Tetrahedron Vol. 42, No. 5, pp. 1275 to 1283, 1986
Printed in Great Britain.

0040-4020/86 $3.00+.00

THE REACTION OF DAMMARANE TRITERPENES WITH m-CHLOROPERBENZOIC ACID[1)]

Motoo Tori, Reiko Matsuda, and Yoshinori Asakawa*

Faculty of Pharmaceutical Sciences, Tokushima Bunri University,
Yamashiro cho, Tokushima 770, Japan

(*Received in Japan* 28 *October* 1985)

Abstract: Dammaran-20(S)-ol was treated with m-chloroperbenzoic acid (mCPBA) to give the corresponding 5α-ol, 7β-ol, 2α-ol, 12β-ol, 25-ol, and 5α,25-diol. 20(S)-Hydroxydammaran-3β-yl acetate was treated with mCPBA to afford the 5α-ol, 7β-ol, 12β-ol, and 25-ol. Their structures were determined by spectroscopic methods.

Dammaranes are a common type of triterpene found in the plant kingdom, and are characteristic constituents of Ginseng[2)] and Gynostemma pentaphyllum Makino.[3)] They are often hydroxylated at various positions besides C-3 and C-20. Tanaka and his co-workers reported that photolysis of 20(S)-hydroxydammaran-3α-yl p-nitrophenylacetate in Bu^tOH according to Breslow's procedure[4)] gave dammarane-3α,12α,20(S)-triol in 13% yield after hydrolysis.[5)] This was the first successful application of the remote oxidation to the triterpenes. No further reports of the application of this method to the triterpenes have appeared presumably due to the rather tedious steps required for derivation of the desired starting material.

We are interested in in vitro reactions which simulate the in vivo oxidation reactions observed in mammalian metabolism. We have been studying the structures of the metabolites obtained from rabbits upon feeding with various terpenoids.[6)] The latter are generally excreted in hydroxylated form for detoxification.[6)] We chose m-chloroperbenzoic acid (mCPBA) as the reagent for the hydroxylation of unactivated carbon atoms. Both concentrated peracids and radicals produced by photolysis of peroxides have previously found use but these reactions are, in principle, dangerous.[7)] Deno et al. demonstrated that trifluoroperacetic acid can be used to hydroxylate aliphatic compounds, e.g. 2-octanol to 2,7-octanediol. It is also useful for the cleavage of steroidal side chains.[8)] mCPBA was first employed by Müller and Schneider for the introduction of a hydroxyl group at a bridgehead position of bicyclic compounds,[9)] whilst Takaishi et al. later subjected polycyclic compounds[10)] to the same procedure. We have recently applied this reaction to natural products, dammarane,[1)] lupane,[11)] and friedelane triterpenoids,[11)] cedrol,[12)] and acyclic compounds[13)] and have found that hydroxylation occurs at not only tertiary but also secondary positions. We describe the details of the reactions of dammarane triterpenes with mCPBA and the assignments of the ^{13}C

NMR spectra of these products by use of INEPT techniques. Revision of the former assignments[14,15] is also discussed.

Dammaran-20(S)-ol (1) was treated with mCPBA (1.2 equiv.) in refluxing chloroform for 6 h. The reaction mixture was worked up and purified by column chromatography over silica gel to give six products (compounds I-VI) as well as the starting material (1) recovered in 77 % yield.

The ^{1}H NMR spectrum of compound I (2; Y. 59%[16]), $C_{30}H_{54}O_2$ [from high resolution mass, m/z 428.4044 (M-H_2O), and ^{13}C NMR spectra], showed no peak between 3 and 5 ppm and in the ^{13}C NMR spectrum thirty signals were observed, two of which were due to oxygen-bearing carbons [δ_C 77.3 (s, C-5) and 75.4 (s, C-20)]. The mass spectrum showed a molecular ion peak at m/z 446 and a characteristic peak at m/z 362 (Fig. 1).[17] These facts suggested that one hydroxyl group had been introduced at the C-5 position. ^{13}C NMR signals having the similar chemical shifts to those of 1 are due to rings C and D and the side chain (Table 1). If the C-5 position is substituted by a hydroxyl group, the chemical shifts of the carbons of rings A and B are well explained by suitable substituent effects. The stereochemistry of the C-5 position was determined by comparing the spectral data of 2 with those of 13 (*vide infra*). Compound I (2) was thus determined to be dammarane-5α,20(S)-diol.

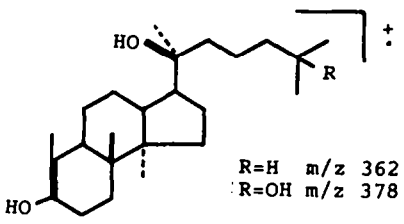
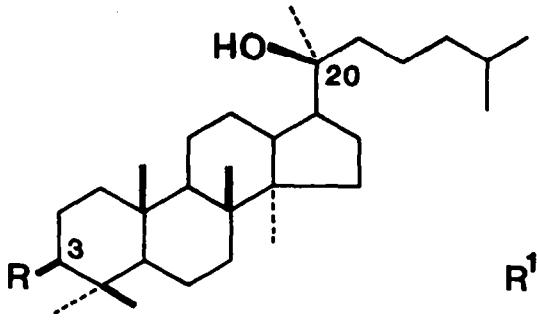


