Bioactive Constituents, Metabolites, and Functions

Enantioselective Synthesis and Antifungal Activity of C18 Polyacetylenes

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Enantioselective Synthesis and Antifungal Activity of C18 Polyacetylenes

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Abstract

Fungal pathogens cause serious crop diseases and decrease crop yields and quality. Polyacetylene alcohols are plant secondary metabolites and bioactive against various pathogenic fungi. They are, however, difficult to synthesize. In the present study, an efficient and highly enantioselective method (> 98% *ee*) was established and employed to achieve the synthesis of the natural C18 polyacetylenes (***S***, ***E***)-octadeca-1,9-dien-4,6-diyn-3-ol **1**, (**3*R***, **10*R***, ***E***)-octadeca-1,8-dien-4,6-diyne-3,10-diol **2** and their analogs. The title compounds were structurally characterized and biologically evaluated for fungicidal activities. The compounds exhibited high potencies against eight pathogenic fungal species tested, such as *Colletotrichum gloeosporioiles*, *Bipolaris sorokiniana*, *Fusarium graminearum* and *Fusarium pseudograminearum*, with half-maximum effective concentrations ranging from 8 to 425 μg/mL, being similar to those of the fungicide thiophanate-methyl (3 - 408 μg/mL). These compounds are potential natural fungicides and fungicide lead candidates for further structural and property improvements.

Key words: Antifungal, Asymmetric synthesis, Fungicide, *Panax Stipuleanatus*, Polyacetylene

INTRODUCTION

Fungal pathogens are widely distributed and cause serious damage to crops. Many plants secrete secondary metabolites to defend against pathogenic invasion. Those metabolites reveal antifungal effects,[1-5] which indicate a great potential for the development of fungicides. A classic example is the discovery of strobilurins from *Strobilurus tenacellus*, which opened the door for the development of one of the best salable fungicides – azoxystrobin.[6-9] Pyrethroid and neonicotinoid insecticides are also good examples of pesticides developed from natural products.[10-11]

Polyacetylene alcohols are widespread defensive secondary metabolites in plants such as *Umbelliferae* and *Araliaceae*. In an early report in 1978, falcarindiol, a C17 polyacetylene alcohol, was discovered to exhibit antifungal activity to *Uromyces fabae* and *Colletotrichum lagenarium*.[12] In 2012, Kim's group found two new C18 polyacetylenes from *Panax stipuleanatus*,[13-14] which showed biological activities against HCT-116 cancer cell better than mitoxantrone. Further studies have shown that polyacetylene alcohols have beneficial effects on acute alcohol-induced oxidative stress and liver damage as well as antioxidant activity, intensive cell differentiation activity and exhibited dose-dependent inhibitory effects on the cancer cell proliferation.[15-20] The diverse biological activities indicate that polyacetylene alcohols have the potential to be developed as natural product drugs and fungicides, especially as natural fungicides from plant origin.[19]

However, the unique structures of chiral conjugated diynol make the compounds difficult to store and synthesize, which restrict the development and application of polyacetylene alcohols.[21] Those limitations prompted us to develop an efficient and highly enantioselective method and to employ it for the synthesis of these natural products with a large structural diversity.

In 2016, our group achieved the C17 polyacetylenes virol A and C by using chiral alkynols, which was proved to be a reasonable synthetic block for the preparation of polyacetylene alcohols (Figure 1).[22] In the present study, a general method was established to synthesize the natural C18 polyacetylenes (***S***, ***E***)-octadeca-1,9-dien-4,6-diyn-3-ol **1** and (**3*R***, **10*R***, ***E***)-octadeca-1,8-dien-4,6-diyne-3,10-diol **2** and their analogs with a large structural diversity. The synthetic compounds inhibited eight pathogenic fungal species tested, such as *Colletotrichum gloeosporioiles*, *Bipolaris sorokiniana*, *Fusarium graminearum* and *Fusarium pseudograminearum*. The half-maximum effective concentrations (EC_{50}) of the synthetic polyacetylene alcohols ranged from 8 to 425 μg/mL, which are comparable to the potencies, but differ in the anti-fungal spectrum of the commercial fungicide thiophanate-methyl. Those polyacetylene alcohols can be fungicide lead candidates.

MATERIALS AND METHODS

General Information. All reactions were performed under argon atmosphere. Solvents were dried according to standard procedures and distilled prior to use.[23] All reagents were purchased commercially and used without further purification unless stated otherwise. ^{1}H, ^{13}C and 2D NMR data were recorded with a Bruker Avance™ III 400MHz spectrometer (Billerica, MA). High-resolution mass spectra were recorded on an Agilent 6224 TOF instrument (Santa Clara, CA) by the TOF MS technique. The fused silica capillary column was a DB-5MS column (Agilent Technologies). Electron impact (70 eV) mass spectra were recorded from m/z 50 to 550. Enantiomeric excess (*ee*) was determined by chiral HPLC (Prominence LC-20A, Kyoto, Japan) analyses using a chiral column (Chiralpak OD-H), and elution with isopropanol-hexane.

Chiralcel OD: 0.8 ml/min, Hexane/2-propanol (65/35), 220 nm. The optical rotations were measured on a Perkin Elmer 341 Polarimeter (Waltham, MA).

Antifungal activity assay. All seven compounds were tested against eight food and cash crop fungal pathogens,[24-25] according to published procedure.[26] The compound (10 mg) was dissolved in 0.5 mL of DMSO and then diluted with distilled water containing 0.1% Tween-80 to a concentration of 200 μg/mL. The solution was then diluted to 1.56, 3.12, 6.25, 12.5, 25, 50, and 100 μg/mL. The fungicidal activity was determined as the rate of mycelial growth inhibition.[26] Thiophanate-methyl is a broad-spectrum systemic commercial fungicide. It was used as a positive control to compare the antifungal potencies of the newly synthesized compounds.

Statistics and EC_{50} calculations. Data were presented as the means ± standard deviations (SD) for at least three independent experiments. Statistical significance (***$p < 0.001$, **$p < 0.01$, *$p < 0.05$) was assessed using Student's *t*-test or one-way analysis of variance (ANOVA) coupled with Dunnett's *t*-test.

