Tetrahedron Vol. 46, No. 10, pp. 3569-3580, 1990
Printed in Great Britain

0040-4020/90 $3.00+.00

PROLONGED TREATMENT OF ACETYLMETHYL URSOLIC ACID WITH H_2O_2/ACETIC ACID — A FACILE ONE-POT SYNTHESIS OF 15α-HYDROXY ANALOGUE

Salimuzzaman Siddiqui, Bina Shaheen Siddiqui*, Qayyum Adil and Sabira Begum

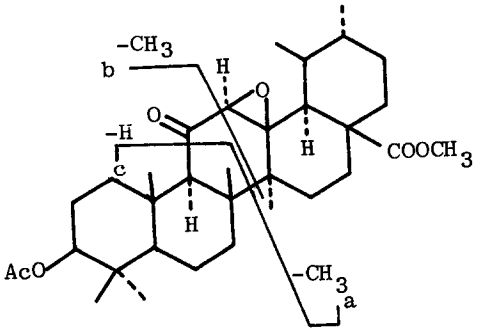
H.E.J.Research Institute of Chemistry, University of Karachi, Karachi-75270, Pakistan

(*Received in UK* 2 *March* 1990)

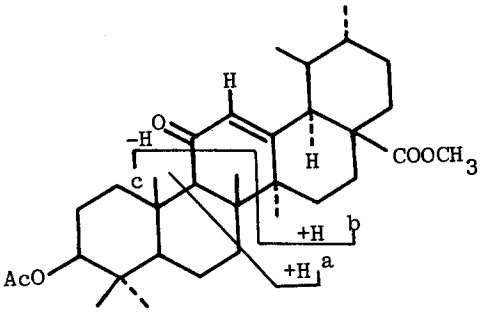
Abstract - With a view to obtain new compounds bearing potential applications in the biogenetic type synthesis of natural products, acetylmethyl ursolic acid (**1**) was subjected to prolonged treatment with H_2O_2/AcOH on boiling water bath. Several such products (**2**,**3**, **5** & **7**) have thus been obtained. A new one-pot partial synthesis of 15α-hydroxy Δ^{12}-analogues of pentacyclic triterpenes of amyrin series has been achieved. ^{13}C-NMR spectral data of the major product are also described.

INTRODUCTION

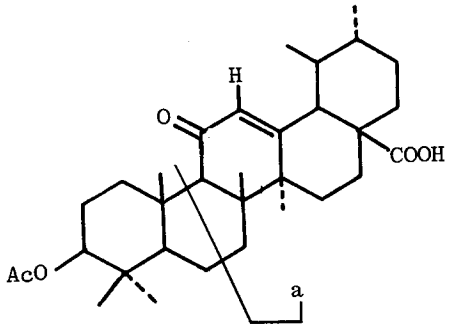
Oxidative transformations[1-5] of pentacyclic triterpenes have attracted the attention of various groups of workers for a long time from synthetic as well as biological point of view. Several such reactions including photochemical transformations[6] leading to biogenetic type total synthesis of natural products have been reported providing a possibility of converting natural products from one into another. One such reaction[4] involved the action of hydrogen peroxide on ursolic acid acetate in hot glacial acetic acid which afforded three products. In the present working the effect of a prolonged treatment of the same reagent on (**1**) was studied. The reaction product after separation through flash column chromatography furnished several interesting products (**2**,**3**,**4**,**5** & **7**) apart from the 11α,12α-epoxy lactone (**6**) generally encountered in peroxide,[2-4] auto-[3] and photochemical[6] oxidation reactions. The reaction furnishes a possible one-pot synthesis of 15α-hydroxy compounds from Δ^{12}-pentacyclic triterpenes of amyrin series and a method of introducing a carbonyl group at C-11, C-C double bond at C-15 and an 11-keto, 12,13-β epoxy moiety. The insertion of these functionalities shows the utilization of this reaction in the synthesis of natural products.


