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ARTICLE *in* JOURNAL OF NATURAL PRODUCTS · DECEMBER 2011

Impact Factor: 3.8 · DOI: 10.1021/np200441h · Source: PubMed

JOURNAL OF NATURAL PRODUCTS

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Proteasome Inhibitors from *Neoboutonia melleri*

Christophe Long,[†] Joséphine Beck,[†] Frédéric Cantagrel,[†] Laurence Marcourt,[†] Laure Vendier,[‡] Bruno David,[†] Fabien Plisson,[†] Fadila Derguini,[†] Isabelle Vandenberghe,[§] Yannick Aussagues,[†] Frédéric Ausseil,[†] Catherine Lavaud,[⊥] François Sautel,[†] and Georges Massiot*[,†]

[†]USR CNRS-Pierre Fabre No. 3388 ETaC, Centre de Recherche et Développement Pierre Fabre, 3 Avenue Hubert Curien, 31035 Toulouse Cedex 01, France

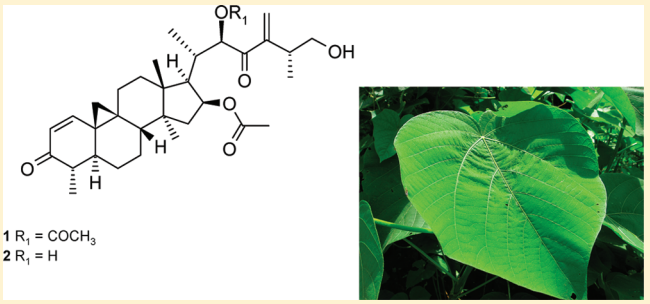
[‡]UPR CNRS No. 8241, Laboratoire de Chimie de Coordination, 205 Route de Narbonne, 31077 Toulouse Cedex 04, France

[§]Institut de Recherche Pierre Fabre, Centre de Recherche en Oncologie Experimentale, Centre de Recherche et Développement Pierre Fabre, 3 Avenue Hubert Curien, 31035 Toulouse Cedex 01, France

[⊥]UMR CNRS-Université Reims Champagne-Ardenne No. 6229, Institut de Chimie Moléculaire de Reims, Laboratoire de Pharmacognosie, 51097 Reims, France

Supporting Information

ABSTRACT: Thirty new cycloartane derivatives (**1**−**3**, **5**−**12**, **14**−**32**) have been isolated from the leaves of *Neoboutonia melleri*. Their novelty stems from the loss of one of the C-4 methyl groups (**1**−**3**, **5**−**12**, **14**−**25**, and **32**) and from the presence of an "extra" carbon atom in the side chain (**1**−**3**, **5**−**12**, **14**−**20**, **26**−**29**, and **30**−**32**). Furthermore, compound **32** possesses a rare triterpene skeleton with the cyclopropane ring fused onto C-1 and C-10, instead of C-9 and C-10. The structures were determined by spectrometric means, chemical correlations, and X-ray crystallography of derivative **1c**. The substitution pattern in ring A, with a cyclopropyl ring conjugated with an α,β-unsaturated carbonyl moiety, confers to the molecule a particular reactivity, giving rise to a formal inversion of the stereochemistry of the cyclopropane ring under UV irradiation. These compounds showed an interesting level of activity on the proteasome pathway, thus motivating their evaluation as possible anticancer agents. The large number of isolated compounds permitted a structure−activity relationship analysis, which showed that the presence of the two enone functions was a requirement for the activity.



For the past decade, proteasome inhibitors have been considered as potentially useful drugs in the treatment of cancer, and this has culminated in the approval of bortezomib for the treatment of multiple myeloma.[1] Epoxomycin, lactacystin, and, the most promising, salinosporamide A, presently in phase 1 clinical trials, are examples of natural products that inhibit the proteasome catalytic functions.[2] In order to find new inhibitors that do not particularly target proteolysis but the whole ubiquitin−proteasome pathway, we have developed an assay based on a human colon cancer cell line that stably expresses a 4-ubiquitin luciferase reporter protein (4Ub-Luc DLD-1).[3] Under normal conditions, the fusion protein (4Ub-Luc) is produced and, due to the ubiquitin tag's presence, it is addressed to and degraded by the proteasome, while inhibitors of the proteasome pathway lead to protein accumulation and luminescence observation.[4]

The assay was applied to a collection of over 12 000 plant extracts and 62 000 pure compounds including natural products and derivatives. An extract from *Neoboutonia melleri* (Muell. Arg.) Prain and two pure compounds isolated from it (**1** and **2**) inhibited the proteasome pathway. The plant was collected in Cameroon, in the late 1980s, following ethnopharmacological observations. Its leaves were locally used to wrap fish, hence, its name Koutencha, "fish leaves" in the Bamum language. *Neoboutonia* is an African tropical genus of the Euphorbiaceae, which, according to the latest classifications, represents numerous varieties combined into three species: *N. melleri*, *N. diaguissensis* Beille, and *N. macrocalyx* Pax.[5] *N. melleri*, a small tree (4−8 m) growing in swamp areas from Cameroon/Angola to Sudan/Mozambique, exhibits alternate, stipulate leaves, broadly ovate or subcircular. It is locally considered as toxic and of interest against diabetes. *N. melleri* was the subject of a single phytochemical investigation,[6] and articles on the genus are rare.[7] Here we describe the isolation and structural characterization of 30 new compounds, evaluation of their biological activity on the proteasome pathway, and discussion of their structure−activity relationship.

RESULTS AND DISCUSSION

The CH_2Cl_2 extract of *N. melleri* was dominated by two compounds, which differed by the presence of an acetyl group and

Received: May 30, 2011
Published: December 14, 2011

dx.doi.org/10.1021/np200441h | *J. Nat. Prod.* 2012, 75, 34−47

Table 1. ^{13}C NMR Data of Compounds 1-8

carbon	1[a]	2[a]	3[b]	4[b]	5[b]	6[b]	7[a]	8[a]
1	153.7, CH	153.9, CH	155.5, CH	155.3, CH	33.5, CH	33.5, CH	154.4, CH	154.6, CH
2	128.3, CH	128.2, CH	128.4, CH	128.5, CH	41.5, CH	41.5, CH	127.4, CH	127.3, CH
3	202.0, C	202.1, C	202.4, C	202.3, C	213.2, C	213.3, C	202.5, C	202.7, C
4	46.9, CH	46.9, CH	47.6, CH	47.5, CH	50.5, CH	50.5, CH	43.8, CH	43.8, CH
5	42.5, CH	42.6, CH	43.6, CH	43.1, CH	47.0, CH	47.0, CH	45.7, CH	45.7, CH
6	23.4, CH_2	23.6, CH_2	24.3, CH_2	23.9, CH_2	26.5, CH_2	26.5, CH_2	65.6, CH	65.6, CH
7	23.5, CH_2	23.6, CH_2	24.3, CH_2	23.8, CH_2	26.2, CH_2	26.2, CH_2	31.9, CH_2	31.9, CH_2
8	44.3, CH	44.7, CH	45.2, CH	43.6, CH	48.6, CH	48.6, CH	40.1, CH	40.2, CH
9	26.0, C	26.1, C	27.2, C	27.4, C	25.4, C	25.4, C	25.3, C	25.4, C
10	31.9, C	31.9, C	32.9, C	33.3, C	30.3, C	30.3, C	29.6, C	29.7, C
11	27.4, CH_2	27.5, CH_2	28.1, CH_2	28.4, CH_2	27.4, CH_2	27.4, CH_2	27.1, CH_2	27.2, CH_2
12	32.1, CH_2	32.1, CH_2	32.9 CH_2	32.8, CH_2	33.3, CH_2	33.5 CH_2	32.3, CH_2	32.3, CH_2
13	47.4, C	47.4, C	46.9, C	44.8, C	46.8, C	46.7, C	46.3, C	46.1, C
14	46.0, C	45.8, C	48.4, C	52.4, C	48.0, C	47.9, C	46.7, C	46.7, C
15	46.0, CH_2	46.2, CH_2	46.7, CH_2	41.7, CH_2	47.6, CH_2	47.8, CH_2	46.8, CH_2	46.9, CH_2
16	76.0, CH	76.9, CH	76.7, CH	82.5, CH	76.9, CH	77.5, CH	76.2, CH	77.1, CH
16a	170.1, C	170.1, C	171.3, C		171.3, C	171.3, C	170.1, C	170.2, C
16b	21.7, CH_3	21.8, CH_3	22.1, CH_3		22.1, CH_3	22.1, CH_3	21.7, CH_3	21.8, CH_3
17	50.3, CH	50.1, CH	51.3, CH	60.4, CH	51.4, CH	51.5,CH	50.5, CH	50.4, CH
18	17.8, CH_3	18.2, CH_3	18.3, CH_3	21.3, CH_3	19.3, CH_3	19.5, CH_3	19.0, CH_3	19.3, CH_3
19	27.0, CH_2	27.4, CH_2	27.6, CH_2	26.5, CH_2	28.0, CH_2	28.0, CH_2	32.6, CH_2	32.7, CH_2
20	32.4, CH	35.9, CH	33.2, CH	38.2, CH	33.2, CH	36.3, CH	32.4, CH	35.8, CH
21	12.8, CH_3	11.7, CH_3	13.3, CH_3	15.8, CH_3	13.2, CH_3	12.2, CH_3	12.7, CH_3	11.5, CH_3
22	77.6, CH	74.8, CH	78.3, CH	111.4, C	78.5, CH	75.8, CH	77.6, CH	74.8, CH
22a	170.6, C		171.6, C		171.7, C		170.6, C	
22b	20.7, CH_3		20.9, CH_3		20.9, CH_3		20.7, CH_3	
23	198.7, C	204.5, C	198.9, C	77.8, CH	199.6, C	205.6, C	198.6, C	204.4, C
24	149.3, C	148.0, C	149.2, C	153.3, C	150.3, C	149.1, C	149.2, C	147.9, C
24a	123.7, CH_2	125.7, CH_2	125.6, CH_2	114.2, CH_2	124.6, CH_2	126.5, CH_2	123.8, CH_2	125.8, CH_2
25	37.2, CH	36.7, CH	34.9, CH	38.3, CH	37.9, CH	37.6, CH	37.1, CH	36.6, CH
26	66.9, CH_2	67.0, CH_2	67.8, CH_2	69.0, CH_2	66.4, CH_2	66.7, CH_2	66.9, CH_2	67.0, CH_2
26a			171.6, C					
26b			21.1, CH_3					
27	16.3, CH_3	16.5, CH_3	17.3, CH_3	17.8, CH_3	17.2, CH_3	17.2, CH_3	16.3, CH_3	16.5, CH_3
28	10.8, CH_3	10.8, CH_3	11.3, CH_3	11.3, CH_3	11.2, CH_3	11.2, CH_3	10.8, CH_3	10.8, CH_3
30	19.6, CH_3	19.6, CH_3	20.0, CH_3	19.3, CH_3	20.6, CH_3	20.7, CH_3	20.3, CH_3	20.3, CH_3

[a]In $CDCl_3$. [b]In CD_3CN.

were named neoboutomellerone (**1**) and 22-de-*O*-acetylneoboutomellerone (**2**). Compound **1** showed a pseudomolecular ion peak at m/z 591.3 $[M + Na]^+$ in the positive ESI mode that analyzed for $C_{34}H_{48}O_7Na$. The UV spectrum exhibited two maxima at 228 and 266 nm; the IR spectrum displayed strong OH vibrations at 3430 cm^{-1} and carbonyl bands at 1735, 1665, and 1602 cm^{-1}. Analysis of the ^{13}C NMR spectrum (DEPTQ and HSQC) permitted identification of seven methyl, eight methylene, ten methine, and nine quaternary carbons, thus accounting for a $C_{34}H_{47}$ partial formula and, therefore, a single OH group. Among the deshielded signals, two were clearly assignable to ketocarbonyls (δ_C 198.7 and 202.0), two ester carbonyls (δ_C 170.1 and 170.6), and an oxymethylene carbon at δ_C 66.9. The esters were identified as acetates on the basis of HMBC correlations between the CO and methyls at δ_H 2.08 and 2.16. The composition determined by HRMS was thus fully explained by NMR. A cyclohexenone moiety with a quaternary carbon atom γ to the CO, a conjugated *exo*-methylene, two acetates, two methyl groups on sp^3 quaternary carbons, and three methyl doublets were particular features of the molecule.The observation of an isolated methylene with its most upfield proton at δ_H 0.58, showing a small geminal coupling constant (J = 4.3 Hz), suggested the presence of a cyclopropane ring. Given the C_{30} composition of the basic skeleton (C_{34} minus two acetyl moieties), the first structural hypothesis for **1** was that of a diacetylated cycloartane derivative. On this basis, the two methyl singlets were assigned to C-18 and C-30, and the HMBC experiment permitted detection of the two quaternary carbons C-13 and C-14 (shared correlations with the methyls, see Table 1 for chemical shifts) as well as C-8, C-17 (CH), C-12, and C-15 (CH_2). The cyclohexenone moiety was located in ring A to account for the observed long-range couplings between the ketocarbonyl (C-3), H-1, H-4, H-5, and CH_3-28; the most upfield cyclopropane proton showed a correlation with H-1. Contrary to most described cycloartanes,[8] **1** had a proton on C-4 and therefore a single methyl group at this position. Analysis of the HMBC and HSQC experiments permitted identification of all the carbon atoms. The acetates were located on C-16 and C-22, and the alcohol function was located on the methyl group that terminates the chain. The location of the second enone moiety and the 30th carbon atom was determined by the couplings observed between the carbonyl at δ_C 198.7, the *exo*-methylene, and H-22, on one hand, and between the *exo*-methylene protons and C-26 (or -27), on the other. The extra carbon atom was thus placed at C-24, a common alkylation site in the cycloartane series.[9] The planar

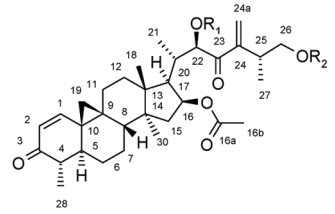
dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34–47

structure of neoboutomellerone (**1**) is thus 16,22-di-*O*-acetyl-26-hydroxy-29-nor-24-methylcycloarta-1,24(24a)-diene-3,23-dione.

The second most abundant molecule was straightforwardly identified as 22-de-*O*-acetylneoboutomellerone (**2**), H-22 appearing shielded at δ_H 4.74 instead of 5.57. Upon acetylation, compounds **1** and **2** yielded the same 26-acetylneoboutomellerone (**3**), also isolated as a minor natural product, thus demonstrating that the three compounds had the same configuration. The latter was established through a ROESY experiment and by the preparation of Mosher esters. The cyclopropyl hydrogen atoms (H-19) were distinguished by the observation that the *exo* hydrogen showed ROESY correlations with H-1 and H-11*eq*, and the *endo* hydrogen with H-4, H-6, and H-8. These observations showed that the A/B ring junction was *trans* (H-5α) and that the C-4 methyl was equatorial (H-4β). In ring D correlations between the angular methyl-18, H-8, and H-20 on one hand and between CH_3-30, H-16, and H-17, on the other, permitted assignment of a C/D *trans* ring junction with OAc-16 and the side chain in a β-orientation. The configuration of C-25 was determined by examination of the Mosher esters of **1** (**1a** and **1b**), according to the method of Ivanchina.[10] Ester **1a**, prepared with *S*-α-methoxy-α-trifluoromethylphenylacetic acid chloride, showed 0.12 ppm nonequivalence between the diastereotopic H-26, while this value was 0.04 ppm for ester **1b** prepared with the *R*-acid chloride, thus implying a 25*R* configuration. The configuration of C-22 was established similarly on diol **2** by considering the chemical shifts of H-20; in this case H-20 in the ester **2a**, prepared from the *S*-acid chloride, was found at higher field than in the ester **2b** made with the *R*-reagent (δ_H 2.57 vs 2.64). Finally, the configuration of C-20 was determined by the examination of the NMR spectra of compound **4**, a saponification product of **1**, in which a *cis*- fused five-membered ring is formed following hydrolysis of the acetate at C-16, α-ketol rearrangement, and ring closure as a hemiketal. In this compound, NOE correlations are observed between H-20 and CH_3-18, between H-17 and CH_3-21, and between CH_3-30, H-16, and H-17, thus establishing a 20*S* configuration. Compounds **1** and **2** could not be crystallized for X-ray analysis. However, phenylcarbamate **1c** was obtained in crystalline form in the course of a chemical modification program. The crystal structure determination confirmed these assignments as depicted in the formulas (see Experimental Section and Supporting Information for details).

