Arch. Pharm. Res. **14**(2), 160-164 (1991)

The Chiroptical Properties and Absolute Configuration of 28-nor-β-amyrins

Won Sick Woo
Natural Products Research Institute, Seoul National University
(Received March 27, 1991)

Abstract □ The existence in nature of two isomers of 28-nortriterpenes is known. One is normal D/E *cis* form and the other is 17α-hydrogen D/E *trans* form. Since the latter cannot exist with ring D in the chair conformation, the chiroptical method is not applicable to determination of the absolute configuration. The stereochemical assignment would now be made by NMR data. Confirmation of this view could be provided by the synthesis of 3β, 21β-dihydroxy-16-keto-28-nor-17α, 18β-olean-12-ene as a model compound.

Keywords □ 28-nor-β-amyrins, melandrigenin, CD, ORD, ^{13}C-NMR

Chiroptical studies of carbonyl compounds can be used to resolve uncertainties in the detailed structure or relative configuration of a substance belonging to a series with known absolute configuration. In the last 30 years, ORD and CD have found many applications to rigid cyclic structures which either were already ketonic or could be converted into ketones by simple chemical transformation such as oxidation of secondary alcohols.

It is known as an example, introduction of a keto group in position 16 of the β-amyrin system imposes a rather strong negative Cotton effect upon the rotary dispersion curve[1]. The negative Cotton effect associated with the 16-keto group is so strong as to overcome the positive one of a 3-keto function. Therefore, the sign of the 16-oxo-Cotton effect was taken as a guide to chirality in this family of compounds[2,3]. Thus, observations of negative Cotton effects led to norechinocystenolone (**1**) and other norterpenoids of the group being allotted the natural β-amyrin configuration[4-6]. Norechinocysterolone is a product prepared from echinocystic acid (**2**) under conditions favoring the thermodynamically preferred isomer[7,8]. The assignment of β-orientation (D/E *cis* juncture) of the hydrogen atom at the invertible position is naturally to be expected since a *trans-cisoid-cis*-perhydrophenanthrene system in all-chair conformation is known to be more stable than the *trans-cisoid-trans* isomer where a chair-boat-chair arrangement is required for rings, C, D and E[9]. Although the premise that the cyclohexane ring is significantly more stable in the chair than the boat form has been well accepted, a few cases are known that the boat conformers are more stable than chair conformers[10,11].

X-ray analysis showed that melandrigenin (**3**) isolated from *Melandrium firmum* took the boat form on D ring[12]. This evidently can be the case when the boat form is more stable than chair form. In the conformation where ring D is a chair, there would be a β,β′-1,3-diaxial interaction between the methyl group at C-14 and the methylene group at C-18. If ring D becomes a "Twist" (13,16 LT form)* this interaction is relieved, a strong negative amplitude might be expected. In effect this accords with the experimental fact (see Figs. 1 and 2).

This observation let to an assumption that the chiroptical method can not be applicable as an aid to determine the configuration of a 28-nor-oleanane. Thus, the absolute configuration of norechinocystenolone should be reinvestigated.**

Except for those having substituents in close proximity, in D/E *cis* fused 12-oleanenes with all rings

* According to the convention proposed by Djerassi and Klyne[10] for describing twist conformations.

** Alves and Noller previously proposed the 17α-orientation (**4**)[8]. Later Belous made public a revision of the structure to 13(18)-ene (**5**)[13].

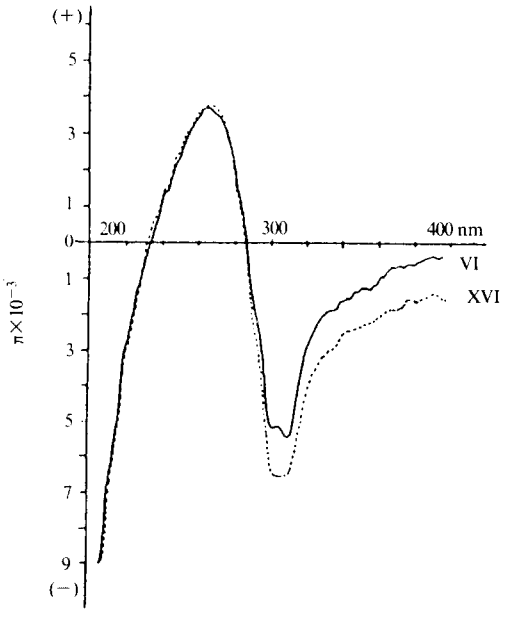


Fig. 1. ORD curve of model compound acetate (16) and melandrigenin acetate (6).