General synthetic method of target compounds. To a blue solution of CuCl (2.9 mg, 0.03 mmol) in degassed 30% n-$BuNH_2$ solution (2.2 mL) was added aqueous NH_2OH-HCl to discharge the blue color at 0 ^oC. A solution of terminal alkyne (1.50 mmol) in methanol (MeOH) (1.0 mL) was then added. After the resulting mixture was stirred for 1 min, bromohydrocarbon (1.00 mmol) in MeOH (1.0 mL) was added slowly. The reaction mixture was stirred for 30 min before it was quenched with water. The aqueous phase was extracted with dichloromethane. The combined organic phases were washed with saturated brine solution, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure. The residue was purified by silica gel chromatography.

RESULTS AND DISCUSSION

The retrosynthetic analysis of (***S***, ***E***)-**1** and (**3*R***, **10*R***, ***E***)-**2** (Figure 2) led to four different fragments: the terminal alkyne **8** and chiral alkynols (***S***)-**12**, (***R***)-**12**, and **18**. All the chiral alkynols could be generated from a chiral alkynol ester, a classic product synthesized from the asymmetric addition of methyl propiolate to aldehydes with the enantioselectivity up to 98% purity. Terminal alkyne **8** could be synthesized by desiliconization of enyne **7**, which could be accomplished by the Cu-catalyzed cross-coupling reaction.

The synthesis of **8** started from the preparation of the long-chain propargyl alcohol **4** (Scheme 1). Under the effect of *n*-butyllithium, propargyl alcohol reacted with 1-bromooctane to afford **4** with 95% yield. Then, **4** was treated with $LiAlH_4$ to generate enol **5** with 91% yield. Bromohydrocarbon **6** was prepared in 99% yield by the substitution of enol **5** with carbon tetrabromide and triphenylphosphine. The key intermediate **7** was achieved by the cross-coupling of bromohydrocarbon **6** with trimethylsilyacetylene in 83% yield under room temperature. After the desiliconization of enyne **7**, the terminal alkyne **8** was synthesized in 90% yield.

Synthesis of the chiral alkynol (***S***)-**12** was consisted of two parts (Scheme 2). Under the effect of prophenol-triphenylphosphine oxide-zinc complex, the asymmetric addition of methyl propiolate **10** to acrolein **9** was achieved to afford (***S***)-**11** with 65% yield and 99% *ee*.[27-28] The second part was the decarboxylation and substitution of (***S***)-**11**. (***S***)-**11** was treated with LiOH, CuCl, NBS and $AgNO_3$ to afford chiral alkynol (***S***)-**12** in 71% yield. The synthesis of the chiral alkynol (***R***)-**12** was similar to that of (***S***)-**12** with the compound (***S***)-**19** instead of (***R***)-**19**.

The synthesis of the chiral enol **18** started from the preparation of the chiral propargyl alcohol (***R***)-**15** (Scheme 3). Under the same condition of the synthesis of (***R***)-**12**, (***R***)-**15** was

120 achieved in 73% yield. (***R***)-**15** was then treated with $LiAlH_4$ and $AlCl_3$ to afford the bromoenol
121 **16** in 91% yield.[29] The key intermediate **17** was achieved by cross-coupling **16** with
122 trimethylsilyacetylene in 87% yield at room temperature. After the desiliconization of **17**, the
123 terminal alkyne **18** was synthesized in 91% yield.

124 The natural product (***S***, ***E***)-**1** was synthesized by the Cadiot-Chodkiewicz cross-coupling of
125 the terminal alkyne **8** and the alkynol (***S***)-**12** in 87% yield.[30] The natural product (**3*R***, **10*R***, ***E***)-**2**
126 was achieved by the across-coupling of chiral enol **18** and (***R***)-**12** in 87% yield (Scheme 4). After
127 (***S***, ***E***)-**1** and (***3R***, ***10R***, ***E***)-**2** were synthesized, we intended to test the antifungal activity of the
128 new C18 structures having different double bonds and chiral alkynol centers. A new *Z*-
129 configuration terminal alkyne **23** was then synthesized in order to achieve a series of (***S***, ***E***)-**1**
130 analogs.

131 The synthesis of the terminal alkyne **23** started from **4** (Scheme 5). The propargyl alcohol **4**
132 was treated with carbon tetrabromide and triphenylphosphine to generate the bromohydrocarbon
133 **20** in 98% yield. The intermediate **21** was achieved by cross-coupling **20** with
134 trimethylsilyacetylene in 85% yield under nitrogen atmosphere. **22** was selectively reduced by
135 the Raney Nickel in 81% yield. After the desiliconization of the enyne **22**, the terminal alkyne **23**
136 was synthesized in 91% yield.

137 Five isomers of (***S***, ***E***)-**1** were achieved by the same synthetic strategy as (***S***, ***E***)-**1** (Scheme
138 6). (***R***, ***E***)-**1** was synthesized by the Cadiot-Chodkiewicz cross-coupling of **8** and (***R***)-**12** in 85%
139 yield. ***Rac***-(***E***)-**1** was accomplished by the cross-coupling of **8** and ***rac***-**12** in 83% yield. Cross-
140 coupling of **23** with (***R***)-**12**, ***rac***-**12** and (***S***)-**12** afforded in (***R***, ***Z***)-**1**, ***Rac***-(***Z***)-**1** and (***S***, ***Z***)-**1**,
141 respectively, in 87%, 87% and 85% yields.

