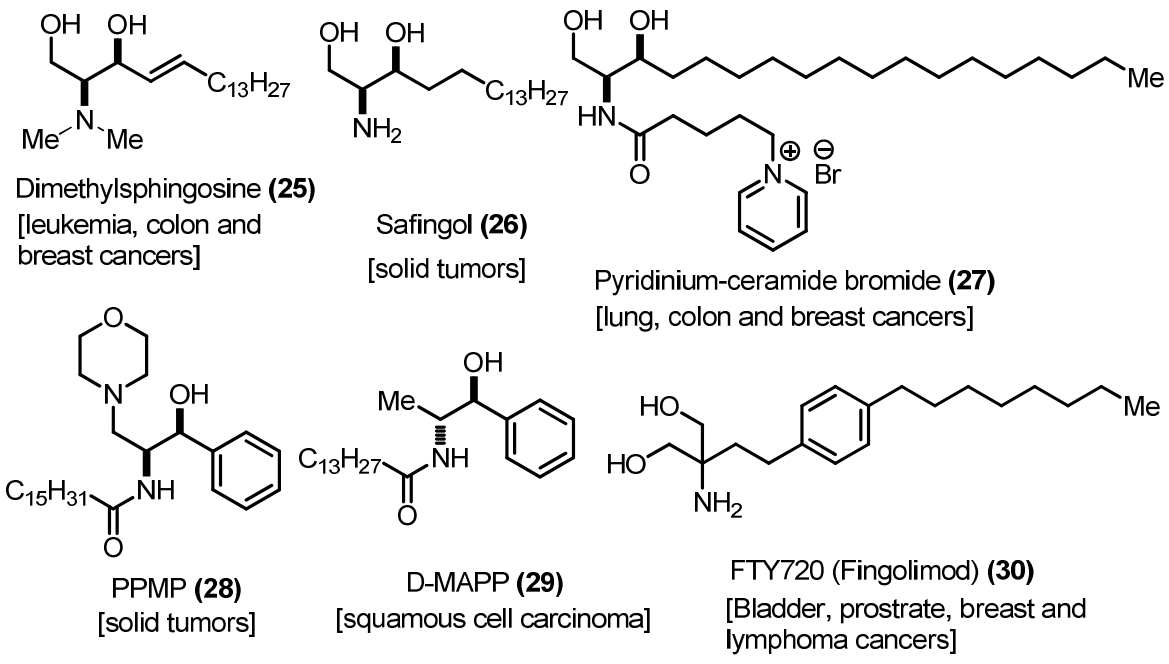
Studies have indicated that altered levels of ceramide and expression of enzymes of sphingolipid metabolism may play a key role in the growth of various cancers.[72] Strategies that mimic or modulate these properties could provide novel avenues for cancer therapy. Chemotherapeutic agents have shown to regulate individual components of the *de novo* pathway of ceramide synthesis on one hand, and also to control the sphingomyelin pathway for the production of ceramide on the other.

The roles of sphingolipids in the modulation of cell behaviors pertinent to cancer prevention and treatment, is a field still in its infancy. Much needs to be done to understand the role of exogenously administered sphingolipid analogs on the metabolism and functions of the endogenous sphingolipids. Based on the current evidence for the anticancer efficacy of the sphingoid base and ceramide backbones of sphingolipids both in cell culture and in vivo, developing new synthetic analogs that would help us to understand more about this class of bioactive compounds seems very reasonable in this regard.

2.5. Synthesis of 1-deoxy-5-hydroxysphinganine and their analogs

The interesting biological activities of sphingolipids, specifically as anticancer principles have resulted in many synthetic endeavors to synthesize these compounds. Many of these sphingolipid analog compounds like dimethylsphingosine **(25)**, safingol **(26)**, pyridinium-ceramide bromide **(27)**, PPMP **(28)**, D-MAPP **(29)**, fingolimod **(30)**, have become pharmaceutical leads for many types of cancer (figure 5).[65, 66,73]

Figure 6. Synthetic sphingolipid based compounds as pharmaceutical leads

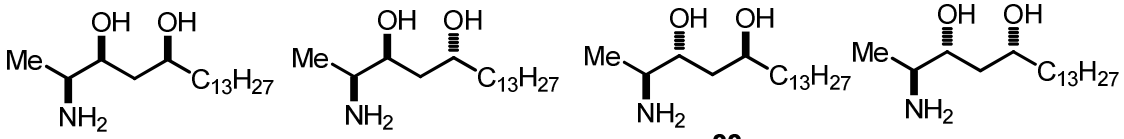


As mentioned before the primary component of sphingolipids is the sphingoid base, specifically the 2-amino-1, 3-diol headgroup. In some sphingoid base like compounds like fumonisins **(9)**, spisulosine **(12)**, and amaminol A **(10**) (figure 3), the primary hydroxyl group is absent or else moved to the C5 position

along the carbon chain and these compounds have also shown potential biological activities.[67] Recent reports indicate that ceramide is a prominent compound responsible for the induction of tumor death via apoptosis.[72] On the other hand the primary hydroxyl group of these compound are substituted by phosphates and carbohydrates and these compounds are responsible for tumor growth, proliferation and angiogenesis.

Given this knowledge it becomes clear that in an analog compound of sphingolipid, the C1 hydroxyl group should be removed to avoid any phosphorylation. This change on the other hand also affects the hydrophobicity of the molecule. To balance this Liotta *et al.* developed a synthetic analog of sphingolipids wherein the primary hydroxyl group was moved to the C5 position called 1-deoxy-5-hydroxysphinganines (figure 7).[74]

Figure 7. 1-deoxy-5-hydroxysphinganine compounds

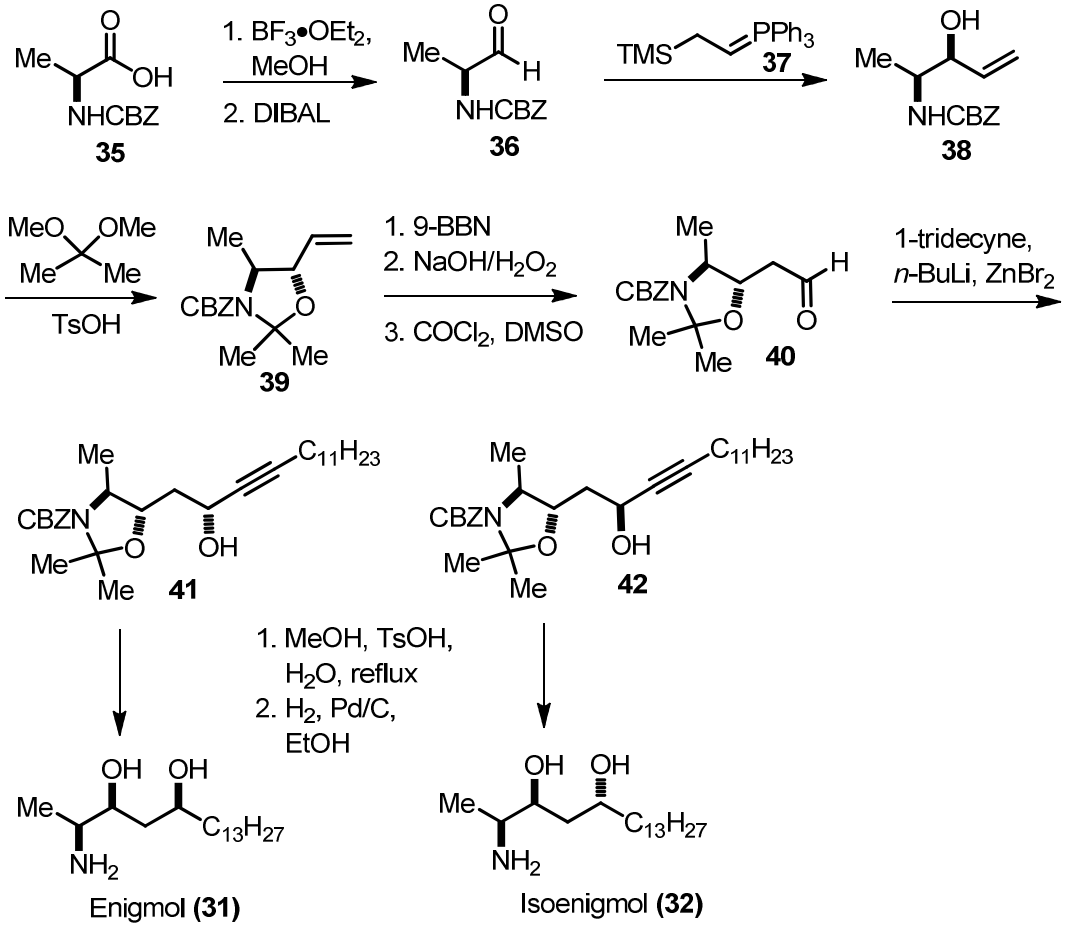


2.5.1. Synthesis of 1-deoxy-5-hydroxysphinganine compounds by Liotta *et al.*

The synthesis of these deoxy compounds began from aldehyde **36** obtained from the CBZ protected aminoacid L-alanine **35** (scheme 1).[75] Reaction of aldehyde **36** with phosphorane **37** gave compound **38** which was converted to aldehyde **40** in 4 steps. Addition of lithiated tridecyne to aldehyde **40** gave

alkynols **41** and **42** in a 9:1 diastereomeric ratio, which were separated and converted to the respective compounds **31** and **32** in 2 steps (scheme 1).

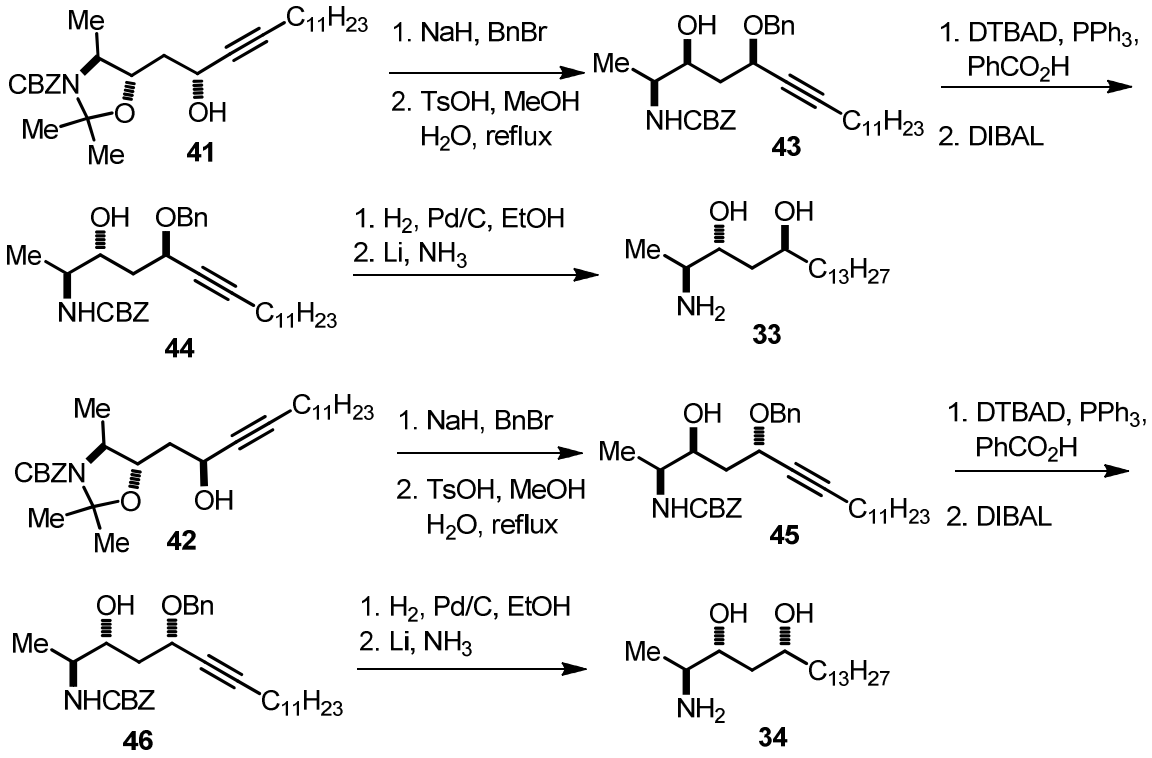
Scheme 1. Synthesis of 1-deoxy-5-hydroxy compounds enigmol **(31)** and isoenigmol **(32)**



The other two enigmol diastereomers **33** and **34** were synthesized starting from intermediates **41** and **42** (scheme 2). Benzyl protection of these compounds followed by acetonide cleavage gave compounds **43** and **45** respectively, which

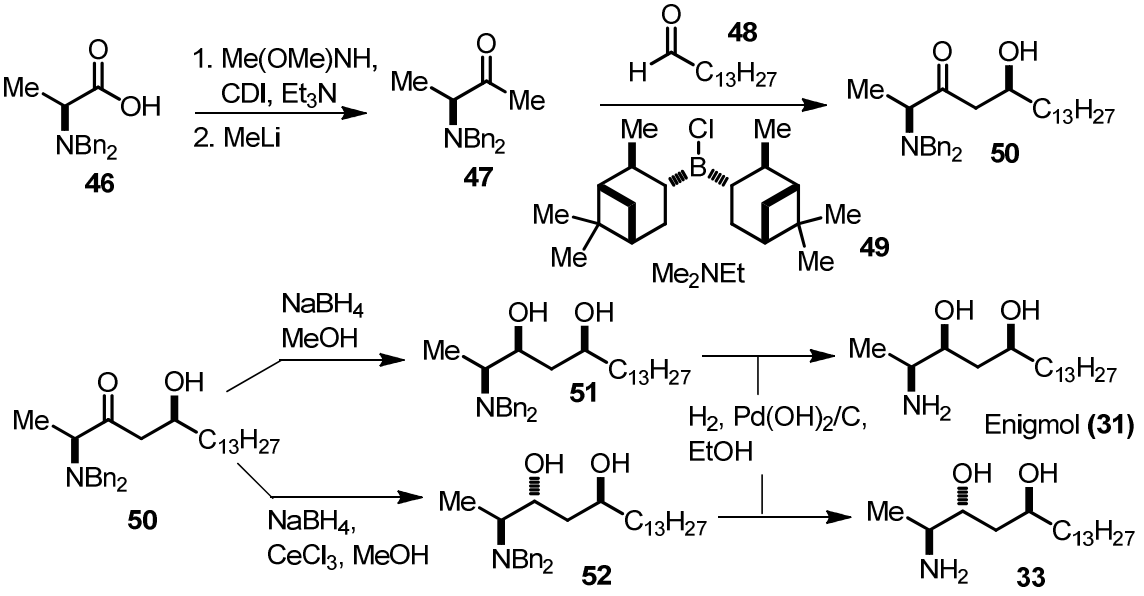
underwent a Mitsunobu inversion to invert the stereocentre at C3. Deprotection of the benzyl and CBZ protecting group gave the diastereomers **33** and **34**.

Scheme 2. Synthesis of 1-deoxy-5-hydroxy diastereomers **33** and **34**



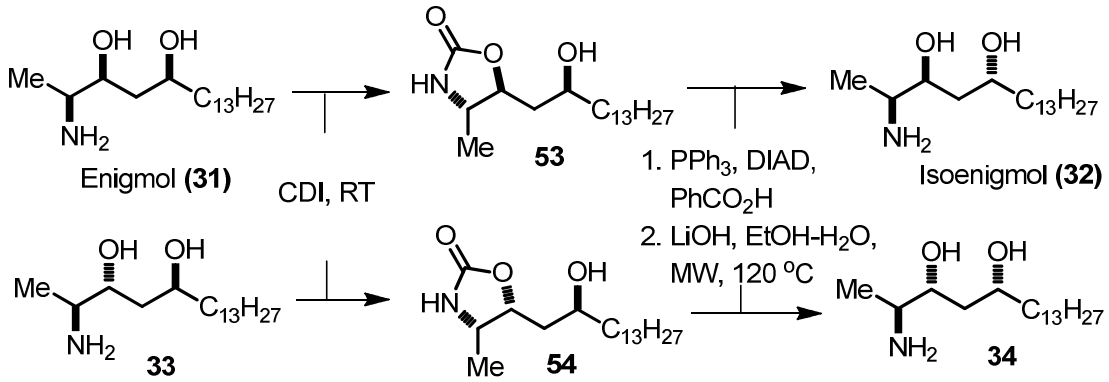
A modified approach of the above synthesis was also reported using an aldol methodology starting from the ketone **47**, obtained from the dibenzyl derivative of the aminoacid L-alanine **46** (scheme 3).[74, 76] The aldol reaction set the C5 stereocentre resulting in ketone compound **50**. The last stereocenter was then set in a diasteroselective manner using $NaBH_4$ in absense and presence of $CeCl_3$ to give deoxy compounds **51** and **52** respectively. Hydrogenation of these intermediates using Pearlman's catalyst gave the 1-deoxy-5-hydroxysphinganine compounds **31** and **33**.

Scheme 3. Aldol based approach to 1-deoxy-5-hydroxy compounds enigmol **(31)** and diastereomer **33**



Compounds isoenigmol **32** and diastereomer **34** were synthesized starting from **31** and **33** by first forming the oxazolidinone **53** and **54** regioselectively followed by Mitsunobu inversion of C5 hydroxyl group and carbamate hydrolysis using LiOH and microwave (scheme 4).[76]

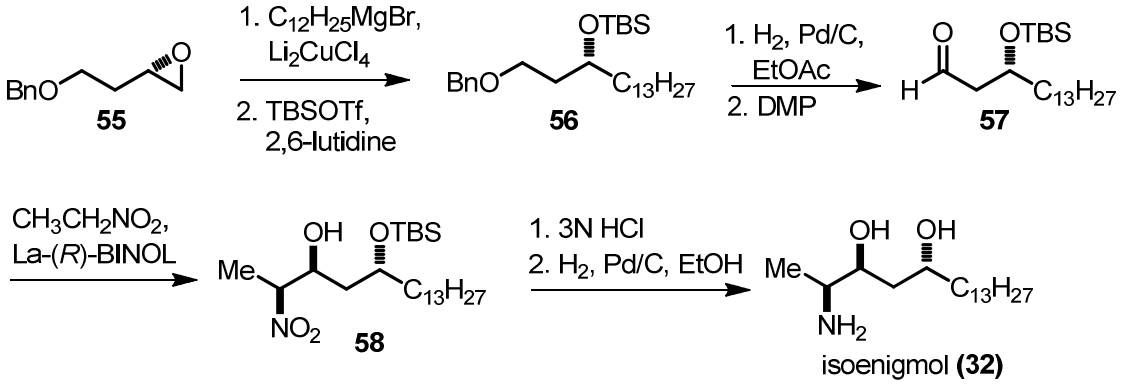
Scheme 4. Synthesis of 1-deoxy-5-hydroxy compound isoenigmol **(32)** and diastereomers **34**



2.5.2. Synthesis of 1-deoxy-5-hydroxy compound isoenigmol (32) by Sakia *et al.*

The synthesis began from the chiral epoxide **55** obtained through a Jacobsen hydrolytic kinetic resolution from racemic epoxide ***rac*-55**.[77] This epoxide **55** was opened using a cuprate formed from $C_{12}H_{25}MgBr$ to give the secondary alcohol, which was protected as the silyl ether **56** (scheme 5). Hydrogenation followed by Dess-Martin-Periodinane oxidation gave the aldeyde **57** which underwent a Henry reaction with nitroethane to give compound **58** as the only diastereomer which was then converted to isoenigmol **32** by removal of TBS and hydrogenation of the nitro group.[77]

Scheme 5. Sakia's approach to 1-deoxy-5-hydroxy compound isoenigmol **(32)**

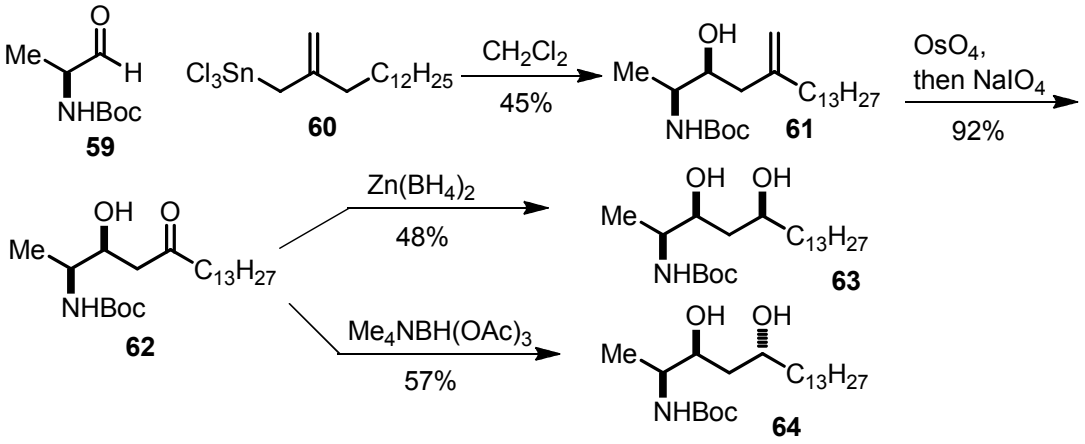


2.5.3. Synthesis of 1-deoxy-5-hydroxy analog compound, 2-*N*-Boc-amino-3,5-diols by Dias *et al.*

Dias et al used an allylation approach to synthesize the 1-deoxy-5-hydroxy analog compounds **63** and **64** (scheme 6). Allylation of α-aminoaldehyde **59** using allyltrichlorostannane **60** gave the corresponding *syn* product **61** (scheme 6).[78] A two step approach of diol formation followed by periodate clevage gave

the ketone **62**. This ketone was then selectively reduced to the required C5 stereocenter of enigmol *N*-Boc-derivative **63** and isoenigmol *N*-Boc-derivative **64** using $Zn(BH_4)_2$ and $Me_4NBH(OAc)_3$ respectively.[79]

Scheme 6. Synthesis of 1-deoxy-5-hydroxy analog compounds **63** and **64**



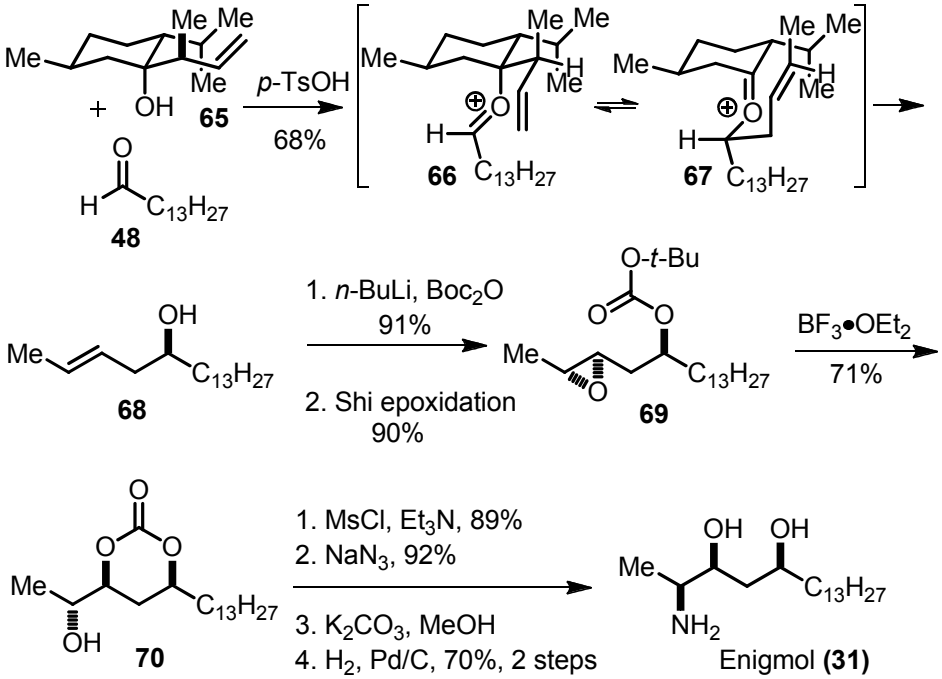
To establish the relative stereochemistry of the 3, 5-diol all the groups relied on the acetonide method reported by Rychnovsky.[36] All the above synthetic approaches used L-alanine aldehyde derivatives as their starting point. These α-aminoaldehydes are usually prone to epimerization under the reaction conditions. For a scalable synthesis and providing material for biological studies one needed a more robust and a reliable method.

2.5.4. Second generation approach towards 1-deoxy-5-hydroxysphinganine compounds enigmol (31) and isoenigmol (32)

The McDonald group entered into the foray of these molecules in collaboration with Liotta group. In this regard a second generation approach was proposed which took into account the above mentioned problems.[31] Through the recently developed allyl transfer chemistry using oxonia-[3, 3]-sigmatropic rearra-

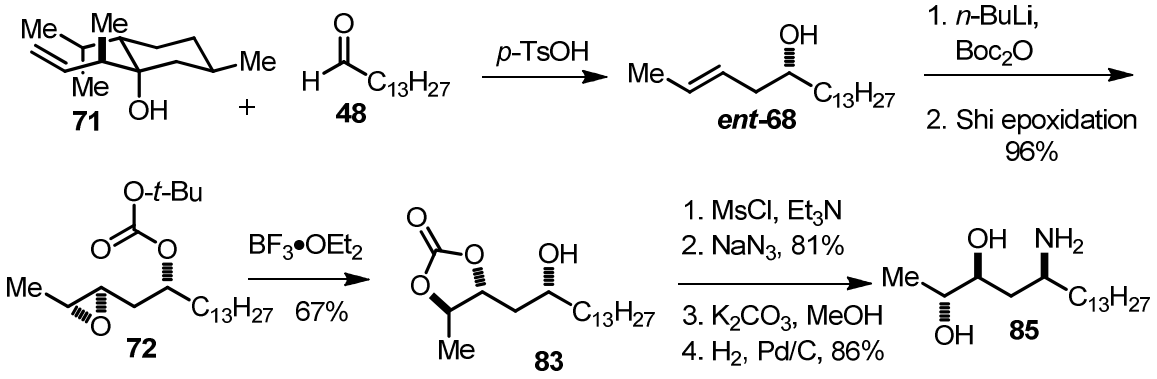
ngement by Nokami,[34] the *trans*-homoallylic alcohol **68** was prepared in high enantioselectivity and *trans* selectivity from the allyl donor **65** (scheme 7). Enigmol **(31)** was then obtained by an enantioselective Shi epoxidation followed by BF_3•OEt_2 mediated oxacyclization of the *tert*-butyl carbonate **69** to obtain the cyclic carbonate **70**.[80] Mesylation of the secondary free hydroxyl group followed by stereoselective azide substitution, methanolysis of the carbonate and hydrogenation of the azide gave enigmol **(31)**. This work was developed by John Wiseman in the McDonald laboratory.[31]

Scheme 7. McDonald approach of enigmol **(31)** through allyl transfer reaction



In a similar approach starting from the (-)-menthone allyl transfer reagent **71** and tetradecanal **48** the enantiomer of *trans*-homoallylic ***ent*-68** was synthesized. Although claimed to lead to isoenigmol **(32)**, the synthesis resulted in compound **85** (scheme 8).[31]

Scheme 8. Synthesis of isoenigmol **(32)** using allyl transfer chemistry



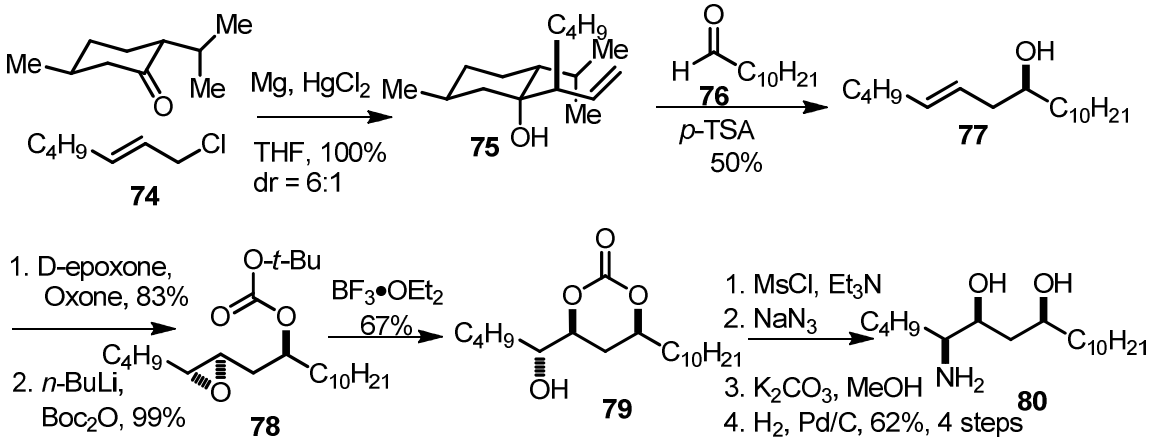
In this synthesis the relative stereochemistry of the 3, 5-diol was not confirmed by any of the methods reported in literature, specifically through acetonide formation.[36] Based on the promising biological results for enigmol and to study the role and effect of the 3 stereocentres, a synthetic endeavor to synthesize the diastereomers and some analog compounds using the established 2nd generation was undertaken. In this regard an enigmol analog **80** (scheme 9) with the sphingoid head group moved to the 5, 6, 8 position of the 18-carbon chain was explored.

2.5.5. Synthesis of internal aminodiol 1-deoxy-5-hydroxy analog compound 80

The synthesis began with the allyl tranfer reagent **75**, obtained in a dr of 6:1 from the Grignard reaction of 1-chloro-2-heptene **74** with (+)-menthone (scheme 9). The allyl transfer reaction with undecanal **76** gave the *trans*-homoallylic alcohol **77** in > 95% *ee* as determined by Mosher ester analysis.[34] Shi epoxidation of compound **77** (dr = 40:1) and *tert*-butyl carbonate formation

was followed by oxacyclization to obtain **79**. The four step sequence of mesylation and azide substitution followed by carbonate methanolysis and hydrogenation of the azide provided internal aminodiol **80**.

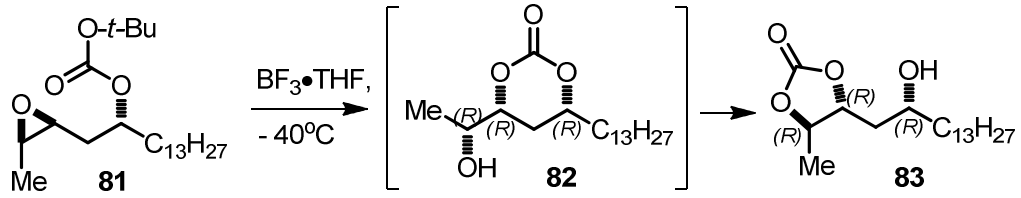
Scheme 9. Synthesis of internal aminodiol enigmol analog **80**



2.5.6. Unexpected rearrangement and establishment of regio- and stereochemistry of the synthesized 1-deoxy-5-hydroxysphinganine compounds enigmol (31) and isoenigmol (32) from 2nd generation approach

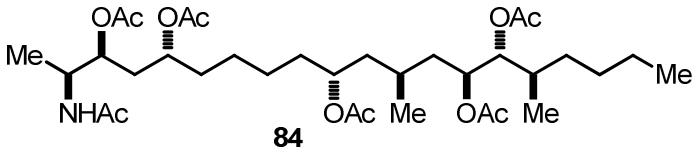
It was during the synthesis of one of the enigmol diastereomers **(34)** that an unexpected outcome was observed for the oxacyclization step for intermediate **81** (scheme 10).

Scheme 10. Unexpected outcome for oxacyclization of intermediate **81**



Two compounds were observed by TLC and characterized by NMR. When the reaction was left for a longer period of time or if the isolated compound was further treated with BF_3•OEt_2, only one major compound was observed (This was not the case with the other diastereomers synthesized). These observed results were further corroborated when the hexaacetate derivative of fumonisin B_1 synthesized in the lab by Dr. Yi-Hung Chen did not match to the data reported in the literature (Figure 8).[22] The possible explanation that was considered was the conversion of the 6-membered cyclic carbonate **82** to the 5- membered cyclic carbonate **83** (scheme 10).

Figure 8. Fumonisin B_1 hexaacetate **(84)** reported by Gurjar



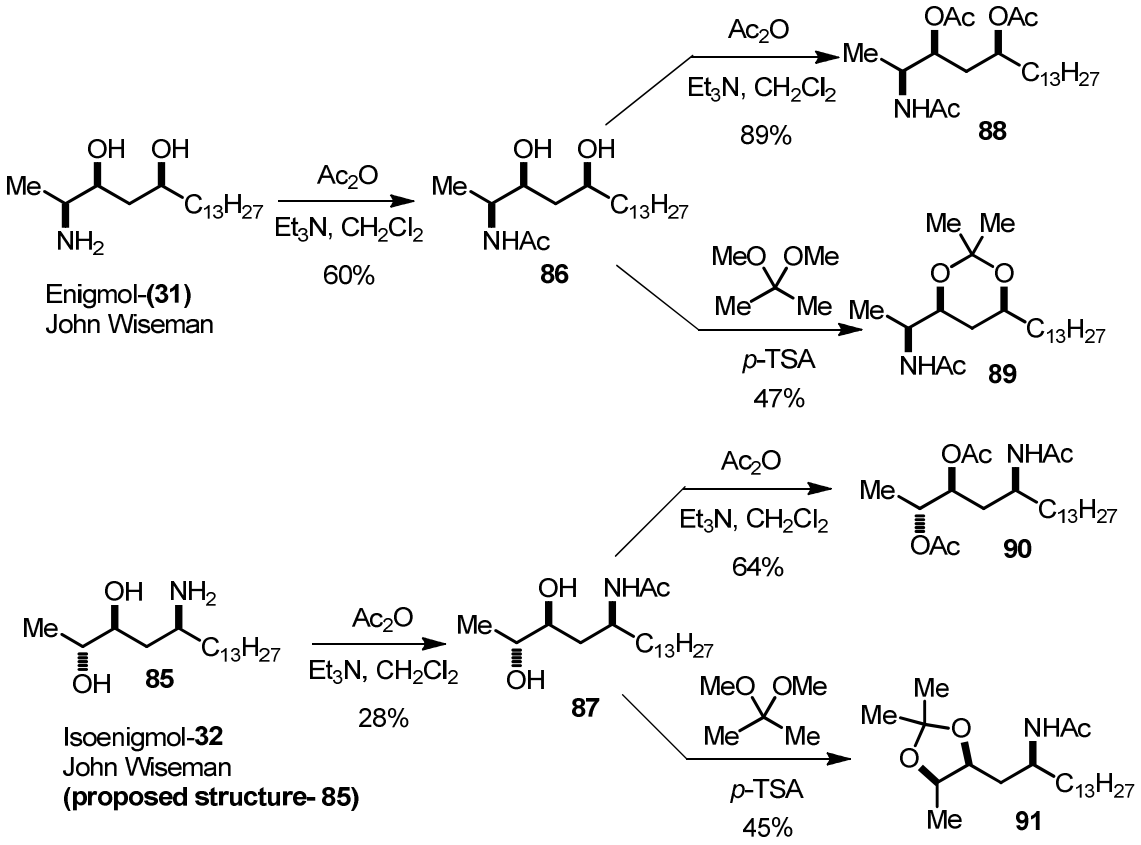
84

This outcome questioned the structure of the already synthesized 1-deoxy-5-hydroxysphinganine compounds enigmol **(31)** and isoenigmol **(32)** in the 2nd generation approach from McDonald laboratory.[31] A thorough literature search and spectroscopic studies were conducted in this regard.

First, to establish the relative stereochemistry of the 3, 5-diol, enigmol **(31)** and isoenigmol **(32)** synthesized by John Wiseman were converted to the acetonide **89** and **91** in two steps and to the peracetate compound **88** and **90** respectively (scheme 11). These compounds were also used to carry out

extensive 2D NMR studies and based on these the correct structure for isoenigmol **(32)** was proposed (now **85**, scheme **11**)

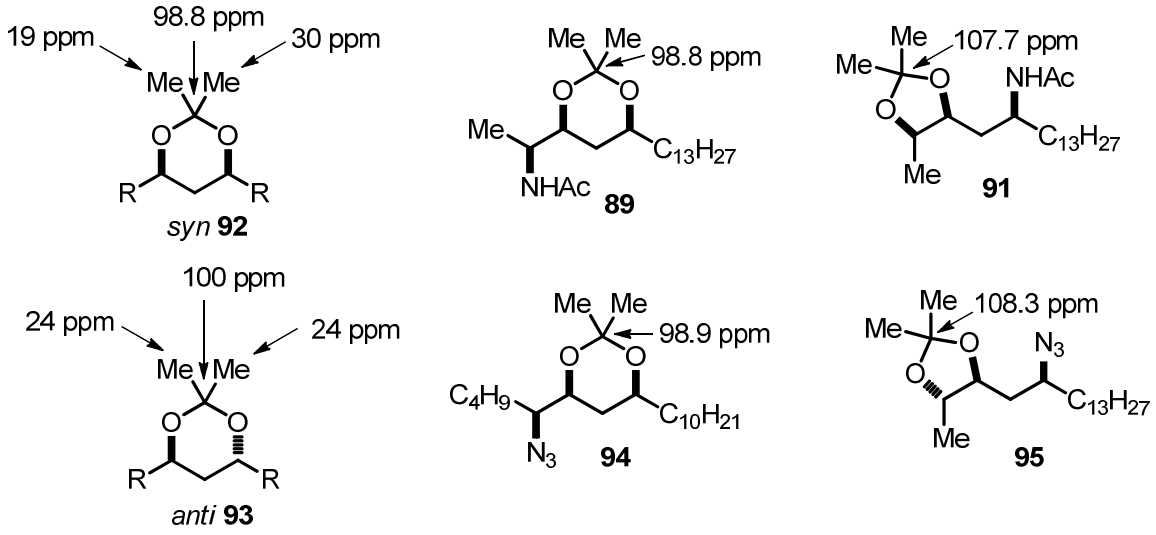
Scheme 11. Synthesis of enigmol and proposed isoenigmol derivatives



From literature it is well known that a 6-membered ring acetonide with *syn* stereochemistry has the quaternary carbon appear at ≤100 ppm (99-98) and the two methyls at 30 and 19 ppm in the ^{13}C NMR.[36] In the case of *anti*-stereochemistry this is observed at ≥100 ppm (100-101) for the quaternary and 24 ppm for the two methyls.[36] The ^{13}C data for compound **89** showed peaks at 98.9, 30.2 and 19.9 ppm, thereby confirming the relative stereochemistry of the 3, 5-diol, whereas for compound **91** it was observed at 107.7 (figure 9). A

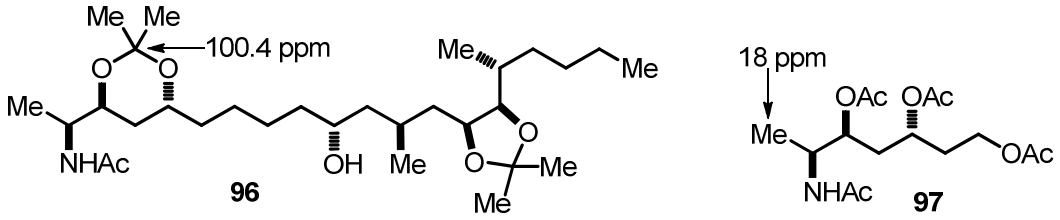
literature search for ^{13}C data for 5-membered acetonide indicated that the quaternary carbon appears between 108-110 ppm.[37]

Figure 9. Acetonide ^{13}C data for 6- vs 5- membered ring for synthesized intermediates during the synthesis of enigmol diastereomers



Shier *et al.* during their study to determine the absolute configuration of fumonisin B_1 had also shown that the ketal carbon of the 6-membered acetonide **96** appeared at 100.4 ppm in the ^{13}C NMR for the *anti*-stereochemistry of the 3, 5-diol (figure 10).[5]

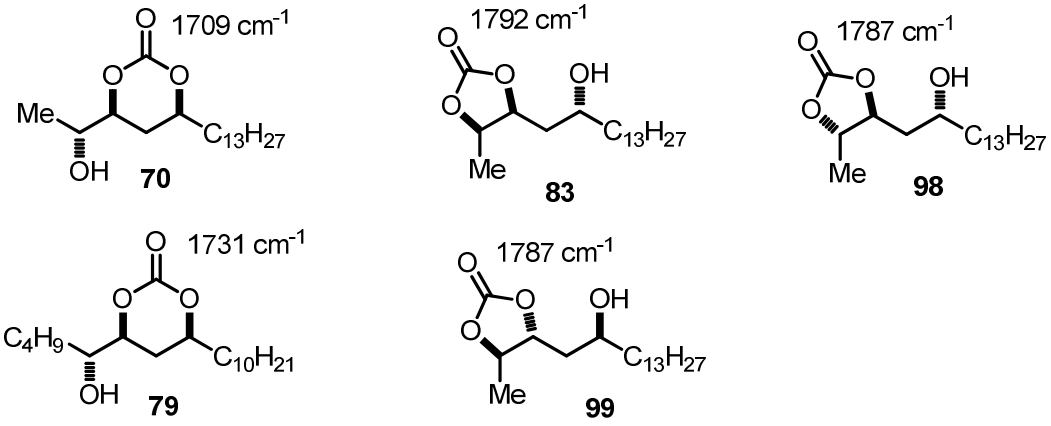
Figure 10. Literature evidence for isoenigmol **(32)** relative stereochemistry



Kishi had shown that in the peracetate derivative **97** and related compounds, the C1 methyl in the ^{13}C appears at 18 ppm when C2 amine and C3 hydroxyl are *syn* and 14 ppm if they are *anti*.[81] In the case of intermediate **88** the C1 methyl was observed at 18.6 ppm whereas for compound **90** it appeared at 14.8 ppm (scheme 53). The data was also correlated with all the other acetonides that were synthesized (figure 9). This was the first evidence that indicated towards the possibility of a 5-membered ring.

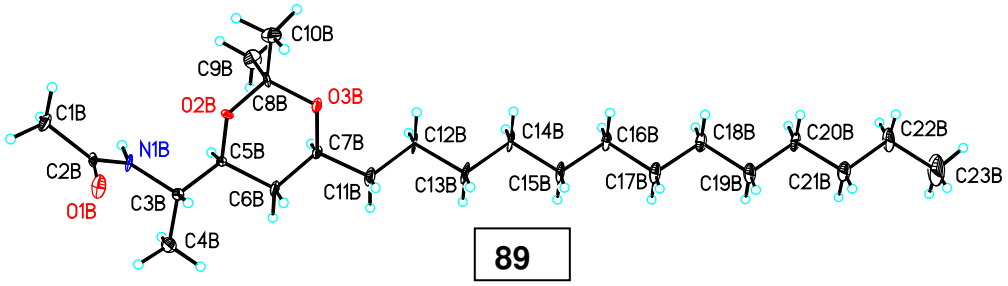
Secondly, the IR data of the cyclic carbonates were compared to what was reported in the literature. 6-membered cyclic carbonate showed the carbonyl peak at around 1750 cm^{-1} (ranged from 1740-1755),[80, 82] whereas the 5-membered cyclic carbonate showed at 1800 cm^{-1} (ranged from 1785-1810).[82] A comparison of this data with compound **70** showed a peak at 1709 cm^{-1} and that for **83** (previously **73**) to be 1792 cm^{-1}. This was also correlated with all the cyclic carbonates that were synthesized in the lab (figure 11). This was the second evidence that indicated towards a 5-membered ring.

Figure 11. IR frequncies of synthesized cyclic carbonate compounds



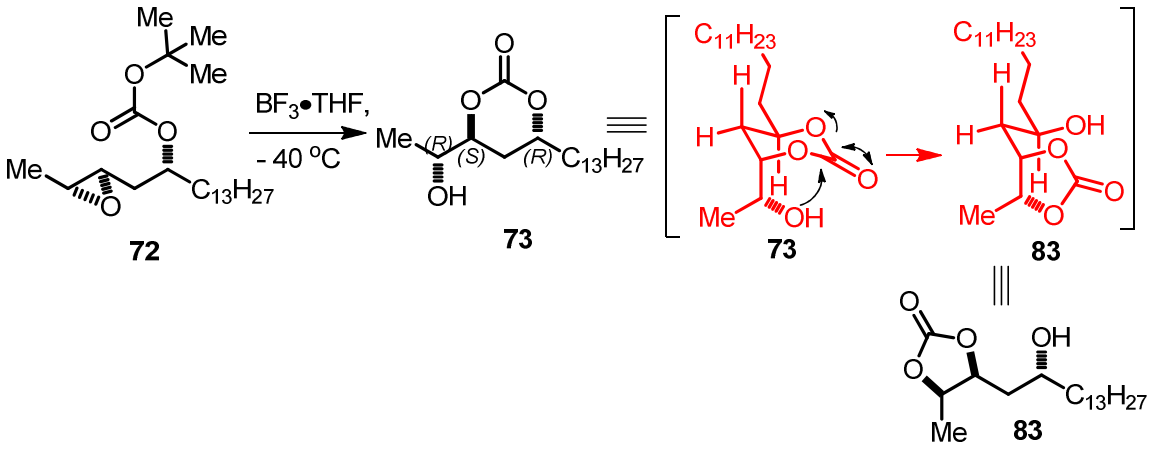
Further evidence was obtained from the 2D NMR studies like COSY and HMQC that were carried out on compounds **88**, **89**, **90** and **91**(scheme 11). A crystal structure was obtained for compound **89** which once and for all proved the stereochemistry as well as the regiochemistry of enigmol **(31)**.

Figure 12. Crystal structure of compound **89**



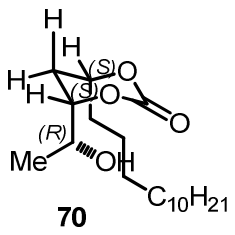
Based on all this a mechanistic rationale was proposed for the observed rearrangement from 6- to 5-membered cyclic carbonate (scheme 12).

Scheme 12. Mechanistic rationale for the observed 5-membered cyclic carbonate formation

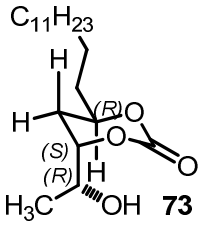


A 3D model shows that in the case of the *syn* 6-membered carbonate **70** there are steric and A (1, 3) strains involved, and this possibly prevents it from forming the 5-membered byproduct (figure 13). In the case of *anti*-compound **73** this is minimized as shown in the 3D model, and that probably leads to the 5-membered product **83**.

Figure 13. 3D representations of the 6-membered cyclic carbonates



70



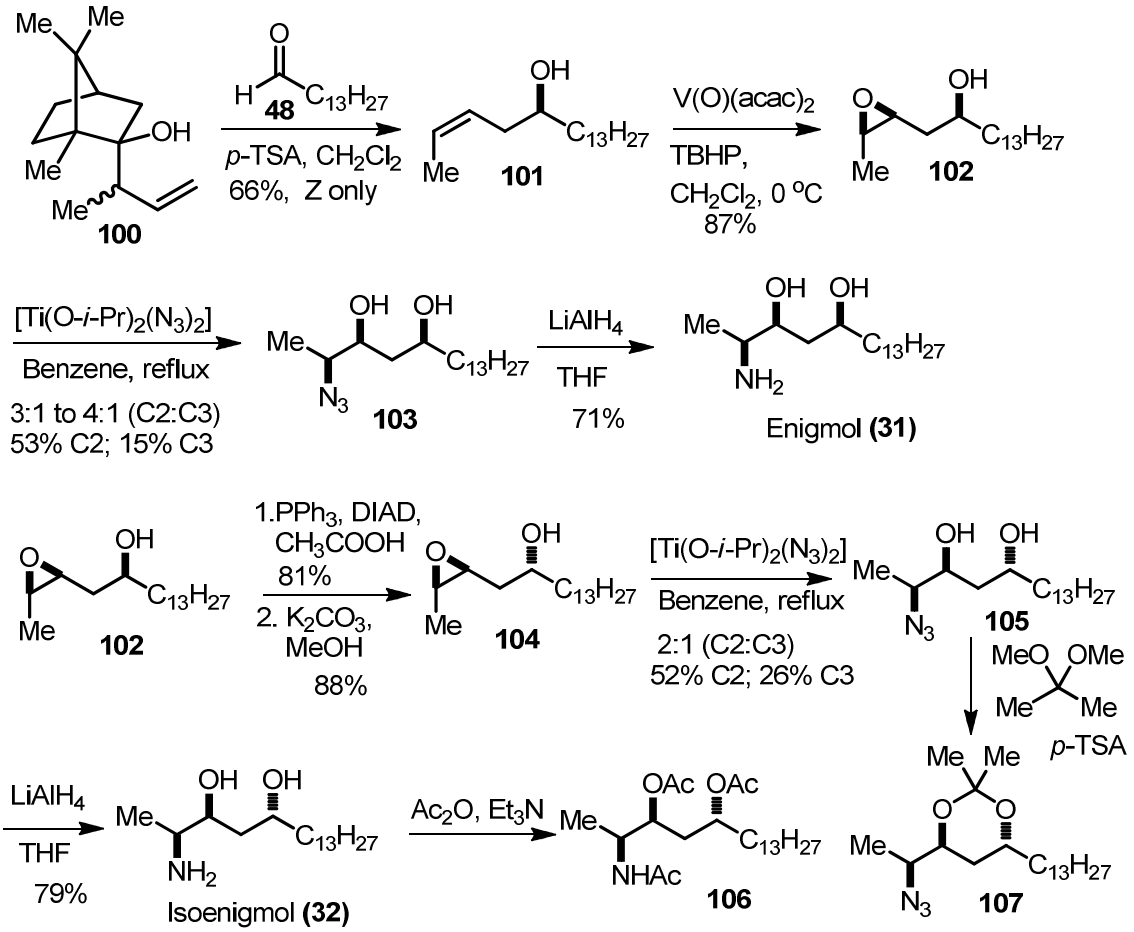
73

2.6. New approach towards the synthesis of enigmol diastereomers

2.6.1. Results and discussions

The allyl transfer methodology that was used in the 2nd generation approach was still a viable choice for the synthesis, although one had to add the azide regioselctively. This meant the addition of azide to C2 of an epoxide requiring only a single inversion of stereochemistry. *Cis*-alkene **101**, obtained from aldehyde **48** and allyl donor **100**[42] through an allyl transfer using Loh's[39] and our recently developed methodology (chapter 4) was converted to the epoxide **102** using a $VO(acac)_2$ catalysed epoxidation[43] in high diastereoselectivity from the C5 hydroxyl (scheme 13). The epoxide diasteromer **104** at C5 was obtained through a Mitsunobu inversion and methanolysis. [44]

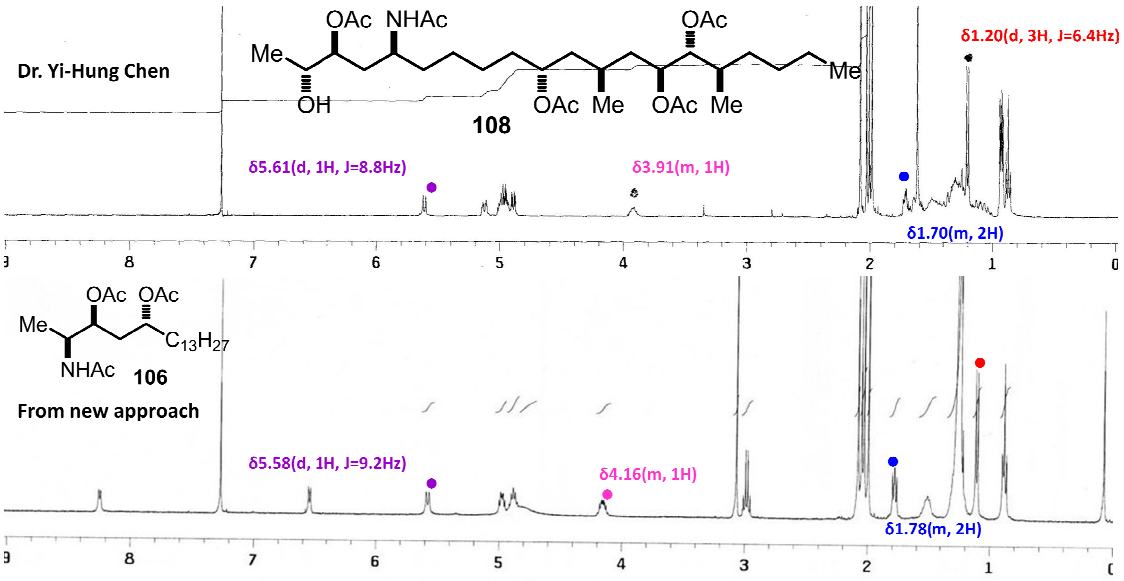
Scheme 13. Synthesis of enigmol **(31)** and isoenigmol **(32)** through the new approach



$Ti(O\text{-}i\text{-}Pr)_2(N_3)_2$ mediated opening of epoxide **102** resulted in the formation of C2 and C3 substituted azidodiols in modest selectivity, with **103** predominating.[45] (There is no precedence for the reaction of the epoxyalcohols arising from homoallylic alcohols compared to allylic alcohols). Other conditions using $LiClO_4$ and NaN_3,[46] Me_2AlN_3,[46] or chiral zirconium-azide reagents resulted in lower yield or selectivity.[46] The azide reduction of compound **103** using $LiAlH_4$ gave enigmol **(31)**. Similarly isoenigmol **(32)** was obtained starting from the epoxide **104** and was thoroughly characterized by NMR (scheme 13).

To establish the relative stereochemistry of the 3, 5 diol of isoenigmol **(31)**, intermediate **105** was converted to the acetonide **107**. The acetonide ^{13}C data for compound **107** now showed the quaternary carbon at 100.9 ppm and the two methyl at 24.5 and 24.6 ppm in accordance with the data reported in figure 9. Kishi had shown that in the peracetate derivative **97** and related compounds, the C1 methyl in the ^{13}C appears at 18 ppm when C2 amine and C3 hydroxyl are *syn* and 14 ppm if they are *anti.*[81] In compound 106 the C1 appeared at 18.6 ppm

Figure 14. ^{1}H NMR of compound **106** in comparison with Dr. Yi-Hung Chen's proposed FB_1-hexacetate **108**

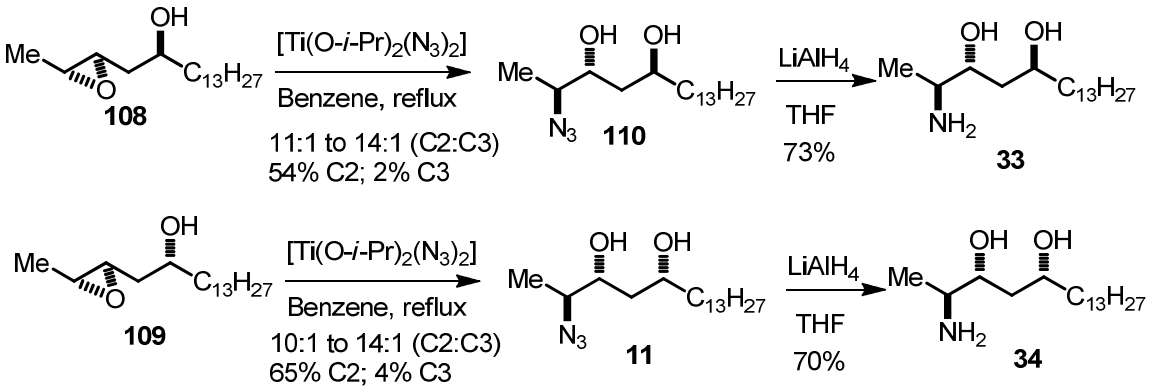


The ^{1}H NMR data of peracetate derivative **106** (figure 14) matched to the hexaacetate derivative of fumonisin B_1 **84** (figure 8), which was not the case in the first synthesis (figure 14) [Gurjar FB_1-hexacetate **84** (400 MHz, $CDCl_3$): δ 0.87 (t, *J* = 6.8 Hz, 3H), 0.93 (d, *J* = 6.8 Hz, 6H), 1.10 (d, *J* = 6.7 Hz, 3H), 1.2-1.65 (m,20H), 1.76 (m, 2H), 1.99 (s, 6H), 2.01, 2.02, 2.07, 2.09 (4s, 12H), 4.17

(m, 1H), 4.8-5.02 (m, 4H), 5.17 (dt, *J* = 2.5, 10.1 Hz, 1H), 5.58 (d, *J* = 9.3 Hz, 1H)]

Now with this new approach enigmol diasteromers **33** and **34** were synthesized starting from epoxide **108** and **109** obtained from the *trans*-alkene **68** and ***ent*-68** through Shi epoxidation (scheme 8 and 9).[31] The epoxide opening of these substrates proceeded with high regioselectivity to afford the azidodiols **110** and **111** which were reduced by $LiAlH_4$ to the respective aminoalcohols **33** and **34** respectively. The stereochemistry of these compounds was confirmed by the now established methods for similar compounds.

Scheme 14. Synthesis of diasteromers **33** and **34** using the new approach



2.6.2. Biological evaluation of 1-deoxy-5-hydroxysphinganine compounds and analogs

All the compounds were submitted to Prof. David Pallas, Emory University for biological studies and were evaluated against DU145 and HT 29 cancer cell lines. The results indicated that the diasteromers showed potency, but were not as effective as enigmol. The results are summarized below.

