PREPARATION AND CONFORMATIONAL STUDY OF 19β,28-EPOXY-18α-OLEAN-5-ENE DERIVATIVES[+]

Martin DRAČÍNSKÝ[a1], Václav RICHTR[b], Václav KŘEČEK[a], † Jan SEJBAL[a], Jiří KLINOT[a] and Miloš BUDĚŠÍNSKÝ[c],*

[a] *Department of Organic Chemistry, Faculty of Science, Charles University, 128 40 Prague 2, Czech Republic; e-mail:* [1] *martindraca@yahoo.com*
[b] *Department of Chemistry, University of West Bohemia, 306 19 Plzeň, Czech Republic; e-mail: richtr@kch.zcu.cz*
[c] *Institute of Organic Chemistry and Biochemistry, Academy of Sciences of the Czech Republic, 166 10 Prague 6, Czech Republic; e-mail: milos.budesinsky@uochb.cas.cz*

Dedicated to Professor Jaroslav Podlaha on the occasion of his 70th birthday.

New oleanane type triterpenoids with the 5(6) double bond were prepared using partial demethylation on carbon C-4. The starting compound was 23-hydroxybetulin (**1b**) and the key reaction was the methylation of 19β,28-epoxy-24-nor-18α-olean-4-en-3-one (**3b**). The 5(6) double bond was used in preparation of new derivatives with an epoxy or oxo substituent in ring B. The conformation of ring A of new type 3-oxo oleanane derivatives with a double bond or a substituent on ring B was elucidated from vicinal coupling constants of hydrogen atoms in positions 1 and 2.

Keywords: Triterpenoids; Triterpenes; Oleananes; Lupanes; NMR spectroscopy; Conformation analysis.

Triterpenoids are naturally occurring compounds, which are widespread in higher plants and some marine organisms. The wide occurrence of triterpenoids in nature has evoked interest in their biological activity. For a long time triterpenoids were considered to have no biological activity at all. In the last decade, a very interesting anti-HIV and anti tumour activities were found for about 50 triterpenoids[2–11]. Moreover, a complete lack of toxicity was found even for very high doses.

[+] Part CXIII in the series Triterpenes; Part CXII see ref.[1]

Triterpenoids substituted in ring B have been occasionally isolated from natural sources[12–17] in small quantities and the published structures were often revised. No synthetic route to these compounds has been published yet and usually no reactions were performed with isolated compounds. Moreover, conformational behaviour of ring A in such B-ring substituted compounds should be interesting especially in comparison with steroids and 4,4-dimethyl steroids.

One of the possibilities for introduction of a functional group to ring B is radical functionalisation, but in our previously published[18,19] attempts on radical functionalisation of lupane and oleanane derivatives we did not find any products with substituted ring B. A second possible route uses common hydroxy group in the position 3. The main problem of this route is quarternary carbon atom in the position 4.

Our strategy was based on two known reaction routes: Key intermediate is 4-monomethyl derivative of oleanane with double bond in position 4, which should be available from saturated 4-monomethyl derivative. Such demethylation of common triterpenoids is known[20] as well as conversion to 4-unsaturated derivatives[20]. Secondly, it was shown previously[21–23] that methylation of 3-oxo steroids with double bond in position 4 using methyl iodide after reaction with potassium *tert*-butoxide prefers position 4. The result of the methylation is usually a mixture of 4-monomethyl and 4,4-dimethyl derivatives with double bond in position 5 in ring B (see also ref.[24]).

An important compound in our route is known 19β,28-epoxy-24-nor-18α-oleanan-3-one (**3a**). It can be prepared either from betulin (**1a**) by a multistep process[20,25–27] or from 23-hydroxybetulin[28] (**1b**). In this work 23-hydroxybetulin (**1b**), obtained by extraction of the bark of *Sorbus aucuparia* L., was used and transformed to norketone **3a** by modification of the method published previously[28].

23-Hydroxybetulin (**1b**) was transformed to 19β,28-epoxy-18α-oleanane-3β,23-diol diformate (**2a**) which was hydrolysed to 19β,28-epoxy-18α-oleanane-3β,23-diol (**2b**). Oxidation of **2b** with the Jones reagent directly to norketone **3a**. Norketone **3a** was brominated and dehydrobrominated in one step to give unsaturated norketone **3b**.

A previously published method[29] was used for the methylation of norketone **3b**. The reaction mixture provided starting norketone **3b** and expected 4,4-dimethyl ketone **4a** with double bond in position 5 in the ratio approximately 1:1. To increase the yield of ketone **4a**, the reaction conditions were modified. Neither the prolongation of the reaction with potassium *tert*-butoxide nor the prolongation of the methylation enhanced the yields. When higher concentrations of potassium *tert*-butoxide and methyl

iodide were used, the yield of ketone **4a** slightly increased but three by-products appeared. When high excess of both potassium *tert*-butoxide and methyl iodide were used, the result of the reaction was a mixture, which did not contain starting norketone **3b** but, in addition to ketone **4a** (24%) three less polar compounds were isolated and identified as 2α-methyl ketone **4b** (12.5%), 2β-methyl ketone **4c** (9.5%) and 2,2-dimethyl ketone **4d** (24%). Because 2-methyl ketones **4b** and **4c** in alkaline conditions (sodium hydroxide in the mixture of ethanol and benzene) isomerise to an equilibrium mixture, in which both ketones are present in the ratio approximately 1:1, it is impossible to decide, which position (2α, 2β or both) is methylated during the reaction.
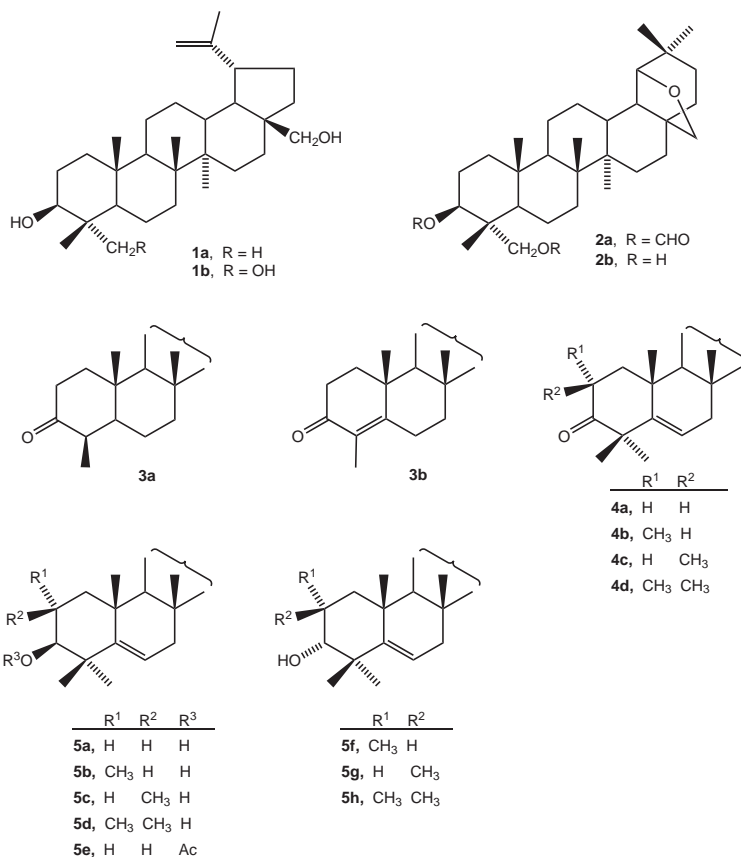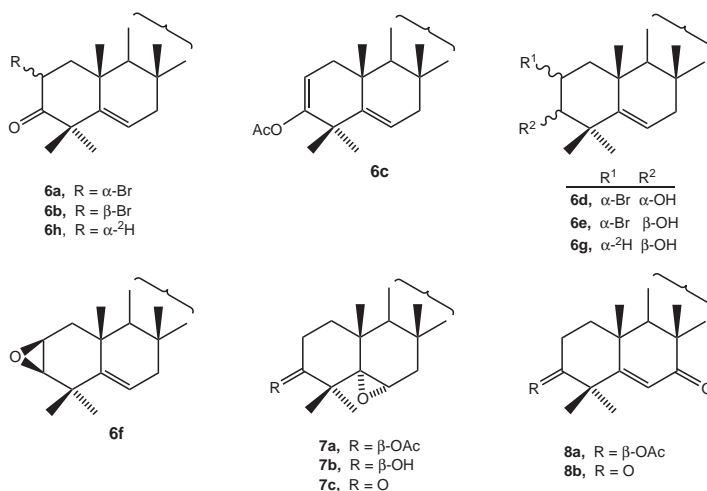
The double bond in position 5 gives in ^{13}C NMR spectra of all four 3-oxo derivatives **4a**–**4d** (for complete assignment see Table I) two signals – singlet at 145–147 ppm (C-5) and doublet at 118–122.4 ppm (C-6). The number of signals in ^{13}C NMR spectra as well as molecular ions in EI mass spectra confirm the number of methyl groups, which were introduced during methylation. In ^1H NMR spectra H-6 appears as doublet of doublets with coupling constants $J(6,7\beta) = 5.5$ and $J(6,7\alpha) = 2.5$ Hz, where H-7α was identified by the long range coupling with 8β methyl group. Multiplet of H-2, splitted by two protons on C-1 and three protons of the methyl group, is present in the ^1H NMR spectra of 2-methyl ketones. Configuration on C-2 is discussed together with conformation later on.

Ketones **4a**–**4d** were reduced to hydroxy derivatives **5a**–**5d** and **5f**–**5h**. Reduction of ketone **4a** with sodium borohydride gave hydroxy derivative **5a**, which afforded acetyl derivative **5e** after reaction with acetic anhydride. Reduction of ketone **4b** with sodium borohydride gave a mixture of 3α- and 3β-alcohols **5b** (60%) and **5f** (7.5%). Lithium aluminium hydride was used for the reduction of ketones **4c** and **4d**, because they could not be reduced with sodium borohydride (obviously because of steric hindrance of carbonyl group). In both cases a mixture of 3β- (**5c**, **5d**) and 3α- (**5g**, **5h**) alcohols appeared. All hydroxy derivatives show in IR spectra a band of OH group and in ^1H NMR spectra signal characteristic of 5-double bond.

For the determination of configuration and conformation of the above prepared triterpenoids and for comparison of the stereochemistry of reactions in compounds with and without double bond in ring B, we prepared some other 2,3-disubstituted derivatives. In ref.[30] the bromination of 4,4-dimethylcholest-5-en-3-one is published. It gives 2α-bromo derivative as a sole product. In our case, reaction of ketone **4a** with bromine in acetic acid with a drop of hydrobromic acid gave a mixture, which contained, in addition to 2α-bromo derivative **6a**, also 10–15% of 2β-bromo derivative **6b**. Attempts to isolate pure 2α-bromo derivative **6a** were not successful. Pure 2α-bromo derivative was obtained by addition of bromine to enol acetate **6c** (prepared from ketone **4a** by heating with isopropenyl acetate under catalysis of 4-toluenesulfonic acid, for analogy, see refs[31,32]) or by oxidation of bromohydrin **6e**. Reduction of 2α-bromo derivative **6a** with sodium borohydride gave a mixture of bromohydrins **6d** and **6e**. Bromohydrin **6e**, when refluxed with ethanolic solution of potassium hydroxide, gives epoxide **6f**. Bromohydrin **6d** under the same conditions gives ketone **4a**.

Compounds with double bond in position 5 may be used as starting material for the preparation of another B-ring substituted derivatives. We stud-

ied three reactions: epoxidation, allylic oxidation and allylic bromination. Both unsaturated 3β-acetoxy derivative **5e** and hydroxy derivative **5a** gave corresponding epoxy derivatives **7a** and **7b** after reaction with 3-chloroperoxybenzoic acid. Epoxy alcohol **7b** was oxidised to epoxy ketone **7c**. In analogy to 4,4-dimethyl steroids[30,33] it is possible to propose α-configuration for epoxy group in derivatives **7a–7c**. All attempts at allylic oxidation with selenium dioxide and chromium trioxide were not successful, on the other hand, acetate **5e** was brominated under the conditions for allyl bromination[34]. A mixture of bromo derivatives was converted without isolation to a mixture of alcohols which was oxidised to ketone **8a**. Acetoxy ketone **8a** was hydrolysed and oxidised to diketone **8b**.