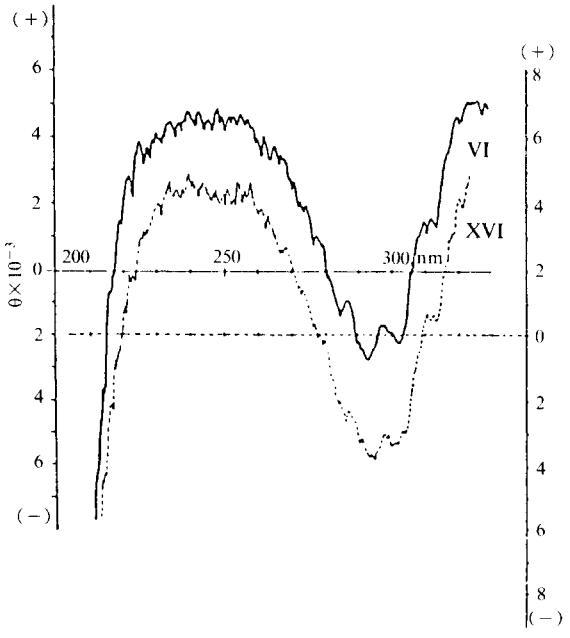
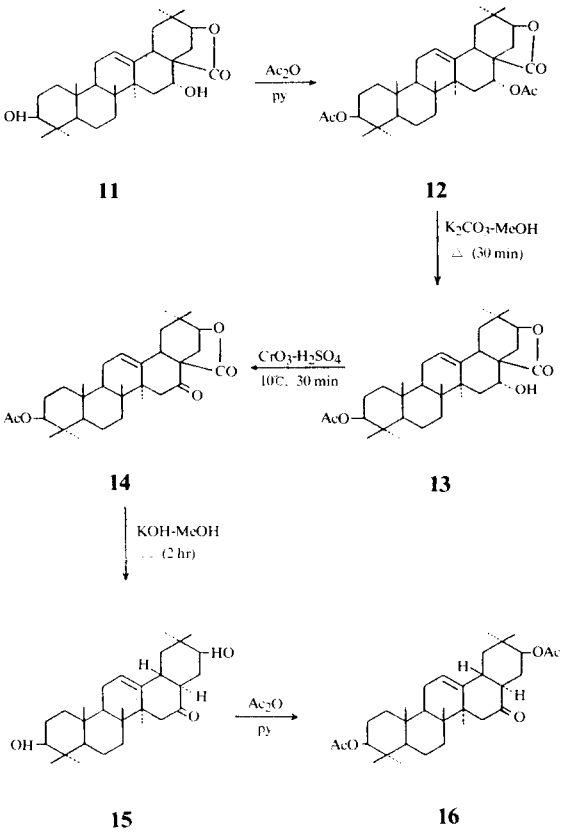


Fig. 2. CD curve of model compound acetate (16) and melandrigenin acetate (6).

in conventional chair conformation, the chemical shift of C-12 appears around δ 122 ppm[14].

However, the signal for melandrigenin acetate (**6**)



Scheme 1. Synthesis of model compound (15).

and its derivative (**7**) appeared in rather higher field region as compared with oleanene series common in nature (**8-10**), which can be used for a distinction between triterpenoids having normal D/E *cis* junction and those having 17α-hydrogen-D/E *trans* junction.

