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Epothilones are a class of promising antibreast cancer candidate. They were synthesized by three strategies. Structure – activity relationships were studied with the synthetic compounds. The first in vivo test of synthetic epothilone B is encouraging. Further chemical and biological studies are on going.

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FOREWORD

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The epothilones, isolated from the cellulose-degrading myxobacterium *Sorangium cellulosum,* are the first class of microtubule(MT)-stabilizing agents with a taxol-like mechanism of action since the original discovery of taxol. [1,2,3] *In vitro* and in cultured cells, the epothilones mimic all of the biological effects of taxol, one of the most effective drugs for the treatment of breast cancer. Competition binding studies reveal that they share the same MT-binding site and bind with an affinity comparable to that of taxol. It is significant that the epothilones appear to possess several advantages over taxol. First, they exhibit a much lower drop in potency compared to taxol against a multiple drug-resistant cell line. Second, they have much better solubility in water. Third, relative to taxol, the epothilones would appear to be more manageable as targets for total synthesis. It is conceivable that the epothilones could emerge as promising drug candidates for the treatment of breast cancer . With respect to clinical utility, these substances may prove superior to taxol. At the present time, it is difficult to obtain significant quantities of the epothilones through fermentation. It remains to be determined whether or not the epothilones and analogues can be obtained in useful amounts by total synthesis.

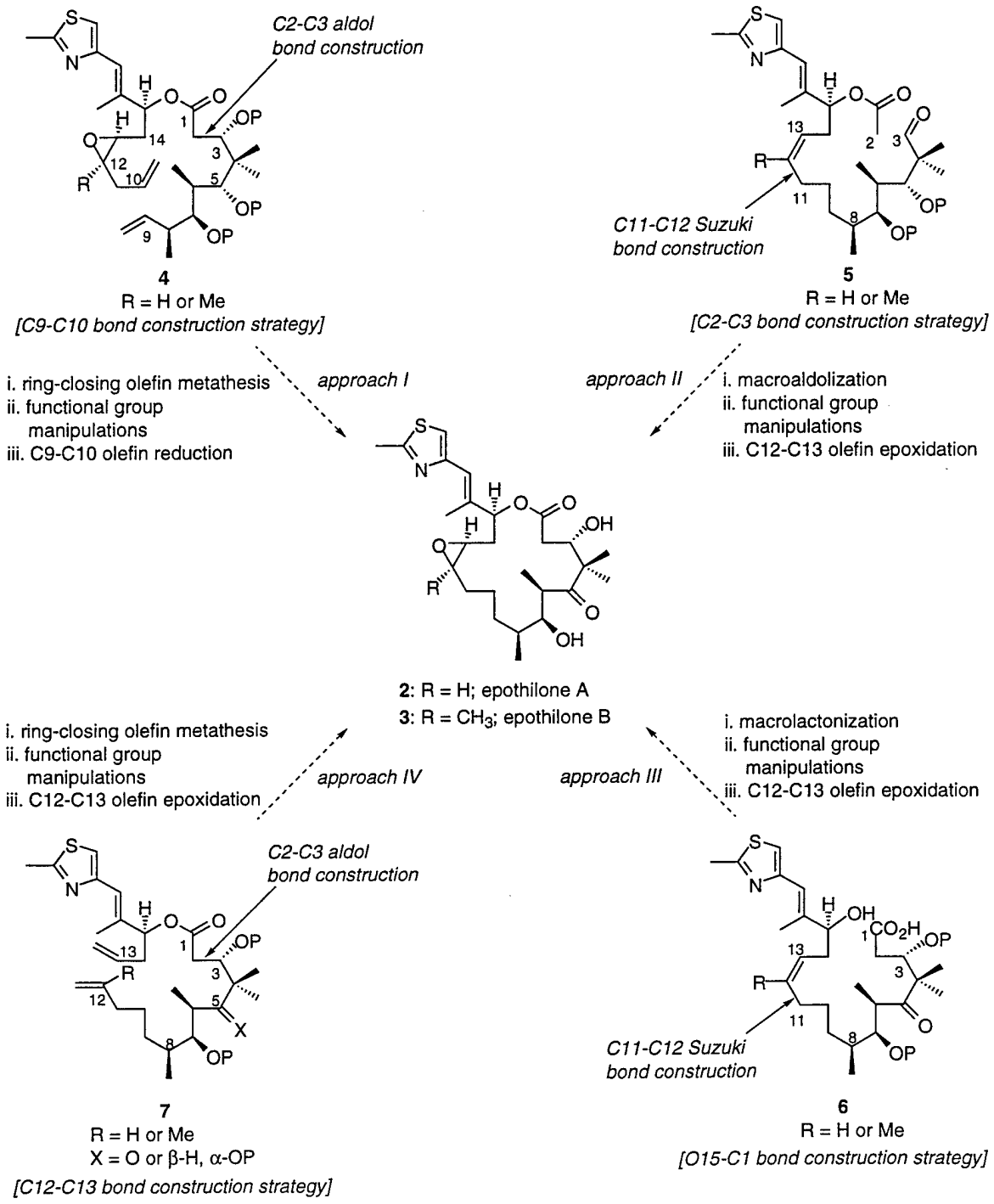
In the proposal, a convergent and stereocontrolled path for a total synthesis of epothilones A and B was described. The long term goals include:

1) Total syntheses of epothilones under academic level;
2) Under the guidance of Molecular Modeling studies, design and synthesize simplified epothilone analogues and evaluate their potential as chemotherapeutic agents for the treatment of breast cancer;
3) The development of a practical synthesis of epothilones which would impact on the actual availability of the drug for serious pre-clinical and clinical evaluation. *

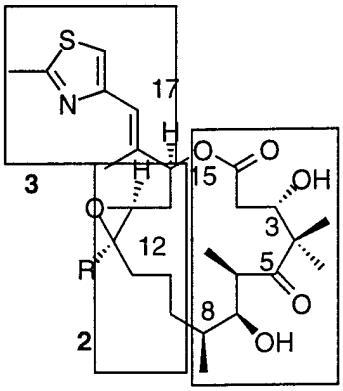
What has been achieved in the past year in this lab includes:

1) Chemically , four synthetic strategies have been attemped(see scheme 1), three of which were successful.[4, 5, 6, 7, 8, 10.] Some new methods and new reaction sequences have been used : a) an unprecedented stereocontrolled macroaldolization was used to close the 16 member ring; b) ring-closing olefin metathesis(RCM), as an arising powerful C-C bond formation, was systematically studied on the most complicated substrates. The theoretically safest strategy to close the ring at C9 – C10 using RCM failed although model compounds worked well in the presence of epoxide and thiozal double bond. The remote functional groups and catalysts both affected the E/Z ratio of the newly formed double bond from metathesis. The trisubstituted olefin required a more reactive molybdenum catalyst without any free hydroxy group on the substrate.
2) Biologically, the cytotoxicity and tubulin polymerization of epothilones and important synthetic intermediates were studied [4, 5, 6, 8, 10]. Furthermore, a library of epothilone analogues were synthesized for the structure-activity relationship study.[9,11] The first in vivo test was done with synthetic epothelones[11]. The result demonstrate that (scheme 2): a) Epothilones are hundreds to thousands times more effective than Taxol to some drug resistant cell lines and show similar MT-stabilizing efficiency; b) The efficacy is very sensitive to ring size and the modification of the acyl sector (sector 1, C1-C8), sector 2 (C9-C15) and 3 (aryl side chain) are more tolerant although C16-C17 spacer is very important.

*The biologiacal test will be done in some other labs.



Scheme 1. Bond construction strategies for total syntheses of epothilone A (**2**) and epothilone B (**3**).



scheme 2. The three arbitrarily defined sectores of the epothilones

Some further work is ongoing:
1) A practical synthesis of epothilones has been started. This synthesis would provide grams to hundreds grams of the epothilones for serious pre-clinical and clinical evaluation.
2) More epothelones will be tested on animals to screen for drug candidates for clinical evaluation.
In conclusion, the primary goals of the proposal have been reached, the results showed epothilones are promising to cure breast cancer.

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Total Syntheses of Epothilones A and B

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Abstract: Convergent, stereocontrolled total syntheses of the microtubule-stabilizing macrolides epothilones A (**2**) and B (**3**) have been achieved. Four distinct ring-forming strategies were pursued (see Scheme 1). Of these four, three were reduced to practice. In one approach, the action of a base on a substance possessing an acetate ester and a nonenolizable aldehyde brought about a remarkably effective macroaldolization see (**89** → **90** + **91**; **99** → **100** + **101**), simultaneously creating the C2−C3 bond and the hydroxyl-bearing stereocenter at C-3. Alternatively, the 16-membered macrolide of the epothilones could be fashioned through a C12−C13 ring-closing olefin metathesis (e.g. see **111** → **90** + **117**; **122** → **105** + **123**) and through macrolactonization of the appropriate hydroxy acid (e.g. see **88** → **93**). The application of a stereospecific *B*-alkyl Suzuki coupling strategy permitted the establishment of a *cis* C12−C13 olefin, thus setting the stage for an eventual site- and diastereoselective epoxidation reaction (see **96** → **2**; **106** → **3**). The development of a novel cyclopropane solvolysis strategy for incorporating the geminal methyl groups of the epothilones (see **39** → **40** → **41**), and the use of Lewis acid catalyzed diene−aldehyde cyclocondensation (LACDAC) (see **35** + **36** → **37**) and asymmetric allylation (see **10** → **76**) methodology are also noteworthy.

The introduction of taxol (paclitaxel) (**1**) into cancer chemotherapy is testimony to the synergism of broadly based contributions from many areas of scientific expertise *en route* to the clinic. The original isolation and structure work, which also served to identify the cytotoxicity of the drug, was accomplished by Wall and co-workers.[1] In a seminal paper, Horwitz identified the *in vitro* mode of action of taxol, demonstrating its ability to stabilize microtubule assemblies.[2] This finding gave impetus to a wider range of pharmacological investigations of critical importance. The development of improved methods from phytochemical sources for obtaining baccatin III, and improved chemical methods for converting baccatin III to paclitaxel, provided the drug in ample quantities for human trials.

On the basis of favorable findings that issued from these evaluations, paclitaxel, developed by the Bristol Myers-Squibb Co., was approved for chemotherapeutic application against ovarian carcinomas. Since then, this drug has been undergoing extensive evaluations for other indications and is being incorporated in a variety of clinical contexts. Though it is often not a curative agent, paclitaxel is emerging as a useful main line chemotherapeutic resource. There being no evidence to the contrary, it is assumed that the *in vivo* mode of action and antitumor properties of paclitaxel arise from inhibition of cellular mitosis through the Horwitz mechanism. The question of whether or not this mode of action is actually operative in the human patient is not easily addressed.

Although having many advantages, paclitaxel is not an ideal drug.[3] One major problem with this agent, useful as it is, has to do with difficulties in its formulation. Paclitaxel is a rather insoluble substance in water, thereby necessitating awkward forms of clinical administration. Perhaps even more serious is the fact that paclitaxel is subject to a significant attenuation of therapeutic value through the onset of multiple drug resistance (MDR). Although there has been extensive structure−activity work in the paclitaxel area, to our knowledge the only modified compound presently being evaluated in human clinical trials is the close relative, taxotere.[4]

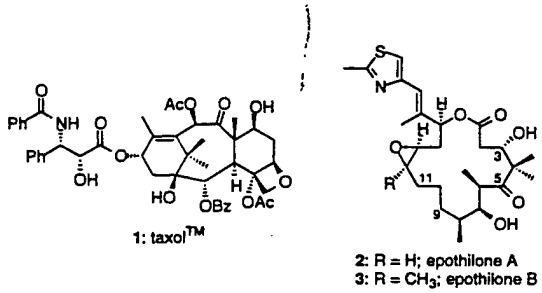

Figure 1. Structures of taxol (**1**) and epothilones A (**2**) and B (**3**).

Given the high interest engendered by taxoids as a consequence of their clinical usefulness, the search for new agents that function by a comparable mechanism is of great interest. It is in this connection that the recently discovered bacterial metabolites epothilone A (**2**) and B (**3**) have attracted considerable attention. These compounds were first identified as antifungal cytotoxic agents by Höfle and co-workers.[5] During the course of a screening program aimed at the identification of substances with a paclitaxel-like mode of action, a group based at Merck found that the epothilones are powerful cytotoxic agents which function through stabilization of cellular microtubules.[6] The strong implication of the Merck research effort was that the epothilones share in the paclitaxel mode of action.

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⊗ Abstract published in *Advance ACS Abstracts,* October 1, 1997.

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Indeed, these compounds seem to adhere to the "taxol binding domains" of the microtubule assemblies. In light of the possibility that the epothilones, or suitably modified derivatives, might find a role in cancer chemotherapy, this series also merits multidisciplinary attention. Among the scientific enterprises which we felt to be warranted in the case of the epothilones would be research directed to their total synthesis.

Unlike the situation with paclitaxel, where it was clear from the outset that total synthesis would be unlikely to impact upon the actual availability of the drug itself,[7] the simpler structures of the epothilones invited the hope that chemical synthesis could improve accessibility to the desired agents. Thus, organic chemistry could contribute to the development of an epothilone-based drug effort through a highly efficient total synthesis or possibly by delivering much simpler structures that still manifest chemotherapeutically useful biological activity.

Moreover, several intrinsically challenging chemical issues require attention in any total synthesis venture aimed at the epothilones. Quickly recognized in the case of epothilone A is the presence of a thiazole moiety, a *cis*-epoxide (C12−C13) and, somewhat unusual for a macrolide, the presence of geminal methyl groups at C4. Not the least noteworthy feature of the synthesis problem is inherent in the array of three contiguous methylene groups which serves to insulate the two domains of the epothilones that bear stereochemical imprints. The acyl section, numbered from carbons 1−8, presents a constellation of four chiral centers whose proper emplacement would require careful management. An agenda dealing with the synthesis of this domain must also include programs for elaborating and maintaining a potentially unstable β-hydroxy ester linkage at C3. The oxidation state at C3 must be cleanly differentiated from that at C5 where a ketonic group is to emerge. This entire polypropionate sector is insulated by carbons 9, 10, and 11 from the chiral *O*-alkyl domain comprising carbons 12−15. The already mentioned *cis*-epoxide, connecting carbons 12 and 13 (disubstituted in the case of epothilone A and trisubstituted in the case of epothilone B), is insulated by a single methylene group from carbon 15, which bears an allylic alcohol and a thiazole-based version of an α-methyl styryl linkage.

None of these issues in isolation pose an insurmountable obstacle to the capabilities of contemporary organic chemistry. However, taken together, they constitute a significant challenge to the goal of a stereocontrolled total synthesis of the epothilones. Below, we provide a summary of our activities that eventually accomplished this goal[8] for the first time for both epothilone A[8c] and epothilone B.[8e] In a concurrent time frame, studies from other laboratories accomplished these goals.[9,10] Moreover, the field of epothilone synthesis has spawned a host of interesting disclosures of potential impact on the long term total synthesis goal.[11]

Overall Synthetic Strategies

In pursuit of this program, a variety of initiatives were considered for assembling epothilone systems, including the natural products themselves. It is well to identify the main themes that were followed, often in parallel. In the earliest phase, our thinking was much influenced by the presence of the achiral domain encompassing carbons 9, 10, and 11. As noted above, this achiral region insulates the chiral "*O*-alkyl and acyl" sectors of the molecule from one another. We felt that the presence of this spacer element would pose a considerable difficulty in communicating stereochemical information from one chiral locus to the other. *Rather, it seemed appropriate to build the two chiral domains independently and to join them through carbon−carbon bond formation somewhere in the C9−C11 sector.* In this way, the integrity of chiral centers at C8 and C12, that are terminal to their respective chiral enclaves, would not be placed at risk in the crucial "merger" phase. One obvious possibility which presented itself in this regard was that of ring-forming olefin metathesis[12−14] (see approach I, Scheme 1). In this connection we were mindful of a seminal precedent disclosed by Hoveyda et al. in the context of synthesizing a macrolactam. Indeed, other laboratories, active in the epothilone field recognized, as did we, the potential pertinence of olefin metathesis to the epothilone area.[13]

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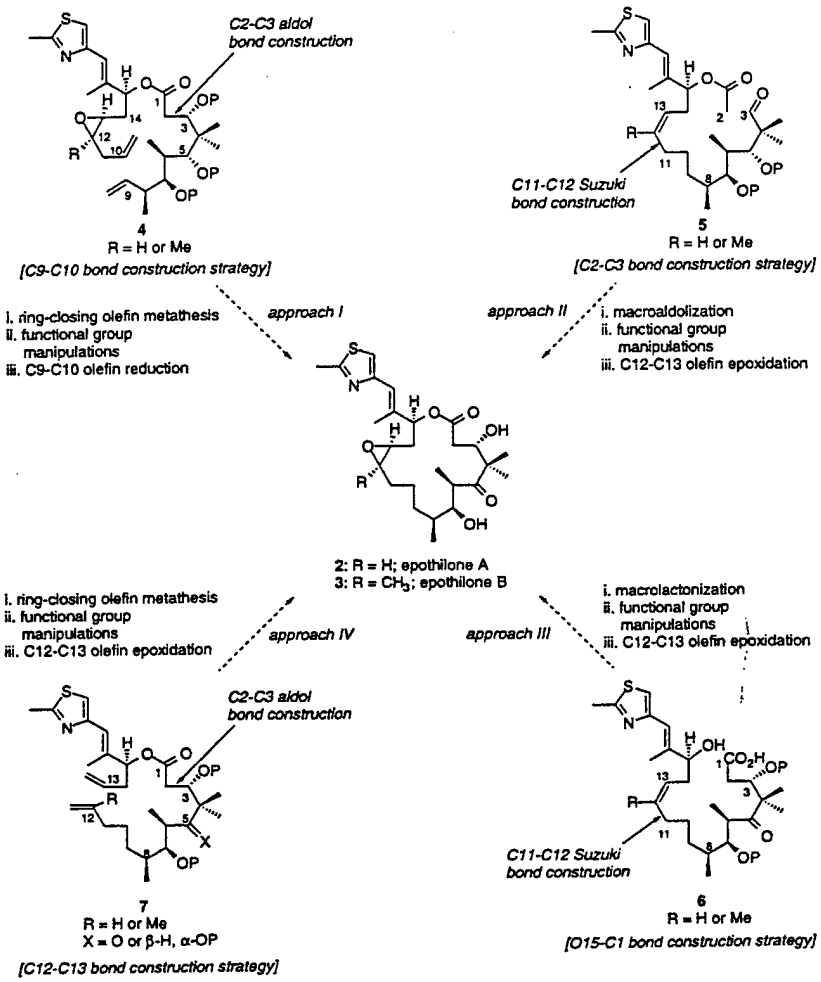
(13) For an outstanding demonstration of ring-closing olefin metathesis in the context of a natural product total synthesis, see: Houri, A. F.; Xu, Z.; Cogan, D. A.; Hoveyda, A. H. *J. Am. Chem. Soc.* **1995**, *117*, 2943. For the first published application on a model basis of the olefin metathesis concepts to the epothilone problem see refs 9a and 8b disclosed in that order. For the disclosures of the total synthesis of epothilone A based on olefin metathesis see refs 9b and 8d disclosed in that order.

(14) For recent examples of ring-closing olefin metathesis, see: (a) Martin, S. F.; Chen, H.-J.; Courtney, A. K.; Liao, Y.; Pätzel, M.; Ramser, M. N.; Wagman, A. S. *Tetrahedron* **1996**, *52*, 7251. (b) Fürstner, A.; Langemann, K. *J. Org. Chem.* **1996**, *61*, 3942.

Scheme 1. Bond Construction Strategies for Total Synthesis of Epothilone A (2) and B (3)



Refining the matter still further, we directed our attentions to a construction in which the olefin metathesis bond would join carbons 9 and 10 (see 4). An alternative approach wherein carbons 10 and 11 would be joined seemed riskier. The latter construction would have involved substrates in which there was an allylic relationship between the epoxide and the C10−C11 unsaturation in both the starting material and the product. Accordingly, the olefin metathesis prospectus that we came to favor called for a precursor of the general type 4 (P = unspecified protecting group). The resultant olefin would comprise carbons 9 and 10 of the goal system. Reduction of the olefin, followed by appropriate functional group manipulations, would then lead to target systems 2 and 3.

As will be shown, some surprising limitations in the ring-forming olefin metathesis reaction surfaced as we attempted to reduce this line of thinking to practice. When the full dimensions of the obstacles associated with a C9−C10 bond construction through ring-closing olefin metathesis were revealed, we came to focus on a fundamentally different assembly strategy wherein a double bond would be established between carbons 12 and 13 through ring-closing olefin metathesis. In this prospectus, the thiazole bearing chiral domain would display sp^3 asymmetry only at C-15. Carbons 12 and 13 would first be presented in the form of a *cis*-olefin, hopefully *en route* to a properly configured epoxide.

We also had occasion to contemplate an alternative synthetic logic, i.e. that of cross coupling, *wherein a bond would be fashioned between future carbons 11 and 12*. Specifically, we came to favor a *B*-alkyl Suzuki motif[15] to achieve this goal. In this line of reasoning, the fragments entering into this Suzuki coupling would be implied by a structure of the type **5**. From such a seco-compound, the 16-membered macrolide ring could be established through an intramolecular aldol addition,[16] giving rise to the C2−C3 bond (approach II). Alternatively, one could envision a post-Suzuki coupling structure of the type **6** which could set the stage for macrolide construction through macrolactonization[17] (approach III).

As will eventually be shown, the double bond at C-12 and C-13, either in the di- or trisubstituted series, could be very

(15) (a) Miyaura, N.; Ishiyama, T.; Sasaki, H.; Ishi-Kawa, M.; Satoh, M.; Suzuki, A. *J. Am. Chem. Soc.* **1989**, *111*, 314. (b) For an excellent review, see: Miyaura, N.; Suzuki, A. *Chem. Rev.* **1995**, *95*, 2457. (c) Johnson, C. R.; Braun, M. P. *J. Am. Chem. Soc.* **1993**, *115*, 11014. (d) Ohba, M.; Kawase, N.; Fujii, T.; Aoe, K.; Okamura, K.; Fathi-Afshar, R.; Allen, T. M. *Tetrahedron Lett.* **1995**, *36*, 6101.

(16) For a prior instance of a keto aldehyde macroaldolization, see: Hayward, C. M.; Yohannes, D.; Danishefsky, S. J. *J. Am. Chem. Soc.* **1993**, *115*, 9345.

(17) For a review of methods for constructing macrolides, see: Paterson, I.; Mansuri, M. M. *Tetrahedron* **1985**, *41*, 3569. The first macrolactonization in this series was demonstrated (though unknown to us at the time of our experiments) by Nicolaou and associates (reference 9c).

effectively exploited for introduction of the β-epoxide required for the final targets. As this feasibility was established, we also directed efforts to establishment of the C12−C13 bond through a ring-closing olefin metathesis reaction. An assessment of this interesting possibility would require the construction of a diolefin of the general type **7** (see approach IV, Scheme 1). This subgoal was accomplished. We now proceed to describe the progress attained in pursuing each of these strategies.

The First Generation Ring-Closing Olefin Metathesis Strategy (Approach I, Scheme 1)

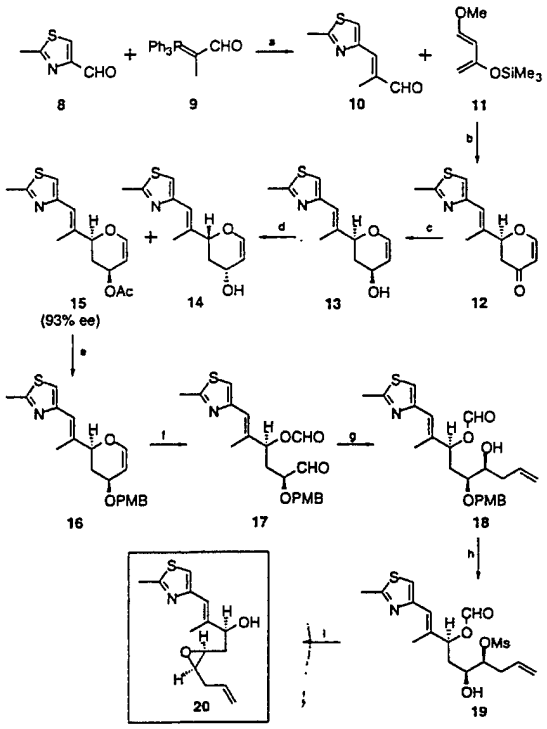
As was alluded to above, several options for constructing the 16-membered macrolide ring of the epothilones were considered as we contemplated strategies for a total synthesis. Initially, we favored a strategy wherein the C9−C10 would be fashioned during the course of the macrocyclization event. It was our hope that a structure of the general type **4** (see Scheme 1) could be induced to undergo a ring-closing olefin methathesis (RCM). Our considerations for favoring such an approach were several. In synthesizing and merging the subunits leading to **4**, all of the stereochemical problems associated with an epothilone total synthesis would have been overcome. The unsaturation at C9−C10 that would emerge from a successful ring-closing olefin metathesis could conceivably be removed by reduction *en route* to the epothilones, or used to introduce new functionality for the purpose of synthesizing novel analog structures.

As will be shown (*vide infra*), the guiding paradigm, *i.e.* the possibility of creating a C9−C10 double bond during the course of an intramolecular RCM process, was not reducible to practice in a subststrate having adequate functionality to reach the natural products. Nonetheless, many of our perceptions pertinent to the epothilone stereochemical problem and, indeed, some of the very compounds used in the pursuit of approach I, did find application to variations for the successful total syntheses. Hence, we describe here the findings pertinent to approach I, Scheme 1.

We defined goal system **20** (see Scheme 2) as a milestone compound under the approach I program. A structure of this genre would be joined to an acyl fragment (*vide infra*) to establish a precursor of the type hitherto generalized as **4**. Our path commenced with the known aldehyde **8**,[18] which was elongated in a Wittig-type construction with the commercially available phosphorane **9**, leading to **10** in 83% yield (see Scheme 2).[8a] At this stage, it was of interest to us to take advantage of a line of chemistry that our laboratory had innovated in the 1980s.[19] Thus, aldehyde **10** served as a "heterodienophile" in the context of a Lewis acid catalyzed diene−aldehyde cyclocondensation (LACDAC) reaction with the synergistic butadiene **11**.[20] The reaction proceeded quite smoothly, giving rise to the racemic dihydropyrone **12** in a yield of 65%.

Reduction of compound **12** via conditions that we had introduced some years ago for synthesizing artificial glycals bearing equatorial alcohols at C3 (glucose numbering),[21] led to racemic **13**. Here we were able to take advantage of more recently introduced methodology, wherein racemic glycals, derived by total synthesis rather than from carbohydrate sources, could be effectively resolved by lipase-mediated kinetic resolution.[22] In the event, we chose to carry out a kinetic resolution

Scheme 2[a]



[a] (a) C_6H_6, reflux (83%); (b) *trans*-1-methoxy-3-((trimethylsilyl)oxy)-1,3-butadiene (**11**), $BF_3{\cdot}OEt_2$, CH_2Cl_2; then CSA (65%); (c) $NaBH_4$, $CeCl_3{\cdot}7H_2O$, MeOH, 0 °C → rt (99%); (d) Lipase-30, vinyl acetate, DME, rt, (−)-**15** (45%; 93% ee); (e) (i) K_2CO_3, MeOH, rt; (ii) PMBCl, NaH, DMF, 0 °C → rt (97% overall); (f) 3,3-dimethyldioxirane, K_2CO_3, CH_2Cl_2, 0 °C; then $NaIO_4$, H_2O/THF (92%); (g) allyl triphenylstannane, $SnCl_4$, CH_2Cl_2, −78 °C (98% of **18** + epimer (4:1)); (h) (i) MsCl, Et_3N, CH_2Cl_2, 0 °C; (ii) DDQ, CH_2Cl_2/H_2O (20:1), 0 °C → rt (93% overall); (i) (i) $LiN(SiMe_3)_2$, THF, −78 → 0 °C; (ii) K_2CO_3, MeOH/H_2O (78% of the *cis* epoxide); PMB = *p*-$MeOC_6H_4CH_2$; Ms = SO_2CH_3.

through acetylation in the "forward sense". Following protocols of Wong,[23] the racemic alcohol was used in a transesterification experiment with vinyl acetate under the influence of Lipase-30 to afford alcohol **14** and acetate **15**. Of course, at this stage, we were in no position to assert the assignments of the absolute configuration to the antipodal glycal and glycal acetates with rigorous confidence. Rather, our tentative formulations arose from extensive precedents that had been garnered in our laboratory some years ago in this general area.[22] The presumed 3*S*-acetate **15** was subjected to deacetylation, giving rise to *ent*-**14**. The latter was subjected to the action of sodium hydride and *p*-methoxybenzyl chloride to afford **16**. In principle, it was initially supposed that compound **14** in the 3*R* series could be utilized in the synthesis program. However, as will be shown, an alternate route not requiring resolution to reach the desired 3*S* pyranoid series (cf. **16**) became available, and the possibility of recycling the 3*R* series available from the lipase chemistry was not pursued.

The next phase of the program called for disconnection of the C1−C2 bond of the artificial glycal **16** in such a fashion that C3 would emerge as C13 of the projected *cis*-epoxide **20**. We proceeded as follows. Drawing once again on chemistry that we had introduced in an earlier era for different purposes,[24]

(18) (a) Shafiee, A.; Mazloumi, A.; Cohen, V. I. *J. Heterocycl. Chem.* **1979**, *16*, 1563. (b) Schafiee, A.; Shahocini, S. *J. Heterocycl. Chem.* **1989**, *26*, 1627.

(19) (a) Danishefsky, S. J. *Aldrichim. Acta* **1986**, *19*, 59. (b) Danishefsky, S. J. *Chemtracts* **1989**, *2*, 273.

(20) Danishefsky, S. J.; Kitahara, T. *J. Am. Chem. Soc.* **1974**, *96*, 7807.

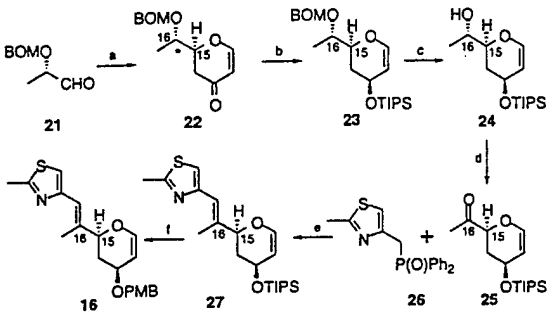
(21) Luche, J.-L. *J. Am. Chem. Soc.* **1978**, *100*, 2226.

(22) (a) Berkowitz, D. B.; Danishefsky, S. J. *Tetrahedron Lett.* **1991**, *32*, 5497. (b) Berkowitz, D. B.; Danishefsky, S. J. Schulte, G. K. *J. Am. Chem. Soc.* **1992**, *114*, 4518.

(23) Hsu, S.-H.; Wu, S.-S.; Wang, Y.-F.; Wong, C.-H. *Tetrahedron Lett.* **1990**, *31*, 6403.

(24) Danishefsky, S. J.; Bilodeau, M. T. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 1381.

Scheme 3[a]



[a] (a) *trans*-1-Methoxy-3-((trimethylsilyl)oxy)-1,3-butadiene (**11**), $MgBr_2 \cdot OEt_2$, THF, −10 °C; then AcOH, H_2O (93%); (b) (i) $NaBH_4$, $CeCl_3 \cdot 7H_2O$, MeOH, 0 °C; (ii) TIPSCl, imidazole, DMF, 0 °C → rt (87% overall); (c) Na^0, NH_3 (l), THF, −78 °C; then MeOH, −78 → 25 °C (92%); (d) Dess–Martin periodinane, pyridine, CH_2Cl_2, rt (98%); (e) **26**, *n*-BuLi, THF, −78 °C; then **25**, THF, −78 °C → rt (87%); (f) (i) *n*-Bu_4NF, THF, rt; (ii) PMBCl, NaH, DMF, 0 °C → rt (97% overall); BOM = CH_2OCH_2Ph; TIPS = Si*i*-Pr_3; PMB = *p*-$MeOC_6H_4CH_2$.

reaction of **16** with 3,3-dimethyldioxirane gave rise to an intermediate epoxide which, on oxidative solvolysis with sodium metaperiodate, afforded a 92% yield of aldehyde **17**. It was hoped that the emergence of the future oxygen at C15 (epothilone numbering) in the form of the formate ester would serve to prevent hemiacetal formation with the aldehyde group. Such a complicating event would have been anticipated if the C15 oxygen were free. While there was considerable apprehension as to whether the formate protecting device would be equal to the challenges with which it would be confronted, in practice this group survived during reaction of compound **17** with allyltriphenylstannane.[25] There was obtained a 96% yield of a 4:1 mixture of diastereomers at the future C12. The major product was assumed (and demonstrated on the basis of subsequent events) to be **18** in the relative configurational sense. Compound **18** was then subjected to mesylation, followed by deprotection of the *p*-methoxybenzyl group to give the hydroxy mesylate **19**. Finally, the sequence was completed by cyclization of the hydroxy mesylate with lithium hexamethyldisilazide. In this way, goal system **20** was obtained. Since compound **20** was indeed a *cis*-epoxide, the assignments of relative stereochemistry to compounds subsequent to the intermediate **12** had been substantiated. As for the absolute configuration of **20**, this assignment was proven by a sharply modified route (*vide infra*), which was undertaken to avoid the need for any resolution.

The pursuit of this modified route was also motivated by the desire to demonstrate still another dimension to the cyclocondensation reaction.[19] The concept involved the leveraging of chirality of heterodienophiles in the LACDAC reaction to create a pyran matrix of defined absolute configurations. This accomplished, the original asymmetries of the hetereodienophile can be abrogated, depending on the needs of the synthesis.

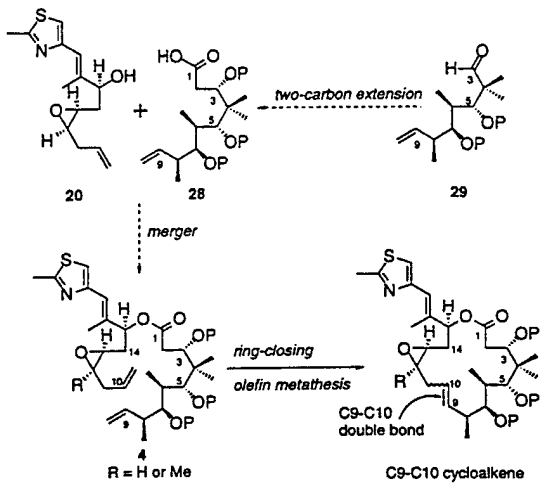
To teach this lesson, we proceeded as follows. Cyclocondensation of the known lactaldehyde derivative **21**[26] with **11** under mediation by magnesium bromide etherate gave rise to a 93% yield of a dihydropyrone (see Scheme 3). On the basis of earlier work,[27] we formulated this pyrone to be structure **22**. The importance of this assignment lies in the statement that it

(25) For a review of allylations, see: Yamamoto, Y.; Asao, N. *Chem. Rev.* **1993**, *93*, 2207.

(26) Heathcock, C. H.; Young, S. D.; Hagen, J. P.; Pirrung, M. C.; White, C. T.; VanDerveer, D. *J. Org. Chem.* **1980**, *45*, 3846.

(27) (a) Danishefsky, S. J.; Pearson, W. H.; Harvey, D. F. *J. Am. Chem. Soc.* **1984**, *106*, 2455. (b) Danishefsky, S. J.; Pearson, W. H.; Harvey, D. F.; Maring, C. J.; Springer, J. P. *J. Am. Chem. Soc.* **1985**, *107*, 1256.

Scheme 4. A C9–C10 Bond Construction through Ring-Closing Olefin Metathesis



makes about the absolute stereochemistry of the center destined to become C15. This center was presumed to be *S* as a consequence of α-chelation control in the cyclocondensation reaction. It will be appreciated that we had introduced an sp^3 chiral element which was *per se* unneeded, at the future trigonal C16 center (see the indicated carbon atom in **22**).

