

Electronic Supplementary Information

Cyclization cascade of the C₃₃-bisorheptaprenoid catalyzed by recombinant squalene cyclase

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Contents

Synthesis of (6*E*, 10*E*, 14*E*, 17*E*, 21*E*)-5,9,13,18, 22,26-hexamethyltricosane-1,5,9,13, 17,21-hexanene **14** S2-S3

EIMS spectra and the analyses of NMR data of products **20-35**, and ¹H and ¹³C NMR spectra of products **20-35** together with 2D NMR spectra of product **29** S4-S56

Synthesis of (6E, 10E, 14E, 17E, 21E)-5,9,13,18, 22,26-hexamethyltricoso-1,5,9,13, 17,21-hexanene 14

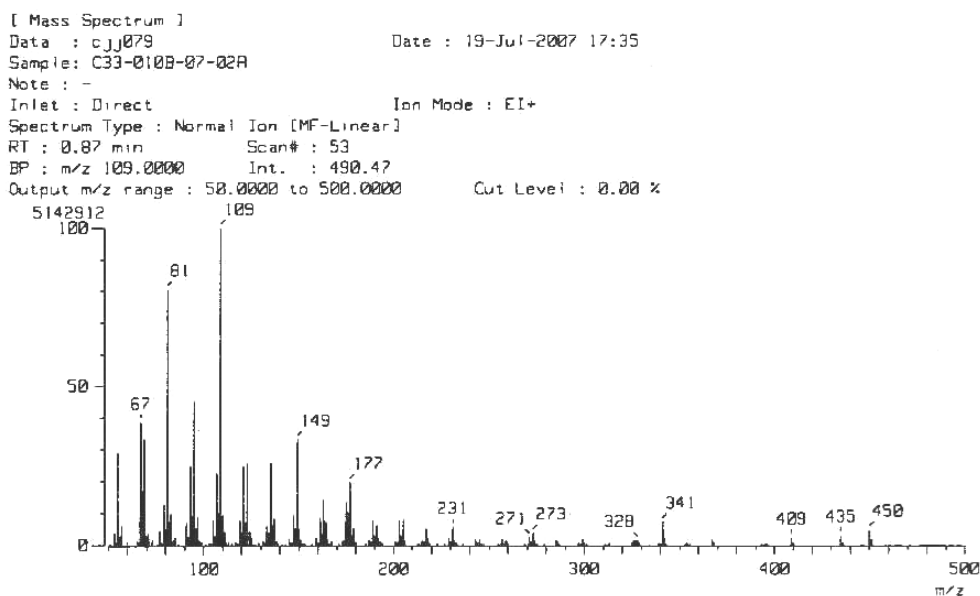
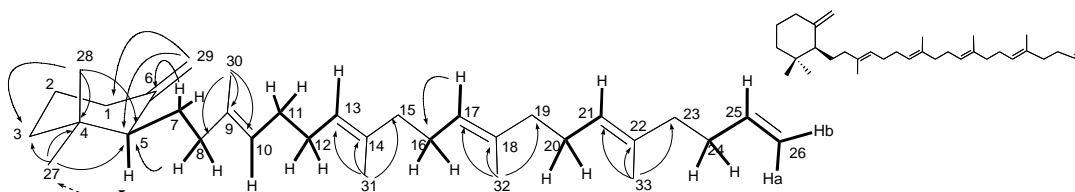
Allylic alcohol **16** was prepared according to the published method (F. W. Sum and L. Weiler, *Tetrahedron*, 1981, **37**, 303-317). Squalene **1** (72.61g, 177mmol) and salicylic acid (2.45g, 17.7mmol) were suspended in 360ml of CH₂Cl₂, and cooled at 0°C. To the suspension was added a mixture of SeO₂ (3.93g, 35.4mmol) and 45.6 g of *n*-BuOOH (70 wt% in H₂O: *n*-BuOOH used was 31.9g, 0.35mol) in a dropwise manner under atmosphere of N₂ gas, and stirred for 1.5 h, followed by addition of aq. Na₂SO₃ to quench the reaction. The hexane-extract from the reaction mixture was dried over anhyd. Na₂SO₄ and then purified by a SiO₂ column chromatography (hexane:EtOAc=100:5~100:10), yielding the desired **16** (8.24g, 10.9% yield). NMR data of **16** in C₆D₆: ¹H (400 MHz), 5.50 (1H, t, *J*=6.8 Hz), ~5.41 (5H, m), 3.93 (2H, s), 2.37~2.16 (20H, m), 1.798 (3H, s), 1.731 (6H, s), 1.722 (3H, s), 1.713(3H, s), 1.689 (3H, s), 1.685 (3H,s); δ_C (100 MHz), 135.4 (s), 135.2 (s), 135.1 (s), 134.9 (s), 134.7 (s), 131.1 (s), 125.3 (d), 124.95 (d), 124.93 (d), 124.9 (d), 124.8 (d), 124.79 (d), 68.73 (t), 40.21 (t, 3xC), 39.86 (t), 28.74 (t, 2xC), 27.22 (t), 27.09 (t, 2xC), 26.66 (t), 25.84 (q), 17.79 (q), 16.16 (q, 2xC), 16.09 (q, 2xC), 13.64 (q).

To the solution of compound **16** (4.13g, 9.7mmol) in dry THF (205ml) at 0°C was added in a dropwise fashion the solution of PBr₃ (3.94g, 14.55mmol) in 1.5 ml of THF, and stirred for 30 min. To the reaction mixture was added saturated brine, followed by extraction with hexane, which was dried over Na₂SO₄, giving **17** (4.5g), and used without further purification. To a mixture of PheSO₂Na·H₂O (1.8g, 11mmol) and DMF (90ml), was added compound **17** ((3.6g, 7mmol)) in a small portion and then the reaction continued for 1.5-2h. The reaction mixture was poured into ice-cooled saturated brine, and extracted with hexane. The hexane-extract was dried over anhyd. Na₂SO₄, and then subjected to SiO₂ column chromatography eluting with hexane:EtOAc (10:1), affording a pure **18** (1.20g, 29.4% yield). NMR data of **18** in CDCl₃: δ_H (400 MHz), 7.83(2H, ddd, *J*=7.2, 1.6, 1.6 Hz), 7.60(1H, ddd, *J*=7.2, 7.2, 1.6 Hz), 7.52(2H, ddd, *J*=7.2,7.2,1.6 Hz), 5.09(4H, m), 5.01(2H, m), 3.70(2H, s), 2.09-1.94 (20H, m), 1.75(3H, s), 1.67(3H, s), 1.59(12H, s, 4×CH₃), 1.52(3H, s); δ_C (100 MHz), 138.3 (s), 135.9 (d), 135.0 (s), 134.9 (s), 134.8 (s), 133.9 (s), 133.4 (d), 131.1 (s), 128.8 (d, 2xC), 128.4(d, 2xC), 124.7 (d), 124.3 (d), 124.27 (d), 124.2 (d, 2xC), 123.1 (s), 66.16 (t), 39.66 (t), 39.64 (t), 39.58 (t), 38.51(t),

28.19(t, 2xC), 26.86(t), 26.68 (t), 26.56 (t), 26.53 (t), 25.61 (q), 17.59(q), 16.57(q), 15.96(q, 2xC), 15.92(q), 15.80(q).

Compound **18** (1.2 g, 2mmol) was dissolved in 60ml of THF:[(CH₃)₂N]₃PO (4:1) at -40°C under N₂ gas. Then *n*-BuLi (1.58M, 4.11ml, 6mmol) was added, giving a brown color, to which the THF solution (1 ml) of allyl bromide (0.4g, 3mmol) was slowly added at -78°C and reacted for 20 min. The reaction mixture was poured into ice-cooled saturated brine, then extracted with hexane, and dried over Na₂SO₄. A SiO₂ column chromatography eluting with a mixture of hexane and EtOAc (3~5% EtOAc in hexane) gave pure compound **19** (869.2mg, yield 67.5%). NMR data of **19** in CDCl₃: ¹H (400 MHz), 7.80(2H, d, *J* = 7.6Hz), 7.61(1H, t, *J* = 7.6Hz), 7.51(2H, t, *J* = 7.6Hz), 5.58(1H, m), 5.12(5H, m), 5.02(3H, m), 3.53(1H, dd, *J* = 11.6, 3.6Hz), 2.86(1H, m), 2.66(1H, m), 2.08-1.90 (20H, m), 1.67 (3H, s), 1.66 (3H, s), 1.59(12H, s, 4×CH₃), 1.51(3H, s); ¹³C (100 MHz), 137.7 (s), 136.1 (d), 135.0 (s), 134.9 (s), 134.8 (s), 133.9 (s), 133.3 (d), 133.2 (d), 131.1 (s), 128.8(d, 2xC), 128.6(d, 2xC), 126.0 (s), 124.6 (d), 124.3 (d), 124.28 (d), 124.2 (d, 2xC), 117.7 (t), 73.65 (d), 39.68 (t), 39.66(t), 39.61 (t), 38.44 (t), 29.45 (t), 28.20(t, 2xC), 26.69 (t), 26.56(t, 3xC), 25.63 (q), 17.61 (q), 15.98(q, 2xC), 15.93(q), 15.77 (q), 13.49 (q).

Compound **19** (869.2mg, 1.47nmol) and C₂₇H₂₆Cl₂P₂Pd (49mg, 0.083nmol) was dissolved in dry Et₂O (160 ml) at 0°C under N₂ atmosphere. To the solution, super hydride reagent (1.0 M, 20 ml) was added slowly until the deep brown color appears, and reacted for 10 min. The reaction mixture was poured in a dropwise manner into an ice-cooled saturated brine, and then extracted with hexane. The hexane extract was dried over Na₂SO₄. The desired C₃₃ analog **14** was obtained after purifying with SiO₂ column chromatography eluting with hexane (585.6mg, 88.30%). NMR data of **14** in C₆D₆: ¹H (400 MHz), 5.79 (1H, m), 5.12 (6H, m), 4.99 (1H, bd, *J*=17.2 Hz), 4.93 (1H, bd, *J*=10.0 Hz), 2.33-1.97 (24H, m), 1.68 (3H, s), 1.60 (18H, s, 6×CH₃); ¹³C (100 MHz), 138.7 (d), 135.1 (s, 2xC), 134.9 (s), 134.8 (s), 134.3 (s), 131.2 (s), 124.6 (d), 124.4 (d), 124.3 (d), 124.28 (d, 2xC), 124.26 (d), 114.2 (t), 39.75(, t, 2xC), 39.72 (t), 39.69 (t), 39.04 (t), 32.37 (t), 28.27 (t, 2xC), 26.75 (t), 26.6t (t, 2xC), 26.59 (t), 25.69 (q), 17.67 (q), 16.04 (q, 2xC), 15.99 (q, 2xC), 15.95 (q). EIMS; *m/z* (%) 67(34), 69 (100), 81 (72), 95(25), 109(40), 121(15), 137 (12), 149 (9), 163(2), 177(2), 203(1), 217(1), 271(1), 299(1), 342(1), 382(1), 408(1), 450(M⁺, 1).

Product 20**Product 20**

$[\alpha]_D^{25} +8.99$ (EtOH, $c=0.2$)

HRMS for calcd. C₃₃H₅₄ (M⁺)450.4226
 Found 450.4218

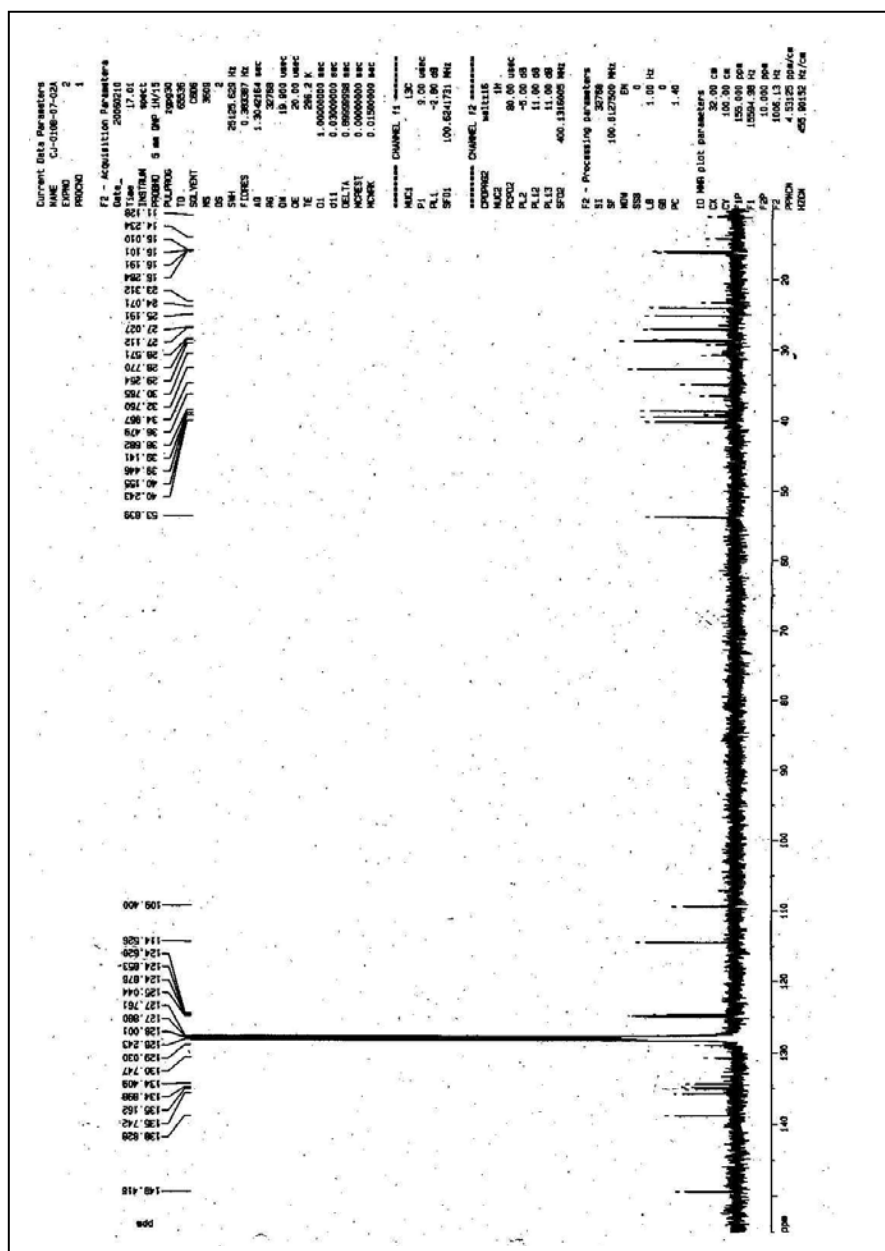
400MHz in C₆D₆
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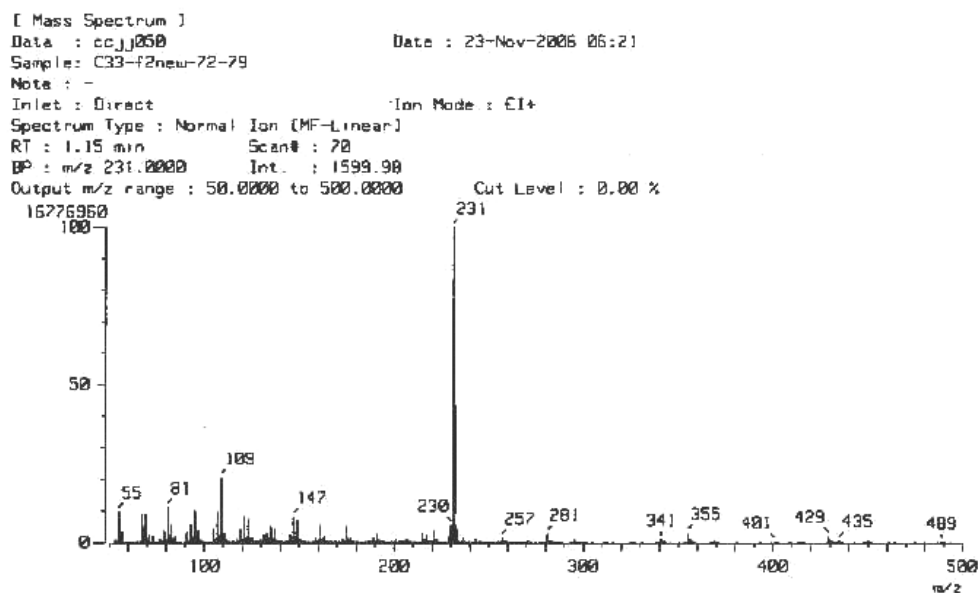
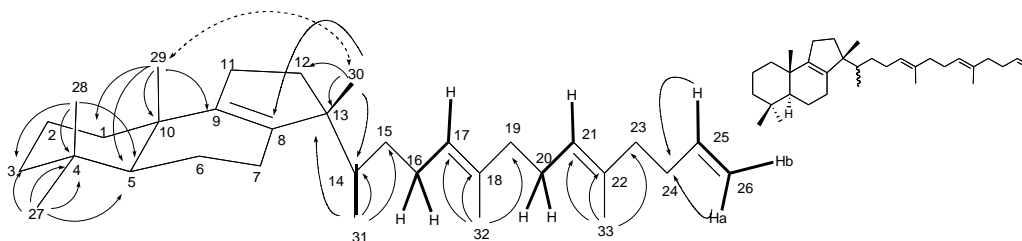
NOE: HMBC:
 COSY:

NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C
1	2.10(m);2.22(m)	32.76	9	—	135.7	17	5.42(1H, t, J=6.8)	124.6	25	5.93(1H, m)	138.8
2	1.61(2H,m)	24.07	10	5.47(m)	124.6	18	—	134.9	26	5.16(1H, bdd, J=16.4, 1.6Hz) 5.12(1H, bd, J=10 Hz)	114.5
3	1.52(1H, m); 1.27(1H, m)	36.48	11	2.27(2H,m)	28.77	19	2.22(2H, m)	40.16 ^a	27	1.075(3H, s)	28.57
4	—	34.96	12	2.27(2H,m)	28.77	20	2.30(2H,m)	27.03 ^b	28	0.972(3H, s)	26.51
5	1.86(1H, J=11.4, 3.0)	53.84	13	5.47(m)	124.9	21	5.36(1H, t, J=6.8)	125.0	29	5.00(1H,bs); 4.82(1H, bs)	109.4
6	—	149.4	14	—	135.2	22	—	134.4	30	1.770(3H, s)	16.28
7	1.64(m);1.77(m)	25.19	15	2.22(2H, m)	40.24 ^a	23	2.17(2H,m)	39.45	31	1.746(3H, s)	16.19 ^c
8	2.05(m);2.23(m)	38.68	16	2.30(2H,m)	27.11 ^b	24	2.22(2H,m)	32.76	32	1.731(3H, s)	16.10 ^c
									33	1.671(3H, s)	16.01

The carbon signals of a-c are exchangeable between the same characters.