In order to verify this assumption, a model compound (**15**) was prepared (Scheme 1). Acacic acid lactone (**11**) obtained from *Albizzia* species was subject to acetylation followed by partial hydrolysis to yield the monoacetate (**13**) and treatment of this compound with chromium trioxide followed by alkali degradation gave a model compound (**15**), which was acetylated to compare with melandrigenin acetate (**6**).

The right hand portion of the model compound molecule is exactly the same as that of melandrigenin and the left hand portion as oleanolic acid. As expected carbon-13 chemical shift values for rings A and B of the model compound acetate

Table I. ^{13}C-NMR chemical shifts of melandrigenin acetate (6), gypsogenin methyl ester acetate (8), tetrahydromelandrigenin acetate (7), hederagenin methyl ester acetate (9), oleanolic acid methyl ester acetate (10) and model compound acetate (16). Methyl and acetate signals are not shown

Carbon No.	6	8	7	9	10	16
1	37.6	37.9	37.7	37.7	38.1	37.9
2	22.4	22.6	23.0	23.0	23.6	23.5
3	73.3	73.5	74.5	74.3	80.7	80.8
4	54.1	54.3	40.7	40.6	37.5	37.8
5	48.0	48.0	48.3	47.7	55.2	55.6
6	20.5	20.4	18.1	18.1	18.2	18.3
7	32.3	32.4	32.4	32.3	32.6	32.9
8	39.0	39.8	39.2	39.3	39.3	38.8
9	47.1	47.1	47.1	47.7	47.5	46.9
10	36.1	35.9	37.0	36.8	36.9	37.3
11	23.2	23.4	23.3	23.4	23.6	23.5
12	118.1	121.9	118.3	122.0	122.1	118.7
13	141.5	144.0	143.0	143.6	143.6	141.3
14	42.6	41.8	42.4	41.6	41.6	42.7
15	43.7	27.8	33.3	27.7	27.7	43.8
16	212.1	23.1	74.6	23.0	23.0	209.2
17	48.0	46.8	39.8	46.6	46.6	47.9
18	36.1	41.5	35.8	41.3	41.3	36.0
19	42.8	46.0	44.8	45.8	45.8	42.9
20	34.5	30.7	34.7	30.6	30.6	34.7
21	78.1	34.0	78.2	33.8	33.8	78.3
22	27.7	32.1	32.8	32.3	32.3	27.7
23	203.9	204.2	65.5	65.3	28.0	27.9
24	9.4	9.5	12.9	13.1	16.8	16.7
25	15.5	15.5	15.8	15.8	15.3	15.6
26	16.8	16.9	16.8	16.8	16.8	16.8
27	25.7	25.9	26.2	25.8	25.8	25.7
28	–	178.0	–	177.8	177.8	–
29	28.8	33.0	18.9	33.1	33.1	28.8
30	19.3	23.6	19.3	23.6	23.6	19.5

(**16**) are very similar to those of oleanolic acid methyl ester acetate (**10**) and the values for rings D and E are similar to those of melandrigenin acetate (**6**) and carbon signals for the double bond of these two compounds appear at high field and shift values are almost the same (Table I).

These evidences clearly indicate that rings C, D and E of the model compound (**15**) represent essentially a *trans-cisoid-trans*-perhydrophenanthrene system. As expected, CD and ORD curves (Figs. 1 and 2) of the model compound acetate (**16**) bear considerable resemblance to that of melandrigenin acetate (**6**).

From the carbon-13 chemical shift values reported, it is obvious that 28-nor-pentacyclic triterpenes isolated from *Camellia japonica*[5], *Celmisia petriei*[6] and *Sapindus mukorossi*[15] belong to a D/E *cis* fused one.

There is, however, no way we can elucidate the structure of maragenin I[14] without its carbon-13 NMR data.

