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Onoceranoid-Type Triterpenoids from *Lansium domesticum*

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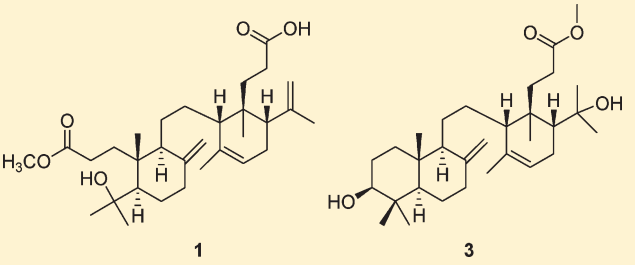
Supporting Information

ABSTRACT: A rare class of onoceranoid-type triterpenoids, represented by lamesticumin A (**1**), the ethanolysis product of lamesticumin A (**2**), lamesticumins B−F (**3**−**7**), lansic acid 3-ethyl ester (**8**), and ethyl lansiolate (**9**), along with four known analogues were isolated from the twigs of *Lansium domesticum*. Their structures were elucidated on the basis of extensive spectroscopic analysis, and the absolute configuration of C-21 in compound **7** was assigned by using Snatzke's method. Compounds **1**−**9** exhibited moderate antibacterial activity against Gram-positive bacteria.



The plant of *Lansium domesticum* Corr. (Meliaceae family), a fruit-bearing tree, grows widely in southeastern Asia. Previous chemical studies on its fruit peel have led to the isolation of a number of onoceranoid-type triterpenoids, of which some show significant inhibition of the leukotriene D4-induced contraction of guinea pig ileum[1] and toxicity against brine shrimp (*Artemia salina*).[2] Methyl lansate[3,4] and lansiolic acid[5] isolated from this plant have been synthesized.[3−5] A chemical study on the plant of *L. domesticum* growing in China has not been documented hitherto. In the current study, nine new onoceranoid-type triterpenoids, namely, lamesticumin A (**1**), the ethanolysis product of lamesticumin A (**2**), lamesticumins B−F (**3**−**7**), lansic acid 3-ethyl ester (**8**), and ethyl lansiolate (**9**), along with four known analogues, were isolated from the ethanolic extract of twigs of *L. domesticum*. Compounds **2**, **8**, and **9** are likely artifacts formed in the extraction process by involving ethanol as the solvent. Compounds **1**−**9** exhibited moderate antibacterial activity against Gram-positive bacteria. Herein we present the isolation, structural elucidation, and antimicrobial evaluation of these isolates.

RESULTS AND DISCUSSION

Compound **1** was obtained as a white, amorphous powder. Its HRESIMS displayed a sodiated molecular ion peak at m/z 525.3559 $[M + Na]^+$, corresponding to the molecular formula of $C_{31}H_{50}O_5$ with seven degrees of unsaturation. The IR absorption bands showed the presence of hydroxy (3433 cm^{-1}), carbonyl (1716 cm^{-1}), and olefinic (1645 cm^{-1}) functional groups. The ^{13}C NMR data (Table 1) with DEPT experiments revealed the presence of seven methyls (one *O*-methyl), 11 methylenes (two olefinic), five methines (one olefinic), and eight quaternary carbons (two carbonyls, three olefinic, and one oxygenated). The two terminal double bonds, a trisubstituted double bond, and two carbonyls accounted for five of the seven degrees of unsaturation, and the remaining two thus required **1** being bicyclic. The above analysis suggested that compound **1** was an onoceranoid-type triterpenoid featuring a 3,4-*seco* and 21,22-*seco* skeleton.[3,5] The structure of **1** was further elucidated by analysis of 2D NMR spectra, especially the HMBC (Figure 1A), in which a terminal $\Delta^{8(26)}$ double bond (δ_H 4.54 and 4.86; δ_C 148.2 and 106.9) (Tables 1 and 2) was located by the correlation networks of H_2-26/C-7 and C-9, H_2-7/C-8, and H-9/C-8; the other terminal $\Delta^{22(29)}$ double bond (δ_H 4.78 and 4.81; δ_C 147.7 and 113.9) was assigned by the correlations of H_2-29/C-30 and C-17, H-17/C-22, and Me-30/C-22. The trisubstituted Δ^{14} double bond (δ_H 5.37; δ_C 136.1 and 121.9) was placed by the correlations of Me-27/C-14 and C-15, H_2-16/C-14 and C-15, and H-15/C-13. The carboxy group (δ_C 177.7) was attached to C-20 by the HMBC correlation between H_2-20 and C-21. The attachment of the only methoxycarbonyl group at C-2 was indicated by the HMBC correlations from H_2-2 and OCH_3 to C-3 (δ_C 176.6). The carbon resonance at δ_C 75.5 was assigned to C-4 bearing a hydroxy group based on the HMBC correlations between Me-23(Me-24) and C-4.

The relative configuration of **1** was defined by analysis of its ROESY spectrum (Figure 1B). The ROESY cross-peaks of Me-25/Me-24 and Me-25/H-11a indicated that Me-25 and CH_2-11 moieties were cofacial and randomly assigned in a β-orientation. In consequence, the ROESY correlation between H-5 and H-9 suggested that they were α-oriented. By the same token, the Me-28 and CH_2-12 moieties were assigned in an α-orientation from the ROESY correlation between Me-28 and H-12b, while the H-13, H-17, and CH_2-19−C-21 moieties were β-oriented based on the correlations of H-13/H-17 and