¹³C-NMR



Product 21**Product 21**

$[\alpha]_D^{25}$ -28.08 (c=0.073, EtOH)

HRMS for calcd. $C_{33}H_{54}$ (M^+) 450.4226
 Found 450.4227

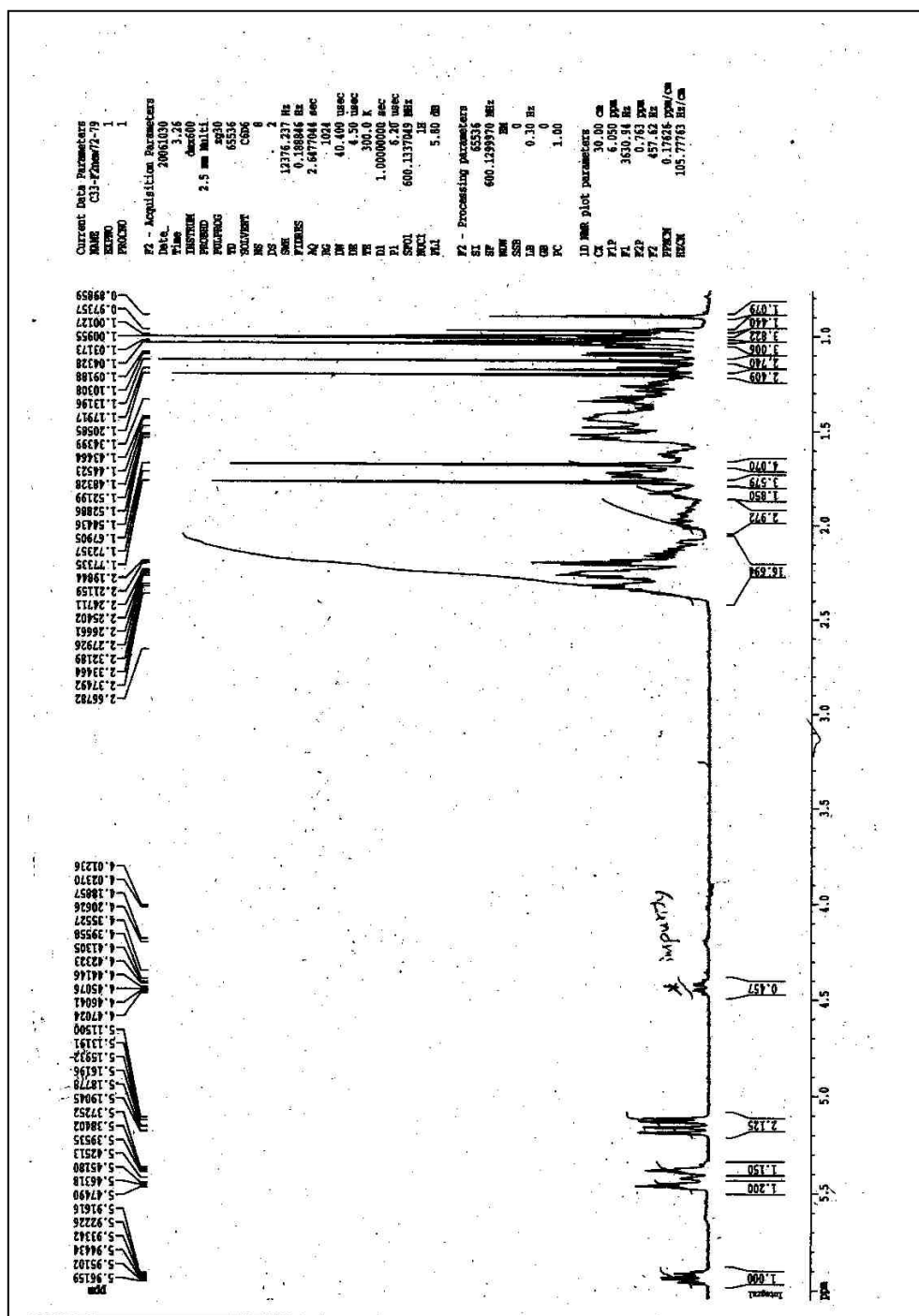
600 MHz in C_6D_6
 the solvent peak 1H : 7.28 ppm; ^{13}C :128.0 ppm

NOE: \dashrightarrow
 COSY: \longrightarrow HMBC: \dashrightarrow

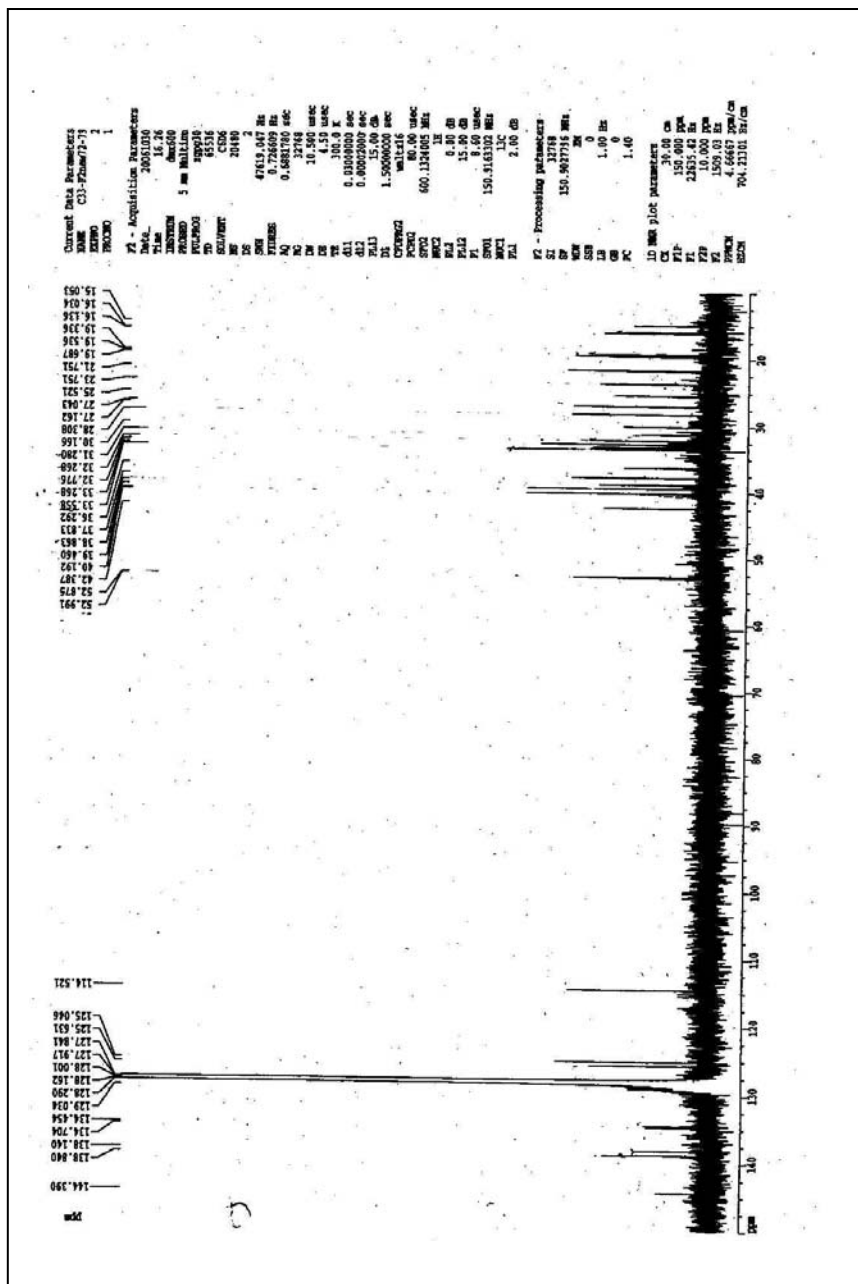
NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	1.26 (1H, m); 1.72 (1H, m)	37.83	9	—	144.4	17	5.46 (1H, t, J=6.6)	125.6	25	5.94 (1H, m)	138.9
2	1.49 (2H, m)	19.34 ^a	10	—	36.29	18	—	134.7	26	a:5.17 (1H, bdd, J=17.1, 1.6); b:5.12 (1H, bdd, J=10.1)	114.5
3	1.29 (1H, m); 1.54 (1H, m)	42.39	11	2.27 (1H, m); 2.36 (1H, m)	28.31	19	2.25 (2H, m)	40.19	27	1.043 (3H, s)	33.56
4	—	33.27	12	1.23 (1H, m); 1.85 (1H, m)	31.28	20	2.33 (2H, m)	27.04 ^b	28	1.010 (3H, s)	21.75
5	1.33 (1H, bd, J=10.7)	52.87	13	—	52.99	21	5.38 (1H, t, 6.6)	125.1	29	1.132 (3H, s)	19.69
6	1.49 (2H, m)	19.54 ^a	14	1.63 (m)	38.86	22	—	134.5	30	1.206 (3H, s)	25.52
7	1.97 (1H, m); 2.12 (1H, m)	23.75	15	1.23 (1H, m); 1.82 (1H, m)	32.27	23	2.20 (2H, m)	39.46	31	1.00 (3H, d, J=6.0)	15.05
8	—	138.1	16	2.15 (m, 2.31 (m))	27.16 ^b	24	2.20 (1H, m); 2.25 (1H, m)	32.78	32	1.773 (3H, s)	16.14
									33	1.679 (3H, s)	16.03

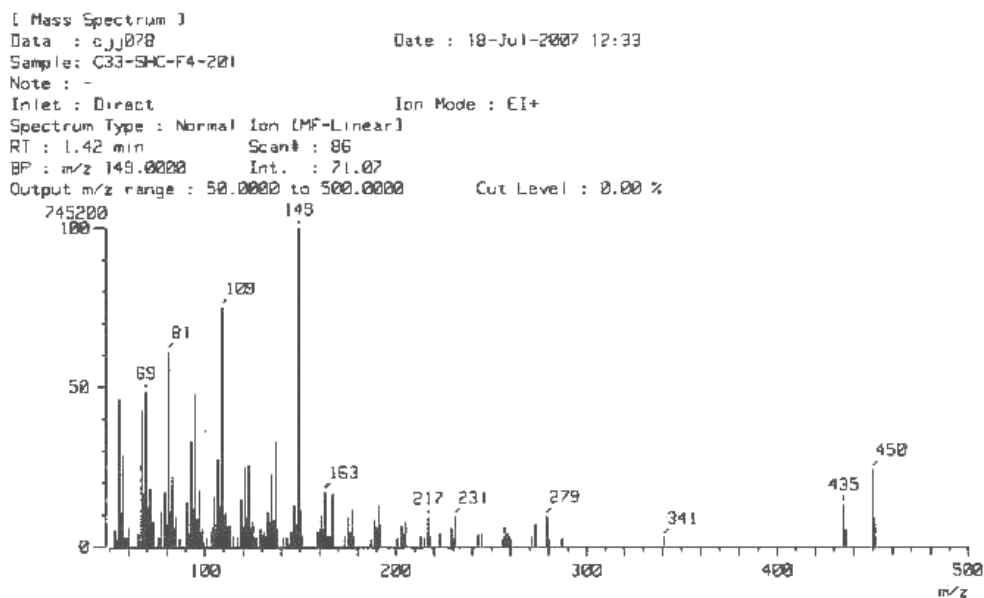
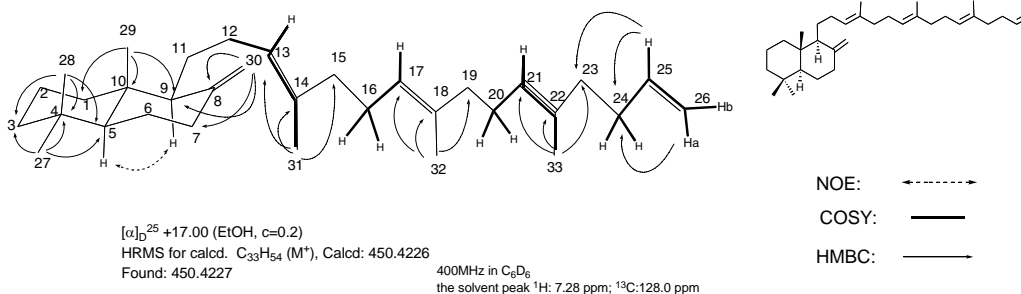
The carbon signals of **a** and **b** may be exchangeable between the same letters.

¹H-NMR



¹³C-NMR

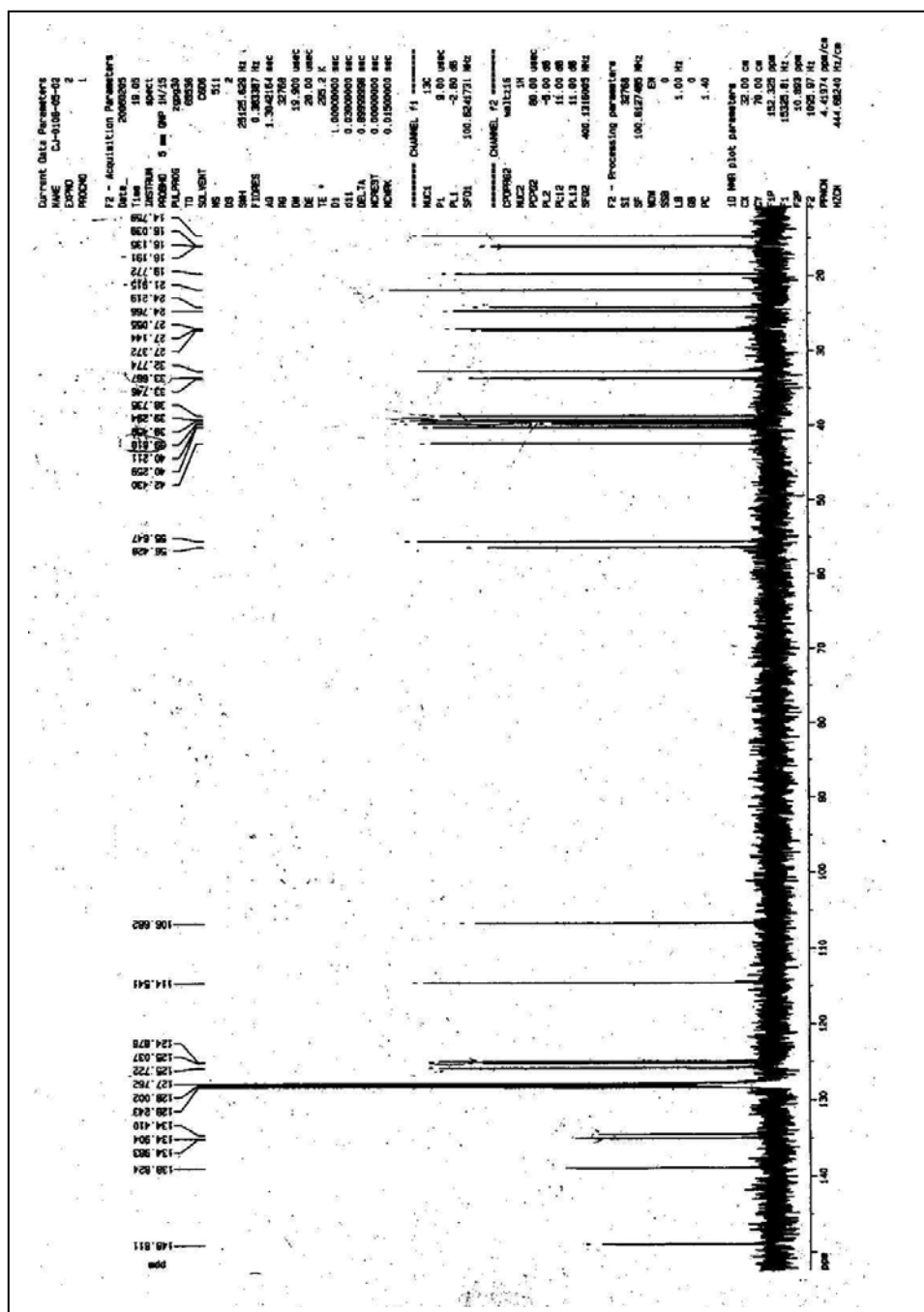


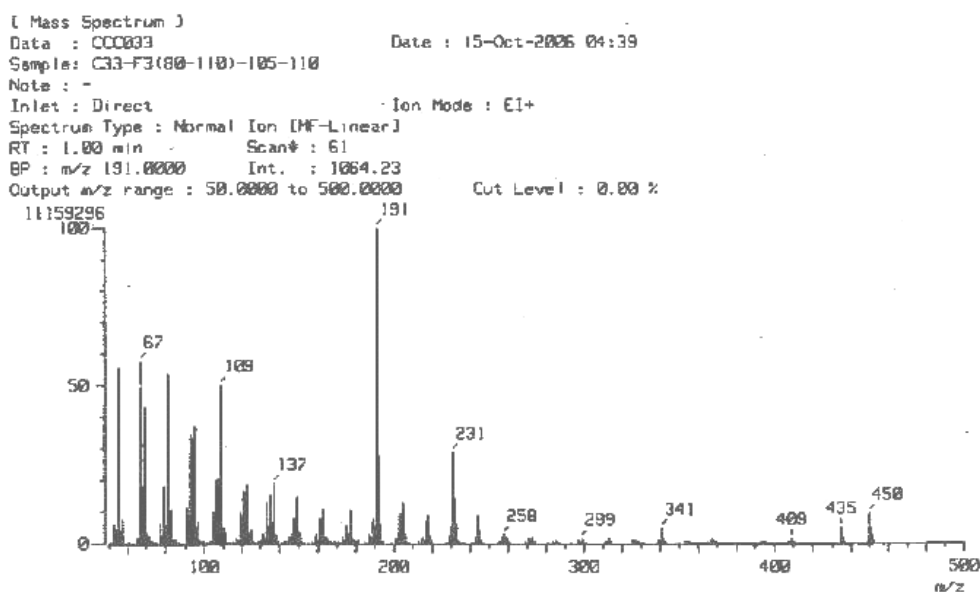
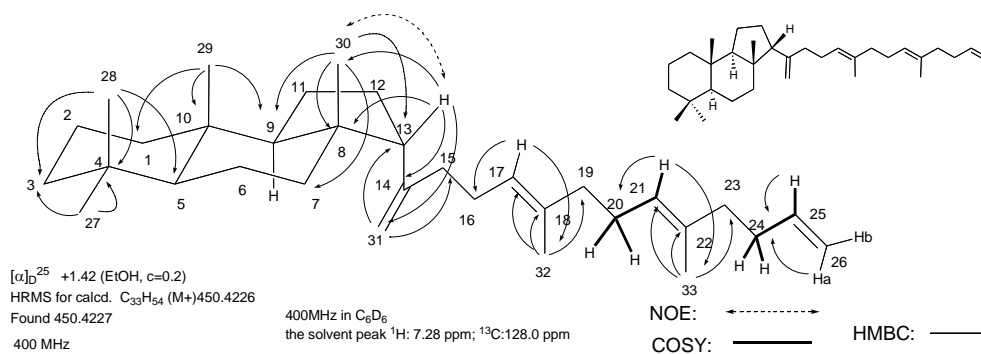
Product 22**Product 22**

NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C
1	1.05 (1H, m); 1.79 (1H, m)	39.29	9	1.76 (1H, m)	56.43	17	5.40 (1H, m)	124.9	25	5.90 (1H, m)	138.8
2	1.53 (1H, m); 1.60 (1H, m)	19.77	10	—	39.82	18	—	134.9	26	a:5.16(1H, dd, J=15.6, 1.6) b:5.11(1H, bd, J=10.0)	114.5
3	1.27 (1H, m); 1.47 (1H, m)	42.43	11	1.63 (1H, m); 1.76 (1H, m)	24.22	19	2.21 (2H, m)	40.21	27	0.939 (3H, s)	33.75
4	—	33.67	12	2.08 (1H, m); 2.40(1H, m)	27.37	20	2.31 (2H, m)	27.06	28	0.892(3H, s)	21.92
5	1.10 (1H, dd, J=12.8, 2.4)	55.65	13	5.42 (1H, m)	125.7	21	5.33 (1H, t, J= 6.8)	125.0	29	0.831(3H, s)	14.76
6	1.39 (1H, m); 1.72 (1H, m)	24.77	14	—	134.9	22	—	134.4	30	5.06 (1H, s); 4.80(1H, s)	106.7
7	2.18 (1H, m); 2.49(1H, m)	38.74	15	2.21 (2H, m)	40.26	23	2.17 (2H, m)	39.46	31	1.74 (3H, s)	16.04
8	—	148.8	16	2.31 (2H m)	27.14	24	2.23 (2H, m)	32.77	32	1.71 (3H, s)	16.14
									33	1.64 (3H, s)	16.19

Positions of 15 and 19 are exchangeable; positions of 12, 16 and 20 are exchangeable; positions of 13, 21 and 17 are exchangeable; positions of 14, 18, 22 are exchangeable

¹³C-NMR

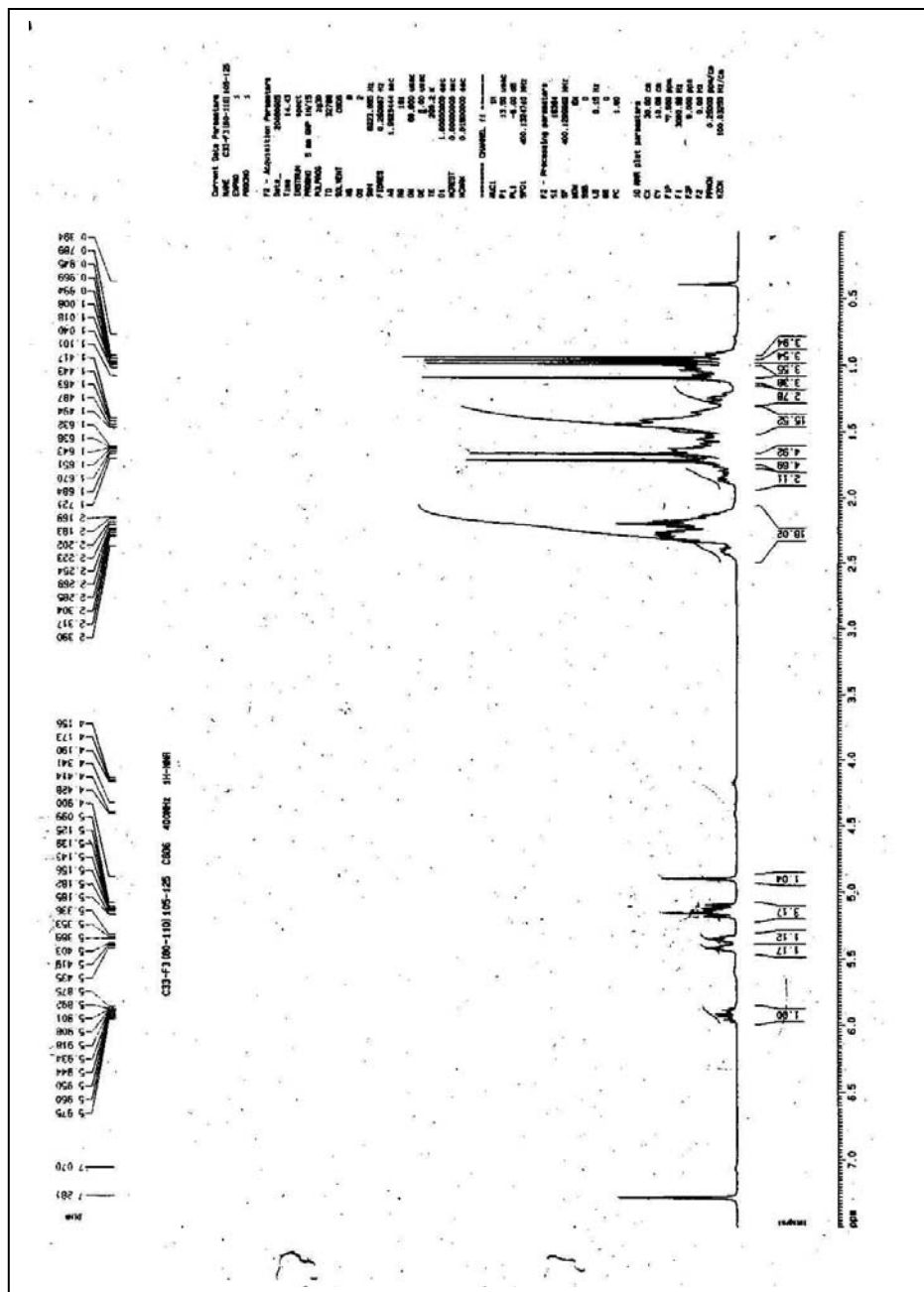


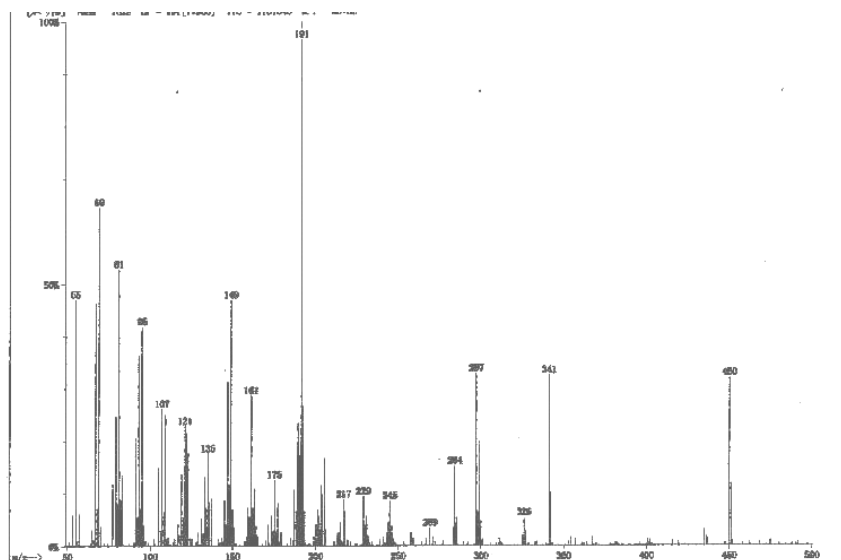
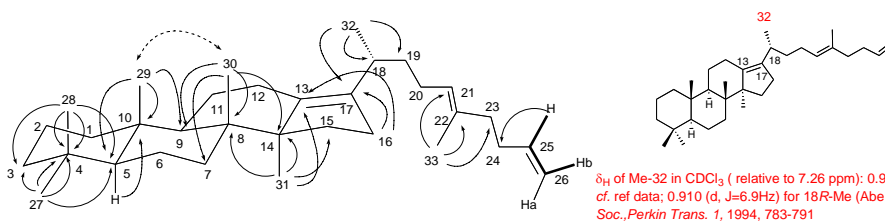
Product 23**Product 23**

NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C
1	1.08(1H, m); 1.59(1H, m)	40.74	9	1.65(1H, m)	55.84	17	5.42(1H, t, J=6.4Hz)	125.0	25	5.93(1H, m)	138.8
2	1.49(1H, m); 1.76(1H, m) ^a	18.83 ^b	10	—	37.05	18	—	135.1	26	a:5.16(1H, bd., 16.8); b:5.11(bd.,10.4Hz)	114.5
3	1.25(1H, m); 1.48(1H, m)	42.68	11	1.56(1H, m); 1.65(1H, m)	21.06	19	2.20(2H, m)	40.13	27	0.945(3H, s)	33.62
4	—	33.14	12	1.84(1H, m); 2.15(1H, m)	28.17	20	2.27(2H, m)	27.00	28	0.969(3H, s)	21.63
5	0.92(1H, m)	57.25	13	2.31(1H, m)	56.98	21	5.35(1H, t, 6.4Hz)	124.8	29	0.994(3H, s)	15.80
6	1.49(1H, m); 1.68(1H, m) ^a	19.63 ^b	14	—	154.6	22	—	134.5	30	1.101(3H, s)	25.01
7	1.44(1H, m); 1.70(1H, m)	37.10	15	2.20(1H, m); 2.34(1H, m)	39.81	23	2.18(2H, m)	39.45	31	4.90(1H, s); 5.16(1H, s)	109.3
8	—	45.86	16	2.30(1H, m); 2.40(1H, m)	27.38	24	2.27(2H, m)	32.77	32	1.721(3H, s)	16.12
									33	1.670(3H, s)	16.04

The assignments of proton and carbon signals at 2- and 6-position may be exchangeable.

¹H-NMR



Product 24**Product 24 (Oil)**

$[\alpha]_D^{25}$ -35.67 (EtOH, $c=0.1$)
 HRMS for calcd. $\text{C}_{33}\text{H}_{54}$ (M^+) 450.4226
 Found 450.4219

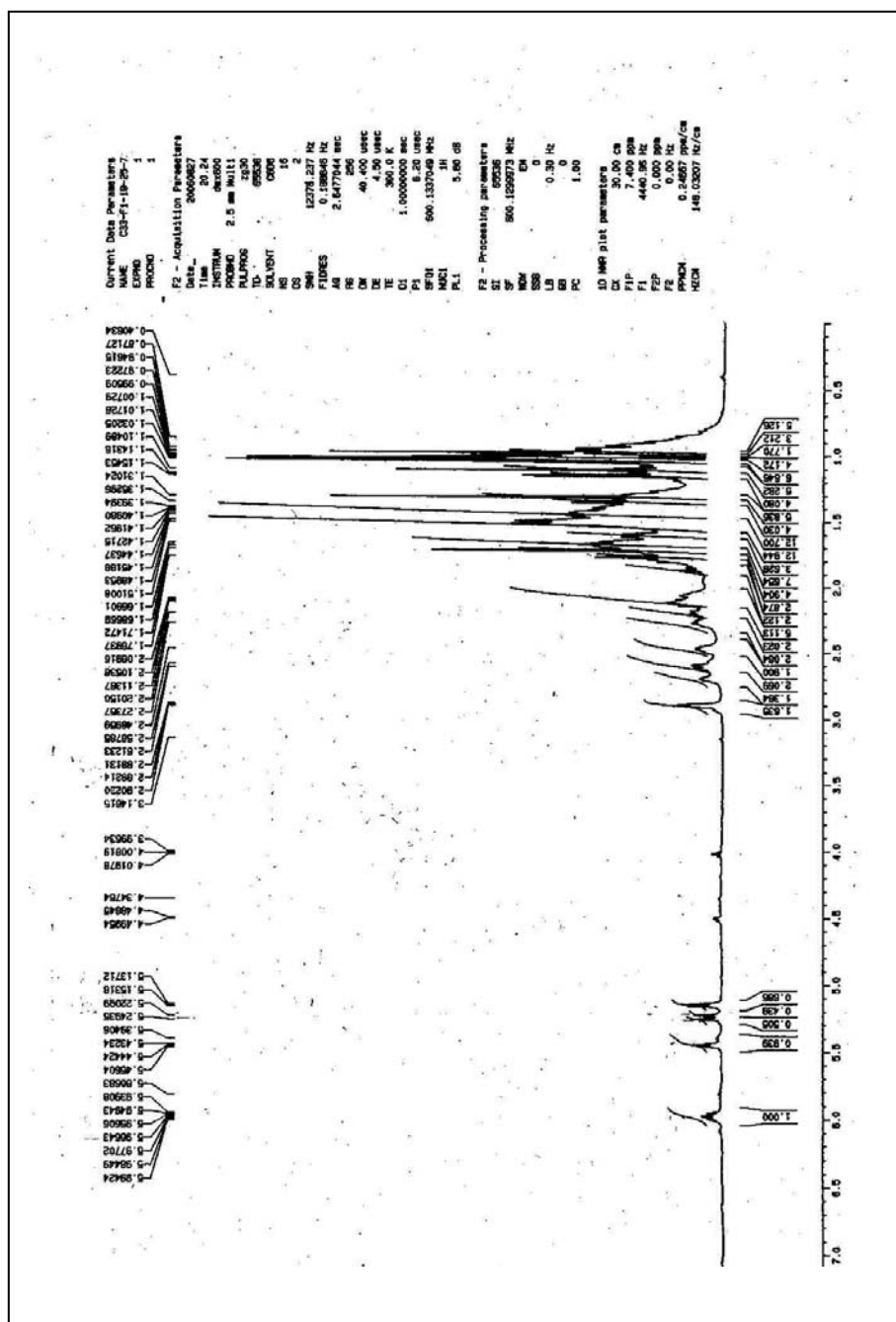
600 MHz in C_6D_6
 the solvent peak ^1H : 7.28 ppm; ^{13}C : 128.0 ppm

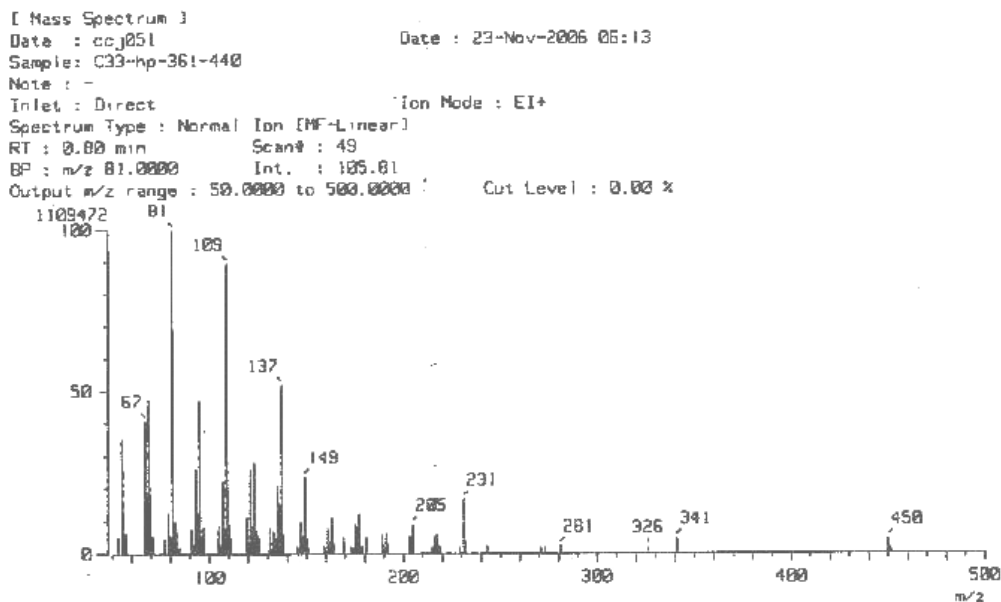
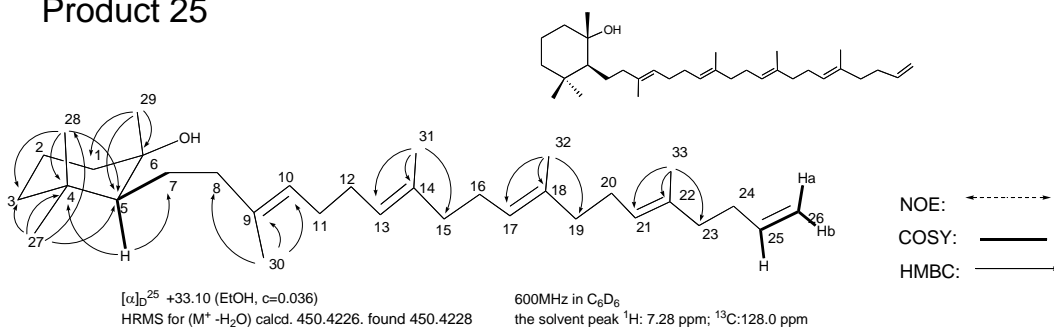
NOE: \dashleftarrow \dashrightarrow
 COSY: --- HMBC: ---

NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C
1	0.95(1H, m); 1.79(1H, m)	40.96	9	1.60(1H, bd, 12.2Hz)	52.25	17	—	135.1	25	5.97(1H, m)	137.6
2	1.45(1H, m); 1.70(1H, m)	18.99	10	—	38.10	18	2.67(1H, m)	32.27	26	a:5.23(1H, bd, 17.1Hz) b:5.14(1H, bd, 9.7Hz)	114.3
3	1.27(1H, m); 1.50(1H, m)	42.41	11	1.43(m); 1.70(m)	22.19	19	1.50(1H, m); 1.82(1H, m)	35.43	27	1.03(3H, s)	33.68
4	—	33.53	12	2.05(1H, m); 2.62(1H, m)	23.41	20	2.18(m); 2.33(m)	32.63	28	0.972(3H, s)	21.91
5	0.94(1H, m)	57.38	13	—	139.6	21	5.44(1H, bt, 7.1)	125.8	29	1.017(3H, s)	16.67
6	1.46(1H, m); 1.67(1H, m)	19.08	14	—	57.00	22	—	134.2	30	1.105(3H, s)	17.67
7	1.50(1H, m); 1.67(1H, m)	35.85	15	2.11 (2H, m)	31.09	23	2.23(2H, m)	39.52	31	1.310(3H, s)	23.13
8	—	41.67	16	2.30(1H, m); 2.48(1H, m)	29.49	24	2.27(2H, m)	32.77	32	1.144(3H, d, 6.7Hz)	20.21
									33	1.714(3H, s)	16.03

The proton and carbon signals at C2 and C6 may be exchangeable.

