University of Arkansas, Fayetteville
ScholarWorks@UARK

Theses and Dissertations

8-2018

Conformationally-Controlled Late-Stage Modifications for SAR Studies of the C-3-Glcp Moiety of Ipomoeassin F

Lucas Whisenhunt
University of Arkansas, Fayetteville

Follow this and additional works at: https://scholarworks.uark.edu/etd

 Part of the Cancer Biology Commons, Cell Biology Commons, Medicinal-Pharmaceutical Chemistry Commons, and the Organic Chemistry Commons

Recommended Citation
Whisenhunt, Lucas, "Conformationally-Controlled Late-Stage Modifications for SAR Studies of the C-3-Glcp Moiety of Ipomoeassin F" (2018). *Theses and Dissertations*. 2926.
https://scholarworks.uark.edu/etd/2926

This Dissertation is brought to you for free and open access by ScholarWorks@UARK. It has been accepted for inclusion in Theses and Dissertations by an authorized administrator of ScholarWorks@UARK. For more information, please contact scholar@uark.edu, ccmiddle@uark.edu.

Conformationally-Controlled Late-Stage Modifications for SAR Studies of the C-3-Gl*c*p Moiety of Ipomoeassin F

A dissertation submitted in partial fulfillment
of the requirements for the degree of
Doctor of Philosophy in Chemistry

by

Lucas Whisenhunt
Henderson State University
Bachelor of Science in Chemistry, 2012

August 2018
University of Arkansas

The dissertation is approved for recommendation to the Graduate Council.

_____
Wei Shi, Ph.D.
Dissertation Chair

_____
Julie Stenken, Ph.D.
Committee Member

_____
Susanne Striegler, Ph.D.
Committee Member

_____
Suresh Thallapuranam, Ph.D.
Committee Member

_____
Neil Allison, Ph.D.
Committee Member

Abstract

The resin glycoside, ipomoeassin F has been shown to be extremely potent against multiple cancer lines (IC_{50} = 4.2-36 nM). However, the mechanism of action of this potent and complex natural product is still not fully understood. The *α,β*-unsaturated esters of the glucosyl moiety have been shown to be vital for the overall cytotoxicity of ipomoeassin F. Nevertheless, the importance of the tigloyl ester of the glucosyl moiety is still largely unknown. This work aimed to study the pharmacophore importance of the tigloyl ester by creating, an efficient, scalable, and flexible synthesis route for various analogs. The 18-linear step synthesis utilized multiple regio-selective and chemo-selective reactions, while not affecting the highly functionalized (1→2)-*β*-disaccharide moiety. The synthesis route modified the C-3 glucosyl position in the penultimate step, making medicinal chemistry studies of the tigloyl moiety possible. The late-stage conformationally-controlled highly regio-selective esterification allowed for the completion of ipomoeassin F and other tigloyl modified analogs. The in-house cytotoxicity data conveyed that large aromatic modifications greatly reduced the cytotoxicity against multiple cancer cell lines. The ipomoeassin F was submitted for NCI-60 cell line screening that showed good sensitivity against breast, renal, and melanoma cell lines (GI_{50} = ~30 nM). To determine the importance of *α,β*-methyl groups of tigloyl moiety, acrylic modifications were envisioned but were difficult to obtain using the developed synthesis route. Therefore, we believe the *α,β*-unsaturated double bond should be tri-substituted to effectively study the tigloyl moiety. This work advanced the scientific knowledge of an underexplored natural product with an unknown mechanism of action and unreached therapeutic potential.

© 2018 by Lucas Whisenhunt
All Rights Reserved

Acknowledgements

I thank my family for the unconditional love, sacrifice, and guidance since I was born. Regardless of where life has taken me, they have always been a phone call or text away.

I am forever thankful to my research advisor, Dr. Wei Shi, for openly accepting me into the group, never allowing me to settle, and guiding me to become a better chemist. His deep knowledge in the field of resin glycosides and curiosity to learn more about carbohydrate chemistry will benefit my entire life as a chemist. I thank Drs. T.M.S. Kumar and Julie Stenken for always reminding me of the "bigger" picture of total synthesis. I thank Dr. Allison for all his advice and calming attitude in my research projections. I also thank Dr. Striegler for her suggestions and guidance in the disaccharide synthesis of ipomoeassin F.

I thank Drs. John Hardee and David Bateman from Henderson State University for opening my mind to the possibility of graduate school and their continued support.

I thank all the members of the Shi group, especially Dr. Mugunthan Govindarajan, Dr. Gunaghui Zong, and Eric Barber for teaching total organic synthesis and become lifelong friends. I also thank Drs. Zhijian Hu and Hazim Al-jewari for their work and assistance in the biology lab. I also thank Dr. Jiang Wang for being my roommate and the ultimate friend.

I thank Celeste Otto for her unconditional support and patience during this journey. Thanks for being my life partner and teaching me to always put others before self.

Most importantly, I thank Jesus Christ accepting me as an imperfect sinner, giving me an eternal hope, and showing me the meaning of Romans 8:28.

Dedication

This work is dedicated to the millions of people that have and will succumb to cancer, especially my grandfather, Coach, and best friend Ronald Whisenhunt. Each of their stories serve as motivation for scientists, like myself, to decrease the impact cancer will have on our future.

Table of Contents

List of Figures

List of Schemes

List of Tables

Abbreviations

μM – micromolar

A2780 – human ovarian cancer cell line

ABPP – activity-based protein profiling

Ac – acetyl or acetic acid

Alloc – allyloxycarbonyl

B-M-S – Bristol-Myers-Squibb

Bn – benzyl

C_{14} ¬tetradecanoic acid

C_{16} ¬ hexadecanoic acid

CA – carboxylic acid

cinn ¬ cinnamoyl or cinnamic acid

CMPI – 2,4,6-trichlorobenzoyl chloride

CSA – camphorsulfonic acid

DABCO - 1,4-diazabicyclo[2.2.2]octane

DCC – *N*,*N*'-dicyclohexylcarbodiimide

DCE – dichloroethane

DCM – dichloromethane

DDQ – 2,3-Dichloro-5,6-Dicyanobenzoquinone

DMAP ¬ 4-dimethylaminopyridine

DMF – Dimethylformamide

DTS – Diverted Total Synthesis

E – trans double bond

E7389 – Eribulin clinical anti-cancer drug

Et_2O – diethyl ether

EtOH – ethanol

FDA – Federal and Drug Administration

Fuc*p* – fucose

Gal – galactose

Glc*p* – glucose

Gr – Grubbs

H522-T1 – non-small cell lung cancer cell line

HCl – hydrochloric acid

Hex – hexanes

H-G – Hoveyda-Grubbs

HPLC – High-performance liquid chromatography

hrs. – hours

HT-29 – human colorectal adenocarcinoma cell line

iba – isobutyric

Ipomo – Ipomoeassin

KOH – potassium hydroxide

KOMe – potassium methoxide

mba – (2S)-methylbutyric

MDA-MBA-435 – metastatic human breast cancer cell line

MeCN – acetonitrile

MeOH- methanol

MS – molecular sieves

n – number of carbons

nla – (2R,3R)-3-hydroxy-2-methylbutyric

nm – nanometer

nM – nanomolar

NME – new molecular entity

NMR – Nuclear Magnetic Resonance

Pen – pentanes

Ph – phenyl

PMB – 4-methoxylbenozyl

PMP– 4-methoxylphenyl

POC – proof of concept

PPI – Protein-Protein Interaction

PySSPy – 4,4'-dipyridyl sulfide

Qui – quinovose

RCAM – ring closing alkyne metathesis

RCM – Ring Closing Metathesis

RCYM – Ring Closing Alkyne Metathesis

Rha – rhamnose

RT – room temperature

SAR – structure -activity relationships

SL – sophorolipid

TASF – trissulfonium difluorotrimethylsilicate

TBS – tert-butyldimethylsilyl

TFA – trifluoracetic acid

THF – tetrahydrofuran

tig – tiglic or tigloyl

TMS – trimethylsilyl

TMSCl – trimethylsilyl chloride

TMSOTf -trimethylsilyl trifluoromethanesulfonate

Tol – toluene

TPP – triphenyl phosphine

TsOH – *p*-toluenesulfonic acid

U937 – human lung(lymphoblast) cell line

WHO – World Health Organization

Xly – xylose

Z – cis double bond

α – alpha configuration

β – beta configuration

CHAPTER 1. BACKGROUND AND INTRODUCTION

1.1. Natural Products

Over the past century, the synthesis of complex natural products has been a challenging yet rewarding task for organic chemists around the world. This field of study, called total synthesis, has bettered the lives of billions of people around the world and made modern life possible. From pharmaceuticals[1] to pesticides[2], fragrance [3-5] to food flavorings[3, 6, 7], countless lives have been positively affected by the synthesis of complex natural products. A natural product can be defined as any chemical compound or substance isolated from a living thing. Historically, the major factors inhibiting the developed of natural products has been structural complexity and limited quantities. Often a natural product is difficult to isolated in pure form, produced in trace amounts (<10 mg), or the biomass is limited (<0.01% by weight). So, total syntheses have been developed to prepare larger quantities of a natural product for further studies (mg to gram scale). There are many different classes of natural products from numerous sources like plants,[8] bacteria,[8, 9] and even microorganisms.[10, 11] However the total syntheses of natural products are still far from routine. In the following chapters we will look at a class of natural product called resin glycosides, specially ipomoeassins, isolated from the morning glory flower.

1.2. Cancer

In 2015, the Worldwide Health Organization (WHO) reported cancer was responsible for 1 in 6 deaths worldwide and 8.8 million people have died to this currently, incurable disease. The United States alone accounted for almost 1.7 million new cancer cases and over 600,000 cancer deaths last year.[12] Even though great advances have been made over the past few decades, such as structure-based drug design,[13-15] protein crystallography,[16, 17] binding methods (surface plasmon

resonance, isothermal calorimetry, *etc.*), and the human genome project, cancer still remains a seemingly never-ending battle.[1, 18] The term "cancer" is said to go back to the Greek physician Hippocrates (460-370 BC), who is considered by many as the "Father of Medicine." He used the Greek words *carcinoma* and *carconis* to explain the crab shaped tumors he observed on his patients. Around 100 BC, the Roman physician Celsus translated the two Greek words to *cancer*, from the Latin word for crab**.** Since this time, people have feared of the term *cancer*.[19, 20]

1.2.1. Natural Products and Cancer Research

Throughout history, cultures have turned to nature for their needs, and the treatment of cancer is no different.[21, 22] The WHO has estimated that between 65-70% of the world is reliant on plant-derived medicines. These "traditional" or "crude" medicines can be very important in underdeveloped countries, where modern health care options are not accessible. In 1983, Hartwell reported over 3000 different plant species are used for the treatment of cancers.[23] In 2000, more than 350 plant species were added to Hartwell's list. However, these numbers can be misleading because the claims of efficacy generally apply to the treatment of a patient's skin. So, many of these treatments should be looked at with skepticism. That said, a few examples of plant-derived anti-cancer agents have been directly or indirectly linked back to ethochemical uses.[24] One example of a direct relationship is the tree *Podophyllum peltatum*, odophylltoxin, and the clinically anti-cancer agent etoposide. Commonly known as the Mayapple or the American mandrake, this tree has a variety of medicinal uses, including the treatment of warts and skin cancers. These traditional uses caused an investigation of the roots resulting in the isolation of podophyllotoxin, the active agent. Finally, a shortened synthesis was developed from the stereoisomers of

epipdophyllotoxin. Currently, etoposide is marketed clinically for the treatment of different skin cancers.[25-27]

1.2.2. Problems with Drug Discovery and Cancer Research

Many factors have contributed to cancer becoming a major problem in modern society. Some of the reasons range from the detection or treatment of the cancer,[28] the social habits of the patient, and the cost of treatment.[29, 30] Researchers cannot control a patient's social activities or government funding, but scientists and physicians can identify better targets, prepare more effective, less toxic drugs, and attempt to control the metastasis once the tumors have developed. In this section some problems with drug discovery and cancer research will be examined in greater detail.

The difficulty of finding "druggable" targets and target identification are problems in all areas of drug discovery. In general, a "druggable" target is anything that can be modified or inhibited by a drug (e.g. nucleic acid, peptide, or protein). Even with advances in the human genome, the number of druggable targets remains very low.[31, 32] In 2015, the FDA stated that only a quarter of the newly approved drugs attack novel or previously un-druggable targets. Later the same year, the FDA stated that ten target families accounted for more than three-fourths (78%) of all FDA-approved new molecular entities (NME).[9, 33-36] Research has shown potential target classes like GPCRs, ion channels, and nuclear receptors remain largely unstudied. Kinhlberg *et al.* stated that only 3000 out of the approximately 30000 genes in the human genome are related to diseases. Less than half of these, 600-1500 genes, are considered targets for inhibition for "traditional" small molecule drugs.[37] So, there is a need and opportunity to increase the number of therapeutic targets.[31, 32] Many pharmaceutical companies only focus on druggable targets

because there is a higher chance for proof of concept (POC) and a greater percentage the NME will proceed to phase 3 trials. One reason for the narrow focus is the large amount of literature data on relatively few "druggable" targets. Chemical biologists and medical chemists have argued and debated the terms "druggable" or "undruggable" target. Fortunately, there is not a single answer for this difficult question. One clear conclusion is researchers in all areas of life sciences (organic chemistry, chemical biology, etc.) must work together to expand the number "druggable" targets for significant progress to be made against diseases such as cancer.

Although there is not a perfect target to inhibit a specific disease, including cancer, here are a few rules that medicinal chemists need to consider (Table 1.1).[38, 39] The most important aspect is the availability of human data, whether it is basic research or preclinical data from tissue

Table 1.1. Some Characteristics of an "Ideal" Drug Target
1. Target is selectivity for the drug of interest (toxicity).
2. Can prove the target is being modified and the pathophysiology of the disease is changing.
3. Modification of the target will not affect normal physiological conditions or possible diseases.
4. Ability to forecast or predict possible side reactions from the phenotype data (e. g. animal model or mutation databases)
5. The target is not evenly spread throughout the body.
6. Ability to obtain a 3-D structure conformational change during the drug-target interaction.
7. Chemical Assays of the target are favorable for high throughput screening.
8. Not many other drugs competing for the target (helps with intellectual property situations and gives more freedom to design drug).

taken from an affected subject. Unwanted side effects will occur when any potential therapeutic is introduced to a biological system, so being able to predict the unwanted side reactions is critical. This can save the pharmaceutical company significant time and financial capital predicting side effects that could happen in humans from a simpler model (mice or pig model, mutation databases, etc.).[38, 40] Similarly, the "undruggable" or "difficult-to-drug" targets have common characteristics. The binding sites of these targets are generally devoid of functionality, either extremely lipophilic or high polar, fluid, or flat.[41, 42] Often, a lack of functionality refers to the limited opportunity for hydrogen bonding. Interestingly, the inability to develop drugs with good oral bioavailability has caused some potential targets to be abandoned and viewed as "undruggable."[37]

Protein-protein interactions (PPI) offer an interesting opportunity to develop new treatments for diseases like cancer. The human proteome is predicted to have between 100,000 and 1,000,000 PPI.[17, 43] Even if only a small number of these PPI are found to be druggable for diseases like cancer, it could be a great prospect for future chemotherapeutic agents. Another interesting aspect in contemporary drug discovery is the often-naïve concept of "one drug-one target." Many clinical examples have been shown to exhibit polypharmacology by inhibiting multiple targets.[44, 45] This makes the number of potential targets almost limitless from the different targets combination that could be utilized. However, this "limitless space" for new chemotherapeutic agents is met with a heightened cause for safety concerns.[46, 47]

1.2.3. Drug-Target Interaction and Validation

The other major obstacle in drug discovery, specifically cancer research, is target validation.[48, 49] It is very important for pharmaceutical companies to validate the drug-target interaction before scaling-up a potential therapeutic agent. Many times target validation is aided

by the development of a chemical probe molecule.[50] The probe molecule is structurally similar to the drug but has some key chemical and structural differences.[48] Although the standards for a chemical probe differ, the criteria for the probe should be more severe than the potential drug because of the increased functionality (Table 1.2). This can sometimes lead to unwanted off-target interactions. One key criteria in the chemical probe is the inactive chemical probe as a negative control. The inactive probe is often an inactive enantiomer of the potential drug.[48]

The medicinal chemists Bunnage, Piatnitski, and Jones, from Pfizer, recently investigated the failure of more than 40 drug programs and tried to determine the reasons for their inefficacy.[51] This led them to coin the term "four pillars of cell-based target validation" (Figure 1.1). The first pillar involves confirmation of cellular penetration and the validation of a chemical probe at the active site of the target (Table 1.2). Pillar 1 confirms the on-target activity of the probe and makes sure the response for the chemical probe is not due to off-target hits. Confirming the drug-target interactions should be one of the first tests conducted, because false positives could lead to misleading pharmacological (pillar 3) or phenotypic (pillar 4) results. Often, techniques like LC-

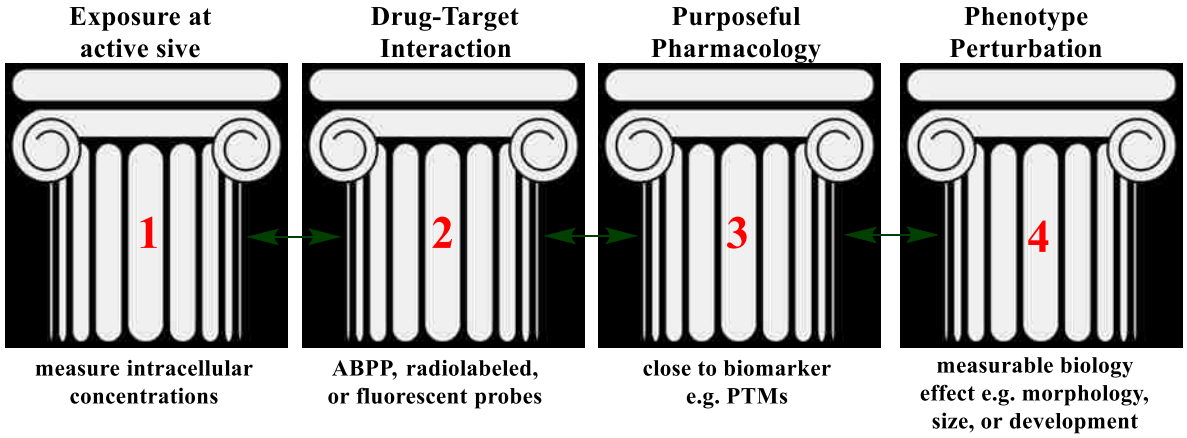
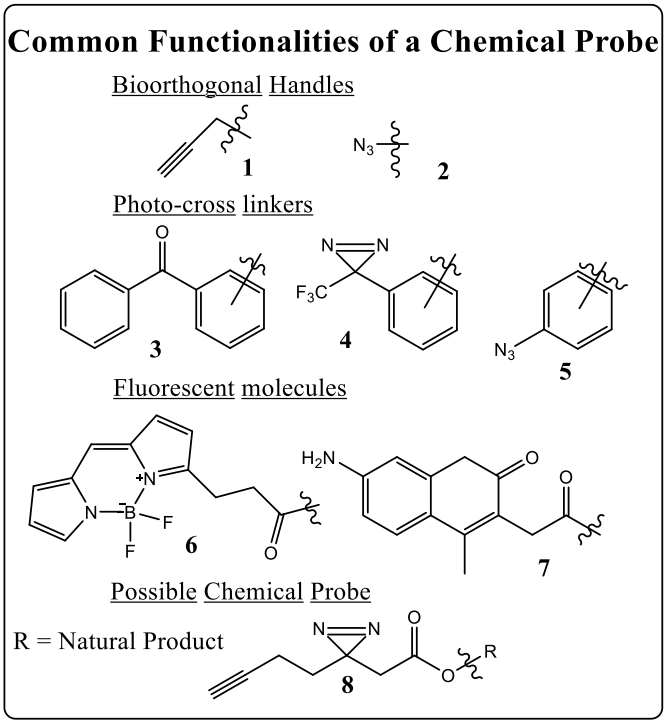


Figure 1.1. The four pillars of cell-based drug-target interaction and validation coined by Pfizer.

MS can be utilized to measure the in-cell concentrations of cell extractions. Sometimes, MS must be combined with other techniques like sophisticated microscopic imaging or radiometric methods because of changing local concentrations of specific subcellular compartments. Pillar 2 builds on pillar 1 and involves selectivity studies and probe-engagement. When designing a chemical probe, the chemist has to be thoughtful of the highly reactive functional groups.[48] These groups can potentially cause unwanted side-reactions, leading to misleading or even false conclusions. Pillar 2 is often found to be the most important, because it links pillar 1 (active site confirmation) to pillars 3 (pharmacology) and 4 (phenotype). Unlike the studies to optimize the selectivity or potency (pillar 1), the determination of a probe's performance can be very challenging for in vivo or live cell studies.

Table 1.2. Criteria for developing and designing a functional chemical probe.

Similar activity to target molecule
Selective (>100 fold)
Potent in in biochemical assay (<100 nM)
Potent in cellular assay (<100 nM)
Aqueous solubility
Several active and chemical classes
Negative control (e.g. enantiomer)
Cell permeability (active/passive transport)



Luckily, major developments have been made in the past decade with techniques like activity-based protein profiling (ABPP) to determine the drug-target interaction. Pillar 3 involves measuring the expression of pharmacology and creating assays to measure the pharmacological effects of the chemical probe. Pillar 4 includes the development of assays to prove or disprove phenotype perturbation in a human disease. The degree of "translatability" of these results from patient to patient should always be considered. Lastly, the researchers should consider factors like nonspecific cell death because this can lead to a waste of time and resources. The "four pillars of cell-based target validation" can help with the target validation of a small molecule, but this is still a risky endeavor.[48] Scientific literature is full of examples of failed projects and un-bridled claims of strong drug-target interactions.[36, 38, 39] Medical chemists, chemical biologists, and biochemists must work together to increase the number of "druggable" targets to make advancements in the treatment of major diseases, such as cancer.

1.3. Organic Synthesis and Medical Chemistry to Advance Cancer Research

The problems with current anti-cancer chemotherapeutic agents range from their extremely difficult syntheses,[52] high cost,[29, 53] and unwanted side-effects.[36, 38, 39] Organic chemistry, specifically medicinal chemistry tries to limit some of these problems by synthesizing biologically active compounds.[18] Organic chemists use diverse chemical transformations and cheap, commercially available starting materials to construct a target molecule for a particular application. An organic chemist may or may not know the application of the target molecule he/she is trying to synthesize, often the challenge of constructing the molecule being the driving force for the research. A medicinal chemist often already knows the application of his/her target molecule, whether to inhibit a disease or upregulate a biological process.

The importance of natural products in drug discovery and cancer research has been stated by numerous reviewers.[1, 24, 54, 55] In 2016, Newman and Cragg published a review on the NCEs from 1981 to 2014.[1] They stated that almost 60% of the 1500 NCEs were natural products, natural products derivatives, or mimicked natural products. For anti-cancer drugs, 83% of the approved chemotherapeutic agents (113 of 136) were natural products or directly derivative.[56] Still, some pharmaceutical companies have moved away from natural products, selling off the rights or closing entire natural product departments. The reasons for the shift away from natural products have been debated. Some of the commonly examined reasons include an inability to reproduce spectroscopic evidence of the natural product during the synthesis, problems scaling-up or producing enough NCEs for commercialization, and difficult to impossible chemical transformations.[52, 57, 58] Despite the problems, most major pharmaceutical companies believe natural products and their derivatives will continue to play a major role in drug discovery and cancer chemotherapy.

The reason for the high percentage of anti-cancer agents being based on natural products has been debated (Table 1.3).[56] One accepted reason is that the complex chemical structure of most natural products can interact with a larger number of biological targets.[1, 18] This may explain why nature has been a leading source of compounds for drug development, when compared to combinatorial libraries and small-molecules. Most natural products exhibit a trend when compared to small molecule drugs.[59] The last three points in limit off-target interactions between the natural product and biological environment.[49] Extreme toxicity is a major problem of cancer chemotherapies and one of the leading causes for attrition during clinical trials.[60]

Table 1.3. *Common Characteristics of Natural Products Compared to Small-Molecules or Combinatorial Libraries*

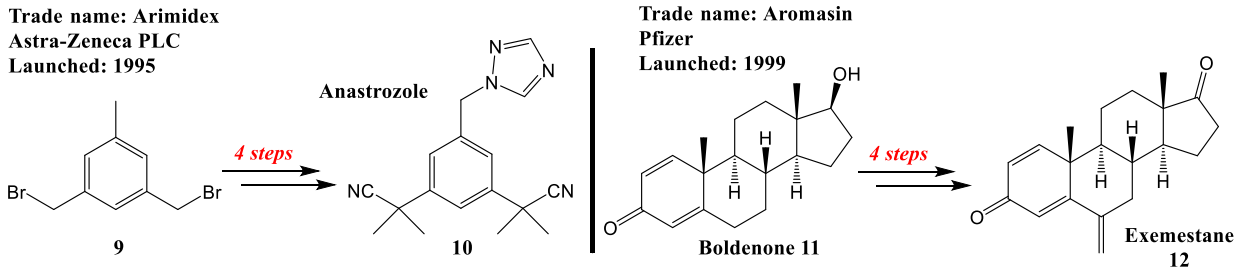
1. More complex ring structure (bridged, fused, and spiro ring fusions)
2. Less nitrogen, sulfurs, and halogens (helps with toxicity)
3. More H^+ bonding (both donors and acceptors)
4. Less rotatable bond but more flexible overall structure
5. Larger number of heteroatoms (oxygen, less common nitrogen)
6. More hydrophilic than many small molecules (especially A. compounds)
7. High percentage of sp^3 carbons (more 3-D structure)

Unfortunately, few examples can be found of a directly isolated natural product being used as the actual drug. Often, the natural product acts as a structural scaffold or lead compound for the synthesis of a library of compounds or analogs. During this time, the organic chemists must work very closely with computational chemists and chemical biologists to determine the pharmacophoric "hot spots" and the moieties of the molecules that are vital to biological activity.[24, 54, 59] Lipinski's rule of five has acted as a guide for medicinal chemists to design potential therapeutic agents with the correct physiological properties and low cytotoxicity.[37, 60] The development of SAR libraries takes significant time and patience to find a molecule with good pharmacological properties. Through collaborative studies, an organic chemist can start to determine the important SAR of a molecule and determine the best synthesis method.

1.3.1. Total Synthesis

Total synthesis is the classic technique for the preparation of complex natural products. Structural complexity and chemical challenges have driven groups from around the world to construct natural products.[1, 24, 54, 55] To begin the total synthesis procedure, a chemist starts from a cheap, commercially available starting material. Then, he/she performs a series of chemical transformations to add structural moieties. Due to the structural complexity, stereochemistry or regio-chemistry may need to be considered for an intermediate with multiple reaction sites. To limit this problem, organic chemists utilize protecting groups or functionalities that can be added and removed without affecting the rest of the molecule. Finally, after a series of chemical transformations, a complex intermediate is obtained, and all the protecting groups can be removed to make the natural products. The total syntheses of natural products fall into two main categories: linear and convergent.

For a linear synthesis, the target molecule is obtained from a series of straight-line reactions from one starting material.[61, 62] During a linear synthesis, a series of chemical transformations are performed until the target compound is reached. Scheme 1.1 shows examples the linear syntheses of anastrozole **10** and exemestane **12** from commercially available starting materials. These drugs are used to treat different types of breast cancers thought an irreversibly binding of Type II



Scheme 1.1. Linear synthesis of Anastrozole **10** (Arimidex) and Exemestane **12** (Aromasin).

aromatase inhibitor. A convergent synthesis utilizes two or more starting materials and the different moieties of the natural product are synthesized independently. Then, a coupling reaction between two separate moieties to form a more complex intermediate. This process along with other chemical transformations are performed until the target natural product is completed.

A convergent synthesis offers specific advantages and benefits over a linear synthesis. One of the main problems with a linear synthesis is the overall yield quickly drops with each chemical transformation. This means most clinical drugs utilizing a linear synthesis are at the maximum 6-8 steps.[1, 25, 54] A convergent synthesis limits this problem by separately synthesizing the different moieties of the natural product. This gives the organic chemists more material to use to optimize reaction conditions and test other factors. Another benefit is analogs can be synthesized for SAR studies without greatly modifying the synthesis route.[126] The amount of time to synthesize a natural product can often be a major limiting factor in drug discovery. Designing a convergent synthesis route can help lessen the time and material needed to prepare a complex natural product because different moieties can prepare concurrently.

1.3.2. Diverted Total Synthesis

Sometimes during the total synthesis of a natural product, a pharmacophore of the natural product can be recognized. The pharmacophore is the vital moiety, or moieties, of the natural product that is necessary for biological activity. Biological information along with a good synthetic approach allows for the removal of needless moieties of the natural product. In most situations, the synthesis of the pharmacophore is easier than the natural product.[24] In the mid-2000s, Danishefsky *et al.* created the term "Diverted Total Synthesis" (DTS) to define this method of synthesis.[63] Danishefsky stated in the paper, he was definitely not the first chemist to use this

method.[64] DTS had been used by numerous groups in both industry and academia in the 1970s and 1980s. Because the target molecule can be less complex, the synthesis is usually more efficient, taking less time to make the biologically-active agent. Another benefit is the reduction in cost of chemical reagents and human manpower. These advantages give the organic chemists more time, financial capital, and materials to synthesize analogs with varying degrees of structural complexity based on a simpler pharmacophore.

Prominent examples of the structurally simpler pharmacophores developed into anti-cancer agents are shown in Figure 1.2. Halichondrin B **13** was isolated in the mid-1980s from the marine sponge *Halichondria* genus and found to be cytotoxic against multiple cancer cell lines.[65, 66] During the total synthesis, Kishi *et al.* found the left half of halichondrin B **13** (blue) was not vital to the biological activity.[67, 68] This led them to develop the analogue E7389 **14** that retained all the biological activity of halichondrin B **13**.[69] Besides making the synthesis easier, the analog **14** showed decreased toxicity and greater in vivo stability. E7389 **14** was developed into Eribulin for the treatment of multiple breast cancers.[70-72]

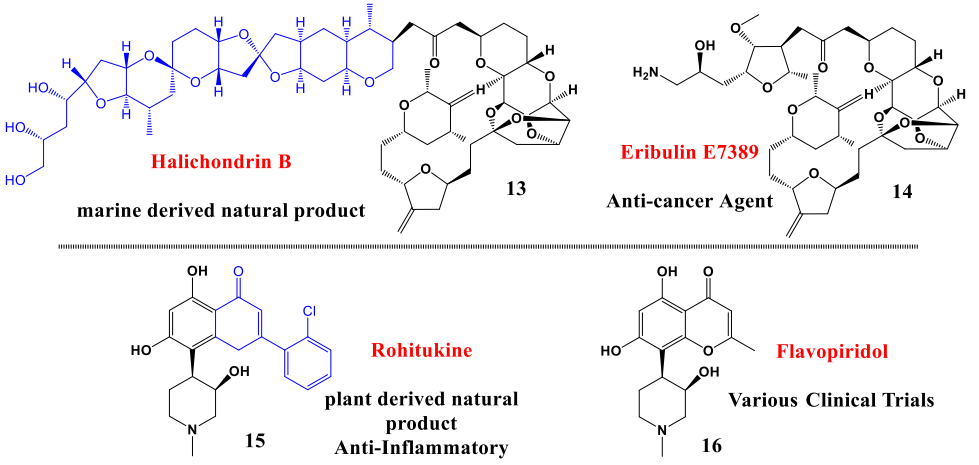


Figure 1.2. Simpler pharmacophores based on a natural product used as anti-cancer agents in clinical medicine.

Flavopiridol **16** was a molecule based on the natural product rohitukine **15** isolated in the early 1990s. Rohitukine **15** was isolated from the Dysoxylum *binectariferum* and is used to treat rheumatoid arthritis. Over 100 analogs were synthesized based on **15**, only flavopiridol **16** was found to inhibit cyclin-dependent kinases and be extremely active against numerous lung and breast carcinoma cell lines.[73] In the past decade, flavopiridol (Alvocidib) has moved into at least nine clinical trials (phase 2 to phase 3) for the treatment of lymphomas, leukemias, and solid tumors. Flavopiridol **16** has also been used in combination with other anticancer agents to increase the potency. Often the isolated natural product is not the chemical entity developed into an anti-cancer agent. Dolastatin 10 **17** is an example of a natural product that failed during clinical trials, while the modified auristatin PE (TZT-1027) **18** is, currently, in clinical trials (Figure 1.3).[24] The removal of the thiazole in **18** caused increased tubulin polymerization, resulting in cell cycle arrest and starting apoptosis. Auristatin PE is being delivered to specific epitopes by use of monoclonal antibodies for the treatment of hematologic cancers.[71, 74, 75] Eribulin **14**, Alvocidib **16**, and Auristatin PE **18** are some examples of a structurally simple compound that contained the biologically important pharmacophore for the treatment of various cancers.

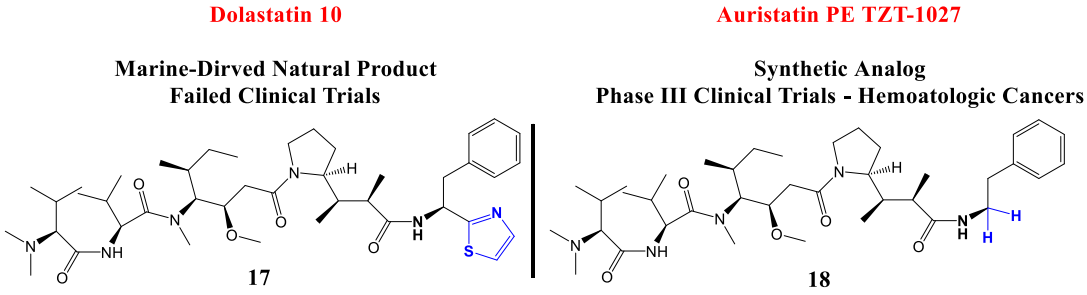
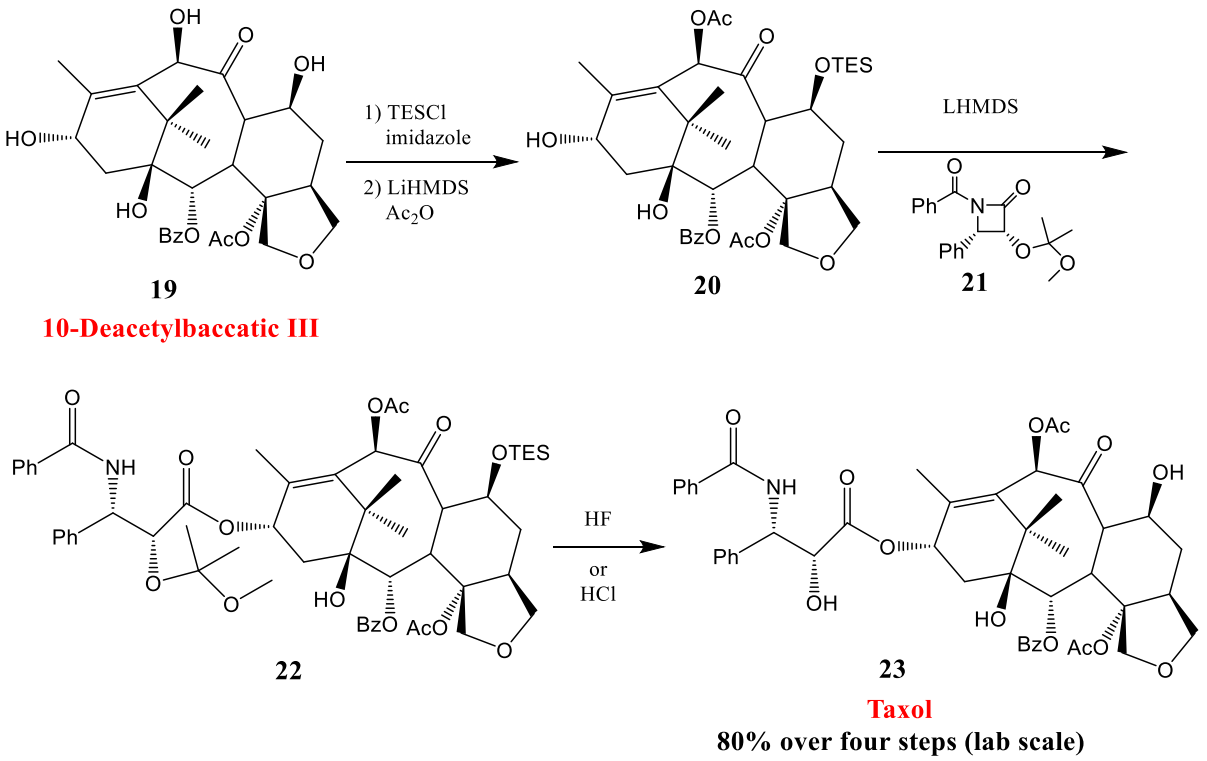


Figure 1.3. The synthetic analog Auristatin PE (Soblidotin or TZT-1027) **18** based on the natural product Dolastatin 10 **17** that was terminated during clinical trials.

1.3.3. Semisynthesis

Starting with a structurally-complex starting material is ideal for the synthesis of a natural product.[25, 54] This method is called the semisynthesis of a natural product. Since a limiting factor in the study of a complex natural product is the amount of material, the development of a semisynthesis can shorten the synthesis of the natural product or a biologically active intermediate.[76] Semisynthetic methods can be very valuable to produce numerous analogs quickly and easily.[71, 77] Often the source of the complex intermediate is from biomass like wood, alcohol fuels, or crops, which are readily available, cheap, and the waste products of other industrial processes. Enzymatic methods are another technique used to install functional groups during a semisynthesis. Enzymes are a great alternative to conventional catalysts and chemical reagents, because enzymes always give the correct product in high yield. Fermenting bacteria has been shown to produce large amounts of complex intermediates or even complete natural product.[76] Unfortunately, some functionalities are difficult to obtain through semisynthetic methods because of functional group tolerance or incompatible reaction conditions. So, a semisynthesis is best utilized for the preparation of the natural product or an already known biologically active intermediate.

One of the best and well-known examples of the impact of a semisynthesis is of paclitaxel **23** (Taxol).[19, 25] The natural product was isolated from the Pacific yew (*Taxus brevifolia*) in 1958 and found to have good therapeutic activity against multiple cancer cell lines (single digit nM). However, paclitaxel **23** could not be commercially developed because of the unusual ring fusion and large number of functional groups. Also, each mature Pacific yew tree (40 feet) would only give approximately 0.5 g of paclitaxel **23**, not allowing direct isolation of large qualities of the



Scheme 1.2. Semi-Synthesis if Taxol from 10-Deacetylbaccatic III.

natural product. This limited the amount of paclitaxel **23** to study, until a semisynthesis was developed from 10-deacetylbaccatin III **19** by Robert A. Holton at Florida State (Scheme 1.2). [78] The 10-deacetylbaccatin III **19** was readily available in large quantities from the needles of multiple *Taxus* species giving a readily available, renewable resource.[79-81] After slight modification in the semisynthesis route, Holton *et al.* obtained a significant amount of **23** for animal testing and commercial use.[78] In the early 1990s, Bristol-Myers-Squibb (B-M-S) purchased the rights to and developed Taxol **23** to treat many different cancers, including breast, ovarian, and specific lung cancers.[11] Taxol **23** has become the most popular chemotherapeutic drug for numerous kinds of cancers and acts as a standard for new potential therapeutic agents.[25,82] The development of Taxol **23** gives an example that shows the potential importance of

semisynthesis for organic chemistry. As enzymic methods and the isolation techniques improve, semisynthesis will become a more important part of natural product synthesis.

1.4. Resin Glycosides

1.4.1. History of Resin Glycosides

Resin glycosides isolated from the morning glory family (Convolvulaceae) have been used as traditional medicines for the treatment of numerous diseases throughout history.[83] The plant species making up the Convolvulaceae family can be found worldwide, but more than 60% are spread throughout Central America.[84, 85] These secondary metabolites are commonly known as glycolipids or lipo-oligosaccharides, a chemical entity special to the morning glory family. The Convolvulaceae family name was derived from the Latin word *convolvo*, meaning wrapping or interlaced vines wrapped around a support. All parts of the plant, including the bulbs, flowers, and the stems have been exploited for different medicinal issues; however, the bulbs have been the common part of the plants exploited (Figure 1.4).[86]

Figure 1.4. The vines and bulbs (seeds) of the morning glory flower. The botanical name, Convolvulaceae, for the morning glory family was derived from the Latin word *convolvo*, meaning interweaving vines. The bulbs or roots were valued for their laxative properties by new world civilizations.

Most of the Convolvulaceae species have been found to possess drastic purgative properties. One of the first recorded uses of a Convolvulaceae species for a therapeutic effect was in Meso-America. Where the Aztecs used multiple species of the tuber-shaped roots to exhibit a diverse range of laxative and cathartic effects (Figure 1.4). More recent studies have shown the roots of genus, *Ipomoea* (*I. purga, I. orizabbensis, I. stans, and I. jalapa*) were most commonly used.[87] During colonial expansion of the Americas a commercial enterprise developed, shipping the roots from the Americas to Europe which continues to the present day. The roots would have been ground to a fine powder and mixed with alcoholic extracts. Sometimes the ground resin glycosides would have been taken alone or in concert with other ingredients to reduce unwanted therapeutic effects. In parts of Mexico the plants were even ground down to a resin and used to prevent invasive weeds during crop production. This ethnopharmacology relationship between resin glycosides and a potential pesticides would be confirmed later in the 20th century.[88] By the end of the 19th century, reports started to be published on the phytochemical and chemical properties of the resin glycosides. However, most of this published data has been recently found to be non-conclusive and even scientifically un-reliable. It was not until the discovery of techniques like HPLC, NMR, and MS that resin glycosides could start to be fully studied in greater detail.[89] Still today, several crude drugs with laxative properties isolated from the morning glory can be found commercially. This shows the potential importance of Old World remedies and why modern techniques should be used to study their potential before finalizing a conclusion.

1.4.2. Structural Diversity and Chemical Complexity

Currently more than 250 resin glycosides have been discovered and fully characterized. The molecular weight of these molecules ranges from a few 100 g/mol to more than 2000 g/mol.[58]

So, there are many resin glycoside congeners that can be observed for this class of natural products.[90] However, most of the resin glycosides have some common structural features in their chemical make-ups (Figure 1.5).[91-94] All resin glycosides are amphipathic secondary metabolites, meaning they contain two distinct regions (hydrophobic and hydrophilic region).[55] These regions make the resin glycoside very interesting to study because they can exist in both a non-polar and polar environment.

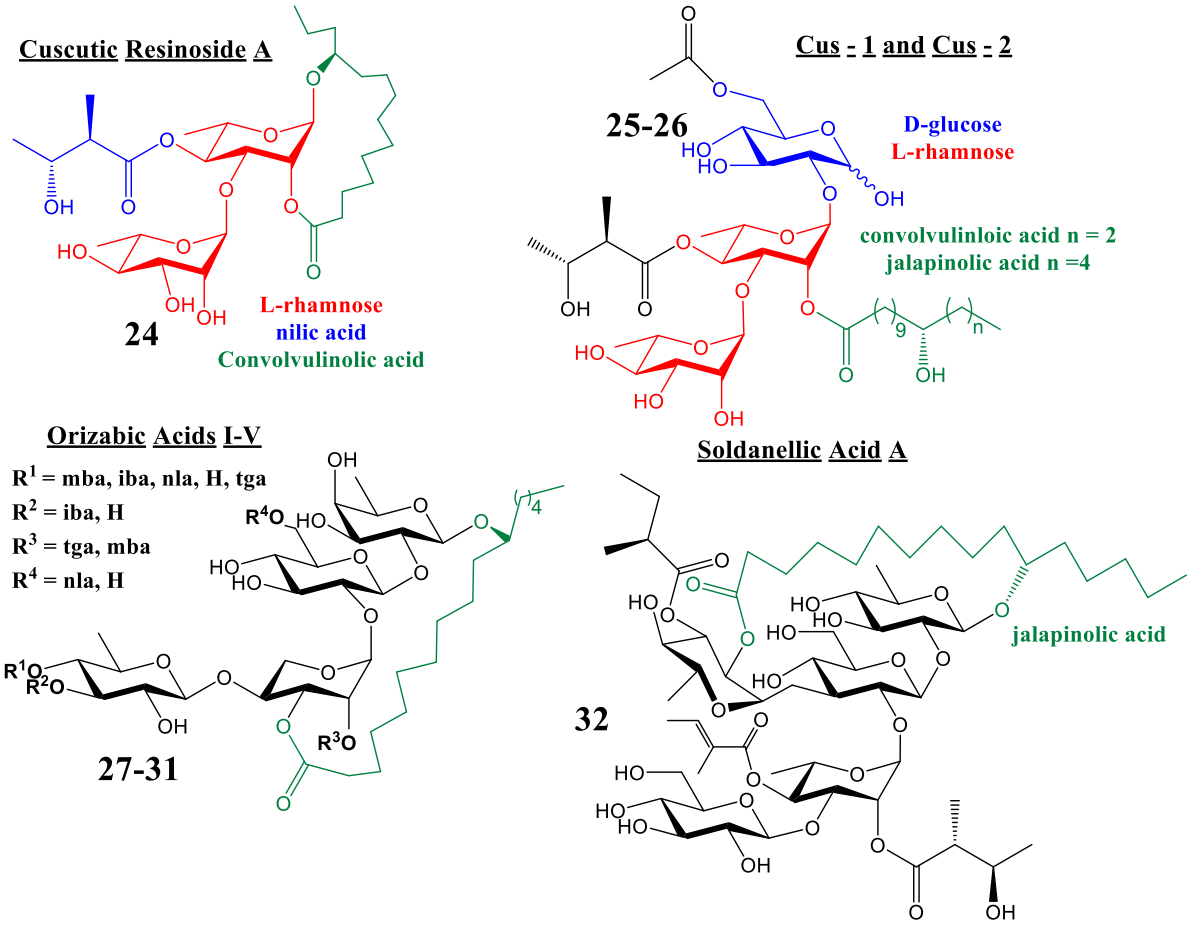


Figure 1.5. Resin glycosides containing different monosaccharides with and without a macrolactone substructure.

The hydrophilic or sugar moiety contains up to six hetero-polysaccharides connected by *O*-glyosidic linkages.[87, 95] However, most of the resin glycosides isolated to date contain a disaccharide core structure (Cusctuic Resinoside A, **24**). This moiety is often called the glycone region of the resin glycoside. The structural diversity in the glycone region comes from two factors, the diversity of the sugars and short-chain fatty acids. Most resin glycosides conatin sugars that are a combination of epimers of pentoses (D-Fuc*p*, D-Qui, L-Rha, and D-Xly) or D-Glc*p*.[87] Few resin glycosides isolated have been found to contain D-Gal or other sugars. Differing fatty acids are the second source of structural diversity in the glycone part of the resin glycoside. These aliphatic acids range from just a few carbons; like acetic (Ac), butyric, 3-hydroxy-2-methylbutyric acid, and propionic acids to longer saturated fatty acids; such as *n*-dodecanoic, *n*-hexanoic, and *n*-hexadecanoic acids. The fatty acids (aglycone region) are always connected to the glycone core of the natural product though a glyosidic bond.

The hydrophobic or aglycone moiety of resin glycosides is made-up of a long chain fatty acid, either tetradecanoic (C_{14}) or hexadecenoic (C_{16}) acid.[87] The structural diversity in the aglycone region comes from the potential macrolactone formation, hydroxylation, or oxygenation. Jalapinolic acid or (11*S*)-hydroxyhexadecanoic acid has been found to be the most common aglycone for resin glycosides (Figure 1.2, green color).[84, 88, 96] The aglycone is always connected to glycone through a glyosidic bond. Often this bond forms a macrocycle or macrolactone across two or more monosaccharides of the glycone region (orizbic acids **27**-**31** and soldanellic acid **32**). However, resin glycosides like, cuscutic acid A_1-A_2 **25**-**26**, contain an acyclic form of the aglycone. For these resin glycosides, the *C-11* hydroxyl does not form a glyosidic bond to the glycone ring. The other difference is the hydroxylation of the resin glycosides. Most of the aglycone fatty acids contain one hydroxyl group (11*S*) which connection the two moieties. However, some examples

contain an aglycone made-up of dihydroxyl fatty acid. The ipomoeassins family of resin glycosides covered later in this section will show some examples of functionality in the aglycone region.

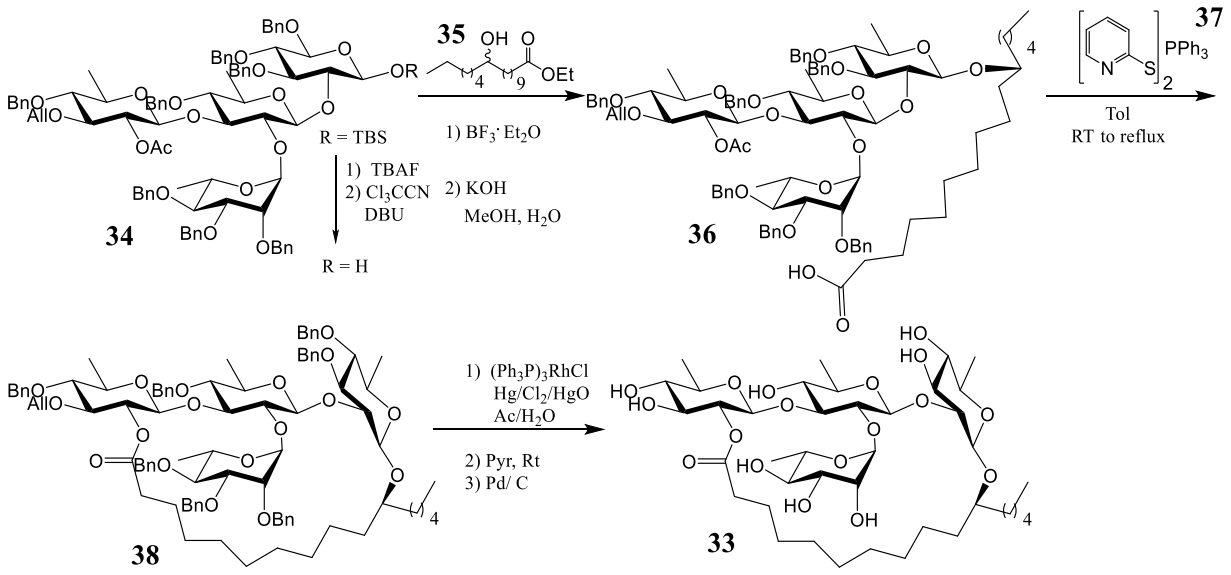
1.5. Common Methods of the Synthesis of Resin Glycosides

1.5.1. Classical Resin Glycoside Syntheses

Despite the advancements in carbohydrate chemistry in the 20th century, only recently have the syntheses of macrocyclic glycolipids and more specifically resin glycosides been successfully completed. Numerous organic groups have made contributions to the resin glycoside field, like Pereda-Miranda, Yang, Xu, and many others.[97] Also, recent advancements have been made to achieve specific glyosidic linkages between the aglycone and glycone moieties. The largest contributions to the glycolipids field has been made by Alois Fürstner *et al.*[98, 99] To date, the preparation of bioactive glycoconjugates can be difficult and far from routine. Still, organic chemists push forward synthesizing complex resin glycosides with interesting structural and biological properties.

1.5.2. Calonyctin A1

Calonyctin A **33** was the first resin glycoside synthesized by Schmidt *et al.* in 1995.[100, 101] Calonyctin A **33** (Scheme 1.3) was a tetrasaccharide that had been shown to inhibit the growth of multiple crops such as beans, peanuts, and sweet potatoes. The tetrasaccharide **34** was constructed through a series of glycosylation reactions using the trichloroacetimidate method. This kind of donor was developed by Schmidt *et al.* in the early 1980s and has become the leading method to make donors for glycosylation reactions.[102-105] The racemic mixture resulted in the 1:1 mixture of



Scheme 1.3. Final steps in Schmidt *et al.* synthesis of calonyctin A **33**.

the glycosylated products that were then treated with KOH and separated to give the carboxylic acid (CA) **36**. At this point, the correct configuration of the aglycone moiety was determined.[100, 101] To form the macrolide**,** an intramolecular coupling was performed under Mukaiyama-Corey conditions with PySSPy **37** and TPP. The 2-pyridyl thioester intermediate **36** had to be heated at reflux and in highly dilute conditions to give the 22-membered macrolactone **38**. The AllOc was isomerized with Wilkinson's catalyst and deprotected by the $HgO/HgCl_2$. Finally, hydrogenation was performed to remove the benzyl protecting group and the total synthesis of calonyctin **33** was achieved. Schmidt *et al.* synthesis of calonyctin A **33** may be inefficient when compared with modern reactions and techniques, but this work opened organic synthesis to resin glycosides.

1.5.3. Tricolorin A and G

The tricolorins are the prototype members and class of the resin glycosides (Figure 1.6). Tricolorins were isolated from the Ipomoea *tricolor Cav* in Mexico where the plant was ground

into a powder and used as a crop protectant for sugar cane.[106, 107] Tricolorin A **39** and G **40** were first synthesized by Heathcock *et al.* [108, 109] in 1996 and by Fürstner *et al.* in 1998.[98, 99]

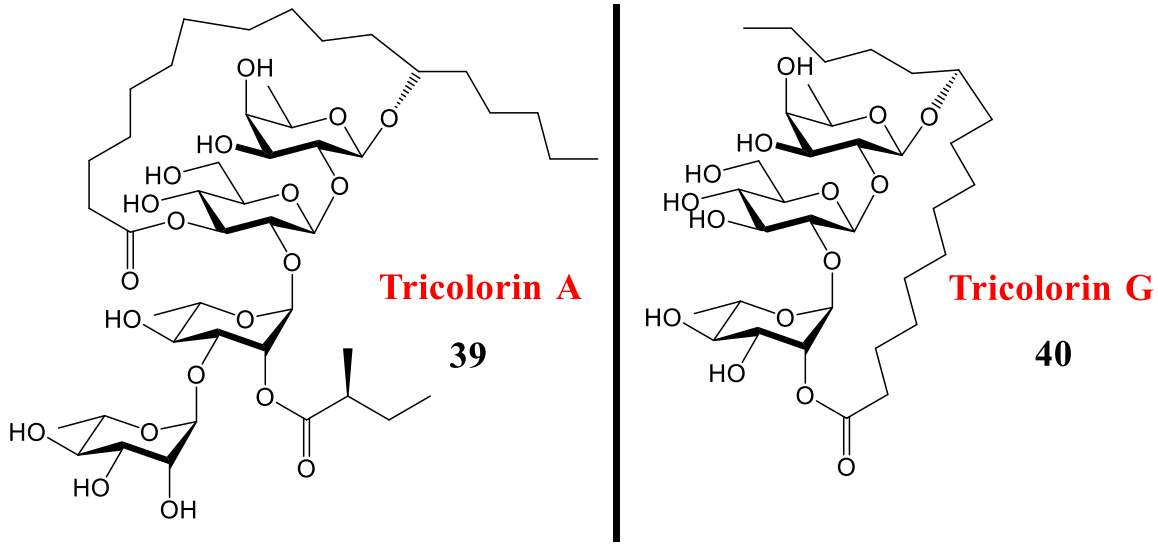
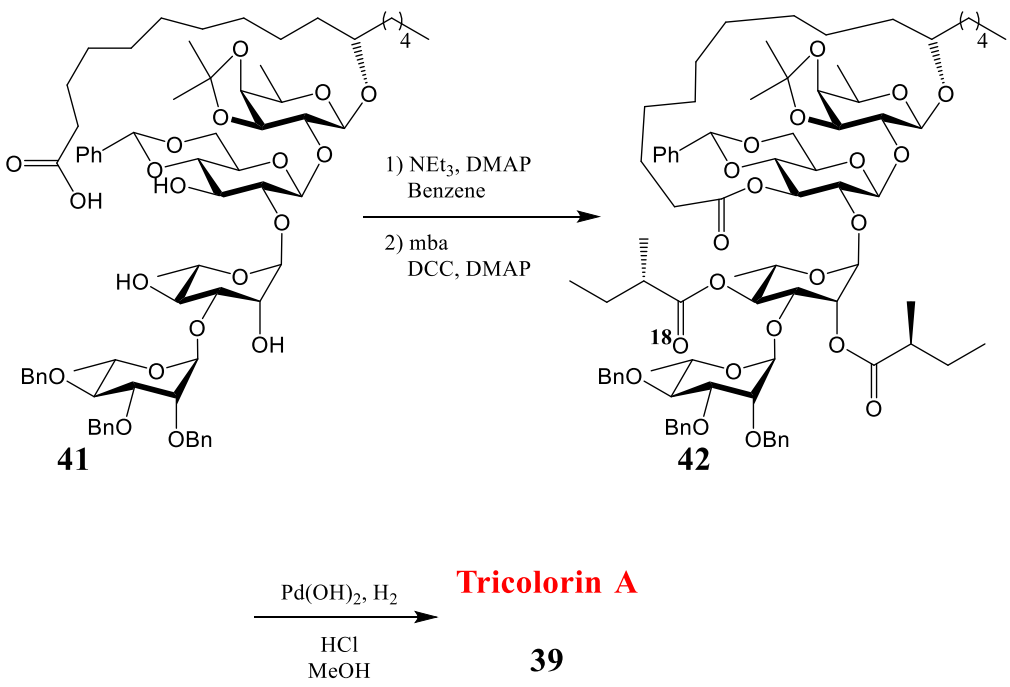


Figure 1.6. Structures of Tricolors A **39** and G **40**.

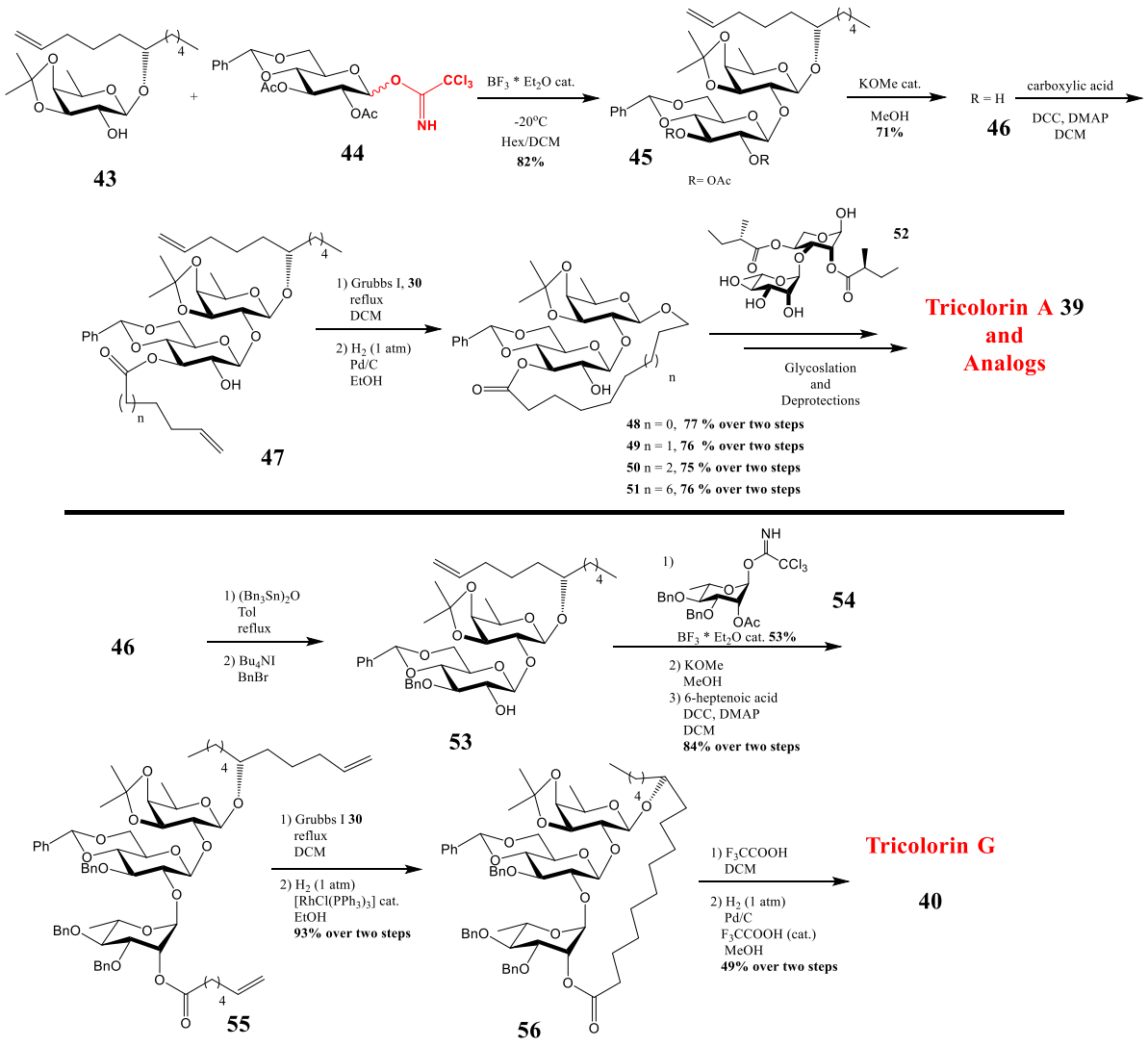
In the synthesis by Heathcock *et al.*, a macrolactonization reaction was performed using Yamaguchi's method to form the macrolactone **42** ring from jalapinolic acid (Scheme 1.4).[109, 110] The macrolactonization method was satisfactory to synthesize the natural products but does not permit a wide range of analogs.[111-113] The resin glycoside contains a large number of free-hydroxyl groups in the glycone region, potentially causing difficulty obtaining the correct regio-chemistry.

Fürstner *et al.* designed a route including ring closing metathesis (RCM) (Scheme 1.5) reaction to form the macrolactone ring of tricolorin A **39** and G **40** (Figure 1.6). The RCM reaction was developed by Grubbs and Hoveyda in the mid-1990s for the formation of new sp^2-sp^2 bonds. This reaction has allowed for the synthesis of numerous resin glycoside and their analogs because



Scheme 1.4. Heathcock's macrolactonization for tricolorin A **39**.

of the wide functional group compatibility.[99] These homogenous transition metal catalysts are stable at RT (solid powder) and readily accessible though many chemical companies. To perform the glycosylation, Fürstner used the Schmidt donor **44** (red) as an effective and reliable way to selectively form the glyosidic linkage. The formation of a Schmidt donor[114-117] (trichloroacetimidate donor) for glycosylation reactions and the RCM reaction have become common synthetic methods for many amphiphilic glycoconjugates.[118] The final few steps in the synthesis of tricolorin G **40** involved the deprotection of benzoyl and isopropylidene protecting groups. The Fürstner *et al.* synthesis of tricolorin A **39** and G **40** gave an example of RCM that improved the flexibility of macrocycle syntheses, allowing for the preparation of analogs for SAR studies. The most common catalysts **57-60** applicable for RCM reactions, are shown in Figure 1.7.



Scheme 1.5. Fürstner *et al.* synthesis of tricolorin A **39** and G **40** using the RCM reaction.

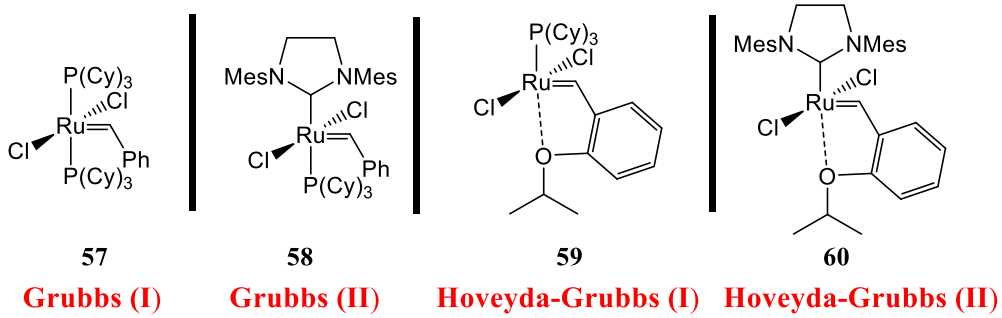


Figure 1.7. Regularly used catalysts **57-60** for the RCM or RCAM reactions.

1.5.4. Sophorolipid Lactone

Another unusual family of resin glycosides are sophorolipids (SL) isolated from the yeast *Candida bombicola*.[118] These glycolipids are used as biodegradable emulsifiers in multiple industries like cosmetic, food production, and pharmaceuticals. Interestingly, Candida *bombicola* has been shown to be able to grow on pure hydrocarbons.[119, 120] In 2017, SL were found to have anti-proliferative effects against human cervical cancer cells.[121] Native SL are made up of a mixture of 14 different natural products with **61** being the most abundant shown in Figure 1.8.

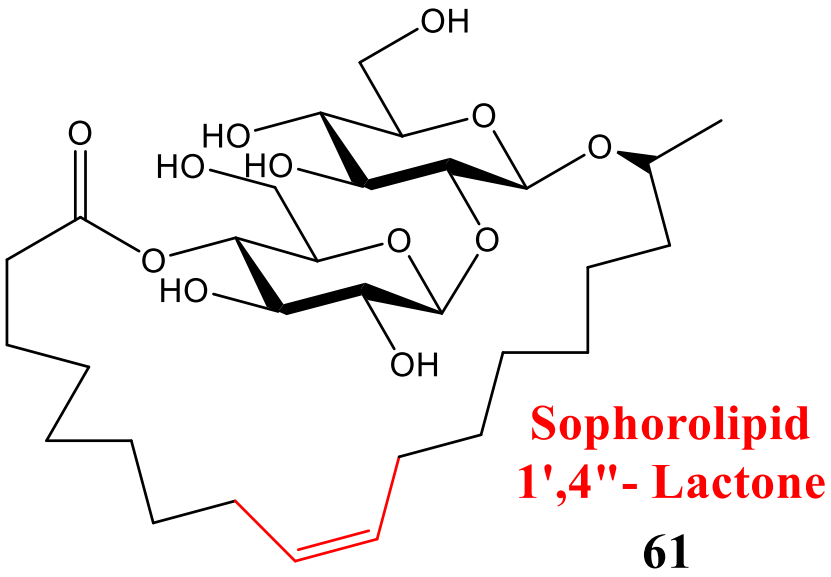
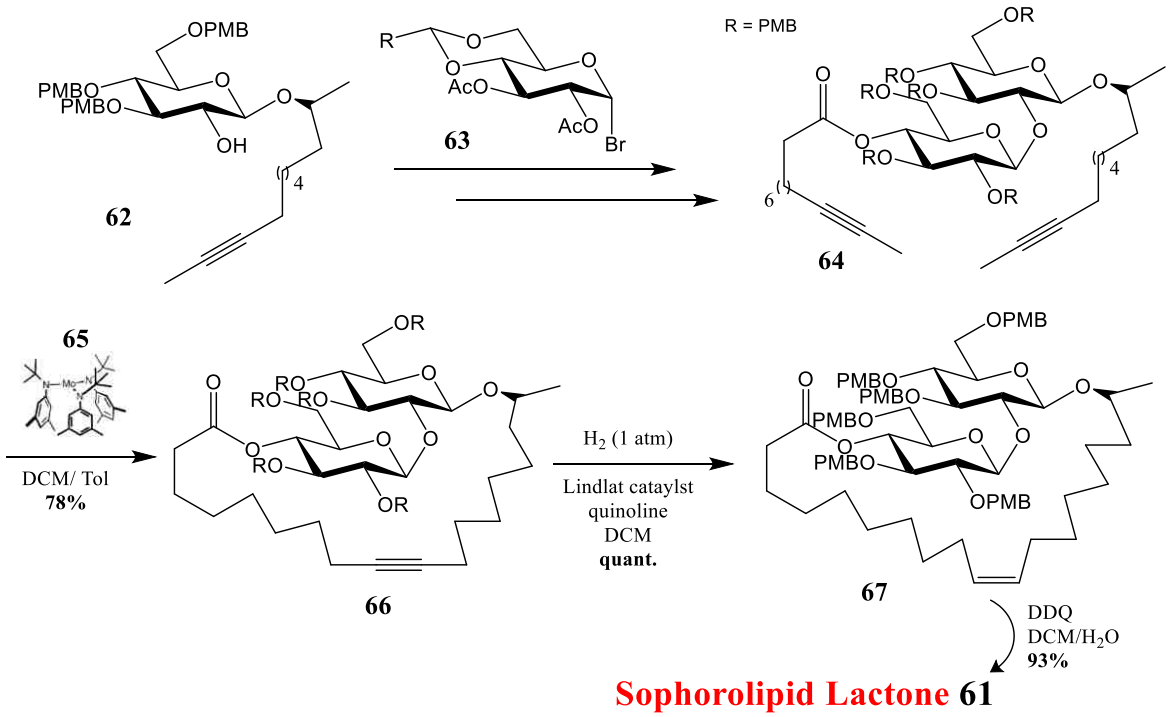


Figure 1.8. The most abundant sorphorolipid **61** containing an unsaturated aglycone.

The glycone moiety of **61** is structurally simple with two glucose (Glc*p*) units making up of a disaccharide backbone (Figure 1.8). However, the aglycone of **61** was not composed of jalalpinolic acid, but a *C-18* fatty acid with a cis double bond between *C-9* and *C-10* (red, Figure 1.8). As shown in the synthesis of tricolorins, RCM became the common method for synthesizing the large, saturated FA of the aglycone region. The RCM reactions tend to give a higher ratio of

the more stable (*E*)-double bond conformation. The resulting double bond can then be reduced though hydrogenation conditions to give the saturated aglycone.[98, 99] This offered an interesting challenge for Fürstner *et al.* synthesis of SL **61** (Scheme 1.6).[122] To give the essential (Z)-alkene in SL **61**, Fürstner *et al.* employed an innovative method called ring closing alkyne metathesis (RCAM) developed in the late-1990s.[123, 124] This transformation gave an internal alkyne instead of the mixture of double bonds observed for the RCM reaction. The triple bond could be selectivity reduced though a Birch reduction (trans formation) or Lindlar hydrogenation (cis formation).[124] The RCAM reaction worked effectively well for Fürstner *et al.'s* first total synthesis of SL **61** (Scheme 1.6).[118] Fürstner *et al.* observed the RCAM proceeding smoothly from **64** to **66** with the Mo catalyst **65**. Neither the glyosidic linkages or the acid-labile *p*-methyloxyphenyl (PMB) groups were affected by this transformation. Afterwards, Lindar's hydrogenation of **66** and the PMB-



Scheme 1.6. Fürstner *et al.* synthesis of SL **61** using RCAM method.

ether protecting groups were deprotected in **67** with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) and the synthesis of SL **61** was completed.[122]

1.6. Ipomoeassins

The ipomoeassin family is a small class of six resin glycosides isolated in the mid-2000s by Kingston *et al.* from the Suriname rainforest.[125] Ipomoeassins are part of the larger family of resin glycosides isolated from the Ipomoea *sp*., more commonly known as the morning glory flower. The *ipomoea* family is made up of about 650 plant species worldwide with almost half of the species located in Central and South America. Like other resin glycosides, many ancient Central and South American cultures used a crude mixture from the morning glories for their purgative and laxative properties. Recently, the biological applications of the ipomoeassins have grown ranging from antifungal, antibacterial, and cytotoxicity.[84, 86] However, the mode of action (MOA) of these bioactive molecules is still not well understood. This small family of glycoresins contain some interesting structural features, while exhibiting potent biological activity.

The ipomoeassins consist of a (1→2)-*β*-disaccharide core structure with different acylation patterns connected by the hydroxylated fatty acid derivatives **68**-**73** (Figure 1.9). The disaccharide or glycone moiety is made up of a Glc*p* and Fuc*p*-derivative. The peripheral acylation of cinnamate (cinn, C-4-Glc*p*) and tiglate (tig, C-3-Glc*p*) are rarely found in other resin glycosides.[94, 126, 127] As seen in tricolorins or batatin VI (Figure 1.2), the saturated or hydrated forms (2-methylbutyate or 3-hydroxy-2-methylburyic acid) are more often found.[128, 129] The C-4-Fuc*p* moiety differs for some of the ipomoeassins with some natural products being acylated (A **68**, C **70**, D **71**), while other are hydroxylated (B **69**, E **72**). The fatty acid or aglycone is made-up of a C_{14} or C_{16} chain connected to the glycone moiety though glyosidic bonds (C-6-Glc*p* and C-1-

Fuc*p*). Like many resin glycosides, the (11*S*)-configuration is found to connect the aglycone back to the glycone moiety.[93] Unlike most resin glycosides, ipomoeassins contain an oxygenation at *C-4* of the aglycone.[125, 130] Also, the aglycone of some ipomoeassins contain an unusual stereogenic center at *C-5*. Ipomoeassins C **70**, D **71**, and E **72** contain either an acylated or free hydroxyl group. This made some of the first syntheses of ipomoeassin E **72** more difficult than the syntheses of A **68** and B **69**. The absolute configuration of *C-5* and *C-11* stereogenic centers of the aglycone were confirmed by the *Mosher's* ester test.[125]

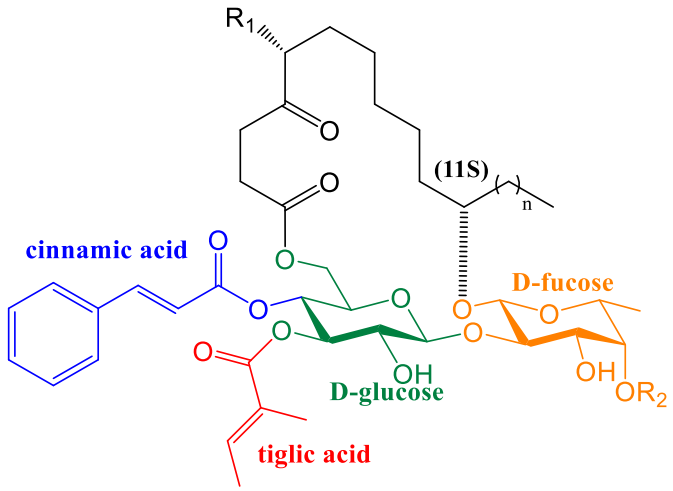


Figure 1.9. The Family of ipomoeassins **68**-**73**.

Table 1.4. Ipomoeassin A-F

		n	R1	R2
68	**Ipomo- A**	2	H	Ac
69	**Ipomo- B**	2	H	H
70	**Ipomo- C**	2	OH	Ac
71	**Ipomo- D**	2	OAc	Ac
72	**Ipomo- E**	2	OAc	H
73	**Ipomo- F**	4	H	Ac

Ipomoeassins offer an interesting case study for research of the potential importance of resin glycosides for modern medicine (Table 1.1). In 2005, Kingston *et al.* found ipomoeassins A-E **68**-**72** showed potent cytotoxicity against the A2780 ovarian cancer cell line, ranging from 35 to 1900 nM.[125] The potential pharmacophore "hot spots" seemed to be the acylation patterns of the C-4-Fuc*p* and *C-5* of the aglycone. Ipomoeassin D **71** only differed from ipomoeassin C **70** by the Ac group at *C-5* of the aglycone moiety, while being two-fold more active (35 nM to 2.9 µM). Ipomoeassin E **72** varies from ipomoeassin D **71** at the C-4-Fuc*p* moiety caused a 100-fold

decrease in the cytotoxicity (35 nM to 3.3 µM). This showed small changes in the ipomoeassins structure that could greatly affect the biological properties.

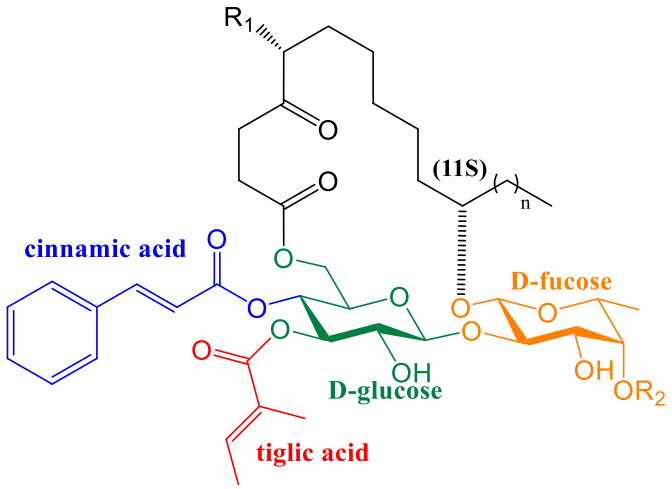


Table 1.5. Cell Growth Inhibitory Data for Ipomoeassin A, B, C, D, E, and F. IC_{50}

		n	R1	R2	A2780	HT-29	MDA-MBA-435	H522-T1	U937
68	**Ipomo- A**	**2**	**H**	**Ac**	**0.5 µM**	**46.1 nM**	**42.6 nM**	**108.9 nM**	**20.2 nM**
69	**Ipomo- B**	**2**	**H**	**H**	**0.4 µM**	**396 nM**	**2700 nM**	**1070nM**	**134 nM**
70	**Ipomo- C**	**2**	**OH**	**Ac**	**2.9 µM**	**–**	**–**	**–**	**–**
71	**Ipomo- D**	**2**	**OAc**	**Ac**	**35 nM**	**11.8 nM**	**19.9 nM**	**23.2 nM**	**7.9 nM**
72	**Ipomo- E**	**2**	**OAc**	**H**	**3.3 µM**	**393 nM**	**1633 nM**	**967 nM**	**163 nM**
73	**Ipomo- F**	**4**	**H**	**Ac**	**36 nM**	**4.2 nM**	**9.4 nM**	**12.9 nM**	**2.6 nM**

In 2007 a new glycoresin, ipomoeassin F **73**, was isolated by Kingston *et al.* [131] Ipomoeassin F **73** was shown to be structurally similar to ipomoeassin A **68** with the only difference being the fatty acid tether (C_{16} vs. C_{14}). This small change had a large effect on the cytotoxicity of the natural products. The published data showed that the addition of two methylene units could affect the lipophilicity of the aglycone of ipomoeassin F **73**.[132] The cell growth inhibitory assays showed ipomoeassin F **73** to be significantly more potent than its congeners against multiple cancer cell lines. Interestingly, ipomoeassin D **71** is only 2-3-fold less potent than

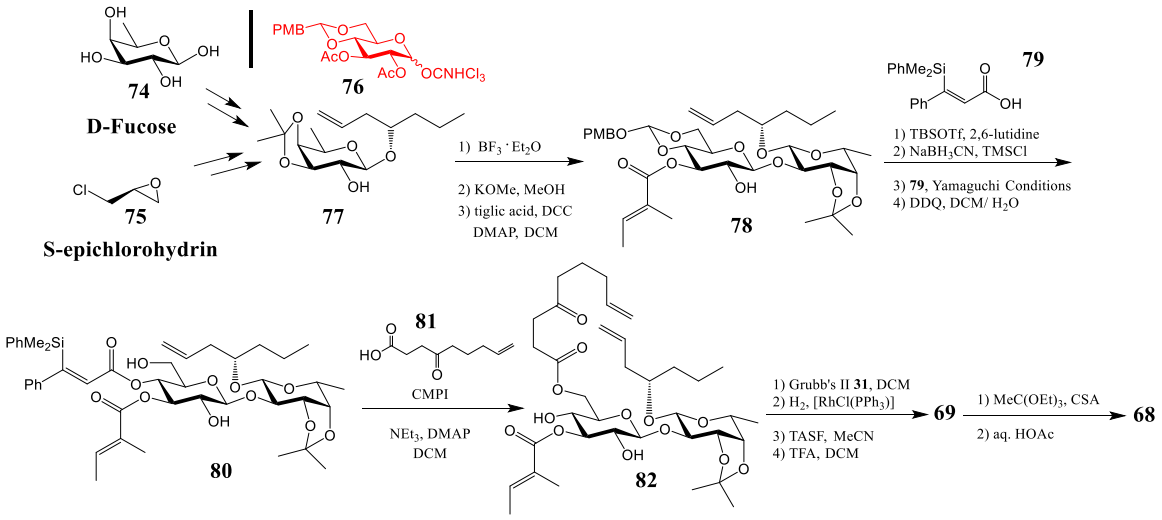
ipomoeassin F **73**. The only structural difference in the two compounds is the *C-5* Ac group of the aglycone moiety. This makes the synthesis of the aglycone for ipomoeassin D **71** considerably more difficult than ipomoeassin F **73**. The superior potency against multiple cell lines and easier aglycone synthesis are the reasons ipomoeassin F **73** was studied.

1.6.1. Previous Syntheses of Ipomoeassins

Multiple syntheses of ipomoeassins have been performed over the past decade by Fürstner [132, 133], Postema,[134] and Shi.[135-138] Fürstner *et al.* reported the first synthesis of ipomoeassins B **69** and E **72** in early 2007,[133] while Postema *et al.* published the first synthesis of the ipomoeassin F **73** in 2009[134] and shortly after Fürstner *et al.* synthesized ipomoeassin A, C, D, and F.[132] Recently, Shi *et al.* have published multiple papers on the synthesis of ipomoeassin F **73** and numerous analogs based on the natural product.[135-138] All the published syntheses utilized RCM (Figure 1.7) to form the macrolactone ring because of the high flexibility and the favorable reactivity profile.[118] The unsaturated ester groups (cinn C-4-Glc*p* and tig C-3-Glc*p*) have caused some difficulty because common reduction methods are not selective enough to reduce just the cis/trans double bond intermediate from the RCM reaction.

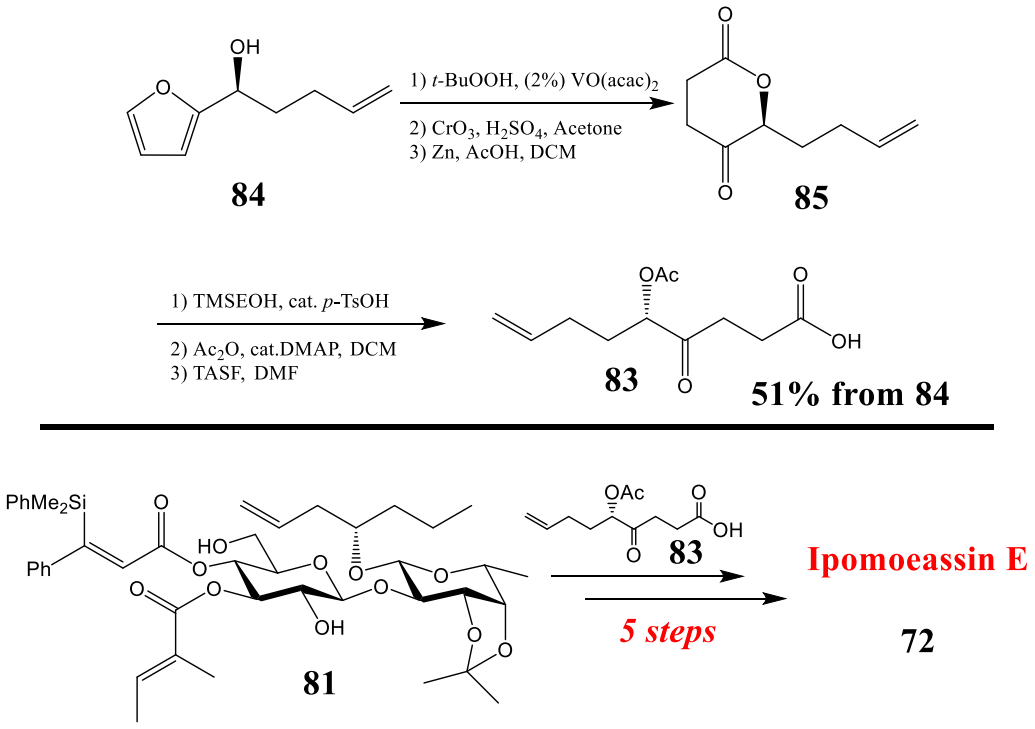
The key steps in Fürstner *et al.* synthesis of ipomoeassins A **68** and B **69** are shown in Scheme 1.7.[133] (*S*)-epichlorohydrin **75** has become a common starting material for resin glycosides because it establishes the (*S*)-configuration at *C-11* of the aglycone. The Fuc*p* acceptor **77** was synthesized in a few transformations from (*S*)-epichlorohydrin **75** and D-Fuc*p* **74**. The glucosyl (Glc*p*) donor **76** was prepared from a previously published hemiketal. After the glycosylation of **76** and **77** to form the (1→2)-*β*-disaccharide and an acetyl de-protection, a regioselective esterification installed the tig group **78**. The reductive opening of substituted benzylidene gave

the unexpected *C-6*-O-PMB ether **80**, causing Fürstner *et al.* to change the envisioned RCM/hydrogenation method. The (1→2)-*β*-disaccharide **78** was exposed to Yamaguchi conditions to couple with cinn acid substitute **79**. Fürstner *et al.* believed the C-silylated derived **79** could survive the hydrogenation with Wilkinson's catalyst because of its tri-substituted double bond. Then, the dimethyl phenylsilyl group **79** could be removed later in the synthesis. The 4-oxo-8-nonenic acid **81** was coupled with the (1→2)-*β*-disaccharide using Yamaguchi conditions to afford the diene **82,** followed by an RCM reaction to give the un-saturated macrocycle.[139] The reduction of the double bond with Wilkinson's catalyst proceeded surprisingly well (81%). Finally, the protecting groups were removed with diluted trifluoracetic acid (TFA) and trissulfonium difluorotrimethylsilicate (TASF) to give ipomoeassin B **69**. Interestingly, they reported other acidic conditions or fluoride sources were not feasible for the removal of TBS and isopropylidene groups. Finally, Ipomoeassin A **68** was afforded by an HOAc-induced orthoester rearrangement to give the observed axial acetate.[133]



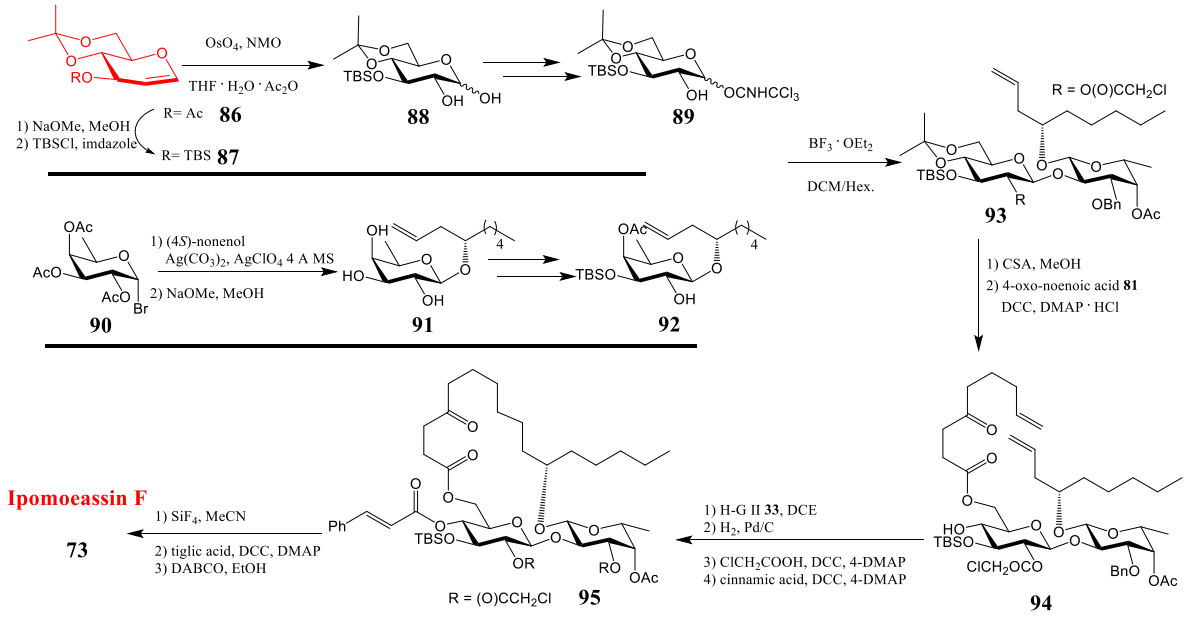
Scheme 1.7. Fürstner *et al.* synthesis of ipomoeassin A **68** and B **69** with the RCM reaction.

Most of Fürstner *et al.* synthesis of ipomoeassins A **68** and E **72** could be extended to ipomoeassin E **72**. The main difference comes from the *C-5* acetyl group on the aglycone moiety. The synthesis of 5-acetyl-4-oxo-8-nonenoic acid **83** by Fürstner *et al.* is shown in Scheme 1.8.[133] Sharpless-type kinetic resolution was performed on the alcohol **84**.[140, 141] Then, a ring expansion with catalytic vandayl acetylacetonate ($VO(acac)_2$), oxidation of the hemiacetal, and conjugate reduction of the enone gave the lactone **85**.[142] The six-membered lactone **85** was opened by trimethylsilyl ethanol (TMSEOH) and acid **83** formed by a fluoride-aided cleavage of the ester.[143, 144] Yamaguchi conditions were used to couple the acid **83** with the (1→2)-*β*-disaccharide **80** (Scheme 1.8). The remaining four steps to ipomoeassin E **72** followed the route of ipomoeassin A **68** (Scheme 1.7).[133]



Scheme 1.8. The scalable route to the 5-acetyl-4-oxonon-8-enoic acid **81** developed by Fürstner for the synthesis of ipomoeassin E **72**.

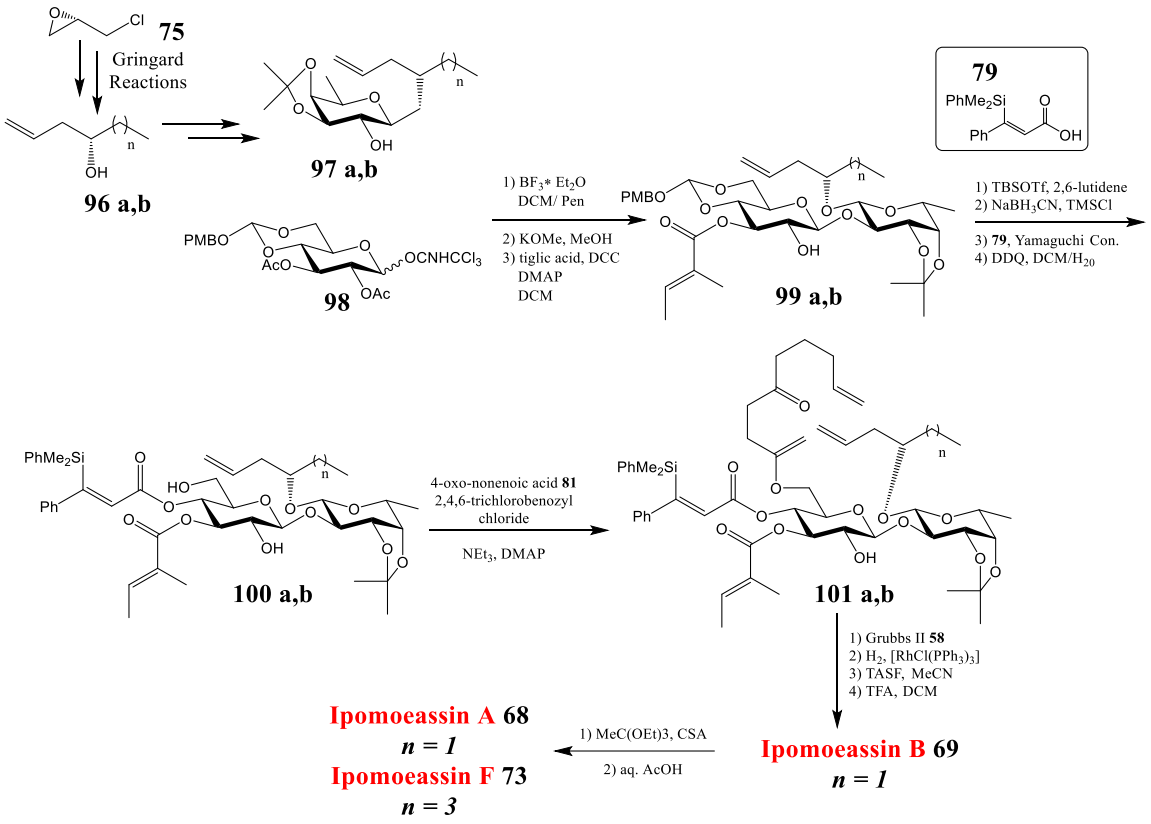
The first synthesis of ipomoeassin F **73** by Postema *et al.* is shown in Scheme 1.9.[134] Three differences from the first synthesis by Fürstner *et al.* were the Glc*p* starting material **86** (glucal), the late stage introduction of the peripheral *α,β*-unsaturated esters (cinn C-4-Glc*p* and tig C-3-Glc*p*), and the RCM catalyst (H-G II, **60**). Postema *et al.* chose to start with the commercially available glucal **86** for the preparation of the Glc*p* donor **89**. After a TBS protection of **87**, a dihydroxylation with osmium tetroxide (OsO_4) was performed to give the mixture of anomers **88**. Studies have shown OsO_4 to be extremely toxic, making this route not favorable for large synthesis of ipomoeassin F **73**.[145, 146] Unlike Fürstner synthesis, Postema *et al.* chose to start with optically pure 1,2-epoxyheptane and with the addition of a vinyl cuprate formed (4*S*)-nonenol **65**. This chemistry was adapted from the chiral epoxidation studies developed by Jacobsen *et al.* in the mid-2000s.[147] The Fuc*p* acceptor **92** was made through a series of transformations after the glycosylation of Fuc*p* bromide **90** and (4*S*)-nonenol. The final two steps in the synthesis of the Fuc*p* acceptor **92** involved the Ac introduction to C-4-Fuc*p* and the de-protection of TBS with a



Scheme 1.9. Postema *et. al.* first synthesis of ipomoeassin F **73** in 2009.

weak acid (HCl, MeOH) to give the HO-2-Fuc*p* (**91**→**92**). Like Fürstner *et al*. synthesis, a (1→2)-*β*-disaccharide **93** was formed between the Schmidt donor **89** and Fuc*p* acceptor **92** catalyzed by $BF_3 \cdot OEt_2$. They also observed a significant amount of the isopropylidene (C-4-Glc*p* and C-6-Glc*p*) removed to give the diol. After a CSA deprotection of the acetonide, the 4-oxo-8-noenoic acid **81**[148] was regio-selectively introduced (Steglich method) to the C-6 hydroxyl of the Glc*p* moiety. The RCM reaction was catalyzed by H-G II **60** catalyst and the mixture of cis-trans isomers hydrogenated (H_2, Pd/C). The simple hydrogenation conditions offered a big advantage over Fürstner *et al*. synthesis, since they needed to use Wilkinson's catalyst to not interfere with the unsaturated esters (cinn and tig). Then, the HO-3-Fuc*p* was protected with chloroacetylation followed by Steglich esterification with cinn acid to give the intermediate **95**. Expectedly, this coupling reaction was very difficult because of the bulky OTBS group blocked the C-4-Glc*p*. The esterification would only proceed if heated under solvent evaporation and replacement conditions. Then, the C-3-Fuc*p* TBS was removed with silicon tetrafluoride (SiF_4) and HO-3-Glu*p* coupling reaction of tig acid (DCC, DMAP). Finally, the ipomoeassin F **73** was obtained after an excess of DABCO removed the α-chloroacetates.

