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Synthesis and Antimalarial Activity of Mallatojaponin C and Related Compounds

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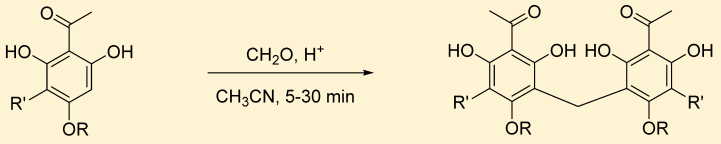
Supporting Information



10 R = Me, R' = H IC_{50} >20 μM
11 R = Me, R' = prenyl IC_{50} 1.7 μM
12 R = Me, R' = geranyl IC_{50} 0.57 μM
13 R = Me, R' = farnesyl IC_{50} 0.56 μM
14 R = Me, R' = $CH_2CH_2C(CH_3)_2OH$ IC_{50} >20 μM
16 R = H, R' = prenyl IC_{50} >20 μM
17 R = H, R' = geranyl IC_{50} 6.6 μM
18 R = H, R' = farnesyl IC_{50} 3.6 μM

1 R = Me, R' = prenyl IC_{50} 0.64 μM
5 R = Me, R' = H; IC_{50} 8.1 μM
6 R = Me, R' = geranyl IC_{50} 1.7 μM
7 R = Me, R' = $CH_2CH_2C(CH_3)_2OH$ IC_{50} >20 μM
8 R = H, R' = prenyl IC_{50} >20 μM
9 R = H, R' = geranyl IC_{50} >10 μM

ABSTRACT: The phloroglucinol mallotojaponin C (**1**) from *Mallotus oppositifolius*, which was previously shown by us to have both antiplasmodial and cytocidal activities against the malaria parasite *Plasmodium falciparum*, was synthesized in three steps from 2′,4′,6′-trihydroxyacetophenone, and various derivatives were synthesized in an attempt to improve the bioactivity of this class of compounds. Two derivatives, the simple prenylated phloroglucinols **12** and **13**, were found to have comparable antiplasmodial activities to that of mallotojaponin C.

The phloroglucinol mallotojaponin C (**1**) was first isolated from *Mallotus oppositifolius* Muell. Arg., a member of the Euphorbiaceae family,[2] and was shown to have both cytostatic and cytocidal activity against the chloroquine/mefloquine-resistant Dd2 strain of *Plasmodium falciparum*. The related compounds methylated mallotojaponin C (**2**), mallotophenone (**3**), and mallotojaponin B (**4**) were also obtained, but had less potent antimalarial activity than **1**, indicating the importance of the alkenyl side chain and the phenolic hydroxy substituents at C-2 and C-6 for antiplasmodial activity. Many monomeric phloroglucinols have been investigated for their antimalarial activity,[3−5] and some additional dimeric compounds from *M. oppositifolius* have recently been shown to have trypanocidal and antileishmanial activities.[6] This paper describes the syntheses and biological evaluations of mallotojaponin C and various derivatives that were reported in 2015 in a Ph.D. dissertation[7] and complements the results in an excellent recent paper by Cariou and Dubois et al.[8]

RESULTS AND DISCUSSION

Synthesis of Mallatojaponin C and Analogues. The prenylated monomeric phloroglucinols **11**−**14** and **16**−**18** (Scheme 1) were synthesized by prenylation of either 2,4,6-trihydroxyacetophenone (**15**) or 2,6-dihydroxy-4-methoxyacetophenone (**10**) (Scheme 1). Prenylation of the precursors **10** and **15** was achieved by reaction of the phenol with either sodium hydroxide or lithium hydroxide and the appropriate alkenyl halide in 80% aqueous ethanol at room temperature; the solvent was chosen based on a previous study of the effect of solvent on the C-alkylation of phloroglucinols and on trials of various solvents for the synthesis of **11**.[9,10] The use of sodium hydroxide led to higher yields of **11**, **12**, **16**, and **17** than the use of lithium hydroxide. Compounds **11**,[8,11−14] **16**,[14−17] **17**,[18,19] and **18**[17,20] have previously been synthesized, and their structures were confirmed by MS and by comparison of their ^1H and ^{13}C NMR spectra with previously published spectra. Thus, treatment of **10** with 3,3-dimethylallyl bromide (prenyl bromide) in 80% aqueous ethanol with 1.1 equiv of NaOH at room temperature for 16 h gave the monoprenylated product **11** in 33% yield. Alkylation of **10** with geranyl bromide and farnesyl bromide under the same conditions yielded the corresponding geranylated and farnesylated products **12** and **13**, with the yields decreasing with increasing chain length. Purification of the final product was carried out in each case by reversed-phase chromatography over C_{18}-silica gel. Purification of **11** over silica gel in the presence of 0.1% formic acid yielded the hydroxylated product **14**.