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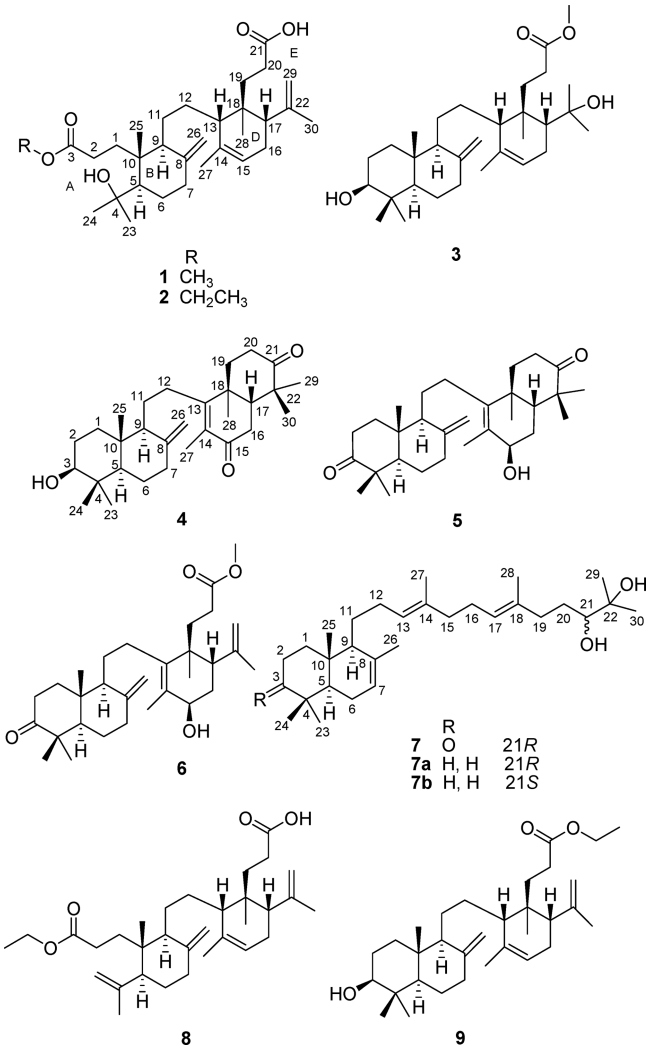

H-17/H-19b. Thus, the structure of compound **1**, lamesticumin A, was assigned as shown.

Compound **2** was obtained as a white, amorphous powder. Its HRESIMS gave a molecular formula of $C_{32}H_{52}O_5$. Its NMR spectra (Table 1 and Experimental Section) were similar to those of **1**, except for the presence of an *O*-ethyl [δ_H 1.25 (t, 7.3, 3H) and 4.12 (q, 7.3, 2H); δ_C 14.2 and 60.7] replacing the *O*-methyl group of **1**. Thus, the structure of **2** was elucidated as the ethanolysis product of lamesticumin A.

Compound **3**, a white, amorphous powder, gave a sodiated molecular ion peak in the HRESIMS at *m/z* 511.3768 [M + Na]$^+$ (calcd for $C_{31}H_{52}NaO_4$, 511.3763), corresponding to the molecular formula of $C_{31}H_{52}O_4$. The IR absorption bands showed the presence of hydroxy (3439 cm^{-1}), carbonyl (1722 cm^{-1}), and olefinic (1643 cm^{-1}) functionalities. The ^{1}H and ^{13}C NMR data (Tables 1 and 2) showed the presence of seven methyls, a methoxy group, a terminal double bond, a trisubstituted double bond, a carbonyl, an oxygenated methine, and an oxygenated quaternary carbon. These data revealed that **3** was also an onoceranoid-type triterpenoid featuring a 21,22-*seco* skeleton and shared the same A, B, and D rings with methyl lansiolate[1] based on the analysis of its HMBC spectrum (Figure S1, Supporting Information). The main difference was the presence of one more oxygenated quaternary carbon at δ_C 75.1 in the opened E ring of **3**, with the concomitant absence

Table 1. ^{13}C NMR Spectroscopic Data of Compounds 1−9[a]

carbon	1	2	3	4	5	6	7	8	9
1	33.5	33.5	37.0	37.1	37.7	37.6	38.0	33.1	38.1
2	28.5	28.7	28.0	27.8	34.7	34.7	34.6	29.2	28.0
3	176.6	176.2	78.8	78.6	216.6	216.8	217.3	175.4	78.8
4	75.5	75.5	39.1	39.1	47.8	47.8	47.4	147.1	39.4
5	52.0	52.0	54.5	54.6	55.2	55.2	51.3	50.7	54.5
6	26.0	25.9	24.0	24.0	25.2	25.2	24.0	26.6	24.0
7	38.0	38.0	38.1	38.2	37.9	37.9	121.7	38.0	37.0
8	148.2	148.2	148.4	147.8	147.4	147.4	135.6	147.9	148.2
9	52.7	52.6	58.9	58.6	58.0	57.8	52.9	50.9	58.3
10	43.3	43.3	39.5	39.7	39.5	39.6	36.4	41.2	39.1
11	29.0	29.2	26.6	22.9	24.2	24.8	27.3	29.2	25.8
12	27.4	27.3	25.9	29.7	28.1	33.1	29.7	27.3	27.1
13	48.2	48.3	49.1	166.1	144.2	141.2	124.4	48.0	48.2
14	136.1	136.1	136.1	130.7	128.1	131.3	134.9	136.1	136.0
15	121.9	121.8	121.6	198.9	70.0	69.2	39.6	121.7	121.6
16	29.4	29.4	27.5	35.3	29.4	28.1	26.4	29.3	29.5
17	49.0	49.0	48.8	49.5	45.0	39.9	124.9	48.6	49.0
18	38.9	38.9	40.0	40.2	39.2	42.7	135.3	38.9	38.6
19	32.3	32.4	33.0	34.9	35.0	31.9	36.8	32.7	32.7
20	28.4	28.4	29.3	34.4	34.3	29.6	29.7	28.6	29.1
21	177.7	177.9	175.4	214.7	217.1	174.4	78.2	178.7	174.1
22	147.7	147.7	75.1	47.0	46.6	146.2	73.0	147.7	147.7
23	34.7	34.6	28.3	28.3	26.0	26.0	22.1	113.6	28.3
24	27.1	27.1	15.4	15.3	21.6	21.6	24.9	23.5	15.4
25	18.7	18.7	14.6	14.5	14.1	14.1	13.2	17.6	14.6
26	106.9	106.9	106.7	106.6	107.3	107.5	22.0	107.5	106.8
27	23.5	23.5	22.8	11.8	18.0	18.4	16.1	23.5	22.9
28	16.2	16.2	17.8	17.7	18.3	22.2	15.9	16.7	16.4
29	113.9	113.9	34.0	21.3	21.1	115.1	23.3	113.9	113.8
30	22.8	22.8	26.4	25.3	26.7	22.9	26.3	23.0	22.9
OMe-3	51.9								
OMe-21			51.5			51.6			
OEt-3		60.7						60.9	
		14.2						14.1	
OEt-21									60.2
									14.3