Table 1. Assay of enigmol and its diastereomers against DU145 cancer cell lines

DU145 Cell Killing Assays					
Date:	**So**	**Eng**	**CLP-090***	**CLP-096**	**CLP-118**
11/4/2007	11.590	4.500	15.42	12.4	15.73
11/7/2007	7.642	4.198	6.014	4.388	7.164
11/18/2007	8.006	5.369	6.482	6.069	7.802
11/30/2007	13.820	7.981	17.560	13.910	16.070
1/28/2007	8.887	6.374	14.370	6.755	8.441
2/2/2008	16.470	9.060	12.190	28.060	16.200
2/12/2008	9.511	7.281	N/A	15.900	N/A

	So	**Eng**	**CLP-090**	**CLP-096**	**CLP-118**
Avg	10.8	6.4	12.0	12.5	11.9
SD	3.3	1.8	4.8	8.1	4.5

Ttest vs. So	N/A	0.0087	0.6162	0.6272	0.6359
Ttest vs. Eng	0.0087	N/A	0.0148	0.0763	0.0126

Figure 15. Representation of the data in chart form for DU145 cell line

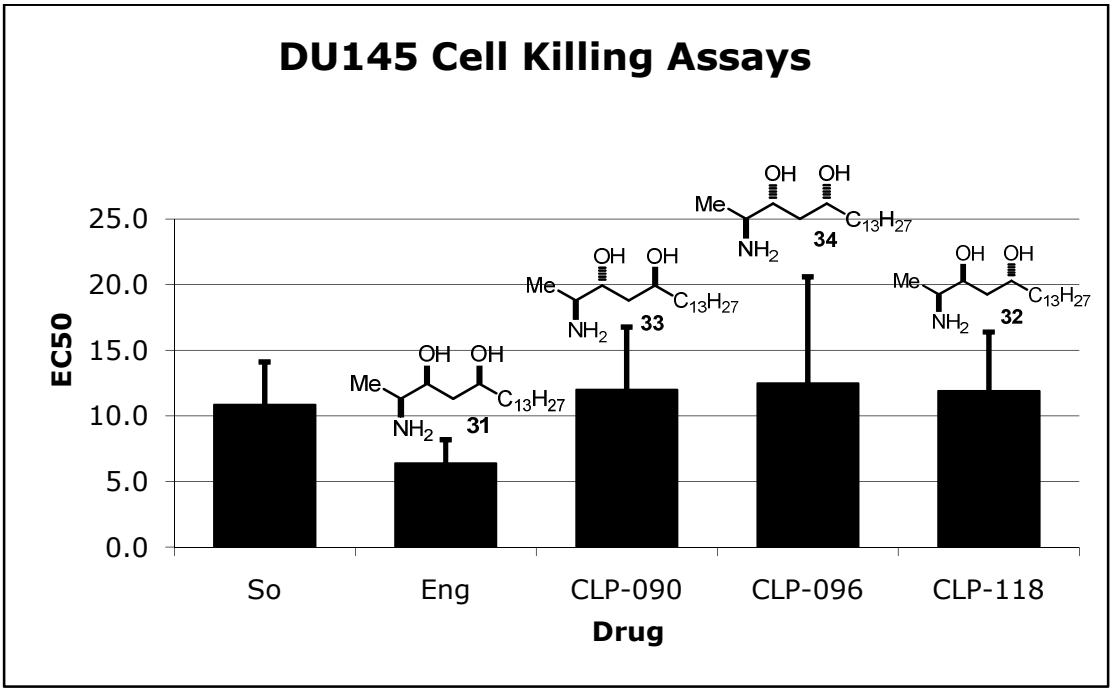


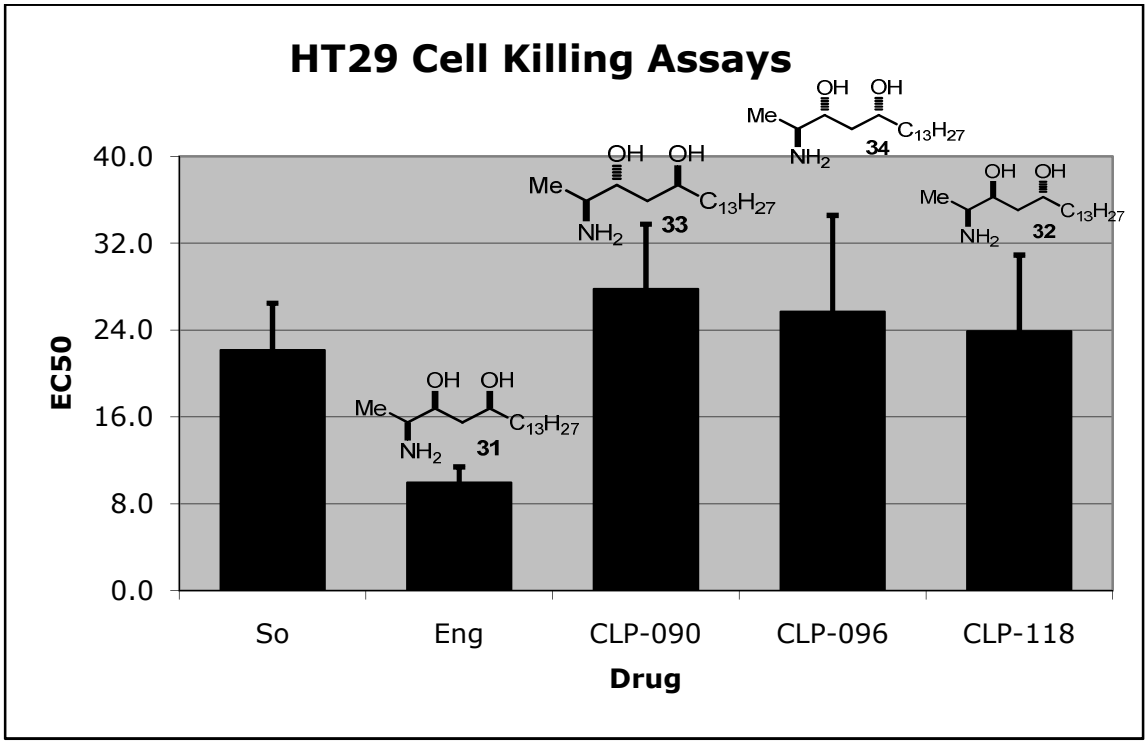
Table 2. Assay of enigmol and its diastereomers against HT29 cancer cell lines

HT29 Cell Killing Assays					
Date:	**So**	**Eng**	**CLP-090**	**CLP-096**	**CLP-118**
2/19/2008	17.14	8.30	20.91	15.85	17.31
3/2/2008	24.69	10.44	31.18	28.29	23.12
3/6/2008	24.63	11.12	31.32	33.01	31.28

	So	**Eng**	**CLP-090**	**CLP-096**	**CLP-118**
Avg	22.2	10.0	27.8	25.7	23.9
SD	4.3	1.5	6.0	8.9	7.0

Ttest vs. So	N/A	0.0100	0.2556	0.5657	0.7320
Ttest vs. Eng	0.0100	N/A	0.0073	0.0385	0.0281

Figure 16. Representation of the data in chart form for HT29 cell line



Enigmol **(31)** had shown potential anticancer activity against the National Cancer Institute's 60 cell lines.[83] According to the study carried out by Merrill *et al.* enigmol showed anticancer activity against colon cancer (HT29 cancer cell line) and prostate cancer (DU145 and PC3 cancer cell line). The studies were also carried *in vivo* on Min mice and nude mouse xenografts wherein enigmol was administered intravenously and orally and was found to suppress the tumor growth by half without causing any host toxicity.[83] Enigmol **(31)** thus represents a novel compound that has potential to be effective against multiple types of cancer.

2.7. Experimental details

General information: ^{1}H NMR and ^{13}C NMR spectra were recorded on Varian INOVA 600, Unity 600 and INOVA 400 spectrometers. NMR spectra were recorded in solutions of deuterated chloroform ($CDCl_3$) with the residual chloroform (7.27 ppm for ^{1}H NMR and 77.23 ppm for ^{13}C NMR) taken as the internal standard, deuterated methanol (CD_3OD) with residual methanol (3.31 ppm for ^{1}H NMR and 49.3 ppm for ^{13}C NMR) taken as the internal standard, or deuterated benzene with residual benzene (7.16 ppm for ^{1}H NMR and 128.23 ppm for ^{13}C NMR) taken as the internal standard, and were reported in parts per million (ppm). Abbreviations for signal coupling are as follows: s, singlet; d, doublet; t, triplet; q, quartet; dd, doublet of doublet; ddd, doublet of doublet of doublet; dt, doublet of triplet; m, multiplet.

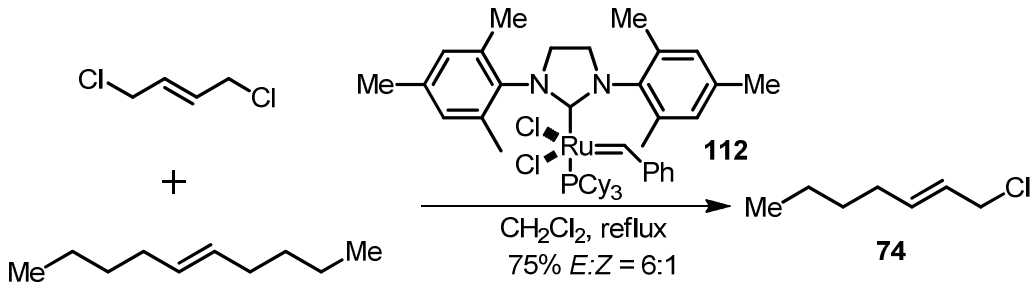
IR spectra were collected on a Mattson Genesis II FT-IR spectrometer as neat films on sodium chloride discs. Mass spectra (high resolution ESI and APCI) were recorded on a Finnigan LTQ FTMS Mass spectrometer. Optical rotations were measured using a Perkin-Elmer 341 polarimeter (concentration in g/100mL). Thin Layer Chromatography (TLC) was performed on precoated glass backed plates purchased from Whatman (silica gel $60F_{254}$; 0.25mm thickness). Flash column chromatography was carried out with silica gel 60 (230-400 mesh ASTM) from EM Science.

All reactions were carried out with anhydrous solvents in oven dried or flame dried and argon-charged glassware. All anhydrous solvents were dried with 4 Å molecular sieves purchased from Sigma-Aldrich and tested for trace

water content with Coulometric KF titrator from Denver instruments. All solvents used in extraction procedures and chromatography were used as received from commercial suppliers without prior purification.

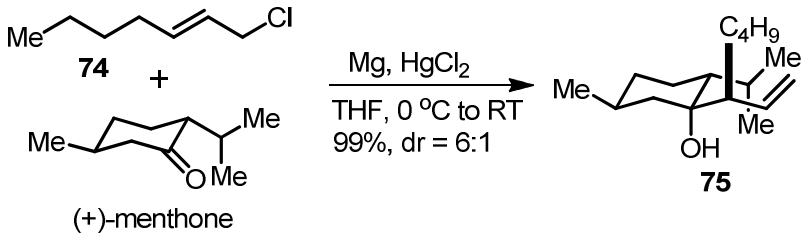
Synthesis of the internal aminodiol 80

Synthesis of vinyl chloride 74



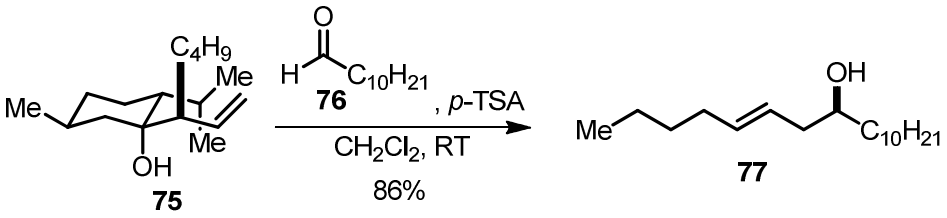
Dissolved *trans*-5-decene (2.0 g, 14.2 mmol) and 1,4-dichloro-2-butene (4.4 g, 35.6 mmol) in dichloromethane (48 mL) and degasified the reaction mixture. Grubbs 2nd generation catalyst **112** (0.06 g, 0.07 mmol) was then added and the reaction mixture heated to reflux overnight. Dichloromethane was then removed under vacuum and purified the crude by flash chromatography using 5% ether in pentane as eluent to obtain the vinyl chloride **74** as yellow oil (2.8 g, 75%) **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.80-5.76 (m, 1H), 5.72-5.66 (m, 1H), 3.95 (d, *J* = 7.2 Hz, 2H), 2.09-2.05 (m, 2H), 1.39-1.30 (m, 5H), 0.93-0.89 (m, 4H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 136.9, 126.4, 33.9, 31.9, 31.1, 22.3, 14.0; **FT-IR**: 2595, 2955, 2917, 2849, 1468, 1349, 1067, 869, 720 cm^{-1} .

Synthesis of allyl donor reagent 75



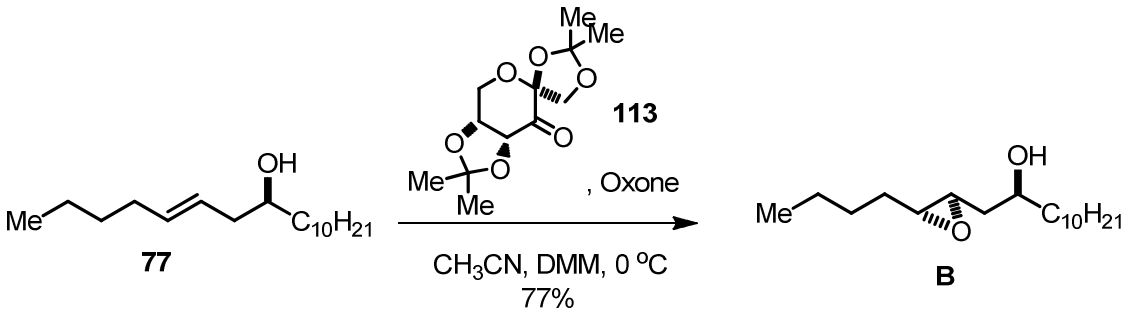
THF (31 mL) was added to flame dried Mg turnings (0.44 g, 18.0 mmol) and cooled to 0 °C. Mercuric chloride (0.007 g, 0.014 mmol) was then added followed by 1-chloro-2-heptene **74** (2.4 g, 13.5 mmol) dropwise and stirred the reaction mixture at RT for 30 min. The reaction was cooled back to 0 °C and (+)-menthone (1.4 g, 9.07 mmol) was added dropwise over a period of 15 min. The reaction was stirred at room temperature for 2 h before quenching it slowly with ice and water. The aqueous layer was extracted with ethyl acetate and the organic layer washed with brine, dried over $MgSO_4$, filtered and concentrated to obtain oil. The crude was purified by flash chromatography using 10% ether in pentane as eluent to obtain compound **75** as yellow oil (1.9 g, 99%) **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.67 (dt, *J* = 9.6, 17.4 Hz, 1H), 5.23 (dd, *J* = 2.4, 10.2 Hz, 1H), 5.15 (dd, *J* = 1.8, 17.1 Hz, 1H), 2.34 (t, *J* = 10.2 Hz, 1H), 2.04-1.99 (m, 1H), 1.79-1.73 (m, 1H), 1.58-1.48 (m, 2H), 1.43 (s, 1H), 1.39-1.22 (m, 6H), 1.21-1.09 (m, 2H), 0.98 (t, *J* = 12.6 Hz, 1H), 0.92-0.88 (m, 9H), 0.84 (d, *J* = 6.0 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 140.0, 119.4, 75.8, 52.3, 46.1, 42.9, 35.5, 30.7, 28.7, 27.7, 25.3, 23.5, 22.8 22.8, 20.8, 18.1, 14.2; **HRMS (ESI)**: *m/z* calcd. For $C_{17}H_{36}NO$ ($M+NH_4^+$) 285.2788, found 285.2788; **FT-IR**: 3375, 2955, 2917, 2849, 1468, 1349, 1067, 869, 720 cm^{-1} ; **$[\alpha]_D^{25}$** = + 3.2 (c=0.525, $CHCl_3$)

Synthesis of *trans*-homoallylic alcohol 77



Allyl donor reagent **75** (1.6 g, 6.4 mmol) and aldehyde **76** (0.55 g, 3.22 mmol) were taken in CH_2Cl_2 (28 mL). *p*-TSA (0.06 g, 0.32 mmol) was then added and the reaction mixture stirred at room temperature for 20 h. The reaction was quenched with $NaHCO_3$ and extracted the aqueous layer with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain yellow oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain homoallylic alcohol **77** (0.75 g, 86%) as a white solid. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.57-5.52 (m, 1H), 5.43-5.38 (m, 1H), 3.61-3.55 (m, 1H), 2.26-2.22 (m, 1H), 2.08-2.01 (m, 3H), 1.58 (d, *J* = 4.2 Hz, 1H), 1.46-1.40 (m, 3H), 1.37-1.26 (m, 19H), 0.91 (m, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 135.0, 126.0, 71.0, 40.9, 36.9, 32.5, 32.1, 31.8, 29.8 (2C), 29.5, 25.9, 22.8, 22.4, 14.3, 14.1; **FT-IR**: 3344, 3028, 2955, 2923, 2853, 1466, 1349, 1077, 968 cm^{-1} ; **Anal. Calcd. for $C_{18}H_{36}O$**: C, 80.53; H, 13.52; O, 5.96 found: C, 80.35; H, 13.49; O, 6.16; **Mp**: 26-28 ^oC; **$[\alpha]_D^{25}$** = + 0.7 (c=0.41, $CHCl_3$).

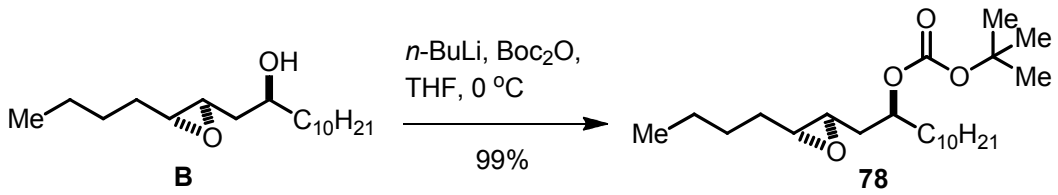
Synthesis of epoxy-alcohol B



Homoallylic alcohol **77** (0.74 g, 2.75 mmol) was dissolved in a mixture of acetonitrile (20.7 mL) and dimethoxymethane (41 mL). A solution of 0.05 M $Na_2B_4O_7.10H_2O$ in 4×10^{-4} M EDTA (40 mL) was added followed by tetrabutyl ammoniumhydrogensulphate (0.03 g, 0.08 mmol) and Shi ketone **113** (0.21 g, 0.81 mmol). The solution was cooled to 0 °C and a solution of oxone (2.71 g, 4.4 mmol) in 4×10^{-4} M EDTA (12.4 mL) and K_2CO_3 (2.56 g, 18.5 mmol) in water (12.4 mL) was added simultaneously over a period of 1.5 h. Stirred for 30 min after addition and diluted the reaction mixture with hexanes. Separated the organic layer and extracted the aqueous with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain epoxyalcohol **B** (0.6 g, 77%) as a white solid. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.82-3.76 (m, 1H), 2.92-2.90 (m, 1H), 2.83-2.81 (m, 1H), 2.07 (d, *J* = 3.6 Hz, 1H), 1.81 (ddd, *J* = 3.6, 8.7, 14.7 Hz, 1H), 1.63 (ddd, *J* = 3.0, 6.0, 14.2 Hz, 1H), 1.57-1.53 (m, 2H), 1.50-1.34 (m, 7H), 1.33-1.26 (m, 15H), 0.91 (t, *J* = 7.2 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 69.6, 58.6, 56.8, 38.6, 37.7, 32.1, 31.9, 29.8, 29.5, 28.2, 25.7, 22.8, 22.7, 14.3, 14.2; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{37}O_2$ ($M+H^+$) 285.2788, found 285.2788; **FT-IR**: 3375, 2955, 2917, 2849,

1468, 1349, 1067, 869, 720 cm^{-1} ; **Anal. Calcd. for $C_{18}H_{36}O_2$**: C, 76.00; H, 12.76; O, 11.25 found: C, 74.99; H, 12.70; O, 10.92; **Mp**: 44-46 ^oC; **$[\alpha]_D^{25}$** = + 19.1 (c=0.745, $CHCl_3$).

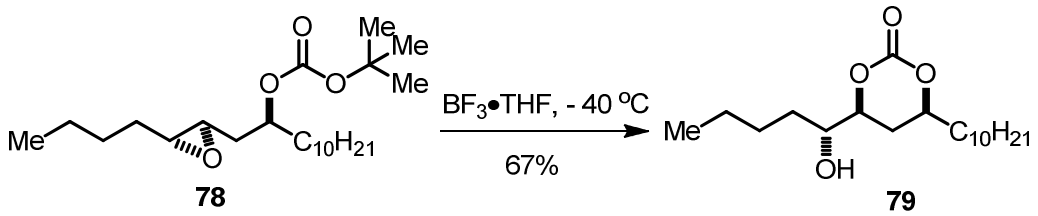
Synthesis of *tert*-butyl epoxycarbonate 78



Epoxy alcohol **B** (0.58 g, 2.03 mmol) was dissolved in THF (15.5 mL) and cooled to 0 ^oC. *n*-BuLi (0.15 g, 2.34 mmol, 2.5 M in hexanes) was then added dropwise and the reaction mixture stirred for 5 min. Boc anhydride (0.88 g, 4.03 mmol) was then added and the reaction mixture stirred at room temperature for 2 h. The reaction mixture was quenched with NH_4Cl and extracted the aqueous with CH_2Cl_2. The organic layer was then washed with brine, dried over $MgSO_4$, filtered and concentrated to obtain the crude as oil. Purified the crude by flash chromatography using 2% ethyl acetate in hexanes as eluent to obtain compound **78** as yellow oil (0.77 g, 99%) **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.84-4.79 (m, 1H), 2.74 (dt, *J* = 1.8, 5.7 Hz, 1H), 2.69 (dt, *J* = 1.8, 5.7 Hz 1H), 1.81-1.73 (m, 2H), 1.66-1.63 (m, 1H), 1.61-1.55 (m, 2H), 1.55-1.50 (m, 5H), 1.49 (s, 9H) 1.46-1.33 (m, 5H) 1.32-1.25 (m, 15H), 0.90 (t, *J* = 7.2 Hz, 3H), 0.88 (t, *J* = 6.9 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 153.5, 82.1, 75.3, 59.2, 55.7, 37.4, 34.8, 32.1, 31.9, 29.8, 29.7 (2C), 29.6, 29.5, 28.2, 27.6 25.4, 22.9, 22.7, 14.3, 14.2; **HRMS (ESI)**: *m/z* calcd. for $C_{23}H_{45}O_4$ ($M+H^+$) 385.3312, found 385.3312; **FT-IR**: 2956, 2927, 2856, 1738, 1461, 1368, 1278, 1166, 1119, 872 cm^{-1} ; **Anal. Calcd. for**

$C_{23}H_{44}O_4$: C, 71.83; H, 11.53; O, 16.64 found: C, 71.89; H, 11.56; O, 16.55; **$[\alpha]_D^{25}$** = + 12.0 (c=0.535, $CHCl_3$).

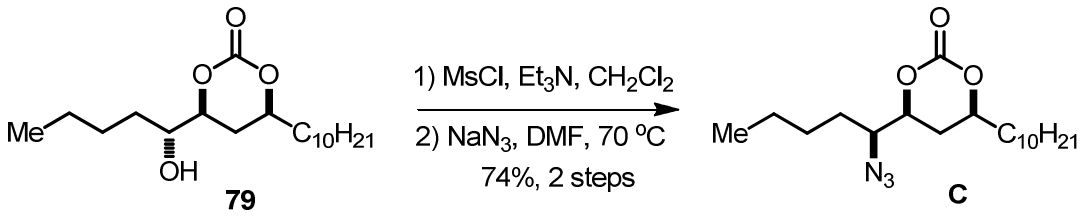
Synthesis of cyclic carbonate 79



78 **79**

Dissolved compound **78** (0.65 g, 1.67 mmol) in anhydrous CH_2Cl_2 (33 mL) and cooled to -40 °C. BF_3•THF (0.23 g, 1.64 mmol) was then added drop wise and the reaction mixture stirred at -40 °C for 1 h. The reaction mixture was quenched with saturated $NaHCO_3$ and the aqueous extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as a white solid. Purified the crude by flash chromatography using 30% ethyl acetate in hexanes as eluent to obtain the cyclic carbonate **79** as white solid (0.37 g, 67%) **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.44-4.39 (m, 1H), 4.35 (dt, *J* = 3.6, 12.0 Hz, 1H), 3.84 (dt, *J* = 4.2, 8.2 Hz, 1H), 2.10 (d, *J* = 4.2 Hz, 1H), 1.99 (dt, *J* = 3.0, 13.8 Hz, 1H) 1.93-1.87 (m, 1H), 1.79-1.73 (m, 1H), 1.69-1.61 (m, 1H), 1.56-1.44 (m, 4H), 1.42-1.30 (m, 9H) 1.26 (s, 9H), 0.90 (t, *J* = 7.2 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 149.7, 81.7, 78.8, 71.9, 35.5, 32.0, 31.4, 29.7 (2C), 29.6, 29.5, 28.0, 26.7, 24.6, 22.8, 22.7, 14.3, 14.1; **HRMS (ESI)**: *m/z* calcd. for $C_{19}H_{37}O_4$ ($M+H^+$) 329.2686, found 329.2688; **FT-IR**: 3435, 2925, 2855, 1731, 1466, 1402, 1247, 1117, 769 cm^{-1} ; **Anal. Calcd. for $C_{19}H_{36}O_4$**: C, 69.47; H, 11.05; O, 19.48 found: C, 69.48; H, 11.19; O, 19.58; **Mp**: 50-52 °C; **$[\alpha]_D^{25}$** = -7.7 (c=1.14, $CHCl_3$).

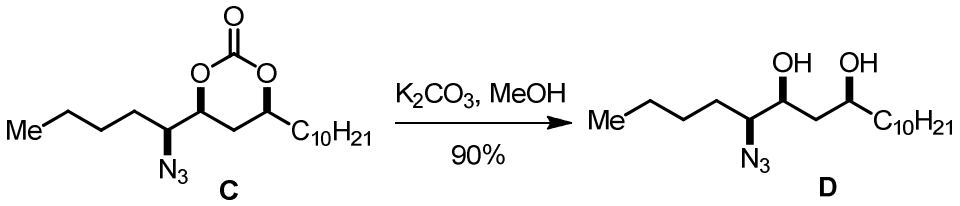
Synthesis of azidocarbonate C



Cyclic carbonate **79** (0.3 g, 0.91 mmol) was dissolved in CH_2Cl_2 (9 mL) and cooled to 0 ^oC. Triethylamine (0.06 g, 1.01 mmol) was then added followed by methanesulphonyl chloride (0.11 g, 0.95 mmol). The reaction mixture was stirred at 0 ^oC for 2 h, before quenching with water. The aqueous layer was extracted with CH_2Cl_2, and the organic layer dried over $MgSO_4$, filtered and concentrated to obtain the crude as oil. The crude mesylate was then taken in DMF (9 mL), and NaN_3 (0.47 g, 7.2 mmol) was added. The reaction mixture was heated at 70 ^oC for 24 h. Water and CH_2Cl_2 were added in equal proportions and extracted the aqueous layer with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as colorless oil. Purified the crude by flash chromatography using 20% ethyl acetate in hexanes as eluent to obtain azidocarbonate **C** as white solid (0.24 g, 74%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 4.46 (dt, *J* = 3.6, 12.0 Hz, 1H), 4.41-4.39 (m, 1H), 3.31 (ddd, *J* = 4.2, 7.2 Hz, 1H), 1.97 (dt, *J* = 11.4, 14.4 Hz, 1H), 1.80-1.74 (m, 1H), 1.74-1.70 (m, 2H), 1.66-1.60 (m, 1H), 1.53-1.45 (m, 2H), 1.44-1.34 (m, 4H) 1.33-1.26 (m, 14H), 0.90 (t, *J* = 7.8 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 148.8, 79.5, 78.6, 63.6, 35.4, 32.0, 29.77, 29.7, 29.6, 29.5, 29.4, 29.2, 28.4, 24.6 , 22.8, 22.6, 14.3, 14.1; **HRMS (ESI)**: *m/z* calcd. for $C_{19}H_{36}N_3O_3$ ($M+H^+$) 354.2751, found 354.2751; **FT-IR**: 2955, 2920, 2100, 1723, 1471, 1280, 1096 cm^{-1} ; **Anal. Calcd. for**

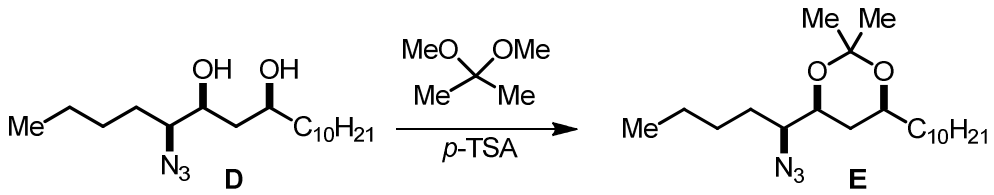
$C_{19}H_{35}N_3O_3$: C, 64.56; H, 9.98; N, 11.89; O, 13.58 found: C, 64.56; H, 9.98; N, 11.81; O, 13.51; **Mp**: 47-49 ^oC; **$[\alpha]_D^{25}$** = - 0.6 (c =1.365, $CHCl_3$).

Synthesis of azidodiol D



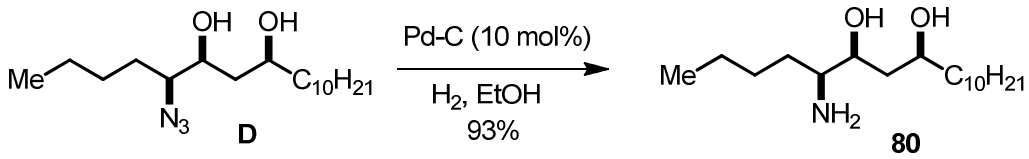
Azido carbamate **C** (0.21 g, 0.59 mmol) and K_2CO_3 (0.57 g, 4.15 mmol) were taken in methanol (6.0 mL) and stirred at room temperature for 4 h. Water and CH_2Cl_2 were added in equal proportions and extracted the aqueous with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain azidodiol **D** (0.17 g, 90%) as yellow oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.87-3.84 (m, 2H), 3.47 (s, 1H), 3.16-3.13 (m, 1H), 2.62 (s, 1H), 1.70-1.59 (m, 4H), 1.54-1.44 (m, 3H), 1.43-1.35 (m, 4H), 1.34-1.26 (m, 16H), 0.93 (t, *J* = 7.8 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 74.7, 73.0, 67.2, 39.8, 38.5, 32.1, 30.1, 29.7, 29.5, 28.6, 25.4, 22.8, 22.7, 14.3, 14.1; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{38}N_3O_2$ ($M+H^+$) 328.2958, found 328.2963; **FT-IR**: 3358, 2924, 2855, 2101, 1463, 1261, 1082, 848 cm^{-1} ; **Anal. Calcd. for $C_{18}H_{37}N_3O_2$**: C, 66.01; H, 11.39; N, 12.83; O, 9.77 found: C, 66.15; H, 11.47; N, 12.59; O, 9.79; **$[\alpha]_D^{25}$** = + 5.8 (c=0.945, $CHCl_3$).

Synthesis of azidoacetonide E



^{1}H NMR (400 MHz, $CDCl_3$) δ 3.87 (ddd, *J* = 2.8, 5.8, 11.8 Hz, 1H), 3.82 - 3.77 (m, 1H), 3.11-3.07 (m, 1H), 1.55-1.46 (m, 4H), 1.44-1.41 (m, 8H), 1.39-1.30 (m, 7H), 1.26 (bs, 13H), 0.92 (t, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 98.9, 72.1, 68.9, 65.5, 36.6, 33.6, 32.1, 30.2, 29.8, 29.8, 29.7, 29.5, 29.3, 28.5, 25.1, 22.9, 22.6, 19.8, 14.3, 14.1.

Synthesis of aminodiol 80

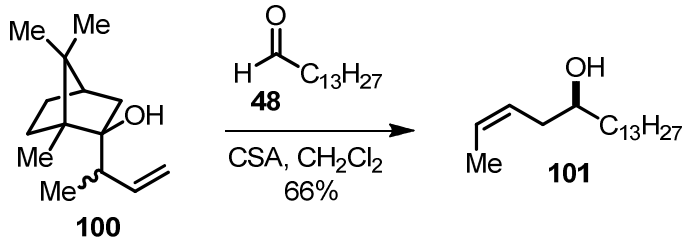


Azidodiol **D** (0.14 g, 0.42 mmol) was dissolved in absolute ethanol (4.3 mL) and purged argon for 5 min before adding Pd-C (0.014 g, 10 mol%). Evacuated the flask before back filling with hydrogen and stirred at room temperature for 8 h. Filtered of the palladium through celite and rinsed the celite with fresh ethanol. Ethanol was removed under vacuum to obtain aminodiol **80** (0.125 g, 93%) as a white solid. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.87 (bs, 1H), 3.62-3.58 (m, 1H), 3.25-2.7 (bs 3H), 2.64-2.62 (m, 1H), 1.65 (d, *J* = 14.4 Hz, 1H), 1.60-1.54 (m, 1H), 1.53-1.47 (m, 2H), 1.45-1.39 (m, 3H), 1.37-1.26 (m, 18H) 0.92 (t, *J* = 7.2 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 74.6, 71.9, 56.2, 40.9, 38.1, 34.0, 32.1, 29.85, 29.8, 29.5, 28.6, 25.7, 22.8, 14.3, 14.1; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{40}NO_2$ ($M+H^+$) 302.3053, found 302.3051; **FT-IR**: 3444, 3367,

2922, 2852, 1573, 1463, 1331, 1143, 931, 846 cm^{-1} ; **Anal. Calcd. for $C_{18}H_{39}NO_2$**: C, 71.70; H, 13.04; N, 4.65; O, 10.61 found: C, 71.04; H, 12.82; N, 4.31; O, 10.86; **Mp**: 50-52 ^oC; **$[\alpha]_D^{25}$** = - 7.5 (c=0.7, $CHCl_3$).

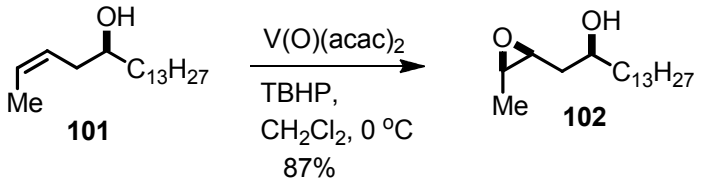
Synthesis of enigmol 31

Synthesis of *cis*-homoallylic alcohol 101



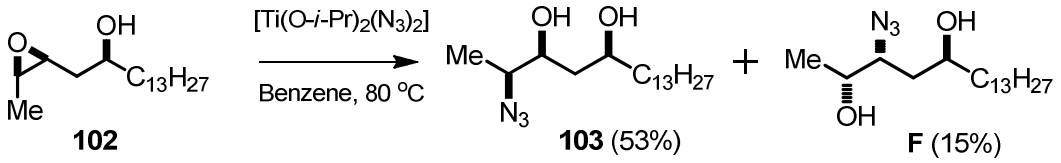
Taken the camphor derivative **100** (11.7 g, 56.1 mmol) and 1-tetradecanal **48** (4.0 g, 18.8 mmol) in CH_2Cl_2 (3 mL). CSA (0.44 g, 1.89 mmol) was then added and the reaction mixture stirred at room temperature for 5-6 days. The reaction mixture was diluted with CH_2Cl_2 and dried the organic layer over $MgSO_4$, filtered and concentrated to obtain colorless oil. Purified the crude by flash chromatography using 6% ethyl acetate in hexanes as eluent to obtain homoallylic alcohol **101** as white solid (3.3 g, 66%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.68-5.62 (m, 1H), 5.46-5.42 (m, 1H), 3.64-3.62 (m, 1H), 2.22 (t, *J* = 6.6 Hz, 2H), 1.64 (dd, *J* = 1.2, 6.6 Hz, 3H), 1.58 (s, 1H), 1.49-1.44 (m, 3H), 1.31-1.26 (m, 21H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 127.4, 126.4, 71.7, 37.0, 35.1, 32.1, 29.9, 29.8, 29.5, 25.9, 22.9, 14.3, 13.2 ; **HRMS (APCI)**: *m/z* calcd. for $C_{18}H_{35}O$ (M^+) 267.2682 found 267.2683; **FT-IR**: 3345, 3021, 2915, 2848, 1469 cm^{-1} ; **Mp**: 28-29 ^oC; **$[\alpha]_D^{25}$** = - 2.7 (c=1.03, $CHCl_3$).

Synthesis of epoxy alcohol 102



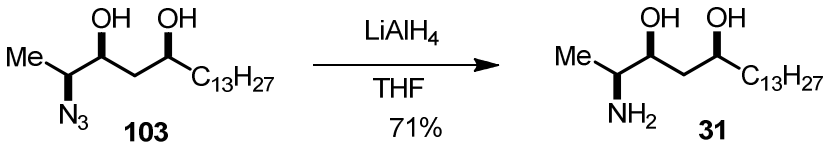
Homoallylic alcohol **101** (3.25 g, 12.1 mmol) and $V(O)(acac)_2$ (0.16 g, 0.60 mmol) where dissolved in CH_2Cl_2 (120 mL) and the resulting solution was cooled to 0 °C. *t*-Butyl hydroperoxide (TBHP, 5.5 M in decane, 3.3 mL, 18.1 mmol) was then added dropwise and the reaction mixture stirred at 0 °C for 3 h before stirring it for another 24 h at room temperature. The reaction mixture was diluted with 10% $Na_2S_2O_3$. The organic layer was separated, washed with brine, dried over $MgSO_4$, filtered and concentrated to obtain the crude as brown oil. The crude was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to obtain epoxy alcohol **102** (3.0 g, 87%) as colorless oil which solidified on standing. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.93-3.87 (m, 1H), 3.12-3.09 (m, 1H), 3.07-3.04 (m, 1H), 2.29 (s, 1H), 1.56-1.40 (m, 4H), 1.30-1.25 (m, 25H), 0.87 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 71.0, 55.5, 52.1, 37.6, 34.6, 32.1, 29.85, 29.8, 29.5, 25.7, 22.8, 14.3, 13.5; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{37}O_2$ ($M+H^+$) 285.2788 found 285.2785; **FT-IR**: 3415, 2994, 2923, 2854, 1463 cm^{-1} ; **Mp**: 32-33 °C; **$[\alpha]_D^{25}$** = - 3.2 (c=1.05, $CHCl_3$).

Synthesis of azidodiol 103



Trimethylsilylazide (0.4 g, 3.47 mmol) was added to a solution of titanium isopropoxide (0.49 g, 1.72 mmol) in benzene (12.5 mL) and the solution heated to 80 ^{o}C for 5 h. A solution of epoxyalcohol **102** (0.5 g, 1.75 mmol) in benzene (5 mL) was then added and the reaction stirred for 15 min before cooling to room temperature. Benzene was removed under vacuum and the crude diluted with ether. A solution of 5% H_2SO_4 was then added and the resulting solution was stirred for 1 h at room temperature. Separated the organic layer and extracted the aqueous with ether. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain brown oil. Purified the crude by flash chromatography using 25% ethyl acetate in hexanes as eluent to obtain the azidodiol **103** (0.3 g, 53%) as colorless oil which solidified on standing. Obtained also the C3 regioisomer **F** (15%). **Compound 103**: **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.88-3.84 (m, 1H), 3.75-3.72 (m, 1H), 3.58 (bs, 1H), 3.43-3.39 (m, 1H), 2.89 (bs, 1H), 1.60-1.55 (m, 2H), 1.54-1.44 (m, 2H), 1.43-1.38 (m, 1H), 1.31-1.25 (m, 24H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 75.6, 72.7, 62.1, 39.2, 38.4, 32.1, 29.85, 29.8, 29.5, 25.5, 22.8, 15.3, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{38}N_3O_2$ ($M+H^+$) 328.2958 found 328.2952; **FT-IR**: 3390, 2923, 2854, 2109, 1463, 1261, 1066 cm^{-1} ; **Mp**: 42-44 ^{o}C; **$[\alpha]_D^{25}$** = + 31.0 (c=1.07, $CHCl_3$).

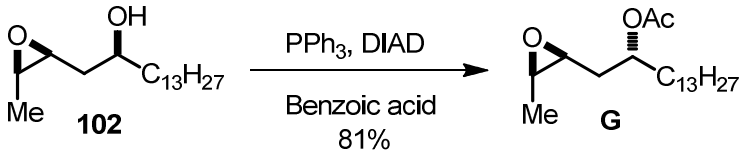
Synthesis of aminodiol enigmol 31



Lithium aluminum hydride (2.0 M in THF) (0.23 g, 6.0 mmol) was added to THF (3.2 mL) at room temperature. A solution of azidodiol **103** (0.1 g, 0.3 mmol) in THF (0.75 mL) was then added dropwise and the reaction mixture stirred at room temperature for 15 min. The reaction was quenched by dropwise addition of Rochelle's salt. The biphasic solution was stirred for 30 min at room temperature, before separating the organic layer. The aqueous layer was extracted with ethyl acetate and the combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain aminodiol enigmol **31** as a white solid (0.065 g, 71%). For spectral details reference 31.

Synthesis of isoenigmol 32

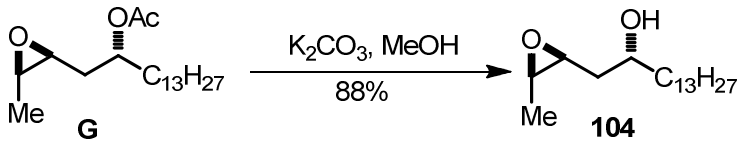
Synthesis of epoxyacetate G



Dissolved epoxy alcohol **102** (3.0 g, 10.5 mmol) in ether (190 mL). Triphenyl phosphine (5.5 g, 21.0 mmol) and acetic acid (1.26 g, 21.0 mmol) were then added and the reaction cooled to 0 ^oC. Diisopropyl azodicarboxylate (4.26 g, 21.0 mmol) was then added and the reaction mixture stirred at 0 ^oC for 2 h. The reaction was diluted with water and the aqueous layer extracted with ether. The

organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as yellow oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain epoxy acetate **G** (2.8 g, 81%) as colorless oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.06-5.02 (m, 1H), 3.06-3.02 (m, 1H), 2.97-2.94 (m, 1H), 2.05 (s, 3H), 1.78-1.76 (m, 2H), 1.64-1.59 (m, 2H), 1.29- 1.25 (m, 26H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.8, 72.4, 54.1, 52.5, 34.5, 32.7, 32.1, 29.8 (2C), 29.7 (2C), 29.6 29.5, 25.4, 22.8, 21.4, 14.3, 13.5; **HRMS (ESI)**: *m/z* calcd. for $C_{20}H_{39}O_3$ ($M+H^+$) 327.2893 found 327.2886; **FT-IR**: 2994, 2925, 2854, 1739 1465, 1373, 1240, 1022, 829 cm^{-1} ; **$[\alpha]_D^{25}$** = + 7.2 (c=1.04, $CHCl_3$).

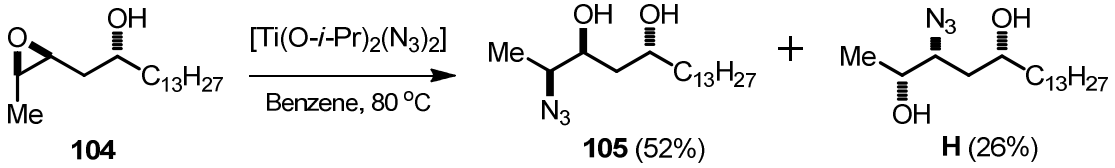
Synthesis of epoxy alcohol 104



Compound **G** (0.3 g, 0.91 mmol) was dissolved in methanol (3 mL). Pottasium carbonate (0.063 g, 0.45 mmol) was then added and the reaction mixture stirred at room temperature for 4 h. Methanol was removed under vacuum and the crude dissolved in water. The aqueous layer was extracted with CH_2Cl_2, dried over $MgSO_4$, filtered and concentrated to obtain the crude as white solid. Purified the crude by flash chromatography using 20% ethyl acetate in hexanes as eluent to obtain epoxy alcohol **104** (0.23 g, 88%) as white solid. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.88-3.83 (m, 1H), 3.15-3.09 (m, 1H), 3.07-3.04 (m, 1H), 1.74-1.70 (m, 2H), 1.62-1.58 (ddd, *J* = 3.6, 7.8, 14.4 Hz, 1H), 1.55-1.51 (m, 2H), 1.48-1.44 (m, 1H), 1.29 (d, *J* = 6.6 Hz, 3H), 1.25 (bs, 22H), 0.87 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR**

(150 MHz, $CDCl_3$) δ 70.3, 54.6, 52.9, 37.9, 35.0, 32.1, 29.8 (2C), 29.5, 25.8, 22.9, 14.3, 13.6: **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{37}O_2$ ($M+H^+$) 285.2788 found 285.2785; **FT-IR**: 3334, 3257, 2915, 2848, 1467, 721 cm^{-1} ; **Mp**: 48-49 °C; **$[\alpha]_D^{25}$** = -13.2 (c=1.0, $CHCl_3$).

Synthesis of azidodiol 105

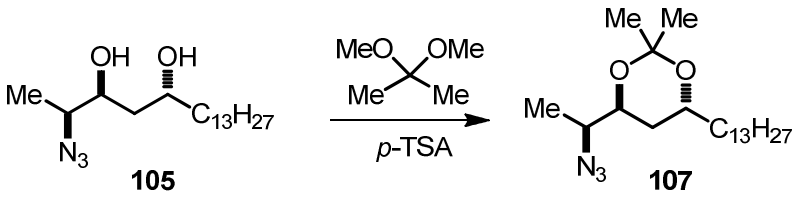


104 **105** (52%) **H** (26%)

Trimethylsilylazide (0.16 g, 1.38 mmol) was added to a solution of titanium tetraisopropoxide (0.19 g, 0.66 mmol) in benzene (5 mL) and the solution heated to 80 °C for 5 h. A solution of epoxyalcohol **104** (0.2 g, 0.7 mmol) in benzene (2 mL) was then added and the reaction stirred for 15 min before cooling to room temperature. Benzene was removed under vacuum and the crude was diluted with ether. A solution of 5% H_2SO_4 was then added and the solution stirred for 1 h at room temperature. Separated the organic layer and extracted the aqueous with ether. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as brown oil. Purified the crude by flash chromatography using 20% ethyl acetate in hexanes as eluent to obtain azidodiol **105** as a white solid (0.12 g, 52%) and 0.06 g of the C3 isomer **H** (26%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.97-3.91 (m, 1H), 3.81-3.76 (m, 1H), 3.49-3.45 (m, 1H), 2.63 (d, *J* = 4.2 Hz, 1H), 2.08 (s, 1H), 1.70 (ddd, *J* = 3.0, 9.6, 14.2 Hz, 1H), 1.60-1.51 (m, 2H), 1.49-1.40 (m, 2H), 1.32-1.26 (m, 24H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 72.2, 69.2, 62.6, 39.5, 37.6, 32.1, 29.8 (2C), 29.5, 25.9, 22.9, 15.8, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{38}N_3O_2$ ($M+H^+$) 328.2958

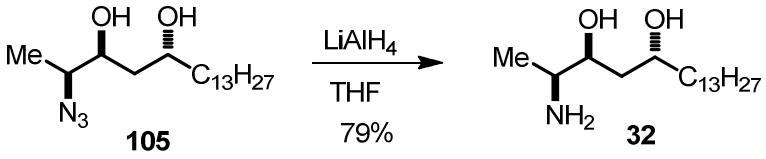
found 328.2952; **FT-IR**: 3305, 2954, 2913, 2848, 2123, 1469, 1022, 981 cm^{-1} ; **Mp**: 56-58°C; $[\alpha]_D^{25}$ = + 17.2 (c=0.52, $CHCl_3$).

Synthesis of azidoacetonide 107



^{1}H NMR (600 MHz, $CDCl_3$) δ 3.80-3.74 (m, 2H), 3.39-3.34 (m, 1H), 1.73 (ddd, *J* = 6.0, 9.9, 12.6 Hz, 1H), 1.54-1.49 (m, 2H), 1.38 (s, 3H), 1.37 (s, 3H), 1.30-1.26 (m, 23H), 1.18 (d, *J* = 7.2 Hz, 3H), 0.89 (t, *J* = 6.0 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 100.9, 70.8, 66.9, 60.2, 36.06, 36.04, 32.1, 29.87, 29.81, 29.7, 29.5, 25.5, 24.6, 24.5, 22.9, 15.3, 14.3.

Synthesis of aminodiol isoenigmol 32

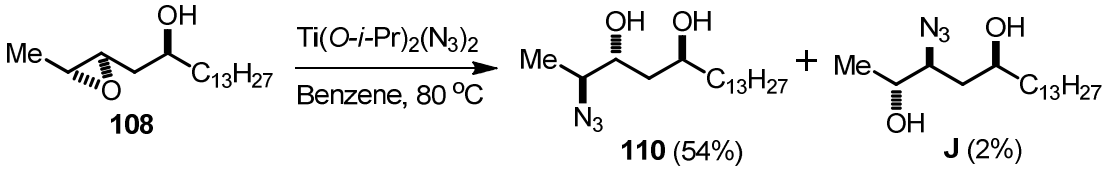


Lithium aluminum hydride (2.0 M in THF, 0.011 g, 0.28 mmol) was added to THF (1.6 mL) at room temperature. A solution of azidodiol **105** (0.05 g, 0.15 mmol) in THF (0.4 mL) was then added dropwise and the reaction mixture stirred at room temperature for 15 min. The reaction was quenched by dropwise addition of saturated Rochelle's salt. The biphasic solution was stirred for 30 min at room temperature and the aqueous layer extracted with ethyl acetate. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain isoenigmol **32** as a white solid (0.036 g, 79%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.92-

3.87 (m, 1H), 3.52-3.49 (m, 1H), 2.85-2.81 (m, 1H), 2.78-2.20 (bs, 3H), 1.66-1.50 (m, 3H), 1.47-1.39 (m, 2H), 1.36-1.25 (bs, 22H), 1.11 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 73.7, 69.2, 51.2, 39.6, 37.8, 32.1, 29.8, 29.5, 26.0, 22.9, 20.8, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{40}NO_2$ ($M+H^+$) 302.3053 found 302.3047; **FT-IR**: 3359, 3307, 2917, 2850, 1592, 1467, 1064 cm^{-1}; **Mp**: 64-66 °C; **$[\alpha]_D^{25}$** = -2.7 (c=0.87, $CHCl_3$).

Synthesis of enigmol diastereomers 33 and 34

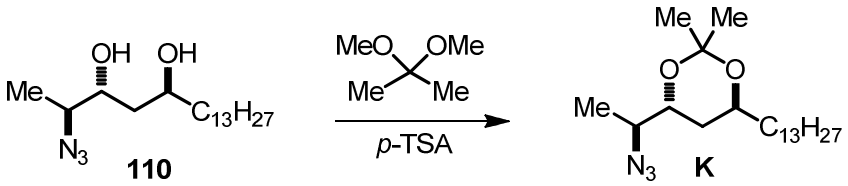
Synthesis of azidodiol 110



Trimethylsilylazide (0.16 g, 1.38 mmol) was added to a solution of titanium tetraisopropoxide (0.19 g, 0.66 mmol) in benzene (5 mL) and the solution heated to 80 °C for 5 h. A solution of epoxyalcohol **108**[31] (0.2 g, 0.7 mmol) in benzene (2 mL) was then added and the reaction stirred for 15 min before cooling to room temperature. Benzene was removed under vacuum and the crude was diluted with ether. A solution of 5% H_2SO_4 was then added and the solution stirred for 1 h at room temperature. Separated the organic layer and extracted the aqueous with ether. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as brown oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain azidodiol **110** as a white solid (0.125 g, 54%) along with C3 isomer **J** (0.01 g, 2%) **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.98-3.94 (m, 1H), 3.90-3.87 (m, 1H), 3.59-3.55 (m, 1H), 2.85 (bs, 1H), 1.69 (ddd, *J* = 3.0, 9.6, 14.1 Hz, 1H), 1.58-1.46 (m, 3H), 1.44-1.39

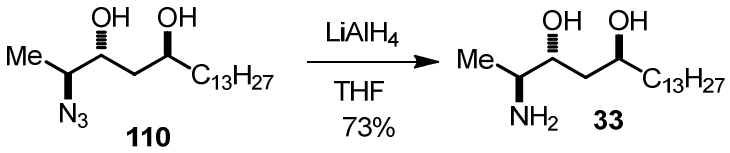
(m, 1H), 1.29 (d, *J* = 6.6 Hz, 3H), 1.26 (bs, 21H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 71.5, 69.6, 61.9 38.1, 37.6, 32.1, 29.8, 29.7, 29.5, 25.9, 22.9, 14.5, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{38}N_3O_2$ ($M+H^+$) 328.2958 found 328.2953; **FT-IR**: 3297, 2954, 2913, 2848, 2129, 1469, 1259, 1022, cm^{-1}; **Mp**: 57-59 °C; **$[\alpha]_D^{25}$** = + 29.8 (c=1.04, $CHCl_3$).

Synthesis of azidoacetonide K



^{1}H NMR (600 MHz, $CDCl_3$) δ 3.79-3.75 (m, 2H), 3.56-3.51 (m, 1H), 1.81 (ddd, *J* = 5.4, 12.6 Hz, 1H), 1.57-1.50 (m, 2H), 1.46-1.39 (m, 2H), 1.35 (s, 6H), 1.30-1.26 (m, 21H), 1.20 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 100.7, 69.9, 66.9, 60.4, 36.0, 34.2, 32.1, 29.87, 29.81, 29.7, 29.5, 25.5, 24.8, 24.7, 22.9, 15.3, 14.3.

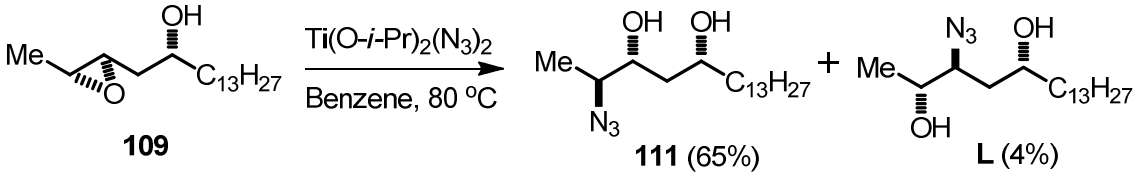
Synthesis of aminodiol diastereomers 33



Lithium aluminum hydride (2.0 M in THF) (0.017 g, 0.44 mmol) was added to THF (2.4 mL) at room temperature. A solution of azidodiol **110** (0.075 g, 0.22 mmol) in THF (0.6 mL) was then added dropwise and the reaction mixture stirred at room temperature for 15 min. The reaction was quenched by dropwise addition of saturated Rochelle's salt. The biphasic solution was stirred for 30 min at room temperature, and the aqueous layer extracted with ethyl acetate. The

combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain aminodiol diastereomer **33** as a white solid (0.05 g, 73%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.94-3.88 (m, 1H), 3.78-3.74 (m, 1H), 2.96-2.92 (m, 1H), 1.65-1.60 (m, 1H), 1.55-1.41 (m, 4H), 1.32-1.25 (bs, 22H), 1.06 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 71.8, 69.2, 51.1, 39.0, 37.8, 32.1, 29.9, 29.8 29.5, 26.1, 22.9, 18.3, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{40}NO_2$ ($M+H^+$) 302.3053 found 302.3051; **FT-IR**: 3307, 2915, 2848, 1469, 1051 cm^{-1} ; **Mp**: 71-73°C; **$[\alpha]_D^{25}$** = +13.9 (c=0.55, $CHCl_3$).

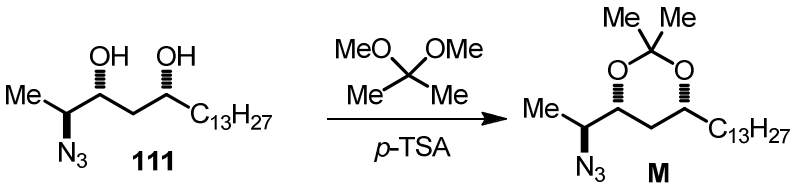
Synthesis of azidodiol 111



Trimethylsilylazide (0.16 g, 1.38 mmol) was added to a solution of titanium isopropoxide (0.19 g, 0.66 mmol) in benzene (5 mL) and the solution heated to 80 °C for 5 h. A solution of epoxyalcohol **109**[31] (0.2 g, 0.7 mmol) in benzene (2 mL) was then added and the reaction stirred for 15 min before cooling to room temperature. Benzene was removed under vacuo and the crude diluted with ether. A solution of 5% H_2SO_4 was then added and the solution stirred for 1h at room temperature. Separated the organic layer and extracted the aqueous with ether. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the crude as brown oil. Purified the crude by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain azidodiol **111** as a white solid (0.15 g, 65%) along with C3 isomer **L** (0.01g, 4%). **^{1}H NMR**

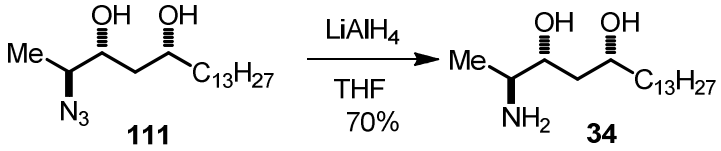
(600 MHz, $CDCl_3$) δ 3.89-3.85 (m, 1H), 3.83-3.80 (m, 1H), 3.52-3.48 (m, 1H), 2.57 (bs, 1H), 1.68-1.66 (m, 1H), 1.53-1.47 (m, 3H), 1.41-1.37 (m, 1H), 1.30-1.26 (m, 21H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 75.5, 73.3, 61.9 38.5, 38.3, 32.1, 29.8, 29.7, 29.5, 25.9, 22.8, 14.3 (2C); **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{38}N_3O_2$ ($M+H^+$) 328.2958 found 328.2959; **FT-IR**: 3369, 2923, 2854, 2115, 2098, 1461, 1261, 1051, cm^{-1} ; **Mp**: 31-33 °C; **$[\alpha]_D^{25}$** = + 10.8 (c=0.86, $CHCl_3$).

Synthesis of azidoacetonide M



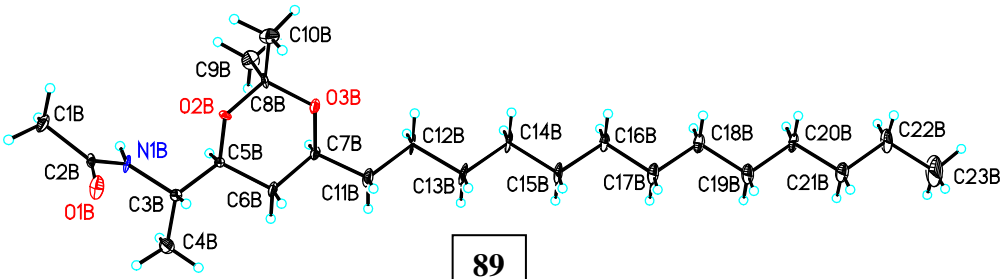
^{1}H NMR (600 MHz, $CDCl_3$) δ 3.80-3.75 (m, 2H), 3.45-3.41 (m, 1H), 1.57-1.53 (m, 1H), 1.42 (s, 3H), 1.40 (s, 3H), 1.31-1.26 (m, 22H), 1.22 (d, *J* = 6.6 Hz, 3H), 0.89 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 98.8, 72.4, 68.9, 60.8, 36.6, 32.4, 32.1, 30.2, 29.8, 29.7, 29.5, 25.5, 22.9, 19.9, 15.1, 14.3.

Synthesis of aminodiol, diastereomer 34



Lithium aluminum hydride (2.0 M in THF) (0.017 g, 0.44 mmol) was added to THF (2.4 mL) at room temperature. A solution of azidodiol **111** (0.075 g, 0.22 mmol) in THF (0.6 mL) was then added dropwise and the reaction mixture stirred at room temperature for 15 min. The reaction was quenched by dropwise addition of saturated Rochelle's salt and stirred the biphasic solution for 30 min at

room temperature. Separated the organic layer and extracted the aqueous layer with ethyl acetate. The combined organic layer was dried over $MgSO_4$, filtered and concentrated to obtain aminodiol **34** as a white solid (0.05 g, 70%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 3.99-3.83 (m, 1H), 3.74-3.70 (m, 1H), 3.03-2.99 (m, 1H), 2.90-2.00 (bs, 3H), 1.58-1.40 (m, 5H), 1.30-1.26 (bs, 21H), 1.57 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 75.7, 72.0, 50.7, 38.3, 38.0, 32.1, 29.8, 29.5, 25.7, 22.9, 18.0, 14.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{40}NO_2$ ($M+H^+$) 302.3053 found 302.3050; **FT-IR**: 3340, 2917, 2848, 1461, 1076 cm^{-1} ; **Mp**: 82-83 °C; **$[\alpha]_D^{25}$** = +3.1 (c=0.98, $CHCl_3$).



89

Table 3. Crystal data and structure refinement for **89**.

Identification code	CLP2_282s
Empirical formula	C_{23} H_{45} N O_3
Formula weight	383.60
Temperature	173(2) K
Wavelength	1.54178 Å
Crystal system	Monoclinic
Space group	P2(1)
Unit cell dimensions	a = 9.2708(10) Å α = 90°.

	b = 8.7296(8) Å β = 90.665(7)°.
	c = 30.257(3) Å γ = 90°.
Volume	2448.6(4) $Å^3$
Z	4
Density (calculated)	1.041 Mg/m^3
Absorption coefficient	0.520 mm^{-1}
F(000)	856
Crystal size	0.24 x 0.10 x 0.07 mm^3
Theta range for data collection	1.46 to 66.22°.
Index ranges	-10<=h<=10, -5<=k<=9, -35<=l<=32
Reflections collected	10761
Independent reflections	5282 [R(int) = 0.0187]
Completeness to theta = 66.22°	88.9 %
Absorption correction	Semi-empirical from equivalents
Max. and min. transmission	0.9645 and 0.8854
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	5282 / 1 / 487
Goodness-of-fit on F^2	1.040
Final R indices [I>2 sigma(I)]	R1 = 0.1023, wR2 = 0.2477
R indices (all data)	R1 = 0.1144, wR2 = 0.2653
Absolute structure parameter	-0.3(4)
Largest diff. peak and hole	0.504 and -0.611 $e.Å^{-3}$

Table 4. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3) for **89**.