Alkylation of **15** with prenyl, geranyl, and farnesyl bromides gave the alkylated derivatives **16**−**18** in low to moderate yields.

The synthesis of dimeric phloroglucinols such as mallatojaponin C (**1**) required the coupling of the monomeric

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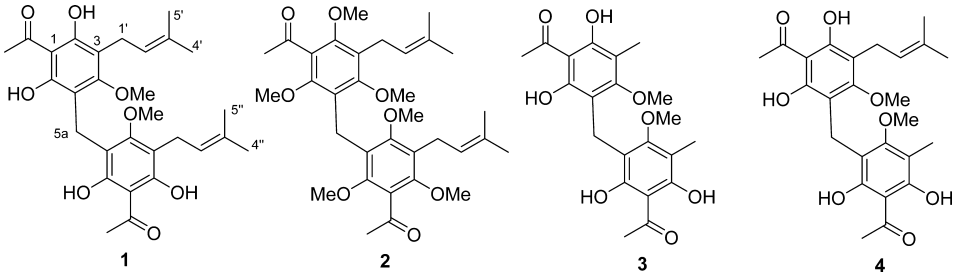
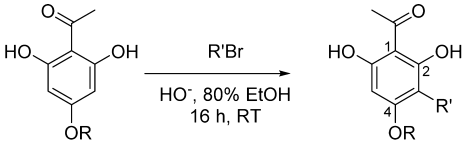


Figure 1. Structures of mallatojapinin C (**1**), tetramethylmallatojapinin C (**2**), mallotophenone (**3**), and mallotojapinin B (**4**).

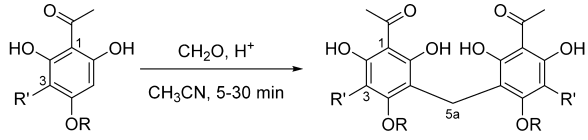
Scheme 1. Synthesis of Alkylated Phloroglucinols 11−18



10 R = Me
15 R = H

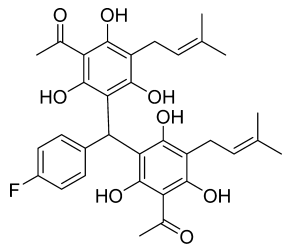
11 R = Me, R' = prenyl; 33%
12 R = Me, R' = geranyl; 30%
13 R = Me, R' = farnesyl; 29%
14 R = Me, R' = $CH_2CH_2C(CH_3)_2OH$; 36%
16 R = H, R' = prenyl; 27%
17 R = H, R' = geranyl; 32%
18 R = H, R' = farnesyl; 27%

Scheme 2. Synthesis of Dimeric Phloroglucinol Derivatives



11 R = Me, R' = prenyl
10 R = Me, R' = H
12 R = Me, R' = geranyl
14 R = Me, R' = $CH_2CH_2C(CH_3)_2OH$
16 R = H, R' = prenyl
17 R = H, R' = geranyl

1 R = Me, R' = prenyl; 10%
5 R = Me, R' = H; 26%
6 R = Me, R' = geranyl; 2%
7 R = Me, R' = $CH_2CH_2C(CH_3)_2OH$; 25%
8 R = H, R' = prenyl; 10%
9 R = H, R' = geranyl; 11%



