Figure 1

Structure of six anticancer Saponins with biangeloyl groups (Y3 (Y), Y2, Y8, Y10, Y1 and Y9)

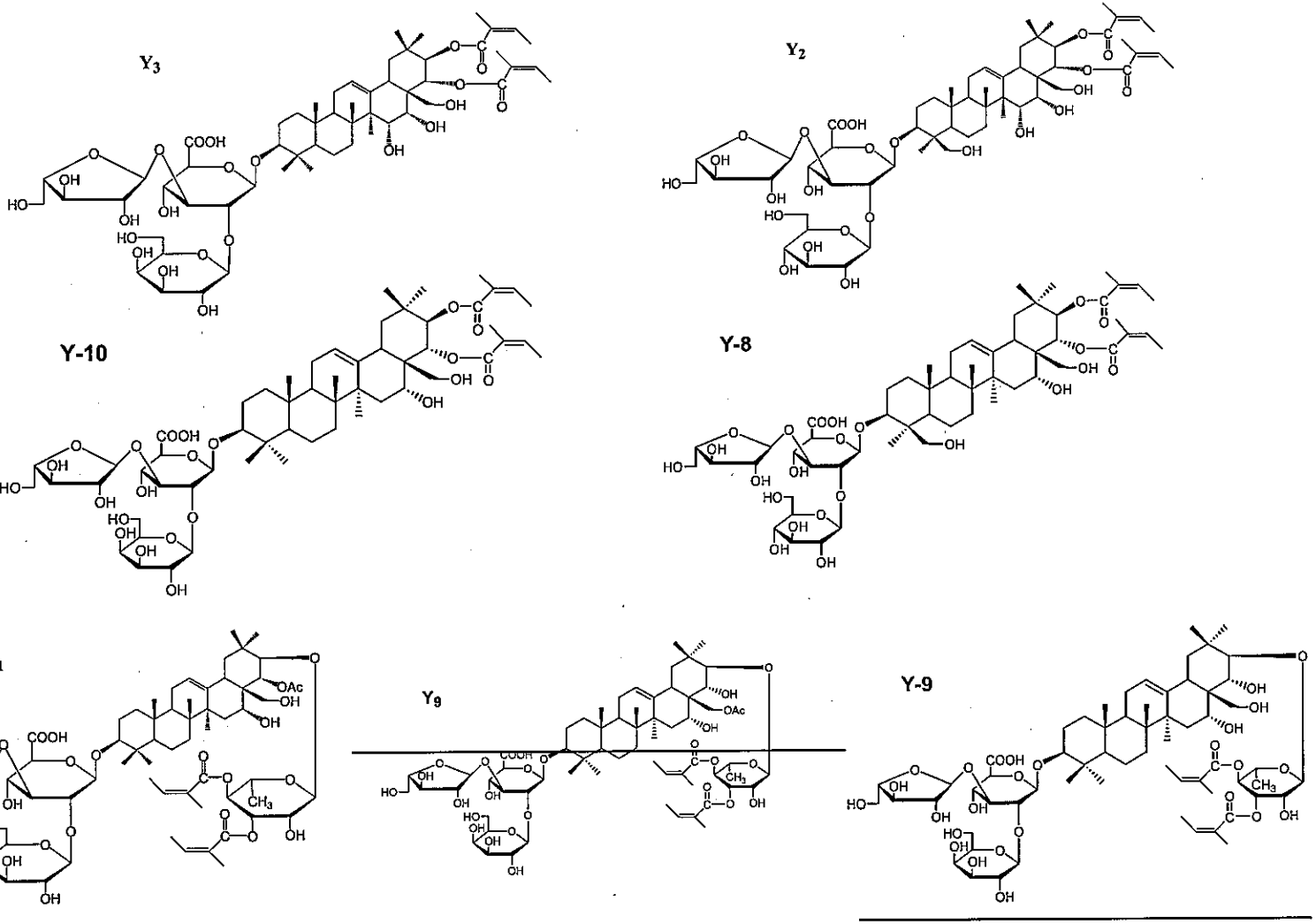
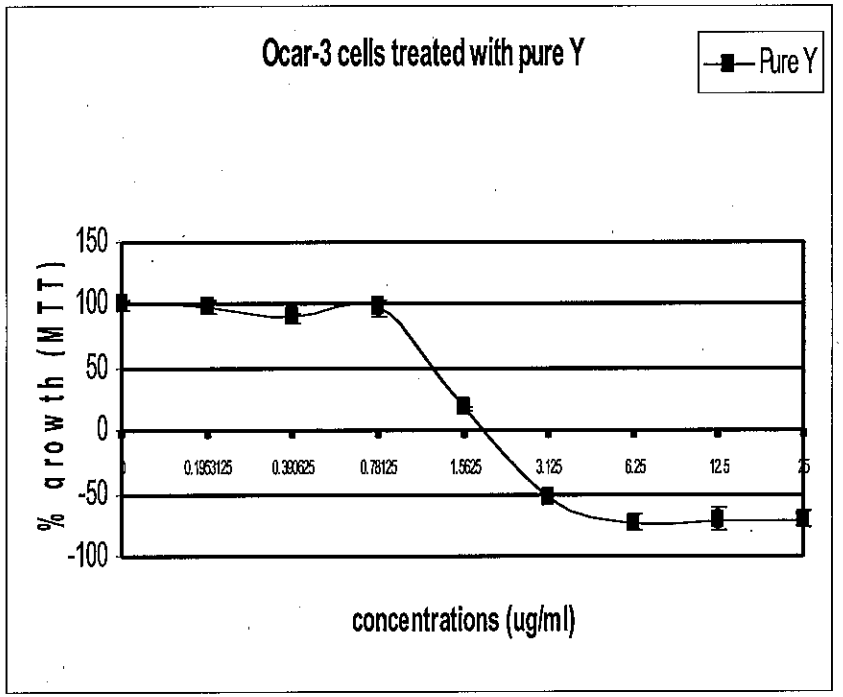


