University of Groningen

Total Synthesis of Enantiopure Biomolecules

van Summeren, Ruben Petrus

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2006

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):
van Summeren, R. P. (2006). *Total Synthesis of Enantiopure Biomolecules: on mycolactones, saturated isoprenoid building blocks and ß-mannosyl phosphomycoketides*. [Thesis fully internal (DIV), University of Groningen]. s.n.

Copyright
Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

The publication may also be distributed here under the terms of Article 25fa of the Dutch Copyright Act, indicated by the "Taverne" license. More information can be found on the University of Groningen website: https://www.rug.nl/library/open-access/self-archiving-pure/taverne-amendment.

Take-down policy
If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): http://www.rug.nl/research/portal. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.

Download date: 24-01-2026

Total Synthesis of Enantiopure Biomolecules

On mycolactones, saturated isoprenoid building blocks
and β-mannosyl phosphomycoketides

Ruben Petrus van Summeren

The work described in this thesis was carried out at the Department of Organic and Molecular Inorganic Chemistry, Stratingh Institute, University of Groningen, The Netherlands.

The work described in this thesis was financially supported by the Dutch Organization for Scientific Research (NWO).

RIJKSUNIVERSITEIT GRONINGEN

Total Synthesis of Enantiopure Biomolecules

On mycolactones, saturated isoprenoid building blocks
and β-mannosyl phosphomycoketides

Proefschrift

ter verkrijging van het doctoraat in de
Wiskunde en Natuurwetenschappen
aan de Rijksuniversiteit Groningen
op gezag van de
Rector Magnificus, dr. F. Zwarts,
in het openbaar te verdedigen op
maandag 27 maart 2006
om 16.15 uur

door

Ruben Petrus van Summeren

geboren op 10 november 1977
te Veghel

Promotores :	Prof. Dr. B.L. Feringa
	Prof. Dr. Ir. A. J. Minnaard
Beoordelingscommissie:	Prof. Dr. J.B.F.N. Engberts
	Prof. Dr. J.G. de Vries
	Prof. Dr. H.S. Overkleeft

ISBN: 90-367-2545-3

Voorwoord

Dat was het dan. De vier jaar van mijn promotieonderzoek zijn werkelijk voorbij gevlogen. En….? Wat heb je nou geleerd? Veel als chemicus, maar nog meer als persoon. Dat heeft natuurlijk alles te maken met de mensen om je heen en die wil ik op deze plaats dan ook bedanken.

Allereerst wil ik mijn promotores prof. dr. B. L. Feringa en prof. dr. A. J. Minnaard bedanken voor de gelegenheid om mijn onderzoek uit te voeren. Ben, jouw enthousiasme voor chemie is inmiddels bijna spreekwoordelijk. Adri, ik ben vereerd om jouw eerste promovendus te mogen zijn. Jij stond altijd klaar met advies en zonodig met bemoedigende woorden en dat was van groot belang met name in de eerste twee jaar. Daarnaast toonde je ook interesse in mij als persoon, bijvoorbeeld toen ik aan mijn knie geopereerd werd. Niets veranderen, al zal dat moeilijk zijn nu je een steeds grotere groep krijgt en bovendien binnenkort naar Duitsland gaat. Succes!

De leden van de beoordelingscommissie prof. dr. J. B. F. N. Engberts, prof. dr. J. G. de Vries en prof. dr. H. S. Overkleeft wil ik bedanken voor de snelle correctie van het manuscript. MD T. S. van der Werf en collegae ben ik dankbaar voor de prettige samenwerking met betrekking tot het Buruli ulcer project. Furthermore, I would like to express my gratitude to Dr. Moody and co-workers in Boston for the successful collaboration on the β-mannosyl phosphomycoketide project.

Pieter Bos ben ik veel dank verschuldigd voor zijn werk als bijvak- en later als hoofdvakstudent. Een gedeelte van dit werk is opgenomen in hoofdstuk 3 van dit proefschrift. Zijn huidige project hoop ik binnenkort te begroeten in artikelvorm. Pieter, je hebt aan den lijve ondervonden dat totaalsynthese niet de makkelijkste weg is naar snelle resultaten. Je hebt echter veel vaardigheden opgedaan als organisch syntheticus en ik heb dan ook alle vertrouwen in de promotie die je hierna wilt gaan doen.

Daarnaast hebben mijn labgenoten: Martin Klopstra, Pedro Pinho, Diego Peña, Richard Jagt, Francesca Gini en Fernando López enorm bijgedragen aan een aangename werkomgeving. Nothing beats a Spanish postdoc as a neighbour. Especially when they also provide all arranged holidays to Galicia. Francesca, dit is mijn laatste poging als ambassadeur van de Nederlandse taal. Ik vond het leuk om met je samen te werken. Je bent,

één van de meest sociale personen die ik ken. Die keren dat je niet zeker wist of ik serieus was: het was een grapje. Ik draag hierbij de radio aan jou over. Richard, jij bent me d'r eentje. Op veel vlakken zijn wij absolute tegenpolen, maar dat heeft het eigenlijk alleen maar interessanter gemaakt. Veel succes met je verdere ondernemingen op mikado-gebied.

Verder zijn er natuurlijk een groot aantal mensen die voor gezelligheid zorgden bij diverse gelegenheden zoals lunches, borrels, etentjes, voetbalwedstrijden, vakanties en wat al niet meer. In willekeurige vollegorde: Hans, Niek, Jaap, Joost, Wesley, Michel, Tieme, Bas, Chris, Rob, Bjorn, Heiko, Lavinia, Suzy, Gabriella, Javier, Edzard en iedereen die ik nog vergeet…bedankt.

De mensen van de subgroep asymmetrische katalyse ben ik dankbaar voor hun suggesties en voor hun geduld wanneer ik het weer eens niet over asymmetrische katalyse had. Theodora Tiemersma-Wegman ben ik veel dank verschuldigd voor hulp bij de analyse van chirale verbindingen. Wim Kruizinga wil ik bedanken voor de NMR-metingen die hij voor mij heeft verricht en Albert Kiewiet voor het bepalen van een enorm aantal massa's.

Een speciaal woord van waardering wil ik richten tot mijn beste vrienden: Erik, Leon, Maarten en Ruud. Samen doorlopen wij nu al ruim 15 jaar de universiteit van het leven. Over oude verhalen en nieuwe bespiegelingen. De discussies, de hilariteit, het geneuzel en het gerommel in de marge. Alles heeft zijn functie. Wanneer is het volgende avondje?

Mijn familie wil ik bedanken voor hun begrip. Groningen? Kan dat niet in Nijmegen dan? Bedankt voor jullie geduld en voor jullie interesse in wat ik doe. Anne, bedankt voor het ontwerpen van de voorkant van o.a. dit boekje.

Philana, jij brengt het beste in mij boven en alleen bij jou kan ik zijn op mijn slechtste momenten. Dank je wel.

Contents

Chapter 1
Introduction

1.1 Organic synthesis

Organic synthesis entails the assembly of complex molecules (usually containing the elements: C, H, O, N, S, P, B and halogens) from relatively simple starting materials and reagents through the formation and breaking of covalent bonds. Traditionally, organic synthesis is divided into method-oriented synthesis and target-oriented synthesis, which is more commonly referred to as total synthesis (Figure 1.1).[1] The central theme of method-oriented synthesis is the invention, discovery and development of new synthetic reactions, reagents and catalysts.[1] In contrast, the ultimate goal of total synthesis is the construction of a defined target molecule *via* a sequence of consecutive reactions. Evidently, progress in the field of total synthesis has been greatly facilitated by progress in the area of method-oriented synthesis.

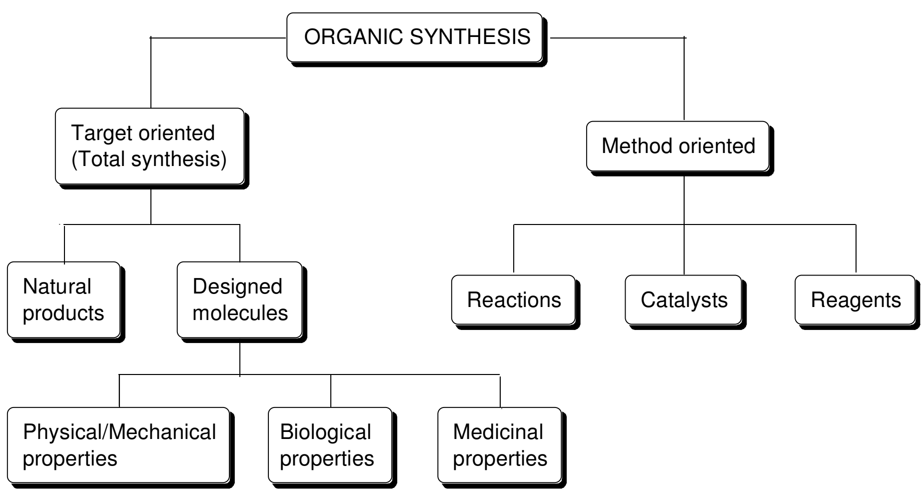


Figure 1.1 *Organic synthesis in perspective.*[1]

1.2 Objectives of total synthesis and target selection

1.2.1 Structure elucidation

Until the late 1960s, an important function of total synthesis was to confirm the molecular structure of natural products, which predefined the target molecule. A landmark success of this approach is the famous example of strychnine, which was first synthesized by Woodward and co-workers as early as 1954 (Figure 1.2).[2]

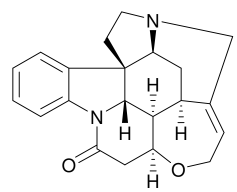
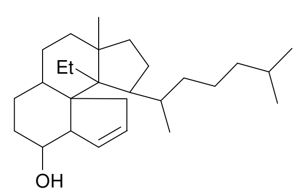
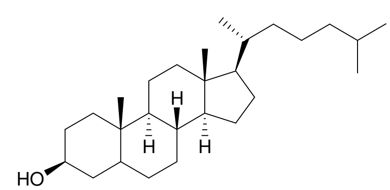


Figure 1.2 *Structure of strychnine*.[2]

Initial structure elucidation usually relied on degradation and/or derivatization of the isolated product (availability in gram quantities required), after which independent assembly from simple precursors was used for verification. This process was of course painstakingly slow and more importantly not flawless.[3] Since chemical transformations do not always proceed along projected lines, incorrect assignments were inevitable. The most notorious misassignment is arguably the proposed structure of cholesterol, for which Wieland and Windaus were awarded the Nobel Prize in Chemistry in 1927 and 1928, respectively.[4] Their proposals had a number of inaccuracies that were revealed in 1932 when Bernal obtained the first X-ray crystal structure of a steroid (Figure 1.3).[3,5]



Wieland/Windaus structure for cholesterol (1927)



Correct structure of cholesterol (1932)

Figure 1.3 *Classical misassignment of the structure of cholesterol*.[3]

Another typical example is the inaccurate structure for patchouli alcohol that was postulated by Büchi and colleagues in the early 1960s.[6] They successfully synthesized patchouli alcohol in four steps from the known compound α-patchoulene (Figure 1.4). Under the assumption that all chemical transformations had taken place according to their synthetic plan, they proposed a structure for the natural product. However, one year later an X-ray structure of a derivative proved that in reality their achievement was a lucky coincidence resulting from an unexpected skeletal rearrangement.[7]

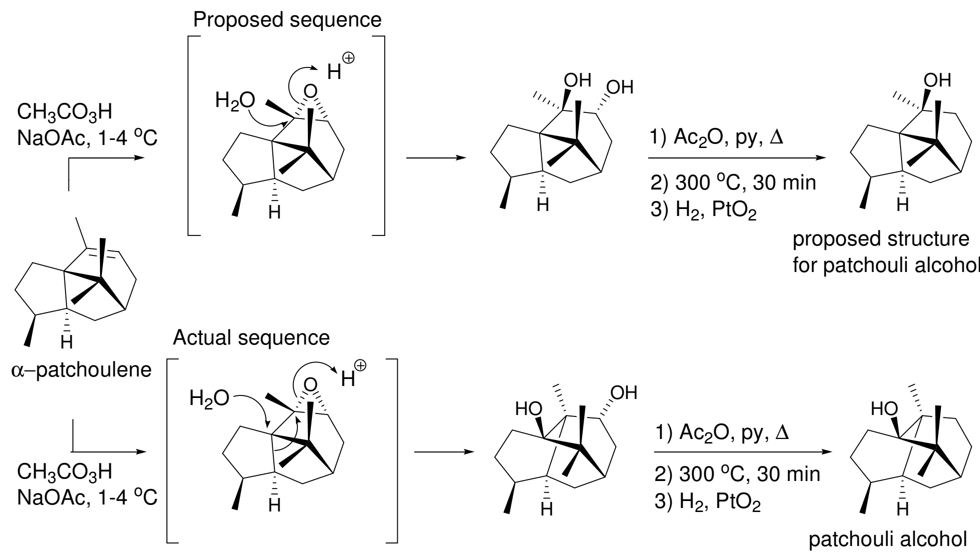


Figure 1.4 *Incorrect assignment of the structure of patchouli alcohol due to an unexpected skeletal rearrangement.*[3,6]

Fortunately, the chances to encounter such disappointments were dramatically reduced with the emergence and improvement of powerful analytical techniques like X-ray crystallography, nuclear magnetic resonance (NMR) and mass-spectrometry (MS).[1] At present, the isolation of minute amounts of a natural product is typically sufficient for a complete structural characterization apart from the assignment of the absolute or relative stereochemistry in specific cases (see, for example, chapter 4). When spectroscopic methods fail to conclusively settle stereochemical issues, total synthesis is still called upon to provide the answer. Needless to say, the laboriousness of this process increases with the number of unknown stereogenic elements.

1.2.2 Privileged structures

Currently, selection of a particular target molecule for total synthesis is often motivated by (potential) interesting properties of the final product like, for example, biological activity, possible medicinal applications, intriguing physical qualities or attractive material properties. Indisputably, total synthesis has indeed contributed to advances in a number of adjacent fields. As a consequence, the field of total synthesis is no longer restricted to natural products, but instead comprises designed molecules as well (Figure 1.1). Especially compounds with remarkable physical/mechanical properties (not covered in this thesis) often find their origin in the imagination of the organic chemist like for example the molecular elevator of Stoddart *et al.*[8] or the molecular motor of Feringa *et al*. (Figure 1.5).[9] For target molecules with an interesting biological/medicinal profile, on the other hand, Nature remains an important source of inspiration.[10] In fact, almost half of the drugs on the market are natural products or derivatives thereof.[11] Nevertheless, in the last decade, research on natural products by the pharmaceutical industry has declined. This is mainly due to a strategic shift toward combinatorial library synthesis of small organic molecules in combination with high-throughput screening for lead discovery as well as optimization.[10,12] However, the latest insights indicate that the generation and screening of a large number of synthetic compounds can at best complement but certainly not replace lead discovery *via* investigation of natural products, which have been carefully selected by evolutionary pressure.[10,12] This rehabilitation of natural products from a pharmacological point of view has led to renewed research activity in the area of total synthesis by industry as exemplified by the recent large scale synthesis of discodermolide[13] (§1.2.2.1) and current efforts to produce apoptolidin (Figure 1.6).[14]

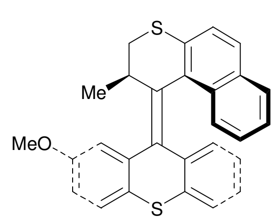


Figure 1.5 *Example of a rationally designed compound with interesting physical and mechanical properties: a unidirectional light driven molecular motor.*[9]

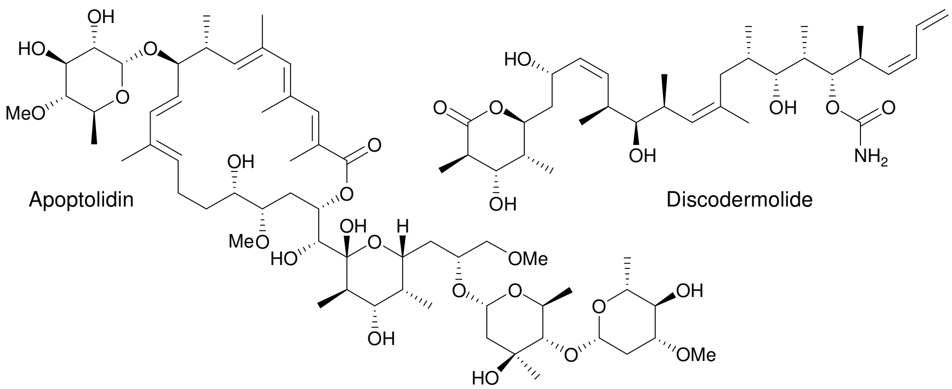


Figure 1.6 *Structures of apoptolidin*[15] *and discodermolide*.[16]

1.2.2.1 Natural product synthesis and drug development; the case of discodermolide

The most eye-catching examples that demonstrate the practical utility of total synthesis are without doubt applications in the preparation of drugs. Due to the low natural abundance of many natural compounds with an interesting biological profile, total synthesis is often the only method to supply the necessary amounts for clinical testing and eventually for distribution to patients. An illustrative example is the synthesis of discodermolide, which is a potent cell growth inhibitory agent, arresting cell development by binding and stabilizing mitotic spindle microtubules. Its mode of action is similar to other well-known anti-cancer agents like paclitaxel (taxol), epothilone B, laulimalide, sarcodictyin A, peloruside A and dictyostatin all of which have been the subject of extensive synthetic research.[17] In the case of discodermolide, six research groups have directed considerable synthetic effort toward providing a practical amount of material, culminating in several total syntheses.[18] Eventually, this has led to the preparation of over 60 grams of discodermolide for clinical trials by Novartis Pharmaceutical Corporation using a hybrid Smith-Paterson route that comprised 39 linear steps.[13] The development of discodermolide has recently been discontinued in part owing to toxic side effects.[19]

1.2.2.2 Structure-activity relationship (SAR)

Like in the case of discodermolide, natural products often turn out to be inadequate as a drug despite their promising pharmalogical profile. However, this does not necessarily mean that they are abandoned as a lead compound, since total synthesis is in principle capable of ‘improving’ the properties of the natural product *via* the preparation of analogues. As a matter of fact, even when the natural product qualifies as a drug, the complexity of the molecule can encourage the synthesis of more easily accessible analogues. In any event, optimization of a natural product requires knowledge regarding the structural features that are responsible for its drug-like properties. A powerful tool in determining the structure-activity relationship (SAR) of the molecule is the construction of a library of analogues with systematic variations in different domains. Using this approach, the ideal drug can be developed in a step-wise manner.

A successful example of such a strategy is the recently launched antithrombotic drug fondaparinux (arixtra; 2002), which is the result of twenty years of research using the natural drug heparin as a vantage point.[20] Heparin is a linear polysulfated polysaccharide isolated from animal organs that has been used clinically since the 1940s. *In vivo*, heparin forms a complex with antithrombin III (AT-III) resulting in allosteric activation of this protease inhibitor (Figure 1.7).[21] The interaction between heparin and AT-III is brought about by a unique sequence-specific pentasaccharide within the polysaccharide, also known as the antithrombin III binding domain (ABD; Figure 1.8). Upon binding, heparin induces a conformational change in AT-III, which results in an increased affinity of AT-III for thrombin and factor Xa, which are two key proteases in the blood-coagulation cascade.[20,21] For binding to factor Xa, activation of AT-III by the pentasaccharide is sufficient, while binding to thrombin additionally requires the presence of negatively-charged saccharide units at a remote position in the polysaccharide (thrombin binding domain: TBD). A shortcoming that is typically associated with heparin as an antithrombotic drug is a lack of selectivity caused by the presence of multiple negative charges on the polysaccharide. Specifically, heparin interacts in a non-selective way with other proteins like for instance platelet factor 4 (PF 4).[22] This can lead to so-called heparin-induced thrombocytopenia (HIT) indicating a shortage of platelets, which is associated with abnormal bleeding.

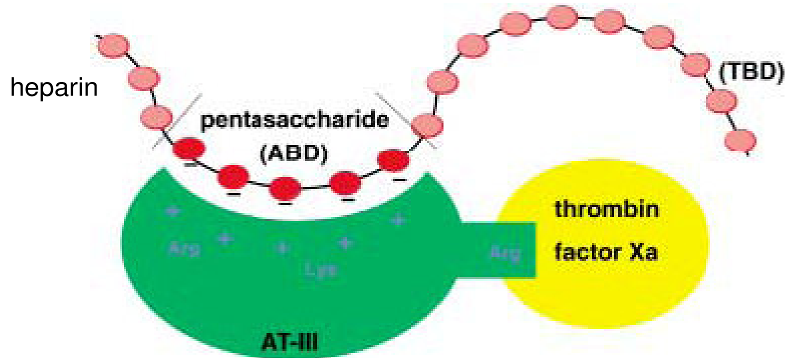


Figure 1.7 *Mode of action of the antithrombotic drug heparin. See text for details.*[21]

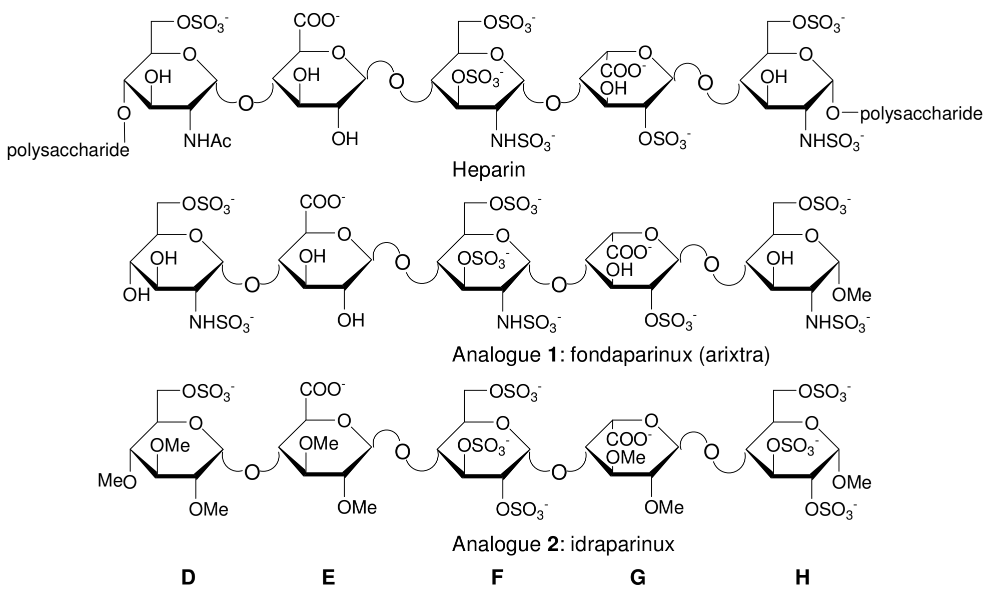


Figure 1.8 *Structure of the antithrombin III binding domain (ABD) in heparin and optimization of its structure during drug development.*[21]

With the objectives to prevent such side-effects and to develop an easier accessible and better characterized drug, a synthetic program was initiated with the primary goal to synthesize an active pentasaccharide for factor Xa inhibition.[20] In 1987, this resulted in the identification of fondaparinux (analogue **1**; Figure 1.8), which contains an *N*-sulfate group instead of an *N*-acetyl group in unit D and a methoxy group at the reducing end for ease of synthesis and purification.[21,23] Analogue **1** proved indeed to possess superior antithrombotic activity in comparison to heparin, while uncompromised by side-effects.[24] In addition, its half-life in humans is approximately 17 h compared to 1 h for heparin, which allows for lower dosages. After a successful clinical development program in the 1990s, fondaparinux is currently produced on multi-kg scale in industry, although its preparation is still lengthy (approximately 55 steps).[20,21]

Further investigations into the structure-activity relationships of heparin-like pentasaccharides, have led to the discovery of idraparinux (analogue **2**; Figure 1.8) which displays both higher activity as well as increased bio-availability (half-life of 120 h).[23,25] Moreover, due to the simplified chemical structure, orthogonal protecting groups are no longer required for the synthesis of **2**. In addition, units E, F, G and H can be obtained from a common disaccharide precursor. As a consequence, the total synthesis of **2** comprises (only) 25 steps. Idraparinux is currently in the last phase of clinical development.[20]

1.2.3 Synthetic challenges

The reluctance of the pharmaceutical industry to apply total synthesis in drug development in the past decade (§ 1.2.2), did not cause a diminished interest from the academic community. On the contrary, total synthesis thrived in that same period, resulting in the construction of complex targets like palytoxin,[26] brevetoxin[27] and vancomycin[28,29] among numerous others.[1,30,31] Part of the explanation for this contrast may be, that the academic community simply disagreed with the new industrial strategy. More importantly, though, the difference can be understood by taking truly academic arguments into account. The most important reason for a synthetic chemist to embark on an endeavor that may take years to complete, is a fundamental fascination for the complex molecular architecture itself and the wish to meet the challenges it poses with respect to the molecular skeleton, functional groups, stereochemistry and so on. In order to succeed, application of known methodologies may not suffice. Even the best retrosynthetic analysis can be subject to failure, due to the unpredictable outcome of well-established methodology once applied to complex substrates. Fine-tuning of reaction conditions, adaptation of the strategy, and if known methods fail, even development of new methodology are often required to prevail.

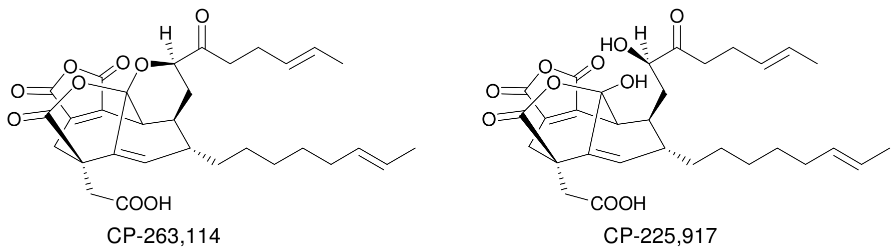


Figure 1.9 *Structures of the CP-molecules*.[32]

A particularly instructive example is the total synthesis of the CP-molecules (Figure 1.9) by Nicolaou and co-workers.[32] Even though CP-molecules exhibit cholesterol-lowering properties as well as anti-cancer activity, the main attraction from a total synthesis point of view stems from the overwhelming molecular frameworks in combination with the presence of sensitive functionalities. One of the hurdles that had to be overcome in the total synthesis of the CP-molecules, was the conversion of carboxylic acid **4** into α-diazoketone **5** (Figure 1.10). Even though this seems a trivial transformation at first sight, it soon appeared that known carboxyl-activation methods failed when addressing such a sterically demanding system.[33] This prompted the invention of a new method based on functionalization with sulfene ($SO_2{=}CH_2$; generated *in situ* from MsCl and Et_3N) followed by displacement with diazomethane. Not only did this novel protocol solve the problem at hand, but in general terms, the utility of acyl mesylates as acyl chloride surrogates for highly hindered carboxylic acids has been demonstrated on various occasions since then.[34]

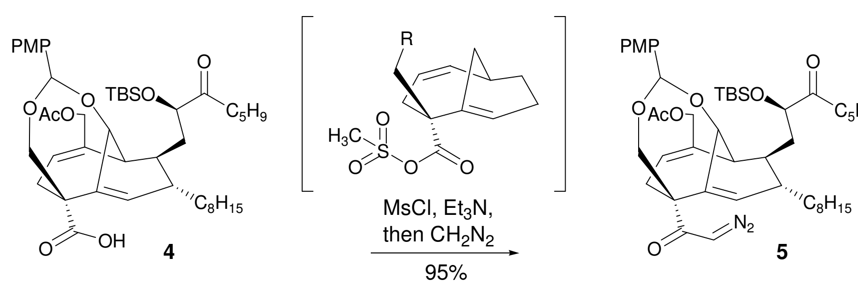


Figure 1.10 *Synthesis of a sterically congested α-diazoketone via mesylation*.[33]

Other valuable methodologies of general use that find their origin in the total synthesis of the CP-molecules are the homologation of sterically hindered aldehydes *via* cyanohydrin formation (Figure 1.11)[35] and (indirectly) the discovery of new applications for the oxidizing agents Dess-Martin periodinane (DMP) and *o*-iodylbenzoic acid (IBX).[36] Incidentally, one of these new applications of IBX is the introduction of unsaturation adjacent to carbonyl moieties,[37] which was used in the first step of the preparation of enantiopure saturated isoprenoid building blocks as described in chapter 3 of this thesis.

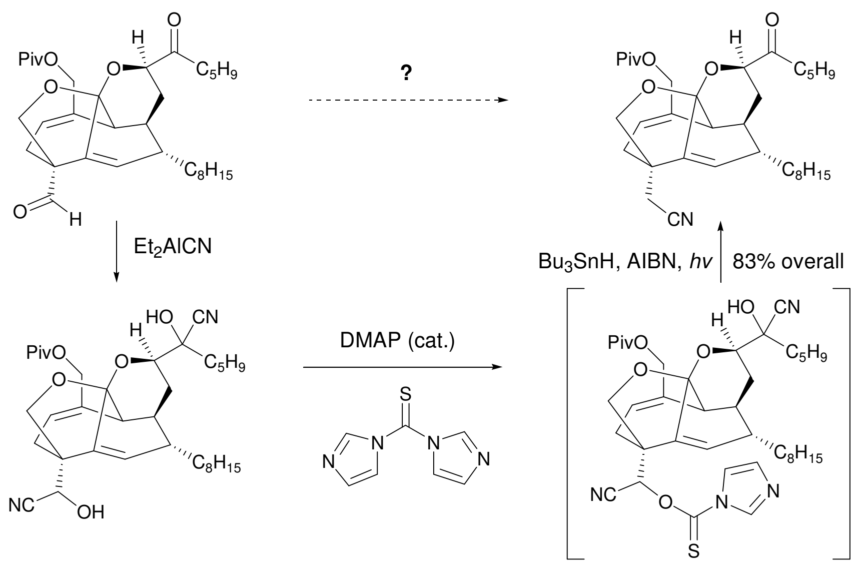


Figure 1.11 *One-carbon homologation of aldehydes to nitriles in the presence of a keton.*[33]

1.2.4 Method-inspired targets

From the previous paragraph it can be concluded, that total synthesis is an important test by which we judge the methods in our chemical toolbox. In the case of the CP-molecules, a new method to functionalize carboxylic acids was developed to facilitate arrival at the target structure (Figure 1.10). In general, though, the reversed process is infinitely more common; on many occasions, method-oriented organic chemists seek to apply a new synthetic protocol to the total synthesis of a more complex molecule with the objective to demonstrate its practical value. In such cases, a target containing a suitable structural motif is chosen to accommodate the method rather than selecting a method in pursuit of the target. A typical example is described elsewhere in this thesis, concerning the development of a method to prepare enantiopure saturated isoprenoid building blocks (chapter 3) followed by applications in the total syntheses of pheromones (chapter 3) and mannosyl *β*-phosphomycoketides (chapter 4).

1.3 Asymmetric catalysis in total synthesis

An important aspect in total synthesis is the introduction of stereogenic elements in an enantio- and/or diastereoselective way (Figure 1.12). In principle, there are three fundamentally different approaches to achieve enantioselectivity: 1) utilizing the *natural chiral pool* consisting of enantiopure starting materials provided by Nature; 2) *enantioselective synthesis*: preparation from achiral precursors using a chiral reagent/auxiliary or catalyst and 3) *resolution*: separation of enantiomers.[38] Resolution can in turn be divided into broadly three classes: 1) *resolution via diastereomers*: involving the use of a stoichiometric amount of a chiral resolving agent; 2) *chiral chromatography*: relying on the use of a chiral stationary phase and 3) *kinetic resolution*: involving the use of a chiral catalyst or reagent to promote selective reaction of one enantiomer over the other.[38] If this process is coupled to an *in situ* racemization protocol of the starting material (dynamic kinetic resolution) it has the same net result as enantioselective synthesis.

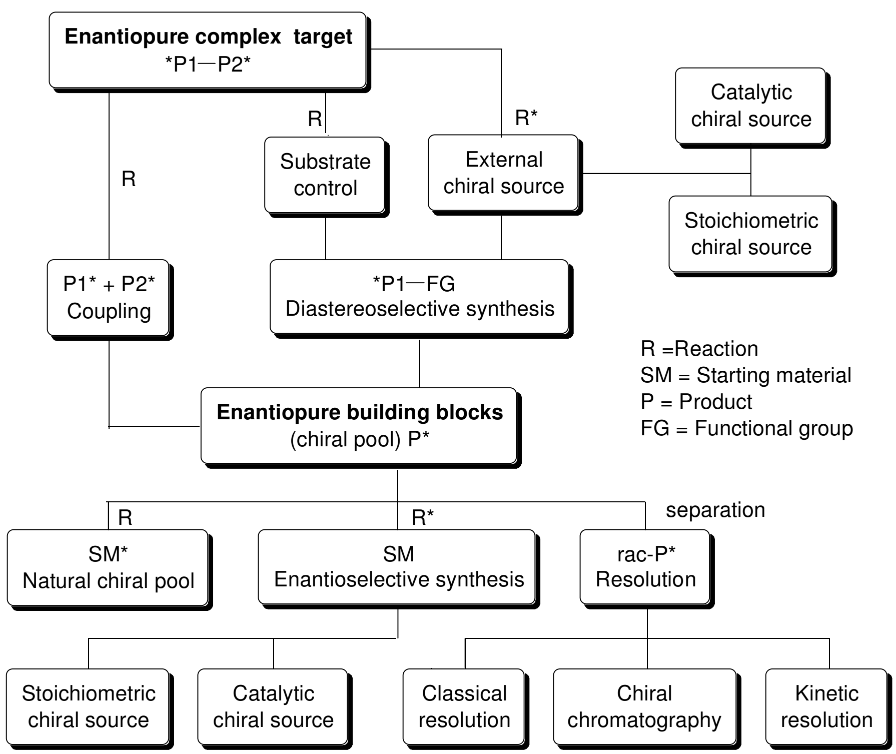


Figure 1.12 *Routes in the total synthesis of enantiopure complex targets.*[38,39]

Once a molecule contains a chiral element, there are basically two ways to selectively construct stereochemically more complex targets. The first strategy merely couples two enantiopure building blocks (Figure 1.12). The second approach creates a new stereogenic element using either an external chiral source like before (reagent-control, catalyst-control etc.) or takes advantage of the previously introduced chiral element in the substrate (substrate control).

After selecting one of the approaches described above, there are numerous methodologies to choose from. Within the field of asymmetric catalysis, for example, there is a distinction between bio-, organometallic- and organocatalysis and each of these methods can be either homogeneous or heterogeneous. In the wake of the rising number of different options, health and regulatory authorities have defined stricter requirements to produce new chiral drugs or agrochemicals.[40] As a consequence, single-enantiomer drug sales show a continuous growth reaching a worldwide sale of $159 billion in 2002.[14,41,42] In fact, in 2004, of all new synthetic chiral drugs (excluding peptides, proteins, monoclonal antibodies etc.) 92% was produced as a single enantiomer or diastereomer *versus* 8% as a racemic mixture.[43] As a result, economic motives have joined fundamental interests in operating as a driving force for the improvement of the various routes to enantiomerically pure compounds.[44]

The question that now presents itself is which of the above approaches is best qualified to do the job in a particular total synthesis. To answer this question it is important to realize, that the choice for an approach not only depends on the structure of the target compound, but also on the underlying motivation for target selection (see § 1.2). In other words: ‘how’ depends as much on ‘why’ as it does on ‘what’.

1.3.1 Trends in methodology

From a methodology point of view, the current paradigm in introducing chirality is to develop catalytic asymmetric versions of commonly used reactions. The emphasis on this particular approach stems from fundamental advantages that asymmetric catalysis holds over other methodologies. Specifically, asymmetric catalysis is in theory capable of delivering all possible diastereomers (in contrast to the natural chiral pool or substrate control) in 100% yield (as opposed to resolution) using only a small amount of chiral material (contrary to stoichiometric approaches).

An early demonstration of the power of asymmetric catalysis was given by Masamune and Sharpless in their synthesis of the L-hexoses in 1983.[39,45] Using their titanium tartrate epoxidation catalyst, they introduced all four contiguous stereocenters in a controlled manner (Figure 1.13). This accomplishment inspired the development of an enormous number of different catalysts for all sorts of asymmetric transformations.[46] However, despite some important successes,[39,47] the rate of implementation of asymmetric catalysis in total synthesis does not seem to follow the rate of discovery of catalysts.

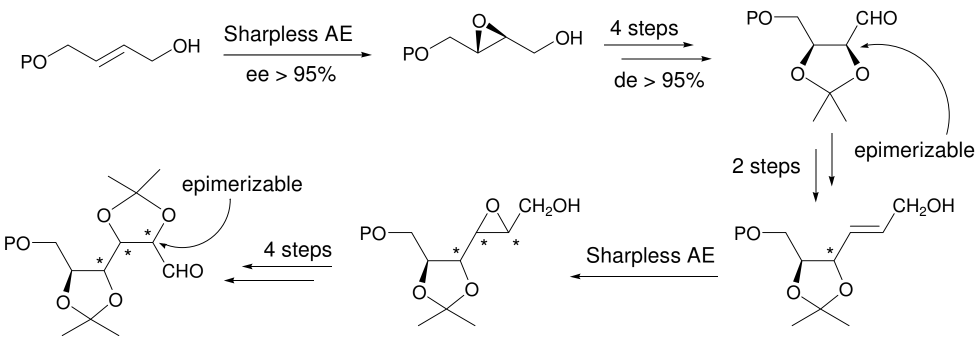


Figure 1.13 *Masamune-Sharpless synthesis of L-hexoses. All stereocenters under control.*[45]

1.3.2 Considerations in total synthesis

What is causing this paradox between the development of asymmetric catalysts and their application in total synthesis? As mentioned before (§ 1.2.4), total synthesis is an important test by which we evaluate the value of a method. It is probable that some catalysts simply fail this test, meaning that they have to be improved in order to be applied. This is particularly evident when a stoichiometric chiral source is used instead of a catalyst. After all, there is no strategic gain in choosing a reagent/auxiliary over a catalyst. The fact that many total syntheses still use stoichiometric approaches therefore indicates that the toolbox of efficient and generally applicable asymmetric catalysts is still relatively empty except for in the areas of oxidation (Sharpless) and hydrogenation.[39] To be fair it should be noted, that total synthesis can be a harsh judge for a catalyst. At a progressed stage of a total synthesis, the substrate is obviously the most valuable component in the reaction so the costs of the chiral source are often negligible. On the other hand, some catalysts are composed of such expensive ligands and/or metals that they are not cost-effective despite the small amount required. The preparation of certain types of ligands requires a multi-step synthesis in itself. In addition, application of some asymmetric catalysts necessitates the use of specialized equipment including autoclaves or glove-boxes.

Even when a suitable asymmetric catalyst is available, it is conceivable that another approach is chosen. This can be explained by the fact, that in specific total syntheses one or more of the fundamental advantages of asymmetric catalysis may be considered irrelevant. In such cases other strategies like resolution, the natural chiral pool or substrate control can be tough competitors.

1.3.2.1 Total synthesis of libraries of stereoisomers

When total synthesis serves the purpose of elucidating the stereochemistry of a natural product or when it is used to determine the influence of the stereochemistry of a biomolecule on the biological activity (SAR), the task of the organic chemist is to synthesize a number of stereoisomers. In such cases, the flexibility of asymmetric catalysis can be fully exploited in two different ways. First of all, it can be used to expand the natural chiral pool with all stereoisomers of simple enantiopure building blocks (for an example see chapter 3). Furthermore, complex intermediates can be converted into different diastereoisomers along the same route (Figure 1.12). Especially, this latter application shows the true potential of a catalyst as it is usually confronted with potentially reactive functional groups and it has to overcome inherent substrate control.[39]

Since the target is a library of stereoisomers, an approach based on resolution is a viable alternative to asymmetric catalysis. However, asymmetric catalysis has the advantage that the same route is still efficient if, at a later stage, one particular stereoisomer is to be prepared. A typical example is the synthesis of the side chain of mycolactones A and B by Kishi and co-workers (chapter 2). For the determination of the stereochemistry of the natural product, two diastereomers were prepared as a 1:1 mixture after which they were separated by column chromatography (Figure 1.14).[48] In contrast, a catalytic asymmetric Sharpless dihydroxylation was used in the eventual total synthesis of the correct diastereomer (scheme 2.6).[49]

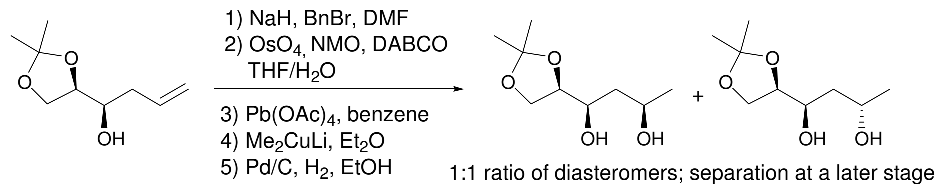


Figure 1.14 *Preparation of a mixture of diastereomers for structure elucidation.*[48]

1.3.2.2 Synthesis of a single stereoisomer

When the challenge is the synthesis of a natural product with known stereochemistry or when the target is an optimized privileged structure, the preparation of a single stereoisomer is required. In such cases, flexibility of the synthetic route is of little concern. Instead, the focus shifts to overall efficiency (yield, convergence, selectivity, costs, etc.). If an abundant chiral building block from Nature can be used as a starting material, this approach is often unbeatable in terms of selectivity (high enantiomeric excess [ee] and diastereomeric excess [de]) and costs. The advantage is proportional to the complexity of the natural starting material. And even when Nature does not provide the desired structural motif, modification of a natural building block may prove an attractive option (see, for example, chapter 2, scheme 2.10 and 2.11). In fact, although counterintuitive for an organic chemist, it can occasionally pay to break down readily available complex natural products to obtain simple enantiopure building blocks (Figure 1.15).[50] The extent to which such processes are efficient depends on the number of steps and the overall yield in comparison to the alternative: asymmetric catalysis (or stoichiometric approaches when a suitable catalyst is unavailable). Hence, while in general the natural chiral pool and asymmetric catalysis (§ 1.3.2.1) are complementary approaches for building block synthesis, there is a large grey area in which they compete.

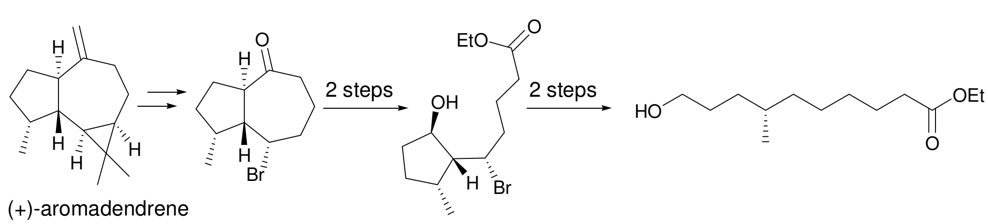


Figure 1.15 *Synthesis of enantiopure building blocks from complex natural products.*[50]

When performing a reaction on an advanced chiral intermediate that creates a new stereogenic element, substrate control can often be used to favor formation of the desired diastereomer. Knowledge of the reaction mechanism is essential since the stereochemical outcome sometimes depends as much on the reaction pathway as it does on the substrate (Figure 1.16). The fact that substrate control operates without an external chiral source makes it an attractive strategy, which is both elegant and efficient. Asymmetric catalysis is in this case truly complementary: it should be applied when the levels of diastereoselectivity induced by the substrate are either unsatisfactory or when the desired stereoisomer cannot be formed altogether (§ 1.3.2.1). An impressive display of how substrate control can be optimally exploited was given by Paterson *et al.* in their recently disclosed optimized route to discodermolide (Figure 1.4).[17] They constructed this complex target from a commercially available enantiopure synthon in 7.8% yield over 24 linear steps (35 steps in total) without using any external chiral source. One of the key reactions in their total synthesis was the Evans protocol to convert *β*-hydroxy ketones into the corresponding *anti* 1,3-diols as depicted in Figure 1.16.[51]

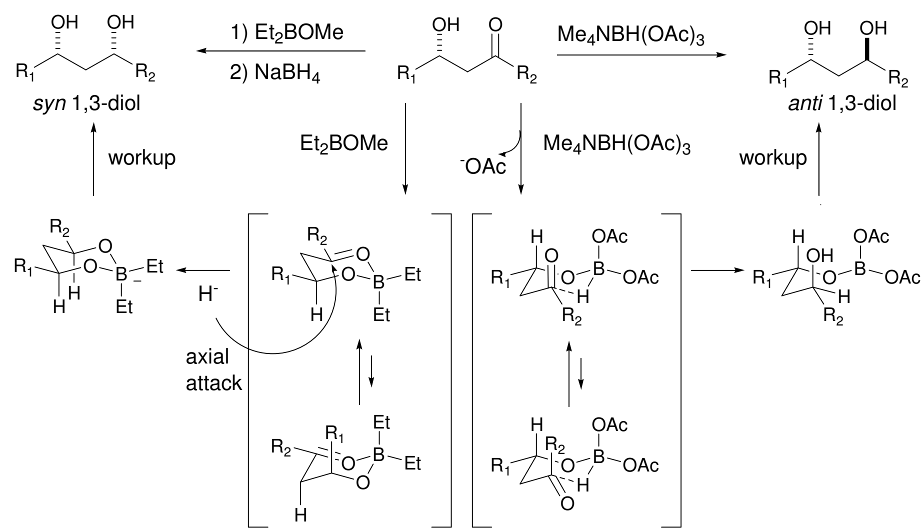


Figure 1.16 *Complementary reduction methods to selectively convert a β-hydroxyketone into either the syn-*[52] *or the anti-1,3-diol.*[51]

1.3.2.3 Commercial considerations

Although total synthesis is a useful tool to judge the performance of asymmetric catalysts, the verdict is sometimes clouded because methods occasionally tend to become more important than the target in an academic environment. This can, for instance, be the case if a target is selected to underscore the power of a catalyst. In such examples it is tempting to undertake an elaborate synthesis for the sake of the key asymmetric transformation(s). The ultimate impartial test in cases like that is commercialization, which takes the whole synthetic route into consideration. An illustrative example is the elegant total synthesis of (R,R,R)-α-tocopherol (Vitamin E),[a] which uses catalytic asymmetric hydrogenation to introduce the asymmetric centers in the side chain (Figure 1.17).[53,54,55] Although the catalysts proved to be extremely successful, the route depicted in Figure 1.17 could in the end not compete with partial synthesis from naturally occurring tocopherols due to inefficient substrate preparation and cross coupling procedures. For example, the separation of *trans*-**7** from its *cis*-isomer formed a technical bottleneck that was difficult to overcome. New catalytic strategies, like the one recently reported by Pfaltz *et al*. employing the asymmetric hydrogenation of unfunctionalized olefins (Figure 1.18),[56] may tip the balance in favour of asymmetric catalysis in the future. However, also in this case, substrate synthesis as well as catalyst loading and other technical aspects have to be critically evaluated.

Commercial application of an asymmetric total synthesis route evaluates not only flexibility and/or efficiency, but more practical aspects as well.[57] For example, the need for high dilution or low reaction temperature can render scale-up extremely difficult or expensive. In addition, purification of the product can be critical, especially in the case of toxic heavy-metal-catalyzed reactions. Furthermore, the often limited development time can be a decisive factor.[58] In the pharmaceutical industry, the typical strategy is therefore to synthesize a certain amount of product for clinical trials as quickly as possible and to optimize the route if the drug is a success. For instance, in the large scale synthesis of

[a] Vitamin E is currently sold as a stereorandom mixture (all-rac-form). Since the stereochemistry of the side chain has no distinct influence on the biological activity, there is no economic incentive to change to the enantiopure form.

discodermolide (§1.2.2.1), all stereogenic elements were introduced using chiral auxiliaries and reagents.[13] To find suitable catalysts to replace them, would have taken years and would eventually have been in vain since the development of discodermolide as a drug was discontinued.[19] Besides, it is doubtful whether such catalysts would have been able to compete with the substrate controlled route of Paterson *et al.* (§1.3.2.2).[17] This all goes to show once more, that the best approach to introduce chirality in total synthesis does not only depend on the target, but also on the motivation for target selection.

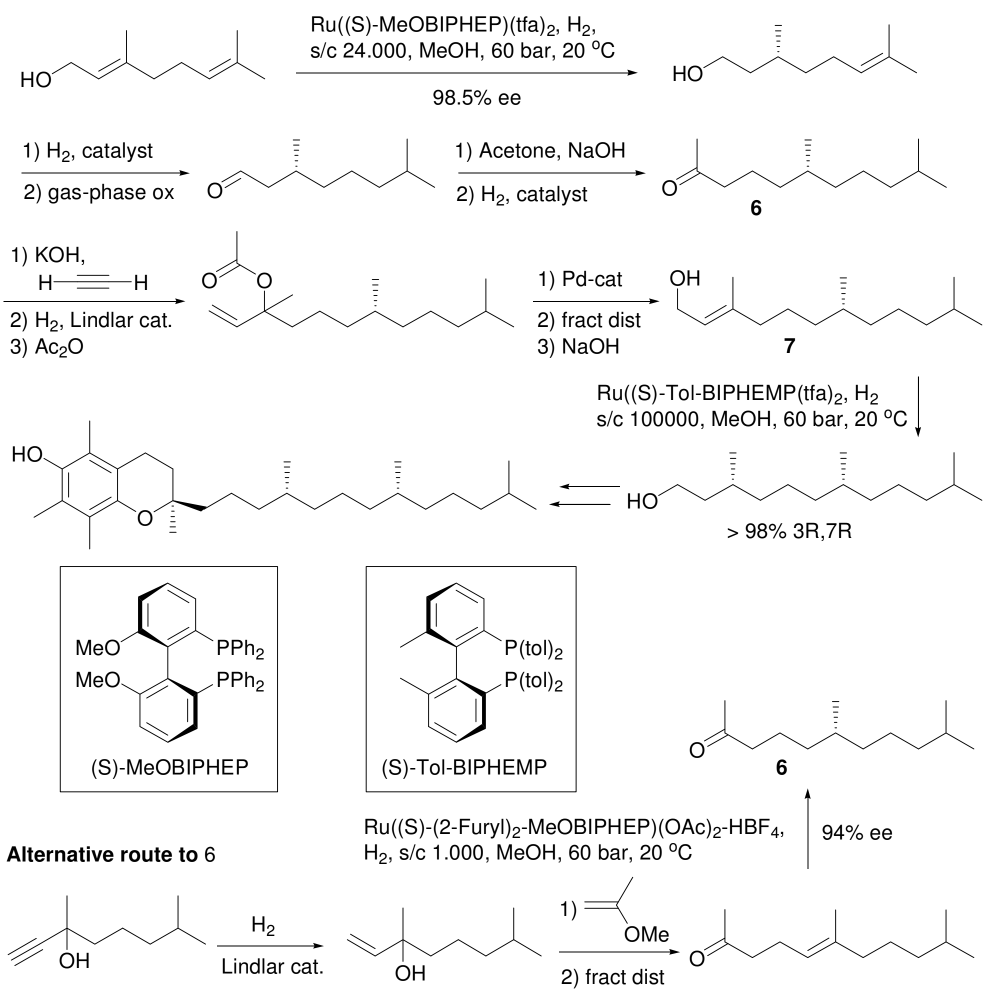


Figure 1.17 *Synthesis of (R,R,R)-α-tocopherol side chain using catalytic asymmetric hydrogenation to introduce the stereogenic elements.*[53]

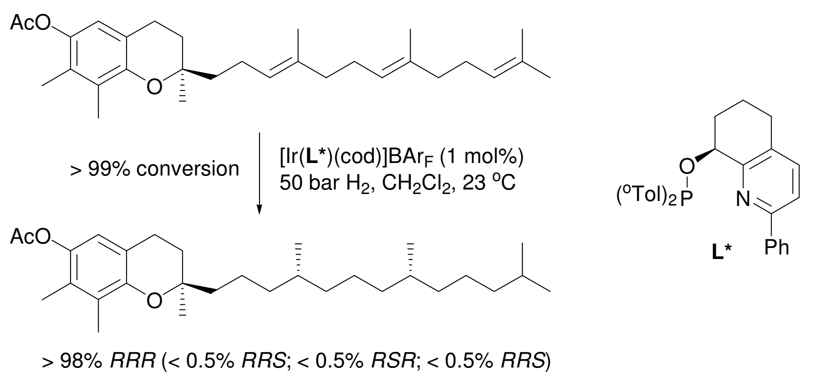


Figure 1.18 *Application of asymmetric hydrogenation of unfunctionalized olefins in the synthesis of γ-tocopherol.*[56]

1.3.3 Conclusion

Asymmetric catalysis is a valuable instrument for total synthesis. Its importance will only increase in the future with the development of a larger toolbox filled with better catalysts for a wider range of transformations. It is particularly useful for expanding the chiral pool with new enantiopure building blocks and for overcoming inherent substrate bias in complex target synthesis.[39] It is therefore important that the power of a catalyst is demonstrated in complex products beyond benchmark substrates. However, with respect to total synthesis, asymmetric catalysis remains a means to an end and should not become a goal in itself. Related to this, the extend to which the fundamental advantages of asymmetric catalysis are in fact beneficial to a specific total synthesis is highly variable. Depending on the target and on the motivation for target selection, there is often an alternative (natural chiral pool, substrate control or resolution). The question should therefore remain: 'What is the best way to synthesize this particular target?' rather than 'How can we make this target using an asymmetric catalyst?'

1.4 Aims and outline of this thesis

The remainder of this thesis will be devoted to the fruits (immature or not) of my efforts in the area of enantiopure total synthesis. The aim of every total synthesis discussed in this thesis is to develop a route to the final product that is both efficient as well as flexible especially in relation to the introduction of stereogenic elements. Both the chiral pool and asymmetric catalysis play an eminent role in achieving this goal. To put the work into a broader context, the biological background as well as the synthetic history of each target compound will be introduced prior to discussion of the actual synthesis.

In chapter 2, new approaches towards the synthesis of the side chain of mycolactones A and B will be discussed. Notable differences with previous approaches include a new strategy to construct the elaborate conjugated system and introduction of chirality through modification of monosaccharides rather than asymmetric catalysis. Chapter 3 deals with the development of an asymmetric catalytic method to prepare all diastereomers of enantiopure saturated isoprenoid building blocks. The practical value of this methodology is demonstrated by its application in the synthesis of two insect pheromones. In chapter 4, the saturated isoprenoid building blocks are used once more in the enantiopure total synthesis of a β-D-mannosyl phosphomycoketide. For this purpose, a protocol is developed to synthesize enantiopure oligoisoprenoid chains of any desired length and stereochemistry. Another important aspect of this total synthesis is the formation of the β-mannosyl glycosidic linkage.

1.5 References

[1] Nicolaou, K. C.; Sorensen, E. J. *Classics in Total Synthesis I* **1996**, VCH, New York.
[2] Woodward, R. B.; Cava, M. P.; Ollis, W. D.; Hunger, A.; Daeniker, H. U.; Schenker, K. *J. Am. Chem. Soc.* **1954**, *76*, 4749-4751.
[3] Nicolaou, K. C.; Snyder, S. A. *Angew. Chem. Int. Ed.* **2005**, *44*, 1012-1044.
[4] See the Nobel Prize website: http://www.nobel.se/chemistry.
[5] Bernal, J. D. *Nature* **1932**, *129*, 721.
[6] (a) Büchi, G.; Erickson, R. E.; Wakabayashi, N. *J. Am. Chem. Soc.* **1961**, *83*, 927-938; (b) Büchi, G.; MacLeod, W. D. *J. Am. Chem. Soc.* **1962**, *84*, 3205-3206.
[7] (a) Dobler, M.; Dunitz, J. D.; Gubler, B.; Weber, H. P.; Büchi, G.; Padilla, O. J. *Proc. Chem. Soc. London* **1963**, 383; (b) Büchi, G.; MacLeod, W. D.; Padilla, O. J. *J. Am. Chem. Soc.* **1964**, *86*, 4438-4444.
[8] Badjić, J. D.; Balzani, V.; Credi, A.; Silvi, S.; Stoddart, J. F. *Science* **2004**, *303*, 1845-1849.
[9] Koumura, N.; Geertsema, E. M.; Meetsma, A.; Feringa, B. L. *J. Am. Chem. Soc.* **2000**, *122*, 12005-12006.
[10] Koehn, F. E.; Carter, G. T. *Nature Rev. Drug Discov.* **2005**, *4*, 206-220.

[11] Grifo, F.; Newman, D.; Fairfield, A. S.; Bhattaharya, B.; Grupenhoff, J. T. *The Origins of Prescription Drugs*. In *Biodiversity and Human Health* Grifo, F.; Rosenthal, J. Eds. Island Press: Washington, DC, **1997**, p 131.
[12] Ortholand, J. -Y.; Ganesan, A. *Curr. Opin. Chem. Biol.* **2004**, *8*, 271-280.
[13] (a) Mickel, S. J.; Sedelmeier, G. H.; Niederer, D.; Daeffler, R.; Osmani, A.; Schreiner, K.; Seeger-Weibel, M.; Bérod, B.; Schaer, K.; Gamboni, R.; Chen, S.; Chen, W.; Jagoe, C. T.; Kinder, F. R. jr.; Loo, M.; Prasad, K.; Repic, O.; Shieh, W. -C.; Wang, R. -M.; Waykole, L.; Xu, D. D.; Xue, S. *Org. Process Res. Dev.* **2004**, *8*, 92-100; (b) Mickel, S. J.; Sedelmeier, G. H.; Niederer, D.; Schuerch, F.; Grimler, D.; Koch, G.; Daeffler, R.; Osmani, A.; Hirni, A.; Schaer, K.; Gamboni, R.; Bach, A.; Chaudhary, A.; Chen, S.; Chen, W.; Hu, B.; Jagoe, C. T.; Kim, H. -Y.; Kinder, F. R. jr.; Liu, Y.; Lu, Y.; McKenna, J.; Prashad, M.; Ramsey, T. M.; Repic, O.; Rogers, L.; Shieh, W. -C.; Wang, R. -M.; Waykole, L. *Org. Process Res. Dev.* **2004**, *8*, 101-106; (c) Mickel, S. J.; Sedelmeier, G. H.; Niederer, D.; Schuerch, F.; Koch, G.; Kuesters, E.; Daeffler, R.; Osmani, A.; Seeger-Weibel, M.; Schmid, E.; Hirni, A.; Schaer, K.; Gamboni, R.; Bach, A.; Chen, S.; Chen, W.; Geng, P.; Jagoe, C. T.; Kinder, F. R. jr.; Lee, G. T.; McKenna, J.; Ramsey, T. M.; Repic, O.; Rogers, L.; Shieh, W. -C.; Wang, R. -M.; Waykole, L. *Org. Process Res. Dev.* **2004**, *8*, 107-112; (d) Mickel, S. J.; Sedelmeier, G. H.; Niederer, D.; Schuerch, Seger, M.; Schreiner, K.; Daeffler, R.; Osmani, A.; Bixel, D.; Loiseleur, O.; Cercus, J.; Stettler, H.; Schaer, K.; Gamboni, R.; Bach, A.; Chen, G. -P.; Chen, W.; Geng, P.; Lee, G. T.; Loeser, E.; McKenna, J.; Kinder, F. R. jr.; Konigsberger, K.; Prasad, K.; Ramsey, T. M.; Reel, N.; Repic, O.; Rogers, L.; Shieh, W. -C.; Wang, R. -M.; Waykole, L.; Xue, S.; Florence, G.; Paterson, I. *Org. Process Res. Dev.* **2004**, *8*, 113-121; (e) Mickel, S. J.; Niederer, D.; Daeffler, R.; Osmani, A.; Kuesters, E.; Schmid, E.; Schaer, K.; Gamboni, R.; Chen, W.; Loeser, E.; Kinder, F. R. jr.; Konigsberger, K.; Prasad, K.; Ramsey, T. M.; Repic, O.; Wang, R. -M.; Florence, Lyothier, I.; Paterson, I. *Org. Process Res. Dev.* **2004**, *8*, 122-130; (f) Mickel, S. J.; Daeffler, R.; Prikoszovich, W. *Org. Process Res. Dev.* **2005**, *9*, 113-120
[14] Rouhi, A. M. *Chem. Eng. News* **2003**, *81*, 45-55.
[15] Hayakawa, Y.; Kim, J. W.; Adachi, H.; Shin-ya, K.; Fujita, K. -i.; Seto, H. *J. Am. Chem. Soc.* **1998**, *120*, 3524-3525.
[16] Gunasekera, S. P.; Gunasekera, M.; Longley, R. E.; Schulte, G. K. *J. Org. Chem.* **1990**, *55*, 4912-4915.
[17] Paterson, I.; Delgado, O.; Florence, G. J.; Lyothier, I.; O'Brien, M.; Scott, J. P.; Sereinig, N. *J. Org. Chem.* **2005**, *70*, 150-160.
[18] (a) Nerenberg, J. B.; Hung, D. T.; Somers, P. K.; Schreiber, S. L. *J. Am. Chem. Soc.* **1993**, *115*, 12621-12622; (b) Hung, D. T.; Nerenberg, J. B.; Schreiber, S. L. *J. Am. Chem. Soc.* **1996**, *118*, 11054-11080; (c) Smith, A. B. III; Qiu, Y.; Jones, D. R.; Kobayashi, K. *J. Am. Chem. Soc.* **1995**, *117*, 12011-12012; (d) Smith, A. B. III; Kaufman, M. D.; Beauchamp, T. J.; LaMarche, M. J.; Arimoto, H. *Org. Lett.* **1999**, *1*, 1823-1826; (e) Smith, A. B. III; Beauchamp, T. J.; LaMarche, M. J.; Kaufman, M. D.; Qui, Y.; Arimoto, H.; Jones, D. R.; Kobayashi, K. *J. Am. Chem. Soc.* **2000**, *122*, 8654-8664; (f) Smith, A. B. III; Freeze, B. S.; Brouard, I.; Hirose, T. *Org. Lett.* **2003**, *5*, 4405-4408; (g) Yang, G.; Myles, D. C. *Tetrahedron Lett.* **1994**, *35*, 1313-1316; (h) Yang, G.; Myles, D. C. *Tetrahedron Lett.*

1994, *35*, 2503-2504; (i) Harried, S. S.; Yang, G.; Strawn, M. A.; Myles, D. C. *J. Org. Chem.* **1997**, *62*, 6098-6099; (j) Harried, S. S.; Lee, C. P.; Yang, G.; Lee, T. I. H.; Myles, D. C. *J. Org. Chem.* **2003**, *68*, 6646-6660; (k) Marshall, J. A.; Lu, Z. -H.; Johns, B. A. *J. Org. Chem.* **1998**, *63*, 817-823; (l) Marshall, J. A.; Johns, B. A. *J. Org. Chem.* **1998**, *63*, 7885-7892; (m) Paterson, I.; Florence, G. J.; Gerlach, K.; Scott, J. P. *Angew. Chem. Int. Ed.* **2000**, *39*, 377-380; (n) Paterson, I.; Florence, G. J. *Tetrahedron Lett.* **2000**, *41*, 6935-6939; (o) Paterson, I.; Florence, G. J.; Gerlach, K.; Scott, J. P.; Sereinig, N. *J. Am. Chem. Soc.* **2001**, *123*, 9535-9544; (p) Paterson, I.; Delgado, O.; Florence, G. J.; Lyothier, I.; Scott, J. P.; Sereinig, N. *Org. Lett.* **2003**, *5*, 35-38; (q) Paterson, I.; Delgado, O. *Tetrahedron Lett.* **2003**, *44*, 8877-8882; (r) Paterson, I.; Lyothier, I. *Org. Lett.* **2004**, *6*, 4933-4936. (s) Arefolov, A.; Panek, J. S. *J. Am. Chem. Soc.* **2005**, *127*, 5596-5603; (t) For a review, see: Paterson, I.; Florence, G. J. *Eur. J. Org. Chem.* **2003**, 2193-2208.

[19] Federsel, H. -J. *Nat. Rev. Drug Discov.* **2005**, *4*, 685-697.

[20] Petitou, M.; Van Boeckel, C. A. A. *Angew. Chem. Int. Ed.* **2004**, *43*, 3118-3133.

[21] Van Boeckel, C. A. A.; Petitou, M. *Angew. Chem. Int. Ed. Engl.* **1993**, *32*, 1671-1690.

[22] Noti, C.; Seeberger, P. H. *Chem. Biol.* **2005**, *12*, 731-756.

[23] Herbert, J. M.; Herault, J. P.; Bernat, A.; Van Amsterdam, R. G. M.; Lormeau, J. C.; Petitou, M.; Van Boeckel, C. A. A.; Hoffmann, P.; Meuleman, D. G. *Blood* **1998**, *91*, 4197-4205.

[24] Bauer, K. A.; Hawkins, D. W.; Peters, P. C.; Petitou, M.; Herbert, J. -M.; Van Boeckel, C. A. A.; Meuleman, D. G. *Cardiovasc. Drug. Rev.* **2002**, *20*, 37-52.

[25] Westerduin, P.; Van Boeckel, C. A. A.; Basten, J. E. M.; Broekhoven, M. A.; Lucas, H.; Rood, A.; Van der Heijden, H.; Van Amsterdam, R. G. M.; Van Dinther, T. G.; Meuleman, D. G.; Visser, A.; Vogel, G. M. T.; Damm, J. B. L.; Overklift, G. T. *Bioorg. Med. Chem.* **1994**, *2*, 1267-1280.

[26] Suh, E. M.; Kishi, Y. *J. Am. Chem. Soc.* **1994**, *116*, 11205-11206 and references therein.

[27] Nicolaou, K. C. *Angew. Chem. Int. Ed.* **1996**, *35*, 589-607 and references therein.

[28] (a) Evans, D. A.; Wood, M. R.; Trotter, B. W.; Richardson, T. I.; Barrow, J. C.; Katz, J. L. *Angew. Chem. Int. Ed.* **1998**, *37*, 2700-2704; (b) Evans, D. A.; Dinsmore, C. J.; Watson, P. S.; Wood, M. R.; Richardson, T. I.; Trotter, B. W.; Katz, J. L. *Angew. Chem. Int. Ed.* **1998**, *37*, 2704-2708.

[29] (a) Nicolaou, K. C.; Natarajan, S.; Li, H.; Jain, N. F.; Hughes, R.; Solomon, M. E.; Ramanjulu, J. M.; Boddy, C. N. C.; Takayanagi, M. *Angew. Chem. Int. Ed.* **1998**, *37*, 2708-2714; (b) Nicolaou, , K. C.; Jain, N. F.; Natarajan, S.; Hughes R.; Solomon, M. E.; Li, H.; Ramanjulu, J. M.; Takayanagi, M.; Koumbis, A. E.; Bando, T. *Angew. Chem. Int. Ed.* **1998**, *37*, 2714-2717; (c) Nicolaou, K. C.; Takayanagi, M.; Jain, N. F.; Natarajan, S.; Koumbis, A. E.; Bando, T.; Ramanjulu, J. M. *Angew. Chem. Int. Ed.* **1998**, *37*, 2717-2719; (d) Nicolaou, K. C.; Mitchell, H. J.; Jain, N. F.; Winssinger, N.; Hughes, R.; Bando, T. *Angew. Chem. Int. Ed.* **1999**, *38*, 240-244.

[30] Nicolaou, K. C.; Snyder, S. A. *Classics in Total Synthesis II* **2003**, VCH, New York.

[31] Nicolaou, K. C. *J. Org. Chem.* **2005**, *70*, 7007-7027.

[32] (a) Nicolaou, K. C.; Baran, P. S.; Zhong, Y. -L.; Choi, H. -S.; Yoon, W. H.; He, Y.; Fong, K. C. *Angew. Chem. Int. Ed.* **1999**, *38*, 1669-1675; (b) Nicolaou, K. C.; Baran, P. S.;

Zhong, Y. -L.; Fong, K. C.; He, Y.; Yoon, W. H.; Choi, H. -S. *Angew. Chem. Int. Ed.* **1999**, *38*, 1676-1678.
[33] Nicolaou, K. C.; Baran, P. S. *Angew. Chem. Int. Ed.* **2002**, *41*, 2678-2720.
[34] Nicolaou, K. C.; Baran, P. S.; Zhong, Y. -L.; Choi, H. -S.; Fong, K. C. ; He, Y. ; Yoon, W. H. *Org. Lett.* **1999**, *1*, 883-886.
[35] Nicolaou, K. C.; Vassilikogiannakis, G.; Kranich, R.; Baran, P. S.; Zhong, Y. -L.; Natarajan, S. *Org. Lett.* **2000**, *2*, 1895-1898.
[36] (a) Nicolaou, K. C.; Mathison, C. J. N. *Angew. Chem. Int. Ed.* **2005**, *44*, 5992-5997 and references therein.
[37] Nicolaou, K. C.; Montagnon, T.; Baran, P. S.; Zhong, Y. -L. *J. Am. Chem. Soc.* **2002**, *124*, 2245-2258.
[38] Keith, J. M.; Larrow, J. F.; Jacobsen, E. N. *Adv. Synth. Catal.* **2001**, *343*, 5-26.
[39] Taylor, M. S.; Jacobsen, E. N. *Proc. Natl. Acad. Sci.* **2004**, *101*, 5368-5373.
[40] U.S. Food and Drug Administration, FDA's policy statement for the development of new stereoisomeric drugs, *Fed Regist* **1992**, *57*, FR22249, available at: http://www.fda.gov/cder/guidance/stereo.htm.
[41] Tucker, G. T. *Lancet* **2000**, *355*, 1085-1087.
[42] Kulig, K.; Nowicki, P.; Malawska, B. *Pol. J. Pharmacol.* **2004**, *56*, 499-508.
[43] American Medical Association, *Geometric Isomerism and Chirality: The USAN Perspective* available at: http://www.ama-assn.org/ama/pub/category/15698.html.
[44] Maier, N. B.; Franco, P.; Lindner, W. *J. Chromatogr. A* **2001**, *906*, 3-33.
[45] Ko, S. Y. ; Lee, A. W. M. ; Masamune, S. ; Reed, L. A. III ; Sharpless, K. B.; Walker, F. J. *Science* **1983**, *220*, 949-951.
[46] Jacobsen, E. N.; Pfaltz, A.; Yamamoto, H. In *Comprehensive Asymmetric Catalysis*, Springer, New York, **1999**, Vol 1-3.
[47] Hoveyda, A. H. In *Stimulating Concepts in Chemistry*, eds. F. J. Stoddart; M. Shibasaki; F. Vogtle, Wiley-VCH, **2000**, 145-160.
[48] Fidanze, S.; Song, F.; Szlosek-Pinaud, M.; Small, P. L. C.; Kishi, Y. *J. Am. Chem. Soc.* **2001**, *123*, 10117-10118.
[49] Song, F.; Fidanze, S.; Benowitz, A. B.; Kishi, Y. *Org. Lett.* **2002**, *4*, 647-650.
[50] Lamers, Y. M. A. W.; Rusu, G.; Wijnberg, J. B. P. A.; De Groot, A. *Tetrahedron* **2003**, *59*, 9361-9369.
[51] Evans, D. A.; Chapman, K. T.; Carreira, E. M. *J. Am. Chem. Soc.* **1988**, *110*, 3560-3578.
[52] Chen, K. -M.; Hardtmann, G. E.; Prusad, K.; Repič, O.; Shapiro, M. J. *Tetrahedron Lett.* **1987**, *28*, 155-158.
[53] Netscher, T.; Scalone, M.; Schmid, R. In *Asymmetric Catalysis on Industrial Scale: Challenges, Approaches and Solutions*, eds. H. U. Blaser; E. Schmidt, Wiley-VCH, Weinheim, Chapter 4, 71-89.
[54] Schmid, R.; Scalone, M. In *Comprehensive Asymmetric Catalysis*, eds. E. N. Jacobsen; A. Pfaltz; H. Yamamoto, Springer, New York, **1999**, 3, 1445-1446.
[55] Schmid, R.; Broger, E. A.; Cereghetti, M.; Crameri, Y.; Foricher, J.; Lalonde, M.; Müller, R. K.; Scalone, M.; Schoettel, G.; Zutter, U. *Pure Appl. Chem.* **1996**, *68*, 131-138.

[56] Bell, S.; Wüstenberg, B.; Kaiser, S.; Menges, F.; Netscher, T.; Pfaltz, A. *Science* **2006**, *311*, 642-644.
[57] Jacobsen, E. N. In *Comprehensive Asymmetric Catalysis*, eds. E. N. Jacobsen; A. Pfaltz; H. Yamamoto, Springer, New York, **1999**, 3, 1475.
[58] Hawkins, J. M.; Watson, T. J. N. *Angew. Chem. Int. Ed.* **2004**, *43*, 3224-3228.

