Natural Products

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Rapid Construction of the Cortistatin Pentacyclic Core**

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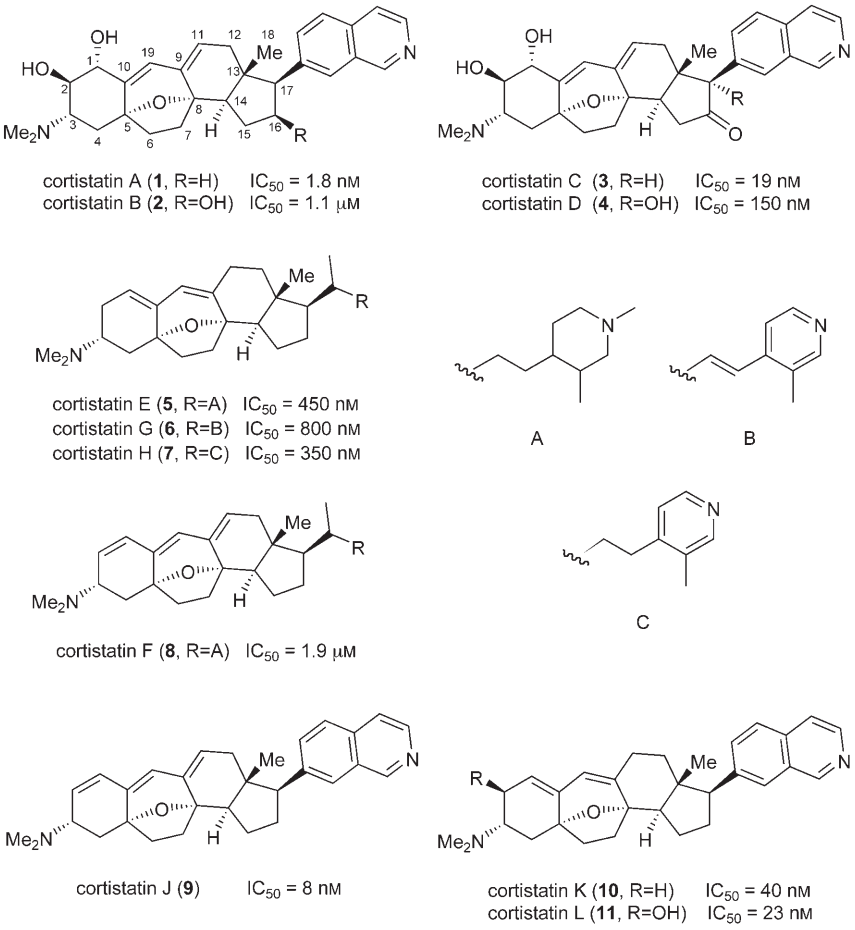
Angiogenesis, the formation of new blood capillaries, is an essential process for embryonic development and wound repair. Under normal circumstances, this process is tightly regulated and is promoted by angiogenic polypeptides such as vascular endothelial growth factor (VEGF), basic fibroblast growth factor (bFGF) and angiogenin.[1] However, angiogenesis also plays a key role in tumor growth and metastasis, and the discovery of angiogenesis inhibitors has generated excitement about their potential use in the treatment of cancer. When administered alongside traditional chemotherapeutics, antiangiogenics suppress the recurrence of tumor growth, leading to curative treatment in some animal models.[2] In contrast to many existing chemotherapies, antiangiogenic therapy typically exhibits low toxicity and drug resistance does not appear to be a significant problem.[1b] Research in this area is in the early stages, but recently Bevacizumab, a monoclonal antibody against VEGF, was approved for the treatment of colon and breast cancers.[3] Because of these promising initial results, there is intense interest in the discovery and study of novel antiangiogenic factors.



Scheme 1. Members of the cortistatin family and their respective IC_{50} values against HUVECs.

Herein, we report our initial synthetic studies on the antiangiogenic compound cortistatin A (**1**, Scheme 1). This compound is among the first members of a new family of rearranged steroidal alkaloids (**1**–**4**) that were isolated from the Indonesian marine sponge *Corticium simplex* in 2006 by Kobayashi and coworkers.[4] All four compounds were found to have significant antiangiogenic activity, with the most potent being cortistatin A (**1**), displaying an IC_{50} value of 1.8 nM against human umbilical vein endothelial cells (HUVECs), which is a standard model for antiangiogenic activity. Importantly, this high activity was coupled with an exquisite selectivity (> 3000) relative to other cell types, thus arguing against general cytotoxicity as a mode of action.

