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Mallophenins A-E, Antibacterial Phenolic Derivatives from the Fruits of *Mallotus philippensis*

Abstract

The chromatographic separation of the components of the acetone extract of *Mallotus philippensis* fruits yielded five new phenolic compounds including two chalcones, 1 and 3, a functionalized phloroglucinol, 2, two flavanones, 4 and 5, and six known compounds. The structures of 1-5 were confirmed by NMR and mass analyses. Racemic compounds 1 and 2 were separated by chiral-phase HPLC, and the absolute configuration of (+)-1 was confirmed by X-ray diffraction studies and ECD spectroscopic data. The configurations of the enantiomers of 2 were defined by comparison of its ECD data with those of (+)-1. Compounds 6 and 7 exhibited significant antibacterial activities, with MIC values ranging from 3.8 to 15.5 μM .

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Mallophenins A-E, Antibacterial Phenolic

Derivatives from the Fruits of *Mallotus philippensis*

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ABSTRACT: The chromatographic separation of the components of the acetone extract of *Mallotus philippensis* fruits yielded five new phenolic compounds including two chalcones **1** and **3**, a functionalized phloroglucinol **2**, two flavanones **4** and **5** and six known compounds. The structures of **1–5** were confirmed by NMR and mass analyses. Racemic compounds **1** and **2** were separated by chiral-phase HPLC and the absolute configuration of (+)-**1** was confirmed by X-ray diffraction studies and ECD spectroscopic data. The configurations of the enantiomers of **2** were defined by comparison of its ECD data with those of (+)-**1**. Compounds **6** and **7** exhibited significant antibacterial activities with MIC values ranging from 3.8 to 15.5 μ M.

Mallotus philippensis (Euphorbiaceae), known locally in Thai as “Kham Saet”, is a perennial shrub or small tree that is widely distributed from East to Southeast Asia, Indomalaysia to New Caledonia, and in Australia.^{1,2} Glandular hairs of the fruits are covered with short soft curved reddish bristles which are a source of rottlerin.³ This plant has long been used for its anthelmintic and purgative properties.^{4,5} In Northern Thailand, the fruits and bark have been used in traditional medicine and as a dye.² Several types of bioactive compounds have been isolated from the fruits which have displayed a range of pharmacological activities including anti-allergic,⁶ anti-inflammatory,⁶ antifungal,⁷ and antimycobacterial activities.^{8,9} Rottlerin, a red powder from the fruits, exhibits an inhibitory effect towards protein kinase C.¹⁰ As part of a search for bioactive compounds from Thai medicinal plants, the acetone extract of *M. philippensis* fruits was screened for antibacterial activity and afforded two new chalcones **1** and **3**, a new functionalized phloroglucinol **2**, two new flavanones **4** and **5**, and six known flavonoids (Chart 1). The known compounds were identified as rottlerin (**6**),¹¹ 4'-hydroxyrottlerin (**7**),⁷ isorottlerin (**8**),¹¹ 4'-hydroxyisorottlerin (**9**),¹¹ isoallorottlerin (**10**),¹¹ and mallotophilippen F (**11**)⁸. Some isolates were also tested for their antibacterial activities.

RESULTS AND DISCUSSION

Compound **1**, racemic orange crystals, had the formula $C_{30}H_{34}O_4$, as indicated by HRESIMS (m/z 458.2452, $[M]^+$) and ^{13}C NMR data, consistent with an index of hydrogen deficiency of 14. The UV absorption bands [λ_{max} 236, 307, and 351 nm] were attributed to conjugated chromophores and its IR spectrum showed absorption bands for a hydroxy (3327 cm^{-1}), and conjugated hydrogen bonded carbonyl (1691 cm^{-1}) groups. Its 1H and ^{13}C NMR spectroscopic data (Table 1) indicated the presence of resonances for a hydrogen-bonded hydroxy group [δ_H 14.39 (1H, s)], a 3,3-dimethylallyl group [δ_H 5.21 (1H, br t, $J = 7.0\text{ Hz}$), 3.25 (2H, m),