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compounds **10−12**, **14**, and **16−17**. The initial coupling conditions used were based upon previous work with similar substrates,[21−23] but it was apparent that the alkene moiety was readily hydrated under the standard acidic conditions. Most of the previously published work was on derivatives not containing an alkene moiety, so this problem did not arise in these cases. The coupling conditions were thus optimized by a study of synthetic conditions for compound **8**, and various solvents, catalysts, and reaction durations were investigated. The best conditions were found to be the use of acetonitrile as solvent and a small amount of concentrated sulfuric acid as catalyst to reduce hydration of the alkene. These conditions allowed the reaction to be completed in less than 30 min, and this short reaction time also reduced the amount of hydration. Although the reaction yield was low, the desired product could easily be purified. These conditions were then applied to the synthesis of mallatojapinin C (**1**) and compounds **5−9**. The latter compounds were prepared in an attempt to improve upon the antiparasitic activity of the coupled compounds and specifically to investigate the effect of the length of the alkenyl chain and the substituent at C-5 on the bioactivity of the resulting compounds. In the case of the known compound **1** the structure was confirmed by comparison of spectroscopic data with the published data,[2] and the structures of the other compounds were established by analysis of their spectroscopic data.

The synthesis of other derivatives was attempted by replacing formaldehyde with various aldehydes. The synthesis of the *p*-fluorophenyl derivative **19** initially appeared to be successful, as judged by its ^{1}H NMR spectrum, consistent with its assigned structure, and the compound showed promising bioactivity, with an IC_{50} value in the 2−4 μM range. Unfortunately, the compound proved to be unstable on storage in the freezer and decomposed before MS data could be obtained and an accurate IC_{50} value could be determined. Attempts to synthesize other proposed derivatives were unsuccessful, presumably because of the stability of the secondary benzylic carbocation that would be formed in an acid-catalyzed retrocoupling reaction.

Antiplasmodial Activities of Mallatojaponin C Analogues. The synthetic compounds **1−18** were evaluated for their antiplasmodial activities against the Dd2 strain of *P. falciparum*. The results are presented in Table 1, together with the data for some of the same compounds from the recent

Table 1. Antiparasitic Activities of Natural and Synthetic Phloroglucinols and Analogues

compound number (this work)	IC_{50} (μM) Dd2 strain	compound number (ref 8)	IC_{50} (μM) FcB1/ Columbia strain
1	0.64 ± 0.1[a]	**2**	0.75 ± 0.11
5	8.1 ± 0.8	**5a**	17.6 ± 4.3
6	1.7 ± 0.8	**12**	6 ± 0.4
7	>10	NR[b]	
8	>20	NR	
9	>10	NR	
10	>20	NR	
11	1.7 ± 0.4	**6**	7.4 ± 1.1
12	0.57 ± 0.09	**7**	4.4 ± 1.3
13	0.56 ± 0.07	NR	
14	>20	NR	
15	>20	NR	
16	>20	NR	
17	6.6 ± 2.4	NR	
18	3.6 ± 0.8	NR	

[a]This value differs from that reported in ref 2. [b]NR = not reported in ref 8.

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Cariou and Dubois et al. paper.[8] The potencies against the two different strains of *P. falciparum* show similar trends, but the Dd2 strain is more sensitive to these compounds than the FcB1/Columbia strain.

For the dimeric phloroglucinols (**1**, **5**−**9**), it was found that the prenyl side chain maximized the bioactivity, with the bisprenylated natural product **1** having the best potency. Both the monoprenylated compound **4** and the bisgeranylated compound **6** were less potent than **1** against the Dd2 strain. Methylation at the 4-position is also very important. Thus, the bisgeranyl compound **6**, with methylation at both the 4- and 4′-hydroxy groups, has an IC_{50} value of 1.7 ± 0.8 μM, while the corresponding unmethylated compound **8** is much less potent, with an IC_{50} value greater than 20 μM. This difference may be partly due to polarity, since the hydrated derivative **7** is much less active than the unhydrated natural product **1**.