[a] Data were measured in $CDCl_3$ at 100 MHz.

of proton and carbon resonances of the $\Delta^{22(29)}$ terminal double bond in methyl lansiolate,[1] indicating that a hydroxy group was located at C-22 of **3**. This analysis was supported by the upfield shift of H_3-30 ($\Delta\delta_H$ 0.45 ppm) and downfield shift of C-30 ($\Delta\delta_C$ 3.5 ppm) as compared with those of methyl lansiolate.[1] The HMBC correlations of H-17/C-22 and Me-29 (or Me-30)/C-22 (Figure S1, Supporting Information) confirmed this assignment. The relative configuration of **3** was established to be identical with that of methyl lansiolate by the analysis of its ROESY spectrum (Supporting Information), as well as their similar NMR patterns.[1] Thus, the chemical structure of **3** (lamesticumin B) was determined as shown.

Compound **4**, a white, amorphous powder, gave a molecular formula of $C_{30}H_{46}O_3$ as determined by the sodiated molecular ion peak at *m/z* 477.3335 [M + Na]$^+$ (calcd for $C_{30}H_{46}NaO_3$, 477.3345) in the HRESIMS. The UV absorption band at 249 nm (log ε 3.53) implied the presence of α,β-unsaturated carbonyl

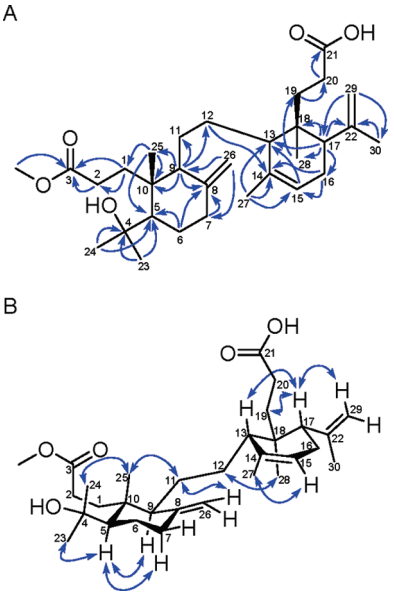

Figure 1. Selected HMBC (H→C) and ROESY (H↔H) correlations of **1**.

group. The IR absorption bands indicated the presence of hydroxy (3446 cm^{-1}), carbonyl (1709 cm^{-1}), and α,β-unsaturated carbonyl (1662 and 1608 cm^{-1}) functional groups. Its ^1H and ^{13}C NMR spectra (Tables 1 and 2) displayed resonances assignable to seven methyls, an oxygenated methine, a terminal double bond, a keto carbonyl, and an α,β-unsaturated keto moiety [δ_C 130.7, 166.1, and 198.9]. The aforementioned data displayed the characteristic features of an onoceranoid-type triterpenoid.[2] The structure of **4** was further defined by analysis of 2D NMR spectra, especially the HMBC (Figure S1, Supporting Information). The keto carbonyl group at δ_C 214.7 was assigned to C-21 by HMBC correlations of Me-29 (or Me-30)/C-21 and H_2-20/C-21. The only oxygenated methine bearing a hydroxy group was located at C-3 by the HMBC correlations of Me-23 (or Me-24)/C-3 and H_2-1/C-3. The $\Delta^{8(26)}$ terminal double bond was assigned by the HMBC correlations from H_2-26 to C-7, C-8, and C-9. This analysis further indicated that compound **4** was an analogue of 3β-hydroxyonocera-8(26),14-dien-21-one,[2] with the main differences being due to the presence of an α,β-unsaturated keto group in the D ring. The remaining keto group at C-15 (δ_C 198.9), conjugated with the Δ^{13} double bond, was assigned on the basis of the multiple HMBC correlations of Me-27/C-13, C-14, and C-15, Me-28/C-13, H_2-12/C-13 and C-14, and H_2-16/C-15 (Figure S1, Supporting Information). The carbon signals of C-12, C-16, and C-18 downfield shifted, respectively, to δ_C 29.7, 35.3, and 40.2 as compared to those of its structural analogues due to the strong deshielding effect of the α,β-unsaturated keto group.[2] The relative configuration of **4** was established by a ROESY experiment (Supporting Information), in which Me-28 was assigned an α-orientation by the correlation between Me-28 and Me-29, while H-17 was assigned a β-orientation by the correlation between H-17 and Me-30. The other stereocenters in the A and B rings were established to be identical to those of 3β-hydroxyonocera-8(26),14-dien-21-one,[2] on the basis of the ROESY spectrum and their similar NMR patterns (Supporting Information). Therefore, the structure of **4** (lamesticumin C) was assigned.