This initial isolation report was followed a year later by the discovery of seven additional family members, cortistatins E–L, from the same source.[5] Although they share the same pentacyclic core as their previously isolated congeners, cortistatins E–H incorporate *N*-methylpiperidine or 3-methylpyridine in their side chains (see **5**–**8**, Scheme 1) in place of the isoquinoline fragment. In addition, the degree and position of unsaturation and oxygenation along the northern edge of the ABC rings differs relative to cortistatins A–D.[5a] Cortistatins J–L (**9**–**11**) show similar variation in the ABC-ring substitution pattern, but retain the isoquinoline ring at C17.[5b] Preliminary structure–activity analysis suggests that the presence of the isoquinoline moiety is crucial for potent activity: while cortistatins A–D and J–L were found to have

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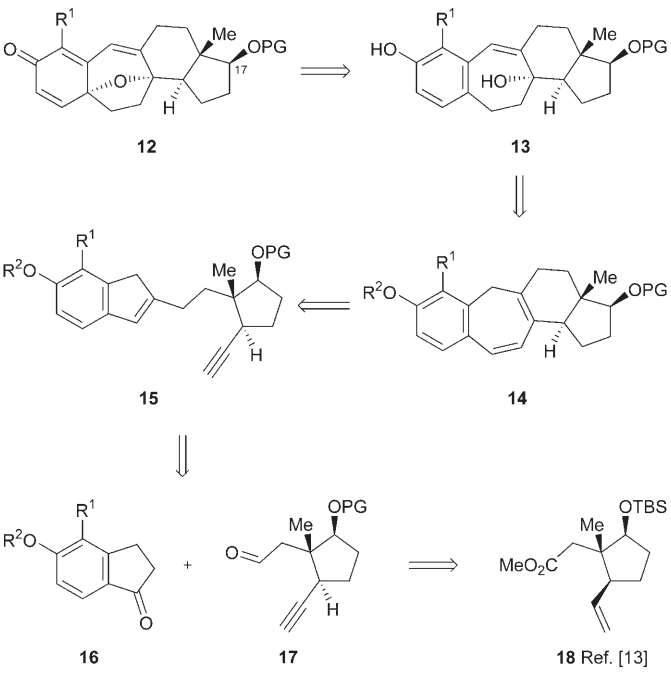
IC_{50} values as low as 1.8 nM, cortistatins E–H were only weakly active (IC_{50} value: 350 nM–1.9 μM). Furthermore, the oxygenation at C16 or C17 of the D ring appears to significantly attenuate activity (see **1**–**4**).[6]

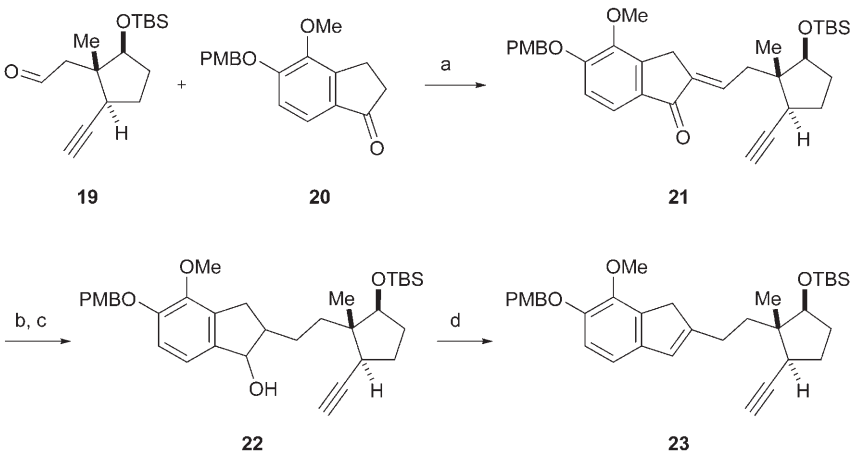
The cortistatins discovered to date share a unique rearranged steroid core where the C19 methyl group has been incorporated into the B ring, and an ether bridge links C5 and C8. There are many steroids in which a cyclopropane ring has formed from C9, C10 and the C19 methyl group. In contrast, the corresponding ring-expanded *seco*-tetracycle bearing a seven-membered B ring is relatively rare. This type of arrangement has only previously been found in other marine sponges in the *Corticium* genus,[7] as well as in terrestrial plant species in the *Buxus*[8] and *Cimicifuga*[9] genera. However, the ether-containing pentacycle that is characteristic of the cortistatins is unprecedented among known natural products. The clear synthetic challenge posed by the cortistatins, coupled with their unique biological activity has generated significant interest from the synthetic community, but as yet a total synthesis has not been reported.[10] Herein, we present a concise approach which has led to the de novo assembly of the pentacyclic core of the cortistatins. This approach now sets the stage for a total synthesis of these intriguing natural products.