The situation is somewhat different with the monomeric phloroglucinol derivatives. With these compounds potency increases as the alkenyl side chain increases in length from prenyl to geranyl to farnesyl in both the 4-methylated series (compounds **11**−**13**) and the unmethylated series (compounds **16**−**18**). As with the dimeric phloroglucinols, the compounds in the 4-methylated series are more potent than those in the unmethylated series, but the differences are smaller. Thus, the unmethylated farnesyl compound **18** is only about 9-fold less potent than its methylated analogue **13**.

Finally, the excellent potencies of the monomeric compounds **12** and **13** are worth noting. Both compounds show submicromolar activities, with the farnesylated compound **13** having an IC_{50} value of 0.56 μM against the Dd2 strain, comparable to the dimeric lead compound **1**. It is thus conceivable that further explorations in this chemical space could yield relatively simple compounds with significant antiparasitic activities.

EXPERIMENTAL SECTION

General Experimental Procedures. Melting points were obtained using a Büchi B-540 melting point apparatus. UV−vis spectra were recorded on a Shimadzu UV-1201 spectrophotometer. NMR spectra were recorded on Varian 400 or Bruker Avance 500 MHz spectrometers. Mass spectra were obtained with an Agilent 6220 LC-TOF-MS. Preparatory HPLC separations were performed using Shimadzu LC-8A pumps coupled with a Shimadzu SPD-M10A diode array detector, a SCL-10A controller, and a Varian Dynamax C_{18} column (250 × 21.4 mm). Semipreparative HPLC separations were performed using Shimadzu LC-10AT pumps coupled with a Shimadzu SPD-M10A diode array detector, a SCL-10A system controller, and a Phenomenex Luna C_{18} column (250 × 10 mm). Bioactive compounds were checked for purity by analytical HPLC analysis using Shimadzu LC-10AT pumps coupled with a Shimadzu SPD-M10A diode array detector, a Sedex 75 evaporative light scattering detector, a SCL-10A system controller, and a Phenomenex Luna C_{18} column (250 × 4.6 mm). Compounds **10** and **15** were purchased from Indofine Chemical Company and tested for bioactivity without further purification. Compounds **2** and **3** were isolated from *M. oppositifolius*, and compound **4** was previously synthesized from **1**.[2] Geranyl bromide was purchased from Alfa Aesar, and prenyl and farnesyl bromides were purchased from Sigma-Aldrich.

Standard Procedure A for Synthesis of Alkylated Phloroglucinols 11−14 and 16−18. The following general procedure was used. Compound **10** or **15** (1 equiv) was dissolved in a solution of 80% EtOH (40 mL) containing LiOH or NaOH (2 equiv). Prenyl, geranyl, or farnesyl bromide (1.1 equiv) was added, and the reaction mixture was stirred for 16 h at rt. The reaction mixture was acidified with 3 N HCl and extracted with CH_2Cl_2 (3 × 50 mL). The organic solution was washed with saturated aqueous NaCl (50 mL) and dried with $MgSO_4$, and the solvent was removed under reduced pressure. The residue was purified utilizing C_{18} HPLC ($MeOH/H_2O$ gradient with 0.1% formic acid) to yield the alkylated phloroglucinol (**11**−**14**, **16**−**18**).

Standard Procedure B for Synthesis of Dimeric Phloroglucinol Derivatives 1 and 5−9. The following general procedure was used. The alkylated phloroglucinol (**10**−**12**, **14**, **16**, or **17**, 0.1−0.5 mmol) was dissolved in CH_3CN. Formaldehyde (37% aqueous, 10 equiv) was added, followed by the addition of concentrated H_2SO_4 (1 drop). The reaction mixture was stirred for 5 min at rt. The reaction mixture was diluted with water (10 mL) and extracted with EtOAc (3 × 10 mL). The organic solution was washed with saturated NaCl (10 mL) and dried with $MgSO_4$, and the solvent was removed under reduced pressure. The residue was purified utilizing C_{18} HPLC ($MeOH/H_2O$ gradient with 0.1% formic acid) to yield the dimeric phloroglucinol derivatives **1** and **5**−**9**.

Antiparasitic Bioassay. This assay was performed at Virginia Tech as previously described.[2]

Synthesis of 11. Standard procedure A was used with NaOH to convert **10** (495 mg, 2.72 mmol) to **11** (228 mg, 0.91 mmol, 33%).