Chapter 2
Total Synthesis of Mycolactones

2.1 Introduction

Buruli ulcer (BU) is a severe necrotizing skin disease, characterized by the formation of large, painless necrotic ulcers in the absence of an acute inflammatory response. It is caused by *Mycobacterium ulcerans*, the third most common mycobacterial pathogen of humans after its more infamous family members *M. tuberculosis* and *M. leprae*. Recently, it has been reported, that *M. ulcerans* secretes a heterogeneous mixture of polyketide toxins known as mycolactones. Clinical evidence shows, that these mycolactone toxins are indeed responsible for tissue destruction and immune suppression.[1]

2.2 Biological context

BU was first reported in 1948 by MacCallum and colleagues,[2] even though large ulcers almost certainly caused by *M. ulcerans* had been described by Sir Albert Cook as early as 1897.[3] The disease primarily occurs in sub-Saharan Africa, but is also known in tropical areas of Papua New Guinea, Malaysia, China, French Guiana, Mexico and Australia among others.[4] BU affects all age groups, but about 50% of the cases concerns children. Unfortunately, the disease has long been neglected, possibly because foci of infection are located in local communities with little economic or political influence.[4] Recently however, the World Health Organization (WHO) has declared the emergence of BU an immediate and serious health hazard. In 1998, this resulted in the Global Buruli Ulcer Initiative (GBUI) aiming to mobilize and coordinate international research as was formulated in the Yamoussoukro Declaration.[5] In 2004, this was followed by the adoption of a resolution on Buruli ulcer calling for additional (scientific) efforts.[6]

2.2.1 Infection

Proximity to marshes and wetlands is a risk factor for infection, but the exact mode of transmission remains unclear.[3] Current evidence indicates that the infection is transmitted through mild traumatic injuries or skin abrasions, after contact with contaminated water, soil or vegetation.[7] The isolation of *M. ulcerans* from the salivary glands of *Naucoris* sp. has incriminated aquatic insects as possible passive hosts serving as the key reservoir species. In support of this hypothesis, mice were infected in the laboratory after insect bites.[8] The involvement of slowly moving or stagnant water for the proliferation of *M. ulcerans* combined with the increased agricultural activities involving irrigation in some of the endemic regions is almost certainly responsible for the steep rise in reported infections over the last decade.[7]

2.2.2 Treatment

At present, complete surgical removal of infected tissue is the only way to effectively treat BU, leaving the patient with extensive scarring or even complete loss of limbs.[3] To date, drug treatment has proven to be ineffective. Even though a number of antibiotics seem to be effective *in vitro*,[9] the results obtained in *in vivo* studies are disappointing.[10] In 2002 however, Dega *et al*. reported an *in vivo* study on mice disclosing that *M. ulcerans* is sensitive to a combination of amikacin and rifampicin.[11] WHO guidelines now recommend the use of these anti-mycobacterial drugs in combination with surgery. Ideally, the development of a vaccine to prevent infection would offer the best structural solution, but attempts in this area have met with limited success so far.[12]

2.2.3 Structure of mycolactones

The increased research activity toward BU led to a first major breakthrough when Small and co-workers isolated and identified mycolactones A and B (Figure 2.1) as the polyketide toxins secreted by *M. ulcerans*.[1,13] Since then, several other mycolactones have been identified (C, D, E and F; Figure 1) all of which show conservation of the lactone core, but modifications in the side chain.[14] Even though the biological effects of mycolactones have been extensively studied and documented as being cytotoxic, apoptotic and immunosuppressive, the mechanisms by which they act remain an enigma. Biological evaluation shows that the lactone core in itself is sufficient for cytopathicity, but that the side chain greatly enhances its virulence factor probably by facilitating entrance into the cell or by interaction with an intracellular target molecule. The mode of action of all mycolactone analogues seems therefore identical, but the potency varies with the composition of the side chain (mycolactones A and B being the most potent). This suggests, that the differences in virulence that are observed between geographical strains of *M. ulcerans* can be directly correlated to their differences in mycolactone profile.[14b]

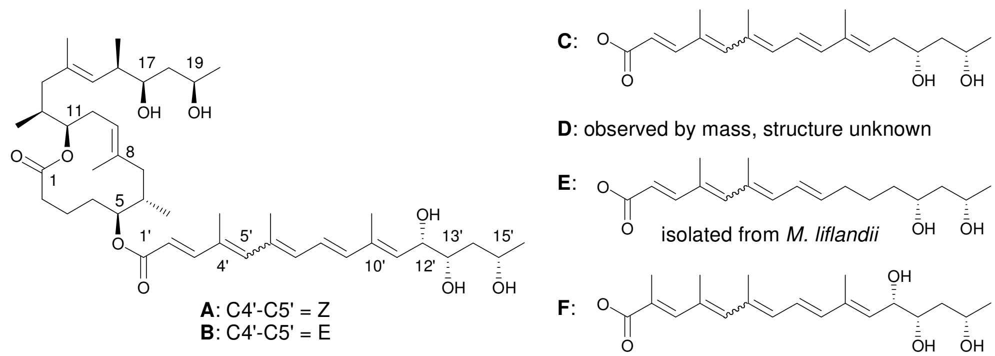


Figure 2.1 *Molecular structures of natural mycolactones*.[a]

2.2.4 Biosynthesis of mycolactones A and B

From the onset of the joint effort against BU, the determination of the complete genome sequence of *M. ulcerans* received high priority. A fundamental understanding of the molecular machinery of the bacterium provides opportunities for rational drug targeting and vaccine design. The sequencing of the entire genome has recently been achieved by Stinear and co-workers.[15] Previously, the genes encoding for the biosynthesis of mycolactones had already been identified.[16] To this end, the DNA from *M. ulcerans* was compared to the DNA from *M. marinum*; a closely related species which does not produce mycolactones.

[a] During the course of my research, it was brought to my attention by Prof. Dr. J. Lugtenburg, that 1,3-steric interactions between the methyl substituents on C4' and C6' of the side chain of mycolactone B (E-configuration) will force the side chain into the 5'-s-*cis* conformation rather than the 5'-s-*trans* conformation which is depicted in figure 2.1. However, for ease of presentation and to avoid unnecessary confusion, I have chosen to follow the literature precedent for the remainder of this chapter.

This approach resulted in the identification of a giant plasmid called pMUM001, most of which consists of six genes coding for proteins involved in mycolactone synthesis.[16] Two polyketide synthases (PKS MLSA1 and MLSA2) are responsible for the synthesis of the mycolactone core, while a third PKS (MLSB) constructs the side chain. The arrangement of the three PKS resembles an 'assembly-line' of enzymes. For the formation of the lactone core by MLSA1 and MLSA2, the growing polyketide chain is extended nine times by a ketosynthase and an acyltransferase adding either a C2- (malonate) or a C3-unit (methylmalonate). The added unit is subsequently reduced to the appropriate degree by enzymes immediately following the acyltransferase in the assembly-line (Figure 2.2). The saturated parts in the chain are obtained by the consecutive action of a ketoreductase, a dehydratase and an enoylreductase (Scheme 2.1). When the enoylreductase is missing an olefin is formed, while a hydroxyl moiety is produced if only a ketoreductase is present.

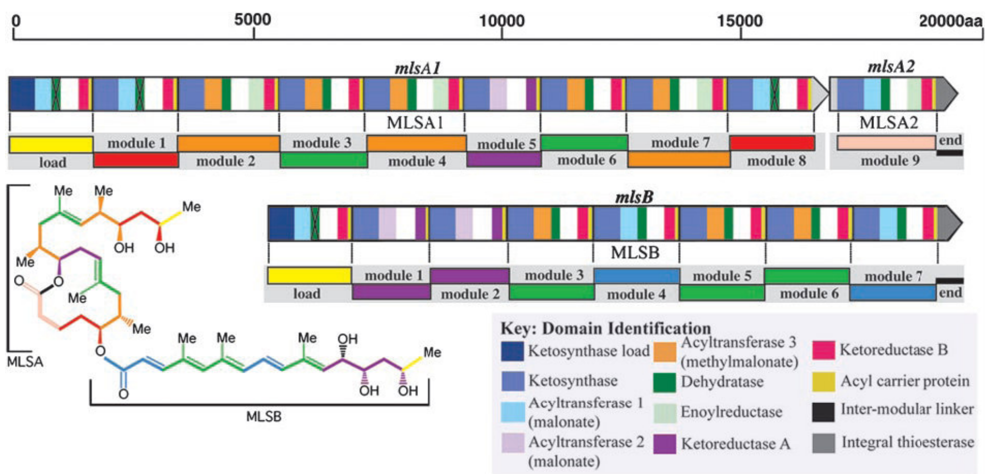
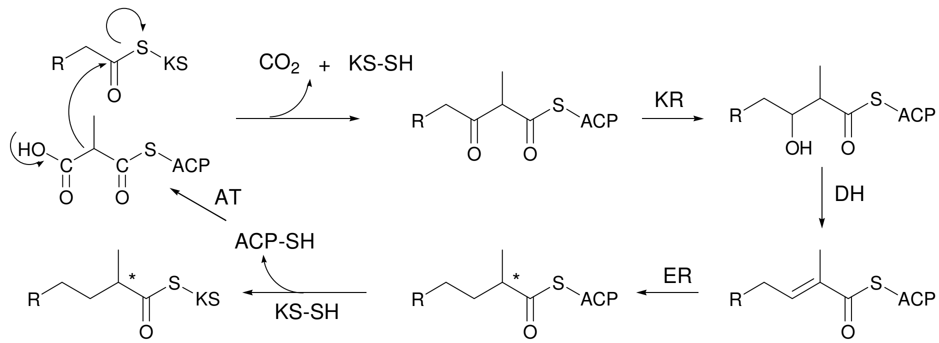


Figure 2.2 *Organization of the mycolactone PKS genes*.[16]

After completion of the chain, a terminal thioesterase/cyclase releases the product as the 12-membered lactone ring. In a similar fashion, MLSB produces the side chain by seven chain extensions. In this case a polyketide-modifying enzyme denoted P450 monooxygenase (also encoded by pMUM001) is believed to introduce the additional hydroxyl moiety at carbon 12. The function of the other two genes on pMUM001 is still unclear, but it is speculated that the protein encoded by one of them catalyzes ester bond formation between the mycolactone core and the side chain.[16]



start of a new cycle

ACP = Acyl carrier protein, KS = Ketosynthase, KR = Ketoreductase, DH = Dehydratase,
ER = Enoylreductase, AT = Acyltransferase, R = growing polyketide chain,

Scheme 2.1 *Example of a chain extension cycle; in casu with a saturated C3-unit.*[17]

Interestingly, the three PKS genes contain a high degree of identical nucleotide sequences. In fact, the entire 105-kb mycolactone locus contains only 9.5 kb of unique DNA sequence and this has important implications. First of all, the relative simplicity of the genome suggests that mycolactones evolved only very recently.[18] Moreover, the enormous size of the plasmid is believed to make it genetically unstable and therefore sensitive to mutation. This hypothesis is supported by the fact that in the absence of purifying selection, deletion of plasmid sequences readily proceeds resulting in loss of mycolactone production.[18] The instability of the plasmid may partly explain why different geographical strains of *M. ulcerans* show a different mycolactone profile (*vide supra*). For example, it has been proven that the extra methyl group in the side chain of mycolactone F (Figure 2.1) arises from the substitution of an acyltransferase 1 domain (malonate) by an acyltransferase 3 domain (methylmalonate) in module 7 of MSLB (Figure 2.2).[14d] Similarly, a mutation in the gene coding for P450 monooxygenase that renders the enzyme inactive would result in the production of mycolactone C instead of A/B. It seems that the circumstances are ideal for the creation of a large family of mycolactones and that the structural diversity that can be found in nature may therefore be much larger than first anticipated.

2.2.5 Production of mycolactones

Access to a wide variety of natural and non-natural mycolactones is essential for a better understanding of the biological mechanisms by which they act and ultimately for the development of better drugs and vaccines. Unfortunately, initial research was hampered by the lack of sufficient material. The bacterium produces its toxins in minute amounts and cultivation of *M. ulcerans* is problematic due to slow growth and formation of aggregates. In 2001, a large scale suspension cultivation process for the isolation of useful amounts of mycolactones was described, but the procedure remained cumbersome and the overall yield was still low.[19] Synthetic organic chemists were therefore called upon to supply larger quantities. In 2002, Kishi and co-workers met this challenge by completion of the total synthesis of mycolactones A and B and more recently they reported the construction of mycolactone C (see also § 2.3).[20] An additional advantage of a (general) synthetic approach is, that it is currently the only method to access non-natural mycolactones as well.

The recent discovery, that pMUM001 encodes all the genes required for the biosynthesis of mycolactones (*vide supra*),[16] may make transformation of this plasmid a possible alternative for the production of natural mycolactones in the future.

2.3 Previous total syntheses of mycolactones

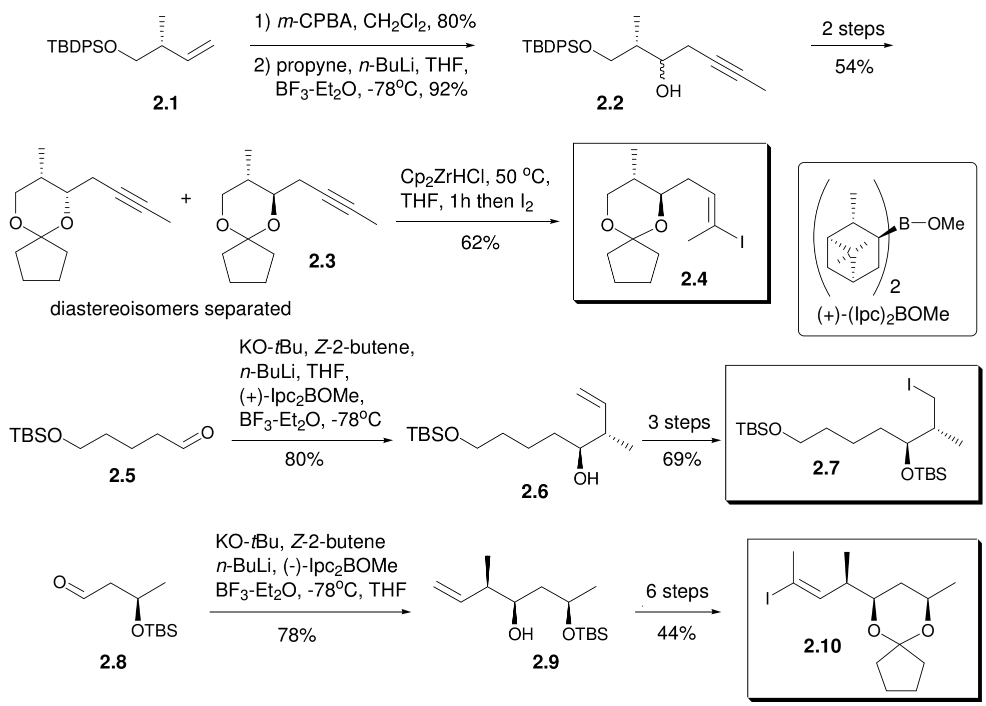
Until today, only Kishi and co-workers have reported a total synthesis of mycolactones.[20] Prior to that, before the elucidation of the stereochemistry by Small and co-workers,[13] Gurjar *et al.* synthesized the side chain, but with the wrong stereochemistry at C15' as appeared later.[21]

2.3.1 Synthesis of the mycolactone core by Kishi

The mycolactone core was assembled by connection of three chiral building blocks (**2.4**, **2.7** and **2.10**; Scheme 2.2).[13a] For the preparation of fragment **2.4** enantiopure olefin **2.1** was converted into the epoxide, which was opened with propyne to give alcohol **2.2** as a mixture of diastereomers. After protective group manipulations, cyclic acetal **2.3** was obtained, which could be separated from its diastereoisomer. The building block was completed by hydrozirconation with Schwartz reagent followed by halogen metal exchange.

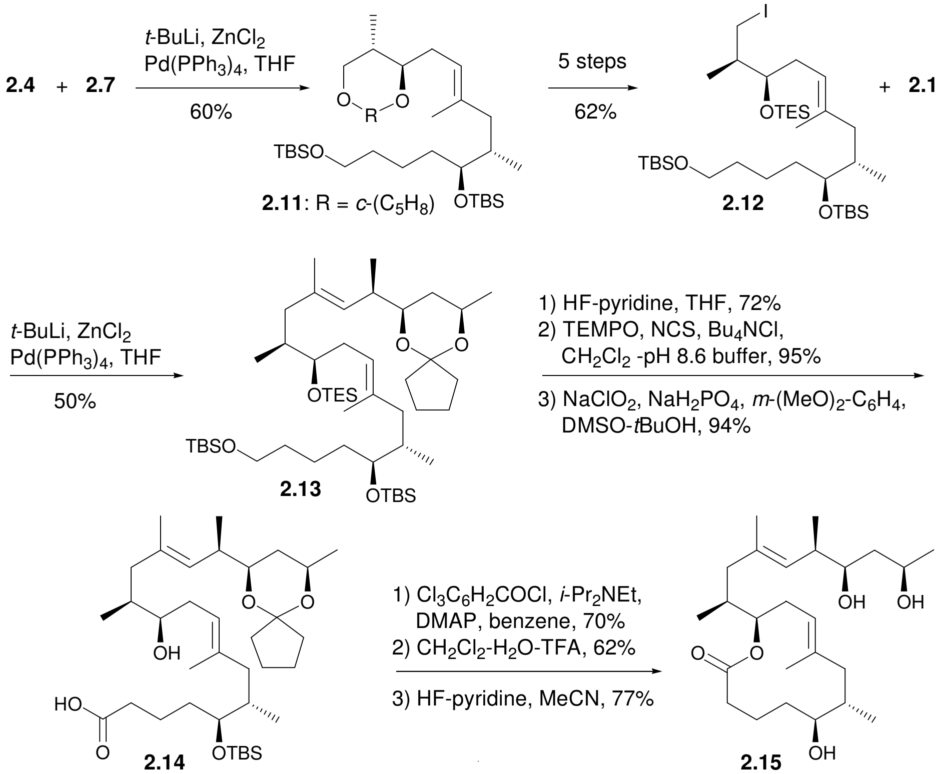
The two stereocenters of fragment **2.7** were installed in a single step by application of the Brown crotylboration protocol[22] to aldehyde **2.5** resulting in terminal olefin **2.6**. Subsequently, **2.7** was obtained in three straightforward steps.

For the introduction of the stereogenic centers of building block **2.10**, the Brown protocol was employed in the key step once again, this time resulting in terminal olefin **2.9**. Appropriate functionalization to obtain vinyl iodine **2.10** was achieved in six additional steps.



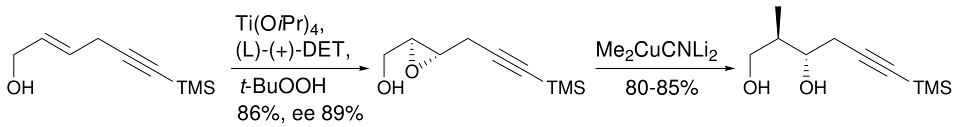
Scheme 2.2 *Synthesis of chiral building blocks for the mycolactone core.*[13a]

The assembly of the mycolactone core from the enantiopure building blocks commenced with the cross-coupling of fragments **2.4** and **2.7** using a modified Negishi coupling protocol (Scheme 2.3).[13b] Product **2.11** was subsequently subjected to a five-step sequence to generate primary iodide **2.12** suitable for connection to **2.10** in a second Negishi coupling. Selective removal of the TES-group and the primary TBS-group of **2.13** was achieved in one pot by treatment with buffered HF·pyridine. Oxidation of the primary alcohol to the corresponding carboxylic acid in two steps then set the stage for a Yamaguchi macrolactonization to form the 12-membered ring. The synthesis of the mycolactone core was completed by deprotection of the remaining hydroxyl moieties to give **2.15**.



Scheme 2.3 *Assembly of the mycolactone core.*[13a]

An important aspect of the synthesis of the mycolactone core as described above, is that it constitutes a general method for the preparation of any desired diastereoisomer. In addition, the convergent approach avoids long linear sequences, which makes the synthesis of analogues relatively straightforward. Nevertheless, there is still room for improvement, for example in the yields of the cross-coupling steps and in reducing the overall number of steps (for instance in the preparation of **2.10**). With regard to the introduction of new chiral centers (i.e. formation of **2.2**, **2.6** and **2.9**), the authors unfortunately fail to mention the obtained levels of diastereoselectivity and/or enantioselectivity. The Brown crotylboration protocol usually gives excellent diastereoselectivity (de > 99%) and high enantioselectivity (ee 90% ± 5%). An indication for the enantiopurity of **2.6** is the preparation of its enantiomer by others in 86% ee using the same method.[23] A definite improvement should be possible in the synthesis of fragment **2.4** where a stereocenter is introduced with low/no selectivity resulting in the loss of a substantial amount of compound two steps later. Alternatively, the stereocenters of this building block can be formed *via* asymmetric Sharpless epoxidation followed by hydroxyl-directed dimethyl cuprate opening of the epoxide as reported by Forsyth *et al*. (Scheme 2.4).[24]

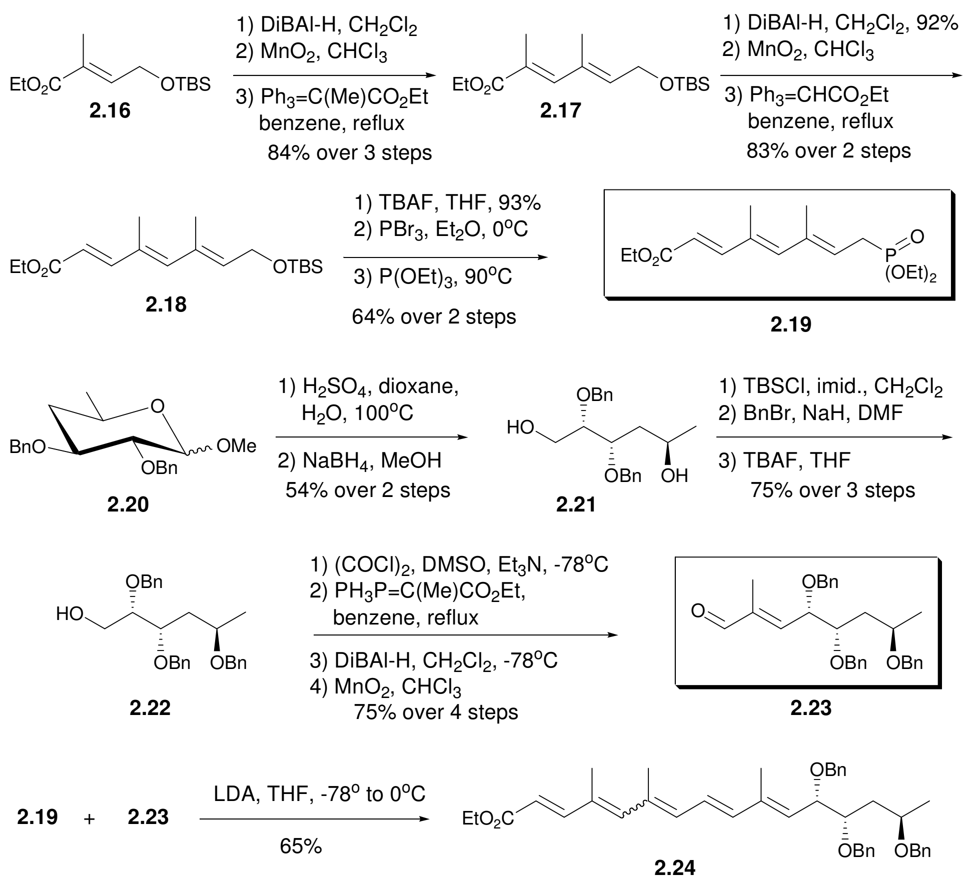


Scheme 2.4 *Alternative preparation of* ***2.4***.[24]

2.3.2 Synthesis of a side chain epimer

Gurjar and Cherian synthesized a C15'-epimer of the side chain of mycolactones A and B by cross-coupling of a conjugated building block (**2.19**: C1'-C8') and a chiral building block (**2.23**: C9'-C16'; Scheme 2.5).[21] The construction of the conjugated fragment started with the reduction of ester **2.16** (available in 3 steps from allyl alcohol) to the corresponding alcohol. Subsequent oxidation to the aldehyde followed by Wittig olefination yielded ester **2.17**. A second reduction-oxidation-Wittig chain elongation sequence resulted in the formation of ester **2.18**. To make the building block suitable for cross-coupling, the allylic alcohol was deprotected and converted into phosphonate **2.19** in two steps.

For the preparation of the chiral fragment, the methyl glycoside bond of 4,6-dideoxy-D-*xylo*-hexopyranoside **2.20** was hydrolyzed and the resulting hemiacetal reduced to give diol **2.21**. By adopting a protection-deprotection protocol, the secondary alcohol was selectively shielded in three steps (**2.22**), after which the primary alcohol was oxidized to the aldehyde. The synthesis of the chiral building block was concluded *via* a Wittig-reduction-oxidation sequence similar to the ones described above. Horner-Wadsworth-Emmons (HWE) coupling of the thus obtained aldehyde **2.23** to phosphonate **2.19** proceeded readily to give the protected side chain **2.24** in 65% yield.

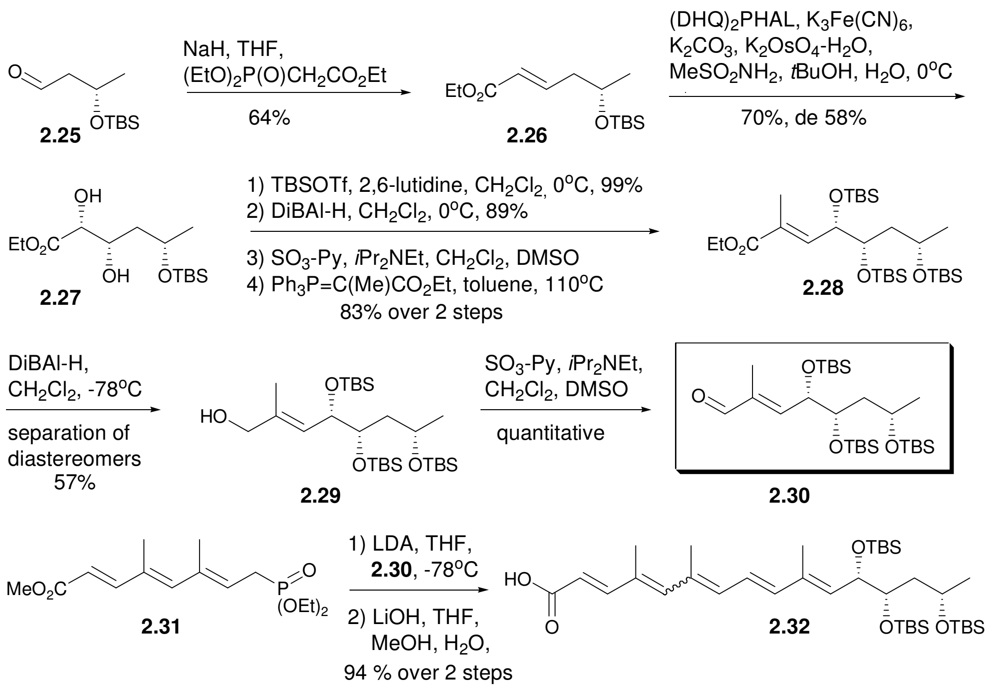


Scheme 2.5 *Synthesis of a C15'-epimer of the side chain.*[21]

2.3.3 Synthesis of the side chain of mycolactones A, B and C

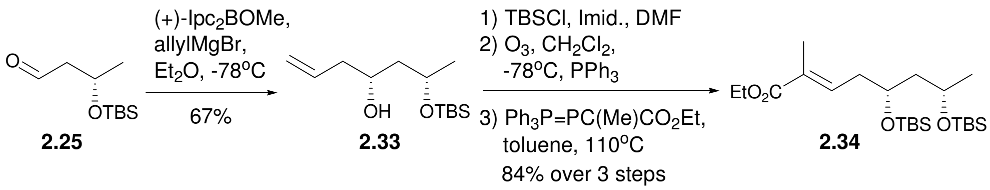
In their synthesis of the side chains of mycolactones A, B and C,[20] Kishi and co-workers adopted the strategy of Gurjar *et al.*[21] to couple a conjugated C1'-C8' fragment to a chiral C9'-C16' fragment. To this end, the methyl ester analogue of conjugated building block **2.19** (**2.31**; Scheme 2.6) was prepared employing similar procedures as described in Scheme 2.5. The synthesis of the chiral building block started from aldehyde **2.25**, which was available from ethyl (*S*)-3-hydroxy-*n*-butyrate in two steps. HWE chain elongation afforded olefin **2.26**, which was subjected to a catalytic asymmetric dihydroxylation to give a 3.8:1 mixture of diol **2.27** and its diastereomer. Protection of the hydroxyl groups followed by a reduction-oxidation-Wittig chain elongation sequence resulted in olefin **2.28**.

Allylic alcohol **2.29** was subsequently obtained by reduction of the ester, at which stage the diastereomers could be separated. Finally, oxidation to the aldehyde afforded conjugated building block **2.30**, which was coupled to phosphonate **2.31** to give carboxylic acid **2.32** after base-mediated hydrolysis.



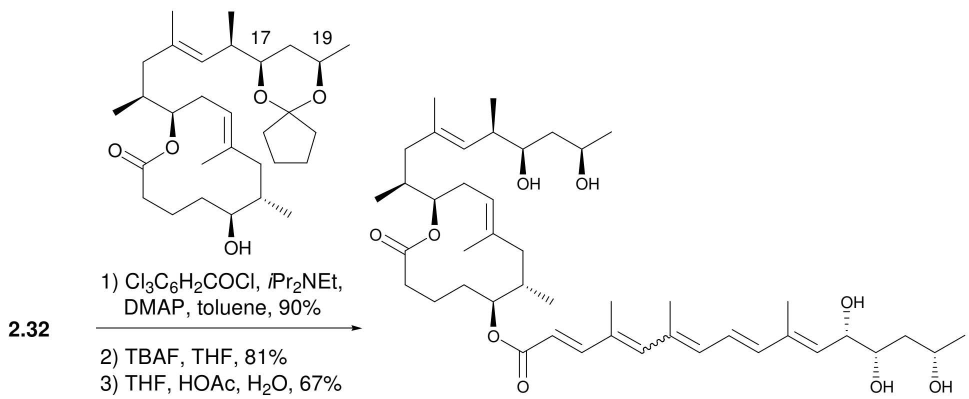
Scheme 2.6 *Synthesis of the side chain of mycolactones A and B.*[20a]

For the synthesis of the side chain of mycolactone C, aldehyde **2.25** was subjected to a Brown allylation protocol to give olefin **2.33** as a single diastereomer.[20b] Protection of the hydroxyl moiety followed by ozonolysis afforded the aldehyde, which was used in a Wittig reaction to obtain ester **2.34** (Scheme 2.7). Beyond this stage, the route established for the side chain of mycolactone A/B was followed (Scheme 2.6).



Scheme 2.7 *Synthesis of the side chain of mycolactone C.*[20b]

The total synthesis of mycolactones A and B was completed *via* a Yamaguchi esterification of the side chain **2.32** with a suitably protected mycolactone core followed by removal of the protecting groups (Scheme 2.8). Because hydrolysis of the cyclopentylidene acetal was accompanied by side reactions on the unsaturated fatty acid moiety, the C17- and C19 hydroxyl groups were protected as TBS-ethers in the total synthesis of mycolactone C.



Scheme 2.8 *Coupling of the side chain and the core unit.*[20a]

2.4 New approaches towards the side chain of mycolactones A and B[25]

The total synthesis of mycolactones A, B and C (*vide supra*) offers new opportunities in the biological and medical field with regard to Buruli ulcer disease (see § 2.2.5). However, even though the synthesis as described in paragraph 2.3 constitutes a general method capable of producing a range of (non-natural) mycolactones, alternative (more efficient) approaches remain a continuing area of research.

2.4.1 Retrosynthesis

For the assembly of the conjugated system, both Kishi and Gurjar employed reduction-oxidation-Wittig sequences that elongate the chain with one double bond at the time (§ 2.3.2). The construction of **2.19** or **2.32** takes nine linear steps from **2.16**, which leaves ample room for improvement. We envisioned the synthesis of a conjugated C1'-C9' system (**2.37** in Figure 2.3) in only 3 steps from 2,4-dimethylfuran. We subsequently planned to connect terminal alkyne **2.37** to chiral C10'-C16' fragment **2.38** *via* a Sonogashira coupling to obtain 8,9-dehydro analogue **2.36**. Finally, partial hydrogenation of the alkyne followed by deprotection should complete the synthesis of the side chain **2.35**.

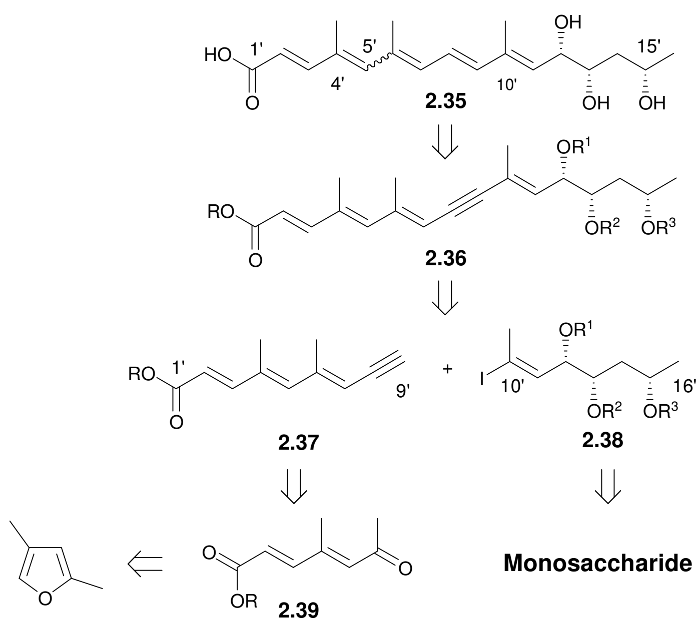


Figure 2.3 *Retrosynthetic analysis.*

With regard to the preparation of the chiral building block, the choice of Kishi to use an asymmetric catalyst in combination with ethyl (*S*)-3-hydroxy-*n*-butyrate (enantiomer also available) can be appreciated, as it gives access to all diastereomers (Scheme 2.6). In contrast, the use of readily available D-monosaccharides admittedly allows variation at C12' and C13', but restricts the configuration at C15' to the non-natural epimer. Preparation of natural C15'-epimers requires the use of L-sugars, which are often rare and expensive

(Figure 2.4). On the other hand, monosaccharides have the advantage that they offer absolute stereocontrol as opposed to the 58% d.e. obtained during the Sharpless dihydroxylation reaction.[20a] Besides, in principle, sugars also provide access to regioisomers with hydroxyl moieties at C14' or C16'.

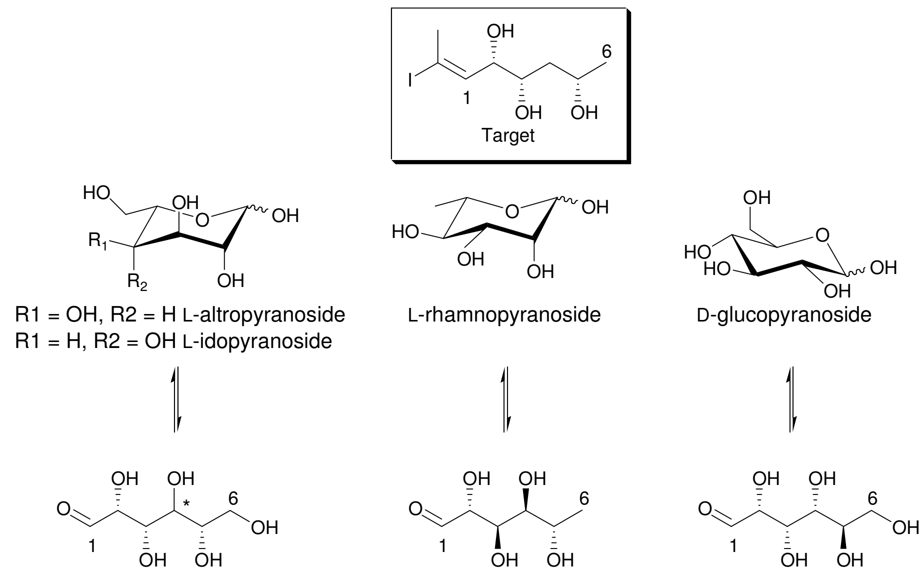
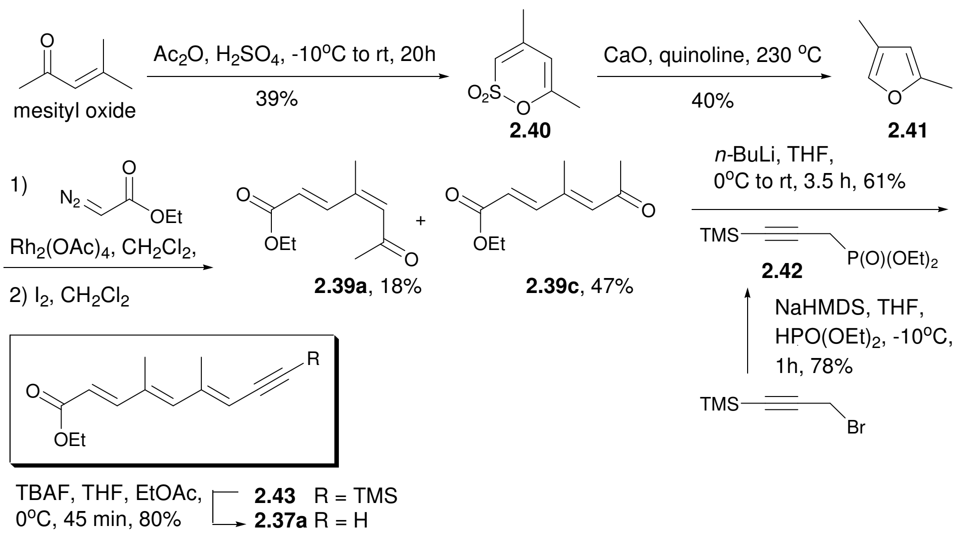


Figure 2.4 *Correlation of stereochemistry between sugars and chiral building blocks.*

In order to benefit from the advantages that monosaccharides offer, while avoiding limitations in the configuration at C15', we attempted to synthesize a chiral building block with the correct stereochemistry starting from cheap monosaccharides by epimerization of a single stereocenter. To this end two routes were explored starting from either D-glucose or L-rhamnose. In case of D-glucose, the C5-position needs to be epimerized and the hydroxyl groups at C4 and C6 need to be removed. L-Rhamnose requires epimerization at C3 and removal of C4-OH (Figure 2.4). After installation of the correct stereochemistry, chain elongation with two carbon atoms and suitable functionalization to obtain vinyl iodine **2.38** was planned to proceed in five steps.

2.4.2 Synthesis of the conjugated building block (2.37)

The preparation of conjugated building block **2.37a** started with the formation of δ-sultone **2.40** (39%) *via* subjection of mesityl oxide to a mixture of acetic anhydride and sulfuric acid (Scheme 2.9). Subsequent pyrolysis in the presence of activated CaO and quinoline afforded 2,4-dimethylfuran (**2.41**) in 40% yield under expulsion of sulfur dioxide.[26]



Scheme 2.9 *Synthesis of the conjugated building block.*

Furan **2.41** was then treated with ethyl diazoacetate (EDA) in the presence of catalytic $Rh_2(OAc)_4$ as described by Wenkert *et al.* to give elaborate conjugated system **2.39** as a mixture of *cis*/*trans* isomers (**2.39a** and **2.39b**) (Figure 2.5).[27] When exposed to iodine, the system equilibrated to a mixture of **2.39a** (18% isolated yield) and the desired all-*trans* **2.39c** (47% isolated yield), which could be separated by column chromatography. Re-exposure of pure **2.39a** to iodine furnished a mixture of **2.39a** and **2.39c** in the same ratio as before.

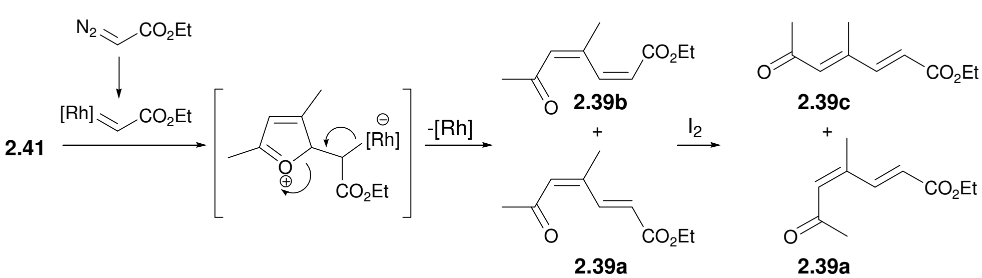
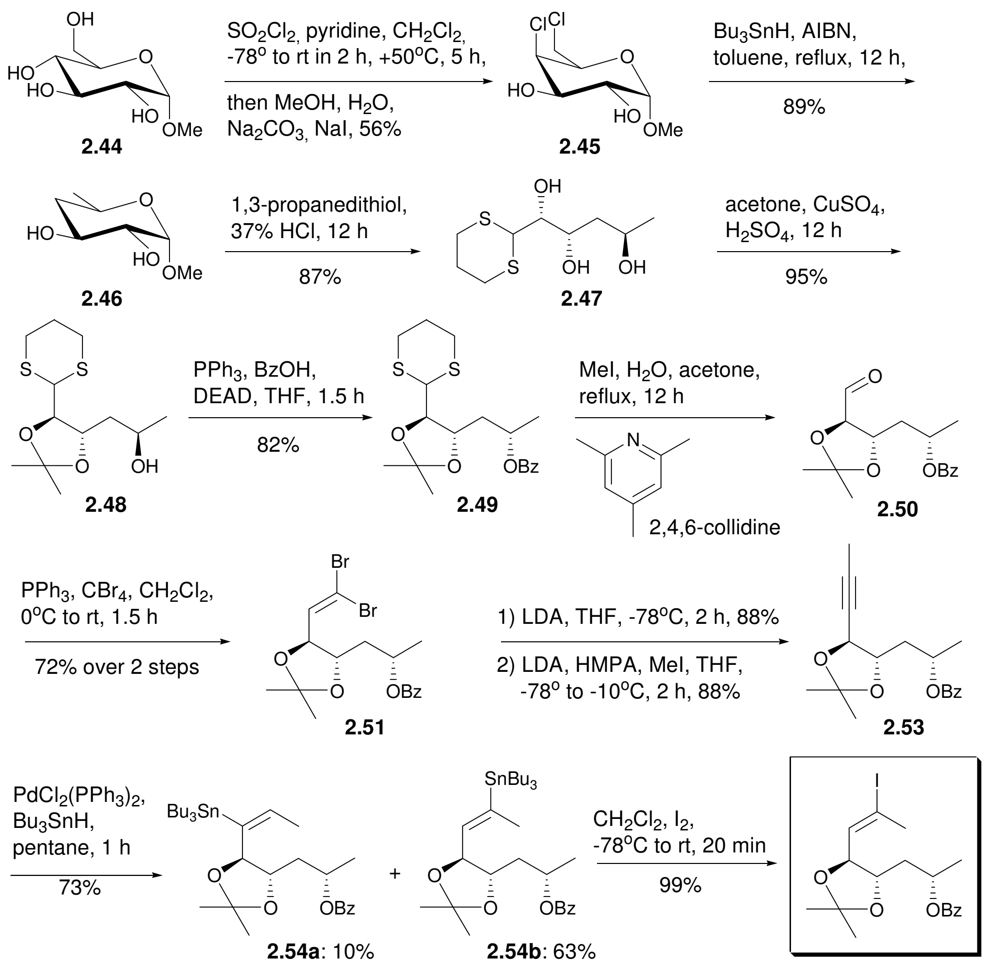


Figure 2.5 *Reaction of EDA with 2,4-dimethylfuran.*

Chain elongation of **2.39c** by a Horner-Wadsworth-Emmons reaction with diethyl (3-trimethylsilyl-2-propynyl) phosphonate (**2.42**)[28] proceeded readily to give **2.43** in 61% yield as a mixture of *cis*/*trans* isomers that could not be separated.[29] Finally, cleavage of the TMS-group with TBAF resulted in the unstable terminal alkyne **2.37a** (80%).

2.4.3 Synthesis of the chiral building block from D-glucose (2.38)

As explained in paragraph 2.4.1, installation of the correct stereochemistry from D-glucose requires reduction of C4-OH and C6-OH and epimerization at C5 (Figure 2.4). To this end, a six step procedure developed by Redlich *et al.* for the synthesis of bicyclic acetals was adopted.[30] In the event, D-glucose was converted into methyl α-D-glucopyranoside (**2.44**) by refluxing in anhydrous methanol in the presence of Dowex 50W (H^+).[31] Reaction of **2.44** with sulfuryl chloride in pyridine followed by dechlorosulfation in the presence of sodium iodide resulted in dichloro compound **2.45** (56%) with exclusively the galacto-configuration as described by Jones *et al.* (Scheme 2.10)[32] Alternatively, treatment of **2.44** with triphenylphosphine, tetrachloromethane and imidazole in acetronitrile/pyridine (1:1 v/v) at 50 °C gave **2.45** as well, but conversion was often incomplete and separation from side products was tedious.[33] Radical reduction of **2.45** with tri-*n*-butyltin hydride in the presence of a catalytic amount of AIBN (α,α'-azoisobutyronitrile) afforded 4,6-dideoxy-hexopyranoside **2.46** in 89% yield. Subsequent deprotection of the anomeric center and opening of the resulting hemi-acetal was achieved in one pot by subjection to 1,3-propanedithiol in 37% HCl to give dithioacetal **2.47** in 87% yield. After regioselective protection of C2-OH and C3-OH as their acetonide by reaction with acetone under acidic conditions (**2.48**, 95%), epimerization at C5 was accomplished *via* a Mitsunobu reaction with benzoic acid (**2.49**, 82%). The mass balance in this reaction was completed by the C4-C5 olefin resulting from dehydration (16%). Comparison of the ^{13}C-NMR-spectra of **2.49** (t, 39.5 ppm) with its C5-epimer (t, 40.5 ppm) obtained from the reaction of **2.48** with benzoyl chloride showed that there was only one diastereoisomer present.



Scheme 2.10 *Synthesis of chiral building block **2.38a** from D-glucose.*

With the stereochemistry in place, the next objective was to elongate the chain of **2.49** in order to make it suitable for coupling. Deprotection of the aldehyde proved not to be straightforward as the dithioacetal was resistant to mercury salts[34] and low yields were obtained with NBS in acetone and water.[35] Eventually, treatment with MeI and 2,4,6-collidine in a refluxing mixture of acetone and water gave aldehyde **2.50** (no epimerization at C2 according to ^{1}H-NMR), which was used without further purification after drying in vacuo over P_2O_5.[36] Reaction of **2.50** with CBr_4 and PPh_3 under Corey-Fuchs conditions gave dibromo-olefin **2.51** in 72% yield over 2 steps.[37,38] Elimination of HBr by treatment with LDA followed by a protic work-up resulted in the terminal alkyne **2.52** (88%),[39] which was converted into the methylated alkyne **2.53** (88%) by reaction with LDA and MeI in the presence of HMPA.[40] The direct synthesis of **2.53** from **2.51** by treatment with MeI and *n*-BuLi or *t*-BuLi was unsuccessful, due to the intolerance of the benzoyl group to these conditions.

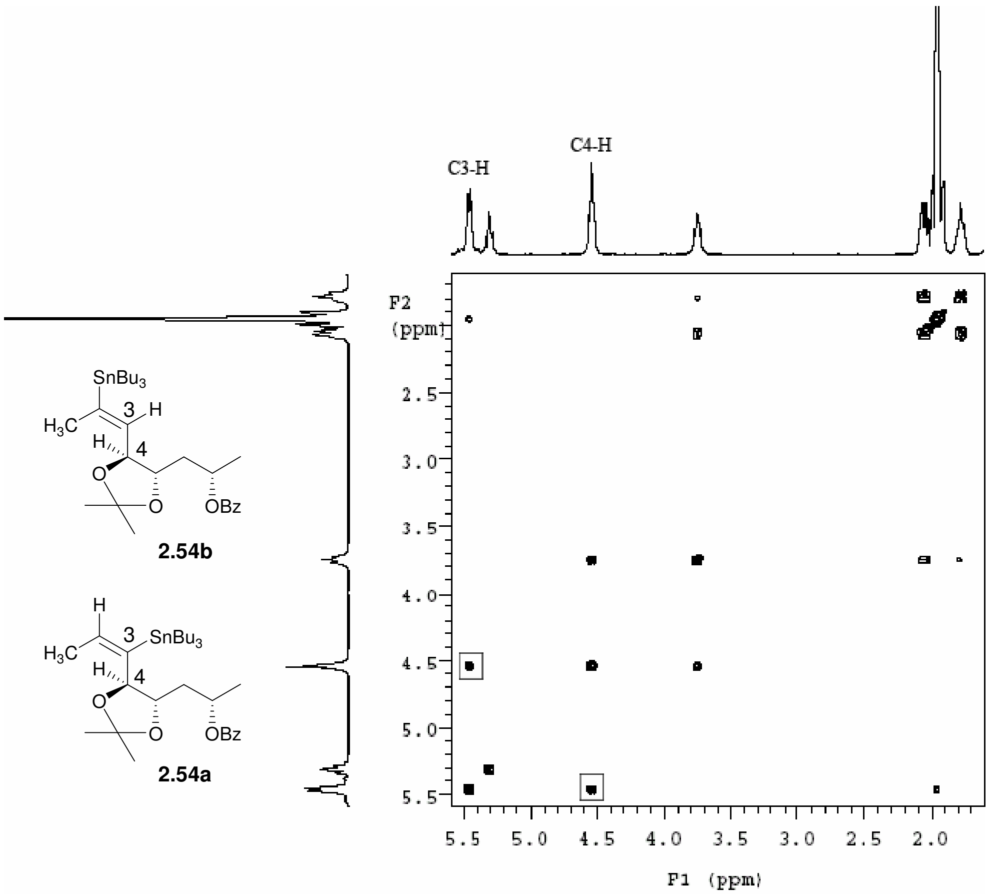


Figure 2.6 *COSY-spectrum of **2.54b** showing interaction between C3-H and C4-H.*

Palladium-catalyzed hydrostannation of the internal alkyne did not go to completion, due to palladium-black formation.[41] Increasing the catalyst loading resulted in full conversion, but was accompanied by additional side-product formation. Fortunately, changing the solvent from THF to *n*-pentane prevented Pd-black formation improving the yield of **2.54** from 63% to 73%.[42] Moreover, the regioselectivity of the reaction was superior in *n*-pentane enhancing the ratio of the terminal (**2.54b**) to internal (**2.54a**) hydrostannation product from 2.6 : 1 in THF to 6.3 : 1 in *n*-pentane. Assignment of **2.54a** and **2.54b** was based on the difference in ^{1}J-value of the vinylic CH_3 with the vinylic proton (J = 6.6 Hz for **2.54a** and J = 1.6 Hz for **2.54b**) and on the COSY-interaction between C3-H and C4-H in **2.54b** (Figure 2.6). Proof that **2.54b** was indeed the *trans*-isomer, was obtained by NOESY-NMR, which showed a clear interaction between C4-H and C1-H3 (Figure 2.7). The synthesis of the chiral moiety was completed by exchange of the tri-*n*-butyltin moiety in **2.54b** with iodine to give **2.38a** (99%).[41]

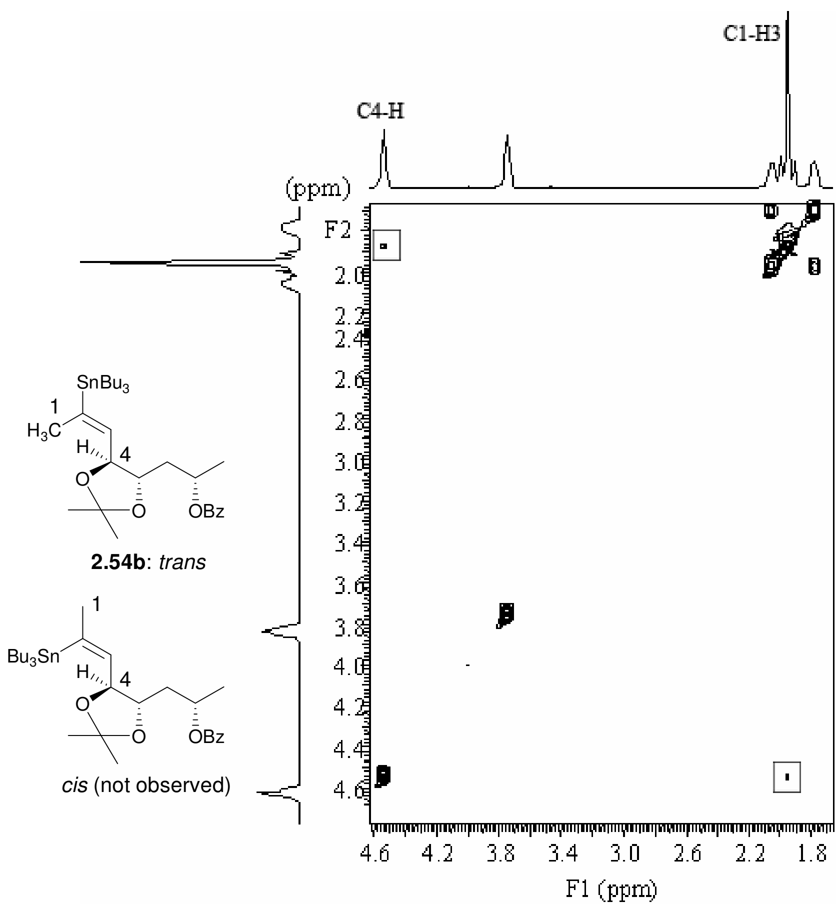
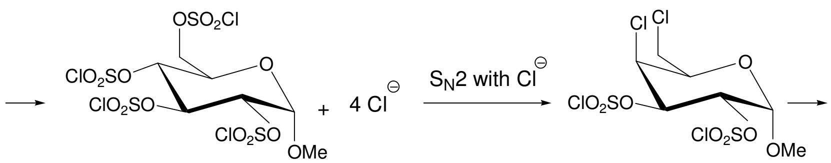


Figure 2.7 *NOESY-spectrum of* ***2.54b*** *showing interaction between C1-H_3 and C4-H.*

The route depicted in Scheme 2.10 constitutes a general procedure for making analogues of **2.38** with any desired stereochemistry starting from different 4,6-dideoxy-D-hexopyranosides (**2.46**-analogues). However, it should be noted, that the conversion of methyl α-D-glucopyranoside (**2.44**) into 4,6-dichloro-compound **2.45** without protection of the C2-OH and C3-OH is not generally applicable to monosaccharides of any configuration. This can be explained by looking at the mechanism in more detail (Figure 2.8).

2.44



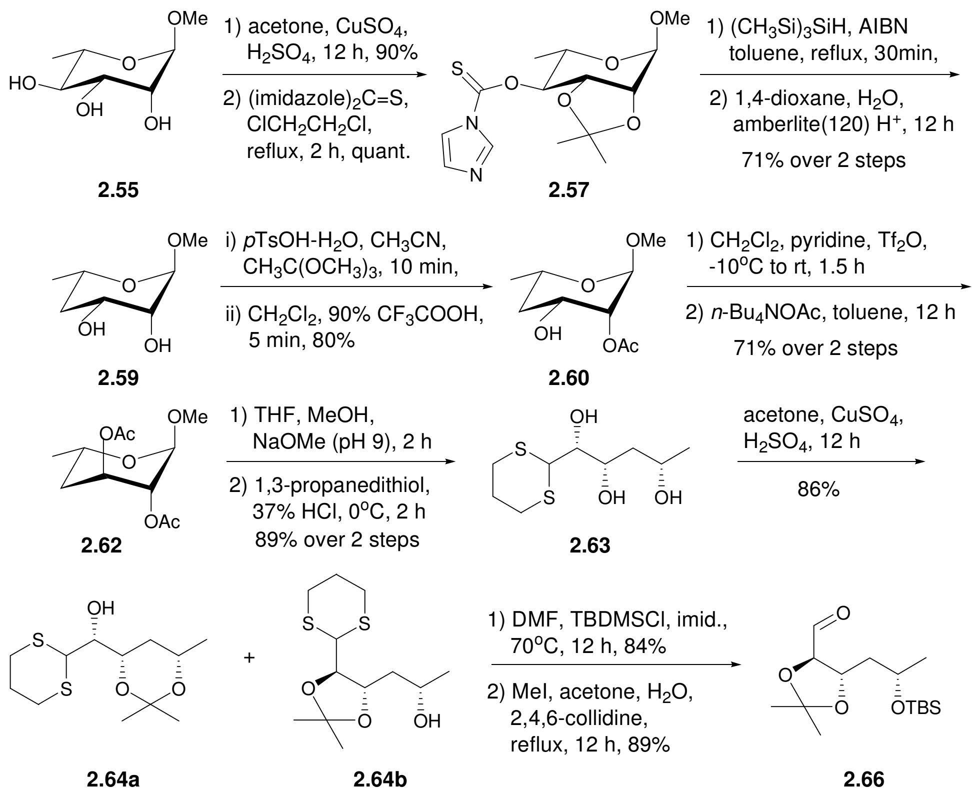
2.45

Figure 2.8 *Formation of chlorodeoxy sugars.*

The process involves initial formation of a tetrachlorosulfate intermediate, followed by displacement of specific chlorosulfates by chloride ions liberated during the chlorosulfation. Whether the S_N2-displacement occurs at a particular center or not, depends on both steric and electronic factors. The primary chlorosulfate at the six-position is the least hindered and is therefore always the first to undergo substitution. The C2-chlorosulfate on the other hand has shown almost complete inertness in every case even when the steric environment is comparable to C4. This is usually explained by the proximity of two *β*-oxygen atoms (compared to one for C4), which are known to considerably retard the rate of nucleophilic substitution.[43,44] The reactivity at the C3-position depends on the anomeric configuration and on neighboring substituents. In case of **2.44**, the C3-position is unreactive because the axial anomeric methoxy group obstructs axial approach of a chloride ion at the C3-position. In concordance, methyl β-D-glucopyranoside gives indeed a mixture of the 3,6- (major) and 4,6-dichloro (minor) compounds.[45] However, formation of the 3,4,6-trichloro-compound is not observed, possibly because a neighboring axial substituent leads to unfavorable interactions in the transition state due to the near-eclipsed conformation of the large substituents on C3 and C4.[43] Analogously, substitution of an equatorial chlorosulfate at C4 is impaired by an axial substituent at C2 or C3, which precludes the synthesis of a C12'- or a C13'-epimer of the side chain employing this particular reduction strategy. This is supported by the observation, that α-D-mannopyranoside gives in fact a 6-monochloro derivative.[43] Alternative routes to obtain 4,6-dideoxy-D-hexopyranosides are therefore required to make the strategy in Scheme 2.10 completely general.

2.4.4 Synthesis of a chiral building block from L-rhamnose

In order to obtain a chiral fragment with the correct stereochemistry from L-rhamnose, reduction of C4-OH and epimerization at C3 is required (Figure 2.4). The synthesis started by protection of the anomeric center *via* acid-catalyzed reaction with methanol to give methyl α-L-rhamnopyranoside (**2.55**; Scheme 2.11). Subsequently, the C2- and C3-hydroxyl moieties were regioselectively protected as their acetonide by reaction with acetone under acidic conditions leading to **2.56** (90%).[46] The remaining free C4-OH was then reacted with 1,1'-thiocarbonyldiimidazole to give the activated precursor for a Barton-MacCombie deoxygenation (**2.57**) in quantitative yield.[47,48] Subsequent radical reduction of **2.57** with tris(trimethylsilyl)silane in the presence of AIBN afforded **2.58** (not shown).[39,49] Cleavage of the acetonide was first attempted with trifluoroacetic acid (55% over 2 steps), but the results with the milder amberlite H^+-resin proved to be superior giving diol **2.59** in 71% over 2 steps.[50] Selective protection of the C2-OH was then achieved by reaction of **2.59** with trimethyl orthoacetate followed by partial hydrolysis of the resulting orthoester leading to the formation of monoacetate **2.60** (80%).[48] It should be noted that the regioselectivity in the hydrolysis step strongly depends on the choice of solvent. In acetonitrile a 3:1 mixture of C2:C3 O-acetylated regioisomers was obtained, while in dichloromethane only the desired C2 O-acetylated product was observed. Subsequent epimerization of the C3-center proved not to be straightforward. Mitsunobu conditions gave only very low conversions (<10%) and the alternative procedure comprising formation of a triflate (**2.61**; not shown) followed by a S_N2-displacement with tetraethylammonium acetate gave no conversion at all. However, changing to tetra-*n-butyl*ammonium acetate led to the isolation of **2.62** in a gratifying 71% yield, with only a minor amount (8%) of side product resulting from dehydration of the C3-C4 bond.[51] This remarkable improvement is most likely due to the superior phase-transfer properties of the tetra-*n*-butyl salt, which can act as a co-solvent when used in large excess (11 eq. in our case). Comparison of the ^1H- and ^{13}C-NMR spectra of **2.62** with the acetylated product of **2.60** proved that epimerization had indeed taken place. An attempt was made to deprotect the hydroxyl moieties and to form the dithioacetal in one pot by stirring **2.62** in HCl (37% aq.) in the presence of 1,3-propanedithiol, but only 21% of **2.63** was isolated. The acetyl groups were therefore first removed under mildly basic conditions (pH 9), after which ring opening proceeded very well, providing **2.63** in 89% yield over two steps. Acetonide protection of two hydroxyl moieties by acid-catalyzed reaction with acetone gave two regioisomers; **2.64a** (8%) and the desired **2.64b** (78%), which were separated by column chromatography.[30] After protection of the remaining C5-OH of **2.64b** with TBDMSCl (**2.65**, 84%),[52] the dithioacetal was deprotected as before to give aldehyde **2.66** in 89% yield. Unfortunately, the TBDMS-ether was not stable under Corey-Fuchs conditions giving a complex mixture of products. Obviously, the target molecule **2.38a** can be synthesized from **2.64b** as described above for D-glucose if a benzoyl ester is chosen as a protecting group.



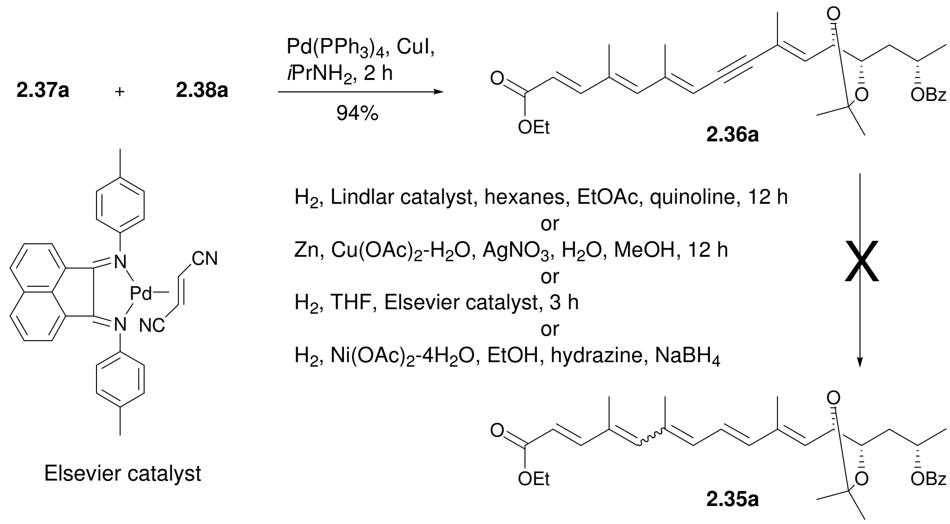
Scheme 2.11 *Chiral building block from L-Rhamnose.*

For the preparation of **2.38a**, the route starting from D-glucose seems more attractive, because it is much more cost-effective and concise; less steps are required and the overall yield is higher. Nevertheless, in light of the fact that preparation of the C13'-epimer of the side chain from D-monosaccharides requires a different (more elaborate) reduction strategy (§ 2.4.3), the route from L-rhamnose may be useful, since the C13'-epimer can be easily accessed by omission of the epimerization steps.

2.4.5 Coupling and partial hydrogenation

Sonogashira coupling of terminal alkyne **2.37a** to vinyl iodine **2.38a** proceeded readily, resulting in the isolation of **2.36a** in an excellent 94% yield (Scheme 2.12). It was anticipated that partial *cis*-hydrogenation of the internal alkyne of 8,9-dehydro analogue **2.36a** would lead to **2.35a** after isomerization to its all *trans* configuration.[53,29] Disappointingly, partial hydrogenation of the internal alkyne to the alkene did not show sufficient selectivity to be useful on a preparative scale. Lindlar catalyst ($Pd/CaCO_3/Pb$) was typically unreactive regardless of the solvent, temperature, catalyst loading, and/or hydrogen pressure.[54,55] Only at 65 bar of hydrogen some conversion was observed, but with a complete lack of selectivity leading to overreduction. An effort was made with a homogeneous palladium catalyst developed by Elsevier for selective alkyne hydrogenation, but once again no conversion was seen at one bar of hydrogen pressure.[56] Ni-catalyzed reduction with $NaBH_4$ on the other hand proved to be too active and gave only overreduced products.[57] Finally, we turned to the Zn(Cu/Ag)-reduction method in aqueous MeOH as developed by Boland and co-workers.[58] This method is known to selectively *cis*-hydrogenate triple bonds which are embedded in a conjugated system and has been successfully used by Nakanishi among others.[29,59] However, in our case Zn(Cu/Ag)-reduction resulted in a mixture of (over)reduced products containing only traces of a compound with the correct mass (as observed with GC-MS). Even though the desired product was likely to be in the mixture, this could not be confirmed by isolation and full characterization. In any case, the lack of selectivity in the partial reduction of the alkyne precludes this strategy as a viable synthetic pathway. As it is known that free hydroxyl moieties occasionally aid the selectivity of Boland-reductions, the isopropylidene-moiety of

2.36a was removed by treatment with acetic acid and water, but this did not have a beneficial effect on the outcome of the partial reduction step.



Scheme 2.12 *Coupling of the building blocks and partial hydrogenation/reduction.*

In light of the difficulties we have encountered in the semi-hydrogenation of **2.36a**, I believe that there is currently no general method for the selective partial reduction of alkynes embedded in an alkene system while in conjugation with an electron withdrawing moiety. In this respect it should be noted, that for example in the elegant total synthesis of 6,7-dehydrostipiamide, the final reduction to stipiamide itself is also not reported (Figure 2.9).[60]

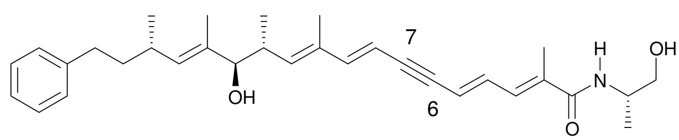
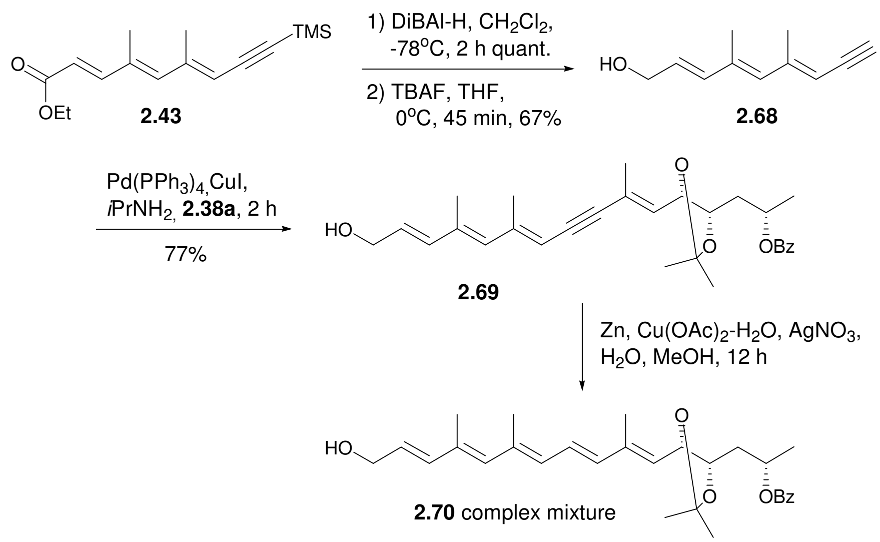


Figure 2.9 *Structure of 6,7-dehydrostipiamide.*

Only the Lindlar catalyst has been used with varying success in the past, but the results are highly substrate-dependent. In some cases good selectivity is observed, but in other examples the catalyst is inactive or the reaction has to be stopped after consumption of a certain amount of hydrogen to avoid overreduction.[61] To the best of my knowledge, the partial reduction of alkynes in conjugation with an ester using Zn(Cu/Ag) has not been reported. It was therefore decided to synthesize **2.69** having a primary alcohol instead of an ester (Scheme 2.13).

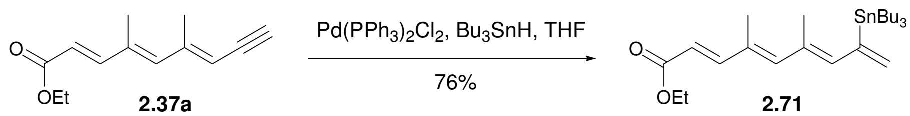


Scheme 2.13 *Alternative approach: removing the conjugated electron withdrawing group.*

Unfortunately, Boland-reduction of **2.69** resulted in a complex mixture from which only a small amount of a compound with the correct mass (GC-MS and EI- and CI-MS) was isolated. Whether this was indeed the desired product **2.70** was unclear and in any event the yield of the reaction was too low to continue with this approach.

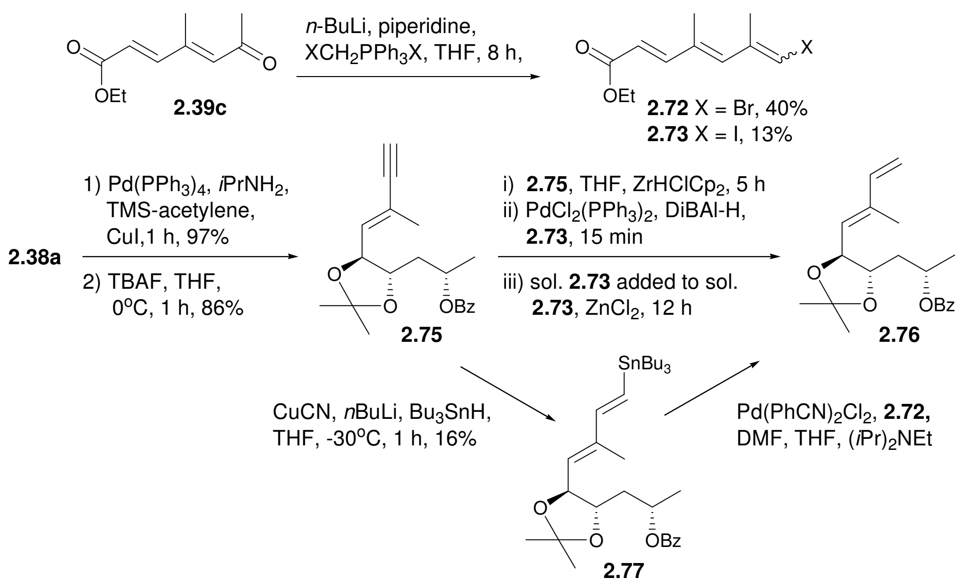
2.4.6 Alternative strategies

As an alternative approach, an attempt was made to functionalize terminal alkyne **2.37a** to obtain an olefin suitable for palladium catalyzed sp^2-sp^2 coupling. In my hands, however, **2.37a** was unreactive towards stannylcupration (Bu_3SnH, CuCN, *n*-BuLi),[62] and hydrozirconation (Schwartz reagent),[63] while palladium-catalyzed hydrostannation resulted exclusively in the undesired internal regioisomer (**2.71**, Scheme 2.14).[64] Even though it is known that palladium-catalyzed hydrostannation of terminal alkynes in direct conjugation with an ester predominately gives the α-addition product,[41,65] it was surprising to find that this also holds true when the ester and terminal alkyne are separated by three double bonds.