6a, R = α-Br
6b, R = β-Br
6h, R = α-²H

6c

	R¹	R²
6d,	α-Br	α-OH
6e,	α-Br	β-OH
6g,	α-²H	β-OH

6f

7a, R = β-OAc
7b, R = β-OH
7c, R = O

8a, R = β-OAc
8b, R = O

Configuration and Conformation Analysis

The structure of prepared compounds was confirmed by ^{1}H and ^{13}C NMR spectra. Proton NMR data are given in Experimental and carbon-13 chemical shifts are presented in Tables I and II.

Configuration and conformation of all the derivatives prepared above was solved together. The solution is based on reduction of 2-methyl ketones. It is known[23,32,35,36] that 4,4-dimethyl-3-oxo steroid derivatives with double bond in position 5 are reduced preferably from the α-side to yield 3β-hydroxy derivatives. This is true also when substituents in position 2α or 2β are present. Large values of vicinal coupling constants $J(2,3) \sim$ 11 Hz in hydroxy derivatives **5b** and **5g** indicate antiperiplanar position of

<small>TABLE I</small>
Carbon-13 chemical shifts of compounds **1–5d** in CDCl$_3$

Carbon	1b	2a^a	2b	4a	4b	4c	4d	5a	5b	5c	5d
1	38.28	38.15	38.61	35.46	44.64	48.16	42.51	39.21	48.79	45.58	53.76
2	26.24	23.19	26.89	33.63	39.53	36.84	52.77	27.47	30.60	32.30	35.73
3	76.71	74.28	76.62	217.32	220.09	219.95	222.97	78.18	83.80	81.17	84.15
4	41.55	41.47	41.86	48.35	48.52	48.29	47.87	42.05	40.06	40.12	37.99
5	49.73	47.91	50.09	146.84	146.66	147.09	114.19	145.85	146.14	147.24	146.01
6	18.24	17.84	18.36	119.14	118.62	119.69	117.97	119.81	119.38	119.84	119.90
7	33.80	33.43	33.67	32.46	32.46	32.32	32.19	33.69	33.69	33.28	33.62
8	40.75	40.62	40.58	38.42	38.32	38.63	37.71	37.83	37.84^b	38.61	37.99
9	50.24	51.05	51.06	44.44	44.06	46.02	44.66	47.25	47.11	48.02	48.18
10	36.89	37.06	33.14	37.33	37.12	37.85	38.51	37.47	37.87^b	37.83	38.24
11	20.69	21.07	21.00	22.97	22.79	23.34	23.35	22.59	22.62	22.71	22.64
12	25.07	26.40^b	26.41	25.98	25.92	25.94	25.97	26.01	25.98	26.78	26.01
13	37.14	34.13	34.11	34.55	34.47	34.57	34.56	34.56	34.56	34.66	34.53
14	42.58	40.75	40.73	39.83	39.79	39.82	39.81	40.07	41.97	41.06	40.16
15	26.85	26.36^b	26.41	26.98	26.89	27.00	26.97	27.04	27.00	26.91	26.89
16	29.02	36.72	36.72	36.69	36.63	36.67	36.68	36.73	36.71	36.78	36.73
17	47.58	41.47	41.46	41.47	41.40	41.45	41.46	41.54	41.51	41.53	41.51
18	48.63	46.80	46.80	46.90	46.84	46.86	46.86	46.96	46.94	47.00	46.97
19	47.67	87.93	87.95	87.97	87.90	87.96	87.95	88.05	88.04	88.05	88.02
20	150.43	36.27	36.25	36.26	36.20	36.25	36.25	36.30	36.27	36.30	36.27
21	29.60	32.69	32.69	32.67	32.61	32.65	32.66	32.73	32.71	32.75	32.72
22	33.80	26.22	26.23	26.07	26.03	26.07	26.11	26.16	26.14	26.05	26.26
23	72.00	64.44	71.88	28.02	29.23^b	28.09	30.54^b	28.02	28.37	28.01	24.85
24	11.17	12.92	11.33	29.89	30.15^b	29.73	30.27^b	20.64	20.95	18.86	21.83
25	16.28	16.94	16.82	17.41	17.80	17.27	18.88	22.73	23.68	23.90	23.44
26	15.80	15.69	15.72	15.43	15.59	14.89	15.08	16.38	16.37	16.11	16.46
27	14.59	13.50	13.54	14.14	14.06	14.26	14.18	14.39	14.34	14.22	14.24
28	59.91	71.24	71.24	71.14	71.07	71.11	71.13	71.17	71.14	71.19	71.16
29	109.47	24.56	24.54	24.52	24.48	24.51	24.52	24.55	24.53	24.56	24.53
30	18.92	28.81	28.81	28.75	28.71	28.73	28.72	28.80	28.78	28.81	28.78
2α-Me	–	–	–	–	19.76	–	32.26^c	–	19.23	–	31.40^b
2β-Me	–	–	–	–	–	20.24	31.26^c	–	–	31.17	33.82^b

^{a} Signals of O-CH=O groups at δ 175.23. $^{b,\ c}$ Signals with identical symbols may be mutually interchanged.

TABLE II
Carbon-13 chemical shifts of compounds **5e–8b** in CDCl$_3$

Carbon	5e	5f	6a	6d	6e	6f	7a	7b	7c	8a	8b
1	38.89	43.54	46.94	42.98	43.50	38.94	34.38	34.70	35.05	38.52	38.58
2	23.86	32.19	42.76	56.03	59.35	53.58	23.74	27.47	33.51	23.16	34.20
3	79.97	81.08	221.09	79.66	81.87	61.03	77.52	75.18	217.02	77.86	213.66
4	40.84	40.18	49.84	45.57	50.39	35.28	37.68	37.65	47.78	51.12	33.14
5	145.05	142.75	145.17	140.55	143.92	144.17	68.29	68.52	68.66	170.53	170.68
6	120.49	122.40	120.02	122.65	121.11	119.92	54.90	54.82	55.27	125.73	124.24
7	33.71	33.86	32.44	33.69	33.55	33.20	32.17	32.23	31.81	206.44	205.21
8	37.84	38.09	38.52	38.19	37.99	38.09	37.06	37.05	36.29	41.20[a]	51.01[a]
9	47.20	47.51	43.74	46.90	46.80	46.95	41.59	41.67	40.67	49.27	46.53
10	37.43	37.93	38.84	40.86	40.84	37.36	40.03	41.06	37.94	37.41	40.80
11	22.57	22.41	22.89	22.29	22.62	22.75	20.32	21.58	21.80	21.12	22.34
12	26.00	26.01	25.94	25.93	25.95	26.00	26.26	26.27	25.91	26.26	25.96
13	34.55	34.51	34.51	34.38	34.47	34.36	34.25	34.25	34.35	35.56	36.08
14	40.08	42.22	39.92	40.26	40.13	40.09	41.22	41.19	40.52	41.43[a]	48.57[a]
15	27.03	27.01	26.90	26.90	26.96	26.85	26.59	26.50	26.74	28.19	28.30
16	36.73	36.76	36.67	36.68	36.69	36.74	36.63	36.64	36.64	36.52	36.53
17	41.53	41.56	41.48	41.51	41.50	41.50	41.52	41.52	41.46	41.43	41.22
18	46.96	46.99	46.87	46.90	46.90	46.77	47.00	47.00	47.00	46.43	45.71
19	88.06	88.09	87.99	88.05	88.02	88.00	87.83	87.84	87.95	87.70	87.85
20	36.30	36.31	36.28	36.28	36.28	36.28	36.22	36.22	36.25	36.23	36.28
21	32.73	32.75	32.66	32.69	32.69	32.71	32.66	32.66	32.64	32.58	32.61
22	26.13	26.12	25.94	25.93	25.95	26.19	26.09	26.10	25.99	26.23	26.23
23	27.93	27.47[a]	32.18	29.15	28.82	30.74	21.66	21.58	23.00	26.26	28.71
24	20.43	18.70[b]	29.09	28.81	23.64[a]	22.41	21.04	19.88	24.29	21.20	26.89
25	23.98	21.90[b]	16.99	21.74	20.71[a]	29.56	20.29[a]	20.29	19.71	22.18	17.24
26	16.42	16.51	15.61	16.63	16.41	16.24	20.38[a]	20.29	19.03	15.12	15.74
27	14.35	14.46	14.11	14.45	14.32	14.16	13.33	13.33	14.11	13.73	13.42
28	71.17	71.19	71.12	71.13	71.12	71.15	71.22	71.21	71.09	71.39	71.41
29	24.55	24.57	24.54	24.54	24.54	24.54	24.53	24.53	24.50	24.54	24.52
30	28.80	28.81	28.75	28.77	28.77	28.78	28.78	28.77	28.74	28.80	28.81
2α-Me	–	28.64[a]	–	–	–	–	–	–	–	–	–
OAc	–	–	–	–	–	–	–	–	–	171.30	–
	–	–	–	–	–	–	–	–	–	21.20	–

[a, b] Signals with identical symbols may be mutually interchanged.

H-2 and H-3 which is possible only in *trans*-configuration. On the other hand, small values of vicinal constant $J(2,3) \sim 2$ Hz in hydroxy derivatives **5f** and **5c** are characteristic of *cis*-configuration. From this it follows that hydroxy derivatives **5b** and **5f** and ketone **4b** have 2α-methyl group and therefore configuration of 2-methyl group in compounds **5c**, **5g** and **4c** must be β. Configuration of hydroxy groups in 2,2-dimethyl-3-hydroxy derivatives **5d** and **5h** is proposed on the above discussed reduction of 3-oxo derivatives and on comparison of chemical shifts of protons in position 3 with those in other 2-methyl-3-hydroxy derivatives.

Configuration of 2α-bromo-3-oxo derivative **6a** and its 2β-isomer **6b** is based on the known[30,37–39] stereochemistry of bromination of 4,4-dimethyl-3-oxo steroid derivatives, where products with 2α-configuration are known to be preferred. From the reactions of bromohydrins **6e** and **6d** with potassium hydroxide it is clear that the former must have *trans*-configuration and the latter gives ketone **4a**, therefore it must have *cis*-configuration. Configuration of epoxide **6f** is also obvious from comparison of its ^{1}H NMR spectrum with the spectrum of saturated epoxy derivative[31,32].

For study of the conformation of ketone **4a**, we prepared 2α-deuterio-3-oxo derivative **6h** from epoxide **6f** using the reaction with the known stereochemistry of the opening of epoxide ring. Triterpenoid and 4,4-dimethylsteroid 2β,3β-epoxides open usually against the Fürst–Plattner rule and diequatorial products appear[32,40,41]. This mechanism was observed also for reduction of epoxides with complex hydrides[40,42]. 2β,3β-Epoxide gave deuterated alcohol **6g** as the main product after reduction with lithium aluminium tetradeuteride. Deuterated alcohol **6g** was oxidised to 2α-deuterated ketone **6h**.

The conformation of ring A in triterpenoids depends on the substitution pattern. Therefore it was interesting to study the conformation of the newly prepared triterpenoids with double bond in position 5 and to compare it with the conformation of saturated analogs. Conformation of ring A in the case of 2,2-dimethyl derivatives **5d**, **5h** and **4d** is not clear because of the lack of spatial information in NMR spectra. On the other hand, a group of 2-substituted-3-hydroxy derivatives **6d**, **6e**, **5b**, **5c**, **5f** and **5g** may be compared with derivatives without double bond in ring B with well proved conformation[36]. Coupling constants $J(1,2)$ and $J(2,3)$ in the ^{1}H NMR spectra of our compounds are in very good agreement with those found in saturated analogs. This means that the double bond in position 5 does not affect the conformation of ring A of these derivatives and all have ring A in the chair conformation with the exception of 2β,3α-derivative **5g**, which prefers a boat conformation.

