CHAPTER II

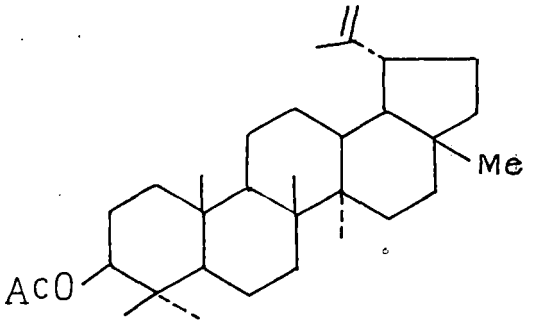
STUDIES ON THE ACTION OF SELENIUM DIOXIDE CONTAINING HYDROGEN PEROXIDE AND TERTIARY BUTANOL ON TRITERPENOIDS CONTAINING ISOPROPENYL GROUP

The study of the selenium dioxide oxidation of 3-keto triterpenoids have been made in detail by various group of workers. A number of various products may be formed dependent on the nature of the substrate and the reaction conditions uses; a brief resume of which has already been given in the review (Chapter I). Since there is no previous report of the systematic study on the selenium dioxide-hydrogen-peroxide oxidation of triterpenoids containing isopropenyl group, it has been felt necessary to study the applicability of this oxidising agent in the oxidation of isoropenyl group. Hence the present author desires to report herein the oxidative reaction of selenium dioxide-hydrogene peroxide in tertiary butanol on triterpenoids containing iso propenyl side chain e.g. lupenyl acetate 1 and methyl acetyl betulinate 2.

A. OXIDATION OF LUPENYL ACETATE 1 WITH SELENIUM DIOXIDE AND HYDROGEN PEROXIDE IN TERTIARY BUTANOL : ISOLATION OF 30-CARBOMETHOXY LUPENYL ACETATE 3, 30- CARBOMETHOXY-LUPAN-3 β, 29-DIYL ACETATE 4 AND 29-CARBOMETHOXY-LUPAN-3 β- YL acetate 5

Lupenyl acetate 1 on oxidation with selenium dioxide (SeO_2) containing hydrogen peroxide in tertiary butanol yielded

an oily residue which could not be separated by chromatography (see Experimental). The eluates were collected, esterified (CH_2N_2) and acetylated (pyridine-acetic anhydride) to give a material which on chromatographic separation afforded three products A, B and C.



1. SeO_2-H_2O_2 → A + B + C
2. Esterification (CH_2N_2)
3. Acetylation (Py/Ac_2O)

Characterization of A : Structure elucidation of 30-carbomethoxy lupenyl acetate 3

The chromatogram on elution with petroleum ether: benzene (2:3) furnished a compound A, analysed for $C_{33}H_{52}O_4$, m.p. 220-21°, responded to the TNM test for unsaturation. The IR spectrum (Fig. 1) of the compound A showed the presence of carbomethoxy and acetate group at 1740-1720, 1250 cm^{-1}, the peaks at 1630, 1125, 990, 825 cm^{-1} ($>C=C<^{H}_{H}$) were also observed. The mass spectrum (Fig. 2) of A showed the molecular ion peak at m/z 512. Thus from elemental analysis and mass spectrum, the molecular formula of compound A is established to be $C_{33}H_{52}O_4$. PMR spectrum (Fig. 3) of the compound A showed the presence of only six tertiary methyls appeared at 0.77, 0.81, 0.83 (2 x Me), 0.99 and 1.02 ppm, a sharp singlet at 3.75

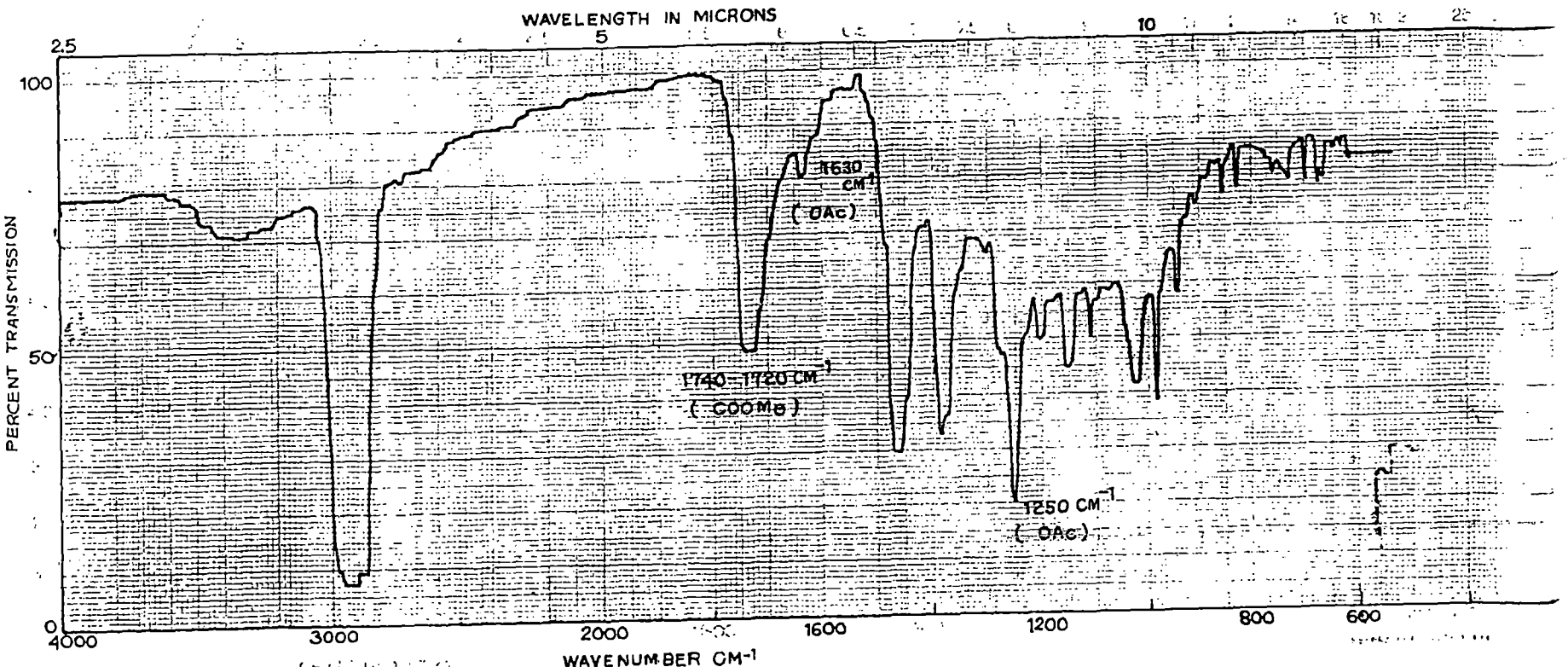


Fig.1 IR Spectrum of 30-carbomethoxy lupenyl acetate(3)

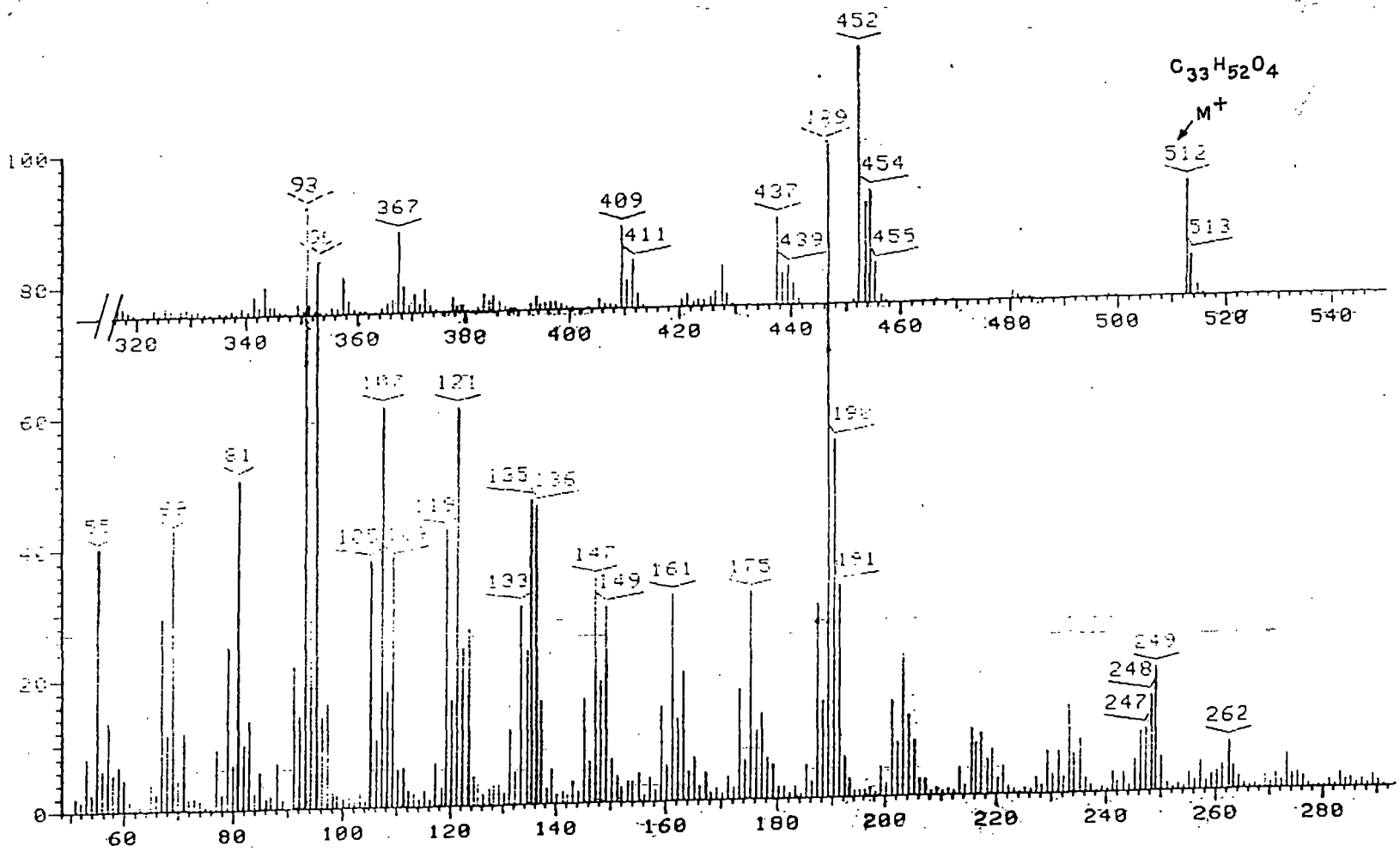


Fig.2 Mass spectrum of 30-carbomethoxy lupenyl acetate(3)

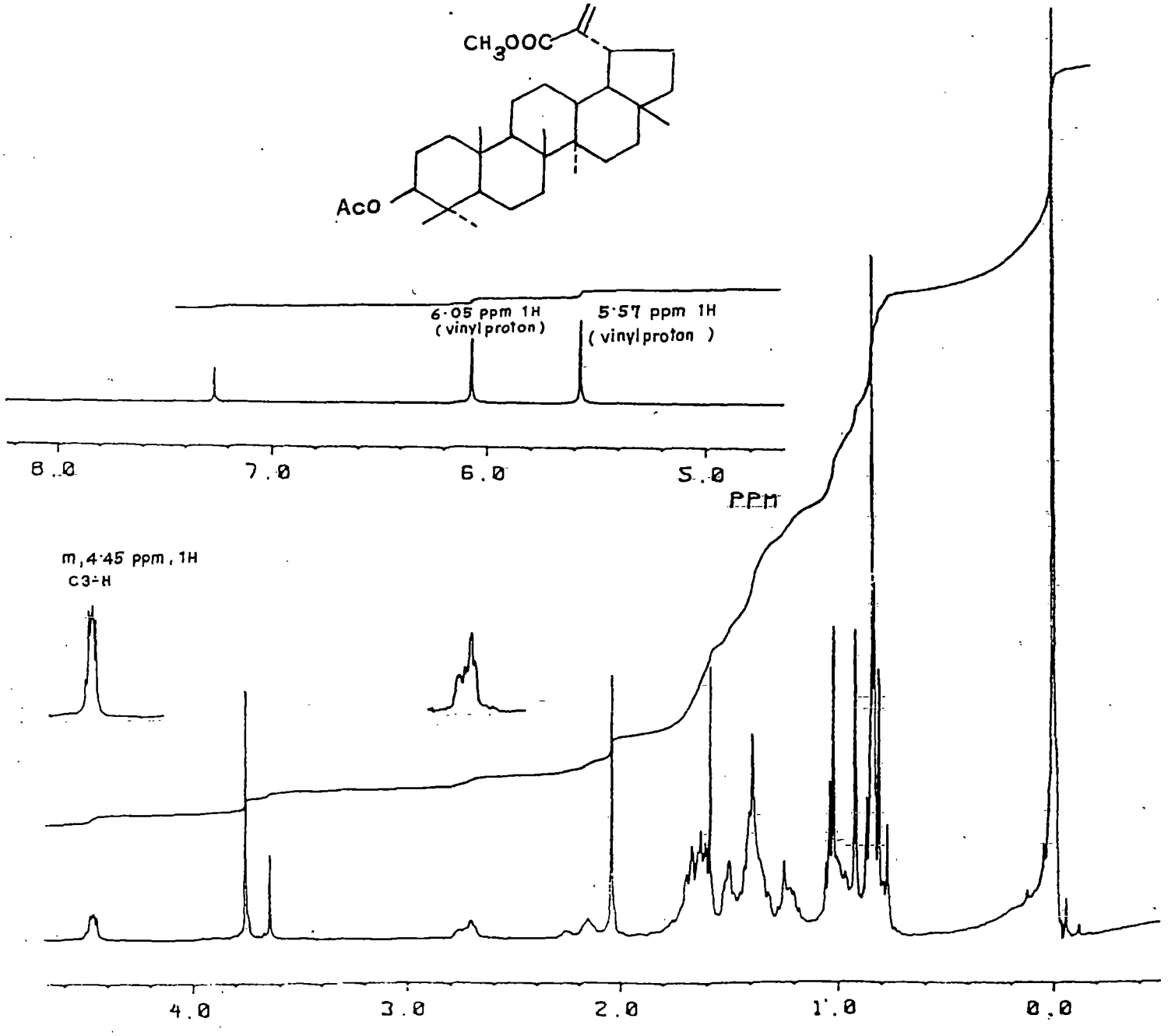
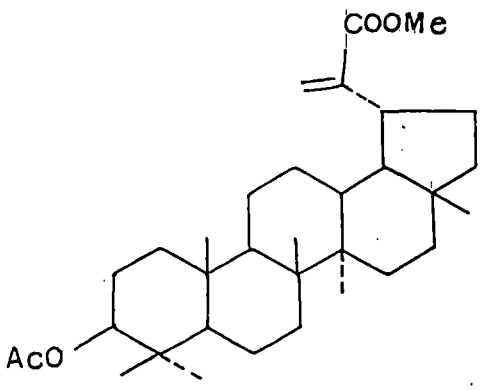


