Fumagillin and Structurally Related Molecules as Source of New Drugs

Diego Gamba-Sánchez*

Lehrstuhl für Organische Chemie I, Department of Chemistry, Technische Universität München, Lichtenbergstr. 4, 85747 Garching, Germany

Abstract: Cancer is one of the most prolific causes of death all around the world. Currently there is a huge focus around the globe on the identification of effective drugs against this disease. One important target is the development of new anti-angiogenic compounds. Fumagillin and other structurally related molecules are some of the most potent anti-angiogenic compounds reported to date with several currently being investigated in clinical studies.

Fumagillin has been used for more than 50 years against microsporidial parasites in animals, however, angiogenesis inhibition constitutes a new activity and an important research topic, this is the reason why nowadays some teams are working to achieve an effective way to obtain fumagillin analogues with more potent anti-angiogenic activity.

Here we present an overview on fumagillin and fumagillin analogues as leads products; we summarize the key structural features for anti-angiogenic activity and discuss the total synthesis and synthesis of analogues and their biological activity.

Keywords: Angiogenesis, anti-angiogenic therapy, fumagillin, ovalicin, structure-activity relationship, TNP470.

INTRODUCTION

Fumagillin (**1,** Fig. **1**) was isolated for the first time in 1951 by Elbe and Hanson from the microbial organism *Aspergillus fumigatus* [1], and its structure was first hypothesized in 1961 [2] by chemical degradation and then confirmed in 1961 by X-ray crystallographic analysis [3], these studies also confirmed the structure of fumagillol (**6**, Fig. **1**) the saponification product of **1**.

Fumagillin is essentially an antimicrobial agent and has been used against microsporidial parasites *Nosema apis* and *Nosema ceranae* in honey bees since 1957 (Flumidil B®). Several studies have been carried out on Fumagillin itself as well as other structurally related molecules (SRM) like TNP-470 (**2**, Fig. **1**) also called AGM-1470 (a semi-synthetic analogue), Ovalicin (**3**, Fig. **1**) a sesquiterpene isolated from cultures of the fungus *Pseudorotium ovalis Stolk* [4, 5], PPI-2458 (**4**, Fig. **1**) (synthetic analogue) [6, 7] and fumarranol (**5**, Fig. **1**) among others. These studies have shown these molecules are potent inhibitors of angiogenesis (formation of new blood vessels from pre-existing vessels), however some of them have sever side-effects, thus synthesis of more active and less toxic analogues has received a lot of attention since 1990 [8].

According to the World Health Organization (WHO) report [9] cancer is one of the largest causes of death all around the world. Anti-angiogenic therapy has shown to be a powerful tool to treat this disease; this is the reason why today many efforts are being made to find more effective anti-angiogenic agents. Fumagillin and other structurally related compounds are some of the most potent anti-angiogenic agents reported to date.

Biological activities of these molecules as well as the structure-activity relationship have been extensively studied and they have shown biological properties other than angiogenesis inhibition.

BIOLOGICAL ACTIVITY

Since its isolation **1** has been known as an antibiotic. Antibacterial and antifungal activity was demonstrated but no antiviral activity was shown *in vivo*. One year later the use of **1** to control Nosema disease of honey bees was described [10] and some studies [11, 12] have also shown it is effective against some myxozoan parasites in fishes.

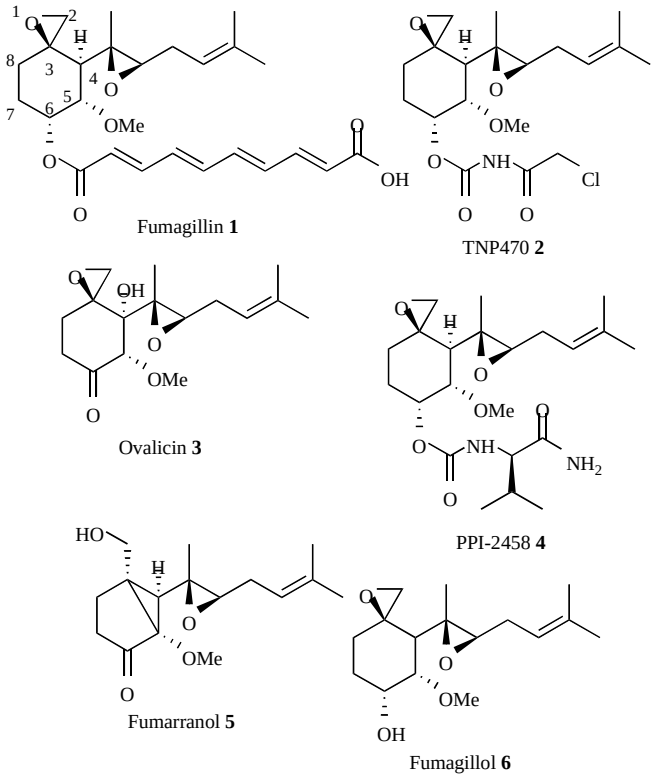


Fig. (1).

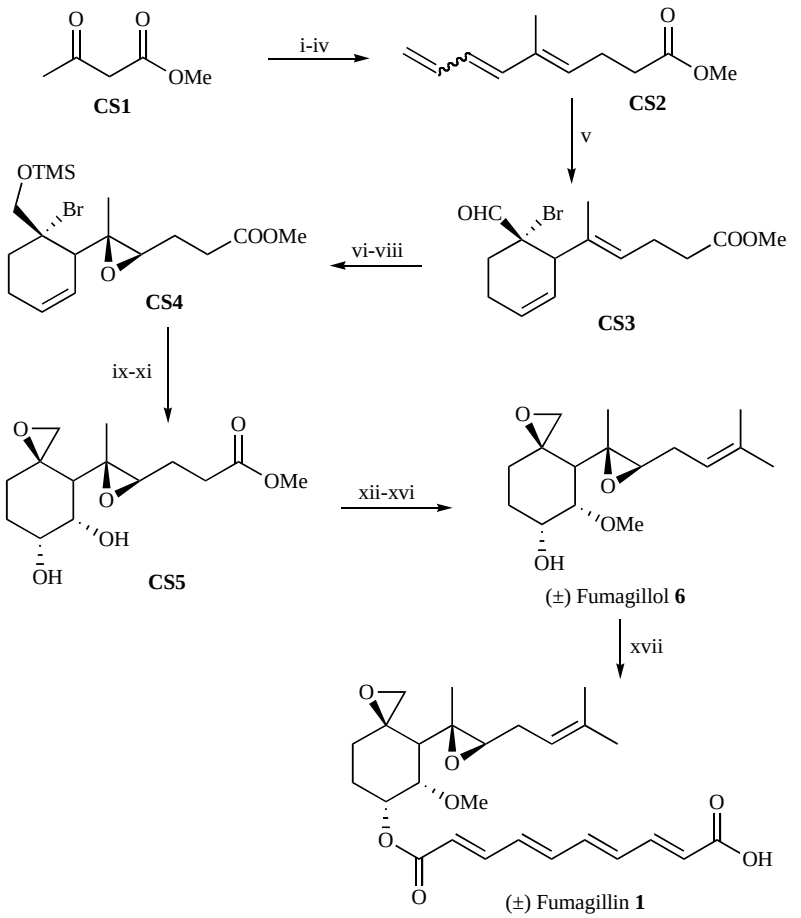
Recently the team of Dr. Molina [13] described the use of **1** for the treatment of intestinal microsporidiosis (or microsporidiasis) in AIDS patients. Microsporidiosis due to *Enterocytozoon bieneusi* is a cause of persistent diarrhea, malabsorption and wasting in immuno-compromised patients, for which there is still no effective treatment.

Amebicidal properties were also described [14] and **1** has been used for treatment of amebiasis in humans [15]. It is necessary to emphasize the fact that at that time no side effects or toxicity for **1** were described.

Angiogenesis is the generation of new blood vessels from pre-existing vessels; this is an essential process in growth, development and wound repair, but it also plays a fundamental role in tumor growth and metastasis as well as in other angiogenic diseases such

*Address correspondence to this author at the Laboratorio de Síntesis Orgánica Bio- y Organocatálisis, Department of Chemistry, Universidad de los Andes, Cra 1 No. 18A-12 Q305, Bogotá-Colombia; Tel: +5713394949; Fax: +5713324366; E-mail: da.gamba1361@uniandes.edu.co

1875-6298/12 $58.00+.00



Conditions: (i) $(CH_3)_2CCHCH_2Br$; (ii) Na/EtOH, 57% (over 2 steps); (iii) SeO_2, $MeO(CH_2)_2OMe$, 41%; (iv) $XPh_3P{=}CHCH{=}CH_2$, 84%; (v) α-bromoacrolein, C_6H_6, K_2CO_3, hydroquinone, 80%; (vi) $NaBH_4$, THF/H_2O, >98%; (vii) TMSCl, Et_3N, THF, 90%; (viii) mCPBA, CH_2Cl_2, $NaHCO_3$, 80%; (ix) TBAF,THF; (x) MeONa; (xi) OsO_4, pyr, 81% (3 steps); (xii) sodium *t*-amylate, THF, MeI, 65%; (xiii) MeLi, THF, 75%; (xiv) Ac_2O, pyr, 95%; (xv) MsCl, Et_3N, THF then TBABr, THF; (xvi) K_2CO_3, MeOH; (xvii) MeLi, decatetraenedioyl chloride, THF.

Scheme 1. Corey's racemic synthesis of fumagillin.

as: diabetic retinopathy, haemangiomas and arthritis among others. Endothelial cells are fundamental in this procedure, and it has been found that type 2 of methionine aminopeptidase (MetAP-2) protein plays a key role in endothelial cells proliferation [16]. The common target for angiogenic inhibitors such as fumagillin, TNP470 or ovalicin (**1**, **2** and **3**, Fig. **1**) is this MetAP-2 [17]. In fact **1** binds covalently to the proteins and inhibits its function. There are two types of methionine aminopeptidase in eukaryotes, but Fig. (**1**) compounds inhibit selectively MetAP-2. This protein is an important target in the development of new anti-angiogenic compounds which can be used in the treatment of different types of cancer but also to control other angiogenic diseases.

More recent studies have demonstrated **1** can be a lead compound for the improvement of novel type of AIDS therapeutic agents that targets Vpr activity [18]. HIV-1 viral protein R (Vpr) is one of the human immunodeficiency virus type 1 proteins that have important roles in viral pathogenesis, nevertheless no effective drugs that target this Vpr has been developed for AIDS therapy.

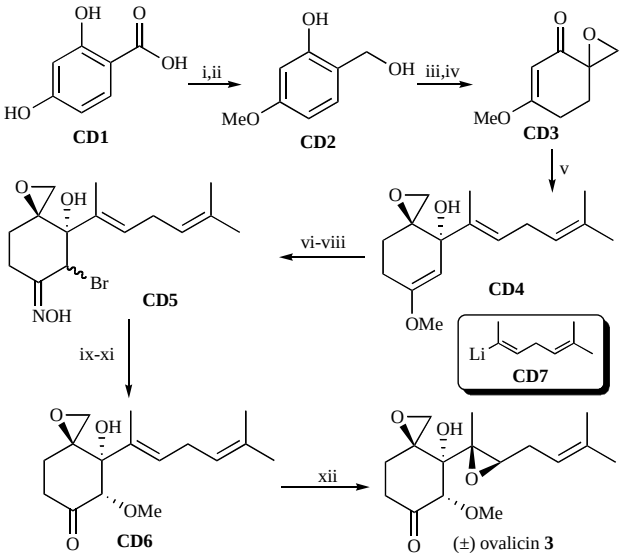
The interaction between Fig. (**1**) compounds and MetAP-2 has another important implication. It was recently demonstrated [19] that **1**, **2** and **5** bound with *Plasmodium falciparum* MetAP-2 (PfMetAP-2) but in a reversible manner emphasizing the difference between the active sites of humans HsMetAP-2 and PfMetAP-2. **1**, **2** and **5** have demonstrated to block malaria growth *in vitro* and *in vivo*; these results suggest that PfMetAP-2 can be a target and **5** an important lead for the discovery of new antimalarial drugs.

FUMAGILLIN AND OVALICIN SYNTHESIS

In this section we will focus on total synthesis or formal synthesis of both fumagillin and ovalicin (**1** and **3** in Fig. **1**) and they will be presented in chronological order; synthesis of analogues will be discussed in a posterior section of this paper.

The first total synthesis of racemic **1** was described in 1972 by Corey and Snider [20]. Actually the goal of that synthesis was fumagillol (**6**, Fig. **1**) (the saponification product of **1**); it was demonstrated that treatment of **6** with MeLi and an excess of decatetraenedioyl chloride in THF gave **1** identical in all aspects with natural samples in good yield.

Their synthetic strategy was based on a Diels-Alder reaction of the intermediate **CS2** (obtained from methyl acetoacetate **CS1**) with α-bromoacrolein (Scheme **1**). Compound **CS3** was reduced, and then protection of the obtained primary alcohol followed by epoxidation gave the intermediate **CS4** which could be transformed in **CS5** by intramolecular Williamson reaction, followed by dihydoxy-



Conditions: (i) MeI, K_2CO_3, acetone, 83%; (ii) bis(methoxyethoxy)aluminiumhydride, Et_2O, 97%; (iii) $NaIO_4$ aq., THF, 61%; (iv) NHNH, DME, 77%; (v) $(CH_3)_2C{=}CHCH_2CH{=}CCH_3I$, *t*BuLi, 83%; (vi) NBS, MeOH; (vii) PTSA cat., 55% (2 steps); (viii) $NH_2OH{\cdot}HCl$, AcOH, pH 6 buffer, quant.; (ix) Et_3N, MeOH, >90%; (x) $TiCl_3$ aq., MeOH, pH 6 buffer; (xi) K_2CO_3, MeOH, 63% (2 steps); (xii) $[VO(acac)_2]$ cat. then *t*BuOOH, toluene, 89%.

Scheme 2. Corey's racemic synthesis of ovalicin.

lation using OsO_4. Selective methylation of equatorial 5-OH group with sodium *t*-amylate and MeI, followed by addition of MeLi to the ester moiety provided a secondary and tertiary diol which was acetylated selectively, and then formation of the trisubstituted double bond was accomplished by mesylation and elimination promotd by TBAB. After that, saponification of acetate provided fumagillol (**6**, Fig. **1**) which could be transformed to **1** under the conditions described above.

More than 10 years later Corey and Dittami published the first total synthesis of ovalicin (**3**, Fig. **1**), using a completely different strategy [21] (Scheme **2**).

2,4-dihydroxybenzoic acid **CD1** was methylated selectively and reduced to furnish primary alcohol **CD2**. Treatment of **CD2** with aqueous $NaIO_4$, followed by reduction with diimide provide the epoxide enol ether **CD3**. Addition of vinyllithiated compound **CD7** to the ketone produced ovalicin precursor **CD4**, bromination of **CD4** gave a mixture of hydrolyzed product and brominated enol ether and this mixture was hydrolyzed with *p*-toluenesulfonic acid PTSA, then oxime generation with hydroxylamine gave **CD5**. This was treated with methanol and triethylamine to produce the α methoxy oxime, and hydrolysis of this oxime was accomplished with aqueous $TiCl_3$ in methanol giving compound **CD6** as a 1:1 mixture of two diastereoisomers. The mixture was treated with K_2CO_3 in methanol to produce the more stable isomer (OCH_3 equatorial) **CD6** as a single diastereomer, which was finally transformed into **3** by epoxidation using a catalytic amount of vanadyl acetylacetonate $[VO(acac)_2]$ and *tert*-butyl hydroperoxide (*t*BuOOH).

In 1994 two enantioselective total synthesis of **3** were published; one of them [22] used commercially available L-Quebrachitol (Fig. **2**) as a chiral template.