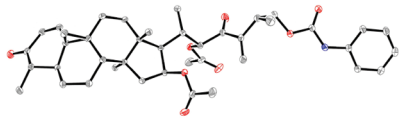
These molecules show a high degree of novelty, and their highly functionalized side chain is unique. Furthermore, there are only a few examples of nor-29-cycloart-1-en-3-ones.[11] With these two major compounds and a valuable biological activity at hand, it was decided to enlarge the panel of molecules in order to evaluate the structural requirements for activity. A derivatization program was thus undertaken,[12] as well as a systematic search for related natural products contained in this particular plant and in a few plants well known as sources of cycloartanes (*Cimicifuga simplex*, *Musa sapientum*, and *Mangifera indica*).

Two minor compounds, **5** and **6**, respectively the 1,2-dihydro derivatives of **1** and **2**, were first isolated. Their HR-MS compositions were in agreement with the expected formulas ($C_{34}H_{50}O_7$ and $C_{32}H_{48}O_6$), and their ^{1}H NMR spectra did not exhibit a deshielded AX system for H-1 and H-2. Both molecules showed a strong shielding of C-28 (δ_C 11.2), as a result of its equatorial position, β to a carbonyl group. All other ^{13}C NMR signals except those of rings A and B were similar to those of **1**−**3** (see Table 1). On the basis of these data, the structures of **5** and **6**



4

1 R_1 = $COCH_3$, R_2 = H
2 R_1 = R_2 = H
3 R_1 = R_2 = $COCH_3$
1a R_1 = $COCH_3$, R_2 = (*R*)-MTPA
1b R_1 = $COCH_3$, R_2 = (*S*)-MTPA
2a R_1 = H, R_2 = (*R*)-MTPA
2b R_1 = H, R_2 = (*S*)- MTPA
1c R_1 = $COCH_3$, R_2 = CONHPh



ORTEP representation of 1c

were identified as 1,2-dihydroneoboutomellerone and 1,2-dihydro-22-de-*O*-acetylneoboutomellerone.

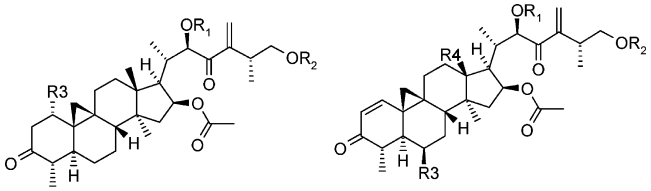
HR-MS and NMR demonstrated that the pair of new compounds **7** and **8** contained an extra oxygen atom compared to their respective parent compounds **1** and **2**. The ^{1}H NMR spectra showed a broad singlet ($W_{1/2}$ = 10 Hz) at δ_H 4.20 in the new compounds, corresponding to a CH at δ_C 65.6. In the COSY experiment, this signal displayed couplings to upfield CH and CH_2, but the HMBC failed to yield any useful correlations. Assuming that the basic skeleton of the neoboutomellerones was conserved, C-6 was left as the unique position for the hydroxylation. In agreement, the neighboring C-5 and C-7 were downfield shifted (4 and 9 ppm, respectively), while C-8 exhibited a γ shielding effect (3 ppm). This substitution also induced a strong deshielding of C-19 ($\Delta\delta$ +6, δ effect) and, most importantly, of the *endo* proton of the cyclopropane ($\Delta\delta_H$ +0.63) moiety, which could be explained by the presence of a 6β-OH group in **7** and **8**. A ROESY experiment led to the same conclusion with the observation of interactions between H-6 and CH_3-28, and between H-4 and H-19 *endo*, thus supporting the configurations of C-4, C-5, and C-6. Consequently, the structures of **7** and **8** were respectively determined to be 6β-hydroxyneoboutomellerone and 6β-hydroxy-22-de-*O*-acetylneoboutomellerone. Along with compound **7**, another isomer (**9**) was isolated, in which either CH_3-18 or CH_3-30 was replaced by a hydroxymethylene function. No HMBC correlations were observed between this CH_2 and the backbone atoms, but the remaining methyl singlet displayed four long-range CH correlations, including one with CH_2-15 (identified by scalar coupling with H-16). The OH was therefore placed at C-18, and this accounted for the chemical shift of the remaining CH_3 at δ_H 1.00 (CH_3-18 at δ_H >1.15, CH_3-30 at δ_H <1). Compound **9** was thus assigned the structure of 18-hydroxyneoboutomellerone.

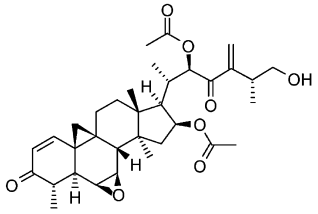
Compound **10** appeared to belong to the same series as **1**, displaying NMR signals for two *O*-acetyl groups, the γ,δ-cyclopropyl-α,β-unsaturated carbonyl, and the full and unmodified side chain. The HRMS indicated a $C_{34}H_{46}O_8$ composition corresponding to the formula of compound **7** plus one degree of unsaturation. Analysis of the ^{1}H NMR data was facilitated by the absence of signal overlap, with the sequence CH_3-28, H-4, H-5, H-6, H-7, H-8 giving rise to well-separated signals, in which H-6 and H-7 appeared as a doublet

dx.doi.org/10.1021/np200441h | *J. Nat. Prod.* 2012, 75, 34−47

of doublets at δ_H 3.15 and 3.00. These two protons were attached to carbon atoms resonating at δ_C 52.8 and 55.0, respectively, suggesting that they were incorporated in an epoxide ring. In this particular compound, H-5 was distinguished as an isolated broad doublet with J = 11.3 Hz (δ_H 2.45), thus proving its axial position and the *trans*-fusion of the A/B rings. The cyclopropane *endo* proton showed an Overhauser interaction with H-8 but not with H-6/7, allowing assignment of a β-configuration for the epoxide moiety. Shielding of the cyclopropane *exo*-proton, possibly as a result of interaction with the epoxide, is noteworthy. Compound **10** is thus 6β,7β-oxidoneoboutomellerone.

The last compound in the "regular" series, **11**, had a $C_{32}H_{48}O_7$ molecular formula, as established by HR-MS, indicative of a hydroxylated analogue of **2**. Absence of the AB system for H-1 and H-2 in the enone, as well as significant differences in the UV spectrum, suggested a 1,4-Michael addition of water to the enone functionality. An HMBC correlation between the *exo*-cyclopropane H-19 and the hydroxymethine allowed the hydroxy group to be placed at C-1. H-1 appeared as a broad triplet with J = 3 Hz (δ_H 3.80), characteristic of an equatorial proton, and, consequently, the hydroxy group was deduced to be axial. This was confirmed by the observation of an NOE interaction between H-1 and the e*xo*-cyclopropane proton. Compound **11** was thus identified as 1,2-dihydro-1α-hydroxy-22-de-*O*-acetylneoboutomellerone.

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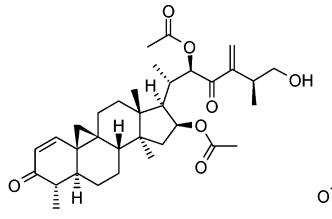
At this point, all isolated compounds could be considered as being derived from **1** and **2** and thus were assigned the same configuration as evidenced by the series of related ^{13}C NMR chemical shifts presented in Tables 1 and 2. Among the minor compounds, several isomers were isolated, including **12**, in which the ^{1}H and ^{13}C NMR spectra were almost superimposable with the corresponding spectra of **1**. The compounds were, however, quite different, with **12** being slightly less polar on reversed-phase HPLC. Since minor differences were observed for the signals of CH-25 and CH_2-26, **12** was proposed to be the C-25 epimer of **1**, i.e., 25-epi-neoboutomellerone. This hypothesis was supported by the Dess-Martin oxidation of **1** into aldehyde **13**, which was found to be unstable, rapidly converting into isomers at C-25. Compound **14**, another isomer of **1**, gave a ^{13}C NMR spectrum remarkably different from that of **1**, despite the presence of all the functionalities, i.e., the two enones, the cyclopropane, and the esters (see Table 2). The most significant chemical shift differences were found for the signals of ring B, with C-7 being almost 7 ppm downfield (Table 2). In acetonitrile-d_3, the ^{1}H NMR spectrum displayed several well-separated signals, including H-17, H_2-12, H-15, and *endo* H-19, while H-5 was masked by the broad acetonitrile residual signal, and *exo* H-19 partially overlapped with H_3-30. A ROESY experiment showed a correlation between H-5 and the *endo* H-19, suggesting that these atoms were cofacial, thus offering two possibilities: H-5β in a "normal" series or H-5α with H_2-19 α-oriented. The presence of a *cis*-fused decalin moiety was deduced by the observation of an NOE correlation between H-1 and *exo* H-19, a feature not observed in any product in the *trans* series. The decision between the two hypotheses came in a serendipitous manner with the observation of a slow conversion of **1** into **14** upon storage in acetonitrile or by irradiation under a UV lamp, which could only be explained by opening of the cyclopropane (cleavage of the C-9/C-10 bond activated by the enone system) and ring closure to yield the thermodynamically more favorable *cis* isomer.[13] The configuration of all stereogenic centers was assumed to be as in compound **1**, and the proposed structure for **14** was 9,10-di-*epi*-25ξ-neoboutomellerone. Compound **15** is isomeric to **2** and was assigned the structure 9,10-di-epi-25ξ-22-de-*O*-acetylneoboutomellerone owing to the strong similarity between the ^{13}C NMR spectra of **15** and **2**.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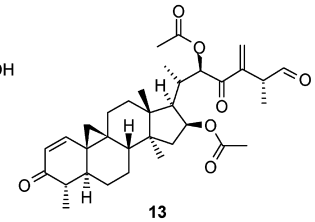
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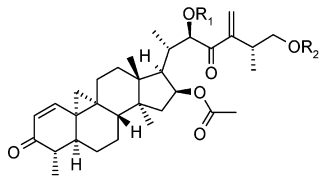

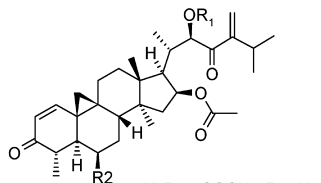

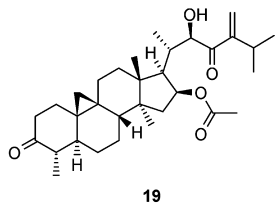
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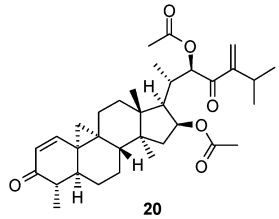
20

The next set of new compounds (**16**–**20**) shared the same side chain as before but without oxidation at C-26. As for **1** and **2**, the difference between **16** and **17** was an acetyl group at C-22. The absence of a deshielded signal for H-26 and the presence of two diastereotopic methyl doublets for H_3-26 and H_3-27 suggested that these compounds were 26-deoxyneoboutomellerone (**16**) and 22-de-*O*-acetyl-26-deoxyneoboutomellerone (**17**), respectively. Compound **18** belonged to the 22-OAc series, but its molecular weight was 16 amu higher than observed for **16** (m/z 591.3287 for $[M + Na]^+$). The ^{1}H NMR spectrum of **18** showed similarity to that of **7**, including a broad deshielded singlet for H-6 and characteristic deshielding for *endo* H-19

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34–47

Table 2. ^{13}C NMR Data of Compounds 9−16

carbon	9[a]	10[b]	11[b]	12[a]	13[b]	14[a]	15[a]	16[b]
1	153.5, CH	154.2, CH	74.0, CH	153.7, CH	155.5, CH	155.4, CH	155.7, CH	155.6, CH
2	128.3, CH	128.4, CH	49.3, CH_2	128.3, CH	128.4, CH	126.5, CH	126.3, CH	128.4, CH
3	202.0, C	202.1, C	212.2, C	202.0, C	202.4, C	200.6, C	200.7, C	202.5, C
4	47.0, CH	45.6, CH	50.5, CH	46.9, CH	47.6, CH	47.7, CH	47.8, CH	47.6, CH
5	43.0, CH	41.5, CH	39.5, CH	42.5, CH	43.6, CH	39.4, CH	39.4, CH	43.6, CH
6	23.5, CH_2	52.8, CH	26.1, CH_2	23.4, CH_2	24.3, CH_2	20.6, CH_2	20.6, CH_2	24.3, CH_2
7	24.0, CH_2	55.0, CH	26.2, CH_2	23.5, CH_2	24.3, CH_2	31.2, CH_2	31.2, CH_2	24.3, CH_2
8	45.0, CH	38.9, CH	48.7, CH	44.3, CH	45.2, CH	40.7, CH	40.7, CH	45.2, CH
9	25.8, C	27.2, C	26.0, C	26.0, C	27.2, C	33.7, C	33.9, C	27.7, C
10	32.0, C	31.8, C	34.0, C	31.9, C	33.4, C	26.6, C	26.6, C	32.9, C
11	27.5, CH_2	27.4, CH_2	26.5, CH_2	27.4, CH_2	28.1, CH_2	29.8, CH_2	29.9, CH_2	28.1, CH_2
12	27.7, CH_2	32.1, CH_2	33.3, CH_2	32.1, CH_2	33.0, CH_2	32.1, CH_2	32.1, CH_2	32.9, CH_2
13	47.5, C	47.1, C	46.6, C	47.4, C	46.9, C	47.8, C	47.8, C	46.8, C
14	51.0, C	47.3, C	47.9, C	46.0, C	48.3, C	46.1, C	46.0, C	48.4, C
15	46.2, CH_2	44.7, CH_2	47.9, CH_2	46.0, CH_2	46.7, CH_2	43.8, CH_2	44.0, CH_2	46.7, CH_2
16	76.6, CH	76.3, CH	77.5, CH	75.9, CH	76.8, CH	76.0, CH	76.9, CH	76.6, CH
16a	169.4, C	171.3, C	171.3, C	170.1, C	171.2, C	170.1, C	170.1, C	171.3, C
16b	21.6, CH_3	22.1, CH_3	22.1, CH_3	21.7, CH_3	22.1, CH_3	21.7, CH_3	21.8, CH_3	22.1, CH_3
17	49.6, CH	49.7, CH	51.5, CH	50.3, CH	51.2, CH	49.5, CH	49.3, CH	51.3, CH
18	64.9, CH_2	15.2, CH_3	19.6, CH_3	17.8, CH_3	18.3, CH_3	15.1, CH_3	15.3, CH_3	18.2, CH_3
19	28.5, CH_2	22.1, CH_2	28.1, CH_2	27.0, CH_2	27.6, CH_2	31.3, CH_2	31.4, CH_2	27.7, CH_2
20	32.8, CH	33.2, CH	36.3, CH	32.2, CH	33.8, CH	32.5, CH	35.9, CH	33.0, CH
21	13.4, CH_3	13.7, CH_3	12.2, CH_3	12.7, CH_3	13.4, CH_3	13.2, CH_3	12.1, CH_3	13.2, CH_3
22	77.4, CH	78.6, CH	75.8, CH	77.8, CH	78.2, CH	77.7, CH	75.0, CH	78.5, CH
22a	170.5, C	171.7, C		170.7, C	171.6, C	170.6, C		171.7, C
22b	20.7, CH_3	20.9, CH_3		20.7, CH_3	20.9, CH_3	20.6, CH_3		20.9, CH_3
23	198.5, C	199.6, C	205.6, C	198.6, C	198.3, C	198.8, C	204.6, C	199.5, C
24	149.3, C	150.3, C	149.1, C	149.2, C	145.8, C	149.3, C	148.0, C	154.3, C
24a	123.6, CH_2	124.7, CH_2	126.5, CH_2	124.3, CH_2	128.6, CH_2	123.7, CH_2	125.6, CH_2	122.3, CH_2
25	37.4, CH	38.0, CH	37.6, CH	37.9, CH	48.9, CH	37.2, CH	36.7, CH	29.7, CH
26	66.8, CH_2	66.4, CH_2	66.7, CH_2	66.8, CH_2	201.7, CH	66.9, CH_2	67.0, CH_2	21.6, CH_3
27	16.4, CH_3	17.2, CH_3	17.2, CH_3	15.5, CH_3	13.8, CH_3	16.3, CH_3	16.4, CH_3	22.4, CH_3
28	10.8, CH_3	11.3, CH_3	11.0, CH_3	10.8, CH_3	11.3, CH_3	12.2, CH_3	12.2, CH_3	11.3, CH_3
30	21.7, CH_3	19.6, CH_3	20.7, CH_3	19.6, CH_3	20.0, CH_3	18.6, CH_3	18.6, CH_3	20.0, CH_3

[a]In $CDCl_3$. [b]In CD_3CN.