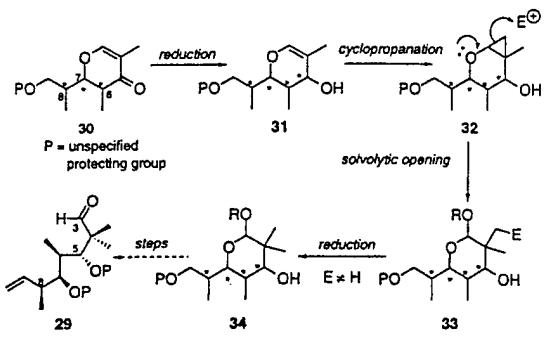
Compound **22** was integrated within the main synthetic pathway as follows. The ketone function of the dihydropyrone was reduced, as before, and the resultant alcohol at C3 (glycal numbering) was protected as its triisopropylsilyl ether (TIPS) derivative **23**. The benzyloxymethyl (BOM) group was discharged through the action of sodium in liquid ammonia and the alcohol function in the resultant **24** was subjected to oxidation using the Dess–Martin periodinane procedure.[28] This sequence provided **25** in 90% yield. This compound was, in turn, successfully condensed with phosphine oxide **26**[8a] in a Horner reaction,[29] thereby producing the elongated structure **27**. For purposes of stereochemical correlation, the silyl group was cleaved (*n*-Bu_4NF), and the resultant alcohol was reprotected as its *p*-methoxybenzyl ether. At this stage, we had achieved an alternate synthesis of compound **16** in a way that rigorously defined the relative stereochemistry as well as the absolute configuration. The conversion of intermediate **16** to **20** has already been discussed above.

With these successes as a platform, it was appropriate to focus on the construction of the acyl fragment projected for the olefin metathesis step. For this purpose, it would be appropriate to reach a carboxylic acid (cf. **28**, Scheme 4) for joining to alcohol **20** to reach system type **4**. We further presumed at the planning level that acid **28** would arise by a two carbon extension of a generic aldehyde (cf. **29**). In this section of the molecule, we would also be dealing with incorporation of the geminal methyl groups at the future C4 as well as the implementation of the appropriate chirality at carbons 6, 7, and 8. The chirality at C3 would have to be established during the course of the two carbon homologation alluded to above.

(28) (a) Dess, D. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4155. (b) Dess, D. B.; Martin, J. C. *J. Am. Chem. Soc.* **1991**, *113*, 7277. (c) Meyer, S. D.; Schreiber, S. L. *J. Org. Chem.* **1994**, *59*, 7549. (d) Ireland, R. E.; Liu, L. *J. Org. Chem.* **1993**, *58*, 2899.

(29) For examples, see: (a) Lythgoe, B.; Nambudiry, M. E. N.; Ruston, S.; Tideswell, J.; Wright, P. W. *Tetrahedron Lett.* **1975**, 3863. (b) Lythgoe, B. *Chem. Soc. Rev.* **1981**, 449. (c) Toh, H. T.; Okamura, W. H. *J. Org. Chem.* **1983**, *48*, 1414. (d) Baggiolini, E. G.; Iacobelli, J. A.; Hennessy, B. M.; Batcho, A. D.; Sereno, J. F.; Uskokovic, M. R. *J. Org. Chem.* **1986**, *51*, 3098.

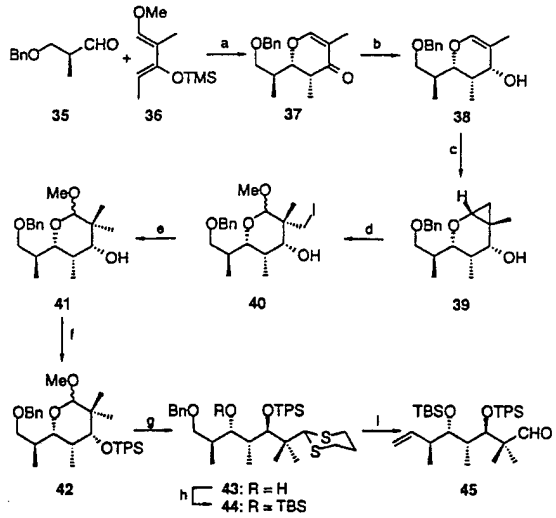
Scheme 5. General Strategy for a Synthesis of the Polypropionate Domain of the Epothilones



A central goal of our plan was the attainment of stereospecificity in the management of chirality at carbons 6, 7, and 8. As our thinking evolved (*vide infra*), it seemed that control over these issues could be facilitated if our scheme included a temporary chirality element at C5. Though C5 is destined to become a ketone, the temporary sp^3-associated chirality at this center would be very valuable in the management of stereochemistry in this region and in the introduction of the geminal methyl groups at C4. The possibility of using a dihydropyrone to address this problem presented itself.[8b] The thought was to translate the C-6, -7, and -8 domain of epothilone to correspond to dihydropyrone **30** (see Scheme 5). The latter could be accessed through cyclocondensation chemistry (*vide infra*). Hence, an artificial glycal (cf. **30**) was seen to be an exploitable intermediate *en route* to subgoal structure **29**. *More specifically, the thought was to utilize a C5 alcohol to facilitate and direct a cyclopropanation of the glycal double bond* (see **31** → **32**, Scheme 5).[30,31] A regiospecific solvolytic fragmentation of the cyclopropane ring in **32**[32] would then provide, in gross terms, an aldehyde equivalent of the type **33**. In the event that the crucial cyclopropane solvolysis step would be conducted in an oxidative sense (i.e. $E^+ \neq H$), it would then be necessary to effect a reduction of **33** to reach a compound of the type **34**. The latter would then be advanced, as appropriate, to reach the desired aldehyde **29**. This general thinking is summarized in Scheme 5.

In practice, titanium-mediated cyclocondensation of the known and optically pure β-(benzyloxy)isobutyraldehyde **35**[33] with diene **36**,[34] following a protocol previously devised in our laboratory,[35] gave rise to dihydropyrone **37** (see Scheme 6).[8b] Reduction of this compound with lithium aluminum hydride in ether provided glycal **38**. The hydroxyl group was then used

Scheme 6[a]



[a] (a) $TiCl_4$, CH_2Cl_2, −78 °C; then CSA, PhH, rt (87%); (b) $LiAlH_4$, Et_2O, −78 °C (91%); (c) Et_2Zn, CH_2I_2, Et_2O, rt (93%); (d) NIS (7 equiv), MeOH, rt; (e) *n*-Bu_3SnH, AIBN (cat.), PhH, reflux (80% from **39**; (f) Ph_3SiCl, imidazole, DMF, rt (97%); (g) 1,3-propanedithiol, $TiCl_4$, CH_2Cl_2, −78 → −40 °C (78%); (h) *t*-$BuMe_2SiOTf$, 2,6-lutidine, CH_2Cl_2, 0 °C (98%); (i) (i) 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ), CH_2Cl_2/H_2O (19:1), rt (89%); (ii) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C; then Et_3N, −78 °C → 0 °C (90%); (iii) CH_3PPh_3Br, $NaN(SiMe_3)_2$, $PhCH_3$, 0 °C → rt (76%); (iv) $PhI(OCOCF_3)_2$, $CH_2Cl_2/CH_3CN/H_2O$, rt (85%); Bn = CH_2Ph; TMS = $SiMe_3$; TPS = $SiPh_3$; TBS = Si*t*-$BuMe_2$.

to direct a cyclopropanation under modified Conia−Simmons−Smith conditions[31a] to afford cyclopropano derivative **39**. Oxidative solvolytic fragmentation of this cyclopropane was accomplished through the agency of *N*-iodosuccinimide in methanol to provide methyl glycoside **40**.[8b] Reductive deiodination of this compound led to the branched artificial methyl glycoside **41**, after which triphenylsilylation afforded the protected derivative **42**.

At this stage, it was timely to cleave the pyran ring with a view toward liberating the future aldehyde corresponding to C3 of the epothilones. Advancement *en route* to this goal involved subjecting compound **42** to the combined action of 1,3-propanedithiol and titanium(IV) chloride. This protocol led to the formation of the dithioacetal **43**.[36] Protection of the future C7 alcohol as shown (see compound **44**) was followed by olefin formation and liberation of the aldehyde function. In this way the specific compound **45** was in hand.

The next stage in the pursuit of approach I involved a two-carbon expansion starting with aldehyde **45**. The goal was the production of an acylation partner for alcohol **20** *en route* to a competent substrate for ring-closing olefin metathesis. In an early experiment, aldehyde **45** was treated with the lithium enolate of *tert*-butyl acetate (Rathke anion, see Scheme 7).[37]

(30) (a) Winstein, S.; Sonnenberg, J. *J. Am. Chem. Soc.* **1961**, *83*, 3235. (b) Dauben, W. G.; Berezin, G. H. *J. Am. Chem. Soc.* **1963**, *85*, 468. (c) Furukawa, J.; Kawabata, N.; Nishimura, J. *Tetrahedron* **1968**, *24*, 53. (d) For recent examples of oxygen-directed cyclpropanations of glycals, see: Hoberg, J. O.; Bozell, J. J. *Tetrahedron Lett.* **1995**, *36*, 6831. (e) For an excellent review of substrate-directable chemical reactions, see: Hoveyda, A. H.; Evans, D. A.; Fu, G. C. *Chem. Rev.* **1993**, *93*, 1307.

(31) For selected examples of glycal cyclopropanations, see: (a) Boeckman, R. K., Jr.; Charette, A. B.; Asberom, T.; Johnston, B. H. *J. Am. Chem. Soc.* **1991**, *113*, 5337. (b) Murali, R.; Ramana, C. V.; Nagarajan, M. *J. Chem. Soc., Chem. Commun.* **1995**, 217. (c) Henry, K. J., Jr.; Fraser-Reid, B. *Tetrahedron Lett.* **1995**, *36*, 8901. (d) Hoberg, J. O.; Claffey, D. J. *Tetrahedron Lett.* **1996**, *37*, 2533. (e) Timmers, C. M.; Leeuwenburgh, M. A.; Verheijen, J. C.; van der Marel, G. A.; van Boom, J. H. *Tetrahedron: Asymmetry* **1996**, *7*, 49. (f) Scott, R. W.; Heathcock, C. H. *Carbohydr. Res.* **1996**, *291*, 205. (g) For an excellent review of cyclopropanes in synthesis, see: Wong, H. N. C.; Hon, M.-Y.; Tse, C.-H.; Yip, Y.-C.; Tanko, J.; Hudlicky, T. *Chem. Rev.* **1989**, *89*, 165.

(32) (a) Collum, D. B.; Mohamadi, F.; Hallock, J. S. *J. Am. Chem. Soc.* **1983**, *105*, 6882. (b) Collum, D. B.; Still, W. C.; Mohamadi, F. *J. Am. Chem. Soc.* **1986**, *108*, 2094. (c) For an important early study, see: Wenkert, E.; Mueller, R. A.; Reardon, E. J., Jr.; Sathe, S. S.; Scharf, D. J.; Tosi, G. *J. Am. Chem. Soc.* **1970**, *92*, 7428.

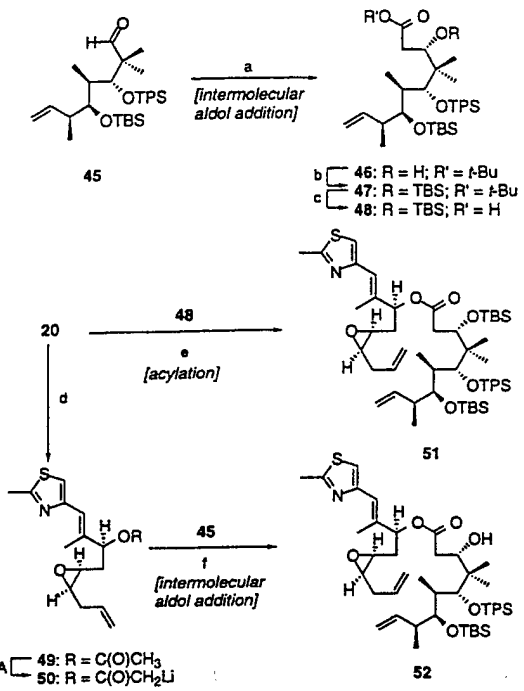
(33) (a) Cohen, N.; Eichel, W. F.; Lopresti, R. J.; Neukom, C.; Saucy, G. *J. Org. Chem.* **1976**, *41*, 3505. (b) Nagaoka, H.; Kishi, Y. *Tetrahedron* **1981**, *37*, 3873. (c) Meyers, A. I.; Babiak, K. A.; Campbell, H. L.; Comins, D. L.; Fleming, M. P.; Henning, R.; Heuschmann, M.; Hudspeth, J. P.; Kane, J. M.; Reider, P. J.; Roland, D. M.; Shimizu, K.; Tomioka, K.; Walkup, R. D. *J. Am. Chem. Soc.* **1983**, *105*, 5015. (d) Roush, W. R.; Palkowitz, A. D.; Ando, K. *J. Am. Chem. Soc.* **1990**, *112*, 6348.

(34) Danishefsky, S. J.; Yan, C.-F.; Singh, R. K.; Gammill, R. B.; McCurry, P. M., Jr.; Fritsch, N.; Clardy, J. *J. Am. Chem. Soc.* **1979**, *101*, 7001.

(35) (a) Danishefsky, S. J.; Pearson, W. H.; Harvey, D. F.; Maring, C. J.; Springer, J. P. *J. Am. Chem. Soc.* **1985**, *107*, 1256. (b) Danishefsky, S. J.; Myles, D. C.; Harvey, D. F. *J. Am. Chem. Soc.* **1987**, *109*, 862.

(36) For an example, see: Egbertson, M.; Danishefsky, S. J. *J. Org Chem.* **1989**, *54*, 11.

Scheme 7[a]



[a] (a) *t*-BuOC(O)CH_2Li, THF, 0 °C (90%; *ca.* 2.5:1 mixture of C-3 epimers in favor of **46**); (b) *t*-$BuMe_2SiOTf$, 2,6-lutidine, CH_2Cl_2, rt; (c) TESOTf, 2,6-lutidine, CH_2Cl_2, rt (90% overall); (d) Ac_2O, Et_3N, 4-DMAP, CH_2Cl_2, rt (94%); (e) **48**, EDC, CH_2Cl_2, 4-DMAP, rt; then **20** (78%); (f) **45** + **49**, LDA, THF, −78 °C (2−6:1 mixture of C3 epimers in favor of **52**; 85%); TBS = Si*t*-$BuMe_2$; TPS = $SiPh_3$.

There was thus generated a mixture of diastereomeric alcohols at the carbon destined to become C3 of epothilone. The major product was shown to have the required 3*S* configuration. The alcohol function in compound **46** was successfully protected as the *tert*-butyldimethylsilyl (TBS) ether derivative (see compound **47**). At this stage, it was possible to cleave the *tert*-butyl ester function to generate the acid **48**.

The coupling of the previously described alcohol **20** with acid **48** was conducted under the influence of 1-(3-(dimethylamino)-propyl)-3-ethylcarbodiimide hydrochloride (EDC) to produce, at long last, the proposed metathesis substrate **51** (see Scheme 7). It was also of interest to investigate the possibility of a more concise construction of a possible metathesis substrate. This approach started with acetylation of compound **20** to provide **49**. This acetate, when treated with lithium diisopropylamide, generated a presumed lithium ester enolate (see proposed structure **50**). This enolate underwent successful union with aldehyde **45** to produce a mixture of C3 epimers with the desired compound **52** predominating. A particularly interesting and efficient method for conducting the coupling of **49** and **45** involved merger of the two units in a "Barbier" sense.[38] *In this mode, the aldehyde and the ester would be concurrently exposed to the action of lithium diisopropylamide.* It was felt that such a treatment might be successful since the aldehyde is nonenolizable. In the event, an 85% yield of a ca. 5:1 mixture of C3 epimers was obtained with the major product being the 3*S* compound shown. This experiment was to have significant implications for our intramolecular aldol addition strategy which will be discussed under approach II.

(37) Rathke, M. W.; Sullivan, D. F. *J. Am. Chem. Soc.* **1973**, *95*, 3050.

(38) For a prior instance of a Barbier aldol reaction, see: Linde, R. G., II; Jeroncic, L. O.; Danishefsky, S. J. *J. Org. Chem.* **1991**, *56*, 2534.

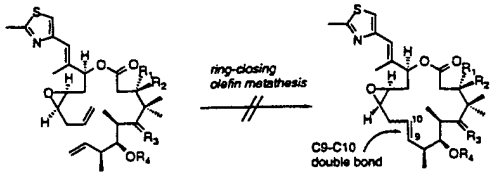
We now had in hand substrates **51** and **52**. Surprisingly, when these substrates were submitted to a range of conditions and catalysts designed to bring about ring-closing olefin metathesis, no convincing indication could be garnered for the presence of cyclized systems even to a small extent. While we could not exclude the possibility that trace amounts of desired products had been produced, the overwhelming bulk of the material was clearly of a very complicated nature, suggesting probable oligomerization. In any case, neither the protected system **51**, nor the structure bearing a free C3 alcohol in **52** served as usable substrates *en route* to epothilone A.

We did not take the setbacks in this early skirmishing with substrates **51** and **52** to necessarily establish the nonviability of the concept. We hoped that the feasibility of the RCM reaction could be sharply influenced by the nature and stereochemistry of the "decorating" substituents along the acyl chain. It seemed possible that certain substrates might be more amenable to cyclization in that conformational factors (in these variants) might help to predispose proximity between the terminal vinyl groups, or that properly selected substituents would be less conformationally obtrusive in the cyclic products which we were hoping to form on metathesis. In that spirit, we synthesized a wide variety of compounds as potential participants in the RCM reaction.[39] Unfortunately, none of these candidate substrates produced workable amounts of cyclized product. At best, mass spectrometric analysis indicated the possible formation of some desired materials. However, attempted isolation of traces of RCM products (assuming they were actually present) from very complex mixtures proved to be unsuccessful.

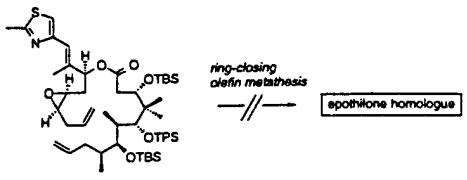
These facts, however disheartening, forced us to conclude that the row of substituents projecting from C3 through C8 had created an unmanageable problem of steric hindrance with respect to participation of the C9 double bond in the metathesis process. To probe this matter further, we went so far as to synthesize a compound which would, in itself, not constitute a promising intermediate for reaching epothilone. However, we thought that the study of the olefin metathesis possibility with this substrate could shed more light on the failures of the previously described entries. Accordingly, we synthesized compound **53**[40] in which the vinyl group on the acyl side was insulated from the secondary methyl group at C8 by another methylene group. Unhappily, homologous olefin **53** also failed to undergo ring-closing olefin metathesis.[40]

As described elsewhere,[8b] we went on to demonstrate to our satisfaction that the "culprit" in preventing the ring forming olefin metathesis reaction was the network of functionality between C3 and C8. There was nothing inherently unworkable

(39) For specific substrates tested in ring-closing olefin metathesis reactions, see Supporting Information.

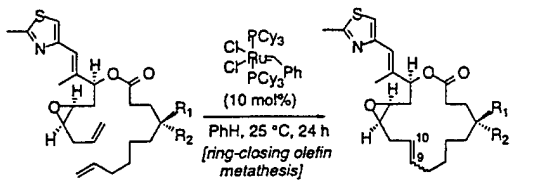


(40) Compound **53** was not a successful substrate for ring-closing olefin metathesis.



53

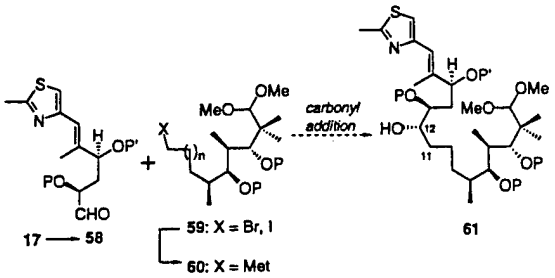
Scheme 8. Construction of the Compounds **56** and **57** through Ring-Closing Olefin Methathesis



54: R_1, R_2 = H
55: R_1, R_2 = CH_3

56: R_1, R_2 = H (*E*:*Z* *ca.* 1:1; 45%)
57: R_1, R_2 = CH_3 (one isomer; 70%)

Scheme 9. Intermolecular Carbonyl Addition Strategy for the Construction of the C11−C12 Bond



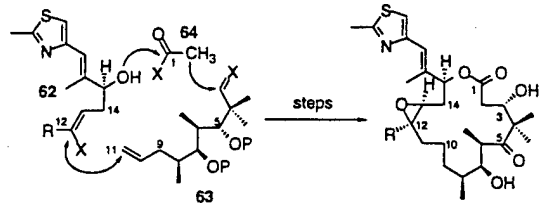
about inclusion of the homoallylic epoxide or the thiazolyl function, which were situated in the alkyl sector of the proposed premacrolide substrate. This "probable cause" argument was demonstrated by successful ring-closing metathesis reactions of substrates **54** and **55** (see Scheme 8). Unfortunately, while products **56** and **57**, respectively, could be obtained from such reactions in excellent yields, they did not display significant biological activity, either in tubulin binding or in cell-culture cytotoxicity studies.

We were now in the horns of what seemed to be an unsolvable dilemma. Those substrates in which olefin metathesis, leading to a C9−C10 cycloalkene (see Scheme 8), could be conducted provided products with nonuseful biological profiles. Conversely, those substrates which were functionalized in the spirit of serious potential epothilone precursors failed to undergo ring-closing olefin metathesis. It was when the full scope of this conundrum became clear that approach I was set aside in favor of other options.

Since we had demonstrated the ability to generate a protected aldehyde of the type **17** (Scheme 2), we turned to the possibility of a convergent coupling of this sort of system (generalized as **58**) with a conventional nucleophile in the form of a metallo derivative (generalized as **60**) (see Scheme 9). In the ideal scenario, **60** would be derived from **59**, in which case a product such as **61** could be anticipated.

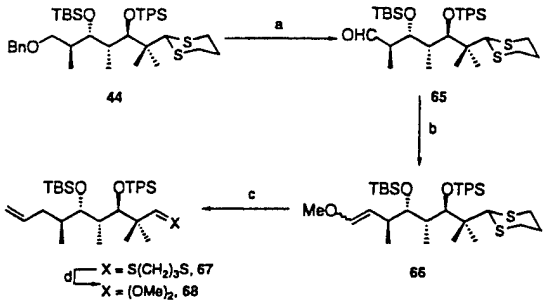
In practice, it proved possible to synthesize potential probe compounds for such metalation reactions (i.e. systems of the type **59**). Surprisingly, at no point were we able to accomplish the metalation of any such derivative to produce a competent organometallic nucleophile corresponding to **60**. In all cases, either unreacted aldehyde was recovered with the protonated metal species or decomposition took place. These failures were documented, not only in attempted couplings to the relatively sophisticated electrophile **17**, but even with much simpler electrophiles. We could garner little evidence to show that we had achieved metalation either in the series $n = 0$ or $n = 1$. These failures, suggesting problems in accessing external agents to the terminus of **59**, mirrored some of the difficulties which were soon to be encountered in the Suzuki coupling scheme (*vide infra*). Fortunately, this problem could be overcome in a novel way.

Scheme 10. C11−C12 Suzuki Bond Construction



2: R = H; epothilone A
3: R = CH_3; epothilone B

Scheme 11[a]



[a] (a) (i) DDQ, CH_2Cl_2/H_2O (89%); (ii) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C; then Et_3N, −78 → 0 °C (90%); (b) $MeOCH_2PPh_3Cl$, *t*-BuOK, THF, 0 °C → rt (86%); (c) (i) *p*-TSOH, dioxane/H_2O, 50 °C (99%); (ii) CH_3PPh_3Br, NaHMDS, $PhCH_3$, 0 °C → rt (76%); (d) PhI(O-$COCF_3)_2$, MeOH/THF, rt, 0.25 h (92%); Bn = CH_2Ph; TPS = $SiPh_3$; TBS = Si*t*-$BuMe_2$.

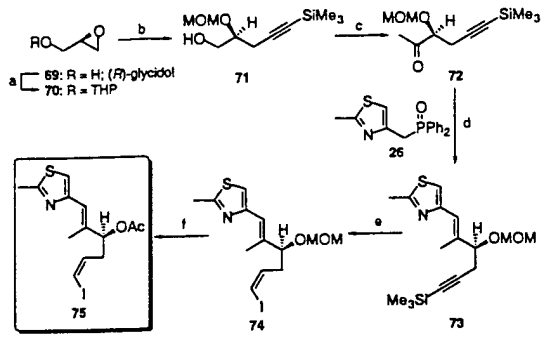
B-Alkyl Suzuki Strategy (Approaches II and III, Scheme 1)

We now report the results of the first successful syntheses of epothilones A and B which were achieved by the alkyl Suzuki method. The concept is generalized in Scheme 10 anticipating a synthesis of epothilones A and B. Through some as yet unspecified method, we envisioned a route to a *Z*-haloalkene **62**. Furthermore, it was expected that the chemistry would lend itself to construction of a terminal vinyl system in the context of a protected C3 substructure, generalized as system **63**. Construction of the C11−C12 bond would be the hallmark of the scheme and would be accomplished through a *B*-alkyl Suzuki coupling[15] (*vide infra*). Also to be dealt with would be a two carbon insert corresponding to carbons 1 and 2 of epothilone (see **64**). The appendage **64** could be incorporated in the scheme at several stages. If these carbon−carbon bond producing maneuvers were to be realized, there would also remain the need for introduction of the C12−C13 β-epoxide through the action of a suitable oxidizing agent. It was from these perceptions that our overall strategy for reaching epothilone A emerged. With suitable modification, a route to epothilone B was also embraced under this paradigm.

In practice, we turned to aldehyde **65** (Scheme 11). Compound **65** had been previously described as arising from **44** and had been converted to terminal vinyl compound **45** (Scheme 6). Now, the same aldehyde was successfully coupled to (methoxymethyl)triphenylphosphorane to give rise to **66**.[8c] The latter was then subjected to sequential hydrolysis and Wittig reactions to afford **67**. Finally, it proved possible to convert the dithiane linkage protecting C3 to the dimethyl acetal **68**. Here it was envisioned that the future C3 aldehyde could be revealed from the dimethyl acetal even in a mutlifunctionalized substrate.

With this chemistry in hand, we turned our attentions to the other projected Suzuki coupling partner. The specific version

Scheme 12[a]



[a] (a) Dihydropyran, PPTS, CH_2Cl_2, rt (73%); (b) (i) $Me_3SiCCLi$, $BF_3{\cdot}OEt_2$, THF, −78 °C (76%); (ii) MOMCl, *i*-Pr_2NEt, $Cl(CH_2)_2Cl$, 55 °C (85%); (iii) PPTS, MeOH rt (95%); (c) (i) $(COCl)_2$, DMSO, CH_2Cl_2, −78 °C; then Et_3N, −78 → rt; (ii) MeMgBr, Et_2O, 0 °C → rt (85% for two steps); (iii) TPAP, NMO, 4 Å mol sieves, CH_2Cl_2, 0 °C → rt (93%); (d) **26**, *n*-BuLi, THF, −78 °C; then **72**, THF, −78 °C → rt (97%); (e) (i) *N*-iodosuccinimide, $AgNO_3$, $(CH_3)_2CO$ (64%); (ii) dicyclohexylborane, Et_2O, AcOH (65%); (f) (i) PhSH, $BF_3{\cdot}OEt_2$, CH_2Cl_2, rt (86%); (ii) Ac_2O, pyr, 4-DMAP, CH_2Cl_2, rt (99%); PPTS = pyridinium *p*-toluenesulfonate; MOMCl = methoxymethyl chloride; TPAP = tetra-*n*-propylammonium perruthenate; NMO = *N*-methylmorpholine *N*-oxide.

of the generic structure **62** which was settled upon was the iodoacetate (see **75**, Scheme 12). Needless to say, it would be necessary to "deliver" this compound with the appropriate olefin geometry and in optically pure form for melding into the epothilone synthesis. In theory, depending on the coupling modality, we could utilize either enantiomeric version at C15; each enantiomer could be interfaced into the synthesis, subject to whether inversion or retention would be required at C15.

In our first route,[8c] we anticipated retention of configuration at this center (see Scheme 12). Accordingly, our program started with the commercially available *R*-(+)-glycidol (**69**).[41] The hydroxyl group was protected in the form of a THP ether (see compound **70**). In a defining step, the epoxide linkage was used to alkylate the lithium salt of (trimethylsilyl)acetylene, under the conditions described, to give rise to compound **71**. It will be recognized that the chiral center of glycidol is retained *en route* to coupling partner **75**.

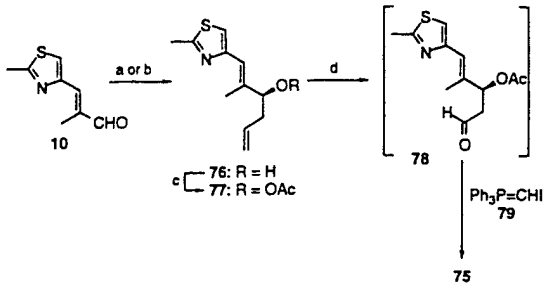
The next phase involved the classical transformation of the primary alcohol linkage to a methyl ketone. This was accomplished, as indicated, to provide ketone **72**. Drawing from an important precedent (see **25** + **26** → **27**, Scheme 3),[8a] we could accomplish the introduction of the thiazolyl nucleus through an Emmons reaction[29] of phosphine oxide **26** with methyl ketone **72**. In the concluding phase of this synthesis, silyl acetylene **73** was converted to the corresponding iodoalkyne which, upon reduction,[42] gave rise to the *cis*-iodoalkene **74**. Finally, cleavage of the MOM protecting group, as shown, followed by acetylation, produced the desired *cis*-vinyl iodide **75**.

When the general concept of the alkyl Suzuki coupling proved to be fruitful (*vide infra*), more concise syntheses of **75** were achieved. For this purpose, we returned to the enal **10**, a substance employed in an earlier stage of the synthesis.[8a] Allylation of this compound with tri-*n*-butylstannane in the presence of the (*S*)-BINOL enantiodirecting ligand, as described by Keck,[43] gave rise to the allylated product **76** in greater than

(41) For an excellent review of the chemistry of glycidol, see: Hanson, R. M. *Chem. Rev.* **1991**, *91*, 437.

(42) Corey, E. J.; Cashman, J. R.; Eckrich, T. M.; Corey, D. R. *J. Am. Chem. Soc.* **1985**, *107*, 713.

Scheme 13[a]



[a] (a) Allyltri-*n*-butylstannane, (*S*)-(−)-BINOL, $Ti(Oi\text{-}Pr)_4$, CH_2Cl_2, −20 °C (60%; >95% ee); (b) $[(-)\text{-Ipc}]_2BCH_2CHCH_2$, Et_2O, −100 °C; then 3 N NaOH, 30% H_2O_2 (83%; >95% ee); (c) Ac_2O, 4-DMAP, Et_3N, CH_2Cl_2 (96%); (d) (i) OsO_4, NMO, 0 °C; (ii) $NaIO_4$, THF/H_2O, rt (iii) **79**, THF, −78 → 0 °C (50% overall).

95% enantiomeric excess (see Scheme 13). Alternatively, we could effect an asymmetric allylation of enal **10** through the use of Brown's procedure.[44,9b] These transformations were generally faster and higher yielding but, of course, lacked the feature of implementation of chirality through catalytic means. The optical purity of allylic alcohol **76** prepared by allyl boration, was established by formation of the Mosher ester and subsequent analysis by ^1H and ^{19}F NMR spectroscopy. Eventually, the assignment and extent of optical purity were corroborated by interfacing this product with compound **75** derived from the glycidol route. Protection of carbinol **76** afforded acetate **77**. In a very delicate set of transformations, this homoallylic acetate was subjected to oxidative cleavage, as shown, to generate the putative β-acetoxy aldehyde **78**. That we had resorted, in the first instance, to the glycidol route reflected our fears that such a structure would be nonviable given its projected vulnerability to β-elimination of the cinnamyl-like acetoxy function. However, in practice, this difficulty could be managed. Wittig-type reaction with the known phosphorane **79**[45] gave rise to **75**. Certainly, this route proved to be more concise for reaching compound **75** than the *R*-glycidol based route shown in Scheme 12. The actual practicality of the process will depend on the feasibility of the scale up of the conversion of **77** to the vulnerable **78** *en route* to **75**.

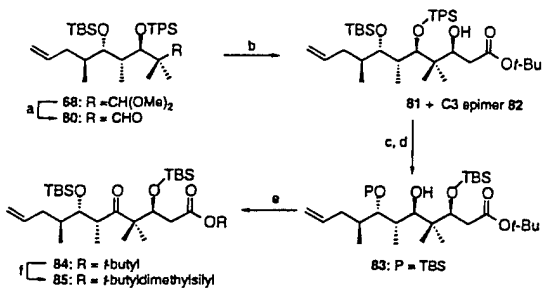
Even while this work was in progress, we were investigating a variation wherein the hypothetical Suzuki coupling would be conducted with a more advanced coupling partner, better positioned to reach epothilone itself. For this purpose, we returned to the acetal **68** which was deprotected to give rise to aldehyde **80** (see Scheme 14). This compound was condensed with lithio *tert*-butyl acetate[37] (i.e. the Rathke anion). There was produced a 63% yield of **81**, as well as its C3 stereoisomer (**82**, not shown here) in a ratio of approximately 2:1. The undesired C3 epimer could be oxidized to the corresponding ketone with Dess−Martin periodinane[28] and subsequently reduced in a stereoselective fashion to give the desired C3 alcohol exclusively. The major product **81** was subjected to the action of buffered HF·pyridine, whereupon the triphenylsilyl function was selectively cleaved from the C5 oxygen. It was further possible to selectively silylate the C3 alcohol through the combined action of TBS triflate and 2,6-lutidine, providing compound **83**. At this point, the C5 alcohol in compound **83** was oxidized to produce the ketone **84**. Finally, the *tert*-butyl

(43) Keck, G. E. Tarbet, K. H.; Geraci, L. S. *J. Am. Chem. Soc.* **1993**, *115*, 8467.

(44) Racherla, U. S.; Brown, H. C. *J. Org. Chem.* **1991**, *56*, 401.

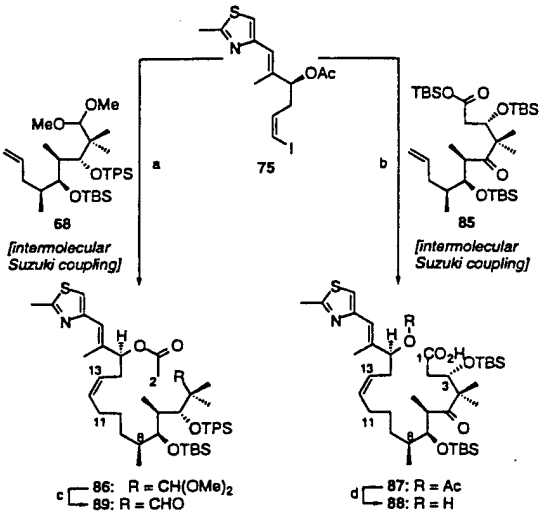
(45) (a) Stork, G.; Zhao, K. *Tetrahedron Lett.* **1989**, *30*, 2173. (b) Stork, G.; Zhao, K. *J. Am. Chem. Soc.* **1990**, *112*, 5875. (c) Chen, J.; Wang, T.; Zhao, K. *Tetrahedron Lett.* **1994**, *35*, 2827.

Scheme 14[a]



[a] (a) *p*-TsOH, dioxane/H_2O (5:1), 50 °C (81% overall); (b) *tert*-butyl acetate, LDA, THF, −78 °C, (95%; *ca.* 2:1 mixture of C-3 epimers); (c) HF·pyr, pyr, THF, rt (98%); (d) TBSOTf, 2,6-lutidine, CH_2Cl_2, −30 °C (96%); (e) Dess–Martin periodinane, CH_2Cl_2, rt (89%); (f) TBSOTf, 2,6-lutidine, CH_2Cl_2, rt (93%); TPS = $SiPh_3$; TBS = Si*t*-BuMe_2.

Scheme 15[a]



[a] (a) **68**, 9-BBN, THF, rt; then **75**, $PdCl_2(dppf)_2$, Cs_2CO_3, Ph_3As, H_2O/DMF, rt (75%); (b) **85**, 9-BBN, THF, rt; then **75**, $PdCl_2(dppf)_2$, Cs_2CO_3, Ph_3As, H_2O/DMF, rt (56%); (c) *p*-TsOH, dioxane/H_2O, 50 °C (85%); (d) K_2CO_3, MeOH/H_2O (84%); 9-BBN = 9-borabicyclo-[3.3.1]nonane; dppf = 1,1′-bis(diphenylphosphino)ferrocene; TPS = $SiPh_3$; TBS = Si*t*-BuMe_2.

ester could be converted to the *tert*-butyldimethylsilyl ester (see compound **85**) through the agency of TBS triflate and 2,6-lutidine.