Fig.3 ^{1}H NMR Spectrum of 30-carbomethoxy lupenyl acetate(3)

ppm showed the presence of carboxy methyl group. A singlet at 2.06 ppm indicated three protons of acetoxy methyl group ($-OCOCH_3$) besides the two vinyl protons appearing as singlets at 5.57 and 6.05 ppm. The downward shift of 1.2 ppm compared to that of methylene protons of 1 could be accounted for the consecutive effect of the carbomethoxy group which is at the allylic position. The ^{13}C NMR spectrum (Fig. 4) of A showed 33 resonating peaks and the ATP experiments indicated the existence of (i) eight methyl carbons as quartets (ii) eleven methylene carbons as triplets (iii) six methine carbons as doublets and (iv) eight tertiary carbons as singlets. The C - 20 carbon at 147.08(S), shifted upfield by about 3.5 ppm and the C-29 carbon significantly shifted downfield by about 13.5 ppm than the parent acetate[75] 1 (See Table 1) due to electromeric effect of C-30 carbomethoxy group. Thus from spectral analysis the structure of the compound A has been established as 30-carbomethoxy lupenyl acetate 3.



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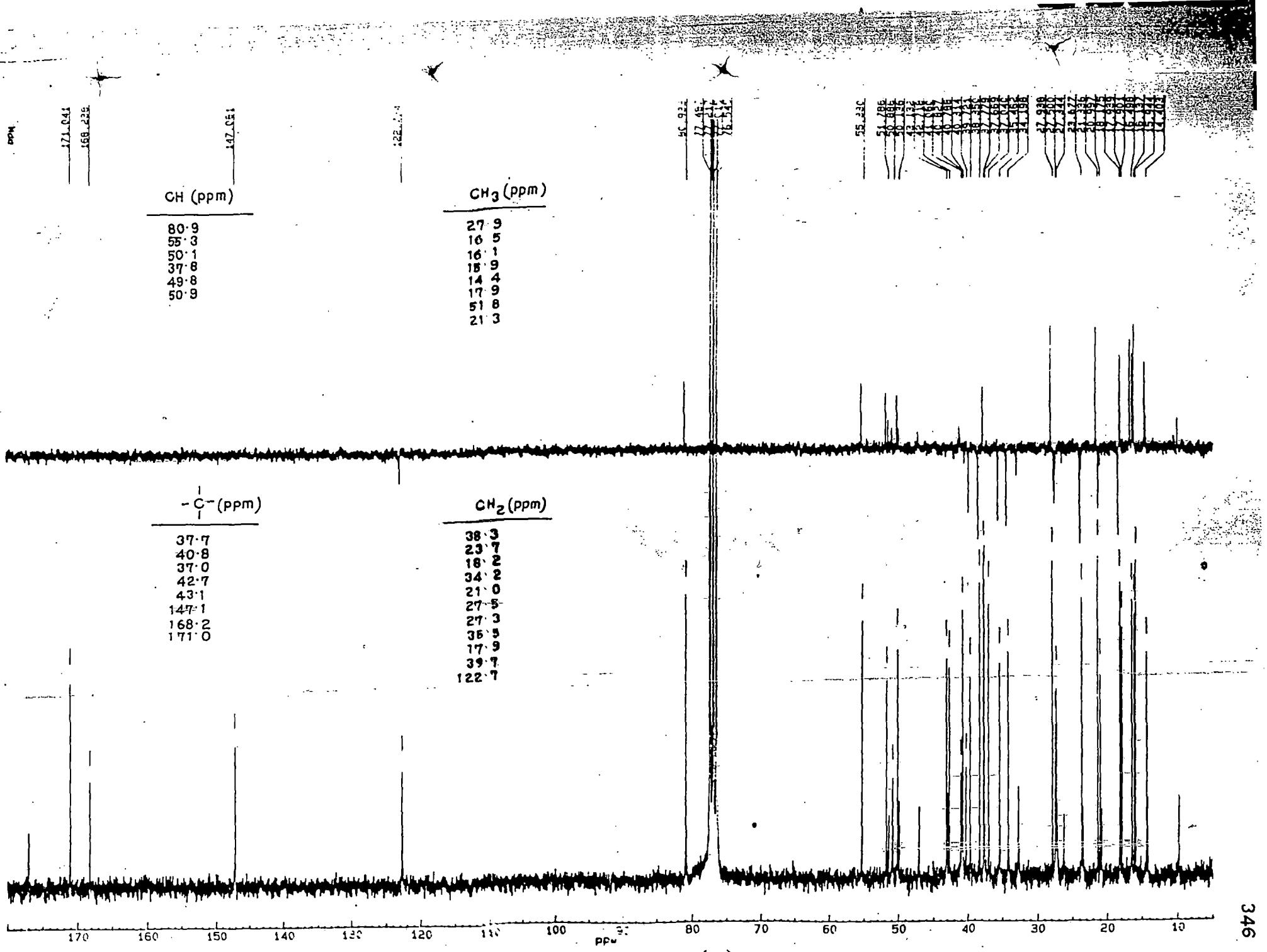
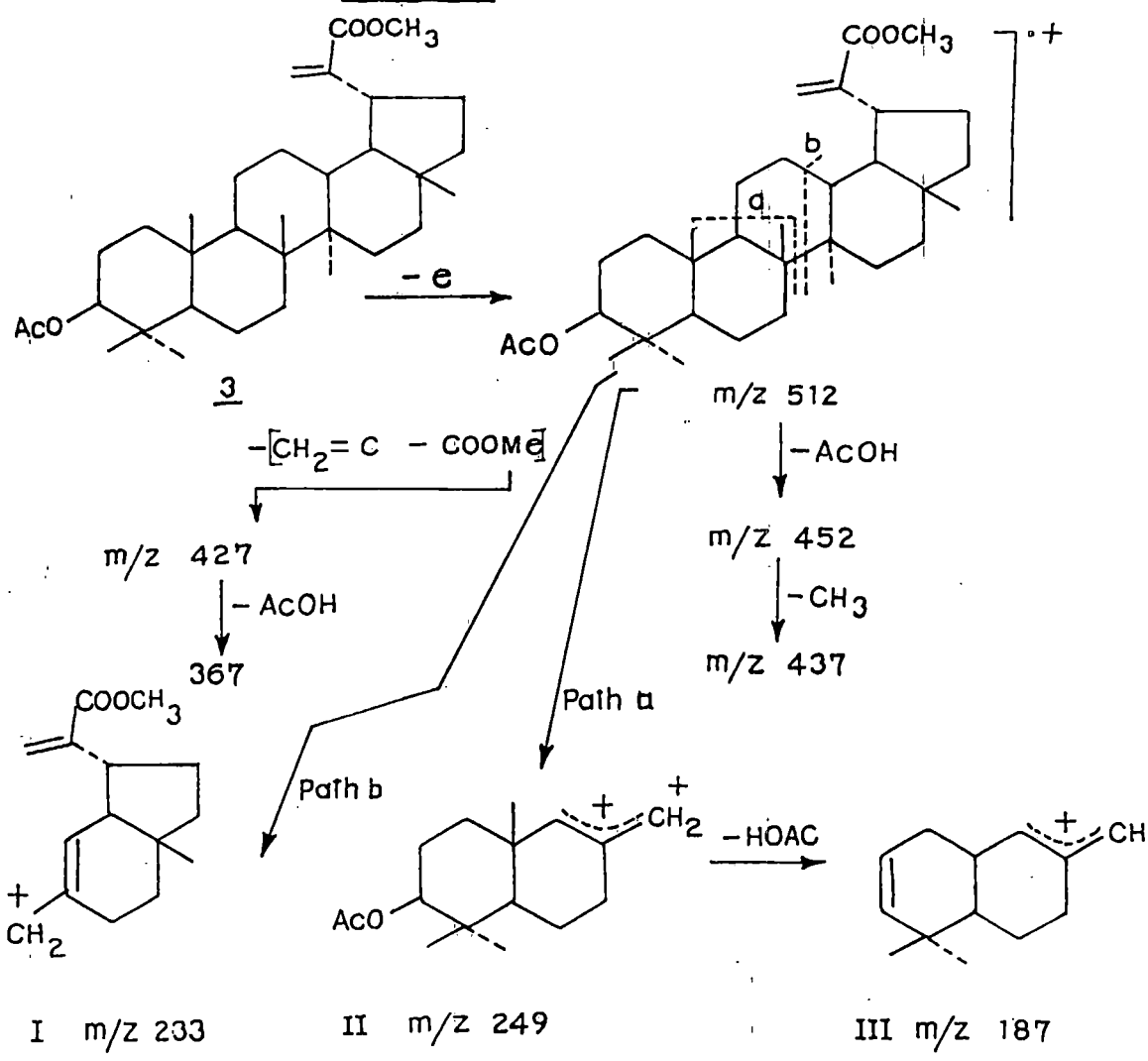


Fig.4 ^{13}c NMR Spectrum of 30-carbomethoxy lupenyl acetate(3)

The structure 3 for compound A gets further support from mass spectrum analysis. The mass spectrum (Fig. 2) of compound A exhibits peaks at m/z 512 $[M]^+$, 452 $[M - CH_3COOH]^+$, 437 $[452 - CH_3]^+$, 427 $[M - CH_2 = C - COOMe]^+$, 411, 409, 367 $[427 - CH_3COOH]^+$, 249, 189 (base peak), 93. A probable mass fragmentation pattern satisfying structure 3 for compound A is depicted in the Scheme I.

Scheme I



Fragments I, II and III are common for lupane skeleton[76].

Characterization of B : Structure elucidation of 30-carbomethoxy-lupan-3 β, 29-diyl acetate 4

Further elution of the chromatogram with petroleum ether : benzene (1:4) furnished compound B, analysed for $C_{35}H_{56}O_6$, m.p. 240-41°. It did not respond to the TNM test for unsaturation indicating the absence of active unsaturation in B. IR spectrum (Fig. 5) of compound B showed a broad band each in the region 1700-1750 cm^{-1} and 1235-50 cm^{-1} for carbonyl groups of acetates and esters. The PMR spectrum (Fig. 6) showed the presence of only six tertiary methyls resonated at 0.75, 0.84, 0.85, 0.87 and 1.09 ppm, two acetoxy methyls at 2.05 and 2.06 ppm and a carboxy methyl at 3.69 (S, 3H)ppm. The AB quartet at 4.29 ppm integrating for two protons with J values of 10.5 and 3.4 Hz was due to the methylene protons geminal to the newly introduced acetoxyl group at C-29. The mass spectrum (Fig. 7) of the compound B showed the molecular ion peak at m/z 512 $[M - CH_3COOH]^+$, other peaks of prominence appeared at 497 $[512 - CH_3]^+$, 469, 452 $[512 - CH_3COOH]^+$, 430, 367, 193, 81 (base peak). These data evidently showed B to be 30-carbomethoxy lupan-3β, 29- diyl acetate 4.