The conformation of 3-oxo derivatives is known to be extremely sensitive to internal effects (substituents) as well as external effects (solvent, crystal structure). We have assumed three possible conformations proposed in ref.[37]: chair (**C**) and two twisted boat conformations (**T₁** and **T₂**, see Fig. 1). Most of this conformational study is based on vicinal coupling constants between protons in positions 1 and 2. Reference $J(1,2)$ values for conformations **C** and **T₁** are taken from ref.[42], for conformation **T₂** is our estimation based on the Karplus-type equation[43] using the geometry parameters described in ref.[44]

In the case of 2α-bromo ketone **6a** we may exclude chair form because the carbonyl band in IR spectrum has a low wavenumber (1717.5 cm^{-1}) not consistent with equatorial bromine atom in chair conformation[45]. This is in agreement with observations for 2α-bromo-4,4-dimethyl-3-oxo steroid derivatives with double bond in position 5 (refs[30,38]). Coupling constants (Table III) agree with conformation **T₂** slightly distorted to relieve the steric strain between bromine and 4α-methyl group. Similar situation is in the case of 2α-methyl derivative **4b**, where some population of a chair form in the equilibrium with **T₂** conformation may be present.
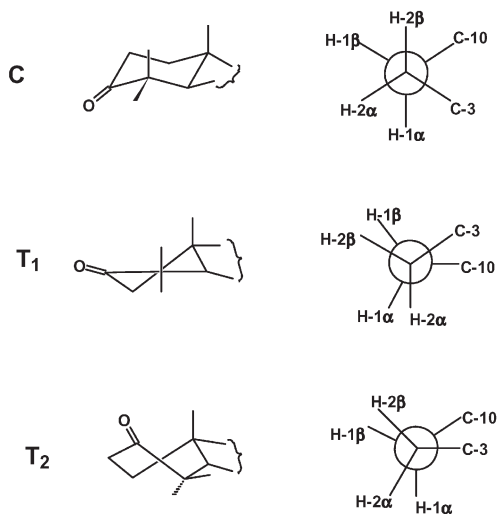
FIG. 1
Three possible conformation of ring A of 3-oxo triterpenoids

The coupling constants between protons at C-1 and C-2 in ketone **4a**, its 2α-deuterio derivative **6h** and diketone **8b** are in good agreement with conformation T_2. In the case of 2β-substituted 3-oxo derivatives **4c** and **6b** the chair conformation of ring A might be excluded on the basis of coupling constants (see Table III). These constants are in agreement with the equilibrium of T_1 and T_2, but the possibility of distorted or classical boat conformation must be also taken into account.

As a result of conformational study we may exclude the chair conformation for all studied 19β,28-epoxy-18α-olean-5-en-3-one derivatives. This applies also to cases, where for the analogs with saturated ring B the chair conformation or a chair–boat equilibrium was found.

EXPERIMENTAL

TLC was performed on silica gel plates G (Merck), visualisation by spraying with 10% sulfuric acid and heating. Kieselgel 60 G (Merck) was used for preparative TLC (spraying with 0.3% methanolic solution of morin). Column chromatography was carried out on silica gel Silpearl (Kavalier, Votice). Mixtures of light petroleum and ether ranging from 6:1 to 2:1 were used as eluents. The usual work-up means dilution of the reaction mixture with water, extraction of products with ether and successive washing of ethereal layer with dilute (1:4) hydrochloric acid (if necessary), water, saturated sodium hydrogencarbonate solution, water,

TABLE III
Vicinal coupling constants of protons in ring A of 3-oxoderivatives

	Substituent in position 2	Coupling constants, Hz			
		1α,2α	1α,2β	1β,2α	1β,2β
C^a	–	5.7	13.2	1.4	5.7
$T_1{}^a$	–	11.2	3.6	9.1	11.2
$T_2{}^b$	–	9.7	10.3	0	9.7
4a	–	9.2	10.5	1.5	9.2
8b	–	8.9	10.9	1.9	8.2
6h	α-^{2}H	–	10.5	–	9.1
4b	α-Me	–	11.7	–	7.6
6a	α-Br	–	11.5	–	8.3
4c	β-Me	11.1	–	5.7	–
6b	β-Br	10.5	–	6.0	–

[a] Taken from ref.[42]. [b] Calculated using the Karplus equation in ref.[43].

drying over anhydrous sodium sulfate and evaporation of solvents under reduced pressure. Samples for elemental analysis were dried over phosphorus pentoxide under reduced pressure. Melting points were determined on a Kofler block and are uncorrected. Optical rotations were carried out in chloroform (if not specified otherwise), c 0.3–0.5 mmol l^{-1}, at ambient temperature on an automatic polarimeter ETL-NPL (Bendix–Ericsson) with the accuracy ±2 and are given in 10^{-1} deg cm^2 g^{-1}. Infrared spectra were recorded in chloroform solution on a PE 684 (Perkin–Elmer) spectrometer. Bands were separated using the spectrometer software and are given in cm^{-1}. Ultraviolet spectra were scanned on a Unicam SP-700 instrument. Mass spectra were recorded on a Finnigan MAT-Incos 50 instrument, ionising electron energy 70 eV and direct inlet temperatures 150–180 °C. NMR spectra were recorded on a Varian UNITY-200 instrument (^{1}H at 200 MHz; ^{13}C at 50.3 MHz) in deuteriochloroform (if not specified otherwise) at room temperature (~20 °C). For ^{1}H NMR spectra, tetramethylsilane (TMS) was used as internal standard, chemical shifts (δ, ppm) and coupling constants (Hz) were obtained by first-order analysis. Carbon-13 chemical shifts were obtained from the spectra measured in deuteriochloroform and referenced to solvent signal (δ(CDCl$_3$) 77.00).

Extraction of Bark of *Sorbus aucuparia* L.

The bark of *Sorbus aucuparia* L. (common names: European mountain ash, mountain ash, rowan) (180 g) was dried to constant weight at 105 °C and then it was extracted in Soxhlet extractor with petroleum ether for 50 h. The solvent was distilled off, the dried extract (1.7 g, 0.9%) did not contain (TLC) 23-hydroxybetulin (**1b**), only a complex mixture of less polar compounds (in agreement with ref.[28]) was present. The bark was then extracted another 50 h with dried ether, the extract (5.73 g, 3.2%) contained (TLC) mainly 23-hydroxybetulin (**1b**). Further 50-h extraction of the bark with methanol gave only more polar compounds. The ethereal extract (200 mg) was filtered through a column of aluminium oxide (20 g). Ether (300 ml) and then ether–methanol mixture (2:1) were used as eluents. Lup-20(29)-ene-3β,23,28-triol (23-hydroxybetulin, **1b**) was obtained in the yield 120 mg, m.p. 260–262 °C (ethyl acetate–light petroleum), [α]$_D$ +22 (ref.[28] m.p. 259–260 °C, [α]$_D$ +24.6). IR: 895 (C=CH$_2$); 1030, 1050 (C–O); 1650 (C=C); 3490 (OH). ^{1}H NMR: 0.868 s, 6 H (2 × CH$_3$); 0.974 d, 3 H (CH$_3$, J = 0.8); 1.023 s, 3 H (CH$_3$); 1.678 dd, 3 H (CH$_3$-30, J(30,29a) = 1.4, J(30,29b) = 0.7); 2.37 m, 1 H (H-19); 3.33 bd, 1 H (H-28a, J(28a,28b) = 10.8); 3.79 bd, 1 H (H-28b, J(28b,28a) = 10.8); 3.41 d, 1 H (H-23a, J(23a,23b) = 10.3); 3.71 d, 1 H (H-23b, J(23b,23a) = 10.3); 3.61 m, 1 H (H-3, ΣJ = 16.2); 4.58 dq, 1 H (H-29a, J(29a,29b) = 2.4, J(29a,30) = 1.4); 4.68 dq, 1 H (H-29b, J(29b,29a) = 2.4, J(29b,30) = 0.7). For ^{13}C NMR, see Table I. EI-MS, m/z (%): 458 (9, M$^+$), 443 (6), 440 (6), 427 (17), 425 (6), 422 (6), 415 (5), 409 (13), 379 (6), 245 (33), 203 (100).

19β,28-Epoxy-18α-oleanane-3β,23-diyl Diformate (2a)

23-Hydroxybetulin (**1b**; 1.87 g, 4.1 mmol) was refluxed for 2 h with 85% formic acid (25 ml). During another 1 h formic acid (17 ml) was distilled off. The residue was cooled to room temperature and dissolved in ether. The ethereal solution was washed with water and saturated solution of sodium hydrogencarbonate and worked up in the usual manner. The crude product (1.69 g) was dried at 105 °C. A sample for analysis was obtained by column chromatography of the crude product (240 g) on silica gel (20 g) with benzene (310 ml) and benzene–ether mixture (20:1, 200 ml) as eluents. Diformate **2a** (150 mg) was obtained after

crystallisation from acetone–water, m.p. 218–220 °C, $[\alpha]_D$ +64 (ref.[28]: m.p. 217–218 °C, $[\alpha]_D$ +62). IR: 1032 (C–O–C); 1185 (HCO–O); 1728 (C=O). ^{1}H NMR: 0.803 s, 3 H (CH₃); 0.864 s, 3 H (CH₃); 0.923 s, 6 H (2 × CH₃); 0.933 s, 3 H (CH₃); 0.984 s, 3 H (CH₃); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.77 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 3.82 d, 1 H (H-23a, J(23a,23b) = 11.7); 3.99 d, 1 H (H-23b, J(23b,23a) = 11.7); 4.95 m, 1 H (H-3, ΣJ = 16.2); 8.06 d, 1 H (3β-OCHO, J(OCHO,3) = 0.9); 8.10 s, 1 H (23-OCHO). For ^{13}C NMR, see Table I.

19β,28-Epoxy-18α-oleanane-3β,23-diol (**2b**)

a) Diformate **2a** (1.45 g, 2.82 mmol) was refluxed for 5 h with a solution of potassium hydroxide (5.0 g, 89.3 mmol) in ethanol (50 ml). The mixture was then diluted with water (150 ml) and extracted with chloroform (3 × 30 ml). The chloroform layer was worked up in the usual manner and the crude product (1.05 g) was dried at 105 °C. The product was purified by column chromatography on aluminium oxide (200 g) with benzene (750 ml) and benzene–ether mixture (1:1, 600 ml) as eluents. Diol **2b** (700 mg, 54%) was obtained after repeated crystallisation from dichloromethane–acetone, m.p. 253–256 °C, $[\alpha]_D$ +47. IR: 1035 (C–O–C); 1050 (C–O); 3458 and 3630 (OH). ^{1}H NMR: 0.796 s, 3 H (CH₃); 0.871 s, 3 H (CH₃); 0.892 s, 3 H (CH₃); 0.910 s, 3 H (CH₃); 0.928 s, 3 H (CH₃); 0.975 s, 3 H (CH₃); 3.41 d, 1 H (H-23a, J(23a,23a) = 10.5); 3.71 d, 1 H (H-23b, J(23b,23a) = 10.5); 3.44 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.77 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 3.63 m (H-3, ΣJ = 16.2). For ^{13}C NMR, see Table I. EI-MS, *m/z* (%): 458 (35, M⁺), 440 (25), 427 (28), 422 (98), 410 (88), 387 (91), 369 (21), 355 (70), 245 (70), 149 (100).

b) The ether extract from the bark of *Sorbus aucuparia* L. (5.73 g) was refluxed for 5 h with 85% formic acid (100 ml). Then formic acid was distilled off and the residue was dried at 110 °C. To the crude diformate (5.89 g) a solution of potassium hydroxide (13 g) in ethanol (130 ml) was added. The mixture was refluxed for 5 h and then it was left standing at room temperature overnight. Then the mixture was diluted with water (600 ml), sodium chloride was added and then it was extracted with chloroform (3 × 120 ml). The chloroform extracts were washed with water (3 × 100 ml) and then worked up in the usual manner. The evaporation residue (4 g) was chromatographed on aluminium oxide (400 g). After elution with benzene (1000 ml) and benzene–ether (1:1, 2800 ml), a mixture of compounds was obtained which were (TLC) less polar then 23-hydroxyallobetulin (**2b**). Further elution with benzene–ether–ethanol (1:1:1, 300 ml) gave pure 23-hydroxyallobetulin (2.37 g, 41.4%) and further elution with the same solvent mixture gave a mixture (200 mg) of more polar compounds.

