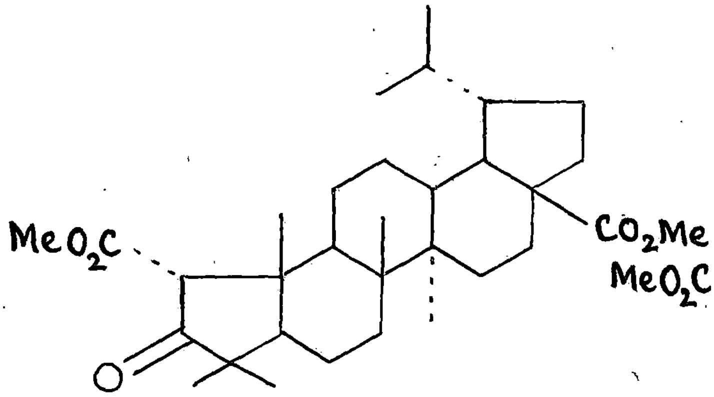
CAHPTER-III

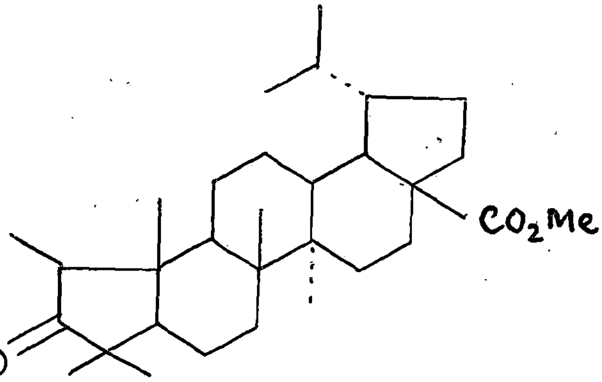
Partial Synthesis of All the Four Stereoisomers of Dimethyl Dihydroceanothate starting from Betulinic Acid.

The objective of the present study was to develop a partial synthesis of all the four stereoisomers of dimethyl dihydroceanothate starting from betulinic acid. In order to achieve this goal, it was necessary (I) to convert the six membered ring A present in the starting material into a five membered ring of the product without affecting the structural and stereochemical features present in the other parts of the starting material and (II) to introduce the substituents at the two asymmetric centres in ring A with proper stereochemistry.

The second problem could best be solved by introducing the stereochemical centres in a stepwise predetermined fashion on some synthetic intermediate. Since Eade et al[22] have shown that the C-3 hydroxyl group in dimethyl ceanothate series (except in one case) could be introduced by sodium borohydride reduction of the corresponding C-3 ketones, it was thought that the β-keto esters (66) and (67) would be the best intermediates for this purpose.



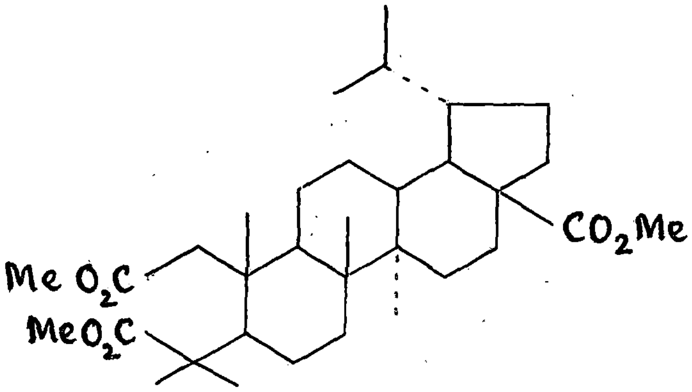
(66)



(67)

These compounds contained the C-2 methoxycarbonyl group in proper stereochemistry and once the synthesis of these could be attained, the introduction of C-3 hydroxyl group in proper stereochemistry would not be very difficult.

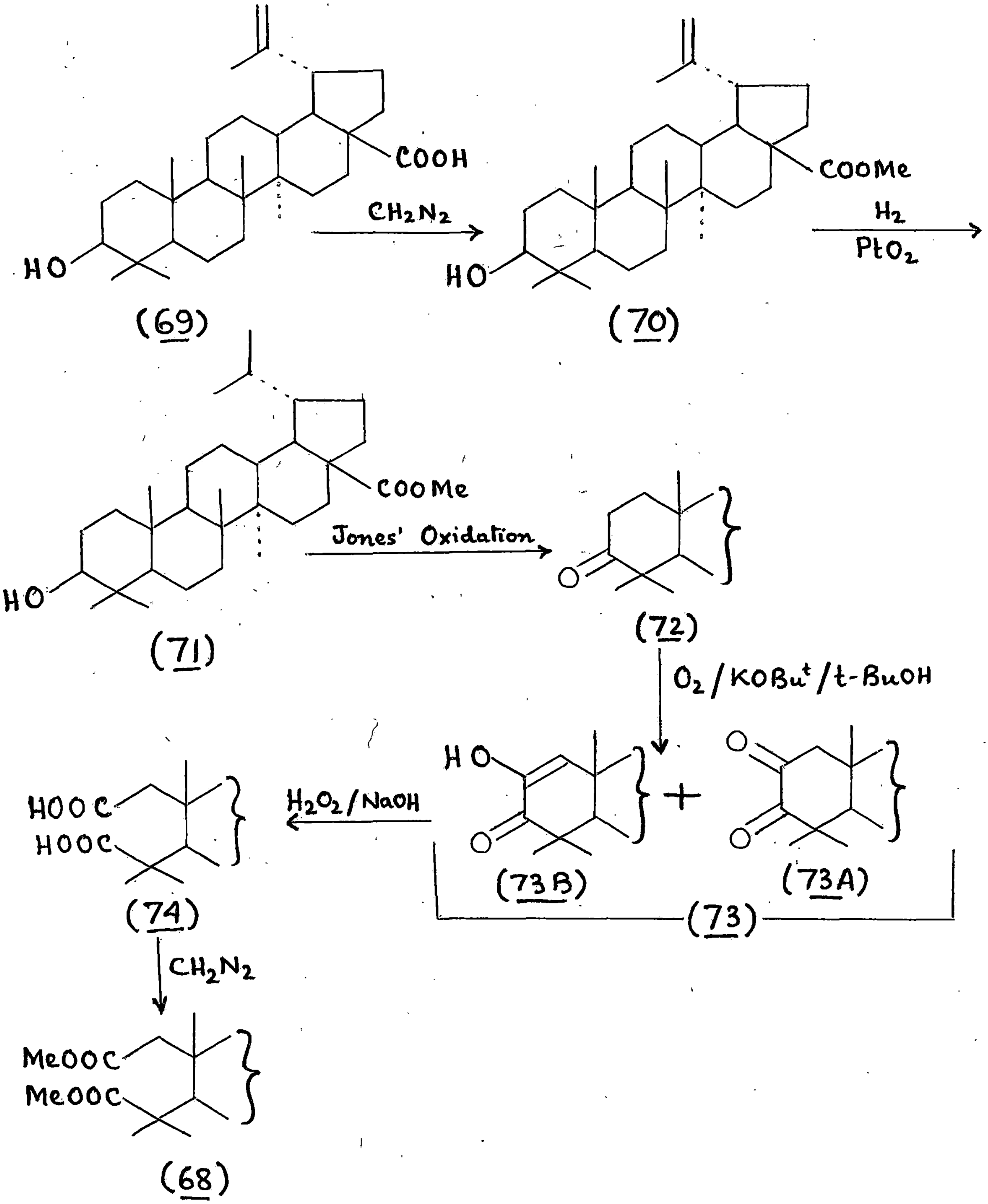
Regarding the first problem, that is, the construction of the five membered A ring skeleton present in (66) and (67), it had to be noted that both these were β-keto esters. Hence, theoretically, they could be prepared by Dieckmann condensation of the corresponding 2, 3-seco trimethyl ester (68). Thus the problem was reduced to the synthesis of the trimethyl ester (68). This trimethyl ester (68) could be obtained by oxidative opening of ring A of a betulinic acid derivative. But betulinic acid