Fig. 1

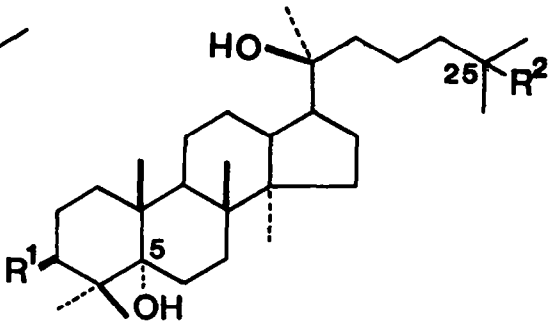
In the ^{1}H NMR spectrum of compound II (3; Y. 2.1%[16]), $C_{30}H_{54}O_2$ [HRMS m/z 428.4014 (M-H_2O) and ^{13}C NMR] a double doublet centered at δ 3.80 (1H, J=11 and 5 Hz) was observed and the ^{13}C NMR spectrum indicated the presence of thirty carbons, two of which bore oxygen functions [δ_C 75.3 (d, C-3) and 75.4 (s, C-20)]. From the coupling pattern of the methine proton, the hydroxyl group could be assigned to the 1β, 7β, or 15α position. Of the five quaternary carbon atoms [δ_C 75.4 (C-20), 49.7 (C-14), 46.2 (C-8), 37.3 (C-10), and 33.4 (C-4)] of 3, four had similar chemical shifts to those of dammaran-20(S)-ol (1) [δ_C 75.3 (C-20), 50.4 (C-14), 40.6 (C-8), 37.5 (C-10), and 33.4 (C-4)], *viz* the carbon signal at δ_C 40.6 assigned to C-8 has undergone a downfield shift of 5.6 ppm, indicating that the hydroxyl group was most probably in the 7β position. The secondary alcohol (3) was oxidized to the corresponding ketone (8), $C_{30}H_{52}O_2$ (m/z 444.3953), by Jones reagent. Methylene protons were observed at δ 2.50 (1H, t, J=15 Hz) and 2.20 (1H, dd, J=15 and 2.5 Hz) in the ^{1}H NMR spectrum of 8, suggesting the partial structure -CH-CH_2-CO-. From these results the secondary alcohol was determined to be dammarane-7β,20(S)-diol (3).

The third product, compound III (4; Y. 1.3%[16]), $C_{30}H_{54}O_2$ [HRMS m/z 410.3920 (M-H_2O) and ^{13}C NMR], was also a secondary alcohol from its ^{1}H NMR [δ 3.60 (1H, td, J=11 and 5 Hz)] and ^{13}C NMR spectra [δ_C 74.4 (s, C-20) and 71.2 (d, C-12)]. The coupling pattern of the methine proton attached to the carbon bearing the newly introduced hydroxyl group indicated that the position of substitution was 6α, 11α, 12β, or 16β. Comparison of the ^{13}C NMR chemical shifts (Table 1) with those of known dammarane triterpenes implied a 12β-ol structure. Jones oxidation of 4 gave the ketone 9, $C_{30}H_{52}O_2$ [m/z 426.3847 (M-



