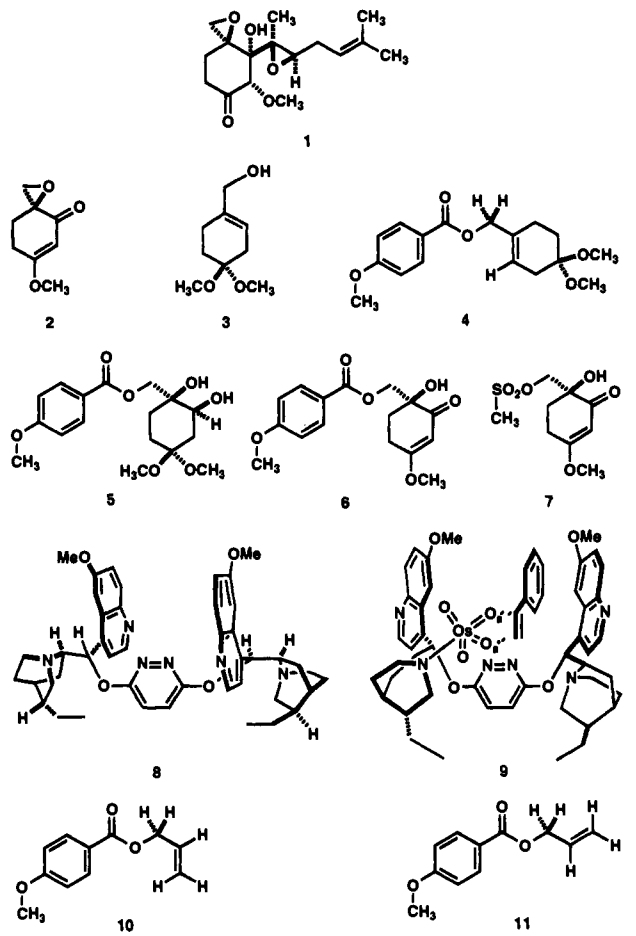
Short Enantioselective Synthesis of (−)-Ovalicin, a Potent Inhibitor of Angiogenesis, Using Substrate-Enhanced Catalytic Asymmetric Dihydroxylation

E. J. Corey,* Angel Guzman-Perez, and Mark C. Noe
Department of Chemistry, Harvard University
Cambridge, Massachusetts 02138

Received September 12, 1994

Inhibition of angiogenesis, the process of development of new blood vessels, is a potentially valuable medical strategy,[1,2] for example, to prevent the growth of solid tumors by cutting off their blood supply.[3] The recent discovery that fumagillin and various ester analogs inhibit angiogenesis and tumor growth at concentrations below 1 ng/mL[4] prompted us to submit for bioassay the related natural product ovalicin (**1**), the synthesis of which (as the racemate) we had reported earlier.[5] Ovalicin was tested by Drs. Judah Folkman and Harold Brem of the Harvard Medical School and was found to be nontoxic, noninflammatory, more potent than fumagillin, and almost as potent as the most active analog AGM-1470.[4a,6] In addition, although fumagillin and AGM-1470 are unstable to storage unless kept in ethanol solution at −20 °C,[6] crystalline ovalicin can be stored at 23 °C for at least 2 years without appreciable decomposition.[7] As a result of these very positive findings we have undertaken the modification of our original synthetic route to (±)-**1** to permit the large-scale synthesis of the natural enantiomer **1** by a catalytic asymmetric process which obviates resolution. This paper reports an effective and simple enantioselective route to the initial chiral intermediate **2** (in >99% ee) from which (−)-ovalicin has been obtained by application of the sequence reported earlier.[5,8,9] The key step in the synthesis of **2** is a new version of the cinchona–OsO_4-catalyzed dihydroxylation reaction[10] in which a *p*-methoxybenzoyl group is attached to the dihydroxylation substrate to produce a dramatic enhancement in enantioselectivity. The broader value of this substrate design approach is supported by the inclusion of several other examples and a mechanistic rationale which provides structural guidance.

The starting point of the present synthesis was the readily available, known allylic alcohol **3**,[11] which was converted to the *p*-methoxybenzoate ester **4** in 98% yield by reaction with