(68)

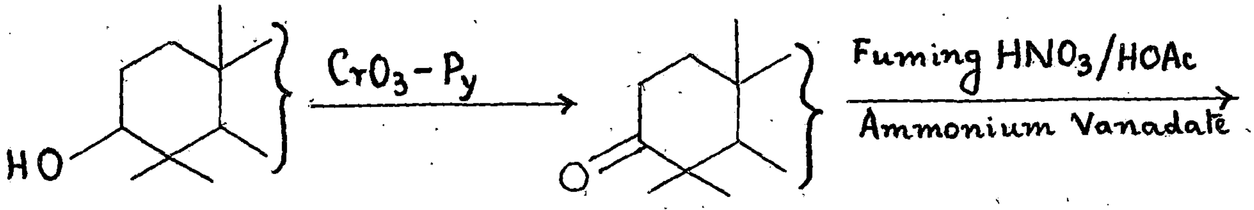
itself contained an isopropenyl side chain, and the exocyclic double bond might be expected to undergo disruption under drastic oxidation condition required for ring opening. Thus, it was necessary to reduce the double bond of betulinic acid at an early stage.

Betulinic acid (69) extracted from the acid part of the benzene extract of the bark of *Bischofia Javonica Blume* on esterification with diazomethane gave methyl betulinate (70), m.p. 223-24°, $(\alpha)_D$ 5°, ν_{max}^{nujol} 3560 (OH), 1715 ($COOCH_3$), 1660 and 880 cm^{-1} (= CH_2). Hydrogenation of methyl betulinate (70) in presence of PtO_2-catalyst in a mixture of acetic acid and ethyl acetate gave methyl dihydrobetulinate (71), m.p. 236-38°, ν_{max}^{nujol} 3560 (OH), 1710 cm^{-1} ($COOCH_3$). Jones' oxidation of methyl

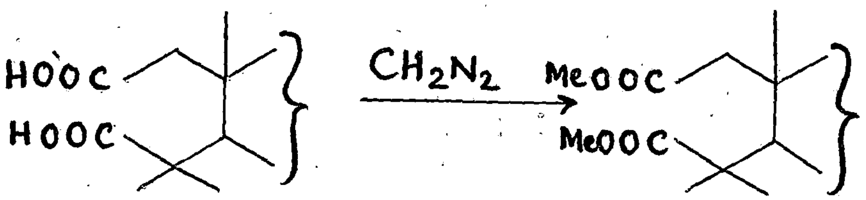

HO
COOH
(69)
CH2N2
HO
COOMe
(70)
H2
PtO2
HO
COOMe
(71)
Jones' Oxidation
(72)
O2/KOBut/t-BuOH
HO
(73B)
(73A)
(73)
H2O2/NaOH
HOOC
HOOC
(74)
CH2N2
MeOOC
MeOOC
(68)

dihydrobetulinate (71) afforded methyl dihydrobetulonate (72), m.p. 191-93°, $(\alpha)_D$ 8°, ν_{max}^{nujol} 1730 ($COOCH_3$), 1705 cm^{-1} (CO). Autoxidation of (72) by passing a stream of oxygen in presence of potassium tertiary butoxide in tertiary butanol furnished a product, m.p. 131-33°, $(\alpha)_D$-1.96°, ν_{max}^{nujol} 3460 (OH), 1730 ($COOCH_3$), 1670, 1650, 860 cm^{-1}, λ_{max} 269 nm (ϵ, 7532). It gave a positive ferric chloride colouration for diosphenol. T.L.C. showed two spots indicating that it was a mixture of two tautomeric forms the α-diketone (73A) and the diosphenol (73B). Hydrogen peroxide oxidation[36] of the mixture of (73A) and (73B) in presence of sodium hydroxide gave after acidification the A-seco acid (74), m.p. 175-77°, ν_{max}^{nujol} 1710 and 1680 cm^{-1} (COOH) which on esterification with diazomethane gave the desired trimethyl ester (68), m.p. 146-47°, ν_{max}^{nujol} 1745 and 1725 cm^{-1} ($COOCH_3$) identical with the compound reported in literature[3,12].

The overall yield of the triester (68) from methyl dihydrobetulinate (71) by the above procedure was very poor. To increase the yield it was felt that (I) a shorter reaction path and (II) a better reaction condition were necessary. After several experimentation such a synthetic pathway could be arrived at. Oxidation of methyl dihydrobetulinate (71) with Chromium trioxide-pyridine complex[37] gave in very good yield methyl dihydrobetulonate (72) which on oxidation with fuming nitric acid in acetic acid in presence of ammonium vanadate[12,38] directly afforded the seco-acid (74). The latter on esterification gave the triester (68) in fairly good overall yield.

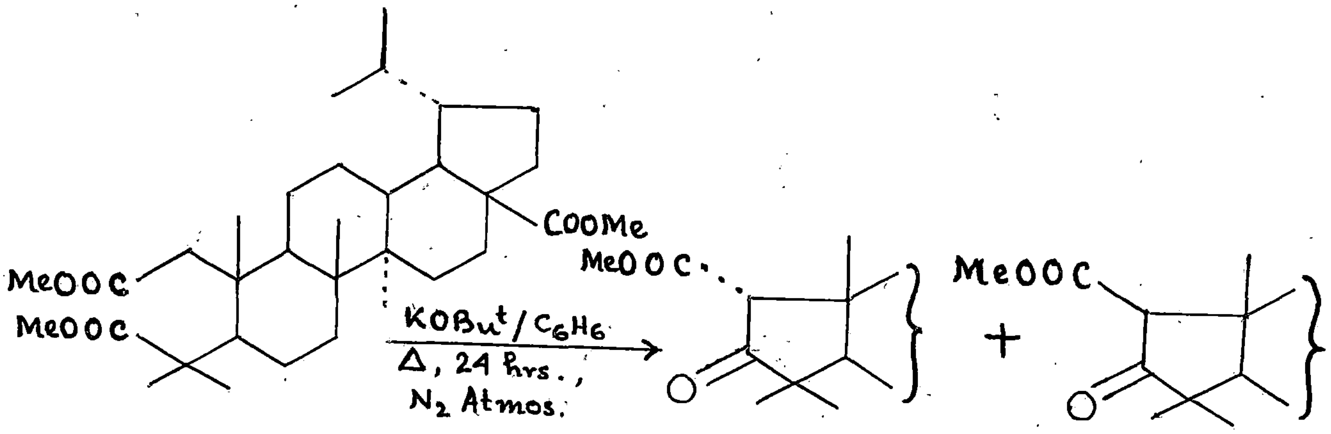


(71) (72)



(74) (68)