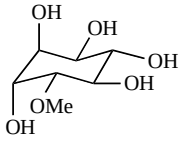
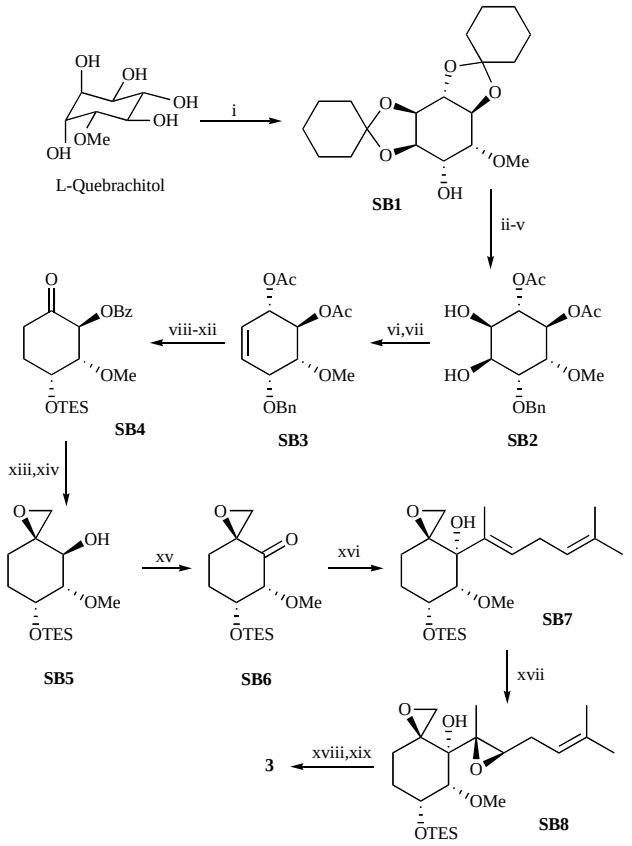


Fig. (2). Selective protection and deprotection sequences were applied (see Scheme **3**) to obtain key intermediate **SB6**.



Conditions: (i) cyclohexanone, PTSA, benzene, 72%; (ii) BnBr, DMF, 90%; (iii) $OH(CH_2)_2OH$, PTSA, CH_2Cl_2, 70%; (iv) Ac_2O, pyr., 98%; (v) TFA, THF/H_2O, 77%; (vi) $CSCl_2$, DMAP, CH_2Cl_2; (vii) $(MeO)_3P$; (viii) NH_3, MeOH, 82% (3 steps); (ix) MnO_2, CH_2Cl_2, 50%; (x) H_2, Pd/C, EtOH, 94%; (xi) BzCl, pyr., 83%; (xii) TESCl, imidazole, DMF, 97%; (xiii) $Ph_3P{=}CH_2$, THF, 70%; (xiv) mCPBA, CH_2Cl_2, 84%; (xv) DMSO, TFAA, CH_2Cl_2, 88%; (xvi) **CD7**, THF/toluene, 72%; (xvii) $[VO(acac)_2]$, *t*BuOOH, benzene, 72%; (xviii) TBAF, THF; (xix) PDC, CH_2Cl_2, 78% (2 steps).

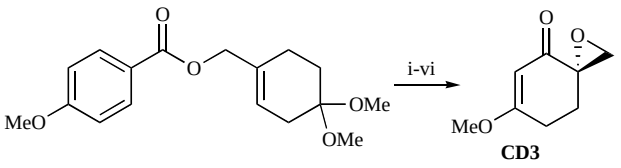
Scheme 3. Samadi's enantioselective synthesis of 3.

Insertion of the side chain using the same vinyllithiated (**CD7**), yielded **SB7** which was then epoxidized to produce **SB8**. TES ether cleavage followed by oxidation provided **3** in 19 steps and 8.5% overall yield.

The other synthesis was described by Corey [23] and used as the key step a Sharpless dihydroxylation reaction to obtain an enantiomerically enriched **CD3** intermediate (Scheme **4**); synthesis of **3** was then achieved using the same reaction sequence described in Scheme **2**.

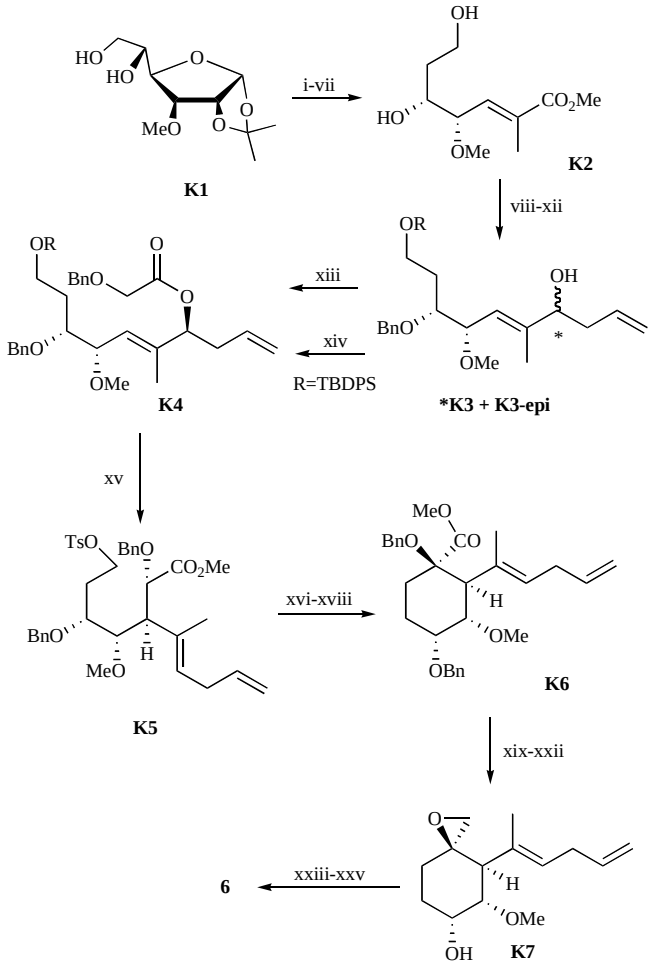
The first asymmetric synthesis of fumagillol (**6**, Fig. **1**) was described by Kim *et al.* [24] using a glycolate Claisen rearrangement and an intramolecular ester enolate alkylation strategy (Scheme **5**).

K1 is obtained from commercially available 1,2:5,6-di-*O*-isopropylidene-α-D-allofuranose, and then easily transformed in 7 steps in **K2**. This methyl ester was reduced with DIBALH then oxidized to produce the corresponding aldehyde, which was then alkylated with allyl magnesium bromide, giving a separable mixture of two epimers **K3** and **K3-epi**. **K3** was esterified using DCC under Steglich conditions and **K3-epi** by Mitsunobu procedure to afford **K4** ester. Claisen-Ireland rearrangement of **K4** under modified condition described by Burke-Fujisawa-Kallmerten [25-27] afforded **K5** as a single diastereomer after silyl ether cleavage and tosylation. The intramolecular ester enolate alkylation strategy was applied to obtain **K6**. This was then reduced and selectively tosylated to afford the precursor of intramolecular Williamson ether synthesis, which proceeded smoothly yielding **K7**. This was then



Conditions: (i) K_2OsO_4, 1 mol% $(DHQ)_2PHAL$, $K_3Fe(CN)_6$, K_2CO_3, $CH_3SO_2NH_2$, *t*BuOH/H_2O, 93%, 99% ee; (ii) Swern oxidation; (iii) PTSA cat. CH_2Cl_2, 87%; (iv) K_2CO_3, MeOH, 93%; (v) MsCl, Et_3N, CH_2Cl_2; (vi) NaOH aq., 82% (2 steps).

Scheme 4. Corey's enantioselective synthesis of CD3.



Conditions: (i) MsCl, TEA, CH_2Cl_2; (ii) NaI, MEK, 77% (2 steps); (iii) 9-BBN, THF then 30% H_2O_2, 3N NaOH, 93%; (iv) Dowex $50x_2$-200 resin, H_2O; (v) $NaIO_4$, acetone/H_2O; (vi) Ph_3P=C $(CH_3)CO_2CH_3$, CH_3CN; (vii) K_2CO_3, MeOH, 63% (4 steps); (viii) TBDPSCl, DMAP, TEA, CH_2Cl_2, 93%; (ix) $CCl_3(C{=}NH)OBn$, CF_3SO_3H cat., cyclohexane/CH_2Cl_2; (x) DIBALH, toluene, 89% (2 steps); (xi) MnO_2, CCl_4, 90%; (xii) CH_2=$CHCH_2MgBr$, THF, 48%; (xiii) $BnOCH_2CO_2H$, DMAP, DCC, CH_2Cl_2, 100%; (xiv) DIAD, $BnOCH_2CO_2H$, PPh_3, THF, 83%; (xv) LHMDS, TMSCl/TEA, THF then Triton B, MeI, THF, 89%; (xvi) TBAF, THF, 95%; (xvii) TsCl, pyr, CH_3Cl; (xviii) KHMDS, THF, 64% (2 steps); (xix) DIBALH, toluene, 87%; (xx) Li/NH_3, 90%; (xxi) TsCl, TEA, DMAP, CH_2Cl_2; (xxii) K_2CO_3, MeOH, 88% (2 steps); (xxiii) mCPBA, $NaHCO_3$, CH_2Cl_2, 92%; (xxiv) O_3, AcOEt; (xxv) Ph_3P=$C(CH_3)_2$, THF, 45% (2 steps).

Scheme 5. Kim's enantioselective synthesis of 6.

epoxidized to install the oxirane ring in the side chain, and ozonolysis followed by Wittig reaction gave finally **6**.

A formal asymmetric synthesis of ovalicin (**3**, Fig. **1**) was described by Pollini and coworkers [28] using (-)-quinic acid (Fig. **3**) as chiral template. They obtained the same key intermediate **SB5** as Samadi described above.

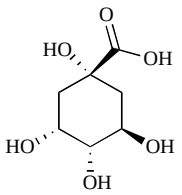
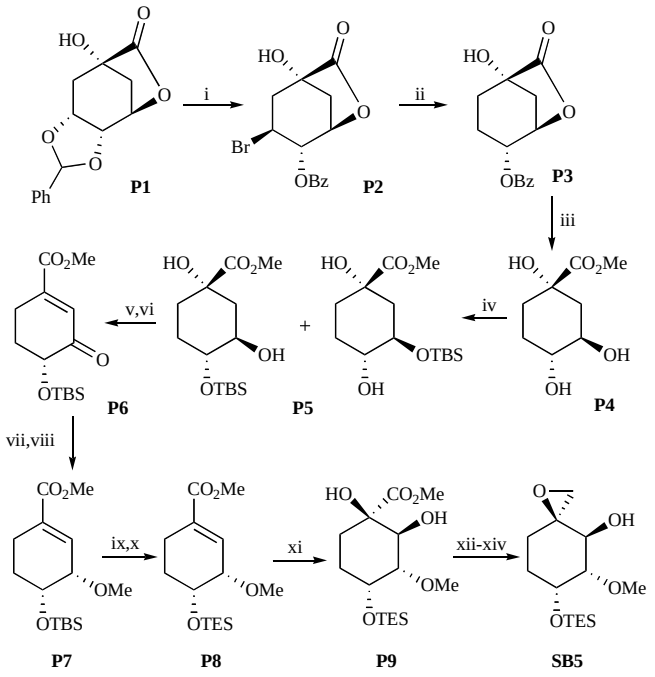

(-)-quinic acid

Fig. (3).

The synthesis started with the preparation of **P1** by a known procedure from (-)-quinic acid [29], then bromobenzoate **P2** was easily obtained in good yield using NBS to promote the regiospecific opening of benzilidenacetal ring. The bromine atom was subsequently removed by a free radical hydride reduction affording **P3**. Saponification gave the triol **P4** which was treated with TBSCl and imidazole to produce an inseparable mixture of regioisomers **P5**.



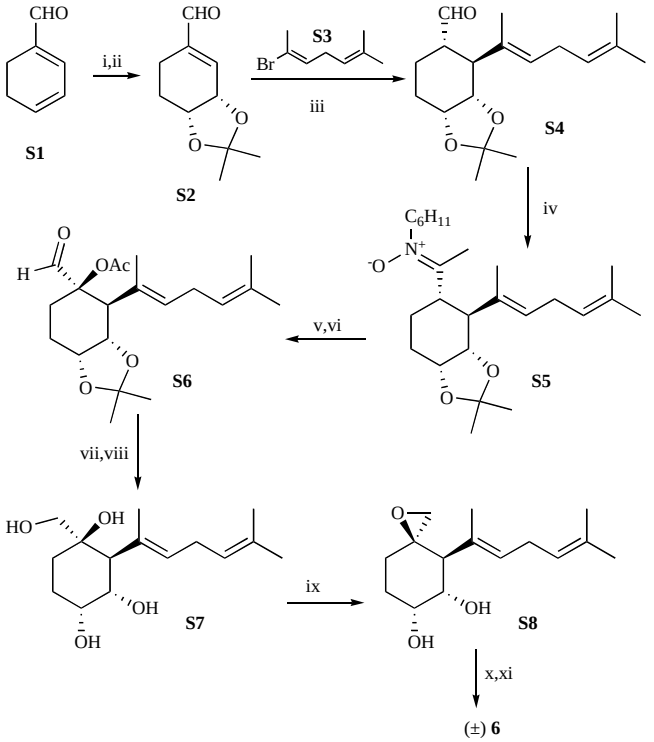
Conditions: (i) NBS, AIBN, CCl_4; (ii) nBu_3SnH, AIBN, C_6H_6, 97%; (iii) K_2CO_3, MeOH, 89%; (iv) TBSCl, imidazole, DMF, 72%; (v) PCC, 3Å molecular sieves, C_5H_5N, CH_2Cl_2; (vi) $POCl_3$, C_5H_5N, 60% (2 steps); (vii) $NaBH_4$, MeOH, 86%; (viii) MeI, Ag_2O, CH_2Cl_2, 88%; (ix) TBAF, THF, 94%; (x) TESCl, C_5H_5N, 83%; (xi) OsO_4, NMO, C_5H_5N, H_2O, *t*BuOH, 91%; (xii) $LiAlH_4$, THF, 60%; (xiii) TsCl, Et_3N, DMAP, CH_2Cl_2; (xiv) K_2CO_3, MeOH, 80% (2 steps).

Scheme 6. Pollini's synthesis of 3.

Fortunately when this mixture was oxidized with pyridinium chromate in the presence of molecular sieves, α,β-unsaturated ketone **P6** was obtained as a single isomer and in 60% overall yield. Reduction of the carbonyl group with sodium borohydride followed by methylation gave compound **P7**. This was then transformed into **P8** by a deprotection-protection sequence; the dihydroxylation gave compound **P9** in 91 % yield.

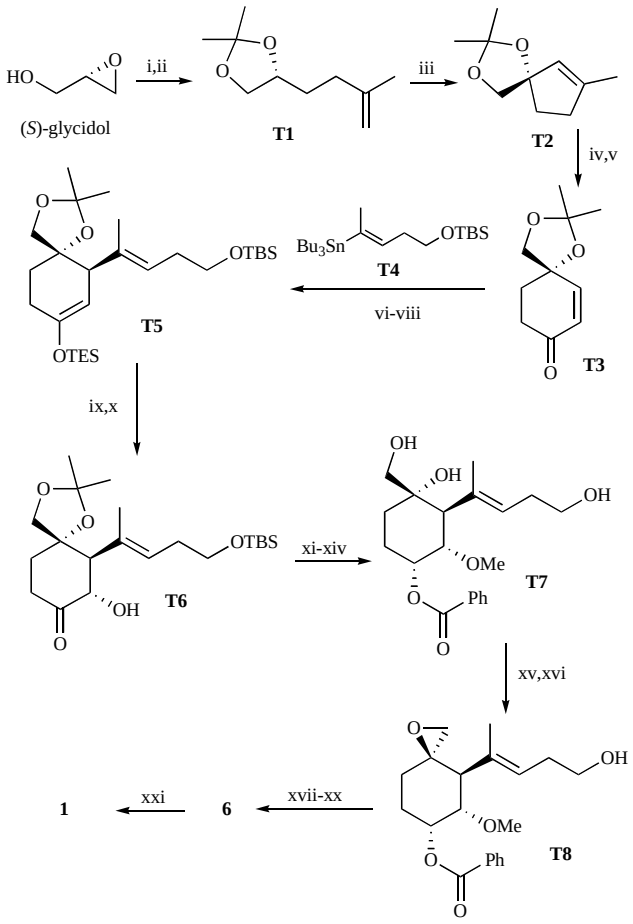
Finally a reduction with $LiAlH_4$, tosylation and intramolecular cyclization yielded **SB5** in 48% yield over three steps.

An elegant [3,3] sigmatropic rearrangement strategy developed by Cummins and Coates [30, 31] for creating α-acetoxycarbonyl compounds was applied to the synthesis of fumagillol (**6**, Fig. **1**) described by Sorensen *et al*. [32] (Scheme **7**).



