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Biology and Chemistry of Sphingosine-Related Metabolites

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Abstract: Metabolites of the thermophilic fungi *Myriococcum albomyces* and *Mycelia sterilia* such as myriocin (**1**), the mycestericins (**2-8**) and the sphingofungins (**13-18**) structurally resemble the sphingosines, important components of cell membranes. All the compounds revealed *in vitro* remarkable immunosuppressive activity and their pharmaceutical potential has led to the development of promising novel immunosuppressants. This review article describes the abundance, biology and chemistry of these metabolites.

1. INTRODUCTION

In 1972, two independent research groups isolated virtually simultaneously compounds named myriocin and thermozymocidin, which were metabolites of the culture broth of thermophilic fungi *Myriococcum albomyces* and *Mycelia sterilia*, respectively. The compounds were, as the more recently isolated ISP-1 from *Isaria sinclairii*, identical with **1** and are homologues of the sphingosines. The sphingosines are important components of all mammalian membranes, where some members play structural roles (e.g. sphingomyelin), whereas others appear to be important in cellular regulation (sphingosine, ceramide, glycosphingolipids). Although myriocin is historically the legitimate and most commonly used name for **1**, the terms thermozymocidin and especially ISP-1 are used frequently and may cause confusion at times.

The initially discovered immunosuppressant **1** is the most thoroughly investigated one of the sphingosine-related metabolites and its biological profile is best known. Several total syntheses with a variety of different approaches have been accomplished during the last two decades. The more recently isolated mycestericins (**2** - **8**) and sphingofungins (**13** - **18**) with similar biological properties have attracted a much lesser interest. In recent years, some related compounds as such as falvovirin (**19**) and malonofungin (**20**) were also discovered.

The discovery of these novel immunosuppressants with completely different activity profiles lead to the development of new highly potential drug candidates. The close relation of the metabolites to the sphingosines suggests an important role in regulatory processes of eukaryotic cell membranes. It is no surprise that there has been considerable synthetic interest for these compounds.

2. ISOLATION AND BIOLOGY OF SPHINGOSINE-ANALOGUES

2.1. Isolation and Biology of Myriocin

Kluepfel and co-workers from the Ayerst Research Laboratories isolated myriocin (**1**) for the first time in 1972 from the fermentation broth of the thermophilic fungus *Myriococcum albomyces*, an ascomycete, and the compound showed strong *in vitro* antifungal, but no significant antibiotic activity. Acute oral toxicity of **1** in mice was 50 times lower than intraperitoneal toxicity, probably due to poor absorption. Subcutaneous injection of 0.25 mg/kg in dog resulted in death of the animals after 2-3 days (Fig. (**1**)) [1].

In the same year, Aragozzini and co-workers reported the discovery of thermozymocidin, a highly effective antifungal metabolite found in the thermophilic mould of *Mycelia sterilia*, with an intraperitoneal LD_{50} of 7.5 mg/kg in mice [2]. Structure and physical properties proved to be identical to those of **1**. In both cases, the constitution of **1** with the *E*-double bond was correctly assigned. However, the initially reported relative configuration of the insoluble natural product, revealed from NMR and IR data of derivatives, [3] proved to be wrong and was corrected with the synthesis of the chiral γ-lactone [4] derived from **1**, and later confirmed with an X-ray structure of (+)-*N*-acetyl-anhydromyriocin (**9**) [5]. Eventually, **1** was completely characterised with the absolute configuration obtained from the (-)-enantiomer (**10**) of (+)-anhydromyriocin (Fig. (**2**)) [6].

Recently, **1** was isolated from the culture broth of *Melanconis flavovirens*, a non-thermophilic fungus of the class of Pyrenomycetes, [7] and as ISP-1 in *Isaria sinclairii* (also known as *Paecilomyces cicadae* [8]), the imperfect stage of the fungus *Cordyceps sinclairii* [9]. *Cordyceps* is a genus of fungus belonging to the Clavicipitacea family and is parasitic on insects larvae like *Lepidoptera adonata* (butterfly larva) or mature insects. Vegetable wasps and plant worms with the parasitic fungus *Cordyceps sinensis* Sacc. have been used in traditional Chinese Medicine as a nostrum for eternal youth [9].

Myriocin (**1**) is a very potent immunosuppressive agent *in vitro* and *in vivo*, equipotent to tacrolimus (FK506) [10] and approximately 5-100 times more potent than Cyclosporin A (CsA), [11] two clinically prescribed agents. Initial studies have demonstrated that **1** acts mechanistically differently than these commercial immunosuppressive drugs. Compound **1** inhibits the proliferation of a murine cytotoxic T lymphocyte cell line (CTLL-2), but unlike CsA and FK506, not the production of IL-2 [12].

Myriocin binding proteins were identified from CTLL-2. The subunits LCB_1 and LCB_2 were isolated from that cell

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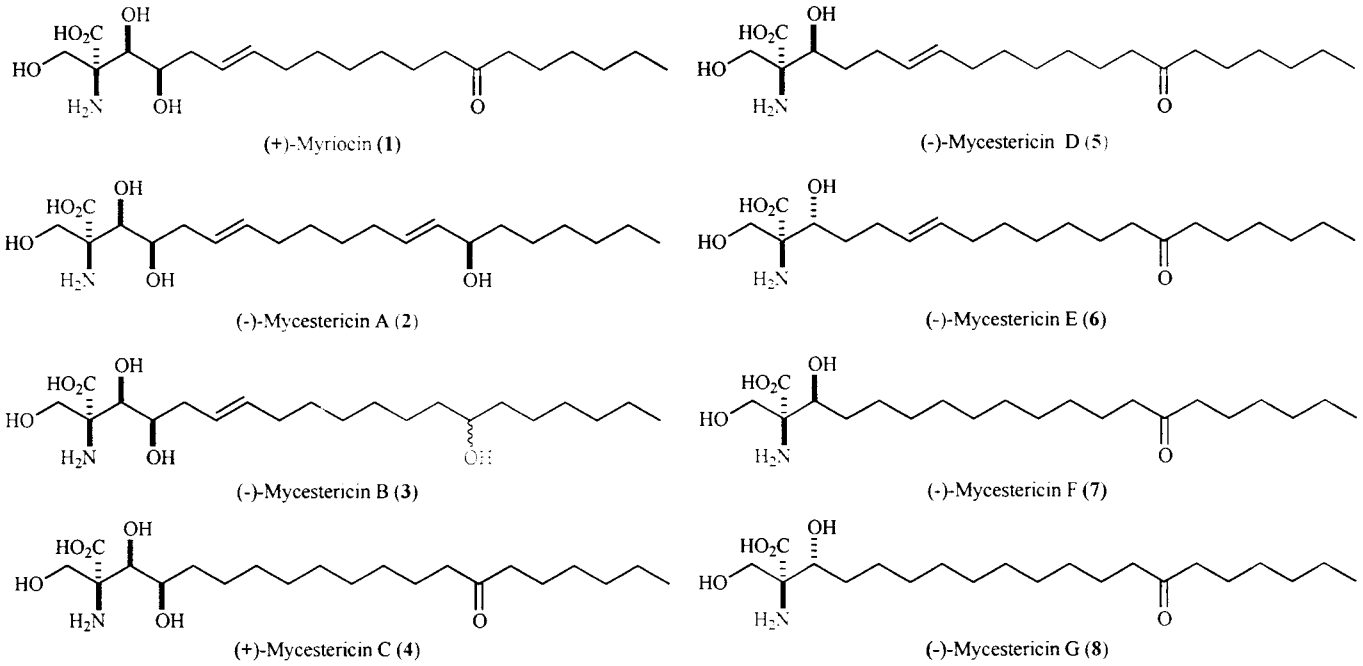


Fig. (1). Metabolites from *Mycelia sterilia.*

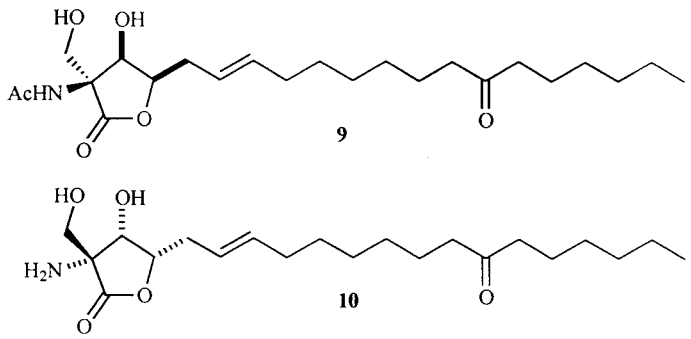


Fig. (2). Andhydromyriocin Derivatives.

line using myriocin derivatives and affinity chromatography, and are mammalian homologues of two yeast proteins that have been genetically linked to sphingolipid biosynthesis [13].

Biosynthetic studies with ^{14}C-labelled substrates confirmed that **1** is assembled in the mould of *Mycelia sterilia* from acetate building blocks and L-serine [14]. Substitution of the α-proton of L-serine with stearoyl-SCoA proceeded with inversion of configuration. Interestingly, the γ-hydroxyl group, the carbonyl group, and the *E*-double bond of **1** were introduced by the enzymatic systems of the mould after the substitution [15].

In CTLL-2-derived microsomes **1** inhibits serine palmitoyltransferase (SPT), an enzyme that catalyses the first step of the sphingolipid biosynthesis, [16] and in *Saccharomyces cerevisiae* the synthesis of the intermediate ceramide is suppressed by **1** by a rapid and specific decrease in the transportation rate of GPI-anchored proteins to the Golgi apparatus, which blocks cell proliferation [17].

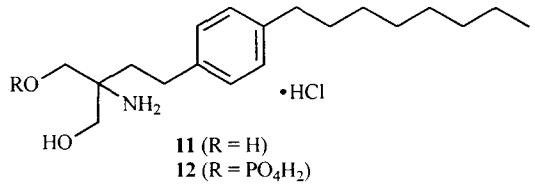
Myriocin (**1**) prevents the accumulation of free sphingoid bases (sphinganine) caused by the mycotoxins fumonisins through inhibition of ceramide synthase, an enzyme in the *de novo* sphingolipid synthesis. The fumonisins B_1, B_2 and B_3 are produced by the fungi *Fusarium moniliforme* and *Fusarium proliferatum* and are found in detectable amounts in most corn containing food and feeds all over the world [18]. Elevated fumonisin concentrations are toxic to animals, but the effect for humans is not clear yet. The accumulation of sphingoid bases in serum, urine, kidney, or liver is correlated with outbreaks of equine leukoencephalomalacia (ELEM) and other farm animal diseases [19]. ELEM is usually mortal to horses, but temporary reduction of sphinganine concentration by **1** induced SPT inhibition could be exploited for promising therapeutic application in animals [20].

2.2. Isolation and Biology of Mycestericins A to G

Re-examination of the culture broth of *Mycelia sterilia* yielded the novel immunosuppressants mycestericin A (**2**) to G (**8**), which are structurally and pharmacologically closely related to **1.** All mycestericins have an equivalent quaternary carbon with identical absolute configuration [21]. Moreover, the polar ends of mycestericin A(**2**), B(**3**), and C(**4**) have identical hydroxyl group substitution patterns as **1**, but **4**

lacks the double bond and **2** and **3** have different alkyl chains. Mycestericin D/E (**5**/**6**) and F/G (**7**/**8**) have only one secondary hydroxyl group and form two pairs of diastereomers (Fig. (**1**)).

In order to reduce toxicity and to improve physicochemical properties, structure-activity relationship studies (SAR) of derivatives of **1** to **8** in mouse allogeneic mixed lymphocyte reaction (MLR) *in vitro* [22] led to the development of symmetrical 2-amino-1,3-propanediols, the essential structures for immunosuppressive activity [23] and, hence, the discovery of the very effective immunosuppressant FTY720 (**11**) (Fig. (**3**)) [24].



11 (R = H)
12 (R = PO_4H_2)

Fig. (3). FTY720 and Phosphorylated Derivative.