Later in 2009, Fürstner *et al.* synthesized ipomoeassin A- F **68-73** for the most complete study of ipomoeassins at the time (Scheme 1.10).[149] They chose to make the (*S*)-alcohols **49 a**, **b** through a series of Grignard reactions catalyzed by copper(I) cyanide (CuCN). After a glycosylation with a bromo Fuc*p*, like Postema *et al*. synthesis (Scheme 1.8 **90**→**91**), they obtained the C-3/C-4 protected Fuc*p* acceptor **97 a**, **b** in two steps. This is a benefit over Postema *et al* synthesis that took five steps to achieve the Fuc*p* acceptor **92** from the bromo-sugar **91** (Scheme



Scheme 1.10. Key transformation in Fürstner's synthesis of ipomoeassins.

1.9). The Schmidt donor **98** was prepared from a hemiketal published a previously reported route.[122] The glycosylation reaction of donor **98** and acceptor **97 a**, **b** was catalyzed by the $BF_3 \cdot Et_2O$, then the C-2-Glc*p* and C-3-Glc*p* deacetylated with potassium methoxide (KOMe). The increased nucleophilicity of the HO-3-Glc*p* was utilized to perform a regioselective esterification to install the tig moiety giving **99 a**, **b**. The benzylidene acetal was opened with $NaBH_3CN$ with chlorotrimethylsilane (TMSCl), followed by Yamaguchi esterification with the C-silyated cinn acid **79**, and oxidative cleavage of the C-6-Glc*p* PMB using 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ) to give the lone free alcohol. Fürstner *et al.* believed trisubstituted alkene **79** would not inhibit the RCM reaction and allow for simpler hydrogenation conditions

from the alkene metathesis product. Then, the sacrificial protecting group was simultaneously removed with the C-2-Fuc*p* TBS at the end of the synthesis. The *C*-silyated cinn acid **79** was derived from 3-phenyl-propargyl alcohol in four steps. A Yamaguchi esterification was performed with 4-oxo-8-noenoic acid **81** to give the RCM precursor **101 a**, **b**. The aglycone intermediates that contained the *C-5* stereocenter (aglycone) were prepared similarly to Scheme 1.8.[110, 140, 150] The RCM reaction proceeded smoothly, and the corresponding double bond was reduced with Wilkinson's catalyst. This choice of catalyst was interesting because they thought the C-silyated group would not interfere with a hydrogenation reaction. Finally, ipomoeassin B **69** was attained by removal of the silyl protecting group (C-3-Fuc*p* and C-4-*Glcp* cinn) with TASF and the isopropylidene acetal deprotected with dilute TFA. Ipomoeassin A **68** and F **73** were achieved by an AA-promoted orthoester rearrangement to give the axial acetate.

Along with the natural product, Fürstner *et al.* prepared a small library of analogs **102-104** to confirm the importance of the oxygenation and acylation patterns (Figure 1.10).[132] They prepared the 4-deoxy-ipomoeassin B **102** in four steps from the esterification of intermediate **99a** with the commercially available 8-nonenoic acid. The deacetylated **103** and the regioisomer **104**

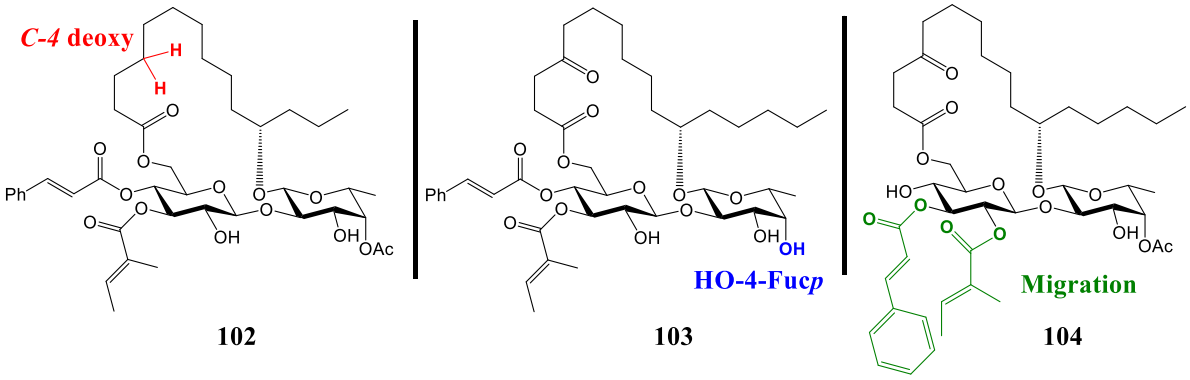
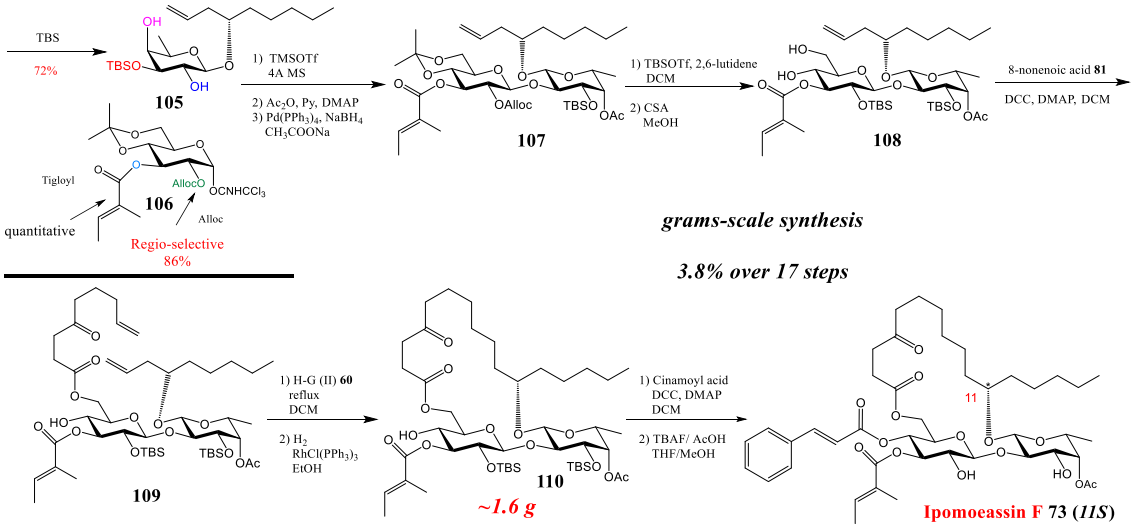


Figure 1.10. First synthetized analogs of the ipomoeassins by Fürstner *et al.* in 2009.

were accidently obtained during the final step, when high concentrations of TFA caused the Ac cleavage and migration. Compared to ipomoeassin F **73**, C-4-Fuc*p* hydroxylated analog **103** showed slight loss in anti-tumor activity (two-fold loss). The 4-deoxy-ipomoeassin B **102** was the most active, indicating the *C-4* ketone is not important to biological activity. Interestingly, they chose not to make 4-deoxy analog **102** based on ipomoeassin F **73**. Later, Shi *et al.* synthesized the 4-deoxy-ipomoeassin F analog that exhibited good cytotoxicity with a only a two-fold loss in the cytotoxicity.[136]

Recently, Shi *et al.* has reported the total synthesis of the ipomoeassin F **73** and numerous analogs based on the natural product.[135-138] In 2015, they published a new synthesis route of ipomoeassin F **73** and the unnatural 11 *R*-epimer (Scheme 1.11).[135] Unlike Postema *et al.* synthesis, Shi *et al.* wanted to design a synthetic route where the cinn group was introduced as late as possible. They believe a late-stage introduction of the cinn moiety could situate important SAR



Scheme 1.11. Shi *et al.* synthesis of ipomoeassin F **73** and its 11*R*-epimer.

studies at the C-4-Glc*p*, since unsaturated esters are rare for resin glycosides (Figure 1.5-1.8). The reactivity HO-3-Fuc*p* was exploited to regio-selectively install the silyl group for the Fuc*p* acceptor **105**. Shi *et al.* chose to use D-Glc*p*, not glucal (Postema *et al.* Scheme 1.9) for the preparation of the Glc*p* moiety. This was a benefit over Postema *et al.* synthesis, since Shi *et al.* did not need to use toxic OsO_4 to form the OH-2-Glc*p* and OH-3-Glc*p*.[134] Similar to the previous synthesis of ipomoeassins, the trichloroacetimidate donor **106** was designed because of the highly effective transformation. Alloc was chosen as a transient protecting group for C-2-Glc*p* to assist the formation of the *β*-(1→2)-disaccharide linkage though neighboring group participation. The de-protection conditions and wide functional group tolerance made Alloc an attractive protecting group.[151] After the glycosylation catalyzed by trimethylsilyl trifluoromethanesulfonate (TMSOT*f*), an acetylation was performed at C-4-Fuc*p*, and the C-2-Glc*p* Alloc removed to give the (1→2)-*β*-disaccharide **107**. The free OH-2-Fuc*p* was protected with TBS and the CSA removed the isopropylidene to afford the diol **108**. Unlike the previous syntheses of ipomoeassin, Shi *et al.* took advantage of the nucleophilicity of the primary OH-6-Glc*p* and regio-selectively added the 4-oxo-8-nonenoic acid **81** to afford the diene **109**. The macrocycle formation was catalyzed by H-G II **60** and the corresponding cis-trans double bond was reduced with H_2. Then, the cinn moiety could be introduced to the OH-4-Glc*p* **110** without the worry of any unwanted regioisomers. The final step in the synthesis involved the removal of the TBS groups to give ipomoeassin F **73**. Since 1.6 g of intermediate **110** was synthesized, this route could be utilized in the future to prepare grams scale quantities of ipomoeassin F.[136]

1.7. Statement of the Problem

By 2015, multiple syntheses have been developed for the resin glycoside, ipomoeassin F (Scheme 1.7-1.11).[133-135, 149] Through these syntheses numerous analogs were prepared to

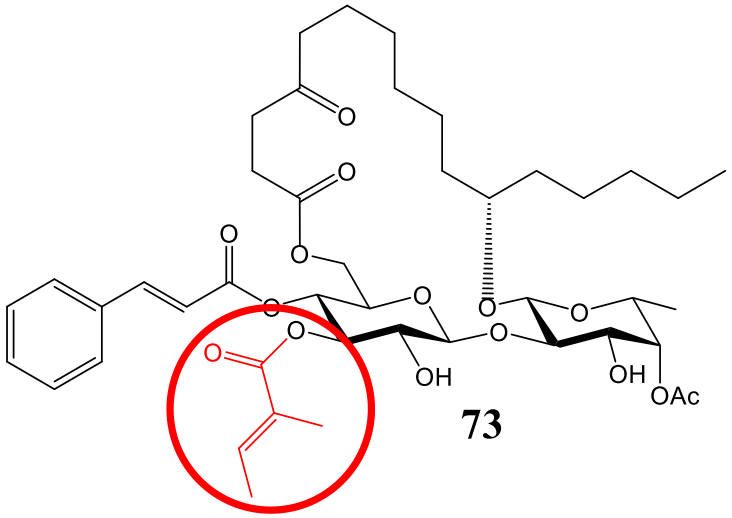
investigate the importance of different moieties of the natural product. In 2016, Shi *et al.* synthetized a library of analogs for the most systematic study of ipomoeassin F **73** to date. They found the *C-4* ketone and the macrolactone ring were not vital to overall activity of the natural product, while modification of the 3-*O*-Gl*p* (tig) or 4-*O*-Gl*p* (cinn) would remarkably spoil the cytotoxicity (150-1000 loss in activity). Even subtle modification to the *α,β*-unsaturated esters resulted in dramatic changes in the cytotoxicity of the analogs. These analogs caused Shi *et al.* to postulate that the cinn group could be a pharmacophoric "hot spot" through π-π stacking interaction with an aromatic AA. This finding caused Shi *et al.* to hypothesize that the *α,β*-unsaturated esters could form a covalent bond with the target through a 1,4-Michael addition.[135,136]

1.7.1. Ipomoeassins… Novel Target?

More significant than SAR studies, was the information found through the NCI 60-cell line screening of ipomoeassin A **68** (most abundant congener of the ipomoeassins family). This screening showed ipomoeassin A **68** to have comparable potency and selective growth inhibition to clinically anti-cancer agents against different cancer cells lines.[58] Interestingly, ipomoeassin A **68** exhibited a pattern of activity evidently different from known anticancer agents.[135, 136] This pattern caused researchers to ask whether ipomoeassins could be inhibiting a novel or "undruggable" target? Since known "druggable" targets make up only a small percentage of the potential targets in the human genome (Table 1.3).[38, 51, 152] This led researchers to ask if ipomoeassins, specifically ipomoeassin F, act as a scaffold to develop a new class of antineoplastic drugs?

One moiety of ipomoeassin F **73** that has not been well-studied is the 3-*O*-Glc*p* and the role of the tig moiety. This position is interesting to explore for a few different reasons. The first reason is the structural aspects of the *α,β*-unsaturated ester of tig acid. It is more common to find

the reduced form of the short-chain aliphatic acid, such as mba or propionic, or saturated fatty acids (Figure 1.5-1.8, *n*-decanoic or palmitic acids).[87, 127] The second reason is similar to the first, involving the *α*,*β*-unsaturated ester (tig moiety). Shi *et al*. comprehensive SAR study showed the two Michael acceptor systems are very interesting.[136] This combination is very uncommon and a distinguishing trait of the ipomoeassins family. Even if one of the *α*,*β*-unsaturated esters forms a covalent bond with the target, the identification of the target protein/proteins could be simplified. However, the roles of the *α*-methyl and *β*-methyl group in tig were not known.



73

Figure 1.11. Design Synthesis route to examine of C-3-Glc*p* tig moiety to overall activity of ipomoeassin F **73**.

1.7.2. Designing a Synthesis Route

To examine the importance of the tig moiety and determine its importance for the cytotoxicity of ipomoeassin F **73**, a scalable and flexible synthesis route needed to be developed. To design a successful synthesis route, late-stage modification to the 3-*O*-Glc*p* needed to be performed. This would make the synthesis more efficient and lessen the need for valuable intermediates. The total synthesis by Fürstner *et al.* could not be adapted because they added the

tig moiety in the early stages, making medicinal chemistry studies of the C-3-Glc*p* inefficient (Scheme 1.8 and 1.10).[133] The synthesis of Postema *et al.* was better because they introduced the tig in the penultimate step (Scheme 1.9). Unfortunately, their overall yield for the synthesis of ipomoeassin F **73** was extremely low (<0.4 %).[134] Since RCM had been found to be extremely efficient for preparing macrocycles and their derivatives, this strategy was adopted. The cinn moiety was also introduced in the later stage because of potential steric problems and the moiety's importance to the cytotoxicity of ipomoeassin F **73**. The synthesis route is highlighted by multiple regioselective esterification reactions, orthogonal protections and deprotections, and the formation of the (1→2)-*β*-disaccharide through a glycosylation reaction.

The goal of this project was to design a successful synthesis route to examine the role of the tig moiety (C-3-Glc*p*) of ipomoeassin F **73**. In the following sections the chemical transformations used to achieve this goal will be presented in detail. These studies will provide significant insight for future studies of ipomoeassins for anti-cancer drug development.

1.8. References

1. Newman, D. J.; Cragg, G. M., Natural Products as Sources of New Drugs from 1981 to 2014. *Journal of Natural Products* **2016,** *79* (3), 629-661.

2. Dayan, F. E.; Cantrell, C. L.; Duke, S. O., Natural products in crop protection. *Bioorganic & Medicinal Chemistry* **2009,** *17* (12), 4022-4034.

3. Abate, A.; Brenna, E.; Fuganti, C.; Gatti, F. G.; Giovenzana, T.; Malpezzi, L.; Serra, S., Chirality and fragrance chemistry: Stereoisomers of the commercial chiral odorants Muguesia and Pamplefleur. *Journal of Organic Chemistry* **2005,** *70* (4), 1281-1290.

4. Brenna, E.; Fuganti, C.; Serra, S.; Kraft, P., Optically active ionones and derivatives: Preparation and olfactory properties. *European Journal of Organic Chemistry* **2002,** (6), 967-978.

5. Brenna, E.; Fuganti, C., Recent Advances in the Synthesis of Fragrances. *Current Organic Chemistry* **2011,** *15* (7), 987-1005.

6. Cioc, R. C.; Ruijter, E.; Orru, R. V. A., Multicomponent reactions: advanced tools for sustainable organic synthesis. *Green Chemistry* **2014,** *16* (6), 2958-2975.

7. Quitmann, H.; Fan, R.; Czermak, P., Acidic Organic Compounds in Beverage, Food, and Feed Production. *Biotechnology of Food and Feed Additives* **2014,** *143*, 91-141.

8. Kingston, D. G. I., Modern Natural Products Drug Discovery and Its Relevance to Biodiversity Conservation. *Journal of Natural Products* **2011,** *74* (3), 496-511.

9. Patridge, E.; Gareiss, P.; Kinch, M. S.; Hoyer, D., An analysis of FDA-approved drugs: natural products and their derivatives. *Drug Discovery Today* **2016,** *21* (2), 204-207.

10. Giddings, L.-A.; Newman, D. J., Bioactive Compounds from Extremophiles. In *Bioactive Compounds from Extremophiles: Genomic Studies, Biosynthetic Gene Clusters, and New Dereplication Methods*, Springer International Publishing: Cham, 2015; pp 1-47.

11. Newman, D. J.; Cragg, G. M., Endophytic and epiphytic microbes as "sources" of bioactive agents. *Frontiers in Chemistry* **2015,** *3*.

12. Siegel, R. L.; Miller, K. D.; Jemal, A., Cancer Statistics, 2017. *Ca-a Cancer Journal for Clinicians* **2017,** *67* (1), 7-30.

13. Rask-Andersen, M.; Almen, M. S.; Schioth, H. B., Trends in the exploitation of novel drug targets. *Nature Reviews Drug Discovery* **2011,** *10* (8), 579-590.

14. van Montfort, R. L. M.; Workman, P., Structure-based design of molecular cancer therapeutics. *Trends in Biotechnology* **2009,** *27* (5), 315-328.

15. Knapp, M.; Bellamacina, C.; Murray, J. M.; Bussiere, D. E., Targeting cancer: The challenges and successes of structure-based drug design against the human purinome. *Current Topics in Medicinal Chemistry* **2006,** *6* (11), 1129-1159.

16. Caliandro, R.; Belviso, D. B.; Aresta, B. M.; de Candia, M.; Altomare, C. D., Protein crystallography and fragment-based drug design. *Future Medicinal Chemistry* **2013,** *5* (10), 1121-1140.

17. Zhang, Q. C.; Petrey, D.; Deng, L.; Qiang, L.; Shi, Y.; Thu, C. A.; Bisikirska, B.; Lefebvre, C.; Accili, D.; Hunter, T.; Maniatis, T.; Califano, A.; Honig, B., Structure-based prediction of protein-protein interactions on a genome-wide scale. *Nature* **2012,** *490* (7421), 556-+.

18. Szychowski, J.; Truchon, J. F.; Bennani, Y. L., Natural Products in Medicine: Transformational Outcome of Synthetic Chemistry. *Journal of Medicinal Chemistry* **2014,** *57* (22), 9292-9308.

19. Lee, W. L.; Shiau, J. Y.; Shyur, L. F., Taxol, Camptothecin and Beyond for Cancer Therapy. *Recent Trends in Medicinal Plants Research* **2012,** *62*, 133-178.

20. Drake, R. R., Glycosylation and Cancer: Moving Glycomics to the Forefront. *Glycosylation and Cancer* **2015,** *126*, 1-10.

21. Alipour, M.; Bigdeli, M.; Rasoulian, B.; Khaksarian, M., Reviews of Natural Products and Herbal Medicines in Treatment of Cancer. *International Journal of Advanced Biotechnology and Research* **2017,** *8* (3), 2127-2139.

22. Bijauliya, R. K.; Alok, S.; Singh, M.; Mishra, S. B., A COMPREHENSIVE REVIEW ON CANCER AND ANTICANCER HERBAL DRUGS. *International Journal of Pharmaceutical Sciences and Research* **2017,** *8* (7), 2740-2761.

23. Graham, J. G.; Quinn, M. L.; Fabricant, D. S.; Farnsworth, N. R., Plants used against cancer - an extension of the work of Jonathan Hartwell. *Journal of Ethnopharmacology* **2000,** *73* (3), 347-377.

24. Cragg, G. M.; Grothaus, P. G.; Newman, D. J., Impact of Natural Products on Developing New Anti-Cancer Agents. *Chemical Reviews* **2009,** *109* (7), 3012-3043.

25. Cragg, G. M.; Newman, D. J., Plants as a source of anti-cancer agents. *Journal of Ethnopharmacology* **2005,** *100* (1-2), 72-79.

26. Kingston, D. G. I.; Newman, D. J., Natural products as drug leads: An old process or the new hope for drug discovery? *Idrugs* **2005,** *8* (12), 990-992.

27. Kingston, D. G. I.; Newman, D. J., The search for novel drug leads for predominately antitumor therapies by utilizing mother nature's pharmacophoric libraries. *Current Opinion in Drug Discovery & Development* **2005,** *8* (2), 207-227.

28. Shieh, Y.; Eklund, M.; Sawaya, G. F.; Black, W. C.; Kramer, B. S.; Esserman, L. J., Population-based screening for cancer: hope and hype. *Nature Reviews Clinical Oncology* **2016,** *13* (9), 550-565.

29. Smieliauskas, F.; Chien, C. R.; Shen, C.; Geynisman, D. M.; Shih, Y. C. T., Cost-Effectiveness Analyses of Targeted Oral Anti-Cancer Drugs: A Systematic Review. *Pharmacoeconomics* **2014,** *32* (7), 651-680.

30. Bhattarai, D.; Singh, S.; Jang, Y.; Han, S. H.; Lee, K.; Choi, Y., An Insight into Drug Repositioning for the Development of Novel Anti-Cancer Drugs. *Current Topics in Medicinal Chemistry* **2016,** *16* (19), 2156-2168.

31. Kim, J. W.; Cochran, J. R., Targeting ligand-receptor interactions for development of cancer therapeutics. *Current Opinion in Chemical Biology* **2017,** *38*, 62-69.

32. Lazo, J. S.; Sharlow, E. R., Drugging Undruggable Molecular Cancer Targets. *Annual Review of Pharmacology and Toxicology, Vol 56* **2016,** *56*, 23-40.

33. Patridge, E. V.; Gareiss, P. C.; Kinch, M. S.; Hoyer, D. W., An analysis of original research contributions toward FDA-approved drugs. *Drug Discovery Today* **2015,** *20* (10), 1182-1187.

34. Kinch, M. S., Post-approval fate of pharmaceutical companies. *Drug Discovery Today* **2015,** *20* (2), 170-174.

35. Kinch, M. S.; Flath, R., New drug discovery: extraordinary opportunities in an uncertain time. *Drug Discovery Today* **2015,** *20* (11), 1288-1292.

36. Kinch, M. S.; Hoyer, D., A history of drug development in four acts. *Drug Discovery Today* **2015,** *20* (10), 1163-1168.

37. Doak, B. C.; Zheng, J.; Dobritzsch, D.; Kihlberg, J., How Beyond Rule of 5 Drugs and Clinical Candidates Bind to Their Targets. *Journal of Medicinal Chemistry* **2016,** *59* (6), 2312-2327.

38. Gashaw, I.; Ellinghaus, P.; Sommer, A.; Asadullah, K., What makes a good drug target? *Drug Discovery Today* **2011,** *16* (23-24), 1037-1043.

39. Elebring, T.; Gill, A.; Plowright, A. T., What is the most important approach in current drug discovery: doing the right things or doing things right? *Drug Discovery Today* **2012,** *17* (21-22), 1166-1169.

40. Zhu, F.; Han, B. C.; Kumar, P.; Liu, X. H.; Ma, X. H.; Wei, X. N.; Huang, L.; Guo, Y. F.; Han, L. Y.; Zheng, C. J.; Chen, Y. Z., Update of TTD: Therapeutic Target Database. *Nucleic Acids Research* **2010,** *38*, D787-D791.

41. Surade, S.; Blundell, T. L., Structural Biology and Drug Discovery of Difficult Targets: The Limits of Ligandability. *Chemistry & Biology* **2012,** *19* (1), 42-50.

42. Perola, E.; Herman, L.; Weiss, J., Development of a Rule-Based Method for the Assessment of Protein Druggability. *Journal of Chemical Information and Modeling* **2012,** *52* (4), 1027-1038.

43. Venkatesan, K.; Rual, J. F.; Vazquez, A.; Stelzl, U.; Lemmens, I.; Hirozane-Kishikawa, T.; Hao, T.; Zenkner, M.; Xin, X. F.; Goh, K. I.; Yildirim, M. A.; Simonis, N.; Heinzmann, K.; Gebreab, F.; Sahalie, J. M.; Cevik, S.; Simon, C.; de Smet, A. S.; Dann, E.; Smolyar, A.; Vinayagam, A.; Yu, H. Y.; Szeto, D.; Borick, H.; Dricot, A.; Klitgord, N.; Murray, R. R.; Lin, C.; Lalowski, M.; Timm, J.; Rau, K.; Boone, C.; Braun, P.; Cusick, M. E.; Roth, F. P.; Hill, D. E.; Tavernier, J.; Wanker, E. E.; Barabasi, A. L.; Vidal, M., An empirical framework for binary interactome mapping. *Nature Methods* **2009,** *6* (1), 83-90.

44. Amelio, I.; Lisitsa, A.; Knight, R. A.; Melino, G.; Antonov, A. V., Polypharmacology of Approved Anticancer Drugs. *Current Drug Targets* **2017,** *18* (5), 534-543.

45. Antolin, A. A.; Workman, P.; Mestres, J.; Al-Lazikani, B., Polypharmacology in Precision Oncology: Current Applications and Future Prospects. *Current Pharmaceutical Design* **2016,** *22* (46), 6935-6945.

46. Tan, Z.; Chaudhai, R.; Zhang, S. X., Polypharmacology in Drug Development: A Minireview of Current Technologies. *Chemmedchem* **2016,** *11* (12), 1211-1218.

47. Zhang, W. J.; Bai, Y. F.; Wang, Y. H.; Xiao, W., Polypharmacology in Drug Discovery: A Review from Systems Pharmacology Perspective. *Current Pharmaceutical Design* **2016,** *22* (21), 3171-3181.

48. Schenone, M.; Dancik, V.; Wagner, B. K.; Clemons, P. A., Target identification and mechanism of action in chemical biology and drug discovery. *Nature Chemical Biology* **2013,** *9* (4), 232-240.

49. Clemons, P. A.; Bodycombe, N. E.; Carrinski, H. A.; Wilson, J. A.; Shamji, A. F.; Wagner, B. K.; Koehler, A. N.; Schreiber, S. L., Small molecules of different origins have distinct distributions of structural complexity that correlate with protein-binding profiles. *Proceedings of the National Academy of Sciences of the United States of America* **2010,** *107* (44), 18787-18792.

50. Carlson, E. E., Natural Products as Chemical Probes. *Acs Chemical Biology* **2010,** *5* (7), 639-653.

51. Bunnage, M. E.; Chekler, E. L. P.; Jones, L. H., Target validation using chemical probes. *Nature Chemical Biology* **2013,** *9* (4), 195-199.

52. Harvey, A. L., Natural products in drug discovery. *Drug Discovery Today* **2008,** *13* (19-20), 894-901.

53. Pauwels, K.; Huys, I.; Casteels, M.; De Nys, K.; Simoens, S., Market access of cancer drugs in European countries: improving resource allocation. *Targeted Oncology* **2014,** *9* (2), 95-110.

54. Cragg, G. M.; Grothaus, P. G.; Newman, D. J., New Horizons for Old Drugs and Drug Leads. *Journal of Natural Products* **2014,** *77* (3), 703-723.

55. Govindarajan, M., Amphiphilic glycoconjugates as potential anti-cancer chemotherapeutics. *European Journal of Medicinal Chemistry* **2018,** *143*, 1208-1253.

56. Lam, K. S., New aspects of natural products in drug discovery. *Trends in Microbiology* **2007,** *15* (6), 279-289.

57. Giupponi, G.; Harvey, M. J.; De Fabritiis, G., The impact of accelerator processors for high-throughput molecular modeling and simulation. *Drug Discovery Today* **2008,** *13* (23-24), 1052-1058.

58. Kingston, D. G. I., A natural love of natural products. *Journal of Organic Chemistry* **2008,** *73* (11), 3975-3984.

59. Feher, M.; Schmidt, J. M., Property distributions: Differences between drugs, natural products, and molecules from combinatorial chemistry. *Journal of Chemical Information and Computer Sciences* **2003,** *43* (1), 218-227.

60. Hughes, J. D.; Blagg, J.; Price, D. A.; Bailey, S.; DeCrescenzo, G. A.; Devraj, R. V.; Ellsworth, E.; Fobian, Y. M.; Gibbs, M. E.; Gilles, R. W.; Greene, N.; Huang, E.; Krieger-Burke, T.; Loesel, J.; Wager, T.; Whiteley, L.; Zhang, Y., Physiochemical drug properties associated with in vivo toxicological outcomes. *Bioorganic & Medicinal Chemistry Letters* **2008,** *18* (17), 4872-4875.

61. Prunet, J., Progress in Metathesis Through Natural Product Synthesis. *European Journal of Organic Chemistry* **2011,** (20-21), 3634-3647.

62. Yang, Y.; Zhang, X. H.; Yu, B., O-Glycosylation methods in the total synthesis of complex natural glycosides. *Natural Product Reports* **2015,** *32* (9), 1331-1355.

63. Oskarsson, T.; Nagorny, P.; Krauss, I. J.; Perez, L.; Mandal, M.; Yang, G. L.; Ouerfelli, O.; Xiao, D. H.; Moore, M. A. S.; Massague, J.; Danishefsky, S. J., Diverted Total Synthesis Leads to the Generation of Promising Cell-Migration Inhibitors for Treatment of Tumor Metastasis: In vivo and Mechanistic Studies on the Migrastatin Core Ether Analog. *Journal of the American Chemical Society* **2010,** *132* (9), 3224-3228.

64. Wilson, R. M.; Danishefsky, S. J., Small molecule natural products in the discovery of therapeutic agents: The synthesis connection. *Journal of Organic Chemistry* **2006,** *71* (22), 8329-8351.

65. Hamel, E., NATURAL-PRODUCTS WHICH INTERACT WITH TUBULIN IN THE VINCA DOMAIN - MAYTANSINE, RHIZOXIN, PHOMOPSIN-A, DOLASTATIN-10 AND DOLASTATIN-15 AND HALICHONDRIN-B. *Pharmacology & Therapeutics* **1992,** *55* (1), 31-51.

66. Bai, R.; Paull, K. D.; Herald, C. L.; Malspeis, L.; Pettit, G. R.; Hamel, E., HALICHONDRIN-B AND HOMOHALICHONDRIN-B, MARINE NATURAL-PRODUCTS BINDING IN THE VINCA DOMAIN OF TUBULIN - DISCOVERY OF TUBULIN-BASED MECHANISM OF ACTION BY ANALYSIS OF DIFFERENTIAL CYTOTOXICITY DATA. *Journal of Biological Chemistry* **1991,** *266* (24), 15882-15889.

67. Aicher, T. D.; Buszek, K. R.; Fang, F. G.; Forsyth, C. J.; Jung, S. H.; Kishi, Y.; Matelich, M. C.; Scola, P. M.; Spero, D. M.; Yoon, S. K., TOTAL SYNTHESIS OF HALICHONDRIN-B

AND NORHALICHONDRIN-B. *Journal of the American Chemical Society* **1992,** *114* (8), 3162-3164.

68. Stamos, D. P.; Kishi, Y., Synthetic studies on halichondrins: A practical synthesis of the C.1-C.13 segment. *Tetrahedron Letters* **1996,** *37* (48), 8643-8646.

69. Namba, K.; Jun, H. S.; Kishi, Y., A simple but remarkably effective device for forming the C8-C14 polycyclic ring system of halichondrin B. *Journal of the American Chemical Society* **2004,** *126* (25), 7770-7771.

70. Shablak, A., Eribulin for Advanced Breast Cancer: A Drug Evaluation. *Journal of Breast Cancer* **2013,** *16* (1), 12-15.

71. Nastrucci, C.; Cesario, A.; Russo, P., Anticancer Drug Discovery from the Marine Environment. *Recent Patents on Anti-Cancer Drug Discovery* **2012,** *7* (2), 218-232.

72. Morris, P. G., Advances in therapy: eribulin improves survival for metastatic breast cancer. *Anti-Cancer Drugs* **2010,** *21* (10), 885-889.

73. Christian, M. C.; Pluda, J. M.; Ho, P. T. C.; Arbuck, S. G.; Murgo, A. J.; Sausville, E. A., Promising new agents under development by the Division of Cancer Treatment, Diagnosis, and Centers of the National Cancer Institute. *Seminars in Oncology* **1997,** *24* (2), 219-240.

74. Law, C. L.; Gordon, K. A.; Toki, B. E.; Yamane, A. K.; Hering, M. A.; Cerveny, C. G.; Petroziello, J. M.; Ryan, M. C.; Smith, L.; Simon, R.; Sauter, G.; Oflazoglu, E.; Doronina, S. O.; Meyer, D. L.; Francisco, J. A.; Carter, P.; Senter, P. D.; Copland, J. A.; Wood, C. G.; Wahl, A. F., Lymphocyte activation antigen CD70 expressed by renal cell carcinoma is a potential therapeutic target for anti-CD70 antibody-drug conjugates. *Cancer Research* **2006,** *66* (4), 2328-2337.

75. Sutherland, M. S. K.; Sanderson, R. J.; Gordon, K. A.; Andreyka, J.; Cerveny, C. G.; Yu, C. P.; Lewis, T. S.; Meyer, D. L.; Zabinski, R. F.; Doronina, S. O.; Senter, P. D.; Law, C. L.; Wahl, A. F., Lysosomal trafficking and cysteine protease metabolism confer target-specific cytotoxicity by peptide-linked anti-CD30-auristatin conjugates. *Journal of Biological Chemistry* **2006,** *281* (15), 10540-10547.

76. Kirschning, A.; Hahn, F., Merging Chemical Synthesis and Biosynthesis: A New Chapter in the Total Synthesis of Natural Products and Natural Product Libraries. *Angewandte Chemie-International Edition* **2012,** *51* (17), 4012-4022.

77. Koehn, F. E., Biosynthetic medicinal chemistry of natural product drugs. *Medchemcomm* **2012,** *3* (8), 854-865.

78. Wani, M. C.; Taylor, H. L.; Wall, M. E.; Coggon, P.; McPhail, A. T., PLANT ANTITUMOR AGENTS .6. ISOLATION AND STRUCTURE OF TAXOL, A NOVEL

ANTILEUKEMIC AND ANTITUMOR AGENT FROM TAXUS-BREVIFOLIA. *Journal of the American Chemical Society* **1971,** *93* (9), 2325-&.

79. Holton, R. A.; Somoza, C.; Kim, H. B.; Liang, F.; Biediger, R. J.; Boatman, P. D.; Shindo, M.; Smith, C. C.; Kim, S. C.; Nadizadeh, H.; Suzuki, Y.; Tao, C. L.; Vu, P.; Tang, S. H.; Zhang, P. S.; Murthi, K. K.; Gentile, L. N.; Liu, J. H., FIRST TOTAL SYNTHESIS OF TAXOL .1. FUNCTIONALIZATION OF THE B-RING. *Journal of the American Chemical Society* **1994,** *116* (4), 1597-1598.

80. Holton, R. A.; Kim, H. B.; Somoza, C.; Liang, F.; Biediger, R. J.; Boatman, P. D.; Shindo, M.; Smith, C. C.; Kim, S. C.; Nadizadeh, H.; Suzuki, Y.; Tao, C. L.; Vu, P.; Tang, S. H.; Zhang, P. S.; Murthi, K. K.; Gentile, L. N.; Liu, J. H., FIRST TOTAL SYNTHESIS OF TAXOL .2. COMPLETION OF THE C-RING AND D-RING. *Journal of the American Chemical Society* **1994,** *116* (4), 1599-1600.

81. Holton, R. A.; Somoza, C.; Kim, H. B.; Liang, F.; Biediger, R. J.; Boatman, P. D.; Shindo, M.; Smith, C. C.; Kim, S.; Nadizadeh, H.; Suzuki, Y.; Tao, C. L.; Vu, P.; Tang, S. H.; Zhang, P. S.; Murthi, K. K.; Gentile, L. N.; Liu, J. H., THE TOTAL SYNTHESIS OF PACLITAXEL STARTING WITH CAMPHOR. In *Taxane Anticancer Agents: Basic Science and Current Status*, Georg, G. I.; Chem, T. T.; Ojima, I.; Vyas, D. M., Eds. 1995; Vol. 583, pp 288-301.

82. Cragg, G. M.; Newman, D. J.; Yang, S. S., Natural product extracts of plant and marine origin having antileukemia potential. The NCI experience. *Journal of Natural Products* **2006,** *69* (3), 488-498.

83. Pereda-Miranda, R.; Rosas-Ramirez, D.; Castaneda-Gomez, J., Resin glycosides from the morning glory family. *Prog. Chem. Org. Nat. Prod.* **2010,** *92*, 77-153.

84. Pereda-Miranda, R.; Bah, M., Biodynamic constituents in the Mexican morning glories: Purgative remedies transcending boundaries. *Current Topics in Medicinal Chemistry* **2003,** *3* (2), 111-131.

85. Huguet-Termes, T., New world materia medica in Spanish renaissance medicine: From scholarly reception to practical impact. *Medical History* **2001,** *45* (3), 359-376.

86. Pereda-Miranda, R.; Fragoso-Serrano, M.; Escalante-Sanchez, E.; Hernandez-Carlos, B.; Linares, E.; Bye, R., Profiling of the resin glycoside content of Mexican jalap roots with purgative activity. *Journal of Natural Products* **2006,** *69* (10), 1460-1466.

87. Pereda-Miranda, R.; Rosas-Ramírez, D.; Castañeda-Gómez, J., Resin Glycosides from the Morning Glory Family. In *Fortschritte der Chemie organischer Naturstoffe / Progress in the Chemistry of Organic Natural Products, Vol. 92*, Kinghorn, A. D.; Falk, H.; Kobayashi, J., Eds. Springer Vienna: Vienna, 2010; pp 77-153.

88. Peredamiranda, R.; Mata, R.; Anaya, A. L.; Wickramaratne, D. B. M.; Pezzuto, J. M.; Kinghorn, A. D., TRICOLORIN-A, MAJOR PHYTOGROWTH INHIBITOR FROM IPOMOEA-TRICOLOR. *Journal of Natural Products* **1993,** *56* (4), 571-582.

89. Bendz, G.; Santesson, J., NOBEL SYMPOSIUM NO 25 CHEMISTRY IN BOTANICAL CLASSIFICATION. *Nobel Symposium* **1973**, 320-(1974) 320.

90. Dembitsky, V. M., Chemistry and biodiversity of the biologically active natural glycosides. *Chemistry & Biodiversity* **2004,** *1* (5), 673-781.

91. Gaspar, E. M. M., Soldanelline B - The first acylated nonlinear tetrasaccharide macrolactone from the European Convolvulaceae Calystegia soldanella. *European Journal of Organic Chemistry* **2001,** (2), 369-373.

92. Noda, N.; Kogetsu, H.; Kawasaki, T.; Miyahara, K., RESIN GLYCOSIDES .12. SCAMMONIN-VII AND SCAMMONIN-VIII, 2 RESIN GLYCOSIDES FROM CONVOLVULUS-SCAMMONIA. *Phytochemistry* **1992,** *31* (8), 2761-2766.

93. Donnapee, S.; Li, J.; Yang, X.; Ge, A. H.; Donkor, P. O.; Gao, X. M.; Chang, Y. X., Cuscuta chinensis Lam.: A systematic review on ethnopharmacology, phytochemistry and pharmacology of an important traditional herbal medicine. *Journal of Ethnopharmacology* **2014,** *157*, 292-308.

94. Takigawa, A.; Muto, H.; Kabata, K.; Okawa, M.; Kinjo, J.; Yoshimitsu, H.; Nohara, T.; Ono, M., Calysolins I-IV, Resin Glycosides from Calystegia soldanella. *J. Nat. Prod.* **2011,** *74* (11), 2414-2419.

95. Ito, Y., My Stroll in the Backyard of Carbohydrate Chemistry. *Trends in Glycoscience and Glycotechnology* **2010,** *22* (125), 119-140.

96. Ono, M.; Yamada, F.; Noda, N.; Kawasaki, T.; Miyahara, K., RESIN GLYCOSIDES .18. DETERMINATION BY MOSHERS METHOD OF THE ABSOLUTE-CONFIGURATIONS OF MONO- AND DIHYDROXYFATTY ACIDS ORIGINATED FROM RESIN GLYCOSIDES. *Chemical & Pharmaceutical Bulletin* **1993,** *41* (6), 1023-1026.

97. Deng, S. J.; Yu, B.; Lou, Y.; Hui, Y. Z., First total synthesis of an exceptionally potent antitumor saponin, OSW-1. *Journal of Organic Chemistry* **1999,** *64* (1), 202-208.

98. Furstner, A.; Muller, T., Metathesis route to resin glycosides: Formal total synthesis of tricolorin A. *Journal of Organic Chemistry* **1998,** *63* (3), 424-425.

99. Furstner, A.; Muller, T., Efficient total syntheses of resin glycosides and analogues by ring-closing olefin metathesis. *Journal of the American Chemical Society* **1999,** *121* (34), 7814-7821.

100. Jiang, Z. H.; Geyer, A.; Schmidt, R. R., THE MACROLIDIC GLYCOLIPID CALONYCTIN-A, A PLANT-GROWTH REGULATOR - SYNTHESIS, STRUCTURAL

ASSIGNMENT, AND CONFORMATIONAL-ANALYSIS IN MICELLAR SOLUTION. *Angewandte Chemie-International Edition in English* **1995,** *34* (22), 2520-2524.

101. Schmidt, R. R., New approaches to glycoconjugate synthesis. *Pure and Applied Chemistry* **1998,** *70* (2), 397-402.

102. Ohashi, I.; Lear, M. J.; Yoshimura, F.; Hirama, M., Use of polystyrene-supported DBU in the synthesis and alpha-selective glycosylation study of the unstable Schmidt donor of L-kedarosamine. *Organic Letters* **2004,** *6* (5), 719-722.

103. Patel, K.; Song, F. L.; Andreana, P. R., Synthesis of substrate analogues as potential inhibitors for Mycobacterium tuberculosis enzyme MshC. *Carbohydrate Research* **2017,** *453-454*, 10-18.

104. Paterson, I.; Davies, R. D. M.; Heimann, A. C.; Marquez, R.; Meyer, A., Stereocontrolled total synthesis of (-)-callipeltoside A. *Organic Letters* **2003,** *5* (23), 4477-4480.

105. Sato, E.; Sato, M.; Tanabe, Y.; Nakajima, N.; Ohkubo, A.; Suenaga, K., Total Synthesis of Biselyngbyaside. *Journal of Organic Chemistry* **2017,** *82* (13), 6770-6777.

106. Bah, M.; PeredaMiranda, R., Detailed FAB mass spectrometry and high resolution NMR investigations of Tricolorins A-E, individual oligosaccharides from the resins of Ipomoea tricolor (Convolvulaceae). *Tetrahedron* **1996,** *52* (41), 13063-13080.

107. Bah, M.; PeredaMiranda, R., Bioactive natural products from traditionally used Mexican plants .5. Isolation and structural characterization of new glycolipid ester type dimers from the resin of Ipomoea tricolor (Convolvulaceae). *Tetrahedron* **1997,** *53* (27), 9007-9022.

108. Larson, D. P.; Heathcock, C. H., Synthesis of the macrolactone disaccharide subunit of tricolorin A. *Journal of Organic Chemistry* **1996,** *61* (16), 5208-5209.

109. Larson, D. P.; Heathcock, C. H., Total synthesis of tricolorin A. *Journal of Organic Chemistry* **1997,** *62* (24), 8406-8418.

110. Hikota, M.; Sakurai, Y.; Horita, K.; Yonemitsu, O., CHIRAL SYNTHESIS OF POLYKETIDE-DERIVED NATURAL-PRODUCTS .32. SYNTHESIS OF ERYTHRONOLIDE A VIA A VERY EFFICIENT MACROLACTONIZATION UNDER USUAL ACYLATION CONDITIONS WITH THE YAMAGUCHI REAGENT. *Tetrahedron Letters* **1990,** *31* (44), 6367-6370.

111. Hough, L.; Shankari.Pn; Stacey, B. E., METABOLISM OF C-14-LABELED D-GLUCOSE, D-FRUCTOSE AND ALLITOL BY ITEA PLANTS. *Phytochemistry* **1973,** *12* (3), 573-582.

112. Leon-Rivera, I.; Miron-Lopez, G.; Estrada-Soto, S.; Aguirre-Crespo, F.; Gutierrez, M. D.; Molina-Salinas, G. M.; Hurtado, G.; Navarrete-Vazquez, G.; Montiel, E., Glycolipid ester-

type heterodimers from Ipomoea tyrianthina and their pharmacological activity. *Bioorganic & Medicinal Chemistry Letters* **2009,** *19* (16), 4652-4656.

113. Rosas-Ramirez, D.; Escalante-Sanchez, E.; Pereda-Miranda, R., Batatins III-VI, glycolipid ester-type dimers from Ipomoea batatas. *Phytochemistry* **2011,** *72* (8), 773-780.

114. Kufner, U.; Schmidt, R. R., DENOVO SYNTHESIS OF CARBOHYDRATES AND RELATED NATURAL-PRODUCTS .20. SYNTHESIS OF DEOXYHEXOSES FROM DIVINYLGLYCOLS - THE SYNTHESIS OF D-CHALCOSE AND L-CHALCOSE. *Angewandte Chemie-International Edition in English* **1986,** *25* (1), 89-89.

115. Abdouni, Y.; Yilmaz, G.; Becer, C. R., Sequence and Architectural Control in Glycopolymer Synthesis. *Macromolecular Rapid Communications* **2017,** *38* (24).

116. Bi, N. N.; Xiong, C. H.; Jin, G. X.; Guo, Z. W.; Gu, G. F., Synthesis of a trisaccharide repeating unit of the O-antigen from Burkholderia cenocepacia and its dimer. *Carbohydrate Research* **2017,** *451*, 1-11.

117. Williams, R.; Galan, M. C., Recent Advances in Organocatalytic Glycosylations. *European Journal of Organic Chemistry* **2017,** (42), 6247-6264.

118. Furstner, A., Total syntheses and biological assessment of macrocyclic glycolipids. *European Journal of Organic Chemistry* **2004,** (5), 943-958.

119. Singh, S. K.; Felse, A. P.; Nunez, A.; Foglia, T. A.; Gross, R. A., Regioselective enzyme-catalyzed synthesis of sophorolipid esters, amides, and multifunctional monomers. *Journal of Organic Chemistry* **2003,** *68* (14), 5466-5477.

120. Karanth, N. G. K.; Deo, P. G.; Veenanadig, N. K., Microbial production of biosurfactants and their importance. *Current Science* **1999,** *77* (1), 116-126.

121. Nawale, L.; Dubey, P.; Chaudhari, B.; Sarkar, D.; Prabhune, A., Anti-proliferative effect of novel primary cetyl alcohol derived sophorolipids against human cervical cancer cells HeLa. *Plos One* **2017,** *12* (4).

122. Furstner, A.; Radkowski, K.; Grabowski, J.; Wirtz, C.; Mynott, R., Ring-closing alkyne metathesis. Application to the total synthesis of sophorolipid lactone. *Journal of Organic Chemistry* **2000,** *65* (25), 8758-8762.

123. Cristobal Lopez, J.; Plumet, J., Metathesis Reactions of Carbohydrates: Recent Highlights in Alkyne Metathesis. *European Journal of Organic Chemistry* **2011,** (10), 1803-1825.

124. Fuerstner, A., Alkyne Metathesis on the Rise. *Angewandte Chemie-International Edition* **2013,** *52* (10), 2794-2819.

125. Cao, S. G.; Guza, R. C.; Wisse, J. H.; Miller, J. S.; Evans, R.; Kingston, D. G. I., Ipomoeassins A-E, cytotoxic macrocyclic glycoresins from the leaves of Ipomoea squamosa from the Suriname rainforest. *Journal of Natural Products* **2005,** *68* (4), 487-492.

126. Ono, M.; Akiyama, K.; Yamamoto, K.; Mineno, T.; Okawa, M.; Kinjo, J.; Miyashita, H.; Yoshimitsu, H.; Nohara, T., Four New Acylated Glycosidic Acid Methyl Esters Isolated from the Convolvulin Fraction of Seeds of Quamoclit pennata after Treatment with Indium(III) Chloride in Methanol. *Chemical & Pharmaceutical Bulletin* **2014,** *62* (8), 830-835.

127. Ono, M., Resin glycosides from Convolvulaceae plants. *J. Nat. Med.* **2017,** *71* (4), 591-604.

128. Escalante-Sanchez, E.; Pereda-Miranda, R., Batatins I and II, ester-type dimers of acylated pentasaccharides from the resin glycosides of sweet potato. *Journal of Natural Products* **2007,** *70* (6), 1029-1034.

129. Escobedo-Martinez, C.; Pereda-Miranda, R., Resin glycosides from Ipomoea pes-caprae. *Journal of Natural Products* **2007,** *70* (6), 974-978.

130. Ono, M., Resin glycosides from Convolvulaceae plants. *Journal of Natural Medicines* **2017,** *71* (4), 591-604.

131. Cao, S.; Norris, A.; Wisse, J. H.; Miller, J. S.; Evans, R.; Kingston, D. G. I., Ipomoeassin F, a new cytotoxic macrocyclic glycoresin from the leaves of Ipomoea squamosa from the Suriname rainforest. *Nat. Prod. Res.* **2007,** *21* (10), 872-876.

132. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Muller, R.; Furstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chemistry-a European Journal* **2009,** *15* (38), 9697-9706.

133. Furstner, A.; Nagano, T., Total syntheses of ipomoeassin B and E. *Journal of the American Chemical Society* **2007,** *129* (7), 1906-+.

134. Postema, M. H. D.; TenDyke, K.; Cutter, J.; Kuznetsov, G.; Xu, O. L., Total Synthesis of Ipomoeassin F. *Organic Letters* **2009,** *11* (6), 1417-1420.

135. Zong, G. H.; Barber, E.; Aljewari, H.; Zhou, J. H.; Hu, Z. J.; Du, Y. C.; Shi, W. Q., Total Synthesis and Biological Evaluation of Ipomoeassin F and Its Unnatural 11R-Epimer. *Journal of Organic Chemistry* **2015,** *80* (18), 9279-9291.

136. Zong, G. H.; Aljewari, H.; Hu, Z. J.; Shi, W. Q., Revealing the Pharmacophore of Ipomoeassin F through Molecular Editing. *Organic Letters* **2016,** *18* (7), 1674-1677.

137. Zong, G. H.; Hirsch, M.; Mondrik, C.; Hu, Z. J.; Shi, W. Q., Design, synthesis and biological evaluation of fucose-truncated monosaccharide analogues of ipomoeassin F. *Bioorganic & Medicinal Chemistry Letters* **2017,** *27* (12), 2752-2756.

138. Zong, G. H.; Whisenhunt, L.; Hu, Z. J.; Shi, W. Q., Synergistic Contribution of Tiglate and Cinnamate to Cytotoxicity of Ipomoeassin F. *Journal of Organic Chemistry* **2017,** *82* (9), 4977-4985.

139. Mlynarski, J.; Ruiz-Caro, J.; Furstner, A., Total synthesis of macroviracin D (BA-2836-4). *Chemistry-a European Journal* **2004,** *10* (9), 2214-2222.

140. Kametani, T.; Tsubuki, M.; Tatsuzaki, Y.; Honda, T., SYNTHESIS OF OPTICALLY-ACTIVE 2-FURYLMETHANOLS AS USEFUL CHIRAL BUILDING-BLOCKS AND ITS APPLICATION TO THE SYNTHESIS OF (5R,6S)-6-ACETOXYHEXADECAN-5-OLIDE AND (+)-DISPARLURE. *Journal of the Chemical Society-Perkin Transactions 1* **1990,** (3), 639-646.

141. Kusakabe, M.; Kitano, Y.; Kobayashi, Y.; Sato, F., PREPARATION OF OPTICALLY-ACTIVE 2-FURYLCARBINOLS BY KINETIC RESOLUTION USING THE SHARPLESS REAGENT AND THEIR APPLICATION IN ORGANIC-SYNTHESIS. *Journal of Organic Chemistry* **1989,** *54* (9), 2085-2091.

142. Ho, T. L.; Sapp, S. G., SHARPLESS EPOXIDATION OF 2-FURANCARBINOLS - MASS-SPECTRA OF 6-HYDROXY-3(2H)PYRANONES. *Synthetic Communications* **1983,** *13* (3), 207-211.

143. Aissa, C.; Riveiros, R.; Ragot, J.; Furstner, A., Total syntheses of amphidinolide T1, T3, T4, and T5. *Journal of the American Chemical Society* **2003,** *125* (50), 15512-15520.

144. Scheidt, K. A.; Chen, H.; Follows, B. C.; Chemler, S. R.; Coffey, D. S.; Roush, W. R., Tris(dimethylamino)sulfonium difluorotrimethylsilicate, a mild reagent for the removal of silicon protecting groups. *Journal of Organic Chemistry* **1998,** *63* (19), 6436-6437.

145. Pescarmona, P. P.; Masters, A. F.; van der Waal, J. C.; Maschmeyer, T., Osmium silsesquioxane as model compound and homogeneous catalyst for the dihydroxylation of alkenes. *Journal of Molecular Catalysis a-Chemical* **2004,** *220* (1), 37-42.

146. Rodushkin, I.; Engstrom, E.; Sorlin, D.; Baxter, D.; Hornfeldt, B.; Nyholm, E.; Ecke, F., Uptake and Accumulation of Anthropogenic Os in Free-Living Bank Voles (Myodes glareolus). *Water Air and Soil Pollution* **2011,** *218* (1-4), 603-610.

147. Schaus, S. E.; Brandes, B. D.; Larrow, J. F.; Tokunaga, M.; Hansen, K. B.; Gould, A. E.; Furrow, M. E.; Jacobsen, E. N., Highly selective hydrolytic kinetic resolution of terminal epoxides catalyzed by chiral (salen)Co-III complexes. Practical synthesis of enantioenriched terminal epoxides and 1,2-diols. *Journal of the American Chemical Society* **2002,** *124* (7), 1307-1315.

148. Lhommet, G.; Freville, S.; Thuy, V.; Petit, H.; Celerier, J. P., A general and versatile synthesis of 4- and 5-oxoacids. *Synthetic Communications* **1996,** *26* (21), 3897-3901.

149. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Mueller, R.; Fuerstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chem. - Eur. J.* **2009,** *15* (38), 9697-9706, S9697/1-S9697/48.

150. Chevalier, R.; Colsch, B.; Afonso, C.; Baumann, N.; Tabet, J. C.; Mallet, J. M., Synthetic sulfated glucuronosyl paragloboside (SGPG) and its use for the detection of autoimmune peripheral neuropathies. *Tetrahedron* **2006,** *62* (4), 563-577.

151. Zong, G. H.; Yan, S. Q.; Liang, X. M.; Zhang, J. J.; Wang, D. Q.; Kong, F. Z., Highly efficient removal of allyloxycarbonyl (Alloc) function provides a practical orthogonal protective strategy for carbohydrates. *Chinese Chemical Letters* **2009,** *20* (2), 127-130.

152. Pan, S. J.; Zhang, H. L.; Wang, C. Y.; Yao, S. C. L.; Yao, S. Q., Target identification of natural products and bioactive compounds using affinity-based probes. *Natural Product Reports* **2016,** *33* (5), 612-620.

CHAPTER 2. SYNTHESIS OF THE C-3 MODIFIED IPOMOEASSIN F ANALOGS

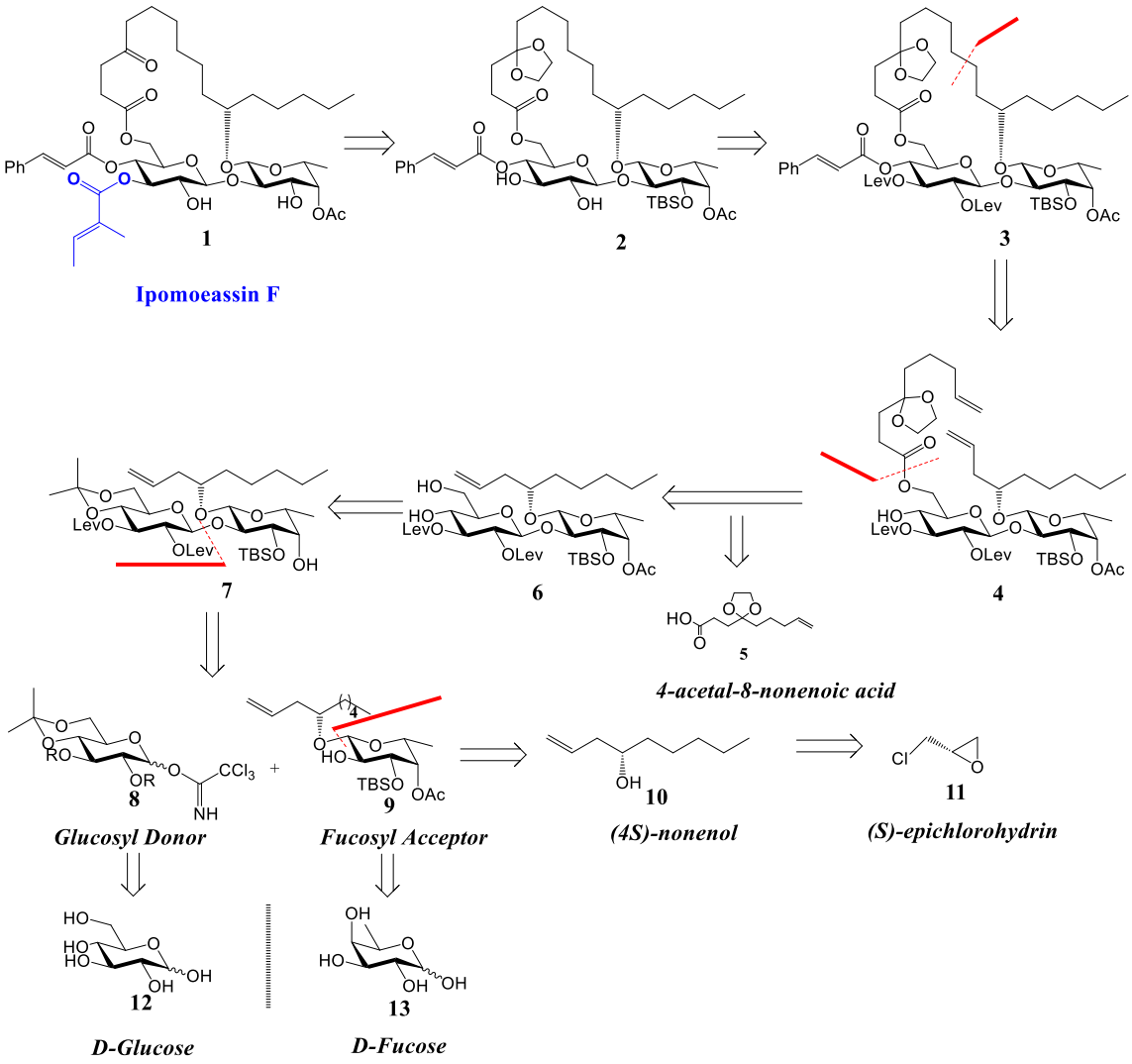
Even with the invention of modern technologies and good reaction profiles carbohydrate synthesis continue to be a challenge task for organic chemists. Carbohydrates possess greater structurally diversity than most other natural products (branching character, stereochemical issues, different glyosidic bonds, etc.). Also, carbohydrates contain a significantly higher number of heteroatoms and hydroxyl groups, when compared to other classes of natural products.[1] This has caused chemists to develop different protecting groups to ensure the correct glyosidic bonds and ester linkages are obtained. Some chemists have started using enzymes to guarantee the correct regio-selectivity and stereoselectivity, but this process is takes time and can be very expensive.[2]

Recently, natural products containing carbohydrates have become relevant because of their potential anti-cancer applications.[3, 4] One interesting class of natural products containing carbohydrates is the family of ipomoeassins.[5, 6] Since 2013, Shi *et al.* have been studying the ipomoeassins, specifically ipomoeassin F **1**, to gain a greater knowledge into the mechanism of action (MOA) of these resin glycosides. One portion of ipomoeassin F **1** that had not been well studied was the C-3-Glc*p* or the tigoyl (tig) moiety. Shi *et al.* knew from previous studies that the C-3-Glc*p* tig moiety was vital to the cytotoxicity but did not know if modifications could be made to extend ipomoeassins into new/advanced area of drug discovery.

2.1 Retro-Synthesis

To study the role of tig, an efficient and scalable synthesis route needed to be developed. We envisioned a route that would introduce the C-4-Glc*p* cinnamoyl (cinn) and the C-3-Glc*p* tig in the late-stage.[7-9] Since, RCM reactions have been shown to be highly efficient and effective in

constructing large macrocycles, this strategy was adopted like in the previous syntheses of ipomoeassins.[10, 11] The retro-synthetic scheme for making C-3-Glc*p* modified ipomoeassin F analogs is shown in Scheme 2.1 (bond disconnections in red). A regioselective esterification could be performed at the C-3-Glc*p* followed by a one-pot deprotection of the *C-4* acetal and the -OTBS at C-3-Fuc*p* to achieve the final analogs from **2**. We believed the final deprotection could happen in one pot based on the previous studies of OSW-1 and other complex natural products.[12, 13] The OH-3-Glc*p* should be favored for the esterification over the OH-2-Glc*p* because of greater nucleophilicity of the OH-3-Glc*p*. Also, Fürstner *et al.* showed that regioselective esterification



Scheme 2.1. Retrosynthesis of the C-3-Glc*p* modified ipomoeassin F **1** analogs.

could be performed at the more nucleophilic *C-3*-Glc*p* site (9:1) in a ipomoeassins B and E synthesis.[10, 14] However, their substrate for the regio-selective esterification was significantly simpler, since they had only constructed the non-cyclical (1→2)-*β*-disaccharide.[8] Also, the -OTBS at C-3-Fuc*p* should help to block the OH-2-Fuc*p* making the regioselective esterification of OH-3-Glc*p* more favorable.

The diol intermediate **2** was the most important intermediate for the synthesis because different modifications were regio-selectively introduced, as discussed above. The levulinoyl (lev) group was chosen as the transient protecting group for the 2-*O*-Glc*p* and 3-*O*-Glc*p* for a few different reasons. The lev groups can be orthogonally deprotected in the late-stage using hydrazine acetate (3→2) ($NH_2NH_2 \cdot AcOH$). Moreover, the neighboring group participation effect of the lev group helped to control the *β*-glycosylation formation found in ipomoeassin F **1** (glycone). However, the *C-4* ketone in the aglycone **5** had be protected to avoid hydrazination when the lev groups were removed. The macrocycle **3** was obtained through an RCM and hydrazination sequence from the diene **4**. Another benefit for the late-stage introduction of the C-4-Glc*p* cinn and the C-3-Glc*p* tig was that the cis/trans RCM intermediate could be effectively reduced through strong hydrogenation conditions. In the previous syntheses of the ipomoeassins, the hydrogenation of the cis/trans double bond intermediate caused Fürstner and Postema some diffcultlies.[9, 15] The diene **4** could be preparaed through a regioselctive esterification between the 4-acetal-8-nonenoic acid **5** and the OH-6-Glc*p* diol **6**. Mutliple esterification methods were tried with Mukaiyama esterification conditions being favored due to a simpler work-up and purification process.

The diol **6** could be assembled though deprotection of the isopropylidene group and acetylation of the OH-4-Fucose (Fuc*p*). The (1→2)-*β*-disaccharide intermediate **7** could be readily

made by a chemo-selective glycosylation of the glucosyl (Glc*p*) donor **8** and fucosyl (Fuc*p*) acceptor **9**. The *β*-configuration could be achieved exclusively via neighboring group participation at C-2-Glc*p*.[2, 16-18] The Glc*p* donor **8** could be assembled through a series of chemical transformations from the commercially available D-glucose (Glc*p*) **13**. The fucosyl (Fuc*p*) acceptor **9** can be prepared from D-fucose (Fuc*p*) **13** though a glycosylation of a Schmidt donor and the (4*S*)-nonenol **10**. Like the glycosylation affording the (1→2)-*β*-disaccharide **7**, neighboring group participation could be utilized to obtain the *β*-configuration.[2, 16-18] To establish the (11*S*)-stereocenter of the aglycone in ipomoeassin F **1**, the (4*S*)-nonenol **10** could be prepared from commercially available (S)-epichlorohydrin **11**. This synthesis route features multiple glycosylation reactions, two regio-selective esterification, and an RCM reaction to form the macrocycle, along with other chemical transformations. The work presented in the following chapter presents an efficient and scalable synthesis route to study the role of the C-3-Glc*p* moiety of ipomoeassin F **1**.

2.2. Synthesis of the Aglycone of Ipomoeassin F

The aglycone moiety of ipomoeassin F **1** is highlighted in red and blue in Figure 2.1. RCM reactions have been found to be efficient for the synthesis of large macrocycles, including

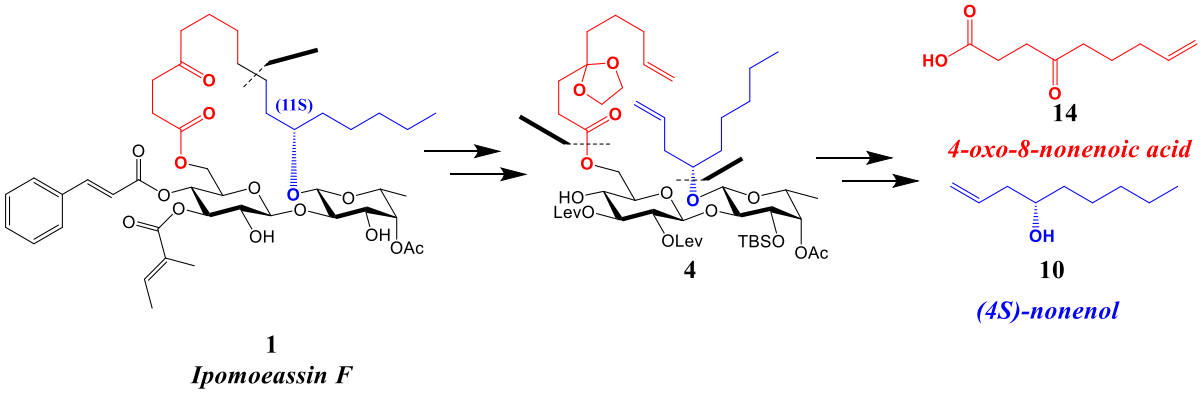
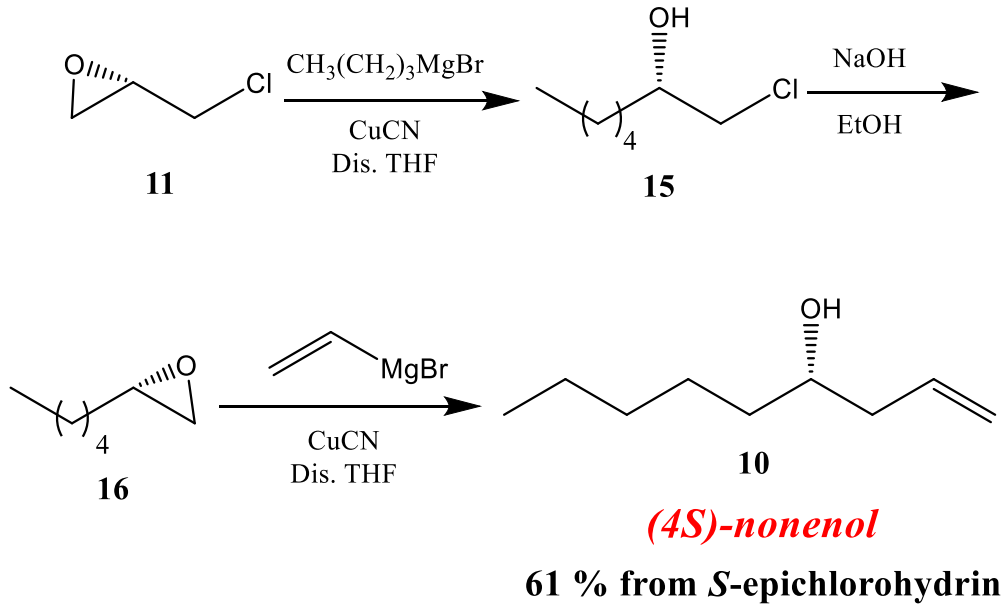


Figure 2.1. Parts of the aglycone moiety of ipomoeassin F **1**.

ipomoeassins. For this project ipomoeassin F **1** aglycone would be composed of the 4-oxo-8-nonenoic acid **14** (left tether) and (4*S*)-nonenol **10** (right tether). So, a retro-synthetic scheme was envisioned that would prepare the left and right tethers of the aglycone separately. Separately preparing the two tethers and performing an RCM reaction would make the synthesis more convergent than if a macrolactonization approach were developed. Also, we wanted to study the importance of the aglycone for the cytotoxicity of ipomoeassin F **1**. Thus**,** preparing each tether of the aglycone separately allowed for flexible modifications without affecting the synthesis of other fragments of ipomoeassin F **1**. The tail of the aglycone (*C-11–C-16*) has been found to be very important to the cytotoxicity of ipomoeassins.[19-21] The cytotoxicity decreased by 5-100-fold, when the length of the aglycone tail was decreased by 2 carbons (ipomo. F vs. ipomo. A). The published data shows the addition of the two methylene units affects the lipophilicity of the backbone of the ipomoeassins.[22] The following section will cover in detail the synthesis of the aglycone of ipomoeassin F **1**.

2.2.1. Preparation of (4S)-nonen-4-ol

The (4*S*)-nonen-4-ol **10** can be prepared from commercially available (*S*)-epichlorohydrin **11** in three steps (Scheme 2.2). Using (*S*)-epichlorohydrin **11** may not be the most cost-effective method for the synthesis of the aglycone; however, the starting material **11** does establish the (*S*)-configuration of the ipomoeassin F **1** aglycone. The only stereogenic center (11*S*) on the aglycone has proven to be of great importance to the cytotoxicity of ipomoeassin F **1**.[7] Also, the five carbon tail seemed to be very important for the cytotoxicity of ipomoeassins.[19-21] Butylmagnesium bromide in the presence of copper (I) cyanide (CuCN) was used to open up the 1,2-exopide ring of **11** to give the 1-chloro-3-heptanol **15**. Since, this Grignard reaction was performed on a

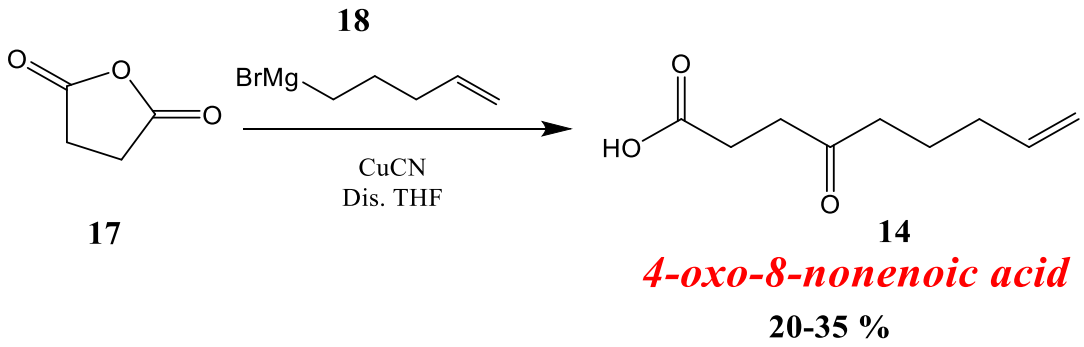


Scheme 2.2. Synthesis of the (4*S*)-nonenol **10** from (*S*)-epichlorohydrin **11**.

relatively large scale (10-15 g), the formation of the Grignard reagent had to be controlled.[23] Treating the α-chloro alcohol **15** with an excess of sodium hydroxide formed a 1,2-expoxide intermediate **16** through an intramolecular S_N2 reaction.[24, 25] Lastly, another Grignard reaction was performed with vinylmagnesium bromide to open-up the 1,2-expoxide ring **16** that give the chiral alcohol **10**.[26, 27] These three reactions were performed in succession without any purification until reaching the (4*S*)-nonen-4-ol **10**. The overall yield from *S*-epichlorohydrin **11** to (4*S*)-nonen-4-ol **10** was 61%.

2.2.2. Synthesis of 4-oxo-8-nonenoic acid

The synthesis of the 4-oxo-noenoic acid **14** was more tedious than the (4*S*)-nonen-4-ol **11**. In previous syntheses of the ipomoeassins, the 4-oxo-8-nonenoic acid **14** (Scheme 2.3) could be prepared through a Grignard reaction in one step from succinic anhydride **44** and 4-pentenylmagnesium bromide **45**. However, the yield of 4-oxx-8-nonenoic acid **14** was low (20-



Scheme 2.3. Original route for the preparation of the 4-oxo-8-nonenoic acid **14**.

35%) even though literature examples showed the yield of similar Grignard reactions could be increased to almost 60%. One of the main reasons for the low yield was any excess Grignard reagent would react another at the *C-4* ketone giving a tertiary alcohol. During the purification of **14**, streaking was observed on the TLC and the column. This situation was resolved by adding a few drops of acetic acid to the eluent system. Despite the drawback in in the Grignard reaction, this method allowed for the preparation of 4-oxo-noenoic acid **14** in one step on a multi-gram scale.

2.3. Preparation of the Fucosyl Acceptor

The synthesis of the Fuc*p* acceptor **9** is more straight forward than the synthesis of the Glc*p* donor **8**. A route was envisioned to start from commercially available D-Fuc*p* **13** and (4*S*)-nonenol **10** to make-up the left tether of the aglycone (Figure 2.2). After the formation of a Fuc*p* Schmidt donor, a glycosylation could be performed with (4*S*)-nonenol **10**. Then, after a few chemical transformations, a regioselective introduction of TBS was envigored based on the nucleophilicity of *H*O-3-Fuc*p*.[7, 15] The increased activity of the *H*O-C-3 of monosaccharides has been well established in the literature. It was believed this method would provide an efficient, scalable route with the use of minimal protections for the preparation of the Fuc*p* acceptor **9**.

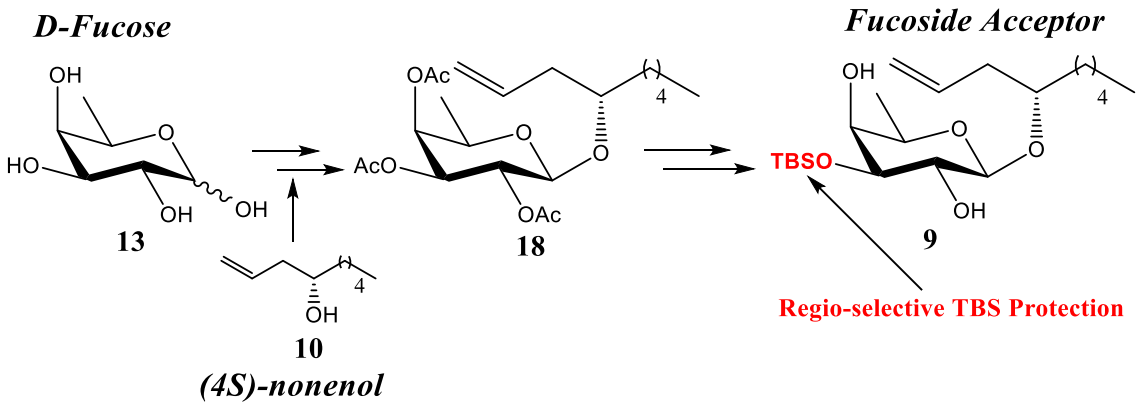


Figure 2.2. The preparation of the Fuc*p* acceptor **9** from D-Fuc*p* **13**.

During the scaling-up process of this project, the focus shifted from the methodology to the scalability of each chemical transformation. Factors such as the design of experiment (DOE) and principle cost analysis (PCA) had to be considered to perform the reaction on a multi-gram scale. One of the biggest cost for an academic research laboratory is the cost of the chemicals and personnel. As shown in the Figure 2.3 most of the starting materials of ipomoeassin F **1** are cheap (< $1/ 1g). However, the price of the (*S*)-epichlorohydrin **11** and D-Fuc*p* **13** are significantly

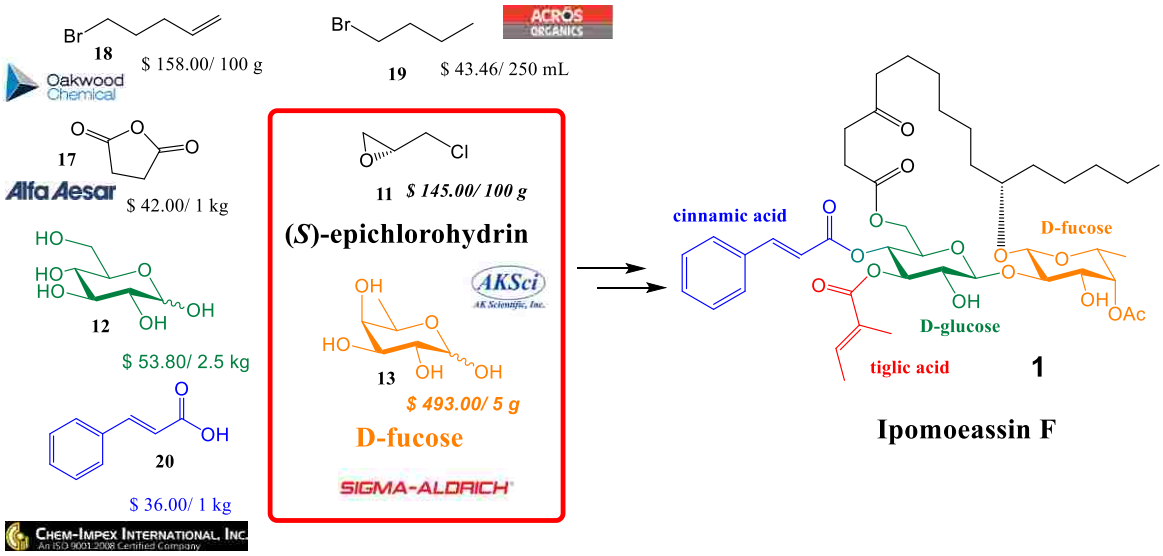


Figure 2.3. Scalability of the synthesis route for ipomoeassin F **1** and C-3-Glcp analogs.

higher than the other starting materials of ipomoeassin F **1**. Using the enantiopure (*S*)-epichlorohydrin **11** was justifiable because it established the (11*S*)-configuration of the overall compound. As, the (11*S*)-configuration has been found to be vital to the cytotoxicity of ipomoeassins F. D-Fuc*p* **13** was the most expensive starting material for this project (Sigma-Aldrich: $493.00/5g). This high price of D-Fuc*p* **13** caused us to think, could a cheaper and more commercially available starting material be used instead of D-Fuc*p* **13**? Then, through a series of chemical transformations prepare the D-Fuc*p* **13**. By starting with a simpler monosaccharide, the number of steps increased, but allowed for a cheaper monosaccharide for the multi-gram synthesis of the Fuc*p* acceptor **9**.

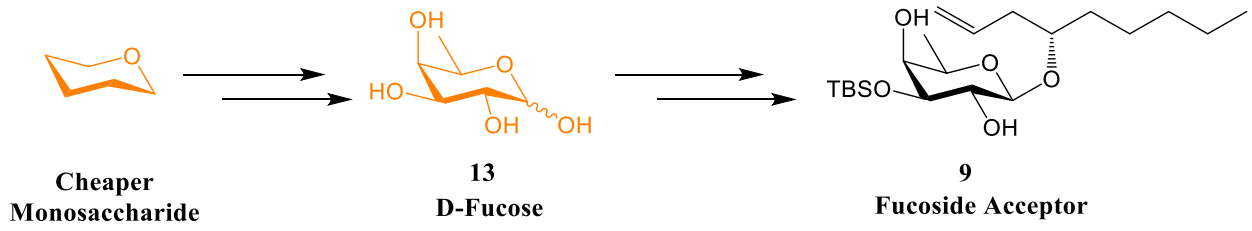
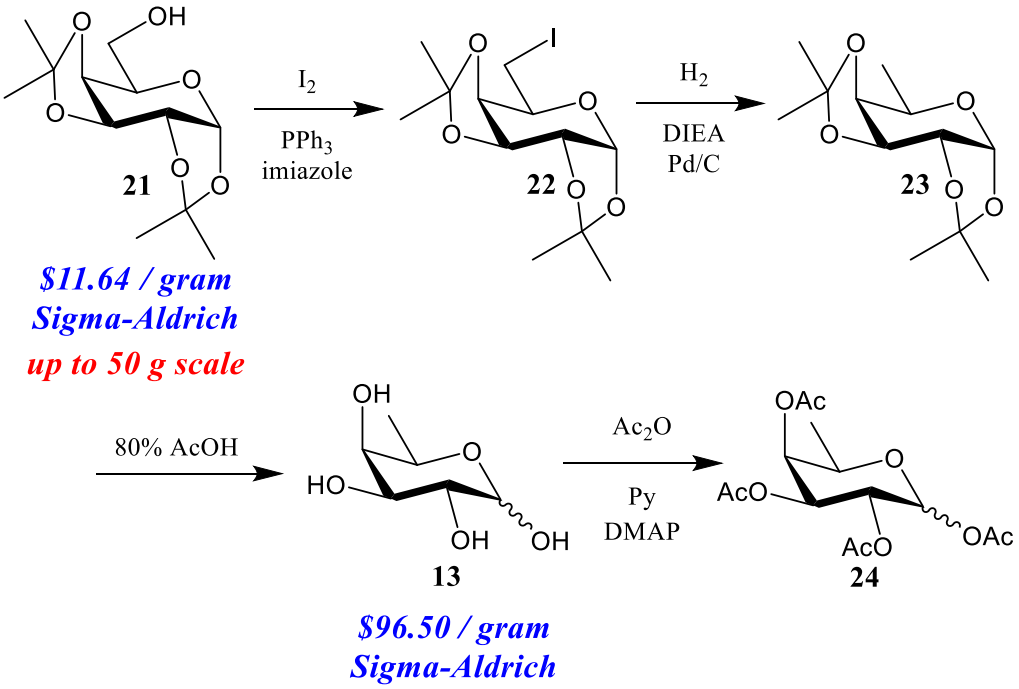


Figure 2.4. The preparation of the Fuc*p* acceptor **9** from D-Fuc*p* **13**.

2.3.1. Synthesis of D-Fucose

To prepare the Fuc*p* acceptor **9** on a multi-gram scale, 1,2:3,4-di-isopropylidene-D-gatactose (Gal) **21** was chosen as a cheaper starting material. The 1,2:3,4-di-isopropylidene-D-gatactose (Gal) **21** was significantly cheaper than D-Fuc*p* **13** ($11.64/ 1 g vs. $96.50/ 1g). The synthesis route of 1,2:3,4-di-isopropylidene-D-Gal **21** to peracetylated-D-Fuc*p* **24** is shown in Scheme 2.4. The first step was a modified Appeal reaction of the alcohol group with iodine to form **22** in 90%.[28] The by-product in this reaction was triphenylphosphine oxide and easily removed by filtration or column chromatography. Then a hydrogenation catalyzed by *N*,*N*-

diisopropylethylamine (DIEA) was be performed to remove iodine and give the *C-6* methyl group **23**. Next, the two di-isopropylidene groups were removed by heating intermediate **23** to 70°C in 80% acetic acid to give the D-Fuc*p* **13** (61% over three steps).[29] Then D-Fuc*p* **13** was globally



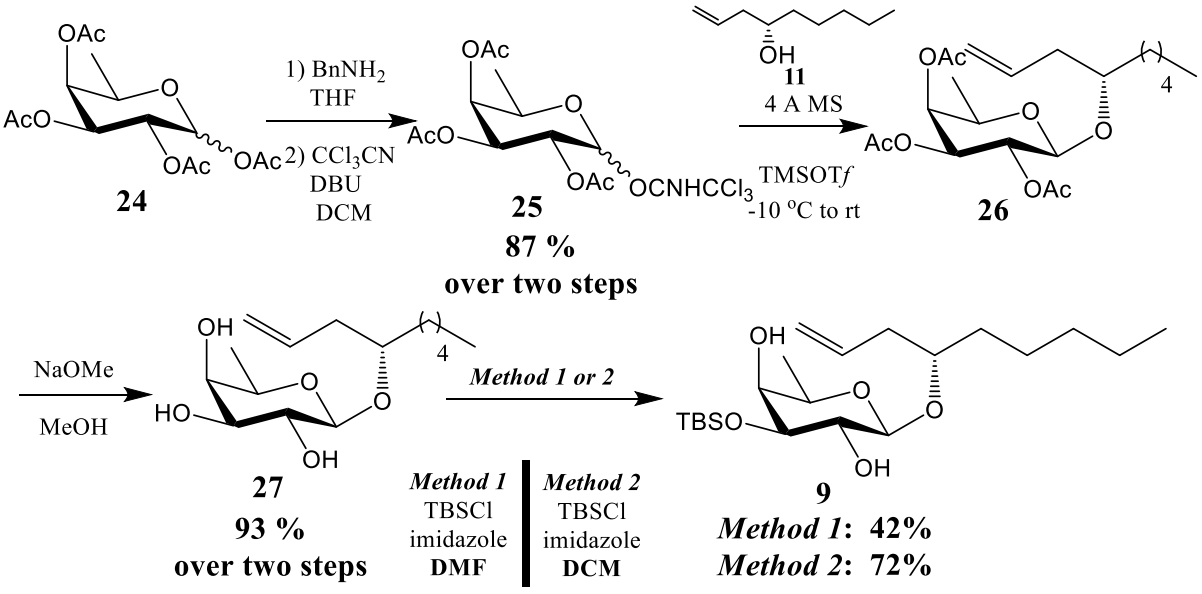
Scheme 2.4. The scalable route of from 1,2:3,4-di-isopropylidene-D-Gal **63** to D-Fuc*p* **13**.

acetylated to deliver **24** using acetic anhydride catalyzed by DMAP.[30, 31] The overall yield from 1,2:3,4-di-isopropylidene-D-galactose **21** to intermediate **24** was 51% over four steps. Scheme 2.4 did make the synthesis of Fuc*p* acceptor **9** longer (three steps) but gave a cheaper starting material for scaling-up of the Fuc*p* acceptor **9**.

2.3.2. Preparation of the Fucosyl Acceptor from D-Fucose

The route to the *β*-Fuc*p* acceptor **9** took five steps from peracetylated Fuc*p* **24** (Scheme 2.5). Scheme 2.5 shows the route followed to achieve the *C-3*-OTBS *β*- Fuc*p* acceptor **9**. The Fuc*p* donor **25** was prepared from the peracetylated Fuc*p* **66** through a de-protection of the

anomeric acetyl group with benzylamine ($BnNH_2$) and the formation of the Schmidt donor **25**.[29, 32] Then, a chemo-selective glycosylation was performed between the (4*S*)-nonenol **11** and the trichloroacetimidate donor **25** catalyzed by TMSOT*f*.[8] Only the *β*-product **26** was obtained because of neighboring group participation of the C-2-Fuc*p*. To obtain the triol intermediate **27**, the acetyl groups were de-protected with NaOMe in MeOH. The regioselective TBS-protection proved to be the most difficult transformation to prepare the *β*- Fuc*p* acceptor **9**. We knew that in the Gal configuration, the reactivity order of the secondary OH groups were 3-OH > 2-OH > 4-OH.[7, 15] So, a regioselective TBS introduction was performed on the OH-3-Fuc*p*. The first method involved the use of DMF as a solvent. Surprisingly, this method did not give a satisfactory yield (<45%) or good selectivity. Along with the two undesired regioisomers, a significant amount of the unreacted starting material **27** was also recovered (15-20%). Nevertheless, the protected *β*-Fuc*p* acceptor **9** was synthesized on a gram scale. During the scaling-up of the Fuc*p* acceptor **9**, this reaction was optimized. After modifying the different parameters (temperature, reaction time,



Scheme 2.5. Synthesis of the TBS Protected fucoside acceptor **9**.

and equivalents) the solvent was changed from DMF to DCM. This change in the solvent increased the yield (almost 30%) and selectivity of the silyl protection. However, the main advantage was the easier work-up and shorter reaction time (overnight vs. 1-2 hrs.). The regioselectivity of the silyl protection was confirmed by both ^{1}H and COSY NMR. The regioselective introduction of the TBS group to the C-3-Fuc improved the preparation of the Fuc*p* acceptor **9** over the previous synthesis of ipomoeassins.[9, 15]

2.4. Synthesis of the Glucosyl Donor

Finding a scalable route for the synthesis of the Glc*p* donor **8** proved to be a more difficult than first envisioned (Figure 2.5). Based on previous syntheses of resin glycosides, especially the ipomoeassins, a Schmidt donor provided a tried and efficient method for the construction of the (1→2)-*β*-disaccharide intermediate **7**. So, an efficient and scalable route was designed for the construction of a Glc*p* donor **8**.[9, 33-35] To design a successful, scalable synthesis route for the Schmidt donor **8** from D-Glc*p* **12** a few key criteria had to be satisfied (Figure 2.6). First, an acyl group had to be installed at C-2-Glc*p* to facilitate the formation of the (1→2)-*β*-disaccharide formation found in ipomoeassin F **1**. This is well-known as neighboring group preparation or the neighboring group effect (red). A 2-*O*-acyl group would block the *α*-face of the glucosyl donor

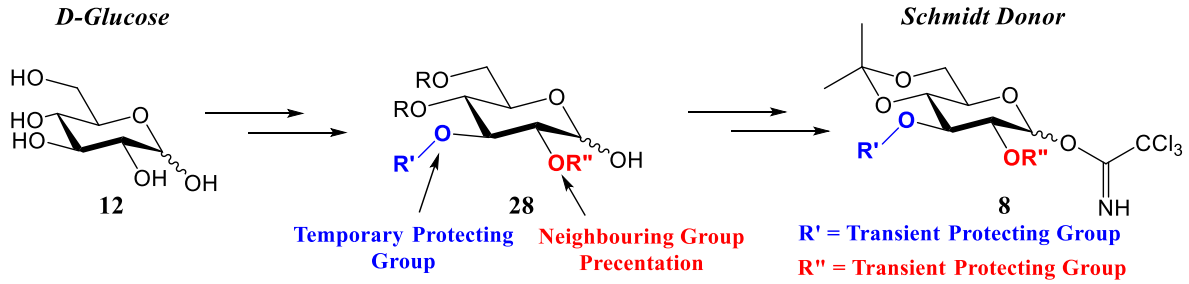


Figure 2.5. Preparation of the Glc*p* donor **8** from *D*-Glc*p* **12**.

during glycosylation, allowing the Fuc*p* acceptor **9** to attack the *β*-face. Secondly, we believed that a transient protecting group at C-3-Glc*p* could selectively be removed later in the synthesis. This would allow for a more convergent synthesis route and later-stage modifications of the C-3-Glc*p* moiety. Thirdly, another temporary group needed to be incorporated at *C-4*/*C-6* to allow the cinn moiety to be introduced late in the synthesis. We believed the best protection group for these positions was an isopropylidene acetal because it is small and can be easily removed in the presents of other functionalities (e.g. glyosidic bonds, esters, and silyl protections). For this work several different Glc*p* were investigated with multiple different anomeric protections. The work to develop a simple, effective synthesis to prepare the Glc*p* donor **8** in an efficient manner are presented in detail in the following section.

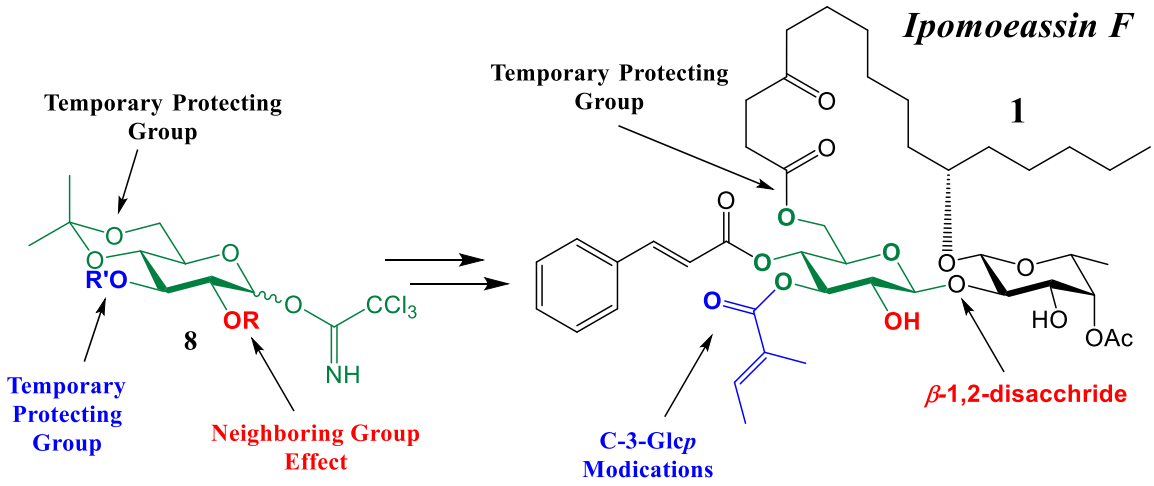
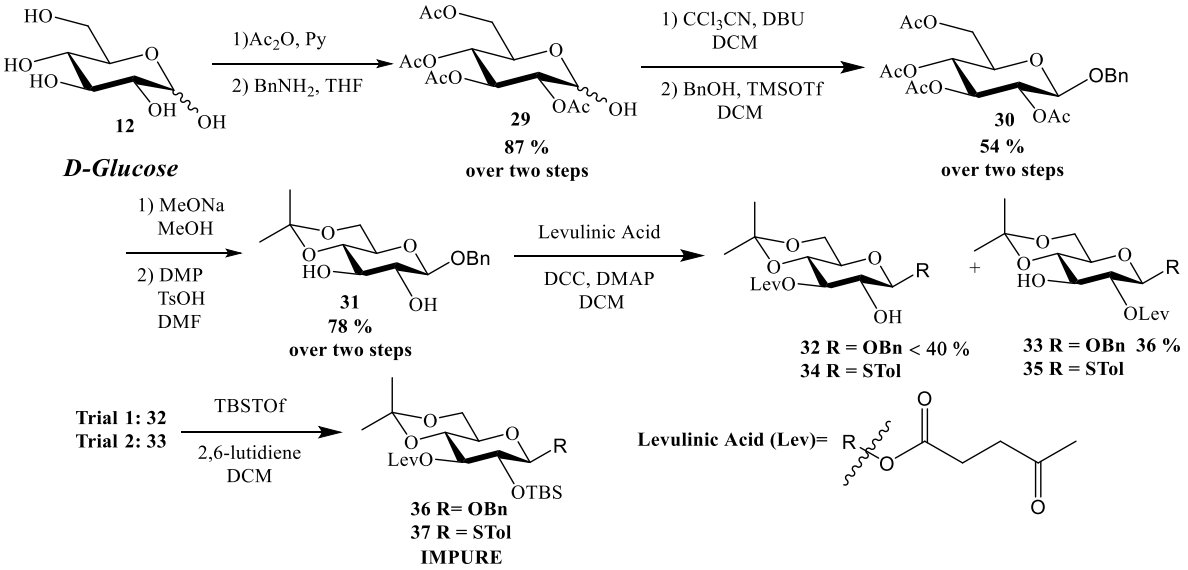


Figure 2.6. Criteria and Parameters to design an efficient and scalable synthesis route for the glucosyl donor **8** of C-3-Glc*p* ipomoeassin F **1** and analogs.