Dieckmann condensation was performed by refluxing the trimethyl ester (68) with potassium tertiary butoxide in benzene solution for 24 hours in an atmosphere of nitrogen[39,40]. During the period of reaction a portion of the solvent was removed with a Dean-Stark trap. The gummy reaction product obtained after usual work up was chromatographed over deactivated alumina. A mixture of petrol and benzene (3:2) first eluted a solid <u>A</u>, m.p. 189-91°. Further elution with the same solvent mixture gave another solid <u>B</u>, m.p. 173-75°. The proportions of two solids varied with the reaction condition and the time of refluxing. Solid <u>A</u> on crystallisation from methanol afforded needle shaped crystals, m.p. 191-93° (TLC-homogenous), $(\alpha)_D$ 89°, ν_{max}^{nujol} 1755 (cyclopentanone), 1725 cm^{-1} (CO_2Me) having the same m.p, rotation and I.R. spectra reported in literature[22] for dimethyl dehydrodihydroceanothate, i.e., methyl-2α-methoxycarbonyl-3-oxo-A(1)-norlupan-28-oate (<u>66</u>). Solid <u>B</u> on crystallisation from methanol afforded needle shaped crystals, m.p. 175-77° (TLC-homogenous), $(\alpha)_D$42°, ν_{max}^{nujol} 1750 (cyclopentanone), 1720 cm^{-1} (CO_2Me) having the same m.p., rotation and I.R. spectra reported in literature[22] for methyl -2β-methoxycarbonyl-3-oxo-A(1)-norlupan-28-oate,(<u>67</u>).



(<u>68</u>) (<u>66</u>) (<u>67</u>)

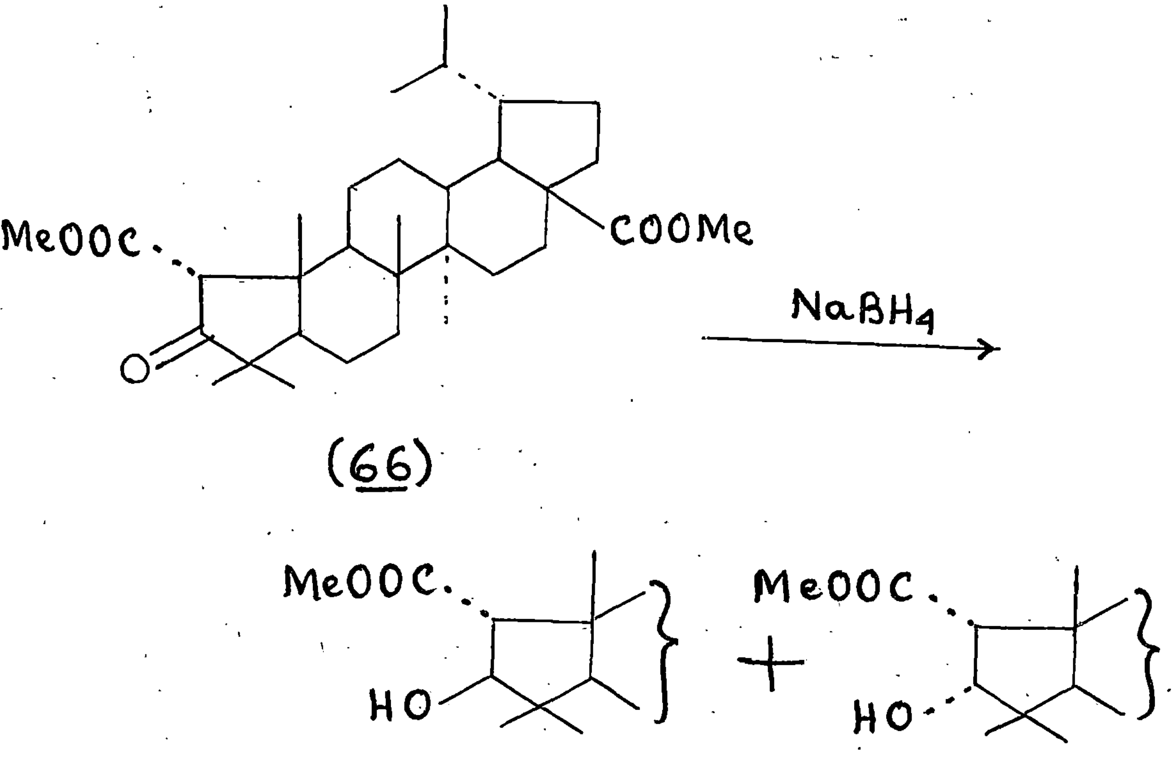
Thus the most important synthetic goal, i.e., the construction of a five membered A-ring without affecting the stereochemistry of various ring junctions was achieved. Furthermore, in the β-ketoesters (66) and (67) the stereochemistry at C-2 was also known.

Sodium borohydride reduction of the β-ketoester (66) in a mixture of methanol and dioxan gave an oily residue which was chromatographed over deactivated alumina. Elution with a mixture of petrol and benzene (2:3) first afforded a solid C, m.p. 257-59°. Further elution with the same solvent gave another solid D, m.p. 140-42°.

Solid C, m.p. 257-59° on crystallisation from methanol gave needle shaped crystals, m.p. 261-63°, $(\alpha)_D$ 22°, ν_{max}^{nujol} 3540 (OH), 1730 (CO_2Me), 1710 cm^{-1} (CO_2Me), and was found to be identical (m.m.p., Co-TLC and I.R. comparison) with an authentic sample of dimethyldihydroceanothate[5] [methyl-3β-hydroxy-2α-methoxycarbonyl-A(1)-norlupan-28-oate] (75) supplied by Professor P. de Mayo.

Solid D on crystallisation from methanol gave crystals, m.p. 140-42°, (TLC-homogenous), ν_{max}^{nujol} 3560 (OH), 1745 (CO_2Me) and 1705 cm^{-1} (CO_2Me). From the method of preparation and by analogy with the previous work[22] it was evident that this compound must be C-3 epimer of dimethyldihydroceanothate, i.e. methyl-3α-

hydroxy-2α-methoxycarbonyl-A(1)-norlupan-28-oate, (76). Further confirmation of this structure by NMR spectra is in progress.



(66)

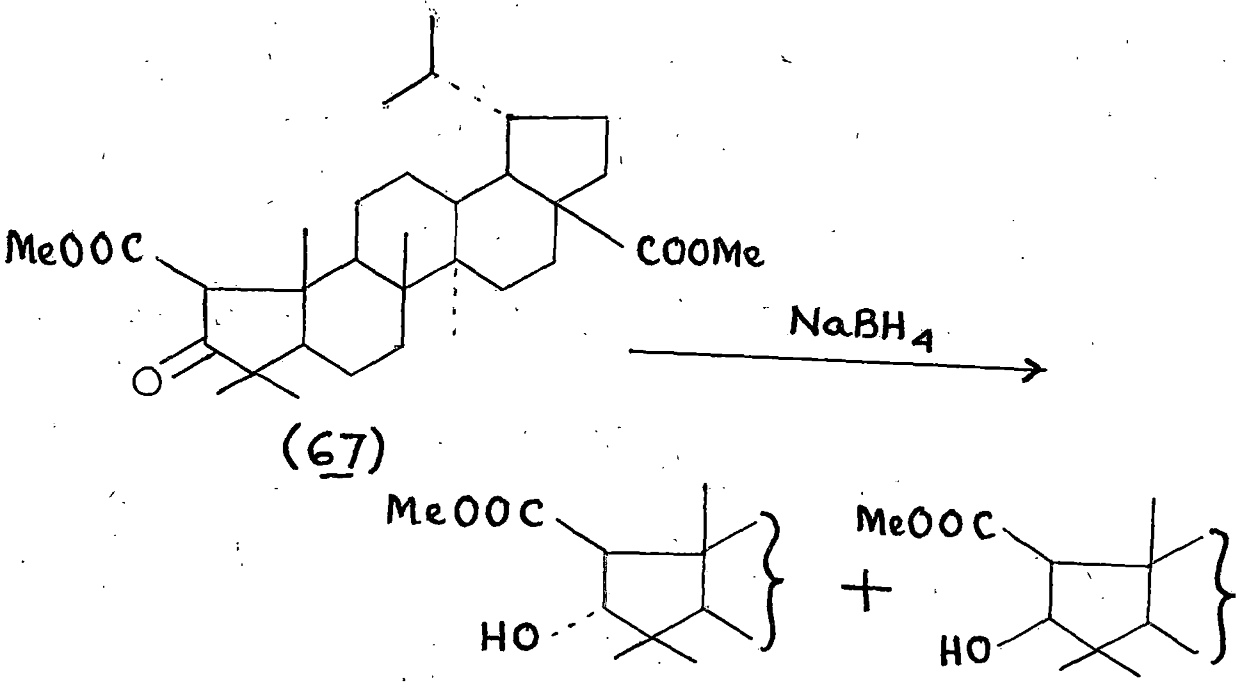
(75) (76)