(δ_H 1.81). This feature, as well as the overall concordance between the spectra of **7** and **18**, indicated that **18** was 6β-hydroxy-26-deoxyneoboutomellerone. Compound **19**, like the preceding compounds, was characterized by four methyl doublets and therefore lacked oxidation at the terminus of the side chain. This compound had a molecular weight of 512, corresponding to a $C_{32}H_{48}O_5$ composition. This confirmed the absence of an oxygen atom and suggested that **19** was a dihydro analogue of a derivative of **2**. Since the characteristic signals for the ring A double bond were missing, it was deduced that **19** was 1,2-dihydro-22-de-*O*-acetyl-26-deoxyneoboutomellerone. Compound **20** was an isomer of **16** ($C_{34}H_{48}O_6$), and, given the previous analyses, it was readily identified as the cyclopropyl isomer of **16**, i.e., 9,10-di-epi-26-deoxyneoboutomellerone.

Compounds **21**−**23** differed from the previous compounds by the absence of the C-24 *exo*-methylene group. Examination of the NMR spectra showed that C-24a was replaced by a proton (δ_H 6.15 ± 0.2) and that the chain was terminated by a 3-methyl-2-butenoyl moiety. In these three compounds, the tetracyclic cycloartane core was intact, and, as for compounds **1** and **2**, **21** and **22** differed only by the presence of a C-22-*O*-acetyl moiety. Compound **21** was thus 24a-nor-24,25-didehydro-26-deoxyneoboutomellerone, and **22** was 22-de-*O*-acetyl-24a-nor-24,25-didehydro-26-deoxyneoboutomellerone. While **21** and **22** were shown by ^{13}C NMR to belong to the normal A/B *trans* series, **23** displayed a markedly different spectrum, characteristic of the *cis* α-series. Consequently, its structure was deduced to be 9,10-di-epi-24a-nor-24,25-didehydro-26-deoxyneoboutomellerone (see Table 3).

Compounds **24** and **25** had molecular weights of 412 and 428, respectively, corresponding to molecular formulas $C_{26}H_{36}O_4$ and $C_{26}H_{36}O_5$. Although six carbon atoms were missing, the ^1H NMR spectra of both compounds showed strong resemblance with those of the neoboutomellerones and displayed signals for an acetate, an α,β-unsaturated carbonyl, a cyclopropane, and the angular methyl groups. Initially, it was proposed that these compounds were lacking C-23 to C-27 of the "regular" series side chain. A formyl proton doublet at δ_H 9.58 (J = 1.8 Hz) coupling with a doublet of doublets of quartets (δ_H 2.86) suggested that truncation had happened between C-22 and C-23. Since the ^{13}C NMR spectrum showed that the tetracyclic core was intact, **24** was proposed to be 23,24,24a,25,26,27-hexa-nor-neoboutomelleron-22-al. This was confirmed by the high-yield oxidation of **2** into **24** by MnO_2. It is worth noting that **1** was recovered unchanged under the same conditions. The ^{13}C NMR spectra of **24** and **25** could barely be distinguished except for the formyl carbonyl carbon, which was replaced by a signal at δ_C 182.0. It was deduced that **25** was the corresponding acid

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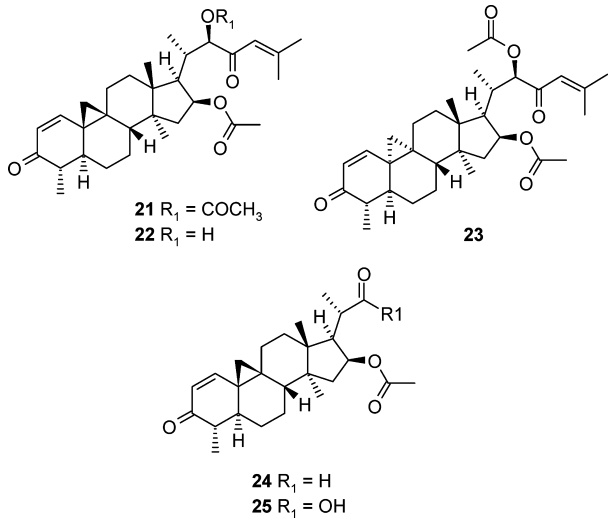
Table 3. ^{13}C NMR Data of Compounds 17−24

carbon	17[a]	18[a]	19[a]	20[a]	21[a]	22[a]	23[a]	24[a]
1	155.7, CH	156.2, CH	33.5, CH_2	157.3, CH	155.7, CH	155.7, CH	157.3, CH	155.4, CH
2	128.3, CH	127.5, CH	41.5, CH_2	126.5, CH	128.4, CH	128.4, CH	126.5, CH	128.5, CH
3	202.5, C	203.4, C	213.3, C	201.0, C	202.6, C	202.5, C	201.0, C	202.3, C
4	47.6, CH	44.9, CH	50.5, CH	48.5, CH	47.6, CH	47.6, CH	48.5, CH	47.6, CH
5	43.6, CH	46.6, CH	47.0, CH	40.4, CH	43.5, CH	43.5, CH	40.4, CH	43.3, CH
6	24.3, CH_2	65.6, CH	26.5, CH_2	21.3, CH_2	24.2, CH_2	24.3, CH_2	21.3, CH_2	24.1, CH_2
7	24.3, CH_2	32.7, CH_2	26.2, CH_2	31.9, CH_2	24.2, CH_2	24.2, CH_2	31.9, CH_2	24.0, CH_2
8	45.3, CH	41.1, CH	48.6, CH	41.5, CH	44.9, CH	45.0, CH	41.4, CH	44.3, CH
9	27.2, C	26.5, C	27.4, C	27.7, C	27.0, C	27.1, C	27.6, C	27.3, C
10	33.0, C	30.8, C	33.5, C	34.9, C	32.9, C	32.8, C	31.5, C	33.0, C
11	28.1, CH_2	27.7, CH_2	27.4, CH_2	30.4, CH_2	28.0, CH_2	28.1, CH_2	30.4, CH_2	28.0, CH_2
12	33.1, CH_2	33.1, CH_2	33.5, CH_2	32.9, CH_2	32.7, CH_2	32.9, CH_2	32.7	32.6, CH_2
13	46.7, C	47.1, C	46.7, C	47.0, C	46.7, C	46.2, C	46.8, C	46.5, C
14	48.3, C	47.7, C	47.9, C	48.8, C	48.7, C	48.2, C	48.8, C	48.4, C
15	46.9, CH_2	47.5, CH_2	47.8, CH_2	44.6, CH_2	46.1, CH_2	46.3, CH_2	44.1, CH_2	45.3, CH_2
16	77.3, CH	76.8, CH	77.5, CH	76.5, CH	76.1, CH	76.5, CH	76.1, CH	75.1, CH
16a	171.3, C	171.3, C	171.3, C	171.3, C	171.3, C	171.3, C	171.3, C	170.9, C
16b	22.1, CH_3	22.1, CH_3	22.1, CH_3	22.1, CH_3	21.8, CH_3	21.8, CH_3	21.8, CH_3	21.3, CH_3
17	51.4, CH	51.5, CH	51.6, CH	50.6, CH	51.1, CH	51.2, CH	50.2, CH	50.7, CH
18	18.5, CH_3	19.4, CH_3	19.5, CH_3	15.4, CH_3	18.1, CH_3	18.3, CH_3	15.4, CH_3	18.4, CH_3
19	27.7, CH_2	33.0, CH_2	28.0, CH_2	31.6, CH_2	27.4, CH_2	27.4, CH_2	31.6, CH_2	26.9, CH_2
20	36.2, CH	33.0, CH	36.2, CH	33.0, CH	32.7, CH	34.4, CH	32.8, CH	45.1, CH
21	12.3, CH_3	13.1, CH_3	12.2, CH_3	13.6, CH_3	13.2, CH_3	12.2, CH_3	13.5, CH_3	13.4, CH_3
22	75.8, CH	78.5, CH	75.7, CH	78.6, CH	81.5, CH	79.0, CH	81.6, CH	205.2, CH
22a		171.7, C		171.7, C	171.7, C		171.6, C	
22b		20.9, CH_3		20.9, CH_3	21.2, CH_3		21.2, CH_3	
23	205.4, C	199.4, C	205.4, C	199.6, C	196.9, C	202.5, C	196.9, C	
24	153.1, C	154.3, C	153.1, C	154.3, C	120.5, CH	120.3, CH	120.5, CH	
24a	124.3, CH_2	122.3, CH_2	124.3, CH_2	122.3, CH_2				
25	29.5, CH	29.7, CH	29.5, CH	29.7, CH	159.8, C	159.9, C	159.7, C	
26	22.0, CH_3	22.4, CH_3	22.0, CH_3	21.6, CH_3	28.0, CH_3	28.1, CH_3	28.0, CH_3	
27	22.6, CH_3	21.7, CH_3	22.6, CH_3	22.4, CH_3	21.0, CH_3	21.3, CH_3	21.0, CH_3	
28	11.3, CH_3	11.0, CH_3	11.3, CH_3	12.6, CH_3	11.3, CH_3	11.3, CH_3	12.6, CH_3	11.3, CH_3
30	20.1, CH_3	20.7, CH_3	20.8, CH_3	18.9, CH_3	19.8, CH_3	19.9, CH_3	18.8, CH_3	19.4, CH_3

[a]In CD_3CN.

of **24**, i.e., 23,24,24a,25,26,27-hexa-nor-neoboutomelleron-22-oic acid.

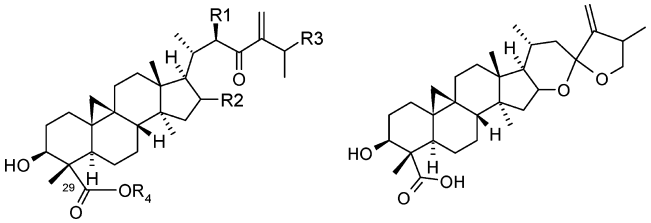
Six more compounds (**26**−**31**) were isolated and, although devoid of any biological activity (*vide infra*), are interesting from a biosynthetic standpoint. They all shared common functionalities in rings A and B: a hydroxy group at C-3 and a quaternary C-4 bearing a hydroxycarbonyl or ester function. These compounds could be considered as the precursors for the aforementioned compounds, undergoing oxidation of a C-4 methyl group of a cycloartane, oxidation of C-3, followed by decarboxylation and introduction of the $\Delta^{1,2}$ double bond, paralleling the well-known biosynthesis of the steroids. All six compounds had a 3*S* configuration (3β-OH) based on the large vicinal coupling between H-3 and H-2α (ca. 11 Hz) and a 4*S* configuration to account for the strong shielding of the axial C-28 methyl group (δ_C ca. 10). In the ^1H NMR spectrum of **27**, H-5 was observed as a doublet of doublets with J = 12.2 and 3.4 Hz and was therefore α-axially oriented. Configurations of the other ring junctions, and of C-16, C-20, and C-22, were most likely the same as for the major compounds given the similarities in chemical shift values. Compounds **26** and **27** were isomers with a $C_{33}H_{50}O_7$ molecular formula, and comparison of their NMR spectra showed that they differed in the location of the side chain hydroxy group: C-22 for **27** and C-25 for **26**.



However, it was not possible to define the configuration of C-25 in compound **26** due to a paucity of material. Compound **29** was not oxidized at C-16 nor C-22, as shown by the presence of two methylene resonances at δ_C 29.3 and 46.5. The C-22 methylene group gave rise to two well-separated doublets of doublets, with 15.6 Hz geminal couplings and vicinal couplings

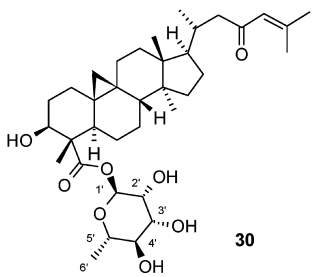
dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34−47

of 3.1 and 10.1 Hz, the smaller constant being close to that observed in all the HO-22 derivatives. The mass spectrum of compound **28** indicated a molecular formula of $C_{31}H_{46}O_5$, corresponding to the loss of HOAc with regard to **26** and **27**. Accordingly, no signal was observed for an acetate group in the ^{1}H NMR spectrum. A noteworthy feature of the ^{13}C NMR spectrum was the presence of a quaternary carbon at δ_C 108, which showed HMBC couplings with two *exo*-methylene protons, sp^2 C-24a, and an oxymethylene corresponding to C-26. These characteristics were best accounted for by a ketal derived from a precursor possessing a C-23 carbonyl group and hydroxy groups at C-16 and C-26. No NOE correlations between protons of the spiroketal rings were evident, and we could not assign configurations to C-23 and C-25. Compounds **30** and **31** shared with **29** the absence of oxidation of C-16 and C-22, but analysis of the NMR spectra suggested that these highly polar compounds were glycosides. In both compounds, the sugar was tentatively identified as an α-L-rhamnose. The difference between the two compounds was in the side chain, with **31** having the same side chain as **16** and **17**, and **30** having the truncated chain of **21** and **22**. Therefore, the respective structures of these compounds were 16-acetyl-3β,26-dihydroxy-24-methyl-25ξ-cycloart-24(24a)-en-23-on-29-oic acid (**26**), 16-acetyl-3β,22β-dihydroxy-24-methylcycloart-24(24a)-en-23-on-29-oic acid (**27**), 3β,16β,22β-trihydroxy-24-methyl-(16,23:23,26)-diepoxycycloart-24(24a)-en-29-oic acid (**28**), 3β-hydroxy-24-methylcycloart-24(24a)-en-23-on-29-oic acid (**29**), 3β-hydroxy-29-*O*-(α-L-rhamnopyranosyl)cycloart-24-en-23-on-29-oic acid (**30**), and 3β-hydroxy-29-*O*-(α-L-rhamnopyranosyl)-24-methylcycloart-24(24a)-en-23-on-29-oic acid (**31**).



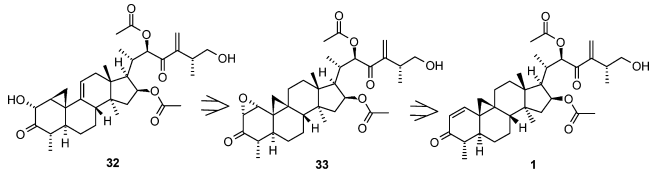
28

26 $R_1 = R_4 = H$, $R_2 = OCOCH_3$, $R_3 = CH_2OH$
27 $R_1 = OH$, $R_2 = OCOCH_3$, $R_3 = CH_3$, $R_4 = H$
29 $R_1 = R_2 = R_4 = H$, $R_3 = CH_3$
31 $R_1 = R_2 = H$, $R_3 = CH_3$, $R_4 = \alpha$-L-rha



30

Compound **32** was an isomer of **7**, with its ^{1}H NMR spectrum displaying all the signals corresponding to rings C and D and of the side chain. However, the enoyl AB system in **7** was missing and the usual upfield doublet for H-19 was replaced by a doublet of doublets at δ_H 0.41 (J = 9.9, 4.7 Hz) and 0.94 [broad triplet (J = 4.7 Hz)], with both signals coupled to a broadened doublet of doublets at δ_H 1.88 (J = 9.9, 4.7 Hz). The values of the vicinal coupling constants in the cyclopropane ring indicated that the most upfield proton was *endo*, since it is known that in such systems $J_{trans} > J_{cis}$.[14] A trisubstituted C-9/C-11 double bond was evident, with H-11 being a broad doublet coupling with H_2-12. This assignment was supported by an HMBC correlation to C-18. A broad hydroxymethine singlet [δ_H 4.16 (δ_C 73.2)] showed HMBC couplings with the methine, the methylene, and the quaternary carbon of the cyclopropane moiety. The COSY experiment showed a weak but definite coupling between this proton and the cyclopropane proton at δ_H 1.88. Consequently, the hydroxy group was placed at C-2, and the cyclopropane bridge between C-1 and C-10. A NOESY experiment showed a correlation between H-2 and the *exo*-cyclopropane proton, suggesting that this proton and the cyclopropane moiety were β-cofocially oriented, and therefore the 2-OH group must be α-oriented. This molecule is another member of a rare family of triterpenes that are linked to the parent compounds through acidic rearrangements, for example, simplexol and *O*-methylcinicimol, arising from cimigenol.[15] In our case, the putative epoxide **33**, an oxidation product of **1**, should be the intermediate between **32** and the regular cycloartanes, and the rearrangement should be favored by an *anti* relationship between the cyclopropane and the oxirane, hence giving further support to the 2α-OH configuration. On the basis of the above evidence, the structure of **32** was proposed to be 16,22-diacetyl-2,26-dihydroxy-29-nor-24-methyl-19(9→1)-*abeo*-cycloart-9(11),24(24a)-diene-3,23-dione.