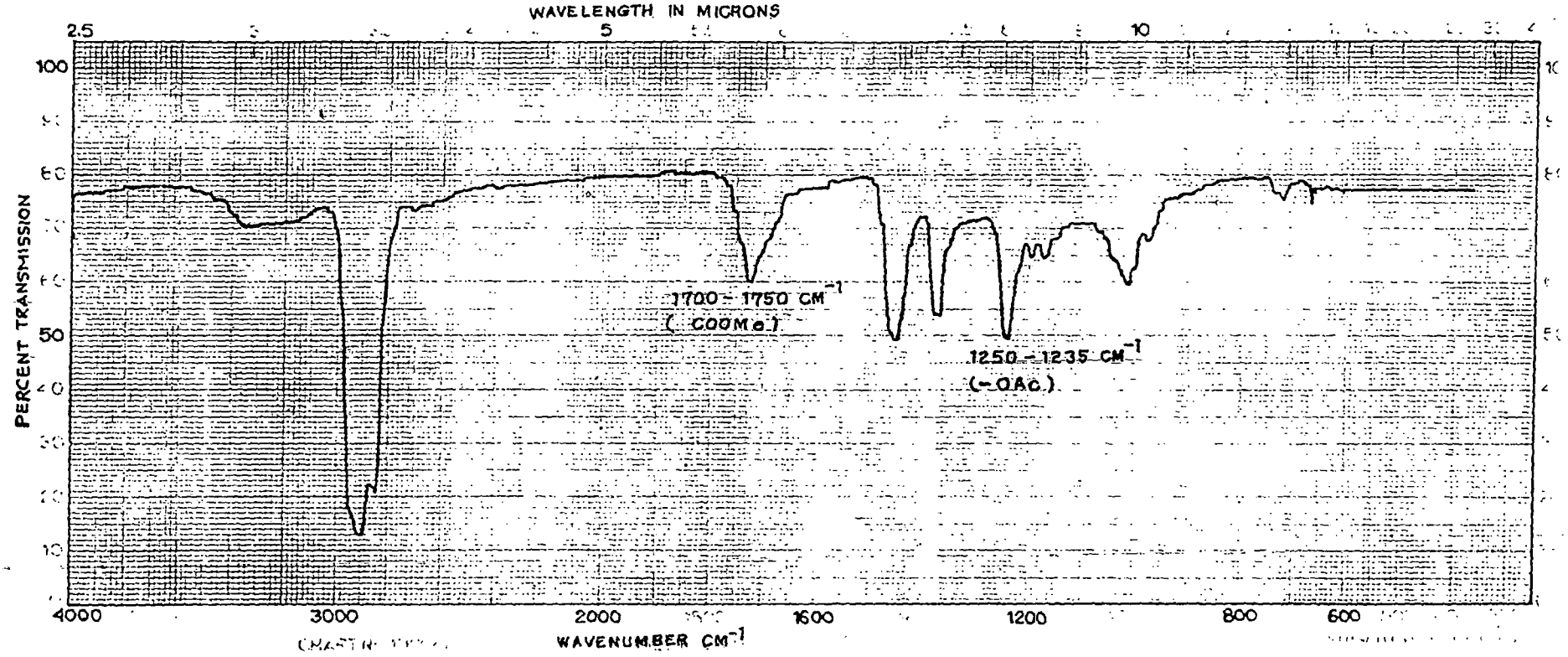


Fig.5 IR Spectrum of 30-carbomethoxy lupan-3β,29-diyl acetate(4)

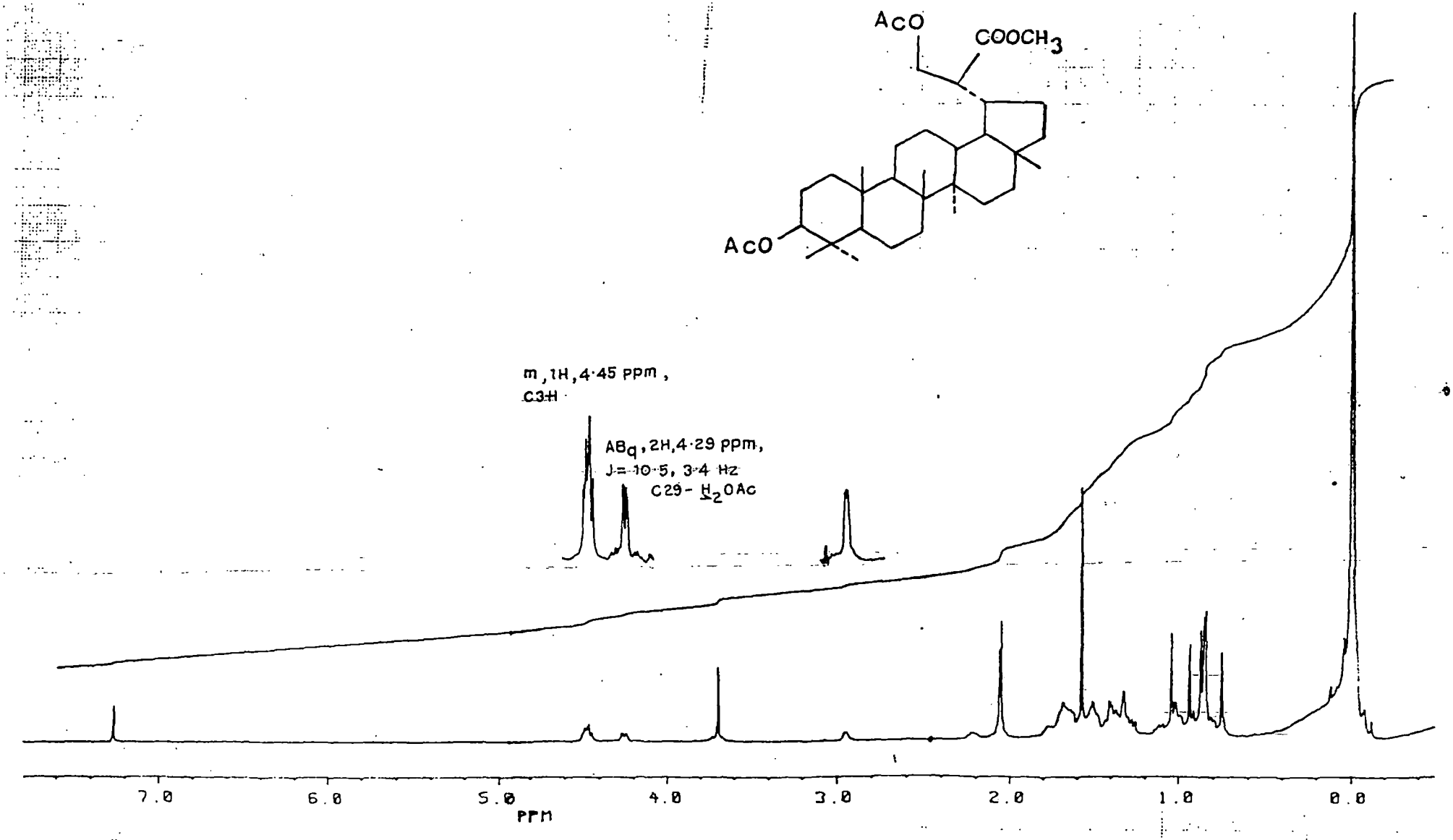


Fig.6 ^{1}H NMR Spectrum of 30-carbomethoxy lupan-3β,29-dilyl acetate(4)

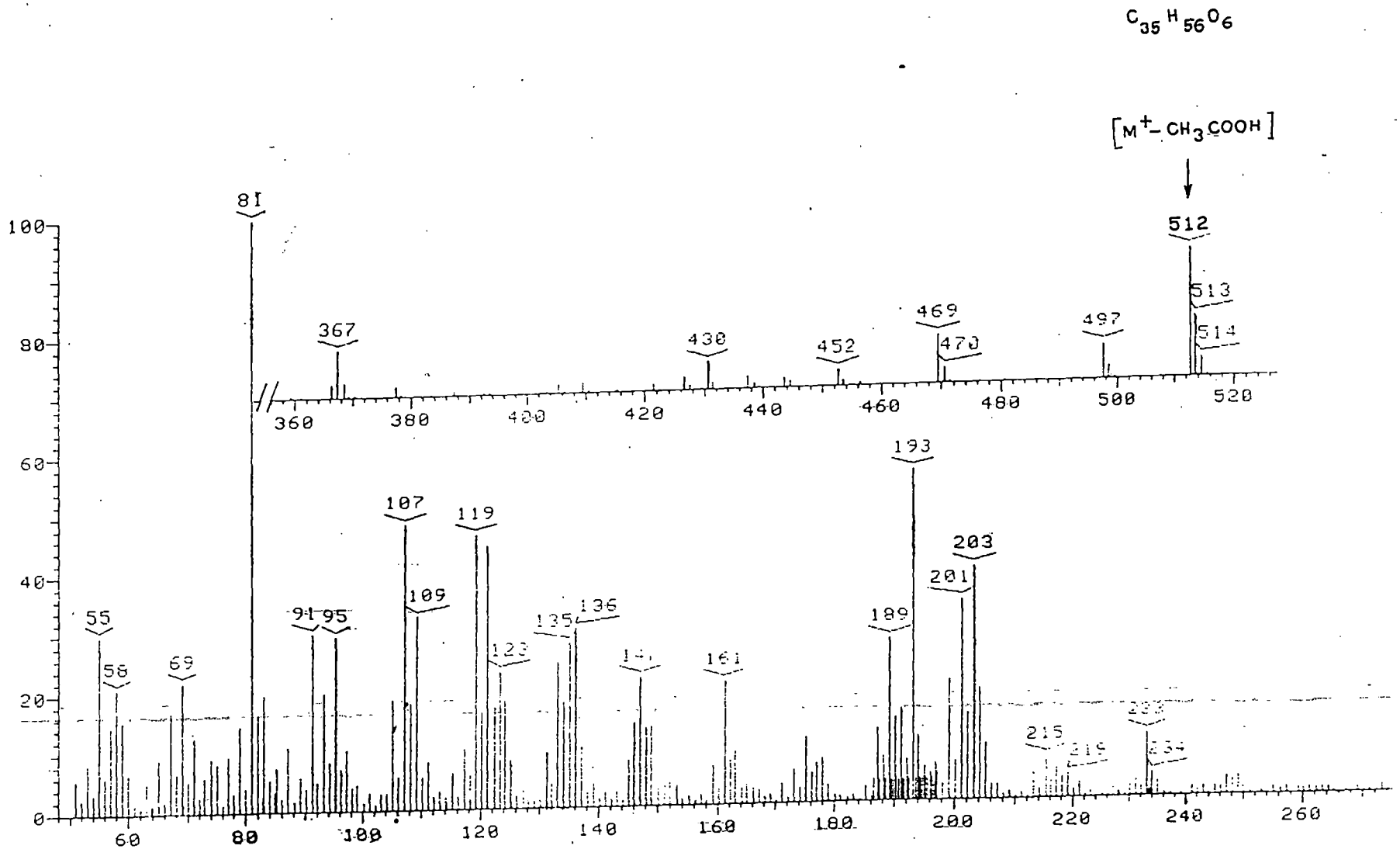
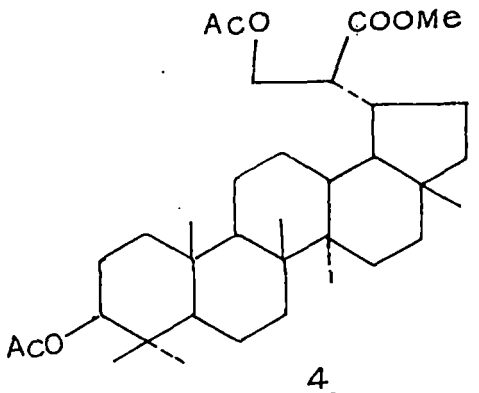


Fig.7 Mass spectrum of 30-carbomethoxy lupan-3β,29-diyl acetate(4)



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The structure 4 for B was substantiated by its ^{13}C NMR spectrum (Fig. 8) which showed the presence of 35 carbons; the DEPT experiment indicated the presence of nine - CH_3 carbons appearing as quartets of which those resonating at 51.8 and 21.1 ppm and 213 ppm were due to a carbomethoxy and two acetoxy methyl carbons respectively, eleven -$\dot{C}H_2$ carbons as triplets and the one at 61.4 ppm was due to methylene carbon having the second acetoxyl group, seven -$\dot{C}H$ carbons as doublets and eight quaternary carbons as singlets, of which the methoxy carbonyl carbon appeared at 174.3 ppm and the two acetoxy carbonyl carbons at 171.0 and 170.9 (Table 1)

Characterization of C : Structure elucidation of 29-carbomethoxy-lupan-3β-yl acetate 5

The more polar fraction eluted with benzene was purified by crystallisation. The crystallisation afforded fine needle shaped crystals of compound C, analysed for $C_{33}H_{54}O_4$, m.p. 237-38°. It did not respond to the TNM test for unsaturation. IR

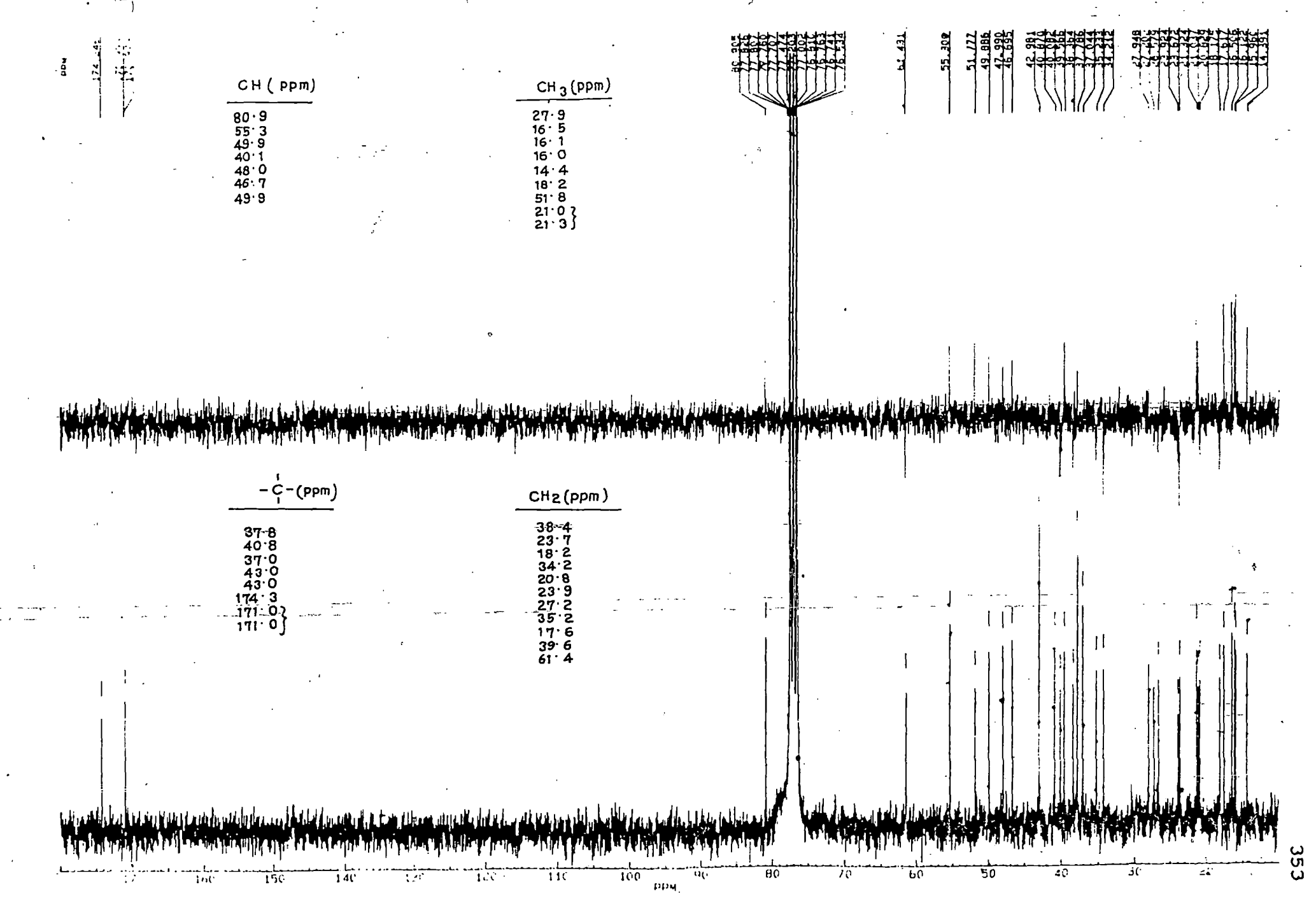
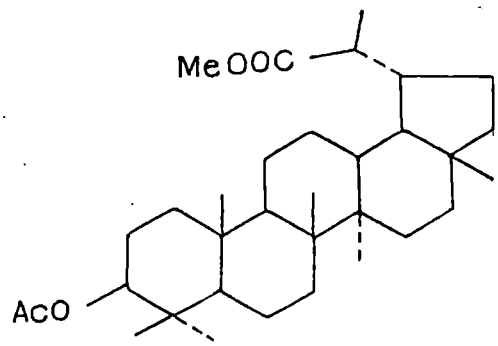


