JOURNAL OF NATURAL PRODUCTS

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# Gram-Scale, Stereoselective Synthesis and Biological Evaluation of (+)-Armillariol C

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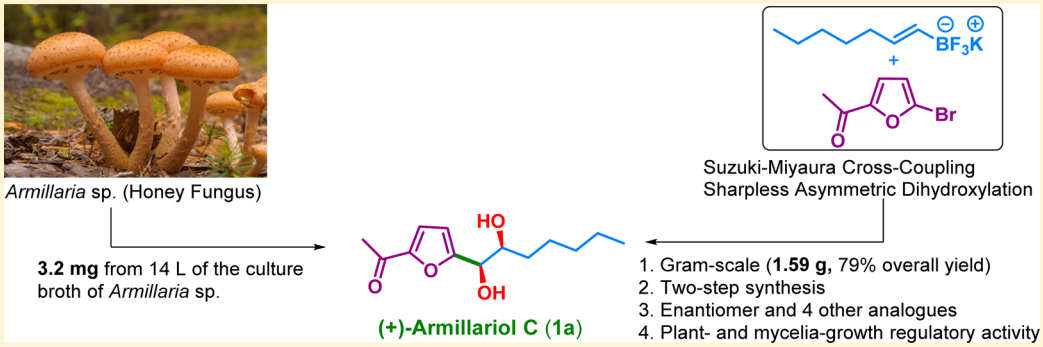
Supporting Information




**ABSTRACT:** Natural products with heteroaromatic cores are ample and widespread in nature, with many compounds exhibiting promising therapeutic properties. (+)-Armillariol C (**1a**) is a furan-based natural product isolated from *Armillaria* species. Herein, we report the first enantioselective synthesis of (+)-armillariol C (**1a**, 79% overall yield), its enantiomer (**1b**), and four other analogues, on a gram-scale, using microwave-mediated, Suzuki−Miyaura cross-coupling and Sharpless asymmetric dihydroxylation reactions. Compounds were tested for plant- and mycelia-growth regulatory activity, with **1b**, **7a**, and **7b** showing the strongest inhibitory properties in a lettuce assay and **7b** and **9b** inhibiting *Flammulina velutipes*.

Heterocycles are ubiquitous and form the core of many agrochemicals and marketed drugs. These compounds function as insecticides, herbicides, plant-growth regulators, fungicides, veterinary products, copolymers, and ligands.[1,2] Furan is a common constituent encountered in numerous biologically active heterocyclic scaffolds including natural products and also serves as a useful building block in several important reactions such as Diels−Alder and Achmatowicz reactions, attaining a prominent role in organic synthesis.[3]

(+)-Armillariol C (**1a**) is a furan-containing natural product isolated as a minor component (3.2 mg) from the culture broth (14 L) of *Armillaria* sp. (honey fungus), possessing plant-growth regulatory activity.[4] *Armillaria* sp. comprise edible mushrooms that display a wide array of biological activity including anticancer,[5,6] anti-inflammatory,[7−9] and antioxidant properties[10] and have been used in traditional Chinese medicine (Tian-ma) to treat hypertension, insomnia, and dizziness.[11,12] Additionally, the genus is responsible for Armillaria root disease, affecting a myriad of woody plants including oak, fruit, and nut trees as well as many herbaceous plants.[13] The structure of (+)-armillariol C encompasses a furan ring with an acetyl group and two asymmetric hydroxy groups on the aliphatic side chain. The synthesis of **1a** represents an attractive challenge for researchers as a way to make the natural product and its analogues in sufficient quantities for further biological testing. Our interest in **1a** stems from its ability to hinder plant growth. We envisioned a concise, stereoselective route to (+)-armillariol C and a variety of analogues that would permit exploration of this biological property. To the best of our knowledge there have been no reports on the synthesis of **1a**. Herein, we describe the first synthesis of (+)-armillariol C in a protecting-group-free manner and our initial exploration of this class of compounds.

Figure 1 outlines a retrosynthetic protocol aimed at the target molecule. The desired natural product (**1a**) could be prepared from conjugated alkene (**2**) using the Sharpless asymmetric dihydroxylation methodology. The alkene (**2**) is easily accessible from heteroaromatic halide **3** and the corresponding alkenyl boronic acid derivative via a Suzuki−Miyaura coupling strategy.

The preparation of armillariol C (**1a**) began with the synthesis of **3** from commercially available 1-(furan-2-yl)ethan-1-one (**4**).[14] Compound **4** was treated with *N*-bromosuccinimide (NBS) in dimethylformamide (DMF) at room temperature, providing compound **3** in 69% yield.

