PART - II

STUDIES ON AUTOXIDATION: SYNTHESIS OF ISOMERIC 2, 3-DIOLS OF ISOHOPANE (MORETANE) AND METHYL OLEAN-12-EN-28-OATES.

PART - II

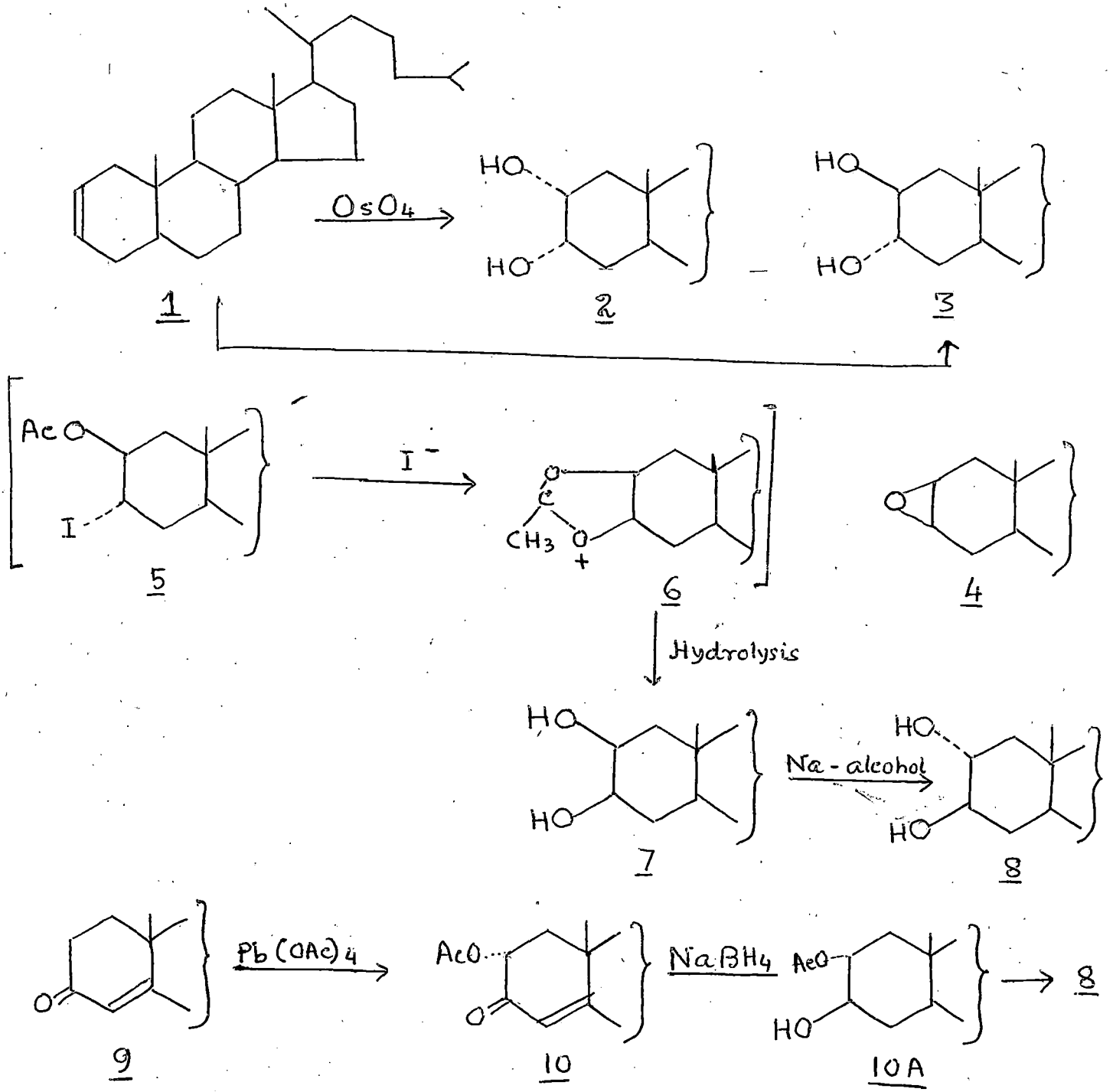
CHAPTER - I

A short review of synthesis of isomeric 2,3-diols of triterpenoids:

Section A: Introduction

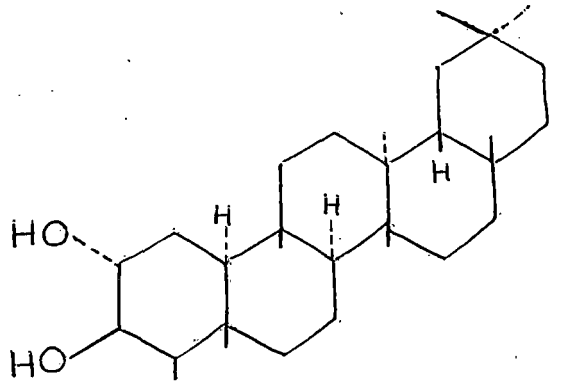
In the course of elucidation of the configurations of sapogenins containing a 2,3-diol system, methods were developed for production of the four possible isomers in this series. The four cholestane 2, 3-diols have also been prepared by the methods developed in this connection[1,2]. Δ^2-Cholestane 1 on reaction with osmium tetroxide gives the 2α, 3α-diol 2 whereas peracetic acid oxidation and subsequent hydrolysis affords 2β, 3α-diol[1-3] 3. 2α, 3α-or 2β, 3β-oxido-cholestanes 4 on diaxial opening also gives the same diol 3. The 2β, 3β diol 7 has also been prepared[1,2] according to the procedure of Winstein and Buckles[4] by treatment of Δ^2-cholestene 1 with silver acetate, iodine and moist acetic acid. The reaction probably involves formation of a cyclic 2α, 3β-iodonium ion which on acetolysis with inversion at C-2 gives 5. Expulsion of iodide ion with inversion at C-3 forms a resonant oxonium-carbonium ion 6 which leads to a mono-acetate which on hydrolysis gives 7. As the diol 7 contains one axial substituent at C-2, it is epimerized by treatment with sodium in ethanol at 180° to the diequatorial 2α, 3β-diol 8. Diol 8 was also obtained from cholestenone 9 which reacts with lead-

tetracetate to give in about 10% yield a product 10 having 2α-acetoxy group[5,6]. Reduction of 10 with sodium borohydride followed by hydrogenation gives the 2α, 3β-diol 8.

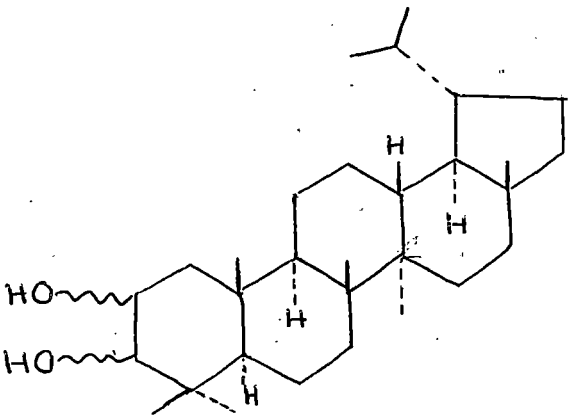


Section B: Synthesis of 2,3-diols of lupane and friedelane series.

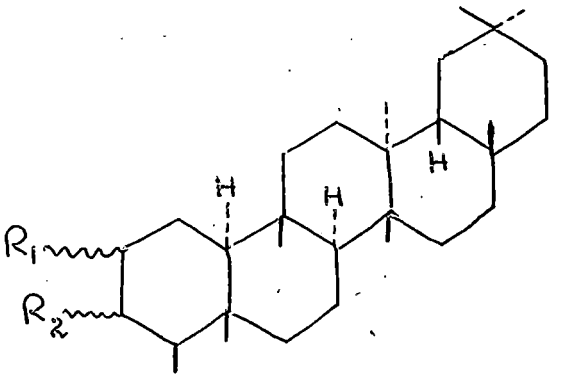
McGinnis[7] et al have reported the preparation of all the four possible stereoisomeric lupane 2,3-diols 12. Samson et al[8] have synthesised 2α, 3α-13A; 2β, 3β-13B; 2α, 3β-13C friedelane diols and shown that the last named isomer is identical with natural pachysandiol A[9] 11.



11



12 (A, B, C, D)



13A $R_1 = R_2 = \alpha$-OH

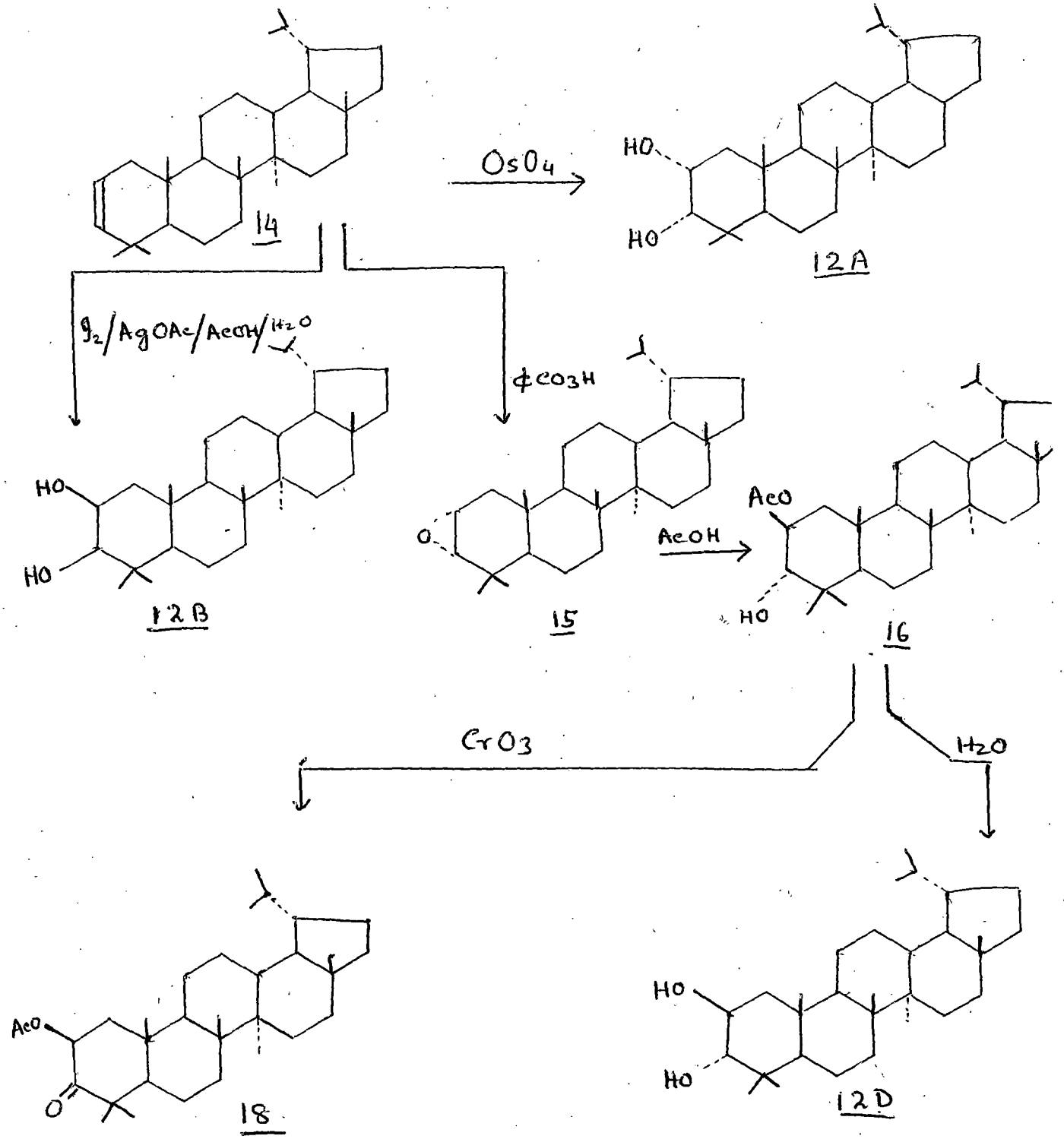
13B $R_1 = R_2 = \beta$-OH

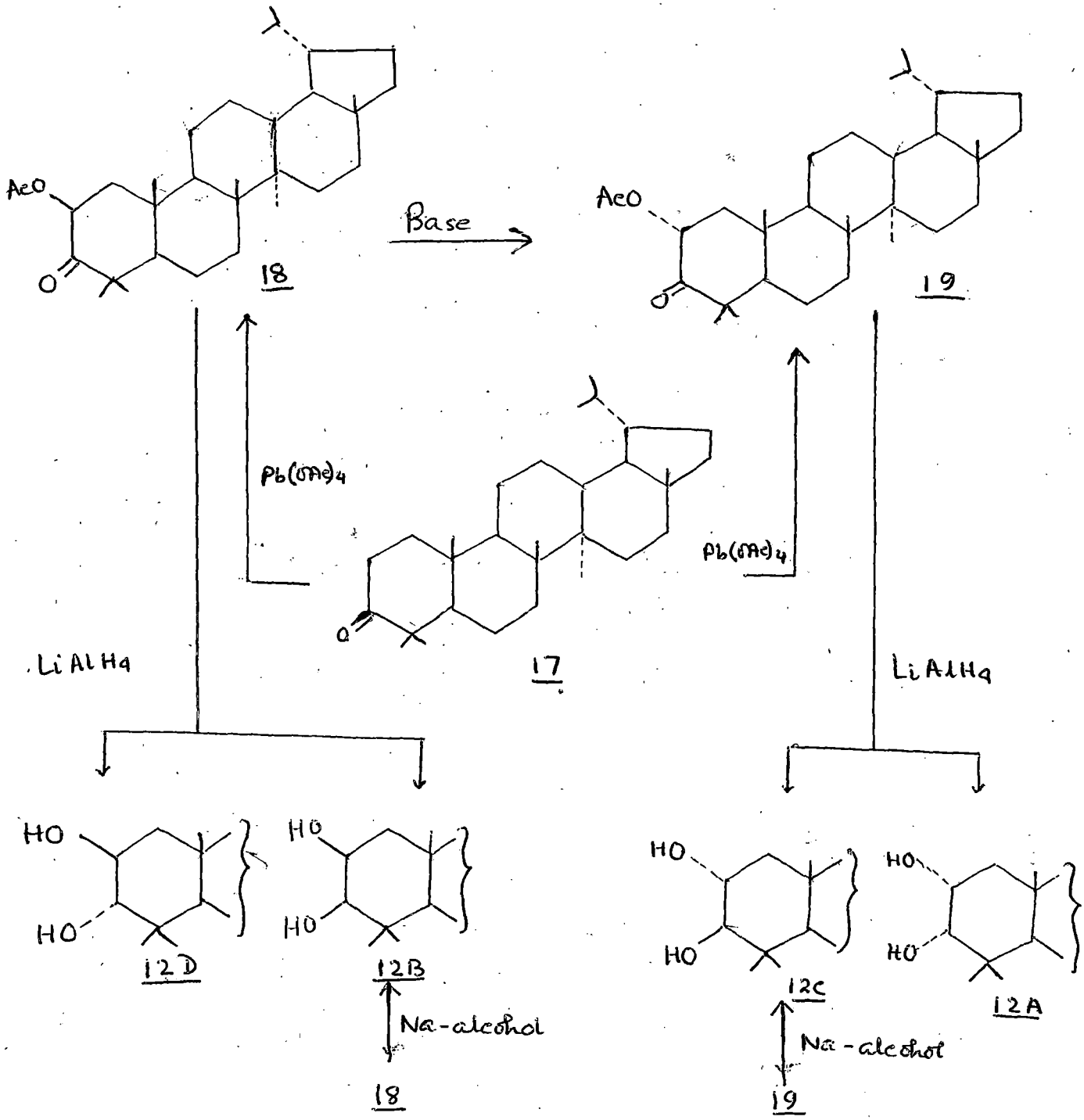
13C $R_1 = \alpha$-OH ; $R_2 = \beta$-OH