1 R = H

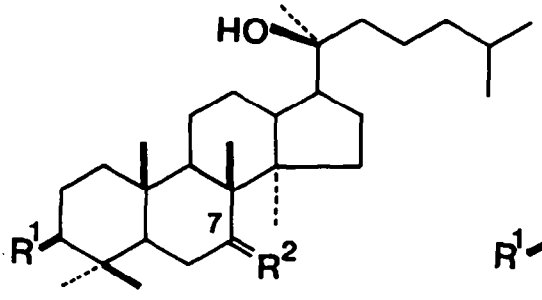
12 R = OAc



2 $R^1 = R^2 = H$

7 $R^1 = H$, $R^2 = OH$

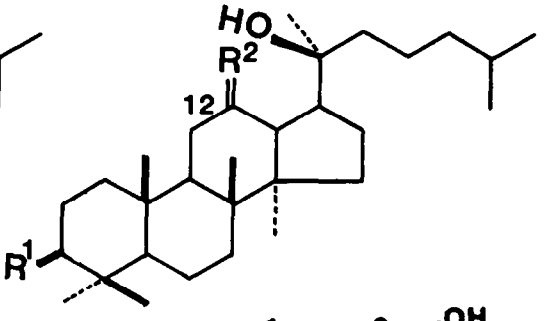
13 $R^1 = OAc$, $R^2 = H$



3 $R^1 = H$, $R^2 = \overset{OH}{H}$

8 $R^1 = H$, $R^2 = O$

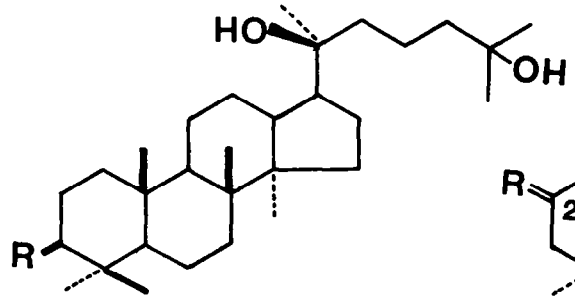
14 $R^1 = OAc$, $R^2 = \overset{OH}{H}$



4 $R^1 = H$, $R^2 = \overset{OH}{H}$

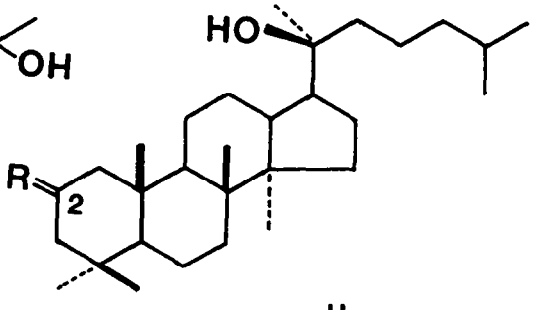
9 $R^1 = H$, $R^2 = O$

15 $R^1 = OAc$, $R^2 = \overset{OH}{H}$



5 R = H

16 R = OAc



6 R = $\overset{H}{OH}$

10 R = $\overset{H}{OAc}$

11 R = O

H_2O)], whose ^1H NMR spectrum showed a doublet at δ 2.85 (1H, J=10 Hz, H-13β), a doublet of doublets at δ 2.30 (1H, J=14 and 5 Hz, H-11α),and a triplet at δ 2.19 (1H, J=14 Hz, H-11β), suggesting the partial structure -CH-CH_2-CO-CH-CH-. From these facts, compound III (4) was concluded to be dammarane-12 ,20(S)-diol (4).

The fourth product, compound IV (5; Y. 28%[16]), $C_{30}H_{54}O_2$ [HRMS m/z 410.3927 (M-H_2O X 2) and ^{13}C NMR], was easily assigned as dammarane-20(S),25-diol (5) from its mass spectrum [m/z 410 (M-H_2O X 2), 191, 145, 127 (base), and 59] and ^1H NMR spectrum (eight singlet methyls were observed at δ 1.23, 1.23, 1.14, 0.96, 0.88, 0.85, 0.84, and 0.81).

A triplet of triplets observed at δ 3.89 (1H, J=11 and 4.5 Hz) in the ^1H NMR spectrum of the penultimate product, compound V (6; Y. 4.2%[16]), $C_{30}H_{54}O_2$ [HRMS m/z 428.4209 (M-H_2O) and ^{13}C NMR], indicated the presence of an equatorial secondary hydroxyl group with a methylene group on each side. The same pattern was observed in the ^1H NMR spectrum [δ 5.06 (1H, tt, J=10 and 6 Hz)] of its acetate (10). The position of substitution is limited only to 2α (equatorial). The corresponding ketone (11), $C_{30}H_{52}O_2$ [m/z 426.3853 (M-H_2O)], obtained by Jones oxidation showed four protons α to the carbonyl group [δ 2.39 (1H, dd, J=13 and 2 Hz), 2.28 (1H, d, J=13 Hz), 2.15 (1H, dd, J=13 and 2 Hz), and 1.98 (1H, d, J=13 Hz)]. These data were fully consistent with structure 6, dammarane-2α,20(S)-diol.

The most polar product, compound VI (7; Y. 1.3%[16]), $C_{30}H_{54}O_2$ [HRMS m/z 444.3999 (M-H_2O) and ^{13}C NMR], showed eight singlet methyls in its ^1H NMR spectrum and fragment peaks at m/z 444 (M-H_2O), 378 (Scheme 4), 127, and 59 in its mass spectrum, suggesting the presence of a 25-hydroxyl group. In the ^{13}C NMR spectrum thirty signals were detected, three of which corresponded to oxygenated carbons [δ_C 77.4 (s, C-5), 75.4 (s, C-20), and 71.1 (s, C-25)]. Comparison of the spectral data with those of 2 and 5 led directly to structure 7.

We next carried out a similar reaction of 20(S)-hydroxydammaran-3β-yl acetate (12) with mCPBA in chloroform. Work up and purification by column chromatography over silica gel and high performance liquid chromatography (HPLC) (μ-PORASIL, see Experimental) afforded four products, compound VII-X (13-16) as well as the recovered 12 (91% yield).

The mass fragmentation pattern (Scheme 4)[17] of the compound VII (13; Y. 16%[16]), $C_{32}H_{56}O_4$ [HRMS m/z 486.4084 (M-H_2O) and ^{13}C NMR], suggested a 5α-ol structure. As the methine proton attached to the carbon bearing the acetoxyl group appeared at δ 5.12 (dd, J=11 and 6 Hz), 0.64 ppm downfield of that of 12, the hydroxyl group was determined to be 5α axial. The ^{13}C NMR signals due to rings B, C, and D and the side chain of 13 were quite similar to those of 2 (*vide supra*). Thus the compound VII (13) was determined to be 5α,20(S)-dihydroxydammaran-3β-yl acetate.

The spectral data [m/z 486, 433, 419, 108, and 95; δ 4.48 (1H, dd, J=11 and 6 Hz), 3.79 (1H, dd, J=11 and 4 Hz), and 2.05 (3H, s); δ_C 171.0 (s, CO), 80.6 (d, C-3), 75.4 (s, C-20), and 70.9 (s, C-12)] of compound VIII (14; Y. 1.6%[16]), $C_{32}H_{56}O_4$ [HRMS m/z 486.4087 (M-H_2O) and ^{13}C NMR], are very well explained in terms of the 3β-acetate of compound II (3) and hence compound VIII (14) should be formulated as 7β,20(S)-dihydroxydammaran-3β-yl acetate.