Phosphorylated FTY720 (**12**), a sphingosine-1-phosphate (S1P) receptor agonist, protects organ grafts by reducing the recirculation of lymphocytes from lymphatics to blood and inflammatory tissues, and has a different mode of action than the commonly used calcineurin-inhibitors CsA and FK506, or the macrolides rapamycin and RAD. Unlike **1**, this aminoalcohol did not inhibit SPT [25]. Drug candidate **11**, which is currently in phase III clinical trials, is expected to become, in combination with the known calcineurin-inhibitors cyclosporin A (CsA) or FK506, the drug of the future for the therapeutic areas of transplantation and autoimmunity. Aminoalcohol **11** was recently found to effectively suppress cellular infiltration and tissue necrosis in a model of acute myocarditis. Clinical phase II studies with humans showed high efficacy in kidney transplantation for the combination of **11** and the novel macrolide RAD [26].

2.3. Isolation and Biology of Sphingofungins

A series of compounds called sphingofungins (**13** - **18**) were found during a screening program for antifungal inhibitors in the Merck, Sharp & Dohme Laboratories (Fig. (**4**)). Sphingofungin A (**13**) to D (**16**) were isolated from a strain of the thermotolerant fungus *Aspergillus fumigatus*, [27] and sphingofungin E (**17**) and F (**18**) from the fermentation broth of the thermophilic fungus *Paecilomyces variotii* (Fig. (**4**)) [28]. The metabolites **13** to **16** have the same backbone skeleton with congruent configuration with the typical quaternary carbon of myriocin and mycestericins missing. It is possible to transform sphingofungin C (**13**) sequentially into sphingofungin D (**14**). E (**15**) and F (**16**) [29]. The previously unknown absolute configuration of the C14 hydroxyl group of **15** was eventually assigned from ozonolysis products [30].

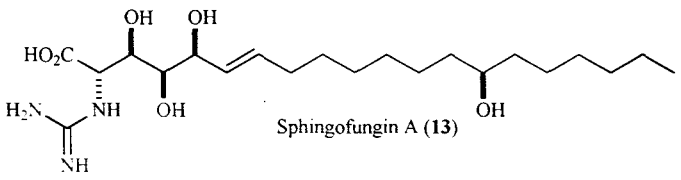
Sphingofungin E (**17**) and F (**18**) resemble myriocin (**1**) and the mycestericins (**2** – **8**). and bear an equivalent quaternary carbon centre with the opposite absolute configuration, respectively.

All sphingofungins are SPT inhibitors [31] and potent antifungal agents against various *Candida* species, but are essentially inactive against filamentous fungi and bacteria. Metabolite **16** is much less potent than **13** to **15** [27,28].

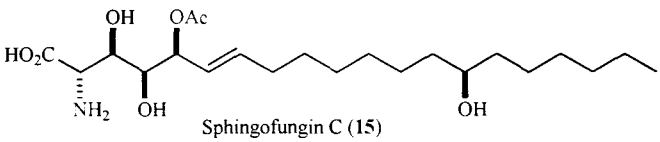
2.4. Isolation of Miscellaneous Sphingosine Analogues

Further examples of sphingosine-related metabolites with a quaternary carbon centre are flavovirin (**19**) and malonofungin (**20**). The antibiotic **19** was found in the pyrenomycete *Melanconis flavovirens* in the course of isolation of **1** and is 5 to 10 times more active than **1** against filamentous fungi, but is ineffective against bacteria [32].

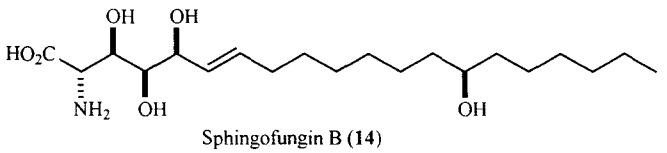
As part of a screening programme for microbial metabolites with growth inhibitory activity against phytopathogenic fungi, the antifungal and antibacterial metabolite **20** was isolated from fermentations of a fungus originating from Jamaican *Panicum maximum* leaves and identified as *Phaeoramularia fusimaculans*. Malonofungin (**20**) shows broad growth inhibitory effects against a range of fungi belonging to the genera Botrytis, Pyricularia,



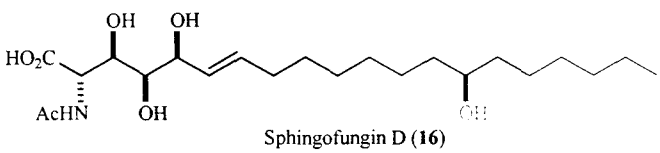
Sphingofungin A (**13**)



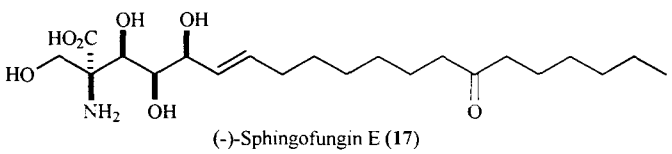
Sphingofungin C (**15**)



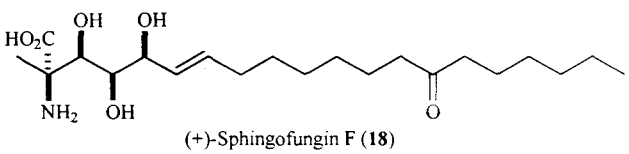
Sphingofungin B (**14**)



Sphingofungin D (**16**)



(-)-Sphingofungin E (**17**)



(+)-Sphingofungin F (**18**)

Fig. (4). Sphingofungins.

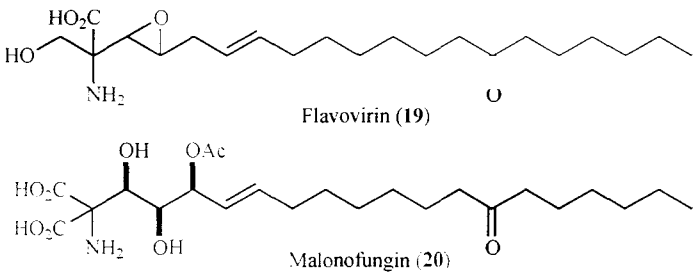
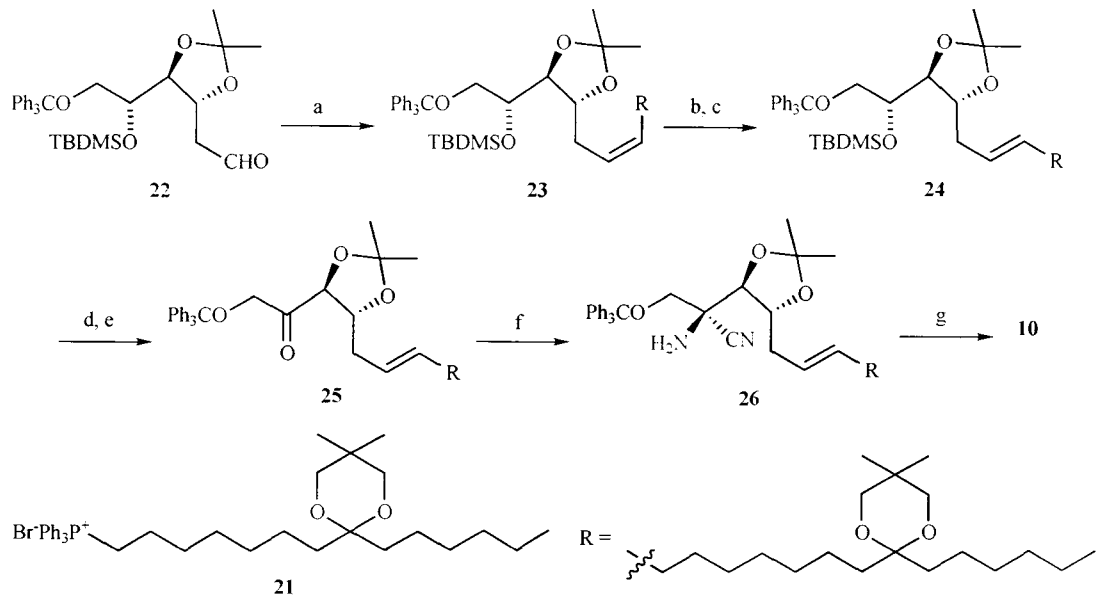


Fig. (5). Miscellaneous Sphingosine Analogues.



Conditions: a) 21, *n*-BuLi, THF; b) *m*-CPBA, cy; c) $LiP(Ph_3)_2$, MeI, THF; d) $(n\text{-}Bu)_4NF{\cdot}H_2O$, PhH, THF; e) $DMSO/Ac_2O$; f) NaCN, NH_4Cl, sat. NH_3/MeOH; g) HCl (g), H_2O/MeOH.

Fig. (6).

Fusarium, and Penicillium, but not against bacteria or yeast (Fig. (5)) [33].

3. TOTAL SYNTHESES OF SPHINGOSINE ANALOGUES

3.1. Introduction

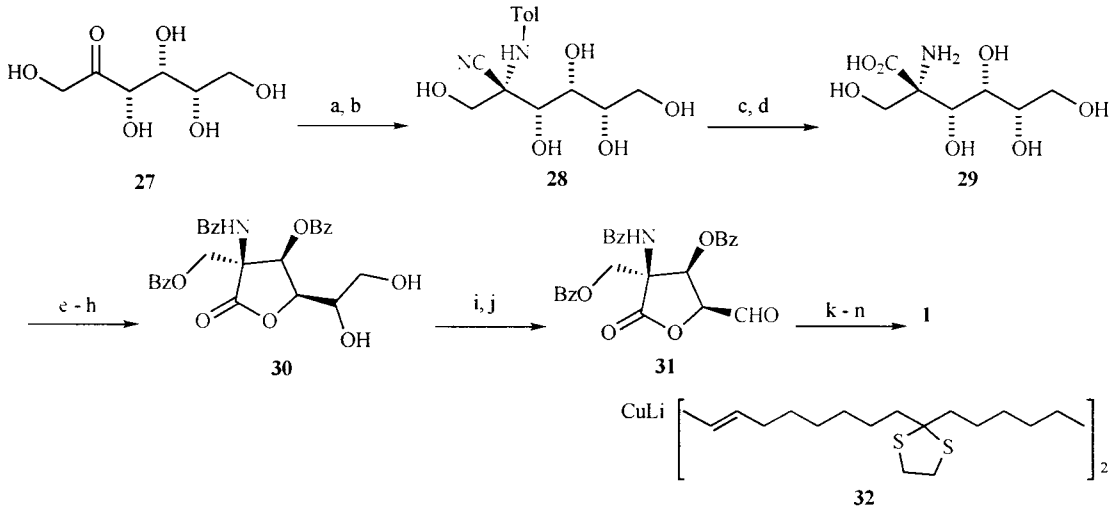
Early synthetic attempts towards myriocin (**1**) were made when Kuo and co-workers synthesised a chiral lactone derived from myriocin and, at the same time, corrected the relative configurations of the 2-amino and 3,4-hydroxyl groups to *syn*, *syn* [4]. Payette and co-workers synthesised the (-)-enantiomer (**10**) of (+)-anhydromyriocin, the γ-lactone derived from **1**, using L-arabinose [6]. They explored a variety of synthetic routes to elongate the pentose moiety and introduced the long alkyl chain tail already at an early stage of the synthesis. However, the Wittig reaction of the alkyl chain ylide **21** with the aldehyde **22** derived from L-arabinose gave predominantly the undesired *Z*-isomer **23** (*Z*:*E* 95:5) in 67% yield , which had to be isomerised to the corresponding *E*-isomer **24** laboriously by the method of Vedejs and Fuchs: epoxidation of the double bond, followed by reaction with lithium diphenylphosphide, quaternisation with methyl iodide, and decomposition of the betaine intermediate gave **24**. The aminocyanation of **25** to install the quaternary amino acid carbon centre of the final product gave an equivalent mixture of diastereomeric cyanoamines, which had to be separated by HPLC. Diastereomer **26** (31% yield) was hydrolysed to yield lactone **10**, the optical antipode of the γ-lactone of **1** (Fig. (6)) [6].

3.2. Total Syntheses of Myriocin

In the following all published total syntheses of natural occurring (+)-myriocin (**1**) are presented in chronological order with focus on the utilised chiral or achiral starting material. The synthesis routes are not in all cases completely described as the present review article concentrates basically on the key parts of each of the described synthetic strategy.

