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Total Syntheses of 2,2′-*epi*-Cytoskyrin A, Rugulosin, and the Alleged Structure of Rugulin

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Abstract: The total syntheses of 2,2′-*epi*-cytoskyrin A, rugulosin, and the alleged structure of rugulin are described. These naturally occurring bisanthraquinones and their relatives are characterized by novel molecular architectures at the core, at which lies a more or less complete, cage-like structural motif termed "skyrane". The strategies developed for their total synthesis feature a cascade sequence called the "cytoskyrin cascade" and deliver these molecules in short order and in a stereoselective manner.

Introduction

The naturally occurring bisanthraquinones cytoskyrin A (**1a**),[1] graciliformin (**1b**),[2] 2,2′-*epi*-cytoskyrin A (**2a**),[3] rugulosin (**2b**),[4] rugulin (**3**),[5] deoxyrubroskyrin (**4**),[4b] 1,1′-bislunatin (**5a**),[3] skyrin (**5b**)[4b,6] and flavoskyrin (**6**),[4d] all shown in Figure 1, represent an impressive and growing class of compounds with varying degrees of structural complexity. Isolated from a number of fungi and lichens, these molecules exhibit a diverse array of biological activities including cytotoxic (**1a**, **1b**, **2b**),[7] antibacterial (**3**),[8] insecticidal (**2b**),[9] anti-influenzal (**2b**),[10] and anti-HIV (**2b**)[11] properties. The most recent members of the family are cytoskyrin A (**1a**),[1] isolated from small-scale cultures of the endophytic fungus CR200 (*Cytospora* sp.) and possessing potency down to 12.5 ng mL^{-1} in the biochemical induction assay (BIA), and 2,2′-*epi*-cytoskyrin A (**2a**)[3] and 1,1′-bislunatin (**5a**),[3] both of which were found in extracts of the endophytic fungus *Diaporthe* sp. residing in a tea plant. Attracted by their intriguing molecular architectures and diverse biological activities, we initiated a program directed toward the total syntheses of these compounds, initially targeting cytoskyrin A (**1a**), the newest member of the class at the time. Herein, we describe in detail our investigations in this field[12] that culminated in the total syntheses of 2,2′-*epi*-cytoskyrin A (**2a**), rugulosin (**2b**), and the alleged structure **3** of rugulin as well as a number of analogues and model systems of these naturally occurring substances. These investigations highlighted the importance of cascade reactions in total synthesis, rendered readily available a number of complex molecular architectures, and enabled us to conclude that **3** does not represent the true structure of rugulin, whose true molecular architecture remains elusive.

Results and Discussion

The present study was inspired by the fascinating structures of the bisanthraquinones and the possibility of sequentially constructing them through cascade reactions starting from monomeric anthraquinone precursors. This expectation was fueled by the work of Shibata and co-workers, who demonstrated the feasibility of such synthetic schemes decades ago.[2b,4] These molecular assemblies are characterized by a central structural motif, more or less complete, as we move from the monomeric units to the more complex bisanthraquinones. The structure of rugulin (**3**) in particular contains a closed cage-like structural motif consisting of 12 carbon atoms arranged in such a way as to form a short cylinder shaped by two six-membered rings (top and bottom faces) and four five-membered rings. The latter systems are uniformly twisted as they reach to form the walls of the molecular cylinder which we termed, in our first

(1) (a) Brady, S. F.; Singh, M. P.; Janso, J. E.; Clardy, J. *Org. Lett.* **2000**, *2*, 4047−4049. (b) Jadulco, R.; Brauers, G.; Edrada, R. A.; Ebel, R.; Wray, V.; Sudarsono Proksch, P. *J. Nat. Prod.* **2002**, *65*, 730−733.
(2) (a) Ejiri, H.; Sankawa, U.; Shibata, S. *Phytochemistry* **1975**, *14*, 277−279. (b) Shibata, S. *Farumashia* **2006**, *42*, 11−14.
(3) Agusta, A.; Ohashi, K.; Shibuya, H. *Chem. Pharm. Bull.* **2006**, *54*, 579−582.
(4) (a) Ogihara, Y.; Kobayashi, N.; Shibata, S. *Tetrahedron Lett.* **1968**, *9*, 1881−1886. (b) Takeda, N.; Seo, S.; Ogihara, Y.; Sankawa, U.; Iitaka, I.; Kitagawa, I.; Shibata, S. *Tetrahedron* **1973**, *29*, 3703−3719. (c) Shibata, S. *Pure Appl. Chem.* **1973**, *33*, 109−128. (d) Seo, S.; Sankawa, U.; Ogihara, Y.; Iitaka, Y.; Shibata, S. *Tetrahedron* **1973**, *29*, 3721−3276. (e) Yang, D.-M.; Sankawa, U.; Ebizuka, Y.; Shibata, S. *Tetrahedron* **1976**, *32*, 333−335.
(5) Sedmera, P.; Podojil, M.; Vokoun, J.; Betina, V.; Nemec, P. *Folia Microbiol.* **1978**, *23*, 64−67.
(6) Franck, B.; Chahin, R.; Eckert, H.-G.; Langenberg, R.; Radtke, V. *Angew. Chem., Int. Ed. Engl.* **1975**, *14*, 819−820.
(7) (a) Umeda, M.; Saito, M.; Shibata, S. *Jpn. J. Expt. Med.* **1974**, *44*, 249−255. (b) Ueno, Y.; Kubota, K. *Cancer Res.* **1976**, *36*, 445−451. (c) Watts, P.; Kittakoop, P.; Veeranondha, S.; Wanasith, S.; Thongwichian, R.; Saisaha, P.; Intamas, S.; Hywel-Jones, N. L. *Mycol. Res.* **2003**, *107*, 581−586.
(8) Betina, V.; Nemec, P. Czech Patent CS187049, Jan 31, 1979.
(9) (a) Dodias, J.; Betina, V.; Nemec, P. *Biologia (Bratislava, Slovakia)* **1980**, *35*, 431−434. (b) Dodias, J.; Nemec, P.; Betina, V. Czech Patent CS189849, May 31, 1979.
(10) Nakamura, S.; Nii, F.; Inoue, S.; Nakanishi, I.; Shimizu, M. *Jpn. J. Microbiol.* **1974**, *18*, 1−7.
(11) Singh, S. B.; et al. *J. Ind. Microbiol. Biotechnol.* **2003**, *30*, 721−731.
(12) For preliminary communications, see: (a) Nicolaou, K. C.; Papageorgiou, C. D.; Piper, J. L.; Chadha, R. K. *Angew. Chem., Int. Ed.* **2005**, *44*, 5846−5851. (b) Nicolaou, K. C.; Lim, Y. H.; Papageorgiou, C. D.; Piper, J. L. *Angew. Chem., Int. Ed.* **2005**, *44*, 7917−7921.

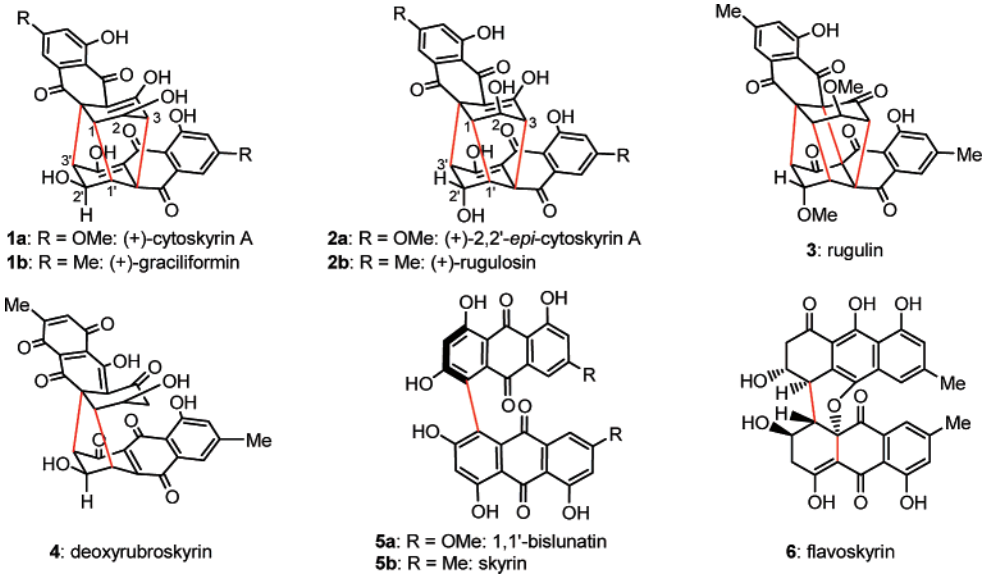


Figure 1. Selected naturally occurring modified bisanthraquinones.

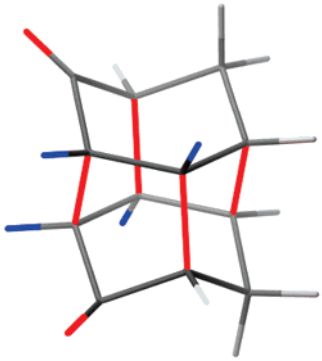


Figure 2. Skyrane, the core, cage-like structural motif of rugulin (**3**).

communication,[12a] "skyrane" (see Figure 2). Depending on its substitutents, this intriguing system may exhibit helical chirality, as will be further discussed below.

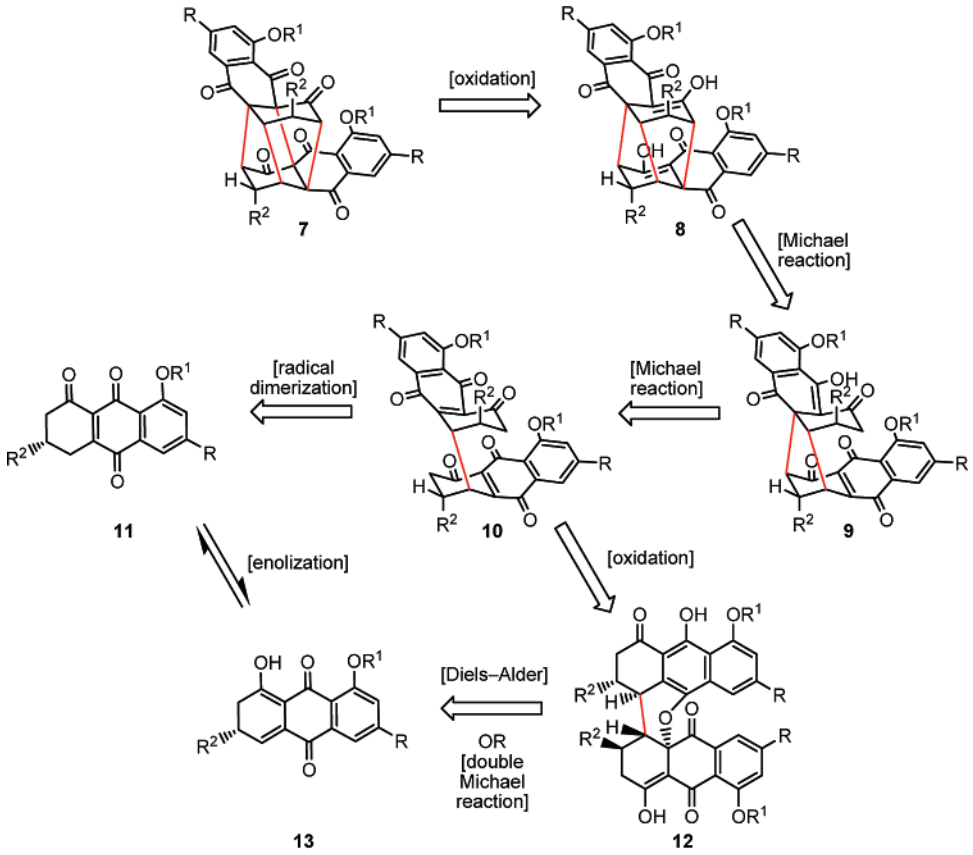
General Retrosynthetic Analysis. Upon inspection, structures **1**−**6** can be arranged as shown in Scheme 1. The ordering is arrived at by placing at the top left (**7**) of the scheme the most complex of the structures (i.e., the one with the complete skyrane structural motif at its core) and proceeding by disconnecting, one at a time, the carbon−carbon bonds linking the two halves of the molecule until the monomeric anthraquinone, placed at the middle left (**11**) of the scheme, is reached. The thus-generated retrosynthetic scheme points to a strategy whereby the anthraquinone precursor **11** can serve as the starting point for a cascade sequence in which enolization is followed by either a double Michael reaction between two monomeric species or a Diels−Alder/dimerization process to form the flavoskyrin-type framework **12**. Ideally, the cascade should be controllable so as to stop at any stage, allowing the isolation of all intermediates along the way before reaching its final destination, the rugulin-type structure **7**. The conversion of the oxygen-bridged heterocyclic compound **12** to the skyrin-type structure **10** was expected to require oxidative conditions, whereas the same framework (**10**) could, in principle, be reached directly from **11** through a radical-based dimerization. The final ring closure to complete the cage (i.e., **8** → **7**) should be possible through a second oxidative process. As the cage-type structure of the most compact molecules of the series is disassembled, the construction of the encountered intermediates should be favored by the expected relief of strain on one hand, but disfavored by the entropy increase associated with each process on the other.