Compound 11: UV (MeOH) λ_{max} (log ε) 215 (4.06), 288 (4.09), 333 (3.36) nm; ^{1}H NMR (MeOH-d_4, 400 MHz) δ 1.60 (3H, s, H-5′), 1.71 (3H, s, H-4′), 2.60 (3H, s, −$COCH_3$), 3.15 (2H, d, J = 7.2 Hz, H-1′), 3.78 (3H, s, −OCH_3), 5.10 (1H, m, H-2′), 5.96 (1H, s, H-5); ^{13}C NMR (MeOH-d_4, 100 MHz) δ 17.8, 22.0, 25.9, 33.1, 55.8, 90.9, 106.0, 108.9, 124.3, 131.1, 162.3, 163.5, 165.0, 205.0; HRESIMS $[M + H]^+$ m/z 251.1274 (calcd for $C_{14}H_{19}O_4^+$ 251.1278), $[M + Na]^+$ m/z 273.1114 (calcd for $C_{14}H_{18}NaO_4^+$ 273.1097).

Synthesis of 12. Standard procedure A was used with LiOH as base to convert **10** (150 mg, 0.82 mmol) to **12** (78 mg, 0.25 mmol, 30%). When NaOH was used as base, the yield of **12** from 100 mg (0.55 mmol) of **10** was 37 mg (0.12 mmol, 22%).

Compound 12: UV (MeOH) λ_{max} (log ε) 216 (4.16), 289 (4.14), 333 (3.38) nm; ^{1}H NMR (MeOH-d_4, 400 MHz) δ 1.60 (3H, s, H-10′), 1.67 (3H, s, H-9′), 1.87 (3H, s, H-8′), 2.08 (4H, m, H-4′, H-5′), 2.67 (3H, s, −$COCH_3$), 3.33 (2H, d, J = 7.0 Hz, H-1′), 3.81 (3H, s, −OCH_3), 5.06 (1H, m, H-6′), 5.19 (1H, m, H-2′), 6.02 (1H, s, H-5); ^{13}C NMR (MeOH-d_4, 100 MHz) δ 16.2, 17.7, 21.4, 25.7, 26.3, 32.9, 39.7, 55.7, 91.8, 105.4, 121.6, 123.7, 131.9, 163.1, 203.7; HRESIMS $[M + H]^+$ m/z 319.1896 (calcd for $C_{19}H_{27}O_4^+$ 319.1904), $[M + Na]^+$ m/z 341.1707 (calcd for $C_{19}H_{26}NaO_4^+$ 341.1723).

Synthesis of 13. Standard procedure A was used with LiOH as base. The yield of **13** from 100 mg (0.55 mmol) of **10** was 61 mg (0.16 mmol, 29%).

Compound 13: UV (MeOH) λ_{max} (log ε) 213 (4.21), 288 (4.14), 333 (3.38) nm; ^{1}H NMR (MeOH-d_4, 400 MHz) δ 1.60 (3H, s, H-10′), 1.67 (3H, s, H-9′), 1.87 (3H, s, H-8′), 2.08 (4H, m, H-4′, H-5′), 2.67 (3H, s, −$COCH_3$), 3.33 (2H, d, J = 7.0 Hz H-1′), 3.81 (3H, s, −OCH_3), 5.06 (1H, m, H-6′), 5.19 (1H, m, H-2′), 6.02 (1H, s, H-5); ^{13}C NMR (MeOH-d_4, 100 MHz) δ 16.2, 17.7, 21.4, 25.7, 26.3, 32.9, 39.7, 55.7, 91.8, 105.4, 121.6, 123.7, 131.9, 163.1, 203.7; HRESIMS $[M + H]^+$ m/z 387.2535 (calcd for $C_{24}H_{35}O_4^+$ 387.2530), $[M + Na]^+$ m/z 409.2346 (calcd for $C_{24}H_{34}O_4Na^+$ 409.2349).