Attempts to reduce the β-keto ester (67) by sodium borohydride under conditions in which the β-ketoester (66) underwent smooth reduction failed. However, when the β-ketoester (67) was stirred with a large excess of sodium borohydride in a mixture of methanol and dioxane for a sufficiently long time the desired reduction took place. The oily reaction product was shown by TLC studies to be a mixture of two compounds which could be resolved by chromatography over a column of deactivated alumina. Elution with a mixture of petrol and benzene (1:1) afforded a solid E, m.p. 197-201°. Further elution with a mixture of petrol and benzene (3:7) gave another solid F, m.p. 168-72°. Evidently E and F must be the C-3 epimers. This observation was at variance with the work of Eade et al[22] who showed that dimethyl epidehydroceanothate (22) on reduction (of course under somewhat milder condition) afforded only the 3β-hydroxy compound (23).

The solid E, m.p. 197-201°, on crystallisation from aqueous methanol afforded crystals, m.p. 202-203° (TLC-single spot), IR ν_{max}^{nujol} 3490 (OH), 1730 (CO_2Me), 1695 cm^{-1} (CO_2Me). The NMR spectrum (80 MH_z) of the compound showed signals at δ 0.7 to 1.1 (seven methyl groups), 2.1 (one proton, OH) 3.06 (1H, doublet, J = 7 H_z, CH-COOMe i.e., C-2 proton), 3.65 (3H, singlet, $-COOCH_3$), 3.7 (3H, singlet, $-COOCH_3$) and 4.18 (1H, doublet = 7 H_z; CHOH i.e., C-3 proton). The signal due to OH at δ 2.1 in the spectrum was a singlet slightly broadened line which moved upfield on heating.

The solid F, m.p. 168-72° on crystallisation from aqueous methanol afforded crystals, m.p. 174-76° (TLC-single spot), IR ν_{max}^{nujol} 3540 (OH), 1740 (CO_2Me), 1690 cm^{-1} (CO_2Me). The NMR spectrum (80 MH_z) of the compound showed signals at δ 0.7 to 1.1 (seven methyl groups), 2.35 (1H, doublet, J = 7.2 H_z; C<u>H</u>-CO_2Me, i.e., the C-2 proton) a doublet centred at 2.8 (1H, doublet, J = 4.5 H_z; O<u>H</u>), 3.65 (3H, singlet, $COOC\underline{H}_3$), 3.7 (3H, singlet, $COOC\underline{H}_3$), 4.02 (1H, doublet of doublets, J = 7.2 H_z and 4.5 H_z; C<u>H</u>OH i.e. C-3 proton). The doublet due to OH moved upfield on heating to 60°C.

It was evident from the method of preparation that solid <u>E</u> and solid <u>F</u> were C-3 epimers, in one the hydroxy at C-3 was α and in the other it was β. However, at variance with the work of Eade <u>et al</u>[22] on analogous compounds, the coupling constant values were found to be nearly equal. Since this vicinal coupling constants were nearly equal there was no way to deduce at this stage in which compound was the 3-hydroxy was β and in which it was α. However, the correct conformations were established by other means. The fact that the proton on C-2 in <u>E</u> was shifted downfield to δ 3.06 indicated that the proton at C-2 and the hydroxy group at C-3 in this compound <u>E</u> was on the same side of the ring. Consequently, this compound <u>E</u> was the <u>trans</u> compound, i.e. methyl - 3α-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (<u>77</u>). Evidently <u>F</u> was the <u>cis</u>-compound, i.e., methyl-3β-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (<u>78</u>).



(67)

(77)

(78)

Additional support for the above interpretation was found in the fact that, although the solution used for NMR of (78) was more dilute, the signal for OH group was found further downfield than that in (77), which meant that (78) was more strongly hydrogen bonded compared to (77). This can only occur when they are on the same side of the ring as in (78). ^{13}C NMR spectra were also run on the compounds (77) and (78). Such a spectrum of the compound (77) showed 31 strong lines, one of which represented two carbons. The ^{13}C NMR spectra of (78) showed 30 strong lines (and 5 weak lines, probably due to noise level) two of which represented two carbons. Thus both compounds had 32 carbons as expected. Both compounds had a peak near 82 ppm which evidently was the carbon bearing the hydroxyl group.

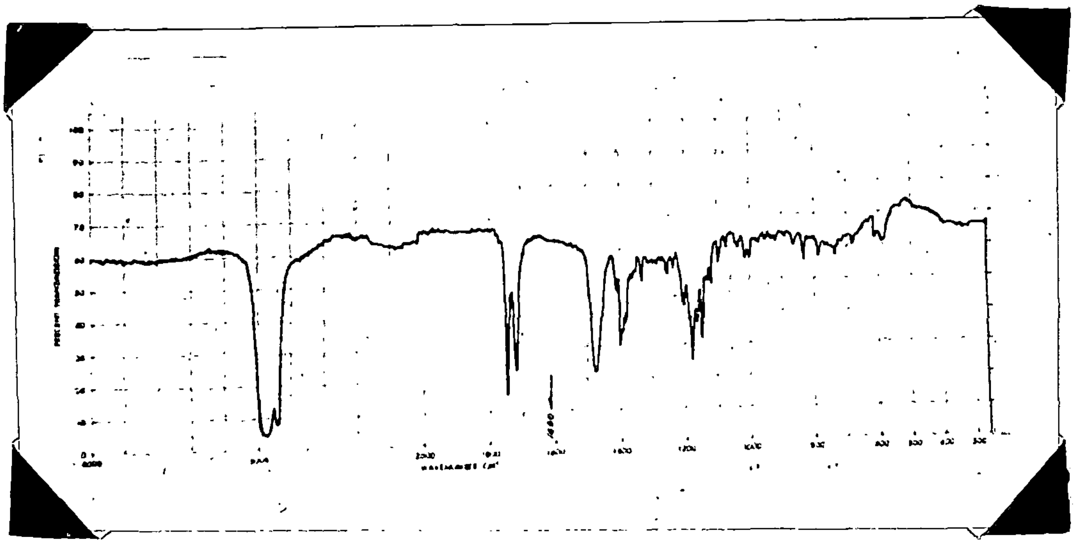


Fig. 1: IR Spectrum of Methyl Dihydrobetulonate (72)