Model Studies. In order to explore the feasibility of the proposed cascade, we chose compound **7**, $R^1 = R^2 = H$, R = OMe (Scheme 1), as a model target molecule. The simplified architecture of this model system was thought to be a good starting point for developing the desired cascade, for the intermediates involved would not be prone to elimination/aromatization side reactions along the way. We first sought to construct the monoanthraquinone **23** (Scheme 3) and its dimer, bisanthraquinone **22** (Scheme 2), as potential starting substrates for the required cascade sequence.

The synthesis of monoanthraquinone **23** and bisanthraquinone **22** features a Hauser annulation[13] starting with nitrile **14** and cyclohexenone **15**, as shown in Scheme 2. Thus, exposure of **14** (1.1 equiv) to LHMDS (1.5 equiv) [for abbreviations of reagents and protecting groups, see legends in schemes] at −78 °C in THF for 1 h, followed by quenching with cyclohexenone **15** (1.0 equiv) and warming to room temperature, furnished the crude dihydroquinone **16**. This product was, without purification, converted to the tetramethylated ethylene ketal **17** by exposure to excess Me_2SO_4 in the presence of K_2CO_3 (acetone, reflux, 12 h) in 85% overall yield over the two steps from **15**. Treatment of **17** with excess ethylene glycol and PPTS (benzene, reflux, 24 h) resulted in the formation of its ethylene ketal, which upon exposure to KO*t*-Bu in DMSO in the presence of oxygen at ambient temperature resulted in the formation of the corresponding benzylic alcohol **18** in 53% overall yield over the two steps. Conversion to chloride **19** was subsequently accomplished through the action of TMSCl on **18** in DMSO in 80% yield. The envisioned dimerization of **19** was carried out by treatment with $[CoCl(PPh_3)_3]$,[14] a reagent presumed to act by abstracting a chlorine atom from the substrate, leaving behind the corre-

(13) Hauser, F. M.; Chakrapani, S.; Ellenberger, W. P. *J. Org. Chem.* **1991**, *56*, 5248−5250.
(14) (a) Momose, D.; Iguchi, K.; Sugiyama, T.; Yamada, Y. *Tetrahedron Lett.* **1983**, *24*, 921−924. (b) Bagal, S. K.; Adlington, R. M.; Marquez, R.; Cowley, A. R.; Baldwin, J. E. *Tetrahedron Lett.* **2003**, *44*, 4993−4996.

Scheme 1. General Retrosynthetic Strategy toward the Modified Bisanthraquinones



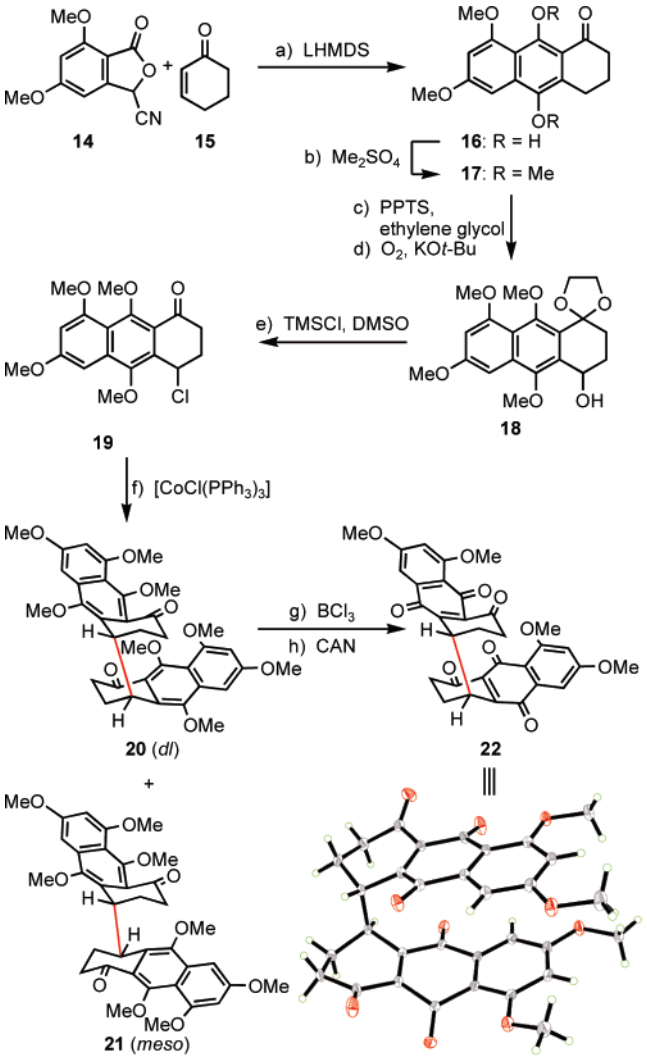
sponding fleeting radical, whose dimerization through a carbon−carbon bond formation led to two isomeric products in ca. 55:45 ratio. The isomers were separated chromatographically and spectroscopically characterized as the *dl* compound **20** (55% yield) and its *meso* isomer **21** (45% yield). An X-ray crystallographic analysis[15] of a subsequently derived intermediate (**22**, see ORTEP structure in Scheme 2) confirmed these assignments. The major (*dl*) isomer was then treated with BCl_3 in CH_2Cl_2 at −78 °C, conditions that selectively cleaved the two methyl ethers nearest to the carbonyl groups, followed by oxidation with CAN to afford the corresponding *dl*-bisanthraquinone **22** in 50% yield over the two steps. The *dl*-bisanthraquinone was crystallized from acetone (mp 295−298 °C, dec), and a crystal was subjected to X-ray crystallographic analysis, which revealed its stereochemical identity and that of its progenitor compound **20** (see ORTEP drawing in Scheme 2) and, by extension, that of its isomer **21**.

A more direct pathway to bisanthraquinone **22** from its monomeric unit, anthraquinone **23**, which was prepared from dihydroquinone **16** in quantitative yield through the action of CAN (Scheme 3), was also considered. To this end, **16** was exposed to catalytic amounts of either CSA or *i*-Pr_2NEt in CH_2Cl_2 at ambient temperature for 1 h, furnishing the flavoskyrin model system **25** in 94 and 90% yields, respectively. It was presumed that this transformation was initiated through enolization of the starting anthraquinone as shown, to afford dienol **24**, whose dimerization to form the observed product may have proceeded through either a double Michael reaction (blue and red arrows) or a hetero Diels−Alder-type process (red arrows), as exemplified in Scheme 3. The structure of **25** was unambiguously confirmed by both spectroscopic and X-ray crystallographic analysis[15] (see ORTEP structure in Scheme 3). The undesired oxygen bridge within **25** was easily dismantled by oxidation with MnO_2 to afford the desired *dl*-bisanthraquinone **22** (83% yield) in a process presumed to proceed through the mechanism shown in Scheme 4 (**25** → **I** →**22**).

With bisanthraquinone **22** readily available from two different routes, its conversion to the deoxyrubroskyrin-type structure **26** was then explored (Scheme 5). Pleasantly, it was found that exposure to Et_3N (2.0 equiv) in CH_2Cl_2 at ambient temperature for 1 h was sufficient inducement for compound **22** to undergo the first Michael reaction, furnishing compound **26** in 65% yield from **22**. After screening a number of conditions (see Table 1, entries 1−7), it was discovered that the second Michael reaction, required to convert **26** to the cytoskyrin-type structure **27**, was induced simply by further treatment with Et_3N (5.0 equiv), this time at 45 °C (16 h, 95% yield), or *i*-Pr_2NEt (2.0 equiv) in $ClCH_2CH_2Cl$ at 80 °C for 20 h (95% yield). The use of acidic conditions (PPTS, 2.0 equiv, $ClCH_2CH_2Cl$, 100 °C, 24 h; see Table 1, entry 4) and other basic conditions (pyridine, 100 °C, 12 h; and pyridine, microwaves, 80 °C, 15 min; then microwaves, 120 °C, 30 min (process repeated twice); see Table 1, entries 5 and 6, respectively) was also effective in carrying out this transformation, albeit in lower yields (65, 63, and 80%, respectively). Finally, exposure of cytoskyrin-type structure **27** to MnO_2 in CH_2Cl_2 at room temperature resulted in the formation of the rugulin-type structure **28** in 95% yield through

(15) CCDC-274275 (**22**), -274271 (**25**), -274272 (**26**), -286831 (**2b**), and -627897 (**3**) contain the crystallographic data for this paper. These can be obtained free of charge from the Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request.cif.

Scheme 2. Synthesis of the Model System: the Radical Route[a]

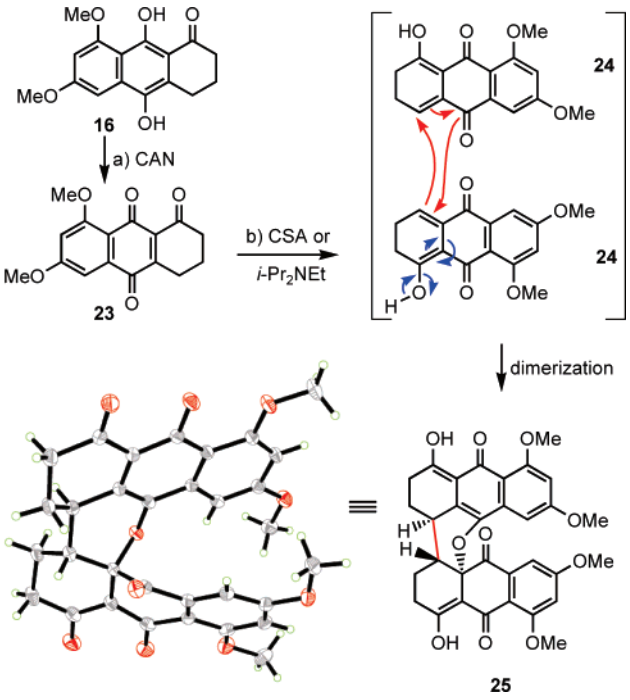


[a] Reagents and conditions: (a) **14** (1.1 equiv), THF, LHMDS (1.5 equiv), −78 °C, 1 h; then **15** (1.0 equiv), −78 → 25 °C over 4 h; (b) Me_2SO_4 (5.0 equiv), K_2CO_3 (5.0 equiv), acetone, reflux, 12 h, 85% over two steps; (c) ethylene glycol (2.0 equiv), benzene, PPTS (0.2 equiv), reflux, 24 h; (d) *t*-BuOK (6.0 equiv), O_2, DMSO, 25 °C, 2 h, 53% over two steps; (e) TMSCl (2.0 equiv), DMSO, 25 °C, 10 min, 80%; (f) $[CoCl(PPh_3)_3]$ (1.2 equiv), benzene, 25 °C, 3 h, 55% (**20**) + 25% (**21**); (g) BCl_3 (2.2 equiv), CH_2Cl_2, −78 → 25 °C, 12 h; (h) CAN (4.0 equiv), MeCN:H_2O (1:1), 0 °C, 10 min, 50% over two steps. Abbreviations: LHMDS, lithium hexamethyldisilazide; PPTS, pyridinium *p*-toluenesulfonate; TMS, trimethylsilyl; DMSO, dimethyl sulfoxide; CAN, cerium ammonium nitrate.

a process presumed to proceed through the mechanism depicted in Scheme 4 (**27** → **II** → **28**). Encouraged by these impressive results, we proceeded to attempt the daring, one-pot cascade conversions **25** → **27**, **25** → **28**, **23** → **27**, and **23** → **28**, as shown in Scheme 5, by screening a number of conditions (see Table 1, entries 8−24) for the various cascade sequences and monitoring the appearance and disappearance of the incipient compounds.

