

Stereocontrolled Synthesis of (\pm)-Melokhanine E via an Intramolecular Formal [3 + 2] Cycloaddition

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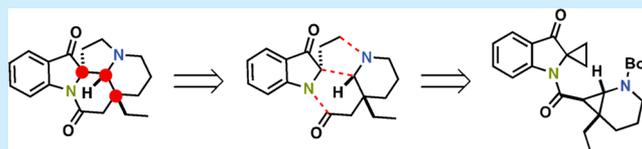


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ABSTRACT: A convergent sequence to access the indole alkaloid (\pm)-melokhanine E in 12-steps (8-step longest linear sequence) and an 11% overall yield is reported. The approach utilizes two cyclopropane moieties as reactive precursors to a 1,3-dipole and imine species to enable stereoselective construction of the core scaffold through a formal [3 + 2] cycloaddition. The natural product was evaluated for its antimicrobial activity based on isolation reports; however, no activity was observed. The reported efforts serve as a synthetic platform to prepare an array of alkaloids bearing this core structural motif.



New natural product skeletons with antimicrobial activity are of significant interest as there is a pressing need for the development of novel antimicrobial agents.^{1–3} Melokhanine E (1, Figure 1) and a series of related compounds were

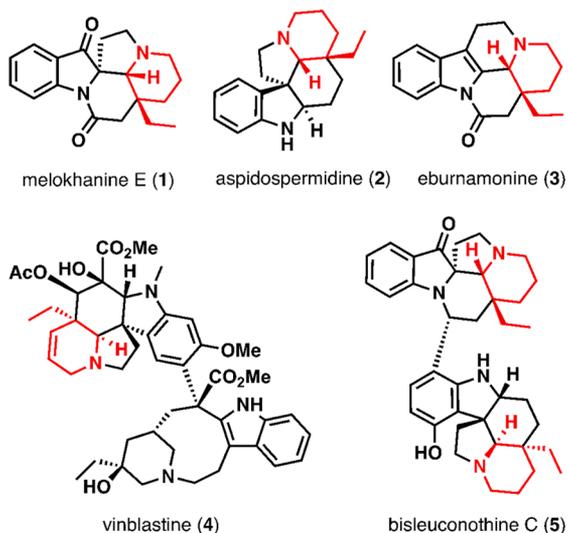


Figure 1. Representative biologically active natural products containing a piperidine C3 all-carbon quaternary center.

isolated in 2016 from *Melodinus khasianus*, a subtropical plant used in traditional Chinese medicine as a treatment for rheumatic heart disease and meningitis.⁴ These indole alkaloids possess a 6/5/5/6/6 pentacyclic structure and are reported to exhibit potent antimicrobial activity against Gram-negative pathogens *P. aeruginosa* and *E. coli* as well as Gram-positive pathogens such as *E. faecalis*. Within the family, melokhanine E is of particular interest due to its superior broad-spectrum activity (MIC = 2 μ M against *P. aeruginosa* and 5 μ M against *E.*

faecalis).⁴ A key structural motif of the melokhanines is a core piperidine heterocycle bearing C3 quaternary substitution. This structural element is found in an array of bioactive natural products and represents a significant synthetic challenge (2–5, Figure 1).^{5,6} We therefore sought to develop a synthetic route to access melokhanine E (1) to confirm its antibacterial activity while developing innovative solutions to access its pentacyclic core. During the course of our efforts, a 14-step synthesis of 1 was reported as part of studies developing rearrangement approaches toward this broad class of natural products.⁷ Herein we describe a short synthetic sequence to stereoselectively access 1, confirm its chemical structure, and explore its potential as an antimicrobial lead.⁸