In the ^{1}H NMR spectrum of compound IX (15; Y. 1.6%[16]), $C_{32}H_{56}O_4$ [HRMS m/z 468.3956 (M-H_2O X 2) and ^{13}C NMR], a triplet of doublets (J=10 and 5 Hz) was observed at δ 3.60, which was exactly the same position and pattern as that of 4. The data were completely identical with those of authentic 12β,20(S)-dihydroxydammaran-3β-yl acetate (15).[14] As this is an acetate of a typical sapogenin of Ginseng, this constitutes a one step synthesis of such types from a 12-deoxy compound.

The last compound X (16; Y. 40.3%[16]),[18] $C_{32}H_{56}O_4$ [HRMS m/z 486.4112 (M-H_2O) and ^{13}C NMR], exhibited eight singlet methyls (δ 1.23, 1.23, 1.14, 0.96, 0.88, 0.87, 0.85, and 0.85) in its ^{1}H NMR spectrum and characteristic fragmentations [m/z 468 (M-H_2O X 2), 191, 189, 145, 127, 109, and 59] in its mass spectrum. Hence it was determined to be 20(S),25-dihydroxydammaran-3β-yl acetate (16).

Thus the hydroxylation reaction reported here takes place by an attack on unactivated carbon atoms of the triterpene framework. The mechanism is presumably a radical process as recently reported by Fossy et. al.[19] The order of reactivity of unactivated carbon atoms is tertiary > secondary > primary. Hydroxylation of the 25 position is the major reaction because it is a tertiary position and is not subject to steric hindrance. The 5α-ol is another major product of both reactions, even though the 5-position would appear to be subject to somewhat more steric hindrance. The other products are all equatorial alcohols (2α, 7β, and 12β-ol), showing that equatorial alcohols are formed much more readily than the corresponding axial alcohols. Although the reaction site cannot fully be predicted at this stage, the oxidation reaction with mCPBA is quite useful for introduction of a functional group at an unactivated carbon atom.

^{13}C NMR Tanaka and his co-workers assigned the ^{13}C NMR spectra of a number of dammarane triterpenes and these results were later used to assign the ^{13}C NMR spectrum of 1.[14,15] However, measurement of ^{13}C NMR spectrum of 1 using the INEPT pulse sequence has revealed that the previously determined multiplicities of two resonances are incorrect. Thus the signal at δ_C 24.8 is due to a methylene carbon whilst that at δ_C 25.6 results from a methyl group (Fig. 2). It is probable that the assignments of Tanaka are also in need of revision. In order to ensure the correctness of other assignments, the ^{13}C NMR spectra of several derivatives in hand were determined and the multiplicities confirmed by use of INEPT pulse sequence. In the case of triterpenes most ^{13}C NMR signals appear in the highly congested region between 10 and 50 ppm and it is hence difficult to determine the multiplicities from the SFORD spectrum. The INEPT or DEPT pulse sequences are the best choice for such compounds.

Table 1. ^{13}C NMR data of dammarane derivatives

	1	2	3	4	5	6	7	13	14	15	16
1	40.6	32.9	40.5	40.5	40.7	49.9	32.8	32.1	36.9	37.9	38.8
2	18.7	17.9	18.7	18.7*	18.7	65.3	17.9	23.6	21.4	23.7	23.7
3	42.2	36.2	41.9	42.1	42.2	51.2	36.1	77.2	80.6	80.8	81.0
4	33.4	38.5	33.2	33.3	33.4	39.7	38.4	39.3	38.5	38.6	38.0
5	57.0	77.3	54.3	57.0	57.0	56.4	77.4	78.9	53.3	55.9	56.0
6	18.7	23.0	28.2	18.6*	18.7	18.3	23.0	23.4	27.7	18.2	18.2
7	35.3	28.7	75.3	34.8	35.3	35.0	28.6	28.4	75.1	34.8	35.2
8	40.6	40.9	46.2	39.9	40.7	41.0	41.0	40.7	46.0	39.8	40.4
9	50.8	42.0	48.8	50.2	50.8	50.7	41.9	42.0	48.7	50.0	50.6
10	37.5	39.6	37.3	37.4	37.5	42.1	39.6	43.1	37.7	37.0	37.1
11	21.5	21.5	21.2	31.2	21.5	21.5	21.5	21.7	21.3	31.2	21.6
12	24.8	24.8	25.3	71.2	24.9	24.8	24.8	24.8	23.7	70.9	24.9
13	42.2	42.3	43.0	47.7	42.4	42.1	42.3	42.4	43.0	47.8	42.3
14	50.4	50.9	49.7	51.7	50.4	50.4	50.8	50.8	49.7	51.6	50.3
15	31.2	31.3	34.7	30.9	31.2	31.1	31.3	31.3	34.7	31.0	31.2
16	27.6	27.8	27.4	26.6	27.7	27.5	27.8	27.8	27.3	26.5	27.5
17	49.8	49.8	50.4	53.5	50.0	49.7	49.9	49.8	50.1	53.5	49.9
18	16.2	16.7	9.8	16.1	16.2	15.6	16.7	16.6	9.7	16.2	16.3
19	15.6	19.2	16.1	15.6	15.6	17.3	19.2	19.3	16.2*	15.7	15.5
20	75.3	75.4	75.4	74.4	75.4	75.4	75.5	75.4	75.4	74.5	75.4
21	25.6	25.6	25.7	27.1	25.6	25.6	25.5	25.7	25.8	27.2	25.6
22	41.1	41.1	40.9	35.1	41.1	40.5	40.9	41.2	40.9	35.0	41.1
23	21.5	21.5	21.5	21.3	18.5	21.6	18.5	21.5	21.5	21.3	18.5
24	39.8	39.8	39.7	39.8	44.6	39.1	44.5	39.8	39.7	39.8	44.5
25	28.0	28.1	28.0	28.2	71.1	28.0	71.1	28.1	28.0	28.2	71.0
26	22.7	22.6	22.6*	22.6+	29.3*	22.6*	29.3*	22.7*	22.6+	22.6*	29.3*
27	22.7	22.6	22.7*	22.7+	29.5*	22.7*	29.4*	22.8*	22.7+	22.7*	29.5*
28	33.4	27.9	33.3	33.4	33.4	33.5	27.9	22.6	28.0	28.0	28.0
29	21.5	23.9	21.5	21.5	21.6	22.4	23.9	18.5	16.5	16.5	16.5
30	16.5	15.7	16.4	16.9	16.6	16.4	15.7	15.7	16.3*	16.8	16.5
CO								170.9	171.0	171.0	171.0
CH_3								21.3	25.3	21.3	21.3