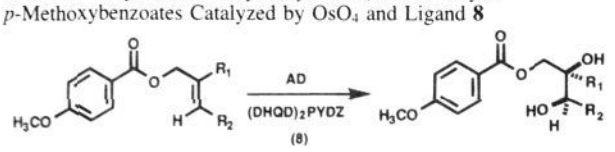


1.1 equiv of *p*-methoxybenzoyl chloride, 1.5 equiv of triethylamine, and 0.05 equiv of 4-(dimethylamino)pyridine at 23 °C for 3 h. The *p*-methoxybenzoate **4** was chosen as a substrate for the critical chirality-producing step, OsO_4–biscinchona alkaloid catalyzed dihydroxylation, on the basis of the mechanistic model recently proposed to explain cases of high enantioselection.[12] The mechanistic reasoning behind this choice is summarized below. Reaction of **4** with 1 mol % of K_2OsO_4, 1 mol % of the $(DHQ)_2PHAL$ biscinchona ligand (Aldrich Co.), 3 equiv of $K_3Fe(CN)_6$, 3 equiv of K_2CO_3, and 1 equiv of $CH_3SO_2NH_2$ in a stirred two-phase mixture of *t*-BuOH–H_2O at 0 °C for 4 h produced the dihydroxylation product **5** in 93% yield and >99% enantiomeric excess (ee).[13] In sharp contrast, the corresponding reaction with the allylic alcohol **3** afforded the dihydroxylation product in only 18% ee and was, therefore, a totally unsuitable substrate. The asymmetric dihydroxylation of the pivalate and triisopropylsilyl derivatives of **3** proceeded with 35% and 13% ee, respectively. Oxidation of the secondary alcohol function in **5** (Swern method, oxalyl chloride, dimethyl sulfoxide, −78 °C for 30 min followed

(1) Folkman, J.; Klagsbrun, M. *Science* **1987**, *235*, 442.
(2) For a recent review, see: Mitchell, M. A.; Wilks, J. W. *Annu. Rep. Med. Chem.* **1992**, *27*, 139.
(3) Neovascularization is crucial not only for solid tumor growth but also in other diseases, for example, diabetic retinopathy, arthritis, psoriasis, and Kaposi's sarcoma. See ref 2 and also the following: Nakamura, S.; Sakurada, S.; Salahuddin, S. Z.; Osada, Y.; Tanaka, N. G.; Sakamoto, N.; Sekiguchi, M.; Gallo, R. C. *Science* **1992**, *255*, 1437.
(4) (a) Ingber, D.; Fujita, T.; Kishimoto, S.; Sudo, K.; Kanamuru, T.; Brem, H.; Folkman, J. *Nature* **1990**, *348*, 555. (b) Kusuka, M.; Sudo, K.; Fujita, T.; Marui, S.; Itoh, F.; Ingber, D.; Folkman, J. *Biochem. Biophys. Res. Commun.* **1991**, *174*, 1070. (c) Marui, S.; Kishimoto, S. *Chem. Pharm. Bull. Jpn.* **1992**, *40*, 575. (d) For inhibition of angiogenesis by genistein, see: Fotsis, T.; Pepper, M.; Adlercreutz, H.; Fleischmann, G.; Hase, T.; Montesano, R.; Schweigerer, L. *Proc. Natl. Acad. Sci. U.S.A.* **1993**, *90*, 2690.
(5) Corey, E. J.; Dittami, J. P. *J. Am. Chem. Soc.* **1985**, *107*, 256.
(6) Written report to E.J.C. from Dr. J. Folkman dated March 3, 1992.
(7) For literature on the isolation and structure of natural ovalicin, see: (a) Sigg, H. P.; Weber, H. P. *Helv. Chim. Acta* **1968**, *51*, 1395. (b) Sassa, T.; Kaise, H.; Munakata, K. *Agric. Biol. Chem.* **1970**, *34*, 649. (c) Bollinger, P.; Sigg, H. P.; Weber, H. P. *Helv. Chim. Acta* **1973**, *56*, 819.
(8) Studies to develop an even shorter route to **1** from **2** are underway.
(9) Subsequent to the development of the route to chiral **2** and **1** described herein, a conversion of L-quebrachitol to (−)-ovalicin in ca. 20 steps was reported; see: Bath, S.; Billington, D. C.; Gero, S. D.; Quiclet-Sire, B.; Samadi, M. *J. Chem. Soc., Chem. Commun.* **1994**, 1495.
(10) (a) Jacobsen, E. N.; Markó, I.; Mungall, W. S.; Schröder, G.; Sharpless, K. B. *J. Am. Chem. Soc.* **1988**, *110*, 1968. (b) Sharpless, K. B.; Amberg, W.; Bennani, Y. L.; Crispino, G. A.; Hartung, J.; Jeong, K.-S.; Kwong, H.-L.; Morikawa, K.; Wang, Z.-M.; Xu, D.; Zhang, X.-L. *J. Org. Chem.* **1992**, *57*, 2768. (c) Vanhessche, K. P. M.; Wang, Z.-M.; Sharpless, K. B. *Tetrahedron Lett.* **1994**, *35*, 3469 and references cited therein.
(11) Prepared from *p*-methoxybenzyl alcohol by Birch reduction (Na–NH_3–THF–EtOH at −33 °C for 30 min; 70%) followed by reaction with MeOH–$HC(OMe)_3$ and a catalytic amount of *p*-toluenesulfonic acid at 23 °C for 2 h (96%). See: (a) Iio, H.; Isobe, M.; Kawai, T.; Goto, T. *Tetrahedron* **1979**, *35*, 941. (b) Berkowitz, W. F.; Amarasekara, A. S.; Perumattam, J. J. *J. Org. Chem.* **1987**, *52*, 1119.
(12) (a) Corey, E. J.; Noe, M. C. *J. Am. Chem. Soc.* **1993**, *115*, 12579. (b) Corey, E. J.; Noe, M. C.; Sarshar, S. *Tetrahedron Lett.* **1994**, *35* 2861. (c) Corey, E. J.; Noe, M. C.; Sarshar, S. *J. Am. Chem. Soc.* **1993**, *115*, 3828. (d) Corey, E. J.; Noe, M. C.; Grogan, M. J. *Tetrahedron Lett.* **1994**, *35*, 6427.
(13) Ee value determined by HPLC analysis using a Chiralcel OD column. The enantioselectivity of the asymmetric dihydroxylation reaction of the *p*-nitro- and *p*-bromo-subsituted and unsubstituted benzoate esters of **3** was also >99%. However, these substrates and esters such as acetates were synthetically less useful than **4** because of the occurrence of significant amounts of acyl migration from primary to tertiary and secondary OH groups.