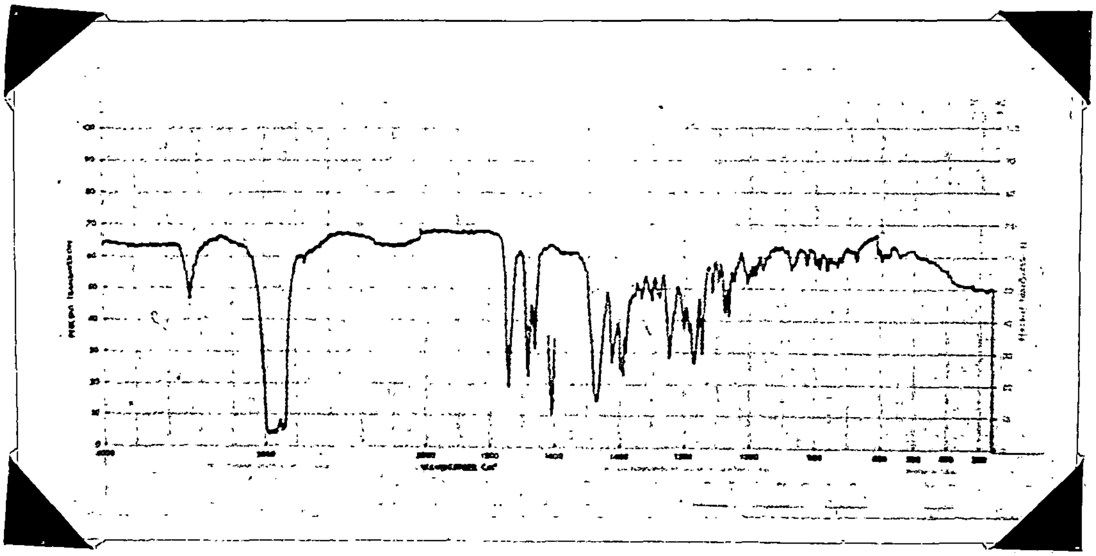
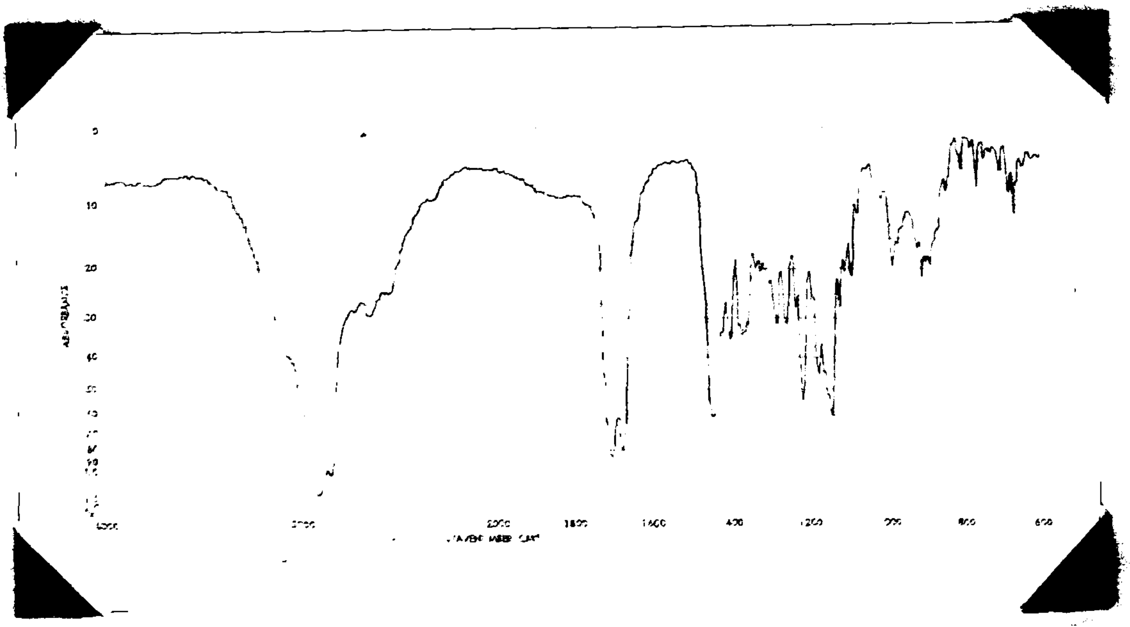
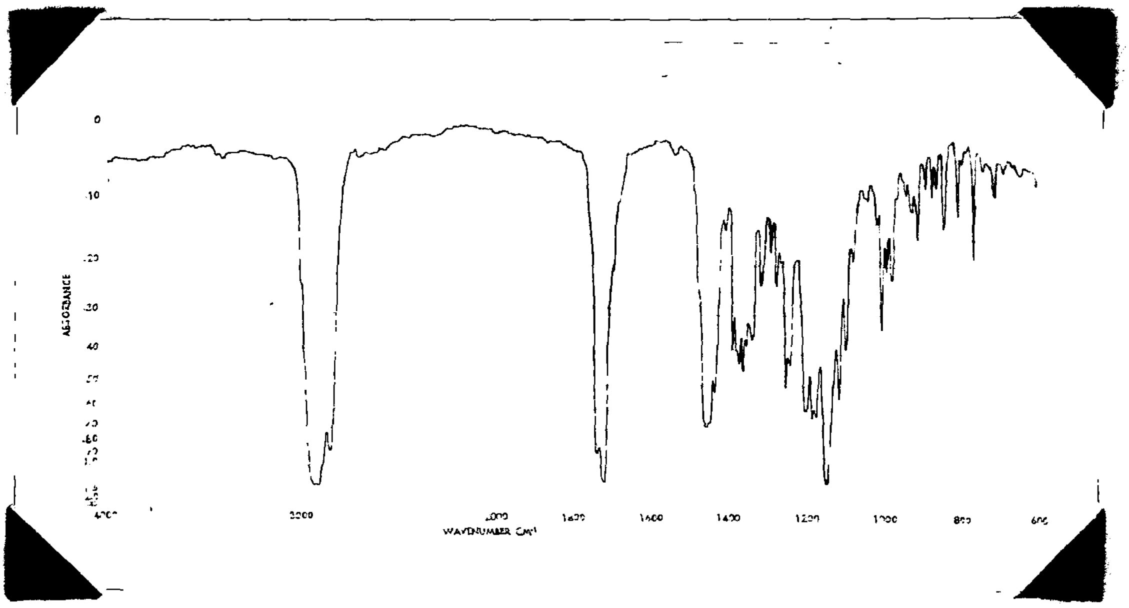


Fig. 2: IR Spectrum of Diosphenol (73)



<u>Fig. 3</u>: IR Spectrum of Seco-A-acid (<u>74</u>)



<u>Fig. 4</u>: IR Spectrum of Trimethyl Ester (<u>68</u>)