**Received:** June 6, 2017





DOI: 10.1021/acs.jnatprod.7b00484
*J. Nat. Prod.* XXXX, XXX, XXX−XXX

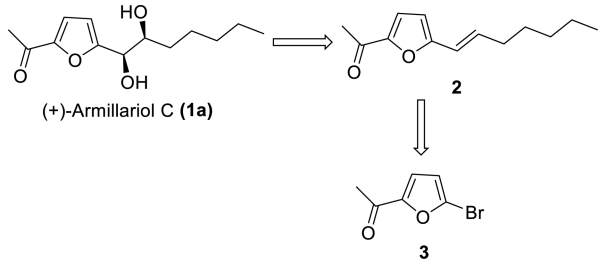
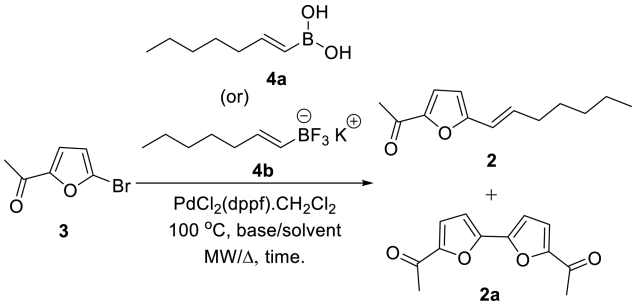



**Figure 1.** Retrosynthetic analysis of target molecule **1a**.

Initial attempts at the preparation of **2** involved the treatment of bromofuran **3** with (*E*)-hept-1-en-1-ylboronic acid (**4a**) under palladium-catalyzed conditions,[15] which furnished **2** in 78% yield, along with 9% of the furan dimer (**2a**, entry 1, Table 1). Attempts to improve the yield of the desired alkene **2** and

**Table 1. Optimization of Reaction Conditions for the Suzuki–Miyaura Reaction**



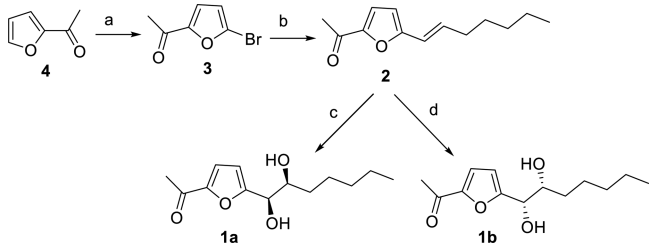

| entry | reagent (**4a**/**4b**) | base/solvent | time (h) | yield % (**2**/**2a**)[b] |
|---|---|---|---|---|
| 1 | **4a** | *t*-BuNH$_2$/*i*-PrOH–H$_2$O | 16 | 78/9 |
| 2[c] | **4a** | *t*-BuNH$_2$/*i*-PrOH–H$_2$O | 0.5 | 81/5 |
| 3[c] | **4a** | *n*-BuNH$_2$/*i*-PrOH–H$_2$O | 0.5 | 56/6 |
| 4[c] | **4a** | EtNH$_2$/*i*-PrOH–H$_2$O | 0.5 | 45/8 |
| 5[c] | **4a** | *t*-BuNH$_2$/1,4-dioxane–H$_2$O | 0.5 | 76/0 |
| 6 | **4b** | *t*-BuNH$_2$/*i*-PrOH–H$_2$O | 12 | 83/0 |
| 7[c] | **4b** | *t*-BuNH$_2$/*i*-PrOH–H$_2$O | 0.5 | 87/0 |

[a]Reagents and conditions: bromide **3** (1 mmol), alkenyl borate (**4a**/**4b**) (1.2 mmol), PdCl$_2$(dppf)·CH$_2$Cl$_2$ (0.02 mol %), base (3 mmol), solvent (5 mL, 2:1). [b]*All* products were characterized by $^1$H and $^{13}$C NMR, IR, and MS. [c]Microwave conditions.

eliminate the production of **2a** using bases such as *n*-BuNH$_2$ and ethylamine and a 1,4-dioxane/water mixture were unsuccessful (entries 2–5, Table 1). Switching to an alternative organoboron, alkenyltrifluoroborate **4b** proved to be advantageous. Suzuki–Miyaura coupling[16−18] of potassium (*E*)-trifluoro(hept-1-en-1-yl)borate (**4b**) with **3** in the presence of a Pd-catalyst at 100 °C for 12 h under conventional heating afforded the desired compound **2** in 83% yield as an exclusive product (entry 6, Table 1).

Likewise, coupling of **4b** with **3** under microwave irradiation gave exclusively **2** in 87% yield after 30 min at 100 °C (entry 7, Table 1). Sharpless asymmetric dihydroxylation[19,20] provided access to both (+)- and (−)-armillariol C (**1a** and **1b**). Treatment of **2** with ADmix-$\alpha$ and methanesulfonamide in *t*-BuOH-H$_2$O at 0 °C for 24 h gave the anticipated (+)-armillariol C (**1a**) in 91% yield and >98% ee (Scheme 1). Similarly, (−)-armillariol C (**1b**, 89% yield, > 98% ee) was synthesized by treating **2** with ADmix-$\beta$ under identical