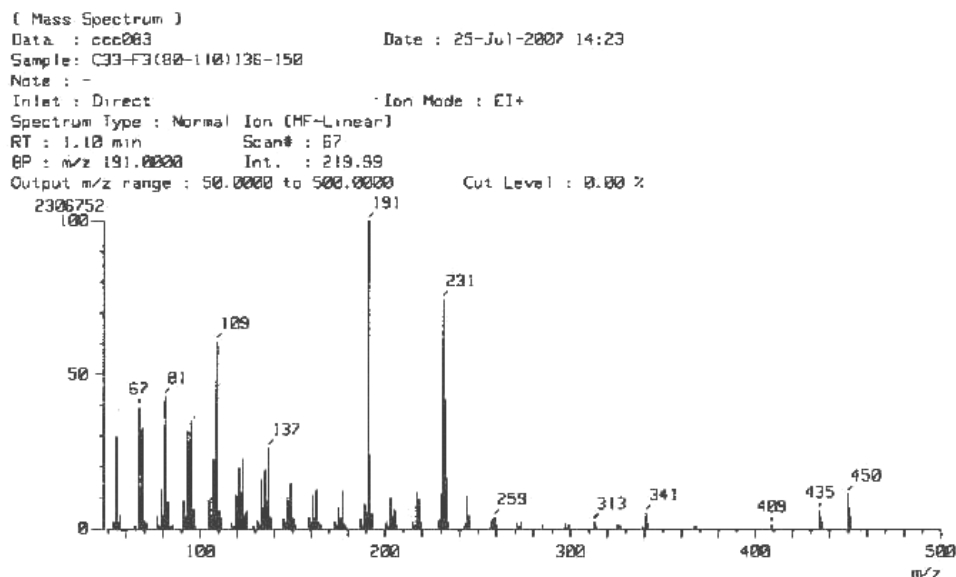
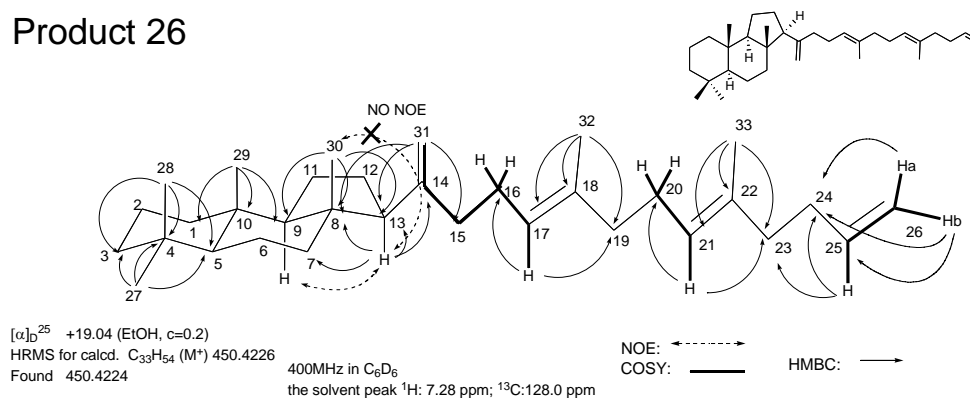
¹H-NMR



Product 25**Product 25**

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	1.23(1H, ddd, 12.5, 12.5, 3.8Hz); 1.72(1H, m)	43.97	9	—	136.6	17	5.42(1H, t, 6.4)	124.9	25	5.93(1H, m)	138.8
2	1.41(1H, m); 1.49(1H, m)	20.85	10	5.54(1H, m)	124.6	18	—	134.9	26	Ha:5.16(1H, bd, 16.6) Hb:5.12(1H, bd, $J=9.3$)	114.5
3	1.22(1H, m); 1.35(1H, m)	41.77	11	2.30(2H, m)	28.79	19	2.22(2H, m)	40.25	27	1.056(3H, s)	32.95
4	—	35.60	12	2.30(2H, m)	28.76	20	2.32(2H, m)	27.04	28	0.872(3H, s)	21.58
5	1.15(1H, m)	56.86	13	5.47(1H, m)	124.9	21	5.37(1H, t, $J=6.6$)	125.1	29	1.203(3H, s)	23.50
6	—	73.51	14	—	135.2	22	—	134.4	30	1.843(3H, s)	16.35
7	1.57 (m); 1.86(m)	25.35	15	2.22(2H, m)	40.16	23	2.23(2H, m)	39.45	31	1.764(3H, s)	16.21
8	2.33(m); 2.47(m)	43.41	16	2.34(2H, m)	27.14	24	2.26(2H, m)	32.76	32	1.734(3H, s)	16.11
									33	1.673(3H, s)	16.02

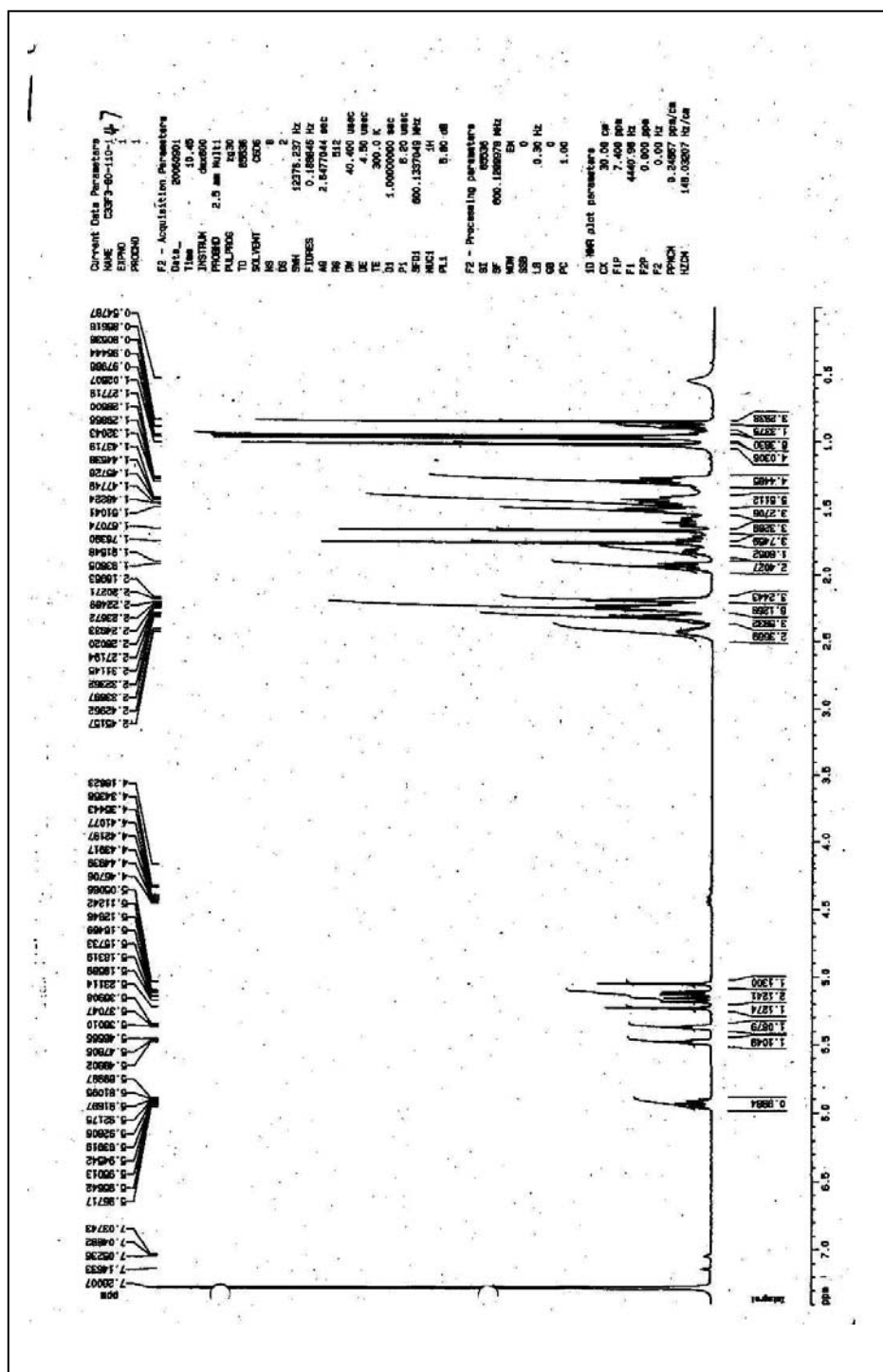
The following positions may be exchangeable between 11 and 12, between 14 and 18, between 15 and 19, between 16 and 20, and between 31 and 32

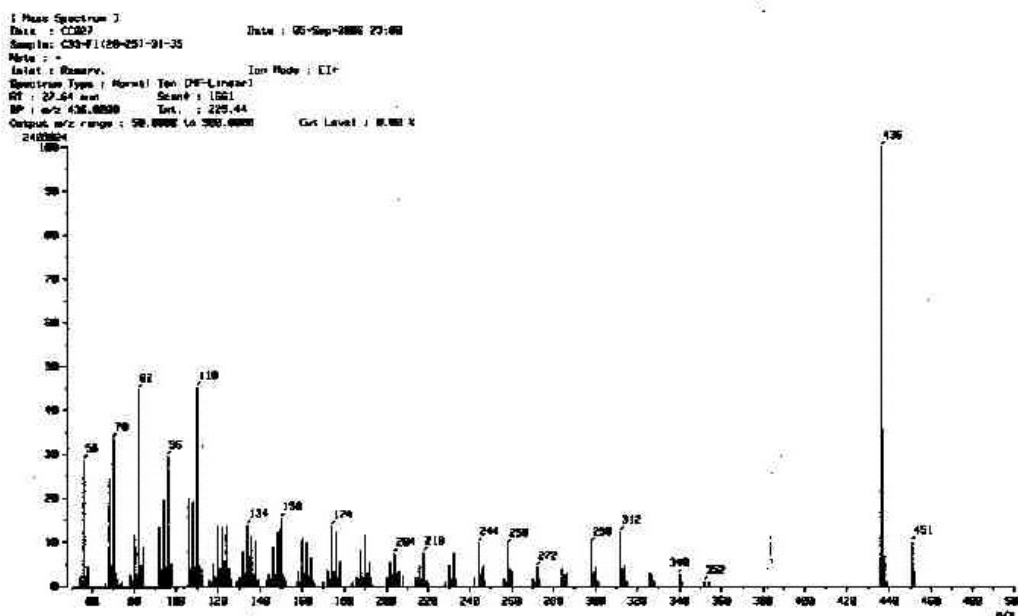
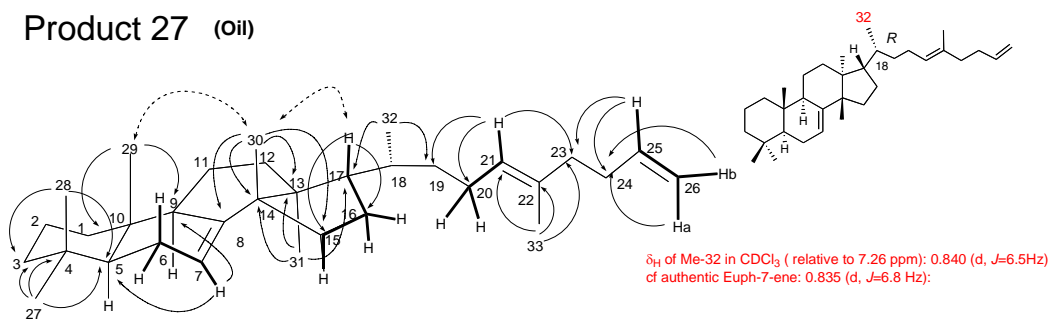
Product 26**Product 26**

NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C
1	0.94(1H, m); 1.52(1H, m)	40.15 ^a	9	1.28(1H, m)	63.36	17	5.46(1H, t, 6.4Hz)	124.9	25	5.93(1H, m)	138.8
2	1.49(1H, m); 1.74(1H, m)	18.77	10	—	37.39	18	—	135.0	26	Ha:5.18(1H, bdd, 16.8, 1.6Hz) Hb:5.12(bd, 10.0Hz)	114.5
3	1.28(1H, m); 1.52(1H, m)	42.90	11	1.45(1H, m); 1.60(1H, m)	19.81	19	2.22 (2H, m)	40.26 ^a	27	1.020(3H, s)	33.74
4	—	33.17	12	1.82(1H, m); 1.95(1H, m)	25.80	20	2.30(2H, m)	26.99	28	0.975(3H, s)	21.50
5	0.88(1H, m)	57.57	13	2.20(1H, m)	57.57	21	5.36(1H, t, 6.0Hz)	124.9	29	0.962 (3H, s)	15.62
6	1.28(1H, m); 1.40(1H, m)	19.81	14	—	149.2	22	—	134.5	30	0.847(3H, s)	15.38
7	1.27(1H, m); 1.92(1H, m)	41.47	15	2.25(1H, m); 2.32(1H, m)	38.12	23	2.19(2H, m)	39.45	31	5.03(1H, s); 5.21(1H, s)	110.6
8	—	43.99	16	2.41(2H, m)	27.53	24	2.26(2H, m)	32.78	32	1.756(3H, s)	16.17
									33	1.667(3H, s)	16.05

The carbon signals of **a** may be exchangeable.

¹H-NMR



Product 27**Product 27 (oil)**

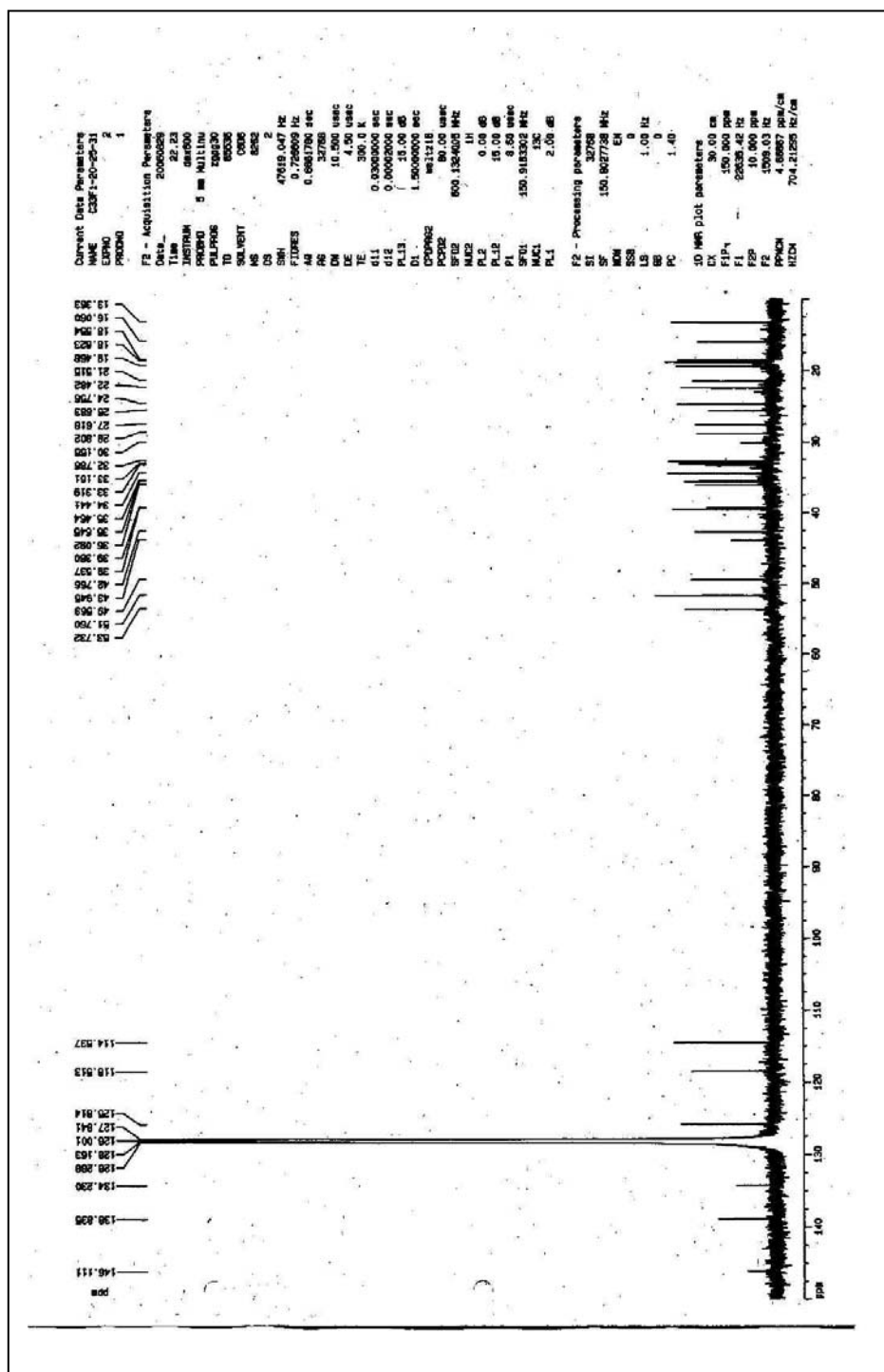
$[\alpha]_{\text{D}}^{25}$ -19.80 (EtOH, $c=0.1$)
 HRMS for calcd. $\text{C}_{33}\text{H}_{54}$ (M^+) 450.4226
 Found 450.4231