We were now in a position to probe the feasibility of establishing the C11−C12 bond by Suzuki coupling in several contexts. Compound **68** was subjected to the action of 9-BBN (see Scheme 15). Remarkably, even the hydroboration of this terminal olefin required surprisingly coercive conditions. This slowness of hydroboration is reminiscent of the difficulties previously discussed in Scheme 9 for metalation of systems of the type **59** as a route to **60**. Fortunately, the hydroboration of **68** could be conducted under more vigorous conditions (as demonstrated by independent oxidative quenching experiments that revealed the anti-Markovnikov hydration product).

Having convinced ourselves that compound **68** had indeed been successfully hydroborated, we now conducted the last phase of the Suzuki reaction under mediation by palladium(II) chloride−1,1′-bis(diphenylphosphino)ferrocene (dppf) in the presence of the (*Z*)-vinyl iodide **75** as shown (Scheme 15). Gratifyingly, there was obtained a 72% yield of the acetate **86**.[8c] Within the limits of our detection, there had been no loss of stereointegrity of the C11−C12 double bond. The remarkable versatility of the *B*-alkyl Suzuki reaction[15] was further demonstrated by successful coupling of keto ester **85** with **75**. Under these circumstances, the TBS ester was cleaved during the course of the reaction, and the acetate was subsequently cleaved through the action of K_2CO_3 in aqueous methanol to give rise to the hydroxy acid **88**.

Our attentions would next be directed to the construction of the 16-membered ring. It was felt that our chances for achieving a stereoselective epoxidation would be better if the framework of the ring system were already in place when the oxidation of the C12−C13 double bond would be conducted. In our first attempt at macrocyclization, we favored a bold possibility. *The thought was to close the ring by connecting the methyl group of the acetate ester (C2) with the aldehyde center (C3) in a construct to be derived from compound 86.* That such a prospect could be even considered, arose from the fact that the *gem*-dimethyl substitution at C4 blocks the possibility of deprotonation of the aldehyde function. It will be recalled that earlier, in converting compound **45** to **52** (Scheme 7), we had exploited this principle by conducting an ester enolate aldol coupling under Barbier-type conditions.[38] Here, we would be drawing from the same concept in a macroaldolization step. To set the stage for this interesting ring-forming possibility, the acetal function in compound **86** was cleaved, thus revealing the electrophilic C3 aldehyde (see **86** → **89**, Scheme 15). In the crucial event, deprotonation of compound **89** (see Scheme 16) was accomplished through the action of potassium hexamethyldisilazide in THF at −78 °C. Remarkably, these conditions allow a stereoselective macroaldolization, resulting in the selective formation (6:1) of the desired (*S*)-C3 alcohol **90**. In some small scale experiments, compound **90** was the *only* product noted at the analytical level. However, optimal results from the standpoint of yield were actually obtained when the aldolate intermediate derived from cyclization was quenched at 0 °C or even at room temperature. When the quenching experiment was conducted at lower temperature, greater amounts of the undesired epimer **91** were obtained with an increase in mass recovery. Apparently, aldolate equilibration favors the formation of the desired 3*S* alcohol, whereas conditions more nearly approximating kinetic control give rise to lower degrees of stereoselectivity. At higher temperatures, it would seem that there is virtually no kinetic control in the stereochemistry of the macroaldolization step; the diastereomeric ratios observed indicate that equilibration occurs. While it is interesting to ponder and sort out methods to control stereoselectivity, in practice, the undesired epimer **91** could be utilized in our synthesis. Thus, oxidation of **91** to the ketone **92** set the stage for a diastereoselective reduction with $NaBH_4$ to provide the desired epimer **90** in high yield. Presumably, this outcome reflects the directing effects of the C5-OTPS function.

Cleavage of the triphenylsilyl ether in **90** could be conducted selectively, producing the C3−C5 diol (see compound **93**). Selective protection of the C3 hydroxyl in this compound was readily achieved, thus exposing the C5 alcohol in **94** for oxidation to a ketone. There was then produced di-TBS C12−C13 desoxyepothilone (**95**). Cleavage of the two silyl protecting groups could be accomplished, giving rise to desoxyepothilone A (**96**).

The whole scheme was now at considerable risk as we approached the matter of epoxidation of the C12−C13 double bond. We had hoped that oxidation would occur from the desired β-face on the basis of local conformational preferences that rendered this face of the molecule more accessible (see substructure **96** in Figure 2).[46] We further hoped to maximize our opportunities for stereocontrol by conducting the reaction

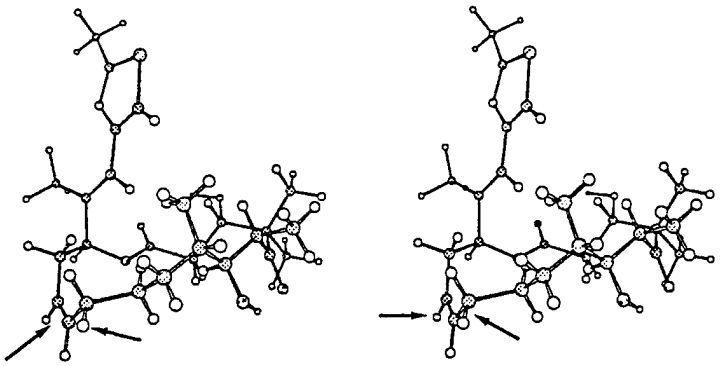


Figure 2. Macromodel minimized stereoview of desoxyepothilone A (**96**).

at low temperature. Given our many successful applications of the oxidizing powers of 3,3-dimethyldioxirane[24] it was not unnatural that we would turn to this reagent.

In practice, the major product of this reaction was the long sought after epothilone A (see **96** → **2**, Scheme 16), *confirmed by spectral and chromatographic comparisons of this material with authentic epothilone A kindly provided by Professor Höfle.* The first total synthesis of epothilone A had thus been accomplished. Two very minor products in the epoxidation were also isolated. One is the corresponding α-epoxide between C12 and C13. Still another product is one in which this epoxide linkage is present, but is accompanied by an epoxide functionality at C16–C17.

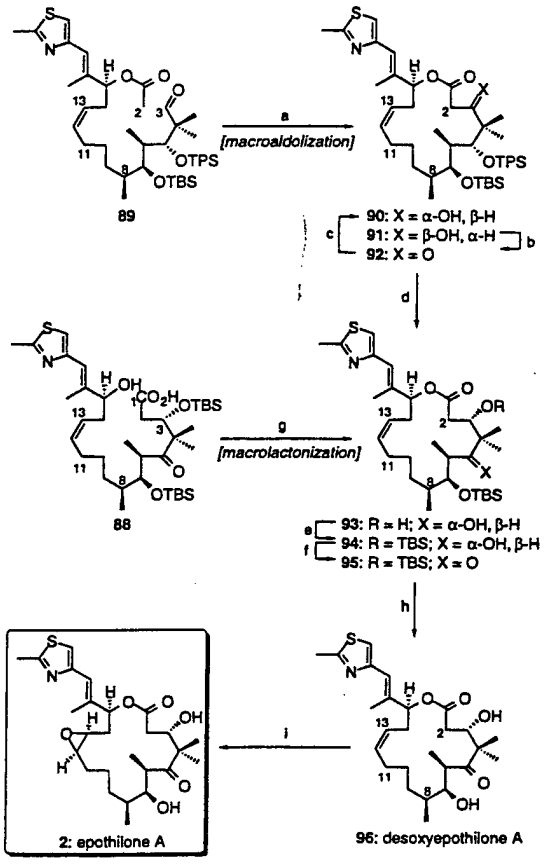
Although we had achieved the epoxidation reaction with high stereoselectivity, our reasoning based on model **96** can be questioned. Nicolaou and co-workers studied the use of the more conventional *m*-chloroperoxybenzoic acid.[9b,c] The use of this reagent produced mixtures of α and β epoxides. Hence, the factors controlling the facial sense of epoxidation are rather more subtle than is reflected in modeling of gross steric accessibility as suggested in **96**, since the course of oxygen delivery is strongly reagent dependent. However, we did have at our disposal a protocol for highly stereoselective epoxidation in the desired sense.

Having demonstrated the macroaldol route, we turned to the possibility of macrolactonization. This goal brought us back to compound **88** which, under Yamaguchi conditions,[47] led to the previously encountered **95**. Thus, we now had available to us two routes to enter the desoxyepothilone series in the form of compound **95** and, shortly thereafter, epothilone itself.

We next turn to our total synthesis of epothilone B (**3**) (see Scheme 17).[8e] We had hoped that this synthesis could be accomplished using, as much as possible, the chemistry that had served so well for the synthesis of epothilone A (**2**).[8c] Indeed, our route started with compound **77**, which was cleaved to the corresponding aldehyde **78**. Condensation of this aldehyde with the appropriate Wittig reagent[45c] gave rise to compound **97**, albeit in only 43% yield. Fortunately, the reaction was highly stereoselective, giving rise to the required *Z*-isomer as the only product. The stage was now set for the key Suzuki coupling. In this instance, we confined ourselves to acetal olefin **68**. Once again, hydroboration of **68** as before was followed by coupling of the resultant borane with (*Z*)-vinyl iodide **97**, giving rise to compound **98** in 77% yield. Cleavage of the acetal linkage led to aldehyde **99**. Once again, aldol condensation occurred in much the same manner as previously noted for compound **89**, thus allowing us to enter into the desoxyepothilone B series.

The protocols to reach desoxyepothilone B from cyclized material were already in hand from our synthesis of the A compound, **90**. Thus, in the case at hand, cleavage of the C5 TPS ether generated the diol **103**, which upon resilylation of

Scheme 16[a]

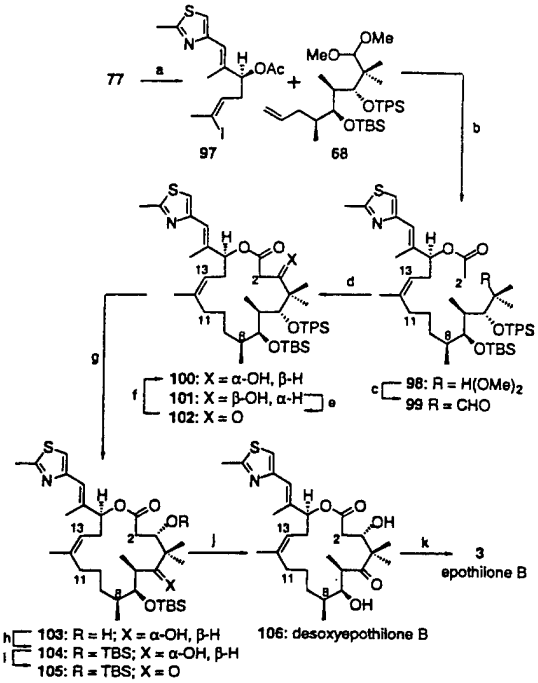


[a] (a) KHMDS, THF, −78 °C, 0.001M (51%, 6:1 α/β); (b) Dess–Martin periodinane, CH_2Cl_2, rt; (c) $NaBH_4$, MeOH, THF, −78 °C → rt (80% for two steps); (d) HF·pyridine, pyridine, THF, rt (99%); (e) TBSOTf, 2,6-lutidine, CH_2Cl_2, −30 °C (93%); (f) Dess–Martin periodinane, CH_2Cl_2, rt (84%); (g) 2,4,6-trichlorobenzoyl chloride, TEA, 4-DMAP, toluene, rt (88%); (h) HF·pyridine, THF, rt (99%); (i) 3,3-dimethyldioxirane, CH_2Cl_2, −35 °C (49%; ≥16:1 mixture of diastereomers in favor of **2**); TPS = $SiPh_3$; TBS = Si*t*-BuMe_2.

(46) Molecular modeling was performed with MacroModel version 5.5; The MMZ force field was used with a Monte Carlo random walk conformational search.

(47) (a) Yamaguchi, M.; Inanaga, J.; Hirata, K.; Saeki, H.; Katsuki, T. *Bull. Chem. Soc. Jpn.* **1979**, *52*, 1989. (b) Mulzer, J.; Mareski, P. A.; Buschmann, J.; Luger, P. *Synthesis* **1992**, 215.

Scheme 17[a]



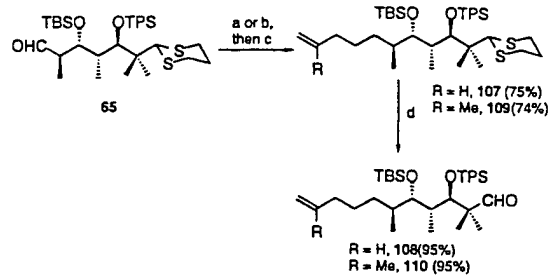
[a] (a) (i) OsO_4, NMO, $(CH_3)_2CO/H_2O$, 0 °C; (ii) $Pb(OAc)_4$, Na_2CO_3, C_6H_6, 0 °C → rt; (iii) $Ph_3P{=}C(I)CH_3$, THF, −20 °C (43% from **77**; *Z* geometrical isomer only); (b) **68**, 9-BBN, THF, rt; then **97**, $PdCl_2(dppf)_2$, Cs_2CO_3, Ph_3As, DMF/H_2O, rt (77%); (c) *p*-TsOH, dioxane/H_2O, 55 °C (71%); (d) KHMDS, THF, −78 °C (60%; **100**:**101**/2.1:1); (e) Dess–Martin periodinane, CH_2Cl_2, rt; (f) $NaBH_4$, MeOH, rt (67% for two steps); (g) HF·pyridine, pyridine, THF, rt (94%); (h) TBSOTf, 2,6-lutidine, CH_2Cl_2, −30 °C (89%); (i) Dess–Martin periodinane, CH_2Cl_2, rt (87%); (j) HF·pyridine, THF, rt (92%); (k) 3,3-dimethyldioxirane, CH_2Cl_2, −50 °C (97%; ≥20:1 mixture of diastereomeric *cis*-epoxides in favor of **3**); NMO = *N*-methylmorpholine *N*-oxide; 9-BBN = 9-borabicyclo[3.3.1]nonane; dppf = 1,1′-bis(diphenylphosphino)ferrocene; KHMDS = $KN(SiMe_3)_2$; TPS = $SiPh_3$; TBS = Si*t*-$BuMe_2$.

the exposed C3 hydroxyl group gave the product **104**. Oxidation of the C5 alcohol led to desoxyepothilone B *bis* (TBS) ether (compound **105**). Cleavage of the silyl blocking groups at C3 and C7 was accomplished, as shown, thereby allowing us to reach desoxyepothilone B (**106**). For obvious reasons, we turned to the use of dimethyl dioxirane in the oxidation of this compound. Happily, *this reaction was even more regio- and stereoselective, producing epothilone B (3) identical in all respects with an authentic sample, kindly provided by Professor Höfle.*

The Second Generation Ring-Closing Olefin Metathesis Strategy (Approach IV, Scheme 1)

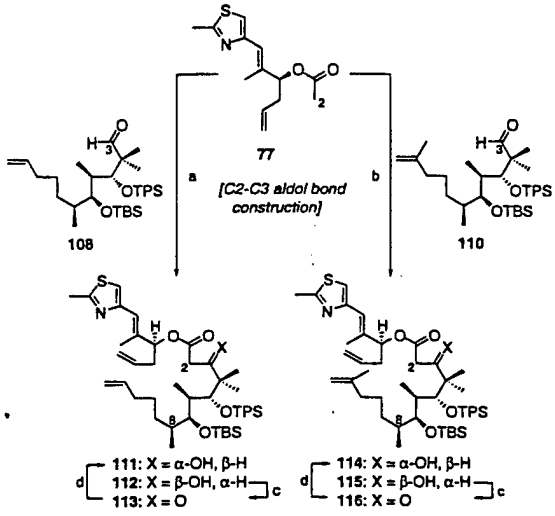
Even though we had accomplished our primary goals of synthesizing epothilones A and and B, it was still of interest to reinvestigate the possibility of intramolecular olefin metathesis.[8d] However, in this case we would be focusing on elaborating the C11–C12 double bond in the course of the metathesis reaction. Since we had known that epoxidation of this double bond could be conducted in a highly stereoselective and favorable direction, we were obviously more disposed to consider syntheses where this double bond would be elaborated in the decisive cyclization step. Toward this end, we returned to thioacetal aldehyde **65**. This compound was itself converted to product **107** through butenylation and deoxygenation at C9 (see Scheme 18). The C3 aldehyde function could be liberated by cleavage of the dithiane, as indicated, to provide aldehyde **108**.

Scheme 18[a]



[a] (a) 3-Butenylmagnesium bromide, Et_2O, −78 → 0 °C; (b) 4-iodo-2-methyl-1-butene, *t*-BuLi (2.1 equiv), Et_2O, −78 → −50 °C; then **65**, Et_2O, −78 → 0 °C; (c) (i) thiocarbonyl diimidazole, 4-DMAP, 95 °C; (ii) *n*-Bu_3SnH, AIBN, C_6H_6, 80 °C; (d) (i) $(CF_3CO_2)_2IC_6H_5$, MeOH/THF, rt; (ii) *p*-TsOH, dioxane/H_2O, 50 °C.

Scheme 19[a]



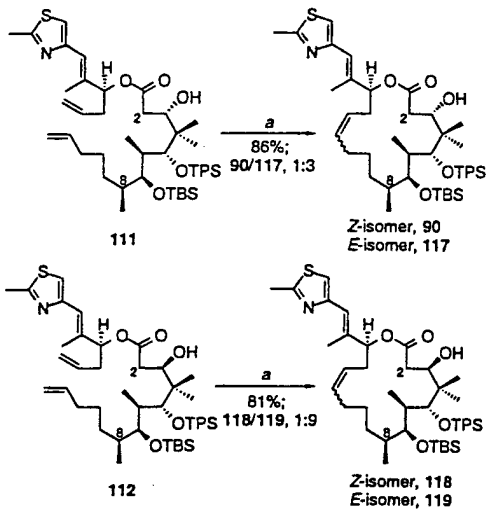
[a] (a) LDA, THF, −78 °C (65%; **111**:**112**/1:1); (b) LDA, THF, −78 °C (70%; **114**:**115**/*ca.* 1:1); (c) Dess–Martin periodinane, CH_2Cl_2, rt; (d) $NaBH_4$, MeOH, THF, −78 °C → rt (*ca.* 92% for two steps).

In order to probe the applicability of such a construction to the total synthesis of epothilone B, we returned to aldehyde dithiane **65**. The latter was converted to compound **109** by isobutenylation and deoxygenation. Once again, cleavage of the dithiane linkage provided aldehyde **110**.

With compounds **77** as well as **108** and **110** in hand, assembly of substrates for RCM were possible. To simplify the initial merger step, we turned, once again, to an intermolecular aldol condensation of the ester enolate, derived from **77**, with aldehydes **108** and **110** (see Scheme 19). In practice, the coupling could be readily conducted to give a mixture of stereoisomers at C3. The product, bearing the *S* configured alcohol, could be separated. The *R*-alcohol, in each case, was recycled through an oxidation/reduction sequence as shown.

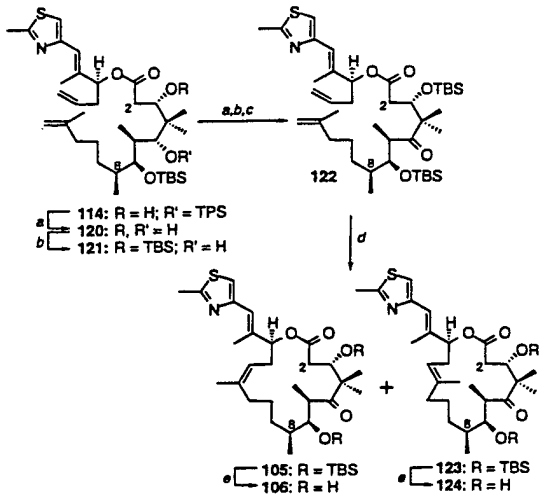
Indeed, with increased spacing between the C12 olefin and the branched positions of the polypropionate domain (see compound **111**), olefin metathesis chemistry proved to be successful.[8d] Cyclizations were conducted as described in Scheme 20 for compounds **111** and **112**. In these studies, we took recourse to both the ruthenium-based catalyst of Grubbs[12b] and the molybdenum-based catalyst of Schrock[12a] to mediate metathesis. In our work, the ruthenium-based system proved to be generally more efficacious for constructing the disubstituted double bonds with the properly configured (3*S*) alcohol.

Scheme 20[a]



[a] (a) $RuBnCl_2(PCy_3)_2$, 50 mol %, C_6H_6, 0.001 M rt, 24 h.

Scheme 21[a]

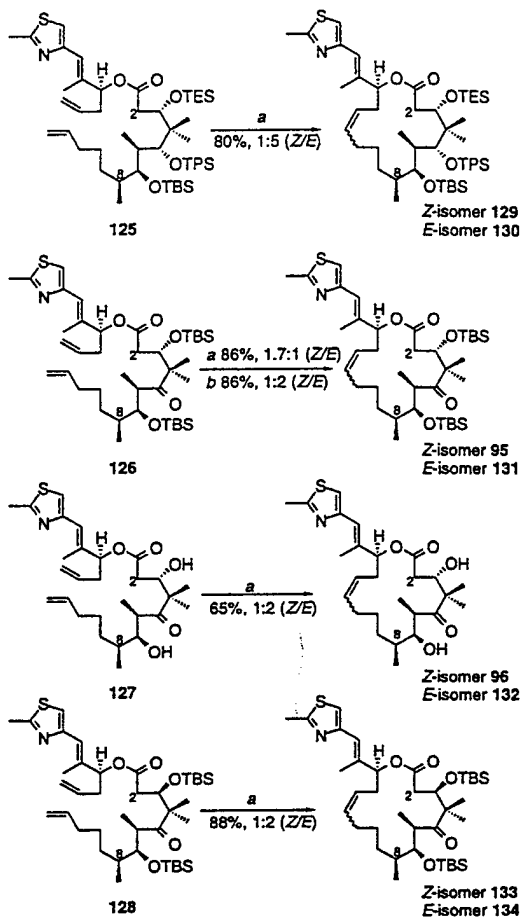


[a] (a) HF·pyr, pyr, THF, rt (93%); (b) TBSOTf, 2,6-lutidine, CH_2Cl_2, −30 °C (85%); (c) Dess−Martin periodinane, CH_2Cl_2 (94%); (d) $Mo(CHMe_2Ph)(N(2,6\text{-}(i\text{-}pr)_2C_6H_3))(OCMe(CF_3)_2)_2$ 20 mol %, C_6H_6, 0.001 M, 55 °C, 2 h, 86%, **105/123** 1:1; (e) HF·pyr, THF, rt, 2 h, 90%.

Although the yields of cyclized products were generally quite good, it was unfortunate that the resultant C12−C13 olefins were produced as a serious mixtures of *E*/*Z* isomers (**90**:**117**). The *Z* compounds could be correlated with earlier intermediates arising from the previously described *B*-alkyl Suzuki pathway. The *E* compounds were independently deprotected and converted to the corresponding *E* desoxyepothilone systems, as shown in Scheme 23 (*vide infra*).

Olefin metathesis was also attempted on compound **114** (see Scheme 21).[8e] In the event, this substrate failed to cyclize with the ruthenium-based system described by Grubbs,[12b] or the molybdenum based catalyst described by Schrock.[12a] However, when compound **122**, derived from **124** as shown, was treated with the Schrock catalyst, cyclization was successful producing a 1:1 mixture of *Z* and *E* isomers **105** and **123**.[8e] These products were independently processed, as shown, leading to the previously encountered *Z*-desoxyepothilone B (**106**) and *E*-desoxyepothilone B (**124**).

Scheme 22[a]

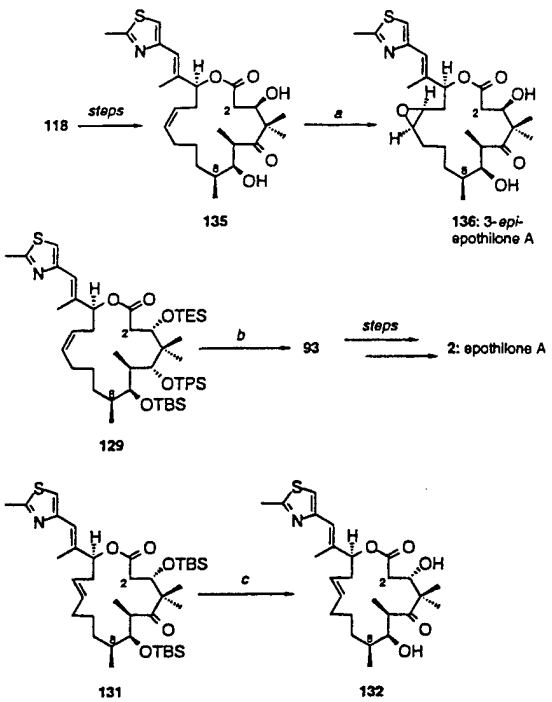


[a] (a) $RuBnCl_2(PCy_3)_2$, 50 mol %, C_6H_6, 0.001 M, rt, 4 h; (b) $Mo(CHMe_2Ph)(N(2,6\text{-}(i\text{-}pr)_2C_6H_3))(OCMe(CF_3)_2)_2$ 20 mol %, C_6H_6, 0.001 M, rt, 1 h, 86%.

As seen with substrates **111** and **112**, a point mutation of stereochemistry at C3 had tilted the process toward substantial stereoselectivity. Unfortunately, it was the 3-*epi* substrate (**112**) in which a high stereochemical margin was obtained, and, in fact, the unnatural *E*-double bond isomer was favored. We then mounted a considerable effort toward improving the stereoselectivity of the olefin metathesis reaction with the goal of reaching the natural *Z* series from the "natural" 3*S* carbinol precursor.

Unfortunately, no hypothesis emerged to guide our experiments in crafting the remote functions in the C3−C7 sector. Accordingly, we took recourse to intermediates that were accessible from the synthetic studies already in place. In this respect, we had occasion to prepare compounds **125** to **128** and to study their olefin metathesis.[8d] Using our collection of substrates, we were able to observe effects on the stereochemical course of olefin metatheses as a function of the nature of the substituents along the acyl chain. However, the only decisive perturbations favoring significant stereoselectivity were in the transformations of **112** to **118** and **119** (Scheme 20) as well as **125** to **129** and **130** (Scheme 22), each of which resulted in the predominant formation of the unnatural *E* compounds.

Scheme 23[a]



[a] (a) 3,3-Dimethyldioxirane, CH_2Cl_2, −30 °C, 70%, 3:1 β/α; (b) HF·pyr, THF, rt, 93%; (c) HF·pyr, pyr, THF, rt, 92%.

As is seen from the data, there is a sensitivity of olefin geometry to variation of substituents at even some distance from the terminal vinyl groups. Presumably, the consequence of these structural permutations reflect subtle effects on the sense of presentation of the vinyl group of one sector to the terminal metal−carbene complex[12] derived from the other sector. In this connection, it is also interesting that the olefin geometry ratio is also sensitive to the catalyst employed (see ratio of **95**:**131**) when this question was probed.

Compounds **118**, **129**, and **131** were processed as shown to afford 3-*epi*-epothilone A (**136**), epothilone A, and (*E*)-12,13-desoxyepothilone A (**132**) (Scheme 23). The results of evaluations of the biological profiles of these epothilone analogs have been published elsewhere[8d,e] and provide a basis for the development of new classes of structurally simpler and synthetically more accessible agents.

Summary

Several total syntheses of epothilones A and B have been described herein. These constructions involved union at either the C11−C12 bond (*B*-alkyl Suzuki coupling) or the C12−C13 bond (ring-forming olefin metathesis). Our routes differ from all others in several important respects. First, the stereochemistry of the polypropionate region accrues from the LACDAC reaction[19] and cyclic matrices elaborated therefrom. *Ultimately, all sterochemistry in this domain is induced from the single chiral center, readily derivable "Roche aldehyde" derivative 35 using sound principles formulated in our group many years ago.*[19]

Of particular importance is that C8 is incorporated in this dynamic. By contrast, the other syntheses have taken recourse to separate syntheses of α-methyl aldehydes, using chiral auxiliaries, to "deliver" the C8 chiral center to the synthetic pool. Furthermore, our syntheses uniquely provide strict control over the geometry of the double bonds of epothilone B as well as A, through adaptations of the B-alkyl Suzuki reaction. Recourse to the separation of *E*:*Z* isomer mixtures arising from olefin metathesis is, at least in our hands, seriously disabling in terms of throughput of significant amounts of material.

A stereoselective conversion of desoxyepothilone A and B to the epoxides in the natural series has been accomplished with the use of dimethyldioxirane (see conversion of **96** → **2** and **106** → **3**). Also illustrated in these studies were the power of catalytic asymmetric allylation (**10** → **76**), the flexibility of glycidol as a multifacated member of the chiral pool (see **69** → **75**), and the power of the LACDAC reaction in assembling polypropionate frameworks (see **35** + **36** → **45**, **68** and **85**). The rather novel cyclopropanation of a glycal (**38** → **39**) followed by oxidative solvolysis (**39** → **40**) and reduction (**40** → **41**) as a route to the introduction of quaternary branching is also deserving of attention. Given the generality of the issues which have been addressed, it is likely that the lessons garnered here would find application to other problems in organic synthesis.

Experimental Section

General. All commercial materials were used without further purifications unless otherwise noted. The following solvents were distilled under positive pressure of dry nitrogen immediately before use: THF and diethyl ether from sodium/potassium−benzophenone ketyl, CH_2Cl_2, toluene, and benzene from CaH_2. All the reactions were performed under N_2 atmosphere. NMR (^1H, ^{13}C) spectra were recorded on Bruker AMX-400 MHz, Bruker Avance DRX-500 MHz, referenced to TMS (^1H-NMR, δ 0.00) or $CDCl_3$ (^{13}C-NMR, δ 77.0) peaks unless otherwise stated. LB = 1.0 Hz was used before Fourier transformation for all of the ^{13}C-NMR. IR spectra were recorded with a Perkin-Elmer 1600 series-FTIR spectrometer, and optical rotations were measured with a Jasco DIP-370 digital polarimeter using 10 cm pathlength cell. Low-resolution mass spectral analysis were performed with a JEOL JMS-DX-303 HF mass spectrometer. Analytical thin-layer chromatography was performed on E. Merck silica gel 60 F_{254} plates (0.25 mm). Compounds were visualized by dipping the plates in a cerium sulfate−ammonium molybdate solution followed by heating. Flash column chromatography was performed using the indicated solvent on E. Merck silica gel 60 (40−63 mm) or Sigma H-Type silica gel (10−40 mm). Melting points are obtained with Electrothermal melting point apparatus (series no. 9100) and are uncorrected.

Preparation of Compound 68. A solution of (methoxymethyl)-triphenylphosphonium chloride (2.97 g, 8.55 mmol) in THF (25 mL) at 0 °C was treated with KO^{t}Bu (8.21 mL, 1 M in THF, 8.1 mmol). The mixture was stirred at 0 °C for 30 min. Aldehyde **65** (3.10 g, 4.07 mmol) in THF (10 mL) was added, and the resulting solution was allowed to warm to rt and stirred at this temperature for 2 h. The reaction was quenched with saturated aqueous NH_4Cl (40 mL), and the resulting solution was extracted with Et_2O (3 × 30 mL). The combined Et_2O fractions were washed with brine (20 mL), dried over $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromatography on silica gel eluting with 5% Et_2O in hexanes to yield the methyl enol ether **66** (2.83 g, 86%) as a colorless foam.

To a solution of the methyl enol ether **66** (2.83 g, 3.50 mmol) in dioxane/H_2O (9:1, 28 mL) was added pTSA·H_2O (1.0 g, 5.30 mmol), and the resulting mixture was heated to 50 °C for 2 h. After cooling to rt, the mixture was diluted with Et_2O (50 mL) and washed successively with saturated aqueous $NaHCO_3$ (15 mL) and brine (20 mL), dried over $MgSO_4$, filtered, and concentrated to provide the corresponding aldehyde (2.75 g, 99%) as a colorless foam: $[\alpha]_D$ = +1.74 (c = 0.77, $CHCl_3$); IR (film) 2929, 1725, 1428, 1253, 1115, 1039 cm^{-1}; ^1H NMR ($CDCl_3$, 500 MHz) δ 9.31 (d, J = 3.4 Hz, 1 H), 7.68 (dd, J = 7.8, 1.4 Hz, 6 H), 7.45−7.37 (band, 9 H), 4.60 (d, J = 6.8 Hz, 1 H), 4.20 (s, 1 H), 3.51 (d, J = 6.7 Hz, 1 H), 2.68 (d, J = 14.0 Hz, 1 H), 2.60 (d, J = 13.5 Hz, 1 H), 2.37 (m, 1 H), 2.24 (m, 1 H), 1.90 (m, 1 H), 1.81 (m, 2 H), 1.68 (m, 2 H), 1.52 (m, 1 H), 1.32 (s, 3 H), 1.14−1.03 (band, 6 H), 0.86 (s, 9 H), 0.75 (d, J = 6.9 Hz, 3 H), −0.03 (s, 3 H), −0.06 (s, 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 202.8, 136.0, 134.6, 130.1, 128.0, 77.6, 76.2, 59.5, 45.1, 44.7, 43.8,

31.8, 30.9, 30.5, 26.3, 25.9, 22.5, 20.9, 18.6, 17.9, 14.7, −3.0, −3.5; HRMS calcd for $C_{39}H_{56}O_3S_2Si_2$: 692.3210; found: 731.2828 (M + K).

Methyltriphenylphosphonium bromide (1.98 g, 5.54 mmol) in THF (50 mL) at 0 °C was treated with lithium bis(trimethylsilyl)amide (5.04 mL, 1 M in THF, 5.04 mmol), and the resulting solution was stirred at 0 °C for 30 min. The aldehyde (2.00 g, 2.52 mmol), prepared above, in THF (5.0 mL) was added, and the mixture was allowed to warm to room temperature and stirred at this temperature for 1 h. The reaction was quenched with saturated aqueous NH_4Cl (15 mL) and extracted with Et_2O (3 × 20 mL). The combined Et_2O fractions were washed with brine (15 mL), dried over $MgSO_4$, filtered, and concentrated. The residue was purified by flash chromatography on silica gel eluting with 5% Et_2O in hexanes to afford compound **67** (1.42 g, 76%) as a colorless foam.

A solution of the dithiane **67** (1.0 g, 1.34 mmol), prepared above, in MeOH/THF (2:1, 13 mL) was treated with [bis(trifluoroacetoxy)-iodobenzene] (0.865 g, 2.01 mmol) at rt. After 15 min, the reaction was quenched with saturated aqueous $NaHCO_3$ (25 mL). The mixture was extracted with Et_2O (3 × 25 mL), and the combined Et_2O fractions were washed once with brine (20 mL), dried over $MgSO_4$, filtered, and concentrated. Purification of the residue by flash chromatography on silica gel eluting with 5% Et_2O in hexanes provided compound **68** (0.865 g, 92%) as a colorless foam: $[\alpha]_D = +1.74$ ($c = 0.77$, $CHCl_3$); IR (film) 1428, 1252, 1114, 1075, 1046 cm^{-1}; ^1H NMR ($CDCl_3$, 500 MHz) δ 7.61 (dd, $J = 7.9$, 1.4 Hz, 6 H), 7.38 (s, 9 H), 5.47 (m, 1 H), 4.87 (d, $J = 10.0$ Hz, 1 H), 4.76 (d, $J = 15.9$ Hz, 1 H), 4.30 (d, $J = 3.7$ Hz, 1 H), 3.95 (s, 1 H), 3.56 (dd, $J = 7.5$, 1.4 Hz, 1 H), 3.39 (s, 3 H), 2.84 (s, 3 H), 2.02 (m, 1 H), 1.64 (m, 2 H), 1.34 (m, 1 H), 1.11 (s, 3 H), 1.02 (d, $J = 7.4$ Hz, 3 H), 0.90 (s, 3 H), 0.85 (s, 9 H), 0.62 (d, $J = 6.8$ Hz, 3 H), −0.04 (s, 3 H), −0.05 (s, 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 138.3, 135.8, 135.0, 129.9, 127.8, 114.9, 110.5, 60.1, 55.6, 46.5, 43.9, 36.8, 34.2, 26.3, 19.6, 18.6, 17.1, 16.16, 13.9, −2.9, −3.8; HRMS calcd for $C_{39}H_{58}O_4Si_2$: 646.3873; found: 685.3491 (M + K).