EXPERIMENTAL

Acetylation of acacic acid lactone (11) and melandrigenin (3)

Acacic acid lactone (**11**), mp. 250-251℃, and melandrigenin (**3**), mp. 294℃, isolated from the acid hydrolysate of BuOH soluble fractions from MeOH extracts of the stem bark of *Albizzia julibrissin*[16] and

the whole plants of *Melandrium firmum*[12], respectively, were acetylated with Ac_2O/pyridine (1:1) in the usual way, separately, to give **12**, mp. 228℃; ^{1}H-NMR ($CDCl_3$) δ : 1.98 (3H, s), 2.04 (3H, s), 4.21 (1H, d, J=6 Hz, H-21), 4.49 (1H, dd, J=7 & 9 Hz, H-3), 5.03 (1H, dd, J=5 & 12 Hz, H-16), 5.43 (1H, m, H-12) and **6**, mp. 267-269℃; ^{1}H-NMR ($CDCl_3$) δ 1.93 (3H, s), 2.02 (3H, s), 4.57 (1H, dd, J=4 & 12 Hz, H-21), 4.98 (1H, dd, J=7 & 9 Hz, H-3), 5.45 (1H, m, H-12); MS (70 eV) *m/z* (rel. int.): 480 (M^+ − HOAc, 59.4), 276 (D/E ring, 29.5).

Partial hydrolysis of 12

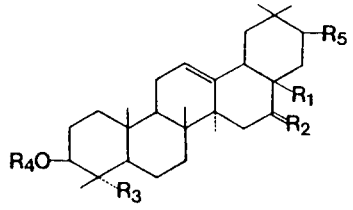
To a solution of diacetate (**12**) (200 mg) in MeOH (50 m*l*), a mixture of dioxane-H_2O (8 m*l*:6 m*l*) containing K_2CO_3 (250 mg) was added and refluxed for 30 min. The reaction mixture was evaporated to remove MeOH, diluted with H_2O, extracted with ether and subjected to column chromatography with C_6H_6-EtOAc (7:3) to give unchanged diacetate (**12**) (33 mg), acacic acid lactone (**11**) (19 mg) and monoacetate (**13**) (130 mg), mp. 296-300℃; ^{1}H-NMR ($CDCl_3$) δ 3.92 (1H, dd, J=5 & 12 Hz, H-16), 4.17 (1H, d, J=6 Hz, H-21), 4.43 (1H, dd, J=7 & 9 Hz, H-3).

Oxidation of 13

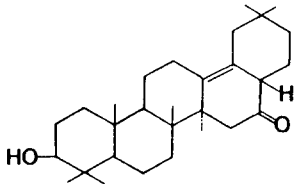
To a solution of monoacetate (**13**) (100 mg) in acetone (5 m*l*) cooled with an ice bath to 0-10℃, the oxidant (CrO_3 2.7 g+conc. H_2SO_4 2.3 m*l*+H_2O 4 m*l*) was added dropwise with stirring until completion of the reaction being shown by the persistence of the brownish orange color and allowed to stand for 30 min. The reaction mixture was poured into ice water and extracted with ether. After evaporation of solvent, the residue was purified by crystallization from MeOH to give needles of **16**, mp. 248-249℃; ^{1}H-NMR ($CDCl_3$) δ 4.18 (1H, d, J=6 Hz, H-21), 4.40 (1H, dd, J=7 & 9 Hz, H-3).

Decarboxylation of 16

A solution of 50 mg of the ketone derivative (**16**) and 0.5 g of KOH in 10 m*l* of MeOH was refluxed for 2 hr. It was poured into water, most of the alcohol removed by boiling, the precipitate filtered, which was crystallized from MeOH as needles of **15**, mp. 233-236℃; MS (70 eV) *m/z* (rel. Int.): 442 (M^+, 6.6), 234 (D/E ring, 20.0), 216 (234-H_2O, 16.4), 190 (A/B ring, 28.8).