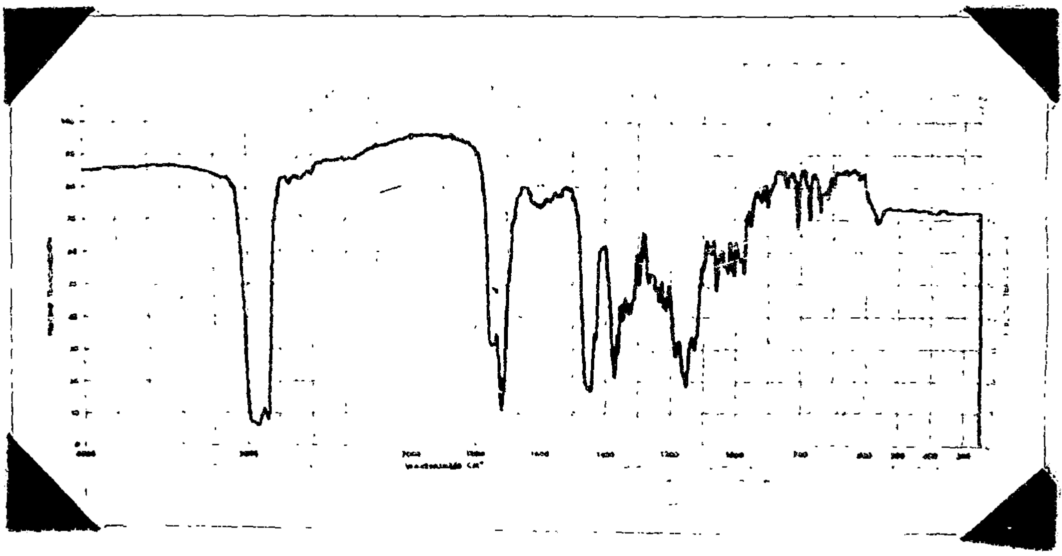


Fig. 5: IR Spectrum of Methyl-2α-methoxycarbonyl-3-Oxo-A(1)-norlupan-28-oate (66)

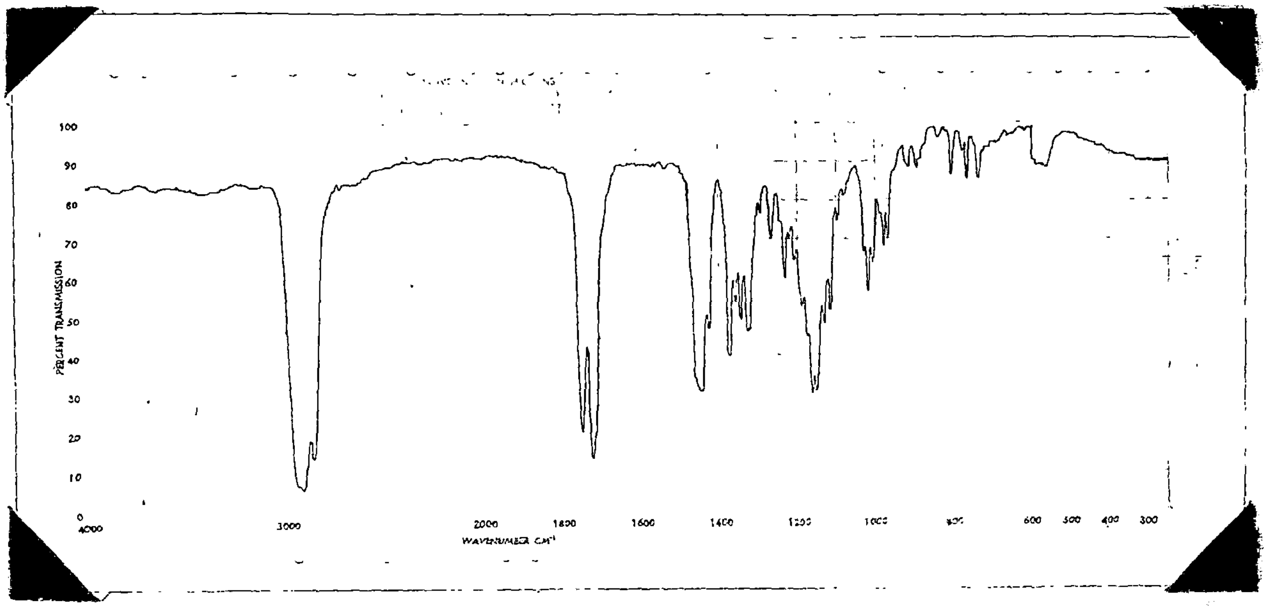
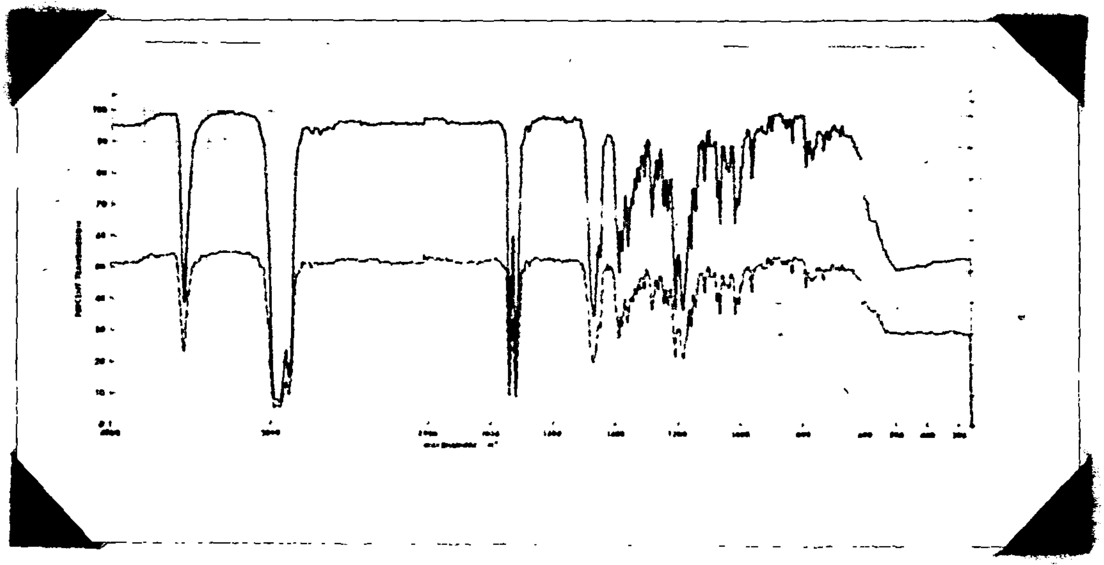
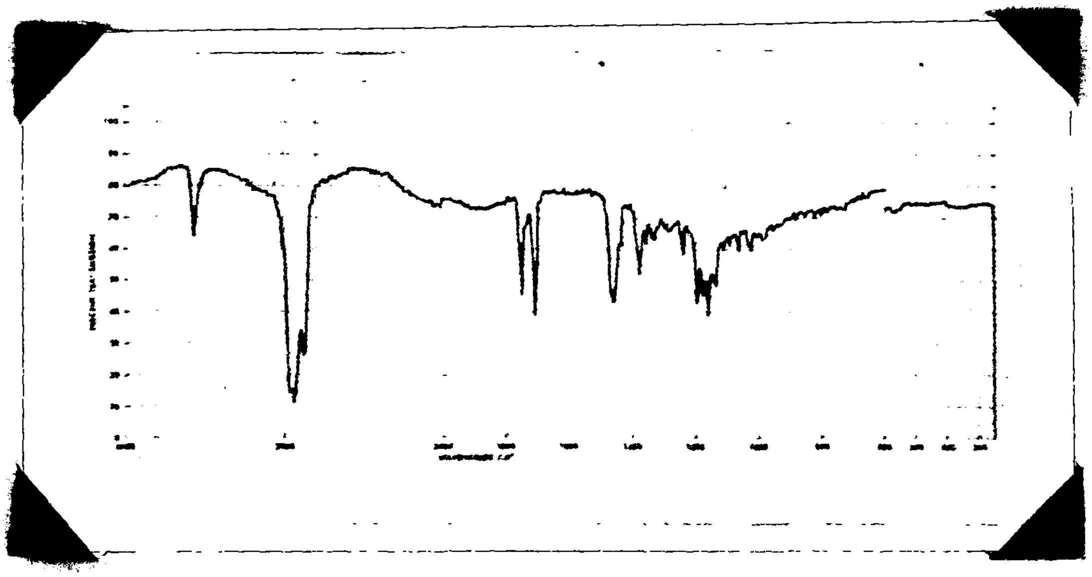


Fig. 6: IR Spectrum of Methyl-2β-methoxycarbonyl-3-Oxo-A(1)-norlupan-28-oate (67)



<u>Fig.7</u>: IR comparison of Dimethyl Dihydroceanothate (<u>75</u>) (solid line) with an authentic specimen (dotted line).