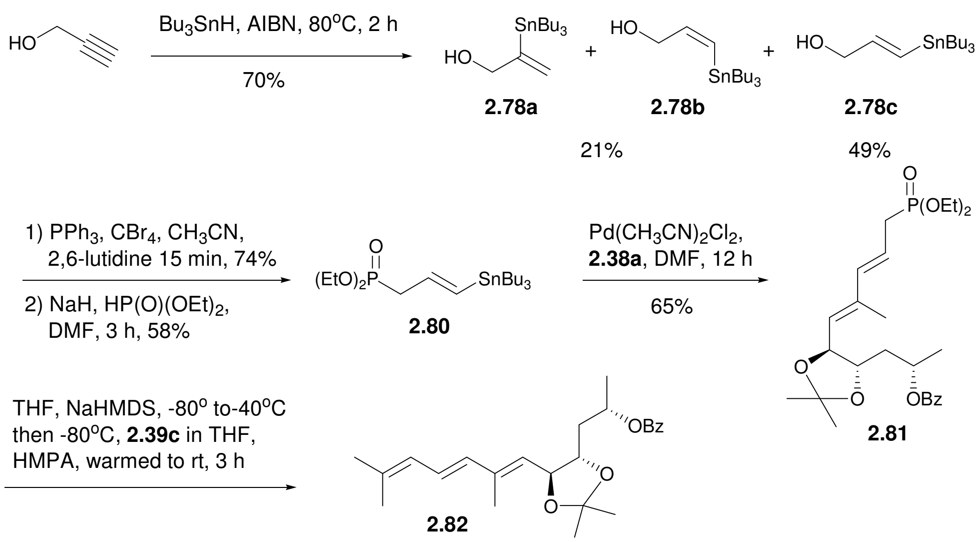
Scheme 2.14 *Hydrostannation of **2.37a**.*

In a second attempt, the point of connection of the building blocks was changed from C9'-C10' to C7'-C8' (see Figure 2.3). This strategy implied that the conjugated building block needed to be one double bond shorter (i.e. **2.72** and **2.73**), while the chiral building block required elongation by two carbon atoms (i.e. **2.75**; Scheme 2.15). The first objective was met by reacting **2.39c** in a Wittig reaction with $BrCH_2PPh_3Br$ (**2.72**, 40% isolated yield) or its iodine analogue (**2.73**, 13% isolated yield).[66] The second goal was realized by Sonogashira coupling of **2.38a** with trimethylsilylacetylene (97%),[52] giving the free alkyne **2.75** (86%) after deprotection with TBAF. Subsequent Negishi coupling of **2.75** to **2.73** resulted in the isolation of terminal alkene **2.76** and starting material **2.73** suggesting that the initial hydrozirconation took place, but that either transmetallation with Zn was unsuccessful or that the catalytic cycle was blocked at some stage.[67,68,69] Similarly, hydrostannation of **2.75** using a stannylcuprate gave **2.77** in low yield (16%),[57a] but Stille coupling to **2.72** afforded alkene **2.76** and the staring vinyl halide once again.[70]



Scheme 2.15 *Alternative coupling strategies (1).*

Finally, an effort was made to couple the building blocks *via* formation of the C6'-C7'-bond using a Horner-Wadsworth-Emmons reaction on conjugated system **2.39c** (Scheme 2.16). To this end, propargyl alcohol was subjected to a radical hydrostannylation resulting in a mixture of isomers from which the desired *trans*-isomer **2.78c** could be isolated by column chromatography in 49% yield.[71] Treatment of **2.78c** with triphenylphosphine and carbon tetrabromide in the presence of 2,6-lutidine afforded the corresponding allylic bromide (**2.79**, 74%), which was converted into diethyl phosphonate **2.80** (58%) by reaction with diethylphosphite and NaH.[72] Subsequent Stille coupling of **2.80** to **2.38a** furnished phosphonate **2.81** in a moderate 65% yield. Regrettably, HWE-reaction with conjugated ketone **2.39c** did not give the desired product, but only side product **2.82** resulting from reaction with acetone, which was most likely formed *in situ* by cleavage of part of the acetonide. The opposite approach to first do a HWE-coupling of **2.80** to ketone **2.39c** followed by a Stille coupling to **2.38a** gave no conversion in the first step.



Scheme 2.16 *Alternative coupling strategies (2).*

2.5 Summary and concluding remarks

In this chapter, new approaches towards the synthesis of natural and non-natural mycolactones were described. In particular, the development of an alternative route to construct the side chain of mycolactones A and B (and analogues thereof) was discussed. It was shown, that the application of monosaccharides is a viable alternative to asymmetric catalysis for the synthesis of the chiral part (**2.38a** and **2.66**) of the side chain. Compound **2.38a** was prepared in twelve steps from D-glucose in 12% yield and with complete stereocontrol (Scheme 2.10). In comparison, Kishi and co-workers synthesized a similar building block (**2.30**) in ten steps from (*S*)-3-hydroxy-*n*-butyrate in 18% yield (Scheme 2.6). In their case, the stereoselective introduction of the hydroxyl moieties at C12' and C13' was accomplished with moderate diastereoselectivity (58% de) using a Sharpless dihydroxylation reaction, after which the undesired diastereomer was removed at a later stage of the synthesis.

Due to the vast number of different, readily available, monosaccharides, the new route is in principle suitable for the synthesis of a wide range of analogues. However, manipulation of monosaccharides to obtain linear enantiopure building blocks containing the structural motif of the desired analogue is not always straightforward. More specifically, new ways have to be explored for the reductive removal of the appropriate hydroxyl moieties (depending on the target).

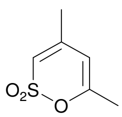
In addition, an efficient route for the rapid assembly of conjugated building block **2.37a** from 2,4-dimethylfuran in 3 steps was described. Unfortunately, employment of **2.37a** is currently obstructed by the lack of an efficient method for the partial reduction of the 8,9-dehydro analogue **2.36**, which was obtained by Sonogashira coupling to **2.38a**. Alternative cross-coupling methods for the direct synthesis of (protected) **2.35** failed so far to give the desired product.

2.6 Experimental section

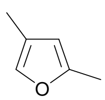
General remarks: reagents were purchased from Aldrich, Acros Chimica, Merck or Fluka and were used as received unless otherwise stated. All solvents were reagent grade and were dried and distilled before use according to standard procedures, after which they were stored under argon. Chromatography: silica gel, Merck type 9385 230-400 mesh, TLC: silica gel 60, Merck, 0.25 mm. Components were visualized by staining with a) $KMnO_4$, b) a mixture of phosphomolybdic acid (25 g), cerium (IV) sulfate (7.5 g), H_2O (500 mL) and H_2SO_4 (25 mL) or c) a mixture of *p*-methoxybenzaldehyde (2.1 mL), AcOH

(1.8 mL), H_2SO_4 (6.3 mL) and EtOH (170 mL). Optical rotations were measured on a Perkin-Elmer 241 or 241 MC polarimeter with a 10 cm cell (*c* given in g/100 mL). Mass spectra (HRMS) were recorded on an AEI MS-902. ^{1}H- and ^{13}C-NMR spectra were recorded on a Varian Gemini-200 (50.32 MHz), a Varian VXR300 (75.48 MHz) or a Varian AMX400 (100.59 MHz) spectrometer in $CDCl_3$. Chemical shift values are denoted in δ values (ppm) relative to residual solvent peaks ($CHCl_3$, ^{1}H $\delta = 7.26$, ^{13}C $\delta = 76.9$). Carbon assignments are based on APT ^{13}C experiments.

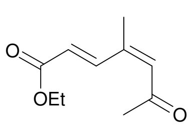
4,6-Dimethyl-[1,2]oxathiine 2,2-dioxide (2.40):[26] Concentrated sulfuric acid (72.3 mL, 1.36 mmol) was added over 1 h at -10 °C to acetic anhydride (300 mL, 3.19 mmol, 2.35 eq), after which mesityl oxide (156 mL, 1.36 mmol) was added over 1 h at the same temperature. The resulting dark red solution was stirred overnight at -5 °C and then for 8 more h at room temperature. The mixture was then poured into ice-water upon which a solid precipitated. Filtration and drying in vacuo gave **2.40** (85 g, 0.53 mmol, 39%) as a sand-colored solid. ^{1}H-NMR ($CDCl_3$, 300 MHz) $\delta = 2.04$ (s, 3H), 2.15 (s, 3H), 5.61 (s, 1H), 6.26 (s, 1H) ppm. Mp: 70 °C, (lit.[26a] 70.5-71.0 °C).



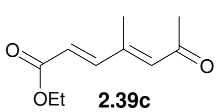
2,4-Dimethylfuran (2.41):[26] CaO was activated by heating it overnight at a 1000°C using a 'moffeloven'. Freshly ignited CaO (17.5 g, 312 mmol), quinoline (1.0 mL, 8.4 mmol) and **2.40** (17.5 g, 109 mmol) were thoroughly mixed in a three-necked flask. The flask was equiped with a destillation device, which was connected to an erlenmeyer containing aqueous KOH (3.3 M, 37 mL) at 0 °C. Upon heating of the three-necked flask to 230 °C a yellow liquid was transferred into the erlenmeyer. The organic layer was separated from the aqueous layer and washed with water, diluted aq. H_2SO_4 (1.0 M) and aq. $NaHCO_3$ (sat.), dried ($MgSO_4$), filtered and purified by distillation (bp 94 °C, lit.[26b] 94.5-95.0) to give **2.41** (4.15 g, 43.2 mmol, 39.6%) as a colorless liquid. ^{1}H-NMR ($CDCl_3$, 300 MHz) $\delta = 1.97$ (s, 3H), 2.24 (s, 3H), 5.83 (s, 1H), 7.03 (s, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) $\delta = 9.6$ (q), 13.3 (q), 108.1 (d), 120.5 (s), 137.1 (d), 152.0 (s) ppm.



Ethyl (2E,4Z)-4-methyl-6-oxohepta-2,4-dienoate (2.39a) and Ethyl (2E,4E)-4-methyl-6-oxohepta-2,4-dienoate (2.39c):[27] $Rh_2(OAc)_4$ (10 mg, 23 µmol), and 2,4-dimethylfuran (**2.41**, 1.0 g, 10.4 mmol, 2.0 eq) were dissolved in dichloromethane (14.3 mL) under argon and a solution of ethyl diazoacetate (0.55 mL, 5.2 mmol) in dichloromethane (3.6 mL) was slowly added over 10 h employing a syringe pump. The resulting solution was stirred for another 5 h at which point the catalyst was removed by filtration over a Florisil column. The green solution was then concentrated and the residue was taken up in dichloromethane (14.3 mL) and stirred overnight under argon in the presence of a catalytic amount of I_2. The resulting black solution was washed with aq.



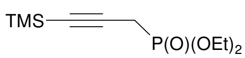
2.39a



2.39c

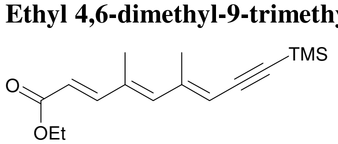
$Na_2S_2O_3$ (10% w/w) and brine (sat.), dried (Na_2SO_4) and concentrated. The product was purified by column chromatography (*n*-hexane-EtOAc 9:1) to give **2.39a** (0.17 g, 0.93 mmol, 18%) and the all-*trans*-isomer **2.39c** (0.44 g, 2.4 mmol, 47%). The latter was a yellow liquid which became crystalline upon standing at 4 ^oC. When **2.39a** was treated with I_2 in dichloromethane, a mixture of **2.39a** and **2.39c** in the same ratio as before was formed.
^{1}H-NMR **2.39a** ($CDCl_3$, 300 MHz) δ = 1.30 (t, *J* = 7.2 Hz, 3H, CH_3CH_2O), 2.01 (s, 3H, CH_3), 2.25 (s, 3H, C7-H), 4.23 (q, *J* = 7.2 Hz, 2H, CH_3CH_2O), 6.17 (d, *J* = 15.9 Hz, 1H, C2-H), 6.27 (s, 1H, C5-H), 8.39 (d, *J* = 16.2 Hz, 1H, C3-H) ppm.
^{1}H-NMR **2.39c** ($CDCl_3$, 300 MHz) δ = 1.30 (t, *J* = 7.2 Hz, 3H, CH_3CH_2O), 2.22 (s, 3H, CH_3), 2.26 (s, 3H, C7-H), 4.23 (q, *J* = 7.2 Hz, 2H, CH_3CH_2O), 6.24 (d, *J* = 15.6 Hz, 1H, C2-H), 6.36 (s, 1H, C5-H), 7.24 (d, *J* = 15.6 Hz, 1H, C3-H) ppm. ^{13}C-NMR **2.39c** ($CDCl_3$, 50.3 MHz) δ = 13.6 (q), 14.1 (q) 32.0 (q), 60.7 (t), 124.2 (d), 131.8 (d), 146.7 (s), 147.3 (d), 166.2 (s), 198.9 (s) ppm. MS(EI) for $C_{10}H_{14}O_3$: m/z = 182 [M^+], HRMS calcd for $C_{10}H_{14}O_3$: 182.094, found: 182.095.

(3-Trimethylsilanyl-prop-2-ynyl)-phosphonic acid diethyl ester (2.42):[28] To a solution



of NaHMDS (1.0 M in THF , 26 mL, 26 mmol) at -10 ^oC was added diethyl phosphonate (3.4 mL, 26 mmol) in THF (8.0 mL) under argon. This solution was stirred for 15 min and then treated with (3-bromo-prop-1-ynyl)-trimethylsilane (3.7 mL, 26 mmol) in THF (8.0 mL) maintaining the temperature at -10 ^oC. After stirring for 1 h, the reaction mixture was quenched with water and the aqueous layer extracted with EtOAc (2x). The combined organic layers were washed with aq. HCl (2.0 M) and water, dried (Na_2SO_4) and concentrated. The product was purified by column chromatography (*n*-pentane-EtOAc 4:1 to 1:1) giving **2.42** (5.1 g, 20 mmol, 78%) as a colorless liquid. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.14 (s, 9H, TMS), 1.34 (t, 6H, CH_3CH_2O, *J* = 7.2 Hz), 2.80 (d, 2H, CH_2, *J* = 22.2 Hz), 4.18 (q, 4H, CH_3CH_2O) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = -0.3 (q), 16.3 (q, $J_{C\text{-}P}$ = 6.1 Hz), 17.8 + 20.6 (t, $J_{C\text{-}P}$ = 145 Hz), 62.9 (t, $J_{C\text{-}P}$ = 6.9 Hz), 94.4 (s) ppm.

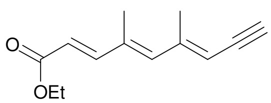
Ethyl 4,6-dimethyl-9-trimethylsilanyl-nona-2,4,6-trien-8-ynoate (2.43): 2.42 (1.0 g, 4.0



mmol, 2.0 eq) was dissolved in THF (20 mL) and *n*-BuLi (1.6 M in *n*-hexane, 2.5 mL, 4.0 mmol) was added at 0 ^oC under argon. After stirring for 30 min of which the last 10 min were at room temperature, the solution had turned dark red. At this point, a solution of **2.39c** (367 mg, 2.01 mmol) in THF (8.0 mL) was added upon which the color slowly changed to brown. The reaction mixture was stirred for 3 h and then quenched with aq. NH_4Cl (sat.). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried (Na_2SO_4) and concentrated. **2.43** (336 mg, 1.22 mmol, 61%, mixture of *cis*/*trans* isomers with a ratio of approximately 4:1) was isolated as a yellow solid after column chromatography (*n*-hexane-EtOAc 98:2 to 95:5 to 4:1). ^{1}H-NMR **major isomer** ($CDCl_3$, 500 MHz) δ = 0.22 (s, 9H, TMS), 1.30 (t, *J* = 7.0 Hz, 3H, CH_3CH_2O), 2.00 (s, 3H, C6-CH_3), 2.14 (s, 3H, C4-CH_3), 4.21 (q, 2H, CH_3CH_2O), 5.59 (s, 1H, C7-H), 5.91 (d, *J* = 15.5 Hz, 1H, C2-H), 6.28 (s, 1H, C5-H), 7.32 (d, *J* = 15.5 Hz, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = -0.1 (q), 13.9 (q), 14.2 (q), 19.7 (q), 60.2 (t), 103.0 (s), 103.5 (s), 113.2 (d), 117.8 (d), 134.5 (s), 140.3 (d), 147.3 (s), 149.7 (d), 167.1 (s) ppm. ^{1}H-NMR **minor isomer** ($CDCl_3$, 500 MHz) δ

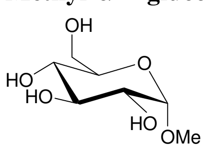
= 0.18 (s, 9H, TMS), 1.31 (t, 3H, CH_3CH_2O), 1.95 (s, 3H, C6-CH_3), 2.04 (s, 3H, C4-CH_3), 4.21 (q, 2H, CH_3CH_2O), 5.53 (s, 1H, C7-H), 5.94 (d, *J* = 14.5 Hz, 1H, C2-H), 6.68 (s, 1H, C5-H), 7.39 (d, *J* = 15.5 Hz, 1H, C3-H) ppm. MS(EI) for $C_{16}H_{24}O_2Si$: m/z = 276 [M^+], HRMS calcd for $C_{16}H_{24}O_2Si$: 276.155, found: 276.155.

Ethyl 4,6-dimethylnona-2,4,6-trien-8-ynoate (2.37a): TBAF (1.0 M in THF, 1.45 mL, 1.45 mmol, 4.0 eq) was stirred for 30 min under argon in the presence of EtOAc (47 µl, 0.48 mmol). The solution was cooled to 0 °C and **2.43** (100 mg, 0.36 mmol) in dry THF (1.8 mL) was added. The mixture was stirred for 45 min at 0 °C and then quenched with aq. NH_4Cl (sat.), dried (Na_2SO_4) and concentrated. **2.37a** (59 mg, 0.29 mmol, 80%) was isolated after column chromatography (*n*-pentane-EtOAc 95:5) as a colorless oil, which turned brown within 15 min. The product was therefore immediately used in the next step. ^{1}H-NMR **major isomer** ($CDCl_3$, 200 MHz) δ = 1.29 (t, *J* = 7.0 Hz, 3H, CH_3CH_2O), 1.99 (d, *J* = 0.8 Hz, 3H, C6-CH_3), 2.13 (s, 3H, C4-CH_3), 3.38 (d, *J* = 2.4 Hz, 1H, C9-H), 4.21 (q, 2H, CH_3CH_2O), 5.54 (s, 1H, C7-H), 5.91 (d, *J* = 15.6 Hz, 1H, C2-H), 6.28 (s, 1H, C5-H), 7.31 (d, *J* = 15.6 Hz, 1H, C3-H) ppm.

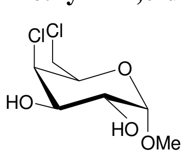


^{1}H-NMR **minor isomer** ($CDCl_3$, 200 MHz) δ = 1.30 (t, *J* = 7.0 Hz, 3H, CH_3CH_2O), 1.95 (d, *J* = 1.0 Hz, 3H, C6-CH_3), 2.04 (s, 3H, C4-CH_3), 3.22 (d, *J* = 2.4 Hz, 1H, C9-H), 4.21 (q, 2H, CH_3CH_2O), 5.49 (s, 1H, C7-H), 5.94 (d, *J* = 15.6 Hz, 1H, C2-H), 6.71 (s, 1H, C5-H), 7.39 (d, *J* = 16.0 Hz, 1H, C3-H) ppm. MS(EI) for $C_{13}H_{16}O_2$: m/z = 204 [M^+], HRMS calcd for $C_{13}H_{16}O_2$: 204.115, found: 204.116.

Methyl α-D-glucopyranoside (2.44):[31] Dowex 50W (*p*TsOH) was stirred in HCl (2.0 M aq) for 2 h and then filtrated and washed with HCl (2.0 M aq), water and MeOH. A suspension of the thus obtained resin (38 g) and D-glucose (42.0 g, 233 mmol) in MeOH (600 mL) was heated under reflux overnight. After cooling to room temperature, the resin was filtered off and the filtrate concentrated to give methyl D-glucopyranoside (α:β = 62:38) in quantitative yield. Pure **2.44** (23.5 g, 122 mmol, 52.2%) was obtained as a white solid after crystallization from ethanol. ^{13}C-NMR (DMSO, 50.3 MHz) δ = 54.3 (q), 61.0 (t), 70.3 (d), 72.0 (d), 72.6 (d), 73.4 (d), 99.7 (d) ppm.

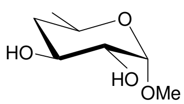


Methyl 4,6-dideoxy-4,6-dichloro-α-D-galactopyranoside (2.45):[32] Methyl α-D-glucopyranoside (**2.44**) (42.5 g, 218 mmol) was dissolved in pyridine-chloroform (430 mL, 1:1 v/v) and sulfuryl chloride (142 mL, 236 g, 1.75 mol, 8eq.) was added dropwise at -78 °C under argon. The resulting yellow solution was stirred for 2 h while slowly warming to room temperature. Subsequently the solution was heated to 50 °C and stirred for another 5 h. After cooling to room temperature the solution was diluted with MeOH and water, neutralized with $Na_2CO_3 \cdot 10H_2O$ and quenched with a NaI-solution (16 g in 40 mL water-MeOH, 1:1 v/v). The resulting solution was concentrated in vacuo by co-evaporation with toluene and purified by continuous liquid-liquid extraction from water with chloroform. Concentration in vacuo gave a brown/red solid which was further purified by crystallization from chloroform to give white crystals (28.1 g, 122 mmol, 55.8%). ^{1}H-NMR



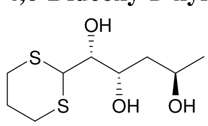
($CDCl_3$, 300 MHz) δ = 2.20 (br s, 2H, C2-OH, C3-OH), 3.48 (s, 3H, C1-OCH_3), 3.68 (d, J = 6.6 Hz, 2H, C6-H), 3.85 (dd, J = 3.6, 9.6 Hz, 1H, C2-H), 3.99 (dd, J = 3.6, 9.6 Hz, 1H, C3-H), 4.14 (t, J = 6.6 Hz, 1H, C5-H), 4.53 (d, J = 3.3 Hz, 1H, C4-H), 4.86 (d, J = 3.6 Hz, 1H, C1-H) ppm. ^{13}C-NMR (DMSO, 50.3 MHz) δ = 44.4 (t), 54.7 (q), 64.9 (d), 67.6 (d), 67.7 (d), 69.3 (d), 99.9 (d) ppm.

Methyl 4,6-dideoxy-α-D-xylo-hexopyranoside (2.46):[30] Bu_3SnH (30 mL, 32 g, 0.11 mol)



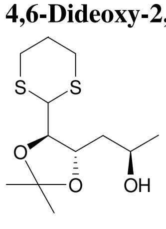
was added dropwise to a solution of **2.45** (5.3 g, 23 mmol) in refluxing dry toluene (180 mL) under argon and AIBN (0.10 g, 0.61 mmol, 2.6 mol%) was added. The resulting solution was heated under reflux overnight and then cooled to room temperature and concentrated in vacuo. The obtained oil was diluted with CH_3CN and washed with *n*-hexanes (2x). Concentration gave a solid which was dissolved in water and washed with Et_2O (3x). Concentration of the aqueous layer afforded a white solid which was further purified by column chromatography (dichloromethane-MeOH 19:1 to 9:1) to give pure **2.46** (3.3 g, 20 mmol, 89%). When the reaction was performed on larger scale, purification was done by crystallization from chloroform and *n*-pentane. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.21 (d, J = 6.3 Hz, 3H, C6-H_3), 1.36 (q, 1H, C4-H), 1.98 (ddd, J = 2.4, 5.1, 12.9 Hz, 1H, C4-H'), 2.05 (d, J = 10.2 Hz, 1H, OH), 2.46 (d, J = 2.1 Hz, 1H, OH), 3.33-3.44 (m, 1H, C2-H), 3.41 (s, 3H, C1-OCH_3), 3.77-3.95 (m, 2H, C3-H, C5-H), 4.75 (d, J = 3.6 Hz, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.7 (q) , 39.6 (t), 55.1 (q), 64.0 (d), 68.9 (d), 74.3 (d), 99.7 (d) ppm.

4,6-Dideoxy-D-xylo-hexose-trimethylen-dithioacetal (2.47):[30] 1,3-propanedithiol (6.8



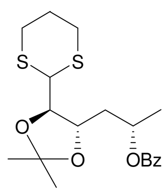
mL, 7.1 g, 65 mmol) was added to a solution of **2.46** (6.0 g, 37 mmol) in 37% aq. HCl (68 mL) and stirred overnight. The solution was then neutralized with 25% aq. ammonia and concentrated in vacuo. The resulting white solid was stirred in acetone for 1 h after which the suspension was filtered and the filtrate concentrated. Purification by column chromatography (dichloromethane-MeOH 9:1) gave **2.47** (7.7 g, 32 mmol, 87%) as a white solid. Alternatively **2.47** could be purified by crystallization from dichloromethane-MeOH. $[\alpha]_D$ -30.3 (c 1.09 in MeOH), (lit.,[9] $[\alpha]_D^{20}$ -29.5 (c 1.00 in MeOH)). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.27 (d, J = 6.3 Hz, 3H, C6-H_3), 1.60 (ddd, J = 3.3, 8.4, 14.7 Hz, 1H, C4-H), 1.89 (m, 1H, C4-H'), 2.03 (m, 2H, dithian-H), 2.70 (m, 2H, dithian-H), 2.92 (m, 2H, dithian-H), 3.74 (d, J = 8.1 Hz, 1H, C2-H), 4.03 (d, J = 8.1 Hz, 1H, C3-H), 4.17 (m, 1H, C5-H), 4.31 (d, J = 9.6 Hz, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 23.6 (q), 25.3 (t) , 26.8 (t), 27.4 (t), 42.3 (t), 47.6 (d), 65.3 (d), 67.7 (d), 73.3 (d) ppm. MS(EI) for $C_9H_{18}O_3S_2$: m/z = 238 [M^+], HRMS calcd for $C_9H_{18}O_3S_2$: 238.070, found: 238.069.

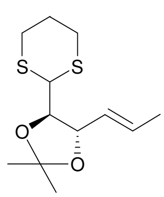
4,6-Dideoxy-2,3-O-isopropyliden-D-xylo-hexose-trimethylen-dithioacetal (2.48):[30] **2.47**



(2.2 g, 9.2 mmol) was dissolved in dry acetone (30 mL) and $CuSO_4$ (2.9 g, 18.5 mmol, 2 eq) and H_2SO_4 (0.14 mL, 0.78 mmol, 8.4 mol%) were added. The resulting green suspension was stirred overnight and then filtered. The filtrate was neutralized with 25% aq. ammonia and the resulting blue suspension was filtered again. The filtrate was concentrated, suspended in brine (sat.) and extracted with dichloromethane (3x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. Purification by column chromatography (*n*-hexane-EtOAc 4:1) gave **2.48** (2.4 g, 8.7 mmol, 95%) as a colorless oil. $[\alpha]_D$ -55.0 (c 1.04 in MeOH), (lit.,[9] $[\alpha]_D^{20}$ -64.8 (c 1.17 in MeOH)). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.24 (d, *J* = 6.0 Hz, 3H, C6-H_3), 1.44 (m, 6H, CMe_2), 1.71-2.17 (m, 4H, C4-H,H', dithian-H), 2.31 (br s, 1H, OH), 2.77-2.96 (m, 4H, dithian-H), 3.95 (dd, *J* = 5.7, 7.8 Hz, 1H, C2-H), 4.08 (m, 1H, C5-H), 4.13 (d, *J* = 5.4 Hz, 1H, C1-H), 4.34 (ddd, *J* = 3.3, 7.8, 7.8 Hz, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 23.5 (q), 25.6 (t), 26.7 (q), 27.3 (q), 29.2 (t), 29.5 (t), 41.3 (t), 48.0 (d), 65.1 (d), 76.1 (d), 82.2 (d), 109.5 (s) ppm. MS(EI) for $C_{12}H_{22}O_3S_2$: m/z = 278 [M^+], HRMS calcd for $C_{12}H_{22}O_3S_2$: 278.101, found: 278.101.

5-O-Benzoyl-4,6-dideoxy-2,3-O-isopropyliden-L-arabino-hexose-trimethylen-dithioacetal (2.49):[30] Triphenylphosphine (13 g, 50 mmol) and benzoic acid (5.8 g, 48 mmol) were added to a solution of **2.48** (6.2 g, 22 mmol) in dry THF (115 mL) at room temperature. Subsequently, a solution of diethylazodicarboxylate (DEAD, 8.4 g, 48 mmol) in dry THF (30 mL) was added over 20 min. The resulting mixture was stirred for 1 h and then quenched with MeOH and concentrated in vacuo. Water was added to the resulting oil and the product was extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. **2.49** (7.0 g, 18 mmol, 82%) was obtained as a yellow oil after purification by column chromatography (*n*-hexane-EtOAc 9:1 to 4:1). The remaining 18% was isolated as a yellow oil, which appeared to be the side product resulting from C4-C5 dehydration according to ^{1}H-NMR. ^{1}H-NMR **2.49** ($CDCl_3$, 200 MHz) δ = 1.39-1.44 (m, 9H, C6-H_3, CMe_2), 1.82-2.20 (m, 4H, C4-H,H', dithian-H), 2.67-2.97 (m, 4H. dithian-H), 3.96 (dd, *J* = 5.2, 7.6 Hz, 1H, C2-H), 4.12 (d, *J* = 5.2 Hz, 1H, C1-H), 4.26 (ddd, *J* = 4.4, 7.6, 7.6 Hz, 1H, C3-H), 5.38 (m, 1H, C5-H) 7.38-7.60 (m, 3H, Bz-H), 8.03-8.09 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 19.8 (q), 25.6 (t), 26.7 (q), 27.3 (q), 29.1 (t), 29.5 (t), 39.5 (t), 48.2 (d), 69.0 (d), 75.5 (d), 82.6 (d), 109.6 (s), 128.1 (d), 129.5 (d), 130.6 (s), 132.6 (d), 165.9 (s) ppm. MS(EI) for $C_{19}H_{26}O_4S_2$: m/z = 282 [M^+], HRMS calcd for $C_{19}H_{26}O_4S_2$: 282.127, found: 282.126.

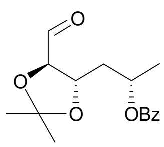




^{1}H-NMR side product ($CDCl_3$, 200 MHz) δ = 1.39-1.49 (m, 6H, CMe_2), 1.74 (dd, *J* = 1.8, 6.6 Hz, 3H, C6-H_3), 1.84-2.17 (m, 2H, dithian-H), 2.74-3.03 (m, 4H, dithian-H), 3.94 (dd, *J* = 4.2, 8.0 Hz, 1H, C2-H), 4.10 (d, *J* = 4.2 Hz, 1H, C1-H), 4.47 (t, *J* = 8.0 Hz, 1H, C3-H), 5.47 (m, 1H), 5.87 (m, 1H) ppm.

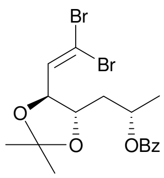
The C5-epimer of **2.49** was synthesized by reaction of **2.48** with benzoyl chloride in pyridine. ^{1}H-NMR C5-epimer of **2.49** ($CDCl_3$, 300 MHz) δ = 1.39-1.43 (m, 9H, C6-H_3, CMe_2), 1.82-2.25 (m, 4H, C4-H,H', dithian-H), 2.74-2.98 (m, 4H, dithian-H), 3.93 (dd, *J* = 5.7, 7.5 Hz, 1H, C2-H), 4.11 (d, *J* = 5.4 Hz, 1H, C1-H), 4.24 (ddd, *J* = 2.1, 8.3, 8.3 Hz, 1H, C3-H), 5.32 (m, 1H, C5-H) 7.41-7.71 (m, 3H, Bz-H), 8.06-8.18 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.6 (q), 25.6 (t), 26.8 (q), 27.3 (q), 29.1 (t), 29.4 (t), 40.5 (t), 48.05 (d), 69.4 (d), 75.6 (d), 82.6 (d), 109.7 (s), 128.2 (d), 129.5 (d), 130.5 (s), 132.7 (d) ppm.

(1S)-2-[(4S,5R)-5-formyl-2,2-dimethyl-1,3-dioxolan-4-yl]-1-methylethyl benzoate: 2.49



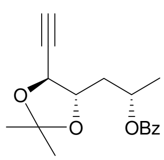
(6.8 g, 18 mmol) was dissolved in acetone (140 mL) and water (35 mL) and 2,4,6-collidine (23,4 mL, 21.5 g, 178 mmol) and MeI (11.1 mL, 25.2 g, 178 mmol) were added. The resulting solution was refluxed under argon for 3.5 h at which point another portion of MeI (11.1 mL) was added. After refluxing for another 4.5 h, the mixture was cooled to room temperature and the acetone was removed in vacuo. The remaining solution was diluted with dichloromethane, washed with aq. HCl (2 M, 3x), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated. Crude **2.50** was used without purification in the next step after overnight drying in vacuo over P_2O_5.

(1S)-2-[4S,5S]-5-(2,2-dibromovinyl)-2,2-dimethyl-1,3-dioxolan-4-yl]-1-methylethyl benzoate (2.51):



Triphenylphosphine (18.6 g, 71.1 mmol, 4 eq.) was dissolved in freshly distilled dry dichloromethane (47 mL) and CBr_4 (sublimed prior to use, 11.8 g, 35.6 mmol, 2 eq.) in dichloromethane (47 mL) was added at 0 °C under argon. The resulting yellow/red solution was stirred for 10 min, after which the crude aldehyde in dichloromethane (42 mL) was added dropwise. The solution was then allowed to reach room temperature and stirred until TLC showed the reaction to be complete (approximately 1 h, *n*-hexane-EtOAc 4:1 to detect the product and dichloromethane-MeOH 98:2 to detect the aldehyde). The reaction was quenched with aq. $NaHCO_3$ (sat.), the aqueous layer was extracted with dichloromethane (2x), the combined organic layers were washed with water and brine (sat.), dried ($MgSO_4$) and concentrated. The resulting brown solid was first filtered over silica ($CHCl_3$) and then further purified by column chromatography (*n*-hexane-EtOAc 9:1) to give **2.51** (5.7 g, 13 mmol, 72% from **2.49**) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 1.36 (m, 6H, CMe_2), 1.42 (d, *J* = 6.3 Hz, 3H, C7-H_3), 1.94 (ddd, *J* = 4.2, 6.2, 14.2 Hz, 1H, C5-H), 2.14 (m, 1H, C5-H'), 3.92 (ddd, *J* = 4.2, 8.0, 8.0 Hz, 1H, C4-H), 4.34 (t, *J* = 8.0 Hz, 1H, C3-H), 5.34 (m, 1H, C6-H), 6.42 (d, *J* = 8.6 Hz, 1H, C2-H), 7.39-7.60 (m, 3H, Bz-H), 8.02-8.07 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.0 (q), 26.5 (q), 27.0 (q) 38.0 (t), 68.9 (d), 76.8 (d), 80.7 (d), 94.5 (s), 109.6 (s), 128.1 (d) 129.4 (d), 130.6 (s), 132.7 (d), 135.0 (d), 165.8 (s) ppm. MS(CI) for $C_{17}H_{20}O_4Br_2$: m/z = 466 $(M + NH_4)^+$.

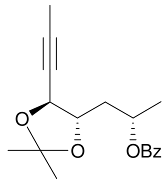
(1S)-2-[(4S,5S)]-5-ethynyl-2,2-dimethyl-1,3-dioxolan-4-yl]-1-methylethyl benzoate (2.52):



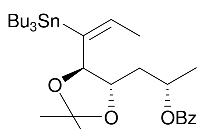
To a solution of **2.51** (3.0 g, 6.7 mmol) in dry THF (77 mL), was added LDA (0.43 M in THF/*n*-hexane, 34 mL, 14.6 mmol, 2.2 eq.) at -78 °C under argon. The resulting solution was stirred for 2 h, after which TLC showed complete conversion. The reaction was quenched with water, extracted with Et_2O (2x), dried ($MgSO_4$), filtered and concentrated. The product was purified by column chromatography (*n*-hexane-EtOAc 9:1) to give the free alkyne (**2.52**, 1.7 g, 5.9 mmol, 88%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 1.38 (s, 3H, CMe_2), 1.43 (s, 3H, CMe_2), 1.43 (d, *J* = 6.2 Hz, 3H, C7-H_3), 1.97 (ddd, *J* = 4.6, 5.4, 14.0 Hz, 1H, C5-H), 2.16 (m, 1H, C5-H') 2.48 (d, *J* = 1.8 Hz, 1H, C1-H), 4.22 (m, 2H, C3-H, C4-H), 5.36 (m, 1H, C6-H), 7.39-7.60 (m, 3H, Bz-H), 8.02-8.08 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.0 (q), 26.0 (q), 26.9 (q), 38.0 (t), 68.7

(d), 70.1 (d), 74.9 (s), 78.4 (d), 110.1 (s), 128.2 (d), 129.4 (d) 130.5 (s), 132.7 (d), 165.8 (s) ppm. MS(CI) for $C_{17}H_{20}O_4$: m/z = 306 $(M + NH_4)^+$, HRMS calcd for $C_{17}H_{20}O_4$-CH_3: 273.113, found: 273.114.

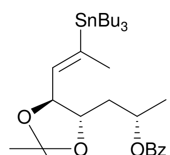
(1S)-2-[(4S,5S)-2,2-dimethyl-5-prop-1-yn-1-yl-1,3-dioxolan-4-yl]-1-methylethyl benzoate (2.53): 2.52 (425 mg, 1.47 mmol) was dissolved in dry THF (3.7 mL) and added to a solution of LDA (0.43 M in THF/*n*-hexane, 10.2 mL, 4.4 mmol, 3.0 eq,) at -78 °C under argon. After 3 min HMPA (1.28 mL, 1.32 g, 7.37 mmol, 5.0 eq.) was added and after an additional 5 min MeI (0.28 mL, 0.63 g, 4.4 mmol, 3.0 eq.) was added. The resulting solution was allowed to warm to -10 °C over 2 h, after which GC-MS showed the reaction to be complete. The reaction was quenched with aq. HCl (1 M), extracted with Et_2O (2x), dried ($MgSO_4$), filtered and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 95:5 to 9:1) gave **2.53** (392 mg, 1.30 mmol, 88%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.36-1.44 (m, 9H, C8-H_3, CMe_2), 1.75 (s, *J* = 2.0 Hz, 3H, C1-H), 1.94 (dt, *J* = 5.2, 14.0 Hz, 1H, C6-H), 2.16 (ddd, *J* = 5.1, 5.7, 14.4 Hz, 1H, C6-H'), 4.11 (ddd, *J* = 5.2, 6.8, 8.0 Hz, 1H, C5-H), 4.24 (ddd, *J* = 2.0, 4.0, 8.0 Hz, 1H, C4-H), 5.34 (m, 1H, C7-H), 7.41-7.57 (m, 3H, Bz-H), 8.04-8.06 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 3.5 (q), 20.1 (q), 26.3 (q), 27.0 (q), 38.0 (t), 68.9 (d), 70.7 (d), 74.7 (s), 78.3 (d), 83.5 (s), 109.3 (s), 128.2 (d), 129.4 (d) 130.6 (s), 132.7 (d), 165.8 (s) ppm. MS(CI) for $C_{18}H_{22}O_4$: m/z = 320 $(M + NH_4)^+$, HRMS calcd for $C_{18}H_{22}O_4$-CH_3: 287.128, found: 287.129.



(1S)-2-{(4S,5S)-2,2-dimethyl-5-[(1E)-2-tri-*n*-butylstannanylprop-1-en-1-yl]-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.54b): 2.53 (230 mg, 761 µmol) was added to a suspension of $PdCl_2(PPh_3)_2$ (27 mg, 38 µmol, 5 mol%) in *n*-pentane (6.9 mL) under argon and after stirring for 10 min, Bu_3SnH (0.83 mL, 3.1 mmol, 4 eq.) was added over 2 min. After 45 min TLC showed complete conversion and the mixture was concentrated. The product was purified by column chromatography (benzene-cyclohexane 9:1) giving **2.54b** (282 mg, 475 µmol, 63%) and the internal hydrostannylation side product **2.54a** (45 mg, 76 µmol, 10%) both as colorless liquids. ^{1}H-NMR **2.54b** ($CDCl_3$, 400 MHz) δ = 0.70-1.03 (m, 15H, Bu-Sn), 1.19-1.60 (m, 21H, C8-H_3, CMe_2, Bu-Sn), 1.77 (ddd, *J* = 3.2, 6.4, 14.0 Hz, 1H, C6-H), 1.96 (d, *J* = 1.6 Hz, 3H, C1-H), 2.06 (ddd, *J* = 6.8, 8.8, 14.0 Hz, 1H, C6-H'), 3.75 (ddd, 3.6, 8.4, 8.4 Hz, 1H, C5-H), 4.54 (t, *J* = 8.4 Hz, 1H, C4-H), 5.31 (m, 1H, C7-H), 5.47 (dd, *J* = 2.0, 8.4 Hz, 1H, C3-H), 7.41-7.56 (m, 3H, Bz-H), 8.02-8.06 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 9.0 (t), 13.6 (q), 19.9 (q), 27.0 (q), 27.2 (q), 27.2 (t), 29.0 (t), 37.8 (t), 69.2 (d), 76.0 (d), 77.7 (d), 108.5 (s), 128.1 (d), 129.4 (d), 130.7 (s), 132.6 (d), 135.6 (d), 147.5 (s), 165.8 (s) ppm. MS(EI) for $C_{30}H_{50}O_4Sn$: m/z = 593 $[M^+]$.



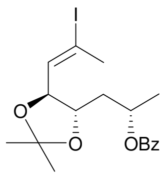
2.54a



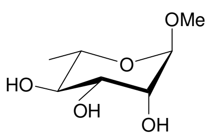
2.54b

^{1}H-NMR **2.54a** ($CDCl_3$, 200 MHz) δ = 0.70-1.03 (m, 15H, Bu-Sn), 1.20-1.60 (m, 21H, C8-H_3, CMe_2, Bu-Sn), 1.72-1.84 (m, 1H, C6-H), 1.79 (d, *J* = 6.6 Hz, 3H, C1-H), 2.04 (ddd, *J* = 5.6, 9.4, 13.6 Hz, 1H, C6-H'), 3.57 (ddd, *J* = 3.4, 9.2, 9.2 Hz, 1H, C5-H), 4.76 (dd, *J* = 1.2, 8.6 Hz, 1H, C4-H), 5.32 (m, 1H, C7-H), 5.84 (dd, *J* = 1.2, 6.6 Hz, 1H, C2-H), 7.38-7.58 (m, 3H, Bz-H), 8.01-8.07 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 10.3 (t), 13.6 (q), 16.4 (q), 19.5 (q), 27.1 (q), 27.3 (t), 29.0 (t), 38.1 (t), 69.5 (d), 77.6 (d), 80.8 (d), 108.1 (s), 128.1 (d), 129.4 (d), 130.7 (s), 132.6 (d), 137.8 (d), 147.7 (s), 165.7 (s) ppm.

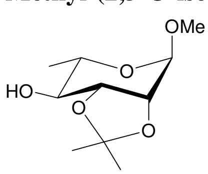
(1S)-2-{(4S,5S)-5-[(1E)-2-iodoprop-1-en-1-yl]-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.38a): 2.54b (200 mg, 0.34 mmol) was dissolved in dichloromethane (2.7 mL) and I_2 (114 mg, 0.45 mmol, 1.3eq) in dichloromethane (0.7 mL) was added at -78 ^oC under argon. The resulting solution was stirred for 10 min at -78 ^oC and then allowed to warm to room temperature. The solution was concentrated and **2.38a** (144 mg, 0.33 mmol, 99%) was isolated as a yellow oil after column chromatography (*n*-hexane-EtOAc 39:1; 2.5 volume-% Et_3N was used during the preparation of the column). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.35 (s, 3H, CMe_2), 1.38 (s, 3H, CMe_2), 1.41 (d, *J* = 6.3 Hz, 3H, C8-H_3), 1.84 (ddd, *J* = 3.9, 5.7, 14.4 Hz, 1H, C6-H), 2.06 (m, 1H, C6-H'), 2.49 (s, 3H, C1-H), 3.84 (ddd, *J* = 3.9, 8.1, 8.1 Hz, 1H, C5-H), 4.29 (t, *J* = 8.7 Hz, 1H, C4-H), 5.31 (m, 1H, C7-H), 6.14 (d, 1H, C3-H, *J* = 8.7 Hz), 7.41-7.58 (m, 3H, Bz-H), 8.04 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 19.9 (q), 26.7 (q), 27.1 (q), 28.4 (q), 37.5 (t), 68.9 (d), 76.9 (d), 77.7 (d), 101.5 (s), 109.0 (s), 128.1 (d), 129.4 (d), 130.5 (s), 132.6 (d), 137.0 (d) 165.7 (s) ppm. MS(CI) for $C_{18}H_{23}IO_4$: m/z = 448 $(M + NH_4)^+$, HRMS calcd for $C_{20}H_{23}IO_4$: 430.064, found: 430.066.



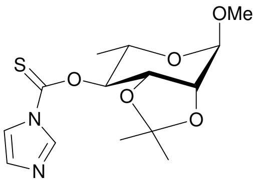
Methyl α-L-rhamnopyranoside (2.55): L-rhamnose (2.0 g, 11 mmol) was dissolved in MeOH (80 mL) and acetyl chloride (1.2 mL, 17 mmol) was added. The resulting solution was stirred under argon for 60 h and then neutralized with Na_2CO_3 (s) and filtered. The filtrate was concentrated and purified by column chromatography (EtOAc-MeOH 97.5 : 2.5) to give **2.55** (1.4 g, 6.9 mmol, 63%) as a white foam. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 17.3 (q), 54.6 (q), 67.9 (d), 70.7 (d), 71.5 (d), 72.5 (d), 100.7 (d) ppm. MS(CI) for $C_7H_{14}O_5$: m/z = 196 $(M + NH_4)^+$.



Methyl-(2,3-O-isopropylidene-α-L-rhamnopyranoside) (2.56):[46] **2.55** (21.5 g, 110 mmol) was dissolved in dry acetone and $CuSO_4$ (34 g, 220 mmol, 2 eq) and H_2SO_4 (1.7 mL) were added. The resulting green suspension was stirred overnight under argon and then filtered. The filtrate was made basic (pH 9) with 25% aq. ammonia and the resulting blue suspension was filtered again. The filtrate was concentrated, suspended in brine (sat.) and extracted with dichloromethane (3x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated. Purification by column chromatography (*n*-hexane-EtOAc 9:1 to 4:1) gave **2.56** as an oil (23.3 g, 98.6 mmol, 90%). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.31 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.35, 1.53 (2s, 6H, CMe_2), 2.16 (d, *J* = 4.5 Hz, 1H, OH), 3.39 (s, 3H, OMe), 3.39 (m, 1H, C4-H), 3.65 (m, 1H, C5-H), 4.07 (t, *J* = 6.6 Hz, 1H, C3-H), 4.13 (d, *J* = 6.0 Hz, 1H, C2-H), 4.85 (s, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 17.3 (q), 26.0 (q), 27.8 (q), 54.8 (q), 65.6 (d), 74.3 (d) 75.6 (d), 78.3 (d), 98.0 (d), 109.3 (s) ppm. MS(CI) for $C_{10}H_{18}O_5$: m/z = 236 $(M + NH_4)^+$.

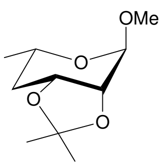


Methyl-4-O-(imidazol-1-ylthiocarbonyl)-2,3-O-isopropylidene-α-L-rhamnopyranoside

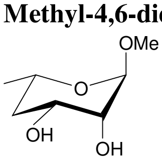


(2.57):[48] A solution of **2.56** (13.5 g, 57.1 mmol) and 1,1'-thiocarbonyldiimidazole (12.8 g, 71.5 mmol, 1.3 eq) in anhydrous 1,2-dichloroethane (200 mL) was refluxed for 2 h under argon. After cooling to room temperature, the solvent was removed and the product was purified by crystallization from ether/*n*-hexane and/or by column chromatography (*n*-hexane-EtOAc 4:1 to 1:1) giving a white solid (19.0 g, 54.8 mmol, 99.7%). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.28 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.36, 1.61 (2s, 6H, CMe_2), 3.43 (s, 3H, OMe), 3.95 (dq, $J_{5,6}$ = 6.3 Hz, 1H, C5-H), 4.22 (d, *J* = 5.4 Hz, 1H, C2-H), 4.37 (dd, *J* = 5.7, 7.5 Hz, 1H, C3-H), 4.95 (s, 1H, C1-H), 5.74 (dd, *J* = 7.8, 9.9 Hz, 1H, C4-H), 7.07, 7.65, 8.40 (3s, 3H, imidazole) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 17.0 (q), 26.1 (q), 27.4 (q), 55.1 (q), 63.6 (d), 75.1 (d), 75.8 (d), 83.0 (d), 97.8 (d), 110.2 (s), 118.0 (d), 130.8 (d) 136.7 (d), 183.8 (s) ppm. MS(CI) for $C_{14}H_{20}N_2O_5S$: m/z = 329 $(M + H)^+$.

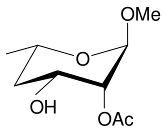
Methyl-4,6-dideoxy-2,3-O-isopropylidene-α-L-lyxo-hexapyranoside (2.58):[48] **2.57** (2.0 g, 6.1 mmol) was dissolved in toluene (27 mL) and AIBN (0.3 g, 1.8 mmol, 30 mol%) and tris(trimethylsilyl)silane (2.3 mL, 7.5 mmol, 1.2 eq) were added. The resulting solution was slowly heated to 110 °C and then refluxed for 30 min under argon, after which TLC showed complete conversion. After cooling to room temperature, aq. $NaHCO_3$ (20% w/w) was added and the product was extracted with EtOAc (3x). The combined organic layers were dried ($MgSO_4$) and concentrated below 30 °C using MeOH to remove the toluene. **2.58** was isolated as a yellow oil after column chromatography (*n*-hexane-EtOAc 98:2 to 95:5). Due to volatility of the product, the yield was determined in the next step. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.22 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.34, 1.52 (2s, 6H, CMe_2), 1.47 (m, 1H, C4-H), 1.86 (ddd, *J* = 2.1, 6.6, 13.2 Hz, 1H, C4-H'), 3.38 (s, 3H, OMe), 3.77 (m, 1H, C5-H), 3.92 (d, *J* = 5.4 Hz, 1H, C2-H), 4.30 (m, 1H, C3-H), 4.91 (s, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 21.1 (q), 26.2 (q), 28.1 (q), 36.0 (t), 54.7 (q), 61.9 (d), 70.9 (d), 72.6 (d), 98.7 (d), 108.6 (s) ppm. MS(CI) for $C_{10}H_{18}O_4$: m/z = 220 $(M + NH_4)^+$.



Methyl-4,6-dideoxy-α-L-lyxo-hexapyranoside (2.59): Amberlite-120-(Na^+)-resin was stirred in an aq. HCl-solution (2 M) for 1.5 h and then filtered off and flushed with aq. HCl-solution (2 M) twice. 4.0 g Of the thus obtained resin was added to a solution of **2.58** (1.23 g, 6.10 mmol) in water (37 mL) and 1,4-dioxane (37 mL). After stirring for 12 h, the resin was filtered off and the solution was neutralized with aq. NaOH (1 M) and then concentrated. After column chromatography ($CHCl_3$-EtOH 9:1) **2.59** (703 mg, 4.33 mmol, 71% from **2.57**) was isolated as a white foam. NMR-data are as reported in literature.[73] ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.23 (d, *J* = 6.0 Hz, 3H, C6-H_3), 1.47 (q, *J* = 12.4 Hz, 1H, C4-H), 1.74-1.87 (m, 3H, C4-H', 2-OH), 3.37 (s, 3H, OMe), 3.72 (s, 1H, C2-H), 3.84 (m, 1H, C3-H), 3.96 (m, 1H, C5-H), 4.73 (s, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 21.0 (q), 36.3 (t), 54.8 (q), 63.7 (d), 65.6 (d), 68.6 (d), 101.2 (d) ppm. MS(CI) for $C_7H_{14}O_4$: m/z = 180 $(M + NH_4)^+$.

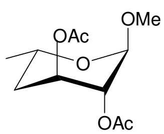


Methyl-2-O-acetyl-4,6-dideoxy-α-L-lyxo-hexapyranoside (2.60): 2.59 (670 mg, 4.13 mmol) was dissolved in acetonitrile (13 mL), and *p*-toluenesulfonic acid monohydrate (41 mg) and trimethyl orthoacetate (0.95 mL, 7.43 mmol, 1.8 eq) were added. The resulting solution was stirred for 10 min and then concentrated. The residue was dissolved in dichloromethane (13 mL) and 90% aqueous trifluoroacetic acid (4.06 mL) was added. The thus obtained mixture was stirred for 5 min after which TLC (*n*-hexane-EtOAc 4:1) showed complete conversion. After concentration, the residue was taken up in dichloromethane, washed with aq. $NaHCO_3$ (5% w/w) and water, dried (Na_2SO_4) and concentrated to give **2.60** (675 mg, 3.30 mmol, 80%) as an oil, which was used in the next step without further purification. ^{1}H-NMR of the crude product showed only 1 regioisomer present: ($CDCl_3$, 300 MHz) δ = 1.24 (d, J = 6.3 Hz, 3H, C6-H_3), 1.56 (q, J = 12.0 Hz, 1H, C4-H), 1.77 (m, 2H, C4-H', OH), 2.14 (s, 1H, OAc), 3.35 (s, 3H, OMe), 3.87 (m, 1H, C5-H), 4.12 (m, 1H, C3-H), 4.71 (s, 1H, C1-H), 4.92 (br s, 1H, C2-H) ppm.

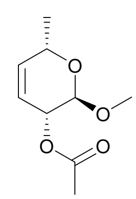


Methyl-2,3-di-O-acetlyl-4,6-dideoxy-α-L-arabino-hexopyranoside (2.62):

Trifluoromethanesulfonic anhydride (7.45 mL, 44.1 mmol, 3.0 eq) was slowly added to a solution of **2.60** (3.0 g, 14.7 mmol) and pyridine (7.2 mL, 88.2 mmol, 6.0 eq) in dichloromethane (70 mL) at -10 ^{o}C under argon. The mixture was slowly warmed to room temperature and after stirring for an additional 1.5 h, the solution was diluted with dichloromethane and poured into ice-cold aq. $NaHCO_3$ (20% w/w). The organic layer was washed with aq. HCl (1 M), water and aq. $NaHCO_3$ (sat.), dried (Na_2SO_4) and concentrated to give **2.61**. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.28 (d, J = 6.3 Hz, 3H, C6-H), 1.98-2.13 (m, 5H, C4-H,H', OAc), 3.35 (s, 3H, OMe), 3.93 (m, 1H, C5-H), 4.72 (s, 1H, C1-H), 5.14 (brs, 1H, C2-H), 5.29 (m, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.6 (2q), 34.5 (t), 55.0 (q), 63.7 (d), 67.5 (d), 81.6 (d), 98.8 (d), 121.4 (s), 169.6 (s) ppm. ^{19}F-NMR ($CDCl_3$) δ = 1.81 ppm.

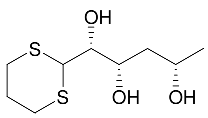


A solution of the crude triflate **2.61** and tetra-*n*-butylammonium acetate (48.7 g, 162 mmol, 11.0 eq) in dry toluene (40 mL) was stirred overnight under argon. The mixture was concentrated and purified by column chromatography (*n*-hexane-EtOAc 4:1) to give **2.62** (2.6 g, 10.4 mmol, 71% from **2.60**) as an oil and a side product resulting from dehydration (0.2 g, 1.2 mmol, 8%). ^{1}H-NMR **2.60** ($CDCl_3$, 500 MHz) δ = 1.22 (d, J = 6.5 Hz, 3H, C6-H_3), 1.70 (m, $J_{4eq,4ax}$ = 14.5, $J_{4eq,5}$ = 2.5 Hz, 1H, C4-Heq), 1.81 (ddd, $J_{3,4ax}$ = 3.5, $J_{4ax,5}$ = 11.0, $J_{4ax,4eq}$ = 14.5 Hz, 1H, C4-ax), 2.08, 2.09 (2s, 6H, 2OAc), 3.38 (s, 3H, OMe), 4.14 (m, 1H, C5-H), 4.60 (s, 1H, C1-H), 4.74 (m, 1H, C2-H), 4.91 (m, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 20.7 (q), 20.8 (q), 21.0 (q), 32.6 (t), 55.1 (q), 59.7 (d), 66.5 (d), 67,2 (d), 98.7 (d), 169.3 (s), 170.0 (s) ppm. MS(EI) for $C_{11}H_{18}O_6$: m/z = 215 [M + H – MeOH]$^+$, MS(CI) = 264 (M + NH_4)$^+$.



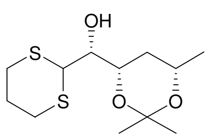
^{1}H-NMR of the side product ($CDCl_3$, 200 MHz) δ = 1.32 (d, J = 7.2 Hz, C6-H_3), 2.09 (s, 3H, OAc), 3.47 (s, 3H, OMe), 4.30 (m, 1H, C5-H), 4.74 (brs, 1H, C1-H), 4.89 (m, 1H, C2-H), 5.78 + 5.83 (2 ddd, J = 1.2, 2.2, 4.8 Hz and J = 1.2, 2.0, 5.0 Hz, 1H, C3-H), 5.97 + 6.02 (2m, 1H, C4-H) ppm. MS(EI) for $C_9H_{14}O_4$: m/z = 186 [M]$^+$, MS(CI) = 204 (M + NH_4)$^+$.

^{1}H-NMR of the C3-epimer of **2.62** obtained from acetylation of **2.59**: ($CDCl_3$, 300 MHz) δ = 1.24 (d, *J* = 6.3 Hz, 3H, C6-H), 1.74 (m, 2H, 2C4-H), 1.99 (s, 3H, OAc), 2.12 (s, 3H, OAc), 3.35 (s, 3H, OMe), 3.94 (m, 1H, C5-H), 4.65 (s, 1H, C1-H), 5.05 (s, 1H, C2-H), 5.21 (m, 1H, C3-H) ppm.

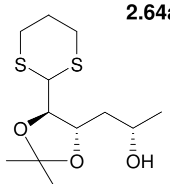


4,6-Dideoxy-L-arabino-hexose-trimethylen-dithioacetal (2.63): 2.62 (100 mg, 0.41 mmol) was dissolved in MeOH-THF (2.0 mL, 1:1 v/v) and NaOMe was added until pH 9 was reached. The thus obtained reaction mixture was stirred for 2 h and then concentrated. The residue was dissolved in 37% aq. HCl (1.0 mL), after which 1,3-propanedithiol (85 µL, 0.82 mmol, 2.0 eq) was added dropwise. The resulting solution was stirred for 24 h and then neutralized with 25% aq. ammonia. Subsequently the aqueous layer was washed with petroleum-ether (40-60; 5x) and concentrated to give a white solid which was suspended in acetone and stirred for 5 min. The solid was filtered off and the filtrate concentrated affording **2.63** (87 mg, 0.36 mmol, 87%) as a white solid. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.24 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.64 (m, 1H, C4-H), 1.81 (m, 1H, C4-H'), 2.05 (m, 2H, dithian-H), 2.67-2.97 (m, 4H, dithian-H), 3.70 (d, *J* = 8.1 Hz, 1H, C2-H), 4.05 (d, *J* = 8.4 Hz, 1H, C3-H), 4.15 (m, 1H, C5-H), 4.31 (d, *J* = 9.6 Hz, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 23.9 (q), 25.3 (t) , 27.0 (t), 27.6 (t), 42.2 (t), 47.7 (d), 67.9 (d), 70.9 (d), 73.5 (d) ppm. MS(EI) for $C_9H_{18}O_3S_2$: m/z = 238 [M^+], HRMS calcd for $C_9H_{18}O_3S_2$: 238.070, found: 238.071.

4,6-Dideoxy-2,3-O-isopropyliden-L-arabino-hexose-trimethylen-dithioacetal (2.64b):



2.64a

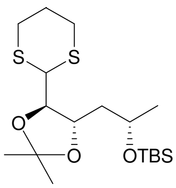


2.64b

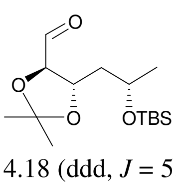
2.63 (600 mg, 2.52 mmol) was dissolved in dry acetone and $CuSO_4$ (778 mg, 3.06 mmol, 1.2 eq.) and a drop of H_2SO_4 were added. The resulting green suspension was stirred overnight and then filtered. The filtrate was neutralized with 25% aq. NH_3 and the resulting blue suspension was filtered again. The filtrate was concentrated, suspended in brine (sat.) and extracted with dichloromethane (3x). The combined organic layers were dried (Na_2SO_4) and concentrated. Purification by column chromatography (*n*-hexane-EtOAc 95:5 to 4:1) gave **2.64b** (549 mg, 1.97 mmol, 78%) and **2.64a** (55 mg, 0.20 mmol, 8%) as colorless oils. ^{1}H-NMR **2.64b** ($CDCl_3$, 200 MHz) δ = 1.21 (d, *J* = 6.0 Hz, 3H, C6-H_3), 1.43, 1.45 (2s, 6H, CMe_2), 1.67 (m, 1H, C4-H), 1.86-2.17 (m, 3H, C4-H', dithian-H), 2.74-3.01 (m, 4H, dithian-H), 3.09 (brs, 1H, OH), 3.93 (dd, *J* = 5.8, 8.0 Hz, 1H, C2-H), 4.05 (m, 1H, C5-H), 4.11 (d, *J* = 5.2 Hz, 1H, C1-H), 4.21 (ddd, *J* = 2.6, 7.6, 10.0 Hz, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 23.2 (q), 25.6 (t), 26.8 (q), 27.2 (q), 29.2 (t), 29.4 (t), 42.3 (t), 47.9 (d), 67.2 (d), 78.8 (d), 82.9 (d), 110.0 (s) ppm. MS(EI) for $C_{12}H_{22}O_3S_2$: m/z = 278 [M^+], HRMS calcd for $C_{12}H_{22}O_3S_2$: 278.101, found: 278.101.

^{1}H-NMR **2.64a** ($CDCl_3$, 300 MHz) δ = 1.20 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.40, 1.46 (2s, 6H, CMe_2), 1.50 (m, 1H, C4-H), 1.92- 2.14 (m, 3H, C4-H', dithian-H), 2.74-2.96 (m, 5H, dithian-H, OH), 3.64 (m, 1H), 4.03 (m, 1H), 4.14 (d, *J* = 7.0 Hz, 1H, C1-H), 4.25 (m, 1H, C3-H) ppm.

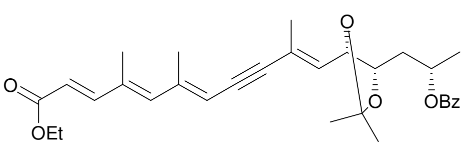
5-O-*tert*-Butyl-dimethyl-silyl-4,6-dideoxy-2,3-O-isopropyliden-L-arabino-hexose-trimethylen-dithioacetal (2.65): 2.64b (500 mg, 1.80 mmol) was dissolved in DMF (3.0 mL) and TBDMSCl (541 mg, 3.59 mmol, 2.0 eq.) and imidazole (245 mg, 3.59 mmol, 2.0 eq.) were added. The resulting solution was stirred for 12 h at 70 °C under argon. After cooling to room temperature, the reaction mixture was diluted with water, extracted with Et_2O (2x), dried (Na_2SO_4) and concentrated. Purification by column chromatography (*n*-hexane-EtOAc 95:5 to 9:1) gave **2.65** (593 mg, 1.51 mmol, 84%) as an oil. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.06 (s, 6H, 2MeSi), 0.88 (s, 9H, *t*BuSi), 1.20 (d, *J* = 6.3 Hz, 3H, C6-H_3), 1.40, 1.42 (2s, 6H, CMe_2), 1.72-2.14 (m, 4H, C4-H,H', dithian-H), 2.74-2.97 (m, 4H, dithian-H), 3.90 (dd, *J* = 5.1, 7.5 Hz, 1H, C2-H), 4.01-4.15 (m, 3H, C1-H, C3-H, C5-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = -4.9 (q), -4.7 (q), 18.0 (s), 22.8 (q), 25.7 (t), 25.8 (q), 26.8 (q), 27.3 (q), 29.1 (t), 29.5 (t), 43.6 (t), 47.9 (d), 66.1 (d), 75.9 (d), 83.2 (d), 109.3 (s) ppm. MS(EI) for $C_{18}H_{36}O_3S_2Si$: m/z = 392 [M^+], MS(CI) for $C_{18}H_{36}O_3S_2Si$: m/z = 393 $(M + H)^+$, 410 $(M + NH_4)^+$, HRMS calcd for $C_{18}H_{36}O_3SiS_2$: 392.188, found: 392.188.



(4R,5S)-5-[(2S)-2-(tert-Butyl-dimethyl-silanyloxy)-propyl]-2,2-dimethyl-1,3-dioxolane-4-carbaldehyde (2.66): 2.65 (137 mg, 0.35 mmol) was converted into **2.66** (94 mg, 0.31 mmol, 89%) using a procedure analogous to the synthesis of **2.50**. ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 0.06 (s, 6H, 2MeSi), 0.87 (s, 9H, *t*BuSi), 1.19 (d, *J* = 6.0 Hz, 3H, C6-H_3), 1.42, 1.47 (2s, 6H, CMe_2), 1.70-1.97 (m, 2H, C4-H,H'), 3.97-4.06 (m, 2H, C2-H, C5-H), 4.18 (ddd, *J* = 5.0, 7.6, 7.6 Hz, 1H, C3-H), 9.72 (d, *J* = 2.4 Hz, 1H, O=CH) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = -5.0 (q), -4.5 (q), 17.9 (s), 23.2 (q), 25.7 (q), 26.1 (q), 27.0 (q), 42.8 (t), 65.6 (d), 73.9 (d), 85.0 (d), 110.8 (s), 200.6 (d) ppm.

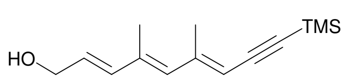


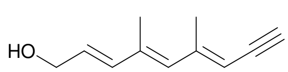
(1S)-2-{(4S,5S)-5-(11-ethoxy-2,6,8-trimethyl-11-oxoundeca-1,5,7,9-tetraen-3-yn-1-yl)-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.36a): 2.38a (122 mg, 0.28 mmol, 1.2 eq) was dissolved in *i*PrNH_2 (1.0 mL) and $Pd(PPh_3)_4$ (5.3 mg, 4.6 µmol, 2 mol%) was added. The solution was stirred under argon at ambient temperature for 5 min, after which CuI (0.9 mg, 4.6 µmol, 2 mol%) was added. After 5 min, **2.37a** (47 mg, 0.23 mmol) in *i*PrNH_2 (0.85 mL) was added and the mixture was stirred for 2 h and then concentrated. The residue was dissolved in Et_2O, washed with aq. NH_4Cl (sat.) and brine (sat.), dried (Na_2SO_4) and concentrated. **2.36a** (109 mg, 0.22 mmol, 94%) was isolated as a yellow oil after purification by column chromatography (*n*-hexane-EtOAc 19:1 to 9:1) and remaining **2.38a** (20 mg, 0.05 mmol, 20%) was recovered. ^{1}H-NMR **major isomer** ($CDCl_3$, 200 MHz) δ = 1.30 (t, *J* = 7.0 Hz, 3H, CH_3CH$_2$O), 1.34-1.42 (m, 9H, C16-H_3, CMe_2), 1.76-2.20 (m, 11H, C4,- C6-, C10-CH_3, C14-H_2), 3.85 (ddd, *J* = 3.8, 8.2, 8.2 Hz, 1H, C13-H), 4.22 (q, *J* = 7.2 Hz, 2H, CH$_3$$CH_2$O), 4.39 (m, 1H, C12-H), 5.31 (m, 1H, C15-H), 5.67 (s, 1H, C7-H), 5.74 (dd, *J* = 1.6, 9.0 Hz, 1H, C11-H), 5.91 (d, *J* = 15.6 Hz, 1H, C3-H), 6.31 (s, 1H, C5-H), 7.30-7.59 (m, 5H, Bz-H, C2-H), 8.03 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 13.9 (q), 14.2 (q), 17.9 (q). 19.5 (q), 19.9 (q), 26.7 (q). 27.1 (q), 37.6 (t), 60.1 (t), 69.0 (d), 71.1 (d), 77.6 (d), 87.0



(s), 99.9 (s), 108.9 (s), 113.4 (d), 117.6 (d), 124.4 (s), 128.1 (d), 129.4 (d), 130.6 (s), 132.2 (d), 132.6 (d), 134.2 (s), 140.5 (d), 146.1 (s), 149.7 (d), 165.7 (s), 167.0 (s) ppm. MS(EI) for $C_{31}H_{38}O_6$: m/z = 506 [M^+], HRMS calcd for $C_{31}H_{38}O_6$: 506.267, found: 506.268.

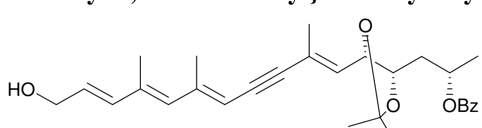
4,6-Dimethyl-9-trimethylsilanyl-nona-2,4,6-trien-8-yn-1-ol (2.67): 2.43 (398 mg, 1.44 mmol) was dissolved in dichloromethane (7.2 mL) and DiBAl-H (1.0 M in *n*-hexanes, 3.17 mL, 3.17 mmol, 2.2 eq) was added at -70 °C. The resulting mixture was stirred for 2 h under argon, while warming to -60 °C. The reaction was quenched with MeOH and the precipitate was dissolved with aq. NH_4Cl (sat.) and aq. HCl (1.0 M). The aqueous layer was extracted with dichloromethane (3x), after which the combined organic layers were washed with aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$), filtered and concentrated. **2.67** (336 mg, 1.44 mmol, quantitative) was isolated as a colorless oil (single isomer) after purification by column chromatography (*n*-pentane-EtOAc 85:15 to 4:1). ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 0.20 (s, 9H, TMS), 1.96 (s, 3H, C6-CH_3), 2.09 (s, 3H, C4-CH_3), 4.23 (d, *J* = 6.0 Hz, 2H, *CH₂*OH), 5.47 (s, 1H, C7-H), 5.87 (dt, *J* = 6.0, 15.8 Hz, 1H, C2-H), 5.95 (s, 1H, C5-H), 7.32 (d, *J* = 15.8 Hz, 1H, C3-H) ppm.



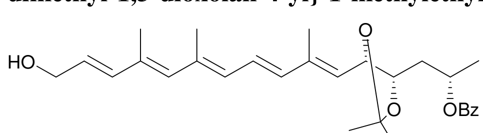


4,6-Dimethylnona-2,4,6-trien-8-yn-1-ol (2.68): 2.67 (50 mg, 0.21 mmol) was converted into **2.68** (23 mg, 0.14 mmol, 67%) using a procedure analogous to the synthesis of **2.37a**. Due to the instability of **2.68**, it was immediately used in the next step without purification.

(1S)-2-{(4S,5S)-5-(11-hydroxy-2,6,8-trimethyl-undeca-1,5,7,9-tetraen-3-yn-1-yl)-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.69): 2.68 (23 mg, 0.14 mmol) was coupled to **2.38a** (88 mg, 0.20 mmol, 1.4 eq) to give **2.69** (51 mg, 0.11 mmol, 77%, single isomer) as described for the preparation of **2.36a**. Column chromatography was performed with dichloromethane-MeOH 99:1 to 98:2. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.34-1.42 (m, 9H, C16-H_3, CMe_2), 1.79-2.20 (m, 11H, C4,- C6-, C10-CH_3, C14-H_2), 3.84 (ddd, *J* = 3.3, 8.1, 8.1 Hz, 1H, C13-H), 4.24 (brs, 2H, *CH₂*OH), 4.39 (m, 1H, C12-H), 5.30 (m, 1H, C15-H), 5.52 (s, 1H, C7-H), 5.70 (d, *J* = 9.0 Hz, 1H, C11-H), 5.89 (dt, *J* = 6.0, 15.6 Hz, 1H, C2-H), 5.98 (s, 1H, C5-H), 6.30 (d, *J* = 15.9 Hz, 1H, C3-H), 7.42 (t, *J* = 7.5 Hz, 2H, Bz-H), 7.54 (t, *J* = 7.8 Hz, 1H, Bz-H), 8.03 (d, *J* = 7.5 Hz, 2H, Bz-H) ppm.