142 Structural characterization: Table 1 shows ^{1}H NMR spectroscopic data (δ_H in ppm, *J* in

Hz) of, (***S*, *E*)-1** (***R*, *Z*)-1** and (**3*R*, 10*R*, *E*)-2** (400 MHz, $CDCl_3$). (***S***, ***E***)-octadeca-1,9-dien-4,6-diyn-3-ol, (***S***, ***E***)-**1**, $[\alpha]_D^{20}$ = + 11.2 (*c* = 0.10, $CHCl_3$), lit[13] $[\alpha]_D^{20}$ = + 11.2 (*c* = 0.10, $CHCl_3$). The molecular formula is $C_{18}H_{26}O$. HRMS ESI $[M-H]^-$ calcd for $C_{18}H_{25}O^-$ 257.1911, found 257.1913. The ^{1}H-NMR spectrum contained alkenyl proton signals H-1 at δ 5.47 (dt, *J* = 17.0, 1.3 Hz, 1H) and 5.24 (dt, *J* = 10.1, 1.3 Hz, 1H). The HMBC and DEPT spectra showed an oxygenated H-3 proton signal at δ 4.93 (d, *J* = 5.0 Hz, 1H), olefinic protons (H-2, H-9, H-10) at δ 5.94 (ddd, *J* = 17.0, 10.1, 5.3 Hz, 1H), 5.39 – 5.30 (m, 1H), 5.67 (dtt, *J* = 15.3, 6.8, 1.8 Hz, 1H), and alkyl protons (H-17, H-16, H-15, H-14, H-13, H-12, H-11, and H-8) at δ 1.37 – 1.32 (m, 2H), 1.26 (s, 10H), 2.03 – 1.98 (m, 2H), 3.00 (dt, *J* = 5.7, 1.4 Hz, 2H), and a methyl proton (H-12) at 0.88 (t, *J* = 6.8 Hz, 3H). The δ 2.10 (d, *J* = 5.6 Hz, 1H) showed no correlation with any carbon in the HSQC spectrum; it was assigned to the hydroxy group (H-19). The ^{13}C-NMR spectrum contained four olefinic carbon signals (C-2, C-10, C-9, and C-1) at δ 136.1, 133.6, 122.0, and 117.0, one oxygenated sp^3 carbon signal (C-3) at δ 63.5, four alkyne carbon signals (C-4, C-5, C-6, and C-7) at δ 79.6, 65.7, 71.2, 74.5, seven alkyl carbon signals (C-8, C-17, C-16, C-15, C-14, C-13, C-12, and C-11) at 22.4, 31.9, 29.4, 29.2, 29.2, 29.1, 22.6, and 32.3, and one methyl carbon signal (C-18) at 14.1. The NOESY-NMR spectrum correlation between H-8 (δ 3.00 (dt, *J* = 5.6, 1.4 Hz, 2H)) and H-11 (δ 2.03 – 1.98 (m, 2H)) revealed that C-9 and C-10 were trans-configured.

(***R***, ***Z***)-Octadeca-1,9-dien-4,6-diyn-3-ol, (***R***, ***Z***)-**1**, $[\alpha]_D^{20}$ = - 30.9 (*c* = 0.10, $CHCl_3$). The molecular formula is $C_{18}H_{26}O$. HRMS ESI $[M-H]^-$ calcd for $C_{18}H_{25}O^-$ 257.1911, found 257.1912. The ^{1}H-NMR spectrum contained alkenyl proton signals (H-1) at δ 5.49 - 5.44 (m, 1H), 5.41 – 5.34 (m, 1H). The HMBC and DEPT spectra showed an oxygenated (H-3) proton signal at δ 4.91 (dt, *J* = 5.4, 1.3 Hz, 1H), olefinic proton signals (H-2, H-10, H-9) located separately at δ 5.94

(ddd, J = 17.1, 10.2, 5.4 Hz, 1H), 5.51 (dt, J = 9.0, 1.7 Hz, 1H), 5.24 (dt, J = 10.2, 1.2 Hz, 1H), and alkyl proton signals (H-8, H-11, H-12, H-13, H-14, H-15 ,H-16, and H-17) at δ, 2.02 (qd, J = 7.3, 1.5 Hz, 2H), 3.03 (dt, J = 7.0, 1.1 Hz, 2H), 1.35 (q, J = 7.0 Hz, 2H), 1.27 (s, 10H), and a methyl proton signal (H-18) at 0.87 (t, J = 6.7 Hz, 3H). The δ 2.02 signal (qd, J = 7.3, 1.5 Hz, 1H) showed no correlation with any carbon in the HSQC spectrum; it was assigned to the hydroxy group (H-19). The ^{13}C-NMR spectrum contained four olefinic carbon signals (C-2, C-10, C-9, and C-1) at δ 136.1, 133.1, 121.9, and 117.0, and one oxygenated sp3 carbon signal (C-3) at δ 63.5, four alkyne carbon signals (C-4, C-5, C-6, and C-7) at δ 80.3, 64.0, 71.3, and 74.2, seven alkyl carbon signals (C-8, C-11, C-12, C-13, C-14, C-15, C-16, and C-17) at δ 17.7, 27.2, 29.2, 29.2, 29.3, 29.4 22.7, and 31.8, and one methyl carbon signal (C-18) at 14.1. The NOESY-NMR spectrum correlation between H-11 (δ 3.03 (dt, J = 7.0, 1.1 Hz, 2H)) and H-8 (2.02 (qd, J = 7.3, 1.5 Hz, 3H)) revealed that C-9 and C-10 were cis-configured.

(**3*R***, **10*R***, ***E***)-Octadeca-1,8-dien-4,6-diyne-3,10-diol, (**3*R***, **10*R***, ***E***)**-2** was obtained as yellow oil with $[\alpha]_D^{20}$ = - 38.4 (c = 0.70, $CHCl_3$), lit[13] $[\alpha]_D^{20}$ = - 38.5 (c = 0.70, $CHCl_3$). The molecular formula is $C_{18}H_{26}O_2$. HRMS ESI $[M-H]^-$ calcd for $C_{18}H_{25}O_2^-$ 273.1860, found 273.1863. The ^{1}H-NMR spectrum contained alkenyl proton signals (H-1) at δ 5.47 (dd, J = 17.1, 1.4 Hz, 1H) and 5.25 (dd, J = 10.1, 1.4 Hz, 1H). The HMBC and DEPT spectra showed two oxygenated proton signals (H-3 and H-10) located separately at δ 4.97 (d, J = 5.0 Hz, 1H), 4.18 (q, J = 6.2 Hz, 1H), and three olefinic proton signals (H-9, H-2, H-8) located separately at δ 6.33 (ddd, J = 15.9, 5.7, 1.5 Hz, 1H), 5.99 – 5.89 (m, 1H), and 5.76 (d, J = 15.9 Hz, 1H), and alkyl proton signals (H-11, H-12, H-13, H-14, H-15, H-16, and H-17) at δ 1.52 (t, J = 6.7 Hz, 2H) and δ 1.27 (q, J = 5.4, 4.5 Hz, 12H), and one methyl proton signal (H-18) at 0.87 (t, J = 6.8 Hz, 3H). The signals at δ 2.52 (d, J = 6.2 Hz, 1H) and 1.92 (s, 1H) showed no correlation with any carbon in the HSQC