**Scheme 1. Synthesis of (+)-Armillariol C (1a) and Its Enantiomer (1b)[a]**

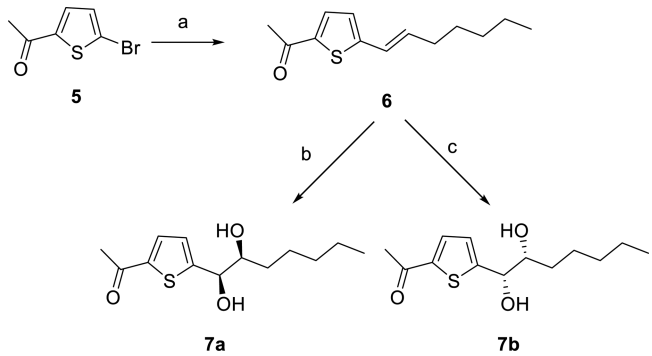



[a]Reagents and conditions: (a) NBS, DMF, rt, 16 h, 69%; (b) see Table 1; (c) ADmix-$\alpha$, MeSO$_2$NH$_2$, *t*-BuOH/H$_2$O (1:1), 0 °C, 24 h, 91%; (d) ADmix-$\beta$, MeSO$_2$NH$_2$, *t*-BuOH/H$_2$O (1:1), 0 °C, 24 h, 89%.

conditions. Enantiomeric excess was determined using the method of Bull and James[21] (see Experimental Section for details).

After successful synthesis of armillariol C, we focused on the synthesis of various analogues. The treatment of **4b** with commercially available 1-(5-bromothiophen-2-yl)ethan-1-one (**5**), under Pd-catalyzed and microwave-irradiated conditions, afforded the corresponding alkene **6** in excellent yield (Scheme 2). Asymmetric dihydroxylation of **6** with ADmix-$\alpha$ and ADmix-$\beta$ gave the enantiomeric diols **7a** in 90% (>98% ee) and **7b** in 86% yield (>98% ee), respectively.

**Scheme 2. Synthesis of Thiophene Analogues of Armillariol C[a]**




[a]Reagents and conditions: (a) **4b**, PdCl$_2$(dppf)·CH$_2$Cl$_2$, *t*-BuNH$_2$, *i*-PrOH/H$_2$O (2:1), 100 °C, MW, 30 min, 90%; (b) ADmix-$\alpha$, MeSO$_2$NH$_2$, *t*-BuOH/H$_2$O (1:1), 0 °C, 24 h, 90%; (c) ADmix-$\beta$, MeSO$_2$NH$_2$, *t*-BuOH/H$_2$O (1:1), 0 °C, 24 h, 86%.

Additionally, the treatment of **3** with potassium (*E*)-trifluoro(styryl)borate (**4c**) under Pd-catalyzed and microwave-irradiated conditions at 100 °C for 45 min afforded the heteroaryl alkene **8** in excellent yield (Scheme 3). Finally, **8** was subjected to ADmix-$\alpha$ and ADmix-$\beta$ to yield the syn, enantiomeric diols (**9a** and **9b**, >98% ee) in good yields.

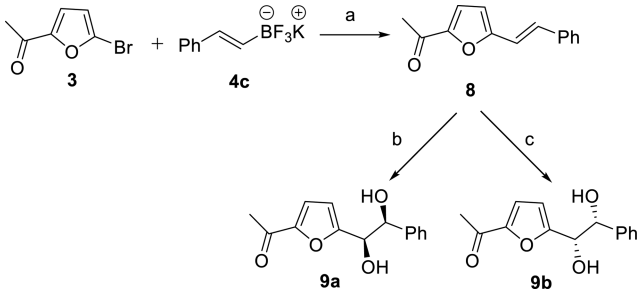
The ability of (+)-armillariol C (**1a**) and its analogues to regulate plant growth was determined at a range of concentrations using lettuce (Figure 2).[22] All synthesized analogues inhibited the growth of lettuce, while analogues **1b**, **7a**, and **7b** showed the strongest inhibition of growth of both the hypocotyl and root.

Further testing was conducted to determine the ability of the diols to inhibit the growth of mycelia from *Flammulina velutipes*.[22] Compounds **7b** and **9b** inhibited the mycelia growth of *F. velutipes* as indicated by the presence of clear zones on



DOI: 10.1021/acs.jnatprod.7b00484
J. Nat. Prod. XXXX, XXX, XXX−XXX

**Scheme 3. Synthesis of Analogues of Armillariol C**[a]




[a]Reagents and conditions: (a) $PdCl_2(dppf)\cdot CH_2Cl_2$, *t*-$BuNH_2$, *i*-PrOH:$H_2O$ (2:1), 100 °C, MW, 45 min, 92%; (b) ADmix-*α*, $MeSO_2NH_2$, *t*-BuOH/$H_2O$ (1:1), 0 °C, 72 h, 87%; (c) ADmix-*β*, $MeSO_2NH_2$, *t*-BuOH/$H_2O$ (1:1), 0 °C, 72 h, 88%.

potato dextrose agar (PDA) plates (see Supporting Information).

In conclusion, the synthesis of (+)- and (−)-armillariol C and their analogues using Suzuki−Miyaura cross-coupling and Sharpless asymmetric dihydroxylation on a gram-scale is described. (+)-Armillariol C (**1a**) was synthesized for the first time and in a two-step protocol from 1-(5-bromofuran-2-yl)ethan-1-one (**3**) in 79% overall yield. Additionally, the plant- and mycelia-growth regulatory activity of the natural product and its analogues were also investigated, with **1b**, **7a**, and **7b** showing the strongest inhibitory properties in a lettuce assay and **7b** and **9b** inhibiting *F. velutipes*.