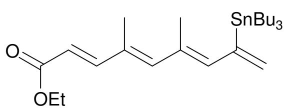


(1S)-2-{(4S,5S)-5-(11-hydroxy-2,6,8-trimethyl-undeca-1,3,5,7,9-pentaen-1-yl)-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.70): HCl (2.0 M aq, 30 mL) was added to a double schlenk containing zinc (5.0 g) and the resulting suspension was stirred under argon for 5 min. The HCl was then removed by filtration and the zinc was washed with HCl (2.0 M aq, 3x 30 mL) and water (10 x 30 mL). The zinc was suspended in water (40 mL) and argon was bubbled

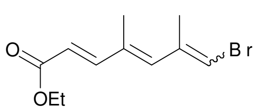


through for 15 min, after which $Cu(OAc)_2{\cdot}H_2O$ (0.5 g) was added and stirring continued for another 15 min. Subsequently, $AgNO_3$ (0.5 g) was added, which resulted in an exothermic reaction and a change of color from light grey to black. After stirring for 30 min, the zinc-amalgam was filtered and washed with water (30 mL), MeOH, acetone and Et_2O. The zinc-amalgam was then suspended in MeOH (15 mL) and water (10 mL) and 3 mL of this suspension was added to **2.69** (25 mg, 54 μmol), after which the reaction mixture was stirred overnight under argon. The amalgam was removed by filtration over celite and the filtrate was partly concentrated in vacuo until all the MeOH was removed. The remaining aqueous solution was subsequently extracted with dichloromethane (3x) and the combined organic layers were washed with brine (sat.), dried (Na_2SO_4) and concentrated. Purification by column chromatography and/or preparative TLC (dichloromethane-MeOH 99:1 to 98:2) gave a small amount of product that could be **2.70**. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.26-1.48 (m, 9H, C16-H_3, CMe_2), 1.72-2.28 (m, 11H, C4,- C6-, C10-CH_3, C14-H_2), 3.83 (ddd, *J* = 3.3, 8.1, 8.1 Hz, 1H, C13-H), 4.24 (m, 2H, *CH₂*OH), 4.43 (m, 1H, C12-H), 5.23-5.42 (m, 3H), 5.80-6.86 (m, 5H), 7.42 (m, 2H, Bz-H), 7.54 (m, 1H, Bz-H), 8.03 (d, *J* = 7.5 Hz, 2H, Bz-H) ppm. MS(EI) for $C_{29}H_{38}O_5$: m/z = 466 [M^+], MS(CI) m/z = 484 $(M + NH_4)^+$.

Ethyl 4,6-Dimethyl-8-tri-*n*-butylstannanylnona-2,4,6,8-tetraenoate (2.71): A solution of **2.37a** (55 mg, 0.27 mmol) in THF (1.0 mL) was added to a solution of $PdCl_2(PPh_3)_2$ (9.5 mg, 14 μmol, 5 mol%) in THF (1.5 mL) and the resulting mixture was stirred for 10 min under argon. Subsequently, Bu_3SnH (0.29 mL, 1.1 mmol, 4.0 eq.) was added over 2 min and stirring was continued for an additional 15 min. The reaction mixture was concentrated and **2.71** was isolated as a colorless oil after purification by column chromatography (*n*-hexane-EtOAc 99:1 to 9:1). ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 0.91 (m, 15H), 1.30 (m, 9H), 1.50 (m, 6H), 1.92 (s, 3H, C6-CH_3), 1.99 (s, 3H, C4-CH_3), 4.21 (q, *J* = 7.0 Hz, 2H, CH_3CH_2O), 5.41 (dd, *J* = 1.2, 3.2 Hz, 1H, C9-H), 5.71 (dd, *J* = 2.0, 3.2 Hz, 1H, C9-H'), 5.84 (d, *J* = 15.4 Hz, 1H, C2-H), 6.24 (s, 1H, C7-H), 6.32 (s, 1H, C5-H), 7.37 (d, *J* = 15.4 Hz, 1H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 9.9 (t), 13.6 (q), 13.7 (q), 14.2 (q), 17.5 (q), 27.2 (t), 28.9 (t), 60.0 (t), 115.7 (d), 127.7 (t), 130.8 (s), 131.1 (s), 140.4 (d), 143.9 (d), 150.8 (d), 151.4 (s), 167.5 (s) ppm. MS(EI) for $C_{25}H_{44}O_2Sn$: m/z = 496 [M^+], HRMS calcd for $C_{25}H_{44}O_2Sn$: 496.236, found: 496.235.

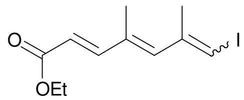


Ethyl (2E,4E)-7-bromo-4,6-dimethylhepta-2,4,6-trienoate (2.72): $BrCH_2PPh_3Br$ (479 mg, 1.10 mmol, 2.0 eq) was suspended in dry THF (2.7 mL) under argon and piperidine (108 μl, 1.10 mmol, 2.0 eq) and *n*-BuLi (1.6 M in *n*-hexane, 0.69 mL, 1.10 mmol, 2.0 eq) were added upon which the solution turned brown/red. After stirring for 15 min, **2.39c** (100 mg, 0.55 mmol) in THF (2.2 mL) was added and the reaction mixture was stirred for 6 h. The reaction was quenched with aq. NH_4Cl (sat.), the aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried (Na_2SO_4) and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 95:5) gave **2.72** (57 mg, 0.22 mmol, 40%) as a mixture of isomers. ^{1}H-NMR ($CDCl_3$, 300 MHz) **major isomer** δ = 1.30 (t, *J* = 7.0 Hz, 3H, CH_3CH_2O), 1.91 (s, 3H, C6-CH_3), 1.94 (s, 3H, C4-CH_3), 4.21 (q, *J* = 7.2 Hz, 2H, CH_3CH_2O), 5.92 (d, *J* = 15.6 Hz, 1H,

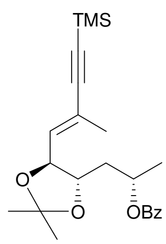


C2-H), 6.17 (s, 1H, C5-H), 6.24 (s, 1H, C7-H), 7.31 (d, J = 15.3 Hz, 1H, C3-H) ppm. **Minor isomer** δ = 1.31 (t, J = 7.0 Hz, 3H, CH_3CH$_2$O), 1.88 (s, 3H, C6-CH$_3$), 1.96 (s, 3H, C4-CH$_3$), 4.21 (q, J = 7.2 Hz, 2H, CH$_3$$CH_2$O), 5.95 (d, J = 15.6 Hz, 1H, C2-H), 6.14 (s, 1H, C5-H), 6.35 (s, 1H, C7-H), 7.39 (d, J = 15.9 Hz, 1H, C3-H) ppm.

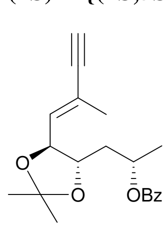
Ethyl (2E,4E)-7-iodo-4,6-dimethylhepta-2,4,6-trienoate (2.73): Preparation as described for **2.72**. **2.73** (70 mg, 0.23 mmol, 13%) was isolated as a single isomer from **2.39c** (321 mg, 1.76 mmol) using 1.91 g of ICH_2PPh_3I (3.53 mmol, 2.0 eq). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.30 (t, J = 7.2 Hz, 3H, CH_3CH$_2$O), 1.89 (s, 3H, C6-CH$_3$), 1.99 (s, 3H, C4-CH$_3$), 4.21 (q, J = 7.2 Hz, 2H, CH$_3$$CH_2$O), 5.93 (d, J = 16.0 Hz, 1H, C2-H), 6.21 (s, 1H, C5-H), 6.31 (s, 1H, C7-H), 7.30 (d, J = 16.0 Hz, 1H, C3-H) ppm.



(1S)-2-{(4S,5S)-2,2-dimethyl-5-[(1E)-2-methyl-4-trimethylsilanyl-but-1-en-3-yn-1-yl]-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.74): Ethynyl-trimethylsilane (99 µl, 0.70 mmol, 1.5 eq) was dissolved in *i*PrNH$_2$ (1.7 mL) and Pd(PPh$_3$)$_4$ (10.7 mg, 9.3 µmol, 2 mol%) was added. The solution was stirred under argon at ambient temperature for 5 min, after which CuI (1.8 mg, 9.3 µmol, 2 mol%) was added. After 5 min, **2.38a** (200 mg, 0.46 mmol) in *i*PrNH$_2$ (1.4 mL) was added and the mixture was stirred for 1 h and then concentrated. The residue was dissolved in Et$_2$O and washed with aq. NH$_4$Cl (sat.). The aqueous layer was extracted with Et$_2$O (3x) and the combined organic layers were washed with brine (sat.), dried (Na$_2$SO$_4$) and concentrated. The product (**2.74**, 180 mg, 0.45 mmol, 97%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 19:1). ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 0.16 (s, 9H, TMS), 1.38 (m, 9H, C9-H$_3$, CMe$_2$), 1.74-1.87 (m, 4H, C7-H, C3-CH$_3$), 2.03 (ddd, J = 6.6, 8.6, 14.0 Hz, 1H, C7-H'), 3.82 (ddd, 3.4, 8.4, 8.4 Hz, 1H, C6-H), 4.34 (t, J = 8.6 Hz, 1H, C5-H), 5.32 (m,1H, C8-H), 5.78 (dd, J = 1.6, 9.0 Hz, 1H, C4-H), 7.41-7.58 (m, 3H, Bz-H), 8.04 (m, 2H, Bz-H) ppm.

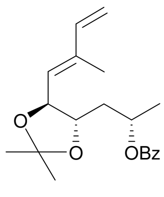


(1S)-2-{(4S,5S)-2,2-dimethyl-5-[(1E)-2-methylbut-1-en-3-yn-1-yl]-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.75): 2.74 (180 mg, 0.45 mmol) was dissolved in dry THF (2.2 mL) and TBAF (1.0 M in THF, 0.90 mL, 0.90 mmol, 2.0 eq) was added at 0 ^{o}C. The resulting solution was stirred under argon for 1 h and then quenched with aq. NH$_4$Cl (sat.). The aqueous layer was extracted with Et$_2$O (3x) and the combined organic layers were washed with brine (sat.), dried (Na$_2$SO$_4$) and concentrated. **2.75** (127 mg, 0.39 mmol, 86%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 97:3 to 95:5). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.35, 1.39 (2s, 6H, CMe$_2$), 1.40 (d, J = 6.4 Hz, 3H, C9-H$_3$), 1.81 (ddd, J = 3.6, 6.4, 14.0 Hz, 1H, C7-H), 1.89 (d, J = 1.2 Hz, 3H, C3-CH$_3$), 2.06 (ddd, J = 6.4, 8.4, 14.0 Hz, 1H, C7-H'), 2.86 (s, 1H, C1-H), 3.84 (ddd, J = 3.6, 8.4, 8.4 Hz, 1H, C6-H), 4.36 (t, J = 8.4 Hz, 1H, C5-H), 5.32 (m, 1H, C8-H), 5.81 (dd, J = 1.2, 9.2 Hz, 1H, C4-H), 7.40-7.57 (m, 3H, Bz-H), 8.03 (m, 2H, Bz-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 17.8 (q), 19.9 (q), 26.7 (q), 27.1 (q), 37.6 (t), 69.0 (d), 77.0 (d), 77.6 (d), 85.2 (s), 109.1 (s), 123.2 (s), 128.1

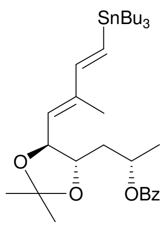


(d), 129.4 (d), 130.7 (s), 132.6 (d), 134.0 (d) 165.8 (s) ppm. MS(EI) for $C_{20}H_{24}O_4$: m/z = 328 [M^+], HRMS calcd for $C_{20}H_{24}O_4$: 328.167, found: 328.167.

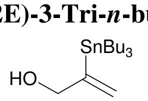
(1S)-2-{(4S,5S)-2,2-dimethyl-5-[(1E)-2-methylbuta-1,3-dien-1-yl]-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.76): ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.39 (m, 9H, C9-H_3, CMe_2), 1.81 (ddd, J = 4.0, 6.4, 14.0 Hz, 1H, C7-H), 1.84 (s, 3H, C3-CH_3), 2.08 (m, 1H, C7-H'), 3.83 (ddd, J = 3.6, 8.4, 8.4 Hz, 1H, C6-H), 4.47 (t, J= 8.8 Hz, 1H, C5-H), 5.08 (d, J = 10.4 Hz, 1H, C1-H), 5.23 (d, J = 17.6 Hz, 1H, C1-H'), 5.30 (m, 1H, C8-H), 5.38 (d, J = 8.8 Hz, 1H, C4-H), 6.32 (dd, J = 10.8, 17.2 Hz, 1H, C2-H), 7.43 (t, J = 7.6 Hz, 2H, Bz-H), 7.55 (t, J = 7.6 Hz, 1H, Bz-H), 8.03 (d, J = 7.6 Hz, 2H, Bz-H) ppm.



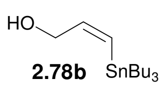
(1S)-2-{(4S,5S)-2,2-dimethyl-5-[(1E,3E)-2-methyl-4-tri-*n*-butylstannanyl-buta-1,3-dien-1-yl]-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.77): CuCN (5.1 mg, 57 µmol, 1.1 eq) was suspended in dry THF (0.27 mL) under argon and *n*-BuLi (1.6 M in *n*-hexane, 71 µl, 114 µmol, 2.2 eq) was added at -40 °C giving a colorless solution after 20 min. The solution was then warmed to -30 °C and Bu_3SnH (31 µl, 114 µmol, 2.2 eq) was added resulting in a yellow solution. **2.75** (17 mg, 52 µmol) in THF (0.11 mL) was added to the cuprate and the mixture was stirred for 75 min. Even though TLC showed that conversion was not complete, the reaction was quenched with first MeOH and then aq. NH_4Cl (sat.)- aq. NH_3 (12.5%) 4:1 (v/v). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were dried (Na_2SO_4) and concentrated. Even though GC-MS showed only 1 product apart from starting material, **2.77** (5 mg, 8.1 µmol, 16%) was isolated in low yield after performing column chromatography twice (AlOx basic, *n*-pentane-EtOAc 39:1). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.89 (m, 15H, Bu-Sn), 1.19-1.54 (m, 21H, Bu-Sn, C9-H_3, CMe_2), 1.83 (m, 4H, C7-H, C3-CH_3), 2.05 (m, 1H, C7-H'), 3.84 (m, 1H, C6-H), 4.48 (m, 1H, C5-H), 5.32 (m, 3H, C1-H, C4-H, C8-H), 6.39 (dd, J = 19.2, J ^{1}H-^{117}Sn = J ^{1}H-^{119}Sn = 72.3 Hz, 1H, C2-H), 7.43 (m, 2H, Bz-H), 7.55 (m, 1H, Bz-H), 8.03 (d, J = 8.1 Hz, 2H, Bz-H) ppm.



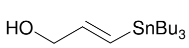
(2E)-3-Tri-*n*-butylstannanylprop-2-en-1-ol (2.78c):[71] Bu_3SnH (6.00 ml, 22.3 mmol, 1.3 eq) and AIBN (0.28 g, 1.7 mmol, 10 mol%) were added to propargyl alcohol (1.00 mL, 17.1 mmol) and the resulting solution was heated under reflux (80 °C) for 2 h. The reaction mixture was cooled to room temperature and the product was then purified by column chromatography (*n*-hexane-Et_2O 95:5 to 4:1) to give a mixture of **2.78a** and **2.78b** (1.25 g, 3.60 mmol, 21.0%, less polar) and pure **2.78c** (2.90 g, 8.35 mmol, 48.9%, more polar) as colorless liquids. ^{1}H-NMR **2.78c** ($CDCl_3$, 300 MHz) δ = 0.78-1.00 (m, 15H), 1.24-1.60 (m, 12H), 4.17 (brs, 2H, CH_2OH), 6.08-6.31 (m, 2H) ppm.



2.78a

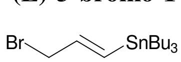


2.78b



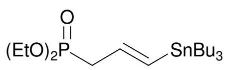
2.78c

(E)-3-bromo-1-tri-*n*-butylstannylpropene (2.79):[72] **2.78c** (2.15 g, 6.19 mmol) was dissolved in acetonitrile (57 mL) and PPh_3 (3.25 g, 12.4 mmol, 2.0 eq), CBr_4 (2.47 g, 7.45 mmol, 1.15 eq) and 2,6-lutidine (144 µl, 1.23 mmol, 0.20 eq) were added. The resulting solution was stirred for 15 min under argon, after which



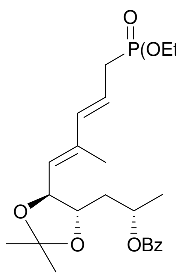
TLC (n-hexane-Et_2O 4:1) showed the reaction to be complete. The reaction mixture was poured into aq. $NaHCO_3$ (sat., 70 mL) and extracted with n-hexane (3 x 70 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated. Purification by column chromatography (n-hexane-Et_2O 99:1) gave **2.79** (1.88 g, 4.59 mmol, 74.2%) as a colorless liquid. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.80-1.02 (m, 15H), 1.22-1.60 (m, 12H), 3.96 (d, J = 6.6 Hz, 2H, CH_2Br), 6.12 (dt, J = 6.6, 18.9 Hz, 1H, C2-H), 6.29 (m, 1H, C1-H) ppm.

(3-Tri-*n*-butylstannanyl-allyl)-phosphonic acid diethyl ester (2.80):[72] To a solution of



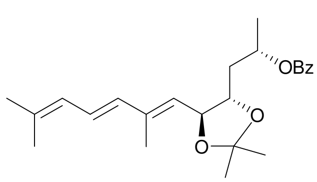
2.79 (0.26 g, 0.63 mmol) in DMF (4.2 mL) were added NaH (50% in oil, 152 mg, 3.17 mmol, 5.0 eq) and a solution of diethyl phosphite (0.40 mL, 3.10 mmol, 4.9 eq) in DMF (4.2 mL) at 0 °C. The resulting mixture was warmed to room temperature and stirred under argon for 3 h, after which it was poured into water (50 mL). The aqueous layer was extracted with Et_2O (3 x 50 mL) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. **2.80** (172 mg, 0.37 mmol, 58%) was isolated as a colorless oil after purification by column chromatography (n-hexane-EtOAc 4:1 to 1:1). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.88 (t, J = 7.5 Hz, 15H), 1.30 (m, 12H), 1.48 (m, 6H), 3.96 (dd, J = 6.6, 21.6 Hz, 2H, CH_2P), 4.09 (m, 4H, $CH_3\mathit{CH_2}OP$), 5.90 (m, 1H, C2-H), 6.15 (dd, J = 4.5, 18.6 Hz, 1H, C3-H) ppm.

(1S)-2-{(4S,5S)-5-[(1E,3E)-5-(diethoxyphosphoryl)-2-methyl-penta-1,3-dien-1-yl]-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.81):



$Pd(CH_3CN)_2Cl_2$ (9.0 mg, 10 mol%) was added to a solution of **2.38a** (150 mg, 0.35 mmol) in DMF (2.3 mL) and stirred for 2 min, after which **2.80** (179 mg, 0.38 mmol, 1.1 eq) in DMF (0.80 mL) was added. The resulting solution was stirred overnight under argon and then quenched with brine (sat.). The product was extracted with Et_2O (3x) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. **2.81** (110 mg, 0.23 mmol, 65%) was isolated as a colorless oil after column chromatography (n-pentane-EtOAc 4:1 to 1:2). ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.30 (t, J = 7.2 Hz, 6H, $\mathit{CH_3}CH_2$), 1.37 (m, 9H, C10-H_3, CMe_2), 1.75-1.85 (m, 1H, C8-H), 1.82 (s, 3H, C4-CH_3), 2.06 (m, 1H, C8-H'), 2.62 (dd, J = 7.2, 22.2 Hz, 2H, CH_2P), 3.80 (ddd, J = 3.9, 8.1, 8.1 Hz, 1H, C7-H), 4.08 (m, 4H, $CH_3\mathit{CH_2}$), 4.44 (t, J= 8.7 Hz, C6-H), 5.29 (m, 2H, C5-H, C9-H), 5.64 (m, 1H, C2-H), 6.12 (dd, J = 4.8, 15.6 Hz, 1H, C3-H), 7.42 (t, J = 7.5 Hz, 2H, Bz-H), 7.54 (t, J = 7.5 Hz, 1H, Bz-H), 8.01 (d, J = 7.8 Hz, 2H, Bz-H) ppm.

(1S)-2-{(4S,5S)-5-[(1E,3E)-2,6-dimethyl-hepta-1,3,5-trien-1-yl]-2,2-dimethyl-1,3-dioxolan-4-yl}-1-methylethyl benzoate (2.82):



2.81 (37 mg, 76 µmol) was dissolved in THF (0.42 mmol) and NaHMDS (1.0 M in THF, 80 µl, 80 µmol, 1.05 eq) was added at -78 °C upon which the solution turned orange/brown. The mixture was warmed to -40 °C over 45 min and then cooled again to -78 °C, after which **2.39c** (9.3 mg, 51 µmol) in THF (0,21 mL) was added. The reaction was allowed to warm to 0 °C over

2 h, at which point HMPA (14 µl) was added. The mixture was stirred for another h while warming to room temperature and was then quenched with brine (sat.). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were dried ($MgSO_4$), filtered and concentrated. Column chromatography (*n*-pentane-EtOAc-Et_3N 90:10:2) afforded starting material **2.39c** and **2.82**, but none of the desired cross-coupled product. ^{1}H-NMR **2.82** ($CDCl_3$, 300 MHz) δ = 1.39 (m, 9H, C1-H_3, CMe_2), 1.75-1.85 (m, 1H, C3-H), 1.80 (s, 3H, CH_3) 1.81 (s, 3H, CH_3), 1.88 (s, 3H, CH_3), 2.08 (m, 1H, C3-H'), 3.81 (ddd, *J* = 4.2, 8.4, 8.4 Hz, 1H, C4-H), 4.47 (t, *J*= 8.7 Hz, C5-H), 5.30 (m, 2H, C2-H, C6-H), 5.85 (d, *J* = 10.8 Hz, 1H, C10-H), 6.07 (d, *J* = 15.3 Hz, 1H, C8-H), 6.44 (dd, *J* = 10.8, 15.3 Hz, 1H, C9-H), 7.42 (t, *J* = 7.5 Hz, 2H, Bz-H), 7.54 (t, *J* = 7.8 Hz, 1H, Bz-H), 8.01 (d, *J* = 7.5 Hz, 2H, Bz-H) ppm.

2.7 References

[1] George, K. M.; Chatterjee, D.; Gunawardana, G.; Welty, D.; Hayman, J.; Lee, R.; Small, P. L. C. *Science* **1999**, *283*, 854-857.
[2] MacCallum P.; Tolhurst, J. C.; Buckle, G.; Sissons H. A. *J. Pathol. Bacteriol.* **1948**, *60*, 93-122.
[3] Van der Werf, T. S.; Stienstra, Y.; Johnson, R. C.; Phillips, R.; Adjei, O.; Fleischer, B.; Wansbrough-Jones, M. H.; Johnson, P. D. R.; Portaels, F.; Van der Graaf, W. T. A.; Asiedu, K. *Bull. World Health Organ.* **2005**, *83*, 785-791.
[4] Johnson, P. D. R.; Stinear, T.; Small, P. L. C.; Plushke, G.; Merritt, R. W.; Portaels, F.; Huygen, K.; Hayman, J. A.; Asiedu, K. *PloS Medicine*, **2005**, *2*, 282-286.
[5] Anonymous, In *Proceedings of the International Conference on Buruli Ulcer Control and Research*, ed. Asiedu, K.; Raviglione, M.; Scherpbier, R., Geneva: World Health Organization, **1998**, 5-10.
[6] World Health Organization, Buruli Ulcer: History and Background. Available: http://www.who.int/gtb-buruli/. Accessed 10 august 2005.
[7] Van der Werf, T.S.; Van der Graaf, W. T. A.; Tappero, J. W.; Asiedu K. *Lancet* **1999**, *354*, 1013-1018.
[8] a) Marsollier, L.; Robert, R.; Aubry, J.; Saint Andre, J. P.; Kouakou, H.; Legras, P.; Manceau A. L.; Mahaza, C.; Carbonnelle, B. *Appl. Environ. Microbiol.* **2002**, *68*, 4623-4628; b) Marsollier, L.; Aubry, J.; Saint Andre, J. P.; Robert, R.; Legras, P.; Manceau A. L.; Bourdon, S.; Audrain, C.; Carbonnelle, B. *Pathol. Biol.* **2003**, *51*, 490-495; c) Marsollier, L.; Aubry, J.; Coutanceau, E.; Saint Andre, J. P.; Small, P. L. C.; Milon G.; Legras, P.; Guadagnini, S.; Carbonelle, B.; Cole, S. T. *Cell. Microbiol.* **2005**, *7*, 935-943.
[9] a) Portaels, F.; Traore, H.; De Ridder, K.; Meyers, W. M. *Antimicrob. Agents Chemother.* **1998**, *42*, 2070-2073; b) Thangaraj, H. S.; Adjei, O.; Allen, B. W.; Portaels, F.; Evans, M. R. W.; Banerjee, D. K.; Wansbrough-Jones, M. H. *J. Antimicrob. Chemother.* **2000**, *45*, 231-233.
[10] Goutzamanis, J. J.; Gilbert, G. L. *Clin. Infect. Dis.* **1995**, *21*, 1186-1192.
[11] a) Dega, H.; Bentoucha, A.; Robert, J.; Jarlier, V.; Grosset, J. *Antimicrob. Agents Chemother.* **2002**, *46*, 3193-3196; b) Marsollier, L.; Prévot, G.; Honoré, N.; Legras, P.;

Manceau, L.; Payan, C.; Kouakou, H.; Carbonnelle, B. *Int. J. Antimicrob. Agents* **2003**, *22*, 562-566.

[12] a) The Uganda Buruli Group, *Lancet* **1969**, *293*, 111-115; b) Smith, P. G.; Revill, W. D. L.; Lukwago, E.; Rykushin, Y. P.; Lukwago, E.; Rykushin, Y. P. *Trans. R. Soc. Trop. Med. Hyg.* **1976**, *70*, 449-457; c) Portaels, F.; Aguiar, J.; Debacker, M.; Guedenon A.; Steunou C.; Zinsou, C.; Meyers, W. M. *Infect. Immun.* **2004**, *72*, 62-65; d) Huygen K. *Expert Rev. Vaccines* **2003**, *2*, 561-569.

[13] a) Benowitz, A. B.; Fidanze, S.; Small, P. L. C.; Kishi, Y. *J. Am. Chem. Soc.* **2001**, *123*, 5128-5129; b) Fidanze, S.; Song, F.; Szlosek-Pinaud, M.; Small, P. L. C.; Kishi, Y. *J. Am. Chem. Soc.* **2001**, *123*, 10117-10118.

[14] a) Hong, H.; Gates, P. J.; Staunton, J.; Stinear, T.; Cole, S. T.; Leadlay, P. F.; Spencer, J. B. *Chem. Comm.* **2003**, 2822-2823; b) Mve-Obiang, A.; Lee, R. E.; Portaels, F.; Small, P. L. C *Infect. Immun.* **2003**, *71*, 774; c) Mve-Obiang, A.; Lee, R. E.; Umstot, E. S.; Trott, K. A.; Grammer, T. C.; Parker, J. M.; Ranger, B. S.; Grainger, R.; Mahrous, E. A.; Small. P. L. C. *Infect. Immun.* **2005**, *73*, 3307-3312; d) Hong, H.; Spencer, J. B.; Porter, J. L.; Leadlay, P. F.; Stinear, T. *ChemBioChem* **2005**, *6*, 643-648.

[15] Stinear, T. P. *Report of the 8th annual WHO advisory group meeting on Buruli ulcer*, **2005**, in press.

[16] Stinear, T. P.; Mve-Obiang, A.; Small, P. L. C.; Frigui, W.; Pryor, M. J.; Brosch, R.; Jenkin, G. A.; Johnson, P. D. R.; Davies, J. K.; Lee, R. E.; Adusumilli, S.; Garnier, T.; Haydock, S. F.; Leadlay, P. F.; Cole, S. T. *Proc. Natl. Acad. Sci.* **2004**, *101*, 1345-1349.

[17] Staunton, J.; Wilkinson, B.; *Chem. Rev.* **1997**, *97*, 2611-2629.

[18] Stinear, T. P.; Hong, H.; Frigui, W.; Pryor, M. J.; Brosch, R.; Garnier, T.; Leadlay, P. F.; Cole, S. T. *J. Bacteriol.* **2005**, *187*, 1668-1676.

[19] Cadapan, L. D.; Arslanian, R. L.; Carney, J. R.; Zavala, S. M.; Small, P. L. C.; Licari, P. *Microbiol. Lett.* **2001**, *205*, 385-389.

[20] a) Song, F.; Fidanze, S.; Benowitz, A. B.; Kishi, Y. *Org. Lett.* **2002**, *4*, 647-650; b) Judd, T. C.; Bischoff, A.; Kishi, Y.; Adusumilli, S.; Small, P. L. C. *Org. Lett.* **2004**, *6*, 4901-4904.

[21] Gurjar, M. K.; Cherian, J. *Heterocycles* **2001**, *55*, 1095-1103.

[22] Brown, H. C.; Bhat, K. S. *J. Am. Chem. Soc.* **1986**, *108*, 5919-5923.

[23] Hermitage, S.A.; Roberts, S.M.; Watson, D. J. *Tetrahedron Lett.* **1998**, *39*, 3567-3570.

[24] Dounay, A. B.; Urbanek, R. A.; Frydrychowski, V. A.; Forsyth, C. J. *J. Org. Chem.* **2001**, *66*, 925-938.

[25] Part of this work has been published: Van Summeren, R. P.; Feringa, B. L.; Minnaard, A. J. *Org. Biomol. Chem.* **2005**, *3*, 2524-2533.

[26] (a) Morel, Th.; Verkade, P. E. *Rec. Trav. Chim. Pays-Bas* **1949**, *68*, 619-638; (b) Morel, Th.; Verkade, P. E. *Rec. Trav. Chim. Pays-Bas* **1951**, *70*, 35-49.

[27] Wenkert, E.; Khatuya, H.; Klein, P.S. *Tetrahedron Lett.* **1999**, *40*, 5171-5174.

[28] Gibson, A. W.; Humphrey, G. R.; Kennedy, D. J.; Wright, S. H. B. *Synthesis* **1991**, 414-416.

[29] Borhan, B.; Souto, M. L.; Um, J. M.; Zhou, B.; Nakanishi, K. *Chem. Eur. J.* **1999**, *5*, 1172-1175.

[30] Redlich, H.; Schneider, B.; Hoffmann, R. W.; Geueke, K. -J. *Liebigs Ann. Chem.* **1983**, 393-411.
[31] Sahai, P.; Chawla, M.; Vishwakarma, R. A. *J. Chem. Soc., Perkin Trans. 1* **2000**, 1283-1290.
[32] Jennings, H. J.; Jones, J. K. N. *Can. J. Chem.* **1963**, *41*, 1151-1159.
[33] Garegg, P. J.; Johansson, R.; Samuelsson, B. *Synthesis* **1984**, 168-170.
[34] Vogel, C.; Bergemann, C.; Ott, A. J.; Lindhorst, T. K.; Thiem, J.; Dahlhoff, W. V.; Hallgren, C.; Palcic, M. M.; Hindsgaul, O. *Liebigs Ann./ Recueil* **1997**, 601-612.
[35] Miljkovic, M.; Dropkin, D.; Hagel, P.; Habash-Marino, M. *Carbohydr. Res.* **1984**, *128*, 11-20.
[36] Redlich, H.; Sudau, W.; Szardenings, A. K.; Vollerthun, R. *Carbohydr. Res.* **1992**, *226*, 57-78.
[37] Corey, E. J.; Fuchs, P. L. *Tetrahedron Lett.* **1972**, *13*, 3769-3772.
[38] Yamada, H.; Aoyagi, S.; Kibayashi, C. *J. Am. Chem. Soc.* **1996**, *118*, 1054-1059.
[39] Aurrecoechea, J. M.; López, B.; Arrate, M. *J. Org. Chem.* **2000**, *65*, 6493-6501.
[40] (a) Marshall, J. A.; Yanik, M. M. *J. Org. Chem.* **1999**, *64*, 3798-3799; (b) Paik, S.; Carmeli, S.; Cullingham, J.; Moore, R. E.; Patterson, G. M. L.; Tius, M. A. *J. Am. Chem. Soc.* **1994**, *116*, 8116-8125.
[41] Zhang, H. X.; Guibé, F.; Balavoine, G. *J. Org. Chem.* **1990**, *55*, 1857-1867.
[42] Semmelhack, M. F.; Hooley, R. J. *Tetrahedron Lett.* **2003**, *44*, 5737-5739.
[43] Jennings, H. J.; Jones, J. K. N. *Can. J. Chem.* **1965**, *43*, 2372-2386.
[44] Tutwiler, F. B.; McKee, R. L. *J. Am. Chem. Soc.* **1954**, *76*, 6342-6344.
[45] Dean, D. M.; Szarek, W. A.; Jones, J. K. N. *Carbohydr. Res.* **1974**, *33*, 383-386.
[46] Percival, E. E.; Percival, E. G. V. *J. Chem. Soc.* **1950**, 690-691.
[47] Barton, D. H. R.; McCombie, S. W. *J. Chem. Soc., Perkin Trans. 1* **1975**, 1574-1585.
[48] Mulard, L. A.; Claudemans, C. P. J. *Carbohydr. Res.* **1995**, *274*, 209-222.
[49] Mathé, C.; Imbach, J. -L.; Gosselin, G. *Carbohydr. Res.* **2000**, *323*, 226-229.
[50] Ashton, P. R.; Brown, C. L.; Menzer, S.; Nepogodiev, S. A.; Stoddart, J. F.; Williams, D. J. *Chem. Eur. J.* **1996**, *2*, 580-591.
[51] Fernández, P.; Jiménez-Barbero, J.; Martín-Lomas, M.; Solís, D.; Díaz-Mauriño, T. *Carbohydr. Res.* **1994**, *256*, 223-244.
[52] Arroyo-Gómez, Y.; López-Sastre, J. A.; Rodríguez-Amo, J. F.; Santos-García, M.; Sanz-Tejedor, M. A. *Tetrahedron: Asymm.* **1999**, *10*, 973-990.
[53] Sonogashira, K.; Yatake, T.; Tohda, Y.; Takahashi, S.; Hagihara, N. *J. Chem. Soc., Chem. Commun.* **1977**, *9*, 291-292.
[54] Lindlar, H. *Helv. Chim. Acta* **1952**, *35*, 446-456.
[55] Ivanova, D. I.; Eremin, S. V.; Shvets, V. I. *Tetrahedron* **1996**, *52*, 9581-9588.
[56] (a) van Laren, M. W.; Elsevier, C. J. *Angew. Chem. Int. Ed.* **1999**, *38*, 3715-3717; (b) Sprengers, J. W.; Wassenaar, J.; Clement, N. D.; Cavell, K. J.; Elsevier, C. J. *Angew. Chem. Int. Ed.* **2005**, *44*, 2026-2029.
[57] (a) Suh, Y. –G.; Min, K. –H.; Lee, Y. -S.; Seo, S. -Y.; Kim, S. -H.; Park, H. -J. *Tetrahedron Lett.* **2002**, *43*, 3825-3828; (b) Brown, C. A.; Ahuja, V. K. *J. Org. Chem.* **1973**, *38*, 2226-2230.

[58] Boland, W.; Schroer, N.; Sieler, C.; Feigel, M. *Helv. Chim. Acta* **1987**, *70*, 1025-1040.
[59] (a) Souto, M. L.; Um, J.; Borhan, B.; Nakanishi, K. *Helv. Chim. Acta* **2000**, *83*, 2617-2628; (b) Fujimoto, Y.; Ishihara, J.; Maki, S.; Fujioka, N.; Wang, T.; Furuta, T.; Fishkin, N.; Borhan, B.; Berova, N.; Nakanishi, K. *Chem. Eur. J.* **2001**, *7*, 4198-4204; (c) Nesnas, N.; Rando, R. R.; Nakanishi, K. *Tetrahedron* **2002**, *58*, 6577-6584. (d) Quesada, E.; Acuña, A. U.; Amat-Guerri, F. *Eur. J. Org. Chem.* **2003**, 1308-1318; (e) Rodríguez, A. R.; Spur, B. W. *Tetrahedron Lett.* **2001**, *42*, 6057-6060.
[60] Zeng, X.; Zeng, F.; Negishi, E. I. *Org. Lett.* **2004**, *6*, 3245-3248.
[61] (a) Ahmad, R.; Weedon, B. C. L. *J. Chem. Soc.* **1953**, 3286-3294; (b) Isler, O.; Guex, W.; Ruegg, R.; Ryser, G.; Saucy, G.; Schwieter, U.; Walter, M.; Winterstein, A. *Helv. Chim. Acta* **1959**, *42*, 864-871; (c) Mead, D; Asato, A. E.; Denny, M.; Liu, R. S. H.; Hanzawa, Y.; Taguchi, T.; Yamada, A.; Kobayashi, N.; Hosoda, A.; Kobayashi, Y. *Tetrahedron Lett.* **1987**, *28*, 259-262.
[62] (a) Lipshutz, B. H.; Ellsworth, E. L.; Dimock, S. H.; Reuter, D. C. *Tetrahedron Lett.* **1989**, *30*, 2065-2068; (b) Le Ménez, P.; Fargeas, V.; Berque, I.; Poisson, J.; Ardisson, J.; Lallemand, J. –Y.; Pancrazi, A. *J. Org. Chem.* **1995**, *60*, 3592-3599; (c) Wada, A.; Fukunaga, K.; Ito, M. *Synlett* **2001**, 800-802.
[63] Hart, D. W.; Blackburn, T. F.; Schwartz, J. *J. Am. Chem. Soc.* **1975**, *97*, 679-680.
[64] For a review on metal-catalyzed hydrostannations: Smith, N. D.; Mancuso, J.; Lautens, M. *Chem. Rev.* **2000**, *100*, 3257-3282.
[65] (a) Cochran, J. C.; Bronk, B. S.; Terrence, K. M.; Phillips, H. K. *Tetrahedron Lett.* **1990**, *31*, 6621-6624; (b) Rossi, R.; Carpita, A.; Cossi, P. *Tetrahedron Lett.* **1992**, *33*, 4495-4498.
[66] Seyferth, D.; Heeren, J. K.; Singh, G.; Grim, S. O.; Hughes, W. B. *J. Organomet. Chem.* **1966**, *5*, 267-274.
[67] Van Horn, D. E.; Negishi, E. -i. *J. Am. Chem. Soc.* **1978**, *100*, 2254-2256.
[68] Shi, J. -C.; Zeng, X.; Negishi, E. -i. *Org. Lett.* **2003**, *5*, 1825-1828.
[69] For reviews on organozirconium chemistry: (a) Wipf, P.; Jahn, H. *Tetrahedron* **1996**, *52*, 12853-12910; (b) Negishi, E. -i. *Dalton Trans.* **2005**, 827-848.
[70] For a review on the use of Stille-type couplings in conjugated systems see: Domínguez, B.; Iglesias, B.; de Lera, A. R. *Tetrahedron* **1999**, *55*, 15071-15098.
[71] Jung, M. E.; Light, L. A. *Tetrahedron Lett.* **1982**, *23*, 3851-3854.
[72] Smith, A. B.; Wan, Z. *J. Org. Chem.* **2000**, *65*, 3738-3753.
[73] Pozsgay, V.; Neszmélyi, A. *Carbohydr. Res.* **1980**, *85*, 143-150.

Chapter 3
Enantiopure Saturated Isoprenoid Building Blocks

3.1 Introduction

The chiral saturated isoprenoid unit is a central structural element in many natural products including pheromones,[1] vitamins,[2] marine natural products,[3] and archaebacterial lipids[4] (Figure 3.1).

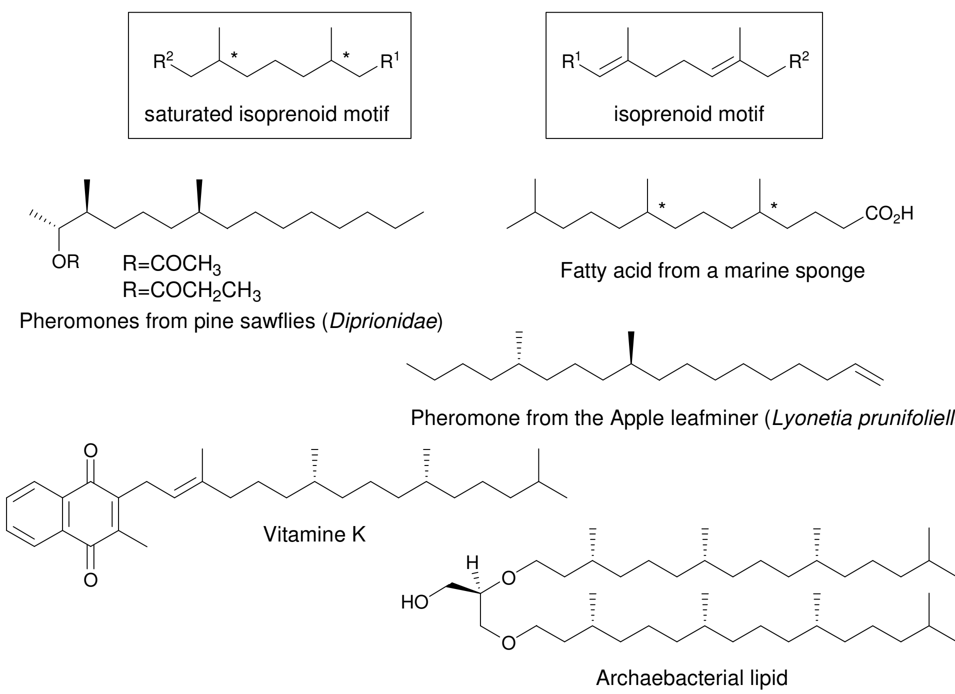


Figure 3.1 *Some natural products containing the saturated isoprenoid motif.*

As a consequence, many efforts have been devoted to the synthesis of versatile and enantiopure isoprenoid building blocks, which can be functionalized at both ends (Figure 3.2). In the quest for efficient and general synthetic protocols, organic chemists have adopted various strategies among which are chiral pool approaches, enzymatic protocols and chiral auxiliary based methods. Remarkably, until recently, asymmetric catalysis has received little attention in the synthesis of enantiopure isoprenoid building blocks.

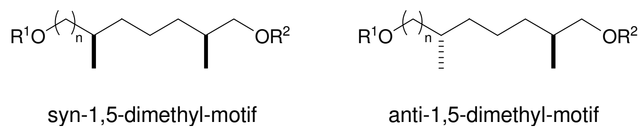
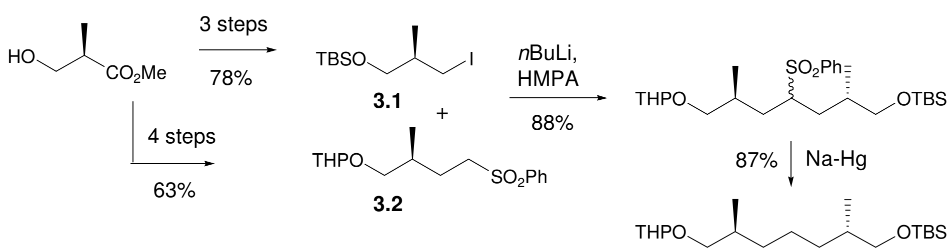


Figure 3.2 *Versatile enantiopure saturated isoprenoid building blocks*.

3.2 Previously reported strategies

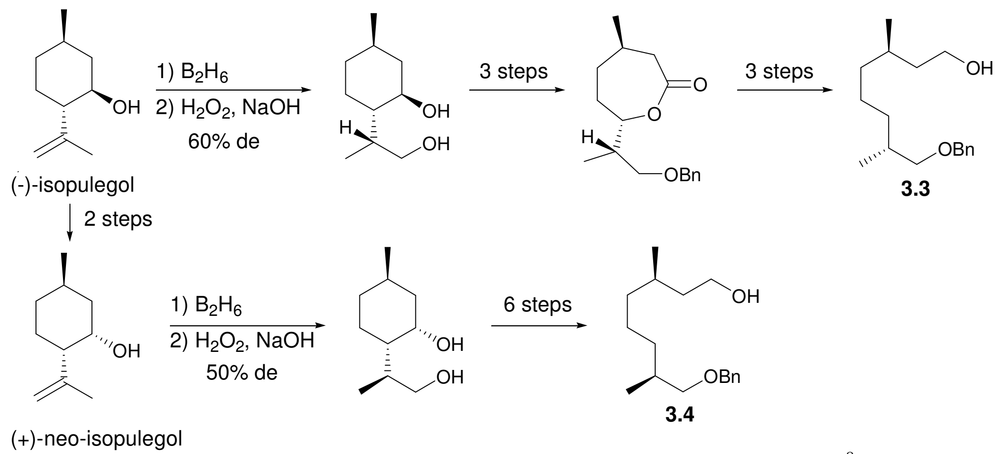
3.2.1 Chiral pool and substrate control strategies

The most straightforward way to prepare chiral building blocks like the ones depicted in Figure 3.2, is to couple two enantiopure fragments. This approach has been used by Eguchi[4d] to synthesize the *syn*-motif and by Mori to make the *anti*-motif (Scheme 3.1).[5] Cross-coupling is achieved by reaction of alkyl halide **3.1** with the anion derived from sulfon **3.2**, followed by reductive removal of the phenylsulfonyl group with sodium amalgam. Similar strategies using Wittig-[6] or Horner-Wadsworth-Emmons[7] cross-coupling protocols followed by hydrogenation of the resulting olefin have also been used.



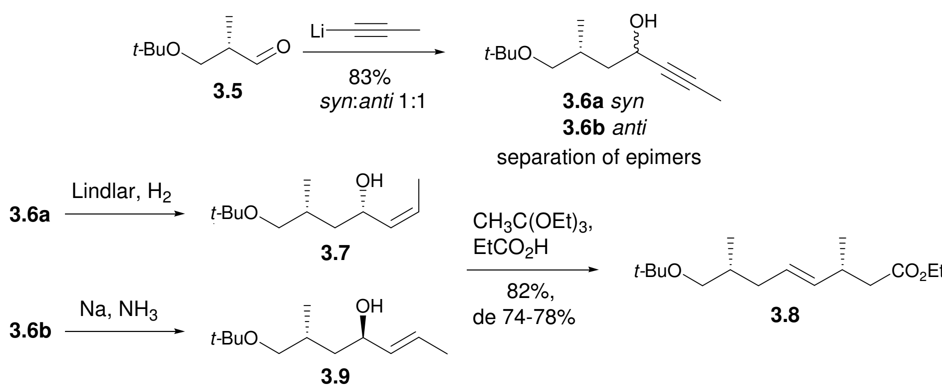
Scheme 3.1 *Synthesis of the anti-1,5-dimethyl-motif by Mori*.[5]

Ferreira *et al*. developed a chiral pool strategy starting from (-)-isopulegol to obtain the *anti*-motif and from (+)-neo-isopulegol to prepare the *syn*-motif (Scheme 3.2).[8] In the key step, the second methyl center is introduced using a hydroboration reaction as described by Schulte-Elte.[9] The stereochemical outcome of this step relies on the configuration of the hydroxyl moiety in the ring. The diastereoselectivity is modest (50-60% de), but the isomers can be separated by conventional column chromatography. The enantiopure building blocks **3.3** or **3.4** are obtained after six additional steps.



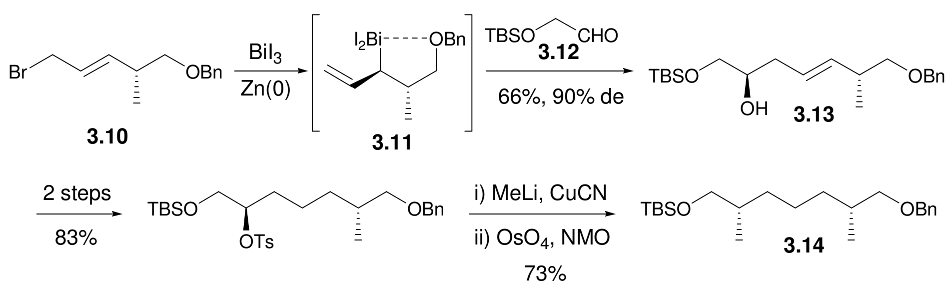
Scheme 3.2 *Synthesis from (-)-isopulegol and (+)-neoisopulegol.*[8]

One of the first procedures to selectively make the *syn*-motif was published by Cohen *et al*. in 1976.[10] They converted enantiopure aldehyde **3.5** into a diastereomeric mixture (1:1) of alcohols (**3.6**) and separated the epimers (Scheme 3.3). Subsequent partial hydrogenation of *syn*-alcohol **3.6a** to *cis*-alkene **3.7** with Lindlar catalyst set the stage for chirality transfer *via* a Claisen rearrangement. Heating of **3.7** in the presence of triethyl orthoacetate and catalytic propionic acid (Johnson-Faulkner conditions)[11] afforded the desired *syn*-product **3.8** (82%, 78% de). Interestingly, Birch reduction of *anti*-alcohol **3.6b** to *trans*-alkene **3.9** followed by a [3,3]-sigmatropic rearrangement under identical conditions gave **3.8** as well (74% de). Since the stereochemical outcome of the reaction seems to depend on the configuration of both the alcohol and the alkene, this method should also be capable of producing the *anti*-motif but the authors did not explore this.



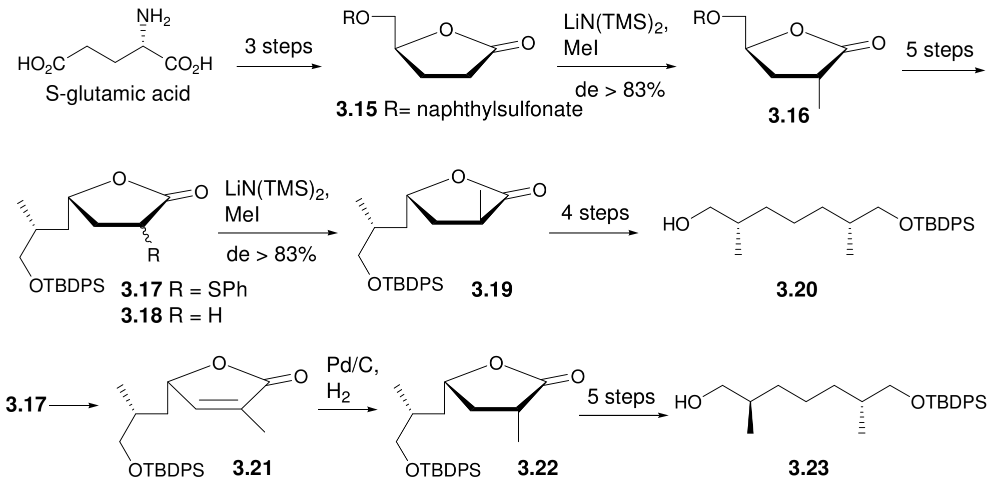
Scheme 3.3 *Chirality transfer via Claisen rearrangement.*[10]

A recently published method for the assembly of a building block containing the *syn*-motif uses remote stereocontrol of a methyl-substituent to selectively introduce a hydroxyl moiety in a 1,5-*anti*-fashion (Scheme 3.4).[12] It was proposed that the stereoselectivity results from reaction of allylic bromide **3.10** with bismuth (0) to form complex **3.11**, which gives olefin **3.13** (90% de) after reaction with aldehyde **3.12**. Reduction of the olefin and substitution of the hydroxyl moiety for a methyl group subsequently gave **3.14** in 3 steps.



Scheme 3.4 *Synthesis of the syn-motif by asymmetric induction followed by substitution.*[12]

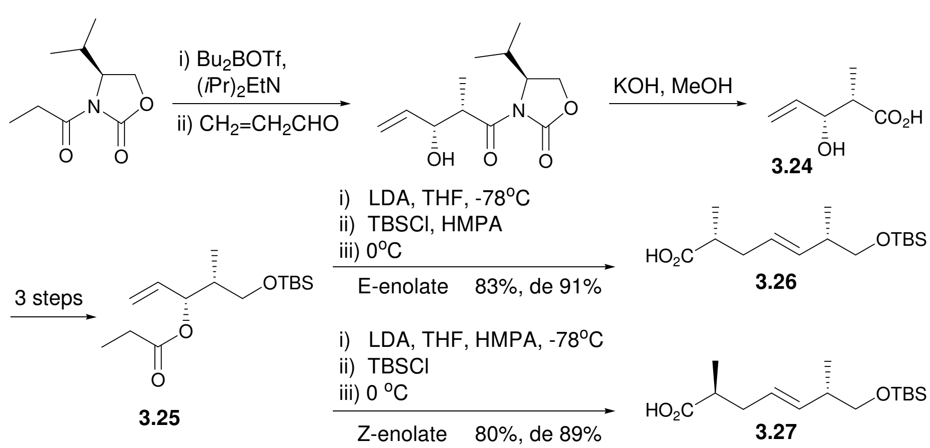
One of the first chiral pool-based methods capable of producing both the *syn*- and the *anti*-motif was developed by Hanessian *et al* in 1985.[13] In their approach, they used a substituted enantiopure butyrolactone derivative to control the stereochemistry of an alpha-methylation process (**3.15** to **3.16** Scheme 3.5). After chain extension and elaborate functional group manipulations they repeated this process (**3.18** to **3.19**) to introduce the second methyl group. Eventually, they obtained *syn*-building block **3.20** in fourteen steps from S-glutamic acid with an overall yield of 17% from **3.15**. A slight modification in the route also gave access to *anti*-building block **3.23** *via* hydrogenation of olefin **3.21**.



Scheme 3.5 *Substituted butyrolactone as a template for α-methylation.*[13]

3.2.2 Chiral auxiliary methods

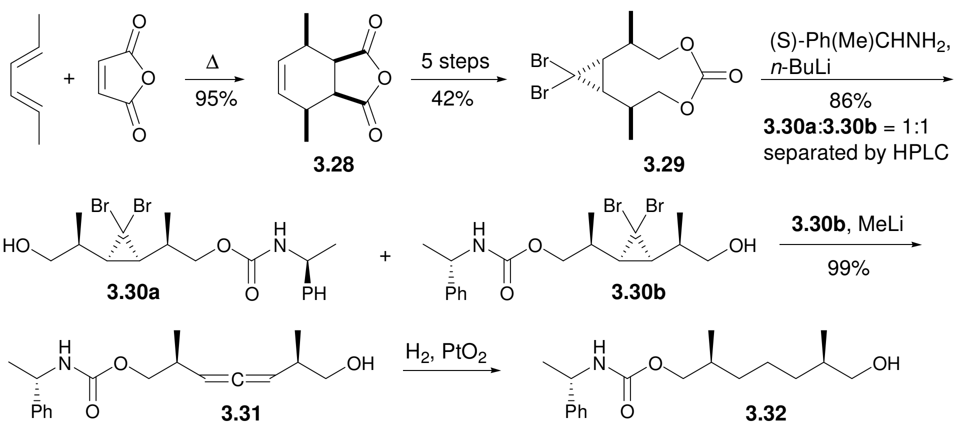
In 1988, Heathcock and Finkelstein employed a chiral auxiliary assisted aldol condensation in combination with chirality transfer *via* a Claisen rearrangement to prepare both the *syn*- and the *anti* motif from a common precursor (Scheme 3.6).[14]



Scheme 3.6 *Chiral auxiliary-assisted aldol condensation followed by transfer of chirality.*[14]

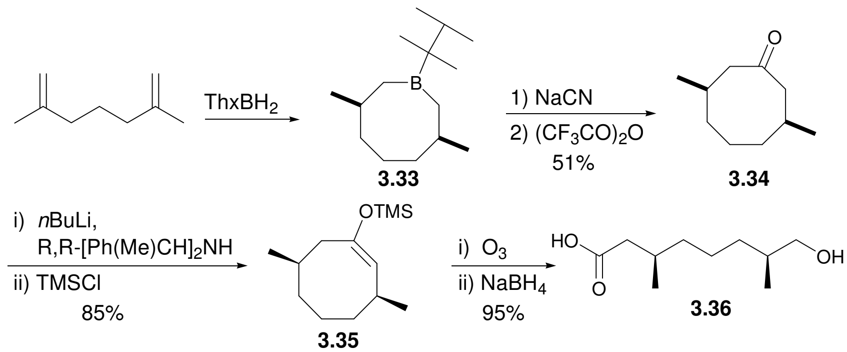
Ester **3.25** was synthesized in three steps from allylic alcohol **3.24**, which was itself obtained with high diastereo- and enantioselectivity using the Evans oxazolidone protocol.[15] Subsequently, **3.25** was selectively converted *via* a Claisen rearrangement into either **3.26** (*syn*, de 91%) or **3.27** (*anti*, de 89%) by formation of the E- or Z-enolate respectively as reported by Ireland and co-workers.[16]

Bérubé and Deslongchamps developed a method to synthesize the *syn*-motif, in which they use a Diels-Alder cyclization to control the relative stereochemistry of two methyl groups (**3.28**, Scheme 3.7).[17] Functional group manipulations subsequently furnish carbonate **3.29** in five steps, after which reaction with lithiated (*S*)-α-methylbenzylamine gives a separable mixture (1:1) of **3.30a** and **3.30b**. A final key element in their approach is the insertion of a methylene unit *via* allene formation and hydrogenation in order to mutate from a 1,4-*syn* to a 1,5-*syn* dimethyl relationship.



Scheme 3.7 *Diels-Alder reaction in combination with diastereomeric separation*.[17]

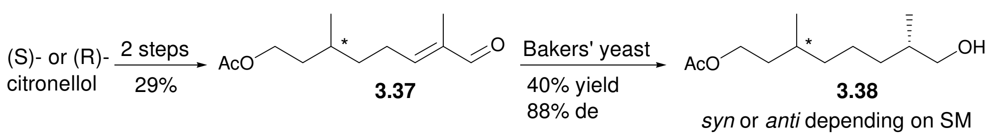
Another strategy to obtain the *syn*-motif was published in 1997 by Berkowitz and Wu (Scheme 3.8).[4c] They used a cyclic hydroboration/oxidation protocol on 2,6-dimethylhepta-1,6-diene to acquire meso-ketone **3.34** with high diastereoselectivity (>99% de).[18] Subsequent desymmetrization by deprotonation with a chiral base and quenching with trimethylsilyl chloride gave silyl enol ether **3.35** (97% ee). Finally, saturated isoprenoid building block **3.36** was isolated after oxidative ring opening and reductive work-up.



Scheme 3.8 *Cyclic hydroboration in combination with desymmetrization using a chiral base followed by ring opening.*[4c]

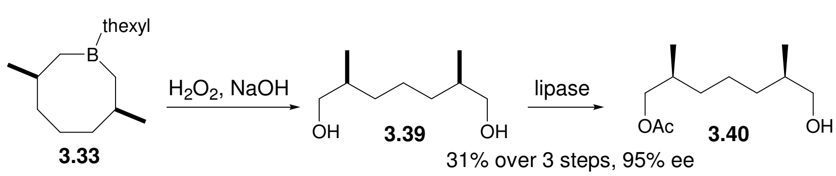
3.2.3 Enzymatic approaches

In 1985, Gramatica *et al*. combined a chiral pool strategy with an enzymatic approach for the preparation of both the *syn*- and the *anti*- isoprenoid motif.[19] Hydrogenation of optically active α-methyl-α,β-unsaturated aldehyde **3.37** in the presence of bakers' yeast resulted in the formation of 1,5-dimethylated acyclic unit **3.38** (Scheme 3.10) in good diastereoselectivity with an overall yield of 11%.



Scheme 3.9 *Enzymatic hydrogenation.*

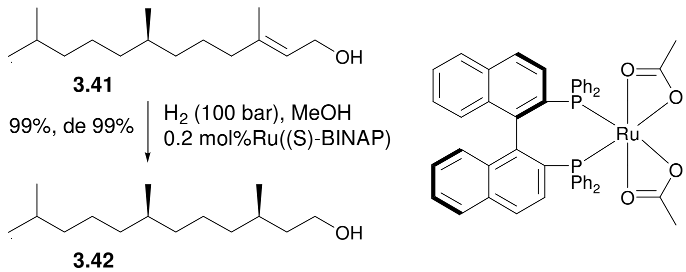
The enzymatic equivalent of Berkowitz' desymmetrization strategy (Scheme 3.8) was already developed by Chênevert and Desjardins in 1996 (Scheme 3.10).[20] They used a lipase for the selective mono-acetylation of *meso*-2,6-dimethyl-1,7-heptanediol (**3.39**), which was obtained *via* oxidative ring opening of borocane **3.33**.[18]



Scheme 3.10 *Enzymatic desymmetrization.*

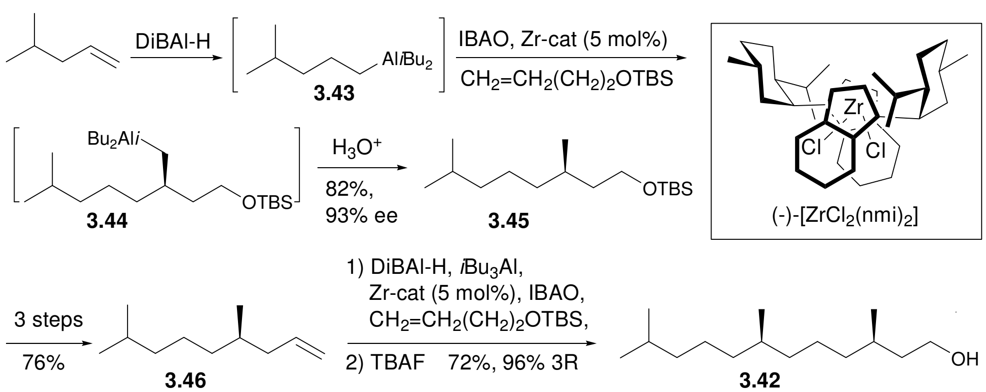
3.2.4 Catalytic asymmetric methods

In 1987, Noyori *et al*. synthesized C_{15}-chain **3.42** (99% de, Scheme 3.11) *via* catalytic asymmetric hydrogenation of allylic alcohol **3.41** using a Ru-BINAP complex.[21] The chiral center of enantiopure starting material **3.41** could have been introduced using the same catalyst, but the authors chose to employ the chiral pool in this particular case.



Scheme 3.11 Catalytic asymmetric hydrogenation with Ru-binap.[21]

More recently, Negishi and co-workers applied the Zr-catalyzed enantioselective carboalumination of alkenes to the preparation of **3.42** (Scheme 3.12).[22] The key step starts with the hydroalumination of a terminal alkene to give intermediate **3.43**. Subsequent addition of a second terminal alkene in the presence of (-)-[$ZrCl_2$(nmi)$_2$] (5 mol%) and isobutylaluminoxane (IBAO) results in enantioselective alkylalumination by **3.43** to form intermediate **3.44**, which gives rise to the desired methyl-branched product **3.45** (82%, ee 93%) after protonation. The second methyl center is introduced (96% 3R) by repetition of this process on terminal alkene **3.46**, which is obtained in three steps from **3.45**.



Scheme 3.12 *Hydroalumination/Zirconium-catalyzed alkylalumination protocol.*[22]

3.3 A general catalytic asymmetric method[23]

Even though an impressive number of methods for the synthesis of enantiopure saturated isoprenoid building blocks have been reported in the past (§ 3.2), there is still ample room for improvement. Some of the approaches discussed above are just too elaborate, while others are incapable of delivering all diastereomers or require a stoichiometric amount of chiral material. Moreover, the obtained levels of stereoselectivity are often far from perfect (ee or de < 98%), which necessitates tedious separations of diastereomers or enantiomers. As for the two asymmetric catalytic methods, it should be noted that in both cases only the *syn*-isoprenoid unit was synthesized and that one end of the molecule was restricted to a saturated alkyl chain. In order to avoid these constraints in synthetic flexibility and stereocontrol, we have focused on the development of a general catalytic method to access all 4 diastereoisomers of a versatile building block, that can be functionalized at both termini.

3.3.1 Strategy

Our approach is based on the Cu-phosphoramidite-catalyzed asymmetric conjugate addition (ACA) of dialkylzinc reagents to cyclic substrates, which was developed in our group in 1997.[24] We anticipated that this protocol, used iteratively in the ACA of Me_2Zn to cycloocta-2,7-dienone (**3.47**) followed by oxidative ring opening, would enable the rapid assembly of enantiopure *syn*- and *anti*-isoprenoid building blocks (Figure 3.3). However, quenching of the zinc enolate after the second 1,4-addition with a proton source would result in a meso compound (**3.49**) in case of the *cis*-adduct. To avoid a racemic product after ring opening, an alternative procedure was explored, involving *in situ* trapping of the zinc enolate as a silyl enol ether.

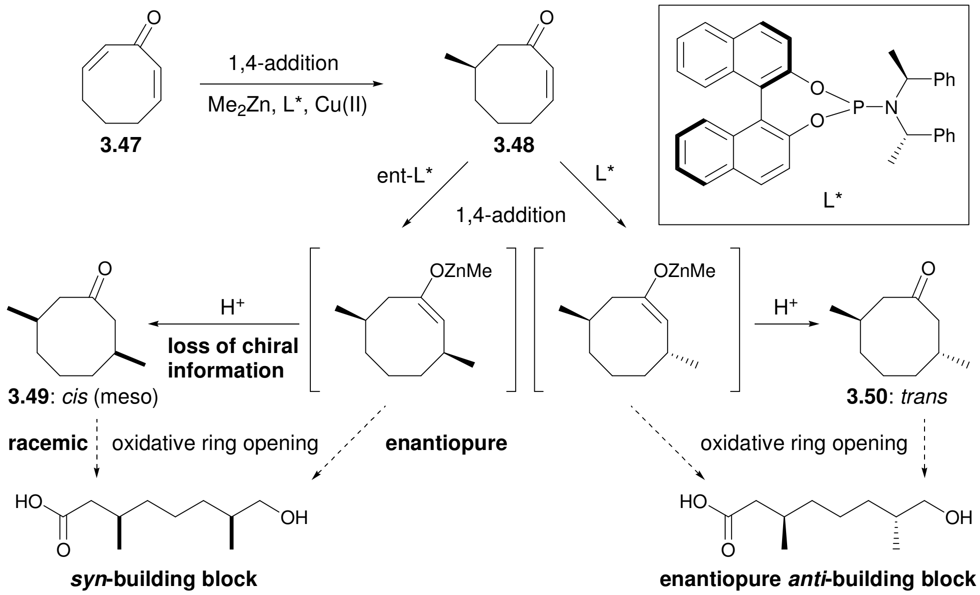


Figure 3.3 *General strategy and the problem of formation of a meso-compound.*

3.3.2 Synthesis

Cycloocta-2,7-dienone (**3.47**) was prepared from cyclooctanone in one step via oxidation with IBX (*o*-iodoxybenzoic acid) as described by Nicolaou et al. (Scheme 3.13).[25] The subsequent conjugate addition of Me_2Zn (1.5 eq) to **3.47** in the presence of copper(II) triflate (2.5 mol%) and chiral phosphoramidite L* (5 mol%)[26] yielded the desired product (7R)-**3.48** with excellent enantioselectivity (ee > 99%). However, due to the formation of side product **3.51** (33%) resulting from partial Michael addition of the intermediate zinc-enolate to **3.47**, the isolated yield amounted to a modest 65%. A range of conditions were tested to suppress this side reaction (Table 3.1), resulting in the isolation of (7R)-**3.48** in 85% yield (>99% ee) when the catalyst loading and the amount of Me_2Zn were doubled and the substrate was slowly added to the reaction mixture over 5 hours (entry 7).

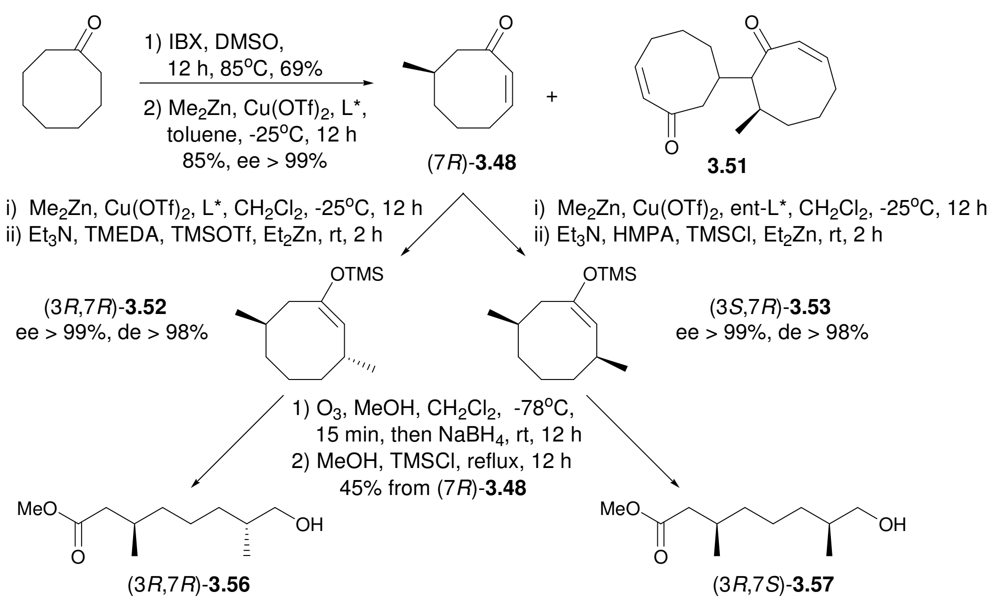
In the second 1,4-addition, Michael reaction of the zinc-enolate was sufficiently suppressed by slow addition of (7R)-**3.48**, allowing for the use of 2.5 mol% of catalyst and 1.5 equivalents of dimethylzinc. The stereochemical outcome of the reaction was solely determined by the nature of the phosphoramidite ligand, overruling any induction of the previously introduced chiral center. Use of L* resulted in the isolation of (3R,7R)-**3.50**

(>98% de) after quenching with a proton source, while ent-L* led to the formation of meso-**3.49** (>98% de) as determined by ^{1}H-NMR (§ 3.3.3).[4c]

Table 3.1 *Optimization of the 1,4-addition of Me_2Zn to cycloocta-2,7-dienone (***3.47***).*[a]

entry	catalyst loading (mol%)	Me_2Zn (eq)	Addition[b]	Temperature (°C)	Ratio[c] **3.48**:**3.51**
1	2.5	1.5	normal	-25	66 : 33
2	2.5	1.5	normal	-50	71 : 29[d]
3	2.5	1.5	reversed	-25	74 : 26
4	2.5	1.5	reversed	0	79 : 9 : 11[e]
5	5.0	3.0	normal	-25	76 : 24
6	5.0	5.0	reversed	-25	> 95 : 5
7	**5.0**	**3.0**	**reversed**	**-25**	**95 : 5**
8	2.5	5.0	reversed	-25	88 : 12

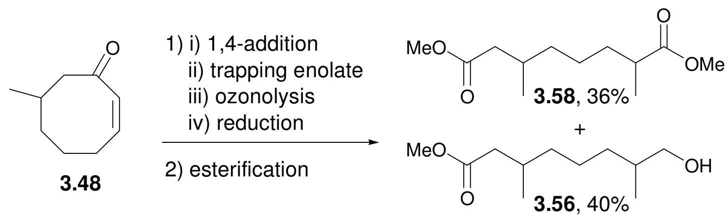
[a] ee > 99% under all conditions. [b] Normal addition means addition of Me_2Zn to a solution of **3.47**, while reversed addition denotes adding **3.47** to a solution of Me_2Zn. [c] Determined by GC-MS. [d] Incomplete conversion. [e] Second (unknown) side product present.



Scheme 3.13 *Synthesis of all diastereomers of an enantiopure building block.*

In situ trapping of the zinc-enolate with TMSOTf or TMSCl in the presence of triethylamine to obtain silyl enol ethers (3R,7R)-**3.52** and (3S,7R)-**3.53** typically required elongated reaction times and often resulted in low conversions.[27] Moreover, the cis-adduct appeared to be prone to racemization under those conditions, probably due to an equilibrium between the (meso-)ketone and the silyl enol ether. In order to change the reactive properties of the zinc-enolate, we explored different additives which are known to break up enolate clusters. In case of the *trans*-adduct, addition of Et_3N and TMEDA followed by quenching with TMSOTf, which was dried with Et_2Zn prior to use, resulted in quantitative formation of **3.52** within 2 hours. When HMPA was used instead of TMEDA and TMSOTf was replaced with the milder TMSCl, similar results were obtained for **3.53** without partial racemization (conversion > 95%, ee > 99%). Unfortunately, due to the volatility of the silyl enol ethers, the isolated yields remained low. This problem was partly overcome by changing the reaction solvent from toluene to dichloromethane, which did not adversely affect the stereochemical selectivity (de and/or ee). Ring opening of **3.52** and **3.53** was accomplished by ozonolysis and the resulting aldehyde was reduced upon work-up with sodium borohydride. The crude carboxylic acids (**3.54/3.55**) were then heated under reflux in MeOH in the presence of TMSCl to give the methyl esters (3R,7R)-**3.56** and (3R,7S)-**3.57** in an overall yield of 45% from (7R)-**3.48**. The enantiomers (3S,7S)-**3.56** and (3S,7R)-**3.57** were isolated in comparable yield and (stereochemical) purity from (7S)-**3.48** by using the opposite enantiomer of the ligand, i.e. ent-L* in place of L* and vice versa.