The first synthesis route to prepare the Glc*p* donor is shown in Scheme 2.6. The chemical transformations from D-Glc*p* **12** to the diol **31** proceeded as expected**.** However, the regioselective esterification to protect the HO-3-Glc*p* of the diol **31** proved to be problematic. A 1:1 ratio of the

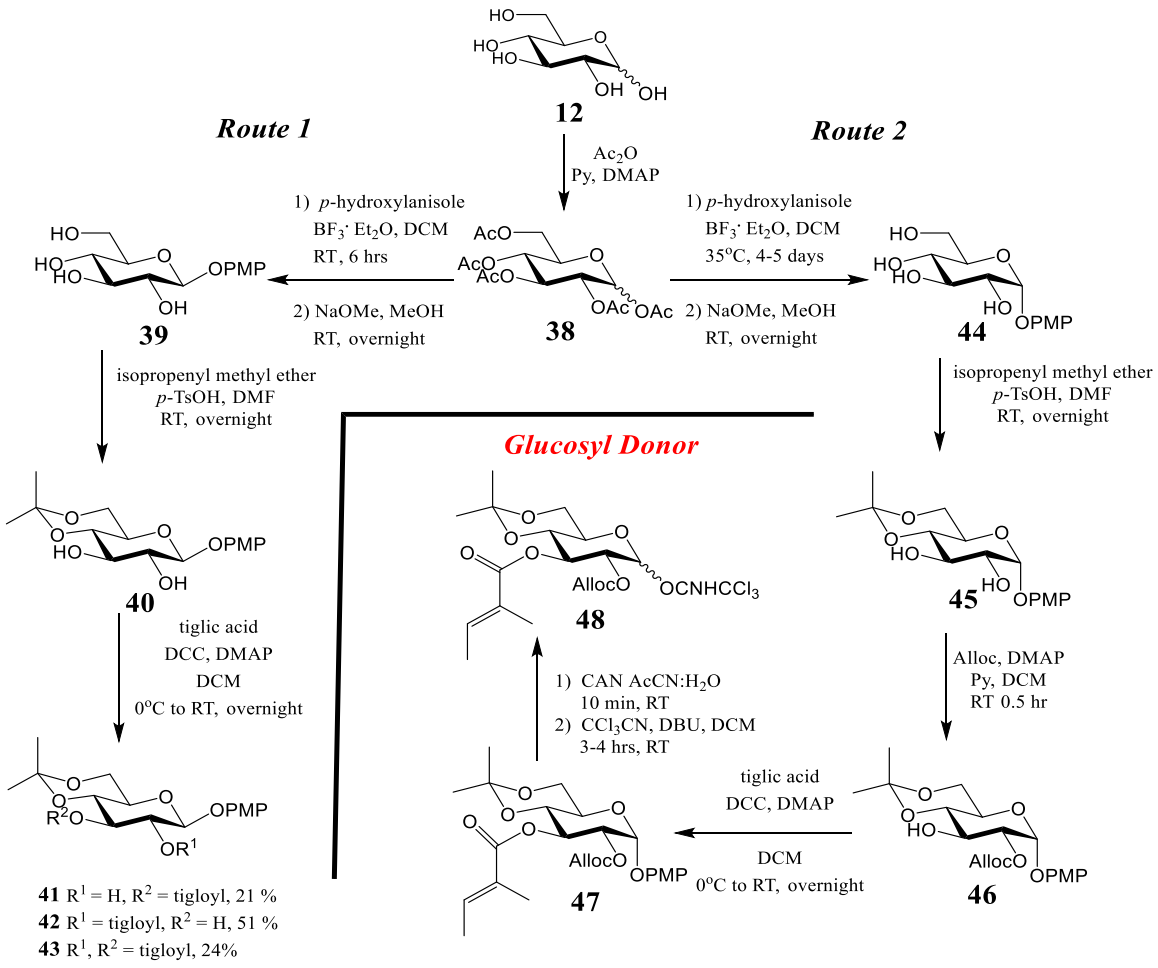
desired product **32** and the unwanted **33** was obtained. Attempts were made to go-ahead and protect 2-OH with TBSOT*f*, but a pure product **36** could not be obtained. To incorporate the lev acid through a different route, 4,6-protected *β*-D-thioGlc*p* **34** and **35** were obtained. Unfortunately, like the benzyl intermediate **32**, the by-product **34** could not be easily separated. The intermediate **33** was collected and attempts were made to protect the OH-2-Glc*p* with TBS. The TBS-protected thioGlc*p* **37** was shown to be impure after column chromatography. After multiple failed attempts to prepare the Glc*p* donor from this route, we determined the *β*-configuration would not allow for regioselective introduction to the HO-3- Glc*p*.



Scheme 2.6. Selective introduction of the lev. group for the protected glucosyl donor.

The next route opted for the allylcarbonyl (Alloc) group as a transient protecting group at C-2-Glc*p*. It was believed Alloc would be a good protecting group because of neighboring group participation required *β*-glycosylation for ipomoeassin F **1**.[36] Also, the mild protection and deprotection conditions were compatible with many functional groups.[37, 38] This route was applicable for the synthesis of ipomoeassin F **1**, the study of the C-4-Glc*p*, and aglycone modified

analogs.[7, 39, 40] The synthesis route of the Glc*p* donor is shown in Scheme 2.7. After a global peracetylation of D-Glc*p* **12**, the *p*-methoxylphenyl (PMP) group was installed at the C-1-Glc*p*. If the reaction was left for a short time (4-6 hrs.), only the PMP-*β*-Glc*p* **39** would be obtained. Whereas, if the reaction was left much longer (4-5 days), the more thermodynamically stable *α*-Glc*p* **44** would be found. Following route 1, the PMP-*β*-Glc*p* **39** deacylated with sodium methoxide followed by isopropylidenation at C-4-Glc*p*/C-6-Glc*p* (65%), to achieve the intermediate **40**. Unpredictably, the Steglich esterification did not occur exclusively at the more reactive OH-3-Glc*p* **41**.[4, 41] The major product was 2-*O*-tig **42** in 51% at a 2.4:1 ratio of 2-*O*-tig vs. 3-*O*-tig. We also attained 24% of the C-2-Glc*p* and the C-3-Glc*p* tig **43**. The non-selectivity

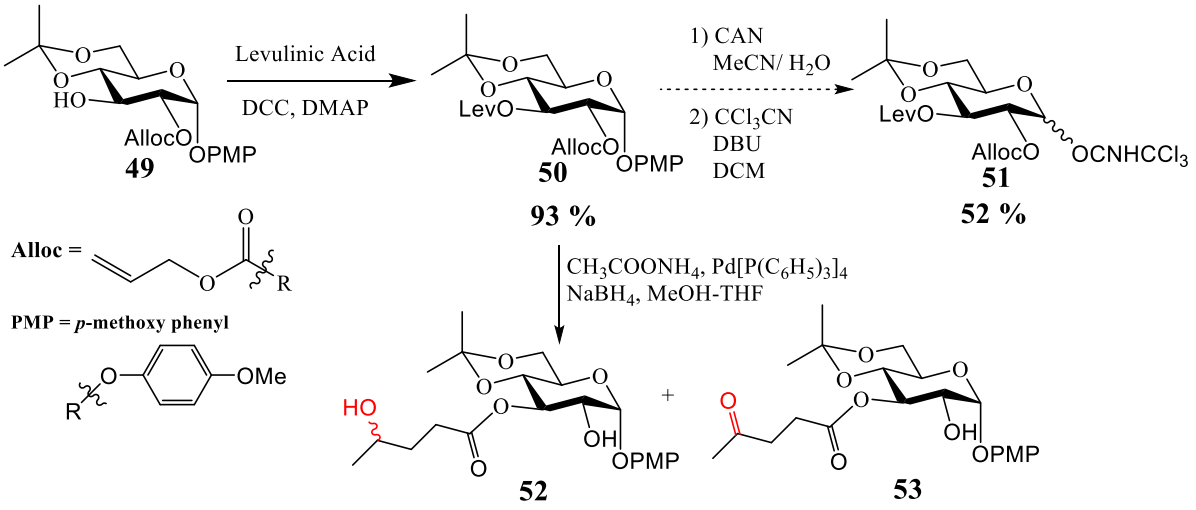


Scheme 2.7. Preparation of the Glc*p* donor **48** using Alloc at C-2-Glc*p*.

of the *β*-Glc*p* would be utilized in a later synthesis route of the Glc*p* donor. Since, it is well-known that *α*-Glc*p* favors OH-2-Glc*p* regioselective esterification, we went back to focus on the (PMB)-*α*-Glc*p* **44** (route 2).[42] After a successful isopropylidenation to protect the C-4-Glc*p*/C-6-Glc*p* **45**, a successful regioselective esterification (80%) was performed at 2-*O*-Glc*p* with AllocCl at 0^{o}C **46**. Then, the tig acid acylated the remaining hydroxyl group with a good yield to give the Glc*p* donor precursor **47** (85%). The PMP group was removed with CAN in AcCN/water to give the hemiacetal in 80 %. Finally, the Glc*p* donor **48** was obtained from the reaction of the trichloroacetonitrile catalyzed by DBU.[43] The synthesis of **48** took eight steps from D-Glc*p* **12** (15-20% yield). As stated earlier, this route was appropriate and efficient for the synthesis of ipomoeassin F **1** and the other modified analogs. However, since the C-3-Glc*p* tig was introduced at an early stage (**46**→**47**) of the overall synthesis route, this route was not applicable for the studies of the C-3-Glc*p* tig moiety. Each analog would require the synthesis of a different Glc*p* donor with a different C-3-Glc*p* modification.

The introduction of a lev group to the C-3-Glc*p* would offer a solution for the apparent drawbacks of Scheme 2.6-2.7. It was well-known that the lev groups could be orthogonally removed in the present of other acylated groups. Also, a lev protection could act as a transient protection for the C-3-Glc*p* and be deprotected later in the synthesis. Then, an esterification reaction could be performed to install modifications to the tig moiety. The OH-3-Glc*p* intermediate **49** was prepared from commercially available D-Glc*p* **13** in four steps (Scheme 2.7). However, the conditions for removing the Alloc protecting group in **50** to obtain **53** were problematic. When using sodium borohydride ($NaBH_4$), the product was found to be **53**, not **52**, where the ketone functionality in the lev moiety was reduced along with the removal of Alloc.

Looking back, the side reaction should have been expected based on reactivity of the $NaBH_4$ and ketones.



Scheme 2.8. Attempted preparation of Glc*p* donor **51** using Alloc at C-2-Glc*p*.

After learning the drawbacks with some of the previous routes, we started to think about using the same transient protection at both the C-2-Glc*p* and C-3-Glc*p* (Figure 2.7). If both positions were protected with the same protection, they could be orthogonally de-protected later in synthesis to give the *C-4* ketone macrocycle diol intermediate **55**. Then, a regioselective esterification was envisioned to modify the C-3-Glc*p* **56** in the late stage of the synthesis. This late stage modification would allow for a more convergent synthesis route than previously considered. Also, this route would allow more time to be spent on synthetizing tig modifications and not preparing complex disaccharide intermediates. Lev groups **57** were finally chosen because they could be installed during the preparation of the Gluc*p* donor synthesis and potentially a late-stage deprotection could be performed. Hydrazine acetate had been shown to orthogonally de-protect lev group **54** in the presents of other esters and glyosidic bonds.[38, 44, 45] Another benefit

was the C-2-Glc*p* lev group could help form the (1→2)-*β*-disaccharide needed in the natural product.

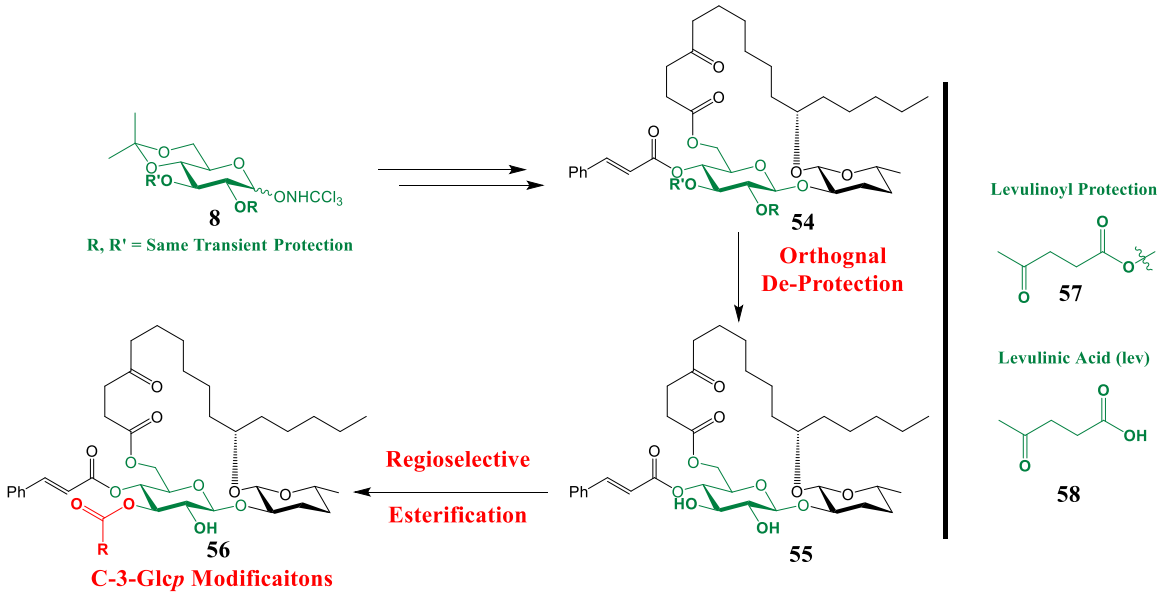
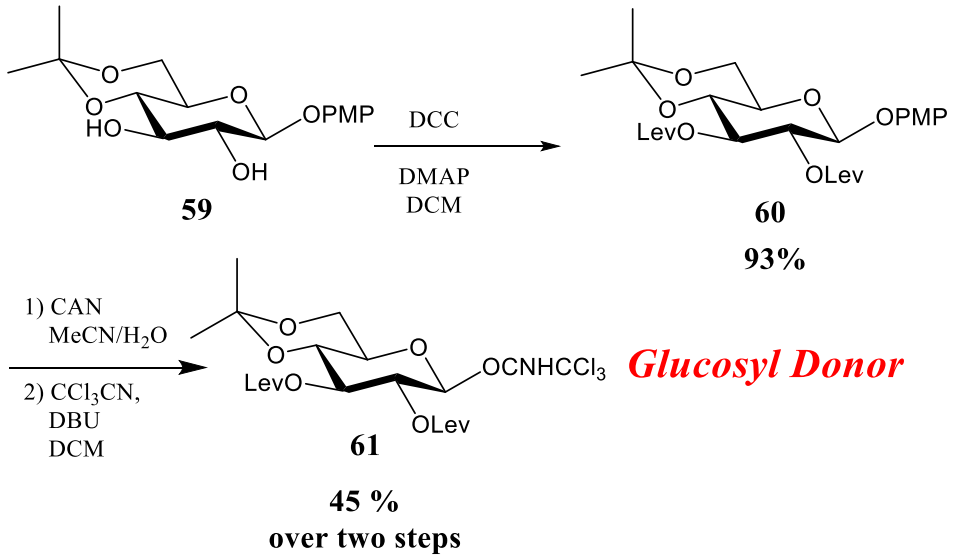


Figure 2.7. Reasoning for using the same transient protection at C-2/C-3-Glc*p*.

Scheme 2.9 shows the first synthesis route developed with C-2 and C-3-Glc*p* lev groups. The diol intermediate **59** was prepared from the D-Glc*p* **12** in four steps by following the steps in Scheme 2.7.[7] **59** was then utilized to introduce the lev groups to the *C-2* and *C-3* positions though Steglich esterification to give **60**. The purification of this intermediate was difficult because of the dicyclohexylurea (DCU) bi-product. The anomeric *p*-methoxyphenyl group (PMP) was removed with CAN in AcCN/H_2O to give the hemiacetal and the trichloroacetimidate group installed to furnish the Schmidt donor **61**. However, the yield was only 45% for these two steps. This low yield was unexpected, because published data showed that these reactions proceeded smoothly (74% **60**→**61**). The low yield for this transformation can possibly be explained by a few reasons. During the work-up of the CAN reaction an emulsion was formed between the aqueous and DCM layers, causing difficulty in obtaining the hemiacetal. Secondly, these deprotection conditions



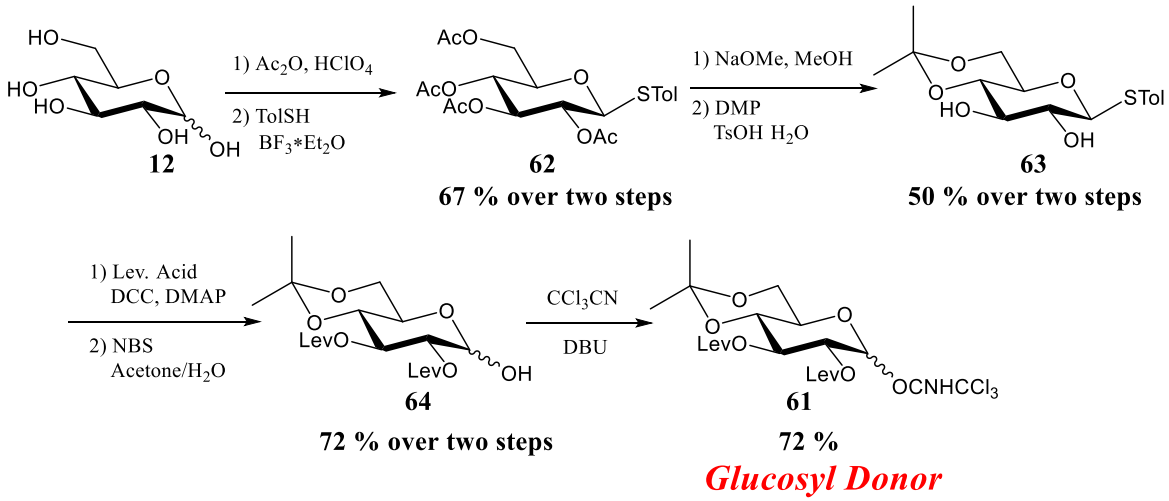
Scheme 2.9. Synthesis of the Glc*p* donor **61** with C-2 and C-3 lev protecting groups.

have been shown to be very sensitive to acid-labile groups, like the C-4/C-6 isopropylidene group. If the reaction was left too long, the PMP and the C-4/C-6 isopropylidene group would be removed. The third reason involved the in-stability of the Schmidt donor. It is well-know that Schmidt donors are un-stable when exposed to acid or moisture, this causes the trichloroacetimidate donors to decompose back to the hemiacetal.[46] Because of the number of steps (eight steps **13**→**61**) and the overall yield, especially in the final two steps, another synthesis had to be developed to efficiently prepare the Glc*p* donor. The experience from this synthesis route helped us gain the knowledge that lev groups at the *C-2* and *C-3* would be good transient protecting groups.

Knowing the disadvantages of the previous routes, a new synthesis route was designed to achieve a new Glc*p* donor. For this method a thioglycoside was utilized, instead of the PMP group used in previous routes (Scheme 2.7-2.9). Anomeric thioglycoside protections have been routinely utilized in carbohydrate synthesis because of their simple preparation and good stability.[1, 44, 45, 47]

They have been very useful for a wide range of glycosylation reactions though activating thiophilic agents like NIS-TMSOTF, NIS-AgTOF, MeOT*f*, DMTST, among others.[31, 32] However, we chose to continue to make a trichloroacetimidate donor for the glycosylation reaction. Literature has shown for the synthesis of the complex glycosides, thiophilic promoters can initiate unwanted side reactions in a multifunctional glycone like ipomoeassin F **1**.[1]

The synthesis route to prepare the Glc*p* donor **61** was achieved from D-Glc*p* **12** in seven steps (Scheme 2.10). The synthesis route offered several benefits compared to previous routes, including decreased reaction times and the number of steps. For this route the D-Glc*p* **12** was globally acetylated with Ac_2O catalyzed by perchloric acid ($HClO_4$) instead of Ac_2O and pyridine. This acylation method decreased the reaction time from overnight to 30 mins and allowed for almost exclusively the *β*-peracetylated Glc*p*. The conversion to the thioGlc*p* **62** did not go to completion giving only a 72% yield. The formation of the *β*-thioGlc*p* **62** was very dependent on the quality of the boron trifluoride diethyletherate ($BF_3{\cdot}OEt_2$). Sometimes a higher ratio of the $BF_3{\cdot}OEt_2$ would still not push the reaction forward, making the purification of the thioGlc*p* **62** more tedious. In the next two steps, the acetyl groups were removed by sodium methoxide, then

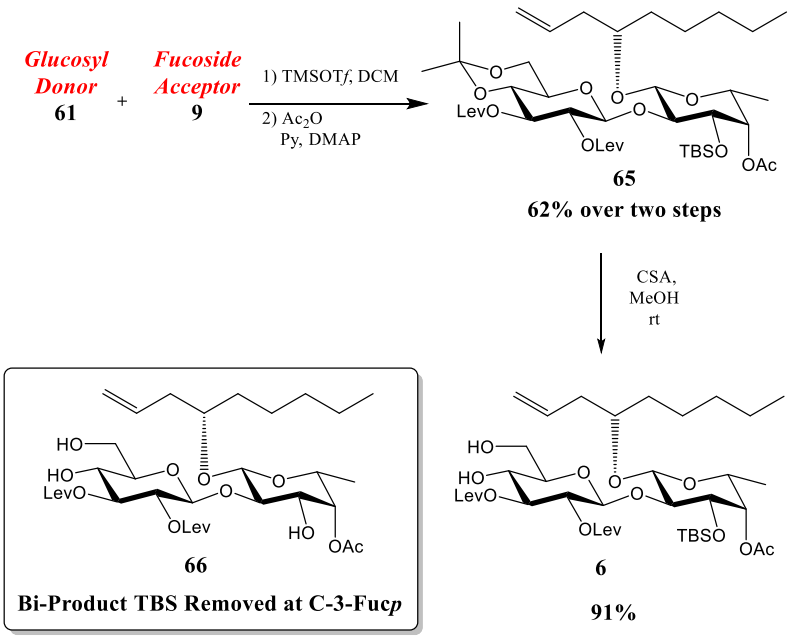


Scheme 2.10. Preparation of the Glc*p* donor **61** using thiotoluene protection.

the C-4/C-6 were protected using dimethoxypropane (DMP) to form the isopropylidene acetal **63**.[48] Then, the lev groups could be easily installed to the *C-2* and *C-3* positions through Steglich esterification. Similar to intermediate **60**, the purification was a difficult because of the DCU by-product.[49] This problem was solved by removing some of the DCM under reduced pressure and adding 2:1 (Hex:Et_2O). This caused most of the DCU to become insoluble and the solution could be filtered through Celite. Then, any remaining DCU was removed by column chromatography. Another benefit of Scheme 2.10 was observed in the removal of the thiotolyl group at the anomeric position. This route allowed for the orthogonal removal of the thiotolyl protection with NBS in acetone/H_2O to afford the hemiacetal **64**.[50, 51] This was a major benefit over previous syntheses of the Glc*p* donor, because CAN was needed to remove anomeric PMB. The deprotection of the thiotolyl group was time and temperature dependent. If the reaction was left longer than 15-20 mins or the temperature rose above -5^{o}C, the isopropylidene acetal would start to be removed. Finally, the trichloroacetimidate group was installed to furnish the Schmidt donor **61**. This synthesis route took seven steps and offered numerous benefits over previous route for the preparation of the Glc*p* donor **61**. The overall efficiency and the number of steps were the primary reasons why Scheme 2.10 eventually was chosen to scale up the Glc*p* donor **61** for the study of the tig moiety of ipomoeassin F **1**.

2.5. Glycosylation of the Two Monosaccharides and Regio-selective Esterification

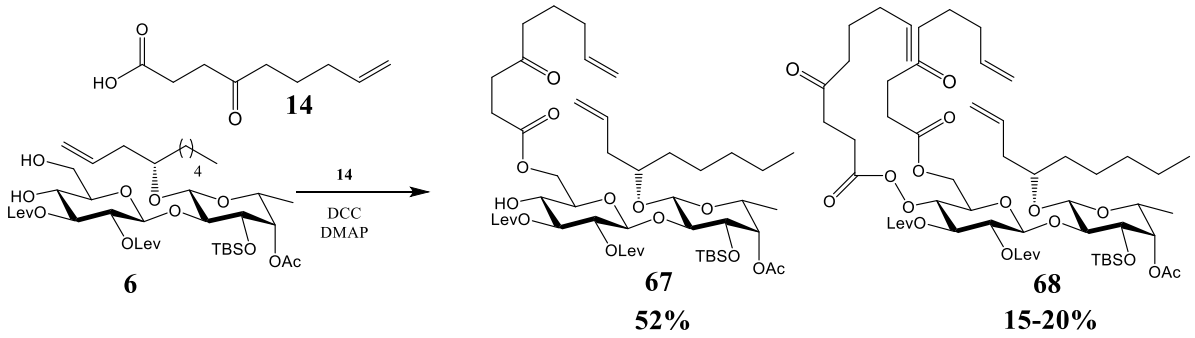
With both the Glcp donor **61** and the Fucp acceptor **9** in hand, a Schmidt regio-selective glycosylation reaction was performed with a 1.2:1 ratio of **61** to **9** (Scheme 2.11).[33, 34] The (1→2)-*β*-disaccharide was obtained for a few specific reasons. First, the bulky-TBS group at the C-3-



Scheme 2.11. Glycosylation of the Glc*p* donor **61** and Fuc*p* acceptor **9**, acylation at C-4-Fuc*p*, and removal of isopropylidene acetal to give the diol **6**.

Fuc*p* helped to block the OH-4-Fuc*p* causing the glycosylation to occur preferentially at the OH-2-Fuc*p*. Second, the lev group at C-2-Glc*p* blocked the α-face of the Schmidt donor **61** resulting in only the β-glycosylation product. Next, an acetylation was performed to the remaining hydroxyl at the C-4-Fuc*p* to give **65**. After the acetyl group was installed, the isopropylidene acetal was removed using camphor-10-sulfonic acid (CSA). This de-protection was time dependent, giving the diol **6** after a few hours at RT. However, if the reaction was allowed stir longer or the CSA was not properly quenched with NEt_3 the acidic conditions would cause the -OTBS at C-3-Fucp to be removed giving **66**. The (1→2)-β-disaccharide diol **6** was a key intermediate in the synthesis of ipomoeassin F analogs because modifications to the left-tether of the aglycone can be easily coupled to the 6-OH-Glc*p*. Next, another trans-esterification reaction was performed with the (1→2)-β-disaccharide **6** and 4-oxo-8-nonenic acid **14** to form the RCM precursor **67** (Scheme

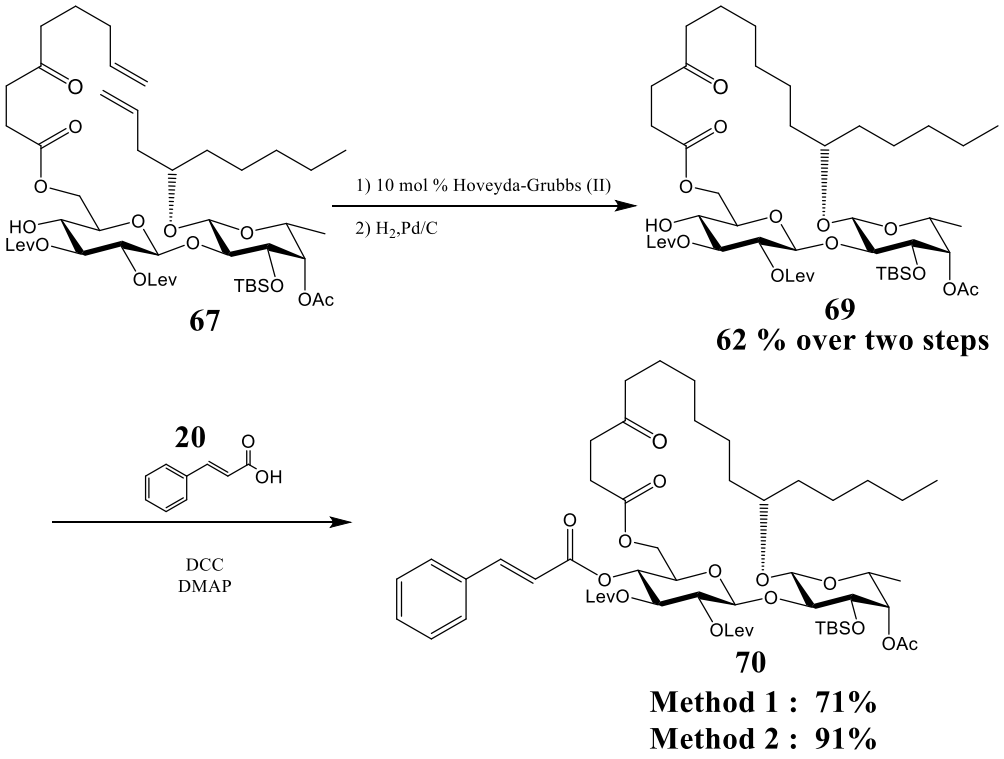
2.12). The greater nucleophilicity of the primary OH-6-Glc*p* over the secondary OH-4-Glc*p* was utilized for the chemo-selective esterification. A significant amount (15-20%) of the di-substituted product **68** was often observed. This drawback could be limited by the controlling the temperature and the equivalents of the 4-oxo-8-nonenoic acid **14** (1.05-1.2 eq).



Scheme 2.12. Coupling of 4-oxo-8-nonenoic acid **14** and the (1→2)-*β*-disaccharide **6** using Steglich esterification conditions.

2.6. Macrocycle Formation and Cinnamoyl Introduction

Because of its favorable reactivity profile and high flexibility, a RCM reaction was believed to be the best method to form the macrolactone ring (Scheme 2.13).[52, 53] RCM was adopted by both Postema[9] and Fűrstner[15] for the formation of the 20-membered macrolactone ring of the ipomoeassins. From the diene **67**, an RCM reaction was performed using Hoveyda-Grubbs catalyst (II) (10 mol%) to yield the E/Z mixture.[54, 55] Dilute conditions were very important for the RCM reaction to prevent the polymerization of the diene **67**. Then hydrogenation with Pd/C in EtOH was performed on the E/Z double bond mixture to afford the saturated macrolactone **69** in 62% over two steps. These hydrogenation conditions were an improvement over previous synthesis where Postema [9]and Fűrstner[15] could not use strong hydrogenation conditions due to



Scheme 2.13. Preparing the macrocycle **69** and coupling of the cinn moiety to make **70**.

incompatibility of the two *α,β*-unsaturated esters (cinn, C-4-Glc*p* and tig, C-3-Glc*p*). The cinn moiety was introduced thorough an esterification reaction with HO-4-Glc*p* through Steglich esterification conditions. A late-stage introduction of the cinn moiety gave the opportunity to perform systematic SAR studies on the C-4-Glc*p* by modifying cinn moiety and then performing final chemical transformations.

2.7. Protection of the *C-4* Ketone of the Aglycone

After installation of the cinn moiety at C-4-Glc*p* **70**, one transformation was needed to obtain the *C-4*-oxo macrocycle diol intermediate **71**, critical for the synthesis of C-3-Glc*p* modified ipomoeassin F analogs (Figure 2.8). Based on the designed synthesis route, we envisioned using hydrazine acetate ($NH_2NH_2{\cdot}AcOH$) to orthogonally remove the C-2/C-3 lev protecting groups to

yield **71**. However, the intermediate **72** was obtained where the lev groups had been deprotected to the hydroxyls (red), but the *C-4* ketone had been converted into hydrazone (blue). Multiple attempts were made to hydrolyzes the hydrazone **72** back to the ketone **71**, but the stability of the hydrazone prevented this transformation from occurring.

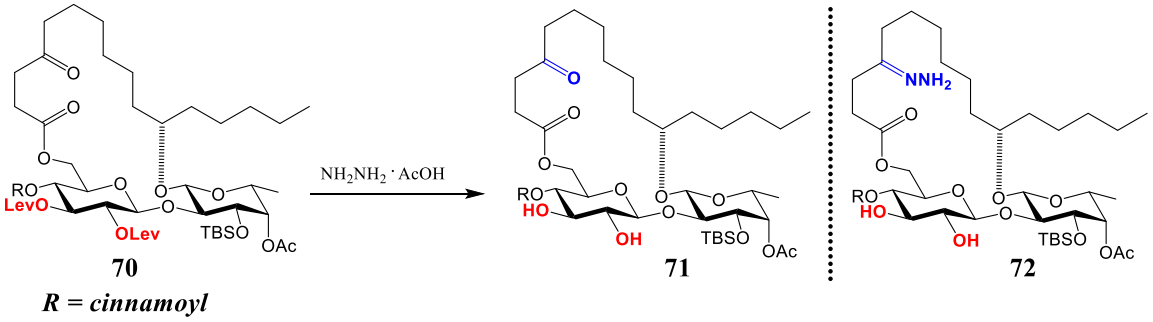
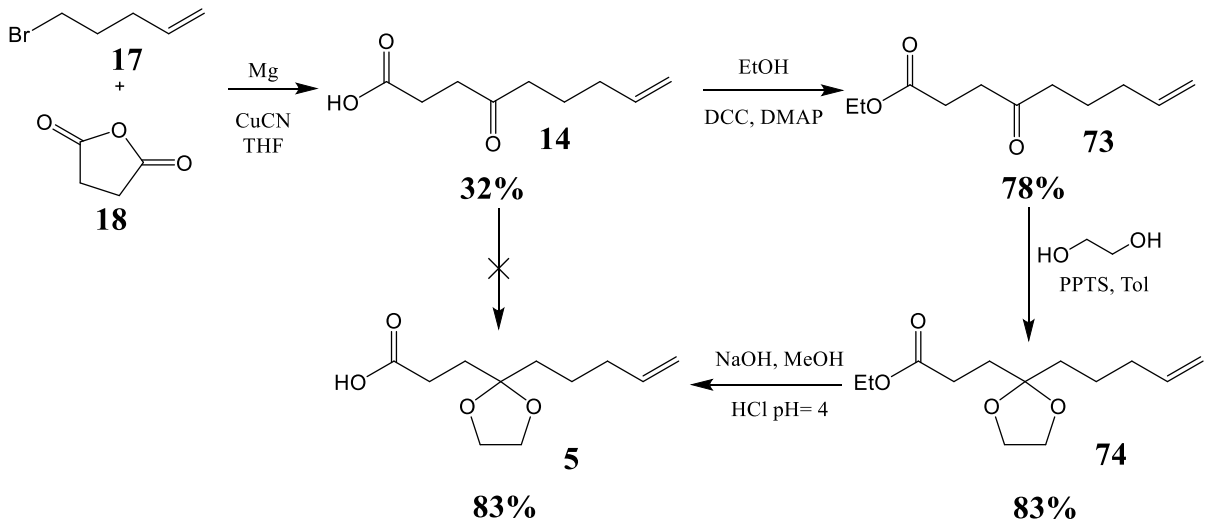


Figure 2.8. Removal of the C-2/C-3-Glc*p* lev groups in presents of *C-4* ketone.

At this point in the synthesis, a decision had to be made about how to obtain the *C-4* ketone macrocycle diol **71**. After considering different possibilities that could be explored, we determined the two best options would be to prepare a different Glc*p* donor **61** or protect the *C-4* ketone of aglycone. Finally, we decided the best option would be to protect the *C-4* ketone for a few reasons. Preparing a different Glc*p* donor **61** was justifiable, but this meant going back to the beginning of the synthesis and spending a significant amount of time exploring different routes. Since, the current Glc*p* donor **61** had already been synthesized on a multi-gram scale. We did not want spend time to develop another Glc*p* donor route. Protection the *C-4* ketone did make the synthesis route a few steps longer but provided an easier alterative than developing another Glc*p* donor synthesis route.

Since the *C-4* ketone to be protected, a synthesis route was developed prepare the 4-acetal-8-nonenoic acid **5** from the 4-oxo-8-nonenoic acid **14** (Scheme 2.14). First, a direct protection of

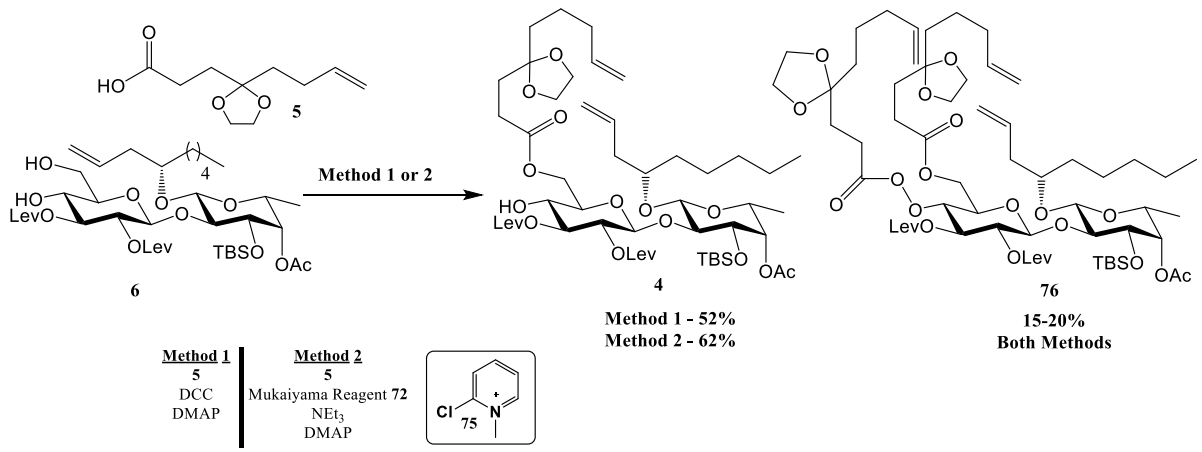
the **14** 4-oxo-8-nonenoic acid to protect the *C-4* ketone was attempted. The direction protection gave a mixture of the desired product **5** and an unwanted ethoxy ester. After multiple reaction parameters (solvent, temperature, and equivalents of the ethylene glycol) were modified without obtaining purely 4-acetal-8-nonenoic acid **5**, we determined a step-wise protection must we performed. So, the 4-oxo-noenoic acid **14** was converted to an ethyl ester **73** by ethanol (78%) using Steglich conditions.[56, 57] Then the *C-4* ketone was protected by forming the acetal **74** with ethylene glycol catalyzed by pyridinium *p*-toluenesulfonate (PPTS) (83%). The R_f values of ketone **73** and acetal **74** were the same (R_f = 0.55), making the transformation difficult to monitor by TLC. When the reaction was left overnight, and the excess of the ethylene glycol was added to the reaction, the transformation easily went to completion. Lastly, the ethyl 4-acetal-8-nonenoate **74** was treated with NaOH in MeOH to form the 4-acetal-8-nonenoic acid **5**. The yield of the four transformations was only 17-30%. Following Scheme 2.14 did make the synthesis route of the left-tether three steps longer but give a scalable method to protect the *C-4* ketone.



Scheme 2.14. The synthesis of the 4-acetal-8-nonenoic acid **5** from 4-oxo-8-nonenoic acid **14**.

2.8. Optimized RCM Precursor and Macrocycle Formation

Originally, a Steglich esterification (Method 1) was performed to couple the diol **6** and the acid **5** (Scheme 2.15). However, the Mukaiyama esterification (Method 2) offered a few advantages over the Steglich esterification (Method 1).[58] One advantage was found in the reaction time of the trans-esterification. The Steglich esterification took 12 hrs. to completely convert the acid **5** and diol **6** into the ester **4**, whereas the Mukaiyama method took only one-two hrs. to complete. This helped improve regioselectivity for the reaction from **6** → **4**, caused a small decrease in the percentage of C-4-Glc*p* ester **76** isolated. The biggest advantage was found in the purification of the diene intermediate **4**. A major drawback in the Steglich method was the formation of DCU **80** as a by-product from the DCC **81** (Figure 2.9). The DCU was difficult to remove with column chromatography, often, taking more than one purification to obtain pure diene **4**. The Mukaiyama conditions made the purification of multiple esterification reactions easier; this esterification method was utilized for future esterification reactions.



Scheme 2.15. Coupling of 4-acetal-8-nonenoic acid **5** and the (1→2)-*β*-disaccharide **6** using Steglich vs. Mukaiyama esterification conditions.

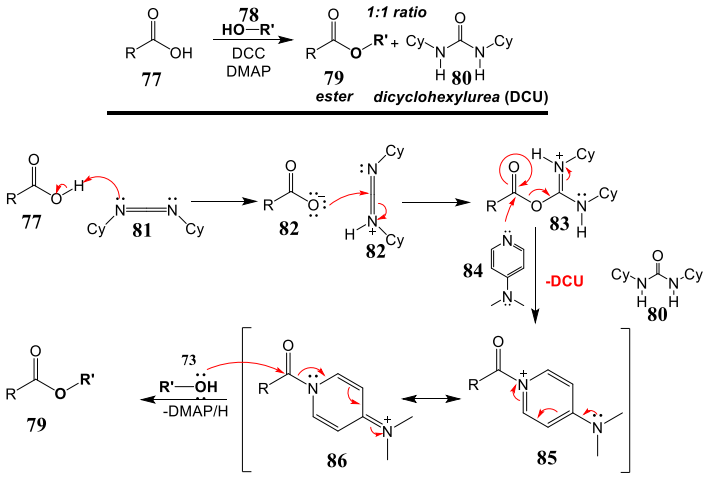
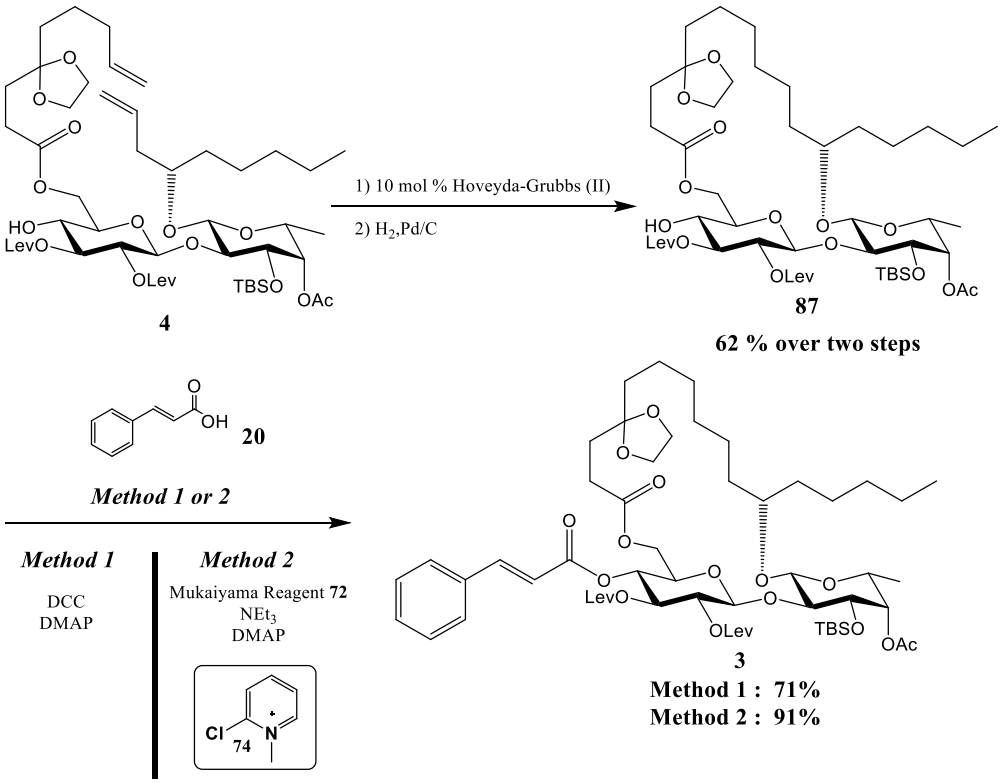


Figure 2.9. General Steglich esterification mechanism 1:1 ratio of Product:DCU.

The macrocycle **87** was formed through a RCM/hydrogenation reaction as shown Scheme 2.16. Then, the cinn moiety **20** was introduced thorough an esterification reaction with the one remaining hydroxyl group (C-4-Glc*p*). Originally, Steglich esterification method was utilized to

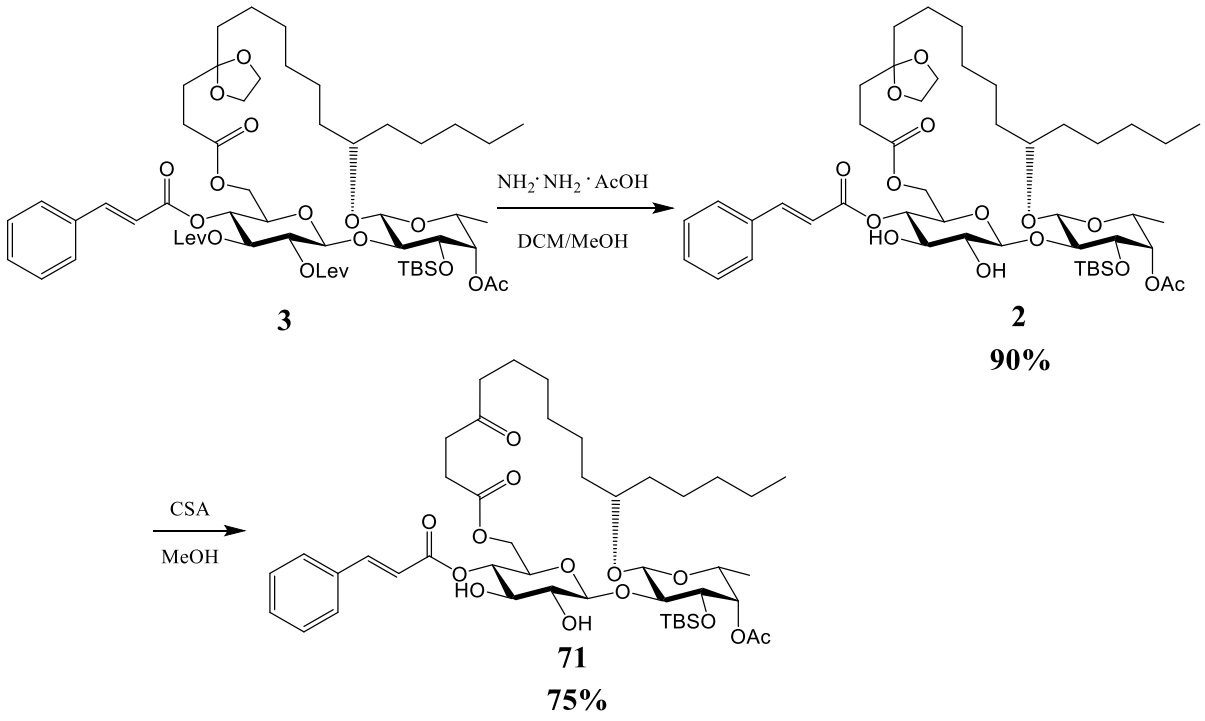


Scheme 2.16. Preparing the macrocycle **84** and coupling of the cinn moiety to construct **3**.

install the cinn moiety. However, during the scaling-up process we chose to use Mukaiyama esterification conditions because of simpler work-up and purification (Figure 2.9).

2.9. Completion of the Key C-2/C-3 Glucosyl Diol Intermediate

After installation of the cinn moiety at C-4-Glc*p* **3**, two transformations were needed to obtain the diol intermediate **71** which was critical for the synthesis of C-3-Glc*p* modified ipomoeassin F analogs (Scheme 2.17). The *C-4* acetate protection allowed the lev groups on the C-2/C-3-Glc*p* moieties to be easily removed with $NH_2NH_2{\cdot}AcOH$ to yield **2** (90 %). The orthogonal de-protection seemed to work best with a DCM–MeOH (9:1) solvent mixture. The *C-4* acetal **2** could be removed by CSA to give the key diol intermediate **71**. This reaction proved to be more problematic than anticipated, because no change could be observed in the R_f values on the

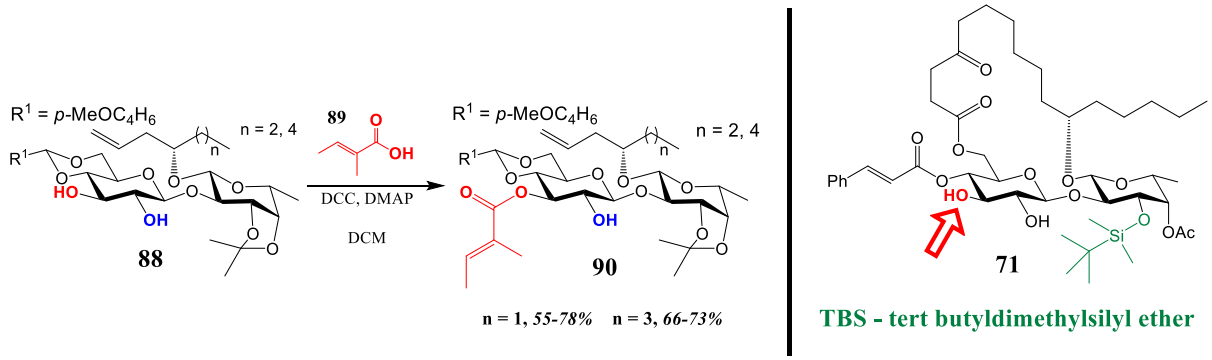


Scheme 2.17. Preparation of key C-2/C-3-Glc*p* diol intermediate **71** from macrocycle **3**.

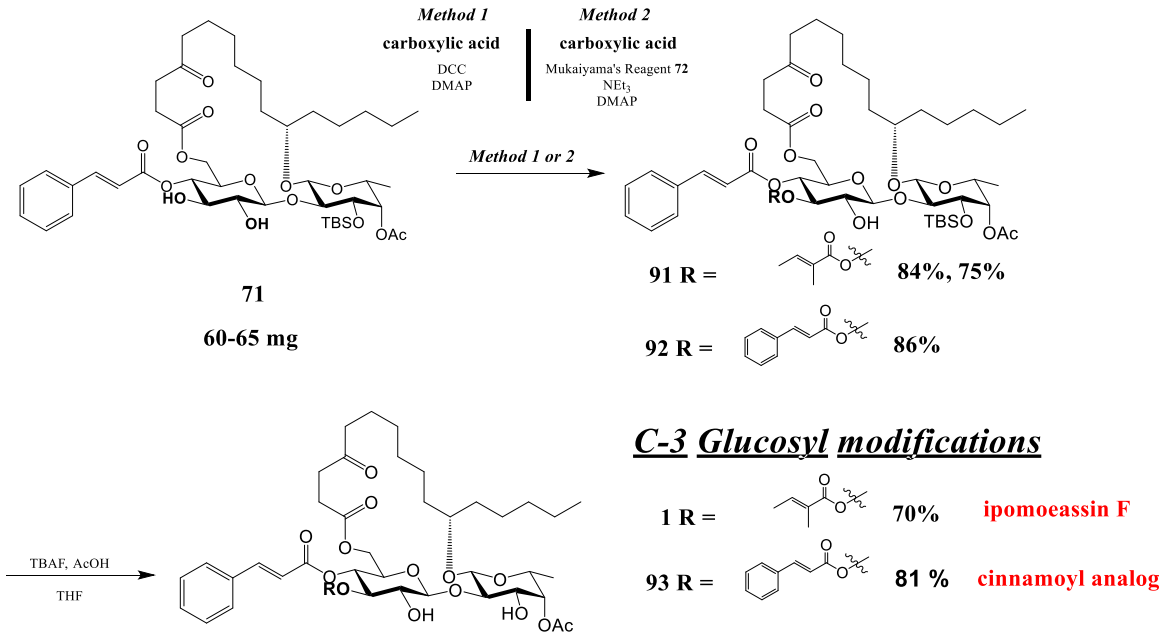
TLC. After multiple trials, the reaction proceeded smoothly with 0.2 eq of CSA and solvent grade MeOH. The transformation was time dependent if the reaction was left too long, the -OTBS protecting group at C-3-Fuc*p* would be removed giving the triol. A trace amount of H_2O was critical for the transformation from **2** to **71**. The creation of simple, straight-forward, and reproducible conditions to achieve the diol intermediate **71** was vital to the overall project.

2.10. Ipomoeassin F and C-3-Glucosyl Modified Analogs

Having completed the important diol intermediate **71**, we wanted to determine if the regioselective esterification to HO-3-Glc*p* would occur as anticipated. There were two main reasons we believe esterification would occur regioselectivity at the HO-3-Glc*p*. In the Fürstner synthesis, they performed a regioselective esterificaiton for tiglic acid (9:1 ratio in favor of 3-OH-Glc*p*), however the Steglich esterification only included a non-cyclic1,2-*β*-disaccharide **88** and tig. acid **89** as the coupling partners (Scheme 2.18). Another reason we believed the esterification could occur regio-selectively is because the -OTBS group at the C-3-Fuc*p* in **71**. Based on preliminary studies, the silyl protecting group would block the OH-2-Glc*p*, causing the esterification to occur almost exclusively at the C-3-Glc*p*. In the final step, -OTBS at C-3-Fuc*p* could be removed with the mixture of tetra-*n*-butylammonium fluoride and AcOH (TBAF/AcOH)



Scheme 2.18. Reasoning for the the regioselective esterification to the HO-3-Glc*p* of ipomoeassin F **1**.



Scheme 2.19. Completion of ipomoeassin F **1** and C-3-Glc*p* cinn acid analog **93**.

to give the analogs. To confirm the hypothesis and literature supported reasonings work began to syntheses ipomoeassin F **1** and C-3-Glc*p* modified analogs (Scheme 2.19). The regioselective esterification of **71** proceeded as expected yielding the 3-*O*-tigloyl intermediate **91** and the 3-*O*-cinn intermediate **92** in high yield. The conformation of the esterification was established by ^{1}H, ^{13}C, COSY and HMBC correlations shown in Figure 2.10. In the final step, the silyl ether was deprotected with tetra-*n*-butylammonium fluoride (TBAF) and AcOH to yield ipomoeassin F **1**. The cinn analog **93** was obtained through the same de-protection conditions without difficulty.

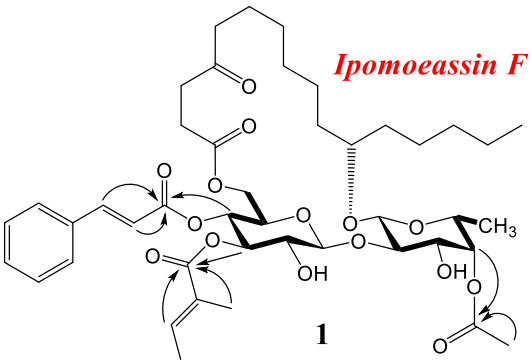


Figure 2.10. Key COSY (Bold) and HMBC (arrows) correlations for ipomoeassin F **1**.

2.10.1. Cytotoxicity Testing of the C-3-Glucosyl Modified Ipomoeassin F. Analogs

In-house cytotoxicity evaluations were performed on ipomoeassin F **1**, the cinn analog **93**, and two other analogs **94-95** (prepared by another lab member) on two human breast cancer cell lines (MDA-MB-231 and MCF7). Table 2.1 shows the concentrations required for 50% cell death (IC_{50} values) compared to the vehicle-treated negative control. Unfortunately, when compared to ipomoeassin F **1**, all three of the analogs displayed significantly decreased potency. The IC_{50} for the cinn analog **93** declined by 161 and 150-fold against MCF7 and MDA-MB-231, respectively. While C-4-Glc*p* analogs **94-95** lost almost all bioactivity, further emphasizing previous studies that showed the cinn moiety is vital to the bio-activity ipomoeassin F **1**. These results proposed the combination of an aliphatic *α,β*-unsaturated ester at C-3-Glcp and the aromatic *α,β*-unsaturated ester at C-4-Glcp provide a synergistic contribution for ipomoeassin F **1** cytotoxicity.

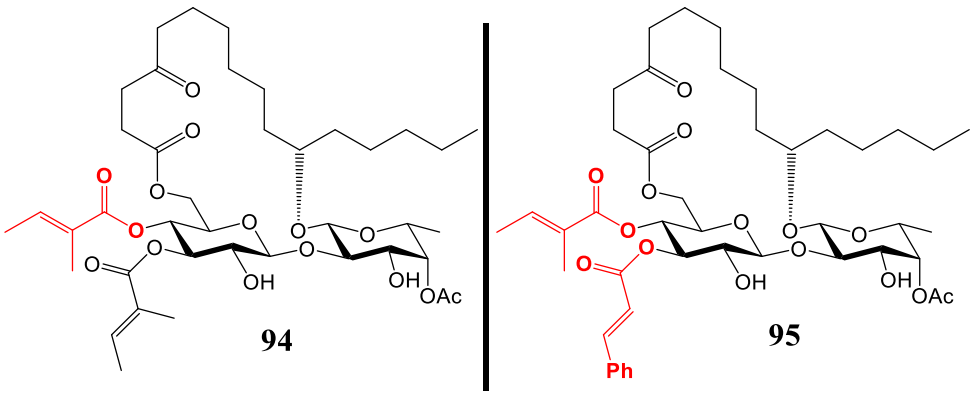


Table 2.1. Cytotoxicity (IC_{50}, *μ*M) of Ipomo. F and C-3-Glc*p* Analogs

	MDA-MB-231[a]	MCF-7[a]
Ipomoeassin F 1	0.014	0.070
93	2.1	11.3
94[b]	17.0	>25
95[b]	>2.5	-[c]

[a] The data were obtained from at least two independent experiments, and standard errors are within 20%. [b] Prepared by another lab member. [c] Not tested.

2.11. C-3-Glucosyl Acylate Ipomoeassin F Analogs

Knowing regioselective esterification of the diol **71** proceeded as expected, work began on a small library of analogs to study the importance of the tig moiety to the cytotoxicity of ipomoeassin F. The C-3-Glc*p* position was chosen to make modifications, because this position has been previously shown by Shi *et al.* to be vital to the activity of ipomoeassin F **1**.[40] From the same study, the *α,β*-unsaturated esters were determined to be vital to the bioactive of ipomoeassin F **1**. When the *α,β*-unsaturated ester was saturated, a loss of 100-150 fold was observed for multiple cell lines. So, we knew that any C-3-Glc*p* analogs needed to contain an *α,β*- unsaturated ester. However, the role and importance of the *α,β*-methyl groups had not yet been determined. Before any further C-3-Glc*p* modification or analogs were synthesized, the role and importance of the *α,β*-methyl groups needed to be determined. Figure 2.11 shows the small library of C-3-Glc*p* analogs **96-98** studied through this work with the modifications displayed in red. For the first analog **96**, the tig moiety is replaced by crotonic acid to investigate the *α*-methyl substituent. The

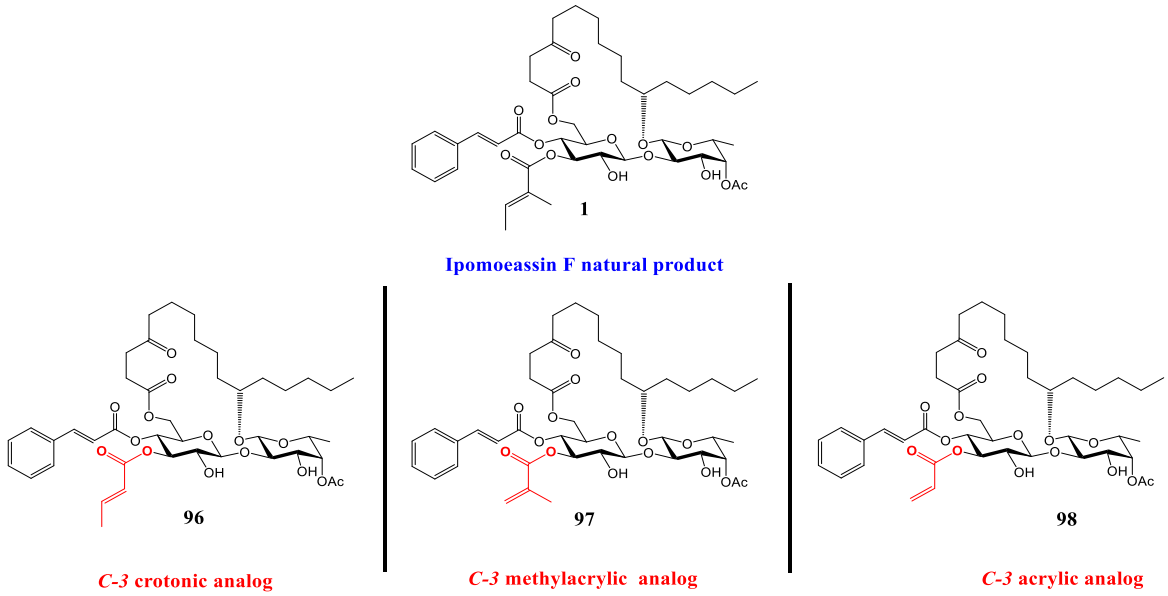
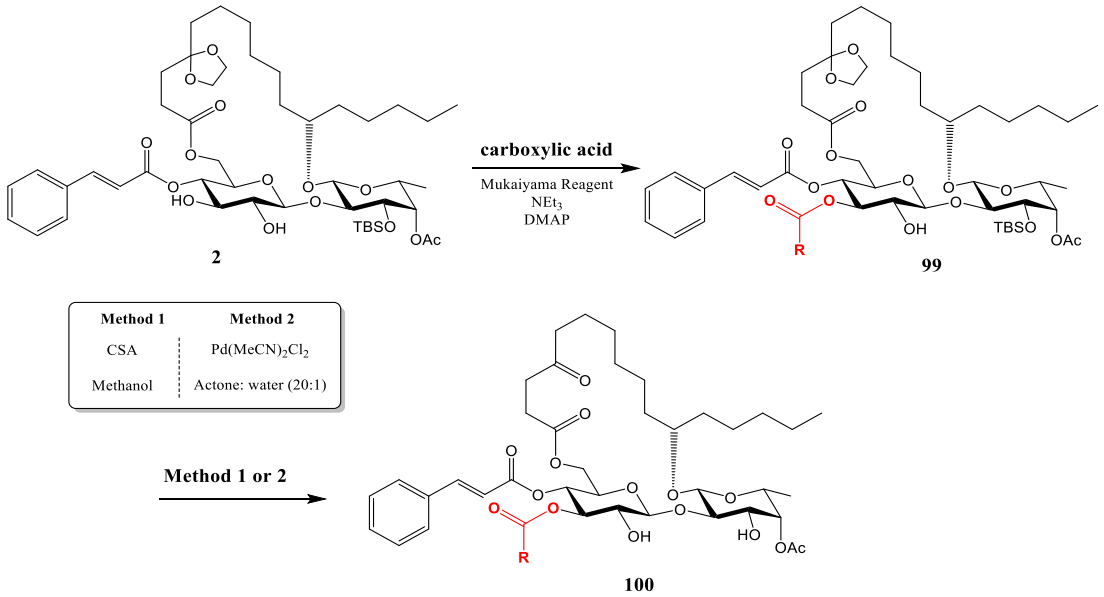


Figure 2.11. C-3-Glc*p* modified ipomoeassin F analogs **96-98**.

second analog **97** focused on the *β*-methyl group of the tig moiety with a replacement of methylacrylic acid. The third analog **98** studied the role of both methyl groups by using acrylic acid. Another reason these modifications were chosen was the three acids needed for the modifications are commercially available. This allowed for more time to be spent on the synthesis of analogs **96-98** and not preparing potential modifications for the C-3-Glc*p* moiety. Systematically, studying each methyl group of the tig moiety would help determine the role and importance of this position in the overall cytotoxicity of ipomoeassin F **1**. The following section presents the details of the syntheses of the C-3-Glc*p* ipomoeassin F analogs **96-98**.

2.11.1.Modified Synthesis Route for Mukaiyama Esterification and One-Pot De-Protection

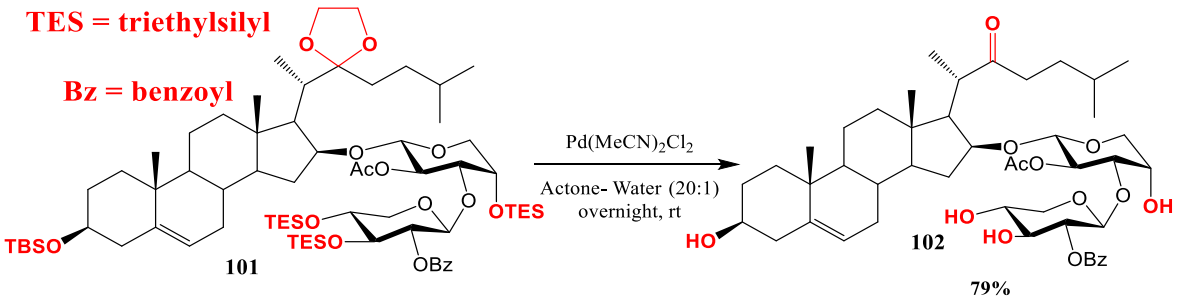
After scaling up the C-4-acetal macrocycle diol intermediate **2**, we started to think about a modified synthesis route to prepare the acrylate analogs **96-98**. For the completion of ipomoeassin F **1** (Scheme 2.20), the final step had been a de-protection of the -OTBS group at the *C-3* of the



Scheme 2.20. Modified synthesis route of *C-3* glucosyl modified ipomoeassin F analogs from the key diol intermediate **2**.

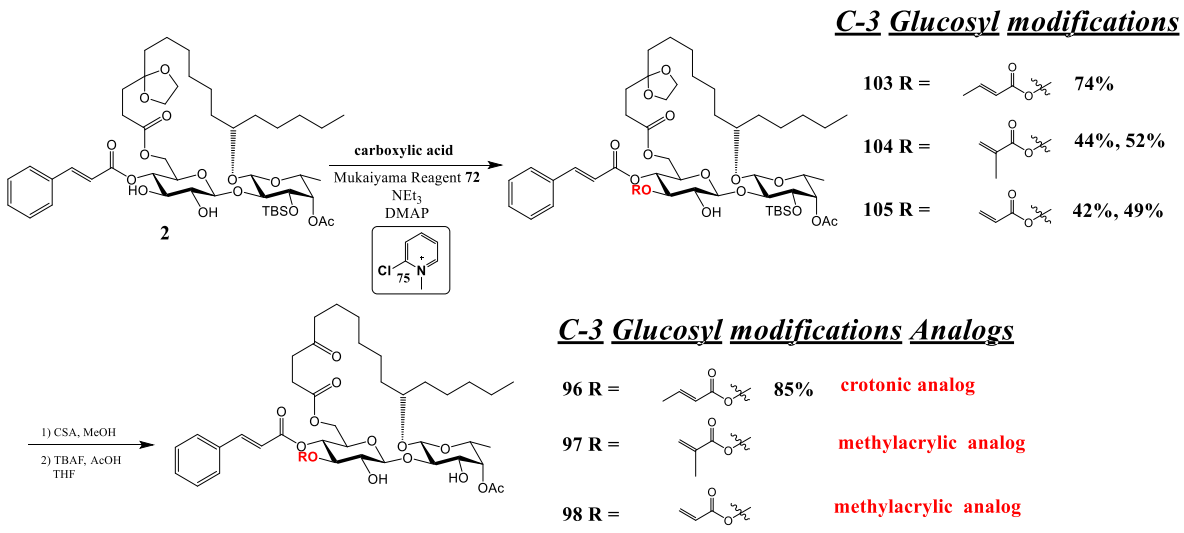
Fuc*p* moiety. However, we believed the *C-4* cyclic acetal could be carried through the synthesis until the final step. Then, a one-pot de-protection could be performed to de-protection both the *C-4* acetal and the silyl ether at C-3-Fuc*p*. It is well-known that silyl ethers, such as -OTBS groups, are acid-labile and can be removed under slightly acidic conditions. This was observed as an unwanted side-product when the isopropylidene acetal was de-protected with CSA (**65**→**6** Scheme 2.11). When the reaction was left too long or not properly quenched, both the isopropylidene acetal and OTBS group would easily be removed giving the triol **66**.

In the synthesis of a saponin family natural product, OSW-1 **102**, Deng *et al*. design a similar reaction where multiple protecting groups (one TBS, one ethylene glycol ketal, and three TES) were easily removed with $Pd(MeCN)_2Cl_2$ as a catalyst (Scheme 2.21). The modified synthesis route (Scheme 2.20) offered a few advantages over the original route (Scheme 2.19). First, Mukaiyama esterification conditions were chosen for the coupling reaction of the OH-3-Glc*p* and the acrylic acids. This esterification method offered advantages over Steglich esterification, including a shorter reaction time, simpler workup, and purification process. Secondly, performing a one-pot deprotection would remove one step in the total synthesis of the ipomoeassin F analogs, allowing more time and material (diol intermediate **2**) to prepare the C-3-Glc*p* analogs. It was believed the changes to the original route would make C-3-Glc*p* analogs more accessible and easier to achieve.



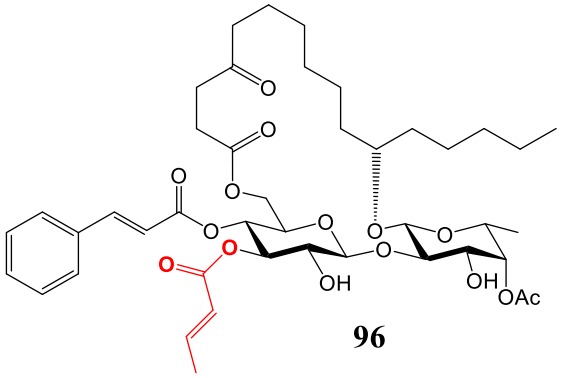
Scheme 2.21. Final step in the first synthesis of OSW-1 **102**.

The modified synthesis route for the acrylic analogs **96-98** is shown in Scheme 2.22. The regioselective esterification of the acids to the 3-*O*-Glc*p* was successful. The coupling reaction with crotonic modification **103** was the fastest, taking only a few hours to complete, while the methylacrylic **104** and acrylic **105** analogs had to be left 12 hrs.-24 hrs. for most of the *C-4* acetal diol intermediate **2** to be consumed (70-80%). Afterwards, the intermediates were concentrated, and column chromatography was performed to obtain the C-3-Glc*p* modified intermediates. The yield for the crotonic modification **103** was >70%, while the yields for the methylacrylic **104** and acrylic **105** modifications were lower and inconsistent. The regioselectivity of the crotonic modification **103** was confirmed by the ^{1}H, ^{13}C, COSY, and HMBC NMR spectra data. The typical COSY and HMBC correlations were analogous to ipomoeassin F **1** shown in Figure 2.9. We speculated that the bulky TBS group at 3-*O*-Fuc*p* blocked the 2-OH-Glc*p*, causing the esterification to only occur at 3-OH-Glc*p*. When 2.5 eq of the crotonic and methylacrylic acids were needed to modify the C-3-Glc*p*, no C-2-Glc*p* acylated by-products were observed.



Scheme 2.22. Modified route for the esterification of the C-3-Glc*p* and one-pot de-protection.

The one-pot deprotection of the -OTBS and the *C-4* cyclic acetal proved to be more difficult than anticipated. The crotonic analog **96** was the first analog attempted using the modified one-pot deprotection. The *C-4* cyclic acetal in **103** was removed with CSA (0.2 eq), after 12 hrs. TBAF (1M in THF) and AcOH was added to the reaction mixture and stirred overnight at RT. At that point, the TLC showed one spot and the reaction to be complete. The mixture was concentrated, then purified by column chromatography. However, after multiple columns using Hex:EtOAc as the mobile phase, a pure ^{1}H or ^{13}C-NMR could not be obtained. Believing the impurities came be from elution mixture, the mobile phase was changed to DCM:MeOH. However, the change in the mobile phase did not increase the purity of the crotonic analog **87**. The highest purity that could be obtained was 80-85% due to a complex mixture of signals observed in the aliphatic region of the ^{1}H- and ^{13}C-NMR (See Chapter 3 Experimental Section, ^{1}H-NMR 2.0-0.8 ppm and ^{13}C-NMR 45-20 ppm). Along with multiple purification methods,



96

Table 2.2. Cytotoxicity (IC_{50}, nM) of Ipomo. F and Crotonic Analog

	MDA-MB-231	MCF-7 (MTT)
Ipomoeassin F 1	14.0	70.0
96	13.4	200

[a] The data were obtained from at least two independent experiments, and the standard errors are within 20%.

numerous work-up procedures were performed to remove any excess TBAF, silyl compounds (TBSF or TBSOH), or acetic acid before performing column chromatography. Like the crotonic analog **96** obtaining pure methacrylic **97** or acrylic **98** analogs was not possible for this synthesis route. For the final deprotection of the acrylic analog **98**, the yield varied when even the TLC of the reaction showed one spot. The small amount of methylacrylic **97** and acrylic **98** analogs obtained after purification by column chromatography showed a complex mixture of signals in the aliphatic region. In-house cytotoxicity evaluations were performed on the crotonic analog **96** on two human breast cancer cell lines (MDA-MB-231 and MCF7). The results showed that in crotonic analog **96** retained most of the cytotoxicity as ipomoeassin F **1** (2-3 fold loss).

In November of 2017, a senior lab member was given 350-400 mg of the versatile diol **2** to attempt to prepare some of the acrylic analogs **96-98**. This lab member had been successful in preparing numerous analogs to facilitate SAR studies of ipomoeassin F **1**. However, some of the same problems were encountered, such as varying yields, multiple isomers in the final de-protection, and difficulty purifying the analogs by column chromatography. Therefore, we concluded that *α*,*β*-substitution of unsaturated ester (tig moiety, tri-substituted) is necessary to easily obtain C-3-Glc*p* modified ipomoeassin F analogs.

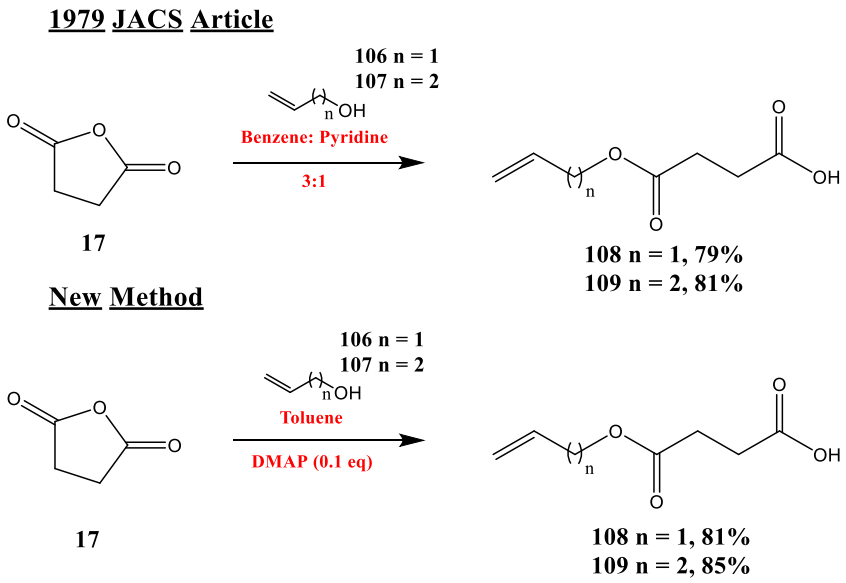
2.12. Left-Tether Modification to Study the *C-4* Ketone and the Macrocycle Ring Size

2.12.1. Synthesis of Bioisosteric Modifications

In 2016, Shi *et al.* become very interested in the role of the aglycone for the cytotoxicity of ipomoeassin F **1**, especially the *C-4* ketone and macrocycle ring size. A *C-4* deoxy aglycone analog showed the *C-4* ketone was not vital to the anti-cancer activity of ipomoeassin F **1**.[40] This

analog made studies of the aglycone moiety very interesting, since almost all resin glycoside contains tetradecanoic (C_{14}) or hexadecenoic (C_{16}) acid without structural functionality.[63, 64] For this study, the *C-4* ketone was replaced with an ester functionality. Replacing the *C-4* ketone with an ester eliminated a few problems associated with the current synthesis route. For the current aglycone synthesis route, the *C-4* ketone had to be protected to selectively remove the C-2/C-3-lev group later in the synthesis (Figure 2.8). Then, the *C-4* acetal had to be de-protected to reform the ketone (Scheme 2.17). These problems caused the synthesis route to become longer, specifically the aglycone (four steps vs. one step). The preparation of the 4-oxo-8-nonenoic acid **14** was another problem in the synthesis of the right tether of the aglycone (Scheme 2.3). This was the first and limiting for the preparation of the left tether of the aglycone (<35%). Also, an oxygen atom could be considered a bioisostere of methylene according to the Grimm's Hydride Displacement. A bioisostere is a compound resulting from the exchange of a specific atom or collection of atoms. Often, the bioisosteric modified atom has the same hybridization or on the same column of periodic table (e.g. carbon vs. silicon). For bioactive molecules, bioisosteric substitutions have long been used by medical chemists to adjust activity, improve selectivity, and/or stability during reduction metabolism.[65-67] Replacement of the *C-4* ketone with the ester functionality shorten the synthesis of the aglycone (one vs. four), increased the yield of the left-tether of the aglycone, and provided critical SAR information about the aglycone of ipomoeassin F **1**.

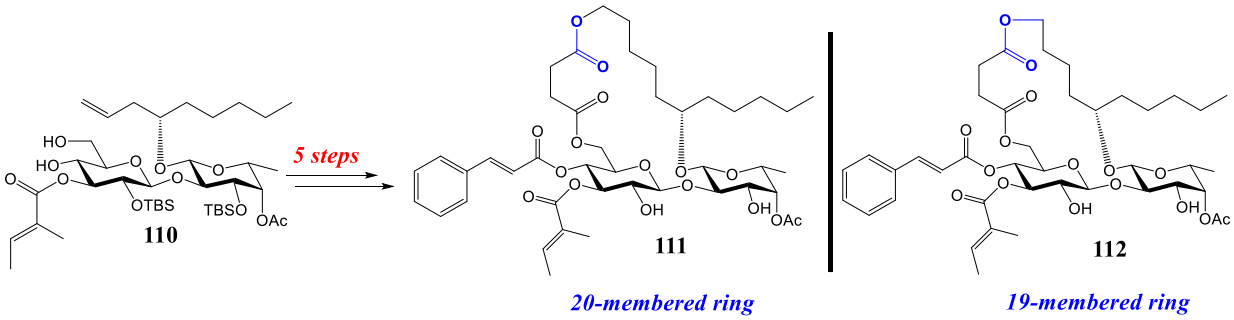
Scheme 2.23 shows the two methods used to prepare 1-(2-propen-1-yl) ester acid 1-(buten-1-yl) ester **108** and butanedioic acid 1-(propen-1-yl) ester **109**. The longer acid **109** formed the same size macrocycle as ipomoeassin F **1** (20-membered ring), while the shorter acid **108** was one member smaller (19-membered ring). The literature called for using succinic anhydride **17** and



Scheme 2.23. Two conditions for preparing the esters **108** and **109** for the left-tether of the aglycone.

allyl alcohol **106** with benzene/ pyridine as a solvent. The reported yield was 79%; however, the solvent mixture of benzene/ pyridine was a potential safety issue. Because of this potential issue another method was utilized based on work by Shuller *et al.* For the new method, toluene (Tol) was the solvent catalyzed by DMAP. The Tol/ DMAP method gave an 85% yield for the butanedioic acid 1-(propen-1-yl) ester **108**. The butanedioic acid 1-(buten-1-yl) ester **109** was prepared by the same method as the shorter acid **108** with 81%.[68] The route was adopted from a method developed by Cheng *et al.*[28, 56] The C-5-oxa analogs of ipomoeassin F **1** were finished by another lab member in five steps from the diol intermediate **110** (Scheme 2.24). The 5-oxa analog **111** was found to be active with only a 3-5-fold loss in activity, making **111** a potential replacement for ipomoeassin F **1** (Table 2.3). To our surprise, 19-membered 5-oxa analog **112** caused a significant loss in the in the cytotoxicity (9-fold for both MDA-BD-231 and MCF7 when compared to ipomoeassin F **1**, Table 2.4). More analogs are needed to better understand the importance of

the macrocycle's size and the effect of the aglycone. These analogs displayed that the ring size seems important for the biological activity of ipomoeassins.



Scheme 2.24. Synthesis of *C-5* ester ipomoeassin F analogs **111** and **112**.

TABLE 2.3. ***C-5 Aglycone Modified Analogs*** **(IC_{50} Data nM)[a,b]**

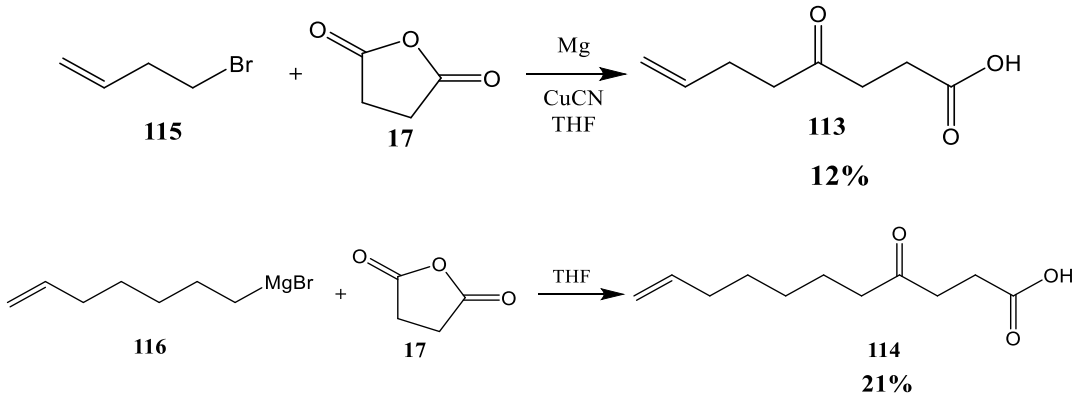
	MDA-MB-231[c]	**MCF7[d]**	**MCF-10A[c]**
Ipomoeassin F 1	7.7	36.7	5.1
111	31.5	81.2	26.9
112	290.3	710.5	–[e]

[a] 3-day treatment.
[b] Data was obtained from at least two independent experiments (standard error <20%).
[c] Alamar Blue
[d] MTT.
[e] "–" = not tested.

2.12.2. Synthesis of Left-Tether Modification to Study Ring Size

Last summer, Shi *et al*. became interested in the role the of the macrocycle ring size for the overall cytotoxicity of ipomoeassin F **1**. Unlike 5-oxa analogs **111** and **112**, for this study focused on the size and not any bioisosteric substitutions or modifications. For most resin glycosides, the aglycone is made up of jalapinolic acid, (11*S*)-hydroxyhexadecanoic acid. The left tether of the aglycone was easier to modify because this moiety was introduced later in the synthesis (Scheme

2.17). So, two carboxylic acids, 4-oxo-7-octenoic acid **113** and 4-oxo-10-undecenoic acid **114**, were synthesized from Scheme 2.25.[69] The route for these two intermediates followed the same procedure as 4-oxo-8-nonenoic acid **14**. Like the synthesis of 4-oxo-8-noenoic acid **14** (Scheme 2.3), the yields for these two carboxylic acids **129** and **130** were rather low (12 % and 21%). The main reason for the low yield was any excess Grignard reagents again to the *C-4* ketone, forming



Scheme 2.25. Preparation of the 4-oxo-7-octenoic acid **113** and 4-oxo-10-undecenoic acid **114**.

the tertiary alcohol. Both carboxylic acids (CA) **113** and **114** will be used to prepare analogs to study the importance of the macrocycle size.

2.13. Potential Modifications to the C-3-Glucosyl Moiety

Since a significant amount of time was spent trying to obtain the acrylic analogs **96**-**98**, research began on further potential modification to the tig moiety of ipomoeassin F. At the time, we believed the C-3-Glc*p* acrylic analogs **96**-**98** were obtainable through the developed synthesis route and the difficulties were related to the purification in the final step (Scheme 2.22). From the cinn analog **93**, it was known aromatic modifications to the tig moiety resulted in a loss in cytotoxicity, while the crotonic analog **96** showed that small modifications of the α,β-methyl groups could be performed without greatly affecting the potency of ipomoeassin F **1**. Based on

these beliefs, the crotonic analog **96** could act as a structural scaffold for further modifications to the tig moiety because of its comparable activity and structural similarity to ipomoeassin F **1**.

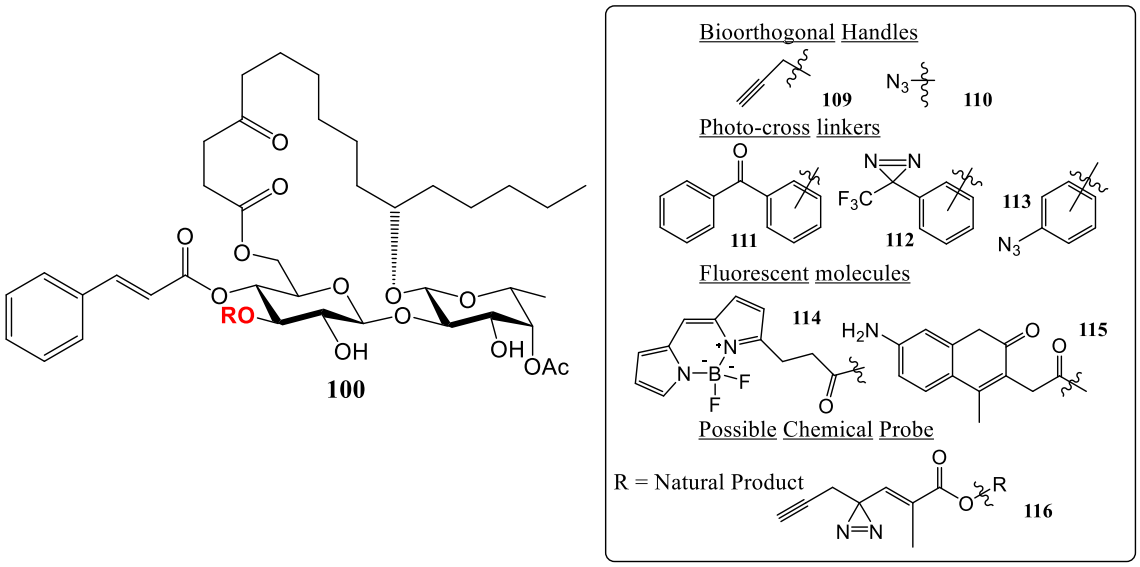


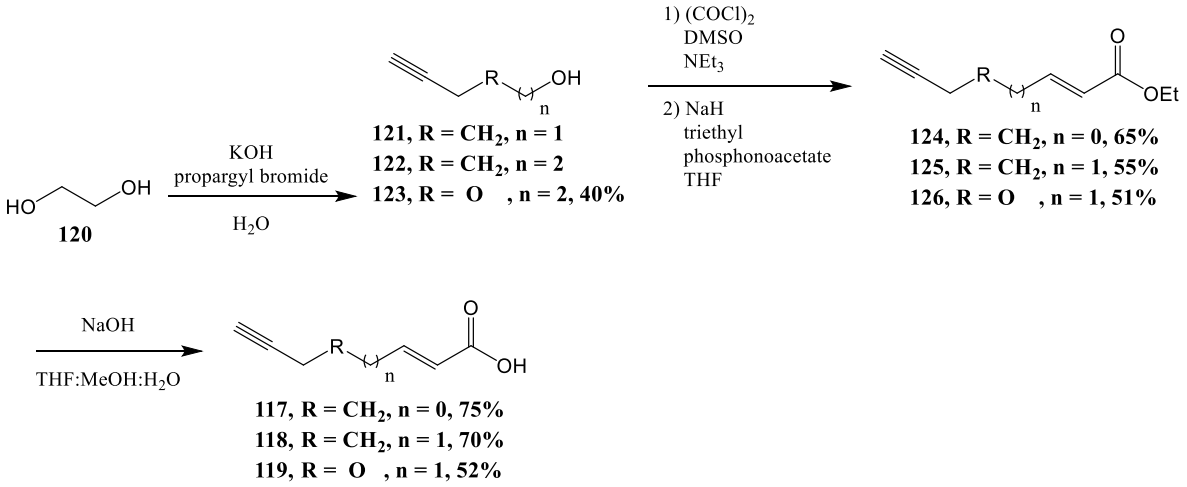
Figure 2.12. The structures of a possible chemical probe of ipomoeassin F **1**.

Based on these findings, we wanted to determine if further modifications could be made to the tig moiety to develop a chemical probe to push ipomoeassins research into new area of drug discovery and chemical biology (Figure 2.12).

2.13.1. The α,β-Unsaturated Acids Containing an Alkyne

The first group of potential modifications were based on the elongation of crotonic acid (Scheme 2.26). Potentially, the terminal alkyne could be used for future click chemistry studies and target identification. These acids were chosen because of their length and methylene carbons between the *α,β*-double and terminal alkyne. If the carbon-chain of the acid was shorter, the terminal alkyne proton would have to be protected. The *α,β*-unsaturated acids **117**-**119** were prepared from the commercially available ethylene glycol **120** and the alkyne alcohols **121**-**122** in three-four steps. The ethyl esters **124** and **125** were obtained using a one-pot Swern oxidation

reaction from **121** and **122** respectively, followed by a Horner-Wadsworth-Emmons (H-W-E) reaction. The conditions for the oxidation reaction and the H-W-E were modified from chemistry developed by Masamune and Roush conditions developed in the late 1980s.[59-61] The ether **121** was obtained from a Williamson ether synthesis of ethylene glycol **120** and propargyl bromide. Then, the same Swern oxidation/H-W-E reaction conditions were utilized to obtain **123** from the 2-(2′-propynloxy) ethanol **120**. Next, a hydrolysis reaction was performed with ethyl esters **124-126** to give the corresponding acids **117-119**. Because of potential polymerization and purification



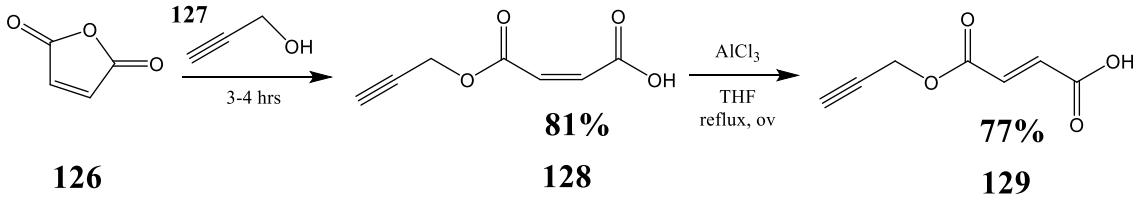
Scheme 2.26. Potential alkyne modifications for C-3-Glc*p* ipomoeassin F analogs.

concerns with acids **117-118**, only the (2*E*)-4-(2′-propynloxy)but-2-enoic acid **119** was used to modify the C-3-Glc*p* moiety. Unfortunately, an inseparable mixture of two isomers was obtained for the one-pot deprotection of the *C-4* cyclic acetal and the -OTBS group at 3-*O*-Fuc*p*.

2.13.2. Conjugated α,β-Unsaturated Double Bond Acids from Maleic Anhydride

The second set of potential modifications of the C-3-Glc*p* are shown in Scheme 2.27. The propargyl maleic acid **128** was obtained by a ring opening reaction of the maleic anhydride **126** and propargyl alcohol **127**. Then, the cis acid **128** was isomerized with cat. $AlCl_3$ to give the trans-

configured propargyl fumaric acid **129** in 77%. Like the cinn analog **93**, these modifications contain a conjugated *α,β*-unsaturated double bond. So, analogs based on the conjugated double

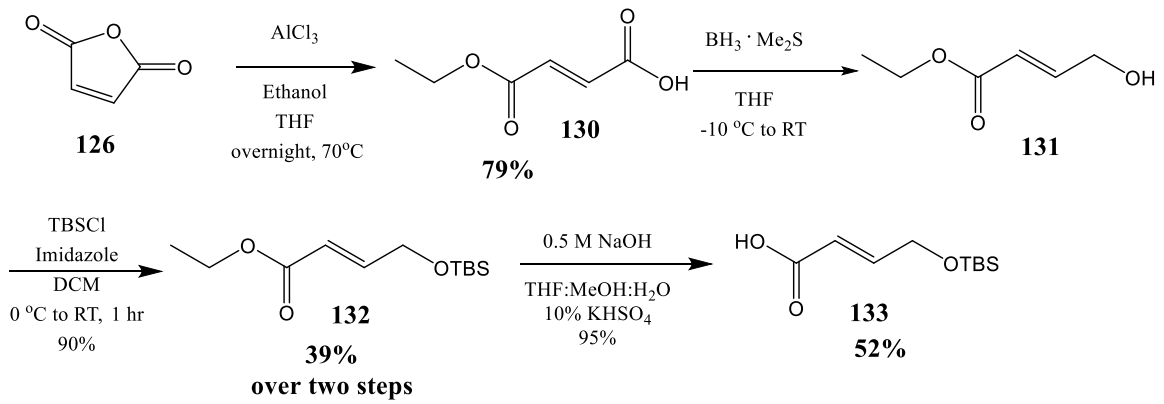


Scheme 2.27. Preparation the propargyl maleic acid **128** and propargyl fumaric acid **129** for C-3-Glc*p* modifications.

bond could be prepared easier than the acrylic analogs (Section 2.11). Unfortunately, the regioselective introduction of propargyl acids **128-129** to the diol intermediate **2** has not been attempted but could be performed to study modifying the C-3-Glc*p*.