## EXPERIMENTAL SECTION

**General Experimental Procedures.** Glassware was dried in an oven (120 °C), heated under reduced pressure, and cooled under argon before use. Unless otherwise noted, materials obtained from commercial suppliers were used without further purification. Reactions were monitored by thin-layer chromatography on Analtech silica gel plates using UV light and ceric sulfate or *β*-naphthol for visualization. Column chromatography was performed on silica gel (230−400 mesh) using hexanes and ethyl acetate as eluent. Evaporation of solvents was conducted under reduced pressure at 50 °C. FTIR spectra were recorded neat on a PerkinElmer Spectrum 65. Specific rotation values were determined using an Anton-Paar MCP 100 polarimeter. Microwave reactions were conducted in a Biotage Initiator Classic. NMR spectra were recorded on a Bruker Avance III 400 NMR spectrometer at 400 MHz ($^1H$) and 100 MHz ($^{13}C$), respectively. Deuterated chloroform was used as the solvent, and spectra were calibrated against the residual solvent peak (7.24 ppm for $^1H$ and 77.0 ppm for $^{13}C$). Chemical shifts ($\delta$) and coupling constants ($J$) are given in ppm (parts per million) and Hz (hertz), respectively. The following abbreviations were used to explain multiplicities: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet, bs = broad singlet. Low-resolution ESI mass spectra were obtained on a Waters Acquity UPLC H-Class with PDA and SQ mass detectors using a Waters BEH $C_{18}$ 1.7 $\mu$m column (2.1 × 50 mm). High-resolution mass spectra were obtained on a VG 70-70H or LC/MSD trapSL spectrometer operating at 70 eV using a direct inlet system.

**Synthesis of 1-(5-Bromofuran-2-yl)ethan-1-one (3).** To a stirred solution of 1-(furan-2-yl)ethan-1-one (**4**, 5 g, 45.4 mmol, 1 equiv) in DMF (50 mL) at 10 °C was added portionwise *N*-bromosuccinimide (8.79 g, 49.9 mmol, 1.1 equiv) over a period of 10 min. The mixture was stirred for another 16 h at room temperature and then poured into ice-cold water (150 mL). The product was extracted with diethyl ether (100 mL × 3), the combined organic layers were washed with cold water (100 mL × 2) and brine (100 mL × 2) and dried over $MgSO_4$, and the solvent was removed *in vacuo*. The crude product was purified by flash column chromatography (silica gel, EtOAc/hexanes, 1:50 to 1:20, $R_f$ = 0.25 in 10% EtOAc in hexanes) to give the brominated compound **3** in 69% yield as a pale yellow solid.

**Potassium (*E*)-Trifluoro(hept-1-en-1-yl)borate (4b).** To a solution of 1-heptyne (5.43 mL, 4 g, 41.6 mmol, 1 equiv) in $CH_2Cl_2$ (25 mL) at 0 °C was added dibromoborane dimethyl sulfide complex ($Br_2BH\cdot SMe_2$) (41.6 mL, 41.6 mmol, 1 M in $CH_2Cl_2$, 1 equiv). The mixture was stirred at room temperature for 4 h. Subsequently, the light green solution was added via a double-ended needle to a mixture of diethyl ether/water (25 mL of diethyl ether and 10 mL of water) at 0 °C. The mixture was stirred at room temperature for 30 min. The aqueous layer was separated, and the organic phase was washed with cold water and then brine. The organic phase was subsequently dried with $MgSO_4$. The solvents were evaporated to provide (*E*)-hept-1-en-1-ylboronic acid (**4a**) as a white solid.

To a solution of the *trans*-hept-1-en-1-ylboronic acid thus prepared (**4a**, 5.3 g) in diethyl ether (60 mL) in a 250 mL plastic beaker/narrow mouth plastic bottle was added $KHF_2$ (9.08 g, 116 mmol, 2.8 equiv), followed by slow addition of water (25 mL) over a period of 30 min. After stirring at room temperature for 3 h, the solution was concentrated, and the crude material was dissolved in acetone, filtered, and concentrated. The resulting white solid was purified by dissolving in hot acetone and precipitating with $Et_2O$, resulting in a white solid (**4b**, 5.52 g, 65%).