189 spectrum; it was assigned to the hydroxy group (H-19 and H-20). The ^{13}C-NMR spectrum
190 contained four olefinic carbon signals (C-9, C-2, C-1, and C-8) at δ 149.8, 135.9, 117.2, 108.1,
191 and two oxygenated sp^3 carbon signals (C-3, C-10) at δ 63.5 and 72.0, four alkyne carbon signals
192 (C-4, C-5, C-6, and C-7) at δ 80.4, 70.9, 73.5, and 77.5, seven alkyl carbon signals (C-11, C-12,
193 C-13, C-14, C-15, C-16, and C-17) at δ 22.6, 31.8, 29.5, 29.4, 29.2, 25.2, and 36.8, and one
194 methyl carbon signal (C-18) at δ 14.1. The NOESY-NMR spectrum correlation between H-9 (δ
195 6.33 (ddd, J = 15.9, 5.7, 1.5 Hz, 1H)) and H-11 (δ 1.55 – 1.49 (m, 2H)) revealed that H-9 and H-
196 11 were trans-configured.

197 (***S***, ***E***)-**1** and (**3*R***, **10*R***, ***E***)-**2** showed good antifungal activities (Table 2). The antifungal
198 activities of (***R***, ***E***)-**1**, ***rac***-(***E***)-**1**, (***S***, ***Z***)-**1**, (***R***, ***Z***)-**1** and ***rac***-(***Z***)-**1** were also shown in Table 2. It
199 is very interesting that the newly synthesized polyacetylene alcohols show half-maximum
200 effective concentrations (EC_{50}) between 8 and 425 μg/mL. Such potencies are in a similar range
201 as those of thiophanate-methyl (EC_{50}, 3 and 408 μg/mL), whereas the anti-fungal spectra of the
202 polyacetylene alcohols largely differ among themselves and from those of thiophanate-methyl.
203 Thiophanate-methyl is the most potent (3 μg/mL) against *M. fructigena* among the eight fungal
204 pathogens tested. Among the seven tested pure isomers or racemic mixtures and eight fungal
205 species, ***rac***-(***E***)-**1** was the most potent against *B. sorokiniana*.

206 The EC_{50} value of (***S***, ***E***)-**1** against *B. sorokiniana* was 36-, 3- and 8-times smaller (i.e.,
207 more potent) than that of (***R***, ***E***)-**1**, (***S***, ***Z***)-**1** and (***3R***, ***10R***, ***E***)-**2**, respectively. The EC_{50} value of
208 (***S***, ***E***)-**1** against *C. gloeosporioides* was 7-, 2- and 2-times smaller than that of (***R***, ***E***)-**1**, (***S***, ***Z***)-**1**
209 and (***3R***, ***10R***, ***E***)-**2**, respectively. The EC_{50} value of (***S***, ***E***)-**1** against *Phytophthora capsici* was
210 2-, 5- and 4-times smaller than (***R***, ***E***)-**1**, (***S***, ***Z***)-**1** and (***3R***, ***10R***, ***E***)-**2**, respectively. The EC_{50} of
211 (***S***, ***E***)-**1** against *F. graminearum* was 3-times smaller than both ***rac***-(***E***)-**1**, and (***R***, ***Z***)-**1**,

212 respectively. The EC_{50} of (***S***, ***E***)-**1** against *A. kikuchiana* Tanaka was 3-, 2- and 3-times smaller
213 than ***rac***-(***E***)-**1**, ***rac***-(***Z***)-**1**, and (***3R***, ***10R***, ***E***)-**2**, respectively. The EC_{50} of (***3R***, ***10R***, ***E***)-**2** against
214 *Rhizoctonia cerealis* was 8-, 4-, and 3-times smaller than (***S***, ***E***)-**1**, (***S***, ***Z***)-**1**, and ***rac***-(***E***)-**1**,
215 respectively. The EC_{50} of (***S***, ***E***)-**1** against *F. pseudograminearum* was similar to that of (***3R***,
216 ***10R***, ***E***)-**2**, but 4-times smaller than ***rac***-(***E***)-**1** and (***R***, ***Z***)-**1**. The EC_{50} of (***S***, ***Z***)-**1** against *M.*
217 *fructigena* was 7-, 6- and 3-times smaller than (***S***, ***E***)-**1**, ***rac***-(***Z***)-**1** and (***3R***, ***10R***, ***E***)-**2**,
218 respectively.

219 The results may indicate that the fungicidal potency is related to the chiral center
220 configurations and geometric isomerism. The bioassay results of the eight fungal species suggest
221 that (***S***, ***E***)-**1** and (***R***, ***E***)-**1**, (***S***, ***Z***)-**1** and (***R***, ***Z***)-**1** are mostly antagonistic. However, (***S***, ***E***)-**1** and
222 (***R***, ***E***)-**1**, (***S***, ***Z***)-**1** and (***R***, ***Z***)-**1** had low activity against *B. sorokiniana*. (***S***, ***E***)-**1** and (***R***, ***E***)-**1** had
223 low activity against *P. capsica* and *M. fructigena*, and were synergistic against *Rhizoctonia*
224 *cerealis*. (***S***, ***Z***)-**1** and (***R***, ***Z***)-**1** were synergistic against *P. capsica*.

225 Compounds having the ***S*** optical configuration and the ***E*** configuration seem to have greater
226 antifungal potential. The spatial structure and activity information may lay a good foundation for
227 the study of the structure-activity relationship of polyacetylenes in the future.

