MINOR SESQUITERPENIC LACTONES FROM *Ursinia anthemoides* (L.) POIRET*

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The authors isolated from the aerial parts of *Ursinia anthemoides* (L.) POIRET another five sesquiterpenic lactones, *IV*, *V*, *VII*, *IX* and *X* and have derived for them their structures and configurations mainly on the basis of ^{1}H NMR spectroscopy and CD measurements; for compounds *IV* and *V* absolute configurations have also been proposed.

In connection with the preceding communications[1,2], concerning the isolation of the main sesquiterpenic lactones of *Ursinia anthemoides* (L.) POIRET and the derivation of their structures *I*–*III*, which were further confirmed[3] by X-ray structural analysis of ursiniolide A (*I*), we investigated the minor sesquiterpenic lactones of the mentioned species.

For their isolation from a relatively very rich mixture of components we made use of high-performance liquid chromatography. We thus isolated compound *IV* ($C_{22}H_{30}O_7$) which according to its physical constants and a comparison of IR and ^{1}H NMR spectra (Table I) was identical with 3-deacetoxyursiniolide B (*IV*) isolated from *U. anethoides* (DC) N.E.BR. (ref.[5]). We derived the absolute configuration of this compound on the basis of its CD spectrum. The value $\Delta\varepsilon = +1{\cdot}4$ at 267 nm, corresponding to a $n\rightarrow\pi^*$ transition of an α,β-unsaturated lactone, characterizes according to Geissman and coworkers[6,7] the absolute configuration of this group when the relative configuration of the α-exomethylene-γ-lactone group is known. Hence, the absolute configuration of 3-deacetylursiniolide B corresponds to formula *IV*. The absolute configuration of the transannularly interacting system of endocyclic double bonds of the discussed lactone is also confirmed by the $\pi\rightarrow\pi^*$ transition of the mentioned system of the double bonds (216 nm with $\Delta\varepsilon = -47{\cdot}7$), which

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is in good agreement with the similar values of the CD spectrum of ursiniolide A (*I*), the absolute configuration of which has been described[1,2]. This fact too indicates the absolute configuration of 3-deacetoxyursiniolide B, as represented by formula *IV*.

The next isolated compound *V*, to which we gave the name ursiniolide D, had the composition $C_{20}H_{28}O_6$. Its IR spectrum indicated the presence of a γ-lactone and ester grouping (intensive band at 1 748 cm^{-1}), a hydroxyl (3 555 cm^{-1}) and a double bond (1 661 cm^{-1}). The mass spectrum showed the molecular peak at *m/z* 364 and characteristic fragments of *m/z* 248 (M−116), and 230 (M−116−18). The CD spectrum displayed a maximum at 267 nm with $\Delta\varepsilon = +1{\cdot}4$ and at 216 nm with $\Delta\varepsilon = -46{\cdot}4$. The ^{1}H NMR spectrum (Table I) proved the presence of a single ester group, *i.e.* of 2,3-dihydroxy-2-methylbutanoate (δ 1·33 s, 3 H; 1·21 d, 3 H, $J = 6{\cdot}3$; 3·92 q, 1 H, $J = 6{\cdot}3$), bound similarly as in ursiniolides *I*–*IV* in position 8. The parameters of all other hydrogen atoms are very similar to analogous parameters of 3-deacetoxyursiniolide B (*IV*), and they thus permit ursiniolide D to be assigned the structure *V* unambiguously. We derived the absolute configuration of ursiniolide D in an analogous manner, as shown for 3-deacetoxyursiniolide B (*IV*). We also confirmed the structure of ursiniolide D chemically by acetylation of the secondary hydroxyl group of this compound with acetic anhydride, which gave 3-deacetoxyursiniolide B (*IV*). On acetylation of the tertiary hydroxyl of lactone *IV* and of both free hydroxyls of compound *V* we prepared a compound identical with diacetylursiniolide D (*VI*), of the composition $C_{24}H_{32}O_8$, the IR spectrum of which contained bands of a γ-lactone grouping (1 760 cm^{-1}), an acetate group (1 743 and 1 236 cm^{-1}) and a double bond (1 660 cm^{-1}). The mass spectrum contained a molecular peak at *m/z* 448 and characteristic peaks at *m/z* 388 (M−60), 230 (M−60−158) and 43 (CH_3CO^+). The CD spectrum contained a maximum at 268 nm with $\Delta\varepsilon = +2{\cdot}2$ and at 216 nm with $\Delta\varepsilon = -69{\cdot}7$. The ^{1}H NMR spectrum (Table I) showed the presence of two acetoxy groups (δ 2·04 s, 2·08 s), localized in one ester group – 2,3-diacetoxy-2-methylbutanoate (δ 1·59 s, 3 H; 1·13 d, 3 H, $J = 6{\cdot}6$; 5·23 q, 1 H, $J = 6{\cdot}6$) – bound in the position 8β. The agreement of other parameters confirms that the common acetylation product has the structure *VI*.

