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Studies on the Constituents of *Marsdenia formosana* Masamune. IV. Photochemical Reaction of Marsformoxide A

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Marsformoxide A (IX) is a triterpene substance possessing an epoxide ring in the molecule, which was previously isolated by us from *Marsdenia formosana* Masamune (Asclepiadaceae). Irradiation of marsformoxide A with a high pressure mercury lamp in acidic medium furnished D-friedours-11,14-dien-3β-yl acetate (XI), and urs-9(11),12-dien-3β-yl acetate (XII), together with recovery of the starting material IX.

Keywords——*Marsdenia formosana* Masamune; Asclepiadaceae; marsformoxide A; D-friedours-11,14-dien-3β-yl acetate; urs-9(11),12-dien-3β-yl acetate; urs-12-en-11β-ol-3β-yl acetate; photochemical reaction

In 1965, Agata and Corey *et al.*[2] reported some interesting biogenetic-type photochemical reactions of pentacyclic triterpenes. It involved a novel skeletal rearrangement of thermo-

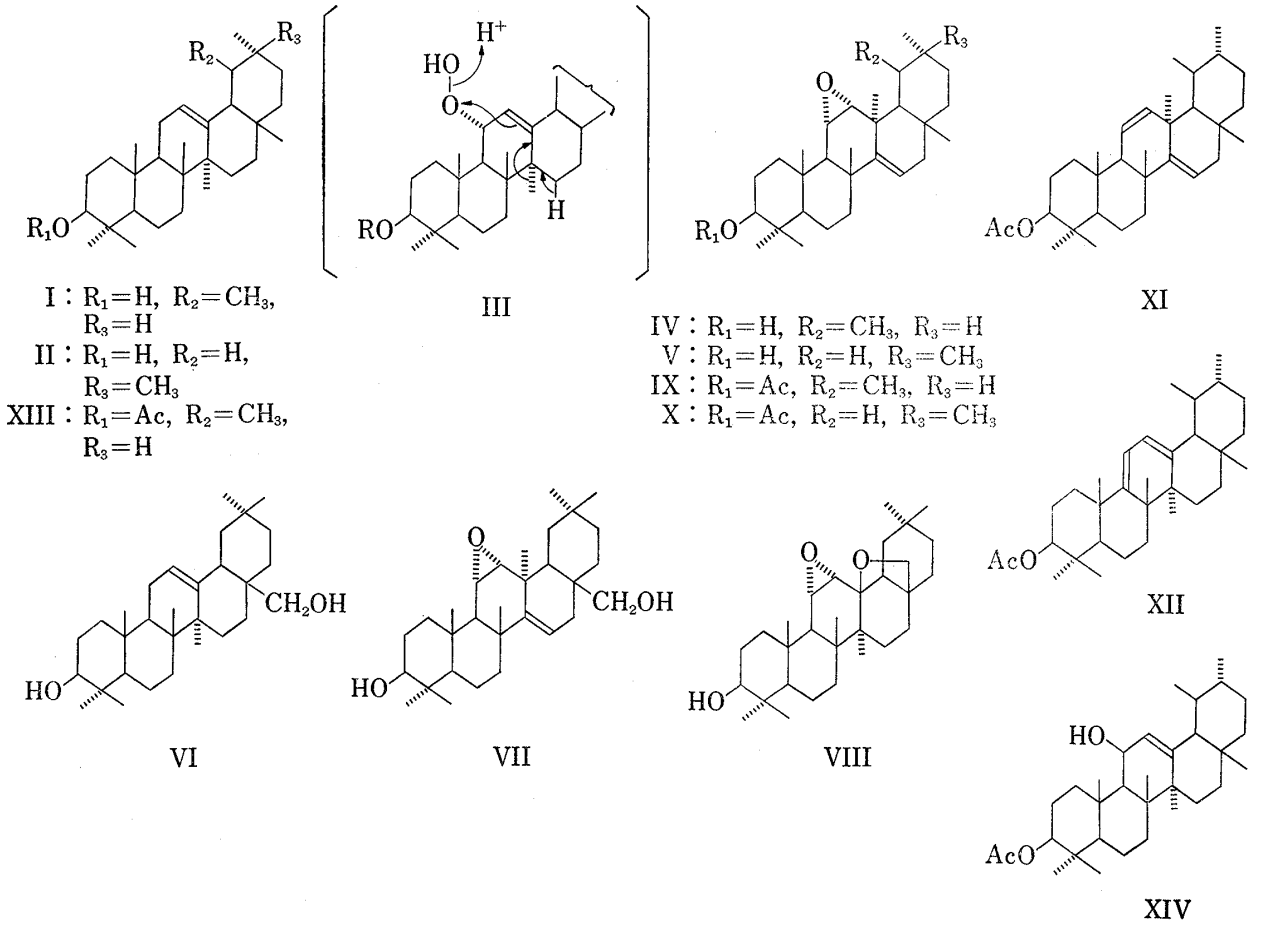


Chart 1

1) Location: *Yagoto-urayama, Tenpaku-cho, Tenpaku-ku, Nagoya.*

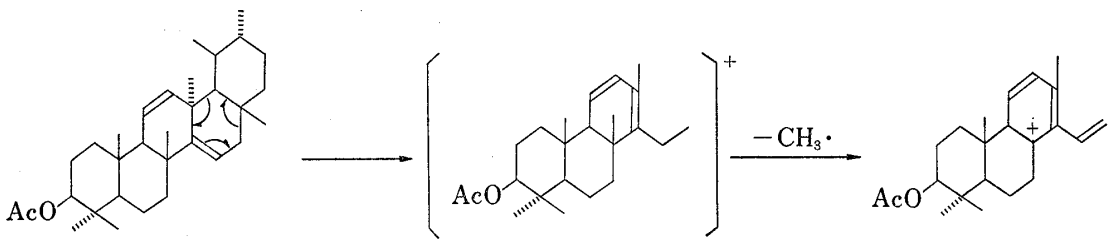
2) I. Agata, E. Corey, A.G. Hortmann, J. Klein, S. Proskow, and J.J. Ursprung, *J. Org. Chem.*, 30, 1698 (1965).

dynamically more favored α-amyrin (I) and β-amyrin (II) into less stable D-friedours-11α, 12α-epoxy-14-en-3β-ol (IV) and D-friedoolean-11α,12α-epoxy-14-en-3β-ol (V) respectively, by the way of photochemical oxidation in the acidic medium. They proposed an intermediate (III) in connection with the independent conversion of α-amyrin and β-amyrin to the same products IV and V by the alternate chemical oxidation. On the other hand, Kitagawa *et al.*[3] also presented a paper which dealt with the photooxidation of erythrodiol (VI) leading to the formation of D-friedoolean-11α,12α-epoxy-14-en-3β,28β-diol (VII) and olean-11α,12α-epoxy-13β,28-oxide (VIII) in the application of Agata's method.[2]