600 MHz in C_6D_6
 the solvent peak ^1H : 7.28 ppm; ^{13}C :128.0 ppm

NOE: \dashrightarrow
 COSY: — HMBC: —

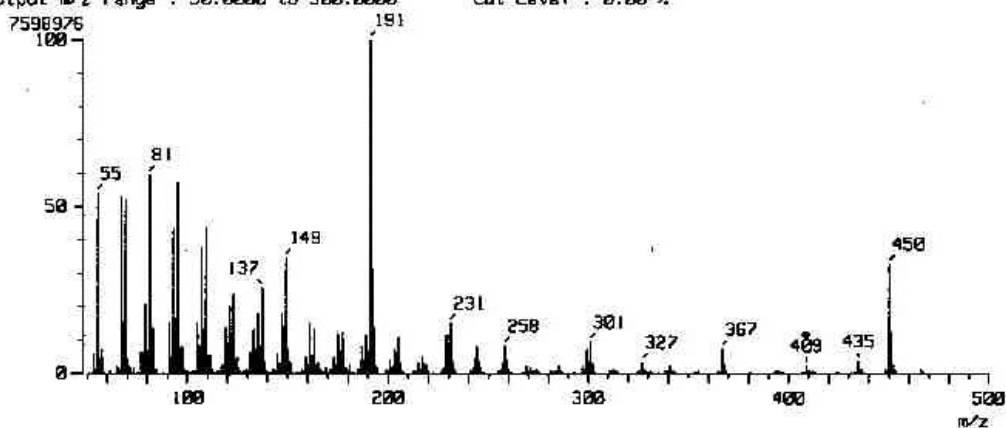
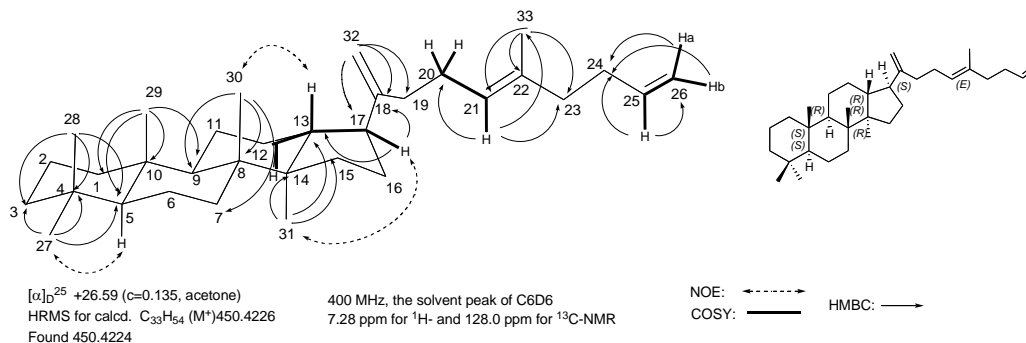
NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C
1	1.06(m);1.75(m)	39.36	9	2.43(m)	49.56	17	1.64(m)	53.73	25	5.93(1H, m)	138.8
2	1.65(2H,m)	18.55	10	—	35.46	18	1.58(m)	36.09	26	5.17(1H, bd, 17.1Hz)	114.5
3	1.28(m);1.55(m)	42.77	11	1.50(m);1.66(m)	19.47	19	1.52(m);1.89(m)	35.65	27	5.11(1H, bd, 9.2Hz)	33.16
4	—	33.32	12	1.78(m);1.95(m)	34.44	20	2.18(m);2.33(m)	25.68	28	1.031(3H,s)	21.51
5	1.48 (1H, m)	51.76	13	—	43.95	21	5.44(1H, bt, 6.9Hz)	125.8	29	0.955(3H,s)	13.36
6	1.98(m);2.25(m)	24.76	14	—	51.76	22	—	134.2	30	1.183(3H,s)	27.62
7	5.48(bs)	118.5	15	1.62(m);1.76(m)	34.44	23	2.23 (2H,m)	39.54	31	1.072(3H,s)	22.48
8	—	146.1	16	1.38(m);2.04(m)	28.80	24	2.26(2H,m)	32.79	32	1.055(3H,d, 6.5Hz)	18.82
									33	1.735(3H, s)	16.06

¹³C-NMR



Product 28

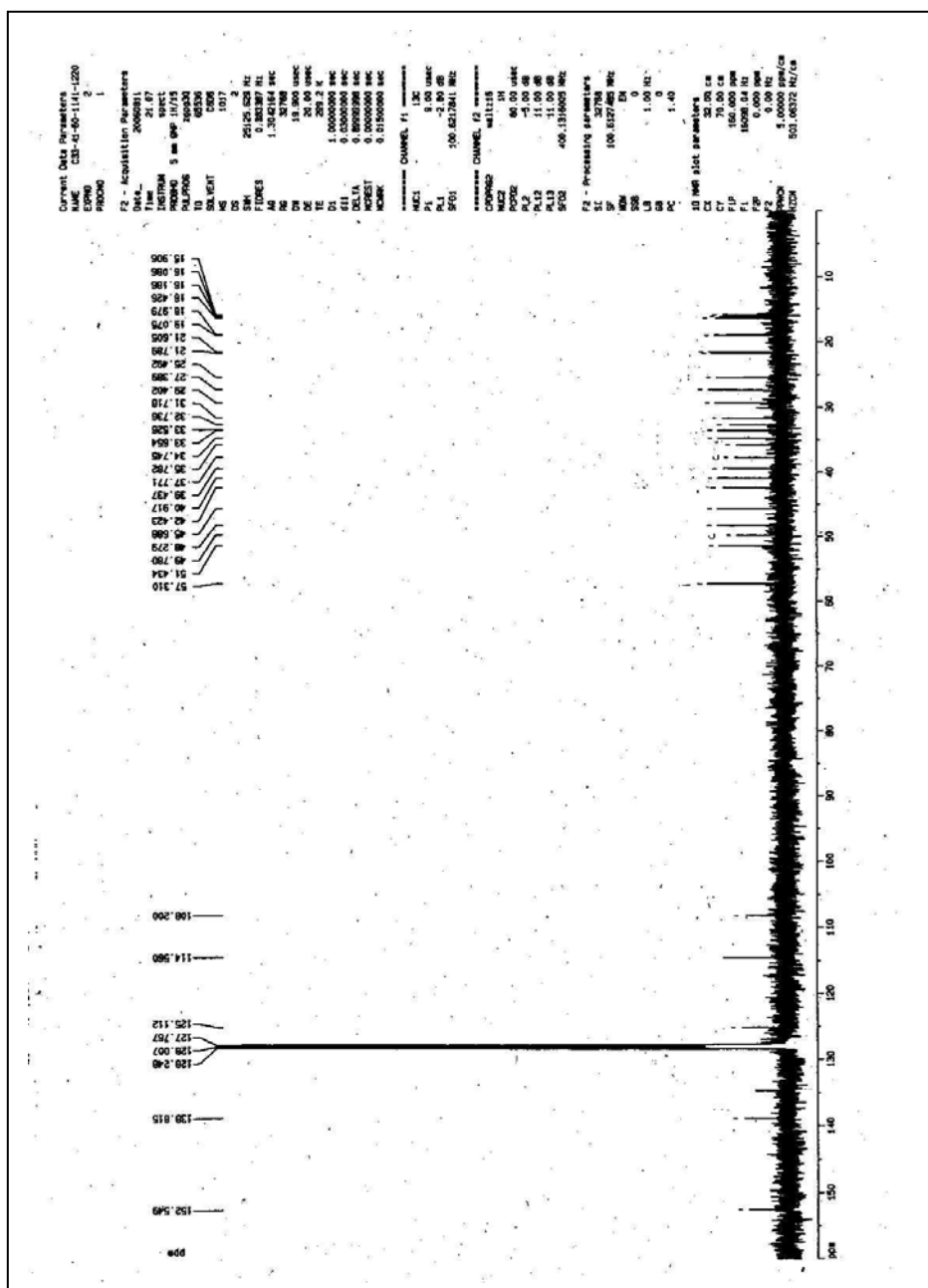
[Mass Spectrum]
 Data : CCG035 Date : 15-Oct-2006 04:49
 Sample: C33-F2-HPLC-1
 Note : -
 Inlet : Direct Ion Mode : EI+
 Spectrum Type : Normal Ion [MF-Linear]
 RT : 0.90 min Scan# : 55
 BP : m/z 191.0000 Int. : 724.70
 Output m/z range : 50.0000 to 500.0000 Cut Level : 0.00 %

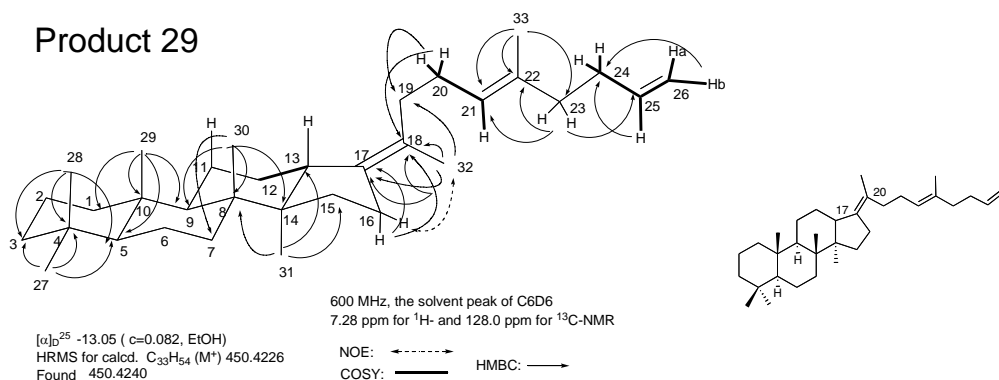
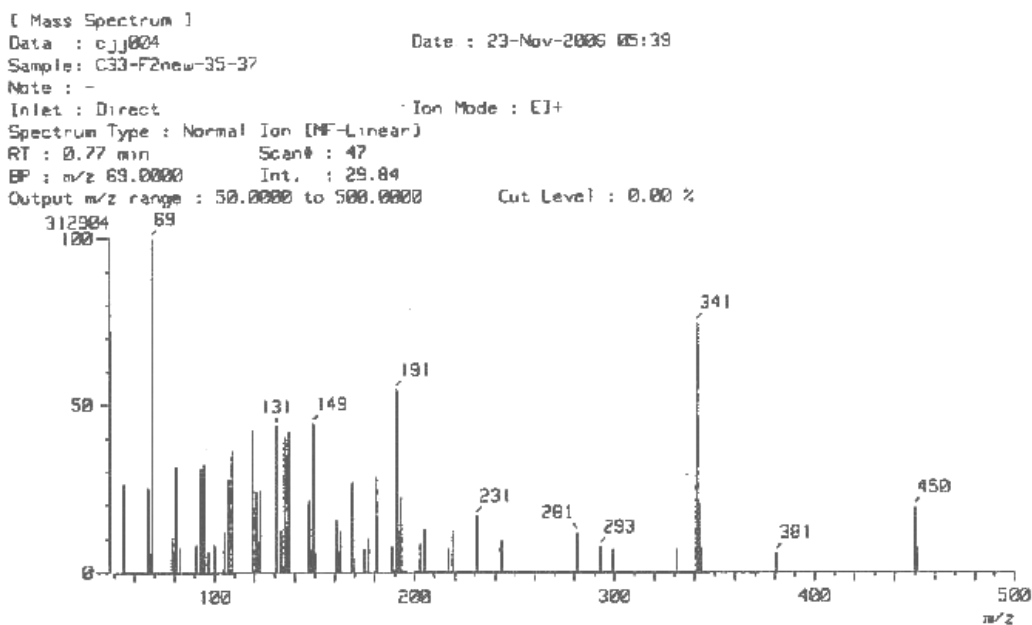
**Product 28**

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	1.72(1H, m); 0.872(1H, ddd, 12.0, 12.0, 4.1Hz)	40.92	9	1.47(1H, m)	51.43	17	2.41(1H, m)	48.28	25	5.93 (1H, m)	138.8
2	1.50(1H, m); 1.63(1H, m)	18.98 ^a	10	—	37.77	18	—	152.5	26	a: 5.17(1H, bdd, 17.2, 1.6Hz) b: 5.12(1H, bd, 11.6)	114.6
3	1.27(1H, m); 1.50(1H, m)	42.42	11	1.28(1H, m); 1.65(1H, m)	21.61	19	2.25(2H, m)	34.75	27	1.030(3H, s)	33.65
4	—	33.53	12	1.30(1H, m); 1.90(1H, m)	25.49	20	2.38(2H, m)	27.39	28	0.984(3H, s)	21.79
5	0.911(1H, dd, 2.0, 12.0Hz)	57.31	13	1.91(1H, m)	45.69	21	5.41(1H, t, 6.4)	125.1	29	1.014(3H, s)	16.43
6	1.50(1H, m); 1.63(1H, m)	19.08 ^a	14	—	49.78	22	—	134.6	30	1.109(3H, s)	16.09
7	1.37(1H, m); 1.72(1H, m)	35.78	15	1.24(1H, m); 1.76(1H, m)	31.72	23	2.18(2H, m)	39.44	31	0.971(3H, s)	16.19
8	—	40.97	16	1.63(1H, m); 2.08(1H, m)	29.40	24	2.22(2H, m)	32.74	32	5.10 (1H, s); 5.05 (1H, s)	108.2