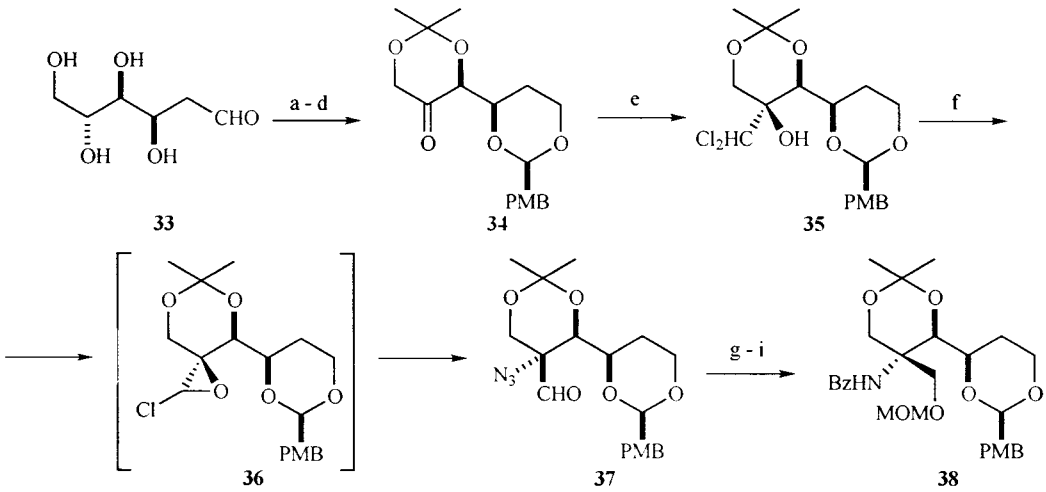
3.2.1. D-Fructose (Scolastico 1982)

The first total synthesis by Scolastico and co-workers used D-fructose (**27**) as the chiral starting material, which was transformed into a diastereomeric mixture of cyanoamines following the Kuhn-procedure. This desired 2*S*-epimer **28** was obtained as a 1:3 mixture of diastereomers in favour of the undesired 2*R*-epimer (80% yield), which was partially converted into soluble **28** by equilibration in $EtOH/H_2O$ in the presence of an excess of liquid hydrogen cyanide. The 2*S*-epimer **28** was hydrolysed and the amino



Conditions: a) *p*-TosNH$_2$; b) HCN, EtOH/H$_2$O; c) HCl; d) H$_2$/Pd, HCl; e) BzCl, Py; f) TEA, MeOH; g) acetone, H$_2$SO$_4$; h) BzCl, Py; i) H$^+$/H$_2$O; j) NaIO$_4$; k) NaBH$_3$(CN), THF; l) TsCl; m) **32**; n) hydrolysis.

Fig. (7).



Conditions: a) DMP, PTS, DMF; b) NaBH$_4$, EtOH; c) *p*-anisyldimethylacetal, *p*-TsOH·H$_2$O; d) DMSO, (COCl)$_2$, TEA, CH$_2$Cl$_2$; e) CH$_2$Cl$_2$, LDA, THF; f) NaN$_3$, 15-crown-5, HMPA; g) NaBH$_4$, EtOH, h) MOMCl, Hünig's base, CH$_2$Cl$_2$; i) H$_2$, Pd/C, EtOH; j) BzCl, Py.

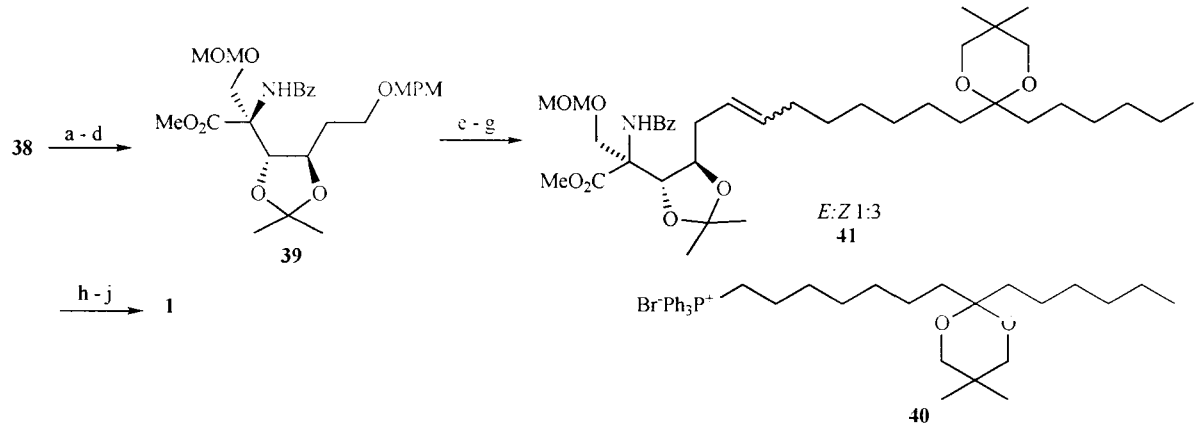
Fig. (8).

tolyl protection cleaved by hydrogenolysis to give 2-amino-2-deoxy-2-hydroxymethyl-D-mannonic acid (**29**). Lactonisation of the amino-benzoylated **29**, temporary acetalisation with acetone and subsequent benzylation of the remaining free hydroxyl groups finally yielded alcohol **30**. Diol **30** was oxidised to the unstable aldehyde **31** with sodium periodate, then, reduced and tosylated. Subsequent nucleophilic substitution of the tosylate with vinyl cuprate **32** in Et$_2$O with HTMP as cosolvent established fully-protected myriocin (28% yield). The lithium *E*-divinylcuprate reagent **32** was generated by standard reactions from 1-morpholinocyclohexene in 9 linear steps. Global deprotection completed the synthesis of **1** in 13 linear steps (Fig. (7)) [34].

3.2.2. 2-Deoxy-D-Glucose (Yoshikawa 1994)

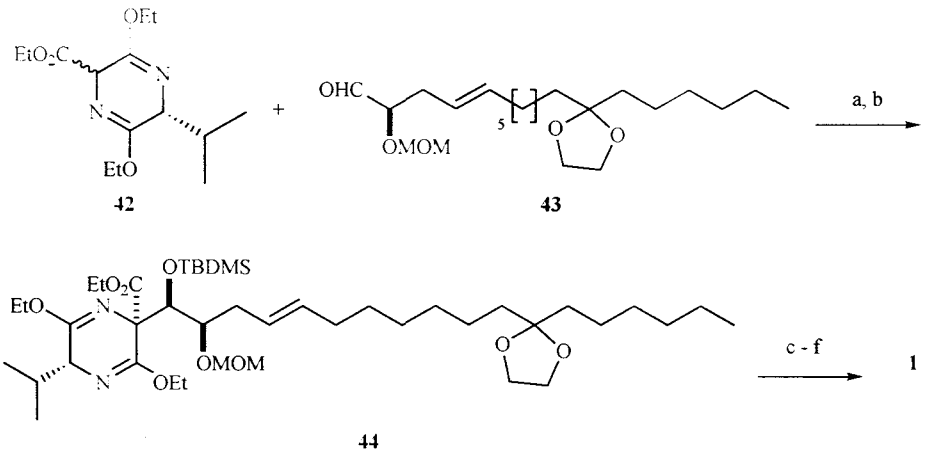
The group of Yoshikawa and co-workers started their stereoselective synthesis with 2-deoxy-D-glucose **33** and used ketone **34** after double acetal protection for a modified Darzen's reaction to obtain the addition product **35** (68% yield for two steps). Treatment with sodium azide in HMPA in the presence of a crown ether as catalyst stereoselectively yielded quantitatively the aldehyde **37** via chloroepoxide **36**. The aldehyde was gradually reduced to the amino acid derivative **38** (Fig. (8)).

The *p*-methoxybenzylidene group (PMB) of **38** was reductively opened to the *p*-methoxybenzyl (MPM) protected primary and free secondary hydroxyl group. Then, migration of the isopropylidene group and oxidation of the liberated primary hydroxyl group to the acid with subsequent esterification with diazomethane yielded the α,α-disubstituted amino acid derivative **39**. The MPM protection-group was cleaved; the resulting primary hydroxyl group oxidised to the corresponding aldehyde, and directly reacted with the phosphonium salt **40**, which was obtained in 8 linear steps from cyclooctanone, [6] in a Wittig reaction



Conditions: a) $NaBH_3CN$, TMSCl, CH_3CN; b) DMSO, $(COCl)_2$, TEA, CH_2Cl_2; c) $NaClO_2$, NH_2SO_3H, dioxane/H_2O; d) CH_2N_2, Et_2O; e) DDQ, CH_2Cl_2/H_2O, f) DMSO, $(COCl)_2$, TEA, CH_2Cl_2; g) **40**, *n*-BuLi, *t*-BuOH/ THF, h) hv, PhSSPh, cy; i) PTS, aq. EtOH; j) 1N NaOH.

Fig. (9).



Conditions: a) $MgBr_2$, TEA, CH_3CN; b) TBDMSOTf, 2,6-Lutidine, CH_2Cl_2; c) DIBALH, CH_2Cl_2; d) HCl, MeOH; e) NaOH, MeOH/H_2O; f) IRC-50 (H^+ type).

Fig. (10).

to give predominantly the undesired *Z*-alkene **41** (*E*:*Z*~1:3) in 65% yield. Photochemical isomerisation in the presence of diphenylsulfide changed the *E*:*Z*-ratio to 4:1 (92% yield). The isomers were separated by HPLC and global deprotection of the *E*-isomer **41** afforded myriocin (**1**) in 20 steps in an overall yield of 5% from **33** (Fig. (**9**)) [35].

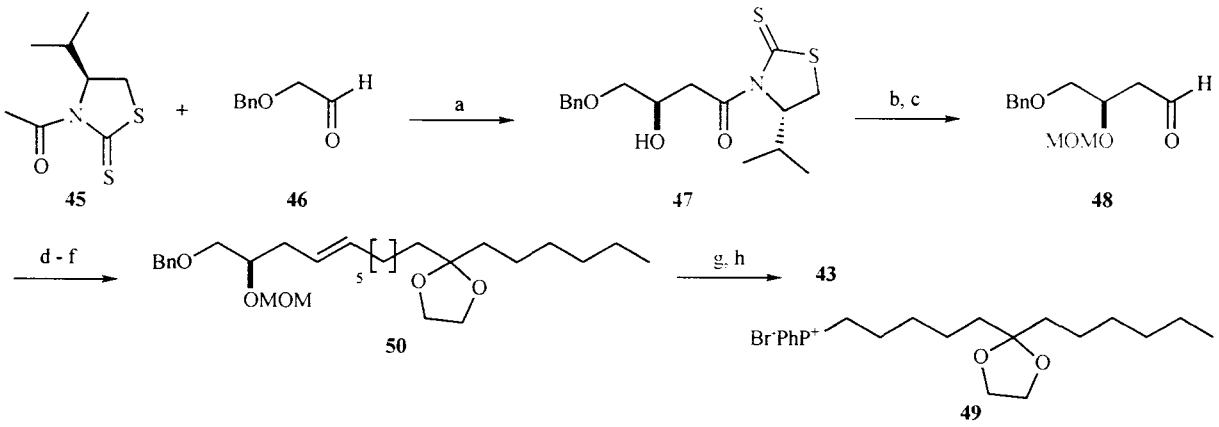
3.2.3. D-Valine (Nagao 1995)

Nagao and co-workers used for the first time highly diastereoselective aldol additions of amino acid derivatives as the key steps in the synthesis of **1**. Unnatural D-valine was transformed into Schöllkopf's bis-lactimether **42** and then added to the linear C-17 aldehyde **43** to yield the fully protected myriocin **44** (47% for two steps). Reduction of the bis-lactimether and stepwise removal of protecting groups gave **1** in 13 linear steps (Fig (**10**)).

The long-chain aldehyde **43** was synthesised starting with a highly diastereoselective aldol addition of the enolate of thiazolidinethione **45** to aldehyde **46** to give the chiral aldehyde **47** in 93% *de* and 79% yield. Subsequent reductive cleavage of the thiazolidinethione auxiliary in **47** yielded the aldehyde **48** with the right absolute configuration of the protected secondary hydroxyl group. Highly *E*-selective Schlosser-type Wittig reaction of the phosphonium salt **49** with **48** then provided the protected diol **50** (*E*:*Z* 96:4; 82% yield), which was, after deprotection of the primary hydroxyl group, oxidised to aldehyde **43** (Fig (**11**)) [36].