Conditions: (i) OsO_4, NMO, H_2O, 2-propanol, 73%; (ii) acetone, PTSA, 87%; (iii) *t*BuLi, Et_2O, then Li(2-thienyl)CuCN, then BF_3-Et_2O, Et_2O, 46%; (iv) $C_6H_{11}NHOH$, EtOH, $NaHCO_3$; (v) AcCl, Et_3N, Et_2O; (vi) AcOH, NaOAc, 51% (3 steps); (vii) $LiAlH_4$, Et_2O, 71%; (viii) 2N HCl/THF, 81%; (ix) MsCl, DMAP, Et_3N, CH_2Cl_2 then 6N NaOH, MeOH, 97% (x) [$VO(acac)_2$], *t*BuOOH, benzene, 75%; (xi) *t*BuONa, MeI, THF, [15]crown-5, 40%.

Scheme 7. Sorensen's synthesis of (±)-6.



Conditions: (i) methallylmagnesiumchloride, Et_2O; (ii) 2,2-dimethoxypropane, PTSA, 90% (2 steps); (iii) Br_2, KHMDS, Et_2O, 72%; (iv) O_3, CH_2Cl_2 then PPh_3; (v) KOH aq, CH_2Cl_2, 85%; (vi) **T4**, nBuLi, CuCN, THF; (vii) **T3**, THF; (viii) TESCl, Et_3N, THF, 74% (3 steps); (ix) mCPBA, hexane; (x) TBAF, THF, 55% (2steps); (xi) MeI/Ag_2O, CH_3CN; (xii) L-selectride, THF then NaOH, 30% H_2O_2, 66% (2 steps); (xiii) PhCOCl, CH_2Cl_2/pyr; (xiv) Dowex 50 resin, MeOH, 52% (2 steps); (xv) $NaIO_4$, CH_2Cl_2/H_2O then $NaHCO_3$; (xvi) $Me_3SO^+I^-$, NaH, THF/DMSO, 78% (2 steps); (xvii) mCPBA, CH_2Cl_2, $NaHCO_3$; (xviii) DMP; (xix) $Ph_3P{=}C(CH_3)_2$, THF, nBuLi, 86% (3 steps); (xx) K_2CO_3, MeOH, 89%; (xxi) MeLi, decatetraenedioyl chloride, THF, 75%.

Scheme 8. Taber's synthesis of 1.

The synthesis started with known **S1** which can be easily transformed in **S2** by selective dihydoxylation followed by formation of the acetonide. Conjugate addition of the organocoprate generated *in situ* from **S3** produced **S4** in moderate yield. This was transformed in *N*-cyclohexylnitrone **S5** with *N*-cyclohexyl hydroxylamine. Then the key [3,3] sigmatropic rearrangement took place using acetyl chloride and triethylamine, the resultant crude imine was then hydrolyzed to give aldehyde **S6**. This aldehyde was reduced and the acetonide moiety was hydrolyzed to afford polyol **S7**, which was transformed into the spiro oxirane **S8** *via* intramolecular nucleophilic substitution of mesylated intermediate. This epoxide was transformed into **6** by epoxidation and selective methylation.

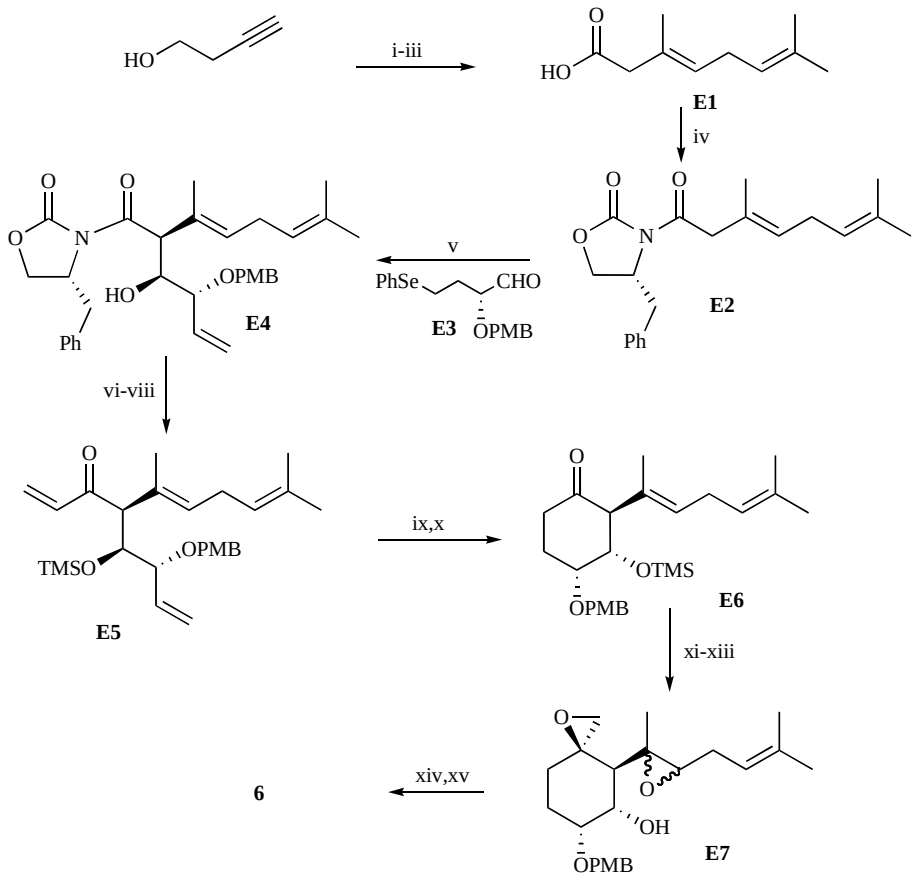
Also in 1999 another asymmetric synthesis of **1** was published [33], the synthetic strategy was based on a carbene mediated cyclization followed by an intramolecular aldol condensation (Scheme **8**).

Addition of methallyl magnesium bromide to (*S*)-glycidol followed by acetonide formation with 2,2-dimethoxypropane produced **T1** which was then treated with bromine and a strong base to yield spiro cyclopentene **T2**. These authors have described this reaction before [34] and they suggest the formation of the corresponding terminal bromo alkene which under basic conditions generates a carbene intermediate; this can insert the C-H bond producing the spiro compound.

Ozonolysis followed by intramolecular aldol condensation yielded **T3**. **T4** (obtained in three steps starting from dihydrofuran) was treated with nBuLi then with CuCN, this mixture was added to the ketone **T3** generating the addition product which was quenched with TESCl to afford silyl enol ether **T5** as a single diastereomer. Rubottom oxidation [35-37] of enol ether **T5** followed by cleavage of the obtained silyl ether provided **T6**, methylation followed by reduction with L-selectride and esterification of the secondary alcohol provided the intermediate acetonide which was hydrolyzed to yield diol **T7**. After oxidative cleavage of this diol, the spiro oxirane ring was introduced by a Corey-Chaykovsky epoxidation. **T8** was transformed in **6** as follow: epoxidation with mCPBA, oxidation of the primary alcohol, Wittig olefination and saponification of benzylic ester. The authors also described the reaction between **6** and decatetraenedioyl chloride to obtain fumagillin (**1**, Fig. **1**).

More recently Eustache *et al*. [38] published an elegant enantioselective synthesis of **6**; their strategy is based on an Evans aldol reaction and ring closing metathesis (RCM) to obtain the unfunctionalized skeleton. This synthesis is presented in the Scheme **9**.

Treatment of homopropargylic alcohol with $CpZrCl_2$ and $AlMe_3$ under Negishi's conditions followed by palladium catalyzed cross-coupling of the resulting allane with 1-bromo-3-methyl-but-2-ene produced a 9:1 mixture of regioisomers; this mixture was oxidized twice, first to yield the double bond oxidation product of the minor isomer, and the resulting mixture was easily separated by flash chromatography and the major alcohol isomer was oxidized with CrO_3, yielding the desired *E*-isogeranic acid **E1**. After forma-



Conditions: (i) Cp_2ZrCl, $AlMe_3$, DCE then 1-bromo-3-methyl-but-2-ene, $Pd(PPh_3)_4$; (ii) CuCl, O_2, $PdCl_2(CH_3CN)_2$ cat. DMF/H_2O, 65% (2 steps); (iii) CrO_3, H_2SO_4, H_2O/acetone, 35%; (iv) *t*BuCOCl, Et_3N then (*R*)-4-benzyl-2-oxazolidinone lithium salt, 70%; (v) LDA, THF, **E3**, then NBu_4IO_4, $CHCl_3$, reflux, 55%; (vi) *N,O*-dimethylhydroxylamine, $AlMe_3$, THF, 71%; (vii) TMSCl, Et_3N, DMAP, THF, 100%; (viii) vinylmagnesium bromide, THF, 87%; (ix) Ti(O*i*Pr)$_4$, Grubbs I cat., CH_2Cl_2, 53%; (x) Raney Ni, THF, 83%; (xi) $Me_3S^+OI^-$, NaH, LiI, DMSO/THF, 53%; (xii) PTSA, H_2O/THF, 75%; (xiii) Ti(O*i*Pr)$_4$, *t*BuOOH, CH_2Cl_2, 65%; (xiv) MeI, NaH, THF/DMF, 97%; (xv) DDQ, CH_2Cl_2/H_2O, 83%.

Scheme 9. Eustache's asymmetric synthesis of **6**.

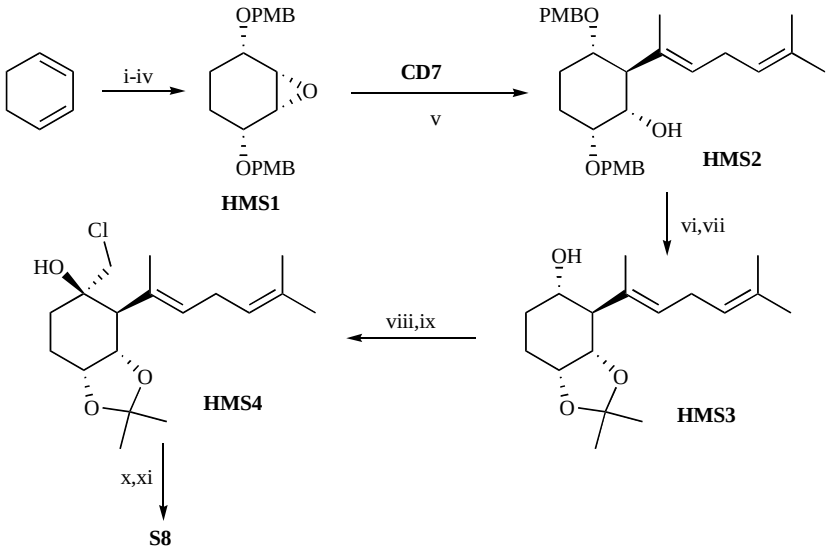
tion of *N*-acyl oxazolidinone **E2**, it was deprotonated and treated with aldehyde **E3**. The aldol product obtained was subjected to periodate oxidation to obtain expected product **E4**. The terminal double bond in **E5** was generated by addition of vinylmagnesium bromide to the Weinreb amide formed after oxazolidinone cleavage. **E5** was the then treated with Grubbs I catalyst in presence of Ti(O*i*Pr)$_4$ to generate the unsaturated six-member ring in 53% yield; this α,β unsaturated ketone was reduced with Raney nickel to provide compound **E6**. Corey-Chaykovsky epoxidation as described above followed by an epoxidation under Sharpless conditions Ti(O*i*Pr)$_4$/*t*BuOOH produced a 1:1 mixture of two epoxide isomers **E7**. Methylation of this mixture worked extremely well and yielded separable methylated isomers; after separation and cleavage of the PMB ether (carried out independently in both isomers) **6** and its epimer were obtained in good yields.

A racemic formal synthesis of **6** (Scheme **10**) was published by Simpkins in 2001 [39]. This synthesis began with a palladium catalyzed *bis*-acetoxylation of 1,3-cyclohexadiene; the obtained acetylated compound was saponified, epoxidized and then protected with a PMB group to afford **HMS1**.

The epoxide ring was opened with the appropriate vinyllithiated reagent **CD7** yielding **HMS2**. Deprotection of the alcohol was accomplished using Birch conditions; the obtained triol was protected selectively with dimethoxypropane giving the expected acetonide **HMS3**.

HMS3 was then oxidized under Swern oxidation conditions or using Dess-Martin periodinane (DMP) and the resulting ketone was treated with an excess of chloromethyllithium reagent, which was prepared as described by Sadhu and Matteson [40], yielding chlorohydrin **HMS4** with excellent diastereoselectivity. Acid treatment to remove the acetonide moiety followed by intramolecular cyclization produced intermediate **S8** which can be transformed in **6** or **1** using the reaction sequence described by Sorensen [32].

Langlois and Haudrechy [41] described in 2004 a stereoselective synthesis of fumagillol (**6**, Fig. **1**) (Scheme **11**). They used diisopropylidenmannitol **L1** as starting material; this was converted into ketone **L3** by a known [42, 43] reaction sequence including oxidative glycol cleavage, and Horner-Emmons reaction with **L2**. This ketone was subjected to a Witting reaction with phosphonium salt **L4**, and then selective removal of acetonide moiety gave diol **L5**, which was selectively protected with a TBDPS group in primary position followed by esterification of remaining secondary alcohol. This sequence provided ester **L6** in good yield.



Conditions: (i) $Pd(OAc)_2$, LiCl, LiOAc, MnO_2, benzoquinone, AcOH, pentane, 93%; (ii) K_2CO_3, MeOH, 43%; (iii) mCPBA, EtOAc, Et_2O, 67%; (iv) NaH, PMBCl, 85%; (v) (2-Th)Li(Cn)Cu, 70%; (vi) Na/NH_3, tBuOH, 91%; (vii) dimethoxypropane, PPTS, acetone, 92%; (viii) Swern or Dess-Martin, 73-77%; (ix) $ClCH_2I$, nBuLi, 64%; (x) 2M HCl, THF; (xi) NaOH, EtOH, 70% (2 steps).

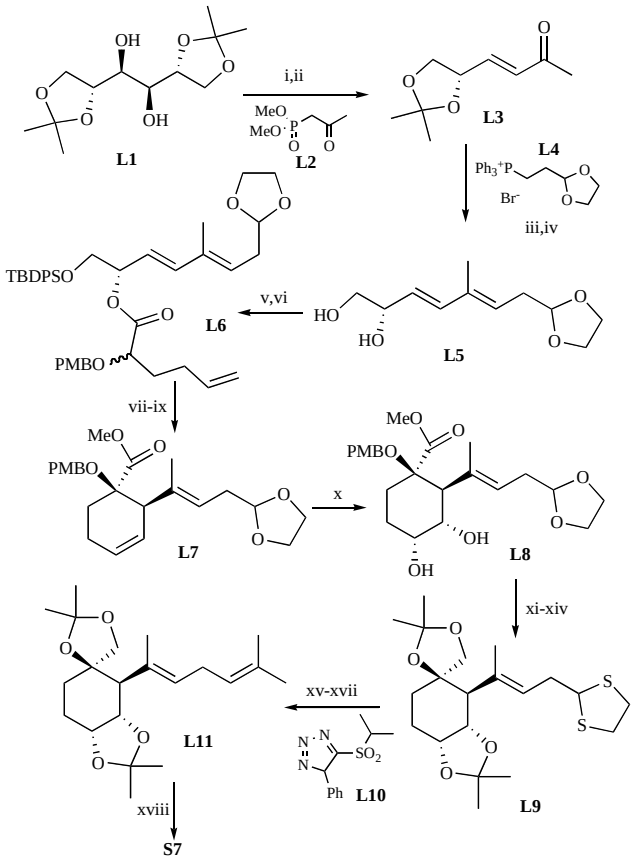
Scheme 10. Simpkins' formal synthesis of (±)-6.

Claisen-Ireland rearrangement of this ester under conditions optimized by the same team [44] followed by hydrolysis and RCM provided the crude cyclic acid which was treated immediately with diazomethane to afford methyl ester **L7**. The authors tried different dihydoxylation conditions but only under Sharpless conditions were they able to obtain the desired glycol **L8** with excellent diastereoselectivity and acceptable yield. The original idea was to perform a Wittig reaction after dioxolane hydrolysis to introduce the trisubstituted double bond, however under acidic conditions the desired aldehyde quickly isomerized to the α,β unsaturated compound; for this reason, it was necessary to transform the dioxolane ring in a dithioacetal after acetylation. Treatment of **L8** with 1,2-ethane dithiol and $BF_3.Et_2O$ also cleaved the PMB ether. After that, reduction of methyl ester and protection of both diols as acetonides provided **L9**, which could be deprotected with $HgO/HgCl_2$ and then subjected to a Julia-Kocienski olefination with sulfone **L10**, yielding the trisubstituted olefin **L11**. Acetonide cleavage with HCl, gave **S7** intermediate, which can be transformed into **S8** and then into fumagillin or fumagillol (**1** and **6**, Fig. **1**) as described above.