a: The carbon signals at the positions of 2 and 6 may be exchangeable

¹³C-NMR

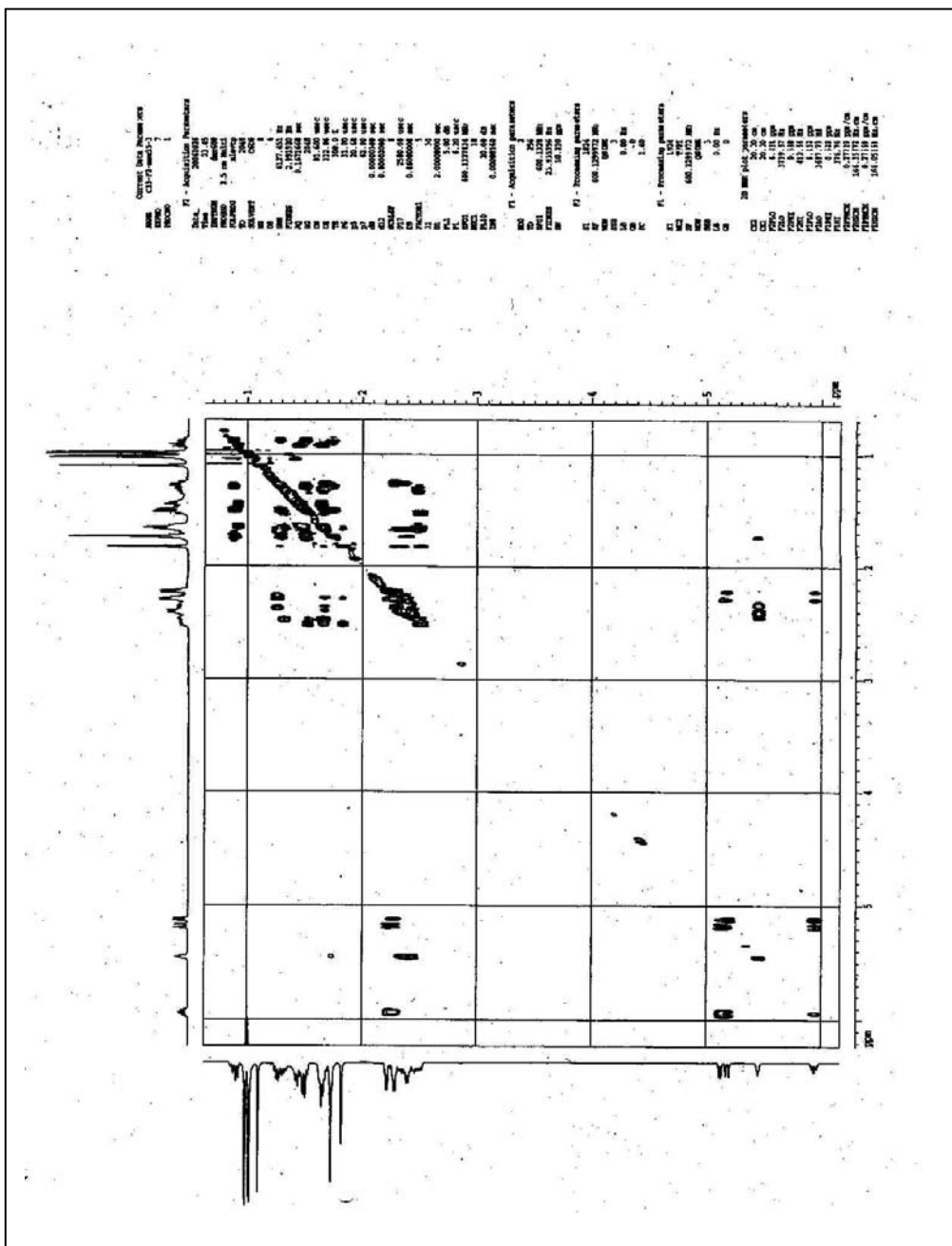


Product 29

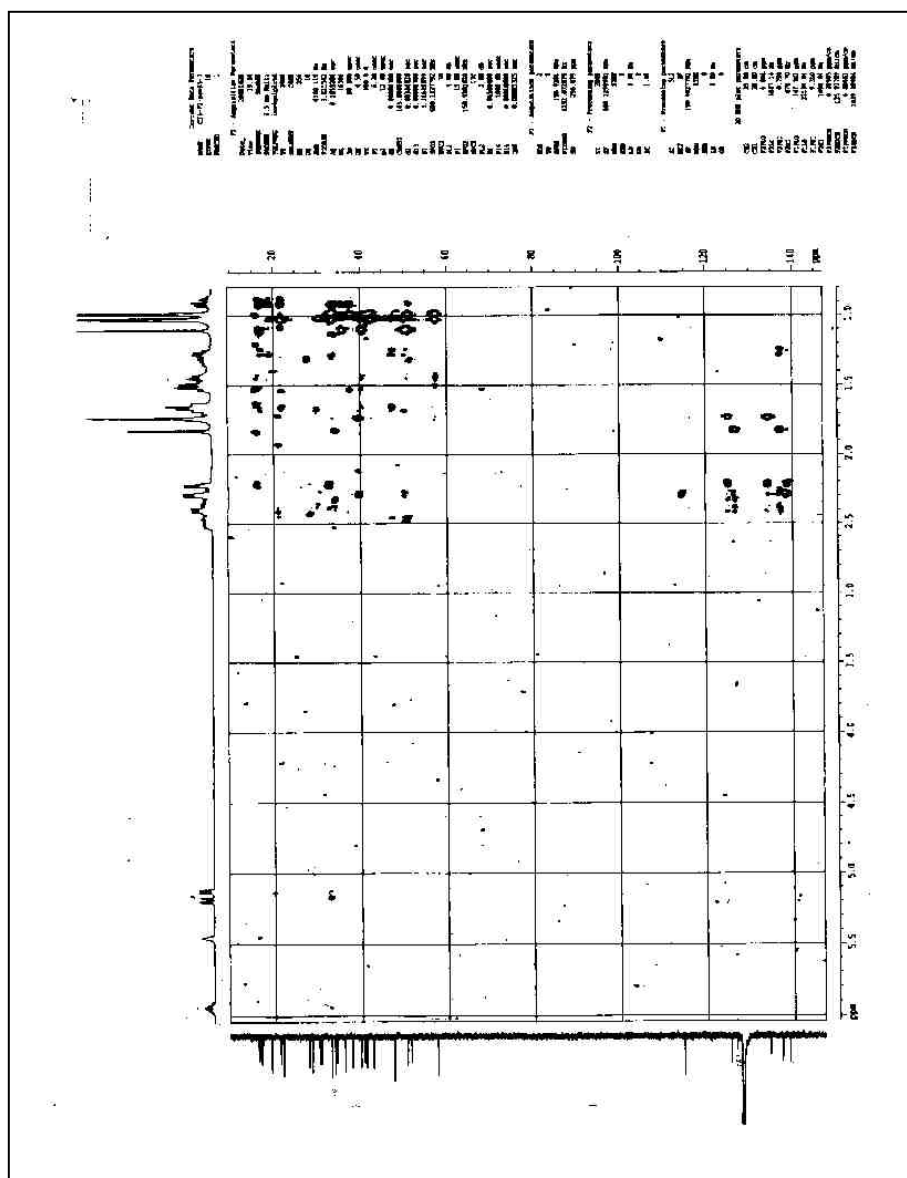
NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C	NO.	¹ H	¹³ C
1	0.88(1H, m); 1.75(1H, m)	40.87	9	1.49(1H, m)	51.08	17	—	137.1	25	5.94(1H, m)	138.8
2	1.47(1H, m); 1.63(1H, m)	18.93 ^a	10	—	37.62	18	—	126.7	26	a: 5.18(1H, bd, 17.1) b: 5.12(1H, bd, 10.0Hz)	114.5
3	1.27(1H, m); 1.47(1H, m)	42.44	11	1.28(1H, m); 1.68(1H, m)	21.78	19	2.41(2H, m)	34.13	27	1.029(3H, s)	33.59
4	—	33.51	12	1.65(1H, m); 2.48(1H, m)	25.49	20	2.30(1H, m), 2.38(1H, m)	28.39	28	0.984(3H, s)	21.74
5	0.91(1H, bd, 11.8Hz)	57.33	13	2.50(1H, m)	47.27	21	5.45(1H, m)	125.3	29	0.991(3H, s)	16.58
6	1.47(1H, m); 1.63(1H, m)	19.05 ^a	14	—	50.25	22	—	134.3	30	1.102(3H, s)	15.92
7	1.43(1H, m); 1.63(1H, m)	35.89	15	1.25(1H, m); 1.65(1H, m)	30.53	23	2.21(2H, m)	39.53	31	1.021(3H, s)	16.90
8	—	40.36	16	2.25(1H, m); 2.36(1H, m)	30.05	24	2.21(1H, m); 2.29(1H, m)	32.77	32	1.826(3H, bs)	21.08
									33	1.736(3H, s)	16.08

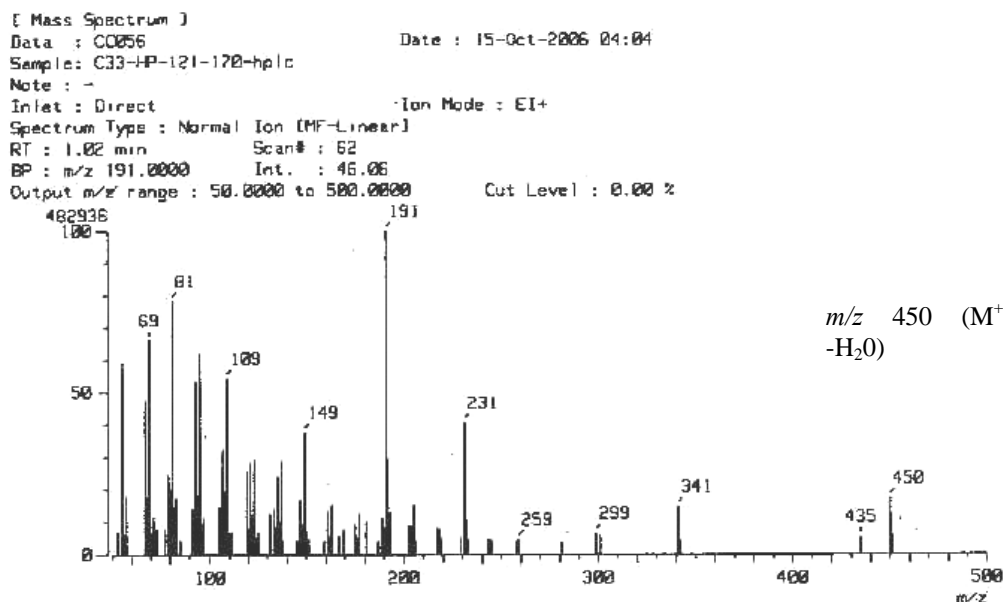
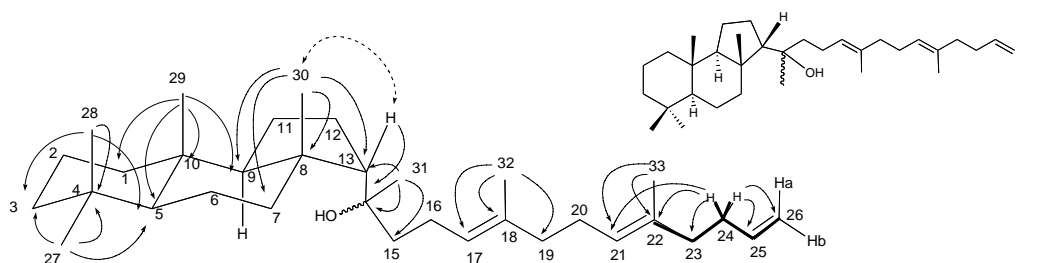
a: the assignments of C2 and C6 may be exchangeable.

HOHAHA



HMBC



Product 30**Product 30 (oil)**

$[\alpha]_D^{25}$ -70.80 (EtOH, c=0.05)

HRMS m/z 450 ($M^+ - H_2O$) Calcd: 450.4226

Found: 450.4228

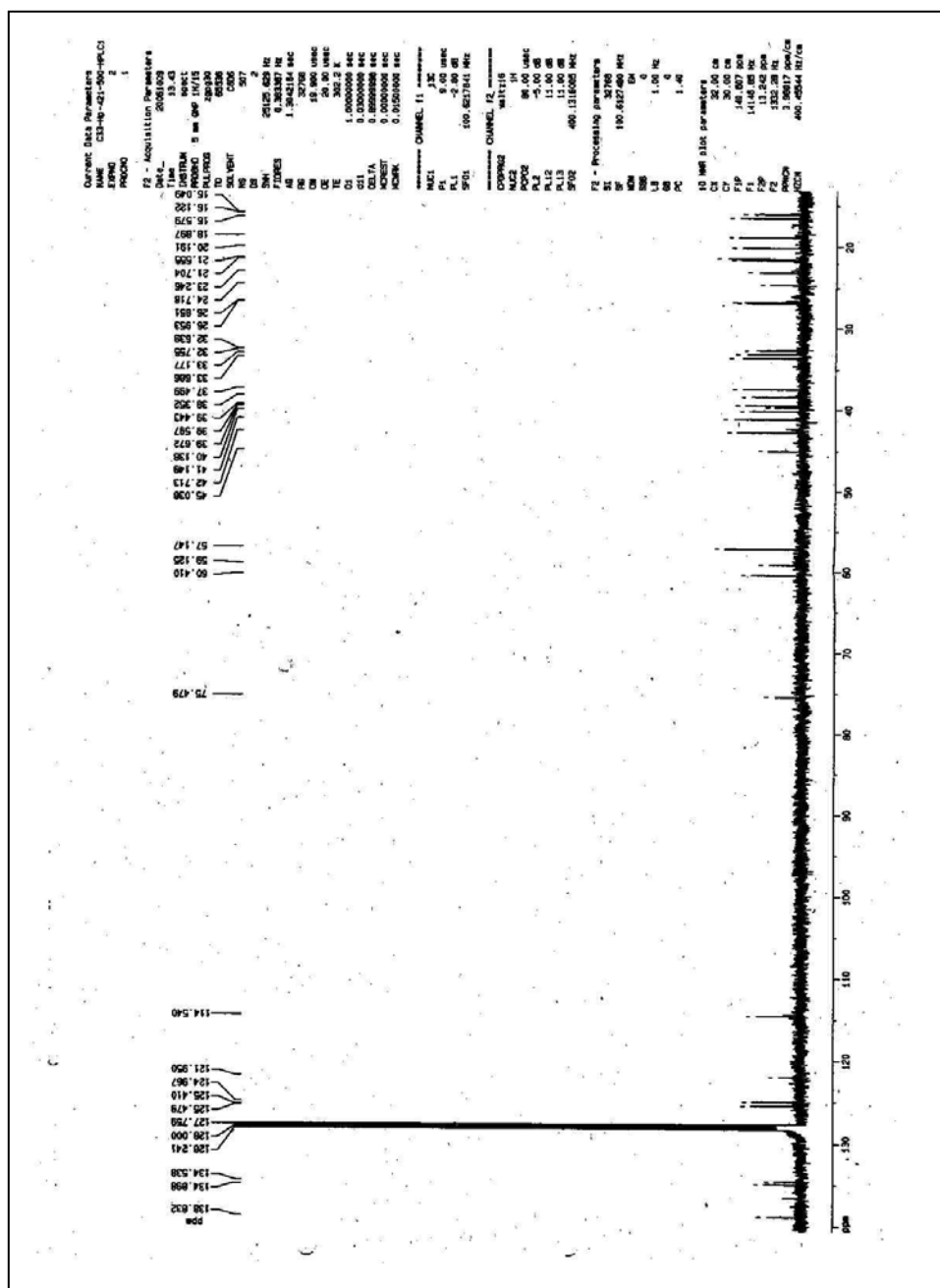
400MHz in C_6D_6
 the solvent peak 1H : 7.28 ppm; ^{13}C : 128.0 ppm

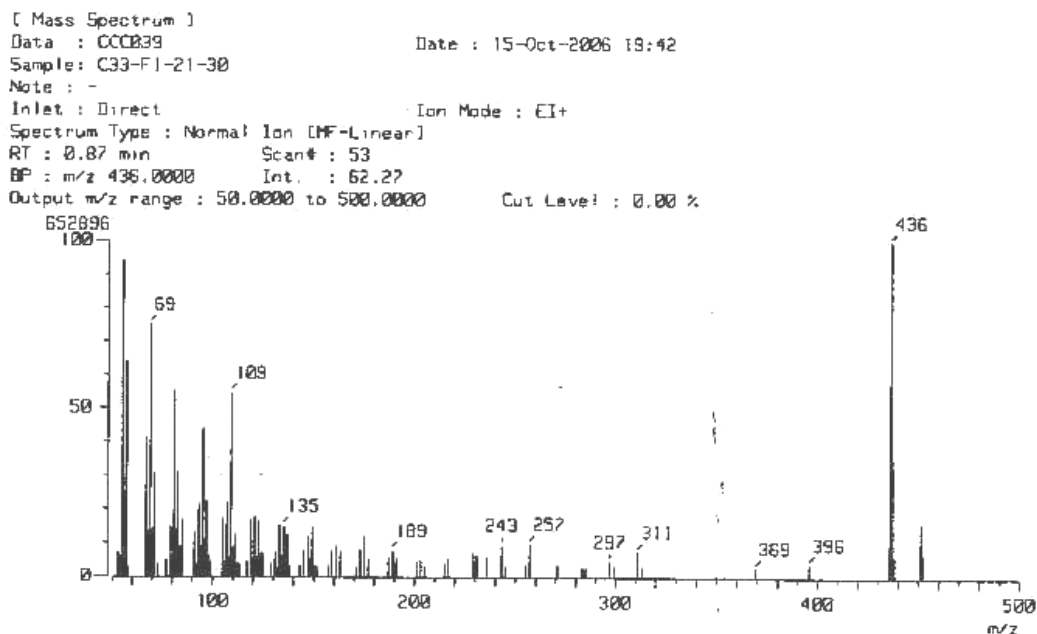
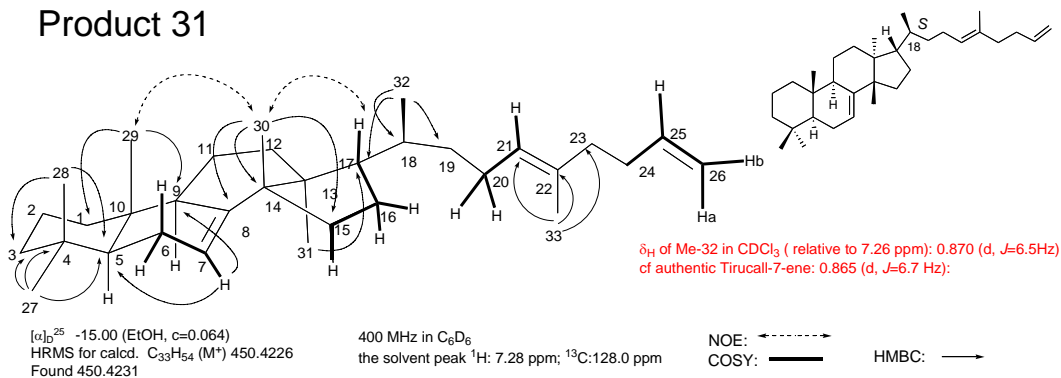
NOE: \longleftrightarrow
 COSY: \longrightarrow HMBC: \longrightarrow

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	1.04(1H, m); 1.58(1H, m)	41.13	9	1.54(1H, m)	60.39	17	5.42(1H, m)	125.4	25	5.94(1H, m)	137.6
2	1.47(1H, m); 1.73(1H, m)	18.89	10	—	37.48	18	—	135.0	26	a:5.21(1H, bdd, 16.8, 2) b:5.13(1H, bd, 10.0)	114.3
3	1.25(1H, m); 1.50(1H, m)	42.69	11	1.47(1H, m); 1.65(1H, m)	21.70	19	2.12(1H, bt, 7.0)	39.67	27	1.002(3H, s)	33.68
4	—	33.16	12	1.68(1H, m); 1.94(1H, m)	24.71	20	2.27(2H, m)	26.63	28	0.975(3H, s)	21.54
5	0.99(1H, m)	57.13	13	1.76(1H, m)	59.11	21	5.42(1H, m)	121.9	29	0.982(3H, s)	16.57
6	1.52(1H, m); 1.73(1H, m)	20.17	14	—	75.49	22	—	136.5	30	1.068(3H, s)	26.83
7	1.94(1H, m); 2.12(1H, m)	38.32	15	1.70(1H, m); 2.10(1H, m)	42.31 ^a	23	2.12(1H, bt, 7.0)	39.59	31	1.308(3H, s)	26.08 ^a
8	—	45.04	16	2.27(2H, m)	23.23	24	2.34(2H, m)	32.70	32	1.760(3H, s)	16.04

a: C-31 and C-15 signal are significantly small and broad.