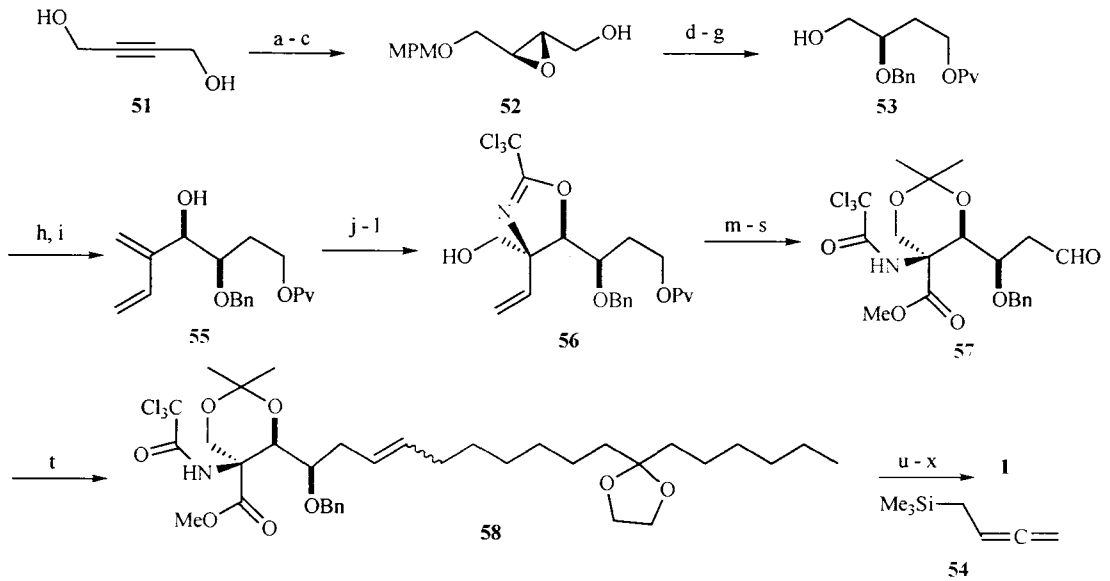
3.2.4. Achiral Starting Material (Hatakeyama 1997)

Recently, Hatakeyama and co-workers published an enantioselective synthesis based on the chemistry of 1-trimethylsilylbuta-2,3-dienes. The utilisation of achiral starting material required stereoselective reaction steps. Catalytic Katsuki-Sharpless asymmetric epoxidation of the *E*-allylic alcohol obtained from reduction of the mono protected 2-butyn-1,4-diol **51** gave the enantiomerically pure epoxide **52** (92% yield). Reduction of **52** followed by a series of protection and deprotection steps gave alcohol **53**. Addition of 1-trimethylsilylbuta-2,3-diene (**54**) to the aldehyde obtained by Swern oxidation of **53** occurred in a highly diastereoselective manner in 78% yield over two steps. The ensuing dienol **55** was diastereoselectively epoxidised and converted to an epoxytrichloroacetamidate. Then, Lewis-acid catalysed cyclisation with inversion to



Conditions: a) $Sn(OTf)_2$, *N*-ethylpiperidine, CH_2Cl_2; b) $MeOCH_2Cl$, Hünig's Base, CH_2Cl_2; c) DIBALH, THF; d) PhLi, **49**, THF/Et_2O;e) PhLi, f) Silica gel; g) Li, liq. NH_3, THF; h) DMSO, $(COCl)_2$, CH_2Cl_2.

Fig. (11).



Conditions: a) PMBCl, KOH, DMSO; b) Red-Al®, Et_2O; c) diisopropyl L-tartrate, $Ti(O\textit{i}\text{-}Pr)_4$, *t*-$BuO_2H$, 4 Å MS, CH_2Cl_2; d) Red-Al®, THF; e) *t*-BuCOCl, Py, CH_2Cl_2; f) TMSCl, TEA, THF; g) PhCHO, TESH, TMSOTf, CH_2Cl_2; h) $(COCl)_2$, DMSO, TEA, CH_2Cl_2; i) **54**, $TiCl_4$, CH_2Cl_2; j) $VO(acac)_2$, *t*-BuO_2H, CH_2Cl_2; k) CCl_3CN, DBU, 4 Å MS, CH_2Cl_2; l) Et_2AlCl, CH_2Cl_2; m) HCl, THF; n) $(MeO)_2CMe_2$, PTS, CH_2Cl_2; o) O_3, CH_2Cl_2, then Me_2S; p) $NaClO_2$, NaH_2PO_4, 2-methyl-2-butene, *t*-BuOH/H_2O; q) CH_2N_2, Et_2O; r) NaOMe, MeOH; s) $(COCl)_2$, DMSO, TEA, CH_2Cl_2; t) **49**, *n*-BuLi, THF; u) NaOH/MeOH; v) Li, THF, liq. NH_3; w) HCl/MeOH; x) Ac_2O, DMPA, Py; y) hv, PhSSPh, PhH; z) NaOH/MeOH, then Amberlite®IRC-76.

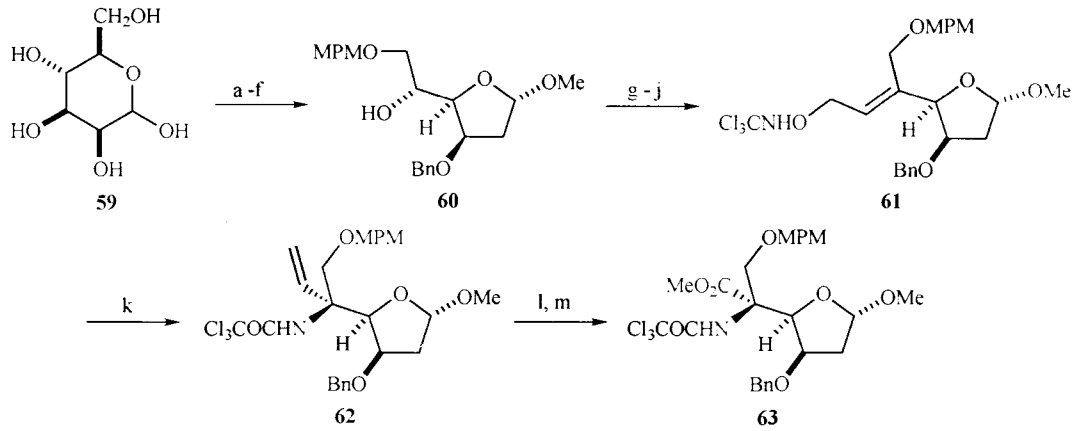
Fig. (12).

oxazoline **56** stereoselectively established the amino group in 69% yield from **55**. Acid hydrolysis and sequential diol protection, ozonolysis, oxidation, esterification and Swern oxidation gave the aldehyde **57**. A Wittig reaction joined **57** with the phosphonium salt **49** (Fig. (**11**)), to yield the fully protected **58** as a 92:8 *Z*:*E*-mixture (48% yield). Conversion of the modified **58** into the corresponding γ-lactone and photochemical isomerisation as in Yoshikawa's synthesis completed, after global deprotection, the synthesis for **1** in 26 linear steps (Fig. (**12**)) [37].

3.2.5. D-Mannose (Chida 2001)

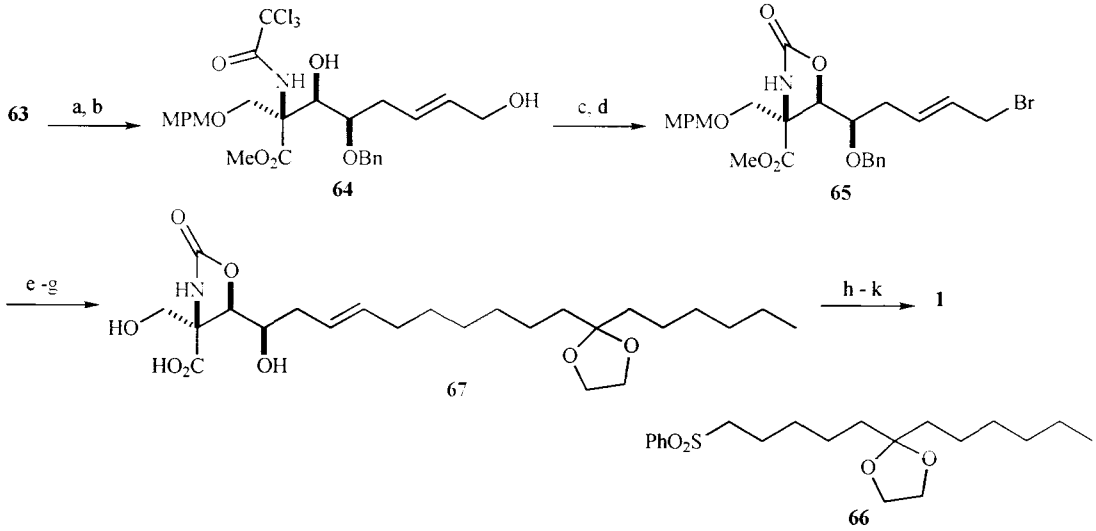
Chida and co-workers used aldohexoses for a long linear total synthesis of **1** and, similarly, sphingofungin E (**17**, see below), where an Overman rearrangement was the key reaction. Compound **1** was synthesised starting from D-mannose (**59**). The α-methyl furanoside **60** was obtained in six steps and 61% overall yield. After Swern oxidation, Horner-Wadsworth-Emmons olefination under the Masamune-Roush conditions (*E*:*Z* 15:1) and reduction, the allylic trichloroacetimidate **61** was subjected to Overman rearrangement to give the allylic amide **62**. Ozonolysis, followed by oxidation and esterification afforded intermediate **63** (Fig. (**13**)).

Isomerically pure *E*-allyl alcohol **64** was obtained by a Wittig reaction of acetal **63** and the stabilised phosphor ylide derived from ethyl bromoacetate (53% yield over two steps). The coupling of the alkyl sulfone **66** with the highly functionalised allyl bromide **65** yielded, after saponification and subsequent Birch reduction, the carboxylic acid **67**.



Conditions: a) H_2SO_4/acetone; b) $(Me_2N)_3P/CCl_4$ then Li, NH_3, THF; c) NaH; BnBr; d) $Hg(OAc)_2$, THF/H_2O, then KI, $NaBH_4$, THF/H_2O; e) $AcOH/H_2O$, then AcCl, MeOH; f) *n*-Bu_2SnO, tol, then MPMCl, CsF, DMF; g) $(COCl)_2$, DMSO, CH_2Cl_2, TEA; h) $(MeO)_2$-$POCH_2CO_2Me$, LiBr, DBU, CH_3CN, 90% from **60** (*E*:*Z* 15:1); i) DIBALH, tol; j) Cl_3CCN, DBU, CH_2Cl_2; k) K_2CO_3, *o*-xylene, 90% for 2 steps; l) O_3, CH_2Cl_2, then Me_2S; m) $NaClO_2$, NaH_2PO_4, $HOSO_2NH_2$, *t*-BuOH/H_2O, then $TMSCHN_2$, MeOH.

Fig. (13).



Conditions: a) aq. HCl/THF, then $Ph_3P{=}CHCO_2Et$, tol, 71%; b) DIBALH, THF/tol, 75%; c) DBU, CH_2Cl_2; d) MsCl, TEA, CH_2Cl_2, then LiBr, acetone; e) *n*-BuLi, THF, then **66**, 80%; f) LiOH, aq. MeOH; g) Li, liq. NH_3, THF; h) aq. HCl/THF; i) aq. NaOH/MeOH; j) Ac_2O, Py; k) aq. NaOH/MeOH.

Fig. (14).

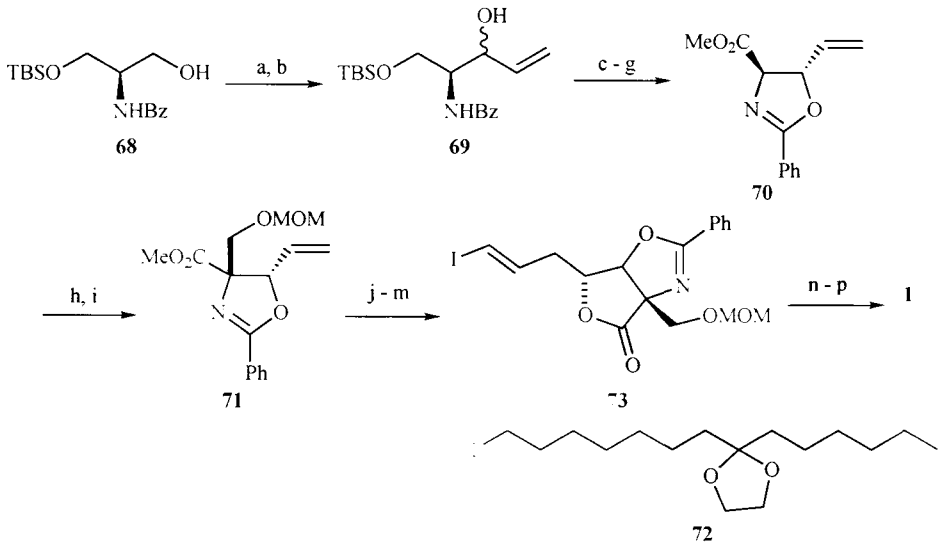
Hydrolysis of the carbamate and the acetal protection group in **67** yielded, via anhydromyriocin, **1** in 24 steps and 5% overall yield (Fig. (**14**)) [38].