The conversion of silyl enol ethers **3.52** and **3.53** into methyl esters **3.56** and **3.57** was performed many times and during the course of time small alterations in the procedure led to the optimal protocol with high reproducibility, which is described above. In general, a trade-off seemed to exist between different factors influencing the eventual isolated yield of the methyl ester. When the conversion of **3.48** into carboxylic acid **3.54** or **3.55** was performed as a one pot procedure, **3.56/3.57** was isolated in 40% yield after esterification along with di-ester **3.58** (36%, Scheme 3.14). In this case the prime loss of product was caused by oxidation of the aldehyde during ozonolysis due to the presence of amines (Figure 3.4).[28] In contrast, extensive purification of the silyl enol ether by column chromatography and concentration resulted in a clean ozonolysis/reduction product, but part of the silyl enol ether was lost during rotary evaporation and/or because of decomposition on the silica.



Scheme 3.14 *Conjugate addition, Zn-enolate trapping and ozonolysis/reduction in one pot.*

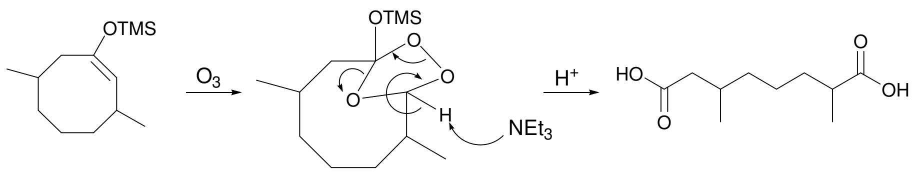


Figure 3.4 *Amine-mediated rearrangement to the dicarboxylic acid during ozonolysis.*[28]

3.3.3 Determination of the relative and absolute stereochemistry

Compounds **3.49** and **3.53** could be distinguished from their respective diastereomers **3.50** and **3.52** by ^{1}H-NMR (Figure 3.5). As expected, comparison with data from the literature[4c] proved that the methyl substituents of **3.49** and **3.53** were indeed in a *cis*-relationship to each other making the relative stereochemistry of **3.50** and **3.52** *trans* by default.

The absolute configurations of the enantiomers of *syn*-building block **3.57** were determined by comparison of their optical rotations with the literature value of (3R,7S)-**3.57**.[4c,29] From this information, the absolute configuratons of **3.48** and of the enantiomers of *anti*-building block **3.56** were deduced. This assignment was confirmed by comparison of the optical rotation of pheromone (10S,14S)-**3.67**, which was synthesized from *anti*-building block (3R,7R)-**3.56** (§ 3.4), to its literature value.[5]

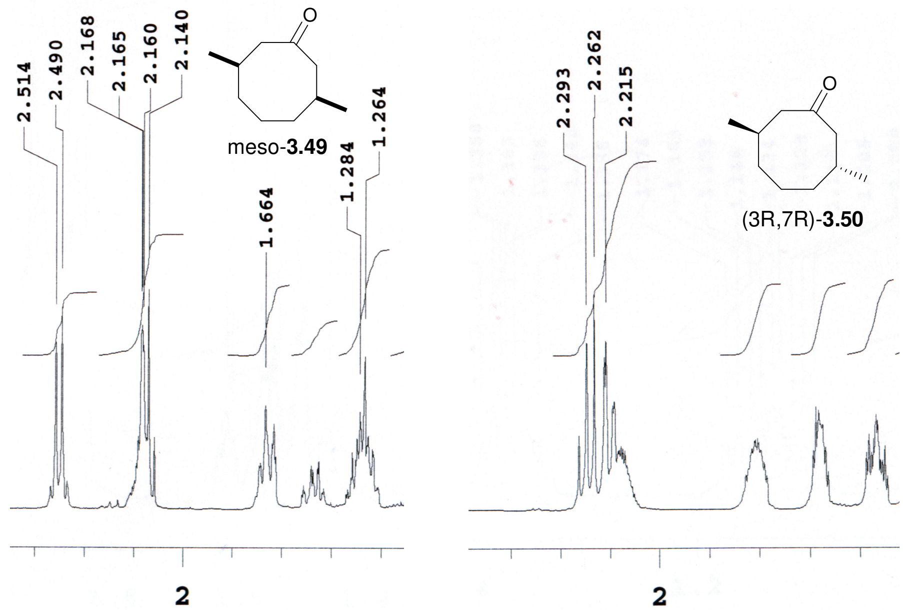


Figure 3.5 *Assignment of the relative stereochemistry of* ***3.49*** *and* ***3.50*** *by* ^{1}*H-NMR.*

3.4 Application in the total synthesis of apple leafminer pheromones

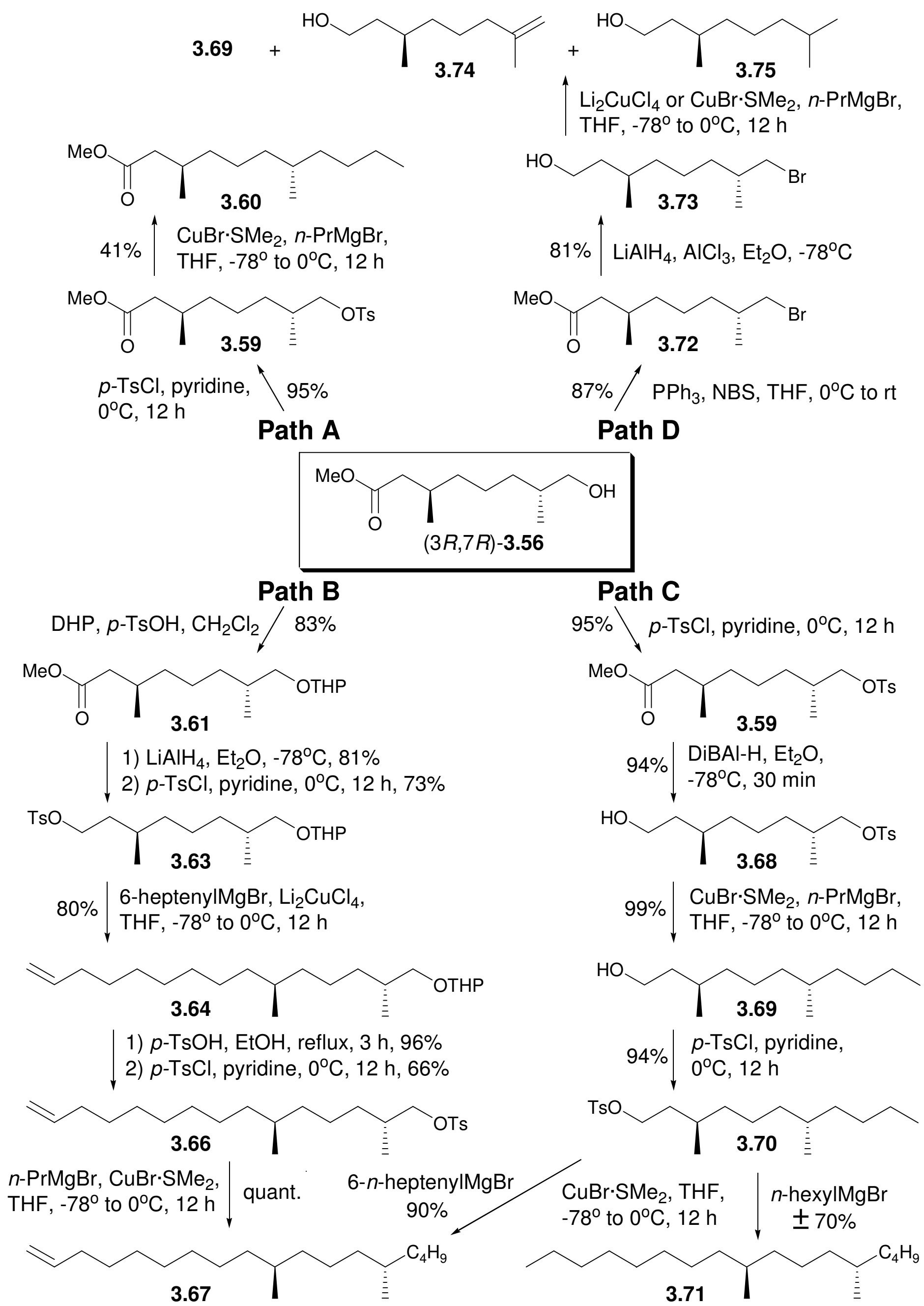
To demonstrate the synthetic value of the catalytic approach described in paragraph 3.3, it was employed in the total synthesis of two synergistic - female-produced - pheromones **3.67** and **3.71** (Scheme 3.15) of the apple leafminer (*Lyonetia prunifoliella*). This insect causes a pest in apple orchards in the eastern regions of North America. Field trapping experiments with mixtures of pheromones extracted from female glands have shown promising results in controlling their procreation.[30] The total synthesis of all diastereoisomers of **3.67** and **3.71** by Mori and co-workers using a chiral pool strategy (Scheme 3.1) showed that (10S,14S)-**3.67** and (5S,9S)-**3.71** are the isomers with the strongest biological activity.[5,31]

In our case, the total synthesis of (10S,14S)-**3.67** and (5S,9S)-**3.71** required elongation of both ends of *anti*-building block (3R,7R)-**3.56** with alkyl chains. To this end, the primary alcohol of **3.56** was treated with *p*-toluenesulfonyl chloride in pyridine to afford **3.59** in 95% yield (Scheme 3.15; Path A). Subsequent chain elongation by reaction with *n*-propylmagnesium bromide in the presence of catalytic CuBr·SMe_2 or Li_2CuCl_4 furnished

3.60 in a disappointing 41% yield. Crude GC-MS showed that the poor yield was caused by partial addition of the Grignard reagent to the methyl ester. This urged us to pursue a different strategy (Path B), which started with the protection of primary alcohol **3.56** as a tetrahydropyranyl (THP) ether (**3.61**, 83%). Reduction of **3.61** with $LiAlH_4$ (**3.62**, 81%) followed by formation of the corresponding tosylate yielded **3.63** (73%). Subjection of **3.63** to 6-heptenylmagnesium bromide (5.0 eq) and catalytic Li_2CuCl_4 (5 mol%) resulted in the isolation of **3.64** in a satisfying 80% yield. The THP-ether was then cleaved under acidic conditions (**3.65**, 96%), after which tosylate **3.66** (66%) was obtained as before. Finally, chain extension with *n*-PrMgBr (16.0 eq) and stoichiometric $CuBr{\cdot}SMe_2$ gave **3.67** as the sole product. In contrast, the use of (semi)-catalytic amounts of $CuBr{\cdot}SMe_2$ or Li_2CuCl_4 and a smaller excess of Grignard reagent (5.0 eq) was less successful, as it resulted in incomplete conversion after 12 hours and partial exchange of the tosyl group by a bromide.

In an attempt (Path C) to avoid the (extra) THP-protection and deprotection steps, methyl ester **3.59** was reduced to alcohol **3.68** (94%) with DiBAl-H. We were happy to find, that the tosyl moiety was not affected under these conditions. The subsequent chain elongation with *n*-PrMgBr once more gave the best result when stoichiometric $CuBr{\cdot}SMe_2$ and a large excess of Grignard reagent (16.0 eq) were used (**3.69**, 99%). Transformation of the hydroxyl moiety into a tosyl group was performed as before to give **3.70** (94%). Finally, Cu-catalyzed (25 mol%) coupling to 6-*n*-heptenylmagnesium bromide (5.0 eq) or *n*-hexylmagnesium bromide (5.0 eq) proceeded readily to furnish **3.67** (90%) and **3.71** (~ 70%), respectively. The yield of **3.71** was estimated by GC as the final product could not be separated from dodecane, which resulted from homocoupling of the Grignard reagent.

It should be mentioned that apart from tosylates, bromides were also tested in Cu-catalyzed coupling reactions with Grignard reagents (Path D). Treatment of alcohol **3.56** with *N*-bromosuccinimide and triphenylphosphine afforded primary bromide **3.72** in 87% yield, after which reduction with $LiAlH_4$ gave alcohol **3.73** (81%). However, reaction with *n*-PrMgBr in the presence of (semi)-catalytic $CuBr{\cdot}SMe_2$ or Li_2CuCl_4 produced a complex mixture containing only traces of the desired compound **3.69**. Investigation by crude ^{1}H-NMR and GC-MS led to the identification of (at least) two side products resulting from *β*-elimination (**3.74**) and *β*-hydride transfer (**3.75**).[32]



Scheme 3.15 *Synthesis of apple leafminer pheromones.*

3.5 Limitations of the catalytic system

As mentioned in paragraph 3.3.2, the stereochemical outcome of the second ACA with Me_2Zn to enone **3.48** was completely dominated by the chiral catalyst. From previous experience, we also knew that in the case of a six-membered ring, the first methyl substituent directs the second conjugate addition towards the *trans*-product (Figure 3.6).[33] We therefore decided to explore the limitations of the catalytic system by investigating the situation for the seven-membered ring.

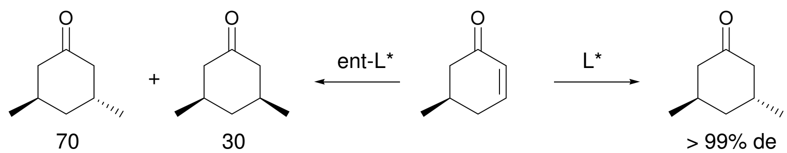
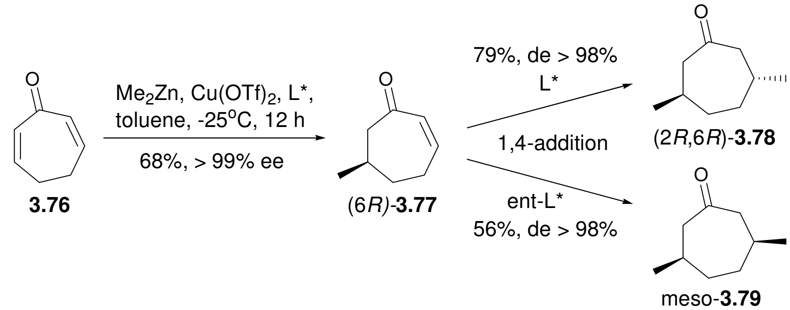


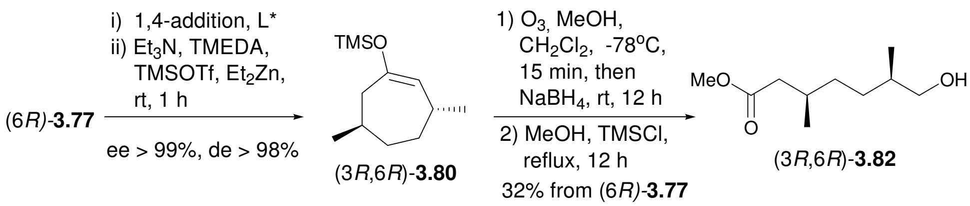
Figure 3.6 *Diastereoselectivity in the conjugate addition to the six-membered ring*.[33]

Interestingly, the one-step synthesis of starting material cyclohepta-2,6-dienone (**3.76**) from cycloheptanone by oxidation with IBX as performed for the eight-membered ring gave no conversion in our hands, even though this transformation had been described in the original paper.[25] We therefore resorted to the traditional method to prepare **3.76**, which consists of a four step sequence involving conversion of cycloheptanone into a cyclic acetal, α,α′-dibromination, elimination and finally deprotection of the ketone (22% yield over 4 steps).[34] Subsequent ACA of dimethylzinc to **3.76** under standard conditions (2.5 mol% $Cu(OTf)_2$, 5.0 mol% L*, 1.5 eq Me_2Zn, normal addition) gave **3.77** (ee > 99%) in 68% isolated yield (Scheme 3.16). Remarkably, coupling of the zinc-enolate to the starting material was not observed in this case. The introduction of the second methyl group was conducted under identical circumstances and proceeded with complete stereocontrol giving either (2*R*,6*R*)-**3.78** in the presence of L* or meso-**3.79** when ent-L* was used. The diastereomeric excess was determined by ^{1}H-NMR to be over 98%.



Scheme 3.16 *Iterative conjugate addition on the seven-membered ring.*

In analogy to the synthesis of saturated isoprenoid building blocks from eight-membered rings, 2,6-dimethyl-substituted seven-membered rings can be used to prepare linear chiral building blocks with a 1,4-dimethyl motif. To prove this principle, (6*R*)-**3.77** was subjected to a 1,4-addition, after which the zinc-enolate was trapped as before to give *trans*-silyl enol ether (2*R*,6*R*)-**3.80** (Scheme 3.17). Ozonolysis of (2*R*,6*R*)-**3.80** followed by a reductive work-up and esterification of the crude carboxylic acid (**3.81**) resulted in the isolation of building block (2*R*,6*R*)-**3.82** with an overall yield of 32% from **3.77**.



Scheme 3.17 *Synthesis of a chiral building block with the 1,4-dimethyl-motif.*

Even though the 1,4-dimethyl motif is not as omnipresent in nature as the saturated isoprenoid unit, there are some natural products which contain this structural element.[14,35]

3.6 Summary and concluding remarks

In this chapter the development of a general catalytic method to prepare all diastereoisomers of an enantiopure saturated isoprenoid building block (**3.56/3.57**) in good yield (26% over 5 steps) and with high selectivity (ee > 99% and de > 98%) was described. The synthetic value of this protocol has been demonstrated by its application in the concise total synthesis of two pheromones in 10 steps with an overall yield of 20% for **3.67** and

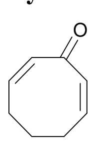
15% for **3.71**. In theory, **3.56** and **3.57** can also be utilized in cross-coupling reactions to access oligoisoprenoids of any desired stereochemistry and length (provided they have an even number of methyl substituents; see chapter 4). This approach should, therefore, show broad application in the total synthesis of a range of natural products and analogues thereof.

In addition, the limitations of the catalytic system have been investigated by comparing the iterative ACA on the seven-membered ring (complete control) with previously obtained results on the six-membered ring (predominantly *trans*-product).

3.7 Experimental section

For general information: see Chapter 2. Optical rotations were measured on a *Schmidt + Haensch* polarimeter (Polartronic MH8) with a 10 cm cell (*c* given in g/100 mL). Enantio- and diastereoselectivities were determined by capillary GC analysis (Chiraldex B-TA column (30 m x 0.25 mm) or Betadex 120 column (30 m x 0.25 mm)) using a flame ionization detector (in comparison with racemic products). Racemic **3.48** was prepared by 1,4-addition of Me_2CuLi to **3.47** in Et_2O at 0 ^oC. A racemic mixture of diastereoisomers **3.56/3.57** was synthesized by 1,4-addition of Me_2CuLi to racemic **3.48** in Et_2O at 0 ^oC followed by trapping of the enolate with TMSCl in the presence of Et_3N. Racemic **3.77** was synthesized by 1,4-addition of Me_2Zn to **3.76** in the presence of $Cu(OTf)_2$ and racemic MonoPhos. A racemic mixture of diastereomers **3.78/3.79** was prepared by repeating this protocol on racemic **3.77**.

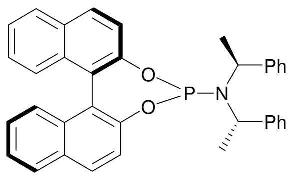
Cycloocta-2,7-dienone (3.47):[25] Preparation of IBX: Oxone (272 g, 0.44 mol, 1.46 eq) was suspended in water (650 mL) and *o*-iodobenzoic acid (75 g, 0.30 mol) was added. The resulting mixture was stirred at 75 ^oC for 3 h and then allowed to cool to room temperature over 1 h. After cooling for an additional 30 min at 0 ^oC, the suspension was filtered and the residue was washed with water (6 x 150 mL) and acetone (2 x 150 mL). The product was dried overnight in the air to give IBX (74.3 g, 0.26 mol, 88%) as a white solid.



Cyclooctanone (4.5 g, 36 mmol) was dissolved in DMSO (548 mL) and IBX (40 g, 143 mmol, 4.0 eq) was added. The resulting solution was slowly heated to 85 ^oC and stirred for 6 h and then overnight at 65 ^oC. After cooling to room temperature, the mixture was diluted with Et_2O until two layers appeared. The DMSO-layer was extracted with Et_2O (9x). The combined ether extracts were washed with aq. $NaHCO_3$ (5% w/w, 3x), water and brine (sat.), dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*- *n*-pentane-Et_2O 4:1) gave **3.47** (3.0 g, 25 mmol, 69%) as a light yellow liquid. ^1H-NMR

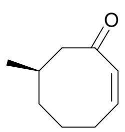
($CDCl_3$, 300 MHz) δ = 1.77 (m, 2H, C5-H), 2.38 (m, 4H, C4-H), 6.35 (m, 4H, C2-H, C3-H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 25.0 (t), 27.1 (t), 135.9 (d), 141.5 (d) ppm.

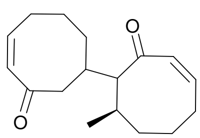
O,O′-(*R*)-(1,1′-dinaphthyl-2,2′-diyl)-N-N′-di-(S,S)-1-phenylethylphosphoramidite[26]



(R,S,S)-L*: (*R*)-(+)-Binol (6.0 g, 21.0 mmol) was dissolved in PCl_3 (24.0 mL, 275 mmol) and the resulting solution was heated under reflux overnight (80 °C) while protected from moisture with a $CaCl_2$-tube. Subsequently, the PCl_3 was removed *via* distillation using a hotgun; traces were removed by co-evaporation with toluene (24.0 mL). Upon drying in vacuo for 30 min, a solid formed, which was dissolved in dry toluene (24.0 mL) and cooled to 0 °C under argon (solution A). In a separate reaction vessel, S-(-)-bis(α-methylbenzyl)amine (5.74 mL, 25.1 mmol) was dissolved in dry THF (27.2 mL) and cooled to 0 °C. To this solution, *n*-BuLi (1.6 M in *n*-hexanes, 17.7 mL, 28.4 mmol, 1.2 eq) was added and the resulting mixture was stirred under argon at 0 °C for 30 min (solution B). Solution B (46 mL, 23 mmol, 1.1 eq) was then added to solution A and the thus obtained mixture was stirred for 2 h under argon at 0 °C (brown). Subsequently, excess amine-salt was precipitated by addition of Et_2O, after which these salts were removed by filtration over a glass frit. The filtrate was concentrated in vacuo to give a yellow foam. The product was purified by crystallization from acetone followed by column chromatography (*n*-pentane-Et_2O 50:1) to give (R,S,S)-L* (7.3 g, 13.6 mmol, 65%) as white crystals. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 1.85 (d, J = 8.5 Hz, 6H), 4.51 (m, 2H), 7.11 (m, 10H), 7.20 (m, 3H), 7.42 (m, 4H), 7.61 (d, J = 4.8 Hz, 1H), 7.95 (m, 4H) ppm. ^{31}P-NMR (80.8 MHz) δ = 144.7 ppm.

(*R*)-7-Methyl-cyclooct-2-enone (3.48): $Cu(OTf)_2$ (240 mg, 0.664 mmol, 5 mol%) and R,S,S-L* (715 mg, 1.33 mmol, 10 mol%) were dissolved in dry toluene (40 mL) and stirred at room temperature for 30 min under argon. The solution was cooled to -25 °C and Me_2Zn (2.0 M in toluene, 20.0 mL, 40.0 mmol, 3.0 eq) was added over 2 min. After stirring for 5 min, **3.47** (1.62 g, 13.3 mmol) dissolved in toluene (40 mL) was added *via* a syringe pump over 5 h. The resulting solution was stirred at -25 °C overnight and then quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x), washed with brine (sat.) and dried (Na_2SO_4). The Et_2O was removed under vacuum and the product purified by column chromatography (*n*-pentane-Et_2O 95:5 to 9:1) to give (*7R*)-**3.48** as a colorless liquid (1.56 g, 11.3 mmol, 85.0%, ee >99%) and **3.51** (< 5%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.01 (d, J = 6.8 Hz, 3H), 1.36 (m, 1H), 1.56-1.74 (m, 3H), 2.13 (m, 1H), 2.46 (dd, J = 8.4, 13.6 Hz, 1H), 2.42-2.62 (m, 2H), 2.70 (dd, J = 5.6, 13.6 Hz, 1H), 6.04 (d, J = 12.0 Hz, 1H), 6.35 (dt, J = 7.2, 12.4 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 21.5 (t), 21.8 (q), 28.0 (t), 28.8 (d), 32.0 (t), 50.0 (t), 133.3 (d), 141.5 (d), 204.4 (s) ppm. Ee determination: GC Chiraldex B-TA, 30 m x 0.25 mm, He-flow = 1.0 mL/min, T_i = 70 °C for 5 min, T_f = 150 °C, rate 10 °C/min, rt 23.5 (S), 23.8 (R) min. MS(EI) for $C_9H_{14}O$: m/z = 138 [M^+], HRMS calcd for $C_9H_{14}O$: 138.104, found: 138.105.

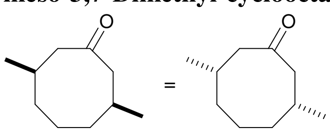




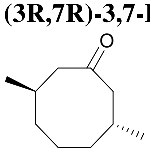
^{1}H-NMR **3.51** ($CDCl_3$, 400 MHz) δ = 1.06 (d, J = 6.8 Hz, 3H), 1.42-1.82 (m, 8H), 2.11-2.27 (m, 3H), 2.40-2.66 (m, 6H), 5.80 (d, J = 12.4 Hz, 1H), 5.86 (d, J = 12.4 Hz, 1H), 6.22 (m, 2H) ppm. ^{13}C-NMR **3.51**

($CDCl_3$, 50.3 MHz) δ = 18.4 (q), 20.3 (t), 22.6 (t), 28.4 (t), 29.8 (d), 30.0 (t), 30.1 (t), 33.4 (d), 48.7 (t), 61.3 (d), 128.6 (d), 138.7 (d), 140.4 (d), 207.4 (s) ppm. MS(CI) for $C_{17}H_{24}O_2$: m/z = 278 $(M + NH_4)^+$.

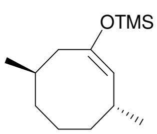
meso-3,7-Dimethyl-cyclooctanone (2.49): The 1,4-addition on **3.48** was performed as described for the synthesis of **3.53**. The reaction was quenched with aq. NH_4Cl (sat.) and work-up was done as for **3.48**. The product was isolated as a colorless oil after purification by column chromatography (*n*-pentane-Et_2O 9:1). NMR data were as reported in the literature[4c]: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.98 (d, *J* = 6.4 Hz, 6H), 1.28 (m, 3H), 1.49 (m, 1H), 1.68 (m, 2H), 2.18 (m, 4H), 2.52 (dd, *J* = 8.4, 18.0 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 22.3 (q), 22.5 (t), 32.4 (d), 36.3 (t), 50.2 (t), 215.2 (s) ppm.



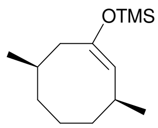
(3R,7R)-3,7-Dimethyl-cyclooctanone (2.50): The 1,4-addition on **3.48** was performed as described for the synthesis of **3.52**. The reaction was quenched with aq. NH_4Cl (sat.) and the work-up was done as for **3.48**. The product was isolated as a colorless oil after purification by column chromatography (*n*-pentane-Et_2O 9:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.99 (d, *J* = 6.8 Hz, 6H), 1.15 (m, 2H), 1.38 (m, 2H), 1.63 (m, 2H), 2.18 (m, 2H), 2.22 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 23.3 (q), 23.7 (t), 32.1 (d), 36.1 (t), 50.3 (t), 215.9 (s) ppm.



[((3R,7R)-3,7-Dimethyl-cyclooct-1-yl)oxy]-trimethylsilane (3.52): $Cu(OTf)_2$ (6.6 mg, 18 μmol, 2.5 mol%) and R,S,S-L* (20 mg, 37 μmol, 5 mol%) were dissolved in dry DCM (2.5 mL) and the mixture was stirred at room temperature for 30 min under argon. Subsequently, the solution was cooled to -25 °C and Me_2Zn (2.0 M in toluene, 0.54 mL, 1.08 mmol, 1.5 eq) was added over 2 min. After stirring for 5 min, (*7R*)-**3.48** (100 mg, 0.72 mmol) dissolved in DCM (2.0 mL) was added *via* a syringe pump over 5 h. The resulting solution was stirred at -25 °C overnight after which Et_3N (0.30 mL, 2.17 mmol, 3.0 eq), TMEDA (0.55 mL, 3.6 mmol, 5.0 eq) and a pre-stirred (30 min) solution of TMSOTf (0.65 mL, 3.62 mmol, 5.0 eq) and Et_2Zn (1.1 M in toluene, 0.27 mL, 0.30 mmol, 0.42 eq) were added. The mixture was then stirred for 2 h while slowly warming to ambient temperature. Et_2O (2.5 mL) was added to the solution and the reaction mixture was flashed over a silica-plug (SiO_2 inactivated with Et_3N prior to use). The Et_2O was evaporated under vacuum and the product purified by column chromatography (*n*-pentane; SiO_2 inactivated with Et_3N prior to use) to give (3*R*,7*R*)-**3.52** (ee > 99%, de > 98%) as a volatile colorless liquid with still some toluene present. **3.52** was immediately used in the next step without further removal of the toluene. For the purpose of analysis the toluene was removed in vacuo from a small sample of the product. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.18 (s, 9H), 0.89 (d, *J* = 6.8 Hz, 3H), 0.97 (d, *J* = 6.4 Hz, 3H), 0.92-1.16 (m, 3H), 1.36-1.78 (m, 5H), 2.12-2.32 (m, 2H), 4.35 (d, *J* = 8.8 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 0.4 (q), 22.5 (q), 23.2 (q), 23.6 (t), 30.8 (d), 33.5 (d), 34.9 (t), 39.7 (t), 40.5 (t), 111.9 (d), 151.0 (s) ppm. MS(EI) for $C_{13}H_{26}OSi$: m/z = 226 [M^+]. Ee determination: Betadex 120, 30 m x 0.25 mm, He-flow = 1.0 mL/min, T = 100 °C for 30 min, rt. 22.9 (3S,7S), 23.7 (3R,7R) min.

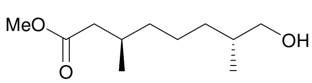


[((3S,7R)-3,7-Dimethyl-cyclooct-1-yl)oxy]-trimethylsilane (3.53): $Cu(OTf)_2$ (16 mg, 45

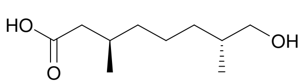


μmol, 2.5 mol%) and S,R,R-L* (49 mg, 90 μmol, 5 mol%) were dissolved in dry DCM (6.3 mL) and the mixture was stirred at room temperature for 30 min under argon. Subsequently the solution was cooled to -25 ^oC and Me_2Zn (2.0 M in toluene, 1.36 mL, 2.71 mmol, 1.5 eq) was added over 2 min. After stirring for 5 min, (*7R*)-**3.48** (250 mg, 1.81 mmol) dissolved in DCM (5 mL) was added *via* a syringe pump over 5 h. The resulting solution was stirred at -25 ^oC overnight, after which Et_3N (0.76 mL, 5.5 mmol, 3.0 eq, freshly distilled from CaH_2), HMPA (1.6 mL, 9.0 mmol, 5.0 eq, freshly distilled from CaH_2 under reduced pressure) and a pre-stirred (30 min) solution of TMSCl (1.15 mL, 9.0 mmol, 5.0 eq, freshly distilled from CaH_2) and Et_2Zn (1.1 M in toluene, 0.69 mL, 0.76 mmol, 0.42 eq) were added. The resulting mixture was stirred for 2 h while slowly warming to ambient temperature. Isolation was performed as for compound **3.52** to give (3*S*,7*R*)-**3.53** (ee > 99%, de >98%) as a volatile colorless liquid with still some toluene present. **3.53** was immediately used in the next step without further removal of the toluene. Complete removal of toluene in vacuo was performed for an analytical sample. NMR data were as reported in the literature[4c]: ^1H-NMR ($CDCl_3$, 400 MHz) δ = 0.18 (s, 9H), 0.95 (d, *J* = 4.8 Hz, 3H), 0.97 (d, *J* = 4.8 Hz, 3H), 1.08-1.18 (m, 1H), 1.21-1.36 (m, 2H), 1.53-1.61 (m, 2H), 1.70-1.79 (m, 2H), 1.87-1.98 (m, 1H), 2.21- 2.32 (m, 1H), 2.60 (dd, *J* = 4.8, 14.0 Hz, 1H), 4.44 (d, *J* = 7.6 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 0.3 (q), 21.5 (q), 23.5 (q), 24.7 (t), 32.0 (d), 33.7 (d), 34.2 (t), 37.1 (t), 39.7 (t), 113.3 (d) 149.9 (s) ppm. MS(EI) for $C_{13}H_{26}OSi$: m/z = 226 [M^+]. Ee determination: Betadex 120, 30 m x 0.25 mm, He-flow = 1.0 mL/min, T = 100 ^oC for 30 min, rt. 23.9 (3R,7S), 25.7 (3S,7R) min.

Methyl 8-hydroxy-(3R,7R)-3,7-dimethyloctanoic acid (3.56): (3*R*,7*R*)-**3.52** (0.72 mmol)



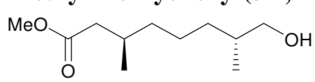
was dissolved in MeOH (3.6 mL) and DCM (3.6 mL) and O_3 was bubbled through the solution at -78 ^oC until the solution colored green/blue (10-15 min). The solution was then purged with argon, after which $NaBH_4$ (274 mg, 7.2 mmol, 10 eq) was added and the reaction mixture was allowed to slowly warm to ambient temperature. After stirring overnight, the reaction mixture was concentrated and the residue was taken up in Et_2O. Subsequently, aq. HCl (10%) was added and the aqueous layer was extracted with Et_2O (3x). The combined ether extracts were dried ($MgSO_4$) and concentrated. The crude product was used in the next reaction without purification. An analytical sample was purified by column chromatography (*n*-pentane-EtOAc 1:2) to give (3*R*,7*R*)-**3.54** as a colorless oil.

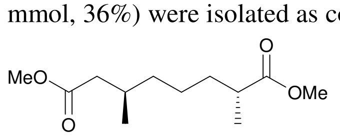


^1H-NMR ($CDCl_3$, 300 MHz) δ = 0.91 (d, *J* = 6.6 Hz, 3H), 0.96 (d, *J* = 6.6 Hz, 3H), 1.06-1.45 (m, 7H), 1.61 (m, 1H), 1.97 (m, 1H), 2.11 (dd, *J* = 7.6, 14.8 Hz, 1H), 2.33 (dd, *J* = 5.7, 14.8 Hz, 1H), 3.46 (m, 2H), 7.04 (brs, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.3 (q), 19.5 (q), 24.0 (t), 29.9 (d), 32.9 (t), 35.3 (d), 36.6 (t), 41.5 (t), 67.9 (t), 178.5 (s) ppm. MS(CI) for $C_{10}H_{20}O_3$: m/z = 206 $(M + NH_4)^+$, HRMS calcd for $C_{10}H_{20}O_3$-O: 172.146, found: 172.147.

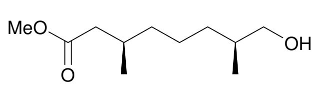
3.54 was dissolved in anhydrous MeOH (11 mL) and TMSCl (0.26 mL, 2.1 mmol, 3.0 eq) was added. The resulting solution was heated under reflux overnight while protected from moisture with a $CaCl_2$-tube, after which TLC (*n*-pentane-EtOAc 4:1) showed complete conversion. The mixture was cooled to room temperature and concentrated, after which aq. $NaHCO_3$ (sat.) was added to the residue and the product was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. The product (3*R*,7*R*)-**3.56** (65 mg, 0.32 mmol, 45% from (*7R*)-**3.48**) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 6:1 to 3:1). (3R,7R)-**3.56**: $[\alpha]_D^{20} = +15.6^o$ (c = 1.27, $CHCl_3$), (3S,7S)-**3.56** (prepared in an identical way from **3.47** by two consecutive conjugate additions using S,R,R-L*): $[\alpha]_D^{20} = -14.4^o$ (c = 0.71, $CHCl_3$). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.90 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.8 Hz, 3H), 1.05-1.42 (m, 7H), 1.60 (m, 1H), 1.94 (m, 1H), 2.11 (dd, *J* = 8.0, 14.4 Hz, 1H), 2.33 (dd, *J* = 6.4, 14.4 Hz, 1H), 3.41 (dd, *J* = 6.4, 10.8 Hz, 1H), 3.48 (dd, *J* = 6.0, 10.8 Hz, 1H), 3.65 (s, 3H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.4 (q), 19.6 (q), 24.1 (t), 30.1 (d), 33.0 (t), 35.5 (d), 36.7 (t), 41.6 (t), 51.3 (q), 68.2 (t), 173.7 (s) ppm. MS(CI) for $C_{11}H_{22}O_3$: m/z = 220 $(M + NH_4)^+$, HRMS calcd for $C_{11}H_{22}O_3$-O: 186.162, found: 186.162.

Methyl 8-hydroxy-(3R,7R)-3,7-dimethyloctanoic acid (3.56) using a one-pot conversion of 3.48 into 3.54: Silyl enol ether **3.52** was formed as described earlier starting from 50 mg (0.36 mmol) of **3.48**. To the reaction mixture was added MeOH (2.0 mL) and O_3 was bubbled through the solution at -78 °C until the solution colored green (5 min). The solution was then purged with argon, after which $NaBH_4$ (137 mg, 3.6 mmol, 10 eq) was added and the reaction mixture was allowed to slowly warm to ambient temperature. After stirring overnight, the reaction mixture was concentrated and the residue was taken up in Et_2O. Subsequently, aq. HCl (10%) was added and the aqueous layer was extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$) and concentrated. The crude carboxylic acid was dissolved in anhydrous MeOH (5.0 mL) and heated under reflux with a $CaCl_2$-tube in the presence of TMSCl (0.14 mL, 1.08 mmol, 3.0 eq). After work-up and purification (see **3.56**), the product (3R,7R)-**3.56** (29 mg, 0.14 mmol, 40%) and a side product which proved to be 2,6-dimethyl-octanedioic acid dimethyl ester **3.58** (30 mg, 0.13 mmol, 36%) were isolated as colorless oils. ^{1}H-NMR **3.58** ($CDCl_3$, 300 MHz) δ = 0.91 (d, *J* = 6.6 Hz, 3H), 1.13 (d, *J* = 6.9 Hz, 3H), 1.12-1.67 (m, 6H), 1.93 (m, 1H), 2.09 (dd, *J* = 7.8, 15.0 Hz, 1H), 2.28 (dd, *J* = 5.7, 15.0 Hz, 1H), 2.42 (m, 1H), 3.65 (s, 6H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 16.9 (q), 19.5 (q), 24.4 (t), 30.0 (d), 33.6 (t), 36.3 (t), 39.2 (d), 41.4 (t), 51.2 (q), 51.3 (q), 173.5 (s), 171.1 (s) ppm. MS(CI) for $C_{12}H_{22}O_4$: m/z = 248 $(M + NH_4)^+$.

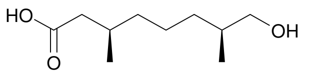




Methyl 8-hydroxy-(3R,7S)-3,7-dimethyloctanoic acid (3.57): (3*S*,7*R*)-**3.53** (1.81 mmol) was dissolved in MeOH (9 mL) and DCM (9 mL) and O_3 was bubbled through the solution at -78 °C until the solution colored green/blue (10-15 min). The solution was then purged with nitrogen, after which $NaBH_4$ (684 mg, 18.1 mmol, 10 eq) was added and the reaction mixture was allowed to slowly warm to ambient temperature. After stirring overnight, the reaction mixture was concentrated and the residue was taken up in Et_2O. Subsequently, aq. HCl (10%) was added and the water layer was extracted with Et_2O (3x). The combined organic layers were dried ($MgSO_4$) and



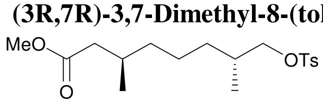
concentrated. The crude product **3.55** (182 mg, 0.97 mmol, 53%) was used in the next reaction without purification. An analytical sample was purified by column chromatography (*n*-pentane-EtOAc 1:2) to give (3*R*,7*S*)-**3.55** as a colorless oil. NMR data



were as reported in the literature[4c]: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.91 (d, *J* = 6.8 Hz, 3H), 0.96 (d, *J* = 6.8 Hz, 3H), 1.05-1.46 (m, 7H), 1.59 (m, 1H), 1.95 (m, 1H), 2.14 (dd, *J* = 7.6, 15.2 Hz, 1H), 2.33 (dd, *J* = 6.4, 15.2 Hz, 1H), 3.42 (dd, *J* = 6.0, 10.4 Hz, 1H), 3.50 (dd, *J* = 5.6, 10.4 Hz, 1H), 5.8-6.8 (s,br 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.4 (q), 19.7 (q), 24.0 (t), 30.0 (d), 33.0 (t), 35.4 (d), 36.7 (t), 41.4 (t), 68.0 (t), 178.7 (s) ppm.

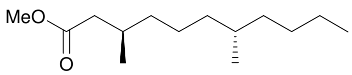
3.55 (182 mg, 0.97 mmol) was dissolved in anhydrous MeOH (15 mL) and TMSCl (0.37 mL, 2.9 mmol, 3.0 eq) was added. The resulting solution was heated under reflux overnight while protected from moisture with a $CaCl_2$-tube, after which TLC (*n*-pentane-EtOAc 4:1) showed complete conversion. The mixture was cooled to room temperature and concentrated, after which aq. $NaHCO_3$ (sat) was added to the residue and the product was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. The product (3*R*,7*S*)-**3.57** (163 mg, 0.81 mmol, 45% from (*7R*)-**3.48**) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 6:1 to 3:1). (3R,7S)- **3.57**: $[\alpha]_D^{20}$ = -3.3^o (c = 1.22, $CHCl_3$), Lit.[4c,29] $[\alpha]_D^{25}$ = -3.9^o, (3S,7R)- **3.57** (prepared in an identical way from **3.47** by two consecutive conjugate additions with S,R,R-L* and R,S,S-L*): $[\alpha]_D^{20}$ = +3.02 (c = 0.91, $CHCl_3$). NMR data were as reported in the literature[4c]: ^{1}H-NMR **3.57** ($CDCl_3$, 400 MHz) δ = 0.92 (t, *J* = 6.8 Hz, 6H), 1.04-1.44 (m, 7H), 1.60 (m, 1H), 1.95 (m, 1H), 2.11 (dd, *J* = 8.0, 14.4 Hz, 1H), 2.30 (dd, *J* = 6.0, 14.8 Hz, 1H), 3.41 (dd, *J* = 6.4, 10.4 Hz, 1H), 3.50 (dd, *J* = 6.0, 10.4 Hz, 1H), 3.66 (s, 3H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.4 (q), 19.6 (q), 24.1 (t), 30.2 (d), 33.0 (t), 35.6 (d), 36.8 (t), 41.5 (t), 51.2 (q), 68.1 (t), 173.7 (s) ppm. MS(CI) for $C_{11}H_{22}O_3$: m/z = 220 $(M + NH_4)^+$, HRMS calcd for $C_{11}H_{22}O_3$-O: 186.162, found: 186.163.

(3R,7R)-3,7-Dimethyl-8-(toluene-4-sulfonyloxy)-octanoic acid methyl ester (3.59):

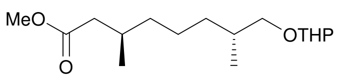


(3R,7R)-**3.56** (116 mg, 0.57 mmol) was dissolved in dry pyridine (1.7 mL) and *p*-TsCl (164 mg, 0.86 mmol, 1.5 eq) was added at 0 ^{o}C. The resulting solution was stirred at 4 ^{o}C under argon overnight and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat.), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated to give **3.59** (194 mg, 0.54 mmol, 95%) as a light yellow oil which was used in the next reaction without purification. An analytical sample was purified by column chromatography (*n*-pentane-EtOAc 4:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.87 (d, *J* = 6.4 Hz, 3H), 0.89 (d, *J* = 6.8 Hz, 3H), 1.00-1.36 (m, 6H), 1.77 (m, 1H), 1.89 (m, 1H), 2.09 (dd, *J* = 8.0, 14.4 Hz, 1H), 2.25 (dd, *J* = 6.4, 14.4 Hz, 1H), 2.44 (s, 3H), 3.65 (s, 3H), 3.80 (dd, *J* = 6.4, 9.2 Hz, 1H), 3.86 (dd, *J* = 6.0, 9.2 Hz, 1H), 7.34 (d, *J* = 8.0 Hz, 2H), 7.78 (d, *J* = 8.0 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 16.2 (q), 19.5 (q), 21.5 (q), 23.8 (t), 30.1 (d), 32.6 (t), 32.7 (d), 36.5 (t), 41.5 (t), 51.2 (q), 74.9 (t), 127.8 (d), 129.7 (d), 173.5 (s) ppm. MS(EI) for $C_{18}H_{28}O_5S$: m/z = 356 [M^+], HRMS calcd for $C_{18}H_{28}O_5S$: 356.166, found: 356.164.

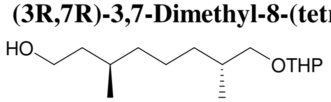
(3R,7S)-3,7-Dimethyl-undecanoic acid methyl ester (3.60): The solution of *n*-PrMgBr (1.5 M in THF) was prepared as described for the synthesis of **3.69**. The Grignard reagent (0.37 mL, 0.56 mmol, 4.0 eq) was added in a dropwise fashion to a solution of **3.59** (50 mg, 0.14 mmol) and CuBr·SMe_2S (7.2 mg, 35 μmol, 25 mol%) in dry THF (1.3 mL) at -78 °C under argon. After stirring for 1 h at -78 °C, the solution was allowed to slowly warm to 0 °C and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Crude GC-MS showed a mixture of products from which **3.60** (13 mg, 0.06 mmol, 41%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane- Et_2O 95:5). ^{1}H-NMR ($CDCl_3$, 200 MHz) δ = 0.82-0.94 (t + 2d, *J* = 6.2 Hz and *J* = 6.6 Hz, 9H), 1.00-1.40 (m, 16H), 1.96 (m, 1H), 2.11 (dd, *J* = 8.0, 14.4 Hz, 1H), 2.31 (dd, *J* = 5.8, 14.4 Hz, 1H), 3.66 (s, 3H) ppm.



(3R,7R)-3,7-Dimethyl-8-(tetrahydro-pyran-2-yloxy)-octanoic acid methyl ester (3.61): To a solution of **3.60** (210 mg, 1.04 mmol) in dichloromethane (3.2 mL) were added 3,4-dihydro-2H-pyran (DHP, 128 μl, 1.40 mmol, 1.35 eq) and *p*-toluenesulfonic acid·H_2O (2.0 mg, 11 μmol, 1.1 mol%). The resulting solution was stirred for 2 h at room temperature, after which aq. $NaHCO_3$ (sat.) was added. The aqueous layer was extracted with dichloromethane (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$), filtered and concentrated. **3.61** (248 mg, 0.87 mmol, 83%) was isolated as a colorless liquid after purification by column chromatography (*n*-pentane-Et_2O 9:1 to 4:1). ^{1}H-NMR ($CDCl_3$, 400 MHz, mixture of diastereomers) δ = 0.88 (d, *J* = 6.8 Hz, 3H), 0.90 (d, *J* = 6.4 Hz, 3H), 1.04-1.60 (m, 10H), 1.65-1.98 (m, 4H), 2.09 (dd, *J* = 8.4, 14.8 Hz, 1H), 2.28 (dd, *J* = 6.0, 14.8 Hz, 1H), 3.11, 3.20 (each dd, *J* = 6.4, 9.6 Hz and *J* = 6.0, 9.6 Hz, 1H), 3.48 (m, 1H), 3.48, 3.57 (each dd, *J* = 6.4, 9.6 Hz, 1H), 3.64 (s, 3H), 3.83 (m, 1H), 4.54 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 17.0 (q), 19.4 (t), 19.5 (q), 24.1 (t), 25.4 (t), 30.2, (d), 30.6 (t), 33.19 (d), 33.25 (d), 33.6 (t), 36.8 (t), 41.5 (t), 51.2 (q), 61.96 (t), 62.03 (t), 72.8 (t), 73.0 (t), 98.7 (d), 98.9 (d), 173.6 (s) ppm. MS(EI) for $C_{16}H_{30}O_4$-H: m/z = 285 $[M-H]^+$, HRMS calcd for $C_{16}H_{30}O_4$-H: 285.207, found: 285.206.

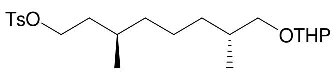


(3R,7R)-3,7-Dimethyl-8-(tetrahydro-pyran-2-yloxy)-octan-1-ol (3.62): A solution of **3.61** (243 mg, 0.85 mmol) in Et_2O (1.4 ml) was added to a suspension of $LiAlH_4$ (35 mg, 0.93 mmol, 1.05 eq) in Et_2O (0.93 mL) at -78 °C. The resulting mixture was stirred for 45 min and then quenched with water followed by aq. NaOH (15% w/w) at 0 °C. After stirring for 30 min, the suspension was filtered through celite and the filtrate was extracted with Et_2O (3x). The combined organic layers were dried (K_2CO_3), filtered and concentrated to give **3.62** (179 mg, 0.69 mmol, 81%) as a mixture of diastereomers. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.90 (m, 6H),1.02-1.88 (m, 16 H), 3.14, 3.23 (each dd, *J* = 6.4, 9.6 Hz and *J* = 6.0, 9.6 Hz, 1H), 3.47-3.53 (m, 1H), 3.50, 3.60 (each dd, *J* = 6.0, 9.6 Hz, 1H), 3.63-3.73 (m, 2H), 3.86 (m, 1H), 4.56 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 17.0 (q), 17.1 (q), 19.4 (t), 19.5 (q), 24.1 (t), 25.4 (t), 29.3 (d), 30.6 (t), 33.22 (d), 33.28 (d), 33.7 (t), 37.2



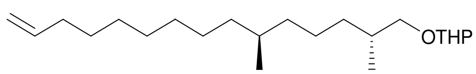
(t), 39.9 (t), 61.1 (t), 62.0 (t), 62.1 (t), 72.9 (t), 73.1 (t), 98.7 (d), 99.0 (d) ppm. MS(EI) for $C_{15}H_{30}O_3$-H: m/z = 257 $[M-H]^+$, HRMS calcd for $C_{15}H_{30}O_3$-H: 257.212, found: 257.211.

[(3R,7R)-3,7-Dimethyl-8-(tetrahydro-pyran-2-yloxy) octyl] *p*-toluenesulfonate (3.63):



3.62 (173 mg, 0.67 mmol) was dissolved in dry pyridine (1.7 mL) and *p*-TsCl (166 mg, 0.87 mmol, 1.3 eq) was added at 0 °C. The resulting solution was stirred at 4 °C under argon overnight and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat.), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated. **3.63** (203 mg, 0.49 mmol, 73%) was obtained as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 92:8 to 9:1). ^{1}H-NMR ($CDCl_3$, 400 MHz, mixture of diastereomers) δ = 0.79 (d, *J* = 6.0 Hz, 3H), 0.89 (t, *J* = 6.4 Hz, 3H), 1.00-1.86 (m, 16 H), 2.44 (s, 3H), 3.12, 3.20 (each dd, *J* = 6.4, 9.2 Hz and *J* = 6.0, 9.6 Hz, 1H), 3.46-3.52 (m, 1H), 3.49, 3.57 (each dd, *J* = 6.4, 9.6 Hz, 1H), 3.85 (m, 1H), 4.03-4.08 (m, 2H), 4.55 (m, 1H), 7.34 (d, *J* = 8.4 Hz, 2H), 7.78 (d, *J* = 8.4 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 17.0 (q), 18.9 (q), 19.46 (t), 19.51 (t), 21.5 (q), 24.0 (t), 25.4 (t), 29.01 (d), 29.03 (d), 30.6 (t), 33.19 (d), 33.26 (d), 33.7 (t), 35.6 (t), 36.70 (t), 36.73 (t), 62.0 (t), 62.1 (t), 68.9 (t), 72.9 (t), 73.0 (t), 98.7 (d), 99.0 (d), 127.7 (d), 129.7 (d), 133.1 (s), 144.5 (s) ppm. MS(CI) for $C_{22}H_{36}O_5S$: m/z = 430 $(M + NH_4)^+$.

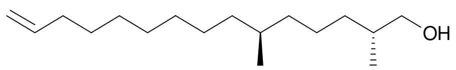
2-((2R,6S)-2,6-Dimethyl-pentadec-14-enyloxy)-tetrahydro-pyran (3.64):



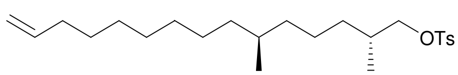
Preparation Grignard: 6-heptenyl bromide (185 μL, 1.21 mmol) in dry THF (2.1 mL) was added dropwise to Mg (44 mg, 1.8 mmol, 1.5 eq) in an argon-purged flask and then stirred for 1 h at 45 °C. The resulting solution was used immediately. Preparation Li_2CuCl_4: $CuCl_2$ (161 mg, 1.20 mmol) and LiCl (102 mg, 2.40 mmol, 2.0 eq) were dissolved in dry THF (12.0 mL) and stirred for 1 h under argon.

To a solution of **3.63** (100 mg, 0.24 mmol) in dry THF (1.3 mL) were added in a dropwise fashion the Grignard reagent (2.2 mL, 1.2 mmol, 5.0 eq) and Li_2CuCl_4 (0.1 M in THF, 121 μl, 12 μmol, 5 mol%) at -78 °C under argon. After stirring for 1 h at -78 °C, the solution was allowed to slowly warm to 0 °C and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. **3.64** (66 mg, 0.19 mmol, 80%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-Et_2O 98:2). ^{1}H-NMR ($CDCl_3$, 400 MHz, mixture of diastereomers) δ = 0.82 (d, *J* = 6.4 Hz, 3H), 0.92 (t, *J* = 7.2 Hz, 3H), 1.04-1.87 (m, 26 H), 2.04 (m, 2H), 3.13, 3.24 (each dd, *J* = 6.8, 9.2 Hz and *J* = 6.0, 9.2 Hz, 1H), 3.47-3.53 (m, 1H), 3.50, 3.60 (each dd, *J* = 6.4, 9.6 Hz, 1H), 3.86 (m, 1H), 4.57 (m, 1H), 4.91-5.02 (m, 2H), 5.82 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 17.0 (q), 17.1 (q), 19.4 (t), 19.50 (t), 19.54 (q), 24.2 (t), 25.4 (t), 27.0 (t), 28.8 (t), 29.0 (t), 29.4 (t), 29.8 (t), 30.6 (t), 32.6 (d), 33.27 (d), 33.33 (d), 33.7 (t), 33.87 (t), 33.90 (t), 37.0 (t), 37.2 (t), 62.0 (t), 62.1 (t), 73.0 (t), 73.2 (t), 98.6 (d), 98.9 (d), 114.0 (t), 139.1 (d) ppm. MS(EI) for $C_{22}H_{42}O_2$: m/z = 338 $[M]^+$, HRMS calcd for $C_{22}H_{42}O_2$: 338.318, found: 338.319.

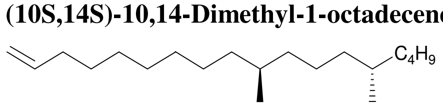
(2R,6S)-2,6-Dimethyl-pentadec-14-en-1-ol (3.65): 3.64 (60 mg, 0.18 mmol) was dissolved in EtOH-H_2O (19:1 v/v, 2.0 mL) and *p*-TsOH·H_2O (3.0 mg, 16 µmol, 8.9 mol%) was added. The resulting solution was heated under reflux for 3 h and then cooled to room temperature. The reaction was quenched with K_2CO_3 (s), after which brine (sat.) was added. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$), filtered and concentrated. **3.65** (41 mg, 0.16 mmol, 91%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane-Et_2O 95:5 to 9:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 6.8 Hz, 3H), 0.91 (d, *J* = 6.8 Hz, 3H), 1.02-1.40 (m, 20 H), 1.60 (m, 1H), 2.03 (m, 2H), 3.41 (dd, *J* = 6.8, 10.4 Hz, 1H), 3.50 (dd, *J* = 5.6, 10.4 Hz, 1H), 4.90-5.01 (m, 2H), 5.81 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 75.5 MHz) δ = 16.4 (q), 19.5 (q), 24.3 (t), 27.0 (t), 28.8 (t), 29.0 (t), 29.4 (t), 29.8 (t), 32.6 (d), 33.3 (t), 33.7 (t), 35.6 (d) 37.0 (t), 37.2 (t), 68.3 (t), 114.0 (t), 139.1 (d) ppm. MS(EI) for $C_{17}H_{34}O$: m/z = 254 $[M]^+$, HRMS calcd for $C_{17}H_{34}O$: 254.261, found: 254.261.

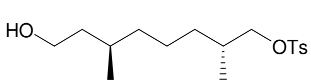


(2R,6S)-2,6-Dimethylpentadec-14-en-1-yl *p*-toluenesulfonate (3.66): 3.65 (38 mg, 0.15 mmol) was dissolved in dry pyridine (0.6 mL) and *p*-TsCl (43 mg, 0.22 mmol, 1.5 eq) was added at 0 °C. The resulting solution was stirred at 4 °C under argon overnight and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat.), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated. **3.66** (40 mg, 0.10 mmol, 66%) and starting material **3.65** (11 mg, 43 µmol, 29%) were obtained as colorless oils after purification by column chromatography (*n*-pentane-Et_2O 9:1 to 4:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.80 (d, *J* = 6.8 Hz, 3H), 0.87 (d, *J* = 6.8 Hz, 3H), 1.00-1.41 (m, 19 H), 1.76 (m, 1H), 2.04 (m, 2H), 2.45 (s, 3H), 3.80 (dd, *J* = 6.4, 9.6 Hz, 1H), 3.87 (dd, *J* = 5.6, 9.2 Hz, 1H), 4.90-5.02 (m, 2H), 5.81 (m, 1H), 7.34 (d, *J* = 8.0 Hz, 2H), 7.78 (d, *J* = 8.4 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 75.5 MHz) δ = 16.3 (q), 19.4 (q), 21.5 (q), 23.9 (t), 26.9 (t), 28.8 (t), 29.0 (t), 29.4 (t), 29.8 (t), 32.5 (d), 32.7 (d), 32.8 (t), 33.7 (t), 36.90 (t) 36.94 (t), 75.1 (t), 114.0 (t), 127.8 (d), 129.7 (d), 133.0 (s), 139.1 (d), 144.5 (s) ppm. MS(EI) for $C_{24}H_{40}O_3S$: m/z = 408 $[M]^+$, HRMS calcd for $C_{24}H_{40}O_3S$: 408.270, found: 408.271.



(10S,14S)-10,14-Dimethyl-1-octadecene (3.67 from 3.66): Procedure as described for the synthesis of **3.69**. A solution of **3.66** (10 mg, 24 µmol) in dry THF (0.31 mL) was reacted with *n*-PrMgBr (0.57 M in THF, 0.69 mL, 0.39 mmol, 16 eq) in the presence of CuBr·SMe_2 (5.0 mg, 24 µmol, 1.0 eq) to give **3.67** (6.9 mg, 24 µmol, quantitative). Analytical data identical to the synthesis of **3.67** from **3.70** (described below).

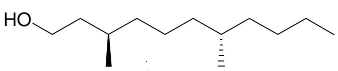




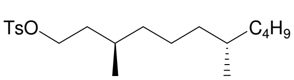
2R,6R)-2,6-Dimethyl-8-hydroxy-octyl *p*-toluenesulfonate (3.68): 3.59 (193 mg, 0.54 mmol) was dissolved in dry Et_2O and DiBAl-H (20% wt in toluene, 2.24 mL, 2.71 mmol, 5.0

eq) was added at -78 ^oC. The resulting solution was stirred for 30 min under argon and then poured into an aqueous solution of potassium sodium tartrate and stirred until the phases became clear. The aqueous layer was extracted with Et_2O (3x), the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. The product **3.68** (168 mg, 0.51 mmol, 94%) was isolated after column chromatography (*n*-pentane-EtOAc 2:1) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.84 (t, *J* = 6.8 Hz, 6H), 0.98-1.36 (m, 7H), 1.51 (m, 2H), 1.74 (m, 1H), 1.86 (brs, 1H), 2.42 (s, 3H), 3.63 (m, 2H), 3.78 (dd, *J* = 6.4, 9.2 Hz, 1H), 3.84 (dd, *J* = 6.0, 9.2 Hz, 1H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.74 (d, *J* = 8.0 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.2 (q), 19.3 (q), 21.4 (q), 23.7 (t), 29.1 (d), 32.6 (d), 32.6 (t), 36.8 (t), 39.6 (t), 60.8 (t), 75.0 (t), 127.7 (d), 129.6 (d), 132.8 (s), 144.5 (s) ppm. MS(EI) for $C_{17}H_{28}O_4S$: m/z = 328 [M^+], HRMS calcd for $C_{17}H_{28}O_4S$: 328.171, found: 328.172.

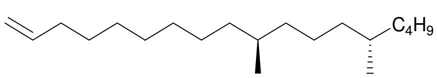
(3R,7S)-3,7-Dimethyl-undecan-1-ol (3.69): *n*-Propyl bromide (0.74 mL, 8.13 mmol) in dry THF (13.6 mL) was added dropwise to Mg (257 mg, 10.6 mmol, 1.3 eq) in an argon-purged flask and then stirred for 1 h at room temperature. The resulting solution was used immediately. The Grignard reagent (13.1 mL, 7.40 mmol, 16 eq) was added in a dropwise fashion to a solution of **3.68** (152 mg, 0.46 mmol) and CuBr·SMe_2S (95 mg, 0.46 mmol) in dry THF (5.7 mL) at -78 ^oC under argon. After stirring for 1 h at -78 ^oC, the solution was allowed to slowly warm to 0 ^oC and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. **3.69** (92 mg, 0.46 mmol, 99%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane- Et_2O 4:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 6.0 Hz, 3H), 0.87 (d, *J* = 6.8 Hz, 3H), 0.87 (t, *J* = 6.4 Hz, 3H), 1.00-1.40 (m, 14H), 1.48-1.70 (m, 3H), 3.66 (m, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 19.5 (q), 19.5 (q), 22.9 (t), 24.2 (t), 29.2 (t), 29.4 (d), 32.6 (d), 36.7 (t), 37.2 (t), 37.3 (t), 39.9 (t), 61.0 (t) ppm. MS(CI) for $C_{13}H_{28}O$: m/z = 218 $(M + NH_4)^+$, HRMS calcd for $C_{13}H_{28}O$-H_2O: 182.203, found: 182.205.



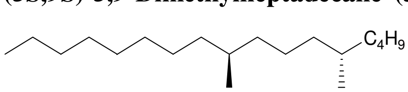
(3R,7S)-3,7-Dimethyl-undecyl *p*-toluenesulfonate (3.70): Compound **3.69** (89 mg, 0.44 mmol) was dissolved in dry pyridine (1.7 mL) and *p*-TsCl (135 mg, 0.71 mmol, 1.6 eq) was added at 0 ^oC. The resulting solution was stirred at 0 ^oC under argon overnight and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat.), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated to give **3.70** (148 mg, 0.42 mmol, 94%) as a light yellow oil which was used in the next reaction without purification. An analytical sample was purified by column chromatography (*n*-pentane-EtOAc 4:1). ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.80 (d, *J* = 6.8 Hz, 3H), 0.82 (d, *J* = 6.8 Hz, 3H), 0.88 (t, *J* = 6.8 Hz, 3H), 0.98-1.70 (m, 16H), 2.45 (s, 3H), 4.06 (m, 2H), 7.34 (d, *J* = 8.4 Hz, 2H), 7.79 (d, *J* = 8.0 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.1 (q), 19.0 (q), 19.5 (q), 21.5 (q), 22.9 (t), 24.0 (t), 29.1 (d), 29.2 (t), 32.6 (d), 35.6 (t), 36.7 (t), 36.8 (t), 37.1 (t), 69.0 (t), 127.8 (d), 129.7 (d), 133.1 (s), 144.5 (s) ppm. MS(EI) for $C_{20}H_{34}O_3S$: m/z = 354 [M^+], HRMS calcd for $C_{20}H_{34}O_3S$-C_4H_9: 297.152, found: 297.151.



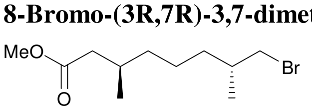
(10S,14S)-10,14-Dimethyl-1-octadecene (3.67 from 3.70): 6-heptenyl bromide (0.38 mL, 2.49 mmol) in dry THF (6.0 mL) was added dropwise to Mg (91 mg, 3.7 mmol, 1.5 eq) in an argon-purged flask and then stirred for 1 h at 45 ^oC. The resulting solution was used immediately. The Grignard reagent (2.41 mL, 0.94 mmol, 4.0 eq) was added in a dropwise fashion to a solution of **3.70** (84 mg, 0.24 mmol) and $CuBr{\cdot}SMe_2$ (10.3 mg, 50 µmol, 21 mol%) in dry THF (1.1 mL) at -78 ^oC under argon. After stirring for 1 h at -78 ^oC, the solution was allowed to slowly warm to 0 ^oC and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. **3.67** (60 mg, 0.21 mmol, 90%) was isolated as a colorless oil after purification by column chromatography (20 g SiO_2, *n*-pentane). (10S,14S)-**3.67**: $[\alpha]_D^{20}$ = +2.09^o (c = 0.91, $CHCl_3$),), Lit.[5] $[\alpha]_D^{25}$ = +1.78^o. NMR data were as reported in the literature[5]: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.835, 0.838 (2d, *J* = 6.4 Hz, 6H), 0.89 (t, *J* = 6.8 Hz, 3H), 1.00-1.42 (m, 26H), 2.04 (q, *J* = 7.6 Hz, 2H), 4.93 (dd, *J* = 1.6, 10.4 Hz, 1H), 4.99 (dd, *J* = 1.6, 17.2, 1H), 5.82 (m, *J* = 6.8, 10.4, 16.8 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.1 (q), 19.6 (q), 22.9 (t), 24.4 (t), 27.0 (t), 28.9 (t), 29.1 (t), 29.3 (t), 29.4 (t), 29.6 (t), 29.8 (t), 32.6 (d), 33.7 (t), 36.7 (t), 37.0 (t), 37.3 (t), 114.0 (t), 139.2 (d) ppm. MS(EI) for $C_{20}H_{40}$: m/z = 280 [M^+], HRMS calcd for $C_{20}H_{40}$: 280.313, found: 280.313.



(5S,9S)-5,9-Dimethylheptadecane (3.71): hexyl bromide (0.38 mL, 2.69 mmol) in dry THF (6.0 mL) was added dropwise to Mg (91 mg, 3.74 mmol, 1.4 eq) in an argon-purged flask and then stirred for 1 h at 45 ^oC. The resulting solution was used immediately. The Grignard reagent (2.23 mL, 0.94 mmol, 5.7 eq) was added in a dropwise fashion to a solution of **3.70** (58 mg, 0.16 mmol) and $CuBr{\cdot}SMe_2$ (10.3 mg, 50 µmol, 31 mol%) in dry THF (1.1 mL) at -78 ^oC under argon. After stirring for 1 h at -78 ^oC, the solution was allowed to slowly warm to 0 ^oC and stirred overnight. Work-up was performed as described for **3.63**. GC-MS of the crude product showed that only **3.71** and dodecane (approximately 40%) were present. As the dodecane could not be removed by column chromatography (20 g SiO_2, *n*-pentane), the isolated yield of the reaction could not be determined. NMR data were as reported in the literature[5]: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.825, 0.827 (2d, *J* = 6.8 Hz, 6H), 0.89 (m, 6H), 1.03-1.40 (m, 28H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 14.1 (q), 19.6 (q), 22.6 (t), 23.0 (t), 24.4 (t), 27.0 (t), 29.3 (t), 29.6 (t), 30.0 (t), 31.9 (t), 32.7 (d), 36.7 (t), 37.1 (t), 37.3 (t) ppm. MS(EI) for $C_{19}H_{40}$: m/z = 268 [M^+], HRMS calcd for $C_{19}H_{40}$: 268.313, found: 268.313.

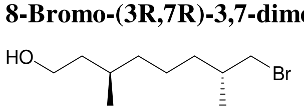


8-Bromo-(3R,7R)-3,7-dimethyl-octanoic acid methyl ester (3.72): (3*R*,7*R*)-**3.56** (120 mg, 0.59 mmol) was dissolved in dry THF (2.0 mL) and PPh_3 (171 mg, 0.65 mmol, 1.1 eq) and NBS (116 mg, 0.65 mmol, 1.1 eq) were added at 0 ^oC. The resulting solution was stirred under argon at 0 ^oC for 10 min and then allowed to warm to room temperature. Because TLC (*n*-pentane-Et_2O 5:1) showed incomplete conversion after 1 h, more PPh_3 (34 mg, 0.13 mmol, 0.2 eq) and NBS (23 mg, 0.13 mmol, 0.2 eq) were added, which resulted in

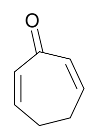


complete conversion after another 15 min. The crude mixture (suspension) was then filtered over cotton and the residue was washed with Et_2O. The filtrate was concentrated and purified by column chromatography (*n*-pentane-Et_2O 5:1) to give **3.72** (137 mg, 0.52 mmol, 87%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.93 (d, *J* = 6.6 Hz, 3H), 1.00 (d, *J* = 6.6 Hz, 3H), 1.10-1.47 (m, 6H), 1.77 (m, 1H), 1.94 (m, 1H), 2.02 (m, 1H), 2.12 (dd, *J* = 8.1, 14.7 Hz, 1H), 2.29 (dd, *J* = 6.0, 14.7 Hz, 1H), 3.28-3.45 (m, 2H), 3.66 (s, 3H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 18.6 (q), 19.6 (q), 24.0 (t), 30.1 (d), 34.8 (t), 35.0 (d), 36.5 (t), 41.3 (t), 41.5 (t), 51.2 (q), 173.5 (s) ppm. MS(CI) for $C_{11}H_{21}BrO_2$: m/z = 282 (M + $NH_4)^+$.

8-Bromo-(3R,7R)-3,7-dimethyl-octan-1-ol (3.73): To prepare a 1.0 M $AlCl_3$-solution, $AlCl_3$ (1.33 g, 9.97 mmol) was dissolved in dry Et_2O (10.0 mL).

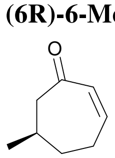


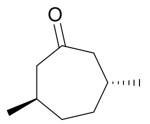
To a suspension of $LiAlH_4$ (19 mg, 0.49 mmol, 1.05 eq) in dry Et_2O (0.50 mL) was added $AlCl_3$ (1.0 M in Et_2O, 0.49 mL, 0.49 mmol, 1.05 eq). The resulting solution was cooled to -78 ^{o}C, after which a solution of **3.72** (123 mg, 0.46 mmol) in dry Et_2O (0.75 mL) was added. The mixture was stirred for 45 min at -78 ^{o}C under argon and then quenched with H_2O and diluted with aq. H_2SO_4 (2.0 M). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$), filtered and concentrated. The product was purified by column chromatography (*n*-pentane-Et_2O 3:1) to give **3.73** (89 mg, 0.38 mmol, 81%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.90 (d, *J* = 6.6 Hz, 3H), 1.01 (d, *J* = 6.3 Hz, 3H), 1.08-1.66 (m, 10H), 1.79 (m, 1H), 3.32 (dd, *J* = 6.0, 9.9 Hz, 1H), 3.39 (dd, *J* = 5.1, 9.9 Hz, 1H), 3.67 (m, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 75.5 MHz) δ = 18.6 (q), 19.4 (q), 24.1 (t), 29.2 (d), 34.9 (t), 35.1 (d), 37.0 (t), 39.8 (t), 41.4 (t), 61.0 (t) ppm. MS(CI) for $C_{10}H_{21}BrO$: m/z = 254 (M + $NH_4)^+$.



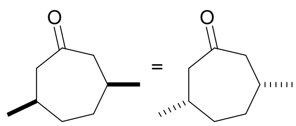
Cyclohepta-2,6-dienone (3.76): Prepared from cycloheptanone in 22% overall yield over four steps.[34] ^{1}H-NMR ($CDCL_3$, 300 MHz) δ = 2.45 (brs, 4H) 6.09 (d, J = 11.7 Hz, 2H), 6.60 (m, 2H) ppm.