¹³C-NMR

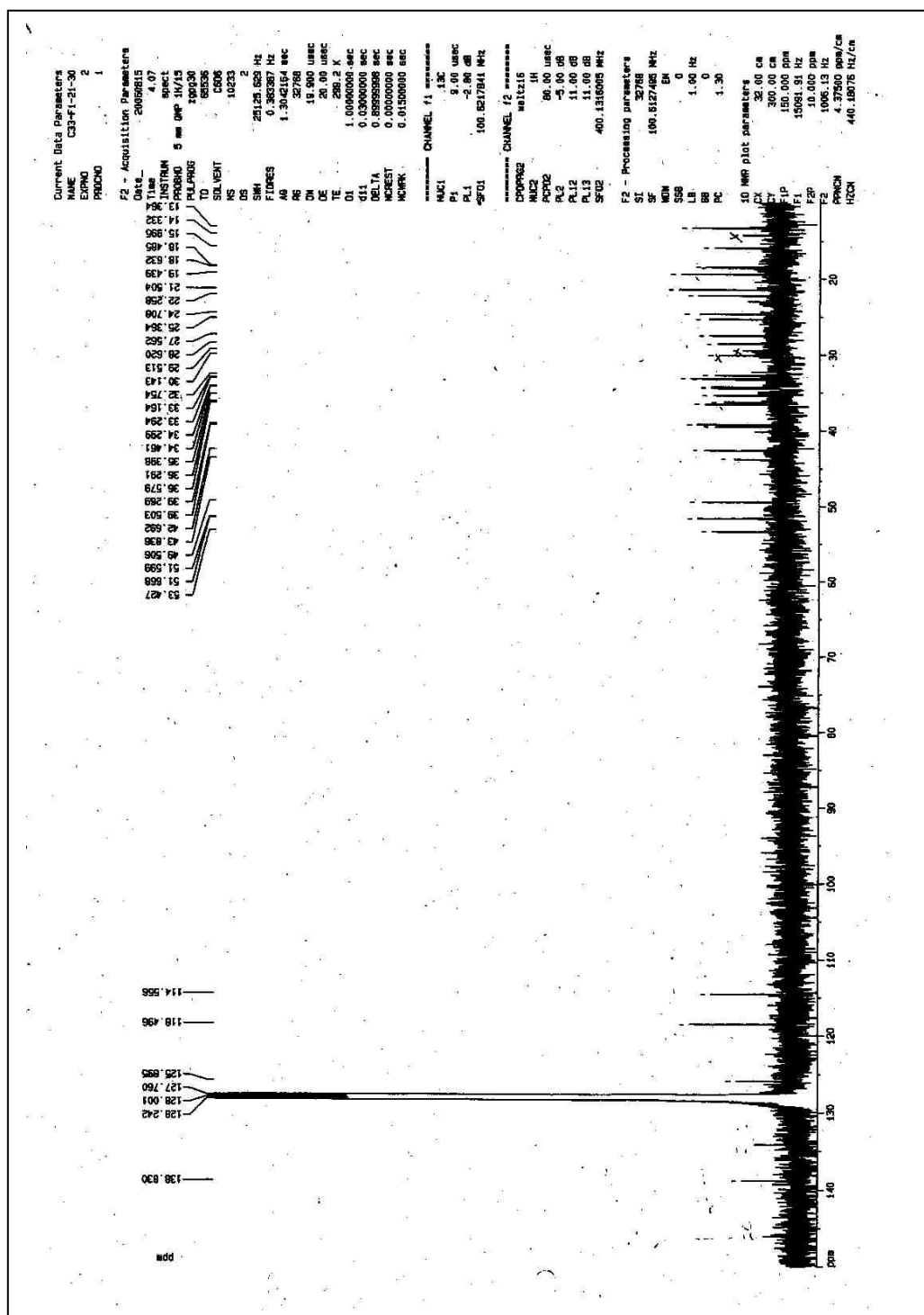


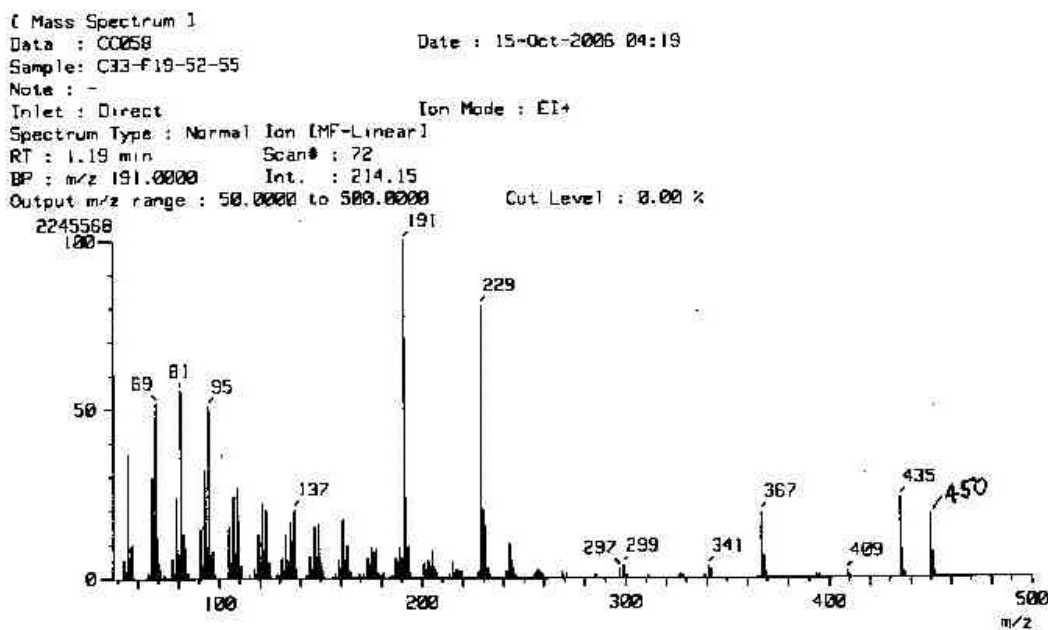
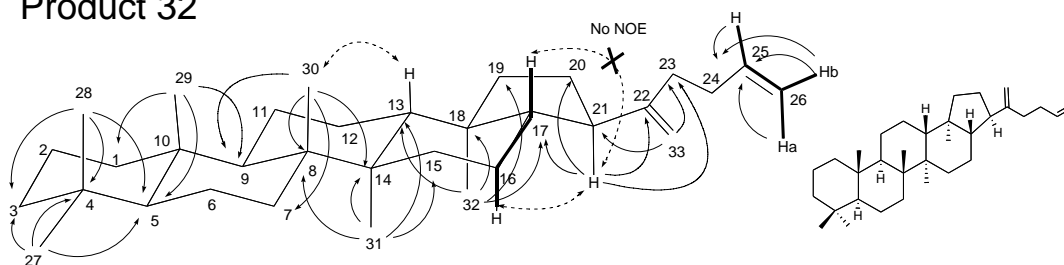
Product 31**Product 31**

NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C	NO.	^1H	^{13}C
1	1.05(1H, m); 1.75(1H, m)	39.27	9	2.43(1H, m)	49.51	17	1.63(1H, m)	53.43	25	5.94(1H, m)	138.8
2	1.60(1H, m); 1.68(1H, m)	18.49	10	—	35.40	18	1.58(m)	36.29	26	b:5.12(1H, d, J 9.5) a:5.18(1H, bd, J 17.2)	114.6
3	1.29(1H, m); 1.55(1H, m)	42.69	11	1.55(1H, m); 1.69(1H, m)	19.44	19	1.27(1H, m); 1.59(1H, m)	36.58	27	0.971(3H, s)	33.16
4	—	33.29	12	1.78(m); 1.89(1H, m)	34.16	20	2.17(1H, m); 2.31(1H, m)	25.36	28	1.034(3H, s)	21.50
5	1.50(1H, m)	51.67	13	—	43.84	21	5.42(1H, t, J 6.4)	125.9	29	0.965(3H, s)	13.36
6	1.97(1H, m); 2.25(1H, m)	24.71	14	—	51.60	22	—	134.1	30	1.186(3H, s)	27.56
7	5.47(1H, bs)	118.5	15	1.65(1H, m); 1.78(1H, m)	34.46	23	2.24(2H, m)	39.50	31	1.058(3H, s)	22.26
8	—	146.0	16	1.40(1H, m); 2.12(1H, m)	28.62	24	2.28(2H, m)	32.75	32	1.107(3H, d, 6.0Hz)	18.63
									33	1.730(3H, s)	15.99

The assignments of the proton and carbon at 12 and 15 position may be exchangeable

¹³C-NMR



Product 32**Product 32**

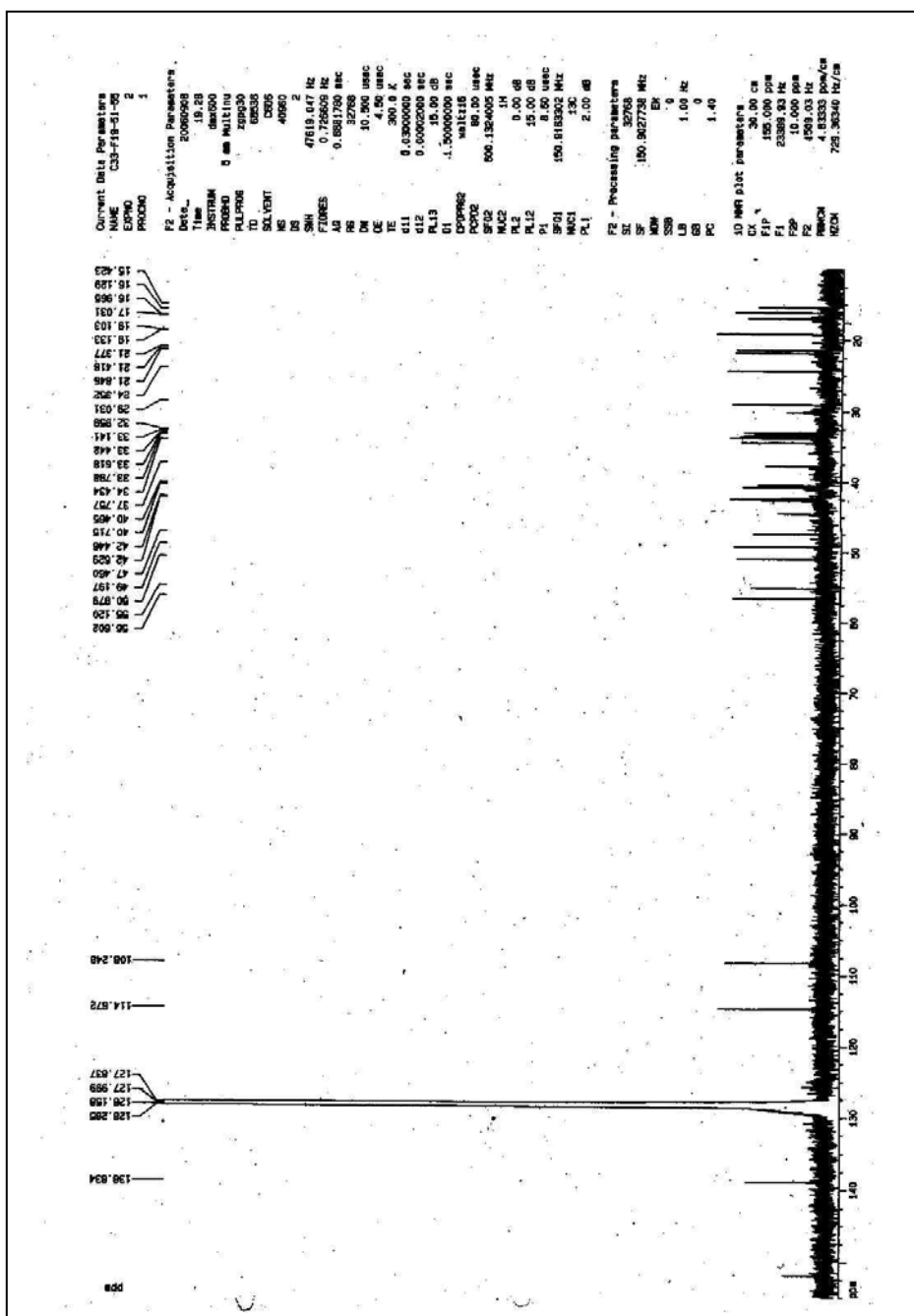
$[\alpha]_D^{25}$ -52.39 (EtOH, c=0.046)
 HRMS for calcd. $C_{33}H_{54}$ (M^+) 450.4226
 Found 450.4213

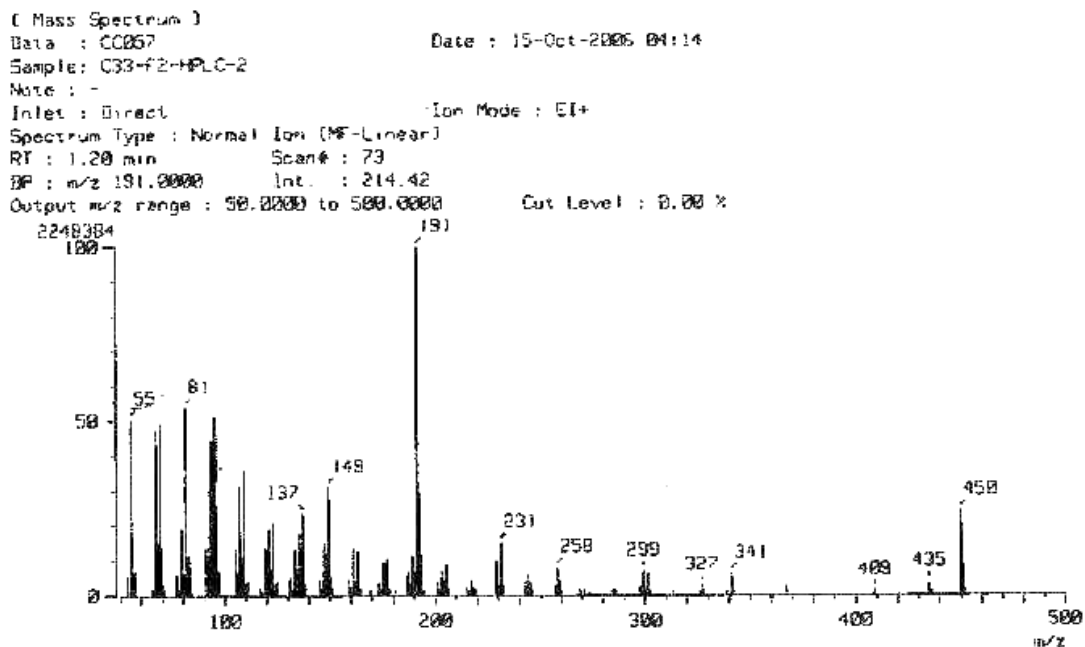
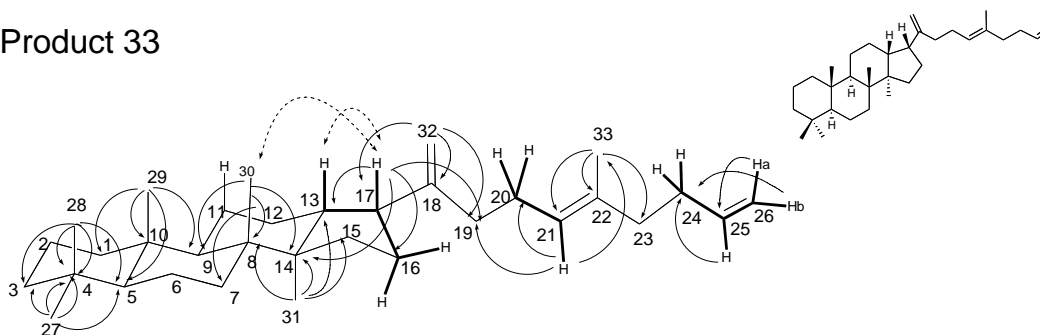
600 MHz, the solvent peak of C_6D_6
 7.28 ppm for 1H - and 128.0 ppm for ^{13}C -NMR

NOE: \dashrightarrow
 COSY: \longrightarrow HMBC: \longrightarrow

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	0.87(1H, m); 1.78(1H, m)	40.71	9	1.41(1H, m)	50.98	17	1.28(1H, m)	55.12	25	5.96(1H, m)	138.8
2	1.42(1H, m); 1.63(1H, m)	19.10	10	—	37.76	18	—	44.55	26	a: 5.20(1H, bd, 17.1, b: 5.13 (1H, bd, 10.5 Hz)	114.7
3	1.25(1H, m); 1.51(1H, m)	42.45	11	1.30(1H, m); 1.42(1H, m)	21.38	19	1.19(1H, m); 1.63(1H, m)	40.47	27	1.037(3H, s)	33.62
4	—	33.44	12	1.55(1H, m); 1.64(1H, m)	24.35	20	1.58(1H, m); 2.06(1H, m)	29.03	28	0.989(3H, s)	21.85
5	0.91(1H, m)	56.60	13	1.58(1H, m)	49.20	21	2.41(1H, m)	47.46	29	0.979(3H, s)	16.13
6	1.51(1H, m); 1.72(1H, m)	19.13	14	—	42.27	22	—	151.9	30	1.106(3H, s)	17.03
7	1.35(1H, m); 1.61(1H, m)	33.80	15	1.33(1H, m); 1.54(1H, m)	33.14	23	2.24(2H, t, 7.7Hz)	34.43	31	1.114(3H, s)	16.96
8	—	42.63	16	1.65(1H, m); 1.72(1H, m)	21.42	24	2.37(2H, m)	32.96	32	0.845(3H, s)	15.42
									33	5.00(1H, bs); 5.07(1H, bs)	108.2

The carbon assignments between 11 and 16-positions, between 2 and 6-positions and between 30 and 31 may be interchangeable

¹³C-NMR

Product 33**Product 33**

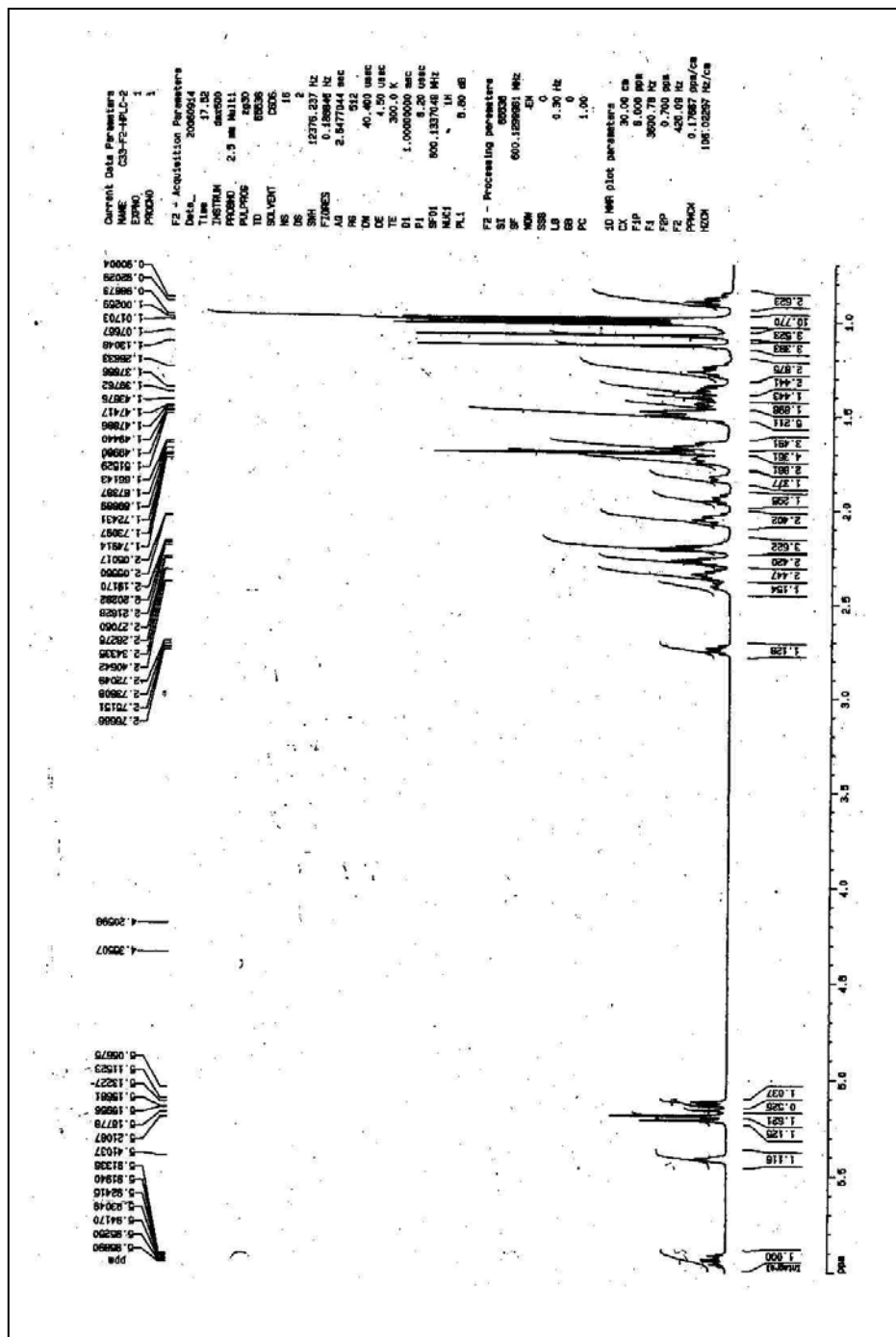
$[\alpha]_D^{25}$ -51.64 (c=0.056, EtOH)
 HRMS for calcd. $C_{33}H_{54}$ (M^+) 450.4226
 Found 450.4229