Compound **5** was obtained as a white, amorphous powder, and its molecular formula was determined as $C_{30}H_{46}O_3$ by HRESIMS at m/z 477.3342 $[M + Na]^+$ (calcd for $C_{30}H_{46}NaO_3$, 477.3345). The IR absorption bands showed the presence of hydroxy (3423 cm^{-1}), carbonyl (1705 cm^{-1}), and olefinic (1641 cm^{-1}) functionalities. The ^1H NMR and ^{13}C NMR data (Tables 1 and 2) of **5** showed the presence of seven methyls, a terminal double bond, a tetrasubstituted double bond, an oxygenated methine, and two keto carbonyls. These data also suggested an onoceranoid-type triterpenoid structure for compound **5**.[2] In its HMBC spectrum (Figure 2A), one keto carbonyl at δ_C 216.6 was located at C-3 by the correlations of Me-23 (or Me-24)/C-3 and of H_2-1 and H_2-2/C-3; the other keto carbonyl at δ_C 217.1 was placed at C-21 by correlations of Me-29 (or Me-30)/C-21 and of H_2-19 and H_2-20/C-21. The terminal double bond $\Delta^{8(26)}$ was fixed by the correlations of H_2-26/C-7, C-8, and C-9. The aforementioned analysis suggested that the structure of compound **5** was closely related to α,γ-onoceradienedione,[4,5] with major differences occurring at the D ring. In the HMBC spectrum (Figure 2A), the Δ^{13} double bond (δ_C 144.2 and 128.1) was located on the basis of the multiple HMBC correlations of Me-27/C-13 and C-14, H_2-12/C-13 and C-14, H-17 and Me-28/C-13, and H-15/C-13 and C-14; the hydroxy group was assigned to C-15 (δ_C 70.0) by the HMBC correlations from Me-27, H_2-16, and H-17 to C-15. In the ROESY spectrum (Figure 2B), the key correlations from H-15 to H_2-16 and Me-27 indicated that the OH-15 was β-oriented. The other stereocenters in **5** were established by the ROESY spectrum to be identical to those of **4**. Thus, the structure of **5** (lamesticumin D) was elucidated.

Compound **6**, a white, amorphous powder, gave a molecular formula of $C_{31}H_{48}O_4$ on the basis of an HRESIMS ion at m/z 507.3449 $[M + Na]^+$ (calcd for $C_{31}H_{48}NaO_4$, 507.3450). The IR absorption bands showed the presence of hydroxy (3435 cm^{-1}), carbonyl (1738 and 1707 cm^{-1}), and olefinic (1645 cm^{-1}) functionalities. Comparison of the 1D NMR data of **6** with those of **5** (Tables 1 and 2) suggested that they shared the same A, B, and D rings, and this assignment was confirmed by the HMBC correlations (Figure S2, Supporting Information). Consequently, according to its molecular formula and NMR data, one methoxycarbonyl group and one terminal double bond were indicative of a 21,22-*seco* feature for **6**.[2] The methoxycarbonyl group was attached to C-20 by the HMBC correlations from H_2-19, H_2-20, and OCH_3 to C-21 (δ_C 174.4). The $\Delta^{22(29)}$ terminal double bond was located on the basis of the HMBC correlations from H_2-29 to C-17, C-22, and C-30. The relative configurations of the stereocenters in **6** were identical to those of **5** as assigned by the ROESY spectrum (Figure S2, Supporting Information). Thus, the structure of **6** (lamesticumin E) was established.

Compound **7**, a colorless oil, gave a molecular formula of $C_{30}H_{50}O_3$, as established on the basis of an HRESIMS ion at m/z 481.3640 $[M + Na]^+$ (calcd for $C_{30}H_{50}NaO_3$, 481.3658) requiring six degrees of unsaturation. The IR absorption bands showed the presence of hydroxy (3442 cm^{-1}) and carbonyl (1707 cm^{-1}) groups. The ^{13}C NMR spectrum of **7** (Table 1) revealed the presence of eight methyls, three trisubstituted double bonds, an oxygenated methine, an oxygenated quaternary carbon, and a keto carbonyl carbon. These functionalities accounted for four of the six degrees of unsaturation, and the remaining two degrees of unsaturation thus required **7** being bicyclic. The aforementioned data revealed that compound **7** was

Table 2. ^{1}H NMR Spectroscopic Data of Compounds 1 and 3−7[a]