A further minor lactone was ursanthemolide (*VII*), of the composition $C_{22}H_{30}O_8$. Its IR spectrum contained bands corresponding to a γ-lactone grouping (1 763 cm^{-1}), an acetate group (1 738 and 1 238 cm^{-1}), a hydroxyl (3 600 and 3 545 cm^{-1}) and a double bond (1 638 and 1 666 cm^{-1}). The mass spectrum did not contain the molecular peak, but it did have characteristic fragments with *m/z* 404 (M−18), 362 (M−60), 264 (M−60−98), 246 (M−60−98−18), 228 (M−60−98−18−18), 71 ($C_4H_7CO^+$), 43 (CH_3CO^+). The CD spectrum displayed a maximum at 224 nm with $\Delta\varepsilon = -6{\cdot}9$ and at 206 nm with $\Delta\varepsilon = +8{\cdot}3$. The ^{1}H NMR spectrum (Table I) indicated the presence of 2-hydroxy-3-acetoxy-2-methylbutanoate as the sole ester group, exomethylene lactone (δ 6·41 bd and 5·77 d), another exomethylene group (δ 5·42 bd and 5·05 d) and only one methyl group on a trisubstituted double bond

TABLE I

^{1}H NMR Parameters of minor sesquiterpenic lactones from *U. anthemoides* and their derivatives