-CH3
b
H
O
O
-H
H
COOCH3
c
H
AcO
-CH3
a

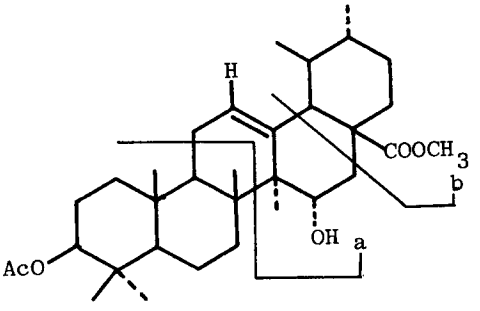
2


H
O
-H
H
COOCH3
c
+H
b
AcO
+H
a

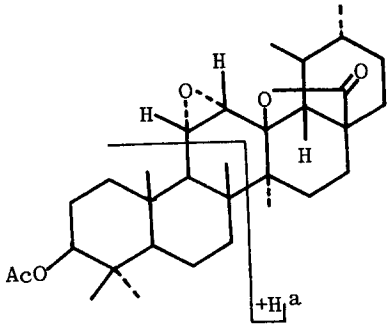
3


H
O
COOH
AcO
a

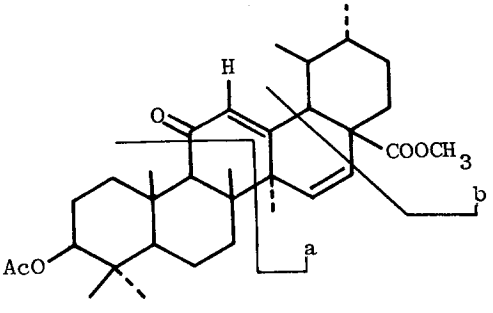
4


H
COOCH3
b
OH
a
AcO

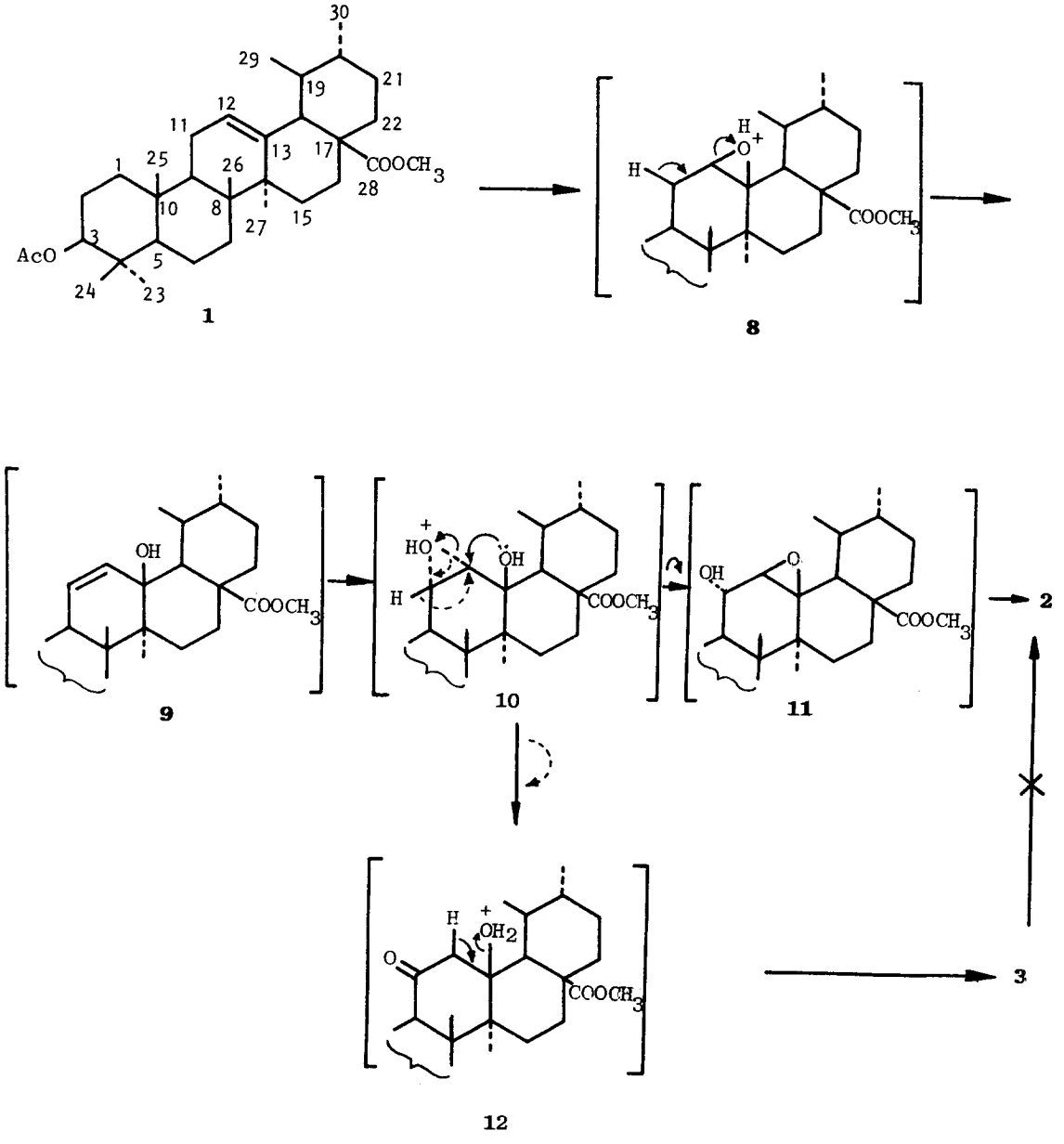
5


H
O
O
O
H
H
AcO
+H
a

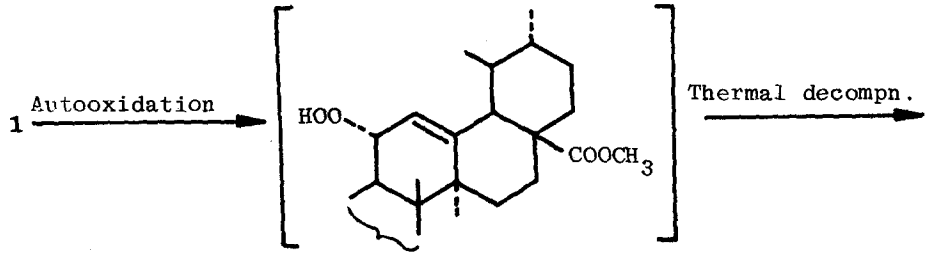
6


H
O
COOCH3
b
a
AcO

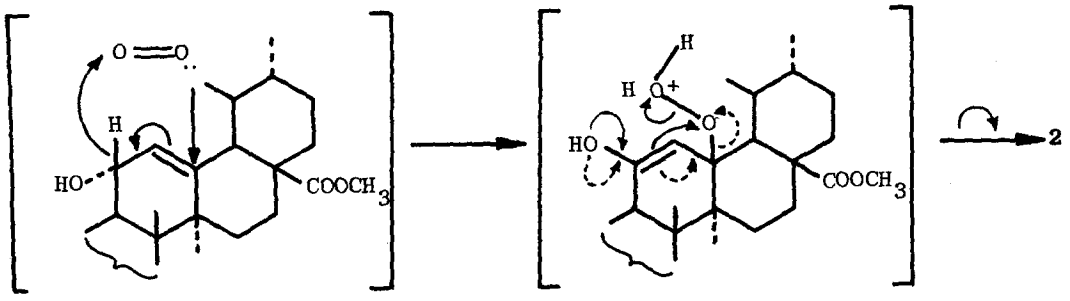
7


30
29
19
21
12
11
22
1
25
26
13
17
COOCH3
28
10
8
27
15
AcO
3
5
24
23
1
H
O+
8
OH
9
HO
10
11
2
12
OH2
3

Scheme 1

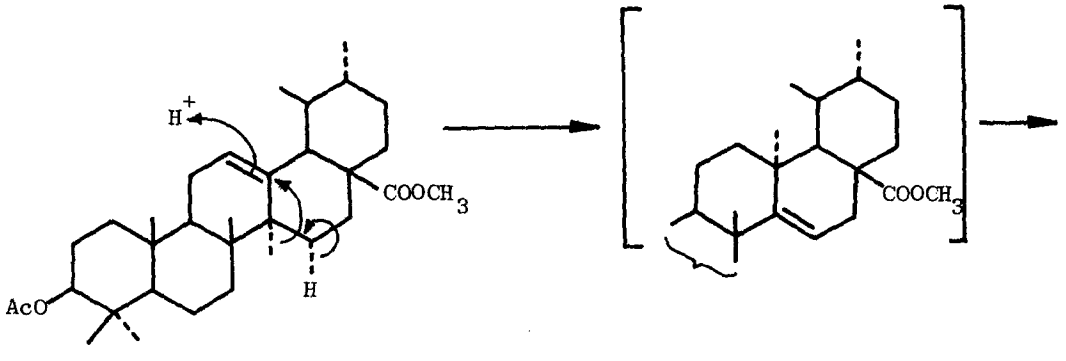


13

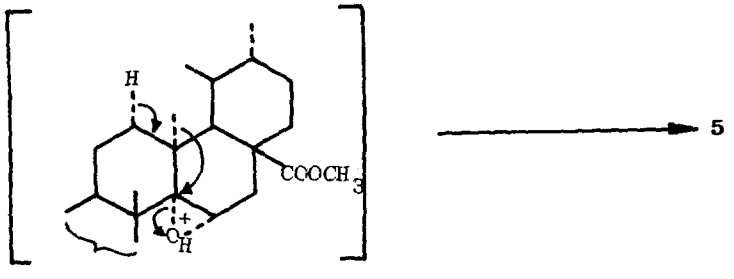


13a **14**

Scheme 2



1 **15**



16

Scheme 3

RESULTS AND DISCUSSION

The molecular formula ($C_{33}H_{50}O_6$; M^+, 542.3637) of **2**, the least polar compound was obtained through the high resolution mass spectrum, which showed an increment of 30 a.m.u. from that of **1** and incorporation of two more oxygen atoms with the loss of two hydrogens. In the ^{1}H-NMR spectrum the signal of H-12 of **1** was replaced by a singlet at δ 3.67 suggesting an epoxy ring between C-12 and C-13. This singlet showed that there must be a ketonic function at C-11 which was also indicated by the IR band at 1705 cm^{-1}. The steric requirements and the mechanism proposed in scheme 1 favour a β-epoxy ring between C-12 and C-13. These features were corroborated by significant fragments at m/z 277.1799 (fragment