Synthesis of 14. Standard procedure A was used with NaOH as base. The residue was purified by column chromatography on silica gel (9:1 hexanes/EtOAc/0.1% formic acid). The yield of **14** from 49.6 mg (0.27 mmol) of **10** was 24 mg (0.096 mmol, 36%).

Compound 14: UV (MeOH) λ_{max} (log ε) 216 (4.03), 290 (4.07), 331 (3.39) nm; ^{1}H NMR (MeOH-d_4, 400 MHz) δ 1.37 (6H, s, H-4′, H-5′), 1.77 (2H, t, J = 6.9 Hz, H-3′), 2.53 (2H, t, J = 6.9 Hz, H-2′), 2.58 (3H, s, −$COCH_3$), 3.83 (3H, s, −OCH_3), 6.01 (1H, s, H-5); ^{13}C NMR (MeOH-d_4, 100 MHz) δ 17.4, 26.9, 32.5, 33.6, 56.2, 77.1, 92.3, 102.1, 107.0, 157.7, 165.2, 166.5, 204.7; HRESIMS $[M - OH]^{+\bullet}$ m/z 251.1278 (calcd for $C_{14}H_{19}O_4^+$ 251.1278), $[M - C_4H_9O]^+$ m/z 195.0646 (calcd for $C_{10}H_{11}O_4^+$ 195.0652).

Synthesis of 16. Standard procedure A was used with LiOH as base. The yield of **16** from 980 mg (5.8 mmol) of **15** was 365 mg (1.55 mmol, 27%).

Compound 16: ^{1}H NMR (MeOH-d_4, 400 MHz) δ 1.63 (3H, s, H-5′) 1.73 (3H, s, H-4′), 2.60 (3H, s, −$COCH_3$), 3.17 (2H, d, J = 7.2

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Hz, H-1′), 5.16 (1H, m, H-2′), 5.89 (1H, s, H-5); ^{13}C NMR (MeOH-d_4, 100 MHz) δ 17.8, 22.1, 26.0, 32.8, 94.7, 105.5, 107.9, 124.5, 131.1, 161.8, 163.9, 164.8, 204.6; HRESIMS $[M + H]^+$ m/z 237.1119 (calcd for $C_{13}H_{17}O_4^+$ 237.1121), $[2M + NH_4]^+$ m/z 490.2405 (calcd for $C_{26}H_{36}NO_8^+$ 490.2435).

Synthesis of 17. Standard procedure A was used. With LiOH as base the yield of **17** from 253 mg (1.5 mmol) of **15** with LiOH was 147 mg (0.48 mmol, 32%), and with NaOH as base the yield of **17** from 100 mg (0.60 mmol) of **15** was 65 mg (0.48 mmol, 35%).

*Compound **17**:* ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.60 (3H, s, H-10′), 1.68 (3H, s, H-9′), 1.82 (3H, s, H-8′), 2.10 (4H, m, H-4′, H-5′), 2.67 (3H, s, $-COCH_3$), 3.37 (2H, d, J = 7.1 Hz, H-1′), 5.05 (1H, m, H-6′), 5.25 (1H, H-2′), 5.85 (1H, s, H-5); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 16.2, 17.7, 21.5, 25.7, 26.2, 32.9, 39.7, 95.3, 105.2, 105.4, 121.4, 123.6, 132.2, 140.1, 161.2, 203.6; HRESIMS $[M + H]+$ m/z 305.1744 (calcd for $C_{18}H_{25}O_4^+$ 305.1747), $[M + Na]^+$ m/z 327.1539 (calcd for $C_{18}H_{24}NaO_4^+$ 327.1567).

Synthesis of 18. Standard procedure A was used with LiOH as base. The yield of **18** from 101 mg (0.60 mmol) of **15** was 61 mg (0.16 mmol, 27%).