Synthesis of Lupane-2, 3-diols[7] 12 (Chart I)

Lup-2-ene 14 was converted to lupane-2α,3α-diol 12A, lupane-2β, 3β-diol 12B and lupane 2α, 3α-epoxides 15 by the action of osmium tetroxide, iodine-silveracetate-acetic acid and perbenzoic acid respectively. The epoxide 15 was opened with acetic acid to give 2β-acetoxy-lupane-3α-ol 16 which on mild alkaline hydrolysis yielded the diaxial trans diol, lupane 2β, 3α-diol 12D. In order to increase the poor yield of the 2β, 3β-diol 12B obtained by the above procedure and also to prepare the remaining isomer: lupane 2α, 3β-diol 12C, lupane-3-one 17 was treated with lead tetracetate to give 2β-acetoxy-lupane-3-one 18 and 2α-acetoxy-lupane-3-one 19 as the major and minor products respectively. The 2β-acetoxy isomer 18 could also be prepared by the chromic acid oxidation of 2β-acetoxy-lupane-3α-ol 16. This acetoxy ketone 18 upon equilibration with base furnished the 2α-acetoxy-3-ketone 19 which was then reduced with lithium aluminium hydride mainly to lupane-2α, 3α diol 12A and with sodium and isopropanol to lupane-2α, 3β-diol 12C. The 2β-acetoxy-3-ketone 18 was similarly reduced with lithium aluminium hydride to give the 2β, 3α-diol 12D as the main product and with sodium and isopropanol to the 2β, 3β-diol 12B.

CHART I

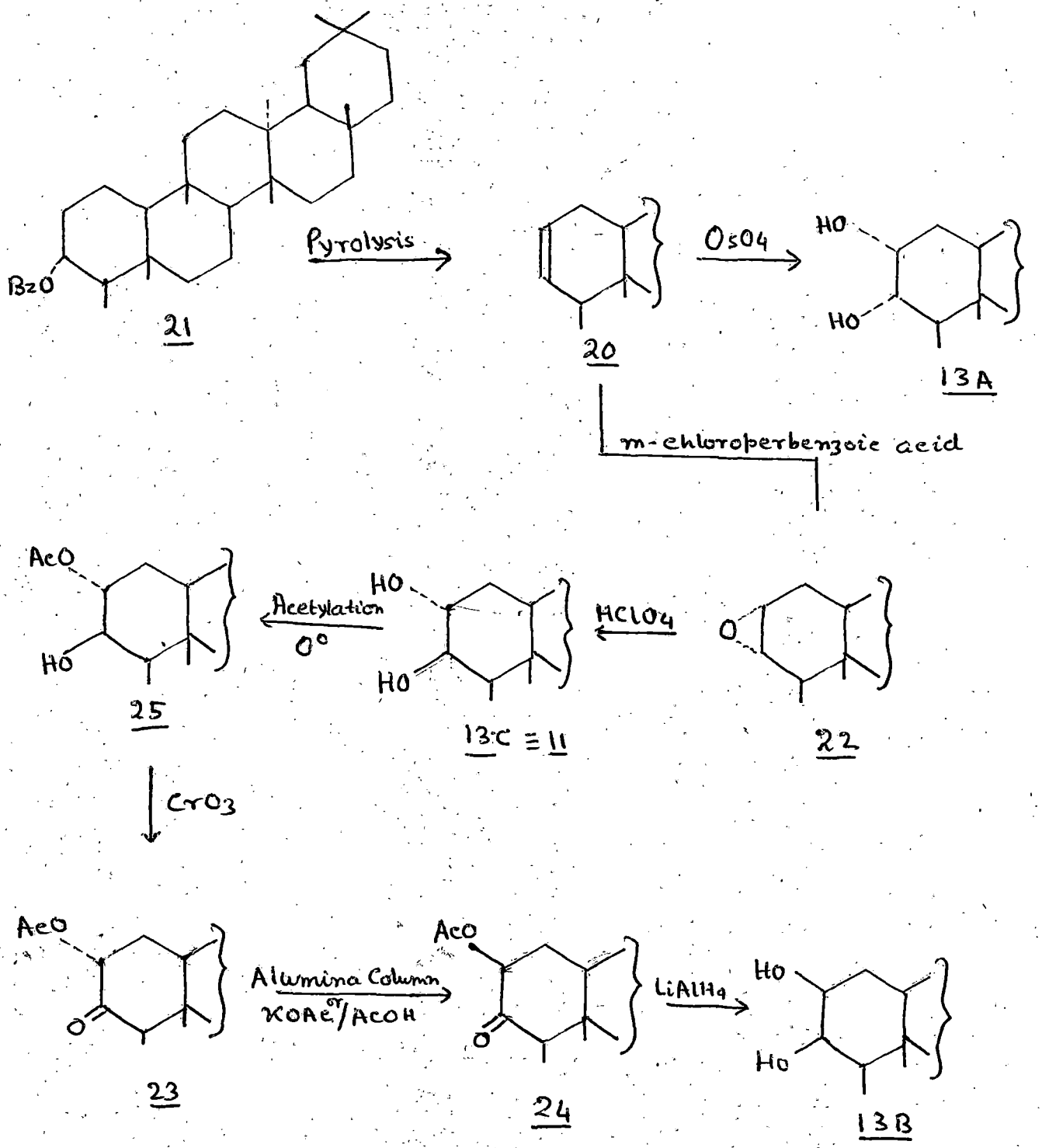

OsO4
HO
HO
12A
14
I2/AgOAc/AcOH/H2O
HO
HO
12B
MCO3H
O
15
AcOH
AcO
HO
16
CrO3
H2O
AcO
O
18
HO
HO
12D


AcO
O
18
Base
AcO
O
19
Pb(OAc)4
O
17
Pb(OAc)4
LiAlH4
LiAlH4
HO
HO
12D
HO
HO
12B
Na-alcohol
18
HO
HO
12C
HO
HO
12A
Na-alcohol
19

Synthesis of Friedelane-2α, 3α 13A; 2β, 3β -13B; 2α, 3β -13C-diols[8]:(Chart II)

Friedel-2-ene 20, obtained by the ~~hydrolysis~~ pyrolysis of friedelanol benzoate 21 was converted to friedelane-2α, 3α-diol 13A by the action of osmium tetroxide and a 2, 3-epoxide 22 by the action of m-chloroperbenzoic acid. The 2,3-epoxide 22 was opened with perchloric acid to yield friedelane 2α, 3β -diol 13C identical with naturally occuring pachy-sandiol A[9] 11. Kikuchi and Toyoda[9] has suggested that cerin acetate was 2α-acetoxy friedelan-3-one 23, contrary to its previous formulation as 2β-acetoxy-friedelane-3-one 24 on the following grounds: Paschysandiol A-2-monoacetate 25 obtained by the acetylation of Pachysandiol A 11 at 0°, could be oxidised to cerin acetate 23 with chromic acid. The cerin acetate so obtained, on prolonged absorption on alumina, was isomerized to another 2-acetoxy-3-ketone 24 which must hence be the more stable 2β-(e, equatorial)-acetoxy isomer. Consequently the original cerin acetate must be less stable 2α-(axial) acetoxy isomer 23. Samson et al[8] isomerized cerin acetate 23 with potassium acetate in acetic acid and reduced the resulting 2β -acetoxy-friedelan-3-one 24 with lithium aluminium hydride and thus synthesised friedelane-2β, 3β -diol 13B . (Chart II)

CHART -II


BzO
21
Pyrolysis
20
OsO4
HO
HO
13A
m-chloroperbenzoic acid
AcO
HO
25
Acetylation
0°
HO
HO
13C ≡ 11
HClO4
O
22
CrO3
AcO
O
23
Alumina Column
KOAc/AcOH
AcO
O
24
LiAlH4
HO
HO
13B

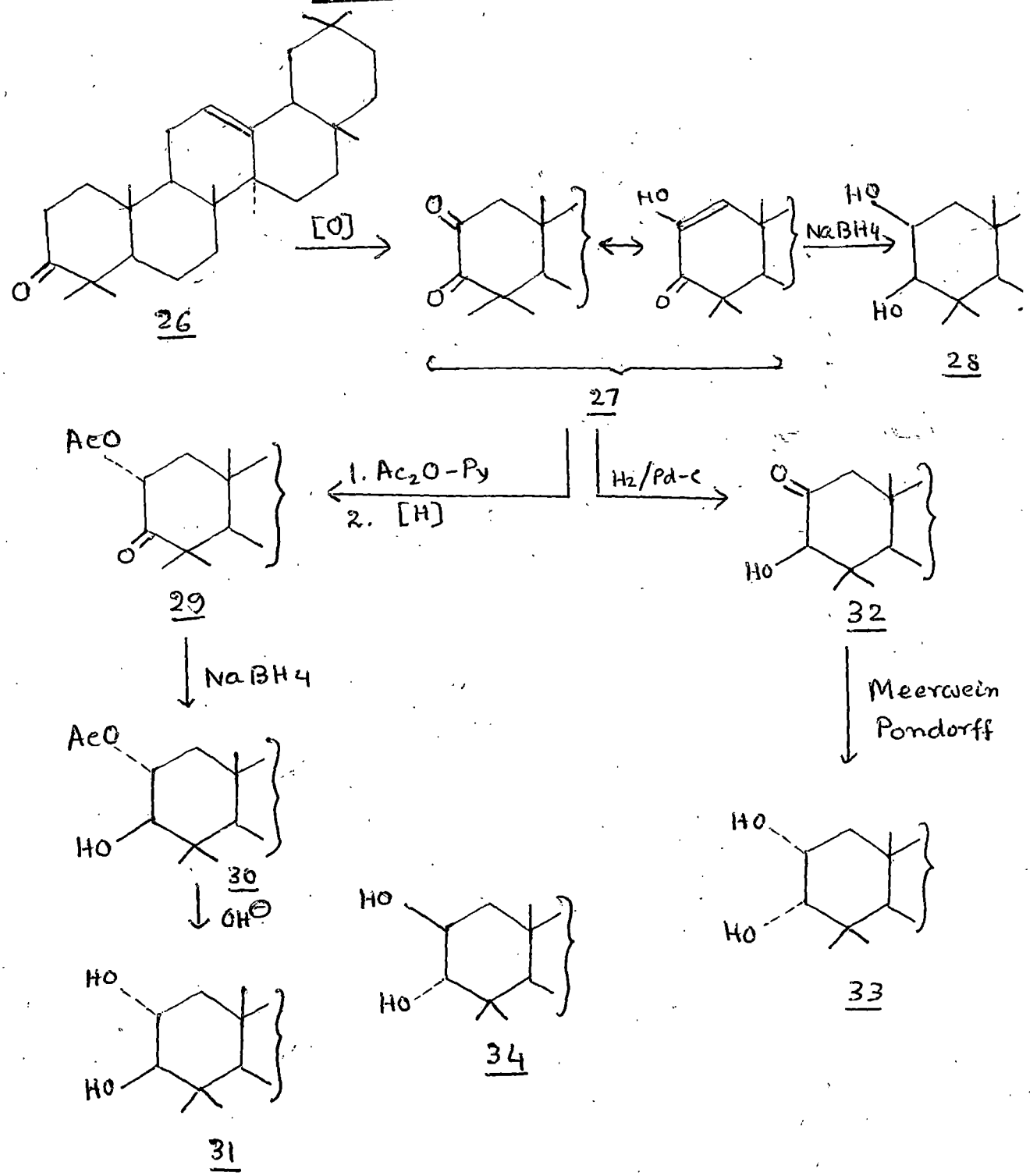
Section C: A short review of synthesis of isomeric Δ^{12}-Oleane-2,3-diols[10]:

A number of 2,3-diols of triterpenoids have been isolated from natural sources. Recently Khastgir et al[10] have synthesised three out of the four isomeric 2,3-diols (Chart IV) by using diosphenol 27 as the intermediate, from β-amyrone 26. Δ^{12}-Oleanene 2β, 3β-diol m.p. 240-42°, $(\alpha)_D$ 101.88° 28 was obtained by sodium borohydride reduction of 27. Acetylation of 27 followed by hydrogenation gave 2α-acetoxy-β-amyrone 29 which on sodium borohydride reduction at pH-8 gave 2α-acetoxy-β-amyrin 30 which on hydrolysis gave the most stable Δ^{12}-oleanene 2α, 3β-diol 31. Hydrogenation of 27 gave 32 which on Meerwein-Pondorff reduction afforded Δ^{12}-Oleanene-2α, 3α-diol, 33 having m.p. 278-81°, $(\alpha)_D$ 71.28°. The sterically most unstable 2β, 3α-diol 34 was also synthesised. The configurations assigned have been confirmed from NMR spectral evidences. The melting points and rotations of the isomeric diols, their diacetates and their acetonide derivatives have been shown in Chart III.