Figure 2

Anticancer activity of Compound Y

(A) Points graph



(B) X,Y linear graph

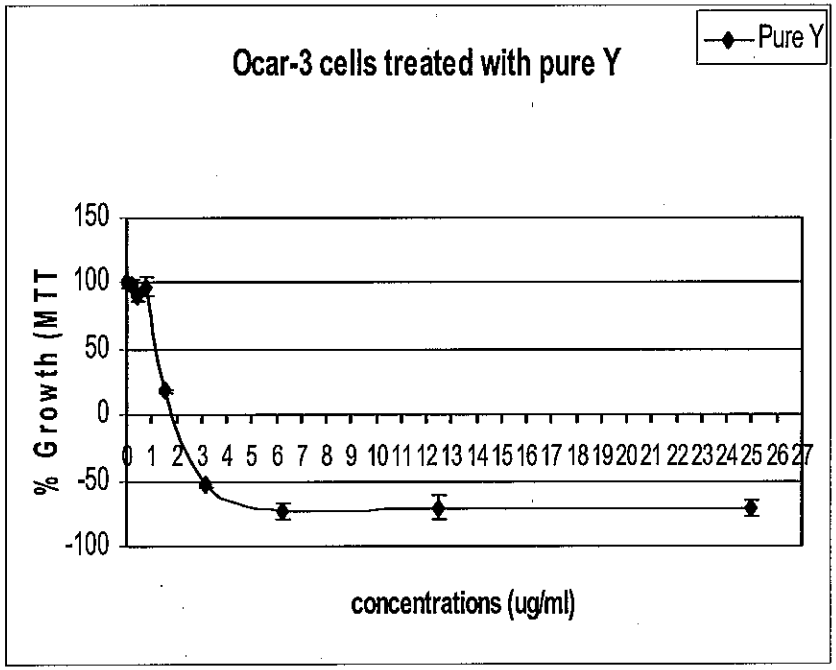


Figure 3

Y1 and Y2 activity on Ovarian caner cells

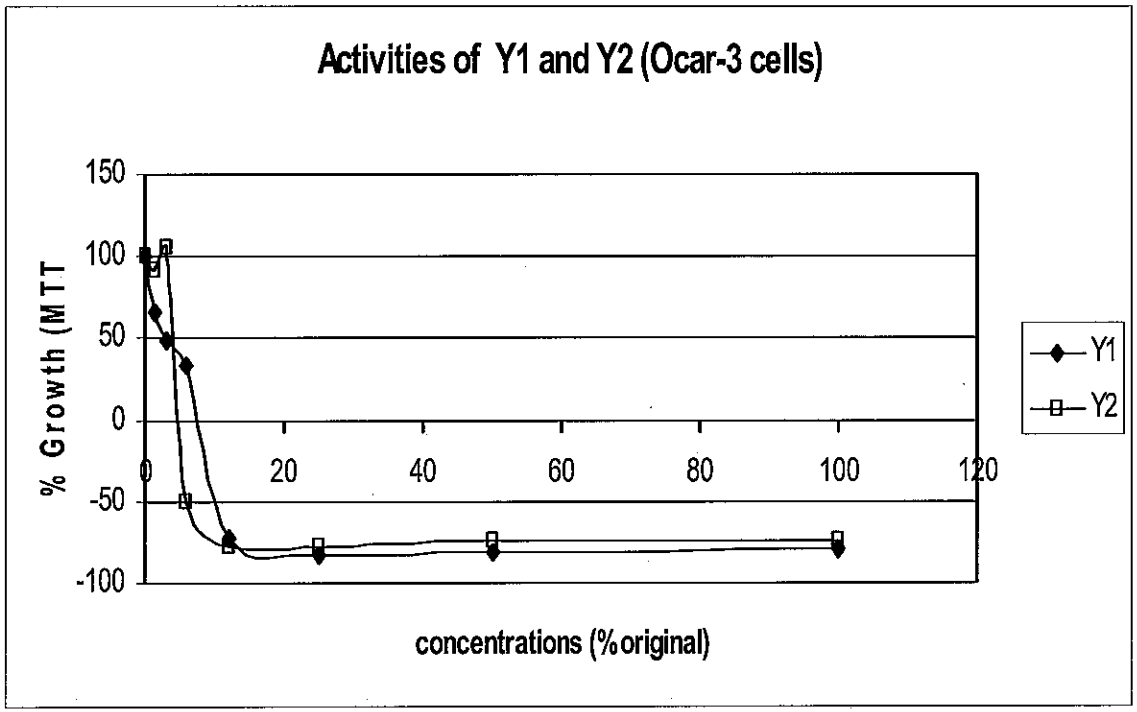
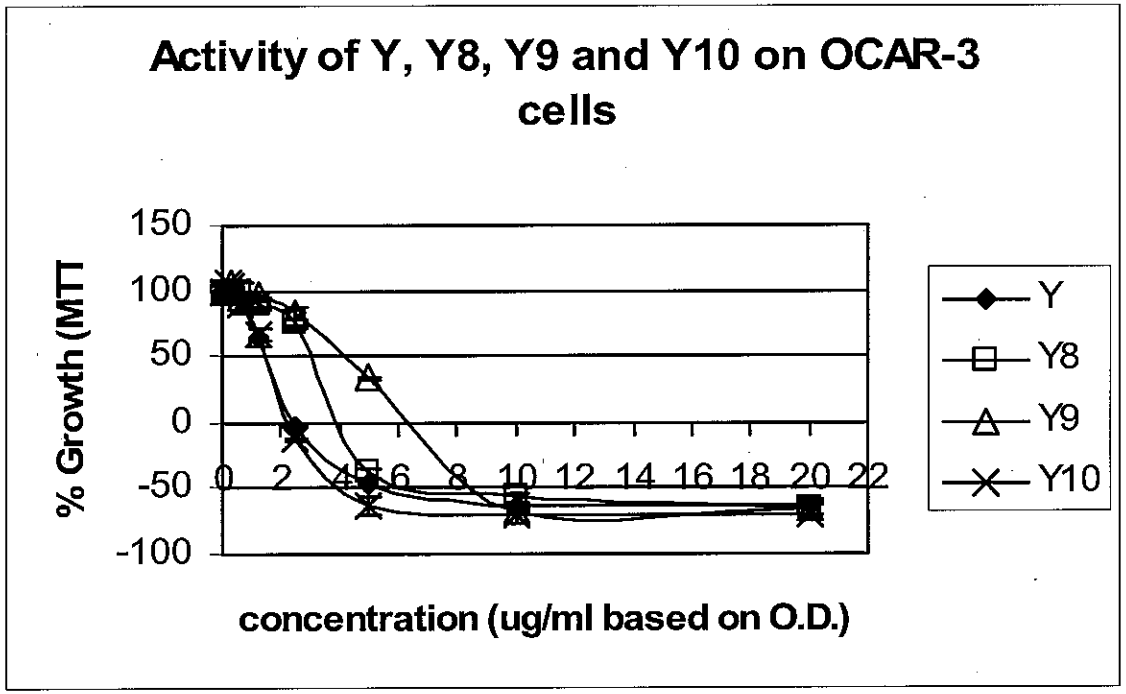


Figure 4

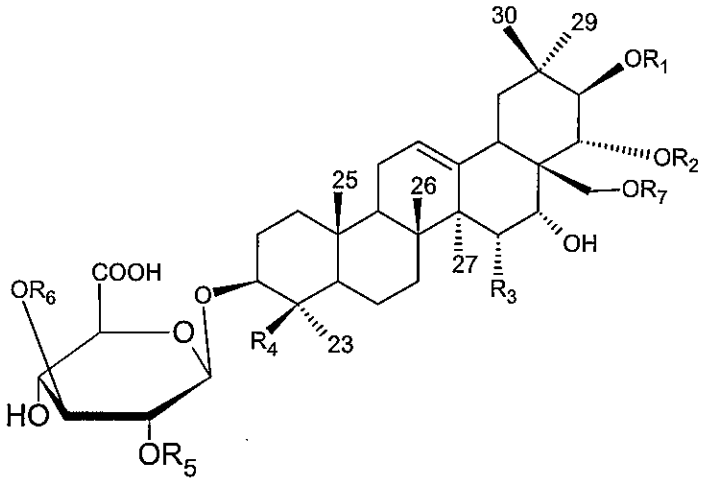
Anticancer activity of Compounds Y, Y8, Y9 and Y10.