*Compound **18**:* light yellow powder; UV (MeOH) λ_{max} (log ε) 214 (4.21), 291 (4.19) nm; ^{1}H NMR (CD_3OD, 400 MHz) δ 1.54 (3H, s, H-15′), 1.56 (3H, s, H-14′), 1.65 (3H, s, H-13′), 1.79 (3H, s, H-12′), 1.97 (8H, m, H-4′, H-5′, H-8′, H-9′), 2.59 (3H, s $-COCH_3$), 3.18 (2H, d, J = 7.1 Hz, H-1′), 5.05 (2H, m, H-6′, H-10′), 5.17 (1H, m, H-2′), 5.89 (1H, s, H-5); ^{13}C NMR (CD_3OD, 100 MHz) δ 14.7, 14.7, 16.3, 20.6, 24.5, 26.0, 26.3, 31.4, 39.4, 39.4, 93.3, 104.1, 106.5, 123.3, 123.9, 124.1, 130.4, 133.0, 134.3, 160.4, 162.5, 163.4, 203.1; HRESIMS $[M + H]^+$ m/z 373.2366 (calcd for $C_{23}H_{33}O_4^+$ 373.2373), $[M + Na]^+$ m/z 395.2169 (calcd for $C_{23}H_{32}NaO_4^+$ 395.2193).

Synthesis of Mallotojaponin C (1). Standard procedure B was used with 3 mL of CH_3CN and HCl instead of H_2SO_4. The yield of **1** from 26.2 mg (0.1 mmol) of **11** was 6.1 mg (0.011 mmol, 10%).

Mallotojaponin C (1): light yellow powder; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.68 (6H, s, H_3-5′, H_3-5″), 1.77 (6H, s, H_3-4′, H_3-4″) 2.70 (6H, s, 2 × $-COCH_3$) 3.31 (4H, d, J = 6.5 Hz, H_2-1′, H_2-1″), 3.68 (2H, s, H_2-5a), 3.98 (2 × $-OCH_3$, 6H, s), 5.21 (2H, m, H-2′, H-2″), 9.06 (2H, s, $-$OH), 13.49 (2H, s, $-$OH); HRESIMS $[M + H]+$ m/z 513.2469 (calcd for $C_{29}H_{37}O_8^+$ 513.2483), $[M + Na]^+$ m/z 535.2297 (calcd for $C_{29}H_{36}O_8Na^+$ 535.2302).

Synthesis of 5. Standard procedure B was used with 3 mL of MeOH replacing CH_3CN. The reaction mixture was stirred for 16 h at rt. The residue was purified utilizing silica gel CC (1:1 hexanes/EtOAc). The yield of **5** from 100 mg (0.55 mmol) of **10** was 53.7 mg (0.14 mmol, 26%).

*Compound **5**:* ^{1}H NMR (DMSO-d_6, 400 MHz) δ 2.56 (6H, s $-COCH_3$), 3.61 (2H, s, H-5a), 3.69 (6H, s, $-OCH_3$), 6.00 (2H, s, H-3), 10.87 (2H, s, $-$OH), 13.48 (2H, s, $-$OH); ^{13}C NMR (DMSO-d_6, 100 MHz) 15.2, 30.7, 32.7, 39.5, 55.4, 90.2, 104.4, 160.8, 162.3, 163.9, 203.1; HRESIMS $[M + H]^+$ m/z 377.1197 (calcd for $C_{19}H_{21}O_8^+$ 377.1231), $[M + Na]^+$ m/z 399.1020 (calcd for $C_{19}H_{20}NaO_8^+$ 399.1050).

Synthesis of 6. Standard procedure B was used. The yield of **6** from 49.9 mg (0.16 mmol) of **12** was 2.2 mg (0.0034 mmol, 2%).

*Compound **6**:* UV (MeOH) λ_{max} (log ε) 204 (4.32), 289 (3.95) nm; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.56 (6H, s, H-10′), 1.63 (6H, s, H-9′), 1.77 (6H, s, H-8′), 1.98 (4H, m, H-5′), 2.05 (4H, bt, J = 7.4 Hz, H-4′), 2.70 (6H, s, $-COCH_3$), 3.31 (4H, d, J = 6.9 Hz, H-1′), 3.68 (2H, s, H-5a), 3.97 (6H, s, $-OCH_3$), 5.04 (2H, m, H-6′), 5.22 (2H, m, H-2′), 9.09 (2H, s, $-$OH), 13.47 (2H, s, $-$OH); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 16.2, 17.7, 22.7, 25.7, 26.6, 33.7, 39.6, 62.8, 108.4, 109.0, 114.1, 122.6, 124.2, 131.3, 135.7, 157.4, 159.6, 162.7, 205.3; HRESIMS $[M - H]^+$ m/z 647.3663 (calcd for $C_{19}H_{27}O_8^+$ 647.3579).