U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
C(1)	4003(6)	5214(8)	-161(2)	26(1)
C(2)	3988(5)	4448(6)	281(2)	14(1)
C(3)	2473(5)	3469(6)	895(2)	8(1)
C(4)	2976(6)	1814(6)	875(2)	18(1)
C(5)	3178(5)	4364(6)	1277(2)	9(1)
C(6)	2975(5)	3602(6)	1724(2)	14(1)
C(7)	3600(5)	4655(6)	2076(2)	12(1)
C(8)	3064(5)	6846(6)	1619(2)	11(1)
C(9)	4624(6)	7344(8)	1523(2)	24(1)
C(10)	2062(6)	8196(7)	1629(2)	22(1)
C(11)	3392(6)	4052(7)	2541(2)	20(1)
C(12)	4128(7)	5027(7)	2896(2)	25(1)
C(13)	4144(7)	4250(8)	3344(2)	28(1)
C(14)	4923(7)	5152(8)	3707(2)	29(1)
C(15)	5028(7)	4287(8)	4142(2)	29(1)
C(16)	5827(7)	5171(8)	4503(2)	29(2)
C(17)	5953(6)	4286(8)	4933(2)	26(1)
C(18)	6754(7)	5170(9)	5294(2)	29(2)

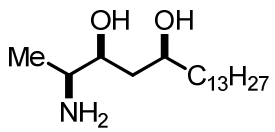
C(19)	6911(7)	4296(8)	5728(2)	30(2)
C(20)	7736(7)	5215(9)	6078(2)	32(2)
C(21)	7884(8)	4392(10)	6521(2)	40(2)
C(22)	8704(8)	5347(10)	6862(2)	45(2)
C(23)	8787(12)	4609(16)	7302(2)	94(5)
C(1B)	1688(5)	8320(7)	10315(2)	18(1)
C(2B)	1071(5)	9339(6)	9965(1)	9(1)
C(3B)	1426(5)	10539(6)	9233(2)	9(1)
C(4B)	2015(6)	12159(6)	9272(2)	21(1)
C(5B)	1916(4)	9727(6)	8817(1)	6(1)
C(6B)	1422(5)	10529(6)	8392(2)	12(1)
C(7B)	1701(5)	9550(6)	7987(2)	10(1)
C(8B)	1589(5)	7302(6)	8447(1)	9(1)
C(9B)	3196(5)	6885(7)	8432(2)	22(1)
C(10B)	657(6)	5888(7)	8490(2)	24(1)
C(11B)	982(6)	10248(7)	7578(2)	21(1)
C(12B)	1135(6)	9329(8)	7152(2)	21(1)
C(13B)	415(7)	10181(8)	6760(2)	26(1)
C(14B)	432(7)	9288(8)	6329(2)	26(1)
C(15B)	-324(7)	10168(8)	5949(2)	27(1)
C(16B)	-430(7)	9278(8)	5519(2)	26(1)
C(17B)	-1204(7)	10176(8)	5150(2)	28(1)
C(18B)	-1366(7)	9281(8)	4726(2)	26(1)

C(19B)	-2131(7)	10170(8)	4353(2)	26(1)
C(20B)	-2315(7)	9235(8)	3932(2)	27(1)
C(21B)	-3049(7)	10102(8)	3552(2)	29(1)
C(22B)	-3229(9)	9160(9)	3133(2)	45(2)
C(23B)	-3894(12)	10055(13)	2750(2)	80(3)
N(1)	2716(4)	4242(5)	472(1)	11(1)
N(1B)	1890(4)	9633(5)	9614(1)	11(1)
O(1)	5152(4)	4001(6)	467(1)	27(1)
O(2)	2524(4)	5843(4)	1283(1)	10(1)
O(3)	2954(4)	6146(4)	2043(1)	12(1)
O(1B)	-197(4)	9876(5)	9999(1)	18(1)
O(2B)	1270(3)	8229(4)	8820(1)	8(1)
O(3B)	1116(4)	8052(4)	8047(1)	13(1)

Compilation of spectroscopic data for enigmol diastereomers:

The diastereomer were synthesized by various methods by McDonald group (John Wiseman and Claney Pereira), Liotta group (Mark Baillie, Anatoliy Bushnev, Jason Holt and David Menaldino) and Liebeskind group (Ethel Garnier). This is just a data compilation of these diastereomers from the above sources.

Enigmol (31):



(2*S*,3*S*,5*S*)-2-aminooctadecane-3,5-diol **(31)**

Since the aminoalcohol compounds are polar and are capable of hydrogen bonding the chemical shifts in the spectral are affected by the concentration (very difficult to match up spectra between individuals). Mixed ^{1}H and ^{13}C was carried out to solve these problems.

John Wiseman[31]

^{1}H NMR ($CDCl_3$) δ 3.85 (m, *J* = 4.2, 2.4, 1.2 Hz, 1H), 3.48 (ddd, *J* = 9.9, 6.6, 2.4 Hz, 1H), 3.28 (bs, 4H), 2.78 (m, *J* = 6.6 Hz, 1H), 1.94 (s, 0.2 H, maybe trace impurity or a result of H-bonding), 1.49 (bm, 1H), 1.42 (m, 3H), 1.25 (bs, 22H), 1.12 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** ($CDCl_3$) δ 76.4, 72.1, 51.8, 40.4, 38.2, 32.1, 29.91, 29.87, 29.83, 29.54, 25.7, 22.9, 20.6, 14.3; **HRMS (EI)** *m/z* calcd for $C_{18}H_{39}O_2NLi$, 308.3141, found 308.3127; **Anal. Calcd. for $C_{18}H_{39}O_2N$**: C, 71.70; H, 13.04; N, 4.65; found C, 70.35; H, 12.86; N, 4.48. **IR** (thin film, CH_2Cl_2): 3401, 2952, 2918, 2850, 1645, 1467, 1018 cm^{-1}; **Mp**: 69-70.5 ^{o}C; **$[\alpha]_D^{25}$** = -16.8 (CH_2Cl_2).

John Wiseman sample (taken by Claney)

^{1}H NMR (400 MHz, CD_3OD) δ 3.82-3.76 (m, 1H), 3.49 (dd, *J* = 3.2, 5.4, 9.0 Hz, 1H), 2.79 (dq, *J* = 6.8 Hz, 1H), 1.66 (dt, *J* = 4.0, 14.4 Hz, 1H), 1.54-1.39 (m, 5H), 1.29 (bs, 23H), 1.09 (d, *J* = 6.4 Hz, 3H), 0.90 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (100 MHz, CD_3OD) δ 75.8, 71.5, 52.6, 41.2, 38.6, 33.2, 31.0, 30.9, 30.6, 26.6, 23.9, 19.1, 14.6; **$[\alpha]_D^{25}$** = -10.3 (c=1.05, CH_3OH).

John Wiseman sample (taken by Claney)

^{13}C NMR (100 MHz, DMSO-*d6*) δ 73.3, 68.3, 50.2, 37.1, 31.3, 29.3, 29.1, 28.7, 25.0, 22.1, 19.5, 13.9.

Avanti sample (taken by Claney)

^{1}H NMR (400MHz, CD_3OD) δ 3.32-3.76 (m, 1H), 3.49 (dd, *J* = 3.2, 5.4, 9.0 Hz, 1H), 2.78 (dq, *J* = 6.4 Hz, 1H), 1.66 (dt, *J* = 4.0, 14.0 Hz, 1H), 1.54-1.39 (m, 5H), 1.29 (bs, 23H), 1.09 (d, *J* = 6.4 Hz, 3H), 0.90 (t, *J* = 6.8 Hz, 3H); **^{13}C NMR** (100MHz, CD_3OD) δ 75.8, 71.5, 52.6, 41.2, 38.6, 33.2, 31.0, 30.9, 30.6, 26.6, 23.9, 19.1, 14.6.

Avanti sample (taken by Claney)

^{1}H NMR (400MHz, $CDCl_3$) δ 3.91-3.85 (m, 1H), 3.45 (ddd, *J* = 2.4, 5.6, 10.2 Hz, 1H), 2.74 (dq, *J* = 6.4 Hz, 1H), 1.64 (dt, *J* = 2.4, 14.0 Hz, 1H), 1.45-1.37 (m, 3H), 1.25 (bs, 20H), 1.12 (d, *J* = 6.4 Hz, 3H), 0.88 (t, *J* = 6.8 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 76.7, 72.1, 51.7, 40.5, 38.2, 32.1, 29.8, 29.5, 25.7, 22.9, 21.2, 14.3.

From new approach (Claney)

^{1}H NMR (400 MHz, $CDCl_3$) δ 3.87 (m, 1H), 3.45 (t, *J* = 7.2 Hz, 1H), 2.76-2.74 (bm, 1H), 1.64 (d, *J* = 13.8 Hz, 1H), 1.53-1.48 (m, 1H), 1.44-1.39 (m, 3H), 1.25 (bs, 22H), 1.12 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 76.6, 72.1, 51.7, 40.5, 38.2, 32.1, 29.8, 29.5, 25.7, 22.9, 21.1, 14.3; **$[\alpha]_D^{25}$** = -12.1 (c=1.05, CH_3OH).

Anatoliy Bushnev sample (compd-61, taken by Claney)

^{1}H NMR ($CDCl_3$) δ 3.87 (bs, 1H), 3.61 (bs, 1H), 2.94 (bs, 1H), 1.67 (d, *J* = 13.2 Hz, 1H), 1.54-1.48 (m, 1H), 1.46-1.38 (m, 2H), 1.26-1.22 (bs, 24H), 0.88 (t, *J* = 6.6 Hz, 3H).

Mixed ^{1}H and ^{13}C of John Wiseman and Anatoliy sample (taken by Claney)

^{1}H NMR (400 MHz, CD_3OD) δ 3.32-3.76 (m, 1H), 3.49 (dd, *J* = 3.2, 5.4, 9.0 Hz, 1H), 2.78 (dq, *J* = 6.4 Hz, 1H), 1.66 (dt, *J* = 4.0, 14.0 Hz, 1H), 1.54-1.39 (m, 5H), 1.29 (bs, 23H), 1.09 (d, *J* = 6.4 Hz, 3H), 0.90 (t, *J* = 6.8 Hz, 3H); **^{13}C NMR** (100 MHz, CD_3OD) δ 75.8, 71.5, 52.6, 41.2, 38.6, 33.2, 31.0, 30.9, 30.6, 26.6, 23.9, 19.1, 14.6.

Mark Baillie, Anatoliy and Jason Holt

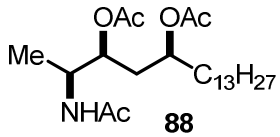
^{1}H NMR (400 MHz, CD_3OD) δ 3.86 (m, 1 H), 3.46 (m, 1 H), 2.75 (quintet, *J* = 6.1 Hz, 1 H), 1.64 (dt, *J* = 14.3, 2.4 Hz, 1 H), 1.55 - 1.35 (m, 5 H), 1.35 - 1.20 (m, 22 H), 1.08 (d, *J* = 6.7 Hz, 2 H), 0.88 (t, *J* = 7.0 Hz, 3 H); **^{13}C NMR** (100 MHz, CD_3OD) δ 72.2 (2 C), 51.8, 40.5, 38.2, 32.1, 29.9 (7 C), 29.7, 25.7, 22.9, 21.1, 14.3; **HRMS (ESI)** *m/z* calcd. for $C_{18}H_{39}NO_2$ ($M+H^+$) 302.30550 found 302.3059; **Anal. Calcd. for** $C_{18}H_{39}NO_2$: C, 71.70; H, 13.01; N, 4.65. Found: C 71.76; H 13.26; N 4.64. **IR**: 3402, 3328, 2917, 2847, 1596, 1463, 1325, 1170, 1142, 1120, 1075, 1020, 984, 919, 845, 818, 721, 607 cm^{-1}; **MP** = 64-65 ^{o}C; **$[\alpha]_D^{24}$** = +3.3 (c = 1.6, $CHCl_3$), -10.6 (c = 1.6, MeOH).

David Menaldino (compound 55b from thesis)

^{1}H NMR (500 MHz, $CDCl_3$) δ 3.87 (m, 1H), 3.53 (m, 1H), 2.85 (qn, J = 6.4 Hz, 1H), 1.65 (dt, *J* = 14.3, 2.2 Hz, 1H), 1.46 (m, 4H), 1.25 (br s, 20H), 1.17 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.0 Hz, 3H); **^{13}C NMR** (75.5 MHz, DMSO-d_6, APT) δ 72.70, 71.02, 53.10, 38.14, 31.92, 29.75, 29.55, 29.37, 25.65, 25.59, 22.68, 15.92, 14.09; **MS (low resolution FAB)**: *m/z* calcd for $C_{18}H_{40}NO_2$ ($M+H^+$)

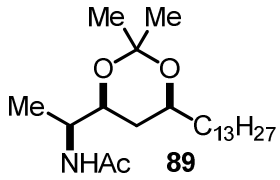
302.3059, found 302.3067; **IR ($CDCl_3$ solution)**: 3650, 3500, 3300, 2920, 2850, 1510, 1490, 1470, 1450, 1380, 1100 cm^{-1}; **Mp**: 104-107.5 ^oC.

Enigmol derivatives: Prepared from John Wiseman sample (by Claney)



88

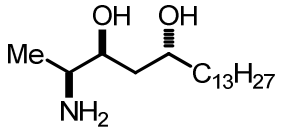
^{1}H NMR (400 MHz, $CDCl_3$) δ 5.51 (d, *J* = 9.6 Hz, 1H), 4.94-4.86 (m, 2H), 4.32-4.24 (m, 1H), 2.10 (s, 3H), 2.04 (s, 3H), 1.99 (s, 3H), 1.85-1.70 (m, 2H), 1.53 (bs, 2H), 1.24 (bs, 26H), 1.09 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 6.4 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 171.0, 170.7, 169.6, 73.6, 71.4, 47.3, 36.6, 34.1, 32.1, 29.8, 29.7 (2C), 29.6, 29.5, 25.3, 23.6, 22.9, 21.4, 21.1, 18.6, 14.3.



89

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.76 (d, *J* = 8.8 Hz, 1H), 3.97 (dq, *J* = 2.0, 7.2 Hz, 1H), 3.83-3.77 (m, 2H), 2.00 (s, 3H), 1.42 (s, 3H), 1.39 (s, 3H), 1.40-1.36 (m, 2H), 1.25 (bs, 23H), 1.16 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 6.4 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 169.9, 98.5, 71.5, 68.9, 48.1, 36.6, 33.8, 32.1, 30.2, 29.8 (2C), 29.7, 29.6, 29.5, 25.1, 2 23.7, 22.9, 19.9, 18.4, 14.3.

Isoenigmol:



(2*S*,3*S*,5*R*)-2-aminooctadecane-3,5-diol **(32)**

From new approach (Claney)

^{1}H NMR (600 MHz, $CDCl_3$) δ 3.92-3.87 (m, 1H), 3.52-3.49 (m, 1H), 2.85-2.81 (m, 1H), 2.78-2.20 (bs, 3H), 1.66-1.50 (m, 3H), 1.47-1.39 (m, 2H), 1.36-1.25 (bs, 22H), 1.11 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 73.7, 69.2, 51.2, 39.6, 37.8, 32.1, 29.8, 29.5, 26.0, 22.9, 20.8, 14.3; **HRMS (ESI)**: *m/z* calcd. For $C_{18}H_{40}NO_2$ ($M+H^+$) 302.3053 found 302.3047; **FT-IR**: 3359, 3307, 2917, 2850, 1592, 1467, 1064 cm^{-1}; **Mp**: 64-66°C; **$[\alpha]_D^{25}$** = -2.7 (c=0.87, $CHCl_3$).

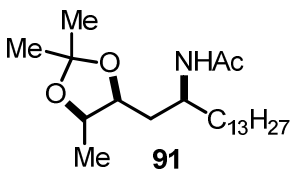
Mark Baillie, Anatoliy Bushnev and Jason Holt (Data same as David Menaldino)

^{1}H NMR (500 MHz, CD_3OD, 60 ˚C) δ 3.80 (m, 1 H), 3.72 (dt, *J* = 6.8, 1.6 Hz, 1 H), 3.10 (dq, *J* = 6.8, 6.8 Hz, 1 H), 1.54 (m, 2 H), 1.45 (m, 2 H), 1.28 (m, 22 H), 1.26 (d, *J* = 5.0 Hz, 3 H), 0.89 (t, *J* = 6.9 Hz, 3 H); **^{13}C NMR** (75.5 MHz, d_6-DMSO) δ 72.3, 67.7, 51.4, 40.5, 37.9, 31.6, 29.4 (6 C), 29.0, 25.4, 22.3, 17.5, 13.0; **HRMS (FAB)**: *m/z* calcd. for $C_{18}H_{39}NO_2$ ($M+H^+$) 302.3074, found 302.30590); **$[\alpha]^{24}_D$** = -12.9 (c = 2.0, MeOH), -4.83 (c = 2.9, $CHCl_3$).

Anatoly sample (compd-65, taken by Claney)

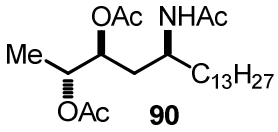
^{1}H NMR (600 MHz, $CDCl_3$) δ 3.88 (bs, 1H), 3.84-3.66 (bs, 2H), 3.65-3.63 (m, 3H), 2.98 (dq, *J* = 7.2 Hz, 1H), 1.66-1.61 (m, 1H), 1.58-1.50 (m, 2H), 1.46-1.42 (m, 2H), 1.26 (bs, 23H), 1.18 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 6.6 Hz, 3H).

John Wiseman sample derivatization (by Claney):



91

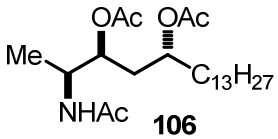
^{1}H NMR (600 MHz, $CDCl_3$) δ 5.43 (d, *J* = 8.4 Hz, 1H), 4.26 (dq, *J* = 6.3 Hz, 1H), 4.09 (ddd, *J* = 4.8, 8.4 Hz, 1H), 4.02-3.96 (m, 1H), 1.97 (s, 3H), 1.69 (ddd, *J* = 4.8, 14.4 Hz, 1H), 1.57-1.50 (m, 3H), 1.45 (s, 3H), 1.31 (s, 3H), 1.25 (bs, 27H), 1.15 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.0, 107.7, 75.9, 74.1, 48.1, 35.7, 35.1, 32.1, 30.2, 29.8, 29.7, 29.5, 28.7, 26.0, 25.9, 23.8, 22.9, 15.9, 14.3.



90

^{1}H NMR (600 MHz, $CDCl_3$) δ 5.52 (d, *J* = 8.4 Hz, 1H), 5.02-4.99 (m, 1H), 4.98-4.96 (m, 1H), 3.99-3.93 (m, 1H), 2.08 (s, 3H), 2.03 (s, 3H), 1.99 (s, 3H), 1.76 (ddd, *J* = 5.4, 14.4 Hz, 1H), 1.69 (ddd, *J* = 7.2, 15.0 Hz, 1H), 1.52-1.46 (m, 1H), 1.24 (bs, 26H), 1.21 (d, *J* = 6.6 Hz, 3H), 0.88 (t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 171.0, 170.6, 169.8, 72.0, 71.2, 47.0, 39.3, 34.9, 34.7, 32.1, 29.8 (3C), 29.7, 29.6, 29.5, 26.0, 23.7, 22.8, 21.3, 14.8, 14.3.

Isoenigmol derivatization (from new approach):



106

^{1}H NMR (400 MHz, $CDCl_3$) δ 5.57 (d, *J* = 9.2 Hz, 1H), 4.99-4.95 (m, 1H), 4.91-4.85 (m, 1H), 4.20-4.11 (m, 1H), 2.07 (s, 3H), 2.02 (s, 3H), 1.99 (s, 3H), 1.79-1.76 (m, 2H), 1.51 (bs, 2H), 1.24 (bs, 25H), 1.10 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 6.8 Hz, 3H); **^{13}C NMR** (100 MHz, $CDCl_3$) δ 171.0, 170.7, 169.6, 72.2, 70.0, 48.4, 45.0, 36.2, 35.0, 32.1, 29.8, 29.7 29.5, 25.3, 23.6, 22.9, 21.3, 21.1, 18.6, 14.3.

Enigmol **(31)** data compilation:

John Wiseman ($CDCl_3$)	**Avanti sample ($CDCl_3$)**	**New approach, Claney ($CDCl_3$)**	**David Menaldino ($CDCl_3$)**
3.85 (m, *J* = 4.2, 2.4, 1.2 Hz, 1H)	3.91-3.85 (m, 1H)	3.87 (m, 1H)	3.87 (m, 1H)
3.48 (ddd, *J* = 9.9, 6.6, 2.4 Hz, 1H)	3.45 (ddd, *J* = 2.4, 5.6, 10.2 Hz, 1H)	3.45 (t, *J* = 7.2 Hz, 1H)	3.53 (m, 1H)
3.28 (bs, 4H)			
2.78 (m, *J* = 6.6 Hz, 1H)	2.74 (dq, *J* = 6.4 Hz, 1H)	2.76-2.74 (bm, 1H)	2.85 (qn, *J* = 6.4 Hz, 1H)
1.94 (s, 0.2 H)	1.64 (dt, *J* = 2.4, 14.0 Hz, 1 H)	1.64 (d, *J* = 13.8 Hz, 1 H)	1.65 (dt, *J* = 2.2, 14.3 Hz, 1 H)
1.49 (bm, 1H)	1.45-1.37 (m, 3H)	1.53-1.48 (m, 1H)	1.46 (m, 4H)
1.42 (m, 3H)		1.44-1.39 (m, 3H)	
1.25 (bs, 22H)	1.25 (bs, 20H)	1.25 (bs, 22H)	1.25 (bs, 20H)
1.12 (d, *J* = 6.6 Hz, 3H)	1.12 (d, *J* = 6.4 Hz, 3H)	1.12 (d, *J* = 6.6 Hz, 3H)	1.17 (d, *J* = 6.6 Hz, 3H)
0.88 (t, *J* = 7.2 Hz, 3H)	0.88 (t, *J* = 6.8 Hz, 3H)	0.88 (t, *J* = 7.2 Hz, 3H)	0.88 (t, *J* = 7.0 Hz, 3H)

Enigmol **(31)** data compilation:

John Wiseman ($CDCl_3$)	**Avanti sample ($CDCl_3$)**	**New approach, Claney ($CDCl_3$)**
76.4	76.7	76.6
72.1	72.1	72.1
51.8	51.7	51.7
40.4	40.5	40.5
38.2	38.2	38.2
32.1	32.1	32.1
29.91		
29.83	29.8	29.8
29.54	29.5	29.5
25.7	25.7	25.7
22.9	22.9	22.9
20.6	21.2	21.1
14.3	14.3	14.3

Chapter 3

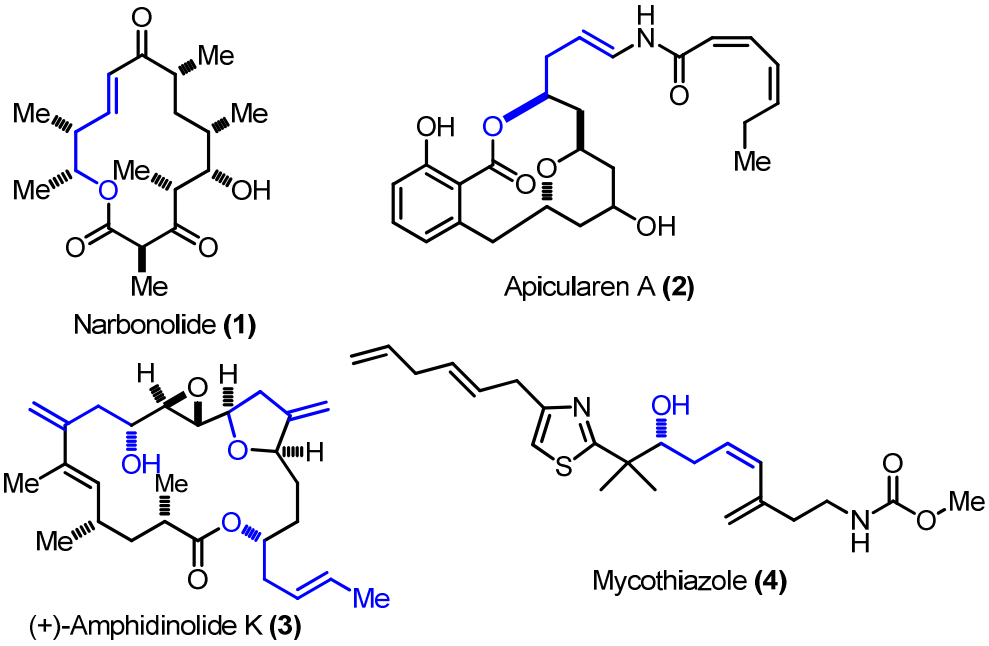
Chapter 3

Development of methodology: synthesis of *cis*- and *trans*-homoallylic alcohols

3.1. Introduction and background

Homoallylic alcohols are important structural motifs that occur in a number of natural products (figure 1, Homoallylic alcohol are highlighted in blue).[84] There are number of ways to synthesize optically active homoallylic alcohols in the literature.[85]

Figure 1. Examples of natural products containing homoallylic alcohols



The widely used methods for synthesizing homoallylic alcohols are carbonyl allylation and carbonyl-ene reactions,[85, 86] which are one of the most attractive ways of forming a C-C bond. The homoallylic alcohol thus formed has both a chiral center and a double bond, which serves as an useful building block

in synthesis. Representative example of a carbonyl allylation and carbonyl-ene reaction is shown in figure 2 and 3.

Figure 2. Homoallylic alcohol **6** by allylation of aldehyde **5** using catalyst **7**[87]

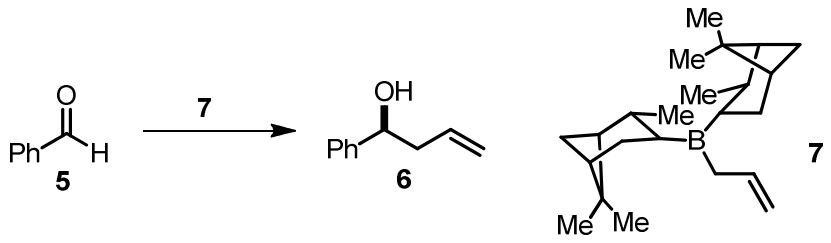
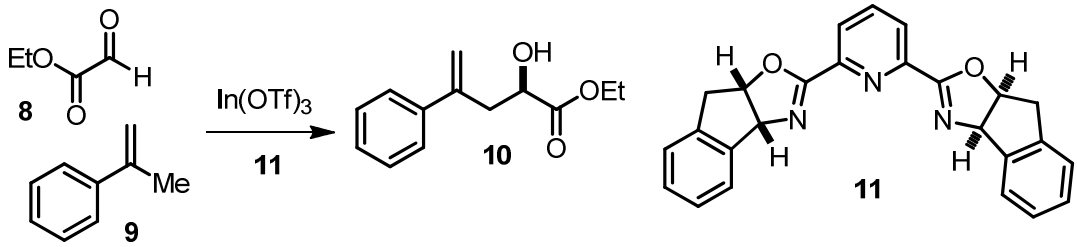
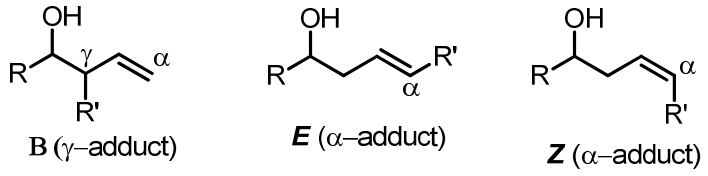


Figure 3. Homoallylic alcohol **10** by carbonyl-ene reaction using catalyst **11**[88]



Enantioselective allylation generates homoallylic alcohol in high *ee's*, but does have its own set of problems. These reactions utilize stoichiometric amounts of allylic metal compounds which by themselves are environmentally unfriendly along with stoichiometric amounts of a chiral catalyst or in combination with stoichiometric amounts of chiral auxillaries. Allylations using catalytic amount of the catalyst have also been developed. The reactions have to be performed under carefully controlled conditions. Furthermore, most of these reactions generate branched homoallylic alcohols of the type **B** (γ-adduct) exclusively (Figure 4). Only allylbarium reagents have thus far been able to generate α-adduct product. Therefore an allylation that generates homoallylic alcohols of the type ***E*** (α-adduct) and ***Z*** (α-adduct) becomes warranted (figure 4).

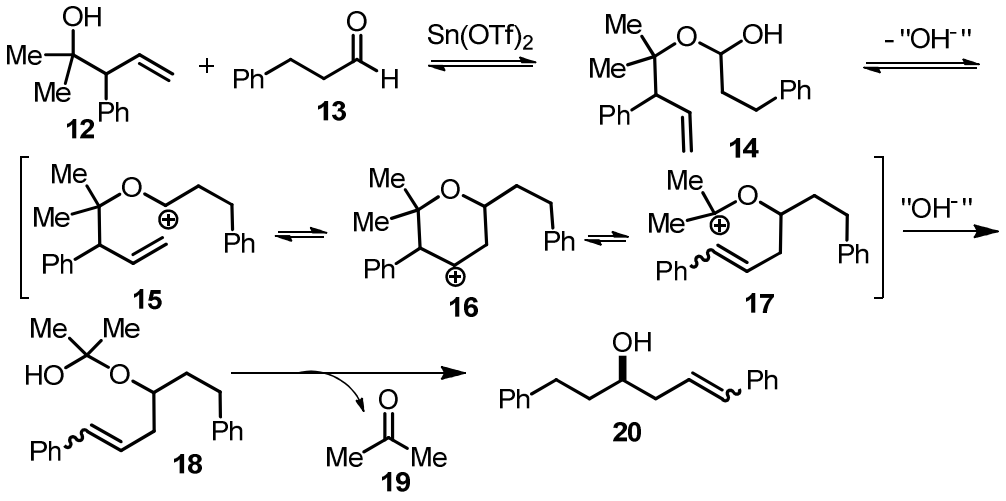
Figure 4. Representative branched **B** vs linear homoallylic alcohol ***E*** and ***Z***



3.2. Allyl transfer and 2-oxonia-[3, 3]-sigmatropic rearrangement

In 1998 Nokami *et al.* reported the first allyl transfer reaction,[89] though at that time it was refered to as crotyl transfer reaction. The term allyl transfer was coined by Nokami in 2001,[34] refering to an allylic functionality transfer from the donor molecule (not a metallic reagent) to acceptor (aldehyde) with 3(γ)- to 1(α)-allylic transposition of the attaching carbon atom. In the first example a branched tertiary homoallylic alcohol **12** reacted with an aldehyde **13** in presense of $Sn(OTf)_2$ to give the linear homoallylic alcohol **20** (scheme 1). [89]

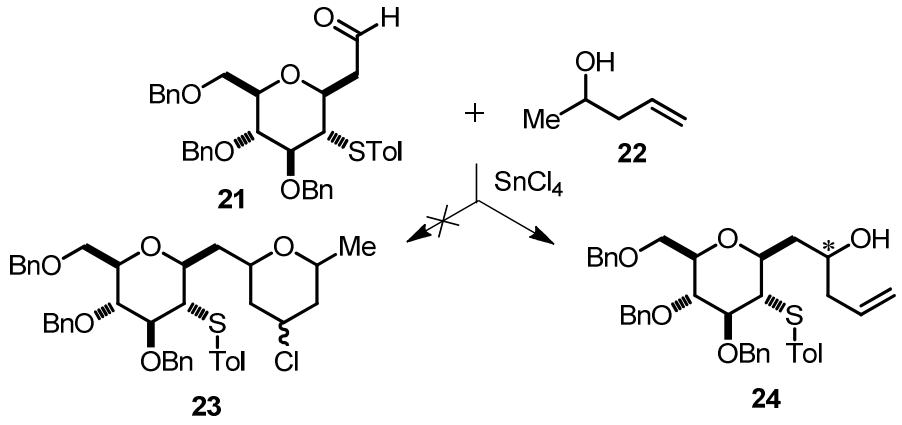
Scheme 1. First example of allyl transfer and proposed mechanism by Nokami



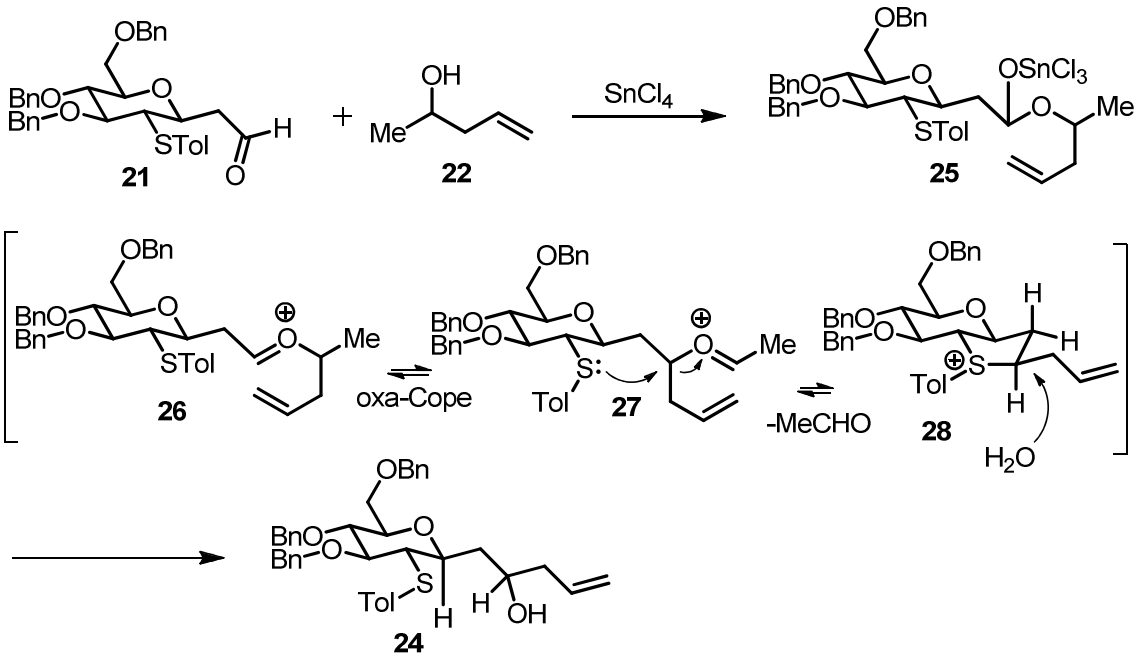
The reaction mechanism was not clear at that point and Nokami proposed a mechanism that is represented in scheme 1. In 1999 Samoshin *et al.* observed

a similar unexpected outcome for a reaction between **21** and **22** that was expected to give a cyclized tetrahydropyran **23** product (scheme 2).[90]

Scheme 2. Unexpected allyl transfer observed by Samoshin *et al*.



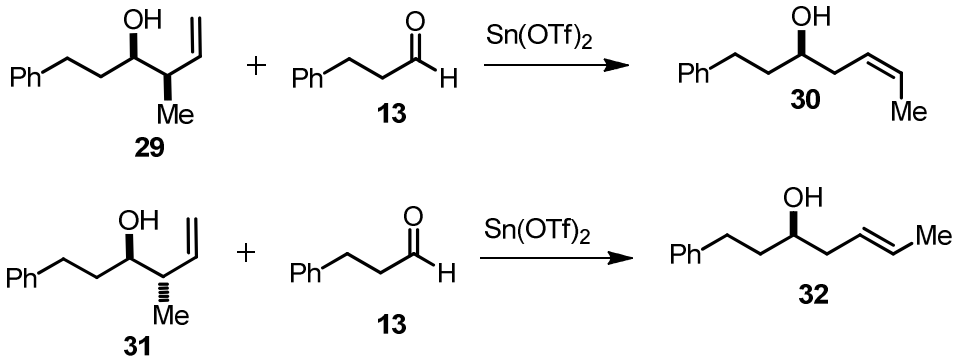
Scheme 3. Proposed mechanism for the above transformation



They proposed the mechanism in terms of an oxa-Cope rearrangement based on the work of Speckamp *et al*.[91] The term 2-oxonia-[3, 3]-sigmatropic rearrangement was first used by Larry Overman in 1987 and was the first precedent for these rearrangement.[92] The allyl transfer reactions that followed

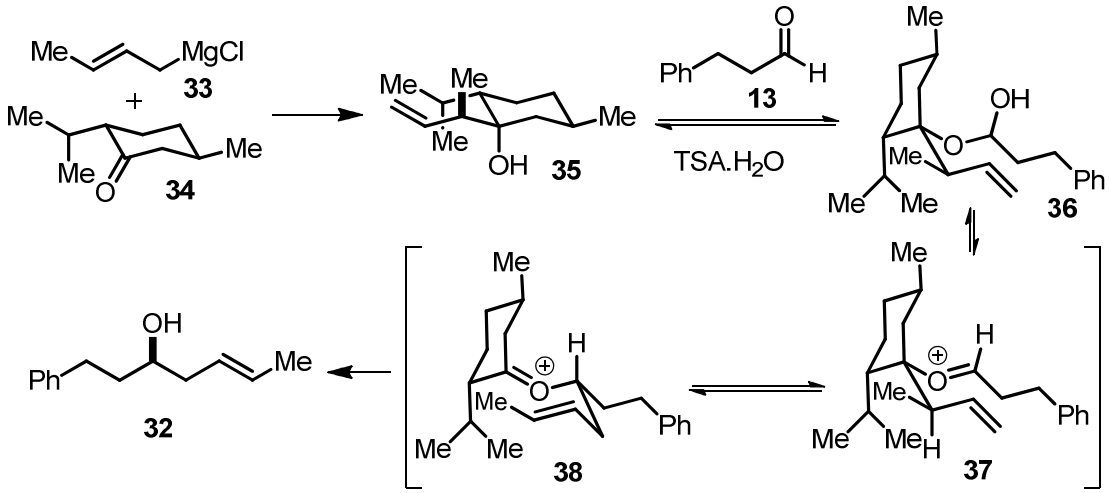
after Nokami's used the term 2-oxonia-[3, 3] sigmatropic rearrangement to explain the outcome. Nokami was able to show that, one obtained *cis*-α-homoallylic alcohol **30** from an optically pure *syn*-γ-adduct **29** and *trans*-α-homoallylic alcohol **32** from an optically pure *anti*-γ-adduct **31**.[93]

Scheme 4. Selectivity of allyl transfer based on the type of γ-adduct used



Based on their initial success Nokami *et al.* reported the first enantioselective allyl transfer reaction using a diastereomerically pure allyl donor **35** obtained from (-)-menthone (scheme 5).[34]

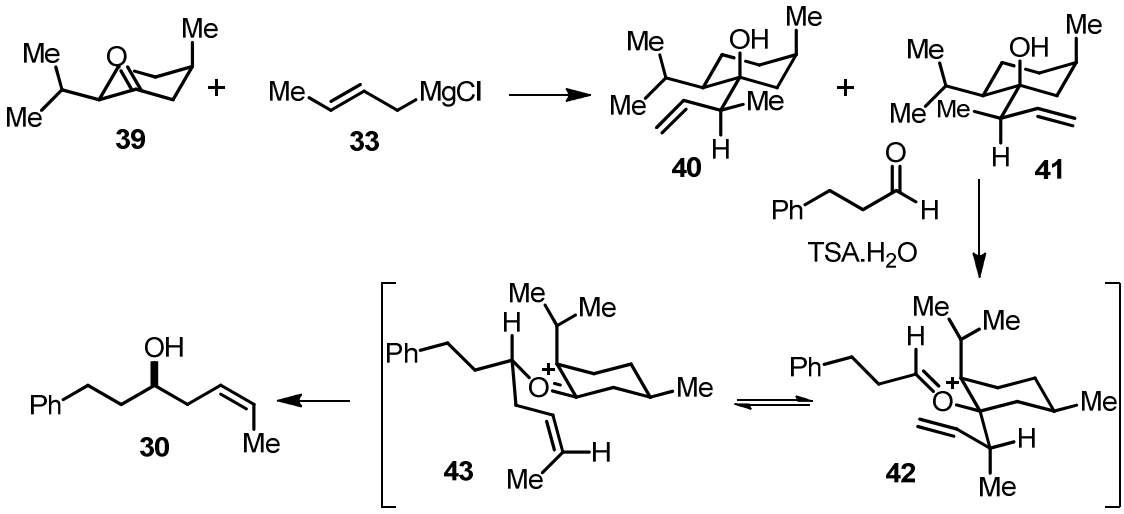
Scheme 5. First enantioselective allyl transfer reaction developed by Nokami



The homoallylic alcohol **32** was obtained in high *trans*-selectivity and >99% ee. The observed selectivity was explained based on a chair like transition state that goes through a 2-oxonia-[3, 3] sigmatropic rearrangement mechanism (scheme 5).

In an extension of the concept Nokami came up with a new allyl donor **40** starting from (+)-Isomenthone (scheme 6).[85d] The allyl transfer reaction was then successfully applied with one of the isolated isomer **41** to obtain *cis*-homoallylic alcohol **30** (scheme 6).

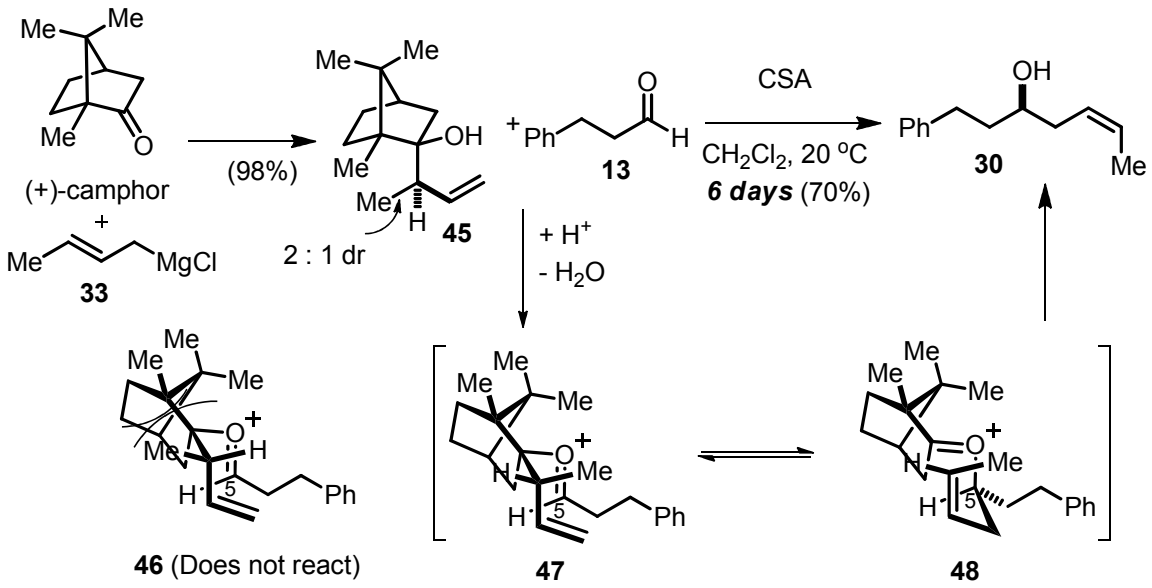
Scheme 6. Synthesis of *cis*-homoallylic alcohol **30** by Nokami



The reaction proceeded through a 2-oxonia-[3, 3]-sigmatropic rearrangement with a six membered chair like transition state (scheme 6). In this synthesis the disadvantage was that the Grignard reaction gave a mixture of isomers **40** and **41** and one needed to separate them and then use allyl isomer **41** for the allyl transfer reaction to give the *cis*-homoallylic alcohol **30**. Using the same allyl transfer concept Loh *et al*. developed their own allyl transfer reagent **45**[42] starting from (R)-(+)-camphor using the crotyl Grignard reagent **33**.[39] Though the

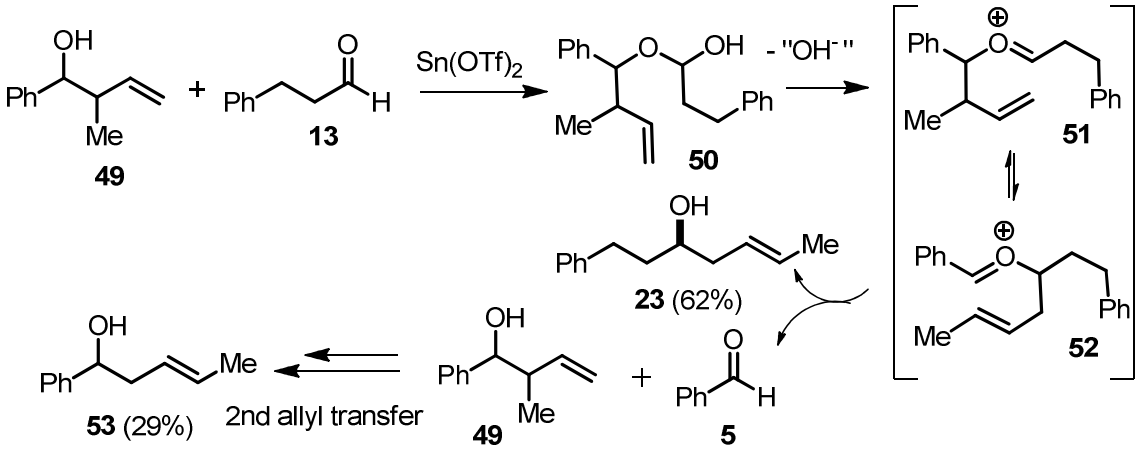
compound **45** was obtained as a 2:1 mixture, it could be taken through the allyl transfer reaction without separating the two isomers. Only isomer **47** reacted to give the *cis*-homoallylic alcohol **30** with high enantioselectivity (scheme 7). The drawback of this approach was the excessive use of the allyl donor reagent **45** and the longer reaction time, typically requiring 5-6 days.[39]

Scheme 7. Synthesis of *cis*-homoallylic alcohol **30** by Loh from (*R*)-(+)-camphor



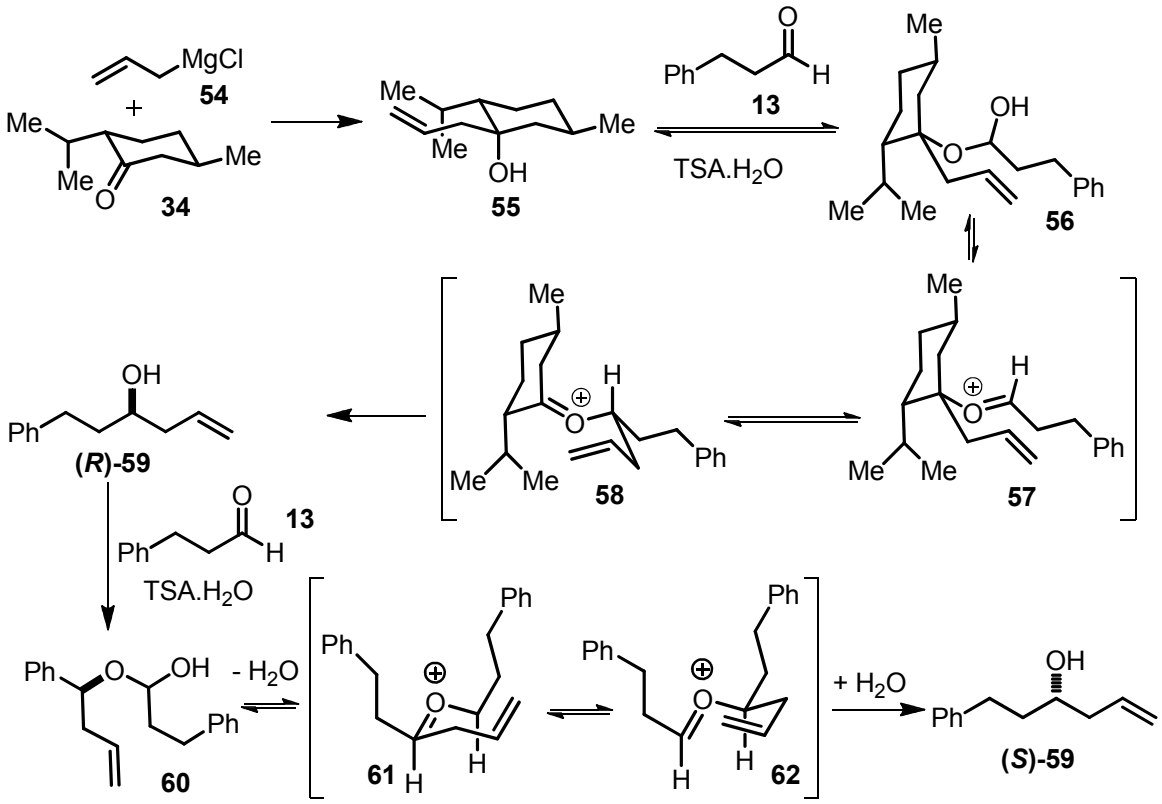
The driving force for these allyl transfer reaction through a 2-oxonia-[3, 3] sigmatropic rearrangement mechanism is the formation of a sterically less hindered and thermodynamically more stable homoallylic alcohol.[89] Though these are novel and useful transformations there are some issues that need to be addressed 1) the unwanted side product **53** that arises from the side reaction of the released aldehyde **5** (scheme 8).[93]

Scheme 8. Byproduct **53** formed during allyl transfer reaction



2) The problem of epimerization that arises due to a second cycle of allyl transfer from the first formed homoallylic alcohol (scheme 9).[94]

Scheme 9. Racemization of **(*R*)-59** to **(*S*)-59** due to allyl transfer



These racemization processes has been observed by many groups involved in both allyl transfer and Prins cyclization reaction and have come up with methods to prevent them.[95]

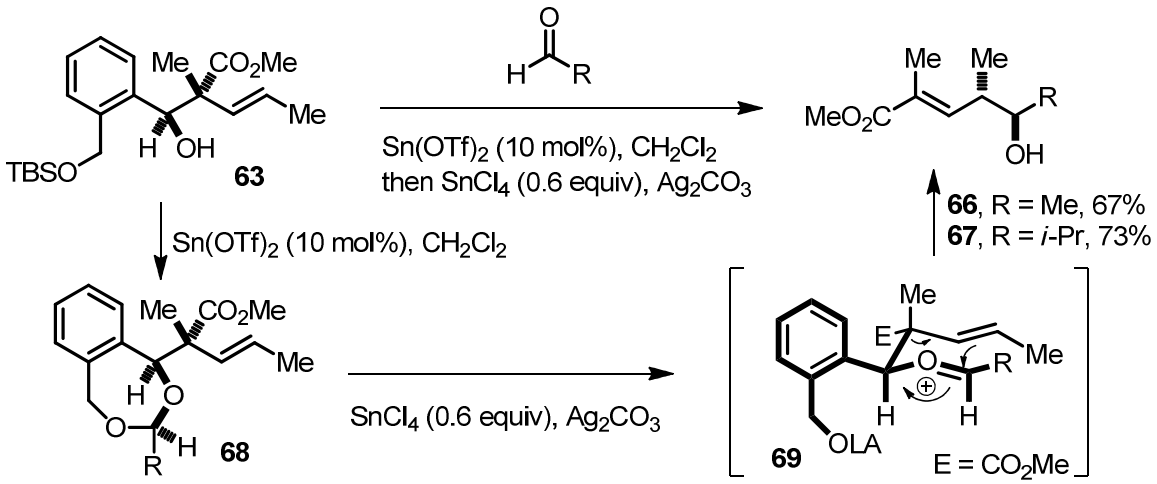
3) The formation of the Prins cyclization product in presence of an external nucleophile to give the tetrahydropyran.[96] Recently there has been a steady rise in the application of 2-oxonia-[3, 3] sigmatropic rearrangement and allyl transfer in the literature. Nokami has used epoxides as an aldehyde equivalent for allyl transfer,[97] allylic sulfones have been synthesized stereoselectively through 2-oxonia-[3, 3]-sigmatropic rearrangement,[98] Loh has used this method to synthesize homoallylic nitrones,[99] Lee *et al.* used it for the diastereoselective 1,3-dimethylallylation of aldehyde,[100] Nokami *et al*. used it for the enylation of aldehyde[101] and lastly there has been the application of Loh's allyl transfer method in the synthesis of natural product (+)- neopeltolide.[102]

3.3. New concept and synthetic design for allyl transfer

The McDonald laboratory is interested in synthesizing complex polyketide based natural products. Since homoallylic alcohols form a key intermediate towards their synthesis, an allyl transfer reaction becomes an obvious choice in this direction. In this regard the McDonald laboratory has developed their own set of reagents to carry out this transformations.[30] The work shown in scheme 10 was carried out by Dr. Yi-Hung Chen, who developed the synthon **63** for the transfer of bispropionates.[30] Very recently this concept was applied in the

synthesis of a key homoallylic alcohol intermediate involved in the total synthesis of fumonisin B_1.[59]

Scheme 10. Allylic rearrangement with synthon **63**

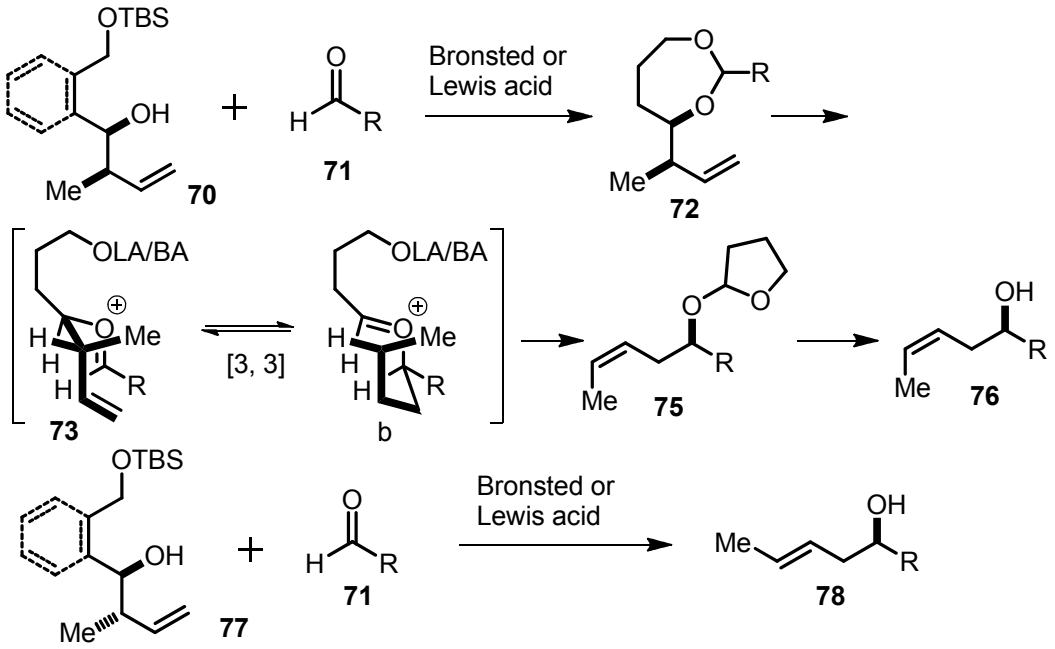


3.4. Results and Discussion

3.4.1. Synthesis of synthon 80

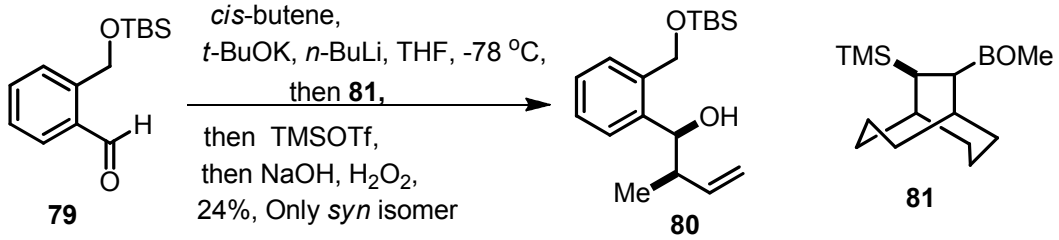
The next question was whether this methodology could be extended to a linear homoallylic alcohol without an α, β-conjugated system. With all the problems of allyl transfer in mind, synthons of the type **70** and **77** were envisioned (scheme 11). The tether on these molecules was positioned in a way that it could intercept the oxonium ion after the 2-oxonia-[3, 3] sigmatropic rearrangement to give **75** so as to prevent the byproduct aldehyde generated from entering into the allyl transfer cycle, thereby totally eliminating the side product mentioned in scheme 8.[103] The *syn*-γ-synthon **70** would give the *cis*-α-homoallylic alcohol **76** and the *anti*-γ-synthon **77** would give the *trans*-α-homoallylic alcohol **78**.

Scheme 11. Development of the concept for linear homoallylic alcohols



To make the method more viable the synthons **70** and **77** should be obtained in high diastereo- and enantioselectivity with fewer steps, preferably in one or two steps. If enantioselectivity is poor, one could use kinetic resolution to enhance it,[104] but higher diastereoselectivity was essential. The first approach towards the synthesis of synthon **80** is outlined in scheme **12** using the Soderquist method of crotylation.[105] The reagent **81** was synthesized as reported and the synthon **80** was obtained in > 95% *ee* and as a single diastereomer starting from aldehyde **79**.

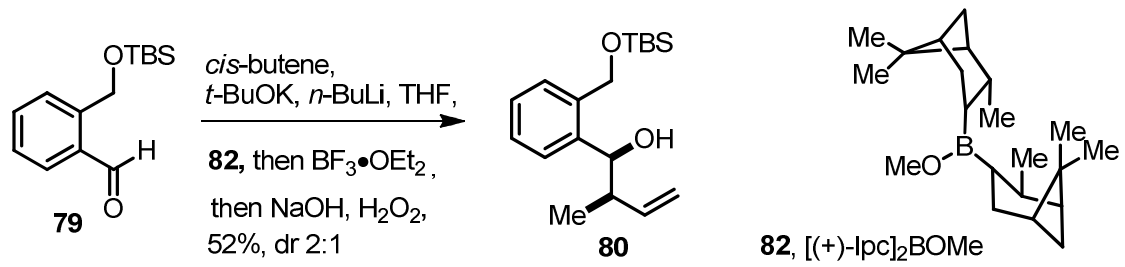
Scheme 12. Synthesis of synthon **80** using Soderquist method



.