A number of strategies have been developed to install the quaternary substituted piperidine motif found in many alkaloids. Pandey and co-workers have utilized a Birch reduction/alkylation followed by auxiliary removal⁹ as well as an approach utilizing a Johnson-Claisen rearrangement that provided greater than 99% *ee*.¹⁰ One of the most successful and broadly used strategies was developed by the Stoltz group, which employs racemic 3,3-disubstituted piperidines and chiral palladium catalysis to obtain enantioenriched 3,3-disubstituted piperidines with good yields and up to 99% enantiomeric excess.^{11–13} While high-yielding and stereoselective, these approaches yield products at nonideal oxidation states for their direct advancement to the target natural products. The approach to the piperidine scaffold presented herein is inspired by Waser and co-workers' efforts on push–pull cyclopropane opening and subsequent cycloadditions, allowing for direct access to piperidines at the desired oxidation states.^{14–16}

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Inspired by Carreira's utilization of a regioisomeric version of our oxindole 1,3-dipole for intermolecular cycloadditions, we set out to devise a concise retrosynthesis of melokhanine E featuring a similar oxindole 1,3-dipole-based intramolecular cycloaddition (Figure 2).^{17–21} Our retrosynthetic approach to

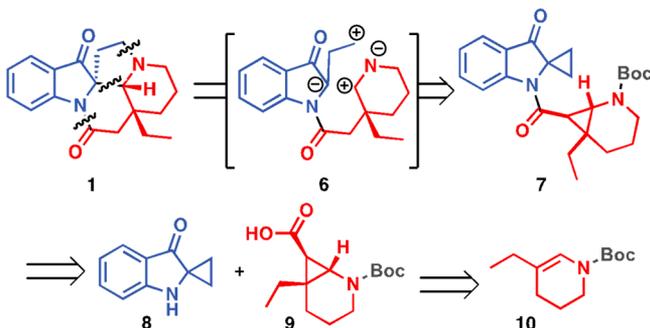


Figure 2. Retrosynthetic access to melokhanine E (1).

1 therefore began by recognizing that intramolecular formal [3 + 2] cycloaddition within biscyclopropane 7 would enable all relative stereochemistry in the molecule to be set in a single transformation based on the configuration of the quaternary piperidine moiety. We envisioned that the stereoselectivity of this transformation would derive from sterically favored exo approach of the revealed oxindole 1,3-dipole functionality into the imine as well as kinetically favored attack of the 1,3-dipole anti- to the ethyl group. We initially proposed that the kinetic preference for anti- addition into the imine would derive from the kinetically favored chairlike transition state that would be formed upon addition onto the antiface of the ring, a process that is likely supplemented by steric predisposition to the antiface of attack. Thus, a late stage intramolecular cycloaddition of an appropriately functionalized 1,3-dipole tethered to an imine or iminium derivative, represented by 6 (Figure 2), enables an extremely rapid approach to the core of 1. Then 6 was envisioned to arise from coupling of spirocyclic cyclopropane 8 with cyclopropanated piperidine 9, followed by subsequent Lewis acid/thermal opening of both cyclopropanes to generate the requisite reactive functionality *in situ*. Both cyclopropane precursors 8 and 9 are available through literature protocols, with 9 arising from cyclopropanation of enecarbamate 10.^{14,22}

The synthesis of coupling partners 8 and 9 was achieved via modification of literature procedures (Figure 3).^{14,22} To target the spirocyclic cyclopropane 8, anthranilic acid (11) was reacted with bromo-lactone 12 to provide amino acid 13 in 72% yield. Activation of the carboxylic acid functionality in the presence of base and heat forges the spirocyclic bicycle 14 in 61% yield. Heating this compound in the presence of NaCl enables a Krapcho decarboxylation-ring contraction sequence that reveals the desired alpha-keto cyclopropane 15 in 73% yield. Finally, methanolysis of 15 affords 8 in four steps and 32% overall yield (Figure 3a).²² Synthesis of the desired fused cyclopropane-piperidine coupling partner 9 begins with an alkylation-protection of delta-valerolactam (16) to generate 17 (61%, two steps). Reduction of the lactam with NaBH₄ followed by dehydration of the resulting hemiaminal with H₂SO₄ affords Boc-protected enamine 10 (82%, two steps). Copper(I) triflate-mediated cyclopropanation of enecarbamate 10 then affords fused ester 18 in 73% yield, which saponifies