32 **33** **1**

Structure–Activity Relationship Analysis. Twenty-four of the isolated compounds the two non-natural derivatives, **4** and **13**, were evaluated in the cellular proteasome assay (Table 5). Cimiracemosides F and G,[16] cycloeucalone,[17] 28-nor-cycloeucalone, mangiferonic, and mangiferolic acids,[18] isolated from *Cimicifuga simplex*, *Musa sapientum*, *and Mangifera indica*, were also tested, but none were found to be active. Table 5 lists the induction factors (IFs) observed for the cycloartanes. The IF is defined as the increase in luciferase signal measured in the 4UB-Luc-DLD-1 cells, after 7 h exposure to the test compound, as compared to untreated cells.[3b] Epoxomycin was used as reference with an IF of 100 at 0.5 μM. Due to the cytotoxicity of the compounds and/or the accumulation of proteasome substrates (known to produce cytotoxic effects), it was not possible to observe luminescence at the highest concentrations. It also explains why a maximum on the induction factor (IF) versus concentration plots is observed (bell-shaped curve). The two major compounds (**1**, **2**) from the plant were also the most active, with IF values of 48 and 45 at 1 μM, respectively, and the activity decreased with the acylation level at the C-26 hydroxy group, with **1**, **2** > **3**. The integrity of the side chain seemed to be another requirement since compounds such as **4** or **24** showed no activity. Simple modifications, such as the removal of the *exo*-methylene in this side chain (**21** and **22**) or the alteration of CH_3-18 into a hydroxymethlene (**9**), also reduced the biological activity. However, a C-6 hydroxy group (**7**, **8**), an epoxy moiety on C-6/C-7 (**10**), a configurational change at C-25 (**12**), a cyclopropyl isomerism (**14**, **15**), or substitutions at C-26 (**13**, **16**, **17**) represent structural modifications that do not significantly affect biological activity.

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34–47

Table 4. ^{13}C NMR Data of Compounds 25−32

carbon	25[a]	26[a]	27[c]	28[c]	29[c]	30[c]	31[c]	32[a]
1	153.7, CH	31.4, CH_2	32.8, CH_2	32.8, CH_2	32.8, CH_2	32.7, CH_2	32.7, CH_2	23.6, CH
2	128.3, CH	29.4, CH_2	30.5, CH_2	30.5, CH_3	30.5, CH_2	30.4, CH_2	30.4, CH_2	73.2, CH
3	202.0, C	75.4, CH	76.3 CH	76.3, CH	76.3, CH	75.9, CH	75.9, CH	213.2, C
4	46.8, CH	54.5, C	56.0, C	55.9, C	55.8, C	56.5, C	56.5, C	40.4, CH
5	42.3, CH	44.0, CH	45.7, CH	45.5, CH	45.6, CH	46.0, CH	46.0, CH	47.6, CH
6	23.4, CH_2	22.8, CH_2	24.1, CH_2	23.9, CH_2	24.1, CH_2	23.9, CH_2	23.9, CH_2	26.6, CH_2
7	23.3, CH_2	25.6, CH_2	26.9, CH_2	27.1, CH_2	26.7, CH_2	26.5, CH_2	26.5, CH_2	29.5, CH_2
8	43.4, CH	47.7, CH	49.7, CH	49.1, CH	49.3, CH	49.3, CH	49.3, CH	44.2, CH
9	n.d.[c]	19.7, C	20.8, C	20.8, C	21.1, C	21.2, C	21.2, C	140.4, C
10	32.0, C	25.1, C	26.6, C	26.8, C	26.4, C	26.3, C	26.3, C	30.0, C
11	27.4, CH_2	26.0, CH_2	27.1, CH_2	27.0, CH_2	27.4, CH_2	27.4, CH_2	27.4, CH_2	116.1, CH
12	32.0, CH_2	32.3, CH_2	33.8, CH_2	34.1, CH_2	34.0, CH_2	33.9, CH_2	33.9, CH_2	36.2, CH_2
13	45.2, C	45.9, C	46.9, C	45.7, C	46.6, C	46.6, C	46.6, C	45.1, C
14	47.5, C	47.1, C	48.3, C	47.4, C	50.1, C	50.1, C	50.1, C	45.2, C
15	44.0, CH_2	45.7, CH_2	48.1, CH_2	44.7, CH_2	36.5, CH_2	36.5, CH_2	36.5, CH_2	45.0, CH_2
16	75.0, CH	75.1, CH	78.1, CH	74.0, CH	29.3, CH_2	29.3, CH_2	29.3, CH_2	76.2, CH
16a	171.8, C	171.0, C	172.3, C					170.1, C
16b	21.1, CH_3	21.2, CH_3	21.9, CH_3					21.7, CH_3
17	52.0, CH	54.7, CH	51.8, CH	57.7, CH	53.8, CH	53.8, CH	53.8, CH	49.1, CH
18	18.0, CH_3	18.5, CH_3	19.5, CH_3	20.9, CH_3	18.5, CH_3	18.6, CH_3	18.6, CH_3	15.1, CH_3
19	26.5, CH_2	30.2, CH_2	31.3, CH_2	31.3, CH_2	30.7, CH_2	30.8, CH_2	30.8, CH_2	15.8, CH_2
20	38.7, CH	27.3, CH	36.2, CH	26.8, CH	35.2, CH	34.9, CH	35.1, CH	32.4, CH
21	17.5, CH_3	19.2, CH_3	12.1, CH_3	21.0, CH_3	19.8, CH_3	19.7, CH_3	19.7, CH_3	12.8, CH_3
22	182.0, C	44.8, CH_2	76.0, CH	43.8, CH_2	46.5, CH_2	52.7, CH_2	46.4, CH_2	77.6, CH
22a								170.6, C
22b								20.7, CH_3
23		203.1, C	205.1, C	108.0, C	205.0, C	204.2, C	204.9, C	198.7, C
24		151.8, C	154.2, C	159.2, C	157.1, C	125.3, CH	157.1, C	149.3, C
24a		123.8, CH_2	123.2, CH_2	106.2, CH_2	122.6, CH_2		122.6, CH_2	123.7, CH_2
25		36.6, CH	29.9, CH	38.7, CH	29.1, CH	157.0, C	29.1, CH	37.2, CH
26		67.6, CH_2	22.6, CH_3	72.7, CH_2	22.3, CH_3	27.7, CH_3	22.3, CH_3	66.9, CH_2
27		16.1, CH_3	21.9, CH_3	14.3, CH_3	22.4, CH_3	20.9, CH_3	22.4, CH_3	16.3, CH_3
28	10.8, CH_3	9.1, CH_3	10.2, CH_3	10.1, CH_3	10.0, CH_3	9.9, CH_3	9.9, CH_3	10.8, CH_3
29		180.8, C	181.7, C	181.4, C	181.2, C	176.2, C	176.2, C	
30	19.0, CH_3	20.0, CH_3	20.7, CH_3	20.0, CH_3	19.7, CH_3	19.7, CH_3	19.8, CH_3	19.4, CH_3
1′						94.9, CH	94.9, CH	
2′						71.2, CH	71.2, CH	
3′						72.4, CH	72.4, CH	
4′						73.3, CH	73.3, CH	
5′						72.4, CH	72.4, CH	
6′						17.9, CH_3	17.9, CH_3	

[a]In $CDCl_3$ [c]In methanol-d_4. [c]n.d., not detected.

Moreover, minor changes in ring A were tolerated; the 1,2-dihydro derivatives (**5**, **6**) and 1-hydroxy derivative (**11**) showed some activity, possibly due to reoxidation *in vivo*. Compounds lacking the A-ring enone function (**26**−**28**, **30**−**32**) or the C-16-ester moiety (**4**, **28**) were also found to be weakly active. Acetylation of the C-22-hydroxy group results in minor decreases of activity among comparable structures (**1**/**2**, **5**/**6**, **7**/**8**, **16**/**17**, and **21**/**22**). Finally, enone functionalities associated with the side chain and in ring A appear to be crucial to maintain the biological activity, thus leaving the C-16-ester group and the C-22-hydroxy function as possible points of modification.

CONCLUSION

From a phytochemical standpoint, it is not surprising to find cycloartanes as major secondary metabolites in a Euphorbiaceae species such as *N. melleri*. Cursory investigation of the terpenes from the twigs did not show the presence of cycloartanes but rather daphnanes, as observed in the only published investigation on similar material.[6] Despite the ubiquity of these triterpenes, the oxidation level of the molecules discussed here is most peculiar, with the rare combination of an α,β-unsaturated carbonyl with a cyclopropane giving rise to photochemical reactivity. The two Michael acceptors in ring A and in the side chain add further biological properties to the neoboutomellerones. In fact, the two moieties may act as reversible or irreversible inhibitors across the ubiquitin−proteasome pathway. From a medicinal point of view, the novelty of the isolated compounds and their interesting pharmacological properties motivate the antitumor activity evaluation of the most abundant molecules **1** and **2**.[19]

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34−47

Table 5. Induction Factors for Cycloartanes

	induction factor (4Ub Luc DLD-1 cell) at dose (μM)			
compound	10	5	1	0.5
1	4.4	37	48	2.1
2	2.9	28	45	1.4
3		48	12	1.1
4	2.6	1.0	0.9	
5	48	45	0.9	
6	77	10	1.2	
7		31	5.1	1.2
8		35	1.1	
9	1.1	12	1.2	
10	15	72	2.8	
11	74	11	1.6	1.3
12	60	71	1.5	1.0
13	31	70	1	
14	24	84	2	1.4
15		9.1	0.9	0.9
16	19	74	7.2	
17	20	65	2.4	
21	44	34	1.1	
22	53	5.2	1.1	
24	3.0	1.3	1.1	
26	2.8	1.3	1.0	
27	1.8	1.3	1.2	
28	1.3	1.3	1.2	
30	1.1	1.0	1.0	
31	1.1	1.0	1.1	
32	15	1.9	1.2	

EXPERIMENTAL SECTION

General Experimental Procedures. Optical rotations were determined on a Perkin-Elmer 341 automatic polarimeter. UV spectra were obtained in MeOH using a UV MC^2 Safas spectrophotometer. An FT-IR Bruker Tensor 27 spectrophotometer was used for scanning IR spectroscopy. The NMR spectra were recorded on a Bruker Avance II spectrometer equipped with a ^{13}C cryoprobe at 500 MHz for ^{1}H and 125 MHz for ^{13}C; 2D experiments were performed using standard Bruker programs. The ESIMS and MS/MS were performed using a Bruker Esquire-LC ion trap mass spectrometer; the samples were introduced by infusion in a solution of MeOH. HR-ESIMS were obtained on a Bruker MicrOTOF. TLC was carried out on precoated silica gel 60F$_{254}$ (Merck) with CH_2Cl_2–MeOH (97:3), and spots were visualized by heating after spraying with 3% H_2SO_4 + 1% vanillin. CC was carried out on a prepacked Kieselgel cartridge (40–60 μm) with cyclohexane–EtOAc (50:50). Analytical HPLC was performed on a Merck-Hitachi apparatus equipped with an L-7200 automated sample injector, an L-7100 pump, a L-7450 diode array detector, a D-7000 interface, and Lachrom HSM or EZChrom software. A prepacked C_{18} reversed-phase column (Lichrospher 100 RP-18, 4 × 125 mm, 5 μm) was used for analytical HPLC with a binary gradient elution (solvent A: H_2O; solvent B: MeCN) and a flow rate of 1 mL·min^{-1}. Semipreparative HPLC was performed on an apparatus equipped with a VWR International LaPrep pump P110, a VWR LaPrep P314 Dual λ absorbance detector, and EZChrom software. A 50 × 250 mm column (NW 50, Merck) with LiChroprep RP-18 (15–25 μm) and a prepacked C_{18} reversed-phase column (Hibar-Lichrospher 100 RP-18, 25 × 250 mm, 5 μm) were used for semipreparative HPLC with a binary gradient elution (solvent A: H_2O; solvent B: MeCN) and a flow rate of 100 or 30 mL.min^{-1}, and the chromatogram was monitored at 210 and 270 nm.

Plant Material. The original plant sample was collected by René Bellé (Pierre Fabre Research Institute) in Malantouen (Cameroon) on June 21, 1988, and an herbarium specimen was deposited under reference RBL-273 in the Pierre Fabre Botanical Conservatory in Cambounet-sur-le-Sor (France). The plant was collected again by one of us (B.D.) at the same location and identified by comparison of the vouchers at the National Herbarium of the Museum National d'Histoire Naturelle in Paris. HPLC of the extracts showed similar profiles.

Extraction and Isolation. The dried leaves (1 kg) of *N. melleri* were powdered and extracted at room temperature with CH_2Cl_2 (15 L) overnight. After filtration, the extract was concentrated under reduced pressure. The residue (45 g) was dissolved in CH_2Cl_2 (1 L) and stirred for 1 h with 100 g of activated vegetable charcoal. After filtration and concentration under reduced pressure, the extract (32 g) was partitioned between MeOH and cyclohexane. The MeOH fraction (15 g) was subjected to silica gel CC (600 g, 57 × 420 mm) using cyclohexane–EtOAc (50:50) to give 50 fractions of 150 mL. All the fractions were analyzed by TLC on silica gel using the solvent mixture CH_2Cl_2–MeOH (97:3) and pooled according to TLC into four fractions (F1–F4). Fraction F1 (6.0 g) was purified by semipreparative HPLC RP-18 chromatography, eluting with a linear gradient (80 to 100% B), to give after repeated HPLC purification **17** (84 mg), **19** (53 mg), **23** (6 mg), **3** (5 mg), **22** (25 mg), **21** (18 mg), **16** (76 mg), **24** (7 mg), **20** (3 mg), and **18** (1 mg). Fractions F2 (5.5 g) and F3 (1.5 g) were purified by semipreparative RP-18 chromatography, eluting with a linear gradient (45–100% B). After repeated HPLC purification of F2, compounds **1** (2.5 g, 0.25%), **6** (180 mg, 0.018%), and **5** (107 mg, 0.0107%) were obtained. Fraction F3, after repeated HPLC purification, afforded the second most abundant compound, **2** (600 mg), **10** (11 mg), and **15** (63 mg). Fraction F4 (1.0 g) was purified by semipreparative RP-18 chromatography, to give after repeated HPLC purification with a linear gradient (90–100% B and 50–100% B) the most polar compounds, **26** (3 mg), **30** (5 mg), **27** (7 mg), **29** (23 mg), **28** (17 mg), **11** (12 mg), **25** (3 mg), **31** (2 mg), 7 (34 mg), **8** (18 mg), **14** (80 mg), **12** (5 mg), **32** (1 mg), and **9** (1 mg).