*,+ Assignments may be reversed.

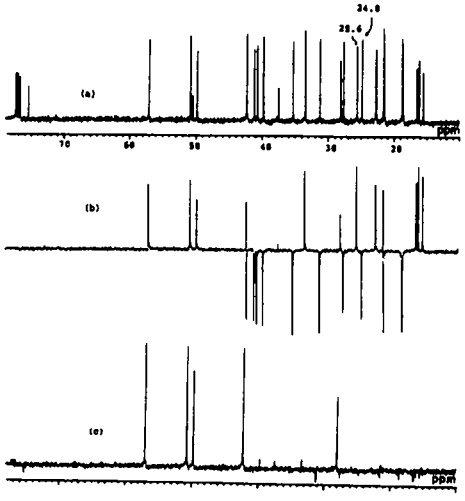


Fig. 2. (a) Complete ^1H decoupled spectrum (100 MHz) of dammaran-20(S)-ol (1). About 20 mg/0.5 ml $CDCl_3$ solution, 100 times accumulation, pulse width 4 μs (ca. 40°), pulse delay 2 sec, data points 16 K, frequency 20000 Hz. (b) INEPT spectrum with PI1(=1/4J)=2 ms, PI2(=3/4J)=6 ms. (c) INEPT spectrum with PI1(=1/4J)=2 ms, PI2(=2/4J)=4 ms.

EXPERIMENTAL

All melting points were measured on a Yanagimoto microscope hot plate and are uncorrected. IR spectra were measured with a Shimadzu IR-27G spectrophotometer. ^1H and ^{13}C NMR spectra were obtained with a JEOL JMN-GX400 spectrometer in $CDCl_3$ unless otherwise stated. Mass spectra were taken on a Shimadzu LKB-9000B or JEOL JMS D-300. Thin layer chromatography (TLC) was performed on Silica gel 60 F_{254} (Merck). Silica gel 60 (70-230 mesh, Merck) was used for column chromatography. mCPBA was purchased from Nakarai Chemical Co. Ltd. and used without further purification.

Oxidation Reaction of Dammaran-20(S)-ol (1) with mCPBA To a solution of dammaran-20(S)-ol (1) (1.0 g) in $CHCl_3$ (30 ml) was added mCPBA (450 mg, 1.2 eq). The mixture was stirred at 65-75°C for 6h and washed successively with 5% Na_2SO_3 aq, 5% $NaHCO_3$ aq, and brine. The dried solution was evaporated in vacuo to give the residue (1.23 g). The product mixture was chromatographed on silica gel, using hexane-Et_2O and benzene-EtOAc solvent systems for elution. The products were further purified by prep. HPLC (μ-PORASIL, 15 % EtOAc-hexane) to afford dammaran-20(S)-ol (1) (140 mg), dammarane-5α,20(S)-diol (2) (140 mg), dammarane-7β,20(S)-diol (3) (5 mg), dammarane-12β,20(S)-diol (4) (3 mg), dammarane-20(S),25-diol (5) (66 mg), dammarane-2α,20(S)-diol (6) (10 mg), and dammarane-5α,20(S),25-triol (7) (3 mg).

Dammarane-5α,20(S)-diol (2): mp 116°C; IR λ_{max} : 3620, 3480 (OH) cm^{-1}; ^1H NMR: δ 0.87 (3H, s), 0.88 (3H, d, J=7 Hz), 0.89 (3H, d, J=7 Hz), 0.96 (3H, s), 0.97 (3H, s), 1.00 (3H, s), 1.01 (3H, s), 1.12 (3H, s, 21-CH_3); ^1H NMR (C_6D_6): δ 0.88 (3H, s), 0.92 (3H, d, J=7 Hz), 0.93 (3H, d, J=7 Hz), 0.94 (3H, s), 0.96 (3H, s), 0.98 (3H, s), 1.05 (3H, s), 1.10 (3H, s, 21-CH_3); MS m/z: 446 (M^+), 428 ($M-H_2O$)$^+$, 410 ($M-H_2O$ x 2)$^+$, 395, 362, 344, 205, 191, 95 (base); HRMS: Found m/z 428.4044 ($M-H_2O$)$^+$, Calcd for $C_{30}H_{52}O$ 428.4018.