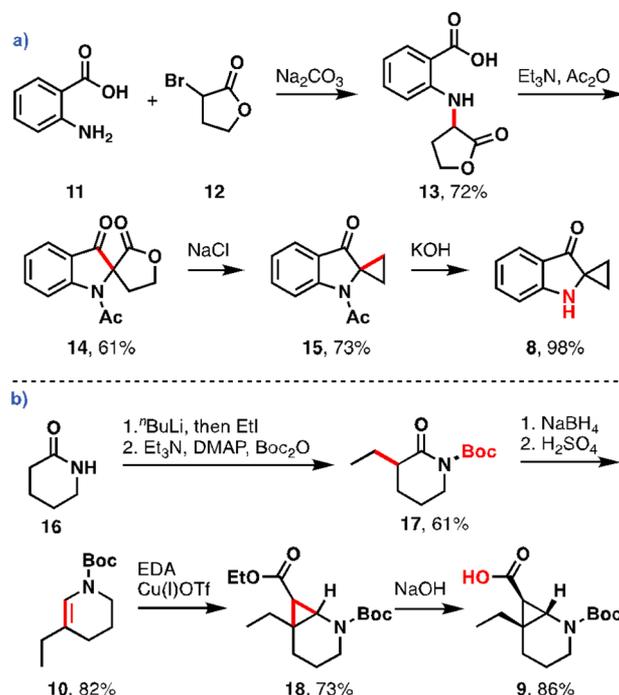


Figure 3. (a) Synthesis of spirocyclic cyclopropane 8; (b) synthesis of fused bicyclic cyclopropane 9.

under basic conditions and is simultaneously epimerized to the more stable exoisomer 9 (Figure 3b).

At this stage, we began to explore coupling conditions to unite 8 and 9 through the amide linkage to set the stage for our key intramolecular cycloaddition. The coupling of indolinone 8 and piperidine 9 proved to be more challenging than anticipated. We postulate that the difficulty of this amide bond formation is a result of steric bulk on both coupling partners as well as deactivation of the nucleophile via resonance with the 3-keto functionality of the indolinone. After exploring various conditions, we discovered that an acid chloride-based coupling approach proved most fruitful, with triphosgene providing our desired acid chloride species with minimal substrate decomposition. Our initial attempt with triphosgene provided 16% yield of the desired coupled product 7 after 16 h at room temperature (Table 1). We found that

Table 1. Optimization of Coupling Conditions for 7

entry	time (h)	temp (°C)	solvent	yield (%)
1	19	rt	CH ₂ Cl ₂	16
2	41	rt	CH ₂ Cl ₂	26
3	3.5	40	CH ₂ Cl ₂	45
4	6	40	CH ₂ Cl ₂	31
5 ^a	6	40	CH ₂ Cl ₂	33
6 ^a	19	60	THF	9
7	52	40	THF	13

^aLutidine was used in place of collidine.

increased temperature allowed for shorter reaction time and higher yield; however, increased reaction time at elevated temperature decreased the overall yield, likely due to slow decomposition of the substrates or product. At this stage, we elected to take these conditions and press forward with the synthesis, having access to **7** in 45% yield. It should be noted that upon isolation and storage **7** can ring-open to the hydrated hemiaminal piperidine motif; however, this has no consequence on the yield of the subsequent cycloaddition chemistry.