0002-7863/94/1516-12109$04.50/0

Table 1. Asymmetric Dihydroxylation (AD) of Allylic *p*-Methoxybenzoates Catalyzed by OsO_4 and Ligand **8**



allylic ester	yield, %	ee,[a] %	allylic ester	yield, %	ee,[a] %
allyl	>99	98	(*E*)-crotyl	96	>99
2-methylallyl	98	97	1-cyclohexenylmethyl	>99	98

[a] Enantioselectivity determined by HPLC analysis using a Chiralpak AS column for the first three entries and a Chiralcel OD column for the fourth entry with isopropyl alcohol–hexane for elution at 23 °C.

by triethylamine at −78 to 23 °C) afforded the corresponding ketone (87%), which upon treatment with a catalytic amount of *p*-toluenesulfonic acid in CH_2Cl_2 at 23 °C for 1 h gave the β-methoxy α,β-enone **6** (93%). The required epoxy ketone **2** was easily obtained from **6** by the following sequence: (1) ester cleavage (K_2CO_3–MeOH at 23 °C for 3 h; 93% yield), (2) mesylate formation (CH_3SO_2Cl–Et_3N in CH_2Cl_2 at −78 to 23 °C for 1 h to give **7**), and (3) reaction of mesylate **7** with aqueous NaOH at 23 °C for 5 min to give **2** (82% for two steps). The key intermediate **2** was converted into synthetic ovalicin as previously described.[5,8] Totally synthetic ovalicin produced in this way was identical with an authentic sample of natural ovalicin by comparison of the ^1H and ^{13}C NMR, IR, and mass spectra, mp and mixture mp, optical rotation, and chromatographic mobility.

The very highly enantioselective dihydroxylation of the *p*-methoxybenzoate **4** to form the diol **5** mandated a study of the scope of this process with a series of allylic *p*-methoxybenzoate substrates using 1 mol % of K_2OsO_4 and 1 mol % of the $(DHQD)_2PYDZ$ reagent (**8**) as the catalytic chiral ligand[12c] for a minimum reaction time to prevent acyl migration. The results for the *p*-methoxybenzoates of allyl alcohol, 2-methylallyl alcohol, (*E*)-crotyl alcohol, and 1-cyclohexenylmethanol were uniformly excellent with regard to both yield and enantioselectivity as shown in Table 1. In contrast to the high enantioselectivities indicated in Table 1 (97 to >99%) for *p*-methoxybenzoates, only poor or mediocre enantioselectivities were observed for free allylic alcohols, or with benzyl or triisopropylsilyl ethers or pivalate esters. For example, the ee value observed for allyl triisopropylsilyl ether was 3% and that for the benzyl ether was 61%.[10c,14]

The application of substrate modification of allylic alcohols as an effective strategy for the achievement of high enantioselectivity in the asymmetric dihydroxylation of allylic alcohols, as described herein, was guided by the mechanistic model which has recently been advanced for the biscinchona alkaloid–OsO_4 system.[12a,b,d] The proposed transition state assembly for the dihydroxylation of styrene using the $(DHQD)_2PYDZ$–OsO_4 system is represented in **9**. The essential features of this dihydroxylation assembly are as follows: (1) a preference for the U-shaped conformation **9** for the OsO_4 complex of **8**; (2) the ability of **9** to hold olefinic substrates such as styrene in a binding pocket composed of the two parallel methoxyquinoline units and the pyridazine connector, as shown; (3) staggered geometry about the Os–N bond of the **8**–OsO_4 complex;[12c] (4) the proximity of one axial oxygen and one equatorial oxygen of the complexed OsO_4 unit to the olefinic carbons of the bound substrate, as shown in **9**; and (5) a minimum motion pathway from this arrangement for the [3 + 2] cycloaddition which directly produces the pentacoordinate osmate ester in the energetically most favorable geometry.[12a] The rate acceleration