Chart III

	DIOL		DIACETATE		ACETONIDE DERIVATIVE	
	m.p.	$(\alpha)_D$	m.p.°	$(\alpha)_D$	m.p.	$(\alpha)_D$
2β, 3β	240-42°	101.88°	221-22°	83.63°	180-2°	102.50°
2α, 3α	278-82°	71.28°	180-82°	40.77°	199-201°	97°
2α, 3β	202-4°	60.00°	216-18°	73.42°	173-4°	-
2β, 3α	250-52°	12°	161-63°	-	-	-

Chart IV

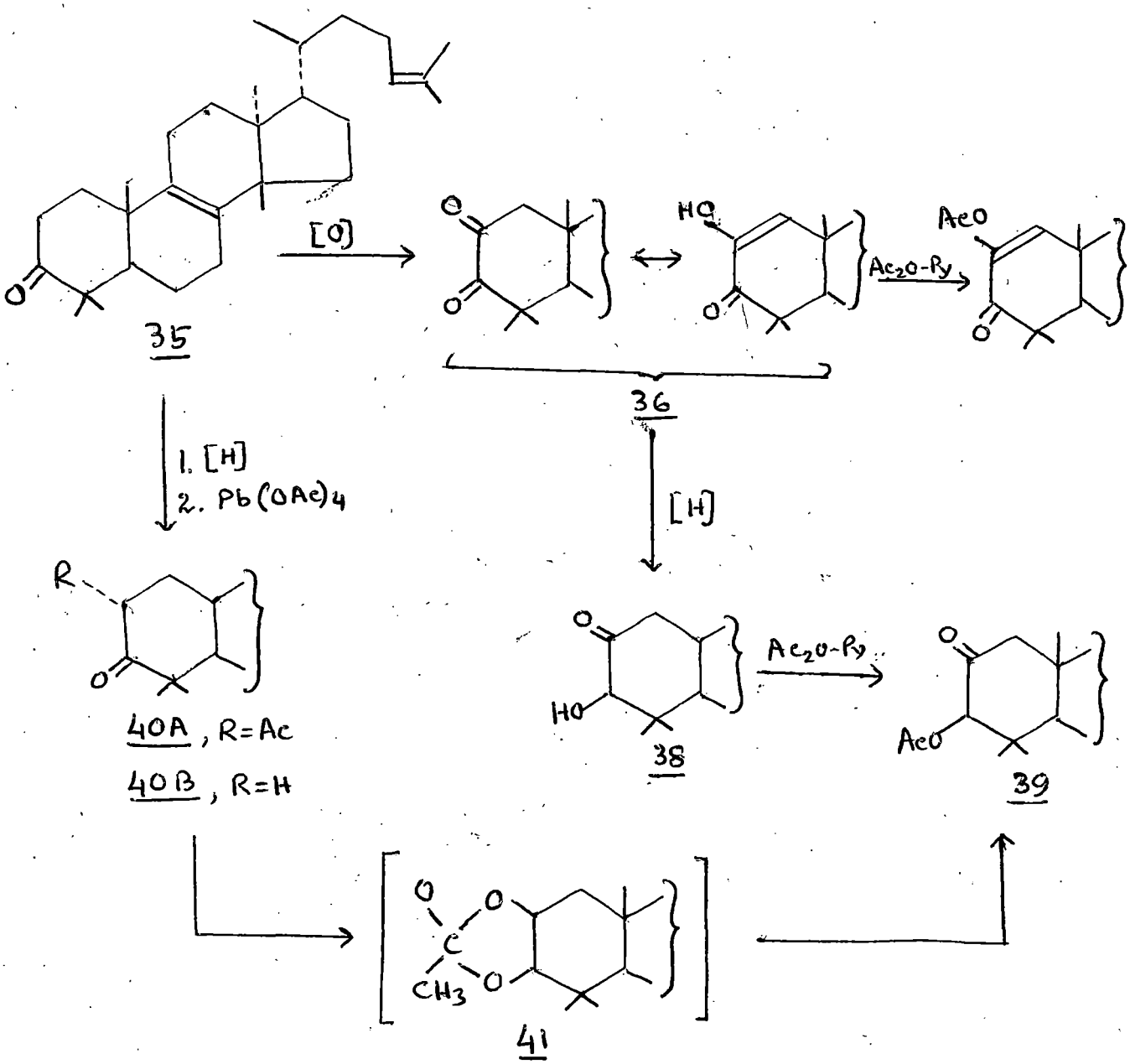

[O]
NaBH4
26
27
28
1. Ac2O-Py
2. [H]
H2/Pd-c
29
32
NaBH4
Meerwein
Pondorff
30
OH⊖
31
34
33

Section D: A short review on autoxidation and isomerization in ring A in triterpenoids :

1. Oxidation in ring A in Euphol.

Lavie and co-workers[11] studied the autoxidation of euphadiene-3-one 35 and the results of their work is summarised in the following lines. Euphadiene-3-one 35 was oxidised by shaking in oxygen in t-butanol saturated with potassium-t-butoxide[12,13]. A tautomeric mixture of diketone and the corresponding diosphenol 36 (two spots on chromatoplate) was produced by absorbing one mole of oxygen UV, λ_{max} 269 (ϵ, 7900), ν_{max} 1715, 1672, and 1653 cm^{-1}. NMR of the compound 36 showed a singlet at τ 3.60 due to vinylic proton at C-1. Acetylation gave the corresponding acetate 37, UV_{max} 236 mμ (ϵ,9000), ν_{max} 1764 cm^{-1}. NMR showed a singlet at τ 3.02 due to C-1 proton. On hydrogenation of the diosphenol 36 over palladium on charcoal (two moles of hydrogen were absorbed-one mole to reduce the side chain double bond and the second mole to reduce the enolic double bond) a non-crystallisable homogeneous solid ν_{max} 1712 cm^{-1}, NMR singlet at τ 5.95 accounting for one hydrogen and two AB type doublets centered at τ 7.69 and τ 7.35 accounting for two hydrogens, was obtained. Upon acetylation a crystalline Ketoacetate was obtained, ν_{max} 1742 and 1730 cm^{-1}, NMR singlet at τ 4.95 for one hydrogen and a broad peak at τ 7.50 accounting for two hydrogens. From the above spectral properties structures 38 and 39 were proposed for hydroxy-ketone

and ketoacetate respectively. 2α-acetoxy (equatorial) derivative

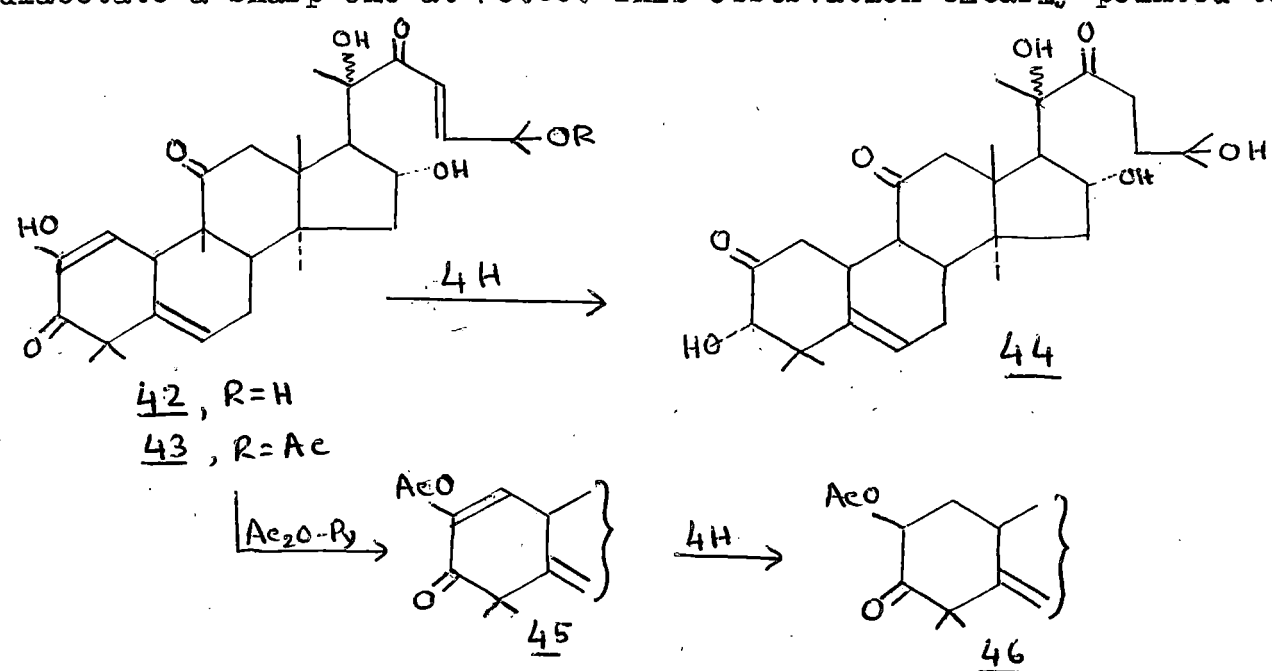


40A was prepared by the reaction of dihydroderivatives of 35 with lead tetra-acetate in acetic acid in presence of boron-trifluoride[14].

The product 40A showed IR bands at 1742 and 1730 cm^{-1} and the NMR spectra showed a quartet of lines centered at τ 4.3 (Jae = 6.5 eps and Jaa = 13.0 eps) for the C-2 proton but no signals for protons α to a keto-functions. The isomerisation of 2α-equatorial acetoxy ketone 40A into the isomer 39 was also observed and they proposed that the migration proceeded through the cyclic intermediate 41[15]. Acid hydrolysis of 40A afforded a compound which has been assigned the 2α-equatorial hydroxy 3-keto derivative 40B on the basis of its IR, ν_{max} 1718 cm^{-1}.

2. Isomerisation in ring A of the Cucurbitacins

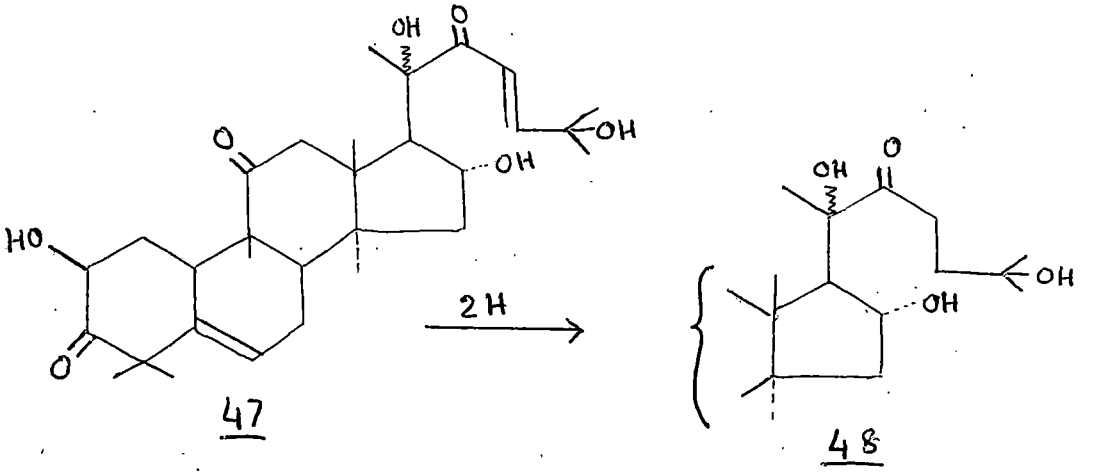
Lavie and co-workers[16,17] reported that hydrogenation of the diosphenol containing cucurbitacins namely elatericin B 42 and elaterin 43 resulted in 1,4 addition of hydrogen during the process of hydrogenation. The NMR spectrum of hydrogenated product of elatericin B was found to show a singlet at τ 6.02 and that of its diacetate a sharp one at τ 5.00. This observation clearly pointed to



the fact that the proton linked to the carbon to which the acetoxy group is also attached had no neighbouring protons and can not therefore be at C-2. The NMR spectra could be explained, if it was considered that 1, 4-addition of hydrogen to the diosphenol system took place, resulting in the conversion of Δ^1-2-hydroxy-3-keto to a 2 keto-3 hydroxy system 44.

Elatericin B diacetate 45 on hydrogenation formed the 2β-equatorial-acetoxy-3-keto derivative 46 by a normal 1,2-addition of hydrogen. This compound showed a quartet of lines related to the 2α-axial proton which is centered at τ4.4 (Jaa = 13.5 eps; Jae = 5.1 eps). The isomerisation of 2-acetoxy-3-ketoderivative 46 on a basic column of alumina as well as on an acedic column was studied. In both the cases the material recovered from the columns showed that it had remained unchanged.

The ORD curves of dihydro elatericin A 48 and tetrahydro elatericin B 46 were also interpreted. Cotton effect curves of both 2 and 3-keto derivatives were found to be positive with the amplitude



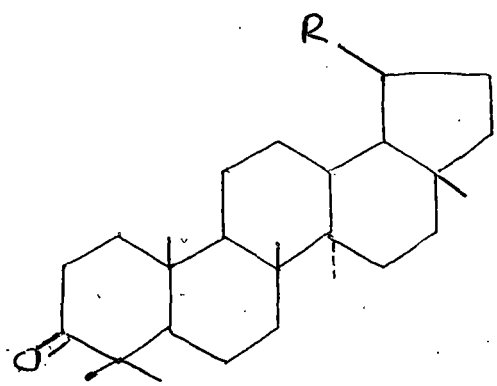
47 48

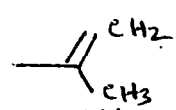
of the 2 keto-derivative being larger than that of the 3-keto form. ORD studies on 2 and 3 ketosteroids[18] and the oxomanoyl oxide series[19] also revealed the same result. The inverted stereochemistry of cucurbitacins at C-10 resulting in a mirror image of the C-10, β - analogy, should give rise to a negative Cotton effect but instead the two compounds displayed positive curves. This can be interpreted as due to the presence of two additional carbonyl chromophore, one in particular at C-11 displaying a large amplitude, which counteracts thereby the inverted rotation of the keto group in ring A as should be expected. The result is a lower positive value instead of a negative one. The peak for dihydro elatericin A 48 (3 keto) at $(\alpha)_{325}+2200^{o}$ is larger, than that of tetrahydroelatericin B 44 (2 keto) $(\alpha)_{325}+1558^{o}$. In both the cases the keto group was flanked by an equatorial (OH) substituent which is either likely to increase the Cotton effect or to render no change at all.

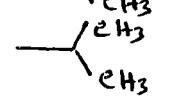
In order to obtain pure tetrahydroelatericin B 45, alkaline hydrolysis of tetrahydroelatericin B diacetate 46 was attempted but the reaction resulted in the formation of dihydroelatericin B[20] 47 λ_{max} 267 mμ (∈, 5700), positive ferric chloride coloration (characteristic of diosphenol). Tetrahydroelatericin B diacetate 48 on alkaline hydrolysis yielded the same dihydroelatericin B 47. The alkali induced autoxidation of α-hydroxy ketone in elatericin was also studied[21] and was found to occur at much slower rate.