With coupled bicyclopropane **7** in hand, we sought to explore the generation of our key cycloaddition intermediates via cyclopropane opening. To this end, **7** was subjected to cycloaddition conditions utilizing MgI_2 to promote spirocycle opening, and thermolysis of the Boc-carbamate to reveal the reactive imine intermediate.^{17–21} We envision a sequence of Boc-deprotection/cyclopropane opening and MgI_2 -promoted cyclopropanation opening to generate the reactive 1,3 dipole **20**, which undergoes stepwise imine addition/nitrogen alkylation to generate the formal [3 + 2] cycloaddition product (Figure 4). In our first attempt (0.43 equiv MgI_2 ,

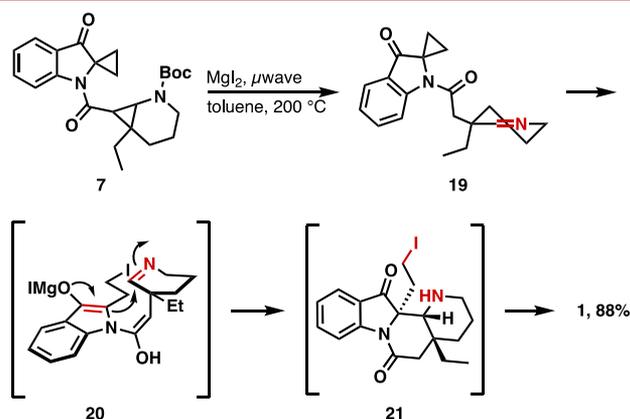


Figure 4. Formal [3 + 2] cycloaddition to synthesize (±)-melokhanine E (**1**).

THF, sealed vessel, 125 °C), we obtained 15% of the desired natural product **1** as a single diastereomer with no other related melokhanine products observed. To our delight, performing the reaction in toluene at 200 °C under microwave heating provided 88% yield of (±)-melokhanine E, again as a single detectable diastereomer (**1**, Figure 4). Interestingly, in the absence of MgI_2 , the reaction still proceeds, albeit at an exceptionally slow rate (29% yield after 8.5 h at 200 °C).

With the racemic natural product in hand, we next sought to confirm the biological activity reported in Luo and co-workers' isolation report.⁴ In the isolation report, Luo and co-workers reported impressive antibiotic activity against Gram-negative bacteria (*P. aeruginosa* MIC 2 μM , 0.62 $\mu g/mL$) as well as Gram-positive bacteria (*E. faecalis* MIC 5 μM , 1.56 $\mu g/mL$). We explored the activity of **1** against both the pathogens reported in the isolation report as well as the ATCC numbers reported²³ and found in all cases the activity of the natural product was $>256 \mu g/mL$. Disappointed by these initial results, we have adjusted the assay conditions to nonstandard protocols (see Supporting Information for biological protocols and Table S1 for full data) and have explored alternate growth media but have not yet uncovered conditions that provide the reported levels of antimicrobial activity. Going forward, access to the

enantiopure natural product will enable us to rule out the unlikely possibility that the unnatural enantiomer has a negative impact on the natural products' antimicrobial activity.

In conclusion, a concise, stereoselective synthesis of (±)-melokhanine E has been reported in eight steps (longest linear sequence) and in 11% overall yield. The key step of our synthesis is a MgI_2 promoted ring opening/cyclization that successfully provides the natural product core in excellent yield and as a single detectable diastereomer. The stereochemical control of this transformation relies on the piperidine C3 all-carbon quaternary center, presenting an opportunity to expand this approach to a number of related natural products with validated biological activities. The current work is focused on developing an asymmetric synthesis of **9** and extending this chemistry to the bisleuconothine family of natural products^{5,24,25} (**5**, Figure 1), and these efforts will be reported in due course.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acs.orglett.9b04546>.

Experimental details, spectroscopic and analytical data of all new compounds (PDF)

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) Wright, P. M.; Seiple, I. B.; Myers, A. G. The Evolving Role of Chemical Synthesis in Antibacterial Drug Discovery. *Angew. Chem., Int. Ed.* **2014**, *53* (34), 8840–8869.