a), 263 (fragment b), 218 (a-$COOCH_3$), 203 (a-$COOCH_3$-CH_3) and 189 (fragment c -CH_3COOH) in the mass spectrum. Thus it was arrived at that **2** is methyl 3β-acetoxy-11-oxo-12β,13β-epoxy-28-oate. In the ^{1}H-NMR spectrum of **3** ($C_{33}H_{50}O_5$, vide HRMS) the H-12 signal was shifted to δ 5.59 with change of its multiplicity from triplet to a singlet showing that 3 is the allylic oxidation product of **1**. The rest of the ^{1}H-NMR features were same as those of **1**. The structure of **3** as methyl 3β-acetoxy-11-oxo-12-ene-28-oate was finally confirmed through fragments a-c (vide structure) described in the experimental. The ^{1}H-NMR spectrum of **4** showed only one difference from that of **3** i.e. the absence of the OCH_3 singlet at δ 3.5, indicating the hydrolysis of the methyl ester at C-17 during the reaction. The structure as 3β-acetoxy, 11-oxo-12-ene-28-oic acid was supported by the molecular ion peak at 512.3501 ($C_{32}H_{48}O_5$), which showed the loss of a-CH_2 moiety and the diagnostic fragments a and b (vide structure).

Two possible pathways (schemes-1 & 2) can be suggested for the formation of **2** and **3**. The first three steps (**1** → **10**) of scheme 1 follow from those reported by Majumder *et al* (4). Nucleophilic attack of the C-13 hydroxyl group at C-12 and opening of the 11α,12α-epoxide ring would give the more stable[7] isomeric hydroxy epoxide **11**, the oxidation of which can afford 11-keto-12β,13β-epoxide **2**. On the other hand, the opening of the epoxide ring of **10** with the migration

of the C-11 proton to C-12 would result in the 11-keto derivative **12** (a similar opening of the 12β,13β-epoxide gave 12-keto ursolic acid),[4] dehydration of which may lead to **3**. The epoxidation of **3** to **2** is not favoured, **3** being an α-β-unsaturated ketone. Evidence of this came when **3** was recovered unchanged on its attempted epoxidation with *m*-CPBA as well as H_2O_2/NaOH under varying reaction conditions.[8] On the other hand **13** may be the auto-oxidation product of **1** which after thermal decomposition to **13a** leads to **2** and **3** through the attack of singlet oxygen formed in situ from the acetic acid and acetic acid anion[9,10] as shown in scheme 2. Therefore, the same reaction was repeated under nitrogen atmosphere employing the same reaction conditions in respect of reagents, temperature and time period which again afforded **2** and **3**, and thus eliminated the possibility of auto-oxidation of **1** to **13** and favoured the mechanism shown in scheme-1

To our knowledge allylic oxidation product (**3**) has not been reported earlier with peracids although such oxidations with H_2O_2 in the presence of oxidants such as Cu are known.[11] Thus the reaction provides an interesting method for the preparation of 11-oxo derivatives.