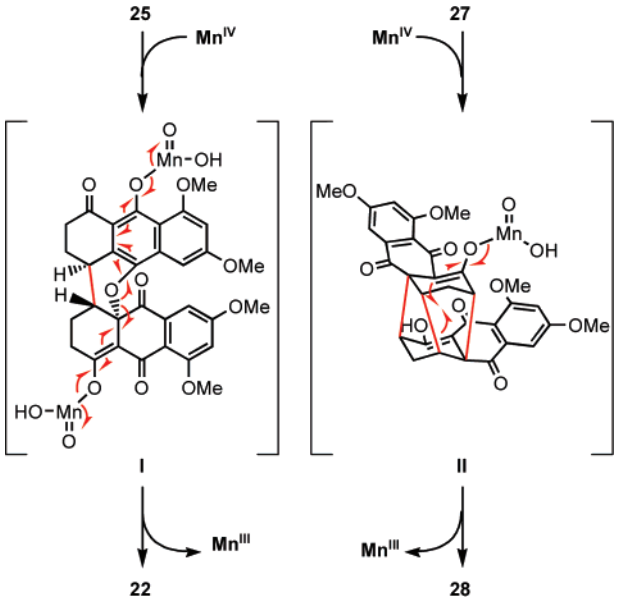
Thus, the one-pot cascade conversions of **25** → **27** and **25** → **28** were investigated under various basic oxidative conditions (Scheme 5 and Table 1, entries 15−19). As suspected, the action of freshly prepared MnO_2 (10.0 equiv, presumed to contain trace amounts of basic species capable of enolizing the carbonyl groups) in CH_2Cl_2 at 45 °C was sufficient to convert compound **25** to the rugulin-type structure **28** in essentially quantitative

Scheme 3. Postulated Mechanism for the Dimerization of the Monomeric Anthraquinone **23** through Enolization and a Diels−Alder (Red Arrows) or a Double Michael (Blue and Red Arrows) Reactions[a]



[a] Reagents and conditions: (a) CAN (2.0 equiv), MeCN:H_2O (1:1), 0 °C, 10 min; (b) CSA (0.125 equiv) or *i*-Pr_2NEt (0.125 equiv), CH_2Cl_2, 25 °C, 1 h, 94% or 90% over two steps, respectively. Abbreviation: CSA, (±)-10-camphorsulfonic acid.

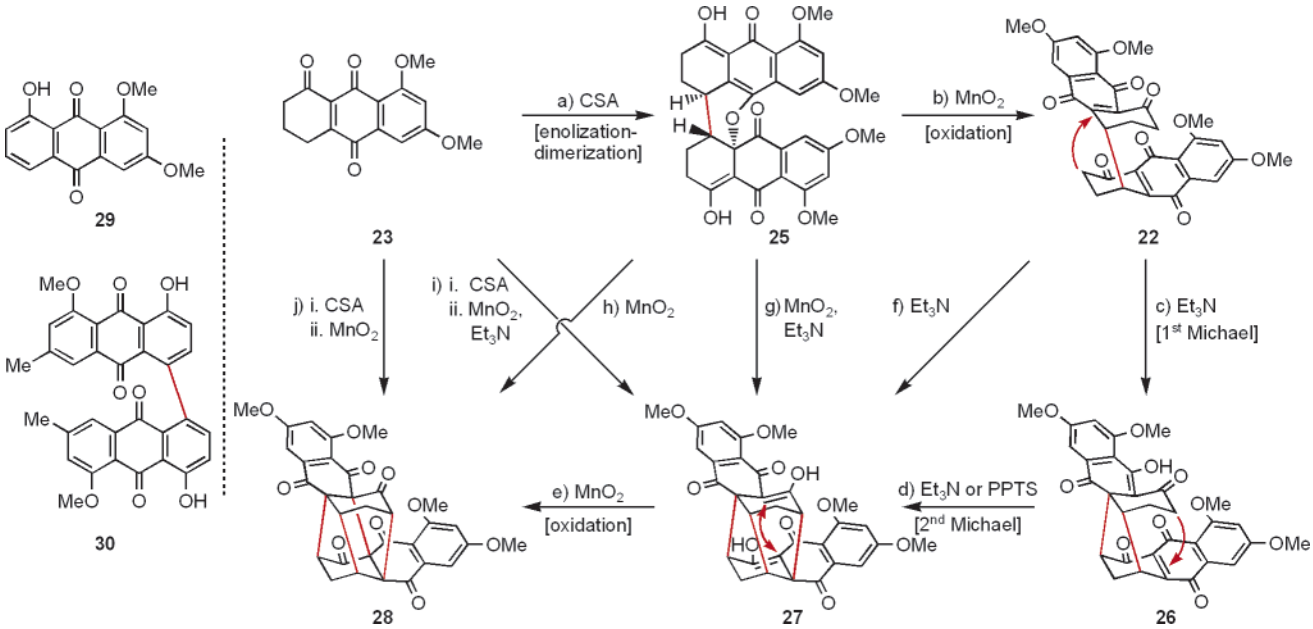
Scheme 4. Postulated Mechanism for the MnO_2 Oxidation of Compound **25** → **I** → **22** and **27** → **II** → **28**[a]



[a] This working hypothesis depiction is not based on any experimental evidence and is not meant to exclude any other mechanism. The proximity of the Mn=O and enol groups within **II** bodes well for an intramolecular interaction between them as part of the mechanism.

yield. TLC monitoring revealed that, indeed, this transformation proceeded through the detectable intermediates **22**, **26**, and **27**. It was also gratifying to observe that addition of Et_3N (10.0 equiv) to the above reaction mixture (**26**, MnO_2, CH_2Cl_2, 45 °C) suppressed the last step of the cascade, resulting in the

Scheme 5. The Cytoskyrin Cascades[a]



[a] Reagents and conditions: (a) CSA (0.125 equiv), CH_2Cl_2, 25 °C, 1 h, 94%; (b) MnO_2 (10.0 equiv), CH_2Cl_2, 25 °C, 1 h, 83%; (c) Et_3N (2.0 equiv), CH_2Cl_2, 25 °C, 1 h, 65%; (d) Et_3N (5.0 equiv), CH_2Cl_2, 45 °C, 16 h, 95% or PPTS (2.0 equiv), $ClCH_2CH_2Cl$, 100 °C, 24 h, 65%; (e) MnO_2 (10.0 equiv), CH_2Cl_2, 25 °C, 20 h, 95%. Cascade sequences: (f) **22** → **27**, Et_3N (5.0 equiv), CH_2Cl_2, 45 °C, 16 h, 95%; (g) **25** → **27**, MnO_2 (10.0 equiv), Et_3N (5.0 equiv), CH_2Cl_2, 25 → 45 °C, 20 h, 100%; (h) **25** → **28**, MnO_2 (10.0 equiv), CH_2Cl_2, 45 °C, 48 h, 100%; (i) **23** → **27**, CSA (0.125 equiv), CH_2Cl_2, 25 °C, 1 h; then MnO_2 (10.0 equiv), Et_3N (10.0 equiv), 45 °C, 36 h, 66%; (j) **23** → **28**, CSA (0.125 equiv), CH_2Cl_2, 25 °C, 1 h; then MnO_2 (10.0 equiv), CH_2Cl_2, 25 °C, 88 h, 75%.

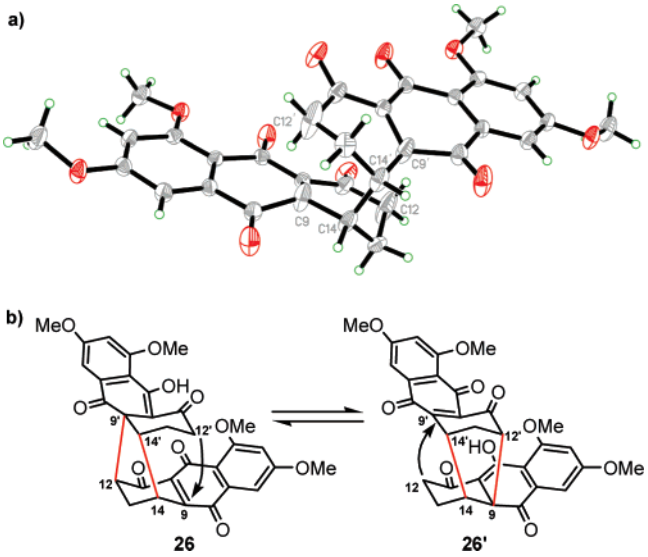


Figure 3. (a) ORTEP drawing of compound **26** (drawn at 30% probability level) obtained by X-ray crystallographic analysis (using *C*2/*c* space group) at ambient temperature. (b) Postulation of a possible equilibration between **26** and **26′**.

formation of the cytoskyrin-type structure **27**, again in essentially quantitative yield.

A more stunning cascade was realized after screening a number of other conditions (see Scheme 5 and Table 1, entries 20−23). Thus, upon exposure to CSA (0.125 equiv) in CH_2Cl_2 at 25 °C, monomeric anthraquinone **23** was observed (by TLC analysis) to transform completely to the heptacyclic dimeric compound **25**, at which time MnO_2 (excess) and Et_3N (10.0 equiv) were added and the reaction mixture was heated to 45 °C with stirring for 36 h, leading, through fleeting intermediates **22** and **26**, to the nonacyclic cytoskyrin-type structure **27** in 66% yield. The ultimate cascade in this series, however, was the one that transformed the monomeric anthraquinone precursor **23** into the rugulin model system **28** (Scheme 5 and Table 1, entry 24), an event that occurred upon sequential exposure of the former compound in CH_2Cl_2 to CSA (0.125 equiv) at 25 °C, followed, upon complete conversion (TLC analysis) to dimer **25**, by addition of MnO_2 (excess) to the same reaction vessel and heating to 45 °C for 36 h. Proceeding in 75% overall yield, this stunning cascade encompasses an initial enolization step followed by a stereoselective dimerization reaction, an oxidative collapse of an oxygen bridge, two intramolecular Michael reactions, and an oxidative coupling to form a carbon−carbon bond.

X-ray Crystallographic Analysis of Compound 26. During these investigations, we had the opportunity to make some interesting observations regarding the structure of the dimeric compound **26**, as revealed by X-ray crystallographic studies and NMR spectroscopy. Thus, our X-ray crystallographic analysis of **26**[15] pointed to a symmetrical structure with only one carbon−carbon bond linking the two monomeric units of the molecule (see ORTEP drawing in Figure 3a), and yet NMR spectroscopic analysis clearly supported the non-symmetrical structure (i.e., **26**) originally assigned to it. This rather rare phenomenon is reminiscent of Ermer's classic observation with secododecahedrane,[16] which exhibited the symmetrical dodecahedrane structure by X-ray analysis but was correctly contradicted by NMR spectroscopic analysis. As in that case, we attributed this abnormality to a crystal disorder[17] or a dynamic equilibrium involving a two-proton shuttle that involves rupture of the C12−C9′ bond within **26** (**26** → **26′**, Figure 3b) while

(16) Ermer, O. *Angew. Chem., Int. Ed. Engl.* **1983**, *22*, 251−252. We thank Prof. Scott Rychnovsky for pointing out this reference to us.
(17) (a) Dunitz, J. D.; Filippini, G.; Gavezzotti, A. *Helv. Chim. Acta* **2000**, *83*, 2317−2335. (b) Dunitz, J. D.; Bernstein, J. *Acc. Chem. Res.* **1995**, *28*, 193−200 and references therein. We thank Prof. Jack Dunitz for pointing out these articles to us and for his stimulating comments.