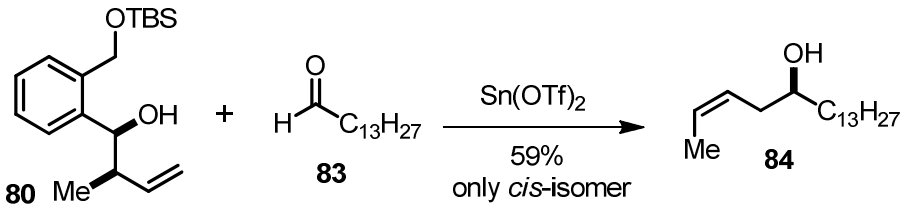
Although this method gave compound **80**, this transformation was very difficult from a practical standpoint because of the number of steps involved to make the reagent **81** needed for the transformation.[105] An attempt to make the synthon **80** using Brown's method resulted in poor diastereoselectivity though obtained in reasonable yields (scheme 13).[106]

Scheme 13. Synthesis of synthon **80** using Brown's method



Before venturing further onto a new approach to make synthon **80** it was important to prove the concept of allyl transfer reaction using the synthon **80**. In this regard a reaction carried out with tetradecanal **83** in presense of $Sn(OTf)_2$ gave exclusively the *cis*-homoallylic alcohol **84** in 59% yield and >95% ee as determined through Mosher ester analysis (scheme 14).[107]

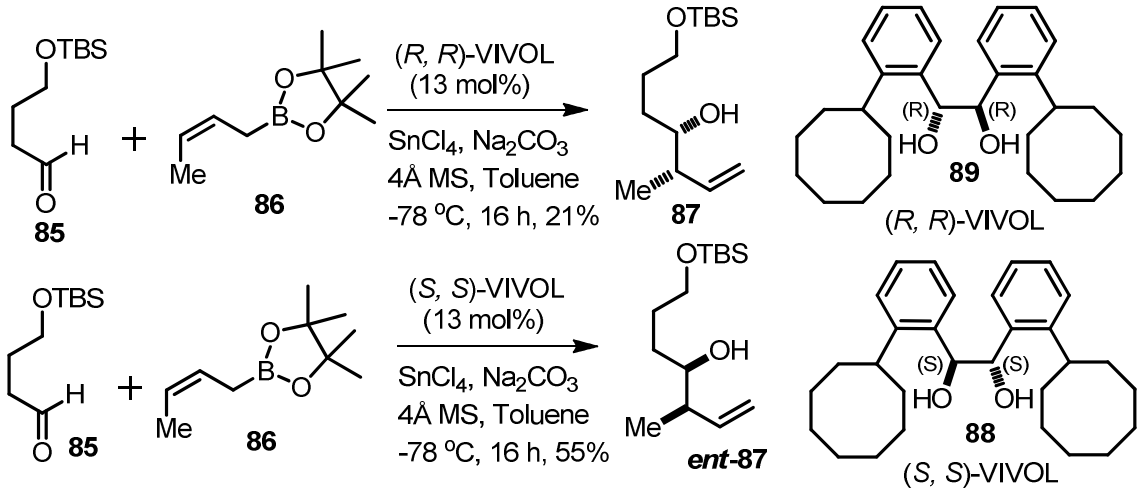
Scheme 14. Synthesis of linear *cis*- homoallylic alcohol **84** from synthon **80**



3.4.2. Synthesis of *syn* synthon 87 and *ent*-87

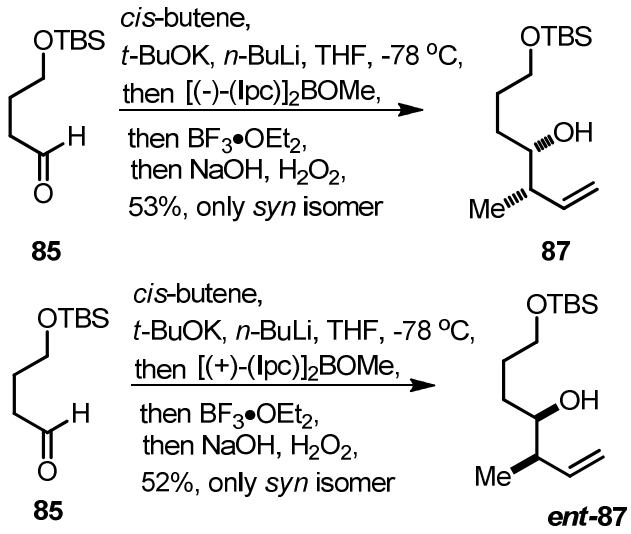
With this result it was important to synthesize the synthon **80** in larger quantities using an effective method and provide evidence for the proposed mechanistic rationale in scheme 11. Dennis Hall had reported the application of their VIVOL based catalyst in the synthesis of similar compounds.[108] The catalyst was found to be more effective in the crotylation of aliphatic aldehydes.

Scheme 15. Synthon **87** and ***ent*-87** from aliphatic aldehyde **85** using VIVOL



The VIVOL catalyst **88** and **89** were synthesized from the literature precedents.[108] (Obtained also through a generous donation from Dennis Hall's laboratory). A reaction carried out with *Z*-pinacol boronate ester **86** with aldehyde **85** resulted in synthons **87** and ***ent*-87** respectively as a single diastereomer with > 95% ee (Scheme 15). Similar results were obtained when the Brown method was used with the aliphatic aldehyde **85** (scheme 16).[106]

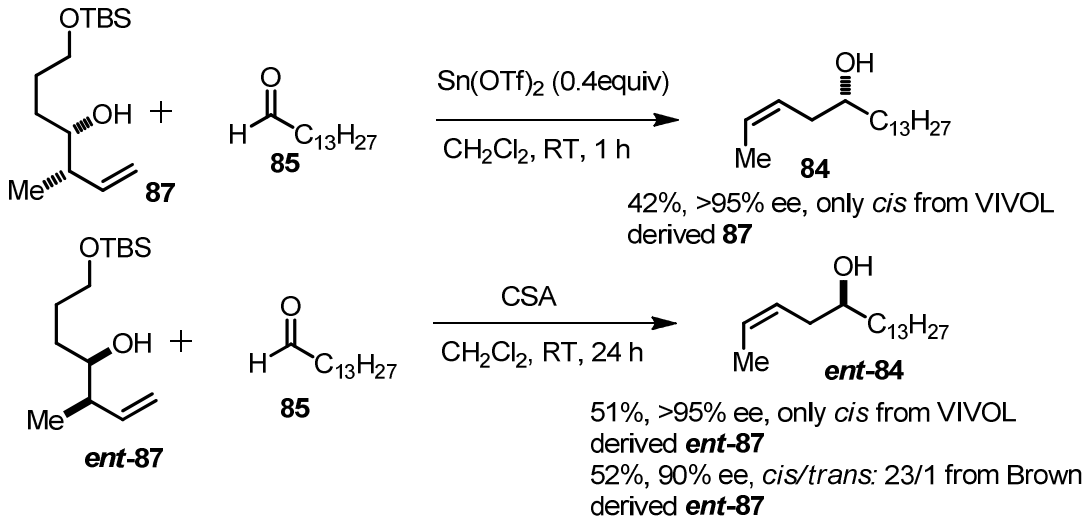
Scheme 16. Synthon **87** and ***ent*-87** from aldehyde **85** using Brown method



3.4.3. Allylic rearrangements studies with synthon 87 and *ent*-87

With these new synthons **87** and ***ent*-87**, the allyl transfer reaction carried out with tetradecanal **83** gave the homoallylic alcohol **84** and ***ent*-84** in reasonable yields and selectivity (scheme 17).

Scheme 17. *cis*-homoallylic alcohol **84** and ***ent*-84** from synthon **87** and ***ent*-87**

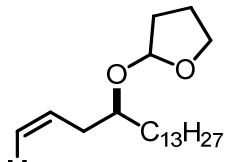


The various reaction conditions that were attempted are summarized in table 1 along with the byproducts (figure 4) that were isolated in some of these reactions.

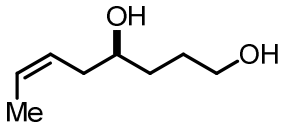
Table 1. Conditions attempted for allyl tranfer with synthon **87** and ***ent*-87** and tetradecanal **85**

Reaction condition	yield	*cis/trans*	byproducts
CSA, CH_2Cl_2, RT, 24 h	44%		9% of **89**
CSA, CH_2Cl_2, RT, 24 h, then MeOH, 1h (0.047 mmol scale)	51%		-
$In(OTf)_3$, toluene, RT, 18 h	16%	7/1	-
TMSOTf, CH_2Cl_2, 0 °C, 30 min	40%	11/1	16% of **89**
TMSOTf, CH_2Cl_2, 0 °C, 30 min, then MeOH, 15 min	40%		Isolated **89**, **90** and **91**
CSA, $CHCl_3$, RT, 24 h	49%	11/1	
CSA, CH_2Cl_2, RT, 24 h, then MeOH, 1 h (1.17 mmol scale)	52%	23/1	17% of **89** (impure)

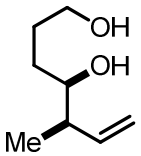
Figure 5. Byproducts observed during allyl transfer reaction



89



90



91

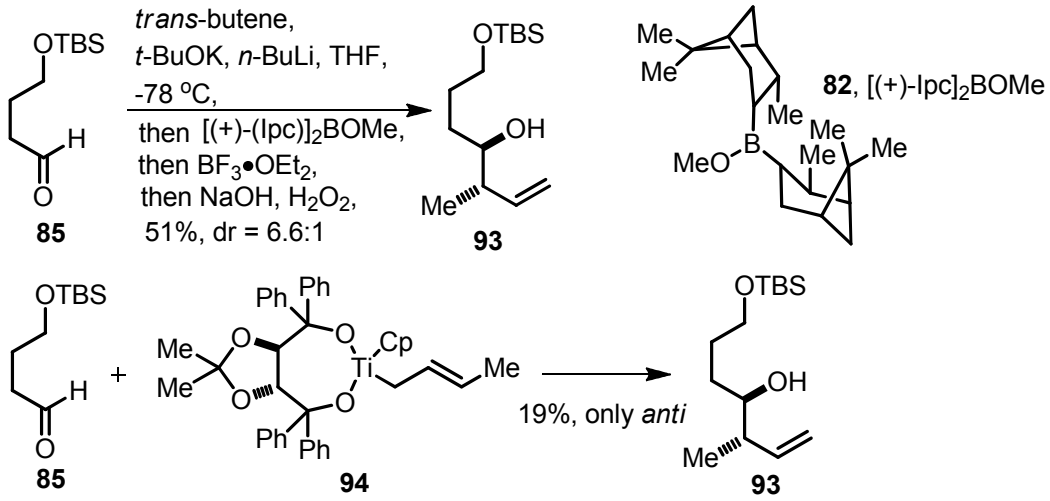
These results clearly indicated the superiority of synthon **87** and ***ent*-87** compared to the one reported by Loh or Nokami for the synthesis of *cis*-homoallylic alcohol especially in terms of shorter reaction time (1 day compared

to 6 days), the equivalence of synthon used (1.1 equiv vs 3.0 equiv) and the synthesis of the synthon **87** and ***ent*-87** in high diastereoselectivity thereby avoiding the separation of the synthon diastereomers as in the case of Nokami. Out of all the methods attempted for the synthesis of synthon **87** and ***ent*-87**, the Brown method was opted as the method of choice because of the commercial availability of the reagent.

3.4.4. Synthesis of *anti*-synthon 93 and *ent*-93 and its allylic rearrangement

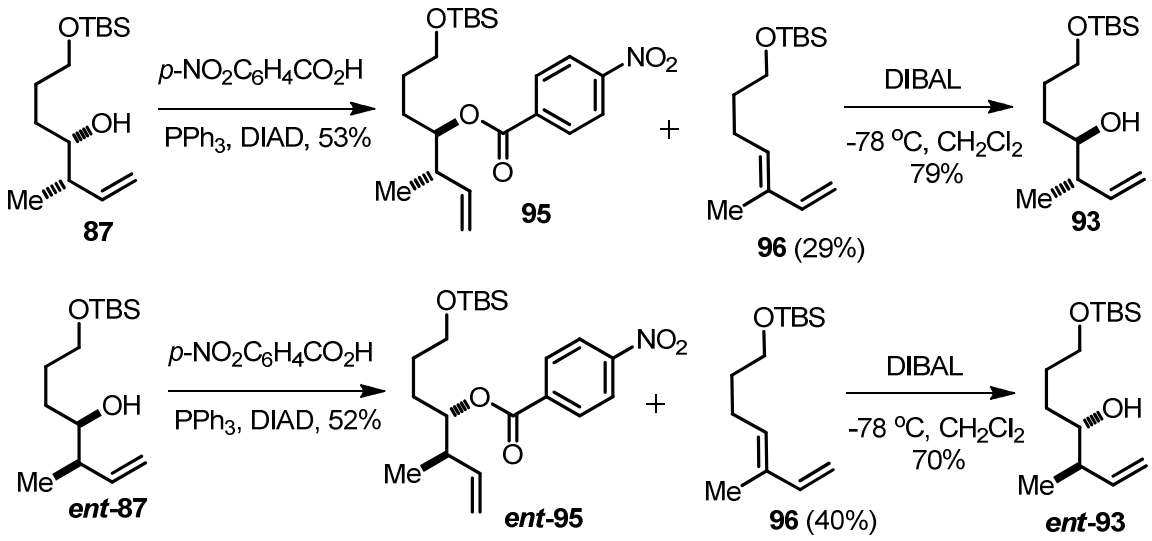
With the success of the *syn*-synthon **87** and ***ent*-87** in the synthesis of *cis*-homoallylic alcohol **84** and ***ent*-84** the next approach was to synthesize the *trans*-homoallylic alcohol. In this regard the Brown method of crotylation was adopted using the aliphatic aldehyde **85** to obtain the synthon **93** only in a dr of 6.6:1 (scheme 18).[106]

Scheme 18. Synthesis of *anti*-synthon **93** using Brown and Hafner reagents



Using the titanium based Hafner catalyst **94**,[109] the crotylation with aldehyde **85** led to the formation of the synthon **93** as a single diastereomer but in low yields (scheme 18). To circumvent this problem of low yield and selectivity a Mitsunobu inversion and hydrolysis protocol was considered.[110] Inversion carried out with *p*-nitrobenzoic acid and DIAD gave compound **106** and **107** and the elimination product **96** (scheme 19). Hydrolysis of these compounds using DIBAL gave *anti*-synthon **93** and ***ent*-93** in >95% ee as determined by Mosher ester analysis.

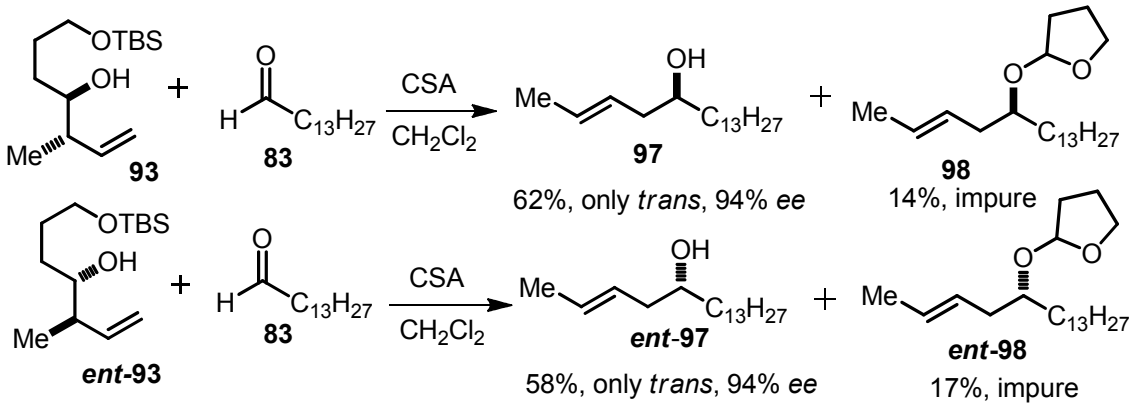
Scheme 19. Synthesis of *anti*-synthon **93** and ***ent*-93** using Mitsunobu protocol



3.4.5. Allylic rearrangement with synthon 93 and *ent*-93

With the required *anti*-synthon **93** and ***ent*-93** an allyl transfer reaction under the optimised conditions of camphorsulphonic acid at room temperature for 24 h using aldehyde **83** resulted in the formations of *trans*-homoallylic alcohol **97** and ***ent*-97** in reasonable yield, *trans* selectivity and enantioselectivity.

Scheme 20. Synthesis of *trans*-homoallylic alcohol **97** and ***ent*-97** from *anti*-synthons **93** and ***ent*-87**



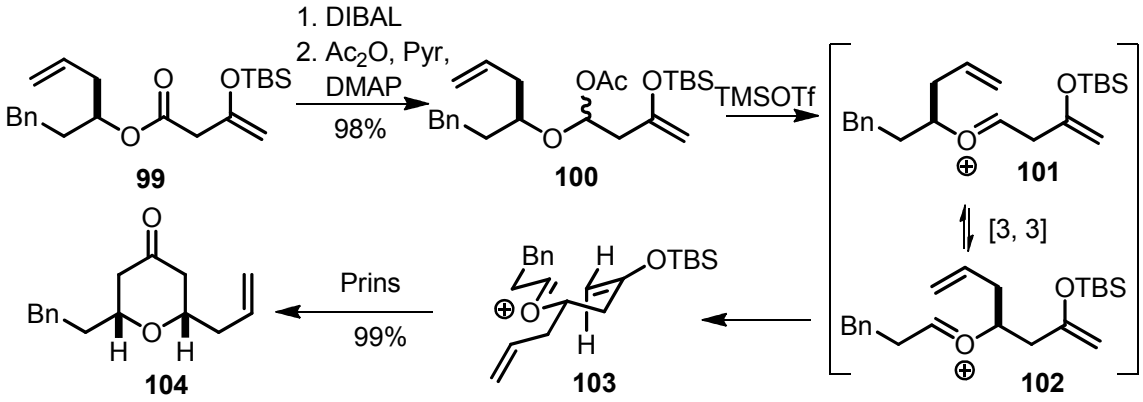
To provide evidence for the proposed mechanism (scheme 11) the allyl transfer reactions were monitored by ^{1}H NMR. The formation of the seven membered ring acetal was very evident within 30 min. (no aldehyde peak was observed) One could even isolate these stable intermediates as observed during the synthesis of fumonisin B_1 (chapter 1, page 52).[59] The tetrahydrofuranyl acetal intermediates **89**, **98** and ***ent*-98**, had never been isolated previously during these rearrangements. In this case these intermediates were isolated and characterized by NMR thereby providing further proof for the proposed mechanism.

3.4.6. Allyl transfer using hemiacetal generated from Rychnovsky method

In the allyl transfer reactions discussed so far the synthon and the aldehyde were mixed together to generate the hemiacetal which then underwent the 2-oxonia-[3, 3] sigmatropic rearrangement to give the product. Rychnovsky

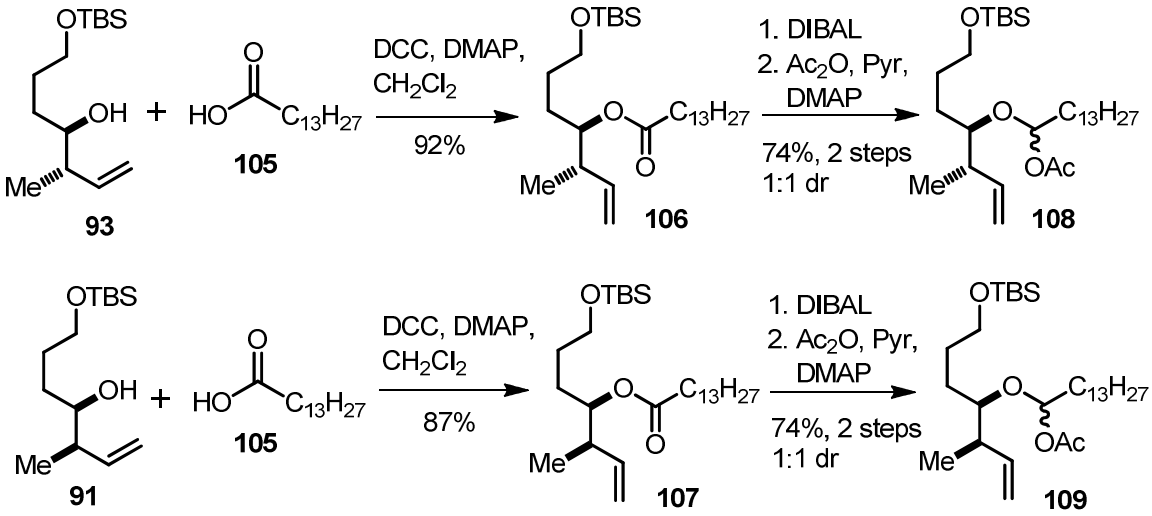
had demonstrated that one could carry out a similar transformation leading to a Prins cyclised product through a 2-oxonia-[3, 3]-sigmatropic rearrangement of a hemiacetal without using an aldehyde source (scheme 21).[111]

Scheme 21. Prins cyclization via 2-oxonia-[3, 3]-sigmatropic rearrangement



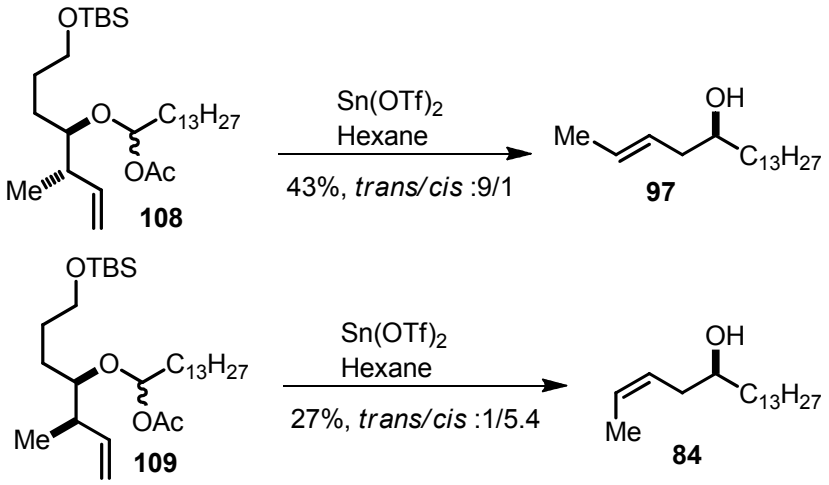
Using this, intermediate **108** was synthesized in three steps starting from synthon **93** through DCC mediated coupling with tetradecanoic acid **105**, followed by a DIBAL reduction and acylation (scheme 22).[111] Similarly intermediate **109** was obtained starting from the synthon **91** (scheme 22).

Scheme 22. Synthesis of hemiacetals **108** and **109** using Rychnovsky method



An allyl transfer reaction carried out with hemiacetals **108** and **109** using $Sn(OTf)_2$ gave *trans* and *cis*- homoallylic alcohol **97** and **84** respectively, but in low yield and reasonable *trans* to *cis* selectivity (scheme 23). Further research is required to find an optimized reaction condition to carry out these transformations successfully.

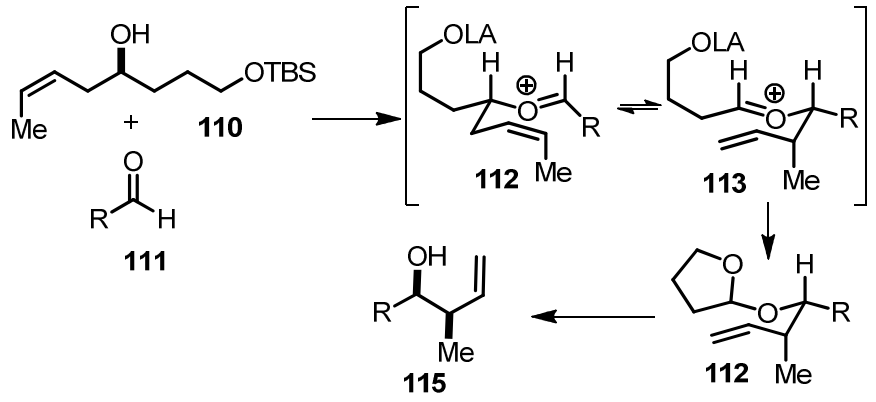
Scheme 23. Synthesis of *trans* and *cis*-homoallylic alcohol **97** and **84**



3.4.7. Synthesis of branched homoallylic alcohol from linear homoallylic alcohol

The allyl transfer reaction is a reversible reaction. Is it then possible to go from a linear homoallylic alcohol to a branched homoallylic alcohol? In other words can we achieve regioselectivity? To test this hypothesis once again we needed to rely on a synthon of the type **110** that could carry out the 2-oxonia-[3, 3] sigmatropic rearrangement (scheme 24).

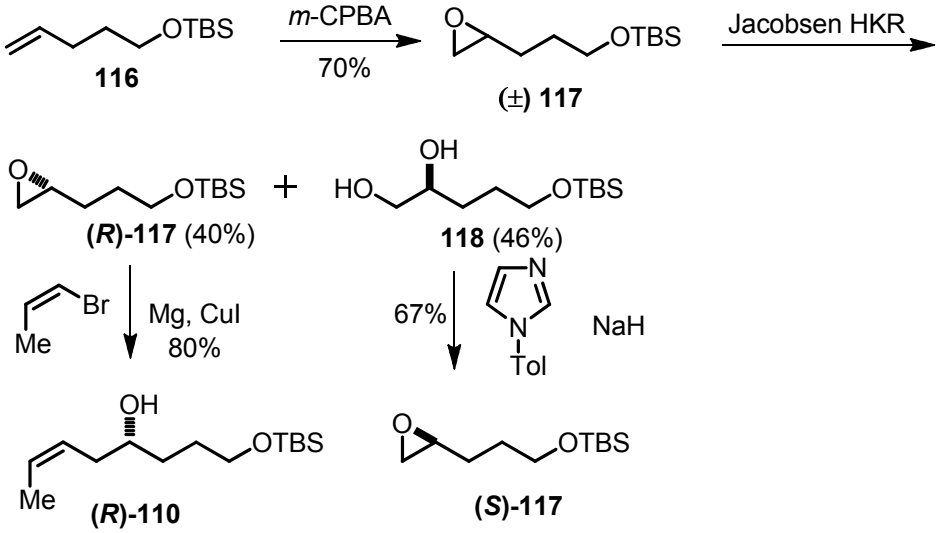
Scheme 24. Hypothesis for the synthesis of branched homoallylic alcohol **115** from linear homoallylic alcohol **110**.



3.4.8. Synthesis of the linear homoallylic alcohol synthon 110

Starting from the terminal alkene **116**, *m*-CPBA epoxidation gave the racemic epoxide **117** which was resolved using Jacobsen's hydrolytic kinetic resolution to obtain (*R*)-epoxide **117** and diol **118** (scheme 25).[112]

Scheme 25. Synthesis of homoallylic alcohol synthon **110**



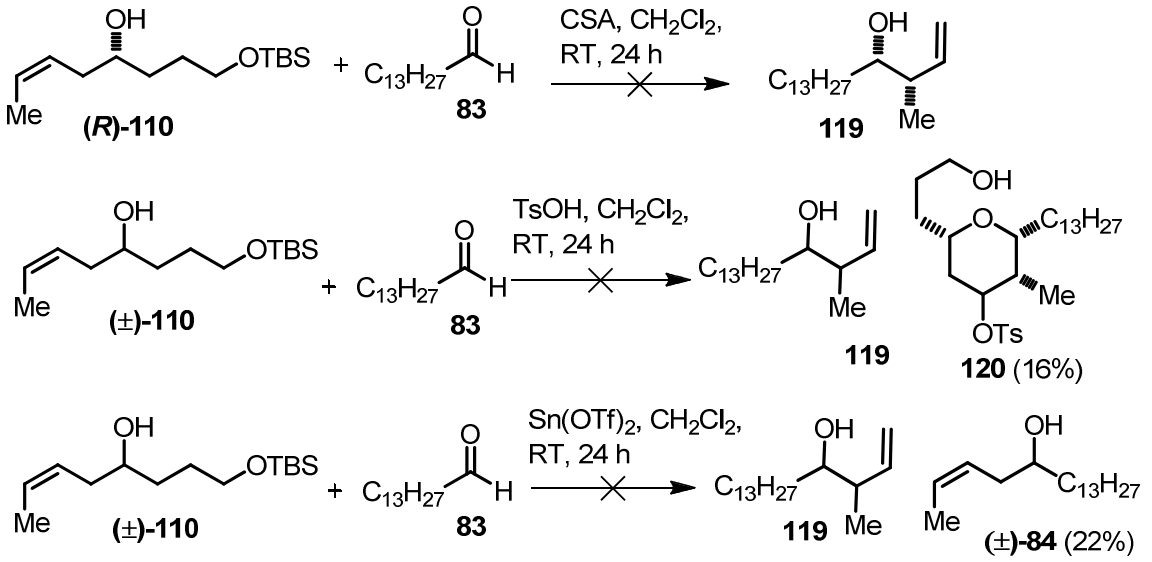
The diol **118** was converted to the enantiomer of epoxide **117** in a single step using tosylimidazole and NaH. After attempting to open the epoxide by different methods, a CuI catalyzed addition of the Grignard reagent of *cis*-1-

bromo-propene gave the required linear homoallylic alcohol **110** in 80% yield (scheme 25).[113] The enantioselectivity was found to be >95% by Mosher ester analysis. The racemic synthon **110** was obtained in 78% yield in a similar manner from ***rac*-117**.

3.4.9. Allylic rearrangement studies with homoallylic alcohol synthon 110

Using the standard conditions developed in the laboratory for allyl transfer through 2-oxonia-[3, 3]-sigmatropic rearrangement, attempted reactions with synthon **110** and tetradecanal **83** failed to give the desired branched homoallylic alcohol **119** (scheme 26). When toluenesulphonic acid was used as the catalyst, a Prins cyclized product **120** was obtained (scheme 26), wherein the TsOH was the nucleophile.

Scheme 26. Attempted synthesis of branched homoallylic alcohol **119** using linear homoallylic alcohol synthon **110**

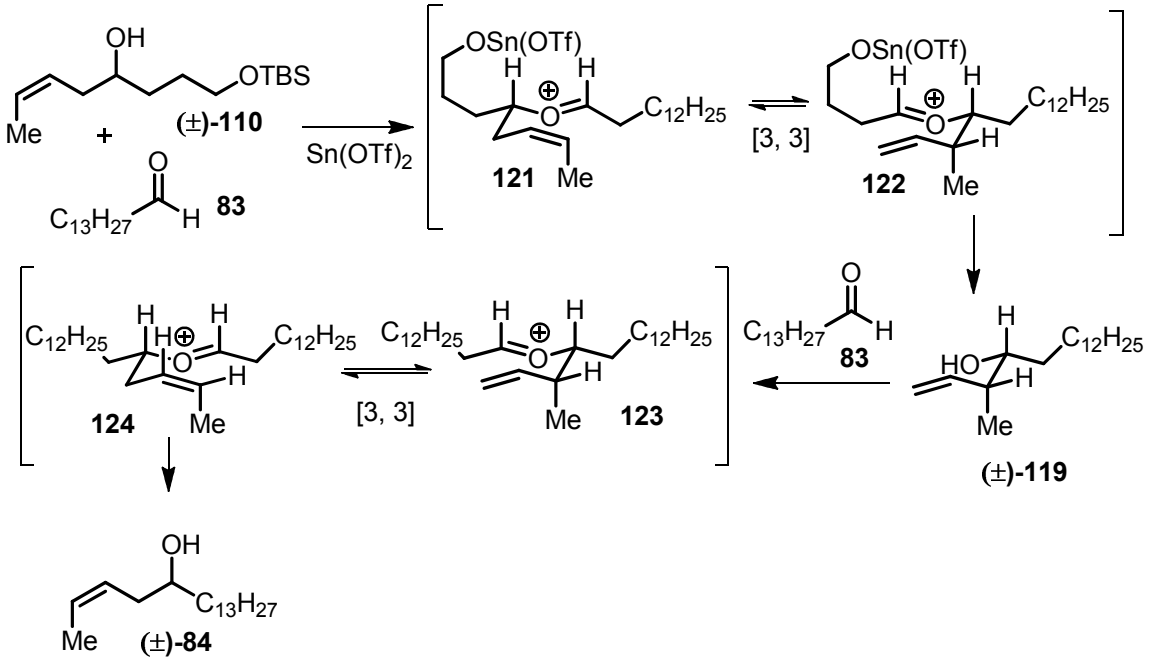


Although some of the byproducts that were obtained in the allyl transfer reaction for the synthesis of linear homoallylic alcohol **84** and **97** might have

been Prins byproducts, the assignment could not be confirmed with certainity because of the impure spectra obtained. For the first time a spectra indicating the formation of a Prins cyclization product, clearly indicates that this might be the case. The use of $Sn(OTf)_2$ as a catalyst for the transformation resulted in the formation of a linear homoallylic alcohol *rac*-**84** from a linear homoallylic alcohol (scheme 26).

This was clear evidence that the branched homoallylic alcohol **119** does form during the reaction but undergoes a second allyl transfer reaction to give the linear homoallylic alcohol *rac*-**84**. The mechanism for the outcome of the reaction is outlined in scheme 27. This outcome was also observed by Loh when he reported the first example of an allyl transfer reaction involving a linear homoallylic alcohol and an aldehyde.[114]

Scheme 27. Mechanistic rationale for the formation of linear homoallylic alcohol ***rac*-84** from linear homoallylic alcohol **110**



At this point in time further research on this transformation is required. Variations in the type of catalyst (Lewis acid or Bronsted acid), temperature and solvent must be considered. As mentioned before the driving force for the allyl transfer reaction is that, from a branched γ-homoallylic alcohol, which is sterically hindered and thermodynamically unstable (1,1 disubstituted alkene), we go to a linear α–homoallylic alcohol which is sterically less hindered and thermodynamically stable (1, 2-disubstituted alkene). Thus reaction conditions need to be optimized with this in mind so as to avoid the second cycle of allyl transfer.

3.5. Experimental details

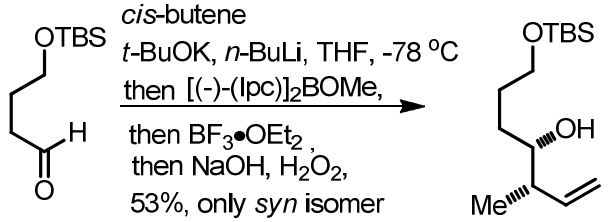
General information: ^{1}H NMR and ^{13}C NMR spectra were recorded on Varian INOVA 600, Unity 600 and INOVA 400 spectrometers. NMR spectra were recorded in solutions of deuterated chloroform ($CDCl_3$) with the residual chloroform (7.27 ppm for ^{1}H NMR and 77.23 ppm for ^{13}C NMR) taken as the internal standard, deuterated methanol (CD_3OD) with residual methanol (3.31 ppm for ^{1}H NMR and 49.3 ppm for ^{13}C NMR) taken as the internal standard, or deuterated benzene with residual benzene (7.16 ppm for ^{1}H NMR and 128.23 ppm for ^{13}C NMR) taken as the internal standard, and were reported in parts per million (ppm). Abbreviations for signal coupling are as follows: s, singlet; d, doublet; t, triplet; q, quartet; dd, doublet of doublet; ddd, doublet of doublet of doublet; dt, doublet of triplet; m, multiplet.

IR spectra were collected on a Mattson Genesis II FT-IR spectrometer as neat films on sodium chloride discs. Mass spectra (high resolution ESI and APCI) were recorded on a Finnigan LTQ FTMS Mass spectrometer. Optical rotations were measured using a Perkin-Elmer 341 polarimeter (concentration in g/100mL). Thin Layer Chromatography (TLC) was performed on precoated glass backed plates purchased from Whatman (silica gel $60F_{254}$; 0.25mm thickness). Flash column chromatography was carried out with silica gel 60 (230-400 mesh ASTM) from EM Science.

All reactions were carried out with anhydrous solvents in oven dried or flame dried and argon-charged glassware. All anhydrous solvents were dried with 4 Å molecular sieves purchased from Sigma-Aldrich and tested for trace

water content with Coulometric KF titrator from Denver instruments. All solvents used in extraction procedures and chromatography were used as received from commercial suppliers without prior purification.

Synthesis of branched *syn*-synthon 87

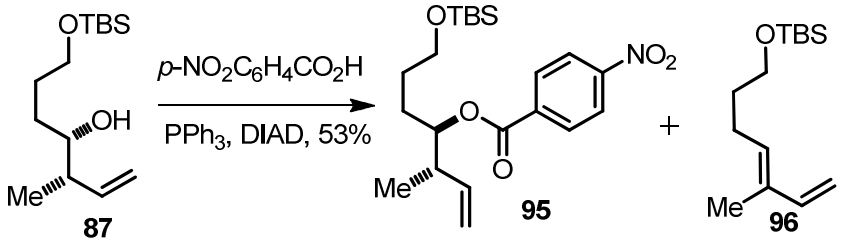


85 **87**

Cis-2-butene (3.7 g, 65.9 mmol) was added to a cooled solution of *t*-BuOK (1.8g, 16.0 mmol) in THF (34 mL) at -78 °C. *n*-BuLi (1.02 g, 16.0 mmol, 2.5 M in hexanes) was then added and the yellow reaction mixture warmed to -45 °C and stirred for 10 min. After cooling back to -78 °C, $[(-)Ipc]_2BOMe$ (5.0 g, 16.0 mmol) in THF (27 mL) was then added and the reaction mixture stirred for 30 min. $BF_3•OEt_2$ (2.27 g, 16.0 mmol) was then added, followed by aldehyde **85** (2.7 g, 13.3 mmol) and the reaction stirred for 5 h. The reaction was quenched by the addition of 3 M NaOH (13.5 mL), followed by 30% H_2O_2 (10.0 mL) and stirred the reaction at room temperature for 18 h. The aqueous layer was then extracted with ethyl acetate, dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 8% ethyl acetate and hexanes to obtain synthon **87** (1.7 g, 50%) as colorless oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.80 (ddd, *J* = 7.2, 10.5, 17.4 Hz, 1H), 5.08-5.04 (m, 2H), 3.66 (t, *J* = 5.7 Hz, 2H), 3.50-3.46 (m, 1H), 2.45 (d, *J* = 4.2, 1H), 2.29-2.26 (m, 1H), 1.68-1.61 (m, 3H), 1.42-1.37 (m, 1H), 1.04 (d, *J* = 6.6 Hz, 3H), 0.90 (s, 9H), 0.07 (s, 6H); **^{13}C NMR**

(150 MHz, $CDCl_3$) δ 141.4, 115.0, 74.7, 63.6, 43.8, 31.4, 29.5, 26.1, 18.5, 14.9, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{14}H_{31}O_2Si$ ($M+H^+$) 258.2087, found 258.2087; **FT-IR**: 3367, 2954, 2857, 1471, 1253, 1095, 995, 832, 773 cm^{-1} ; $[\alpha]_D^{25}$ = + 19.0 (c=1.23, $CHCl_3$).

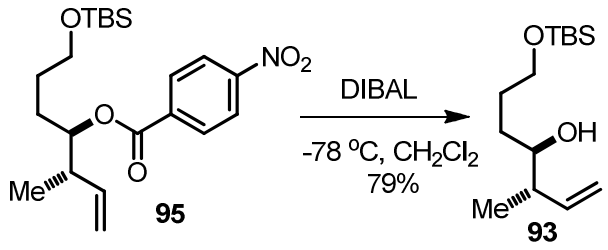
Synthesis of Mitsunobu inversion ester 95



To a solution of the synthon **87** (0.8 g, 3.0 mmol), PPh_3 (1.6 g, 6.19 mmol), and *p*-nitrobenzoic acid (1.0 g, 6.19 mmol) in benzene (40 mL) at room temperature was added DIAD (1.25 g, 6.19 mmol). The solvents were removed under vacuum after stirring the reaction for 24 h to obtain a semisolid. Purified the crude by flash chromatography using 1% ether in hexanes to obtain compound **95** (0.67 g, 53%) as oil. We obtained also the elimination product **96** in 29% yield. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 8.29 (d, *J* = 8.4 Hz, 2H), 8.20 (d, *J* = 9.6 Hz, 2H), 5.80 (ddd, *J* = 7.8, 10.2, 17.7 Hz, 1H), 5.15 (ddd, *J* = 4.8, 9.0 Hz, 1H), 5.08 (dd, *J* = 16.8, 18.3 Hz, 2H), 3.65-3.59 (m, 2H), 2.59-2.53 (m, 1H), 1.81-1.70 (m, 2H), 1.60-1.51 (m, 2H), 1.08 (d, *J* = 6.6 Hz, 3H), 0.08 (s, 9H), 0.03 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 164.5, 150.6, 139.2, 136.2, 130.8, 123.7, 116.2, 78.8, 62.7, 42.0, 28.9, 28.0, 26.1, 18.5, 16.3, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{21}H_{34}NO_5Si$ ($M+H^+$) 408.2200, found 408.2202; **FT-IR**: 2954, 2856, 1721, 1528, 1318, 1270, 1098, 872, 831, 717 cm^{-1} ; $[\alpha]_D^{25}$ = - 11.1 (c=0.65, $CHCl_3$). Byproduct **96**: **^{1}H NMR** (600 MHz, $CDCl_3$) δ 6.37 (dd, *J* = 11.4, 17.4 Hz, 1H), 5.50

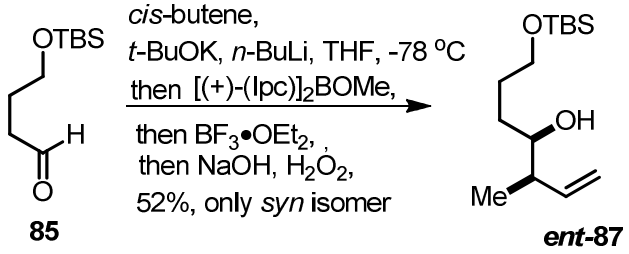
(app t, *J* = 7.2 Hz, 1H), 5.08 (d, *J* = 17.4 Hz, 1H), 4.93 (d, *J* = 10.8 Hz, 1H), 3.62 (t, *J* = 6.6 Hz, 2H), 2.20 (q, *J* = 7.8 Hz, 2H), 1.74 (s, 3H), 1.63-1.59 (m, 2H), 0.90 (s, 9H), 0.05 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 141.7, 134.4, 133.0, 110.6, 62.7, 32.8, 31.8, 26.1, 24.7, 22.8, 18.5, 14.3, 11.8, -5.0.

Synthesis of *anti*-synthon 93



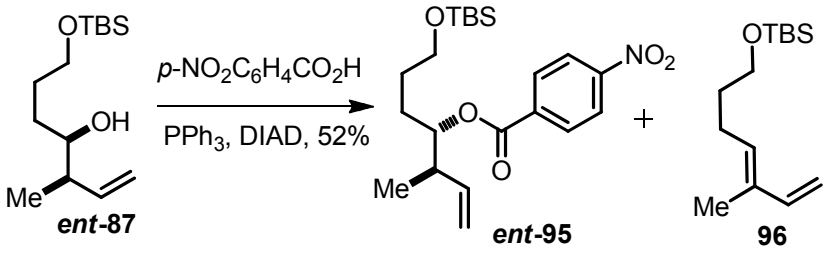
DIBAL (0.7 g, 4.9 mmol, 1.0 M in hexanes) was added to a cooled solution of ester **95** (0.67 g, 1.64 mmol) in CH_2Cl_2 (21 mL) at -78 °C and the reaction mixture stirred for 30 min. The reaction was quenched with ethanol (0.5 mL) and saturated Rochelle's salt. After stirring the reaction mixture at room temperature for 1 h extracted the aqueous with CH_2Cl_2, dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 2-5% ethyl acetate and hexanes to obtain *anti*-synthon **93** (0.27 g, 79%) as colorless oil. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.79 (ddd, *J* = 9.0, 19.2 Hz, 1H), 5.10-5.07 (m, 2H), 3.69-3.63 (m, 2H), 3.45-3.43 (m, 1H), 2.38 (d, *J* = 3.6 Hz, 1H), 2.26-2.20 (m, 1H), 1.68-1.62 (m, 3H), 1.44-1.38 (m, 1H), 1.04 (d, *J* = 6.6 Hz, 3H), 0.90 (s, 9H), 0.06 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 140.8, 115.9, 74.7, 63.6, 44.2, 31.2, 29.3, 26.1, 18.5, 16.2, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{14}H_{31}O_2Si$ ($M+H^+$) 258.2087, found 258.2086; **FT-IR**: 3399, 2954, 2856, 1471, 1253, 1094, 832, 773 cm^{-1} ; **$[\alpha]_D^{25}$** = - 0.5 (c=0.54, $CHCl_3$).

Synthesis of branched *syn*-synthon *ent*-87



^{1}H NMR (600 MHz, $CDCl_3$) δ 5.80 (ddd, *J* = 7.2, 10.5, 17.4 Hz, 1H), 5.08-5.04 (m, 2H), 3.66 (t, *J* = 5.7 Hz, 2H), 3.50-3.46 (m, 1H), 2.45 (d, *J* = 4.2, 1H), 2.29-2.26 (m, 1H), 1.68-1.61 (m, 3H), 1.42-1.37 (m, 1H), 1.04 (d, *J*= 6.6 Hz, 3H), 0.90 (s, 9H), 0.07 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 141.4, 115.0, 74.7, 63.6, 43.8, 31.4, 29.5, 26.1, 18.5, 14.9, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{14}H_{31}O_2Si$ ($M+H^+$) 258.2087, found 258.2087; **FT-IR**: 3367, 2954, 2857, 1471, 1253, 1095, 995, 832, 773 cm^{-1} ; **$[\alpha]_D^{25}$** = - 17.4 (c=1.14, $CHCl_3$).

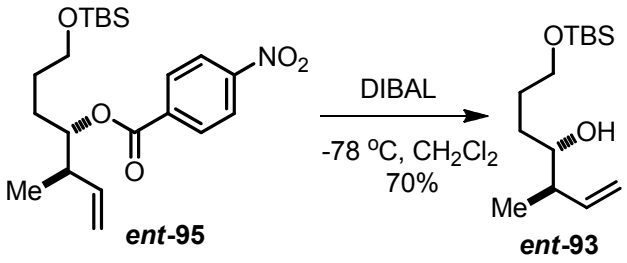
Synthesis of Mitsunobu inversion ester *ent*-95



^{1}H NMR (600 MHz, $CDCl_3$) δ 8.29 (d, *J* = 8.4 Hz, 2H), 8.20 (d, *J* = 9.6 Hz, 2H), 5.80 (ddd, *J* = 7.8, 10.2, 17.7 Hz, 1H), 5.15 (ddd, *J* = 4.8, 9.0 Hz, 1H), 5.08 (dd, *J* = 16.8, 18.3 Hz, 2H), 3.65-3.59 (m, 2H), 2.59-2.53 (m, 1H), 1.81-1.70 (m, 2H), 1.60-1.51 (m, 2H), 1.08 (d, *J* = 6.6 Hz, 3H), 0.08 (s, 9H), 0.03 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 164.5, 150.6, 139.2, 136.2, 130.8, 123.7, 116.2, 78.8, 62.7, 42.0, 28.9, 28.0, 26.1, 18.5, 16.3, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{21}H_{34}NO_5Si$

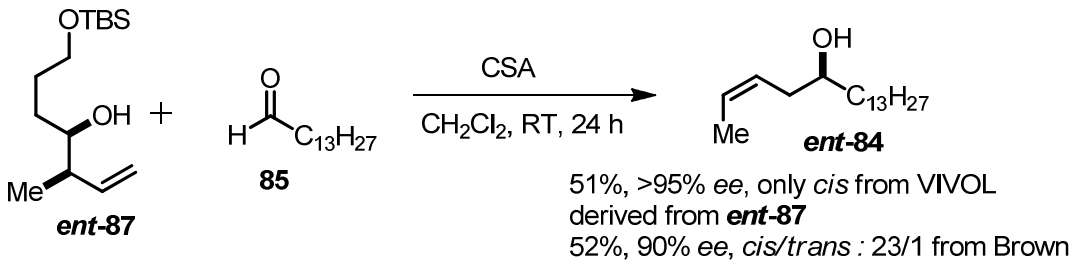
(M+H$^+$) 408.2200, found 408.2202; **FT-IR**: 2954, 2856, 1721, 1528, 1318, 1270, 1098, 872, 831, 717 cm^{-1} ; **[α]$_D^{25}$** = +9.7 (c=0.875, $CHCl_3$).

Synthesis of *anti*-synthon *ent*-93



^{1}H NMR (600 MHz, $CDCl_3$) δ 5.79 (ddd, *J* = 9.0, 19.2 Hz, 1H), 5.10-5.07 (m, 2H), 3.69-3.63 (m, 2H), 3.45-3.43 (m, 1H), 2.38 (d, *J* = 3.6 Hz, 1H), 2.26-2.20 (m, 1H), 1.68-1.62 (m, 3H), 1.44-1.38 (m, 1H), 1.04 (d, *J* = 6.6 Hz, 3H), 0.90 (s, 9H), 0.06 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 140.8, 115.9, 74.7, 63.6, 44.2, 31.2, 29.3, 26.1, 18.5, 16.2, -5.1; **HRMS (ESI)**: *m/z* calcd. for $C_{14}H_{31}O_2Si$ (M+H$^+$) 258.2087, found 258.2086; **FT-IR**: 3399, 2954, 2856, 1471, 1253, 1094, 832, 773 cm^{-1} ; **[α]$_D^{25}$** = + 0.8 (c=0.625, $CHCl_3$).

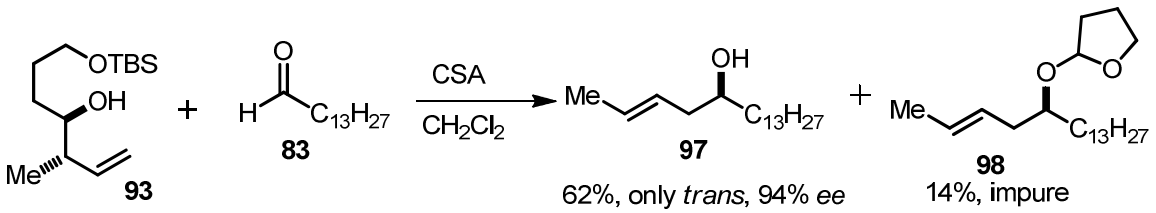
Synthesis of *cis*-homoallylic alcohol *ent*-84



To a solution of the *syn*-synthon ***ent*-87** (0.34 g, 1.29 mmol) and tetradecanal **85** (0.25 g, 1.17 mmol) in CH_2Cl_2 (8 mL) at room temperature was added camphorsulphonic acid (0.33 g, 1.4 mmol). (TLC after 30 min showed the absence of the aldehyde and the formation of the 7-membered ring acetal as

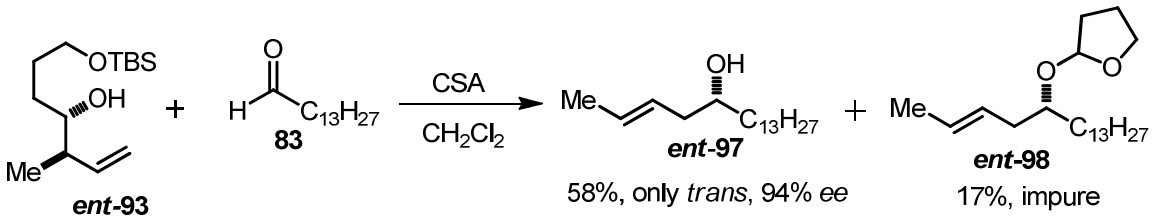
indicated by crude ^{1}H NMR). Stirred the reaction mixture for 24 h (a crude NMR at this point showed the absence of the 7-membered ring acetal compound) before adding MeOH. Stirred for 1h and removed the solvent under vacuum. Purified the crude by flash chromatography using 2-3% ethyl acetate and hexanes to obtain *cis*-homoallylic alcohol ***ent*-84** (0.16 g, 52%) as a white solid. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.68-5.62 (m, 1H), 5.46-5.42 (m, 1H), 3.64-3.62 (m, 1H), 2.22 (t, *J* = 6.6 Hz, 2H), 1.64 (dd, *J* = 1.2, 6.6 Hz, 3H), 1.58 (s,1H), 1.49-1.44 (m, 3H), 1.31-1.26 (m, 21H), 0.88 (t, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 127.4, 126.4, 71.7, 37.0, 35.1, 32.1, 29.9, 29.8, 29.5, 25.9, 22.9, 14.3, 13.2 ; **HRMS (APCI)**: *m/z* calcd. for $C_{18}H_{35}O$ (M^+) 267.2682 found 267.2683; **FT-IR**: 3345, 3021, 2915, 2848, 1469 cm^{-1} ; **Mp**: 28-29 °C; **$[\alpha]_D^{25}$** = - 2.7 (c=1.03, $CHCl_3$).

Synthesis of *trans*-homoallylic alcohol 97



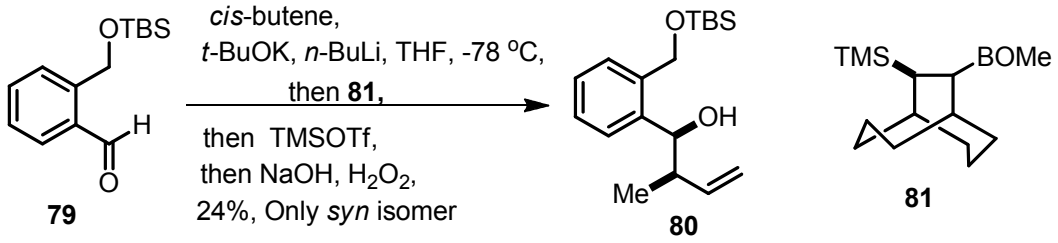
Procedure same as *cis*-homoallylic alcohol ***ent*-84**. *anti*-synthon **93** (0.24 g, 0.93 mmol), tetradecanal **83** (0.18 g, 0.84 mmol) in CH_2Cl_2 (5.5 mL) and camphorsulphonic acid (0.24 g, 1.01 mmol). Obtained *trans*-homoallylic alcohol **97** (0.14 g, 62%) as a white solid. For spectral details reference 31.

Synthesis of *trans*-homoallylic alcohol *ent*-97



Procedure same as *cis*-homoallylic alcohol ***ent*-84**. *anti*-synthon ***ent*-93** (0.20 g, 0.77 mmol), tetradecanal **83** (0.15 g, 0.70 mmol) in CH_2Cl_2 (4.5 mL) and camphorsulphonic acid (0.20 g, 0.84 mmol). Obtained *trans*-homoallylic alcohol ***ent*-97** (0.11 g, 58%) as a white solid. For spectral details reference 31.

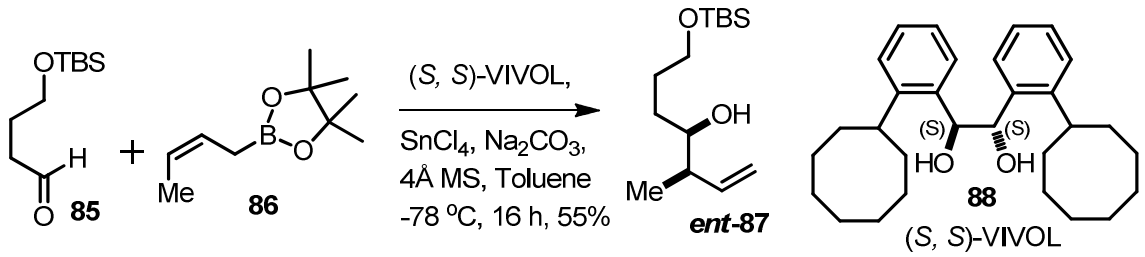
Synthesis of synthon 80 using Soderquist method



cis-2-butene (0.64 g, 11.4 mmol) was added to a cooled solution of *t*-BuOK (0.61 g, 5.49 mmol) in THF (3.0 mL) at -78 ^{o}C. *n*-BuLi (0.35 g, 5.49 mmol, 2.5M in hexanes) was then added and the yellow reaction mixture warmed to -78 ^{o}C and stirred for 1.5 h. Cannulated this solution to a cooled solution of the Soderquist reagent **81** (1.3 g, 5.49 mmol) in ether (54 mL) at -78 ^{o}C and stirred for 30 min. TMSOTf (1.73 g, 7.81 mmol) was then added and stirred for 15 min before adding the aldehyde **79** (1.1 g, 4.39 mmol). After stirring for 3 h at -78 ^{o}C removed the solvents under vacuum. Acetronitrile (8.7 mL) was then added followed by (*1R, 2R*)-pseudoephedrine (0.9 g, 5.49 mmol) and the reaction heated to reflux for 2 h. After cooling the reaction to room temperature, filtered

out the solid and evaporated the filtrate to obtain the crude as a semisolid. Purified the crude by flash chromatography using 5% ethyl acetate and hexanes to obtain synthon **80** as colorless oil (0.32 g, 24%). Recovered also the unreacted aldehyde **79** (0.63 g, 57%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.46 (dd, *J* = 1.6, 8.0 Hz, 1H), 7.36 (dd, *J* = 2.0, 7.2 Hz, 1H), 7.33-7.24 (m, 2H), 5.73 (ddd, *J* = 6.8, 17.4 Hz, 1H), 5.05-5.00 (m, 1H), 4.98 (d, *J* = 11.2 Hz), 4.82 (d, *J* = 12.4 Hz, 1H), 4.73 (d, *J* = 12.4 Hz, 1H). 4.72 (d, *J* = 4.8 Hz, 1H), 2.74-2.66 (m, 1H), 1.16 (d, *J* = 6.8 Hz, 3H), 0.93 (s, 9H), 0.12 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 141.0, 141.0, 137.8, 128.3, 127.7, 127.5, 127.3, 115.0, 74.3, 63.9, 43.2, 26.1, 18.5, 14.9, -5.0.

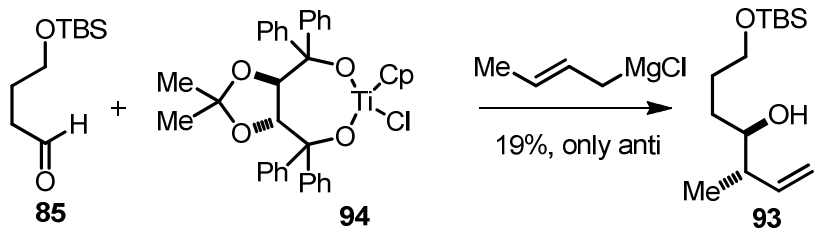
Synthesis of synthon *ent*-87 using Dennis Hall VIVOL catalyst 88



To preactivated 4Å MS (0.11 g) was added (*S*, *S*)-VIVOL catalyst **88** (0.028 g, 0.064 mmol), anhydrous Na_2CO_3 (0.01 g, 0.049 mmol) and toluene (2.0 mL). After stirring for 5 min at room temperature added $SnCl_4$ (0.012 g, 0.049 mmol) and cooled the reaction to -75 °C and stirred for 15 min. *cis*-crotyl boronic acid pinacol ester (0.1 g, 0.54 mmol) was then added and stirrred for 30 min before adding the aldehyde **85** (0.1 g, 0.49 mmol). The reaction mixture was stirred at -67 °C for 19 h before quenching with DIBAL (0.9 mL, 1.0 M in toluene). After 15 min added 1.0 M HCl (1.8 mL) and stirred at room temperature for 1 h. The aqueous layer was extracted with hexanes, dried over $MgSO_4$, filtered and

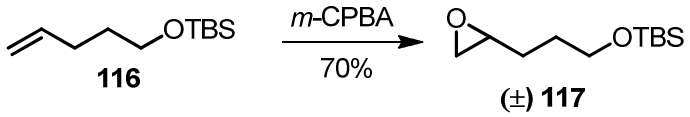
concentrated to obtain oil. Purified the crude by flash chromatography using 6% ethyl acetate and hexanes to obtain synthon ***ent*-87** as colorless oil (0.07 g, 55%). Spectral data similar to compound **87**.

Synthesis of *anti*-synthon 93 using Hafner reagent 94



To a solution of **(*R, R*)-94** (0.21 g, 0.34 mmol) in ether (2.5 mL) at 0 ^{o}C was added 2-butenylmagnesium chloride (0.032 g, 0.28 mmol, 0.5 M in THF) and stirred for 2.5 h. Cooled to -78 ^{o}C and added a solution of the aldehyde **85** (0.05 g, 0.24 mmol) in ether (0.37 mL) and stirred for 17 h at -65 ^{o}C. Deionised water (0.55 mL) was then added and the reaction mixture stirred at room temperature for 3 days. After passing the reaction through a bed of celite extracted with ether, dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 5% ethyl acetate and hexanes to obtain *anti*-synthon **93** as colorless oil (0.012 g, 19%). Spectral data similar to compound **93**.

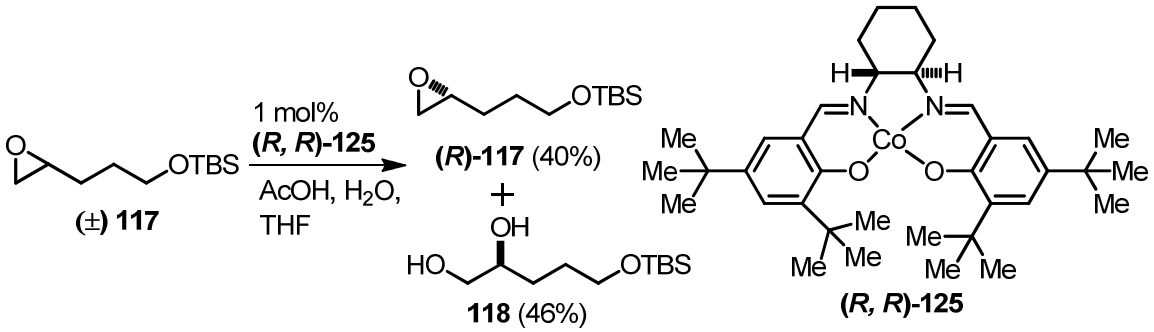
Synthesis of racemic epoxide 117



To a cooled solution of the alkene **116** (5.0 g, 24.9 mmol) in CH_2Cl_2 (80 mL) at 0 ^{o}C was added a solution of *m*-CPBA (6.5 g, 37.5 mmol) in CH_2Cl_2 (20 mL) and stirred for 4 h. The reaction was quenched with sat. $NaHCO_3$ and the aqueous extracted with CH_2Cl_2. The organic layer was then washed with brine,

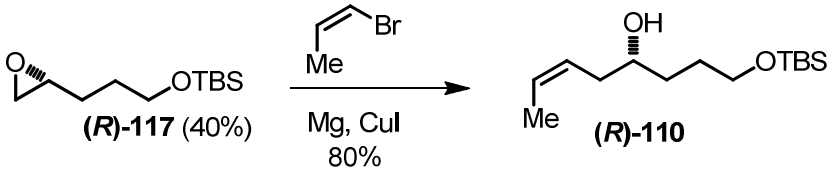
dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 5% ethyl acetate and hexanes to obtain **117** as colorless oil (3.8 g, 70%). Spectral data similar to **(*R*)-117**.

