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Total Synthesis of Rhizoxin D

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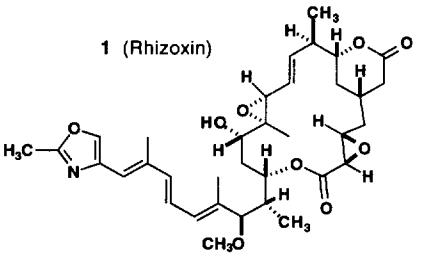
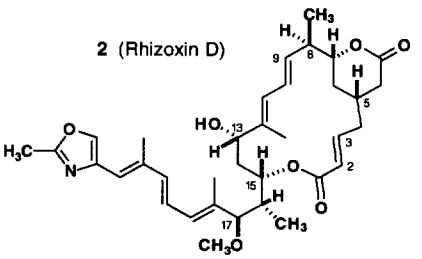
Abstract: The convergent, enantiocontrolled synthesis of a significant antimitotic agent for cancer chemotherapy is presented with completion of rhizoxin diene **2**.

The rhizoxins are a family of sixteen-membered macrolactones with exceptionally potent antitumor and antifungal activity. Produced by *Rhizopus chinensis*, rhizoxin (**1**) was first discovered as the causal agent of rice seedling blight.[1] Subsequent explorations[2] have led to characterizations of several related structures, including the macrocyclic diene **2** (rhizoxin D),[3] which is a putative precursor to the *bis*-epoxide **1**. Studies revealing the absolute configuration of **1** and a biosynthetic pathway have been reported.[4]



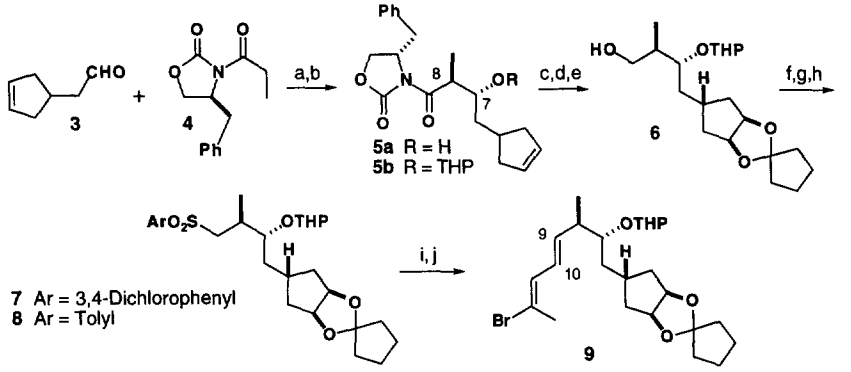


The rhizoxins exhibit powerful antimitotic effects as reversible inhibitors of the polymerization of intracellular α- and β-tubulin to prevent microtubule formation.[5] One striking feature is that the rhizoxins exhibit no cross-resistance to agents such as cisplatin, doxorubicin, and etoposide.[6] Rhizoxin and maytansine share a related binding site, which is non-identical to the receptor for other inhibitors, including colchicine, vinblastine, dolastatins, and halichondrin.[7] Limited structure-activity information suggests that **2** exhibits equivalent potency to rhizoxin itself.[5,6a] *In vivo* activity has been demonstrated for solid tumors, including five human tumor xenografts: LOX melanoma, MX-1 breast cancer, non-small cell lung cancer A549, and small cell lung cancers LXFS605 and LXFS650.[8] Phase II clinical studies of rhizoxin have been completed, and Phase III evaluations are underway. In 1993, Ohno and coworkers[9] communicated the first synthesis of rhizoxin. Recently, Kende has reported a route for the preparation of **2**,[10] and studies have described various structural fragments.[11] Herein we present our efforts culminating in the enantiocontrolled total synthesis of rhizoxin D (**2**).

From the onset, our plans were directed with two underlying assumptions. Our experiences with macrolactonizations suggested that ring closure of a hindered C-15 alcohol with an activated acyl derivative of the stabilized α,β-unsaturated acid (C_1) would be difficult compared to the intramolecular Horner-Emmons process envisioned from a C-2/C-3 disconnection. Secondly, this strategy introduced an element of pseudosymmetry with a C-3 aldehyde and the carbonyl of the valerolactone arising from a two-carbon branch at C-5 of **2**. These concepts were realized beginning with 3-cyclopentenylacetaldehyde[12] for preparation of the C_3-C_{12} fragment

(Scheme I). Using an Evans aldol reaction,[13] the boron Z(O) enolate derived from the (*S*)-4-benzyloxazolidinone **4** provided exclusive formation of the *syn*-adduct securing the asymmetry at C_7 and C_8 of **5**.