1.79 (3H, s), 1.67 (3H, s); δ_{C} 123.2, 21.3, 17.9, 25.8, 130.9] and an (*E*)-cinnamoyl unit [δ_{H} 8.12 and 7.77 (each 1H, d, $J = 15.6$ Hz), 7.38-7.60 (5H, m); δ_{C} 128.0, 141.4, 128.9 ($\times 2$), 128.8, 128.3 ($\times 2$)]. The resonances at δ_{H} 2.84 (1H, brs, H-7), 2.25 (1H, dt, $J = 12.8, 3.5$ Hz, H-8a), 2.04 (1H, m, H-13), 1.89 (1H, dd, $J = 12.8, 1.1$ Hz, H-8b), 1.85 (1H, m, H-11a), 1.46 (1H, m, H-11b), 1.33 (1H, m, H-12a) and 0.92 (1H, m, H-12b) indicated a sequence of aliphatic methine and methylene coupled protons of a CH₂–CH–CH–CH₂–CH₂ backbone from analysis of the COSY spectrum (Figure 1). Three methyl singlets [δ_{H} 1.56 (Me-16), 1.53 (Me-10) and 1.08 (Me-15)] were also observed. The 3,3-dimethylallyl and cinnamoyl moieties were linked to C-3 and C-1, respectively, based on the correlations in the HMBC data of OH-2 to C-1 (δ_{C} 106.5), C-2 (δ_{C} 163.8), and C-3 (δ_{C} 11.5), of H-18 to C-1, and of H-1' to C-2, C-3, and C-4 (δ_{C} 161.1) (Figure 1). The key HMBC correlation between Me-10 and C-8 (δ_{C} 35.3), C-9 (δ_{C} 29.1), and C-11 (δ_{C} 37.6) indicated that Me-10 was attached to C-9. The Me-15 and Me-16 groups were located at C-14 based on cross-peaks in the HMBC data of both methyl groups to C-13 (δ_{C} 46.0) and C-14 (δ_{C} 84.9). From these HMBC and COSY correlations, a cyclohexane moiety was attached through C-9 and C-13 of the two 3,4-dihydro-2H-pyran moieties. Cross-peaks of H-7/H-8a, H-13, and Me-16, and of H-8a/H-8b and Me-10 (Figures 2 and S7, Supporting Information) in the NOESY spectrum indicated that these protons were cofacial. The *E*-configuration of the $\Delta^{18(19)}$ double bond was defined by the magnitude of $J_{18,19}$ (15.6 Hz). Additionally, resolution of compound **1** using semi-preparative chiral-phase HPLC (Figure S9, Supporting Information) afforded the two enantiomers of (+)-**1** (t_{R} 12.5 min), $[\alpha]_{\text{D}}^{24} +28.4$ (c 0.03, CHCl₃) and (–)-**1** (t_{R} 14 min), $[\alpha]_{\text{D}}^{24} -24.6$ (c 0.03, CHCl₃). A single crystal was obtained and the (7*S*,9*S*,13*R*) configuration of enantiomer (+)-**1** (Figure 3) was defined by X-ray diffraction data. The ECD curve of (+)-**1** gave positive Cotton effects at 245 and 304 nm and negative Cotton effects at 224 and 363 nm, whereas (–)-**1** had opposite Cotton effects

at the aforementioned wavelengths (Figure 4). Therefore, the structures of (+)-mallophenin A [(+)-**1**] and (–)-mallophenin A [(–)-**1**] were assigned as shown in Chart 1.