Kinetic resolution of racemic epoxide 117 using Jacobsen HKR



To a soluion of (*R*, *R*)-Jacobsen catalyst **125** (0.028 g, 0.046 mmol) and epoxide ***rac*-117** (1.0 g, 4.62 mmol) in THF (1.0 mL) was added acetic acid (0.0055 g, 0.092 mmol) at room temperature. After cooling to 0 ^{o}C added water (0.046 g, 2.54 mmol) and then stirred the reaction at room temperature for 22 h. Loaded the reaction mixture directly onto the column for purification and purified using 6-8% ethyl acetate and hexanes for the epoxide and 70% for diol to obtain the epoxide **(*R*)-117** (0.4 g, 40%) and diol **118** (0.5 g, 46 %) as colorless oil. **Epoxide:** ^{1}**H NMR** (600 MHz, $CDCl_3$) δ 3.69-3.61 (m, 2H), 2.95-2.92 (m, 1H), 2.75 (app t, *J* = 4.2 Hz, 1H), 2.47 (dd, *J* = 4.8 Hz, 1H), 1.73-1.54 (m, 4H), 0.89 (s, 9H), 0.04 (s, 6H); ^{13}**C NMR** (150 MHz, $CDCl_3$) δ 62.8, 52.3, 47.3, 29.3, 29.2, 26.1, 18.5, -5.1; **Diol 118** : ^{1}**H NMR** (600 MHz, $CDCl_3$) δ 3.79 (bs, 1H), 3.70-3.59 (m, 4H), 3.45-3.41 (m, 1H), 2.91 (bs, 1H), 1.68-1.62 (m, 2H), 1.61-1.55 (m, 1H), 1.49-1.42 (m, 1H), 0.89 (s, 9H), 0.05 (s, 6H); ^{13}**C NMR** (150 MHz, $CDCl_3$) δ 72.1, 66.9, 63.6, 30.8, 29.2, 26.0, 18.4, -5.2.

Synthesis of linear homoallylic alcohol (*R*)-110



To a round bottom flask containing a solution of Mg turnings (0.56 g, 23.2 mmol) and a crystal of iodine in THF (10.0 mL), attached a cold finger condensor and cooled to -78 ^{o}C. *Cis*-1-bromo-propene (1.2 g, 9.98 mmol) was then added and the reaction stirred at room temperature for 1 h. This solution was then cannulated into a suspension of CuI (0.41 g, 2.16 mmol) in ether (53 mL) at -78 ^{o}C. After stirring for 30 min epoxide **(*R*)-117** (0.36 g, 1.66 mmol) was added and the reaction allowed to warm to room temperature and stirred overnight. The reaction was diluted with saturated NH_4Cl and extracted the aqueous with ether, dried over $MgSO_4$, filtered and concentrated to obtain oil. Purified the crude by flash chromatography using 8% ethyl acetate and hexanes to obtain **(*R*)-110** as oil (0.33 g, 80%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.64-5.59 (m, 1H), 5.47-5.42 (m, 1H), 3.70-3.62 (m, 3H), 2.62 (d, *J* = 3.6 Hz, 1H), 2.29-2.19 (m, 2H), 1.69-1.63 (m, 7H), 0.90 (s, 9H), 0.07 (s, 6H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 126.9, 126.6, 71.5, 63.7, 35.1, 34.1, 29.4, 26.1, 18.5, 13.2, -5.1.

Chapter 4

Chapter 4

Human Milk Oligosaccharides: Synthesis of natural and unnatural oligomers

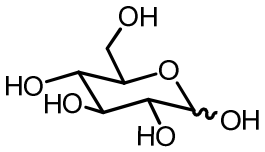
4.1. Introduction and background

One of the major classes of biological molecules that are essential for life is carbohydrates.[115] In mammalian cells there are nine common monosaccharide units present, which form a wide variety of structurally complex oligomers. Carbohydrates were once considered to be nutritional molecules whose main function was as energy source, without any biological function. Recent developments in glycomics and improved synthetic protocols for oligosaccharide synthesis have painted a new picture on the biological role of carbohydrates.[116]

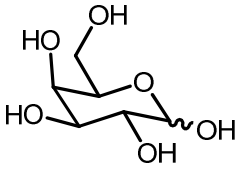
One of the areas in carbohydrates that has seen less development is the human milk oligosaccharide (HMO). Besides lactose and lipids, HMO's represent the third component of human milk.[117] Over 200 different complex mixtures of oligosaccharides have been identified so far in human milk.[117] This complexity is achieved from a combination of five different monosaccharide, (a) Glucose (Glc) **(1)**; (b) galactose (Gal) **(2)**; (c) sialic acid (Neu5Ac) **(3)**; (d) fucose (Fuc) **(4)**; and (e) *N*-acetylglucoseamine (GlcNAc) **(5)** which are present in human milk (figure 1).[118] A liter of milk contains about 5-10 g of unbound oligosaccharide. The HMO's range from three to thirty two monosaccharide residues in length with lactose usually occupying the reducing end. The composition or concentration of

these oligosaccharides are highest after birth and decrease during the first 3 months after giving birth.

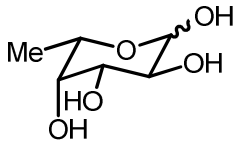
Figure 1. The five monosaccharides present in human milk



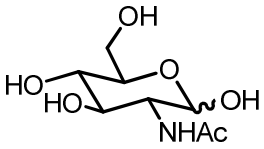
Glucose **(Glc) (1)**



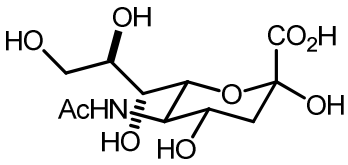
Galactose **(Gal) (2)**



Fucose **(Fuc) (3)**



N-Acetylglucosamine **(GlcNAc) (4)**

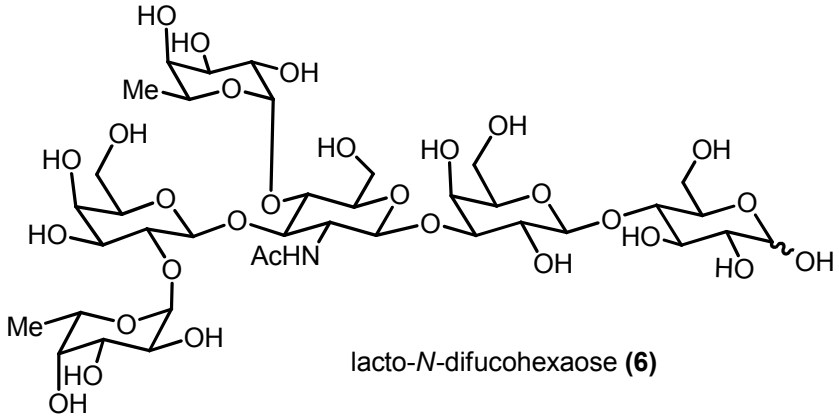


Sialic acid (*N*-Acetyl neuraminic acid, **Neu5Ac (5)**

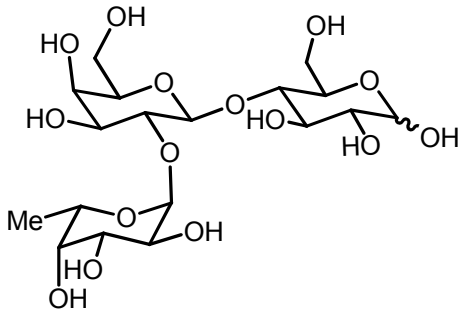
HMO's are manufactured in the breast in the Golgi apparatus in cells lining the alveoli and ductules only during lactation. The initial step is the synthesis of lactose in a reaction between UDP galactose and glucose using the enzyme galactotransferase.[119] Lactose then acts as a building block to which other transferases like fucosyl, sialyl, *N*-acetylglucosaminyl add monosaccharides to form complex HMO's. The core molecule is characterized by the repetitive addition of galactose or *N*-acetylglucosamine in β-glycosidic linkages to lactose. This in itself creates a lot of complexity which is further amplified by the attachment of α-glycosidic linkages of fucose or sialic acid to the core molecules.[120] The attachment of fucose is based on the Lewis blood group of the individual mother.

Out of the many HMO's, the major components are monofucosyllactose **(7)**, **(9)**, difucosyllactose lacto-*N*-hexaoses **(6)** and lacto-*N*-pentaoses **(8)** (figure 2).[118]

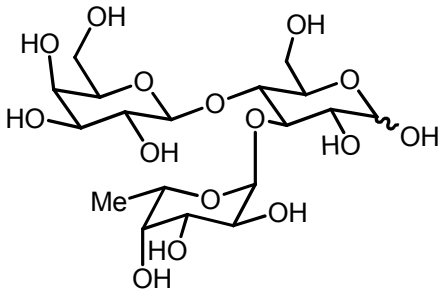
Figure 2. Some representative HMO's



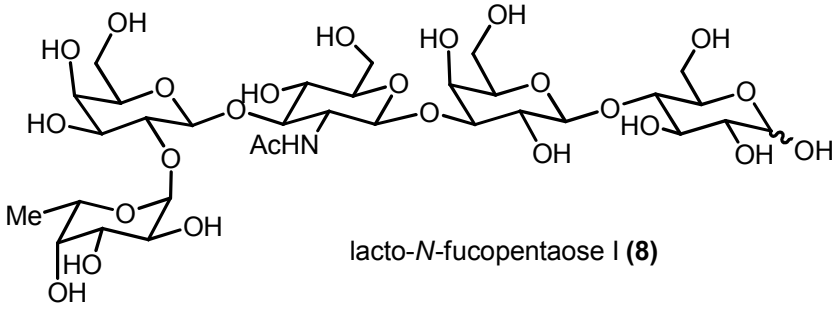
lacto-*N*-difucohexaose **(6)**



2'-fucosyllactose **(7)**



3-fucosyllactose **(9)**



lacto-*N*-fucopentaose I **(8)**

The oligosaccharide content of human milk is higher than that of bovine milk. Table 1 lists the amount and composition of oligosaccharides present in human milk in comparison to cow's milk.[118]

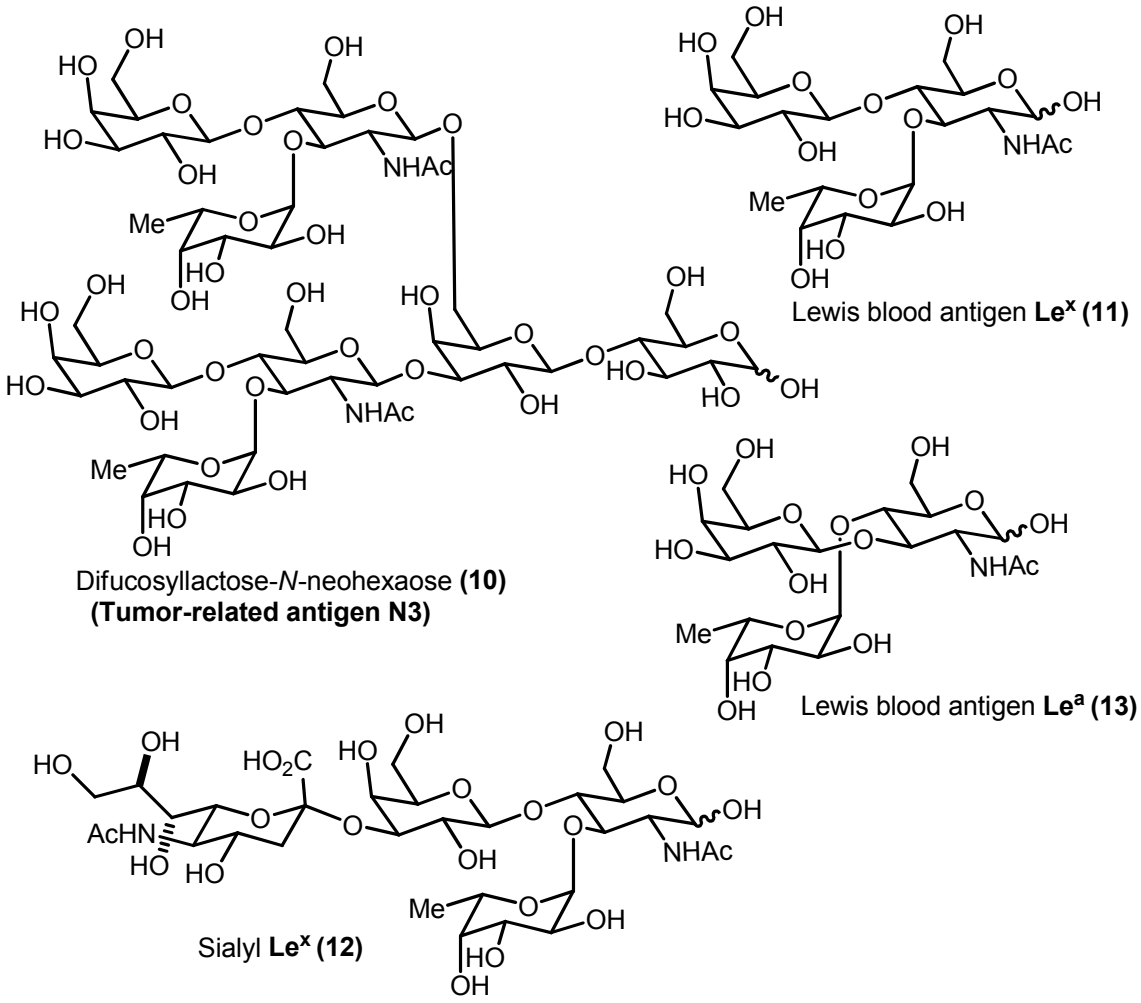
Table 1. Oligosaccharides in human and cow's milk

Components	Amount (g/liter)	
	Human milk	Cow's milk
Lactose	55-70	40-50
Oligosaccharides		
Lacto-*N*-tetraose	0.5-1.5	traces
Lacto-*N*-fucopentaose I	1.2-1.7	
Lacto-*N*-fucopentaose II	0.3-1.0	
Lacto-*N*-fucopentaose III	0.01-0.2	
Lacto-*N*-difucohexaose I	0.1-0.2	
NeuAc (α 2-6) lactose	0.3-0.5	0.03-0.06
NeuAc (α 2-3) lactose	0.1-0.3	
NeuAc-lacto-*N*-tetraose a	0.03-0.2	traces
NeuAc-lacto-*N*-tetraose c	0.1-0.6	traces
$NeuAc_2$-lacto-*N*-tetraose	0.2-0.6	traces
Oligosaccharides (total)	5.0-8.0	traces

The composition of HMO's depends largely on the blood type of the mother. They can be classified into three groups according to the Lewis acid blood groups Le(a-b+), Le(a+b-) and Le(a-b-).[121] Based on the presence or absence of silalic acid, HMO's are classified as acidic or neutral. The structures

of about 93 neutral or acidic HMO's have been identified. Some of the HMO's are similar to tumor related antigens and Lewis blood group antigens (figure 3).[122]

Figure 3. Some representative Lewis blood antigens



4.2. Biological significance of HMO's

The physiological effects of HMO's may be local or systemic and may be classified as prebiotic, anti-adhesive, glycome-modifying, immunomodulatory and other unknowns (table 2).[118]

Table 2: Postulated HMO effects

Postulated effects	Affected organs
Prebiotic	Colon
Anti-adhesive	Laryngopharynx, Stomach, Small intestine, Colon, Urinary tract
Glycome-modifying	Intestine
Immunomodulatory	Inflamed tissues, Immune system

Listed below are the local effects of HMO's

1. Prebiotic effects:

HMO's are delivered intact into the colon and thus form a main source of energy for intestinal bacteria like *Bifidobacterium bifidum,* which are known to be beneficial to the health of the host.[120] HMO's act as precursors for the biosynthesis of muramic acid, a component of bacterial cell wall. These organisms decrease the intestinal pH by producing lactic acid. These factors inhibit the development of any pathogenic microorganisms, thus protecting the infant from infection.[123]

2. Anti-adhesive effects:

The virulence of pathogens depends on their ability to adhere to host's epithelial surface. HMO's serve as soluble receptor analogues of epithelial cell surface carbohydrates and thus compete with epithelial cells surface for pathogenic bacteria and viruses and thus prevent their adhesion.[120] Infants are

thus protected from infections and diarrhea by HMO's.[124] The incidences of diarrhea in breastfed infants are inversely related to the amount of 2'-fucosyllactose in the mother's milk. Since HMO's bathe the mucosal lining of the nasopharyngeal, gastrointestinal and urinary tracts of infants, they protect them from infection.[125]

3. Glycome-modifying effects:

In vitro studies have suggested that HMO's might modify the epithelial cell surface glycans, the attachment sites of the pathogens and thus regulate the bacteria-host interactions.[120]

Systemic effects:

HMO's are partially absorbed by the intestine and appear in the infant urine and serve as receptor analogs for urinary pathogens. Neutral HMO's inhibit the adhesion to uroepithelial cells of *E. coli* strain isolated from an infant with urinary tract infection.[118] Their presence in urine proves their presence in systemic circulation, though they have not yet been detected in blood. Their presence may thus alter protein carbohydrate interaction on a systemic level. Selectins for example are involved in cell-cell interaction and they bind to fucosylated or sialylated oligosaccharides (sialyl Le^x) on their respective glycoconjugate ligands which resemble some HMO's.[122] Similar is the case with galectins and siglecs. Out of the many HMO's isolated so far half of them are sialylated. Since sialic acids are an integral part of gangliosides, which are present in the plasma membrane of nerve cells, especially in nerve endings and dendrites, HMO's might thus play a vital role in postnatal brain development.[126]

Functional foods and therapeutics:

Some companies are already exploring the possibility of adding these oligosaccharides in infant formulas and food stuffs thus increasing the beneficial health promoting microbes.[119, 123] HMO's are becoming attractive therapeutic agents. Since they are similar to endogenous human oligosaccharide they may not be toxic and cause allergic reactions. They are highly water soluble and stable to heat which also gives them an ability to be sterilized easily. Biological studies are currently underway in this regard.

4.3. Why synthesize these complex HMO's?

HMO's are synthesized exclusively in the mammary gland during lactation, so the study of biosynthetic pathway becomes difficult. HMO's are unique to humans compared to other species of placental mammals and so this limits the use of animal models to mimic absorption, metabolism, pharmacokinetics and function. For further biological evaluation naturally occurring oligosaccharides from the milk of other species could be used, but large scale isolation may not be possible and their effect on human infants remains to be studied. With these limitations it means that a synthetic approach to produce these HMO's seems more viable. The recent developments in automated solid phase oligosaccharide synthesis have been crucial to synthesize HMO's, though a large scale synthesis has not been reported so far.[127]

Some of these HMO's are commercially available from companies like Carbosynth and Dextra but are very expensive.[128] 2'-Fucosyllactose which

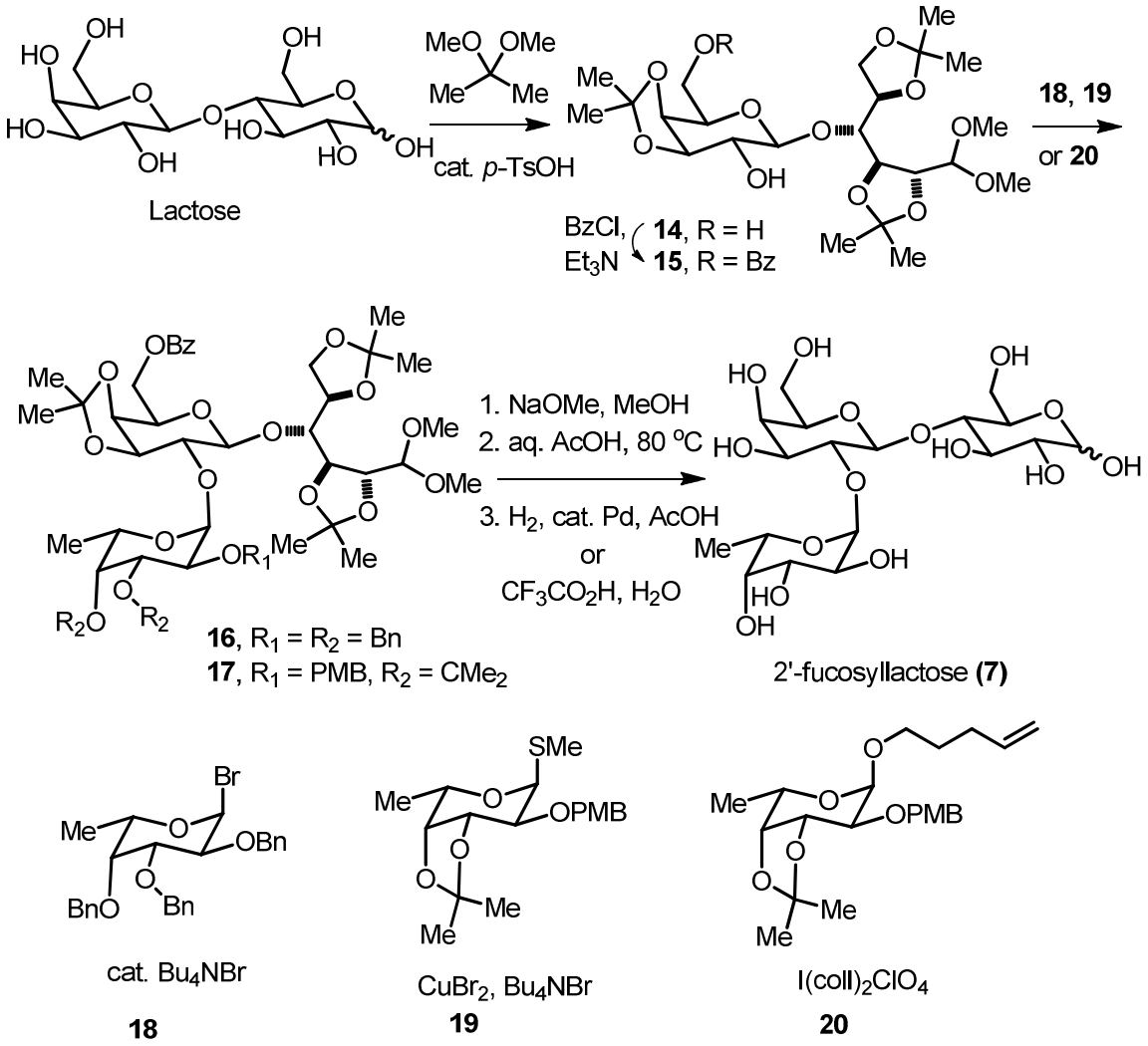
accounts for half or one third of the HMO's costs about $110/mg (Carbosynth), difucosyllacto-*N*-hexaose is $800/mg, Le^x antigen is $110/mg and so are the others. Thus a large scale synthesis would be beneficial not only to make the material available for further biological evaluation but also would substantially lower the cost.

4.4. Synthetic approaches towards HMO's

There have been few synthetic efforts towards the synthesis of HMO's. For a HMO to be viable for human consumption, for biological studies or for process development, the choice of method, reagent and purification must be considered. The synthetic approaches considered thus far have been carried out without any of the above limitations.

The first synthesis of 2'-fucosyllactose **(7)**, a specific ligand for inhibiting the diarrhea-causing bacterium *Campylobacter* was reported by Matta *et al.*[129] Starting from lactose with the tris-acetonide dimethylketal **(15)** as intermediate the 2'-fucosyllactose **(7)** was obtained in 6 steps (scheme 1). Although it is a short synthesis, the issues encountered are the stability of donor **18**, the odor and toxicity of methanediol used for the synthesis of donor **19** and lastly the corrosive and expensive silver nitrate that was used for the preparation of the glycosyl promoter $I(coll)_2ClO_4$.[130]

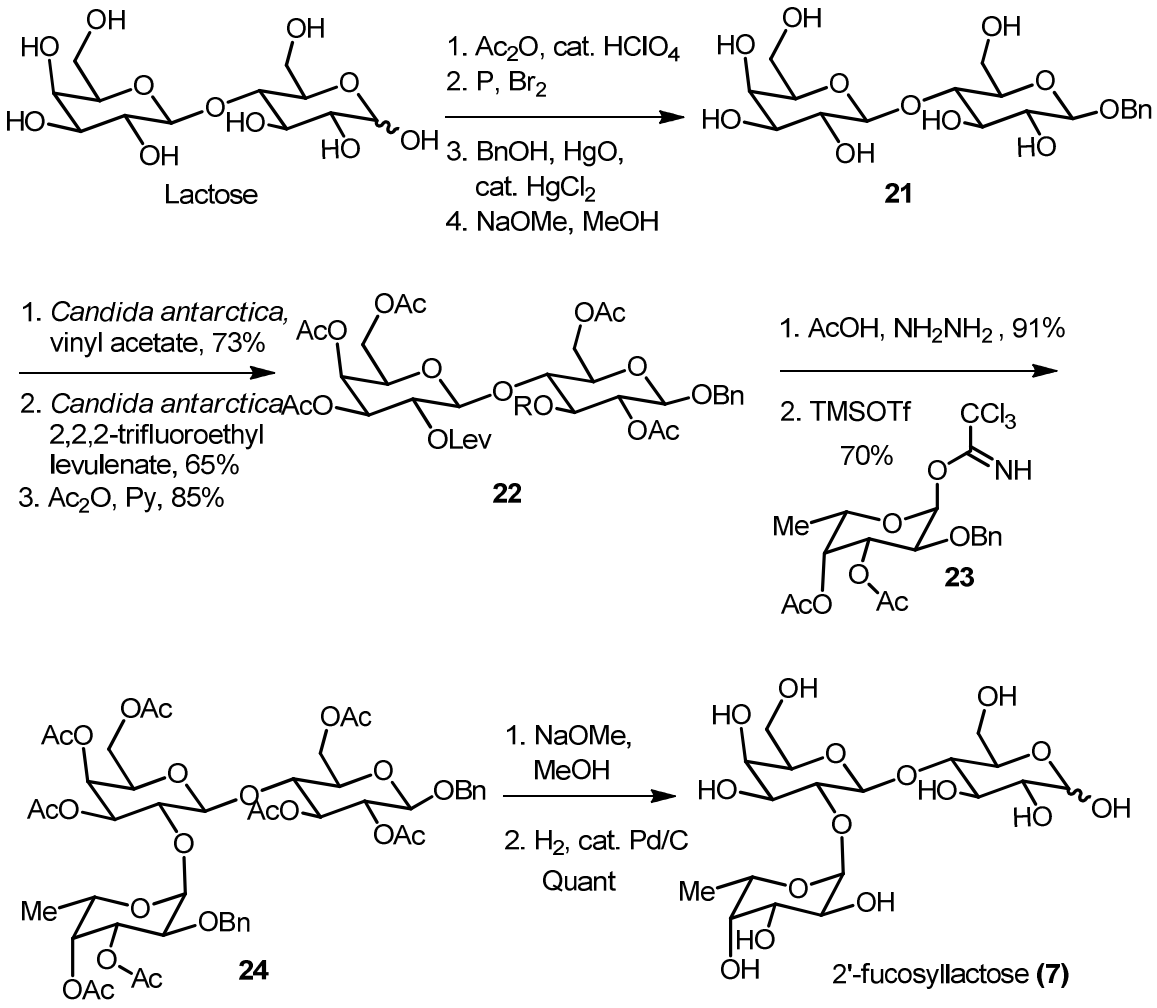
Scheme 1. Synthesis of 2'-fucosyllactose **(7)**, Matta's approach



Another method towards the synthesis of 2'-fucosyllactose **(7)** was reported by Lay *et al.* using a chemoenzymatic approach.[131] Lactose was converted to the benzylether intermediate **21** in a 4 step protocol (scheme 2). Selective acylation of the 6' primary hydroxyl group using *Candida antarctica* and vinyl acetate, followed by selective protection of the 2' secondary hydroxyl group using *Candida antarctica* and 2,2,2-trifluoroethyl levulenate gave intermediate **22**. Acylation followed by the selective removal of levulenate ester and glycosylation using the trichloroacetamidate donor **23** and deprotection gave 2'-fucosyllactose

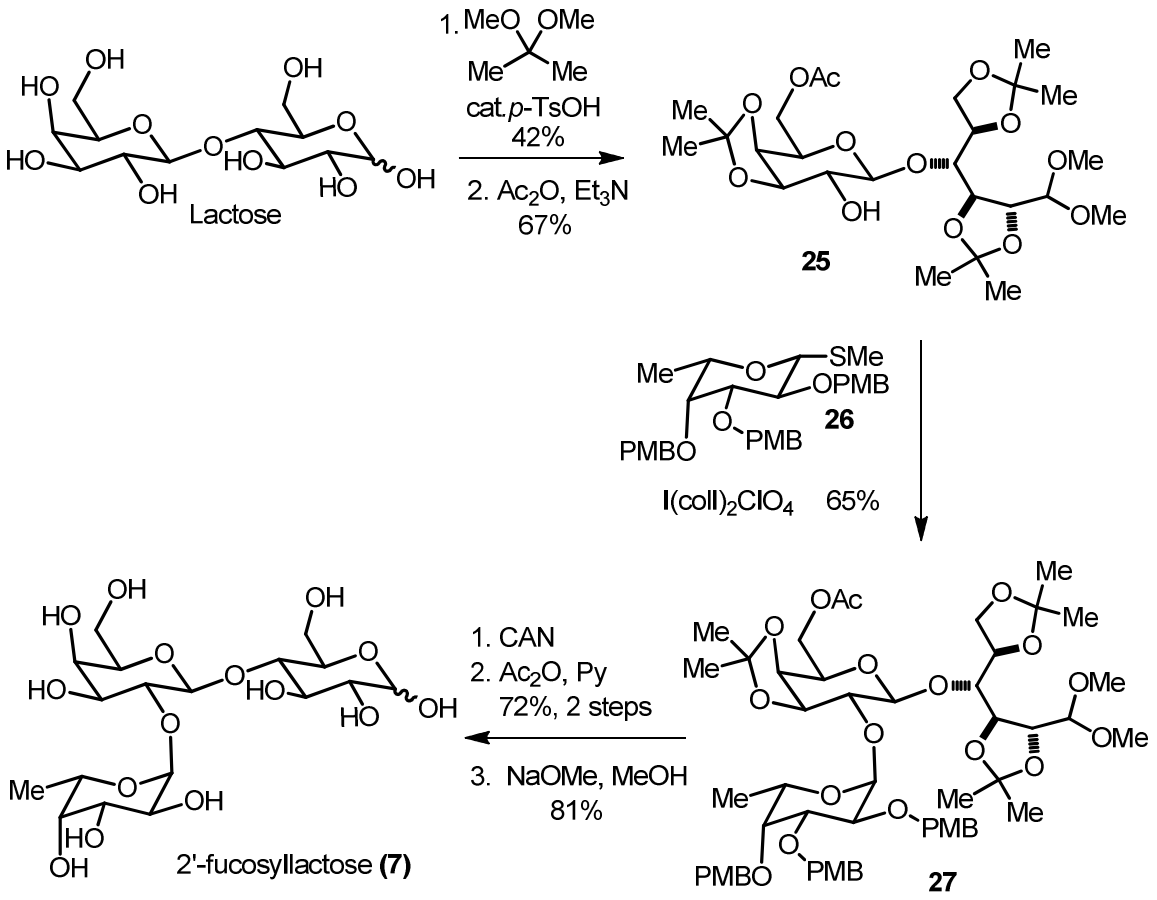
(7). Though it is a streamlined synthesis, the approach used toxic mercuric oxide to promote the glycosylation to prepare **21**, which would be a health hazard.

Scheme 2. Synthesis of 2'-fucosyllactose **(7)**, Lay's chemoenzymatic approach



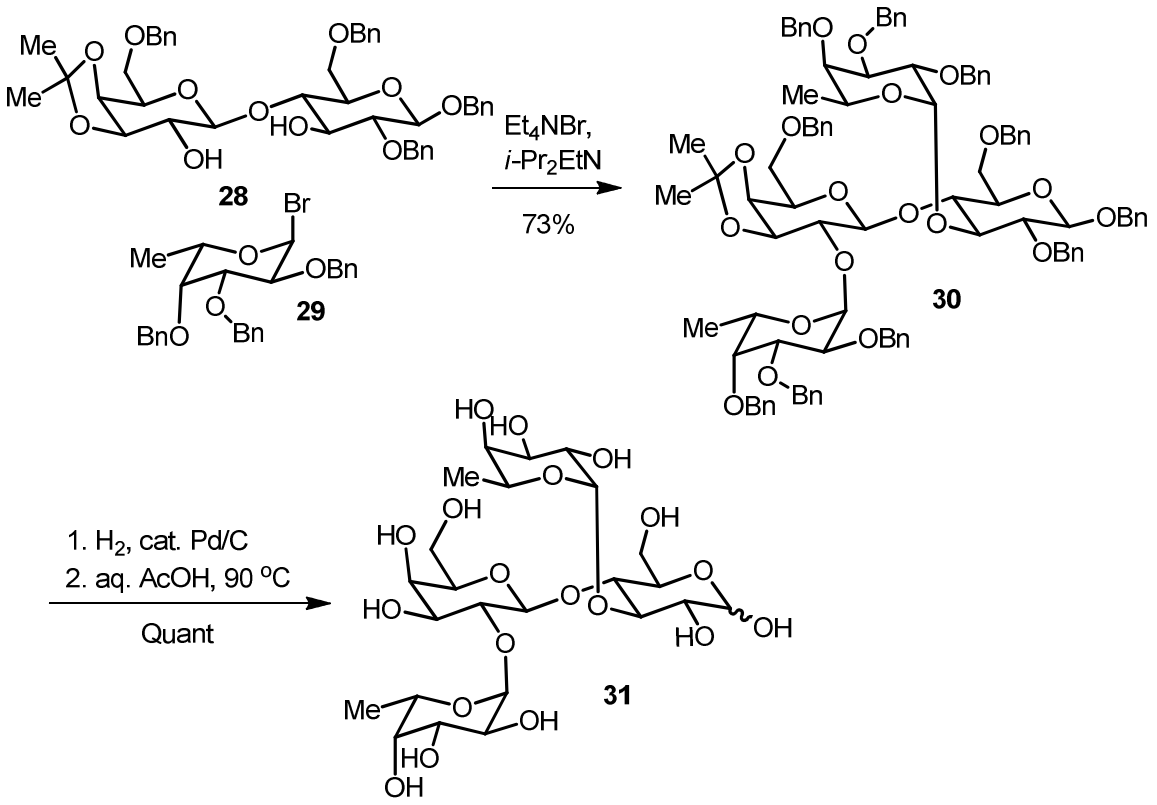
In an approach similar to Matta's, Hashimoto *et al.* used the lactose derived intermediate tris-acetonide dimethylketal **25** and the thioglycoside donor **26** to obtain 2'-fucosyllactose **(7)** in 6 steps (scheme 3).[132] The issues with this synthesis are similar to that of Matta's mentioned earlier.

Scheme 3. Synthesis of 2'-fucosyllactose **(7)**, Hashimoto approach



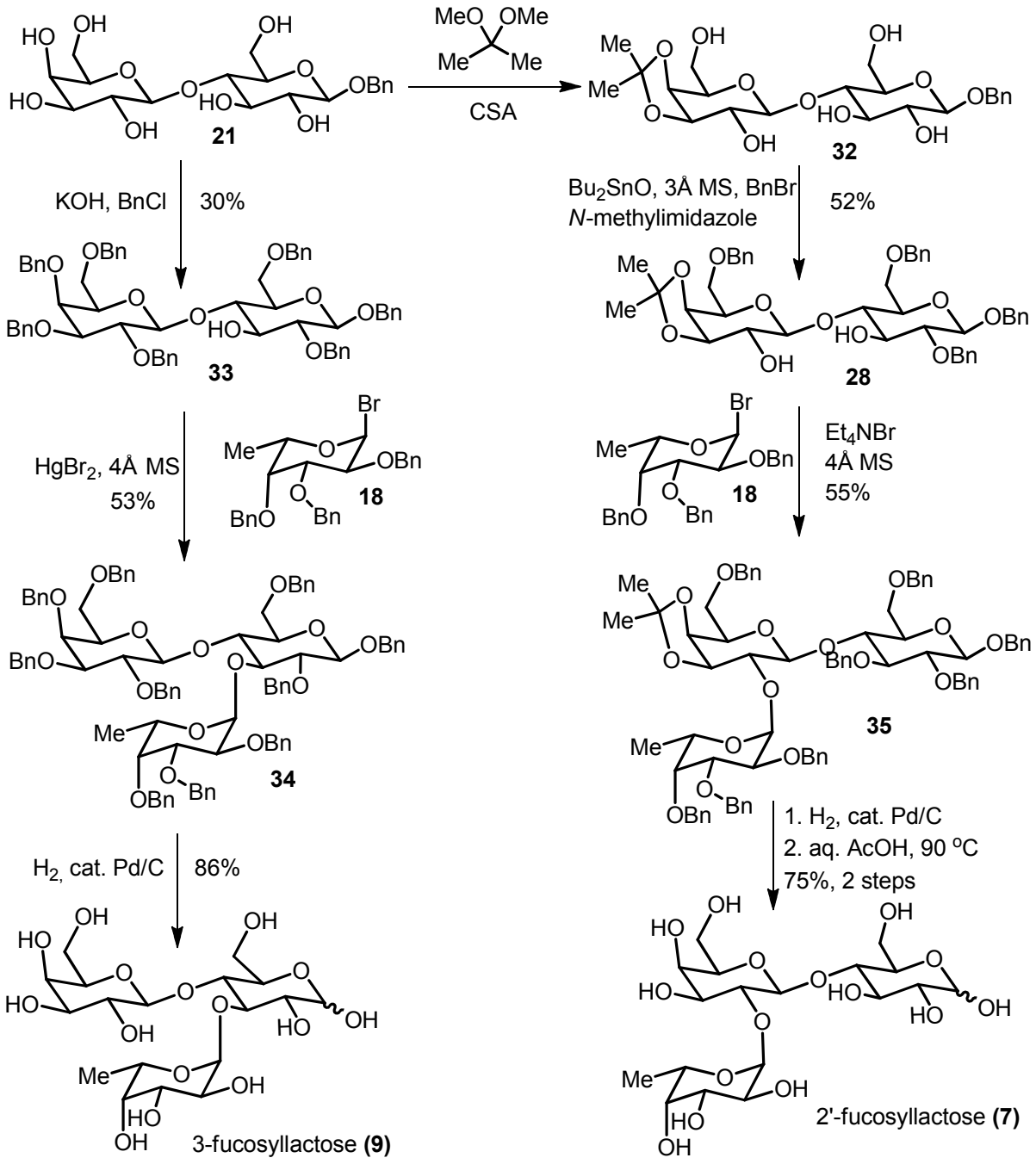
Manuel *et al.* reported the first synthesis of 2',3-difucosyllactose **(31)**.[133] Glycosylation carried out with acceptor **28**, synthesized in 6 steps starting from lactose (scheme 2 and 5) and the bromide donor **29** gave the tetrasaccharide **30**. (scheme 4). A two step deprotection gave 2',3-difucosyllactose **(31)**. The synthesis of 2'-fucosyllactose **(7)** and the first synthesis of 3-fucosyllactose **(9)** was also reported by Manuel *et al.* (scheme 5). [133]

Scheme 4. Synthesis of 2',3-difucosyllactose **(31)**, Manuel approach



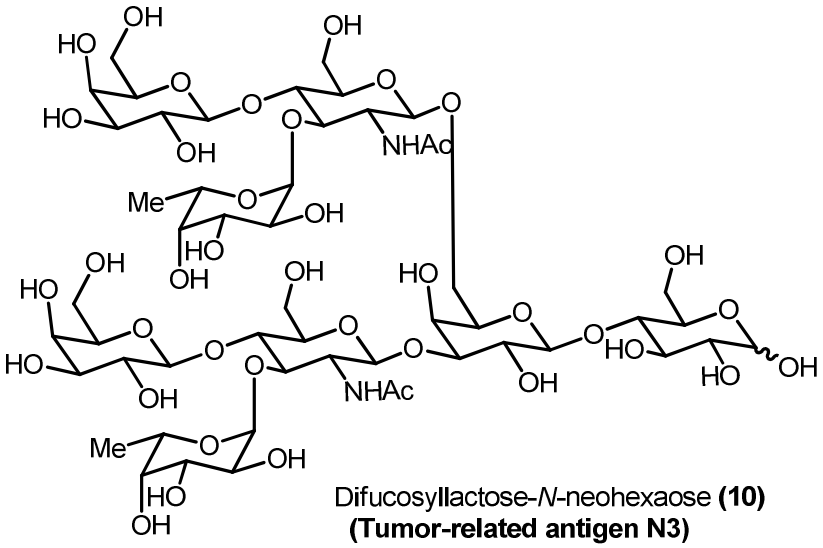
The advantage of this synthesis was the use of a common precursor **21**, which was selectively benzylated to give intermediate **28** and **33** (scheme 5). Glycosylation using the bromide donor **18** gave the trisaccharide **34** and **35** which were then converted to the respective 2'-fucosyllactose **(7)** and 3-fucosyllactose **(9)**. The drawback of the synthesis is the low yields for the selective protection of intermediate **21** in addition to the use of toxic metals like tin and mercury.

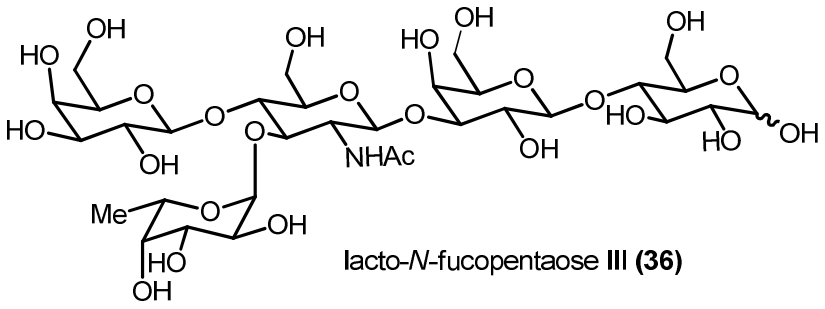
Scheme 5. Synthesis of 2' and 3-fucosyllactose, Manuel approach



The most complex HMO's synthesized thus far has been the lacto-*N*-fucopentaose III **(36)** by Schmidt *et al.*,[134] and the difucosyllacto-*N*-hexaose **(10)** by Danishefsky (figure 4).[135]

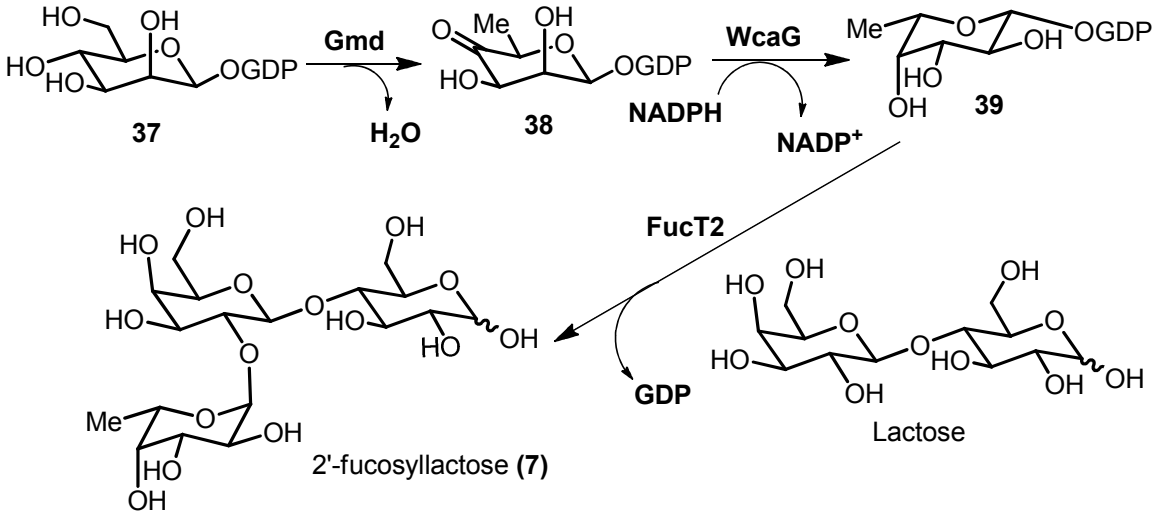
Figure 4. Complex HMO's synthesized thus far





Apart from the chemo and chemoenzymatic approaches, literature also indicates the synthesis of HMO's in a purely enzymatic way.[136] The first reported enzymatic synthesis of 2'-fucosyllactose **(7)** was using recombinant bacterial enzymes.[137] GDP-D-mannose **37** was converted to GDP-4-keto-6-deoxymannose **38** using GDP-mannose-4, 6-dehydratase (Gmd) and cofactor NADP+ (scheme 6). This was then converted to GDP-L-fucose **39** by the bifunctional enzyme GDP-fucose-synthetase in presence of co-substrate NADPH. Fucose **39** was then transferred to the acceptor lactose using the fusion protein GST-FucT2 to obtain 2'-fucosyllactose **(7)**.

Scheme 6. Enzymatic approach towards 2'-fucosyllactose **(7)**



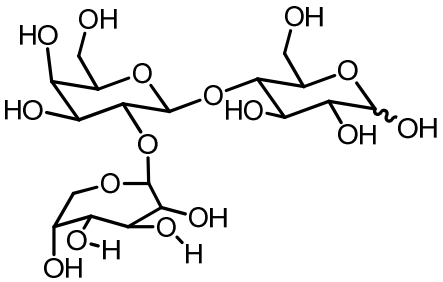
The best approach thus far based on the amount of 2'-fucosyllactose **(7)** produced was reported by Sophie *et al*.[138] Using enzymatic method they were able to produce 11 g/L of 2'-fucosyllactose. With all the above limitation it was necessary to design a synthetic approach which took into account all the drawbacks of the previous synthesis and utilized the recent developments that have taken place in the field of complex oligosaccharide synthesis.

4.5. Synthetic design and evaluation

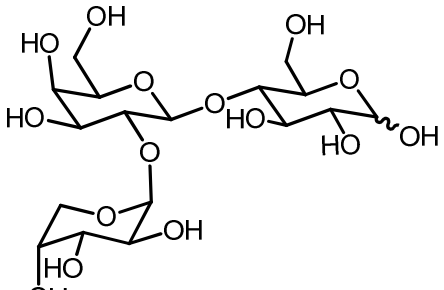
The synthetic challenge in terms of oligosaccharide synthesis is the selective glycosylation to give either α or β isomer and also the selective protection of hydroxyl groups.[115] The outcome depends mostly on the choice of protecting groups used. The design of the synthesis was based mostly upon green chemistry, and so thiols, stannanes, mercury, silver, silyl ethers, and other toxic reagents were out of the question. Initial studies would use common solvents like CH_2Cl_2, $ClCH_2CH_2Cl$, THF, ether and so on which would also be

eventually replaced by greener solvents like water, ionic liquids, trifluorotoluene, 2-methyl-THF, cyclopentyl methyl ether, dimethyl carbonate etc. In addition to this, reducing the number of chromatographic purifications involved in the synthesis might then lead to a viable process which could then be utilized for a process scale synthesis with minor modification. The approach should thus allow putting together some analogues of the type **40** and **41** by using either unnatural sugar during the glycosylation step and then evaluate their biological profile with respect to the natural HMO's (figure 5).

Figure 5. Some proposed unnatural analogs of HMO's



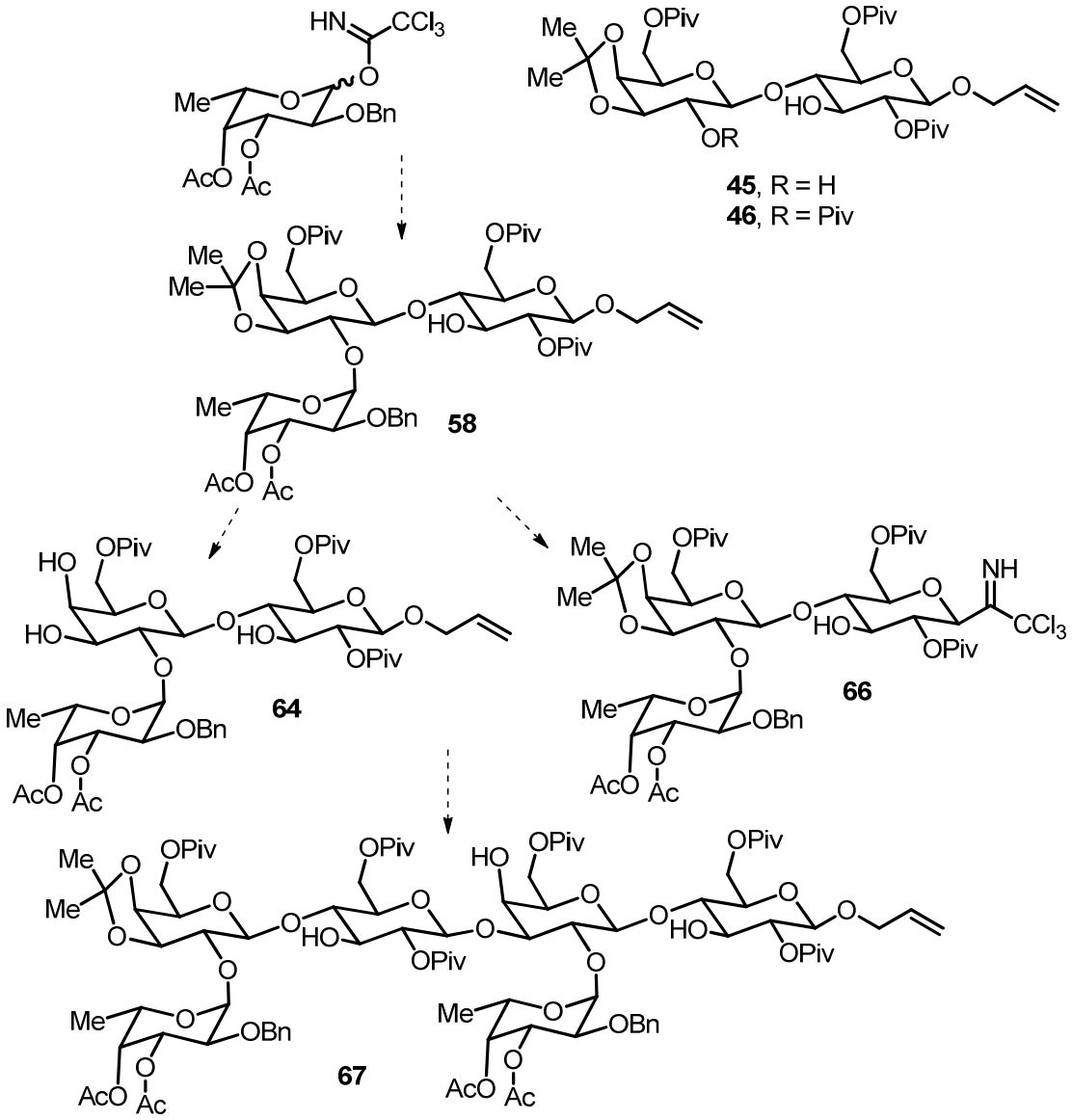
2'-glucoseptanosyllactose **(40)**



2'-arabinolactose **(41)**

The synthesis was envisioned to involve a common acceptor of the type **45** and **46**. Glycosylation of these acceptors with donor **23** would then produce the respective trisaccharide which could be converted to the natural and analog HMO's (scheme 7). From these trisaccharides using some protecting group manipulation one could then produce a donor and acceptor of the type **64** and **65** respectively, which could be used to carry out a modular approach like a 3 + 3 or a 6 + 6 to put together the complex pattern of HMO's.

Scheme 7. Synthetic design to synthesize simple and more complex HMO's



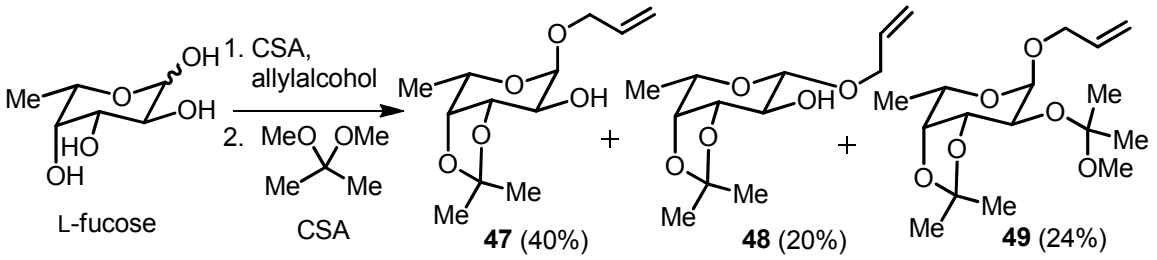
4.6. Results and Discussion

4.6.1. Synthesis of the fucosyl donor 23

The synthesis began from the commercially available L-fucose with the protection of the anomeric hydroxyl group with allyl alcohol to obtain a mixture of both α and β anomer (scheme 8).[139] Subjecting the crude mixture to acetonide

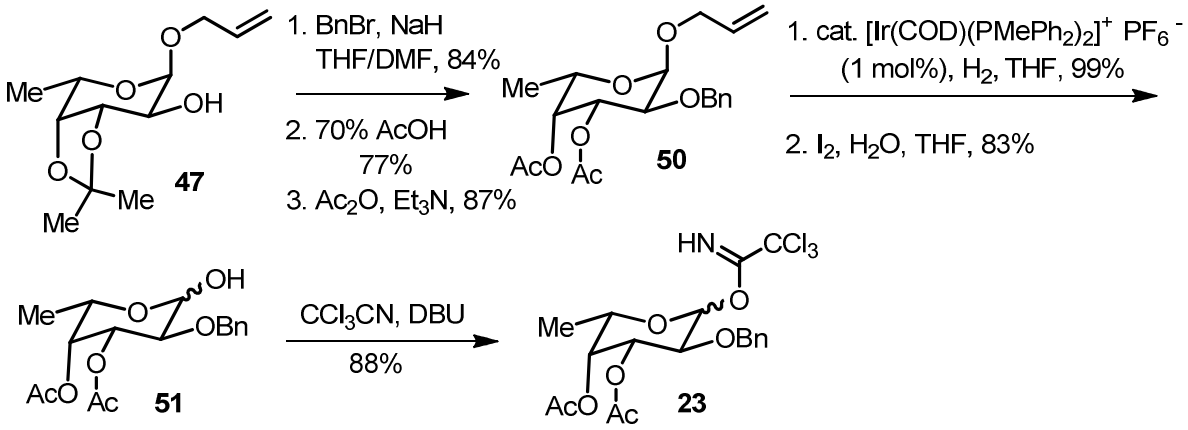
formation and purification resulted in the isolation of the acetonide protected α-anomer **47**, β-anomer **48** and the acetonide-ketal byproduct **49**.

Scheme 8. Synthesis of allylacetonide-L-fucose **47** and **48**



Benzyl protection of the free hydroxyl group followed by acetonide cleavage gave the diol, which was protected as the bisacetate to obtain compound **50** (scheme 9).[139, 140]

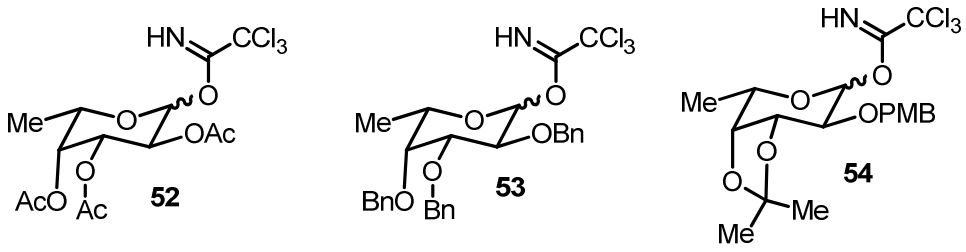
Scheme 9. Synthesis of the trichloroacetamidate donor **23**



Removal of the allyl protecting group by isomerization followed by the clevage of the vinyl ether using iodine generated the free anomeric hydroxyl bearing compound **51** as a mixture of α and β anomer (scheme 9).[141] The free hydroxyl group was then converted to trichloroacetamidate donor **23** using trichloroacetonitrile and DBU.[142] Other fucosyl donors **52**, **53** and **54** with varying

protecting groups were also synthesized to establish good selectivity and yield for the glycosylation step (figure 6).

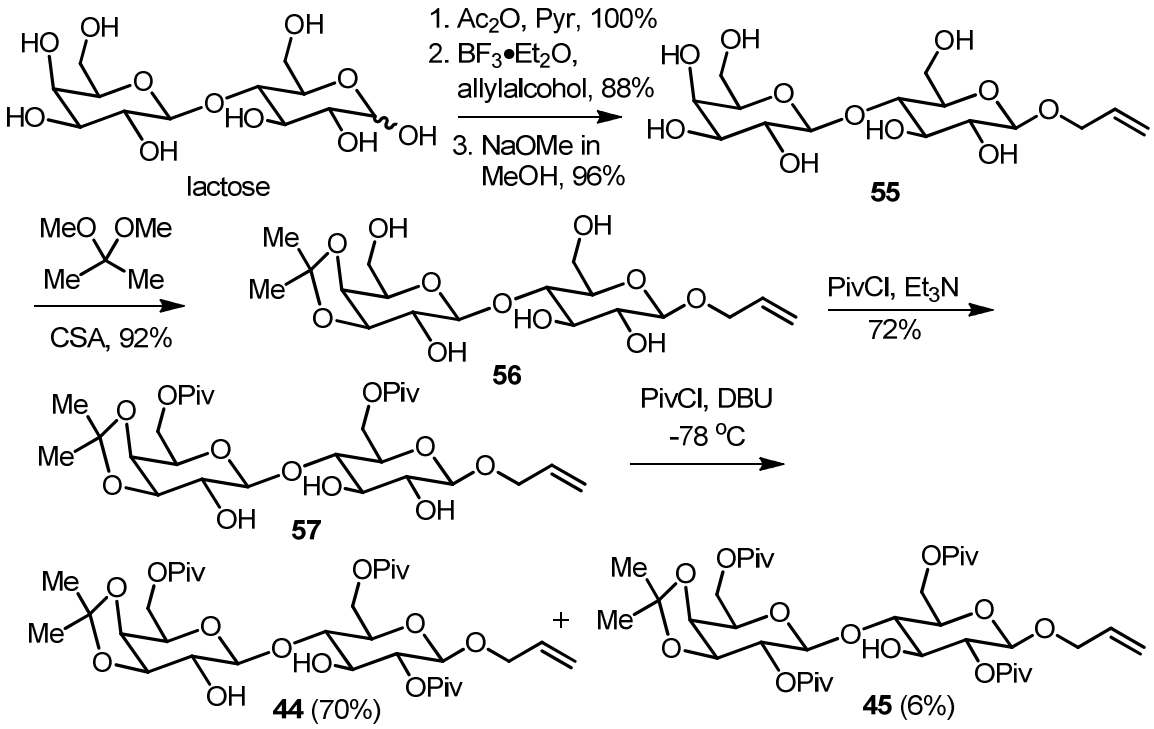
Figure 6. Other fucose donors synthesized for optimization



4.6.2. Synthesis of the lactose acceptor

The synthesis of lactose acceptor began from the commercially available lactose. Our initial approach was to protect the anomeric hydroxyl group as the allylether in a single step which proved to be difficult under varying reaction conditions. Alternatively a three step synthesis from lactose gave the allyl lactose **55** in excellent overall yield (scheme 10).[143] Selective formation of 3', 4' acetonide followed by pivaloation of the primary alcohols gave intermediate **57**.[144] The selective protection of the C2 hydroxyl group using pivaloyl chloride and DBU at -78^{o}C gave the tripivaloate **44** (70%) with some amount of tetrapivaloate **45** (6%) formation.[145] Attempts to carry out the pivaloation in a single step starting from **56** resulted in poor yields of the required compound.