	R_1	R_2	R_3	R_4	R_5
1	β-H	O	CH_3	H	H
2	COOH	H, α-OH	CH_3	H	H
3	α-H	O	CHO	H	OH
4	α-H	O	CH_3	H	H
6	α-H	O	CHO	Ac	OAc
7	α-H	H, α-OAc	CH_2OAc	Ac	OAc
8	COOMe	H_2	CHO	Ac	H
9	CH_2OAc	H_2	CH_2OAc	Ac	H
10	COOMe	H_2	CH_3	Ac	H



5

Acetylation of 15

Diacetate (**16**) was prepared from the model compound (**15**) by acetylation with pyridine-Ac_2O; needles from MeOH, mp. 224-230℃; ^{1}H-NMR ($CDCl_3$) δ 2.04 (6H, s, acetate × 2), 4.49 (t, J=8.1 Hz, H-3), 4.59 (1H, dd, J=4.5 & 12 Hz, H-21), 5.47 (1H, m, H-12); MS (70 eV) *m/z* (rel. int.): 466 (M^+ - HOAc, 13.5), 276 (D/E ring, 5.3), 216 (276- HOAc, 25.0), 190 (A/B ring- HOAc, 27.6).

LITERATURE CITED

1. Djerassi, C., Osiecki, J. and Closson, W.: Optical rotatory dispersion studies. XXV. Effect of carbonyl groups in pentacyclic triterpenes. *J. Am. Chem. Soc.* **81**, 4587 (1959).
2. Varshney, I. P., Shamsuddin, K. M. and Beyler, R. E.: The structure, stereochemistry and conformation of acacic acid. *Tetrahedron Lett.* 1187 (1965).
3. Varshney, I. P. and Shamsuddin, K. M.: Absolute structure of acacic acid. *Bull. Chem. Soc.*

Japan **43**, 3830 (1970).
4. Hylands, P. J. and Salama, A. M.: Maragenins I, II and III, new pentacyclic triterpenes from *Marah macrocarpus*. *Tetrahedron* **35**, 417 (1979).
5. Itokawa, H., Nakajma, H., Ikuta, A. and Iitaka, Y.: Two triterpenes from the flowers of *Camellia japonica*. *Phytochemistry* **20**, 2539 (1981).
6. Rowan, D. D. and Newman, R. H.: Noroleanane saponins from *Celmisia petriei*. *Phytochemistry* **23**, 639 (1984).
7. White, W. R. and Noller, C. R.: Saponins and sapogenins. IX. Oxidation of echinocystic acid and derivatives. *J. Am. Chem. Soc.* **61**, 983 (1939).
8. Alves, F. A. and Noller, C. R.: Saponins and sapogenins. XXX. The isomeric norechinocystenediones and some related compounds. *J. Am. Chem. Soc.* **74**, 4043 (1952).
9. Johnson, W. S.: The relative stability of stereoisomeric forms of fused ring systems. *Experientia* **7**, 315 (1951).
10. Djerassi, C. and Klyne, W.: Optical rotatory dispersion and the twist conformation of cyclohexanones, *Proc. Nat. Acad. Sci.* **48**, 1093 (1962).
11. Morita, M., Kojima, Y., Kato, N., Miwa, K., Tanaka, I., Yamane, T. and Ashida, T.: Absolute stereochemistries and conformations of clerodin and caryoptin. Why conflicting results in absolute stereochemistry based on CD and ORD spectra. *Tetrahedron Lett.* **24**, 5631 (1983).
12. Chang, H. S., Han, Y. B., Woo, W. S., Kang, S. S., Lotter, H. and Wagner, H.: Sapogenins from *Melandrium firmum*. *Planta Med.* **55**, 544 (1989).
13. Belous, V. N.: Structure of norechinocystenolone. *Vestn. Leningrad. Univ.* **22**, 162 (1967) (CA. **67**, 116987q).
14. Kang, S. S.: ^{13}C-NMR spectroscopy of amyrins. *Kor. J. Pharmacogn.* **18**, 151 (1987).
15. Linde, H: Über Inhaltsstoffe der Perikarpien von *Sapindus mukorossi* Gaertn. *Arch. Pharm.* **312**, 416 (1979).
16. You, Y. H., Woo, W. S., Choi, J. S. and Kang, S. S.: Isolation of a new sapogenin from *Albizzia julibrissin*. *Arch. Pharm. Res.* **5**, 33 (1982).