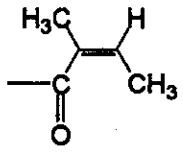
~~5/77~~ 5/66

Figure 5

Consensus structure of compound Ys
(Y,Y2, Y8 and Y10)



R1 = Angeloyl
R2 = Angeloyl
R3 = OH or H
R4 = CH3 or CH_2OH
R5 = D- glucose, D-galactose
~~R6 = L- arabinose~~
~~R7 = H,~~

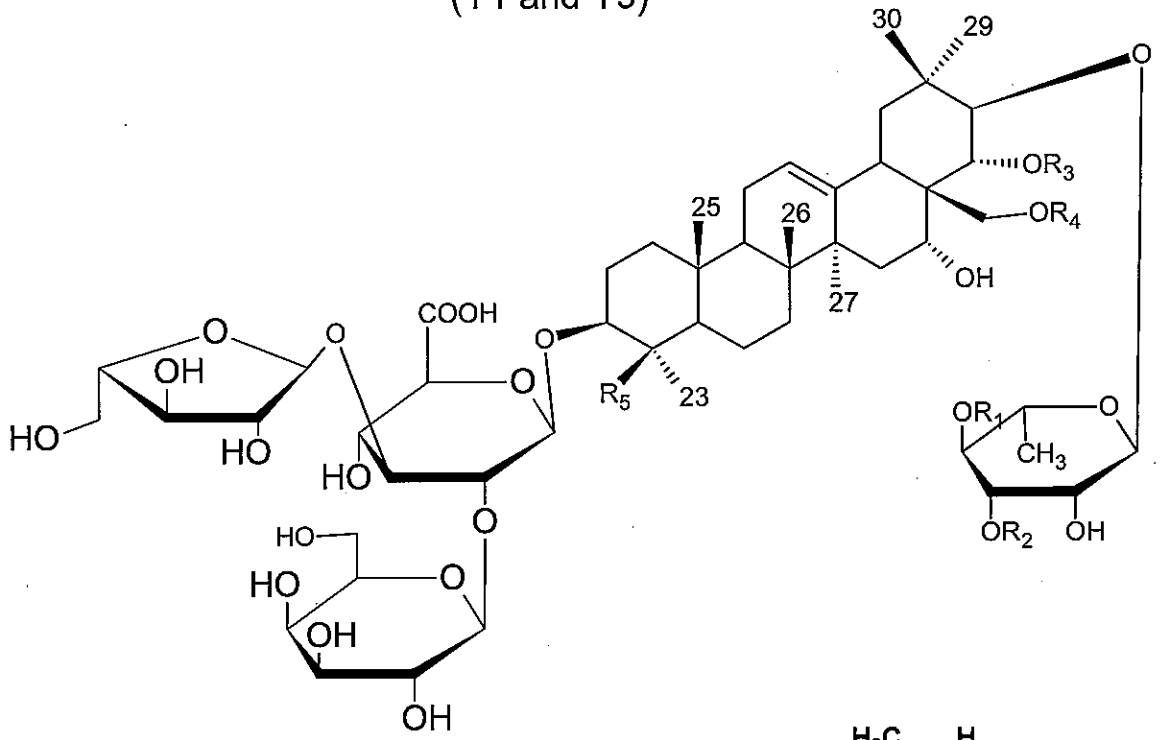
Angeloyl = 

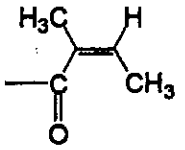
~~6/77~~ 6/66

Figure 6

Consensus structure of Ys

(Y1 and Y9)