Table 1. Screening of Conditions for the Various Steps and Cascade Sequences Depicted in Scheme 5[a]

Entry	Starting material	Conditions	Product (% yield)
		26 → **27** transformation	
1	**26**	proton sponge, CH_2Cl_2, 45 °C, 3 h	NR
2	**26**	MW, 110 °C, 30 min	NR
3	**26**	PPTS, toluene, 100 °C, 2 h	NR
4	**26**	PPTS, $ClCH_2CH_2Cl$, 80 °C, 48 h	**27** (65)
5	**26**	py, 100 °C, 12 h	**27** (63)
6	**26**	py, MW, 15 min, 80 °C, then 30 min, 120 °C (x2)	**27** (80)
7	**26**	Et_3N, CH_2Cl_2, 45 °C, 16 h	**27** (95)
		22 → **27** transformation	
8	**22**	DBU, THF, 25 °C, 12 h	**29** or **30** mainly[a]
9	**22**	*hv*, CH_2Cl_2, 3 h	complex mixture
10	**22**	*hv*, THF, 5 h	complex mixture
11	**22**	CF_3SO_3H, CH_2Cl_2, 25 °C, 2 h	**29** or **30** mainly[a]
12	**22**	LDA, THF, –78 → 25 °C, 8 h	**25** (50)
13	**22**	Et_3N, CH_2Cl_2, MW, 80 °C, 6 h	**27** (91)
14	**22**	Et_3N, CH_2Cl_2, 45 °C, 16 h	**27** (95)
		25 → **27** transformation	
15	**25**	py, O_2, 45 °C, 2 h	**29** mainly[a]
16	**25**	MW, py, O_2, 80 °C, 30 min	**29** mainly[a]
17	**25**	DDQ, Et_3N, CH_2Cl_2, 25 °C, 16 h	**22** (10) + **26** (20)[a]
18	**25**	dry Cs_2CO_3, THF, 25 °C, 3 h	**27** (57)
19	**25**	MnO_2, Et_3N, CH_2Cl_2, 25 → 45 °C, 20 h	**27** (100)
		23 → **27** transformation	
20	**23**	Dowex, CH_2Cl_2, 25 °C, 48 h	**29** mainly[a]
21	**23**	Et_3N, CH_2Cl_2, 25 °C, 3 h	**29** (70) + **25** (30)
22	**23**	*i*-Pr_2NEt, CH_2Cl_2, 25 °C, 2 h	**25** (90)
23	**23**	CSA, CH_2Cl_2, 25 °C, 1 h; then MnO_2, Et_3N, 45 °C, 36 h	**27** (66), **29** (15)
		23 → **28** transformation	
24	**23**	CSA, CH_2Cl_2, 25 °C, 1 h; then MnO_2, 25 °C, 88 h	**28** (75)

[a] Complex reaction. Abbreviations: NR, no reaction; MW, microwaves; DDQ, 2,3-dichloro-5,6-dicyano-1,4-benzoquinone; LDA, lithium diisopropylamine.

the C12′−C9 bond within **26′** forms. Structures **26** and **26′** are identical, and the X-ray crystallographic analysis reveals only an average picture of the two, with bond lengths C12−C9′ = C12′−C9 = 2.372 Å and C14−C14′ = 1.56 Å.[18]

Abortive Attempts toward Cytoskyrin. Following our successful model studies described above, we then considered the challenging task of synthesizing cytoskyrin (**1a**) and its relatives, all of which contain a β-hydroxy group at the C2 and C2′ positions. A cursory inspection of these structures reveals that, while they may be immune to H_2O elimination by virtue of Bredt's rule,[19] which forbids the formation of a bridgehead double bond—the product of such elimination—the precursors leading up to them are not. On the contrary, all such intermediates (see Scheme 1) are prone to elimination/aromatization and/or fragmentation/elimination; therefore, our initial inclination was to introduce a surrogate moiety for the desired hydroxy group at the C2 and C2′ positions.

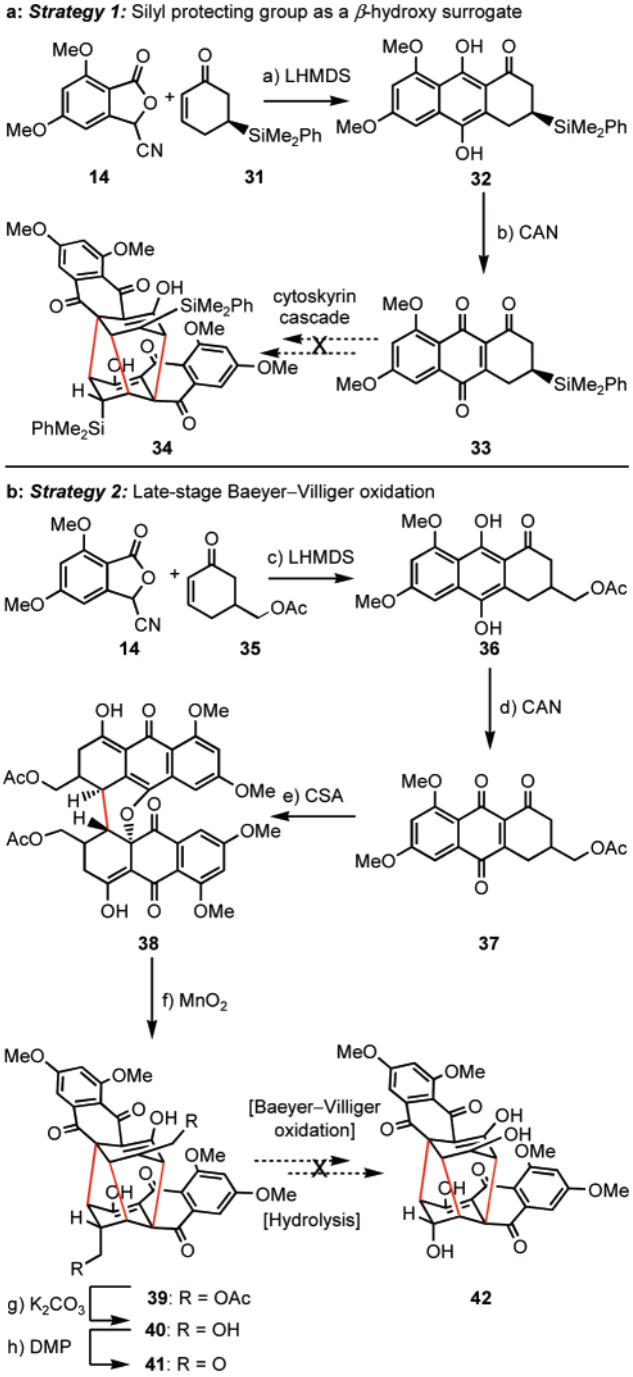
In our first attempt to test this hypothesis, we adopted the $SiMe_2Ph$ group as a surrogate moiety with the expectation that we could convert it to the desired hydroxy group under Tamao−Fleming oxidation conditions[20] at the end of the cytoskyrin cascade (Scheme 6a). Thus, Hauser annulation of the known cyclohexenone **31**[21] with nitrile **14** led to dihydroquinone **32** (65% yield). CAN oxidation of the latter compound led to the formation of the labile quinone **33** (detected by TLC analysis, not stable enough for isolation), which, however, failed to enter into a productive cascade or even dimerization to any characterizable products. We attributed the failure of quinone **33** to participate in the expected cytoskyrin cascade to the bulkiness of the $SiMe_2Ph$ group.

We then reasoned that a CH_2OAc group—a considerably smaller group than the $SiMe_2Ph$ moiety—may provide us with a more willing substrate for the desired cytoskyrin cascade, at the end of which we were planning a short sequence (deprotection, oxidation, Baeyer−Villiger oxidation,[22] and hydrolysis of the resulting formate ester) for its conversion to the desired hydroxy group. To this end, we combined nitrile **14** with the known cyclohexenone **35**[23] in the standard Hauser annulation reaction and obtained dihydroquinone **36** in 49% yield (Scheme 6b). Oxidation of the latter compound with CAN then furnished, in quantitative yield, quinone **37**, whose exposure to catalytic amounts of CSA in CH_2Cl_2 at ambient temperature led to dimer **38** in 84% yield. When treated with MnO_2 and Et_3N in CH_2Cl_2 at ambient temperature, compound **38** underwent dismantling and reconstitution through the cytoskyrin cascade to afford the cytoskyrin-like structure **39** in 63% yield. Basic cleavage of the acetate groups from the latter compound was effected by the action of K_2CO_3 in MeOH for 3 h at ambient temperature, leading to tetraol **49** in 96% yield. Oxidation of the primary alcohols in **49** with Dess−Martin periodinane in CH_2Cl_2 at ambient temperature then led to quantitative formation of the bisaldehyde **41**. All attempts, however, to effect the intended Baeyer−Villiger reaction proved unproductive, leading to decomposition and/or unidentifiable products instead. The abortive attempts to realize this transformation in similar substrates by the Snider group[24] added further reasons for abandoning this route, and so we opted for a different strategy.

(18) The X-ray measurements were initially performed at room temperature, and the crystal structure was solved and refined in the centrosymmetric space group *C*2/*c* with $Z = 4$, following the general conventions and statistical analysis of data ($|E^2 - 1| = 0.91$). Accordingly, the molecules of **26** were observed to reside on (average) crystallographic twofold axes. The distance between the carbon atoms C9 and C12′, however, was as short as 2.37(2) Å, which was far smaller than the expected van der Waals sum of 3.5 Å. Furthermore, the anisotropic temperature factor ellipsoids of both these atoms were distinctly elongated along their line of separation, suggesting that one Michael addition reaction could have taken place, resulting in bond formation between C9 and C12′ and leading to the nonsymmetrical structure **26**, which could exist in dynamic equilibrium with its identical "regioisomer" (**26′**, Figure 3b). Subsequent crystal structure solution and refinement in the lower noncentrosymmetric space group *Cc*[32] gave a slightly nonsymmetrical molecular structure with C9−C12′ and C9′−C12 distances of 2.24(2) and 2.52(1) Å, respectively. Interestingly, when the same crystal was subjected to low-temperature X-ray measurements (123 K), these bond lengths diverged even further, as revealed by the values of 1.91(2) and 2.81(2) Å computed by structure solution and refinement in *Cc*. When the *C*2/*c* space group was used, the C9−C12′ distance was approximately the same as the room-temperature distance of 2.37(2) Å. These distances were even more drastically different [1.66(2) and 2.95(2) Å, respectively] in *Cc* when the atoms were refined isotropically. Thus, apparently partial dynamic ordering of the crystal structure occurs on cooling. Interestingly, while both secododecahedrane[16] and the present example involve carbon atoms at close proximity to each other, in the former case the X-ray analysis revealed a bond, whereas in the latter it missed a bond.

(19) (a) Bredt, J.; Thouet, H.; Schmitz, J. *Liebigs Ann. Chem.* **1924**, *437*, 1−13. (b) Wiseman, J. R. *J. Am. Chem. Soc.* **1967**, *89*, 5966−5968. (c) Maier, W. F.; Schleyer, P. v. R. *J. Am. Chem. Soc.* **1981**, *103*, 1891−1900.
(20) (a) Fleming, I.; Henning, R.; Plaut, H. *J. Chem. Soc., Chem. Commun.* **1984**, 29−31. (b) Tamao, K.; Ishida, N.; Tanaka, T.; Kumada, M. *Organometallics* **1983**, *2*, 1694−1696. (c) See also a series of papers from Fleming in *J. Chem. Soc., Perkin Trans. 1* **1992**, 3295−3302.
(21) Sarakinos, G.; Corey, E. J. *Org. Lett.* **1999**, *1*, 811−814.
(22) Renz, M.; Meunier, B. *Eur. J. Org. Chem.* **1999**, 737−750.
(23) (a) Smith, A. B.; Richmond, R. E. *J. Am. Chem. Soc.* **1983**, *105*, 575−585. (b) van Tamelen, E. E.; Hildahl, G. T. *J. Am. Chem. Soc.* **1956**, *78*, 4405−4412.
(24) Snider, B. B.; Gao, X. *J. Org. Chem.* **2005**, *70*, 6863−6869.

Scheme 6. Abortive Strategies toward 2,2′-*epi*-Cytoskyrin A (**2a**)[a]



[a] Reagents and conditions: (a) **14** (1.1 equiv), THF, LHMDS (1.1 equiv), −78 °C, 1 h; then **31** (1.0 equiv), −78 → 25 °C, 4 h, 65%; (b) CAN (2.0 equiv), MeCN:H_2O (9:1), 0 °C, 10 min, product not stable enough for isolation; (c) **14** (1.01 equiv), THF, LHMDS (1.01 equiv), −78 °C, 1 h; then **35** (1.0 equiv), −78 → 25 °C, 1 h, 49%; (d) CAN (2.0 equiv), MeCN:H_2O (9:1), 0 °C, 1 h, 100%; (e) CSA (0.1 wt equiv), CH_2Cl_2, 0.2 M, 25 °C, 12 h, 84%; (f) MnO_2 (5.0 equiv), CH_2Cl_2; then Et_3N (10.0 equiv), 25 → 40 °C, 16 h, 63%; (g) K_2CO_3, MeOH, 25 °C, 3 h, 96%; (h) DMP, CH_2Cl_2, 25 °C, 1 h, 92%. Abbreviation: DMP, Dess−Martin periodinane.

Total Syntheses of 2,2′-*epi*-Cytoskyrin A and Rugulosin and Analogues. Having failed to employ a surrogate group for the hydroxy groups at C2 and C2′ positions, and despite the danger of elimination, we decided to attempt the cytoskyrin cascade with substrates carrying free or protected hydroxy groups at those positions, as shown in Figure 4. We were encouraged to do so by our hypothesis that nature may very well be using such substrates in the biogenesis of these bisanthraquinone systems and by the expectation that, should we succeed in entering into a cytoskyrin-type scaffold and beyond under suitably mild conditions, those structures should be protected toward elimination by Bredt's rule.