228 In conclusion, an efficient and highly enantioselective general method (> 98% *ee*, 32-56%
229 yield) was established to synthesize C18 polyacetylenes (***S***, ***E***)-octadeca-1,9-dien-4,6-diyn-3-ol,
230 (**3*R***, **10*R***, ***E***)-octadeca-1,8-dien- 4,6-diyne-3,10-diol and their analogs for the first time. The
231 configuration of the double bond and the chiral alkynol centers have shown varying selectivity
232 against the tested fungi. The EC_{50} values ranged from 8 to 425 μg/mL. The EC_{50} values of (***S***,
233 ***E***)-**1**, ***rac***-(***E***)-**1**, (***S***, ***Z***)-**1**, ***rac***-(***Z***)-**1**, and (**3*R***, **10*R***, ***E***)-**2** against *B. sorokiniana* were 9, 8, 26, 24,
234 and 74 μg/mL, respectively, which provide useful information to the structural optimization of

new polyacetylene fungicide candidates.

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SUPPORTING INFORMATION

^{1}H and ^{13}C NMR, 2D NMR spectra, HPLC chromatograms of the products. This material is available free of charge via the Internet at http://pubs.acs.org.

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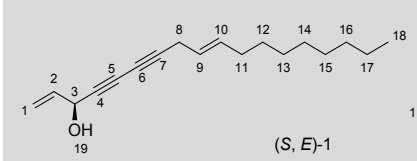
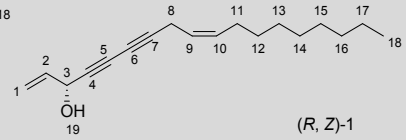
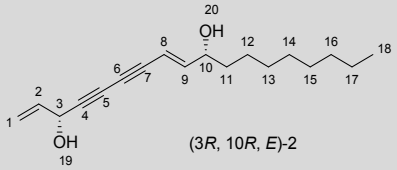
Table 1. ^{1}H NMR spectroscopic data (δ_H in ppm, J in Hz) of (***S*, *E*)-1**, (***R*, *Z*)-1** and (**3*R*, 10*R*, *E*)-2**. (400 MHz, $CDCl_3$)



(*S*, *E*)-1



(*R*, *Z*)-1



(3*R*, 10*R*, *E*)-2

No[a]	δ_C	δ_H (J in Hz)	HMBC	δ_C	δ_H (J in Hz)	HMBC	δ_C	δ_H (J in Hz)	HMBC
1	117.0, CH_2	H_a 5.47 dt, (17.0, 1.3) H_b 5.24 dt, (10.1, 1.3)	H_a 2, 3, 4, 5 H_b 2, 3, 4	117.0, CH_2	H_a 5.49 – 5.44, m H_b 5.41 – 5.34, m	H_a 2, 3 H_b 2, 3	117.2 CH_2	H_a 5.47, dd (17.1, 1.4) H_b 5.25, dd (10.1, 1.4)	H_a 2, 3, 4 H_b 3, 4
2	136.1, CH	5.94 ddd, (17.0, 10.1, 5.3)	3, 4	136.1, CH	5.94, ddd (17.1, 10.2)	3, 4, 5	135.9, CH	5.99 – 5.89, m	1, 3, 4
3	63.5, CH	4.93 d, (5.0)	1, 2, 4, 5	63.5, CH	4.91, dt (5.4, 1.3)	1, 2, 4, 5	63.5, CH	4.97, d (5.0)	1, 2, 4,
4	79.6, C			80.3, C			80.4, C		
5	65.7, C			64.0, C			70.9, C		
6	71.2, C			71.3, C			73.5, C		
7	74.5, C			74.2, C			77.5, C		
8	22.4, CH_2	3.00 dt, (5.6, 1.4)	3, 4, 5, 6, 7, 9, 10, 11	17.7, CH_2	2.02, qd (7.3, 1.5)	6, 7, 9, 10	108.1, CH	5.76, d (15.9)	6, 7, 9, 10
9	122.0, CH	5.39 – 5.30, m	7, 8, 10, 11	121.9, CH	5.24, dt (10.2, 1.2)	7, 8, 10, 11	149.8, CH	6.33, ddd (15.9, 5.7, 1.5)	7, 8, 10, 11
10	133.6, CH	5.67 dtt, (15.3, 6.8, 1.8)	7, 9, 11, 12	133.1, CH	5.51, dt (9.0, 1.7)	7, 8, 9, 11,12	72.0, CH	4.18, q (6.2)	8, 9, 11
11	32.3, CH_2	2.03 – 1.98, m	11, 12, 13, 14, 16, 17	27.2, CH_2	3.03, dt (7.0, 1.1)	9, 10, 12, 13, 14, 15	36.8, CH_2	1.55 – 1.49, m	8, 9, 10, 12, 13, 14, 15, 16
12	22.6, CH_2	1.26, s	11, 12, 13, 14, 16, 17	29.2, CH_2	1.27, s	9, 10, 11, 13, 14, 15	25.2, CH_2	1.27, q (5.4, 4.5)	11, 13, 15, 16, 17
13	29.1, CH_2	1.26, s	11, 12, 14, 15, 16, 17	29.2, CH_2	1.27, s	11, 12, 14, 15, 16	29.2, CH_2	1.27, q (5.4, 4.5)	10, 11, 12, 14, 15, 16, 17

14	29.2, CH_2	1.26, s	11, 12, 13, 15, 16, 17	29.3, CH_2	1.27, s	11, 12, 13, 15, 16, 17	29.4, CH_2	1.27, q (5.4, 4.5)	11, 12, 13, 15, 16, 17
15	29.2, CH_2	1.26, s	11, 12, 13, 14, 16, 17	29.4, CH_2	1.27, s	12, 13, 14, 16, 17	29.5, CH_2	1.27, q (5.4, 4.5)	11, 13, 14, 16, 17, 18
16	29.4, CH_2	1.26, s	12, 13, 14, 15, 17, 18	22.7, CH_2	1.27, s	12, 13, 14, 15, 17, 18	31.8, CH_2	1.27, q (5.4, 4.5)	11, 12, 13, 14, 15, 17, 18
17	31.9, CH_2	1.37 – 1.32, m	12, 13, 14, 15, 16, 18	31.8, CH_2	1.35, q (7.0)	12, 13, 14, 15, 16, 18	22.6, CH_2	1.27, q (5.4, 4.5)	11, 12, 13, 14, 15, 16, 18
18	14.1, CH_3	0.88 t, (6.8)	9, 13, 14, 15, 16, 17	14.1, CH_3	0.87, t (6.7).	13, 14, 15, 16, 17	14.1, CH_3	0.87, t (6.8).	13, 14, 15, 16, 17
19		2.10 d, (5.6)			2.02, qd (7.3, 1.5)			2.52, d (6.2)	
20								1.92, s	

[a] Carbon position.