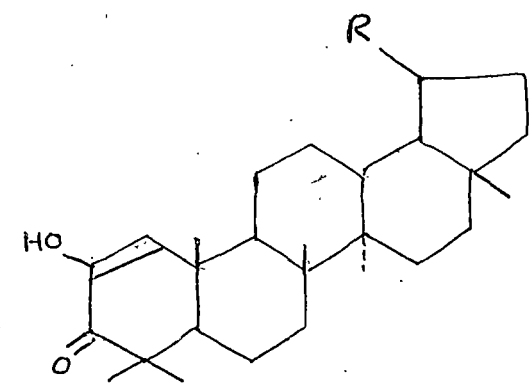
3. Oxidation in ring A in Lupeol

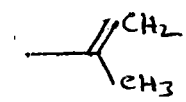
Ganguly and co-workers[22] carried out the oxidation of lupenone 49 and lupanone 50 to the corresponding diosphenols 51 and 52 respectively by passing oxygen in dry t-butyl alcohol containing potassium tertiary butoxide. Diosphenol 52 on hydrogenation afforded a non-crystalline alcohol which on acetylation yielded the keto-acetate 53. The structure 53 was assigned to the keto-acetate by examining its NMR spectra (a sharp singlet at δ, 4.95) ascribed to the C-3 proton.

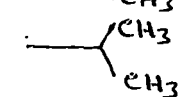


49, R= 

50, R= 



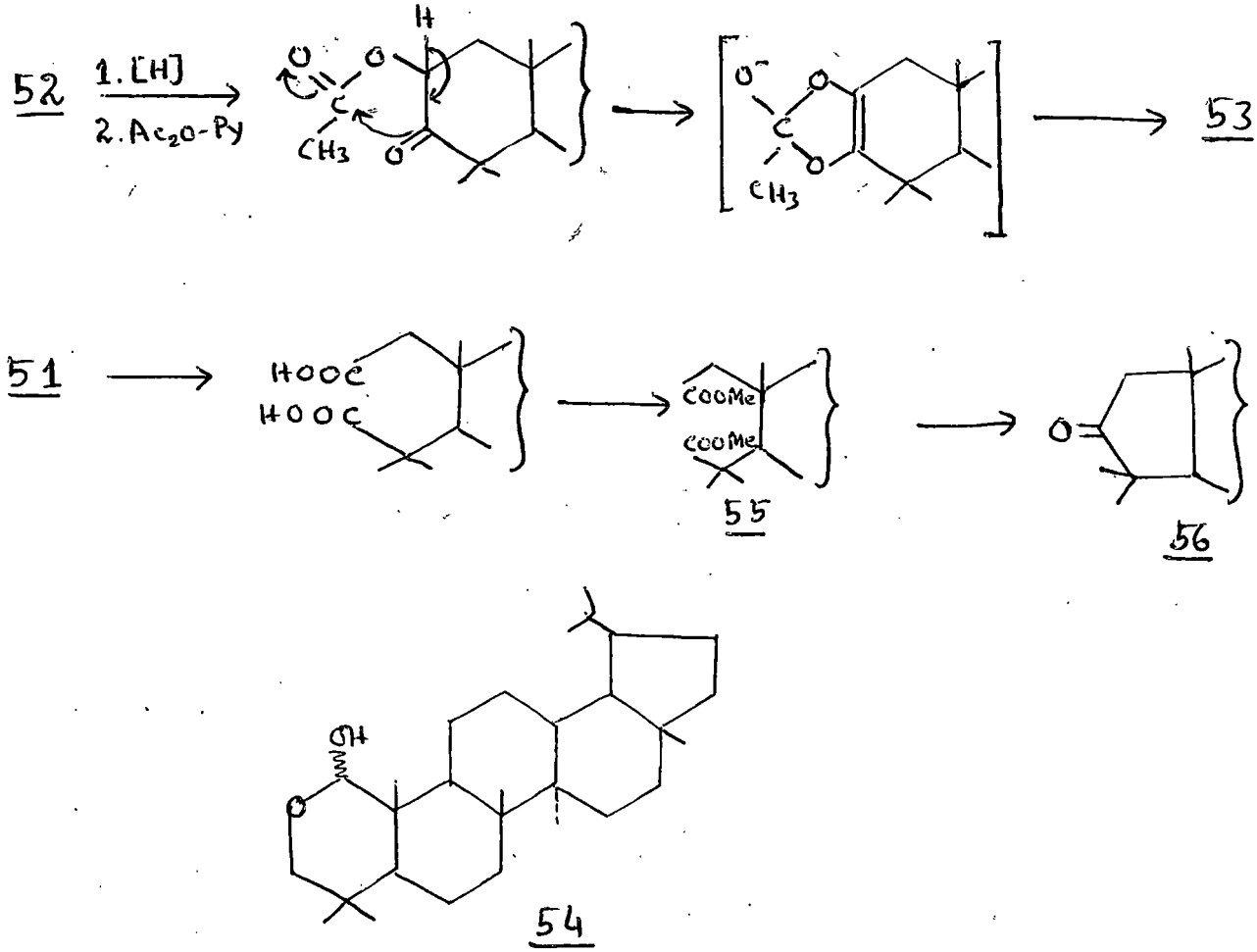
51, R= 

52, R= 

53

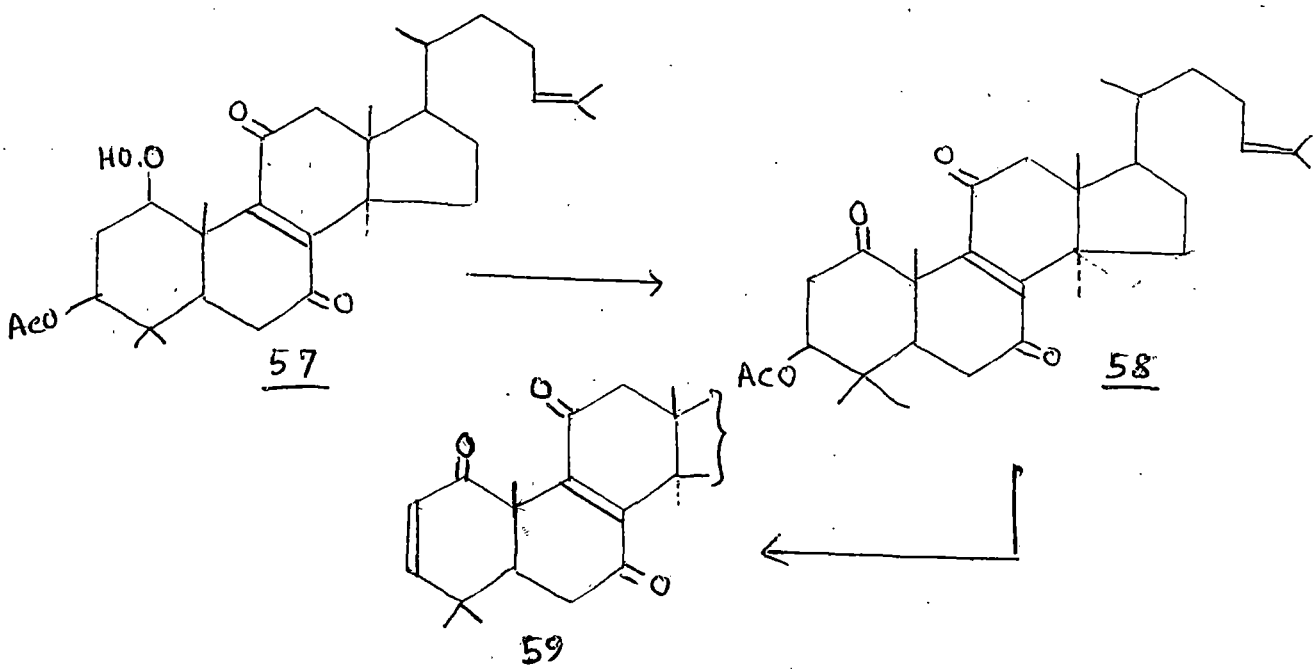
Formation of 53 from 52 was explained by the mechanism shown in Chart V. Diosphenol 52 on ozonisation gave a neutral compound $C_{29}H_{48}O_3$, whose structure was assigned as 54 on the basis of mode of formation, spectral characteristics and elemental composition. Diosphenol 51 was cleaved by alkaline hydrogen peroxide to the dicarboxylic acid $C_{30}H_{48}O_4$. The acid was converted into the dimethyl ester, 55 which on refluxing with alcoholic alkali yielded a neutral crystalline compound 56.

Chart-V



4. Autoxidation of Lanostenyl acetate

Horn and Ilse[23] stated that lanostenyl acetate in ethyl acetate was extensively converted into a mixture of 7-hydroperoxy and 7, 11-dihydroperoxy lanostenyl acetates by treatment with gaseous oxygen at 50° for 48 hours. After that Scotney and Truter[24] found that the autoxidation of lanostenyl acetate in ethyl acetate at 50° after 14 days was a mixture of at least eight peroxides (laminar chromatography). The two most plentiful peroxides were recovered and shown to be 7β-and 11β- hydroperoxy lanostenyl acetates. The structure of 7β-hydroperoxy-lanostenyl acetate was obtained by reducing it with sodium borohydride to 7β-hydroxylanostenyl acetate. The structure of 11β-hydroperoxide was proved by converting it to 11-oxo-lanostenyl acetate with ferrous ion. Furthermore, lithium aluminium hydride reduction of the 11-hydroperoxide afforded one product, which was identical with 11β-hydroxylanostenol.



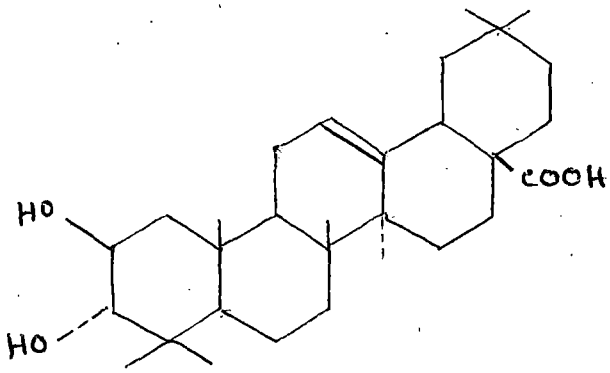
Autoxidation of 7,11-dioxolanost-8-enyl-3β-acetate in cyclohexane at 40° proceeded via 1β-hydroperoxy-7, 11-dioxolanostenyl acetate to 1, 7, 11-trioxolanost-8-enyl acetate[25]. The location of ketone at 1-position was deduced from the behaviour of the trione acetate with alkali. With alkali 1, 7, 11-trixolanost-8-enyl acetate yielded 1,7,11-trioxolanosta-2,8-diene and it had been derived from the trione acetate by elimination of the 3β-acetate group and the formation of a conjugated unsaturated grouping (57, 58, 59). That the precursor for the trione is a mono-hydroperoxide of 7,11-dioxolanostenyl acetate was established by the fact it was decomposed by ferrous ion to 1,7,11-trioxolanostenyl acetate.

In an experiment a solution of lanost-8-en-3βyl acetate in cyclohexane at 40° was oxidised by passing oxygen through it[26]. After twelve months treatment the neutral fraction was examined and was found to contain at least sixteen components. From the R_f values several components have been identified e.g. 1, 7, 11-trioxolanostenyl acetate, 1,7,11-trioxolanosta-2,8-diene. Besides these 15β-hydroxy-7-oxo, 15α-hydroxy-7-oxo, 7, 15-dioxo-and 11, 15-dioxolanostan 3β-yl acetate were also identified.

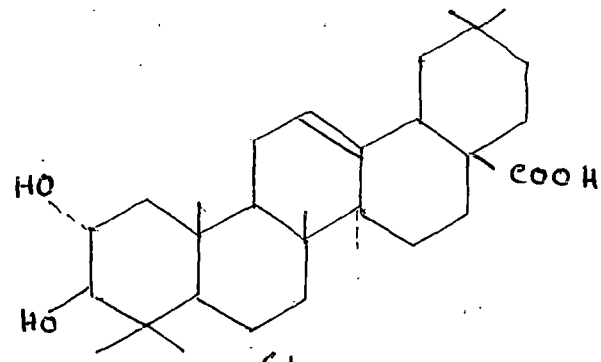
5. Oxidation of ring A in oleanolic acid.

In connection with their work to confirm the structure of bredemolic acid 60 and crategolic acid 61 Tschesche and coworkers[27,28] performed the autoxidation of ring A in methyl oleanonate 62. Methyl oleanonate was stirred in t-butanol containing

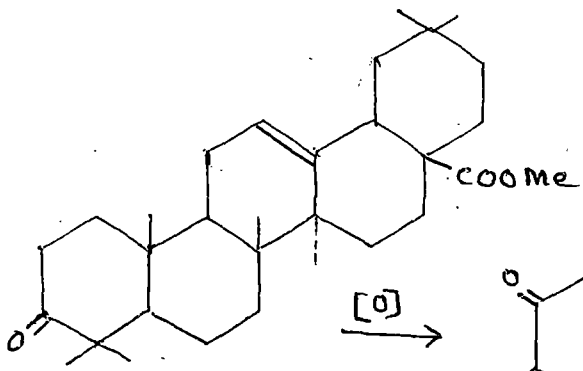
potassium metal at 25-50° with simultaneous introduction of oxygen. The reaction mixture on acidification and usual working up gave an amorphous solid for which structure 63 was proposed. The diosphenol 63 m.p. 130-35°, $(\alpha)_D$ 104 ± 4° on sodium borohydride reduction gave 2β, 3β-dihydroxy-12-en-olean-28-oate 64 which on oxidation with kilani solution gave a mixture of several compounds in which 10% of 63 was found to be present as was shown by its UV spectrum.



60

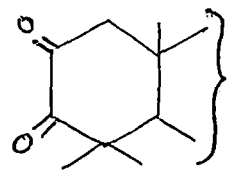


61



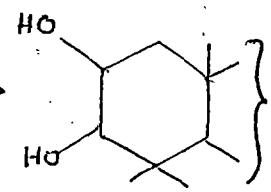
62

[O]



63

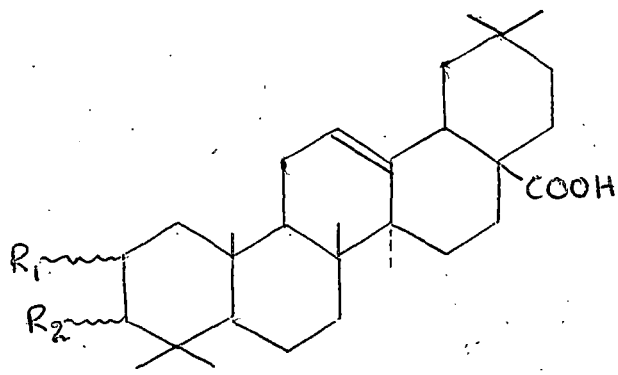
NaBH4



64

Section D: A short review on 2,3 -dihydroxy triterpene acids from natural sources:

The four stereo-isomeric 2,3-dihydroxy olean-12-en-28-oic acids are known to occur in nature viz: (1) the 2α, 3α -dihydroxyolean-12-en-28-oic acid 65A[29] (2) the 2α, 3β -dihydroxyolean-12-en-28-oic acid (crategolic/maslinic acid) 65B[30] (3) the 2β, 3β -dihydroxy olean-12-en-28 oic acid[31] 65C (4) the 2β, 3α-dihydroxy olean-12-en-28-oic acid 65D[28] (bredemolic acid).