19β,28-Epoxy-24-nor-l8α-oleanan-3-one (**3a**)

Jones reagent (5 ml) was dropwise added to a stirred acetic acid solution (30 ml) of the alcohol **2b** (500 mg, 1.1 mmol) during 2 min. After another 3 min of stirring, methanol (1 ml) and water (20 ml) were added. The mixture was refluxed for 3 h, then water (50 ml) was added dropwise and the mixture was left standing at room temperature overnight. The precipitate was filtered off, washed with water and dried at 105 °C. The crude product (380 mg) contained (TLC), in addition to norketone **3a**, small amounts of more polar compounds. The product was purified by column chromatography on aluminium oxide (200 g) with benzene as eluent. After crystallisation from chloroform–heptane, norketone **3a** (306 mg, 66%) was obtained, m.p. 214–216 °C, $[\alpha]_D$ +83 (ref.[28]: m.p. 214–215 °C, $[\alpha]_D$ +84; ref.[20]: m.p.

215–216 °C, [α]$_D$ = +82). IR and ^{1}H NMR spectra were identical with spectra of norketone **3a** described in ref.[20]

19β,28-Epoxy-24-nor-18α-olean-4-en-3-one (3b)

To a solution of norketone **3a** (520 mg, 1.2 mmol) in acetic acid (30 ml), one drop of 46% hydrobromic acid was added and then a solution of bromine (212 mg, 1.3 mmol) in acetic acid (30 ml) was dropwise added during 30 min with stirring. Five minutes after addition of the last portion of bromine, 46% hydrobromic acid (10 ml) was added and the mixture was allowed to stand in the dark at room temperature for 10 days. Then the mixture was diluted with water and extracted with chloroform. The chloroform layer was washed with water, a saturated solution of sodium hydrogencarbonate and with a solution of sodium sulfite and then it was worked up in the usual manner. Norketone **3b** (500 mg, 95%) was obtained, pure enough (TLC) for the use in another reaction. A sample for analysis was obtained by column chromatography on silica gel with benzene–ether mixture (10:1) as eluent. M.p. 237–240 °C, [α]$_D$ +127 (ref.[20]: m.p. 238–241 °C, [α]$_D$ +128). IR and ^{1}H NMR spectra were identical to those of ketone **3b** described in ref.[20]

Methylation of 19β,28-Epoxy-24-nor-18α-olean-4-en-3-one (3b)

Method A. Potassium (600 mg, 15.4 mmol) was dissolved in warm *tert*-butyl alcohol (120 ml). Norketone **3b** (2.8 g, 6.6 mmol) was added to the solution under nitrogen. The mixture was refluxed under nitrogen for 1 h and then a solution of methyl iodide (6 g, 4.22 mmol) in *tert*-butyl alcohol (50 ml) was added dropwise within 5 h and then the mixture was left standing under nitrogen at room temperature overnight. Then 100 ml of solvents were distilled off, the mixture was diluted with water (300 ml) and extracted with chloroform (3 × 50 ml). The chloroform extract was washed with water and then worked up in the usual manner. The evaporation residue was separated by column chromatography on aluminium oxide (300 g), benzene and benzene–ether mixture (10:1) were used as eluents. Two compounds obtained were crystallised from chloroform–methanol (they are introduced according to their increasing polarity).

a) 19β,28-Epoxy-18α-olean-5-en-3-one (**4a**; 1.4 g, 49%), m.p. 236–239 °C, [α]$_D$ +90. IR: 1033 (C–O–C); 1670 (C=C); 1711 (C=O). UV: 296 nm (ε 42). ^{1}H NMR: 0.814 s, 3 H (CH$_3$-29); 0.846 d, 3 H (CH$_3$-25, *J* = 0.5); 0.929 s, 3 H (CH$_3$-27); 0.945 s, 3 H (CH$_3$-30); 1.000 d, 3 H (CH$_3$-26, *J* = 1.0); 1.230 s, 3 H (CH$_3$-24); 1.248 s, 3 H (CH$_3$-23); 1.58 m, 1 H (H-1α); 2.01 ddd, 1 H (H-1β, *J*(1β,1α) = 13.4, *J*(1β,2α) = 1.5, *J*(1β,2β) = 9.2); 2.44 m, 1 H (H-2β); 2.54 m, 1 H (H-2α, *J*(2α,1α) = 9.2, *J*(2α,1β) = 1.5, *J*(2α,2β) = 18.9); 1.74 dd, 1 H (H-7β, *J*(7β,6) = 5.8, *J*(7β,7α) = 17.5); 2.16 bd, 1 H (H-7α, *J*(7α,7β) = 17.5); 3.47 d, 1 H (H-28a, *J*(28a,28b) = 7.8); 3.81 bd, 1 H (H-28b, *J*(28b,28a) = 7.8); 3.55 s, 1 H (H-19); 5.55 dd, 1 H (H-6, *J*(6,7α) = 2.4, *J*(6,7β) = 5.8). For ^{13}C NMR, see Table I. EI-MS, *m/z* (%): 438 (25, M$^+$), 423 (3), 420 (6), 410 (3), 407 (2), 405 (3), 395 (6), 377 (2), 367 (9), 315 (11), 245 (16), 124 (100). For C$_{30}$H$_{46}$O$_2$ (438.7) calculated: 82.13% C, 10.57% H; found: 82.25% C, 10.34% H.

b) Starting ketone **3b** (1.3 g, 35%).

Method B) Potassium (10 g, 256 mmol) was dissolved in warm *tert*-butyl alcohol (200 ml). To the solution norketone **3b** (3.0 g, 7.1 mmol) was added under nitrogen. The mixture was refluxed under nitrogen for 3 h and then methyl iodide (25 ml, 401 mmol) was dropwise added during 20 min while refluxing and then the mixture was refluxed for another 2 h. Then the mixture was cooled to room temperature, diluted with water and worked up as in

method *A*. The mixture was separated by column chromatography on silica gel (120 g) with petroleum ether–ether mixture (8:1) as eluent. The following compounds were obtained (according to increasing polarity).

a) 19β,28-Epoxy-2,2-dimethyl-18α-olean-5-en-3-one (**4d**; 0.8 g, 24%), m.p. 275–277 °C (chloroform–methanol), $[\alpha]_D$ +56. IR: 1036 (C–O–C); 1663 (C=C); 1696 (C=O). UV: 298 nm (ε 35). [1H] NMR: 0.818 s, 3 H (CH$_3$-29); 0.863 bs, 3 H (CH$_3$-25); 0.926 s, 3 H (CH$_3$-27); 0.946 s, 3 H (CH$_3$-30); 0.986 d, 3 H (CH$_3$-26, *J* = 0.9); 1.140 s, 3 H (2β-CH$_3$); 1.197 s, 3 H (2α-CH$_3$); 1.270 s, 3 H (CH$_3$-23); 1.307 s, 3 H (CH$_3$-24); 1.51 bd, 1 H (H-1α, *J*(1α,1β) = 13.6); 1.73 dd, 1 H (H-7β, *J*(7β,6) = 6.0, *J*(7β,7α) = 17.4); 1.98 d, 1 H (H-1β, *J*(1β,1α) = 13.6); 2.16 bd, 1 H (H-7α, *J*(7α,7β) = 17.4); 3.46 d, 1 H (H-28a, *J*(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, *J*(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 5.53 dd, 1 H (H-6, *J*(6,7α) = 2.3, *J*(6,7β) = 6.0). For [13C] NMR, see Table I. EI-MS, *m/z* (%): 466 (26, M$^+$), 451 (3), 448 (3), 438 (2), 435 (2), 433 (1), 423 (5), 395 (3), 315 (7), 245 (17), 180 (16), 152 (100). For C$_{32}$H$_{50}$O$_2$ (466.8) calculated: 82.35% C, 10.80% H; found: 82.49% C, 10.67% H.

b) 19β,28-Epoxy-2β-methyl-18α-olean-5-en-3-one (**4c**; 0.3 g, 9.5%), m.p. 249–251 °C (chloroform–methanol), $[\alpha]_D$ +105. IR: 1030 (C–O–C); 1663 (C=C); 1707 (C=O). UV: 292 nm (ε 36). [1H] NMR: 0.814 s, 3 H (CH$_3$-29); 0.826 d, 3 H (CH$_3$-25, *J* = 0.9); 0.937 s, 3 H (CH$_3$-27); 0.944 s, 3 H (CH$_3$-30); 0.950 d, 3 H (CH$_3$-26, *J* = 0.8); 1.084 d, 3 H (2β-CH$_3$, *J* = 6.7); 1.245 s, 6 H (CH$_3$-23,24); 1.57 dd, 1 H (H-1β, *J*(1β,1α) = 13.3, *J*(1β,2α) = 5.7); 1.72 dd, 1 H (H-7β, *J*(7β,6) = 6.0, *J*(7β,7α) = 17.3); 2.04 ddq, 1 H (H-1α, *J*(1α,1β) = 13.3, *J*(1α,2α) = 11.1, *J*(1α,25) = 0.9); 2.19 bd, 1 H (H-7α, *J*(7α,7β) = 17.3); 2.66 ddq, 1 H (H-2α, *J*(2α,1α) = 11.1, *J*(2α,1β) = 5.7, *J*(2α,CH$_3$) = 6.7); 3.46 d, 1 H (H-28a, *J*(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, *J*(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 5.61 dd, 1 H (H-6, *J*(6,7α) = 2.3, *J*(6,7β) = 6.0). For [13C] NMR, see Table I. EI-MS, *m/z* (%): 452 (32, M$^+$), 437 (4), 434 (5), 421 (2), 419 (3), 409 (4), 381 (8), 315 (9), 245 (18), 166 (18), 138 (100). For C$_{31}$H$_{48}$O$_2$ (452.7) calculated: 82.25% C, 10.69% H; found: 82.39% C, 10.47% H.

c) 19β,28-Epoxy-2α-methyl-18α-olean-5-en-3-one (**4b**; 0.4 g, 12.5%), m.p. 230–233 °C (chloroform–methanol, at 205–225 °C modification change), $[\alpha]_D$ + 67. IR: 1029 (C–O–C); 1663 (C=C); 1696 (C=O). UV: 296 nm (ε 40). [1H] NMR: 0.816 s, 3 H (CH$_3$-29); 0.881 s, 3 H (CH$_3$-25); 0.925 s, 3 H (CH$_3$-27); 0.945 s, 3 H (CH$_3$-30); 1.017 bs, 3 H (CH$_3$-26); 1.177 d, 3 H (2β-CH$_3$, *J* = 7.4); 1.220 s, 3 H (CH$_3$-23); 1.260 s, 3 H (CH$_3$-24); 1.24 dd, 1 H (H-1α, *J*(1α,1β) = 13.2, *J*(1α,2) = 11.7); 1.73 dd, 1 H (H-7β, *J*(7β,6) = 5.7, *J*(7β,7α) = 17.6); 2.13 dd, 1 H (H-1β, *J*(1β,1α) = 13.2, *J*(1β,2) = 7.6); 2.15 bd, 1 H (H-7α, *J*(7α,7β) = 17.6); 2.51 m, 1 H (H-2, *J*(2,1α) = 11.7, *J*(2,1β) = 7.6, *J*(2,CH$_3$) = 7.4); 3.46 d, 1 H (H-28a, *J*(28a,28b) = 7.8); 3.81 bd, 1 H (H-28b, *J*(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 5.51 dd, 1 H (H-6, *J*(6,7α) = 2.5, *J*(6,7β) = 5.7). For [13C] NMR, see Table I. EI-MS, *m/z* (%): 452 (70, M$^+$), 437 (10), 434 (13), 421 (5), 419 (8), 409 (13), 381 (15), 315 (13), 245 (39), 166 (26), 138 (100). For C$_{31}$H$_{48}$O$_2$ (452.7) calculated: 82.25% C, 10.69% H; found: 82.10% C, 10.83% H.

d) Ketone **4a** (1.3 g, 42%).

Isomerisation of Methyl Ketones **4b** and **4c**

Both methyl ketones were isomerised in this way: To a solution of a ketone (30 mg, 0.066 mmol) in benzene (2 ml), a solution of sodium hydroxide (0.5 g, 12.5 mmol) in ethanol (10 ml) was added. The mixture was refluxed for 4 h and then diluted with water and extracted with ether. The ether layer was worked up in the usual manner. The isomerisation

products of both ketones were nearly identical (TLC) – both mixtures contained 2α-methyl ketone **4b** and 2β-methyl ketone **4c** in the ratio 1:1.

