DOI: 10.1002/anie.200806269

Natural Product Synthesis

Total Synthesis of Cribrostatin 6**

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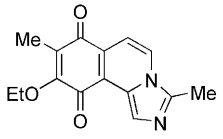
The spread of multi-drug-resistant Gram-positive bacteria has spurred the search for novel structural classes of antibiotics. Resistant strains of enterococci and *Staphylococcus aureus, Pseudomonas aeruginosa*, and *Streptococcus pneumoniae* are emerging with troubling frequency. Indeed, *Streptococcus pneumoniae*, which is the most common bacterial cause of acute respiratory infection and otitis media, causes millions of deaths each year.[1] It is thus significant that in 2003, Pettit et al. reported the isolation and characterization of cribrostatin 6 (**1**) from the blue marine sponge *Cribrochalina* sp. (Figure 1).[2] Although cribrostatin 6 was found to inhibit the growth of a number of antibiotic-resistant Gram-positive bacteria and pathogenic fungi, it was most active against *S. pneumoniae*.[1] In addition to its potent antimicrobial activity, **1** displayed antineoplastic activity against murine and human cancer cell lines at micromolar concentrations (P388 ED_{50} = 0.3 $\mu g\,mL^{-1}$).[2]

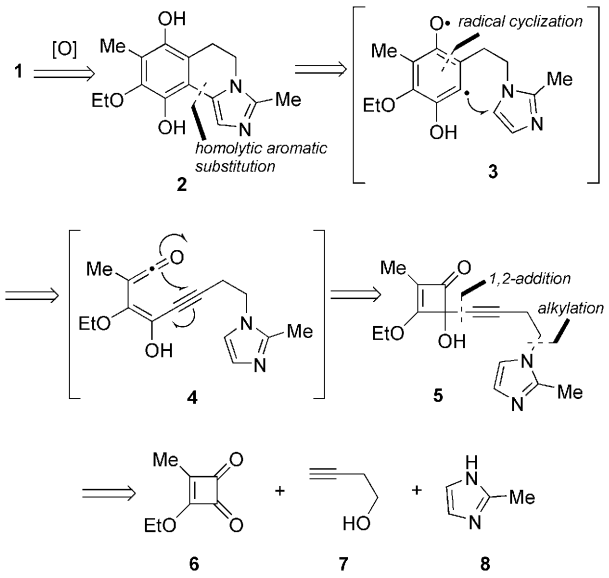


cribrostatin 6 (**1**)

Figure 1.

Owing to its important biological activity and its tricyclic imidazo[5,1-*a*]isoquinolinedione architecture, which is unique amongst known natural products, cribrostatin 6 (**1**) has inspired interest in its synthesis. Indeed the research groups of Nakahara[3] and Kelly[4] have each completed the total synthesis of **1**, although the syntheses were somewhat lengthy requiring 18 and 13 steps (15 steps in total), respectively. In the context of several ongoing projects, it occurred to us that we might be able to develop a more concise, and hopefully more efficient route to **1**, that might also be amenable for the facile preparation of analogues for further biological evaluation. We report herein the realization of this goal and the completion of a concise synthesis of cribrostatin 6 (**1**).

Our approach to cribrostatin 6 (**1**) is outlined in retrosynthetic format in Scheme 1 and features a novel application of chemistry pioneered some years ago by Moore and co-workers.[5] In particular, we envisioned that it might be possible to effect the one-pot conversion of the squarate derivative **5** into **1** through a sequence involving ring opening of **5** to give the ketene **4** that would cyclize to generate the putative diradical **3**. Spontaneous cyclization of **3** by an intramolecular homolytic aromatic substitution reaction would then produce the penultimate intermediate **2**,[6] oxidation of which would generate cribrostatin 6 (**1**). The conversion of squarate derivatives into benzenoid compounds by ring opening/cyclization is well known.[5,7] There is also some precedent for the cyclizations of benzenoid intermediates generated from enynyl ketenes having pendant alkenyl and alkynyl groups.[8] However, we are aware of only two closely related reports of cyclizations of benzenoid diradicals, derived from enynyl ketenes, onto appending aryl groups.[9] Hence, the successful reduction of our strategy to practice would significantly expand the utility of such processes. The assembly of the key intermediate **5** would only require the use of known 3-ethoxy-4-methylcyclobutene-1,2-dione (**6**)[10] and the commercially available 3-butyn-1-ol (**7**) and 2-methylimidazole (**8**).



Scheme 1. Retrosynthetic analysis of cribrostatin 6 (**1**).

Turning our attention to the forward synthesis, alcohol **7** was transformed into known tosylate **9**[11] in 95% yield by

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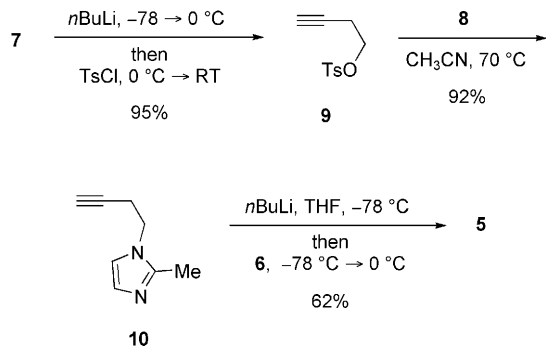
[**] Acknowledgement is made to The National Institutes of General Medical Sciences (GM31077) and The Robert A. Welch Foundation for support of this research. D.K. would like to thank the National Science Foundation for a Graduate Research Fellowship. We also thank Genevieve Pease for the preparation of starting materials.

Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/anie.200806269.

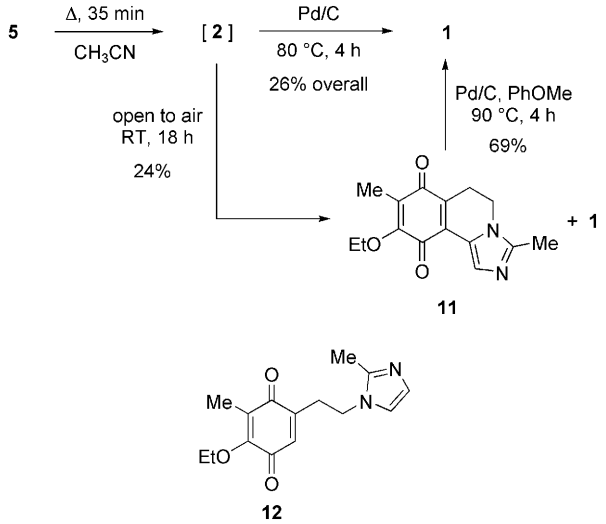
slight modification of the literature procedure (Scheme 2). Initial attempts to generate alkyne **10** from 2-methylimidazole (**8**) and **9** in the presence of additional bases such as



Scheme 2. Synthesis of key intermediate **5**. THF = tetrahydrofuran, Ts = 4-toluenesulfonyl.

*t*BuOK, Cs_2CO_3, and NaH did not afford significant amounts of the desired **10**.[12] However, we discovered that the reaction of **9** with excess imidazole provided the alkyne **10** in 92% yield. Deprotonation of **10** with *n*BuLi and subsequent addition of the acetylide anion thus formed to the squarate **6**[10] delivered the desired key intermediate **5**.

With the key intermediate **5** in hand, the stage was set to investigate its conversion into cribrostatin 6. In the event, a dilute solution of **5** was heated in CH_3CN (0.001M) to give a mixture containing a compound whose ^1H NMR spectroscopic characteristics corresponded to those expected for the hydroquinone **2** (Scheme 3). However, it was not possible to isolate pure **2** from this mixture owing to its facile oxidation to quinone **11**. When **5** in CH_3CN (0.001M) was heated at reflux for 35 minutes and the resulting solution stirred overnight at room temperature open to the air, a mixture (2:1) of **11** and **1** was obtained in 24% yield. Heating this mixture in the presence of Pd/C led to complete dehydrogenation of **11** and formation of **1** in 69% yield. The synthetic **1** thus obtained

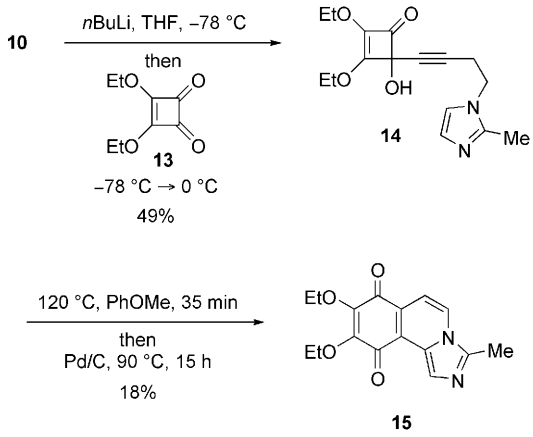


Scheme 3. Key cyclization reaction and synthesis of cribrostatin 6 (**1**).

gave ^1H and ^{13}C NMR data consistent with those reported for the natural product.[2–4] A simple expedient of this experimental protocol was then developed to generate **1** in a single operation from **5**. Namely, **5** was first heated in CH_3CN (0.001M); the majority of the solvent was evaporated, Pd/C was added, and the mixture was heated at 80 °C to furnish **1** in 26% overall yield.

All attempts to improve the yield of this sequence by varying solvent, concentration, and temperature were unavailing. For example, when a more concentrated solution of **5** (0.01M) was heated in CH_3CN and then stirred overnight in a flask open to the air, a mixture (1:1) of quinones **11** and **12** was obtained. Upon heating a solution of **5** in PhCl (0.02M), quinone **12** was isolated as the sole product in about 25% yield. This result seems to suggest that the intermediate diradical **3** can undergo not only cyclization to the desired tricycle, but can also suffer intermolecular hydrogen-atom transfer to deliver a hydroquinone that is readily oxidized to **12**.[8b,c]

To probe the generality of this new approach to imidazo-[5,1-*a*]isoquinolinediones, **15**, an analogue of cribrostatin 6 was prepared (Scheme 4). Toward this goal, the substituted



Scheme 4. Synthesis of the cribrostatin 6 analogue **15**.

squarate **14** was synthesized by coupling the anion of the acetylenic imidazole **10** with commercially available diethyl squarate (**13**). Although **14** was stable to heating at reflux in CH_3CN, heating it at 120 °C in anisole (0.001M) and subsequent oxidation as before gave **15** in 18% overall yield from **14**.