As mentioned above, although the biogenetic-type photooxidation of α-amyrin, β-amyrin and erythrodiol have been studied extensively, any example of the further photochemical study of these epoxide products has never been reported. Recently, we have reported the isolation and characterization of the two biogenetically interested compounds marsformoxide A (IX: D-friedours-11α,12α-epoxy-14-en-3β-yl acetate) and marsformoxide B (X: D-friedoolean-11α,12α-epoxy-14-en-3β-yl acetate) from the *Marsdenia formosana* MASAMUNE. In that paper, we assumed that the α-amyrin and β-amyrin would be the *in vivo* precursors of IV and V respectively.[4] In connection with our studies on the constituents of *Marsdenia formosana* MASAMUNE (Asclepiadaceae), in the present paper we intend to report the biogenetic-type photochemical reaction of marsformoxide A.

Irradiation of marsformoxide A(IX) with a high pressure mercury lamp in 0.003 N HCl–dioxane for 7 hr at room temperature furnished three pure products tentatively designated as MF-I (9%), MF-II (6%), MF-III (trace) along with recovery of the starting material (43%). Independently, treatment of marsformoxide A with 0.003 N HCl–dioxane for 7 hr at room temperature in the dark gave no reaction product except for the starting material on thin-layer chromatography (TLC) examination.

MF-I, colorless needles, mp 178—180°, has a molecular formula $C_{32}H_{50}O_2$. Its infrared (IR) spectrum indicated acetoxyl absorption at 1728 and 1250 cm^{-1}, together with the absorption of double bond at 1638 cm^{-1}, and ultraviolet (UV) spectrum gave no evidence for the existence of conjugated double bonds. The nuclear magnetic resonance (NMR) spectrum of this compound discloses the presence of eight methyl groups (δ 0.86—1.12) and a C-3α-proton at δ 4.52 (1H, d.d, J=9, 7 Hz), together with a one proton signal at δ 5.53 (1H, d.d, J=4, 8 Hz) due to an olefinic proton at C-15. In addition, two olefinic proton signals at δ 5.64 (1H, d.d, J=11, 3.5 Hz) and 5.99 (1H, d.d, J=11, 2.5 Hz), could be assignable to the AM part of an AMX system in the NMR spectrum.[5] Furthermore, the mass (MS) spectrum of this compound exhibits the prominent fragment ion peaks at *m/e* 342 and *m/e* 327 assignable