Preparation of Compound 76. A mixture of (*S*)-(−)-1,1′-bi-2-naphthol (0.259 g, 0.91 mmol), $Ti(O\text{-}i\text{-}Pr)_4$ (261 mL; 0.90 mmol), and 4 Å sieves (3.23 g) in CH_2Cl_2 (16 mL) was heated at reflux for 1 h. The mixture was cooled to rt, and aldehyde **10** was added. After 10 min, the suspension was cooled to −78 °C, and allyltri-*n*-butyltin (3.60 mL, 11.6 mmol) was added. The reaction mixture was stirred for 10 min at −78 °C and then placed in a −20 °C freezer for 70 h. Saturated aqueous $NaHCO_3$ solution (2 mL) was added, and the mixture was stirred for 1 h, poured over Na_2SO_4, and then filtered through a pad of $MgSO_4$ and Celite. The crude material was purified by flash chromatography (hexanes/ethyl acetate, 1:1) to give alcohol **76** as a yellow oil (1.11 g, 60%): $[\alpha]_D = -15.9$ (*c* 4.9, $CHCl_3$); IR (film) 3360, 1641, 1509, 1434, 1188, 1017, 914 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$, 25 °C) δ 6.92 (s, 1 H), 6.55 (s, 1 H), 5.82 (m, 1 H), 5.13 (dd, $J = 17.1$, 1.3 Hz, 1 H), 5.09 (d, $J = 10.2$ Hz, 1 H), 4.21 (t, $J = 6.0$ Hz, 1 H), 2.76 (br s, 1 H), 2.69 (s, 3 H), 2.40 (m, 2 H), 2.02 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$, 25 °C) δ 164.5, 152.6, 141.5, 134.6, 119.2, 117.6, 115.3, 76.4, 39.9, 19.0, 14.2; HRMS calcd for $C_{11}H_{15}NOS$: 209.0874 found: 209.0872 (M + H).

Preparation of Compound 77. To a solution of alcohol **76** (0.264 g; 1.26 mmol) in CH_2Cl_2 (12 mL) were added 4-DMAP (0.015 g, 0.098 mmol), Et_3N (0.45 mL; 3.22 mmol), and Ac_2O (0.18 mL; 1.90 mmol). After 2 h, the reaction was quenched by the addition of H_2O (20 mL) and extracted with EtOAc (4 × 20 mL). The combined organic extracts were dried with $MgSO_4$, filtered, and concentrated. Flash chromatography on SiO_2 (EtOAc/hexanes, 1:3) afforded acetate **77** as a yellow oil (0.302 g; 96%): $[\alpha]_D = -40.0$ (*c* 7.3, $CHCl_3$); IR (film) 1738, 1505, 1436, 1370, 1236, 1019 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$, 25 °C) δ 6.95 (s, 1 H), 6.52 (s, 1 H), 5.72 (m, 1 H), 5.33 (t, $J = 6.6$ Hz, 1 H), 5.10 (ddd, $J = 17.1$, 3.1, 1.5 Hz, 1 H), 5.07 (ddd, $J = 10.2$, 3.3, 1.7 Hz, 1 H), 2.70 (s, 3 H), 2.48 (dt, $J = 5.9$, 1.3 Hz, 2 H), 2.08 (s, 3 H), 2.07 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$, 25 °C) δ 170.1, 164.5, 152.4, 137.0, 133.4, 120.6, 117.6, 116.2, 77.9, 37.5, 21.1, 19.1, 14.7; HRMS calcd for $C_{13}H_{17}O_2NS$: 251.0980; found: 251.0983 (M^-).

Preparation of Compound 75. To a solution of acetate **77** (0.099 g; 0.39 mmol) in acetone (10 mL) at 0 °C were added H_2O (4 drops), OsO_4 (2.5% wt in butyl alcohol; 0.175 mL; 0.018 mmol), and *N*-methylmorpholine *N*-oxide (0.069 g; 0.59 mmol). The mixture was stirred at 0 °C for 2 h and then quenched with saturated aqueous Na_2SO_3 solution (10 mL). The solution was poured into H_2O (10 mL) and extracted with EtOAc (8 × 10 mL). The combined organic layer was dried over $MgSO_4$, filtered, and concentrated.

To a solution of the crude diol in THF/H_2O (4 mL, 3:1) was added $NaIO_4$ (0.260 g; 1.22 mmol). After 1.25 h, the reaction mixture was quenched with H_2O (10 mL) and concentrated. The aqueous layer was extracted with EtOAc (5 × 10 mL) and dried over $MgSO_4$. Flash chromatography (SiO_2, EtOAc/hexanes, 1:1) on a short pad of silica gave the crude aldehyde **78** as a yellow oil (0.080 g) which contained unidentified byproduct(s). This mixture was used without further purification.

To a solution of $(Ph_3P^+CH_2I)I^-$ (0.100 g; 0.19 mmol) in THF (0.25 mL) at rt was added sodium bis(trimethylsilyl)amide (1 M soln in THF, 0.15 mL, 0.15 mmol). To the resulting solution at −78 °C were added HMPA (0.022 mL; 0.13 mmol) and the crude aldehyde **78** from the previous step (0.016 g) in THF (0.25 mL). The reaction mixture was then stirred at rt for 30 min. After the addition of saturated aqueous NH_4Cl (10 mL), and the solution was extracted with EtOAc (4 × 10 mL). The combined organic extracts were dried ($MgSO_4$), filtered, and concentrated. The residue was purified by preparative thin-layer chromatography (prep-TLC) (EtOAc/hexanes, 2:3) to give the vinyl iodide **75** as a yellow oil (0.014 g; 50% for three steps).

Preparation of Compound 80. The acetal **68** (0.930 g, 1.44 mmol) was dissolved in dioxane/H_2O (9:1, 20 mL), and *p*TSA·H_2O (0.820 g, 4.32 mL) was added. The mixture was heated at 55 °C for 2 h. After cooling to rt, the solution was poured into Et_2O (200 mL) and washed once with saturated aqueous $NaHCO_3$ solution (30 mL) and once with brine (30 mL) and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/ethyl acetate (9:1) gave 0.702 g (81%) of the aldehyde **80** as a white foam: $[\alpha]_D = -12.8$ ($c = 3.4$, $CHCl_3$); IR (film) 2929, 1722, 1472, 1429, 1256, 1115, 1059 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 9.76 (s, 1 H), 7.71 (d, $J = 6.4$ Hz, 6 H), 7.42 (m, 9 H), 5.49 (m, 1 H), 4.87 (m, 1 H), 4.75 (m, 1 H), 4.08 (d, $J = 1.6$ Hz, 1 H), 3.56 (dd, $J = 1.6$, 8.7 Hz, 1 H), 2.18 (m, 1 H), 1.70 (m, 1 H), 1.46 (m, 2 H), 1.10 (s, 3 H), 0.89 (d, $J = 6.4$ Hz, 3 H), 0.79 (s 9 H), 0.60 (d, $J = 6.5$ Hz, 3 H), −0.8 (s, 3 H), −0.83 (s, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 205.5, 137.6, 135.6, 134.2, 130.0, 127.9, 115.4, 80.7, 76.4, 51.7, 43.5, 38.2, 34.9, 26.1, 20.9, 20.1, 18.5, 15.5, 12.9, −3.3, −3.9; HRMS calcd for $C_{37}H_{52}O_3S_2$: 600.3455; found: 623.3333 (M + Na).

Preparation of Compound 81. The aldehyde **80** (0.702 g, 1.17 mmol) was dissolved in THF (50 mL), and *tert*-butyl acetate (1.26 mL, 9.36 mmol) was added. The solution was cooled to −78 °C, and LDA (2.0 M soln, 3.51 mL, 7.02 mmol) was added. After 20 min, the reaction was quenched with MeOH (10 mL) and H_2O (100 mL). The mixture was extracted with Et_2O (3 × 100 mL). The combined organics were washed once with brine (30 mL) and dried over anhydrous $MgSO_4$. The crude mixture contained a 2:1 ratio (**81**:**82**) of diastereomers. Purification was done by flash chromatography on silica gel eluting with hexanes/ethyl acetate (19:1) to give 0.527 g (63%) of the desired isomer **81** as a white foam: $[\alpha]_D = -11.2$ ($c = 1.4$, $CHCl_3$); IR (film) 3493, 2929, 1710, 1429, 1153, 1115, 1045 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 7.35 (dd, $J = 1.2$, 7.6 Hz, 6 H), 7.38 (m, 9 H), 5.50 (m, 1 H), 4.86 (d, $J = 9.5$ Hz, 1 H), 4.73 (d, $J = 17.1$ Hz, 1 H), 4.09 (d, $J = 3.5$ Hz, 1 H), 3.93 (br d, $J = 10.0$ Hz, 1 H), 3.75 (d, $J = 7.1$ Hz, 1 H), 3.34 (d, $J = 2.6$ Hz, 1 H), 2.29 (dd, $J = 2.5$, 16.4 Hz, 1 H), 2.18 (m, 2 H), 1.67 (m, 2 H), 1.44 (m, 1 H), 1.41 (s, 9 H), 1.05 (d, $J = 7.4$ Hz, 3 H), 0.95 (s, 3 H), 0.86 (s, 12 H), 0.64 (d, $J = 6.7$ Hz, 3 H), −0.03 (s, 3 H), −0.05 (s, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 173.2, 138.0, 136.7, 134.8, 129.8, 127.6, 115.0, 81.2, 78.8, 75.9, 71.3, 44.4, 43.5, 38.0, 37.2, 34.9, 28.0, 26.3, 21.2, 20.5, 18.8, 16.0, 14.1, −3.0, −4.1; HRMS calcd for $C_{43}H_{64}O_5Si_2$: 572.4293; found: 573.4390 (M + H).

Preparation of Compound 83. The alcohol **81** (0.110 g, 0.0153 mmol) was treated with pyridine buffered HF·pyridine solution (3.0 mL) (stock solution was prepared from 20 mL of THF, 11.4 mL of pyridine, and 4.2 g of hydrogen fluoride−pyridne (Aldrich Co.)) at rt and stirred for 2 h. The reaction mixure was poured into saturated aqueous $NaHCO_3$ (50 mL) and extracted with ether (3 × 50 mL). The organic layer was washed in sequence with saturated aqueous $CuSO_4$ (3 × 10 mL) and saturated aqueous $NaHCO_3$ (10 mL) and then dried over Na_2SO_4 and concentrated in vacuo. The residue was purified by

flash chromatography on silica gel eluting with hexanes/ethyl acetate (7:1) to give the corresponding diol (0.070 g, 98%) as a white foam.

The diol (0.231 g, 0.48 mmol) was dissolved in CH_2Cl_2 (5.0 mL) and cooled to −30 °C. 2,6-Lutidine (0.168 mL, 1.44 mmol) was added followed by TBSOTf (0.131 mL, 0.570 mmol). After 1 h at −30 °C, the reaction was poured into Et_2O (300 mL), washed once with 1 N HCl (50 mL), once with saturated aqueous $NaHCO_3$ (50 mL), and once with brine (30 mL), and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/ethyl acetate (20:1) gave alcohol **83** (0.276 g, 96%) as a clear oil: $[\alpha]_D = -5.4$ (c = 0.9, $CHCl_3$); IR (film) 3466, 2930, 1728, 1462, 1369, 1254, 1156 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 5.76 (m, 1 H), 4.99 (m, 2 H), 4.15 (t, J = 4.4 Hz, 1 H), 3.91 (d, J = 4.0 Hz, 1 H), 3.60 (bs, 1 H), 3.75 (dd, J = 3.3, 7.8 Hz, 1 H), 2.81 (dd, J = 4.7, 17.3 Hz, 1 H), 2.41 (m, 1 H), 2.22 (dd, J = 4.2, 17.3 Hz, 1 H), 1.88 (m, 1 H), 1.73 (m, 2 H), 1.45 (s, 9 H), 0.94 (d, J = 7.0 Hz, 3 H), 0.92 (s, 3 H), 0.90 (s, 9 H), 0.89 (s, 12 H), 0.85 (s, 3 H), 0.08 (s, 3 H), 0.07 (s, 3 H), 0.06 (s, 3 H), 0.05 (s, 3H, s); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 172.1, 138.2, 115.5, 80.8, 77.7, 74.4, 46.3, 43.7, 40.9, 38.7, 37.9, 39.7, 28.1, 26.2, 26.0, 19.5, 19.2, 18.5, 18.1, 16.9, 15.1, −3.9, −4.1, −4.2, −4.9; HRMS calcd for $C_{31}H_{64}O_5Si_2$: 572.4293; found: 573.4390 (M + H).

Preparation of Compound 85. The alcohol **83** (0.275 g, 0.46 mmol) was dissolved in CH_2Cl_2 (5.0 mL), and Dess−Martin periodinane (0.292 g, 0.690 mmol) was added. After 2 h, a 1:1 mixture of saturated aqueous $NaHCO_3$/saturated aqueous $Na_2S_2O_3$ (2.0 mL) was added. After 10 min, the mixture was poured into Et_2O (40 mL), and the organic layer was washed with brine (3.0 mL) and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/ethyl acetate (19:1) gave ketone **84** (0.244 g, 89%) as a clear oil.

The olefin **84** (0.420 g, 0.76 mmol) was dissolved in CH_2Cl_2 (10 mL) and treated successively with 2,6-lutidine (1.75 mL, 15 mmol) and TBSOTf (1.72 mL, 7.5 mmol). After 7 h, the reaction was poured into Et_2O (150 mL), washed successively with 0.2 N HCl (25 mL) and brine (20 mL), and dried over anhydrous $MgSO_4$. The residue was purified by flash chromatography on a short pad of silica gel with fast elution with hexanes/ethyl acetate (20:1) to give TBS ester **85** (0.611 g, 93%) as a clear oil. The purification must be done quickly to avoid hydrolysis of the silyl ester: $[\alpha]_D = -35.4$ (c = 0.4, $CHCl_3$); IR (film) 2930, 1730, 1692, 1472, 1367, 1253, 1155, 1084 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 5.73 (m, 1 H), 4.97 (m, 2 H), 4.33 (dd, J = 3.8, 5.5 Hz, 1 H), 3.78 (dd, J = 1.9, 7.3 Hz, 1 H), 3.19 (m, 1 H), 2.53 (dd, J = 3.8, 17.3 Hz, 1 H), 2.25 (m, 2 H), 1.85 (m, 1 H), 1.40 (m, 1 H), 1.24 (s, 3 H), 1.07 (s, 3 H), 1.04 (d, J = 6.8 Hz, 3 H), 0.92 (s, 12 H), 0.91 (s, 9 H), 0.87 (s, 9 H), 0.26 (s, 3 H), 0.26 (s, 3 H), 0.10 (s, 3 H), 0.06 (s, 6 H), 0.05 (s, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 217.1, 170.6, 137.2, 115.0, 79.8, 77.1, 73.2, 52.9, 45.0, 40.8, 37.5, 34.7, 27.5, 25.6, 25.4, 22.4, 19.7, 17.9, 17.6, 17.3, 14.9, −4.2, −4.3, −4.9, −5.4.

Preparation of Compound 87. To a solution of olefin **85** (0.053 g, 0.081 mmol) in THF (0.8 mL) was added 9-BBN (0.5 M soln in THF, 0.323 mL, 0.162 mmol). In a separate flask, the iodide **75** (0.036 g, 0.097 mmol) was dissolved in DMF (1.0 mL). Cs_2CO_3 (0.053 g, 0.162 mmol) was then added with vigorous stirring followed by sequential addition of Ph_3As (0.0025 g, 0.0081 mmol), $PdCl_2(dppf)_2$ (0.0067 g, 0.0081 mmol), and H_2O (0.052 mL, 2.91 mmol). After 4 h, the borane in THF was added to the iodide mixture in DMF. The reaction quickly turned dark brown in color and slowly became pale yellow after 2 h. The reaction was then poured into saturated aqueous NH_4Cl (10.0 mL) and extracted with $CHCl_3$ (3 × 30 mL). The combined organics were washed with H_2O (2 × 50 mL) and once with brine (50 mL) and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/ethyl acetate (4:1 → 3:1) gave 0.036 g (56%) of the coupled product **87** as a pale yellow oil: $[\alpha]_D = -29.2$ (c = 0.3, $CHCl_3$); IR (film) 3500−2600, 1738, 1710, 1691, 1236, 988 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 6.95 (s, 1 H), 6.59 (s, 1 H), 5.49 (m, 1 H), 5.31 (m, 1 H), 5.25 (dd, J = 5.7, 7.7 Hz, 1 H), 4.38 (dd, J = 3.8, 6.5 Hz, 1 H), 3.78 (dd, J = 1.9, 6.7 Hz, 1 H), 3.13 (m, 1 H), 2.71 (s, 3 H), 2.49 (m, 2 H), 2.42 (m, 1 H), 2.31 (dd, J = 6.5, 16.3 Hz, 1 H), 2.07 (s, 3 H), 2.04 (s, 3 H), 1.95 (m, 1 H), 1.89 (m, 2 H), 1.48 (m, 3 H), 1.21 (s, 3 H), 1.15 (m, 2 H), 1.12 (s, 3 H), 1.06 (d, J = 6.9 Hz, 3 H), 0.90 (s, 12 H), 0.88 (s, 9 H), 0.10 (s, 3 H), 0.07 (s, 3 H), 0.06 (s, 3 H), 0.05 (s, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 217.9, 176.7, 170.2, 164.9, 152.2, 137.6, 132.7, 123.9, 120.3, 115.9, 78.4, 73.6, 53.5, 45.0, 40.1, 38.8, 31.0, 30.6, 27.9, 27.7, 26.2, 26.0, 23.5, 21.9, 21.2, 19.2, 18.9, 18.4, 18.1, 17.5, 15.7, 14.9, −3.7, −3.9, −4.3, −4.7; HRMS calcd for $C_{40}H_{71}O_6NSSi_2$: 765.4490; found: 766.4571 (M + H).

Preparation of Compound 88. The acetate **87** (0.035 g, 0.044 mmol) was dissolved in MeOH/H_2O (2:1, 1.5 mL), and K_2CO_3 (0.050 g) was added. After 3 h, the reaction was diluted with saturated aqueous NH_4Cl (5.0 mL) and extracted with $CHCl_3$ (5 × 10 mL). The hydroxy acid **88** was purified by flash chromatography on silica gel eluting with hexanes/ethyl acetate (4:1 → 2:1) to give the pure hydroxy acid **88** (0.030 g, 84%): $[\alpha]_D = -19.8$ (c = 16.5, $CHCl_3$); IR (film) 3600−2450, 1710, 1700, 1472, 1253, 988 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 6.95 (s, 1 H), 6.64 (s, 1 H), 5.55 (m, 1 H), 5.42 (m, 1 H), 4.39 (dd, J = 3.6, 6.4 Hz, 1 H), 4.15 (t, J = 6.7 Hz, 1 H), 3.79 (dd, J = 1.7, 6.9 Hz, 1 H), 3.13 (m, 1 H), 2.71 (s, 3 H), 2.49 (dd, J = 3.6, 16.4 Hz, 1 H), 2.40 (m, 2 H), 2.31 (dd, J = 6.5, 16.4 Hz, 1 H), 2.11 (m, 1 H), 2.01 (s, 3 H), 1.38 (m, 3 H), 1.20 (s, 3 H), 1.15 (m, 2 H), 1.13 (m, 3 H), 1.12 (s, 3 H), 1.05 (d, J = 6.8 Hz, 3 H), 0.91 (d, J = 6.7 Hz, 3 H), 0.87 (s, 9 H), 0.10 (s, 3 H), 0.06 (s, 6 H), 0.05 (s, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 218.0, 176.2, 165.0, 152.5, 141.8, 133.2, 124.9, 118.9, 115.2, 73.5, 53.6, 44.9, 40.1, 38.9, 33.3, 30.8, 28.0, 27.9, 26.2, 26.0, 23.5, 19.2, 18.9, 18.5, 18.2, 17.4, 15.9, 14.5, −3.7, −3.8, −4.2, −4.6; HRMS calcd for $C_{38}H_{69}O_6NSSi_2$: 723.4384; found: 746.4285 (M + Na).

Preparation of Compound 89. To a solution of the the olefin **68** (0.680 g, 1.07 mmol) in THF (8.0 mL) was added 9-BBN (0.5 M soln in THF, 2.99 mL, 1.50 mmol). In a separate flask, the iodide **75** (0.478 g, 1.284 mmol) was dissolved in DMF (10.0 mL). Cs_2CO_3 (0.696 g, 2.14 mmol) was then added with vigorous stirring followed by sequential addition of Ph_3As (0.034 g, 0.111 mmol), $PdCl_2(dppf)_2$ (0.091 g, 0.111 mmol), and H_2O (0.693 mL, 38.5 mmol). After 4 h, the borane solution was added to the iodide mixture in DMF. The mixture quickly turned dark brown in color and slowly became pale yellow after 2 h. The reaction was then poured into H_2O (100 mL) and extracted with Et_2O (3 × 50 mL). The combined organics were washed with H_2O (2 × 50 mL) and once with brine (50 mL) and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/EtOAc (7:1) gave 0.630 g (75%) of the coupled product **86** as a pale yellow oil. This compound could not be separated completely from residual borane impurities and was taken forward in impure form.

The acetate **86** (0.610 g, 0.770 mmol) was dissolved in dioxane/H_2O (9:1, 15 mL), and pTSA·H_2O (0.442 g, 2.32 mmol) was added. The mixture was then heated to 55 °C. After 3 h, the mixture was cooled to rt and poured into Et_2O. This solution was washed once with saturated $NaHCO_3$ (30 mL) and once with brine (30 mL) and dried over anhydrous $MgSO_4$. Purification by flash chromatography on silica gel eluting with hexanes/EtOAc (7:1) gave 0.486 g (85%) of the aldehyde **89** as a pale yellow oil: $[\alpha]_D = -18.7$ (c = 0.53, $CHCl_3$); IR (film) 1737, 1429, 1237, 1115, 1053 cm^{-1}; ^1H NMR ($CDCl_3$, 500 MHz) δ 9.74 (s, 1 H), 7.61 (dd, J = 7.8, 1.2 Hz, 6 H), 7.38 (m, 9 H), 6.94 (s, 1 H), 6.53 (s, 1 H), 5.39 (m, 1 H), 5.31 (m, 1 H), 5.29 (t, J = 6.9 Hz, 1 H), 4.61 (d, J = 4.3 Hz, 1 H), 3.50 (dd, J = 5.2, 2.6 Hz, 1 H), 2.70 (s, 3 H), 2.48 (m, 2 H), 2.14 (m, 1 H), 2.09 (s, 3 H), 2.07 (s, 3 H), 1.83 (m, 2 H), 1.41 (m, 1 H), 1.18 (m, 1 H), 1.01 (s, 3 H), 0.99 (s, 3 H), 0.91 (d, J = 7.4 Hz, 3 H), 0.85 (s, 9 H), 0.69 (m, 1 H), 0.58 (d, J = 6.8 Hz, 3 H), −0.05 (s, 3 H), −0.06 (s, 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 205.46, 170.01, 164.49, 152.46, 137.10, 135.60, 134.22, 132.55, 130.65, 127.84, 123.82, 120.66, 116.19, 81.09, 78.47, 76.73, 51.66, 43.14, 38.98, 30.99, 30.42, 27.63, 26.10, 21.15, 20.92, 20.05, 19.15, 18.49, 15.12, 14.70, 12.75, −3.25, −4.08; HRMS calcd for $C_{50}H_{69}O_5NSSi_2$: 851.4435; found: 890.4100 (M + K).

Preparation of Compounds 90 and 91. To a solution of the acetate−aldehyde **89** (0.084 g, 0.099 mmol) in THF (99 mL) at −78 °C was added potassium bis(trimethylsilyl)amide (0.5 M in toluene, 1.0 mL, 0.5 mmol) dropwise. The resulting solution was stirred at −78 °C for 30 min. Then the reaction mixure was transfered via cannula to a short pad of silica gel and washed with Et_2O. The residue was purified by flash chromatography (silica, 12% EtOAc in hexane) to give the 3*S* product **90** and the 3*R* product **91** in a 6:1 ratio in 51% combined yield:

Compound 90: $[\alpha]_D = -39.4$ (c 0.52, $CHCl_3$); IR (film) 3508, 1733, 1428, 1254, 1113, 1034 cm^{-1}; ^1H NMR (500 MHz, C_6D_6, 60 °C) δ 7.85 (dd, J = 7.3, 2.1 Hz, 6 H), 7.22 (m, 9 H), 6.54 (s, 1 H), 6.49 (s, 1 H), 5.53 (d, J = 6.0 Hz, 1 H), 5.42 (m, 2 H), 4.22 (d, J =

5.9 Hz, 1 H), 4.19 (d, J = 3.5 Hz, 1 H), 4.17 (br s, 1 H), 2.58 (m, 1 H), 2.45 (m, 2 H), 2.31 (s, 3 H), 2.29 (m, 2 H), 2.13 (s, 3 H), 2.00 (m, 2 H), 1.81 (m, 1 H), 1.56 (m, 1 H), 1.35−1.28 (band, 2 H), 1.24 (d, J = 7.1 Hz, 3 H), 1.20 (m, 1 H), 1.11 (s, 3 H), 1.07−0.92 (band, 13 H), 0.88 (d, J = 6.7 Hz, 3 H), 0.12 (s, 6 H); ^{13}C NMR (125 MHz, C_6D_6, 60 °C) δ 171.1, 164.3, 153.7, 137.4, 136.6, 136.0, 130.1, 128.5, 125.5, 120.2, 116.7, 78.5, 75.8, 72.8, 44.6, 41.6, 38.3, 32.7, 31.8, 30.1, 28.5, 27.9, 26.6, 22.2, 21.3, 18.9, 18.8, 15.8, 15.7, 1.30, −2.76, −3.66; HRMS calcd for $C_{50}H_{69}O_5NSSi_2$: 851.4435; found: 852.4513 (M + H).

Compound 91: $[\alpha]_D$ = −53.9 (c 0.37, $CHCl_3$); IR (film) 2927, 1734, 1428, 1114, 1036 cm^{-1}; ^{1}H NMR (500 MHz, C_6D_6, 60 °C) δ 7.82 (m, 6 H), 7.22−7.19 (band, 9 H), 6.59 (s, 1 H), 6.53 (s, 1 H), 5.58−5.53 (band, 2 H), 5.49 (m, 1 H), 4.39 (d, J = 9.7 Hz, 1 H), 3.98 (d, J = 4.7 Hz, 1 H), 3.86 (dd, J = 1.9, 5.6 Hz, 1 H), 3.59 (br s, 1 H), 2.49−2.43 (band, 3 H), 2.34−2.26 (band, 6 H), 2.18 (s, 3 H), 1.98 (m, 2 H), 1.51 (m, 1 H), 1.50−1.30 (band, 2 H), 1.21−1.19 (band, 6 H), 1.03−0.99 (band, 10H), 0.85 (d, J = 5.4 Hz, 3 H), 0.78 (s, 3 H), 0.10 (s, 3 H), 0.07 (s, 3 H); HRMS calcd for $C_{50}H_{69}O_5NSSi_2$: 851.4435; found: 852.4489 (M + H).

Preparation of Compound 93. The lactone **90** (0.032 g, 0.0376 mmol) was treated with pyridine-buffered HF·pyridine solution (1 mL) (stock solution was prepared from 20 mL of THF, 11.4 mL of pyridine, and 4.2 g of hydrogen fluoride−pyridne (Aldrich Co.)) at room temperature for 2 h. The reaction mixure was poured into saturated aqueous $NaHCO_3$ (15 mL) and extracted with Et_2O (3 × 30 mL). The organic layer was washed in sequence with saturated aqueous $CuSO_4$ (3 × 10 mL) and saturated aqueous $NaHCO_3$ (10 mL) and then dried over Na_2SO_4 and concentrated in vacuo. The residue was purified by flash chromatography (silica, 25% EtOAc in hexane) to give diol **93** (0.022 g, 99%) as a white foam: $[\alpha]_D$ = −111.7 (c = 0.7, $CHCl_3$); IR (film) 3463, 2928, 1729, 1253 cm^{-1}; ^{1}H NMR (C_6D_6, 500 MHz, 60 °C) δ 6.65 (s, 1 H), 6.57 (s, 1 H), 5.50 (dd, J = 2.58, 9.42 Hz, 1 H), 5.39 (m, 2 H), 4.34 (s, 1 H), 4.07 (dd, J = 2.2, 9.9 Hz, 1 H), 3.47 (t, J = 4.5 Hz, 1 H), 3.11 (br s, 1 H), 2.69 (m, 2 H), 2.57 (m, 2 H), 2.31 (s, 3 H), 2.18 (m, 6 H), 2.0 (m, 1 H), 1.78 (m, 1 H), 1.53 (m, 2 H), 1.27 (m, 1 H), 1.09 (d, J = 4.5 Hz, 3 H), 0.98 (m, 15H), 0.91 (s, 3 H), 0.14 (s, 3 H), 0.13 (s, 3 H); ^{13}C NMR (C_6D_6, 125 MHz, 60 °C) δ 171.4, 164.3, 153.8, 137.8, 133.2, 128.5, 125.8, 120.3, 116.6, 83.2, 78.7, 75.8, 73.9, 42.6, 40.9, 39.4, 35.5, 34.4, 32.3, 28.1, 26.3, 22.5, 22.4, 18.8, 18.6, 17.0, 16.5, 15.5, −3.6, −4.4; LRMS calcd for $C_{32}H_{55}O_5NSSi$: 593.4; found: 616.3 (M + Na).

Preparation of Compound 94. To a cooled (−30 °C) solution of diol **93** (0.029 g, 0.048 mmol) and 2,6-lutidine (0.017 mL, 0.147 mmol) in anhydrous CH_2Cl_2 (1.0 mL) was added TBSOTf (0.015 mL, 0.065 mmol). The resulting solution was then stirred at −30 °C for 30 min. The reaction was quenched with 0.5 M HCl (10 mL) and extracted with Et_2O (15 mL). The ether layer was washed with saturated aqueous $NaHCO_3$ (5 mL), dried (Na_2SO_4) and concentrated in vacuo. Purification of the residue by flash chromatogrphy (silica, 8% EtOAc in hexane) afforded TBS ether **94** (0.032 g, 93%) as white foam: $[\alpha]_D$ = −21.7 (c 0.35, $CHCl_3$); IR (film) 3471, 2928, 1742, 1253, 1076 cm^{-1}; ^{1}H NMR (500 MHz, C_6D_6, 38 °C) δ 6.62 (s, 1 H), 6.53 (s, 1 H) 5.49−5.46 (band, 3 H), 4.41 (br s, 1 H), 4.10 (br s, 1 H), 3.49 (br s, 1 H), 2.70−2.64 (band, 2 H), 2.44 (dd, J = 16.1, 6.5 Hz, 1 H), 2.34 (d, J = 15.5 Hz, 1 H), 2.28 (s, 3 H), 2.22−2.15 (band, 5H), 2.02 (m, 1 H), 1.81 (m, 1 H), 1.68 (m, 1 H), 1.50 (m, 1 H), 1.34 (m, 1 H), 1.18 (s, 3 H), 1.14 (d, J = 20.9 Hz, 3 H), 1.02−0.98 (band, 23 H), 0.90 (s, 3 H), 0.16−0.15 (band, 9 H), 0.10 (s, 3 H); ^{13}C NMR (125 MHz, C_6D_6, 43 °C) δ 171.3, 164.2, 153.9, 137.9, 133.0, 127.9, 120.1, 116.6, 78.9, 75.1, 44.7, 41.0, 32.8, 31.9, 27.9, 27.6, 26.4, 26.2, 25.9, 21.7, 18.9, 18.51, 18.49, 17.4, 15.53, −3.4, −3.6, −4.3, −4.4; HRMS calcd for $C_{38}H_{69}O_5NSSi_2$: 707.4435; found: 746.4062 (M + K).

Preparation of Compound 95. To a solution of alcohol **94** (0.030 g, 0.0424 mmol) in CH_2Cl_2 (2.0 mL) at 25 °C was added Dess−Martin periodinane (0.036 g, 0.0848 mmol) in one portion. The resulting solution was then allowed to stir at 25 °C for 1.5 h. The reaction was quenched by the addition of 1:1 saturated aqueous $NaHCO_3$:$Na_2S_2O_3$ (10 mL) and stirred for 5 min. The mixture was then extracted with Et_2O (3 × 15 mL). The organic layer was dried (Na_2SO_4), filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (silica, 8% EtOAc in hexane) provided ketone **95** (0.025 g, 84%) as white foam: $[\alpha]_D$ = −21.93 (c = 1.4, $CHCl_3$); IR (film): 2928, 1745, 1692, 1254, 1175, 836 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.97 (s, 1 H), 6.57 (s, 1 H), 5.53 (dt, J = 3.4, 11.1 Hz, 1 H), 5.37 (dd, J = 16.4, 9.9 Hz, 1 H), 5.00 (d, J = 10.3 Hz, 1 H), 4.02 (d, J = 9.7 Hz, 1 H), 3.89 (d, J = 8.7 Hz, 1 H), 3.00 (m, 1 H), 2.82 (d, J = 6.5 Hz, 1 H), 2.71 (m, 5H), 2.36 (q, J = 10.7 Hz, 1 H), 2.12 (, 3 H), 2.07 (dd, J = 8.2 Hz, 1 H), 1.87 (bs, 1 H), 1.49 (m, 3 H), 1.19 (m, 5H), 1.14 (s, 3 H), 1.08 (d, J = 6.8 Hz, 3 H), 0.94 (m, 12 H), 0.84 (s, 9 H), 0.12 (s, 3 H), 0.10 (s, 3 H), 0.07 (s, 3 H), −0.09 (s, 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 218.7, 170.1, 164.5, 152.6, 137.9, 133.9, 124.8, 119.6, 115.9, 72.7, 53.2, 43.9, 41.0, 40.3, 32.9, 32.3, 28.4, 27.1, 26.3, 26.1, 26.0, 19.2, 19.1, 18.3, 18.2, 17.1, 16.0, 15.2, 14.3, −4.2, −4.4, −4.6, −4.8; HRMS calcd for $C_{38}H_{67}O_5NSSi_2$: 705.4315, found: 706.4357 (M + H).

Macrolactonization To Produce Compound 95. To a solution of hydroxy acid **88** (0.094 g, 0.133 mmol) in THF (1 mL) were added Et_3N (0.11 mL, 0.79 mmol) and 2,4,6-trichlorobenzoyl chloride (0.104 mL, 0.66 mmol). The mixture was stirred at rt for 0.25 h, diluted with toluene (15 mL), and added dropwise over a period of 3 h to a solution of DMAP (0.167 mg, 1.37 mmol) in toluene (50 mL). After complete addition, the mixture was stirred for additional 0.5 h and concentrated in vacuo. Purification of the residue by flash chromatography on silica gel eluting with hexanes/ethyl acetate (9:1) gave 0.081 g (88%) of the previously described lactone **95**.