**General Procedure for the Preparation of *trans*-Alkenes (2/6/8).** A solution of potassium *trans*-alkene trifluoroborate (**4a**/**4b**/**4c**) (6.32 mmol, 1.2 equiv), $PdCl_2(dppf)\cdot CH_2Cl_2$ (86 mg, 0.106 mmol,

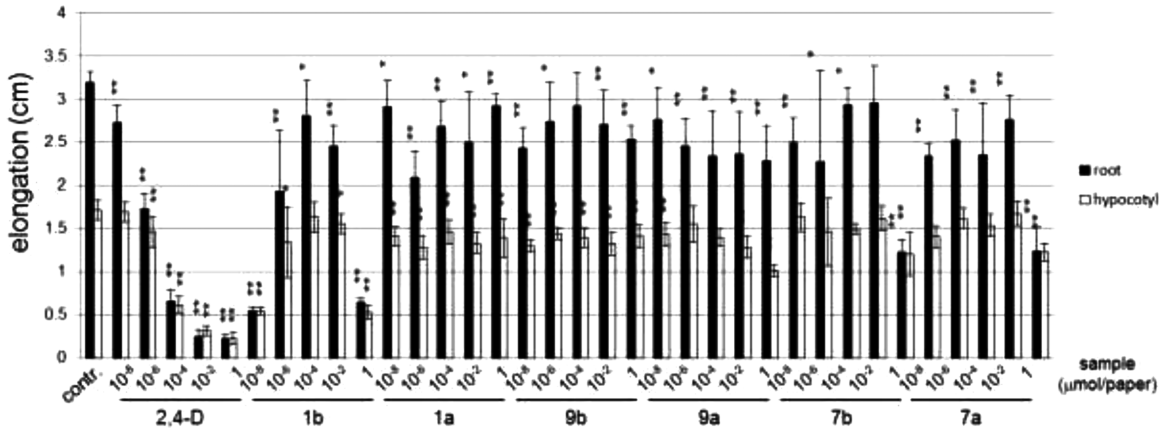



**Figure 2.** Lettuce growth regulating activity of armillariol C (**1a**) and analogues against root or hypocotyl. 2,4-Dichlorophenoxyacetic acid (2,4-D) was used as the positive control. Results are the mean ± standard deviation ($n$ = 7). The asterisk indicates significant difference from control (Student's $t$ test; *, $p < 0.05$; **, $p < 0.01$).



DOI: 10.1021/acs.jnatprod.7b00484
J. Nat. Prod. XXXX, XXX, XXX−XXX

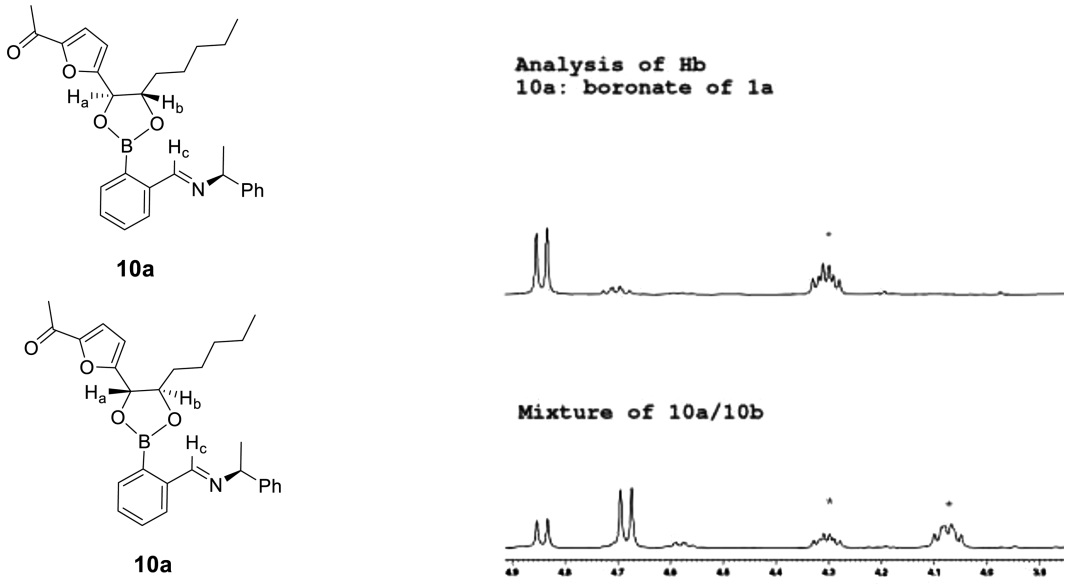



**Figure 3.** Determination of enantiomeric excess of **1a** via $^1$H NMR of boronate **10a**.[21]

0.02 equiv), 1-(5-bromofuran-2-yl)ethan-1-one (**3**, 1 g, 5.29 mmol, 1 equiv)/1-(5-bromothiophen-2-yl)ethan-1-one (**5**, 1 g, 4.88 mmol, 1 equiv), and *t*-BuNH$_2$ (15.81 mmol, 3 equiv) in *i*-PrOH/H$_2$O (2:1, 24 mL) was heated at 100 °C under an argon atmosphere for 30 min under microwave conditions (12 h in the case of conventional heating on an oil bath), then cooled to room temperature (RT) and diluted with water (20 mL), followed by extraction of diethyl ether (50 mL × 2). The organic layers were combined and washed with 1 N HCl (80 mL) and brine (80 mL), dried over MgSO$_4$, and then filtered. The solvent was removed under vacuum, and the crude product was purified by silica gel chromatography (eluting with hexanes/ethyl acetate, 20:1 to 5:1) to afford the corresponding alkene derivative (**2**/**6**/**8**) in good yields.