	1	3	4	5	6	7
proton position	(mult., *J* in Hz)	(mult., *J* in Hz)	(mult., *J* in Hz)	(mult., *J* in Hz)	(mult., *J* in Hz)	(mult., *J* in Hz)
1α	1.79, m	1.17, m	1.16, m	1.55, m	1.59, m	1.46, m
1*β*	2.54, m	1.79, m	1.74, m	2.46, m	2.45, m	2.14, m
2α	2.34, 2H, m	1.61, m	1.59, m	2.42, m	2.40, m	2.24, m
2*β*		1.70, m	1.72, m	2.63, m	2.62, m	2.71, m
3		3.26, dd (11.4, 4.5)	3.25, dd (11.8, 4.2)			
5	1.66, m	1.09, m	1.09, m	1.58, m	1.61, m	1.56, m
6α	1.39, m	1.38, m	1.78, 2H, m	1.69, 2H, m	1.71, 2H, m	1.90, m
6*β*	1.70, m	1.78, m				2.06, m
7α	1.95, dt (12.9, 4.3)	1.96, m	2.00, m	2.02, m	2.04, 2H, m	5.41, t (1.8)
7*β*	2.30, m	2.39, ddd (12.8, 4.2, 2.4)	2.45, m	2.47, m		
9	1.80, m	1.57, m	1.63, m	1.66, m	1.68, m	1.66, m
11a	2.16, m	1.19, m	1.48, m	1.43, 2H, m	1.50, 2H, m	1.30, m
11b	2.66, m	1.38, m	1.60, m			1.46, m
12a	1.44, 2H, m	1.39, m	1.25, m	1.61, m	1.66, m	1.99, m
12b		1.65, m	2.53, m	2.29, m	1.84, m	2.16, m
13	1.85, brs	1.83, m				5.10, t (7.0)
15	5.37, brs	5.33, brd (3.8)		4.01, brs	3.94, brs	2.00, 2H, m
16α	1.85, brs	1.84, m	2.50, 2H, m	1.72, 2H, m	1.74, m	2.08, 2H, m
16*β*	1.25, d (2.0)	2.02, m				
17	2.23, m	1.69, dd (12.5, 2.7)	2.15, m	1.98, m	2.53, dd (13.4, 2.7)	5.16, t (6.3)
19α	1.68, m	1.78, m	1.87, m	1.74, m	1.72, m	2.04, m
19*β*	1.77, brs	2.26, m	2.17, m	1.96, m	1.94, m	2.21, m
20α	1.73, m	2.18, m	2.53, m	2.54, 2H, m	1.93, m	1.42, m
20*β*	2.34, m	2.72, m	2.74, ddd (18.9, 12.5, 6.5)		2.29, brt (11.4)	1.62, m
21						3.50, dd (10.4, 1.8)
23	1.27, 3H, s	0.99, 3H, s	1.00, 3H, s	1.09, 3H, s	1.10, 3H, s	1.08, 3H, s
24	1.19, 3H, s	0.77, 3H, s	0.77, 3H, s	1.02, 3H, s	1.02, 3H, s	1.04, 3H, s
25	0.82, 3H, s	0.68, 3H, s	0.67, 3H, s	0.84, 3H, s	0.84, 3H, s	0.97, 3H, s
26a	4.86, brs	4.82, brs	4.91, brs	4.94, brs	4.93, brs	1.72, 3H, s
26b	4.54, brs	4.55, brs	4.62, brs	4.69, brs	4.66, d (1.1)	
27	1.77, 3H, s	1.74, 3H, s	1.80, 3H, s	1.79, 3H, s	1.78, 3H, s	1.59, 3H, s
28	0.80, 3H, s	0.85, 3H, s	1.24, 3H, s	0.99, 3H, s	0.93, 3H, s	1.60, 3H, s
29a	4.81, brs	1.20, 3H, s	1.12, 3H, s	1.06, 3H, s	4.97, brs	1.15, 3H, s
29b	4.78, brs				4.71, t (1.6)	
30	1.77, 3H, s	1.29, 3H, s	1.10, 3H, s	1.13, 3H, s	1.76, 3H, s	1.19, 3H, s
OMe-3	3.66, 3H, s					
OMe-21		3.66, 3H, s			3.63, 3H, s	

[a] Data were measured in $CDCl_3$ at 400 MHz.

likely an oxygenated derivative of two synthetic products, **7a** and **7b**, a pair of C-21 epimers showing nearly identical NMR data.[6] The keto carbonyl was assigned to C-3 by the multiple HMBC correlations from H_2-1, H_2-2, Me-23, Me-24, and H-5 to C-3 (Figure S2, Supporting Information). The carbon signals of C-2 and C-4 shifted downfield by 15.4 and 14.3 ppm, respectively, as compared to those of **7a** and **7b**.[6] The relative configuration of the bicyclic core of **7** was assigned by the ROESY spectrum (Supporting Information), in which the correlations of H-5/H-9 and Me-25/Me-24 were observed.

The absolute configuration of C-21 in **7** was assigned by using the *in situ* dimolybdenum CD method.[7−9] Theoretically, the formation of a chiral metal complex required the vicinal diol in a *gauche* arrangement and would give two diastereomorphous structures (Figure 3A), in which the favored conformation prefers the bulkier groups in a pseudo-equatorial position away from the core of the metal complex. According to the empirical rule proposed by Snatzke,[7] the observed induced CD curve at around 300 nm showing the same sign with the O−C−C−O torsion angle in the favored conformation will allow the assignment of the absolute configuration. The metal complex of compound **7** in DMSO gave a significant induced CD spectrum (ICD) (Figure 3B), in which the negative Cotton effect at 295 nm permitted the assignment of a 21*R*-configuration for **7**.[7−9] Therefore, the structure of **7** was assigned as shown and was named lamesticumin F.

Compound **8**, a white, amorphous powder, displayed a molecular formula of $C_{32}H_{50}O_4$ as determined by the HRESIMS

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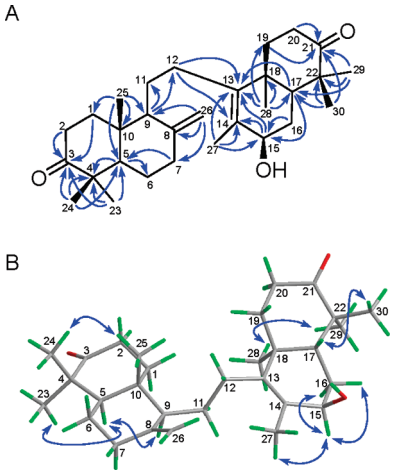

Figure 2. Selected HMBC (H→C) and ROESY (H↔H) correlations of **5**.

spectrum. The NMR data (Table 1 and Experimental Section) of compound **8** showed many similarities to those of lansic acid,[1,3−5] except for the presence of the typical proton and carbon signals of an ethoxycarbonyl group [δ_H 1.24 (t, 7.0, 3H) and 4.13 (q, 7.0, 2H); δ_C 14.1, 60.9, and 175.4] replacing the C-3 carboxy group of lansic acid. Thus, the C-3 signal was upfield shifted and the C-1 and C-2 signals were downfield shifted as compared to those of lansic acid.[1] The carbon resonances in the D ring and opened E ring of **8** were similar to those of **1** and **2**, confirming that the ethoxycarbonyl group was located at C-2. The structure of **8** was thus assigned as lansic acid 3-ethyl ester.

Compound **9** was obtained as a white, amorphous powder and gave a molecular formula of $C_{32}H_{52}O_3$ as assigned by the HRESIMS spectrum. Its NMR data (Table 1 and Experimental Section) showed similarity to those of methyl lansiolate,[1] except for the existence of a typical *O*-ethyl [δ_H 1.25 (t, 7.1, 3H) and 4.10 (q, 7.1, 2H), δ_C 14.3 and 60.2] replacing the *O*-methyl group of methyl lansiolate.[1] Therefore, the structure of compound **9** was assigned and named ethyl lansiolate.