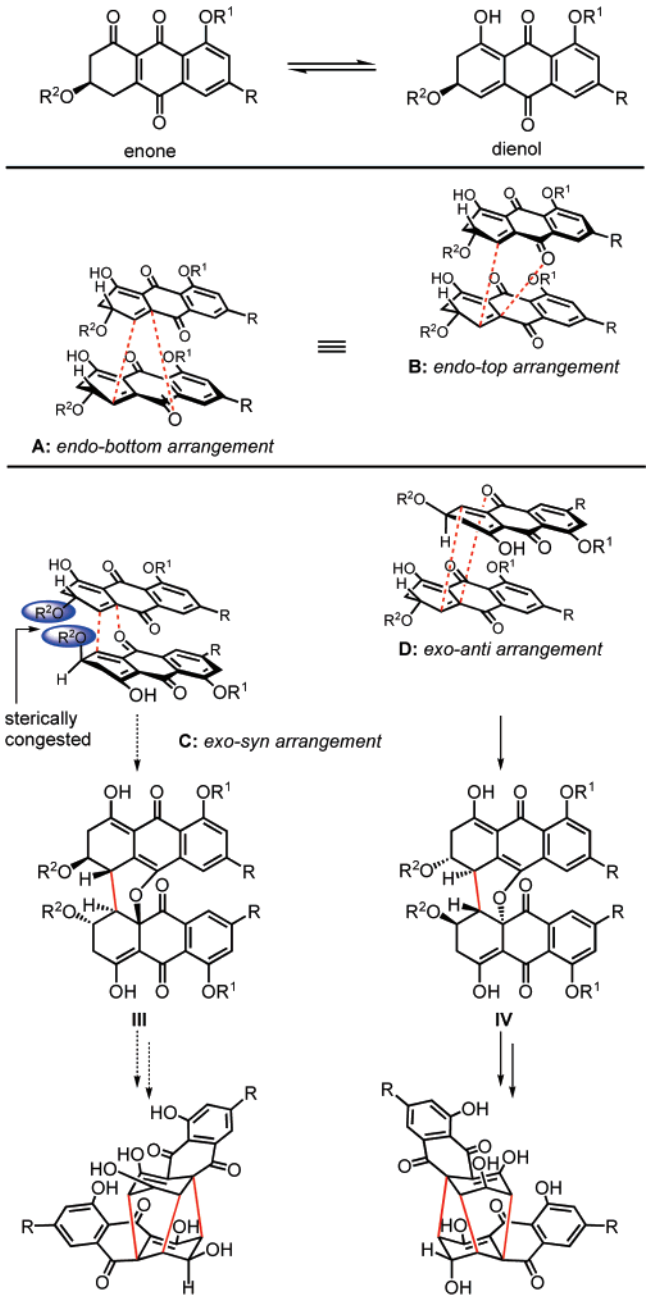


Figure 4. Stereoselectivity considerations for the dimerization step of anthradihydroquinones to modified bisanthraquinones. The shown enantiomers of the starting anthraquinones correspond to *ent*-**1a**, *ent*-**1b**, **2a**, and **2b**. Should the pathway involving arrangement **C** be possible, the opposite enantiomers would be needed in order to obtain the naturally occurring form of cytoskyrin A (**1a**) and graciliformin (**1b**).

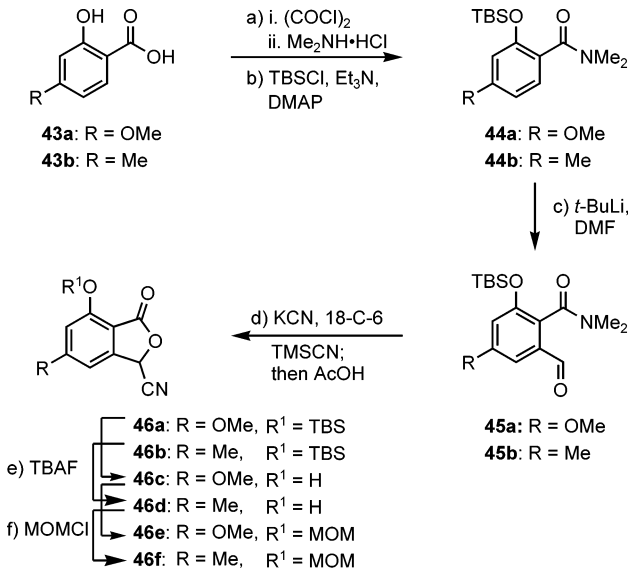
Once formed from their enone counterparts (Figure 4, top), the dienol forms of the monomeric anthraquinone substrates may approach each other for dimerization in four different ways (**A**−**D**, Figure 4). **A** and **B** represent the *endo* arrangements, while **C** and **D** depict the *exo* arrangements, each pair being further

distinguished by the faciality of the approach (assuming one of the monomers is stationary, as shown), a differentiation that leads to the *endo*-bottom (**A**), the *endo*-top (**B**), the *exo*-syn (**C**), and the *exo*-anti (**D**) alignments. The two *endo* arrangements (**A** and **B**) are equivalent and are highly unlikely due to steric constraints (R′O syn to H). The *exo*-syn arrangement (**C**) is even more constrained (R′O syn to R′O) and, therefore, unlikely to occur. Such an arrangement (**C**) may lead to *ent*-cytoskyrin (*ent*-**1a**) or *ent*-graciliformin (*ent*-**1b**) through the intermediacy of the dimeric compounds **III**, depending on the substrates used. The most likely arrangement is that represented by **D** (*exo*-anti), in which the two R′O groups are placed opposite to each other, thereby allowing optimum comfort with regard to steric interactions. As the most favorable arrangement, this approach (**D**) is geared to form 2,2′-*epi*-cytoskyrin A (**2a**) and rugulosin (**2b**) through the intermediacy of dimer **IV**, as shown in Figure 4. It should be noted that the chirality of the starting monoquinone dictates the absolute stereochemistry of the cage (helical stereochemistry) structural motif in the product, while the diastereoselectivity of the dimerization step controls the stereochemistry of the C2 and C2′ centers relative to the rest (epimeric stereochemistry). According to this stereochemical analysis, the depicted enantiomer of the starting anthraquinones (Figure 4) should correspond to *ent*-**1a**, *ent*-**1b**, **2a**, and **2b**. The chiral centers at C2 and C2′ of the compounds **III** and **IV**, in combination with the helical nature of the cage formed thereafter, improvise for the occurrence of two possible diastereoisomers for each intermediate member of the cascade (one set of which is not expected to form on the basis of the above stereochemical considerations). Should the cascade involving arrangement **C** be possible, the opposite enantiomer of the starting anthraquinones would be required in order to obtain the naturally occurring forms of cytoskyrin (**1a**) and graciliformin (**1b**).

The next task was the generation of the required monomeric anthradihydroquinones (i.e., **59a**−**h**, Scheme 10) with different protecting groups (R^2) on the C2 hydroxyl group in order to explore their dimerization and subsequent cascade reactions. As previously, we chose the Hauser annulation as the most expedient means to construct these substrates, requiring the availability of a series of nitriles (**46a**−**f**, Scheme 7) and enones (**51a**−**e**, Scheme 8).

Scheme 7 summarizes the synthesis of nitriles **46a**−**f** from commercially available salicylic acids **43a** and **43b**. Exposure of **43a** or **43b** to $(COCl)_2$ and catalytic amounts of DMF in CH_2Cl_2 at 0 °C afforded the corresponding acid chlorides, which were quenched with $Me_2NH{\cdot}HCl$ to give the expected amides. The resulting amides were subsequently protected with TBSCl in the presence of Et_3N and catalytic amounts of DMAP in CH_2Cl_2 at ambient temperature to give the desired protected amides **44a** and **44b** in 93 and 87% yields, respectively, over the three steps. *ortho*-Lithiation of the amides **44a** or **44b** using *t*-BuLi and TMEDA at −78 °C and subsequent quenching with freshly distilled DMF yielded the corresponding aldehydes **45a** and **45b** in 88 and 85% yields, respectively. Exposure of the latter compounds to TMSCN and catalytic amounts of KCN and [18]-crown-6, followed by treatment with AcOH, afforded the deprotected nitriles **46c** (78%) and **46d** (65%) as the major compounds, along with the TBS-protected nitriles **46a** (15%) and **46b** (20%), respectively. TBAF deprotection of the latter compounds afforded the free hydroxy nitriles **46c** and **46d** in 80 and 78% yields, respectively, which were then treated with MOMCl and *i*-Pr_2NEt at 0 °C to furnish the required nitrile building blocks, **46e** and **46f**, in 90 and 85% yields, respectively, as shown in Scheme 7.

Scheme 7. Construction of Nitriles **46a**−**46f**[a]



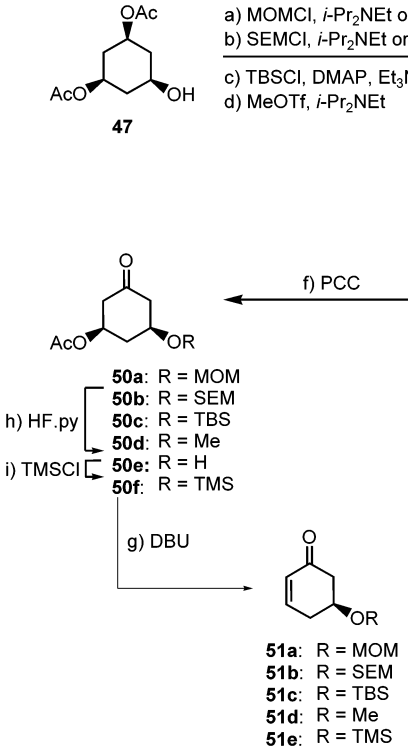
[a] Reagents and conditions: (a) i. $(COCl)_2$ (1.25 equiv), DMF (cat.), CH_2Cl_2, 0 → 25 °C, 2 h; ii. $Me_2NH{\cdot}HCl$ (1.5 equiv), Et_3N (4.0 equiv), CH_2Cl_2, 0 °C, 30 min, 25 °C, 16 h; (b) TBSCl (1.1 equiv), Et_3N (1.5 equiv), CH_2Cl_2, 25 °C, 16 h; 93% over three steps (**44a**), 87% (**44b**); (c) TMEDA (2.5 equiv), *t*-BuLi (2.5 equiv), DMF (3.0 equiv), THF, −78 → 25 °C, 12 h, 88% (**45a**), 86% (**45b**); (d) KCN (0.2 equiv), 18-C-6 (0.2 equiv), TMSCN (1.4 equiv), CH_2Cl_2, 25 °C, 4 h; then AcOH, 12 h, 15% (**46a**), 78% (**46c**), 20% (**46b**), 65% (**46d**); (e) TBAF (2.0 equiv), CH_2Cl_2, 25 °C, 5 h, 80% (**46c**), 78% (**46d**); (f) MOMCl (1.5 equiv), *i*-Pr_2NEt (1.2 equiv), CH_2Cl_2, 0 °C, 1 h, 90% (**46e**), 85% (**46f**). Abbreviations: DMAP, 4-dimethylaminopyridine; TBS, *tert*-butyldimethylsilyl; DMF, *N*,*N*′-dimethylformamide; TMEDA, *N*,*N*,*N*′,*N*′-tetramethylethylenediamine; TBAF, *tetra*-*n*-butylammonium fluoride; MOM, methoxymethyl.

The protected enones **51a**−**e** required for the Hauser annulation were synthesized as summarized in Scheme 8. Thus, protection of the known diacetate **47**[25] with MOMCl, SEMCl, TBSCl, or MeOTf, followed by desymmetrisation using porcine liver esterase (PLE) in pH 8 buffer and *t*-BuOH as cosolvent, afforded the corresponding alcohols **49a**−**d** with the following respective yields over the two steps: **49a**, 95%; **49b**, 100% (based on 60% recovered starting material **48b**); **49c**, 92% (based on 51% recovered starting material **48c**); and **49d**, 58% (based on 40% recovered starting material **48d**). PCC oxidation, followed by brief exposure to DBU at 25 °C, then furnished the desired enones **51a**−**d** in 68, 96, 71, and 90% yields, respectively, over the two steps. Dilsilyation of **50c** using HF·py in CH_2Cl_2 at 25 °C for 1 h, followed by protection with TMSCl and subsequent brief exposure to DBU, afforded the TMS-protected enone **51e** in 84% yield over the three steps. The enantiomeric excesses of the enones **50a**−**e** were determined by Mosher's ester analysis of the corresponding alcohols **49a**−**d** (**49a**, 70% ee; **49b**, −20% ee; **49c**, 90% ee;[25] **49d**, 65% ee).

In an effort to improve the asymmetric synthesis of the enone partner, we sought to develop an alternative pathway, as exemplified for enone **51a** in Scheme 9. Featuring a Brown allylation[26] and a ring-closing metathesis reaction, this route

(25) Kalkote, U. R.; Ghorpade, S. R.; Chavan, S. P.; Ravindranathan, T. *J. Org. Chem.* **2001**, *66*, 8277−8281.

