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Biomimetic Synthesis of the Antimalarial Flindersial Alkaloids

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Supporting Information

ABSTRACT: A biomimetic strategy for the synthesis of the antimalarial flindersial alkaloids is described. Flinderoles A, B, and C, desmethylflinderole C, isoborreverine, and dimethylisoborreverine were all synthesized in three steps from tryptamine. The key step is an acid-promoted dimerization of the natural product borrerine. This approach is thought to mirror the biosynthesis of these compounds.

Historically, successful malaria treatments have been derived from natural products.[1] Emerging malarial drug resistance compels additional investigations in this area, and natural product research remains a promising ground for finding new treatments.[2] In a recent disclosure of the isolation and structural determination of flinderole B (**3**) and C (**5**) (Figure 1), Avery and co-workers reported significant

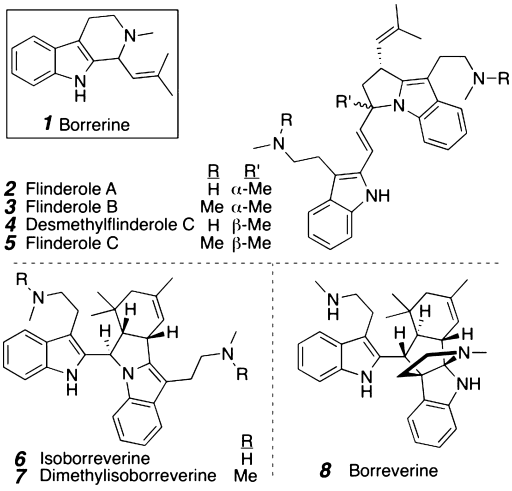
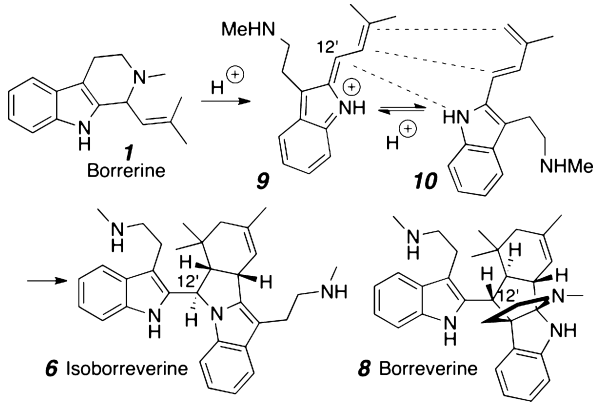


Figure 1. The flindersial alkaloids.

antimalarial activity for the alkaloids **2**, **3**, and **5**–**7** of the plant genus *Flindersia*.[3] A follow-up paper reported more detailed biological activities and suggested that these alkaloids interrupt parasitic hemoglobin metabolism through a different mechanism than that of chloroquine.[4] Consequently, these alkaloids show parasitic toxicity even in chloroquine-resistant strains of *Plasmodium falciparum*. To gain access to these compounds and generate structural analogues in significant amounts for further testing, we have developed a biomimetic synthesis[5] of alkaloids **2**–**7** in three synthetic steps from tryptamine. This route is significantly shorter than reported strategies for synthesizing the flinderoles and allows access to all of the bioactive members of the family. Additionally, these studies shed light on the probable biosynthetic pathway for these alkaloids.

The report of the structure of the flinderoles and their bioactivity quickly drew synthetic interest, culminating in two strategies consisting of 14[6] and 19[7] synthetic steps. Concurrent with the initiation of those efforts, we became interested in synthesizing the flinderoles and their constitutional isomers, the borreverines.[8] Studies of the likely biosyntheses of borreverine and isoborreverine were conducted in the 1970s.[9] These alkaloids have an obvious biosynthetic origin in naturally occurring borrerine (**1**). The proposed mechanism of their formation begins with acidic opening of borrerine to an equilibrium mixture of iminium **9** and indolodiene **10**, which is followed by Diels–Alder cycloaddition (Scheme 1).[10] Sub-

Scheme 1. Biosynthesis of Borreverines



sequent N1 or C3 attack by the indole of **10** at C12′ in **9** would produce isoborreverine or borreverine, respectively. Experimentally, treatment of borrerine with trifluoroacetic acid (TFA) at 65 °C was reported by Koch to produce a 50:50 mixture of isoborreverine and borreverine in 80% yield.[8] Prolonged exposure to acid was said to produce a greater percentage of the former product, indicating that it is favored thermodynamically. Together with the observation that these compounds naturally occur as racemates, these studies strongly supported a biosynthesis of the borreverines from borrerine.