*Neoboutomellerone (**1**):* white fluffy solid, $[\alpha]^{20}_D$ −55 (*c* 0.16, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 224 (3.77), 266 (3.83) nm; IR (film) ν_{max} 3430, 1735, 1665, 1602, 1230 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.85 (1H, d, *J* = 10.1 Hz, H-1), 6.13 (1H, s, H-24a), 6.02 (1H, d, *J* = 10.1 Hz, H-2), 5.89 (1H, br s, H-24a), 5.57 (1H, d, *J* = 2.1 Hz, H-22), 5.09 (1H, dt, *J* = 4.6, 7.6 Hz, H-16), 3.59 (2H, d, *J* = 5.8 Hz, 2 H-26), 2.88 (1H, sext, *J* = 6.5 Hz, H-25), 2.60 (1H, dqd, *J* = 11.0, 7.0, 2.1 Hz, H-20), 2.31 (1H, dd, *J* = 14.0, 7.9 Hz, H-15a), 2.28 (1H, dd, *J* = 11.1, 7.5 Hz, H-17), 2.19 (1H, dq, *J* = 12.7, 7.9 Hz, H-4), 2.16 (3H, s, OAc-22), 2.08 (3H, s, OAc-16), 1.75–1.65 (3H, m, H-6, 2 H-12), 1.54 (1H, m, H-11), 1.47 (1H, m, H-7), 1.32 (1H, dd, *J* = 13.7, 4.6 Hz, H-15b), 1.20 (1H, d, *J* = 4.3 Hz, H-19a), 1.18 (3H, s, H-18), 1.12 (3H, d, *J* = 7.0 Hz, H-27), 1.10 (3H, d, *J* = 6.7 Hz, H-28), 0.95 (3H, s, H-30), 0.90 (3H, d, *J* = 6.7 Hz, H-21), 0.58 (1H, d, *J* = 4.3 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS *m*/*z* 591.3297 (calcd for $C_{34}H_{48}O_7Na$ 591.3292).

*Preparation of Compound **1c**.* To a stirred solution of **1** (53 mg, 0.093 mmol) in 1 mL of CH_2Cl_2 were added DMAP (6 mg, 0.047 mmol, 0.5 equiv), phenyl isocyanate (11 μL, 0.102 mmol, 1.1 equiv), and Et_3N (11 μL, 0.102 mmol, 1.1 equiv). The mixture was stirred at room temperature, and after 22 h, it was diluted with EtOAc and the organic solution was successively washed with 4% aqueous HCl, $NaHCO_3$, and brine. The organic solution was dried over $MgSO_4$ and concentrated under vacuum. The residue was purified by chromatography on silica gel (elution: cyclohexane–EtOAc, 1:0 to 0:1) to give **1c** as a white solid (30 mg, 47%): ^{1}H NMR (500 MHz, CD_3CN) δ 7.68 (1H, br s, H-26b), 7.41 (2H, br d, *J* = 8.3 Hz, H-26d, H-26 h), 7.29 (2H, dd, *J* = 7.6, 8.3 Hz, H-26 g, H-26e), 7.03 (1H, tt, *J* = 7.6, 1.2 Hz, H-26f), 6.94 (1H, d, *J* = 10.1 Hz, H-1), 6.15 (1H, s, H-24a), 6.02 (1H, d, *J* = 0.6 Hz, H-24a), 5.90 (1H, d, *J* = 10.1 Hz, H-2), 5.54 (1H, d, *J* = 2.1 Hz, H-22), 5.09 (1H, dt, *J* = 4.4, 7.7 Hz, H-16), 4.13 (1H, dd, *J* = 10.7, 7.0 Hz, H-26a), 4.06 (1H, dd, *J* = 10.7, 6.4 Hz, H-26b), 3.07 (1H, sext, *J* = 7.0 Hz, H-25), 2.61 (1H, ddq, *J* = 2.3, 10.9, 7.0 Hz, H-20), 2.30 (1H, dd, *J* = 11.0, 7.3 Hz, H-17), 2.10 (3H, s, OAc-22), 2.03 (3H, s, OAc-16), 1.55 (1H, ddd, *J* = 6.6, 9.0, 14.8 Hz, H-11b), 1.37 (1H, dd, *J* = 13.9, 4.1 Hz, H-15b), 1.24 (1H, d, *J* = 4.6 Hz, H-19a), 1.18 (3H, s, H-18), 1.11 (3H, d, *J* = 7.0 Hz, H-27), 1.02

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34–47

(3H, d, J = 6.7 Hz, H-28), 0.95 (3H, s, H-30), 0.85 (3H, d, J = 7.0 Hz, H-21), 0.57 (1H, d, J = 4.6 Hz, H-19b); ^{13}C NMR (125 MHz, CD_3CN) δ 202.4 (C-3), 198.8 (C-23), 171.8 (C-22a), 171.3 (C-16), 155.5 (C-1), 149.2 (C-24), 139.9 (C-26c), 129.9 (C-26e, 26 g), 128.4 (C-2), 125.7 (C-24a), 123.9 (C-26f), 119.5 (C-26d, 26 h), 78.5 (C-22), 76.7 (C-16), 68.5 (C-26), 51.3 (C-17), 48.3 (C-14), 47.6 (C-4), 46.8 (C-13), 46.7 (C-15), 45.2 (C-8), 43.6 (C-5), 35.0 (C-25), 33.4 (C-20), 32.9 (C-12), 28.1 (C-11), 27.6 (C-19), 27.2 (C-9), 24.3 (C-6, 7), 22.1 (C-16b), 21.0 (C-22b), 20.0 (C-30), 18.3 (C-18), 17.1 (C-27), 13.4 (C-21), 11.3 (C-28); ESIMS m/z: 688.24 $[M + H]^+$.

X-ray Crystallographic Analysis of Compound 1c. Data were collected at low temperature (180 K) on a Bruker Kappa Apex II diffractometer using graphite-monochromated Mo Kα radiation (λ = 0.71073 Å) and equipped with an Oxford Cryosystems Cryostream cooler device. The structure was solved by direct methods using SHELXS-86[20] and refined by means of least-squares procedures on F^2 with the aid of the program SHELXL97[20] included in the software package WinGX version 1.63.[21] The atomic scattering factors were taken from International Tables for X-ray Crystallography.[22] All hydrogen atoms were placed geometrically and refined by using a riding model, except for the atoms H30A and H30B, which were located by Fourier differences. All non-hydrogen atoms were anisotropically refined, and in the last cycles of refinement, a weighting scheme was used, where weights were calculated from the following formula: $w = 1/[\sigma^2(F_o^2) + (aP)^2 + bP]$ where $P = (F_o^2 + 2F_c^2)/3$. Drawing was performed with the program ORTEP32 with 30% probability displacement ellipsoids for non-hydrogen atoms.[23]

Crystal Data for ***1c****:* orthorhombic, $C_{41}H_{53}NO_8$, space group $P2_12_12_1$ with a = 11.5565(7) Å, b = 14.6892(10) Å, c = 22.0210(15) Å, V = 3738.2(4) Å^3, Z = 4, D_{calcd} 1.222 g/cm^3, m = 0.084 mm^{-1}, $F(000)$ = 1480. Crystal size: 0.35 × 0.2 × 0.08 mm^3. Independent reflections: 6975 with R_{int} = 0.0333. The final agreement factors are R1 = 0.0394 and wR2 = 0.0972 [$I > 2\sigma(I)$]. Crystallographic data are deposited at the Cambridge Crystallographic Data center (deposition no. CCDC 826141).

*22-De-O-acetylneoboutomellerone (**2**):* $[\alpha]^{20}_D$ −55 (c 0.16, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 228 (3.71), 266 (3.86) nm; IR (film) ν_{max} 3453, 1725, 1661, 1601, 1245, 1043 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.85 (1H, d, J = 10.1 Hz, H-1), 6.18 (1H, s, H-24a), 6.02 (1H, s, H-24a), 5.99 (1H, d, J = 10.1 Hz, H-2), 5.35 (1H, dt, J = 4.6, 7.5 Hz, H-16), 4.74 (1H, dd, J = 5.5, 1.5 Hz, H-22), 3.65 (2H, d, J = 6.1 Hz, H-26), 3.52 (1H, d, J = 5.8 Hz, OH-22), 2.96 (1H, sext, J = 6.7 Hz, H-25), 2.52 (1H, dd, J = 11.0, 7.3 Hz, H-17), 2.46 (1H, m, H-20), 2.35 (1H, dd, J = 14.0, 7.9 Hz, H-15a), 2.19 (1H, dq, J = 12.5, 6.7 Hz, H-4), 2.10 (3H, s, OAc-16), 2.05 (1H, m, H-11a), 1.99 (1H, dt, J = 5.2, 13.4 Hz, H-5), 1.97 (1H, dd, J = 10.5, 6.9 Hz, H-8a), 1.65 (1H, m, H-12a), 1.34 (1H, dd, J = 14.0, 4.4 Hz, H-15b), 1.18 (1H, d, J = 4.5 Hz, H-19a), 1.18 (3H, s, H-18), 1.14 (3H, d, J = 7.0 Hz, H-27), 1.10 (3H, d, J = 7.0 Hz, H-28), 0.99 (3H, s, H-30), 0.88 (1H, m, H-6b), 0.66 (3H, d, J = 7.0 Hz, H-21), 0.57 (1H, d, J = 4.5 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 549.3179 (calcd for $C_{32}H_{46}O_6Na$ 549.3187).

Preparation of Mosher Ester Derivatives 1a, 1b, 2a, and 2b. Compounds **1** and **2** (10 mg) in dry pyridine (2.0 mL) were transferred into an HPLC vial (2 mL) capped with a septum and were treated with 30 μL (175 μmol) of (*S*)-(+)-α-methoxy-α-trifluoromethylphenylacetyl chloride (MTPA chloride). The mixtures were stirred at room temperature for 2 h. After concentration under reduced pressure, they were purified by semipreparative HPLC RP-18 chromatography, eluting with a linear gradient (55−100% B) to afford **1a** and **2a**. Derivatives **1b** and **2b** were prepared in a similar fashion by using (*R*)-(−)-α-methoxy-α-trifluoromethylphenylacetyl chloride.

*1-(R)-MTPA ester (**1a**):* ^{1}H NMR (CD_3CN, 500 MHz) δ 4.39 (1H, dd, J = 6.2, 10.9 Hz, H-26a), 4.27 (1H, dd, J = 5.7, 10.9 Hz, H-26b); ESIMS m/z 807 $[M + Na]^+$.

*1-(S)-MTPA ester (**1b**):* ^{1}H NMR (CD_3CN, 500 MHz) δ 4.34 (2H, AB part of an ABX system, H-26); ESIMS m/z 807 $[M + Na]^+$.

*2-(R)-MTPA ester (**2a**):* ^{1}H NMR (methanol-d_4, 500 MHz) δ 4.44 (1H, dd, J = 6.0, 10.9 Hz, H-26a), 4.35 (1H, dd, J = 5.1, 10.9 Hz, H-26b), 2.59 (1H, m, H-20); ESIMS m/z 981 $[M + Na]^+$.

*2-(S)-MTPA ester (**2b**):* ^{1}H NMR (methanol-d_4, 500 MHz) δ 4.40 (2H, m, H-26), 2.65 (1H, m, H-20); ESIMS m/z 981 $[M + Na]^+$.

*26-Acetylneoboutomellerone (**3**):* yellow gum; $[\alpha]^{20}_D$ −61 (c 0.05, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 222 (3.78), 266 (3.74) nm; IR (film) ν_{max} 3299, 2933, 2874, 1736, 1699, 1596, 1372, 1230, 1022 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.94 (1H, d, J = 10.1 Hz, H-1), 6.10 (1H, s, H-24a), 5.95 (1H, d, J = 0.6 Hz, H-24a), 5.88 (1H, d, J = 10.1 Hz, H-2), 5.52 (1H, d, J = 2.1 Hz, H-22), 5.09 (1H, dt, J = 4.6, 7.6 Hz, H-16), 4.04 (1H, dd, J = 7, 11 Hz, H-26a), 3.99 (1H, dd, J = 6.5, 11 Hz, H-26b), 3.00 (1H, sext, J = 6.9 Hz, H-25), 2.57 (1H, ddq, J = 11.0, 2.1, 7.0 Hz, H-20), 2.28 (1H, dd, J = 11.0, 7.6 Hz, H-17), 2.08 (3H, s, OAc-22), 2.05 (3H, s, OAc-16), 1.96 (3H, s, OAc-26), 1.56 (1H, ddd, J = 6.3, 8.5, 15 Hz, H-11b), 1.45 (1H, m, H-7a), 1.37 (1H, dd J = 14.5, 4.5 Hz, H-15b), 1.24 (1H, d, J = 4.6 Hz, H-19a), 1.19 (1H, m, H-7b), 1.18 (3H, s, H-18), 1.07 (3H, d, J = 7.0 Hz, H-27), 1.02 (3H, d, J = 6.7 Hz, H-28), 0.95 (3H, s, H-30), 0.94 (1H, m, H-6b), 0.84 (3H, d, J = 7.0 Hz, H-21), 0.57 (1H, d, J = 4.6 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 633.3399 (calcd for $C_{36}H_{50}O_8Na$ 633.3398).

Preparation of Compound ***3****.* To a stirred solution of compound **2** (50 mg, 0.088 mmol) in 4 mL of CH_2Cl_2 at 0 °C were added pyridine (300 μL, 3.7 mmol) and acetyl chloride (60 μL, 0.88 mmol, 10 equiv). The mixture was stirred overnight, MeOH (0.5 mL) was added, and the mixture was concentrated under vacuum. The crude material was purified by chromatography on silica gel (elution: from cyclohexane−EtOAc, 7:3 to 6:4) to yield **3** (52 mg, 75%), identical by all means with the natural product.

*Preparation of 22,23,26-Trihydroxy-29-nor-24-methyl-16,22-epoxycycloarta-1,24(24a)-diene-3,23-dione (**4**).* To a stirred solution of compound **1** (20 mg, 0.038 mmol) in CH_3CN (300 μL) was added NaOH (570 μL, 0.437 mmol, c = 1 mol/L, 15 equiv). The mixture was stirred for 22 h at room temperature, then diluted with EtOAc, and filtered over Celite. After concentration under vacuum, the product was purified by chromatography on silica gel (elution: cyclohexane−EtOAc, 1:1) to obtain **4** as a translucent film (3 mg, 17%): ^{1}H NMR (CD_3CN, 500 MHz) δ 6.96 (1H, d, J = 10.1 Hz, H-1), 5.91 (1H, d, J = 10.1 Hz, H-2), 5.24 (1H, s, H-24a), 5.11 (1H, s, H-24a), 4.66 (1H, dt, J = 7.2, 8.5 Hz, H-16), 3.97 (1H, d, J = 5.6 Hz, H-23), 3.79 (1H, d, J = 5.6 Hz, OH-23), 3.48 (1H, s, OH-22), 3.47 (1H, m, H-26a), 3.40 (1H, ddd, J = 9.6, 8.7, 4.6 Hz, H-26b), 3.23 (1H, t, J = 4.7 Hz, OH-26), 2.63 (1H, sext, J = 7.0 Hz, H-25), 2.47 (1H, quint, J = 6.8 Hz, H-20), 1.31 (1H, d, J = 4.6 Hz, H-19a), 1.27 (1H, m, H-7b), 1.13 (3H, s, H-18), 1.03 (3H, d, J = 6.7 Hz, H-28), 1.00 (3H, d, J = 7.0 Hz, H-27), 0.95 (3H, d, J = 6.7 Hz, H-21), 0.92 (3H, s, H-30), 0.50 (1H, d, J = 4.6 Hz, H-19b); ^{13}C NMR, see Table 1; ESIMS m/z 507.3 $[M + Na]^+$, 991.6 $[2M + Na]^+$.

*1,2-Dihydroneoboutomellerone (**5**):* yellow gum; $[\alpha]^{20}_D$ +36 (c 0.08, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 216 (3.78), 290 (2.63) nm; IR (film) ν_{max} 3513, 2965, 2871, 1734, 1705, 1452, 1376, 1231 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.05 (1H, s, H-24a), 5.90 (1H, d, J = 0.9 Hz, H-24a), 5.53 (1H, d, J = 2.1 Hz, H-22), 5.08 (1H, dt, J = 4.3, 7.6 Hz, H-16), 3.53 (1H, dt, J = 10.6, 5.8 Hz, H-26a), 3.39 (1H, dt, J = 10.6, 6.1 Hz, H-26b), 2.77 (1H, sext, J = 6.7 Hz, H-25), 2.68 (1H, t, J = 5.6 Hz, OH-26), 2.60 (1H, ddq, J = 10.7, 1.8, 6.7 Hz, H-20), 2.43 (1H, td, J = 13.6, 6.4 Hz, H-2a), 2.20 (1H, dd, J = 14.0, 7.9 Hz, H-15a), 2.09 (3H, s, OAc-22), 2.11 (1H, m, H-11a), 2.03 (3H, s, OAc-16), 1.82 (1H, m, H-1a), 1.60−1.55 (2H, m, H-1b, H-5), 1.36 (1H, dd, J = 14.5, 4.1 Hz, H-15b), 1.23 (3H, s, H-18), 1.10 (1H, m, H-7b), 1.03 (3H, d, J = 7.0 Hz, H-27), 0.97 (3H, s, H-30), 0.91 (3H, d, J = 6.7 Hz, H-28), 0.84 (3H, d, J = 7.0 Hz, H-21), 0.75 (1H, dq, J = 2.4, 12.6 Hz, H-6b), 0.65 (1H, br d, J = 4.0 Hz, H-19a), 0.47 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 593.3451 (calcd for $C_{34}H_{50}O_7Na$ 593.3449).