Scheme 10. Synthesis of lactose acceptors **44** and **45**

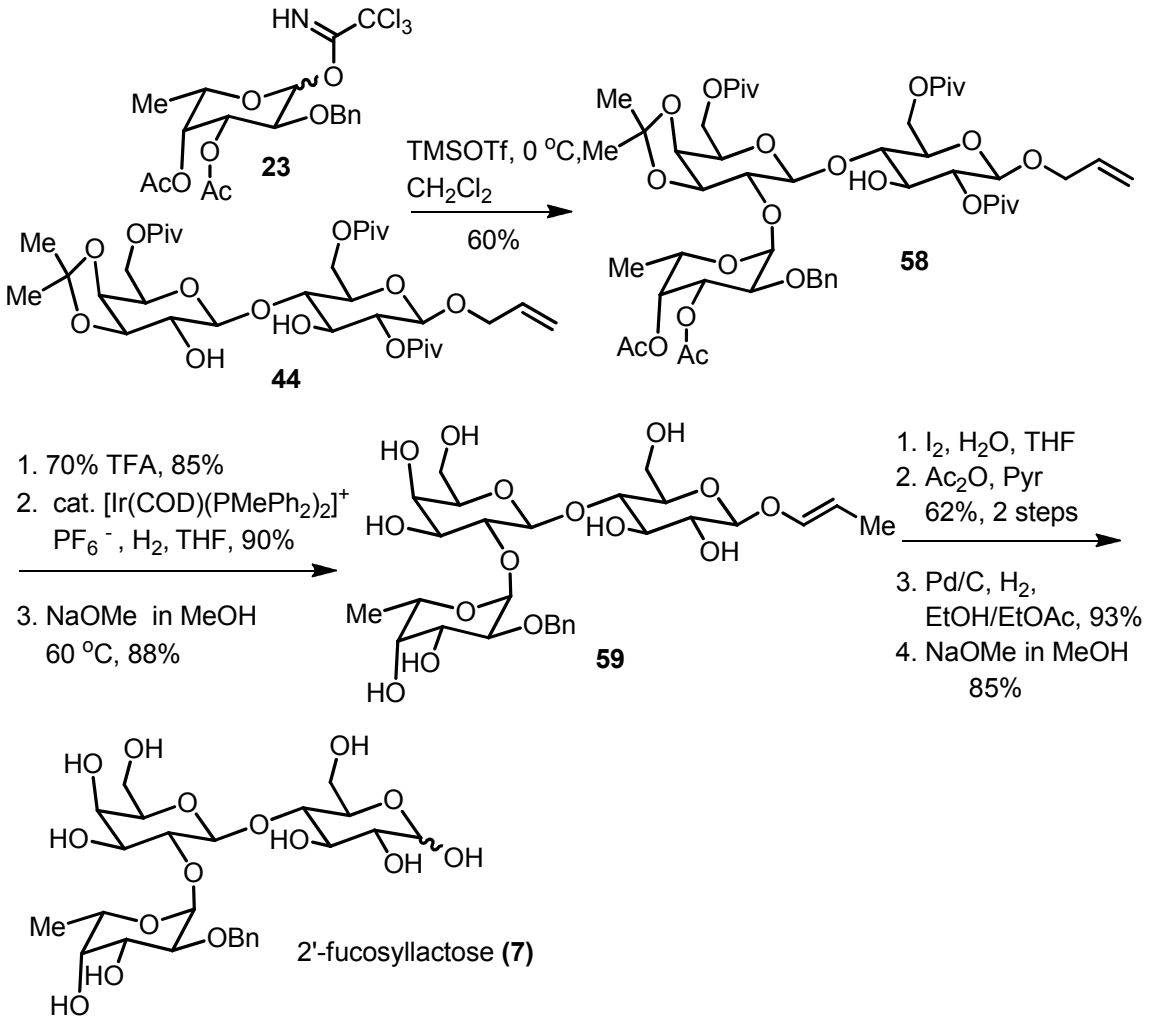


4.6.3. Glycosylation studies and the synthesis of HMO's

With substantial amount of acceptor **44** material in hand glycosylation studies were carried out with the donors **23**, **52** and **54**. The use of donor **54** resulted in the formation of the desired trisaccharide, but in low yield and poor selectivity under Lewis acids such as TMSOTf and BF_3•OEt_2. Similar results were observed with the peracetate donor **52**. The best results for the glycosylation were observed when the donor **23** was added to the acceptor **44** containing the Lewis acid TMSOTf under an inverse demand protocol (scheme 11).[146] Only the α- anomer was observed, which clearly indicates the importance of the participating C3 and C4 acetate groups. A substantial amount of an unknown product (see experimental details), presumably the tetrasaccharide was also observed. Use of molecular sieves and slower addition of the donor to the

reaction mixture containing the Lewis acid and the acceptor may further increase the yield of the reaction. The regioselctivity of glycosylation was confirmed by 2D-NMR, COSY and TOCSY.

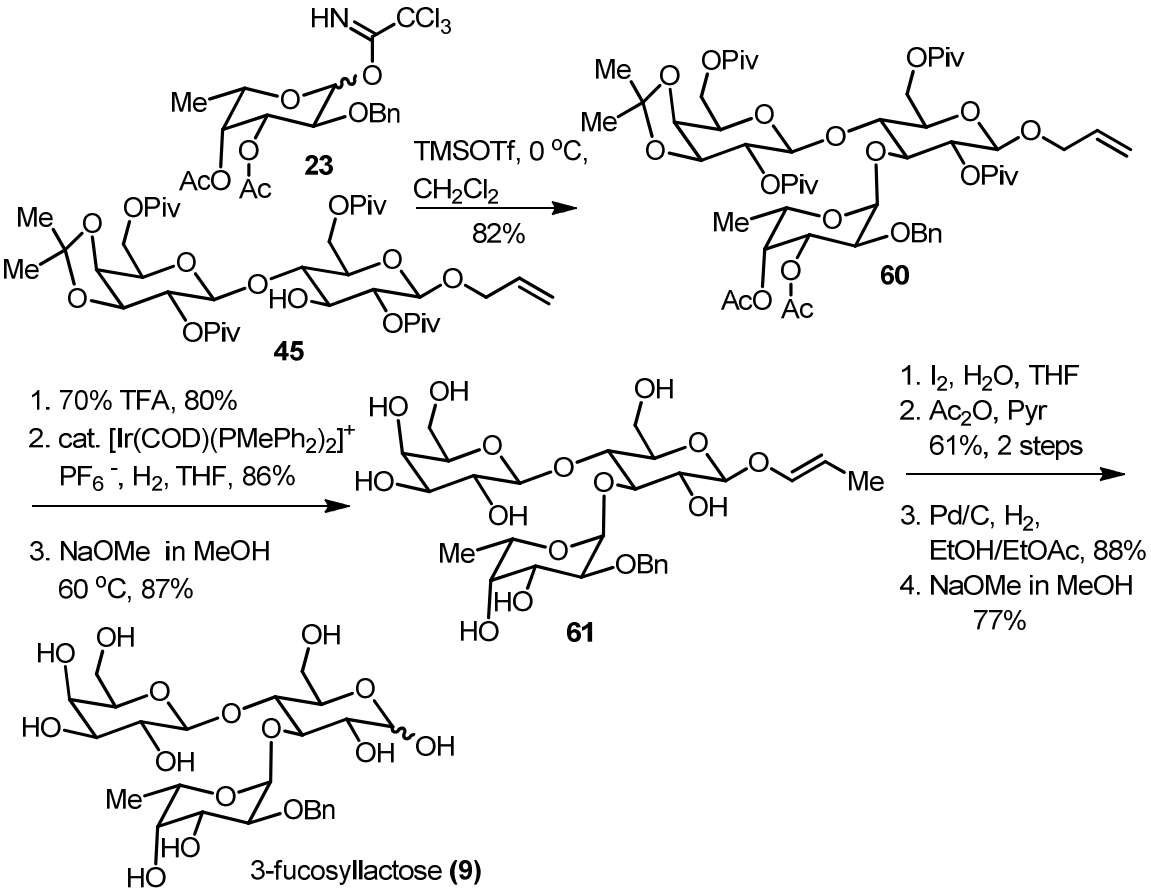
Scheme 11. Synthesis of 2'-fucosyllactose **(7)**



After establishing a procedure for the preparation of the trisaccharide **58**, the next synthetic strategy was the deprotection of the protecting groups. An initial attempt of removal of the allyl protecting group after the removal of the acetonide and esters proved impossible because of the solubility of the compound, under those reaction conditions. In a modified approach after the

removal of the acetonide protecting group using TFA, the allyl group was isomerized to the vinyl ether followed by methanolysis of the pivaloate and acetate groups to obtain intermediate **59** (scheme 11).[147] Removal of the vinyl ether using iodine and water gave a mixture of anomers which were then peracylated. Removal of the benzyl ether using Pd/C under an atmosphere of hydrogen and methanolysis gave the HMO 2'-fucosyllactose **(7)** as a mixture of anomers. The spectroscopic data matched in all respect to the one reported in literature.[148]

Scheme 12. Synthesis of 3-fucosyllactose **(9)**

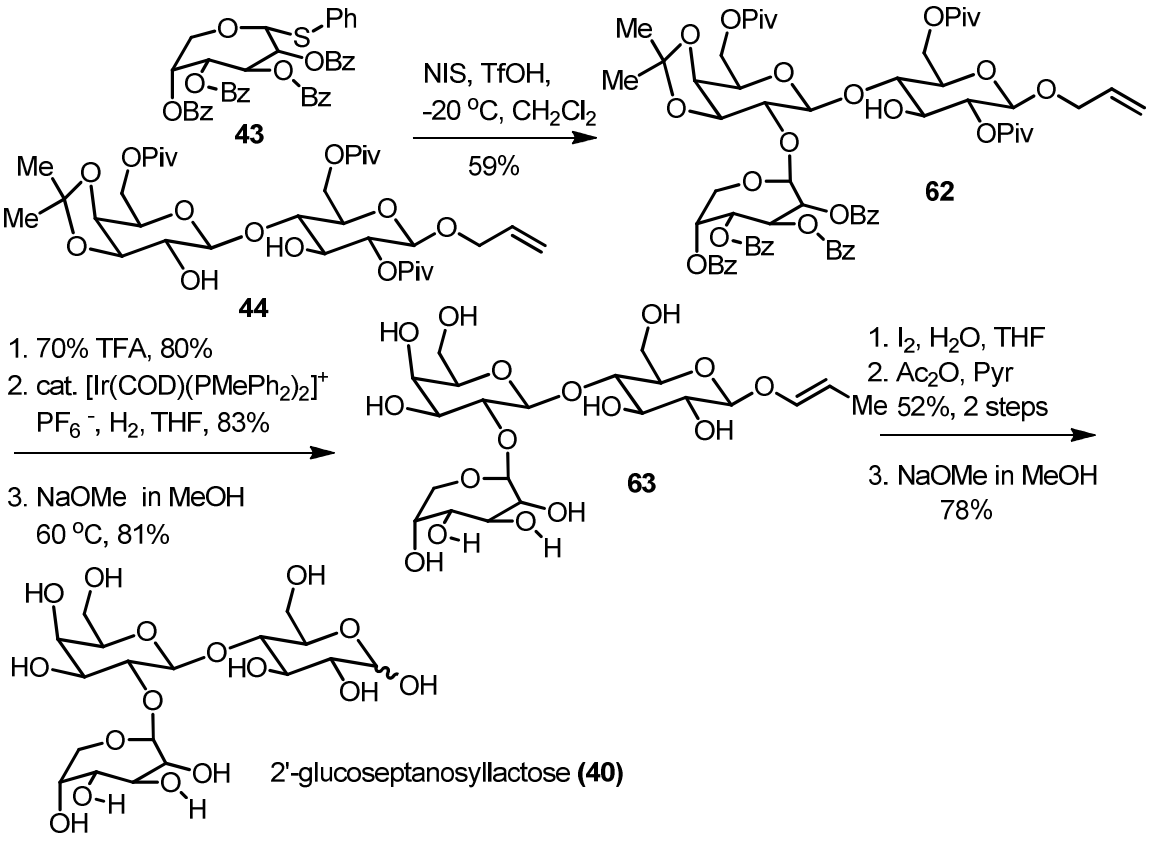


Based on the successful outcome of the synthesis of 2'-fucosyllactose **(7)**, starting from the tetrapivaloate **45** the trisaccharide **60** was synthesized in good

yield and exclusive α-selectivity. Using the similar deprotection strategy the 3-fucosyllactose **(9)** was obtained as a mixture of anomers and the spectroscopic data matched the literature.[147]

Now with two natural HMO's synthesized with the designed protocol, an approach to an unnatural HMO **(40)** was envisioned. The McDonald group is known for the synthesis of unnatural septanose sugars of the type **43** [149] and these were chosen as donor substrate for the glycosylation with the tripivaloate acceptor **44**. Thus far in the synthesis of the two HMO's **(7)** and **(9)** no toxic metal or malodorous chemicals were used.

Scheme 13. Synthesis of 2'-glucoseptanosyllactose **(40)**

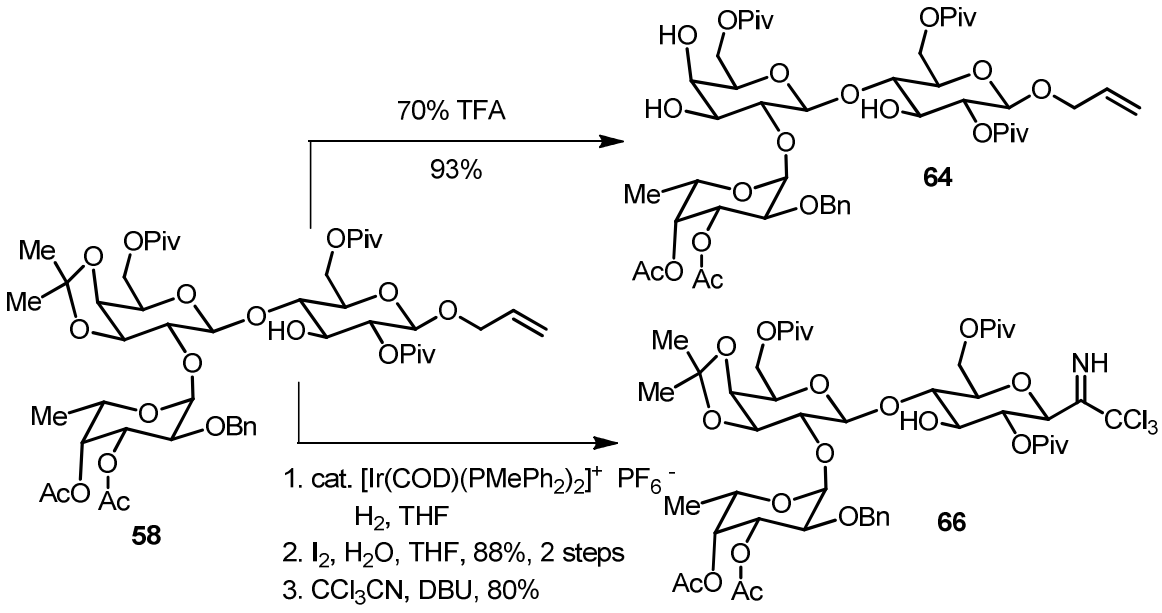


In the case of HMO **(40)** because of the ready availability of these septanose sugars in the laboratory the thioglygoside **43** was chosen as the

donor. The glycosylation carried out with acceptor **44** in presence of NIS and TfOH provided the trisaccharide **62** (scheme 13).[148] Removal of the acetonide protecting group, followed by isomerization and methanolysis provided intermediate **63**. Removal of the anomeric vinyl ether followed by peracylation and methanolysis gave the unnatural HMO 2'-glucoseptanosyllactose **(40)**.

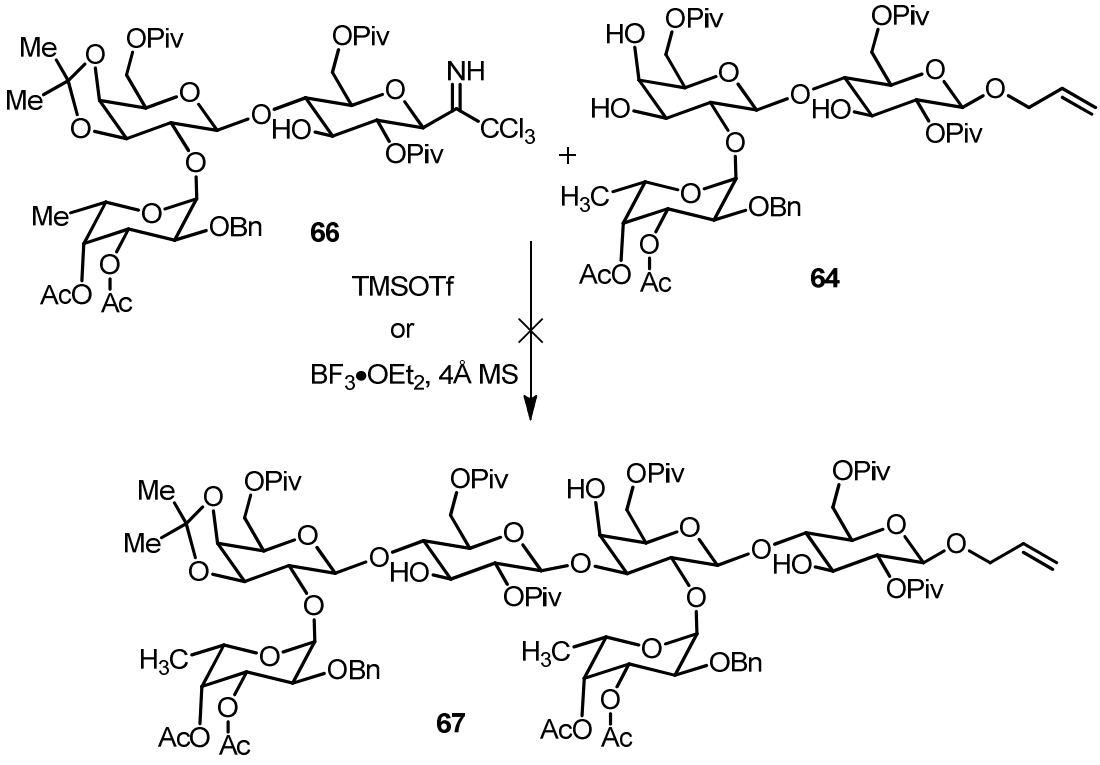
To date the most complex of the HMO's that have been put together are the lacto-*N*-fucopentaose III **(36)** by Schmidt *et al*. and difucosyllacto-*N*-hexaose **(10)** (figure 3). In these cases the fucose was introduced at a late stage in the synthetic process. Based on the successful synthesis of the trisaccharides **58**, **60** and **62** (schemes 11, 12 and 13) the next step was to try a modular approach of putting together two trisaccharides by a 3+3 approach to obtain a hexasaccharide. If the method became viable the modular approach could then be extended to put together more complex HMO's.

Scheme 14. Synthesis of trisaccharide donor **66** and acceptor **64**



In this regard the trisaccharide **58** was converted into the donor **66** by removal of the allyl group as before and converting the free anomeric alcohol to the trichloroacetamidate (scheme 14).[145] The acceptor was compound **64** obtained during the synthesis of 2'-fucosyllactose **(7)** (scheme 14). Initial attempts of glycosylation carried out using Lewis acids such as TMSOTf and BF_3•OEt_2 were unsuccessful.

Scheme 15. 3+3 modular approach for the synthesis of hexasaccharide **67**



More research needs to be done to find out the best coupling partners for this glycosylation. The trichloroacetamidate donor might need to be replaced by either a glycosylfluoride or a thiol. The reactivity could also be fine tuned by replacing some of the protecting groups, preferably the pivaloate by acetate.

4.7. Experimental details

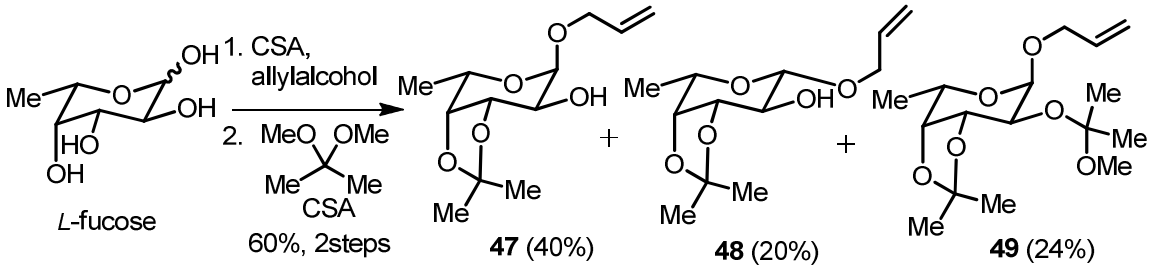
General information: ^{1}H NMR and ^{13}C NMR spectra were recorded on Varian INOVA 600, Unity 600 and INOVA 400 spectrometers. NMR spectra were recorded in solutions of deuterated chloroform ($CDCl_3$) with the residual chloroform (7.27 ppm for ^{1}H NMR and 77.23 ppm for ^{13}C NMR) taken as the internal standard, deuterated methanol (CD_3OD) with residual methanol (3.31 ppm for ^{1}H NMR and 49.3 ppm for ^{13}C NMR) taken as the internal standard, or deuterated benzene with residual benzene (7.16 ppm for ^{1}H NMR and 128.23 ppm for ^{13}C NMR) taken as the internal standard, and were reported in parts per million (ppm). Abbreviations for signal coupling are as follows: s, singlet; d, doublet; t, triplet; q, quartet; dd, doublet of doublet; ddd, doublet of doublet of doublet; dt, doublet of triplet; app d, apparent doublet; app t, apparent triplet; m, multiplet.

IR spectra were collected on a Mattson Genesis II FT-IR spectrometer as neat films on sodium chloride discs. Mass spectra (high resolution ESI and APCI) were recorded on a Finnigan LTQ FTMS Mass spectrometer. Optical rotations were measured using a Perkin-Elmer 341 polarimeter (concentration in g/100mL). Thin Layer Chromatography (TLC) was performed on precoated glass backed plates purchased from Whatman (silica gel $60F_{254}$; 0.25mm thickness). Flash column chromatography was carried out with silica gel 60 (230-400 mesh ASTM) from EM Science.

All reactions were carried out with anhydrous solvents in oven dried or flame dried and argon-charged glassware. All anhydrous solvents were dried

with 4 Å molecular sieves purchased from Sigma-Aldrich and tested for trace water content with Coulometric KF titrator from Denver instruments. All solvents used in extraction procedures and chromatography were used as received from commercial suppliers without prior purification.

Synthesis of allyl acetonide 47 and 48



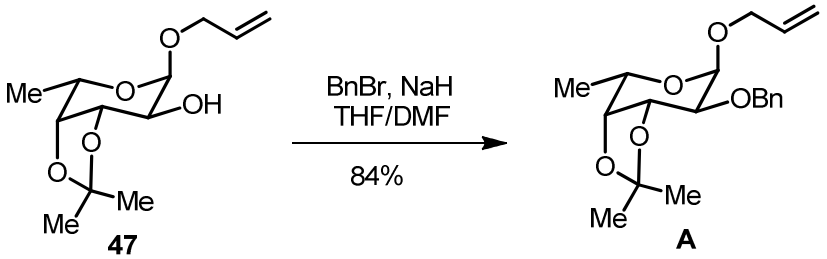
To a solution of L-fucose (25 g, 152.2 mmol) in allyl alcohol (250 mL) was added 10-camphorsulphonic acid (0.7 g, 3.0 mmol) and the reaction heated to reflux (90^{o}C external) for 2 h. Triethylamine (1.0 mL) was then added and the solvents removed under vacuum. Removed any trace amount of allylacohol by azeotroping with toluene and dried the semisolid obtained under vacuum and carried on to the next step without purification.

To the above crude added 2, 2-dimethoxypropane (120 mL), acetone (120 mL) and 10-camphorsulphonic acid (0.7 g, 3.0 mmol) and stirred the reaction at room temperature for 24 h. Triethylamine (2.0 mL) was then added and the solvents removed under vacuum. The crude was purified by flash chromatography using 20% ethyl acetate in hexanes as eluent to obtain the α-anomer **47** (15.0 g, 40%), β-anomer **48** (7.5 g, 20%) and compound **49** as the β-anomer (11.0 g, 24%) **^{1}H NMR** (600 MHz, $CDCl_3$) **β-anomer**: δ 5.96-5.90 (m, 1H), 5.30 (dd, *J* = 2.4, 17.4 Hz, 1H), 5.21 (d, *J* = 10.8 Hz, 1H), 4.38 (dd, *J* = 2.7,

12.6 Hz, 1H), 4.20 (d, *J* = 8.4 Hz, 1H), 4.08 (dd, *J* = 6.6, 13.2 Hz, 1H), 4.04 (dd, *J* = 6.0, 7.5 Hz, 1H), 4.00 (dd, *J* = 2.4, 5.1 Hz, 1H), 3.85 (dq, *J* = 2.4, 6.0, 13.2 Hz, 1H), 3.56 (dd, *J* = 7.8 Hz, 1H), 1.53 (s, 3H), 1.42 (d, *J* = 6.0 Hz, 3H), 1.35 (s, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 133.9, 118.2, 110.0, 101.1, 78.9, 76.4, 73.7, 70.1, 69.3, 28.4, 26.5, 16.7; **HRMS (ESI)**: *m/z* calcd. for $C_{12}H_{20}O_5Na$ ($M+Na^+$) 267.1203, found 267.1200; **FT-IR**: 3444, 2984, 2873, 1379, 1066, 1030, 868 cm^{-1} ; **$[\alpha]_D^{25}$** = - 30.8 (c=0.825, $CHCl_3$).

^{1}H NMR (600 MHz, $CDCl_3$) **α-anomer**: δ 5.95-5.88 (m, 1H), 5.30 (dd, *J* = 1.2, 17.1 Hz, 1H), 5.21 (d, *J* = 0.6, 10.5, Hz, 1H), 4.87 (d, *J* = 3.6 Hz, 1H), 4.26-4.20 (m, 2H), 4.14 (dq, *J* = 1.8, 6.6, 13.5 Hz, 1H), 4.08-4.02 (m, 2H), 3.80 (dd, *J* = 3.6 , 6.9 Hz, 1H), 1.52 (s, 3H), 1.36 (s, 3H), 1.32 (d, *J* = 6.0 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 133.9, 117.8, 109.4, 96.8, 76.4, 75.8, 69.6, 68.7, 64.1, 28.0, 26.1, 16.4; **HRMS (ESI)**: *m/z* calcd. for $C_{12}H_{20}O_5Na$ ($M+Na^+$) 267.1203, found 267.1200; **FT-IR**: 3444, 2941, 2935, 2873, 1067, 1025, 867 cm^{-1} ; **$[\alpha]_D^{25}$** = - 124.9 (c=1.105, $CHCl_3$).

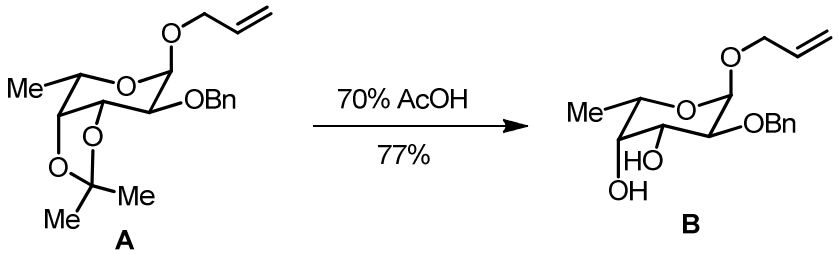
Synthesis of benzyl ether fucose A



To a solution of the compound **47** (17.8 g, 72.8 mmol) in a mixture of THF (150 mL) and DMF (15 mL) (10:1) at 0 ^oC was added NaH (2.1 g, 87.4 mmol, 60% in mineral oil) and the reaction stirred at room temperature for 15 min. The reaction

was cooled to 0 °C and added benzyl bromide (14.9 g, 87.4 mmol) and tetrabutylammonium iodide (TBAI) (25 mg). The reaction was stirred at room temperature for 16h. The reaction was diluted with water and the aqueous was extracted with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 5% ethyl acetate in hexanes as eluent to obtain the compound **A** as oil (20.5 g, 84%).**^{1}H NMR** (600 MHz, $CDCl_3$) **α-anomer**: δ 7.38 (d, *J* = 7.2 Hz, 2H), 7.34 (t, *J* = 7.2 Hz, 2H), 7.28 (t, *J* = 7.2 Hz, 1H), 5.97-5.91(m, 1H), 5.35 (dd, *J* = 1.8, 17.1 Hz, 1H), 5.23 (d, *J* = 10.8 Hz, 1H), 4.81 (d, *J* = 3.6 Hz, 1H), 4.81 (d, *J* = 11.4 Hz, 2H) 4.73 (d, *J* = 12.6 Hz, 2H), 4.36 (dd, *J* = 6.0, 7.8 Hz, 1H), 4.19-4.12 (m, 2H), 4.06 (dd, *J* = 2.4, 5.4 Hz, 1H), 4.01 (ddd, *J* = 0.6, 6.3, 12.6 Hz, 1H), 3.53 (dd, *J* = 3.6, 7.8 Hz, 1H), 1.42 (s, 3H), 1.36 (s, 3H), 1.33 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.4, 133.9, 128.4, 128.0, 127.8, 117.9, 108.8, 96.1, 76.3, 76.2, 76.0, 72.3, 68.4, 63.2, 28.3, 26.5, 16.4; **HRMS (ESI)**: *m/z* calcd. for $C_{19}H_{26}O_5Na$ ($M+Na^+$) 357.1672, found 357.1669; **FT-IR**: 2984, 2901, 1379, 1074, 869, 735 cm^{-1} ; **$[\alpha]_D^{25}$** = - 107.1 (c=1.115, $CHCl_3$).

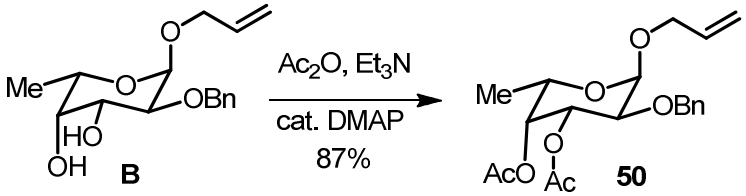
Synthesis of the diol B



A solution of the compound **A** (20.5 g, 61.3 mmol) in 70% acetic acid (102 mL) was heated at 45 °C for 4 h. After cooling to room temperature diluted the

reaction mixture with water and extracted the aqueous with ethyl acetate. The organic layer was washed with saturated $NaHCO_3$ solution, dried over $MgSO_4$ filtered and concentrated to obtain compound **B** as oil (14.0 g, 77%). Carried onto the next step without purification.**^{1}H NMR** (600 MHz, $CDCl_3$) **α-anomer**: δ 7.36-7.34 (m, 4H), 7.32-7.29 (m, 1H), 5.95-5.88 (m, 1H), 5.32 (d, *J* = 16.5 Hz, 1H), 5.21 (d, *J* = 10.2 Hz, 1H), 4.86 (d, *J* = 3.6 Hz, 1H), 4.66 (d, *J* = 11.4 Hz, 1H) 4.61 (d, *J* = 11.4 Hz, 1H), 4.14 (dd, *J* = 4.8, 12.9 Hz, 1H), 4.02 (d, *J* = 10.2 Hz, 1H), 3.99-3.93 (m, 2H), 3.81 (s, 1H), 3.71 (dd, *J* = 3.6, 9.6 Hz, 1H), 2.66 (s, 1H), 2.48 (s, 1H), 1.27 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 138.1, 134.0, 128.7, 128.3, 128.2, 117.9, 95.6, 76.5, 72.6, 71.6, 69.5, 68.5, 65.6, 16.2; **HRMS (ESI)**: *m/z* calcd. for $C_{16}H_{22}O_5Na$ ($M+Na^+$) 317.1359, found 317.1353; **FT-IR**: 3424, 2984, 2901, 1366, 1089, 869, 736 cm^{-1} ; **$[\alpha]_D^{25}$** = - 127.9 (c=1.03, $CHCl_3$).

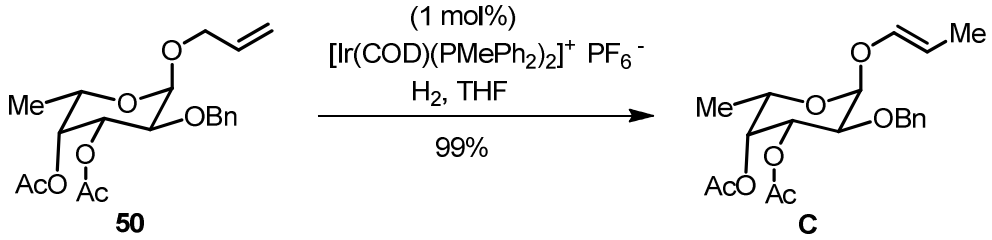
Synthesis of diacetate fucose 50



To a solution of the compound **B** (14.0 g) in pyridine (42 mL) was added acetic anhydride (42 mL) and catalytic amount of dimethyl aminopyridine. The reaction was stirred at room temperature for 16 h. The reaction was diluted with water and extracted the aqueous with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain compound **50** as oil (15.8 g, 87%). **^{1}H NMR** (600 MHz, $CDCl_3$) **α-anomer**: δ 7.35-7.32 (m, 4H), 7.31-7.27 (m,

1H), 5.95-5.88 (m, 1H), 5.36-5.28 (m, 3H), 5.22 (d, *J* = 10.2 Hz, 1H), 4.88 (d, *J* = 3.6 Hz, 1H), 4.69 (d, *J* = 12.0 Hz, 1H) 4.60 (d, *J* = 12.6 Hz, 1H), 4.18-4.14 (m, 2H), 4.00 (dd, *J* = 6.0, 13.5 Hz, 1H), 3.84 (dd, *J* = 3.6, 10.8 Hz, 1H), 2.13 (s, 3H), 1.99 (s, 3H), 1.10 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.7, 170.2, 138.3, 133.8, 128.5, 128.0, 127.9, 118.1, 96.4, 73.6, 73.0, 71.8, 70.2, 68.7, 64.5, 21.0, 20.8, 16.0; **HRMS (ESI)**: *m/z* calcd. for $C_{20}H_{26}O_7Na$ ($M+Na^+$) 401.1570, found 401.1571; **FT-IR**: 2983, 2903, 1740, 1238, 1366, 1033, 738, 696 cm^{-1} ; **$[\alpha]_D^{25}$** = - 81.8 (c=1.0, $CHCl_3$).

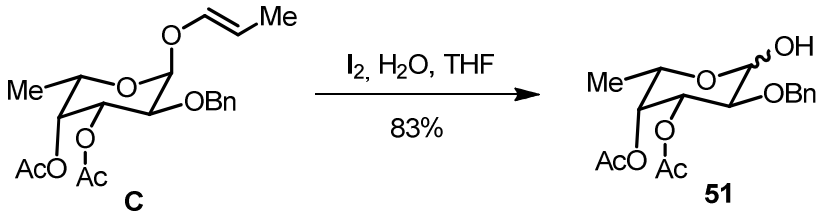
Isomerization of compound 50 to vinylether C



A solution of the catalyst $Ir(COD)(PMePh_2)_2]^+PF_6^-$ (0.35 g, 0.41 mmol) in THF (21 mL) was degassified and then stirred for 15 min under an athmosphere of hydrogen (red color of the solution decolorised). This was then cannulated into a solution of compound **50** (15.8 g, 41.7 mmol) in THF (316 mL) at room temperature and stirred for 30 min. The reaction was diluted with saturated $NaHCO_3$ and extracted the aqueous with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain compound **C** as oil. The compound was carried forward without any further purification. **^{1}H NMR** (600 MHz, $CDCl_3$) **α-anomer**: δ 7.35-7.28 (m, 5H), 6.14 (d, *J* = 12.6 Hz, 1H), 5.36 (dd, *J* = 3.0, 10.5, Hz, 1H), 5.29 (d, *J* = 3.6 Hz, 1H), 5.19-5.14 (m, 1H), 5.03 (d, *J* = 3.0 Hz, 1H), 4.69 (d, *J* = 12.6 Hz, 1H), 4.62 (d, *J* = 12.6 Hz, 1H), 4.13 (dd, *J* =

6.6, 13.2 Hz, 1H), 3.85 (dd, *J* = 3.0, 10.5 Hz, 1H), 2.13 (s, 3H), 1.99 (s, 3H), 1.56 (d, *J* = 6.6 Hz, 3H), 1.09 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.6, 170.2, 143.1, 138.1, 128.6, 128.0, 127.9, 104.8, 96.9, 73.2, 73.1, 71.6, 70.1, 65.0, 21.0, 20.8, 16.0, 12.6; **HRMS (ESI)**: *m/z* calcd. for $C_{20}H_{26}O_7Na$ ($M+Na^+$) 401.1570, found 401.1563; **FT-IR**: 2937, 1741, 1238, 1078, 738, 696 cm^{-1} ; **$[\alpha]_D^{25}$** = - 92.2 (c=1.065, $CHCl_3$).

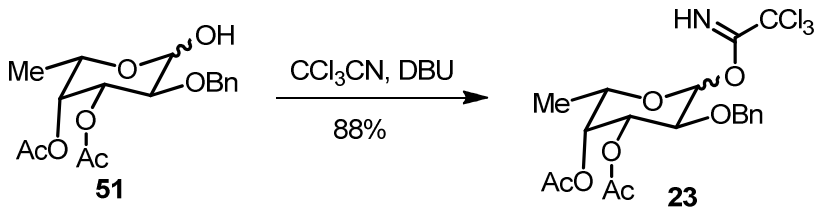
Synthesis of deallylated compound 51



To a solution of the above compound **C** in a mixture of THF (880 mL) and water (220 mL) (4:1) was added iodine (21.2 g, 83.5 mmol) at room temperature and the brown reaction mixture stirred for 30 min. The reaction mixture was diluted with ethyl acetate and the organic layer washed with saturated $Na_2S_2O_3$, water and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 10% ethyl acetate in hexanes as eluent to obtain compound **51** as a solid (11.8 g, 83%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.35-7.32 (m, 4H), 7.31-7.27 (m, 1H), 5.95-5.88 (m, 1H), 5.36-5.28 (m, 3H), 5.22 (d, *J* = 10.2 Hz, 1H), 4.88 (d, *J* = 3.6 Hz, 1H), 4.69 (d, *J* = 12.0 Hz, 1H) 4.60 (d, *J* = 12.6 Hz, 1H), 4.18-4.14 (m, 2H), 4.00 (dd, *J* = 6.0, 13.5 Hz, 1H), 3.84 (dd, *J* = 3.6, 10.8 Hz, 1H), 2.13 (s, 3H), 1.99 (s, 3H), 1.10 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.7, 170.2, 138.3, 133.8, 128.5, 128.0, 127.9, 118.1, 96.4, 73.6, 73.0, 71.8, 70.2, 68.7, 64.5, 21.0, 20.8, 16.0; **HRMS**

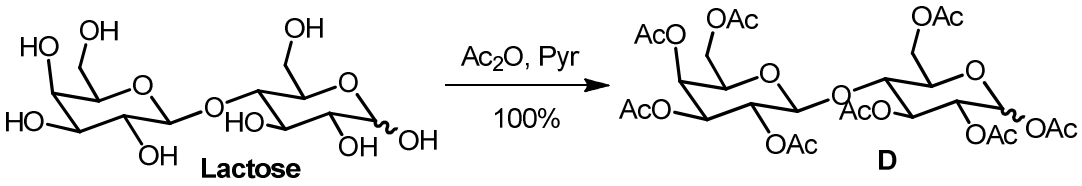
(ESI): *m/z* calcd. for $C_{20}H_{26}O_7Na$ ($M+Na^+$) 401.1570, found 401.1571; **FT-IR**: 2983, 2903, 1740, 1238, 1033, 738, 696 cm^{-1} ; $[\alpha]_D^{25}$ = - 52.1 (c=1.0, $CHCl_3$); **Mp:** 80-83 ^oC.

Synthesis of trichloroacetamidate donor 23



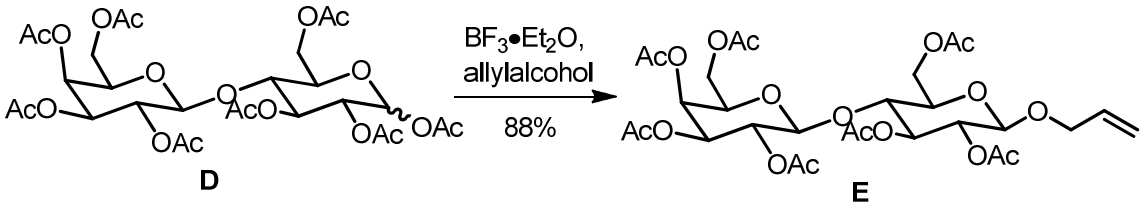
To a solution of compound **51** (5.0 g, 14.7 mmol) in CH_2Cl_2 at room temperature was added trichloroacetonitrile (10.6 g, 73.8 mmol) followed by DBU (0.45 g, 2.9 mmol) and stirred the brown reaction mixture for 20 h. The solvent was removed under vacuum and the crude was purified by flash chromatography using 10% ethyl acetate in hexanes containing 2% Et_3N as eluent to obtain compound **23** as a white solid (5.4 g, 76%, α-anomer) and a mixture of α and β-anomer (12%). **^{1}H NMR** (600 MHz, $CDCl_3$) **α-anomer**: δ 8.6 (s, 1H), 7.34-7.27 (m, 5H), 6.52 (d, *J* = 3.6 Hz, 1H), 5.39-5.36 (m, 2H), 4.70 (d, *J* = 12.0 Hz, 1H), 4.65 (d, *J* = 12.0 Hz, 1H), 4.35 (q, *J* = 6.6, 13.2 Hz, 1H), 4.02 (dd, *J* = 2.4, 10.2 Hz, 1H), 2.15 (s, 3H), 2.00 (s, 3H), 1.15 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.6, 170.2, 161.4, 137.9, 128.5, 127.9, 127.6, 94.6, 73.0, 72.7, 71.1, 70.0, 67.5, 21.0, 20.8, 16.1; **HRMS (ESI)**: *m/z* calcd. For $C_{19}H_{22}Cl_3NO_7Na$ ($M+Na^+$) 504.0354, found 504.0353; **FT-IR**: 3292, 2991, 1743, 1262, 1159, 1027, 798, 638 cm^{-1} ; $[\alpha]_D^{25}$ = - 84.9 (c=1.225, $CHCl_3$); **Mp:** 150-152 ^oC.

Synthesis of peracylated lactose D



To a stirred solution of lactose (80 g, 233 mmol) in pyridine (328 mL) was added acetic anhydride (358 g, 3505 mmol) and the reaction stirred for 48 h at room temperature. The solvent was removed under vacuum and azetroped the crude with toluene. The crude was dissolved in water and extracted with ethyl acetate. The organic layer was washed with saturated $NaHCO_3$ solution, dried over $MgSO_4$, filtered and concentrated to obtain oil. Repeated washing with hexanes followed by decantation resulted in a white solid. Recrystallization using acetone and hexanes resulted in compound **D** as a white solid (158 g, 100%).[93]

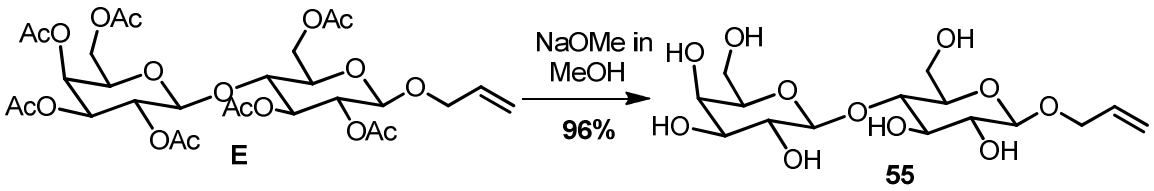
Synthesis of allyllactose octaacetate E



To a solution of compound **D** (20 g, 29.4 mmol) and allyl alcohol (2 g, 35.3 mmol) in CH_2Cl_2 (400 mL) at 0 ^oC was added BF_3•OEt_2 (6.2 g, 44.2 mmol) and the reaction stirred for 30 min. After stirring for 68 h at room temperature added 12 g of K_2CO_3 and stirred the reaction for 30 min. The solids were filtered and the filtrate diluted with water. The aqueous layer was extracted with CH_2Cl_2 and the combined organic layer was washed with brine, dried over $MgSO_4$, filtered and

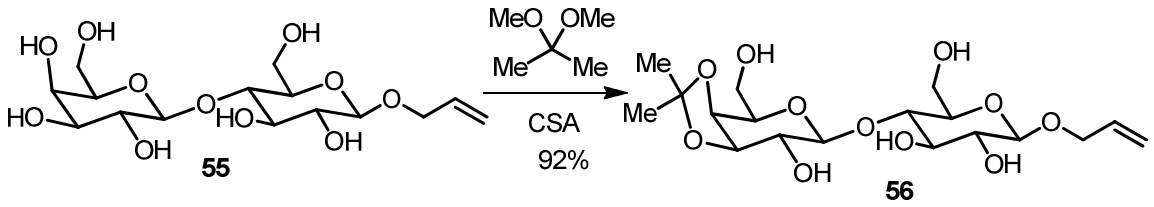
concentrated to obtain oil. Repeated washing with hexanes followed by decantation gave the allylated lactose **E** as a white solid (17.5 g, 88%).[93, 94]

Synthesis of allyllactose 55



To a solution of compound **E** (17.5 g, 25.8 mmol) in methanol (175 mL) at room temperature was added a 0.5 M solution of NaOMe in MeOH (5.2 mL, 2.58 mmol) and stirred for 3.5 h. The reaction was diluted with methanol and neutralized with amberlyst 120(H^+) resin. Filtered the resin and removed the solvent under vacuum to obtain the allyllactose **55** as a white solid (9.5 g, 96%).[93]

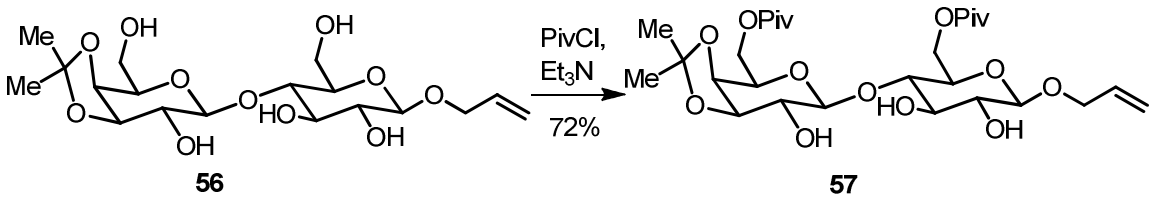
Synthesis of allyllactose-acetonide 56



To a solution of compound **55** (9.6 g, 25.1 mmol) in 2,2-dimethoxypropane (576 mL) was added CSA (0.3 g, 1.25 mmol) and the reaction stirred at room temperature for 108 h. Et_3N (2.5 ml) was then added and the reaction stirred for 15 min before removing the solvents under vacuum. Azeotroped the reaction with toluene and added a mixture of methanol and water (250 mL : 50 mL) and heated the reaction to reflux for 3 h. The solvent was removed under vacuum and the water azeotroped using toluene to obtain a white solid. The solid was redissolved using a mixture of ethyl acetate and dichloromethane and

precipitated out using hexanes, filtered and dried to obtain compound **56** (9.8 g, 92%).[93, 94]

Synthesis of dipivaloate-lactose 57

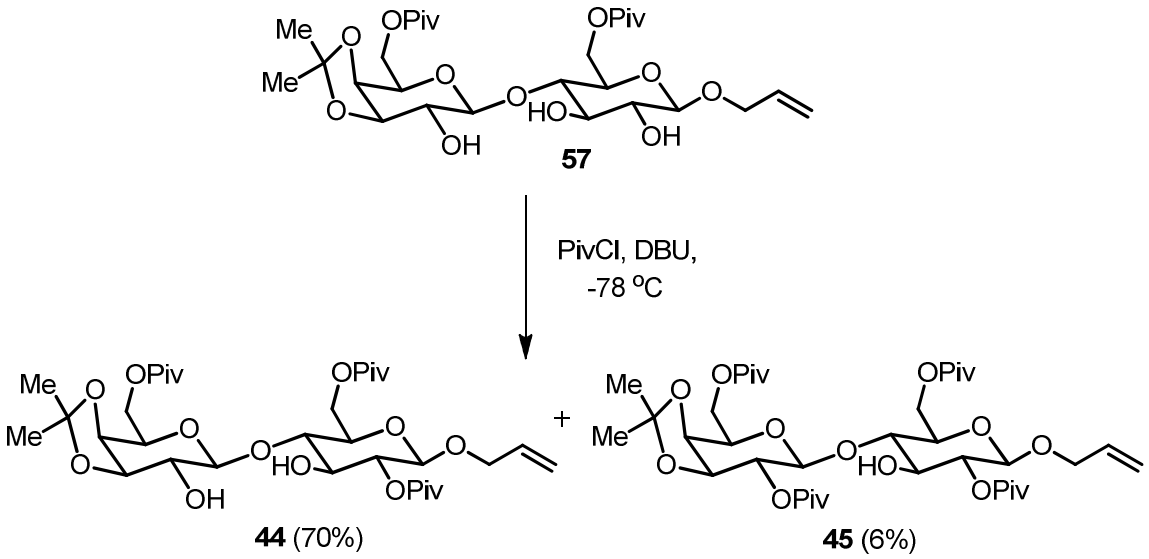


56 **57**

To a solution of compound **56** (2.1 g, 4.9 mmol), DMAP (0.06 g, 0.49 mmol) and pyridine (10.5 mL) in 1,2-dichloroethane (31 ml) at 0 ^{o}C was added pivaloyl chloride (1.2 g, 9.9 mmol) and the reaction stirred for 3 h and then at room temperature for 30 min. The reaction mixture was diluted with water and the aqueous was extracted with CH_2Cl_2. The organic layer was washed with water, brine, dried over $MgSO_4$, filtered and concentrated to obtain oil. Toluene was then added and trace amount of pyridine was removed under vacuo to obtain a white solid. Finally washed the solid with pentane and filtered to obtain compound **57** (2.1 g, 72%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.98-5.92 (m, 1H), 5.32 (dd, *J* = 1.2, 17.4 Hz, 1H), 5.23 (d, *J* = 10.8 Hz, 1H), 4.63 (d, *J* = 12.0 Hz, 1H), 4.44 (dd, *J* = 3.6, 12.0 Hz, 1H), 4.37-4.34 (m, 1H), 4.36 (d, *J* = 7.8 Hz, 1H), 4.24 (dd, *J* = 8.4, 12.0 Hz, 1H), 4.23 (d, *J* = 8.4 Hz, 1H), 4.18-4.06 (m, 6H), 3.64-3.60 (m, 2H), 3.50 (dd, *J* = 6.0, 9.9 Hz, 1H), 3.45-3.41 (m, 2H), 3.30 (dd, *J* = 9.6 Hz, 1H), 2.56 (s, 1H), 1.53 (s, 3H), 1.34 (s, 3H), 1.22 (s, 9H), 1.21 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.9, 178.5, 133.8, 118.4, 110.8, 103.6, 101.1, 81.5, 78.8, 75.0, 73.9, 73.4, 73.3, 73.2, 71.9, 70.5, 63.6, 63.4, 39.0 (2), 28.2, 27.3, 27.2, 26.4; **HRMS (ESI)**: *m/z* calcd. For $C_{28}H_{46}O_{13}Na$ ($M+Na^{+}$) 613.2830, found

613.2831; **FT-IR**: 3458, 3419, 2978, 1733, 1722, 1286, 1169, 1072, 1020 cm^{-1} ; $[\alpha]_D^{25}$ = + 33.4 (c=1.135, $CHCl_3$); **Mp**: 173-175 °C.

Preparation of tri- and tetrapivaloate lactose 44 and 45

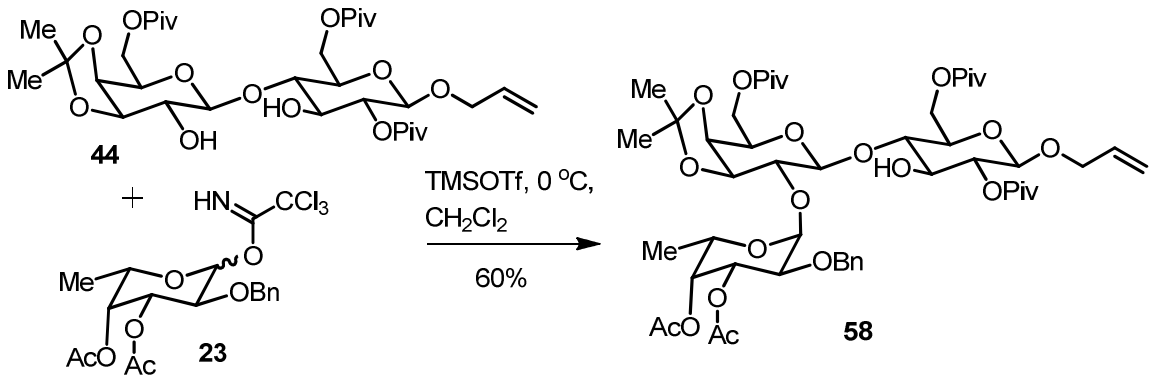


To a solution of compound **57** (4.2 g, 7.1 mmol) and DMAP (0.086 g, 0.71 mmol) in CH_2Cl_2 (105 mL) at -78 °C was added DBU (1.19 g, 7.8 mmol) and the reaction stirred for 15 min. Pivaloyl chloride (0.94 g, 7.8 mmol) was then added dropwise and the reaction stirred for 30 min at -78 °C. The reaction was quenched with MeOH at -78 °C and then warmed to room temperature. After diluting the reaction with saturated $NaHCO_3$ the aqueous was extracted with CH_2Cl_2. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 15% acetone in hexanes as eluent to obtain tripivaloate compound **44** as a white solid (3.37 g, 70%). Obtained tetrapivaloate **45** as a white solid as the byproduct (0.35 g, 6%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.86-5.79 (m, 1H), 5.24 (dd, *J* = 1.8, 17.1 Hz, 1H), 5.16 (dd, *J* = 1.2, 10.5 Hz, 1H), 4.87 (dd, *J* = 8.4 Hz, 1H), 4.65 (d, *J* = 12.0 Hz, 1H),

4.44 (d, *J* = 8.4 Hz, 1H), 4.41 (dd, *J* = 3.6, 12.0 Hz, 1H), 4.29 (dd, *J* = 4.8, 12.9 Hz, 1H), 4.23 (dd, *J* = 8.4, 12.0 Hz, 1H), 4.14-4.07 (m, 5H), 4.05 (dd, *J* = 6.0, 12.9 Hz, 1H), 3.71 (dd, *J* = 8.4 Hz, 1H), 3.61 (dt, *J* = 3.0, 8.1 Hz, 1H), 3.49 (dd, *J* = 6.0, 9.6 Hz, 1H), 3.44 (d, *J* = 3.0 Hz, 1H), 3.36 (dd, *J* = 9.6 Hz, 1H), 1.51 (s, 3H), 1.33 (s, 3H), 1.21 (s, 9H), 1.19 (s, 9H), 1.19 (2s, 18H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.9, 178.7, 176.9, 133.7, 117.7, 110.8, 103.8, 99.7, 82.7, 78.9, 73.8, 73.5, 73.4, 73.1, 72.1, 71.9, 70.0, 63.6, 63.5, 39.0 (2C), 38.9, 28.2, 27.2 (2C), 26.4; **HRMS (ESI)**: *m/z* calcd. For $C_{33}H_{54}O_{14}Na$ ($M+Na^+$) 674.3405, found 674.3406; **FT-IR**: 3463, 2973, 1731, 1284, 1157, 1072 cm^{-1} ; **$[\alpha]_D^{25}$** = + 24.0 (c=1.025, $CHCl_3$); **Mp**: 160-162 ^oC.

Tetrapivaloate-lactose 45: ^{1}H NMR (600 MHz, $CDCl_3$) δ 5.84-5.78 (m, 1H), 5.23 (dd, *J* = 1.2, 17.1 Hz, 1H), 5.16 (dd, *J* = 1.8, 10.5 Hz, 1H), 5.01-4.98 (m, 1H), 4.86 (app t, *J* = 9.0 Hz, 1H), 4.30-4.24 (m, 3H), 4.17-4.14 (m, 3H), 4.07 (dd, *J* = 3.6, 8.7 Hz, 1H), 3.75 (app t, *J* = 8.4 Hz, 1H), 3.41 (app t, *J* = 9.0, Hz, 1H), 1.53 (s, 3H), 1.31 (s, 3H), 1.21 (4s, 36H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.6, 178.0, 177.1, 176.8, 133.7, 117.6, 111.2, 100.9, 99.6, 81.3, 73.4, 73.1, 72.5 (2C), 72.4, 71.7, 69.9, 63.2, 62.7, 39.0, 38.9, 27.5, 27.4, 27.3, 27.2, 26.4; **HRMS (ESI)**: *m/z* calcd. For $C_{38}H_{62}O_{15}Na$ ($M+Na^+$) 781.3980, found 781.3982; **FT-IR**: 3471, 2973, 1735, 1280, 1141, 1056 cm^{-1} ; **$[\alpha]_D^{25}$** = + 26.1 (c=1.15, $CHCl_3$); **Mp**: 184-186 ^oC.