Scheme I[a]

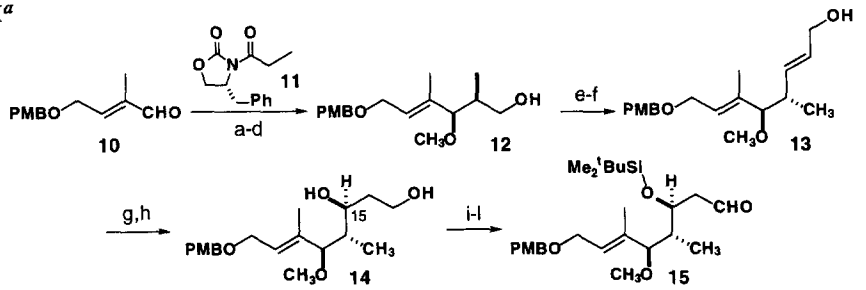


[a]Key: a) *n*-Bu_2BOTf, Et_3N, CH_2Cl_2, 0 °C to –78 °C then **3**, 95%; b) DHP, PPTs, CH_2Cl_2, 100%; c) OsO_4, NMO, Acetone/H_2O, 88%; d) cyclopentanone, PPTs, $HC(OEt)_3$, CH_2Cl_2, 92%; e) $LiBH_4$, MeOH, Et_2O, 0 °C, 91%; f) TsCl, Et_3N, CH_2Cl_2, 98%; g) ArSH, NaH, DMF/THF, 0 °C, 95%; h) *m*CPBA, $NaHCO_3$, CH_2Cl_2, 98%; i)LDA, HMPA, THF, –78 °C then (*E*)-1,3-dibromo-2-butene, 88%; j) DBU, tol., 105 °C, 70%.

Dihydroxylation of **5b** afforded a mixture (7:1 ratio) of cis-diols.[14] Diastereofacial selectivity of the oxidation was only evident from ^{13}C-NMR spectra of crude triols obtained via removal of the C_7 THP ether. In all other aspects, the compounds of the synthesis route behaved as simple (1:1 ratios) THP isomers. Ketalization of the diols from **5** followed by hydride reductive cleavage[15] of the chiral auxiliary yielded alcohol **6**. Standard transformations gave **7**, and deprotonation to the corresponding α-sulfonyl carbanion led cleanly to C-alkylation.[16] Subsequent E_2 elimination of the intermediate allylic sulfone produced *trans*-diene **9** in 70% isolated yield. In contrast, use of the *para*-tolylsulfone **8** led to only 30% yield of **9** with evidence of decomposition at higher temperatures. Base-induced sulfone elimination with KO^{t}Bu (1 M in THF) at 22 °C afforded the analogous (*E*)-enyne (80% yield) via the further loss of HBr from **9**.

Synthesis of the $C_{13} \rightarrow C_{20}$ fragment was accomplished on a multigram scale (Scheme II) by initially establishing the C_{16} and C_{17} asymmetry with high enantiocontrol via the Evans aldol condensation[13,15] with α,β-unsaturated aldehyde **10**. The remaining stereocenter at C_{15} was installed by the Sharpless asymmetric

Scheme II[a]