*1,2-Dihydro-22-de-O-acetylneoboutomellerone (**6**):* yellow gum; $[\alpha]^{20}_D$ +11 (c 0.12, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 214 (3.74), 262 (3.23) nm; IR (film) ν_{max} 3435, 2932, 2873, 1731, 1705, 1700, 1452, 1376, 1237, 1022, 997 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.12 (1H, s, H-24a), 5.99 (1H, d, J = 0.9 Hz, H-24a), 5.20 (1H, dt, J = 4.4, 7.6 Hz, H-16), 4.72 (1H, dd, J = 6.1, 2.1 Hz, H-22), 3.56 (1H, dt, J = 10.5, 5.8 Hz, H-26a), 3.54 (1H, d, J = 6.1 Hz, OH-22), 3.41 (1H, dt,

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34−47

J = 10.5, 5.8 Hz, H-26b), 2.83 (1H, sext, J = 6.7 Hz, H-25), 2.67 (1H, t, J = 5.8 Hz, OH-26), 2.07 (1H, m, H-11a), 2.03 (3H, s, OAc-16), 1.82 (1H, m, H-1a), 1.37 (1H, ddd, J = 14.2, 4.4, 0.9 Hz, H-15b), 1.22 (3H, s, H-18), 1.12 (1H, dq, J = 2.7, 12.8 Hz, H-7b), 1.05 (3H, d, J = 7.3 Hz, H-27), 0.97 (3H, d, J = 0.6 Hz, H-30), 0.91 (3H, d, J = 6.7 Hz, H-28), 0.75 (1H, dq, J = 2.4, 12.5 Hz, H-6b), 0.65 (1H, m, H-19a), 0.64 (3H, d, J = 6.4 Hz, H-21), 0.47 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 551.3342 (calcd for $C_{32}H_{48}O_6Na$ 551.3343).

*6β-Hydroxyneoboutomellerone (**7**):* yellow gum; $[\alpha]^{20}_D$ −43 (c 0.13, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 212 (3.84), 220 (3.84), 266 (3.87) nm; IR (film) ν_{max} 3409, 2937, 2876, 1733, 1662, 1454, 1374, 1232, 1022 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.76 (1H, d, J = 10 Hz, H-1), 6.14 (1H, s, H-24a), 5.96 (1H, d, J = 10 Hz, H-2), 5.90 (1H, s, H-24a), 5.59 (1H, d, J = 2.1 Hz, H-22), 5.10 (1H, dt, J = 4.4, 7.7 Hz, H-16), 4.20 (1H, br s, H-6), 3.60 (2H, d, J = 5.8 Hz, H-26), 2.89 (1H, sext, J = 6.4 Hz, H-25), 2.35 (1H, dd, J = 14.0, 7.9 Hz, H-15a), 2.20 (1H, m, H-11a), 2.16 (3H, s, OAc-22), 2.08 (3H, s, OAc-16), 1.99 (1H, m, H-5), 1.86 (1H, d, J = 3.7 Hz, H-19a), 1.76 (2H, m, 2 H-12), 1.56 (1H, m, H-7a), 1.48 (1H, dt, J = 1.7, 13.3 Hz, H-7b), 1.25 (3H, s, H-18), 1.24 (3H, d, J = 7.0 Hz, H-28), 1.13 (3H, d, J = 7.0 Hz, H-27), 1.0 (3H, s, H-30), 0.91 (3H, d, J = 6.7 Hz, H-21), 0.72 (1H, d, J = 3.7 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 607.3243 (calcd for $C_{34}H_{48}O_8Na$ 607.3241).

*6β-Hydroxy-22-de-O-acetylneoboutomellerone (**8**):* yellow gum; $[\alpha]^{20}_D$ −8 (c 0.14, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (3.94), 266 (3.78) nm; IR (film) ν_{max} 3435, 2934, 2876, 1729, 1666, 1453, 1375, 1236, 1023 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz)) δ 6.76 (1H, d, J = 10 Hz, H-1), 6.18 (1H, s, H-24a), 6.03 (1H, s, H-24a), 5.94 (1H, d, J = 10 Hz, H-2), 5.35 (1H, dt, J = 4.4, 7.1 Hz, H-16), 4.74 (1H, s, H-22), 4.19 (1H, br s, H-6), 3.64 (2H, d, J = 6.1 Hz, H-26), 3.59 (1H, d, J = 5.5 Hz, OH-22), 2.96 (1H, sext, J = 6.4 Hz, H-25), 2.58 (1H, m, H-4), 2.52 (1H, m, H-17), 2.45 (1H, m, H-20), 2.37 (1H, dd, J = 13.7, 7.9 Hz, H-15a), 2.30 (1H, dd, J = 12.7, 4.4 Hz, H-8), 2.22 (1H, m, H-11a), 2.09 (3H, s, OAc-16), 1.99 (1H, dd, J = 12.8, 2.4 Hz, H-5), 1.84 (1H, d, J = 3.1 Hz, H-19a), 1.75 (2H, m, 2 H-12), 1.58 (1H, dt, J = 4.4, 12.9 Hz, H-7a), 1.48 (1H, t, J = 13.1 Hz, H-7b), 1.40−1.20 (2H, m, H-15b, H-11b), 1.24 (3H, d, J = 7.3 Hz, H-28), 1.23 (3H, br s, H-18), 1.14 (3H, d, J = 7.0 Hz, H-27), 1.04 (3H, s, H-30), 0.70 (1H, d, J = 3.1 Hz, H-19b), 0.67 (3H, d, J = 6.4 Hz, H-21); ^{13}C NMR, see Table 1; HR-ESIMS m/z 565.3133 (calcd for $C_{32}H_{46}O_7$ Na 565.3136).

*18-Hydroxyneoboutomellerone (**9**):* yellow gum; $[\alpha]^{20}_D$ −16 (c 0.03, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 214 (3.14), 266 (2.96) nm; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.80 (1H, d, J = 10 Hz, H-1), 6.07 (1H, s, H-24a), 5.98 (1H, d, J = 10 Hz, H-2), 5.86 (1H, br d, H-24), 5.57 (1H, d, J = 2.1 Hz, H-22), 5.18 (1H, dt, J = 4.3, 7.6 Hz, H-16), 4.06 (1H, m, H-18a), 3.94 (1H, d, J = 12.2 Hz, H-18b), 3.58 (2H, m, 2 H-26), 2.86 (1H, sext, J = 7.0 Hz, H-25), 2.72 (1H, m, H-20), 2.36 (1H, dd, J = 11.3, 7.6 Hz, H-17), 2.28 (1H, dd, J = 14.3, 7.9 Hz, H-15a), 2.19 (1H, m, H-4), 2.14 (3H, s, OAc-22), 2.09 (3H, s, OAc-16), 2.03 (2H, m, H-5, H-8), 1.85 (1H, m, H-12a), 1.70 (1H, m, H-6a), 1.63 (1H, m, H-12b), 1.60 (1H, m, H-15b), 1.35 (2H, m, H-7a, H-11b), 1.19 (1H, d, J = 4.3 Hz, H-19a), 1.16 (1H, m, H-7b), 1.11 (3H, d, J = 7.3 Hz, H-27), 1.09 (3H, d, J = 6.9 Hz, H-28), 1.00 (3H, s, H-30), 0.93 (3H, d, J = 6.7 Hz, H-21), 0.90 (1H, m, H-6b), 0.60 (1H, d, J = 4.3 Hz, H-19b); ^{13}C NMR, see Table 2; HR-ESIMS m/z 607.3216 (calcd for $C_{34}H_{48}O_8Na$ 607.3241).

*6β,7β-Oxidoneoboutomellerone (**10**):* yellow gum; $[\alpha]^{20}_D$ −86 (c 0.16, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 224 (3.92), 262 (3.93) nm; IR (film) ν_{max} 3411, 2966, 2878, 1735, 1669, 1375, 1233, 1022 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.94 (1H, d, J = 10 Hz, H-1), 6.06 (1H, s, H-24a), 5.91 (1H, d, J = 10.0 Hz, H-2), 5.90 (1H, br s, H-24a), 5.54 (1H, d, J = 2.1 Hz, H-22), 5.16 (1H, dt, J = 4.7, 7.7 Hz, H-16), 3.53 (1H, dd, J = 10.4, 6.4 Hz, H-26a), 3.39 (1H, dd, J = 10.4, 6.6 Hz, H-26b), 3.15 (1H, dd, J = 4.3, 1.5 Hz, H-6), 3.00 (1H, dd, J = 4.3, 1.8 Hz, H-7), 2.83 (1H, br d, J = 1.2 Hz, H-8), 2.77 (1H, sext, J = 6.7 Hz, H-25), 2.70 (1H, br s, OH-26), 2.63 (1H, ddq, J = 10.8, 2.1, 7.0 Hz, H-20), 2.55 (1H, dq, J = 12.5, 7.0 Hz, H-4), 2.45 (1H, br d, J = 11.3 Hz, H-5), 2.33 (1H, dd, J = 13.4, 7.9 Hz, H-15a), 2.28 (1H, dd, J = 10.8, 7.8 Hz, H-17), 2.08 (3H, s, OAc-22), 2.06 (3H, s, OAc-16), 2.04 (2H, m, H-11a, H-19a), 1.40 (1H, ddd, J = 15.9, 3.1, 1.6 Hz, H-11b), 1.23 (3H, d, J = 7.0 Hz, H-28), 1.20 (3H, s, H-18), 1.03 (3H, d, J = 7.0 Hz, H-27), 0.94 (3H, s, H-30), 0.86 (3H, d, J = 7.0 Hz, H-21), 0.03 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 2; HR-ESIMS m/z 605.3091 (calcd for $C_{34}H_{46}O_8Na$ 605.3085).

*1,2-Dihydro-1α-hydroxy-22-de-O-acetylneoboutomellerone (**11**):* yellow gum; $[\alpha]^{20}_D$ +30 (c 0.12, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 216 (3.920) nm; IR (film) ν_{max} 3401, 2934, 2876, 1707, 1669, 1376, 1237, 1021 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.12 (1H, s, H-24a), 5.99 (1H, d, J = 0.9 Hz, H-24a), 5.21 (1H, dt, J = 4.3, 7.5 Hz, H-16), 4.72 (1H, dd, J = 5.3, 1.7 Hz, H-22), 3.80 (1H, t, J = 3.1 Hz, H-1), 3.55 (2H, dd, J = 10.9, 6.1 Hz, H-26a + OH-22), 3.42 (1H, dd, J = 10.9, 7 Hz, H-26b), 2.83 (1H, sext, J = 6.4 Hz, H-25), 2.80 (1H, br s, OH-1), 2.69 (1H, br s, OH-26), 2.64 (1H, ddd, J = 14.0, 3.7, 0.5 Hz, H-2a), 2.46 (1H, ddq, J = 11.0, 2.1, 6.7 Hz, H-20), 2.42 (1H, dd, J = 11.0, 7.0 Hz, H-17), 2.14 (1H, m, H-5), 2.03 (3H, s, OAc-16), 1.74 (1H, m, H-6a), 1.21 (3H, s, H-18), 1.05 (3H, d, J = 7.0 Hz, H-27), 1.01 (3H, s, H-30), 0.92 (3H, d, J = 6.4 Hz, H-28), 0.80 (1H, dq, J = 2.4, 12.6 Hz, H-6b), 0.73 (1H, d, J = 4.3 Hz, H-19a), 0.64 (3H, d, J = 6.6 Hz, H-21), 0.48 (1H, d, J = 4.3 Hz, H-19b), ^{13}C NMR, see Table 2; HR-ESIMS m/z 567.3296 (calcd for $C_{32}H_{48}O_7Na$ 567.3292).

*25-Epi-neoboutomellerone (**12**):* yellow gum; $[\alpha]^{20}_D$ +9 (c 0.03, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (3.54), 264 (3.54) nm; IR (film) ν_{max} 3386, 2927, 1732, 1667, 1570, 1375, 1233, 1022 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.82 (1H, d, J = 9.8 Hz, H-1), 6.11 (1H, s, H-24a), 5.98 (1H, d, J = 9.8 Hz, H-2), 5.83 (1H, d, J = 0.9 Hz, H-24a), 5.52 (1H, d, J = 2.1 Hz, H-22), 5.07 (1H, dt, J = 4.4, 7.7 Hz, H-16), 3.56 (2H, t, J = 6.0 Hz, 2 H-26), 2.96 (1H, sext, J = 6.7 Hz, H-25), 2.59 (1H, m, H-20), 2.28 (1H, dd, J = 7.4, 14.4 Hz, H-17), 2.25 (1H, m, H-15a), 2.18 (1H, m, H-4), 2.14 (3H, s, OAc-22), 2.06 (3H, s, OAc-16), 1.45 (1H, m, H-7), 1.30 (1H, dd, J = 14.2, 4.7 Hz, H-15b), 1.18 (1H, d, J = 4.6 Hz, H-19a),1.16 (3H, s, H-18), 1.09 (3H, d, J = 7.0 Hz, H-27), 1.08 (3H, d, J = 6.7 Hz, H-28), 0.93 (3H, s, H-30), 0.89 (3H, d, J = 7.0 Hz, H-21), 0.55 (1H, d, J = 4.6 Hz, H-19); ^{13}C NMR, see Table 2; HR-ESIMS m/z 591.3291 (calcd for $C_{34}H_{48}O_7Na$, 591.3292).

*Preparation of 25ξ-Neoboutomelleron-26-al (**13**).* To a stirred solution of **1** (100 mg, 0.18 mmol) in CH_2Cl_2 (3.5 mL) was added 350 μL of pyridine (2 mL/mmol), and the mixture was cooled to 0 °C. The Dess-Martin reagent (1.76 mL, 0.528 mmol, c = 3 mol/L, 3 equiv) was added dropwise, and the mixture was allowed to warm. After 3 h, the mixture was diluted with EtOAc, and a saturated solution of $Na_2S_2O_3$ was added. The aqueous layer was extracted three times with EtOAc (10 mL), and the combined organic layers were washed successively with $NaHCO_3$ (10 mL) and brine. The organic solution was dried over $MgSO_4$ and concentrated under vacuum. The residue was purified by chromatography on silica gel (cyclohexane−EtOAc, 7:3 to 6:4) to give **13** (66 mg, 66%). ^{1}H NMR (500 MHz, CD_3CN) δ 9.52 (1H, d, J = 0.6 Hz, H-26), 6.94 (1H, d, J = 10.1 Hz, H-1), 6.31 (1H, s, H-24a), 6.06 (1H, s, H-24), 5.90 (1H, d, J = 10.1 Hz, H-2), 5.58 (1H, d, J = 2.1 Hz, H-22), 5.10 (1H, dt, J = 4.4, 7.7 Hz, H-16), 3.45 (1H, q, J = 7.2 Hz, H-25), 2.64 (1H, m, H-20), 2.31 (1H, dd, J = 11.0, 7.6 Hz, H-17), 2.09 (3H, s, OAc-22), 2.04 (3H, s, OAc-16), 1.38 (1H, dd, J = 13.7, 4.0 Hz, H-15b), 1.25 (1H, d, J = 4.4 Hz, H-19a), 1.18 - 1.22 (7H, m, H-7, -27, -18), 1.03 (3H, d, J = 6.7 Hz, H-28), 0.96 (3H, s, H-30), 0.94 (1H, qd, J = 12.8, 3.7 Hz, H-6b), 0.85 (3H, d, J = 7.0 Hz, H-21), 0.58 (1H, d, J = 4.4 Hz, H-19b);); ^{13}C NMR, see Table 2; ESI-MS m/z 567.3 [M + H]$^+$.