Preparation of Compound 96. To a solution of TBS ether **95** (0.027 g, 0.038 mmol) in THF (1.0 mL) at 25 °C in a plastic vial was added dropwise HF·pyridine (0.5 mL). The resulting solution was allowed to stir at 25 °C for 2 h. The reaction mixture was diluted with chloroform (2 mL) and very slowly added to saturated aqueous $NaHCO_3$ (20 mL). The mixture was extracted with $CHCl_3$ (20 mL × 3). The organic layer was dried (Na_2SO_4), filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (silica, 30% EtOAc in hexane) provided diol **96** (0.018 g, 99%) as white foam: $[\alpha]_D$ = −84.7 (c = 0.85, $CHCl_3$); IR (film): 3493, 2925, 1728, 1689, 1249 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.96 (s, 1 H), 6.59 (s, 1 H), 5.44 (dt, J = 4.3, 10.4 Hz, 1 H), 5.36 (dt, J = 5.1, 10.2 Hz, 1 H), 5.28 (dd, J = 1.7, 9.8 Hz, 1 H), 4.11 (d, J = 7.2 Hz, 1 H), 3.74 (s, 1 H), 3.20 (d, J = 4.5 Hz, 1 H), 3.14 (dd, J = 2.2, 6.8 Hz, 1 H), 3.00 (s, 1 H), 2.69 (m, 4 H), 2.49 (dd, J = 11.3, 15.1 Hz, 1 H), 2.35 (dd, J = 2.5, 15.1 Hz, 1 H), 2.27 (m, 1 H), 2.05 (m, 1 H), 2.04 (s, 3 H), 2.01 (m, 1 H) 1.75 (m, 1 H), 1.67 (m, 1 H), 1.33 (m, 4 H), 1.21 (s, 1 H), 1.19 (m, 2 H), 1.08 (d, J = 7.0 Hz, 3 H), 1.00 (s, 3 H), 0.93 (d, J = 7.1 Hz, 3 H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 226.5, 176.5, 171.1, 158.2, 144.7, 139.6, 131.1, 125.7, 122.0, 84.6, 80.2, 78.6, 59.4, 47.9, 45.4, 44.6, 38.5, 37.9, 33.7, 33.6, 28.7, 25.1, 25.0, 21.9, 21.7, 19.6; HRMS calcd for $C_{26}H_{39}O_5NS$ 477.2549, found 478.2627 (M + H).

Preparation of Epothilone A (2). To a cooled (−50 °C) solution of desoxyepothilone A (**96**) (0.009 g, 0.0189 mmol) in dry CH_2Cl_2 (1 mL) was added freshly prepared 3,3-dimethyldioxirane (0.95 mL, 0.1 M in acetone). The resulting solution was allowed to warm to −30 °C for 2 h. A stream of nitrogen was then bubbled through the solution to remove excess dimethyldioxirane. The residue was purified by flash chromatography (silica, 40% EtOAc in hexane) and afforded epothilone A (**2**) (0.0046 g, 49%) as colorless solid and 0.0003 g of the *cis*-epoxide diastereomer: $[\alpha]_D$ = −41.6 (c = 0.51, MeOH); IR (film): 3464, 2926, 1737, 1689, 978, 755 cm^{-1}; ^{1}H NMR (CD_2Cl_2, 500 MHz) δ 7.00 (s, 1 H), 6.56 (s, 1 H), 5.39 (dd, J = 9.2, 2.0 Hz, 1 H), 4.16 (br d, J = 10.0 Hz, 1 H), 3.73 (dd, J = 8.6, 4.2 Hz, 1 H), 3.59 (br s, 1 H), 3.21 (m, 1 H), 2.99 (m, 1 H), 2.87 (m, 1 H), 2.68 (s, 3 H), 2.50−2.44 (band, 2 H), 2.38 (dd, J = 15.0, 3.2 Hz, 1 H), 2.14−2.08 (band, 4 H), 1.86 (m, 1 H), 1.75−1.66 (band, 3 H), 1.55 (m, 1 H), 1.41 (m, 4 H), 1.35 (s, 3 H), 1.14 (d, J = 6.9 Hz, 3 H), 1.06 (s, 3 H), 0.98 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (CD_2Cl_2, 125 MHz) δ 220.2, 170.9, 165.6, 152.4, 138.1, 120.2, 116.7, 77.2, 74.9, 73.4, 57.8, 55.1, 53.7, 39.5, 36.8, 32.1, 30.9, 30.1, 27.7, 23.8, 22.0, 20.2, 19.3, 17.3, 15.6, 14.3; HRMS calcd for $C_{26}H_{39}O_6NS$: 493.2498, found: 494.2578 (M + H).

Preparation of Compound 97. To a suspension of ethyltriphenylphosphonium iodide (0.250 g, 0.60 mmol) in THF (6 mL) was added *n*BuLi (2.5 M soln in hexanes, 0.24 mL, 0.60 mmol) at rt. After disappearance of the solid material, the solution was added to a mixture of iodine (0.152 g, 0.60 mmol) in THF (4 mL) at −78 °C. The resulting suspension was vigorously stirred for 5 min at −78 °C and then warmed to −20 °C and treated with sodium bis(trimethylsilyl)amide (1 M soln in THF, 0.56 mL, 0.56 mmol). The resulting red solution was stirred for 5 min followed by the slow addition of aldehyde **78** (0.074 g, 0.30

mmol). The mixture was stirred at −20 °C for 40 min, diluted with pentane (50 mL), filtered through a pad of Celite, and concentrated in vacuo. Purification of the residue by flash column chromatography (hexanes/ethyl acetate, 85:15) gave 0.141 g (43% overall from acetate **77**) of the vinyl iodide **97** as a yellow oil: $[\alpha]_D = -20.7$ (*c* 2.45, $CHCl_3$); IR (film) 2920, 1738, 1369, 1234 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 6.98 (s, 1 H), 6.53 (s, 1 H), 5.43 (t, J = 6.5, 5.4 Hz, 1 H), 5.35 (t, J = 6.6, 6.5 Hz, 1 H), 2.71 (s, 3 H), 2.58−2.50 (band, 5H), 2.08 (s, 6 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.0, 164.6, 152.4, 136.9, 130.3, 120.7, 120.6, 116.4, 103.6, 40.3, 33.7, 19.2, 19.1, 14.9; HRMS calcd for $C_{14}H_{18}O_2NIS$: 391.0103; found: 392.0181 (M + H).

Preparation of Compound 98. To a solution of olefin **68** (0.977 g, 1.55 mmol) in THF (3 mL) was added 9-BBN (0.5 M soln in THF, 3.4 mL, 1.7 mmol). In a separate flask, iodide **97** (0.749 g, 1.92 mmol) was dissolved in DMF (5 mL). Cs_2CO_3 (1.154 g, 3.54 mmol) was then added with vigorous stirring followed by sequential addition of $PdCl_2(dppf)_2$ (0.162 g, 0.198 mmol), Ph_3As (0.061 g, 0.20 mmol), and H_2O (0.42 mL, 23.4 mmol). After 5 h, the borane solution was added to the iodide mixture in DMF. The reaction quickly turned dark brown in color and slowly became pale yellow after 3 h. The solution was then poured into H_2O (10 mL) and extracted with Et_2O (3 × 15 mL). The combined organic layers were washed with H_2O (3 × 15 mL), brine (1 × 20 mL), dried over $MgSO_4$, filtered, and concentrated in vacuo. Flash column chromatography (hexanes/ethyl acetate, 9:1) gave the coupled product **98** (1.073 g; 77%) as a yellow oil: IR (film) 2931, 1738, 1429, 1239, 1072, 709 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 7.41 (m, 15 H), 6.98 (s, 1 H), 6.59 (s, 1 H), 5.30 (t, J = 6.9 Hz, 1 H), 5.15 (t, J = 6.7 Hz, 1 H), 4.38 (d, J = 3.5 Hz, 1 H), 4.02 (s, 1 H), 3.60 (m, 2 H), 3.41 (t, J = 6.9 Hz, 2 H), 2.88 (s, 3 H), 2.79 (s, 3 H), 2.41 (m, 2 H), 2.16−2.10 (band, 6 H), 1.80 (m, 2 H), 1.70 (s, 3 H), 1.30−1.01 (band, 6 H), 0.85 (s, 15H), 0.73 (d, J = 6.8 Hz, 3 H), 0.62 (d, J = 6.7 Hz, 3 H), 0.00 (br s, 6 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.2, 164.5, 152.7, 138.6, 137.5, 135.8, 135.3, 129.9, 127.8, 120.6, 119.1, 116.2, 110.6, 110.5, 78.9, 77.8, 63.3, 60.4, 60.2, 55.5, 46.6, 43.9, 43.4, 38.0, 32.3, 31.7, 30.4, 26.2, 26.1, 26.0, 23.6, 21.3, 19.6, 19.4, 19.2, 18.6, 17.1, 15.8, 15.4, 14.8, 14.2, 13.6, −2.9, −3.9, −4.1; HRMS calcd for $C_{53}H_{77}O_6NSSi_2$ 911.5010, found 950.4613 (M + K).

Preparation of Compound 99. The acetal **98** (0.069 g, 0.077 mmol) was dissolved in dioxane/H_2O (9:1, 1 mL), and pTSA·H_2O (0.045 g, 0.237 mmol) was added. The mixture was then heated to 55 °C. After 3 h, the mixture was cooled to rt, poured into saturated aqueous $NaHCO_3$, and extracted with Et_2O (4 × 15 mL). The combined ether extracts were washed with saturated aqueous $NaHCO_3$ (1 × 30 mL), brine (1 × 30 mL), dried over $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/EtOAc, 3:1) gave aldehyde **99** (0.046 g, 71%) as a pale yellow oil: $[\alpha]_D = -13.3$ (*c* 0.95, $CHCl_3$); IR (film) 3070, 2929, 2856, 1737, 1429, 1238, 1116, 1056 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 9.79 (s, 1 H), 7.71−7.69 (band, 6 H), 7.52−7.44 (band, 9 H), 7.01 (s, 1 H), 6.60 (s, 1 H), 5.31 (t, J = 6.6 Hz, 1 H), 5.12 (t, J = 6.9 Hz, 1 H), 4.85 (d, J = 4.5 Hz, 1 H), 3.58 (br s, 1 H), 2.77 (s, 3 H), 2.47 (m, 2 H), 2.14 (s, 3 H), 2.13 (s, 3 H), 1.87 (m, 2 H), 1.72 (s, 3 H), 1.49 (m, 1 H), 1.30 (m, 2 H), 1.10 (m, 1 H), 1.09 (s, 3 H), 1.06 (s, 3 H), 0.98 (d, J = 1.5 Hz, 3 H), 0.92 (s, 9 H), 0.80 (m, 2 H), 0.65 (d, J = 6.7 Hz, 3 H), 0.02 (s, 3 H), 0.00 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 205.7, 170.2, 164.6, 152.6, 138.4, 137.4, 135.7, 135.1, 134.9, 134.3, 130.1, 127.9, 120.6, 119.2, 116.2, 81.2, 78.9, 51.7, 43.1, 39.1, 32.3, 31.7, 30.9, 26.1, 26.0, 23.6, 21.3, 20.8, 20.2, 19.2, 18.6, 14.9, 14.8, 12.8, 0.00, −3.2, −4.0; HRMS calcd for $C_{51}H_{71}O_5NSSi_2$ 865.4592, found 904.4201 (M + K).

Preparation of Compounds 100 and 101. To a solution of aldehyde **99** (0.290 g, 0.341 mmol) in THF (34 mL) at −78 °C was added potassium bis(trimethylsilyl)amide (0.5 M soln in toluene, 3.4 mL, 1.7 mmol). The solution was stirred at −78 °C for 1 h and then quenched with saturated aqueous NH_4Cl and extracted with Et_2O (3 × 15 mL). The combined organic layers were dried with $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/EtOAc, 7:1) gave 0.120 g of the desired α-alcohol **100** and 0.055 g of β-alcohol **101** (60% combined yield) as pale yellow oils.

Conversion of Compound 101 to Compound 100. To a solution of β-alcohol **101** (0.075 g, 0.088 mmol) in 0.5 mL of CH_2Cl_2 at rt was added Dess−Martin periodinane (0.188 g, 0.44 mmol). The resulting solution was stirred at rt for 1 h and then treated with Et_2O (2 mL) and saturated aqueous $NaHCO_3$:$Na_2S_2O_3$ (3 mL, 1:1), poured into H_2O (20 mL), and extracted with Et_2O (4 × 10 mL). The combined ether solutions were washed with H_2O (1 × 30 mL), brine (1 × 30 mL), dried with $MgSO_4$, filtered, and concentrated in vacuo. To a solution of crude ketone **102** in MeOH/THF (2 mL, 1:1) at −78 °C was added $NaBH_4$ (0.017 g, 0.447 mmol). The resulting solution was stirred at rt for 1 h, quenched with saturated aqueous NH_4Cl (10 mL), and extracted with Et_2O (3 × 15 mL). The organic layers were dried with $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/ethyl acetate, 9:1) gave 0.052 g (67%) of the α-alcohol **100** as a pale yellow oil and 0.009 g of β-alcohol **101**.

Compound 100: $[\alpha]_D = -45.4$ (*c* 1.0, $CHCl_3$); IR (film) 3490, 2929, 2856, 1729, 1428, 1114, 1037, 708 cm^{-1}; ^1H NMR (500 MHz, C_6D_6, 60 °C) δ 7.85 (m, 6 H), 7.22 (m, 9 H), 6.55 (s, 1 H), 6.49 (s, 1 H), 5.56 (br d, J = 6.9 Hz, 1 H), 5.25 (m, 1 H), 4.24 (br s, 1 H), 4.17 (m, 2 H), 2.60 (m, 1 H), 2.47 (m, 2 H), 2.29−2.22 (band, 6 H), 2.15 (s, 3 H), 2.08 (m, 2 H), 1.96 (m, 1 H), 1.84 (br s, 1 H), 1.68 (s, 3 H), 1.43 (m, 1 H), 1.32 (m, 2 H), 1.24 (d, J = 7.1 Hz, 3 H), 1.11 (s, 6 H), 1.04 (s, 9 H), 0.91 (d, J = 6.3 Hz, 3 H), 0.12 (s, 6 H); HRMS calcd for $C_{51}H_{71}O_5NSSi_2$: 865.4591; found: 866.4716 (M + H).

Preparation of Compound 103. The silyl ether **100** (0.210 g, 0.247 mmol) was dissolved in HF·pyridine/pyridine/THF (15 mL). The solution was stirred at rt for 2 h and then diluted with Et_2O (1 mL), poured into a mixture of Et_2O/saturated $NaHCO_3$ (20 mL, 1:1), and extracted with Et_2O (4 × 10 mL). The Et_2O extracts were washed with saturated aqueous $CuSO_4$ (3 × 30 mL), saturated aqueous $NaHCO_3$ (1 × 30 mL), and brine (1 × 30 mL), dried with $MgSO_4$, filtered, and concentrated in vacuo. Flash column chromatography (hexanes/EtOAc, 9:1) gave diol **103** (0.141 g, 94%) as a pale yellow oil: $[\alpha]_D = -41.7$ (*c* 0.65, $CHCl_3$); IR (film) 3460, 2928, 1728, 1252, 1037, 757 cm^{-1}; ^1H NMR (500 MHz, C_6D_6, 58 °C) δ 6.69 (s, 1 H), 6.56 (s, 1 H), 5.51 (dd, J = 9.4, 3.0 Hz, 1 H), 5.20 (m, 1 H), 4.30 (br s, 1 H), 4.10 (dd, J = 9.7, 2.6 Hz, 1 H), 3.50 (br s, 1 H), 3.07 (br s, 1 H), 2.74 (t, J = 9.6, 5.7 Hz, 1 H), 2.70 (dd, J = 16.0, 3.5 Hz, 1 H), 2.52 (dd, J = 16.0, 9.7 Hz, 1 H), 2.31−2.25 (band, 4 H), 2.24 (s, 3 H), 2.22−2.18 (band, 3 H), 1.91 (m, 1 H), 1.81 (m, 1 H), 1.61−1.58 (band, 4 H), 1.50 (m, 1 H), 1.37 (m, 2 H), 1.11 (d, J = 6.9 Hz, 3 H), 1.04 (d, J = 7.0 Hz, 3 H), 0.99 (s, 12 H), 0.93 (s, 3 H), 0.16 (s, 3 H), 0.14 (s, 3 H); ^{13}C NMR (125 MHz, C_6D_6, 58 °C) δ 171.3, 164.3, 153.9, 138.2, 137.6, 122.1, 120.4, 116.6, 82.8, 79.2, 73.8, 42.7, 39.5, 34.0, 32.9, 31.8, 26.3, 22.4, 21.9, 18.8, 18.6, 17.0, 15.5, −3.5, −4.5; LRMS calcd for $C_{33}H_{57}O_5NSSi$ 607.4, found 608.4 (M + H).

Preparation of Compound 104. To a solution of diol **103** (0.0066 g, 0.011 mmol) in 0.5 mL of CH_2Cl_2 at −78 °C were added 2,6-lutidine (7 μL, 0.060 mmol) and TBSOTf (5 μL, 0.022 mmol). The resulting solution was stirred at −30 °C for 0.5 h and then quenched with H_2O (5 mL) and extracted with Et_2O (4 × 10 mL). The ether solutions were washed with 0.5 M HCl (1 × 10 mL) and saturated aqueous $NaHCO_3$ (1 × 10 mL), dried over $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/EtOAc, 93:7) gave alcohol **104** (0.0070 g, 89%) as a pale yellow oil: $[\alpha]_D = -2.6$ (*c* 3.15, $CHCl_3$); IR (film) 3453, 2929, 1726, 1252, 1030 cm^{-1}; ^1H NMR (500 MHz, C_6D_6, 60 °C) δ 7.15 (s, 1 H), 6.66 (s, 1 H), 6.58 (s, 1 H), 5.54 (t, J = 6.8, 6.7 Hz, 1 H), 5.21 (m, 1 H), 4.64 (br s, 1 H), 3.80 (br s, 1 H), 2.99 (m, 1 H), 2.84 (dd, J = 14.0, 6.7 Hz, 1 H), 2,45 (dd, J = 14.1, 1.5 Hz, 1 H), 2.37 (s, 3 H), 2.29 (s, 3 H), 2.17−2.08 (band, 4 H), 1.80 (m, 1 H), 1.70−1.60 (band, 4 H), 1.49 (m, 1 H), 1.12−1.08 (band, 5H), 1.04 (d, J = 6.8 Hz, 3 H), 1.00−0.90 (band, 25H), 0.18 (s, 3 H), 0.12 (s, 3 H), 0.11 (s, 3 H), 0.06 (s, 3 H); ^{13}C NMR (125 MHz, C_6D_6) δ 171.2, 164.1, 154.2, 138.8, 121.4, 120.8, 116.6, 80.1, 77.6, 74.7, 44.7, 41.8, 33.5, 32.5, 31.9, 27.1, 26.4, 26.8, 23.3, 20.2, 18.9, 18.4, 18.3, 17.4, 15.1, −3.8, −4.3, −4.4; LRMS calcd for $C_{39}H_{71}NO_5SSi_2$: 721.5; found: 722.7 (M + H).

Preparation of Compound 105. To a solution of alcohol **104** (0.107 g, 0.148 mmol) in CH_2Cl_2 (3 mL) at rt was added Dess−Martin periodinane (0.315 g, 0.743 mmol). The resulting solution was stirred at rt for 2 h, treated with Et_2O (1 mL) and saturated aqueous $Na_2S_2O_3$/saturated aqueous $NaHCO_3$ (2 mL, 1:1), poured into H_2O (20 mL), and extracted with Et_2O (4 × 10 mL). The ether solution was washed with saturated aqueous $NaHCO_3$ (1 × 20 mL), dried with $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/EtOAc, 9:1) gave ketone **105** (0.092 g, 87%) as a pale yellow oil: $[\alpha]_D = -18.7$ (*c* 3.65, $CHCl_3$); IR (film) 2931, 2856, 1742, 1696, 1463, 1255, 835 cm^{-1}; ^1H NMR (500 MHz, $CDCl_3$) δ 6.85 (s, 1 H), 6.46 (s,

1 H), 5.06 (t, J = 8.4, 7.7 Hz, 1 H), 4.86 (d, J = 10.0 Hz, 1 H), 3.92 (d, J = 9.3 Hz, 1 H), 3.78 (d, J = 8.9 Hz, 1 H), 2.92 (m, 2 H), 2.69 (d, J = 15.5 Hz, 1 H), 2.60−2.55 (band, 1 H), 2.36 (m, 1 H), 2.06 (d, J = 3.5 Hz, 1 H), 2.00−1.93 (band, 5H), 1.63−1.58 (band, 6 H), 1.44 (m, 2 H), 1.15 (s, 3 H), 1.08 (s, 3 H), 1.03 (s, 3 H), 0.99 (d, J = 6.8 Hz, 3 H), 0.87 (d, J = 6.9 Hz, 3 H), 0.85−0.73 (band, 12 H), 0.00−0.06 (band, 9 H), −0.21 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 215.1, 171.2, 164.5, 152.5, 140.6, 138.8, 119.3, 119.1, 115.9, 79.8, 76.2, 53.4, 39.1, 32.5, 31.9, 31.4, 29.2, 27.4, 26.3, 26.1, 24.5, 24.3, 23.0, 19.1, 18.7, 18.6, 17.8, 15.3, −3.3, −3.7, −5.6; LRMS calcd for $C_{39}H_{69}O_5NSSi_2$: 719.4; found: 720.6 (M + H).

Preparation of Compound 106. To a solution of ketone **105** (0.092 g, 0.128 mmol) in THF (4.5 mL) at 0 °C was added HF·pyridine (2.25 mL) dropwise. The solution was stirred at rt for 2 h, diluted with $CHCl_3$ (2 mL), poured into saturated aqueous $NaHCO_3/CHCl_3$ (20 mL, 1:1) slowly, and extracted with $CHCl_3$ (4 × 10 mL). The combined $CHCl_3$ layers were dried with $MgSO_4$, filtered, and concentrated. Flash column chromatography (hexanes/EtOAc, 3:1) gave desoxyepothilone B (**106**)-(0.057 g, 92%) as a pale yellow oil: $[\alpha]_D$ = −61.4 (c 2.85, $CHCl_3$); IR (film) 3465, 2969, 1735, 1691, 1377, 1181, 1148 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.95 (s, 1 H), 6.58 (s, 1 H), 5.21 (d, J = 9.3 Hz, 1 H), 5.15 (dd, J = 10.7, 5.5 Hz, 1 H), 4.28 (br d, J = 9.2 Hz, 1 H), 3.80−3.50 (band, 3 H), 3.17 (m, 1 H), 3.16 (br s, 1 H), 2.69 (s, 3 H), 2.66−2.61 (band, 3 H), 2.46 (dd, J = 14.6, 3.4 Hz, 1 H), 2.34−2.22 (band, 3 H), 2.18 (s, 3 H), 2.07 (s, 3 H), 1.88 (m, 1 H), 1.75 (m, 1 H), 1.34 (s, 3 H), 1.02 (d, J = 7.0 Hz, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 220.6, 210.7, 170.4, 164.9, 151.9, 139.1, 138.4, 120.8, 119.3, 115.6, 78.9, 74.1, 72.3, 69.4, 53.7, 53.4, 41.7, 39.6, 38.3, 32.5, 31.7, 29.2, 22.9, 19.0, 18.0, 15.7; HRMS calcd for $C_{27}H_{41}NO_5NS$: 491.2705; found $C_{27}H_{42}NO_5S$: 492.2782 (M + H).

Preparation of Epothilone B (3). To a solution of desoxyepothilone B (**106**) (0.090 g, 0.183 mmol) in CH_2Cl_2 (1.8 mL) at −78 °C was added freshly prepared dimethyldioxirane (0.087 M soln in acetone, 3.60 mL, 0.313 mmol) dropwise. The resulting solution was warmed to −50 °C for 1 h, and another portion of dimethyldioxirane (1.0 mL, 0.087 mmol) was added. After stirring at −50 °C for additional 1.5 h, any excess dimethyldioxirane and solvent were removed by a stream of N_2 at −50 °C. The crude reaction mixture was determined to be >20:1 ratio of diastereomeric *cis*-epoxides by ^{1}H NMR spectroscopy. The resulting residue was purified by flash column chromatography (hexanes/EtOAc, 1:1) to give epothilone B (**3**) (0.090 g, 97%) as a white solid: $[\alpha]_D$ = −31.0 (c 0.045, $CHCl_3$); mp 93.6−94.7 °C; IR (film) 3454, 2962, 1727, 1690, 1263, 978 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.98 (s, 1 H), 6.52 (s, 1 H), 5.43 (dd, J = 7.7, 2.4 Hz, 1 H), 4.22 (m, 2 H), 3.78 (t, J = 4.3 Hz, 1 H), 3.30 (m, 1 H), 2.81 (dd, J = 7.5, 4.6 Hz, 1 H), 2.70 (s, 3 H), 2.54 (m, 1 H), 2.37 (d, J = 12.7 Hz, 1 H), 2.09 (s, 3 H), 1.93 (m, 1 H), 1.72 (m, 2 H), 1.49 (m, 2 H), 1.43 (m, 3 H), 1.37 (s, 3 H), 1.32 (s, 3 H), 1.17 (d, J = 6.8 Hz, 3 H), 1.08 (s, 3 H), 1.01 (d, J = 6.9 Hz, 3 H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 220.5, 170.5, 165.1, 151.7, 137.5, 119.6, 116.1, 74.1, 72.8, 69.5, 61.6, 61.3, 53.7, 53.0, 42.9, 39.1, 36.3, 32.2, 32.0, 31.7, 30.6, 29.2, 22.7, 22.3, 19.6, 19.0, 17.1, 15.7, 13.7; LRMS calcd for $C_{27}H_{41}O_5NS$ 507.3, found 508.4 (M + H).

Preparation of Compound 109. To a solution of *tert*-butyllithium (3.6 mL of a 1.7 M solution in Et_2O; 6.08 mmol) in Et_2O (5 mL) at −78 °C was added a solution of 4-iodo-2-methyl-1-butene (0.596 g; 3.04 mmol) in Et_2O (20 mL). After 0.5 h, a solution of aldehyde **65** (1.03 g; 1.52 mmol) in Et_2O (5 mL) was added. After 5 min at −78 °C, the cooling bath was removed, and the solution was allowed to warm to 0 °C and stirred for 0.5 h. The reaction mixture was then poured into saturated aqueous NH_4Cl solution (100 mL) and extracted with Et_2O (2 × 100 mL). The organic layer was dried ($MgSO_4$), filtered, and concentrated. The residue was purified by flash chromatography (silica, 5−7% Et_2O/hexanes) to give a 3:1 mixture of diastereomeric alcohols (1.01 g; 89%) as a white foam.

A solution of the mixture of alcohols prepared above (1.01 g, 1.35 mmol), thiocarbonyl diimidazole (0.801 g, 4.05 mmol), and 4-DMAP (0.164 g, 1.35 mmol) in THF (10 mL) was heated to 90 °C. A stream of N_2 was then used to evaporate the THF completely. The sticky residue was maintained at 90 °C for 2 h, cooled to rt, and diluted with CH_2Cl_2 (5 mL). Purification of the residue by flash chromatography (SiO_2, 20% EtOAc:hexane) provided an epimeric mixture of thionoimidazolides (1.25 g, 99%) as a pale yellow foam.

A solution of the mixture of thionoimidazolides prepared above (1.25 g, 1.34 mmol), n-Bu_3SnH (0.66 mL, 2.01 mmol), and AIBN (0.022 g, 0.13 mmol) in benzene (14 mL) was refluxed for 1 h. The reaction mixture was cooled to rt and diluted with Et_2O (14 mL). To this solution, DBU (0.32 mL, 2.01 mmol) was added, and the resulting mixture was titrated with a solution of iodine in Et_2O until the solution color turned yellow and white precipitate formed. This slurry was filtered through a 5 cm thick pad of silica gel and concentrated in vacuo. The residue was purified by flash chromatography (SiO_2, 25% CH_2Cl_2:hexanes) to give the dithiane **109** (0.823 g, 84%) as a white foam: $[\alpha]_D$ = 5.97 (c 15.9, $CHCl_3$); IR (film) 2931, 1428, 1252, 1114, 1057 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.70 (dd, J = 7.8, 1.3 Hz, 6 H), 7.45−7.38 (band, 9 H), 4.69 (s, 1 H), 4.64 (s, 1 H), 4.56 (d, J = 3.9 Hz, 1 H), 4.16 (s, 1 H), 3.53 (d, J = 5.2 Hz, 1 H), 2.71 (d, J = 13.4 Hz, 1 H), 2.54−2.43 (band, 2 H), 2.08 (m, 1 H), 1.79−1.73 (band, 4 H), 1.73 (s, 3 H), 1.66−1.61 (band, 2 H), 1.35 (s, 3 H), 1.32 (m, 1 H), 1.16 (s, 1 H), 1.03 (d, J = 7.3 Hz, 3 H), 0.91−0.85 (band, 11 H), 0.65 (d, J = 6.8 Hz, 3 H), −0.03 (s, 3 H), −0.05 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 146.3, 135.9, 134.9, 129.8, 127.8, 109.3, 77.4, 77.1, 38.0, 37.8, 31.0, 30.4, 29.6, 26.2, 25.9, 25.5, 22.6, 22.4, 20.9, −3.1, −4.0; HRMS calcd for $C_{43}H_{64}O_2S_2Si_2$: 732.3886; found: 755.3784 (M + Na).

Preparation of Compound 110. A solution of compound **109** (0.95 g, 1.3 mmol) in MeOH/THF (2:1, 18 mL) was treated with [bis-(trifluoroacetoxy)iodobenzene] (1.118 g, 2.6 mmol) at rt. After 15 min, the reaction was quenched with saturated aqueous $NaHCO_3$ (25 mL). The mixture was extracted with Et_2O (3 × 25 mL), and the organic layer was dried ($MgSO_4$) and concentrated in vacuo. The residue was dissolved in dioxane/water (5:1, 12 mL) and treated with pTSA·H_2O (0.74 g, 3.9 mmol), and the resulting mixture was heated at 50 °C for 2 h. After cooling to rt, the mixture was diluted with Et_2O (50 mL) and washed successively with aqueous $NaHCO_3$ (15 mL) and brine (20 mL). The organic layer was then dried over $MgSO_4$, filtered, and concentrated. Purification of the residue by flash chromatography (SiO_2, 25% CH_2Cl_2:hexanes) afforded aldehyde **110** (0.79 g, 95%) as a white foam: $[\alpha]_D$ = −15.2 (c 11.8, $CDCl_3$); IR (film) 2931, 1722, 1472, 1429 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.75 (s, 1 H), 7.64 (d, J = 6.6 Hz, 6 H), 7.46−7.37 (band, 9 H), 4.69 (s, 1 H), 4.59 (s, 1 H), 4.04 (d, J = 4.2 Hz, 1 H), 3.52 (dd, J = 5.3, 2.5 Hz, 1 H), 2.16 (m, 1 H), 1.77 (m, 2 H), 1.70 (s, 3 H), 1.54 (m, 1 H), 1.44−1.26 (band, 3 H), 1.00 (s, 6 H), 0.92−0.80 (band, 15H), 0.62 (d, J = 6.8 Hz, 3 H), 0.08 (s, 3 H), 0.02 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 205.6, 146.0, 135.7, 134.4, 130.1, 127.9, 109.6, 81.1, 76.9, 51.7, 43.3, 38.9, 38.0, 30.4, 26.2, 25.6, 22.4, 21.1, 20.1, 18.6, 15.1, 12.8, −3.2, −4.0; HRMS calcd for $C_{40}H_{58}O_3Si_2$ 642.3925, found 665.3842 (M + Na).

Preparation of Compound 114. To a solution of acetate **77** (0.285 g, 1.16 mmol) and aldehyde **110** (0.49 g, 0.772 mmol) in THF (1 mL) at −78 °C was added LDA (2 M soln in THF, 0.772 mL, 1.54 mmol). The yellow mixture was stirred at −78 °C for 40 min and then quenched with saturated aqueous NH_4Cl (10 mL). The mixture was extracted with Et_2O (3 × 15 mL), dried ($MgSO_4$), filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (SiO_2, 6% EtOAc:hexanes) provided α-alcohol **114** (0.239 g, 35%) and β-alcohol **115** (0.238 g, 35%).

To a solution of β-alcohol **115** (0.238 g, 0.27 mmol) in CH_2Cl_2 (3 mL) was added Dess−Martin periodinane (0.689 g, 1.62 mmol) at rt. The resulting solution was stirred for 1 h and then quenched by the addition of 1:1 saturated aqueous $NaHCO_3$:$Na_2S_2O_3$ (10 mL). The mixture was extracted with Et_2O (3 × 10 mL). The organic layer was dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The resulting C-3 ketone **116** was *immediately* dissolved in a mixture of MeOH (4 mL) and THF (2 mL) and cooled to −78 °C. Sodium borohydride (0.102 g, 2.7 mmol) was then added, and the mixture was allowed to warm to rt and stirred for 1 h. The reaction was then quenched with saturated aqueous NH_4Cl (10 mL). The mixture was extracted with ether (3 × 15 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo*. Purification of the residue by flash chromatography (SiO_2, 8% EtOAc:hexanes) provided α-alcohol **114** (0.230 g, 92%): $[\alpha]_D$ = −45.3 (c 0.29, $CHCl_3$); IR (film) 3502, 2927, 1715, 1428 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.64 (d, J = 6.5 Hz, 6 H), 7.45−7.30 (band, 9 H), 6.93 (s, 1 H), 6.50 (s, 1 H), 5.65 (m, 1 H), 5.31 (t, J = 6.7 Hz, 1 H), 5.05 (m, 2 H), 4.69 (s, 1 H), 4.63 (s, 1 H), 4.13 (d, J = 3.5 Hz, 1 H), 4.00 (m, 1 H), 3.73 (d, J = 4.5 Hz, 1 H), 3.25 (d, J = 2.5 Hz, 1 H), 2.71 (s, 3 H), 2.42 (m,

4 H), 2.06 (m, 1 H), 2.02 (s, 3 H), 1.87−1.78 (band, 2 H), 1.75 (s, 3 H), 1.70 (m, 1 H), 1.31−1.20 (band, 2 H), 1.12 (m, 1 H), 1.02 (d, J = 8.0 Hz, 3 H), 0.96 (s, 3 H), 0.91 (s, 3 H), 0.79 (s, 9 H), 0.60 (d, J = 7.0 Hz, 3 H), −0.06 (s, 3 H), −0.07 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 131.1, 164.6, 152.4, 146.2, 136.4, 135.7, 134.9, 133.2, 129.8, 127.7, 121.4, 117.9, 116.5, 109.4, 79.0, 78.7, 76.3, 71.8, 44.2, 43.0, 38.7, 38.0, 37.4, 36.5, 30.4, 26.2, 26.1, 25.7, 22.5, 21.0, 20.5, 19.2, 18.5, 15.2, 14.5, 13.4, −3.1, −4.3; HRMS calcd for $C_{53}H_{75}O_5NSSi_2$: 893.4904, found: 932.4529 (M + K).

Preparation of Compound 120. The aldol product **114** (0.219 g, 0.27 mmol) was treated with buffered HF·pyridine in THF (8.0 mL) at rt (the stock solution was prepared from 20 mL THF, 11.4 mL pyridine and 4.2 g hydrogen fluoride−pyridne (Aldrich Co.)). After 2 h, the reaction was poured into saturated aqueous $NaHCO_3$ and extracted with Et_2O. The organic layer was washed in sequence with saturated aqueous $CuSO_4$ (3 × 30 mL) and saturated aqueous $NaHCO_3$ (50 mL), dried over Na_2SO_4, and concentrated in vacuo. The residue was purified by flash chromatography (silica, 10 → 20% EtOAc in hexane) to give diol **120** (0.15 g, 93%) as a white foam: $[\alpha]_D = -40.5$ (c 3.8, $CDCl_3$); IR (film) 3457, 2930, 1732, 1472, 1386, 1252 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.93 (s, 1 H), 6.51 (s, 1 H), 5.72 (m, 1 H), 5.35 (t, J = 6.7 Hz, 1 H), 5.08 (m, 2 H), 4.68 (s, 1 H), 4.65 (s, 1 H), 4.07 (d, J = 10.0 Hz, 1 H), 3.92 (br s, 1 H), 3.80 (br s, 1 H), 3.49 (br s, 1 H), 2.68 (s, 3 H), 2.61−2.45 (band, 4 H), 2.07 (d, J = 1.2 Hz, 3 H), 2.01 (br s, 1 H), 1.69 (s, 3 H), 1.55 (m, 1 H), 1.36 (m, 1 H), 0.99 (d, J = 6.9 Hz, 3 H), 0.88 (s, 9 H), 0.80 (s, 3 H), 0.09 (s, 3 H), 0.07 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 172.7, 164.5, 152.5, 145.8, 136.8, 133.3, 120.9, 117.7, 116.3, 109.8, 78.5, 41.5, 38.0, 37.5, 37.1, 32.9, 25.9, 25.5, 22.3, 21.1, 19.1, 18.2, 16.1, 14.6, −4.4; LRMS calcd for $C_{35}H_{61}O_5NSSi$: 635.4; found: 658.5 (M + Na).