Scheme 8. Construction of Enones **51a**−**51e** through Enzymatic Resolution[a]



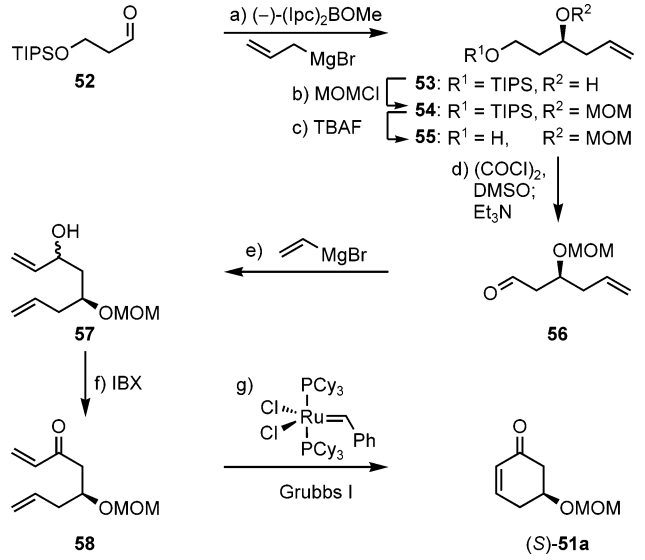
[a] Reagents and conditions: (a) MOMCl (3.8 equiv), *i*-Pr_2NEt (3.8 equiv), CH_2Cl_2, 25 °C, 1.5 h; (b) SEMCl (3.8 equiv), *i*-Pr_2NEt (3.8 equiv), CH_2Cl_2, 25 °C, 1.5 h; (c) TBSCl (2.0 equiv), DMAP (0.25 equiv), Et_3N (2.5 equiv), CH_2Cl_2, 25 °C, 24 h; (d) MeOTf (2.0 equiv), *i*-Pr_2NEt (2.2 equiv), CH_2Cl_2, 25 °C, 24 h; (e) PLE (1.0 equiv), phosphate buffer pH = 8 (0.1 M), *t*-BuOH (8% v/v), 25 °C, 4−48 h, 95% over two steps, 70% ee (**49a**), 40% (100% based on 60% recovered starting material), −20% ee (**49b**), 45% (92% based on 51% recovered starting material), 90% ee (**49c**), 35% (58% based on 40% recovered starting material), 65% ee (**49d**). For R = MOM, SEM, TBS, or Me: (f) PCC (3.0 equiv), CH_2Cl_2, 25 °C, 12 h; (g) DBU (1.0 equiv), CH_2Cl_2, 25 °C, 10 min, 68% over two steps (**51a**), 96% (**51b**), 71% (**51c**), 90% (**51d**). For R = TMS: (h) HF·py (10.0 equiv), CH_2Cl_2, 25 °C, 1 min; then TMSOMe, 30 min; (i) TMSCl (2.0 equiv), *i*-Pr_2NEt (2.2 equiv), CH_2Cl_2, 25 °C, 1.5 h, (g) DBU (1.0 equiv), 10 min, 25 °C, 84% over three steps (**51e**). Abbreviations: SEM, 2-(trimethylsilyl)ethoxy; PLE, porcine liver esterase; PCC, pyridinium chlorochromate; DBU, 1,8-diazabicyclo[5.4.0]undec-7-ene.

began with aldehyde **52**,[27] whose addition to a preformed solution of chiral allylborane, generated from (−)-B-methoxydiisopinocamphenylborane and allyl magnesium bromide in Et_2O at −78 °C, afforded allylic alcohol **53** in 83% yield. Mosher's ester analysis of this alcohol indicated that it was formed in 86% ee. Sequential protection of **53** with MOMCl in the presence of *i*-Pr_2NEt at 0 → 25 °C in CH_2Cl_2 and desilylation (TBAF, THF, 25 °C) furnished hydroxy compound **55**, which was oxidized under Swern conditions to yield the corresponding ketone **56**, to which was added vinyl magnesium bromide (3.0 equiv), leading to a 1:1 mixture of diastereomeric alcohols **57** in 64% yield over the four steps. IBX oxidation of this mixture led to ketone **58**, which underwent smooth ring-closing metathesis in the presence of Grubbs I catalyst to afford the desired enone **51a** in 91% yield for the two steps.

(26) (a) Hertweck, C.; Boland, W. *J. Prakt. Chem.* **1999**, *341*, 83−87. (b) Jadhav, P. K.; Bhat, K. S.; Perumal, T.; Brown, H. C. *J. Org. Chem.* **1986**, *51*, 432−439. (c) Nicolaou, K. C.; Ahn, K. H. *Tetrahedron Lett.* **1989**, *30*, 1217−1220.

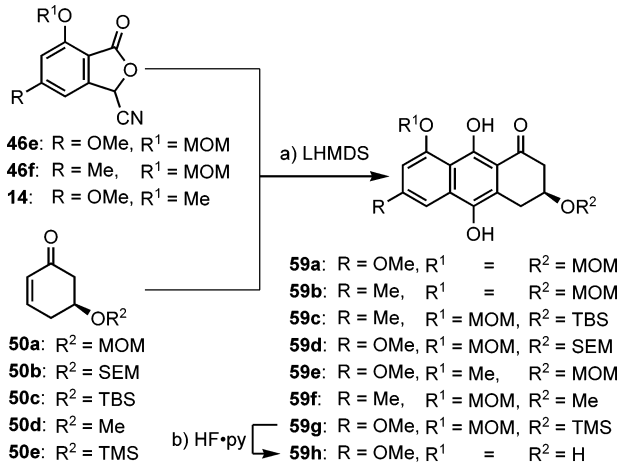
(27) Kopecky, D. J.; Rychnovsky, S. D. *J. Am. Chem. Soc.* **2001**, *123*, 8420−8421.

Scheme 9. Construction of Enone (*S*)-**51a** through Ring-Closing Metathesis[a]



[a] Reagents and conditions: (a) (−)-B-methoxydiisopinocamphenylborane (3.0 equiv), Et_2O, −78 °C, allylmagnesium bromide (3.0 equiv); then **52** (1.0 equiv), 3 h; (b) MOMCl (1.5 equiv), *i*-Pr_2NEt (1.25 equiv), CH_2Cl_2, 25 °C, 12 h; (c) TBAF (1.1 equiv), 0 → 25 °C, 8 h, 74% over three steps; (d) $(COCl)_2$ (1.2 equiv), DMSO (2.4 equiv), Et_3N (3.9 equiv), CH_2Cl_2, 0 °C, 1 h; (e) vinylmagnesium bromide (3.0 equiv), THF, −78 °C, 30 min; then 0 °C, 30 min, 72% over two steps; (f) IBX (1.2 equiv), DMSO, 25 °C, 2 h; (g) Grubbs I catalyst (0.04 equiv), 0.1 M solution, CH_2Cl_2, 30 °C, 6 h, 91% over two steps. Abbreviation: IBX, 2-iodoxybenzoic acid.

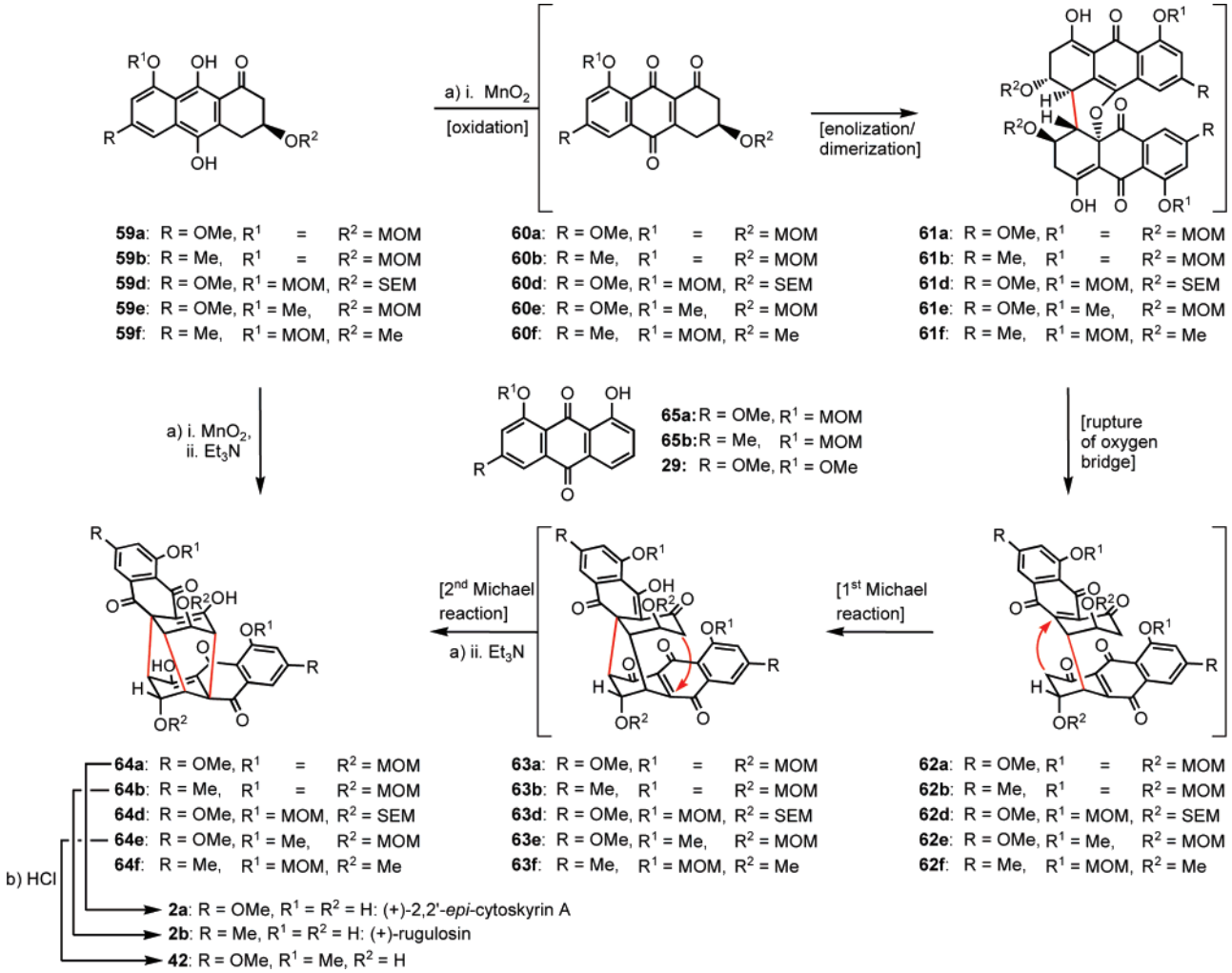
Scheme 10. Synthesis of the Anthradihydroquinone Monomers **59a**−**59h**[a]



[a] General conditions for Hauser annulation: (a) nitrile (1.01 equiv), LHMDS (1.01 equiv), THF, −78 °C, 1 h, enone (1.00 equiv), −78 °C, 1 h, −78 → 25 °C, NH_4Cl workup, **59a** (80%), **59b** (91%), **59c** (69%), **59d** (98%), **59e** (83%), **59f** (52%), **59g** (41%); (b) HF·py (10.0 equiv), CH_2Cl_2, 1 min, 75%.

Total Syntheses of 2,2′-*epi*-Cytoskyrin A and Rugulosin. With all the required fragments available, we then proceeded to construct the monomeric anthradihydroquinones needed for the pending cascades toward the targeted bisanthraquinone natural products. Thus, treatment of the corresponding nitriles (**46e**,**f**, **14**) in THF with LHMDS at −78 °C, followed by addition of the appropriate enone (**51a**−**e**) (−78 → 0 °C), furnished, upon addition of saturated aqueous NH_4Cl and standard workup, the anthradihydroquinones (**59a**−**h**) required for the intended cytoskyrin cascade. In considering these cascades, we were encouraged and guided by a number of

Scheme 11. Cytoskyrin Cascades: Synthesis of (+)-2,2′-*epi*-Cytoskyrin A (**2a**), (+)-Rugulosin (**2b**), and Analogues **64a**−**e** and **42** from Anthradihydroquinone **59a**,**b**,**d**−**f**[a]



[a] Reagents and conditions: (a) MnO_2 (1.0 wt equiv), CH_2Cl_2 (0.4−0.5 M), 25 °C, 1 h; then Et_3N (5.0 equiv), CH_2Cl_2, 25 → 45 °C, 12 h; 60% over seven steps (**64a**), 50% (**64b**), 34% (**64d**), 40% (**64e**), 40% (**64f**); (b) HCl (conc; excess), MeOH:THF (20:1), 60 °C, 12 h, 93% (**2a**), 98% (**2b**), 91% (**42**).

observations and considerations. First, we took comfort in the fact that freshly prepared MnO_2 was capable of initiating and propagating such reactions, as described above. Second, we were cognizant of the fact that elimination of the alkoxy group followed by aromatization could present a problem, but we were confident with regard to the stability of the advanced intermediates and final products by virtue of Bredt's rule. Third, we knew from our model studies that impurities carried through from the Hauser annulation reaction impeded the cascade, causing a shutdown after the initial dimerization step and funneling of varying amounts of material into the undesired pathway toward aromatized products (i.e., **65a**,**b** and **29**).