Proton	Chemical shifts and coupling constants									
	IV[b]	*V*[c]	*VI*[d]	*VII*[d]	*VIII*[d]	*IX*[d]	*X*[d]	*XI*[e]	*XII*	*XIII*[f]
$H_{(1)}$	4·96	4·99	5·30	3·85	4·67	4·56	4·52	5·05	3·41	4·80
$H_{(2)}$	|	2·22	|	|	|	|	|	|	1·83	|
$H_{(2')}$	2·11—	2·12	1·93—	2·00—	2·00—	2·03—	2·00—	2·11—	1·56	1·60—
	—2·36		—2·34	—2·15	—2·20	—2·26	—2·25	—2·46		—2·46
$H_{(3)}$	|	2·33	|	|	|	|	|	|	2·38	|
$H_{(3')}$	1·90	1·91	|	|	|	|	|	|	2·05	|
$H_{(5)}$	5·34	5·32	5·43	5·13	5·21	5·17	5·15	4·58	1·84	|
$H_{(6)}$	5·34	5·32	5·30	5·43	5·40	5·40	5·39	5·71	4·88	4·89
$H_{(7)}$	3·41	3·39	3·37	3·62	3·67	3·60	3·65	—	3·66	3·56
$H_{(8)}$	5·34	5·32	5·30	5·13	5·21	5·10	5·22	—	5·39	5·42
$H_{(9)}$	2·73	2·71	2·75	2·61	2·87	2·74	2·78	3·50	2·15	1·98
$H_{(9')}$	2·40	2·45	2·34	2·35	2·52	2·48	2·48	3·10	1·68	1·85
$H_{(13)}$	6·40	6·38	6·40	6·41	6·38	6·42	6·35	1·91	6·35	6·33
$H_{(13')}$	5·77	5·79	5·77	5·77	5·79	5·79	5·77	—	5·97	5·92
$H_{(14)}$	1·40	1·44	1·43	5·42	5·42	5·29	5·32	1·53	0·83	1·00
$H_{(14')}$	—	—	—	5·05	5·33	5·07	5·20	—	—	—
$H_{(15)}$	1·75	1·76	1·75	1·70	1·69	1·69	1·68	1·64	5·04	5·11
$H_{(15')}$	—	—	—	—	—	—	—	—	4·88	4·95
CH	4·92	3·92	5·23	5·06	5·24	5·04	5·23	—	5·02	5·27
CH_3	1·44	1·33	1·59	1·39	1·77	1·39	1·77	—	1·38	1·70
	1·18	1·21	1·13	1·30	1·31	1·29	1·31	—	1·28	1·28
OAc	2·04	—	2·04	1·94	2·06	1·93	2·00	—	2·00	2·09
			2·08			2·00	2·06			
NH	—	—	—	—	8·40	—	8·41	—	—	8·45
					8·43					8·48
$J_{1,2}$	[a]	5·5	[a]	7·9	[a]	8	8	5	4·7	4·4
$J_{1,2'}$	[a]	10·6	[a]	2·0	[a]	3	3	11·5	11·4	11·4
$J_{5,6}$	[a]	[a]	11·0	9·3	[a]	9·3	9·3	9·5	10·2	10·0
$J_{5,15}$	0	0	0	1·3	1	1·3	1·2	1·2	0	0
$J_{6,7}$	[a]	[a]	7·1	8·6	8·5	8·6	8·6	—	7·6	7·6
$J_{7,8}$	[a]	[a]	[a]	3·0	3	3·3	3	—	5·7	5
$J_{7,13}$	2·5	2·1	2·4	3·6	3·5	3·5	3·6	—	3·2	2·7
$J_{7,13'}$	2·3	1·9	2·2	3·2	3	3·2	3·2	—	2·9	2·4
$J_{8,9}$	3	3	2·8	3·9	[a]	3·8	3·8	—	4·8	5·0
$J_{8,9'}$	4·6	5·3	5·2	3·9	[a]	3·8	4·0	—	10·4	7·4
$J_{9,9'}$	15·4	15·2	15·2	17·2	[a]	17·4	17·5	10·2	13·3	14·4
J_{H,CH_3}[g]	6·5	6·3	6·6	6·4	6·4	6·4	6·5	—	6·4	6·6

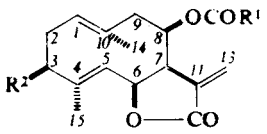
[a] The value could not be determined; [b] $J_{3',2} = 5·9$; $J_{3',2'} = 10·8$; $J_{3,3'} = 10·9$; [c] $J_{2,2'} = 12·8$; $J_{2',3} = 3·8$; $J_{2',3'} = 11·2$; $J_{2,3} = 3·0$; $J_{2,3'} = 5·8$; $J_{3,3'} = 11·1$; [d] $J_{1,14} = 1$; $J_{1,14'} = 0$; $J_{9,14} = 2·5$; $J_{9,14'} = 2·3$; $J_{9',14} = 1·2$; $J_{9',14'} = 0$; [e] $J_{1,14} = 1$; [f] $J_{2,2'} = 13$; $J_{2,3} = 2·4$; $J_{2,3'} = 5·2$; $J_{2',3} = 4·9$; $J_{2',3'} = 12·4$; $J_{3,3'} = 13·9$ Hz; [g] in an ester group.

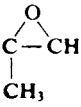
(δ 1·70 d, 3 H, J = 1·3 and 5·13 bd, 1 H, J = 9·3 and 1·3 Hz). The broad doublet at δ 3·85 was assigned to the CH—OH hydrogen atom of the secondary hydroxyl.* The multiplet of the $H_{(7)}$ atom at δ 3·62 can be assigned on the basis of allylic interactions (3·6 and 3·2 Hz) with the exomethylene $H_{(13)}$ and $H_{(13')}$ atoms. The vicinal couplings of $H_{(7)}$ (8·6 and 3·0 Hz) led to: *1*) assignment of $H_{(6)}$ at δ 5·43 ($J_{6,7}$ = = 8·6 Hz) the further coupling of which leads to $H_{(5)}$ at δ 5·13 ($J_{6,5}$ = 9·3 Hz) which must be an olefinic proton of a trisubstituted double bond, as following from the allylic interaction of 1·3 Hz with the methyl $H_{(15)}$ atoms at δ 1·69; *2*) assignment of $H_{(8)}$ at δ 5·13 ($J_{7,8}$ = 3·0 Hz), and thus to the localization of the ester group into the position $C_{(8)}$. $H_{(8)}$ has another two vicinal couplings, both equal to 3·9 Hz, with the methylene $H_{(9)}$ and $H_{(9')}$ atoms at δ 2·61 or 2·35, respectively. The high value of their geminal interaction ($J_{9,9'}$ = 17·5 Hz) and the allylic interaction with the hydrogen atoms of the second exomethylene (δ 5·42 and 5·05) confirm the position $C_{(10)}$ of this exomethylene group.