Angeloyl = 

R1 = Angeloyl

R2 = Angeloyl

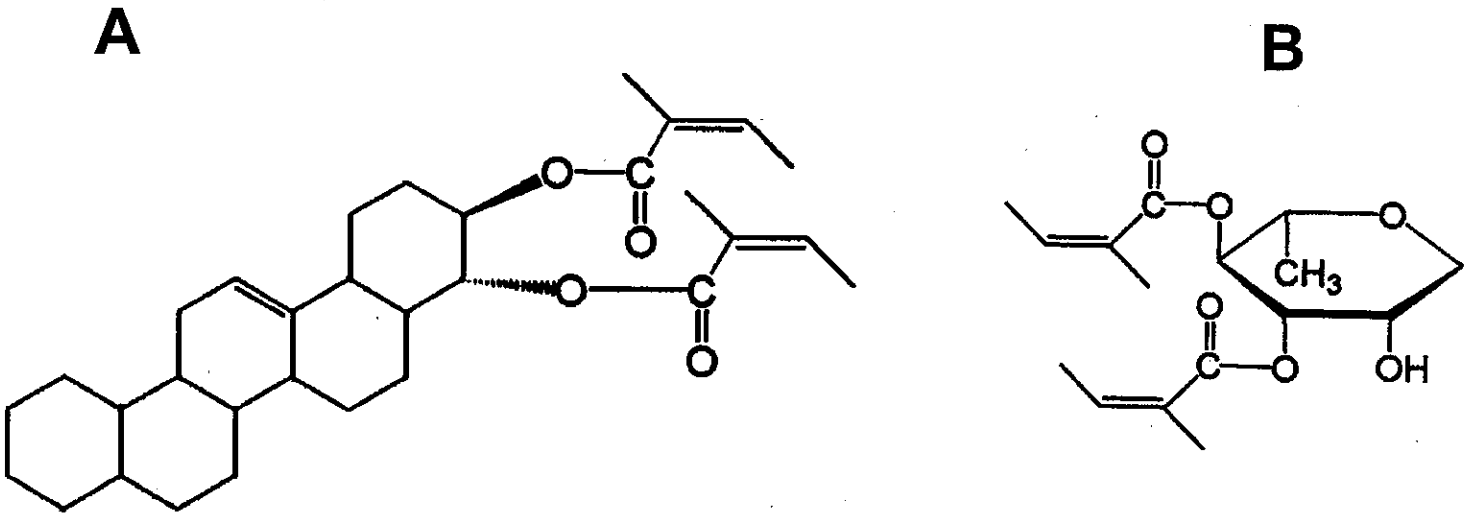
R3 = Ac or H

~~R4 = Ac or H~~

~~R5 = CH3 or CH2OH~~

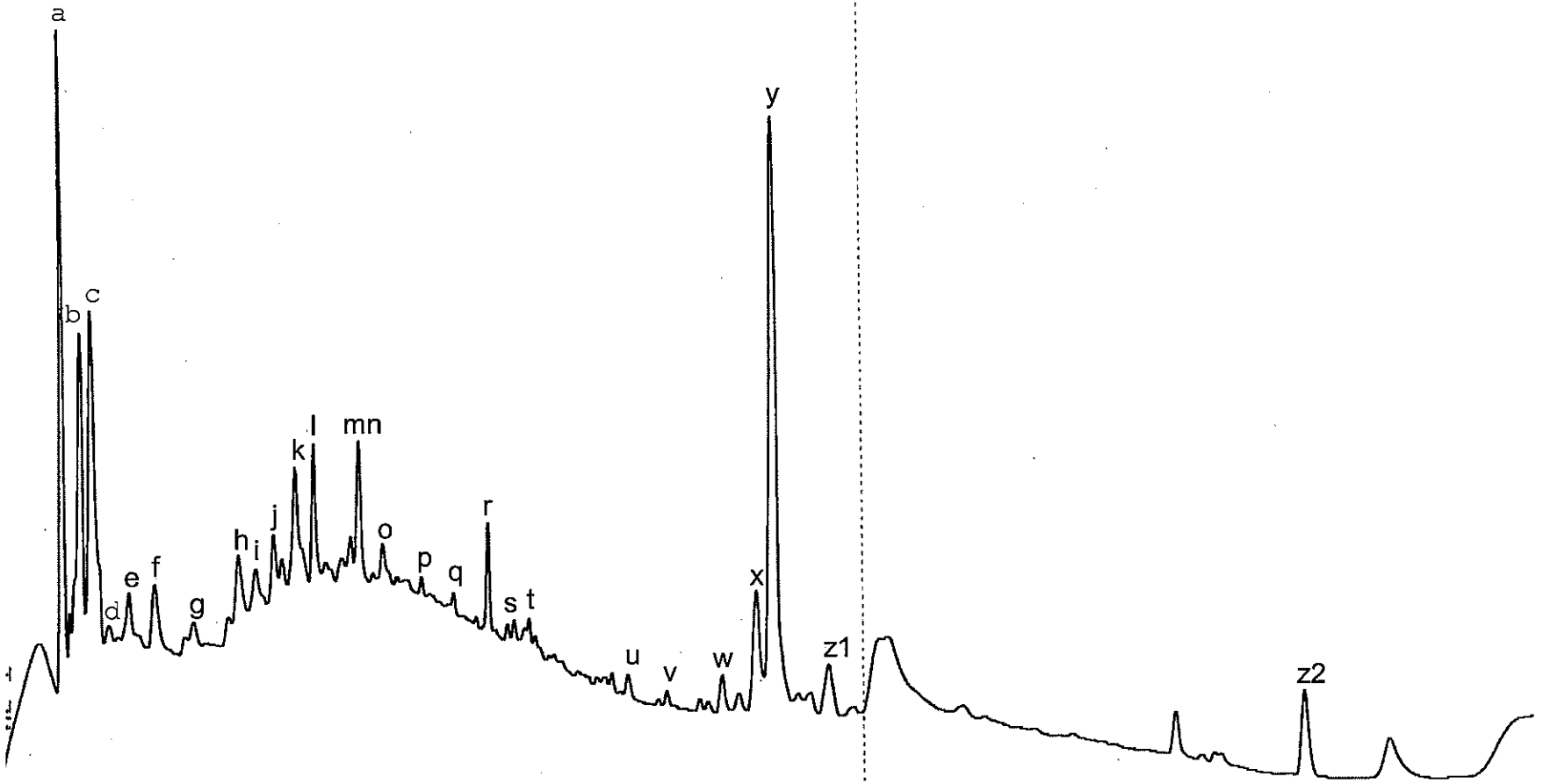
Figure 7

Biangeloyl group situated in trans-position on a structure.