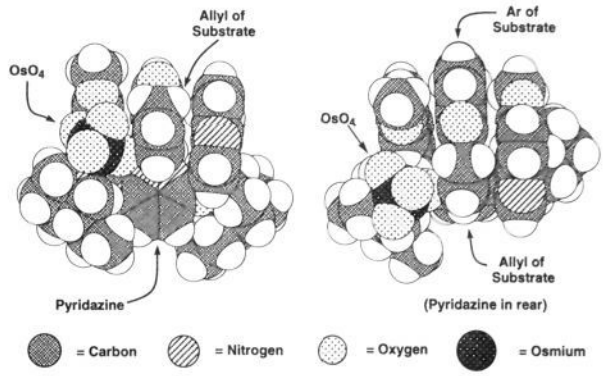


Figure 1. Two views of the proposed geometry for the complex between the allyl *s-cis* form of allyl *p*-methoxybenzoate, OsO_4, and ligand **8**. The catalyst geometry was based on the X-ray crystal structure of **8**–CH_3I[12b] with the following modifications: (a) The methyl group of the methiodide salt was replaced by OsO_4 with the staggered arrangement about the N–Os bond and the bond distances demonstrated from X-ray studies.[12c] (b) The H(8)–C(8)–C(9)–H(9) dihedral angle was adjusted to ca. 90°.[12b]

for the observed enantioselective pathway relative to other modes is due to the favorable free energy of activation for reaction from the complex **9** in which the reactants are held in a manner which is ideal for formation of the thermodynamically more stable osmate ester. Dihydroxylation of the olefin face opposite to that shown in **9** is unfavorable due to the fact that there is no three-dimensional arrangement for simultaneous π-facial approach of the olefin to the oxygens labeled as O_a and O_e and favorable interaction with the binding pocket. X-ray crystallographic data suggest that the pyridazine ring at the bottom of the U-shaped cavity tends to be oriented so as to allow conjugation of the ring and the two alkoxy substituents, with the N–N side of the ring participating in binding to the substrate,[12b] though the exact tilt of this ring during reaction probably varies slightly with substrate.

When allyl *p*-methoxybenzoate is used as reactant, excellent binding of this substrate can be expected because of extensive π-contact of both faces of the allyl and anisoate moieties with the two methoxyquinoline units and edge contact with the pyridazine ring as shown by the two views in Figure 1. An important factor for this binding is the conformational preference and restriction of the anisoate ester substrate. The conformation used for the allyl anisoate in this complex is the *s-cis* allylic conformer **10**, which has been found by MM2 (Macromodel version 3.5a) force field calculations to be more stable than the *s-trans* allylic conformer **11** by 2.1 kcal/mol and within 20 cal/mol of the global minimum in energy. The allylic *s-cis* conformation shown in **4** also fits very well into the binding cavity of the **8**–OsO_4 complex and can react via a transition state assembly analogous to that shown in Figure 1 for **10**.

In summary, this paper has presented the following significant new findings: (1) (−)-ovalicin (**1**) shows outstanding promise as an inhibitor of angiogenesis and tumor growth, (2) a short and practical enantioselective synthesis of **1** is possible using catalytic dihydroxylation of a designed substrate to achieve fully asymmetric synthesis, and (3) an effective strategy has been developed for maximizing enantioselectivity in the dihydroxylation of allylic alcohol substrates.[15]

Supplementary Material Available: Experimental procedures for the ovalicin synthesis and for the preparation and proof of absolute configuration of the products in Table 1 (13 pages). This material is contained in many libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

(14) The asymmetric dihydroxylation of a series of substituted aryl allyl ethers has been reported with ee's ranging from 28 to 95% (average ca. 70%). See: Wang, Z.-M.; Zhang, X.-L.; Sharpless, K. B. *Tetrahedron Lett.* **1993**, *34*, 2267. More recently the dihydroxylation of allyl *p*-methoxyphenyl ether was reported by Bittman et al. in 90–92% ee; see: Vilchèze, C.; Bittman, R. *J. Lipid Res.* **1994**, *35*, 734.

(15) This research was assisted financially by graduate fellowships to M.C.N. (NSF) and A.G.-P. (Schering-Plough) and a grant from the National Institutes of Health. We are indebted to Drs. Judah Folkman and Harold Brem of the Harvard Medical School for bioassays of ovalicin.