Fig.8 ^{13}c NMR Spectrum of 30-carbomethoxy lupan-3β,29-diyl acetate(4)

spectrum (Fig. 9) of the compound C showed peaks at 1730-40 cm^{-1} and 1252 cm^{-1} for methoxy carbonyl and acetate carbonyl groups. Its PMR spectrum (Fig. 10) showed the presence of six tertiary methyl groups that appeared as singlets in the region 0.74 to 1.04 and a secondary methyl appearing as a doublet at 1.24 ppm (J = 7 Hz); the latter must be the methyl group at position C-30; a singlet at 2.05 ppm integrated for three protons indicated the presence of one acetoxy methyl group ($-OCOCH_3$). The singlets appearing at 3.64 and 3.67 ppm integrated for three protons suggesting that the carbomethoxy group at C-29 exists in two stereochemical orientations. Thus assuming that the oxidation has occurred only at the C-29-carbomethoxy lupan-3β-yl acetate 5. The isomeric nature of C was also exhibited in the ^{13}C NMR spectrum (Fig. 11), which displayed double resonances



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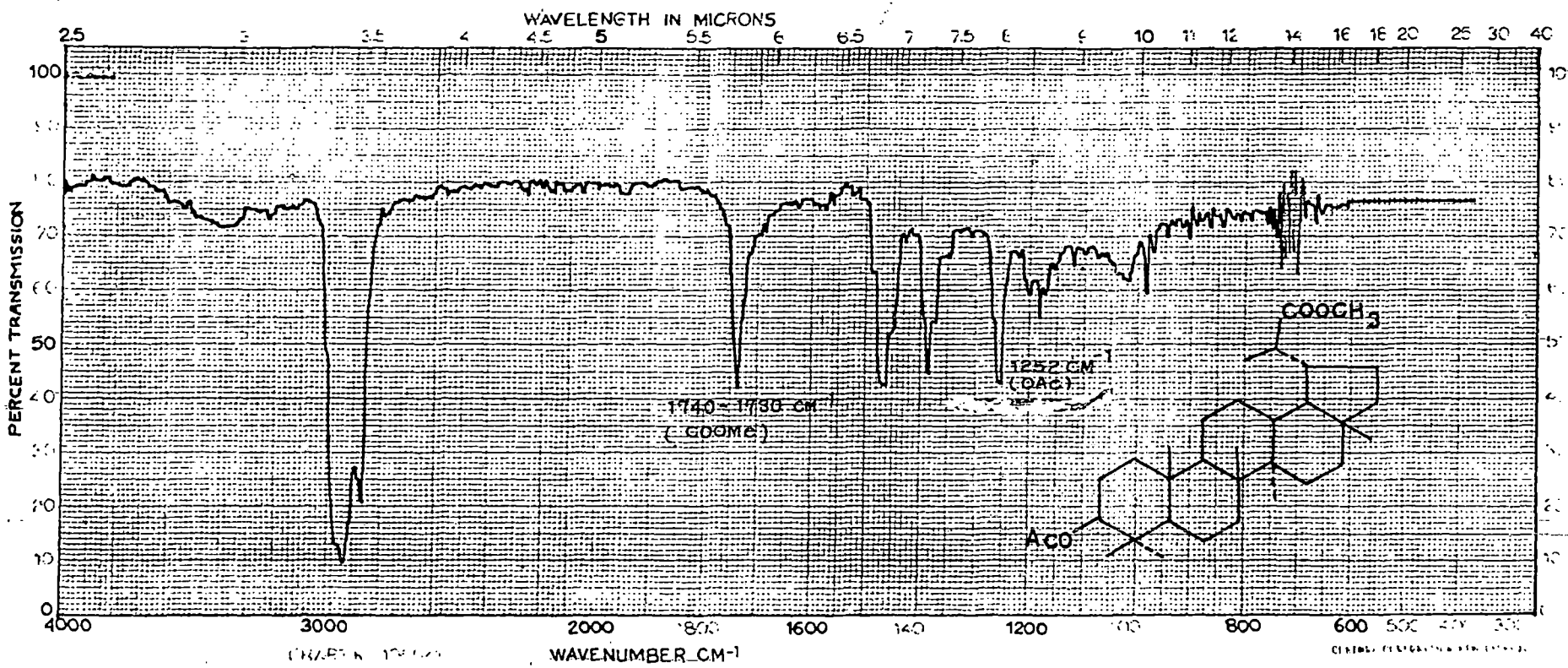


Fig.9 IR Spectrum of 29-carbomethoxy lupan-3B-yl acetate(5)

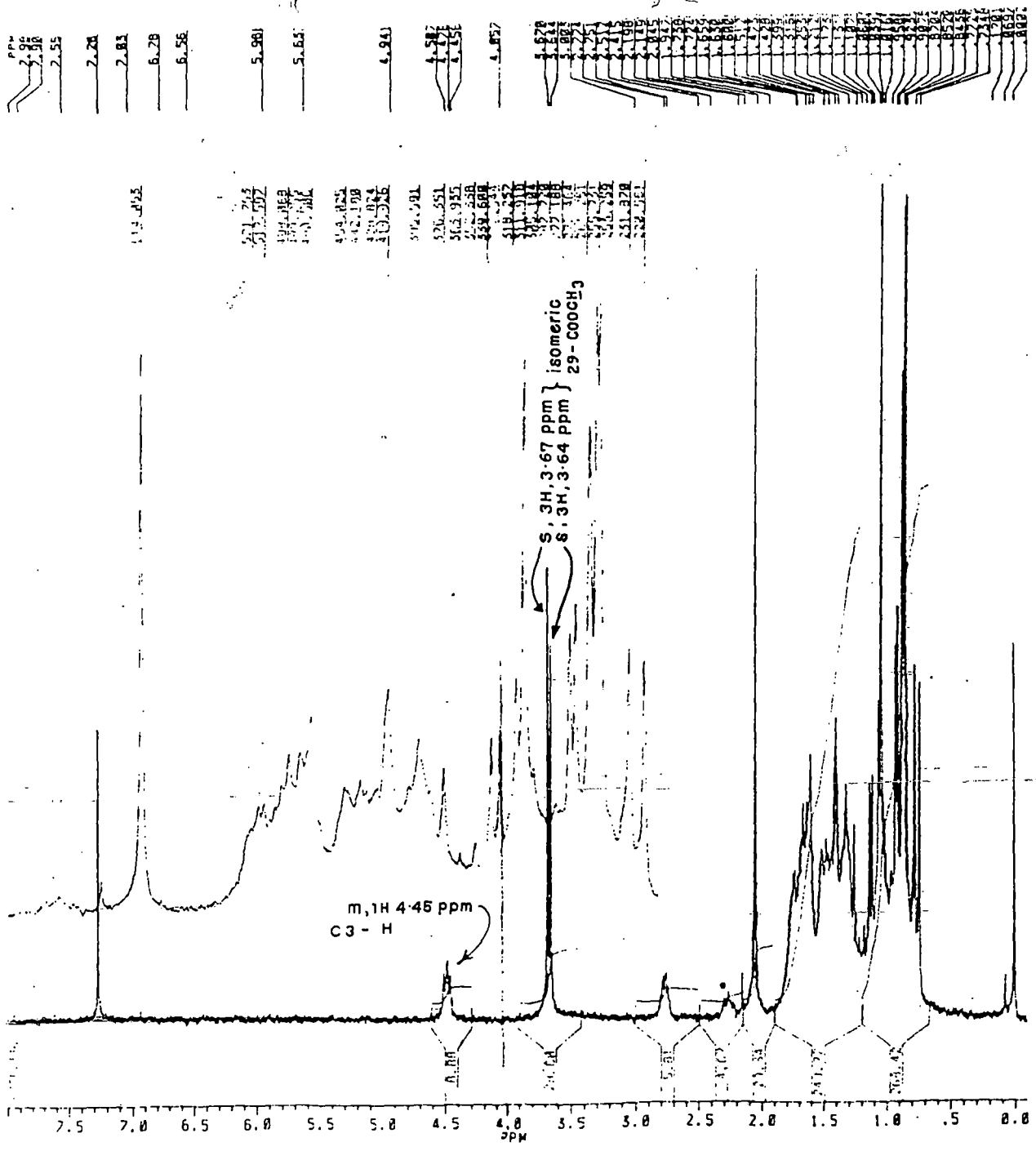


Fig.10 ^{1}H NMR Spectrum of 29-carbomethoxy lupan-3β-yl acetate(5)

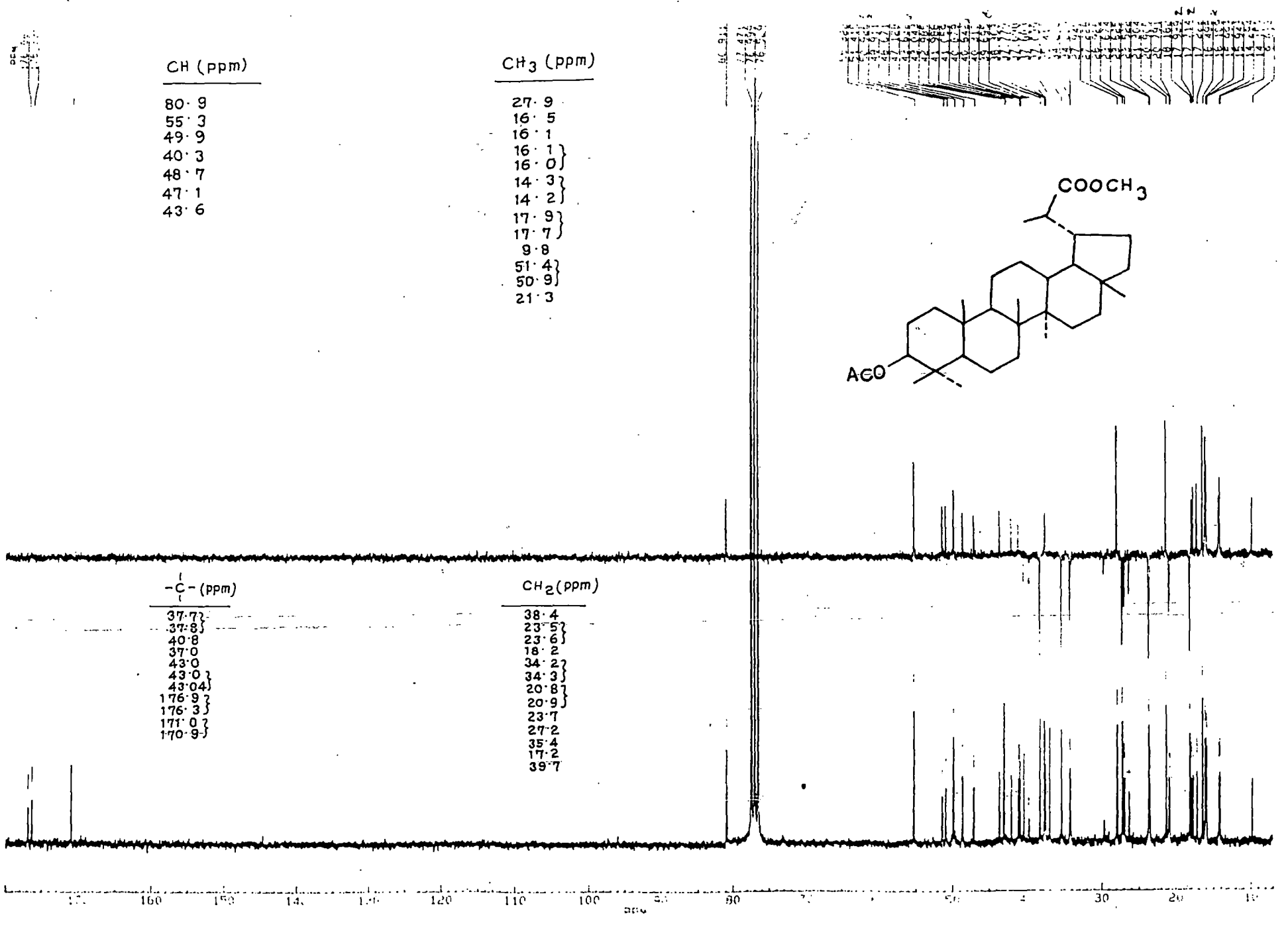


Fig.11 ^{13}c NMR Spectrum of 29-carbomethoxy lupan-3β-yl acetate(5)

for some of its carbons (Table 1). The isomeric 29-carbomethoxy lupan-3 β-yl acetate has been reported in literature[77]. The structure 5 for compound C is further established by mass spectrum analysis. Besides molecular ion peak at m/z 514 $[M^+]$, the mass spectrum (Fig. 12) of compound C shows other peaks at m/z 499 $[M - CH_3]^+$, 454 $[M - CH_3COOH]^+$, 439 $[454 - CH_3]^+$, 427 $[M - CH_3 - CH - COOMe]^+$, 411 $[427 - CH_4]^+$, 367 $[427 - CH_3COOH]^+$, 275, 249, 233, 189, 175, 163, 107, 95, 57 (base peak). A probable mass fragmentation pattern assuming structure 5 for compound C is represented in Scheme II.