We hypothesized that the flinderole skeleton could also arise from a dimerization of borrerine through reaction of indolodiene **10** with imine **9** or **11**. However, this would proceed through a formal [3+2] cycloaddition pathway in lieu

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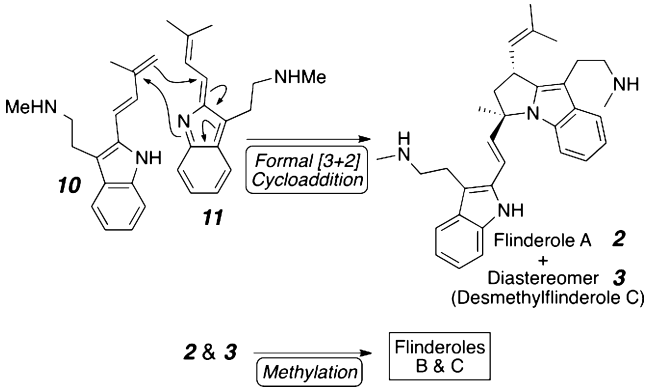
of the initial [4+2] cycloaddition assumed for the borreverines (Scheme 2). Further support for this possibility was found in

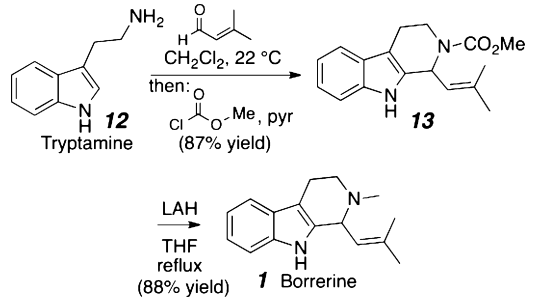
Scheme 2. Proposed Flinderole Biosynthesis



Dethe's recent report of a biosynthesis-inspired [3+2] cycloaddition between two strategically protected alkene-appended indoles.[6] We began to wonder whether the flinderoles had actually been produced in the biosynthetic study of the borreverines[9] but were overlooked as the flinderoles' structures were not known at the time.

To test whether flinderole formation occurs alongside borreverine production, borrerine was first synthesized (Scheme 3). This was readily accomplished using Sakai's

Scheme 3. Synthesis of Borrerine (1)



approach.[11] In fact, by optimizing this sequence, we obtained **1** in 77% overall yield from tryptamine in two steps.[12]

The synthetic borrerine was treated with 2 equiv of TFA in benzene at 65 °C for 40 min (Table 1, entry 2) and was consumed, as observed by thin-layer chromatography. Two new closely spaced product spots appeared, and ^{1}H NMR analysis of the crude mixture showed evidence of isoborreverine as one of the major products. The remaining ^{1}H NMR peaks corresponded to the spectra of flinderole A and desmethylflinderole C. *In fact, the flinderole diastereomers appeared to be produced in roughly the same amount as isoborreverine!* Contrary to the previous report, only trace amounts of borreverine (**8**) were observed. While isoborreverine was readily isolated via chromatography, the separation of the flinderoles required the use of reversed-phase HPLC. This purification allowed us to confirm the alkaloid identities and the ratios of their formation as observed in the ^{1}H NMR spectra of the crude mixtures.