Synthesis of 7. Standard procedure B was used. The yield of **7** from 12.1 mg (0.045 mmol) of **14** was 6.2 mg (0.011 mmol, 25%).

*Compound **7**:* UV (MeOH) λ_{max} (log ε) 213 (4.33), 287 (4.25), 354 (3.70) nm; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.37 (12H, s, H-4′, H-5′), 1.74 (4H, t, J = 6.7 Hz, H-2′), 2.63 (6H, s, 2 × $-COCH_3$), 2.66 (4H, t, J = 6.8 Hz, H-1′, H-1″), 3.65 (2 × $-OCH_3$, 6H, s), 3.94 (H-5a, 2H, s), 13.80 ($-$OH, 2H, s); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 17.1, 27.0, 31.9, 33.8, 60.3, 75.6, 104.8, 108.2, 114.4, 155.2, 162.7, 163.4, 204.4.

Synthesis of 8. Standard procedure B was used. The reaction mixture was stirred for 30 min at rt. The precipitate was filtered and rinsed with MeOH to yield **8**. The yield of **8** from 14.3 mg (0.061 mmol) of **16** was 3 mg (0.0062 mmol, 10%). The compound appeared to be >90% pure by ^{1}H NMR spectroscopy.

*Compound **8**:* ^{1}H NMR (DMSO-d_6, 400 MHz) δ 1.59 (6H, s, H-5′, H-5″), 1.68 (6H, s, H-4′, H-4″), 2.63 (6H, s, 2 × $-COCH_3$,), 3.23 (4H, d, J = 6.7 Hz, H-1′, H-1″), 3.68 (2H, s, H-5a), 5.04 (2H, m, H-2′, H-2″); ^{13}C NMR (DMSO-d_6, 100 MHz) δ 17.0, 17.8, 21.5, 25.5, 32.6, 48.6, 105.8, 106.4, 108.1, 123.0, 130.4, 159.5, 203.7; HRESIMS $[M + H]^+$ m/z 485.2140 (calcd for $C_{27}H_{33}O_8^+$ 485.2170), $[M + Na]^+$ m/z 507.1946 (calcd for $C_{27}H_{32}O_8Na^+$ 507.1989).

Synthesis of 9. Standard procedure B was used. The yield of **9** from 49.3 mg (0.16 mmol) of **17** was 10.5 mg (0.017 mmol, 11%). Attempts at recrystallizing the compound in MeOH/H_2O were unsuccessful and resulted in decomposition before MS data could be obtained.

*Compound **9**:* UV (MeOH) λ_{max} (log ε) 208 (4.27), 230 (4.17), 292 (4.11) nm; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 1.60 (6H, s, H-10′, H-10″), 1.68 (6H, s, H-9′, H-9″), 1.83 (6H, s, H-8′, H-8″), 2.11 (8H, m, H-4′, H-5′, H-4″, H-5″), 2.67 (2 × $-COCH_3$, 6H, s), 3.41 (4H, d, J = 6.7 Hz, H-1′, H-1″), 3.79 (2H, s, H-5a,), 5.05 (2H, m, H-6′, H-6″), 5.21 (2H, m, H-2′, H-2″); partial ^{13}C NMR ($CDCl_3$, 100 MHz) δ 16.2, 17.7, 25.7, 26.2, 32.7, 39.6, 105.7, 121.4, 123.4, 132.2, 140.7, 204.2.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.jnatprod.6b00347.

^{1}H and ^{13}C NMR spectra of compounds **5**−**9**, **11**−**14**, and **16**−**18**, ^{1}H NMR spectra of **1** and **5**, and HPLC chromatograms of **11**−**13** and **16**−**18** (PDF)

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Notes

The authors declare no competing financial interest.

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