(6R)-6-Methyl-cyclohept-2-enone (3.77):[33] $Cu(OTf)_2$ (42 mg, 0.12 mmol, 2.5 mol%) and R,S,S-L* (125 mg, 0.23 mmol, 5.0 mol%) were dissolved in dry toluene (8.0 mL) and stirred at room temperature for 1 h under nitrogen. Subsequently, a solution of **3.76** (500 mg, 4.62 mmol) in toluene (17 mL) was added, after which the mixture was cooled to -25 ^{o}C. Me_2Zn (2.0 M in toluene, 3.47 mL, 6.93 mmol, 1.5 eq) was added over 5 min and the resulting solution was stirred at -25 ^{o}C overnight and then quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x), washed with brine (sat.) and dried (Na_2SO_4). The Et_2O was removed under vacuum and the product was purified by column chromatography (*n*-pentane-Et_2O 9:1 to 4:1) to give (6*R*)-**3.77** (395 mg, 3.18 mmol, 68.8%) as a colorless liquid. ^{1}H-NMR ($CDCL_3$, 300 MHz) δ = 1.01 (d, *J* = 6.6 Hz, 3H), 1.53 (m, 1H), 1.89 (m, 1H), 2.07 (m, 1H), 2.41 (m, 3H), 2.67 (dd, *J* = 4.5, 14.4 Hz, 1H), 5.99 (d, *J* = 11.7 Hz, 1H), 6.62 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 75.5 MHz) δ = 21.9 (q), 28.2 (t), 28.4 (d), 34.7 (t), 51.3 (t), 132.6 (d), 147.2 (d), 203.0 (s) ppm.

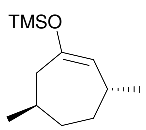




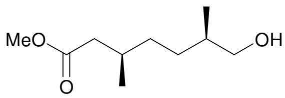
(3R,6R)-3,6-Dimethyl-cycloheptanone (3.78): The procedure was performed analogous to the preparation of **3.77**. (3*R*,6*R*)-**3.78** (44 mg, 0.31 mmol, 79%) was obtained from (6*R*)-**3.77** (50 mg, 0.40 mmol) using Me_2Zn (2.0 M in toluene, 0.30 mL, 0.60 mmol, 1.5 eq), R,S,S-L* (11 mg, 20 μmol, 5.0 mol%) and $Cu(OTf)_2$ (3.6 mg, 10 μmol, 2.5 mol%) in toluene (2.5 mL). Purification was achieved by column chromatography (*n*-pentane-Et_2O 9:1 to 4:1) to give **3.78** as a colorless liquid. ^{1}H-NMR ($CDCL_3$, 300 MHz) δ = 0.99 (d, *J* = 6.9 Hz, 6H), 1.25 (m, 2H), 1.82 (m, 4H), 2.38 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 24.1 (q), 31.5 (d), 38.2 (t), 52.2 (t), 213.5 (s) ppm.



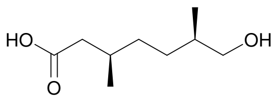
Meso-3,6-Dimethyl-cycloheptanone (3.79): The procedure was performed analogous to the preparation of **3.77**. Meso-**3.79** (63 mg, 0.45 mmol, 56%) was obtained from (6*R*)-**3.77** (100 mg, 0.81 mmol) using Me_2Zn (2.0 M in toluene, 0.61 mL, 1.21 mmol, 1.5 eq), S,R,R-L* (22 mg, 40 μmol, 5.0 mol%) and $Cu(OTf)_2$ (7.2 mg, 20 μmol, 2.5 mol%) in toluene (5.0 mL). Purification was achieved by column chromatography (*n*-pentane-Et_2O 9:1 to 4:1) to give **3.79** as a colorless liquid. ^{1}H-NMR ($CDCL_3$, 300 MHz) δ = 0.96 (d, *J* = 7.2 Hz, 6H), 1.54-1.70 (m, 4H), 2.04 (m, 2H), 2.23 (dd, *J* = 9.0, 14.7 Hz, 2H), 2.62 (dd, *J* = 3.6, 15.0 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 21.2 (q), 29.5 (d), 34.2 (t), 51.2 (t), 213.4 (s) ppm. MS(CI) for $C_9H_{16}O$: m/z = 158 $(M + NH_4)^+$.



((3R,6R)-3,6-Dimethyl-cyclohept-1-enyloxy)-trimethyl-silane (3.80): $Cu(OTf)_2$ (14.6 mg, 40 μmol, 2.5 mol%) and R,S,S-L* (43 mg, 81 μmol, 5.0 mol%) were dissolved in dry toluene (3.0 mL) and stirred at room temperature for 1 h under nitrogen. Subsequently, a solution of (6*R*)-**3.77** (200 mg, 1.61 mmol) in toluene (7.0 mL) was added, after which the mixture was cooled to -25 ^{o}C. Me_2Zn (2.0 M in toluene, 1.21 mL, 2.42 mmol, 1.5 eq) was added over 5 min and the resulting solution was stirred at -25 ^{o}C overnight. Subsequently, Et_3N (0.67 mL, 4.83 mmol, 3.0 eq), TMEDA (1.21 mL, 8.05 mmol, 5.0 eq) and a pre-stirred (30 min) solution of TMSOTf (0.87 mL, 4.83 mmol, 3.0 eq) and Et_2Zn (1.0 M in *n*- hexanes, 0.37 mL, 0.37 mmol, 0.23 eq) were added. The solution was stirred for 1 h while slowly warming to ambient temperature, after which the crude reaction mixture was flashed over a silica-plug (SiO_2 inactivated with Et_3N prior to use). The Et_2O was evaporated under vacuum and the product was purified by column chromatography (*n*-pentane; SiO_2 inactivated with Et_3N prior to use) to give (3*R*,6*R*)-**3.80** (254 mg, 1.20 mmol, 74%) as a volatile colorless liquid with still some toluene present. **3.80** was immediately used in the next step without further removal of the toluene. For the purpose of analysis the toluene was removed in vacuo from a small sample of the product. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.15 (s, 9H), 0.89 (d, *J* = 6.6 Hz, 3H), 0.97 (d, *J* = 6.6 Hz, 3H), 1.23 (m, 2H), 1.55 (m, 2H), 1.87 (m, 2H), 2.12-2.42 (m, 2H), 4.72 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 0.2 (q), 23.8 (q), 24.4 (q), 31.0 (d), 31.5 (d), 36.5 (t), 40.3 (t), 43.7 (t), 116.1 (d), 152.9 (s) ppm.



7-Hydroxy-(3R,6R)-3,6-dimethyl-heptanoic acid methyl ester (3.82): The procedure was performed analogous to the preparation of (3*R*,7*R*)-**3.56** from (3*R*,7*R*)-**3.52**. (3*R*,6*R*)-**3.80** (250 mg, 1.18 mmol) was converted into (3*R*,6*R*)-**3.82** (103 mg, 0.55 mmol, 46%) *via* carboxylic acid (3*R*,6*R*)-**3.81**. The carboxylic acid was used in the next reaction without purification. An analytical sample was purified by column chromatography (*n*-pentane-EtOAc 3:7) to give a colorless oil. Purification of **3.82** was achieved by column chromatography (*n*-pentane-Et_2O 2:1) to give a colorless oil as well.



(3*R*,6*R*)-**3.81**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.91 (d, *J* = 6.9 Hz, 3H), 0.97 (d, *J* = 6.6 Hz, 3H), 1.16-1.42 (m, 5H), 1.58 (m, 1H), 1.94 (m, 1H), 2.18 (dd, *J* = 7.8, 15.0 Hz, 1H), 2.34 (dd, *J* = 6.3, 15.0 Hz, 1H), 3.47 (m, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 50.3 MHz) δ = 16.3 (q), 19.5 (q), 30.1 (t), 30.3 (d), 33.6 (t), 35.6 (d), 41.3 (t), 68.1 (t), 178.3 (s) ppm. (3*R*,6*R*)-**3.82**: ^{1}H-NMR ($CDCl_3$, 300 MHz) δ = 0.90 (d, *J* = 6.9 Hz, 3H), 0.93 (d, *J* = 6.9 Hz, 3H), 1.12-1.41 (m, 5H), 1.57 (m, 1H), 1.93 (m, 1H), 2.14 (dd, *J* = 7.8, 14.7 Hz, 1H), 2.30 (dd, *J* = 6.3, 14.7 Hz, 1H), 3.42 (dd, *J* = 6.3, 10.5 Hz, 1H), 3.49 (dd, *J* = 6.0, 10.5 Hz, 1H), 3.66 (s, 3H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.3 (q), 19.5 (q), 30.1 (t), 30.4 (d), 33.7 (t), 35.7 (d), 41.5 (t), 51.3 (q), 68.1 (t), 173.6 (s) ppm. MS(CI) for $C_{10}H_{20}O_3$: m/z = 206 $(M + NH_4)^+$.

3.8 References

[1] (a) Mori, K. In *The Total Synthesis of Natural Products*, ed. J. ApSimon, Wiley-Interscience, New York, **1992**, 9, 1-524; (b) Nakamura Y.; Mori, K. *Eur. J. Org. Chem.* **1999**, 2175-2182; (c) Aldrich, J. R.; Oliver, J. E.; Lusby, W. R.; Kochansky J. P.; Borges, M. J. *J. Chem. Ecol.* **1994**, *20*, 1103-1111.

[2] (a) Mercier C.; Chabardes, P. *Pure Appl. Chem.* **1994**, *66*, 1509-1518; (b) Dowd, P.; Hershline, R.; Ham, S. W.; Naganathan, S. *Nat. Prod. Rep.* **1994**, *11*, 251-264; (c) Gramatica, P.; Manitto, P.; Monti, D.; Speranza, G. *Tetrahedron* **1986**, *42*, 6687-6692.

[3] (a) Minale, L. In *Marine Natural Products*, ed. P. J. Scheuer, Academic Press, **1978**, 1*,* ch. 4; (b) Sankaranarayanan, S.; Sharma A.; Chattopadhyay, S. *Tetrahedron: Asymm.* **2002**, *13*, 1373-1378.

[4] (a) Heathcock, C. H.; Finkelstein, B. L. *Science* **1985**, *229*, 862-864; (b) Zhang, D.; Poulter, C. D. *J. Am. Chem. Soc.* **1993**, *115*, 1270-1277; (c) Berkowitz, W. F.; Wu, Y. *J. Org. Chem.* **1997**, *62*, 1536-1539; (d) Eguchi, T.; Arakawa, K.; Terachi, T.; Kakinuma, K. *J. Org. Chem.* **1997**, *62*, 1924-1933.

[5] Nakamura, Y.; Mori, K. *Eur. J. Org. Chem.* **2000**, 2745-2753.

[6] Keyling-Bilger, F.; Schmitt, G.; Beck, A.; Luu, A. *Tetrahedron* **1996**, *52*, 14891-14904.

[7] Mulzer, J.; Berger, M. *Tetrahedron Lett.* **1998**, *39*, 803-806.

[8] Ferreira, J. T. B.; Zarbin, P. H. G. *Bioorg. Med. Chem.* **1996**, *4*, 381-388.

[9] Schulte-Elte, K. H.; Ohloff, G. *Helv. Chim. Acta* **1967**, *50*, 153-165.

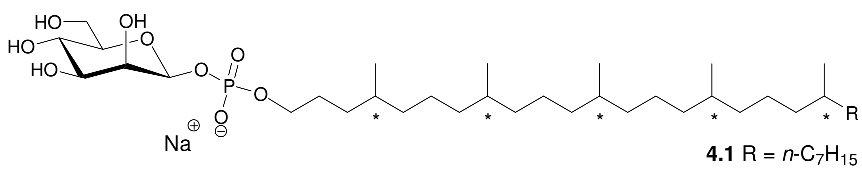
[10] Cohen, N.; Eichel, W. F.; Lopresti, R. J.; Neukom, C.; Saucy, G. *J. Org. Chem.* **1976**, *41*, 3512- 3515.

[11] Johnson, W. S.; Werthemann, L.; Bartlett, W. R.; Brocksom, T. J.; Li, T. -T.; Faulkner, D. J.; Peterson, M. R. *J. Am. Chem. Soc.* **1970**, *92*, 741-743.
[12] Donnelly, S.; Thomas, E. J.; Fielding, M. *Tetrahedron Lett.* **2004**, *45*, 6779-6782.
[13] Hanessian, S.; Murray, P. J.; Sahoo, S. P. *Tetrahedron Lett*. **1985**, *26*, 5623-5626.
[14] Heathcock, C. H.; Finkelstein, B. L.; Jarvi, E. T.; Radel, P. A.; Hadley, C. R. *J. Org. Chem.* **1988**, *53*, 1922-1942
[15] Evans, D. A.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2127-2129.
[16] Ireland, R. E.; Mueller, R. K.; Willard, A. K. *J. Am. Chem. Soc.* **1976**, *98*, 2868-2877.
[17] (a) Bérubé, G.; Deslongchamps, P. *Can. J. Chem.* **1984**, *62*, 1558-1560; (b) Bérubé, G.; Deslongchamps, P. *Bull. Soc. Chim. Fr.* **1987**, 103-115
[18] Still, W. C.; Darst, K. P. *J. Am. Chem. Soc.* **1980**, *102*, 7387-7389.
[19] Gramatica, P.; Manitto, P.; Poli, L. *J. Org. Chem.* **1985**, *50*, 4625-4628.
[20] Chênevert R.; Desjardins, M. *J. Org. Chem.* **1996**, *61*, 1219-1222.
[21] Takaya, H.; Ohta, T.; Sayo, N.; Kumobayashi, H.; Akutagawa, S.; Inoue, S.; Kasahara, I.; Noyori, R. *J. Am. Chem. Soc.* **1987**, *109*, 1596-1597.
[22] Huo, S.; Shi, J.; Negishi, E. -i. *Angew. Chem. Int. Ed.* **2002**, *41*, 2141-2143.
[23] Van Summeren, R. P.; Reijmer, S. J. W.; Feringa, B. L.; Minnaard, A. J. *Chem. Comm.* **2005**, *11*, 1387-1389.
[24] (a) Feringa, B. L.; Pineschi, M.; Arnold, L. A.; Imbos, R.; De Vries, A. H. M. *Angew. Chem. Int. Ed.* **1997**, *36*, 2620-2623; (b) Feringa, B. L. *Acc. Chem. Res.* **2000**, *33*, 346-353.
[25] Nicolaou, K. C.; Montagnon, T.; Baran, P. S.; Zhong, Y. -L. *J. Am. Chem. Soc.* **2002**, *124*, 2245-2258.
[26] Arnold, L. A.; Imbos, R.; Mandoli, A.; De Vries, A. H. M.; Naasz, R.; Feringa, B. L. *Tetrahedron* **2000**, *56*, 2865-2878.
[27] Knopff, O.; Alexakis, A. *Org. Lett.* **2002**, *4*, 3835-3837.
[28] Hon, Y. S.; Lu, L. *Tetrahedron Lett.* **1993**, *34*, 5309-5312.
[29] Gambacorta, A.; Tofani, D.; Lupattelli P.; Tafi, A. *Tetrahedron Lett.* **2002**, *43*, 2195-2198.
[30] Gries, R.; Gries, G.; King, G. G. S.; Maier, C. T. *J. Chem. Ecol.* **1997**, *23*, 1119-1130.
[31] Tamagawa, H.; Takikawa, H.; Mori, K. *Eur. J. Org. Chem.* **1999**, 973-978.
[32] Arrayás, R. G.; Cabrera, S.; Carretero, J. C. *Org. Lett.* **2003**, *5*, 1333-1336.
[33] Jagt, R. B. C.; Imbos, R.; Naasz, R.; Minnaard, A. J.; Feringa, B. L. *Isr. J. Chem.* **2001**, *41*, 221-229.
[34] (a) Garbisch, E. W. *J. Org. Chem.* **1965**, *30*, 2109-2120; (b) Krabbenhoft, H. O. *J. Org. Chem.* **1979**, *44*, 4285-4294.
[35] Tokuyama, T.; Tsujita, T. *Tetrahedron* **1991**, *47*, 5415-5424.

Chapter 4
β-D-Mannosyl Phosphomycoketides

4.1 Introduction

β-Mannosyl phosphomycoketides (MPM's) **4.1** and **4.2** are unusual mycobacterial antigens that were recently isolated in minute amounts by Moody and co-workers[1] from the cell wall of *Mycobacterium tuberculosis* and *Mycobacterium avium*, respectively (Figure 4.1). Initial structural characterization was based on degradation and mass spectrometric studies and did not include elucidation of the stereochemistry of the oligoisoprenoid chain.[1] In 2002, Dudkin and Crich reported the synthesis of **4.2**, confirming the overall molecular structure.[2] However, as a stereorandom alkyl chain (mixture of 32 diastereoisomers) was used, the stereochemistry of the lipid part remained unsolved.



4.1 R = *n*-C_7H_{15}
4.2 R = *n*-C_5H_{11}

Figure 4.1 *Molecular structure of β-mannosyl phosphomycoketides.*

Structurally, **4.1** and **4.2** resemble a partially saturated *β*-mannosyl heptaprenyl phosphate isolated from *M. smegmatis* (Figure 4.2), that serves as a carrier of mycolic acid.[3] However, an important difference from a biological point of view is that **4.1** and **4.2** violate the isoprene rule at both ends of their alkyl chain, which has implications for the biosynthetic pathway by which these saturated isoprenoid chains are assembled (see § 4.4).

From our perspective, **4.1** constituted a challenge to apply our procedure for the preparation of enantiopure saturated isoprenoid building blocks (Chapter 3) to a more complex target. Hence, we aimed to develop a general protocol for the construction of enantiopure MPM's with oligoisoprenoid chains of any desired stereochemistry and length.

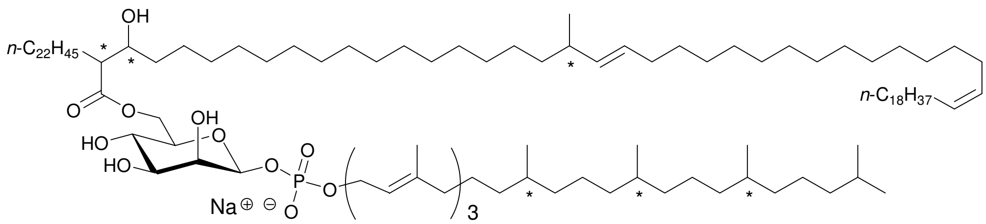


Figure 4.2 *Related β-mannosyl heptaprenyl phosphate isolated from M. Smegmatis.*[3]

4.2 Lipid-antigen mediated immune response

The success of the (human) immune system depends on its ability to distinguish foreign antigens from self-elements and to initiate appropriate effector mechanisms that destroy alien intruders without inflicting damage to self-elements.[4] The mammalian immune system utilizes two major recognition strategies: innative and adaptive. Innate immune responses use pattern-recognition receptors such as lectins and the Toll-like receptors to recognize specific foreign molecules that are characteristic for microbial species. By focusing on conserved structural motifs that are common among pathogens, the innate immune system is able to respond immediately in case of infection. In contrast, the adaptive immune system uses recombinatorial antigen-receptors and is therefore able to interact with an almost infinite number of structures, although its response is generally slower. The main mediators of adaptive immune responses are T and B lymphocytes. While B-cells produce antibodies that can interact with epitopes of any molecular species, T-cells were believed until recently to be activated exclusively by peptides which are presented by major histocompatibility (MHC) class I and II proteins.[5] In 1992, though, Porcelli *et al.* showed that a second class of antigen presenting proteins, known as the Cluster of Differentiation 1 (CD1) family, is capable of inducing proliferation of T-cells which are specific for microbial pathogens.[6] Two years later they disclosed, that CD1 molecules have in fact the ability to present lipid antigens rather than peptides.[7] Finally, in 2000, Moody *et al.* provided strong evidence that CD1-lipid complexes indeed play an important role in resistance to microbial infections *in vivo via* activation of T-cells.[1,8] Since then, four members of the CD1 family (CD1a to CD1d) have been proven to present at least five classes of (glyco)lipids:[9] mycolates,[7] glycosphinglolipids,[10] phospholipids,[11] sulfoglycolipids[12] and lipopeptides.[13] Based on sequence similarities, the CD1 isoforms have been divided in two groups: CD1a, b, and c (group 1) and CD1d (group 2). The fact that many CD1d restricted T-cells express a semi-invariant T-cell antigen receptor (TCR) suggests that they may function as a pattern-recognition receptor of the innate immune system, while CD1a, b and c are part of the adaptive immune system.[5]

From a chemical point of view, lipids seem to be an unlikely target for recognition by T-cells. They are insoluble in the aqueous environment in which antigen-presenting molecules and T-cells function. Moreover, the high degree of conformational freedom of a long aliphatic chain and the absence of functional groups for hydrogen bonding, makes specific interactions with T-cell receptors (TCRs) unlikely. This paradox was partly resolved, when crystal structures of CD1 proteins showed the presence of large hydrophobic cavities.[14] Based on these crystal structures and on investigations to establish the molecular determinants for antigen recognition,[15] a model was proposed in which van der Waals interactions are maximized by embedding of the aliphatic chain(s) into the hydrophobic groove, while the polar headgroup is presented at the surface to interact with the TCR (Figure 4.3).[4,14a]

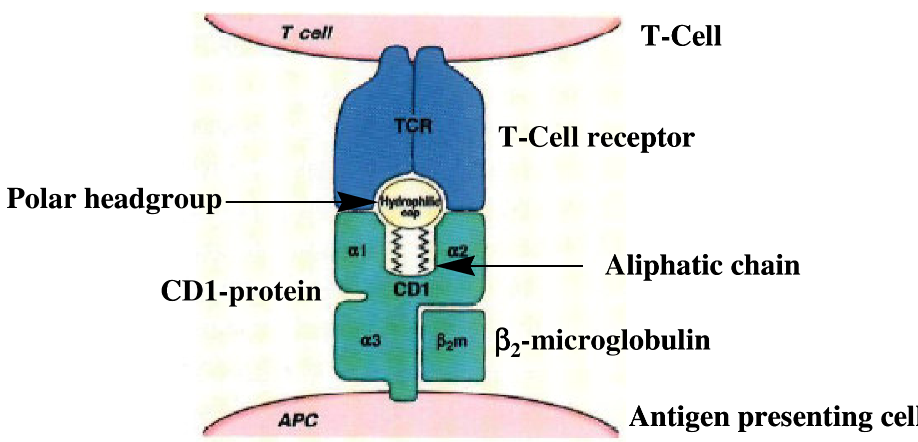


Figure 4.3 *Trimolecular model of CD1-glycolipid antigen-T-Cell receptor interactions*.[4]

4.2.1 CD1 proteins and lipid antigen presentation

Considering the limited α-amino acid sequence homology (< 36%) between CD1 proteins and MHC class I proteins, their three-dimensional structures are remarkably similar. Both antigen presenting molecules are integral membrane glycoproteins containing three domains (α1, α2 and α3), that form non-covalent heterodimers with β_2-microglobulin (Figure 4.3).[4] When evaluating tissue expression, CD1 proteins are more reminiscent of MHC class II proteins, in that they are mainly present on specialized antigen-presenting cells (APC's) like B cells, macrophages and dendritic cells. However, unlike MHC class I and class II proteins, human CD1 proteins are nonpolymorphic so that genetically unrelated individuals express nearly identical CD1 gene products. It is speculated that the overall lack of polymorphism of CD1 alleles is related to the limited structural variation in lipid tails of microbial species. In contrast to proteins, lipids and glycolipids are products of multi-enzyme biosynthetic pathways and their structures are therefore highly conserved.[4,16] Moreover, (glyco)lipids are responsible for the integrity of the mycobacterial cell envelope, so that small changes in their structure are often detrimental for the survival of the bacterium.[17] Incidentally, this conservation of structure makes these non-mammalian (glyco)lipids interesting biological targets for both drugs (such as isoniazid) and vaccines.

CD1 molecules are synthesized in the endoplasmic reticulum (ER), where N-linked glycans are attached and association with β_2-microglobulin occurs.[18] ER chaperones and self lipids subsequently assist proper folding,[19] after which the CD1 complex is transported from the Golgi apparatus to the plasma membrane, presumably along the secretory pathway (Figure 4.4).[20] Surface CD1 proteins are then internalized, in case of CD1b and CD1c through the interaction of a tyrosine-motif in the cytoplasmic domain with adaptor protein complex 2 (AP2). Specific CD1 molecules are transported to particular intracellular compartments where the previously bound self lipids are exchanged for distinct foreign- or other self lipids. CD1a is predominantly targeted to recycling endosomes, CD1b is sorted to late endosomes and lysosomes, CD1c broadly traffics to all compartments and CD1d is found mainly in early and late endosomes.

The transport of lipids from hydrophobic environments like membranes and CD1 grooves to the aqueous lumens of intracellular compartments is a thermodynamically unfavorable process. Recent studies on the regulation of antigen presentation by human

CD1b and CD1d show that the loading of lipids is assisted by sphinglolipid activator proteins (SAP), presumably through their interaction with the polar heads of lipids.[9,21] The pathways that direct the CD1 molecules from the internal compartments back to the plasma membrane have not been elucidated yet.

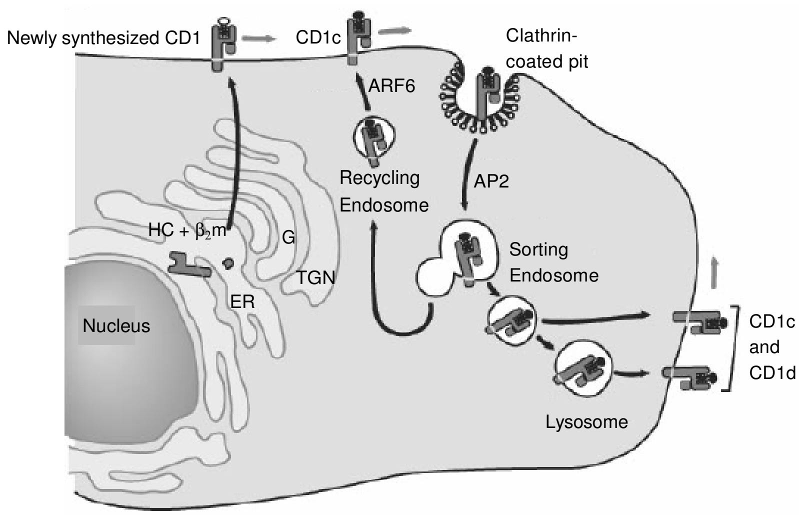


Figure 4.4 *Intracellular trafficking and loading of CD1c and CD1d molecules.*[16]

4.2.2 Recognition of CD1-lipid complexes by T-cell antigen receptors

T-cell antigen receptors (TCRs) that recognize peptide antigens bound in the groove of MHC molecules are heterodimers consisting of an α and a β chain. The specific interaction is mediated by the complementarity-determining residue (CDR) loops of the α and β chains which contact exposed residues of the peptide antigen and α-amino acids along the MHC α helices. The three-dimensional resemblance of CD1 molecules to MHC class I proteins (§ 4.2.1) allows the same type of α/β TCR heterodimers to specifically interact with CD1-lipid complexes as well. Indeed, the primary structure of TCRs that recognize CD1a, b and c in the presence of lipid antigens are indistinguishable from those that recognize MHC class I or II complexes with peptides.[22]

In general, TCRs are highly specific for the epitope that is formed by the combination of a particular CD1 family member and a certain lipid antigen, even though there are some reports of self-recognition of CD1 in the absence of an antigen.[23] The molecular basis of the

CD1-lipid-TCR interaction awaits the determination of the structure of the tri-molecular complex (Figure 4.3). Frequent usage of basic α-amino acids in the CDR regions of the TCR suggests a model in which these CDR loops project directly between the CD1 α helices to participate in electrostatic interactions with the polar headgroups of the lipid antigens.[22] In concurrence with this model, TCRs can distinguish even small changes in the structure of the hydrophilic headgroup of antigens,[7,15,24,25] while alterations in the aliphatic tail often have less pronounced effects. Nevertheless, the influence of the aliphatic tail cannot be dismissed as inconsequential, since it often provides the (only) principal basis for discrimination of self- and non-self structures.[26]

4.2.3 Effector functions of T-cells

In the peptide mediated system, class I MHC-peptide complexes can be found at the surface of infected cells, where they specifically bind to cytolytic T-cells having a $CD8^+$TCR. Upon recognition, the cytolytic T-cells initiate an immune response which eventually leads to the lysis of the infected cells as well as killing of the invading pathogen. Class II MHC-peptide complexes on the other hand are presented at the surface of B cells, macrophages and dendritic cells where they specifically interact with T-helper cells (Th1/Th2) having a $CD4^+$TCR. In this case, recognition results in the secretion of a variety of cytokines which stimulate the maturation, proliferation and activation of a range of different T-cells, B-cells, macrophages and so on. *De facto*, a cell is either killed or proliferated depending on the presented MHC-peptide complex and a strict separation of these activities is obviously instrumental for the proper functioning of the immune system.

In this light, it is noteworthy, that group 1 CD1-lipid complexes are recognized by both T-cells expressing a $CD8^+$TCR as well as by T-cells having a $CD4^+$TCR among others. To make the situation even more complex, some CD1-restricted T-cells display both effector functions characteristic for cytolytic T-cells as well as features of T-helper cells.[5] Indeed, the same CD1-restricted T-cell that is involved in the lysis of infected cells and in direct antimicrobial activity *via* the secretion of perforin and granulysin, respectively,[27] can also be responsible for the production of interferon-γ (IFN-γ) and tumor necrosis factor-α (TNF-α), which activate the microbicidal functions of macrophages (Figure 4.5).[5,16] How the lipid-based immune system distinguishes infected cells, which have to be lysed, from B cells, macrophages and dendritic cells, which have to be proliferated, remains to be elucidated.

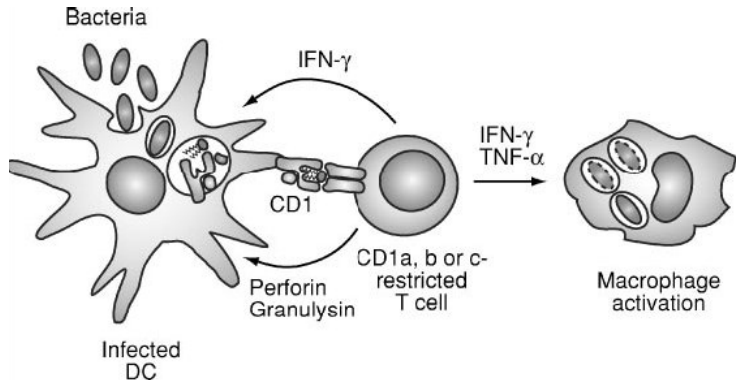


Figure 4.5 *Effector functions of CD1-restricted T cells.*[16]

4.2.4 Rationale for a lipid-antigen mediated immune response

The rationale for a lipid-antigen mediated immune response is still a subject of debate. It may be that the lipid-antigen mediated pathway serves to identify infected cells that inefficiently present protein antigens, thus providing a complementary mechanism.[5] In addition, CD1 molecules are expressed on immature dendritic cells, suggesting that CD1-restricted T-cells may function at an earlier phase of the immune response than MHC-restricted T-cells.[5] Finally, evasion of immune responses mediated by protein antigens may readily occur by point mutations that do not affect protein function, while (glyco)lipid structures are highly conserved (*vide supra*). However, by definition, the tailor-made system for an instant immune response against antigens with a common motif is the innate immune system. Hence, the first explanation for the existence of an adaptive immune system based on (glyco)lipids seems to be more plausible than the latter two arguments.

4.3 CD1c-mediated T-cell recognition of isoprenoid glycolipids

In 2000, it was reported that CD1c-presentation of two previously unknown *β*-D-mannosyl phospholipids (**4.1** and **4.2**) isolated from *Mycobacterium tuberculosis* resulted in recognition by T-cells (CD8-1) expressing a $CD8^+$TCR.[1] Notably, this was the first example of a lipid antigen having a single alkyl chain instead of the more common motif of two straight aliphatic hydrocarbon tails connected by a central hydrophilic cap. To gain further insight into the molecular determinants that affect recognition, a range of semi-synthetic glycosyl-β1-phosphoisoprenoid compounds were prepared and tested. As predicted by the tri-molecular TCR-CD1-antigen model (§ 4.2.2 and Figure 4.3), the outcome demonstrated a fine specificity for the hydrophilic headgroup. Both removal of the mannose or exchange for glucose resulted in a complete loss of activity. In addition, a saturated α-prenyl unit was required, but both *cis* and *trans* double bonds elsewhere in the alkyl chain were tolerated. Finally, T-cell response appeared inversely proportional to prenyl length. A glycolipid with an alkyl tail of 35 carbons responded stronger than one with a C55-tail, while the analogue with a C95-tail was completely inactive.

Since the above study was performed using living cells rather than immobilized CD1 proteins, it is unclear what is causing changes in TCR recognition. It could be that changes in the structure of the lipid antigen result in different affinity for the CD1 protein or that the CD1-antigen complex interacts differently with the TCR. However, it is also possible that modification of the lipid antigen influences intracellular trafficking and delivery of the glycolipids to relevant antigen-loading compartments. Likewise, it is also uncertain whether the fact that only CD1c presents lipid antigens with a single alkyl chain is due to specialization of the CD1c groove or to the loading pathway of the antigen. Related to this, the dilemma remains how the immune system discriminates between self and foreign mannosyl-β1-phosphate lipids.[28] Even though a mannosyl-β1-phosphate moiety with a saturated α-prenyl unit and a short tail seems both necessary and sufficient for recognition, the saturated isoprenoid chain of *β*-D-mannosyl phosphomycoketides has to be the key element responsible for selectivity. To obtain insight into the subtle mechanisms that underlie selectivity, complementary information should be acquired by *in vitro* and *in vivo* tests. A necessity for such experiments is the availability of sufficient and pure (glyco)lipid antigens of any desired composition. Considering the slow growth of *M. tuberculosis* and the minute amounts of *β*-mannosyl phosphomycoketide that it produces, total synthesis seems to be the only way to fulfill this requirement at present. Assembly by total synthesis has the additional advantage that non-natural antigens are accessible as well.

4.4 Biosynthesis of mycoketides[26]

In the original paper on CD1c-mediated T-cell recognition of isoprenoid glycolipids the authors commented on the molecular structure of the antigens (**4.1** and **4.2**) in the following way: 'Unusual features of the newly discovered mycobacterial isoprenoids (…) included the complete saturation of their alkyl chains, and a deviation from the repeating five carbon prenyl unit at the proximal and distal ends of these chains.'[1] Since the structure of the saturated alkyl chain could not be explained by a conventional C_5 isopentenyl pyrophosphate biosynthetic pathway, an investigation was started to determine the genetic and enzymatic basis of **4.1** and **4.2**. Metabolic labeling and mass spectrometric analysis suggested an elongation mechanism using alternating C_2 and C_3 units, which is indicative for a polyketide synthase pathway. Inspection of the *M. tuberculosis* genome identified one candidate gene, *pks12*, which was predicted to encode a large protein consisting of 12 contiguous but separately functioning catalytic domains. Knockout and complementation of *pks12* established that this gene was indeed responsible for the production of the saturated isoprenoid tails of **4.1** and **4.2**. As a consequence, **4.1** and **4.2** were thereafter referred to as mannosyl phosphomycoketides (MPM) rather than isoprenoids. Interestingly, mannosyl-β-1-phospholipids with true polyisoprenol lipid moieties, with their biosynthesis not disrupted by *pks12* genetic deletion, did not substitute as potent antigens for the T-cell response.

During the elucidation of the biosynthetic pathway, MPM's with a tail length of 31, 33 and 34 carbons were observed next to the already known species with 30 (**4.2**) and 32 (**4.1**) carbons. A plausible polyketide based mechanism for the biosynthesis of all these structurally related mycoketides starts from different carboxylate primers of varying length. Subsequently, (two) acyltransferases and (two) ketosynthases add malonate (C_2) and methylmalonate (C_3) units in an alternating fashion. After every C_2- or C_3-addition, the consecutive action of a ketoreductase (to the alcohol), a dehydratase (to the alkene) and an enoylreductase provides the saturated system (see Scheme 2.1). Since C_2-addition is followed by C_3-addition and vice versa rather than at random, it is likely that two sets of catalytic domains are required, one for malonate units and one for methyl malonate units. The stereochemistry of the five asymmetric centers is determined by the enoylreductase following C_3-addition. The final carboxylate moiety has to be reduced to the alcohol and phosphorylated to yield a substrate for glycosylation.

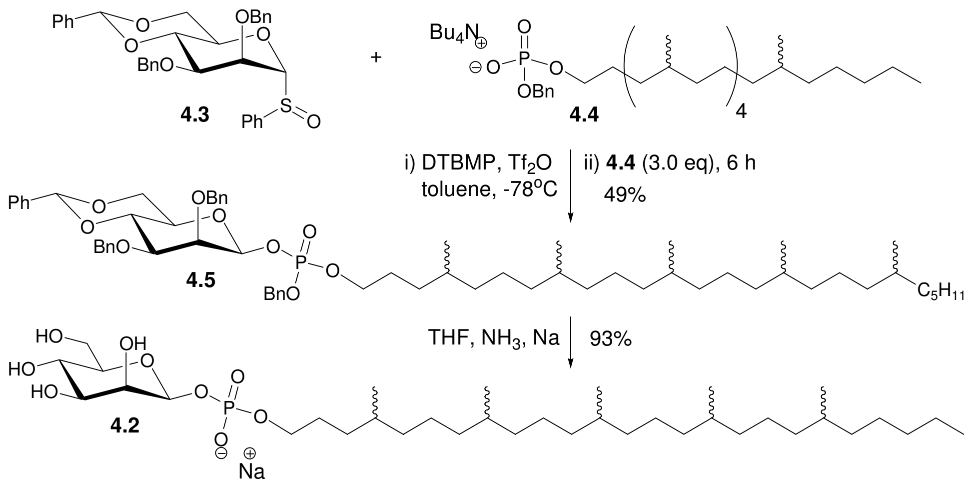
The unraveling of the biosynthetic pathway of MPM's has raised speculations regarding their biological function. Their complex biosynthesis is energetically expensive and conserved in infectious mycobacteria while lacking in non-virulent mycobacteria. This suggests that they may have a highly specialized role that is essential for intracellular growth. In agreement with this hypothesis, MPM's are found at low absolute levels; they make up about 1 ppm of the total weight of the cell wall. Possibly MPM's are involved in mannose transmembrane transport or transfer.[26]

4.5 Synthesis of stereorandom *β*-D-mannosyl phosphomycoketides

The central theme in the synthesis of *β*-D-mannosyl phosphomycoketides by Crich and Dudkin,[2] was the formation of the *β*-mannosyl phosphate linkage. As a result of steric repulsion caused by the 1,2-*cis* configuration and instability due to the anomeric effect, *β*-mannosyl glycosidic linkages in general are notoriously difficult to bring about.[29] However, in a series of publications, Crich *et al*. disclosed an elegant method for the direct formation of such linkages.[30] Their procedure involves *in situ* conversion of a 4,6-*O*-benzylidene protected α-mannosyl sulfoxide (for example **4.3**; Scheme 4.1) into the corresponding α-anomeric triflate. Subsequent addition of an acceptor leads to S_N2 type displacement to give the desired β-mannoside with high selectivity.[31] The origin of this high selectivity lies in the torsional disarming propterties of the 4,6-*O*-benzylidene moiety, which opposes flattening of the molecule and thus prevents the formation of the oxocarbenium ion necessary for S_N1 type reactions.[32]

In order to construct MPM **4.2**, the general mannosylation protocol discussed above was applied to the coupling of sulfoxide donor **4.3**, which was prepared in six steps from D-mannose, to phosphate acceptor **4.4** (Scheme 4.1). Since formation of the *β*-D-mannosyl phosphate linkage was the main goal of the investigation, a stereorandom mixture of **4.4** was used consisting of 32 stereoisomers. In the event, **4.3** was reacted with Tf_2O (1.1 eq) in toluene at -78 ^{o}C in the presence of acid scavenger 2,6-di-*tert*-butyl-4-methylpyridine (DTBMP), followed by the addition of three equivalences of **4.4** to give the desired product

(**4.5**) in 49% yield after six hours. Subsequent deprotection *via* Birch reduction (Na, NH_3) gave the final product **4.2** with exclusively the β-configuration in 93% yield.



Scheme 4.1 *Synthesis of racemic β-D-mannosyl phosphomycoketide **4.2** by Crich et al.*[2]

4.6 Total synthesis of enantiopure *β*-D-mannosyl phosphomycoketides

To study the effect of the stereochemistry of the saturated alkyl chain on the activity of MPM's, a synthetic protocol that is capable of accessing any desired diastereomer is required. Hence, we aimed at the development of a general (catalytic) strategy using enantiopure saturated isoprenoid building blocks (see chapter 3) as key intermediates.

As explained in § 4.4, the stereochemistry of natural phosphomycoketides is determined by an enoylreductase following the introduction of methylmalonate. Provided that a single catalytic domain is responsible for this transformation, the product most likely has an all-*syn* configuration. Since the authors, who elucidated the biosynthetic pathway, predicted the stereochemistry to be all-S (private communication), we decided to direct our initial efforts to the preparation of this diastereomer of **4.1**.

4.6.1 Retrosynthesis

When we started the synthesis of enantiopure MPM's, we envisioned that three requirements had to be fulfilled in order to prepare useful amounts of final product. First of all, since connection of two saturated isoprenoid units (**A** and **B** in Figure 4.6) would generate an alkyl chain with four stereogenic centers, an additional (catalytic asymmetric) method had to be found for the introduction of the fifth asymmetric center (**C**). Conveniently, the catalytic asymmetric conjugate addition of Grignard reagents to α,β-unsaturated thioesters was under investigation in our group at that moment and we planned to employ this procedure to obtain fragment **C**.[33] Secondly, as the various linear chiral building blocks (**A**, **B** and **C**) are relatively valuable, an efficient procedure had to be found to cross-couple them without using a large excess of either coupling partner. Related to this, formation of the *β*-D-mannosyl phosphate linkage as described by Crich and Dudkin was undesirable,[2,30g] since it would require three equivalents of precious oligoisoprenoid chain. We therefore intended to either adapt their methodology in such a way that an excess of sulfoxide donor **4.3** could be used or to employ an entirely different coupling strategy.

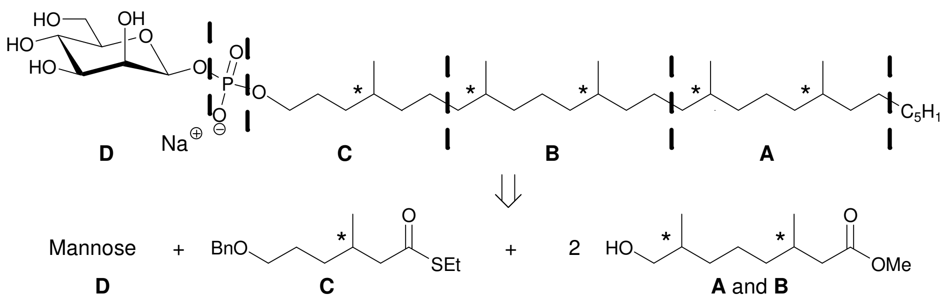


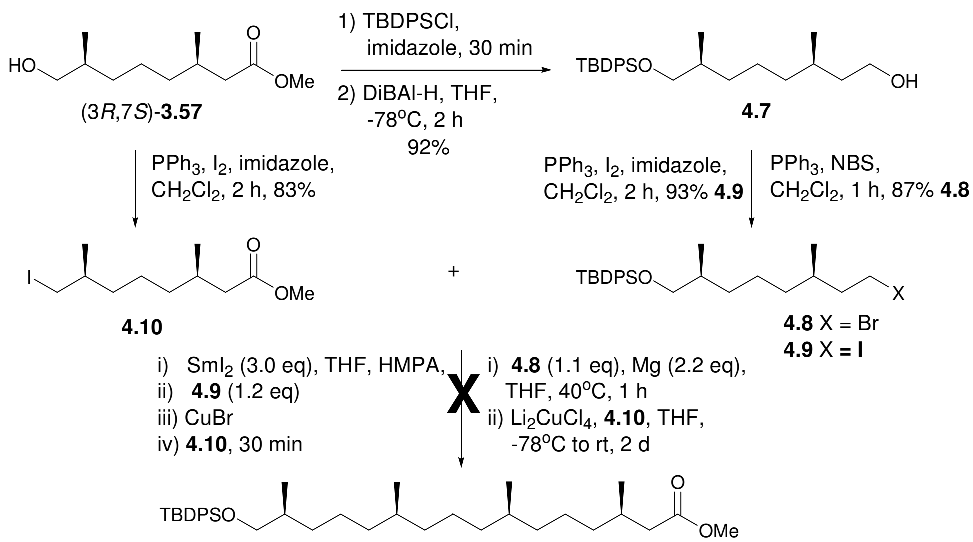
Figure 4.6 *Retrosynthetic analysis of β-mannosyl phosphomycoketide **4.1**.*

4.6.2 Synthesis of diisoprenoids

The direct formation of sp^3-sp^3 carbon bonds in the presence of other functionalities, like for example (protected) alcohols or esters, is still a difficult transformation in organic synthesis. This particularly holds true when a large excess of either coupling partner has to be avoided, like in the synthesis of an expensive homo-chiral saturated isoprenoid chain. The classical method to cross-couple isoprenoids in order to obtain saturated diisoprenoids relies on an indirect approach. It consists of a S_N2 reaction between an α-deprotonated alkyl sulfone and an alkyl halide followed by a metal reduction (see scheme 3.1).[34] However, the reductive removal of the sulfone proceeds under harsh conditions, that preclude the presence of, for example, esters. Moreover the yield in this second step is somewhat unreliable as it ranges from modest to good (50-85%). For the connection of our linear fragments, we decided to search for a milder and less capricious direct or indirect coupling procedure.

4.6.2.1 Copper-catalyzed Grignard and SmI_2-assisted cross-couplings

In 1997, Berkowitz and Wu described the direct formation of saturated diisoprenoids *via* copper-catalyzed cross-couplings using Grignard and samarium chemistry.[35] Our attempt to reproduce their results started with the protection of the hydroxyl moiety of (3R,7S)-**3.57** *via* reaction with *tert*-butyldiphenylsilyl chloride (TBDPSCl) in the presence of imidazole (**4.6**, 95%, Scheme 4.2). The methyl ester was then reduced with DiBAl-H to afford primary alcohol **4.7** (97%). Subsequent treatment of **4.7** with either triphenylphosphine and NBS or with PPh_3, iodine and imidazole furnished primary alkyl halides **4.8** (87%) and **4.9** (93%), respectively. In an analogous fashion, (3R,7S)-**3.57** was converted into primary iodide **4.10** in 83% yield.



Scheme 4.2 *Preparation of diisoprenoids using Grignard/samarium cross-coupling*.

Disappointingly, Li_2CuCl_4-catalyzed Grignard coupling of **4.8** to **4.10** did not result in the formation of the saturated diisoprenoid in our hands (lit.[35] 71%). Furthermore, SmI_2

mediated cross-coupling of **4.9** and **4.10** in the presence of HMPA and catalytic copper bromide (20 mol%) was unsuccessful as well (lit.[35] 81%).

Careful optimization of the SmI_2-mediated reaction using comparable (non-chiral) compounds eventually gave yields of up to 40% in model reactions, while the isolated yields with magnesium were even lower. In the latter case, the problems were partly due to inefficient formation or instability (degradation and/or homocoupling) of the Grignard reagent. Titration with *sec*-butanol in the presence of 1,10-phenanthroline revealed that approximately only 60% of the alkyl bromide was converted into the corresponding active Grignard reagent. With regard to the SmI_2-mediated reaction, the initial $RSmI_2$ species (**A**, Figure 4.7) was readily formed as no starting alkyl halide was recovered. However, beside the desired cross-coupled product, side-products resulting from β-elimination, β-hydride transfer as well as homocoupling were observed by GC-MS.

According to the proposed mechanism, acceleration of the desired reaction by increasing the catalyst loading is not possible unless the excess of starting alkyl halide (RI, Figure 4.7) is also increased.[36] To start the catalytic cycle, part of species **A** has to form a complex (**B**) with CuBr and this amount is thereafter permanently tied up in the catalytic cycle. As a consequence, an amount of the starting alkyl halide RI equal to the catalyst loading is lost by definition. All things considered, both coupling procedures were abandoned as they consistently gave poor yields and were unlikely to perform at an acceptable level in equimolar cross-coupling reactions.

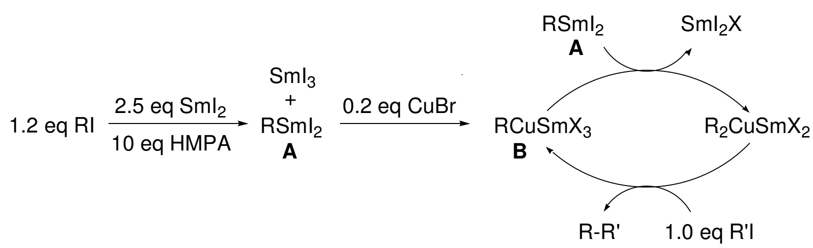


Figure 4.7 *Proposed mechanism of the copper-catalyzed cross coupling of alkyl samarium reagents with alkyl halides.*[36]

4.6.2.2 Cross-couplings via formation of an olefin

As an alternative approach, we focused our attention on coupling procedures which generate a carbon-carbon double bond, which would then later be reduced to the saturated system. In a comparative study, we evaluated the performance of the Wittig, the Horner-Wadsworth-Emmons (HWE) and the Julia-Kocienski[37] coupling for our system. The Julia-Kocienski coupling is a one-pot procedure (Figure 4.8) derived from the Julia-Lythgoe olefination,[38] which required a separate reductive elimination step.

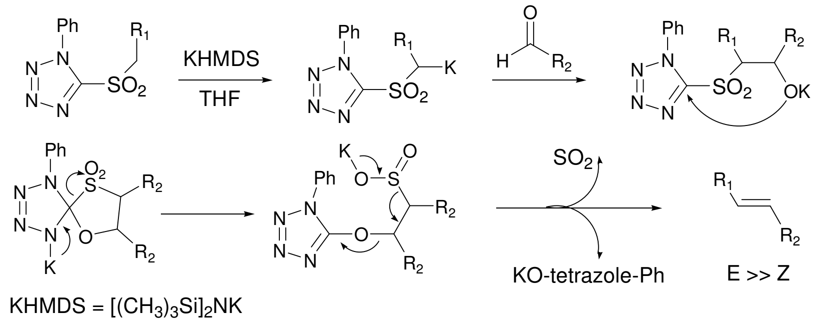
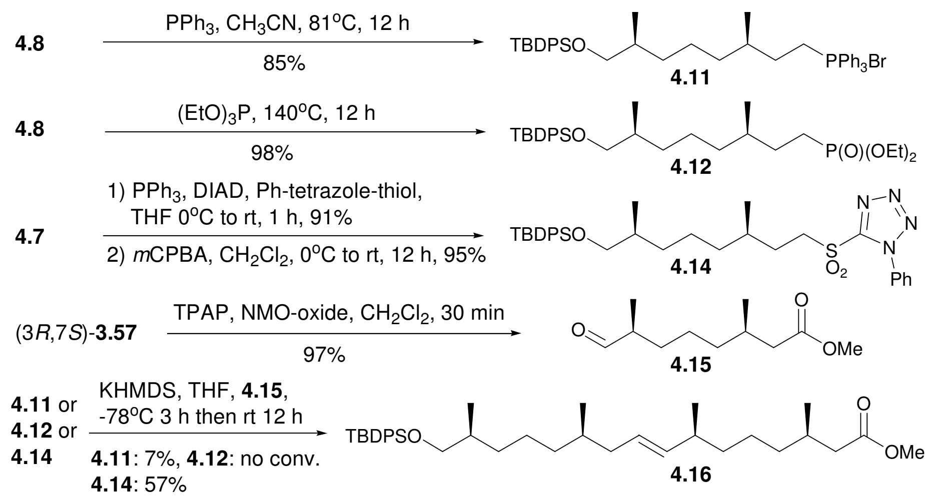


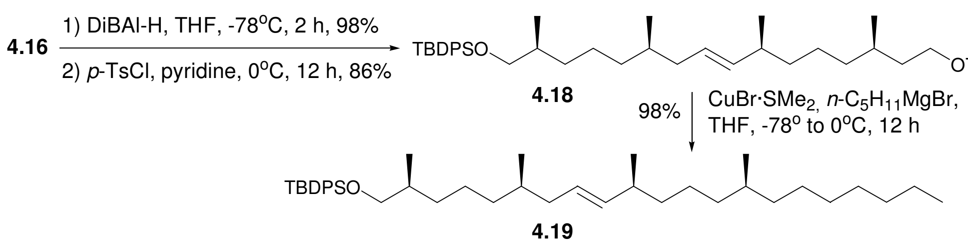
Figure 4.8 *Mechanism of the Julia-Kocienski coupling*.[37]

The Wittig reagent **4.11** was obtained in 85% yield from alkyl bromide **4.8** *via* reaction with PPh_3 (Scheme 4.3). Subjection of **4.8** to an Arbuzov reaction with triethylphosphite furnished HWE-reagent **4.12** (98%). Finally, the Julia-Kocienski sulfone was prepared from alcohol **4.7** *via* Mitsunobu reaction with 1-phenyl-1H-tetrazole-5-thiol (**4.13**, 91%) followed by oxidation of the resulting thioether with *meta*-chloroperbenzoic acid (*m*CPBA) to give **4.14** (95%).[39] Compounds **4.11**, **4.12** and **4.14** were subjected to coupling reactions with aldehyde **4.15**, which was obtained in 97% yield from (3R,7S)-**3.57** *via* oxidation with 4-methylmorpholine N-oxide (NMO-oxide) in the presence of a catalytic amount of tetra-*n*-propylammonium perruthenate (TPAP).[40] All three olefination reactions were performed in THF using a slight excess of **4.15** (1.1 eq) and potassium hexamethyldisilazane (KHMDS 1.0 eq) as the base. The Wittig reaction afforded diisoprenoid **4.16** in minute amounts (7%), while the desired product was not formed at all in the HWE-reaction. The Julia-Kocienski coupling gave the best result with an isolated yield of 57%. The reason why the Julia-Kocienski coupling performed better for this system than conventional Wittig- and HWE-reactions is unclear.

The next goal was to convert the methyl-ester of **4.16** into a terminal linear saturated C7 chain, for which we intended to employ the Grignard extension methodology already used in the synthesis of the apple leafminer pheromones (§ 3.4). In the event, the methyl ester was reduced to the corresponding primary alcohol **4.17** (98%) with DiBAl-H, after which reaction with *p*-toluenesulfonyl chloride in pyridine afforded **4.18** in 86% yield (Scheme 4.4). Subsequent treatment with *n*-pentylmagnesium bromide and semi-catalytic $CuBr{\cdot}SMe_2$ yielded **4.19** in 98%.

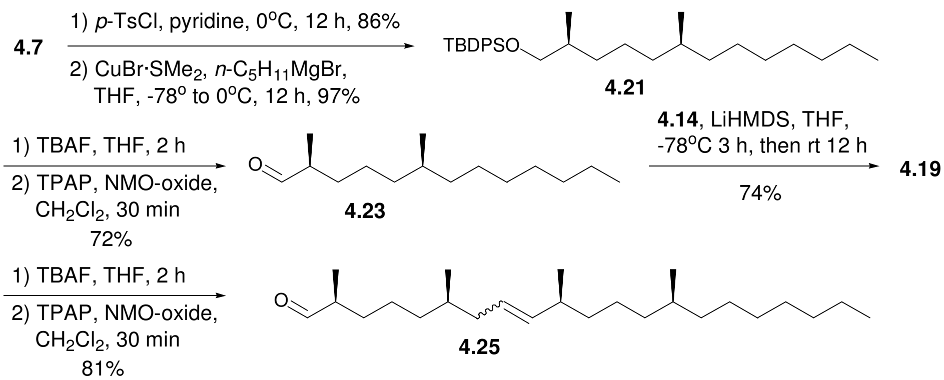


Scheme 4.3 *Comparison of carbon-carbon double bond cross-coupling methods.*



Scheme 4.4 *Introduction of the saturated alkyl terminal chain.*

Although an isolated yield of 57% in the preparation of diisoprenoid **4.16** was acceptable, we decided to investigate whether removal of the ester in **4.15** would be beneficial for the Julia-Kocienski coupling. To this end, an isoprenoid building block was elongated to obtain the terminal *n*-heptyl function prior to cross-coupling (Scheme 4.5). This slightly modified approach had the additional advantage that it made the route somewhat more convergent.



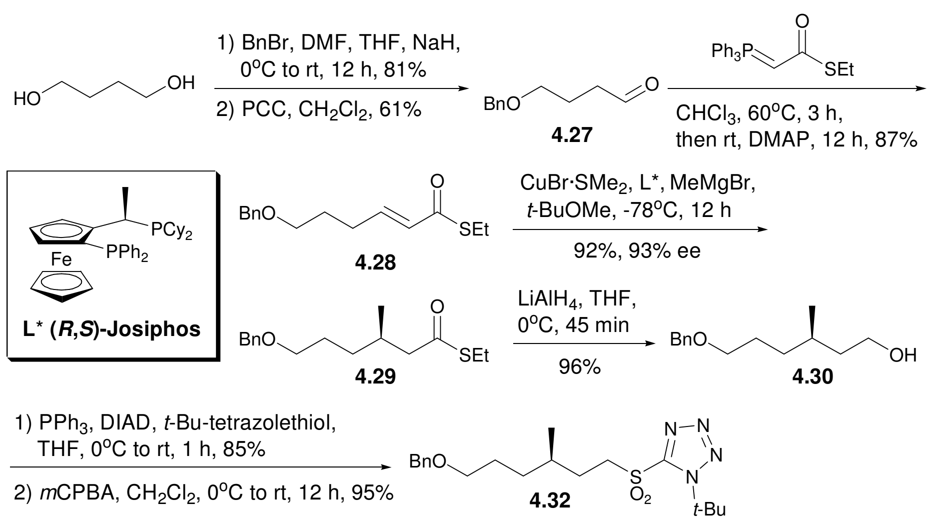
Scheme 4.5 *Synthesis of diisoprenoid **4.25**.*

The conversion of **4.7** into the corresponding tosylate **4.20** (86%) and the subsequent coupling with *n*-pentylMgBr to obtain **4.21** (97%) was performed as described for the transformation of **4.17** into **4.19**. Treatment of **4.21** with TBAF yielded alcohol **4.22**, after which oxidation with NMO-oxide and TPAP furnished aldehyde **4.23** (72% over 2 steps). Julia-Kocienski coupling of **4.23** to sulfone **4.14** under comparable conditions as for the preparation of **4.16**, initially gave diisoprenoid **4.19** in similar yield (50-60%). However, changing the base from KHMDS to LiHMDS and mixing **4.23** and **4.14** prior to addition of the base (Barbier conditions) improved the yield to a gratifying 74%. It should be noted, that under these conditions, **4.19** was isolated as a mixture of *cis*/*trans* isomers (predominantly *trans*), whereas complete stereocontrol (only *trans*) was observed when KHMDS was used. Finally, the TBDPS-ether of **4.19** was cleaved and the resulting primary alcohol **4.24** was oxidized as before to give aldehyde **4.25** in 81% yield over two steps.

4.6.3 Completion of the oligoisoprenoid chain

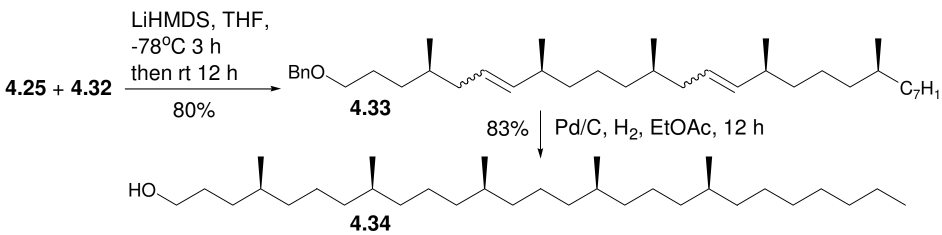
As mentioned in paragraph 4.6.1, we intended to introduce the fifth methyl-branched stereogenic center (fragment **C**) using the asymmetric conjugate addition of Grignard reagents to *α*,*β*-unsaturated thioesters.[33]

Synthesis of a suitable substrate for the key reaction started from 1,4-butanediol, which was mono-protected *via* reaction with benzyl bromide and sodium hydride to give **4.26** in 81% yield (Scheme 4.6). Even though the subsequent oxidation with pyridinium chlorochromate (PCC) proceeded quantitatively, the resulting aldehyde was isolated in a modest 61% yield due to the volatility of **4.27**. Finally, Wittig reaction of **4.27** with $PPh_3CHCOSEt$ afforded *α,β*-unsaturated thioester **4.28** as a mixture of *cis/trans* isomers.[41] Stirring of the mixture in the presence of 4-dimethylaminopyridine (DMAP) shifted the equilibrium towards the desired *trans*-isomer, which was isolated in 87% yield after separation from remaining traces of the *cis*-isomer by column chromatography. Conjugate addition of MeMgBr to **4.28** in the presence of catalytic $CuBr{\cdot}SMe_2$ (5 mol%) and chiral ferrocene-based ligand L* (6 mol%) resulted in the formation of (3R)-**4.29** (92%) with excellent stereoselectivity (93% ee).[33]



Scheme 4.6 *Synthesis of linear chiral fragment C.*

In order to make fragment C suitable for cross-coupling, the thioester of **4.29** was reduced with $LiAlH_4$ to provide the corresponding primary alcohol **4.30** in 96% yield. Subsequent Mitsunobu reaction of **4.30** with 1-*tert*-butyltetrazole-5-thiol (**4.31**, 85%) followed by oxidation with *m*CPBA provided sulfone (3R)-**4.32** (95%). The rationale for the altered substitution moiety on the tetrazole ring in comparison to **4.14**, is that *tert*-butyltetrazolesulfones are known to often give superior yields, probably because of enhanced stability.[42] In our case the improvement was not distinct, as LiHMDS-mediated Julia-Kocienski coupling of sulfone **4.32** to aldehyde **4.25** under Barbier conditions resulted in the formation of oligoisoprenoid **4.33** in 80% yield as a mixture of geometrical isomers (predominantly *trans*; Scheme 4.7). In comparison, coupling of **4.14** to **4.23** under identical conditions gave **4.19** in 74% yield (Scheme 4.5). The synthesis of the oligoisoprenoid chain was completed by simultaneous deprotection of the primary alcohol and hydrogenation of the double bonds *via* exposure to hydrogen over Pd/C to give **4.34** in 83% yield. With the privilege of hindsight, a diimide reduction or hydrogenation over a different catalyst like, for example, Wilkinson catalyst ($(PPh_3P)_3RhCl$) would have been a better choice to obtain the fully saturated system. Pd/C is known to occasionally cause migration of double bonds through reversible hydrometalation, which could lead to partial racemization of the stereogenic centers adjacent to the double bonds in our case.[43] Extensive evaluation of **4.34** by GC-analysis did not indicate that partial racemization/epimerization had indeed taken place.

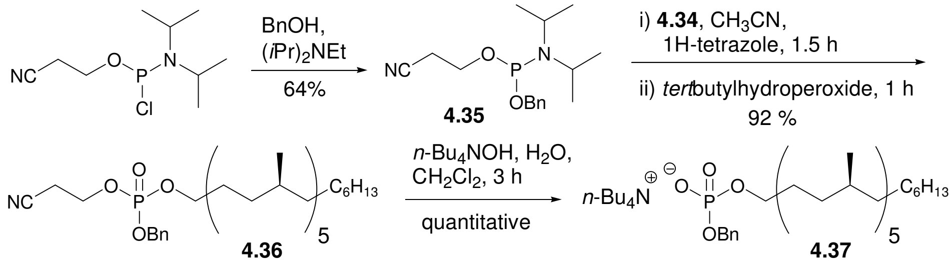


Scheme 4.7 *Completion of the oligoisoprenoid chain **4.34**.*

4.6.4 Formation of the *β*-D-mannosyl phosphate linkage

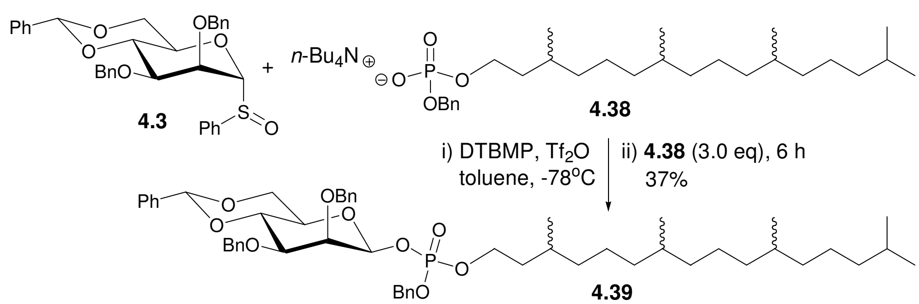
4.6.4.1 *Modification of Crich's β-mannosylation protocol*

Our first attempt to install the *β*-D-mannosyl phosphate linkage, was based on modification of Crich's *β*-mannosylation protocol (Scheme 4.1). We wondered how their approach would perform when a surplus of sulfoxide donor **4.3** was used rather than an excess of phosphate acceptor. To synthesize the appropriate phosphate acceptor, phosphoramidite **4.35** was prepared in 64% yield by reaction of benzyl alcohol with commercial 2-cyanoethyl *N*,*N*-diisopropylchlorophosphoramidite in the presence of Hunig's base (Scheme 4.8).[2] Subsequent tetrazole-mediated substitution of the diisopropylamine moiety of **4.35** by enantiopure saturated oligoisoprenoid **4.34** followed by oxidation of the resulting phosphite with *tert*-butyl hydroperoxide yielded phosphate ester **4.36** in 92%. Finally, treatment of **4.36** with tetra-*n*-butylammonium hydroxide in a dichloromethane/water biphasic system quantitatively afforded phosphate acceptor **4.37**.[2]

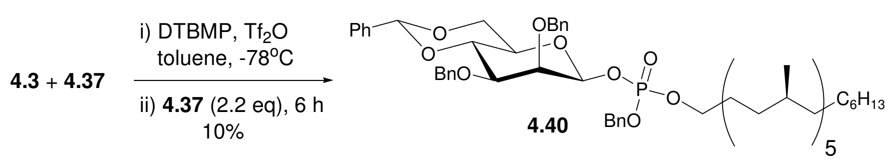


Scheme 4.8 *Synthesis of tetra-n-butylammonium phosphate acceptor **4.37**.*

To find the optimal coupling conditions, sulfoxide **4.3** was first connected to **4.38**, a racemic analogue of **4.37** which was synthesized from phytol (Scheme 4.9). Reproduction of Crich's procedure using three equivalents of **4.38** resulted in the isolation of **4.39** in 37% yield (literature[30g]: 56%). The mass balance was made up mainly by the hydrolysis product of **4.3**, indicating that formation of the triflate was quantitative. When the amount of **4.38** was lowered to 1.5 equivalences, the yield of **4.39** dropped to 16%. Disappointingly, all reactions in which an excess of **4.3** was used failed to provide the desired product at all. The only attempt to couple **4.3** to enantiopure acceptor **4.37** (2.2 eq.) furnished the coupled product **4.40** in an unacceptable 10% yield (Scheme 4.10).



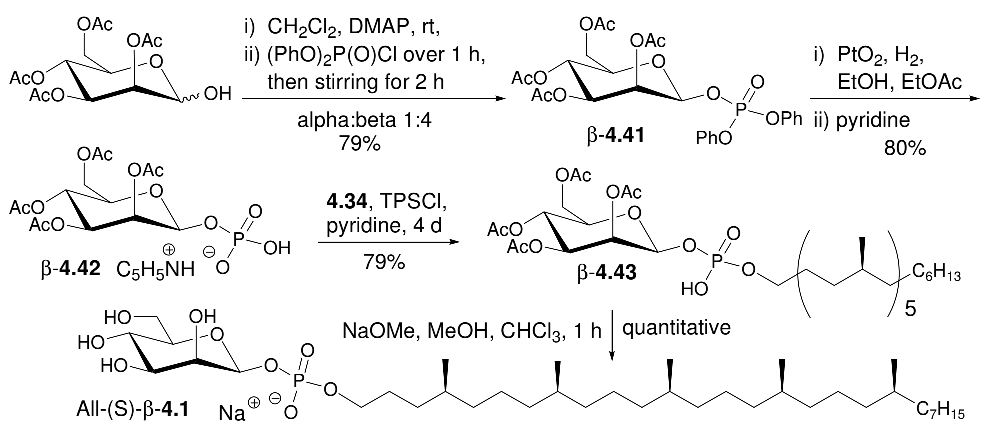
Scheme 4.9 *Model system for the formation of the β-mannosyl phosphate linkage.*



Scheme 4.10 *Formation of the β-mannosyl phosphate linkage using Crich's protocol.*

*4.6.4.2 Reversed strategy; synthesis of a β-mannosyl phosphate ester and coupling to **4.34***

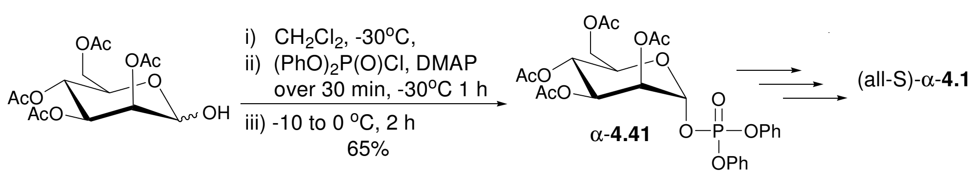
As an alternative approach, we decided to first install a *β*-phosphate moiety on a suitably protected mannopyranose, and then to connect the resulting *β*-mannosyl phosphate ester to the enantiopure oligoisoprenoid alcohol **4.34** (Scheme 4.11). To this end, diphenyl chlorophosphate was slowly added to a solution of 2,3,4,6-tetra-*O*-acetyl-D-mannose in the presence of excess DMAP at room temperature to give predominantly the *β*-anomer of mannosyl phosphate triester **4.41** (79%, α:β = 1:4).[44] The anomers were separated by column chromatography and the *β*-anomer proved to be stable in solution ($CDCl_3$) at room temperature. Remarkably, epimerization to the *α*-anomer did take place when **4.41** was concentrated, but this could be prevented by storage at -18 °C. Removal of the phenyl groups of *β*-**4.41** was achieved by hydrogenation over Adam's catalyst (PtO_2)[44] to give pyridinium salt *β*-**4.42** (80%) after quenching with pyridine. Subsequent coupling of an excess of readily available *β*-**4.42** (2.0 eq) to enantiopure **4.34** (1.0 eq) in the presence of 2,4,6-triisopropylbenzenesulfonyl chloride (TPSCl; 3.0 eq) proceeded readily to give **4.43** in a gratifying 79% yield after four days.[45] The employment of a sterically hindered arenesulfonyl chloride served to prevent sulfonylation of the primary alcohol of **4.34** which could otherwise compete with the desired phosphorylation reaction.[46] Finally, deacetylation with NaOMe in a mixture of chloroform and methanol afforded the target molecule (all-S)-*β*-**4.1** in quantitative yield (see § 4.6.6 for characterization).



Scheme 4.11 *Completion of the synthesis; selective formation of a β-mannosyl phosphate ester followed by coupling to enantiopure alcohol **4.34** and deprotection.*

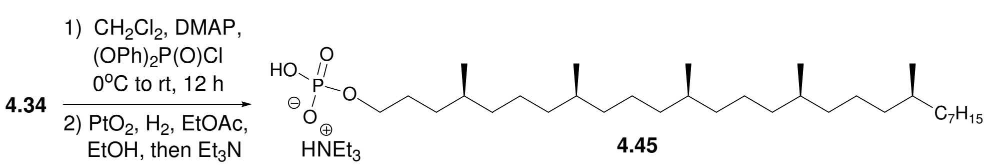
4.6.5 Synthesis of analogues

For analytical and biological comparison, the *α*-anomer of (all-S)-**4.1** was synthesized as well. In the event, α-**4.41** was selectively prepared in 65% isolated yield by simultaneous addition of diphenyl chlorophosphate and DMAP to a solution of 2,3,4,6-tetra-*O*-acetyl-D-mannose at -30 °C (Scheme 4.12).[44] The origin of this remarkable change in the stereochemical outcome of the phosphorylation reaction by a slight modification of the reaction conditions is related to the equilibrium between the two anomers of the starting material and their difference in reactivity. Due to the anomeric effect as well as the axial substituent at the neighboring carbon, the starting mannosylpyranose exists for over 85% as the α-anomer.[44] However, the same effects that cause instability of the *β*-anomer are also grounds for increased reactivity.[47] Hence, reaction in the presence of excess base (DMAP), which catalyzes the anomerization of the *α*- to the less stable *β*-form, mainly gives the kinetic *β*-product at room temperature. In contrast, simultaneous addition of the base and the phosphorylating agent results in the formation of exclusively the thermodynamic *α*-product at -30 °C. Conversion of *α*-**4.41** into (all-S)-*α*-**4.1** was performed in 49% overall yield following the same route as for the preparation of (all-S)-*β*-**4.1**.