- (2) Rossiter, S. E.; Fletcher, M. H.; Wuest, W. M. Natural Products as Platforms To Overcome Antibiotic Resistance. *Chem. Rev.* **2017**, *117* (19), 12415–12474.
- (3) Abouelhassan, Y.; Garrison, A. T.; Yang, H.; Chávez-Riveros, A.; Burch, G. M.; Huigens, R. W. Recent Progress in Natural-Product-Inspired Programs Aimed To Address Antibiotic Resistance and Tolerance. *J. Med. Chem.* **2019**, *62*, 7618.
- (4) Cheng, G.-G.; Li, D.; Hou, B.; Li, X.-N.; Liu, L.; Chen, Y.-Y.; Lunga, P.-K.; Khan, A.; Liu, Y.-P.; Zuo, Z.-L.; et al. Melokhanines A–J, Bioactive Monoterpenoid Indole Alkaloids with Diverse Skeletons From *Melodinus khasianus*. *J. Nat. Prod.* **2016**, *79* (9), 2158–2166.
- (5) Hirasawa, Y.; Shoji, T.; Arai, T.; Nugroho, A. E.; Deguchi, J.; Hosoya, T.; Uchiyama, N.; Goda, Y.; Awang, K.; Hadi, H. A.; et al. Bisleuconothine A, an Eburnane–Aspidosperma Bisindole Alkaloid from *Leuconotis Griffithii*. *Bioorg. Med. Chem. Lett.* **2010**, *20* (6), 2021–2024.
- (6) Sears, J. E.; Boger, D. L. Total Synthesis of Vinblastine, Related Natural Products, and Key Analogues and Development of Inspired Methodology Suitable for the Systematic Study of Their Structure–Function Properties. *Acc. Chem. Res.* **2015**, *48* (3), 653–662.
- (7) Li, G.; Piemontesi, C.; Wang, Q.; Zhu, J. Stereoselective Total Synthesis of Eburnane-Type Alkaloids Enabled by Conformation-Directed Cyclization and Rearrangement. *Angew. Chem., Int. Ed.* **2019**, *58* (9), 2870–2874.
- (8) Pierce, J. G.; Cholewczynski, A. E.; Williams, P. C. Stereocontrolled Synthesis of Melokhanine E via an Intramolecular Formal [3 + 2] Cycloaddition. *ChemRxiv.* **2019**, DOI: 10.26434/chemrxiv.10022858.v1.
- (9) Pandey, G.; Prasanna, K. C. Iminium Ion Cascade Reaction in the Total Synthesis of (+)-Vincadifformine. *Org. Lett.* **2011**, *13* (17), 4672–4675.
- (10) Pandey, G.; Khamrai, J.; Mishra, A. Generation of All-Carbon Quaternary Stereocenters at the C-3 Carbon of Lactams via [3,3]-Sigmatropic Rearrangement and Revision of Absolute Configuration: Total Synthesis of (–)-Physostigmine. *Org. Lett.* **2018**, *20* (1), 166–169.
- (11) Behenna, D. C.; Liu, Y.; Yurino, T.; Kim, J.; White, D. E.; Virgil, S. C.; Stoltz, B. M. Enantioselective Construction of Quaternary N-Heterocycles by Palladium-Catalyzed Decarboxylative Allylic Alkylation of Lactams. *Nat. Chem.* **2012**, *4* (2), 130.
- (12) Numajiri, Y.; Pritchett, B. P.; Chiyoda, K.; Stoltz, B. M. Enantioselective Synthesis of A-Quaternary Mannich Adducts by Palladium-Catalyzed Allylic Alkylation: Total Synthesis of (+)-Sibirinine. *J. Am. Chem. Soc.* **2015**, *137* (3), 1040–1043.