<u>Fig. 8</u>: IR Spectrum of Methyl-3α-hydroxy-2α-methoxycarbonyl-A(1)-norlupan-28-oate (<u>76</u>)

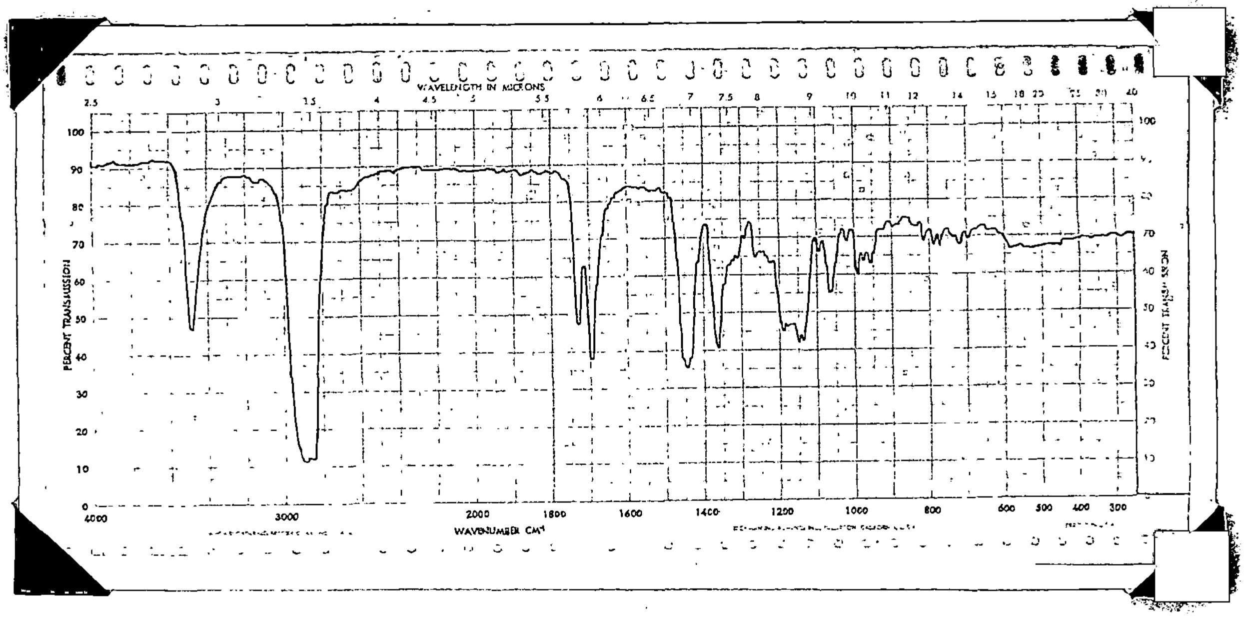


Fig.9: IR Spectrum of Methyl-3α-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (77)

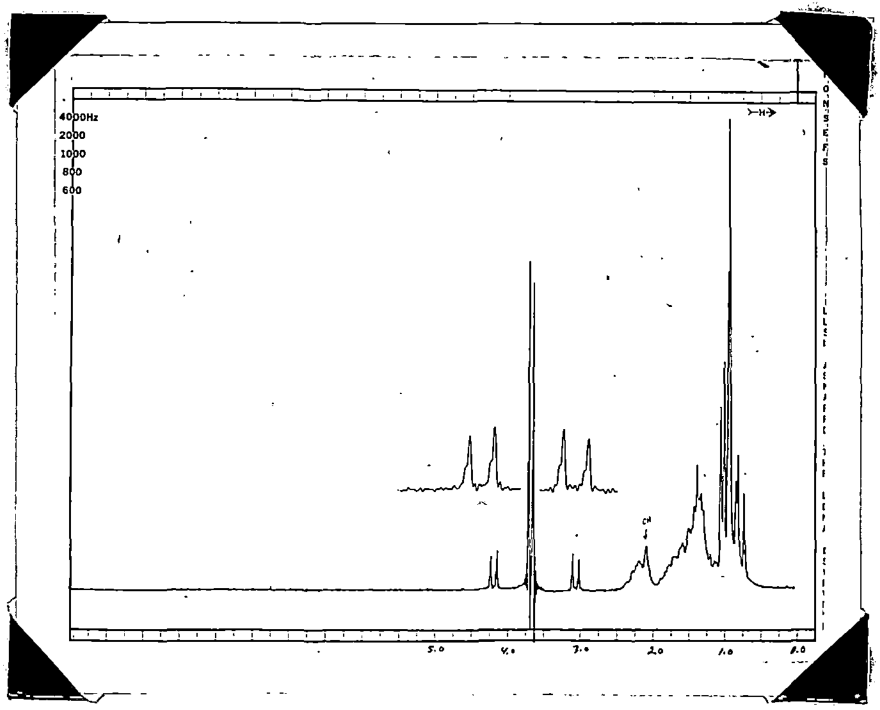


Fig.10: ^{1}H-NMR spectrum of Methyl-3α-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (77)

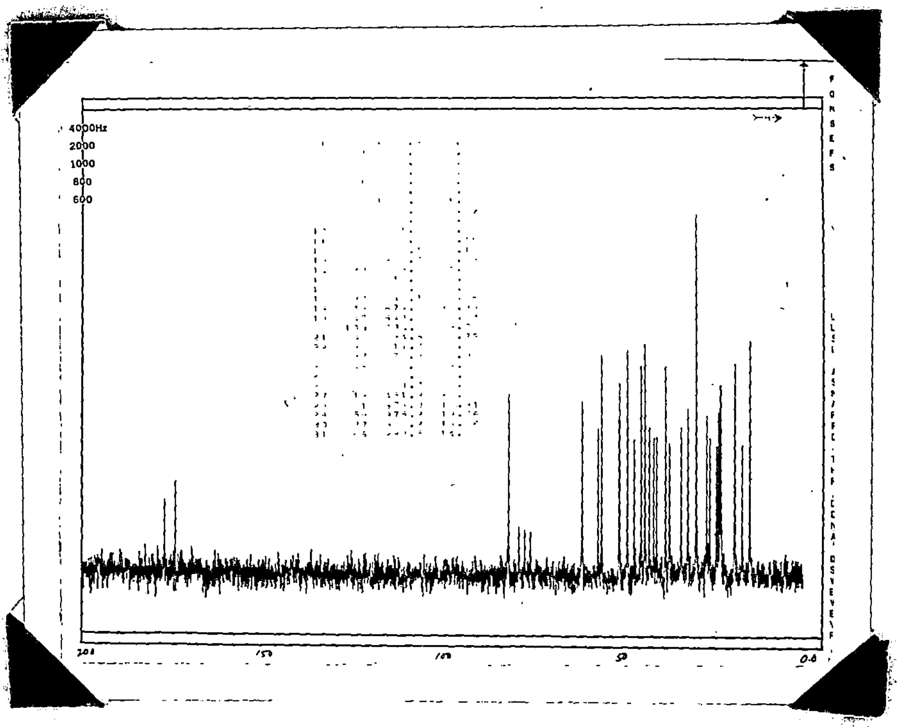


Fig. 11: ^{13}C-NMR spectrum of Methyl-3α-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (77)