2.13.3. Synthesis of 4-OTBS-Crotonic Acid for the Development of a Chemical Probe

The third modification was based on crotonic acid (Scheme 2.28). This modification is potentially interesting because the *C-4* hydroxyl group could act as a handle for further derivatization to develop a functional chemical probe (Figure 2.9). The route to reach 4-OTBS crotonic acid **133** takes five transformations from the maleic anhydride **126** developed by Ortiz *et al.* in 2015.[62] The first step was altered from the published route, where the maleic anhydride **126** was opened up by ethanol to give the monoethyl maleic acid. Then, cat. $AlCl_3$ was used to isomerize the double bond to give the trans, monoethyl fumaric acid **130**. We chose to perform both transformations in one-pot, by refluxing the maleic anhydride **126** and ethanol in THF with cat. $AlCl_3$. The isomerization of the double bond to give the monoethyl fumaric acid **130** was monitored by NMR and the difference in the coupling constant of the cis vs. trans double bond (trans J = 16 Hz and cis J = 12 Hz). Next, a selective reduction of the carboxylic acid **130** to the allyl alcohol **131** was performed in the presence of borane dimethylsulfide, followed by protection



Scheme 2.28. The synthesis of 4-OTBS-crotonic acid **133** for the alteration the C-3-Glc*p* of ipomoeassin F **1**.

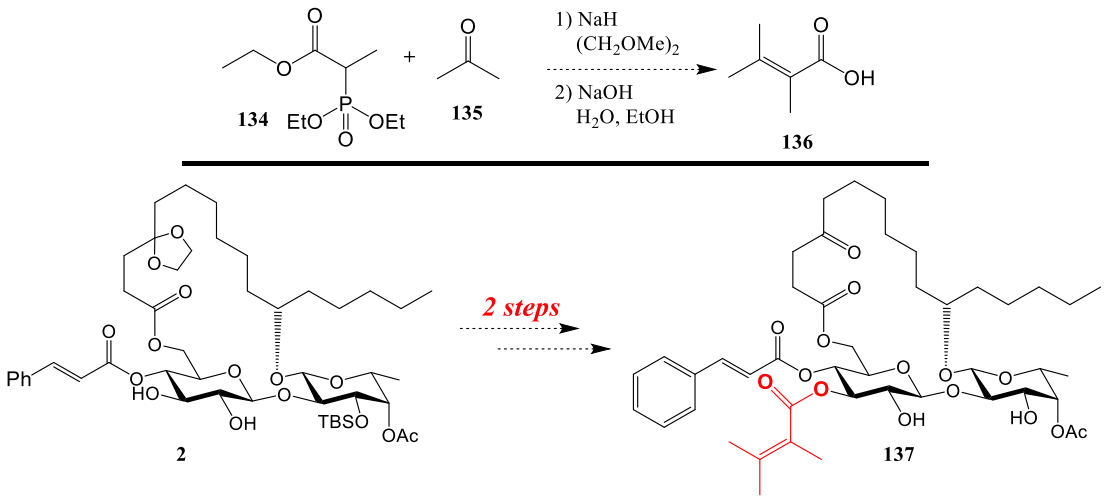
of the C-4 hydroxyl group with *tert*-butyldimethylsilyl chloride (TBSCl) to give **132** in 39% over two steps. Finally, a hydrolysis of ethyl ester **132** with NaOH in THF:MeOH:H_2O (4:1:1), followed by acidification of the solution (pH = 4-5), gave the acid **133**. The hydrolysis proved to be more difficult than anticipated. The reaction could not be performed in MeOH and the -OTBS was very acid-labile making the work-up very tedious. If the hydrolysis was attempted in MeOH, a mixture of acid **133** and methyl 4-OTBS-crotonate would be obtained.

2.14. Future Studies on the C-3-Glc*p* Moiety of Ipomoeassin F

From this work, significant knowledge was gained about the importance of the tig and cinn moieties for the overall anti-cancer activity of ipomoeassin F **1**. Unfortunately, the acrylic analogs **96-98** to systematically study the *α,β*-methyl groups of the tig moiety were not obtainable by the developed synthesis routes. These findings suggest the *α,β*-unsaturated double bond at C-3-Glc*p* must be at least tri-substituted (tig moiety, ipomoeassin F **1**) or part of an sp^2 conjected system (cinn analog **93**) to easily and successfully study the C-3-Glc*p* moiety of ipomoeassin F **1**.[70] The following section presents three potential classes of C-3-Glc*p* analogs that could be easily prepared and applied to the developed synthesis route.

2.14.1. Tetra-Substituted α,βUnsaturated Double Bond Analog

The first potential modification is 2,3-dimethylbutenoic acid **132** (Scheme 2.29). This modification would examine the importance of the *α,β*-methyl groups of the tig moiety, while possibly allowing easier purification than the less substituted acrylic analogs **96-98**. The 2,3-dimethylcrotonic acid **136** is commercially available (Sigma-Aldrich $509/1 g), but it can be synthesized in two steps from triethyl 2-phosphonopionate **134** and acetone **135**.[71] Then, the 2,3-dimethylcrotonate analog **137** can be obtained in two steps from the diol **2** through a regioselective esterification and the one-pot de-protection of the transient protecting groups.

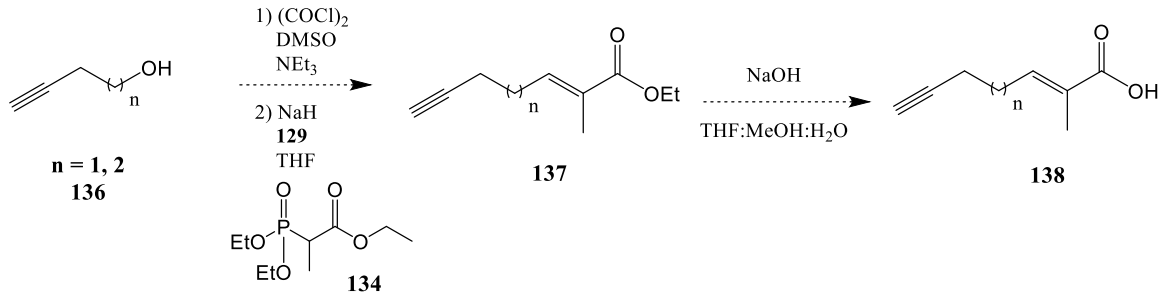


Scheme 2.29. Synthesis of 2,3-dimethylcrotonic analog **137**.

2.14.2. Tri-substituted α,β-Unsaturated Double Bond Acids for Click Chemistry

Some potential C-3-Glc*p* modifications are shown in Scheme 2.30. These possible modifications could be used for Click Chemistry and are like the ones shown in Scheme 2.26. The only difference is that these analogs contain a tri-substituted *α-β*-unsaturated double bond that

closely mimics the tig moiety in ipomoeassin F **1**. We do not believe that the terminal alkyne proton should to be protected, based on a C-4-Glc*p* cinn analog that contained a propargyl group. The major difference from Scheme 2.26 comes from triethyl 2-phosphonopionate **134** needed to form on the tri-substituted double bond, instead of triethyl phosphonoacetate. An H-W-E reaction can be adapted from chemistry published by Ambudka and Tang to form the ethyl ester **137**.[71-73] Then, a hydrolysis reaction with NaOH or LiOH can afford the acids **138.** After preparing the carboxylic acids, a regioselective trans-esterification can be performed between the acids **138** and the diol intermediate **2**, followed by removal of the transient protecting groups to obtain C-3-Glc*p*



Scheme 2.30. Potential modification for click chemistry based on the tri-substituted tiglic acid.

ipomoeassin F analogs (Scheme 2.22).

2.14.3. Terminal Hydroxyl Modification for Cell Imaging or Target Identification

The final class of potential analogs **139** discussed are based on the 4-hydroxylcrotonic modification (Figure 2.13). The hydroxyl group can act as a handle to introduce different structural motifs to the tig moiety. Analogs **140-142** could be utilized in cellular localization studies and protein pull-down experiments for activity-based protein profiling (ABPP). The coumarin **141** and biotin **142** modifications should be easily incorporated into the synthesis for the studies of the C-3-Glc*p* of ipomoeassin F; however, the BODIPY analog **140** may be more difficult to synthesize

because of stability concerns. If the BODIPY analog **140** is not obtainable, other fluorescent molecules can be used, such as pyrene or anthracene.

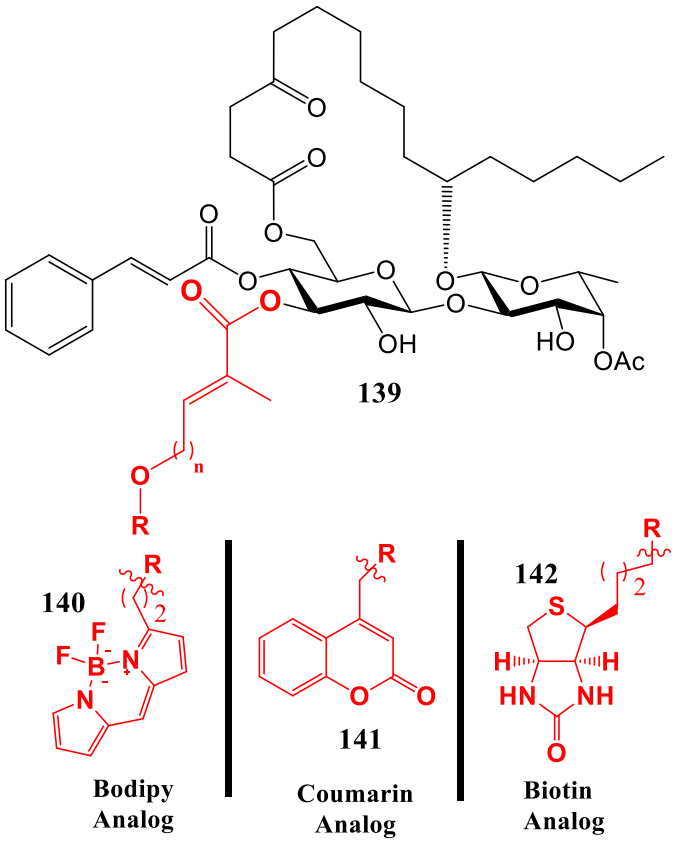
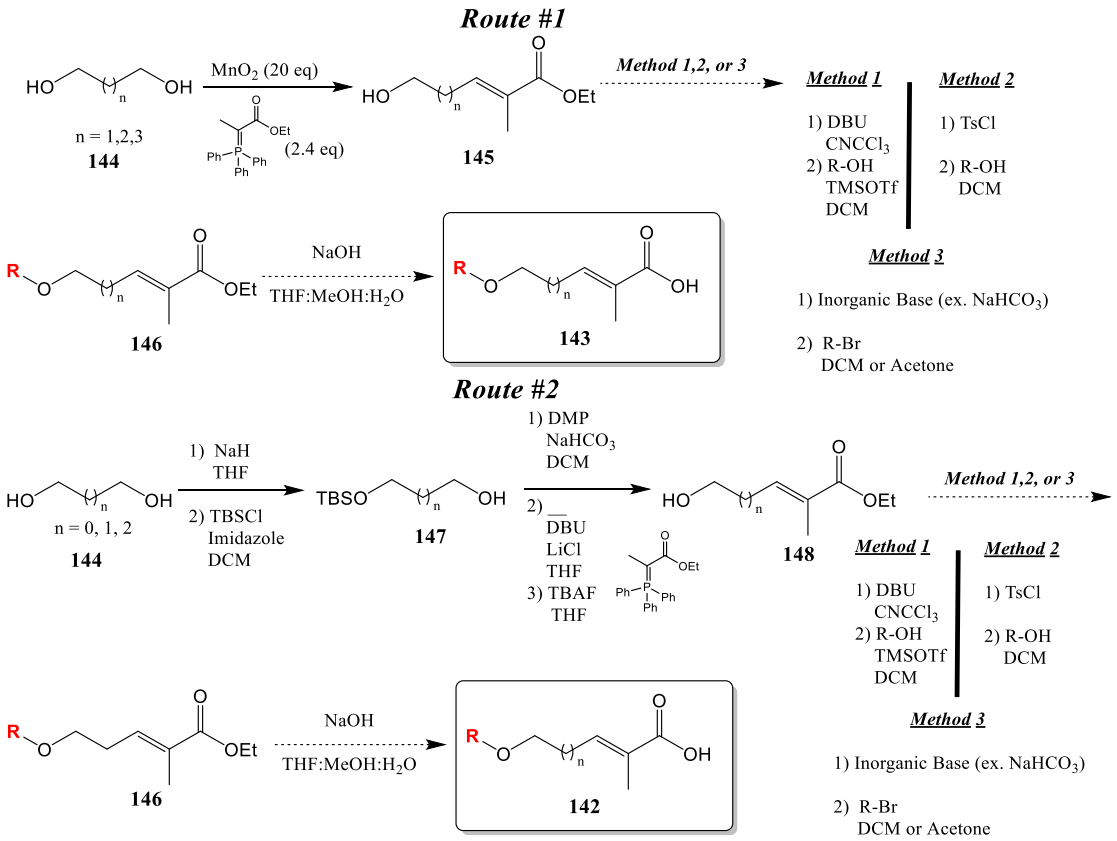


Figure 2.13. Potential analogs **139-141** based on 4-hydroxyl tig mod. of ipomoeassin F **1**.

The syntheses of the modified 4-hydroxyl tig acids **143** are shown in Scheme 2.31. Route 1 is a more direct path to achieve the tig modifications **143**, while route 2 offers a back-up plan if the hydroxyl group needs be protected with a silyl ether. For route 1, the **144** diol can be partially oxidized by MnO_2, then an H-W-E reaction can be performed to get the ethyl ester alcohol **145**.[74, 75] At this point, the potential modifications can be introduced through a donor-acceptor reaction (method 1), a tosylate (method 2), or an inorganic base method (method 3). Method 1 or 2 should be the ideal process to install the modifications because an inorganic base (method 3) could potentially cause hydrolysis of the ethyl ester **146**. The last step in route 1 is hydrolysis of the



Scheme 2.31. The possible synthesis route of 4-hydroxyl tig modifications for ipomoeassin F analogs.

ethyl ester **145** to achieve the carboxylic acids **143**. Route 2 is a few steps longer but allows for the protection of one of the alcohols with a silyl ether. First, the alcohol **144** can be protected with TBSCl by using sodium hydride to give **147**. Then, an H-W-E reaction can be performed, and the TBS group can be removed with TBAF to give the primany alcohol **148**. The last two steps in route 2 utilized the same transformations as in route #1 to get the carboxylic acids **143**. Like the other potential C-3-Glc*p* modification, these tig modifications can be introduced to the diol **2** in the next to last step through a regioselective esterification. The modifications to the C-3-Glc*p* analogs presented in this section build on cinn analog **93** and acrylic analogs **96-98** (Section 2.11). These C-3-Glc*p* modifications could limit some of the problems experienced with the acrylic analogs **96-98** and provide new insights into the mechanism of action (MOA) of ipomoeassin F **1**.

2.15. Prospective and Future Direction for C-3-Glucosyl Modifications of Ipomo. F

Many challenges were encountered designing the synthesis route for late-stage modification of the C-3-Glc*p* moiety of ipomoeassin F **1**. The previous syntheses of ipomoeassin F **1**, provided a firm base to start designing a synthesis route. We knew the RCM strategy would be the best method to construct the macrocycle and the increased nucleophilicity of C-3-OH could be utilized to perform regioselective transformations.[6, 7, 9, 15] To develop a convergent synthesis route, the four moieties or building blocks of ipomoeassin F **1** were prepared separately (Section 2.2-2.5). This made the synthesis more efficient and allowed for modifications of the different building blocks without greatly affecting the rest of the molecule. The synthesis of the (4*S*)-nonenol **10** and the Fuc*p* acceptor **9** were straight forward and proceeded as expected. The (4*S*)-nonenol **10** was prepared from the (*S*)- epichlorohydrin **11** in three steps. While, the Fuc*p* acceptor **9** was synthesized in five steps from the D-Fuc*p* **13**. During the scaling-up process, a cheaper starting material needed to be utilized for a multi-gram synthesis. Diacetone-D-Gal **21** provided a cheaper starting material for the Fuc*p* acceptor **9** but did make the synthesis route 3-4 steps longer.

The synthesis of the Glc*p* donor **61** and 4-acetal-8-nonenoic acid **5** proved to be more difficult. The criteria to develop an efficient, effective synthesis route for the Glc*p* donor were transient protecting groups at the C-3-Glc*p* and C-4/C-6-Glc*p* positions. After a few ineffective synthesis routes were investigated, we started to think about using the same transient group at C-2/C-3 Glc*p* (Section 2.7). Lev groups were chosen as the transient protection for these two positions. The lev groups helped form the β-(1→2)-disaccharide (neighboring group participation) and allowed for an orthogonally deprotection in the late-stage of the synthesis ($H_2NNH_2 \cdot$ AcOH). Unfortunately, we found out the *C-4* ketone was transformed to the stable *C-4* hydrazone during

the lev de-protection step. At this point a decision had to be made about the direction of the synthesis, did we want to design another Glc*p* donor or protect the *C-4* ketone? Since, the Glc*p* donor **61** had already been prepared on a multi-gram scale, we chose to protect the *C-4* ketone to for the preparation of 4-acetal-8-nonenoic acid **5**. This did make the synthesis of the left-tether of the glycone four steps (previous syntheses, one step), but did allow for late-stage de-protection of the C-2/C-3-lev groups (Section 2.9).

The macrocycle was successfully constructed by an RCM/hydrogenation reaction scheme (Section 2.8 and 2.10). Since we chose to install the *α*,*β*-unsaturated esters in the late stage, stronger hydrogenation conditions could be used reduce the cis/trans double bond. For the previous syntheses, the cinn group had to be protected or a specific catalyst to achieve a selective reduction of the cis/trans macrocycle.[7-9] After the orthogonal de-protection of lev groups, a regio-selective esterification to the HO-3-Glc*p* could be performed. Ipomoeassin F **1** and a C-3-Glc*p* cinn analog **93** were completed after the de-protection of the silyl ether at C-3-Fuc*p* (Section 2.10). Ipomoeassin F **1**, the cinn analog **93**, and other two analogs **94**-**95** were tested against two breast cancer cell lines to determine their IC_{50}. When compared to ipomoeassin F **1**, all the analogs were significantly less potent (Table 2.1, μM range). Suggesting the positioning of the aromatic *α*,*β*-unsaturated ester (C-4-Glc*p* cinn) and the small aliphatic ester (C-3-Glc*p* tig) provide a synergetic contribution to the cytotoxicity of ipomoeassin F **1**.

After scaling-up the versatile *C-4* acetal macrocycle diol intermediate **2**, we needed to determine the importance of the *α*,*β*-methyl groups before further modifications could be made to the C-3-Glc*p* moiety. So, a series of acrylate analogs **96**-**98** were envisioned, from a regioselective esterification of the *C-4* macrocycle diol **2** and commercially available acrylic acids. Ultimately, this synthesis route did not allow for the preparation of the acylate analogs **96**-**98**. The

regioselective esterification proceeded as expected with slight drop in yield, but difficulties were encountered during the purifications of the final one-pot de-protection. These results lead us to believe that the *α*,*β*-unsaturated ester must be at least tri-substituted (tig ipomoeassin F **1**) or part of the a sp^2 conjugated system (C-3-Glc*p* cinn analog **93**).

As the project concluded in the spring of 2018, we started to think about two questions. First, is the current route better than previous synthesis routes for ipomoeassin F **1**? Before the current work, three syntheses of ipomoeassin F **1** had been published since 2009.[7, 9, 15] All of these syntheses utilized RCM for the construction of the macrocycle, and Schmidt donors for the glycosylation reactions.[11, 35, 76, 77] Each synthesis had advantages and disadvantages, but none of the syntheses were efficient for medicinal chemistry studies of the tig moiety. The current work built on the previous syntheses by incorporating a late-stage regioselective esterification reaction to modify the C-3-Glc*p* moiety. This synthesis route can be utilized to further explore the importance of the C-3-Glc*p* moiety for the bioactivity of ipomoeassin F **1**.

The second question involved, "what could be the future direction for studying the C-3-Glc*p* moiety for ipomoeassin F **1**?" When thinking about the future direction of this project, it is important to remember the lack of published literature data on ipomoeassins.[19] Currently, very little is known about the MOA of ipomoeassins.[19, 39, 40] So, future work must be centered around chemistry to develop analogs to push ipomoeassins research into new areas of drug discovery and chemical biology. Section 2.13-14 discussed some potential modifications for the C-3-Glc*p* moiety to facilitate further studies. The modifications are based on tig moiety of the natural product (tri-substituted). We believe these modifications could limit some of the problems experienced with the purification of the acylate analogs **96**-**98**. These proposed analogs in could provide vital insight into the MOA and potential biological target/targets of ipomoeassins.

Another area of future research evolves modifications and optimization to the current synthesis route. The synthesis of the left-tether of the aglycone was significantly longer (four steps vs. one step) and give a lower overall yield when compared to previous syntheses (Section 2.2). Developing a shorter, optimized synthesis route for the 4-acetal-8-nonenoic acid **5** would make the current synthesis route more efficient. Schmidt donors were utilized for multiple glycosylation reactions in the current route, because of their versatility and reliability.[34, 43, 76] However, when looking at the synthesis of the monosaccharides building blocks this presented a disadvantage in the current synthesis route (Section 2.2-2.4). When preparing the Schmidt donor, we first must selectively de-protect the anomeric group to give a hemiacetal, then make the Schmidt donor with trichloroacetonitrile. This forced us to perform two extra chemical transformation and give a moisture sensitive donor. Thioglycosides have been shown to be useful as donors for glycosylation reactions. Developing a glycosylation reaction with the thioglycoside (Scheme 2.4) would decrease the number of steps (2-3 steps) and give an air/moisture stable donor for glycosylation reactions ($\beta(1\rightarrow2)$-disaccharide). The regioselective esterification of the diol **6** and 4-acetal-8-nonenoic acid **5** is another transformation that could be optimized (Section 2.7). The yield of this reaction could not be fully optimized (<60%) and the purification of the desired product was difficult because of the similar polarity of the triene bi-product. These modifications and optimizations are just a few aspects of the synthesis route that could investigated further. More research is needed by both organic chemists and chemical biologists before ipomoeassins can reach their full potential as a potential anti-cancer chemotherapeutic. This work provides a firm footing for researchers to continue investigating the role of the tig moiety for the bioactive of ipomoeassin F **1**.

2.16. Conclusion

The goal of this project was to design a synthesis route to study the role of the C-3-Glc*p* moiety of ipomoeassin F **1**. From a previous work, it was known the C-3-Glc*p* position was important to the overall activity of ipomoeassin F **1**, but a systematic study of the tig moiety had not been performed. The developed synthesis route for the C-3-Glc*p* modified ipomoeassin F analogs highlighted multiple regioselective reactions and improved reaction conditions over previous syntheses. The two glycone moieties (Glc*p* **12** and Fuc*p* **13**) were prepared separately from cheap, commercially available starting materials through straightforward transformations and (1→2)-*β*-disaccharide **7** was formed from a regioselective glycosylation reaction. The aglycone moiety was, also, divided into two parts, (4*S*)-nonenol **10** and 4-acetal-8-nonenoic acid **5**. These two moieties of the aglycone were connected after the glycosylation reaction through an RCM reaction forming the macrocycle of ipomoeassin F **1**. Separately preparing the two moieties allowed for multiple aglycone analogs to be prepared without modifications complex (1→2)-*β*-disaccharide. Completion of the *C-4* ketone macrocycle diol intermediate **71** was vital for the developed synthesis route. Through a regioselective esterification reaction, ipomoeassin F **1** and a cinn analog **93** analog were successfully synthesized. Proposed acrylic analogs **96**-**98** were not obtainable through the developed synthesis route. A few different reaction conditions and eluent systems were attempted without significant success. Through the synthesis of ipomoeassin F **1**, the cinn **93** and crotonic analog **96**, this work provided new insights into the importance of the C-3-Glc*p* moiety of ipomoeassin F **1**. Aromatic modifications like the cinn analog **93** greatly decrease the bioactivity, while a small modification like the crotonic analog **96** retained most of the cytotoxicity of ipomoeassin F **1**. The developed synthesis route provided a simple, scalable, and effective synthesis route to study a vital moiety of ipomoeassin F **1**. Future work based on

established synthesis route and knowledge gained through this project will continue to push ipomoeassins research into new areas of chemical biology and drug discovery research.

2.17. References

1. Christensen, H. M.; Oscarson, S.; Jensen, H. H., Common side reactions of the glycosyl donor in chemical glycosylation. *Carbohydrate Research* **2015,** *408*, 51-95.

2. Williams, R.; Galan, M. C., Recent Advances in Organocatalytic Glycosylations. *European Journal of Organic Chemistry* **2017,** (42), 6247-6264.

3. Danishefsky, S. J.; Allen, J. R., From the laboratory to the clinic: A retrospective on fully synthetic carbohydrate-based anticancer vaccines. *Angewandte Chemie-International Edition* **2000,** *39* (5), 836-863.

4. Tamura, J.; Neumann, K. W.; Ogawa, T., Synthetic studies of glycosyl serines in the carbohydrate-protein region of protoglycans. *Liebigs Annalen* **1996,** (8), 1239-1257.

5. Escobedo-Martinez, C.; Pereda-Miranda, R., Resin glycosides from Ipomoea pes-caprae. *Journal of Natural Products* **2007,** *70* (6), 974-978.

6. Furstner, A.; Nagano, T., Total syntheses of ipomoeassin B and E. *Journal of the American Chemical Society* **2007,** *129* (7), 1906-+.

7. Zong, G. H.; Barber, E.; Aljewari, H.; Zhou, J. H.; Hu, Z. J.; Du, Y. C.; Shi, W. Q., Total Synthesis and Biological Evaluation of Ipomoeassin F and Its Unnatural 11R-Epimer. *Journal of Organic Chemistry* **2015,** *80* (18), 9279-9291.

8. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Mueller, R.; Fuerstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chem. - Eur. J.* **2009,** *15* (38), 9697-9706, S9697/1-S9697/48.

9. Postema, M. H. D.; TenDyke, K.; Cutter, J.; Kuznetsov, G.; Xu, O. L., Total Synthesis of Ipomoeassin F. *Organic Letters* **2009,** *11* (6), 1417-1420.

10. Furstner, A.; Muller, T., Efficient total syntheses of resin glycosides and analogues by ring-closing olefin metathesis. *Journal of the American Chemical Society* **1999,** *121* (34), 7814-7821.

11. Furstner, A., Total syntheses and biological assessment of macrocyclic glycolipids. *European Journal of Organic Chemistry* **2004,** (5), 943-958.

12. Deng, S. J.; Yu, B.; Lou, Y.; Hui, Y. Z., First total synthesis of an exceptionally potent antitumor saponin, OSW-1. *Journal of Organic Chemistry* **1999,** *64* (1), 202-208.

13. Deng, S. J.; Yu, B.; Xie, J. M.; Hui, Y. Z., Highly efficient glycosylation of sapogenins. *Journal of Organic Chemistry* **1999,** *64* (19), 7265-7266.

14. Furstner, A.; Muller, T., Metathesis route to resin glycosides: Formal total synthesis of tricolorin A. *Journal of Organic Chemistry* **1998,** *63* (3), 424-425.

15. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Muller, R.; Furstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chemistry-a European Journal* **2009,** *15* (38), 9697-9706.

16. Wang, H. Y.; Blaszczyk, S. A.; Xiao, G. Z.; Tang, W. P., Chiral reagents in glycosylation and modification of carbohydrates. *Chemical Society Reviews* **2018,** *47* (3), 681-701.

17. Yang, B.; Yang, W. Z.; Ramadan, S.; Huang, X. F., Pre-Activation-Based Stereoselective Glycosylations. *European Journal of Organic Chemistry* **2018,** (9), 1075-1096.

18. Leng, W. L.; Yao, H.; He, J. X.; Liu, X. W., Venturing beyond Donor-Controlled Glycosylation: New Perspectives toward Anomeric Selectivity. *Accounts of Chemical Research* **2018,** *51* (3), 628-639.

19. Cao, S.; Norris, A.; Wisse, J. H.; Miller, J. S.; Evans, R.; Kingston, D. G. I., Ipomoeassin F, a new cytotoxic macrocyclic glycoresin from the leaves of Ipomoea squamosa from the Suriname rainfores. *Natural Product Research* **2007,** *21* (10), 872-876.

20. Cao, S. G.; Guza, R. C.; Wisse, J. H.; Miller, J. S.; Evans, R.; Kingston, D. G. I., Ipomoeassins A-E, cytotoxic macrocyclic glycoresins from the leaves of Ipomoea squamosa from the Suriname rainforest. *Journal of Natural Products* **2005,** *68* (4), 487-492.

21. Cao, S. G.; Guza, R. C.; Wisse, J. H.; Evans, R.; Miller, J. S.; Kingston, D. G. I., Ipomoeassins A-E, five new cytotoxic macrocyclic glycoresins, from the leaves of Ipomoea squamosa from the Suriname rainforest. *Abstracts of Papers of the American Chemical Society* **2005,** *229*, U184-U184.

22. Ito, Y., My Stroll in the Backyard of Carbohydrate Chemistry. *Trends in Glycoscience and Glycotechnology* **2010,** *22* (125), 119-140.

23. Moriarty, R. M.; Rani, N.; Enache, L. A.; Rao, M. S.; Batra, H.; Guo, L.; Penmasta, R. A.; Staszewski, J. P.; Tuladhar, S. M.; Prakash, O.; Crich, D.; Hirtopeanu, A.; Gilardi, R., The intramolecular asymmetric Pauson-Khand cyclization as a novel and general stereoselective route to benzindene prostacyclins: Synthesis of UT-15 (treprostinil). *Journal of Organic Chemistry* **2004,** *69* (6), 1890-1902.

24. Berkessel, A.; Rollmann, C.; Chamouleau, F.; Labs, S.; May, O.; Groger, H., Practical two-step synthesis of an enantiopure aliphatic terminal (S)-epoxide based on reduction of

haloalkanones with "Designer Cells". *Advanced Synthesis & Catalysis* **2007,** *349* (17-18), 2697-2704.

25. Ritschel, J.; Sasse, F.; Maier, M. E., Synthesis of a benzolactone collection using click chemistry. *European Journal of Organic Chemistry* **2007,** (1), 78-87.

26. Quitmann, H.; Fan, R.; Czermak, P., Acidic Organic Compounds in Beverage, Food, and Feed Production. *Biotechnology of Food and Feed Additives* **2014,** *143*, 91-141.

27. Huang, X. R.; Pan, X. H.; Lee, G. H.; Chen, C. P., C-2-Symmetrical Bipyridyldiols as Promising Enantioselective Catalysts in Nozaki-Hiyama Allylation. *Advanced Synthesis & Catalysis* **2011,** *353* (11-12), 1949-1954.

28. Verma, A.; Jana, S.; Prasad, C. D.; Yadav, A.; Kumar, S., Organoselenium and DMAP co-catalysis: regioselective synthesis of medium-sized halolactones and bromooxepanes from unactivated alkenes. *Chemical Communications* **2016,** *52* (22), 4179-4182.

29. Fais, M.; Karamanska, R.; Allman, S.; Fairhurst, S. A.; Innocenti, P.; Fairbanks, A. J.; Donohoe, T. J.; Davis, B. G.; Russell, D. A.; Field, R. A., Surface plasmon resonance imaging of glycoarrays identifies novel and unnatural carbohydrate-based ligands for potential ricin sensor development. *Chemical Science* **2011,** *2* (10), 1952-1959.

30. Wang, Q.; Grkovic, T.; Font, J.; Bonham, S.; Pouwer, R. H.; Bailey, C. G.; Moran, A. M.; Ryan, R. M.; Rasko, J. E. J.; Jormakka, M.; Quinn, R. J.; Holst, J., Monoterpene Glycoside ESK246 from Pittosporum Targets LAT3 Amino Acid Transport and Prostate Cancer Cell Growth. *Acs Chemical Biology* **2014,** *9* (6), 1369-1376.

31. Cheng, J. M. H.; Dangerfield, E. M.; Timmer, M. S. M.; Stocker, B. L., A divergent approach to the synthesis of iGb3 sugar and lipid analogues via a lactosyl 2-azido-sphingosine intermediate. *Organic & Biomolecular Chemistry* **2014,** *12* (17), 2729-2736.

32. Milhomme, O.; John, C.; Djedaini-Pilard, F.; Grandjean, C., Access to Antigens Related to Anthrose Using Pivotal Cyclic Sulfite/Sulfate Intermediates. *Journal of Organic Chemistry* **2011,** *76* (15), 5985-5998.

33. Kufner, U.; Schmidt, R. R., DENOVO SYNTHESIS OF CARBOHYDRATES AND RELATED NATURAL-PRODUCTS .20. SYNTHESIS OF DEOXYHEXOSES FROM DIVINYLGLYCOLS - THE SYNTHESIS OF D-CHALCOSE AND L-CHALCOSE. *Angewandte Chemie-International Edition in English* **1986,** *25* (1), 89-89.

34. Schmidt, R. R., NEW METHODS FOR THE SYNTHESIS OF GLYCOSIDES AND OLIGOSACCHARIDES - ARE THERE ALTERNATIVES TO THE KOENIGS-KNORR METHOD. *Angewandte Chemie-International Edition in English* **1986,** *25* (3), 212-235.

35. Tiwari, V. K., Prof. Dr. Richard R Schmidt A tribute. *Arkivoc* **2013**, 1-5.

36. Zong, G. H.; Yan, S. Q.; Liang, X. M.; Zhang, J. J.; Wang, D. Q.; Kong, F. Z., Highly efficient removal of allyloxycarbonyl (Alloc) function provides a practical orthogonal protective strategy for carbohydrates. *Chinese Chemical Letters* **2009,** *20* (2), 127-130.

37. Hansen, S. G.; Skrydstrup, T., Studies directed to the synthesis of oligochitosans - Preparation of building blocks and their evaluation in glycosylation studies. *European Journal of Organic Chemistry* **2007,** (20), 3392-3401.

38. Fang, T.; Mo, K. F.; Boons, G. J., Stereoselective Assembly of Complex Oligosaccharides Using Anomeric Sulfonium Ions as Glycosyl Donors. *Journal of the American Chemical Society* **2012,** *134* (17), 7545-7552.

39. Zong, G. H.; Hirsch, M.; Mondrik, C.; Hu, Z. J.; Shi, W. Q., Design, synthesis and biological evaluation of fucose-truncated monosaccharide analogues of ipomoeassin F. *Bioorganic & Medicinal Chemistry Letters* **2017,** *27* (12), 2752-2756.

40. Zong, G. H.; Aljewari, H.; Hu, Z. J.; Shi, W. Q., Revealing the Pharmacophore of Ipomoeassin F through Molecular Editing. *Organic Letters* **2016,** *18* (7), 1674-1677.

41. Slaghek, T. M.; Nakahara, Y.; Ogawa, T.; Kamerling, J. P.; Vliegenthart, J. F. G., SYNTHESIS OF OLIGOSACCHARIDES RELATED TO HYALURONIC-ACID .2. SYNTHESIS OF HYALURONIC ACID-RELATED DI-SACCHARIDES, TRI-SACCHARIDES, AND TETRA-SACCHARIDES HAVING AN N-ACETYLGLUCOSAMINE RESIDUE AT THE REDUCING END. *Carbohydrate Research* **1994,** *255*, 61-85.

42. Sokolov, V. M.; Zakharov, V. I.; Studentsov, E. P., Stereoselectivity of reactions at the glycoside center of carbohydrates: VII. Synthesis of aryl alpha- and beta-D-glucopyranosides by Helferich, catalyzed by boron trifluoride etherate. *Russian Journal of General Chemistry* **2002,** *72* (5), 806-811.

43. Schmidt, R. R., New approaches to glycoconjugate synthesis. *Pure and Applied Chemistry* **1998,** *70* (2), 397-402.

44. Boltje, T. J.; Li, C. X.; Boons, G. J., Versatile Set of Orthogonal Protecting Groups for the Preparation of Highly Branched Oligosaccharides. *Organic Letters* **2010,** *12* (20), 4636-4639.

45. Boltje, T. J.; Zhong, W.; Park, J.; Wolfert, M. A.; Chen, W. X.; Boons, G. J., Chemical Synthesis and Immunological Evaluation of the Inner Core Oligosaccharide of Francisella tularensis. *Journal of the American Chemical Society* **2012,** *134* (34), 14255-14262.

46. Zhu, X. M.; Schmidt, R. R., New Principles for Glycoside-Bond Formation. *Angewandte Chemie-International Edition* **2009,** *48* (11), 1900-1934.

47. Kapoor, V. K.; Kaur, A., Drug-Glycosidation and Drug Development. *Mini-Reviews in Medicinal Chemistry* **2013,** *13* (4), 584-596.

48. Zhang, X. R.; Zhang, C. Q., Nano n-Propylsulfonated Magnetic -Fe2O3 as an Efficient and Reusable Catalyst for the Synthesis O-Isopropylidene Derivatives of Carbohydrates. *Journal of Carbohydrate Chemistry* **2013,** *32* (4), 249-258.

49. Miermont, A.; Zeng, Y. L.; Jing, Y. Q.; Ye, X. S.; Huang, X. F., Syntheses of lewis(x) and dimeric lewis(x): Construction of branched oligosaccharides by a combination of preactivation and reactivity based chemoselective one-pot Glycosylations. *Journal of Organic Chemistry* **2007,** *72* (23), 8958-8961.

50. de Jong, A. R.; Volbeda, A. G.; Hagen, B.; van den Elst, H.; Overkleeft, H. S.; van der Marel, G. A.; Codee, J. D. C., A Second-Generation Tandem Ring-Closing Metathesis Cleavable Linker for Solid-Phase Oligosaccharide Synthesis. *European Journal of Organic Chemistry* **2013,** *2013* (29), 6644-6655.

51. Coste, G.; Gerber-Lemaire, S., Efficient synthesis of cyclic glycolipid analogues. *Synlett* **2007,** (7), 1121-1123.

52. Fu, G. C.; Grubbs, R. H., THE APPLICATION OF CATALYTIC RING-CLOSING OLEFIN METATHESIS TO THE SYNTHESIS OF UNSATURATED OXYGEN HETEROCYCLES. *Journal of the American Chemical Society* **1992,** *114* (13), 5426-5427.

53. Fu, G. C.; Grubbs, R. H., SYNTHESIS OF NITROGEN-HETEROCYCLES VIA CATALYTIC RING-CLOSING METATHESIS OF DIENES. *Journal of the American Chemical Society* **1992,** *114* (18), 7324-7325.

54. Garber, S. B.; Kingsbury, J. S.; Gray, B. L.; Hoveyda, A. H., Efficient and recyclable monomeric and dendritic Ru-based metathesis catalysts. *Journal of the American Chemical Society* **2000,** *122* (34), 8168-8179.

55. Gessler, S.; Randl, S.; Blechert, S., Synthesis and metathesis reactions of a phosphine-free dihydroimidazole carbene ruthenium complex. *Tetrahedron Letters* **2000,** *41* (51), 9973-9976.

56. Cheng, Y. A.; Chen, T.; Tan, C. K.; Heng, J. J.; Yeung, Y. Y., Efficient Medium Ring Size Bromolactonization Using a Sulfur-Based Zwitterionic Organocatalyst. *Journal of the American Chemical Society* **2012,** *134* (40), 16492-16495.

57. Padwa, A.; Kline, D. N.; Norman, B. H., SYNTHESIS OF THE BENZAZEPIN-4-ONE RING-SYSTEM VIA DIPOLAR CYCLO-ADDITION OF N-PHENYLNITRONES WITH ACTIVATED ALLENES. *Journal of Organic Chemistry* **1989,** *54* (4), 810-817.

58. El-Faham, A.; Albericio, F., Peptide Coupling Reagents, More than a Letter Soup. *Chemical Reviews* **2011,** *111* (11), 6557-6602.

59. Grafton, M. W.; Farrugia, L. J.; Sutherland, A., Synthesis of Amino-Substituted Indanes and Tetralins via Consecutive Multibond-Forming Tandem Processes. *Journal of Organic Chemistry* **2013,** *78* (14), 7199-7207.

60. Blanchette, M. A.; Choy, W.; Davis, J. T.; Essenfeld, A. P.; Masamune, S.; Roush, W. R.; Sakai, T., HORNER-WADSWORTH-EMMONS REACTION - USE OF LITHIUM-CHLORIDE AND AN AMINE FOR BASE SENSITIVE COMPOUNDS. *Tetrahedron Letters* **1984,** *25* (21), 2183-2186.

61. Ireland, R. E.; Norbeck, D. W., APPLICATION OF THE SWERN OXIDATION TO THE MANIPULATION OF HIGHLY REACTIVE CARBONYL-COMPOUNDS. *Journal of Organic Chemistry* **1985,** *50* (12), 2198-2200.

62. Jimenez, J.; Ramirez, J.; Huelgas, G.; Melendrez, R.; Cabrera-Vivas, B. M.; Sansinenea, E.; Ortiz, A., 'Syn-effect' in the diastereoselective alkylation of 3- (E)-alpha,beta-unsaturated-gamma-substituted -N-acyloxazolidinones. *Tetrahedron* **2015,** *71* (28), 4590-4597.

63. Pereda-Miranda, R.; Rosas-Ramirez, D.; Castaneda-Gomez, J., Resin glycosides from the morning glory family. *Prog. Chem. Org. Nat. Prod.* **2010,** *92*, 77-153.

64. Ono, M., Resin glycosides from Convolvulaceae plants. *Journal of Natural Medicines* **2017,** *71* (4), 591-604.

65. Patani, G. A.; LaVoie, E. J., Bioisosterism: A rational approach in drug design. *Chemical Reviews* **1996,** *96* (8), 3147-3176.

66. Lusher, S. J.; McGuire, R.; van Schaik, R. C.; Nicholson, C. D.; de Vlieg, J., Data-driven medicinal chemistry in the era of big data. *Drug Discovery Today* **2014,** *19* (7), 859-868.

67. Seddon, M. P.; Cosgrove, D. A.; Gillet, V. J., Bioisosteric Replacements Extracted from High-Quality Structures in the Protein Databank. *Chemmedchem* **2018,** *13* (6), 607-613.

68. Gulten, S., The Synthesis and Characterization of Solvatochromic Maleimide-Fused N-Allyl- and N-Alkyl-Substituted 1,4-Dithiines and Diels-Alder Reactions with Anthracene. *Journal of Heterocyclic Chemistry* **2010,** *47* (1), 188-193.

69. Shuler, S. A.; Yin, G. Y.; Krause, S. B.; Vesper, C. M.; Watson, D. A., Synthesis of Secondary Unsaturated Lactams via an Aza-Heck Reaction. *Journal of the American Chemical Society* **2016,** *138* (42), 13830-13833.

70. Zong, G. H.; Whisenhunt, L.; Hu, Z. J.; Shi, W. Q., Synergistic Contribution of Tiglate and Cinnamate to Cytotoxicity of Ipomoeassin F. *Journal of Organic Chemistry* **2017,** *82* (9), 4977-4985.

71. Borszeky, K.; Mallat, T.; Baiker, A., Enantioselective hydrogenation of alpha,beta-unsaturated acids. Substrate-modifier interaction over cinchonidine modified Pd/Al2O3. *Tetrahedron-Asymmetry* **1997,** *8* (22), 3745-3753.

72. Shu, X. Z.; Huang, S. Y.; Shu, D. X.; Guzei, I. A.; Tang, W. P., Interception of a Rautenstrauch Intermediate by Alkynes for 5+2 Cycloaddition: Rhodium-Catalyzed

Cycloisomerization of 3-Acyloxy-4-ene-1,9-diynes to Bicyclo 5.3.0 decatrienes. *Angewandte Chemie-International Edition* **2011,** *50* (35), 8153-8156.

73. Andrus, M. B.; Turner, T. M.; Sauna, Z. E.; Ambudkar, S. V., The synthesis and evaluation of a solution phase indexed combinatorial library of non-natural polyenes for reversal of p-glycoprotein mediated multidrug resistance. *Journal of Organic Chemistry* **2000,** *65* (16), 4973-4983.

74. Chen, Y. G.; Shuai, B.; Ma, C.; Zhang, X. J.; Fang, P.; Mei, T. S., Regioselective Ni-Catalyzed Carboxylation of Allylic and Propargylic Alcohols with Carbon Dioxide. *Organic Letters* **2017,** *19* (11), 2969-2972.

75. Phillips, D. J.; Pillinger, K. S.; Li, W.; Taylor, A. E.; Graham, A. E., Diol desymmetrization as an approach to the synthesis of unsymmetrical dienyl diesters. *Tetrahedron* **2007,** *63* (42), 10528-10533.

76. Zhang, Y. L.; Knapp, S., Glycosylation of Nucleosides. *Journal of Organic Chemistry* **2016,** *81* (6), 2228-2242.

77. Lecourt, C.; Dhambri, S.; Allievi, L.; Sanogo, Y.; Zeghbib, N.; Ben Othman, R.; Lannou, M. I.; Sorin, G.; Ardisson, J., Natural products and ring-closing metathesis: synthesis of sterically congested olefins. *Natural Product Reports* **2018,** *35* (1), 105-124.

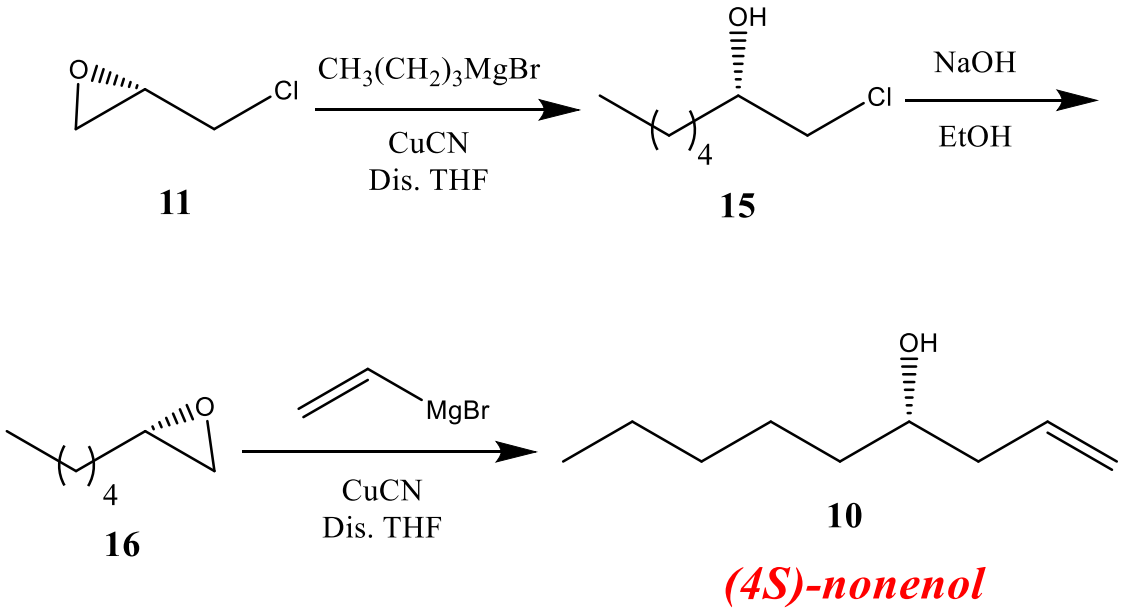
CHAPTER 3. EXPERIENTIAL PROCEDURE

3.1. General Methods

Reactions were carried out in oven-dried glassware. All reagents were purchased from commercial sources and were used without further purification unless specified. Except stated otherwise, all reactions were carried out under a nitrogen atmosphere and monitored by thin layer chromatography (TLC) using Silica Gel GF_{254} plates (Agela) with detection by charring with 5% (v/v) H_2SO_4 in EtOH or by visualizing in UV light (254 nm). Column chromatography was performed on silica gel (230–450 mesh, Sorbent). The ratio between silica gel and crude product ranged from 100 to 50:1 (w/w). NMR data were collected on a Bruker 300 or 400 MHz NMR spectrometer and a Bruker 300 or 400 MHz system. ^1H NMR spectra were obtained in deuterochloroform ($CDCl_3$) with chloroform ($CHCl_3$, $\delta_H = 7.27$ for ^1H) as an internal reference. ^{13}C NMR spectra were proton decoupled and were in $CDCl_3$ with $CHCl_3$ ($\delta_C = 77.0$ for ^{13}C) as an internal reference. Chemical shifts are reported in ppm (δ). Data are presented in the form: chemical shift (multiplicity, coupling constants, and integration). ^1H data are reported as though they were first order. The errors between the coupling constants for two coupled protons were less than 0.5 Hz, and the average number was reported. Proton assignments, when made, were done so with the aid of COSY NMR spectra. For some compounds, HSQC and HMBC NMR were also applied to assign the proton signals. Optical rotations were measured on an Autopol III Automatic Polarimeter at 25 ± 1 ^oC for solutions in a 1.0 dm cell. High resolution mass spectrum (HRMS) and were acquired in the ESI mode.

3.2. Synthesis of the Aglycone of Ipomoeassin F

3.2.1. Preparation of the (4S)-nonen-4-ol



(2*S*)-1-chloro-2-heptanol **15**. A mixture of Mg (4.08 g, 144.4 mmol), cat I_2 and Dry THF (120 mL) were heated to reflux (68-70 ^oC). While heating 0.5 mL of bromo-butane was added to the mixture. One the refluxing temperature was reached, the bromo-butane (17.4 mL, 144.4 mmol) was added dropwise (bubbling N_2 atmosphere) to the mixture. After all the Mg had been converted to the Grignard reagent the solution was cooled to RT. The butyl magnesium bromide was added dropwise to a solution of (*S*)-epichlorohydrin **11** (10.16 g, 130.3 mmol), CuCN (1.17 g, 116.0 mmol) at -78 ^oC over 1 hr. The reaction was slowly warmed to -20 ^oC over 3-4 hrs (green to black prep.). The reaction was quenched with sat. aq. NH_4Cl (200 mL) and the solvent removed under reduced pressure. The organic layer was extracted with Et_2O (250 mL · 2), dried over $NaSO_4$, filtered, and solvent carefully removed under reduce pressure. The α-chloro alcohol **15** was taken to the next step without further purification. ^1H NMR (300 MHz, $CDCl_3$) δ_H 3.80 (m, 1H, 2 C*H*),

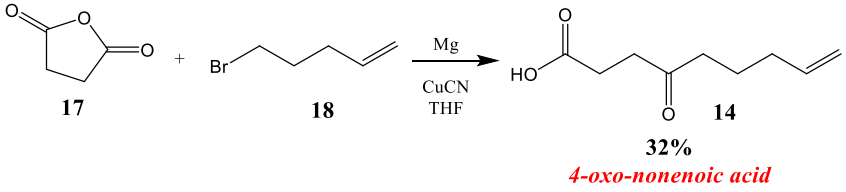
3.49 (dd, *J* = 6.12 Hz, 1H Cl-C*H*$_2$), 2.27 (m, 1H), 1.52 (m, 2H), 1.31 (m, 6H), 0.89 (t, *J* = 6 Hz, 3H, –C*H*$_3$). ^{13}C NMR ($CDCl_3$, 75 MHz) δ_C 71.5 (–CH_2), 50.6 (–CH_2), 34.3 (–CH_2), 31.7 (–CH_2), 25.2 (–CH_2), 22.6 (–CH_2), 14.0 (–CH_3). The ^{1}H and ^{13}C NMR were identicatial to published spectra data.[17]

2-pentyl-(2*S*)-oxirane **16**. NaOH (36.0 g, 361.2 mmol) was added to a solution of the crude *α*-chloro alcohol **15** (18.14 g, 120.4 mmol) at RT and allowed stirred for 4-5 hrs. The reaction was monitored by TLC (silica, Hex-EtOAc 4:1, R_f = 0.74). Upon completion the reaction poured into ice water and extracted with Et_2O (200 mL · 3). The organic layers were combined, dried over $NaSO_4$, filtered, and the Et_2O was removed under reduced pressure. The epoxide **16** was used direct in the next step. ^{1}H NMR ($CDCl_3$, 400 MHz) δ_H 2.94–2.88 (m, 1 H), 2.75 (dd, *J* = 5.0, 4.0, Hz, 1H), 2.46 (dd, *J* = 5.0, 2.8 Hz, 1H), 1.56–1.28 (m, 8H), 0.90 (m, 3H, –C*H*$_3$).^{13}C NMR (100 MHz, $CDCl_3$) δ_C 195.6 (O–C–O), 64.2 (O–CH_2), 32.2(–CH_2), 31.3 (–CH_2), 25.4 (–CH_2), 22.6 (–CH_2), 14.1 (–CH_3). The spectroscopic data matched previously published literature data.[18, 19]

(4*S*)-nonenol **10**. Vinylmagnesium bromide (175 mL, 109.4 mmol) was added dropwise at -78 ^{o}C to a solution of the crude epoxide **16** (9.61 g, 66.6 mmol), CuCN (845 mg, 66.6 mmol), and Dry THF (200 mL). The reaction was kept at -78 ^{o}C for 0.5 hrs and slowly warmed to 0 ^{o}C over 3-4 hrs. The reaction was quenched with sat. aq. NH_4Cl (150 mL) and the THF removed under reduce pressure. Th organic layer was extracted with Et_2O (225 mL · 3) and the combined organic layers washed with BRINE (200 mL). The organic layer was dried over $NaSO_4$, filtered, and solvent removed under reduced pressure. The oil was purified by column chromatography (silica, Hex-EtOAc, 10:1 → 6:1) to give the alcohol **10** in 64% (12.12 g) from (*S*)-epichlorohydrin. ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 5.84 (dd, *J* =17.2, 10.2, 7.1 Hz, 1H, *H*C=), 5.19–5.09 (m, 2H,

=C*H*$_2$), 3.65 (br, s, 1H, C–O*H*), 2.37–2.25 (m, 1H, *H*C–O), 2.20–2.07 (m, 2H), 1.67–1.56 (m, 2H), 1.53–1.21(m, 8H), 0.89 (t, *J* = 6.6 Hz, 3H, –C*H*$_3$). ^{13}C NMR δ_C (75 MHz, $CDCl_3$) δ_C 134.9 (=CH), 118.0 (H_2C=), 70.7 (C–H), 41.9 (–CH_2), 36.8 (–CH_2), 31.8 (–CH_2), 25.3 (–CH_2), 22.6 (–CH_2), 14.0 (–CH_3). The spectroscopic data matched those previously reported.[20-22]

3.2.2. Synthesis of 4-oxo-8-nonenoic acid

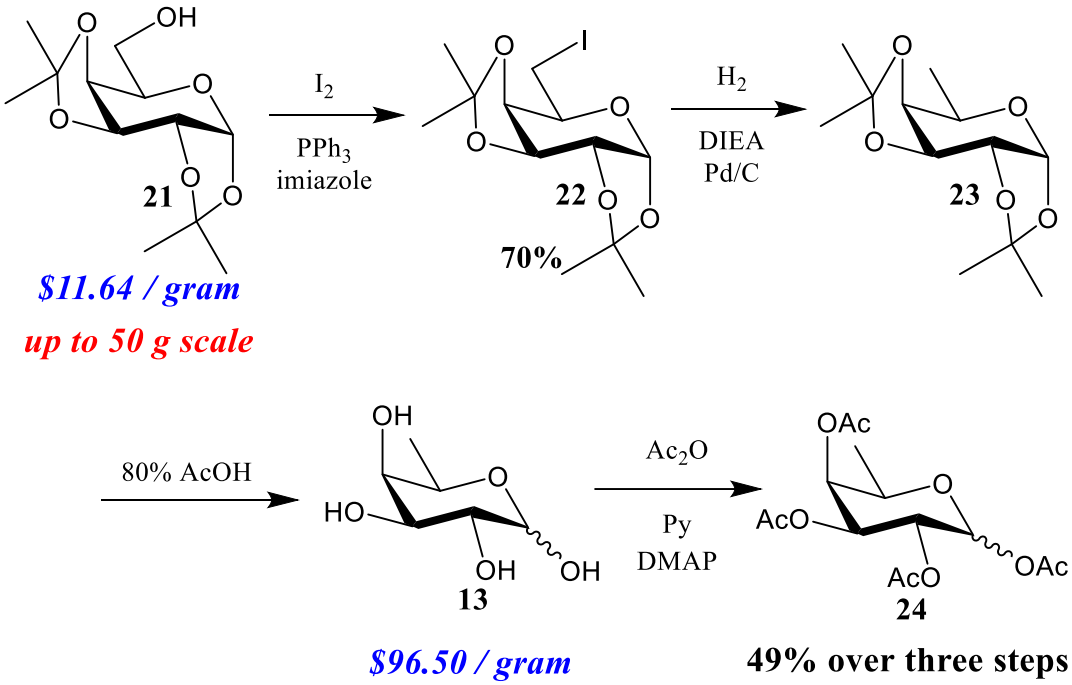


4-oxo-8-nonenoic acid **14**. Mg (1.64 g, 6.8 mmol), I_2 (trace), and, 5-bromopentene **18** (few drops, initiate the Grignard reaction) Dry THF (100 mL) were combined, then heated to reflux under a N_2 atmosphere. Then, 5-bromopentene **18** (8.1 mL, 67.1 mmol) was added to the reaction mixture dropwise over 30 mins and continued to heat at reflux for 2 hrs (trace amount of Mg). The Grignard formation was cooled to RT, while succinic anhydride (7.35 g, 73.8 mmol) in THF (60 mL) cooled to 0^{o}C. After cooling to RT, the Grignard reagent was added dropwise (1 hr.) to the SA **17** and THF, then slowly warmed to RT overnight. TLC showed the reaction complete (Hex:EtOAc:AcOH, 79.5:20:0.5). 1 M HCl (75 mL) quenched the reaction and the mixture was stirred for 10 mins. Then, the THF was removed under reduced pressure. The aqueous layer was the extracted with DCM (75 mL · 2) then acidified with contc. HCl (pH =2-3). The aqueous layer was extracted with DCM (40 mL · 3). The combined organic layers were dried with $NaSO_4$, filtered, and removed under reduced pressure. Purification by column chromatography gave the acid **14** in 32% (silica, Hex–EtOAc–AcOH 100:0:0→75:25:0.5). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.81-5.71 (m, 1H), 5.04-4.97 (m, 2H), 2.72 (t, 2H, *J* = 6.4 Hz), 2.63 (t, 2H, *J* = 6.4 Hz), 2.46 (t,

2H, *J* = 7.2), 2.05 (t, 2H, *J* = 6.0 Hz), 1.70 (app. qui., 2H). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 208.7 (C=O), 178.1, 137.8 (C=C), 115.9 (C=C), 41.7, 36.8, 32.9, 27.7, 24.7. The ^{1}H and ^{13}C NMR data were in accordance with the literature.[23-25]

3.3. Preparation of the Fucosyl Acceptor

3.3.1. Preparation of the D-Fucose from Diisopropylidene -Galactose



1,2:3,4-bis-*O*-(1-methylethylidene)-6-deoxy-6-iodo-*α*-D-galactopyranose **22**. Toluene (500 mL) was added to flask of the diisopropylidene-galactose **21** (52.28 g, 200.8 mmol), PPh_3 (63.21 g, 241.0 mmol), and imidazole (49.22 g, 723.1 mmol) at RT. Then, I_2 (63.21 g, 241.0 mmol) was added to the reaction mixture and allowed to stir at 80 °C overnight as the reaction turned a dark brown color. After completion of the reaction (silica, Hex-EtOAc 2:1, R_f = 0.76), the reaction mixture was quenched with MeOH (10 mL), and the resulting solution was concentrated to a residue. The reaction mixture was filtered through a pad of Celite and $Na_2S_2O_3$ added until the

slurry turned a almost clear (yellow). The aqueous layer was extracted with DCM (400 mL·2), dried over $NaSO_4$, and the solvent evaporated under reduced pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc 5:1→3:1) to give **22** in 70% (52.47 g). ^{1}H NMR ($CDCl_3$, 400 MHz) δ_H 5.54 (d, *J* = 5.1 Hz, 1H, 1-H-Glc*p*), 4.61 (dd, *J* = 7.9, 2.5 Hz, 1H), 4.41 (dd, *J* = 7.9, 1.8 Hz, 1H), 4.30 (dd, *J* = 5.1, 2.5 Hz, 1H), 3.95 (td, *J* = 7.0, 1.8 Hz, 1H), 3.32 (dd, *J* = 10.1, 6.9 Hz, 1H), 3.20 (dd, *J* = 10.1, 7.2 Hz, 1H), 1.55 (s, 3H, –C*H*$_3$), 1.45 (s, 3H, –C*H*$_3$), 1.36 (s, 3H, –C*H*$_3$), 1.37 (s, 3H, –C*H*$_3$), 1.33 (s, 3H, –C*H*$_3$); ^{13}C NMR ($CDCl_3$, 100 MHz) δ_C 109.6 (O–C–O), 108.8 (O–C–O), 96.8 (C-1), 71.7, 71.1, 70.6, 69.1, 26.1 (–CH_3), 26.0 (–CH_3), 25.1 (–CH_3), 24.6 (–CH_3). The ^{1}H and ^{13}C-NMR data matched previously published reports.[33, 34]

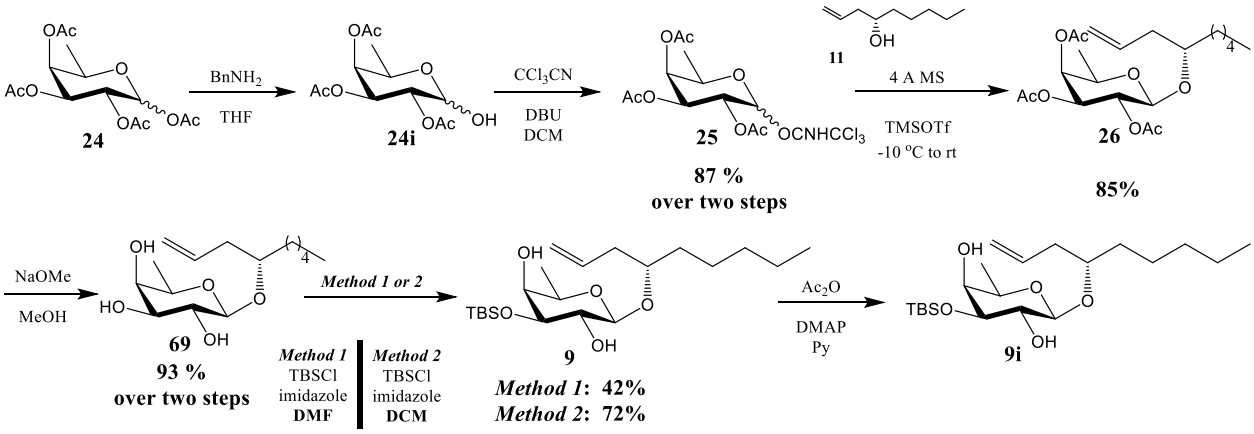
1,2:3,4-bis-*O*-(1-methylethylidene)-6-deoxy-*α*-D-galactopyranose **23**. Pd/C (10 g) was added to a solution of the iodo intermediate **22** (31.23 g, 84.3 mmol), DIEA (21.9 mL, 126.5 mmol), and MeOH (150 mL). Under a H_2 atmosphere, the pressure was increased to 80 pounds per square inch (PSI) and allowed to stir at RT overnight. TLC (silica, Hex-EtOAc 2:1) showed the reaction was complete and the reaction was quenched with NEt_3 and filtered through a pad of Celite. The crude 6-deoxy-Gat **23** was used directly in the next step without further purification. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.57 (d, *J* = 5.2 Hz, 1H, H-1-Fuc*p*), 5.48 (dd, *J* = 8.1 Hz, 2.8 Hz, 1H, H-3-Fuc*p*), 4.26 (dd, *J* = 5.3 Hz, 2.4 Hz, 1H, H-2-Fuc*p*), 4.06 (dd, *J* = 1.6 Hz, 8.0 Hz, 1H, H-4-Fuc*p*), 3.89 (ddd, 1H, H-5-Fuc*p*), 1.50 (s, 3H, –C*H*$_3$), 1.44 (s, 3H, –C*H*$_3$), 1.33 (s, 3H, –C*H*$_3$), 1.30 (s, 3H, –C*H*$_3$), 1.23 (d, *J* = 6.4 Hz, 3H, –C*H*$_3$). ^{13}C NMR (75 MHz, CDCl3) δ_C 109.0, 108.3, 96.6, 73.5, 70.9, 70.4, 63.5, 26.0 • 2, 24.9, 24.4, 15.9. The spectroscopic data matched those previously reported.[35, 36]

6-deoxy-*α*,*β*-D-galactopyranose **13**. The crude 6-deoxy-gataltose intermediate **23** (44 g) was dissolved in 80% acetic acid (400 mL) and heated to reflux (125 ^{o}C). The reaction was allowed to stir at reflux overnight, when TLC (silica, Hex-EtOAc 6:1, R_f = 0.11) indicated the reaction was finished. The reaction was cooled, then co-evaporated with Tol (200 mL· 3). The slurry was dissolved in sat. aq. $NaHCO_3$ (300 mL) and extracted with DCM (300 mL · 3). The combined organic layers were dried over $NaSO_4$, filtered, and solvent removed under reduce pressure. The crude D-Fuc*p* **13** was used directly in the next step without further purification. ^{1}H NMR (400 MHz, D_2O) δ_H 5.05 (d, *J* = 3.8 Hz, 1H, 1-H-Fuc*p*), 4.41 (d, *J* = 7.9 Hz, 1H, H-1-Fuc*p*), 4.06 (q, *J* = 6.7 Hz, 1H, H-5-Fuc*p*), 3.74-3.64 (m, 4H, H-2-Fuc*p*, H-3-Fuc*p*, H-4-Fuc*p*, H-5-Fuc*p*), 3.59 (d, *J* = 2.8, 4-OH-Fuc*p*), 3.50 (dd, *J* = 10.1 Hz, 3.4 Hz, 3-OH-Fuc*p*), 3.31 (dd, *J* = 10.1 Hz, 7.9 Hz, 2-OH-Fuc*p*), 1.10 (d, *J* = 6.3 Hz, 3H, H-6-Fuc*p*), 1.07 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*).[37, 38]

1,2,3,4-*O*-tetracetate-6-deoxy-*α*,*β*-D-galactopyranose **24**. The crude D-Fuc*p* **13** (32.1 g, 195 mmol) was dissolved in acetic anhydride (75 mL), Py (140 mL), cat DMAP. The reaction was allowed to stir at RT overnight, while turning a deep orange color. TLC (silica, Hex-EtOAc 2:1, R_f = 0.61) showed the reaction to be complete and the Py/AA was co-evaporated with Tol (200 · 3). The slurry was dissolved in sat. aq. $NaHCO_3$ (300 mL) and extracted with DCM (300 mL · 3). The combined organic layers were dried over $NaSO_4$, filtered, and solvent removed under reduce pressure. The residue was purified by column chromatography (silica, Hex-EtOAc, 4:1→3:1) to the peracetylated D-Fuc*p* **66** in 48% three steps from **13**. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 6.30 (d, *J* = 3.0 Hz, 1H), 5.65 (d, *J* = 8.5 Hz, 1H), 5.30-5.23 (m, 5H), 5.05 (dd, *J* = 10.5, 3.5 Hz, 1H), 4.24 (q, *J* = 6.5 Hz, 1H), 3.93 (q, *J* = 6.5 Hz, 1H), 2.15-1.95 (series of s, 3H, –CH_3), 1.19 (d, *J* = 6.5 Hz, 3H, –CH_3), 1.12 (d, *J* = 6.5 Hz, 3H, –CH_3). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 170.48, 170.46, 170.11, 169.94, 169.90, 169.4, 169.1, 166.4, 92.2, 89.9, 71.3, 70.6, 70.3, 70.0, 68.0, 67.9,

67.3, 66.5, 22.1, 20.9, 20.8, 20.63, 20.60, 20.57, 20.53, 15.92, 15.91. The ^{1}H- and ^{13}C-NMR were identical to previous published data.[39-41]

3.3.2. Synthesis of the Fucosyl Acceptor from D-Fucose



2,3,4-*O*-triacetate-6-deoxy-*α*,*β*-D-galactopyranose **24i**. The peracetylated D-Fuc*p* **24** (10.55 g, 31.7 mmol) was dissolved in Dry THF (150 mL) and $BnNH_2$ (4.6 mL, 47.6 mmol). The reaction was stirred at RT overnight and TLC showed the reaction to be complete (silica, Hex-EtOAc 1:1, R_f = 0.54, 0.49). The THF was removed under reduced pressure, the slurry diluted with DCM (200 mL), washed with 5% HCl, then sat. aq. $NaHCO_3$. The organic layer was collected, dried over $NaSO_4$, filtered, and DCM removed under reduced pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc, 2:1→1:2) to give the *α*,*β*-hemiacetal D-fucose **24i** in 77% (7.15 g) and carried directly to the next step.[42]

1-(2,2,2-trichloroethanimidate)-2,3,4-triacetate-6-deoxy-*α*,*β*-D-galactopyranose **25**. Trichloroacetonitrile (12.8 mL, 128 mmol) and DBU (0.68 mL, 3.2 mmol) were added to a solution of DCM (100 mL) and the hemiacetal **24i** (9.30 g, 32.0 mmol) at RT. The reaction was stirred at RT for 3-4 hrs, when TLC (silica, Hex-EtOAc 1:1, R_f = 0.71 and 0.77) indicated starting had been used up. The reaction was quenched with NEt_3 (0.5 mL) and the DCM reduced under reduced

pressure. The residue was purified by column chromatography (silica, Hex-EtOAc, 4:1→1:1, 0.1% NEt_3) to give the *α,β*-Schmidt donor **25** in 86% (10.6 g). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 8.64 (s, 1H), 6.59 (d, *J* = 3.5 Hz, 1H), 5.51-5.35 (m, 3 H), 4.43-4.37 (br q, J= 6.6 Hz, 1 H), 2.22, 2.05, and 2.04 (3 s, 3H), 1.15 (d, *J* = 6.6 Hz, 3 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 170.4-170.0 · 3, 161.1, 93.9, 77.2, 70.5, 67.9, 66.9, 67.5, 20.7-20.6 · 3, 15.9. The ^{1}H and ^{13}C NMR data were identical to the published literature data.[43, 44]

((1*S*)-1-(2-propen-1-yl))hexyl-,2,3,4-*O*-triacetate-6-deoxy-*β*-D-galactopyranoside **26**. The Schmidt donor **25** (11.6 g, 26.7 mmol), (4*S*)-nonenol **11** (3.8 g, 24.0 mmol), and 4 Å MS (11 g) were dissolved in Dis. DCM (100 mL) and cooled to −10 °C over 30 mins. TMSOT*f* (400 *μ*L, 2.7 mmol) was slowly added to the mixture and the reaction mixture was slowly warmed to RT and was stirred for 1-3 hrs. TLC (silica, Hex-EtOAc, 3:1, R_f = 0.59) indicated that the all the acceptor had been consumed. The reaction mixture was quenched with NEt_3 (0.5 mL) and filtrated through a pad of Celite. The filtrate was under reduced pressure to give a residue, then purified by column chromatography (silica, Hex:EtOAc, 8:1 → 4:1) to give intermediate **26** in a 85% (9.4 g) as a yellowish syrup. $[\alpha]^{25}_D$ −9.4 (*c* 1 $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.82−5.70 (m, 1H, CH_2=C*H*CH_2−), 5.23−5.11 (m, 2H, H-4-Fuc*p*, H-2-Fuc*p*), 5.08−4.96 (m, 3H, H-3-Fuc*p*, C*H*$_2$=CHCH_2−), 4.47 (d, *J* = 7.7 Hz, 1H, H-1-Fuc*p*), 3.80−3.73 (m, 1H, H-5-Fuc*p*), 3.65−3.58 (m, 1H, −CH_2C*H*CH_2−), 2.25−2.20 (m, 2H), 2.16 (s, 3H, C*H*$_3$C=O), 2.02 (s, 3H, C*H*$_3$C=O), 1.98 (s, 3H, C*H*$_3$C=O), 1.63−1.46 (m, 2H), 1.39−1.22 (m, 6H), 1.20 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.88 (t, *J* = 6.7 Hz, 3H, –C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 170.9 170.3, 169.4, 134.4, 117.0, 100.4, 80.2, 71.3, 70.3, 69.3, 68.9, 38.4, 34.4, 31.7, 24.1, 22.5, 20.9, 20.7, 20.6, 16.1, 14.0. The spectroscopic data were in accordance with the literature reports.[45, 46]

[(1*S*)-1-(2-propen-1-yl)]hexyl-6-deoxy-*β*-D-galactopyranoside **27**. NaOMe (cat) was added to a solution of Fuc*p* **26** (12.2 g, 29.4 mmol) and MeOH (150 mL) at RT. The reaction was stirred at RT overnight, while being monitored by TLC (silica, Hex-EtOac, 1:1 R_f = 0.15). The reaction was neutralized with Amberlite IR-120 (H) ion exchange resin and filtered. The solvent was removed under reduced pressure. The residue was purified by column chromatography (silica, Hex-EtOAc, 2:1→1:1) to give the triol **27** in 85% (7.26 g). $[\alpha]^{25}_D$ −21.2 (*c* $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.91−5.78 (m, 1H, CH_2=$CHCH_2$−), 5.20−5.02 (m, 2H, CH_2=$CHCH_2$−), 4.24 (d, *J* = 7.6 Hz, 1H, H-1-Fuc*p*), 3.74−3.53 (m, 5H, H-2-Fuc*p*, H-3-Fuc*p*, H-4-Fuc*p*, H-5-Fuc*p*, −CH_2CHCH_2−), 3.36 (d, *J* = 4.8 Hz, 1H, –O*H*), 2.95 (br, 1H, –O*H*), 2.74 (d, *J* = 5.6 Hz, 1H, -OH), 2.40−2.22 (m, 2H), 1.67−1.48 (m, 2H), 1.42−1.21 (m, 9H), 0.89 (d, J = 6.9 Hz, 3H, H-6-Fuc*p*). ^{13}C NMR (100 MHz, CDCl3) δ_C 134.9, 117.7, 102.3 (C-1), 79.2, 73.9, 72.1, 71.6, 70.5, 38.6, 34.7, 31.8, 24.8, 22.6, 16.3, 14.1. The ^{1}H and ^{13}C NMR data were in accordance with the literature.[45,46]

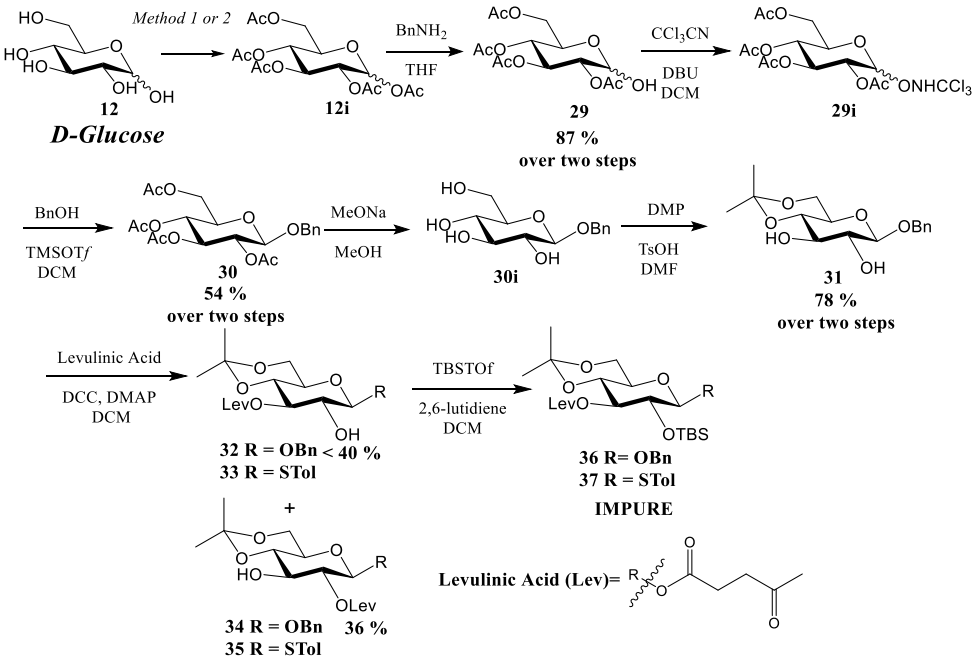
[(1*S*)-1-(2-propen-1-yl)]hexyl-6-deoxy-3-*O*-[(1,1-dimethylethyl)dimethylsilyl]-*β*-D-galactopyranoside **9**. *Method 1.* The triol **27** (3.37 g, 11.6 mmol) and the 1H-imidazole (2.38 g, 35.0 mmol) was dissolved in the minimal amount of the DMF (35 mL). After the reaction was cooled to -10 ^{o}C, TBSCl (2.29 g, 15.1 mmol) was added to the reaction. The reaction was slowly warm to RT and stirred for additional 2-3 hrs. When the TLC (silica, Hex-EtOAc 6:1, R_f = 0.69) showed the reaction was starting to form by-product. The DMF was removed by reduced pressure. The slurry was diluted with ether (50 mL) and the reaction was washed with water (40 mL 2) and BRINE (35 mL). The combined organic layers were collected, dried over $NaSO_4$, filtered, and concentrated. The residue was purified by column chromatography (silica, Hex-EtOAc, 14:1→8:1) to give the Fuc*p* acceptor **9** in 42% (1.97 g). $[\alpha]^{25}_D$ −6.9 (*c* 1 $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.91−5.74 (m, 1H, CH_2=C*H*CH_2−), 5.14−4.98 (m, 2H, C*H*$_2$=$CHCH_2$−), 4.20 (d,

J = 7.8 Hz, 1H, H-1-Fuc*p*), 3.73−3.65 (m, 1H, −$CH_2C\mathit{H}CH_2$−), 3.63 (dd, *J* = 9.2, 3.6 Hz, 1H, H-3-Fuc*p*), 3.59−3.48 (m, 3H, H-2-Fuc*p*, H-4-Fuc*p*, H-5-Fuc*p*), 2.60 (br, 1H, −O*H*), 2.37−2.19 (m, 2H), 2.17 (d, *J* = 1.6 Hz, 1H, −O*H*), 1.67−1.47 (m, 2H), 1.39−1.22 (m, 9H), 0.97−0.84 (m, 12H), 0.15 (s, 3H, Si−CH_3), 0.13 (s, 3H, Si−CH_3). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 135.1, 117.4, 102.2, 78.9, 74.9, 72.1, 72.1, 70.0, 38.5, 34.7, 31.8, 25.8(3), 24.7, 22.6, 18.1, 16.4, 14.1, −4.3, −4.9; IR (film) ν = 3537, 2930, 2857, 1655, 1109, 1066, 997, 910, 839, 781; HRMS (ESI) m/z calcd. for $C_{21}H_{42}NaO_5Si$ $[M + Na]^+$ 425.2699, found 425.2698.

(1*S*)-1-(2-propen-1-yl)hexyl-6-deoxy-3-*O*-[(1,1-dimethylethyl)dimethylsilyl]-2,4-diacetate-*β*-D-galactopyranoside **9i**. The diol **9** (50 mg, 0.12 mmol) was dissolved in Py (4 mL, then acetic anhydride (5100 μL) and DMAP were added to the solution at RT. The reaction mixture was stirred at RT overnight, when TLC (silica, Hex-EtOAc) indicated that the reaction was complete. The mixture was concentrated under reduced pressure and then co-evaporated with toluene (2 · 20 mL) to give the crude product. The residue was purified by column chromatography (silica, Hex-EtOAc 10:1) to give the acylated **9i** (38 mg, 60%) as a colorless syrup. $[\alpha]^{25}_D$ −1.9 (c 1 $CHCl_3$).^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.84−5.69 (m, 1H, CH_2=C$\mathit{H}CH_2$−), 5.11−4.97 (m, 4H, CH_2=CHCH_2−, H-2-Fuc*p*, H-4-Fuc*p*), 4.38 (d, *J* = 8.0 Hz, 1H, H-1-Fuc*p*), 3.78 (dd, *J* = 9.6, 3.6 Hz, 1H, H-3-Fuc*p*), 3.70−3.63 (m, 1H, H-5- Fuc*p*), 3.61−3.56 (m, 1H, −$CH_2C\mathit{H}CH_2$−), 2.26−2.14 (m, 2H), 2.11 (s, 3H, CH_3C=O), 2.03 (s, 3H, CH_3C=O), 1.63−1.19 (m, 8H), 1.17 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.88 (t, *J* = 6.8 Hz, 3H, −CH_3), 0.83 (s, 6H), 0.07 (s, 3H, Si−CH_3), 0.06 (s, 3H, Si−CH_3). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 170.9, 169.2, 134.7, 116.9, 100.6, 79.9, 72.9, 72.3, 71.5, 69.0, 38.4, 34.4, 31.8, 25.4 · 3 , 24.7, 22.6, 21.3, 20.9, 17.8, 16.3, 14.1, −4.8, −5.1. IR (film) ν = 2928, 2855, 1744, 1371, 1231, 1119, 1063, 837, 781.

3.4. Preparation of the Glucosyl Donor

3.4.1. Selective introduction of the lev. group for the protected glucosyl donor



1,2,3,4,6-pentaacetate-α,β-D-glucopyranose **12i**. *Method 1.* Pyridine (60 mL), acetic anhydride (30mL) was added to D-Glc*p* **13** (30.0 g, 0.167 mol) and allowed to stir at RT overnight. At this point, TLC (silica, 1:1 Hex-EtOAc) showed the reaction was complete. The reaction was concentrated under reduced pressure and the residue was dissolved in sat. aq. $NaHCO_3$ (150 mL). The aq. layer was extracted twice with DCM (150 mL). The organic layers were combined, dried with $NaSO_4$, filtered, and concentrated under reduced pressure. The yellow syrup **12i** (65 g) was used in the following step without any further purification. (1:1 Hex-EtOAc, R_f = 0.55) ^{1}H NMR (300 MHz; $CDCl_3$) δ_H 6.29 (d, *J* = 3.2 Hz, 1 H, H-1$_\alpha$), 5.67 (d, *J* = 7.7 Hz, c0.4 H, H-1$_\beta$), 5.44-5.35 (dd, *J* = 10.2 and 10.0 Hz, 1 H, H-3$_\alpha$), 5.29-5.21 (m, 0.4 H, H-3$_\beta$), 5.17-5.04 (m, 2.8 H, H-2$_\alpha$, H-2$_\beta$, H-4$_\beta$, H-4$_\alpha$), 4.30-4.26 (m, 1.4 H, H-6$_\alpha$, H-6$_{\alpha\beta}$), 4.12-4.01 (m, 2.4 H, H-6$_\alpha$, H-6$_\beta$, H-5$_\alpha$), 3.84-3.79 (0.4 H, H-5$_\beta$), 2.19 (s, 4.2 H), 2.13 (s, 1.2 H), 2.09 (s, 4.2 H), 2.02 (s, 8.4 H). ^{13}C NMR (75

MHz, $CDCl_3$) δ_c 170.4, 170.1, 169.5, 169.3, 168.6, 92.0 (C-1β), 89.4 (C-1α), 73.2 (Cβ), 70.6 (Cβ), 70.2 (2Cα,β), 69.5 (Cα), 68.2 (Cα), 61.7 (Cα,β), 21.0, 21.0, 20.8, 20.7. Spectral data matched those previously reported. *Method 2*. D-Glc*p* **12** (20.65 g, 0.14 mol) was slowly added to a mixture of acetic anhydride (90 mL) and $HClO_4$ (0.7 mL) over 0.5-1 hr at 0 ^oC. The reaction was allowed to warm slowly over 30 mins as the reaction turned from yellow to clear color. TLC (silica, 2:1 Hex-EtOAc) showed the reaction to be complete. The reaction mixture was co-evaporated with Tol (150 mL) three times after quenching reaction. The mixture was dissolved in sat. aq. $NaHCO_3$ (100 mL) and extracted twice with DCM (150 mL). The organic layers were combined, dried with $NaSO_4$, filtered, and concentrated under reduced pressure. The ^1H and ^{13}C-NMR matched those of the *Method 1*.[1, 2]

2,3,4,6-tetraacetate-*α,β*-D-glucopyranose **29**. Benzylamine (4.56 mL, 47.6 mmol) was added at to a solution of peracetate D-Glc*p* **12i** (10.55 g, 31.7 mmol) and THF (150 mL) at RT and allowed to stir for 24 hrs. The THF was removed under reduced pressure, diluted with DCM (150 mL), washed with 5% aq. HCl (100 mL), washed with sat. aq. $NaHCO_3$ (100 mL). The organic layer was dried with $NaSO_4$, filtered, and concentrated under reduce pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc 2:1→1:1) to give the hemiacetate **29** (6.33 g, 21.8 mmol, *α*:*β* 1:4) in 68.7%. ^1H NMR (400 MHz, $CDCl_3$) δ_H 5.44 (t, 1H, *J* = 9.9 Hz, H3α), 5.35 (t, 1H, *J* = 3.5 Hz, H-1α), 5.14 (t, 1H, *J* = 9.8 Hz, Hβ), 4.98 (t, 2H, *J* = 9.9 Hz, H-4$\alpha+\beta$), 4.93 (d, 1H, *J* = 8.2 Hz, Hβ) 4.79 (dd, 2H, *J* = 10.1 Hz, 3.5 Hz), 4.20-4.14 (m, 2H), 4.07 (m, 2H), 3.66 (t, 2H, *J* = 6.6 Hz), 2.00 (6H), 2.00 (6H), 1.95(6H), 1.93 (6H).^{13}C NMR (100 MHz, $CDCl_3$): δ_C 171.0, 170.9, 170.3, 170.3, 170.3, 169.8, 169.6, (*β*): 95.1 (C-1), 72.9 (C-2), 72.5 (C-3), 71.8 (C-4), 68.4 (C-5), 67.9 (C-6) (*α*), 89.1 (C-1), 71.2 (C-2), 69.9 (C-3), 68.5 (C-4), 66.9 (C-5), 62.0 (C-

6), 25.49 (−CH_3), 20.7 (−CH_3), 20.7 (−CH_3), 20.6 (−CH_3), 20.6 (−CH_3), 20.5 (−CH_3). The ^{1}H- and ^{13}C-NMR data were identical to those previously reported.[3, 4]

1-(2,2,2-trichloroethanimidate)-2,3,4,6-tetraacetate-*α,β*-D-glucopyranose **29i**. CCl_3CN (6.42 mL, 62.8 mmol) and DBU (0.34 mL, 1.5 mmol) were are added to the mixture of the hemiacetal **29** (4.56 g, 15.7 mmol). The reaction was allowed to stir at RT for 3 hrs as the mixture turned an orange-brown color. The reaction was quenched with 1 mL of NEt_3 after the TLC (silica Hex-EtOAc 1:1 R_f = 0.65) showed the reaction to be complete. The mixture was quenched with NEt_3 (0.5 mL) concentrated under reduced pressure. The Schmidt donor **29i** was obtained in 85.6% (5.86g), after purification by column chromatography (silica, Hex-EtOAc 4:1→1:1, few drops of NEt_3). ^{1}H NMR (400 MHz, $CDCl_3$): δ_H 8.68 (s, 1H, C=N*H*), 6.51 (d, 1H, *J* = 3.9 Hz, H-1-Glc*p*), 5.51 (t, 1H, *J* = 9.9 Hz, H-4-Glc*p*), 5.13 (t, 1H, *J* = 9.9 Hz, H-3-Glc*p*), 5.09 (dd, 1H, *J* = 10.3 Hz, 3.7 Hz, H-2-Glc*p*), 4.24 (dd, 1H, *J* = 12.0, 3.8 Hz, H-6$_\alpha$-Glc*p*), 4.19-4.15 (m, 1H, H-5-Glc*p*) 4.09 (dd, 1H, *J* = 12.0 Hz, 1.8 Hz, H-6$_\beta$-Glc*p*), 2.02 (s, 3H, −CH_3), 2.00 (s, 3H, −CH_3), 1.98 (s, 3H, −CH_3), 1.97 (s, 3H, −CH_3). The ^{13}C-NMR was not obtained because of the stability of the donor. The ^{1}H-NMR spectra match previously reported data.[5, 6]

Benzyl-2,3,4,6-tetraacetate-*β*-D-glucopyranoside **30**. BnOH (4.2 mL, 40.2 mmol) and 4 Å MS (5 g) were added to a solution of the Schmidt donor **29i** and Dry DCM (200 mL). The reaction was cooled to 0 ^{o}C and TMSOT*f* (0.1 eq) was added dropwise to the solution. The reaction was allowed to slowly warm to RT overnight. The reaction was monitored by TLC (silica, Hex-EtOAc 2:1 R_f = 0.65) and showed the reaction went to 80-90% completion. After quenching the reaction with NEt_3, filtered through a pad of Celite using acetone as an eluent, and concentrated under reduced pressure. The residue was purified by column chromatography (silica Hex-EtOAc 6:1→

1:1) to give the *β*-benzyl glucoside **30** in 69% (6.078 g). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.28–7.45 (m, 5H, Ar−H), 5.25–5.01 (m, 3H), 4.91 (d, *J* = 6.8 Hz, 1H, 1-H-Glc*p*), 4.51–4.72 (m, 2H), 4.24–4.12 (m, 2H), 3.71–3.62 (d, *J* = 4.1 Hz, 1H), 2.09 (s, 3H, $-CH_3$), 2.05 (s, 3H, $-CH_3$), 2.03 (s, 3H, $-CH_3$), 2.02 (s, 3H, $-CH_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.80 (C=O), 170.50 (C=O), 169.6 (C=O), 169.4 (C=O), 136.3 (C=C), 129.7 (C=C), 129.8 · 2 (C=C), 129.9 · 2 (C=C), 128.3 (C=C), 98.8 (C-1), 77.1, 73.6, 73.1, 71.6, 71.6, 71.5, 69.0, 68.3, 68.0, 62.6, 62.5, 21.2 ($-CH_3$), 21.0 ($-CH_3$), 20.8 ($-CH_3$), 20.8 ($-CH_3$). The spectroscopic data matched previous literature reports.[5,6]

Benzyl-*β*-D-glucopyranoside **30i**. The *β*-benzyl glucoside **30** (2.27 g, mmol) was dissolved in the MeOH (15 mL) and cat NaOMe (35 mg, 30 mmol). The reaction was allowed to stir at RT overnight, when the TLC (silica, EtOAc: MeOH 10:1, R_f = 0.15) showed the reaction to me complete. The reaction was neutralized with Amberlite IR-120 (H), ion exchange resin and filtered. The solvent was removed under reduced pressure. The crude tetraol **30i** was re-crystallized in methanol and used directly in the next step. (400 MHz, CD_3OD) δ_H 7.33-7.17 (m, 5H, Ar-H), 4.88 (d, *J* = 11.8, 1H, H_2C-Ar), 4.57(d, *J* = 11.8, 1H, H_2C-Ar), 4.25 (d, *J* = 7.7, 1H, Hz, H-1-Glc*p*), 3.80 (dd, *J* = 11.9, 2.1 Hz, 1H, H-6-Glc*p*), 3.59 (dd, *J* = 11.9, 5.5 Hz, 1H, H-6-Glc*p*), 3.25-3.13 (m, 4H, H-2, H-3, H-4 & H-5); (100 MHz, $CDCl_3$) δ_C 137.8 (C=C), 128.8 · 2 (C=C), 128.5 · 2 (C=C), 128.2 (C=C), 102.3, (C-1) 76.7, 75.9, 73.8, 71.6, 69.7, 61.6. The spectral data matched those previously reported.[7, 8]

Benzyl-4,6-*O*-(1-methylethylidene)-*β*-D-glucopyranoside. **31**. The tetraol **30i** (1.54 g, 5.6 mmol) was dissolved in the minimum amount DMF and dimethoxypropane (2.8 mL, 22.8 mmol). Cat. *p*-TsOH (21 mg, 0.1 mmol) was added to the reaction mixture and allowed to stir for 36 hrs. The reaction was quenched with 0.2 mL of NEt_3 and co-evaporated with Tol 3 times. The reside

was purified by column chromatography to the give in the diol **31** (silica, Hex-EtOAc, 4:1→1:1, 0.1% NEt_3). (TLC silica, R_f = 0.63 Hex:EtOAc 6:1) ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.40-7.28 (m, 5H, Ar−H), 4.91 (d, *J* = 11.2 Hz, 1H, $-H_2C-Ar$), 4.61 (d, *J* = 11.6, 1H, $-H_2C-Ar$), 4.44 (d, *J* = 7.6, 1H, H-1-Glc*p*), 3.95 (dd, *J* = 8.4 3.2 Hz, 1H, H-6-Glc*p*), 3.82 (t, *J* = 10.4 Hz, 1H, H-6-Glc*p*), 3.67-3.57 (m, 2H, H-2-Glc*p*, H-3-Glc*p*), 3.50 (t, *J* = 8 Hz, 1H, H-4-Glc*p*), 3.30-3.26 (m, 1H, H-5-Glc*p*), 1.49 (s, 3H, $-CH_3$), 1.44 (s, 3H, $-CH_3$); δ_C (100 MHz, CDCl3) 136.4 (C=C), 128.3· 2 (C=C), 127.9· 2 (C=C), 127.9 (C=C), 101.9 (O-C-O), 99.5 (C-1), 74.41, 73.2, 72.9, 71.2, 67.1 (C-6), 61.8, 28.8 ($-CH_3$), 18.8 ($-CH_3$).[9, 10]

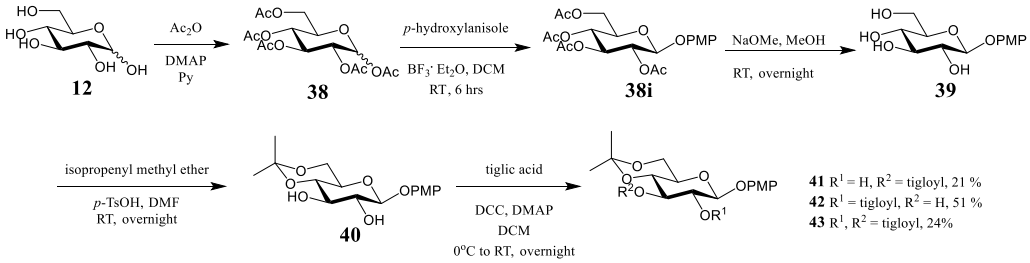
Benzyl-4,6-*O*-(1-methylethylidene)-2 or 3-levulinic-*β*-D-glucopyranoside. **32** or **34**. The solution of the acetal **31** (261 mg, 0.84 mmol), lev acid (110 mg, 0.93 mmol) DMAP (1.5 mg, 0.084 mmol), Dry DCM DCM (20 mL) was cooled to 0 ^oC. Then, DCC (191 mg, 0.93 mmol) was slowly added to the reaction mixture and allowed to slowly warm to RT overnight. TLC showed the reaction to be complete with a mixture of the two products (silica, Hex-EtOAc, 1:1 R_f = 0.65). The DCM was partially removed under reduced pressure, then diluted with Et_2O (40 mL) and Hex (20 mL) to precipitate any DCU. The mixture was filtered through a pad of Celite and the solvent removed under reduced pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc 4:1→1:1, NEt_3). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.29-7.20 (m, 5 H), 4.87 (t, *J* = 9.2 Hz, 1H) 4.79 (d, *J* = 12.0 Hz, $-CH_2$), 4.52 (d, *J* = 12.4 Hz, $-CH_2$), 3.89 (dd, *J* = 8.1 Hz, 2.9 Hz, 1H), 3.75 (t, *J* = 10.4 Hz, 1H), 3.66 (t, *J* = 9.2 Hz, 1H), 3.58 (t, *J* = 9.2 Hz, 1H), 3.49-3.39 (b, OH), 3.19 (m, 1H), 2.67 (m, CH_2), 2.48 (m, CH_2), 2.08 (s,CH_3), 1.44 (s, CH_3), 1.37 (s, CH_3); (100 MHz, $CDCl_3$) δ_C 206.9 (C=O), 171.6 (C=O) 136.7, 128.0 · 2, 127.5, 127.4 · 2, 99.6, 99.5 (C-1), 74.2, 72.9, 71.9, 70.4, 66.9 (C-6), 61.6 ($-CH_2$), 37.7 ($-CH_3$), 29.4 ($-CH_3$), 28.6 ($-CH_3$), 27.6 ($-CH_3$), 18.7 ($-CH_3$).