Scheme 4.12 *Synthesis of the α-mannosyl phophomycoketide analogue.*

To determine whether the (all-S)-phosphomycoketide, without the mannose headgroup, would show any biological activity, this compound was synthesized in two steps from enantiopure alcohol **4.34**. DMAP-assisted phosphorylation of **4.34** provided diphenyl phosphate triester **4.44**, after which hydrogenation over Adam's catalyst followed by quenching with triethylamine gave phosphate salt **4.45** in quantitative yield (Scheme 4.13).



Scheme 4.13 *Synthesis of a phosphomycoketide analogue.*

4.6.6 Proof of the anomeric configuration

Proof of the anomeric configuration of (all-S)-β-**4.1** commenced with a mass spectrometric investigation employing the same technique as was used in the structural elucidation of the natural product. Indeed, low-energy collisionally induced dissociation tandem mass spectrometry (ESI-CID-MS/MS) of (all-S)-β-**4.1** nicely reproduced the spectrum of the natural product (Figure 4.9).[1] Thus, a cross-ring fragmentation (*m/z* 587.7) of the carbohydrate moiety and a dehydration fragment (*m/z* 689.5) which are both characteristic for *cis*-1,2-glycosyl phosphates were observed.[48] In contrast, (all-S)-α-**4.1** produced a clearly different spectrum with the main fragment (*m/z* 545.5) originating from cleavage of the phosphomycoketide chain at the anomeric center.

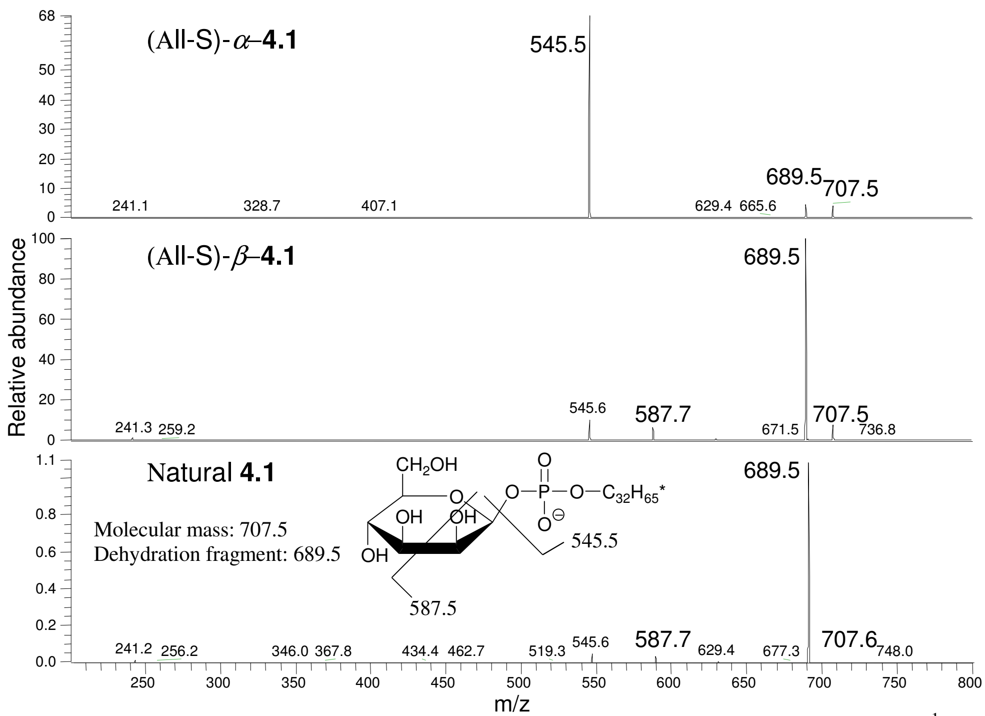


Figure 4.9 *Low energy CID-MS/MS spectra of α-and β-(all-S)-**4.1** and natural **4.1**.*[1]

The anomeric configuration of (all-S)-β-**4.1** was confirmed by the NOE correlation between the anomeric hydrogen and H_3 and H_5 of the mannose moiety (1,3-diaxial interactions). Further evidence was obtained by comparison of the anomeric ${}^1J_{CH}$ coupling constants, which as predicted by literature,[49] amounted to 169 Hz for the *α*-anomer and 159 Hz for the *β*-anomer. A final clue came from the ^{1}H-NMR spectra, which showed a clear difference in the chemical shift of the alpha- (δ = 5.38 ppm) and the beta (δ = 5.05 ppm) anomeric hydrogen (Figure 4.10; anomeric hydrogens depicted in inset).

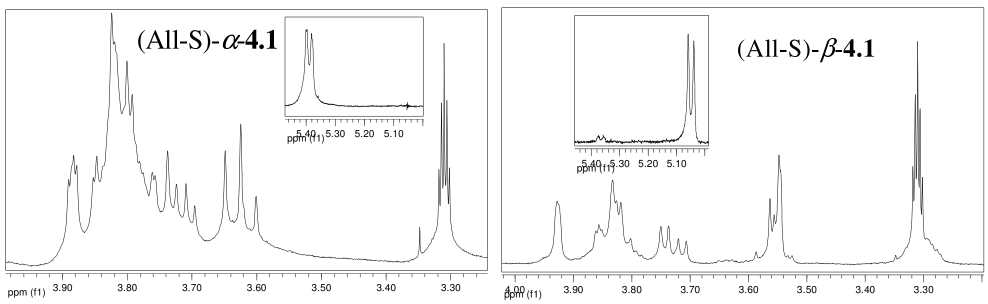


Figure 4.10 *Comparison of part of the ^{1}H-NMR spectra of the two anomers of (all-S)-**4.1**; insert: anomeric protons.*

4.7 Biological activity

In order to evaluate the biological activity of (all-S)-*β*-**4.1**, (all-S)-*α*-**4.1** and (all-S)-**4.45**, the compounds were tested in a dose-dependent manner in T-cell proliferation assays by Dr. Moody and co-workers.[50] CD8-1 T-cells (CD8$^+$TCR) were cultured with CD1c$^+$ antigen presenting cells in the presence or absence of synthetic antigen. As an index of antigen-specific activation, interleukin-2 production was measured, which is secreted by CD8-1 T-cells to stimulate (among others) the proliferation of type 2 helper T-cells (HT-2 cells). In practice, an aliquot of the supernatant was removed from the bio-assay and diluted, after which HT-2 cells were cultured in this medium in the presence of [^{3}H]thymidine. The amount of [^{3}H]thymidine incorporated in DNA was measured with a liquid scintillation counter to give the Interleukin-2 production in counts per minute (CPM).

In agreement with the tri-molecular CD1-lipid-TCR model (§ 4.2.2), phosphomycoketide **4.45**, without the mannose moiety, did not show any activity unless elevated concentrations were used (> 10 μM) in which case activity might be caused by *in situ* mannosylation (Figure 4.11). In addition, comparison of (all-S)-*β*-**4.1** and (all-S)-*α*-**4.1** showed, that a relatively small structural change in the polar epitope already results in a significant decrease in activity. The most important finding, though, was that (all-S)-*β*-**4.1**

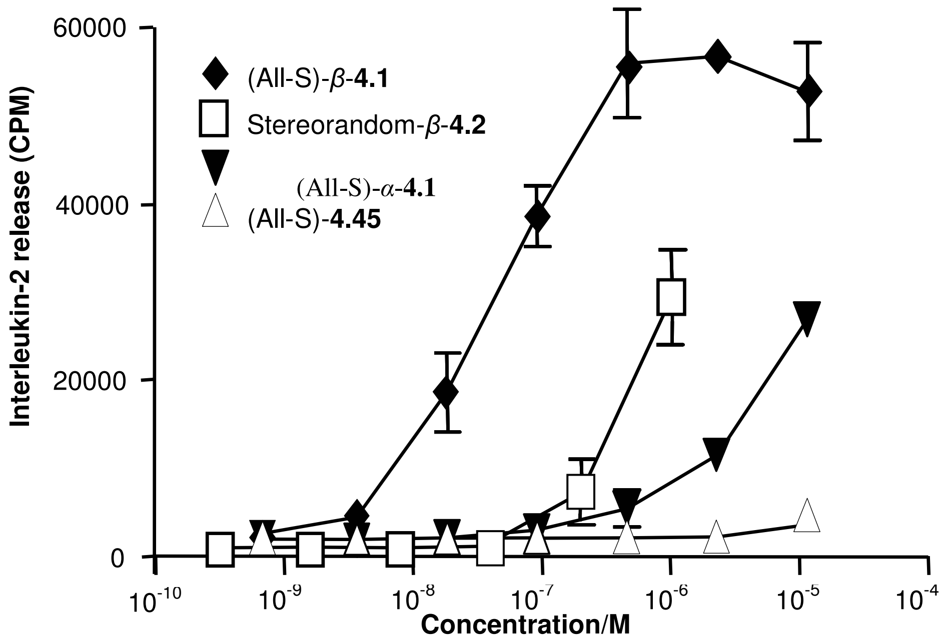


Figure 4.11 *Biological activity of different synthetic MPM's.*

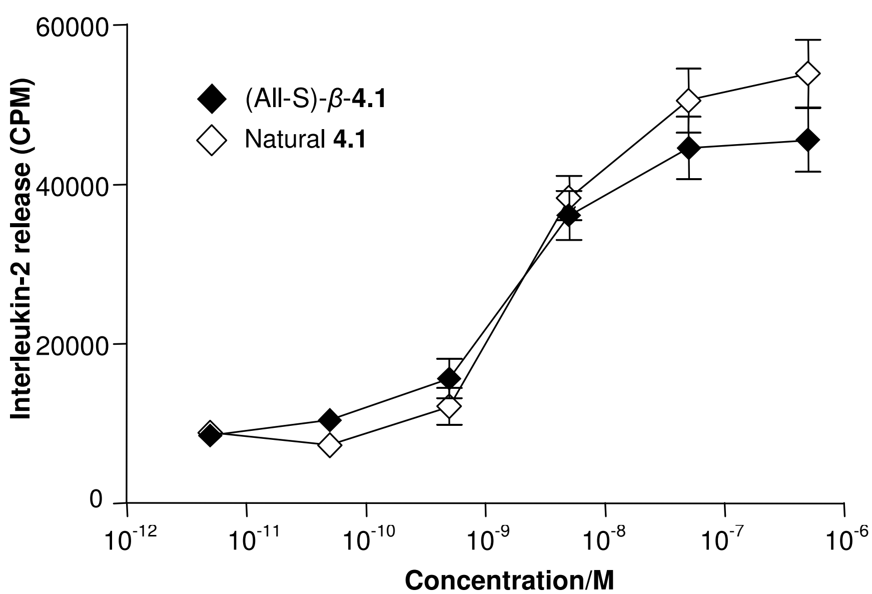


Figure 4.12 *Biological activity of synthetic (all-S)-β-**4.1** compared to the natural product.*

has a considerably higher activity than stereorandom-β-**4.2**. It appears that, contrary to what was assumed so far, (the stereochemistry of) the oligoisoprenoid side-chain does have a pronounced effect on the activity. Finally, assessment of (all-S)-β-**4.1** in comparison to the natural product demonstrated that both compounds have identical activity within the margin of error (Figure 4.12). Obviously, this is an indication that the stereochemistry of the natural product could very well be all-S, although it is certainly no definite proof.

4.8 Summary and concluding remarks

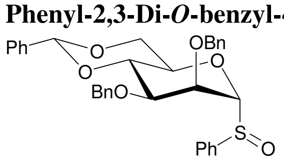
In this chapter the first total synthesis of an enantiopure β-mannosyl phosphomycoketide (MPM) was described. To this end, a general and fully catalytic method for the preparation of enantiopure, saturated oligoisoprenoids of any desired length and stereochemistry was developed. In the event, two different conjugate addition reactions were employed to introduce the stereogenic centers with complete control over the absolute configuration, after which a Julia-Kocienski sequence was used to cross-couple the various linear enantiopure fragments. Using this strategy, penta-methyl-branched aliphatic alcohol **4.34** was prepared, with all stereogenic centers having the S-configuration. Low yields at the final stage of the synthesis due to the difficult nature of the β-mannosyl phosphate linkage were circumvented by installation of the phosphate moiety on a suitably protected mannopyranose prior to coupling to enantiopure **4.34**. Using this convergent approach, the total synthesis of (all-S)-β-**4.1** was achieved in 6.7% overall yield and a longest linear sequence of eighteen steps. In addition, the analogues (all-S)-α-**4.1** as well as (all-S)-**4.45**, which lacks the mannose unit, were prepared following similar routes.

Biological evaluation revealed that (all-S)-β-**4.1** and the natural product have identical activity, whereas an analogue with an oligoisoprenoid chain consisting of a mixture of 32 stereoisomers is significantly less active. The latter result implicates that the fine structure of the saturated alkyl chain influences biological activity to a far greater extent than assumed until now. From a biological point of view, *in vitro* and *in vivo* testing of a library of MPM's with variations in both the length and the stereochemistry of the oligoisoprenoid chain would be very helpful in acquiring detailed knowledge regarding the relationship between structure and activity. Since the synthesis of such a library is a laborious process, an X-ray structure of a CD1c-protein complexed to a β-mannosyl phosphomycoketide antigen would be useful to set certain initial boundaries. At present, attempts are being made to co-crystallize a CD1c-protein and a molecule of synthetic (all-S)-*β*-**4.1**. If succesful, the next objective would be to include a T-cell-receptor as well, which would provide the first direct structural evidence for the existence of the trimolecular complex (Figure 4.3).

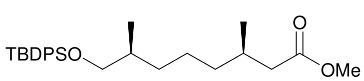
4.9 Experimental section

For general information: see Chapter 2 and 3. HPLC analysis to determine the enantiomeric excess of **4.29** was carried out on a Shimadzu LC-10AD*VP* HPLC equipped with a Shimadzu SPD-M10A*VP* diode array detector. Racemic **4.29** was prepared by reaction of **4.28** with MeMgBr at -78°C in THF in the presence of CuI and TMSCl.

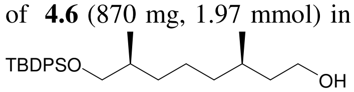
Phenyl-2,3-Di-*O*-benzyl-4,6-*O*-benzylidene-1-deoxy-1-thio-α-D-mannospyranoside S-oxide (4.3):[30f,51c] Sulfoxide donor **4.3** was prepared in six steps from mannose using literature procedures.[30f,51] NMR-data were as reported in the literature.[30f,51c] ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 3.77 (t, *J* = 10.0 Hz, 1H), 4.12 (m, 1H), 4.23 (dd, *J* = 4.8, 10.4 Hz, 1H), 4.28-4.41 (m, 3H), 4.52 (brs, 1H), 4.57 (d, *J* = 11.6 Hz, 1H), 4.61 (d, *J* = 12.0 Hz, 1H), 4.68 (d, *J* = 12.0 Hz, 1H), 4.84 (d, *J* = 12.0 Hz, 1H), 5.65 (s, 1H), 7.21-7.43 (m, 15H), 7.49-7.58 (m, 5H) ppm. MS(CI) for $C_{33}H_{32}O_6S$: m/z = 574 $(M + NH_4)^+$.



8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octanoic acid methyl ester (4.6):[35] (3R,7S)-**3.57** (1.62 g, 8.01 mmol) was dissolved in DMF (12.4 mL) and imidazole (1.09 g, 16.0 mmol, 2.0 eq) and TBDPSCl (4.15 mL, 16.0 mmol, 2.0 eq) were added. The resulting solution was stirred for 30 min under argon at room temperature, after which TLC showed complete conversion. The solution was diluted with water, extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried (Na_2SO_4) and concentrated. **4.6** (3.34 g, 7.58 mmol, 95%) was isolated as a colorless oil after column chromatography (*n*-pentane-Et_2O 9:1). (3R,7S)-**4.6**: $[\alpha]_D^{22}$ = +1.7° (c = 1.97, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.91 (d, *J* = 6.4 Hz, 6H), 1.05 (s, 9H), 1.06-1.45 (m, 6H), 1.63 (m, 1H), 1.93 (m, 1H), 2.09 (dd, *J* = 8.4, 14.8 Hz, 1H), 2.29 (dd, *J* = 6.0, 14.8 Hz, 1H), 3.43 (dd, *J* = 6.4, 9.6 Hz, 1H), 3.50 (dd, *J* = 5.6, 9.6 Hz, 1H), 3.66 (s, 3H), 7.35-7.44 (m, 6H), 7.65-7.68 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.8 (q), 19.1 (s), 19.6 (q), 24.1 (t), 26.8 (q), 30.2 (d), 33.1 (t), 35.5 (d), 36.9 (t), 41.5 (t), 51.2 (q), 68.7 (t), 127.4 (d), 129.4 (d), 134.0 (s), 135.5 (d), 173.5 (s) ppm. MS(CI) for $C_{27}H_{40}O_3Si$: m/z = 458 $(M + NH_4)^+$, HRMS calcd for $C_{27}H_{40}O_3Si$-C_4H_9: 425.251, found: 425.252.

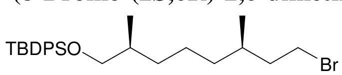


8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octan-1-ol (4.7):[35] To a solution of **4.6** (870 mg, 1.97 mmol) in THF (8.5 mL) at -78 °C was added a solution of Dibal-H



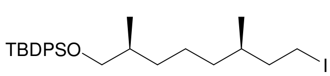
(20 wt% in toluene, 8.3 mL, 10.1 mmol, 5.0 eq) and the resulting mixture was stirred for 2 h at -78 °C under argon, after which TLC showed complete conversion. The reaction was quenched with aq. NH_4Cl (sat.) and then diluted with Et_2O and aq. HCl (1.0 M) until a clear solution was obtained. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $NaHCO_3$ (sat.) and brine (sat.), dried (Na_2SO_4) and concentrated. The product **4.7**, 792 mg, 1.92 mmol, 97%) was isolated as a colorless oil after column chromatography (*n*-pentane-EtOAc 4:1). (3R,7S)-**4.7**: $[\alpha]_D^{22}$ = +0.5° (c = 2.00, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.88 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.4 Hz, 3H), 1.05 (s, 9H), 1.06-1.70 (m, 10H), 3.43 (dd, *J* = 6.4, 9.6 Hz, 1H), 3.51 (dd, *J* = 5.6, 9.6 Hz, 1H), 3.67 (m, 2H), 7.35-7.44 (m, 6H), 7.65-7.68 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.8 (q), 19.2 (s), 19.5 (q), 24.1 (t), 26.7 (q), 29.3 (d), 33.2 (t), 35.5 (d), 37.3 (t), 39.7 (t), 61.0 (t), 68.7 (t), 127.4 (d), 129.3 (d), 133.9 (s), 135.5 (d) ppm. MS(CI) for $C_{26}H_{40}O_2Si$: m/z = 430 $(M + NH_4)^+$, HRMS calcd for $C_{26}H_{40}O_2Si$-C_4H_9: 355.209, found: 355.207.

(8-Bromo-(2S,6R)-2,6-dimethyl-octyloxy)-*tert*-butyl-diphenyl-silane (4.8):[35] To a



solution of **4.7** (188 mg, (0.46 mmol) in dichloromethane (1.6 mL) were added PPh_3 (143 mg, 0.55 mmol, 1.2 eq) and then NBS (97 mg, 0.55 mmol, 1.2 eq) at 0 °C. The resulting solution was stirred under argon at 0 °C for 10 min and then warmed to room temperature over 1 h. The reaction mixture was quenched with aq. $NaHCO_3$ (sat.) and the aqueous layer was extracted with dichloromethane (3x). The combined organic layers were washed with aq. $Na_2S_2O_3$ (10% w/w) and brine (sat.), dried (Na_2SO_4), filtered and concentrated. The resulting brown solid was suspended in *n*-pentane and filtered, after which the residue was washed with *n*-pentane. The filtrate was concentrated and further purified by column chromatography (*n*-pentane-Et_2O 99:1) to give **4.8** (191 mg, 0.40 mmol, 87%) as a colorless oil. (2S,6R)-**4.8**: $[\alpha]_D^{22}$ = -4.1° (c = 1.36, $CHCl_3$),^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.93 (d, *J* = 6.4 Hz, 3H), 1.00 (d, *J* = 6.8 Hz, 3H), 1.14 (s, 9H), 1.10-1.54 (m, 7H), 1.70 (m, 2H), 1.92 (m, 1H), 3.41-3.53 (m, 2H), 3.52 (dd, *J* = 6.4, 10.0 Hz, 1H), 3.59 (dd, *J* = 5.6, 9.6 Hz, 1H), 7.40-7.50 (m, 6H), 7.72-7.77 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 18.8 (q), 19.2 (s), 24.0 (t), 26.8 (q), 31.5 (d), 32.0 (t), 33.2 (t), 35.5 (d), 36.6 (t), 39.9 (t), 68.7 (t), 127.4 (d), 129.4 (d), 133.9 (s), 135.5 (d) ppm. MS(CI) for $C_{26}H_{39}BrOSi$: m/z = 494 $(M + NH_4)^+$. HRMS calcd for $C_{26}H_{39}BrOSi$-C_4H_9: 417.125, found: 417.126.

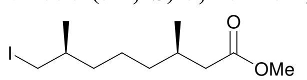
***tert*-Butyl-(8-iodo-(2S,6R)-2,6-dimethyl-octyloxy)-diphenyl-silane (4.9):**[35] To a solution



of PPh_3 (272 mg, 1.04 mmol, 1.2 eq) in dichloromethane (2.5 mL) were added imidazole (70 mg, 1.04 mmol, 1.2 eq) and iodine (262 mg, 1.04 mmol, 1.2 eq) and the resulting mixture was stirred under argon for 5 min. Subsequently, a solution of **4.7** (356 mg, 0.86 mmol) in dichloromethane (1.2 mL) was added and the mixture was stirred for 2 h, after which the reaction was quenched with aq. $NaHCO_3$ (sat.). The aqueous layer was extracted with dichloromethane (3x) and the combined organic layers were washed with aq. $Na_2S_2O_3$

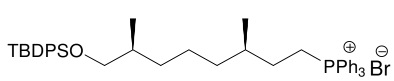
(10% w/w) and brine (sat.), dried (Na_2SO_4), filtered and concentrated. The resulting white solid was suspended in *n*-pentane and filtered, after which the residue was washed with *n*-pentane. The filtrate was concentrated and further purified by column chromatography (*n*-pentane-Et_2O 99:1 to 98:2) to give **4.9** (418 mg, 0.80 mmol, 93%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.86 (d, *J* = 6.4 Hz, 3H), 0.92 (d, *J* = 6.4 Hz, 3H), 1.06 (s, 9H), 1.06-1.67 (m, 9H), 1.86 (m, 1H), 3.17 (m, 1H), 3.24 (m, 1H), 3.44 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.51 (dd, *J* = 5.6, 10.0 Hz, 1H), 7.36-7.45 (m, 6H), 7.65-7.68 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 5.2 (t), 16.9 (q), 18.6 (q), 19.2 (s), 24.0 (t), 26.8 (q), 33.2 (t), 33.7 (d), 35.6 (d), 36.4 (t), 40.8 (t), 68.7 (t), 127.4 (d), 129.4 (d), 134.0 (s), 135.5 (d) ppm. MS(CI) for $C_{26}H_{39}IOSi$: m/z = 540 $(M + NH_4)^+$.

8-Iodo-(3R,7S)-3,7-dimethyl-octanoic acid methyl ester (4.10):[35] To a solution of PPh_3



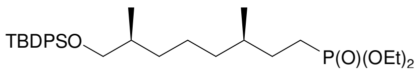
(207 mg, 0.79 mmol, 1.2 eq) in dichloromethane (1.9 mL) were added imidazole (54 mg, 0.79 mmol, 1.2 eq) and iodine (200 mg, 0.79 mmol, 1.2 eq) and the resulting mixture was stirred under argon for 5 min. Subsequently, a solution of (3R,7S)-**3.57** (133 mg, 0.66 mmol) in dichloromethane (0.94 mL) was added and the mixture was stirred for 2 h, after which the reaction was quenched with aq. $NaHCO_3$ (sat.). The aqueous layer was extracted with dichloromethane (3x) and the combined organic layers were washed with aq. $Na_2S_2O_3$ (10% w/w) and brine (sat.), dried (Na_2SO_4), filtered and concentrated. Purification by column chromatography (*n*-pentane-Et_2O 85:15) provided **4.10** (171 mg, 0.55 mmol, 83%) as a colorless liquid. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.90 (d, *J* = 6.4 Hz, 3H), 0.94 (d, *J* = 6.4 Hz, 3H), 1.11-1.45 (m, 7H), 1.92 (m, 1H), 2.09 (dd, *J* = 8.0, 14.8 Hz, 1H), 2.26 (dd, *J* = 6.0, 14.8 Hz, 1H), 3.12 (dd, *J* = 5.6, 9.6 Hz, 1H), 3.19 (dd, *J* = 4.8, 9.6 Hz, 1H), 3.63 (s, 3H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 17.6 (t), 19.5 (q), 20.4 (q), 24.0 (t), 30.0 (d), 34.4 (d), 36.3 (t), 36.4 (t), 41.4 (t), 51.2 (q), 173.4 (s) ppm. MS(CI) for $C_{11}H_{21}IO_2$: m/z = 330 $(M + NH_4)^+$.

[8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octyl]-triphenyl-phosphonium; bromide (4.11):



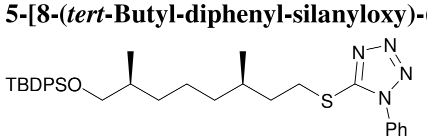
To a solution of **4.8** (197 mg, 0.41 mmol) in acetonitrile (4.0 mL) was added PPh_3 (130 mg, 0.50 mmol, 1.2 eq) and the resulting mixture was heated under reflux overnight. After cooling to room temperature, the reaction mixture was concentrated in vacuo. Purification by column chromatography (first *n*-pentane-Et_2O 98:2 to regain **4.8** and then dichloromethane-MeOH 95:5) furnished **4.11** (259 mg, 0.35 mmol, 85%) as a white solid. (3R,7S)-**4.11**: $[\alpha]_D^{22}$ = -4.4^o (c = 1.13, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.82 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.8 Hz, 3H), 0.99 (s, 9H), 1.00-1.42 (m, 7H), 1.55 (m, 2H), 1.68 (m, 1H), 3.34 (dd, *J* = 6.4, 10.0 Hz, 1H), 3.43 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.59 (m, 2H), 7.28-7.36 (m, 6H), 7.58-7.68 (m, 10H), 7.72-7.81 (m, 9H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.6 (q), 18.9 (q), 19.0 (s), 20.4 + 20.9 (t, J_{C-P} = 50.7 Hz), 23.8 (t), 26.6 (q), 29.0 (t, J_{C-P} = 4.6 Hz), 33.0 (t), 33.3 (d), 35.4 (d), 36.5 (t), 68.6 (t), 117.5 + 118.4 (s, J_{C-P} = 85.9 Hz), 127.3 (d), 129.2 (d), 130.2 (d, *J* = 13.1 Hz), 133.4 (d, J_{C-P} = 10.0 Hz), 133.8 (s), 134.8 (d, *J* = 3.0 Hz), 135.3 (d) ppm.

[8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octyl]-phosphonic acid diethyl ester (4.12):



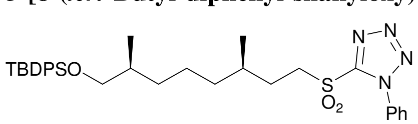
A solution of **4.8** (197 mg, 0.41 mmol) in triethylphosphite (1.0 mL) was heated overnight at 140 ^{o}C while protected from moisture by a $CaCl_2$-tube. The reaction mixture was cooled to room temperature and then concentrated in vacuo. Purification by column chromatography (*n*-pentane-EtOAc 4:1 to 1:1) gave **4.12** (215 mg, 0.40 mmol, 98%) as a colorless oil. (3R,7S)-**4.12**: $[\alpha]_D^{22}$ = +1.7^{o} (c = 1.39, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.84 (d, *J* = 6.4 Hz, 3H), 0.90 (d, *J* = 7.2 Hz, 3H), 1.04 (s, 9H), 1.04-1.44 (m, 7H), 1.30 (t, *J* = 7.2 Hz, 6H), 1.53-1.80 (m, 3H), 3.42 (dd, *J* = 6.4, 10.0 Hz, 1H), 3.49 (dd, *J* = 6.0, 10.0 Hz, 1H), 4.00-4.15 (m, 6H), 7.30-7.42 (m, 6H), 7.63-7.67 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.3 (q, $J_{C\text{-}P}$ = 6.1 Hz), 16.7 (q), 18.9 (q), 19.1 (s), 22.4 + 23.8 (t, $J_{C\text{-}P}$ = 140 Hz), 24.1 (t), 26.7 (q), 28.9 (t, *J* = 5.3 Hz), 33.2 (t), 33.3 (d), 35.5 (d), 36.4 (t), 61.2 (t, $J_{C\text{-}P}$ = 6.1 Hz), 68.7 (t), 127.4 (d), 129.3 (d), 133.9 (s), 135.4 (d) ppm. MS(EI) for $C_{30}H_{49}O_4PSi\text{-}CH_3$: m/z = 517 $[M - CH_3]^+$. HRMS calcd for $C_{30}H_{49}O_4PSi\text{-}CH_3$: 517.290, found: 517.291.

5-[8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octylthio]-1-phenyl-1H-tetrazole (4.13):



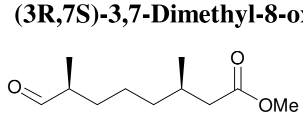
To a solution of **4.7** (450 mg, 1.09 mmol) and 1-phenyl-1H-tetrazole-5-thiol (389 mg, 2.18 mmol, 2.0 eq) in THF (10.0 mL) was added PPh_3 (430 mg, 1.64 mmol, 1.5 eq) at 0 ^{o}C. Subsequently, DIAD (0.38 mL, 1.9 mmol, 1.8 eq) was added dropwise over 2 min and the resulting solution was stirred under argon while warming to room temperature. After 1 h TLC showed complete conversion and the reaction was quenched with brine (sat.). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 9:1) gave **4.13** (571 mg, 1.00 mmol, 91%) as a colorless oil. (3R,7S)-**4.13**: $[\alpha]_D^{22}$ = -4.3^{o} (c = 1.96, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.92 (dd, *J* = 5.6, 6.4 Hz, 6H), 1.06 (s, 9H), 1.06-1.70 (m, 9H), 1.81 (m, 1H), 3.34-3.54 (m, 4H), 7.36-7.44 (m, 6H), 7.51-7.60 (m, 5H), 7.67-7.69 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.8 (q), 19.0 (q), 19.2 (s), 24.0 (t), 26.7 (q), 31.2 (t), 32.0 (d), 33.2 (t), 35.5 (d), 35.8 (t), 36.7 (t), 68.7 (t), 123.7 (d), 127.4 (d), 129.3 (d), 129.6 (d), 129.9 (d), 133.6 (s), 133.9 (s), 135.5 (d) 154.3 (s) ppm. MS(CI) for $C_{33}H_{44}N_4OSSi$: m/z = 573 $(M + H)^+$, 590 $(M + NH_4)^+$, HRMS calcd for $C_{33}H_{44}N_4OSSi\text{-}C_4H_9$: 515.230, found: 515.230.

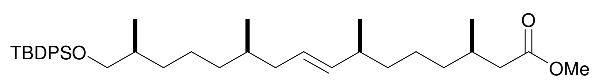
5-[8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octane-1-sulfonyl]-1-phenyl-1H-tetrazole (4.14):



4.13 (570 mg, 0.99 mmol) was dissolved in dichloromethane (5.5 mL) and *m*CPBA (70%, 1.23 g, 4.97 mmol, 5.0 eq) was added at 0 ^{o}C. The resulting suspension was warmed to room temperature and stirred overnight. The reaction was quenched with aq. $Na_2S_2O_3$ (10% w/w) and diluted with dichloromethane. The organic layer was washed with aq. $Na_2S_2O_3$ (10% w/w, 3x; no more peroxides present) and the combined aqueous layers were subsequently extracted with dichloromethane (3x). The combined organic layers were washed with aq. $NaHCO_3$ (sat. 3x) and brine (sat.), dried ($MgSO_4$) and concentrated. The product was purified by column chromatography (*n*-pentane-EtOAc 9:1) to give **4.14** (575 mg, 0.95 mmol, 95%) as a colorless oil. (3R,7S)-**4.14**: $[\alpha]_D^{22}$ = -4.1° (c = 1.74, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.94 (t, *J* = 7.2 Hz, 6H), 1.07 (s, 9H), 1.07-1.48 (m, 6H), 1.64 (m, 2H), 1.77 (m, 1H), 1.96 (m, 1H), 3.49 (m, 2H), 3.74 (m, 2H), 7.37-7.45 (m, 6H), 7.57-7.64 (m, 3H), 7.67-7.72 (m, 6H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.7 (q), 18.9 (q), 19.2 (s), 23.9 (t), 26.7 (q), 28.2 (t), 31.8 (d), 33.1 (t), 35.5 (d), 36.4 (t), 54.1 (t), 68.6 (t), 124.9 (d), 127.4 (d), 129.4 (d), 129.5 (d), 131.3 (d), 132.9 (s), 133.9 (s), 135.5 (d) 153.3 (s) ppm. MS(CI) for $C_{33}H_{44}N_4O_3SSi$: m/z = 622 $(M + NH_4)^+$, HRMS calcd for $C_{33}H_{44}N_4O_3SSi$-C_4H_9: 547.220, found: 547.223.

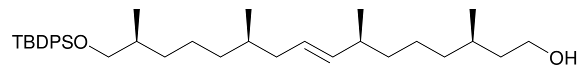
(3R,7S)-3,7-Dimethyl-8-oxo-octanoic acid methyl ester (4.15): (3R,7S)-**3.57** (50 mg, 0.25 mmol) was dissolved in dichloromethane (2.3 mL) and NMO-oxide (43 mg, 0.37 mmol, 1.5 eq) and TPAP (17 mg, 49 μmol, 0.20 eq) were added. The resulting black solution was stirred for 30 min under argon, after which TLC showed conversion into a single product. The reaction mixture was brought directly onto a silica column and flushed with a mixture of *n*-pentane-EtOAc (95:5 to 9:1) to give pure **4.15** (48 mg, 0.24 mmol, 97%) as a colorless liquid after concentration. Because the product was particularly sensitive to oxidation, the pressure after concentration in vacuo was equalized with nitrogen instead of air and **6** was immediately used in the next step. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.91 (d, *J* = 6.4 Hz, 3H), 1.08 (d, *J* = 6.8 Hz, 3H), 1.12-1.40 (m, 5H), 1.69 (m, 1H), 1.94 (m, 1H), 2.11 (dd, *J* = 8.0, 14.8 Hz, 1H), 2.28 (dd, *J* = 6.0, 14.8 Hz, 1H), 2.32 (m, 1H), 3.65 (s, 3H), 9.59 (d, *J* = 2.0 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 13.2 (q), 19.5 (q), 24.1 (t), 30.0 (d), 30.4 (t), 36.4 (t), 41.4 (t), 46.1 (d), 51.3 (q), 173.5 (s) 205.1 (d) ppm.



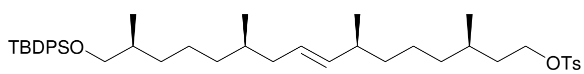


16-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S,11R,15S)-3,7,11,15-tetramethyl-hexadec-8-enoic acid methyl ester (4.16): The experiment using sulfon **4.14** is described; procedures with Wittig reagent **4.11** and HWE-reagent **4.12** were performed in an analogous fashion. KHMDS (0.5 M in toluene, 0.69 mL, 0.35 mmol, 1.0 eq) was added in a dropwise fashion to a solution of **4.14** (196 mg, 0.32 mmol, 1.0 eq) in THF (4.0 mL) at -78 °C resulting in a bright yellow solution. This mixture was stirred for 30 min under argon, after which **4.15** (73 mg, 0.36 mmol, 1.1 eq) in THF (4.0 mL) was added over 10 min. The solution was stirred for 3 h at -78 °C and then stirred overnight while slowly warming to room temperature. The reaction was quenched with water and the aqueous layer was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane:Et_2O 98:2) gave **4.16** (106 mg, 0.18 mmol, 57%) as a colorless oil. In addition to **4.16**, several side products were observed, but due to the complexity of the mixture their structure could not be elucidated. (3R,7S,11R,15S)-**4.16**: $[\alpha]_D^{22}$ = +9.4° (c = 1.70, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 6.8 Hz, 3H), 0.92 (m, 6H), 0.95 (d, *J* = 6.8 Hz, 3H), 1.06 (s, 9H), 1.07-1.46 (m, 13H), 1.64 (m, 1H), 1.79 (m, 1H), 1.88-2.10 (m, 3H), 2.10 (dd, *J* = 8.0, 14.4 Hz, 1H), 2.30 (dd, *J* = 6.0, 14.4 Hz, 1H), 3.44 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.52 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.66 (s, 3H), 5.21 (dd, *J* = 7.6, 15.2 Hz, 1H), 5.31 (m, 1H), 7.36-7.44 (m, 6H), 7.66-7.69 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 19.2 (s), 19.3 (q), 19.6 (q), 21.0

(q), 24.3 (t), 24.5 (t), 26.8 (q), 30.2 (d), 33.1 (d), 33.3 (t), 35.6 (d), 36.6 (t), 36.6 (d), 36.7 (t), 37.0 (t), 39.8 (t), 41.6 (t), 51.2 (q), 68.8 (t), 126.9 (d), 127.4 (d), 129.3 (d), 134.0 (s), 135.5 (d), 137.4 (d), 173.7 (s) ppm. MS(CI) for $C_{37}H_{58}O_3Si$: m/z = 597 $(M + NH_4)^+$, HRMS calcd for $C_{37}H_{58}O_3Si$-C_4H_9: 521.345, found: 521.346.

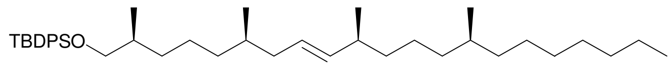


16-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S,11R,15S)-3,7,11,15-tetramethyl-hexadec-8-en-1-ol (4.17): To a solution of **4.16** (100 mg, 0.173 mmol) in THF (6.0 mL) at -78 °C was added a solution of Dibal-H (20 wt% in toluene, 0.71 mL, 0.86 mmol, 5.0 eq) and the resulting mixture was stirred for 2 h at -78 °C under argon, after which TLC showed complete conversion. The reaction was quenched with aq. NH_4Cl (sat.) and then diluted with Et_2O and aq. HCl (1.0 M) until a clear solution was obtained. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $NaHCO_3$ (sat.) and brine (sat.), dried (Na_2SO_4) and concentrated. The product (**4.17**, 93 mg, 0.169 mmol, 98%) was isolated as a colorless oil after column chromatography (*n*-pentane:EtOAc 4:1). (3R,7S,11R,15S)-**4.17**: $[\alpha]_D^{22}$ = +7.9° (c = 1.12, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 6.8 Hz, 3H), 0.88 (d, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.4 Hz, 3H), 0.95 (d, *J* = 6.8 Hz, 3H), 1.06 (s, 9H), 1.07-1.69 (m, 17H), 1.80 (m, 1H), 1.98 (m, 1H), 2.06 (m, 1H), 3.41 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.51 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.67 (m, 2H), 5.22 (dd, *J* = 7.6, 15.2 Hz, 1H), 5.32 (m, 1H), 7.36-7.44 (m, 6H), 7.66-7.68 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 19.2 (s), 19.4 (q), 19.5 (q), 21.0 (q), 24.3 (t), 24.5 (t), 26.8 (q), 29.3 (d), 33.1 (d), 33.3 (t), 35.6 (d), 36.69 (t), 36.71 (d), 37.0 (t), 37.3 (t), 39.87 (t), 39.90 (t), 61.1 (t), 68.8 (t), 126.8 (d), 127.4 (d), 129.3 (d), 134.0 (s), 135.5 (d), 137.5 (d) ppm. MS(CI) for $C_{36}H_{58}O_2Si$: m/z = 568 $(M + NH_4)^+$, HRMS calcd for $C_{36}H_{58}O_2Si$-C_4H_9: 493.350, found: 493.349.



16-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S,11R,15S)-3,7,11,15-tetramethyl-hexadec-8-enyl *p*-toluenesulfonate (4.18): 4.17 (90 mg, 0.16 mmol) was dissolved in dry pyridine (0.62 mL) and and *p*-TsCl (47 mg, 0.25 mmol, 1.5 eq) was added at 0 °C. The resulting solution was stirred at 0 °C under argon overnight and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat.), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated to give **4.18** (99 mg, 0.14 mmol, 86%) as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 95:5). (3R,7S,11R,15S)-**4.18**: $[\alpha]_D^{22}$ = +7.9° (c = 1.46, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.78 (d, *J* = 6.4 Hz, 3H), 0.82 (d, *J* = 6.4 Hz, 3H), 0.92 (t, *J* = 6.8 Hz, 6H), 1.05 (s, 9H), 1.06-1.70 (m, 17H), 1.78 (m, 1H), 2.00 (m, 2H), 2.45 (s, 3H), 3.43 (dd, *J* = 6.4, 9.6 Hz, 1H), 3.51 (dd, *J* = 5.2, 9.6 Hz, 1H), 4.05 (m, 2H), 5.20 (dd, *J* = 7.6, 15.2 Hz, 1H), 5.30 (m, 1H), 7.33-7.44 (m, 8H), 7.66-7.68 (dd, *J* = 1.6, 7.6 Hz, 4H), 7.79 (d, *J* = 8.4 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.9 (q), 19.0 (q), 19.2 (s), 19.3 (q), 20.9 (q), 21.5 (q), 24.3 (t), 26.8 (q), 29.0 (d), 33.0 (d), 33.3 (t), 35.60 (d), 35.63 (t), 36.5 (t), 36.6 (d), 36.7 (t), 37.1 (t), 39.8 (t), 68.8 (t), 69.0 (t), 126.9 (d), 127.4 (d), 127.8 (d), 129.3 (d), 129.7 (d), 133.1 (s), 134.0 (s), 135.5 (d), 137.4 (d), 144.5 (s) ppm.

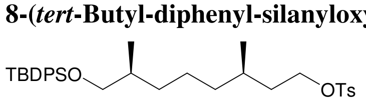
MS(CI) for $C_{43}H_{64}O_4SSi$: m/z = 722 $(M + NH_4)^+$, HRMS calcd for $C_{43}H_{64}O_4SSi$-C_4H_9: 647.359, found: 647.360.



***tert*-Butyl-diphenyl-((2S,6R,10S,14S)-2,6,10,14-tetramethyl-henicos-8-enyloxy)-silane (4.19):** *n*-Pentyl bromide (0.50 mL, 4.0 mmol) in dry THF (9.5 mL) was added dropwise to Mg (147 mg, 6.05 mmol, 1.5 eq) in an argon-purged flask and then stirred for 1 h at 45 ^oC. The resulting solution was cooled to room temperature and the precipitate was allowed to settle overnight. Titration with *sec*-butanol in the presence of 1,10-phenanthroline showed the concentration of $C_5H_{11}MgBr$ to be 0.43 M. The Grignard reagent (1.72 mL, 0.740 mmol, 4.3 eq) was added in a dropwise fashion to a solution of **4.18** (120 mg, 0.171 mmol) and CuBr·SMe_2 (9.5 mg, 46 μmol, 27 mol%) in dry THF (1.6 mL) at -78 ^oC under argon. After stirring for 1 h at -78 ^oC, the solution was allowed to warm to 0 ^oC and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. **4.19** (101 mg, 0.167 mmol, 98%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane). (2S,6R,10S,14S)-**4.19**: $[\alpha]_D^{22}$ = +6.0^o (c = 1.51, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.82-0.97 (m, 15H), 1.06 (s, 9H), 1.07 (m, 4H), 1.14-1.46 (m, 22H), 1.65 (m, 1H), 1.80 (m, 1H), 1.94-2.12 (m, 2H), 3.43 (dd, *J* = 6.0, 10.0 Hz, 1H), 3.51 (dd, *J* = 5.6, 10.0 Hz, 1H), 5.20-5.36 (m, 2H), 7.36-7.45 (m, 6H), 7.66-7.70 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 16.9 (q), 19.2 (s), 19.4 (q), 19.6 (q), 21.0 (q), 22.6 (t), 24.3 (t), 24.7 (t), 26.8 (q), 27.0 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.6 (d), 33.1 (d), 33.3 (t), 35.6 (d), 36.70 (t), 36.72 (d), 37.0 (t), 37.4 (t), 39.9 (t), 68.8 (t), 126.7 (d), 127.4 (d), 129.3 (d), 134.0 (s), 135.5 (d), 137.7 (d) ppm. MS(CI) for $C_{41}H_{68}OSi$: m/z = 623 $(M + NH_4)^+$, HRMS calcd for $C_{41}H_{68}OSi$-C_4H_9: 547.434, found: 547.434.

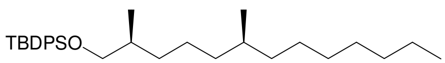
***tert*-Butyl-diphenyl-((2S,6R,10S,14S)-2,6,10,14-tetramethyl-henicos-8-enyloxy)-silane (4.19) from 4.14 and 4.23:** To a solution of **4.14** (964 mg, 1.59 mmol) and **4.23** (415 mg, 1.83 mmol, 1.15 eq) in THF (19 mL) at -78 ^oC was added LiHMDS (1.0 M in THF, 1.67 mL, 1.67 mmol, 1.05 eq) in a dropwise fashion over 5 min. The resulting yellow solution was stirred at -78 ^oC for 3 h and then allowed to warm to room temperature overnight. The reaction was quenched with aq. NH_4Cl (sat.) and the aqueous layer was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane) gave **4.19** (713 mg, 1.18 mmol, 74%) as a colorless oil. Analytical data are shown above.

8-(*tert*-Butyl-diphenyl-silanyloxy)-(3R,7S)-3,7-dimethyl-octyl *p*-toluenesulfonate

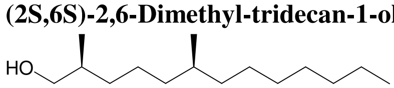


(4.20): 4.7 (1.96 g, 4.76 mmol) was dissolved in dry pyridine (17.7 mL) and *p*-TsCl (1.36 g, 7.13 mmol, 1.5 eq) was added at 0 ^oC. The resulting solution was stirred at 0 ^oC under argon for 36 h and then quenched with water. The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with aq. $CuSO_4$ (sat. 2x), aq. $NaHCO_3$ (sat.) and brine (sat.), dried ($MgSO_4$) and concentrated to give **4.20** (2.33 g, 4.11 mmol, 86%) as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 95:5). (3R,7S)-**4.20**: $[\alpha]_D^{22}$ = -1.0 ^{o} (c = 1.27, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.79 (d, *J* = 6.4 Hz, 3H), 0.90 (d, *J* = 6.8 Hz, 3H), 1.06 (s, 9H), 1.06-1.52 (m, 8H), 1.64 (m, 2H) 2.44 (s, 3H), 3.43 (dd, *J* = 6.4, 10.0 Hz, 1H), 3.50 (dd, *J* = 5.6, 10.0 Hz, 1H), 4.06 (m, 2H), 7.32-7.44 (m, 8H), 7.66-7.68 (m, 4H), 7.80 (d, *J* = 8.4 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 16.8 (q), 19.0 (q), 19.2 (s), 21.5 (q), 24.0 (t), 26.8 (q), 29.1 (d), 33.2 (t), 35.5 (d), 35.6 (t), 36.8 (t), 68.7 (t) 69.0 (t), 127.4 (d), 127.8 (d), 129.4 (d), 129.7 (d), 133.1 (s), 134.0 (s), 135.5 (d), 144.5 (s) ppm. MS(CI) for $C_{33}H_{46}O_4SSi$: m/z = 584 $(M + NH_4)^+$, HRMS calcd for $C_{33}H_{46}O_4SSi$-C_4H_9: 509.218, found: 509.219.

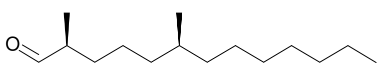
***tert*-Butyl-((2S,6S)-2,6-dimethyl-tridecyloxy)-diphenyl-silane (4.21):** Pentyl bromide (2.50 mL, 20.2 mmol) in dry THF (47.5 mL) was added dropwise to Mg (735 mg, 30.2 mmol, 1.5 eq) in an argon-purged flask and then stirred for 1 h at 45 °C. The resulting solution was cooled to room temperature and the precipitate was allowed to settle overnight. Titration with *sec*-butanol in the presence of 1,10-phenanthroline showed the concentration of $C_5H_{11}MgBr$ to be 0.39 M. The Grignard reagent (41.7 mL, 16.3 mmol, 4.0 eq) was added over 40 min to a solution of **4.20** (2.30 g, 4.06 mmol) and $CuBr{\cdot}SMe_2$ (209 mg, 1.02 mmol, 25 mol%) in dry THF (38 mL) at -78 °C under argon. After stirring for 2 h at -78 °C, the solution was allowed to warm to 0 °C and stirred overnight. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. **4.21** (1.84 g, 3.94 mmol, 97%) was isolated as a colorless oil after purification by column chromatography (*n*-pentane). (2S,6S)-**4.21**: $[\alpha]_D^{20}$ = -0.8° (c = 1.27, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.86 (d, *J* = 6.8 Hz, 3H), 0.92 (t, *J* = 7.2 Hz, 3H), 0.96 (d, *J* = 6.8 Hz, 3H), 1.09 (s, 9H), 1.06-1.52 (m, 19H), 1.67 (m, 1H) 3.47 (dd, *J* = 6.4, 9.6 Hz, 1H), 3.50 (dd, *J* = 5.6, 9.6 Hz, 1H), 7.38-7.46 (m, 6H), 7.70-7.72 (m, 4H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 16.9 (q), 19.2 (s), 19.6 (q), 22.6 (t), 24.3 (t), 26.8 (q), 27.0 (t), 29.3 (t), 29.9 (t), 31.9 (t), 32.6 (d), 33.4 (t), 35.6 (d), 37.0 (t), 37.3 (t), 68.8 (t), 127.4 (d), 129.4 (d), 134.1 (s), 135.5 (d) ppm. MS(CI) for $C_{31}H_{50}OSi$: m/z = 484 $(M + NH_4)^+$, HRMS calcd for $C_{31}H_{50}OSi$-C_4H_9: 409.293, found: 409.294.

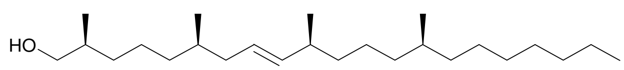


(2S,6S)-2,6-Dimethyl-tridecan-1-ol (4.22): TBAF (1.0 M in THF, 15.7 mL, 15.7 mmol, 4.0 eq) was added to a solution of **4.21** (1.83 g, 3.92 mmol) in THF (26.5 mL) and the resulting mixture was stirred for 2 h at room temperature, after which TLC showed complete conversion. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Because **4.22** was difficult to separate from the TBDPS-waste products, the crude product was used in the next step. An analytical sample was purified by column chromatography (benzene:Et_2O 95:5 and then a second column using *n*-pentane-Et_2O 9:1 to 4:1) to give a colorless oil. (2S,6S)-**4.22**: $[\alpha]_D^{22}$ = -8.0° (c = 1.05, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.84 (d, *J* = 6.4 Hz, 3H), 0.87 (t, *J* = 6.8 Hz, 3H), 0.91 (d, *J* = 6.8 Hz, 3H), 1.07 (m, 3H), 1.18-1.40 (m, 16H), 1.46 (brs, 1H), 1.61 (m, 1H), 3.40 (dd, *J* = 6.8, 10.4 Hz, 1H), 3.50 (dd, *J* = 5.6, 10.4 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 16.5 (q), 19.6 (q), 22.6 (t), 24.3 (t), 27.0 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.6 (d), 33.4 (t), 35.7 (d), 36.9 (t), 37.2 (t), 68.3 (t) ppm. MS(EI) for $C_{15}H_{32}O$: m/z = 210 $[M^+]$, HRMS calcd for $C_{15}H_{32}O$: 210.235, found: 210.235.

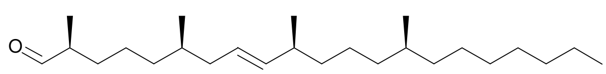


(2S,6S)-2,6-Dimethyl-tridecanal (4.23): Crude **4.22** (3.92 mmol) was dissolved in dry dichloromethane (37 mL) in the presence of crushed molsieves (4Å) and stirred for 20 min. Subsequently, NMO-oxide (668 mg, 5.70 mmol, 1.50 eq) and TPAP (80 mg, 0.23 mmol, 0.06 eq) were added. The resulting black solution was stirred for 45 min, after which TLC showed conversion into a single product. The reaction mixture was filtered over a silica plug (dichloromethane) and concentrated. After column chromatography (*n*-pentane-EtO_2 98:2) **4.23** (639 mg, 2.82 mmol, 72% from **4.21**) was obtained as a colorless liquid. Because the product was sensitive to oxidation, the pressure after concentration in vacuo was equalized with nitrogen instead of air and (2S,6S)-**4.23** was immediately used in the next step. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.84 (d, *J* = 6.4 Hz, 3H), 0.88 (t, *J* = 6.8 Hz, 3H), 1.07 (m, 2H), 1.09 (d, *J* = 6.8 Hz, 3H), 1.18-1.40 (m, 16H), 1.69 (m, 1H), 2.34 (ddd, *J* = 2.0, 6.8, 13.6 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 50.32 MHz) δ = 13.3 (q), 14.0 (q), 19.5 (q), 22.6 (t), 24.3 (t), 26.9 (t), 29.3 (t), 29.8 (t), 30.8 (t), 31.8 (t), 32.5 (d), 33.8 (t), 36.9 (t), 46.3 (d), 205.2 (d) ppm. MS(CI) for $C_{15}H_{30}O$+O (oxidized to carboxylic acid): m/z = 260 $(M + NH_4)^+$.



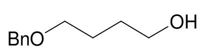


(2S,6R,10S,14S)-2,6,10,14-Tetramethyl-henicos-8-en-1-ol (4.24): TBAF (1.0 M in THF, 6.3 mL, 6.3 mmol, 4.0 eq) was added to a solution of **4.23** (958 mg, 1.58 mmol) in THF (10.6 mL) and the resulting mixture was stirred for 2 h, after which TLC showed complete conversion. The reaction was quenched with aq. NH_4Cl (sat.), extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Because **4.24** was difficult to separate from the TBDPS-waste products, the crude product was used in the next step. An analytical sample was purified by column chromatography (benzene-Et_2O 95:5) to give a colorless oil. (2S,6R,10S,14S)-**4.24**: $[\alpha]_D^{22}$ = +6.1^o (c = 1.06, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 7.2 Hz, 3H), 0.84 (d, *J* = 7.2 Hz, 3H), 0.88 (t, *J* = 6.8 Hz, 3H), 0.92 (d, *J* = 6.8 Hz, 3H), 0.95 (d, *J* = 6.4 Hz, 3H), 1.06 (m, 4H), 1.14-1.48 (m, 23H), 1.61 (m, 1H), 1.81 (m, 1H), 1.98 (m, 1H), 2.05 (m, 1H), 3.41 (dd, *J* = 6.4, 10.0 Hz, 1H), 3.51 (dd, *J* = 5.6, 10.0 Hz, 1H), 5.22 (dd, *J* = 7.2, 15.2 Hz, 1H), 5.31 (m, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 16.5 (q), 19.4 (q), 19.6 (q), 21.0 (q), 22.6 (t), 24.3 (t), 24.7 (t), 27.0 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.7 (d), 33.1 (d), 33.3 (t), 35.7 (d), 36.65 (t), 36.72 (d), 37.0 (t), 37.4 (t), 39.9 (t), 68.3 (t), 126.6 (d), 137.8 (d) ppm. MS(EI) for $C_{25}H_{50}O$: m/z = 366 [M^+], HRMS calcd for $C_{25}H_{50}O$: 366.386, found: 366.385.



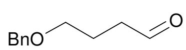
(2S,6R,10S,14S)-2,6,10,14-Tetramethyl-henicos-8-enal (4.25): Crude **4.24** (1.58 mmol) was dissolved in dry dichloromethane (15.4 mL) in the presence of crushed molsieves (4Å) and stirred for 20 min. Subsequently, NMO-oxide (278 mg, 2.37 mmol, 1.50 eq) and TPAP (33 mg, 0.10 mmol, 0.06 eq) were added. The resulting black solution was stirred for 30 min, after which TLC showed conversion into a single product. The reaction mixture was filtered over a silica plug (dichloromethane) and concentrated. After column chromatography (*n*-pentane-EtO_2 98:2) **4.25** (468 mg, 1.28 mmol, 81% from **4.23**) was obtained as a colorless oil. Because the product was sensitive to oxidation, the pressure after concentration in vacuo was equalized with nitrogen instead of air and **4.25** was immediately used in the next step. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.82 (d, J = 6.4 Hz, 3H), 0.84 (d, J = 6.8 Hz, 3H), 0.88 (t, J = 6.8 Hz, 3H), 0.95 (d, J = 6.4 Hz, 3H), 1.09 (d, J = 6.8 Hz, 3H), 1.09 (m, 4H), 1.16-1.50 (m, 21H), 1.69 (m, 1H), 1.82 (m, 1H), 1.97 (m, 1H), 2.05 (m, 1H), 2.33 (m, 1H), 5.20-5.34 (m, 2H), 9.61 (d, J = 2.0 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 13.2 (q), 14.0 (q), 19.3 (q), 19.6 (q), 21.0 (q), 22.6 (t), 24.3 (t), 24.6 (t), 27.0 (t), 29.3 (t), 29.9 (t), 30.7 (t), 31.8 (t), 32.6 (d), 32.9 (d), 36.3 (t), 36.7 (d), 37.0 (t), 37.4 (t), 39.8 (t), 46.2 (d), 126.4 (d), 137.9 (d) 205.3 (d) ppm. MS(EI) for $C_{25}H_{48}O$: m/z = 364 [M^+], HRMS calcd for $C_{25}H_{48}O$: 364.371, found: 364.373.

4-Benzyloxy-butan-1-ol (4.26): To a solution of butane-1,4-diol (4.95 mL, 55.5 mmol) in



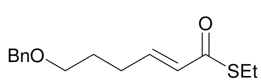
dry THF (36 mL) was added NaH (50%, 2.66 g, 55.5 mmol) at 0 °C. Subsequently, BnBr (6.60 mL, 55.5 mmol) in THF (12 mL) was added to the mixture and a small amount of DMF to increase solubility. The resulting suspension was stirred overnight under argon at room temperature, after which the reaction was quenched with aq. NH_4Cl (sat.). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. The product was purified by column chromatography (*n*-pentane-EtOAc 4:1 to 7:3) to give **4.26** (8.10 g, 44.9 mmol, 81%) as a light yellow oil. NMR-data were as reported in the literature.[52]

3-Benzyloxy-propionaldehyde (4.27):[52] To a solution of PCC (5.3 g, 24 mmol, 2.2 eq) in



dichloromethane (38 mL) was added a solution of **4.26** (2.0 g, 11 mmol) in dichloromethane (6.8 mL). The resulting dark green mixture was stirred under argon overnight and then filtered over a silica plug (dichloromethane). After concentration, **4.27** (1.2 g, 6.7 mmol, 61%) was obtained as a colorless liquid, which was used without further purification. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.95 (m, 2H), 2.54 (dt, J = 1.6, 6.8, 14.4 Hz, 2H), 3.50 (t, J = 6.4 Hz, 2H), 4.49 (s, 2H), 7.24-7.37 (m, 5H), 9.77 (t, J = 1.6 Hz, 1H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 22.3 (t), 40.7 (t), 68.9 (t), 72.7 (t), 127.4 (d), 128.1 (d), 138.0 (s), 202.0 (d) ppm.

S-Ethyl (2E)-6-(benzyloxy)hex-2-enethioate (4.28): Preparation of the Wittig reagent:[41]

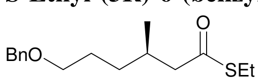


Bromoacetic acid (13.9 g, 100 mmol), ethanethiol (9.65 mL, 130 mmol) and DMAP (1.22 g, 10.0 mmol) were dissolved in dichloromethane (450 mL) and cooled to 0 °C. To the resulting mixture was added DCC (21.7 g, 105 mmol) in three equal portions, after which the solution was warmed to room temperature and stirred overnight under argon. The resulting white suspension was filtered over celite and the residue was washed with dichloromethane (3x). The filtrate was partially concentrated in vacuo and then washed with aq. $NaHCO_3$ (sat.), water and brine (sat.). The organic layer was dried (Na_2SO_4), filtered and concentrated to give a yellow oil, which was dissolved in benzene (150 mL). To this solution was added PPh_3 (26.3 g, 100 mmol), after which the mixture was left to crystallize for 4 days. The crystals were filtered off and washed with cold toluene, after which they were dissolved in dichloromethane. The solution was washed with aq. Na_2CO_3 (10% w/w) and the aqueous layer was extracted with dichloromethane (2x). The combined organic layers were dried ($MgSO_4$), filtered and partially concentrated. The product was crystallized from a mixture of dichloromethane and *n*-pentane to give EtSC(O)C=PPh_3 (25 g, 69 mmol, 69%) as white/colorless crystals. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.25 (t, *J* = 7.6 Hz, 3H), 2.84 (q, *J* = 7.6 Hz, 2H), 3.66 (d, *J* = 21.6 Hz, 1H), 7.44-7.65 (m, 15H) ppm.

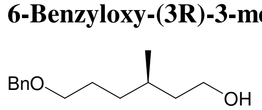
EtSC(O)C=PPh_3 (1.21 g, 3.33 mmol, 1.1 eq) was added to a solution of **4.27** (540 mg, 3.03 mmol) in chloroform (35 mL) and the resulting mixture was refluxed under argon for 3 h. The reaction mixture was cooled to room temperature and then stirred overnight in the presence of DMAP (100 mg, 0.82 mmol), after which the solvent was removed in vacuo. *Trans*-**4.28** (more polar, 697 mg, 2.64 mmol, 87%) and a small amount of *cis*-**4.28** (less polar) were obtained as colorless oils after purification by column chromatography (*n*-pentane-EtOAc 95:5 to 9:1). ^{1}H-NMR *trans*-**4.28** ($CDCl_3$, 400 MHz) δ = 1.28 (t, *J* = 7.6 Hz, 3H), 1.78 (m, 2H), 2.31 (m, 2H), 2.94 (q, *J* = 7.6 Hz, 2H), 3.49 (t, *J* = 6.4 Hz, 2H), 4.50 (s, 2H), 6.11 (d, *J* = 15.2 Hz, 1H), 6.90 (dt, *J* = 7.2, 15.2 Hz, 1H) 7.26-7.37 (m, 5H) ppm. ^{13}C-NMR *trans*-**4.28** ($CDCl_3$, 100.6 MHz) δ = 14.6 (q), 22.9 (t), 27.9 (t), 28.7 (t), 69.0 (t), 72.8 (t), 127.42 (d), 127.44 (d), 128.2 (d), 128.8 (d), 138.2 (s), 144.3 (d) 189.9 (s) ppm. MS(EI) for $C_{15}H_{20}O_2S$-C_2H_5: m/z = 235 [M^+].

^{1}H-NMR *cis*-**4.28** ($CDCl_3$, 400 MHz) δ = 1.28 (t, *J* = 7.6 Hz, 3H), 1.78 (m, 2H), 2.74 (m, 2H), 2.92 (q, *J* = 7.6 Hz, 2H), 3.50 (t, *J* = 6.4 Hz, 2H), 4.50 (s, 2H), 5.98-6.08 (m, 2H), 7.26-7.36 (m, 5H) ppm. ^{13}C-NMR *cis*-**4.28** ($CDCl_3$, 100.6 MHz) δ = 14.6 (q), 23.1 (t), 26.7 (t), 29.1 (t), 69.6 (t), 72.8 (t), 126.5 (d), 127.4 (d), 127.5 (d), 128.2 (d), 138.3 (s), 146.0 (d) 189.7 (s) ppm. MS(EI) for $C_{15}H_{20}O_2S$-C_2H_5S: m/z = 203 [M^+].

S-Ethyl (3R)-6-(benzyloxy)-3-methylhexanethioate (4.29):[33] (R,S)-Josiphos (L*, 67 mg, 0.11 mmol, 6 mol%) and CuBr·SMe_2 (19 mg, 0.094 mmol, 5 mol%) were dissolved in *t*BuOMe (17 mL) and stirred for 30 min under argon at room temperature. The mixture was cooled to -78 °C and MeMgBr (3.0 M in Et_2O, 0.78 mL, 2.3 mmol, 1.3 eq) was added dropwise over 2 min. After stirring for 10 min, a solution of *trans*-**4.28** (495 mg, 1.87 mmol) in *t*BuOMe (1.9 mL) was added over 5 h by syringe pump and the resulting mixture was stirred overnight at -78 °C. The reaction was quenched with MeOH at -78 °C, removed from the cold bath and diluted with H_2O, aq. NH_4Cl (sat.) and Et_2O. The product was extracted with Et_2O (3x), the combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. (3R)-**4.29** (485 mg, 1.73 mmol, 92%, 93% ee) was isolated as a colorless liquid after purification by column chromatography (*n*-pentane-EtOAc 95:5). (3R)-**4.29**: $[\alpha]_D^{22}$ = +5.1° (c = 1.88, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.94 (d, *J* = 6.8 Hz, 3H), 1.24 (t, *J* = 7.6 Hz, 3H), 1.22-1.31 (m, 1H), 1.41 (m, 1H), 1.63 (m, 2H), 1.94 (m, 1H), 2.36 (dd, *J* = 8.4, 14.4 Hz, 1H), 2.54 (dd, *J* = 5.6, 14.4 Hz, 1H), 2.87 (q, *J* = 7.2 Hz, 2H), 3.45 (t, *J* = 6.4 Hz, 2H), 4.50 (s, 2H), 7.26-7.37 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.7 (q), 19.3 (q) 23.2 (t), 27.0 (t), 30.8 (d), 32.9 (t), 51.2 (t), 70.3 (t), 72.8 (t), 127.4 (d), 127.5 (d), 128.2 (d), 138.4 (s), 199.1 (s) ppm. MS(EI) for $C_{16}H_{24}O_2S$: m/z = 280 [M^+], HRMS calcd for $C_{16}H_{24}O_2S$: 280.150, found: 280.149. Ee determination by chiral HPLC analysis, chiralcel OD-H column, *n*-heptane-*i*PrOH 99.5:0.5, retention times: 20.77 (R) / 22.19 (S) min.

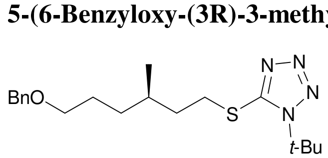


6-Benzyloxy-(3R)-3-methyl-hexan-1-ol (4.30): 4.29 (555 mg, 1.98 mmol) was dissolved



in THF (6.0 mL) and $LiAlH_4$ (188 mg, 4.95 mmol, 2.5 eq) was added at 0 °C. The resulting suspension was stirred for 45 min at 0 °C under argon and then quenched with water. NaOH (3 M aq.) was added and the mixture was stirred until two clear layers were obtained. The layers were separated and the aqueous layer was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 7:3) gave **4.30** (421 mg, 1.89 mmol, 96%) as a colorless liquid. (3R)-**4.30**: $[\alpha]_D^{22}$ = +4.9° (c = 1.10, $CHCl_3$), Lit. $[\alpha]_D$[53] = +5.6°, ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.90 (d, *J* = 6.8 Hz, 3H), 1.22 (m, 1H), 1.40 (m, 2H), 1.54-1.73 (m, 3H), 3.46 (t, *J* = 6.4 Hz, 2H), 3.67 (m, 2H), 4.50 (s, 2H), 7.26-7.37 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 19.4 (q), 27.0 (t), 29.2 (d), 33.3 (t), 39.7 (t), 61.0 (t), 70.6 (t), 72.8 (t), 127.4 (d), 127.5 (d), 128.2 (d), 138.5 (s) ppm. MS(EI) for $C_{14}H_{22}O_2$: m/z = 222 [M^+], HRMS calcd for $C_{14}H_{22}O_2$: 222.162, found: 222.164.

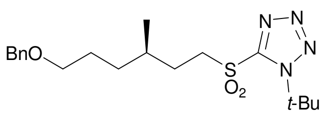
5-(6-Benzyloxy-(3R)-3-methyl-hexylthio)-1-*tert*-butyl-1H-tetrazole (4.31): Preparation



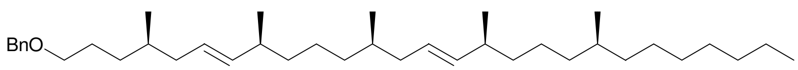
of 1-*tert*-butyl-1H-tetrazole-5-thiol:[54] Sodium azide (5.64 g, 86.8 mmol) was dissolved in water (27 mL) and heated under reflux, after which a solution of *tert*-butylisothiocyanate (10.0 g, 86.8 mmol) in *i*-PrOH (21 mL) was added over 2 h. The resulting mixture was heated under reflux overnight and then cooled to 0 °C. Subsequently, concentrated aq. HCl (37%, 13 mL) was added and the solution was concentrated to 25-30 mL upon which a solid appeared. The suspension was stored at 0 °C for 5 h, after which the solid was filtered off. The residue was washed with ice-water and then crystallized from cyclohexane. The crystals were dried in vacuo at 50 °C for 5 h, after which recrystallization from cyclohexane afforded 1-*tert*-butyl-1H-tetrazole-5-thiol (8.85 g, 55.9 mmol, 64.4%) as a mixture of yellow and white crystals.

To a solution of PPh_3 (779 mg, 2.96 mmol, 1.5 eq) and **4.30** (440 mg, 1.98 mmol) in THF (12.0 mL) at 0 °C was added DIAD (0.68 mL, 3.5 mmol, 1.8 eq) over 2 min resulting in a yellow suspension. Subsequently, a solution of 1-*tert*-butyl-1H-tetrazole-5-thiol (626 mg, 3.96 mmol, 2.0 eq) in THF (6.0 mL) was added over 5 min and the reaction mixture was warmed to room temperature. After 1 h TLC showed complete conversion and the reaction was quenched with brine (sat.). The aqueous layer was extracted with Et_2O (3x) and the combined organic layers were dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 9:1) gave **4.31** (610 mg, 1.68 mmol, 85%) as a colorless oil. (3R)-**4.31**: $[\alpha]_D^{20}$ = -3.7° (c = 1.18, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.95 (d, *J* = 6.0 Hz, 3H), 1.24 (m, 1H), 1.43 (m, 1H), 1.54-1.70 (m, 3H), 1.71 (s, 9H), 1.71-1.86 (m, 2H), 3.31-3.50 (m, 2H), 3.45 (t, *J* = 6.4 Hz, 2H), 4.49 (s, 2H), 7.24-7.36 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 50.32 MHz) δ = 19.0 (q), 27.0 (t), 28.5 (q), 31.8 (t), 32.1 (d), 32.8 (t), 35.8 (t), 60.7 (s), 70.4 (t), 72.8 (t), 127.3 (d), 127.5 (d), 128.2 (d), 138.4 (s), 152.6 (s) ppm. MS(EI) for $C_{19}H_{30}N_4OS$ m/z = 362 [M^+], HRMS calcd for $C_{19}H_{30}N_4OS$: 362.214, found: 362.213.

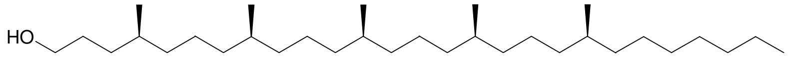
5-(6-Benzyloxy-(3R)-3-methyl-hexane-1-sulfonyl)-1-*tert*-butyl-1H-tetrazole (4.32): 4.31



(610 mg, 1.68 mmol) was dissolved in dichloromethane (18.0 mL) and *m*CPBA (70%, 2.44 g, 9.90 mmol, 5.9 eq) was added at 0 °C. The resulting suspension was warmed to room temperature and stirred overnight. The reaction was quenched with aq. $Na_2S_2O_3$ (10% w/w) and diluted with dichloromethane. The organic layer was washed with aq. $Na_2S_2O_3$ (10% w/w, 3x; no more peroxides present) and the combined aqueous layers were subsequently extracted with dichloromethane (3x). The combined organic layers were washed with aq. $NaHCO_3$ (sat. 3x) and brine (sat.), dried ($MgSO_4$) and concentrated. The product was purified by column chromatography (*n*-pentane-EtOAc 9:1) to give **4.32** (631 mg, 1.60 mmol, 95%) as a colorless oil. (3R)-**4.32**: $[\alpha]_D^{20}$ = -3.8° (c = 1.00, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.99 (d, *J* = 6.8 Hz, 3H), 1.30 (m, 1H), 1.47 (m, 1H), 1.55-1.73 (m, 3H), 1.80 (m, 1H), 1.85 (s, 9H), 1.99 (m, 1H), 3.47 (t, *J* = 6.4 Hz, 2H), 3.77 (ddd, *J* = 5.2, 10.8, 14.4 Hz, 1H), 3.84 (ddd, *J* = 5.2, 11.2, 14.4 Hz, 1H), 4.50 (s, 2H), 7.26-7.37 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 18.9 (q), 26.9 (t), 28.4 (t), 29.5 (q), 31.8 (d), 32.6 (t), 54.8 (t), 65.2 (s), 70.1 (t), 72.8 (t), 127.4 (d), 127.5 (d), 128.2 (d), 138.3 (s), 153.8 (s) ppm. MS(EI) for $C_{19}H_{30}N_4O_3S$ m/z = 394 [M^+], HRMS calcd for $C_{19}H_{30}N_4O_3S$-C_4H_9: 337.133, found: 337.133.