Dammarane-7β,20(S)-diol (3): Oil; IR λ_{max} : 3600, 3450 (OH) cm^{-1}; ^1H NMR: δ 0.82 (3H, s), 0.84 (3H, s), 0.87 (3H, s), 0.88 (6H, d, J=7 Hz), 0.95 (3H, s), 0.97 (3H, s), 1.13 (3H, s, 21-CH_3), 3.80 (1H, dd, J=11, 5 Hz, 7-Ha); MS m/z: 428 ($M-H_2O$)$^+$, 410, 361, 343, 318, 300, 285, 247, 123, 109, 95, 69 (base); HRMS: Found m/z 428.4014 ($M-H_2O$)$^+$, Calcd for $C_{30}H_{52}O$ 428.4019.

Dammarane-12β,20(S)-diol (4): Oil; IR λ_{max} : 3370 (OH) cm^{-1}; ^1H NMR: δ 0.81 (3H, s), 0.85 (3H, s), 0.87 (3H, s), 0.88 (6H, d, J=6.5 Hz, 26-CH_3, 27-CH_3), 0.89 (3H, s), 0.99 (3H, s), 1.18 (3H, s, 21-CH_3), 3.60 (1H, td, J=11, 5 Hz, 12-Hα); MS m/z: 446 (M^+), 428, 410, 191, 109, 69 (base); HRMS: Found m/z 410.3920 ($M-H_2O$ x 2)$^+$, Calcd for $C_{30}H_{50}$ 410.3912.

Dammarane-20(S),25-diol (5): mp 153°C (MeOH); IR λ_{max} : 3600, 3400 (OH) cm^{-1}; ^1H NMR: δ 0.81, 0.84, 0.85, 0.88, 0.96 (each 3H, s, 18-CH_3, 19-CH_3, 28-CH_3, 29-CH_3, 30-CH_3), 1.14 (3H, s, 21-CH_3), 1.23 (6H, s, 26-CH_3, 27-CH_3); MS m/z: 410 ($M-H_2O$ x 2)$^+$, 345, 191, 127 (base), 109; HRMS: Found m/z 410.3927 ($M-H_2O$)$^+$, Calcd for $C_{30}H_{50}$ 410.3912.

Dammarane-2α,20(S)-diol (6): mp 104°C (MeOH); IR λ_{max} : 3600, 3450 (OH) cm^{-1}; ^1H NMR: δ 0.85 (3H, s), 0.88 (6H, d, J=6.5 Hz, 26-CH_3, 27-CH_3), 0.89 (6H, s), 0.92 (3H, s), 0.96 (3H, s), 1.13 (3H, s, 21-CH_3), 3.89 (1H, tt, J=11, 4.5 Hz, 2-Hβ); MS m/z: 428 ($M-H_2O$)$^+$, 413, 395, 247, 207, 191, 95, 69 (base); HRMS: Found m/z 428.4029 ($M-H_2O$)$^+$, Calcd for $C_{30}H_{52}O$ 428.4019.

Dammarane-5α,20(S),25-triol (7): mp 165°C (MeOH); IR λ_{max}: 3600, 3450 (OH) cm^{-1}; ^1H NMR: δ 0.91, 0.96, 0.97, 1.00, 1.01 (each 3H, s, 18-CH_3, 19-CH_3, 28-CH_3, 29-CH_3, 30-CH_3), 1.14 (3H, s, 21-CH_3), 1.23 (6H, s, 26-CH_3, 27-CH_3); MS m/z: 444 ($M-H_2O$)$^+$, 426, 411, 408, 378, 145, 127 (base), 109, 69; HRMS: Found m/z 444.3999 ($M-H_2O$)$^+$, Calcd for $C_{30}H_{52}O_2$ 444.3967.

Oxidation of Diol 3 The diol 3 (2.2 mg) in acetone (0.6 ml) was oxidized with Jones reagent (2 drops) at rt for 30 min to give 7-oxodammaran-20(S)-ol (8) (1.9 mg): Oil; IR λ_{max} : 3400 (OH), 1700 (C=O) cm^{-1}; ^1H NMR: δ 0.83 (3H, s), 0.84 (3H, s), 0.88 (6H, d, J=6.5 Hz, 26-CH_3, 27-CH_3), 0.89 (3H, s), 1.00 (3H, s), 1.13 (3H, s), 1.16 (3H, s), 2.20 (1H, dd, J=15, 2.5 Hz, 6-Ha), 2.50 (1H, t, J=15 Hz, 6-Hb); ^1H NMR (C_6D_6): δ 0.67 (3H, s), 0.71 (3H, s), 0.77 (3H, s), 0.92 (3H, d, J=6.5 Hz), 0.93 (3H, d, J=6.5 Hz), 1.04 (3H, s), 1.06 (3H, s), 1.08 (3H, s); MS m/z: 426 ($M-H_2O$)$^+$, 410, 359, 316, 248, 220, 123, 95, 69 (base); HRMS: Found m/z 444.3953 (M^+), Calcd for $C_{30}H_{52}O_2$ 444.3965.