Employing purified anthradihydroquinones **59a** or **59b** and sequential addition of MnO_2 and Et_3N led to the development of an impressive seven-step cascade sequence that involved the conversion of these tricyclic substrates to the nonacyclic systems **63a** and **64b** in 60 and 50% yields, respectively. Proceeding through the intermediacy of **60a** or **60b**, **61a** or **61b**, **62a** or **62b**, and **63a** or **63b**, these sequences feature alternate oxidations and double Michael reactions, as exemplified in Scheme 11. These cascades were initiated by treatment of **59a** or **59b** with MnO_2 (10 wt equiv) in CH_2Cl_2 (0.35−0.5 M) for 30 min at ambient temperature, conditions that led to the initial generation of **61a** or **61b** and the complete consumption of the starting material as revealed by TLC analysis, followed by dilution with CH_2Cl_2 (5- to 7-fold) and stirring until the formation of **63a** or **63b** was complete. Addition of Et_3N (5.0 equiv) and warming to 45 °C with stirring for 6 h then furnished **64a** or **64b**. Treatment of the latter compounds with concentrated HCl in a mixture of MeOH:THF (20:1) at 60 °C resulted in global deprotection, affording (+)-2,2′-*epi*-cytoskyrin A (**2a**, 93% yield) and (+)-rugulosin (**2b**, 98% yield). The ^{1}H NMR spectroscopic data of synthetic rugulosin matched those reported[4b] for the naturally occurring substance. Although (+)-2,2′-*epi*-cytoskyrin A (**2a**) had not yet been isolated from nature at the time of its total synthesis, its stereochemistry was anticipated on the basis of the arguments presented above (Figure 4) and proven by spectroscopic techniques. Thus, the ^{1}H NMR spectrum of the synthetic (+)-2,2′-*epi*-cytoskyrin A (**2a**) exhibited doublets for H3 and H3′ (δ = 2.85 ppm and J = 4.8 Hz, respectively) and H2 and H2′ (δ = 4.49 ppm and J = 4.8 Hz, respectively), while in the reported spectrum of natural cytoskyrin A (**1a**), both signals for H3 and H3′, and H2 and H2′, appeared as singlets at δ = 2.85 and 4.00 ppm, respectively. Had (+)-2,2′-*epi*-cytoskyrin A (**2a**) possessed the same stereochemistry at C2 and C2′ as cytoskyrin A (**1a**), its dihedral

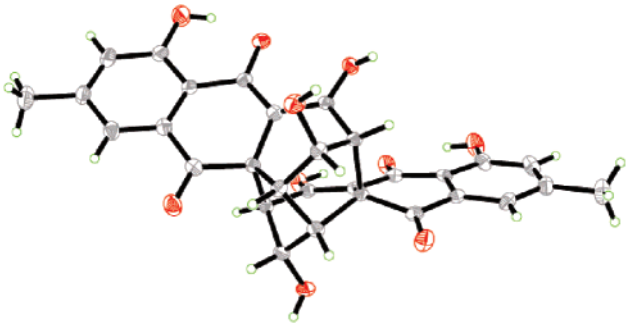
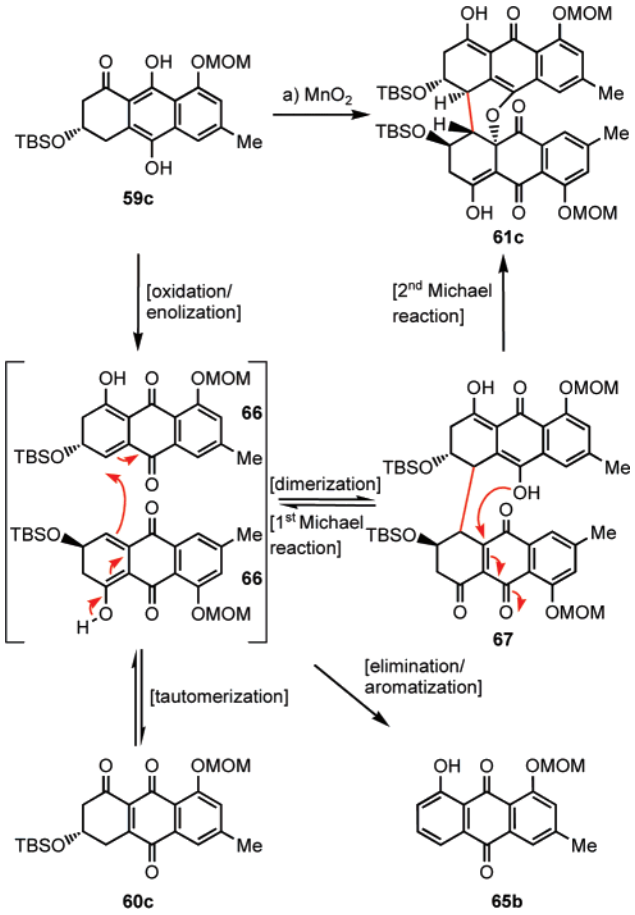


Figure 5. ORTEP drawing of rugulosin (**2b**) derived from X-ray crystallographic analysis. The molecule of Et_3N per molecule of **2b** is not shown. Crystallographic analysis did not reveal the absolute stereochemistry of the **2b**, which was assigned as shown on the basis of the absolute stereochemistry of its precursor **59a**.

angles H1/H1′−H2/H2′ and H2/H2′−H3/H3′ would have been 78 and 85°, respectively, on the basis of manual molecular models, and hence the ^{1}H NMR signals for H2/H2′ and H3/H3′ would have been singlets according to Ejiri et al.[2a] In addition, the X-ray crystallographic analysis[15] (see ORTEP drawing, Figure 5) of a crystal of synthetic rugulosin (**2b**), obtained by slow crystallization from acetone containing 1% Et_3N over MeOH vapors, confirmed its structure and indirectly rendered further support for the assigned structure for (+)-2,2′-*epi*-cytoskyrin A (**2a**) by virtue of the striking similarities between the ^{1}H NMR spectra of the two compounds.

Mechanistic Studies. As mentioned above, the cytoskyrin cascade was envisaged to proceed from the monoanthradihydroquinone substrates, through initial oxidation to the monoanthraquinone systems, followed by regioselective enolization and dimerization. The latter process could, in principle, proceed through either a concerted [4+2] cycloaddition reaction (hetero Diels−Alder reaction, see Scheme 3) or a sequential double Michael process, as shown in Scheme 12. In order to distinguish between the two pathways, we chose the TBS-protected anthradihydroquinone **59c** as a substrate for the cascade in the hope that the bulkiness of the TBS moiety as compared to the MOM group would serve to retard the progress of the cascade. It was further reasoned that such a slow-down of some of the reactions within the cascade may allow the isolation of some of the intermediates along the sequence, thus shining light on the mechanism involved. Thus, exposure of **59c** to MnO_2 (1.5 wt equiv) in CH_2Cl_2 (0.35 M) at ambient temperature for 30 min produced, in sequence, anthraquinone **60c**, dimer **67**, *O*-bridged dimer **61c**, and aromatized compound **65b** (Scheme 13). While compounds **61c** and **65b** were stable, **60c** and **67** were rather labile, but stable enough for purification and spectroscopic characterization (^{1}H NMR spectroscopy). It was also observed that, upon standing in $CDCl_3$ at ambient temperature, dimer **67** was slowly and quantitatively converted to *O*-bridged dimer **61c** and aromatized product **65b** (**61c**:**65b** ca. 1:3 by ^{1}H NMR spectroscopy). This finding implies that compounds **60c**, **66**, and **67** exist in equilibrium with each other and that the rate of aromatization of **66** to **65b** is considerably faster than the second Michael reaction leading from **67** to **61c**. These observations were consistent with a stepwise mechanism for the cytoskyrin cascade involving two sequential Michael reactions as opposed to a concerted mechanism involving a hetero Diels−Alder process, at least in the case of the TBS-protected substrate **59c**.

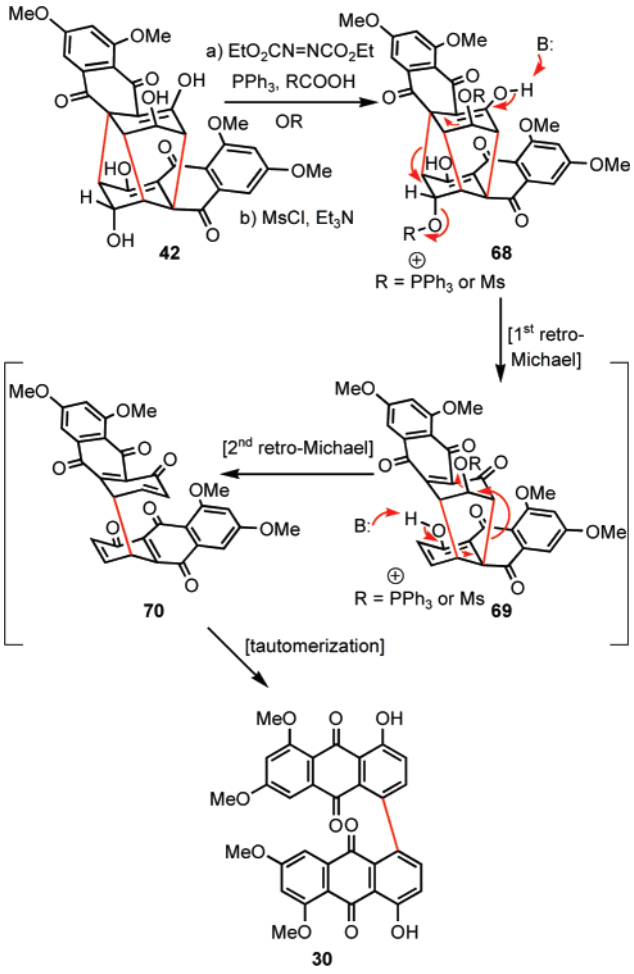
Scheme 12. Mechanistic Investigation of the Dimerization Cascade (**59c** → **61c**)[a]



[a] Reagents and conditions: (a) MnO_2 (1.5 wt. equiv), CH_2Cl_2 (0.35 M), 25 °C, 1 h, 20% (**61c**), 30% (**67**), 45% (**65b**), 5% (**60c**); ratio of products dependent on reaction time.

Attempts To Synthesize Cytoskyrin A. Realizing the difficulties associated with the direct conversion of stereochemically appropriate anthraquinones to cytoskyrin-like structures discussed above and summarized in Figure 4, we opted to make use of the facile entry into the epimeric series of compounds with the expectation that their hydroxy groups at the C2 and C2′ positions could be inverted at the end of the synthesis. To this end, the required anthradihydroquinone substrates **59d** and **59e** (Scheme 10) were synthesized and subjected to the cytoskyrin cascade under the standard conditions shown in Scheme 11, affording *epi*-cytoskyrin-like bisanthraquinones **64d** and **64e** in 34 and 40% overall yields (unoptimized), respectively. While the SEM-protected compound **64d** resisted selective deprotection to the desired diol system needed for the intended inversion (under various fluoride or acidic conditions), the bis-MOM derivative **64e** yielded smoothly the desired product **42**, and in 91% yield, upon exposure to concentrated HCl at 60 °C (12 h). Unfortunately, attempts to invert the two cyclohexane-bound hydroxy groups within compound **42** by either Mitsunobu reaction (PPh_3, EtOOCN=NCOOEt, 4-nitrobenzoic acid) or mesylation (MsCl, Et_3N), followed by nucleophilic displacement, led to exclusive formation of the double retro-Michael reaction product **30** (85% for the Mitsunobu attempt; 91% for the mesylation attempt), as shown in Scheme 13.