65A $R_1 = R_2 = \alpha$-OH
65B $R_1 = \alpha$-OH, $R_2 = \beta$-OH
65C $R_1 = R_2 = \beta$-OH
65D $R_1 = \beta$-OH, $R_2 = \alpha$-OH

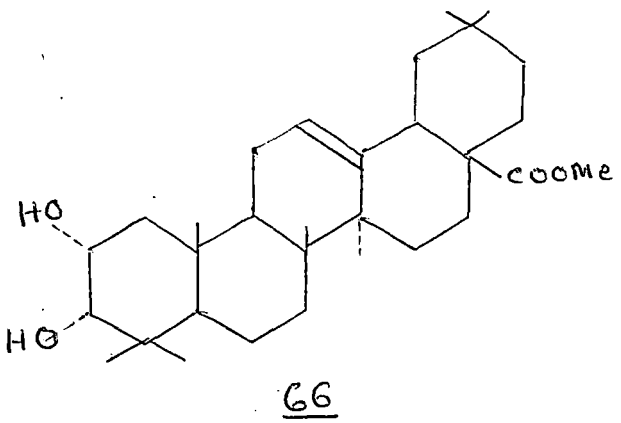
Alphitolic acid[33] in the lupeol series and 2α-hydroxy ursolic acid[34] in the ursane series are known to occur in nature.

1. The 2α, 3α-dihydroxy olean-12-en-28-oic acid:

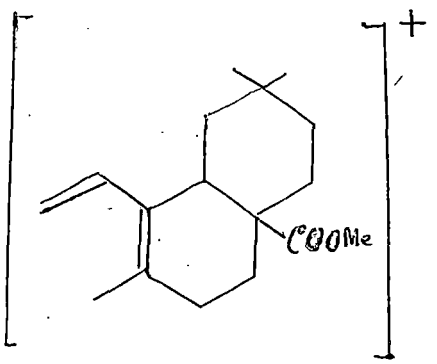
Cheung et al[29] isolated a triterpene acid 65A as its methyl ester m.p. 296-99° from shorea accuminata resin, which has been shown to be the 2α, 3α-dihydroxyolean-12-en-28 oic acid. The methyl ester 66 (ν_{max} 3340, 1725 cm^{-1}) formed a diacetate, a monoacetate and a O, O-isopropylidene derivative indicating the presence of two hydroxyl groups. All these compounds showed NMR signals due to

methyl ester group (3H, singlet δ 3.6) and a triplet for an olefinic proton (J 4Hz, δ 5.3).

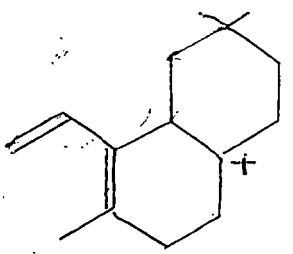
The mass spectrum of the diol methyl ester or its acetonide showed intense peaks at m/e 262, 203 corresponding to ions A and B[35]



66



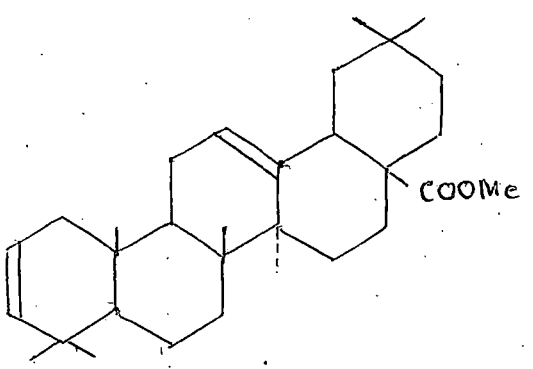
A



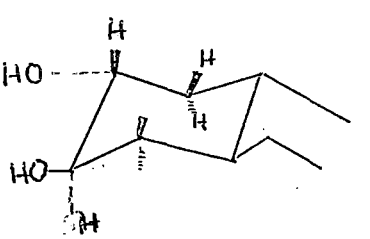
B

The mass fragmentation pattern established that the diol methyl ester belongs to 12-oleanene or 12-Ursene series with a 28-methoxycarbonyl group. They concluded from the NMR signal[36] of the allylic 18β-hydrogen at δ 2.8 (AB quartet), that the diol ester belonged to oleanene group. Further evidences for assignment of a 2α, 3α-configuration of the diol is supported by the following observations. In this diol, the C-3 proton showed a doublet (J 3Hz) at δ 3.35 due to vicinal coupling of equatorial and axial protons. Upon saturation (by double irradiation) of this signal, the multiplet near δ3.9 due to the proton at C-2 simplified to a four-line signal characteristics of X part of an ABX type spectrum.

The cis isomers, 2α, 3α-diol and 2β, 3β-diol were first prepared[37] by Djerassi et al[37] by osmium tetroxide oxidation of methyl oleana-2, 12-diene-28-oate 67. Cheung et al also repeated the oxidation and obtained two cis-diols and the one with higher melting point was identical to the methyl ester of m.p. 296-99°, isolated from shorea acuminata. Tschesche et al[27,28] assigned a 2β, 3β-configuration to this diol and an 2α, 3α-configuration to the one with lower m.p. (258-60°), from consideration of the infrared absorption due to O-H stretching. Cheung et al demonstrated that the configurations assigned by Tschesche et al should be reversed. By comparing the methyl resonance frequencies from published substitution effects[36] with those observed for the two cis diols and their acetate derivatives Cheung et al suggested that, contrary to the views of Tschesche et al[27,28], the diol m.p. 296-99° must have the 2α, 3α-and the diol m.p. 258-60° the 2β, 3β-configuration.



67



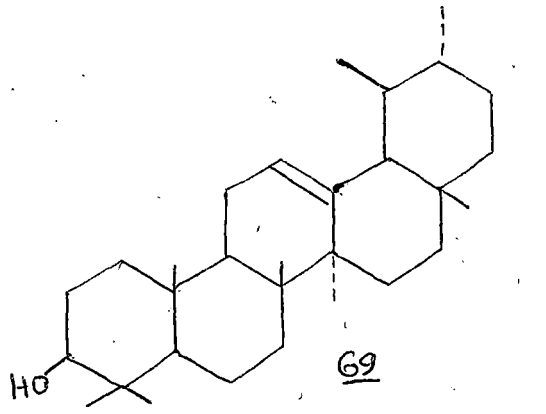
68

In the 2α, 3α-diol, with a chair ring A, 68, the 2β-proton is axial and is expected[39] to be subject to a large ax-ax coupling with the 1-α proton and to small ax-eq coupling with the 3β and 1β -protons. Of the two cis diols from osmium tetroxide only one with m.p. 296-99°, showed a signal due to C-2 proton of sufficient width at half-height (Wh/2 21Hz) to be compatible with a 2α, 3α-diol structure. The other diol m.p. 258-60° having a corresponding signal of Wh/2 8Hz should have a 2β, 3β- arrangement.

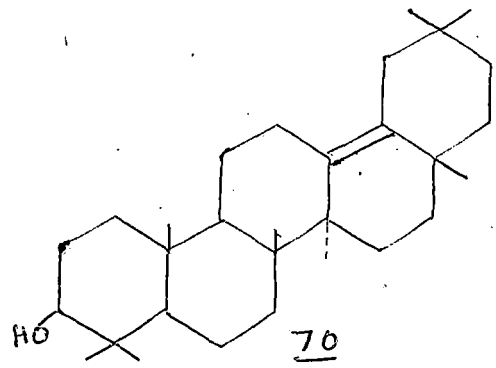
2. The 2α, 3β -dihydroxyolean-12-en-28-oic acid (Crategolic/maslinic acid):

Bachler[40] was the first to isolate an amorphous acid "Crategus acid" from the leaves of Crategus Oxyacantha L . He, however, erroneously assigned the molecular formula $C_{32}H_{54}O_4$ to it. The acid was also observed to occur in the leaves of Psidium guaijava by Arthur and Hui[40]. This acid was subsequently studied by a number of workers[40]. However a more detailed study of the acid was made by Tschesche et al[40,41], who succeeded in obtaining the acid in crystalline form and establishing the correct molecular formula $C_{30}H_{48}O_4$. They designated the acid as crategolic acid, established the presence of a double bond resistant to catalytic hydrogenation and suggested the presence of two hydroxyl groups, although they could not prepare a diacetate. From a consideration of the behaviour of the acid towards acylation, decarboxylation and lactonisation, they erroneously

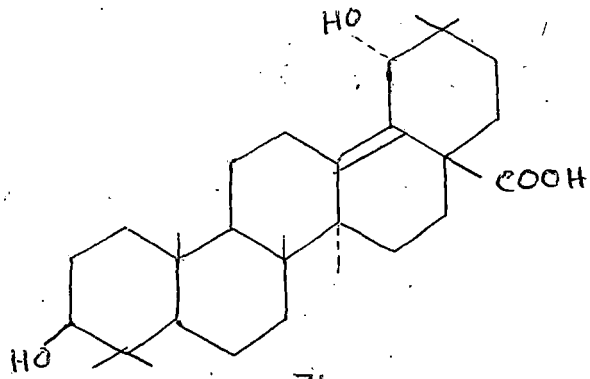
concluded that crategolic acid was an α-amyrin derivative and even suggested the revision of the acdepted structure 69 of α-amyrin to the δ-amyrin structure 70. On the basis of their proposed new formula of α-amyrin 70, they suggested without much valid reason that crategolic acid had the structure 71.



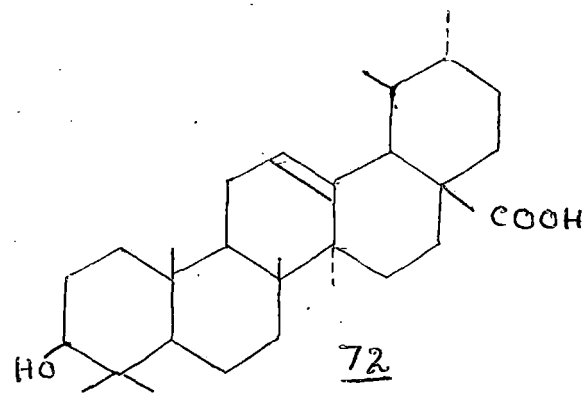
69



70



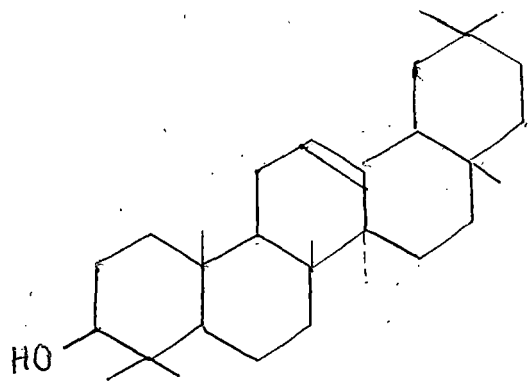
71



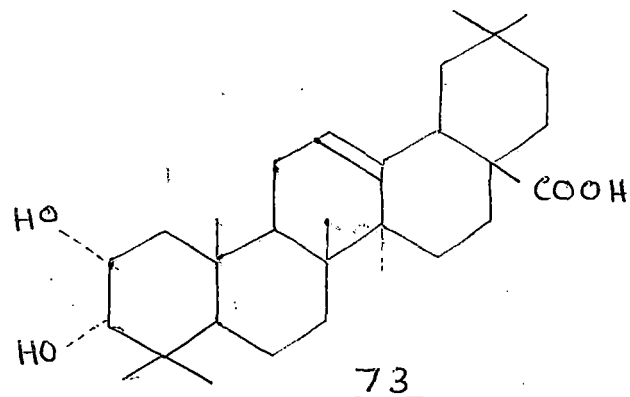
72

However, they themselves latter showed[41] that their 'crategolic acid' was impure, being contaminated with 60-65% of ursolic acid 72 which could not be easily separated. Arthur et al[42] drew attention

to this fact and suggested further work. Tschesche et al in a subsequent paper[43] correctly recognised crategolic acid 73 as a derivative of β-amyrin 74. The impure acid mixture could be



74



73

resolved by them by paper chromatography or column chromatography of the methyl esters derived from it. They were also able to prepare a diacetate, a monoacetate and a keto-monoacetate from methyl ester of crategolic acid 73.