19β,28-Epoxy-18α-olean-5-en-3β-ol (5a)

To a solution of ketone **4a** (770 mg, 1.8 mmol) in a mixture of benzene (10 ml), dioxane (10 ml) and ethanol (10 ml), sodium borohydride (770 mg, 20 mmol) was slowly added while stirring. The mixture was left standing at room temperature for 48 h and then a solution of ammonium chloride (2 g, 37 mmol) in water (20 ml) was added. The mixture was stirred for 1 h and then extracted with benzene. The organic layer was worked up in the usual manner. Hydroxy derivative **5a** (700 mg, 90%) was obtained; it was pure enough to be used in another reaction. A sample for analysis was obtained by preparative TLC with benzene–ether mixture (5:1) as eluent. M.p. 273–275 °C (chloroform–octane), $[\alpha]_D$ +45. IR: 1035 (C–O–C); 3630 (OH). ^{1}H NMR: 0.805 s, 3 H (CH$_3$-29); 0.895 s, 3 H (CH$_3$-27); 0.939 s, 3 H (CH$_3$-30); 0.979 d, 3 H (CH$_3$-26, J = 0.8); 1.078 s, 6 H (CH$_3$-24,25); 1.151 s, 3 H (CH$_3$-23); 1.69 dd, 1 H (H-7β, J(7β,7α) = 18.1, J(7β,6) = 5.3); 2.19 bd, 1 H (H-7α, J(7α,7β) = 18.1); 3.20 m, 1 H (H-3, ΣJ = 16.0); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 5.57 dd, 1 H (H-6, J(6,7α) = 2.9, J(6,7β) = 5.3). For ^{13}C NMR, see Table I. EI-MS, m/z (%): 440 (16, M$^+$), 425 (9), 422 (32), 407 (22), 397 (16), 391 (13), 379 (13), 369 (15), 368 (16), 357 (78), 354 (15), 187 (100), 175 (71), 133 (97), 121 (65), 119 (80). For C$_{30}$H$_{48}$O$_2$ (440.7) calculated: 81.76% C, 10.98% H; found: 81.53% C, 10.82% H.

19β,28-Epoxy-18α-olean-5-en-3β-yl Acetate (5e)

To a solution of compound **5a** (380 mg, 0.86 mmol) in pyridine (10 ml), acetic anhydride (2 ml, 20 mmol) was added and the mixture was left standing at room temperature for two days. The crystals were filtered off, washed with 2 M hydrochloric acid and with water and then dried in air. The filtrate was diluted with 2 M hydrochloric acid and extracted with chloroform. The organic layer was washed with water, saturated solution of sodium hydrogencarbonate and then it was worked up in the usual manner. After crystallisation from chloroform–octane, another portion of product **5e** was obtained (total 290 mg, 74%). M.p. 268–270 °C, $[\alpha]_D$ +52. IR: 1037 (C–O–C); 1265 (CH$_3$COO); 1666 (C=C); 1734 (CH$_3$COO). ^{1}H NMR: 0.806 s, 3 H (CH$_3$-29); 0.894 s, 3 H (CH$_3$-27); 0.940 s, 3 H (CH$_3$-30); 0.979 d, 3 H (CH$_3$-26, J = 0.8); 1.032 s, 3 H (CH$_3$-25); 1.106 s, 3 H (CH$_3$-23); 1.151 s, 3 H (CH$_3$-24); 1.69 dd, 1 H (H-7β, J(7β,7α) = 18.3; J(7β,6) = 5.3); 2.06 s, 3 H (CH$_3$COO); 2.20 bd, 1 H (H-7α, J(7α,7β) = 18.3); 3.46 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.81 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 4.47 m, 1 H (H-3, ΣJ = 16.4); 5.58 dd, 1 H (H-6, J(6,7α) = 2.9, J(6,7β) = 5.3). For ^{13}C NMR, see Table II. EI-MS, m/z (%): 482 (4, M$^+$), 422 (17), 411 (5), 407 (22), 389 (9), 379 (9), 354 (33), 187 (100), 175 (39), 133 (47), 119 (39). For C$_{32}$H$_{50}$O$_3$ (482.7) calculated: 79.62% C, 10.44% H; found: 79.73% C, 10.20% H.

Reduction of 19β,28-Epoxy-2α-methyl-18α-olean-5-en-3-one (4b)

To a solution of 2α-methyl ketone **4b** (120 mg, 0.27 mmol) in benzene (4 ml), a solution of sodium borohydride (50 mg, 1.3 mmol) in methanol (8 ml) was added and the reaction mixture was left standing at room temperature for 36 h. Then the mixture was worked up as in the case of reduction of ketone **4a**, the evaporation residue was separated by column

chromatography on silica gel (15 g) with ether–petroleum ether (1:5) as eluent. The following compounds were obtained after crystallisation from dichloromethane–methanol.

a) 19β,28-Epoxy-2α-methyl-18α-olean-5-en-3β-ol (**5b**; 72 mg, 60%), m.p. 252–253 °C (at 180–220 °C modification change), $[\alpha]_D$ +25. IR: 1029 (C–O–C); 3632 (OH). ^{1}H NMR: 0.808 s, 3 H (CH$_3$-29); 0.892 s, 3 H (CH$_3$-27); 0.941 s, 3 H (CH$_3$-30); 0.979 d, 3 H (CH$_3$-26, J = 0.9); 0.994 d, 3 H (2α-CH$_3$, J = 6.2); 1.090 s, 3 H (CH$_3$-23); 1.109 d, 3 H (CH$_3$-25, J = 0.7); 1.155 s, 3 H (CH$_3$-24); 1.69 dd, 1 H (H-7β, J(7β,6) = 5.3, J(7β,7α) = 18.2); 2.19 bd, 1 H (H-7α, J(7α,7β) = 18.2); 2.74 bd, 1 H (H-3, J(2,3) ~ 10.2); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H, (H-28b, J(28b,28a) = 7.8); 3.47 s, 1 H (OH); 3.54 s, 1 H (H-19); 5.56 dd, 1 H (H-6, J(6,7α) = 2.8, J(6,7β) = 5.3). For ^{13}C NMR, see Table I. For C$_{31}$H$_{50}$O$_2$ (454.7) calculated: 81.88% C, 11.08% H; found: 81.71% C, 11.24% H.

b) 19β,28-Epoxy-2α-methyl-18α-olean-5-en-3α-ol (**5f**; 9 mg, 7.5%), m.p. 263–264 °C, $[\alpha]_D$ +28. IR: 1030 (C–O–C); 3584 (OH). ^{1}H NMR: 0.810 s, 3 H (CH$_3$-29); 0.898 s, 3 H (CH$_3$-27); 0.943 s, 3 H (CH$_3$-30); 0.948 d, 3 H (2α-CH$_3$, J = 6.7); 0.980 d, 3 H (CH$_3$-26, J = 0.9), 1.124 d, 3 H (CH$_3$-25, J = 0.8); 1.147 s, 3 H (CH$_3$-23); 1.195 s, 3 H (CH$_3$-24); 1.68 dd, 1 H (H-7β, J(6,7β) = 5.1, J(7β,7α) = 18.2); 2.19 m, 1 H (H-2, J(2,1α) ~ 12.5, J(2,1β) ~ 4.0, J(2,CH$_3$) ~ 6.7, J(2,3) ~ 2.5); 2.25 bd, 1 H (H-7α, J(7α,7β) = 18.2); 3.16 bs, 1 H (H-3); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 5.53 dd, 1 H (H-6, J(6,7α) = 2.9, J(6,7β) = 5.1). For ^{13}C NMR, see Table II.

Reduction of 19β,28-Epoxy-2,2-dimethyl-18α-olean-5-en-3-one (**4d**)

Method A. To a solution of ketone **4d** (100 mg, 0.21 mmol) in benzene (5 ml), a solution of sodium borohydride (80 mg, 2.1 mmol) in methanol (10 ml) was added and the mixture was left standing at room temperature overnight. Then the mixture was worked up as in the case of the reduction of ketone **4a**, the evaporation residue contained only the starting ketone **4d**.

Method B. To a refluxing solution of ketone **4d** (200 mg, 0.43 mmol) in ether (30 ml), lithium aluminium hydride (60 mg, 1.6 mmol) was added by extracting from Soxhlet extractor during 4 h. The mixture was refluxed for another 3 h and then ethyl acetate, water and 2 M hydrochloric acid were dropwise added. The organic layer was washed with water and sodium hydrogencarbonate and then it was worked up in the usual manner. The evaporation residue (185 mg) was separated by column chromatography on silica gel (25 g) with petroleum ether–ether (3:1) as eluent. The following compounds were obtained.

a) 19β,28-Epoxy-2,2-dimethyl-18α-olean-5-en-3β-ol (**5d**; 85 mg, 42%), m.p. 294–297 °C (benzene–ethanol), $[\alpha]_D$ +63. IR: 1029 (C–O–C); 1652 (C=C); 3631 (OH). ^{1}H NMR: 0.808 s, 3 H (CH$_3$-29); 0.893 s, 3 H (CH$_3$-27); 0.943 s, 3 H (CH$_3$-30); 0.953 s, 3 H (2β-CH$_3$); 1.009 d, 3 H (CH$_3$-26, J = 0.9); 1.134 s, 3 H (2α-CH$_3$); 1.180 bs, 3 H (CH$_3$-23); 1.215 s, 3 H (CH$_3$-24); 1.237 s, 3 H (CH$_3$-25); 1.69 dd, 1 H (H-7β, J(7β,6) = 5.3, J(7β,7α) = 18.0); 1.69 bd, 1 H (OH, J(OH,3) = 6.2); 1.80 d, 1 H (H-1β, J(1β,1α) = 13.6); 2.18 bd, 1 H (H-7α, J(7α,7β) = 18.0); 3.00 bd, 1 H (H-3, J(3,OH) = 6.2); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 5.61 dd, 1 H (H-6, J(6,7α) = 2.8, J(6,7β) = 5.3). For ^{13}C NMR, see Table I. For C$_{32}$H$_{52}$O$_2$ (468.8) calculated: 81.99% C, 11.18% H; found: 82.15% C, 11.01% H.

b) 19β,28-Epoxy-2,2-dimethyl-18α-olean-5-en-3α-ol (**5h**; 45 mg, 22%), m.p. 275–278 °C (benzene–ethanol), $[\alpha]_D$ +78. IR: 1030 (C–O–C); 3630 (OH). ^{1}H NMR: 0.809 s, 3 H (CH$_3$-29); 0.844 s, 3 H (CH$_3$-23); 0.890 s, 3 H (CH$_3$-27); 0.942 s, 3 H (CH$_3$-30); 0.984 d, 3 H (CH$_3$-26,

J = 1.1), 1.012 s, 3 H (CH$_3$-24); 1.138 d, 3 H (CH$_3$-25, J = 0.4); 1.148 s, 3 H (2α-CH$_3$); 1.218 s, 3 H (2β-CH$_3$); 1.28 bd, 1 H (H-1α, J(1α,1β) = 14.0); 1.63 d, 1 H (H-1β, J(1β,1α) = 14.0); 1.68 dd, 1 H (H-7β, J(7β,6) = 5.5, J(7β,7α) = 17.8); 2.16 bd, 1 H (H-7α, J(7α,7β) = 17.8); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 3.80 d, 1 H (H-3, J(3,OH) ~ 6.8); 5.60 dd, 1 H (H-6, J(6,7α) = 2.7, J(6,7β) = 5.5). For C$_{32}$H$_{52}$O$_2$ (468.8) calculated: 81.99% C, 11.18% H; found: 81.87% C, 11.34% H.

Reduction of 19β,28-Epoxy-2β-methyl-18α-olean-5-en-3-one (4c)

Method A. Reduction with sodium borohydride under the conditions described in previous experiment, procedure *A*, gave only traces of the product; starting ketone **4c** (93%) was isolated.