3.2.6. L-Serinol (Ham 2002)

Most recently, Ham and co-workers published a convergent total synthesis employing the diastereoselectively formed oxazoline **70** as a key intermediate, which was prepared from L-serinol derivative **68**. Oxidation of **68** and subsequent Grignard reaction afforded a 1:1 *syn*/*anti*-mixture of allyl alcohol **69** in 70% yield over two steps. Acetyl protection of **69** followed by highly diastereoselective Pd(0)-catalysed intramolecular cyclisation yielded only *trans*-oxazoline **70** in 60% yield from **69**. Hydroxymethylation of the α-amino carbon of **70** gave the desired *anti* adduct **71** in a diastereomeric ratio of 20:1 in favour of the desired isomer in 52% for two steps. Ozonolysis of the terminal double bond and $MgBr_2$-promoted allylic stannane addition of the obtained unstable aldehyde was followed by another ozonolysis and iodomethylation to give the vinyl iodide **73** in 51% yield over four steps. Negishi-coupling of alkyl chain **72** as an organozinc reagent with the halide **73** and final hydrolysis finished the synthesis in 19 steps from **68** and 4.6% overall yield (Fig. (**15**)) [39].

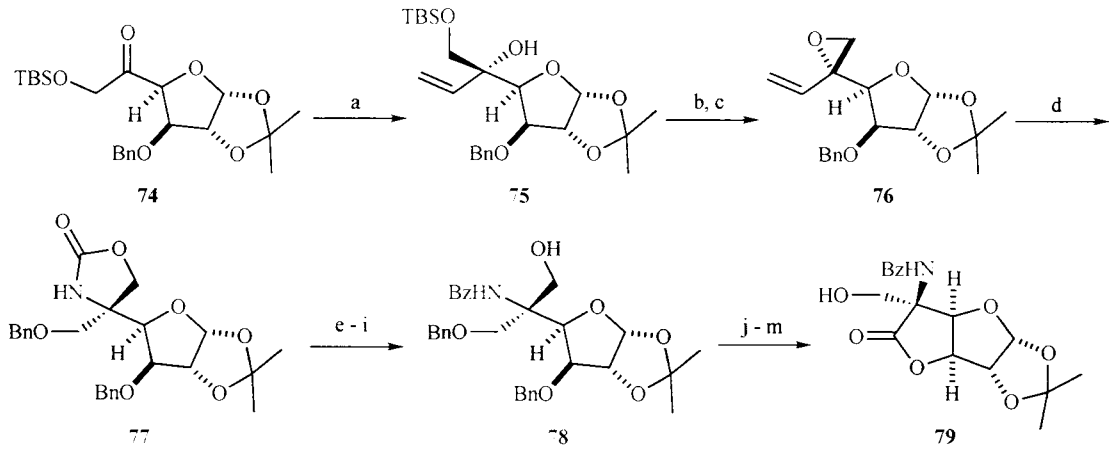
3.3. Formal Syntheses of Myriocin from D-Glucose (Rao 1993, Olesker 1994)

The first and longer formal synthesis of (**1**) by Rao and co-workers utilised the Trost Pd-catalysed *cis*-hydroxyamination of chiral vinyl epoxides as the key step. The 5-ulose derivative **74**, readily obtained from D-glucose, was alkylated with vinylmagnesium bromide. The alkene **75** was selectively converted to the epoxide **76** under Mitsunobu conditions. The resulting vinyl epoxide **76** underwent *cis*-hydroxyamination with *p*-methoxyphenyl-isocyanate and oxazolidinone **77** was obtained as the single product in 76%



Conditions: a) Dess-Martin periodinane, CH_2Cl_2; b) $CH_2CHMgBr$, THF, 70% for two steps; c) Ac_2O, Py, CH_2Cl_2; d) $Pd(PPh_3)_4$, K_2CO_3, CH_3CN, 60%; e) TBAF, THF; f) $RuCl_3$, $K_2S_2O_8$, NaOH, CH_3CN; g) CH_2N_2, Et_2O; h) HCHO, KHMDS, HMPA, THF; i) MOMCl, Hünig's Base, CH_2Cl_2; j) O_3, MeOH, then Me_2S. k) $Bu_3SnCH_2CH{=}CH_2$, $MgBr_2Et_2O$, CH_2Cl_2, 82% for two steps; l) O_3, MeOH, then Me_2S. m) $CrCl_2$, CHI_3, THF, 63% for two steps; n) **72**, *t*-BuLi, then $ZnCl_2$, $Pd(PPh_3)_4$, THF, 78%; o) HCl, THF, p) NaOH.

Fig. (15).



Conditions: a) $CH_2CHMgBr$, THF, 80%; b) Bu_4NF, THF; c) PPh_3, DEAD, tol; d) TosNCO, $Pd(PPh_3)_4$, THF, $(CH_3)_2CHO)_3P$, 76%; e) O_3, CH_2Cl_2, $NaBH_4$; f) NaH, BnBr, THF; g) CAN, CH_3CN/H_2O; h) NaOH, THF; i) Bz_2O, MeOH; j) $RuCl_3$, $NaIO_4$, H_2O, CCl_4, CH_3CN; k) CH_2N_2, Et_2O; l) H_2/Pd, MeOH, 35 psi, 85%; m) NH_3, MeOH.

Fig. (16).

yield. After a series of transformations the key intermediate **79** for both of the formal syntheses was achieved in 13 steps from **74**. The final known aldehyde **31** (Fig. (7)) was obtained after few transformations (Fig. (**16**)) [40].

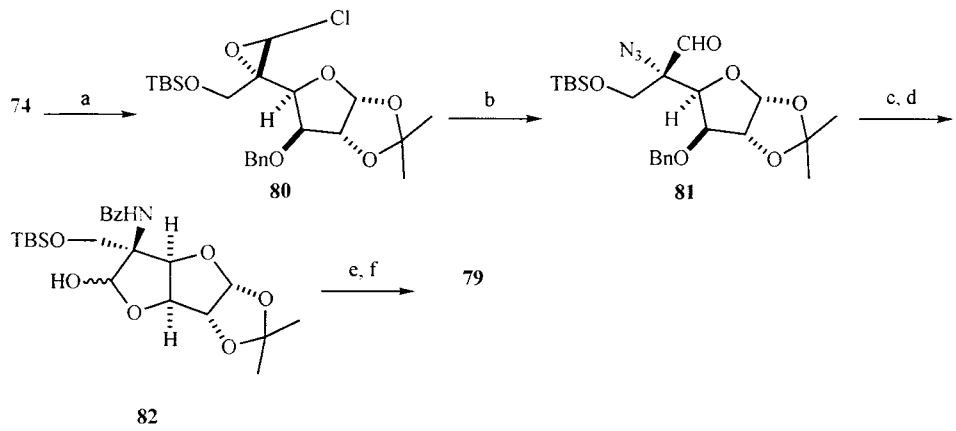
The related formal synthesis by Olesker and co-workers used the same 5-ulose derivative as the starting material, but reduced the number of reaction steps to 6 for the route from **74** to **79** (Fig. (**17**)) [41]. Instead of the alkylation/*cis*-hydroxyamination sequence, the key intermediate **79** was obtained through a highly stereoselective Darzen's condensation with chloromethyl *p*-tosylsulfone. An S_N2 reaction of the α,β-epoxy chloride **80** in the presence of azide ion gave the α-azido aldehyde **81** with the desired (*S*)-configuration in 55% yield over 2 steps.

3.4. Total Syntheses of Mycestericins

The scientific focus in total syntheses of mycestericin A to G (**2** - **8**) was mainly on the two pairs of diastereomers, D/F (**5**,**7**) and E/G (**6**,**8**), since **7** and **8** are easily obtained after reduction of the double bond of **5** and **6** respectively. Here, we present the published syntheses in chronological order.

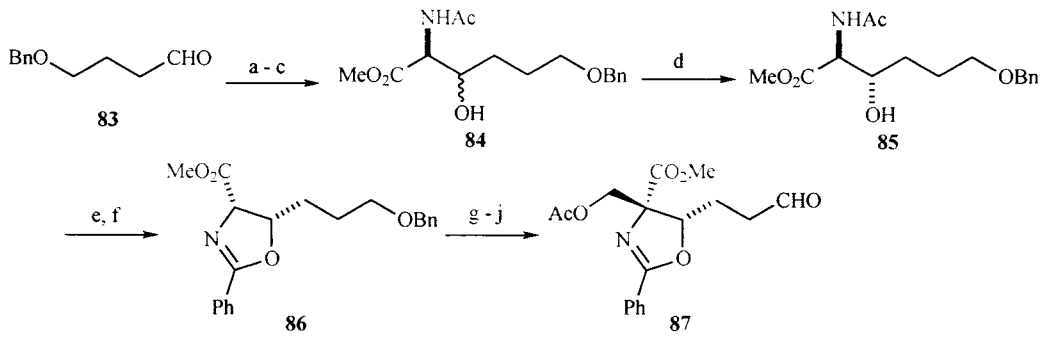
3.4.1. L-Threonine Aldolase (Mycestericin D 1996 and F 2000)

An enzymatic aldol reaction was used in the synthesis of mycestericin D (**5**) and F (**7**). L-Threonine aldolase from *Candida humicola* catalysed the reaction of



Conditions: a) LDA, CH_2Cl_2, THF; b) NaN_3, DMPU, 15-crown-5-ether, 55%; c) H_2/Pd, abs. EtOH; d) BzO_2, MeOH, 46% for two steps; e) $(COCl)_2$, DMSO, TEA, CH_2Cl_2, 86%; f) TBAF, THF, 97%.

Fig. (17).



Conditions: a) L-Threonine aldolase, glycine; b) Ac_2O, TEA, CH_2Cl_2; c) CH_2N_2, Et_2O; d) Chromatography; e) HCl, MeOH; f) PhC(NH)OMe, Et_2O/H_2O; g) DBU, $(CH_2O)_n$, DMF; h) Ac_2O, Py, 83% for two steps; i) $AlCl_3$, NaI, CH_3CN, 88%; j) PCC, CH_2Cl_2, 92%.

Fig. (18).

benzyloxybutanal **83** and glycine to a mixture of *threo/erythro* products **84** with high diastereoselectivity in favour of the *erythro* product. Pure **85** was obtained after derivatisation and chromatography and used for the subsequent formation of *cis*-oxazoline **86** with methyl benzimidate in 76% yield and with >99% *ee*. The addition of formaldehyde occurred stereoselectively and cleavage of the benzyl ether with $AlCl_3$ and oxidation of the primary alcohol stereoselectively gave the aldehyde **87** in 67% yield from **86** (Fig. (**18**)). Addition of the known phosphonium salt **49** (Fig. (**11**)) and subsequent photochemical isomerisation [35] established the backbone of the mycestericin D (**5**) and F (**7**). Finally, global deprotection (**5**) and double bond reduction (**7**) gave the natural products [42].