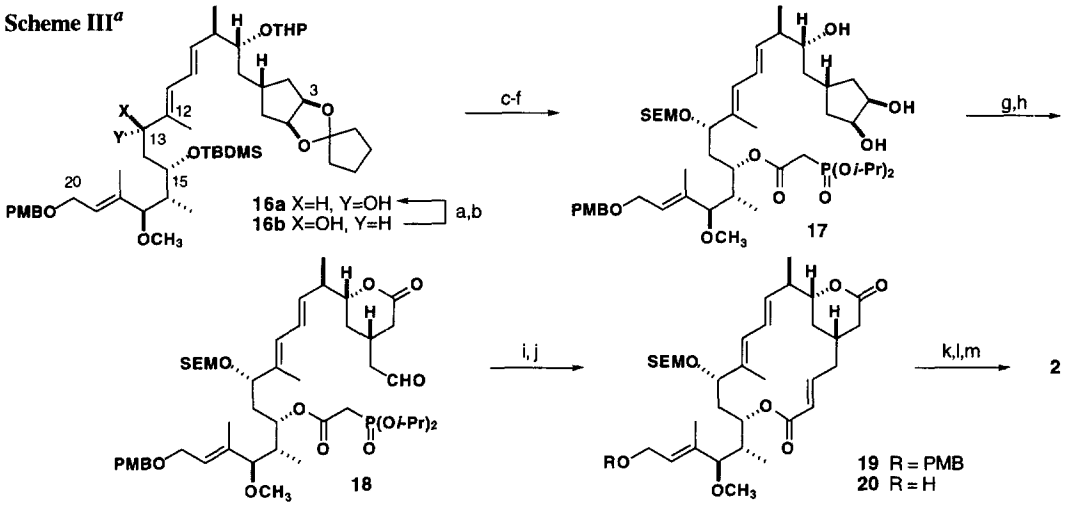


[a]Key: a) *n*-Bu_2BOTf, Et_3N, CH_2Cl_2, 0 °C to –78 °C then **10**, 87%; b) Bu_3B, $LiBH_4$, HOAc, THF, 77%; c) TBDMSCl, Imid., CH_2Cl_2, (100%); d) *i.* NaH, CH_3I, THF, *ii.* PPTs, EtOH, 66 °C, 97% (two steps); e) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, –78 °C to 22 °C, then $(C_6H_5)_3PCHCO_2CH_3$, 88%; f) DIBAL, CH_2Cl_2, 93%; g) $Ti(O^iPr)_4$, (–)-DET, ^{t}BuOOH, CaH_2, 4Å sieves, CH_2Cl_2, 93%; h) Red-Al, THF, –78 °C to 22 °C, 90%; i) Piv-Cl, pyr, CH_2Cl_2, 90%; j) $Me_2{}^t$BuSiOTf, collidine, CH_2Cl_2, 98%; k) DIBAL, CH_2Cl_2, –78 °C, 87%; l) $(COCl)_2$, DMSO, CH_2Cl_2 then Et_3N, –78 °C to 22 °C, 92%.

epoxidation (88% de) of *E*-allylic alcohol **13** provided from the Wittig chain extension of primary alcohol **12**. Regioselective hydride opening[17] of the epoxyalcohol from **13** gave diol **14** for transformation into β-silyloxy-aldehyde **15**. The choice of the silyl protection at C_{15} was important for steric shielding to prevent base-induced α-deprotonation in **15**. This was apparent in the BOM ether and the analogous C_{15} benzoate of **15**, which suffered from β-eliminations in the Swern oxidation or in further attempts for nucleophilic additions.

Linkage of our nonracemic fragments was achieved through halogen-metal exchange of **9** (^{t}BuLi, 2 equiv., THF/Et_2O/pentane (4:1:1) at −120 °C; warming to −90 °C) followed by addition of aldehyde **15** (−90 °C to −78 °C). The process afforded a mixture (1:1 ratio) of C_{13} alcohol diastereomers **16** in 77% yield (Scheme III).[18] Undesired isomer **16b** was readily separated by flash chromatography and effectively converted into **16a** via TPAP oxidation[19] and borohydride reduction using (*R*)-2-methyl-CBS-oxazaborolidine,[20] giving a 68% overall yield of the desired C_{13} alcohol. Interestingly, the conjugated enone system displayed an inherent facial bias upon reaction with simple hydrides ($LiAlH_4$) for highly selective production of **16b**. After C_{13} hydroxyl protection and fluoride treatment, esterification with diisopropylphosphonoacetic acid was followed by

Scheme III[a]



[a]Key: a) TPAP, NMO, 4Å sieves, CH_2Cl_2, 80%; b) (*R*)-2-methyl-CBS-oxazaborolidine, BH_3, THF, −10 °C, 85%; c) SEMCl, $^iPr_2EtN$, CH_2Cl_2, 94%; d) TBAF, THF, 94%; e) diisopropylphosphonoacetic acid, 1-cyclohexyl-3-(2-morpholinoethyl)-carbodiimide methyl-*p*-toluenesulfonate, DMAP, 4Å sieves, CH_2Cl_2, 97%; f) HOAc/H_2O (4/1), 48 h, 56%; g) $NaIO_4$, THF/H_2O; h) TPAP, NMO, 4Å sieves, CH_2Cl_2, 67%, 2 steps; i) DBU (2 equiv), CH_3CN, LiCl (15 equiv), 4.6×10^{-4} M, 84%; j) DDQ, H_2O, CH_2Cl_2, 98%; k) MnO_2, 70%; l) KHMDS, 4-[3-(diphenylphosphinoyl)-2-methylpropenyl]-2-methyloxazole, 44%; m) Me_2BBr, THF/CH_2Cl_2, 0 °C, 65%

simultaneous hydrolysis of the THP ethers and cyclopentylidene ketal to afford the key intermediate triol **17**.[21] Oxidative cleavage of the vicinal diol provided two diastereomeric lactols (ratio 3:2). Development of stereochemistry at C_5 in the lactols was based on a clear thermodynamic preference for diequatorial (C_5/C_7) substitution. These lactols underwent a mild TPAP oxidation to a single lactone **18** in 67% overall yield. Finally, intramolecular Horner-Emmons cyclization[22] of **18** was achieved in 84% yield necessitating strictly anhydrous conditions to avoid hydrolysis of the δ-lactone in **19**. Quantitative DDQ removal[23] of the *para*-methoxybenzyl ether produced the parent rhizoxin macrocycle **20**.

Completion of the total synthesis of **2** was realized by oxidation of the allylic alcohol **20**, and Wittig olefination (–78 °C, THF) with the carbanion generated from 4-[3-(diphenylphosphinoyl)-2-methyl-propenyl]-2-methyloxazole.[9] The all-*trans*-triene was obtained in 44% yield,[24] and deprotection of the C_{13} SEM ether afforded synthetic rhizoxin D, $[\alpha]_D^{24}$ + 293° (c 0.53, MeOH), with characterization data which were identical to the literature for the natural product.[2a]

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