Oxidation of Diol 4 To a stirred solution of the diol 4 (1.0 mg) in acetone (0.4 ml) was added Jones reagent (2 drops) at rt. The mixture was stirred for 30 min and acetone was removed. Work up as usual afforded 12-oxodammaran-20(S)-ol (9) (0.7 mg): Oil: IR λ_{max} : 3400 (OH), 1700 (C=O) cm^{-1}; ^1H NMR: δ 0.81 (3H, s), 0.84 (3H, s), 0.87 (3H, s), 0.87 (3H, d, J=7 Hz), 0.88 (3H, d, J=7 Hz), 0.93 (3H, s), 1.10 (3H, s), 1.17 (3H, s, 21-CH_3), 2.19 (1H, t, J=14 Hz, 11-Hb), 2.30 (1H, dd, J=14, 5 Hz, 11-Ha), 2.40 (1H, td, J=10, 7 Hz, 17-H),

2.85 (1H, d, J=10 Hz, 13-H); MS m/z: 426 $(M-H_2O)^+$, 411, 359, 316, 234, 191, 124 (base); HRMS: Found m/z 426.3847 $(M-H_2O)^+$, Calcd for $C_{30}H_{50}O$ 426.3859.

Acetylation of Diol 6 The diol 6 (8.0 mg) was treated with acetic anhydride (1 ml) in pyridine (1 ml) and worked up as usual to give 20(S)-hydroxydammaran-2α-yl acetate (10) (8 mg): mp 144°C (EtOAc); IR λ_{max} : 3450 (OH), 1720 (C=O) cm^{-1}; ^1H NMR: δ 2.02 (3H, s, $OCOCH_3$), 5.06 (1H, tt, J=10, 6 Hz, 2-Hβ); MS m/z: 470 $(M-H_2O)^+$, 410, 403, 300, 285, 191, 189, 129 (base); HRMS: Found m/z 470.4110 $(M-H_2O)^+$, Calcd for $C_{32}H_{54}O_2$ 470.4122.

Oxidation of Diol 6 The diol 6 (9.0 mg) in acetone (1.6 ml) was oxidized with Jones reagent (4 drops) at rt for 1h to give 2-oxodammaran-20(S)-ol (11) (8.4 mg): Oil; IR λ_{max} : 3450 (OH), 1700 (C=O) cm^{-1}; ^1H NMR: δ 0.86 (3H, s), 0.88 (6H, d, J=7 Hz), 0.89 (3H, s), 0.92 (3H, s), 0.97 (3H, s), 1.06 (3H, s), 1.13 (3H, s), 1.98 (1H, d, J=13 Hz), 2.15 (1H, dd, J=13, 2 Hz); ^1H NMR (C_6D_6): δ 0.78 (6H, s), 0.79 (3H, s), 0.84 (3H, s), 0.86 (3H, s), 0.93 (3H, d, J=7 Hz), 0.94 (3H, d, J=7 Hz), 1.09 (3H, s), 1.57 (1H, d, J=13 Hz), 1.96 (1H, d, J=13 Hz), 2.13 (1H, dd, J=13, 2 Hz), 2.37 (1H, dd, J=13, 2 Hz); MS m/z: 426 $(M-H_2O)^+$, 316, 217, 205, 95, 69 (base); HRMS: Found m/z 426.3853 $(M-H_2O)^+$, Calcd for $C_{30}H_{50}O$ 426.3860.

Oxidation Reaction of 20(S)-Hydroxydammaran-3β-yl acetate (12) with mCPBA mCPBA (843 mg, 1.2 eq) was added to a solution of 20(S)-hydroxydammaran-3β-yl acetate (12) (1.44 g) in $CHCl_3$ (45 ml) in one portion and the mixture was refluxed for 6h. The reaction mixture was worked up as usual. The residue (1.65 g) was purified by silica gel column chromatography (benzene-EtOAc) to give 12 (1.32 g), a mixture of 13, 14, 15, and 16 (50 mg). The mixture of 13, 14, and 15 was further purified by prep. HPLC (μ-PORASIL, 5% EtOAc-hexane and 20% EtOAc-hexane) to give 5α,20(S)-dihydroxydammaran-3β-yl acetate (13) (20 mg), 7β,20(S)-dihydroxydammaran-3β-yl acetate (14) (2 mg), and 12β,20(S)-dihydroxydammaran-3β-yl acetate (15) (2 mg).

5α,20(S)-Dammaran-3β-yl acetate (13): mp 194°C; IR λ_{max} : 3600, 3450 (OH), 1725, 1255 (OCO) cm^{-1}; ^1H NMR: δ 0.88 (6H, d, J=7 Hz, 26-CH_3, 27-CH_3), 0.91 (3H, s), 0.94 (3H, s), 0.98 (3H, s), 1.00 (3H, s), 1.02 (3H, s), 1.13 (3H, s, 21-CH_3), 2.03 (3H, s, $OCOCH_3$), 5.12 (1H, dd, J=11, 6 Hz, H-3α); MS m/z: 486 $(M-H_2O)^+$, 468, 426, 393, 362 (base), 344, 259, 95, 69; HRMS: Found m/z 486.4084 $(M-H_2O)^+$, Calcd for $C_{32}H_{54}O_3$ 486.4072.