Three *O*-ethyl-bearing compounds, **2**, **8**, and **9**, are likely artifacts formed in the extraction process by involving EtOH as the solvent.

Four known analogues, lansic acid,[1,3−5] lansiolic acid,[1,5] methyl lansiolate,[1] and α,γ-onoceradienediol,[5,10] were also obtained and were identified on the basis of ^{1}H NMR, ^{13}C NMR, and ESIMS data.

Compounds **1**−**9** were evaluated for antibacterial activity against 11 bacteria *in vitro* by the microdilution assay.[11,12] A natural antimicrobial agent, magnolol,[13] was used as the positive control in this test. The antibacterial MICs of compounds **1**−**9** are listed in Table 3. Compounds **1**−**9** all showed moderate antibacterial activity against Gram-positive bacteria.

EXPERIMENTAL SECTION

General Experimental Procedures. Optical rotations were measured on a Perkin-Elmer 341 polarimeter at room temperature. The CD spectrum was obtained on a Jasco 810 spectrometer. UV spectra were recorded on a Shimadzu UV-2550 spectrophotometer. IR spectra were recorded on a Perkin-Elmer 577 IR spectrometer. NMR spectra were obtained on a Bruker AM-400 NMR spectrometer with TMS as internal standard. HRESIMS was carried out on a Bruker

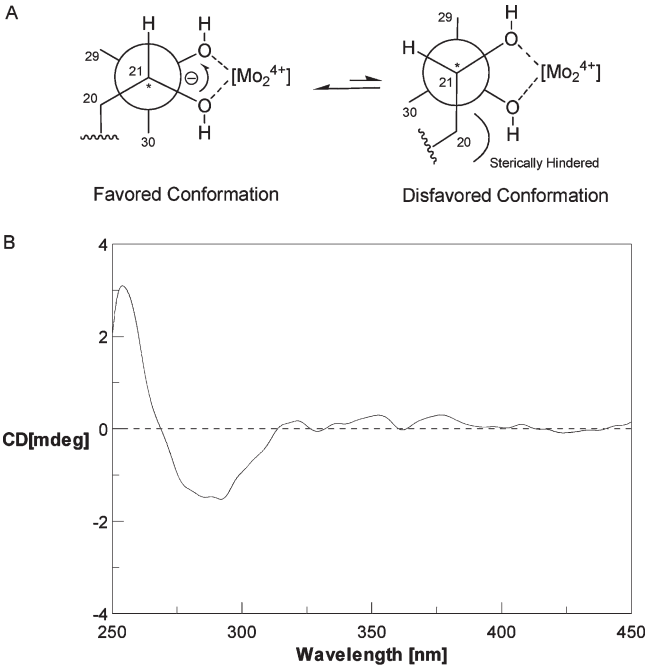

Figure 3. (A) Conformations of the Mo_2^{4+} complex of **7**. (B) ICD spectrum of the Mo_2^{4+} complex of **7** in DMSO.

Daltonics micrOTOFQ II and a Waters Q-TOF Ultima mass spectrometer. Semipreparative HPLC was carried out on a Waters 515 pump with a Waters 2487 detector (254 nm) and a YMC-Pack ODS-A column (250 × 10 mm, S-5 μm, 12 nm). Silica gel (300−400 mesh), C_{18} reversed-phase silica gel (250 mesh, Merck), and MCI gel (CHP20P, a kind of refined polystyrene-based separation stuffing, 75−150 μM, Mitsubishi Chemical Industries, Ltd.) were used for column chromatography, and precoated silica gel GF254 plates (Qingdao Marine Chemical Plant, Qingdao, People's Republic of China) were used for TLC. All solvents used were of analytical grade (Shanghai Chemical Plant, Shanghai, People's Republic of China).

Plant Material. The twigs of *Lansium domesticum* were collected in August 2005 from Xishuangbanna, Mengla County, Yunnan Province, China, and were authenticated by Professor You-Kai Xu of Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences. A voucher specimen (access number: Ldom-2005-1Y) has been deposited in Shanghai Institute of Materia Medica.

Extraction and Isolation. The air-dried powder of twigs of *L. domesticum* (5.2 kg) was extracted three times with 95% EtOH (each 25 L, three days) at room temperature to give an ethanolic extract (240 g), which was partitioned between EtOAc and H_2O to obtain the EtOAc-soluble fraction (175 g). The EtOAc-soluble fraction was subjected to passage over MCI gel column chromatography (MeOH/H_2O, 50/50 to 90/10) to produce three fractions, A−C. Fraction C (100 g) was chromatographed over a silica gel column, eluted with petroleum ether/acetone (10/1 to 1/2), to give five major fractions, C1−C5. Fraction C2 was subjected to silica gel column chromatography, eluted with $CHCl_3$/MeOH (pure $CHCl_3$ to 40/1), to give two major fractions, C2A and C2B. Fraction C2A was purified by semipreparative HPLC (90% MeOH in H_2O, 11.0 min) to give compound **6** (4 mg). Fraction C2B was subjected to a reversed-phase C_{18} silica gel column eluted with MeOH/H_2O (80/20 to 95/5) to give three fractions, each of which was purified by semipreparative HPLC with 100% MeOH (10.0, 8.0, 10.3 min) to yield compounds **5** (6 mg), **7** (10 mg), and α,γ-onoceradienediol (20 mg), respectively. Fraction C3 was chromatographed over a reversed-phase C_{18} silica gel column eluted with MeOH/H_2O (60/40 to