600 MHz, the solvent peak of C_6D_6
 7.28 ppm for 1H - and 128.0 ppm for ^{13}C -NMR

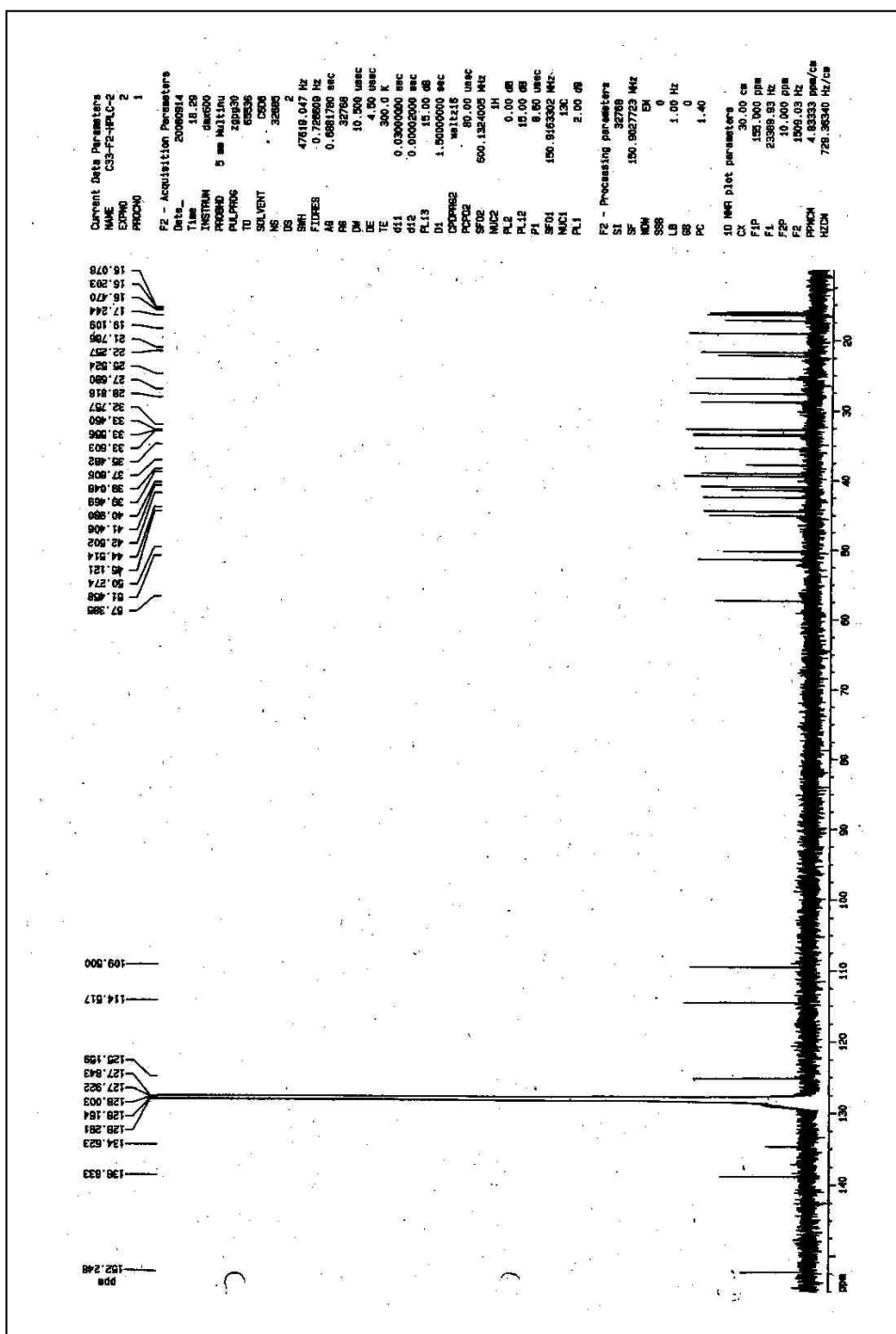
NOE: \longleftrightarrow
 COSY: \longrightarrow HMBC: \longrightarrow

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	0.88(1H, m); 1.75(1H, m)	40.98	9	1.48(1H, m)	51.46	17	2.74(1H, m)	44.51	25	5.93 (1H, m)	138.8
2	1.48(1H, m); 1.65(1H, m)	19.11	10	—	37.81	18	—	152.2	26	a: 5.17 (1H, bd, 16.9Hz) b: 5.12 (1H, d, 10.1Hz)	114.5
3	1.25(1H, m); 1.50(1H, m)	42.50	11	1.27(1H, m); 1.70(1H, m)	22.26	19	2.36(1H, m); 2.20(1H, m)	39.05	27	1.017(3H, s)	33.60
4	—	33.56	12	1.45(1H, m); 1.83(1H, m)	25.52	20	2.42(1H, m); 2.35(1H, m)	27.68	28	0.989(3H, s)	21.79
5	0.910(1H, bd, 12.1Hz)	57.39	13	2.06(1H, m)	45.12	21	5.41(1H, bs)	125.2	29	1.003(3H, s)	16.47
6	1.48(1H, m); 1.65(1H, m)	19.11	14	—	50.27	22	—	134.6	30	1.077(3H, s)	16.09
7	1.40(1H, m); 1.72(1H, m)	35.48	15	1.34(1H, m); 1.68(1H, m)	33.46	23	2.36(1H, m); 2.20(1H, m)	39.47	31	1.131(3H, s)	17.24
8	—	41.41	16	1.93(1H, m); 2.03(1H, m)	28.82	24	2.27(1H, m); 2.21(1H, m)	32.76	32	5.21(1H, s); 5.19(1H, s)	109.5
									33	1.699(3H, s)	16.20

¹H-NMR

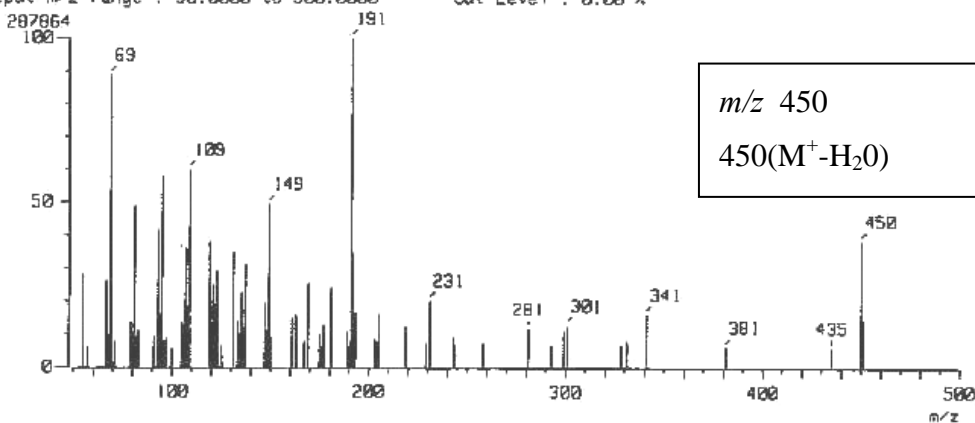
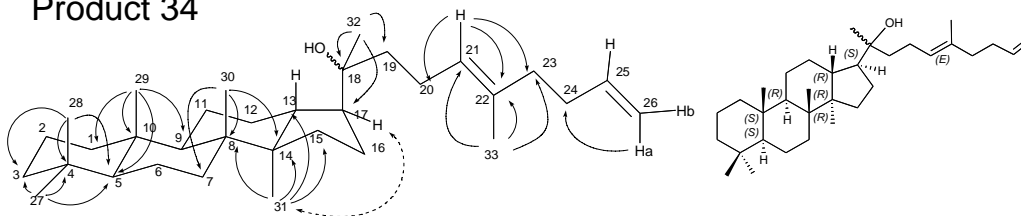


¹³C-NMR



Product 34

[Mass Spectrum]
 Data : ccj050 Date : 23-Nov-2006 06:04
 Sample: C33-tp-161-180
 Note : -
 Inlet : Direct Ion Mode : EI+
 Spectrum Type : Normal Ion [MF-Linear]
 RT : 0.85 min Scan# : 52
 BP : m/z 191.0000 Int. : 27.45
 Output m/z range : 50.0000 to 500.0000 Cut Level : 0.00 %

**Product 34**

$[\alpha]_D^{25}$ +20.28 (EtOH, c=0.036)
 HRMS for calcd. $C_{33}H_{54}$ ($M^+ - H_2O$) 450.4226
 Found 450.4247

600 MHz, the solvent peak of C_6D_6
 7.28 ppm for 1H - and 128.0 ppm for ^{13}C -NMR

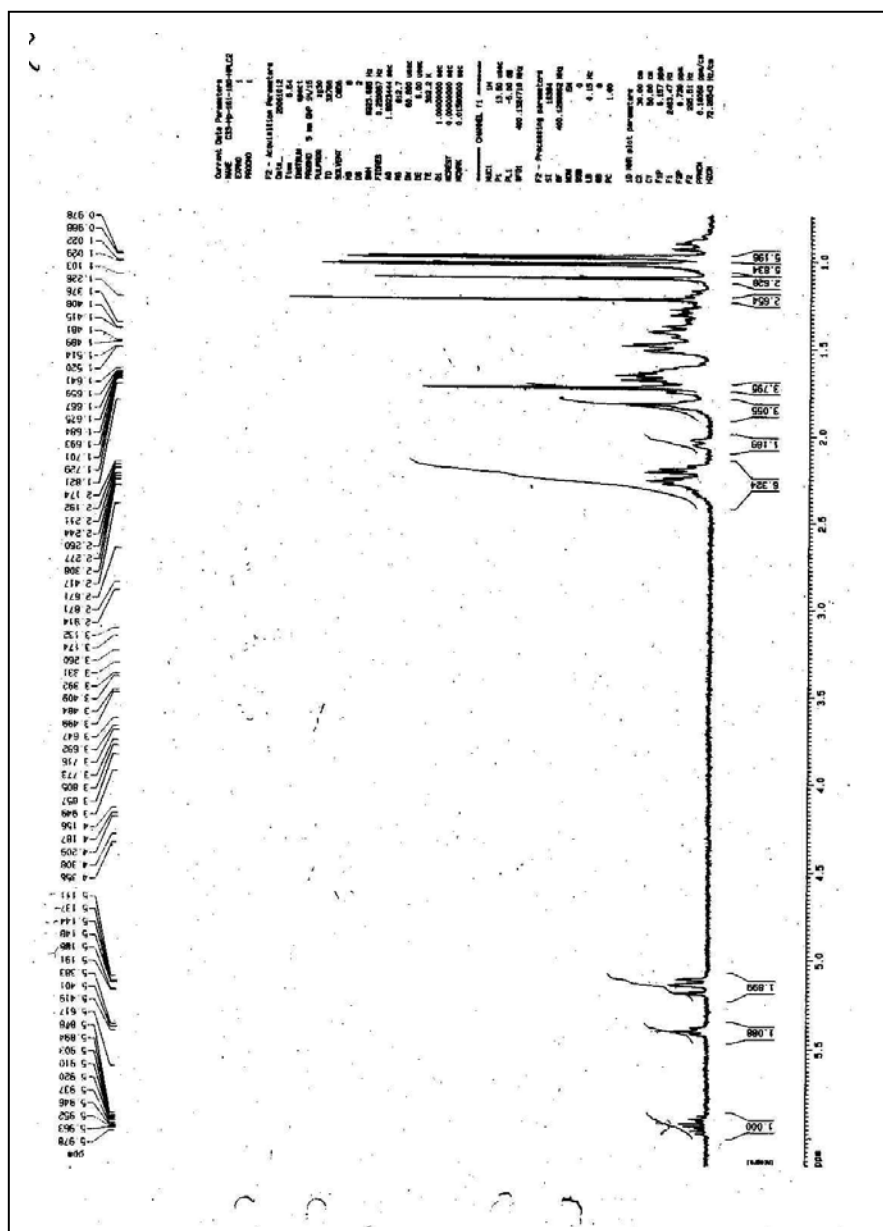
NOE: \longleftrightarrow HMBC: \longrightarrow
 COSY: \longrightarrow

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	0.88(1H, m); 1.75(1H, m)	40.89	9	1.46(1H, m)	51.20	17	1.80(1H, m)	50.40	25	5.93(1H, m)	138.8
2	1.48(1H, m); 1.62(1H, m)	19.00	10	—	37.70	18	—	74.77	26	Ha: 5.19(1H, bd, 17.2); Hb: 5.13(1H, bd, 10.4)	114.6
3	1.27(1H, m); 1.48(1H, m)	42.48	11	1.27(1H, m); 1.61(1H, m)	21.76	19	1.66(2H, m)	40.98	27	1.029(3H, s)	33.63
4	—	33.52	12	1.66(1H, m); 1.72(1H, m)	25.18	20	2.26(2H, m)	22.93	28	0.988(3H, s)	21.76
5	0.93(1H, dd, 12.0, 1.6)	57.34	13	1.81(1H, m)	42.48	21	5.40(1H, t, 7.2)	125.7	29	0.978(3H, s)	16.43
6	1.48(1H, m); 1.62(1H, m)	19.09	14	—	50.62	22	—	134.4	30	1.103(3H, s)	15.79
7	1.37(1H, m); 1.68(1H, m)	35.64	15	1.19(1H, m); 1.62(1H, m)	31.55	23	2.19(2H, m)	39.44	31	1.022(3H, s)	16.73
8	—	40.89	16	1.32(1H, m); 2.03(1H, m)	28.02	24	2.22(2H, m)	32.70	32	1.226(3H, s)	25.82
									33	1.729(3H, s)	16.03

1. The assignments of H2 and H6 and those of C2 and C6 may be exchangeable.
 2. C15 and C24 are exchangeable due to the close values of chemical shifts.

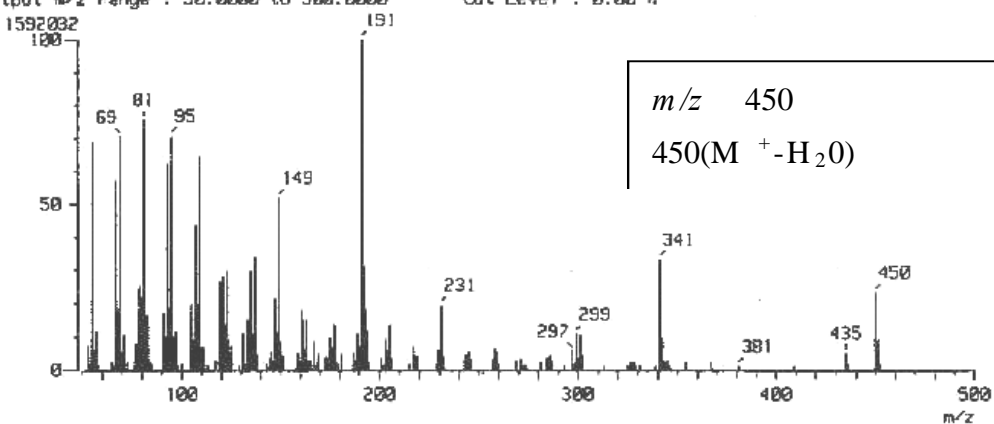
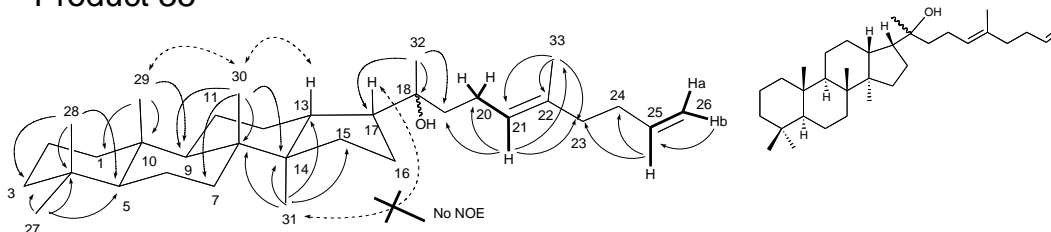
3. The assignments of C11, C12 and C16 may be exchangeable together with H11, H12 and H16.
 4. The assignments of C1 and C19 may be exchangeable due to the close chemical shifts.

$^1\text{H-NMR}$



Product 35

[Mass Spectrum]
 Data : CC054 Date : 15-Oct-2009 03:49
 Sample: C33-HP-261-340
 Note : -
 Inlet : Direct Ion Mode : EI+
 Spectrum Type : Normal Ion (MF-Linear)
 RT : 1.73 min Scan# : 105
 BP : m/z 191.0000 Int. : 151.83
 Output m/z range : 50.0000 to 500.0000 Cut Level : 0.00 %

**Product 35**

$[\alpha]_D^{25}$ -18.21 (EtOH, c=0.2)

HRMS for calcd. $C_{33}H_{54}$ ($M^+ - H_2O$) 450.4226
 Found 450.4247

400 MHz, the solvent peak of C_6D_6
 7.28 ppm for 1H - and 128.0 ppm for ^{13}C -NMR

NOE: \dashrightarrow COSY: \longrightarrow HMBC: \dashrightarrow

NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C	NO.	1H	^{13}C
1	0.92(1H, m); 1.77(1H, m)	40.88	9	1.54(1H, m)	51.14	17	2.10(1H, m)	48.38	25	5.91(1H, m)	138.8
2	1.49(1H, m); 1.67(1H, m)	19.11	10	—	37.71	18	—	74.59	26	Hb: 5.12(1H, bd, 10.0 Hz) Ha: 5.17(1H, bd, 16.8 Hz)	114.6
3	1.31(1H, m); 1.51(1H, m)	42.49	11	1.22(1H, m); 1.68(1H, m)	22.88	19	1.62(2H, m)	42.67	27	1.025(3H, s)	33.62
4	—	33.54	12	1.64(1H, m); 2.04(1H, m)	26.42	20	2.21(2H, m)	23.42	28	0.994(3H, s)	21.78
5	0.94(1H, dd, 12.0, 2.0)	57.34	13	2.13(1H, m)	44.07	21	5.37(1H, t, 6.0)	125.6	29	1.008(3H, s)	16.50
6	1.52(1H, m); 1.75(1H, m)	19.15	14	—	49.48	22	—	134.4	30	1.092(3H, s)	16.17
7	1.41(1H, m); 1.73(1H, m)	35.73	15	1.26(1H, m); 1.56(1H, m)	32.76	23	2.20(2H, m)	39.43	31	1.218(3H, s)	17.16
8	—	41.19	16	1.77(2H, m)	26.75	24	2.26(2H, m)	32.72	32	1.308(3H, s)	27.69
									33	1.722(3H, s)	16.03

1. The assignments of H2 and H6 and those of C2 and C6 may be exchangeable.
 2. C15 and C24 are exchangeable due to the close chemical shifts.

3. The assignments of C11, C12 and C16 may be exchangeable together with H11, H12 and H16

$^1\text{H-NMR}$