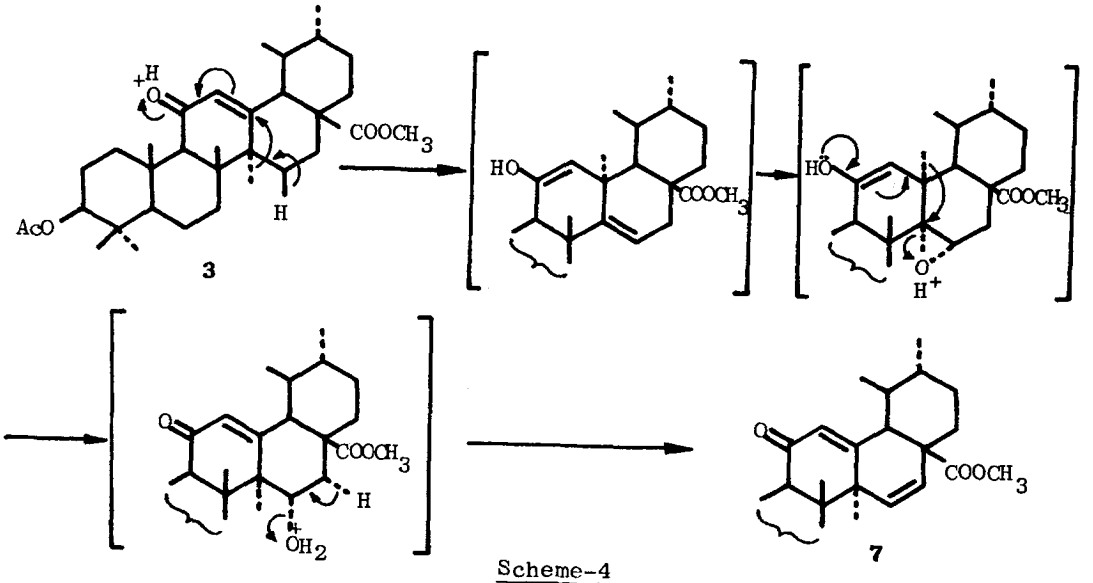
The molecular ion peak of **5** was observed at m/z 528, the exact measurement (528.3810) of which gave the molecular formula as $C_{33}H_{52}O_5$, demonstrating the incorporation of an oxygen atom in the acetylmethyl ursolate molecule, which was identified as a hydroxyl function from its IR (3600 cm^{-1}) and acetylation (Ac_2O/pyridine) to **5a** (δ 2.1, $COCH_3$, δ 4.4dd, J=10,5Hz, H-15). The ^{1}H-NMR spectrum showed a close similarity of **5** with **1**, viz. the triplet of H-12 at δ 5.22 (J=3.6 Hz), the dd of H-3 at 4.43 (J=10.68, 5.43 Hz), and the singlet of OCH_3 and OCOCH at 3.57 and 2.01 respectively. Apart from these it showed a signal at δ 4.18 (1H dd, J=10.89, 5.64 Hz, H-15β) attributable to the carbinylic proton.[12] An ion at m/z 278.1875 ($C_{17}H_{26}O_3$, fragment a) resulting from the retro Diels Alder cleavages around ring C exhibited that the hydroxyl function is in ring D/E. The fragment b at m/z 168.1158 ($C_{10}H_{16}O_2$) further limited its possibility to ring E and thus left only two possible positions i.e. either at C-15 or at C-16. An upfield shift (6.56 ppm) of C-27 (δ 17.04) in the ^{13}C-NMR spectrum (table 2) of **5** as

compared to that of ursolic acid (δ 23.60) showed that the hydroxyl group is at C-15 with an α-disposition (which is also evident from the coupling constants of H-15β) and thus exerts a γ-gauche effect[13] on C-27. In the alternate position (C-16) a similar effect would have been observed for C-22 which is not noted in this case and C-22 appears at almost same chemical shift (δ 36.67) as in ursolic acid (δ 36.7). These observations led to place the hydroxyl group at C-15. Considering the oleanane-taraxane interconversion in acidic conditions,[14] a mechanism for the formation of **5** is proposed in scheme 3.

Hydroxylation at an inactivated centre (i.e. C-15) of **1** to form **5** may be envisaged to go through the Δ^{14}-derivative (**15**) formed by acid catalysed rearrangement.[14] Its epoxidation to the unstable 14α,15α-epoxide (**16**), subsequent acid catalysed ring opening followed by migration of the methyl group from C-13 to C-14 would form the 15α-hydroxy derivative (**5**) of the Δ^{12}-ursane skeleton (scheme 3). A similar transformation[15,16] of β-amyrin to 15α-hydroxy β-amyrin has been demonstrated earlier through several steps. The present reaction exemplifies a one-pot synthesis of a 15α-hydroxy derivative from Δ^{12}-analogues.

The molecular ion peak at 512.3501 ($C_{32}H_{48}O_5$), and other spectral data of **6** particularly the similarity of the chemical shifts and coupling constants of H-11, H-12 with those reported[4] revealed that **6** is 3β-acetoxy-11α,12α-epoxy-urs-13β, 28-olide.