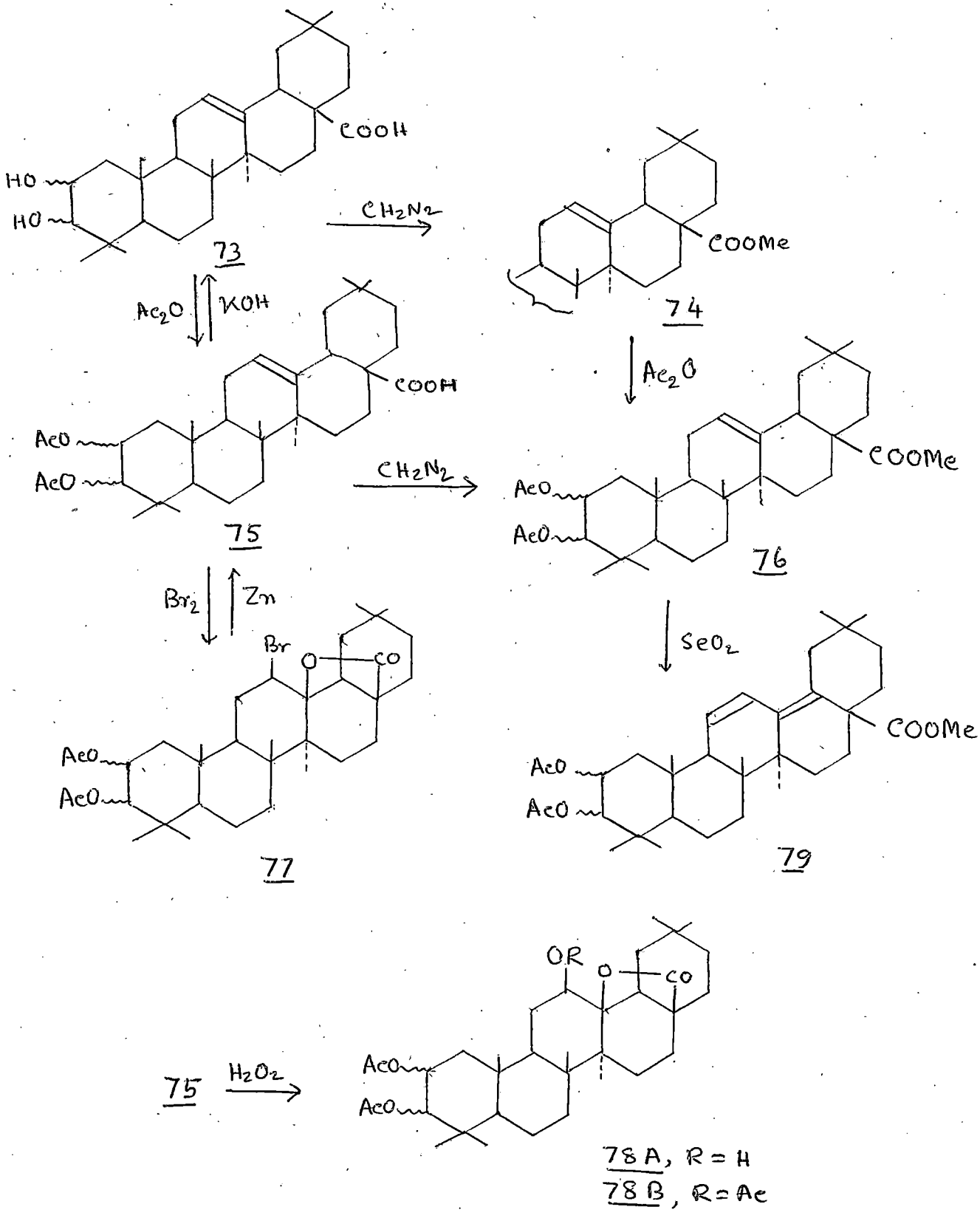
However, in the mean time, Caglioti et al reported[30] the isolation from the cakes of Olean europa of a new acid, maslinic acid, which latter proved identical with crategolic acid 73 of Tschesche et al. The Italian workers[30] were able to show that maslinic acid was a pentacyclic triterpenic acid, probably belonging to the β-amyrin group, containing two acylable hydroxyl groups and a non-hydrogenizable double bond γ- to the carboxyl group.

In their subsequent investigations[44,45] Caglioti et al were

able to elucidate the complete structure of the acid as a 2, 3-dihydroxy olean-12-en-28-oic acid. Their work of structure elucidation of crategolic acid 73 is shown schematically in Chart VI. Crategolic acid 73 formed a methyl ester 74, a diacetate 75 and a methyl ester diacetate 76. The diacetoxy acid 75 with bromine gave a bromolactone 77 which on treatment with zinc and acetic acid regenerated the diacetoxy acid 75. The latter 75 with hydrogen peroxide gave a hydroxy-diacetoxy-lactone 78A which gave a triacetoxy lactone 78B on acetylation. Sclenium dioxide oxidation of the methyl ester diacetate 76 gave a conjugated diene ester 79 showing U.V absorption maxima at 260, 251, 243 mμ, characteristics of β-amyrin derivatives. The presence of an α glycol system was shown by the consumption of one mole of periodic acid of methyl crategolate 74.

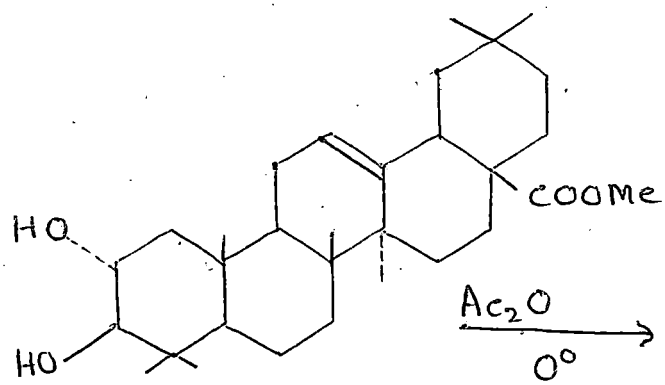
Finally crategolic acid 73 was correlated with the β-amyrin group by its elegant conversion into methyl oleanonate 81 and methyl olean-12-en-28 oate 82. This conversion inter alia settled the position of the two hydroxyl group at C-2 and C-3. When methyl crategolate 74 was acetylated at 0^{o}, the major product was the 2-acetate 79, which could be oxidised with chromic acid to give the 3-keto-2-acetate 80. The latter 80 on treatment with calcium in liquid ammonia was converted to methyl oleanonate 81, a substance of known structure methyl crategolate 74 itself on oxidation with chromic acid followed by Huang Minlon reduction furnished the known ester 82. These transformations are shown schematically in Chart VI.

Chart VI



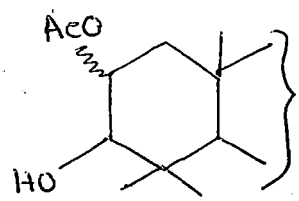
78 A, R = H

78 B, R = Ac



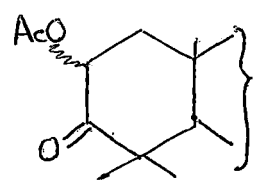
74

Ac_2O, 0°



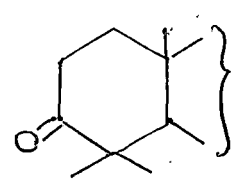
79

CrO_3



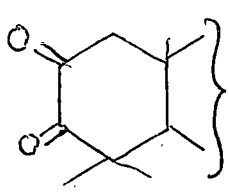
80

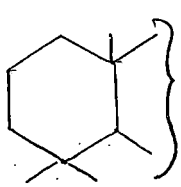
Ca / liq. NH_3



81

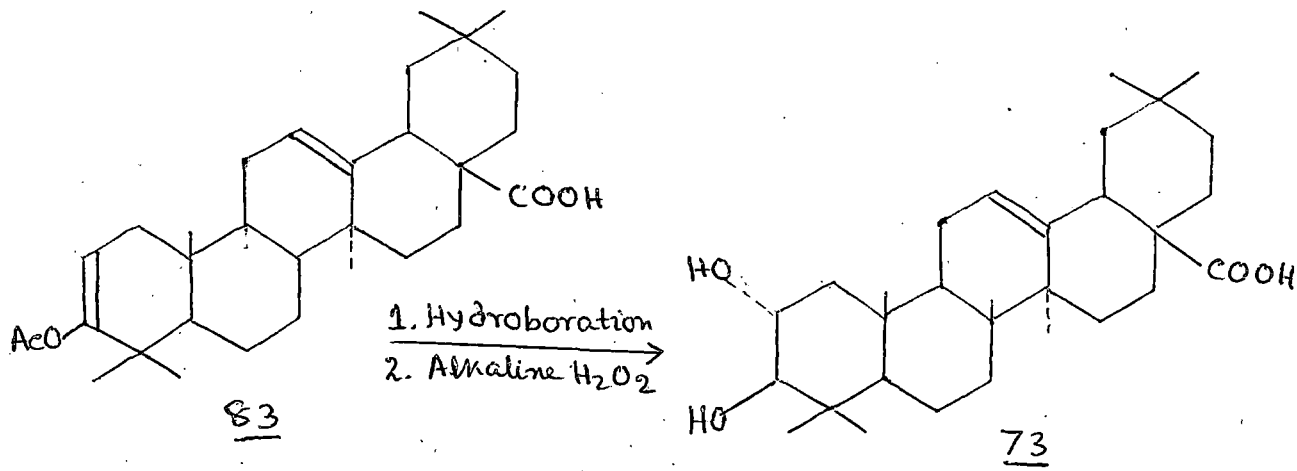
CrO_3





82

Subsequently, the configuration of the two hydroxyl groups was elucidated by Caglioti et al[46]. Preferential formation of the 2-monoacetate 79 suggested the 2α, 3β-trans diequatorial configuration of the diol moiety, which was confirmed by successful synthesis[46] of crategolic acid 73 from the enol-acetate 83 of oleanonic acid by hydroboration, which was known to be a stereospecific process[47,48].



83 73

The correctness of the above assignment of configuration of the two hydroxyl groups of crategolic acid 73 was further supported by the work of Tschesche et al[38] on the structure of bredemolic acid (discussed in page 118, Part II of this thesis).

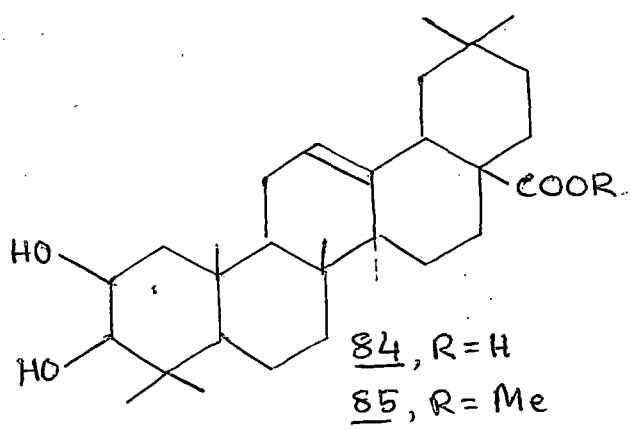
Sengupta et al[49] has also isolated crategolic acid (maslinic acid) 73 from the flowers of Eugenia jambolana Lam as its methyl ester along with oleanolic acid.

The present author has also synthesised methyl crategolate and the details have been discussed on page 163 Part II of this thesis.

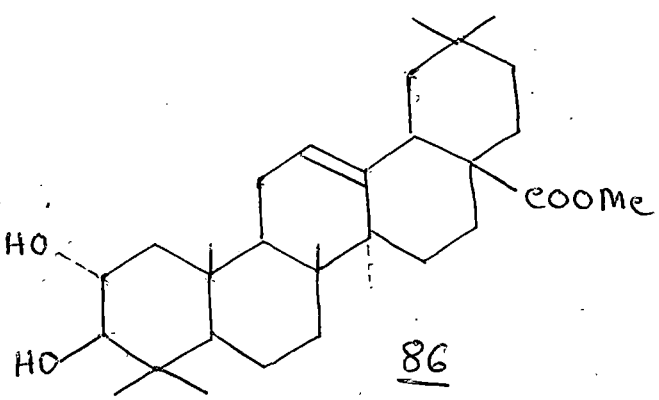
3. The 2β, 3β -dihydroxyolean-12-en-28 oate.

Bannon et al[31] recently reported the isolation of 2β, 3β -dihydroxyolean-12-en-28-oic acid 84 from the sapogenin mixture prepared from the extract of the wood of Castanosperum australe Cunn and Fras. These authors established the identity of 84 (2β, 3β-dihydroxyolean-12-en-28 oic acid) by a high yielding stereospecific synthesis (Chart VII) from methyl crategolate 86.

Bannon et al isolated 84 as its methyl ester 85. The structure was suggested by its IR, NMR, mass spectrum[29b] . The melting point of the methyl ester 276-80°, 85 was in agreement with that published previously for methyl 2β, 3β-dihydroxyolean-12-en-28 oate (lit 278-82°[37] and 276-84°[28]). These authors carried out a stereospecific synthesis of methyl 2β, 3β dihydroxyolean-12-en-28-oate 85 in high yield from methyl crategolate 86 and thereby concluded that the product islated from C. australe is in fact methyl 2β, 3β-dihydroxy-olean-12-en-28 oate.

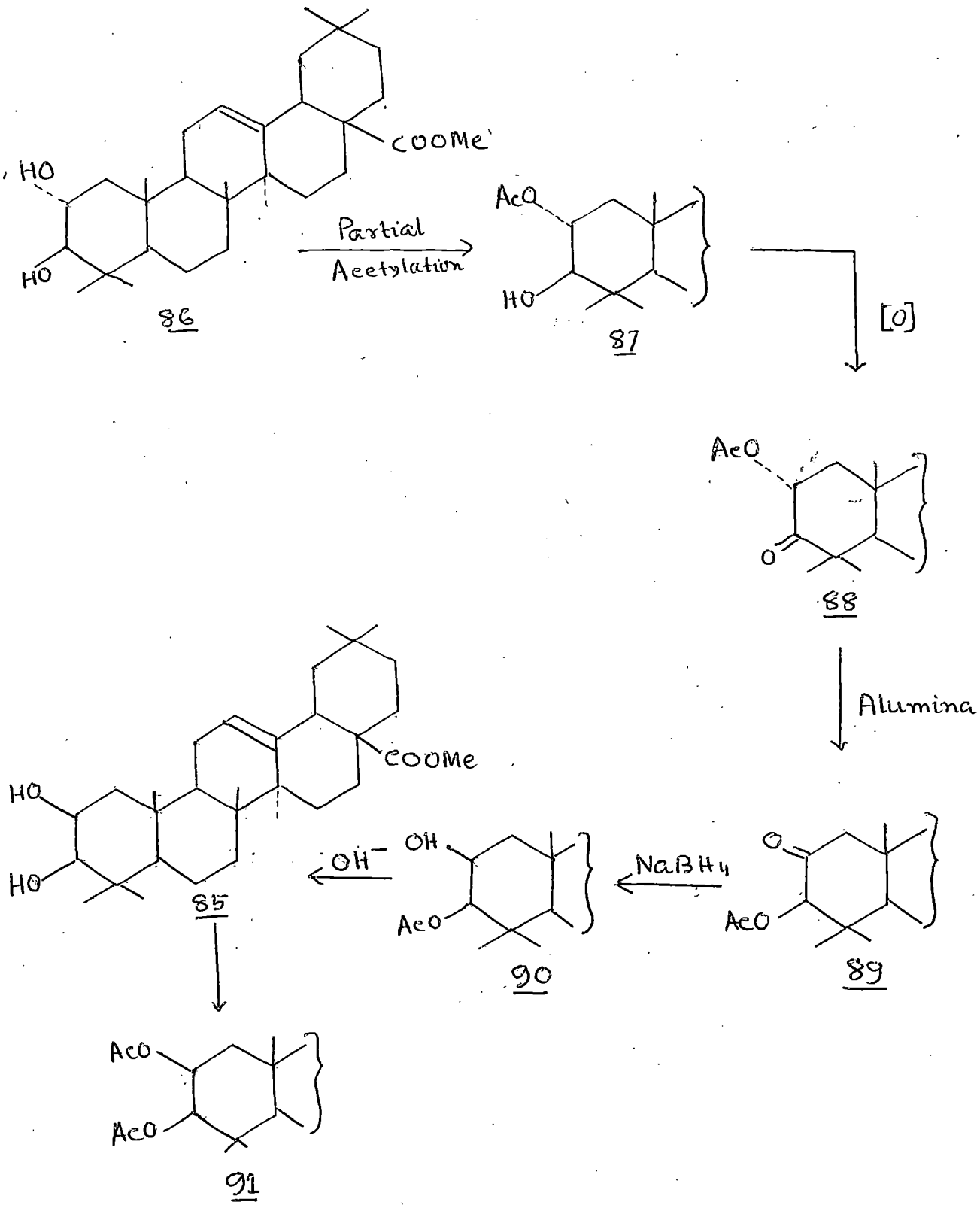


84, R = H
85, R = Me



86

Chart VII


HO
COOMe
HO
Partial
Acetylation
AcO
HO
[O]
AcO
O
Alumina
COOMe
HO
HO
OH⁻
OH
AcO
NaBH4
O
AcO
AcO
AcO
86
87
88
85
90
89
91