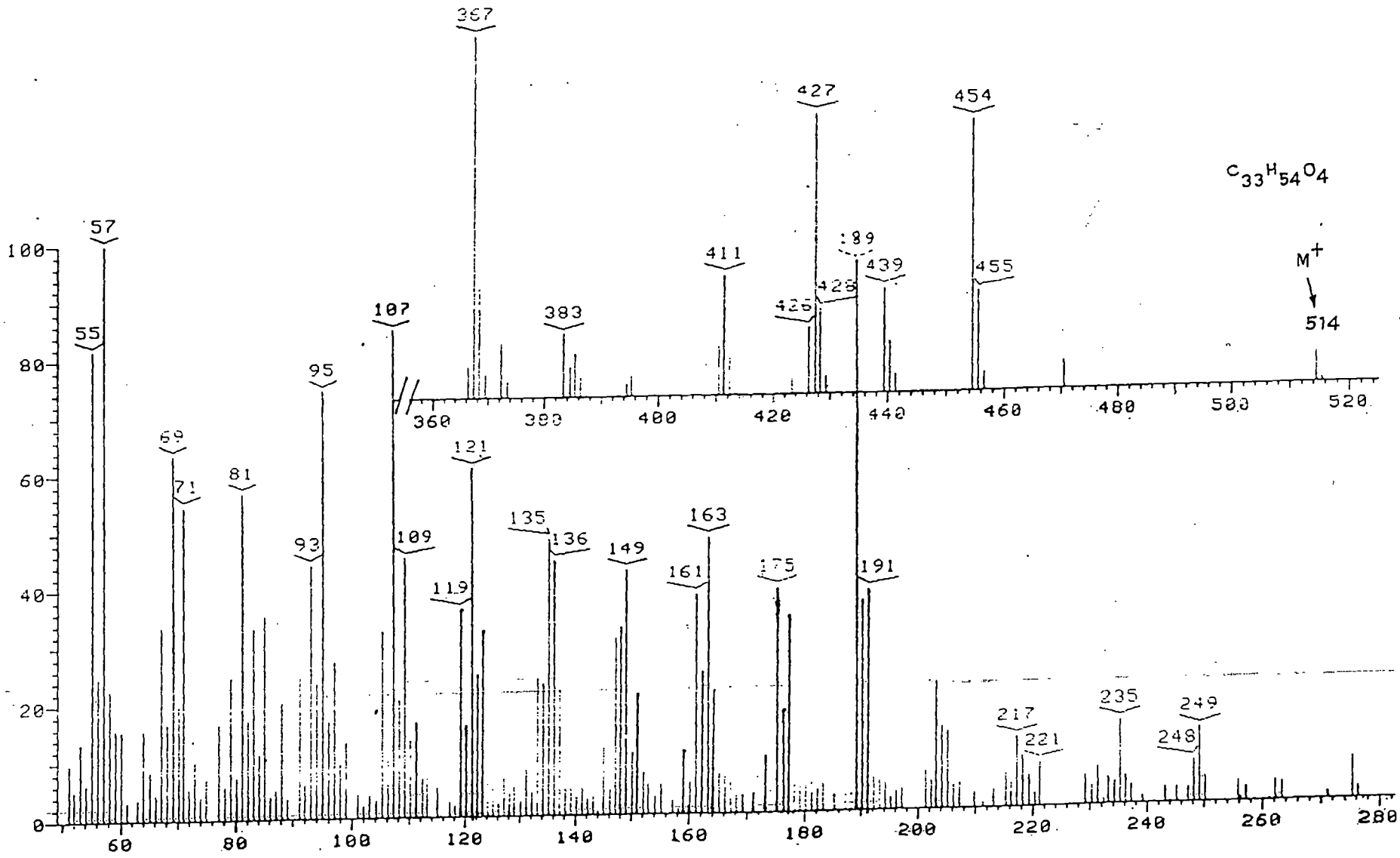
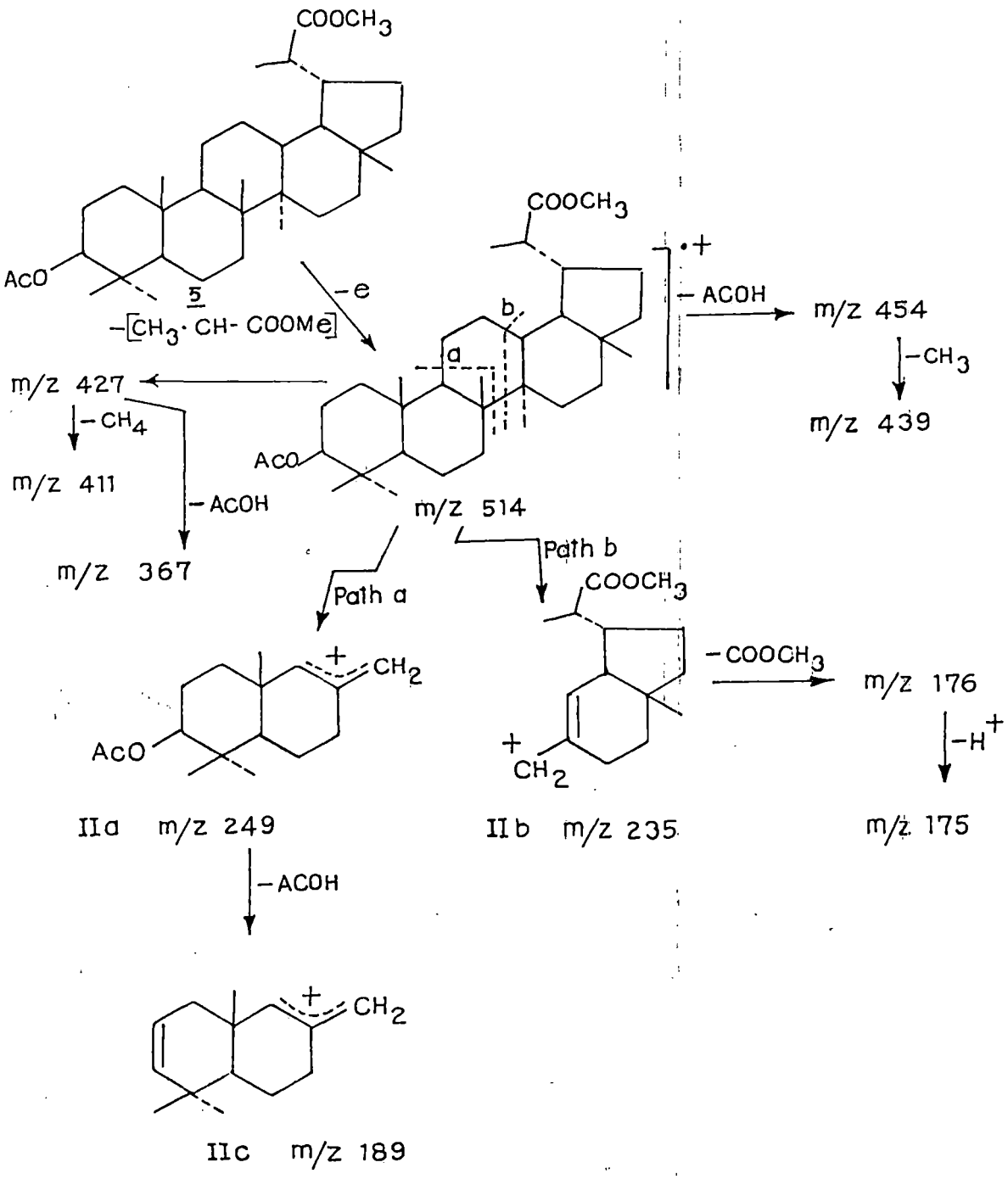


Fig.12 Mass spectrum of 29-carbomethoxy lupan-3β-yl acetate (5)

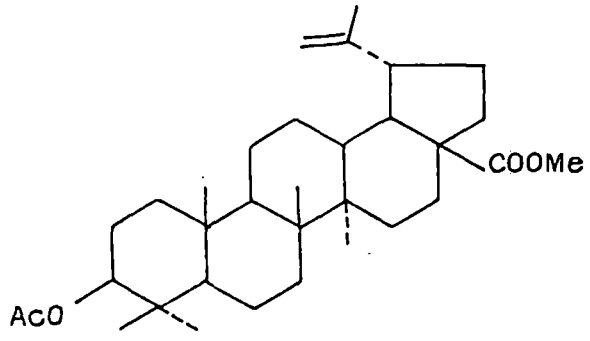
Scheme II



The fragments IIa, IIb and IIc are characteristics for lupane skeleton[76].

B. OXIDATION OF METHYL ACETYL BETULENATE 2 WITH SELENIUM DIOXIDE AND HYDROGEN PEROXIDE IN TERTIARY BUTANOL: ISOLATION OF 28, 30-DICARBOMETHOXY LUPENYL ACETATE 6 AND 28, 29-DICARBOMETHOXY-LUPAN-3β - YL ACETATE 7.

Methyl acetyl betulenate on similar treatment (as discussed above) with selenium dioxide-hydrogen peroxide in tertiary butanol yielded an oily residue which on esterification (CH_2N_2) and acetylation (pyridine-acetic anhydride) gave a material. Chromatographic separation of the material so obtained, afforded only two products D and E.



2

1. SeO_2-H_2O_2 → D + E
2. Esterification (CH_2N_2)
3. Acetylation (Py/Ac_2O)

Characterization of D : Structure elucidation of 28, 30-dicarbomethoxy lupenyl acetate 6

Elution of the column with petroleum ether: benzene (2:3) furnished a compound, which on crystallisation afforded fine crystals of compound D, analysed for $C_{34}H_{52}O_6$, m.p. 150-51°. Compound D gave yellow colouration with TNM indicating the presence of double bond. Its IR spectrum (Fig. 13) exhibited absorptions at 1730-40 and 1250 cm^{-1} for the ester and the acetate

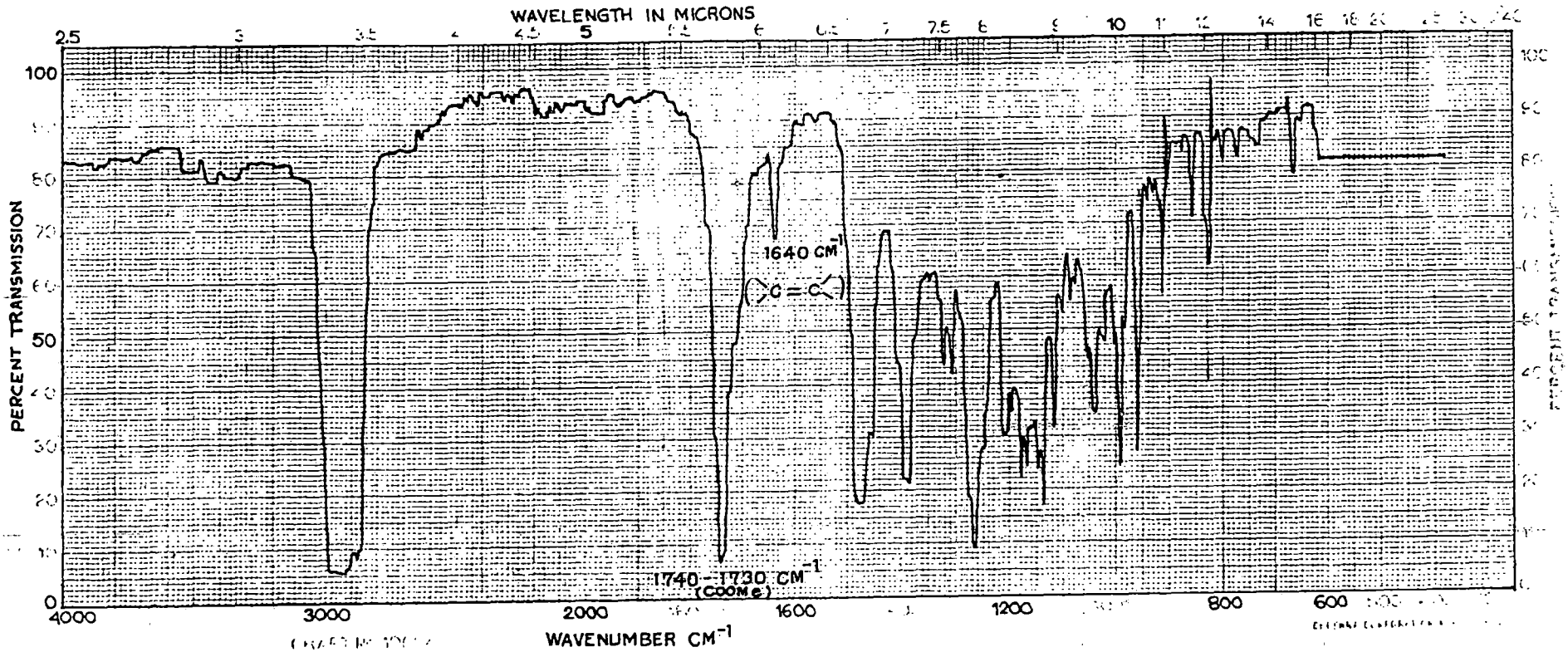


Fig.13 IR Spectrum of 28,30-dicarbomethoxy lupenyl acetate(6)