Compound **2**, a racemic pale yellow solid, was assigned a molecular formula, C₂₃H₃₀O₄ from the HRESIMS ion at m/z 371.2216 [M + H]⁺ (calcd for 371.2222) and ¹³C NMR data. The UV absorptions [λ_{max} 217, 302, and 354 nm] were indicative of a phenyl ring with a carbonyl group and the IR absorption bands implied the stretching bands of hydroxy (3282 cm^{–1}) and hydrogen bonded carbonyl groups (1713 cm^{–1}). The NMR data of **2** (Table 1) were comparable to those of **1**. The major difference was that the cinnamoyl moiety of **1** was replaced by an acetyl unit, which was evident from resonances at δ_{H} 2.61 (3H, s)/ δ_{C} 32.7 and 202.8. The position of this substituent was determined from HMBC analysis (Figures 2 and S15, Supporting Information) from Me-18 to C-1 (δ_{C} 106.1) and C-17 (δ_{C} 202.8). Compound **2** was also resolved by chiral-phase HPLC (Figure S18, Supporting Information) to yield (+)-**2** [t_{R} 13.0 min, $[\alpha]_{\text{D}}^{24}$ +17.4 (c 0.02, CHCl₃) and (–)-**2** [t_{R} 13.5 min, $[\alpha]_{\text{D}}^{24}$ –11.8 (c 0.02, CHCl₃)]. The absolute configurations of (7*S*,9*S*,13*R*) for (+)-**2** and (7*R*,9*R*,13*S*) for (–)-**2** were defined by comparing its ECD spectra (Figure 4) with those of (+)-**1** and (–)-**1**, respectively. Thus, the structures of (+)-mallophenin B [(+)-**2**] and (–)-mallophenin B [(–)-**2**] were defined as shown in Chart 1.

Compound **3**, a pale yellow solid, was assigned a molecular formula, C₃₀H₃₄O₄ based on its HRTOFESIMS ([M + H]⁺ at m/z 459.2505) and ¹³C NMR data. Its UV and IR spectra also resembled those of **1**. The NMR spectroscopic data of **3** suggested high structural similarities compared to **1** (Table 1, Figures S19 and S20, Supporting Information), indicating that compound **3** had the same a 3,4-dihydro-2H-benzopyran framework as **1** with the 3,3-dimethylallyl and cinnamoyl substituents at C-3 and C-1, respectively. A major difference was found in the NMR splitting patterns for the aliphatic methine and methylene resonances [δ_{H} 3.00 (1H, d, J = 10.7 Hz,

H-7), 2.60 (1H, dd, $J = 10.7, 8.1$ Hz, H-8), 2.34 (1H, brt, $J = 8.1$ Hz, H-13), 2.04 (1H, m, H-11a), 1.67 (2H, m, H-11b, H-12a), 1.58 (1H, m, H-12b), 1.41 (3H, s, Me-10), 1.29 (3H, s, Me-16), 0.70 (3H, s, Me-15); δ_{C} 34.7, 36.5, 45.2, 37.6, 24.8, 26.7, 32.6, 16.8]. The singlet resonance at δ_{H} 6.00 (1H, s) was assigned to a hydroxy group due to its lack of a cross peak in the HMQC experiment. However, this resonance showed cross-peaks in the HMBC spectrum (Figure 1) with C-3 (δ_{C} 104.6), C-4 (δ_{C} 159.5), and C-5 (δ_{C} 102.4). The connectivity of H-7/H-8, H-11/H-12, H-12/H-13 and H-13/H-8 in the COSY spectrum led to assignment of all aliphatic protons. The *gem*-dimethyl moiety was located at C-14 based on cross peaks in the HMBC spectrum from Me-15 to C-7 (δ_{C} 34.7), C-13 (δ_{C} 45.2), C-14 (δ_{C} 38.2), and C-16 (δ_{C} 32.6), from H-7 to C-8 (δ_{C} 36.5), C-9 (δ_{C} 83.7), C-13, C-14, C-15 (δ_{C} 16.8), and C-16, and from Me-10 to C-8. The relative configurations of C-7, C-8, and C-13 were assigned tentatively by the coupling constants of 10.7 and 8.1 Hz, suggesting β -orientations of the attached protons as shown in Figure 2. The NOESY correlations (Figure 2) of H-7/H-8/Me-16, H-8/Me-10, H-8/H-13, and H-13/Me-16 supported the assignments. Compound **3** was also analyzed by chiral-phase HPLC, but only a single peak was observed. The structure of mallopinin C (**3**) was thus assigned as shown in Chart 1.