~~8/77~~ 8/66

Figure 8



~~9/77~~ 9/66

Figure 9

FPLC profile of Plant extract

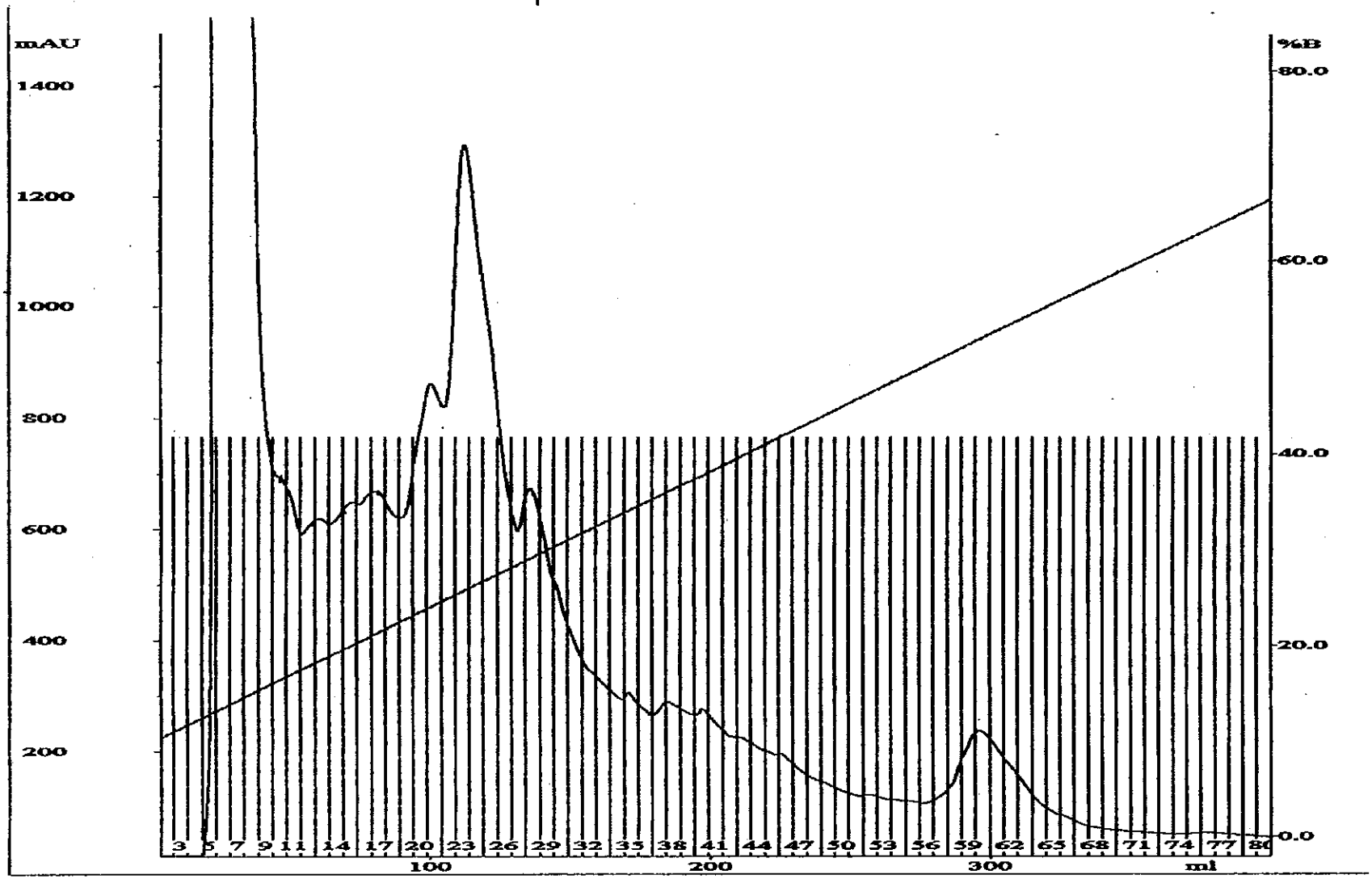


Figure 10

Cell growth activity of fractions of plant extract

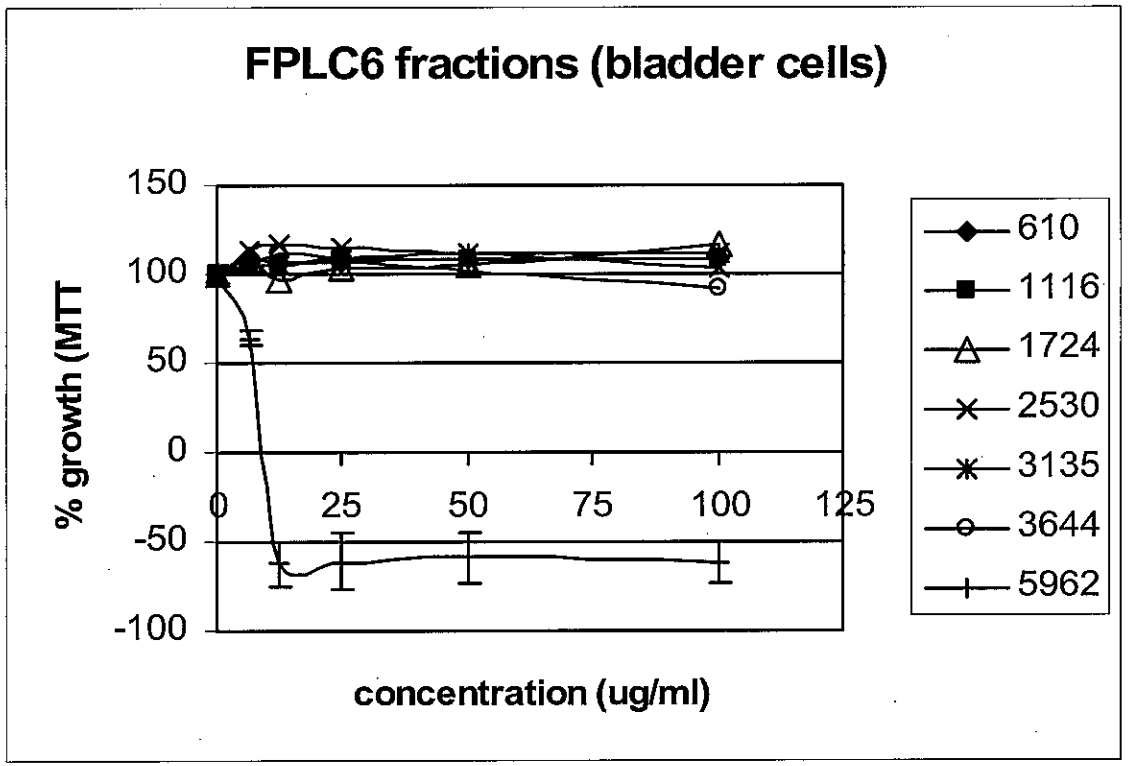
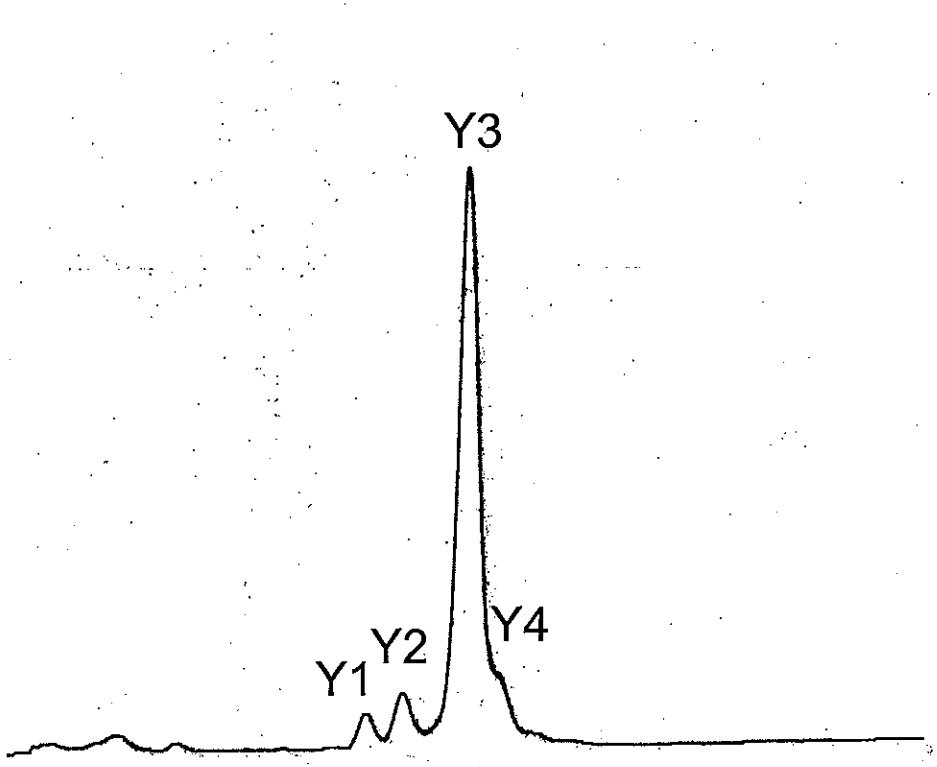
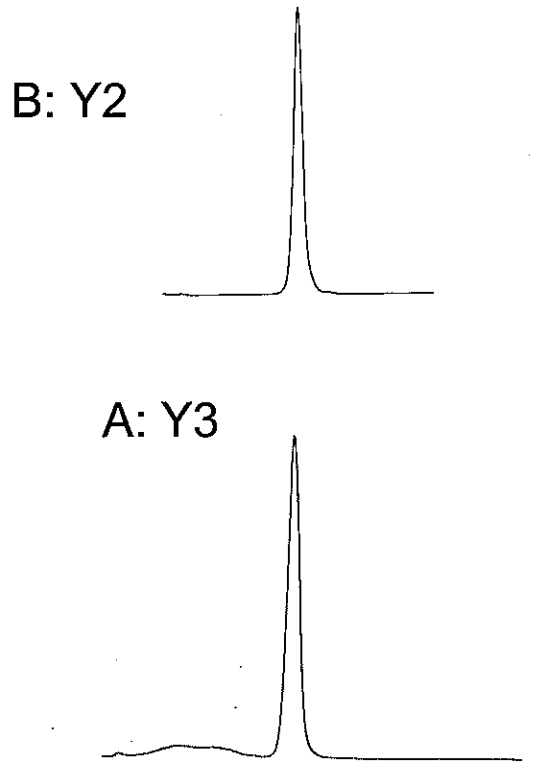