Benzyl-2-*O*-dimethyl-tert-butyl-silyl-4-oxopentanoate-4,6-*O*-(1-methylethylidene)-*β*-D-glucopyranoside **36**. The alcohol **32** (101 mg, 0.25 mmol) was dissolved in the Dry DCM and 2,6-lutidiene (0.143 mL, 1.24 mmol). After the mixture was cooled to 0°C, TMSO*f* (0.11 mL, 0.5 mmol) was added dropwise. The reaction was warmed to RT overnight. The TLC (silica, Hex-EtOAc 6:1, R_f = 0.64) said the reaction was complete and the solvent was removed under reduced pressure. The residue was purified by column chromatography (silica, Hex-EtOAc 10:1→5:1, 0.1% NEt_3) to the C-2-*O*-silyl ether **22** in 89% (0.557). However, the ^{1}H- and ^{13}C-NMR of **22** was not definitive, so another method was developed to the glucosyl donor for glycosylation.

4-(methylthio)phenyl-2-*O*-dimethyl-tert-butyl-silyl-4-oxopentanoate-4,6-*O*-(1-methylethylidene) *β* -D-glucopyranoside **37**. The was attempted through the same method as the *β*-benzyl-glucoside **37**. (silica, Hex-EtOAc 6:1→ 4:1, R_f = 0.68, 0.78, 0.1% NEt_3). A pure ^{1}H- or ^{13}C-NMR of the *β*-thiol-glucoside **37** could not be obtained, causing the need another route to prepare the glucosyl donor.

3.4.2. Preparation of the Glucosyl Donor using Alloc at C-2-Glcp

Route 1 using *β*–OPMB Protecting Group



1,2,3,4,6-pentaacetate-*α,β*-D-glucopyranoside **38**. Followed *Method 1* previous discussed as **12i**. The ^{1}H and ^{13}C-NMR matched those of the *Method 1*.

4-methoxyphenyl,2,3,4,6-tetraacetate-*β*-D-glucopyranoside **38i**. The peracetylated glucoside **24** (10 g, 25.6 mmol) and DCM (100 mL) was cooled to 0°C, then slowly added $BF_3 \cdot Et_2O$ (8.1 mL, 64.1 mmol). The reaction was stirred for 4-6 hrs at RT. The reaction was quenched with sat. aq. $NaHCO_3$, then extracted with DCM (100 mL · 2). The organic layers were collected, dried over $MgSO_4$ and the filtrate was removed under reduced pressure. The residue was purified with column chromatography (silica, Hex-EtOAc, 5:1 → 2:1) to the *β*-glycoside **38i** in 73%. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.21 (d, *J* = 9 Hz, 2H, Ar−*H*), 6.88 (d, *J* = 9 Hz, 2H, Ar−*H*), 5.20-5.20-5.00 (m, 3H), 4.82 (d, *J* = 12.1 Hz, 1H, H_2C−O−Ar), 4.56 (d, *J* = 12.1 Hz, 1H, H_2C−O−Ar), 4.52 (d, *J* = 7.9 Hz, 1H, 1-H-Glc*p*), 4.28 (dd, *J* = 12.1, 5.3 Hz, 1H, H-6-Glc*p*), 4.19 (dd, *J* = 12.1, 5.3 Hz, 1H, H-6-Glc*p*), 3.81 (s, 3H, Ar-O-CH_3), 3.87 (ddd, *J* = 10.1, 5.2, 2.5 Hz, 1H, 5-H-Glc*p*), 2.11 (s, 3H, −CH_3), 2.02 (s, 3H, −CH_3), 2.00 (s, 3H, −CH_3), 1.99 (s, 3H, −CH_3). (100 MHz, $CDCl_3$) δ_C 170.5 (C=O), 170.2 (C=O), 169.3 (C=O), 169.2 (C=O), 159.4 (C=C), 129.4 · 2 (C=C), 129.4 · 2 (C=C), 113.7 (C=C), 98.7 (C-1), 72.7, 71.7, 71.1, 70.3, 68.3, 61.9, 55.1, 20.6 (−CH_3), 20.5 (−CH_3), 20.5 (−CH_3), 20.4 (−CH_3). The spectra data matched previously reported data.[11]

4-methoxyphenyl-*β*-D-glucopyranoside **39**. The *β*-glucoside **38i** (5.8 g, 12.8 mmol) was dissolved in MeOH (60 mL) and cat. NaOMe (70 mg, 1.28 mmol). The reaction was stirred at RT overnight, when the TLC (silica, EtOAc: MeOH 10:1, R_f = 0.19) showed the reaction to me complete. The reaction was neutralized with Amberlite IR-120 (H) ion exchange resin and filtered. The solvent was removed under reduced pressure. The crude tetraol **39** was used directly in the next step without further purification. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.31 (d, *J* = 7.8 Hz, 2H, Ar−*H*), 6.89 (d, *J* = 7.8, 2H, Ar−*H*), 5.19-5.00 (br, 4H), 4.74 (d, *J* = 11.8 Hz, 1H, H_2C−O−Ar), 4.49 (d, *J* = 11.8 Hz, 1H, H_2C−O−Ar), 4.20 (d, *J* = 7.9 Hz, 1H, 1-H-Glc*p*), 3.72 (s, 3H, Ar−O−CH_3), 3.71-3.67 (m, 1H), 3.48 (dd, *J* = 11.1, 5.2 Hz, 1H), 3.15-3.01 (m, 4H). (100 MHz,

$CDCl_3$) δ_C 158.7 (C=C), 129.9 · 2 (C=C), 129.4 · 2 (C=C), 113.5 (C=C) 101.8 (C-1) 77.0, 76.8, 73.5, 70.2, 69.2, 61.2, 55.1.

4-methoxyphenyl-4,6-*O*-(1-methylethylidene)-*β*-D-glucopyranoside **40**. The crude *β*-tetraol **39** (1.54 g, 5.6 mmol) was dissolved in the minimum amount DMF (10 mL) and DMP (2.8 mL, 22.8 mmol). Cat. *p*-TsOH (21 mg, 0.1 mmol) was added to the reaction mixture and allowed to stir for 36 hrs. The reaction was quenched with 0.2 mL of NEt_3 and co-evaporated with Tol 3 times. The reside was purified by column chromatography (silica, Hex-EtOAc, 2:1→1:2, 0.1% NEt_3) to give **40** in 69% (1.07 g). (TLC silica, R_f = 0.75 EtOAc-MeOH 10:1).

4-methoxyphenyl-3-*O*-tigloyl-4,6-*O*-isopropylidene-*β*-D-glucopyranoside. After cooling to 0°C, DCC (566.7 mg, 2.74 mmol) was added to a cold solution of the *β*-diolglucoside **40** (746.6 g, 2.28 mmol), tig acid (247 mg, 2.74 mmol), and DMAP (27.9 mg, 0.228 mmol) in DCM (25 mL). The reaction was warmed to RT and stirred overnight. At this point, TLC (silica, Hex-EtOAc, 2:1, R_f = 0.53) showed the reaction was complete. The solvent was partially removed, then Hex (60 mL) and Et_2O (30 mL) were added and stirred for 30 mins. The precipitate (DCU) formed was filtered through a pad of Celite using DCM (20 mL) as the eluent, and the filtrate was concentrated in vacuo. The residue was purified by column chromatography (silica, Hex-EtOAc 5:1 → 1:2, 0.1% NEt_3) to give **41** (195 mg, 21%), **42** (420 mg, 50%), and **43** (273 mg, 26%) as white amorphous solids.

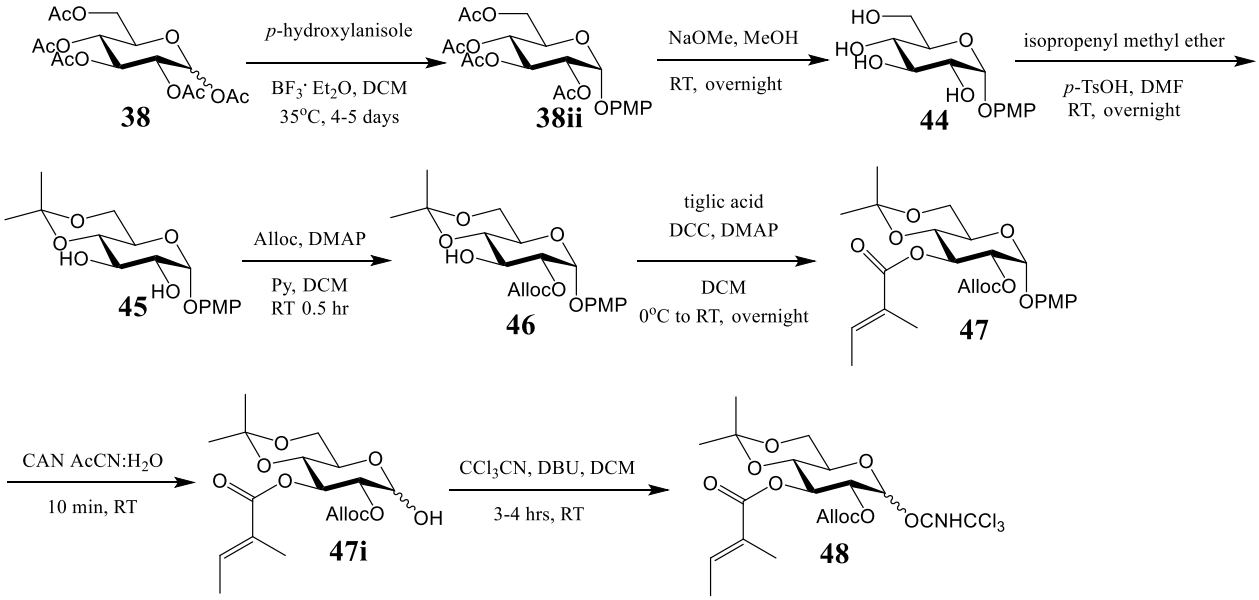
Compound **41**. $[\alpha]^{25}_D$ −64.4 (*c* 1 $CHCl_3$). ^{1}H NMR (400 MHz; $CDCl_3$): δ_H 7.03−6.97 (m, 2H, 2 · Ar−*H*), 6.98−6.91 (m, 1H, MeC*H*C(Me)C=O), 6.85−6.80 (m, 2H, 2 · Ar−*H*), 5.12 (t, *J* = 9.6 Hz, 1H, H-3-Glc*p*), 4.93 (d, *J* = 7.6 Hz, 1H, H-1-Glc*p*), 3.98 (dd, *J* = 10.8, 5.2 Hz, 1H, H-6-Glc*p*), 3.85−3.75 (m, 6H, H-2-Glc*p*, H-4, H-6-Glc*p*, −OC*H*$_3$), 3.48−3.41 (m, 1H, H-5-Glc*p*), 3.21

(d, *J* = 3.6 Hz, 1H, *H*O-2-Glc*p*), 1.90−1.80 (m, 6H, C*H*$_3$CHC(C*H*$_3$)C=O), 1.48 (s, 3H, (C*H*$_3$)$_2$C-O), 1.39 (s, 3H, (C*H*$_3$)$_2$C-O). ^{13}C NMR (100 MHz, CDCl3) δ_C 168.6 (C=O), 155.6 (C=C), 150.9 (C=C), 138.7 (C=C), 128.1 (C=C), 118.6 · 2 (C=C), 114.52 · (C=C), 102.9 (O–C–O), 99.7 (C-1), 74.8, 73.6, 71.1, 67.4, 62.1, 55.6, 28.9, 18.9, 14.5, 12.1.

Compound **42**. $[\alpha]^{25}_D$ −57.1 (*c* 1 CHCl3); ^{1}H NMR (400 MHz, CDCl$_3$) δ_H 6.99−6.89 (m, 1H, MeCHC(Me)C=O, Ar−*H*), 6.84− 6.77 (m, 2H, Ar−*H*), 5.15 (dd, J = 9.2, 8.0 Hz, 1H, H-2-Glc*p*), 4.99 (d, J = 7.6 Hz, 1H, H-1-Glc*p*), 3.99 (dd, J = 10.8, 5.6 Hz, 1H, H-6-Glc*p*), 3.90−3.80 (m, 2H, H-3-Glc*p*, H-6-Glc*p*), 3.78−3.72 (m, 4H, H-4-Glc*p*, −OC*H*$_3$), 3.43−3.35 (m, 1H, H-5-Glc*p*), 2.89 (br, 1H, *H*O-3-Glc*p*), 1.90−1.78 (m, 6H, C*H*$_3$CHC(C*H*$_3$)C=O), 1.55 (s, 3H, (C*H*$_3$)$_2$C), 1.46 (s, 3H, (C*H*$_3$)$_2$C). ^{13}C NMR (100 MHz, CDCl$_3$) δ_C 167.7 (C=O), 155.6 (C=C), 151.1 (C=C), 138.9 (C=C), 127.9 (C=C), 118.7 · 2 (C=C), 114.5 · 2 (C=C), 101.1 (O–C–O), 99.9 (C-1), 74.8, 73.5, 73.0, 67.2, 61.9, 55.6, 28.9, 19.0, 14.5, 12.2.

Compound **43**. $[\alpha]^{25}_D$ +8.0 (*c* 1 CHCl$_3$); ^{1}H NMR (400 MHz, CDCl$_3$) δ_H 6.96−6.88 (m, 2H, Ar–*H*), 6.86−6.76 (m, 4H, 2 ·Ar−*H*, 2 ·H$_3$CHCCH$_3$C=O), 5.38−5.27 (m, 2H, H-2-Glc*p*, H-3-Glc*p*), 5.10−5.00 (d, *J* = 7.2 Hz, 1H, H-1-Glc*p*), 4.00 (dd, *J* = 11.2, 5.6 Hz, 1H, H-6-Glc*p*), 3.94−3.80 (m, 2H, H-4-Glc*p*, H-6-Glc*p*), 3.76 (s, 3H, Ar−OC*H*$_3$), 3.54−3.46 (m, 1H, H-5-Glc*p*), 1.82−1.73 (m, 12H, 2 · C*H*$_3$CHC(C*H*$_3$)C=O), 1.49 (s, 3H, (C*H*$_3$)$_2$C−), 1.39 (s, 3H, (C*H*$_3$)$_2$C−). ^{13}C NMR (100 MHz, CDCl$_3$) δ_C 167.1 (C=O), 166.5 (C=O), 155.6 (C=C), 151.1 (C=C), 138.3 (C=C), 138.0 (C=C), 128.0 (C=C), 127.8 (C=C), 118.8 · 2 (C=C), 114.5 · 2 (C=C), 101.2 (O–C–O), 99.7, 72.1, 72.0, 71.4, 67.6, 62.1, 55.6, 28.9, 18.9, 14.4, 14.4, 12.1, 12.1.

Route 2 using the α-OPMP Propecting Group



Glucosyl Donor

4-methoxyphenyl, 2,3,4,6-tetraacetate-α-D-glucopyranoside **38ii**. The α-PMP-glucoside **38ii** was achieved through the same method as the β-PMP-glucoside **38i**. The reaction time was lengthened to 4-5 days to allow the β-intermediate to isomerize to the α-PMP in 80% (4.11 g).[12] H-NMR data.

4-methoxyphenyl-α-D-glucopyranoside **44**. To obtain the tetraol **44** the same procedure was followed as the β-PMP-glucoside **39**. The tetraol **44** was attained in 90% (2.15 g) and carried to the next step without further purification.

4-methoxyphenyl-4,6-*O*-(1-methylethylidene)-α-D-glucoside **44i**. To a cold (0 °C) solution of the **44** (70.0 g, 179 mmol) and p-methoxyphenol (33.4 g, 269 mmol) in Dry DCM (800 mL) was added $BF_3 \cdot OEt_2$ (111 mL, 897 mmol) over 30 min. The mixture was then heated to 35 °C and agitated for 4 days, at the end of which time TLC (Hex−EtOAc, 2:1) indicated that the reaction was complete. The reaction mixture was poured onto crushed ice, and the excess $BF_3 \cdot$

OEt_2 was neutralized by the careful addition of saturated aqueous $NaHCO_3$ solution. The organic layer was washed with brine, dried (Na_2SO_4), and concentrated in vacuo to provide a syrup. The syrup was dried under high vacuum for 2 h before being dissolved in MeOH (500 mL). NaOMe (800 mg) was added to the reaction mixture at RT and stirred for 2 hrs. The residue was purified by column chromatography (silica, EtOAc−MeOH, 1:0 → 10:1) to afford *p*-methoxyphenyl-α-D-glucopyranoside **44i** (30.8 g, 60% over two steps) as a white solid. To a solution of compound **44i** (28.8 g, 101 mmol) in DMF (200 mL) containing TsOH · H_2O (0.38 g, 2.0 mmol) was added DMP (11.6 mL, 121 mmol) under nitrogen atmosphere. The mixture was stirred for 6 hrs. at RT, when TLC showed the reaction to be complete (Hex−EtOAc, 1:2). The reaction was quenched with NEt_3 (0.5 mL) and then diluted with EtOAc (400 mL) and washed with water (400 mL · 3) and brine (100 mL). The aqueous layer was extracted with EtOAc (400 mL · 2). The combined organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex−EtOAc, 2:1 → 1:1) to give compound **45** (30.2 g, 92%) as a white foam: $[\alpha]^{25}_D$ +124.7 (*c* 1 $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.02−6.96 (m, 2H, 2 · Ar−*H*), 6.84−6.79 (m, 2H, 2 · Ar−*H*), 5.41 (d, J = 3.6 Hz, 1H, H-1-Glc*p*), 4.00 (td, *J* = 9.2, 2.0 Hz, 1H), 3.88−3.66 (m, 7H, H-2, H-5, 2 · H-6-Glc*p*, −OCH_3), 3.61 (t, *J* = 8.8 Hz, 1H), 3.45−3.40 (m, 1H, −O*H*), 3.00−2.92 (m, 1H, −O*H*), 1.52 (s, 3H, (CH_3)$_2$C−), 1.44 (s, 3H, (CH_3)$_2$C−). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 155.4, 150.2, 118.4 · 2, 114.6 ·2, 99.8, 98.6, 73.4, 72.8, 71.9, 64.0, 62.1, 55.6, 29.0, 19.1.

4-methoxyphenyl-2-*O*-(allyloxycarbonyl)-4,6-*O*-isopropylidene-*α*-D-glucopyranoside **46**. The diol **45** (13.50 g, 41.6 mmol) was dissolved in a mixture of DCM (200 mL), Py (13.3 mL, 166.7 mmol) and DMAP (506.7 mg, 4.2 mmol), then under an N_2 atmosphere, allyl chloroformate (4.86 mL, 45.9 mmol) in DCM (50 mL) was added dropwise to the solution at −30 °C over 30

min. The reaction mixture was gradually warm to 0 °C over 30 min, when TLC (silica, Hex-EtOAc, 2:1) showed the reaction was complete. The reaction mixture was then quenched with MeOH (0.5 mL) and washed with 1 M HCl (100 mL), sat. aq. $NaHCO_3$ (100 mL), and brine sol. (100 mL). The aqueous layer was extracted with DCM (200 mL). The combined organic layer was dried over Na_2SO_4 and concentrated under reduced pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc, 5:1→2:1, 0.1% NEt_3) to give **46** in 81% (14.1 g) as a white foam. $[\alpha]^{25}_D$ +152.9 (*c* 1 $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.02−6.95 (m, 2H, 2 · Ar−*H*), 6.85−6.77 (m, 2H, 2 · Ar−*H*), 5.96−5.85 (m, 1H, CH_2=C*H*CH_2−), 5.65 (d, *J* = 3.8 Hz, 1H, H-1-Glc*p*), 5.38−5.22 (m, 2H, C*H*$_2$=CHCH_2−), 4.72 (dd, *J* = 9.6, 3.6 Hz, 1H, H-2-Glc*p*), 4.66−4.58 (m, 2H, CH_2=CHC*H*$_2$−), 4.26 (t, *J* = 9.3 Hz, 1H, H-3-Glc*p*), 3.91−3.81 (m, 2H, H-5-Glc*p*, H-6-Glc*p*), 3.81−3.74 (m, 4H, H-6-Glc*p*, −OC*H*$_3$), 3.75−3.63 (m, 1H, H-4-Glc*p*), 3.04 (br, 1H, –O*H*), 1.54 (s, 3H, (C*H*$_3$)$_2$C–O), 1.45 (s, 3H, (C*H*$_3$)$_2$C–O). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 155.3, 154.4, 150.3, 131.1, 119.2, 118.2 · 2, 114.5 · 2, 100.0, 95.8, 76.7, 73.7, 68.97, 68.8, 63.7, 62.1, 55.6, 28.9, 19.1.

4-methoxyphenyl-2-*O*-(allyloxycarbonyl)-3-*O*-tigloyl-4,6-*O*-isopropylidene-*α*-D-gluco-pyranoside **47**. DCC (5.73 g, 27.8 mmol) was added in one portion to a -10 °C DCM (100 mL) solution of alcohol **46** (7.6 g, 18.5 mmol), tig acid (2.78 g, 27.8 mmol), and DMAP (226.7 mg, 1.85 mmol). The reaction was slowly warmed to RT and stirred overnight. At this point, TLC (silica, Hex-EtOAc, 2:1) showed the reaction was complete. The reaction mixture was diluted with Et_2O (100 mL) and Hex (50 mL), stirred for 15 min, and then filtered through a pad of Celite using Et_2O (50 mL) as the eluent, and the filtrate was concentrated in vacuo. The residue was purified by column chromatography (silica, Hex-EtOAc, 8:1 → 5:1, 0.1% NEt_3) to give intermediate **47** in 85% (8.10 g) as a colorless syrup. $[\alpha]^{25}_D$ +127.5 (*c* 1 $CHCl_3$); ^{1}H NMR (400

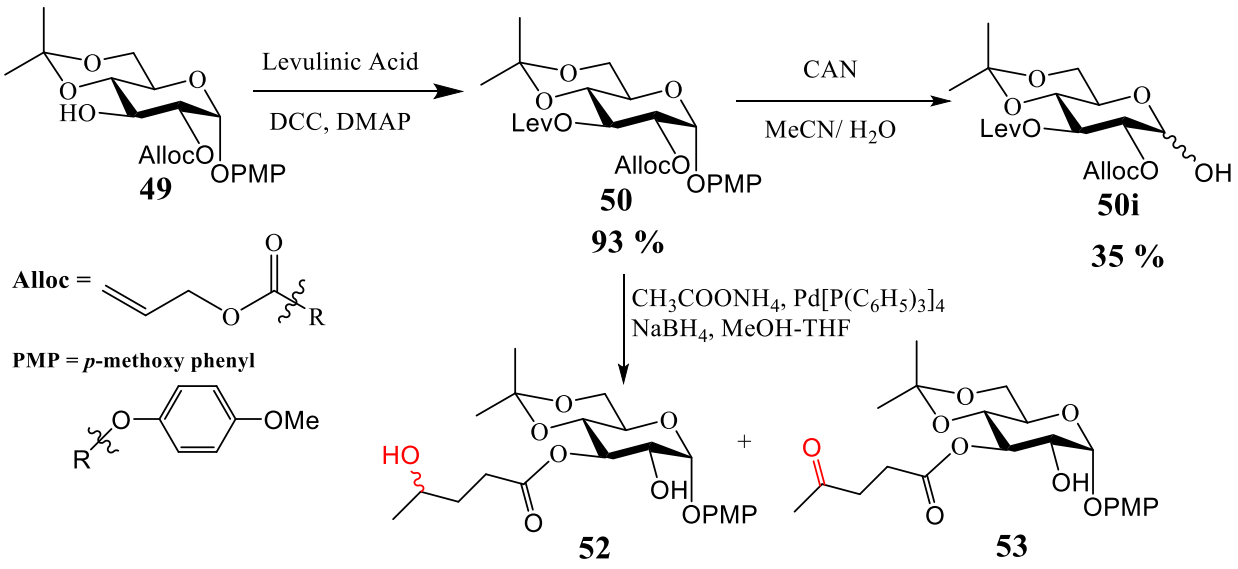
MHz, MeOD) δ_H 7.05−6.98 (m, 2H, 2 · Ar–*H*), 6.93−6.83 (m, 3H, MeC*H*C(Me)$_2$, 2 · Ar–*H*), 5.96−5.75 (m, 1H, –CH_2= $CHCH_2$−), 5.67 (d, *J* = 3.6 Hz, 1H, H-1-Glc*p*), 5.57 (t, *J* = 10.0 Hz, 1H, H-3-Glc*p*), 5.29−5.15 (m, 2H, CH_2=CHCH_2−), 4.96 (dd, *J* = 10.1, 3.6 Hz, 1H, H-4-Glc*p*), 4.65−4.51 (m, 2H, CH_2=$CHCH_2$−), 4.01−3.79 (m, 4H, H-4-Glc*p*, H-5-Glc*p*, 2 · H-6-Glc*p*), 3.75 (s, 3H, −OCH_3), 3.75 (d, *J* = 4.7 Hz, 3H), 1.86−1.74 (m, 6H, CH_3CHC(CH_3)–C=O), 1.51 (s, 3H, (CH_3)$_2$C–O), 1.35 (s, 3H, (CH_3)$_2$C–O). ^{13}C NMR (100 MHz, MeOD) δ 168.4, 157.0, 155.6, 151.7, 139.6, 132.8, 129.2, 119.4 · 2, 118.9, 115.7 ·2, 101.2, 97.7, 75.9, 73.1, 70.8, 69.8, 65.7, 63.1, 56.0, 29.3, 19.4, 14.5, 12.2. IR (film) ν = 2993, 2940, 1747, 1718, 1652, 1510, 1277, 1244, 1211, 1042; HRMS (ESI) m/z called for $C_{25}H_{32}NaO_{10}$ $[M + Na]^+$ 515.1893, found 515.1875.

hemiacetal-2-*O*-(allyloxycarbonyl)-3-*O*-tigloyl-4,6-*O*-isopropylidene-*α,β*-D-gluco-pyranoside **47i**. To a solution of the *α*-PMP-glucoside **47** (7.26 g, 14.7 mmol) in AcCN/H_2O (9:1 72 mL: 8 mL) was added a solution of CAN (16.2 g, 29.5 mmol) in H_2O (10 mL) over 5 min at −10 °C. The mixture was stirred for 10 mins at the same temperature, at the end of which time TLC (silica, 2:1, Hex-EtOAc) indicated that the reaction was complete. The reaction was quenched with saturated aqueous $NaHCO_3$ and extracted with EtOAc (400 mL · 2). The combined organic phase was dried over Na_2SO_4 and concentrated under reduced pressure. The obtained *α,β*-hemiacetal glucoside **47i** was purified by column chromatography (silica, Hex-EtOAc, 4:1→1:1, 0.1% NEt_3) and proceeded to the next step.

trichloroacetimidate-2-*O*-[(allyloxy)carbonyl]-3-*O*-tigloyl-4,6-*O*-isopropylidene-*α,β*-D-glucopyranoside **48**. Trichloroacetonitrile (5.93 mL, 58.7 mmol) was added to a solution of the obtained *α,β*-hemiacetal **47i** in DCM (60 mL) at RT, then DBU (0.22 mL, 1.47 mmol). The mixture was stirred at RT overnight (turned red-orange color), TLC (silica, Hex-EtOAc, 4:1, R_f =

0.55, 0.49) showed the reaction was complete. The reaction was quenched with NEt_3 (1 mL) and the solvent was removed under reduced pressure. The residue was purified by column chromatography (silica, Hex-EtOAc, 6:1 → 1:1) to afford the glucosyl donor **48** (4.23 g, 51% over two steps) as a colorless syrup. $[\alpha]^{25}_D$ +59.6 (*c* 1 $CHCl_3$). ^{1}H NMR (400 MHz, CDCl3) δ_H 8.66 (s, 1H, CN*H*CCl_3), 6.90−6.83 (m, 1H, MeC*H*C(Me)C=O), 6.57 (d, *J* = 4.0 Hz, 1H, H-1-Glc*p*), 5.93−5.76 (m, 1H, CH_2=C*H*CH_2−), 5.61 (t, *J* = 9.6 Hz, 1H, H-3-Glc*p*), 5.36−5.17 (m, 2H, C*H*$_2$=CHCH_2−), 5.03 (dd, *J* = 9.6, 4.0 Hz, 1H, H-2-Glc*p*), 4.65−4.54 (m, 2H, CH_2=CHC*H*$_2$−), 4.05−3.72 (m, 4H, H-2-Glc*p*, H-4-Glc*p*, H-5-Glc*p*, 2 · H-6-Glc*p*), 1.90−1.76 (m, 6H, C*H*$_3$CHC(C*H*$_3$)C=O), 1.49 (s, 3H, (C*H*$_3$)$_2$C–O), 1.41 (s, 3H, (C*H*$_3$)$_2$C–O). ^{13}C NMR (100 MHz, CDCl3) δ_C 166.5, 161.2, 154.2, 138.0, 131.1, 128.0, 118.7, 100.0, 93.6, 74.1, 71.7, 69.07, 68.97, 66.2, 62.0, 28.9, 18.8, 14.4, 12.1.

3.4.3. Attempted Preparation of Glucosyl Donor using Alloc at C-2-Glcp

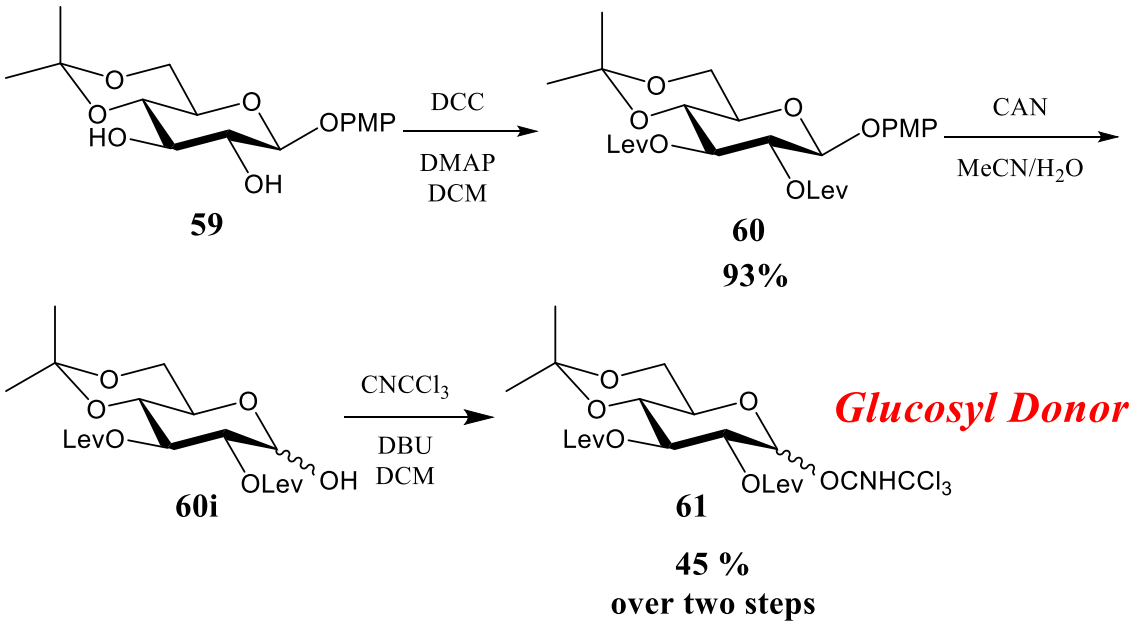


4-methoxyphenyl-2-*O*-(allyloxycarbonyl)-3-*O*-levulinoyl-4,6-*O*-isopropylidene-D-*α*-gluco-pyranoside **50**. The alcohol **50** was obtained in four steps from D-Glc*p* **12** through the procedure

discussed previously (3.1.2, Route 2). A solution of the alcohol **49** (3.51 g, 8.5 mmol), lev. acid (1.19 g, 10.2 mmol) (DMAP (1.25 g, 4.2 mmol), and DCM (50 mL) were cooled to -10 ^{o}C. Then DCC (2.12 10.2 mmol) was added to the solution in portion and the reaction allowed to slowly warm to RT overnight. The TLC (silica, Hex-EtOAc, 2:1, R_f = 0.61). The solvent was partially removed, then Hex (40 mL) and Et_2O (20 mL) were added, then stirred for 20 min to precipitate the DCU. The mixture was filtered through a pad of Celite using Et_2O (20 mL) as the eluent, and the filtrate was concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex-EtOAc 5:1 → 1:2, 0.1% NEt_3) to give intermediate **50** in 93% (3.91 g). **50** was not fully characterized and continued to the next step.

4-hemiacetal-2-*O*-(allyloxycarbonyl)-3-*O*-levulinoyl-4,6-*O*-isopropylidene-*α*,*β*-D-gluco-pyranoside **50i**. After cooling to 0 ^{o}C, the CAN (5.53 g, 9.77 mmol) dissolved in H_2O (8 mL) and added dropwise to a solution of compound **50** (1.24 g, 2.4 mmol), AcCN (30 mL), and H_2O (7 mL). The reaction was kept at 0^{o}C for 10 mins as the reaction was monitored by TLC (silica, Hex-EtOAc 1:1, R_f = 0.47). Sat. aq. $NaHCO_3$ (40 mL) was added to the reaction causing the reaction to change colors (yellow → clear) and extracted with EtOAc (50 mL· 3). The organic layers were combined, dried over $NaSO_4$, filtered and solvent removed under reduced pressure. The syrup was purified by column chromatography (silica, Hex-EtOAc 3:1 → 1:1, NEt_3 0.1%) to give hemiacetal **50i** in 35.4% (0.348 g).

3.4.4. Synthesis of the Glucosyl Donor with C-2 and C-3 Lev Protecting Groups.



4-methoxyphenyl-2,3-*O*-levulinoyl-4,6-*O*-isopropylidene-*β*-D-glucopyranoside **60**. The alcohol **59** was obtained in four steps from D-Glc*p* **12** through the procedure discussed previously (Section 3.1.2). A solution of the diol **59** (6.25 g, 19.15 mmol), lev. acid (6.68 g, 57.5 mmol), DMAP (4.75 g, 3.75 mmol), and DCM (50 mL) were cooled to -10 °C. Then DCC (7.11 g, 34.5 mmol) was added to the solution in portion and the reaction allowed to slowly warm to RT overnight. The TLC (silica, Hex-EtOAc, 2:1, R_f = 0.51). The solvent was partially removed, then Hex (10 mL) and Et_2O (20 mL) were added and stirred for 15 min to precipitate the DCU. The mixture was filtered through a pad of Celite using Et_2O (20 mL) as the eluent, and the filtrate was concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex-EtOAc 4:1→1:2, 0.1% NEt_3) to give intermediate in **60** in 85% (9.2 g). $[\alpha]^{25}_D$ –21.1° (*c* 1 $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.01 – 6.90 (m, 2H, 2 · Ar–*H*), 6.86 – 6.76 (m, 2H, 2 ·

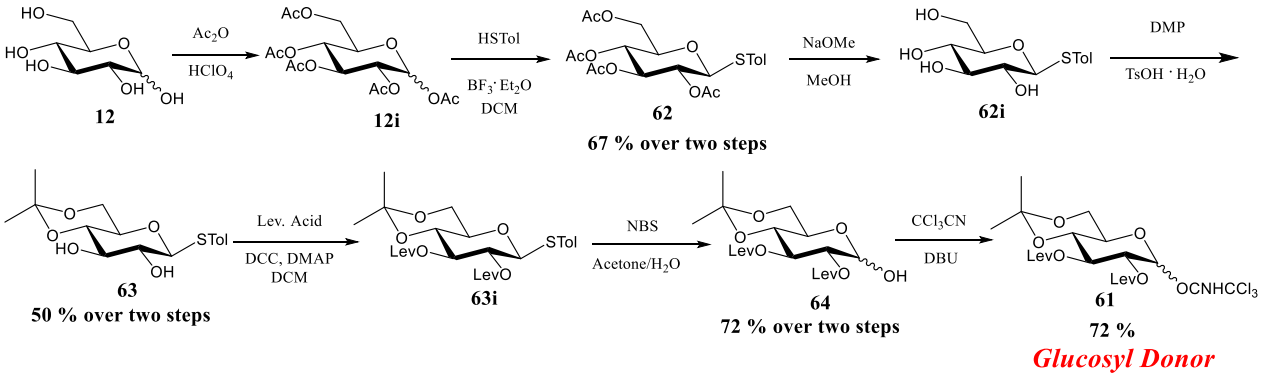
Ar–*H*), 5.27 – 5.13 (m, 2H, H-2-Glc*p*, H-3-Glc*p*), 5.00 – 4.90 (m, 1H, H-1-Glc*p*), 3.98 (dd, *J* = 10.8, 5.2 Hz, 1H, H-6-Glc*p*), 3.88 – 3.70 (m, 5H, H-4- Glc*p*, H-6- Glc*p*, –OCH_3), 3.50 – 3.35 (m, 1H, H-5-Glc*p*), 2.89 – 2.69 (m, 4H, CH_3-C=O(CH_2)$_2$C=O), 2.69 – 2.52 (m, 4H, CH_3-C=O(CH_2)$_2$C=O), 2.18 (s, 3H, CH_3-C=O(CH$_2$)$_2$C=O), 2.16 (s, 3H, CH_3-C=O(CH$_2$)$_2$C=O), 1.48 (s, 3H, (CH_3)$_2$C), 1.38 (s, 3H, (CH_3)$_2$C). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 206.35, 206.23, 171.89, S9 171.42, 155.69, 150.97, 118.73 · 2, 114.51· 2, 100.98, 99.80, 71.99, 71.93, 71.08, 67.48, 61.92, 55.58, 37.75, 37.69, 29.76, 29.73, 28.85, 27.89, 27.76, 18.84.

hemiacetal-2,3-*O*-levulinoyl-4,6-*O*-isopropylidene-*α,β*-D-glucopyranoside **60i**. To a solution of the *β*-PMP-glucoside **60** (2.68 g, 5.13 mmol) in AcCN/H_2O (8:1 45 mL) and was added a solution of CAN (5.62 g, 10.2 mmol) in H_2O (5 mL) over 5 min at −10 °C. The mixture was stirred for 10 mins at the same temperature, when TLC (silica, 2:1, Hex-EtOAc) indicated that the reaction was complete. The reaction was quenched with sat. aqus. $NaHCO_3$ and extracted with Et_2O (60 mL · 2). The combined organic phase was dried over Na_2SO_4 and concentrated under reduced pressure. The obtained *α,β*-hemiacetal Glc*p* **60i** was purified by column chromatography (silica, Hex-EtOAc, 4:1→1:1, 0.1% NEt_3) and proceeded to the next step.

trichloroacetimidate-2,3-*O*-levulinoyl-4,6-*O*-isopropylidene-*α,β*-D-glucopyranoside **60**. CCl_3CN (8.35 g, 83.0 mmol) and DBU (0.31 mL, 2.01 mmol) were are added to the mixture of the α,*β*-hemiacetal **59i** (8.64 g, 20.8 mmol). The reaction was allowed to stir at RT for 3-5 hrs. as the mixture turned an orange-brown color. The reaction was quenched with 0.5 mL of NEt_3 after the TLC (silica Hex-EtOAc 1:1 R_f = 0.65) showed the reaction to be complete. The mixture was concentrated under reduced pressure. The Schmidt donor **60** was obtained in 85% (9.90 g), after purification by column chromatography (silica, Hex-EtOAc 4:1 →1:1, few drops of NEt_3). $[\alpha]^{25}_D$

+ 52.4° (*c* 1 $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 8.71, 8.64 (2s, 1H, OCN*H*CCl_3), 6.45 (d, *J* = 4.5 Hz, 1H, H-1-Glc*p*), 5.49 (t, *J* = 9.6 Hz, 1H, H-3-Glc*p*), 5.12 (dd, *J* = 10.0, 4.0 Hz, 1H, H-2-Glc*p*), 3.98 – 3.87 (m, 2H, H-5, H-6-Glc*p*), 3.84 – 3.68 (m, 2H, H-4-Glc*p*, H-6-Glc*p*), 2.90 – 2.46 (m, 8H, 2 · CH_3–C=O(CH_2)$_2$C–O), 2.17 (s, 3H, CH_3-C=O(CH_2)$_3$C-O), 2.16, 2.15 (2s, 3H, CH_3–C=O(CH_2)$_2$C–O), 1.49, 1.48 (2s, 3H, (CH_3)$_2$C–O), 1.40, 1.39 (2s, 3H, (CH_3)$_2$C–O). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 206.3, 206.2, 171.81, 171.7, 160.9, 99.9, 93.6, 90.7, 71.3, 70.3, 69.1, 66.0, 61.94, 37.7, 37.5, 29.7, 29.7, 28.7, 27.8, 27.5, 18.9.

3.4.5. Preparation of the Glucosyl Donor using Thiotolyl Protection



4-(methylthio)phenyl-2,3,4,6-tetraacetate-*β*-D-glucopyranoside **62**. The peracetylated glucose **12i** (27 g, 69.3 mmol) obtained though *Method 2* (Section 3.4.1) was dissolved in Dry DCM (300 mL) with 4-methylthiophenol (12.9 g, 138.7 mmol) and cooled to -10 ºC. Then, the $BF_3 \cdot OEt_2$ was slowly added and reaction stirred 12-48 hrs at RT. The reaction was monitored by TLC (silica, Hex-EtOAc 2:1, R_f = 0.54). The reaction time can be decrease with the use of an ultrasonic bath. The reaction mixture was slowly poured into crushed ice and stirred for 10 mins. The reaction was neutralized with careful addition of sat. aq. $NaHCO_3$ (200 mL) and extracted (300 mL · 2) with DCM. The combined organic layers were dried over $NaSO_4$, filtered, and the solvent removed under reduced pressure. The residue was purified with column chromatography

(silica, Hex-EtOAc, 5:1-1:1) to the *β*-thioGlc*p* **62** in 70% (85% BORSM). ^{1}H NMR (400 MHz, CDCl3) δ_H 7.38 (d, *J* = 7.8 Hz, 2H, 2 · Ar–*H*), 7.12 (d, *J* = 7.8 Hz, 2H, 2 · Ar–*H*), 5.20 (t, *J* = 9.6 Hz, 1H, H-3-Glc*p*), 5.01 (t, *J* = 9.6 Hz, 1H, H-4-Glc*p*), 4.93 (t, *J* = 9.7 Hz, 1H, H-2-Glc*p*), 4.62 (d, *J* = 10.1 Hz, 1H, H-1-Glc*p*), 4.23-4.14 (m, 2H, H-6-Glc*p*), 3.69 (ddd, *J* = 2.3, 4.3, 7.4 Hz, 1H, H-5-Glc*p*), 2.34 (s, 3H, S–Tol–C*H*$_3$), 2.08 (s, 3H, –C*H*$_3$), 2.08 (s, 3H, –C*H*$_3$), 2.00 (s, 3H, –C*H*$_3$), 1.99 (s, 3H, –C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 170.6 (C=O), 170.1 (C=O), 169.4 (C=O), 169.3 (C=O), 138.7 (C=C), 133.7 · 2 (C=C), 129.6 · 2 (C=C), 127.4 (C=C), 85.7 (C-1), 75.8, 73.8, 69.8, 68.2, 62.1, 21.2 (–CH_3), 20.7 (–CH_3), 20.7 (–CH_3), 20.5(–CH_3), 20.4 (–CH_3). The ^{1}H- and ^{13}C-NMR spectroscopic data matched previous report literature.[13, 14]

4-(methylthio)phenyl-*β*-D-glucopyranoside **62i**. The *β*-thioGlc*p* **62** (23.6 g, 51.9 mmol) was dissolved in MeOH (200 mL) and cat NaOMe (0.280 g, 5.2 mmol). The reaction was stirred at RT for 6-12 hrs, when the TLC (silica, EtOAc: MeOH 10:1, R_f = 0.15) showed the reaction to be complete. The reaction was neutralized with Amberlite IR-120 (H) ion exchange resin and filtered. The solvent was removed under reduced pressure. The crude tetraol **62i** was used directly in the next step without further purification. ^{1}H NMR (400 MHz, CD_3OD) δ_H 7.44 (d, *J* = 7.7, 2H, 2 · Ar–*H*), 7.11 (d, *J* = 7.8, 2H, 2 · Ar–*H*), 4.50 (d, *J* = 9.5, 1H, H-1-Glc*p*), 3.89 (d, *J* = 2.5, 1H), 3.76-3.71 (m, 2H), 3.59 (t, *J* = 9.2, 1H), 3.56-3.48 (m, 2H, H-5-Glc*p*), 2.30 (s, 3H, Ph –C*H*$_3$). ^{13}C NMR (100 MHz, CD_3OD) δ 138.5 (C=C), 133.0 · 2 (C=C), 132.2 · 2 (C=C), 130.7 (C=C), 90.8 (C-1), 80.7, 76.5, 71.1, 70.5, 62.7, 21.2 (Ph–CH_3). The ^{1}H- and ^{13}C-NMR spectra data match those previously reported.[13, 15]

4-(methylthio)phenyl-4,6-*O*-isopropylidine-*β*-D-glucopyranoside **63**. The crude tetraol **62i** (16.8 g, 58.6 mmol) was dissolved in the minimal amount of DMF (80 mL), then DMP (66

mL, 234 mmol) and cat. *p*-TsOH (223 mg, 1.1 mg) added to the solution at RT. The reaction was allowed to at RT overnight, while monitoring the reaction though TLC (Hex-EtOAc, 2:1, R_f = 0.49). The reaction was quenched with NEt_3 (0.5 mL), diluted with Tol, and solvent removed under reduced pressure. The crude syrup was dissolved in EtOAc (150 mL), washed with BRINE · 2, dried over $NaSO_4$, filtered, and the EtOAc under reduced pressure. The syrup was purified using column chromatography (silica, Hex-EtOAc 4:1→1:2) to the C-4,C-6-protected glucoside **63** in 75% (15.75 g) clear syrup; ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.40 (d, *J* = 8.2 Hz, 2H, 2 · Ar–*H*), 7.13 (d, *J* = 8.1 Hz, 2H, 2 · Ar–*H*), 4.58 (d, *J* = 9.6 Hz, 1H, C-1-Glc*p*), 3.94 (dd, *J* = 10.4, 5.2 Hz, 1H), 3.76 (t, *J* = 10.8 Hz, 1H), 3.68 (t, *J* = 8.4 Hz, 1H), 3.51 (t, *J* = 9.6 Hz, 1H), 3.41– 3.27 (m, 2H), 3.10 (br, 1H, –O*H*), 2.90 (br, 1H, –O*H*), 2.61 (s, 3H, Ph–CH_3), 1.49 (s, 3H, O–C–CH_3), 1.42 (s, 3H, O–C–CH_3). ^{13}C NMR (100 MHz, $CDCl_3$) δ_H 138.68 (C=C), 133.43 · 2 (C=C), 129.84 · 2 (C=C), 127.50 (C=C), 99.80 (O–C–O), 88.80 (C-1), 74.91, 72.89, 72.66, 71.52, 61.97, 28.94 (Ph–CH_3), 21.12 (O–C–CH_3), 19.10 (O–C–CH_3). All spectroscopic data matched previously published data.[16]

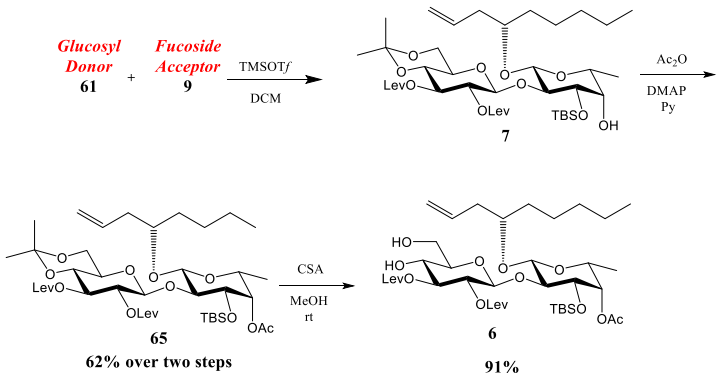
4-(methylthio)phenyl-2,3-*O*-levulinoyl-4,6-isopropylidine-*β*-D-glucopyranoside **63i**. DCC (23.13 g, 112 mmol) was added one portion to a cooled solution (0^{o}C) of DCM (300 mL), the diol **63** (12.2 g, 37.3 mmol), lev. acid (13.0 g, 112mmol), and DMAP (991 mg, 7.4 mmol). The reaction was stirred at RT overnight, as the reaction started to turn form white precipitate (DCU), and the reaction turned a reddish-brown color. After TLC (silica, Hex-EtOAc, 3:1, R_f = 0.58) showed the reaction to be complete the DCM was partially removed under reduced pressure. The reaction was diluted with Et_2O (200 mL) and Hex (100 mL) to cause the DCU to crash out of the solution. The slurry was filtered through a pad of Celite and concentrated under reduced

pressure. The residue was purified through column chromatography (silica, Hex-EtOAc, 2:1→1:1) to give the 2,3-lev-glucoside **63i** in 90% (17.5 g). Went directly to the next step.

Hemiacetal-2,3-*O*-levulinoyl-4,6-*O*-isopropylidene-*α,β*-D-glucopyranoside **64**. The 2,3-*O*-lev glucoside **63i** was dissolved in the mixture of Ac/H_2O (9:1, 108 mL: 12 mL) was cooled to -10 ^oC. Then, NBS (15.1 mmol, 89.2 mmol) was slowly added over five mins to the mixture and stirred at -10 ^oC for 5-10 mins. After TLC (silica, Hex-EtOAc, R_f = 0.51, 0.42) showed the reaction to be complete, sat. aq. $Na_2S_2O_3$ (40 mL, orange to clear) followed by sat. aq. $NaHCO_3$ (120 mL, bubbling) were added to the reaction. The mixture was partially removed under reduced pressure and the organic layer extracted with DCM · 2 (150 mL). The combined organic layers were collected, dried over $NaSO_4$, filtered, and removed under reduced pressure. The residue was purified with column chromatography (silica, Hex-EtOAc, 4:1→EtOAc) to the give the α,β-hemiacetal **64** in 80%.

Trichloroacetimidate-2,3-*O*-levulinoyl-4,6-*O*-isopropylidene-*α,β* -D-glucopyranoside **61**. The Schmidt donor **61** was prepare from the hemiacetal glucoside **60** through the same method as **60i → 61**.

3.5. Glycosylation, Acylation, C-4/C-6 Glucosyl De-protection



(4*S*)1-nonenyl2,3-di-*O*-levulinoyl-4,6-*O*-isopropylidene-*β*-D-glucopyranosyl-(1→2)-3-*O* tertbutyldimethylsilyl-*β*-D- fucopyranoside **7**. in Dis DCM (50 mL), the Fuc*p* acceptor **9** (6.10 g, 15.1 mmol), Glc*p* donor **61** (9.83 g, 16.7 mmol), and 4 Å molecular sieves (9.8 g) were cooled to –10 ^{o}C while stirring. Then, TMSOT*f* (310 µL, 1.51 mmol) was added to the mixture. The reaction was slowly added to warm over 1hr, when TLC (silica, Hex:EtOAc 2:1, R_f = 0.41) showed the fucosyl acceptor to be consumed. The reaction mixture was quenched with NEt_3 (0.5 mL) and filtrated through a pad of Celite (DCM 25 mL). The filtrate was removed under reduced pressure to give a syrup that was purified by column chromatography (silica, Hex-EtOAc, 4:1 → 1:1) to give the (1→2)-*β*-disaccharide **7** (8.61 g, 71%) as a colorless syrup. $[\alpha]_D^{25}$ –27.3° (*c* 1 $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 6.01 – 5.86 (m, 1H, CH_2=C*H*–CH_2–), 5.13 (d, *J* = 8.0 Hz, 1H, H-1-Gl*p*), 5.10 – 5.00 (m, 3H, C*H*$_2$=CH–CH_2–, H-3-Glu*p*), 4.88 (dd, *J* = 9.2, 7.6 Hz, 1H, H-2-Glu*p*), 4.29 (d, *J* = 7.6 Hz, 1H, H-1-Fuc*p*), 3.95 (dd, *J* = 10.8, 5.2 Hz, 1H, H-6-Glu*p*), 3.83 – 3.62 (m, 5H, H-4-Glu*p*, H-6-Glu*p*, H-2-Fuc*p*, H-3-Fuc*p*, H-4-Fuc*p*), 3.54 – 3.47 (m, 2H, H-5-Fuc*p*, –CH_2–C*H*–CH_2–), 3.30–3.24 (m, 1H, H-5-Glu*p*), 2.85–2.50 (m, 9H, O*H*-4-Fuc*p*, 2 · CH_3–C(O)C*H*$_2$C*H*$_2$C=O), 2.34 – 2.24 (m, 2H), 2.17 (2s, 6H, 2 · C*H*$_3$C(O)CH_2CH_2C=O), 1.57 – 1.48 (m, 2H), 1.46 (s, 3H, (C*H*$_3$)$_2$C), 1.40 – 1.20 (m, 12H, (C*H*$_3$)$_2$C, H-6-Fuc*p*), 0.92 (s, 9H), 0.89 (t, *J* = 6.8 Hz, 3H, –C*H*$_3$), 0.17 (s, 3H, C*H*$_3$–Si), 0.12 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 206.37, 206.26, 171.96, 171.23, 135.24, 116.47, 101.05, 99.65, 99.49, 79.07, 75.86, 75.45, 73.78, 72.63, 72.52, 71.32, 69.59, 67.35, 62.10, 38.27, 37.72, 37.63, 34.28, 31.82, 29.79, 29.77, 28.87, 27.88, 27.80, 25.93(3), 24.54, 22.58, 18.84, 18.03, 16.34, 14.07, –4.25, – 4.62.

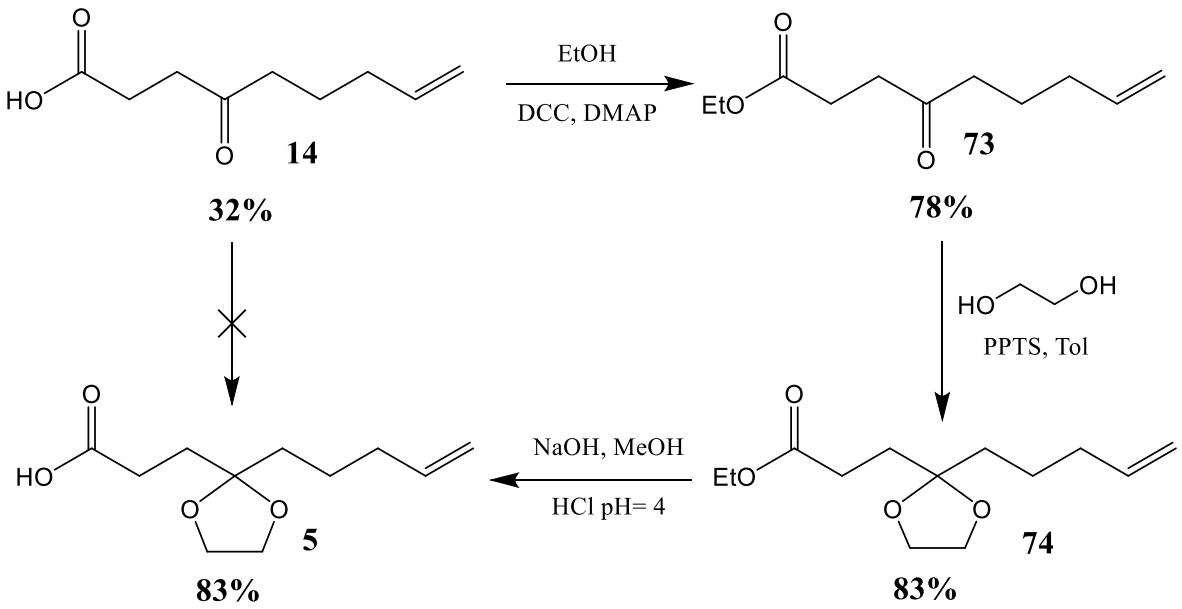
(4*S*)-1-nonen-4S-yl-2,3-di-*O*-levulinoyl-4,6-*O*-isopropylidene-*β*-D-glucopyranosyl-(1→2)-4-*O*-acetyl-3-*O*-tert-butyldimethylsilyl-*β*-D-fucopyranoside **65**. Acetic anhydride was added to a cold solution (-10 ^{o}C) of compound **7** (4.10 g, 5.15 mmol) and DMAP (314 mg, 2.1

mmol) in Py (35 mL). The reaction mixture was heated to 40°C and stirred overnight, when TLC (silica, Hex–EtOAc 2:1, R_f = 0.51) indicated that the reaction was complete. The reaction mixture was concentrated under reduced pressure, then co-evaporated with toluene (40 mL · 2). The residue was purified by column chromatography (silica, Hex–EtOAc, 4:1 → 1:1) to afford C-4-Fuc*p* acetylated intermediate **65** in 72% (3.10 g) as a colorless syrup. $[\alpha]^{25}_{D}$ –19.8° (*c* 1 $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_C 6.04 – 5.94 (m, 1H, CH_2=C*H*–CH_2–), 5.14 – 5.04 (m, 4H, C*H*$_2$=CH–CH_2–, H-1-Glu*p*, H-3-Glu*p*), 5.00 (m, 1H, H-4-Fuc*p*), 4.90 (dd, *J* = 9.2, 7.6 Hz, 1H, H-2-Glc*p*), 4.29 (d, *J* = 7.6 Hz, 1H, H-1-Fuc*p*), 3.98 (dd, *J* = 10.8, 5.2 Hz, 1H, H-6-Glu*p*), 3.88 (dd, *J* = 9.6, 8.0 Hz, 1H, H-2-Fuc*p*), 3.83 – 3.74 (m, 2H, H-6-Glc*p*, H-3-Fuc*p*), 3.68 (t, *J* = 9.6 Hz, 1H, H-4-Glc*p*), 3.64 – 3.57 (m, 2H, H-5-Fuc*p*, –CH_2–C*H*–CH_2–), 3.33 – 3.25 (m, 1H, H-5-Glc*p*), 2.86 – 2.53 (m, 8H, 2 · $CH_3C(O)CH_2CH_2C$=O), 2.35 – 2.27 (m, 2H), 2.17 (2s, 6H, 2 · $CH_3C(O)CH_2CH_2C$=O), 2.16 (s, 3H, C*H*$_3$–C=O), 1.59 – 1.48 (m, 2H), 1.47 (s, 3H, (C*H*$_3$)$_2$C), 1.41 – 1.21 (m, 9H, (C*H*$_3$)$_2$C), 1.12 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.95 – 0.84 (m, 12H), 0.14 (s, 3H, C*H*$_3$–Si), 0.10 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 206.39, 206.18, 171.92, 171.35, 170.75, 135.37, 116.35, 101.44, 99.67, 99.38, 80.35, 75.22, 73.42 · 3, 72.39, 71.51, 68.81, 67.36, 62.16, 38.33, 37.71 · 2, 34.19, 31.79, 29.77, 29.76, 28.86, 27.85 · 2, 25.81 · 3, 24.54, 22.58, 20.88, 18.82, 17.74, 16.46, 14.02, –4.46 · 2.

(4*S*)-1-nonen-4S-yl-2,3-di-*O*-levulinoyl-*β*-D-glucopyranosyl-(1→2)-4-*O*-acetyl-3-*O*-tert-butyldimethylsilyl-*β*-D-fucopyranoside **6**. CSA (140 mg, 0.63 mmol) was added in one portion to a solution of **65** (2.67 g, 3.16 mmol) in MeOH (50 mL) at room temperature. The reaction mixture was stirred for 3 hrs. at which point TLC (silica, Hex−EtOAc 1:1, R_f = 0.25) showed the starting material was gone. Water (0.5 mL) was added to the reaction mixture and stirred for another 20 min. The reaction was quenched with NEt_3 (few drops) and concentrated. The residue was purified

by column chromatography (silica, Hex–EtOAc, 2:1 to 1:1) gave diol **6** (2.31 g, 91%) as a colorless syrup. $[\alpha]^{25}_{D}$ −13.6° (*c* 1, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 6.01−5.86 (m, 1H, CH_2=C*H*–CH_2–), 5.12− 4.95 (m, 5H, C*H*$_2$=CH–CH_2–, H-1-Glc*p*, H-3-Glc*p*, H-4-Fuc*p*), 4.89 (dd, *J* = 9.6, 7.6 Hz, 1H, H-2-Glc*p*), 4.26 (d, J = 7.6 Hz, 1H, H-1-Fuc*p*), 3.96−3.83 (m, 2H, H-6-Glc*p*, H-2-Fuc*p*), 3.82−3.68 (m, 3H, H-4-Glc*p*, H-6-Glc*p*, H-3-Fuc*p*), 3.64−3.55 (m, 2H, H-5-Fucp, −CH_2−C*H*−CH_2−), 3.52 (d, *J* = 3.6 Hz, 1H, –O*H*), 3.44−3.35 (m, 1H, H-5-Glc*p*), 2.88−2.48 (m, 9H), 2.34−2.24 (m, 2H), 2.17 (s, 3H, C*H*$_3$C(O)CH_2CH_2C=O), 2.15 (s, 3H, C*H*$_3$C(O)CH_2CH_2C=O), 2.13 (s, 3H, C*H*$_3$–C=O), 1.60−1.42 (m, 2H), 1.37−1.20 (m, 6H), 1.11 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.89−0.81 (m, 12H), 0.12 (s, 3H, C*H*$_3$–Si), 0.09 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 208.2, 206.1, 173.0, 171.4, 170.8, 135.1, 116.7, 101.3, 98.7, 80.6, 76.2, 74.9, 74.8, 73.5, 73.3, 72.1, 69.6, 68.8, 61.9, 38.3 · 2, 37.6, 34.3, 31.7, 29.7 · 2, 28.0, 27.9, 25.7 · 3, 24.7, 22.5, 20.9, 17.7, 16.5, 14.0, −4.4, −4.6. HRMS (ESI) *m/z* calcd for $C_{39}H_{66}NaO_{15}Si$ [M + Na]$^+$ 825.4063, found 825.4061.

3.6. Synthesis of 4-acetyl-8-nonenoic acid



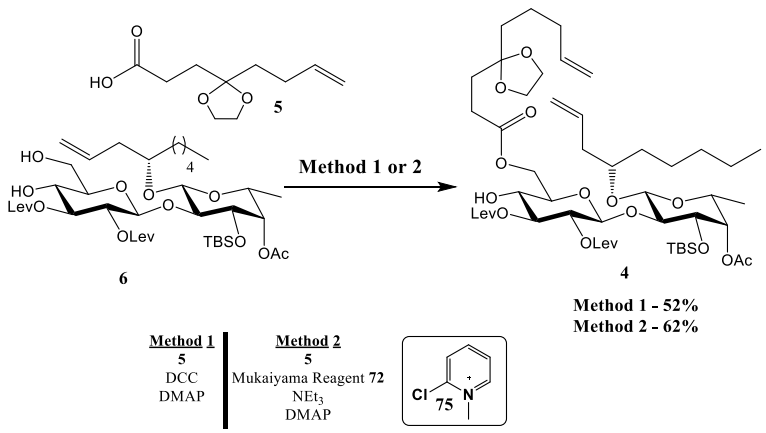
Monoethyl 8-nonenoate **73**. DCC (1.75 g, 8.47 mmol) was added in one portion to a 0ºC CH_2Cl_2 (20 mL) solution of 4-oxo-8-nonenoic acid **14** (1.20 g, 7.06 mmol), EtOH (2.1 mL, 35.3 mmol) and 4-dimethylaminopyridine (86 mg, 0.071 mmol). The reaction was allowed to warm to ambient temperature and stirred overnight. At this point, TLC (silica, Hex–EtOAc, 2:1) showed the reaction was complete. The reaction mixture was diluted with Et_2O (20 mL) and Hex (10 mL), stirred for 20 mins. then filtered through a pad of Celite using ether (10 mL) as the eluent and the filtrate concentrated *in vacuo*. The residue was purified by column chromatography (silica, Hex–EtOAc, 10:1→6:1) gave **73** (1.15 g, 79%) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.83 – 5.70 (m, 1H, CH_2=C*H*-CH_2-), 5.08 – 4.94 (m, 2H, C*H*$_2$=CH-CH_2-), 4.13 (dd, *J* = 14.4, 7.2 Hz, 2H), 2.72 (t, *J* = 6.4 Hz, 2H), 2.58 (t, *J* = 6.4 Hz, 2H), 2.47 (t, *J* = 7.4 Hz, 2H), 2.10 – 2.02 (m, 2H), 1.75 – 1.65 (m, 2H), 1.26 (t, *J* = 7.0 Hz, 3H, –C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 208.9, 172.8, 137.9, 115.2, 60.6, 41.9, 37.1, 33. 0, 28.0, 22.7, 14.1. HRMS (ESI) m/z calcd for $C_{11}H_{18}NaO_3$ $[M + Na]^+$ 221.1148, found 221.1151.

Monoethyl 4-acetal-8-nonenoate **74**. Ethyl 4-oxo-8-nonenoic ester **73** (914 mg, 4.61 mmol), ethylene glycol (2.32 mL, 41.5 mmol) and PPTS (174 mg, 0.69 mmol) were dissolved in toluene (20 mL). The reaction mixture was heated to reflux under Dean-Stark conditions overnight. The mixture was cooled to RT and extracted with sat. aq. $NaHCO_3$ (50 mL). The organic layer was dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex–EtOAc, 5:1 → 4:1) gave the acetal **74** (966 mg, 87%) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 5.85 – 5.70 (m, 1H, CH_2=C*H*–CH_2–), 5.04 – 4.90 (m, 2H, C*H*$_2$=CH–CH_2–), 4.11 (dd, *J* = 14.0, 7.2 Hz, 2H), 3.91 (s, 4H), 2.37 – 2.31 (m, 2H), 2.07 – 2.00 (m, 2H), 2.00 – 1.94 (m, 2H), 1.62 – 1.55 (m, 2H), 1.50 – 1.40 (m, 2H), 1.24 (t, *J* = 7.0 Hz, 3H, –CH_3). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 173.5, 138.4, 114.7, 110.7, 65.0 · 2, 60.2,

36.7, 33.7, 32.0, 28.9, 23.0, 14.2. HRMS (ESI) m/z calcd. for $C_{13}H_{22}NaO_4$ $[M + Na]^+$ 265.1410, found 265.1414.

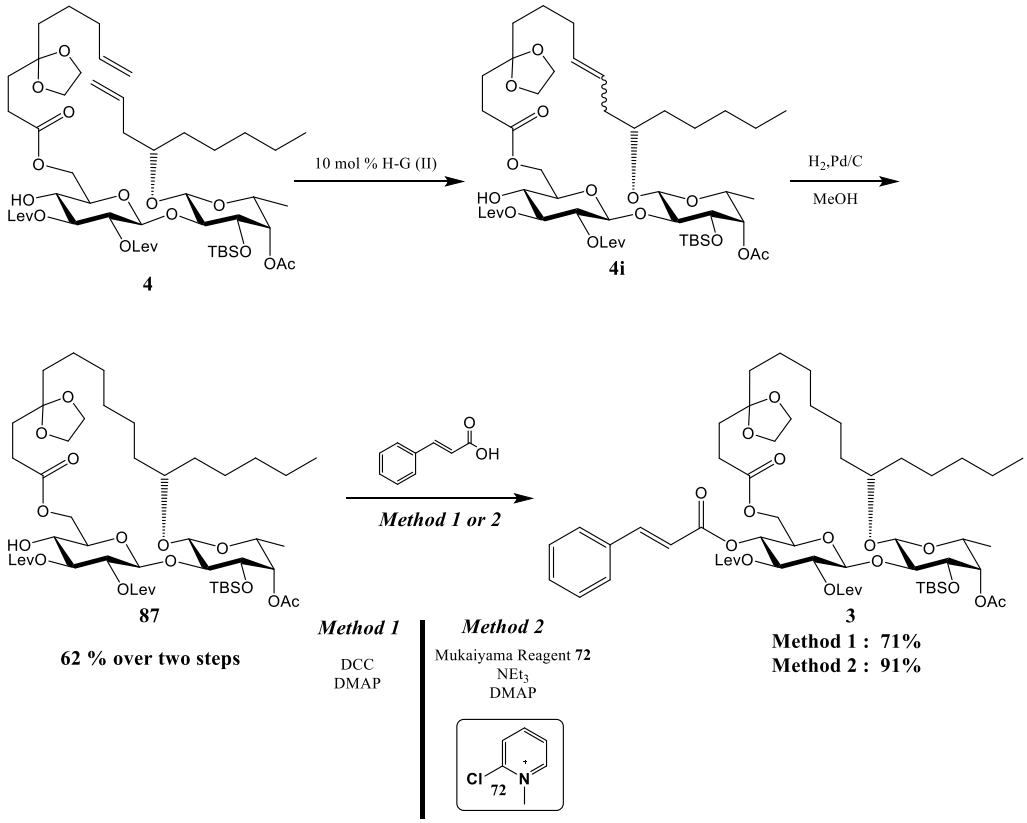
4-acetal-8-nonenoic acid **5**. To a solution of ethyl ester **74** (738 mg, 3.05 mmol) in MeOH (10 mL) was added NaOH (2 M solution in H_2O, 6.1 mL, 12.2 mmol) at 0 ºC. The mixture was allowed to slowly warm to RT and was stirred overnight. TLC analysis (silica, Hex–EtOAc, 1:1) showed it was complete. The mixture was concentrated under reduced pressure and the residue was dissolved in sat. aq. $NaHCO_3$ (20 mL) and extracted with EtOAc (40 mL · 2). Next, the aqueous layer was acidified with 10% w/v aq. Weak HCl (pH = 4) and extracted again twice with EtOAc. The latter organic layers were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex–EtOAc–AcOH, 2:1:0.05 → 1:1:0.05) gave compound **5** (550 mg, 82%) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 11.59 (br, 1H, COO*H*), 5.84 – 5.72 (m, 1H, CH_2=C*H*-CH_2–), 5.05 – 4.91 (m, 2H, CH_2=CH-CH_2–), 3.95 (s, 4H), 2.41 (t, *J* = 7.6 Hz, 2H), 2.10 – 1.96 (m, 4H), 1.64 – 1.55 (m, 2H), 1.52 – 1.40 (m, 2H). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 179.8, 138.4, 114.8, 110.7, 65.1 · 2, 36.8, 33.7, 31.8, 28.6, 23.0. HRMS (ESI) m/z calcd for C11H18NaO4 $[M + Na]^+$ 237.1097, found 237.1101.

3.7. Macrocycle Formation and Cinnamoyl Coupling



Diene **4**. *Method 1.* The (1→2)-*β*-disaccharide **6** (1.70 g, 2.11 mmol), 4-acetal-8-nonenoic acid **5** (0.50 g, 2.32 mmol), and DMAP (25 mg, 0.21 mmol) were dissolved in DCM (30 mL) and cooled to 0°C. Then, DCC (478 mg, 2.11 mmol) was added in one portion and the reaction was allowed to slowly warm to RT, overnight. TLC (silica, Hex–EtOAc 1:1, R_f = 0.59) showed the reaction was complete. The solvent was partially under reduced pressure, then diluted with Et_2O (40 mL) and Hex (20 mL), stirred for 15 min then filtered through a pad of Celite using ether (20 mL) as the eluent and the filtrate under reduced pressure. The residue was purified by column chromatography (silica, Hex−EtOAc, 5:1 → 1:1) gave diene **4** in 52% (1.64 g) as a colorless syrup. *Method 2.* The diol **6** (1.19 g, 1.48 mmol) was dissolved in the DCM, then added 1-methyl-2-chloropyrdium iodide (CMPI) (750 mg, 2.97 mmol), DMAP (90 mg, 0.7mmol), and the CA **5** (90 mg, 0.74 mmol). After the reaction was cooled to 0°C, then NEt_3 (2.1 mL, 14.8 mmol) was added and the reaction was allowed to slowly warm to RT over 1-2 hrs. At this point TLC showed the reaction to be complete (silica, Hex–EtOAc 1:1, R_f = 0.62). The reaction was concentrated under reduced pressure and purified by column chromatography (silica, Hex−EtOAc, 5:1 → 1:1) to give the diene **4** in 62%. $[\alpha]_D^{25}$ −18.0° (*c* 1, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 6.09−5.95 (m, 1H, CH_2=C*H*–CH_2–), 5.85−5.71 (m, 1H, CH_2=C*H*–CH_2–), 5.12−4.84 (m, 8H, 2 · C*H*$_2$=CH–CH_2–, H-1-Glc*p*, H-2-Glc*p*, H-3-Glc*p*, H-4-Fuc*p*), 4.46 (dd, *J* = 12.0, 3.8 Hz, 1H, H-6-Glc*p*), 4.32−4.23 (m, 2H, H-6-Glc*p*, H-1-Fuc*p*), 4.01−3.85 (m, 5H, H-2-Fuc*p*, −OC*H*$_2$C*H*$_2$O−), 3.79 (dd, *J* = 9.6, 3.8 Hz, 1H, H-3-Fuc*p*), 3.71−3.55 (m, 3H, H-4-Glc*p*, H-5-Fuc*p*, −CH_2−C*H*−CH_2−), 3.52−3.42 (m, 2H, H-5-Glc*p*, –O*H*), 2.86−2.49 (m, 8H), 2.45−2.35 (m, 2H), 2.32−2.27 (m, 2H), 2.17 (2s, 6H, 2 · C*H*$_3$C (O)CH_2CH_2C=O), 2.13 (s, 3H, C*H*$_3$–C=O), 2.09−1.98 (m, 4H), 1.62−1.41 (m, 6H), 1.38−1.20 (m, 6H), 1.12 (d, *J* = 6.5 Hz, 3H, H-6-Fuc*p*), 0.91−0.83 (m, 12H), 0.13 (s, 3H, C*H*$_3$–Si), 0.10 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 207.4, 206.1, 174.1, 172.9, 171.3,

170.7, 138.4, 135.6, 116.4, 114.8, 110.8, 101.5, 98.9, 80.7, 75.4, 75.0, 73.9, 73.6, 73.5, 72.4, 68.8, 68.7, 65.1, 65.0, 62.9, 38.5, 38.1, 37.7, 36.9, 34.2, 33.7, 32.1, 31.8, 29.8 · 2, 28.7, 28.0, 27.8, 25.8 · 3, 24.6, 23.0, 22.6, 20.9, 17.7, 16.5, 14.0, −4.4, −4.5. HRMS (ESI) m/z calcd. for $C_{50}H_{82}NaO_{18}Si$ $[M + Na]^+$ 1021.5163, found 1021.5150.



RCM Product **4i**. To a solution of diene **4** (2.41 g, 2.41 mmol) in DCM (800 mL) was added Hoveyda-Grubbs catalyst 2nd generation (151 mg, 0.24 mmol) in one portion at room temperature. The reaction mixture was refluxed (40 °C) overnight. At this point, TLC (silica, Hex–EtOAc 1:1, R_f =0.49) showed the reaction was complete. The reaction was cooled to ambient temperature and then concentrated. Flash chromatography (silica, Hex–EtOAc, 2:1 → 1:1) gave RCM products **4i** (1.91 g, 82%) as a colorless syrup which was not fully characterized. The

obtained isomers were subjected to hydrogenation in next step and the product was fully characterized.

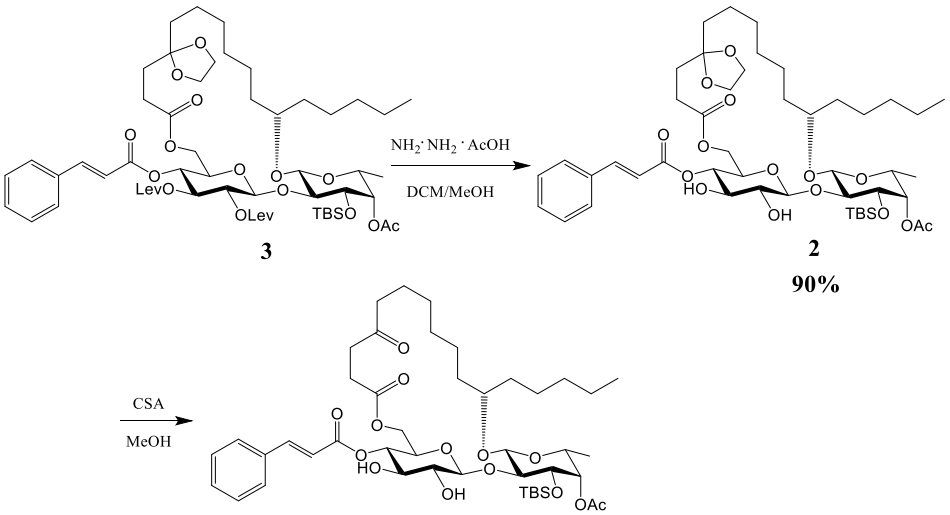
Hydrogenation Product **87**. The cis/trans RCM products **4i** (1.91 g, 1.97 mmol) were dissolved in EtOH (40 mL) was added 10% Pd/C (100 mg) in one portion at room temperature. The reaction was then stirred under an atmosphere of hydrogen overnight at the same temperature. At this point, TLC (silica, 1:1 Hex–EtOAc) showed the reaction was complete. The reaction mixture was filtered through a pad of Celite using EtOAc (10 mL) as the eluent and the resulting filtrate concentrated. Flash chromatography (silica, Hex:EtOAc, 2:1 → 1:1) gave **87** (1.55 g, 81 %) as a white foam. $[\alpha]_D^{25}$ –26.7° (*c* 1 $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 5.11 (d, *J* = 7.8 Hz, 1H, H-1-Glu*p*), 5.05 (t, *J* = 9.6 Hz, 1H, H-3-Glu*p*), 5.01 – 4.96 (m, 1H, H-4-Fuc*p*), 4.91 (dd, *J* = 12.6, 2.4 Hz, 1H, H-6-Glu*p*), 4.84 (dd, *J* = 9.6, 8.1 Hz, 1H, H-2-Glu*p*), 4.20 (d, *J* = 7.8Hz, 1H, H-1-Fuc*p*), 4.13 – 3.83 (m, 8H, H-4-Glu*p*, H-6-Glu*p*, H-2-Fuc*p*, H-3-Fuc*p*, –OC*H*$_2$C*H*$_2$O–), 3.68 – 3.31 (m, 4H, H-5-Glu*p*, H-5-Fuc*p*, O*H*, –CH_2-C*H*-CH_2–), 2.84 – 2.71 (m, 4H), 2.68 – 2.35 (m, 6H), 2.32 – 2.20 (m, 1H), 2.16 (s, 6H, C*H*$_3$–C=O), 2.10 (s, 3H, C*H*$_3$–C=O), 2.09 – 1.87 (m, 1H), 1.71 – 1.20 (m, 20H), 1.11 (d, *J* = 6.3 Hz, 3H, H-6-Fuc*p*), 0.95 – 0.81 (m, 12H), 0.17 (s, 3H, C*H*$_3$–Si), 0.13 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 206.9, 206.3, 175.5, 172.3, 171.3, 170.8, 111.0, 102.0, 98.9, 82.4, 75.0, 74.8, 74.6, 73.6, 73.5, 72.1, 68.7, 67.3, 64.6, 64.5, 62.3, 37.9, 37.7, 35.9, 34.5, 33.9, 31.9, 31.5, 29.8, 29.7, 29.0, 28.5, 27.9, 25.8 · 3, 25.0, 23.3, 22.6, 20.9, 17.6, 16.6, 14.0, –4.1, –4.6. HRMS (ESI) *m*/*z* calcd for $C_{48}H_{80}NaO_{18}Si$ $[M + Na]^+$ 995.5006, found 995.5012.

Compound **3**. *Method 1*. The macrocycle **87** (106 mg, 0.11 mmol) was dissolved in DCM (5-7 mL), cinnamic acid (40 mg, 0.17 mmol) and DMAP (3.2 mg, 0.01 mmol). The mixture was cooled to 0^{o}C and DCC (40 mg, 0.17 mmol) in one portion. The reaction was allowed to slowly

warm to RT overnight. TLC showed the reaction to be complete and the DCM was partially removed under reduced pressure. The mixture was diluted with Et_2O (4 mL) and Hex (2 mL) and stirred for 20 min. Then, filtered through a pad to Celite to remove the DCU and washed with Et_2O. The solvent was removed under reduced pressure and purified by column chromatography (silica, Hex−EtOAc, 3:1 → 1:2) to give the **87** in 71%. *Method 2.* CMPI (138 mg, 0.541 mmol), DMAP (22.0 mg, 0.180 mmol) and NEt_3 (0.50 mL, 3.6 mmol) were added to a solution of **87** (351 mg, 0.361 mmol) and cinn acid (80.0 mg, 0.539 mmol) in dry DCM (3 mL) at -10 °C. The reaction was allowed to slowly warm to ambient temperature and stirred overnight. At this point, TLC (silica, 2:1 Hex–EtOAc) showed the reaction was complete. The reaction mixture was diluted with DCM (15 mL) washed with sat. $NaHCO_3$ (10 mL) and extracted with DCM (15 mL). The combined organic extracts were dried over Na_2SO_4. Evaporation of the solvent followed by purification of the residue by column chromatography (silica, Hexanes−EtOAc, 3:1 → 1:2) to give **3** (345 mg, 88%) as a colorless syrup. $[\alpha]_D^{25}$ –16.9° (*c* 1 $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.64 (d, *J* = 15.9 Hz, 1H, Ph-C*H*=C-), 7.56 – 7.47 (m, 2H, 2 × Ar–*H*), 7.43 – 7.35 (m, 3H, 3 × Ar–*H*), 6.35 (d, *J* = 15.9 Hz, 1H, Ph–CH=C*H*-), 5.32 – 5.18 (m, 3H, H-3-Glc*p*, H-4-Glc*p*, H-1-Glc*p*), 5.04 – 4.92 (m, 2H, H-4-Fuc*p*, H-2-Glc*p*), 4.35 (dd, *J* = 12.3, 3.0 Hz, 1H, H-6-Glc*p*), 4.27 (d, *J* = 7.5 Hz, 1H, H-1-Fuc*p*), 4.12 (dd, *J* = 12.1, 2.4 Hz, 1H, H-6-Glc*p*), 4.04 – 3.83 (m, 6H, H-2-Fuc*p*, H-3-Fuc*p*, –OCH_2CH_2O–), 3.76 – 3.53 (m, 3H, H-5-Glu*p*, H-5-Fuc*p*, –CH_2–C*H*–CH_2–), 2.83 – 2.75 (m, 2H), 2.70 – 2.31 (m, 8H), 2.17 (s, 3H, CH_3C=OCH_2CH_2C=OO), 2.11 (s, 3H, CH_3-C=OO), 2.08 (s, 3H, CH_3–COCH_2CH_2C=OO), 2.05 – 1.84 (m, 2H), 1.73 – 1.21 (m, 20H), 1.13 (d, *J* = 6.3 Hz, 3H, H-6-Fuc*p*), 0.95 – 0.83 (m, 12H), 0.19 (s, 3H, CH_3–Si), 0.14 (s, 3H, CH_3–Si). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 206.2, 206.1, 173.0, 171.9, 171.2, 170.8, 165.0, 146.1, 134.1, 130.5, 128.8 · 2, 128.3 · 2, 116.7, 111.3, 103.5, 101.4, 98.9, 81.9, 74.5, 74.1, 73.6, 73.0, 72.2, 71.9, 68.3, 68.5,

64.5, 61.7, 37.8, 37.7, 34.6, 34.3, 33.7, 32.0, 31.2, 30.0, 29.8, 29.6, 29.0, 28.3, 27.9, 27.8, 25.9 · 3, 24.6, 24.0, 22.9, 22.6, 20.9, 17.8, 16.6, 14.1, –4.3, –4.5. HRMS (ESI) m/z calcd for $C_{57}H_{86}NaO_{19}Si$ $[M + Na]^+$ 1125.5425, found 1125.5414.

3.8. De-protection of the *C-2* and *C-3* Glucosyl Lev Groups and the *C-4* Cyclic Acetal



3

2
90%

71
75%

Compound **2.** Hydrazine acetate (0.350 g, 3.70 mmol) was added in one portion to a solution of intermediate **3** (0.680 mg, 0.620 mmol) in 2:1 DCM/MeOH (6 mL) at room temperature. The reaction mixture was stirred for 0.5 hr, at which point TLC (silica, 2:1 Hex–EtOAc, R_f = 0.51) showed the reaction was complete. Then the reaction was quenched with sat. aq. $NaHCO_3$ (10 mL) and extracted with DCM (15 mL · 2). The combined organic extracts were dried over Na_2SO_4, evaporated and purified by column chromatography (silica, Hex–EtOAc, 3:1→ 1:1) to afford compound **2** (520 mg, 90%) as a colorless syrup. $[\alpha]^{25}_D$ 0.8° (*c*1 $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.73 (d, *J* = 16.0 Hz, 1H, Ph–C*H*=C–), 7.56 – 7.50 (m, 2H, 2 · Ar–*H*), 7.42 – 7.37 (m, 3H, 3 · Ar–*H*), 6.46 (d, *J* = 16.0 Hz, 1H, Ph–CH=C*H*–), 5.15 – 5.06 (m, 2H, H-4-Glu*p*,

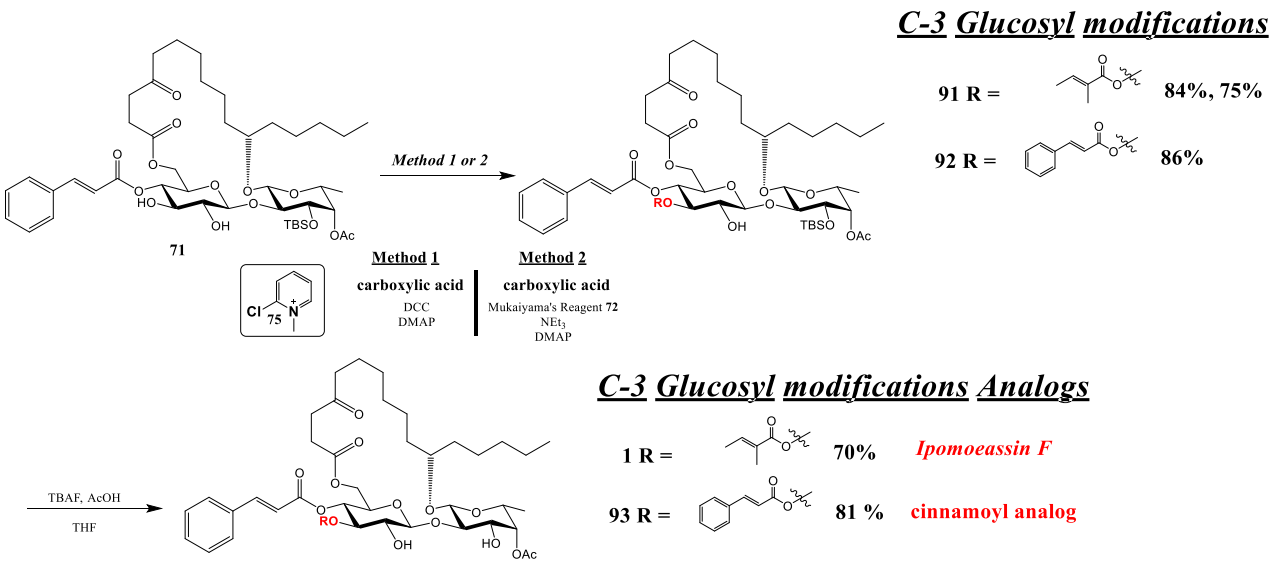
H-4-Fuc*p*), 4.65 (d, *J* = 7.6 Hz, 1H, H-1-Glc*p*), 4.41 (d, *J* = 7.6 Hz, 1H, H-1-Fuc*p*), 4.26 (dd, *J* = 12.0, 4.0 Hz, 1H, H-6-Glc*p*), 4.18 (dd, *J* = 12.0, 4.0 Hz, 1H, H-6-Glc*p*), 3.98 – 3.60 (m, 11H, H-3-Glu*p*, H-5-Glc*p*, H-2-Fuc*p*, H-3-Fuc*p*, H-5-Fuc*p*, O*H*, –OC*H*$_2$C*H*$_2$O–, -CH$_2$–C*H*–CH$_2$–), 3.50 – 3.41 (m, 1H, H-2-Glu*p*), 2.64 (d, *J* = 2.4 Hz, 1H, –O*H*), 2.40 – 2.25 (m, 2H), 2.13 (s, 3H, C*H*$_3$–C=O), 2.05 – 1.86 (m, 2H), 1.61 – 1.48 (m, 6H), 1.41 – 1.20 (m, 14H), 1.15 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.95 – 0.86 (m, 12H), 0.22 (s, 3H, C*H*$_3$–Si), 0.18 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 173.1, 170.7, 166.2, 146.2, 134.1, 130.5, 128.9 · 2, 128.2 · 2, 117.1, 111.2, 103.5, 101.1, 79.8, 78.5, 76.0, 73.6, 73.0, 72.7, 72.5, 71.3, 68.7, 64.6, 64.5, 63.5, 35.1, 33.9, 33.3, 32.0, 31.3, 29.7, 28.9, 28.4, 25.9 · 3, 24.3, 24.0, 22.6 · 2, 20.8, 17.9, 16.5, 14.1, –4.3, –4.6. HRMS (ESI) *m*/*z* calcd for $C_{47}H_{74}NaO_{15}Si$ $[M + Na]^+$ 929.4689, found 929.4692.

Compound **71**. CSA (58.9 mg, 0.254 mmol) was added in one portion to a solution of **2** (1.15 g, 1.27 mmol) in MeOH (30 mL) and H_2O (0.3 mL) at room temperature. The reaction mixture was stirred for 12 h at which point TLC (silica, 1:1 Hex−EtOAc, $R_{f=}$ 0.51) showed it was complete. The reaction was quenched with NEt_3 (70 μL, few drops) and concentrated. The residue was purified by column chromatography (silica, Hex−EtOAc, 1:3 → 1:2) gave compound **4** (72%) as a colorless syrup. $[\alpha]^{25}_D$ −13.4° (*c* $CHCl_3$). ^{1}H NMR (400 MHz, CDCl3) δ_H 7.77 (d, *J* = 16.0 Hz, 1H, Ph–C*H* =C–), 7.58−7.51 (m, 2H, 2 ·Ar–*H*), 7.43−7.37 (m, 3H, 3 ·Ar–*H*), 6.51 (d, *J* = 16.0 Hz, 1H, Ph–CH=C*H*), 5.16−5.08 (m, 2H, H-4- Glc*p*, H-4-Fuc*p*), 4.59 (d, *J* = 7.6 Hz, 1H, H-1-Glc*p*), 4.40 (d, *J* = 7.6 Hz, 1H, H-1-Fuc*p*), 4.34 (dd, *J* = 12.0, 2.4 Hz, 1H, H-6-Glc*p*), 4.19 (dd, *J* = 12.0, 5.2 Hz, 1H, H-6-Glc*p*), 4.02 (d, *J* = 1.6 Hz, 1H, –OH), 3.96−3.89 (m, 1H, H-2-Fuc*p*), 3.85 (dd, *J* = 9.6, 3.2 Hz, 1H, H-3- Fuc*p*), 3.83−3.75 (m, 1H, H-3-Glc*p*), 3.73−3.60 (m, 3H, H-5-Glc*p*, H-5-Fuc*p*, −CH$_2$−CH−CH$_2$−), 3.48−3.40 (m, 1H, H-2-Glc*p*), 2.75− 2.40 (m, 6H), 2.14 (s, 3H, CH$_3$–C=O), 1.70−1.61 (m, 2H), 1.59− 1.45 (m, 4H), 1.39−1.20 (m, 12H), 1.15 (d, *J* = 6.0 Hz, 3H,

H-6- Fuc*p*), 0.95−0.84 (m, 12H), 0.22 (s, 3H, CH_3-Si), 0.19 (s, 3H, CH_3-Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ 210.3, 171.6, 170.7, 166.2, 146.1, 134.2, 130.6, 129.0 ·2, 128.2 ·2, 117.2, 104.0, 101.2, 80.3, 79.0, 76.1, 73.5, 73.1, 72.8, 72.2, 71.6, 68.8, 63.3, 42.0, 37.3, 34.1, 33.3, 32.0, 29.5, 29.1, 28.5, 25.9 3, 25.1, 24.4, 23.6, 22.6, 20.9, 17.9, 16.5, 14.1, −4.3, −4.6. HRMS (ESI) *m*/*z* calcd for $C_{45}H_{70}NaO_{14}Si$ $[M + Na]^+$ 885.4427, found 885.4432.