One could imagine utilizing a commercially available steroid as a starting point to arrive at a given member of the cortistatin family through appropriate functional group manipulations. Indeed, such partial syntheses are common in the steroid literature,[11] and this approach would likely be successful in devising a route to a specific cortistatin target, as evidenced by the recent synthesis of **1** by Baran and coworkers.[10] In considering our synthetic strategy, we sought a flexible intermediate that would provide facile access to any of the cortistatin natural products as well as any desired synthetic analogues. To that end, we envisioned that pentacycle **12** (Scheme 2) could be tailored to a variety of substitution patterns utilizing the trienone handle in the A and B rings and the oxygen substituent at C17. The ether bridge could be constructed by oxidative dearomatization of phenol **13**. We were especially intrigued by the possibility of arriving at **13** from benzocycloheptadiene **14**, which we anticipated would be readily available from alkynyl indene **15** by an enyne cycloisomerization reaction, which was developed in our laboratories and was previously applied to the total synthesis of several icetexane diterpenoid natural products.[12] Finally, **15** could be derived from indanone **16** and aldehyde **17**, which in turn could be prepared from the known ester **18**.[13] Not only would this strategy be amenable to the preparation of synthetic analogues modified in the A and D rings, but also changes in the B- and C-ring substitution could easily be accommodated by appropriate manipulations prior to the construction of the pentacycle.



Scheme 2. Retrosynthetic analysis. PG = protecting group, TBS = *tert*-butyldimethylsilyl.

Our synthesis of the cortistatin pentacyclic core commenced with aldehyde (±)-**19** (Scheme 3), which was readily prepared in four steps and with 53% overall yield from **18**.[14] Notably, **18** has been previously synthesized in enantiopure form, thus providing a straightforward avenue for an enantioselective synthesis. Fragment coupling of **19** with indanone **20**[15] was achieved by aldol condensation to give enone **21** as a single diastereomer in 51% yield following recrystallization. Selective 1,4-reduction of the enone moiety of **21** was achieved by treatment with K-Selectride[16] to yield indanol **22** (as an inconsequential mixture of diastereomers) and the corresponding indanone in approximately a 1:2 ratio. The



Scheme 3. Synthesis of indene **23**. Reagents and conditions: a) **20** (1.0 equiv), **19** (1.0 equiv), KOH (1.0 equiv), $EtOH/CH_2Cl_2$ (3:1), RT, 2.5 h, 51%; b) K-Selectride (3.0 equiv), THF, −78 °C, 2 h; then −78 °C→RT, 2 h; c) $NaBH_4$ (1.0 equiv), $MeOH/CH_2Cl_2$ (1:1), 0 °C, 1 h; d) $KHSO_4$ (1.0 equiv), toluene, 50 °C, 18 h, 65% (over 3 steps).