Method B. The reduction of ketone **4c** (80 mg, 0.18 mmol) with lithium aluminium hydride under the conditions described in previous experiment, procedure *B*, afforded a mixture of products. The mixture was separated by preparative TLC on silica gel (10 g), petroleum ether–ether (2:1) was used as eluent. The following compounds were obtained.

a) *19β,28-Epoxy-2β-methyl-18α-olean-5-en-3β-ol* (**5c**; 55 mg, 68%), m.p. 281–282 °C (chloroform–methanol, around 240 °C modification change), [α]$_D$ +70. IR: 1030 (C–O–C); 3632 (OH). ¹H NMR: 0.808 s, 3 H (CH$_3$-29); 0.905 s, 3 H (CH$_3$-27); 0.940 s, 3 H (CH$_3$-30); 0.950 d, 3 H (2β-CH$_3$, J = 6.2); 0.968 d, 3 H (CH$_3$-26, J ~ 1); 1.111 s, 3 H (CH$_3$-23); 1.203 s, 3 H (CH$_3$-24); 1.278 s, 3 H (CH$_3$-25); 1.63 dd, 1 H (H-7β, J(6,7β) = 5.4, J(7β,7α) = 17.8); 2.16 bd, 1 H (H-7α, J(7α,7β) = 17.8); 3.39 bs, 1 H (H-3); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 5.53 dd, 1 H (H-6, J(6,7α) = 2.7, J(6,7β) = 5.4). For ¹³C NMR, see Table I. For C$_{31}$H$_{50}$O$_2$ (454.7) calculated: 81.88% C, 11.08% H; found: 82.12% C, 10.89% H.

b) *19β,28-Epoxy-2β-methyl-18α-olean-5-en-3α-ol* (**5g**; 5 mg, 6%), m.p. 204–205 °C (chloroform–ethanol). ¹H NMR: 0.808 s, 3 H (CH$_3$-29); 0.894 s, 3 H (CH$_3$-27); 0.941 s, 3 H (CH$_3$-30); 0.974 d, 3 H (CH$_3$-26, J = 1.1); 1.036 d, 3 H (2β-CH$_3$, J = 6.2); 1.039 s, 3 H (CH$_3$-23); 1.099 s, 3 H (CH$_3$-24); 1.204 s, 3 H (CH$_3$-25); 1.67 dd, 1 H (H-7β, J(7β,6) = 5.4, J(7β,7α) = 18.0); 2.15 bd, 1 H (H-7α, J(7α,7β) = 18.0); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.80 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.56 s, 1 H (H-19); 3.75 d, 1 H (H-3, J(2,3) ~ 11.5); 5.56 dd, 1 H (H-6, J(6,7α) = 2.7, J(6,7β) = 5.4).

2α-Bromo-19β,28-epoxy-18α-olean-5-en-3-one (6a) and
2β-Bromo-19β,28-epoxy-18α-olean-5-en-3-one (6b)

a) To a solution of ketone **4a** (100 mg, 0.23 mmol) in acetic acid (10 ml), one drop of 46% hydrobromic acid was added and then a solution of bromine (37 mg, 0.23 mmol) in acetic acid (10 ml) was added during 15 min while stirring. Then the mixture was left standing for 15 min, then diluted with water and extracted with chloroform. The chloroform layer was washed with a solution of sodium hydrogencarbonate and then worked up in the usual manner. The evaporation residue (110 mg) gave 2α-bromo ketone **6a** (85 mg, 72%) after crystallisation from chloroform–heptane, m.p. 220 °C. According to ¹H NMR the product contained 85–90% of 2α-bromo ketone **6a** and 10–15% of 2β-bromo ketone **6b**. After preparative TLC (petroleum ether–ether 4:1) and multiple crystallisation from chloroform–methanol and from ether pure 2α-bromo derivate **6a** was obtained, m.p. 268–274 °C, [α]$_D$ –20. IR: 1035 (C–O–C); 1675 (C=C); 1720 (C=O). ¹H NMR: 0.817 s, 3 H (CH$_3$-29); 0.852 s,

3 H (CH$_3$-25); 0.925 s 3 H (CH$_3$-27); 0.944 s, 3 H (CH$_3$-30); 0.998 bs, 3 H (CH$_3$-26); 1.301 s, 3 H (CH$_3$-24); 1.494 s, 3 H (CH$_3$-23); 1.76 dd, 1 H (H-7β, J(7β,6) = 5.7, J(7β,7α) = 17.7); 1.97 bdd, 1 H (H-1α, J(1α,1β) = 13.4, J(1α,2) = 11.5); 2.17 bd, 1 H (H-7α, J(7α,7β) = 17.7, J(7α,6) = 2.4); 2.65 dd, 1 H (H-1β, J(1β,1α) = 13.4, J(1β,2) = 8.3); 3.46 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.79 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.54 s, 1 H (H-19); 4.37 dd, 1 H (H-2, J(2,1α) = 11.5, J(2,1β) = 8.3); 5.60 dd, 1 H (H-6, J(6,7α) = 2.4, J(6,7β) = 5.7). For ^{13}C NMR, see Table II. ^{1}H NMR of minor isomer **6b**: 1.75 dd, 1 H (H-7β, J(7β,6) = 6.0, J(7β,7α) = 17.6); 2.41 dd, 1 H (H-1β, J(1β,1α) = 14.3, J(1β,2) = 6.0); 2.54 bdd, 1 H (H-1α, J(1α,1β) = 14.3, J(1α,2) = 10.5); 4.96 dd, 1 H (H-2, J(2,1α) = 10.5, J(2,1β) = 6.0); 5.67 dd, 1 H (H-6, J(6,7α) = 2.3, J(6,7β) = 6.0); other signals were overlapped with the spectrum of major 2α-bromo derivate **6a**. For C$_{30}$H$_{45}$BrO$_2$ (517.6) calculated: 69.61% C, 8.76% H; found: 69.40% C, 8.87% H.

b) To a solution of enol acetate **6c** (109 mg, 0.23 mmol) and sodium acetate (22 mg) in acetic acid (15 ml), a solution of bromine (36 mg, 0.23 mmol) in acetic acid (1 ml) was added dropwise while stirring. The mixture was diluted with water and extracted with chloroform. The chloroform layer was washed with a solution of sodium hydrogencarbonate and then worked up in the usual manner. The evaporation residue (104 mg, 88%) gave 2α-bromo derivative **6a** (identical with the product from procedure *a*) after crystallisation from chloroform–petroleum ether.

c) Bromohydrin **6e** (21 mg, 0.04 mmol), sodium acetate trihydrate (100 mg) and sodium dichromate (300 mg, 1.15 mmol) were dissolved in acetic acid (100 ml). The mixture was stirred for 6.5 h and then it was left standing for 11 days. Then methanol (1 ml) was added and the mixture was diluted with water and extracted (2×) with ether. The ether layer was washed with a solution of sodium hydrogencarbonate and then worked up in the usual manner. The evaporation residue (20 mg) gave bromo ketone **6a** (13.9 mg, 66%) after crystallisation from chloroform–petroleum ether.

19β,28-Epoxy-18α-oleane-2,5-dien-3-yl Acetate (**6c**)

A mixture of ketone **4a** (290 mg, 0.66 mmol) and of 4-toluenesulfonic acid monohydrate (40 mg, 0.23 mmol) was refluxed for 2 h with isopropenyl acetate (10 ml). During another 6 h, 6 ml of the solvent was distilled off. From the distillation residue crude enol acetate **6c** (270 mg) crystallised. The product was chromatographed on a column of silica gel (20 g) with benzene as eluent. Enol acetate **6c** (227 mg, 71%) was obtained, m.p. 271–275 °C (chloroform–petroleum ether), [α]$_D$ +71. IR: 1036, 1097 (C–O–C); 1700 (C=C); 1762 (CH$_3$COO). For C$_{32}$H$_{48}$O$_3$ (480.7) calculated: 79.95% C, 10.06% H; found: 79.81% C, 10.24% H.

Reduction of 2α-Bromo-19β,28-epoxy-18α-olean-5-en-3-one (**6a**)

To a solution of bromo ketone **6a** (390 mg, 0.75 mmol) and boric acid (1.2 g) in ethanol (250 ml), sodium borohydride (600 mg, 15.9 mmol) was added during 5 min. The mixture was stirred for 2 h and then it was allowed to stand for 13 h. Then ammonium chloride (6 g) in water (50 ml) was added and the mixture was stirred for 90 min. Then the mixture was diluted with water (1000 ml) and extracted with chloroform. The chloroform layer was worked up in the usual manner. The evaporation residue gave after column chromatography on silica gel (25 g; eluent benzene).

a) 2α-Bromo-19β,28-epoxy-18α-olean-5-en-3α-ol (**6d**; 41.5 mg, 10%), m.p. 237–243 °C (chloroform–petroleum ether), [α]$_D$ +50. IR: 1035 (C–O–C); 3588 (OH). ^{1}H NMR: 0.808 s, 3 H (CH$_3$-29); 0.899 s, 3 H (CH$_3$-27); 0.939 s, 3 H (CH$_3$-30); 0.966 d, 3 H (CH$_3$-26, J = 0.9); 1.161 d, 3 H (CH$_3$-25, J = 0.7); 1.234 s, 6 H (CH$_3$-23,24); 1.69 dd, 1 H (H-7β, J(7β,6) = 5.0, J(7β,7α) = 18.5); 1.92 bt, 1 H (H-1α, J(1α,1β) = 12.5, J(1α,2) = 13.0); 2.04 d, 1 H (OH, J(OH,3) = 3.1); 2.13 bdd, 1 H (H-1β, J(1β,1α) = 12.5, J(1β,2) = 4.4); 2.29 bd, 1 H (H-7α, J(7α,7β) = 18.5); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.79 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 3.62 bs, 1 H (H-3); 4.94 ddd, 1 H (H-2, J(2,1α) = 13.0, J(2,1β) = 4.4, J(2,3) = 2.1); 5.57 dd, 1 H (H-6, J(6,7α) = 2.9, J(6,7β) = 5.0). For ^{13}C NMR, see Table II. EI-MS, *m/z* (%): 520 (3, M$^+$), 518 (3, M$^+$), 505 (34), 503 (34), 487 (28), 485 (28), 449 (5), 447 (5), 438 (60), 315 (12), 203 (66), 191 (94), 124 (100). For C$_{30}$H$_{47}$BrO$_2$ (519.6) calculated: 69.34% C, 9.12% H; found: 69.03% C, 8.88% H.

b) 2α-Bromo-19β,28-epoxy-18α-olean-5-en-3β-ol (**6e**), m.p. 272–276 °C (chloroform–methanol), [α]$_D$ +46. IR: 1036 (C–O–C); 3410 and 3596 (OH). ^{1}H NMR: 0.812 s, 3 H (CH$_3$-29); 0.897 s, 3 H (CH$_3$-27); 0.943 s, 3 H (CH$_3$-30); 0.973 d, 3 H (CH$_3$-26, J = 0.8); 1.134 s, 3 H (CH$_3$-24); 1.141 d, 3 H (CH$_3$-25, J = 0.6); 1.259 s, 3 H (CH$_3$-23); 1.60 bt, 1 H (H-1α, J(1α,1β) = 12.9, J(1α,2) = 12.8); 1.72 dd, 1 H (H-7β, J(7β,6) = 5.3, J(7β,7α) = 18.2); 2.21 bd, 1 H (H-7α, J(7α,7β) = 18.2); 2.47 dd, 1 H (H-1β, J(1β,1α) = 12.9, J(1β,2) = 4.2); 2.50 d, 1 H (OH, J(OH,3) = 2.5); 3.26 dd, 1 H (H-3, J(2,3) = 10.3, J(3,OH) = 2.5); 3.46 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.79 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.53 s, 1 H (H-19); 4.53 ddd, 1 H (H-2, J(2,1α) = 12.8, J(2,1β) = 4.2, J(2,3) = 10.3); 5.64 dd, 1 H (H-6, J(6,7α) = 2.8, J(6,7β) = 5.3). For ^{13}C NMR, see Table II. EI-MS, *m/z* (%): 520 (38, M$^+$), 518 (38, M$^+$), 505 (24), 503 (24), 487 (31), 485 (32), 449 (50), 447 (50), 438 (90), 405 (55), 382 (100), 381 (98), 367 (40), 363 (44), 191 (82). For C$_{30}$H$_{47}$BrO$_2$ (519.6) calculated: 69.34% C, 9.12% H; found: 69.08% C, 9.30% H.