Scheme 13. Attempts To Synthesize Cytoskyrin A (**2a**)[a]

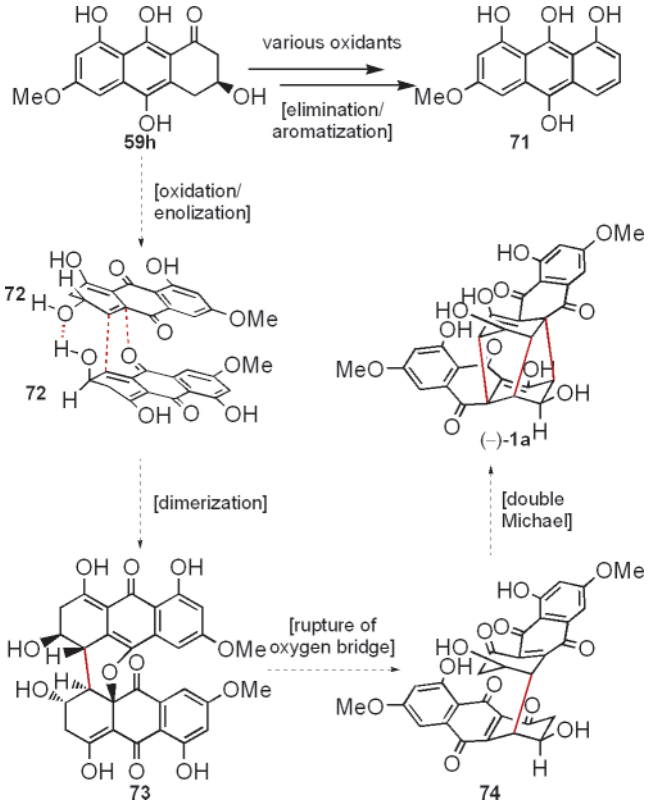


[a] Reagents and conditions: (a) **42** (1.0 equiv), *p*-nitrobenzoic acid (4.1 equiv), PPh_3 (3.8 equiv); then EtOOCN═NCOOEt (4.5 equiv), THF, 0 °C, 10 min, 85%; (b) **42** (1.0 equiv), MsCl (3.0 equiv), 2,6-lutidine (3.0 equiv), 0 °C, 10 min, 91%. Abbreviation: Ms, methanesulfonyl.

Having failed to achieve the required inversion of the two hydroxyl groups of the *epi*-cytoskyrin-type compound **42**, we then attempted to use the monomeric anthradihydroquinone **59h** with a free hydroxy group as a precursor, reasoning that the relatively small size of this group will allow at least partial entry into the desired stereochemical course of the initial dimerization step of the cascade. The suspicion that cytoskyrin and its close relatives, graciliformin and rugulosin, may very well be formed in nature through such hydroxyl-free intermediates fueled our drive to test this daring approach. The required substrate, compound **59h**, was synthesized by a Hauser annulation involving the MOM-protected nitrile **46e** and the TMS-protected enone **51f** (41% yield), followed by global deprotection through brief exposure (<1 min, 75% yield) to HF·py, as shown in Scheme 10. Exposure of this rather labile monomeric anthradihydroquinone to the standard MnO_2 cytoskyrin cascade conditions (Scheme 14), however, disappointingly resulted in rapid decomposition and aromatization to **71** (ca. 45%, yield dependent on reaction time). No detectable dimerization products were observed. Other oxidants (e.g., $PhI(OTFA)_2$, Ag_2O, CAN) employed led to similar results.

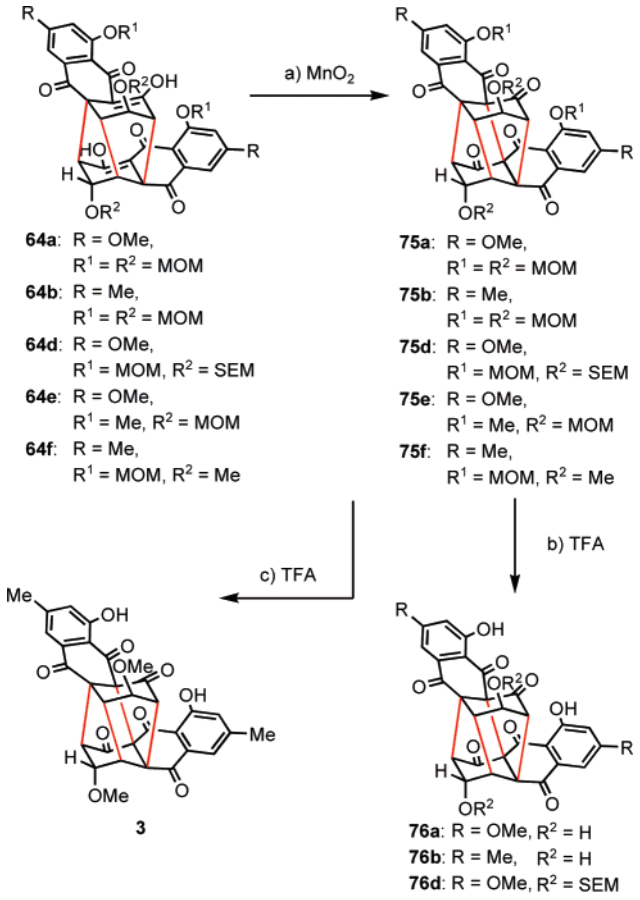
Total Synthesis of the Alleged Structure of Rugulin and Analogues Thereof. The naturally occurring substance rugulin (**3**) is reported to include in its structure the novel "skyrane"

Scheme 14. Attempt To Synthesize Cytoskyrin A (**2a**) Using the Monomeric Anthradihydroquinone **59h** with Free Hydroxy Group



structural motif (Figure 2) and, as such, provided an unusual and attractive target for total synthesis. It so happened that rugulin's stereochemistry at the C2 and C2′ positions is in line with the expected outcome of the most favored dimerization mode of the corresponding monoanthraquinone precursors, as outlined in Figure 4. Indeed, the facile formation of the model system **28** from **27** (Scheme 5) and the previous work of Shibata[4b,c] were encouraging in this regard. Initial concerns about the methoxy group eliminating at the early stages of the sequence quickly dissipated following our successful construction of the required enone **51d**, as shown in Scheme 8. Thus, the requisite monomeric anthradihydroquinone **59f** was synthesized, as shown in Scheme 10, by the Hauser annulation involving nitrile **46f** (Scheme 7) and enone **51d** (Scheme 8). As expected, when **59f** was subjected to the standard conditions for the cytoskyrin cascade [MnO_2 (1.0 wt. equiv), CH_2Cl_2, 25 °C, 1 h; then Et_3N (5.0 equiv), CH_2Cl_2, 25 to 45 °C, 12 h], the cytoskyrin-like cage compound **64f** was formed in 40% overall yield (unoptimized), as shown in Scheme 11. Further oxidation of the latter compound with MnO_2 (10.0 wt equiv) in CH_2Cl_2 at 25 °C for 18 h furnished the desired protected rugulin-type structure **75f** in 70% yield, as depicted in Scheme 15. Finally, global deprotection of **75f** under acidic conditions (TFA:CH_2Cl_2, 1:1, 0 °C, 10−15 min) resulted in the formation of the targeted rugulin structure **3** in quantitative yield. Much to our dismay, however, the ^1H NMR spectral data of the synthetic material did not match those reported in the literature[5] for the natural rugulin. That our synthetic material crystallized (acetone, slow crystallization) and yielded to X-ray crystallographic analysis[15] (see ORTEP drawing, Figure 6) provided the unambiguous proof that the originally reported[5] structure for rugulin was erroneous. In fact, there was previous skepticism about the

Scheme 15. Synthesis of the Alleged Rugulin Structure and Analogues Thereof[a]



[a] Reagents and conditions: (a) MnO_2 (10.0 wt equiv), CH_2Cl_2, 25 °C, 18 h, 100% (**75a**), 82% (**75b**), 73% (**75d**), 98% (**75e**), 70% (**75f**); (b) TFA: CH_2Cl_2 (1:1), 0 °C, 1.5 h, 97% (**76a**), 98% (**76b**), 89% (**76d**); (c) TFA: CH_2Cl_2 (1:1), 0 °C, 10 − 15 min, 100%.

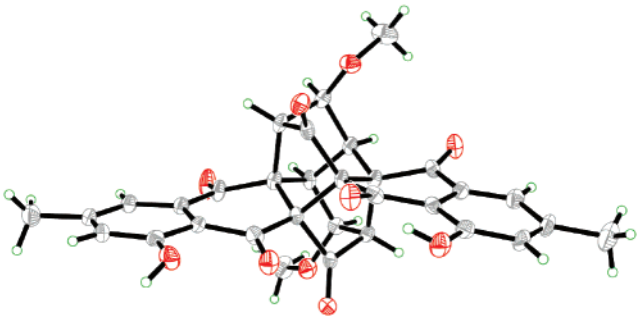


Figure 6. ORTEP drawing of the alleged rugulin (**3**) derived from X-ray crystallographic analysis.

originally assigned structure for rugulin based on its IR spectrum, as noted by Snider et al.[24] Attempts to contact the discoverers of rugulin for a sample proved in vain, leading us to abandon our quest for the true structure of rugulin, a puzzle that will have to await further experimentation for its solution. Despite the rugulin mystery, however, we were able to construct several siblings of its alleged structure through the developed technology. Thus, the rugulosin-type compounds **64a**, **64b**, **64d**, and **64e** (Scheme 15) were converted to the protected rugulin-type structures **75a** (100%), **75b** (82%), **75d** (73%), and **75e** (98% yield), respectively. From these compounds, **75a**, **75b**, and **75d** were globally deprotected by the action of TFA in CH_2Cl_2 (1:1, 0 °C, 1.5 h) to afford **76a** (97%), **76b** (98%), and **76d** (89% yield), respectively.

Conclusion

The described cascade reactions provide a stunning demonstration of the power of such synthetic sequences in constructing high molecular complexity from relatively simple building blocks. Originally introduced by Sir Robert Robinson in 1917[28] with his impressive synthesis of tropinone, and subsequently highlighted most impressively by W. S. Johnson in his biomimetic steroid synthesis,[29] and by us in our endiandric acid cascades,[30] this concept has by now been adopted in myriad cases of total synthesis.[31] The present sequences, termed "the cytoskyrin cascade", provided rapid entries into the growing class of the bisanthraquinone natural products exemplified by 2,2′-*epi*-cytoskyrin A (**2a**) and rugulosin (**2b**) and their precursors. Due to the rather strict stereochemical requirements of the initial dimerization step, however, this cascade failed to deliver cytoskyrin A (**1a**), and, although successful in delivering the reported structure of rugulin (**3**), it only proved that the latter was erroneous. The true structure of rugulin, therefore, remains a mystery. These shortcomings notwithstanding, the reported chemistry provides an example of aesthetically pleasing cascades orchestrated by the simplest of reagents (i.e., MnO_2, Et_3N, CSA). Furthermore, besides being efficient, these high-complexity-yielding processes are both economical and environmentally friendly, for they require minimal amounts of reagents and solvents.

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Supporting Information Available: Complete ref 11, experimental procedures, and full compound characterization (PDF, CIF). This material is available free of charge via the Internet at http://pubs.acs.org.

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(28) Robinson, R. *J. Chem. Soc.* **1917**, 762−768.
(29) (a) Johnson, W. S.; Gravestock, M. B.; Parry, R. J.; Myers, R. F.; Bryson, T. A.; Miles, D. H. *J. Am. Chem. Soc.* **1971**, *93*, 4330−4332. (b) Johnson, W. S.; Gravestock, M. B.; McCarry, B. E. *J. Am. Chem. Soc.* **1971**, *93*, 4332−4334. (c) Gravestock, M. B.; Johnson, W. S.; McCarry, B. E.; Parry, R. J.; Ratcliffe, B. E. *J. Am. Chem. Soc.* **1978**, *100*, 4274−4282.
(30) (a) Nicolaou, K. C.; Petasis, N. A.; Zipkin, R. E.; Uenishi, J. *J. Am. Chem. Soc.* **1982**, *104*, 5555−5557. (b) Nicolaou, K. C.; Petasis, N. A.; Uenishi, J.; Zipkin, R. E. *J. Am. Chem. Soc.* **1982**, *104*, 5557−5558. (c) Nicolaou, K. C.; Zipkin, R. E.; Petasis, N. A. *J. Am. Chem. Soc.* **1982**, *104*, 5558−5560. (d) Nicolaou, K. C.; Petasis, N. A.; Zipkin, R. E. *J. Am. Chem. Soc.* **1982**, *104*, 5560−5562.
(31) Nicolaou, K. C.; Edmonds, D. J.; Bulger, P. G. *Angew. Chem., Int. Ed.* **2006**, *45*, 7134−7186.
(32) We thank Prof. O. Ermer for suggesting that we revert to this space group (*Cc*) and for critically reviewing our conclusions regarding this anomalous X-ray crystallographic analysis.