In summary, we have accomplished a concise and novel total synthesis of cribrostatin 6 (**1**) in 14.1% overall yield by a sequence that employed no protecting groups and required only four steps in the longest linear sequence and five total steps from commercially available starting materials. In the key step, squarate **5** underwent a tandem 4π electrocyclic ring opening, radical cyclization, and homolytic aromatic substitution sequence to afford the tricyclic core of the target molecule, which was directly oxidized to the natural product in one pot. This approach to tricyclic products from readily available squarate derivatives thus represents a significant extension of chemistry originally developed by Moore and co-

workers. Moreover, the synthesis of these compounds is highly modular so that modification of the starting materials used as inputs will lead to related compounds for biological testing. In this context, the versatility of the approach was demonstrated by preparing analogue **15**. Extension of this chemistry to the facile preparation of additional analogues of **1** is currently underway and will be reported in due course.

Received: December 22, 2008
Published online: February 23, 2009

Keywords: cyclobutenediones · natural products · quinones · rearrangement · total synthesis

[1] R. K. Pettit, R. F. Bridget, J. C. Knight, C. A. Weber, G. R. Pettit, G. D. Cage, S. Pon, *J. Med. Microbiol.* **2004**, *53*, 61 – 65, and references therein.
[2] G. R. Pettit, J. C. Collins, J. C. Knight, D. L. Herald, R. A. Nieman, M. D. Williams, R. K. Pettit, *J. Nat. Prod.* **2003**, *66*, 544 – 547.
[3] a) S. Nakahara, A. Kubo, *Heterocycles* **2004**, *63*, 2355 – 2362; b) S. Nakahara, A. Kubo, Y. Mikami, J. Ito, *Heterocycles* **2006**, *68*, 515 – 520.
[4] M. D. Markey, T. R. Kelly, *J. Org. Chem.* **2008**, *73*, 7441 – 7443.
[5] For reviews of ring expansions of cyclobutenones, see: a) H. W. Moore, B. R. Yerxa, *Chemtracts* **1992**, *5*, 273 – 313; b) H. W. Moore, B. R. Yerxa, *Adv. Strain Org. Chem.* **1995**, *4*, 81 – 162.
[6] For reviews of homolytic aromatic substitutions, see: a) M. Tiecco in *Free Radical Reactions* (Eds: D. H. Hey, W. A. Walters), Butterworths, London, **1975**, pp. 25 – 45; b) H. Togo in *Advanced Free Radical Reactions for Organic Synthesis*, Elsevier, Oxford, **2004**, pp. 75 – 83.
[7] L. S. Liebeskind, *Tetrahedron* **1989**, *45*, 3053 – 3060.
[8] a) S. Xu, H. W. Moore, *J. Org. Chem.* **1991**, *56*, 6104 – 6109; b) H. Xia, H. W. Moore, *J. Org. Chem.* **1992**, *57*, 3765 – 3766; c) Y. Xiong, H. Xia, H. W. Moore, *J. Org. Chem.* **1995**, *60*, 6460 – 6467; d) P. Wipf, C. R. Hopkins, *J. Org. Chem.* **1999**, *64*, 6881 – 6887.
[9] a) Y. Xiong, H. W. Moore, *J. Org. Chem.* **1996**, *61*, 9168 – 9177; b) A. R. Hergueta, H. W. Moore, *J. Org. Chem.* **1999**, *64*, 5979 – 5983.
[10] M. W. Reed, D. J. Pollart, S. T. Perri, L. D. Foland, H. W. Moore, *J. Org. Chem.* **1988**, *53*, 2477 – 2482.
[11] a) E. Negishi, L. D. Boardman, H. Sawada, V. Bagheri, A. T. Stoll, J. M. Tour, C. L. Rand, *J. Am. Chem. Soc.* **1988**, *110*, 5383 – 5396; b) J. R. Falck, A. He, H. Fukui, H. Tsutsui, A. Radha, *Angew. Chem.* **2007**, *119*, 4611 – 4613; *Angew. Chem. Int. Ed.* **2007**, *46*, 4527 – 4529.
[12] a) M. Scobie, M. D. Threadgill, *J. Chem. Soc. Chem. Commun.* **1992**, 939 – 941; b) H. He, D. Zatorska, J. Kim, J. Aguirre, L. Llauger, Y. She, N. Wu, R. M. Immormino, D. T. Gewirth, G. Chiosis, *J. Med. Chem.* **2006**, *49*, 381 – 390; c) S. Luo, X. Mi, L. Zhang, S. Liu, H. Xu, J.-P. Cheng, *Angew. Chem.* **2006**, *118*, 3165 – 3169; *Angew. Chem. Int. Ed.* **2006**, *45*, 3093 – 3097.