*9,10-Di-epi-25ξ-neoboutomellerone (**14**):* yellow gum; $[\alpha]^{20}_D$ −48 (c 0.13, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (3.73), 270 (3.93) nm; IR (film) ν_{max} 3417, 2937, 2875, 1734, 1666, 1373, 1233, 1022 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.39 (1H, d, J = 10.4 Hz, H-1), 6.13 (1H, s, H-24a), 5.99 (1H, d, J = 10.4 Hz, H-2), 5.88 (1H, d, J = 0.6 Hz, H-24a), 5.58 (1H, d, J = 2.1 Hz, H-22), 5.15 (1H, dt, J = 5.0, 7.7 Hz, H-16), 3.60 (2H, t, J = 5.5 Hz, 2 H-26), 2.88 (1H, sext, J = 6.4 Hz, H-25), 2.62 (1H, m, H-20), 2.28 (1H, dd, J = 11.9, 7.3 Hz, H-17), 2.26 (1H, m, H-15a), 2.20 (1H, m, H-4), 2.16 (3H, s, OAc-22), 2.10 (3H, s, OAc-16), 1.96 (1H, m, H-8), 1.85 (1H, dt, J = 5.0, 13.0 Hz, H-12a), 1.6 (1H, dd, J = 13.0, 4.1 Hz, H-12a), 1.28 (1H, d, J = 5.0 Hz, H-19a), 1.14 (3H, s, H-18), 1.12 (3H, d, J = 6.1 Hz, H-28), 1.10 (3H, s, H-27), 0.93 (3H, br s, H-30), 0.91 (3H, d, J = 7.0 Hz, H-21), 0.88

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34−47

(1H, d, J = 5.0 Hz, H-19b); ^{13}C NMR, see Table 2; HR-ESIMS m/z 591.3295 (calcd for $C_{34}H_{48}O_7Na$, 591.3292).

9,10-Di-epi-22-de-O-acetyl-25ξ-neoboutomellerone (**15**): yellow gum; $[\alpha]^{20}_D$ −53 (c 0.13, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (4.12), 270 (4.20) nm; IR (film) ν_{max} 3421, 2937, 2877, 1732, 1666, 1609, 1453, 1376, 1240, 1024 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.39 (1H, d, J = 10.1 Hz, H-1), 6.19 (1H, s, H-24a), 6.02 (1H, s, H-24a), 5.99 (1H, d, J = 10.1 Hz, H-2), 5.39 (1H, dt, J = 5.0, 7.6 Hz, H-16), 4.73 (1H, br s, H-22), 3.64 (1H, d, J = 5.8 Hz, 2 H-26), 3.54 (1H, br s, OH-22), 2.96 (1H, sext, J = 6.7 Hz, H-25), 2.53 (1H, dd, J = 11.0, 7.6 Hz, H-17), 2.48 (1H, m, H-20), 2.31 (1H, dd, J = 13.3, 7.8 Hz, H-15), 2.11 (3H, s, OAc-16), 1.84 (1H, dt, J = 4.7,, 13.4 Hz, H-12), 1.60 (1H, ddd, J = 13.4, 4.0, 1.5 Hz, H-12b), 1.33 (1H, m, H-15), 1.28 (1H, d, J = 4.9 Hz, H-19a), 1.13 (6H, d, J = 7.0 Hz, H-27, H-28); 1.12 (3H, s, H-18), 0.97 (3H, s, H-30), 0.88 (1H, d, J = 4.9 Hz, H-19b), 0.67 (3H, d, J = 6.4 Hz, H-21); ^{13}C NMR, see Table 2; HR-ESIMS m/z 549.3190 (calcd for $C_{32}H_{46}O_6Na$, 549.3187).

26-Deoxyneoboutomellerone (**16**): yellow gum; $[\alpha]^{20}_D$ −102 (c 0.08, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.95), 264 (3.77) nm; IR (film) ν_{max} 3437, 2962, 2931, 2873, 1734, 1669, 1599, 1455, 1407, 1237 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.94 (1H, d, J = 10 Hz, H-1), 5.94 (1H, s, H-24a), 5.90 (1H, d, J = 10.0 Hz, H-2), 5.83 (1H, d, J = 1.2 Hz, H-24a), 5.51 (1H, d, J = 2.4 Hz, H-22), 5.09 (1H, dt, J = 4.3, 7.6 Hz, H-16), 2.81 (1H, dsept, J = 0.9, 7.0 Hz, H-25), 2.56 (1H, ddq, J = 11.0, 2.1, 7.0 Hz, H-20), 2.29 (1H, dd, J = 11.0,, 7.3 Hz, H-17), 2.18 (2H, m, H-4, H-15a), 2.09 (3H, s, OAc-22), 2.02 (3H, s, OAc-16), 1.56 (1H, ddd, J = 7.0, 9.0, 15.0 Hz, H-11b), 1.44 (1H, ddt, J = 7.0, 13.5, 4.0 Hz, H-7a), 1.36 (1H, ddd, J = 14.0, 4.3, 0.9 Hz, H-15b), 1.24 (1H, d, J = 4.6 Hz, H-19a), 1.18 (3H, s, H-18), 1.07 (3H, d, J = 6.7 Hz, H-26), 1.02 (3H, d, J = 7.0 Hz, H-28), 1.00 (3H, d, J = 7.0 Hz, H-27), 0.95 (3H, s, H-30), 0.85 (3H, d, J = 7.0 Hz, H-21), 0.58 (1H, d, J = 4.6 Hz, H-19b); ^{13}C NMR, see Table 2; HR-ESIMS m/z 575.3341 (calcd for $C_{34}H_{48}O_6Na$, 575.3343).

22-De-O-acetyl-26-deoxyneoboutomellerone (**17**): yellow gum; $[\alpha]^{20}_D$ −91 (c 0.12, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.87), 266 (3.86) nm; IR (film) ν_{max} 3457, 2933, 2931, 2873, 1734, 1669, 1456, 1375, 1237 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.93 (1H, d, J = 10.0 Hz, H-1), 6.02 (1H, s, H-24a), 5.93 (1H, d, J = 1.2 Hz, H-24a), 5.89 (1H, d, J = 10.0 Hz, H-2), 5.20 (1H, dt, J = 4.4, 7.6 Hz, H-16), 4.71 (1H, d, J = 5.8 Hz, H-22), 3.54 (1H, d, J = 5.8 Hz, OH-22), 2.85 (1H, dsept, J = 0.9, 6.9 Hz, H-25), 2.42 (2H, m, H-17, H-20), 2.22 (1H, dd, J = 13.9, 7.8 Hz, H-15a), 2.17 (1H, dq, J = 12.7, 6.9 Hz, H-4), 2.02 (3H, s, OAc-16), 1.54 (1H, ddd, J = 7.0, 8.5, 15.0 Hz, H-11), 1.45 (1H, ddt, J = 7.0, 14.0, 4.0 Hz, H-7a), 1.37 (1H, ddd, J = 13.9, 4.4, 1.0 Hz, H-15), 1.23 (1H, d, J = 4.3 Hz, H-19a), 1.17 (3H, s, H-18), 1.09 (3H, d, J = 7.0 Hz, H-26), 1.03 (3H, d, J = 6.7 Hz, H-28), 1.02 (3H, d, J = 6.7 Hz, H-27), 0.96 (3H, d, J = 0.6 Hz, H-30), 0.64 (3H, d, J = 6.4 Hz, H-21), 0.57 (1H, d, J = 4.3 Hz, H-19b); ^{13}C NMR, see Table 3; HR-ESIMS m/z 533.3234 (calcd for $C_{32}H_{46}O_5Na$, 533.3237).

6β-Hydroxy-26-deoxyneoboutomellerone (**18**): yellow gum; $[\alpha]^{20}_D$ −61 (c 0.12, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.83), 266 (3.74) nm; IR (film) ν_{max} 3414, 2965, 2939, 2876, 1737, 1668, 1600, 1376, 1232 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.84 (1H, d, J = 9.9 Hz, H-1), 5.95 (1H, s, H-24a), 5.84 (1H, d, J = 1.2 Hz, H-24a), 5.82 (1H, d, J = 9.9 Hz, H-2), 5.53 (1H, d, J = 2.1 Hz, H-22), 5.10 (1H, dt, J = 4.3, 7.6 Hz, H-16), 4.05 (1H, br s, H-6), 2.81 (1H, dsept, J = 1.0, 6.7 Hz, H-25), 2.61 (1H, d, J = 3.4 Hz, OH-6), 2.57 (1H, ddq, J = 2.2, 10.0, 6.9 Hz, H-20), 2.48 (1H, dq, J = 13.1, 6.7 Hz, H-4), 2.32 (1H, dd, J = 11.0, 7.6 Hz, H-17), 2.29 (1H, dd, J = 12.8, 4.9 Hz, H-8a), 2.22 (1H, dd, J = 13.7, 7.9 Hz, H-15a), 2.21 (1H, m, H-11), 2.10 (3H, s, OAc-22), 2.03 (3H, s, OAc-16), 1.81 (1H, d, J = 3.1 Hz, H-19a), 1.75 (2H, m, 2 H-12), 1.52 (1H, dt, J = 13.4, 4.7 Hz, H-7), 1.24 (3H, s, H-18), 1.14 (3H, d, J = 6.7 Hz, H-28), 1.08 (3H, d, J = 6.7 Hz, H-26), 1.01 (3H, d, J = 7.0 Hz, H-27), 1.00 (3H, s, H-30), 0.86 (3H, d, J = 7.0 Hz, H-21), 0.67 (1H, d, J = 3.4 Hz, H-19b); ^{13}C NMR, see Table 3; HR-ESIMS m/z 591.3287 (calcd for $C_{34}H_{48}O_7Na$, 591.3292).

1,2-Dihydro-22-de-O-acetyl-26-deoxyneoboutomellerone (**19**): yellow gum; $[\alpha]^{20}_D$ +19 (c 0.09, $CHCl_3$); IR (film) ν_{max} 3401, 2930, 2871, 1734, 1707, 1453, 1377, 1240 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.03 (1H, s, H-24a), 5.94 (1H, d, J = 1.1 Hz, H-24a), 5.20 (1H, td, J = 4.2, 7.7 Hz, H-16), 4.70 (1H, d, J = 1.4 Hz, H-22), 2.86 (1H, dsept, J = 0.9, 7.0 Hz, H-25), 2.03 (3H, s, OAc-16), 1.86−1.78 (2H, m, H-1, H-12), 1.22 (3H, s, H-18), 1.10 (3H, d, J = 6.7 Hz, H-27), 1.02 (3H, d, J = 6.7 Hz, H-26), 0.97 (1H, d, J = 0.6 Hz, H-30), 0.91 (3H, d, J = 6.8 Hz, H-28), 0.76 (1H, dq, J = 2.3, 12.5 Hz, H-6b), 0.64 (1H, m, H-19a), 0.64 (3H, d, J = 6.1 Hz, H-21), 0.47 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 3; HR-ESIMS m/z 535.3393 (calcd for $C_{32}H_{48}O_5Na$, 535.3394).

9,10-Di-epi-26-deoxyneoboutomellerone (**20**): yellow gum; $[\alpha]^{20}_D$ −98 (c 0.11, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 212 (4.08), 270 (4.21) nm; IR (film) ν_{max} 3451, 2936, 2873, 2876, 1734, 1667, 1609, 1452, 1374, 1238 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.50 (1H, d, J = 10.2 Hz, H-1), 5.95 (1H, s, H-24a), 5.88 (1H, d, J = 10.2 Hz, H-2), 5.82 (1H, d, J = 1.2 Hz, H-24a), 5.52 (1H, d, J = 2.0 Hz, H-22), 5.14 (1H, dt, J = 4.5, 7.7 Hz, H-16), 2.81 (1H, dsept, J = 1.0, 7.0 Hz, H-25), 2.57 (1H, ddq, J = 10.9, 2.1, 6.9 Hz, H-20), 2.29 (1H, dd, J = 10.8, 7.7 Hz, H-17), 2.22 (2H, m, H-4, H-8a), 2.09 (3H, s, OAc-22), 2.04 (3H, s, OAc-16), 2.00 (1H, m, H-7), 1.83 (1H, dt, J = 4.4, 12.5, H-12a), 1.63 (1H, ddd, J = 2.0, 5.0, 13.0 Hz, H-12b), 1.42 (1H, ddd, J = 2.2, 5.0, 14.0, H-11b), 1.36 (1H, dd, J = 4.7, 14.0 Hz, H-15b) 1.30 (1H, d, J = 4.7 Hz, H-19a), 1.13 (3H, s, H-18), 1.07 (3H, d, J = 7.0 Hz, H-26), 1.04 (3H, d, J = 6.6 Hz, H-28), 1.00 (3H, d, J = 6.9 Hz, H-27), 0.93 (3H, s, H-30), 0.92 (1H, m, H-19), 0.86 (3H, d, J = 6.9 Hz, H-21); ^{13}C NMR, see Table 3; HR-ESIMS m/z 575.3338 (calcd for $C_{34}H_{48}O_6Na$, 575.3343).

24a-Nor-24,25-didehydro-26-deoxyneoboutomellerone (**21**): yellow gum; $[\alpha]^{20}_D$ −73 (c 0.03, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 242 (3.96) nm; IR (film) ν_{max} 3433, 2932, 2931, 2874, 1735, 1671, 1615, 1378, 1234 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.94 (1H, d, J = 10.1 Hz, H-1), 6.14 (1H, sept, J = 1.3 Hz, H-24), 5.90 (1H, d, J = 10.1 Hz, H-2), 5.10 (1H, dt, J = 4.6, 7.8 Hz, H-16), 4.85 (1H, d, J = 1.8 Hz, H-22), 2.59 (1H, ddq, J = 10.9, 1.5, 7.0 Hz, H-20), 2.12 (3H, d, J = 1.2 Hz, H-27), 2.09 (3H, s, OAc-22), 2.08 (3H, s, OAc-16), 1.98 (3H, m, H-26), 1.26 (1H, d, J = 4.3 Hz, H-19a), 1.20 (3H, s, H-18), 1.02 (3H, d, J = 6.7 Hz, H-28), 0.93 (3H, d, J = 0.9 Hz, H-30), 0.85 (3H, d, J = 7.0 Hz, H-21), 0.56 (1H, d, J = 4.3 Hz, H-19b); ^{13}C NMR, see Table 1; HR-ESIMS m/z 561.3182 (calcd for $C_{33}H_{46}NaO_6$, 561.3187).

22-De-O-acetyl-24a-nor-24,25-didehydro-26-deoxyneoboutomellerone (**22**): yellow gum; $[\alpha]^{20}_D$ −80 (c 0.04, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 242 (3.95) nm; IR (film) ν_{max} 3464, 2933, 2874, 1732, 1671, 1618, 1449, 1378, 1237 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.94 (1H, d, J = 10 Hz, H-1), 6.17 (1H, sept, J = 1.3 Hz, H-24), 5.89 (1H, d, J = 10.0 Hz, H-2), 5.19 (1H, dt, J = 4.7, 7.9 Hz, H-16), 3.99 (1H, dd, J = 4.9, 1.2 Hz, H-22), 3.54 (1H, d, J = 5.2 Hz, OH-22), 2.47 (1H, ddq, J = 11.3, 1.8, 7.0 Hz, H-20), 2.35 (1H, dd, J = 11.0, 7.6 Hz, H-17), 2.18 (1H, m, H-4), 2.17 (3H, d, J = 1.2 Hz, H-27), 2.05 (3H, s, OAc-16), 1.94 (3H, m, H-26), 1.56 (1H, ddd, J = 7.5, 8.5, 14.0 Hz, H-11), 1.47 (1H, m, H-7), 1.39 (1H, dd, J = 13.6, 5.0 Hz, H-15), 1.25 (1H, d, J = 4.6 Hz, H-19a), 1.20 (3H, s, H-18), 1.02 (3H, d, J = 6.7 Hz, H-28), 0.95 (3H, d, J = 0.6 Hz, H-30), 0.65 (3H, d, J = 6.7 Hz, H-21), 0.56 (1H, d, J = 4.6 Hz, H-19b); ^{13}C NMR, see Table 3; HR-ESIMS m/z 519.3076 (calcd for $C_{31}H_{44}O_5Na$, 519.3081).