The exact measurement of the molecular ion peak (m/z 524.3527) of **7**, the most polar reaction product gave its molecular formula as $C_{33}H_{48}O_5$ and the IR spectrum showed carbonyl stretching at 1710 and 1690 cm^{-1}. The salient features of the ^{1}H-NMR spectrum were: a singlet at δ 5.59 for H-12 suggesting a keto group at C-11; two AB doublets at δ 4.18 and 4.13 with the same coupling constant of 10.23 Hz. These data along with the molecular formula indicated one more double bond in the molecule. The ions at m/z 274.1568 (fragment a) and m/z 109.0944 (fragment b) conclusively decided the position of this double bond at C-15. The creation of this double bond may be imagined through the dehydration of the hydroxyl group at C-15 formed during the reaction as proposed in scheme 4.



Scheme-4

The spectral data of one of the column fractions revealed the presence of 12-keto derivatives analogous to the observation of Majumder *et al*[4]. The ^{13}C-NMR showed that it is a mixture of 13αH- and 13βH-isomers (vide δ_{C12}, 210.8, 214.8 and δ_{C13}, 51.7, 60.6),[17] but the limited quantity did not allow its purification.

Conclusions

i). Prolonged treatment of acetylmethyl ursolic acid with H_2O_2/acetic acid affords 15α-hydroxy acetylmethyl ursolic acid as the major product. ii). Formation of the 11α,12α-epoxy lactone (**6**) and the 12-keto derivative is in analogy with the earlier reaction of ursolic acid acetate with the same reagent.[4] iii). Formation of **6** shows that lactonization is also possible with the 17-carbomethoxy group as noted by Pradhan *et al*[2] with H_2O_2/SeO_2. iv). Allylic oxidation with peracids is noted for the first time. v). Products **2**,**3**,**5** and **7** have not been reported earlier either as natural or synthetic products, while **4** has earlier been obtained through chromic acid oxidation.[18]

Experimental

IR and UV spectra were measured on JASCO IRA-I and Pye-Unicam sp-800 spectrometers respectively. Mass spectra were recorded on Finnigan MAT-112. Exact

masses of various fragments were obtained through their peak matching and high resolution mass spectrum. ^{1}H and ^{13}C-NMR spectra were recorded in $CDCl_3$ on a Bruker AM 300 spectrometer operating at 300 MHz and 75 MHz respectively. The chemical shifts are reported in δ (ppm) and coupling constants in Hz. Assignments of ^{13}C-NMR chemical shifts are based on hetero-COSY and comparison with similar compounds.[19] Flash column (model Eyela) was used with silica gel (9385) as adsorbant and hexane and hexane: ethyl acetate, (gradually increasing the ratio of ethyl acetate) as eluant. Purity of the products was checked on t.l.c. plates using silica gel 60 PF_{254}. Ursolic acid, isolated from the leaves of Nerium oleander, was acetylated with pyridine and acetic anhydride and methylated with diazomethane following usual procedures.

Acetylmethyl ursolic acid (500 mg) was dissolved in glacial acetic acid (50 ml) and heated for 30 minutes on the boiling water bath. A mixture of glacial acetic acid (20 ml) and H_2O_2 (10 ml) was then added dropwise to the solution which was further heated for 33 hours. Working up of the reaction mixture at this stage in the usual manner afforded the reaction product containing a mixture of several compounds along with the unreacted starting material. The separation of these was achieved through flash column chromatography when compounds **1-7** were obtained with the solvent system hexane:ethyl acetate with the ratio noted in the parenthesis against each of these. **1** (9.75:0.25), **2-6** (9.50:0.50, eluted in the order of polarity and **7** (9.0:1.0).

Methyl 3β-acetoxy-11-oxo-12β,13β-epoxy-28-oate, (**2**). Amorphous powder. (4 mg) $\nu_{max}(CHCl_3)$: 2900, 2850, 1705 br, 1640, 1250-1180 and 1025 cm^{-1}; λ_{max}(MeOH): 202 nm; HRMS m/z = 542.3614 ($C_{33}H_{50}O_6$), 514.3637 ($C_{32}H_{50}O_5$, M^+-CO), 277.1799 ($C_{17}H_{25}O_3$, fragment a), 249 (fragment c), 263.1785 ($C_{20}H_{23}$, fragment b-H), 218.1677 ($C_{15}H_{22}O$, fragment a-$COOCH_3$), 203.1684 ($C_{11}H_{23}O_3$, fragment a-$COOCH_3$-CH_3), 189.1603 ($C_{14}H_{21}$, fragment c-H-CH_3COOH). ^{1}H-NMR (table-1).