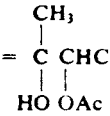
Since both exomethylene hydrogen atoms on $C_{(14)}$ have non-zero allylic couplings with the hydrogen atom of CH—OH type at δ 3·85, the secondary hydroxyl must be localized in the vicinity of the exomethylene, *i.e.* in the position 1. The remaining four hydrogen atoms on $C_{(2)}$ and $C_{(3)}$ form a strongly interacting system affording a multiplet in the δ 2·00 to 2·15 region. The mentioned NMR arguments led us to the structure *VII*, with a germacra-4,10(14)-dien-6,12-olide basis. On irradiation of the methylene hydrogens $H_{(15)}$, no NOE-enhancement of the $H_{(5)}$ signal was observed which indicates a *trans* configuration on the double bond between $C_{(4)}$ and $C_{(5)}$. The values of the couplings $J_{6,7}$ = 8·6, $J_{7,13}$ = 3·5 and $J_{7,13'}$ = 3·2 Hz indicate a *trans*-annellation of the α-exomethylene-γ-lactone group with respect to the homocycle, and $J_{7,8}$ = 3·0 Hz a *cis*-arrangement of the $H_{(7)}$ and $H_{(8)}$ hydrogens – and thus, the 8β-configuration of the ester group. The coupling constants in the fragment $C_{(5)}$ to $C_{(9)}$ and an analysis of Dreiding models show that the ten-membered ring assumed the preferred conformation with *cis*-oriented $C_{(4)}$-methyl and $C_{(10)}$--exomethylene which are directed above the ring plane. In this conformation the $C_{(1)}$-hydroxyl group can be assigned α-configuration on the basis of the coupling constants $J_{1,2}$ = 8·0 and $J_{1,2'}$ = 2·0 Hz. Hence ursanthemolide should have the structure *VII*.

Ursanthemolide acetate (*IX*), prepared by acetylation of the native substance *VII*, had m.p. 115–117°C and the composition $C_{24}H_{32}O_9$, while the IR and the mass spectrum were in accord with the assumed structure. The CD spectrum had maxima at 221 nm, with $\Delta\varepsilon$ = −8·1, and 205 nm, with $\Delta\varepsilon$ = +9·3. The ^{1}H NMR spectrum

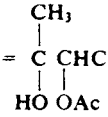
* The correctness of the assignment of the CH—OH fragment and the presence of the second (tertiary) hydroxyl in the molecule were confirmed by *in situ* acylation with trichloroacetyl isocyanate (TAI). The product of acylation *VIII* afforded the signals of two NH protons in the ^{1}H NMR spectrum, together with induced shifts of CH—OR and CH_3—C—OR hydrogen atoms (Table I).