Compound **4**, characterized as a pale yellow viscous oil with $[\alpha]_{\text{D}}^{25} -31.6$ (c 0.2, MeOH), has a molecular formula of $\text{C}_{22}\text{H}_{22}\text{O}_5$ based on the HRTOFESIMS (m/z 389.1375; calcd for 389.1365, $\text{C}_{22}\text{H}_{22}\text{O}_5\text{Na}$) and ^{13}C NMR data. The UV spectrum is typical of a flavanone chromophore with absorption bands at λ_{max} 234, 264, 274, and 298 nm, while the IR absorption band at 1701 cm^{-1} implied a conjugated carbonyl moiety. The ^1H NMR data (Table 2, Figure S27, Supporting Information) indicated the presence of a deshielded hydrogen-bonded hydroxy group (δ_{H} 12.37), a monosubstituted aromatic ring [δ_{H} 7.44 (5H)], a 2,2-dimethyl-2H-pyran unit (δ_{H}

6.64, 5.52, 1.47 and 1.44), an ABX spin system (δ_{H} 5.45, 3.06 and 2.89), and a methoxy group (δ_{H} 3.35). The NMR data were similar to those of 6,6-dimethylpyrano[b,g]-5-hydroxy-8-methylflavanone,¹¹ except for the presence of two doublets of an oxygenated methylene group (δ_{H} 4.49 and 4.43) instead of a singlet for an aromatic methyl substituent. These proton resonances showed correlations with C-7 (δ_{C} 160.9), C-8 (δ_{C} 105.3), C-9 (δ_{C} 161.0), and 11-OMe (δ_{C} 57.9) in the HMBC spectrum (Figure 1), indicating their attachment at C-8. A negative Cotton effect at 280 nm in the ECD spectrum defined the (2*S*) absolute configuration of **4**.¹² Thus, the structure of mallopinin D (**4**) was assigned as shown in Chart 1.

Compound **5**, a pale yellow solid, has a formula of C₂₂H₂₀O₆ based on the HRTOFESIMS ion at m/z 381.1341 [M + H]⁺ (calcd for 381.1338) and the ¹³C NMR data. The UV and IR spectra were consistent with a flavanone skeleton. Compound **5** had the same skeleton as **4**, by comparison of their NMR data (Table 2, Figures S35 and S36, Supporting Information). The lack of the OMe resonance at δ_{H} 3.35 indicated a presence of a hydroxycarbonyl group at C-11 confirmed by the HMBC correlations (Figure 1) between H-11 and C-7 (δ_{C} 159.2), C-8 (δ_{C} 110.8), C-9 (δ_{C} 159.5), and C-12 (δ_{C} 176.5). The (2*S*) configuration of **5** was elucidated based on its ECD data. Therefore, the structure of mallopinin E (**5**) was identified as shown in Chart 1.

Possible biosynthetic pathways for compounds **1-3** and **11** are illustrated in Scheme 1. These compounds could be obtained from a cinnamoyl-CoA starter and three malonyl-CoA extenders in a multi-step process.¹³ Epoxidation of metabolite **i** followed by ring closure and dehydration could yield chromene **iv**. The intramolecular cyclization of **iv** would give chalcone **1**, which could undergo further C-C cleavage to form compound **2**. A formal [2+2] cycloaddition

(possibly either photochemically induced or via a non-concerted process) of the precursor **iv** could produce compound **3**.

The MIC values of some isolated compounds against Gram-positive and Gram-negative bacteria are summarized in Table 3. Only compounds **6** and **7** showed significant antibacterial activities against *M. luteus*, *S. mutans*, *B. cereus*, *S. aureus* and *E. coli* with MIC values ranging from 3.8-15.5 μ M. With MIC values of 30 μ M, compounds **9** and **10** exhibited moderate activity against *B. cereus*, while compound **10** also showed moderate activity against all tested Gram-positive strains. The remaining compounds showed weak or no activity. It is interesting to note that compound **7** with a hydroxy group at C-4' had better activity than **6**, which lacked this substituent.