Methyl 3β-acetoxy-11-oxo-12-ene-28-oate, (**3**). Amorphous powder (5.2 mg) $\nu_{max}(CHCl_3)$: 2920, 2840, 1705 br. 1690 (sh), 1650, 1255-1190 and 1030 cm^{-1};

λ_{max}(MeOH): 202 nm; HRMS m/z = 526.3687 ($C_{33}H_{50}O_5$), 483.3465 ($C_{31}H_{47}O_4$), 317.2134 ($C_{20}H_{29}O_3$, fragment a), 277.1799 ($C_{16}H_{21}O_4$, fragment b), 249.1835 ($C_{16}H_{25}O_2$, fragment c), 218.1677 ($C_{15}H_{22}O$, fragment b-$COOCH_3$), 203.1684 (fragment b-$COOCH_3$), 189.1603 ($C_{14}H_{21}$, fragment c-CH_3COOH). ^{1}H-NMR (table-1).

3β-Acetoxy-11-oxo-12-ene-28-oic acid, (**4**). Amorphous powder (15.0 mg). ν_{max}($CHCl_3$): 3450-2610 br, 2910, 2850, 1705br, 1690(sh), 1650, 1250-1180 and 1025 cm^{-1}; λ_{max}(MeOH): 201 nm; EIMS m/z = 512.3501 ($C_{32}H_{48}O_5$), 452.3290 ($C_{30}H_{44}O_3$, M^+-CH_3COOH), 316.2038 ($C_{20}H_{28}O_3$, fragment a). ^{1}H-NMR (table-1).

Methyl 3β-acetoxy-12-ene-15α-hydroxy-28-oate, (**5**). Amorphous powder (57.0 mg). ν_{max}($CHCl_3$): 3600, 2950-2850, 1710br, 1640 and 1260-1180 cm^{-1}; λ_{max}(MeOH): 203nm; HRMS m/z = 528.3810 ($C_{33}H_{52}O_5$), 513.3582 ($C_{32}H_{49}O_5$, M^+-CH_3), 469.3653 ($C_{31}H_{49}O_3$, M^+-$COOCH_3$), 454.3411 ($C_{30}H_{46}O_3$, M^+-$COOCH_3$-CH_3), 278.1875 ($C_{17}H_{26}O_3$, r.D.A. fragment a), 218.1678 ($C_{15}H_{22}O$, fragment a-CH_3COOH), 215.1704 ($C_{12}H_{23}O_3$), 203.1786 ($C_{15}H_{23}$, fragment a-CH_3COOH-CH_3), 201.1602 ($C_{15}H_{21}$, fragment a-$COOCH_3$-H_2O), 189.1622 ($C_{14}H_{21}$, 203-CH_2), 168.1158 ($C_{10}H_{16}O_2$).

3β-Acetoxy-11α,12α-epoxy-urs-13β,28-olide, (**6**). Yield 19.2 mg; ν_{max}($CHCl_3$): 2900-2825, 1755, 1710 and 1255-1180 cm^{-1}; λ_{max}(MeOH): 202.2 nm; EIMS m/z = 512.3501 ($C_{32}H_{48}O_5$), 496 (M^+-CH_3-H), 484.3552 ($C_{31}H_{48}O_4$, M^+-CO), 466.3446, 263.1647 (fragment a), 216, 203. ^{1}H-NMR (table-1).

Methyl 3β-acetoxy-11-oxo-12,15-diene-28-oate, (**7**). Amorphous powder (9.0 mg ν_{max}($CHCl_3$): 2890, 2825, 1710br., 1690(sh), 1650, 1640, 1280-1180 and 1025 cm^{-1}; λ_{max}(MeOH): 227.4, 202.6 nm; HRMS m/z = 524.3527 ($C_{33}H_{48}O_5$), 465.3357 ($C_{31}H_{45}O_3$, M^+-$COOCH_3$), 274.1568 ($C_{17}H_{22}O_3$, r.D.A. fragment a), 145.0948 ($C_7H_{13}O_3$), 215.1512 ($C_{15}H_{19}O$, fragment a-$COOCH_3$), 168 (fragment b), 109.0944 (C_8H_{13}, fragment b-$COOCH_3$). ^{1}H-NMR (table-1).

Table 1. ^{1}H-NMR spectral data of **2-7**.