3.4.2. L-Serine (Mycestericin E and G 1995)

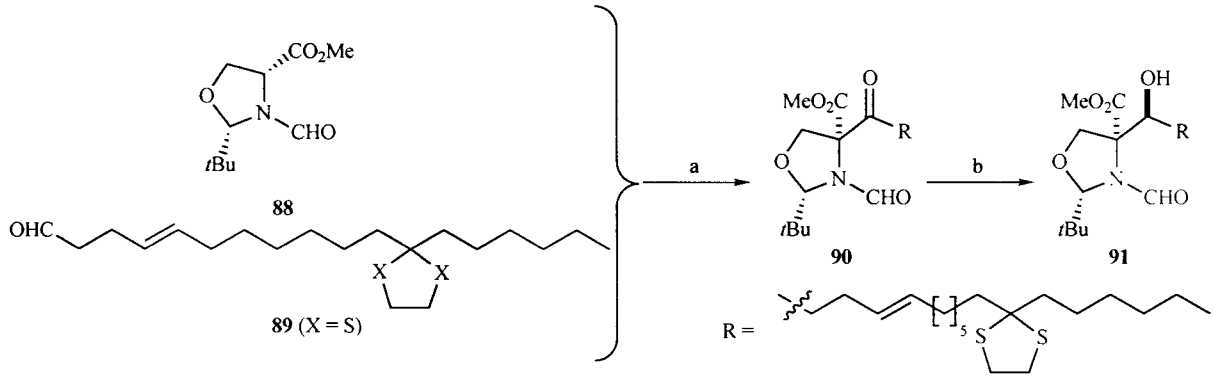
Mycestericins E (**6**) and G (**8**) were synthesised with the methodology of self-regeneration of stereocentres (SRS) for the formation of the quaternary stereocentres [43]. Addition of the enolate of oxazolidine **88**, obtained selectively in three steps from unnatural D-serine, to acid chloride **89** gave the β-keto ester **90** in 46% yield. Reduction with $NaBH_4$ resulted in a 85:15-mixture of the 1'*R* and 1'*S* hydroxyl esters, which could be separated by chromatography. The compounds were unstable on silica gel and, especially, the undesired *S*-isomer decomposed easily. Mycestericin E (**6**) was obtained after global deprotection of *R*-isomer **91** and reduction of the double afforded mycestericin G (**8**) (Fig. (**19**)) [44].

3.4.3. Achiral Starting Material (Mycestericin E 2001)

A second enantioselective synthesis of mycestericin E (**6**) used a *Cinchona* alkaloid-catalysed asymmetric Baylis-Hillman reaction where hexafluoroisopropyl acrylate was added to aldehyde **89** (X = O) in the presence of catalyst **92** (yield 47%) [45]. The resulting allylic hydroxyester **93** was epoxidised and converted to the epoxytrichloroacetimidate **94** with trichloroacetonitrile in the presence of DBU in 42% yield from **93**. The nitrogen was stereoselectively installed by a Lewis-acid-promoted cyclisation of **94**, a methodology already used by the same group in the synthesis of **1** (Fig. (**20**)) [37].

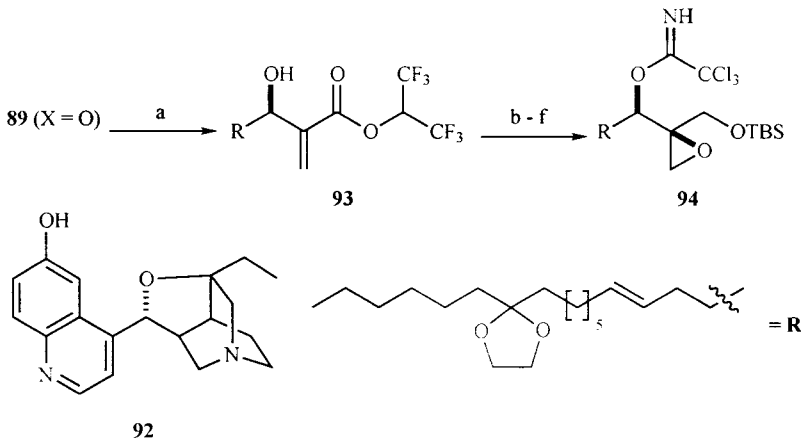
3.5. Total Syntheses of Sphingofungins

Only the sphingofungins E (**17**) and F (**18**) have the characteristic quaternary carbon present in myriocin (**1**) or mycestericins (**2** - **8**). The remaining sphingofungins A to D (**13** – **16**) have the same substitution pattern as the sphingosines. A formal synthesis of sphingofungin D (**16**)



Conditions: a) CH_3CH_2COCl, LDA, THF, 46%; b) $NaBH_4$, MeOH,70%.

Fig. (19).



Conditions: a) **92**, $CH_2CHCO_2CH(CF_3)_2$, DMF/CH_2Cl_2; b) NaOMe, MeOH, then Dowex-50 (H^+), 95%; c) $Ti(Oi\text{-}Pr)_4$, $t\text{-}BuO_2H$, 4 Å MS, CH_2Cl_2, 73%; d) $NaBH_4$, THF/MeOH, 79%; e) TBSCl, DMAP, TEA, CH_2Cl_2, 95%; f) DBU, CCl_3CN, CH_2Cl_2, 81%.

Fig. (20).

was already published in 1994 [46]. It was shown that A (**13**), B (**14**), and D (**16**) are synthetically accessible from the most abundant sphingofungin C (**15**) [29].

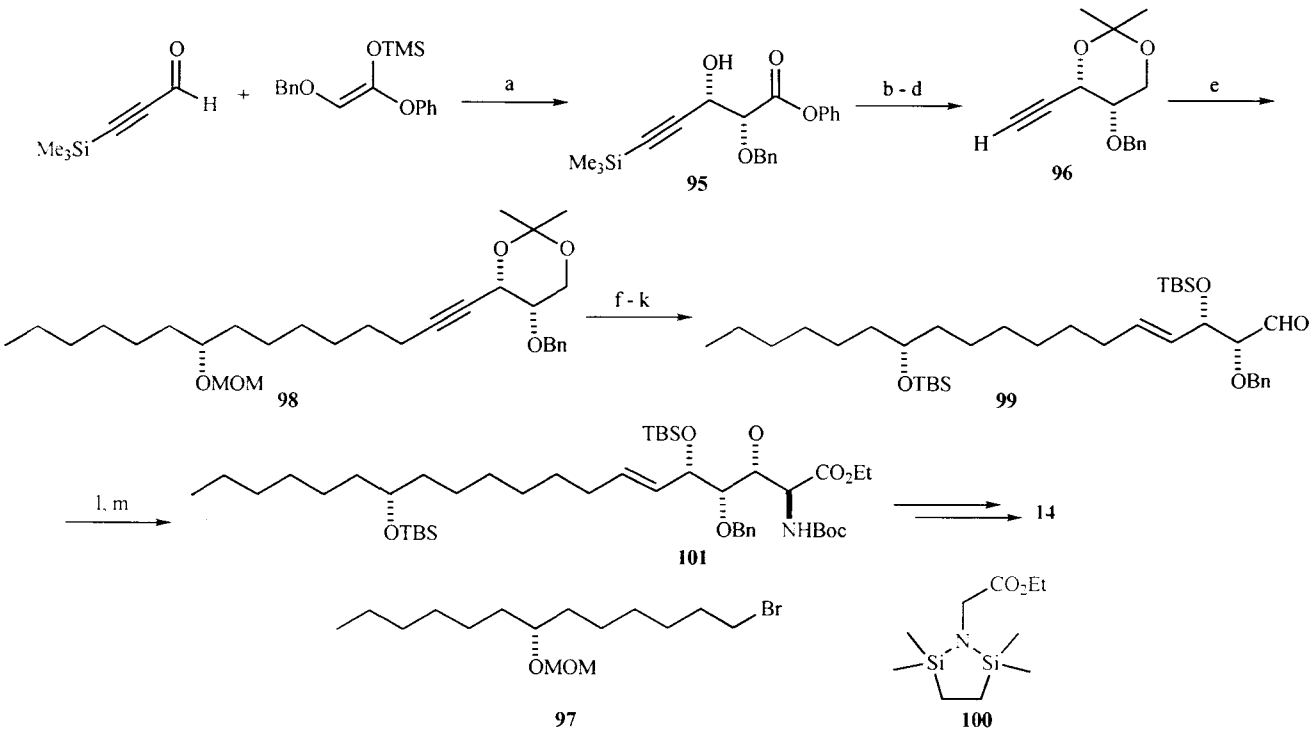
3.5.1. Kobayashi (Sphingofungin B 1996 and F 1997)

Kobayashi and co-workers published the first asymmetric total synthesis of sphingofungin B (**14**) in 1996. The chiral Lewis-acid-controlled (CLAC) aldol addition of 2-benzyloxy-1-trimethylsiloxy-1-phenoxyethene and trimethylsilylpropynal afforded the phenyl ester **95** in a *syn/anti* ratio of 97:3 (87% yield). In general, it is possible to synthesise all four stereoisomers of **95** and, hence, also of **14**, based on the CLAC synthesis. Reduction of the phenyl ester in **95** and protection of the resulting diol was followed by desilylation and acetylene **96** was obtained. Compound **96** was coupled with the bromide **97** to afford ether **98** in 86% yield. An initial Sn(II)-catalysed asymmetric aldol reaction based on the CLAC methodology was also the initial step in the synthesis of **97**. Ether **98** was hydrolysed and converted in 5 steps to the aldehyde **99** (63% yield from **98**), which in turn underwent the key aldol reaction with the zinc enolate of **100** to give the desired adduct in 93% yield and 67% *ds*. Final hydrolysis, and sequential removal of the protecting groups afforded **14** in 17 linear steps and 12% overall yield from trimethylsilylpropynal (Fig. (**21**)) [47].

The first total synthesis of sphingofungin F (**18**) in 1997 was based on an analogous methodology, but the formation of the quaternary amino acid carbon was accomplished utilising Schöllkopf's bis-lactimether method [48]. Here, the diastereoselective Sn(II)-catalysed aldol reaction of bis-lactimether **103**, an alanine equivalent, and keto-aldehyde **102**, obtained after oxidation of **99**, gave the major diastereomer **104** (*dr* 70:25:5:0) in 83% yield and, eventually, **18** was obtained after a series of deprotection steps (Fig. (**22**)) [49].

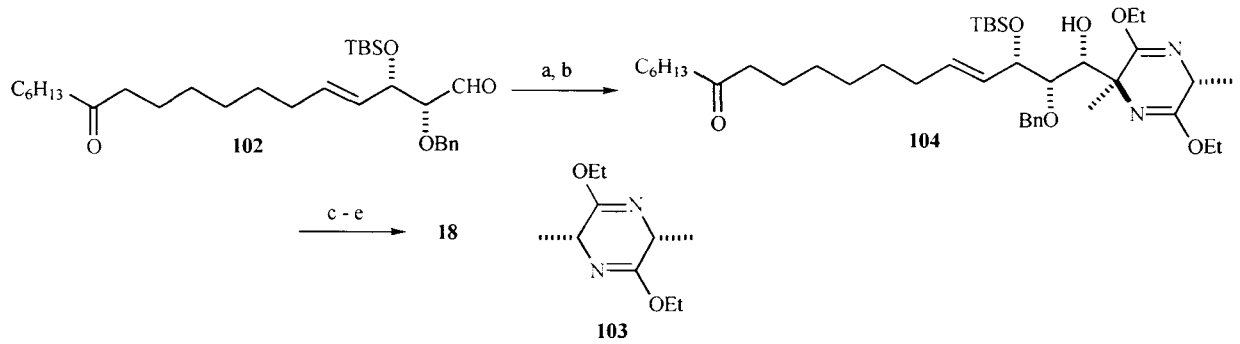
3.5.2. Lin (Sphingofungin E 2001 and F 2000)

An efficient and convenient approach to sphingofungin F (**18**) started from L-(+)-tartaric acid and employed the methodology developed by Hatakeyama and co-workers used for the total synthesis of mycestericin E (**6**) [45]. Aldehyde **105** underwent a Wittig reaction with ylide **106** to give the α,β-unsaturated ester **107**. After reduction of the methyl ester in **107**, a Katsuki-Sharpless asymmetric epoxidation (AE) gave a 3:1 mixture of the desired diastereomer **108** (73% yield), which was joined with the known phosphonium salt



Conditions: a) $Sn(OTf)_2$, (*R*)-1-methyl-2-[(*N*-1-naphtylamino)methyl]pyrrolidine, SnO, C_2H_5CN, 87%, 91% *ee* (*syn*); b) DIBALH, CH_2Cl_2, 83%; c) DMP, PTS, DMF, 97%; d) TBAF, CH_2Cl_2, 89%; e) 97, BuLi, THF, HMPA, 87%; f) HCl, MeOH, 96%; g) MMTrCl, TEA, DMAP, CH_2Cl_2; h) LAH, THF; i) TBSCl, imid., DMF, 98% for three steps; j) HCO_2H, Et_2O, 83%; k) $(COCl)_2$, DMSO, TEA, CH_2Cl_2, 95%; l) LDA, **100**, $ZnCl_2$, THF, 93%, 67% *ds*; m) $(Boc)_2O$, CH_2Cl_2.