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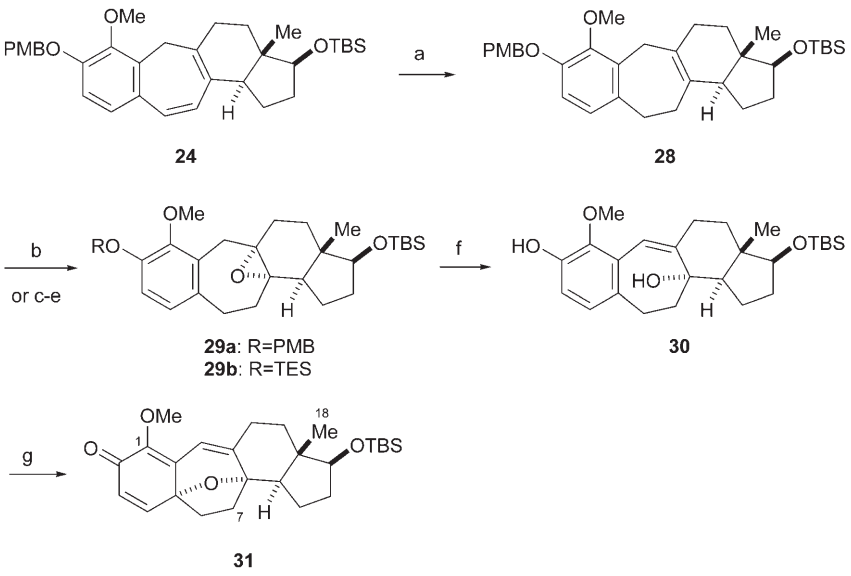
remaining indanone was smoothly converted into indanol **22** by treatment of the crude mixture with $NaBH_4$. Subsequent dehydration of the secondary alcohol was achieved by gentle heating of **22** with $KHSO_4$ in toluene to give indene **23** in 65% yield over the three-step sequence. Notably, while thermally promoted double-bond isomerization of indenes has previously been observed in related systems,[12a] no trace of the isomerization product of **23** could be detected even after 36 h at 50 °C.

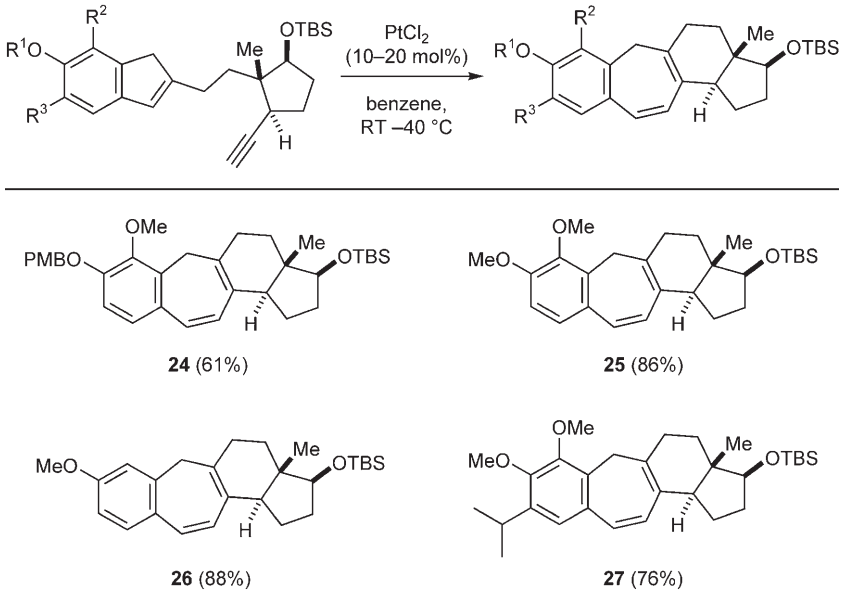
With indene **23** in hand, we anticipated that cycloisomerization could be achieved with $GaCl_3$ as a catalyst on the basis of previous observations.[12a,17] However, preliminary studies indicated that cleavage of the TBS ether was a competing side reaction. Gratifyingly, the desired cycloheptadiene (**24**, Scheme 4) could be readily obtained by treatment of **23** with a catalytic amount of $PtCl_2$.[18] In contrast to the icetexane substrates from our previous studies, cycloisomerization of **23** with $PtCl_2$ occurred without significant formation of isomeric by-products. We have also examined the cycloisomerization of several other indene substrates (Scheme 4), which were easily synthesized from the corresponding indanones[12a,19] by a sequence analogous to that shown in Scheme 3. Pleasingly, each of these alkynyl indenes underwent smooth cycloisomerization to the corresponding cycloheptadiene tetracycle (see **25**–**27**) upon treatment with a catalytic amount of $PtCl_2$.