Protons	Compounds					
	2	**3**	**4**	**5**	**6**	**7**
H-3	4.52 dd J=9.98,5.9Hz	4.51 dd J=9.96,6.27Hz	4.45 dd J=11.82,5.37Hz	4.43 dd J=10.68,5.43Hz	4.53 dd J=9.57,5.73Hz	4.29 dd J=10.07,4.47Hz
H-12	3.67 s	5.59 s	5.59 s	5.22 t J=3.66 Hz	2.93 d J=3.87Hz	5.59 s
H-9	2.30 s	-	-	-	-	-
H-18	2.23 d J=11.52Hz	2.23 d J=11.52Hz	2.21 d J=11.20Hz	2.20 d J=11.43Hz	2.12 ddd J=12.93,5.70Hz	2.30 d J=10.44Hz
O-Me	3.61 s	3.61 s	3.66 s	3.57 s	-	3.60 s
O-Ac	2.02 s	2.02 s	2.02 s	2.01 s	2.04 s	2.03 s
Me 29/30	1.13 d J=5.46Hz	0.98 d J=7.65Hz	0.92 d J=5.22Hz	0.93 d J=5.13Hz	1.21 d J=5.79Hz	0.98 d J=5.16Hz
"	0.96 d J=5.7Hz	0.89 d J=5.6Hz	0.88 d J=6.9Hz	0.84 d J=6.3Hz	0.98 d J=5.22Hz	0.93 d J=5.25Hz
Me	1.06 s	1.06 s	0.97 s	1.05 s	1.10 s	1.12 s
"	0.90 s	0.93 s	0.96 s	0.96 s	1.07 s	0.91 s
"	0.86 s	0.91 s	0.86 s	0.90 s	1.05 s	0.87 s
"	0.86 s	0.90 s	0.86 s	0.75 s	0.87 s	0.85 s
"	0.85 s	0.85 s	0.85 s	0.72 s	0.86 s	0.79 s
H-15	-	-	-	4.18 dd J=10.89,5.64Hz	-	4.13 d J=10.23Hz
H-11	-	-	-	-	3.10 dd J=3.9,2.16Hz	-
H-16	-	-	-	-	-	5.34 d J=5.16Hz

Table 2. ^{13}C-NMR spectral data of **5**

Carbon		Carbon	
C-1	38.67	C-16	28.06
C-2	27.27	C-17	48.13
C-3	79.07	C-18	52.94
C-4	38.77	C-19	39.09
C-5	55.28	C-20	38.91
C-6	18.20	C-21	30.69
C-7	33.01	C-22	36.67
C-8	39.69	C-23	28.16
C-9	47.61	C-24	15.45
C-10	37.01	C-25	15.63
C-11	23.33	C-26	16.93
C-12	125.64	C-27	17.04
C-13	138.23	C-28	178.12
C-14	42.03	C-29	15.27
C-15	80.48	C-30	21.19
		$\underline{C}O-CH_3$	170.95
		$CO-\underline{C}H_3$	21.25
		OCH_3	55.11

REFERENCES

1) Manson, W.; Spring, F.S. J.Chem.Soc., 1951, 3332-3336.
2) Pradhan, B.P.; Chakraborty, S. Tetrahedron, 1987, 43, 4487-4495.
3) Agata, I.; Corey, E.J.; Hortmann, A.G.; Klein, J.; Proskow, S.; Ursprung, J.J. J.Org.Chem., 1965, 30, 1698-1710.
4) Majumder, P.L.; Chakraborty, M. Tetrahedron, 1979, 35, 2397-2403.
5) Tori, M.; Matsuda, R.; Asakawa, Y. Tetrahedron, 1986, 42, 1275-1283.
6) Kitagawa, I.; Kitazawa, K.; Yosioka, I. Tetrahedron, 1972, 28, 907-921.
7) Brill, W.F. J.Am.Chem.Soc., 1963, 85, 141-145.
8) Valente, V.R.; Wolfhagen, J.L. J.Org.Chem., 1966, 31, 2509-2512.
9) Mckeown, E.; Waters, W.A. J.Chem.Soc.(B), 1966, 1040-1046.
10) Nickon, A.; Mendelson, W.L. J.Am.Chem.Soc., 1965, 87, 3921-3928.
11) Kharasch, M.S.; Sosnovsky, G.; Yang, N.C. J.Am.Chem.Soc., 1959, 81, 5819-5824.
12) Hui, W.H.; Li, M.M. Phytochemistry, 1976, 15, 1313-1315.
13) Wehrli, F.W; Wirthlin, T. Interpretation of Carbon-13 NMR Spectra, 1978, Heyden and Son Inc. U.S.A.
14) Simonsen, J.; Ross, W.C.J. The Terpenes, 1957, 4, Cambridge, The University Press, 212, 284-285.
15) Takeda, K.; J.Pharm.Soc.Japan, 1943, 63, 193-197.
16) Beaton, J.M.; Spring, F.S.; Stevenson, R.; Stewart, J.L. J.Chem.Soc., 1955, 2131-2137.
17) Farina, C.; Pinza, M. Gazzetta Chimica Italiana, 1987, 117, 561.
18) Mezzetti, T.; Orzalesi, G.; Bellavita, V. Planta Medica, 1971, 20, 244-251.
19) Seo, S.; Tomita, Y.; Tori, K. J.C.S.Comm., 1975, 954-955.