**(*E*)-1-(5-(Hept-1-en-1-yl)furan-2-yl)ethan-1-one (2).** 1-(5-Bromofuran-2-yl)ethan-1-one (**3**, 1 g, 5.29 mmol, 1 equiv), potassium (*E*)-trifluoro(hept-1-en-1-yl)borate (**4b**, 1.30 g, 6.32 mmol, 1.2 equiv), PdCl$_2$(dppf)·CH$_2$Cl$_2$ (86 mg, 0.106 mmol, 0.02 equiv), and *t*-BuNH$_2$ (1.661 mL, 1.156 g, 15.81 mmol, 3 equiv) in *i*-PrOH/H$_2$O (2:1, 24 mL), 30 min, 100 °C. After column chromatography (gradient, hexanes/EtOAc 50:1 to 20:1) 948 mg (87%) of a pale, yellow oil was obtained.

**(*E*)-1-(5-(Hept-1-en-1-yl)thiophen-2-yl)ethan-1-one (6).** 1-(5-Bromothiophen-2-yl)ethan-1-one (**5**, 1 g, 4.88 mmol, 1 equiv), potassium (*E*)-trifluoro(hept-1-en-1-yl)borate (**4b**, 1.19 g, 5.85 mmol, 1.2 equiv), PdCl$_2$(dppf)·CH$_2$Cl$_2$ (80 mg, 0.098 mmol, 0.02 equiv), and *t*-BuNH$_2$ (1.07 g, 14.63 mmol, 3 equiv) in *i*-PrOH/H$_2$O (2:1, 24 mL), 30 min, 100 °C. After column chromatography (gradient, hexanes/EtOAc, 50:1 to 20:1) 975 mg (90%) of a pale, yellow solid was obtained.

**(*E*)-1-(5-Styrylfuran-2-yl)ethan-1-one (8).** 1-(5-Bromofuran-2-yl)ethan-1-one (**3**, 1 g, 5.29 mmol, 1 equiv), potassium (*E*)-trifluoro(styryl)borate (**4c**, 1.33 g, 6.35 mmol, 1.2 equiv), PdCl$_2$(dppf)·CH$_2$Cl$_2$ (86 mg, 0.106 mmol, 0.02 equiv), and *t*-BuNH$_2$ (1.161 g, 15.87 mmol, 3 equiv) in *i*-PrOH/H$_2$O (2:1, 24 mL), 45 min, 100 °C. After column chromatography (gradient, hexanes/EtOAc, 50:1 to 20:1) 1.03 g (92%) of a pale, yellow solid was obtained.

**1,1′-([2,2′-Bifuran]-5,5′-diyl)bis(ethan-1-one) (2a).** The dimer byproduct was isolated from the reaction of 1-(5-bromofuran-2-yl)ethan-1-one with alkenyl boronic acids; 15 mg (9% of dimer **2a** in the synthesis of (*E*)-1-(5-(hept-1-en-1-yl)furan-2-yl)ethan-1-one, **2**) of a pale, yellow solid was obtained.

**(+)-Armillariol C (1a).** To an ice-cold mixture of (*E*)-1-(5-(hept-1-en-1-yl)furan-2-yl)ethan-1-one (**2**, 1.5 g, 7.27 mmol, 1 equiv), *t*-BuOH (50 mL), and H$_2$O (50 mL) were added MeSO$_2$NH$_2$ (692 mg, 7.27 mmol, 1 equiv) and ADmix-$\alpha$ (10.0 g, ∼1.4 g for 1 mmol of alkene **2**). The mixture was stirred for 24 h at 0 °C and diluted with brine (40 mL). The product was extracted with ethyl acetate (50 mL × 3). The combined organic layers were washed with brine (100 mL), dried over MgSO$_4$, and concentrated. The residue was purified by chromatography (hexanes/ethyl acetate, 70:30) to afford (+)-armillariol C (**1a**) as a pale, yellow oil (1.59 g) in 91% yield (>98% ee).

**(−)-Armillariol C (1b).** Following the general procedure used for the synthesis of (+)-**1a**: (*E*)-1-(5-(hept-1-en-1-yl)furan-2-yl)ethan-1-one (**2**, 1.5 g, 7.27 mmol, 1 equiv), MeSO$_2$NH$_2$ (692 mg, 7.27 mmol, 1 equiv), ADmix-$\beta$ (10.0 g, ∼1.4 g for 1 mmol of alkene **2**) in *t*-BuOH/H$_2$O (1:1, 100 mL), 24 h, 0 °C. After column chromatography (hexanes/EtOAc, 90:10 to 70:30) 1.55 g (89%) of a pale, yellow oil was obtained (>98% ee).

**(−)-1-(5-((1*R*,2*S*)-1,2-Dihydroxyheptyl)thiophen-2-yl)ethan-1-one (7a).** Following the general procedure used for the synthesis of (+)-**1a**: (*E*)-1-(5-(hept-1-en-1-yl)thiophen-2-yl)ethan-1-one (**6**, 1.5 g, 6.75 mmol, 1 equiv), MeSO$_2$NH$_2$ (642 mg, 5 mmol, 1 equiv), ADmix-$\alpha$ (9.6 g, ∼1.4 g for 1 mmol of alkene) in *t*-BuOH/H$_2$O (1:1, 100 mL), 24 h, 0 °C. After column chromatography (hexanes/EtOAc, 90:10 to 70:30) 1.55 g (90%) of a pale, yellow oil was obtained (>98% ee).