carbonyl groups (O - $\overset{\overset{O}{\|}}{C}$ - CH_3) and bands at 1640 cm^{-1}, 830 cm^{-1} for the methylenic double bond. PMR spectrum (Fig. 14) of the compound D showed the presence of five tertiary methyl groups that appeared as singlets in the region 0.81 to 0.92 ppm, a singlet at 2.02 ppm integrated for three protons indicated presence of one acetoxy methyl group (O-$COCH_3$), two carboxy methyls at 3.68 and 3.74 ppm and two methylene protons appeared as singlets at 5.57 and 6.05 ppm indicated that the C-30 carbon of 2 had been converted to carbomethoxy group giving rise to 6. ^{13}C NMR spectrum (Fig. 15) of the compound D and DEPT experiment indicated the existence of eight methyl carbons as quartets, eleven methylene carbons as triplets six methine carbons as doublets and nine tertiary carbons as singlets; the singlets at 170.9 (3 - $\overset{|}{C}$ - O-$COCH_3$), 176.6 (28 - $COOCH_3$) and 167.7 (30 - $COOCH_3$) ppm were due to ester carbonyls; the singlets at 146.5 ppm and the triplet at 123.5 were due to the olefinic carbon at C-20 and C-29 (Table 1). It is to be noted that compound 6 also shows similar shifts in the resonance positions due to C-29 and C-30 carbons in comparison to 2. Thus from above spectral data (PMR, IR and ^{13}C NMR) the compound D has been identified as 28, 30-dicarbomethoxy lupenyl acetate 6.

The structure 6 for compound D is finally supported by its mass spectrum (Fig. 16). Besides the molecular ion peak at m/z 556 $[M]^+$, the other significant peaks at m/z 496 $[M - CH_3COOH]^+$, 481 $[49 - CH_3]^+$, 437 $[496 - COOMe]^+$,

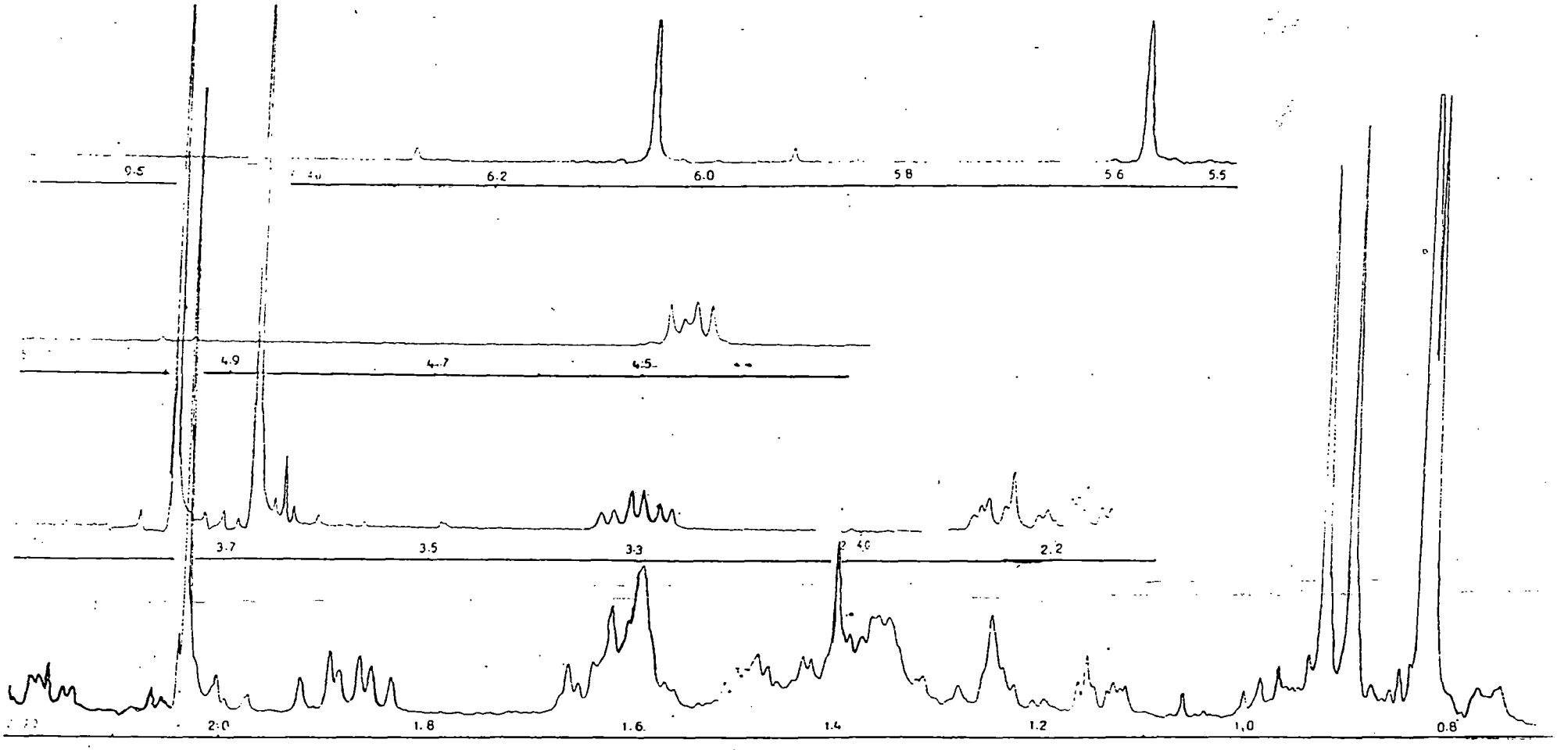


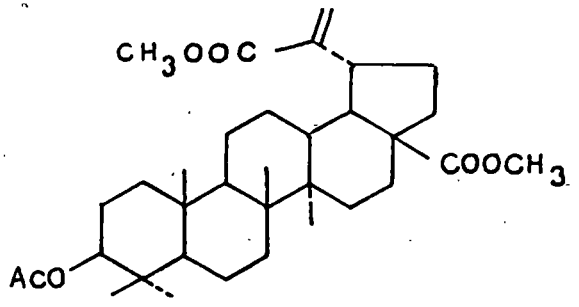
Fig.14 ^{1}H NMR Spectrum of 28,30-dicarbomethoxy lupenyl acetate (6)

CH (ppm)
80·8
55·4
50·2
38·0
50·8
50·8

CH_3 (ppm)
27·9
16·4
16·1
15·9
14·6
51·4
21·3

$-\overset{|}{\underset{|}{C}}-$ (ppm)
37·7
40·6
37·0
42·7
56·6
150·1
176·6
167·7
171·0

CH_2 (ppm)
38·2
23·6
18·1
34·2
20·9
26·9
29·6
32·0
32·3
36·5
123·5



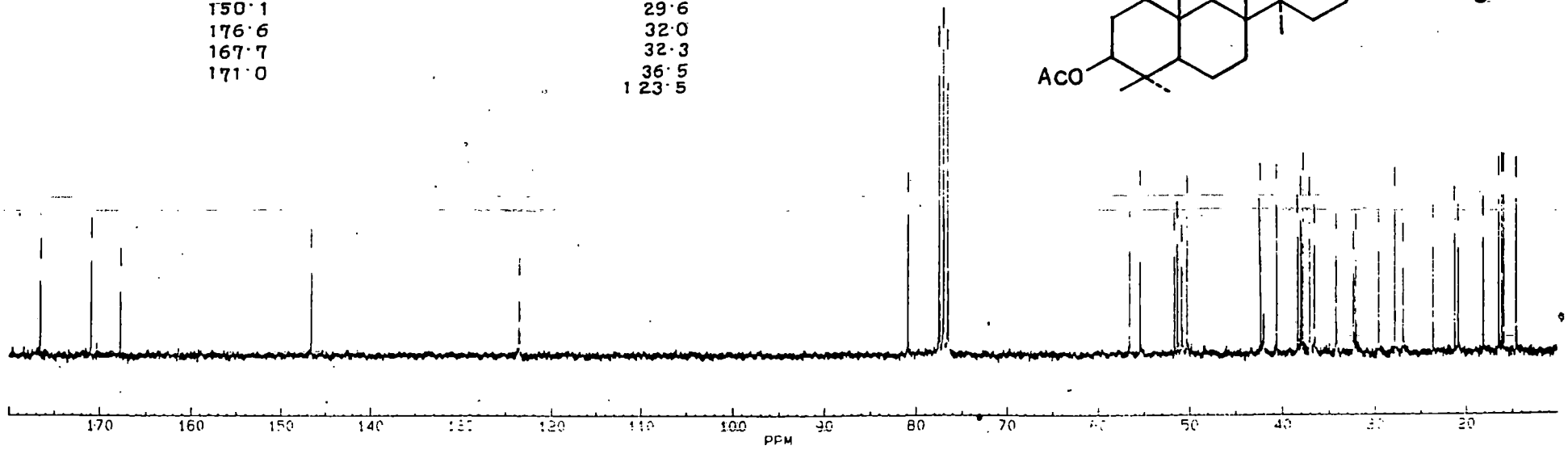


Fig.15 ^{13}c NMR Spectrum of 28,30-dicarbomethoxy lupenyl acetate(6)

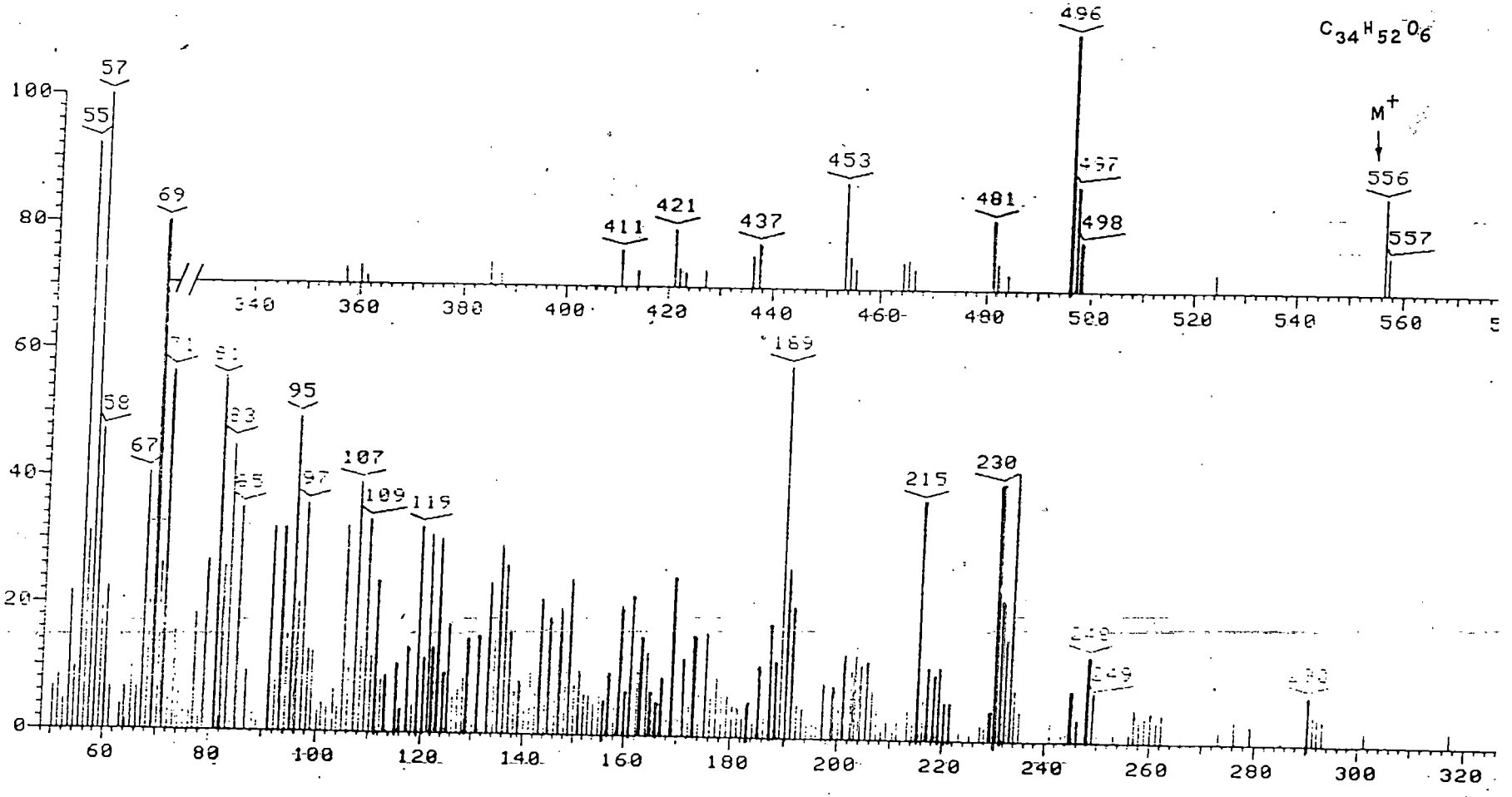
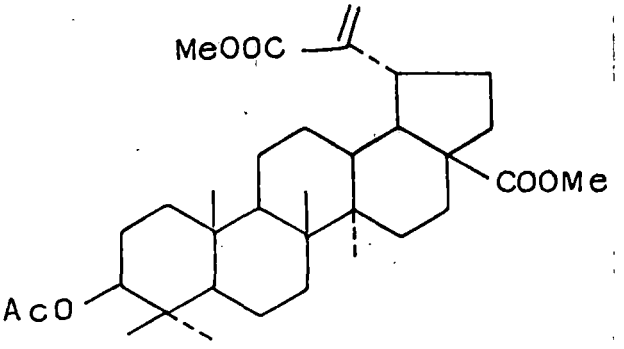


Fig.16 Mass spectrum of 28,30-dicarbomethoxy lupenyl acetate(6)

421 $[481 - CH_3COOH]^+$, 290, 230, 215, 189 (base peak) and 95 appearing in mass spectrum of D can be best rationalised in terms of structure 6.