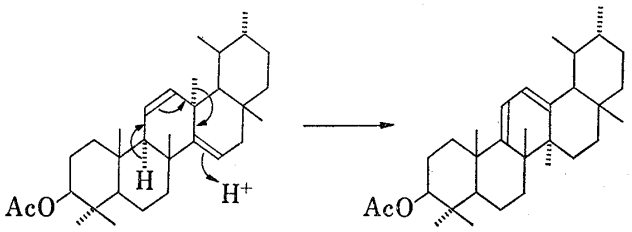
XI (a) *m/e* 342 (b) *m/e* 327

Chart 2

3) I. Kitagawa, K. Kitazawa, and I. Yosioka, *Tetrahedron Lett.*, **1968**, 509.
4) K. Ito and J. Lai, *Chem. Pharm. Bull.* (Tokyo), **26**, 1908 (1978).
5) *a*) T. Mezzetti, G. Oralesi, and V. Bellavita, *Planta. Med.*, **20**, 244 (1971); *b*) I. Kitagawa, K. Kitazawa, and I. Yosioka, *Tetrahedron*, **28**, 907 (1972); *c*) N. Aimi, H. Fujimoto, and S. Shibata, *Chem. Pharm. Bull.* (Tokyo), **16**, 641 (1968).

to the ions (a) and (b) which would be formed by the retro Diels–Alder cleavage[6] (Chart 2). These physical data suggest that MF-I would possess the D-friedoursane skeleton and be represented by D-friedours-11,14-dien-3β-yl acetate (XI).

A number of compounds possessing the D-friedoursane and D-friedooleane skeletons have been reported to be relatively unstable, and easily rearranged to the ursane and oleane skeletons respectively, in the presence of acid catalysis.[7] Therefore, if our suggestion is correct, XI would be isomerized to urs-9(11),-12-dien-3β-yl acetate (XII). In fact, treatment of XI with 0.05 N HCl–dioxane (Chart 3) or direct irradiation of XI with a high pressure mercury lamp in 0.003 N HCl–dioxane afforded XII, which was identical with an authentic sample, yielded by the alternate chemical oxidation from α-amyrin acetate (XIII).[8] From the above physical and chemical data, the structure of MF-I is proved to be represented by D-friedours-11,14-dien-3β-yl acetate (XI).



XI XII

Chart 3

MF-II, colorless needles, mp 165—167°, has a molecular formula $C_{32}H_{50}O_2$. From the investigation of its various spectra, we confirmed that MF-II would be the same compound as urs-9(11),12-dien-3β-yl acetate (XII). In fact, this compound was completely identical with an authentic sample in comparison of their mixed mp, and IR, UV and MS spectra.

MF-III, amorphous powder; its IR spectrum indicated absorption band due to the acetoxyl group at 1730 and 1255 cm^{-1}, together with hydroxyl group at 3610 cm^{-1}. MS spectrum showed a molecular ion peak at *m/e* 482. However, because of considerable unstability of this compound to reagents and shortage of the material, its structure has not yet been clarified.

Although D-friedours-11α,12α-epoxy-14-en-3β-ol (IV) formed in the course of photooxidation of α-amyrin (I), has been reported as a final product, our present experiments clarified that its acetate (IX) will undergo further reaction to afford D-friedours-11,14-dien-3β-yl acetate (XI), which was readily rearranged into urs-9(11),12-dien-3β-yl acetate (XII).

Furthermore, in connection with our extensive studies on the chemistry of the triterpenoids, we considered that urs-9(11),12-diene derivatives would be easily obtained by the photolysis of urs-12-en-11-ol derivatives. In fact, on irradiation of urs-12-en-11β-ol-3β-yl acetate (XIV), derived from α-amyrin acetate (XIII)[4] with a high pressure mercury lamp in 0.002 N HCl–dioxane for 1.5 hr, it was readily converted into urs-9(11),12-dien-3β-yl acetate (XII).[9]

Although this substance XII which frequently occurs in the plants can be readily obtained by irradiation of urs-12-en-11β-ol-3β-yl acetate (XIV), XII can be also obtained by the photochemical transformation of marsformoxide A (IX) (*via* an intermediate XI).

As the results of our present experiments, it seems reasonable to assume that marsformoxide A which was isolated from *Marsdenia formosana* MASAMUNE might be an important *in vivo* intermediate of the biogenetic route from α-amyrin (I) to XI and XII.