**(+)-1-(5-((1*S*,2*R*)-1,2-Dihydroxyheptyl)thiophen-2-yl)ethan-1-one (7b).** Following the general procedure used for the synthesis of (+)-**1a**: (*E*)-1-(5-(hept-1-en-1-yl)thiophen-2-yl)ethan-1-one (**6**, 1.5 g, 6.75 mmol, 1 equiv), MeSO$_2$NH$_2$ (642 mg, 6.75 mmol, 1 equiv), ADmix-$\beta$ (9.6 g, ∼1.4 g for 1 mmol of alkene) in *t*-BuOH/H$_2$O (1:1, 100 mL), 24 h, 0 °C. After column chromatography (gradient, hexanes/EtOAc, 90:10 to 70:30) 1.49 g (86%) of a pale, yellow oil was obtained (>98% ee).

**(−)-1-(5-((1*R*,2*S*)-1,2-Dihydroxy-2-phenylethyl)furan-2-yl)ethan-1-one (9a).** Following the general procedure used for the synthesis of (+)-**1a**: (*E*)-1-(5-styrylfuran-2-yl)ethan-1-one (**8**, 1.5 g, 7.07 mmol, 1 equiv), MeSO$_2$NH$_2$ (672 mg, 7.07 mmol, 1 equiv), ADmix-$\alpha$ (9.90 g, ∼1.4 g for 1 mmol of alkene) in *t*-BuOH/H$_2$O (1:1, 100 mL), 72 h, 0 °C. After column chromatography (hexanes/EtOAc, 90:10 to 70:30) 1.51 g (87%) of a pale, yellow oil was obtained (>98% ee).

**(+)-1-(5-((1*S*,2*R*)-1,2-Dihydroxy-2-phenylethyl)furan-2-yl)ethan-1-one (9b).** Following the general procedure used for the synthesis of (+)-**1a**: (*E*)-1-(5-styrylfuran-2-yl)ethan-1-one (**8**, 1.5 g, 7.07 mmol, 1 equiv), MeSO$_2$NH$_2$ (672 mg, 7.07 mmol, 1 equiv), ADmix-$\beta$ (9.90 g, ∼1.4 g for 1 mmol of alkene) in *t*-BuOH/H$_2$O (1:1, 40 mL), 24 h, 0 °C. After column chromatography (hexanes/EtOAc,


DOI: 10.1021/acs.jnatprod.7b00484
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90:10 to 70:30) 1.53 g (88%) of a pale, yellow oil was obtained (>98% ee).

**General Procedure for the Determination Enantiomeric Excess.**[21] The diol (1.0 equiv), 2-formylphenylboronic acid (1.0 equiv), and (*S*)-(α)-benzylamine (1.0 equiv) were dissolved in $CDCl_3$ under an argon atmosphere. After stirring at RT for 15 min, an aliquot was analyzed by $^1H$ NMR. The doublet for $H_a$, multiplet for $H_b$, or singlet for $H_c$ was utilized to determine enantiomeric excess. Figure 3 shows the resonance peaks for $H_b$ of **10a** and **10b**. In each instance, none of the corresponding enantiomer was detected; thus, owing to the limits of $^1H$ NMR, the enantiomeric excess of each isomer was reported as >98%.

**Bioassay.**[22] Lettuce seeds were put on filter paper (Advantec No. 2, ϕ 55 mm; Toyo Roshi Kaisha, Japan), soaked in distilled water in a Petri dish (ϕ 60 × 20 mm), and incubated in a growth chamber under darkness at 25 °C for 1 day. Each sample was dissolved in 1 mL of methanol (1, $10^{-2}$, $10^{-4}$, $10^{-6}$, and $10^{-8}$ μmol/mL) and then poured on the filter paper (ϕ 55 mm) in a Petri dish (ϕ 60 × 20 mm). After the sample-loaded paper had been air-dried, 1 mL of distilled water was poured on the sample-loaded paper or intact filter paper (control). The preincubated lettuce seedlings ($n$ = 9 in each Petri dish) were transferred onto the sample-loaded filter paper or control filter paper and incubated in a growth chamber under darkness at 25 °C for 3 days. The lengths of the hypocotyl and the root were measured using a ruler.

Growth activity against the mycelia of *F. velutipes* was examined as follows. The mycelia of *F. velutipes* were placed onto a PDA plate and incubated at 25 °C for 1 day in an incubator. Meanwhile, the test compounds or control solution (in MeOH) was added to autoclaved paper disks (Advantec ϕ 8 mm; Toyo Roshi Kaisha, Ltd., Japan) and then air-dried. Each air-dried paper disk containing 1 μmol/paper disk of the compounds was placed directly onto the incubated plate. Plates were further incubated at 25 °C for 1−2 weeks. After the incubation, the inhibitory activity was evaluated by observation of clear zones due to growth inhibition of mycelia. The observation was repeated in duplicate for all of the test plates over a period of 2 weeks.