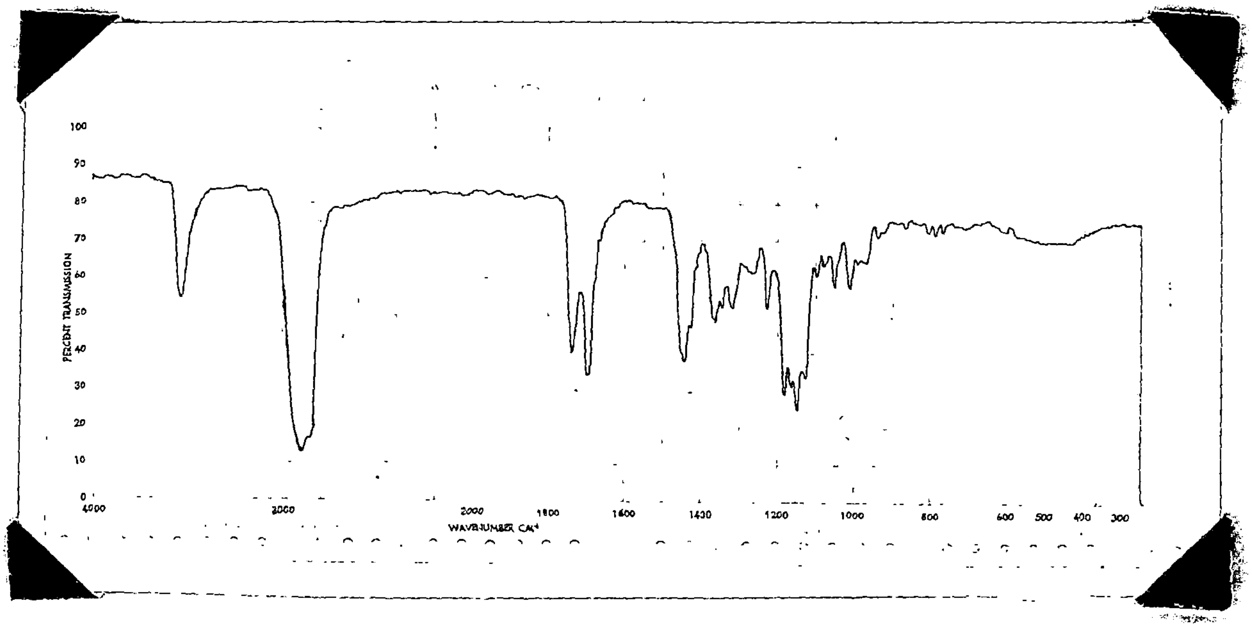


Fig. 12: IR spectrum of Methyl-3β-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (78)

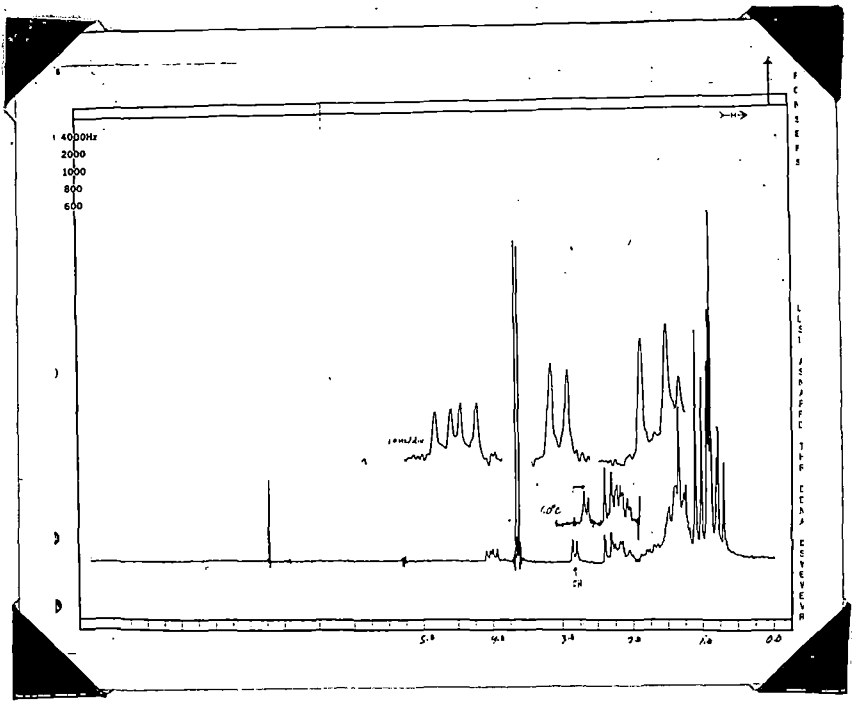


Fig. 13: ^{1}H-NMR spectrum of Methyl-3β-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (78)

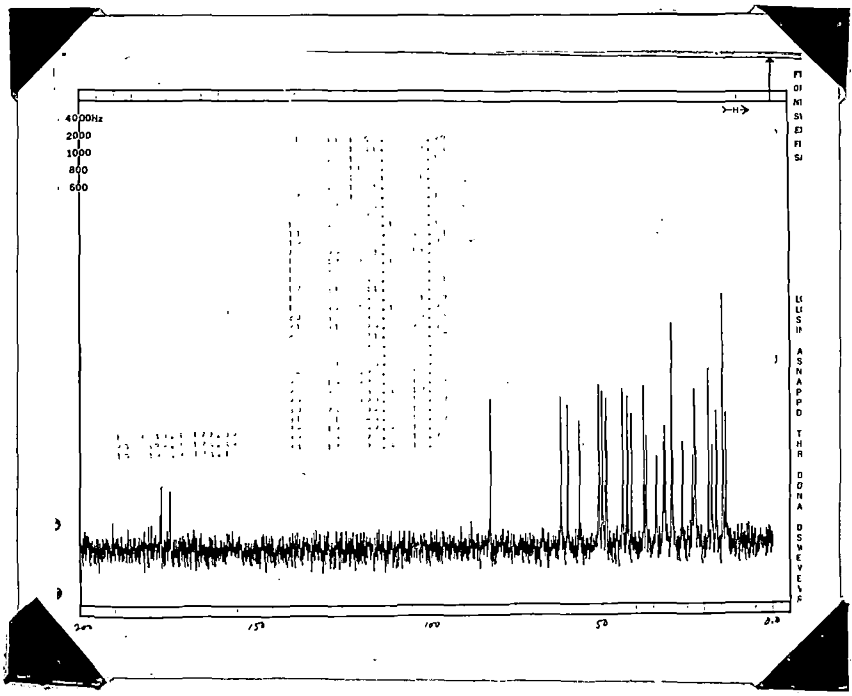


Fig. 14: ^{13}C-NMR spectrum of Methyl-3β-hydroxy-2β-methoxycarbonyl-A(1)-norlupan-28-oate (78).