EXPERIMENTAL SECTION

General Experimental Procedures. The instruments and materials in this study were the same as previously described.¹⁴⁻¹⁶

Plant Material. The fruits of *M. philippensis* were collected from Doi Tung (Chiang Rai Province, Thailand, GPS: 20°17.351' N, 99°48.661' E) in August 2015 and authenticated by Mr. Martin Van de Bult, Doi Tung Development Project. A specimen of the material (MFU-NPR0123) is stored at the Natural Products Research Laboratory, School of Science, Mae Fah Luang University.

Extraction and Isolation. Ground dried fruits of *M. philippensis* (7.0 kg) were soaked with acetone (2 $\times$ 5 L, 3 days) and the mixture was concentrated under reduced pressure to afford the acetone extract (648.6 g). The extract was separated by quick column chromatography (QCC) using hexanes as eluent followed by gradient elution with increasing polarity with EtOAc (100:0

– 0:100) to give 10 fractions (Fr. 1–Fr. 10). Fraction Fr. 2 (8.05 g) was subjected to CC using EtOAc–hexanes (1:19) as mobile phase to afford eight subfractions (Fr. 2a–Fr. 2h). Compound **2** (4.7 mg) was obtained from subfraction Fr. 2d (503.5 mg) by CC over Sephadex LH-20, with 100% MeOH as mobile phase, followed by recrystallization with MeOH–CH₂Cl₂ (1:4). Subfraction Fr. 2e (1.17 g) was chromatographed by CC using EtOAc–hexanes (1:99) as mobile phase followed by CC over Sephadex LH-20, with MeOH–CH₂Cl₂ (1:1) as mobile phase to provide **3** (7.3 mg). Compound **1** (255.5 mg) was purified by recrystallization from MeOH–CH₂Cl₂ (9:1) from fraction Fr. 4 (2.01 g). Fraction Fr. 5 (131.4 g) was subjected to CC with EtOAc–hexanes (0:100 –100:0) as mobile phase to give six subfractions (Fr. 5a – Fr. 5f). Subfraction Fr. 5d (50.8 mg) was separated by Sephadex LH-20 CC [eluted with MeOH–CH₂Cl₂ (1:1)] to give eight subfractions (Fr. 5d1–Fr. 5d8). Purification of subfraction Fr. 5d2 (3.25 g) by CC using EtOAc–hexanes (1:19) as mobile phase and by CC over Sephadex LH-20, with MeOH–CH₂Cl₂ (1:1) as mobile phase afforded **4** (11.2 mg), **6** (1.8 g), **7** (115.8 mg), **10** (89.8 mg) and **11** (52.1 mg). Fraction Fr. 6 (1.50 g) was chromatographed by CC eluting with EtOAc–hexanes (1:9) as mobile phase, followed by CC over RP-18 with MeOH–H₂O (7:3) yielding **5** (4.8 mg), **8** (455.0 mg) and **9** (43.7 mg).

Mallophenin A (1): orange crystals; mp 188.1–189.3 °C; $[\alpha]_D^{25}$ 0 (c 0.11, CHCl₃); UV (MeOH) $\lambda_{\max}$ (log ϵ) 236 (2.86), 307 (2.80), 351 (2.90) nm; IR (KBr) $\nu_{\max}$ 3327, 2968, 2932, 1691, 1631, 1555, 1131, 1051, 947 cm⁻¹; ¹H and ¹³C NMR, see Table 1; HRESIMS $[M]^+$ m/z 458.2452 (calcd for 458.2457, C₃₀H₃₄O₄).

Chiral-phase HPLC Analysis and ECD Spectra of (+)-1 and (–)-1. The racemate **1** (4.0 mg) was resolved by chiral-phase HPLC on a CHIRALCEL OD-H column, using *n*-hexane–iPrOH (99:1) at 1.5 mL/min. Compound (+)-**1** (t_R = 12.5 min) [(1.6 mg), $[\alpha]_D^{24}$ +28.4 (c

0.03, CHCl₃]; ECD (c 4.0×10^{-4} M, MeOH) $\lambda_{\max}$ ($\Delta\epsilon$); 224 (-5.01×10^{-1}), 245 ($+1.93 \times 10^{-1}$), 304 ($+1.10 \times 10^{-1}$) and 363 (-1.51×10^{-1}) nm] and (-)-**1** (t_R = 14 min) [(1.5 mg), $[\alpha]_D^{24}$ -24.6 (c 0.03, CHCl₃); ECD (c 7.0×10^{-4} M, MeOH) $\lambda_{\max}$ ($\Delta\epsilon$); 222 (+8.93), 246 (-2.07×10^{-1}), 302 (-8.73) and 366 ($+1.57 \times 10^{-1}$) nm] were obtained.