7β,20(S)-Dammaran-3β-yl acetate (14): Oil; IR λ_{max} : 3450 (OH), 1720, 1250 (OCO) cm^{-1}; ^1H NMR: δ 0.86 (3H, s), 0.87 (6H, s), 0.88 (6H, d, J=7 Hz), 0.97 (3H, s), 1.14 (3H, s, 21-CH_3), 2.05 (3H, s, $OCOCH_3$), 3.79 (1H, dd, J=11, 4 Hz, H-7α), 4.48 (1H, dd, J=11, 6 Hz, H-3α); MS m/z: 486 $(M-H_2O)^+$, 433, 419, 343, 303, 298, 283, 249, 189, 129, 108, 95 (base), 69, 43; HRMS: Found m/z 486.4087 $(M-H_2O)^+$, Calcd for $C_{32}H_{54}O_3$ 486.4072.

12β,20(S)-Dihydroxydammaran-3β-yl acetate (15): oil: IR λ_{max} : 3350 (OH), 1725, 1250 (OCO) cm^{-1}; ^1H NMR: δ 0.86 (3H, s), 0.87 (3H, d, J=7 Hz), 0.88 (3H, d, J=7 Hz), 0.89 (3H, s), 0.91 (3H, s), 0.99 (3H, s), 1.19 (3H, s, 21-CH_3), 2.05 (3H, s, $OCOCH_3$), 3.60 (1H, td, J=10, 5 Hz, H-12α), 4.48 (1H, dd, J=11, 6 Hz, H-3α); MS m/z: 468 $(M-H_2O \times 2)^+$, 419, 341, 249, 189, 108 (base), 43; HRMS: Found m/z 468.3956 $(M-H_2O \times 2)^+$ Calcd for $C_{32}H_{52}O_2$ 468.3957.

20(S),25-Dihydroxydammaran-3β-yl acetate (16): mp 189°C (MeOH)(lit. mp 190°C)[20]; IR λ_{max} : 3400 (OH), 1730, 1250 (OCO) cm^{-1}; ^1H NMR: δ 0.85 (6H, s), 0.87 (3H, s), 0.88 (3H, s), 0.96 (3H, s), 1.14 (3H, s, 21-CH_3), 1.23 (6H, s, 26-CH_3, 27-CH_3), 2.05 (3H, s, $OCOCH_3$), 4.48 (1H, dd, J=11, 6 Hz, H-3α); MS m/z: 468 $(M-H_2O \times 2)^+$, 189, 145, 136, 127, 109 (base), 95; HRMS: Found m/z 486.4112 $(M-H_2O)^+$, Calcd for $C_{32}H_{54}O_3$ 486.4072.

Acknowledgements--We thank Professors T. Takahashi and T. Tsuyuki, The University of Tokyo, for their generous gift of the material used in this work and Dr. M. Ishibashi for measurements of the high resolution mass spectra. We are very grateful to Professor O. Tanaka, Hiroshima University, for his kind gift of dammar-24-ene-3β,12β,20(S)-triol. We are also indebted to Dr. L. J. Harrison for reading the manuscript prior to submission. This work was supported in part by a Grant-in-Aid for Cancer Research from the Ministry of Health and Welfare.

REFERENCES AND NOTES

1) A part of this paper was published in a preliminary form: M. Tori, R. Matsuda, Y. Asakawa, Tetrahedron Lett., 26, 227 (1985).
2) S. Shibata, M. Fujita, S. Itokawa, O. Tanaka, T. Ishii, Chem. Pharm. Bull., 11, 759 (1963).
3) T. Takemoto, S. Arihara, T. Nakajima, M. Okuhira, Yakugaku Zasshi, 103, 173 (1983).
4) R. Breslow, Chem. Soc. Rev., 1, 553 (1972).
5) R. Kasai, K. Shinzo, O. Tanaka, K. Kawai, Chem. Pharm. Bull., 25, 1213 (1974).
6) T. Ishida, Y. Asakawa, T. Takemoto, J. Pharm. Sci., 71, 965 (1982) and references cited therein.
7) e. g. D. L. Heywood, B. Phillips, H. A. Stansbury, Jr., J. Org. Chem., 61, 281 (1961).
8) N. C. Deno, L. A. Messer, Chem. Commun., 1976, 1051; N. C. Deno, M. D. Meyer, J. Org. Chem.., 44, 3383 (1979).
9) W. Müller, H. -J. Schneider, Angew. Chem., Int. Ed. Engl., 18, 407 (1979).
10) N. Takaishi, Y. Fujikura, Y. Inamoto, Synthesis, 1983, 293.
11) M. Tori, R. Matsuda, Y. Asakawa, Chem. Lett., 1985, 167.
12) M. Tori, R. Matsuda, Y. Asakawa, Bull. Chem. Soc. Jpn., 58, 2523 (1985).
13) M. Tori, M. Sono, Y. Asakawa, Bull. Chem. Soc. Jpn., 58, 2669 (1985).
14) J. Asakawa, R. Kasai, K. Yamasaki, O. Tanaka, Tetrahedron, 33, 1935 (1977).
15) M. Tori, T. Tsuyuki, T. Takahashi, Bull. Chem. Soc. Jpn., 50, 3349 (1977).
16) Yields shown are isolation yields based on consumed starting material.
17) T. R. Govindachari, N. Viswanathan, P. A. Mohamed, Tetrahedron, 27, 4991 (1971).
18) T. Ikeda, K. Kitao, Mokuzai Gakkaishi, 20, 460 (1974).
19) J. Fossy, D. Lefort, M. Massoudi, J. -Y. Nedelec, J. Sorba, Can. J. Chem., 63, 678 (1985).