2β,3β,19β,28-Diepoxy-18α-olean-5-ene (**6f**)

A solution of bromohydrin **6e** (145 mg, 0.28 mmol) and potassium hydroxide (800 mg) in ethanol (80 ml) was refluxed for 4 h and then it was left standing at room temperature overnight. Then the mixture was diluted with water and extracted with chloroform. The chloroform layer was worked up in the usual manner. Epoxide **6f** (113 mg, 92%) was obtained, m.p. 261–263 °C (chloroform–petroleum ether), [α]$_D$ +72. IR: 1035 (C–O–C). ^{1}H NMR: 0.806 s, 3 H (CH$_3$-29); 0.882 s, 3 H (CH$_3$-27); 0.940 s, 3 H (CH$_3$-30); 0.965 d, 3 H (CH$_3$-26, J = 1.0); 1.229 d, 3 H (CH$_3$-25, J = 0.7); 1.246 s, 3 H (CH$_3$-23); 1.307 s, 3 H (CH$_3$-24); 1.65 dd, 1 H (H-7β, J(7β,7α) = 18.0, J(7β,6) = 5.5); 2.14 bd, 1 H (H-7α, J(7α,7β) = 18.0); 2.39 dd, 1 H (H-1β, J(1β,1α) = 14.8, J(1β,2) = 2.4); 2.89 d, 1 H (H-3, J(3,2) = 4.1); 3.26 m, 1 H (H-2, J(2,3) = 4.1, J(2,1β) = 2.4, J(2,1α) = 1.7); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.9); 3.79 bd, 1 H (H-28b, J(28b,28a) = 7.9); 3.53 s, 1 H (H-19); 5.59 dd, 1 H (H-6, J(6,7α) = 2.7, J(6,7β) = 5.5). For ^{13}C NMR, see Table II. EI-MS, *m/z* (%): 438 (11, M$^+$), 423 (6), 420 (4), 407 (5), 405 (4), 395 (5), 394 (7), 367 (7), 133 (67), 121 (75), 119 (100), 107 (70), 105 (66). For C$_{30}$H$_{46}$O$_2$ (438.7) calculated: 82.13% C, 10.57% H; found: 81.90% C, 10.64% H.

Ketone **4a** from Bromohydrin **6d**

Bromohydrin **6d** (30 mg, 0.06 mmol) was refluxed for 2 h with 3% methanolic solution of potassium hydroxide (25 ml). After cooling to the room temperature, the reaction mixture

was diluted with water and extracted with ether. The ether layer was worked up in the usual manner. Ketone **4a** (19 mg, 74%) was obtained after crystallisation from methanol.

2α-[^{2}H]-19β,28-Epoxy-18α-olean-5-en-3β-ol (6g)

Epoxide **6f** (51 mg, 0.12 mmol) was dissolved in a mixture of dried benzene (5 ml) and dried ether (30 ml) and lithium aluminium deuteride (100 mg, 2.3 mmol) was added. The mixture was refluxed for 8 h under dry nitrogen and then it was left standing at the room temperature under nitrogen overnight. Then ethyl acetate and hydrochloric acid were added and the mixture was extracted with ether. The ether layer was worked up in the usual manner. The evaporation residue (50 mg) was purified by preparative TLC on silica gel with benzene–ether (4:1) as eluent. Deuterated alcohol **6g** (34 mg, 66%) was obtained. M.p. 272–275 °C (ether).

2α-[^{2}H]-19β,28-Epoxy-18α-olean-5-en-3-one (6h)

To a solution of deuterated alcohol **6g** (34 mg, 0.08 mmol) and sodium acetate trihydrate (100 mg) in acetic acid (5 ml), sodium dichromate (100 mg, 0.38 mmol) was added and the mixture was stirred for 6.5 h. Then methanol (1 ml) was added and the mixture was diluted with water and extracted with ether. The ether layer was washed with water and with saturated solution of sodium hydrogencarbonate and then worked up in the usual manner. Deuterated ketone **6h** (30 mg, 88%) was obtained, m.p. 229–235 °C (ether–petroleum ether). ^{1}H NMR: 0.814 s, 3 H (CH$_3$-29); 0.847 bs, 3 H (CH$_3$-25); 0.929 s, 3 H (CH$_3$-27); 0.945 s, 3 H (CH$_3$-30); 1.000 d, 3 H (CH$_3$-26, J = 0.9); 1.227 s, 3 H (CH$_3$-24); 1.247 s, 3 H (CH$_3$-23); 1.57 dd, 1 H (H-1α, $J(1α,1β)$ = 14.0, $J(1α,2β)$ = 10.5); 2.00 dd, 1 H (H-1β, $J(1β,1α)$ = 14.0, $J(1β,2β)$ = 9.1); 2.42 bt, 1 H (H-2β, $J(2β,2α)$ = 13.3, $J(2β,1α)$ = 10.5, $J(2β,1β)$ = 9.1); 1.74 dd, 1 H (H-7β, $J(7β,6)$ = 5.8, $J(7β,7α)$ = 17.4); 2.16 bd, 1 H (H-7α, $J(7α,6)$ = 2.4, $J(7α,7β)$ = 17.4); 3.46 d, 1 H (H-28a, $J(28a,28b)$ = 7.8); 3.80 bd, 1 H (H-28b, $J(28b,28a)$ = 7.8); 3.54 s, 1 H (H-19); 5.54 dd, 1 H (H-6, $J(6,7α)$ = 2.4, $J(6,7β)$ = 5.8).

5α,6α,19β,28-Diepoxy-18α-oleanan-3β-yl Acetate (7a)

A solution of acetyl derivative **5e** (170 mg, 0.35 mmol) and 3-chloroperoxybenzoic acid (60%, 120 mg, 0.42 mmol) in chloroform (5 ml) was left standing at room temperature for 2 h. Then the mixture was diluted with a solution of sodium hydrogencarbonate and extracted with chloroform. The chloroform layer was worked up in the usual manner. Epoxy derivative **7a** (165 mg, 94%) was obtained, m.p. 291–295 °C (chloroform–octane), [α]$_D$ +34. IR: 1037 (C–O–C); 1265 (CH$_3$COO); 1666 (C=C); 1734 (CH$_3$COO). ^{1}H NMR: 0.754 s, 3 H (CH$_3$-23); 0.791 s, 3 H (CH$_3$-29); 0.924 s, 6 H (CH$_3$-27,30); 1.027 s, 3 H (CH$_3$-26); 1.150 s, 3 H (CH$_3$-25); 1.163 s, 3 H (CH$_3$-24); 1.43 dd, 1 H (H-7β, $J(7β,7α)$ = 16.5, $J(7β,6)$ = 3.0); 2.04 s, 3 H (CH$_3$COO); 2.26 bd, 1 H (H-7α, $J(7α,7β)$ = 16.5); 3.15 dd, 1 H (H-6, $J(6,7α)$ = 1.7, $J(6,7β)$ = 3.0); 3.44 d, 1 H (H-28a, $J(28a,28b)$ = 7.8); 3.75 bd, 1 H (H-28b, $J(28b,28a)$ = 7.8); 3.51 s, 1 H (H-19); 4.77 m, 1 H (H-3, ΣJ = 16.6). For ^{13}C NMR, see Table II. EI-MS, m/z (%): 498 (4, M$^+$), 480 (2), 438 (50), 427 (4), 423 (9), 420 (15), 407 (5), 405 (9), 395 (11), 367 (6), 317 (17), 290 (17), 288 (34), 275 (27), 273 (63), 257 (35), 217 (34), 206 (66), 203 (100), 187 (79), 185 (68), 175 (79). For C$_{30}$H$_{50}$O$_4$ (498.7) calculated: 77.06% C, 10.11% H; found: 76.82% C, 10.25% H.

5α,6α,19β,28-Diepoxy-18α-oleanan-3β-ol (**7b**)

A solution of alcohol **5a** (120 mg, 0.27 mmol) and 3-chloroperoxybenzoic acid (60%, 90 mg, 0.31 mmol) in chloroform (5 ml) was left standing at room temperature for 2 h. Then the mixture was diluted with a solution of sodium hydrogencarbonate and extracted with chloroform. The chloroform layer was worked up in the usual manner. Epoxy derivative **7b** (90 mg, 72%) was obtained, m.p. 288–290 °C (chloroform–benzene), $[\alpha]_D$ +30. IR: 1037 (C–O–C); 3630 (OH). ^{1}H NMR: 0.789 s, 3 H (CH$_3$-29); 0.875 s, 3 H (CH$_3$-23); 0.921 s, 6 H (CH$_3$-27,30); 1.025 s, 3 H (CH$_3$-26); 1.089 s, 3 H (CH$_3$-24); 1.125 s, 3 H (CH$_3$-25); 1.43 dd, 1 H (H-7β, J(7β,7α) = 16.4, J(7β,6) = 3.0); 2.25 bd, 1 H (H-7α, J(7α,7β) = 16.4); 3.15 dd, 1 H (H-6, J(6,7α) = 1.8, J(6,7β) = 3.0); 3.46 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.75 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.51 s, 1 H (H-19); 3.54 m, 1 H (H-3, ΣJ = 16.4). For ^{13}C NMR, see Table II. EI-MS, *m/z* (%): 456 (5, M$^+$), 438 (40), 423 (15), 420 (9), 413 (3), 407 (6), 405 (6), 395 (25), 385 (14), 367 (9), 290 (45), 288 (67), 275 (29), 273 (76), 257 (55), 217 (27), 206 (51), 203 (100). For C$_{30}$H$_{48}$O$_3$ (456.7) calculated: 78.90% C, 10.59% H; found: 78.75% C, 10.41% H.

5α,6α,19β,28-Diepoxy-18α-oleanan-3-one (**7c**)

To a solution of hydroxyepoxide **7b** (53.4 mg, 0.12 mmol) and sodium acetate (40 mg) in acetic acid (11 ml), a solution of sodium dichromate dihydrate (100 mg, 0.34 mmol) in acetic acid (4 ml) was added under stirring and the mixture was left standing at room temperature for 26 h. Then the mixture was diluted with water and extracted with chloroform. The chloroform layer was washed with water and with a sodium hydrogencarbonate solution and worked up in the usual manner. Ketone **7c** (46.2 mg, 87%) was obtained, m.p. 259–263 °C (chloroform–petroleum ether), $[\alpha]_D$ +106. IR: 1037 (C–O–C); 1720 (C=O). ^{1}H NMR: 0.796 s, 3 H (CH$_3$-29); 0.880 s, 3 H (CH$_3$-23); 0.924 s, 3 H (CH$_3$-27); 0.930 s, 3 H (CH$_3$-30); 1.011 d, 3 H (CH$_3$-25, J = 0.7); 1.021 d, 3 H (CH$_3$-26, J = 0.85); 1.260 s, 3 H (CH$_3$-24); 1.55 dd, 1 H (H-7β, J(6,7β) = 4.2, J(7β,7α) = 16.0); 1.84 m, 1 H (H-1β); 1.93 m, 1 H (H-1α, J(1α,1β) = 12.8, J(1α,2α) = 11.6, J(1α,2β) = 4.2); 2.37 ddd, 1 H (H-2β, J(2β,1α) = 4.2, J(2β,1β) = 9.9, J(2β,2α) = 16.2); 2.77 ddd, 1 H (H-2α, J(2α,1α) = 11.6, J(2α,1β) = 5.2, J(2α,2β) = 16.2); 2.15 bd, 1 H (H-7α, J(7α,7β) = 16.0); 3.12 bd, 1 H (H-6, J(6,7α) ~ 0, J(6,7β) = 4.2); 3.45 d, 1 H (H-28a, J(28a,28b) = 7.8); 3.76 bd, 1 H (H-28b, J(28b,28a) = 7.8); 3.50 s, 1 H (H-19). For ^{13}C NMR, see Table II. EI-MS, *m/z* (%): 454 (28, M$^+$), 439 (34), 436 (22), 426 (22), 421 (18), 411 (8), 383 (31), 290 (15), 288 (17), 275 (18), 273 (34), 257 (21), 233 (45), 219 (67), 217 (44), 203 (69), 201 (100). For C$_{30}$H$_{46}$O$_3$ (454.7) calculated: 78.90% C, 10.59% H; found: 78.75% C, 10.41% H.