In an effort to reproduce the original report of the formation of borreverine,[13] a range of TFA equivalents, reaction times, and reaction temperatures were screened (entries 1–7). ^{1}H NMR peaks associated with **8** were occasionally observed, but

Table 1. Treatment of 1 with TFA

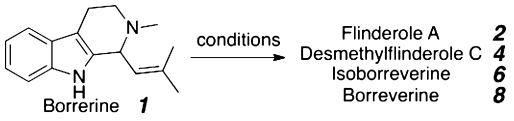
entry	TFA equiv	solvent	T (°C)	time	**2**:**4**:**6**:**8**[a]
1	1.0	benzene	65	2 h	0:0:0:0[b]
2	2.0	benzene	65	40 min	41:30:28:1
3	2.0	benzene	55	80 min	38:33:29:0
4	3.0	benzene	40	2 h	34:32:34:0
5	3.0	benzene	65	30 min	28:28:43:1
6	6.0	benzene	0–22	24 h	16:13:34:0[b]
7	6.0	benzene	40	30 min	17:17:21:0[b]
8	2.0	$CHCl_3$	45	30 min	28:28:7:1[b]
9	2.0	none	65	48 h	0:0:100:0

[a]Determined by integration of ^{1}H NMR peaks. The combined yields were all above 95%. [b]The balance of the material was **1**.

integration indicated that it was produced in only trace amounts. In retrospect, this is unsurprising, as the stereochemistry of borreverine would have to arise from a less-favorable exo-Diels–Alder reaction in the initial step (Scheme 1). This observation may have relevance to the alkaloids' biosynthesis, as Avery and co-workers did not observe any borreverine in the *Flindersia* species that produced the flinderoles.[3] Changing the solvent to chloroform gave the flinderoles as a majority of the crude mixture (entry 8). The use of tetrahydrofuran (THF), dioxane, or *N*,*N*-dimethylformamide (DMF) as the solvent resulted in no reaction. Interestingly, the omission of solvent resulted in exclusive formation of isoborreverine.

A variety of Brønsted and Lewis acids were also introduced to the reaction (Table 2). Many of these acids either failed to promote the reaction or caused decomposition of the material to mixtures of unidentifiable products (entries 1–5). When dimerization cleanly occurred, isoborreverine was almost exclusively produced (entries 6–9). The use of $BF_3{\cdot}OEt_2$ for an extended time led to the formation of only isoborreverine

Table 2. Effects of Acids on Dimerization



entry	acid[a]	solvent	T (°C)	time	**2**:**4**:**6**:**8**[b]
1	TfOH	benzene	0–22	40 min	0:0:0:0[c]
2	TfOH	benzene	60	40 min	decomp.
3	pTsOH	benzene	0–22	60 min	0:0:0:0[c]
4	TMSOTf	benzene	60	40 min	0:0:0:0[c]
5	$AlCl_3$	benzene	0–22	120 min	0:0:0:0[c]
6	$AlCl_3$	benzene	60	40 min	0:0:40:0[c]
7	$Sc(OTf)_3$	benzene	60	40 min	0:0:38:0[c]
8	Tf_2O	benzene	0	60 min	0:0:50:0[c]
9	$BF_3{\cdot}OEt_2$	CH_2Cl_2	22	3 days	0:0:100:0
10	1 N HCl[d]	methanol	55	2 h	43:26:30:1
11	CH_3COOH[e]	none	55	18 h	38:33:0:0[c]

[a]Unless otherwise noted, 2 equiv of acid was used. [b]Determined by integration of ^{1}H NMR peaks. The combined yields were all above 95%. [c]The balance of the material was **1**. [d]6 equiv of acid. [e]150 equiv of acid.

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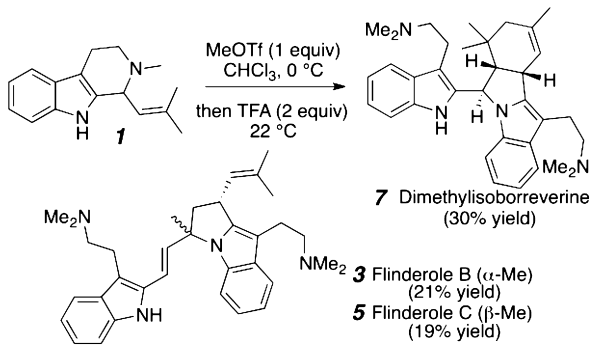
with complete conversion (entry 9). Methanolic HCl, however, provided the flinderoles as the major product (entry 10). Impressively, neat acetic acid produced only the flinderoles (entry 11)! This selectivity is particularly notable given that neat TFA favors isoborreverine formation.