In 2005 Mootoo [45] applied an oxocarbenium ion cyclization to the synthesis of intermediate **HMS3**. This synthesis constituted a formal synthesis of fumagillin (**1**, Fig. **1**) (Scheme **12**).

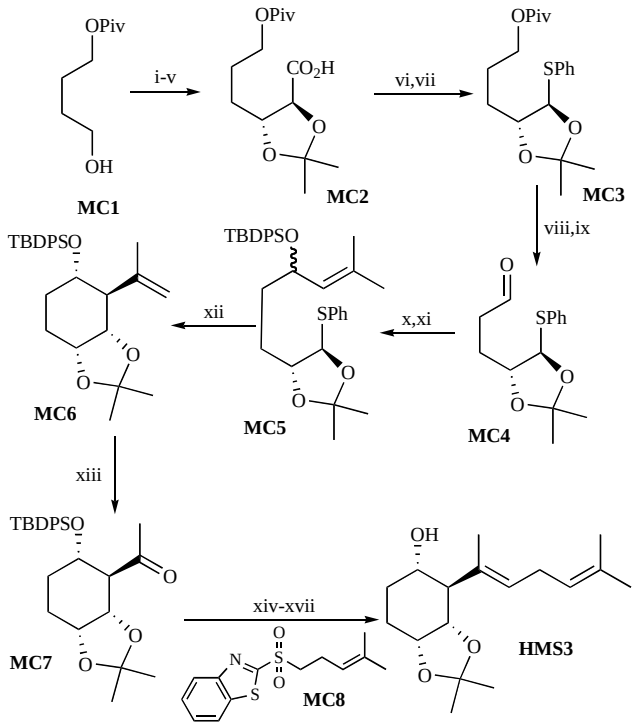
Oxidation of alcohol **MC1** with PCC followed by olefination of the obtained aldehyde provided (*E*)-1-pivaloyloxy-2-hexenoate, which was treated under Sharpless dihydroxylation conditions then acetonization of the diol produced **MC2** in good yield and an enantiomeric excess greater than 95%. The treatment of **MC2** with iodobenzene diacetate under anhydrous conditions provided an inseparable mixture of acetoxy isopropylidene acetals; exchange of acetate group by thiophenol produced a 9:1 separable mixture of sulfides in favor of the desired isomer **MC3**. This was then deprotected and oxidized to yield aldehyde **MC4**.

Addition of 2-propenyl lithium gave a 1:1 mixture of alcohol isomers, the mixture was protected with a TBDPS group (**MC5**) and then the key step of the synthesis was carried out using methyl triflate and 4-methyl-2,6-di-*tert*-butylpyridine; after cyclization the obtained mixture was separable and **MC6** was produced in 44% yield. After ozonolysis, a Sylvestre Julia olefination gave an insepa-



Conditions: (i) $NaIO_4$; (ii) Base, **L2**, >95%; (iii) nBuLi, THF, **L4**, 70%; (iv) AcOH, TFA, 76%; (v) TBDPSCl, TEA, DMAP, THF, 82%; (vi) DCC, DMAP, CH_2Cl_2, 80%; (vii) KHMDS, Tol.; (viii) TMSCl, 77% (2 steps); (ix) Grubbs I, CH_2Cl_2, then CH_2N_2, 94%; (x) AD-mix-α, $MeSO_2NH_2$, tBuOH, H_2O, Acetone, 50%; (xi) Ac_2O, DMAP, Pyr., CH_2Cl_2, 86%; (xii) $(CH_2SH)_2$, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 95%; (xiii) $LiAlH_4$, THF; (xiv) $Me_2C(OMe)_2$, CSA, 79% (2 steps); (xv) HgO, $HgCl_2$, Acetone/H_2O; (xvi) KI; (xvii) LiHMDS, THF, **L10**, 42% (3 steps); (xviii) HCl, THF/H_2O, 71%.

Scheme 11. Langlois' formal synthesis of **6**.



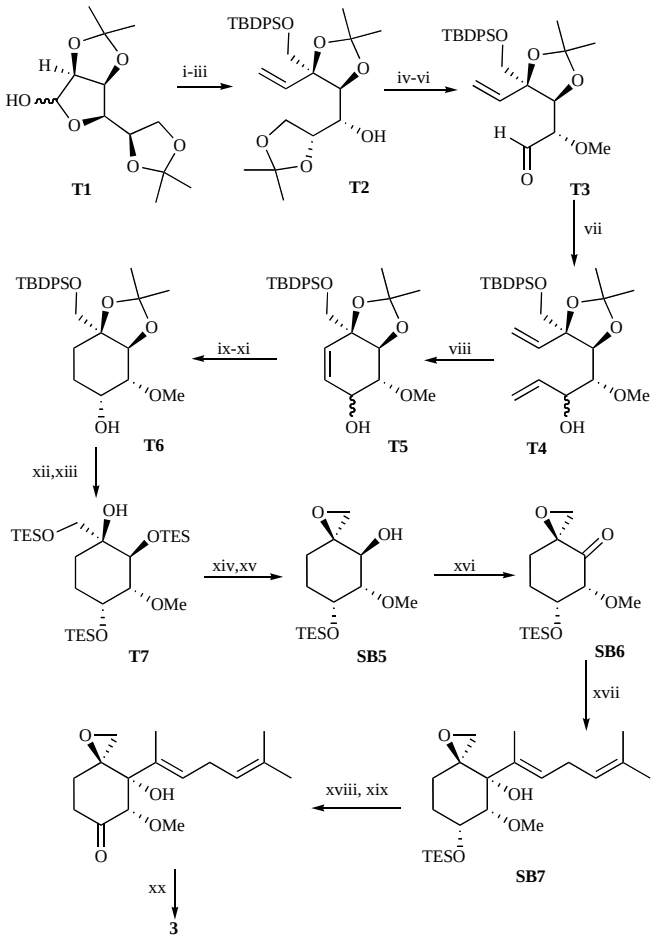
Conditions: (i) PCC, CH_2Cl_2; (ii) $Ph_3P{=}CHCO_2Me$, CH_3CN, 88% (2 steps); (iii) AD-mix-β, *t*BuOK-H_2O, $MeSONH_2$, 90%; (iv) $(MeO)_2CMe_2$, CSA, CH_2Cl_2; (v) THF-KOH aq., 88% (2 steps); (vi) $PhI(OAc)_2$, I_2, CH_2Cl_2; (vii) PhSH, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 80% (2 steps); (viii) DIBALH, CH_2Cl_2, 94%; (ix) Swern, 94%; (x) 2-methyl-1-bromopropene, *t*BuLi, Et_2O, 61%; (xi) TBDPSCl, imidazole, DMF, 100%; (xii) MeOTf, 2,6-di-*tert*-butyl-4-methylpyridine, molecular sieves, CH_2Cl_2, 44%; (xiii) O_3, CH_2Cl_2/MeOH then PPh_3, 85%; (xiv) **MC8**, LIHMDS, THF, 87%; (xv) TBAF, THF, 100%; (xvi) $[VO(acac)_2]$, TBHP, CH_2Cl_2; (xvii) Ph_2PLi, THF then MeI, 67% (2 steps).

Scheme 12. Mooto's formal synthesis of **1**.

rable mixture of alkenes; unfortunately the *Z* undesired isomer was the main product. Direct isomerization with photochemical or thermal conditions was unsuccessful. However using the Vedejs' two step sequence [46, 47] they were able to obtain **HMS3** in 67% yield starting from the original mixture of homoallyllic alcohols.

Also in 2005 Takahashi *et al.* [48] published a total synthesis of ovalicin (**3**, Fig. **1**) starting from D-mannose (Scheme **13**).

Compound **T1** was obtained from D-mannose using a known procedure [49], then a sequence of reactions including acetal formation, sillylation and Wittig reaction of the resulting silyl ether provided compound **T2**. Methylation, acetal cleavage followed by an oxidative cleavage of the obtained diol produced aldehyde **T3** which was then treated with vinylmagnesium chloride to afford a separable mixture of alcohol isomers **T4**. All subsequent reactions were carried out independently on both isomers, starting from RCM with Grubbs II catalyst, producing cyclohexenol **T5**. Hydrogenation provided alcohol **T6** and its epimer which was subjected to a Mitsunobu reaction to afford the corresponding benzoate, whose benzoyl group was removed using sodium methoxide to yield **T6** in 95% overall yield. To transform **T6** into **T8** the following reaction sequence was performed: first all protecting groups were removed using hydrogen chloride in methanol, then tetraol sillylation with TESCl provided a tri-TES ether **T7**; this was treated with TBAF at low temperature to afford mono TES ether which was transformed in the spiro compound **SB5** after tosylation and intramolecular cyclization.



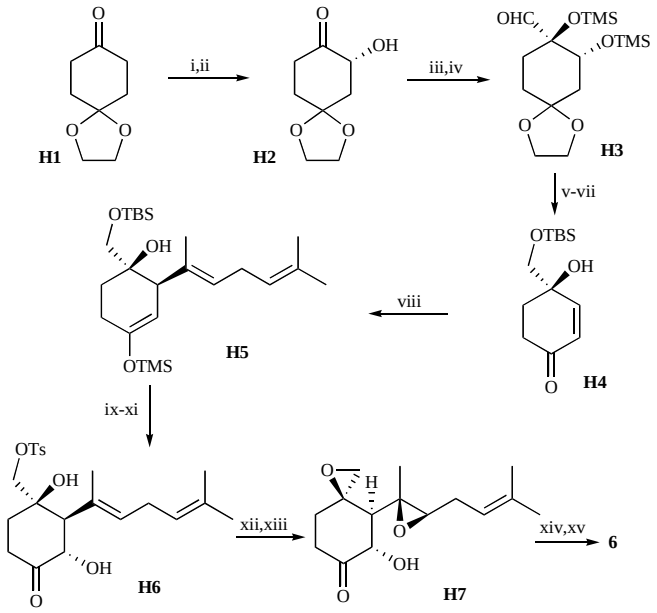
Conditions: (i) K_2CO_3, MeOH, CH_2O 37% aq., 80%; (ii) TBDPSCl, DMAP, Et_3N, CH_2Cl_2, 85%; (iii) $Ph_3P^+CH_3Br^-$, *t*BuOK, toluene, 97%; (iv) NaH, MeI, THF, 97%; (v) AcOH aq., 72%; (vi) $NaIO_4$, THF/H_2O then $(CH_2OH)_2$, quant; (vii) vinylmagnesium chloride, THF, 68%a, 28%b; (viii) Grubbs II, toluene, 94%a, 84%b; (ix) 10%Pd/C, H_2, AcOEt, 99%a, 95%b; (x) DEAD, PPh_3, benzoic acid; (xi) NaOMe, MeOH, 95% (2 steps); (xii) HCl/MeOH, 97%; (xiii) TESCl, Imidazole, DMF, 95%; (xiv) TBAF, THF, 87%; (xv) *p*-TsCl, DMAP, Et_3N, CH_2Cl_2 then, K_2CO_3 MeOH, 93%; (xvi) DMP, $NaHCO_3$, CH_2Cl_2, 98%; (xvii) THF, **CD7**, 85%; (xviii) TBAF, THF, 90%; (xix) TPAP, NMO, 4Å molecular sieve, CH_2Cl_2, 97%; (xx) $[VO(acac)_2]$, *t*BuOOH, benzene/decane, 64%.

Scheme 13. Takahashi's synthesis of **3**.

Oxidation with DMP gave product **SB6** which was already described by Samadi [22], however after alkylation to **SB7** they did not use the same reaction sequence described before. They cleaved the silyl ether then oxidized the secondary alcohol with n-tetrapropyl ammonium perruthenate (TPAP) to obtain **T8** and finally epoxidized the allylic alcohol to produce **3**.

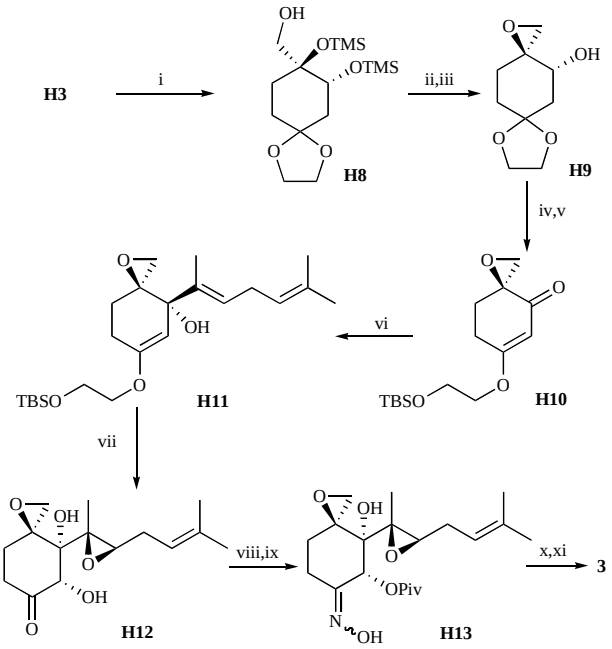
Hayashi *et al.* [50], described an asymmetric synthesis of fumagillol and ovalicin (**6** and **3**, Fugure 1); they applied for the first time the same strategy to the synthesis of these two natural products using a key intermediate **H3** (Schemes **14** and **15**). And it constitutes also the last described synthesis for **6** at present.

The synthesis of key intermediate **H3** started with 1,4-cyclohexadione monoethylene ketal **H1**, which was then subjected to an α-aminoxylation catalyzed by L-proline then hydrogenated to produce **H2** in very good yield and excellent enantiomeric excess. Treatment of **H2** with TMSCN and triethyl amine afforded after 2.5 hours, bis(trimethylsilyloxy)cyanocyclohexane in good yield and excellent diastereoisomeric ratio; this was then reduced with DIBALH to afford aldehyde **H3**, which was then reduced to the corresponding alcohol. Treatment of this alcohol with Amberlyst for several days removed all protecting groups with concomitant dehy-



Conditions: (i) L-proline, PhN=O, DMF, 93%, >99%ee; (ii) Pd/C, H_2, 90%; (iii) TMSCN, Et_3N cat., CH_2Cl_2, 68%; (iv) DIBALH, Et_2O, 72%; (v) DIBALH, CH_2Cl_2; (vi) Amberlyst 15, THF; (vii) TBSCl, Et_3N, DMAP cat., CH_2Cl_2, 57% (3 steps); (viii) **S3**, *t*BuLi, Me_2Zn, THF, then TMSCl, Et_3N, 61%; (ix) DMD, MeOH; (x) TBAF, THF, 74% (2 steps); (xi) TsCl, Et_3N, CH_2Cl_2 71%; (xii) $VO(acac)_2$, TBHP, CH_2Cl_2; (xiii) K_2CO_3, MeOH, 75% (2 steps); (xiv) MeI, Ag_2O CH_3CN, 75%; (xv) K-selectride, THF, 94%.

Scheme 14. Hayashi's total synthesis of **6**.



Conditions: (i) DIBALH, CH_2Cl_2; (ii) MsCl, Et_3N, DMAP, CH_2Cl_2; (iii) K_2CO_3, MeOH, 81% (3 steps); (iv) DMP, CH_2Cl_2 then TLC; (v) TBSCl, imidazole, DMF, 60% (2 steps); (vi) **S3**, *t*BuLi, Et_2O/toluene, 91%; (vii) $VO(OiPr)_3$, TBHP, toluene, 64%; (viii) PivCl, Et_3N, DMAP cat., 84%; (ix) $NH_2OH{\cdot}HCl$, Et_3N, EtOH, 90%; (x) K_2CO_3, MeOH; (xi) MeOTf, 2,6-*t*Bu_2Py, CH_2Cl_2, 72% (2 steps).

Scheme 15. Hayashi's total synthesis of **3**.