Preparation of Compound 121. To a cooled (−30 °C) solution of diol **120** (0.110 g, 0.173 mmol) and 2,6-lutidine (0.121 mL, 1.04 mmol) in anhydrous CH_2Cl_2 (2 mL) was added TBSOTf (0.119 mL, 0.519 mmol). The resulting solution was then stirred at −30 °C for 30 min. The reaction was quenched with 0.5 M HCl (50 mL) and extracted with Et_2O (150 mL). The Et_2O layer was washed with saturated aqueous $NaHCO_3$ (50 mL), dried (Na_2SO_4), and concentrated in vacuo. Purification of the residue by flash chromatogrphy (silica, 5 → 8% EtOAc in hexane) afforded TBS ether **121** (0.112 g, 85%) as white foam: $[\alpha]_D = -33.7$ (c 1.6, $CHCl_3$); IR (film) 3478, 2929, 1737, 1471, 1253 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.93 (s, 1 H), 6.50 (s, 1 H), 5.71 (m, 1 H), 5.30 (t, J = 6.8 Hz, 1 H), 5.08 (m, 2 H), 4.67 (s, 1 H), 4.65 (s, 1 H), 4.20 (m, 1 H), 3.81 (br s, 1 H), 3.40 (br s, 1 H), 2.90 (dd, J = 17.2, 3.9 Hz, 1 H), 2.69 (s, 3 H), 2.47 (m, 2 H), 2.35 (dd, J = 17.2, 5.4 Hz, 1 H), 2.07 (s, 3 H), 2.01 (m, 2 H), 1.99 (m, 1 H), 1.69−1.60 (band, 4 H), 1.54−1.48 (band, 2 H), 1.25 (m, 1 H), 0.94 (d, J = 7.0 Hz, 3 H), 0.91 (s, 3 H), 0.89−0.87 (band, 12 H), 0.86 (s, 3 H), 0.83 (s, 9 H), 0.07 (s, 3 H), 0.02 (s, 3 H), 0.01 (s, 3 H), −0.05 (s, 3 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 172.7, 164.5, 152.5, 146.2, 136.7, 133.4, 121.2, 117.8, 116.4, 109.7, 78.7, 78.1, 74.4, 43.4, 39.8, 38.1, 37.5, 32.9, 26.0, 25.9, 25.6, 22.4, 19.9, 19.2, 18.2, 18.1, 14.2, −4.0, −4.2, −4.22, −4.9; HRMS calcd for $C_{41}H_{75}O_5NSSi_2$: 749.4904, found: 788.4518 (M + K).

Preparation of Compound 122. To a solution of alcohol **121** (0.110 g, 0.147 mmol) in CH_2Cl_2 (2.0 mL) at rt was added Dess−Martin periodinane (0.249 g, 0.583 mmol) in one portion. The resulting solution was then allowed to stir at 25 °C for 1.5 h. The reaction was quenched by the addition of 1:1 saturated aqueous $NaHCO_3$:$Na_2S_2O_3$ (10 mL) and stirred for 5 min. The mixture was then extracted with Et_2O (3 × 15 mL). The organic layer was dried (Na_2SO_4), filtered, and concentrated in vacuo. Purification of the residue by flash chromatography (silica, 6% EtOAc in hexane) provided ketone **122** (0.099 g, 94%) as white foam: $[\alpha]_D = -55.1$ (c 0.85, $CHCl_3$); IR (film) 2929, 1737, 1695, 1253 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.95 (s, 1 H), 6.49 (s, 1 H), 5.70 (m, 1 H), 5.29 (t, J = 6.8 Hz, 1 H), 5.05 (m, 2 H), 4.68 (s, 1 H), 4.65 (s, 1 H), 4.34 (dd, J = 5.9, 3.4 Hz, 1 H), 3.72 (d, J = 5.4 Hz, 1 H), 3.15 (m, 1 H), 2.70 (s, 3 H), 2.53−2.42 (band, 3 H), 2.28 (dd, J = 17.0, 6.1 Hz, 1 H), 2.06 (s, 3 H), 1.99 (m, 2 H), 1.69 (s, 3 H), 1.41 (m, 1 H), 1.33−1.29 (band, 3 H), 1.25−1.21 (band, 4 H), 1.04−1.02 (band, 6 H), 0.92−0.83 (band, 21 H), 0.10 (s, 3 H), 0.05 (s, 3 H), 0.03 (s, 6 H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 171.2, 164.5, 152.5, 146.0, 136.7, 133.4, 121.1, 117.8, 116.4, 109.8, 78.6, 77.7, 74.0, 53.3, 45.2, 40.3, 38.7, 38.3, 37.5, 31.6, 26.2, 26.0, 25.6, 23.1, 22.4, 20.3, 19.3, 18.5, 18.2, 17.1, 15.5, 14.5, −3.6, −3.8, −4.3, −4.7; HRMS calcd for $C_{41}H_{73}O_5NSSi_2$: 747.4748; found: 786.4362 (M + K).

Preparation of Compound 124. To a solution of diene **122** (0.015 g; 0.02 mmol) in dry, degassed benzene (20 mL) at rt was added Schrock's metathesis catalyst [Mo(CHMe$_2$Ph)(N-(2,6-(i-Pr)$_2$C$_6$H$_3$))-(OCMe(CF$_3$)$_2$)$_2$] (0.0038 g; 0.005 mmol) in a dry box. The reaction mixture was then warmed to 55 °C and stirred for 2 h. The reaction mixture was concentrated in vacuo, and the residue was purified by flash chromatography (silica, 4% EtOAc/hexanes) to give an inseparable 1:1 mixture of stereoisomeric alkenes **105** and **123** (0.013 g; 86%).

To a solution of the mixture of alkenes prepared above (0.013 g; 0.018 mmol) in THF (1 mL) was added HF·pyridine (0.5 mL), and the resulting solution was stirred for 1.5 h. The reaction mixture was then poured into saturated aqueous $NaHCO_3$ solution (50 mL) and extracted with chloroform (3 × 30 mL). The organic layer was dried ($MgSO_4$), filtered, and concentrated. The residue was purified by flash chromatography (silica, 30% EtOAc/hexanes) to give an equimolar mixture of stereoisomeric alkenes **106** and **124** (0.008 g; 90%). The mixture was separated by preparative thin-layer chromatography (2% MeOH/CH_2Cl_2, four elutions). **124**: IR (film) 3484, 2922, 1732, 1693, 1464, 1260 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.98 (s, 1 H), 6.57 (s, 1 H), 5.30 (m, 1 H), 5.11 (t, J = 6.7 Hz, 1 H), 4.32 (d, J = 10.0 Hz, 1 H), 3.67 (m, 1 H), 3.29−3.25 (band, 3 H), 2.70 (s, 3 H), 2.65−2.45 (band, 5 H), 2.16 (m, 1 H), 2.07 (s, 3 H), 1.98 (m, 1 H), 1.73−1.61 (band, 3 H), 1.60 (s, 3 H), 1.31 (m, 1 H), 1.28 (s, 3 H), 1.17 (d, J = 6.8 Hz, 3 H), 1.05 (s, 3 H), 0.98 (d, J = 7.0 Hz, 3 H); HRMS calcd for $C_{27}H_{41}O_5NS$: 491.2705; found: 492.2795 (M + H).

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Supporting Information Available: Experimental procedures for the preparation of compounds **10**, **12**, **13**, **15**, **16**−**20**, **22**, **24**, **27**, **37**−**44**, **55**−**57**, **65**, **70**−**75**, **107**, **108**, **111**, **112**, **117**−**119**, and **125**−**136** as well as full characterization data are included (60 pages). See any current masthead page for ordering and Internet access instructions.

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Communications to the Editor

Remote Effects in Macrolide Formation through Ring-Forming Olefin Metathesis: An Application to the Synthesis of Fully Active Epothilone Congeners

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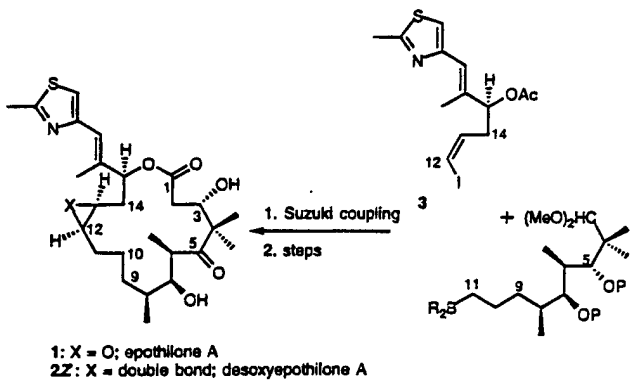
Recently, we achieved the first synthesis of epothilone A (structure **1**).[1] Aside from numerous chemical issues which must be addressed in accomplishing such a synthesis, interest in the epothilone class of compounds is further heightened by claims (thus far based solely on *in vitro* measurements) that the epothilones may constitute a useful group of anticancer agents, operating through the same mechanism of action as paclitaxel.[2] It has further been suggested, again on the basis of *in vitro* data, that the epothilones offer advantages relative to paclitaxel in terms of ease of formulation and potency toward drug resistant cell lines.

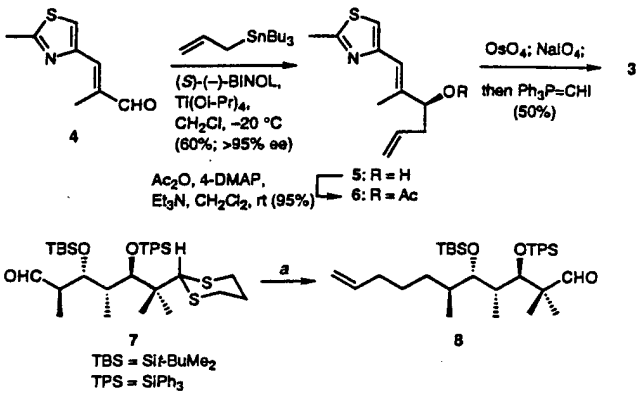
In our synthesis of epothilone A (**1**), we passed through the desoxycompound **2Z**. We showed, for the first time, that the action of dimethyldioxirane on compound **2Z** results in a highly diastereoselective epoxidation, providing compound **1**. The strategy we employed to construct compound **2Z** provided strict control over the geometry of the C12−C13 double bond through a *B*-alkyl Suzuki coupling reaction of *cis* vinyl iodide **3** with an appropriate borane (Scheme 1).

The studies described herein focused on a different method for the construction of desoxyepothilone A (**2Z**). In particular, we investigated the possibility of a ring-forming olefin metathesis reaction to construct the C12−C13 bond.[3] We were particularly mindful of a precedent furnished by Hoveyda et al.[3b] It was hoped that such an assembly strategy involving components of the type **6** and **8** might lead to an even more direct route to the natural series and analogs thereof. *These studies became of particular interest when it was found, surprisingly, that desoxyepothilone A (2Z) has the full biological activity of epothilone A as manifested through independent investigations at the level of cytotoxicity and polymerization of stable microtubules in the absence of GTP.* Herein we describe a straightforward route to reach substrates needed for olefin metathesis. We also disclose the results of these cyclizations which indicate a remarkable sensitivity to permutations of functionality and stereochemistry at centers far removed from the site of olefin metathesis. Finally, we describe some early but exciting SAR results which indicate that significant structural variances can be introduced in this series with maintenance of full biological function.

Scheme 1



Scheme 2[a]



[a] Key: (a) (i) 3-butenylmagnesium bromide, Et_2O, −78 to 0 °C (92%); (ii) thiocarbonyldiimidazole, DMAP, 95 °C; (iii) Bu_3SnH, AIBN, C_6H_6, 80 °C (83% for two steps); (iv) $(CF_3CO_2)_2IC_6H_5$, MeOH, THF; (v) *p*TSA, dioxane, H_2O, 50 °C (85% for two steps).

Our new strategy commences with aldehyde **4**, a substance available in multigram quantities.[1b,c] An important technological advance in the area was registered when it was found that subjection of aldehyde **4** to the catalytic asymmetric allylation protocol previously described by Keck leads to **5** in >95% enantiomeric excess (Scheme 2).[4] As an aside, we note that **5** was converted in two steps to the previously mentioned vinyl iodide **3**, thereby effecting a major economy in the earlier synthesis. For purposes to be described, compound **5** was simply converted to the ester **6**. The pre-acyl construct **8** was assembled from the dithiane aldehyde **7**[1a,b] in the manner indicated in Scheme 2. We thus had in hand the two subunits required to study ring forming olefin metathesis *en route* to the C12−C13 bond.

The compounds **6** and **8** were joined through a simple intermolecular aldol addition. That this reaction produced an approximately 1:1 mixture of the epimers **9** and **10** was *per se*

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(2) See: Höfle, G.; Bedorf, N.; Steinmetz, H.; Schomberg, D.; Gerth, K.; Reichenbach, H. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 1567 and references therein.

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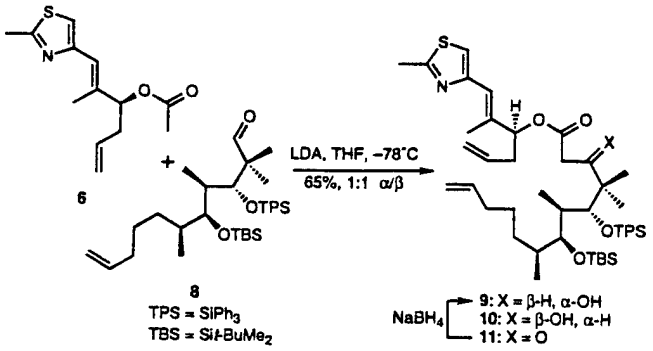
Scheme 3



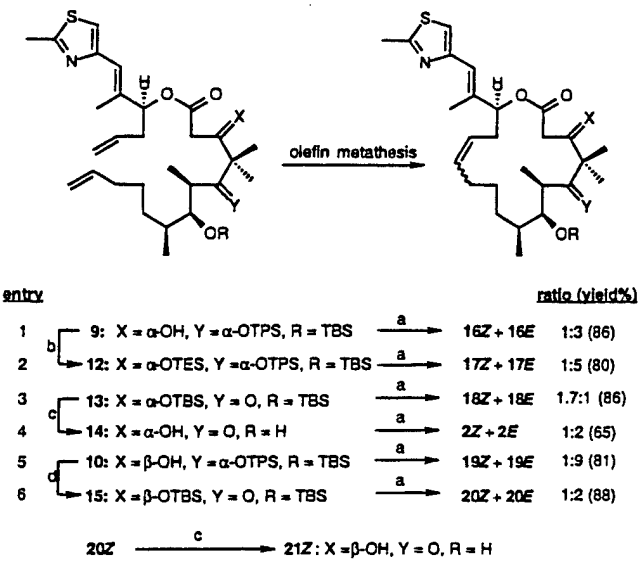
of no consequence, since the latter could be converted to the former through an oxidation/reduction sequence (i.e., **10** → **11** → **9**) (Scheme 3). Indeed, much was learned chemically and biologically from having both the 3*S* (cf. **9**) and 3*R* (cf. **10**) series available to us. From the core compounds **9** and **10**, we could easily fashion substrates **12**−**14**. We were then in a position to study the ring-forming olefin methathesis (ROM) reaction (Scheme 4).

Cyclization reactions were conducted under the conditions shown in Scheme 4 for compounds **9**, **10**, and **12**−**14**. As seen, we could readily obtain products containing the *E* C12−C13 double bond (see formation of **19*E*,*Z***). However, at this writing the highest ratio for the *Z* product is only 1.7:1 (see formation of **18*E*,*Z***). We note that, with all protecting groups identical, the proportion of *E* product increases upon changing from the 3*S* to 3*R* series (see formation of **19*E*,*Z*** and **20*E*,*Z***). Similarly, keeping C-3 and C-7 constant but permutating C-5 (see ROM substrates **13** and **14**) affords more of the *Z* olefin product (see formation of **18*E*,*Z*** and **2*E*,*Z***).

Using this chemistry, we could easily access the fully deprotected *cis*-desoxyepothilone A (**2*Z***). Of course, this work constitutes a second synthesis of **2*Z*** and a formal total synthesis of **1**.[5] It is noteworthy that the concise route to enantiomerically pure vinyl iodide **3** (see Scheme 2) renders our initial approach to **2*Z*** more practical. Nevertheless, the ROM chemistry described herein provides an eminently more workable route to *trans*-desoxyepothilone A (**2*E***). *Remarkably, compound (**2*E***) is fully active as measured by cytotoxicity and microtubule assays. Perhaps equally surprising, biological activity is abrogated in the 3R compounds **21*Z*** and 3-epi-epothilone A (3-epi-**1**).*[6]

(5) It has been brought to our attention through the popular media that another total synthesis of epothilone A has been subsequently completed utilizing ring-closing olefin metathesis. Nicolaou, K. C.; et al. *Angew. Chem., Int. Ed. Engl.* **1997**, *37*, 166.

Scheme 4[a]



[a] Key: (a) $RuBnCl_2(PCy_3)_2$ (50 mol %), C_6H_6, 0.001 M, rt, 24 h; (b) TESCl, imidazole, DMF, (80%); (c) pyridine hydrofluoride, THF, rt; (d) (i) pyridine hydrofluoride, pyridine, THF, rt, (93%); (ii) TBSOTf, 2,6-lutidine, −35 °C, (95%); (iii) Dess−Martin periodinane, (87%) (TBS = *tert*-butyldimethylsilyl; TPS = triphenylsilyl; TES = triethylsilyl).

In summary, a route to (*E*)- and (*Z*)-desoxyepothilones using ROM technology has been accomplished. Through this and related methodology to be described soon, highly biologically active congeners have been obtained, and a total synthesis driven mapping of the SAR of epothilones is well underway.

Acknowledgment. This research was supported by the National Institutes of Health (grant numbers CA-28824 (S.J.D.) CA-39821 (S.B.H.)). Postdoctoral fellowship support is gratefully acknowledged by E.J.S. (NSF, CHE-9504805), A.B. (NIH, CA-GM 72231), P.B. (NIH, CA 62948). We gratefully acknowledge Dr. George Sukenick (NMR Core Facility, Sloan-Kettering Institute) for NMR and mass spectral analyses.

Supporting Information Available: Preparation of substrates for olefin methathesis (**9**, **10** and **12**−**15**) and compounds **21*Z*** and 3-*epi*-epothilone A and relevant biological data (IC_{50} values) as well as all relevant spectral data for compounds **2**−**21** (43 pages). See any current masthead page for ordering and Internet access instructions.

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(6) The compound 3-*epi*-epothilone A was produced by treatment of 3-*epi*-desoxyepothlone A (**21*Z***) with dimethyldioxirane at −35 °C.

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Stereoselective Syntheses and Evaluation of Compounds in the 8-Desmethylepothilone A Series: Some Surprising Observations Regarding Their Chemical and Biological Properties

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[b]Yu-Huang Zheng, [b]Ting-Chao Chou, [d]Lifeng He and [d]Susan B. Horwitz

Contribution from [a]The Laboratory for Bioorganic Chemistry, [b]The Laboratory for Biochemical Pharmacology, The Sloan-Kettering Institute for Cancer Research, 1275 York Ave., New York, N.Y. 10021,

[c]The Department of Chemistry, Columbia University, Havemeyer Hall, New York, N.Y. 10027 and

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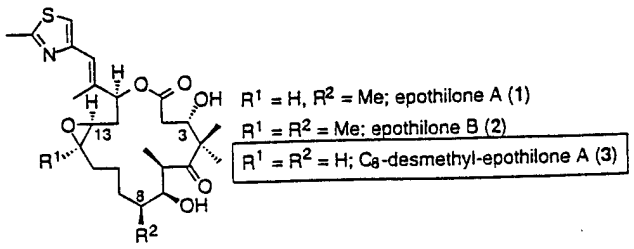
Abstract: The title compounds have been synthesized in a convergent way by recourse to a Weiler type dianion construction. © 1997 Elsevier Science Ltd.

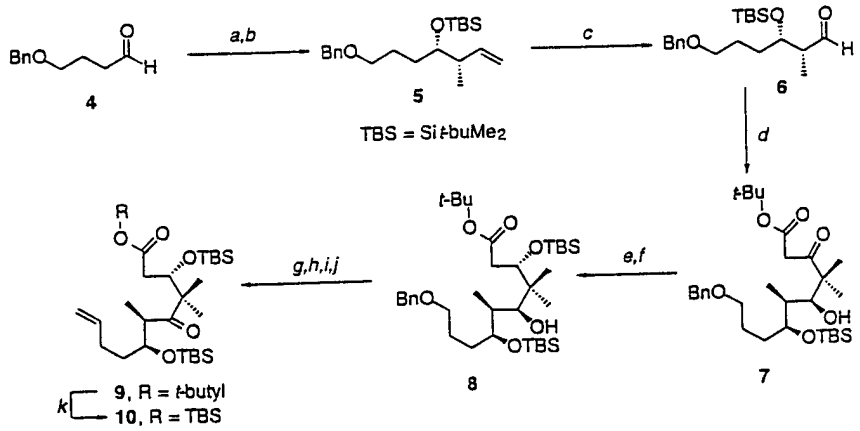
Recently, several groups have described total syntheses of epothilones A (**1**) and B (**2**)[1,2] whose mode of antitumor action closely mimics that of taxol™.[3] Although taxol™ (Paclitaxel) is a clinically proven drug, its formulation continues to be difficult. In addition, taxol induces the multidrug resistance (MDR) phenotype. Hence, any novel agent that has the same mechanism of action as taxol and has the prospect of having superior therapeutic activity warrants serious study.[4]

A central challenge now is that of generating epothilone analogs that are more effective and more readily synthesized than is the case for **1** and **2**. Though the syntheses of the natural products can provide ample material for preliminary biological evaluation, the prospect of producing adequate amounts of these compounds for full development would be daunting. One particular area where a structural change could bring significant relief from the complexities of the synthesis would be in the deletion of the C_8 methyl group from the polypropionate domain (see goal system **3**). The need to deal with this C_8 chiral center complicates all of the syntheses of epothilone thus far reported. In the context of our own program, deletion of the C_8 methyl group would prompt a major change in synthetic strategy relative to our earlier diene-aldehyde cyclocondensation route.[5]



Asymmetric crotylation[6] (87% ee) of **4** followed by protection led to the TBS ether **5**. The double bond was readily cleaved to give rise to aldehyde **6**. The aldehyde was coupled to the dianion derived from *t*-butyl isobutyrylacetate to provide **7**. The ratio of the C_{5S} (**7** shown here): C_{5R} compound (not shown) is *ca* 10:1. That the Weiler-type β-ketoester dianion chemistry[7] can be conducted in the context of the isobutyryl group prompts several alternate perceptions for still more concise syntheses. Directed reduction of the C_3 ketone of **7** following literature precedents,[8] followed by selective silylation of the C_3 hydroxyl gave a 50% yield of a 10:1 ratio of the required C_{3S} (see compound **8**): to C_{3R} isomer (not shown).[9] The carbinol, produced upon debenzylation, was oxidized to an aldehyde which, following methylenation through a simple Wittig reaction, afforded olefin **9**. Treatment of this compound with TBSOTf provided ester **10** which was used directly in the Suzuki coupling with the vinyl iodide **12** (*vide infra*).

Scheme 1.[a]



[a] a) (Z)-Crotyl-B[(–)-Ipc]$_2$, –78°C, Et_2O, then 3N NaOH, 30% H_2O_2; b) TBSOTf, 2,6-lutidine, CH_2Cl_2 (74% for two steps, 87% ee); c) O_3, CH_2Cl_2/MeOH, –78°C, then DMS, (82%); d) *t*-butyl isobutyrylacetate, NaH, BuLi, 0°C, then **6** (60%, 10:1); e) $Me_4NBH(OAc)_3$, –10°C (50%, 10:1 α/β) or $NaBH_4$, MeOH, THF, 0°C, (88%, 1:1 α/β); f) TBSOTf, 2,6-lutidine, –40°C, (88%). g) Dess-Martin periodinane, (90%); h) $Pd(OH)_2$, H_2, EtOH, (96%); i) DMSO, oxalyl chloride, CH_2Cl_2, –78°C (78%); j) Methyl triphenylphosphonium bromide, NaHMDS, THF, 0°C (85%); k) TBSOTf, 2,6-lutidine, CH_2Cl_2, rt (87%).

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conformational

[a] a) $Pd(dppf)_2Cl_2$, F
HF•pyr, THF, rt (8:

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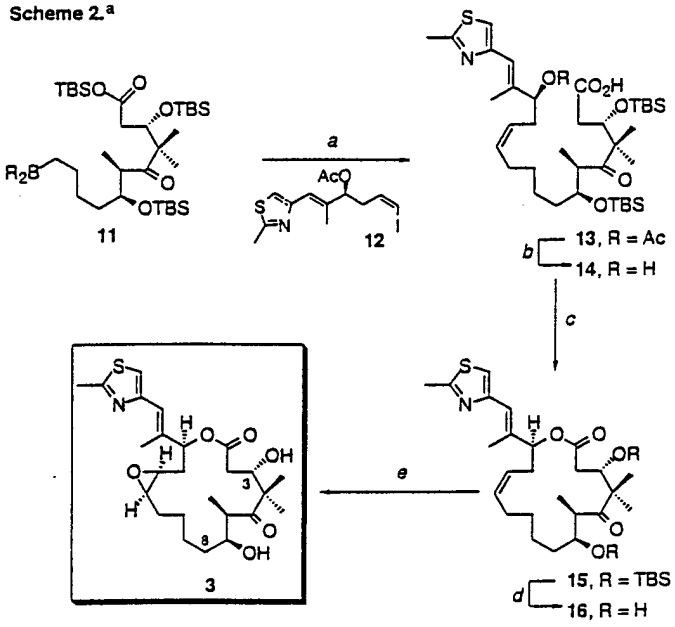
The hydroboration of **10** with 9-BBN produced intermediate **11** which, on coupling with the vinyl iodide **12** and *in situ* cleavage of the TBS ester led to **13**. After de-acetylation, the hydroxy acid **14** was in hand. Marcolactonization[10] of this compound produced **15** which, after desilylation, afforded C_3-desmethyldesoxyepothilone (**16**). Finally, epoxidation of this compound with dimethyldioxirane produced the goal structure **3**. The stereoselectivity of epoxidation was surprisingly poor (1.5:1) given that epoxidation of desoxyepothilone A occurred with >20:1 stereoselectivity. *Apparently, the deletion of the C_8 methyl group tilts the conformational distribution of 16 to forms in which the epoxidation by dimethyl dioxirane is less β-selective.*[11]

Scheme 2.[a]



[a] a) $Pd(dppf)_2Cl_2$, Ph_3As, Cs_2CO_3, H_2O, DMF, rt (62%); b) K_2CO_3, MeOH, H_2O (78%); c) DCC, 4-DMAP, 4-DMAP•HCl, $CHCl_3$ (78%); d) HF•pyr, THF, rt (82%), e) 3,3-dimethyl dioxirane, CH_2Cl_2, -35°C (72%, i.5:1).

Compounds **3** and **16** were tested for cytotoxicity in cell cultures and assembly of tubulin in the absence of GTP. Surprisingly, neither macrolide displayed significant tubulin polymerization.[12] Cytotoxicity studies showed drastically reduced activity in the 8-desmethyl series. Compounds **3** and **16** were approximately 200 times less active than their corresponding epothilone A counterparts (see Table). Recalling earlier SAR findings at both C_3 and C_5, in conjunction with the findings reported here, the polypropionate sector of the epothilones emerges as a particularly sensitive locus of biological function.[2,13] Further studies on the SAR of epothilones congeners, enabled by improved access through synthesis, are ongoing and will be disclosed in due course.

Table 1. *Relative efficacy of epothilone compounds against drug-sensitive and resistant human leukemic CCRF-CEM cell lines.*[a]

Compound	CCRF-CEM IC_{50} (μM)[b]	CCRF-CEM/VBL IC_{50} (μM)[b]	CCRF-CEM/VM_1 IC_{50} (μM)[b]
16	5.00	5.75	6.29
3	0.439	2.47	0.764
epothilone A (1)	0.003	0.020	0.003
desoxyepothilone A	0.022	0.012	0.013
epothilone B (2)	0.0004	0.003	0.002
desoxyepothilone B	0.009	0.017	0.014
taxol®	0.002	3.390	0.002

[a]The cytotoxicities of test compounds were determined by the growth of human lymphoblastic leukemic cells CCRF-CEM, or their sublines resistant to vinblastine and taxol (CCRF-CEM/VBL) or resistant to etoposide (CCRF-CEM/VM-1). XTT-microculture tetrazolium/formazan assays were used.

[b]The IC_{50} values were calculated from 5-6 concentrations based on the median-effect plot using computer software.

Acknowledgments. This research was supported by the National Institutes of Health (Grant numbers CA 28824 (S.J.D.) and CA 39821 (S.B.H.)). Postdoctoral fellowship support is gratefully acknowledged to E.J.S. (NSF CHE-9504805), A.B. (NIH, CA-GM 72231), P.B. (NIH CA 62948)). We gratefully acknowledge Dr. George Sukenick (NMR Core Facility, Sloan-Kettering Institute) for NMR and mass spectral analyses.

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9. Reduction with sodium borohydride afforded a ca. 1:1 mixture of C_3 epimers.
10. Boden, E. P.; Keck, G. E. *J. Org. Chem.* **1985**, *50*, 2394.
11. It is interesting to speculate as to whether the effect of the C_8 methyl on the stereoselectivity of epoxidation by dimethydioxirane and the dramatic reduction of biological activity are related. Molecular modeling studies designed to explore this theme are under way.
12. Microtubule protein (MTP) was purified from calf brains by two cycles of temperature dependent assembly and disassembly.[14] In control assembly experiments, MTP (1 mg/mL) was diluted in assembly buffer containing 0.1 M MES (2-(N-morpholino) ethanesulfonic acid), 1 mM EGTA, 0.5 nM $MgCl_2$, 1mM GTP and 3M glycerol, pH 6.6. The concentration of tubulin in MTP was estimated to be about 85%. Assembly was monitored spectrophotometrically at 350 nm, 35°C for 40 min by following changes in turbidity as a measure of polymer mass.[15] Drugs were tested at a concentration of 10 μM, in the absence of GTP. Microtubule formation was verified by electron microscopy. To determine the stability of microtubules assembled in the presence of GTP or drug, turbidity was followed for 40 min after the reaction temperature was shifted to 4°C.
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Total Synthesis of (−)-Epothilone A**

Aaron Balog, Dongfang Meng, Ted Kamenecka, Peter Bertinato, Dai-Shi Su, Erik J. Sorensen, and Samuel J. Danishefsky*

Epothilones A and B were isolated from the myxobacteria of the genus *Sorangium*. The full structures of these compounds, determined from an X-ray crystallographic analysis, were recently communicated by Höfle.[1] Interest in the total synthesis of the epothilones arises from several considerations. First, they contain substructural motifs that pose intrinsic problems for a total synthesis. Not uncommonly, solutions to novel structural challenges carry with them important lessons for organic chemistry, which transcend the particular goal under investigation. Moreover, the biological profiles of the epothilones warrant multidisciplinary scientific attention. As is well known by now, taxol is already a useful resource in chemotherapy against ovarian and breast cancers.[2] Furthermore, its range of applicability seems to be expanding under continuing clinical scrutiny. The mechanism of the cytotoxic action of taxol, at least at the in vitro level, involves stabilization of microtubule assemblies.[3] A series of complementary in vitro investigations with the epothilones indicated that they function by the same mechanism as the taxoids, apparently down to the binding sites to their protein target.[4] Moreover, the epothilones surpass taxol in terms of cytotoxicity and far surpass it as regards in vitro efficacy against drug-resistant cells. Since multiple drug resistance (MDR) is one of the serious limitations of taxol,[5] any agent that promises relief from this problem merits serious attention. Furthermore, formulatability of the epothilones is claimed to be more straightforward than is the case with taxol. Accordingly, we have undertaken the total synthesis of epothilones, focusing first on epothilone A (**1**). Herein we report for the first time the total synthesis of this target.

Carbons 9 through 11 insulate the chiral domains embracing carbons 3 through 8 on the acyl side of the macrolactone, and carbons 12 through 15 on the alkyl side. We reasoned from the outset that the prospects of transmitting stereochemical information from one of the segments to the other were bleak. Accordingly, it seemed more prudent to deal with the stereochemistry of each segment individually. In the acyl segment this required solution to both the relative and absolute configurations of the "polypropionate-like" network. In the alkyl segment, two possibilities presented themselves. In one instance, the C12–C13 epoxide would be included in the unit to be merged with the acyl-related substructure. In that case it would be necessary to secure the relative as well as absolute stereochemical relationships of carbons 15, 13, and 12. As matters transpired, we came to consider omitting the epoxide from the alkyl-side moiety undergoing coupling. This strategy would be feasible only if the epoxide could be introduced with acceptable stereocontrol after closure of the macrocycle.

In an earlier disclosure[6] we described the synthesis of compound **4**, which contains most of the requisite stereochemical information required for the acyl fragment. This intermediate was reached by a novel, oxidatively induced, solvolytic cleavage of the cyclopropanopyran **3**. We also described a construct containing the alkyl-side coupling partner embodying the absolute and relative stereochemistry at carbons 15, 13, and 12, which was not used in the studies described herein.[7]

Several potential connection sites attracted attention for the union of the alkyl and acyl domains. At some point an acylation would be required to establish an ester (or lactone) bond (see bold arrow *2*, Scheme 1). Furthermore, an aldol condensation seemed to be called for in fashioning a C2–C3 connection. Less obvious was the timing of this aldol step. It could be considered for the elongation of the C3–C9 construct to prepare it for acylation of the C-15 hydroxyl group. As matters developed, however, we employed a bolder possibility: the closure of the macrolide by a virtually unprecedented macroaldolization.[8] This risky but otherwise attractive option is implied by bold arrow *3*.

Considerable debate and experimentation attended the decision on the first union between the acyl and alkyl fragments (see bold arrow *1*). As alluded to in earlier publications,[6, 7] and as will be expanded upon in subsequent disclosures, significant resistance was encountered against proposed bond formation between carbons 9 and 10 or between carbons 10 and 11, for which the epoxide would be included in the alkyl coupling partner. Complications had arisen from unanticipated difficulties in fashioning acyl and alkyl reactants with the appropriate complementarity for merger across either of these bonds. We thus turned to the possibility of establishing the initial merger between carbons 11 and 12. This approach dictated deletion of the oxirane linkage from the O-alkyl coupling partner. After examination of several permutations, we settled upon generalized systems **5** and **6** to enter the first-stage coupling reaction. A de novo synthesis of a usable substrate corresponding to generalized system **5**, starting from **4**, would be necessary.

The steps leading from **4** to **11** are shown in Scheme 2. Protection of the future C-7 alcohol (see compound **7**) was followed by cleavage of the benzyl ether and oxidation to form aldehyde **8**. Elongation of the aldehyde to the terminal allyl-containing fragment **10** proceeded through enol ether **9** (mixture of *E* and *Z* geometrical isomers). Finally, the dithiane linkage was oxidatively cleaved under solvolytic trapping conditions[9] to give rise to specific coupling component **11**.

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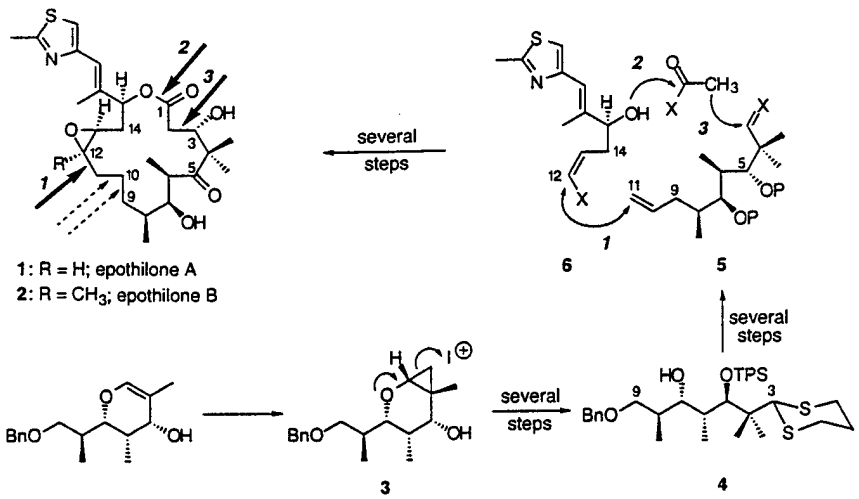
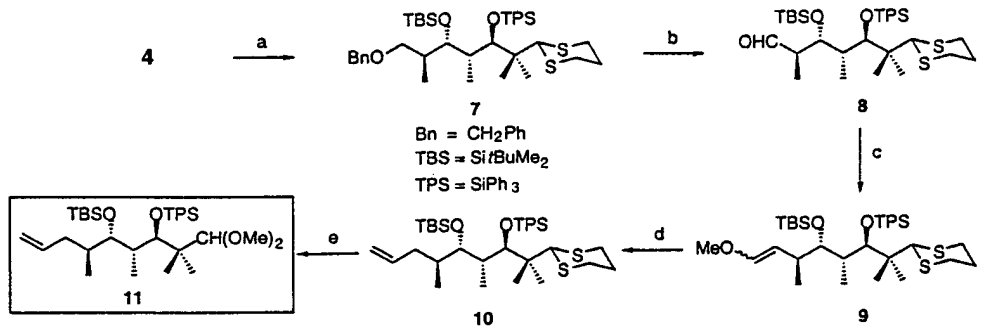


Scheme 1.