6) H. Budzikiewicz, J.M. Wilson, and C. Djerassi, *J. Am. Chem. Soc.*, **85**, 3688 (1963).
7) *a*) J.M. Beaton, F.S. Spring, R. Stevenson, and J.L. Stewart. *J. Chem. Soc.*, **1955**, 2131; *b*) C.J.W. Brooks, *ibid.*, **1955**, 1675; *c*) W. Laird, F.S. Spring, and R. Stevenson, *ibid.*, **1961**, 2638.
8) K. Yagishima and M. Nishimura, *Agr. Biol. Chem.*, **25**, 844 (1961).
9) Treatment of urs-12-en-11β-ol-3β-yl acetate (XIV) with 0.002 N HCl–dioxane for 1.5 hr at room temperature in the dark gave no reaction product except for the starting material.

Experimental[10]

Marsformoxide A[4] **(IX: D-Friedours-11α,12α-epoxy-14-en-3β-yl Acetate)**——Colorless needles, mp 212—214°. $[\alpha]_D^{25}$ −25° (c=0.5, $CHCl_3$). IR ν_{max}^{KBr} cm^{-1}: 1728, 1250 (OAc), 1638 (C=C), 895 (epoxy). NMR ($CDCl_3$) δ: 5.51 (1H, d.d, J=4, 8 Hz, C_{15}–H), 4.50 (1H, d.d, J=9, 6.5 Hz, C_3–H), 3.14 (1H, d.d, J=5, 5.7 Hz, C_{11}-β-H), 2.98 (1H, d, J=5 Hz, C_{12}-β-H), 2.04 (3H, s, OAc), 1.11 (3H, d, J=6 Hz, *sec.* CH_3), 1.09 (3H, s, *tert.* CH_3), 1.07 (3H, s, *tert.* CH_3), 0.99 (3H, d, J=6 Hz, *sec.* CH_3), 0.89 (9H, s, 3 *tert.* CH_3), 0.86 (3H, s, *tert.* CH_3). MS *m/e*: 482 (M^+), 358, 343, 108 (base peak). *Anal.* Calcd. for $C_{32}H_{50}O_3$: C, 79.62; H, 10.44. Found: C, 79.55; H, 10.55.

Irradiation of Marsformoxide A (IX)——A solution of IX (500 mg) in 0.003 N HCl–dioxane (450 ml) was put into Pyrex tube and irradiated with a 100 W high pressure mercury lamp at room temperature for 7 hr. The reaction mixture was poured into water and extracted with ether. The ether extracts were washed thoroughly with 2% NaOH and with water, and then dried over anhydrous Na_2SO_4. Evaporation of the solvent gave an amorphous solid, which was carefully chromatographed on silica gel (200 g) packed in benzene containing 30% cyclohexane. Elution with benzene–cyclohexane (4: 1) gave MF-II (30 mg, 6%) and MF-I (45 mg, 9%). Further elution of the column with benzene–$CHCl_3$ (5: 1) afforded marsformoxide A (215 mg, 43%) and MF-III (5 mg).

MF-I (XI: D-Friedours-11,14-dien-3β-yl Acetate)——It was recrystallized from $CHCl_3$–MeOH to give colorless needles. mp 178—180°. IR ν_{max}^{KBr} cm^{-1}: 1728, 1250 (OAc), 1638 (C=C). NMR ($CDCl_3$) δ: 0.86—1.12 (8 CH_3), 2.04 (3H, s, OAc), 4.52 (1H, d.d, J=9, 7 Hz, C_3–H), 5.53 (1H, d.d, J=4, 8 Hz, C_{15}–H), 5.64 (1H, d.d, J=11, 3.5 Hz, C_{11}–H), 5.99 (1H, d.d, J=11, 2.5 Hz, C_{12}–H). MS *m/e*: 466 (M^+), 451, 406, 391, 342, 327, 282, 267, 255, 188, 173, 159, 131, 123. *Anal.* Calcd. for $C_{32}H_{50}O_2$: C, 82.34; H, 10.80. Found: C, 82.51; H, 10.95.

MF-II (XII: Urs-9(11),12-dien-3β-yl Acetate)——It was recrystallized from acetone to afford colorless needles, mp 165—167°. $[\alpha]_D^{25}$ +336° (c=0.5, $CHCl_3$). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 1725, 1255 (OAc), 1640 (C=C). UV λ_{max}^{MeOH} mμ (ε): 282 (9660). MS *m/e*: 466 (M^+). *Anal.* Calcd. for $C_{32}H_{50}O_2$: C, 82.34; H, 10.80. Found: C, 82.45; H, 10.83. This compound was completely identical with an authentic sample in comparison of their mixed mp, and IR, UV and MS spectra.