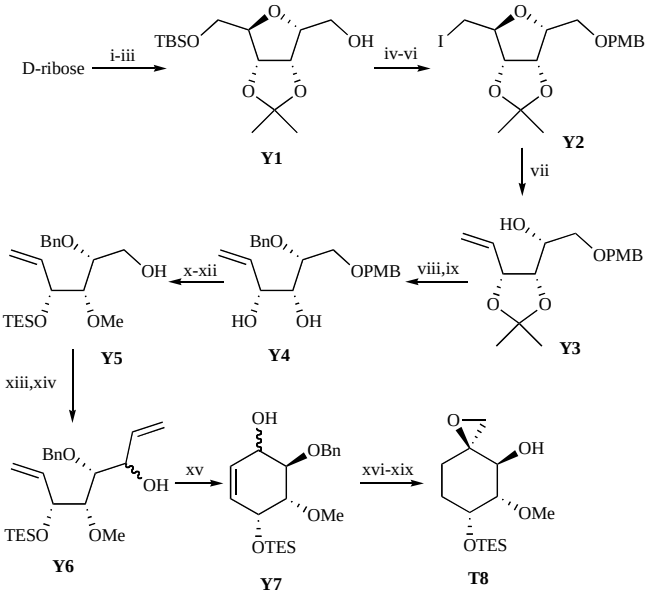
dration affording a cyclohexene diol, which was protected selectively with TBSCl to afford **H4**.

Michael addition of a vinyl zincate prepared from **S3** and trapping the enolate with TMSCl afforded **H5**. This was then oxidized (Rubottom oxidation), then silyl group cleaved and the primary alcohol transformed into the tosylate **H6**. Epoxidation and intramolecular cyclization yielded **H7** which was easily transformed into **6** by methylation and reduction.

In the same paper Hayashi described the synthesis of **3**.

First alcohol **H8** was generated starting from **H3**, then mesylation and intramolecular cyclization afforded **H9**. Oxidation of **H9** with DMP, followed by acid treatment yielded the 3-(2-hydroxyethyloxy)cyclohex-2-enone derivative **H10**. Insertion of the side chain using **S3** provided compound **H11** which was treated with a catalytic amount of $VO(OiPr)_3$ promoting the epoxidation of both the side chain and the enol ether to afford **H12**. Conversion of the alcohol to its methyl ether by traditional methods failed, so the authors decided to use a modified multistep sequence: first transformation into the pivalate, followed by treatment with hydroxylamine yielded oxime **H13**; this was transformed in **3** by a pivalate cleavage and methyl ether formation followed by generation of the ketone with MeOTf.

Another formal synthesis of **3** using carbohydrates as chiral template was published by Yadav in 2007 [51], starting with D-ribose (Scheme **16**).



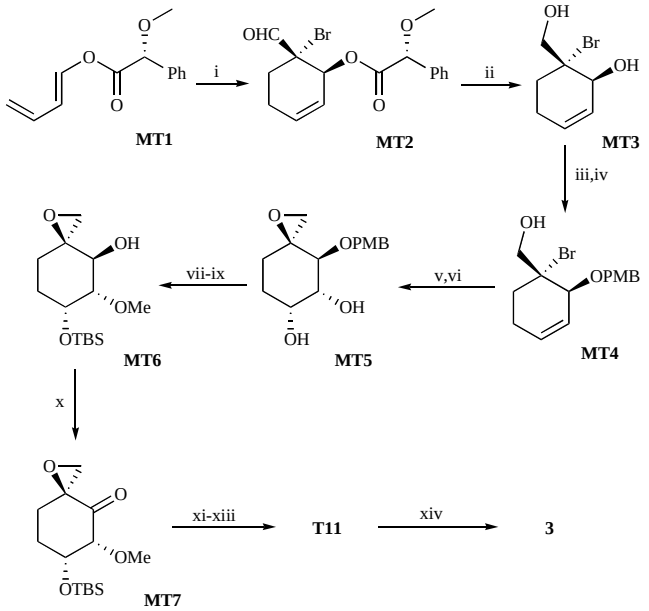
Conditions: (i) 2,2-dimetoxypropane, acetone, PTSA, 72%; (ii) TBSCl, imidazole, CH_2Cl_2, 98%; (iii) $Me_2SO{=}CH_2$, DMSO, 60%; (iv) NaH, PMBBr, Et_2O, 97%; (v) TBAF, THF, 98%; (vi) I_2, PPh_3, imidazole, toluene, 99%; (vii) Zn, EtOH, 95%; (viii) NaH, THF, 85%; (ix) PTSA, MeOH, 98%; (x) TESCl, imidazole, 84%; (xi) NaH, MeI, THF, 95%; (xii) DDQ, CH_2Cl_2/H_2O (8:2), 61%; (xiii) TPAP, NMO, CH_2Cl_2, 80%; (xiv) vinylmagnesium bromide, THF, 75%; (xv) Grubbs I, CH_2Cl_2, 98%; (xvi) IBX, DMSO, CH_2Cl_2, 69%; (xvii) $Pd(OH)_2$, THF, H_2, 87%; (xviii) $MePPh_3I$, *t*BuOK, THF, 87%; (xix) mCPBA, CH_2Cl_2, 85%.

Scheme 16. Yadav's formal synthesis of **3**.

D-ribose was transformed in **Y1** by a known method [52]. The alcohol group was then protected as a PMB ether while he TBS was transformed into the primary iodide **Y2** by TBS cleavage and reaction with I_2 and PPh_3, then transformation into **Y3** was accomplished applying the commonly used protocol for zinc mediated ring opening. Protection by a benzyl group followed by the acetonide cleavage yielded compound **Y4**, which was selectively protected by a TES group at the allylic position followed by methylation and finally PMB cleavage to afford primary alcohol **Y5**. This was oxidized and vinyl group added to the aldehyde to produce the RCM precursor **Y6**. Treatment of this with Grubbs I catalyst yielded cyclohexenol **Y7** in excellent yield. Oxidation, hydrogenation of the double bond, followed by a Wittig reaction and epoxidation provided intermediate **T8** which can be transformed in **3** as described before.

Later the same year Mulzer *et al.* [53] described the application of a Diels-Alder strategy using a chiral auxiliary to the synthesis of ovalicin (**3**, Fig. **1**) (Scheme **17**).

Known compound **MT1** [54] was subjected to a Diels-Alder reaction with 2-bromoacrolein to yield compound **MT2** in good yield and good diastereomeric ratio (8:1). This was then transformed into diol **MT3** with $BH_3.NH_3$. The obtained 1,3-diol was transformed into the *p*-methoxybenzylidene acetal and reduced to the PMB protected alcohol **MT4** with DIBALH. Intramolecular Williamson reaction followed by dihydroxylation yielded **MT5** which was selectively protected and methylated before removal of PMB group.



Conditions: (i) 2-bromoacrolein, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 75%; (ii) $BH_3{\cdot}NH_3$, Et_2O, 89%; (iii) *p*-$MeOC_6H_4CH(OMe)_2$, CSA, CH_2Cl_2, 89%; (iv) DIBALH, CH_2Cl_2, 94%; (v) NaH, THF, MeOH, 98%; (vi) OsO_4 1mol%, NMO, actone/H_2O, 92%; (vii) TBSCl, imidazole, CH_2Cl_2, 89%; (viii) NaH, MeI, THF, 99%; (ix) DDQ, CH_2Cl_2, H_2O, 89%; (x) DMP, $NaHCO_3$, CH_2Cl_2, 92%; (xi) **S3**, *t*BuLi, Et_2O, toluene, 76%; (xii) TBAF, THF, 94%; (xiii) DMP, $NaHCO_3$, CH_2Cl_2, 90%; (xiv) [$VO(acac)_2$], *t*BuOOH, benzene, 71%.

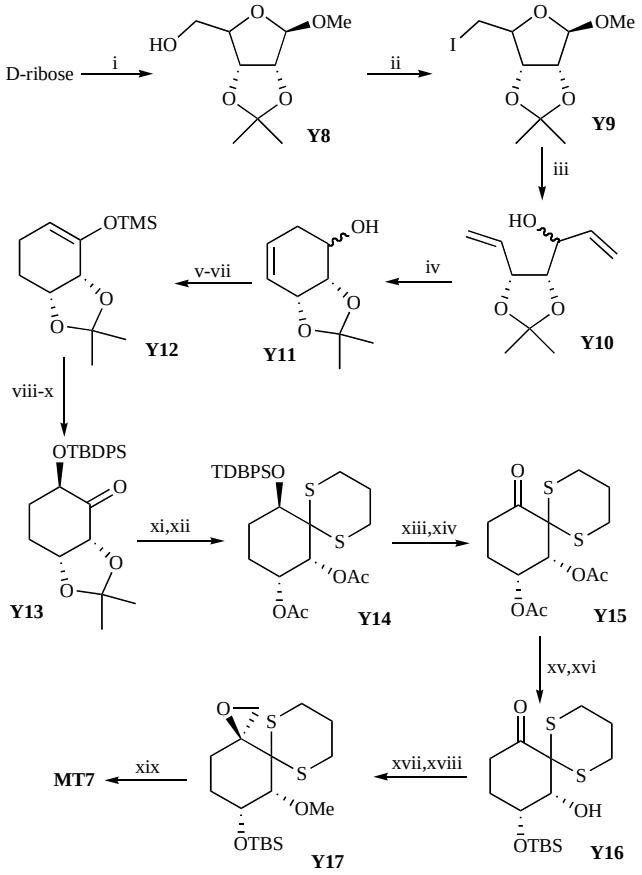
Scheme 17. Mulzer's synthesis of **3**.

Oxidation of **MT6** with DMP yielded compound **MT7** which differs from **T8** only in the silyl group. Thus the strategy used to finish the synthesis was the same as described above: alkylation, deprotection, oxidation and epoxidation.

The last total synthesis of **3** described until now was also published by Yadav [55] in the beginning 2010.

In this synthesis he also used a carbohydrate as chiral template and it presents small differences with the synthesis described in scheme **16**.

First D-ribose was transformed into compound **Y8** by treatment with 2,2-dimethoxypropane, then primary alcohol was transformed ino the corresponding iodide **Y9**. Treatment of **Y9** with allyl bromide and activated Zn, provided in a single step the diene **Y10**, which was treated with Grubbs I catalyst to produce cyclohexenol **Y11**. Reduction of the double bond with $NaBH_4$ using a substoichiometric amount of $NiCl_2{\cdot}7H_2O$, oxidation of the alcohol, followed by generation of the enolate and trapping with TMSCl, provided the silyl enol ether **Y12**. This was oxidized and the obtained alcohol protected with a TBDPS group to afford compound **Y13**. This ketone was protected as a dithiane and the acetonide was cleaved by treatment with $Yb(OTf)_3$ at reflux of CH_3CN. Diol acetylation was accomplished with acetic anhydride to obtain **Y14**. Cleavage of TBDPS group with TBAF followed by oxidation gave the di-acetoxy ketone **Y15**, which was saponified and selectively protected with a TBS group (**Y16**). Corey-Chaykovsky epoxidation followed by methylation yielded compound **Y17**; this was deprotected with mCPBA to afford ketone **MT7** which was transformed in **3** as described above.



Conditions: (i) 2,2-dimethoxypropane, acetone, 2% HCl/MeOH; (ii) PPh_3, imidazole, I_2, toluene; (iii) allyl bromide, Zn, THF/H_2O (4:1), 79%; (iv) Grubbs I, CH_2Cl_2, 90%; (v) $NaBH_4$, MeOH, $NiCl_2{\cdot}7H_2O$; (vi) DMP, $NaHCO_3$, CH_2Cl_2; (vii) LDA, THF, TMSCl; (viii) mCPBA, CH_2Cl_2; (ix) TBAF, 55% (2 steps); (x) TBDPSCl, imidazole, DMAP; (xi) 1,3-propanedithiol, $Yb(OTf)_3$, MeCN, 79%; (xii) Ac_2O, Et_3N, DMAP, CH_2Cl_2, 93%; (xiii) TBAF, THF, 95%; (xiv) DMP, $NaHCO_3$, CH_2Cl_2, 89%; (xv) K_2CO_3, MeOH; (xvi) TBSCl, imidazole, CH_2Cl_2, 88%; (xvii) $Me_3S^+OI^-$, NaH, LiI, DMSO/THF, 50% (xviii) MeI, Ag_2O, Et_2O, 74%; (xix) mCPBA, Ac_2O, Et_3N, H_2O, 59%.

Scheme 18. Yadav's second synthesis of **3**.

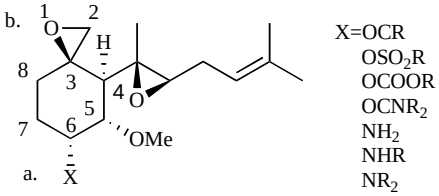
In summary we described in this section all total and formal synthesis reported for fumagillin, ovalicin and fumagillol (**1**, **3** and **6**, Fig. **1**). During the preparation of this manuscript, Hayashi and Yamagushi published a review [56] in total synthesis of **1**, **3** and other natural products with similar structures; another excellent review including also non successful approaches to the synthesis of **1** was published by Langlois [57] in 2003. Synthesis of analogues will be discussed in another section of this manuscript.

STRUCTURE ACTIVITY RELATIONSHIP

In this section we are going to discuss some important results in the synthesis of analogues of **1** and also in the structure determination of MetAP-2-Fumagillin complex, particularly how these studies have helped the scientific community to understand which functions in **1**, **3**, **5** and structurally related molecules (SRM) are crucial for anti-angiogenic activity, and which of them can be transformed without losing activity.

The study of the structure activity relationship for **1** and SRM began in 1992 with a series of three papers published by Marui *et al.* called chemical modifications of fumagillin. In these papers the authors selectively modified the structure of **1**, producing a series of

analogues. Anti-angiogenic activity of these compounds was evaluated and they were able to obtain important conclusions in this area. The authors first focused on modification at position 6 (a. Fig. **4**) and then in modification of spiro epoxide moiety (b. Fig. **4**).



a. Modification of side chain ester
b. Modification of spiro epoxide

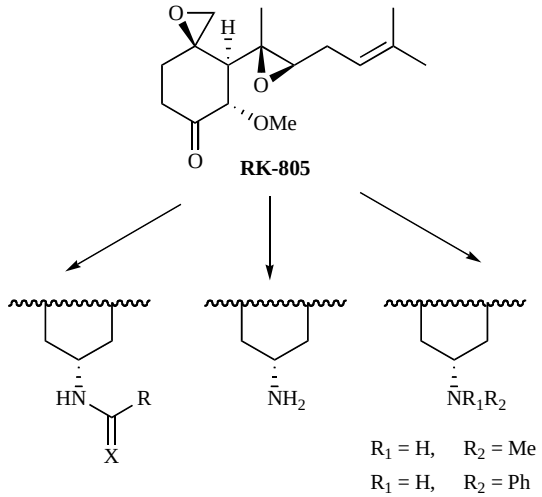
Fig. (4).

In the first paper [58] they worked with fumagillol (**6**, Fig. **1**) (saponification product of **1**) which was transformed into a variety of esters, sulfonic esters, carbonates and carbamates to determine the influence of polyenecarboxylic acid group of **1** in the anti-angiogenic activity.

All molecules they tested exhibited an important inhibitory effect on the proliferation of human umbilical vein endothelial (HUVE) cells, showing that the side-chain ester is not crucial for anti-angiogenic activity and that it can be modified with no loss of activity; all molecules showed more potent anti-angiogenic activity than fumagillin (**1**, Fig. **1**), nevertheless the best results were obtained for TNP-470 (**2**, Fig. **1**) as it was already described [8].

Then authors were focused in the substitution of 6-OH group by a amino group [59]. They anticipated that substitution of the hydroxyl group might result in some attractive changes in biological activity, due to changes in pharmacological behavior, and physicochemical properties.

So they oxidized **6** with CrO_3-Pyr to obtain **RK-805** intermediate, which they used as starting material as indicated in the Scheme **19**.



Scheme 19. C-6 aminated analogues of **1** and **6**.

All compounds tested showed comparable biological activity to **1**, except for **M1** (Fig. **5**); which showed a lower activity. It was demonstrated by MNDO calculations that this compound adopted a different conformation from the other analogues; this caused a different relative location of two epoxides and it was suggested that these differences might play an important role in the expression of anti-angiogenic activity. After these results, the authors turned their interest in modification of the spiro epoxide to determine its importance in anti-angiogenic activity.

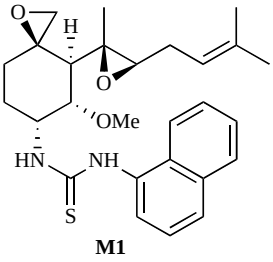
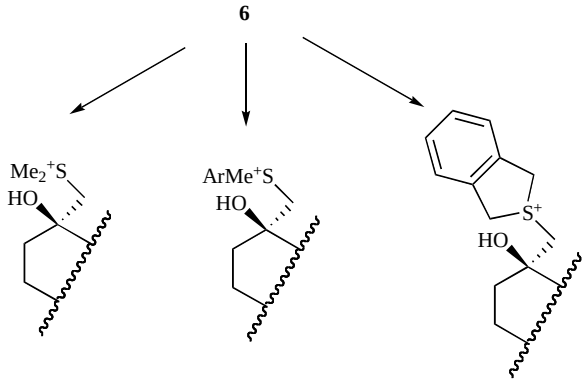


Fig. (5).