- (13) Pritchett, B. P.; Kikuchi, J.; Numajiri, Y.; Stoltz, B. M. Enantioselective Pd-Catalyzed Allylic Alkylation Reactions of Dihydropyrido[1,2-a]Indolone Substrates: Efficient Syntheses of (–)-Goniomitine, (+)-Aspidospermidine, and (–)-Quebrachamine. *Angew. Chem., Int. Ed.* **2016**, *55* (43), 13529–13532.
- (14) De Simone, F.; Gertsch, J.; Waser, J. Catalytic Selective Cyclizations of Aminocyclopropanes: Formal Synthesis of Aspidospermidine and Total Synthesis of Goniomitine. *Angew. Chem., Int. Ed.* **2010**, *49* (33), 5767–5770.
- (15) De Simone, F.; Saget, T.; Benfatti, F.; Almeida, S.; Waser, J. Formal Homo-Nazarov and Other Cyclization Reactions of Activated Cyclopropanes. *Chem. - Eur. J.* **2011**, *17* (51), 14527–14538.
- (16) Frei, R.; Staedler, D.; Raja, A.; Franke, R.; Sasse, F.; Gerber-Lemaire, S.; Waser, J. Total Synthesis and Biological Evaluation of Jerantinine E. *Angew. Chem., Int. Ed.* **2013**, *52* (50), 13373–13376.
- (17) Alper, P. B.; Meyers, C.; Lerchner, A.; Siegel, D. R.; Carreira, E. M. Facile, Novel Methodology for the Synthesis of Spiro[Pyrrolidin-3,3'-oxindoles]: Catalyzed Ring Expansion Reactions of Cyclopropanes by Aldimines. *Angew. Chem., Int. Ed.* **1999**, *38* (21), 3186–3189.
- (18) Fischer, C.; Meyers, C.; Carreira, E. M. Efficient Synthesis of (±)-Horsfiline through the MgI₂-Catalyzed Ring-Expansion Reaction of a Spiro[Cyclopropane-1,3'-indol]-2'-one. *Helv. Chim. Acta* **2000**, *83* (6), 1175–1181.
- (19) Lerchner, A.; Carreira, E. M. First Total Synthesis of (±)-Strychnofoline via a Highly Selective Ring-Expansion Reaction. *J. Am. Chem. Soc.* **2002**, *124* (50), 14826–14827.
- (20) Meyers, C.; Carreira, E. M. Total Synthesis of (–)-Spirotryprostatin B. *Angew. Chem., Int. Ed.* **2003**, *42* (6), 694–696.
- (21) Marti, C.; Carreira, E. M. Total Synthesis of (–)-Spirotryprostatin B: Synthesis and Related Studies. *J. Am. Chem. Soc.* **2005**, *127* (32), 11505–11515.
- (22) Kawada, M.; Kawano, Y.; Sugihara, H.; Takei, S.; Imada, I. Spirocyclopropane Compounds. I. Synthesis and Reactivity of Spiro[Cyclopropane-1, 2'-[2H] Indol]-3'(1'H)-Ones. *Chem. Pharm. Bull.* **1981**, *29* (7), 1900–1911.
- (23) There is a discrepancy between the bacteria code identifiers and the strains reported; both the strain reported, and the code identifier were used in these studies. See Supporting Information for full MIC data and the SI for assay protocols.
- (24) Wong, C.; Seki, A.; Horiguchi, K.; Shoji, T.; Arai, T.; Nugroho, A.; Hirasawa, Y.; Sato, F.; Kaneda, T.; Morita, H. Bisleuconothine A Induces Autophagosome Formation by Interfering with AKT-MTOR Signaling Pathway. *J. Nat. Prod.* **2015**, *78* (7), 1656–1662.
- (25) Nugroho, A.; Zhang, W.; Hirasawa, Y.; Tang, Y.; Wong, C.; Kaneda, T.; Hadi, H. A.; Morita, H. Bisleuconothines B–D, Modified Eburnane–Aspidosperma Bisindole Alkaloids from *Leuconotis Griffithii*. *J. Nat. Prod.* **2018**, *81* (11), 2600–2604.