9,10-Di-epi-24a-nor-24,25-didehydro-26-deoxyneoboutomellerone (**23**): yellow gum; $[\alpha]^{20}_D$ −55 (c 0.07, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (3.77), 244 (3.98) nm; IR (film) ν_{max} 3409, 2936, 2874, 1734, 1669, 1612, 1447, 1374, 1235 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 6.50 (1H, d, J = 10.1 Hz, H-1), 6.15 (1H, sept, J = 1.2 Hz, H-24), 5.88 (1H, d, J = 10.1 Hz, H-2), 5.16 (1H, dt, J = 4.8, 7.7 Hz, H-16), 4.86 (1H, d, J = 1.7 Hz, H-22), 2.62 (1H, ddq, J = 11.0, 1.5, 6.9 Hz, H-20), 2.12 (3H, d, J = 1.1 Hz, H-27), 2.10 (6H, 2 s, OAc-16, OAc-22), 1.44−1.35 (2H, m, H-11, H-15), 1.30 (1H, d, J = 4.9 Hz, H-19a), 1.15 (3H, s, H-18), 1.05 (3H, d, J = 6.8 Hz, H-28), 0.93 (1H, m, H-19b), 0.91 (3H, d, J = 1.0 Hz, H-30), 0.86 (3H, d, J = 7.0 Hz, H-21); ^{13}C NMR, see Table 3; HR-ESIMS m/z 561.3187 (calcd for $C_{33}H_{46}O_6Na$, 561.3187).

23,24,24a,25,26,27-Hexa-nor-neoboutomelleron-22-al (**24**): yellow gum; $[\alpha]^{20}_D$ −74 (c 0.11, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210

dx.doi.org/10.1021/np200441h | J. Nat. Prod. 2012, 75, 34−47

(3.51), 266 (3.83) nm; IR (film) ν_{max} 3640, 2938, 2874, 1732, 1669, 1603, 1455, 1375, 1238 cm^{-1}; ^{1}H NMR (CD_3CN, 500 MHz) δ 9.58 (1H, d, J = 1.8 Hz, H-22), 6.95 (1H, d, J = 10.0 Hz, H-1), 5.90 (1H, d, J = 10.0 Hz, H-2), 5.24 (1H, dt, J = 5.5, 8.2 Hz, H-16), 2.86 (1H, ddq, J = 11.0, 2.0, 7.3 Hz, H-20), 2.39 (1H, dd, J = 11.0, 8.2 Hz, H-17), 2.16 (1H, m, H-4), 1.89 (3H, s, OAc-16), 1.47 (1H, m, H-7), 1.33 (1H, ddq, J = 13.4, 5.6, 1.1 Hz, H-15), 1.27 (1H, d, J = 4.3 Hz, H-19a), 1.17 (3H, s, H-18), 1.09 (3H, d, J = 7.3 Hz, H-21), 1.02 (3H, d, J = 6.7 Hz, H-28), 0.96 (3H, d, J = 0.9 Hz, H-30), 0.55 (1H, d, J = 4.3 Hz, H-19b); ^{13}C NMR, see Table 3; HR-ESIMS m/z 435.2503 (calcd for $C_{26}H_{36}O_4Na$, 435.2506).

*23,24,24a,25,26,27-Hexa-nor-neoboutomelleron-22-oic acid (**25**):* yellow gum; $[\alpha]^{20}_D$ −57 (c 0.06, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 208 (3.52), 266 (3.74) nm; IR (film) ν_{max} 3406, 2930, 2875, 1725, 1666, 1456, 1376, 1247 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.86 (1H, d, J = 10.0 Hz, H-1), 6.01 (1H, d, J = 10.0 Hz, H-2), 5.46 (1H, br q, J = 7.3 Hz, H-16), 2.73 (1H, m, H-20), 2.42 (1H, dd, J = 10.2, 9.1 Hz, H-17), 2.20 (1H, dq, J = 12.0, 6.9 Hz, H-4), 2.00 (3H, s, OAc-16), 1.75 (1H, m, H-12), 1.35 (1H, m, H-15), 1.23 (1H, d, J = 4.5 Hz, H-19a), 1.21 (3H, d, J = 7.0 Hz, H-21), 1.16 (3H, s, H-18), 1.10 (3H, d, J = 6.8 Hz, H-28), 0.96 (3H, s, H-30), 0.55 (1H, d, J = 4.5 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 451.2428 (calcd for $C_{26}H_{36}O_5Na$, 451.2455).

*16-Acetyl-3β,26-dihydroxy-24-methyl-25ξ-cycloart-24(**24a**)-en-23-on-29-oic acid (**26**):* yellow gum; $[\alpha]^{20}_D$ −7 (c 0.04, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.57) nm; IR (film) ν_{max} 3310, 2933, 2871, 1732, 1669, 1555, 1378, 1247, 1028 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.03 (1H, s, H-24a), 5.75 (1H, s, H-24a), 5.26 (1H, dt, J = 4.9, 7.8 Hz, H-16), 4.07 (1H,br d, J = 7.9 Hz, H-3), 3.55 (1H, dd, J = 10.6, 5.2 Hz, H-26a), 3.49 (1H, dd, J = 10.6, 7.0 Hz, H-26b), 2.95 (1H, sext, J = 6.5 Hz, H-25), 2.52 (3H, m, H-20, 2 H-22), 2.11 (1H, dd, J = 13.7, 8.2 Hz, H-15), 1.95 (3H, s, OAc-16), 1.89 (1H, dd, J = 9.9, 8.1 Hz, H-17), 1.80 (1H, m, H-2), 1.17 (3H, s, H-18), 1.13 (3H, br.s, H-28), 1.05 (3H, d, J = 7.0 Hz, H-27), 0.91 (3H, s, H-30), 0.88 (3H, d, J = 5.8 Hz, H-21), 0.62 (1H, d, J = 4.0 Hz, H-19a), 0.40 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 4; HRESIMS m/z 581.3443 (calcd for $C_{33}H_{50}O_7Na$, 581.3449).

*16-Acetyl-3β,22β-dihydroxy-24-methylcycloart-24(24a)-en-23-on-29-oic acid (**27**):* yellow gum; $[\alpha]^{20}_D$ 0 (c 0.03, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 216 (3.58) nm; IR (film) ν_{max} 3302, 2929, 2864, 1732, 1671, 1542, 1454, 1385 cm^{-1}; ^{1}H NMR (methanol-d_4, 500 MHz) δ 5.99 (1H, s, H-24a), 5.89 (1H, d, J = 1.2 Hz, H-24a), 5.27 (1H, dt, J = 4.0, 7.3 Hz, H-16), 4.71 (1H, d, J = 1.5 Hz, H-22), 4.03 (1H, dd, J = 11.3, 4.6 Hz, H-3), 2.87 (1H, dsept, J = 1, 6.7 Hz, H-25), 2.24 (1H, dd, J = 14.0, 7.9 Hz, H-15a), 2.06 (3H, s, OAc-16), 1.94 (1H, dd, J = 12.2, 3.4 Hz, H-5), 1.72 (1H, m, H-2), 1.19 (3H, s, H-18), 1.11 (3H, d, J = 7.0 Hz, H-27), 1.07 (3H, s, H-28), 1.03 (3H, d, J = 7.0 Hz, H-26), 0.99 (3H, s, H-30), 0.69 (3H, d, J = 6.1 Hz, H-21), 0.63 (1H, d, J = 4.1 Hz, H-19a), 0.42 (1H, d, J = 4.1 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 581.3455 (calcd for $C_{33}H_{50}O_7Na$, 581.3449).

*3β,16β,22β-Trihydroxy-24-methyl-(16,23:23,26)-diepoxycycloart-24(24a)-en-29-oic acid (**28**):* yellow gum; $[\alpha]^{20}_D$ −38 (c 0.05, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 208 (3.41), 270 (2.74) nm; IR (film) ν_{max} 3300, 2929, 2864, 1650, 1541, 1385 cm^{-1}; ^{1}H NMR (methanol-d_4, 500 MHz) δ 5.24 (1H, d, J = 3.1 Hz, H-24a), 4.92 (1H, d, J = 3.1 Hz, H-24a), 4.27 (1H, dt, J = 8.6, 7.1 Hz, H-16), 4.03 (1H, dd, J = 11.3, 4.6 Hz, H-3), 3.94 (1H, t, J = 7.8 Hz, H-27a), 3.30 (1H, t, J = 7.8 Hz, H-27b), 2.75 (1H, m, H-25), 2.07 (1H, ddd, J = 15.3, 10.5, 5.3 Hz, H-7a), 1.95 (1H, dd, J = 12.4, 4.1 Hz, H-5), 1.90 (1H, m, H-20), 1.84 (1H, dd, J = 12.8, 7.9 Hz, H-15a), 1.78 (1H, dd, J = 13.7, 2.7 Hz, H-22a), 1.74 (1H, m, H-2a), 1.49 (1H, t, J = 13.7 Hz, H-22b), 1.41 (1H, br dd, J = 12.5, 6.7 Hz, H-15b), 1.18 (3H, s, H-18), 1.08 (3H, s, H-28), 1.07 (3H, d, J = 6.8 Hz, H-26), 0.93 (3H, d, J = 7.0 Hz, H-21), 0.92 (3H, s, H-30), 0.68 (1H, d, J = 4.0 Hz, H-19a), 0.41 (1H, d, J = 4.0 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 521.3230 (calcd for $C_{31}H_{46}O_5Na$, 521.3237).

*3β-Hydroxy-24-methylcycloart-24(24a)-en-23-on-29-oic acid (**29**):* yellow gum; $[\alpha]^{20}_D$ −34 (c 0.06, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.78) nm; IR (film) ν_{max} 3380, 2930, 2869, 1678, 1552, 1406, 1374 cm^{-1}; ^{1}H NMR (methanol-d_4, 500 MHz) δ 6.05 (1H, s, H-24a), 5.75 (1H, d, J = 0.9 Hz, H-24a), 4.02 (1H, dd, J = 11.4, 4.4 Hz, H-3), 2.87 (1H, dsept, J = 1.0, 6.8 Hz, H-25), 2.72 (1H, dd, J = 15.6, 3.1 Hz, H-22a), 2.48 (1H, dd, J = 15.6, 10.1 Hz, H-22b), 1.08 (3H, s, H-28), 1.04 (3H, s, H-18), 1.02 (3H, d, J = 6.7 Hz, H-27), 1.01 (3H, d, J = 7.0 Hz, H-26), 0.95 (3H, s, H-30), 0.84 (3H, d, J = 6.4 Hz, H-21), 0.62 (1H, d, J = 4.2 Hz, H-19a), 0.41 (1H, d, J = 4.2 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 507.3441 (calcd for $C_{31}H_{48}O_4Na$, 507.3445).

*3β-Hydroxy-29-O-(α-L-rhamnopyranosyl)cycloart-24-en-23-on-29-oic acid (**30**):* yellow gum; $[\alpha]^{20}_D$ −22 (c 0.17, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 238 (4.27), 300 (3.09) nm; IR (film) ν_{max} 3291, 2932, 2867, 1723, 1677, 1607, 1445, 1378, 1311 cm^{-1}; ^{1}H NMR (methanol-d_4, 500 MHz) δ 6.16 (1H, sept, J = 1.2 Hz, H-24), 5.99 (1H, d, J = 1.8 Hz, H-1′), 4.03 (1H, dd, J = 11.3, 4.3 Hz, H-3), 3.86 (1H, dd, J = 3.4, 1.8 Hz, H-2′), 3.69 (1H, dd, J = 9.5, 3.4 Hz, H-3′), 3.61 (1H, dq, J = 9.5, 6.2 Hz, H-5′), 3.44 (1H, t, J = 9.5 Hz, H-4′), 2.52 (1H, dd, J = 15.0, 3.1 Hz, H-22a), 2.11 (3H, d, J = 0.9 Hz, H-27), 1.99 (1H, dd, J = 12.1, 3.8 Hz, H-5), 1.90 (3H, d, J = 0.9 Hz, H-26), 1.22 (3H, d, J = 6.1 Hz, H-6′), 1.13 (3H, s, H-28), 1.04 (3H, s, H-18), 0.94 (3H, s, H-30), 0.86 (3H, d, J = 6.4 Hz, H-21), 0.64 (1H, d, J = 4.1 Hz, H-19a), 0.44 (1H, d, J = 4.1 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 639.3856 (calcd for $C_{36}H_{56}O_8Na$, 639.3867).

*3β-Hydroxy-29-O-(α-L-rhamnopyranosyl)-24-methylcycloart-24-(24a)-en-23-on-29-oic acid (**31**):* yellow gum; $[\alpha]^{20}_D$ −16 (c 0.13, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 220 (3.95) nm; IR (film) ν_{max} 3291, 2935, 2865, 1725, 1659, 1462, 1376, 1248, 1147, 1105 cm^{-1}; ^{1}H NMR (methanol-d_4, 500 MHz) δ 6.05 (1H, s, H-24a), 5.99 (1H, d, J = 1.8 Hz, H-1′), 5.76 (1H, d, J = 1.2 Hz, H-24a), 4.03 (1H, dd, J = 4.3, 11 Hz, H-3), 3.86 (1H, dd, J = 3.5, 1.8 Hz, H-2′), 3.70 (1H, dd, J = 9.5, 3.5 Hz, H-3′), 3.61 (1H, dq, J = 9.5, 6.2 Hz, H-5′), 3.45 (1H, t, J = 9.5 Hz, H-4′), 2.87 (1H, sept, J = 6.7 Hz, H-25), 2.72 (1H, dd, J = 15.6, 2.7 Hz, H-22a), 2.48 (1H, dd, J = 15.6, 9.8 Hz, H-22b), 1.22 (3H, d, J = 6.2 Hz, H-6′), 1.12 (3H, s, H-28), 1.04 (3H, s, H-18), 1.02 (3H, d, J = 7.0 Hz, H-27), 1.01 (3H, d, J = 7.0 Hz, H-26), 0.95 (3H, s, H-30), 0.84 (3H, d, J = 6.4 Hz, H-21), 0.64 (1H, d, J = 4.1 Hz, H-19a), 0.44 (1H, d, J = 4.1 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 653.4014 (calcd for $C_{37}H_{58}O_8Na$, 653.4024).

*16,22-Diacetyl-2,26-dihydroxy-29-nor-24-methyl-19(9→1)-abeo-cycloart-9(11),24(24a)-dien-3,23-dione (**32**):* yellow gum; $[\alpha]^{20}_D$ +77 (c 0.13, $CHCl_3$); UV (MeOH) λ_{max} (log ε) 210 (3.97), 268 (3.17) nm; IR (film) ν_{max} 3394, 2926, 2854, 1734, 1694, 1566, 1376, 1229, 1021 cm^{-1}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 6.12 (1H, s, H-24a), 5.87 (1H, d, J = 0.6 Hz, H-24a), 5.56 (1H, d, J = 2.1 Hz, H-22), 5.20 (1H, d, J = 6.1 Hz, H-11), 5.09 (1H, dt, J = 4.4, 7.6 Hz, H-16), 4.16 (1H, d, J = 2.1 Hz, H-2), 3.60 (3H, m, OH-2, 2 H-26), 2.88 (1H, sext, J = 6.5 Hz, H-25), 2.56 (1H, m, H-20), 2.36 (1H, dd, J = 14.0, 7.9 Hz, H-15a), 2.27 (1H, dd, J = 11.1, 7.5 Hz, H-17), 2.15 (3H, s, OAc-22), 2.07 (3H, s, OAc-16), 1.88 (1H, dd, J = 9.9, 4.7 Hz, H-1), 1.37 (1H, dd, J = 14.2, 4.1 Hz, H-15b), 1.11 (3H, d, J = 7.3 Hz, H-27), 1.05 (3H, d, J = 6.4 Hz, H-28), 0.94 (1H, bt, J = 4.7 Hz, H-19a), 0.88 (3H, d, J = 7.0 Hz, H-21), 0.81 (3H, s, H-30), 0.79 (3H, s, H-18), 0.41 (1H, dd, J = 9.9, 4.7 Hz, H-19b); ^{13}C NMR, see Table 4; HR-ESIMS m/z 607.3245 (calcd for $C_{34}H_{48}O_8Na$, 607.3241).

Proteasome Assay. Experimental procedures for the assay are detailed in ref 3.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

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■ AUTHOR INFORMATION

Corresponding Author

*Tel: +33-5-34506517. E-mail: georges.massiot.externe@pierre-fabre.com.

■ ACKNOWLEDGMENTS

The authors are grateful to R. Bellé for historical plant collection, to P. Arimondo, C. Bailly, and N. Guilbaud for their

interest and attentive support during the research program, to A. Samson and P. Vergnes for driving the screening process, to C. Gau, C. Menendez, N. Molinier, and M. J. Serrano for their highly skilled contribution to the isolation of the compounds, and to N. Chansard for pharmacological evaluation.

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