It was already known that the spiro-epoxide of **6** is more reactive to the nucleophilic attack of hydrides than the side chain epoxide [2], based on that information and their own former results, Marui *et al*. [60] decided to convert the spiro epoxide into sulfonium salts as indicated in Scheme **20**.



Scheme 20. Sulfonium analogues of **1** and **6**.

They expected that sulfonium moiety would increase molecule hydrophilicity and that such a synthetic equivalent to an epoxide would show similar biological activities.

Yamaoka *et al*. [61] have measured the weight of mice as an indicator of toxicity, they reported a change in body weight of mice when **2** was used. Among different sulfonium analogues, **M2** showed the best toxicity and solubility profiles compared with **2**. Weight of mice increased similarly to the control mice using **M2**; this may suggest low toxicity of the sulfonium compound.

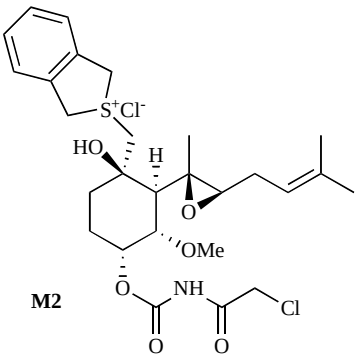
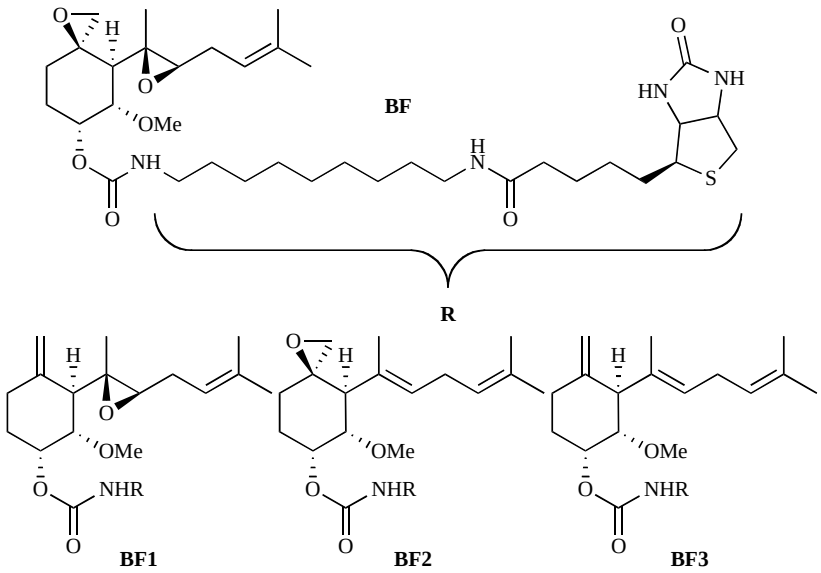


Fig. (6).



Scheme 21. Biotin-fumagillin conjugate and analogues.

Continuing with the study of epoxides influence in biological activity of **1** or SRM, Griffit and coworkers [62] published the structural modification of **1**, producing biotin-fumagillin conjugate (**BF**) and three analogues (**BF1**, **BF2** and **BF3**) these analogues differed in the existence of epoxide moiety: **BF1** contained only the side chain epoxide, **BF2** only the spiro epoxide and **BF3** did not have any epoxides.

In comparison with **BF**, removal of spiro epoxide (**BF1**) caused a decrease in potency to 1/1000 original levels. This results suggested that this epoxide plays a crucial role in the binding of **1** to MetAP-2, it is also in accord with Marui's observations.

BF3 also appeared to be ineffective against angiogenesis. However **BF2** showed a comparable activity to **BF**, suggesting that the side chain epoxide can be suppressed without losing anti-angiogenic activity, and it is not important in the binding with MetAP-2.

Recently isolation of 5-demethoxyfumagillol [63] (**8**, Fig. **7**) and 5-demethoxifumagillin (**7**, Fig. **7**) permitted to determine influence of 5-methoxy group.

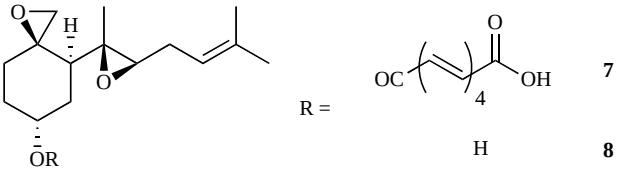


Fig. (7).

Results showed that **8** is a potent angiogenesis inhibitor. Nevertheless, its activity is at least 1000 times lower than **2**; this suggest that 5-methoxy group is important but not crucial in anti-proliferation activity.

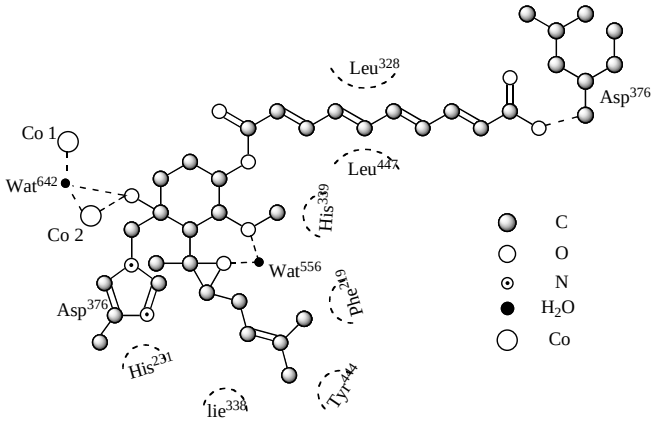
Perhaps the most important result in SAR investigations was published in 1998 by the team of professor Clardy [64]. They were able to determine the crystal structure of MetAP-2 complexed with **1**. This structure confirmed results described above and showed the different interactions of **1** with the active site of the enzyme as showed in Scheme **22**.

The active site of MetAP-2 is a deep pocket with two cobalt atoms at its base and a totally covered side pocket that seems to serve as the specificity pocket for the NH_2-terminal methionine side chain of natural substrates.

In the absence of **1** the cobalts are coordinated by Asp251, Asp262, His331, Glu364, Glu459 and a water molecule.

When **1** is present, the cobalts are forming a hydrogen bond whit an equidistant molecule of water which is also forming a hydrogen bond with the oxygen of the cleaved epoxide.

On the other hand the carbon atom of former spiro epoxide forms a new C-N bond with a nitrogen atom of imidazole in His231. Another molecule of water forms two hydrogen bonds with methoxy oxygen and the oxygen of remained epoxide which occupies the completely covered pocket near to the active site. The long unsaturated side chain makes two hydrophobic contacts with Leu328 and Leu447. Finally terminal carboxyl group of side chain forms a hydrogen bond with Asp376.

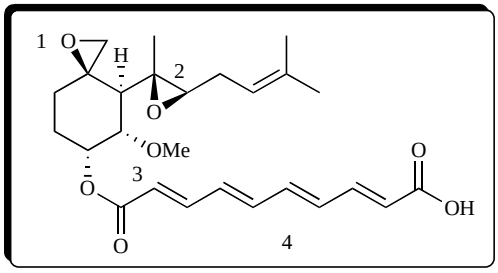


Scheme 22. Schematic structure of 1 in the active site of MetAP-2.

As is visible in the scheme, the polyene chain only has a few hydrophobic interactions with the active site of the enzyme. Using a MetAP-2 model a Korean team published [65] the synthesis of some very portent analogues of **1**. These analogues have non polar (hydrophobic) substituents instead the polyene chain. The results obtained showed the activity of some of these analogues is comparable and even better than the activity of **2**.

Summarizing, the spiro epoxide in **1** or SRM seems to be essential for the anti-angiogenic activity, but it can be replaced by a sulfonium salt without significant decrease of its activity, however it is possible that the remaining hydroxyl group at C3 cyclize to reform

the epoxide ring. This hypothesis remains to be proven and also if it is possible to change this epoxide for other functional groups to give high affinity and to improve the pharmacokinetic properties of analogues.



1. Essential, but can be replaced by an epoxide precursor and maybe other electrophilic functional groups.
2. Not essential, polar functional groups which can form hydrogen bonds can be used.
3. Apparently can be suppressed without significant effect.
4. Can be removed or transformed, better if hydrophobic chains are used.

Fig. (8).

The polyene side chain can be transformed and apparently it is better if it contains hydrophobic groups. The 5-methoxy group is not essential for activity but it is necessary to have a polar group in this position. The same observations were made for the side chain epoxide.

On the other hand the conformation of the analogues is very important for anti-angiogenic activity.

SYNTHESIS OF ANALOGUES

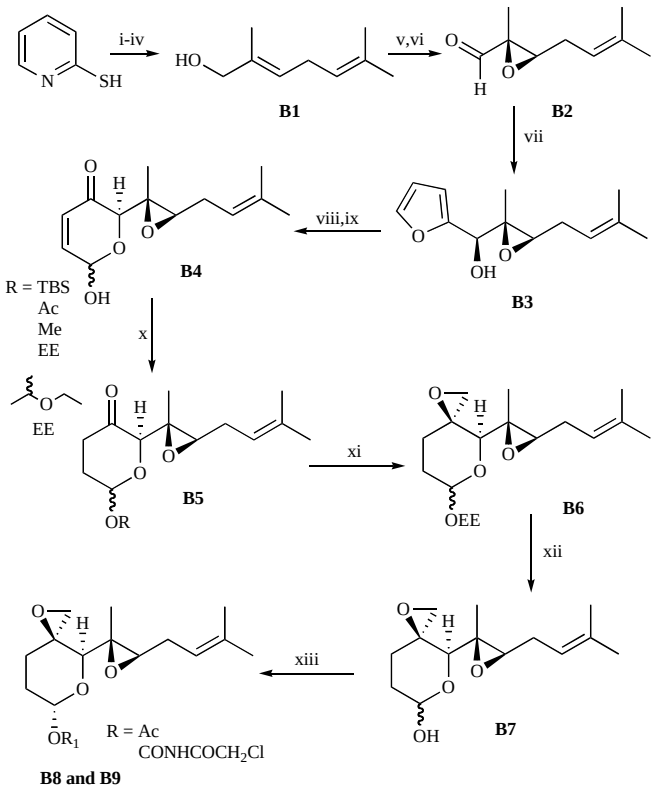
In the previous section we reviewed different synthesis of analogues which permitted to establish the SAR; however those were semi-synthesis starting in general with **6** or even **1**. Here we are going to show other synthesis of analogues which used a different approach from total synthesis or semi-synthesis described above and they will be presented in chronological order.

It has to be noted that synthesis of fumagillol esters has been extensively studied and here we are not going to present these results.

A novel class of non-carbocyclic ring analogues of **1** and **2** have been enantioselectively and efficiently synthesized by Baldwin *et al.* [66]. They anticipate that oxygen atom in position 5 can also form an hydrogen bond with water and the side chain epoxide.

They formed the six-member ring core by a well-known rearrangement [67] of hydroxy furans like **B3**, to piranones like **B4**.

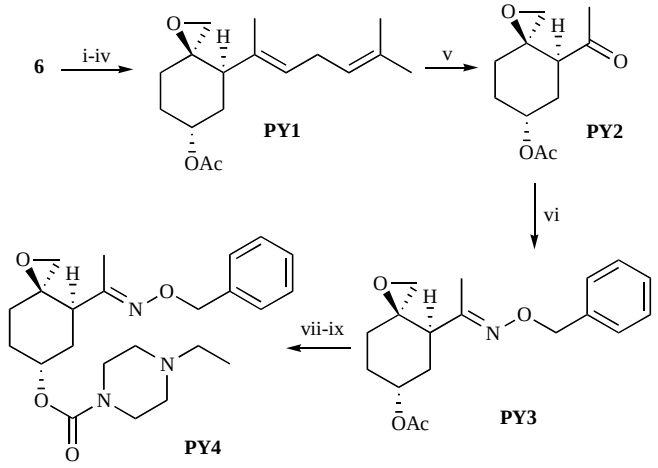
The synthesis started with the preparation of alcohol **B1** in four steps starting from commercially available 2-mercaptopyridine. Asymmetric epoxidation under Sharpless conditions followed by Swern oxidation of the primary alcohol provided α-epoxyaldehyde **B2** in good yield and >98% ee. Then addition of lithiated furan gave the desired carbinol **B3**, which was treated under the conditions reported by Martin [68], to afford pyranone **B4** as a mixture of α and β isomers. Reduction of the double bond with Raney-Ni followed by protection of hydroxyl group with different protecting groups yielded saturated piranones **B6**. Then epoxidation of these ketones was accomplished with bromomethyllithium. Removal of ethoxyethyl group was easily carried out with PPTS in acetone/water at 45°C. Hemiacetal **B7** was then converted into carbamate **B8** or acetate **B9** under classical conditions. The α/β was >10:1 in both cases. This was the first synthesis of analogues of **1** different from esters of **6**. Unfortunately the results of biological activity evaluation of these analogues have not been reported.



Conditions: (i) 3-chloro-2-methylpropene, NaOEt; (ii) phenyl bromide, nBuLi; (iii) mCPBA, Et_2NH, 53% (4 steps); (v) D-(-)-DIPT, $Ti(OiPr)_4$, TBHP; (vi) Swern, 73% (2 steps); (vii) furan, nBuLi, 59%; (viii) $[VO(acac)_2]$, TBHP, 51%; (ix) RX, base or ethylvinyl ether, PPTS, 53-84%; (x) Raney-Ni, 65-92%; (xi) CH_2Br_2, nBuLi, 79-82%; (xii) PPTS, acetone, water, 85%; (xiii) $ClCH_2COCOCNO$, 64%, or Ac_2O, pyr, 72%.

Scheme 23. Baldwin's synthesis of non-carbocyclic analogues of **1**.

Analogue **PY4** was synthesized by Pyum *et al*. [69] and it was found to exhibit similar anti-angiogenic effects as **2**.



Conditions: (i) LiCl, AcOH, THF, 96%; (ii) Ac_2O, DMAP, CH_2Cl_2, 86%; (iii) WCl_6, nBuLi, THF, 92%; (iv) *t*BuOK, THF, 77%; (v) O_3, CH_2Cl_2, MeOH then Me_2S, 74%; (vi) NH_2OBn, PTSA, 4 Å sieves, THF, 40% (*E*-isomer), 17% (*Z*-isomer); (vii) K_2CO_3, MeOH, 100%; (viii) ClCOOPh, DMAP, CH_2Cl_2, 100%; (ix) $HN(CH_2CH_2)_2NEt$, CH_2Cl_2, 78%.

Scheme 24. Pyun's synthesis of oxime analogue of **1**.

Compound **6** was transformed into acetate **PY1** in a four- step sequence including opening of spiro epoxide, acetylation, reduction of side chain epoxide and intramolecular cyclization in basic medium to reform the spiro epoxide. Ozonolysis provided ketone **PY2**

which was then transformed in oxime **PY3** as a separable mixture of *Z* and *E* isomers. Then saponification, formation of benzyl carbonate and substitution for piperazine moiety yielded **PY4**. Anti-angiogenic activity was tested and the results were comparables to these obtained for **2**, confirming that the side chain epoxide is not crucial for biological activity and it can be replaced by other functional groups.