Figure 11
Purification of Factions Ys

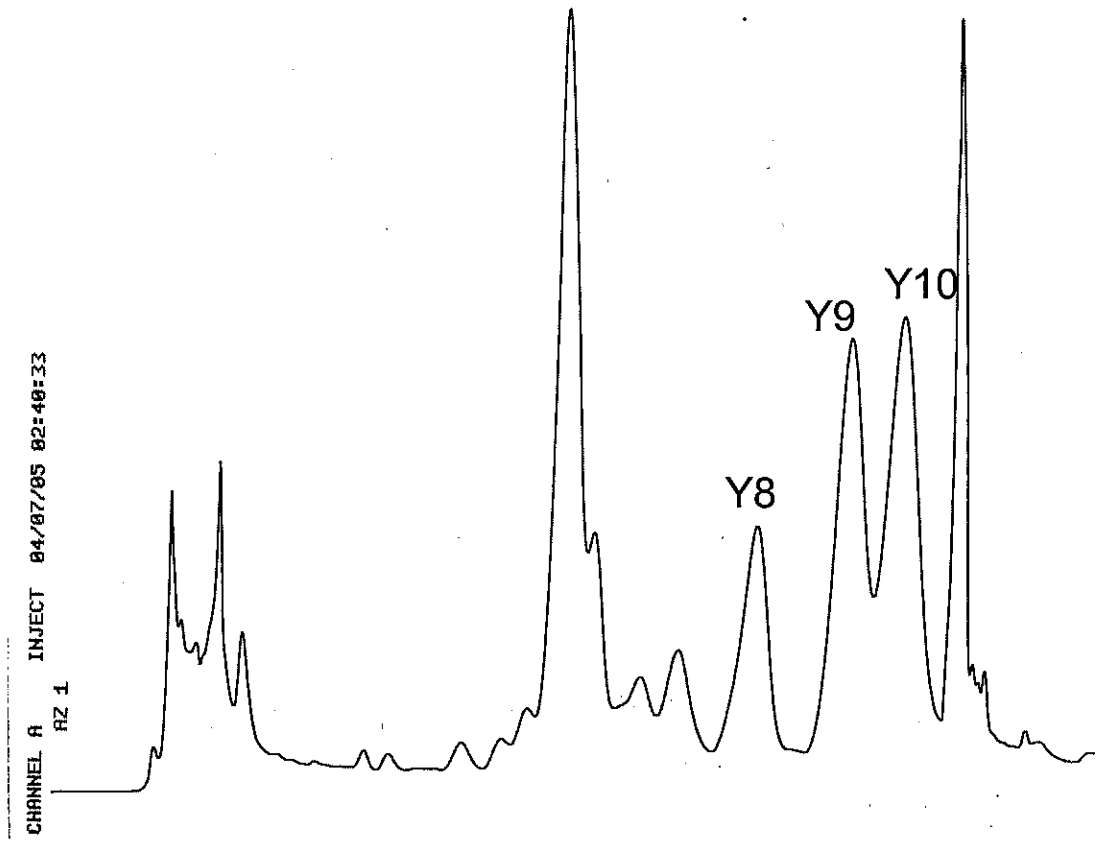




~~12/77~~ 12/66

Figure 12

Purification of Y8-Y10 with HPLC (iso-45)



~~13/77~~ 13/66

Figure 13

Purity of Y8, Y9 and Y10

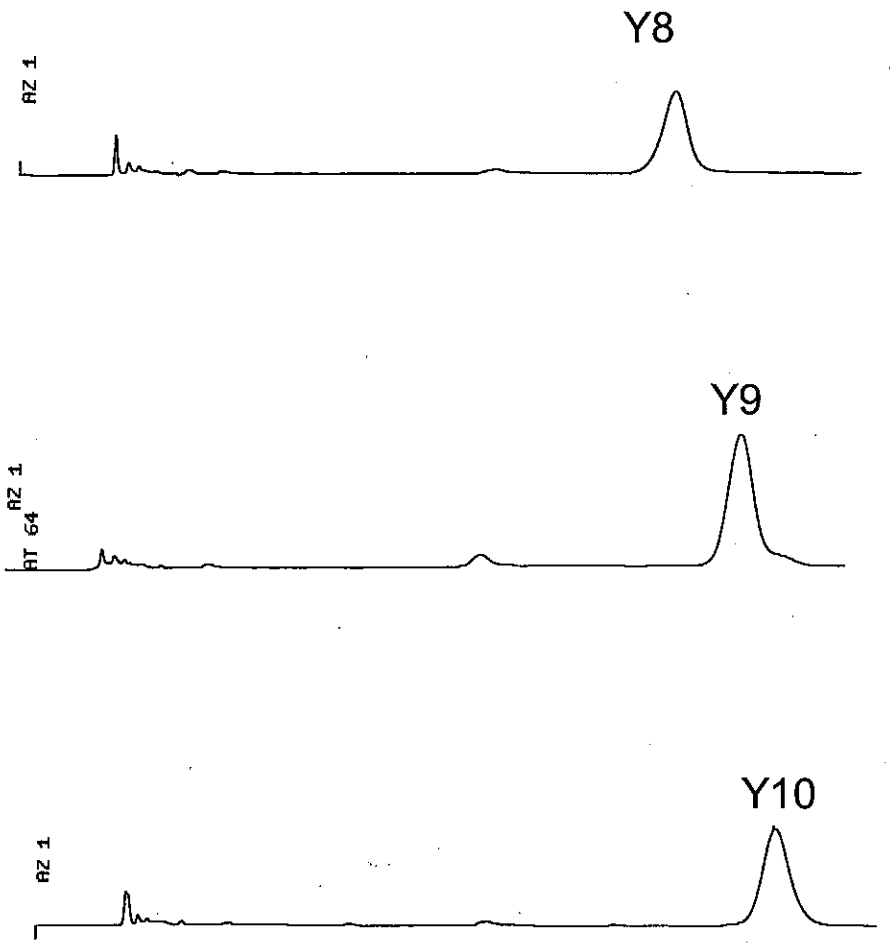
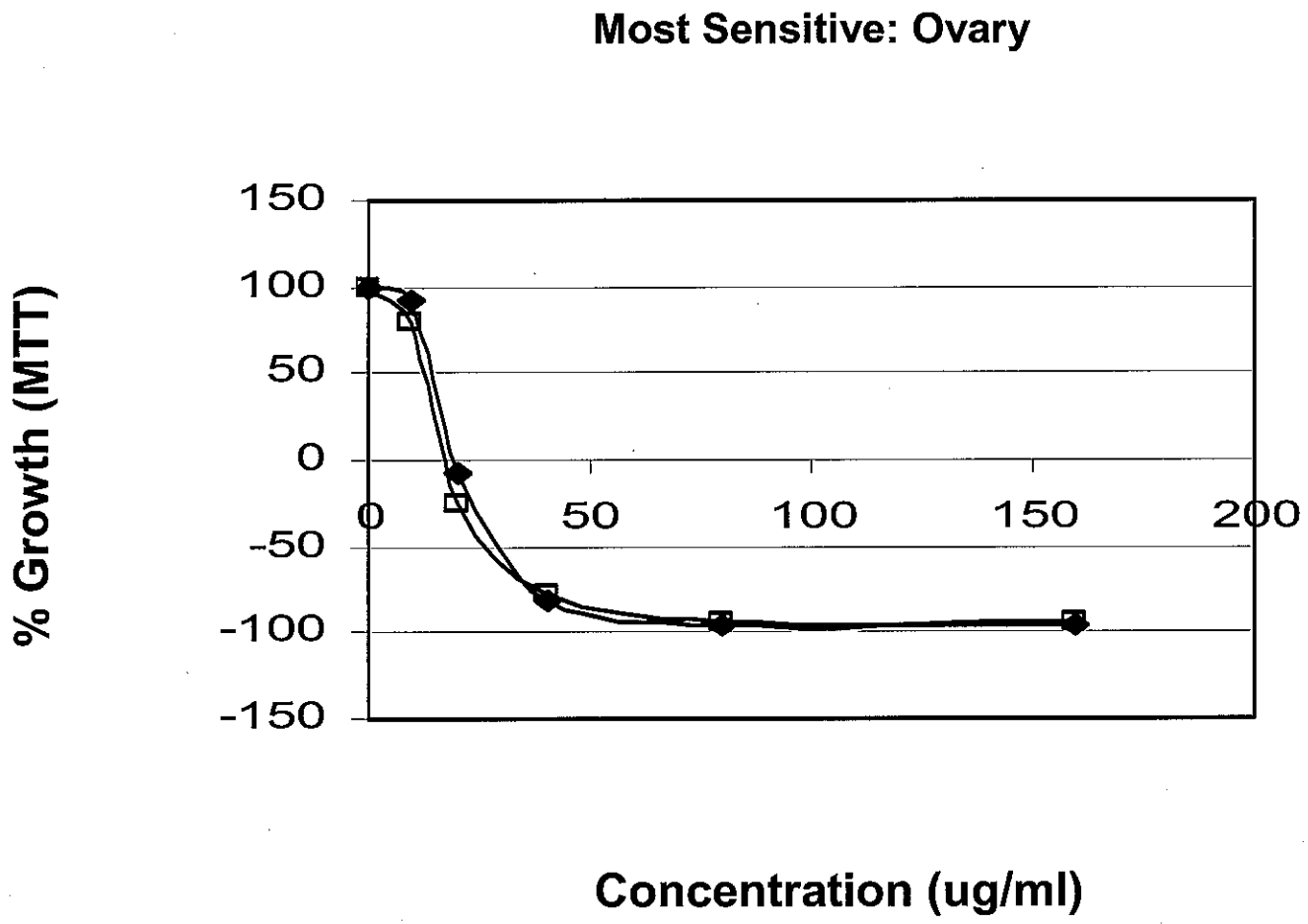