MF-III——Amorphous, IR ν_{max}^{KBr} cm^{-1}: 3610 (OH), 1730, 1255 (OAc). MS *m/e* 482 (M^+).

Isomerization of D-Friedours-11,14-dien-3β-yl Acetate (XI)——a) A solution of XI (10 mg) in 0.05 N HCl–dioxane (15 ml) was heated at 50° for 6 hr. The solvent was then evaporated under reduced pressure and the residue was crystallized from acetone to give colorless needles (5 mg). mp 165—167°. $[\alpha]_D^{25}$ +335° (c=0.5, $CHCl_3$).

b) A solution of XI (15 mg) in 0.003 N HCl–dioxane (200 ml) was put into pyrex tube and irradiated with a 100 W high pressure mercury lamp at room temperature for 50 min. The reaction mixture was poured into water and extracted with ether. The ether extracts were washed thoroughly with 2% NaOH and with water, and then dried over anhydrous Na_2SO_4. Evaporation of the solvent gave amorphous solid, which was carefully chromatographed on silica gel (15 g) packed in benzene containing 30% cyclohexane. Elution with benzene–cyclohexane (4: 1) gave colorless solid, which was recrystallized from acetone to afford colorless needles (3.5 mg). mp 165—167°, $[\alpha]_D^{25}$ +330° (c=0.3, $CHCl_3$).

The compounds obtained from the above two methods were completely identical with urs-9(11),12-dien-3β-yl acetate (XII) which was given by the alternate reaction from α-amyrin acetate (XIII) in comparison of their mixed melting point, and IR, UV and MS spectra.

Formation of Urs-9(11),12-dien-3β-yl Acetate (XII) from α-Amyrin Acetate (XIII)——To a solution of α-amyrin acetate (500 mg) in CCl_4 (50 ml) was added 60 mg of NBS (97%), and the resulting solution was refluxed for 1.5 hr. Filtration and removal of the solvent afforded a yellowish residue, which was dissolved in 200 ml of ether. The ethereal layer was washed with water, and dried with Na_2SO_4. Evaporation of the solvent gave a yellowish material (480 mg), which was chromatographed on alumina column (150 g) packed in petroleum ether solution and then eluted with the same solvent. Removal of the solvent afforded the solid, which was recrystallized repeatedly from acetone to give XII as a colorless needles. mp 166—167°.

Urs-12-en-11β-ol-3β-yl Acetate (XIV)——A solution of urs-12-en-3β,11β-diol (1.0 g) which was derived from α-amyrin acetate in pyridine (70 ml) was maintained at −20°, and then cold acetic anhydride (100 mg) was added at −20°. After standing for 18 hr at −20°, the excess acetic anhydride was decomposed by the addition of 100 ml of 95% ethanol. The solution was concentrated to 60 ml *in vacuo* and then poured into water and extracted with CH_2Cl_2. The CH_2Cl_2 was washed with water, dried over anhydrous Na_2SO_4, and evaporated to dryness. The residue was carefully chromatographed on silica gel (50 g) packed in *n*-

10) All melting points were taken on a micro hot-stage and are uncorrected. IR spectra were recorded on a JASCO A-3 spectrometer. NMR spectra were determined on a JEOL PS-100 spectrometer operating at 100 MHz with tetramethylsilane (TMS) as an internal standard. MS were taken on a Hitachi M-52 mass spectrometer with a heated direct inlet system. Optical rotations were measured on a JASCO DIP-SL polarimeter.

hexane–benzene (1: 1). Elution with *n*-hexane–benzene (1: 2) gave 650 mg of crystalline XIV which was recrystallized from $CHCl_3$–MeOH to afford colorless needles. mp 267—269°. $[\alpha]_D^{25}$ +101° (c=0.5, $CHCl_3$). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3620 (OH), 1725, 1255 (OAc).

Irradiation of Urs-12-en-11β-ol-3β-yl Acetate (XIV)——A solution of XIV (500 mg) in 0.002 N HCl–dioxane (450 ml) was put into pyrex tube and irradiated with a 100 W high pressure mercury lamp at room temperature for 1.5 hr. The reaction mixture was poured into water and extracted with ether. The ethereal extracts were washed thoroughly with 2% NaOH and with water, and then dried over anhydrous Na_2SO_4. Evaporation of the solvent gave an amorphous solid, which was carefully chromatographed on silica gel (200 mg) packed in benzene containing 30% cyclohexane. Elution with benzene–cyclohexane (4: 1) gave XII (65 mg) and urs-12-en-11-on-3β-yl acetate (35 mg). Both of them were identical with the authentic samples in comparison of their mixed melting point, and IR and MS spectra.

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