Malloopenin B (2): pale yellow solid; mp 129.5–130.0 °C; $[\alpha]_D^{25}$ 0 (c 0.07, CHCl₃); UV (MeOH) $\lambda_{\max}$ (log ϵ) 217 (3.92), 302 (2.58), 354 (2.13) nm; IR (neat) $\nu_{\max}$ 3282, 2923, 1713, 1596, 1494, 1156, 1081, 811, 664 cm⁻¹; ¹H and ¹³C NMR, see Table 1; HRESIMS [M + H]⁺ m/z 371.2216 (calcd for 371.2222, C₂₃H₃₁O₄).

Chiral-phase HPLC Analysis and ECD Spectra of (+)-2 and (-)-2. The racemate **2** (3.0 mg) was resolved by chiral-phase HPLC on a CHIRALCEL OD-H column, using *n*-hexane–iPrOH (99:1) at 1.0 mL/min. Compound (+)-**2** (t_R = 13.0 min) [(1.1 mg), $[\alpha]_D^{24}$ +17.4 (c 0.02, CHCl₃); ECD (c 1.0×10^{-3} M, MeOH) $\lambda_{\max}$ ($\Delta\epsilon$); 275 ($+1.62 \times 10^{-1}$), 316 (-7.48) and 355 (-1.05×10^{-1}) nm] and (-)-**2** (t_R = 13.5 min) [(1.0 mg), $[\alpha]_D^{24}$ -11.8 (c 0.02, CHCl₃); ECD (c 2.1×10^{-3} M, MeOH) $\lambda_{\max}$ ($\Delta\epsilon$); 273 (-1.34×10^{-1}), 319 (+9.98) and 352 ($+1.09 \times 10^{-1}$) nm] were obtained.

Malloopenin C (3): pale yellow solid; mp 134.7–136.2 °C; $[\alpha]_D^{24}$ +7.6 (c 0.02, MeOH); UV (MeOH) $\lambda_{\max}$ (log ϵ) 220 (4.12), 301 (2.36), 350 (2.31) nm; IR (KBr) $\nu_{\max}$ 3325, 3191, 2961, 1675, 1622, 1517, 1201, 1109, 742 cm⁻¹; ¹H and ¹³C NMR, see Table 1; HRTOFESIMS [M + H]⁺ m/z 459.2505 (calcd for 459.2535, C₃₀H₃₄O₄).

Malloopenin D (4): pale yellow solid; mp 144.5–145.8 °C; $[\alpha]_D^{25}$ -31.6 (c 0.2, MeOH); ECD (c 0.5×10^{-4}) $\lambda_{\max}$ ($\Delta\epsilon$) 280 (-1.05) nm; UV (MeOH) $\lambda_{\max}$ (log ϵ) 234 (3.46), 264 (4.12), 274 (4.31),

298 (2.96) nm; IR (KBr) $\nu_{\max}$ 2917, 1701, 1629, 1584, 1246, 1067, 943 cm^{-1} ; ^1H and ^{13}C NMR, see Table 2; HRTOFESIMS $[\text{M} + \text{Na}]^+ m/z$ 389.1375 (calcd for 389.1365, $\text{C}_{22}\text{H}_{22}\text{O}_5\text{Na}$).