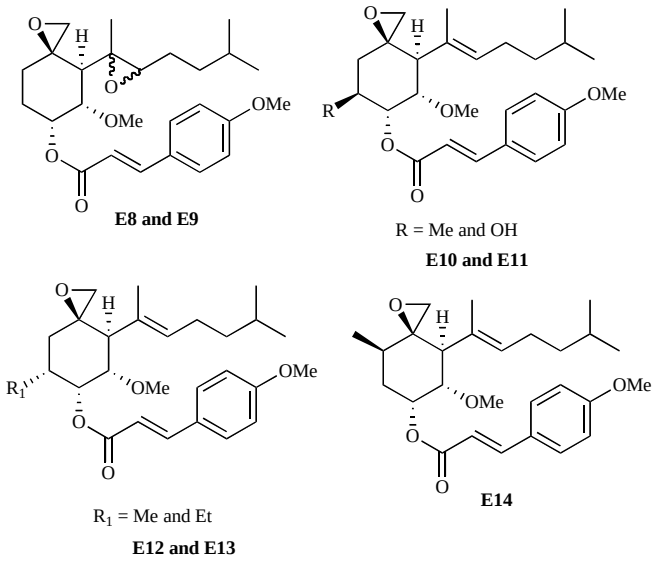
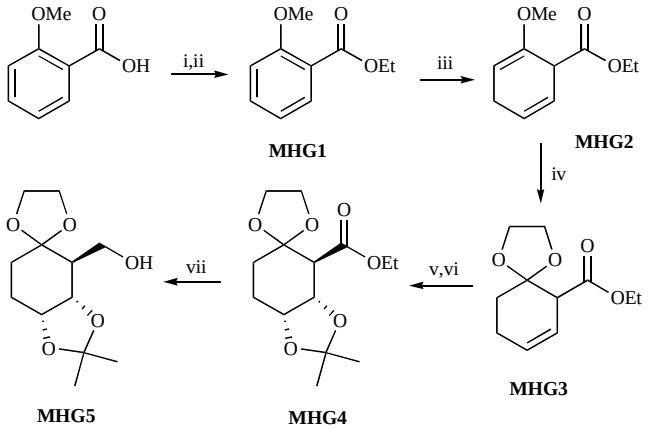


Fig. (9).

Using the same strategy based on RCM of their total synthesis Eustache *et al.* [70] obtained a series of analogues with modifications in the side chain and in the western part of molecule (Fig. **9**).

Their results suggested that even if the side chain epoxide is dispensable, using natural configuration epimer may orient the molecule in a different way rendering difficult the recognition by MetAP-2; however more research to check the validity of this hypothesis is necessary. Also utilization of **E13** (very low activity) showed that modification of western part of the molecule is limited to the introduction of small groups.

The last report we are going to describe here was published in 2005 [71]. The authors described a new strategy for the synthesis of **1** and **3** analogues based on a Birch reduction to generate the unsaturated cyclohexene ring (Scheme **25**).



Conditions: (i) oxalyl chloride, DCM; (ii) EtOH, pyr, DCM, 100% (2 steps); (iii) Na/NH_3, THF, 84%; (iv) ethylen glycol, PTSA, DMF, 70%; (v) NMO, OsO_4, acetone/H_2O; (vi) 2,2-dimethoxypropane, PTSA, DMF, 85% (2 steps); (vii) LAH, THF, 95%.

Scheme 25. Giannis' synthesis of **1** and **3** analogues.

Esterification of commercially available *o*-methoxy benzoic acid gave compound **MHG1** in quantitative yield; this ester was reduced with sodium in liquid ammonia according to the Birch procedure to provide **MHG2** in good yield. Treatment of **MHG2** with ethylene glycol in acidic medium yielded compound **MHG3**; this protected keto ester was then dihydoxylated and the obtained diol protected as its corresponding acetonide to give **MHG4**. Finally reduction of the ester with LAH gave the analogues precursor **MHG5**.

Different protocols were used to transform this precursor into the various analogues shown in Fig. (**10**).

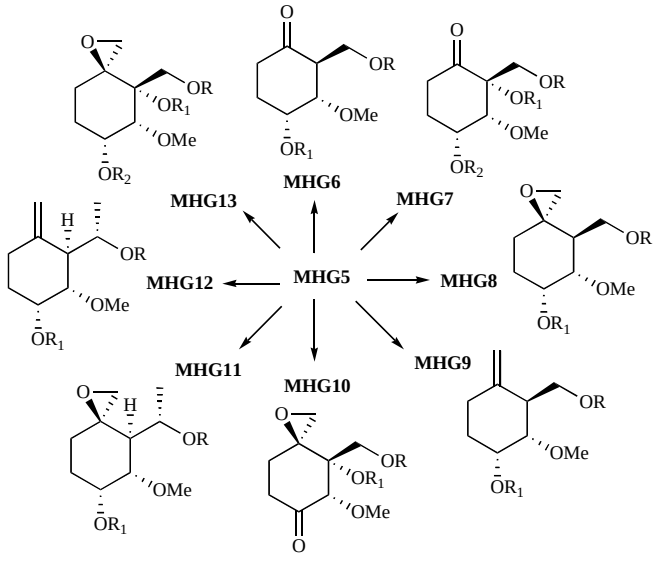


Fig. (10).

First the alcohol **MHG5** could be directly etherified and analogues like **MHG6**, **MHG8** and **MHG9** were obtained. It has to be mentioned that generation of spiro epoxide, methylation and introduction of the ether chain were accomplished by different methods already described here.

Also **MHG5** was oxidized and RMgBr reagents added to generate a secondary alcohol which after etherification provided analogues like **MHG11** and **MHG12.**

Finally dehydration of primary alcohol **MHG5**, followed by epoxidation and opening of this oxirane ring with oxygenated nucleophiles, provided analogues like **MHG7**, **MHG10** and **MHG13**.

This series of analogues were tested *in vitro* to evaluate their anti-angiogenic activity. And a very interesting result was obtained. Compound **MGH14** (Fig. **11**) showed the best angiogenesis inhibition, showing that maybe a ketone can be also a substitute for the spiro epoxide ring, and that the side chain oxirane can be substitutes by an ether.

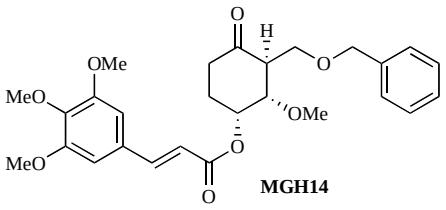


Fig. (11).

OTHER STRUCTURALLY RALATED NATURAL PRODUCTS AND IMPORTANT ASPECTS ABOUT TNP470

In this section we are going to present some important aspects about the molecules described before. Also we are going to present

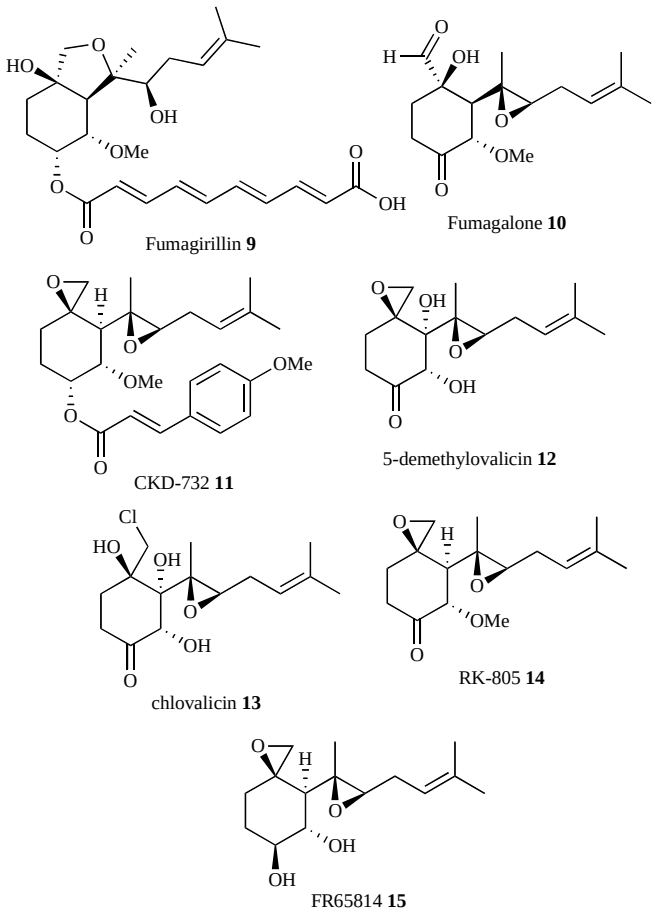


Fig. (12).

some natural products with similar structures to fumagillin (**1**, Fig. **1**) isolated more recently and which have also shown interesting biological properties.

Compound **2** was prepared in 1990 [8] since that time it has been extensively studied and it was the first fumagillin analogue to reach the stage of clinical trials and to be used in humans as anti-angiogenic drug. However it has encountered some problems. Among them are short serum half-life and dose-limiting neurotoxicity [72-74] sometimes attributed to the spiro epoxide ring.

In Fig. (**12**) are shown other structurally related natural products as well as other semi-synthetic analogues.

Fumagirillin (**9**, Fig. **12**) was isolated together with **1** in 2004 [75] from a strain fungus *Aspergillus fumigatus* and biological testing against the murine leukemia cell. Line P388 established that **9** was not cytotoxic (IC_{50}>125 μg/mL).

Fumagallone (**10**, Fig. **12**) is a semi-synthetic analogue of **1**, it was synthesized in 2003 by Liu and coworkers [76] and it exhibited inhibition of MetAP-2 and endothelial cell proliferation. It was also demonstrated by dialysis and competition essays that **10** binds of MetAP-2 in reversible manner.

CKD-732 (**11**, Fig. **12**) (a semi-synthetic analogue) [65] is at this time undergoing clinical trials and exhibited better effectiveness and less toxicity than **2**.

5-demethylovalicin (**12**, Fig. **12**) was isolated in 2002 [77] from the fermentation broth *Chrysosporium lucknowense* and showed to be as potent as **3** in angiogenesis inhibition.

Chlovalicin (**13**, Fig. **12**) was isolated in 1996 by Omura [78] from *Sporothrix* sp. FO-4669 I; it is an antibiotic and showed an inhibitory effect in Interleukin-6 (IL-6) dependent cell growth and Immunoglobulin E (IgE) production. **13** was expected to be a new type of anti-allergic drug possessing the inhibitory effects of both IgE production and IL-6 activities.

RK-805 (**14**, Fig. **12**) was isolated from fungus *Neosartorya* sp. [79] and showed also anti-angiogenesis activity.

FR65814 (**15**, Fig. **12**) [80] isolated in 1988 from the cultured broth of *Penicillium jensenii* F-2883 regardless of its similar structure with Fig. (**12**) molecules it showed a very different biological activity. It exhibited an excellent activity as immunosuppressant.

SUMMARY

We have shown an overview on fumagillin and SRM synthesis, biological activity evaluation, structure activity relationship as well as different efforts made to synthesize more potent analogues. Today the mechanism of action and the SAR is well known. This is an extremely important topic in research for new anti cancer drugs and will certainly continue to grow.

ACKNOWLEDGEMENTS

Author acknowledges the Ecole Polytechnique (Palaiseau, France) for financial support, Dr Joëlle Prunet (Department of Chemistry, University of Glasgow) and Dr. Bill Hawkins (DCSO, Ecole Polytechnique), for the careful reading of this manuscript and their valuable comments.

CONFLICT OF INTEREST

None declared.

REFERENCES

[1] Elbe, T. E.; Hanson, F. R. Fumagillin, an antibiotic from Aspergillus fumigatus H-3. *Antibiot. Chemother.*, **1951**, *1*, 54-58.
[2] Tarbell, D. S.; Carman, R. M.; Chapman, D. D.; Cremer, S. E.; Cross, A. D.; Huffman, K. R.; Kunstmann, M.; McCorkindale, N. J.; McNally, J. G.; Rosowsky, A.; Varino, F. H. L.; West, R. L. The Chemistry of Fumagillin1. *J. Am. Chem. Soc.*, **1961**, *83*(14), 3096-3113.
[3] McCorkindale, N.; Sime, J. G. Configuration of Fumagillin. *Proc. Chem. Soc.*, **1961**, 331.
[4] Sigg, H. P.; Weber, H. P. Isolierung und Strukturaufklärung von Ovalicin. *Helv. Chim. Acta*, **1968**, *51*(6), 1395-1408.
[5] Bollinger, P.; Sigg, H.-P.; Weber, H.-P. Die Struktur von Ovalicin. *Helv. Chim. Acta*, **1973**, *56*(3), 819-830.
[6] Bernier, S. G.; Lazarus, D. D.; Clark, E.; Doyle, B.; Labenski, M. T.; Thompson, C. D.; Westlin, W. F.; Hannig, G. A methionine aminopeptidase-2 inhibitor, PPI-2458, for the treatment of rheumatoid arthritis. *Proc. Natl. Acad. Sci. U.S.A.*, **2004**, *101*(29), 10768-10773.
[7] Cooper, A. C.; Karp, R. M.; Clark, E. J.; Taghizadeh, N. R.; Hoyt, J. G.; Labenski, M. T.; Murray, M. J.; Hannig, G.; Westlin, W. F.; Thompson, C. D. A Novel Methionine Aminopeptidase-2 Inhibitor, PPI-2458, Inhibits Nonâ€"Hodgkin's Lymphoma Cell Proliferation In vitro and In vivo. *Clin. Cancer Res.*, **2006**, *12*(8), 2583-2590.
[8] Ingber, D.; Fujita, T.; Kishimoto, S.; Sudo, K.; Kanamaru, T.; Brem, H.; Folkman, J. Synthetic analogues of fumagillin that inhibit angiogenesis and suppress tumour growth. *Nature*, **1990**, *348*(6301), 555-557.
[9] WHO, *National Cancer Control Programmes: policies and managerial guidelines* 2nd ed.; World Health Organisation: Geneva, **2002**.
[10] Katznelson. H; Jamieson. C, A. Control of Nosema Disease of Honeybees with Fumagillin. *Science*, **1952**, *115*, 70-71.
[11] Molnar. K; Baska. F; Szekely. C. Fumagillin, an efficacious drug against renal sphaerosporosis of the common carp Cyprinus carpio. *Dis. aquat. Org.*, **1987**, *2*, 187-190.
[12] Hedrick. R, P.; Groff. J, M.; Foley. P; McDowell. T. Oral administration of Fumagillin DCH protects chinook salmon Oncorhynchus tshawytscha from experimentally-induced proliferative kidney disease. *Dis. aquat. Org.*, **1988**, *4*, 165-168.
[13] Molina, J.-M.; Tourneur, M.; Sarfati, C.; Chevret, S.; de Gouvello, A.; Gobert, J.-G.; Balkan, S.; Derouin, F.; the Agence Nationale de Recherches sur le SIDA 090 Study Group. Fumagillin Treatment of Intestinal Microsporidiosis. *N. Engl. J. Med.*, **2002**, *346*(25), 1963-1969.
[14] McCowen, M. C.; Callender, M. E.; Lawlis Jr, J. F. Fumagillin (H-3), a new Antibiotic with Amebicidal Properties. *Science*, **1951**, *153*, 202-203.
[15] Killough. J, H.; Magill. G, B.; Smith. R, C. The Treatment of Amebiasis with Fumagillin. *Science*, **1952**, *115*, 71-72.

[16] Griffith, E. C.; Su, Z.; Turk, B. E.; Chen, S.; Chang, Y.-H.; Wu, Z.; Biemann, K.; Liu, J. O. Methionine aminopeptidase (type 2) is the common target for angiogenesis inhibitors AGM-1470 and ovalicin. *Chem. Biol.*, **1997**, *4*(6), 461-471.

[17] Sin, N.; Meng, L.; Wang, M. Q. W.; Wen, J. J.; Bornmann, W. G.; Crews, C. M. The anti-angiogenic agent fumagillin covalently binds and inhibits the methionine aminopeptidase,â€‰MetAP-2. *Proc. Natl. Acad. Sci. U.S.A.*, **1997**, *94*(12), 6099-6103.

[18] Nobumoto, W.; Yoshifumi, N.; Tomoyuki, Y.; Atsushi, K.; Hiroyuki, M.; Hideaki, K.; Hiroyuki, O. Fumagillin suppresses HIV-1 infection of macrophages through the inhibition of Vpr activity. *FEBS letters*, **2006**, *580*(11), 2598-2602.

[19] Chen, X.; Xie, S.; Bhat, S.; Kumar, N.; Shapiro, T. A.; Liu, J. O. Fumagillin and Fumarranol Interact with P. falciparum Methionine Aminopeptidase 2 and Inhibit Malaria Parasite Growth In Vitro and In Vivo. *Chem. Biol.*, **2009**, *16*(2), 193-202.

[20] Corey, E. J.; Snider, B. B. Total synthesis of (+-)-fumagillin. *J. Am. Chem. Soc.*, **1972**, *94*(7), 2549-2550.

[21] Corey, E. J.; Dittami, J. P. Total synthesis of (.+-.)-ovalicin. *J. Am. Chem. Soc.*, **1985**, *107*(1), 256-257.