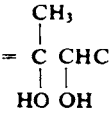


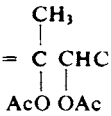
I: R^1 =  R^2 = OAc

II: R^1 =  R^2 = OAc

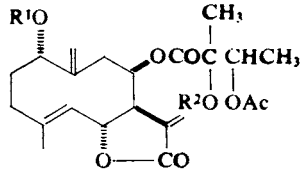
III: R^1 =  R^2 = OH

IV: R^1 =  R^2 = H

V: R^1 =  R^2 = H

VI: R^1 =  R^2 = H

Ac = $COCH_3$

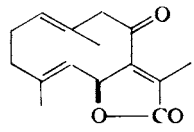


VII: R^1, R^2 = H

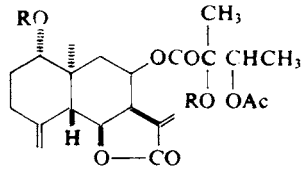
VIII: R^1, R^2 = $CONHCOCCl_3$

IX: R^1 = Ac; R^2 = H

X: R^1 = Ac; R^2 = $CONHCOCCl_3$



XI



XII: R = H

XIII: R = $CONHCOCCl_3$

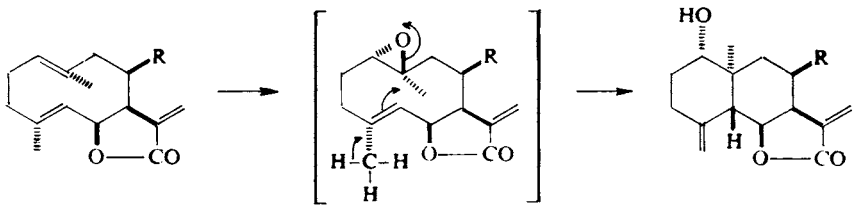
(Table I) confirmed the introduction of the second acetoxy group (δ 1·93 s and 2·00 s), accompanied by the characteristic acylation shift of the $H_{(1)}$ atom to δ 4·56 ($\Delta\delta$ = = 0·71 ppm), and indistinct downfield shifts of the atoms $H_{(9)}$ and $H_{(9')}$ ($\Delta\delta$ = 0·13 ppm) and an upfield shift of the exomethylene atom $H_{(14)}$ ($\Delta\delta$ = −0·13 ppm). The other ^{1}H NMR parameters remained practically unchanged. From these facts the expected structural relatedness of acetate *IX* with lactone *VII* can be inferred. The free tertiary hydroxyl in the ester group of *IX* was proved by *in situ* TAI-acylation. The derivative *X* formed was characterized by its ^{1}H NMR spectrum (Table I).

From the mixture of compounds we further isolated compound *XI* with m.p. 116–117°C of the composition $C_{15}H_{18}O_3$ (M 246·125) the IR spectrum of which indicated the presence of the γ-lactone grouping (1 758 cm^{-1}) and a conjugated keto group (1 688 cm^{-1}). The CD spectrum of lactone *XI* contained a maximum at 313 nm with $\Delta\varepsilon = +6{\cdot}7$, 245 nm with $\Delta\varepsilon = -5{\cdot}7$ and 218 nm with $\Delta\varepsilon = +53{\cdot}0$. All the mentioned data, with the exception of the $\Delta\varepsilon$ value in the region about 215 nm in the CD spectrum, the same as the ^{1}H NMR parameters (Table I), indicate that this substance is identical with the lactone isolated from *Wunderlichia mirabilis* RIEDEL (*Compositae*, tribe *Mutisiae*)[8] to which the structure *XI* has been assigned[8].