Figure 14

Inhibition of Ovarian cancer growth by plant extract



~~15/77~~ 15/66

Figure 15

Comparison of Potency of Y in ovary and cervix cancer cells

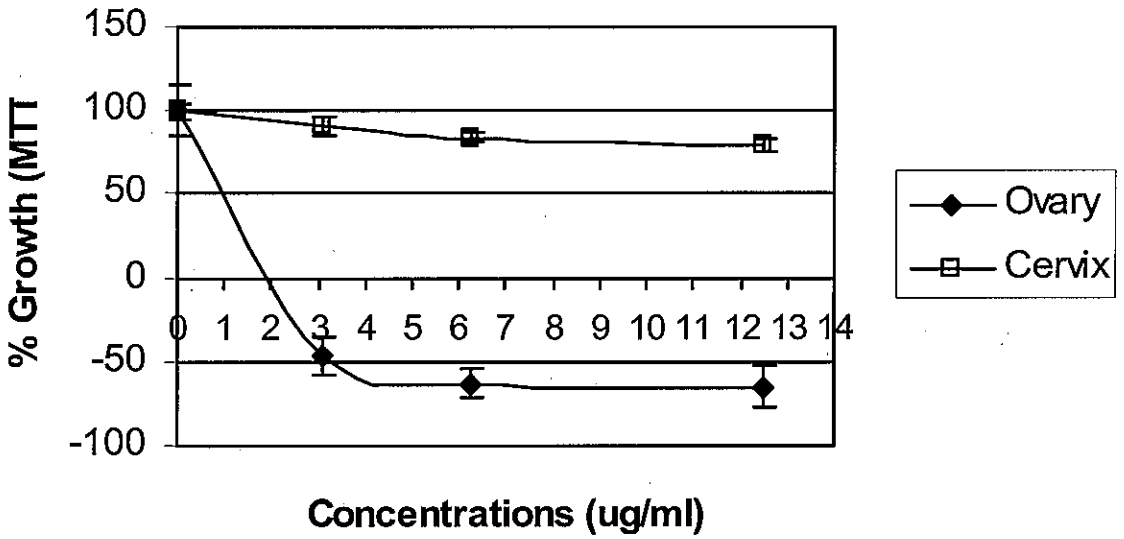
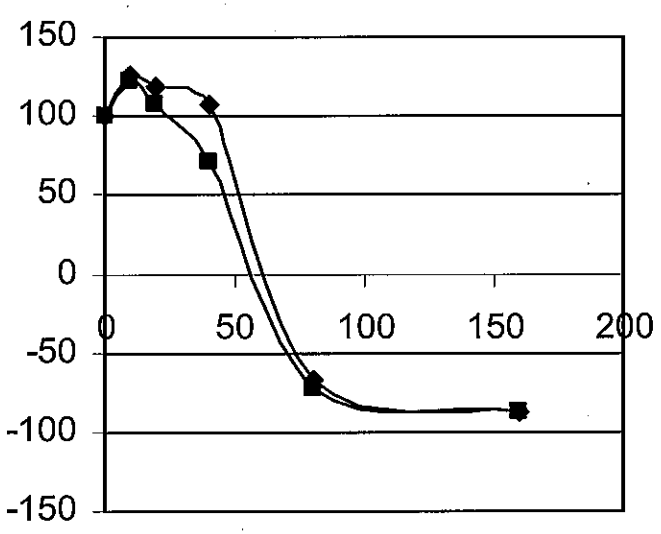


Figure 16A

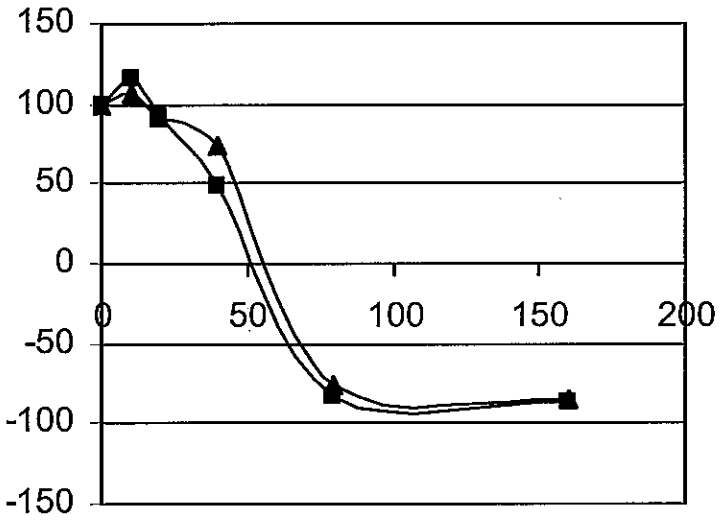
Growth curve of cancer cells after treatment with extract