3.9. Ipomoeassin F and C-3-Glucosyl Modified Analogs

3.9.1. DCC/DMAP Esterification and -OTBS De-protection Method



Ipomoeassin F **1**. Compound **91**. *Method 1*. DCC (14.3 mg, 69.6 μmol) was added in one portion to a 0 °C DCM (3-4 mL) solution of **71** (30 mg, 34.8 μmol), tig acid (7.0 mg, 69.6 μmol) and DMAP (0.5 mg, 3.5 μmol). The reaction was warmed to ambient temperature and stirred overnight at RT. The TLC (silica, 3:1 Hex−EtOAc, R_f = 0.41) showed the reaction was complete. The reaction mixture was diluted with Et_2O (2 mL) and Hex (1 mL), stirred for 20 min then filtered through a pad of Celite using Et_2O (5 mL) as the eluent and the filtrate concentrated in vacuo. The

residue was purified by column chromatography (silica, Hex−EtOAc, 6:1 → 3:1) in 81% (26 mg) as a colorless syrup. $[\alpha]^{25}_{D}$ −38.7° (*c* 0.5, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.67 (d, *J* = 16.0 Hz, 1H, Ph–C*H*=C–), 7.55−7.48 (m, 2H, 2 Ar–*H*), 7.43−7.36 (m, 3H, 3 ·Ar–*H*), 6.87−6.78 (m, 1H, Me−C*H*−C (Me) –C=O), 6.38 (d, *J* = 16.0 Hz, 1H, Ph–CH=C*H*–), 5.33 (t, *J* = 9.6 Hz, 1H, H-3-Glc*p*), 5.23 (t, *J* = 9.6 Hz, 1H, H-4-Glc*p*), 5.10−5.08 (m, 1H, H-4-Fuc*p*), 4.70 (d, 1H, *J* = 8.0 Hz, H-1-Glc*p*), 4.38 (d, 1H, *J* = 7.6 Hz, H-1-Fuc*p*), 4.31−4.18 (m, 2H, H-6-Glc*p*), 3.94 (dd, *J* = 7.6, 10 Hz, 1H, H-2-Fuc*p*), 3.89− 3.80 (m, 2H, –O*H*, H-3-Fuc*p*), 3.78−3.72 (m, 1H, H-5-Glc*p*), 3.71− 3.55 (m, 3H, H-2-Glc*p*, H-5-Fuc*p*, −CH_2−C*H*−CH_2−), 2.70−2.42 (m, 6H), 2.11 (s, 3H, C*H*$_3$–C=O), 1.79−1.63 (m, 8H), 1.60−1.43 (m, 4H), 1.40−1.20 (m, 12H), 1.14 (d, *J* = 6.4 Hz, 3H, H-6-Fuc*p*), 0.94−0.83 (m, 12H), 0.22 (s, 3H, C*H*$_3$–Si), 0.18 (s, 3H, C*H*$_3$–Si). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 210.1, 171.4, 170.8, 167.5, 165.6, 146.0, 138.1, 134.0, 130.6, 128.9 ·2, 128.2 ·2, 127.9, 116.8, 104.2, 101.2, 80.2, 79.0, 74.5, 72.9, 72.8, 72.0, 69.9, 68.8, 63.3, 42.0, 37.4, 34.1, 33.2, 32.0, 29.6, 29.0, 28.6, 25.8 ·3, 25.1, 24.4, 23.7, 22.6, 20.9, 17.9, 16.5, 14.4, 14.1, 12.0, −4.4, −4.6. HRMS (ESI) m/z calcd for $C_{50}H_{76}NaO_{15}Si$ $[M + Na]^+$ 967.4846, found 967.4834. *Method 2.* CMPI **71** (16.3 mg, 67.1 μmol), DMAP (0.5 mg, 0.007 mmol) and NEt_3 (48 μL, 0.330 mmol) were added to a solution of **71** (29.0 mg, 33.6 μmol) and tig acid (6.5 mg, 67.1 μmol) in dry DCM (4 mL) at 0 °C. The reaction was allowed to warm to ambient temperature and stirred for 4 h. At this point, TLC (silica, Hex−EtOAc, 2:1 R_f =0.21) showed the reaction was complete. Evaporation of the solvent followed by purification of the residue by column chromatography (silica, Hex−EtOAc, 5:1 → 3:1) to give **91** (25.4 mg, 79%) as a colorless syrup. The ^{1}H and ^{13}C NMR data of **91** obtained via method 2 were identical to those of **91** obtained via method 1.

Ipomoeassin F **1**. To a solution of **91** (20 mg, 22.6 μmol) in THF (2 mL) was added AcOH (67 μL, 2.23 mmol) and TBAF (1 M solution in THF, 0.58 mL, 1.06 mmol) at 0 °C. The reaction

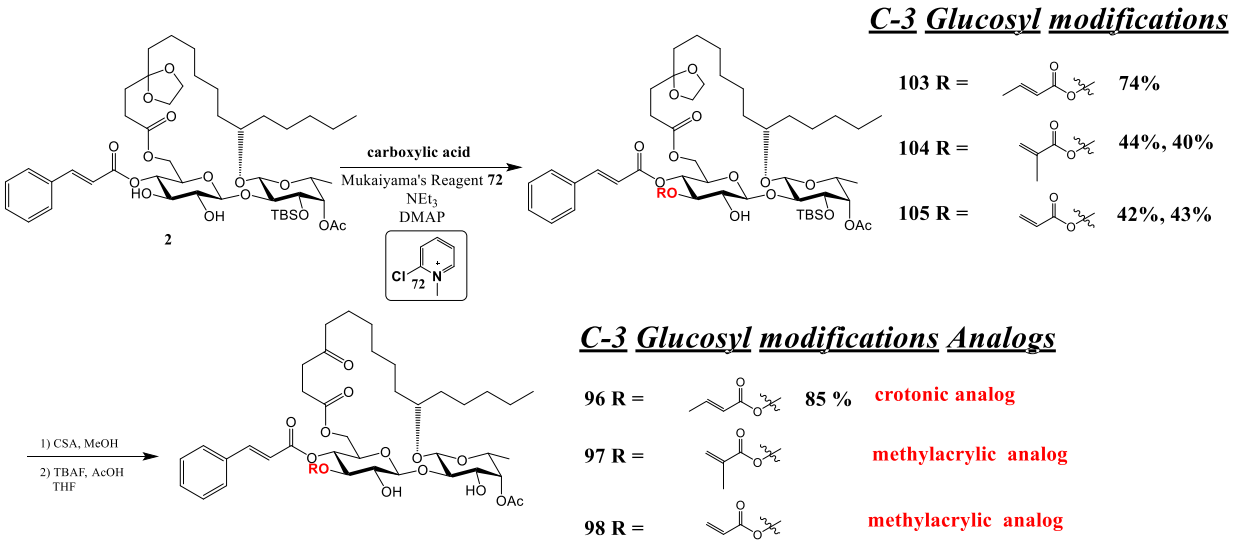
was allowed to warm to ambient temperature and stirred overnight. At this point, TLC (silica, Hex−EtOAc, 1:1, R_f = 0.15) showed the reaction was complete. The reaction mixture was concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex−EtOAc, 2:1 → 1:1) gave synthetic ipomoeassin F **1** (12.1 mg, 70%) as a colorless film. $[\alpha]^{D}_{25}$ −51.2° (*c* $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.63 (d, *J* = 16.0 Hz, 1H, Ph−C*H*=C−), 7.55−7.45 (m, 2H, 2 ·Ar−*H*), 7.45−7.35 (m, 3H, 3 · Ar−*H*), 6.93−6.87 (m, 1H, CH_3−C*H*−CCH_3−C=O), 6.35 (d, *J* = 16.0 Hz, 1H, Ph−CH=C*H*−), 5.33 (t, *J* = 9.6 Hz, 1H, H-4-Glc*p*), 5.18−5.11 (m, 2H, H-3-Glc*p*, H-4-Fuc*p*), 4.62 (d, *J* = 7.6 Hz, 1H, H-1-Glc*p*), 4.56 (d, *J* = 1.6 Hz, 1H, −O*H*), 4.47 (dd, *J* = 12.4, 3.6 Hz, 1H, H-6-Glc*p*), 4.41 (d, *J* = 8.0 Hz, 1H, H-1-Fuc*p*), 4.15 (dd, *J* = 12.4, 2.4 Hz, 1H, H-6-Glc*p*), 3.96−3.88 (m, 2H, −O*H*, H-3-Fuc*p*), 3.78−3.73 (m, 1H, H-5-Glc*p*), 3.73−3.59 (m, 4H, H-2-Glc*p*, H-2-Fuc*p*, H-5-Fuc*p*, −CH_2−C*H*−CH_2−), 2.85−2.38 (m, 6H), 2.18 (s, 3H, C*H*$_3$−C=O), 1.80−1.72 (m, 6H, C*H*$_3$−CH−C (C*H*$_3$)−C=O), 1.72−1.62 (m, 2H), 1.58− 1.46 (m, 4H), 1.41−1.22 (m, 12H), 1.19 (t, *J* = 6.5 Hz, 3H, H-6-Fuc*p*), 0.90 (t, *J* = 6.8 Hz, 3H, −C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 210.0, 171.8, 171.7, 168.9, 165.4, 146.2, 139.9, 134.0, 130.6, 128.9 ·2, 128.3 · 2, 127.5, 116.7, 105.7, 100.2, 82.9, 79.8, 75.8, 74.0, 72.7, 72.6, 72.6, 68.8, 67.4, 61.8, 41.9, 37.6, 34.4, 33.1, 31.9, 29.1, 29.0, 28.4, 24.7, 24.5, 23.5, 22.7, 20.9, 16.3, 14.6, 14.1, 12.0. The ^{1}H, ^{13}C NMR data were in accordance with the literature. [47-49]

Cinnamoyl Analog **93**. Compound **92**. *Method 1*. DCC (5.7 mg, 27.8 μmol) was added in one portion to a 0 °C DCM (3-4 mL) solution of **71** (16 mg, 18.5 μmol), cinn acid (4.1 mg, 27.8 μmol) and DMAP (0.3 mg, 2.8 μmol). The reaction was allowed to warm to ambient temperature and stirred overnight at RT. The TLC (silica, 3:1 Hex−EtOAc, R*f* = 0.51) showed the reaction was complete. The reaction mixture was diluted with diethyl ether (2 mL) and hexanes (1 mL), stirred for 20 min then filtered through a pad of Celite using diethyl ether (5 mL) as the eluent and

the filtrate concentrated in vacuo. The residue was purified by column chromatography (silica, Hex−EtOAc, 6:1 → 3:1) in 86% **92** (15.3 mg) as a colorless syrup. *Method 2*. CMPI (13.0 mg, 0.051 mmol), DMAP, 9.3 mg, 0.076 mmol) and NEt_3 (36 μL, 0.25 mmol) were added to a solution of **71** (22.0 mg, 0.025 mmol) and cinnamic acid (7.6 mg, 0.051 mmol) in dry CH2Cl2 (2 mL) at 0 °C. The reaction was allowed to warm to ambient temperature and stirred for 1 hr. At this point, TLC (silica, 2:1 Hex−EtOAc) showed the reaction was complete. Evaporation of the solvent followed by purification of the residue by column chromatography (silica, Hex−EtOAc, 5:1 → 3:1) to give **92** (20.4 mg, 81%) as a colorless syrup. Then to a solution of **92** (11.6 mg, 11.7 μmol) in THF (2 mL) was added AcOH (67 μL, 11.7 μmol) and TBAF (1 M solution in THF, 0.58 mL, 58.o mmol) at 0 °C. The reaction was allowed to warm to ambient temperature and stirred overnight. At this point, TLC (silica, Hex:EtOAc, 1:1 R_f = 0.15) showed the reaction was complete. The reaction mixture was concentrated under reduced pressure. The residue was purified by column chromatography (silica, Hex−EtOAc, 2:1 → 1:2) gave the cinnamoyl analog (8.0 mg, 78%) as a white powder. $[\alpha]^{D}_{25}$ −127° (*c* 0.2, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.68 (d, *J* = 16.0 Hz, 1H, Ph−C*H*=C−), 7.64 (d, *J* = 16.0 Hz, 1H, Ph−C*H*=C−), 7.53− 7.43 (m, 4H, 4 ·Ar–*H*), 7.39−7.31 (m, 6H, 6 ·Ar–*H*), 6.41 (d, *J* = 16.0 Hz, 1H, Ph−CH=C*H*), 6.35 (d, *J* = 16.0 Hz, 1H, Ph−CH=C*H*−), 5.33 (t, *J* = 9.5 Hz, 1H, H-4-Glc*p*), 5.28 (t, *J* = 9.4 Hz, 1H, H3-Glc*p*), 5.14 (d, *J* = 3.2 Hz, 1H, H-4-Fuc*p*), 4.65 (d, *J* = 8.1 Hz, 1H, H-1-Glc*p*), 4.55−4.41 (m, 3H, H-6-Glc*p*, H-1-Fuc*p*, −O*H*), 4.18 (dd, *J* = 12.4, 2.0 Hz, 1H, H-6-Glc*p*), 3.95 (dd, *J* = 9.6, 3.6 Hz, 1H, H-3-Fuc*p*), 3.83−3.62 (m, 6H, H-2-Glc*p*, H-5-Glc*p*, H-2-Fucp, H-5-Fucp, $-CH_2-CH-CH_2-$, −O*H*), 2.85−2.40 (m, 6H), 2.19 (s, 3H, CH_3−C=O), 1.72−1.22 (m, 18H), 1.20 (d, *J* = 6.5 Hz, 3H, H-6-Fuc*p*), 0.90 (t, *J* = 6.7 Hz, 3H, −C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 210.0, 172.0, 171.7, 167.3, 165.5, 146.6, 146.3, 134.1, 134.0, 130.6, 130.6, 128.9 ·2, 128.8 ·2, 128.3 2, 128.3 2, 116.7, 116.6,

105.6, 100.3, 82.4, 79.7, 75.5, 73.8, 72.8, 72.7, 72.5, 68.8, 67.8, 61.9, 41.9, 37.6, 34.3, 33.0, 31.9, 29.1, 29.0, 28.3, 24.6, 24.5, 23.4, 22.7, 20.9, 16.3, 14.1. HRMS (ESI) m/z calcd for $C_{48}H_{62}NaO_{15}$ $[M + Na]^+$ 901.3981, found 901.3978.

3.9.2. Modified Synthesis Route of Mukaiyama Esterification and One-Pot De-Protection



Crotonic Analog **96**. CMPI **75** (18.6 mg, 0.0721 mmol), DMAP (2.2 mg, 0.018 mmol) and NEt_3 (0.05 mL, 0.320 mmol) were added to a solution of **2** (29.8 mg, 0.320 mmol) and crotonic acid (6.2 mg, 0.072 mmol) in dry DCM (2 mL) at -10 ^oC. The reaction was allowed to slowly warm to ambient temperature and stirred overnight. The TLC showed the reaction was 60% complete. So, the reaction was left to stir for 24 hrs, when the TLC (silica, 2:1 Hexanes–EtOAc) showed the reaction was complete. The reaction mixture was diluted with DCM (5 mL) in a centrifuge tube, washed with sat. $NaHCO_3$ (10 mL) and extracted with DCM (5 mL · 2). The combined organic extracts were dried over Na_2SO_4. Evaporation of the solvent followed by purification of the residue by column chromatography (silica, Hexanes−EtOAc, 3:1 → 1:1) to give **103** in 74% (24 mg, 74%) as a foamy syrup. **103** was not fully characterized and all the protecting group removed in the following transformations. CSA (2.50 mg, 0.0108 mmol) was added to a

solution of MeOH (2 mL) and the **103** (20.9 mg, 0.0215 mmol). The reaction was heated to 30 ^{o}C and stirred for 12 hrs to remove the *C-4* cyclic acetal. Then, acetic acid (61 μL, 1.08 mmol) and TBAF (1 M in THF, 0.538 μL) were added to the reaction and stirred at 30 ^{o}C for 12 hrs. The TLC (1:1Hex−EtOAc, R_f = 0.15) showed the reaction was complete. The reaction mixture was diluted with DCM (3 mL) in a centrifuge tube, washed with water (3 mL), and extracted with DCM (4 mL). The combined organic extracts were dried over Na_2SO_4 and concentrated. Column chromatography (silica, Hex−EtOAc, 2:1 → 1:1, DCM−MeOH, 100:1 → 20:1) was utilized to purify the crotonic analog **96** (16 mg, 85%) as a clear syrup. ^{1}H NMR (400 MHz, $CDCl_3$) δ_H 7.64 (d, *J* = 16.0 Hz, 1H, Ph−C*H*=C−), 7.52−7.50 (m, 2H, 2 ·Ar–*H*), 7.40−7.39 (m, 3H, 3 Ar–*H*), 6.93−6.87 (m, 1H, Me−C*H*−CH−C=O), 6.35 (d, *J* = 16.0 Hz, 1H, Ph−CH=C*H*−), 5.82 (d, *J* = 17.2 Hz, 1H, Me−CH−C*H*−C=O), 5.27 (t, *J* = 19.6 Hz, 1H, H-4-Glc*p*), 5.19 (t, *J* = 9.2 Hz, 1H, H-3-Glc*p*), 5.15 (d, *J* = 4.5 Hz, H-4-Fuc*p*), 4.61 (d, *J* = 7.6 Hz, 1H, H-1-Glc*p*), 4.63-4.41 (m, −O*H*, H-1-Fuc*p*), 4.13 (dd, *J* = 12.4, 3.6 Hz, 1H, H-6-Glc*p*), 3.93 (dd, *J* = 12.4, 2.4 Hz, 1H, H-6-Glc*p*), 3.76 (m, 6H, −OH, H-3-Fuc*p*, H-5-Glcp, H-2-Fuc,−CH_2−C*H*−CH_2), 2.82-2.41 (m, 5 H) 2.85−2.38 (m, 6H), 2.18 (s, 3H, C*H*$_3$−C=O), 1.83 (d, *J* = 6.8 Hz 3H, C*H*$_3$−CH−CH−C=O), 1.72−1.62 (m, 2H), 1.58− 1.46 (m, 4H), 1.41−1.22 (m, 12H), 1.19 (t, *J* = 6.5 Hz, 3H, H-6-Fuc*p*), 0.90 (t, *J* = 6.8 Hz, 3H, −C*H*$_3$). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 210.0, 171.8, 171.7, 168.9, 165.4, 146.2, 139.9, 134.0, 130.6, 128.9 · 2, 128.3 · 2, 127.5, 116.6, 105.5, 100.1, 82.9, 79.8, 75.8, 74.0, 72.7, 72.6, 72.6, 68.8, 67.4, 61.8, 41.9, 37.6, 34.4, 33.1, 31.9, 29.1, 29.0, 28.4, 24.7, 24.5, 23.5, 22.7, 20.9, 16.3, 14.1. Unfortunately, the crotonic analog **87** was not good enough to publish by this method.

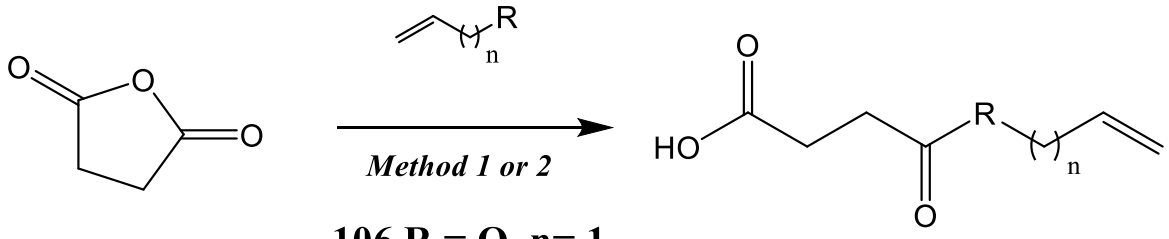
Methylacrylic Analog **97**. The diol **2** (37 mg, 40.8 μmol) was dissolved in DCM (2-3 mL), then CMPI **74** (26.0 mg, 102.0 μmol), methylacrylic acid (16 μL, 102 μmol), and DMAP (2.5 mg, 21 μmol). NEt_3 was added to the reaction mixture in one portion at 0^{o}C and reaction was slowly

allowed to warm to RT overnight. TLC (Hex−EtOAc, 1:1 R_f = 0.15) showed the reaction to complete to be complete. The reaction mixture was diluted with DCM (3 mL) and washed with water and extracted with DCM (5 mL). The combined organic layers were dried over Na_2SO_4 and concentrated. The residue purified though column chromatography (Hex−EtOAc, 2:1 → 1:1, DCM−MeOH, 100:1 → 20:1) to give **104** in 44% (19 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.65 (d, *J* = 16.0 Hz, 1H), 7.52-7.49 (m, 2H), 7.39- 7.37 (m, 3H), 6.36 (d, *J* = 16.2 Hz, 1H), 6.07 (s, 1H), 5.52 (s, 1H), 5.34 (t, *J* = 9.6, 1H), 5.39 (t, *J* = 9.6, 1H), 5.08 (d, *J* = 3.0 Hz, 1H), 4.76 (d, *J* = 7.8 Hz, 1H), 4.41 (d, *J* = 7.5 Hz, 1H), 4.21 (m, 2H), 3.96 (t, *J* = 9.9 Hz, 1H), 3.85 (m, 6H), 3.82 (t, *J* = 3.6 Hz, 1H), 3.69 (t, *J* =5.4 Hz, 1H), 3.62 (m, 2H), 2.34 (m, 2H), 2.12 (s, 3H), 1.87 (s, 3H), 1.56 (m, 8H), 1.34 (m, 18H), 1.14 (d, *J* = 6.3 Hz), 0.91-0.87 (m, 12H), 0.21 (s, 3H), 0.17 (s, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 173.2, 170.8, 166.8, 165.6, 146.2, 135.6, 134.0, 130.5, 128.9 ·2, 128.3 · 2, 126.2, 116.7, 111.3, 103.0, 101.01, 79.5, 78.7, 77.2, 74.3, 73.3, 72.9, 72.7, 72.4, 69.4, 68.8, 64.6, 64.5, 63.4, 36.6, 35.1, 33.9, 33.2, 31.9, 29.7, 29.0, 28.5, 25.8 · 3, 24.6, 24.3, 24.0, 22.7, 22.6, 20.9, 18.2, 17.8, 16.5, 14.1, -4.4, -4.6. **104** was not fully characterized before going to next step. CSA (2.50 mg, 0.0108 mmol) was added to a solution of MeOH (2 mL) and the **104** (20.9 mg, 0.0215 mmol). The reaction was heated to 30^{o}C and stirred for 12 hrs. to remove the *C-4* cyclic acetal. Then, acetic acid (61 μL, 1.08 mmol) and TBAF (1 M in THF, 0.538 μL) were added to the reaction and stirred at 30^{o}C for 12 hrs. The TLC (1:1 Hex−EtOAc, R_f = 0.15) showed the reaction was complete. The reaction mixture was diluted with DCM (3 mL) in a centrifuge tube, washed with water (3 mL), and extracted with DCM (4 mL). The combined organic extracts were dried over Na_2SO_4 and concentrated. Column chromatography (silica, Hexanes−EtOAc, 2:1 → 1:1, DCM−MeOH, 100:1 → 20:1) was utilized to purify the **97** methylacrylic analog (16 mg, 85%)

as a clear syrup. A complex mixture of signals was observed in the H-NMR and C-NMR of the methylacrylic analog **97**.

Acrylic Analog **98**. The diol **2** (30 mg, 33.1 μmol) was dissolved in DCM (2-3 mL), then CMPI **75** (21.0 mg, 82.8 μmol), acrylic acid (6 μL, 82.8 μmol), and DMAP (2.0 mg, 16.6 μmol). NEt_3 (0.46 mL, 33.1 μmol) was added to the reaction mixture in one portion at 0^{o}C and reaction was slowly allowed to warm to RT overnight. TLC (Hex−EtOAc, 1:1 R_f = 0.15) showed the reaction to complete to be complete. The reaction mixture was diluted with DCM (3 mL) and washed with water and extracted with DCM (5 mL). The combined organic layers were dried over Na_2SO_4 and concentrated. The residue purified though column chromatography (Hex−EtOAc, 2:1 → 1:1, DCM−MeOH, 100:1 → 20:1) to give **105** in 44% (14 mg). The **105** was not characterized and proceeded directly to the next step. CSA (2.2 mg, 0.0108 mmol) was added to a solution of MeOH (2 mL) and **105** (15 mg, 14.6 mol). The reaction was heated to 35 ^{o}C and stirred for 12 hrs. to remove the *C-4* cyclic acetal. Then, acetic acid (44 μL, 730 μmol) mol) and TBAF (1 M in THF, 0.384 μL) were added to the reaction and stirred at 30^{o}C for 12 hrs. The TLC (1:1 Hex−EtOAc, R_f = 0.15) showed the reaction was complete. The reaction mixture was diluted with DCM (3 mL) in a centrifuge tube, washed with water (3 mL), and extracted with DCM (4 mL). The combined organic extracts were dried over Na_2SO_4 and concentrated. Column chromatography (silica, Hexanes−EtOAc, 2:1 → 1:1, DCM−MeOH, 100:1 → 20:1) was utilized to purify the **98** acrylic analog as white crystals. An insufficient amount of **98** was obtained and making spectroscopy data unattainable.

3.10. Left-tether Aglycone Modifications



17

106 R = O, n= 1
107 R = O, n =2
107i R = NH_2 = 1

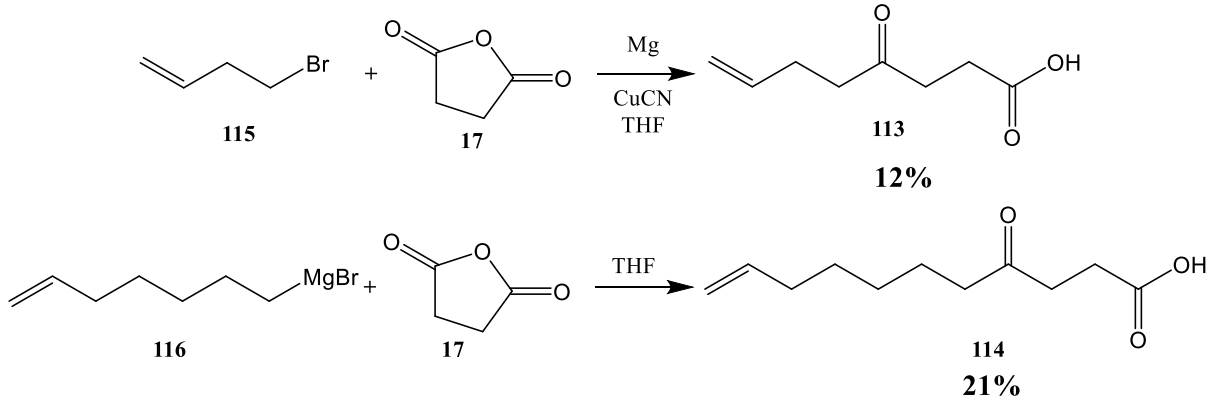
108 R=O, n = 1 81%
109 R= O, n = 2 85%
109i R=NH, n = 1 82%

1-(3-buten-1-yl) ester butanedioic acid **109**. *Method 1:* 1-buten-4-ol **107** (0.92 mL, 10.9 mmol) was added dropwise to a solution of succinic anhydride **17** (1.00 g, 9.99 mmol), and benzene: pyridine (3:1 4 mL). The reaction mixture was heated to reflux (120 ^{o}C) and stirred overnight. At this point, TLC (Hex-EtOAc 3:2, R_f = 0.65) showed the reaction was complete. The reaction was cooled to RT, then diluted with Et_2O (50 mL), washed with 5% HCl solution (25 mL), and a BRINE solution. The organic layer was dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (Hex:EtOAc 5:1→2:1) to give the acid **109** (1.4 g 81%) as a colorless oil. *Method 2.* 1-buten-4-ol **107** (0.424 mL, 4.99 mmol) was added dropwise to a solution of succinic anhydride **17** (0.495 g, 4.99 mmol), DMAP (61 mg, 0.500 mmol), and toluene (7 mL). The reaction mixture was heated to reflux (120 ^{o}C) and stirred overnight. TLC (Hex:EtOAc 3:2, R_f = 0.65) showed the reaction was complete. The reaction was cooled to RT and concentrated under reduced pressure. The residue was purified by column chromatography (Hex:EtOAc 5:1→2:1) to give **108** (0.845 g 85%) as a colorless oil. ^{1}H NMR ($CDCI_3$, 400 MHz) δ_H 10.1 (br s, 1 H COO*H*), 5.83-5.71 (m, 1H, C*H*=), 5.15-5.06 (m, 2H, =C*H*$_2$), 4.16 (t, *J*= 9, 2H, OC*H*$_2$), 2.72-2.60 (m, 2 H), 2.42-2.36 (m, 2H), 2.41 (t, *J*= 20 Hz, 2H), 2.07-1.97 (m, 4H). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 178.5 (O=*C*–O), 172.3, 134.0, 117.5, 64.1,

33.2, 29.1, 29.0. The ^{1}H and ^{13}C NMR of both methods were the same, matching published spectra data.[23]

1-(2-propen-1-yl) ester butanedioic acid **108**. The acid **108** was synthesized in 81% from succinic anhydride **17** (1.00 g, 9.99 mmol) and allyl alcohol (0.66 mL, 15.00 mmol) from Method 2 of the synthesis of **108** ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 11.66 (br, 1H), 6.00−5.81 (m, 1H), 5.39−5.15 (m, 2H), 4.65−4.50 (m, 2H), 2.78−2.59 (m, 4H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 178.6, 172.0, 132.0, 118.5, 65.6, 29.0, 28.9.[26-28]

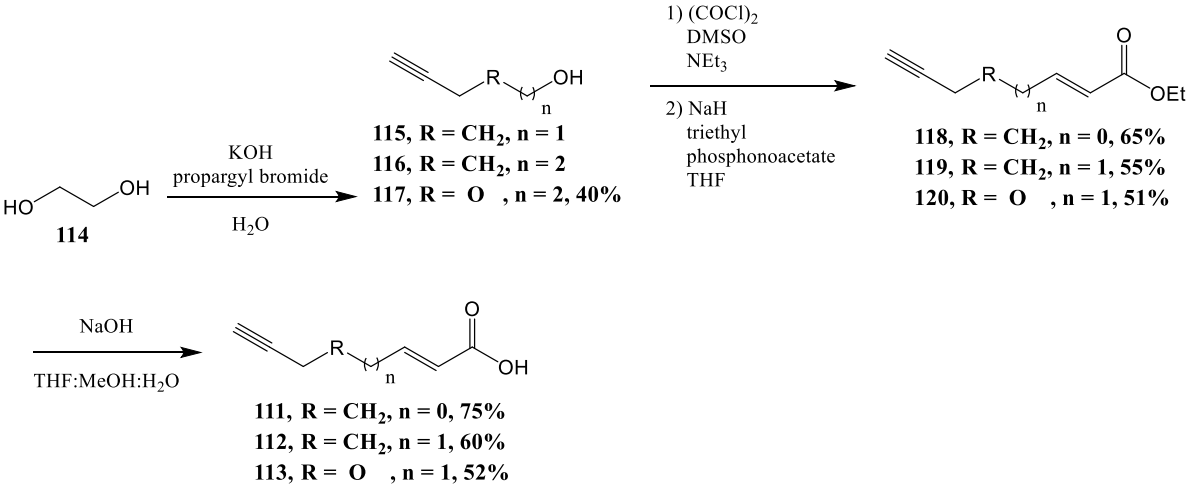
4-oxo-4-(2-propen-1-ylamino)-butanoic acid **107i**. The allyl amine (0.91 mL, 13.01 mmol) was added to a solution of Dis. THF (8 mL) and succinic anhydride **17** (1.01 g, 10.1 mmol) at 0 ^{o}C. The reaction was left to stir at RT overnight (white crystals). The solvent was removed under reduced pressure. The crude acid was recrystallized in Et_2O to give the acid **107i** in 82% (1.31 g). ^{1}H NMR (400 MHz, DMSO-d6) 10.34 (br s, 1H, O=C-O*H*), 6.33-5.98 (br s, 1H, N*H*), 5.79 (m, 1H, CH_2=C*H*), 5.20-5.15 (m, 2H,CH_2=CH), 3.90 (M, 2H, HNCH_2), 2.72 (t, *J* = 6.8 Hz, 2H, CH_2C=O), 2.54 (t, *J* = 6.7 Hz); ^{13}C NMR (100 MHz, DMSO-d6) 174.4 (COOH), 171.5 (CONH), 135.0 ($HC=CH_2$), 115.4 ($HC=CH_2$), 41.6 ($O=CNHCH_2$), 30.6 (H_2C-COOH), 29.7 (H_2C-CONH). Spectral data matched those previously reported.[29, 30]



4-oxo-7-octenoic acid **113**. The acid **113** was prepared from the succinic anhydride **17** (856 mg, 85.5 mmol) and **115** 4-maganism-bromide-1-butene (0.75 mL, 80.1 mmol) by the same method as 4-oxo-8-nonenoic acid **14** in 12% (164 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 10.51 (br s, 1H, COO*H*), 5.80 (ddt, *J* = 16.8, 12.9, 6.5 Hz Hz, 1H, *H*C=CH_2), 5.10-4.90 (m, 2H, HC=CH_2), 2.76-2.67 (m, 2H, CH_2), 2.40-2.28 (m, 2H, CH_2). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 207.9, 179.8, 136.8, 115.3, 41.7, 36.9, 27.7, 27.5. The ^{1}H NMR and ^{13}C NMR were identical to literature published data.[31, 32]

4-oxo-9-undecenoic acid **114**. The longer left-tether acid **114** was obtain through same procedure as 4-oxo-8-nonenoic acid **14** from succinic anhydride **17** (684 mg, 6.83 mmol) and 7-maganism-bromide-1-heptene **116** (0.94 mL, 61.8 mmol) in 21%. ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 209.2, 178.9,139.0,114.6, 42.8,36.9, 33.7, 28.8, 28.8, 28.6, 24.5.

3.11. Potential Modifications to the C-3-Glc*p* Moiety of Ipomoeassin F



Ethyl (2*E*)-hept-2-en-6-ynoate **118**. Oxalyl chloride (1.21 mL, 14.2 mmol) was added to THF (10 mL) and cool to -78 ^{o}C over 30 mins. Then a solution of DMSO (1.08 mL, 15.6 mmol) in THF (10 mL) was added dropwise over 15 mins and allowed to stir at -78 ^{o}C for 30 mins. The

4-pentynol **115** (1.11 mL, 12.0 mmol) in THF (7 mL) was dropwise over 15 mins and stirred at -78 ^{o}C for 45 mins. To the THF solution, NEt_3 (5.0 mL, 36.0 mmol) was added dropwise over 15 mins and stirred for 1 hr. at -78 ^{o}C for 1 hr. As each the reagents were added, white precipitate started to form. After stirring at -78 ^{o}C for 1hr, the reaction mixture slowly warmed to RT over 3-4 hrs. The reaction mixture was filtered through Celite and washed with 15-20 mL of THF. The filtrate was carefully removed under reduced pressure and stored under nitrogen at -5 ^{o}C for 30 mins. While the Swern Ox. was warming to RT, a solution of NaH (358 mg, 15.0 mmol) and THF (10 mL) was cooled to -5 ^{o}C over 15 mins. Then, triethyl phosphonoacetate (3.01 mL, 15.6 mmol) was added dropwise over 15 mins. The mixture turned into a homogenous solution over the next 45 mins while being stirred at 0 ^{o}C. Then, solution was warmed slowly to RT overnight. TLC (Hex−EtOAc 3:1 R_f = 0.75) showed the reaction was complete and the solvent was concentrated to give a yellow oil. Saturated ammonium chloride was added to the oil and extracted with diethyl ether (25 mL ·4). The ether layer was dried with $NaSO_4$, filtered, and the removed under reduced pressure. The yellow oil was purified by flash column chromatography (Hex−EtOAc, 12:1 to 8:1) to give the (2*E*)-ethyl ester **118** in 65% (1.30 g). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 6.95 (dt, *J* = 15.6, 6.3 Hz, 1H), 5.87 (dt, *J* = 15.6, 1.5, 1H), 4.17 (q, *J* = 6.9 Hz, 2H), 2.42 (m, 2H), 2.35 (m, 2H), 1.99 (t, *J* = 2.4 Hz, 1H), 1.27 (t, *J* = 7.2 Hz, 3H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 166.5, 146.5, 122.7, 82.8, 69.6, 60.5, 31.15, 17.15, 14.4. The ^{1}H NMR and ^{13}C NMR were identical to literature published data.[50, 51]

(2*E*)-hept-2-en-6-ynoic acid **111**. A mixture of the THF: MeOH: water (4:1:1) was added to the ethyl ester **118** (101 mg, 0.66 mmol) at RT followed by 0.5 LiOH (3.9 mL, 1.91 mmol). The reaction was stirred at RT overnight. The THF and MeOH was removed under reduced pressure, then acidified by 10% $KHSO_4$ (pH= 3-4). The acidic aqueous layer was extracted with

DCM (3 * 20 mL). The organic layer was dried with $NaSO_4$, filtered and removed under reduced pressure. The crude acid was purified with column chromatography (14:1 to 6:1, Hex: EtOAc) to give **118** (60 mg, 75%). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 10.8 (b, 1H), 7.11 (dt, *J* =15.6, 6.6 Hz, 1H), 5.91 (dt, *J* = 15.6, 1.6 Hz, 1H), 2.47 (m, 2H), 2.47 (m, 2H), 2.38 (m, 2H), 2.01 (t, *J* = 2.4, 1H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 171.9, 149.3, 121.9, 82.4, 69.6, 31.0, 17.2. The spectroscopic data matched previous published data.[52]

Ethyl (2*E*)-octa-2-en-7-ynoate **119**. The same Swern Ox/H-W-E reaction procedure was used to get the ethyl ester **119** as **118** except the starting material was 5-hexynol **115** (1 g) in 55 % (925 mg) over two steps. ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 6.95 (dt, *J* =14. 9, 6.9 Hz, 1H), 5.86 (dt, 1H, *J* = 15.1, 1.5 Hz, 1H), 4.19 (q, *J* = 7.2 H, 2H), 2.34 (q, *J* = 6.9 Hz, 2H), 2.23 (dt, *J* = 6.9 Hz, 2.7 Hz, 2H), 1.98 (t, *J* = 2.7 Hz, 1H), 1.70 (m, 1H) 1.29 (t, *J* = 7.2, 3H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 166.5, 147.8, 122.1, 83.5, 69.0, 60.2, 30.9, 26.7, 17.8, 14.2. The ^{1}H NMR and ^{13}C NMR were identical to literature published data.[50, 53]

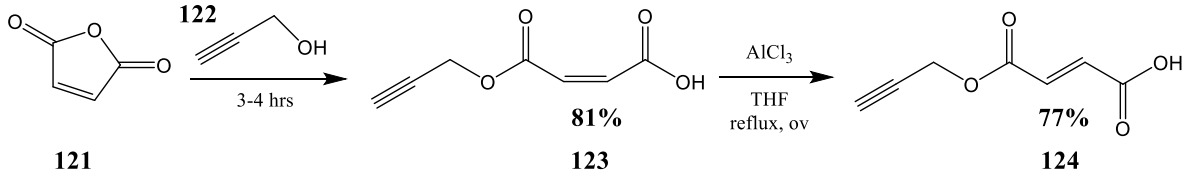
(2*E*)-octa-2-en-7-yonic acid **112**. The same hydrolysis conditions were used to obtain the acid **112** as in 60%. ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 11.28 (b, 1H), 7.07 (dt, *J* = 15.1 Hz, 6.9 Hz,1H), 5.87 (d, 1H), 2.37 (m, 2H), 2.41 (dt, *J* = 8.2 Hz, 2.4 Hz, 2H), 1.99 (t, *J* = 2.7 Hz, 1H), 1.71 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 172.1, 150.8, 121.3, 83.2, 69.1, 30.9, 26.4, 17.2. The NMR data was consistent with published literature data.[54]

2-(Prop-2-yn-1-yloxy)ethanol **116**. A mixture of ethylene glycol (3.49 mL, 62.5 mmol), H_2O (2.5 mL), and KOH (1.40 g, 25.1 mmol) was cooled to 0 ^{O}C. Then propargyl bromide (1.39 mL, 125 mmol) was added dropwise and slowly warmed to RT overnight. H_2O was added to reaction mixture to cause hydrolysis, stirred for 10 mins, and extracted with DCM (30 mL · 3).

The combined organic layers were dried with $NaSO_4$ and removed under reduced pressure. The crude ether was purified with column chromatography to (silica, Hex: EtOAc 3:1 to 1:1, R_f = 0.35) to give **116** in 42 % (2.58 g). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 4.15 (d, *J* = 2.4 Hz, 2H), 3.72-3.69 (m, 2H), 3.61-3.58 (m, 2H), 2.73 (m, 2H), 2.44 (t, *J* = 2.4,1H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 79.4, 74.61, 71.10, 61.35, 58.20. The spectroscopic data matched those previous published.[55, 56]

Ethyl (2*E*)-4-(2′-propynloxy)but-2-enoate **120**. The same procedure was followed as **118** ethyl (2E)-hept-2-en-6-ynoate. The monoethyl ester **120** was obtained in 51% after column chromatography (silica, Hex: EtOAc 35:1 to 28:1). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 6.93 (dt, *J* = 15.9, 4.6 Hz, 1H), 6.08 (dt, *J* = 15.9, 1.8 Hz, 1H), 4.24-4.18 (m, 6H), 2.46 (t, *J* = 2.4 Hz, 1H,), 1.28 (t, *J* = 7.2, 3H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 166.0, 143.1, 121.7, 79.0, 74.9, 68.0, 60.3, 57.8, 14.1. The NMR data was consistent with published literature data.[50]

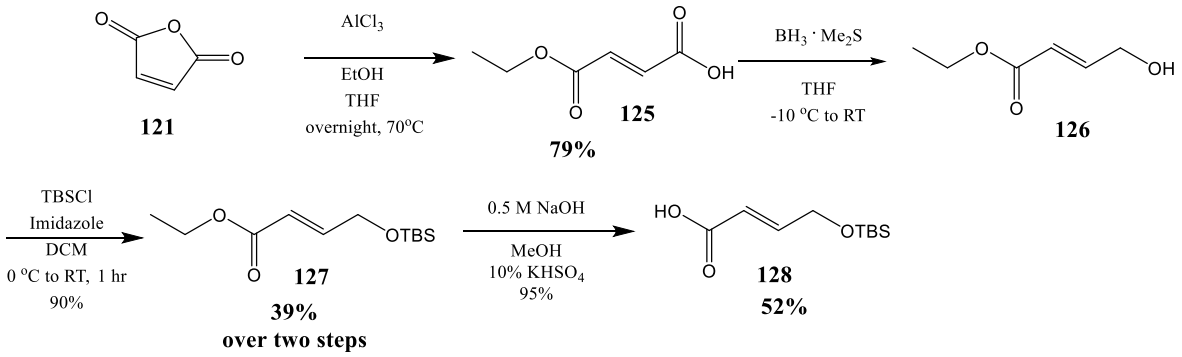
(2*E*)-4-(2′-propynloxy)but-2-enoic acid **113**. The same procedure was followed as the previous acids **111**-**112** in 52%.



Propargyl maleic acid **123**. Maleic anhydride **121** (1.0 g, 10 mmol) was added to propargyl alcohol (10 mL) heated to 60 ^{o}C and stirred overnight. The reaction was cooled to RT and propargyl alcohol was removed under reduced pressure and purified by column chromatography to give **123** in 81% (1.25 g) (silica, Hex:EtOAc 8:1 to 3:1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 9.4 (b,

1H, 1H), 6.41 (d, *J* = 6.15 Hz, 1H), 6.36 (d, *J* = 6.15 Hz, 1H), 4.83 (d, *J* = 2.4 Hz, 1H), 2.56 (t, *J* = 2.4 Hz, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 167.8, 165.2, 131.6, 130.2, 76.3, 75.9, 53.23.

Propargyl fumaric acid **124**. THF (10 mL), propargyl maleic acid **123** (0.95 g, 6.16 mmol), and $AlCl_3$ (200 mg) were in dry THF (20 mL) and heated to reflux (70 ^{o}C). The reaction mixture was stirred at reflux overnight. Sat. $NaHCO_3$ (25 mL) quenched the reaction, then the THF removed under reduced pressure after cooling to RT. The aqueous layer was extracted with DCM (2 · 30 mL), then acidified with conc. HCl (pH= 2-3). The acidic aqueous layer was extracted with DCM (3 · 35 mL). The combined organic layers were dried with $NaSO_4$, filtered and removed under reduced pressure. The acid was purified with column chromatography (silica, Hex:EtOAc 8:1 to 3:1) to give the fumarate 77% (751 mg). ^{1}H NMR (300 MHz, CDCl3) δ_H 10.0 (b, 1H), 6.99 (d, *J* = 15.9 Hz, 1H), 6.91 (d, *J* = 15.9, 1H), 4.83 (d, *J* = 2.4 Hz, 2H), 2.54 (t, *J* = 2.4 Hz, 1H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 169.7, 164.0, 134.6, 134.0, 76.8, 75.8, 53.12.



Monomethyl Fumarate **121**. Maleic Anhydride (2 g, 20.3 mmol), EtOH (1.32 mL, 22.4 mmol), and $AlCl_3$ (200 mg) were dissolved in 10 mL of dis tetrahydrofuran. The reaction mixture was heated to reflux (75 ^{o}C) and stirred overnight. The mixture was cooled to RT and the THF removed under reduced pressure. $NaHCO_3$ (30 mL) was added the oil and extracted with DCM (2 · 40 mL). Next, the aqueous layer was acidified with concentrated HCl until pH 2-3 and extracted

again with DCM (3 · 40 mL). The latter organic layers were combined, dried over Na_2SO_4, and concentrated under reduced pressure to give white crystals **125** (79%, 2.35 g). 1 H NMR (400 MHz, $CDCl_3$) δ_C 10.15 (b, 1H), 6.95 (d, *J* = 15.8 Hz, 1H), 6.85 (d, *J* = 15.8 Hz, 1H), 4.29 (q, *J* = 7.1 Hz, 2H), 1.34 (t, *J* = 7.1 Hz, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ_C 170.1, 164.86, 115.2, 136.1, 132.7, 61.82, 14.30. The NMR data was consistent with published literature data.[57, 58]

(2*E*)-2-Butenoic acid, 4-[(1,1-dimethyl-tert-butyl-silyl)oxy]-, ethyl ester **127**. $BF_3 \cdot Me_2S$ in 1 M THF (7.34 mL, 32.0 mmol) was added dropwise at 0 ^oC to a solution of monoethyl furmate **125** (2.31 g, 16.0 mmol) in THF (25 mL). The solution was allowed gradually warm to room temperature overnight. TLC showed the reaction was complete (4:1, R_f = 0.21). AcOH/H_2O (1:1, v:v, 2.5 mL) was added to the reaction mixture to quench the reaction. The THF was removed under reduced pressure to give a residual slurry. Ice-cold saturated $NaHCO_3$ was added to slurry (20 mL) and stirred for 10 mins. Then the aqueous layer was extracted with EtOAc (3 · 30 mL). The resulting organic layers were combined then washed with saturated $NaHCO_3$ solution, dried with $NaSO_4$. Then, the organic layer was filtered and concentrated under reduced pressure to give (E)-ethyl-4-hydroxy-2-butenoate **126**. After drying, the oil was re-dissolved in Dis. DCM and treated with DMAP (250 mg, 0.70 mmol) and TBSCl (4.86 g, 32.0 mmol). The mixture was cooled to 0^oC and Et_3N (6.05 mL, mmol) was added while the reaction was slowed warmed to RT. TLC (6:1 Hex: EtOAc, R_f = 0.81) showed the reaction was complete. The reaction mixture was washed with H_2O (25 mL) and BRINE (25 mL). The organic layer was dried with $NaSO_4$ and concentrated under reduced pressure. Column chromatography (silica gel, Hex:EtOAc 15:1) was used to give the oil **127** (1.55 g 39% over two steps). ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.00 (dt, *J* =15.3, 3.2 Hz, 1H), 6.10 (dt, *J* =15.3, 2.0 Hz, 1H), 4.35 (dd, *J* = 3.2, 2.0 Hz, 2H), 4.20 (q, *J* = 7.4 Hz, 2H), 1.32 (t, *J* =7.4 Hz, 3H), 0.92 (s, 9H), 0.06 (s, 6H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 166.8,

147.5, 119.8, 62.30, 132.7, 61.82, 14.30. The ^{1}H NMR and ^{13}C NMR were identical to literature published data.[59, 60]

(2*E*)- 4-[[(1,1-dimethyl-tert-butyl)dimethylsilyl]oxy]-2-Butenoic acid **128**. The ethyl ester **128** (115 mg, 4.1 mmol) was dissolved in MeOH (9 mL) and 0.5 M NaOH (aq.) (0.5 mL). The reaction mixture was stirred at RT for 3 hrs and TLC (4:1 Hex:EtOAc R_f = 0.4) showed the reaction was complete. Then, the MeOH was evaporated under reduced pressure. The residue was cooled to 0^{o}C, then acidified with 10% $KHSO_4$ (aq). The aqueous layer was extracted with diethyl ether (3X 25 mL). The organic layers were combined, dried with $NaSO_4$, and concentrated under reduced pressure to give **128** in 52%. ^{1}H NMR (300 MHz, $CDCl_3$) δ_H 7.14 (dt, *J* = 15.3, 3.2 Hz, 1H), 6.14 (dt, *J* =15.3, 2.0 Hz, 1H), 4.39 (m, 2H), 0.93 (s, 9H), 0.1 (s, 6H). ^{13}C NMR (75 MHz, $CDCl_3$) δ_C 171.9, 150.3, 119.0, 62.2, 26.0, 18.4, -5.4. The NMR data was consistent with published literature data.[59, 61]

3.12. Biology and Cytotoxicity Assays

3.12.1. Cell Culture

Two breast cancer cell lines MCF7 and MDA-MB-231 were maintained in a DMEM/HIGH culture medium supplemented with 10% bovine calf serum (BCS) and 2 mM L-glutamine, so-called complete medium. Cell cultures were grown in monolayers in a humidified atmosphere of 5% CO_2 and 95% air at 37 °C. The culture medium was changed every 48−72 hrs. Cell cultures were passaged once or twice a week using trypsin-EDTA (0.25%) to detach the cells from their culture flasks/dishes.

3.12.2. MTT Cytotoxicity Assay

The viable cells were counted before each experiment was done. The assays were performed in triplicate. First, 100 μL of cell suspension at the density of 25,000 cells/mL was seeded in a 96-well plate (2,500 cells/well), which was incubated at 37 °C in 5% CO2 for 24 hrs. Ipomoeassin F, analog **86**, and **87** were dissolved in DMSO (dimethyl sulfoxide) to make drug stocks (10 mM). The stock solutions were diluted with the complete DMEM/HIGH medium to make a series of gradient fresh working solutions right before each test. Then, the cells were treated with 100 μL of the freshly made gradient working solution in the total volume of 200 μL/well for 72 hrs. After that, the media were discarded and 200 μL of the fresh complete medium containing 10% of MTT stock solution (5 mg/mL) was added to each well. The plate was then incubated at 37 °C in 5% CO_2 atmosphere for another 3 hrs. Next, 180 μL of the medium was discarded from each well. The formed formazan crystals were dissolved with 180 μL of DMSO. Absorbance of formazan was detected by a microplate reader (BioTek Synergy H1) at 570 nm with 650 nm as the reference wavelength. The percentage of viability compared to the negative control (DMSO-treated cells) was determined. Activities of the synthesized ipomoeassin F, analog **86**, and **87** were tested against breast cancer cell line MCF7 by the MTT cytotoxicity assay. GraphPad Prism 6 software was used to make a plot of % viability versus sample concentration and to calculate the concentration at which a compound exhibited 50% cytotoxicity (IC_{50}).

3.12.3. AlamarBlue Cytotoxicity Assay

The viable cells were counted before each experiment was done. The assays were performed in triplicates. First, 100 μL of cell suspension at the density of 25,000 cells/mL was seeded in a 96-well plate (2,500 cells/well), which was incubated at 37 °C in 5% CO_2 for 24 hrs. Ipomoeassin F, analogs **86** and **87** were dissolved in DMSO (dimethyl sulfoxide) to make drug

stocks (10 mM). The stock solutions were diluted with the complete DMEM/ HIGH medium to make a series of gradient fresh working solutions right before each test. Subsequently, the cells were treated with 100 μL of the freshly made gradient working solution in the total volume of 200 μL/well for 72 hrs. After that, the media were discarded and 200 μL of the fresh complete medium containing 10% of AlamarBlue (resazurin) stock solution (3 mg/27.15 mL) was added to each well. The plate was then incubated at 37 °C in 5% CO2 atmosphere for another 3 hrs. Next, 180 μL of the medium was discarded from each well. The formed formazan crystals were dissolved with 180 μL of DMSO. Absorbance of formazan was detected by a microplate reader (BioTek Synergy H1) at excitation 580 nm with emission 620 nm as the reference wavelength. The percentage of viability compared to the negative control (DMSO-treated cells) was determined. Activities of synthesized Ipomoeassin F **1**, analogs **86**, and **87** were tested against breast cancer cell line MDA-MB-231 by the AlamarBlue cytotoxicity assay. GraphPad Prism 6 software was used to make a plot of % viability versus sample concentration and to calculate the concentration at which a compound exhibited 50% cytotoxicity (IC_{50}).

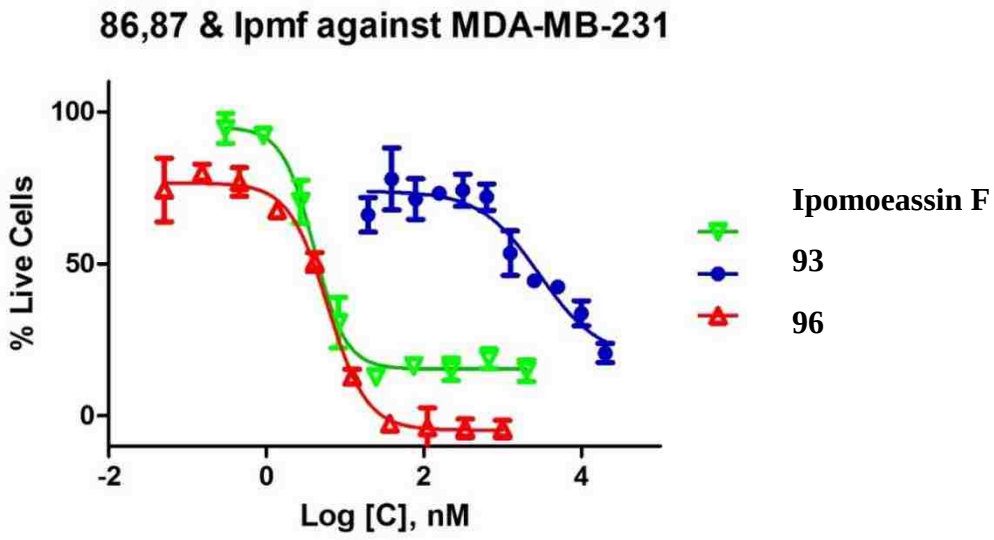


Figure 3.1. IC_{50} curves of Ipomoeassin F and analogs 86-87 against the MDA-MB-231 cell line.

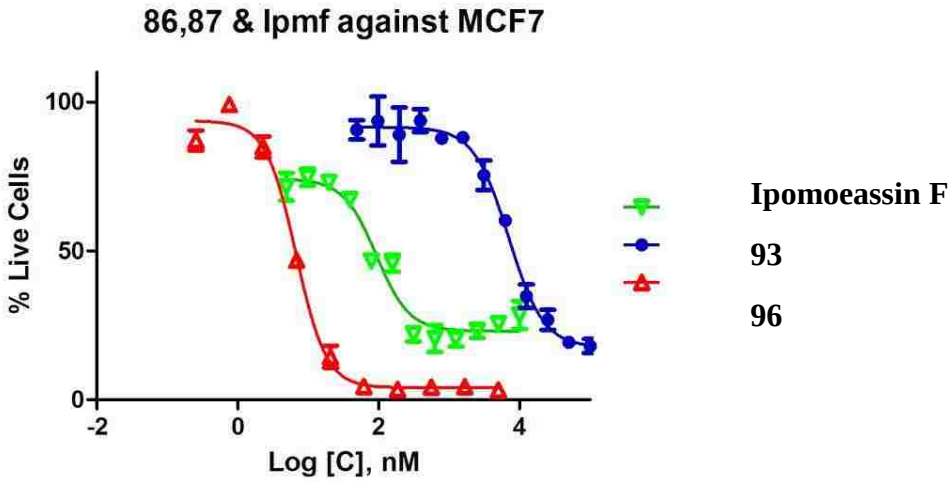


Figure 3.2. IC_{50} curves of Ipomoeassin F and analogs 86-87 against the MCF-7 cell line.

3.13. References

1. Chen, C. T.; Kuo, J. H.; Pawar, V. D.; Munot, Y. S.; Weng, S. S.; Ku, C. H.; Liu, C. Y., Nucleophilic acyl substitutions of anhydrides with protic nucleophiles catalyzed by amphoteric, oxomolybdenum species. *Journal of Organic Chemistry* **2005,** *70* (4), 1188-1197.

2. Fenger, T. H.; Marinescu, L. G.; Bols, M., Cyclodextrin Ketones with the Catalytic Group at the Secondary Rim and Their Effectiveness in Enzyme-Like Epoxidation of Stilbenes. *European Journal of Organic Chemistry* **2011,** (12), 2339-2345.

3. Ikeda, K.; Morimoto, T.; Kakiuchi, K., Utilization of Aldoses as a Carbonyl Source in Cyclocarbonylation of Enynes. *Journal of Organic Chemistry* **2010,** *75* (18), 6279-6282.

4. Pilgrim, W.; Murphy, P. V., SnCl4- and TiCl4-Catalyzed Anomerization of Acylated O- and S-Glycosides: Analysis of Factors That Lead to Higher alpha:beta Anomer Ratios and Reaction Rates. *Journal of Organic Chemistry* **2010,** *75* (20), 6747-6755.

5. Rye, C. S.; Withers, S. G., Elucidation of the mechanism of polysaccharide cleavage by chondroitin AC lyase from Flavobacterium heparinum. *Journal of the American Chemical Society* **2002,** *124* (33), 9756-9767.

6. Kong, L. B.; Almond, A.; Bayley, H.; Davis, B. G., Chemical polyglycosylation and nanolitre detection enables single-molecule recapitulation of bacterial sugar export. *Nature Chemistry* **2016,** *8* (5), 461-469.

7. Zheng, S. J.; Laraia, L.; Connor, C. J. O.; Sorrell, D.; Tan, Y. S.; Xu, Z. C.; Venkitaraman, A. R.; Wu, W. J.; Spring, D. R., Synthesis and biological profiling of

tellimagrandin I and analogues reveals that the medium ring can significantly modulate biological activity. *Organic & Biomolecular Chemistry* **2012,** *10* (13), 2590-2593.

8. Marton, Z.; Tran, V.; Tellier, C.; Dion, M.; Drone, J.; Rabiller, C., Engineering of glucoside acceptors for the regioselective synthesis of beta-(1 -> 3)-disaccharides with glycosynthases. *Carbohydrate Research* **2008,** *343* (17), 2939-2946.

9. Grindley, T. B.; Wickramage, C., A NOVEL-APPROACH TO THE SYNTHESIS OF C-GLYCOSYL COMPOUNDS - THE WITTIG REARRANGEMENT. *Journal of Carbohydrate Chemistry* **1988,** *7* (3), 661-685.

10. Fugedi, P., SYNTHESIS OF 4-O-(ALPHA-L-RHAMNOPYRANOSYL)-D-GLUCOPYRANURONIC ACID. *Journal of Carbohydrate Chemistry* **1987,** *6* (3), 377-398.

11. Tian, F.; Migaud, M. E.; Frost, J. W., myo-inositol 1-phosphate synthase: Does a single active-site amino acid catalyze multiple proton transfers? *Journal of the American Chemical Society* **1999,** *121* (24), 5795-5796.

12. Sokolov, V. M.; Zakharov, V. I.; Studentsov, E. P., Stereoselectivity of reactions at the glycoside center of carbohydrates: VII. Synthesis of aryl alpha- and beta-D-glucopyranosides by Helferich, catalyzed by boron trifluoride etherate. *Russian Journal of General Chemistry* **2002,** *72* (5), 806-811.

13. Bruneau, A.; Roche, M.; Hamze, A.; Brion, J. D.; Alami, M.; Messaoudi, S., Stereoretentive Palladium-Catalyzed Arylation, Alkenylation, and Alkynylation of 1-Thiosugars and Thiols Using Aminobiphenyl Palladacycle Precatalyst at Room Temperature. *Chemistry-a European Journal* **2015,** *21* (23), 8375-8379.

14. Veleti, S. K.; Lindenberger, J. J.; Thanna, S.; Ronning, D. R.; Sucheck, S. J., Synthesis of a Poly-hydroxypyrolidine-Based inhibitor of Mycobacterium tuberculosis GlgE. *Journal of Organic Chemistry* **2014,** *79* (20), 9444-9450.

15. Cai, C.; Dickinson, D. M.; Li, L. Y.; Masuko, S.; Suflita, M.; Schultz, V.; Nelson, S. D.; Bhaskar, U.; Liu, J.; Linhardt, R. J., Fluorous-Assisted Chemoenzymatic Synthesis of Heparan Sulfate Oligosaccharides. *Organic Letters* **2014,** *16* (8), 2240-2243.

16. Zhang, X. R.; Zhang, C. Q., Nano n-Propylsulfonated Magnetic -Fe2O3 as an Efficient and Reusable Catalyst for the Synthesis O-Isopropylidene Derivatives of Carbohydrates. *Journal of Carbohydrate Chemistry* **2013,** *32* (4), 249-258.

17. Moriarty, R. M.; Rani, N.; Enache, L. A.; Rao, M. S.; Batra, H.; Guo, L.; Penmasta, R. A.; Staszewski, J. P.; Tuladhar, S. M.; Prakash, O.; Crich, D.; Hirtopeanu, A.; Gilardi, R., The intramolecular asymmetric Pauson-Khand cyclization as a novel and general stereoselective route to benzindene prostacyclins: Synthesis of UT-15 (treprostinil). *Journal of Organic Chemistry* **2004,** *69* (6), 1890-1902.

18. Kang, B.; Mowat, J.; Pinter, T.; Britton, R., Development of a Concise and General Enantioselective Approach to 2,5-Disubstituted-3-hydroxytetrahydrofurans. *Organic Letters* **2009,** *11* (8), 1717-1720.

19. Toda, H.; Imae, R.; Itoh, N., Bioproduction of Chiral Epoxyalkanes using Styrene Monooxygenase from Rhodococcus sp ST-10 (RhSMO). *Advanced Synthesis & Catalysis* **2014,** *356* (16), 3443-3450.

20. Wadsworth, A. D.; Furkert, D. P.; Brimble, M. A., Total Synthesis of the Macrocyclic N-Methyl Enamides Palmyrolide A and 2S-Sanctolide A. *Journal of Organic Chemistry* **2014,** *79* (22), 11179-11193.

21. Huang, X. R.; Pan, X. H.; Lee, G. H.; Chen, C. P., C-2-Symmetrical Bipyridyldiols as Promising Enantioselective Catalysts in Nozaki-Hiyama Allylation. *Advanced Synthesis & Catalysis* **2011,** *353* (11-12), 1949-1954.

22. Feng, J. P.; Shi, Z. F.; Li, Y.; Zhang, J. T.; Qi, X. L.; Chen, J.; Cao, X. P., An improved asymmetric synthesis of malyngamide U and its 2 '-epimer. *Journal of Organic Chemistry* **2008,** *73* (17), 6873-6876.

23. Padwa, A.; Hornbuckle, S. F.; Fryxell, G. E.; Stull, P. D., REACTIVITY PATTERNS IN THE RHODIUM CARBENOID INDUCED TANDEM CYCLIZATION CYCLO-ADDITION REACTION. *Journal of Organic Chemistry* **1989,** *54* (4), 817-824.

24. Killen, J. C.; Leonard, J.; Aggarwal, V. K., A Novel Asymmetric Azaspirocyclisation Using a Morita-Baylis-Hillman-Type Reaction. *Synlett* **2010,** (4), 579-582.

25. Hodgson, D. M.; Stupple, P. A.; Pierard, F.; Labande, A. H.; Johnstone, C., Development of dirhodium(II)-catalyzed generation and enantioselective 1,3-dipolar cycloaddition of carbonyl ylides. *Chemistry-a European Journal* **2001,** *7* (20), 4465-4476.

26. Catry, M. A.; Madder, A., Synthesis of functionalised nucleosides for incorporation into nucleic acid-based serine protease mimics. *Molecules* **2007,** *12* (1), 114-129.

27. Cheng, Y. A.; Chen, T.; Tan, C. K.; Heng, J. J.; Yeung, Y. Y., Efficient Medium Ring Size Bromolactonization Using a Sulfur-Based Zwitterionic Organocatalyst. *Journal of the American Chemical Society* **2012,** *134* (40), 16492-16495.

28. Casimir, J. R.; Turetta, C.; Ettouati, L.; Paris, J., FIRST APPLICATION OF THE DAKIN-WEST REACTION TO FMOC CHEMISTRY - SYNTHESIS OF THE KETOMETHYLENE TRIPEPTIDE FMOC-N-ALPHA-ASP(TBU)-(R,S)TYR(TBU)PSI(CO-CH2)GLY-OH. *Tetrahedron Letters* **1995,** *36* (27), 4797-4800.

29. Ozay, O., Synthesis and Characterization of Novel pH-Responsive Poly(2-hydroxylethyl methacrylate-co-N-allylsuccinamic acid) Hydrogels for Drug Delivery. *Journal of Applied Polymer Science* **2014,** *131* (1).

30. Gulten, S., The Synthesis and Characterization of Solvatochromic Maleimide-Fused N-Allyl- and N-Alkyl-Substituted 1,4-Dithiines and Diels-Alder Reactions with Anthracene. *Journal of Heterocyclic Chemistry* **2010,** *47* (1), 188-193.

31. Shuler, S. A.; Yin, G. Y.; Krause, S. B.; Vesper, C. M.; Watson, D. A., Synthesis of Secondary Unsaturated Lactams via an Aza-Heck Reaction. *Journal of the American Chemical Society* **2016,** *138* (42), 13830-13833.

32. Chatupheeraphat, A.; Soorukram, D.; Kuhakarn, C.; Tuchinda, P.; Reutrakul, V.; Pakawatchai, C.; Saithong, S.; Pohmakotr, M., Synthesis of gem-Difluoromethylenated Spiro-gamma-butyrolactones by Employing PhSCF2Si(CH3)(3) as a gem-Difluoromethylenating Agent. *European Journal of Organic Chemistry* **2013,** *2013* (30), 6844-6858.

33. Ghosal, P.; Shaw, A. K., A Chiron Approach to Aminocytitols by Petasis-Borono-Mannich Reaction: Formal Synthesis of (+)-Conduramine E and (-)-Conduramine E. *Journal of Organic Chemistry* **2012,** *77* (17), 7627-7632.

34. Gille, A.; Hiersemann, M., (-)-Lytophilippine A: Synthesis of a C1-C18 Building Block. *Organic Letters* **2010,** *12* (22), 5258-5261.

35. Zeng, J.; Sun, G. F.; Yao, W.; Zhu, Y. B.; Wang, R. B.; Cai, L.; Liu, K.; Zhang, Q.; Liu, X. W.; Wan, Q., 3-Aminodeoxypyranoses in Glycosylation: Diversity-Oriented Synthesis and Assembly in Oligosaccharides. *Angewandte Chemie-International Edition* **2017,** *56* (19), 5227-5231.

36. Park, H. S.; Lee, H. Y.; Kim, Y. H., Facile Barton-McCombie deoxygenation of alcohols with tetrabutylammonium peroxydisulfate and formate ion. *Organic Letters* **2005,** *7* (15), 3187-3190.

37. Lowary, T. L.; Hindsgaul, O., RECOGNITION OF SYNTHETIC DEOXY AND DEOXYFLUORO ANALOGS OF THE ACCEPTOR ALPHA-L-FUCP-(1- 2)-BETA-D-GALP-OR BY THE BLOOD-GROUP-A AND BLOOD-GROUP-B GENE-SPECIFIED GLYCOSYLTRANSFERASES. *Carbohydrate Research* **1993,** *249* (1), 163-195.

38. Wei, J. H.; Lv, X.; Lu, Y.; Yang, G. Z.; Fu, L. F.; Yang, L.; Wang, J. J.; Gao, J. H.; Cheng, S. H.; Duan, Q.; Jin, C.; Li, X. B., Glycosynthase with Broad Substrate Specificity - an Efficient Biocatalyst for the Construction of Oligosaccharide Library. *European Journal of Organic Chemistry* **2013,** (12), 2414-2419.

39. Wang, Q.; Grkovic, T.; Font, J.; Bonham, S.; Pouwer, R. H.; Bailey, C. G.; Moran, A. M.; Ryan, R. M.; Rasko, J. E. J.; Jormakka, M.; Quinn, R. J.; Holst, J., Monoterpene Glycoside ESK246 from Pittosporum Targets LAT3 Amino Acid Transport and Prostate Cancer Cell Growth. *Acs Chemical Biology* **2014,** *9* (6), 1369-1376.

40. Cheng, J. M. H.; Dangerfield, E. M.; Timmer, M. S. M.; Stocker, B. L., A divergent approach to the synthesis of iGb3 sugar and lipid analogues via a lactosyl 2-azido-sphingosine intermediate. *Organic & Biomolecular Chemistry* **2014,** *12* (17), 2729-2736.

41. Bording, S.; Bach, T., An enantioselective synthesis of the C-24-C-40 fragment of (-)-pulvomycin. *Chemical Communications* **2014,** *50* (38), 4901-4903.

42. Faust, T.; Theurer, C.; Eger, K.; Kreis, W., SYNTHESIS OF URIDINE 5'-(ALPHA-D-FUCOPYRANOSYL DIPHOSPHATE) AND (DIGITOXIGENIN-3-BETA-YL)-BETA-D-FUCOPYRANOSIDE AND ENZYMATIC BETA-D-FUCOSYLATION OF CARDENOLIDE AGLYCONES IN DIGITALIS-LANATA. *Bioorganic Chemistry* **1994,** *22* (2), 140-149.

43. Milhomme, O.; John, C.; Djedaini-Pilard, F.; Grandjean, C., Access to Antigens Related to Anthrose Using Pivotal Cyclic Sulfite/Sulfate Intermediates. *Journal of Organic Chemistry* **2011,** *76* (15), 5985-5998.

44. Zhang, Y. Y.; Dong, D. X.; Zhou, T.; Zhang, Y. M., Study of carbohydrate-carbohydrate interactions: total synthesis of 6d-deoxy Lewis(x) pentaosyl glycosphingolipid. *Tetrahedron* **2010,** *66* (37), 7373-7383.

45. Postema, M. H. D.; TenDyke, K.; Cutter, J.; Kuznetsov, G.; Xu, O. L., Total Synthesis of Ipomoeassin F. *Organic Letters* **2009,** *11* (6), 1417-1420.

46. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Muller, R.; Furstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chemistry-a European Journal* **2009,** *15* (38), 9697-9706.

47. Nagano, T.; Pospisil, J.; Chollet, G.; Schulthoff, S.; Hickmann, V.; Moulin, E.; Herrmann, J.; Mueller, R.; Fuerstner, A., Total Synthesis and Biological Evaluation of the Cytotoxic Resin Glycosides Ipomoeassin A-F and Analogues. *Chem. - Eur. J.* **2009,** *15* (38), 9697-9706, S9697/1-S9697/48.

48. Zong, G. H.; Barber, E.; Aljewari, H.; Zhou, J. H.; Hu, Z. J.; Du, Y. C.; Shi, W. Q., Total Synthesis and Biological Evaluation of Ipomoeassin F and Its Unnatural 11R-Epimer. *Journal of Organic Chemistry* **2015,** *80* (18), 9279-9291.

49. Zong, G. H.; Aljewari, H.; Hu, Z. J.; Shi, W. Q., Revealing the Pharmacophore of Ipomoeassin F through Molecular Editing. *Organic Letters* **2016,** *18* (7), 1674-1677.

50. Grafton, M. W.; Farrugia, L. J.; Sutherland, A., Synthesis of Amino-Substituted Indanes and Tetralins via Consecutive Multibond-Forming Tandem Processes. *Journal of Organic Chemistry* **2013,** *78* (14), 7199-7207.

51. Batsomboon, P.; Dudley, G. B., Synthesis of C1-C15 of palmerolide A: tactical advances that can lead to better design strategies for polyketide synthesis. *Tetrahedron Letters* **2016,** *57* (33), 3757-3759.

52. Komiya, M.; Asano, S.; Koike, N.; Koga, E.; Igarashi, J.; Nakatani, S.; Isobe, Y., Synthesis of Novel Benzo-Fused Heteroaryl Derivatives as Ca2+/Calmodulin-Dependent Protein Kinase II Inhibitors. *Chemical & Pharmaceutical Bulletin* **2013,** *61* (10), 1094-1097.

53. Majmudar, J. D.; Konopko, A. M.; Labby, K. J.; Tom, C.; Crellin, J. E.; Prakash, A.; Martin, B. R., Harnessing Redox Cross-Reactivity To Profile Distinct Cysteine Modifications. *Journal of the American Chemical Society* **2016,** *138* (6), 1852-1859.

54. Ikeuchi, Y.; Taguchi, T.; Hanzawa, Y., Copper-catalyzed acylations of o-vinylbenzylzirconocene intermediate and synthetic application toward steroid skeleton. *Tetrahedron Letters* **2004,** *45* (23), 4495-4498.

55. Cserep, G. B.; Baranyai, Z.; Komaromy, D.; Horvati, K.; Bosze, S.; Kele, P., Fluorogenic tagging of peptides via Cys residues using thiol-specific vinyl sulfone affinity tags. *Tetrahedron* **2014,** *70* (35), 5961-5965.

56. Bucher, J.; Wurm, T.; Nalivela, K. S.; Rudolph, M.; Rominger, F.; Hashmi, A. S. K., Cyclization of Gold Acetylides: Synthesis of Vinyl Sulfonates via Gold Vinylidene Complexes. *Angewandte Chemie-International Edition* **2014,** *53* (15), 3854-3858.

57. Ma, Y. L.; Zhou, R. J.; Zeng, X. Y.; An, Y. X.; Qiu, S. S.; Nie, L. J., Synthesis, DFT and antimicrobial activity assays in vitro for novel cis/trans-but-2-enedioic acid esters. *Journal of Molecular Structure* **2014,** *1063*, 226-234.

58. Wang, G. J.; Tang, Y.; Zhang, Y.; Liu, X. H.; Lin, L. L.; Feng, X. M., Enantioselective Synthesis of N-H-Free 1,5-Benzothiazepines. *Chemistry-a European Journal* **2017,** *23* (3), 554-557.

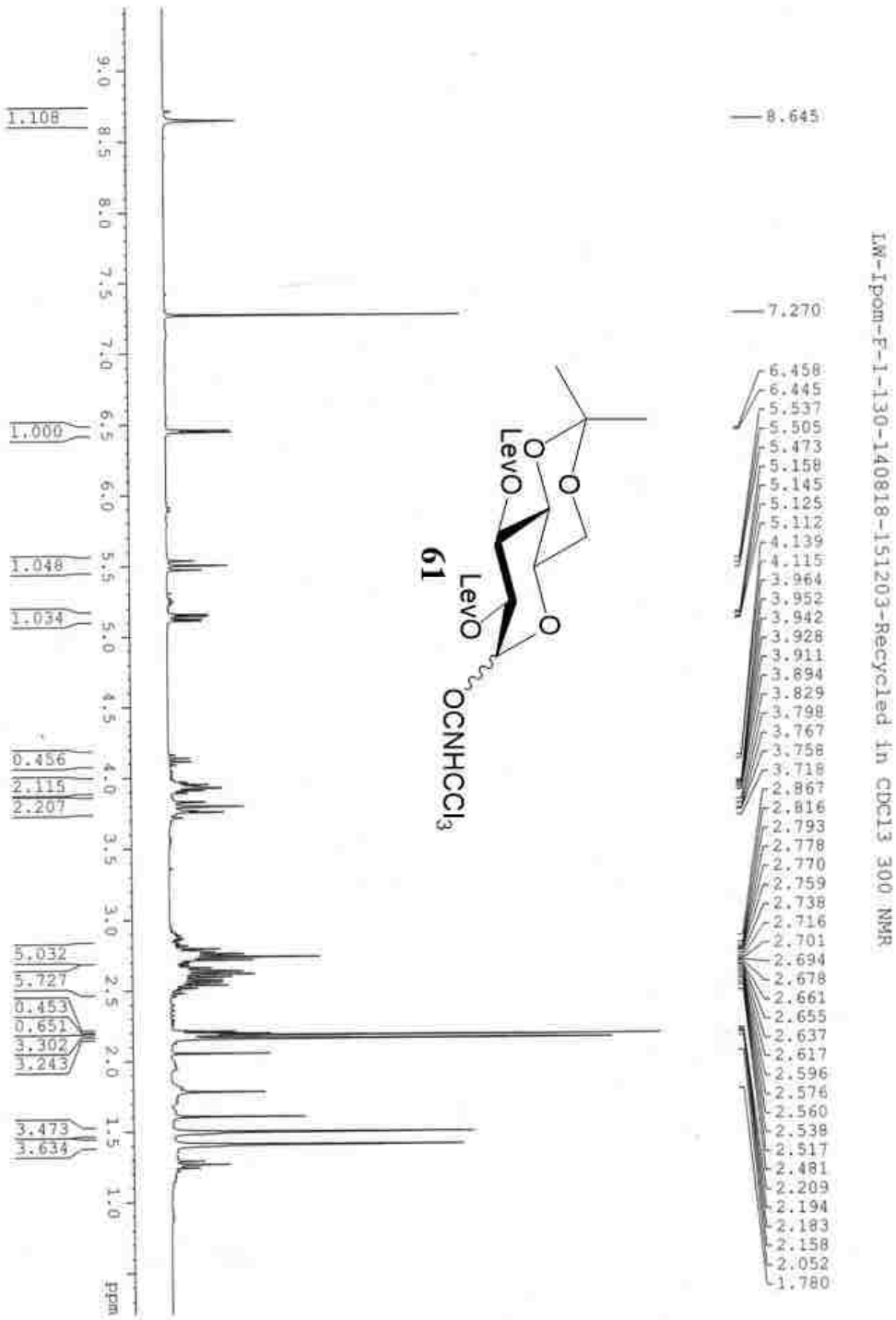
59. Jimenez, J.; Ramirez, J.; Huelgas, G.; Melendrez, R.; Cabrera-Vivas, B. M.; Sansinenea, E.; Ortiz, A., 'Syn-effect' in the diastereoselective alkylation of 3- (E)-alpha,beta-unsaturated-gamma-substituted -N-acyloxazolidinones. *Tetrahedron* **2015,** *71* (28), 4590-4597.

60. Henderson, J. R.; Parvez, M.; Keay, B. A., A Concise Diels-Alder Strategy for the Asymmetric Synthesis of (+)-Albicanol, (+)-Albicanyl Acetate, (+)-Dihydrodrimenin, and (-)-Dihydroisodrimeninol. *Organic Letters* **2009,** *11* (15), 3178-3181.

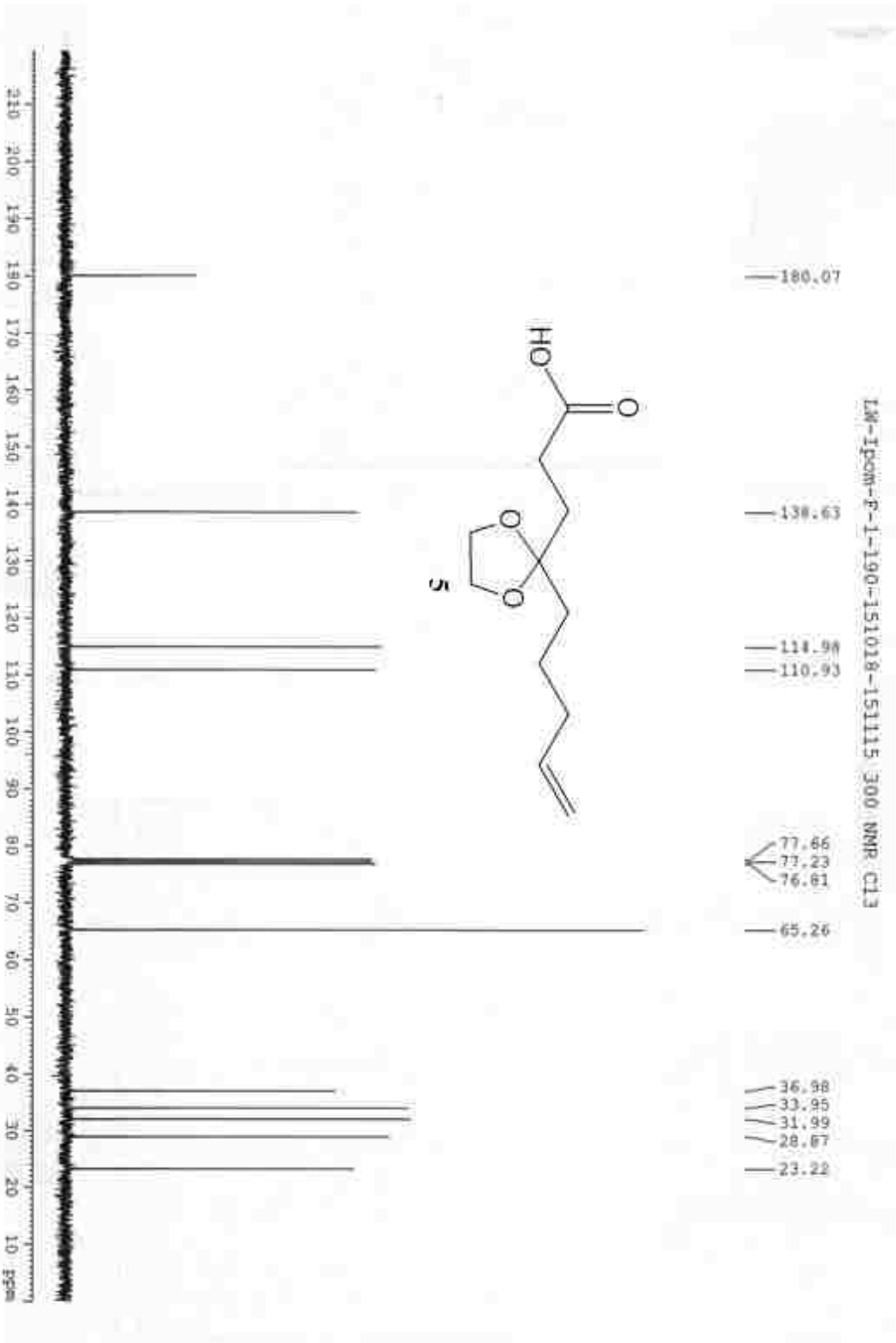
61. Tilley, S. D.; Reber, K. P.; Sorensen, E. J., A Rapid, Asymmetric Synthesis of the Decahydrofluorene Core of the Hirsutellones. *Organic Letters* **2009,** *11* (3), 701-703.