[22] Bath, S.; Billington, D. C.; Gero, S. D.; Quiclet-Sire, B.; Samadi, M. Total synthesis of (–)-ovalicine from L-quebrachitol. *J. Chem. Soc. Chem. Commun.*, **1994**, 1495-1496.

[23] Corey, E. J.; Guzman-Perez, A.; Noe, M. C. Short Enantioselective Synthesis of (-)-Ovalicin, a Potent Inhibitor of Angiogenesis, Using Substrate-Enhanced Catalytic Asymmetric Dihydroxylation. *J. Am. Chem. Soc.*, **1994**, *116*(26), 12109-12110.

[24] Kim, D.; Ahn, S. K.; Bae, H.; Choi, W. J.; Kim, H. S. An asymmetric total synthesis of (-)-fumagillol. *Tetrahedron Lett.*, **1997**, *38*(25), 4437-4440.

[25] Burke, S. D.; Fobare, W. F.; Pacofsky, G. J. Chelation control of enolate geometry. Acyclic diastereoselection via the enolate Claisen rearrangement. *J. Org. Chem.*, **1983**, *48*(26), 5221-5228.

[26] Sato, T.; Tajima, K.; Fujisawa, T. Diastereoselective synthesis of erythro- and threo-2-hydroxy-3-methyl-4-pentenoic acids by the ester enolate claisen rearrangement of 2-butenyl 2-hydroxyacetate. *Tetrahedron Lett.*, **1983**, *24*(7), 729-730.

[27] Kallmerten, J.; Gould, T. J. Enolate claisen rearrangement of glycolate esters. *Tetrahedron Lett.*, **1983**, *24*(47), 5177-5180.

[28] Barco, A.; Benetti, S.; De Risi, C.; Marchetti, P.; Pollini, G. P.; Zanirato, V. Enantioselective formal synthesis of (-)-ovalicin using quinic acid as a chiral template. *Tetrahedron: Asymmetry*, **1998**, *9*(16), 2857-2864.

[29] Lesuisse, D.; Berchtold, G. A. (-)-Methyl 4,5-O-benzylidene-4-epi-shikimate: an intermediate for the synthesis of (-)-chorismic acid and analogs. *J. Org. Chem.*, **1985**, *50*(6), 888-890.

[30] Cummins, C. H.; Coates, R. M. .alpha.-Oxygenation of aldehydes and cyclic ketones by acylation-rearrangement of nitrones. *J. Org. Chem.*, **1983**, *48*(12), 2070-2076.

[31] Coates, R. M.; Cummins, C. H. Preparation of vicinal N-alkylamino alcohols via acylation-rearrangement of nitrones followed by hydride reduction. *J. Org. Chem.*, **1986**, *51*(9), 1383-1389.

[32] Vosburg, D. A.; Weiler, S.; Sorensen, E. J. A Concise Synthesis of Fumagillol. *Angew. Chem. Int. Ed.*, **1999**, *38*(7), 971-974.

[33] Taber, D. F.; Christos, T. E.; Rheingold, A. L.; Guzei, I. A. Synthesis of (â˜)-Fumagillin. *J. Am. Chem. Soc.*, **1999**, *121*(23), 5589-5590.

[34] Taber, D. F.; Sahli, A.; Yu, H.; Meagley, R. P. Efficient Intramolecular C-H Insertion by an Alkylidene Carbene Generated from a Vinyl Chloride. *J. Org. Chem.*, **1995**, *60*(20), 6571-6573.

[35] Hassner, A.; Reuss, R. H.; Pinnick, H. W. Synthetic methods. VIII. Hydroxylation of carbonyl compounds via silyl enol ethers. *J. Org. Chem.*, **1975**, *40*(23), 3427-3429.

[36] Rubottom, G. M.; Vazquez, M. A.; Pelegrina, D. R. Peracid oxidation of trimethylsilyl enol ethers: A facile [alpha]-hydroxylation procedure. *Tetrahedron Lett.*, **1974**, *15*(49-50), 4319-4322.

[37] Brook, A. G.; Macrae, D. M. 1,4-Silyl rearrangements of siloxyalkenes to siloxyketones during peroxidation. *J Organomet Chem*, **1974**, *77*(2), C19-C21.

[38] Boiteau, J.-G.; Van de Weghe, P.; Eustache, J. A New, Ring Closing Metathesis-Based Synthesis of (â˜)-Fumagillol. *Org. Lett.*, **2001**, *3*(17), 2737-2740.

[39] Hutchings, M.; Moffat, D.; Simpkins, N. S. A Concise Synthesis of Fumagillol. *Synlett*, **2001**, *2001*(05), 0661-0663.

[40] Sadhu, K. M.; Matteson, D. S. (Chloromethyl) lithium in an efficient conversion of carbonyl compounds to chlorohydrins or oxiranes. *Tetrahedron Lett.*, **1986**, *27*(7), 795-798.

[41] Bedel, O.; Haudrechy, A.; Langlois, Y. A Stereoselective Formal Synthesis of (-)-Fumagillol. *Eur. J. Org. Chem.*, **2004**, *2004*(18), 3813-3819.

[42] Takano, S.; Ogasawara, K. Synthesis of Versatile Chiral Bulding-Blocks Starting From D-Mannitol. *J. Synth. Org. Chem. Jpn.*, **1987**, *45*(12), 1157-1170.

[43] Ohira, S.; Ida, T.; Moritani, M.; Hasegawa, T. Synthesis of (-)-malyngolide using reactions of alkylidenecarbenes. *J. Chem. Soc. Perkin Trans. 1*, **1998**, (2), 293-297.

[44] Picoul, W.; Urchegui, R.; Haudrechy, A.; Langlois, Y. A novel stereoselective route to a fumagillin and ovalicin synthetic intermediate. *Tetrahedron Lett.*, **1999**, *40*(26), 4797-4800.

[45] Camara, F.; Angarita, J.; Mootoo, D. R. Oxocarbenium Ion Cyclizations for C-Branched Cyclitols: Synthesis of a Relay Intermediate for Fumagillin Analogues. *J. Org. Chem.*, **2005**, *70*(17), 6870-6875.

[46] Vedejs, E.; Snoble, K. A. J.; Fuchs, P. L. Phosphorus betaines derived from cycloheptene and cyclooctene oxides. Inversion of cyclooctene. *J. Org. Chem.*, **1973**, *38*(6), 1178-1183.

[47] Zhang, H.; Mootoo, D. R. Acetal Templates for the Synthesis of trans-2,5-Disubstituted Tetrahydrofurans. *J. Org. Chem.*, **1995**, *60*(25), 8134-8135.

[48] Takahashi, S.; Hishinuma, N.; Koshino, H.; Nakata, T. Synthesis of Ovalicin Starting from d-Mannose. *J. Org. Chem.*, **2005**, *70*(24), 10162-10165.

[49] Witczak, Z. J.; Whistler, R. L.; R. Daniel, J. Synthesis of 3-C-(hydroxymethyl)erythritol and 3-C-methylerythritol. *Carbohydr. Res.*, **1984**, *133*(2), 235-245.

[50] Yamaguchi, J.; Toyoshima, M.; Shoji, M.; Kakeya, H.; Osada, H.; Hayashi, Y. Concise Enantio- and Diastereoselective Total Syntheses of Fumagillol, RK-805, FR65814, Ovalicin, and 5-Demethylovalicin13. *Angew. Chem. Int. Ed.*, **2006**, *45*(5), 789-793.

[51] Yadav, J. S.; Pamu, S.; Bhunia, D. C.; Pabbaraja, S. Formal Total Synthesis of Ovalcin by Carbohydrate Approach. *Synlett*, **2007**, *2007*(06), 0992-0994.

[52] Fréchou, C.; Dheilly, L.; Beaupère, D.; Uzan, R.; Demailly, G. Réaction d'Ylures du soufre sur des sucres réducteurs: Application à la synthèse d'hydroxyméthyl C-glycosides. *Tetrahedron Lett.*, **1992**, *33*(35), 5067-5070.

[53] Tiefenbacher, K.; Arion, Vladimir B.; Mulzer, J. A Diels-Alder Approach to (-)-Ovalicin13. *Angew. Chem. Int. Ed.*, **2007**, *46*(15), 2690-2693.

[54] Trost, B. M.; Chupak, L. S.; Lubbers, T. Short Preparation of (S)-(E)-1-(O-Methylmandeloxy)butadiene. *J. Org. Chem.*, **1997**, *62*(3), 736-736.

[55] Yadav, J. S.; Reddy, P. N.; Reddy, B. V. S. Stereoselective Total Synthesis of (-)-Ovalicin. *Synlett*, **2010**, (03), 457-461.

[56] Yamaguchi, J.; Hayashi, Y. Syntheses of Fumagillin and Ovalicin. *Chem. Eur. J.*, **2010**, *16*(13), 3884-3901.

[57] Picoul, W.; Bedel, O.; Haudrechy, A.; Langlois, Y. Progress in fumagillin synthesis. *Pure Appl. Chem.*, **2003**, *75*(2-3), 235-249.

[58] Marui, S.; Itho, F.; Kozai, Y.; Sudo, K.; Kishimoto, S. Chemical Modification of Fumagillin. I. 6-O-Acyl, 6-O-Sulfonyl, 6-O-Alkyl, and 6-O-(N-Substituted-carbamoyl)fumagillols. *Chem. Pharm. Bull.*, **1992**, *40*(1), 96-101.

[59] Marui, S.; Kishimoto, S. Chemical Modification of Fumagillin. II. 6-Amino-6-deoxyfumagillol and Its Derivatives. *Chem. Pharm. Bull.*, **1992**, *40*(3), 575-579.

[60] Marui, S.; Yamamoto, T.; Sudo, K.; Akimoto, H.; Kishimoto, S. Chemical Modification of Fumagillin. III. Modification of the Spiro-epoxide. *Chem. Pharm. Bull.*, **1995**, *43*(4), 588-593.

[61] Yamaoka, M.; Yamamoto, T.; Masaki, T.; Ikeyama, S.; Sudo, K.; Fujita, T. Inhibition of Tumor Growth and Metastasis of Rodent Tumors by the Angiogenesis Inhibitor O-(Chloroacetyl-carbamoyl)fumagillol (TNP-470; AGM-1470). *Cancer Res.*, **1993**, *53*(18), 4262-4267.

[62] Griffith, E. C.; Su, Z.; Niwayama, S.; Ramsay, C. A.; Chang, Y.-H. Molecular recognition of angiogenesis inhibitors fumagillin and ovalicin by methionine aminopeptidase 2. *Proc. Natl. Acad. Sci. U.S.A.*, **1998**, *95*(26), 15183-15188.

[63] Kim, D.; Min, J.; Ahn, S. K.; Lee, H. W.; Choi, N. S.; Moon, S. K. 5-Demethoxyfumagillol, a Potent Angiogenesis Inhibitor Isolated from Aspergillus fumigatus. *Chem. Pharm. Bull.*, **2004**, *52*(4), 447-450.

[64] Liu, S.; Widom, J.; Kemp, C. W.; Crews, C. M.; Clardy, J. Structure of Human Methionine Aminopeptidase-2 Complexed with Fumagillin. *Science*, **1998**, *282*(5392), 1324-1327.

[65] Han, C. K.; Ahn, S. K.; Choi, N. S.; Hong, R. K.; Moon, S. K.; Chun, H. S.; Lee, S. J.; Kim, J. W.; Hong, C. I.; Kim, D.; Yoon, J. H.; No, K. T. Design and synthesis of highly potent fumagillin analogues from homology modeling for a human MetAP-2. *Bioorg. Med. Chem. Lett.*, **2000**, *10*(1), 39-43.

[66] Baldwin, J. E.; Bulger, P. G.; Marquez, R. Fast and efficient synthesis of novel fumagillin analogues. *Tetrahedron*, **2002**, *58*(27), 5441-5452.

[67] Sammes, P. G.; Street, L. J.; Kirby, P. Preparation and properties of some phytotoxic 2-benzyloxy-8-oxabicyclo[3.2.1]octane derivatives. *J. Chem. Soc. Perkin Trans. 1*, **1983**, *11*, 2729-2774.

[68] Martin, S. F.; Zinke, P. W. The furan approach to higher monosaccharides. A concise total synthesis of (+)-KDO. *J. Am. Chem. Soc.*, **1989**, *111*(6), 2311-2313.

[69] Pyun, H.-J.; Fardis, M.; Tario, J.; Yang, C. Y.; Ruckman, J.; Henninger, D.; Jin, H.; Kim, C. U. Investigation of novel fumagillin analogues as angiogenesis inhibitors. *Bioorg. Med. Chem. Lett.*, **2004**, *14*(1), 91-94.

[70] Rodeschini, V.; Boiteau, J.-G.; Van de Weghe, P.; Tarnus, C.; Eustache, J. MetAP-2 Inhibitors Based on the Fumagillin Structure. Side-Chain Modification and Ring-Substituted Analogues. *J. Org. Chem.*, **2003**, *69*(2), 357-373.

[71] Mazitschek, R.; Huwe, A.; Giannis, A. Synthesis and biological evaluation of novel fumagillin and ovalicin analogues. *Org. Biomol. Chem.*, **2005**, *3*(11), 2150-2154.

[72] Placidi, L.; Cretton-Scott, E.; de Sousa, G.; Rahmani, R.; Placidi, M.; Sommadossi, J.-P. Disposition and Metabolism of the Angiogenic Moderator O-(Chloroacetyl-carbamoyl) Fumagillol (TNP-470; AGM-1470) in Human Hepatocytes and Tissue Microsomes. *Cancer Res.*, **1995**, *55*(14), 3036-3042.

[73] Tran, H. T.; Blumenschein, G. R.; Lu, C.; Meyers, C. A.; Papadimitrakopoulou, V.; Fossella, F. V.; Zinner, R.; Madden, T.; Smythe, L. G.; Puduvalli, V. K.; Munden, R.; Truong, M.; Herbst, R. S. Clinical and pharmacokinetic

study of TNP-470, an angiogenesis inhibitor, in combination with paclitaxel and carboplatin in patients with solid tumors. *Cancer Chemother. Pharmacol.*, **2004**, *54*(4), 308-314.

[74] Bhargava, P.; Marshall, J. L.; Rizvi, N.; Dahut, W.; Yoe, J.; Figuera, M.; Phipps, K.; Voon S. Ong, V. S.; Kato, A.; Hawkins, M. J. A Phase I and Pharmacokinetic Study of TNP-470 Administered Weekly to Patients with Advanced Cancer. *Clin. Cancer Res.*, **1999**, *5*, 3036-3042.

[75] Jiao, W.; Blunt, J. W.; Cole, A. L. J.; Munro, M. H. G. Fumagiringillin, a New Fumagillin Derivative from a Strain of the Fungus Aspergillus fumigatusâ€ *J. Nat. Prod.*, **2004**, *67*(8), 1434-1437.

[76] Zhou, G.; Tsai, C. W.; Liu, J. O. Fumagalone, a Reversible Inhibitor of Type 2 Methionine Aminopeptidase and Angiogenesis. *J. Med. Chem.*, **2003**, *46*(16), 3452-3454.

[77] Son, K.-H.; Kwon, J.-Y.; Jeong, H.-W.; Kim, H.-K.; Kim, C.-J.; Chang, Y.-H.; Choi, J.-D.; Kwon, B.-M. 5-Demethylovalicin, as a Methionine Aminopeptidase-2 Inhibitor Produced by Chrysosporium. *Bioorg. Med. Chem. Lett.*, **2002**, *10*(1), 185-188.

[78] Hayashi, M.; Kim, Y. P.; Takamatsu, S.; Preeprame, S.; Komiya, T.; Masuma, R.; Tanaka, H.; Komiyama, K.; Omura, S. Chlovalicin, a New Cytocidal Antibiotic Produced by Sporothrix sp. FO-4649. *J. Antibiot.*, **1996**, *49*(7), 631-634.

[79] Asami, Y.; Kakeya, H.; Onose, R.; Chang, Y.-H.; Toi, M.; Osada, H. RK-805, an endothelial-cell-growth inhibitor produced by Neosartorya sp., and a docking model with methionine aminopeptidase-2. *Tetrahedron*, **2004**, *60*(33), 7085-7091.

[80] Hatanaka, H.; Kino, T.; Hashimoto, M.; Tsurumi, Y.; Kuroda, A.; Tanaka, H.; Goto, T.; Okuhara, M. FR65814, A Novel Immunosuppressant Isolated Fron a Penicillium Strain. *J. Antibiot.*, **1988**, *41*(8), 999-1008.

Received: April 15, 2010 Revised: August 09, 2010 Accepted: November 02, 2010