Methyl crategolate 86 on partial acetylation yielded the 2α-acetoxy-3β-alcohol[43] 87 which was oxidised with dimethyl sulfoxide in acetic anhydride to give 2α-acetoxy-3-ketone 88. The latter 88 on isomerisation on alumina gave the 3β-acetoxy-2-ketone 89. The structure of this rearranged acetoxy ketone followed from analogy with the rearrangements of similar groups in lupane[51], lanostane and 4, 4-dimethyl cholestane derivatives[52] and from its NMR and IR spectra. Reduction of the 3β-acetoxy-2-ketone 89 with sodium borohydride proceeded quantitatively to give a single product 90 in which the introduced hydroxyl group at C-2 could be assigned the β-configuration on the assumption that attack has occured from less hindered α-side of the molecule. Mild alkaline hydrolysis of 3β-acetoxy-2β-hydroxyolean-12-en-28-oate 90 gave the diol 85 m.p. 278-80°, $(\alpha)_D$ 88 (lit. m.p. 258-60°, $(\alpha)_D$ 97°[43], m.p. 258-62°[28], m.p. 258-62° $(\alpha)_D$ 85°[37], m.p. 258-61°[29b]). 85 on acetylation afforded the diacetate, methyl 2β, 3β-diacetoxyolean-12-en-28-oate 91 m.p. 232-4°, $(\alpha)_D$ 82°, (lit[29b] m.p. 227-31°).

We have also synthesised the 2β, 3β-dihydroxyolean-12-en-28-oic acid from diosphenol of methyl oleanonate (details discussed on page 162 Part II of this thesis).

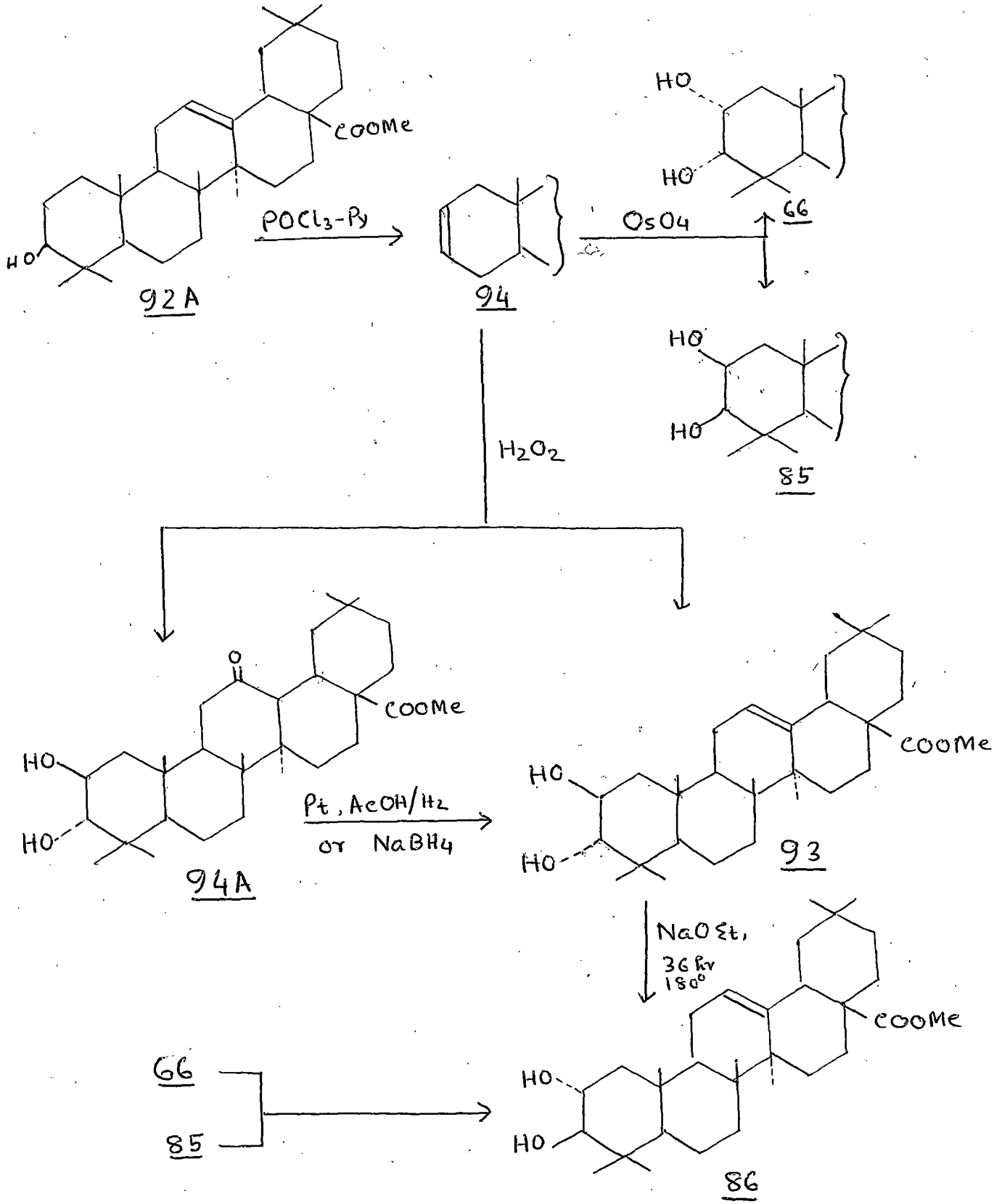
4. The 2β, 3α-dihydroxyolean-12-en-28-oic acid (bredemolic acid):

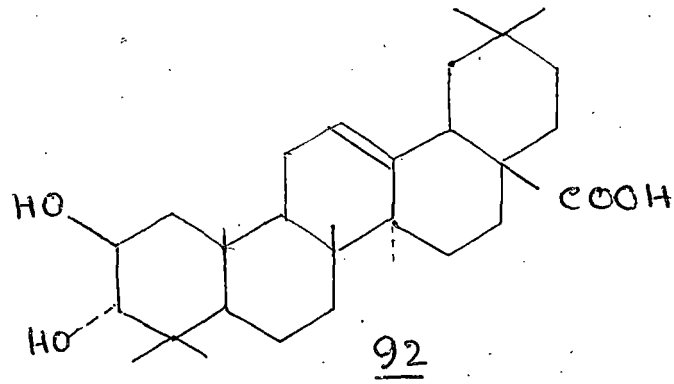
Bredemolic acid 92, isolated from Bredemeyera floribunda willd., was also a 2,3 -dihydroxyolean-12-en-28-oic acid and hence

must be an epimer of crategolic acid 73. Since the 2β, 3β-84, and the 2α, 3α-65A isomers were already known by synthesis[37] and crategolic acid 73 was shown to be the 2α, 3β-dihydroxyolean-12-en-28-oic acid by Caglioti et al[44] bredemolie acid 92 must then be the remaining 2β, 3α-dihydroxy isomer. However bredemolic acid 92 was found to form an acetonide, a somewhat unexpected behaviour on the part of a normal 1,2-diaxial trans cyclohexane diol derivative. Therefore Tschesche et al synthesised[27,28] unambiguously all the four stereoisomeric 2,3-dihydroxy olean-12-en-28-oic acids as their respective methyl esters. The synthetic work of Tschesche et al is shown in Chart VIII.

The key compound in the synthesis of the above epimeric methyl esters Chart VIII; methyl oleana-2, 12-dien-28-oate 94 was prepared from methyl oleanolate 92A by dehydration with phosphorous oxychloride and pyridine. The dien-ester 94 gave two cis diols: the 2α, 3α diol 66 and the 2β, 3β diol 85 by treatment with osmium tetroxide. On the other hand treatment of the diene-ester 94 with hydrogen peroxide gave methyl 12-keto-2β, 3α-dihydroxyolean-28-oate 94A as the major product and only a trace of the desired 2β, 3α-diol 93. The above 12-keto-ester 94A, however, was converted by reduction into methyl bredomolate 93. Finally, all the above three diol-esters 66, 85 and 93 on equilibration with base gave methyl crategolate 86, which must consequently have the stablest diequatorial 2α, 3β-configuration of the two hydroxyl groups.

Chart VIII


COOMe
HO
92A
POCl3-Py
94
HO
HO
66
OsO4
HO
HO
85
H2O2
O
COOMe
HO
HO
94A
Pt, AcOH/H2
or NaBH4
COOMe
HO
HO
93
NaOEt,
36 hr
180°
66
85
COOMe
HO
HO
86

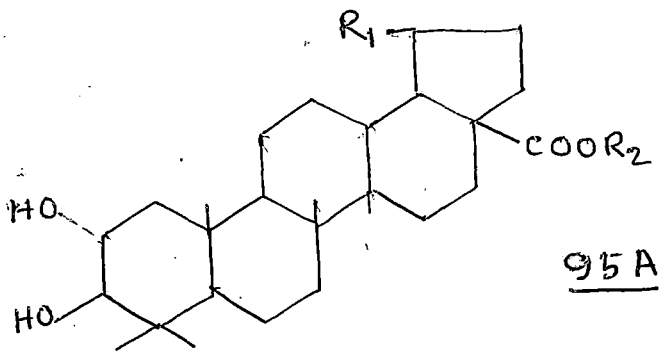


92

5. Alphitolic Acid:

Guise et al[33] isolated alphitolic acid 95A as its methyl ester, m.p. 233-35° from the wood of Alphitonia petrici Braid and White. The ester, methyl alphitolate 95B (ν_{max} 3634, 3587, 1735, 887 cm^{-1}) formed a diacetate (no hydroxyl absorption in the infrared) and a dihydroderivative 95C (no absorption in the infrared at 887 cm^{-1}) namely methyl dihydroalphitolate. The ester 95B consumed one mole of lead tetra-acetate. Guise et al converted dihydromethyl alphitolate 95C to a dialdehyde 96 by treatment with 1 mole equivalent of sodium metaperiodate. The latter on oxidation with chromic anhydride in acetic acid followed by methylation gave a trimethyl ester 97 identical with trimethyl ester of the Seco-A acid derived from dihydrobetulic acid[54]. From the above physical and chemical evidences Guise et al confirmed the structure of methyl

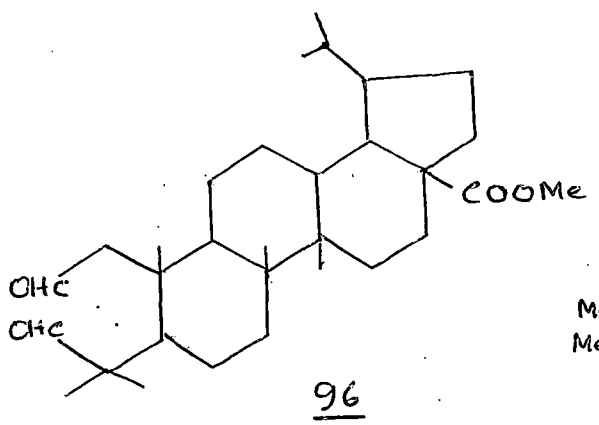
alphitolate as depicted in 95B. The stereochemistry of 1,2-glycol grouping in 95B was based on quantitative lead tetra-acetate titra-