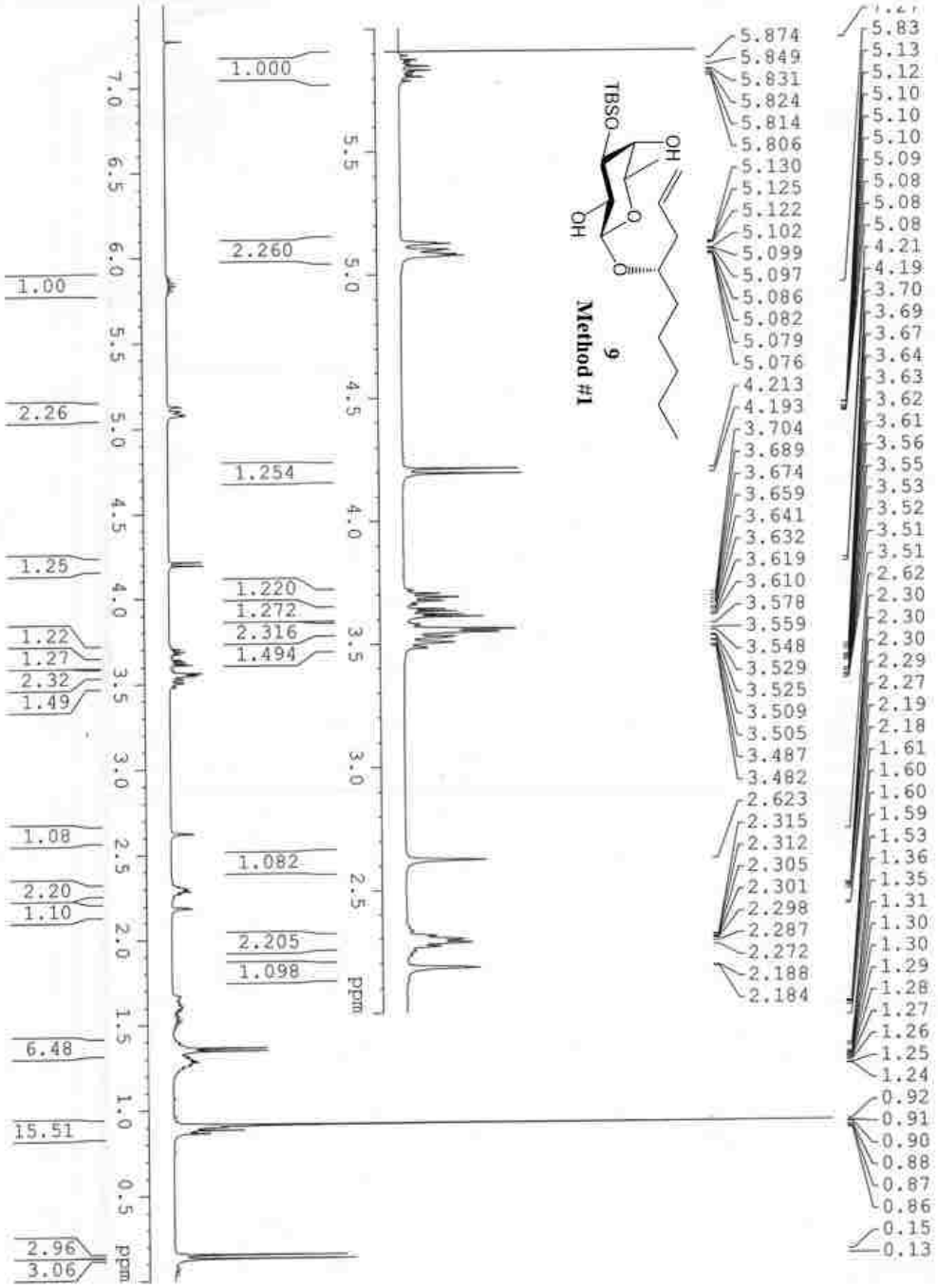
CHAPTER 4. APPENDIX

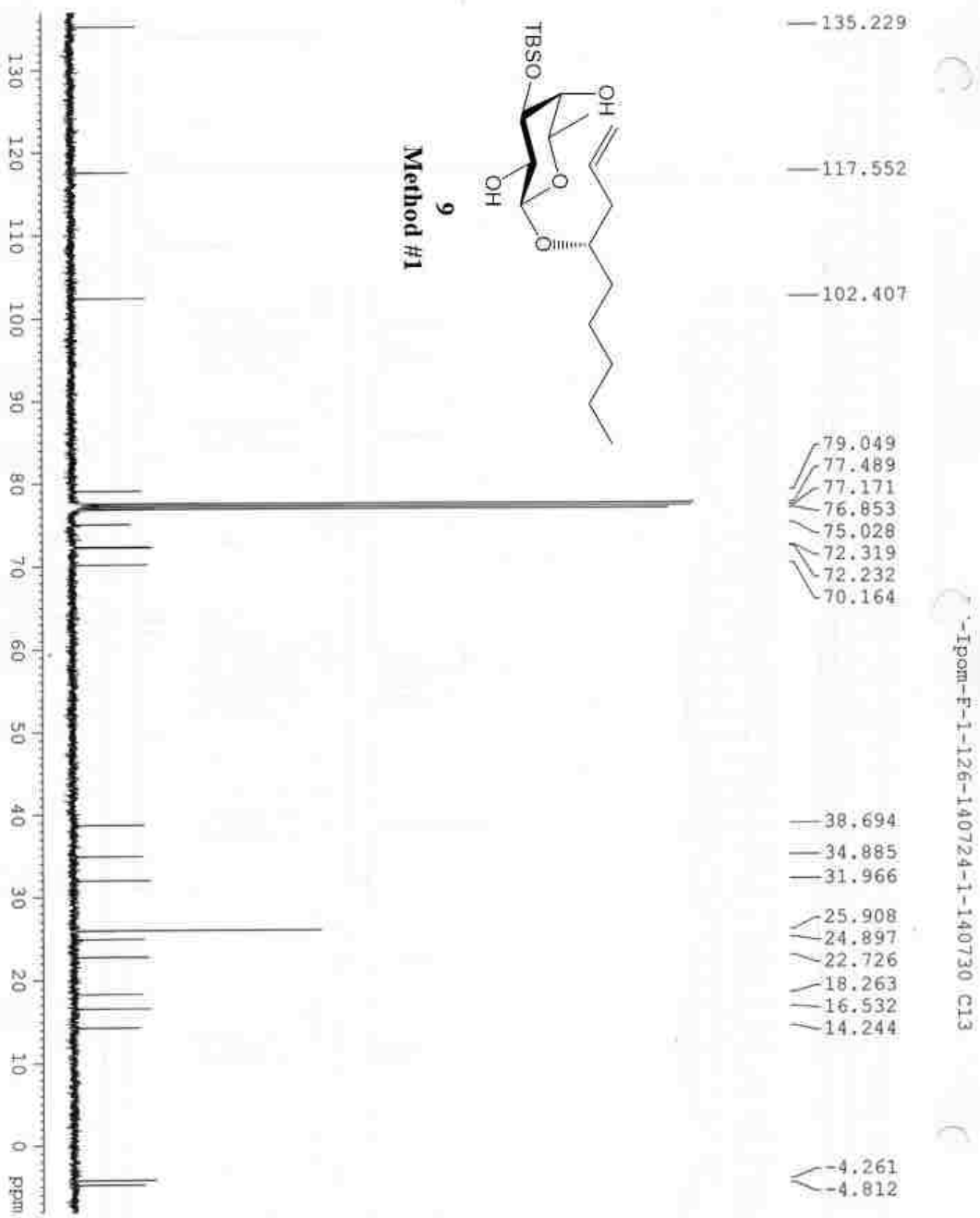
Table of Contents

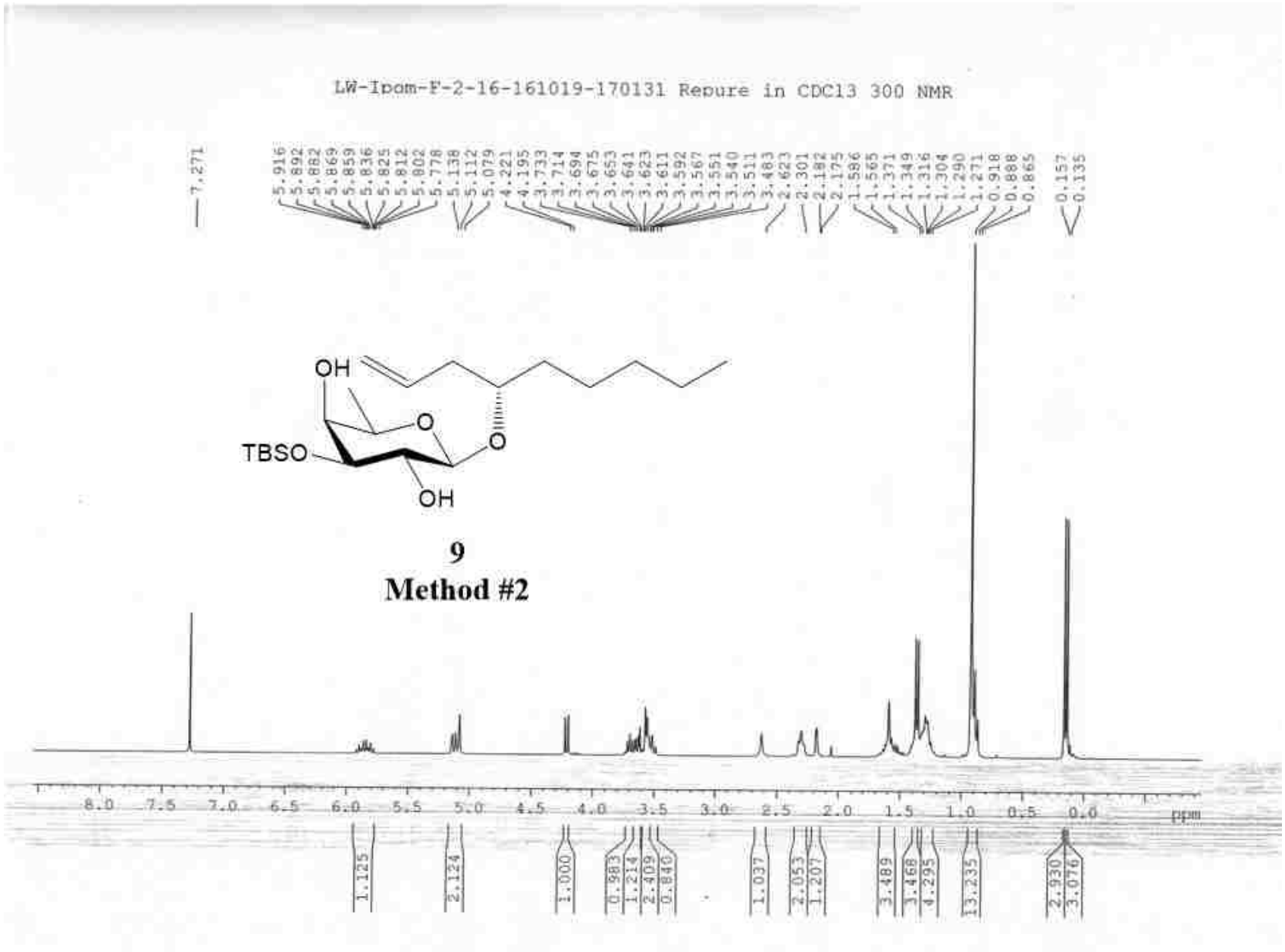

LW-Ipom-F-1-130-140818-151203-Recycled in CDCl3 300 NMR
8.645
7.270
6.458
6.445
5.537
5.505
5.473
5.158
5.145
5.125
5.112
4.139
4.115
3.964
3.952
3.942
3.928
3.911
3.894
3.829
3.798
3.767
3.756
3.718
2.867
2.816
2.793
2.778
2.770
2.759
2.738
2.716
2.701
2.694
2.678
2.661
2.655
2.637
2.617
2.596
2.576
2.560
2.538
2.517
2.481
2.209
2.194
2.183
2.158
2.052
1.780
LevO
LevO
OCNHCCl3
61
1.108
1.000
1.048
1.034
0.456
2.115
2.207
5.032
5.727
0.453
0.651
3.302
3.243
3.473
3.634
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm

IM-1Pam-P-1-10-13021 1H CDCl3 300 NMR
7.270
5.843
5.828
5.821
5.809
5.794
5.787
5.771
5.764
5.752
5.741
5.737
5.730
5.715
5.708
5.032
5.027
5.021
4.969
4.965
4.934
4.931
3.944
3.937
2.436
2.427
2.411
2.409
2.403
2.392
2.386
2.377
2.079
2.074
2.057
2.031
2.023
2.004
2.000
1.957
1.979
1.973
OH
O
O
O
S
0.745
1.000
2.020
4.019
2.018
4.085
2.027
4.019
ppm

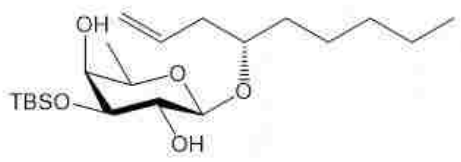

LM-Ipsm-F-1-190-151018-151115 300 NMR C13
180.07
138.63
118.98
110.93
77.66
77.23
76.61
65.26
36.98
33.95
31.99
28.87
23.22
OH
O
O
O
5
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm


TBSO
OH
OH
O
O
9
Method #1
5.874
5.849
5.831
5.824
5.814
5.806
5.130
5.125
5.122
5.102
5.099
5.097
5.086
5.082
5.079
5.076
4.213
4.193
3.704
3.689
3.674
3.659
3.641
3.632
3.619
3.610
3.578
3.559
3.548
3.529
3.525
3.509
3.505
3.487
3.482
2.623
2.315
2.312
2.305
2.301
2.298
2.287
2.272
2.188
2.184
5.83
5.13
5.12
5.10
5.10
5.10
5.09
5.08
5.08
5.08
4.21
4.19
3.70
3.69
3.67
3.64
3.63
3.62
3.61
3.56
3.55
3.53
3.52
3.51
3.51
2.62
2.30
2.30
2.30
2.29
2.27
2.19
2.18
1.61
1.60
1.60
1.59
1.53
1.36
1.35
1.31
1.30
1.30
1.29
1.28
1.27
1.26
1.25
1.24
0.92
0.91
0.90
0.88
0.87
0.86
0.15
0.13
1.000
2.260
1.254
1.220
1.272
2.316
1.494
1.082
2.205
1.098
1.00
2.26
1.25
1.22
1.27
2.32
1.49
1.08
2.20
1.10
6.48
15.51
2.96
3.06
ppm


TBSO
OH
OH
O
O
9
Method #1
135.229
117.552
102.407
79.049
77.489
77.171
76.853
75.028
72.319
72.232
70.164
38.694
34.885
31.966
25.908
24.897
22.726
18.263
16.532
14.244
-4.261
-4.812
-Ipom-F-1-126-140724-1-140730 C13
130
120
110
100
90
80
70
60
50
40
30
20
10
0
ppm

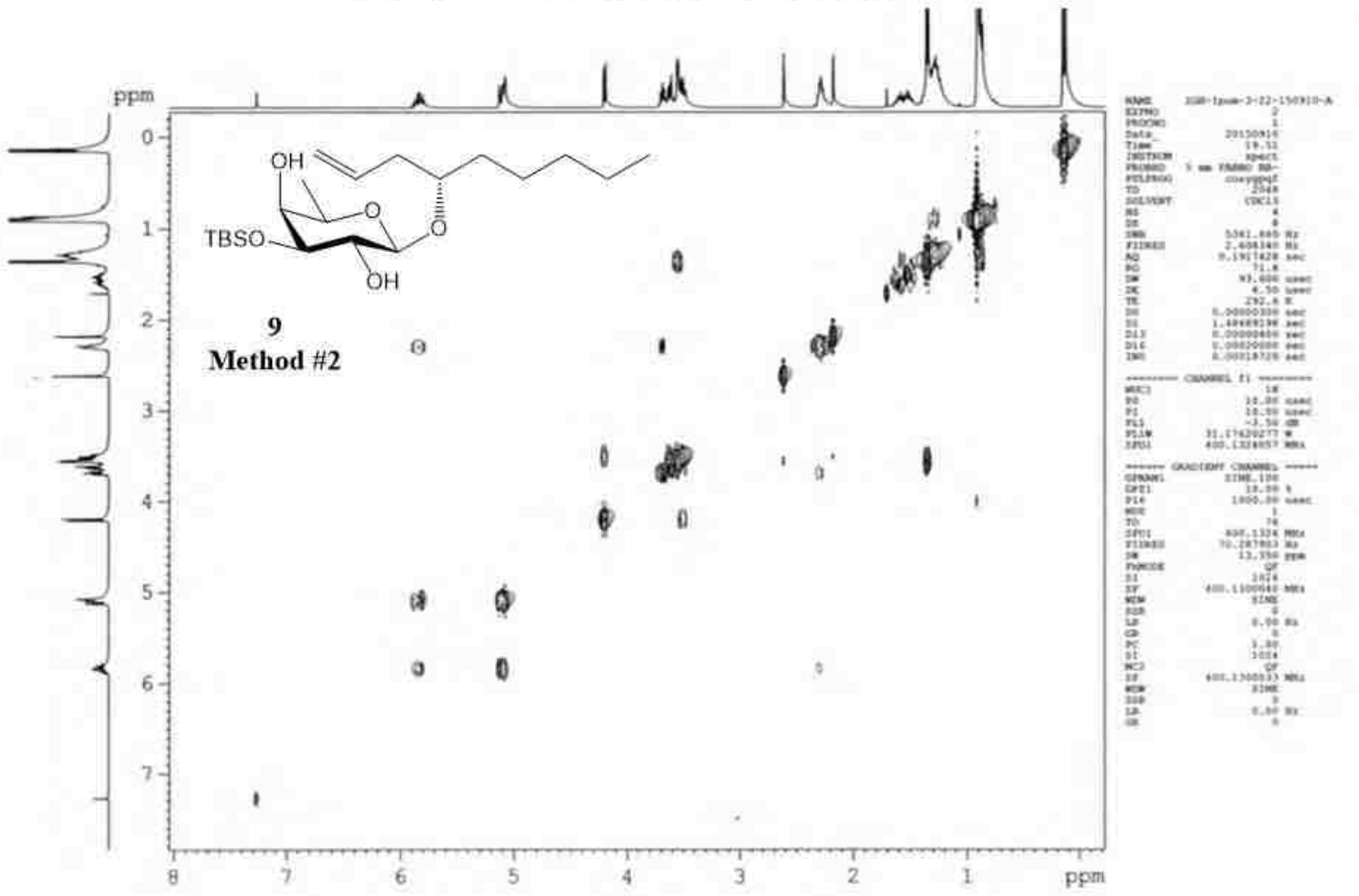

LW-Ipam-F-2-16-161019-170131 Repure in CDCl3 300 NMR
7.271
5.916
5.892
5.882
5.869
5.859
5.836
5.825
5.812
5.802
5.778
5.138
5.112
5.079
4.221
4.195
3.733
3.714
3.694
3.675
3.653
3.641
3.623
3.611
3.592
3.567
3.551
3.540
3.511
3.483
2.623
2.301
2.182
2.175
1.586
1.565
1.371
1.349
1.316
1.304
1.290
1.271
0.918
0.888
0.865
0.157
0.135
OH
O
O
TBSO
OH
9
Method #2
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
ppm
1.125
2.124
1.000
0.983
1.214
2.409
0.840
1.037
2.053
1.207
3.489
3.468
4.295
13.235
2.930
3.076

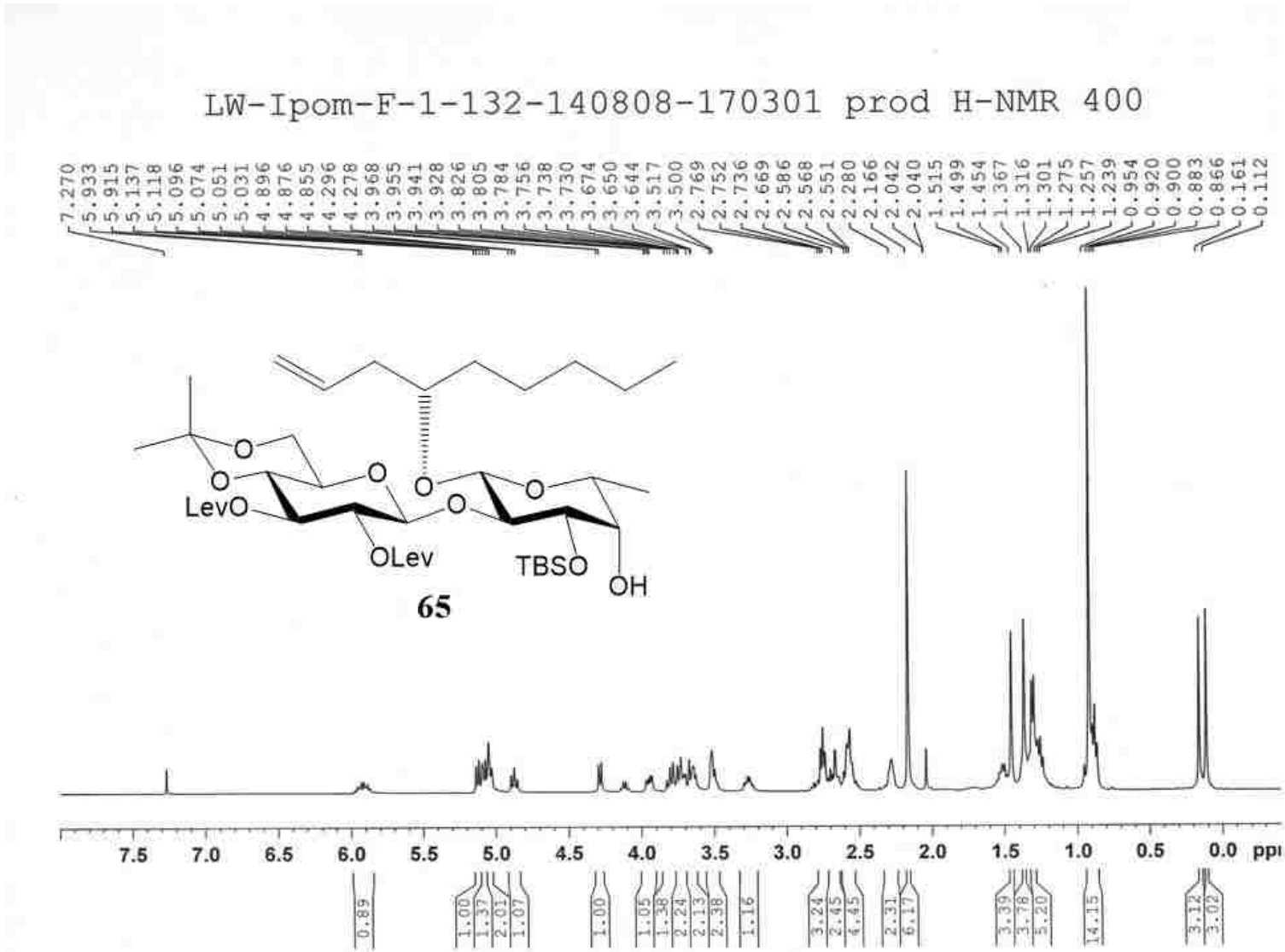
LW-Ipom-F-2-16-16019-17013 mixture of batches pure C13 300 NMR



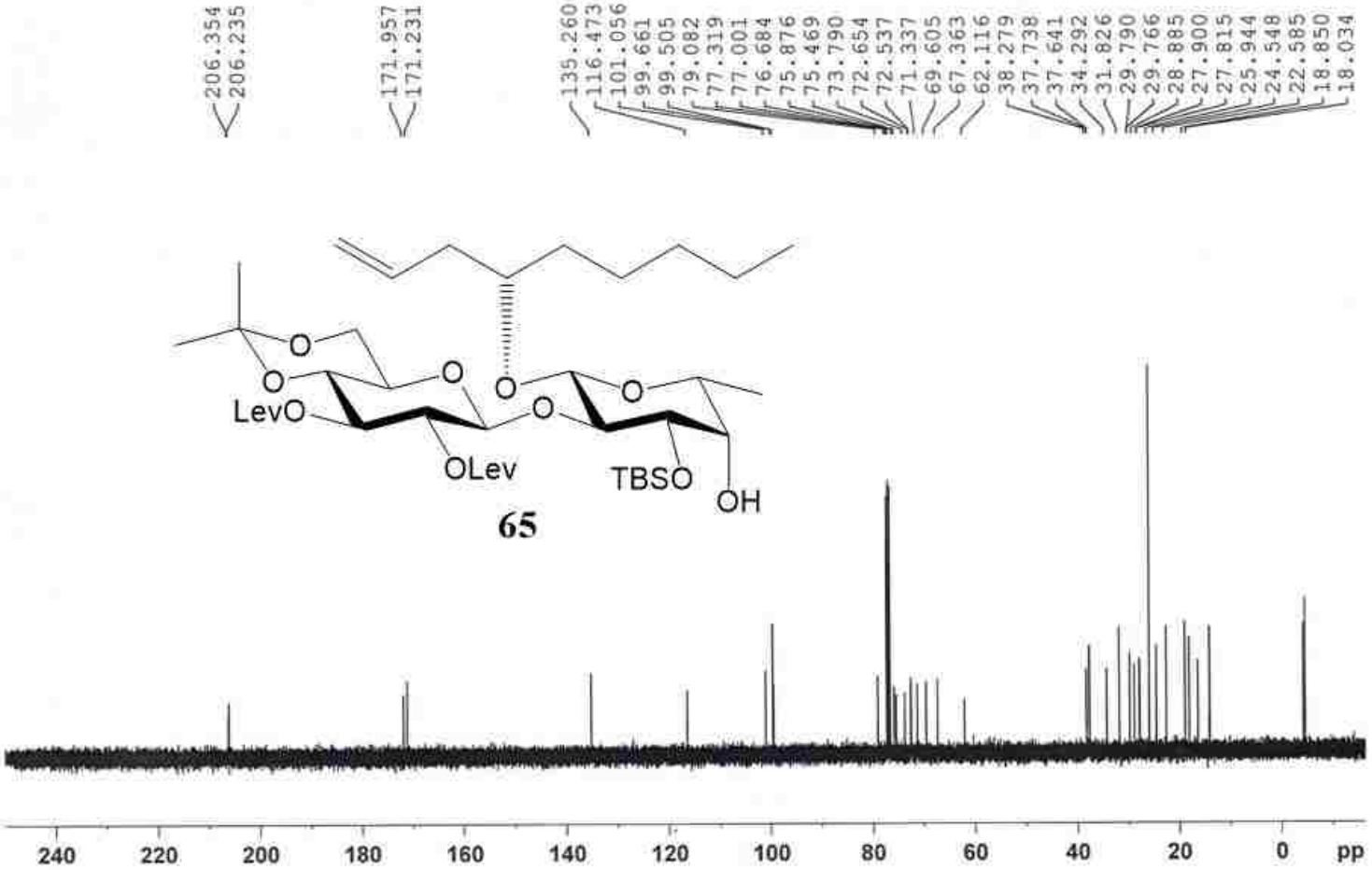
9
Method #2

LW-Ipom-F-1-126-140726 400 COSY

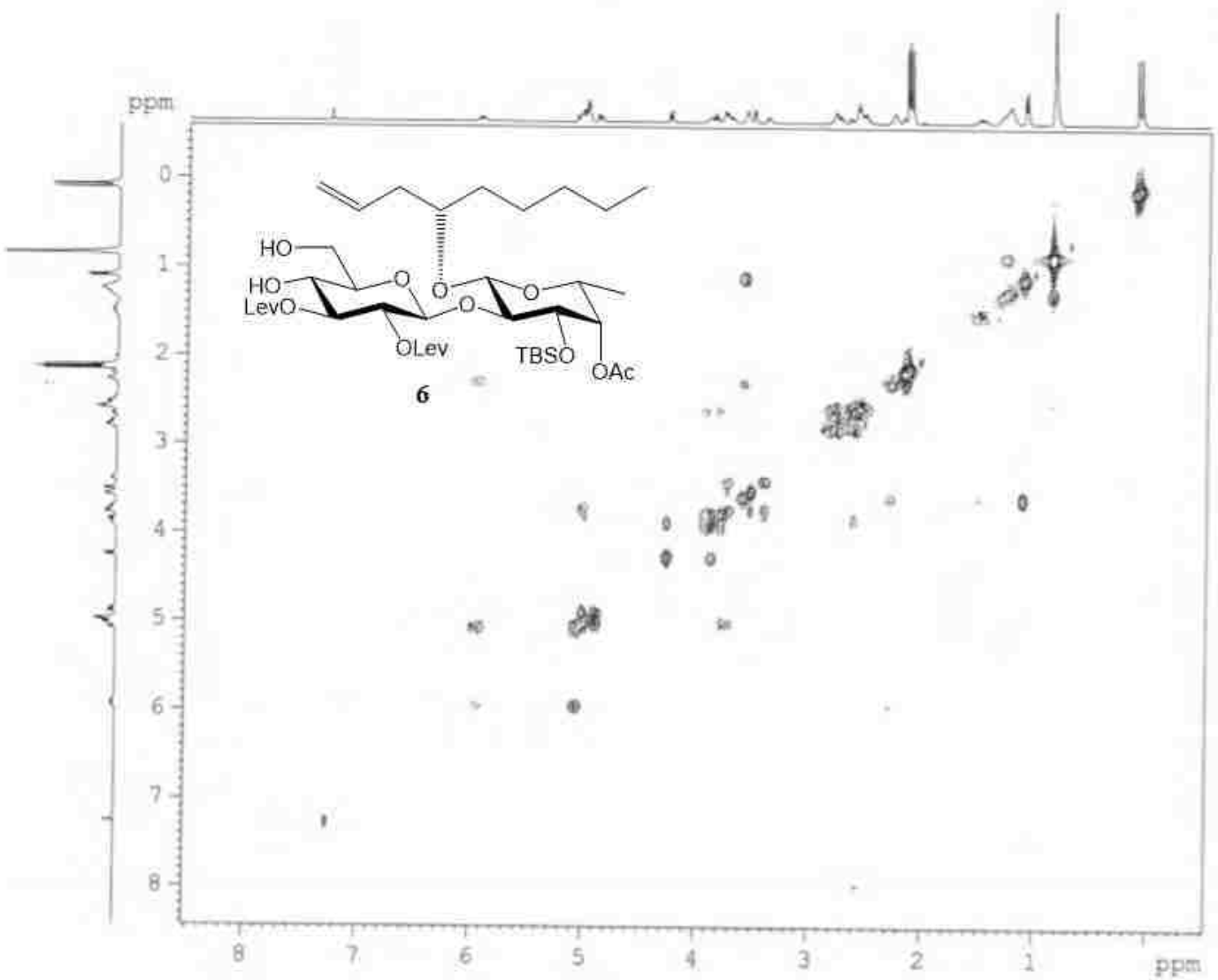


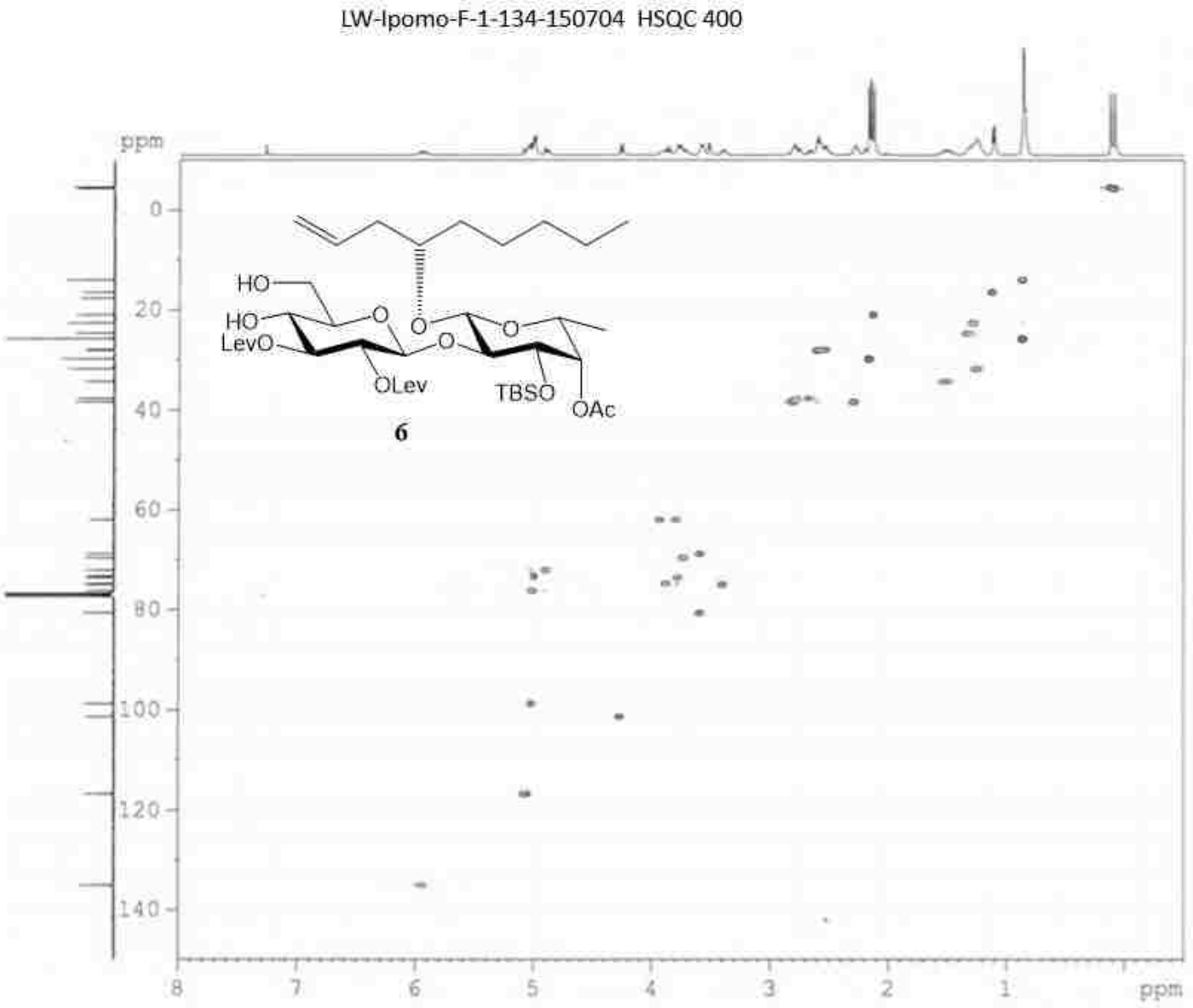

LW-Ipom-F-1-132-140808-170301 prod H-NMR 400
7.270
5.933
5.915
5.137
5.118
5.096
5.074
5.051
5.031
4.896
4.876
4.855
4.296
4.278
3.968
3.955
3.941
3.928
3.826
3.805
3.784
3.756
3.738
3.730
3.674
3.650
3.644
3.517
3.500
2.769
2.752
2.736
2.669
2.586
2.568
2.551
2.280
2.166
2.042
2.040
1.515
1.499
1.454
1.367
1.316
1.301
1.275
1.257
1.239
0.954
0.920
0.900
0.883
0.866
0.161
0.112
LevO
OLev
TBSO
OH
65
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
ppm
0.89
1.00
1.37
2.01
1.07
1.00
1.05
1.38
2.24
2.13
2.38
1.16
3.24
2.45
4.45
2.31
6.17
3.39
3.78
5.20
14.15
3.12
3.02

LW-Ipom-F-1-132-140808-170301 Prod C-NMR 400

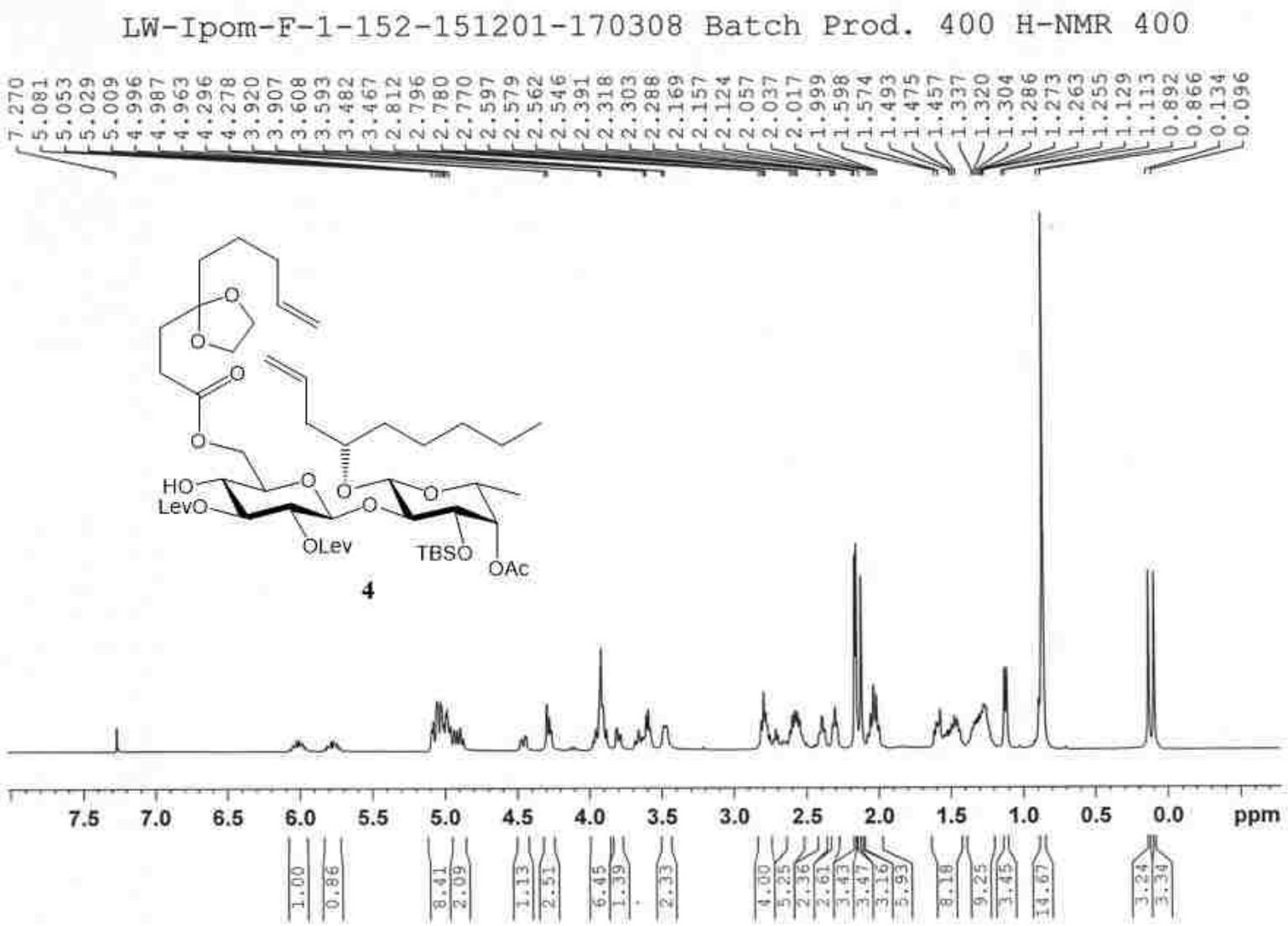


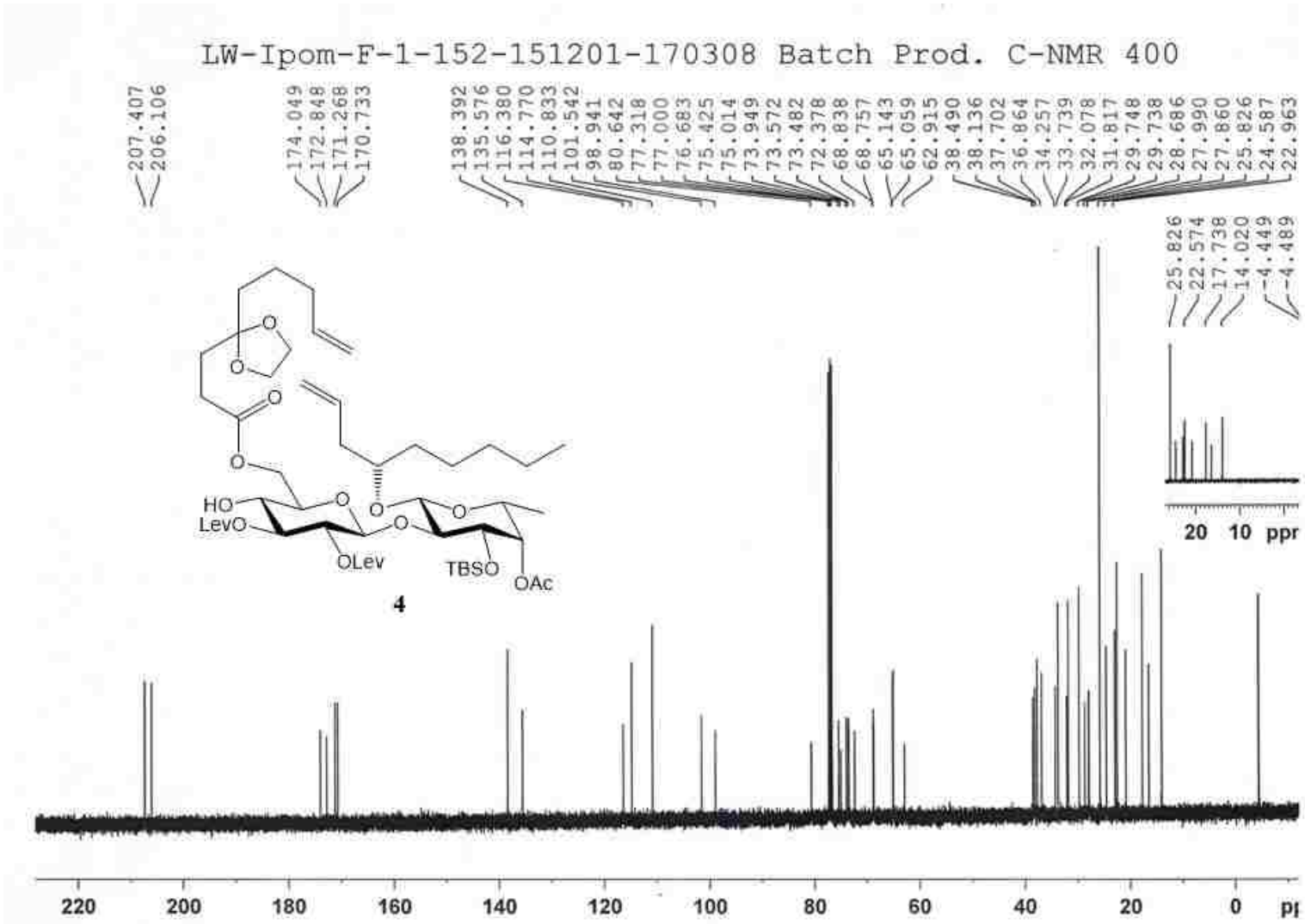
LW-Ipomo-F-1-134-150704 COSY 400



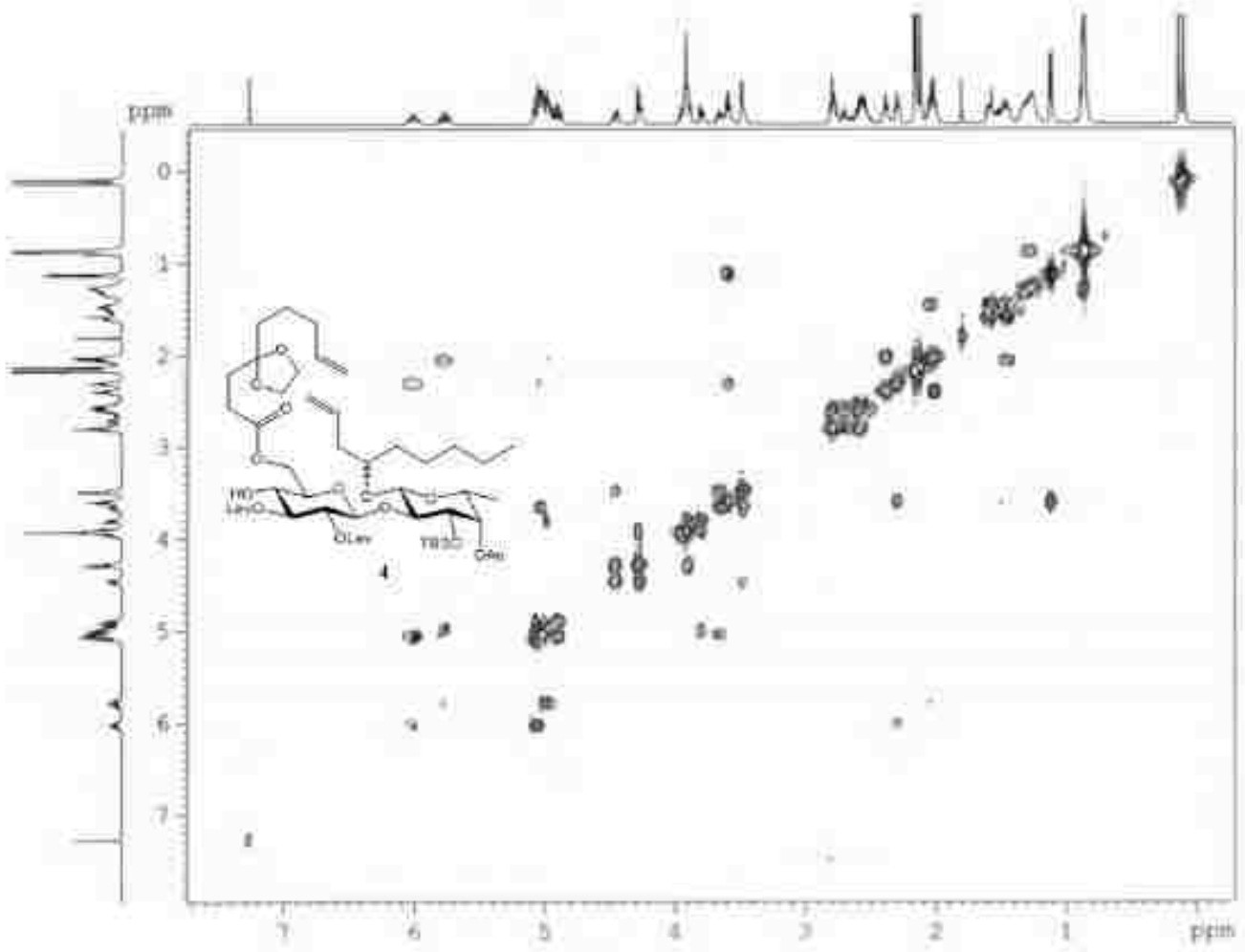

LW-Ipomo-F-1-134-150704 HSQC 400
ppm
0
20
40
60
80
100
120
140
8
7
6
5
4
3
2
1
ppm
HO
HO
LevO
OLev
TBSO
OAc
O
O
O
O

6

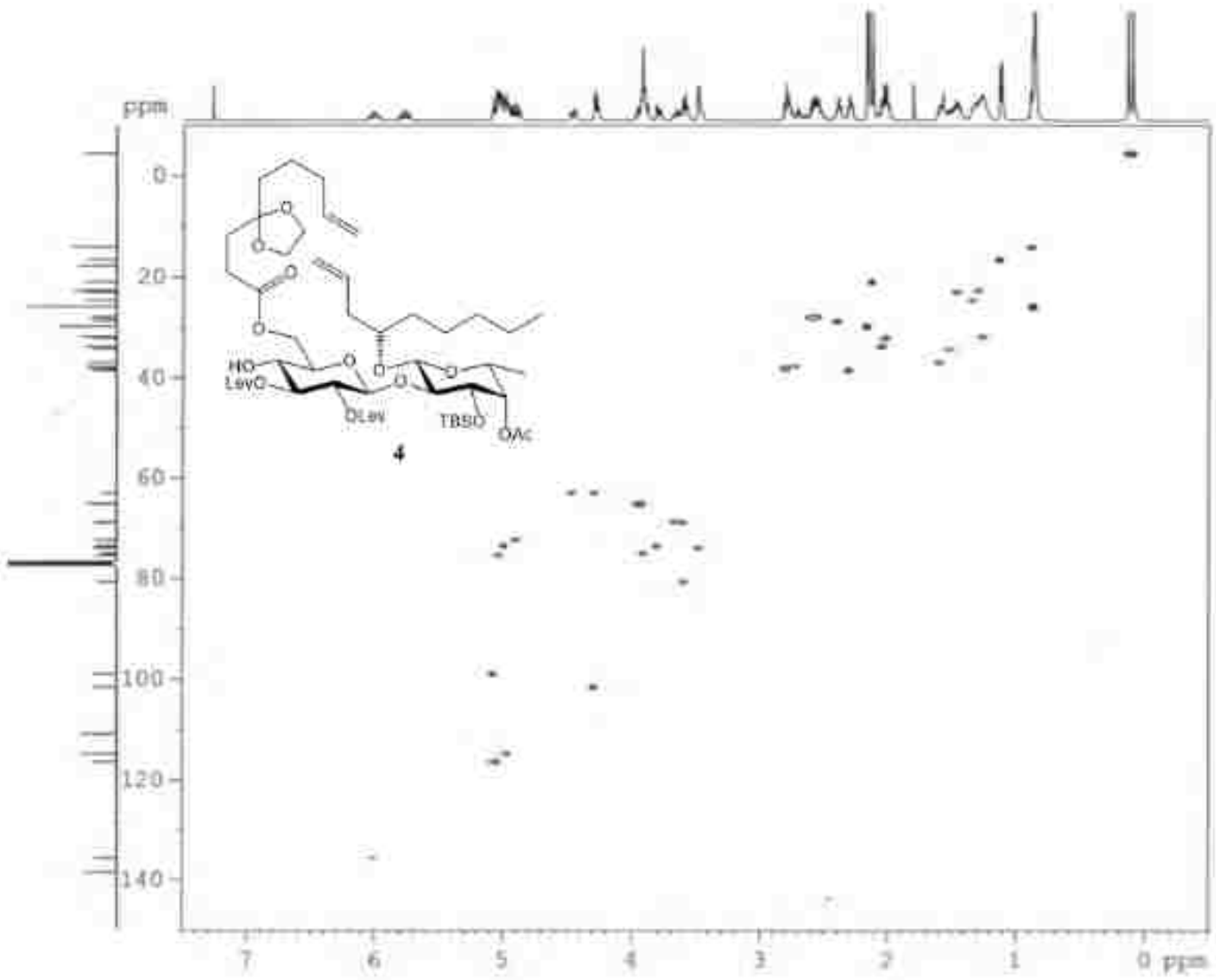

LW-Ipom-F-1-152-151201-170308 Batch Prod. 400 H-NMR 400
7.270
5.081
5.053
5.029
5.009
4.996
4.987
4.963
4.296
4.278
3.920
3.907
3.608
3.593
3.482
3.467
2.812
2.796
2.780
2.770
2.597
2.579
2.562
2.546
2.391
2.318
2.303
2.288
2.169
2.157
2.124
2.057
2.037
2.017
1.999
1.598
1.574
1.493
1.475
1.457
1.337
1.320
1.304
1.286
1.273
1.263
1.255
1.129
1.113
0.892
0.866
0.134
0.096
HO
LevO
OLev
TBSO
OAc
4
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
ppm
1.00
0.86
8.41
2.09
1.13
2.51
6.45
1.39
2.33
4.00
5.25
2.36
2.61
3.43
3.47
3.16
5.93
8.18
9.25
3.45
14.67
3.24
3.34

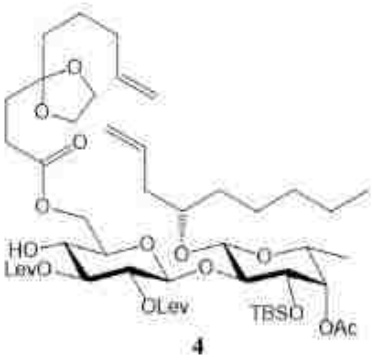

LW-Ipom-F-1-152-151201-170308 Batch Prod. C-NMR 400
207.407
206.106
174.049
172.848
171.268
170.733
138.392
135.576
116.380
114.770
110.833
101.542
98.941
80.642
77.318
77.000
76.683
75.425
75.014
73.949
73.572
73.482
72.378
68.838
68.757
65.143
65.059
62.915
38.490
38.136
37.702
36.864
34.257
33.739
32.078
31.817
29.748
29.738
28.686
27.990
27.860
25.826
24.587
22.963
25.826
22.574
17.738
14.020
-4.449
-4.489
HO
LevO
OLev
TBSO
OAc
4
20 10 ppr
220 200 180 160 140 120 100 80 60 40 20 0 pp

LW-Ipom-152-151201-170308 Product Batch Prod COSY



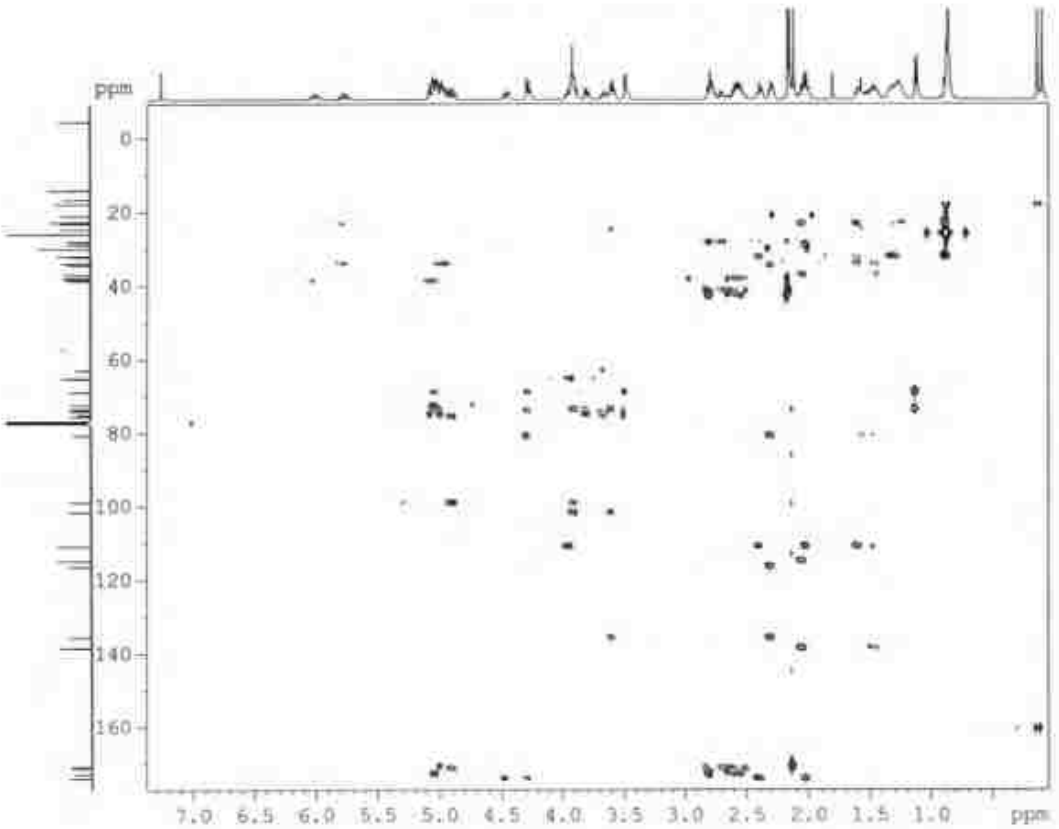
LW-Ipom-152-151201-170308 Product Batch Prod HSQC



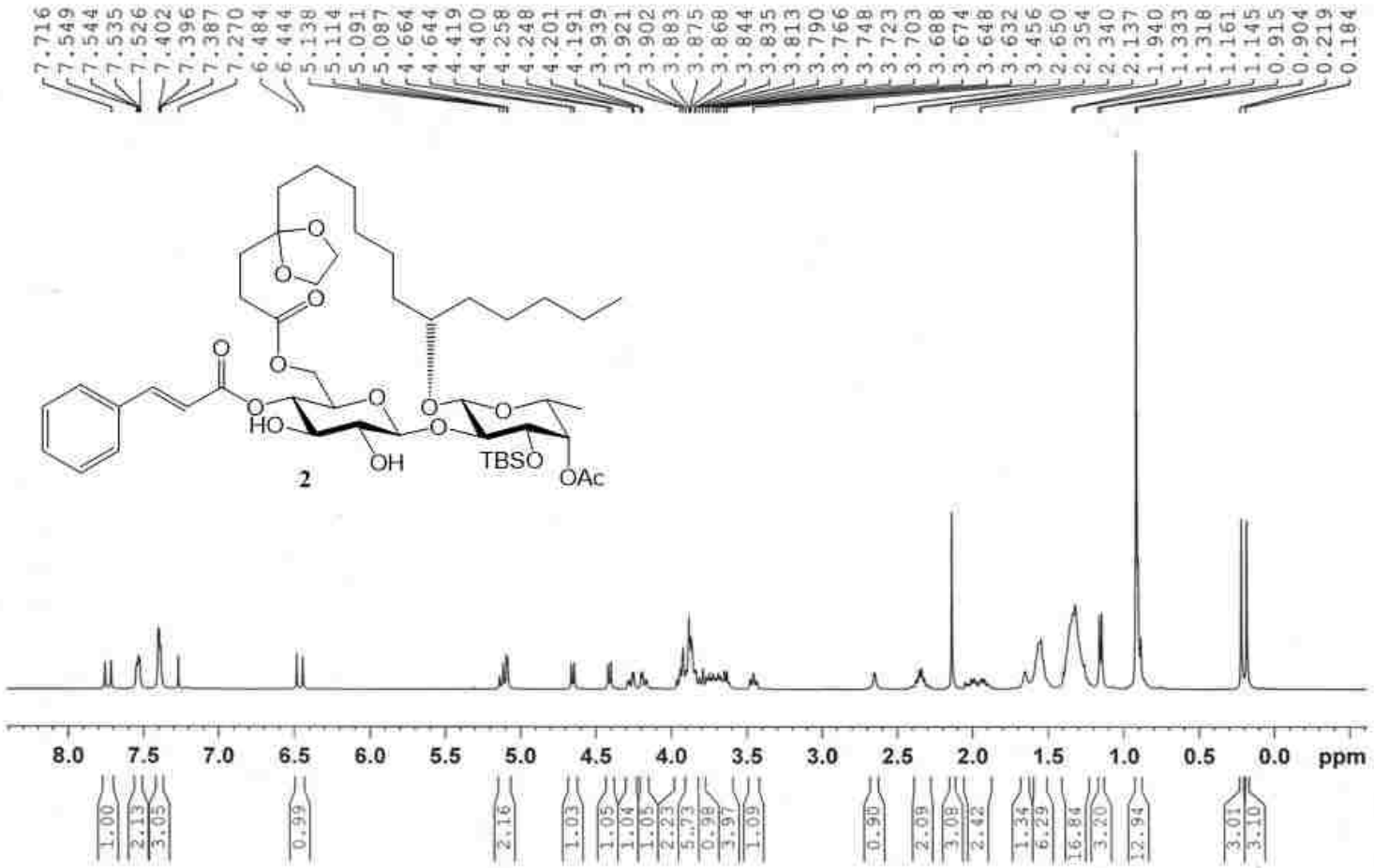


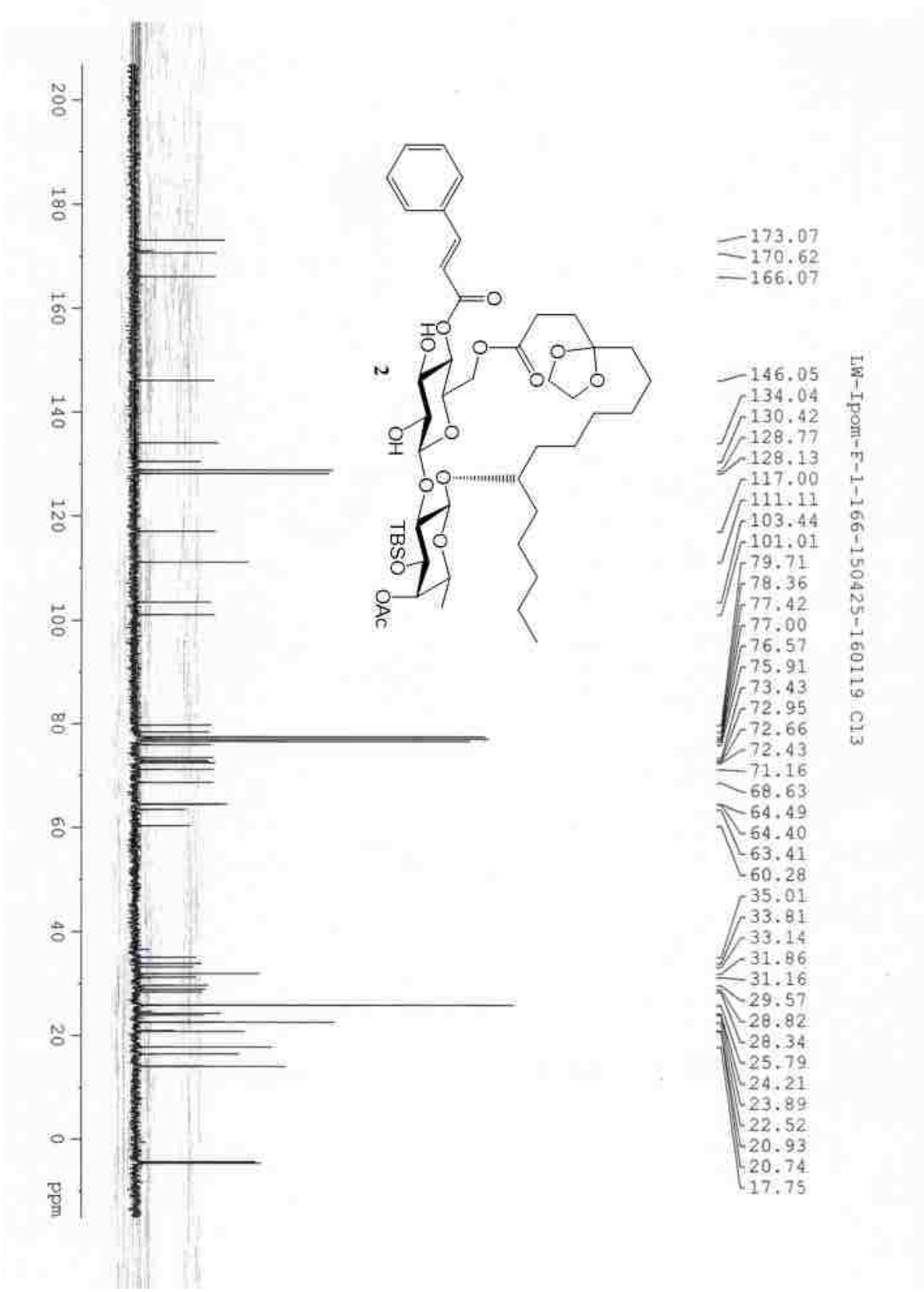
4

LW-Ipom-152-151201-170308 Product Batch Prod HMBC

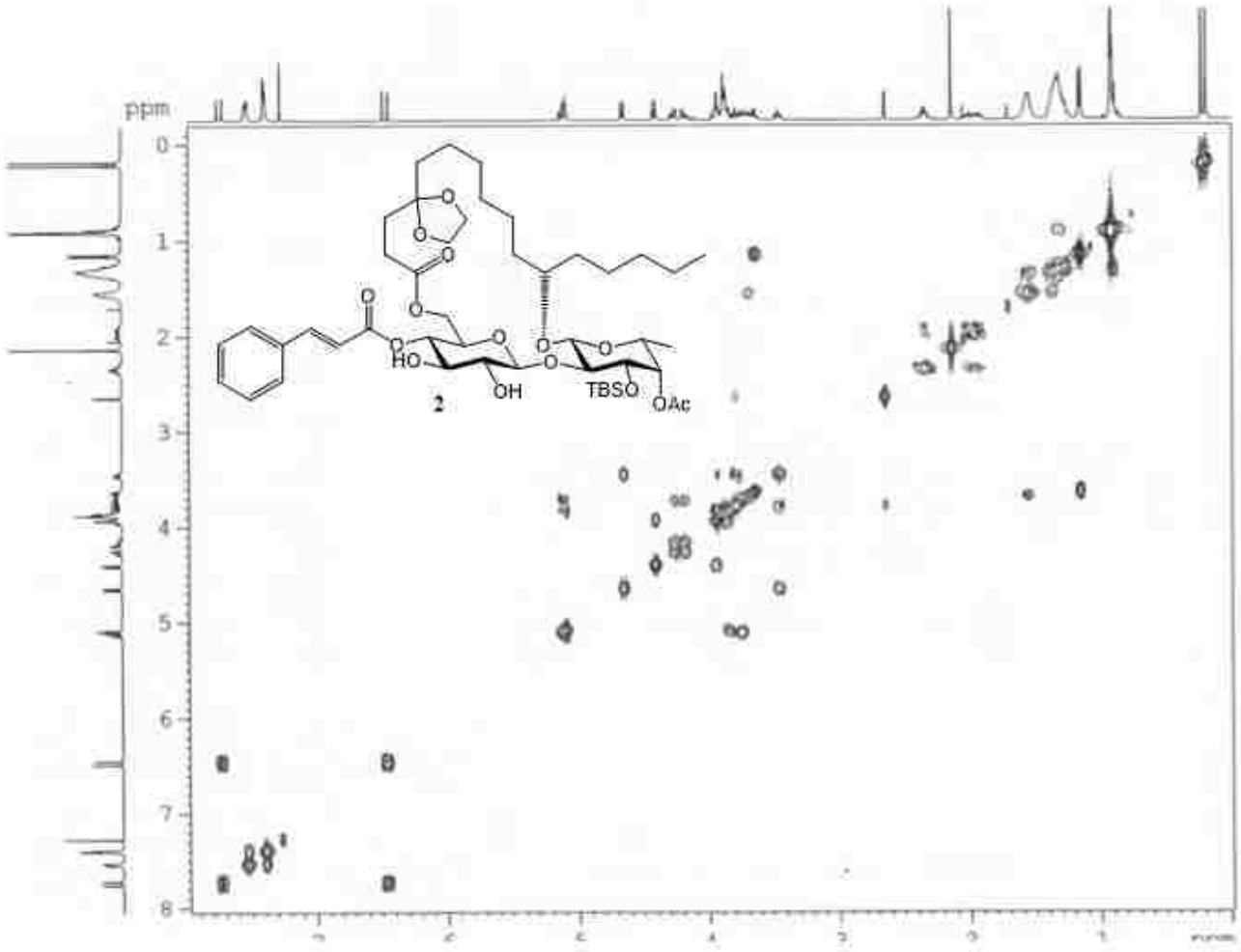


LW-Ipom-F-1-166-150425-170328 Prod. diluted H-NMR 400

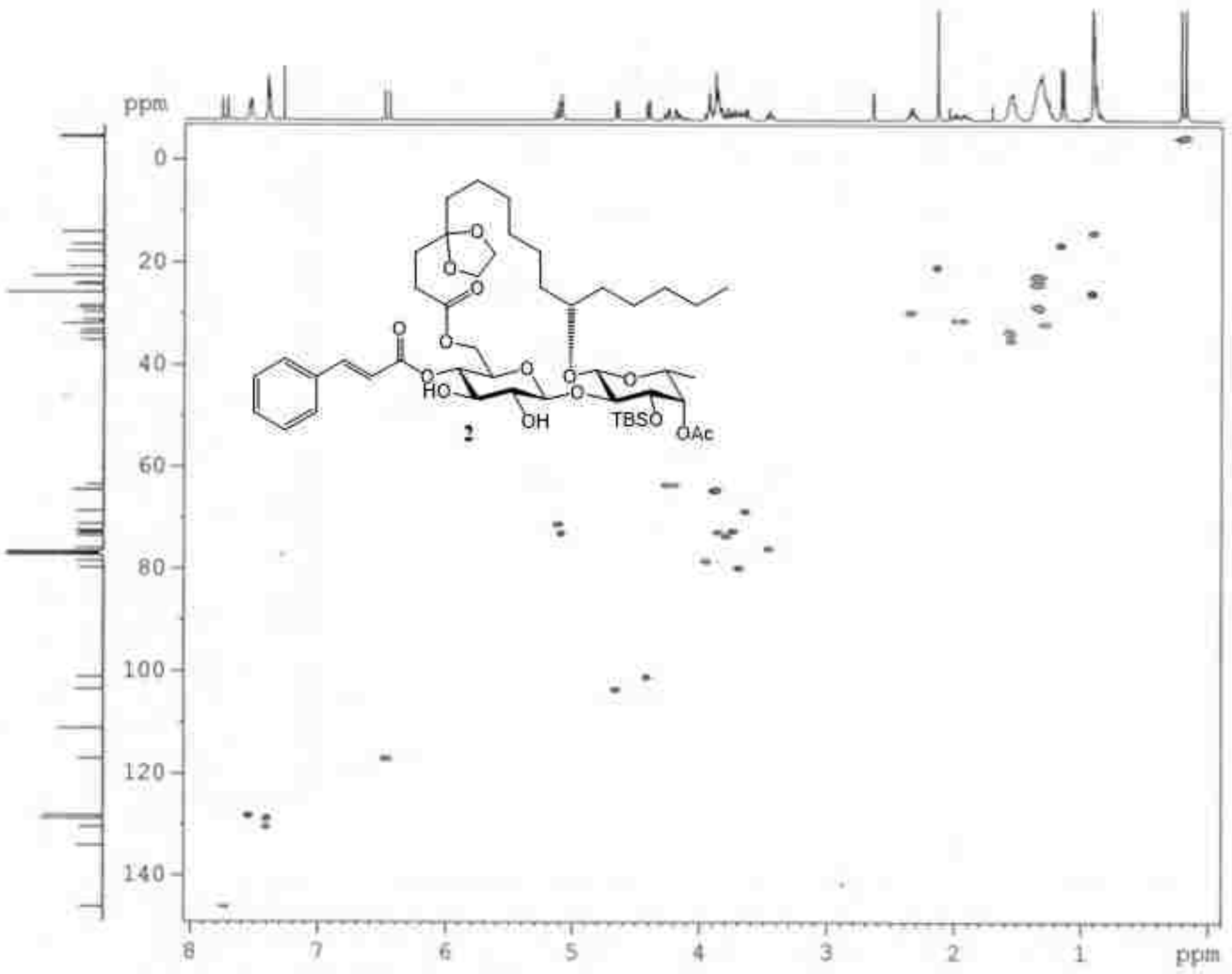



IW-Ipom-F-1-166-150425-160119 C13
173.07
170.62
166.07
146.05
134.04
130.42
128.77
128.13
117.00
111.11
103.44
101.01
79.71
78.36
77.42
77.00
76.57
75.91
73.43
72.95
72.66
72.43
71.16
68.63
64.49
64.40
63.41
60.28
35.01
33.81
33.14
31.86
31.16
29.57
28.82
28.34
25.79
24.21
23.89
22.52
20.93
20.74
17.75
HO
OH
TBSO
OAc
2
200
180
160
140
120
100
80
60
40
20
0
ppm

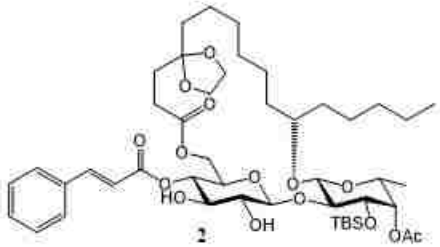
LW-Ipom-F-1-166-150425-160119 COSY

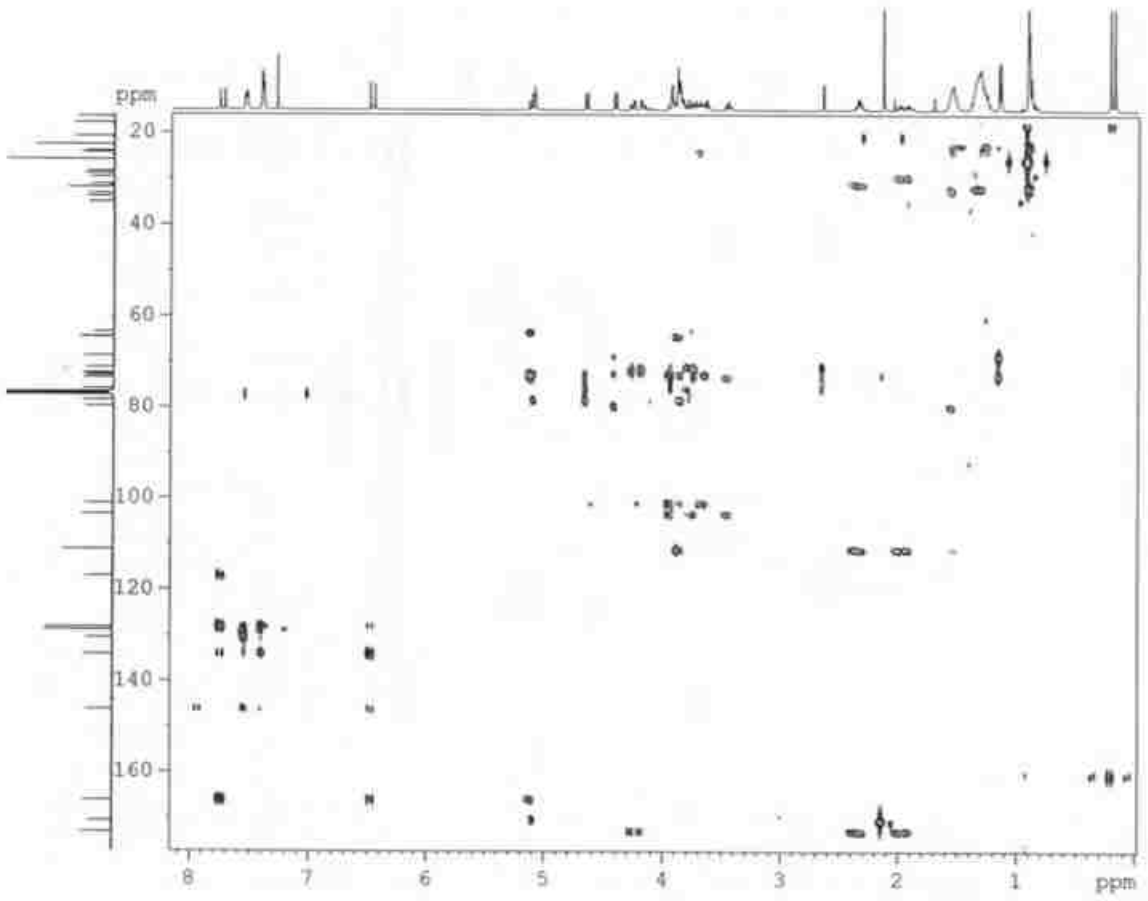


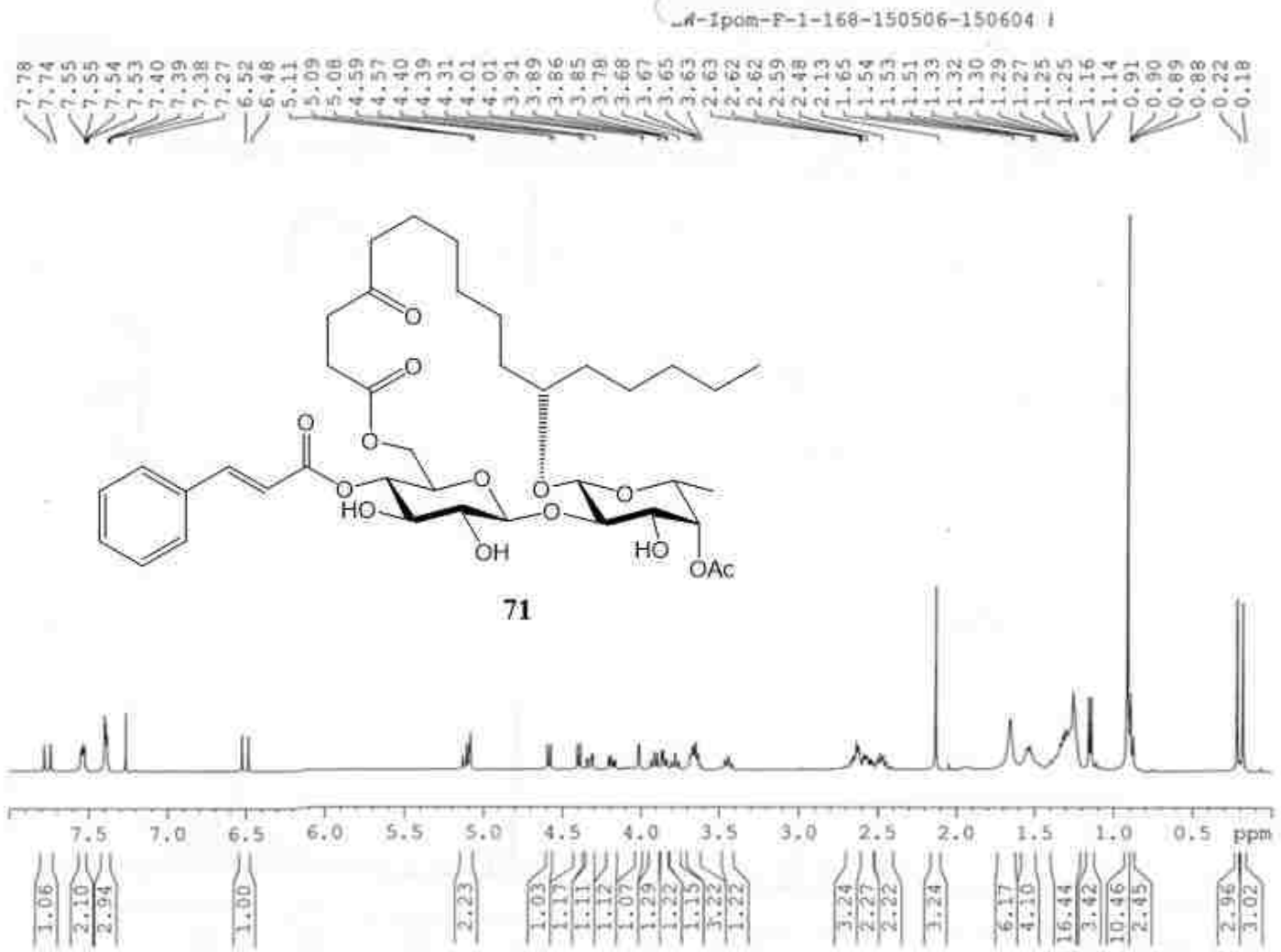
LW-Ipom-F-1-166-150425-160119 HSQC



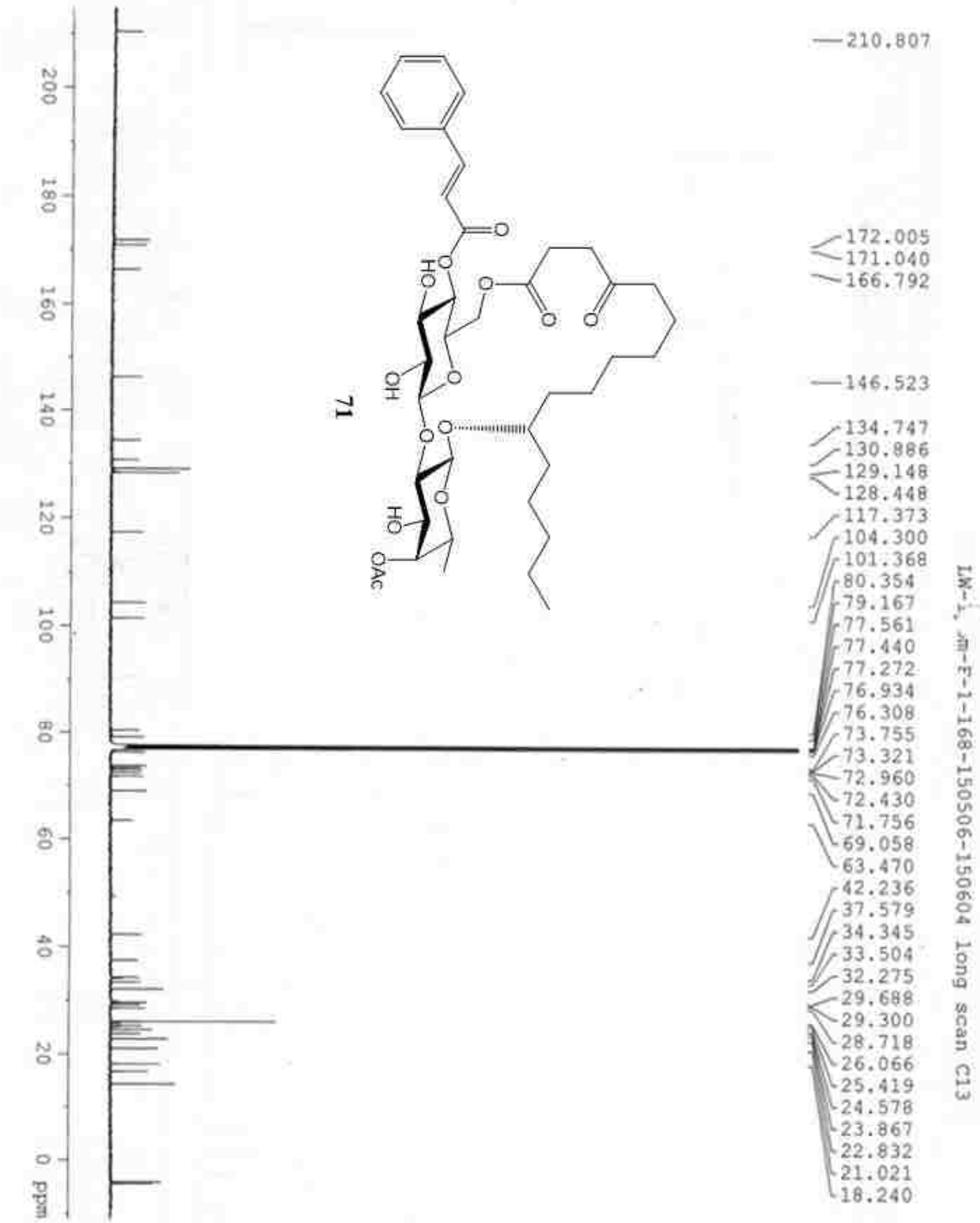
LW-Ipom-F-1-166-150425-160119 HMBC

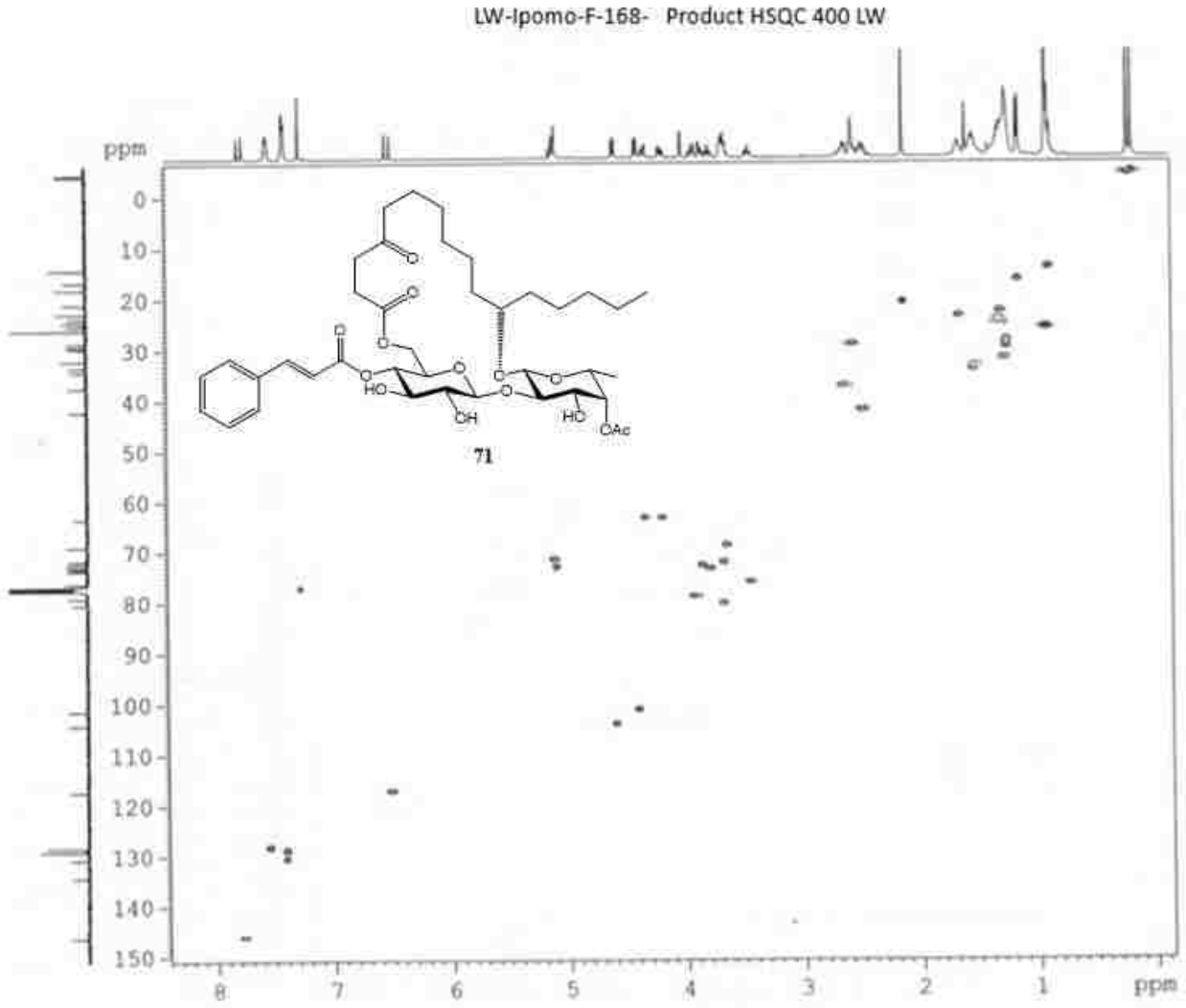




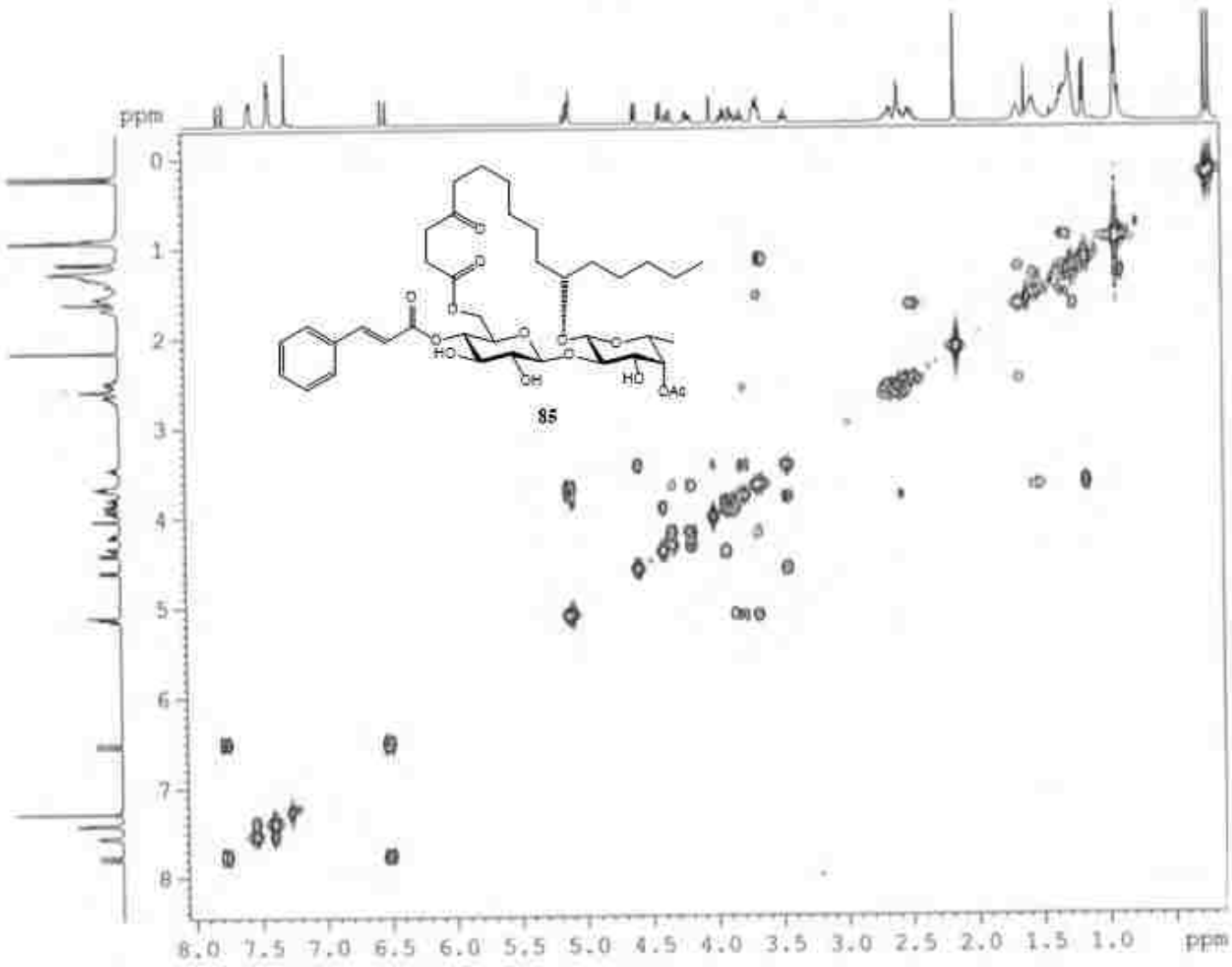

_M-Ipam-F-1-168-150506-150604 1
7.78
7.74
7.55
7.55
7.54
7.53
7.40
7.39
7.38
7.27
6.52
6.48
5.11
5.09
5.08
4.59
4.57
4.40
4.39
4.31
4.01
4.01
3.91
3.89
3.86
3.85
3.79
3.68
3.67
3.65
3.63
2.63
2.62
2.62
2.59
2.48
2.13
1.65
1.54
1.53
1.51
1.33
1.32
1.30
1.29
1.27
1.25
1.25
1.16
1.14
0.91
0.90
0.89
0.88
0.22
0.18
O
O
O
O
O
HO
OH
O
O
O
HO
OAc
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
ppm
1.06
2.10
2.94
1.00
2.23
1.03
1.17
1.11
1.12
1.07
1.29
1.22
1.15
3.22
1.22
3.24
2.27
2.22
3.24
6.17
4.10
16.44
3.42
10.46
2.45
2.96
3.02

71

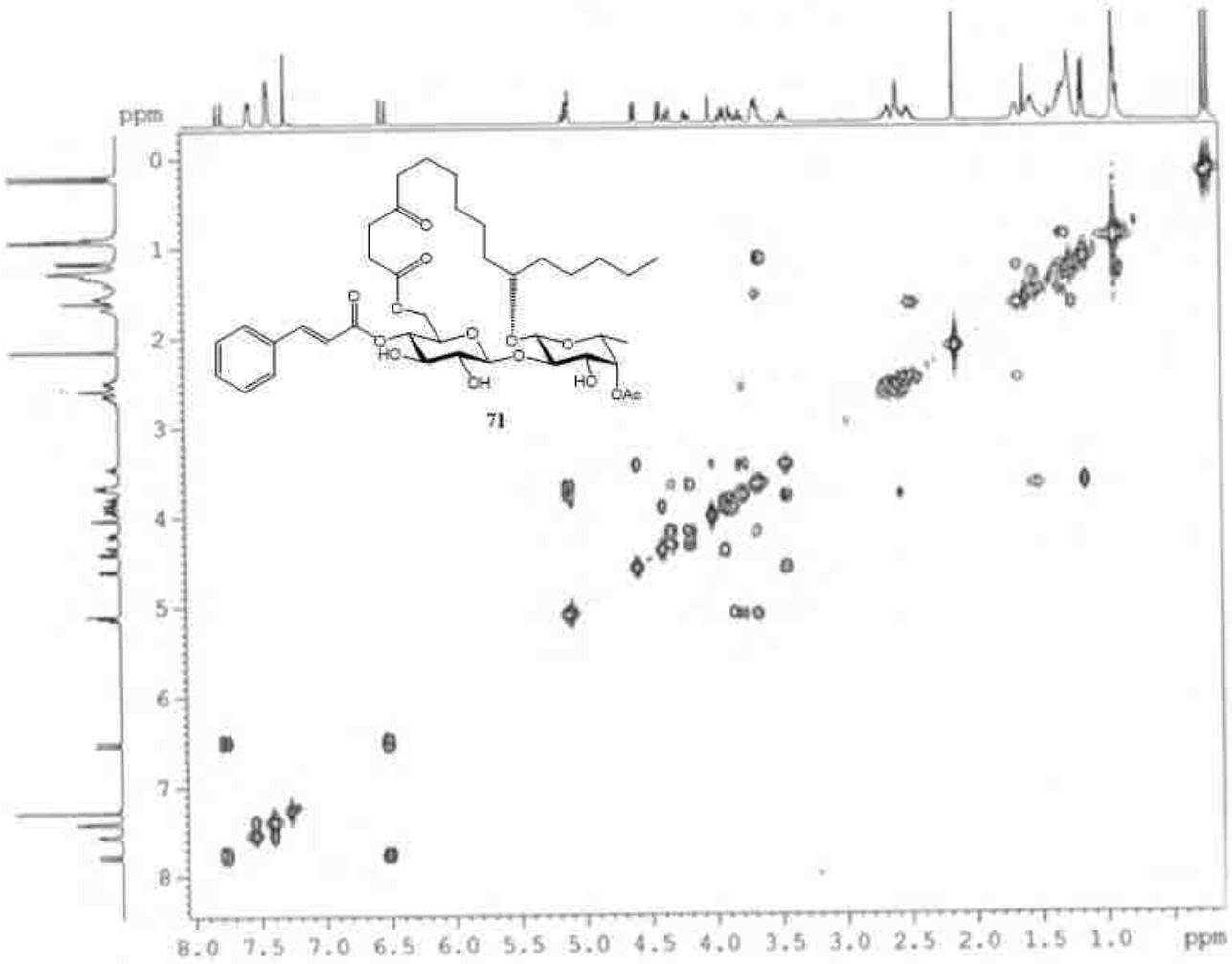

210.807
172.005
171.040
166.792
146.523
134.747
130.886
129.148
128.448
117.373
104.300
101.368
80.354
79.167
77.561
77.440
77.272
76.934
76.308
73.755
73.321
72.960
72.430
71.756
69.058
63.470
42.236
37.579
34.345
33.504
32.275
29.688
29.300
28.718
26.066
25.419
24.578
23.867
22.832
21.021
18.240
LW-1, JM-F-1-168-150506-150604 long scan C13
71
HO
OH
HO
OAc
O
200
180
160
140
120
100
80
60
40
20
0
ppm


LW-Ipomo-F-168- Product HSQC 400 LW
ppm
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
8
7
6
5
4
3
2
1
ppm
HO
OH
HO
OAc
71