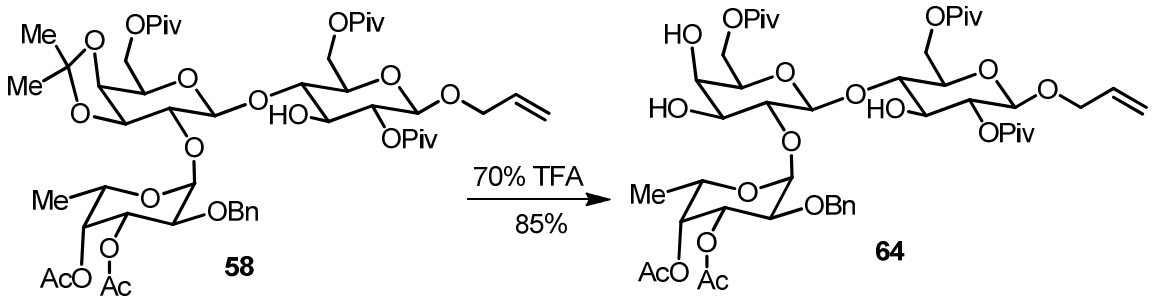
Synthesis of 2'-fucosyllactose trisaccharide 58



To a solution of acceptor **44** (0.7 g, 1.03 mmol) in CH_2Cl_2 (3.5 mL) was added 0.1 M TMSOTf (2.3 mg, 0.01 mmol) in CH_2Cl_2 at 0 °C. A solution of donor **23** (0.75 g, 1.55 mmol) in CH_2Cl_2 (14 mL) was then added using a syringe pump over a period of 1 h. (Monitored the consumption of acceptor by TLC). The reaction was quenched by the addition of Et_3N (0.2 mL) and the solvents removed under vacuum. The crude was purified by flash chromatography using 25% ethyl acetate in hexanes as eluent to obtain trisaccharide **58** as white solid (0.6 g, 60%). Obtained also a white solid as a byproduct (0.37 g, complex mixture of compound by ^{1}H NMR). **^{1}H NMR** (600 MHz, $CDCl_3$) 7.33-7.32 (m, 3H), 7.31 (m, 2H), 5.84-5.78 (m, 1H), 5.57 (d, *J* = 3.6 Hz, 1H), 5.33 (d, *J* = 3.6 Hz, 1H), 5.25-5.21 (m, 2H), 5.15 (d, *J* = 10.2 Hz, 1H), 4.86 (app t, *J* = 8.4 Hz, 1H), 4.69 (dd, *J* = 1.8, 11.7 Hz, 1H), 4.67 (d, *J* = 12.6 Hz, 1H), 4.64 (d, *J* = 12.0 Hz, 1H), 4.43 (d, *J* = 7.8 Hz, 1H), 4.40-4.36 (m, 3H), 4.29-4.23 (m, 2H), 4.20 (app t, *J* = 6.0 Hz, 1H), 4.13 (dd, *J* = 7.2, 12.0 Hz, 1H), 4.09 (dd, *J* = 1.8, 5.4 Hz, 1H), 4.03-4.00 (m, 3H), 3.84 (dd, *J* = 3.0, 10.8 Hz, 1H), 3.77 (app t, *J* = 6.0, 7.8 Hz, 1H), 3.70 (app t, *J* = 9.0 Hz, 1H), 3.66-3.63 (m, 1H), 3.43 (app t, *J* = 9.3 Hz, 1H), 2.12 (s, 3H), 1.96 (s, 3H), 1.50 (s, 3H), 1.33 (s, 3H), 1.21 (s, 9H), 1.20 (2s, 18H), 1.14 (d, *J* = 6.6 Hz,

3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.7, 177.9, 176.7, 170.7, 169.7, 138.3, 133.7, 128.5, 127.9, 127.8, 117.5, 110.8, 101.2, 99.7, 95.6, 81.4, 79.6, 75.3, 73.6, 73.4, 73.0, 72.9, 72.7, 72.3, 71.9, 69.9, 69.4, 65.0, 63.1, 62.7, 39.0, 38.9 (2C), 28.0, 27.3, 27.2, 26.4, 20.9 (2C), 16.0; **HRMS (ESI)**: *m/z* calcd. For $C_{50}H_{74}O_{20}Na$ ($M+Na^+$) 1017.4665, found 1017.4648; **FT-IR**: 3463, 2973, 1743, 1369, 1241, 1149, 1041 cm^{-1} ; **$[\alpha]_D^{25}$** = - 35.5 (c=1.05, $CHCl_3$); **Mp**: 92-94 °C.

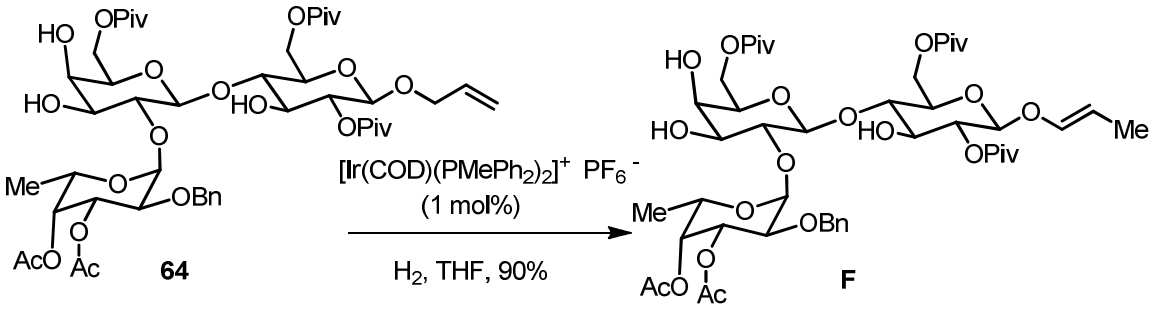
Synthesis of 2'-fucosyllactose trisaccharide diol 64



To a solution of the trisaccharide **58** (0.5 g, 0.5 mmol) in CH_2Cl_2 (20 mL) at 0 °C was added 70% trifluoroacetic acid (2 mL) and the reaction stirred for 2 h followed by 2 h at room temperature. The reaction mixture was diluted with water and the aqueous extracted with CH_2Cl_2. The organic layer was washed with saturated $NaHCO_3$, dried over $MgSO_4$, filtered and concentrated to obtain the diol **64** as a white solid (0.41 g, 85%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.42-7.33 (m, 3H), 7.32 (d, *J* = 6.6 Hz, 2H), 5.85-5.78 (m, 1H), 5.36 (dd, *J* = 3.6, 10.5 Hz, 1H), 5.29 (d, *J* = 3.6 Hz, 1H), 5.23 (d, *J* = 17.4 Hz, 1H), 5.16 (d, *J* = 10.8 Hz, 1H), 5.11 (d, *J* = 3.6 Hz, 1H), 4.85 (app t, *J* = 9.0 Hz, 1H), 4.73 (d, *J* = 11.4 Hz, 1H), 4.64 (d, *J* = 10.8 Hz, 1H), 4.48 (s, 1H), 4.44-4.38 (m, 3H), 4.33-4.24 (m, 4H), 4.01 (dd, *J* = 6.0, 13.5 Hz, 1H), 3.93 (dd, *J* = 3.0, 10.5 Hz, 1H), 3.80 (s, 1H), 3.76 (bs, 1H), 3.71 (app t, *J* = 8.4, Hz, 1H), 3.67 (dd, *J* = 3.6, 9.0 Hz, 1H), 3.64-3.59 (m, 4H),

2.15 (s, 3H), 2.03 (s, 3H), 1.21 (s, 18H), 1.20 (s, 9H), 1.13 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.7, 178.0, 176.8, 170.5, 170.0, 136.3, 133.8, 129.0, 128.9, 128.7, 117.5, 101.8, 99.8, 80.6, 79.1, 75.4, 74.9, 72.9 (2C), 72.8, 72.6 (2C), 71.6, 70.3, 69.8, 68.1, 66.6, 63.1, 62.3, 39.0 (2C), 38.9, 27.4, 27.3, 27.2, 21.0, 20.8, 16.2; **HRMS (ESI)**: *m/z* calcd. for $C_{47}H_{70}O_{20}Na$ ($M+Na^+$) 977.4352, found 977.4339; **FT-IR**: 3440, 2969, 1743, 1241, 1137, 1072 cm^{-1} ; **$[\alpha]_D^{25}$** = - 34.4 (c=0.605, $CHCl_3$); **Mp**: 204-206 °C.

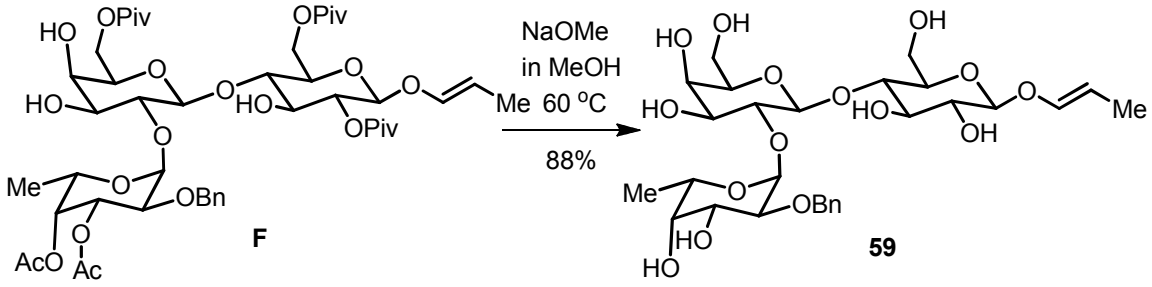
Synthesis of 2'-fucosyllactose trisaccharide vinylether F



A solution of the catalyst $Ir(COD)(PMePh_2)_2]^+PF_6^-$ (3.0 mg, 0.0034 mmol) in THF (0.5 mL) was degassified and then stirred for 15 min under an athmosphere of hydrogen (red color of the solution decolorised). This was then cannulated into a solution of compound **64** (0.33 g, 0.34 mmol) in THF (7.0 mL) at room temperature and stirred for 30 min. The reaction was diluted with saturated $NaHCO_3$ and the aqueous was extracted with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the vinylether **F** as white solid (0.3 g, 90%). The compound was carried forward without any further purification. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.40-7.36 (m, 3H), 7.34 (m, 2H), 6.14 (dd, *J* = 1.2, 12.3 Hz, 1H), 5.36 (dd, *J* = 3.0, 10.2 Hz, 1H), 5.29 (d, *J* = 2.4 Hz,

1H), 5.11 (d, *J* = 3.6 Hz, 1H), 5.06 (dq, *J* = 6.6, 7.2, 11.7 Hz, 1H), 4.89 (dd, *J* = 8.4, 9.3 Hz, 1H), 4.73 (d, *J* = 11.4 Hz, 1H), 4.64 (d, *J* = 10.8 Hz, 1H), 4.49 (s, 1H), 4.45 (dd, *J* = 4.2, 12.0 Hz, 1H), 4.37 (dd, *J* = 1.8, 12.0 Hz, 1H), 4.34-4.24 (m, 3H), 4.22 (dd, *J* = 7.2 Hz, 1H), 3.93 (dd, *J* = 3.6, 10.5 Hz, 1H), 3.84 (d, *J* = 1.2 Hz, 1H), 3.76- 3.71 (m, 2H), 3.68-3.59 (m, 5H), 2.32 (s, 1H), 2.15 (s, 3H), 2.03 (s, 3H), 1.51 (dd, *J* = 1.8, 7.2 Hz), 1.21 (s, 18H), 1.20 (s, 9H), 1.13 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.6, 178.0, 176.8, 170.5, 170.0, 143.6, 136.3, 129.0, 128.9, 128.7, 101.8, 101.1, 99.9, 80.5, 79.0, 75.4, 74.9, 73.0, 72.8, 72.7, 72.6, 71.6, 70.3, 68.1, 66.7, 63.1, 39.0 (2C), 27.4, 27.2 (2C), 21.0, 20.8, 16.2, 12.4; **HRMS (ESI)**: *m/z* calcd. for $C_{47}H_{70}O_{20}Na$ ($M+Na^+$) 977.4352, found 977.4338; **FT-IR**: 3448, 2969, 1747, 1241, 1137, 1072 cm^{-1} ; **$[\alpha]_D^{25}$** = - 29.6 (c=0.55, $CHCl_3$); **Mp**: 207-209 °C.

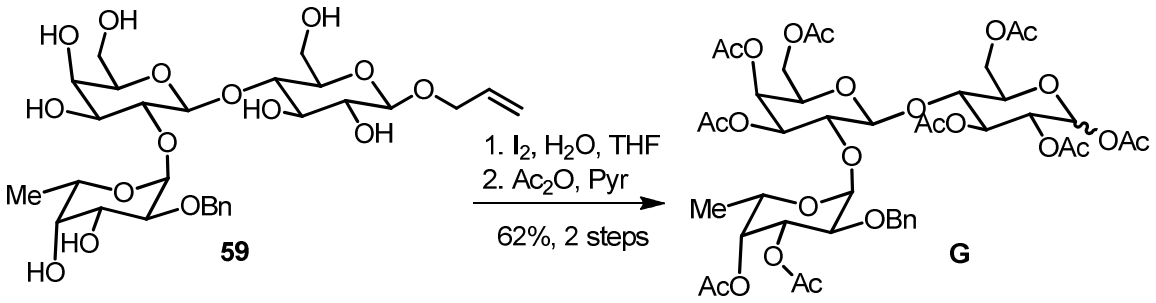
Synthesis of 2'-fucosyllactose trisaccharide vinylpolyol 59



To a solution of compound **F** (0.27 g, 0.28 mmol) in MeOH (1.5 mL) at room temperature was added 0.5 M solution of NaOMe in MeOH (7.6 mg, 0.14 mmol) and heated the reaction to 64 °C for 14 h. Added furthur 0.5 equiv of 0.5 M solution of NaOMe in MeOH (7.6 mg, 0.14 mmol) and continued heating for another 24 h. The reaction was cooled to room temperature, neutralized using amberlyst 120(H^+) resin, filtered and concentrated to obtain oil. Repeated

washing of the crude with hexanes followed by ether and decantation resulted in the formation of vinylpolyol **59** as a pale yellow solid (0.154 g, 88%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.39 (d, *J* = 7.8 Hz, 2H), 7.25 (t, *J* = 7.8 Hz, 2H), 7.18 (t, *J* = 7.2 Hz, 1H), 6.25 (d, *J* = 12.0 Hz, 1H), 5.45 (d, *J* = 3.0 Hz, 1H), 5.00 (dq, *J* = 7.2, 13.2 Hz, 1H), 4.79 (d, *J* = 11.4 Hz, 1H), 4.61 (d, *J* = 11.4 Hz, 1H), 4.41 (dd, *J* = 5.4, 7.2 Hz, 1H), 4.15 (q, *J* = 6.6 Hz, 1H), 3.81-3.78 (m, 2H), 3.73-3.68 (m, 3H), 3.65 (dd, *J* = 3.0, 9.6 Hz, 1H), 3.62- 3.57 (m, 4H), 3.49-3.47 (m, 1H), 3.41 (app t, *J* = 9.3, 10.3 Hz, 1H), 3.27-3.22 (m, 4H), 1.46 (d, *J* = 6.6 Hz, 3H), 1.11 (d, *J* = 6.0 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 146.0, 139.7, 129.6, 129.4, 128.8, 104.7, 103.5, 102.4, 98.7, 77.8, 77.2, 77.0, 76.6, 76.3, 76.2, 74.5, 73.8, 73.2, 71.0, 70.9, 67.6, 62.6, 61.5, 16.6, 12.5; **HRMS (ESI)**: *m/z* calcd. for $C_{28}H_{42}O_{15}Na$ ($M+Na^+$) 641.2415, found 641.2409; **FT-IR**: 3394, 2927, 2885, 1677, 1400, 1079, 1041 cm^{-1} ; **$[\alpha]_D^{25}$** = - 37.9 (c=0.315, $CHCl_3$); **Mp**: 131-133 °C.

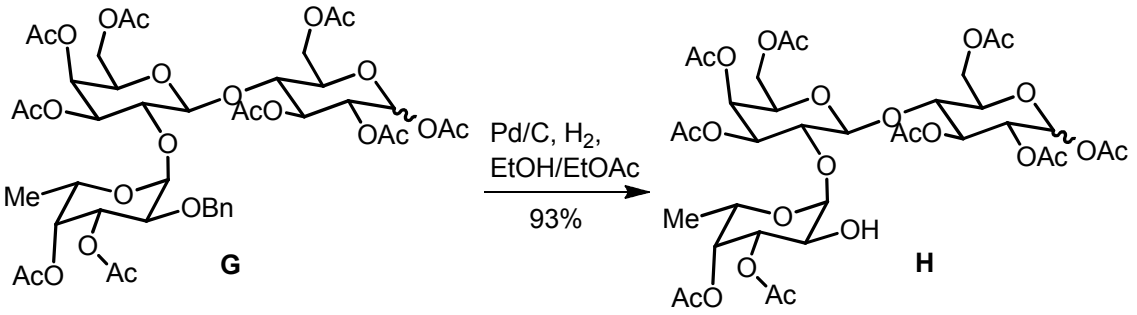
Synthesis of peracylated 2'-fucosyllactose trisaccharide G



To a solution of compound **59** (0.13 g, 0.210 mmol) in a mixture of THF (10.8 mL) and water (3.0 mL) (4:1) was added iodine (0.1 g, 0.42 mmol) at room temperature and the brown reaction mixture stirred for 30 min. The solvents were removed under vacuum and azeotroped the crude with toluene to remove water.

Redissolved the crude in pyridine (1.73 mL), added acetic anhydride (0.86 mL) DMAP (6 mg), and stirred the reaction at room temperature for 20 h. The reaction mixture was diluted with ethyl acetate and washed the organic layer with saturated $Na_2S_2O_3$, $NaHCO_3$, water and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 10% acetone in CH_2Cl_2 as eluent to obtain the peracylated compound **G** as a white solid (0.125 g, 62%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.34-7.31 (m, 3H), 7.28-7.25 (m, 6H), 6.29 (d, *J* = 3.6 Hz, 0.75H), 5.68 (d, *J* = 8.4, 1H), 5.41 (app t *J* = 10.2 Hz, 0.85H), 5.33 (d, *J* = 3.0, 8.1 Hz, 2H), 5.29 (dd, *J* = 3.6, 7.5 Hz, 2H), 5.25 (bs, 2H), 5.23 (app t, *J* = 9.6 Hz, 1H), 5.18-5.14 (m, 2H), 5.10-5.07 (m, 1H), 5.05-5.00 (m, 2H), 4.60 (app t, *J* = 11.4, 12.6 Hz, 2H), 4.56 (app t, *J* = 10.2, 11.1 Hz, 1H), 4.51-4.48 (m, 2H), 4.45-4.40 (m, 3H), 4.38-4.30 (m, 3H), 4.16-4.11 (m, 2H), 4.10-4.04 (m, 3H), 3.95-3.91 (m, 1H), 3.90-3.81 (m, 7H), 2.19 (s, 3H), 2.13-2.10 (m, 12H), 2.08-2.05 (m, 12H), 2.04 (s, 3H), 1.93 (s, 3H), 1.92 (s, 3H), 1.87 (s, 3H), 1.85 (s, 3H), 1.18 (d, *J* = 6.6 Hz, 2H), 1.14 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.7, 170.6, 170.5, 170.4, 170.3, 170.2, 170.1, 169.9, 169.8, 169.5, 169.1, 169.0, 138.1, 128.5, 127.9, 127.5, 101.1, 100.8, 97.5, 97.3, 91.8, 89.1, 74.0, 73.9, 73.6, 73.2, 73.0, 72.9, 72.7, 72.0, 71.7, 71.0, 70.8, 70.4, 61.9, 61.7, 61.2, 61.0, 21.1, 21.0 (2C), 20.9, 20.8, 20.7, 20.6 (2C), 15.8, 15.7; **HRMS (ESI)**: *m/z* calcd. for $C_{43}H_{56}O_{24}Na$ ($M+Na^+$) 979.3053, found 979.3047; **FT-IR**: 2942, 1751, 1369, 1222, 1045 cm^{-1} ; **Mp**: 94-96 ^oC.

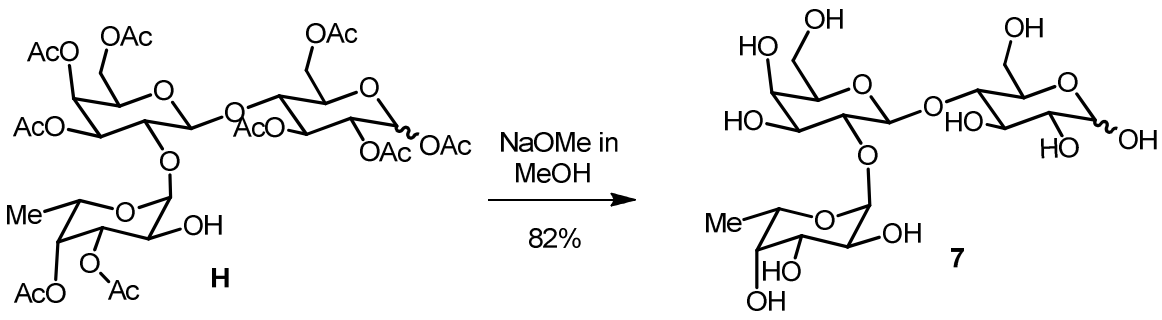
Synthesis of debenzylated 2'-fucosyllactose trisaccharide H



To a degassed solution of compound **G** (0.053 g, 0.055 mmol) in a mixture of ethanol (0.8 mL) and ethyl acetate (0.4 mL) was added 5% Pd/C (0.045 g). The reaction mixture was degassed again under argon and stirred under an atmosphere of hydrogen for 48 h. Filtered off the catalyst through a bed of celite, rinsed with ethanol and again filtered the filtrate through a Whatman filter. Removed the solvents under vacuum to obtain compound **H** as a white solid (0.045 g, 93%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 6.30 (d, *J* = 3.6 Hz, 0.75H), 5.68 (d, *J* = 8.4 Hz, 1H), 5.42 (app t *J* = 10.2 Hz, 0.84H), 5.35 (dd, *J* = 3.0, 6.6 Hz, 2H), 5.25-5.22 (m, 3H), 5.14 (dd, *J* = 4.2, 10.8 Hz, 2H), 5.09 (app t, *J* = 8.7 Hz, 1H), 5.04 (dd, *J* = 3.0, 10.2 Hz, 1H), 5.01-4.97 (m, 3H), 4.45-4.42 (m, 2H), 4.38-4.29 (m, 4H), 4.27 (q, *J* = 6.6 Hz, 1H), 4.17-4.12 (m, 2H), 4.09-4.06 (m, 2H), 3.91-3.80 (m, 8H), 2.18 (s, 3H), 2.15 (s, 3H), 2.13-2.10 (m, 12H), 2.08-2.06 (m, 12H), 2.04 (s, 3H), 2.02 (m, 12H), 1.19 (d, *J* = 6.6 Hz, 2H), 1.17 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.7 (2C), 170.6, 170.5 (2C), 170.2, 170.1 (2C), 169.5, 169.0, 100.9, 100.6, 100.1 (2C), 91.9, 89.1, 74.1, 73.9 (2C), 73.8, 73.0, 71.9, 71.4, 71.0 (2C), 70.6, 70.5, 70.4, 69.3, 69.2, 67.6, 67.3, 66.1, 66.0, 62.0, 61.9, 61.1, 60.9, 21.1, 21.0, 20.9, 20.8, 20.7, 20.6, 16.0, 15.8; **HRMS (ESI)**: *m/z*

calcd. for $C_{36}H_{50}O_{24}Na$ ($M+Na^+$) 889.2584, found 889.2588; **FT-IR**: 3486, 2942, 1747, 1373, 1226, 1076 cm^{-1} ; **Mp**: 114-116 ^oC.

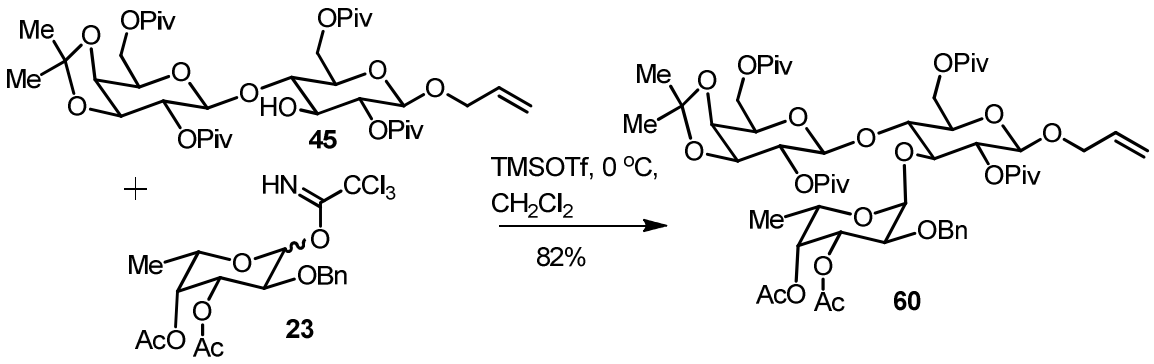
Synthesis of 2'-fucosyllactose (7)



To a solution of the peracylated compound **H** (0.042 g, 0.048 mmol) in methanol (4.2 mL) was added a 0.5 M solution of NaOMe in MeOH (0.005 mL, 0.0024 mmol) and the reaction stirred at room temperature for 24 h. Neutralized the reaction with Amberlite 120(H+) resin, filtered and removed the solvent under vacuum. The crude solid obtained was washed with ether and decanted. Dried under vacuum to obtain 2'-fucosyllactose **(7)** as a white solid (0.023 g, 82%). **^{1}H NMR** (600 MHz, $CDCl_3$) 5.31 (d, *J* = 3.6 Hz, 1H), 5.22 (d, *J* = 3.6 Hz, 0.5H), 4.64 (d, *J* = 7.8 Hz, 0.5H), 4.52 (d, *J* = 7.8 Hz, 1H), 4.26 (q, *J* = 6.6 Hz, 0.5H), 4.23 (q, *J* = 6.6 Hz, 0.5H), 3.95 (dd, *J* = 1.8, 12.0 Hz, 1H), 3.92- 3.85 (m, 3H), 3.82-3.79 (m, 5H), 3.77-3.65 (m, 6H), 3.60-3.57 (m, 1H), 3.48 (ddd, *J* = 1.8, 5.4, 9.9 Hz, 0.6H), 3.29 (dd, *J* = 9.3 Hz, 0.5H), 1.23 (d, *J* = 6.6 Hz, 1.5H), 1.22 (d, *J* = 6.6 Hz, 1.5H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 100.4, 100.3 99.4, 96.0, 91.9, 76.4, 76.0, 75.9, 75.4, 75.3, 74.4, 74.0, 73.7, 71.8, 71.4 (2C), 70.5, 69.7 (2C), 69.3, 68.3, 67.0, 61.2, 60.3, 60.1, 15.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{32}O_{15}Na$ ($M+Na^+$)

5111.1633, found 511.1636; $[\alpha]_D^{25}$ = - 48.1 (c=0.625, H_2O) after 72h $[\alpha]_D^{25}$ = - 49.3 (c=0.625, H_2O).

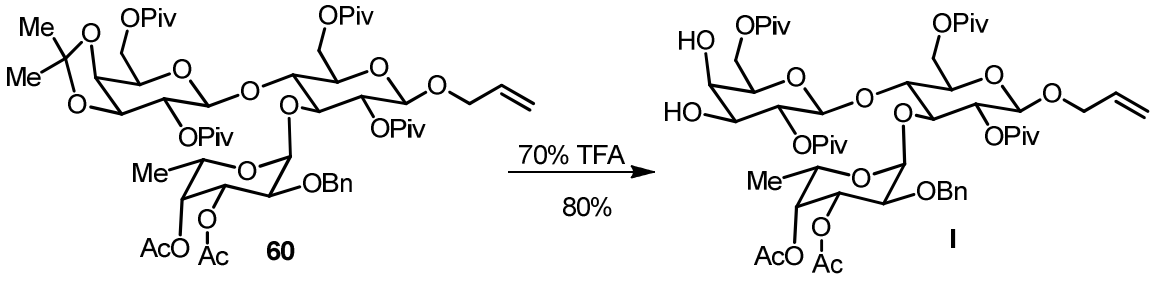
Synthesis of 3-fucosyllactose trisaccharide 60



To a solution of acceptor **45** (0.7 g, 0.92 mmol) in CH_2Cl_2 (3.5 mL) was added 0.1 M TMSOTf (2.0 mg, 0.009 mmol) in CH_2Cl_2 at 0 °C. A solution of donor **23** (0.89 g, 1.84 mmol) in CH_2Cl_2 (14 mL) was then added using a syringe pump over a period of 1 h. (Monitored the consumption of acceptor by TLC). The reaction was quenched by the addition of Et_3N (0.2 mL) and removed the solvents under vacuum. The crude was purified by flash chromatography using 25% ethyl acetate in hexanes as eluent to obtain the trisaccharide **60** as a white solid (0.81 g, 82%). Obtained also white solid as the byproduct (0.37 g, complex mixture of compound by ^{1}H NMR). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.34-7.29 (m, 3H), 7.25-7.23 (m, 2H), 5.84-5.78 (m, 1H), 5.37 (d, *J* = 3.6 Hz, 1H), 5.35 (d, *J* = 2.4 Hz, 1H), 5.31 (dd, *J* = 1.8, 10.5 Hz, 2H), 5.22 (dd, *J* = 1.8, 17.1 Hz, 1H), 5.16 (dd, *J* = 1.2, 11.1 Hz, 1H), 5.07 (dd, *J* = 7.8, 9.0 Hz, 1H), 4.96-4.91 (m, 2H), 4.60-4.56 (m, 3H), 4.46 (d, *J* = 11.4 Hz, 2H), 4.39 (d, *J* = 8.4 Hz, 1H), 4.28 (d, *J* = 9.0 Hz, 1H), 4.28-4.25 (m, 1H), 4.19 (dd, *J* = 4.8, 12.0 Hz, 1H), 4.11-4.05 (m, 3H), 3.97-3.93 (m, 2H), 3.87 (dd, *J* = 4.8, 10.5 Hz, 1H), 3.81 (app t, *J* = 9.6 Hz, 1H), 3.47 (dq, *J*

= 2.4, 9.6 Hz, 1H), 2.14 (s, 3H), 1.92 (s, 3H), 1.52 (s, 3H), 1.31 (s, 3H), 1.22 (2s, 18H), 1.21 (d, *J* = 6.0 Hz, 3H), 1.16 (s, 9H), 1.04 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.6, 178.1, 176.7, 170.7, 169.6, 138.3, 133.4, 128.2, 127.9, 127.5, 117.9, 111.0, 100.2, 97.1, 77.8, 74.7, 74.4 (2C), 74.1, 73.6, 73.5, 72.9, 72.7, 72.1, 71.7, 70.5, 70.1, 64.3, 62.6, 61.8, 39.1, 38.9, 38.8, 27.9, 27.4 (2C), 27.2, 27.0, 26.4, 21.0, 20.9, 16.3; **HRMS (ESI)**: *m/z* calcd. for $C_{55}H_{82}O_{21}Na$ ($M+Na^+$) 1101.5300 found 1101.5300; **FT-IR**: 2973, 1743, 1241, 1133, 1045 cm^{-1} ; **$[\alpha]_D^{25}$** = - 14.7 (c=0.955, $CHCl_3$); **Mp**: 96-98 °C.

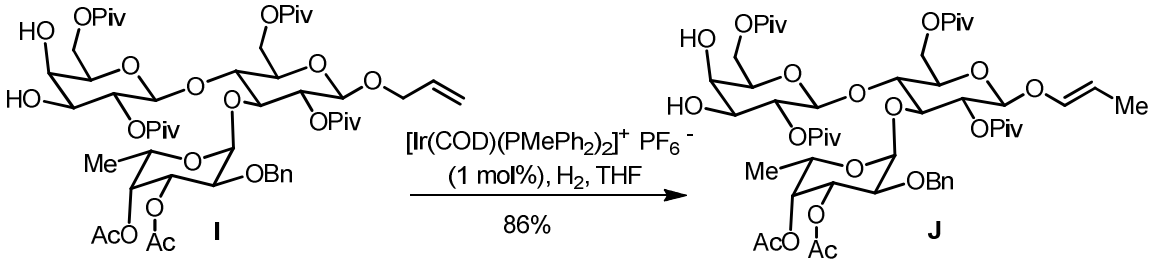
Synthesis of 3-fucosyllactose trisaccharide diol I



To a solution of trisaccharide **60** (0.5 g, 0.5 mmol) in CH_2Cl_2 (20 mL) at 0 °C was added 70% trifluoroacetic acid (2 mL) and the reaction stirred for 2 h followed by 2 h at room temperature. The reaction mixture was diluted with water and the aqueous was extracted with CH_2Cl_2. The organic layer was washed with saturated $NaHCO_3$, dried over $MgSO_4$, filtered and concentrated to obtain the diol **I** as white solid (0.38 g, 80%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.32-7.29 (m, 4H), 7.25-7.23 (m, 1H), 5.85-5.78 (m, 1H), 5.33 (d, *J* = 3.0 Hz, 1H), 5.26 (dd, *J* = 3.0, 10.8 Hz, 1H), 5.23 (dd, *J* = 1.2, 18.0 Hz, 1H), 5.16 (d, *J* = 10.2 Hz, 1H), 5.12 (app t, *J* = 7.8 Hz, 1H), 4.93 (app t, *J* = 7.8 Hz, 1H), 4.88 (q, *J* = 6.6 Hz, 1H), 4.59 (d, *J* = 11.4 Hz, 3H), 4.50-4.39 (m, 6H), 4.26 (dd, *J* = 4.8, 12.6 Hz, 2H), 4.19 (dd, *J* =

4.8, 12.0 Hz, 1H), 4.07 (app t, *J* = 9.6 Hz, 1H), 3.97 (dd, *J* = 6.6, 12.0 Hz, 1H), 3.94 (app t, *J* = 9.6 Hz, 1H), 3.90-3.86 (m, 2H), 3.61 (app t, *J* = 6.6 Hz, 1H), 3.57-3.51 (m, 2H), 3.46 (d, *J* = 3.6 Hz, 1H), 2.69 (d, *J* = 9.6, 1H), 2.12 (s, 3H), 1.93 (s, 3H), 1.72 (s, 1H), 1.24 (s, 9H), 1.21 (s, 9H), 1.18 (s, 9H), 1.18 (d, *J* = 6.6 Hz, 3H), 1.12 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.5, 178.1, 178.0, 176.4, 170.9, 170.8, 138.3, 133.5, 128.3, 127.9, 127.6, 117.9, 100.3, 100.1, 96.9, 74.4, 73.8, 73.6, 73.5 73.1 (2C), 72.8, 72.1, 72.0, 70.7, 70.0, 68.5, 64.5, 62.2, 39.1, 39.0 (2C), 38.8, 27.4, 27.3, 27.2, 21.1, 20.9, 16.0; **HRMS (ESI)**: *m/z* calcd. for $C_{52}H_{78}O_{21}Na$ ($M+Na^+$) 1061.4927 found 1061.4927; **FT-IR**: 3486, 2973, 1739, 1276, 1145, 1045 cm^{-1} ; **$[\alpha]_D^{25}$** = - 25.9 (c=0.83, $CHCl_3$); **Mp**: 98-100 °C.

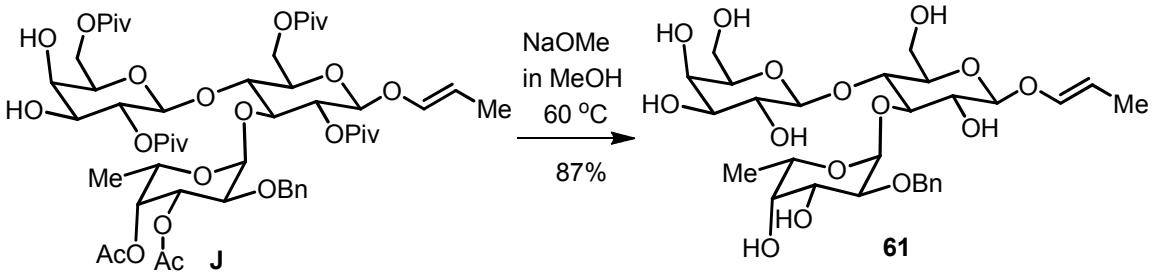
Synthesis of 3-fucosyllactose trisaccharide vinylether J



A solution of the catalyst $Ir(COD)(PMePh_2)_2]^+PF_6^-$ (2.8 mg, 0.0033 mmol) in THF (0.56 mL) was degassified and then stirred for 15 min under an athmosphere of hydrogen (red color of the solution decolorised). This was then cannulated into a solution of compound **I** (0.35 g, 0.36 mmol) in THF (7.7 mL) at room temperature and stirred for 30 min. The reaction was diluted with saturated $NaHCO_3$ and the aqueous was extracted with hexanes, dried over $MgSO_4$, filtered and concentrated to obtain the compound **J** as white solid (0.3 g, 86%). The compound was carried forward without any further purification. **^{1}H NMR** (600

MHz, $CDCl_3$) δ 7.31-7.30 (m, 4H), 7.26-7.24 (m, 1H), 6.12 (dd, *J* = 1.8, 12.3 Hz, 1H), 5.32 (m, 1H), 5.31 (d, *J* = 3.6 Hz, 1H), 5.25 (dd, *J* = 3.6, 10.5 Hz, 1H), 5.16 (app t, *J* = 7.8 Hz, 1H), 5.09-5.03 (m, 1H), 4.93 (app t, *J* = 9.0 Hz, 1H), 4.87 (q, *J* = 6.0, 7.2 Hz, 1H), 4.61 (d, *J* = 11.4 Hz, 1H), 4.56 (d, *J* = 7.2 Hz, 1H), 4.49-4.41 (m, 6H), 4.21 (dd, *J* = 5.4, 12.0 Hz, 1H), 4.08 (app t, *J* = 9.0 Hz, 1H), 3.97 (app t, *J* = 9.6 Hz, 1H), 3.62-3.53 (m, 3H), 3.46 (d, *J* = 3.0 Hz, 1H), 2.70 (d, *J* = 9.6, 1H), 2.11 (s, 3H), 1.93 (s, 3H), 1.72 (s, 1H), 1.51 (d, *J* = 6.6, 3H), 1.23 (s, 9H), 1.22 (s, 9H), 1.18 (s, 9H), 1.18 (d, *J* = 6.0 Hz, 3H), 1.13 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 178.5, 178.0, 176.4, 170.9, 170.8, 143.5, 138.3, 128.3, 127.9, 127.6, 105.1, 100.2, 100.0, 96.9, 74.1, 73.8, 73.5, 73.3 73.1 (2C), 72.8, 72.2, 71.9, 70.7, 68.4, 64.6, 62.2, 61.9, 39.0, 38.9, 27.4, 27.3 (2C), 27.2, 21.1, 20.9, 16.0, 12.4; **HRMS (ESI)**: *m/z* calcd. for $C_{52}H_{78}O_{21}$ ($M+Na^+$) 1061.4927 found 1061.4919; **FT-IR**: 3486, 2969, 1739, 1276, 1141, 1076 cm^{-1} ; **$[\alpha]_D^{25}$** = - 26.3 (c=0.96, $CHCl_3$); **Mp**: 100-102 °C.

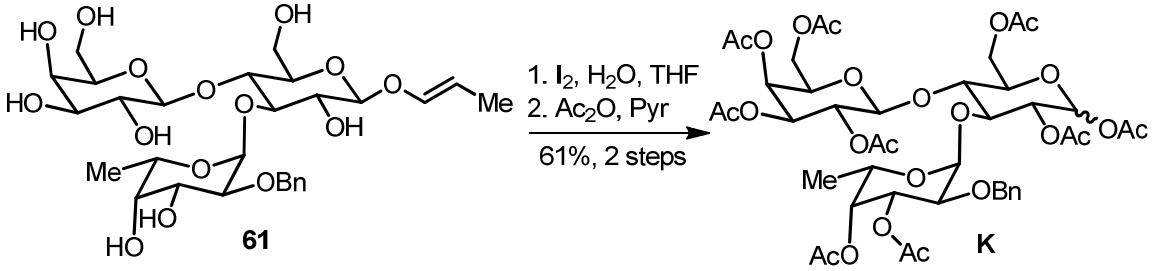
Synthesis of 3-fucosyllactose trisaccharide vinylether polyol 61



To a solution of compound **J** (0.27 g, 0.28 mmol) in MeOH (1.5 mL) at room temperature was added 0.5 M solution of NaOMe in MeOH (7.6 mg, 0.14 mmol) and heated the reaction to 60 °C for 14 h. Added furthur 0.5 equiv of 0.5 M solution of NaOMe in MeOH (7.6 mg, 0.14 mmol) and continued heating for

another 24 h. The reaction was cooled to room temperature, neutralized using amberlyst 120(H^+) resin, filtered and concentrated to obtain oil. Repeated washing of the crude with hexanes followed by ether and decantation gave the polyol **61** as a pale yellow solid (0.14 g, 87%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.48 (d, *J* = 7.2 Hz, 2H), 7.33 (app t, *J* = 7.2 Hz, 2H), 7.26 (app t, *J* = 7.2 Hz, 1H), 6.34 (dd, *J* = 1.2, 12.0 Hz, 1H), 5.70 (d, *J* = 3.6 Hz, 1H), 5.14-5.09 (m, 1H), 4.89 (d, *J* = 11.4 Hz, 1H), 4.66 (d, *J* = 11.4 Hz, 1H), 4.50 (dd, *J* = 7.8 Hz, 1H), 4.40 (d, *J* = 7.2 Hz, 1H), 3.93-3.89 (m, 3H), 3.82-3.79 (m, 4H), 3.75 (dd, 10.5, 1.8 Hz, 1H). 3.65 (dd, *J* = 3.6, 10.6 Hz, 1H), 3.60 (dd, *J* = 4.2, 12.0 Hz, 1H), 3.56 (app t, *J* = 8.4 Hz, 1H), 3.50-3.47 (m, 2H), 3.31-3.30 (m, 1H), 1.56 (d, *J* = 6.6 Hz, 3H), 1.16 (d, *J* = 6.0 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 144.0, 137.8, 127.7, 127.4, 126.8, 102.8, 102.0, 101.9, 95.9, 76.1, 75.7, 75.1, 74.5, 72.9, 72.2, 71.9, 71.0, 70.7, 68.3, 68.1, 65.2, 61.2, 59.2, 14.6, 10.5; **HRMS (ESI)**: *m/z* calcd. for $C_{28}H_{42}O_{15}Na$ ($M+Na^+$) 641.2415, found 641.2417; **FT-IR**: 3394, 2935, 2885, 1454, 1087 cm^{-1} ; **$[\alpha]_D^{25}$** = - 75.8 (c=0.69, $CHCl_3$); **Mp**: 142-144 ^oC.

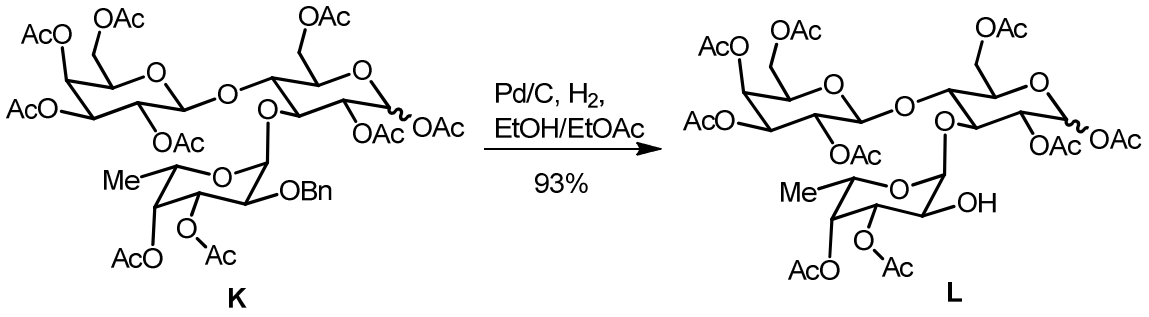
Synthesis of peracylated 3-fucosyllactose trisaccharide K



To a solution of compound **61** (0.12 g, 0.193 mmol) in a mixture of THF (10.0 mL) and water (2.8 mL) (4:1) was added iodine (0.1 g, 0.38 mmol) at room temperature and the brown reaction mixture stirred for 30 min. Removed the

solvents under vacuum and azeotroped the crude with toluene to remove water. The crude was dissolved in pyridine (1.6 mL), added acetic anhydride (0.8 mL) DMAP (5 mg), stirred the reaction at room temperature for 20 h. The reaction mixture was diluted with ethyl acetate and washed the organic layer with saturated $Na_2S_2O_3$, $NaHCO_3$, water and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 10% acetone in CH_2Cl_2 as eluent to obtain compound **K** as a white solid (0.11 g, 61%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.34-7.23 (m, 9H), 6.28 (d, *J* = 3.6 Hz, 0.7H), 5.60 (d, *J* = 7.8 Hz, 1H), 5.40-5.39 (m, 2H), 5.37-5.36 (m, 2H), 5.27 (d, *J* = 4.2 Hz, 2H), 5.24-5.20 (m, 3H), 5.13-5.05 (m, 3H), 5.00-4.97 (m, 3H), 4.91(q, *J* = 6.0, 7.2 Hz, 1H), 4.63-4.54 (m, 6H), 4.53 (dd, *J* = 4.8, 10.2 Hz, 2H), 4.48 (dd, *J* = 7.2, 14.4 Hz, 2H), 4.33 (app t, *J* = 7.8 Hz, 1H), 4.31 (app t, *J* = 7.8 Hz, 1H), 4.19 (app t, *J* = 7.8 Hz, 1H), 4.13-4.09 (m, 2H), 4.07-4.04 (m, 2H), 4.00 (app t, *J* = 6.6 Hz, 1H), 3.93-3.90 (m, 2H), 3.89-3.85 (m, 3H), 3.65 (bd, 1H), 2.20-2.13 (m, 19H), 2.08-2.05 (3s, 9H), 2.03 (s, 6H), 1.98-1.97 (3s, 8H), 1.91-1.90 (2s, 5H), 1.86 (s, 2H), 1.26 (d, *J* = 6.0 Hz, 3H), 1.22 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.8, 170.6, 170.4, 170.1, 169.9, 169.4, 169.3, 169.2, 169.1, 168.8, 138.0, 128.5 (2C), 128.2, 128.5, 127.6, 100.7, 100.5, 97.7, 97.4, 92.1, 89.2, 74.5, 74.0, 73.6, 73.5, 73.2, 73.0, 72.9, 72.0 (2C), 71.6, 71.5, 71.3, 71.2, 71.1 (2C), 70.5 (2C), 69.2, 69.1, 66.8 (2C), 66.8, 64.5, 61.6, 61.2, 61.0, 60.9, 21.1, 20.9, 20.8, 20.7, 16.0; **HRMS (ESI)**: *m/z* calcd. for $C_{43}H_{56}O_{24}Na$ ($M+Na^+$) 979.3053, found 979.3064; **FT-IR**: 2977, 1751, 1373, 1226, 1049 cm^{-1} ; **Mp**: 110-112 ^oC.

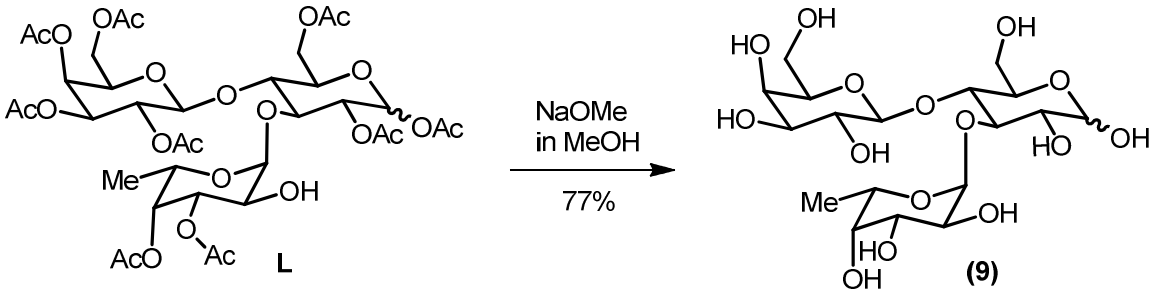
Synthesis of debenzylated 3-fucosyllactose trisaccharide L



To a degassed solution of compound **K** (0.06 g, 0.062 mmol) in a mixture of ethanol (0.48 mL) and ethyl acetate (0.24 mL) was added 5% Pd/C (0.02 g). The reaction mixture was degassed again under argon and stirred under an atmosphere of hydrogen for 48 h. Filtered off the catalyst through a bed of celite, rinsed with ethanol and again filtered the filtrate through a Whatman filter. The solvents were removed under vacuum to obtain compound **L** as a white solid (0.047 g, 88%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 6.17 (d, *J* = 3.6 Hz, 0.7H), 5.60 (d, *J* = 8.4, 1H), 5.43-5.42 (m, 1.8H), 5.33-5.30 (m, 2H), 5.17-5.05 (m, 7H), 4.97 (dt, *J* = 3.0, 10.5 Hz, 2H), 4.89-4.84 (m, 2H), 4.69 (dd, *J* = 6.6, 11.7 Hz, 1H), 4.66 (dd, *J* = 7.8, 11.7 Hz, 1H), 4.58 (dd, *J* = 1.8, 12.0 Hz, 1H), 4.56 (dd, *J* = 1.8, 12.3 Hz, 1H), 4.44 (app t, *J* = 8.4 Hz, 2H), 4.15 (app t, *J* = 9.6 Hz, 1H), 3.96-3.89 (m, 4H), 3.84-3.80 (m, 4H), 3.68-3.66 (m, 1H), 2.20-2.13 (3s, 8H), 2.15-2.14 (2s, 11H), 2.02-1.97 (4s, 11H), 1.26 (d, *J* = 6.6 Hz, 2.75H), 1.23 (d, *J* = 6.6 Hz, 3.3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 170.9, 170.8, 170.7, 170.6 (2C), 170.3, 170.2, 170.1, 169.8, 169.3, 169.2 (2C), 169.0, 100.8, 99.7, 99.6, 91.6, 89.5, 75.3, 74.2, 73.9, 73.8, 72.3, 72.2, 72.1, 71.9, 71.8 (2C), 71.1, 71.0, 70.9, 68.9, 68.8, 66.9, 66.8, 65.2, 65.0, 61.5, 61.3, 21.1, 21.0, 20.8 (2C), 20.7 (2C), 16.0 (2C); **HRMS**

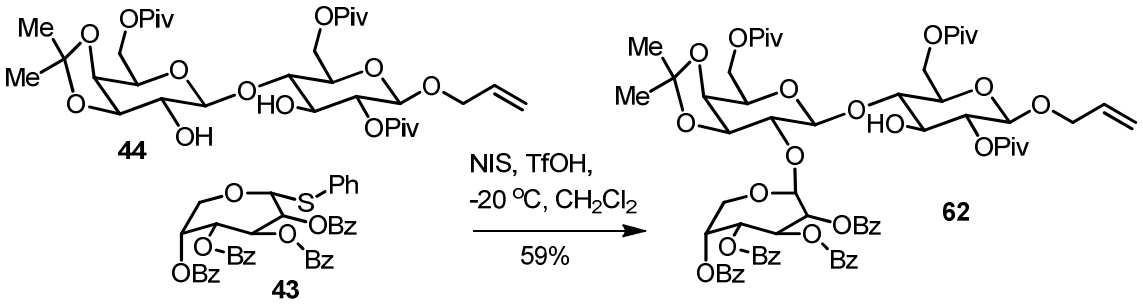
(ESI): *m/z* calcd. for $C_{36}H_{50}O_{24}Na$ ($M+Na^+$) 889.2584, found 889.2572; **FT-IR**: 3494, 2962, 1747, 1373, 1226, 1045 cm^{-1} ; **Mp**: 134-136 ^oC.

Synthesis of 3-fucosyllactose (9)



To a solution of the peracylated compound **L** (0.023g, 0.026 mmol) in methanol (2.3 mL) was added a 0.5 M solution of NaOMe in MeOH (0.0026 mL, 0.0013 mmol) and the reaction stirred at room temperature for 24 h. Neutralized the reaction with amberlite 120(H^+) resin, filtered and removed the solvent under vacuum. The crude solid obtained was washed with ether and decanted. Dried under vacuum to obtain 3-fucosyllactose **(9)** as a white solid (0.01 g, 77%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 5.44 (d, *J* = 3.6 Hz, 1H), 5.38 (d, *J* = 3.6 Hz, 1H), 5.18 (d, *J* = 3.6 Hz, 1H), 4.84-4.81 (m, 3H), 4.65 (d, *J* = 7.8 Hz, 1H), 4.43 (d, *J* = 7.8 Hz, 2H), 3.98-3.93 (m, 5H), 3.90-3.86 (m, 6H), 3.84-3.70 (m, 14H), 3.66-3.64 (m, 3H), 3.62-3.59 (m, 3H), 3.50-3.44 (m, 3H), 1.19 (d, *J* = 6.0 Hz, 3H), 1.18 (d, *J* = 6.6 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 100.4, 100.3 99.4, 96.0, 91.9, 76.4, 76.0, 75.9, 75.4, 75.3, 74.4, 74.0, 73.7, 71.8, 71.4 (2C), 70.5, 69.7 (2C), 69.3, 68.3, 67.0, 61.2, 60.3, 60.1, 15.3; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{32}O_{15}Na$ ($M+Na^+$) 511.1633, found 511.1639; **$[\alpha]_D^{25}$** = - 29.8 (c=0.45, H_2O) after 92 h **$[\alpha]_D^{25}$** = - 22.1 (c=0.45, H_2O).

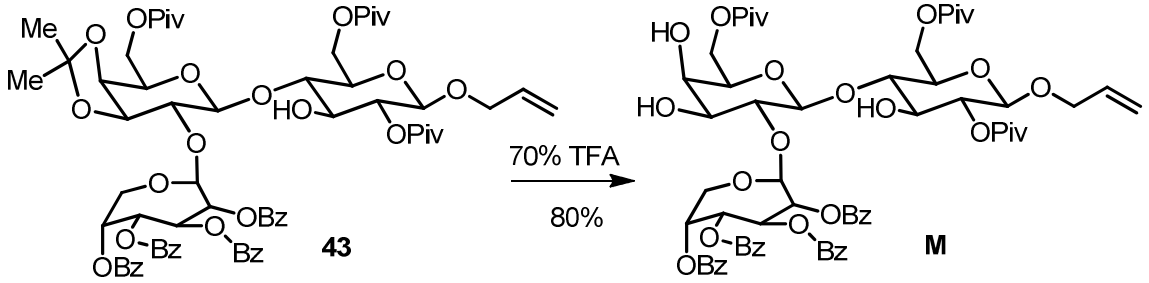
Synthesis of 2'-glucoseptanosyllactose trisaccharide 62



To a -25 °C cooled solution of donor **43** (0.36 g, 0.52 mmol), acceptor **44** (0.32 g, 0.475 mmol) and 4Å MS (1.35 g) in CH_2Cl_2 (4.8 mL) was added NIS (0.1 g, 0.475 mmol) and TfOH (0.018 g, 0.118 mmol) and the reaction stirred for 30 min and then at 0 °C for 30 min. The reaction was quenched with Et_3N (0.45 mL) at 0 °C. Filtered off the inorganics through celite and removed the solvent under vacuum. The crude was purified by flash chromatography using 30% ethyl acetate in hexanes as eluent to obtain the trisaccharide **62** as a white solid (0.35 g, 59%). Recovered 0.085 g of the donor **43** and 0.081 g of the acceptor **44**). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 8.17 (d, *J* = 7.8 Hz, 2H), 7.88 (d, *J* = 8.2 Hz, 2H), 7.81 (d, *J* = 7.8 Hz, 2H), 7.77 (d, *J* = 7.8 Hz, 1H), 7.62 (app t, *J* = 7.6 Hz, 1H), 7.50 (app t, *J* = 7.6 Hz, 2H), 7.44 (app t, *J* = 7.2 Hz, 1H), 7.39 (app t, *J* = 7.6 Hz, 1H), 7.33 (app t, *J* = 7.6 Hz, 1H), 7.27 (app t, *J* = 7.6 Hz, 3H), 7.21 (app t, *J* = 8.0 Hz, 2H), 7.18 (app t, *J* = 7.6 Hz, 2H), 6.26 (dd, *J* = 9.6 Hz, 1H), 6.02 (dd, *J* = 3.2, 9.8 Hz, 1H), 5.90-5.80 (m, 1H), 5.77-5.68 (m, 3H), 5.26 (dd *J* = 1.2, 17.4 Hz, 1H), 5.11 (dd, *J* = 1.2, 10.4 Hz, 1H), 5.01 (dd, *J* = 1.6, 9.4 Hz, 1H), 4.89 (dd, *J* = 1.6, 9.4 Hz, 1H), 4.66 (dd, *J* = 2.4, 13.6 Hz, 1H), 4.49 (d, *J* = 8.0 Hz, 1H), 4.36-4.29 (m, 2H), 4.23-4.18 (m, 2H), 4.09-3.96 (m, 2H), 3.87 (s, 1H), 3.84 (dd, *J* = 6.4, 8.0 Hz, 2H), 3.77-3.71 (m, 2H), 3.33 (app t, *J* = 9.2 Hz, 1H), 1.29 (s, 9H), 1.19 (s, 9H), 1.18 (s, 9H);

^{13}C NMR (150 MHz, $CDCl_3$) δ 178.6, 178.4, 176.8, 165.9, 165.7, 165.1, 164.8, 133.5, 133.2, 133.1, 130.1, 129.9, 129.7, 129.6, 129.3, 129.1, 128.7, 128.3, 128.2, 117.7, 110.9, 101.9, 99.6, 99.2, 82.8, 79.5, 75.4, 74.0, 73.5, 73.2, 72.7, 72.6, 72.3, 71.7, 70.0, 68.9, 63.2, 39.1, 39.0, 38.9, 28.0, 27.4, 27.3, 27.2, 26.2; **HRMS (ESI)**: *m/z* calcd. for $C_{67}H_{80}O_{23}Na$ ($M+Na^+$) 1275.4982, found 1275.4993; **FT-IR**: 3475, 2969, 1731, 1265, 1149, 1095, 709 cm^{-1} ; **$[\alpha]_D^{25}$** = - 59.5 (c=0.815, $CHCl_3$).

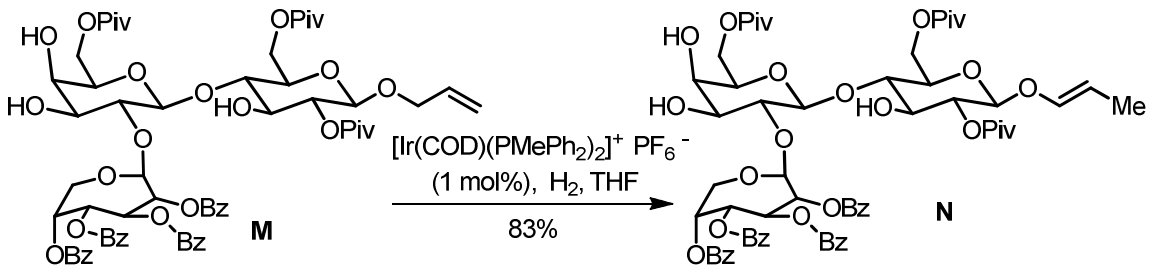
Synthesis of 2'-glucoseptanosyllactose trisaccharide diol M



To a solution of trisaccharide **43** (0.325 g, 0.25 mmol) in CH_2Cl_2 (13 mL) at 0 ^oC was added 70% trifluoroacetic acid (1.3 mL) and the reaction stirred for 2 h followed by 2 h at room temperature. The reaction mixture was diluted with water and the aqueous was extracted with CH_2Cl_2. The organic layer was washed with saturated $NaHCO_3$ solution, dried over $MgSO_4$, filtered and concentrated to obtain compound **M** as a white solid (0.25 g, 80%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 8.16 (d, *J* = 7.8 Hz, 2H), 7.87 (d, *J* = 7.8 Hz, 2H), 7.77 (d, *J* = 7.8 Hz, 2H), 7.75 (d, *J* = 7.2 Hz, 2H), 7.61 (app t, *J* = 7.8 Hz, 1H), 7.50 (app t, *J* = 7.8 Hz, 2H), 7.43 (app t, *J* = 6.6 Hz, 1H), 7.37 (app t, *J* = 7.2 Hz, 1H), 7.32 (app t, *J* = 7.2 Hz, 1H), 7.27 (app t, *J* = 7.2 Hz, 3H), 7.19 (app t, *J* = 7.8 Hz, 2H), 7.16 (app t, *J* = 7.2 Hz, 2H), 6.26 (app t, *J* = 9.0 Hz, 1H), 6.04 (dd, *J* = 3.2, 9.9 Hz, 1H), 5.89-5.72 (m,

4H), 5.27 (d *J* = 17.4 Hz, 1H), 5.20 (d, *J* = 10.8 Hz, 1H), 4.99 (d, *J* = 11.4, Hz, 1H), 4.89 (app t, *J* = 8.4 Hz, 1H), 4.69 (d, *J* = 13.8 Hz, 1H), 4.49 (d, *J* = 8.4 Hz, 1H), 4.41-4.38 (m, 2H), 4.32 (dd, *J* = 3.6, 13.2 Hz, 1H), 4.25 (d, *J* = 7.8 Hz, 1H), 4.16 (dd, *J* = 6.6, 11.4 Hz, 1H), 4.10-4.02 (m, 3H), 3.89 (app t *J* = 8.4 Hz, 1H), 3.74-3.60 (m, 6H), 3.38 (app t, *J* = 9.6 Hz, 1H), 3.00 (bs, 1H), 1.32 (s, 9H), 1.21 (s, 9H), 1.17 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 179.3, 178.7, 166.0, 165.7, 165.5, 165.1, 133.7, 133.6, 133.3, 133.2, 133.1, 130.1, 129.8 (2C), 129.7, 129.4, 129.2, 129.1, 128.7, 128.6, 128.4, 128.3, 128.2, 117.8, 102.4, 100.6, 99.6, 74.7, 74.3, 74.2, 73.2, 72.8, 72.7, 72.6, 72.3, 70.8, 70.1, 68.8, 68.5, 63.4, 63.0, 62.3, 39.1, 39.0, 38.9, 27.4, 27.2 (2C); **HRMS (ESI)**: *m/z* calcd. for $C_{64}H_{76}O_{23}Na$ ($M+Na^+$) 1235.4669, found 1235.4690; **FT-IR**: 3475, 2969, 1727, 1268, 1153, 1095, 709 cm^{-1} ; **$[\alpha]_D^{25}$** = - 65.2 (c=1.15, $CHCl_3$).