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Characterization of E : Structure elucidation of 28, 29-dicarbomethoxy-lupan-3 β-yl acetate 7:

Crystallisation of the more polar compound with chloroform-methanol mixture yielded crystals of compound E, analysed for $C_{34}H_{54}O_6$, m.p. 185-86°. It did not respond to the TNM test for unsaturation. IR spectrum (Fig. 17) of the compound E showed absorptions for the ester and the acetate carbonyls at 1730-40 and 1250 cm^{-1}. The PMR spectrum (Fig. 18). showed the presence of five tertiary methyls in the region 0.83 to 0.96 ppm appearing as singlets and a secondary methyl appearing as a doublet at 1.04 ppm with J value equal to 7 Hz

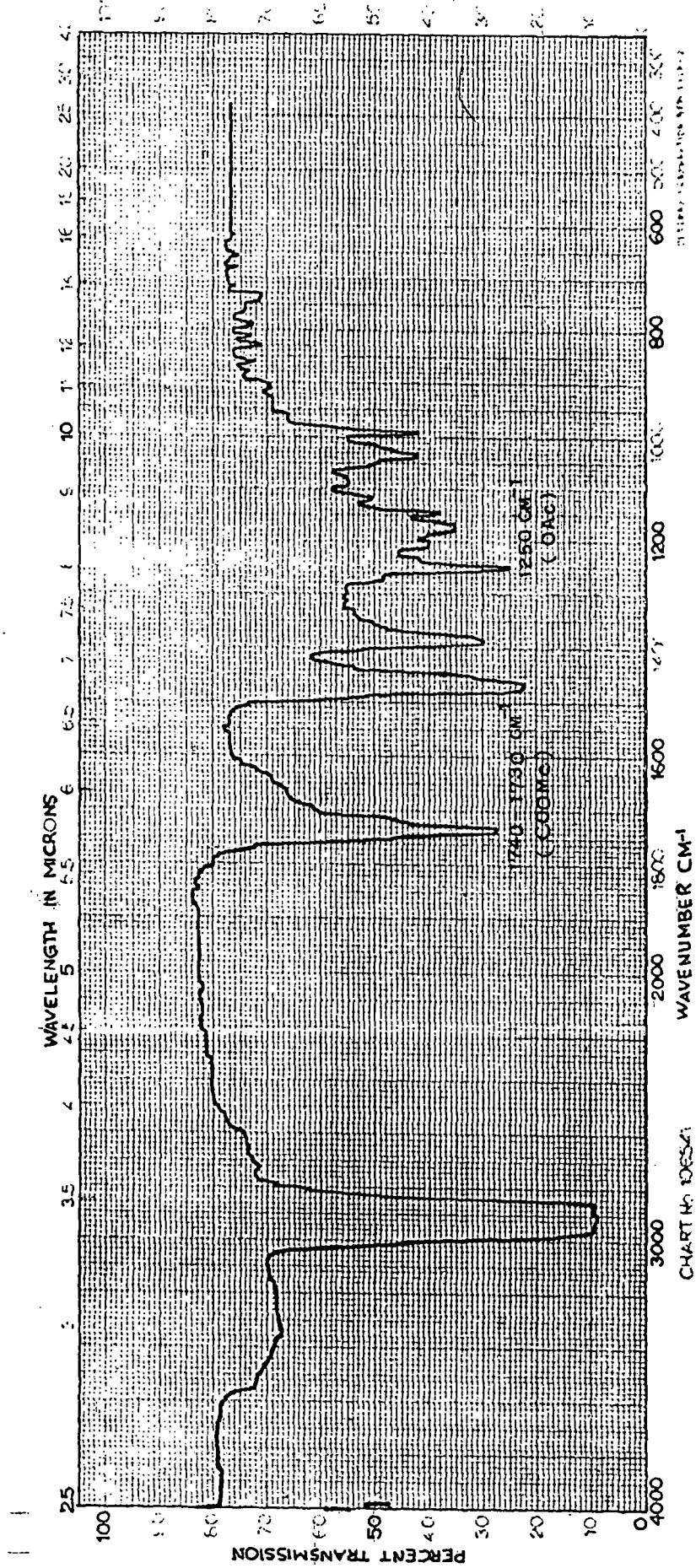


Fig.17 IR Spectrum of 28;29-dicarbomethoxy lupan-3B-yl acetate(7)

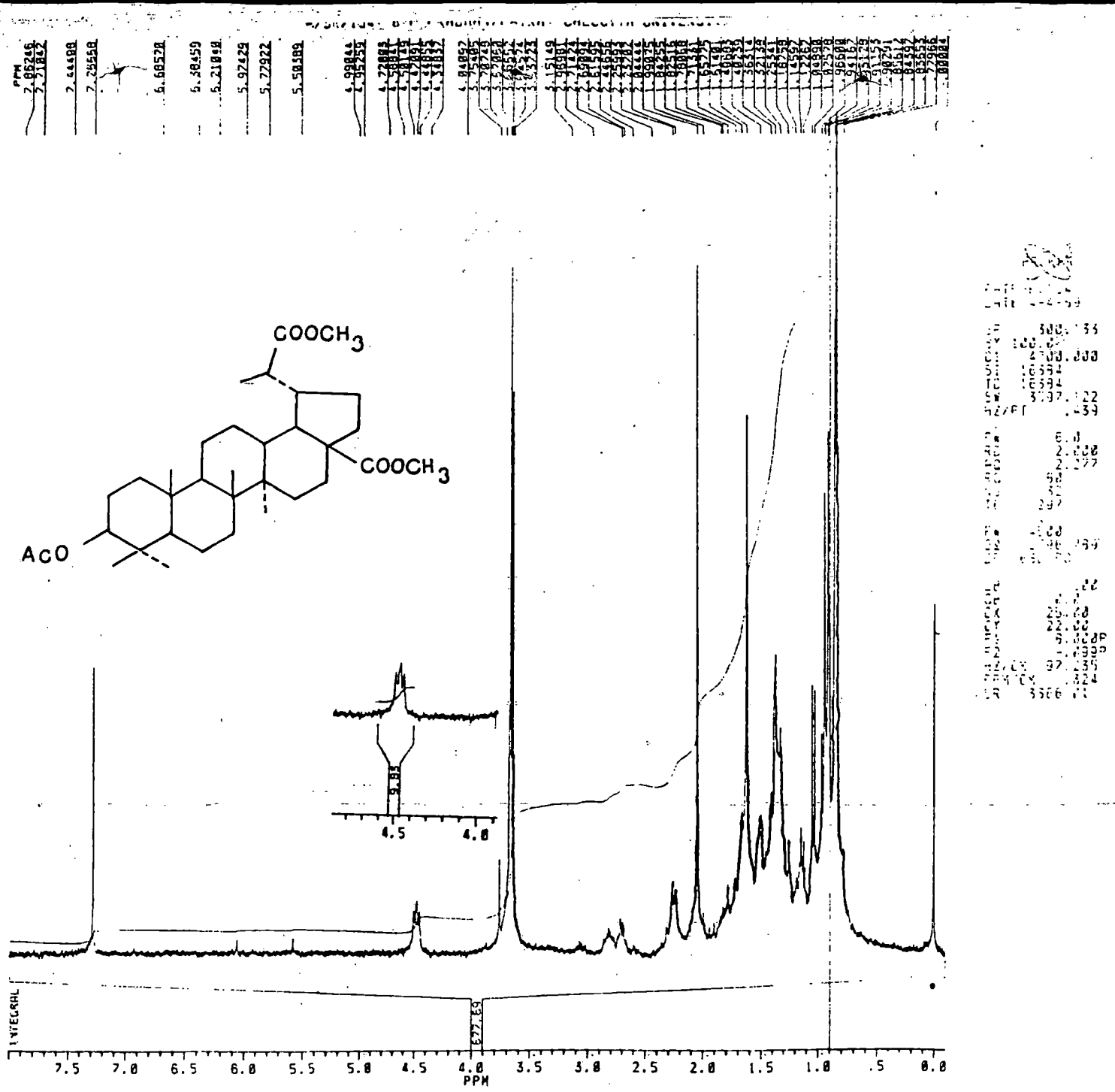
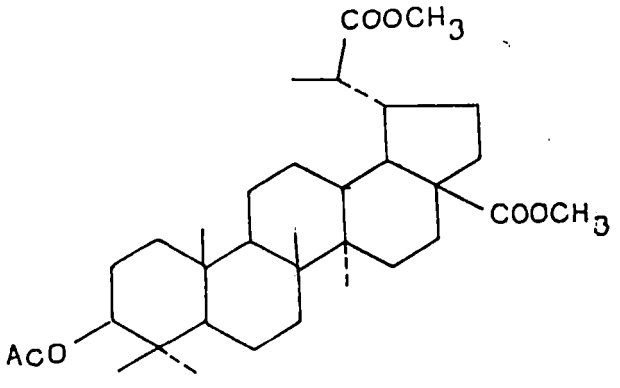


Fig.18 ^{1}H NMR Spectrum of 28,29-dicarbomethoxy lupan-3β-yl acetate(7)

which must be the methyl group at position C-30; a singlet at 2.06 ppm indicated three protons of acetoxy methyl group ($-OCOCH_3$) and two singlets at 3.63 and 3.67 ppm for the two carboxy methyls. This led to the establishment of the structure E as 28, 29-dicarboxy lupan-3 β-yl acetate 7.



7

The structure 7 for compound E gets further support from mass spectrum analysis. The mass spectrum (Fig. 19) of compound E exhibits peaks at m/z 558 $[M]^+$, 498 $[M - CH_3COOH]^+$, 483 $[498 - CH_3]^+$, 439 $[498 - COOMe]^+$, 423 $[483 - CH_3COOH]^+$, 411, 220, 189 (base peak), 175, 147, 136.

A probable fragmentation pattern satisfying structure 7 for compound E is depicted in the Scheme III.