Table 2. Antifungal activity of (***3R***, ***10R***, ***E***)-**2** and all the isomers of natural product **1**.

Fungus	Compound	Toxicity regression equation	R^2	EC_{50} (μg/mL)
B. sorokiniana	(***S***, ***E***)-**1**	y=6.87+0.61x	0.983	9
	(***R***, ***E***)-**1**	y=6.23+0.83x	0.956	330
	rac-(***E***)-**1**	y=6.47+0.47x	0.966	8
	(***S***, ***Z***)-**1**	y=6.55+0.60x	0.984	26
	(***R***, ***Z***)-**1**	y=6.28+0.64x	0.968	100
	rac-(***Z***)-**1**	y=6.47+0.56x	0.978	24
	(***3R***, ***10R***, ***E***)-**2**	y=7.94+1.38x	0.986	74
	Thiophanate-methyl	y=5.50+0.36x	0.976	408
C. gloeosporioides	(***S***, ***E***)-**1**	y=6.75+0.75x	0.991	46
	(***R***, ***E***)-**1**	y=6.22+0.84x	0.982	353
	rac-(***E***)-**1**	y=6.69+0.90x	0.989	133
	(***S***, ***Z***)-**1**	y=7.54+1.21x	0.970	80
	(***R***, ***Z***)-**1**	y=6.79+0.87x	0.969	88
	rac-(***Z***)-**1**	y=6.90+0.96x	0.991	105
	(***3R***, ***10R***, ***E***)-**2**	y=6.32+0.63x	0.960	80
	Thiophanate-methyl	y=6.67+0.97x	0.993	190
Phytophthora capsica	(***S***, ***E***)-**1**	y=6.88+0.87	0.991	69
	(***R***, ***E***)-**1**	y=5.85+0.48x	0.993	169
	rac-(***E***)-**1**	y=6.35+0.63x	0.997	72
	(***S***, ***Z***)-**1**	y=5.79+0.54x	0.994	344
	(***R***, ***Z***)-**1**	y=6.33+0.97x	0.979	425
	rac-(***Z***)-**1**	y=6.61+0.85x	0.973	128
	(***3R***, ***10R***, ***E***)-**2**	y=6.47+0.94x	0.975	273
	Thiophanate-methyl	y=7.13+0.71x	0.981	10
F. graminearum	(***S***, ***E***)-**1**	y=6.52+0.71x	0.951	72
	(***R***, ***E***)-**1**	y=6.05+0.49x	0.966	72
	rac-(***E***)-**1**	y=6.60+0.96x	0.969	216
	(***S***, ***Z***)-**1**	y=6.19+0.59x	0.984	96
	(***R***, ***Z***)-**1**	y=6.53+0.94x	0.969	236
	rac-(***Z***)-**1**	y=5.77+0.41x	0.980	132
	(***3R***, ***10R***, ***E***)-**2**	y=6.66+0.76x	0.964	66
	Thiophanate-methyl	y=8.24+1.20x	0.963	200
A. kikuchiana Tanaka	(***S***, ***E***)-**1**	y=6.75+0.73x	0.992	41

	(***R***, ***E***)-**1**	y=6.17+0.51x	0.988	51
	rac-(***E***)-**1**	y=6.08+0.57x	0.982	127
	(***S***, ***Z***)-**1**	y=6.39+0.64x	0.971	67
	(***R***, ***Z***)-**1**	y=6.76+0.76x	0.984	48
	rac-(***Z***)-**1**	y=6.45+0.73x	0.987	103
	(***3R***, ***10R***, ***E***)-**2**	y=5.89+0.475x	0.994	134
	Thiophanate-methyl	y=9.90+2.01x	0.984	37
Rhizoctonia cerealis	(***S***, ***E***)-**1**	y=5.73+0.48x	0.968	301
	(***R***, ***E***)-**1**	y=5.72+0.39x	0.989	143
	rac-(***E***)-**1**	y=6.44+0.74x	0.981	113
	(***S***, ***Z***)-**1**	y=6.38+0.72x	0.978	121
	(***R***, ***Z***)-**1**	y=6.33+0.65x	0.990	90
	rac-(***Z***)-**1**	y=6.53+0.77x	0.992	103
	(***3R***, ***10R***, ***E***)-**2**	y=6.58+0.65x	0.987	37
	Thiophanate-methyl	y=6.39+0.67x	0.987	84
F. pseudograminearum	(***S***, ***E***)-**1**	y=6.12+0.46x	0.885	37
	(***R, E***)-**1**	y=6.86+0.83x	0.978	57
	rac-(***E***)-**1**	y=6.11+0.64x	0.990	184
	(***S***, ***Z***)-**1**	y=6.90+0.83x	0.989	51
	(***R***, ***Z***)-**1**	y=6.14+0.63	0.987	155
	rac-(***Z***)-**1**	y=6.00+0.53x	0.986	130
	(***3R***, ***10R***, ***E***)-**2**	y=6.98+0.84x	0.968	44
	Thiophanate-methyl	y=8.26+1.20x	0.942	19
M. fructigena	(***S***, ***E***)-**1**	y=5.73+0.50x	0.969	347
	(***R***, ***E***)-**1**	y=6.38+0.66x	0.994	81
	rac-(***E***)-**1**	y=6.46+0.70x	0.985	82
	(***S***, ***Z***)-**1**	y=6.33+0.59x	0.993	56
	(***R***, ***Z***)-**1**	y=7.01+0.99x	0.971	93
	rac-(***Z***)-**1**	y=6.10+0.74x	0.993	326
	(***3R***, ***10R***, ***E***)-**2**	y=6.06+0.601x	0.948	172
	Thiophanate-methyl	y=7.20+0.610x	0.949	3

Figure and scheme captions

Figure 1. Structures of strobilurin's family and polyacetylenes.

Figure 2. Retrosynthetic analysis of (***S***, ***E***)-**1** and (**3*R***, **10*R***, ***E***)-**2**.

Scheme 1. Synthesis of the terminal alkyne **8**.