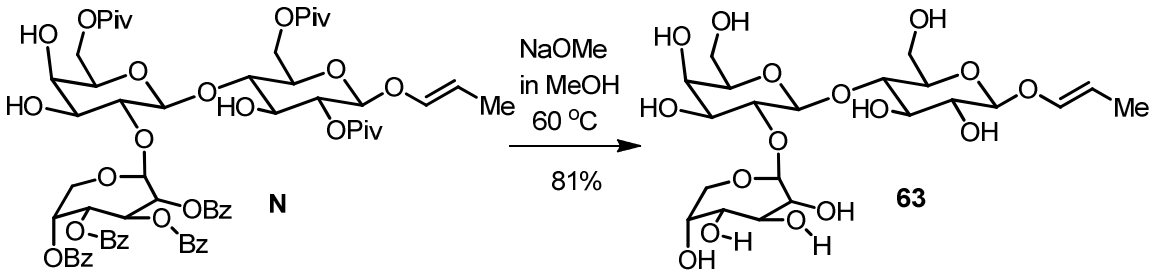
Synthesis of 2'-glucoseptanosyllactose trisaccharide vinylether N



A solution of the catalyst $Ir(COD)(PMePh_2)_2]^+PF_6^-$ (1.7 mg, 0.0019 mmol) in THF (0.4 mL) was degassified and then stirred for 15 min under an athmosphere of hydrogen (red color of the solution decolorised). This was then cannulated into a solution of compound **M** (0.24 g, 0.19 mmol) in THF (4.8 mL) at room temperature and stirred for 30 min. The reaction was diluted with saturated $NaHCO_3$ and the aqueous was extracted with hexanes. The organic layer was

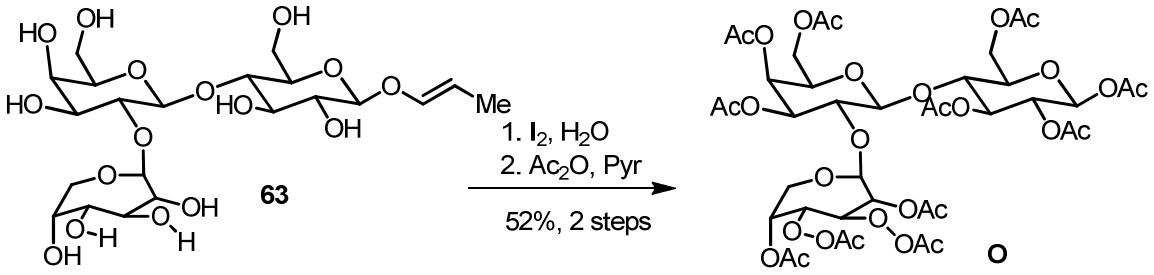
dried over $MgSO_4$, filtered and concentrated to obtain compound **N** as white solid (0.2 g, 83%). The compound was carried forward without any further purification. **^{1}H NMR** (600 MHz, $CDCl_3$) δ 8.17 (d, *J* = 7.8 Hz, 2H), 7.87 (d, *J* = 7.8 Hz, 2H), 7.79 (d, *J* = 7.8 Hz, 2H), 7.75 (d, *J* = 7.2 Hz, 2H), 7.62 (dd, *J* = 7.2 Hz, 1H), 7.50 (app t, *J* = 7.2 Hz, 2H), 7.43 (app t, *J* = 7.8 Hz, 1H), 7.38 (app t, *J* = 7.2 Hz, 1H), 7.32 (app t, *J* = 7.2 Hz, 1H), 7.29-7.27 (m, 3H), 7.20 (app t, *J* = 7.8 Hz, 2H), 7.17 (app t, *J* = 7.8 Hz, 1H), 6.27 (app t, *J* = 9.6 Hz, 1H), 6.18 (d, *J* = 12.0 Hz, 1H), 6.04 (dd, *J* = 3.0, 9.9 Hz, 1H), 5.84 (d, *J* = 7.2 Hz, 1H), 5.79 (d, *J* = 3.0 Hz, 1H), 5.74 (app t, *J* = 8.4 Hz, 1H), 5.14-5.08 (m, 1H), 4.99-4.93 (m, 2H), 4.69 (d, *J* = 12.6 Hz, 1H), 4.64 (d, *J* = 8.4 Hz, 1H), 4.40 (dd, *J* = 6.0, 12.0 Hz, 1H), 4.25 (d, *J* = 7.2 Hz, 1H), 4.15 (dd, *J* = 7.2, 12.0 Hz, 1H), 4.10 (dd, *J* = 7.8, 11.7 Hz, 1H), 4.05 (dd, *J* = 3.6, 14.1 Hz, 1H), 3.89 (app t, *J* = 8.4 Hz, 1H), 3.81-3.69 (m, 4H), 3.65-3.59 (m, 2H), 3.40 (dd, *J* = 9.6 Hz, 1H), 2.99 (dd, *J* = 7.2 Hz, 1H), 1.54 (d, *J* = 6.6 Hz, 3H), 1.32 (s, 9H), 1.21 (s, 9H), 1.17 (s, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 179.3, 178.7, 177.0, 165.9, 165.7, 165.5, 165.1, 143.8, 133.6, 133.3, 133.2, 133.1, 130.2, 129.8 (2C), 129.7, 129.4, 129.2, 129.1, 128.7, 128.4, 128.3, 128.2, 104.8, 102.4, 100.6, 99.7, 82.0, 74.7, 74.3, 74.1, 73.1, 72.9, 72.8, 72.3, 72.2, 70.8, 68.8, 68.5, 63.4, 63.1, 62.3, 39.1, 39.0 (2C), 30.5, 27.4, 27.2 (2C), 12.4; **HRMS (ESI)**: *m/z* calcd. for $C_{64}H_{76}O_{23}Na$ ($M+Na^+$) 1235.4675, found 1235.4703 **FT-IR**: 3475, 2969, 1731, 1268, 1149, 1095, 709 cm^{-1} ; **$[\alpha]_D^{25}$** = - 60.1 (c=1.13, $CHCl_3$).

Synthesis of 2'-glucoseptanosyllactose trisaccharide vinylether polyol 63



To a solution of compound **N** (0.2 g, 0.16 mmol) in MeOH (0.9 mL) at room temperature was added 0.5 M solution of NaOMe in MeOH (4.4 mg, 0.082 mmol) and heated the reaction to 60 °C for 14 h. Added furthur 0.5 equiv of 0.5 M solution of NaOMe in MeOH (4.4 mg, 0.082 mmol), and continued heating for another 24 h. The reaction was cooled to room temperature, neutralized using amberlyst 120(H^+) resin, filtered and concentrated to obtain oil. Repeated washing of the crude with hexanes followed by ether and decantation gave compound **63** as a pale yellow solid (0.072 g, 81%).

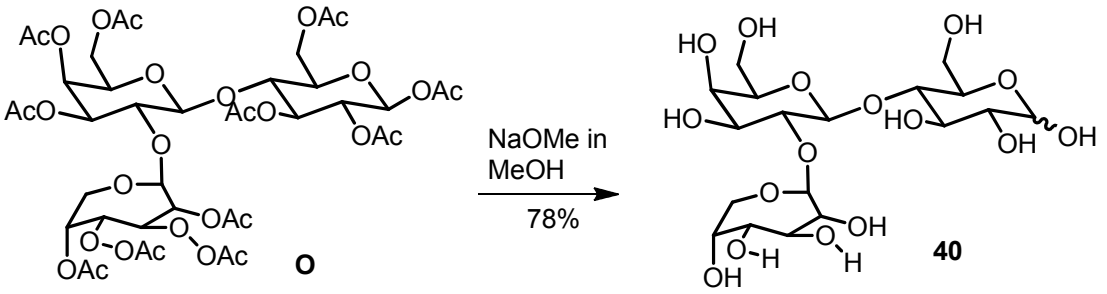
Synthesis of peracylated 2'-glucoseptanosyllactose trisaccharide O



Peracylated trisaccharide: To a solution of the polyol compound **63** (0.072 g, 0.13 mmol) in a mixture of THF (6.0 mL) and water (1.7 mL) was added iodine (0.067 g, 0.26 mmol) at room temperature and the brown reaction mixture stirred for 1 h. Removed the solvents under vacuum and azeotroped the crude with

toluene to remove water. Redissolved the crude in pyridine (2 mL), added acetic anhydride (1 mL), DMAP (5 mg), stirred the reaction at room temperature for 20 h. The reaction mixture was diluted with ethyl acetate and the organic layer was washed with saturated $Na_2S_2O_3$, $NaHCO_3$, water and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated. The crude was purified by flash chromatography using 10% acetone in CH_2Cl_2 as eluent to obtain peracylated compound **O** as a white solid (0.062 g, 52%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 6.29 (d, *J* = 3.6 Hz, 0.5H), 5.68 (d, *J* = 8.4 Hz, 1H), 5.47 (app t, *J* = 8.4 Hz, 1.5H), 5.41-5.30 (m, 5H), 5.26 (dd, *J* = 3.6, 8.1 Hz, 2H), 5.20-5.16 (m, 2H), 5.13-5.10 (m, 1H), 5.08-5.01 (m, 2.5H), 5.00-4.94 (m, 3.5H), 4.86-4.83 (m, 0.8H), 4.49-4.44 (m, 0.7H), 4.34-4.20 (m, 4H), 4.15-4.01 (m, 6.5H), 4.00 (dd, *J* = 7.2, 12.0 Hz, 1.5H), 3.96-3.91 (m, 1H), 3.89-3.79 (m, 5H), 3.78-3.64 (m, 4H), 2.19 (s, 1.5H), 2.17(s, 5H), 2.15 (s, 4H), 2.13-2.10 (app m, 12H), 2.07-2.06 (2s, 9H), 2.04-2.03 (app m, 12H), 2.01-2.00 (2s, 6H), 1.00-1.96 (app m, 9H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 171.0, 170.5 (2C), 170.3 (2C), 170.2, 170.1, 170.0 (2C), 169.8, 169.6, 169.5, 169.1, 168.9, 168.8, 101.4, 100.1 (2C), 91.8, 89.1, 76.0, 75.7, 74.2, 73.5, 73.4, 72.5, 72.4, 72.3, 71.8, 71.7, 71.4, 71.3, 71.1, 70.9, 70.5, 69.5, 68.9, 68.8, 67.2 (2C), 66.9, 66.7, 62.5, 62.3, 61.8, 61.5, 61.0 (2C), 21.1, 21.0 (3C), 20.9, 20.8 (3C), 20.7, 20.6, 20.4; **HRMS (ESI)**: *m/z* calcd. for $C_{40}H_{54}O_{27}Na$ ($M+Na^+$) 989.2744, found 989.2730; **FT-IR**: 2946, 1751, 1369, 1226, 1045 cm^{-1} ; **$[\alpha]_D^{25}$** = - 34.2 (c=0.755, $CHCl_3$).

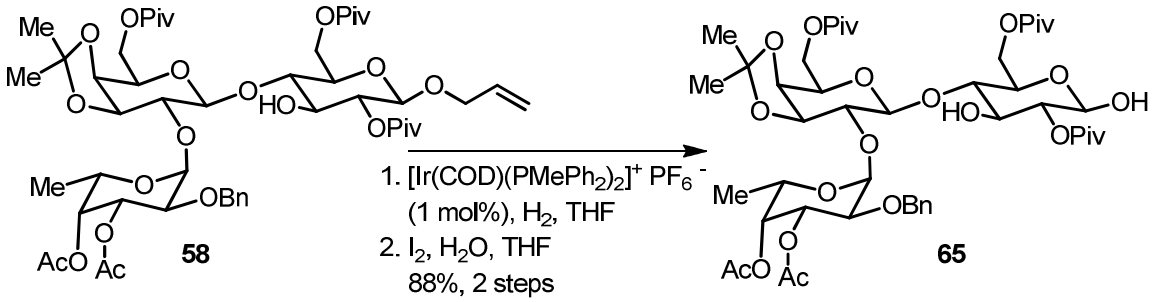
Synthesis of 2'-glucoseptanosyllactose (40)



To a solution of peracylated compound **O** (0.062 g, 0.064 mmol) in methanol (6.2 mL) was added a 0.5 M solution of NaOMe in MeOH (0.006 mL, 0.0032 mmol) and the reaction stirred at room temperature for 24 h. Neutralized the reaction with amberlite 120(H^+) resin, filtered and removed the solvent under vacuum. The crude solid obtained was washed with ether, hexanes and decanted. Dried under vacuum to obtain 2'-glucoseptanosyllactose **40** as a white solid (0.025 g, 78%). **^{1}H NMR** (600 MHz, D_2O) δ 5.21 (d, *J* = 3.6 Hz, 1H), 4.98 (dd, *J* = 6.0 Hz, 1H), 4.63 (d, *J* = 7.8 Hz, 1H), 4.49 (d, *J* = 7.8 Hz, 1H), 4.00-3.50 (m, 34H), 3.27 (app t, *J* = 9.0 Hz, 1H); **^{13}C NMR** (150 MHz, D_2O) δ 103.0, 101.0, 95.9, 95.8, 91.9, 77.3, 77.2, 75.7, 75.5, 75.3, 75.2, 74.4, 74.1, 74.0, 73.5, 72.6, 71.3, 69.4, 69.1, 68.9, 63.5, 61.1, 60.1; **HRMS (ESI)**: *m/z* calcd. for $C_{18}H_{32}O_{16}Na$ ($M+Na^+$) 527.1582, found 527.1575; **$[\alpha]_D^{25}$** = - 27.4 (c=0.725, H_2O).

3+3 modular approach

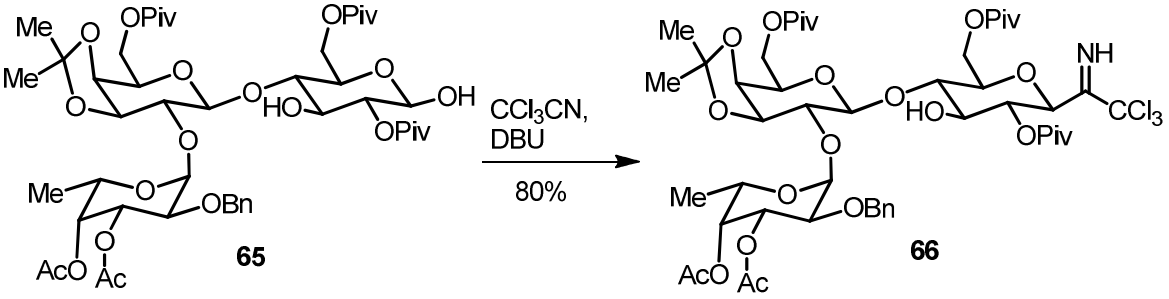
Synthesis of deallylated 2'-fucosyllactose trisaccharide 65



A solution of the catalyst $Ir(COD)(PMePh_2)_2]^+PF_6^-$ (3.6 mg, 0.0042 mmol) in THF (0.6 mL) was degassified and then stirred for 15 min under an athmosphere of hydrogen (red color of the solution decolorised). This was then cannulated into a solution of compound **58** (0.42 g, 0.42 mmol) in THF (8.4 mL) at room temperature and stirred for 30 min. The reaction was diluted with saturated $NaHCO_3$ and the aqueous was extracted with hexanes. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain the compound as white solid. To this compound in a mixture of THF (7.0 mL) and water (3.0 mL) was added iodine (0.21 g, 0.84 mmol) at room temperature and the brown reaction mixture stirred for 30 min. The reaction mixture was diluted with ethyl acetate and the organic layer washed with saturated $Na_2S_2O_3$, saturated $NaHCO_3$, water and brine. The organic layer was dried over $MgSO_4$, filtered and concentrated to obtain compound **65** as solid (0.35 g, 88%). **^{1}H NMR** (600 MHz, $CDCl_3$) δ 7.34-7.28 (m, 5H), 5.53 (app t, *J* = 4.8 Hz, 1H), 5.33-5.28 (m, 2H), 5.24 (dt, *J* = 3.0, 7.2 Hz, 1H), 4.72-4.58 (m, 5H), 4.41-4.33 (m, 3H), 4.29-4.25 (m, 1H), 4.23-4.18 (m, 2H). 4.10-4.00 (m, 4H), 3.87-3.84 (m, 1H), 3.78-3.75 (m, 2H), 3.67 (dd, *J* =

7.8 Hz, 0.5H), 3.54 (app t, *J* = 8.2 Hz, 1H), 3.47 (app t, *J* = 9.6 Hz, 0.6H), 2.12 (s, 3H), 1.97-1.96 (2s, 3H), 1.51-1.50 (2s, 3H), 1.33 (s, 3H), 1.23 (s, 9H), 1.21 (s, 18H), 1.13 (app t, *J* = 7.2 Hz, 3H); **^{13}C NMR** (150 MHz, $CDCl_3$) δ 179.6, 178.5, 178.1, 177.9, 177.7, 170.7, 170.0, 169.8, 139.3, 128.5, 127.9, 127.8, 110.9, 101.1, 100.7, 95.7, 90.3, 80.8, 80.3, 79.4, 79.3, 75.9, 75.3, 73.7, 73.6, 73.4, 73.3 72.8, 72.7, 72.6, 72.2, 71.8 (2C), 69.7, 69.5, 68.7, 68.2, 65.0, 64.9, 63.2, 62.2, 61.4, 39.1, 39.0, 28.0, 27.9, 27.4, 27.3, 27.2, 26.5, 26.4, 20.9 (2C), 16.0, 15.9.

Synthesis of 2'-fucosyllactose trisaccharide trichloroacetamidate donor 66



To a solution of compound **65** (0.1 g, 0.014 mmol) in CH_2Cl_2 at room temperature was added trichloroacetonitrile (0.075 g, 0.052 mmol) followed by DBU (0.0032 g, 0.0029 mmol) and stirred the brown reaction mixture for 20 h. The solvent was removed under vacuum and the crude purified by flash chromatography using 10% ethyl acetate in hexanes containing 2% Et_3N as eluent to obtain the donor **66** as a white solid (0.089 g, 80% α-anomer). **^{1}H NMR** (600 MHz, C_6D_6) 8.45 (s, 1H), 7.36 (d, *J* = 7.2 Hz, 2H), 7.18 (app t, *J* = 7.8 Hz, 2H), 7.08 (app t, *J* = 7.2 Hz, 1H), 6.83 (d, *J* = 3.6 Hz, 1H), 5.89 (d, *J* = 3.6 Hz, 1H), 5.77 (dd, *J* = 3.6, 10.8 Hz, 1H), 5.75-5.74 (m, 1H), 5.28 (dd, *J* = 3.6, 9.9 Hz, 1H), 4.81-4.77 (m, 2H), 4.64 (d, *J* = 11.4 Hz, 1H), 4.60 (t, *J* = 9.6 Hz, 1H), 4.49 (d, *J* = 11.4 Hz, 1H), 4.42-4.36 (m,

4H), 4.25 (dd, *J* = 9.6, 11.7 Hz, 1H), 3.86 (d, *J* = 8.4 Hz, 1H), 3.59-3.57 (m, 1H), 3.49 (dd, *J* = 1.8, 6.0 Hz, 1H), 3.19 (app t, *J* = 10.2 Hz, 1H), 1.77 (s, 3H), 1.72 (s, 3H), 1.36 (s, 12H), 1.32 (s, 9H), 1.30 (d, *J* = 7.2 Hz, 3H), 1.22 (s, 9H), 1.10 (s, 3H); **^{13}C NMR** (150 MHz, C_6D_6) δ 178.2 (2C), 177.3, 170.2, 169.7, 161.0, 139.2, 128.8, 128.5, 128.4, 128.3, 128.1, 127.8, 110.9, 101.7, 96.1, 93.5, 82.8, 80.5, 76.0, 74.7, 74.0, 72.7, 72.6, 72.2, 70.7, 70.0, 69.9, 65.8, 63.7, 63.6, 47.0, 39.3, 39.2, 28.0, 27.6, 27.5, 27.4, 26.5, 20.8, 20.5, 16.4.

References

1. Rheeder, J. P.; Marasas, W. F. O.; Vismer, H. F., *Appl. Environ. Microbiol.* **2002**, *68*, 2101.

2. Henry, M. H.; Wyatt, R. D., *J. Appl. Poult. Res.* **1993**, *2*, 188.

3. (a) Bezuidenhout, S. C.; Gelderblom, W. C. A.; Gorst-Allman, C. P.; Horak, R. M.; Marasas, W. F. O.; Spiteller, G.; Vleggaar, R., *J. Chem. Soc. Chem. Commun.* **1988**, 743; (b) Gelderblom, W. C. A.; Jaskiewicz, K.; Marasas, W. F. O.; Thiel, P. G.; Horak, R. M.; Vleggaar, R.; Kriek, N. P. J.; Fumonisins, n., *Appl. Environ. Microbiol.* **1988**, *54*, 1806; (c) Marasas, W. F. O., *Environ. Health Perspect. Suppl.* **2001**, *109*, 239.

4. (a) Bottini, A. T.; Bowen, J. R.; Gilchrist, D. G., *Tetrahedron Lett.* **1981**, *22*, 2723; (b) Bottini, A. T.; Gilchrist, D. G., *Tetrahedron Lett.* **1981**, *22*, 2719.

5. (a) Blackwell, B. A.; Edwards, O. E.; ApSimon, J. W.; Fruchier, A., *Tetrahedron Lett.* **1995**, *36*, 1973; (b) ApSimon, J. W.; Blackwell, B. A.; Edwards, O. E.; Fruchier, A., *Tetrahedron Lett.* **1994**, *35*, 7703; (c) Boyle, C. D.; Kishi, Y., *Tetrahedron Lett.* **1995**, *36*, 4579; (d) Hoye, T. R.; Jimenez, J. I.; Shier, W. T., *J. Am. Chem. Soc.* **1994**, *116*, 9409; (e) Shier, W. T.; Abbas, H. K.; Badria, F. A., *Tetrahedron Lett.* **1995**, *36*, 1571; (f) Poch, G. K.; Powell, R. G.; Plattner, R. D.; Weisleder, D., *Tetrahedron Lett.* **1994**, *35*, 7707; (g) Hartl, M.; Humpf, H., *Tetrahedron: Asymmetry.* **1998**, *9*, 1549.

6. (a) Desai, K.; Sullards, M. C.; Allegood, J.; Wang, E.; Schmelz, E. M.; Hartl, M.; Humpf, H.-U.; Liotta, D. C.; Peng, Q.; Merrill, A. H., *BBA-Mol. Cell. Biol. L.* **2002**, *1585*, 188; (b) Soriano, J. M.; González, L.; Catalá, A. I., *Prog. Lipid Res.* **2005**, *44*, 345; (c) Zitomer, N. C.; Mitchell, T.; Voss, K. A.; Bondy, G. S.;

Pruett, S. T.; Garnier-Amblard, E. C.; Liebeskind, L. S.; Park, H.; Wang, E.; Sullards, M. C.; Merrill, A. H.; Riley, R. T., *J. Biol. Chem.* **2009**, *284*, 4786.

7. Stockmann-Juvala, H.; Savolainen, K., *Hum. Exp. Toxicol.* **2008**, *27*, 799.

8. Wang, E.; Norred, W. P.; Bacon, C. W.; Riley, R. T.; Merrill, A. H., *J. Biol. Chem.* **1991**, *266*, 14486.

9. (a) Merrill, A. H., *J. Biol. Chem.* **2002**, *277*, 25843; (b) Ogretmen, B.; Hannun, Y. A., *Nat. Rev. Cancer.* **2004**, *4*, 604.

10. Hartl, M.; Humpf, H. U., *J. Agric. Food. Chem.***1999**, *47*, 5078.

11. (a) Sydenham, E. W.; Thiel, P. G.; Marasas, W. F. O.; Shephard, G. S.; Van Schalkwyk, D. J.; Koch, K. R., *J. Agric. Food. Chem.* **1990**, *38*, 1900; (b) Chu, F. C.; Li, G. Y., *Appl. Environ. Microbiol.* **1994**, *60*, 847.

12. (a) Hendricks, K., *Epidemiology* **1999**, *10*, 198; (b) Missmer, S. A.; Suarez, L.; Felkner, M.; Wang, E.; Merrill, A. H., Jr.; Rothman, K. J.; Hendricks, K. A., *Environ. Health Perspect.* **2006**, *114*.

13. Bhat, R. V.; Shetty, P. H.; Amruth, R. P.; Sudershan, R. V., *J. Toxicol. Clin. Toxicol.* **1997**, *35*, 249

14. (a) *Lyon: IARC.* **1993**, *56*, 445; (b) *Lyon: IARC.* **2002**, *82*, 301.

15. (a) Marasas, W. F. O.; Kellerman, T. S.; Gelderblom, W. C. A.; Coetzer, J. A. W.; Thiel, P. G.; van der Lugt, J. J., *Onderstepoort J. Vet. Res.* **1988**, *55*, 197; (b) Kellerman, T. S.; Marasas, W. F. O.; Thiel, P. G.; Gelderblom, W. C. A.; Cawood, M.; Coetzer, J. A. W., *Onderstepoort J. Vet. Res.* **1990**, *57*, 269; (c) Wilson, T. M.; Ross, F. P.; Owens, D. L.; Rice, L. G.; Green, S. A.; Jenkins, S. J.; Nelson, H. A., *Mycopathalogia* **1992**, *11*, 115.

16. (a) Haschek, W. M.; Motelin, G.; Ness, D. K.; Harlin, K. S.; Hall, W. F.; Vesonder, R. F.; Peterson, R. E.; Beasley, V. R., *Mycopathologia* **1992**, *117*, 83; (b) Haschek, W. M.; Gumprecht, L. A.; Smith, G.; Tumbleson, M. E.; Constable, P.D., *Environ. Health Perspect.* **2001**, *109*, 251.

17. Smith, G. W.; Constable, P. D.; Eppley, R. M.; Tumbleson, M. E.; Gumprecht, L. A.; Haschek-Hock, W. M., *Toxicol. Sci.* **2000**, *56*, 240.

18. (a) Wentzel,C. A.; Gelderblom, S. A.; Smuts, C. M.; Marnewick, J.; Marasas, W.F.O.; Lemmer, E. R.; Ramljak, D., *Environ. Health. Perspect.* **2001**, *109*, 291; (b) Gelderblom, W. C. A.; Cawood, M. E.; Snyman, S. D.; Vleggaar, R.; Marasas, W. F. O., *Food Chem. Toxicol.* **1993**, *31*, 407.

19. Oikawa, H.; Yamawaki, D.; Kagawa, T.; Ichihara, A., *Tetrahedron Lett.* **1999**, *40*, 6621.

20. Shi, Y.; Peng, L. F.; Kishi, Y., *J. Org. Chem.* **1997**, *62*, 5666.

21. Kraus, G. A.; Applegate, J. M.; Reynolds, D., *J. Agric. Food. Chem.* **1992**, *40*, 2331.

22. Gurjar, M. K.; Rajendran, V.; Rao, B. V., *Tetrahedron Lett.* **1998**, *39*, 3803.

23. Blackwell, B. A.; Edward, O. E.; Fruchier, A.; ApSimon, J. W.; Miller, J. D., *Adv. Exp. Med. Biol.* **1996**, *392*, 75.

24. Wilson, M. J.; Haggart, C. L.; Gallagher, S. P.; Walsh, D., *J. Biotechnol.* **2001**, *88*, 67.

25. (a) Gorbet, M. B.; Sefton, M. V., *Biomaterials* **2005**, *26*, 6811; (b) Wakelin, S. J.; Sabroe, I.; Gregory, C. D.; Poxton, I. R.; Forsythe, J. L. R.; Garden, O. J.; Howie, S. E. M., *Immunol. Lett.* **2006**, *106*, 1.

26. Trent, M. S.; Stead, C. M.; Tran, A. X.; Hankins, J. V., *J. Endotoxin Res.* **2006**, *12*, 205.

27. Beutler, B.; Rietschel, E. T., *Nat. Rev. Immunol.* **2003**, *3*, 169.

28. Merrill, A. H, Private communication

29. Developing Chiral Synthons for Polyketide-Based Natural Product Synthesis via Stereospecific Oxonia-Cope Rearrangement. Chen, Y.-H, Emory University, Thesis, 2007.

30. Chen, Y. H.; McDonald, F. E., *J. Am. Chem. Soc.* **2006**, *128*, 4568.

31. Wiseman, J. M.; McDonald, F. E.; Liotta, D. C., *Org. Lett.* **2005**, *7*, 3155.

32. Marshall, J. A.; Bourbeau, M. P., *Org. Lett.* **2003**, *5*, 3197.

33. (a) Midland, M. M., *Chem. Rev.* **1989**, *89*, 1553; (b) Matsumura, K.; Hashiguchi, S.; Ikariya, T.; Noyori, R., *J. Am. Chem. Soc.* **1997**, *119*, 8738; (c) Helal, C. J.; Magriotis, P. A.; Corey, E. J., *J. Am. Chem. Soc.* **1996**, *118*, 10938.

34. (a) Nokami, J.; Ohga, M.; Nakamoto, H.; Matsubara, T.; Hussain, I.; Kataoka, H., *J. Am. Chem. Soc.* **2001**, *123*, 9168; (b) Nokami, J.; Nomiyama, K.; Matsuda, S.; Imai, N.; Kataoka, H., *Angew. Chem. Int. Ed.* **2003**, *42*, 1273.

35. (a) López, F.; Harutyunyan, S. R.; Meetsma, A.; Minnaard, A. J.; Feringa, B. L., *Angew. Chem, Int. Ed.* **2005**, *44*, 2752; (b) Schuppan, J.; Minnaard, A. J.; Feringa, B. L., *Chem. Commun.* **2004**, 792; (c) Feringa, B. L.; Badorrey, R.; Peña, D.; Harutyunyan, S. R.; Minnaard, A. J., *J. Proc. Natl. Acad. Sci. U.S.A.* **2004**, *101*, 5834; (d) Des Mazery, R.; Pullez, M.; López, F.; Harutyunyan, S. R.; Minnaard, A. J.; Feringa, B. L., *J. Am. Chem. Soc.* **2005**, *127*, 9966.

36. (a) Rychnovsky, S. D.; Rogers, B.; Yang, G., *J. Org. Chem.* **1993**, *58*, 3511; (b) Rychnovsky, S. D.; Skalitzky, D. J., *Tetrahedron Lett.* **1990**, *31*, 945.

37. (a) Richardson, T. I.; Rychnovsky, S. D., *J. Org. Chem.* **1996**, *61*, 4219; (b) Rychnovsky, S. D.; Griesgraber, G.; Schlegel, R., *J. Am.Chem. Soc.* **1995**, *117*, 197.

38. (a) Wang, S. Y.; Lum, T. K.; Ji, S. J.; Loh, T. P., *Adv. Synth. Catal.* **2008**, *350*, 673; (b) Lum, T. K.; Wang, S. Y.; Loh, T. P., *Org. Lett.* **2008**, *10*, 761.

39. Lee, C. K.; Lee, C. A.; Tan, K.; Loh, T., *Org. Lett.* **2004**, *6*, 1281.

40. Harmat, N. J. S.; Mangani, S.; Perrotta, E.; Giannotti, D.; Nannicini, R.; Altamura, M., *Tetrahedron Lett.* **2000**, *41*, 1261.

41. Layton, M. E.; Morales, C. A.; Shair, M. D., *J. Am. Chem. Soc.* **2002**, *124*, 773.

42. Dimitrov, V.; Simova, S.; Kostova, K., *Tetrahedron* **1996**, *52*, 1699.

43. Mihelich, E. D.; Daniels, K.; Eickhoff, D. J., *J. Am. Chem. Soc.* **1981**, *103*, 7690.

44. Mitsunobu, O., *Synthesis* **1981**, 1.

45. Caron, M.; Carlier, P. R.; Sharpless, K. B., *J. Org. Chem.* **1988**, *53*, 5187.

46. (a) Azzena, F.; Calvani, F.; Crotti, P.; Gardelli, C.; Macchia, F.; Pineschi, M., *Tetrahedron* **1995**, *51*, 10601; (b) Benedetti, F.; Berti, F.; Norbedo, S., *Tetrahedron Lett.* **1998**, *39*, 7971; (c) Nugent, W. A., *J. Am. Chem. Soc.* **1992**, *114*, 2768; (d) DiFuria, F.; Licini, G.; Modena, G.; Motterle, R.; Nugent, W. A., *J. Org. Chem.* **1996**, *61*, 5175; (e) McCleland, B. W.; Nugent, W. A.; Finn, M. G., *J.*

Org. Chem. **1998**, *63*, 6656; (f) Nugent, W. A.; Harlow, R., *J. Am. Chem. Soc.* **1994**, *116*, 6142.

47. Evans, D. A.; Sjogren, E. B.; Bartroli, J.; Dow, R. L., *Tetrahedron Lett.* **1986**, *27*, 4957.

48. Ahmed, M. M.; Cui, H.; O'Doherty, G. A., *J. Org. Chem.* **2006**, *71*, 6686.

49. (a) Evans, D. A.; Nelson, J. V.; Vogel, E.; Taber, T. R., *J. Am. Chem. Soc.* **1981**, *103*, 3099; (b) Kühnel, E.; Laffan, D. D. P.; Lloyd-Jones, G. C.; Martínez del Campo, T.; Shepperson, I. R.; Slaughter, J. L., *Angew. Chem. Int. Ed.* **2007**, *46*, 7075.

50. (a) Evans, D. A.; Adams, D. J., *J. Am. Chem. Soc.* **2007**, *129*, 1048; (b) Morita, A.; Kuwahara, S., *Org. Lett.* **2006**, *8*, 1613; (c) Morita, A.; Kiyota, H.; Kuwahara, S., *Biosci. Biotechnol. Biochem.* **2006**, *70*, 2564.

51. (a) Eustache, F.; Dalko, P. I.; Cossy, J., *Tetrahedron Lett.* **2003**, *44*, 8823; (b) Evans, D. A.; Nagorny, P.; McRae, K. J.; Reynolds, D. J.; Sonntag, L. S.; Vounatsos, F.; Xu, R., *Angew. Chem. Int. Ed.* **2007**, *46*, 537.

52. Gao, Y.; Klunder, J. M.; Hanson, R. M.; Masamune, H.; Ko, S. Y.; Sharpless, K. B., *J. Am. Chem. Soc.* **1987**, *109*, 5765.

53. (a) Suzuki, T.; Saimoto, H.; Tomioka, H.; Oshima, K.; Nozaki, H., *Tetrahedron Lett.* **1982**, *23*, 3597; (b) Inoue, M.; Carson, M. W.; Frontier, A. J.; Danishefsky, S. J., *J. Am. Chem. Soc.* **2001**, *123*, 1878.

54. (a) Shintani, R.; Kimura, T.; Hayashi, T., *Chem. Commun.* **2005**, 3213; (b) Blake, S.; Giunta. A. J., Giunta, D.; Shannon, D.; Shannon, J.; Solinas, J.;

Solinas, M.; Walzer, W.; Walzer, F.; Woodward, F.; Woodward, S.,*Collect. Czech. Chem. Commun.* **2007**, *78*, 1107.

55. Poon, K. W. C.; Dudley, G. B., *J. Org. Chem.* **2006**, *71*, 3923.

56. Abdel-Sattar, S. H. E., *Curr. Org. Chem.* **2004**, *8*, 1405.

57. Oikawa, H.; Kagawa, T.; Kobayashi, T.; Ichihara, A., *Tetrahedron Lett.* **1996**, *37*, 6169.

58. (a) Lelais, G.; Campo, Marino A.; Kopp, S.; Seebach, D., *Helvetica Chimica Acta.* **2004**, *87*, 1545; (b) Arvanitis, E.; Motevalli, M.; Wyatt, P. B., *Tetrahedron Lett.* **1996**, *37*, 4277.

59. Pereira, C. L.; Chen, Y. H.; McDonald, F. E., *J. Am. Chem. Soc.* **2009**, *131*, 6066.

60. (a) Alberts, J. F.; Gelderblom, W. C.; Thiel, P. G.; Marasas, W. F.; Van Schalkwyk, D. J.; Behrend, Y., *Appl. Environ. Microbiol.* **1990**, *56*, 1729; (b) Plattner, R. D.; Norred, W. P.; Bacon, C. W.; Voss, K. A.; Paterson, R.; Schackelford, D. D.; Weisleder, D., *Mycologia* **1990**, *82*, 698; (c) Wilkes, J. G.; Sutherland, J. B.; Churchwell, M. I.; Williams, A. J., *J. Chromatogr. A.* **1995**, *695*, 319; (d) Dilkin, P.; Mallmann, C. A.; de Almeida, C. A. A.; Corrêa, B., *J. Chromatogr. A.* **2001**, *925*, 151; (e) Kim, E.-K.; Maragos, C. M.; Kendra, D. F., *J. Agric. Food. Chem.* **2003**, *52*, 196; (f) Thakur, R. A.; Smith, J. S., *J. Agric. Food. Chem.* **1996**, *44*, 1047.

61. (a) Cawood, M. E.; Gelderblom, W. C. A.; Vleggaar, R.; Behrend, Y.; Thiel, P. G.; Marasas, W. F. O., *J. Agric. Food Chem.* **1991**, *39*, 1958; (b) Meredith, F. I.; Bacon, C. W.; Plattner, R. D.; Norred, W. P., *J. Agric. Food.*

Chem. **1996**, *44*, 195; (c) Dantzer, W. R.; Hopmans, E.; Clark, A.; Hauck, C.; Murphy, P. A., *J. Agric. Food. Chem.* **1996**, *44*, 3730; (d) Bailly, J. D.; Querin, A.; Tardieu, D.; Guerre, P., *Revue Méd. Vét.* **2005**, *156*, 547.

62. Seiferlein, M.; Humpf, H. U.; Voss, K. A.; Sullards, M. C.; Allegood, J. C.; Wang, E.; Merrill, A. H., *Mol. Nutr. Food. Res.* **2007**, *51*, 1120.

63. Refer experimental details.

64. Merrill, A. H, Private communication.

65. (a) Vesper, H.; Schmelz, E.-M.; Nikolova-Karakashian, M. N.; Dillehay, D. L.; Lynch, D. V.; Merrill, A. H., Jr., *J. Nutr.* **1999**, *129*, 1239; (b) Howell, A. R.; Ndakala, A. J., *Curr. Org. Chem.* **2002**, *6*, 365.

66. (a) Liao, J.; Tao, J.; Lin, G.; Liu, D., *Tetrahedron* **2005**, *61*, 4715; (b) Merrill, A. H.; Schmelz, E. M.; Dillehay, D. L.; Spiegel, S.; Shayman, J. A.; Schroeder, J. J.; Riley, R. T.; Voss, K. A.; Wang, E., *Toxicol. Appl. Pharmacol.* **1997**, *142*, 208.

67. (a) Pruett, S. T.; Bushnev, A.; Hagedorn, K.; Adiga, M.; Haynes, C. A.; Sullards, M. C.; Liotta, D. C.; Merrill, A. H., Jr., *J. Lipid Res.* **2008**, *49*, 1621; (b) Shier, W.; Shier, A., *J. Toxicol. Toxin. Rev.* **2000**, *19*, 189.

68. Padrón, J. M., *Curr. Med. Chem.* **2006**, *13*, 755.

69. (a) Fahy, E.; Subramaniam, S.; Brown, H. A.; Glass, C. K.; Jr., A. H. M.; Murphy, R. C.; Raetz, C. R. H.; Russell, D. W.; Seyama, Y.; Shaw, W.; Shimizu, T.; Spener, F.; Meer, G. v.; VanNieuwenhze, M. S.; White, S. H.; Witztum, J. L.; Dennis, E. A., *Eur. J. Lipid Sci. Technol.* **2005**, *107*, 337; (b) Fahy, E.; Subramaniam, S.; Brown, H. A.; Glass, C. K.; Merrill, A. H., Jr.; Murphy, R. C.;

Raetz, C. R. H.; Russell, D. W.; Seyama, Y.; Shaw, W.; Shimizu, T.; Spener, F.; van Meer, G.; VanNieuwenhze, M. S.; White, S. H.; Witztum, J. L.; Dennis, E. A., *J. Lipid Res.* **2005**, *46*, 839.

70. (a) Woodcock, J., *IUBMB Life.* **2006**, *58*, 462; (b) Hannun, Y. A.; Obeid, L. M., *J. Biol. Chem.* **2002**, *277*, 25847.

71. (a) Sabbadini, R. A., *Br. J. Cancer* **2006**, *95*, 1131; (b) Milstien, S.; Spiegel, S., *Cancer Cell* **2006**, *9*, 148; (c) Spiegel, S.; Milstien, S., *J. Biol. Chem.* **2002**, *277*, 25851.

72. (a) Ogretmen, B., *FEBS Letters* **2006**, *580*, 5467; (b) Modrak, D. E.; Gold, D. V.; Goldenberg, D. M., *Mol. Cancer Ther.* **2006**, *5*, 200; (c) Sahar, A.; Saddoughi, P. S.; Ogretmen, B., *Subcell. Biochem.* **2008**, *49*, 413.

73. (a) Radin, N. S., *Bioorg. Med. Chem.* **2003**, *11*, 2123; (b) Curfman, C. L.; Kirkland, K.; Merrill, A. H., *Expert. Opin. Ther. Pat.* **2006**, *16*, 1129.

74. Menaldino, D. S.; Bushnev, A.; Sun, A.; Liotta, D. C.; Symolon, H.; Desai, K.; Dillehay, D. L.; Peng, Q.; Wang, E.; Allegood, J.; Trotman-Pruett, S.; Sullards, M. C.; Merrill, A. H., *Pharmacol. Res.* **2003**, *47*, 373.

75. Asymmetric synthesis of amino alcohols as bioactive compounds. Menaldino, D. S, Emory University, Thesis, 1992.

76. Bushnev, A. S.; Bailley, M. T.; Holt, J.; Menaldino, D. S.; Merrill Jr, A. H.; Liotta, D. C., **2010**, paper submitted.

77. Saikia, P. P.; Goswami, A.; Baishya, G.; Barua, N. C., *Tetrahedron Lett.* **2009**, *50*, 1328.

78. Dias, L. C.; Fattori, J.; Perez, C. C., *Tetrahedron Lett.* **2008**, *49*, 557.

79. Dias, L. C.; Fattori, J.; Perez, C. C.; de Oliveira, V. M.; Aguilar, A. M., *Tetrahedron* **2008**, *64*, 5891.

80. (a) Duan, J. J. W.; Smith, A. B., *J. Org. Chem.* **1993**, *58*, 3703; (b) Bartlett, P. A.; Meadows, J. D.; Brown, E. G.; Morimoto, A.; Jernstedt, K. K., *J. Org. Chem.* **1982**, *47*, 4013.

81. Harmange, J. C.; Boyle, C. D.; Kishi, Y., *Tetrahedron Lett.* **1994**, *35*, 6819.

82. (a) Bongini, A.; Cardillo, G.; Orena, M.; Porzi, G.; Sandri, S., *J. Org. Chem.* **1982**, *47*, 4626; (b) Ahmar, M.; Romain, I.; Bloch, R., *J. Org. Chem.* **1993**, *58*, 2953.

83. Symolon, H.; Bushnev, A.; Peng, Q.; Ramaraju, H.; Mays, S.; Allegood, J.; Pruett, S. E.; Pack, C. D.; Zheng, W.; Sullards, C.; Petros, J.; Dillehay, D. I.; Liotta, D. C.; Merrill Jr, A. H., **2010**, manuscript in preparation.

84. (a) Andreou, T.; Costa, A. M.; Esteban, L.; Gonzàlez, L.; Mas, G.; Vilarrasa, J., *Org. Lett.* **2005**, *7*, 4083; (b) Le Flohic, A.; Meyer, C.; Cossy, J., *Org. Lett.* **2004**, *7*, 339.

85. (a) Yamamoto, Y.; Asao, N., *Chem. Rev.* **1993**, *93*, 2207; (b) Denmark, S. E.; Almstead, N. G.; Otera, J., 2000; p 299; (c) Brown, H. C., *J. Org. Chem.* **1991**, *56*, 401; (d) Nokami, J.; Nomiyama, K.; Shafi, S. M.; Kataoka, K., *Org. Lett.* **2004**, *6*, 1261.

86. (a) Mikami, K.; Shimizu, M., *Chem. Rev.* **1992**, *92*, 1021; (b) Mikami, K.; Terada, M.; Narisawa, S.; Nakai, T., *Synlett* **1992**, 255.

87. Brown, H. C.; Jadhav, P. K., *J. Am. Chem. Soc.* **1983**, *105*, 2092.

88. (a) Evans, D. A.; Wu, J., *J. Am. Chem. Soc.* **2005**, *127*, 8006; (b) Zhao, J.-F.; Tsui, H.-Y.; Wu, P.-J.; Lu, J.; Loh, T.-P., *J. Am. Chem. Soc.* **2008**, *130*, 16492.

89. Nokami, J.; Yoshizane, K.; Matsuura, H.; Sumida, S., *J. Am. Chem. Soc.* **1998**, *120*, 6609.

90. Samoshin, V. V.; Smoliakova, I. P.; Han, M.; Gross, P. H., *Mendeleev Commun.* **1999**, *9*, 219.

91. (a) Lolkema, L. D. M.; Hiemstra, H.; Semeyn, C.; Speckamp, W. N., *Tetrahedron* **1994**, *50*, 7115; (b) Lolkema, L. D. M.; Semeyn, C.; Ashek, L.; Hiemstra, H.; Speckamp, W. N., *Tetrahedron* **1994**, *50*, 7129.

92. (a) Hopkins, M. H.; Overman, L. E., *J. Am. Chem. Soc.* **1987**, *109*, 4748; (b) Brown, M. J.; Harrison, T.; Herrinton, P. M.; Hopkins, M. H.; Hutchinson, K. D.; Overman, L. E.; Mishra, P., *J. Am. Chem. Soc.* **1991**, *113*, 5365; (c) Hopkins, M. H.; Overman, L. E.; Rishton, G. M., *J. Am. Chem. Soc.* **1991**, *113*, 5354.

93. Nokami, J.; Anthony, L.; Sumida, S., *Chem. Eur. J.* **2000**, *6*, 2909.

94. (a) Hussain, I.; Komasaka, T.; Ohga, M.; Nokami, J., *Synlett* **2002**, 640; (b) Loh, T. P.; Hu, Q. Y.; Chok, Y. K.; Tan, K. T., *Tetrahedron Lett.* **2001**, *42*, 9277.

95. Lee, C.-H. A.; Loh, T.-P., *Tetrahedron Lett.* **2004**, *45*, 5819.

96. (a) Rychnovsky, S. D.; Marumoto, S.; Jaber, J. J., *Org. Lett.* **2001**, *3*, 3815; (b) Jasti, R.; Anderson, C. D.; Rychnovsky, S. D., *J. Am. Chem. Soc.* **2005**, *127*, 9939; (c) Crosby, S. R.; Harding, J. R.; King, C. D.; Parker, G. D.; Willis, C. L., *Org. Lett.* **2002**, *4*, 577; (d) Crosby, S. R.; Harding, J. R.; King, C. D.; Parker,

G. D.; Willis, C. L., *Org. Lett.* **2002**, *4*, 3407; (e) Kataoka, K.; Ode, Y.; Matsumoto, M.; Nokami, J., *Tetrahedron* **2006**, *62*, 2471.

97. Nokami, J.; Maruoka, K.; Souda, T.; Tanaka, N., *Tetrahedron* **2007**, *63*, 9016.

98. Jung, S.-Y.; Min, J.-H.; Oh, J. T.; Koo, S., *J. Org. Chem.* **2006**, *71*, 4823.

99. Cheng, H.-S.; Seow, A.-H.; Loh, T.-P., *Org. Lett.* **2008**, *10*, 2805.

100. Yuan, Y.; Lai, A. J.; Kraml, C. M.; Lee, C., *Tetrahedron* **2006**, *62*, 11391.

101. Shafi, S. M.; Chou, J.; Kataoka, K.; Nokami, J., *Org. Lett.* **2005**, *7*, 2957.

102. Woo, Sang K.; Kwon, Min S.; Lee, E., *Angew. Chem. Int. Ed.* **2008**, *47*, 3242.

103. Cheng, H.-S.; Loh, T.-P., *J. Am. Chem. Soc.* **2003**, *125*, 4990.

104. Mandai, T.; Oshitari, T.; Susowake, M., *Synlett* **2002**, *2002*, 1665.

105. Burgos, C. H.; Canales, E.; Matos, K.; Soderquist, J. A., *J. Am. Chem. Soc.* **2005**, *127*, 8044.

106. (a) Brown, H. C.; Bhat, K. S., *J. Am. Chem. Soc.* **1986**, *108*, 5919; (b) Statsuk, A. V.; Liu, D.; Kozmin, S. A., *J. Am. Chem. Soc.* **2004**, *126*, 9546.

107. Hoye, T. R.; Jeffrey, C. S.; Shao, F., *Nat. Protocols* **2007**, *2*, 2451.

108. (a) Rauniyar, V.; Hall, D. G., *J. Org. Chem.* **2009**, *74*, 4236; (b) Rauniyar, V.; Zhai, H.; Hall, D. G., *J. Am. Chem. Soc.* **2008**, *130*, 8481.

109. Hafner, A.; Duthaler, R. O.; Marti, R.; Rihs, G.; Rothe-Streit, P.; Schwarzenbach, F., *J. Am. Chem. Soc.* **1992**, *114*, 2321.

110. (a) Swamy, K. C. K.; Kumar, N. N. B.; Balaraman, E.; Kumar, K. V. P. P., *Chem. Rev.* **2009**, *109*, 2551; (b) But, T. Y. S.; Toy, Patrick H., *Chem. Asian J.* **2007**, *2*, 1340.

111. (a) Rychnovsky, S. D.; Thomas, C. R., *Org. Lett.* **2000**, *2*, 1217; (b) Jaber, J. J.; Mitsui, K.; Rychnovsky, S. D., *J. Org. Chem.* **2001**, *66*, 4679.

112. (a) Smith, A. B.; Kim, D.-S., *J. Org. Chem.* **2006**, *71*, 2547; (b) Tokunaga, M.; Larrow, J. F.; Kakiuchi, F.; Jacobsen, E. N., *Science* **1997**, 936.

113. Rodríguez, D.; Mulero, M.; Prieto, J. A., *J. Org. Chem.* **2006**, *71*, 5826.

114. Loh, T. P.; Lee, C. L. K.; Tan, K. T., *Org. Lett.* **2002**, *4*, 2985.

115. (a) Nicolaou, K. C.; Mitchell, H. J., *Angew. Chem. Int. Ed.* **2001**, *40*, 1576; (b) Hölemann, A.; Seeberger, P. H., *Curr. Opin. Biotechnol.* **2004**, *15*, 615.

116. (a) Seeberger, P. H., *Nat Chem Biol.* **2009**, *5*, 368; (b) Seeberger, P. H.; Werz, D. B., *Nat. Rev. Drug. Discov.* **2005**, *4*, 751.

117. Donovan, S. M., *Br. J. Nutr.* **2009**, *101*, 1267.

118. (a) Bode, L., *J. Nutr.* **2006**, *136*, 2127; (b) Boehm, G.; Stahl, B., *J. Nutr.* **2007**, *137*, 847S; (c) Kunz, C.; Rudloff, S.; Baier, W.; Klein, N.; Strobel, S., *Annu. Rev. Nutr.* **2000**, *20*, 699.

119. McVeagh, P.; Miller, J. B., *J. Paediatr. Child. Health.* **1997**, *33*, 281.

120. (a) Miller, J. B.; McVeagh, P., *Br. J. Nutr.* **1999**, *82*, 333; (b) Bode, L., *Nutr. Rev.* **2009**, *67*, S183.

121. Thurl, S.; Henker, J.; Siegel, M.; Tovar, K.; Sawatzki, G., *Glycoconjugate J.* **1997**, *14*, 795.

122. Yan, L.; Kahne, D., *J. Am. Chem. Soc.* **1996**, *118*, 9239.

123. Vandenplas, Y., *Br. J. Nutr.* **2002**, *87*, S293.

124. Coppa, G. V.; Zampini, L.; Galeazzi T.; Facinelli, B.; Ferrante, L.; Capretti, R.; Orazio, G., *Pediatr Res.* **2006**, *59*, 377.

125. Maria Espinosa, R.; Tamiz, M.; Prieto, P., *Br. J. Nutr.* **2007**, *98*, S74.

126. (a) Wang, B.; Brand-Miller, J., *Eur. J. Clin. Nutr.* **2003**, *57*, 1351; (b) Wang, B.; Brand-Miller, J.; McVeagh, P.; Petocz, P., *Am. J. Clin. Nutr.* **2001**, *74*, 510.

127. Werz, D. B.; Seeberger, P. H., *Chem. Eur. J.* **2005**, *11*, 3194.

128. (a) carbosynth, www.carbosynth.com*;* (b) Jennewein, www.jennewein-biotech.de.

129. Abbas, S. A.; Barlow, J. J.; Matta, K. L., *Carbohydr. Res.* **1981**, *88*, 51.

130. Jain, R. K.; Locke, R. D.; Matta, K. L., *Carbohydr. Res.* **1991**, *212*, C1.

131. (a) Rencurosi, A.; Poletti, L.; Russo, G.; Lay, L., *Eur. J. Org. Chem.* **2003**, *2003*, 1672; (b) Rencurosi, A.; Poletti, L.; Panza, L.; Lay, L., *J. Carbohydr. Chem.* **2001**, *20*, 761.

132. Izumi, M.; Tsuruta, O.; Harayama, S.; Hashimoto, H., *J. Org. Chem.* **1997**, *62*, 992.

133. (a) Fernandez-Mayoralas, A.; Martin-Lomas, M., *Carbohydr. Res.* **1986**, *154*, 93; (b) Fernandez-Mayoralas, A.; Martin-Lomas, M.; Villanueva, D., *Carbohydr. Res.* **1985**, *140*, 81.

134. Leonardo, M.; Lay, L.; Schmidt, R. R., *J. Carbohydr. Chem.* **1998**, *17*, 739.

135. Kim, H. M.; Kim, I. J.; Danishefsky, S. J., *J. Am. Chem. Soc.* **2000**, *123*, 35.

136. (a) Priem, B.; Gilbert, M.; Wakarchuk, W. W.; Heyraud, A.; Samain, E., *Glycobiology.* **2002**, *12*, 235; (b) Murata, T.; Morimoto, S.; Zeng, X.; Watanabe, S.; Usui, T., *Carbohydr. Res.* **1999**, *320*, 192.

137. Albermann, C.; Piepersberg, W.; Wehmeier, U. F., *Carbohydr. Res.* **2001**, *334*, 97.

138. Drouillard, S.; Driguez, H.; Samain, E., *Angew. Chem. Int. Ed.* **2006**, *45*, 1778.

139. (a) Eisele, T.; Windmüller, R.; Schmidt, R. R., *Carbohydr. Res.* **1998**, *306*, 81; (b) Vermeer, H. J.; Kamerling, J. P.; Vliegenthart, J. F. G., *Tetrahedron: Asymmetry* **2000**, *11*, 539; (c) Liu, M.; Yu, B.; Hui, Y., *Tetrahedron Lett.* **1998**, *39*, 415; (d) Liu, M.; Yu, B.; Wu, X.; Hui, Y.; Fung, K.-P., *Carbohydr. Res.* **2000**, *329*, 745; (e) Garegg, P. J.; Norberg, T., *Carbohydr. Res.* **1976**, *52*, 235; (f) Takeo, K. i.; Aspinall, G. O.; Brennan, P. J.; Chatterjee, D., *Carbohydr. Res.* **1986**, *150*, 133.

140. Vermeer, H. J.; van Dijk, C. M.; Kamerling, J. P.; Vliegenthart, J. F. G., *Eur. J. Org. Chem.* **2001**, 193.

141. (a) Inamura, S.; Fujimoto, Y.; Kawasaki, A.; Shiokawa, Z.; Woelk, E.; Heine, H.; Lindner, B.; Inohara, N.; Kusumoto, S.; Fukase, K., *Org. Biomol. Chem.* **2006**, *4*, 232; (b) Hamza, D.; Lucas, R.; Feizi, T.; Chai, W.; Bonnaffé, D.; Lubineau, A., *ChemBioChem.* **2006**, *7*, 1856; (c) Ronchi, S.; Compostella, F.; Lay, L.; Ronchetti, F.; Toma, L., *Eur. J. Org. Chem.* **2005**, 4459.

142. Windmüller, R.; Schmidt, R. R., *Tetrahedron Lett.* **1994**, *35*, 7927.

143. (a) Youssef, R. H.; Azmy Silwanis, B.; El-Sokkary, R. I.; Nematalla, A. S.; Nashed, M. A., *Carbohydr. Res.* **1993**, *240*, 287; (b) Koto, S.; Hirooka, M.; Tashiro, T.; Sakashita, M.; Hatachi, M.; Kono, T.; Shimizu, M.; Yoshida, N.; Kurasawa, S.; Sakuma, N.; Sawazaki, S.; Takeuchi, A.; Shoya, N.; Nakamura, E., *Carbohydr. Res.* **2004**, *339*, 2415; (c) Takeo, K. i.; Imai, T., *Carbohydr. Res.* **1987**, *165*, 123; (d) Zheng, G.; Graham, A.; Shibata, M.; Missert, J. R.; Oseroff, A. R.; Dougherty, T. J.; Pandey, R. K., *J. Org. Chem.* **2001**, *66*, 8709; (e) Okamoto, K.; Kondo, T.; Goto, T., *Tetrahedron* **1987**, *43*, 5919.

144. Matsuzaki, Y.; Ito, Y.; Ogawa, T., *Tetrahedron Lett.* **1992**, *33*, 4025.

145. Tomoo, T.; Kondo, T.; Abe, H.; Tsukamoto, S.; Isobe, M.; Goto, T., *Carbohydr. Res.* **1996**, *284*, 207.

146. Lay, L.; Windmüller, R.; Reinhardt, S.; Schmidt, R. R., *Carbohydr. Res.* **1997**, *303*, 39.

147. (a) Nicolaou, K. C.; Caulfield, T.; Kataoka, H.; Kumazawa, T., *J. Am. Chem. Soc.*. **1988**, *110*, 7910; (b) Xia, C.; Schümann, J.; Emmanuel, R.; Zhang, Y.; Chen, W.; Zhang, W.; De Libero, G.; Wang, P. G., *J. Med. Chem.* **2007**, *50*, 3489.

148. (a) Rundlöf, T.; Widmalm, G., *Magn. Reson. Chem.* **2001**, *39*, 381; (b) Jenkins, G. A.; Bradbury, J. H.; Messer, M.; Trifonoff, E., *Carbohydr. Res.* **1984**, *126*, 157; (c) Ishizuka, Y.; Nemoto, T.; Fujiwara, M.; Fujita, K.; Nakanishi, H., *J. Carbohydr. Chem.* **1999**, *18*, 522.

149. (a) Chapter 1. Part 1: Novel Synthesis and Biological Evaluation of 1,5-Linked D-Mannoseptanosides. Part 2: Synthesis of D-Glucoseptanosides

Chapter 2. Biomimetic Total Synthesis of the Proposed Structure of ent-Muzitone. Boone, M. A, Emory University, Thesis, 2009; (b) Boone, M. A.; McDonald, F. E.; Lichter, J.; Lutz, S.; Cao, R.; Hardcastle, K. I., *Org. Lett.* **2009**, *11*, 851.