Thus, conditions for selective formation of either the flinderoles or isoborreverine were found. Apparently, only a few acids promote just enough reactivity to form the flinderoles. Other acids are either too weak to initiate the reaction or are so reactive that the flinderoles are bypassed for the more thermodynamically stable isoborreverine.[7b,8] In fact, prolonged reaction times generally produce greater percentages of isoborreverine. Furthermore, isolated samples of the flinderoles are converted to isoborreverine in strong acid, illustrating the preference for that structure under thermodynamic conditions.

As flinderole B (**3**), flinderole C (**5**), and dimethylisoborreverine (**7**) exhibit the strongest antimalarial activity,[4] these were next targeted for synthesis. While reductive amination to add an additional methyl group to the secondary amine of the isolated products **2**, **4**, and **6** is certainly feasible, a more direct approach would be to add the methyl concomitantly with borrerine dimerization. Thus, we postulated that borrerine could be exposed to a methylating agent to generate an ammonium salt. Upon treatment with acid and an increase in the temperature, ring opening would form intermediates similar to **9**–**11** but having a pendant tertiary amine. Dimerization could then directly provide **7** (Scheme 4) as well as **3** and **5**.

Scheme 4. Direct Methylative Dimerization



Indeed, when borrerine was first treated with methyl triflate in chloroform at 0 °C and then treated with 2 equiv of TFA and warmed to 22 °C, the dimethylamine alkaloids were formed directly in a combined 70% yield. Dimethylisoborreverine constituted 30% of the mixture, and flinderoles B and C were present as 21 and 19% of the mixture, respectively.[12,14] A longer exposure to TFA afforded dimethylisoborreverine as the only product. These results may have implications for the biosynthesis of flinderoles B and C, which were isolated from the same plant species as dimethylisoborreverine.[3b] In that species, bisindole alkaloid synthesis may be initiated by methylation.

To demonstrate that this biomimetic synthesis can practically provide access to these compounds, the most selective conditions were repeated on a larger scale to generate isolable quantities of each alkaloid. For example, when 50 mg of borrerine was treated with $BF_3{\cdot}OEt_2$ in benzene for 3 days, 20 mg of pure isoborreverine was isolated. Similarly, 28 mg of flinderole A and desmethylflinderole C was obtained from the reaction of 50 mg borrerine with TFA in benzene at 65 °C. A 60 mg yield of dimethylisoborreverine was obtained as the sole product from the reaction of 100 mg of borrerine with 1 equiv of MeOTf and then 2 equiv of TFA for 35 min at room temperature.[12] When this latter reaction was run for less time, 20 mg of dimethylisoborreverine, 13 mg of flinderole C, and 13 mg of flinderole B were obtained.

In conclusion, a biomimetic synthesis of all of the antimalarial flindersial alkaloids has been established. Variations in the reaction conditions allow for the acid-promoted dimerization of borrerine to produce isoborreverine or the flinderoles selectively. We have demonstrated that the outcome of this reaction is different from that previously reported. A novel strategy utilizing an initial methylation followed by acid treatment allows for the synthesis of the flindersial alkaloids appended with tertiary amines. Remarkably, the sequence allows selective access to these alkaloids in only three synthetic steps. Furthermore, the use of tryptamine analogues, alternate α,β-unsaturated aldehydes, and a variety of acylating agents will enable the synthesis of analogues of isoborreverine and the flinderoles.

■ ASSOCIATED CONTENT

Supporting Information

Additional optimization data, experimental procedures, and new characterization data for all compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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(12) See the Supporting Information for full experimental details, compound characterization, and representative NMR spectra.
(13) Equivalents of acid and reaction concentration were not specified in the original procedure.
(14) The balance of the material was made up of a fourth dimeric component that appears to be the result of initial indole methylation; complete structural elucidation of this compound is ongoing.