Table 3. Antibacterial Activities of Compounds 1−9

	MIC (μg/mL)[a]									
bacteria	A	1	2	3	4	5	6	7	8	9
S. aureus	25	6.25	>50	6.25	6.25	>50	>50	>50	50	6.25
S. epidermidis	12.5	12.5	12.5	12.5	12.5	>50	>50	>50	12.5	12.5
M. luteus	12.5	6.25	>50	3.12	6.25	>50	>50	>50	>50	6.25
B. subtilis	12.5	3.12	12.5	3.12	3.12	6.25	12.5	12.5	12.5	3.12
M. pyogenes	25	3.12	50	3.12	3.12	>50	6.25	>50	12.5	3.12
B. cereus	12.5	3.12	3.12	3.12	3.12	3.12	3.12	3.12	3.12	3.12
E. coli	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50
S. flexneri	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50
P. aeruginosa	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50
S. marcescens	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50
A. faecalis	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50

[a] MIC was defined as the lowest concentration that inhibited visible growth. All the tests were conducted in triplicate. The MIC > 50 μg/mL was defined to be inactive. **A**, representing magnolol, was applied as the positive control.

90/10) to give five major fractions, C3A−C3E. Fraction C3A was purified by silica gel column chromatography, eluted with petroleum ether/$CHCl_3$ (1/2 to 1/3), to give compound **4** (3 mg). Fraction C3B was subjected to a Sephadex LH-20 column eluted with MeOH to give two fractions, each of which was purified by a semipreparative HPLC (100% MeOH, 8.3 min, 8.0 min) to yield compounds **1** (3 mg) and **2** (10 mg), respectively. Fraction C3C was subjected to silica gel column chromatography, eluted with $CHCl_3$/MeOH (200/1 to 100/1), to give lansic acid (30 mg) and another three fractions, C3C1−C3C3. Fraction C3C1 was further purified to give compound **8** (4 mg) by reversed-phase C_{18} silica gel column chromatography eluted with MeOH/H_2O (80/10 to 90/1). Fraction C3C2 was subjected to semipreparative HPLC eluted with 100% MeCN (at 17.2 min) to give lansiolic acid (15 mg). Fraction C3C3 was purified to give compound **3** (10 mg) by semipreparative HPLC (100% MeOH, 9.3 min). Fraction C3E was chromatographed over a silica gel column eluted with petroleum ether/EtOAc (6/1 to 4/1) to give two major components, each of which was purified by semipreparative HPLC (mobile phase, 100% MeOH) to give methyl lansiolate (5 mg) and compound **9** (5 mg), respectively.

*Lamesticumin A (**1**):* white, amorphous powder; $[\alpha]^{17}_D$ +20 (*c* 0.06, MeOH); IR (KBr) ν_{max} 3433, 2926, 2872, 1716, 1645, 1441, 1383, 1200, 889 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 525.3559 $[M + Na]^+$ (calcd for $C_{31}H_{50}NaO_5$, 525.3556).

*Ethanolysis product of lamesticumin A (**2**):* white, amorphous powder; $[\alpha]^{17}_D$ +30 (*c* 0.05, MeOH); IR (KBr) ν_{max} 3442, 2966, 1736, 1713, 1647, 1450, 1379, 1290, 1182, 1155, 893 cm^{-1}; ^1H NMR ($CDCl_3$) δ_H 5.37 (1H, brs, H-15), 4.85 and 4.54 (each 1H, brs, H_2-26), 4.81 and 4.77 (each 1H, brs, H_2-29), 4.12 and 1.25 (2H, 3H, q, t, *J* = 7.3, OEt-3), 2.66 and 2.16 (each 1H, m, H_2-11), 2.54 and 1.79 (each 1H, m, H_2-1), 2.34 (2H, m, H_2-2), 2.33 and 1.73 (each 1H, m, H_2-10), 2.30 and 1.95 (each 1H, m, H_2-7), 2.23 (1H, m, H-17), 1.85 (1H, brs, H-13), 1.84 and 1.25 (each 1H, m, H_2-16), 1.80 (1H, m, H-9), 1.77 (6H, s, H_3-27 and H_3-30), 1.76 and 1.68 (each 1H, m, H_2-19), 1.70 and 1.39 (each 1H, m, H_2-6), 1.66 (1H, m, H-5), 1.44 (2H, m, H_2-12), 1.27 (3H, s, H_3-23), 1.19 (3H, s, H_3-24), 0.82 (3H, s, H_3-25), and 0.80 (3H, s, H_3-28); ^{13}C NMR data, see Table 1; HRESIMS *m/z* 539.3709 $[M + Na]^+$ (calcd for $C_{32}H_{52}NaO_5$, 539.3712).

*Lamesticumin B (**3**):* white, amorphous powder; $[\alpha]^{17}_D$ +10 (*c* 0.12, MeOH); IR (KBr) ν_{max} 3439, 2943, 1722, 1643, 1439, 1385, 1175, 1032, 887 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 511.3768 $[M + Na]^+$ (calcd for $C_{31}H_{52}NaO_4$, 511.3763).

*Lamesticumin C (**4**):* white, amorphous powder; $[\alpha]^{17}_D$ +34 (*c* 0.24, MeOH); UV (MeOH) λ_{max} (log ε) 249 (3.53) nm; IR (KBr) ν_{max} 3446, 2929, 2850, 1709, 1662, 1608, 1458, 1385, 1338, 1034, 887 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 477.3335 $[M + Na]^+$ (calcd for $C_{30}H_{46}NaO_3$, 477.3345).

*Lamesticumin D (**5**):* white, amorphous powder; $[\alpha]^{17}_D$ +86 (*c* 0.15, MeOH); IR (KBr) ν_{max} 3423, 2939, 1705, 1641, 1458, 1385, 1051, 1003, 885 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 477.3342 $[M + Na]^+$ (calcd for $C_{30}H_{46}NaO_3$, 477.3345).

*Lamesticumin E (**6**):* white, amorphous powder; $[\alpha]^{17}_D$ +54 (*c* 0.10, MeOH); IR (KBr) ν_{max} 3435, 2943, 1738, 1707, 1645, 1439, 1385, 1057, 893 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 507.3449 $[M + Na]^+$ (calcd for $C_{31}H_{48}NaO_4$, 507.3450).