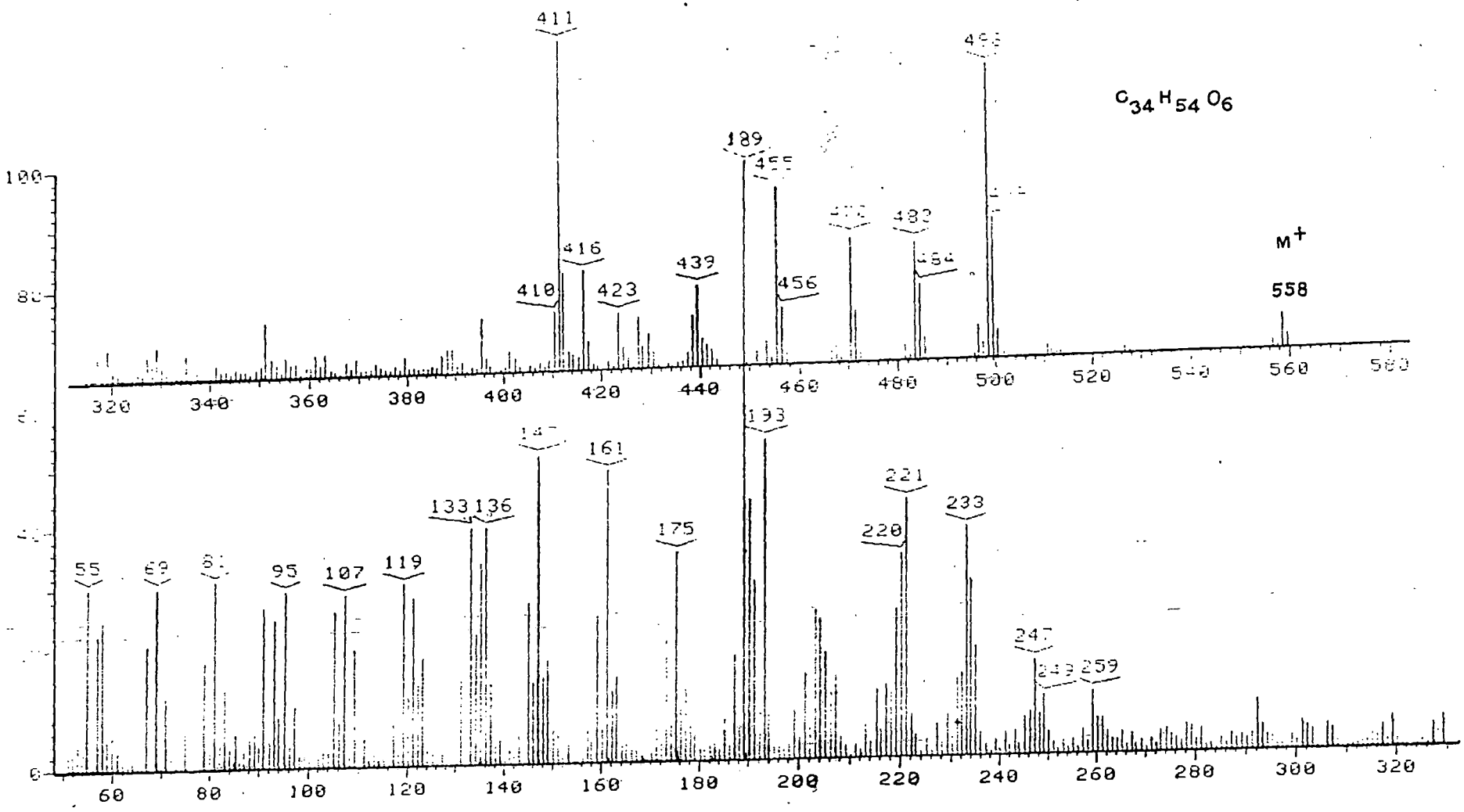
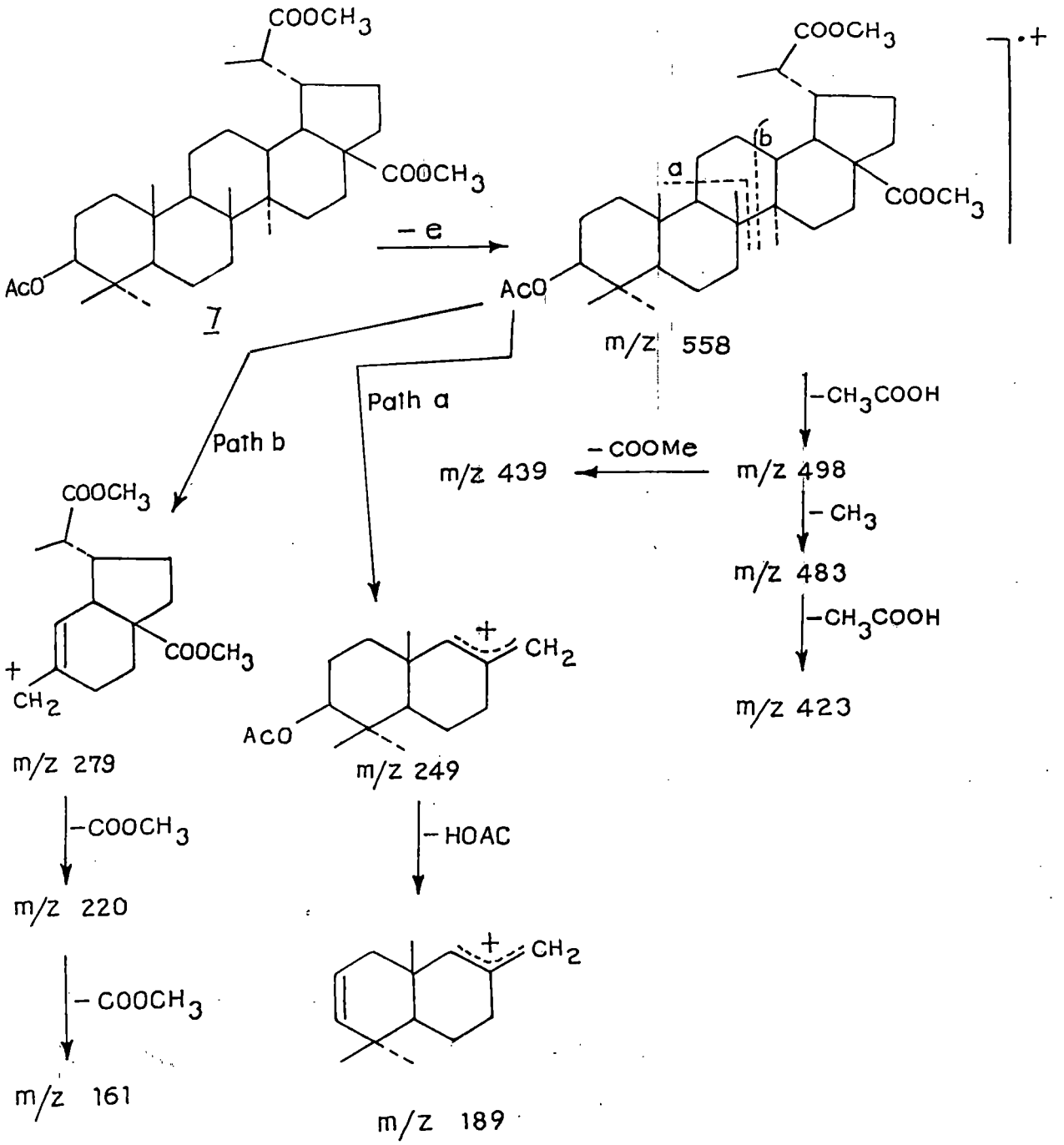


Fig.19 Mass spectrum of 28,29-dicarbomethoxy lupan-3β-yl acetate(7)

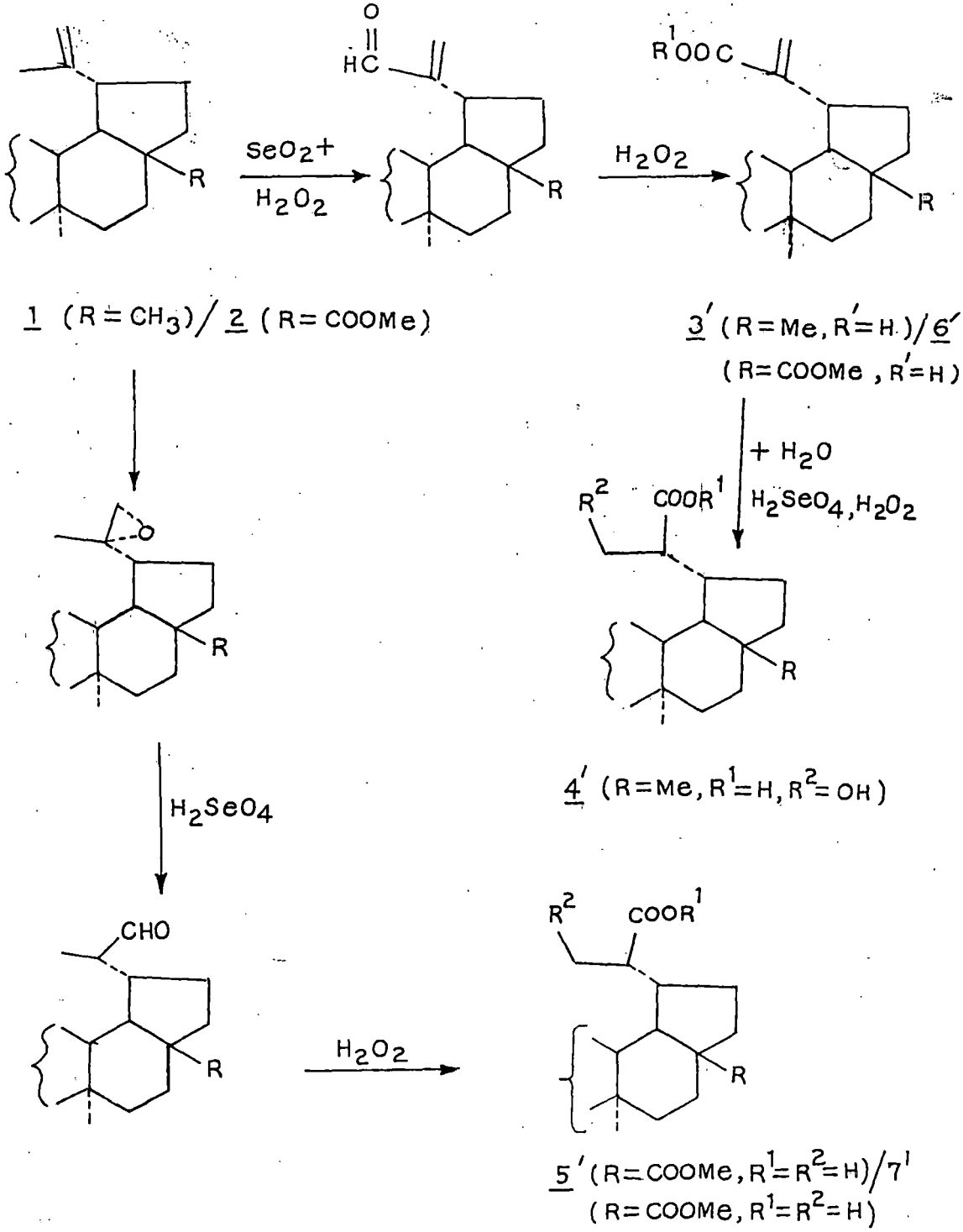
Scheme III


COOCH3
COOCH3
AcO
I
- e
COOCH3
COOCH3
AcO
a
b
m/z 558
Path a
Path b
-CH3COOH
-COOMe
m/z 439
m/z 498
-CH3
m/z 483
-CH3COOH
m/z 423
COOCH3
COOCH3
CH2
m/z 279
-COOCH3
m/z 220
-COOCH3
m/z 161
AcO
CH2
m/z 249
-HOAC
CH2
m/z 189

Discussion of the reaction mechanism:

The formation of 3'/6' from 1/2 can take place via the formation of an aldehydic group at C-30 by SeO_2 oxidation which then gets oxidised easily with H_2O_2 or perhydroxy selenic acid to C-30 carboxylic acid (3'/6'). Formation of aldehyde at the allylic position using SeO_2 in acetic acid has been reported in literature[78] which on further oxidation with chromic acid in acetic acid furnishes the acid 3'/6'. However, the formation of saturated C-29 carboxylic acid (5'/7') can take place by the formation of epoxide at C-20-29 position that opens up to form a saturated aldehyde that gives rise to saturated C-29 carboxylic compound (5'/7') (Cf. Scheme IV). Formation of similar products has been reported on treatment of 2 with chromic acid giving only one isomer 7. The formation of 4' probably takes place by the hydroxylation of 3' with selenic acid attacking the C-30 carbon in the presence of hydrogen peroxide (anti Markownikoff's rule).

Scheme IV



1 (R = CH_3) / 2 (R = COOMe)

3′ (R = Me, R′ = H) / 6′ (R = COOMe, R′ = H)

4′ (R = Me, R^1 = H, R^2 = OH)

5′ (R = COOMe, R^1 = R^2 = H) / 7′ (R = COOMe, R^1 = R^2 = H)

Table 1

Comparison of Carbon-13 Chemical shift (ppm) of compound 3-6 with lupenyl acetate[83] 1

Carbon	3	4	5	6	Lupenyl acetate[83]
1	38.35(t)	38.36(t)	38.36(t)	38.23(t)	38.3(t)
2	23.67(t)	23.67(t)	23.52 23.62 (t)	23.63(t)	23.6(t)
3	80.93(d)	80.91(d)	80.91(d)	80.85(d)	80.70(d)
4	37.72(s)	37.79(s)	37.70 37.78 (s)	37.74(s)	37.7(s)
5	55.33(d)	55.3(d)	55.32(d)	55.36(d)	53.3(d)
6	18.17(t)	18.2(t)	18.18(t)	18.12(t)	18.2(t)
7	34.20(t)	34.20(t)	34.24 34.29 (t)	34.2(t)	34.1(t)
8	40.79(s)	40.8(s)	40.84(s)	40.58(s)	40.7(s)
9	50.14(d)	49.90(d)	49.91(d)	50.22(d)	50.2(d)
10	37.04(s)	37.0(s)	37.04(s)	37.03(s)	37.0(s)
11	21.0(t)	20.8(t)	20.79 20.90 (t)	20.86(t)	20.9(t)
12	27.5(t)	23.92(t)	23.67(t)	26.9(t)	25.0(t)
13	37.78(d)	40.08(d)	40.31(d)	37.98(d)	37.9(d)
14	42.7(s)	42.98(s)	42.98(s)	42.72(s)	42.7(s)
15	27.34(t)	27.2(t)	27.23(t)	29.58(t)	27.4(t)
16	35.46(t)	35.2(t)	35.41(t)	32.00(t)	35.5(t)
17	43.13(s)	42.98(s)	42.98 43.04 (s)	56.56(s)	42.9(s)
18	49.80(d)	48.0(d)	48.67(d)	50.79(d)	48.2(d)
19	50.89(d)	46.69(d)	47.13(d)	50.79(d)	47.9(d)
20	147.08(s)	49.89(d)	43.63(d)	150.1(s)	150.5(s)
21	17.93(t)	17.62(t)	17.22(t)	32.28(t)	29.8(t)
22	39.7(t)	39.56(t)	39.69(t)	36.53(t)	39.9(t)

Contd..

Table 1 (Contd..)

23	27.94(q)	27.95(q)	27.94(q)	27.88(q)	27.9(q)
24	16.5(q)	16.5(q)	16.498(q)	16.44(q)	16.5(q)
25	16.14(q)	16.12(q)	16.11(q)	16.09(q)	16.1(q)
26	15.94(q)	15.96(q)	16.11 15.99 (q)	15.9(q)	15.9(q)
27	14.4(q)	14.4(q)	14.29 14.16 (q)	14.6(q)	14.4(q)
28	17.90(q)	18.18(q)	17.93 17.71 (q)	176.56(s)	18.0(q)
29	122.73(t)	61.43(t)	9.81(q)	123.5(t)	109.2(t)
30	168.24(s)	174.25(s)	176.88 176.34 (s)	167.74(s)	19.2(q)
$-COO\underline{C}H_3$	51.79(q)	51.77(q)	51.41 50.92 (q)	51.42(q)	
$-O-\overset{\overset{O}{\|}}{C}-$	171.04(s)	170.98 171.00 (s)	170.98 170.95 (s)	170.9(s)	
$-O-\overset{\overset{O}{\|}}{C}-\underline{C}H_3$	21.33(q)	21.04 21.32 (q)	21.30(q)	21.26(q)	