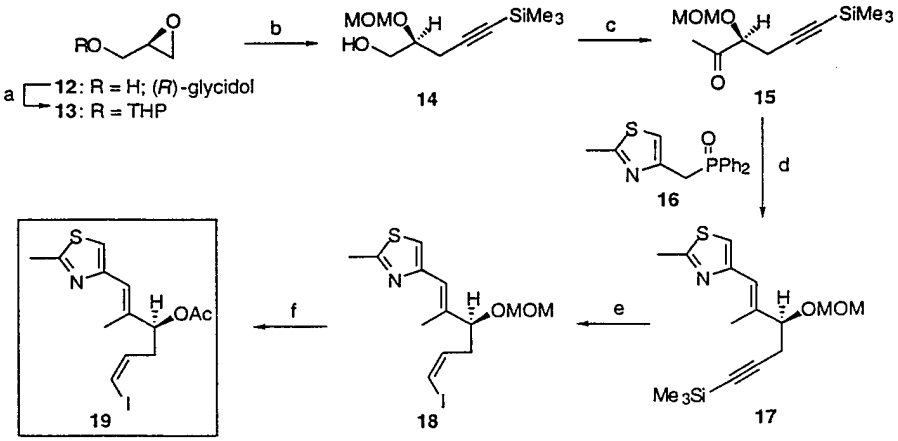


Scheme 2. a) t-BuMe$_2$OTf(Tf = trifluoromethanesulfonate), 2,6-lutidine, CH_2Cl_2, 98%; b) 1. 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ), CH_2Cl_2/H_2O, 89%; 2. $(COCl)_2$, DMSO, CH_2Cl_2, $-78\,^\circ C$; then Et_3N, $-78\,^\circ C \rightarrow RT$, 90%; c) $MeOCH_2PPh_3Cl$, t-BuOK, THF, $0\,^\circ C \rightarrow RT$, 86%; d) 1. p-TsOH, dioxane/H_2O, 50 °C, 99%; 2. CH_3PPh_3Br, sodium hexamethyldisilazide (NaHMDS), $PhCH_3$, $0\,^\circ C \rightarrow RT$, 76%; e) $PhI(OCOCF_3)_2$, MeOH/THF, RT, 0.25 h, 92%.

The coupling of the two fragments, the all-critical first coupling, was achieved by a *B*-alkyl Suzuki carbon–carbon bond construction.[12] Hydroboration of the pre-acyl fragment **11** with 9-BBN furnished the mixed borane, which under the conditions indicated cross-coupled to iodoolefin **19** to give **20** in 71% yield. Upon cleavage of the acetal, aldehyde **21** was in hand.

With **21** we could explore the aggressive strategy of employing the methyl group of the C-1 bound acetoxy function as the nucleophilic component in a macroaldolization.[8] In the event, deprotonation was accomplished with potassium hexamethyldisilazide in THF at $-78\,^\circ C$. Remarkably, these conditions gave rise to a highly stereoselective macroaldolization, resulting in the formation of the C-3 (*S*)-alcohol **22** (Scheme 4). The heavy preponderance of **22** was favored when its precursor potassium aldolate was quenched at about 0 °C. When the aldolate was protonated at lower temperature, higher amounts of the C-3 (*R*) compound were detected. In fact, under some treatments, the C-3 (*R*) epimer predominates. This matter remains to be fully sorted out. At present, we are able to generate highly favorable C-3 (*R*):C-3 (*S*) ratios if the quenching is performed on an analytical scale. In preparative-scale experiments, we can reliably obtain **22** and its C-3 epimer in a 6:1 ratio.

The synthesis of the alkyl fragment started with commercially available (*R*)-glycidol **12**, which was converted, via its THP derivative **13**, into alcohol **14**. After cleavage of the tetrahydropyran blocking group, the resultant alcohol was smoothly converted into the methyl ketone **15**, as shown. The latter underwent an Emmons-type homologation with phosphane oxide **16**.[7, 10] The resultant alkyne **17** was then converted, via compound **18**, into *Z*-iodoalkene **19**[11] (Scheme 3).

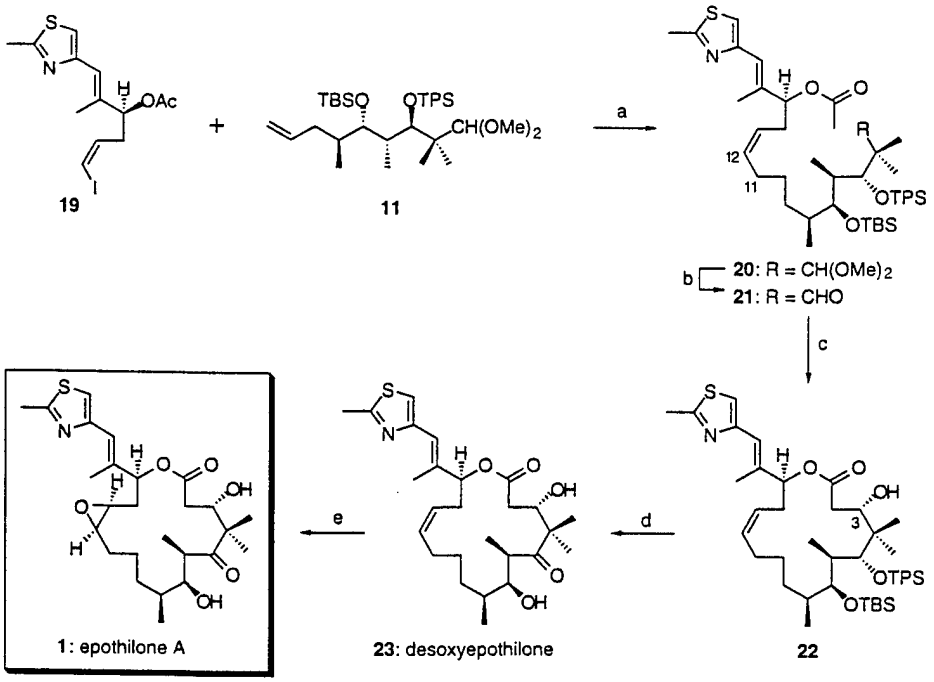


Scheme 3. a) Dihydropyran (DHP), pyridinium p-toluenesulfonate (PPTS), CH_2Cl_2, RT; b) 1. $Me_3SiCCLi$, $BF_3\cdot OEt_2$, THF, $-78\,^\circ C$; 2. methoxymethyl chloride (MOMCl), iPr_2NEt, $Cl(CH_2)_2Cl$, 55 °C; 3. PPTS, MeOH, RT; c) 1. $(COCl)_2$, DMSO, CH_2Cl_2, $-78\,^\circ C$, then Et_3N, $-78\,^\circ C \rightarrow RT$; 2. MeMgBr, Et_2O, $0\,^\circ C \rightarrow RT$; 3. tetra-*n*-propylammonium perruthenate (TPAP), *N*-methylmorpholine *N*-oxide (NMO), 4 Å mol. sieves, CH_2Cl_2, $0\,^\circ C \rightarrow RT$; d) **16**, nBuLi, THF, $-78\,^\circ C$, then **15**, THF, $-78\,^\circ C \rightarrow RT$; e) 1. *N*-iodosuccinimide, $AgNO_3$, $(CH_3)_2CO$; 2. Cy_2BH, Et_2O, AcOH; f) PhSH, $BF_3\cdot OEt_2$, CH_2Cl_2, RT; 2. Ac_2O, Py, 4-dimethylaminopyridine (4-DMAP), CH_2Cl_2, RT.

Having fashioned compound **22**, we could converge on our subgoal, desoxyepothilone (**23**). This objective was accomplished by selective removal of the triphenylsilyl (TPS) group in **22**, followed, sequentially, by selective silylation of the C-3 alcohol, oxidation of the C-5 alcohol, and, finally, fluoride-induced cleavage of the two silyl ethers.

Examination of a model based on the crystal structure of epothilone[1] suggested that the oxirane is disposed on the exterior face of the macrolide. In the event, oxidation of **23** was carried out with dimethyl dioxirane under the conditions shown. The major product of this reaction was (−)-epothilone A (**1**), whose identity was established by NMR and infrared spectroscopy, mass spectrometry, optical rotation, and chromatographic comparisons with authentic material kindly provided by Professor Höfle.[13]

While subject to further improvements, the synthesis in its present form, already provides us with workable amounts of epothilone A. More importantly, it provides routes to congeners not available from the natural product itself. The biological properties of some of these now accessible probe structures, as well as several of the chemical issues raised during the course of this synthesis, are receiving continuing attention.[14]



Scheme 4. a) **11**, 9-borabicyclo[3.3.1]nonane (9-BBN), THF, RT, then $PdCl_2(dppf)_2$, (dppf = 1,1'-bis(diphenylphosphino)ferrocene) $CsCO_3$, Ph_3As, H_2O, DMF, **19**, RT, 71%; b) TsOH, dioxane H_2O, 50 °C; c) KHMDS, THF, −78 °C, 51%; d) 1. HF·Py, Py, THF, RT, 97%; 2. *t*BuMe$_2$SiOTf, 2,6-lutidine, CH_2Cl_2, −25 °C, 93%; 3. Dess–Martin periodinane, CH_2Cl_2, 87%; 4. HF·Py, THF, RT, 99%; e) dimethyl dioxirane, CH_2Cl_2, 0.5 h, −50 °C, 45% (≥20:1).

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[13] In addition to epothilone A (**1**), small amounts of a diepoxide mixture, as well as traces of the diastereomeric *cis* C12–C13 monoepoxide (≥20:1) were detected.
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Total Synthesis of (−)-Epothilone B: An Extension of the Suzuki Coupling Method and Insights into Structure–Activity Relationships of the Epothilones**

Dai-Shi Su, Dongfang Meng, Peter Bertinato, Aaron Balog, Erik J. Sorensen, Samuel J. Danishefsky,* Yu-Huang Zheng, Ting-Chao Chou, Lifeng He, and Susan B. Horwitz

Recently, synthetic studies directed to epothilone A (**3**)[1, 2] culminated in its first total synthesis.[3–5] Our synthesis passed through the *Z*-desoxy compound (**4**), which underwent highly stereoselective epoxidation with 2,2-dimethyldioxirane, under carefully defined conditions, to yield the desired β-epoxide. The same myxobacterium of the genus *Sorangium* that produces **3** also produces epothilone B (**1**). The latter is significantly more potent than **3** both in antifungal screens and in cytotoxicity assays in some cell lines.[6, 7] Clearly then, there was a strong rationale for preparing epothilone B (**1**).

Our interim goal structure was desoxyepothilone B (**2**) or a suitable derivative thereof. With access to such a compound, we could investigate the regio- and stereoselectivity of the epoxidation of the C12–C13 double bond. Not the least interesting issue in the project was the synthesis of *Z*-trisubstituted olefinic precursors of **2** with high margins of stereoselection. In our synthetic route to epothilone A[3] we had employed a palladium-mediated B-alkyl Suzuki coupling[8, 9] of the *Z*-vinyl iodide **5** with borane **7** derived from hydroboration of compound **6** with 9-BBN (Scheme 1).

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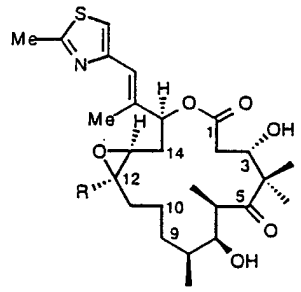
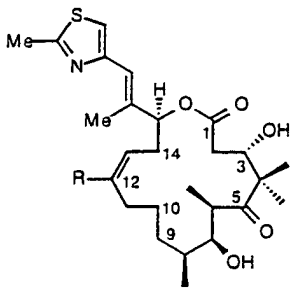
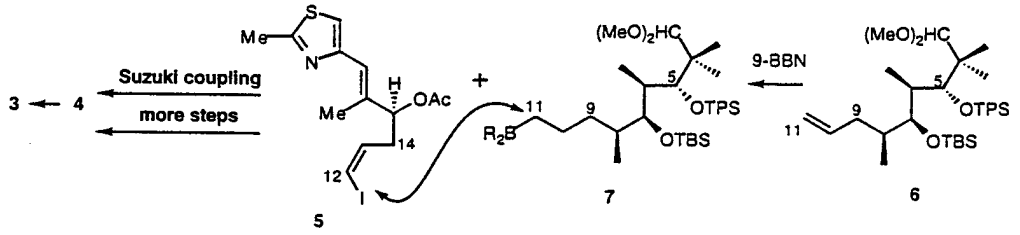


1: R = Me, epothilone B
3: R = H, epothilone A



2: R = Me, X = desoxyepothilone B
4: R = H, X = desoxyepothilone A



Scheme 1.

Naturally, our first instinct was to apply the same line of thinking to reach a *Z*-trisubstituted olefin en route to **2**. Here, two serious issues had to be addressed. First, it would be necessary to devise a method to prepare vinyl iodide **8**, the trisubstituted analog of **5**. If this goal could be accomplished, we would still face the challenge of conducting the required B-alkyl Suzuki coupling reaction to reach a *Z*-trisubstituted olefin. Such an intermolecular transformation, with a B-alkyl as opposed to a B-alkenyl group, and where the vinyl iodide is not partof a β-iodoenoate (or β-iodoenone), was not precedented.[10]

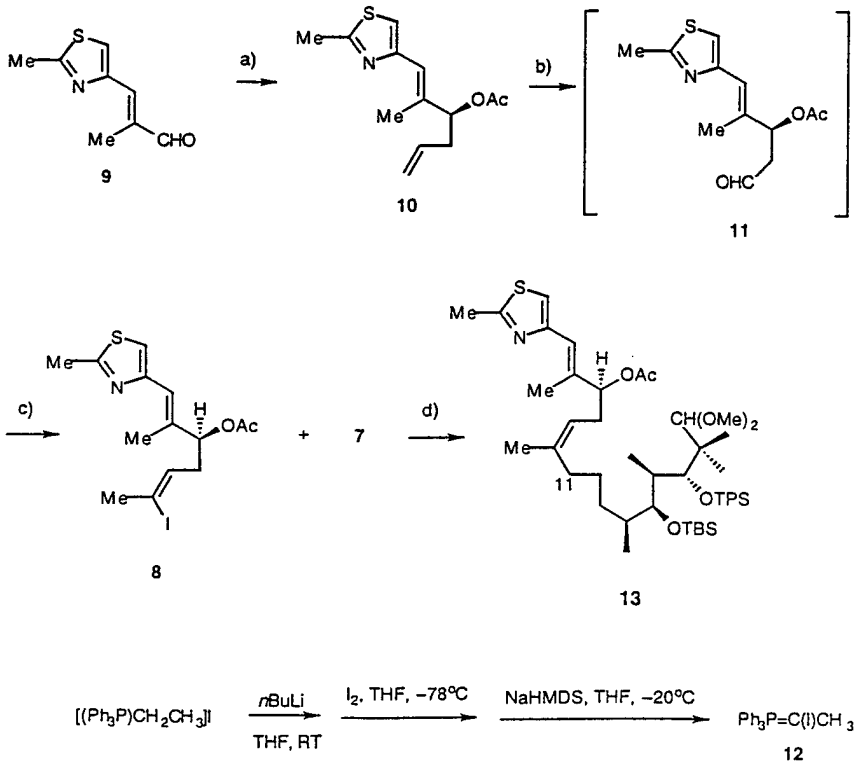
We first dealt with the synthesis of compound **8** (Scheme 2). Our route started with olefin **10**, which was prepared[4] by catalytic asymmetric allylation of **9**[11] followed by acetylation. Site-selective dihydroxylation of **10** followed by oxidative cleavage of the glycol generated the unstable aldehyde **11**. Remarkably, **11** reacted with phosphorane **12**[12] (preparation shown at the bottom of Scheme 2) to afford the *Z*-iodide **8** albeit in modest overall yield. Borane **7** was generated from **6** as previously described.[3] Gratifyingly, the coupling of compound **7** and iodide **8** could be conducted to produce the pure *Z*-olefin **13**.

With compound **13** in hand, we apply protocols similar to those employed in the synthesis of **4**.[3] Thus, hydrolysis of the acetal linkage led to aldehyde **14**, which was now subjected to macroaldolization (Scheme 3). The highest yield of aldol product was obtained by carrying out the reaction under conditions that produced a 1.5:1 mixture of α/β epimers at C3. Fortunately, we could convert the 3*R* isomer to the required 3*S* epimer by reduction of its derived C3 ketone (→**15**).[13] Cleavage of the C5 triphenylsilyl ether was followed sequentially by monoprotection (*tert*-butyldimethylsilyl) of the C3 hydroxyl group, oxidation at C5 (→**16**) and, finally, cleavage of the silyl protecting groups to expose the C3 and C7 hydroxyl groups (→**2**).

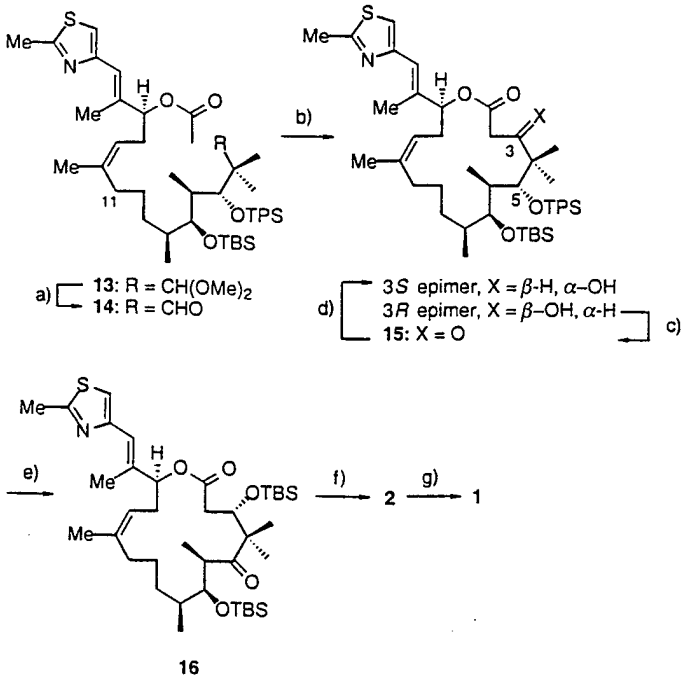
Z-Desoxyepothilone B (**2**) does indeed undergo very rapid and substantially regio- and stereoselective epoxidation[14] to afford epothilone B (**1**), which is identical with an authentic sample (^{1}H NMR, MS, IR , $[\alpha]_D$). Thus, the first total synthesis of (−)-epothilone B has been accomplished.

In retrospect, the total synthesis carries with it several important teachings. Certainly, the stereospecific preparation of the vinyl iodide **8** by use of the rarely employed phosphorane **12**, even with the unstable β-acetoxyaldehyde **11** could not have been predicted.[12] Moreover, the successful Suzuki coupling to construct the trisubstituted *Z*-double bond constitutes an important extension of the prior art.[10, 15] Finally, the highly regio- and stereoselective epoxidation of *Z*-desoxyepothilone B (**2**) with dimethyldioxirane was highly gratifying.

With epothilone B (**1**) and its *Z*-12,13-desoxy precursor (**2**) in hand through total synthesis, we were in a sound position to explore their biological activities. Also available to us from our previous studies[3, 4] were epothilone A (**3**), its *Z*-12,13-desoxy precursor **4**, and the *E* variant (**17**) of the latter. Through an alternative

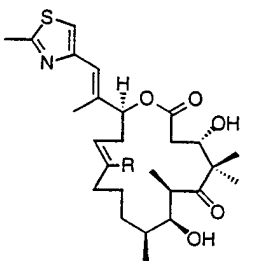


Scheme 2. a) 1. Allyl(tributyl)tin, (*S*)-(−)-2,2'-dihydroxy-1,1'-biphenyl, $Ti(OiPr)_4$, CH_2Cl_2, −20 °C, 60%, >95% *ee*; 2. Ac_2O, Et_3N, 4-dimethylaminopyridine (DMAP), CH_2Cl_2, room temperature (rt) 95%; b) 1. OsO_4, *N*-methylmorpholine *N*-oxide, acetone/H_2O, 0 °C; 2. $Pb(OAc)_4$, C_6H_6, 0 °C; c) **12**, THF, −20 °C, *Z* isomer only, 43% from **10**; d) $[Pd(dppf)_2]$ (dppf = 1,1'bis(diphenylphosphano)-ferrocene), Cs_2CO_3, Ph_3As, H_2O, DMF, rt 77%. THP = tetrahydropyran; TPS = triphenylsilyl.



Scheme 3. a) *p*-TsOH, dioxane/H_2O, 55 °C, 71%; b) potassium bis(trimethylsilyl)-amide (KHMDS), THF, −78 °C, 67%, α/β: 1.5:1; c) Dess–Martin periodinane, CH_2Cl_2, rt; d) $NaBH_4$, MeOH, rt, 80% for two steps; e) 1. HF·pyridine, pyridine, THF, rt, 93%; 2. TBSOTf, 2,6-lutidine, CH_2Cl_2, −30 °C, 89%; 3. Dess–Martin periodinane, CH_2Cl_2, rt, 67%; f) HF·pyridine, THF, rt, 80% g) dimethyldioxirane, CH_2Cl_2, −50 °C, 70%, 14:1 ratio of *cis* epoxides. TBS = *tert*-butyldimethylsilyl; OTf = trifluoromethanesulfonate.

route we also gained access to the *E* compound in the B series (**18**).[16] All of these compounds manifested the ability to bind to microtubules in the absence of guanosine triphosphate (GTP), biological activity reminiscent of that of Taxol™.[17] They also manifest high levels of cell killing as seen in Table 1. By the cytotoxicity standard, the trisubstituted compounds **1** and **2** outperform their disubstituted counterparts **3** and **4**. The desoxy compounds **2** and **4** are comparable and, in some instances, superior to the natural products (**1** and **3**). In this regard it is interesting that though **18** is still quite active, it is significantly less potent than the corresponding *Z* systems **2** and **4**, and the disubstituted *E* system **17**.



R = H, *trans*- desoxyepothilone A **17**
R = Me, *trans*- desoxyepothilone B **18**

Table 1. Relative efficacy of epothilone compounds against drug-sensitive and resistant CCRF-CEM cell lines [a].

Compound	CCRF-CEM IC_{50} [μM] [b]	CCRF-CEM/VBL IC_{50} [μM] [b]	CCRF-CEM/VM_1 IC_{50} [μM] [b]
epothilone A (**3**)	0.003	0.020	0.003
desoxyepothilone A (**4**)	0.022	0.012	0.013
trans-desoxy A **17**	0.052	0.035	0.111
epothilone B (**1**)	0.0004	0.003	0.002
desoxyepothilone B (**2**)	0.009	0.017	0.014
trans-desoxy B **18**	0.090	0.262	0.094
Taxol™	0.002	3.390	0.002

[a] The cytotoxicities of test compounds were determined by the growth of human lymphoblastic leukemic cells CCRF-CEM or their sublines resistant to vinblastine and taxol (CCRF-CEM/VBL) or resistant to etoposide (CCRF-CEM/VM-1). XTT-microculture tetrazolium/formazan assays [18] were used. [b] The IC_{50} values were calculated based on five or six assays at various concentrations; a median-effect plot [19] was generated with computer software [20].

All proposals addressed to the pharmacological modeling of the epothilones must now take notice of these interesting structural findings. Certainly they are of significant consequence in our own analog strategy program, which is well underway.

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Keywords: antitumor agents · epothilone · natural products · total synthesis

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[13] The stereochemically controlled aldol condensation giving a product with the natural 3*S* configuration as previously described in the epothilone A series [3] was accomplished. However, the overall yield for reaching the 3*S* epimer is better with the protocol described here.
[14] Although precise comparisons are not available, the epoxidation of **2** appears to be more rapid and regioselective than that of **4** [3].
[15] To the best of our knowledge, this is the first successful Suzuki coupling between a B-alkyl system and an unconjugated, trisubstituted alkene.
[16] Compound **18** was prepared by a stereorandom olefin metathesis route which also led to **2**. These geometric isomers could be separated only with great difficulty. Details of the nonstereoselective route will be provided in a subsequent fuller disclosure.
[17] Experiments conducted by Dr. Susan Horwitz at the Albert Einstein College of Medicine, Bronx, NY (USA): Microtubule protein (MTP) was purified from calf brains by two cycles of temperature-dependent assembly and disassembly [21]. In control assembly experiments, MTP (1 mg mL^{-1}) was diluted in assembly buffer containing 0.1 M MES (2-(*N*-morpholino)ethanesulfonic acid), 1 mM EGTA (1,2-di(2-aminoethoxy)ethane-*N*,*N*,*N*′,*N*′-tetraacetic acid), 0.5 mM $MgCl_2$, 1 mM GTP, and 3 M glycerol, pH 6.6. The concentration of tubulin in MTP was estimated to be about 85%. Assembly was followed spectrophotometrically at 350 nm, 35 °C for 40 min, by monitoring changes in turbidity as a measure of polymer mass [22]. Drugs were tested at a concentration of 10 μM in the absence of GTP. Microtubule formation was verified by electron microscopy. To determine the stability of microtubules assembled in the presence of GTP or drug, turbidity was monitored for 40 min after the reaction temperature was shifted to 4 °C.
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Studies toward a Synthesis of Epothilone A: Use of Hydropyran Templates for the Management of Acyclic Stereochemical Relationships

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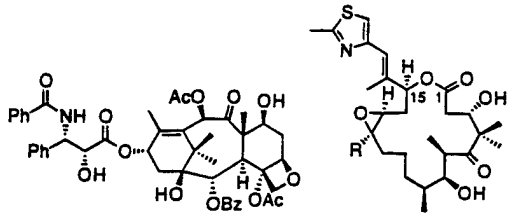
Taxol has been approved for chemotherapeutic clinical application against ovarian carcinomas. It is also undergoing extensive evaluation for other indications. While taxol is not a curative agent, it is already a useful chemotherapeutic resource.[1]

The best indications arising from tissue culture and *in vitro* experiments are that taxol functions by inhibition of cellular mitosis through binding to and stabilization of microtubule assemblies.[2] Presumably, this property is pertinent to the human patient.

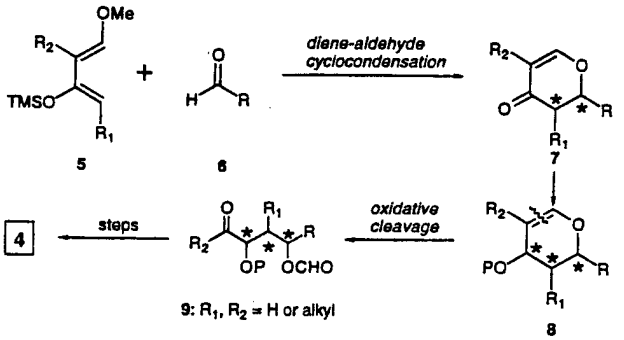
Unfortunately, taxol is far from an ideal drug. Thus, difficulties with respect to formulation and susceptibility to multiple drug resistance (MDR) complicate its applicability.[3] At the present writing, no major improvements in drug performance have been realized from any substantially modified analogs of taxol or its close relative, taxotere.[4]

New agents that function by microtubule stabilization are clearly of great interest.[2] In this connection, there has already been considerable attention directed toward the bacterial-derived metabolites epothilone A (**2**) and B (**3**), which were first identified as antifungal cytotoxic agents by Höefle *et al.*[5a,b] and subsequently encountered by a group based at the Merck corporation.[6] The report of the Merck scientists on the epothilones indicated that they are powerful cytotoxic agents that seem to function through stabilization of microtubules by binding to taxol-binding domains. Given the possibilities that these agents themselves, or appropriately modified derivatives, might function as alternatives to taxol, attention from the standpoint of organic synthesis is warranted.

Augmenting the biological rationale for such a venture are the chemical incentives associated with several novel structural features of the epothilones. Thus, the presence of a thiazole moiety, as well as a *cis* epoxide and geminal dimethyl groups are among the issues to be addressed. Not the least intriguing feature is the array of three contiguous methylene groups that serves to insulate the two functional domains of the molecules. This achiral "spacer element" actually complicates prospects for continuous chirality transfer and seems to call for a strategy of merging two stereochemically committed substructures. Herein, we direct our attention to a synthesis of compound **4**, confident that, in principle, such a structure could be converted to the epothilones themselves, and to related screening candidates.



The identification of compound **4** as a synthetic intermediate provided an opportunity to illustrate the power of hydropyran matrices in addressing problems associated with the control of stereochemistry in acyclic intermediates. Some years ago, we described the synthesis of dihydropyrones through what amounts to overall cyclocondensation of suitably active dienes and aldehydic heterodienophiles.[7]



High margins of stereoselectivity can be realized in assembling such matrices (*cf.* **5** + **6** → **7**). Moreover, the hydropyran platforms service various stereoselective reactions (see formalism **7** → **8**). Furthermore, the products of these reactions are amenable to ring-opening schemes, resulting in the expression of acyclic fragments with defined stereochemical relationships (*cf.* **8** → **9**).[8]

We describe the application of two such routes for the synthesis of compound **4**. Route 1, which does not *per se* involve control over the issue of absolute configuration, commences with the known aldehyde **10**.[9] Homologation, as shown, provided enal **12**. Cyclocondensation of **12** with the known diene,[10] under BF_3 catalysis, led to racemic dihydropyrone **13**. Luche reduction[11] of **13** provided compound **14**. At this point we were well positioned to take advantage of our previously introduced lipase methodology for resolution of glycal derivatives through enzymatically mediated kinetic resolution.[12] Thus, carbinol **14** was treated with lipase 30 and isopropenyl acetate (following the prescriptions of Wong),[13] and

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‡ Columbia University.

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(2) Schiff, P. B.; Fant, J.; Horwitz, S. B. *Nature (London)* **1979**, *277*, 665.
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(7) Danishefsky, S. J. *Aldrichim. Acta* **1986**, *19*, 59.
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(9) (a) Shafiee, A.; Mazloumi, A.; Cohen, V. I. *J. Heterocycl. Chem.* **1979**, *16*, 1563. (b) Schafiee, A.; Shahocini, S. *J. Heterocycl. Chem.* **1989**, *26*, 1627.
(10) Danishefsky, S. J.; Kitahara, T. *J. Am. Chem. Soc.* **1974**, *96*, 7807.
(11) Luche, J.-L. *J. Am. Chem. Soc.* **1978**, *100*, 2226.

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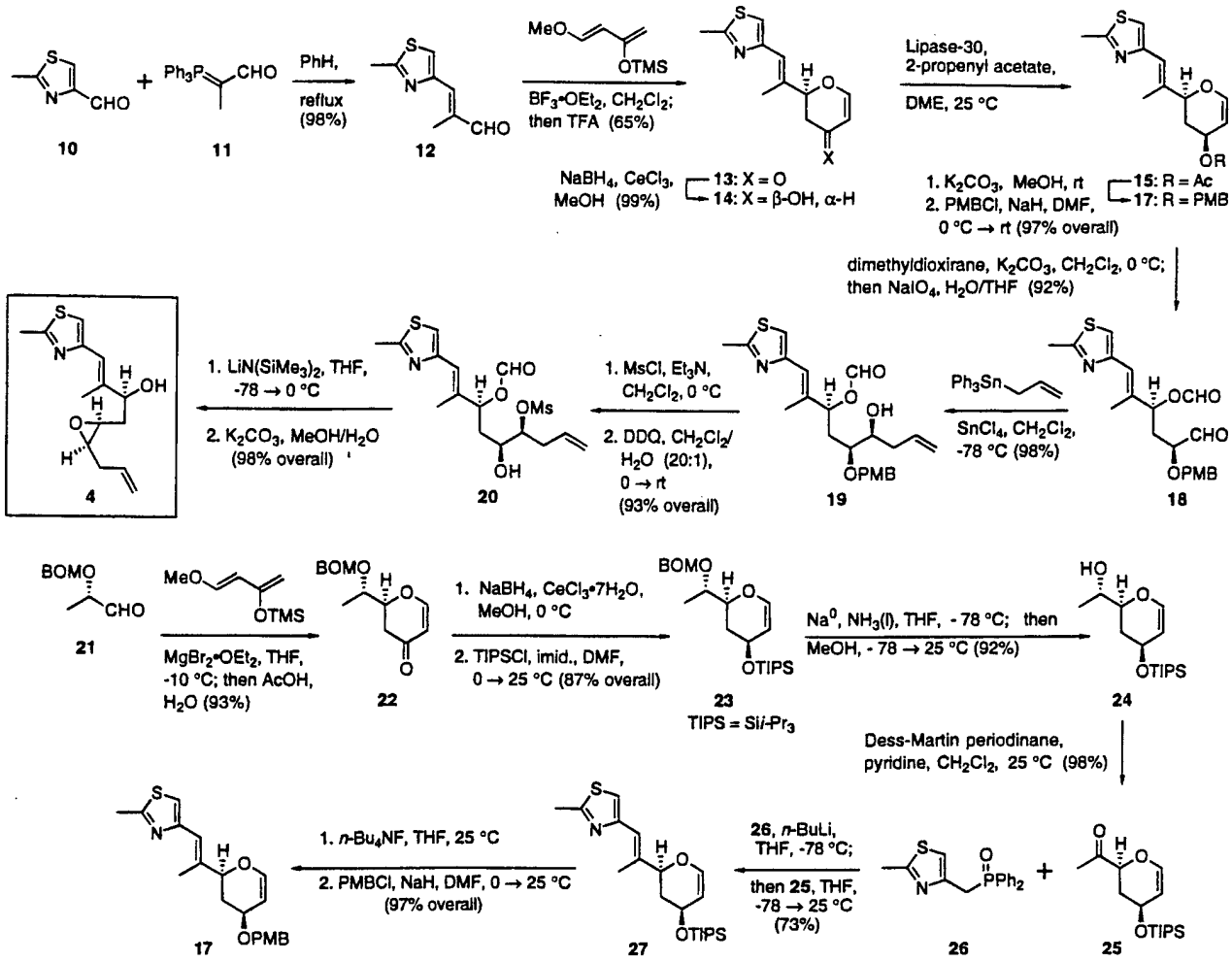

the reaction was stopped after *ca.* 48% conversion, providing acetate **15** in addition to the enantiomerically related free glycal **16**. Compound **15** was further advanced to the *p*-methoxybenzyl (PMB)-protected system **17**. At this juncture, reaction of **17** with dimethyldioxirane[14] generated an intermediate (presumably the corresponding glycal epoxide) that, upon treatment with sodium metaperiodate, gave rise to aldehyde formate **18**. Lewis acid-promoted allylation of **18** afforded carbinol **19** in which the formate ester had nicely survived. Unfortunately, **19** was accompanied by its *anti* stereoisomer (not shown here) (4:1). Although this 4:1 mixture of diastereomers was obtained in an excellent yield of 98%, it was not possible to obtain the desired stereoisomer **19** in pure form at this stage. Mesylation of the secondary alcohol, followed by deprotection (see **19** → **20**) and cyclization, as indicated, gave compound **4**, a substance that could be separated from the stereoisomeric *trans* epoxide.