Scheme 2. Synthesis of (***S***)-**12** & (***R***)-**12**.

Scheme 3. Synthesis of (***R***, ***E***)-tridec-3-en-1-yn-5-ol **18**.

Scheme 4. Synthesis of (***S***, ***E***)-**1** and (**3*R***, **10*R***, ***E***)-**2**.

Scheme 5. Synthesis of the terminal alkyne **23**.

Scheme 6. Synthesis of (***S***, ***E***)-**1** analogs.

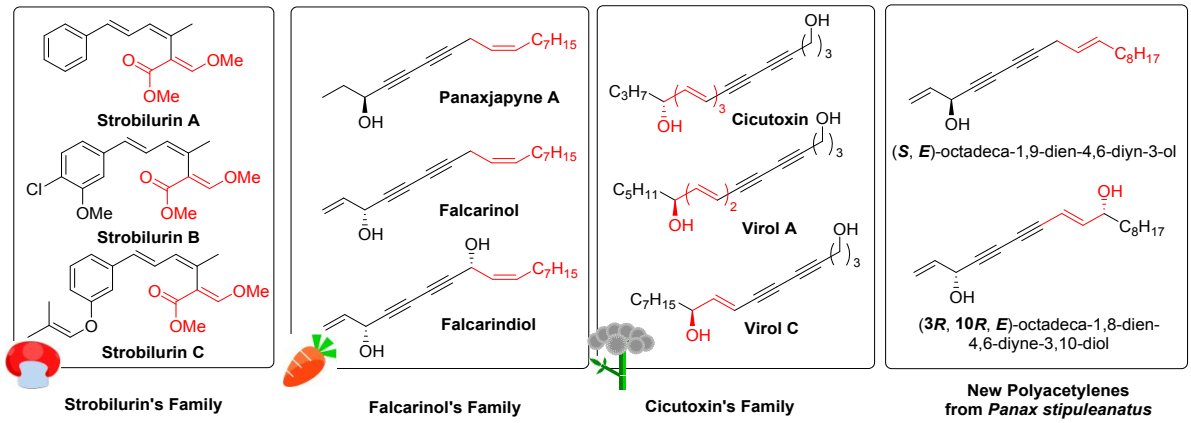


Figure 1. Structures of strobilurin's family and polyacetylenes.

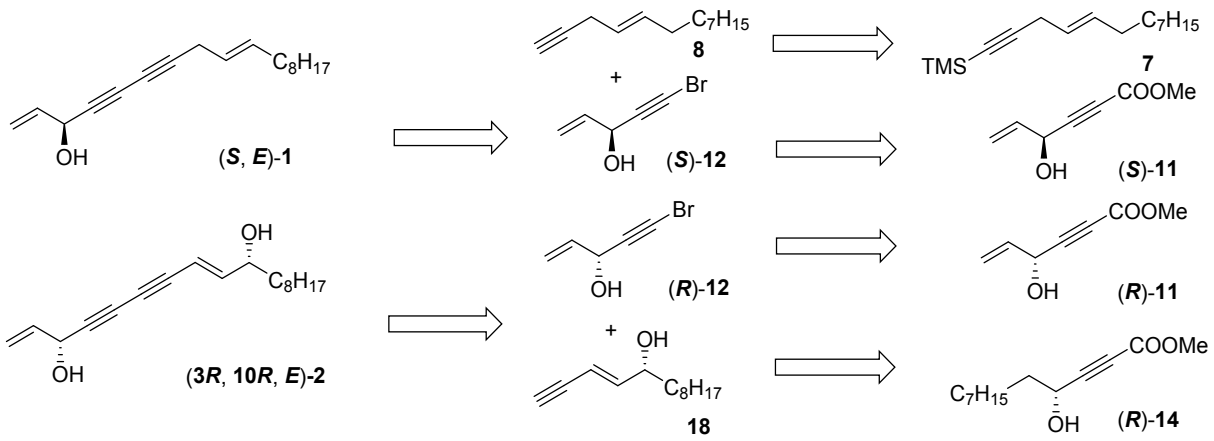
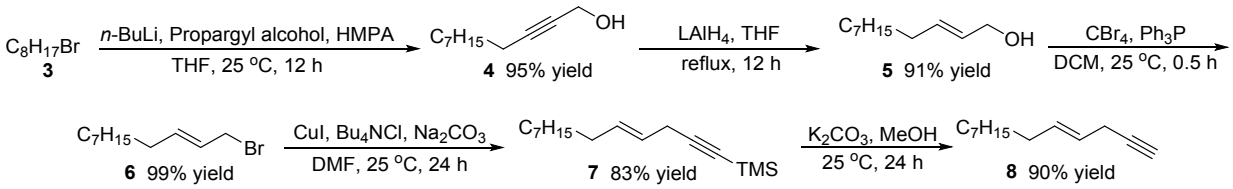
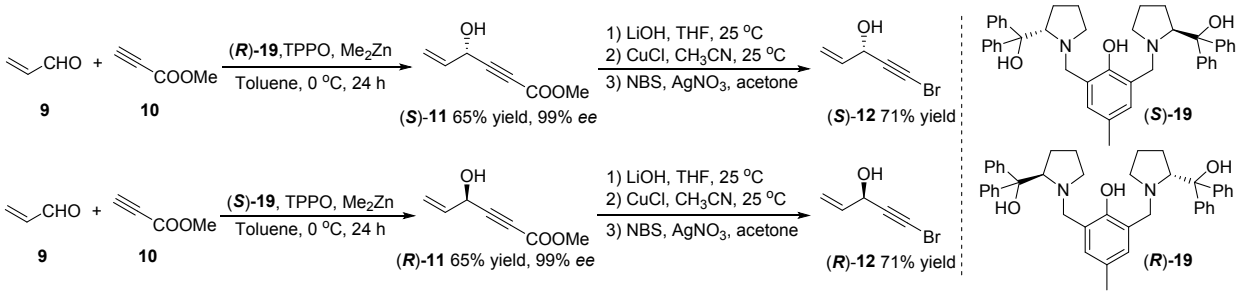


Figure 2. Retrosynthetic analysis of (***S***, ***E***)-**1** and (**3*R***, **10*R***, ***E***)-**2**.