Mallopenin E (5): pale yellow solid; mp 215.9–217.3 °C; $[\alpha]_{\text{D}}^{21} -23.5$ (c 0.23, EtOH); ECD (c 1.8×10^{-3}) $\lambda_{\max}$ ($\Delta\epsilon$) 272 (–2.06), 297 (+0.43) nm; UV (EtOH) $\lambda_{\max}$ (log ϵ) 305 (3.02), 297 (4.71) nm; IR (KBr) $\nu_{\max}$ 3490, 3020, 2976, 2930, 1709, 1624, 1445, 1128, 755, 699 cm^{-1} ; ^1H and ^{13}C NMR, see Table 2; HRTOFESIMS $[\text{M} + \text{H}]^+ m/z$ 381.1341 (calcd for 381.1338, $\text{C}_{22}\text{H}_{20}\text{O}_6$).

X-ray Crystallographic Analysis of (+)-1. The measurement of (+)-1 was performed on a Bruker APEX-II CCD diffractometer with Cu K α radiation ($\lambda = 1.54178$ Å). The structure was solved and refined using the Bruker SHELXTL Software Package. Crystal data for (+)-1: $\text{C}_{30}\text{H}_{34}\text{O}_4$, $M_r = 458.57$, orthorhombic, $P2_12_12_1$ (No. 19), $a = 7.8303(2)$ Å, $b = 10.9599(2)$ Å, $c = 28.0066(6)$ Å, $\alpha = \beta = \gamma = 90^\circ$, $V = 2403.51(9)$ Å³, $T = 90(2)$ K, $Z = 4$, $Z' = 1$, $\mu(\text{CuK}\alpha) = 0.655$, 26458 reflections measured, 4384 unique ($R_{\text{int}} = 0.0395$) which were used in all calculations. The final wR_2 was 0.0790 (all data) and R_1 was 0.0314 ($I > 2(I)$). The Flack parameter value was 0.06(7). Crystallographic data for (+)-1 have been deposited at the Cambridge Crystallographic Data Centre (no. CCDC 1896823). These data can be obtained free of charge via the Internet at www.ccdc.cam.ac.uk/data_request/cif.

Antibacterial Assay. MIC assessments were carried out following the CLSI guidelines by broth microdilution in 96-well microtiter plates that was described in detail previously.¹⁶⁻¹⁸

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at <http://pubs.acs.org>.

1D and 2D NMR, HRTOFESIMS spectra of **1–5** (PDF)

X-ray crystallography for **1** (CIF)

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Notes

The authors declare no competing financial interest.

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Chart 1

Figure 1. $^1\text{H} \rightarrow ^1\text{H}$ COSY and key HMBC ($^1\text{H} \rightarrow ^{13}\text{C}$) correlations of **1**–**5**.

Figure 2. Selected NOSEY correlations of (+)-**1** and **3**.

Figure 3. ORTEP diagram for (+)-**1**.

Figure 4. Experimental ECD spectra of compound (+) and (-)-**1**, and (+) and (-)-**2**.

Scheme 1. Putative Biosynthetic Pathways for **1-3** and **11**.

Table 1. ^1H (400 MHz, CDCl_3) and ^{13}C NMR (100 MHz, CDCl_3) Spectroscopic Data of **1-3**