The last isolated was compound *XII*, m.p. 182–184°C, composition $C_{22}H_{30}O_8$, to which we gave the name cycloursiniolide. Its mass spectrum did not contain the molecular peak. The fragments were at *m/z* 404 (M–18), 336 (M–86), 318 (M–86 to 18), 246 (M–176), 228 (M–176–18). The IR spectrum indicated the presence of a hydroxyl group (3 555 and 3 525 cm^{-1}), a γ-lactone ring (1 765 cm^{-1}), an acetate group (1 735 and 1 243 cm^{-1}) and a double bond (1 642 and 1 664 cm^{-1}). The CD spectrum had a maximum at 210 nm (the last reading) with $\Delta\varepsilon = -2{\cdot}8$. The structure of compound *XII* was determined from its ^{1}H NMR spectrum (Table I). In it the signals of a 2-hydroxy-3-acetoxy-2-methylbutanoate group could be identified (δ 1·28 d, 3 H, J = 6·4; 1·38 s, 3 H; 5·02 q, 1 H, J = 6·4 Hz), further the signals of an α-exomethylene-γ-lactone (δ 6·35 d, J = 3·2; 5·97 d, J = 2·9 Hz) and another exomethylene group (δ 5·04 t, J = 1·8; 4·88 um), and a tertiary sp^3--methyl (δ 0·83 s). The doublet of doublets at δ 3·41 (J = 11·4 and 4·7 Hz) was assigned to the CH—O hydrogen of the secondary hydroxyl. The presence of two hydroxy groups in the molecule was demonstrated by *in situ* acylation with trichloroacetyl isocyanate which gave derivative *XIII* (NH signals at δ 8·48 and 8·45) with a characteristic α-acylation shift of the CH—O hydrogen at δ 3·41 ($\Delta\delta$ = 1·39 ppm) and a β-shift of the methyl of the ester group ($\Delta\delta$ = 0·32 ppm) in the ^{1}H NMR spectrum (Table I). The analysis of the spectrum of the native compound *XII* permitted the structural assignment of all the hydrogen atoms and the determination of the eudesmanolide basis, with the lactone ring in positions $C_{(6)}$ and $C_{(7)}$, the ester group on $C_{(8)}$, the hydroxyl group on $C_{(1)}$ and the exocyclic double bond at 4(15). The coupling constants of the fragment $C_{(5)}$ to $C_{(9)}$ and allylic coupling values $J_{13,7}$ and $J_{13',7}$ led to the determination of a skeleton of *trans*-decalin type, the *trans*-configuration of the $H_{(5)}$ and $H_{(6)}$ atoms ($J_{5,6}$ = 10·2), the *cis*-lactone group of the conformational type *S* ($J_{6,7}$ = 7·6, $J_{13,7}$ = 3·2, $J_{13',7}$ = 2·9 Hz)[9,10], toward which the ester group on $C_{(8)}$ is also *cis*-oriented ($J_{7,8}$ = 5·7, $J_{8,9}$ = 4·8, $J_{8,9'}$ = = 10·4 Hz). According to the values $J_{1,2}$ = 4·7 and $J_{1,2'}$ = 11·4 Hz, the atom $H_{(1)}$ is in the axial position and therefore the hydroxyl must be in an equatorial position. Hence, the isolated compound the structure represented by formula *XII* was assigned, with the relative configurations as shown there.

Among the native lactones described 3-deacetoxyursiniolide B (*IV*) and ursinio-

lide D (*V*) belong – according to their structure – to the group of ursiniolides (6αH,7αH-germacra-1(10)(*E*),4(*E*)-dien-6,12-olides[1,2]). Ursanthemolide (*VII*) with a *trans*-annellated γ-lactone ring and a double bond between $C_{(1)}$ and $C_{(14)}$ belongs to a different group of germacradienolides. In *U. anthemoides* compound *VII*



IV *XII*

R = OCOC(CH₃)(HO)CHCH₃OCOCH₃

SCHEME 1

(ursanthemolide) is the first so far described sesquitepenic lactone with a *trans*-annellated γ-lactone group. In other species of this genus, for example in *U. alpina*[11] or *U. nana*[12] several lactones with the mentioned arrangement have been described. The so far undescribed lactone cycloursiniolide (*XII*) has (the same as the newly described stereostructural type of eudesmanolides, characterized by the 5βH,6αH, 7αH,10αCH_3-eudesman-6,12-olide basis[13]) *trans*-annellated six-membered homocycles, a *cis*-annelated γ-lactone ring and *trans*-configuration of $H_{(5)}$ and $H_{(6)}$ atoms. Very probably cycloursiniolide is in a close biogenetical relationship with the lactones of the ursiniolide type[1,2], especially with 3-deacetoxyursiniolide B (*IV*), from which its formation could be inferred according to the sequence in Scheme 1, which is considered general in the biogenesis of eudesmanolides from germacra--1(10),4-dienolides[14,15].

EXPERIMENTAL

The melting points were determined on a Kofler block and they were not corrected. The mass spectra were measured on an AEI 902 spectrometer. The IR spectra were measured in chloroform on a Zeiss UR 20 (Jena) spectrometer, unless stated otherwise. Optical rotation was determined on an objective polarimeter (Perkin–Elmer) in methanol. The CD spectra were recorded on a Roussel–Jouan Dichrographe CD 185. The ^{1}H NMR spectra were measured on a Varian XL-200 (200 MHz) instrument in deuteriochloroform, using tetramethylsilane as internal reference.