*Lamesticumin F (**7**):* colorless oil; $[\alpha]^{17}_D$ −13.4 (*c* 0.70, MeOH); IR (KBr) ν_{max} 3442, 2931, 1707, 1452, 1383, 1163, 1076 cm^{-1}; ^1H and ^{13}C NMR data, see Tables 1 and 2; HRESIMS *m/z* 481.3640 $[M + Na]^+$ (calcd for $C_{30}H_{50}NaO_3$, 481.3658).

*Lansic acid 3-ethyl ester (**8**):* white, amorphous powder; $[\alpha]^{17}_D$ +28 (*c* 0.05, MeOH); IR (KBr) ν_{max} 3423, 2931, 1736, 1709, 1643, 1381, 1178, 893 cm^{-1}; ^1H NMR ($CDCl_3$) δ_H 5.37 (1H, brs, H-15), 4.90 and 4.60 (each 1H, brs, H_2-26), 4.85 and 4.70 (each 1H, brs, H_2-23), 4.81 and 4.78 (each 1H, brs, H_2-29), 4.13 and 1.24 (2H, 3H, q, t, *J* = 7.0, OEt-3), 2.36 and 2.18 (each 1H, m, H_2-11), 2.35 (2H, m, H_2-2), 2.34 and 2.00 (each 1H, m, H_2-7), 2.32 and 1.67 (each 1H, m, H_2-1), 2.24 (1H, m, H-5), 2.21 (1H, m, H-17), 1.83 (1H, m, H-13), 1.82 (2H, m, H_2-16), 1.80 (1H, m, H-9), 1.77 (6H, s, H_3-27 and H_3-30), 1.73 (3H, s, H_3-24), 1.70 (2H, m, H_2-19), 1.65 and 1.37 (each 1H, m, H_2-20), 1.64 and 1.33 (each 1H, m, H_2-6), 1.43 and 1.22 (each 1H, m, H_2-12), 0.83 (3H, s, H_3-28), and 0.71 (3H, s, H_3-25); ^{13}C NMR data, see Table 1; HRESIMS *m/z* 521.3605 $[M + Na]^+$ (calcd for $C_{32}H_{50}NaO_4$, 521.3607).

*Ethyl lansiolate (**9**):* white, amorphous powder; $[\alpha]^{17}_D$ +27 (*c* 0.06, MeOH); IR (KBr) ν_{max} 3435, 2941, 1738, 1641, 1383, 1180, 1032, 891 cm^{-1}; ^1H NMR ($CDCl_3$) δ_H 5.36 (1H, t, *J* = 1.4, H-15), 4.83 and 4.55 (each 1H, brs, H_2-26), 4.81 and 4.70 (each 1H, brs, H_2-29), 4.10 and 1.25 (2H, 3H, q, t, *J* = 7.1, OEt-21), 3.25 (1H, brd, *J* = 11.4, H-3), 2.21 (1H, dd, *J* = 13.4 and 2.7, H-17), 2.15 (2H, m, H_2-7), 1.82 (1H, m, H-13), 1.81 (2H, m, H_2-16), 1.81 and 1.17 (each 1H, m, H_2-1), 1.78 (3H, s, H_3-27), 1.76 and 1.62 (each 1H, m, H_2-2), 1.73 (3H, s, H_3-30), 1.72 (2H, m, H_2-19), 1.65 and 1.33 (each 1H, m, H_2-20), 1.64 and 1.40 (each 1H, m, H_2-6), 1.61 (1H, m, H-9), 1.59 and 1.28 (each 1H, m, H_2-11), 1.39 and 1.20 (each 1H, m, H_2-12), 1.17 (1H, m, H-5), 1.02 (3H, s, H_3-24), 0.98 (3H, s, H_3-23), 0.80 (3H, s, H_3-28), 0.76 (3H, s, H_3-25); ^{13}C NMR data, see Table 1; HRESIMS *m/z* 507.3821 $[M + Na]^+$ (calcd for $C_{32}H_{52}NaO_3$, 507.3814).

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Determination of the Absolute Configuration of C-21 in 7. According to the published procedure,[8] a mixture of compound **7** (1.5 mg) and $Mo_2(OAc)_4$ (1.4 mg) was prepared for CD measurement. The mixture was kept for 30 min to form a stable chiral metal complex, after which the CD spectrum was recorded. The observed sign of the diagnostic ICD curve at around 300 nm was correlated to the absolute configuration of C-21 in **7**.[7−9]

Antibacterial Test. The *in vitro* antibacterial activities against *Staphylococcus aureus* ATCC 25923, *Staph. epidermidis* ATCC 12228, *Micrococcus luteus* ATCC 9341, *Bacillus subtilis* ATCC 6633, *Micrococcus pyogenes* (a clinical isolate), *Bacillus cereus* (a clinical isolate), *Escherichia coli* ATCC 25922, *Shigella flexneri* ATCC 12022, *Pseudomonas aeruginosa* ATCC 14502, *Serratia marcescens* ATCC 25419, and *Alcaligenes faecalis* ATCC 8750 were assessed by following the published methods.[11] Briefly, the microbial cells were suspended in Mueller Hinton broth to form a final density of 5×10^{-5}−10^{-6} cfu/mL and incubated at 37 °C for 18 h under aerobic conditions with the respective compounds and positive control, which have been dissolved in DMSO. The blank controls of microbial culture were incubated with limited DMSO under the same conditions. DMSO was determined not to be toxic at a limited amount, 1% at most in the solution, under the experimental conditions.

■ ASSOCIATED CONTENT

Supporting Information. IR, HRESIMS, and 1D and 2D NMR spectra of compounds **1**−**9**. This material is available free of charge via the Internet at http://pubs.acs.org.

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