Fig. (21).



Conditions: a) **103**, BuLi, $SnCl_2$, THF, 83%; b) TBAF, THF, 94%; c) PTS, THF/H_2O; d) NaOH, $MeOH/H_2O$ 58% for two steps; e) BCl_3, CH_2Cl_2, 72%.

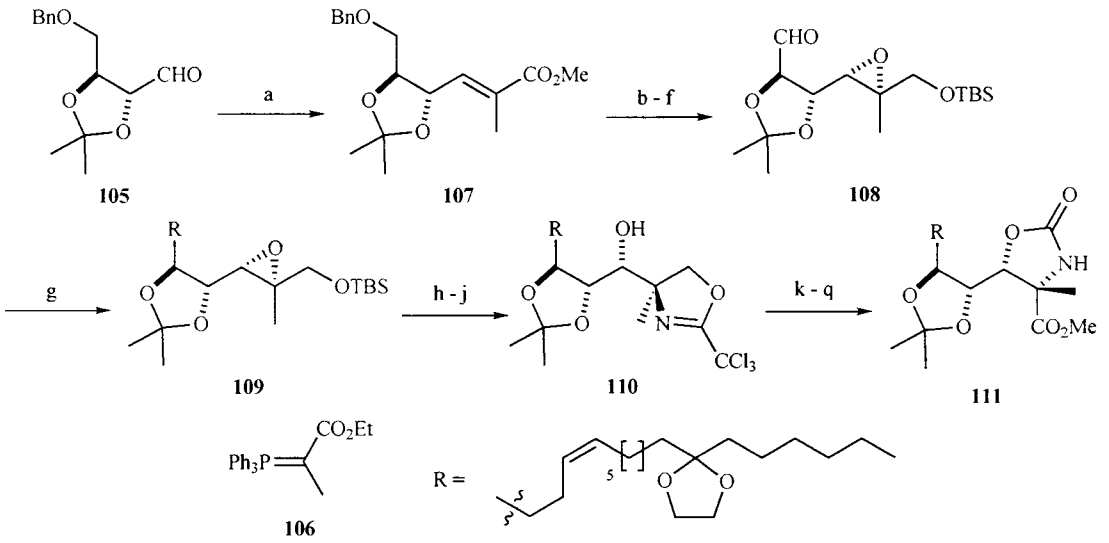
Fig. (22).

49 (Fig. (**11**)) in another Wittig reaction to yield **109** (85%). Application of the Hatakeyama procedure gave the oxazoline **110**, which was transformed, via ester **111** and photoisomerisation [35] with phenyl disulfide, to **18** in total 22 linear steps and 3.7% overall yield (Fig. (**23**)) [50].

The same method was used for the first total synthesis of sphingofungin E (**17**), which was achieved in 19 steps with 8% overall yield (Fig. (**24**)) [51]. Here, the known aldehyde **105** underwent a Baylis-Hillman reaction with methylacrylate to afford the methyl ester **112** in 70% yield. Dihydroxylation with substrate control and protection of the primary hydroxyl group gave the 2,3-*syn*-triol **113** as the sole product. Mesylation of the secondary hydroxyl group in **113** and ring closure was followed by reduction and protection to afford epoxide **114**. Then, Hatakeyama's methodology produced oxazoline **115**, with all stereogenic centres of the final product, which was transformed to the key intermediate carbamate **116**.

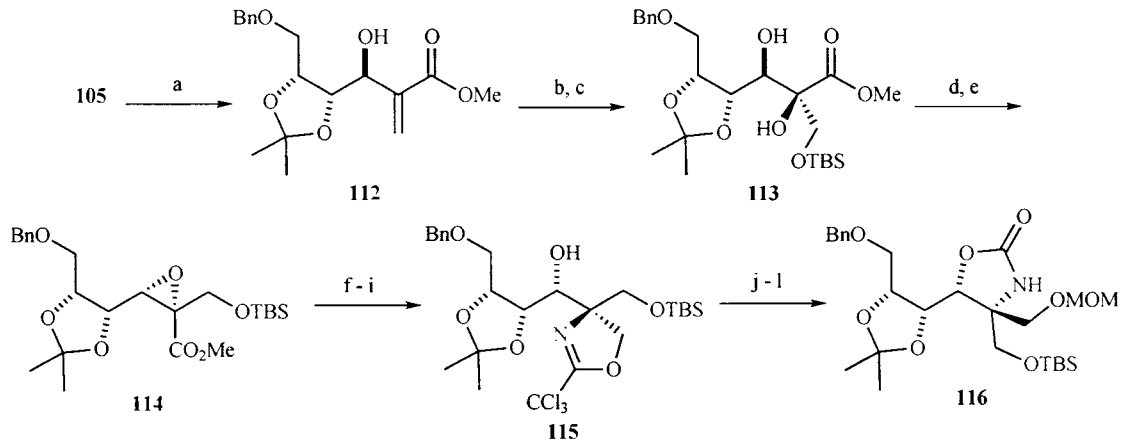
3.5.3. Trost (Sphingofungin E 2001 and F 1997)

The utilisation of *gem*-diacetates as carbonyl surrogates by Trost and co-workers presented two more asymmetric total syntheses of sphingofungins E (**17**) and F (**18**). The key step to establish two of the four stereocentres is a Pd(0)-catalysed asymmetric allylic alkylation reaction (AAA) of the *gem*-diacetate **117** with azlactone **118**. The desired alkylation product **120** is obtained as the major diastereomer in a ratio of 11:1 in 70% isolated yield. Alkylated azlactone **120** was converted to the corresponding methyl ester and dihydroxylation exclusively gave the lactone **121** in 86%



Conditions: a) **106**, CH_2Cl_2 (*E*/*Z* 17:1); b) DIBALH, CH_2Cl_2, 90%; c) TBHP, L-(+)-DIPT, Ti(O*i*-Pr)$_4$, 4 Å MS, CH_2Cl_2, 73%; d) TBSCl, TEA, DMAP, CH_2Cl_2, 80%; e) Pd(OH)$_2$/C, H_2, EtOAc/MeOH, 97%; f) $(COCl)_2$, DMSO, TEA, CH_2Cl_2, 87%; g) **49**, BuLi, THF, 85%; h) TBAF, THF, 95%; i) Cl_3CCN, DBU, CH_2Cl_2; j) BF_3Et_2O, CH_2Cl_2, 80% for two steps; k) $CO(OCCl_3)_2$, Py, CH_2Cl_2; l) K_2CO_3, MeOH, 82% for two steps; m) PDC, DMF; n) CH_2N_2, Et_2O, 78% for two steps; o) PhSSPh, hv, cy. 1,4-dioxane, 95%; p) PTS. EtOH, 81%; q) NaOH, EtOH, 51%.

Fig. (23).



Conditions: a) methylacrylate, DABCO, 70%; b) OsO_4, NMO, acetone/H_2O; c) TBSCl, TEA, DMAP, 89% for two steps; d) MsCl, Py, CH_2Cl_2; e) K_2CO_3, MeOH, 91% for two steps; f) DIBALH, CH_2Cl_2, g) $NaBH_4$, MeOH, 90% for two steps; h) Cl_3CCN, DBU, CH_2Cl_2; i) Et_2AlCl, CH_2Cl_2, 97% for two steps; j) $CO(OCCl_3)_2$, Py, CH_2Cl_2; k) K_2CO_3, MeOH, 81% for two steps; l) MOMCl, DIPEA, CH_2Cl_2, 87%.

Fig. (24).

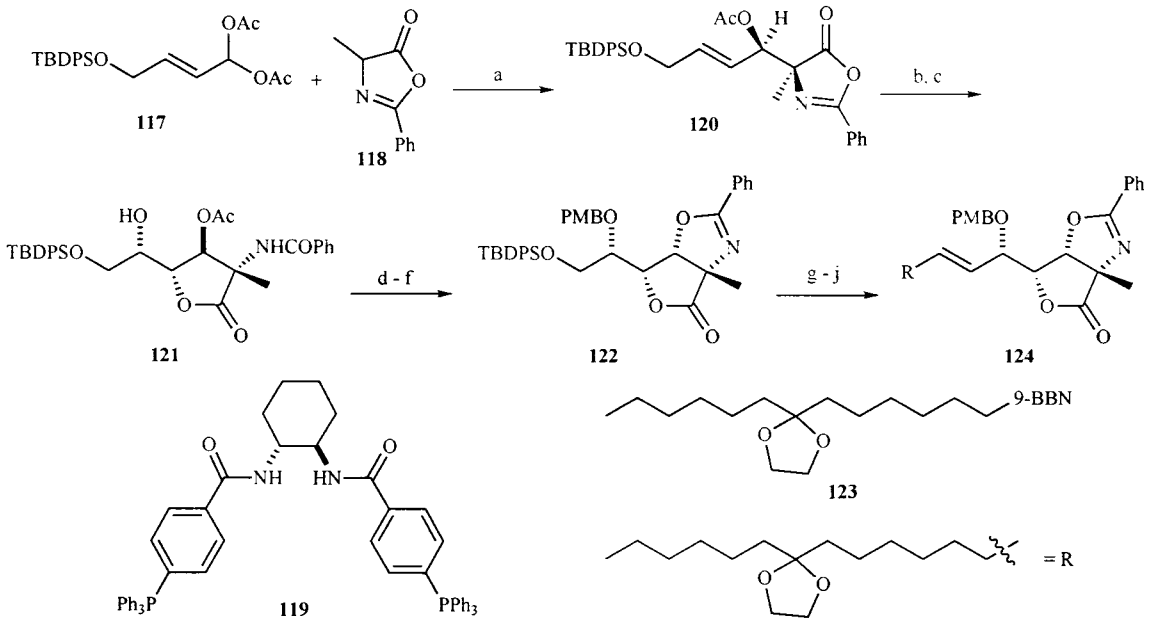
isolated yield. The acetyl protected hydroxyl group in **121** was transformed into a tosylate and an intramolecular nucleophilic substitution yielded oxazoline **122** with the desired absolute configuration of the final product. Removal of the silyl protecting group, oxidation of the primary alcohol and iodomethylenation afforded the coupling partner for the Suzuki cross coupling with borane **123** and the sphingofungin derivative **124** was obtained in very good yield. The natural product **18** was obtained after deprotection and hydrolysis in 15 steps and 17% overall yield. Analogously, the synthesis of **17** was accomplished in 17 steps and 5.1% overall yield (Fig. (**25**)) [52].

3.5.4. Shiozaki (Sphingofungin E 2001)

Shiozaki and co-workers used a combination of known methods for a long total synthesis of sphingofungin E (**17**). The known benzylidene derivative **125** was oxidised and chloromethylated to give alcohol **126**, which in turn was treated according to the method of Olesker [41] to afford azide **127**. The absolute configuration of the secondary hydroxyl group in **128** was inverted with a combination of oxidation and selective reduction in a ratio of >95:5 in favour of the desired azide **128**. The key intermediate **130**, obtained after hydrolysis and a series of deprotection steps, was transformed to the final product **17** in 29 steps and 1% overall yield with the above-mentioned procedure by Trost, consisting of iodoolefination and Suzuki-Miyaura cross-coupling [52] (Fig. (**26**)) [53].