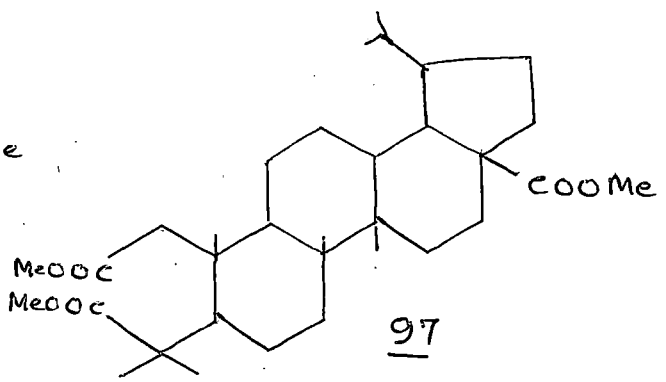
95A, $R_1 = C(CH_3)=CH_2$; $R_2 = H$

95B, $R_1 = C(CH_3)=CH_2$; $R_2 = Me$

95C, $R_1 = HC(CH_3)-CH_3$; $R_2 = Me$



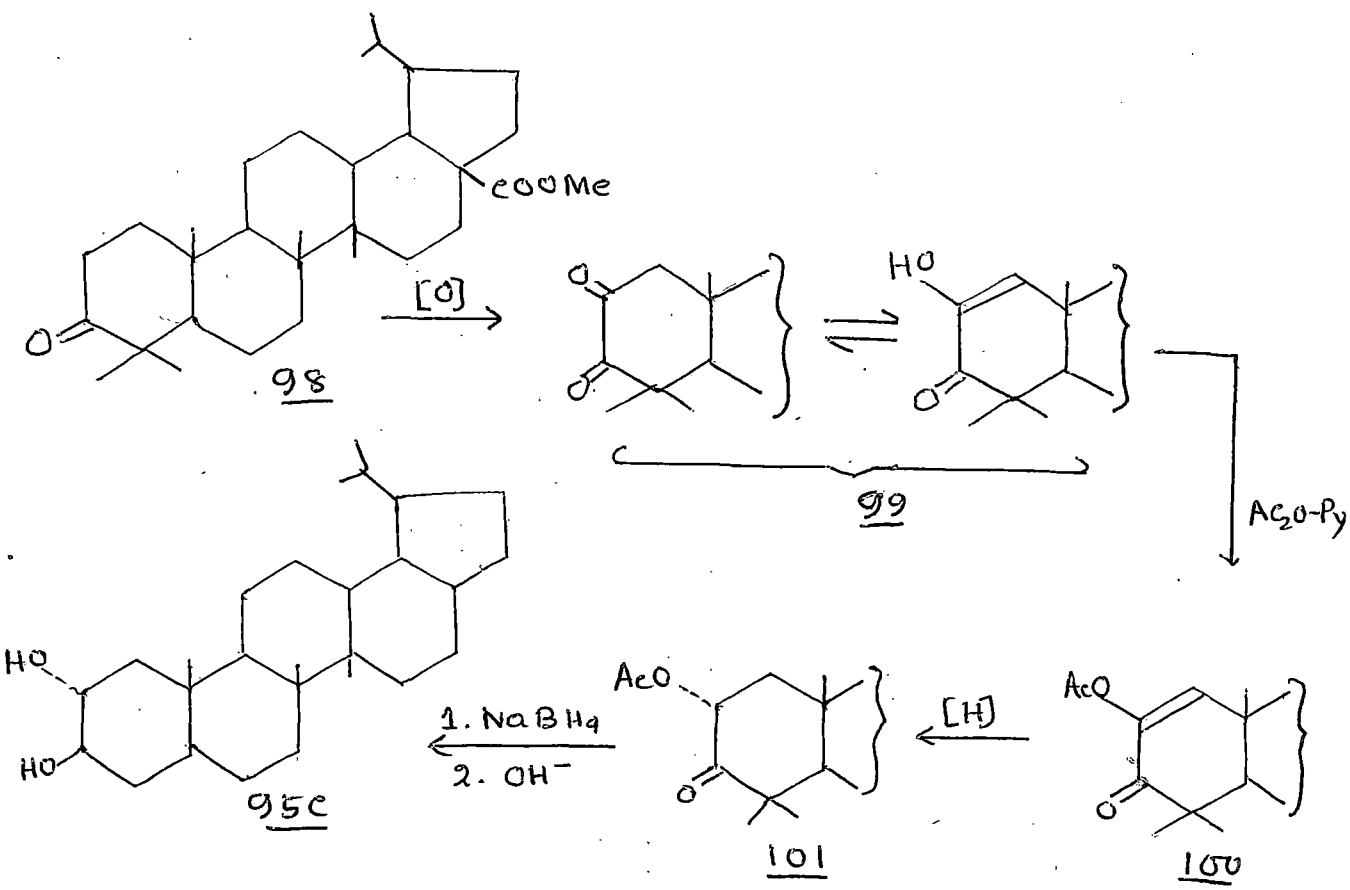
96



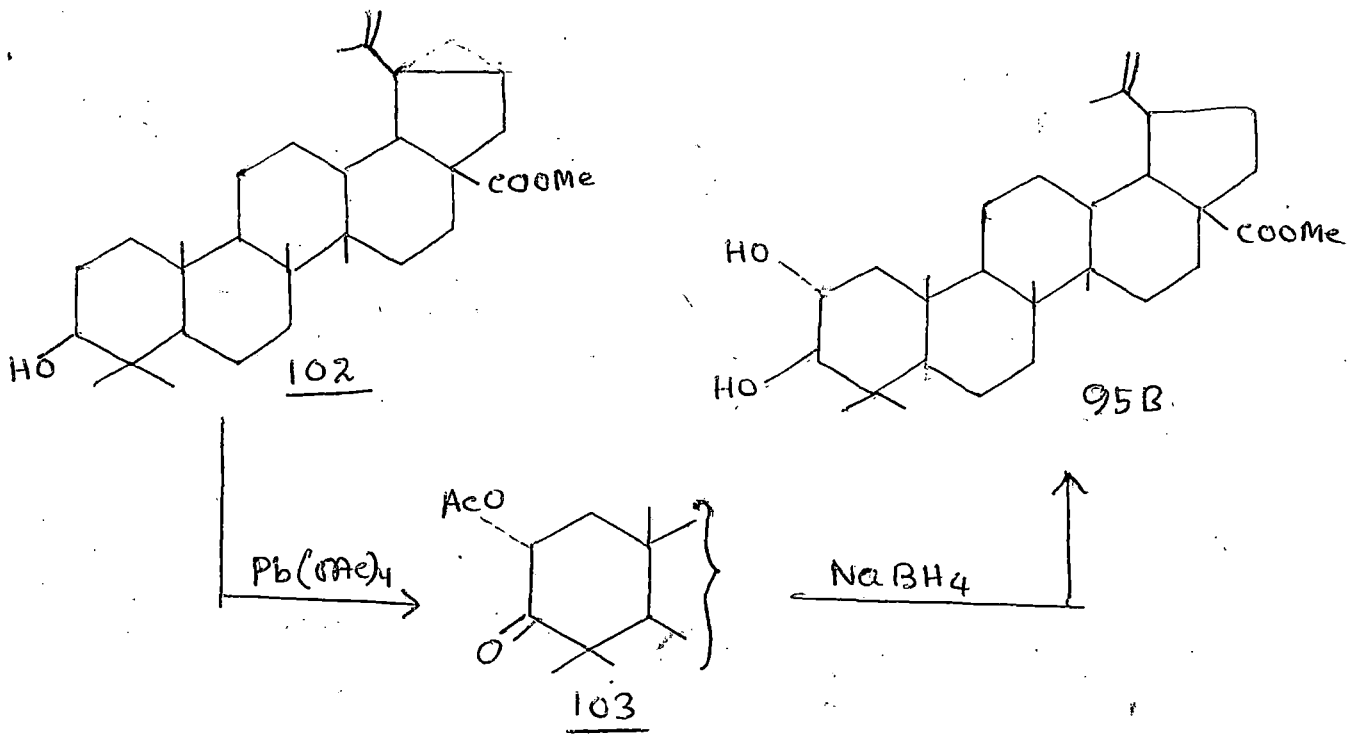
97

tions under the defined conditions of Djerassi and Ehrlich[55], where a value $k = 2.7 \times 10^{-3}$ L mole^{-1} Sec^{-1} was obtained, identical for triterpene 2α, 3β-glycols.

Khastgir et al[56] also reported the synthesis of methyl dihydroalphitolate 95C. Methyl dihydrobetulonate 98 prepared from betulinic acid, was oxidised by passing oxygen through a suspension of K -tertiary butoxide in dry tertiary-butanol to give diosphenol 99, m.p. 131-33°, $\lambda_{max}^{m\mu}$ 269 (ε, 7532) which on acetylation gave the corresponding acetate 100 m.p. 194-96°, λ_{max} 237 mμ (ε, 9968). This acetate 100 on hydrogenation in presence of 10% palladium-on-charcoal catalyst gave 2α-acetoxy methyl dihydrobetulonate m.p. 223-25°, 101. Reduction of 101 with sodium borohydride at PH 8 gave 2α-acetoxy dihydrobetulinate which on alkaline hydrolysis gave methyl dihydroalphitolate 95C identical with an authentic sample of methyl dihydroalphitolate.

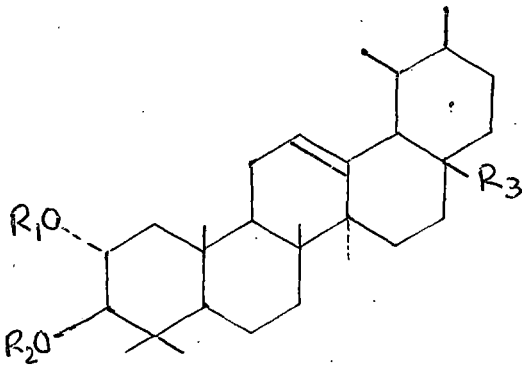


Rastogi et al[57] have very recently described the synthesis of methyl alphitolate 95B from methyl betulonate 102. Methyl betulonate 102 on treatment with lead tetra-acetate yielded 2α-acetoxy-3-ketoderivative 103 m.p. 191° as the major product. That the acetoxy group on C-2 is equatorial was shown by the NMR signal at 2.12 ppm for the acetoxy group and a quartet at 5.6 ppm (J = 13 and 6 Hz) for axial proton at C-2 containing the acetoxy group. The acetoxy ketone 103 on reduction with sodium borohydride gave the glycol namely methyl alphitolate 95B m.p. 235°. NMR signal at 3.66 ppm (multiplet about 26 Hz wide) confirmed the position and 2α, 3β configuration of the hydroxy groups in methyl alphitolate 95B.



6. 2α-hydroxy ursolic acid:

Glen et al[34] reported the isolation of 2α-hydroxy ursolic acid 104A as its methyl ester from the leaves of Rose-bay Willow-herb (chamaenorion angustifolium). That the methyl ester 104B was an ursane derivate was indicated by the presence of three peaks in the region 1400-1350 cm^{-1} and two peaks in the region 1320-1240 cm^{-1} in the infrared spectrum[59]. NMR spectrum revealed the presence of one olefinic proton (τ 4.73) and one methyl ester group (τ 6.38). Further evidence came from mass spectrum of 104B which showed intense peaks at m/e 263, 203 and at 223 corresponding to ions A, B and C, a pattern frequently associated with Δ^{12}-triterpenoids[35]. A similar mass fragmentation pattern was observed for the 2α-hydroxy-uvaol 104C formed by lithium aluminium hydride reduction of the dihydroxy methyl ester 104B. The methyl ester 104B formed a diacetate 104D (twin distinct peaks in the NMR at τ 7.97 and 8.04[60]). This physical evidence for the presence of a 2,3-diol system in 104B was further substantiated by the preparation of an isopropylidene derivative and also by the formation of the diosphenol 105 by chromium trioxide-pyridine oxidation. The presence of an ursane skeleton in the ester 104B was shown by the Wolff Kishner reduction of the diosphenol 105 to give urs-12-en-28 oic acid 106 identical

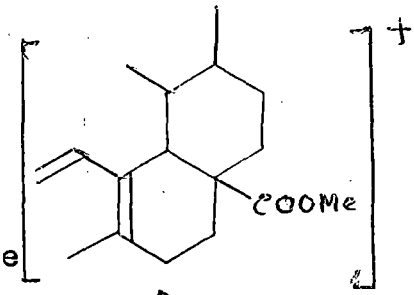


104A $R_1 = R_2 = H$; $R_3 = COOH$

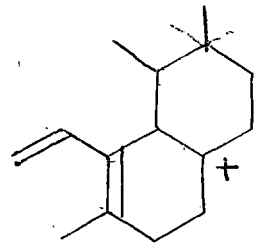
104B $R_1 = R_2 = H$; $R_3 = COOMe$

104C $R_1 = R_2 = H$; $R_3 = CH_2OH$

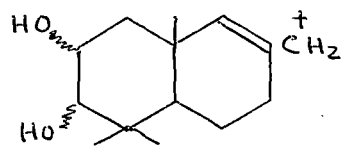
104D $R_1 = R_2 = Ac$; $R_3 = COOMe$



A

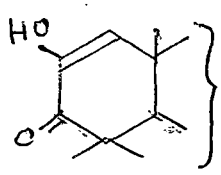


B

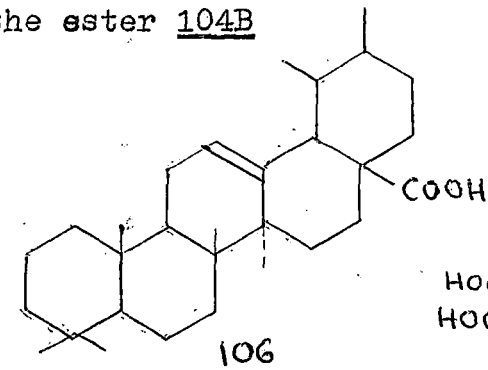


C

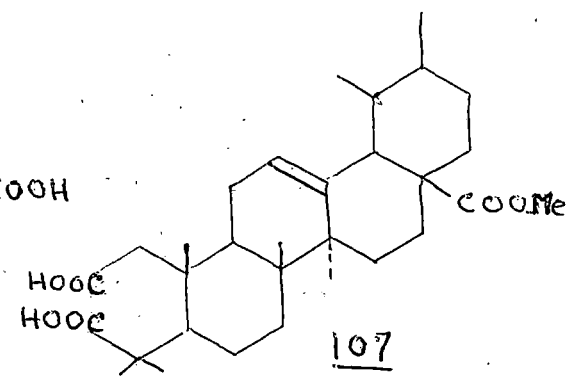
with an authentic sample. Cleavage of the diosphenol 105 with alkaline hydrogen peroxide gave 2,3 seco-urs-12-en-2,3,28-trioic acid-28 methyl ester 107 which confirmed the presence of the ursane-skeleton in the ester 104B



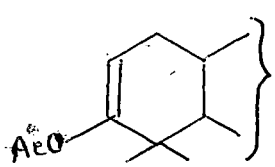
105



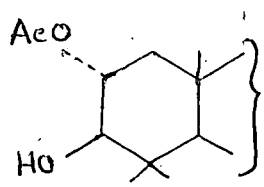
106



107



108



109

The confiiguration of the hydroxyl group in the dihydroxy ester 104B was settled by the synthesis of the trans diequatorial diol 104B. The enol acetate 108 on treatment with diborane and then subsequent oxidation of the intermediate afforded three products viz. methyl ursonate, methyl ursolate and methyl 2α-, 3β-dihydroxy urs-12-en-28-oate 104B, the latter was identical with that obtained from the natural dihydroxy acid 104A.

Further support for the aissignment of theα-configuration to the hydroxyl group at C-2 in 104B was revealed by the NMR spectrum of the monoacetate, methyl 2α-acetoxy-3β-hydroxy urs-12-en-28-oate 109. The NMR spectrum showed signals in the region τ4.9 - 5.4 due to C-2 proton. The breadth of the resonance indicated that at least two couplings of about 8 c/sec are present and this can only occur when the proton at C-2 occupies an axial conformation allowing an axial interaction with the proton at C-3 and a similar interaction with the axial proton at C-1. In addition, there is an axial-equatorial interaction with the other proton at C-1. This indicated that the proton at C-2 has the axial conformation and acetoxy group at C-2 has the equatorial (α) conformation.

Rastogi et al[57] repeated Glen's[34] work and obtained the enol acetate m.p. 198^{o} in 46%. Yield but subsequent steps of hydroboration and oxidation yielded only the starting material methyl ursolate.

Rastogi et al employed lead tetra-acetate oxidation on methyl ursolate 110 and obtained 2α-acetoxy-3-keto derivative 111 as an amorphous powder, the latter was characterized by NMR spectrum. Reduction of 110 with sodium borohydride gave the methyl 2α-hydroxy ursolate 104B in an overall yield of 26.7%, characterized by IR and NMR data.