Needless to say, in this synthesis, only *ca.* half of the dihydropyrone was secured through the process of kinetic resolution. While, in theory, several of our synthetic strategems contemplate the possible use of each enantiomer of **15** to reach epothilone itself, we sought to implement another route to allow for full enantiomeric convergence. The logic of this route is that the chirality of a "dummy" stereogenic center is communicated to the emerging pyran following previously established principles of tunable diastereoselection in the cyclocondensation reaction.[7,8] We proceeded as follows. Cyclocondensation of lactaldehyde derivative **21**[15] with the indicated diene, under ostensible chelation control, afforded **22**. The side chain ether could then be converted to the methyl ketone **25** as shown (see **22** → **23** → **24** → **25**). Finally, an Emmons condensation of **25** with the phosphine oxide **26**[16] as shown in Scheme 4 afforded compound **27** as a single geometrical isomer. A straightforward protecting group adjustment then afforded the previously encountered **17**. This route cogently illustrates the concept of stereochemical imprinting through a carbon center that eventually emerges in planar form after conferring enantioselection to subsequently derived stereocenters. The use of the dihydropyrone-based logic for securing the stereochemical elements of the epothilones, as well as the identification of a possible strategy for macrocyclization will be described in the paper that follows.

Acknowledgment. This work was supported by NIH Grant No. CA 28824. Postdoctoral Fellowships are gratefully acknowledged by E.J.S. (NSF, CHE-9504805) and P.B. (NIH, CA 62948).

Supporting Information Available: Experimental procedures and spectroscopic data for the compounds illustrated in the reactions (compounds **12**–**15**, **17**–**20**, **4**, and **22**–**27**) (9 pages).

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(15) Heathcock, C. H.; Young, S. D.; Hagen J. P.; Pirrung, M. C. White, C. T.; VanDerveer, D. *J. Org. Chem.* **1980**, *45*, 3846.

(16) The known 2-methyl-4-chloromethylthiazole (see: Marzoni, G. *J. Heterocycl. Chem.* **1986**, *23*, 577) was transformed to **26** (*cf.* Toh, H. T.; Okamura, W. H. *J. Org. Chem.* **1983**, *48*, 1414.

J. Org. Chem. 1996, 61, 8000–8001

Studies toward a Synthesis of Epothilone A: Stereocontrolled Assembly of the Acyl Region and Models for Macrocyclization

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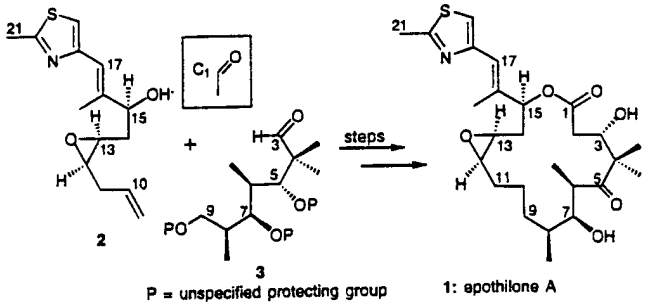
Received August 30, 1996

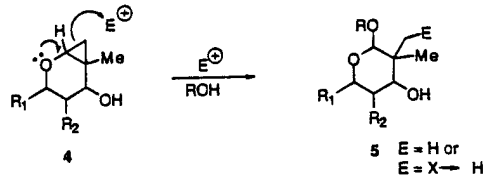
In our previous paper, we described a synthesis of the "alkoxy" segment of epothilone A **1** (see compound **2**, Scheme 1) encompassing carbons 10–21.[1] In this paper, we address the synthesis of another fragment encoding the stereochemical information of acyl section carbons 3–9. It was envisioned that the aldehydo center (C_3) of the formal target **3** would serve as an attachment site to a nucleophilic construct derived from compound **2** (requiring placement of a two-carbon insert, as suggested in Scheme 1), through either inter- or intramolecular means. In such a context, it would be necessary to deal independently with the stereochemistry of the secondary alcohol center eventually required at C_3. One of the interesting features of system **3** is the presence of geminal methyl groups at carbon 4 (epothilone numbering). It was our hope to again use a dihydropyran strategy to assemble a cyclic matrix corresponding, after appropriate disassembly, to a viable equivalent of system **3**. *We hoped to expand upon our dihydropyran paradigm to include the synthesis of gem dimethyl containing cyclic and acyclic fragments.* The particular reaction type we had in mind for this purpose is generalized under the heading of transformation of **4** → **5** (see Scheme 2). At this juncture, we deliberately avoid commitment as to the nature of the electrophile, E. Accordingly, we leave for the moment unaddressed the question as to whether a reduction would or would not be necessary in going from structure type **5** to reach the intended generalized target **3**.

Once again, our opening step consisted of a stereochemically tunable version of the diene–aldehyde cyclocondensation reaction[2] (Scheme 3)—in this instance drawing upon chelation control in the merger of the readily available enantiomerically homogeneous aldehyde **6** with the known diene **7**.[3] Indeed, as precedent would have it, under the influence of titanium tetrachloride there was produced substantially a single isomer shown as compound **8**.[4] In the usual and stereochemically reliable way,[5] the dihydropyrone was reduced to the corresponding glycal **9**. At this point, we utilized a directed Simmons–Smith reaction for the conversion of glycal **9** to cyclopropane **10**.[6] This compound is indeed an interesting structure in that it corresponds in one sense to a cyclopropano version of a *C*-glycoside. At the same time, the cyclopropane is part of a cyclopropylcarbinyl alcohol system with attendant possibilities for rearrangement.[7] It was our intention to cleave the *C*-glycosidic bond of the cyclopropane in a fashion that would elaborate the geminal methyl groups, leaving in its wake a solvent-derived glycoside with the desired aldehyde oxidation state at C-3 (see hypothesized transformation **4** → **5**, Scheme 2). In early efforts, the nonoxidative version of the projected reaction (*i.e.*, $E^+ = H^+$) could not be reduced to practice. Instead, products clearly attributable to the ring-expanded system **11**[8] were identified.

Scheme 1. Convergent Strategy for a Total Synthesis of Epothilone A (1)



Scheme 2. Glycal Cyclopropane Solvolysis Strategy for the Introduction of Geminal Methyl Groups



Fortunately, however, the desired sense of cyclopropane opening, under the influence of the ring oxygen, was achieved by subjecting compound **10** to oxidative opening with *N*-iodosuccinimide.[9] The intermediate iodomethyl compound, obtained as a methyl glycoside **12**, when exposed to the action of tri-*n*-butyltin hydride, gave rise to pyran **13** containing the geminal methyl groups. Protection of this alcohol (see **13** → **14**), followed by cleavage of the glycosidic bond, revealed the acyclic dithiane derivative **15** which can serve as a functional version of the hypothetical aldehyde **3**.

We have also begun to explore possible ways of combining fragments relating to **2** and **3** in a fashion to reach epothilone and congeners thereof. Mindful of the pio-

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‡ Columbia University.

(1) Meng, D.; Sorensen, E. J.; Bertinato, P.; Danishefsky, S. J. *J. Org. Chem.* **1996**, *61*, 7998–7999.
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(3) Danishefsky, S. J.; Yan, C.-F.; Singh, R. K. Gammill, R. B.; McCurry, P. M., Jr.; Fritsch, N.; Clardy, J. *J. Am. Chem. Soc.* **1979**, *101*, 7001.
(4) (a) Danishefsky, S. J.; Pearson, W. H.; Harvey, D. F.; Maring, C. J.; Springer, J. P. *J. Am. Chem. Soc.* **1985**, *107*, 1256. (b) Danishefsky, S. J.; Myles, D. C.; Harvey, D. F. *J. Am. Chem. Soc.* **1987**, *109*, 862.
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(8) For example, exposure of **10** to acidic methanol gave rise to an epimeric mixture of seven- membered mixed acetals, presumably through the addition of methanol to oxocarbenium ion **11**. This most interesting transformation is under active study in our laboratory.
(9) For interesting Hg(II)-induced solvolyses of cyclopropanes that are conceptually similar to the conversion of **10** to **12**, see: (a) Collum, D. B.; Still, W. C.; Mohamadi, F. *J. Am. Chem. Soc.* **1986**, *108*, 2094. (b) Collum, D. B.; Mohamadi, F.; Hallock, J. S. *J. Am. Chem. Soc.* **1983**, *105*, 6882. Following this precedent, we did, in fact, accomplish a Hg(II)-induced solvolysis of cyclopropane **10**, although this transformation proved to be less efficient than the reaction shown in Scheme 3.

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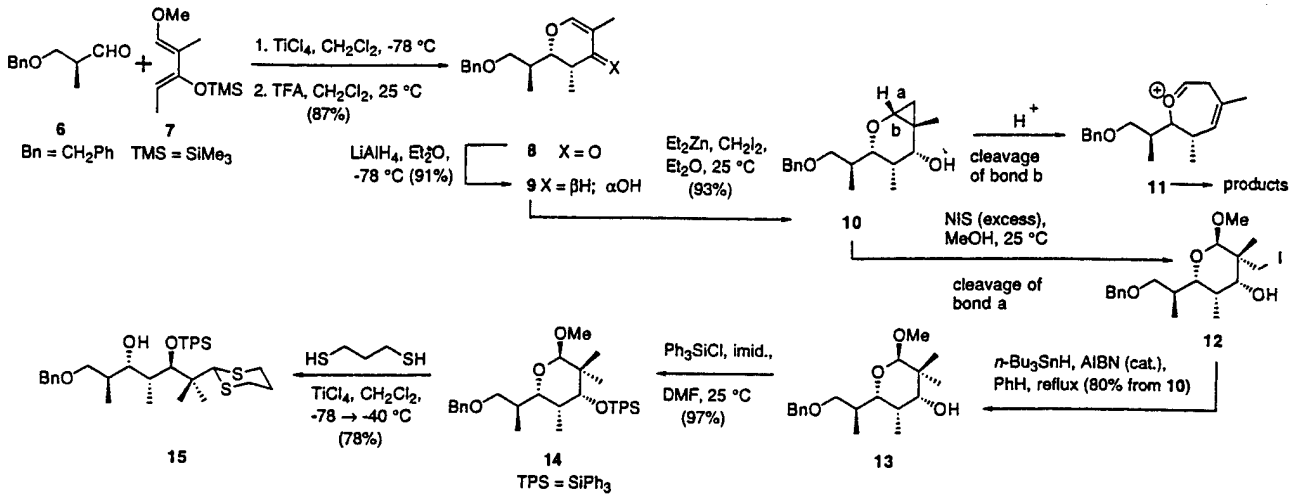
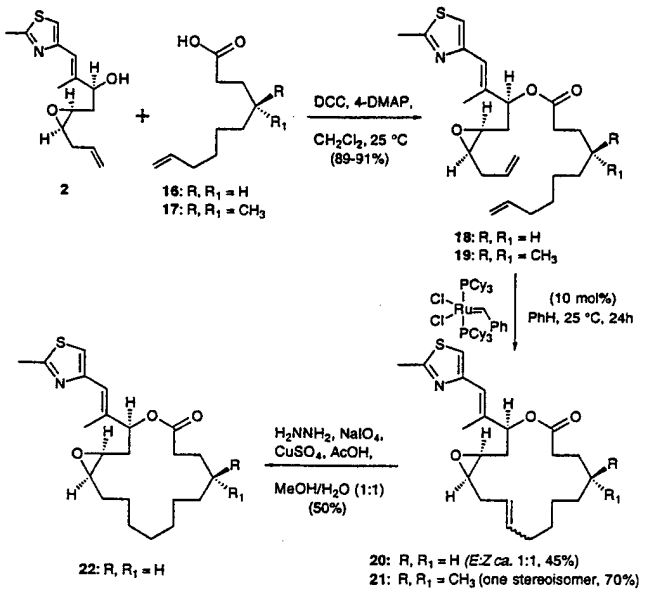
Scheme 3. Enantioselective Synthesis of Compound 15



Scheme 4. Construction of Epothilone Model Systems 20–22 by Ring-Closing Olefin Metathesis



neering studies of Schrock[10a] and Grubbs[10b] and the recent ground-breaking disclosure of Hoveyda,[11] we wondered about the possibility of realizing such an approach *en route* to our goal.[12] The matter was first examined with two model ω-unsaturated acids **16** and **17** that were used to acylate alcohol **2** to provide esters **18** and **19**, respectively (see Scheme 4). These compounds did indeed undergo olefin metathesis macrocyclization in the desired manner under the conditions shown. In the case of substrate **18**, **20** was obtained as a mixture of *E*- and *Z*- stereoisomers (*ca.* 1:1). Diimide reduction of **20** was then conducted to provide homogeneous **22** in 50% yield. The olefin metathesis reaction was also extended to compound **19** bearing geminal methyl groups corresponding to their placement at C4 of epothilone A. Once again, olefin metathesis occurred, this time curiously producing olefin **21** as a single entity in 70% yield (stereochemisty tentatively assigned as *Z*). Substantially identical results were obtained through the use of Schrock's molybdenum alkylidene metathesis catalyst.

Having shown that olefin metathesis is equal, in principle, to the challenge of constructing the 16-membered ring containing both the required epoxy and thiazolyl functions of our target system, we have started to project a synthesis of epothilone A itself. Clearly, with these fragments in hand, a variety of strategies for their combination, culminating in either carbon–carbon bond formation or macrolactonization, can be entertained, and these are being evaluated. At the present writing, however, it is appropriate to point out that no successful olefin metathesis reaction has yet been realized from *seco*-systems bearing a full compliment of functionality required to reach epothilone. These negative outcomes may merely reflect a failure to identify, as yet, a suitable functional group constraint pattern appropriate for macrocylization.[13] Many possibilities remain to be screened. Accordingly, intramolecular olefin metathesis is still included in a variety of ring-forming options currently being evaluated for reaching epothilone A.

Acknowledgment. This work was supported by NIH Grant No. CA 28824. Postdoctoral Fellowships are gratefully acknowledged by E.J.S. (NSF, CHE-9504805) and P.B. (NIH, CA 62948). We gratefully acknowledge Dr. George Sukenick (NMR Core Facility, Sloan-Kettering Institute) for NMR and mass spectral analyses.

Supporting Information Available: Experimental procedures and spectroscopic data for the compounds illustrated in the schemes (compounds **8**–**10**, **12**–**16**, and **18**–**22**) (6 pages).

JO961674O

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(11) Houri, A. F.; Xu, Z.; Cogan, D. A.; Hoveyda, A. H. *J. Am. Chem. Soc.* **1995**, *117*, 2943.

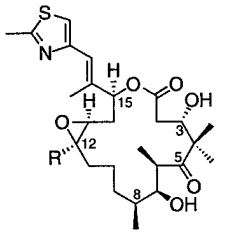
(12) For recent examples of ring-closing metathesis, see: (a) Martin, S. F.; Chen, H.-J.; Courtney, A. K.; Liao, Y.; Pätzel, M.; Ramser, M N.; Wagman, A. S. *Tetrahedron* **1996**, *52*, 7251. (b) Fürstner, A.; Langemann, K. *J. Org. Chem.* **1996**, *61*, 3942.

(13) Substrates containing the full complement of oxygenated functionality, including the trisubstituted olefin and thiazolyl moiety, were screened for ring-closing olefin metathesis. In an effort to favor ring closure through the rigidification of the carbon backbone, a *seco* structure possessing a cyclic isopropylidene ketal bridging a C3–C5 diol relationship was prepared and subjected to ring-closing metathesis. In one instance, we also screened a substrate containing functionality that would lead to the C12–C13 epoxide, but lacking this function, *per se*. In spite of these setbacks, efforts to fashion the macrolide of epothilone A through ring-closing metathesis are continuing A full account of these studies will be disclosed in due course.

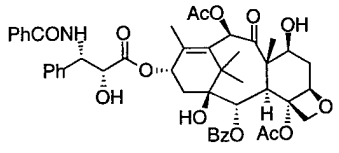
Structure–Activity Relationships of the Epothilones and the First In Vivo Comparison with Paclitaxel**

Dai-Shi Su, Aaron Balog, Dongfang Meng, Peter Bertinato, Samuel J. Danishefsky,* Yu-Huang Zheng, Ting-Chao Chou, Lifeng He, and Susan B. Horwitz

The discovery and characterization of the bacterial natural products epothilones A (**1**) and B (**2**) have evoked a great deal of interest.[1] Their high levels of cytotoxicity and their potent stabilization of microtubules are reminiscent of the biological activity of paclitaxel (**3**), a clinically valuable resource in cancer



1, R = H: epothilone A
2, R = Me: epothilone B



3: paclitaxel

chemotherapy.[2] Paclitaxel (**3**), which has been in use for the treatment of ovarian and breast carcinomas, is also being evaluated against a variety of other tumors. Nonetheless, its application is hampered by difficulties in formulation and susceptibility to multiple drug resistance (MDR). Though a massive amount of analog synthesis based on the paclitaxel structure has been accomplished, the framework appears to be intolerant to major simplification with maintenance of biological activity. To the best of our knowledge, no compound which is of a significantly lower level of structural complexity than paclitaxel has been of clinical interest as a replacement for the parent drug.

Since the epothilones are more water-soluble than paclitaxel and, in preliminary in vitro studies, seemed to perform better against several MDR cell lines, this series warranted evaluation. Our laboratory and several others have attacked the problem of the total synthesis of **1** and **2**.[3–9] This goal, in the case of **1**, was first accomplished in a stereocontrolled fashion by using a boron-alkyl Suzuki coupling strategy to establish the C11–C12

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[**] This research was supported by the National Institutes of Health (grants to S. J. D. (CA-28824) and S. B. H. (CA-39821) and postdoctoral fellowships to A. B. (CA-GM 72231) and P. B. (CA-62948)). We gratefully acknowledge Dr. George Sukenick (NMR Core Facility, Sloan-Kettering Institute) for NMR and mass spectrometric analyses. Professor Dr. G. Höfle, of the Gesellschaft für Biotechnologische Forschung, Braunschweig (Germany), is gratefully acknowledged for providing natural epothilone A and B for comparative analysis. We also thank Professor Gunda Georg of the University of Kansas, Lawrence, KS (USA), for bringing the epothilone problem to our attention.

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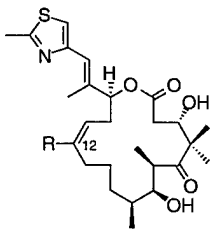
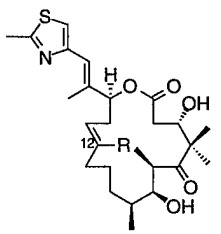
bond.[3c, e] Our total synthesis was followed by three reports of total syntheses involving nonstereoselective ring-forming olefin metathesis reactions to establish the C12–C13 bond.[3d, 4, 5] More recently, a stereoselective Wittig reaction to establish the C12–C13 bond en route to **1** has also been described.[4c]

The first synthesis of epothilone B (**2**) was accomplished in our laboratory, again, through the use of a highly stereoselective boron-alkyl Suzuki coupling reaction to form the trisubstituted olefin.[3e] A Wittig-based route, generating a mixture of olefin isomers leading to **2**, was recently reported.[4d] An important advance in the field was the stereocontrolled epoxidation of the C12–C13 double bond in desoxyepothilones A (**4**) and B (**5**) through the agency of 3,3-dimethyldioxirane.[3c–e]

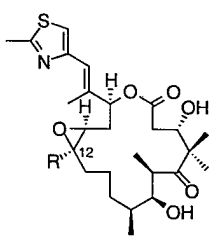
While none of our syntheses described thus far are practical for the full-scale pharmaceutical development of an epothilone, they have provided more than ample quantities for biological evaluations of the natural products both in vitro and in vivo, and for early evaluations of a range of fully synthetic analogs. Presently, we are examining compounds obtained by obvious explorations of key intermediates in the total syntheses of **1** and **2**. At this writing, we have not yet generated analogs derived from extended operations on the completed natural products themselves. We have focused on structure–activity relationships (SAR) to determine zones of the molecule that are tolerant or intolerant of structural change. Such insights would be valuable in focusing studies on molecular modeling. Furthermore, it was hoped to gather in vivo data for a direct comparison of epothilone B and paclitaxel.



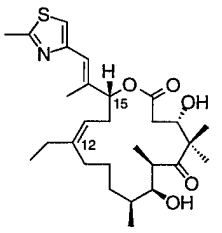
4, R = H
5, R = methyl
8, R = ethyl
10, R = propyl
11, R = 



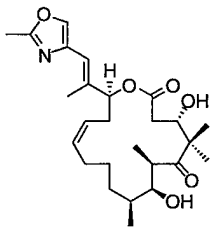
6, R = H
7, R = methyl
9, R = propyl



12, R = ethyl
13, R = propyl
14, R = hexyl



15, C15-epimer



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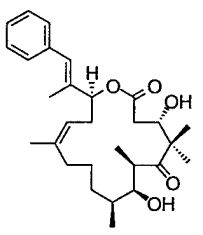

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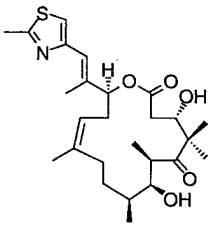

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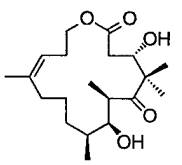

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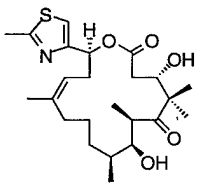

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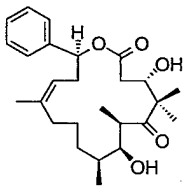

21

For the definition of the SAR of the epothilones, it is convenient to divide the structure of the drug into an acyl sector (shown arbitrarily as C1–C8), an O-alkyl sector (C9–C15), and a pendant aryl sector projecting from C15. In our preliminary SAR studies on epothilones we found that the acyl sector is rather intolerant of modification. For instance, inversion of stereochemistry at C3 ($S \rightarrow R$), or reduction at C5 results in serious abrogation of activity. Wholesale deletion of functionality at C3, C5, C6, C7, and C8 results in a loss of cytoxicity and of activity in the tubulin/microtubule system.[10] Indeed, as we recently demonstrated, a single permutation—deletion of the 8-methyl group—has a highly deleterious effect on biological function.[11] We now report that deletion of the "C9" methylene group (see the 15-membered macrolide **18**) also results in a major loss of activity in the tubulin polymerization/depolymerization assays, which have correlated rather closely with other indicators of biological function.[12]

Turning to the O-alkyl sector, we have expanded on the original[13] and subsequent findings to the effect that epothilone B (**2**) is more potent than epothilone A (**1**) with regards to tubulin polymerization/depolymerization and cytotoxicity (Figure 1 and Table 1). It was also found that the (*Z*)-desoxy compounds **4** and **5** retain a function closely comparable to their natural counterparts, **1** and **2**, respectively. Once again, the "B" precursor, compound **5**, is more active than the "A" precursor, compound **4**. Even the (*E*)-desoxy compounds **6** and **7** maintain significant biological activity, although less than that of the (*Z*) analogs, **4** and **5**, respectively.[3d, e, 4d] We now report that epothilone B is approximately 3400 times more active than paclitaxel against the resistant human leukemic cell line CCRF-CEM/VBL in cell-culture cytotoxicity studies.

The alkyl sector has been studied in greater detail.[14] Thus, substitution of an ethyl group at C12 in epothilone B is well tolerated (see compounds **8** and **12**, the ethyl versions of **5** and **2**, respectively) as are the higher homologues **10**, **13** and **14**.[15] Interestingly, biological activity is not lost in compound **11** bearing the polar acetal functionality on C12, nor in the *E*-desoxy compound **9**.[16] Further, activity is still retained, though in attenuated form, upon inversion of stereochemistry at C15 (see compound **15**). In summary, the O-alkyl sector is remarkably tolerant of modification with basic maintenance of in vitro function.

We also investigated the aryl sector.[14] Our studies indicate that this sector is also quite tolerant to permutation. For instance, the substitution of oxygen for sulfur was well tolerated. Thus, compound **16** (an oxazole rather than a thiazole) is equipotent with **4** in the tubulin polymerization assay (Figure 1)[17] as well as in cytotoxicity assays (Table 1).[18] We then studied a more drastic variant of the aryl domain with phenyl in place of the thiazolyl unit, and found that compound **17** retains 60% of the activity of **2** in the tubulin polymerization assay. Though there is a loss of one order of magnitude in the cytotoxicity assay, compound **17** is still highly cytotoxic.

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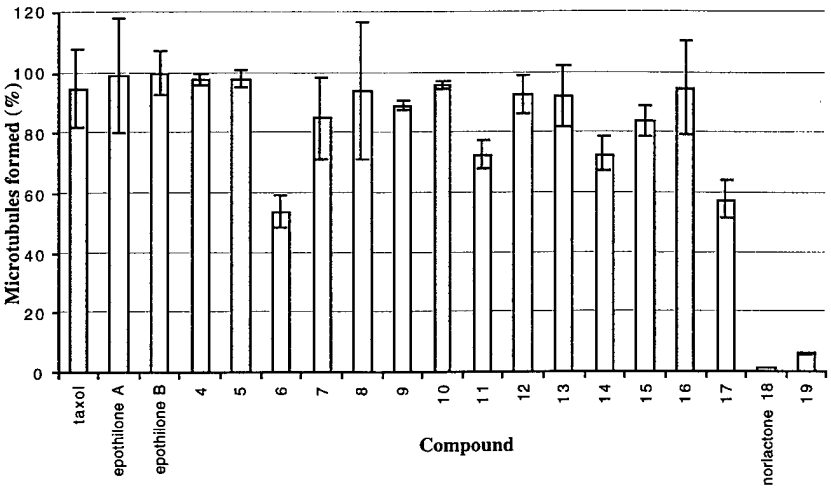


Figure 1. Formation of microtubules in the presence of 10 μM of the tested compounds. (Microtubules formed in the presence of 10 μM epothilone B defined as 100%.)

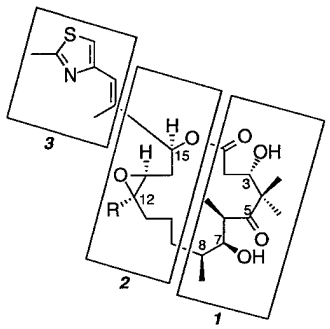
Table 1. Relative efficacy of epothilone compounds against drug-sensitive and -resistant CCRF-CEM cell lines[18].

Compound	CCRF-CEM IC_{50} [μM]	CCRF-CEM/VBL IC_{50} [μM]
paclitaxel (**3**)	0.002	4.140
epothilone A (**1**)	0.003	0.020
epothilone B (**2**)	0.0002	0.001
4	0.022	0.012
5	0.009	0.017
6	0.052	0.035
7	0.090	0.262
8	0.021	0.077
9	0.090	0.254
10	0.039	0.067
11	0.003	0.009
12	0.001	0.007
13	0.004	0.006
14	0.027	0.049
15	0.055	0.197
16	0.030	0.049
17	0.098	0.146
18	>10.0	8.95
19	>10.0	>10.0
20	3.52	1.20
21	1.80	>5.00

Given these data, we wondered whether the aryl sector is even necessary. Could it be replaced by a single hydrogen at C15? Toward this end, we prepared compound **19**. However, it has very low cytotoxicity (IC_{50} > 10.0 μM) and shows almost no activity in tubulin polymerization/depolymerization assays. We then asked whether the olefinic spacer element connecting the aromatic rings with C15 is needed. Therefore, we prepared compounds **20** and **21**. These substances showed a major loss of cytotoxicity. Accordingly, the aryl sector can now be defined with greater precision: it requires an olefinic spacer linking the lactone at C15 to an aromatic subsection which is substantially tolerant to modification.

Of course, it will be necessary to prepare and evaluate many more compounds to provide a definitive mapping of the SAR profile of this series with single carbon atom resolution. However, it is already clear that the acyl sector constitutes a "hot spot" with great sensitivity to structural change (Scheme 1). By contrast, the O-alkyl and aryl sectors exhibit significant tolerance, both in the tubulin assays and in cytotoxicity screens.

We have also initiated in vivo evaluations of the epothilones. As a first line of inquiry, we conducted a direct comparison of fully synthetic epothilone B (**2**) with paclitaxel (**3**). Since the susceptibility to drug resistance is one of the vulnerabilities of paclitaxel, we scrutinized the in vivo efficacy of the two drugs against resistant tumor tissues.[19] In the initial studies, CCRF-CEM/VBL tumor tissue was implanted subcutaneously in SCID mice. The tumor-bearing mice were treated periodically, according to a set protocol, with epothilone B (**2**, 0.7 mg kg^{-1}) and paclitaxel (**3**, 2 mg kg^{-1}). Although the concentration of paclitaxel used was higher than that of epothilone B (ca. 1.7:1 molar ratio), the reduction in tumor size was substantially greater with epothilone B at each interval of measurement (Table 2).



Scheme 1. The three arbitrarily defined sectors of the epothilones: aryl (1), alkyl (2), and acyl (3) sectors. Sector 1 is very sensitive to modifications; sectors 2 and 3 are more tolerant.

Table 2. Chemotherapeutic effect of daily treatments with epothilone B and paclitaxel in CB-17 SCID mice bearing drug-resistant human CCRF-CEM/VBL xenografts[a].

Drug	Dose [mg kg^{-1}][c]	Average tumor volume (T/C)[b] day 7	day 12	day 17	day 22
control	0	1.0	1.0	1.0	1.0
epothilone B	0.7	1.0	0.32	0.40	0.33
paclitaxel	2.0	1.0	0.60	0.58	0.70

[a] Multidrug resistant CCRF-CEM/VBL tumor tissue, 50 μL per mouse, was implanted subcutaneously on day 0. Solutions of the drugs in DMSO were injected intraperitoneally on days 7, 8, 9, 10, 14, and 15. There were seven CB-17 SCID mice in each treated group and in the control group. [b] The tumor volumes for each group on day 7 were about 1 mm^3. The average tumor volumes of the control group on days 12, 17, and 22 were 35, 107 and 278 mm^3, respectively. [c] On day 12, the average decreases in body weight due to treatment with epothilone B and paclitaxel were 2.7 and 3.4%, respectively.

In a subsequent experiment, epothilone B and paclitaxel were administered weekly (Table 3).[22] In these experiments both the sensitive (CCRF-CEM) and resistant (CCRF-CEM/VBL) tumor tissues were implanted in SCID mice. Epothilone B was given both intraperitoneally (H_2O) and intravenously (DMSO) for comparison. Epothilone B and paclitaxel show similar reduction of tumor size in treatment of the sensitive tumor (CCRF-CEM). Against the resistant tumor (CCRF-CEM/VBL), epothilone B shows a significant advantage over paclitaxel. It is also interesting to note that epothilone B is more

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Table 3. Chemotherapeutic effect of weekly treatments with epothilone B and paclitaxel in CB-17 SCID mice bearing human CCRF-CEM and CCRF-CEM/VBL xenografts[a].

Tumor	Drug	Route	Dose [mg kg^{-1}][b]	Average tumor volume (T/C) [c] day 10	day 15	day 20	day 25
CCRF-CEM	control	i.p.	0	1.0	1.0	1.0	1.0
	epothilone B	i.p.	1.5	0.40	0.40	0.37	0.34
		(H_2O)	3.0[d]	0.41	0.35	0.38	0.50
	epothilone B	i.v.	1.5	0.38	0.34	0.42	0.37
		(DMSO)	3.0[e]	0.28	0.38	0.29	0.24
	paclitaxel	i.p.	20.0[f]	0.33	0.28	0.31	0.34
		(DMSO)	30.0[g]	0.43	0.25	0.23	0.25
CCRF-CEM/ VBL	control	i.p.	0	1.0	1.0	1.0	1.0
	epothilone B	i.p.	1.5[h]	0.23	0.34	0.27	0.26
		(H_2O)	3.0[i]	0.26	0.25	0.20	0.22
	epothilone B	i.v.	1.5	0.21	0.19	0.27	0.26
		(DMSO)	3.0	0.24	0.27	0.14	0.20
	paclitaxel	i.p.	20.0[j]	0.77	0.51	0.60	0.59
		(DMSO)	30.0[k]	0.58	0.46	0.42	0.61

[a] CCRF-CEM and CCRF-CEM/VBL tumor tissue, 50 µL per mouse, implanted subcutaneously on day 0. The drugs were administered on day 5, 12, 19 (i.p. = intraperitoneally, i.v. = intravenously). There were five CB-17 SCID mice in each group. [b] On day 20 the average decrease in body weight due to epothilone B doses of 1.5 and 3.0 mg kg^{-1} was 5.0 and 6.6%, respectively. [c] The tumor volumes for each group on day 5 was about 8 mm^3. The average tumor volumes in the CCRF-CEM control group on days 10, 15 and 20 were 57, 145, 335 mm^3; those of the CCRF-CEM/VBL control group were 62, 173, and 386 mm^3, respectively. [d]–[k] A number of mice died of drug toxicity: two on day 23 (d), two on day 19 (e), three on days 13, 21, and 21 (f), three on days 6, 6, and 21 (g), one on day 24 (h), two on day 24 (i), two on day 13 (j), and four on day 6 (k).

effective against the resistant tumor tissues than sensitive tissues in these experiments.

At present, none of our analogs are more potent in vitro than the naturally occurring epothilone B (**2**), although the ethyl compound **12** is fully competitive in all comparisons thus far available. However, we have established zones of the molecule that are most likely to be responsive to molecular modification (see Scheme 1). Also, early in vivo data suggest a potentially valuable margin of advantage of epothilone B (**2**) relative to paclitaxel (**3**) against a highly resistant MDR tumor implant. In all in vivo experiments conducted so far, epothilone B and paclitaxel were equally effective in reducing the size of sensitive (CCRF-CEM) tumor tissues. While it is far too early to argue that we have demonstrated a superior alternative to paclitaxel, the data already in hand mark the epothilones as worthy of continuing interest. Our present focus is on selecting the most promising compounds for further development, and on a major restructuring of our synthesis for the preparation of any desirable compound in the series in an eminently practical fashion.

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[12] Identification of the deleted carbon in compound **18** as C9 is arbitrary; it could also be looked upon as deletion of C10 or C11. For the purpose of this discussion, we look upon this compound as a "C9 deletion" corresponding to an alteration of the substituents at C8.

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[15] The (*Z*)-desoxy compound with *n*-hexyl at C12 was also found to have significant biological activity; however, the solubility of this compound in DMSO is very low. Therefore, the results are not included in Figure 1.

[16] Compound **9** was treated with 3,3-dimethyldioxirane to create the *trans*-epoxide, which was found to have significant biological activity. However, the absolute configuration of the *trans*-epoxide has not been determined.

[17] Microtubule protein (MTP) was purified from calf brains by two cycles of temperature-dependent assembly and disassembly [20]. The concentration of tubulin in the preparation was approximately 85%. Assembly experiments were done in the presence or absence of 10 µM drug plus MTP (1 mg mL^{-1} diluted in assembly buffer containing 0.1 M 2-(*N*-morpholino)ethanesulfonic acid (MES), 1 mM 1,2-di(2-aminoethoxy)ethane-*N*,*N*,*N*',*N*'-tetracetic acid (EGTA), 0.5 mM $MgCl_2$, and 3 M glycerol, pH 6.6). This concentration of drug gives an approximate 1:1 ratio to tubulin dimer. Assembly was followed spectrophotometrically at 350 nm, 37 °C for 40 min, by monitoring changes in turbidity as a measure of polymer mass [21]. Aliquots (200 µL) were taken from each assembly reaction and centrifuged at 28000 rpm, 27 °C for 30 min. The amount of protein remaining in the supernatant was determined, allowing the protein concentration in the microtubule pellet to be calculated. Controls with assembly buffer or DMSO were included. The value obtained for microtubules formed with 10 µM epothilone B was defined as 100%.

[18] The cytotoxicities of test compounds were determined by the growth of human lymphoblastic leukemic cells CCRF-CEM or their sublines resistant to vinblastine and taxol (CCRF-CEM/VBL). XTT-microculture tetrazolium/fromazan assays were used. The IC_{50} values were calculated based on five or six assays at various concentrations; a median-effect plot was generated using computer software [23].

[19] The multidrug resistant CCRF-CEM/VBL tumor tissue was implanted subcutaneously to CB-17 SCID mice, and the tumor-bearing mice were treated on days 7, 8, 9, 10, 14, and 15 with 0.7 mg kg^{-1} epothilone B intraperitoneally. The average tumor sizes were reduced by 68, 60, and 67% on days 12, 17, and 22, respectively. In the parallel experiments, 2 mg kg^{-1} taxol reduced tumor size by 40, 42, and 30%, respectively.

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[22] Experiments were also conducted with daily (0.7 and 1.0 mg kg^{-1}) and twice daily (0.15 and 0.25 mg kg^{-1}) treatments of epothilone B. The efficacy of once and twice daily treatments with epothilone B was similar to the weekly treatments, but the drug toxicity was slightly higher.

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