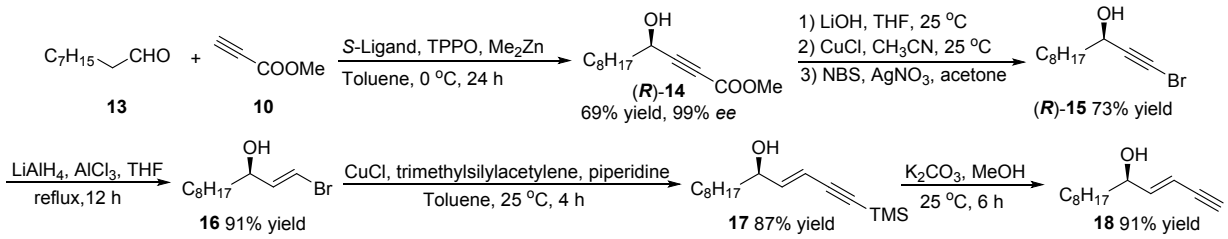
Scheme 1. Synthesis of the terminal alkyne 8



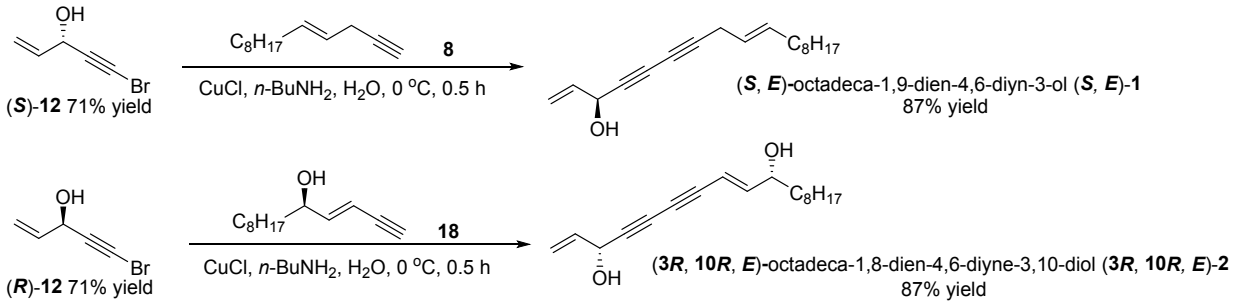
Scheme 2. Synthesis of (*S*)-12 and (*R*)-12.



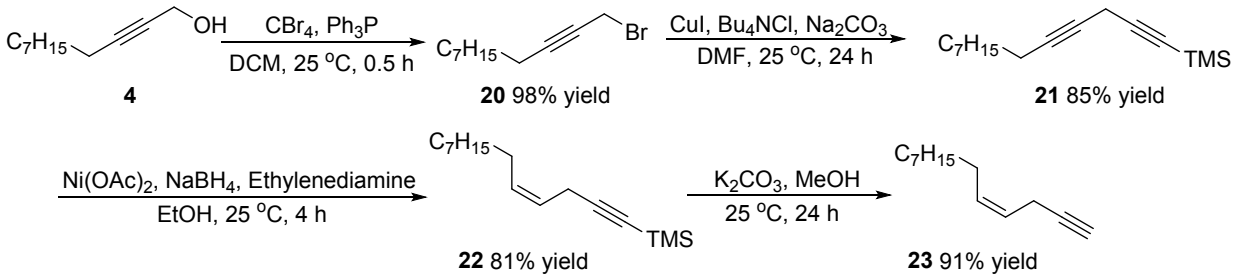
Scheme 3. Synthesis of (*R*, *E*)-tridec-3-en-1-yn-5-ol, 18.



Scheme 4. Synthesis of (*S*, *E*)-1 and (3*R*, 10*R*, *E*)-2.



Scheme 5. Synthesis of the terminal alkyne 23.



4

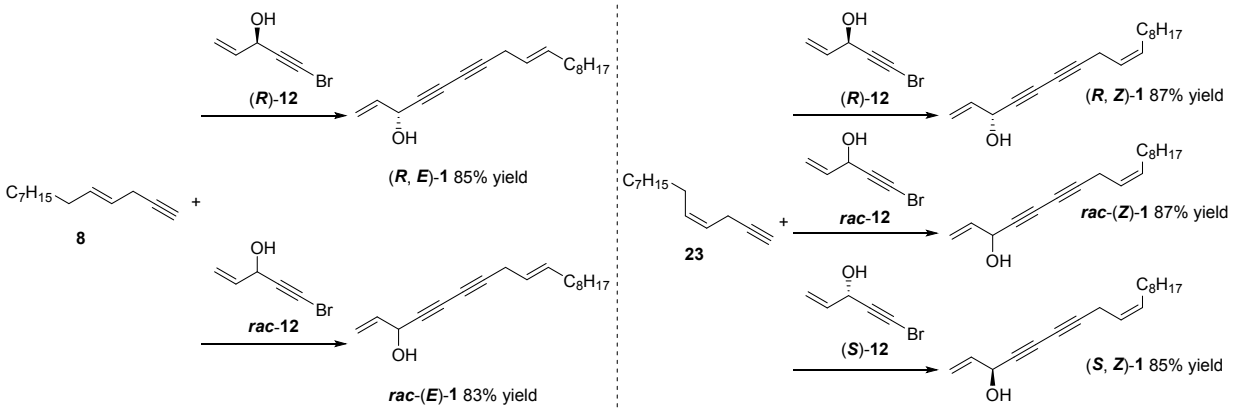
20 98% yield

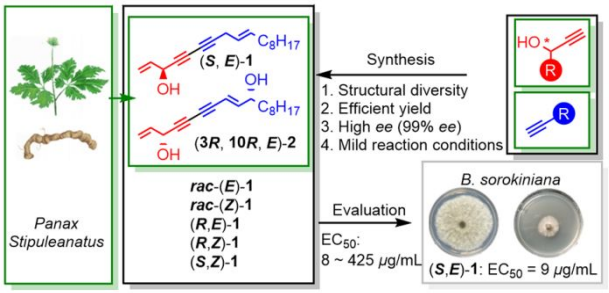
21 85% yield

22 81% yield

23 91% yield

Scheme 6. Synthesis of (*S*, *E*)-1 analogs.





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