Isolation of the Lactones

The so-called lactone fraction[4] (3·0 g) was dissolved in chloroform and chromatographed on 100 g

of silica gel under mild pressure (0·2 MPa) using benzene with increasing content of acetone (5—15%) as eluent. The first fractions (residue 200 mg) were rechromatographed with benzene with 2% of acetone under mild pressure to afford 30 mg of compound *XI*, m.p. 116—117°C (acetone, hexane). IR spectrum (cm^{-1}); 1 758 (γ-lactone), 1 688 (conjugated ketone). Mass spectrum (*m/z*): 246 (M), 231, 228, 217, 204, 203, 163, 149, 124, 121, 108, 107, 93, 67. CD spectrum (nm, Δε): 313, +6·7; 286, ±0; 245, −5·7; 237, ±0; 218, +53·0. For $C_{15}H_{18}O_3$ (246·3) calculated: 73·13% C, 7·37% H; found: 73·02% C, 7·42% H. Further chromatographic fractions contained ursiniolide A (*I*) (ref.[1]), identified by IR and ^1H NMR spectra and mixture melting point with a standard, which was undepressed. Further fractions (residue 500 mg) afforded 3-deacetoxyursiniolide B (*IV*; 300 mg), m.p. 116—118°C (acetone, hexane) and $[\alpha]_D^{20}$ −170° (*c* 0·100). IR spectrum (cm^{-1}): 3 540 (hydroxyl), 1 761 (γ-lactone), 1 638 (ester), 1 660 (double bond). Mass spectrum (*m/z*): 406 (M), 388 (M−18), 362, 320, 302, 258 ($C_{14}H_{20}O_3$), 230 ($C_{15}H_{18}O_2$), 215, 188, 185, 169, 159, 145, 131, 119, 89, 71, 43. CD spectrum (nm, Δε): 267, +1·4; 252, +0; 216, −47·7. For $C_{22}H_{30}O_7$ (406·5) calculated: 65·01% C, 7·44% H, 0·25% act. H; found: 64·75% C, 7·39% H, 0·32% act. H. Further eluates contained ursiniolide B (*II*) (ref.[1]), identified by IR and ^1H NMR spectra and mixture melting point with a standard (undepressed). In further fractions ursiniolide C (*III*) (ref.[1]) was found, identified as mentioned in the case of ursiniolide A (*I*) and B (*II*). From further fractions (residue 120 mg) ursiniolide D (*V*; 45 mg) was isolated, m.p. 119—121°C (acetone, hexane), $[\alpha]_D^{20}$ −199° (*c* 0·100). IR spectrum (cm^{-1}): 3 550 (hydroxyl), 1 748 (γ-lactone, saturated ester), 1 661 (double bond). Mass spectrum (*m/z*): 364 (M), 320, 248 ($C_{15}H_{20}O_3$), 230 ($C_{15}H_{18}O\alpha$), 215, 188, 185, 145, 119, 105. CD spectrum (nm, Δε): 267, +1·4; 252, ±0; 216, −46·4. For $C_{20}H_{28}O_6$ (364·4) calculated: 65·92% C, 7·74% H, 0·55% act. H; found: 65·75% H, 7·39% H, 0·67% act. H. The residue of further fractions (700 mg) was rechromatographed on silica gel (100 g) under mild pressure, using chloroform with 4% of 2-propanol as eluent. The residue of first fractions (300 mg) was chromatographed under the conditions of HPLC on a series of 4 columns (9 × 300 mm) packed with Lichrosorb Si 60 15—25 μ, flow rate 5 ml/min, elution with chloroform with 2·5% of 2-propanol, to afford ursanthemolide (*VII*; 57 mg), m.p. 74—76°C and $[\alpha]_D^{20}$ −25·8°. IR spectrum (cm^{-1}); 3 600, 3 545 (hydroxyl), 1 763 (γ-lactone), 1 738, 1 238 (acetate), 1 638, 1 666, (double bond). Mass spectrum (*m/z*): 404 (M), 361, 347, 336, 264 ($C_{15}H_{20}O_4$), 247, 246 ($C_{15}H_{18}O_3$), 229, 228 ($C_{15}H_{16}O_2$), 213, 204, 183, 167, 149, 131, 105, 89, 71, 43. CD spectrum (nm, Δε): 224, −6·9; 215, ±0; 206, +8·3. For $C_{22}H_{28}O_7$ (404·5) calculated: 65·32% C, 6·98% H, 0·50% act. H; found: 65·64% C, 6·71% H, 0·63% act. H. The residue from further fractions of low-pressure chromatography (140 mg), obtained by elution with chloroform with 4% of 2-propanol was chromatographed under the conditions of HPLC, as mentioned above, but the eluent was a mixture of hexane with ethyl acetate (3 : 1). Cycloursiniolide (*XII*; 22 mg) was obtained, m.p. 182 to 184°C. IR spectrum (cm^{-1}); 3 555, 3 525 (hydroxyl), 1 765 (γ-lactone), 1 735, 1 243 (acetate), 1 642, 1 664 (double bond). Mass spectrum (*m/z*): 404 (M−18), 336, 318, 246 ($C_{15}H_{18}O_3$), 228 ($C_{15}H_{16}O_2$) 213, 202, 183, 181. CD spectrum (nm, Δε): 210 (last reading), −2·8. For $C_{22}H_{30}O_8$ (422·5) calculated: 62·34% C, 7·16% H, 0·48% act. H; found: 62·51% C, 7·13% H, 0·58% act. H.