LW-Ipomo-F-168- Product HMBC 400 LW

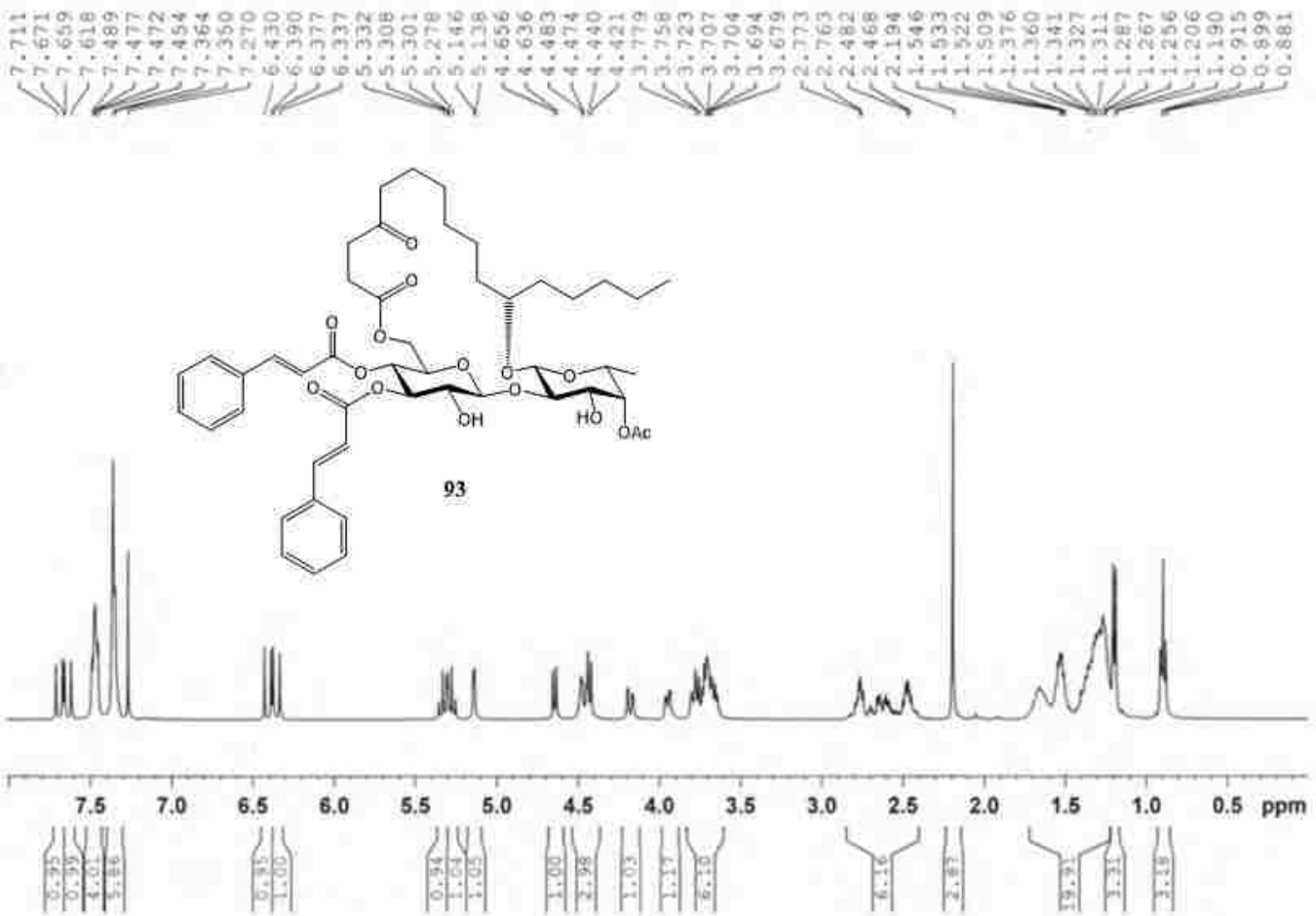


LW-Ipomo-F-168- Product HMBC 400 LW

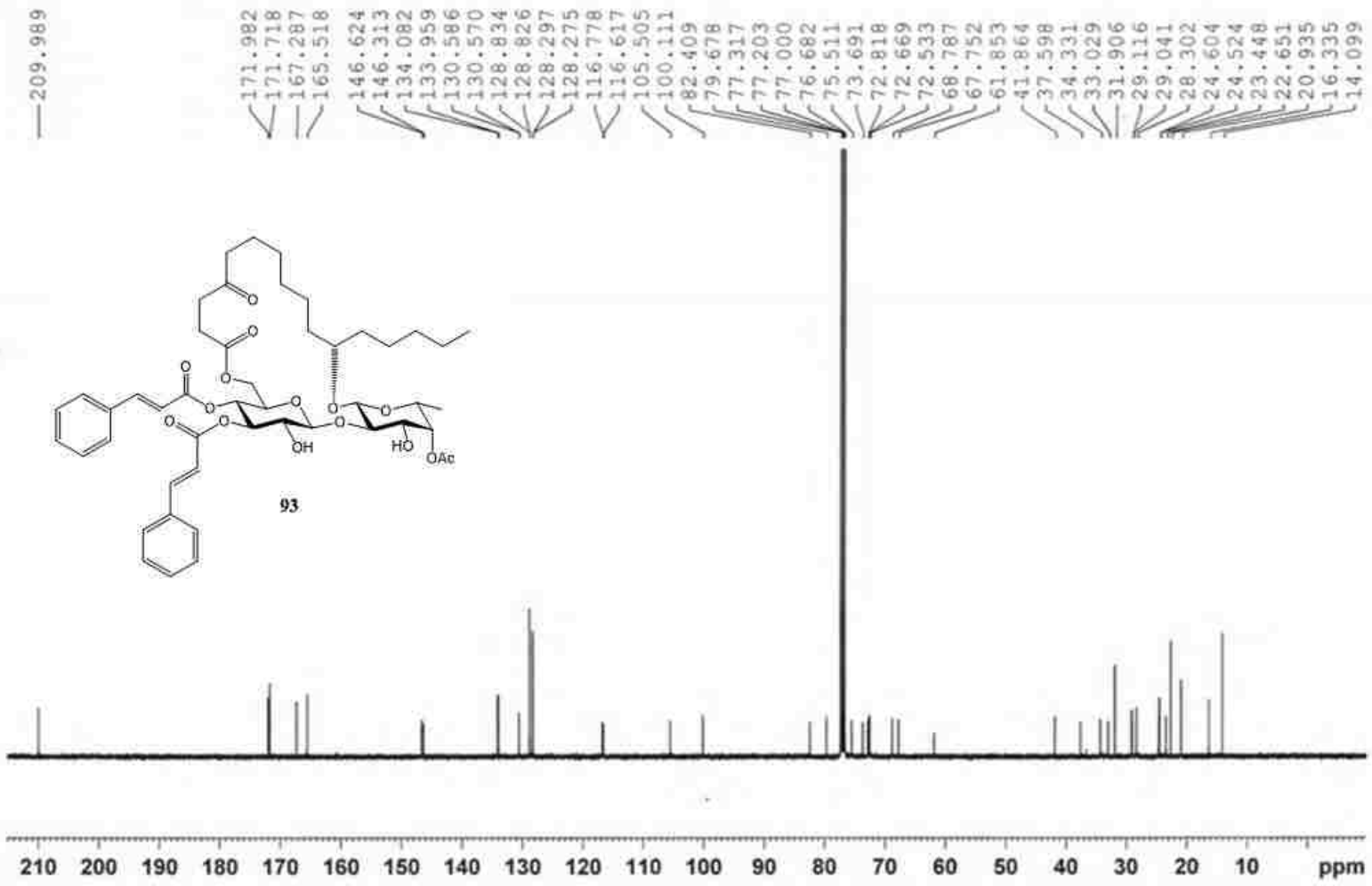


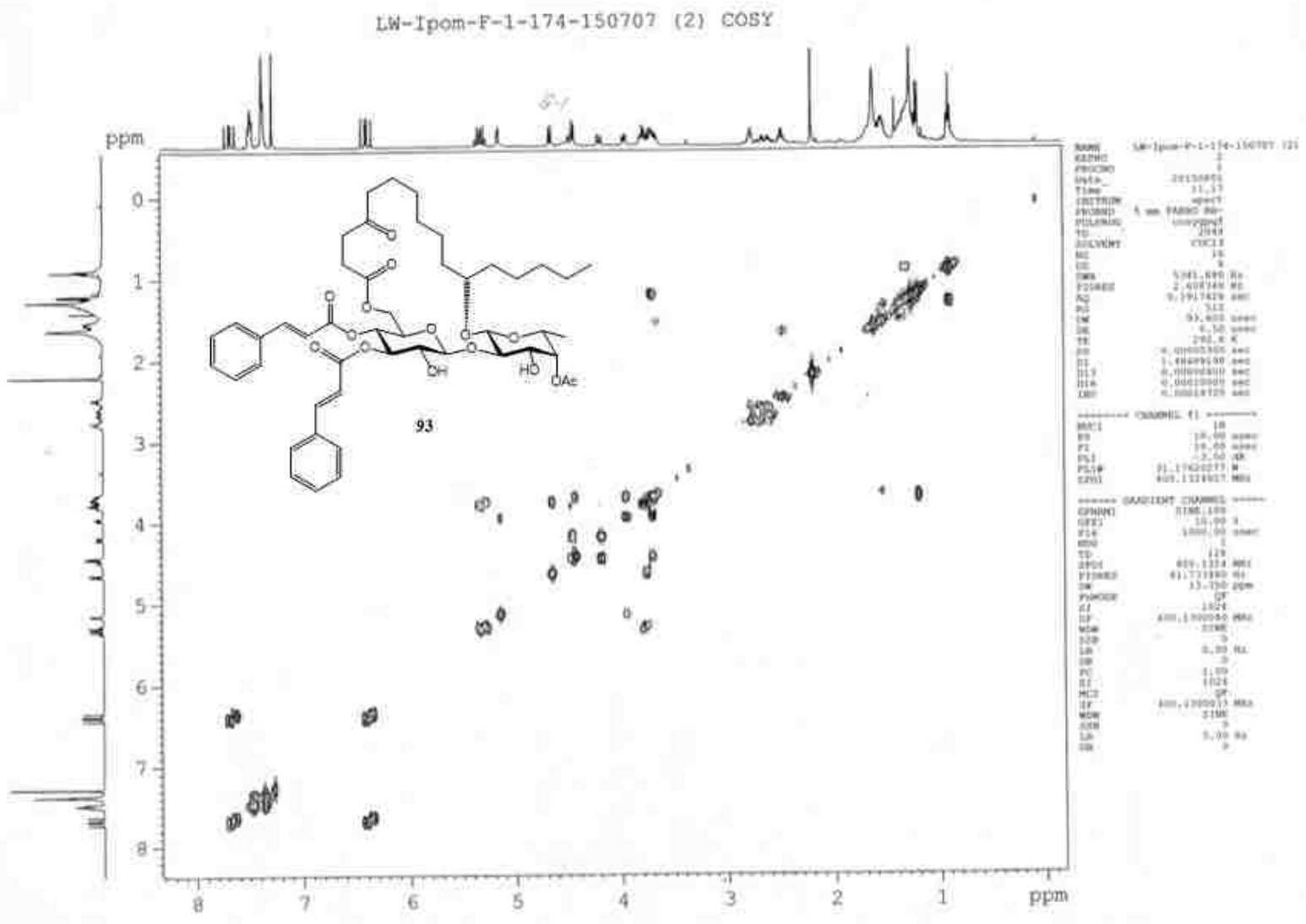
71

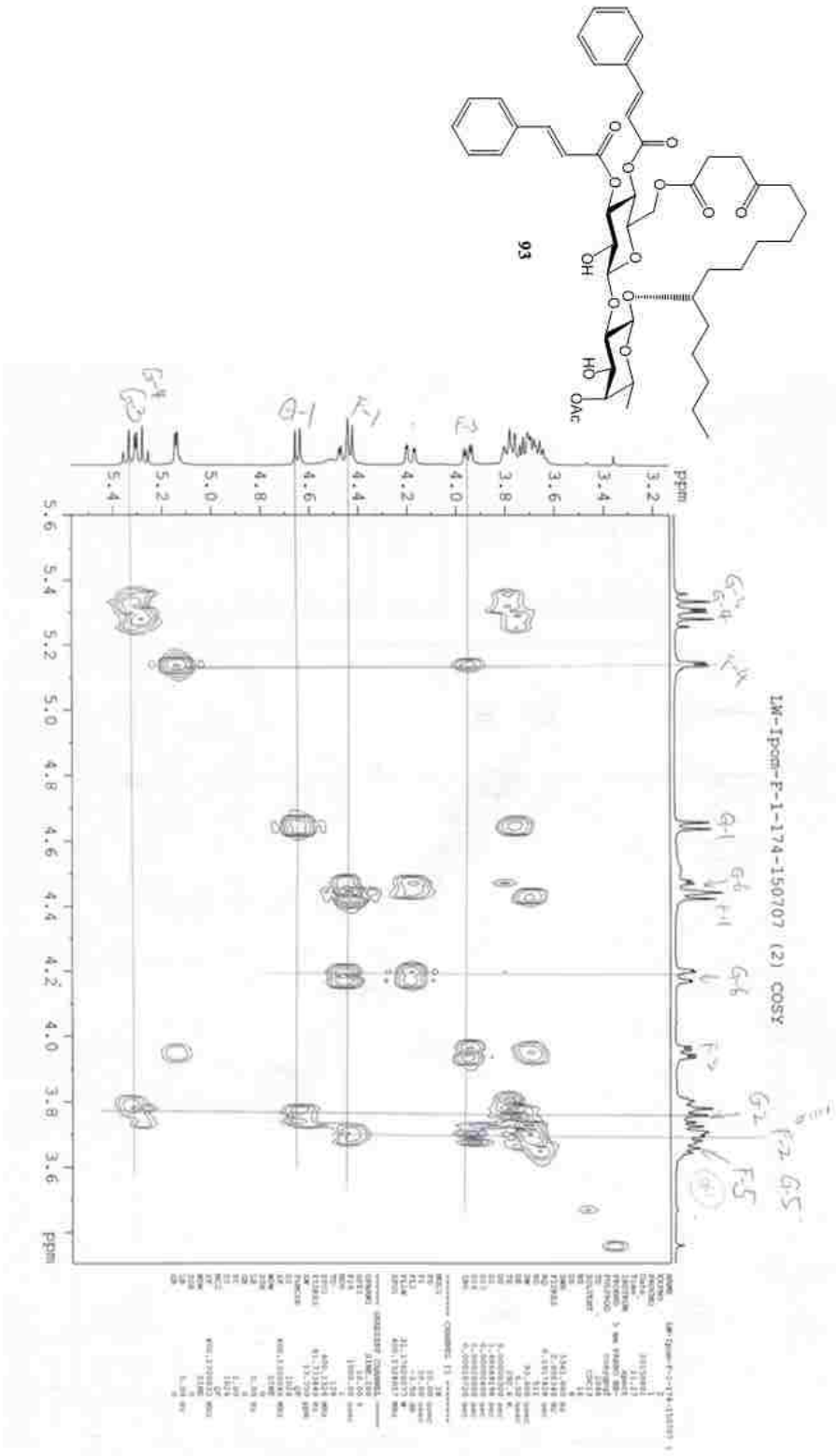
LW-Ipomo-F-1-174-150507 C-NMR 400

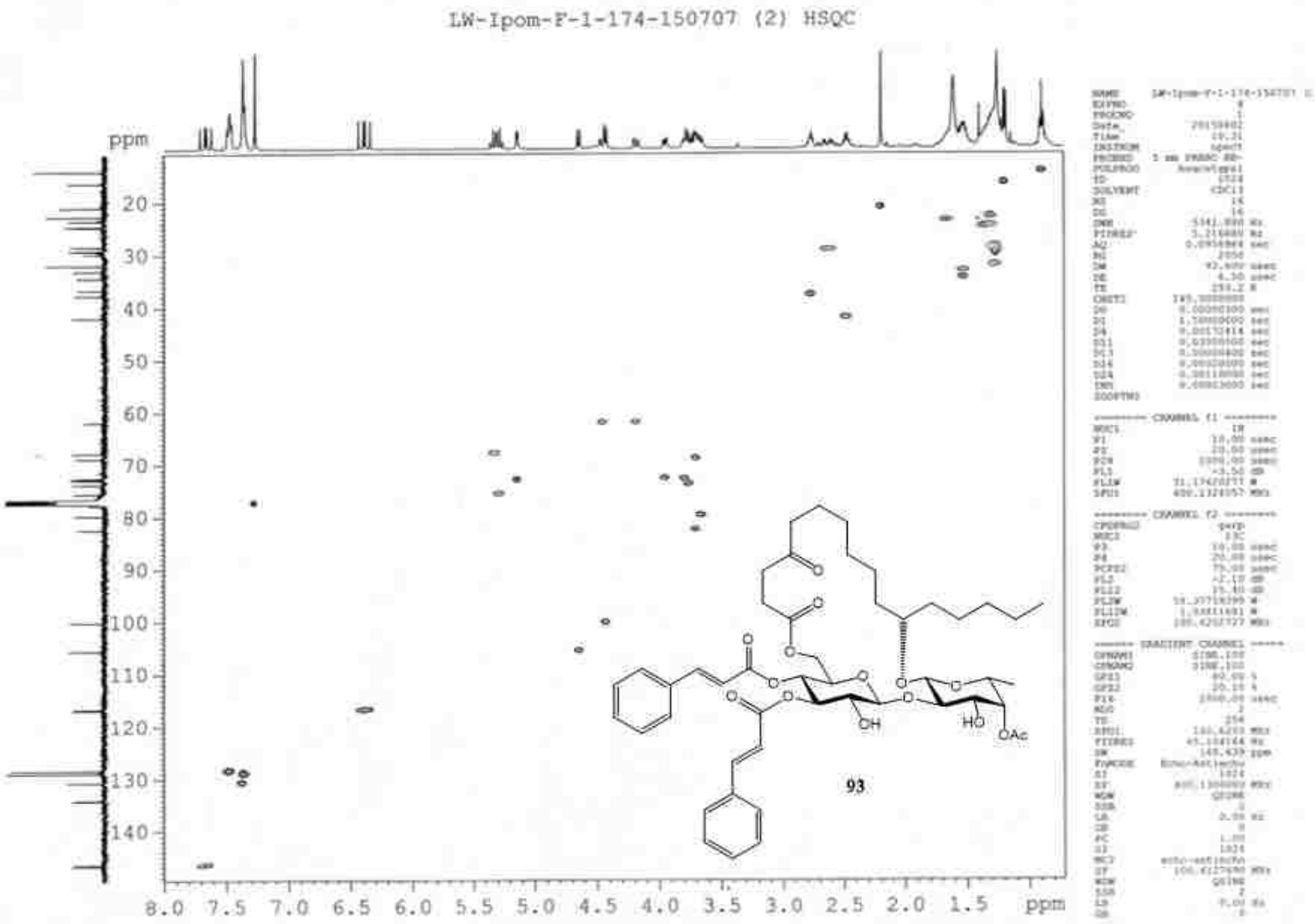


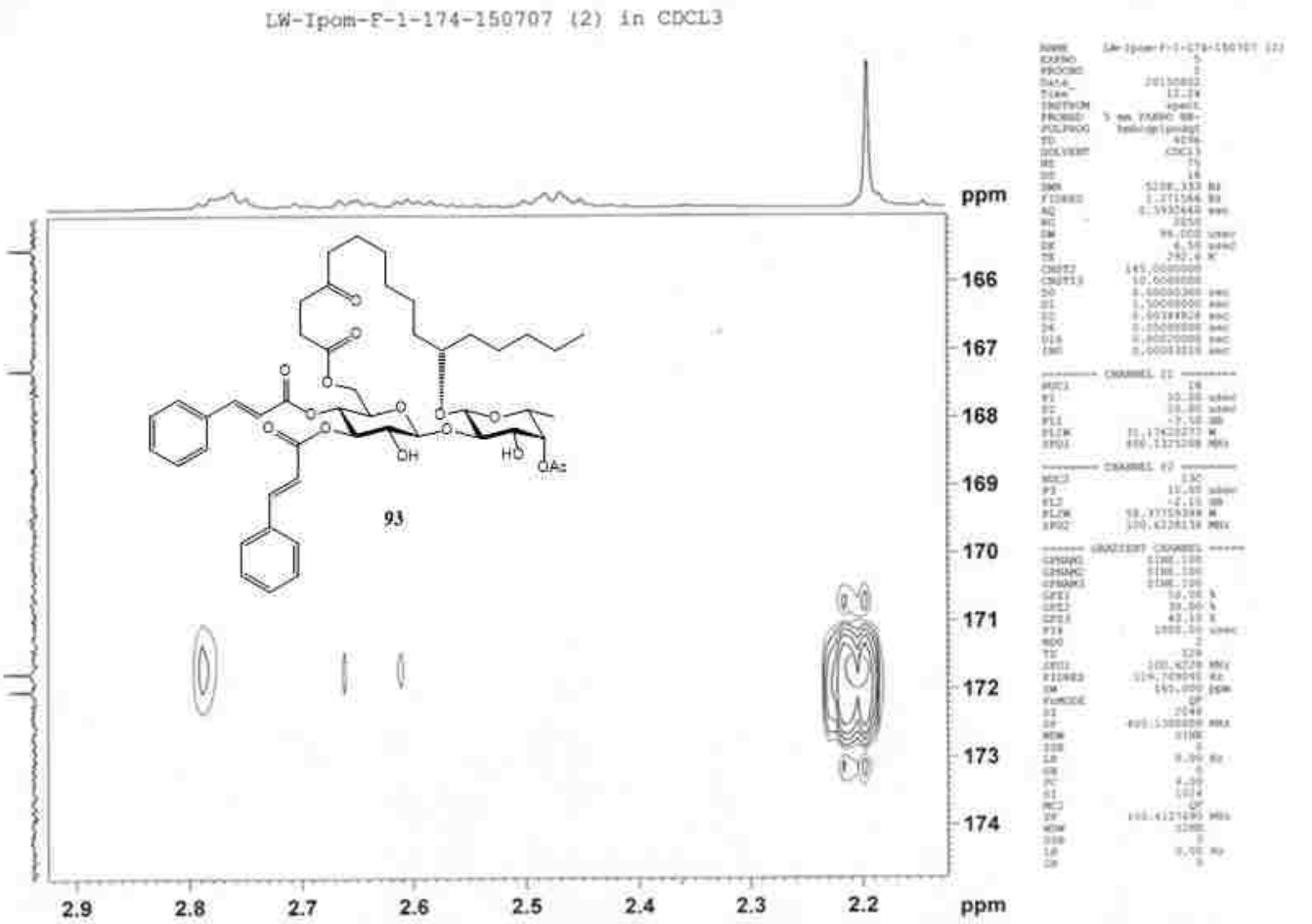
LW-Ipomo-F-1-174-150507 C-NMR 400



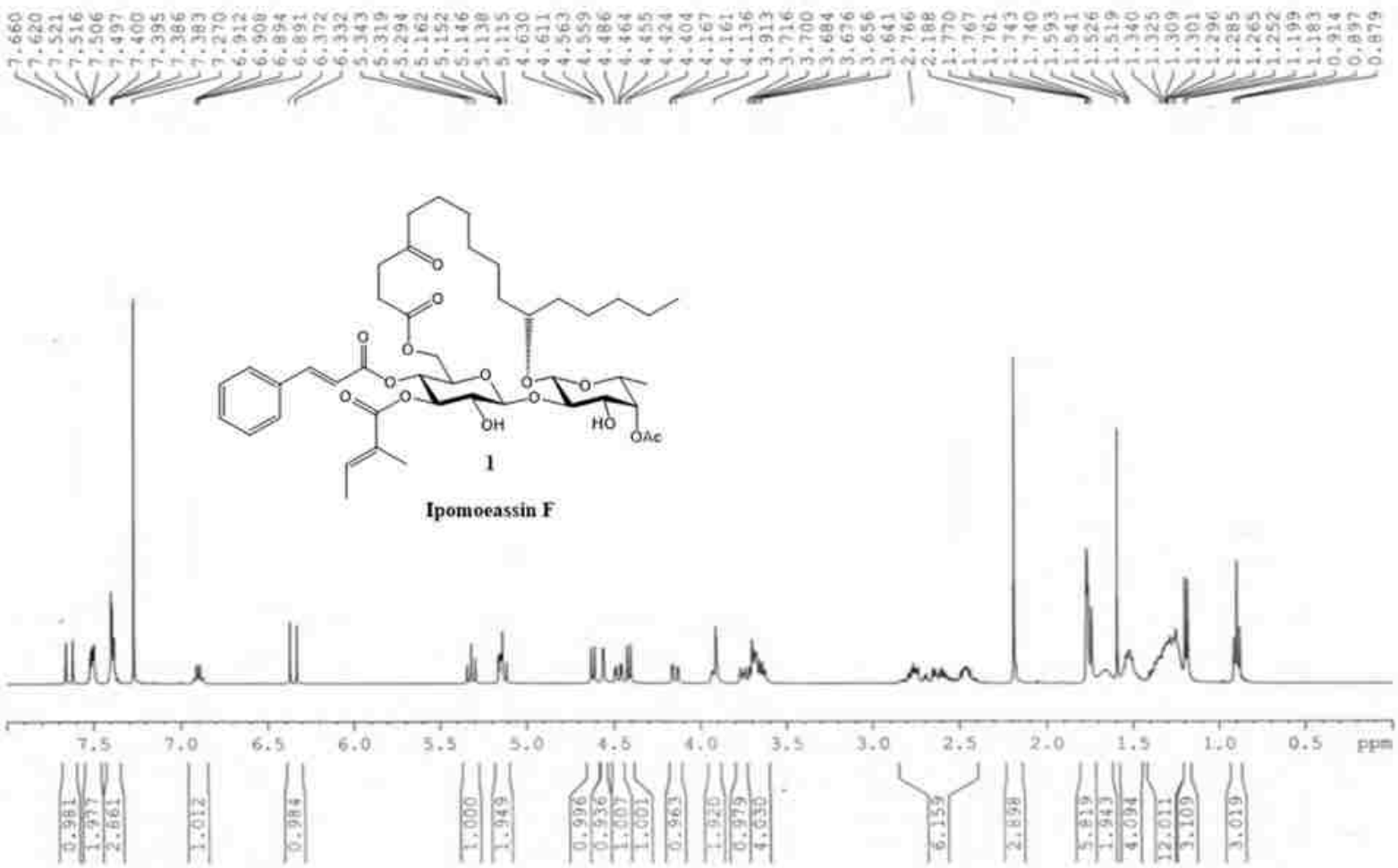

LW-Ipom-F-1-174-150707 (2) COSY
ppm
93
OH
HO
OAc
8
7
6
5
4
3
2
1
ppm


93
OH
HO
OAc
LW-Ipom-F-1-174-150707 (2) COSY
ppm
ppm

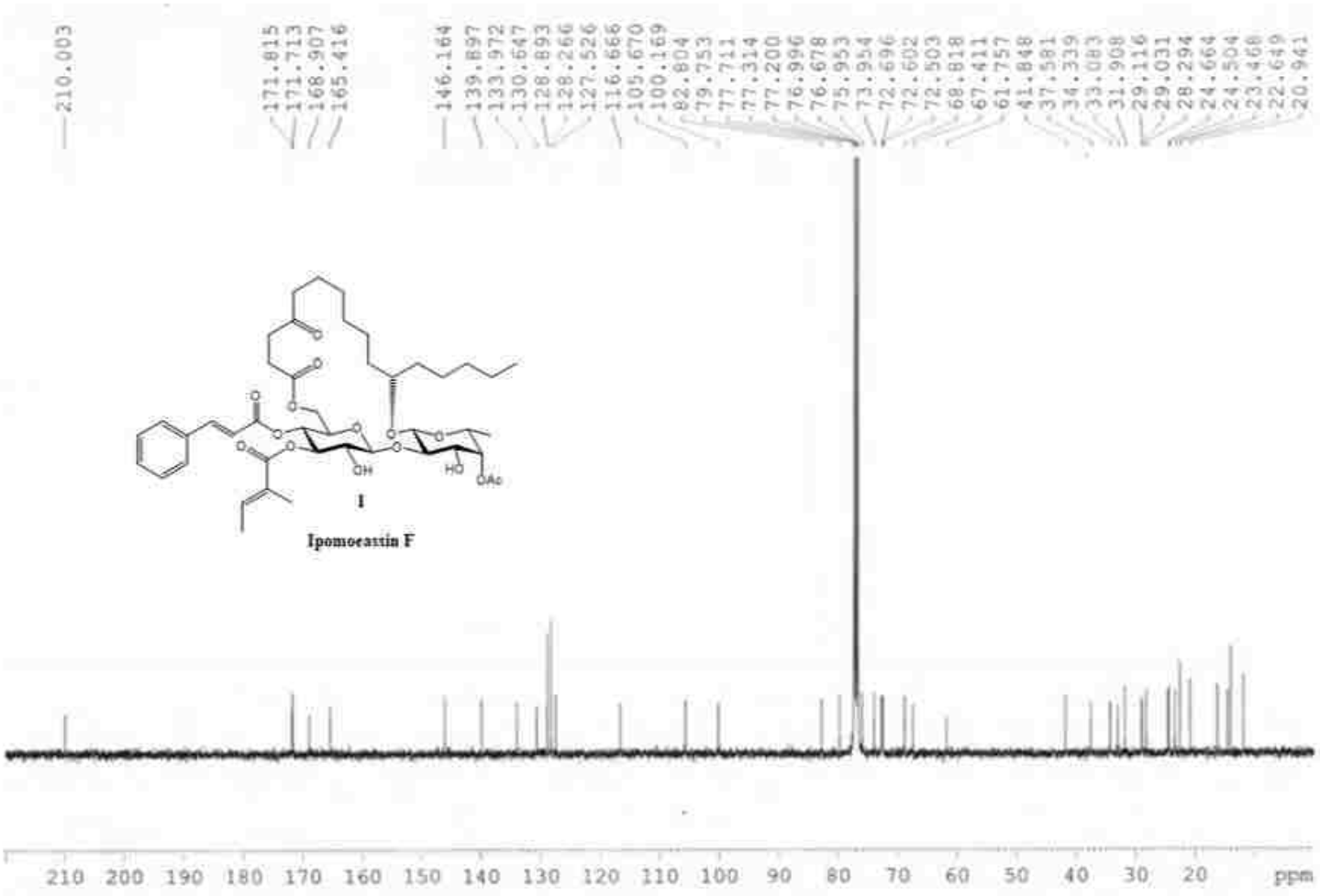

LW-Ipom-P-1-174-150707 (2) HSQC
ppm
20
30
40
50
60
70
80
90
100
110
120
130
140
8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 ppm
OH
HO
OAc
93

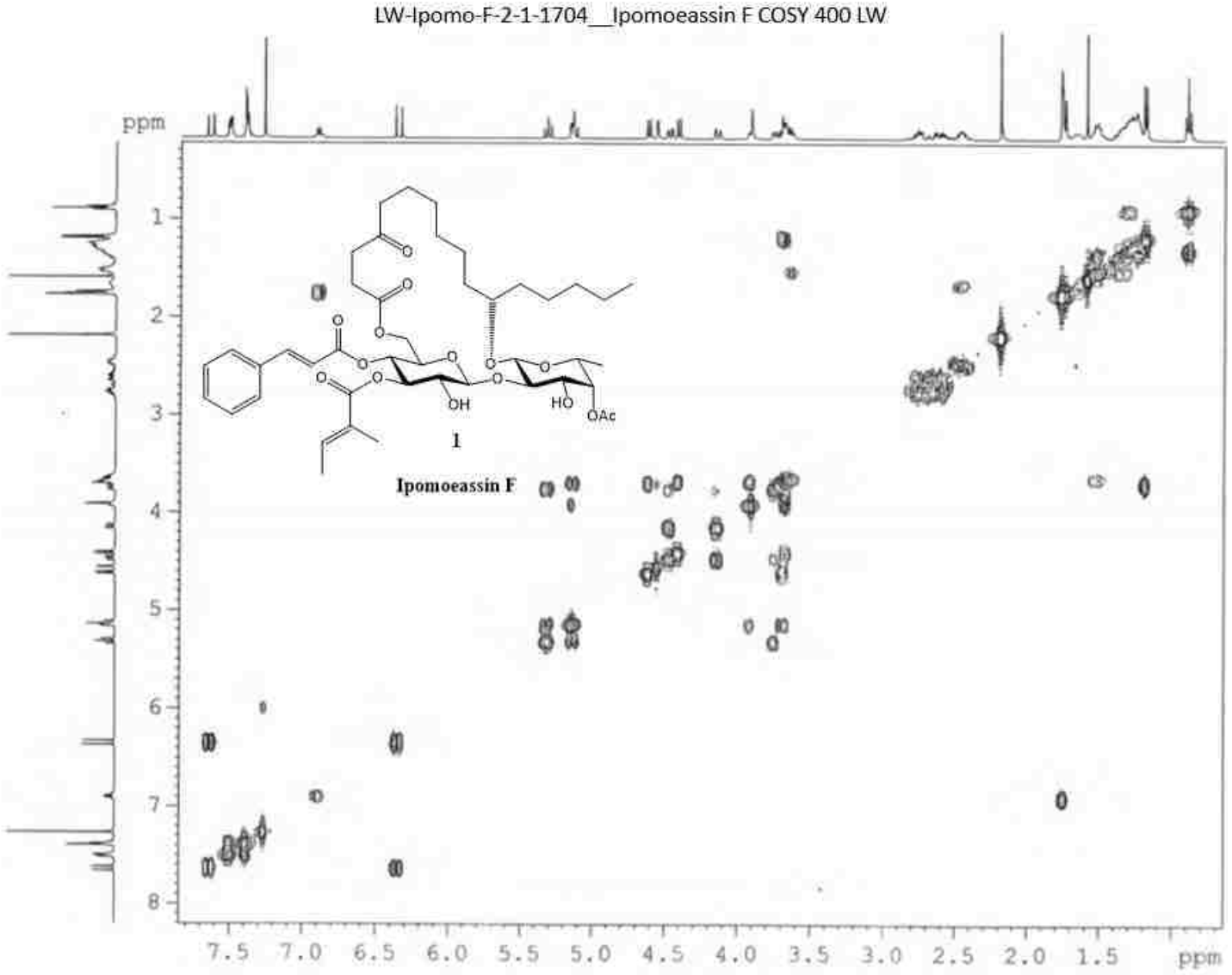

LW-Ipom-F-1-174-150707 (2) in CDCL3
ppm
166
167
168
169
170
171
172
173
174
2.9
2.8
2.7
2.6
2.5
2.4
2.3
2.2
ppm
OH
HO
OAc
93

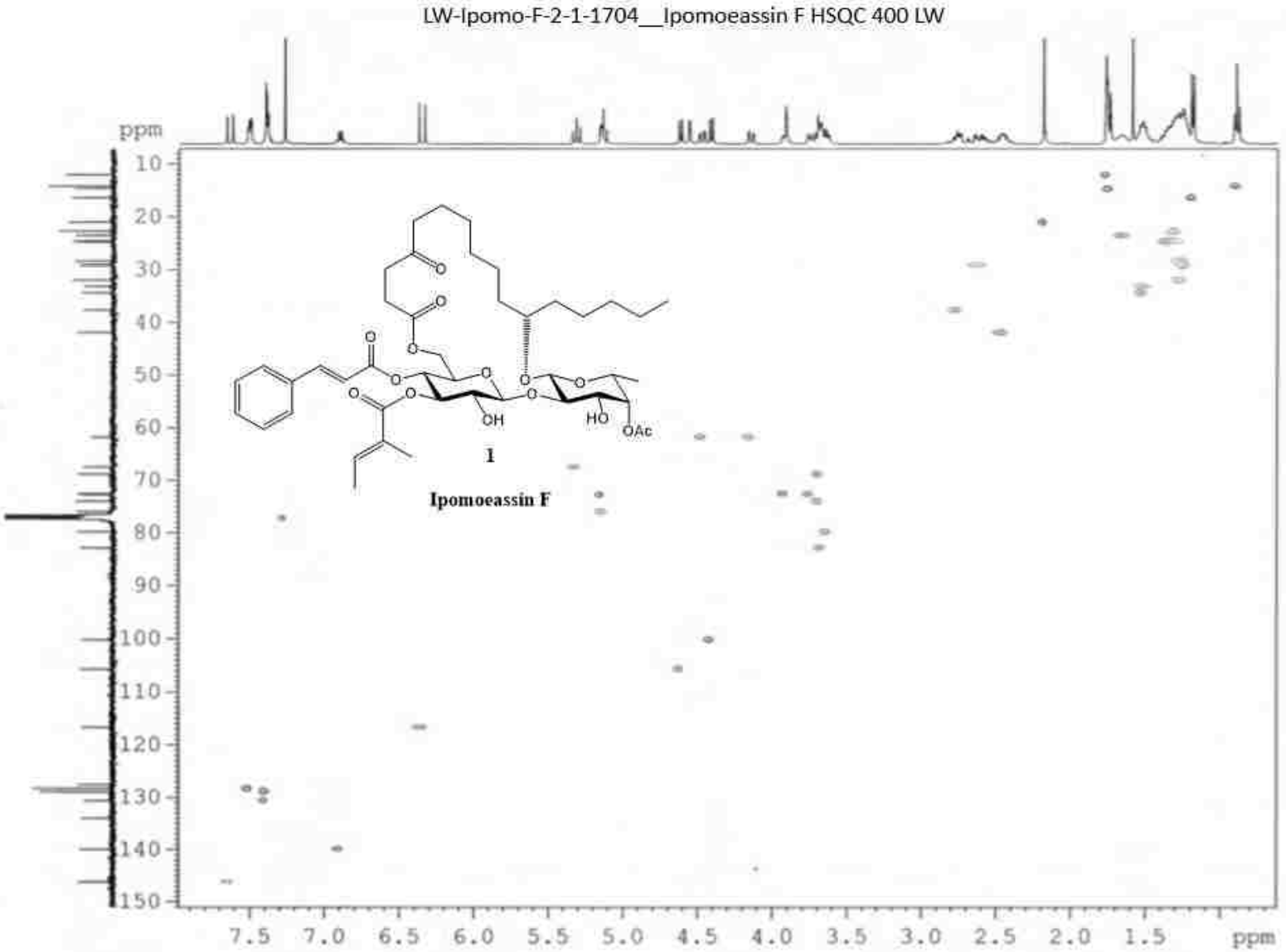
LW-Ipomo-F-2-1-1704___ H-NMR 400

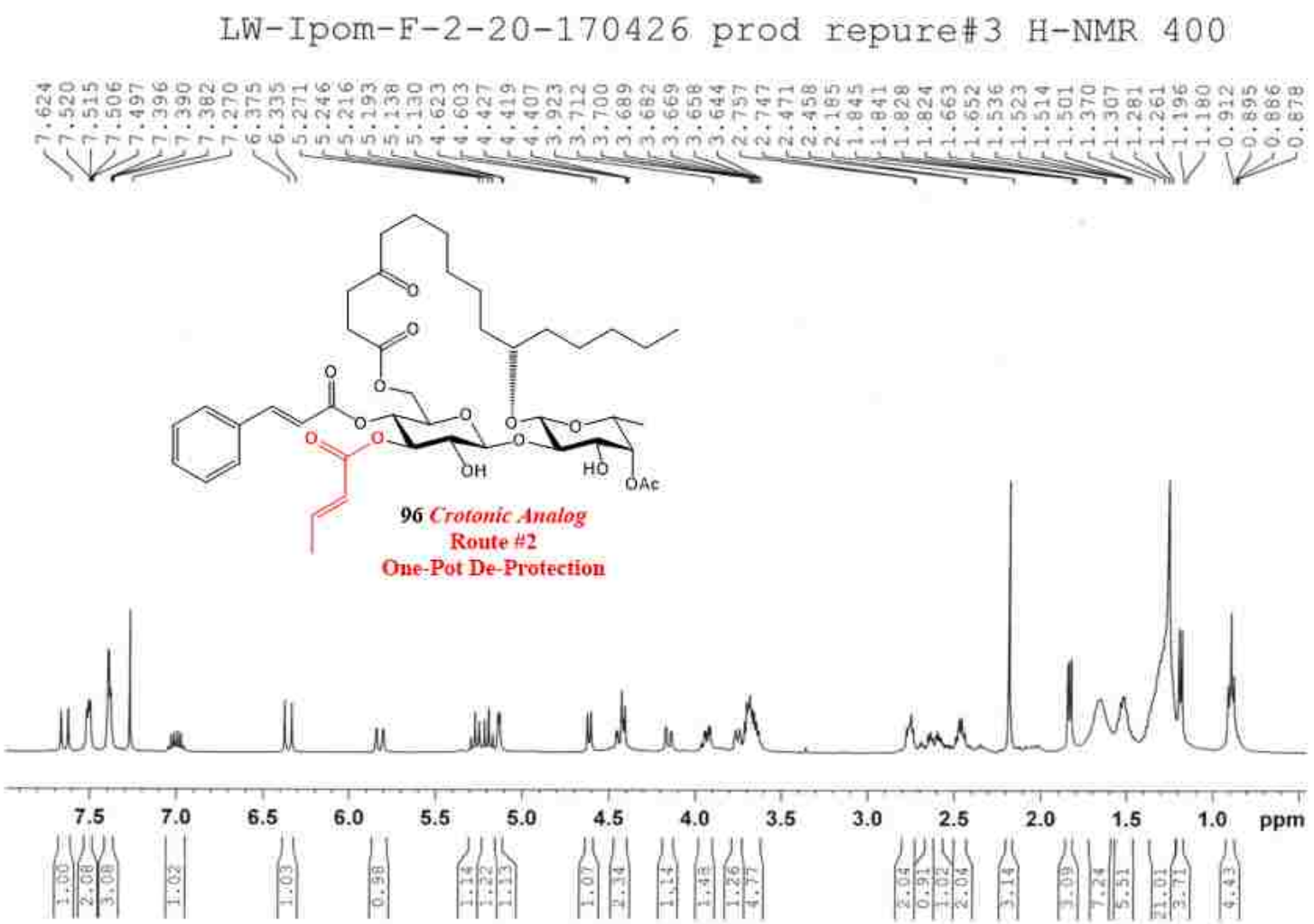


LW-Ipomo-F-2-1-1704___ C-NMR 400

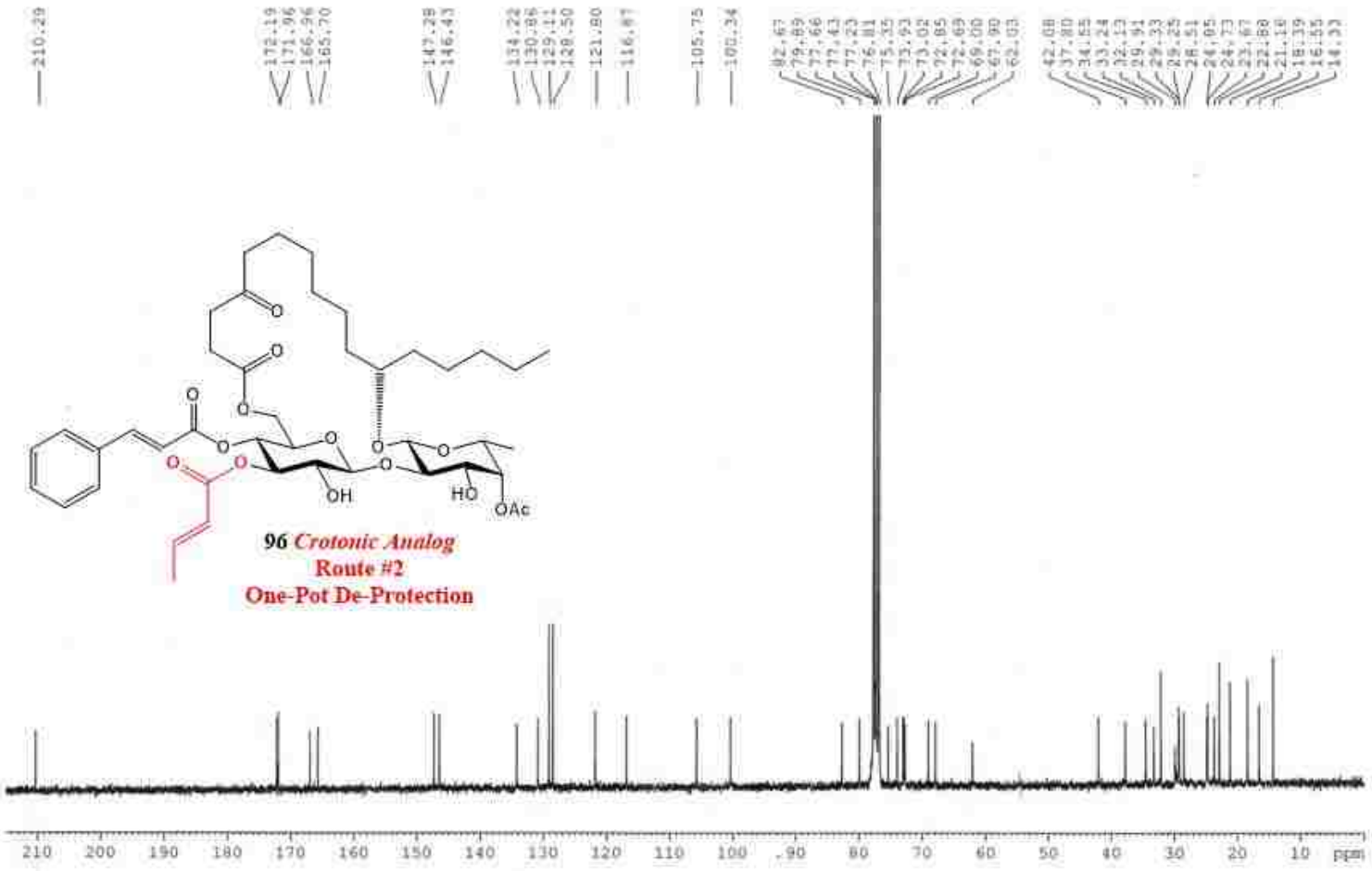



LW-Ipomo-F-2-1-1704__Ipomoeassin F COSY 400 LW
ppm
1
2
3
4
5
6
7
8
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
ppm
OH
HO
OAc
1
Ipomoeassin F

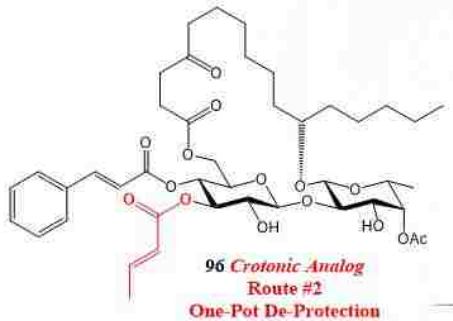

LW-Ipomo-F-2-1-1704__Ipomoeassin F HSQC 400 LW
ppm
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
ppm
OH
HO
OAc
1
Ipomoeassin F


LW-Ipom-F-2-20-170426 prod repure#3 H-NMR 400
7.624
7.520
7.515
7.506
7.497
7.396
7.390
7.382
7.270
6.375
6.335
5.271
5.246
5.216
5.193
5.138
5.130
4.623
4.603
4.427
4.419
4.407
3.923
3.712
3.700
3.689
3.682
3.669
3.658
3.644
2.757
2.747
2.471
2.458
2.185
1.845
1.841
1.828
1.824
1.663
1.652
1.536
1.523
1.514
1.501
1.370
1.307
1.281
1.261
1.196
1.180
0.912
0.895
0.886
0.878
OH
HO
OAc
96 Crotonic Analog
Route #2
One-Pot De-Protection
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm
1.00
2.08
3.08
1.02
1.03
0.98
1.14
1.22
1.13
1.07
2.34
1.14
1.48
1.26
4.77
2.04
0.91
1.02
2.04
3.14
3.09
7.24
5.51
21.01
3.71
4.43

LW-Ipom-F-20-20170426 repure#2 300 C13

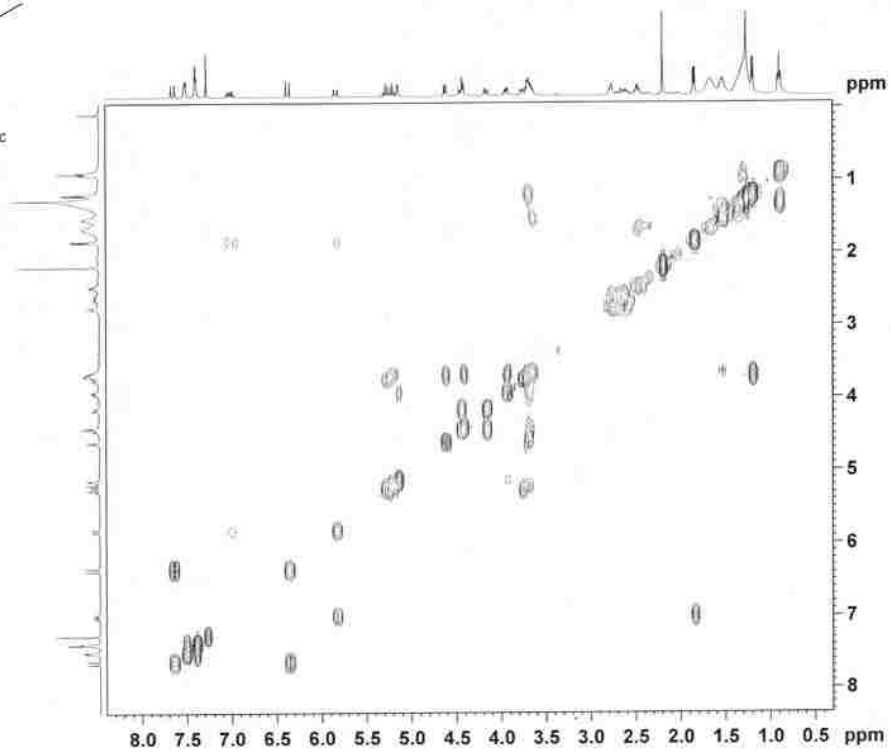


96 ***Crotonic Analog***
Route #2
One-Pot De-Protection

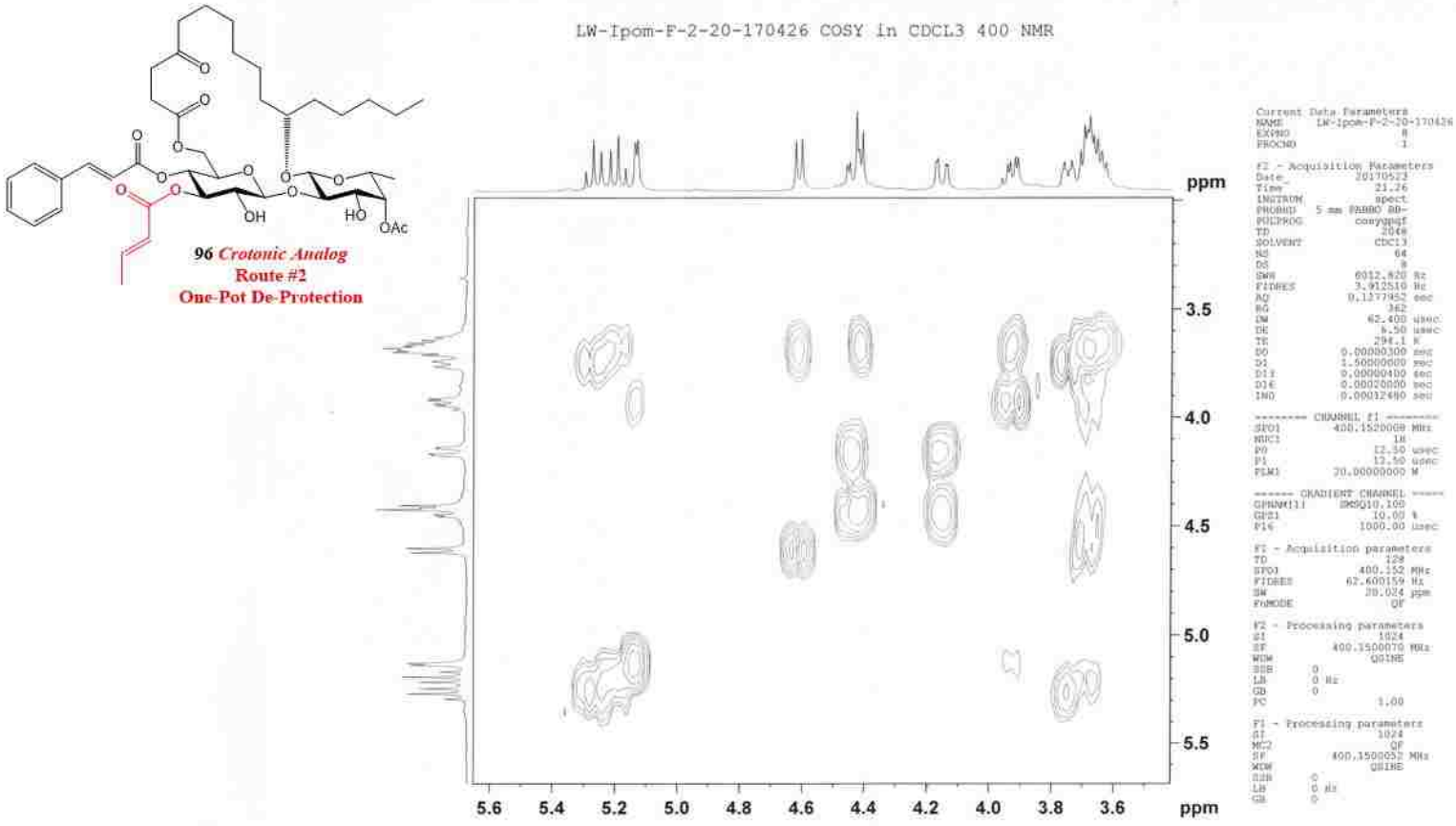


96 *Crotonic Analog*
Route #2
One-Pot De-Protection

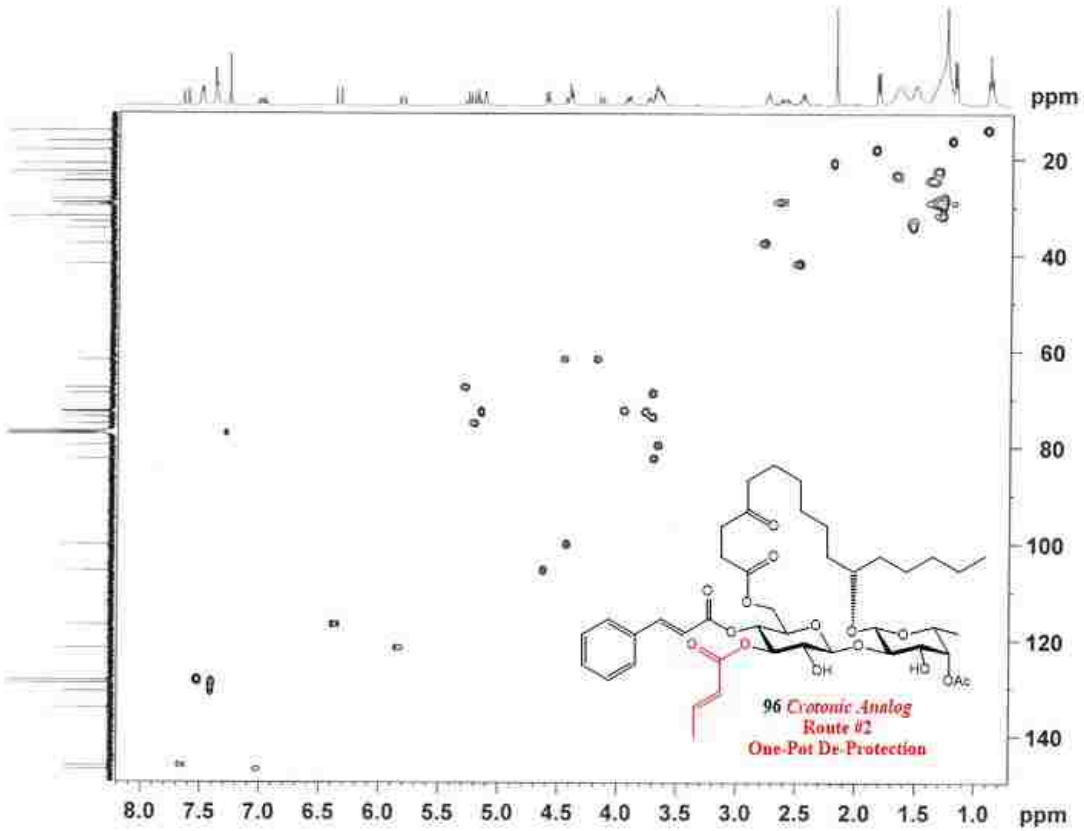
LW-Ipom-F-2-20-170426 COSY in CDCL3 400 NMR

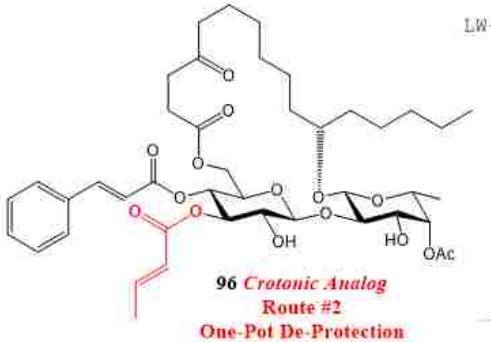



```
Current Data Parameters
NAME      LW-Ipom-F-2-20-170426
EXPNO                 6
PROCNO                1

F2 - Acquisition Parameters
Date_           20170523
Time              21.26
INSTRUM             spect
PROBHD   5 mm PABBO BB-
PULPROG          cosygpqf
TD                2048
SOLVENT           CDCl3
NS                  64
DS                   8
SWH            8012.820 Hz
FIDRES         3.910510 Hz
AQ             0.1277952 sec
RG                 362
DW                62.400 usec
DE                 6.50 usec
TE                294.1 K
D0             0.00000300 sec
D1             1.50000000 sec
D13            0.00000400 sec
D16            0.00020000 sec
IN0            0.00012480 sec

======== CHANNEL f1 ========
SFO1          400.1520000 MHz
NUC1                  1H
P0                 12.50 usec
P1                 12.50 usec
PLW1          20.00000000 W

====== GRADIENT CHANNEL =====
GPNAM[1]       SMSQ10.100
GPZ1              10.00 %
P16             1000.00 usec

F1 - Acquisition parameters
TD                 128
SFO1           400.152 MHz
FIDRES         62.600159 Hz
SW             20.024 ppm
FnMODE              QF

F2 - Processing parameters
SI                 1024
SF          400.1500070 MHz
WDW                QSINE
SSB         0
LB          0 Hz
GB          0
PC                 1.00

F1 - Processing parameters
SI                 1024
MC2                QF
SF          400.1500053 MHz
WDW                QSINE
SSB         0
LB          0 Hz
GB          0
```


LW-Ipom-F-2-20-170426 COSY in CDCL3 400 NMR
96 Crotonic Analog
Route #2
One-Pot De-Protection
OH
HO
OAc
ppm
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.5
4.0
4.5
5.0
5.5
Current Data Parameters
NAME LW-Ipom-F-2-20-170426
EXPNO 8
PROCNO 1
F2 - Acquisition Parameters
Date_ 20170523
Time 21.26
INSTRUM spect
PROBHD 5 mm PABBO BB-
PULPROG cosygpqf
TD 2048
SOLVENT CDCl3
NS 64
DS 8
SWH 6012.820 Hz
FIDRES 3.912510 Hz
AQ 0.1277952 sec
RG 362
DW 62.400 usec
DE 6.50 usec
TE 294.1 K
D0 0.00000300 sec
D1 1.50000000 sec
D13 0.00000400 sec
D16 0.00020000 sec
IN0 0.00012480 sec
======== CHANNEL f1 ========
SFO1 400.1520008 MHz
NUC1 1H
P0 12.50 usec
P1 12.50 usec
PLW1 20.00000000 W
====== GRADIENT CHANNEL =====
GPNAM[1] SMSQ10.100
GPZ1 10.00 %
P16 1000.00 usec
F1 - Acquisition parameters
TD 128
SFO1 400.152 MHz
FIDRES 62.400159 Hz
SW 20.024 ppm
FnMODE QF
F2 - Processing parameters
SI 1024
SF 400.1500070 MHz
WDW QSINE
SSB 0
LB 0 Hz
GB 0
PC 1.00
F1 - Processing parameters
SI 1024
MC2 QF
SF 400.1500052 MHz
WDW QSINE
SSB 0
LB 0 Hz
GB 0

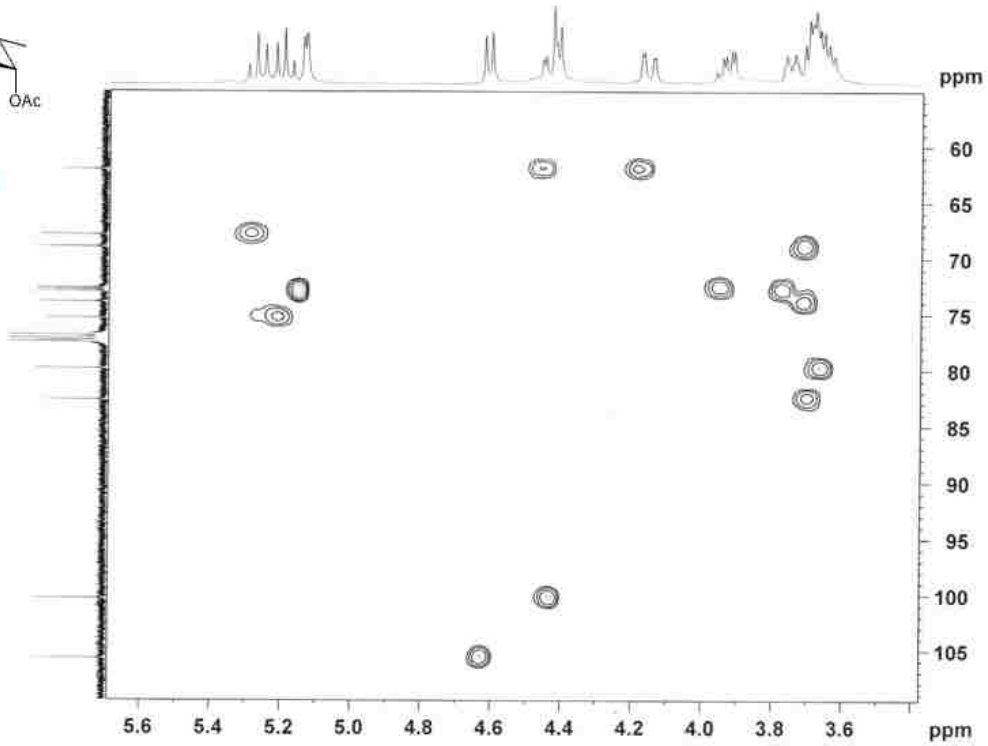
LW-Ipom-F-2-20-170426 HSQC in CDCl3 400 NMR



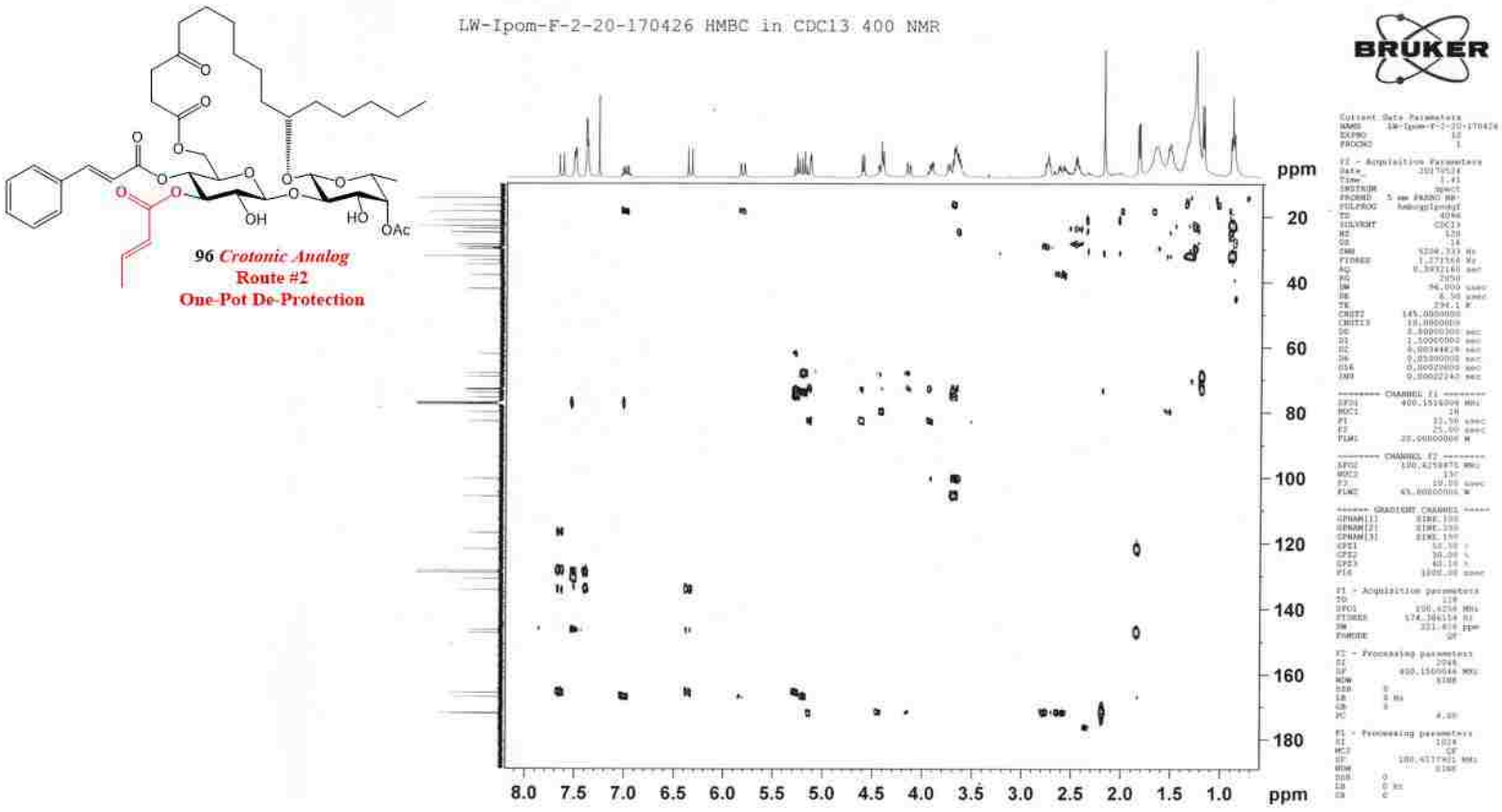


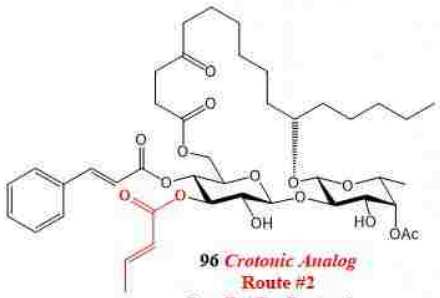
96 *Crotonic Analog*
Route #2
One-Pot De-Protection

LW-Ipom-F-2-20-170426 HSQC in CDCl3 400 NMR

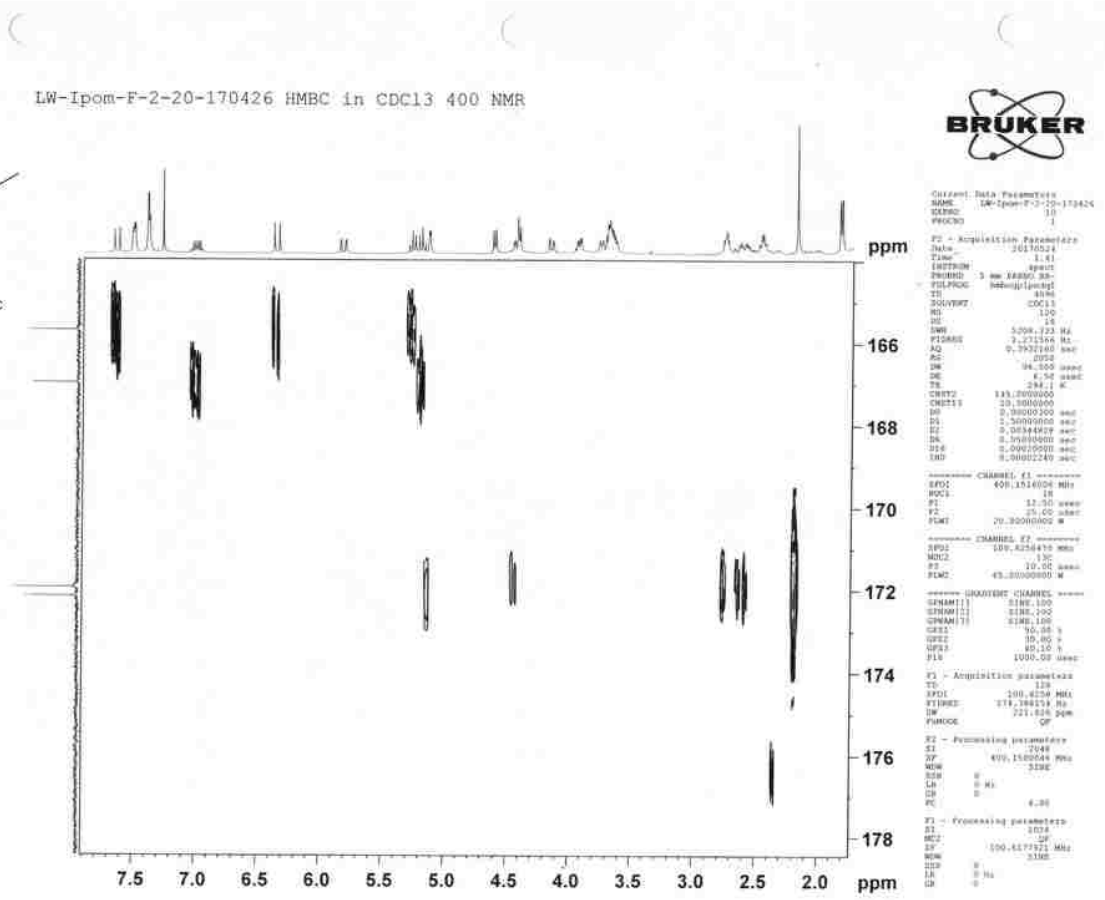


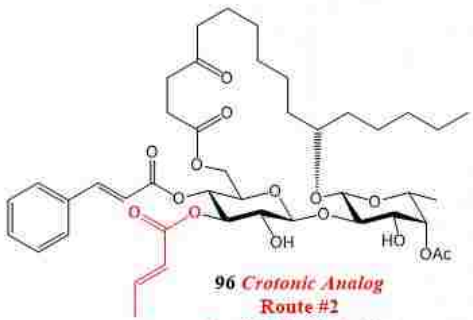
BRUKER


LW-Ipom-F-2-20-170426 HMBC in CDCl3 400 NMR
BRUKER
OH
HO
OAc
96 Crotonic Analog
Route #2
One-Pot De-Protection
ppm
8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 ppm
20 40 60 80 100 120 140 160 180

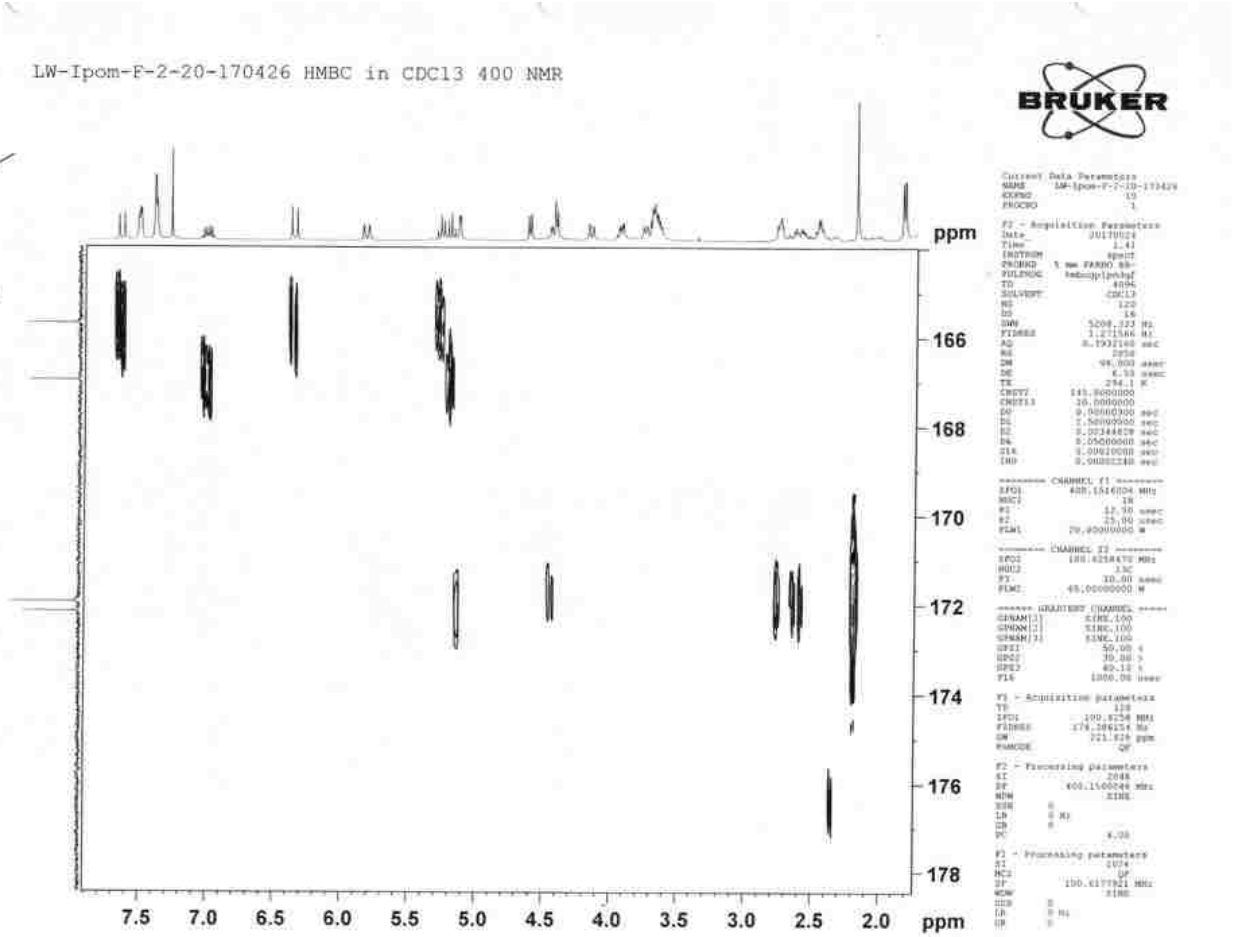


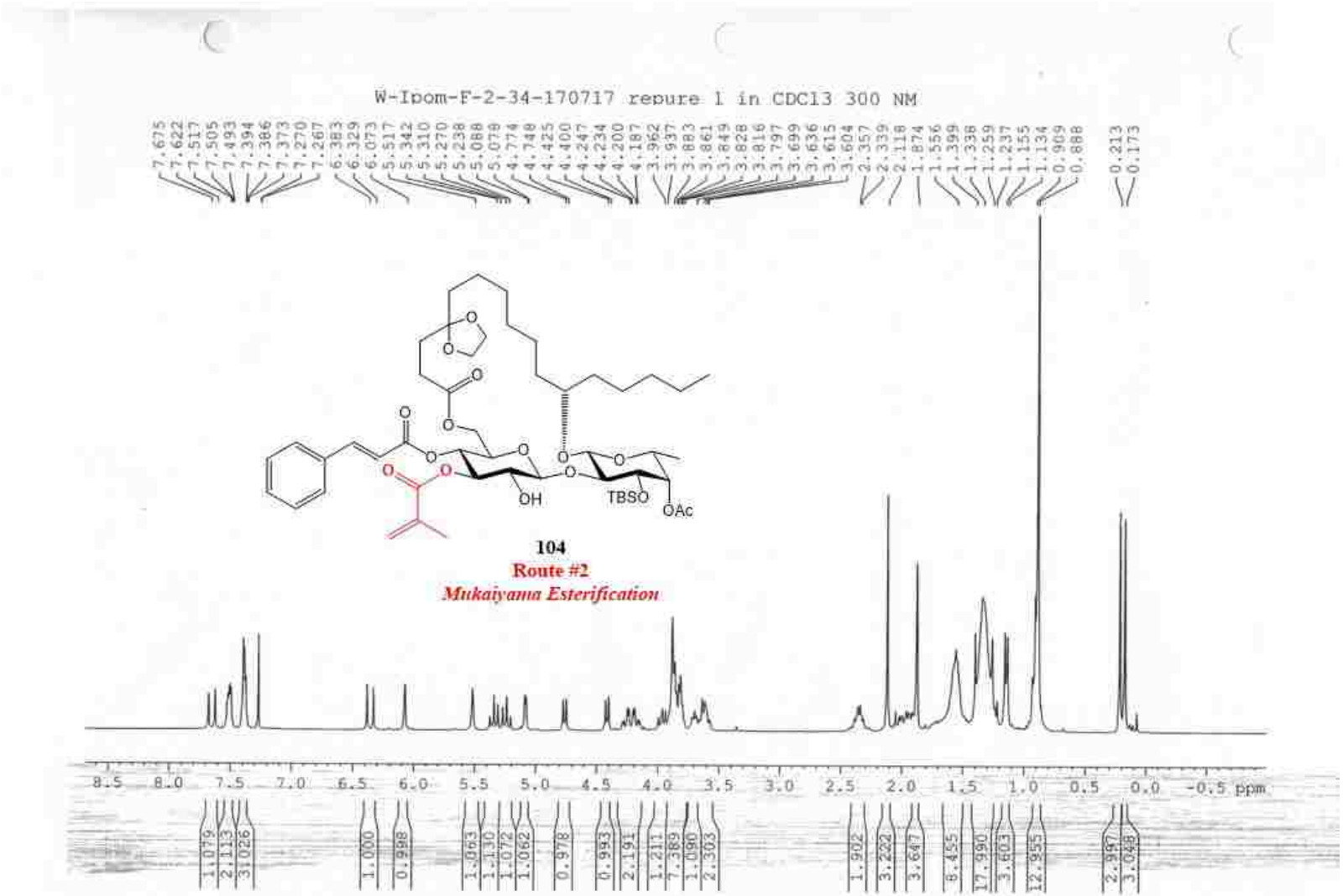
96 ***Crotonic Analog***
Route #2
One-Pot De-Protection

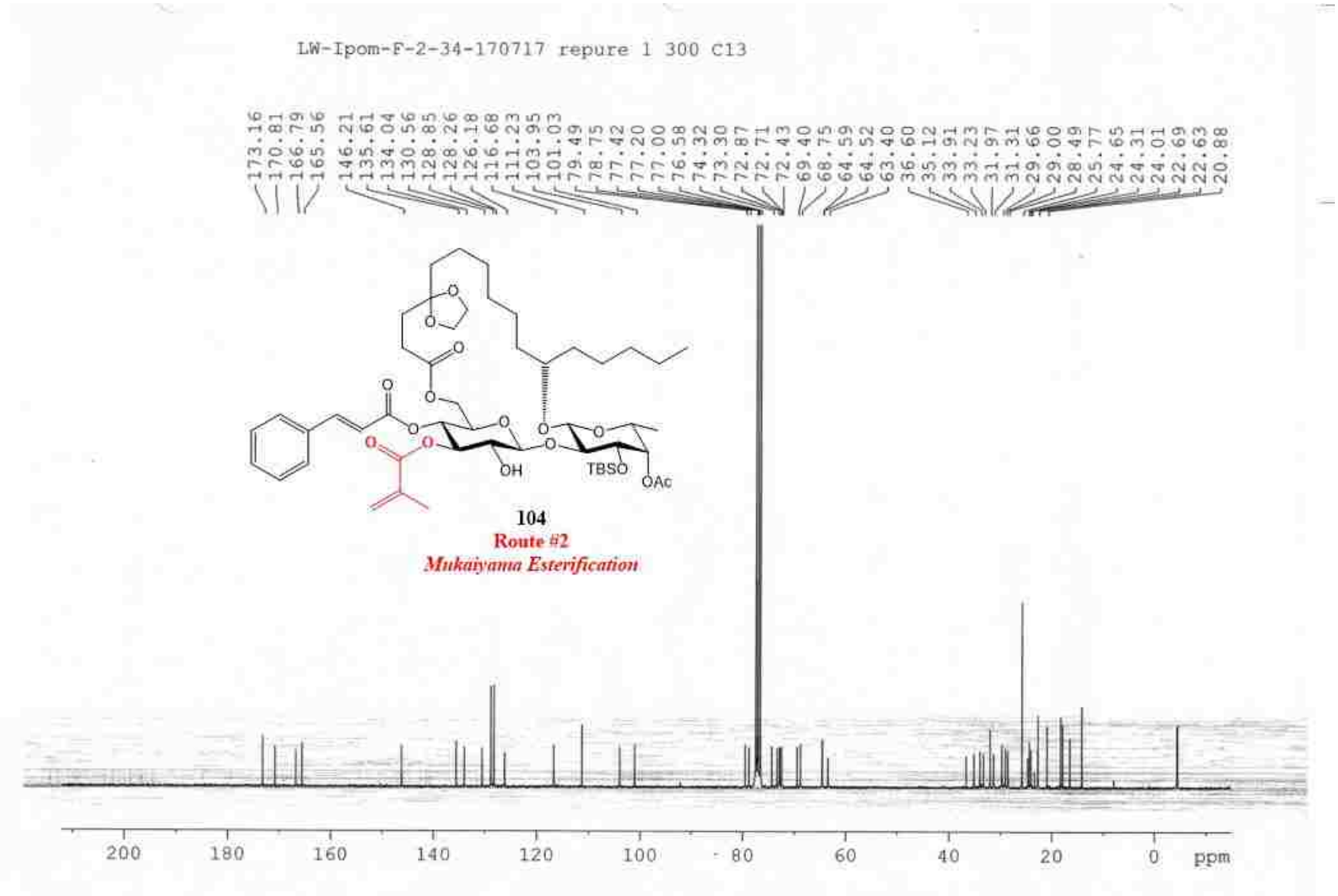




96 ***Crotonic Analog***
Route #2
One-Pot De-Protection



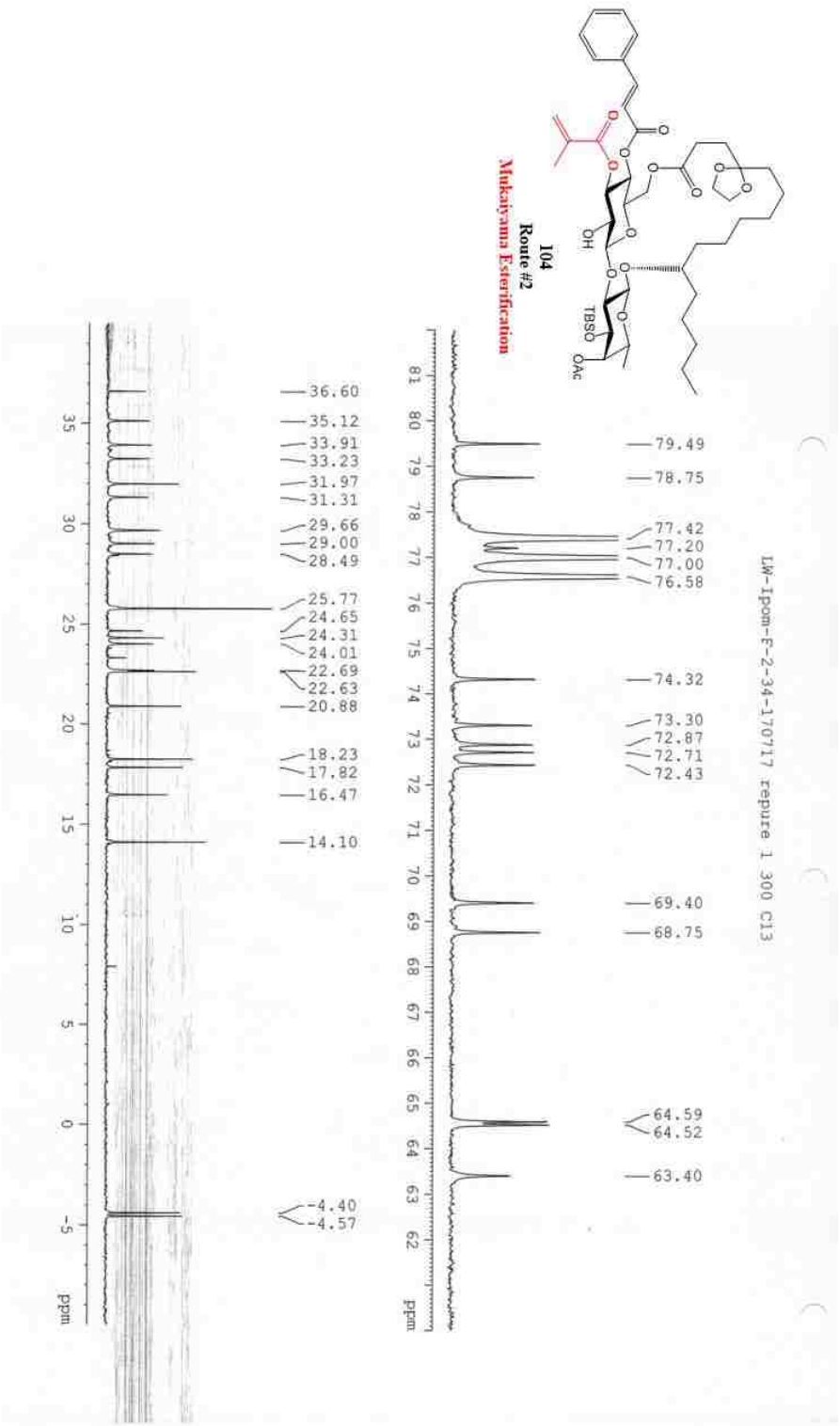

W-Ipom-F-2-34-170717 repure 1 in CDCl3 300 NM
OH
TBSO
OAc
104
Route #2
Mukaiyama Esterification

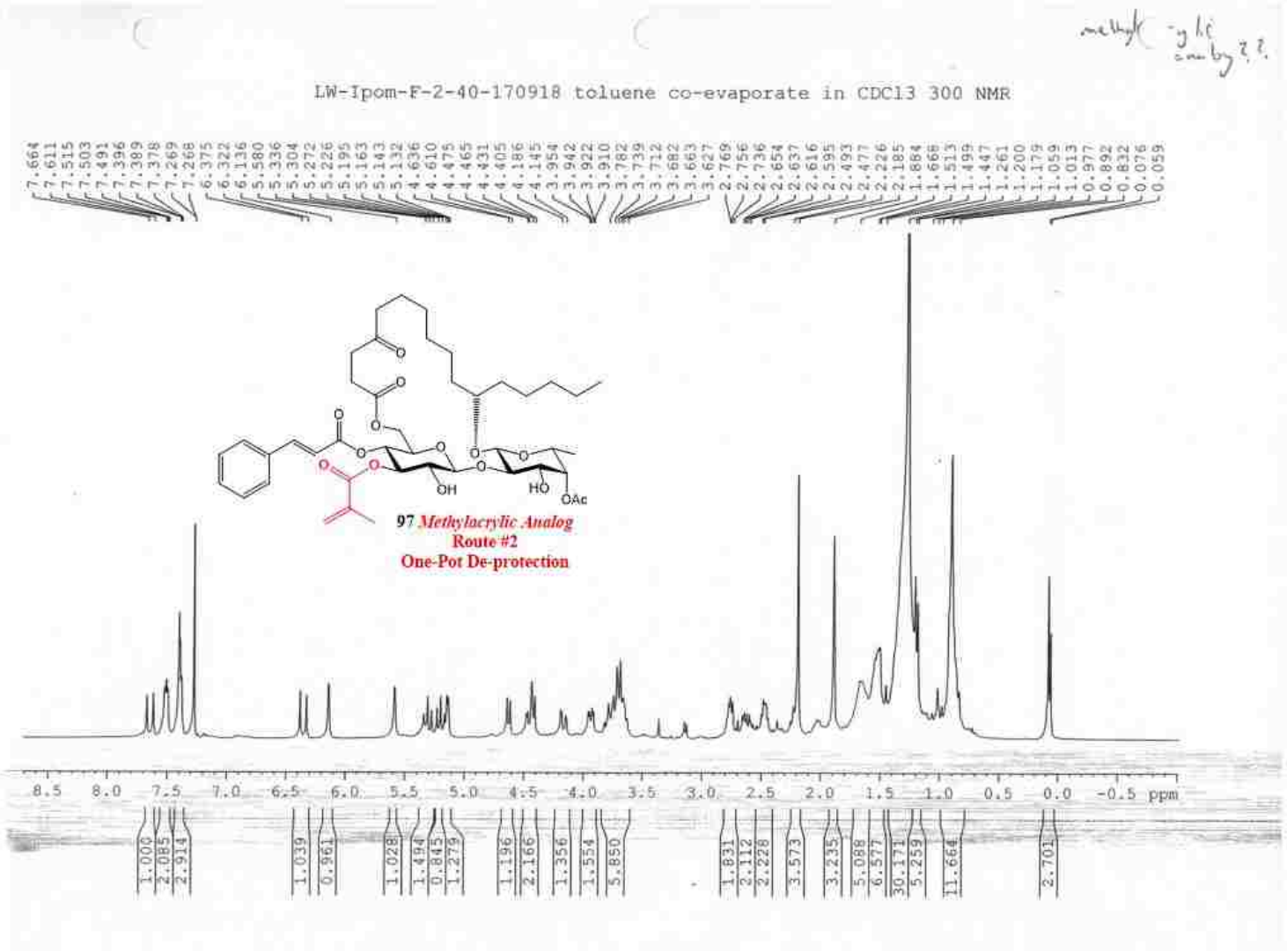


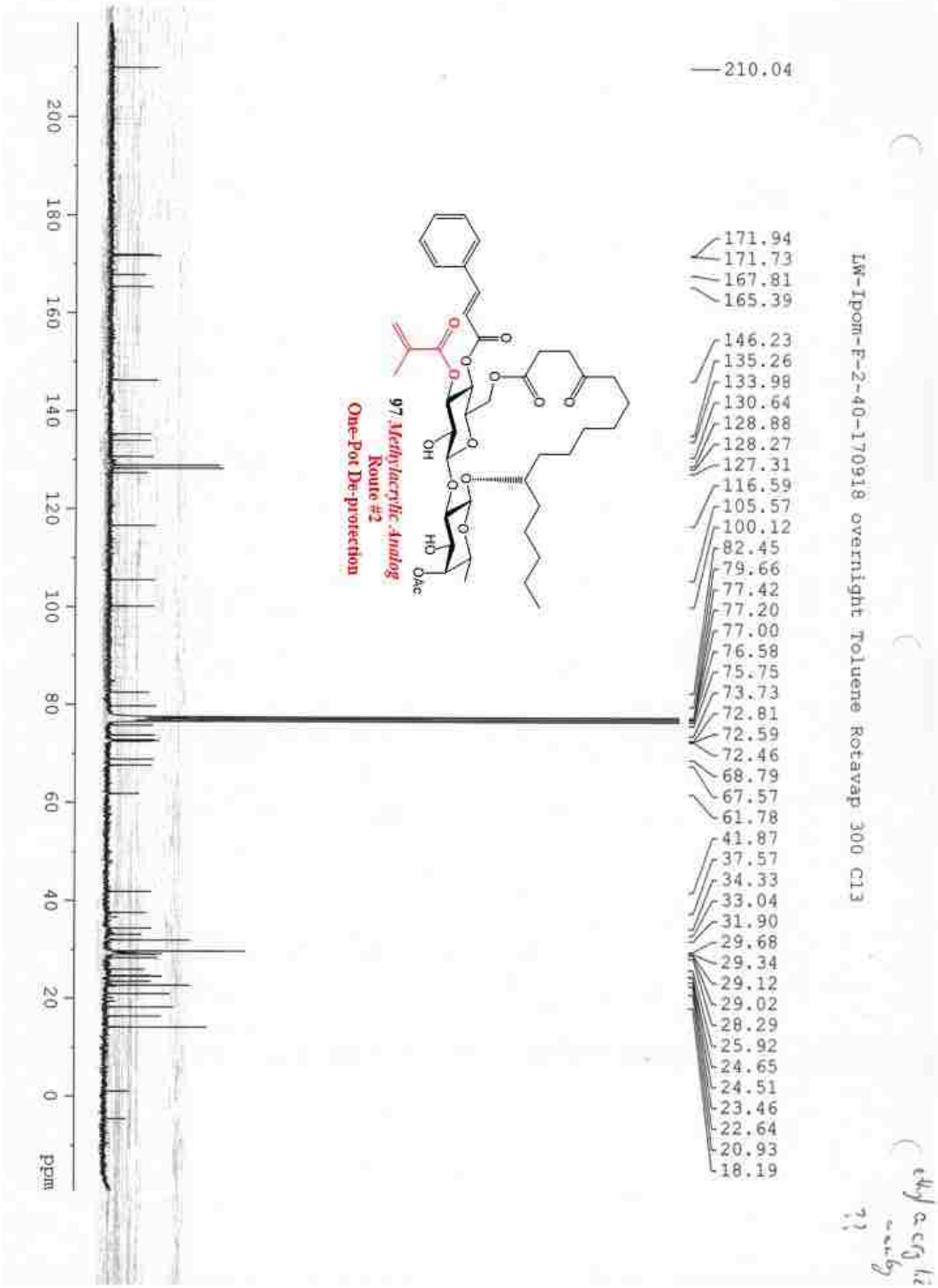
104

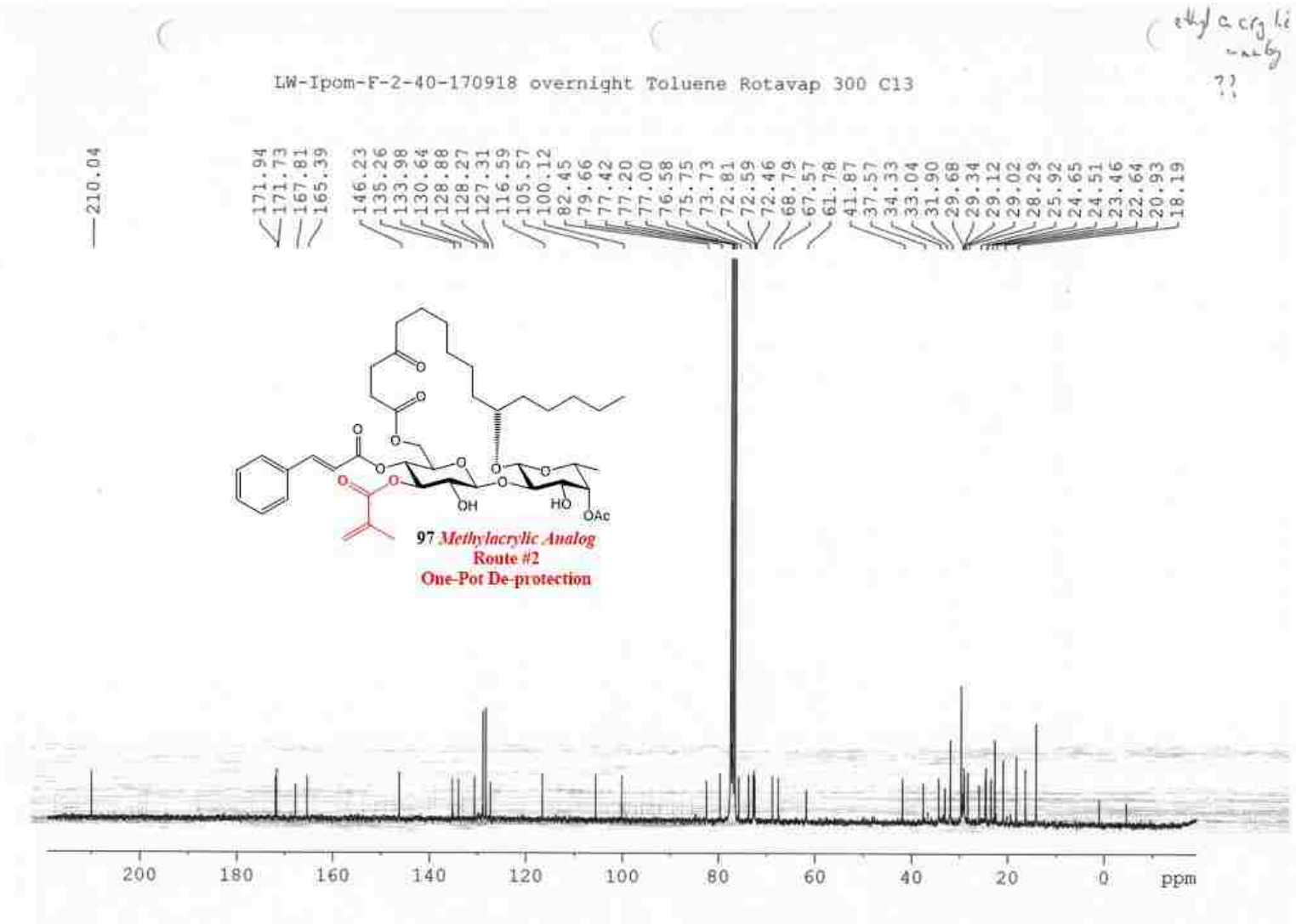
Route #2

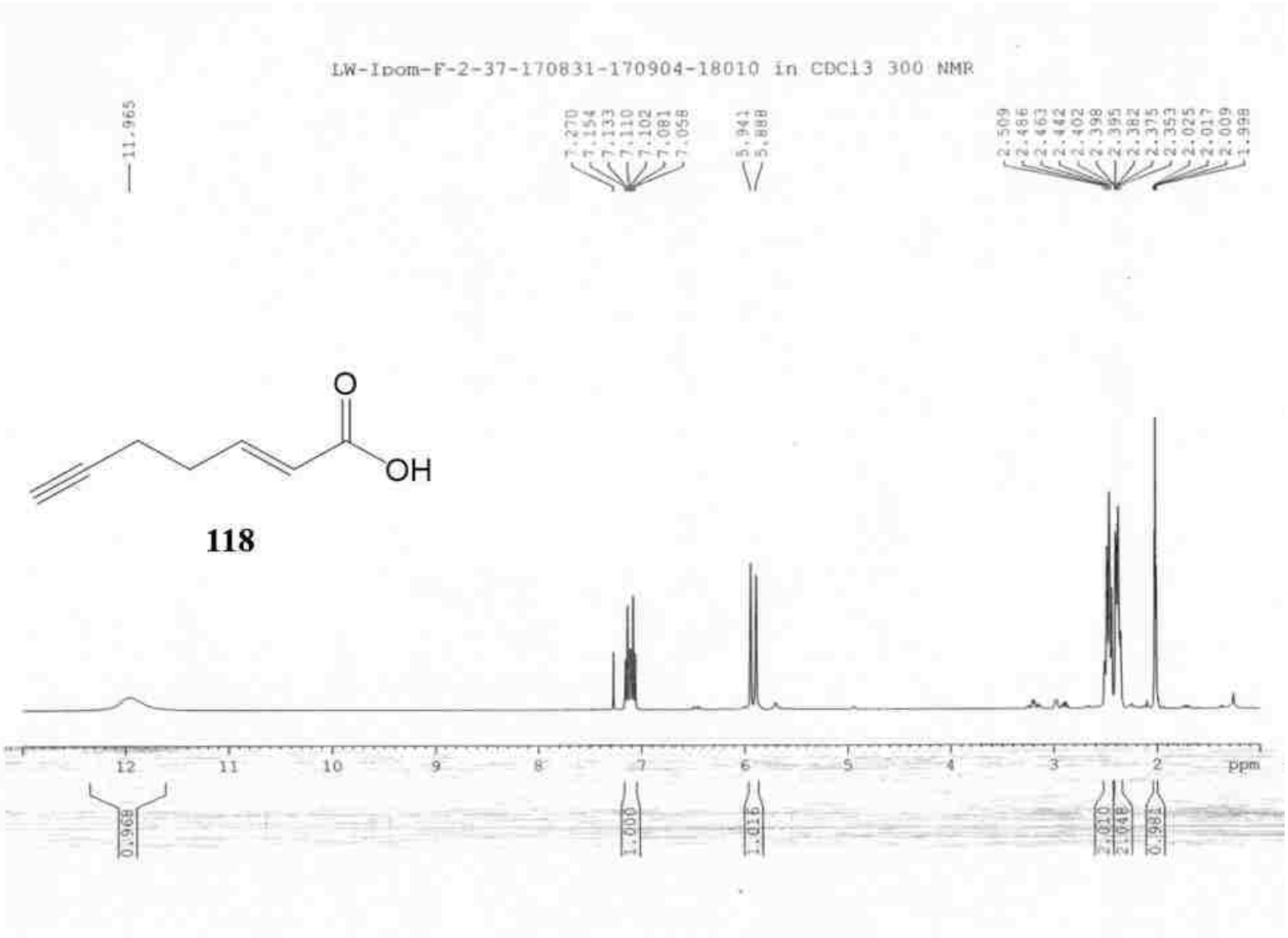
Mukaiyama Esterification

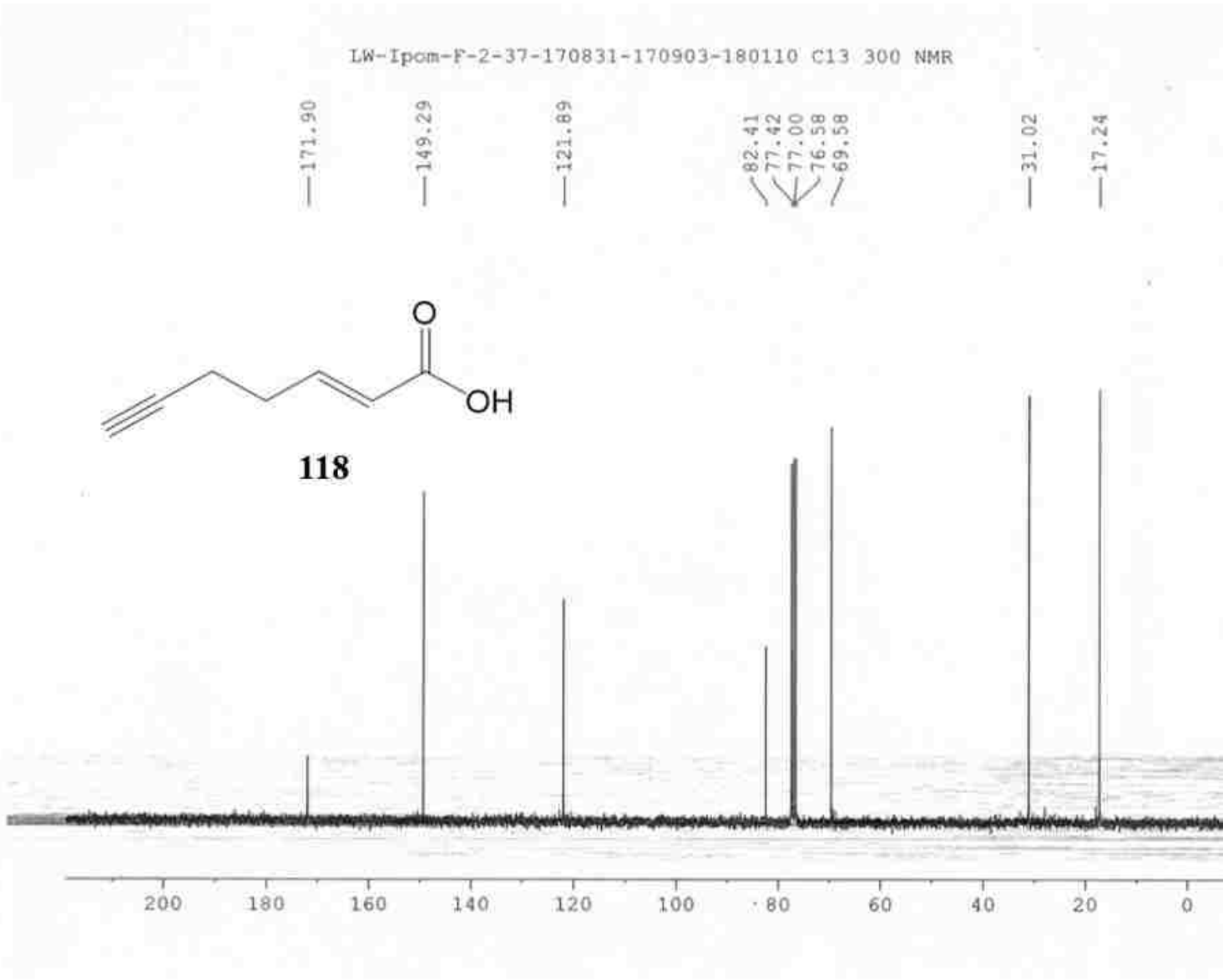

104
Route #2
Mukaiyama Esterification
LW-Ipom-F-2-34-170717 repure 1 300 C13
79.49
78.75
77.42
77.20
77.00
76.58
74.32
73.30
72.87
72.71
72.43
69.40
68.75
64.59
64.52
63.40
36.60
35.12
33.91
33.23
31.97
31.31
29.66
29.00
28.49
25.77
24.65
24.31
24.01
22.69
22.63
20.88
18.23
17.82
16.47
14.10
-4.40
-4.57
ppm


LW-Ipom-F-2-40-170918 toluene co-evaporate in CDCl3 300 NMR
97 Methylacrylic Analog
Route #2
One-Pot De-protection
OH
HO
OAc
ppm


LW-Ipom-F-2-40-170918 overnight Toluene Rotavap 300 C13
210.04
171.94
171.73
167.81
165.39
146.23
135.26
133.98
130.64
128.88
128.27
127.31
116.59
105.57
100.12
82.45
79.66
77.42
77.20
77.00
76.58
75.75
73.73
72.81
72.59
72.46
68.79
67.57
61.78
41.87
37.57
34.33
33.04
31.90
29.68
29.34
29.12
29.02
28.29
25.92
24.65
24.51
23.46
22.64
20.93
18.19
97 Methylacrylic Analog
Route #2
One-Pot De-protection
OH
HO
OAc
200
180
160
140
120
100
80
60
40
20
0
ppm


LW-Ipom-F-2-40-170918 overnight Toluene Rotavap 300 C13
210.04
171.94
171.73
167.81
165.39
146.23
135.26
133.98
130.64
128.88
128.27
127.31
116.59
105.57
100.12
82.45
79.66
77.42
77.20
77.00
76.58
75.75
73.73
72.81
72.59
72.46
68.79
67.57
61.78
41.87
37.57
34.33
33.04
31.90
29.68
29.34
29.12
29.02
28.29
25.92
24.65
24.51
23.46
22.64
20.93
18.19
OH
HO
OAc
97 Methylacrylic Analog
Route #2
One-Pot De-protection
200
180
160
140
120
100
80
60
40
20
0
ppm


LW-Ipom-F-2-37-170831-170904-18010 in CDCl3 300 NMR
11.965
7.270
7.154
7.133
7.110
7.102
7.081
7.058
5.941
5.888
2.509
2.486
2.463
2.442
2.402
2.398
2.395
2.382
2.375
2.353
2.025
2.017
2.009
1.998
O
OH
118
12
11
10
9
8
7
6
5
4
3
2
ppm
0.968
1.000
1.016
2.010
2.048
0.981


LW-Ipom-F-2-37-170831-170903-180110 C13 300 NMR
171.90
149.29
121.89
82.41
77.42
77.00
76.58
69.58
31.02
17.24
O
OH
200
180
160
140
120
100
80
60
40
20
0

118