19β,28-Epoxy-7-oxo-18α-olean-5-en-3β-yl Acetate (**8a**)

A mixture of acetate **5e** (100 mg, 0.21 mmol) and *N*-bromosuccinimide (60 mg, 0.34 mmol) in tetrachloromethane (10 ml) was refluxed for 15 min with a 500 W photobulb. Then it was cooled to the room temperature, the arisen succinimide was filtered off and ethyl acetate (10 ml), methanol (10 ml), benzene (10 ml) and aluminium oxide (2 g) were added to the filtrate. After 2-h shaking aluminium oxide was filtered off and the solvents were distilled off. The evaporation residue was dissolved in acetic acid (20 ml) and the Jones reagent (0.2 ml) was added with stirring. After 5 min methanol (1 ml) was added to remove the reagent and the mixture was diluted with water and extracted with benzene. The organic layer was worked up in the usual manner. The evaporation residue (84.5 mg) was purified by pre-

parative TLC on silica gel (50 g, 50 × 20 cm) with benzene–ether (5:1) as eluent. Acetoxy ketone **8a** (32 mg, 31%) was obtained, m.p. 303 °C (chloroform–methanol), $[\alpha]_D$ –20. IR: 1038 (C–O–C); 1622, 1665 (C=C–C=O); 1738 (CH_3COO). UV: 231 nm (ε 15 000). ^1H NMR: 0.797 s, 3 H (CH_3-29); 0.908 s, 3 H (CH_3-27); 0.935 s, 3 H (CH_3-30); 1.113 bs, 3 H (CH_3-23); 1.181 s, 3 H (CH_3-25); 1.205 s, 3 H (CH_3-24); 1.221 bs, 3 H (CH_3-26); 2.08 s, 3 H (CH_3COO); 3.47 d, 1 H (H-28a, $J(28a,28b)$ = 7.9); 3.82 bd, 1 H (H-28b, $J(28b,28a)$ = 7.9); 3.56 s, 1 H (H-19); 4.60 m, 1 H (H-3, ΣJ = 16.3); 5.91 s, 1 H (H-6). For ^{13}C NMR, see Table II. EI-MS, m/z (%): 496 (18, M^+), 481 (2), 465 (2), 436 (55), 425 (6), 421 (43), 406 (11), 405 (5), 403 (7), 393 (3), 289 (40), 263 (51), 261 (34), 229 (25), 217 (24), 203 (100), 201 (30). For $C_{32}H_{48}O_4$ (496.7) calculated: 77.38% C, 9.74% H; found: 77.45% C, 9.58% H.

19β,28-Epoxy-18α-olean-5-en-3,7-dione (**8b**)

A mixture of acetoxy ketone **8a** (110 mg, 0.22 mmol) and potassium hydroxide (150 mg, 2.7 mmol) was refluxed in methanol (10 ml) for 2.5 h. Then water (20 ml) was added and the mixture was extracted with chloroform (4 × 5 ml). The combined organic layers were worked up in the usual manner. The evaporation residue (78 mg, 77%) was used for oxidation without purification. Sodium dichromate dihydrate (120 mg, 0.28 mmol) was added to the stirred mixture of crude 3β-hydroxy-19β,28-epoxy-18α-olean-5-en-7-one (68 mg) and sodium acetate (50 mg) in acetic acid (15 ml). The reaction mixture was stirred for 20 h, then water (20 ml) was added and the mixture was extracted with chloroform (3 × 10 ml). The combined organic layers were worked up in the usual manner. The evaporation residue was purified by preparative TLC on silica gel (50 g, 50 × 20 cm) with petroleum ether–ether (3:1) as eluent. Diketone **8b** (13 mg, 19%) was obtained, m.p. 215–228 °C (chloroform–methanol), $[\alpha]_D$ +36.5. IR: 1031 (C–O–C); 1634 (C=C); 1658 (7-oxo); 1712 (3-oxo). UV: 234 nm (ε 11 170). ^1H NMR: 0.804 s, 3 H (CH_3-29); 0.943 s, 3 H (CH_3-30); 1.007 s, 3 H (CH_3); 1.038 bs, 3 H (CH_3); 1.253 s, 3 H (CH_3); 1.311 s, 3 H (CH_3); 1.322 s, 3 H (CH_3); 1.67 ddd, 1 H (H-1α, $J(1\alpha,1\beta)$ = 13.9, $J(1\alpha,2\alpha)$ = 8.9, $J(1\alpha,2\beta)$ = 10.9); 2.20 ddd, 1 H (H-1β, $J(1\beta,1\alpha)$ = 13.9, $J(1\beta,2\alpha)$ = 1.9, $J(1\beta,2\beta)$ = 8.2); 2.57 ddd, 1 H (H-2β, $J(2\beta,1\alpha)$ = 10.9, $J(2\beta,1\beta)$ = 8.2, $J(2\beta,2\alpha)$ = 19.1); 2.62 ddd, 1 H (H-2α, $J(2\alpha,1\alpha)$ = 8.9, $J(2\alpha,1\beta)$ = 1.9, $J(2\alpha,2\beta)$ = 19.1); 3.49 d, 1 H (H-28a, $J(28a,28b)$ = 7.9); 3.82 bd, 1 H (H-28b, $J(28b,28a)$ = 7.9); 3.56 s, 1 H (H-19); 5.83 s, 1 H (H-6). For ^{13}C NMR, see Table II. EI-MS, m/z (%): 452 (39, M^+), 437 (18), 422 (4), 421 (4), 419 (4), 407 (18), 381 (6), 219 (94), 135 (50), 43 (100). For $C_{30}H_{44}O_3$ (452.7) calculated: 79.52% C, 9.72% H; found: 79.63% C, 9.87% H.

REFERENCES

1. Tišlerová I., Richtr V., Klinot J.: *Collect. Czech. Chem. Commun.* **2006**, *71*, 368.
2. Kim D. S. L., Pezzuto J. M., Pisha E.: *Bioorg. Med. Chem. Lett.* **1998**, *8*, 1707.
3. a) Sun I. C., Shen J. K., Wang H. K., Cosentino L. M., Lee K. H.: *Bioorg. Med. Chem. Lett.* **1998**, *8*, 1267; b) Lee S. S., Chen W. C., Huang C. F., Su Y.: *J. Nat. Prod.* **1998**, *61*, 1343.

4. Sun I. C., Wang H. K., Kashiwada Y., Shen J. K., Cosentino L. M., Chen C. H., Yang L. M., Lee K. H.: *J. Med. Chem.* **1998**, *41*, 4648.

5. Ryu S. Y., Choi S. U., Lee S. H., Lee C. O., Zaesung N., Ahn J. W.: *Arch. Pharm. Res.* **1994**, *17*, 375.

6. Fujioka T., Kashiwada Y., Kilkuskie R. E., Cosentino L. M., Ballos L. M., Jiang J. B., Janyen W. P., Chen I. S., Lee K. H.: *J. Nat. Prod.* **1994**, *57*, 243.

7. Soler F., Poujade Ch., Evers M., Carry J.-Ch., Hénin Y., Bousseau A., Huet T., Pauwels R., De Clercq E., Mayaux J.-F., Le Pecq J.-B., Dereu N.: *J. Med. Chem.* **1996**, *39*, 1069.

8. Ngassapa O. D., Soejarto D. D., Che Ch. T., Pezzuto J. M., Farnsworth N. R.: *J. Nat. Prod.* **1991**, *54*, 1353.

9. Sturm S., Gil R. R., Chai H.-B., Ngassapa O. D., Santisuk T., Reutrakul V., Howe A., Moss M., Besterman J. M., Yang S.-L., Farthing J. E., Tait R. M., Lewis J. A., O'Neill M. J., Farnsworth N. R., Cordell G. A., Pezzuto J. M., Kinghorn A. D.: *J. Nat. Prod.* **1996**, *59*, 658.

10. Rice G. K., Yokoi T., Hayashi T., Suzuki H., Lee K.-H., McPhail A. T.: *J. Chem. Soc., Chem. Commun.* **1986**, 1397.

11. Šarek J., Klinot J., Džubák P., Klinotová E., Nosková V., Křeček V., Kořínková G., Thomson J. O., Janošťáková A., Wang S., Parsons S., Fischer P. M., Zhelev N. Z., Hajdúch M.: *J. Med. Chem.* **2003**, *46*, 5402.

12. Kouzi S. A., Chatterjee P., Pezzuto J. M., Hamann M. T.: *J. Nat. Prod.* **2000**, *63*, 1653.

13. Hisham A., Kumar G. J., Fujimoto Y., Hara N.: *Phytochemistry* **1996**, *42*, 789.

14. Facundo V. A., Andrade C. H. S., Silveria E. R., Braz-Filho R., Hufford C. D.: *Phytochemistry* **1993**, *32*, 411.

15. Adnyana I. K., Tezuka Y., Awale S., Banskota A. H., Tran K. Q., Kadota S.: *Chem. Pharm. Bull.* **2000**, *48*, 1114.

16. Adnyana I. K., Tezuka Y., Banskota A. H., Tran K. Q., Kadota S.: *J. Nat. Prod.* **2001**, *64*, 360.

17. Jiang Z.-H., Tanaka T., Kouno I.: *Phytochemistry* **1995**, *40*, 1223.

18. Sejbal J., Klinot J., Buděšínský M.: *Collect. Czech. Chem. Commun.* **1996**, *61*, 1371.

19. Sejbal J., Klinot J., Buděšínský M., Protiva J.: *Collect. Czech. Chem. Commun.* **1997**, *62*, 1905.

20. Klinot J., Hořejší M., Buděšínský M., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1975**, *40*, 3712.

21. Kirk D. N., Hartshorn M. P.: S*teroid Reaction Mechanisms*, p. 168. Elsevier, Amsterdam 1968.

22. Woodward R. B., Patchett A. A., Barton D. H. R., Ives D. A. J., Kelly R. B.: *J. Am. Chem. Soc.* **1954**, *76*, 2852.

23. Mauli R., Ringold H. J., Djerassi C.: *J. Am. Chem. Soc.* **1960**, *82*, 5494.

24. Baran J. S., Langford D. D., Liang C. D., Pitzele B. S.: *J. Med. Chem.* **1974**, *17*, 184.

25. Klinot J., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1962**, *27*, 377.

26. Schulze H., Pieroh K.: *Ber. Dtsch. Chem. Ges.* **1922**, *55*, 2332.

27. Vystrčil A., Klinot J.: *Collect. Czech. Chem. Commun.* **1959**, *24*, 3273.

28. Lawrie W., Mc Lean J., Taylor G. R.: *J. Chem. Soc.* **1960**, 4303.

29. Atwater N. W.: *J. Am. Chem. Soc.* **1960**, *82*, 2847.

30. Lablache-Combier A., Lacoume B., Levisalles J.: *Bull. Soc. Chim. Fr.* **1965**, 2595.

31. Klinot J., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1966**, *31*, 1079.

32. Klinot J., Kliment M., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1974**, *39*, 3357.

33. Midgley J. M., Parkin J. E., Whalley W. B.: *J. Chem. Soc., Perkin Trans. 1* **1977**, 834.

34. Ringold H. J.: *J. Am. Chem. Soc.* **1960**, *82*, 961.

35. Lablache-Combier A., Levisalles J., Pete J. P., Rudler H.: *Bull. Soc. Chim. Fr.* **1963**, 1689.

36. Klinot J., Světlý J., Kudláčková D., Buděšínský M., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1979**, *44*, 211.

37. Tsuda Y., Kiuchi F.: *Chem. Pharm. Bull.* **1984**, *32*, 4806.

38. Abraham R. J., Holker J. S. E.: *J. Chem. Soc.* **1963**, 806.

39. Burkert U., Allinger N. L.: *Tetrahedron* **1978**, *34*, 807.

40. Lacoume B., Levisalles J.: *Bull. Soc. Chim. Fr.* **1965**, 2602.

41. Klinot J., Richtr V., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1975**, *40*, 1758.

42. Klinot J., Buděšínský M., Klinotová E., Vystrčil A.: *Collect. Czech. Chem. Commun.* **1986**, *51*, 593.

43. Abraham R. J., Holker J. S. E.: *J. Chem. Soc.* **1963**, 806.

44. Ferguson G., Marsh W. C., Midgley J. M., Whalley W. B.: *J. Chem. Soc., Perkin Trans. 2* **1978**, 272.

45. Jones R. N., Ramsay D. H., Herling F., Dobriner K.: *J. Am. Chem. Soc.* **1952**, *74*, 2828.