Interconversion of Compounds *IV* to *VI*

A) Ursiniolide D (*V*; 20 mg) in 2 ml of a mixture of pyridine and acetic anhydride (1 : 1) was allowed to stand at room temperature for 18 h. After working up and chromatography of the residue (20 mg) on silica gel (2 g) 3-deacetoxyursiniolide B (*IV*) was obtained, m.p. 115—117°C, which was identified by IR and ^1H NMR spectra and mixture melting point with a standard (undepressed).

B) Ursiniolide D (*V*; 20 mg) in 2 ml of pyridine and acetic anhydride (1 : 1) was heated at 40°C for 4 h. After working up diacetylursiniolide D (*VI*) was obtained, m.p. 155—159°C and $[\alpha]_D^{20}$ −187° (*c* 0·111). IR spectrum (cm^{-1}): 1 760 sh (γ-lactone), 1 743, 1 236 (acetone), 1 660 (double bond). Mass spectrum (*m/z*): 448 (M), 388 (M−60), 362, 320, 302, 230 ($C_{15}H_{18}O_2$), 215, 201, 188, 185, 159, 145, 131, 43. CD spectrum (nm, Δε): 268, +2·2; 251, ±0; 216, −69·7. For $C_{24}H_{32}O_8$ (448·4) calculated: 64·27% C, 7·19% H; found: 64·06% C, 7·24% H.

C) 3-Deacetoxyursiniolide B (*IV*; 20 mg) was acetylated as in the preceding case. Diacetylursiniolide D (*VI*) with m.p. 154—156°C was obtained which was identified by its IR and ^{1}HNMR spectrum and mixture melting point with a sample from the preceding experiment.

Acetylursanthemolide (*IX*)

Ursanthemolide (*VII*; 20 mg) in 2 ml of a mixture of pyridine and acetic anhydride (1 : 1) was allowed to stand at room temperature for 18 h. After working up the residue (20 mg) was chromatographed on silica gel (2 g) under a mild pressure. Elution with benzene with 10% of acetone gave acetylursathemolide (*IX*), m.p. 115—117°C. IR spectrum (cm^{-1}): 3 545 (hydroxyl), 1 764 (γ-lactone), 1 739, 1 245 (acetate), 1 659, 1 641 (double bond). Mass spectrum (*m/z*): 464 (M), 446 (M−18), 404 (M−60), 378 ($C_{20}H_{26}O_7$), 246 ($C_{15}H_{18}O_3$), 228 ($C_{15}H_{16}O_2$), 131, 89, 71, 43. CD spectrum (nm, Δε): 221, −8·1; 212, ±0; 205 (last reading), +9·3. For $C_{24}H_{32}O_9$ (464·5) calculated: 62·06% C, 6·95% H, 0·22% H act.; found: 61·87% C, 7·13% H, 0·32% H act.

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