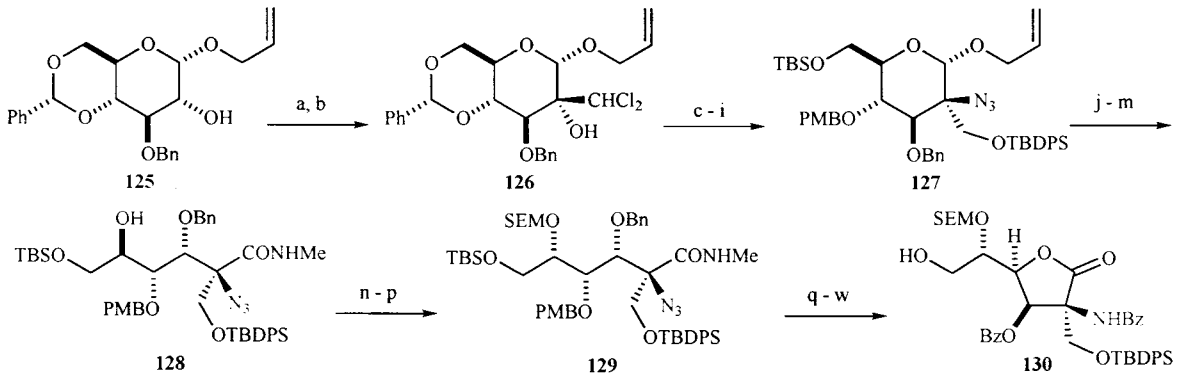
3.5.5. Chida (Sphingofungin E 2002) and Ham (Sphingofungin F 2002)

Sphingofungin E (**17**) and F (**18**) were synthesised with the same methodology as used for the total synthesis of **1**. Chida and co-workers synthesised **17** from D-glucose in 26



Conditions: a) **106**, CH_2Cl_2 (*E*/*Z* 17:1); b) DIBALH, CH_2Cl_2, 90%; c) TBHP, L-(+)-DIPT, Ti(O*i*-Pr)$_4$, 4 Å MS, CH_2Cl_2, 73%; d) TBSCl, TEA, DMAP, CH_2Cl_2, 80%; e) $Pd(OH)_2/C$, H_2, EtOAc/MeOH, 97%; f) $(COCl)_2$, DMSO, TEA, CH_2Cl_2, 87%; g) **49**, BuLi, THF, 85%; h) TBAF, THF, 95%; i) Cl_3CCN, DBU, CH_2Cl_2; j) BF_3Et_2O, CH_2Cl_2, 80% for two steps; k) $CO(OCCl_3)_2$, Py, CH_2Cl_2; l) K_2CO_3, MeOH, 82% for two steps; m) PDC, DMF; n) CH_2N_2, Et_2O, 78% for two steps; o) PhSSPh, hv, cy. 1,4-dioxane, 95%; p) PTS, EtOH, 81%; q) NaOH, EtOH, 51%.

Fig. (25).

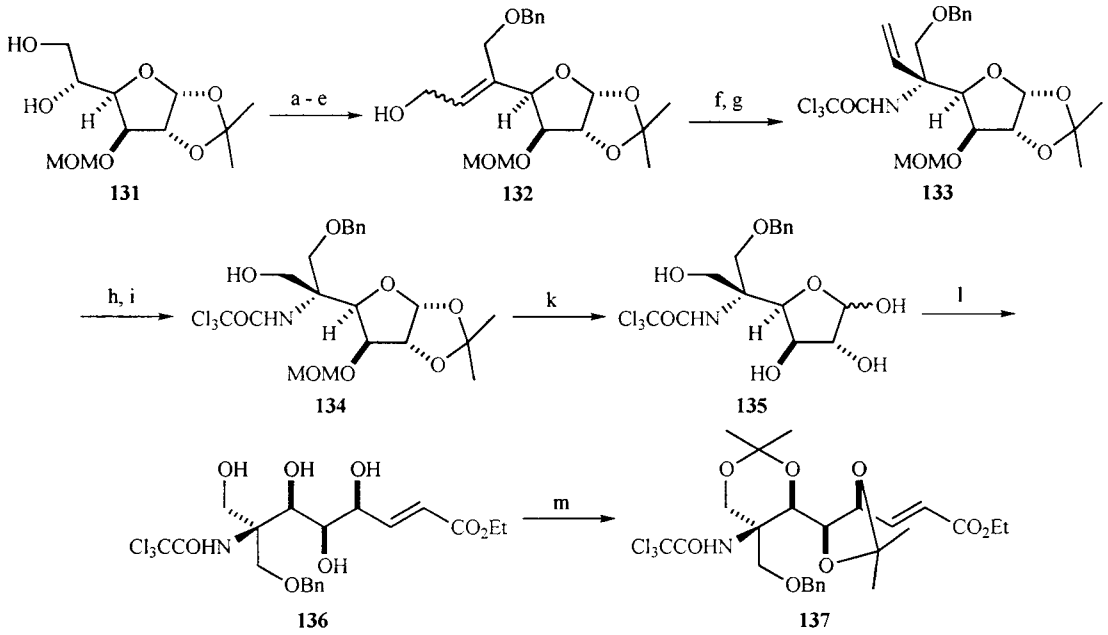


Conditions: a) $(COCl)_2$, DMSO, TEA, CH_2Cl_2, 76%; b) $LiCHCl_2$, THF, 70%; c) DBU, DMSO; d) NaN_3, 15-crown-5, HMPA; e) $NaBH_4$, MeOH, 84% for three steps; f) TBDPSCl, imid., DMF; g) CSA, MeOH, 86% for two steps; h) TBSCl, imid., DMF; i) PMBCl, NaH, DMF, 64% for two steps; j) $[Ir(COD)(PMePh)_2]PF_6$, THF; k) NBS, H_2O/THF, 82% for two steps; l) Dess-Martin periodinane, CH_2Cl_2; m) H_2NMe, MeOH, 94% for two steps; n) $(COCl)_2$, DMSO, TEA, CH_2Cl_2; o) L-Selectride, THF, 82% for two steps; p) SEMCl, DIPEA, DCE, 85%; q) MeI, NaH, DMF, 97%; r) DDQ, H_2O, CH_2Cl_2; s) PPTS, tol, 62% for two steps; t) $NaBrO_3$, $NaHSO_3$, H_2O/EtOAC, 69%; u) Pd, H_2, EtOAc; v) PhCOCl, TEA, CH_2Cl_2, 80% for two steps; w) aq. H_2SO_4, acetone, 87%.

Fig. (26).

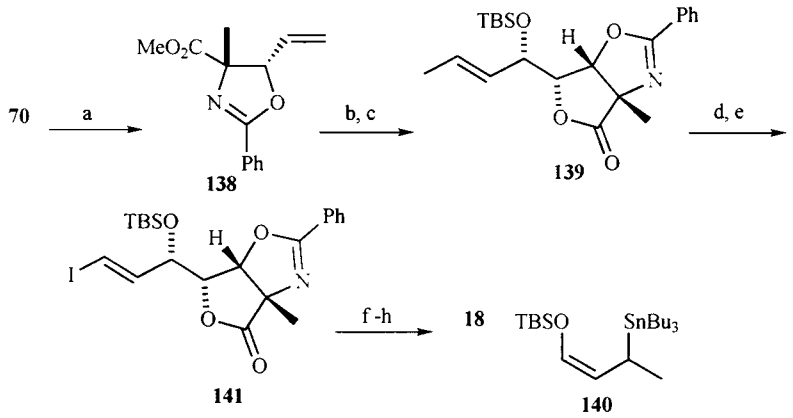
linear steps with the known strategy from myriocin (**1**) [38] with an Overman rearrangement of an allylic trichloroacetimidate as the key step. The primary hydroxyl group of known diol **131**, prepared from D-glucose in three steps, was protected and the secondary hydroxyl group oxidised to the corresponding ketone. Wittig reaction with the stabilised ylide (Ph_3P=$CHCO_2Et$) yielded, after reduction, a mixture of allyl alcohol **132** (*E*:*Z* 1:4). The stereoselective Overman rearrangement of the separated *Z*-isomer resulted in a mixture of isomers (*R*:*S* 64:14). The (*R*)-isomer **133** was transformed via reductive ozonolysis and subsequent reduction to the alcohol **134**. Acid hydrolysis removed the MOM and the acetonide protecting group to give furanose **135**, and after another Wittig reaction with the previously used stabilised ylide (Ph_3P=$CHCO_2Et$) the unsaturated ester **136** was formed exclusively. The hydroxyl groups of **136** were simultaneously protected and, then, the obtained diacetonide **137**, analogously to **1**, transformed to the final product **17**. (Fig. (**27**)) [54].

Ham and co-workers applied their method of Pd(0)-catalysed diastereoselective oxazoline formation used in the total synthesis of **139** to accomplish a total synthesis of **18**. The additional secondary hydroxyl group was introduced via $MgBr_2$-promoted γ-alkoxy allylic stannane addition of **140**



Conditions: a) *n*-Bu_2SnO, tol; b) CsF, BnBr, DMF, 88% for two steps; c) Swern oxidation; d) $Ph_3P{=}CHCO_2Et$, tol, 98% for 2 steps, *E*:*Z* = 1:4; e) DIBALH, tol, 73%; f) Cl_3CCN, DBU, CH_2Cl_2; g) xylene, 140 °C, sealed tube, 64% for two steps; h) O_3, CH_2Cl_2, then Me_2S; i) $Zn(BH_4)_2$, Et_2O, 91% for two steps; k) aq. HCl, THF; l) $Ph_3P{=}CHCO_2Et$, CH_2Cl_2; m) $Me_2C(OMe)_2$, CSA, 46% for three steps.

Fig. (27).



Conditions: a) MeI, KHMDS, HMPA, THF, 73%; b) O_3, MeOH, then Me_2S; c) **140**, $MgBr_2{\cdot}Et_2O$, CH_2Cl_2, 78% for two steps; d) O_3, MeOH, then Me_2S; e) $CrCl_2$, CHI_3, THF, 63% for two steps; f) **72**, *t*-BuLi, then $ZnCl_2$, $Pd(PPh_3)_4$, THF, 68%; g) HCl, THF, 80%; h) NaOH, 79%.

Fig. (28).

to the aldehyde obtained from ozonolysis of **139** (Fig. (**28**)) [55].

CONCLUSIONS

The antifungal and immunosuppressive metabolite myriocin (**1**) was simultaneously discovered in 1972 by two independent research groups. The first group isolated **1** from the culture broth of the thermophilic fungus *Myriococcum albomyces* and named it myriocin while the second group found the compound in *Mycelia sterilia* and named it thermozymocidin. More recently, **1** was also isolated from the culture mash of the non-thermophilic organism *Melanconis flavovirens* and, termed as ISP-1, from the thermophilic fungus *Isaria sinclairii*. Myriocin binding proteins were identified from mammalian homologues of two yeast proteins that have been genetically linked to sphingolipid biosynthesis. Myriocin (**1**) inhibits in CTLL-2-derived microsomes SPT, an enzyme which catalyses the first step of the sphingolipid biosynthesis. In horses the accumulation of free sphingoid bases (sphinganine) serum, urine, kidney, or liver caused by the mycotoxins fumonisins is suppressed by inhibition of ceramide synthase. The accumulation of sphingoid bases is correlated with outbreaks of mortal equine leukoencephalomalacia (ELEM) and other farm animal diseases.

Six total syntheses of naturally occurring (+)-myriocin (**1**) have been published since its discovery and characterisation. Despite the fact that **1** is a α-substituted

amino acid, most of the syntheses have used carbohydrate derivatives as chiral starting materials because of the predisposition of the carboxy and hydroxyl groups. The alternative choice of an amino acid proved to be versatile in several of syntheses, though, in these cases all stereocentres of the final product had to be established during synthesis. The two formal synthesis of **1** published so far started from D-glucose and followed similar synthetic strategies.

Mycestericin A to C (**2** – **4**), structurally and biologically closely related to **1**, have been of minor synthetic interest so far, and only a few total syntheses are published. Scientific focus was mainly on the two pairs of diastereomers, mycestericins D/F (**5**/**7**) and E/G (**6**/**8**).

The very potent immunosuppressive activity of **1** - **8** *in vitro* and *in vivo* with a completely different mechanism than the commercial immunosuppressive tacrolimus (FK506) CsA led to the development of symmetrical 2-amino-1,3-propanediols and, hence, the very effective immunosuppressant FTY720 (**11**). Aminoalcohol **11** is currently in phase III clinical trials and is expected to become in combination with the known calcineurin-inhibitors CsA or FK506 a novel potent drug for the therapeutic areas of transplantation and autoimmunity.

From the sphingofungin family only metabolites E (**17**) and F (**18**) have the characteristic quaternary carbon present in **1** - **8**. Therefore, **17** and **18** caught the main synthetic interest and a series of total synthesis with completely different methodologies have been published. In a few cases just a modified synthesis of **1** was applied to the sphingofungins.

Although much effort in the synthesis of sphingosin-related metabolites has been done, many challenges in research still remain. The interest of the pharmaceutical industry in derivatives of theses immunosuppressive metabolites and their need for optical active compounds might lead to the development of short and highly stereoselective methods and syntheses to obtain the natural compounds and their chiral derivatives.

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