Scheme 5. Completion of the cortistatin core synthesis. Reagents and conditions: a) $TsNHNH_2$ (8.0 equiv), Et_3N (16 equiv), 1,2-dichloroethane, 65 °C, 24 h, 95% after one recycle; b) *m*-CPBA (1.2 equiv), $NaHCO_3$ (2.0 equiv), CH_2Cl_2, 0 °C, 2.5 h, 66% (**29a**); c) $MgBr_2{\cdot}OEt_2$ (6.0 equiv), Me_2S (20 equiv), CH_2Cl_2, −78 °C→RT, 3 h; d) TESCl (1.5 equiv), imidazole (3.0 equiv), DMF, RT, 3 h; e) *m*-CPBA (1.2 equiv), $NaHCO_3$ (2.0 equiv), CH_2Cl_2, 0 °C, 1.5 h, 46% (**29b** over 3 steps from **28**); f) *n*BuLi (5.0 equiv), THF, 0 °C, 1 h; g) $PhI(OAc)_2$ (1.5 equiv), CH_2Cl_2/*i*PrOH/TFE (5:3:2), 0 °C, 30 min, 60% (over 2 steps from **29b**). *m*-CPBA = *meta*-chloroperbenzoic acid, DMF = *N*,*N*-dimethylformamide, Ts = *para*-toluenesulfonyl.

To elaborate cycloheptadiene **24** (Scheme 5) to the cortistatin core, the disubstituted double bond was chemoselectively reduced with diimide to give tetrasubstituted alkene **28** in excellent yield.[20] Subsequent treatment with *m*-CPBA at 0 °C gave epoxide **29a** as a single diastereomer.[21] Although **29a** proved to be exceedingly acid sensitive, regioselective epoxide opening could be achieved by treatment with *n*-butyllithum, which proceeded with concomitant cleavage of the *para*-methoxybenzyl (PMB) ether to give phenol **30**.[22] Even though this reaction proceeded satisfactorily on a small amount of material (1 mg), scale-up led to a significant increase in the amount of side-products and to variable yields of **30**. To circumvent this impediment, the PMB group was removed under non-oxidative conditions ($MgBr_2{\cdot}OEt_2$/Me_2S),[23] which prevented oxidation of the electron-rich benzene ring, and the resulting phenol was directly converted into the triethylsilyl (TES) ether.[24] Epoxidation of the TES ether proceeded to give epoxide **29b** in 46% yield over the three-step sequence (from **28**).[21] Gratifyingly, base-mediated epoxide opening of **29b** was accompanied by the clean removal of the silyl group to give **30** as a single product that did not require purification.

With a reliable route to **30** in hand, we turned our attention to the key tandem oxidative dearomatization/cyclization,



Scheme 4. Cycloisomerization products.

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which we expected could be achieved using hypervalent iodine.[25] Although protic solvents are typically more effective at stabilizing the cationic intermediates generated under the oxidation conditions, we posited that competitive solvent addition at C1 could be problematic. Indeed, treatment of **30** with $PhI(OAc)_2$ in 1:1 CH_2Cl_2/MeOH led to a 1:1 mixture of the desired pentacycle **31** and the C1 dimethyl ketal.[26] Replacement of MeOH with the less nucleophilic, protic solvent 2,2,2-trifluoroethanol (TFE) led to **31** as the major product, but this was accompanied by a significant amount of decomposition products. We hypothesized that a bulky, non-fluorinated, protic solvent might stabilize the reactive intermediates without acting as a nucleophile. In the event, treatment of **30** in CH_2Cl_2/*i*PrOH with a solution of $PhI(OAc)_2$ in TFE gave pentacyclic trienone **31** in 60% yield over two steps (from **29b**), with no detectable products arising from solvent addition. The structure of **31** was supported by extensive 2D NMR analysis (COSY, HMQC, and NOESY spectroscopy). The stereochemistry of the ether bridge was secured by observation of an nOe interaction between the C18 methyl group and one of the C7 protons, which is analogous to that observed for cortistatin A.[4]

In summary, we have reported the construction of the pentacyclic core of the cortistatin steroidal alkaloids through a concise strategy featuring a $PtCl_2$-catalyzed enyne cycloisomerization and an oxidative dearomatization/cyclization as the key steps. The entire sequence proceeds in 11 steps from aldehyde **19** and is directly amenable to an asymmetric synthesis given the ready availability of enantiopure **18**. Application of the strategy disclosed here to the total synthesis of cortistatin A is underway and will be reported in due course.[27]

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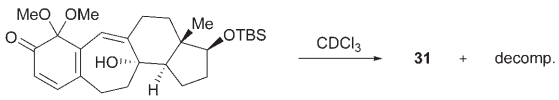

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[27] After online publication of our manuscript, we became aware of a relevant manuscript: M. Dai, S. J. Danishefsky, *Heterocycles* **2008**, DOI: COM-08-S(F)6.

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