position	1		2		3^a	
	δ_{C}	δ_{H} (mult., J in Hz)	δ_{C}	δ_{H} (mult., J in Hz)	δ_{C}	δ_{H} (mult., J in Hz)
1	106.5		106.1		106.2	
2	163.8		162.7		161.8	
3	111.5		110.5		104.6	
4	161.1		160.9		159.5	
5	106.4		106.0		102.4	
6	156.4		156.7		156.6	
7	27.8	2.84, br s	27.7	2.80, br s	34.7	3.00, d (10.7)
8	35.3	2.25, dt (12.8, 3.5) 1.89, dd (12.8, 1.1)	34.9	2.19, m 1.84, m	36.5	2.60, dd (10.7, 8.1)
9	76.3		76.1		83.7	
10	29.1	1.53, s	28.9	1.43, s	26.7	1.41, s
11	37.6	1.85, m 1.46, m	37.7	1.81, m 1.49, m	37.6	2.04, m 1.67, m
12	22.1	1.33, m 0.92, m	22.1	1.30, m 0.87, m	24.8	1.67, m 1.58, m
13	46.0	2.04, m	46.0	2.03, m	45.2	2.34, br t (8.1)
14	84.9		84.8		38.2	
15	24.4	1.08, s	24.4	1.05, s	16.8	0.70, s
16	29.8	1.56, s	29.8	1.56, s	32.6	1.29, s
17	192.2		202.8		192.2	
18	128.0	8.12, d (15.6)	32.5	2.61, s	127.3	7.80, d (15.7)
19	141.4	7.77, d (15.6)			140.1	7.66, d (15.7)
1'	21.3	3.25, m	21.3	3.22, m	20.7	3.39, dd (16.4, 6.7) 3.30, dd (16.4, 7.1)
2'	123.2	5.21, br t (7.0)	123.2	5.18, br s	120.8	5.19, br t (7.1)
3'	130.9		130.9		135.1	
4'	17.9	1.79, s	17.8	1.77, s	16.9	1.76, s
5'	25.8	1.67, s	25.8	1.65, s	24.8	1.71, s
1''	135.9				134.9	
2''/6''	128.3	7.60, m			127.2	7.53, m
3''/5''	128.9	7.38, m			127.9	7.32, m
4''	128.8	7.38, m			128.7	7.32, m
4-OH						6.00, s
2-OH		14.39, s		13.88, s		14.21, s

^aRecorded at 500 MHz.

Table 2. ^1H (400 MHz, CDCl_3) and ^{13}C NMR (100 MHz, CDCl_3) Spectroscopic Data of **4** and **5**

position	4		5^a	
	δ_{C}	δ_{H} (mult., J in Hz)	δ_{C}	δ_{H} (mult., J in Hz)
2	78.9	5.45, br d (12.9)	78.6	5.44, dd (12.9, 3.0)
3	43.2	3.06, dd (16.7, 12.9)	42.9	3.06, dd (17.1, 12.9)
		2.89, dd (16.7, 3.0)		2.86, dd (17.1, 3.0)
4	196.2		195.6	
5	158.3		157.2	
6	102.9		102.2	
7	160.9		159.2	
8	105.3		110.8	
9	161.0		159.5	
10	102.5		102.4	
11	61.8	4.49, d (11.0)	27.3	3.63, s
		4.43, d (11.0)		
12			176.5	
1'	138.6		137.9	
2'/6'	125.9	7.44, m	125.4	7.42, m
3'/5'	128.8	7.44, m	128.4	7.42, m
4'	128.6	7.44, m	128.3	7.42, m
1''	115.5	6.64, d (9.1)	114.9	6.65, d (10.1)
2''	126.3	5.53, d (9.1)	125.7	5.52, d (10.1)
3''	78.6		78.5	
4''	28.3	1.47, s	27.9	1.43, s
5''	28.3	1.45, s	28.0	1.44, s
5-OH		12.37, s		12.30, s
11-OMe	57.9	3.35, s		

^aRecorded at 600 MHz.

Table 3. Antibacterial Activity of Compounds **2-11**

Compound	MIC (μM)								
	Gram-positive bacteria					Gram-negative bacteria			
	<i>M. luteus</i>	<i>S. mutans</i>	<i>S. epidermidis</i>	<i>B. cereus</i>	<i>S. aureus</i>	<i>S. typhimurium</i>	<i>Ps. aeruginosa</i>	<i>E. coli</i>	<i>S. flexneri</i>
($\pm$)-2	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
3	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
4	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
5	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
6	15.5	15.5	> 100	15.5	15.5	> 100	> 100	15.5	> 100
7	30.1	3.8	> 100	3.8	15.0	> 100	> 100	7.5	> 100
8	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
9	> 100	> 100	> 100	30.1	> 100	> 100	> 100	> 100	> 100
10	30.9	30.9	> 100	30.9	61.8	> 100	> 100	30.9	> 100
11	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100	> 100
Vancomycin	0.2	0.2	0.2	0.1	0.2				
Gentamicin						1.0	1.0	0.5	1.0

Table of Content

Mallophenins A-E, Antibacterial Phenolic Derivatives from the Fruits of *Mallotus philippensis*

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