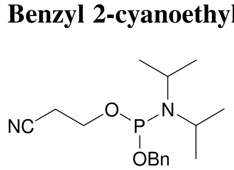
((4R,8S,12R,16S,20S)-4,8,12,16,20-Pentamethyl-heptacosa-6,14-dienyloxymethyl)-benzene (4.33): To a solution of **4.25** (455 mg, 1.25 mmol) and **4.32** (608 mg, 1.54 mmol, 1.24 eq) in THF (19 mL) at -78 °C was added LiHMDS (1.0 M in THF, 1.41 mL, 1.41 mmol, 1.13 eq) in a dropwise fashion over 5 min. The resulting yellow solution was stirred at -78 °C for 3 h and then allowed to warm to room temperature overnight. The reaction was quenched with aq. NH_4Cl (sat.) and the aqueous layer was extracted with Et_2O (3x). The combined organic layers were washed with brine (sat.), dried ($MgSO_4$) and concentrated. Purification by column chromatography (*n*-pentane-Et_2O 98:2) gave **4.33** (549 mg, 0.99 mmol, 80%) as a colorless oil. (4R,8S,12R,16S,20S)-**4.33**: $[\alpha]_D^{22}$ = +15.0° (c = 0.55, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.83 (d, *J* = 6.4 Hz, 6H), 0.87 (d, *J* = 6.4 Hz, 3H), 0.89 (t, *J* = 6.8 Hz, 3H), 0.95 (d, *J* = 6.8 Hz, 6H), 1.00-1.50 (m, 29H), 1.63 (m, 2H), 1.81 (m, 2H), 1.93-2.09 (m, 4H), 3.45 (t, *J* = 6.8 Hz, 2H), 4.51 (s, 2H), 5.19-5.35 (m, 4H), 7.25-7.37 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 19.3 (q), 19.4 (q), 19.6 (q), 20.9 (q), 21.0 (q), 22.6 (t), 24.66 (t), 24.68 (t), 27.0 (t), 27.2 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.6 (d), 32.7 (t), 33.0 (d), 33.1 (d), 36.5 (t), 36.69 (d), 36.73 (d), 37.03 (t), 37.04 (t), 37.3 (t), 37.4 (t), 39.9 (t), 40.0 (t), 70.8 (t), 72.8 (t), 126.5 (d), 126.8 (d), 127.3 (d), 127.5 (d), 128.2 (d), 137.7 (d), 137.8 (d), 138.6 (s) ppm. MS(EI) for $C_{39}H_{68}O$ m/z = 552 [M^+], HRMS calcd for $C_{39}H_{68}O$: 552.527, found: 552.528.

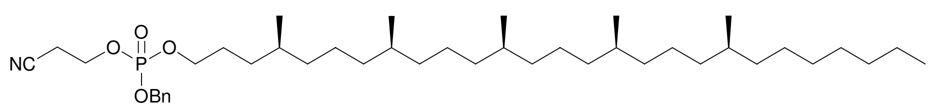


(4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethyl-heptacosan-1-ol (4.34): 4.33 (549 mg, 0.99 mmol) was dissolved in EtOAc (50 mL) and Pd/C (10%, 750 mg) was added. The resulting suspension was degassed with 3 vacuum-argon cycles and then saturated with H_2

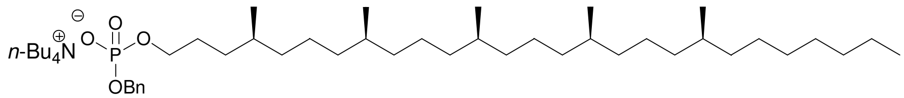
by 5 vacuum-H_2 cycles. After stirring overnight under an H_2-atmosphere (1 bar), TLC showed complete conversion. The Pd/C was removed by filtration over a Florisil-plug and the resulting clear solution was concentrated in vacuo. After purification by column chromatography (*n*-pentane-EtOAc 9:1 to 9:2) **4.34** (381 mg, 0.82 mmol, 83%) was isolated as a colorless oil. (4S,8S,12S,16S,20S)-**4.34**: $[\alpha]_D^{22} = -0.8^o$ (c = 1.16, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.84-0.90 (m, 18H), 1.00-1.42 (m, 44H), 1.57 (m, 2H), 3.63 (t, *J* = 6.8 Hz, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 19.6 (q), 19.7 (q), 22.6 (t), 24.4 (t), 27.0 (t), 29.3 (t), 29.9 (t), 30.3 (t), 31.8 (t), 32.5 (d), 32.7 (d), 32.8 (t), 37.0 (t), 37.3 (t), 63.4 (t) ppm. MS(EI) for $C_{32}H_{66}O$-H_2O m/z = 448 [M^+], MS(CI) for $C_{32}H_{66}O$: m/z = 484 $(M + NH_4)^+$, HRMS calcd for $C_{32}H_{66}O$-H_2O: 448.501, found: 448.501. GC-characterization: HP1-kolom, 30 m x 0.25 mm, He-flow = 1.0 mL/min, T_i = 70 ^{o}C for 70 min, T_f = 300 ^{o}C, rate 10 ^{o}C/min, rt. 79.8 min (1 peak only).

Benzyl 2-cyanoethyl *N,N*-Diisopropylphosphoramidite (4.35):[2] Hunig's base (EtN(*i*Pr)$_2$, 1.47 mL, 8.44 mmol) was added to benzyl alcohol (0.44 mL, 4.27 mmol) and after stirring for 5 min, 2-cyanoethyl *N,N*-diisopropylchlorophosphoramidite (1.0 g, 4.23 mmol) was added. The resulting mixture was stirred for 1 h at room temperature and was then diluted with dichloromethane and aq. $NaHCO_3$ (sat.). The aqueous layer was extracted with dichloromethane (2x) and the combined organic layers were dried (Na_2SO_4), filtered and concentrated. Purification by column chromatography (*n*-pentane-Et_2O-Et_3N 80:20:1) furnished **4.35** (829 mg, 2.69 mmol, 64%) as a colorless liquid. NMR-data were as reported in the literature:[2] ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.20 (t, *J* = 7.2 Hz, 12H), 2.62 (t, *J* = 6.4 Hz, 2H), 3.66 (m, 2H), 3.85 (m, 2H), 4.67 (dd, *J* = 8.8, 12.4 Hz, 1H), 4.76 (dd, *J* = 8.4, 12.4 Hz, 1H), 7.25-7.36 (m, 5H) ppm.

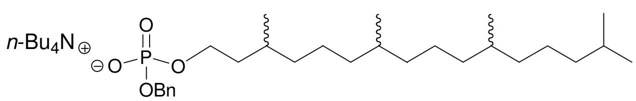




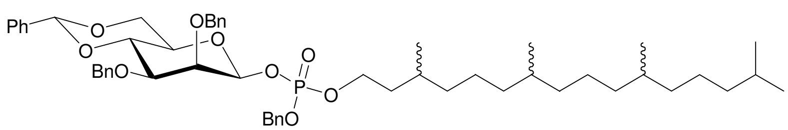
Benzyl-2-cyanoethyl-(4S,8S,12S,16S,20S)-4,8,12,16,20-pentamethyl-heptacosylphosphate (4.36): To an emulsion of **4.34** (76 mg, 0.16 mmol) and **4.35** (60 mg, 0.20 mmol, 1.2 eq) in acetonitrile (0.83 mL) was added ^{1}H-tetrazole (34 mg, 0.49 mmol, 3.0 eq) and the resulting mixture was stirred for 1.5 h. Subsequently, *tert*-butylhydroperoxide (5.0-6.0 M in decane, 0.16 mL, 0.80-0.96 mmol, 5.0-6.0 eq) was added in a dropwise fashion and stirring was continued for 1 h. The resulting clear solution was concentrated in vacuo and **4.36** (104 mg, 0.15 mmol, 92%) was obtained as a colorless oil after purification by column chromatography (*n*-pentane-EtOAc 4:1 to 2:1 to 1:1). (4S,8S,12S,16S,20S)-**4.36**: $[\alpha]_D^{22} = -0.2^o$ (c = 1.03, $CHCl_3$), ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.81-0.90 (m, 18H), 1.00-1.42 (m, 43H), 1.65 (m, 2H), 2.66 (dt, *J* = 2.8, 6.4 Hz, 2H), 3.03 (q, *J* = 6.8 Hz, 2H), 4.15 (m, 2H), 5.10 (d, *J* = 9.2 Hz, 2H), 7.35-7.40 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 19.3-19.6 (q + t), 22.6 (t), 24.4 (t), 27.0 (t), 27.6 (t), 27.7 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.3 (d), 32.4 (t), 32.6-32.7 (d), 36.9-37.3 (t), 61.4 (t, J_{C-P} = 4.6 Hz), 68.8 (t, *J* = 6.9 Hz), 69.6 (t, J_{C-P} = 5.3 Hz), 128.0 (d), 128.6 (d), 128.7 (d), 135.4 (s) ppm.



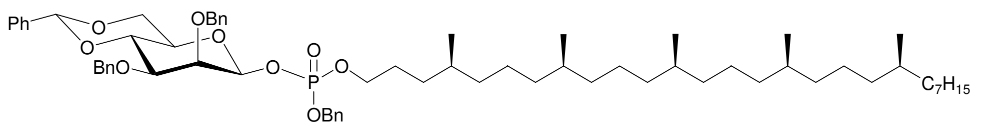
Tetrabutylammonium-benzyl-(4S,8S,12S,16S,20S)-4,8,12,16,20-pentamethyl-heptacosylphosphate (4.37): To a solution of **4.36** (96 mg, 0.14 mmol) in dichloromethane (0.56 mL) was added a solution of tetra-*n*-butylammonium hydroxide (54 mg, 0.21 mmol, 1.5 eq) in water (0.56 mL) and the resulting mixture was stirred vigorously for 1 h. The reaction mixture was diluted with dichloromethane and water and the aqueous layer was extracted with dichloromethane (2x). The combined organic layers were dried (Na_2SO_4), filtered and concentrated to give **4.37** (122 mg, quantitative) as a yellow oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.76-0.86 (m, 18H), 0.93 (t, *J* =7.2 Hz, 12H), 0.93-1.42 (m, 51H), 1.59 (m, 10H), 3.30 (m, 8H), 3.79 (q, *J* = 6.4 Hz, 2H), 4.91 (d, *J* = 6.0 Hz, 2H), 7.14-7.40 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 13.5 (q), 14.0 (q), 19.3-19.6 (q + t), 22.5 (t), 23.9 (t), 24.3 (t), 26.9 (t), 28.4 (t), 28.5 (t), 29.2 (t), 29.8 (t), 31.7 (t), 32.58 (d), 32.61 (d), 33.0 (t), 33.1 (t), 36.9-37.4 (t), 58.6 (t), 65.3 (t), 66.4 (t), 126.5 (d), 127.1 (d), 127.7 (d) ppm.



Tetra-*n*-butylammonium-benzyl-3,7,11,15-tetramethyl-hexadecylphosphate (4.38):[2,30g] Prepared as described for the synthesis of **4.37**. The alcohol was obtained from commercial phytol *via* hydrogenation over PtO_2.[30g] NMR-data were as reported in the literature: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.80-0.86 (m, 15H), 0.97 (t, *J* =7.2 Hz, 12H), 0.95-1.43 (m, 28H), 1.51 (m, 2H), 1.62 (m, 10H), 3.33 (m, 8H), 3.86 (m, 2H), 4.93 (d, *J* = 6.4 Hz, 2H), 7.14-7.45 (m, 5H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 13.6, 19.5-19.6, 22.5, 22.6, 24.0, 24.3, 24.4, 24.7, 27.9, 29.5 32.7, 37.1-37.4, 39.2, 58.7, 63.3, 66.5, 126.6, 127.2, 127.7 ppm.

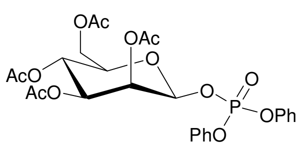


Benzyl 3,7,11,15-tetramethyl-hexadecylphosphoryl 2,3-di-*O*-benzyl-4,6-benzylidene-*β*-D-mannopyranoside (4.39):[2,30g] To a solution of **4.3** (152 mg, 0.27 mmol) and 2,6-di-*tert*-butyl-4-methylpyridine (DTBMP, 56 mg, 0.27 mmol) in toluene (11 mL) at -78 °C was added Tf_2O (55 µl, 0.33 mmol, 1.2 eq) in a dropwise fashion. The resulting mixture was stirred for 15 min under argon, after which a solution of **4.38** (581 mg, 0.82 mmol, 3.0 eq) in toluene (2.7 mL) was added. The mixture was stirred at -78 °C for 6 h and then quenched at this temperature with aq. $NaHCO_3$ (sat.). After warming to room temperature, the aqueous layer was extracted with EtOAc (2x) and the combined organic layers were washed with brine (sat.), dried ($MgSO_4$), filtered and concentrated. Because **4.39** was difficult to separate from the hydrolysis product of **4.3**, the crude product was dissolved in pyridine (1.8 mL), after which Ac_2O (0.2 mL) was added. The mixture was stirred overnight and then concentrated in vacuo. The remaining oil was dissolved in EtOAc and washed with aq. $NaHCO_3$ (sat.), aq. $CuSO_4$ (sat.) and brine (sat.). The organic layer was dried ($MgSO_4$), filtered and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 4:1 to 2:1) gave **4.39** (72 mg, 0.10 mmol, 37%, mixture of two diastereomers at P) as a yellow oil. NMR-data were as reported in the literature: ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.78-0.90 (m, 15H), 0.99-1.50 (m, 21H), 1.51 (m, 2H), 1.68 (m, 1H), 3.40 (m, 1H), 3.63 (2dd, *J* = 2.8, 10.0 Hz for both isomers, 1H), 3.81-4.15 (m, 4H), 4.20 (t, *J* = 9.6 Hz, 1H), 4.29 (dd, *J* = 4.8, 10.4 Hz, 1H), 4.61 + 4.62 (2d, isomer 1 *J* = 12.4 Hz, isomer 2 *J* = 12.0 Hz, 1H), 4.73 (d, *J* = 12.4 Hz, 1H), 4.77 (s, 1H), 4.89 (m, 1H), 5.05 (d, *J* = 8.0 Hz, 1H), 5.08 (m, 1H), 5.24 (brt, *J* = 6.8 Hz, 1H), 5.59 + 5.60 (2s, one for each isomer, 1H), 7.26-7.54 (m, 20H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 19.1, 19.2, 19.5, 19.6, 22.5, 22.6, 24.2, 24.4, 24.7, 27.8, 29.15, 29.18, 29.6, 32.7, 36.8-37.4, 39.2, 66.7, 66.77, 66.84, 66.9, 67.8, 68.0, 68.1, 69.2, 69.3, 69.38, 69.44, 69.9, 72.6, 75.1, 75.2, 76.3, 76.4, 77.4, 77.5, 77.8-77.9, 96.8, 96.9, 101.4, 125.9, 127.4-128.8, 128.9, 131.5, 137.2, 137.9 ppm.



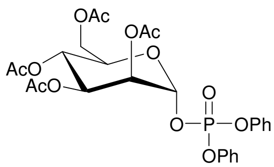
Benzyl (4S,8S,12S,16S,20S)-4,8,12,16,20-pentamethyl-heptacosylphosphoryl 2,3-di-*O*-benzyl-4,6-benzylidene-*β*-D-mannopyranoside (4.40): Sulfon **4.3** (36 mg, 64 µmol) was coupled to phosphate salt **4.37** (127 mg, 0.14 mmol, 2.3 eq) using DTBMP (13 mg, 64 µmol) and Tf_2O (12 µl, 71 µmol, 1.1 eq) in toluene (2.5 + 0.5 mL) to yield **4.40** (7.0 mg, 6.6 µmol, 10%, mixture of two diastereomers at P). The reaction was performed as described for the preparation of **4.39**. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.74-0.90 (m, 18H), 0.96-1.44 (m, 43H), 1.64 (m, 2H), 3.34-3.51 (m, 1H), 3.63 (m, 1H), 3.70-4.41 (m, 6H), 4.61 (m, 1H), 4.70 (m, 1H), 4.80 (m, 1H), 4.90 (m, 1H), 5.07 (m, 2H), 5.25 (m, 1H), 5.63 (s, 1H), 7.23-7.54 (m, 20H) ppm.

Diphenyl-(2,3,4,6-tetra-*O*-acetyl-*β*-D-mannopyranosyl)-phosphate (*β*-4.41):[44] To a solution of 2,3,4,6-tetra-*O*-acetyl-D-mannopyranose (1.90 g, 5.46 mmol) and DMAP (1.52 g, 12.4 mmol, 2.28 eq) in dichloromethane (32 mL) at room temperature was added a solution of diphenyl chlorophosphate (2.66 mL, 12.8 mmol, 2.35 eq) in dichloromethane (13 mL) by syringe pump over 1 h. After stirring the resulting solution under argon for 2 h, the reaction mixture was diluted with dichloromethane and washed with cold water, cold aq. HCl (0.5 M) and cold aq. $NaHCO_3$ (sat.). The organic layer was dried ($MgSO_4$), filtered and concentrated to give **4.41** (2.50 g, 4.31 mmol, 79%) as a mixture of anomers (ratio α:β = 1:4). The pure *β*-anomer of **4.41** (0.98 g, 1.69 mmol, 31%) was obtained after purification by column chromatography (*n*-pentane-EtOAc 8:3 to 3:2) together with a mixed fraction of anomers (1.40 g, 2.41 mmol, 44%). *β*-**4.41** was stable in solution at room temperature (even in chloroform) and for several months at -18°C when concentrated. However, when stored at room temperature after concentration, we observed that the fraction of *α*-anomer increased either by epimerization or by selective degradation of the *β*-anomer. ^{1}H-NMR ***β*-4.41** ($CDCl_3$, 400 MHz) δ = 1.94 (s, 3H, Ac), 2.00 (s, 6H, Ac), 2.05 (s, 3H, Ac), 3.77 (m, 1H, C5-H), 4.07 (dd, *J* = 2.4, 12.4 Hz, 1H, C6-H), 4.23 (dd, *J* = 5.6, 12.4 Hz, 1H, C6-H'),

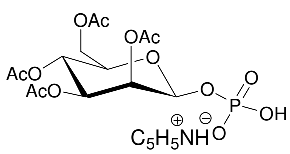


5.07 (dd, J = 3.6, 9.6 Hz, 1H, C3-H), 5.22 (t, J = 9.6 Hz, 1H, C4-H), 5.46 (d, J = 3.2 Hz, 1H, C2-H), 5.61 (d, J = 6.8 Hz, 1H, C1-H), 7.12-7.32 (m, 10H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 20.2 (q), 20.3 (q), 20.37 (q), 20.38 (q), 61.7 (t), 65.0 (d), 67.8 (d, J_{C-P} = 8.5 Hz), 69.8 (d), 72.7 (d), 94.5 (d, J_{C-P} = 4.6 Hz), 120.0 (d, J_{C-P} = 4.5 Hz), 120.1 (d, J_{C-P} = 4.5 Hz), 125.4 (d), 125.6 (d), 129.4 (d), 129.6 (d), 149.7 (s, J_{C-P} = 7.6 Hz), 150.1 (s, J_{C-P} = 8.5 Hz), 169.3 (s), 169.4 (s), 169.5 (s) 170.3 (s) ppm.

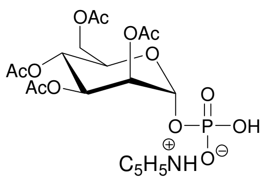
Diphenyl-(2,3,4,6-tetra-*O*-acetyl-*α*-D-mannopyranosyl)-phosphate (*α*-4.41):[44] A solution of DMAP (0.80 g, 6.6 mmol, 2.3 eq) and diphenyl chlorophosphate (1.4 mL, 6.8 mmol) in dichloromethane (6.7 mL) was added over 30 min to a solution of 2,3,4,6-tetra-*O*-acetyl-D-mannopyranose (1.00 g, 2.87 mmol) in dichloromethane (16.7 mL) at -30 °C. The resulting mixture was stirred under argon for 1 h at -30 °C and then maintained for 2 h between -10 and 0 °C. Work-up and purification was performed as described for ***β*-4.41** to give **α-4.41** (1.09 g, 1.88 mmol, 65%) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 1.95 (s, 3H, Ac), 1.98 (s, 3H, Ac), 2.02 (s, 3H, Ac), 2.13 (s, 3H, Ac), 3.89 (dd, J = 2.0, 12.4 Hz, 1H, C6-H), 4.05 (m, 1H, C5-H), 4.16 (dd, J = 4.8, 12.4 Hz, C6-H'), 5.23-5.37 (m, 3H, C2-H, C3-H and C4-H), 5.85 (dd, J = 2.0, 6.8, 1H, C1-H), 7.18-7.37 (m, 10H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 20.30 (q), 20.31 (q), 20.36 (q), 20.41 (q), 61.4 (t), 64.8 (d), 67.9 (d), 68.4 (d, J_{C-P} = 12.3 Hz), 70.5 (d), 95.9 (d, J_{C-P} = 5.3 Hz), 119.8 (d, J_{C-P} = 4.6 Hz), 120.0 (d, J_{C-P} = 4.5 Hz), 125.6 (d), 125.7 (d), 129.7 (d), 129.8 (d), 149.8 (s, J_{C-P} = 6.9 Hz), 150.0 (s, J_{C-P} = 6.8 Hz), 169.3 (s), 169.5 (s), 170.3 (s) ppm.

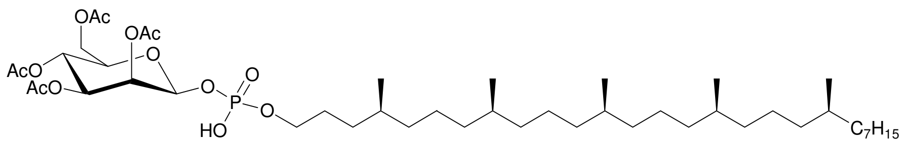


Pyridinium (2,3,4,6-tetra-*O*-acetyl-*β*-D-mannopyranosyl)-phosphate (*β*-4.42): *β*-**4.41** (667 mg, 1.15 mmol) was dissolved in EtOH-EtOAc (14,4 mL, 1:1 v/v) and PtO_2 (29 mg, 0.13 mmol, 0.11 eq) was added. The resulting suspension was degassed with 3 vacuum-argon cycles and then saturated with H_2 by 5 vacuum-H_2 cycles. After stirring over for 60 h under an H_2-atmosphere (1 bar), TLC (chloroform/MeOH/water 60/35/6 v/v/v) showed complete conversion. The catalyst was removed by filtration over celite and the solution was neutralized with pyridine. After concentration *β*-**4.41** (469 mg, 0.92 mmol, 80%) was isolated as a white foam. ^{1}H-NMR (D_2O, 400 MHz) δ = 1.88 (s, 3H, Ac), 1.96 (s, 3H, Ac), 1.99 (s, 3H, Ac), 2.10 (s, 3H, Ac), 3.93 (ddd, J = 2.0, 3.6, 10.0 Hz, 1H, C5-H), 4.07 (dd, J = 2.0, 12.8 Hz, 1H, C6-H), 4.31 (ddd, J = 3.6, 12.8 Hz, 1H, C6-H'), 5.09 (t, J = 10.0 Hz, 1H, C4-H), 5.21 (dd, J = 3.2, 10.4 Hz, 1H, C3-H), 5.33 (dd, J = 0.8, 8.8 Hz, 1H, C1-H), 5.40 (d, J = 2.8 Hz, 1H, C2-H), 7.95 (t, J = 7.2 Hz, 2H, Py), 8.50 (m, 1H, Py), 8.65 (d, J = 5.2 Hz, 2H, Py) ppm. ^{13}C-NMR (D_2O, 50.32 MHz) δ = 18.6 (q), 60.3 (t), 64.0 (d), 68.5 (d, J_{C-P} = 6.8 Hz), 69.6 (d), 70.3 (d), 91.8 (d), 125.9 (d), 171.1 (s), 171.6 (s), 171.7 (s), 172.2 (s) ppm. Positive ion ESI-HRMS calcd for $C_{14}H_{21}O_{13}NaP$ $(M-C_5H_6N^+ + Na^+ + H^+)^+$: 451.062, found: 451.062.

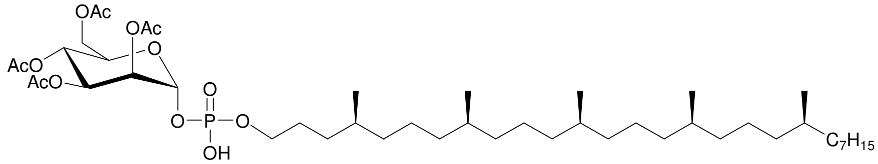


Pyridinium (2,3,4,6-tetra-*O*-acetyl-*α*-D-mannopyranosyl)-phosphate (*α*-4.42): *α*-**4.41** (1.0 g, 1.7 mmol) was converted into *α*-**4.42** (831 mg, 1.64 mmol, 95%) using a procedure analogous to the synthesis of *β*-**4.42**. ^{1}H-NMR (D_2O, 400 MHz) δ = 1.92 (s, 3H, Ac), 2.00 (s, 3H, Ac), 2.03 (s, 3H, Ac), 2.10 (s, 3H, Ac), 4.07 (d, *J* = 12.8, 1H, C6-H), 4.28 (d, *J* = 10.0 Hz, 1H, C5-H), 4.36 (dd, *J* = 2.8, 12.8 Hz, 1H, C6-H'), 5.20 (t, *J* = 10.0 Hz, 1H, C4-H), 5.23 (brs,1H, C2-H), 5.32 (dd, *J* = 3.2, 10.0 Hz, 1H), 5.40 (d, *J* = 8.0 Hz, 1H, C1-H), 7.99 (t, *J* = 7.2 Hz, 2H, Py), 8.50 (m, 1H, Py), 8.69 (d, *J* = 5.6 Hz, 2H, Py) ppm. ^{13}C-NMR (D_2O, 50.32 MHz) δ = 18.6 (q), 60.4 (t), 64.0 (d), 67.3 (d), 67.9 (d), 68.3 (d, $J_{C\text{-}P}$ = 10.3 Hz), 91.6 (d, *J* = 4.6 Hz), 125.9 (d), 171.1 (s), 171.3 (s), 171.7 (s), 172.2 (s) ppm.

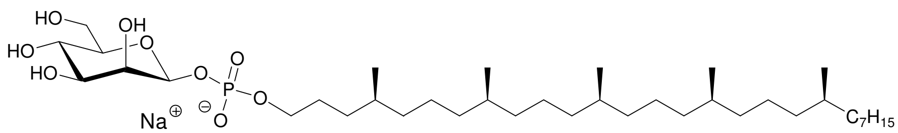




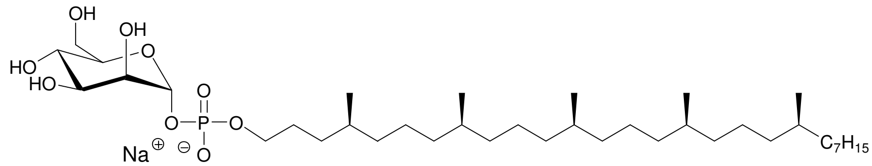
(4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethylheptacosylphosphoryl-2,3,4,6-tetra-*O*-acetyl-*β*-D-mannopyranoside (*β*-4.43): *β*-**4.42** (129 mg, 0.25 mmol, 2.0 eq) was brought into a dry two-neck flask (50 mL) and co-evaporated with freshly distilled dry toluene (2x; pressure equalized with nitrogen). Subsequently a solution of **4.34** (59 mg, 0.13 mmol) in dry toluene was added and the mixture was co-evaporated with dry toluene (2x). Finally TPSCl (115 mg, 0.38 mmol, 3.0 eq) was added and after two more additions and evaporations of dry toluene, the residue was dissolved in dry pyridine (2.7 mL) and stirred under argon for 4 days. The reaction was quenched with MeOH (2.5 mL) and stirred for 2 more h before concentration (co-evaporation with toluene). The residue was dissolved in chloroform and washed with water (3x) and brine (sat.), dried ($MgSO_4$), filtered and concentrated. The product was purified by column chromatography (chloroform-MeOH 95:5 to 85:15) giving *β*-**4.43** (90 mg, 0.10 mmol, 79%) as a white foam. (4S,8S,12S,16S,20S)-*β*-**4.43**: $[\alpha]_D^{22}$ = -9.7° (c = 1.22, $CHCl_3$), ^{1}H-NMR (CD_3OD + 1 drop of $CDCl_3$, 400 MHz) δ = 0.82-0.92 (m, 18H), 1.03-1.46 (m, 43H), 1.61 (m, 2H), 1.95 (s, 3H, Ac), 2.04 (s, 3H, Ac), 2.06 (s, 3H, Ac), 2.16 (s, 3H, Ac), 3.86 (m, 3H, C5-H and CH_2-OP), 4.16 (dd, *J* = 2.4, 12.4 Hz, 1H, C6-H), 4.28 (dd, *J* = 4.4, 12.0 Hz, 1H, C6-H'), 5.17-5.25 (m, 2H, C3-H, C4-H), 5.42 (d, *J* = 8.8 Hz, 1H, C1-H), 5.48 (brs, 1H, C2-H) ppm. ^{13}C-NMR ($CDCl_3$-CD_3OD 1:1 (v/v), 100.6 MHz) δ = 14.3 (q), 19.7-20.0 (q), 20.7 (q), 20.8 (q), 23.0 (t), 24.8-24.9 (t), 27.4 (t), 28.5 (t), 28.6 (t), 29.7 (t), 30.3 (t), 32.3 (t), 33.1-33.2 (d), 33.4 (t), 33.5 (t), 37.4 (t), 37.5 (t), 37.8-37.9 (t), 62.6 (t), 66.0 (d), 67.1 (t), 70.1 (d, $J_{C\text{-}P}$ = 6.1 Hz), 71.5 (d), 72.8 (d), 94.0 (d), 170.5 (s), 170.6 (s), 171.2 (s), 171.5 (s) ppm. ^{31}P-NMR ($CDCl_3$-CD_3OD 1:1 (v/v), 161.6 MHz) δ = -5.6 ppm. Positive ion ESI-HRMS calcd for $C_{46}H_{85}O_{13}NaP$ $(M + Na)^+$: 899.563, found: 899.559.



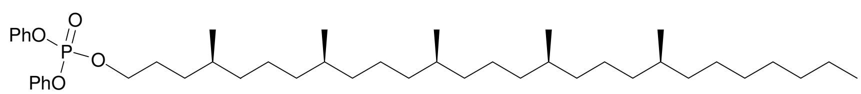
(4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethylheptacosylphosphoryl-2,3,4,6-tetra-*O*-acetyl-*α*-D-mannopyranoside (*α*-4.43): *α*-**4.42** (110 mg, 0.21 mmol, 2.0 eq) was coupled to **4.34** (50 mg, 0.11 mg, 1.0 eq) in the presence of TPSCl (98 mg, 0.32 mmol, 3.0 eq) to give *α*-**4.43** (49 mg, 56 µmol, 52%) using a procedure analogous to the synthesis of *β*-**4.43**. ^{1}H-NMR (CD_3OD + 1 drop of $CDCl_3$, 400 MHz) δ = 0.82-0.92 (m, 18H), 1.03-1.46 (m, 43H), 1.61 (m, 2H), 1.96 (s, 3H, Ac), 2.04 (s, 3H, Ac), 2.07 (s, 3H, Ac), 2.15 (s, 3H, Ac), 3.89 (m, 2H, CH_2-OP), 4.11 (dd, *J* = 2.4, 12.0 Hz, 1H, C6-H), 4.22 (m, 1H, C5-H), 4.29 (dd, *J* = 4.0, 12.4 Hz, 1H, C6-H'), 5.28-5.37 (m, 3H, C2-H, C3-H and C4-H), 5.49 (d, *J* = 6.8 Hz, 1H, C1-H) ppm. ^{13}C-NMR ($CDCl_3$-CD_3OD 1:1 (v/v), 100.6 MHz) δ = 14.5 (q), 20.0-20.3 (q), 20.7 (q), 20.81 (q), 20.85 (q), 23.4 (t), 25.2-25.3 (t), 27.8 (t), 29.1 (t), 29.2 (t), 30.2 (t), 30.7 (t), 32.7 (t), 33.5-33.6 (d), 33.9 (t), 37.8 (t), 37.9 (t), 38.15-38.23 (t), 63.1 (t), 66.5 (d), 67.4 (t), 70.0 (d), 70.3 (d), 70.6 (d, $J_{C\text{-}P}$ = 9.2 Hz), 94.5 (d), 170.99 (s), 171.02 (s), 171.1 (s), 171.9 (s) ppm. ^{31}P-NMR ($CDCl_3$-CD_3OD 1:1 (v/v), 161.6 MHz) δ = -5.8 ppm. Negative ion ESI-HRMS calcd for $C_{46}H_{84}O_{13}P^-$ (M-H)$^-$: 875.566, found: 875.563.



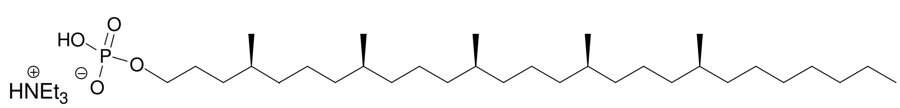
Sodium (4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethylheptacosylphosphoryl-*β*-D-mannopyranoside (*β*-4.1): *β*-**4.43** (130 mg, 0.15 mmol) was dissolved in chloroform-MeOH (3.5 mL, 1:2.5 v/v) and NaOMe was added (14 mg). The resulting solution was stirred for 60 min and then diluted with 1-butanol (30 mL) and quenched with aq. NH_4Cl (sat. 20 mL). The organic layer was washed with water (3 x 20 mL) and then concentrated by co-evaporation with toluene. The resulting solid was placed on a pad of Celite and washed with acetone (50 mL) and then chloroform-MeOH (70 mL, 1:1 v/v). The chloroform-MeOH extract was concentrated to give *β*-**4.1** (108 mg, 0.15 mmol, quantitative) as a white solid. (4S,8S,12S,16S,20S)-*β*-**4.1**: $[\alpha]_D^{22}$ = -3.6^o (c = 0.55, $CHCl_3$-CH_3OH-H_2O 95:95:10 v/v/v), mp = 125 ^{o}C (decomp.), ^{1}H-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v), 400 MHz) δ = 0.82-0.88 (m, 18H), 1.00-1.42 (m, 43H), 1.61 (m, 2H), 3.28-3.32 (m, 1H, C5-H), 3.52-3.59 (m, 2H, C3-H and C4-H), 3.73 (dd, *J* = 5.6, 12.0 Hz, 1H, C6-H), 3.82-3.88 (m, 3H, C6-H' and CH_2-OP), 3.93 (d, *J* = 2.4 Hz, 1H, C2-H), 5.07 (d, *J* = 8.0 Hz, 1H, C1-H) ppm. ^{13}C-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v),125.7 MHz) δ = 14.5 (q), 19.9 (q), 20.2-20.3 (q), 23.3 (t), 25.0 (t), 25.1 (t), 25.2 (t), 25.3 (t), 27.7 (t), 28.96 (t), 29.02 (t), 30.0 (t), 30.6 (t), 32.6 (t), 33.3-33.5 (d), 33.8 (t), 33.9 (t), 37.6 (t), 37.7 (t), 38.0-38.2 (t), 61.8 (t), 67.3 (d) 67.4 (t, $J_{C\text{-}P}$ = 6.2 Hz), 72.1 (d), 74.0 (d), 77.9 (d), 96.3 (d) ppm. ^{31}P-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v),161.6 MHz) δ = -1.6 ppm. Negative ion ESI-HRMS calcd for $C_{38}H_{76}O_9P^-$ (M-Na$^+$)$^-$: 707.523, found: 707.524.



Sodium (4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethylheptacosylphosphoryl-*α*-D-mannopyranoside (*α*-4.1): *α*-**4.43** (42 mg, 48 µmol) was converted into *α*-**4.1** (35 mg, 48 µmol, quantitative) as described for the preparation of *β*-**4.1**. ^{1}H-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v), 400 MHz) δ = 0.82-0.88 (m, 18H), 0.98-1.42 (m, 43H), 1.61 (m, 2H), 3.58-3.65 (m, 2H), 3.71 (dd, *J* = 5.6, 11.2 Hz, 1H, C6-H), 3.75-3.85 (m, 4H), 3.90 (m, 1H) 5.38 (dd, *J* = 1.6, 7.6 Hz, 1H, C1-H) ppm. ^{13}C-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v),125.7 MHz) δ = 14.4 (q), 14.5 (q), 19.8 (q), 19.9 (q), 20.1-20.3 (q), 23.3 (t), 25.0 (t), 25.1-25.3 (t), 27.6 (t), 28.9 (t), 29.0 (t), 30.0 (t), 30.5 (t), 32.5 (t), 33.3-33.4 (d), 33.7 (t), 33.8 (t), 37.6 (t), 37.7 (t), 38.0 (t), 61.7 (t), 67.1 (t, $J_{C\text{-}P}$ = 5.3 Hz), 67.5 (d), 71.0 (d), 71.6 (d, $J_{C\text{-}P}$ = 8.4 Hz), 74.5 (d), 77.9 (d), 96.6 (d, $J_{C\text{-}P}$ = 5.4 Hz) ppm. ^{31}P-NMR (CD_3OD-$CDCl_3$-D_2O 0.45:0.45:0.05 (v/v/v),161.6 MHz) δ = -1.8 ppm. Negative ion ESI-HRMS calcd for $C_{38}H_{76}O_9P^-$ (M-Na^+)$^-$: 707.523, found: 707.521.



(4S,8S,12S,16S,20S)-4,8,12,16,20-Pentamethylheptacosyl diphenyl phosphate (4.44): DMAP (30 mg, 0.25 mmol, 2.3 eq) was added to a solution of **4.34** (50 mg, 0.11 mmol) in dichloromethane (0.63 mL) and the resulting mixture was cooled to 0 ^{o}C. Subsequently, a solution of diphenyl chlorophosphate (51 µl, 0.25 mmol, 2.3 eq) in dichloromethane (0.26 mL) was added over 5 min. The solution was allowed to warm to room temperature and stirred overnight. The reaction mixture was diluted with dichloromethane and then washed with water, aq. HCl (1.0 M), aq. $NaHCO_3$ (sat.) and brine (sat.). The organic layer was dried ($MgSO_4$), filtered and concentrated. Purification by column chromatography (*n*-pentane-EtOAc 95:5 to 92.5:7.5) yielded **4.44** (75 mg, 0.11 mmol, quantitative) as a colorless oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.81-0.95 (m, 18H), 1.01-1.44 (m, 43H), 1.71 (m, 2H), 4.25 (q, *J* = 6.8 Hz, 2H), 7.19 (t, *J* = 7.2 Hz, 1H), 7.25 (m, 2H), 7.35 (m, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 14.0 (q), 19.4 (q), 19.6 (q), 22.6 (t), 24.3 (t), 24.4 (t), 27.0 (t), 27.65 (t), 27.72 (t), 29.3 (t), 29.9 (t), 31.8 (t), 32.25 (d), 32.34 (t), 32.4 (t), 32.7 (d), 36.9 (t), 37.1 (t), 37.3 (t), 69.7 (t, $J_{C\text{-}P}$ = 6.9 Hz), 119.9 (d, *J* = 5.3 Hz), 125.1 (d), 129.6 (d), 150.6 (s) ppm. ^{31}P-NMR ($CDCl_3$, 161.6 MHz) δ = -12.1 ppm.



Triethylammonium-(4S,8S,12S,16S,20S)-4,8,12,16,20-pentamethyl-heptacosylphosphate (4.37): Alcohol **4.44** (70 mg, 0.10 mmol) was dissolved in EtOH-EtOAc (3.0 mL, 1:1 v/v) and PtO_2 (5 mg, 22 µmol, 0.22 eq) was added. The resulting suspension was degassed with 3 vacuum-argon cycles and then saturated with H_2 by 5 vacuum-H_2 cycles. After stirring for 60 h under an H_2-atmosphere (1 bar), ^{1}H-NMR showed complete conversion. The catalyst was removed by filtration over celite and the solution was neutralized with triethylamine. After concentration **4.44** (65 mg, 0.10 mmol, quantitative) was isolated as a yellow oil. ^{1}H-NMR ($CDCl_3$, 400 MHz) δ = 0.78-0.90 (m, 18H), 0.96-1.40 (m, 44H), 1.29 (t, J = 7.2 Hz, 9H), 1.59 (m, 2H),3.03 (q, J = 7.2 Hz, 6H), 3.84 (m, 2H) ppm. ^{13}C-NMR ($CDCl_3$, 100.6 MHz) δ = 0.9 (q), 8.8 (q), 14.0 (q), 19.6 (q), 22.5 (t), 24.4 (t), 26.9 (t), 28.5 (t), 29.2 (t), 29.8 (t), 31.8 (t), 32.6 (d), 33.0 (t), 36.9 (t), 37.3 (t), 45.3 (t), 65.3 (t) ppm. ^{31}P-NMR ($CDCl_3$, 161.6 MHz) δ = 1.8 ppm.

4.10 References

[1] Moody, D. B.; Ulrichs, T.; Mühlecker W.; Young, D. C.; Gurcha, S. S.; Grant, E.; Rosat, J.-P.; Brenner, M. B.; Costello, C. E.; Besra, G. S.; Porcelli, S. A. *Nature* **2000**, *404*, 884-888.
[2] Crich, D.; Dudkin, V. *J. Am. Chem. Soc.* **2002**, *124*, 2263-2266.
[3] Besra, G. S.; Sievert, T.; Lee, R. E.; Slayden, R. A.; Brennan, P. J.; Takayama, K. *Proc. Natl. Acad. Sci.* **1994**, *91*, 12735-12739.
[4] Moody, D. B. ; Besra, G. S.; Wilson, I. A.; Porcelli, S. A. *Immunol. Rev.* **1999**, *172*, 285-296.
[5] Gumperz, J. E.; Brenner, M. B.; *Curr. Opin. Immunol.* **2001**, *13*, 471-478.
[6] Porcelli, S. A.; Morita, C. T.; Brenner, M. B. *Nature* **1992**, *360*, 593-597.
[7] Beckman, E. M.; Porcelli, S. A.; Morita, C. T.; Behar, S. M.; Furlong, S. T.; Brenner, M. B. *Nature* **1994**, *372*, 691-694.
[8] Ulrichs, T.; Moody, D. B.; Grant, E.; Kaufmann, S. H.; Porcelli, S. A. *Infect. Immun.* **2003**, *71*, 3076-3087.
[9] Lawton, A. P.; Kronenberg, M. *Immunol. Cell Biol.* **2004**, *82*, 295-306.
[10] (a) Shamshiev, A.; Donda, A.; Carena, I.; Mori, L.; Kappos, L.; De Libero, G. *Eur. J. Immunol.* **1999**, *29*, 1667-1675; (b) Shamshiev, A.; Gober, H. -J.; Donda, A.; Mazorra Z.; Mori, L.; De Libero, G. *J. Exp. Med.* **2002**, *195*, 1013-1021.
[11] Sieling, P. A.; Chatterjee, D.; Porcelli, S. A.; Prigozy, T. I.; Mazzaccaro, R. J.; Soriano, T.; Bloom, B. R.; Brenner, M. B.; Kronenberg, M.; Brennan, P. J.; Modlin, R. L. *Science* **1995**, *269*, 227-230.
[12] Gilleron, M.; Stenger, S.; Mazorra, Z.; Wittke, F.; Mariotti, S.; Böhmer, G.; Prandi, J.; Mori, L.; Puzo, G.; De Libero, G. *J. Exp. Med.* **2004**, *199*, 649-659.
[13] Moody, D. B.; Young, D. C.; Cheng, T. -Y.; Rosat, J. -P.; Roura-mir, C.; O'Connor, P. B.; Zajonc, D. M.; Walz, A.; Miller, M. J.; Levery, S. B.; Wilson, I. A.; Costello, C. E.; Brenner, M. B. *Science* **2004**, *303*, 527-531.
[14] (a) Zeng, Z. -H.; Castaño, A. R.; Segelke, B. W.; Stura, E. A.; Peterson, P. A.; Wilson, I. A. *Science* **1997**, 339-345; (b) Gadola, S. D.; Zaccai, N. R.; Harlos, K.; Shepherd, D.; Castro-Palomino J. C.; Ritter, G.; Schmitt, R. R.; Jones, E. Y.; Cerundolo, V. *Nat. Immunol.* **2002**, *3*, 721-726; (c) Zajonc, D. M.; Elsliger, M. A.; Teyton, L.; Wilson, I. A. *Nat. Immunol.* **2003**, *4*, 808-815.

[15] (a) Moody, D. B.; Reinhold, B. B.; Guy, M. R.; Beckman, E. M.; Frederique, D. E.; Furlong, S. T.; Ye, S.; Reinhold, V. N.; Sieling, P. A.; Modlin, R. L.; Besra, G. S.; Porcelli, S. A. *Science* **1997**, *278*, 283-286; (b) Kawano, T.; Cui, J. Q.; Koezuka, Y.; Toura, I.; Kaneko, Y.; Motoki, K.; Ueno, H.; Nakagawa, R.; Sato, H.; Kondo, E.; Koseki, H.; Taniguchi, M. *Science* **1997**, *278*, 1626-1629; (c) Moody, D. B.; Guy, M. R.; Grant, E.; Cheng, T. Y.; Brenner, M. B. *J. Exp. Med.* **2000**, *192*, 965-976.
[16] Brigl, M.; Brenner, M. B. *Annu. Rev. Immunol.* **2004**, *22*, 817-890.
[17] Minnikin, D. E.; Kremer, L.; Dover, L. G.; Besra, G. S. *Chem. Biol.* **2002**, *9*, 545-553.
[18] Sugita, M.; Porcelli, S. A.; Brenner, M. B. *J. Immunol*. **1997**, *159*, 2358-2365.
[19] (a) Huttinger, R.; Staffler, G.; Majdic, O.; Stockinger, H. *Int. Immunol.* **1999**, *11*, 1615-1623; (b) Kang, S. J.; Cresswell. P. *J. Biol. Chem.* **2002**, *277*, 44838-44844.
[20] (a) Jayawardena-Wolf, J.; Benlagha, K.; Chiu, Y. H.; Mehr, R.; Bendelac, A. *Immunity* **2001**, *15*, 897-908. (b) Briken, V.; Jackman, R. M.; Dasgupta, S.; Hoening, S.; Porcelli, S. A. *EMBO J.* **2002**, *21*, 825-834.
[21] (a) Zhou, D.; Cantu, C. 3rd; Sagiv, Y.; Schrantz, N.; Kulkarni, A. B.; Qi, X.; Mahuran, D. J.; Morales, C. R.; Grabowski, G. A.; Benlagha, K.; Savage, P.; Bendelac, A.; Teyton, L. *Science* **2004**, *303*, 523-527; (b) Kang, S. J.; Cresswell, P. *Nat. Immunol.* **2004**, *5*, 175-181.
[22] Grant, E. P.; Degano, M.; Rosat, J. -P.; Stenger, S.; Modlin, R. L.; Wilson, I. A.; Porcelli, S. A.; Brenner, M. B. *J. Exp. Med.* **1999**, *189*, 195-205.
[23] (a) Porcelli, S. A.; Brenner, M. B.; Greenstein, J. L.; Balk, S. P.; Terhorst, C.; Bleicher, P. A. *Nature* **1989**, *341*, 447-450; (b) Spada, F. M.; Grant, E. P.; Peters, P. J.; Sugita, M.; Melián, A.; Leslie, D. S.; Lee, H. K.; Van Donselaar, E.; Hanson, D. A.; Krensky, A. M.; Majdic, O.; Porcelli, S. A.; Morita, C. T.; Brenner, M. B. *J. Exp. Med.* **2000**, *191*, 937-948.
[24] Grant, E. P.; Beckman, E. M.; Behar, S. M.; Degano, M.; Frederique, D.; Besra, G. S.; Wilson, I. A.; Porcelli, S. A.; Furlong, S. T.; Brenner, M. B. *J. Immunol.* **2002**, *168*, 3933-3940.
[25] Spada, F. M.; Koezuka, Y.; Porcelli, S. A. *J. Exp. Med.* **1998**, *188*, 1529-1534.
[26] Matsunaga, I.; Bhatt, A.; Young, D. C.; Cheng, T. -Y.; Eyles, S. J.; Besra, G. S.; Briken, V.; Porcelli, S. A.; Costello, C. E.; Jacobs Jr., W. R.; Moody, D. B. *J. Exp. Med.* **2004**, *200*, 1559-1569.
[27] Stenger, S.; Mazzaccaro, R. J.; Uyemura, K.; Cho, S.; Barnes, P. F.; Rosat, J. -P.; Sette, A.; Brenner, M. B.; Porcelli, S. A.; Bloom, B. R.; Modlin, R. L. *Science* **1997**, *276*, 1684-1687.
[28] Roura-Mir, C.; Moody, D. B. *Microbes Infect.* **2003**, *5*, 1137-1148.
[29] Toshima, K.; Tatsuta, K. *Chem. Rev.* **1993**, *93*, 1503-1531.
[30] (a) Crich, D.; Sun, S. *J. Org. Chem.* **1996**, *61*, 4506-4507; (b) Crich, D.; Sun, S. *J. Org. Chem.* **1997**, *62*, 1198-1199; (c) Crich, D.; Sun, S. *J. Am. Chem. Soc.* **1997**, *119*, 11217-11223; (d) Crich, D.; Sun, S. *J. Am. Chem. Soc.* **1998**, *120*, 435-436. (e) Crich, D.; Chandrasekera, N. S. *Angew. Chem. Int. Ed.* **2004**, *43*, 5386-5389; (f) Crich, D.; Sun, S. *Tetrahedron* **1998**, *54*, 8321-8348; (g) Crich, D.; Dudkin. V. *Org. Lett.* **2000**, *2*, 3941-3943.
[31] Litjens, R. *Sulfonium Salt Activation in Oligosaccharide Synthesis*. Ph.D. Thesis, Leiden, **2005**, 18.
[32] Andrews, C. W.; Rodebaugh, R.; Fraser-Reid, B. *J. Org. Chem.* **1996**, *61*, 5280-5289.

[33] Des Mazery, R.; Pullez, M.; López, F.; Harutyunyan, S. R.; Minnaard A. J.; Feringa, B. L. *J. Am. Chem. Soc.* **2005**, *127*, 9966-9967.
[34] a) Heathcock, C. H.; Finkelstein, L.; Jarvi, E. T.; Radel, P. A.; Hadley, C. R. *J. Org. Chem.* **1988**, *53*, 1922-1942; b) Eguchi, T.; Arakawa, K.; Terachi, T.; Kakinuma, K. *J. Org. Chem.* **1997**, *62*, 1924-1933.
[35] Berkowitz, W. F.; Wu, Y. *Tetrahedron Lett.* **1997**, *38*, 8141-8144.
[36] Berkowitz, W. F.; Wu, Y. *Tetrahedron Lett.* **1997**, 38, 3171-3174.
[37] Blakemore, P. R.; Cole, W. J.; Kocienski, P. J.; Morely, A. *Synlett*, **1998**, 26-28.
[38] a) Julia, M.; Paris. J. -M. *Tetrahedron Lett.* **1973**, *14*, 4833-4836; b) Lythgoe, B.; Ruston, S. *J. Chem. Soc., Perkin Trans. 1*, **1978**, 829-834; c) Kocienski, P. J.; Lythgoe, B.; Waterhouse, I. *J. Chem. Soc., Perkin Trans. 1*, **1980**, 1045-1050.
[39] Lautens, M.; Colucci, J. T.; Hiebert, S.; Smith, N. D.; Bouchain, G. *Org. Lett.* **2002**, *4*, 1879-1882.
[40] Griffith, W. P.; Ley, S. V.; Whitcombe, G. P.; White, A. D. *J. Chem. Soc. Chem. Commun.* **1987**, 1625-1627.
[41] Keck, G. E.; Boden, E. P.; Mabury, S. A. *J. Org. Chem.* **1985**, *50*, 709-710.
[42] Kocienski, P. J.; Bell, A.; Blakemore, P. R. *Synlett* **2000**, 365-366.
[43] (a) Smith, G. V.; Roth, J. A.; Desai, D. S.; Kosco, J. L. *J. Catal.* **1973**, *30*, 79-85; (b) Cohen, N.; Eichel, W. F.; Lopresti, R. J.; Neukom, C.; Saucy, G. *J. Org. Chem.* **1976**, *41*, 3512- 3515; (c) Schwartz, B. D.; Hayes, P. Y.; Kitching, W.; De Voss, J. J. *J. Org. Chem.* **2005**, *70*, 3054-3065; (d) Dandapani, S.; Jeske, M.; Curran, D. P. *J. Org. Chem.* **2005**, *70*, 9447-9462.
[44] Sabesan, S.; Neira, S. *Carbohydr. Res.* **1992**, *223*, 169-185.
[45] Warren, C. D.; Liu, I. Y.; Herscovics, A.; Jeanloz, R. W. *J. Biol. Chem.* **1975**, *250*, 8069-8078.
[46] (a) Jacob, T. M.; Khorana, H. G. *J. Am. Chem. Soc.* **1964**, *86*, 1630-1635; (b) Lohrmann, R.; Khorana, H. G. *J. Am. Chem. Soc.* **1966**, *88*, 829-833.
[47] Schmidt, R. R.; Michel, J. *Tetrahedron Lett.* **1984**, *25*, 821-824.
[48] Wolucka, B. A.; Rush, J. S.; Waechter, C. J.; Shibaev, V. N.; De Hoffman, E. *Anal. Biochem.* **1998**, *255*, 244-251.
[49] Bock, K.; Pedersen, C. *J. Chem. Soc., Perkin Trans. 2* **1974**, *293-297.*
[50] Brigham and Women's Hospital, Harvard Medical School, Boston, USA.
[51] (a) Martín-Lomas, M.; Khiar, N.; García, S.; Koessler, J.-L.; Nieto, P. M.; Rademacher, T. W. *Chem. Eur. J.* **2000**, *6*, 3608-3621; (b) Oshitari, T.; Shibasaki, M.; Yoshizawa, T.; Tomita, M.; Takao, K.-i.; Kobayashi, S. *Tetrahedron* **1997**, *53*, 10993-11006; (c) Silva, D. J.; Sofia, M. J. *Tetrahedron Lett.* **2000**, *41*, 855-858.
[52] (a) Hashimoto, M.; Kan, T.; Nozaki, K.; Yanagiya, M.; Shirahama, H.; Matsumoto, T. *J. Org. Chem.* **1990**, *55*, 5088-5107; (b) Harding, K. E.; Jones, M. W. *Heterocycles* **1989**, *28*, 663-668.
[53] Shibuya, H.; Yamamoto, Y.; Miura, I.; Kitagawa, I. *Heterocycles* **1982**, *17*, 215-218.
[54] Quast, H.; Bieber, L. *Chem. Ber.* **1981**, *114*, 3253-3272.

Samenvatting

Totaalsynthese van Enantiomeerzuivere Biomoleculen

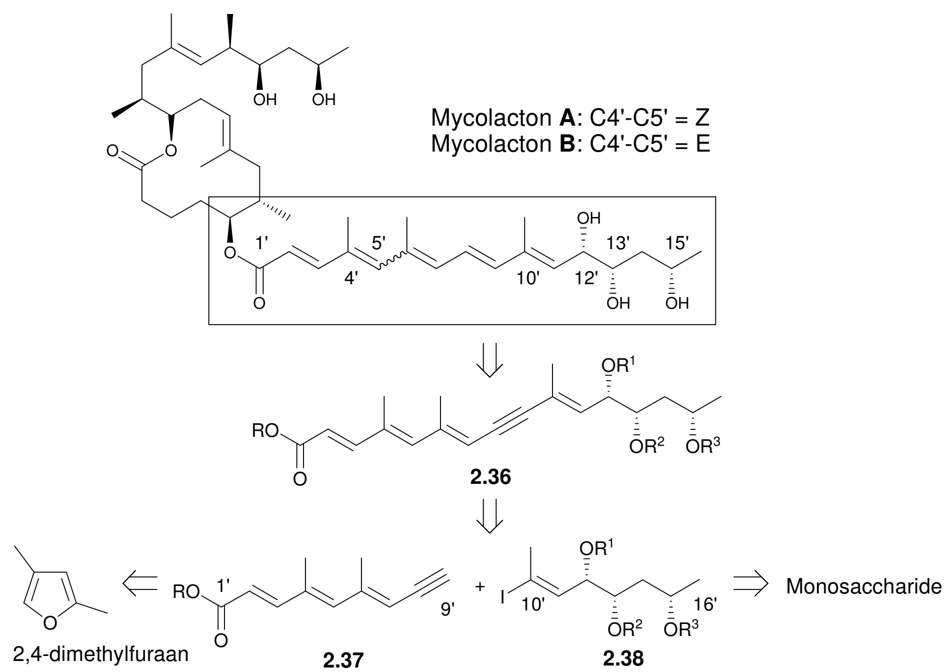
Over mycolactonen, verzadigde isoprenoide bouwstenen en β-mannosyl fosfomycoketiden

Totaalsynthese is een discipline binnen de organische synthese die zichzelf als doel stelt om complexe moleculen op te bouwen uit relatief eenvoudige uitgangsmaterialen. Oorspronkelijk was het zogenaamde 'doelmolecuul' vrijwel altijd een uit de natuur geïsoleerde verbinding. De functie van totaalsynthese bestond dan uit het verifiëren van de voorgestelde structuur van het molecuul. Tegenwoordig richt totaalsynthese zich ook op rationeel ontworpen complexe verbindingen. De natuur blijft echter de belangrijkste inspiratiebron wanneer het doelmoleculen betreft die interessant zijn vanwege hun (potentiële) biologische werking. Redenen die vandaag de dag ten grondslag liggen aan het bedrijven van totaalsynthese zijn ondermeer: a) opheldering van de stereochemie van een natuurproduct; b) onderzoek naar de relatie tussen de structuur en de (biologische) activiteit van een molecuul; c) het synthetiseren van een bruikbare hoeveelheid van een actieve verbinding voor medische applicaties; d) de synthetische uitdaging van een complexe structuur; e) het aantonen van de algemene bruikbaarheid van nieuwe synthetische methodes en f) het maken van verbindingen met interessante fysische of materiële eigenschappen.

Een belangrijk aspect in totaalsynthese is het introduceren van chirale elementen. Er zijn in principe drie manieren om een verbinding enantiomeerzuiver in handen te krijgen: 1) gebruik maken van enantiomeerzuivere bouwstenen (uit de natuur); 2) selectief een nieuw chiraal element creëren middels een reactie in aanwezigheid van een enantiomeerzuivere informatiedrager. Voorbeelden van informatiedragers zijn katalysatoren, reagentia of, in geval van diastereoselectiviteit, eventueel het uitgangsmateriaal zelf; 3) het scheiden van mengsels van enantio- en/of diastereoisomeren.

Welke aanpak het meest geschikt is voor een specifieke totaalsynthese hangt niet alleen af van de structuur van het doelmolecuul, maar ook van de motivatie voor het synthetiseren van dit molecuul. Bijvoorbeeld, indien een bibliotheek van stereoisomeren gemaakt dient te worden dan is flexibiliteit een belangrijk criterium. Wanneer daarentegen één specifiek stereoisomeer gesynthetiseerd dient te worden, bijvoorbeeld in geval van een geneesmiddel, dan zijn een efficiënte en korte route alsmede een hoge opbrengst en selectiviteit belangrijke uitgangspunten.

Hoofdstuk 2: In dit hoofdstuk worden nieuwe strategieën beschreven voor de synthese van de zijketens van mycolactonen (Figuur 1). Mycolactonen zijn toxines die uitgescheiden worden door *Mycobacterium ulcerans*, een bacterie die de ziekte Buruli ulcer veroorzaakt. Buruli ulcer is een aan lepra gerelateerde ziekte die gekenmerkt wordt door de vorming van grote zweren op de huid en het uitblijven van een immunologische reactie. Om de werking van de toxines goed te kunnen bestuderen, is het noodzakelijk om een efficiënte synthetische route te ontwikkelen die eventueel ook gebruikt kan worden om analoge verbindingen te maken.

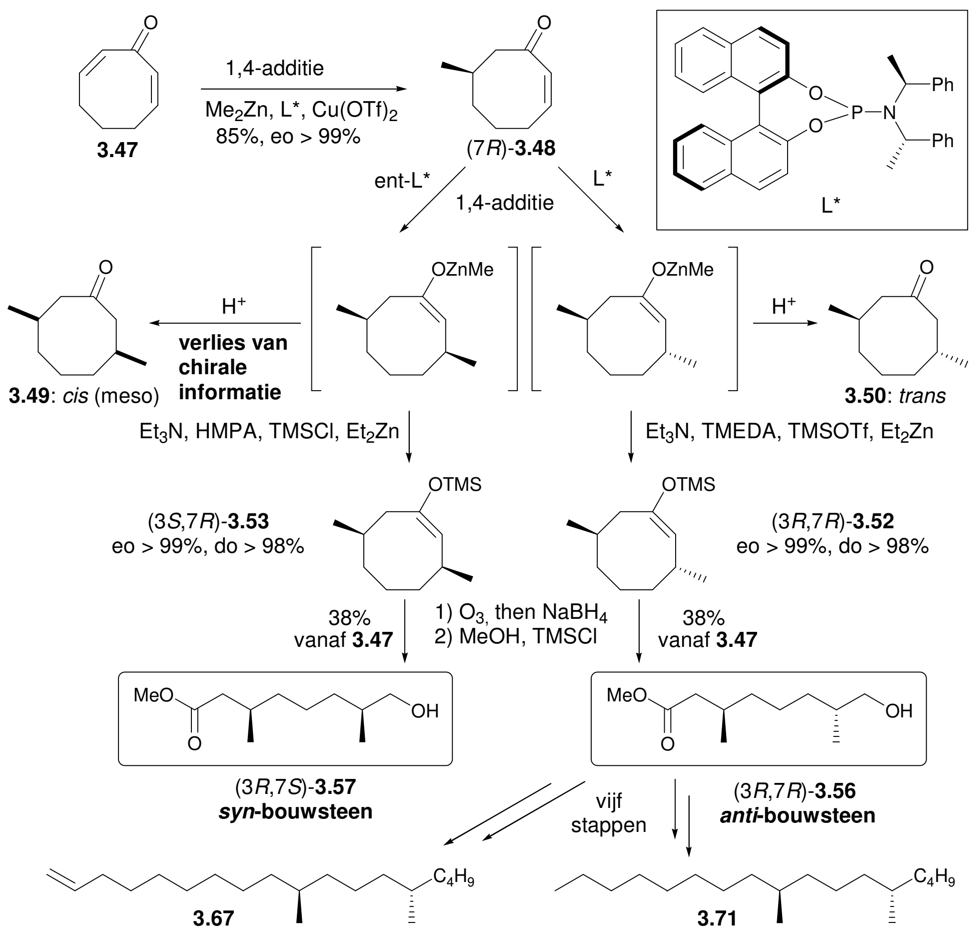


Figuur 1 *Structuur van mycolactonen en retrosynthetische analyse van de zijketen.*

Retrosynthetisch kan de zijketen verdeeld worden in een geconjugeerd fragment (**2.37**) en een chiraal gedeelte (**2.38**, Figuur 1). Voor de bereiding van het chirale gedeelte is gekozen voor het modificeren van natuurlijke bouwstenen, *in casu* monosacchariden. Door de grote verscheidenheid aan natuurlijke monosacchariden is de route in principe geschikt om analoga te maken, hoewel de bereiding daarvan wellicht niet altijd volgens gebaande paden mogelijk is. Voorts is er een nieuwe procedure ontwikkeld voor de synthese van een uitgebreid geconjugeerd fragment (**2.37**) in slechts drie stappen uitgaande van 2,4-dimethylfuraan. De koppeling van het geconjugeerde gedeelte aan de chirale bouwsteen resulteerde in de vorming van een 8,9-dehydroanaloog (**2.36**). Ondanks de inzet van een keur aan reductie- en hydrogeneringsmethoden is het tot op heden niet gelukt om het geconjugeerde alkyn selectief om te zetten in het corresponderende alkeen.

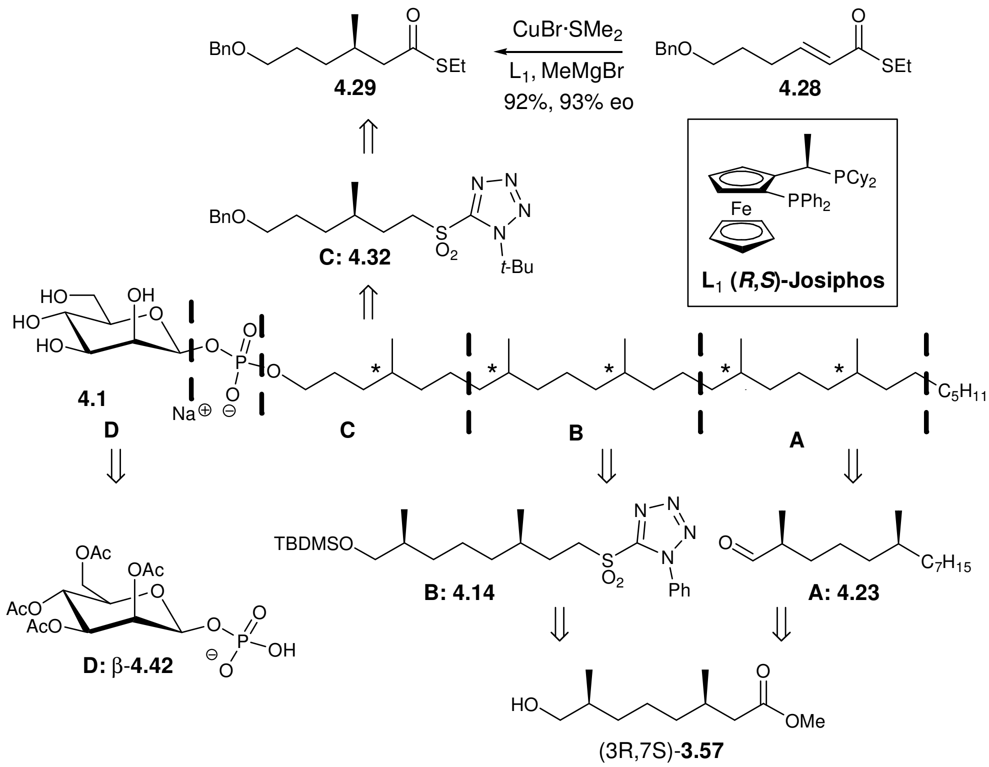
Hoofdstuk 3: Hierin komt de ontwikkeling van een katalytische methode voor de synthese van enantiomeerzuivere verzadigde isoprenoide bouwstenen aan bod. Dit structurele motief is alom aanwezig in verschillende klassen van biomoleculen en is daarom al decennia lang onderwerp van intensief synthetisch onderzoek. Tot voor kort ontbrak het echter aan een katalytische methode die in hoge opbrengst en selectiviteit toegang verleent tot alle vier mogelijke diastereomeren van flexibele bouwstenen zoals **3.56** en **3.57** (Schema 1).

Onze aanpak om invulling te geven aan dit hiaat is gebaseerd op de koper-fosforamidiet gekatalyseerde geconjugeerde additie van dialkylzinkreagentia aan cyclische α,β-onverzadigde ketonen. We anticipeerden erop dat twee opeenvolgende addities van dimethylzink aan cycloocta-2,7-dienon (**3.47**) middels dit protocol, gevolgd door oxidatieve ringopening zou leiden tot de gewenste bouwstenen. Hierbij dient aangetekend te worden dat, na de tweede geconjugeerde additie, het gevormde zink enolaat vastgelegd diende te worden om te voorkomen dat chirale informatie verloren zou gaan door vorming van *meso*-keton **3.49** in geval van een *cis*-adduct (Schema 1). Uiteindelijk werden alle vier bouwstenen bereid in vijf stappen (38% opbrengst vanaf **3.47**) met een enantiomere overmaat van 99% en een diastereomere overmaat (d.o.) van 98% volgens de route zoals afgebeeld in Schema 1. Om de synthetische bruikbaarheid van het nieuw ontwikkelde protocol te illustreren, is het vervolgens toegepast in the synthese van twee appelbladmineerder (*Lyonetia prunifoliella*) feromonen **3.67** en **3.71** (10 stappen, 20% opbrengst vanaf **3.47**), die het *anti*-isoprenoide motief bevatten (Schema 1).



Schema 1 *Synthese van enantiomeerzuivere verzadigde isoprenoide bouwstenen en toepassing in de synthese van twee appelbladmineerder feromonen.*

Hoofdstuk 4: Dit hoofdstuk is gewijd aan de totaalsynthese van β-mannosyl fosfomycoketide (MFM) **4.1** (Schema 2). MFM **4.1** is een glycolipide antigen dat onlangs is geïsoleerd uit de celwand van *Mycobacterium tuberculosis*. De stereochemie van de zijketen van **4.1** is momenteel nog onbekend en hieraan gerelateerd geldt in meer algemene zin dat er nog veel onduidelijkheid bestaat over de relatie tussen de structuur en de functie van de zijketen. Bij het ontwikkelen van een synthetische route voor **4.1** ging de aandacht dan ook met name uit naar de flexibiliteit van het protocol ten aanzien van de zijketen. Om precies te zijn, diende de route in staat te zijn om MFM's te vervaardigen met oligoisoprenoide ketens van elke gewenste lengte en stereochemische samenstelling. Als aanvangspunt is de diastereomeer van **4.1** met alle stereocentra van de zijketen in de S-configuratie gesynthetiseerd. Deze stereochemie wordt ook waarschijnlijk geacht die van de natuurstof te zijn op basis van de biosynthetische route.



Schema 2 *Structuur van β-mannosyl fosfomycoketide **4.1** en synthesestrategie voor de bereiding van de diastereomeer met de volledige-S configuratie van alle asymmetrische C-atomen in de zijketen.*

Om dit te bewerkstelligen, werden twee enantiomeerzuivere oligoisoprenoide bouwstenen **4.14** en **4.23** aan elkaar gekoppeld middels een Julia-Kocienski procedure (Schema 2). Op vergelijkbare wijze werd het aldus ontstane diisoprenoide gekoppeld aan een derde bouwsteen **4.32** met één methylgesubstitueerd asymmetrisch centrum. Voor de invoering van dit laatste asymmetrische centrum werd gebruik gemaakt van de onlangs ontwikkelde katalytische geconjugeerde additie van Grignard reagentia aan lineaire α,β-onverzadigde thioesters (**4.28** naar **4.29** Schema 2).

Een lastige hindernis in de synthese van (volledig-S)-*β*-**4.1** was de vorming van de *β*-mannosylfosfaatbinding. Om lage opbrengsten aan het einde van de totaalsynthese te voorkomen werd er eerst een fosfaatgroep op een afdoende beschermd mannopyranose geïnstalleerd (β-**4.42**, Schema 2) alvorens over te gaan tot koppeling met de enantiomeerzuivere zijketen. Op deze manier werd (volledig-S)-*β*-**4.1** uiteindelijk verkregen in een totale opbrengst van 6.7% na een langste lineaire sequentie van achttien stappen. Ter vergelijking werd het anomere analoog (volledig-S)-α-**4.1** volgens een vergelijkbare procedure bereid.

Biologische evaluatie wijst uit, dat synthetisch (volledig-S)-*β*-**4.1** en het natuurproduct een identieke activiteit aan de dag leggen, terwijl een analoog met een keten bestaande uit een statistisch mengsel van 32 diastereomeren aanzienlijk minder actief is. Dit resultaat impliceert dat de invloed van de zijketen op de biologische activiteit veel groter is dan tot nu toe werd verondersteld. Verder onderzoek moet uitwijzen wat het exacte verband is tussen de activiteit van MFM’s en de structuur van hun zijketen.