## ■ ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.jnatprod.7b00484.

Physical data, assay results, and copies of $^1H$ and $^{13}C\{^1H\}$ NMR spectra (PDF)

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### Notes

The authors declare no competing financial interest.

## ■ ACKNOWLEDGMENTS

This research was funded by Union University. The authors are grateful for the HRESIMS data provided by D. R. Phillips and C.-W. Chou [Proteomics and Mass Spectrometry (PAMS) Facility, NIH grant 1S10RR1028859] at the University of Georgia, Department of Chemistry, Athens, Georgia.

## ■ REFERENCES


(1) Pozharskiĭ, A. F.; Katritzky, Alan, R.; Soldatenkov, A. T. *Heterocycles in Life and Society: An Introduction to Heterocyclic Chemistry, Biochemistry and Applications*, 2nd ed.; Wiley: Chichester, West Sussex, 2011.

(2) Lukevits, É.; Demicheva, L. *Chem. Heterocycl. Compd.* **1993**, *29*, 243−267.

(3) Reddy, C. R.; Krishna, G.; Reddy, M. D. *Org. Biomol. Chem.* **2014**, *12*, 1664−1670.

(4) Kobori, H.; Sekiya, A.; Yasuda, N.; Noguchi, K.; Suzuki, T.; Choi, J.-H.; Hirai, H.; Kawagishi, H. *Tetrahedron Lett.* **2013**, *54*, 5481−5483.

(5) Li, Z.; Wang, Y.; Jiang, B.; Li, W.; Zheng, L.; Yang, X.; Bao, Y.; Sun, L.; Huang, Y.; Li, Y. *J. Ethnopharmacol.* **2016**, *184*, 119−127.

(6) Chen, C.-C.; Kuo, Y.-H.; Cheng, J.-J.; Sung, P.-J.; Ni, C.-L.; Chen, C.-C.; Shen, C.-C. *Molecules* **2015**, *20*, 9994−10003.

(7) Geng, Y.; Zhu, S.; Lu, Z.-M.; Xu, H.-Y.; Cheng, P.; Shi, J.-S.; Xu, Z.-H. *Int. J. Med. Mushrooms* **2014**, *16*, 319−325.

(8) Chang, C.-W.; Lur, H.-S.; Lu, M.-K.; Cheng, J.-J. *Food Res. Int.* **2013**, *54*, 239−245.

(9) Geng, Y.; Zhu, S.; Cheng, P.; Lu, Z.-M.; Xu, H.-Y.; Shi, J.-S.; Xu, Z.-H. *Phytomedicine* **2017**, *26*, 55−61.

(10) Zhang, S.; Liu, X.; Yan, L.; Zhang, Q.; Zhu, J.; Huang, Na; Wang, Z. *Molecules* **2015**, *20*, 5680−5697.

(11) Chen, Y.-J.; Wu, S.-Y.; Chen, C.-C.; Tsao, Y.-L.; Hsu, N.-C.; Chou, Y.-C.; Huang, H.-L. *J. Funct. Foods* **2014**, *6*, 196−204.

(12) Donatini, B. *Phytotherapie* **2013**, *11*, 39−41.

(13) Robinson-Bax, C.; Fox, R. T. V. *Mycologist* **2002**, *16*, 21−22.

(14) Ismail, M. A.; Brun, R.; Wenzler, T.; Tanious, F. A.; Wilson, W. D.; Boykin, D. W. *J. Med. Chem.* **2004**, *47*, 3658−3664.

(15) Thimmaiah, M.; Zhang, X.; Fang, S. *Tetrahedron Lett.* **2008**, *49*, 5605−5607.

(16) Alacid, E.; Nájera, C. *J. Org. Chem.* **2009**, *74*, 8191−8195.

(17) Molander, G. A.; Bernardi, C. R. *J. Org. Chem.* **2002**, *67*, 8424−8429.

(18) Noreen, M.; Rasool, N.; El Khatib, M.; Molander; Gary, A. *J. Org. Chem.* **2014**, *79*, 7243−7249.

(19) Kobayashi, Y.; Nakano, M.; Kumar, G. B.; Kishihara, K. *J. Org. Chem.* **1998**, *63*, 7505−7515.

(20) Ren, J.; Liu, Y.; Song, L.; Tong, R. *Org. Lett.* **2014**, *16*, 2986−2989.

(21) Kelly, A. M.; Perez-Fuertes, Y.; Arimori, S.; Bull, S. D.; James, T. D. *Org. Lett.* **2006**, *8*, 1971−1974.

(22) Kobori, H.; Sekiya, A.; Suzuki, T.; Choi, J.-H.; Hirai, H.; Kawagishi, H. *J. Nat. Prod.* **2015**, *78*, 163−167.




DOI: 10.1021/acs.jnatprod.7b00484
*J. Nat. Prod.* XXXX, XXX, XXX−XXX