Sensitive

Bladder



Bone



~~17/77~~ 17/66

Figure 16B

Growth curve of cancer cells after treatment with extract

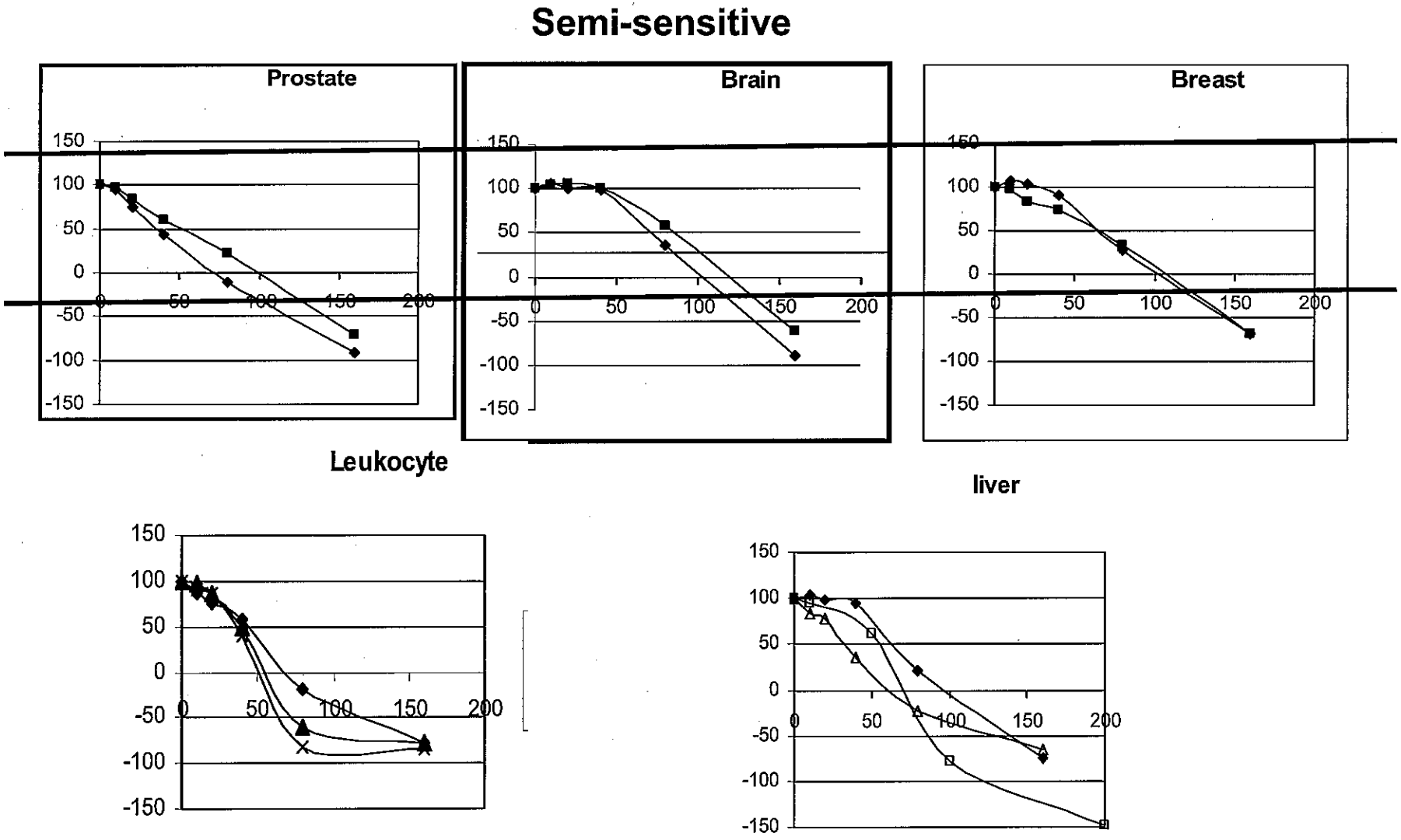
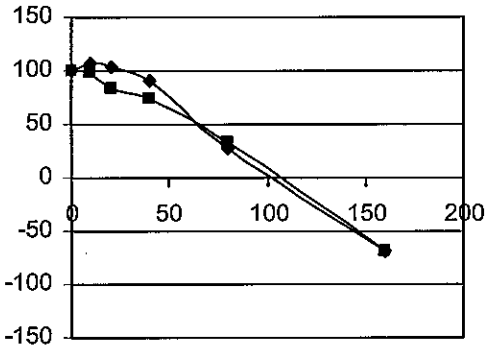


Figure 16C

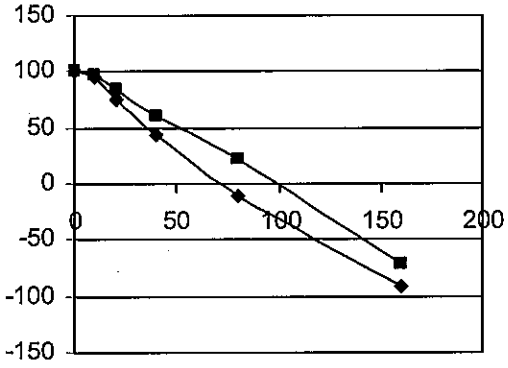
Growth curve of cancer cells after treatment with extract

Semi-sensitive

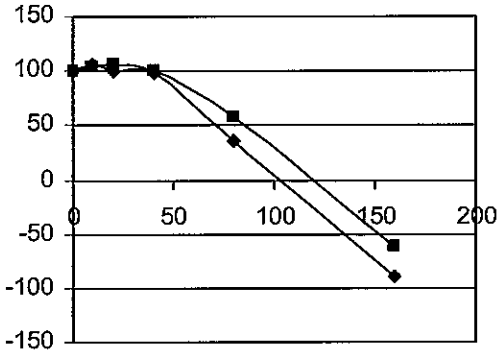
Breast



Prostate



Brain



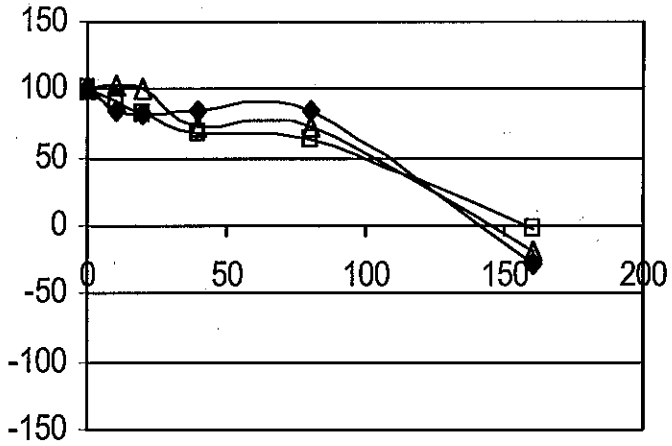
~~18/77~~ 19/66

Figure 16~~C~~ D

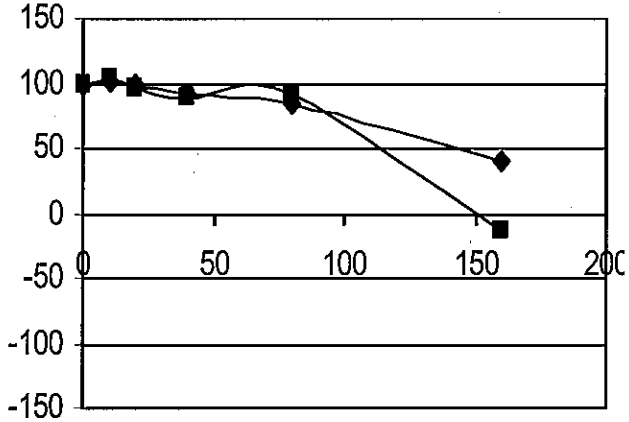
Growth curve of cancer cells after treatment with extract

Least sensitive

Colon



Cervix



Lung

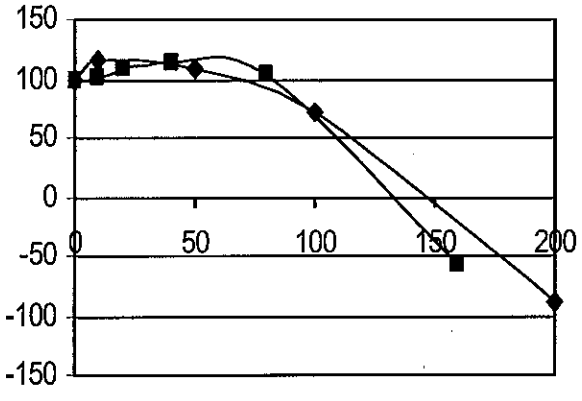
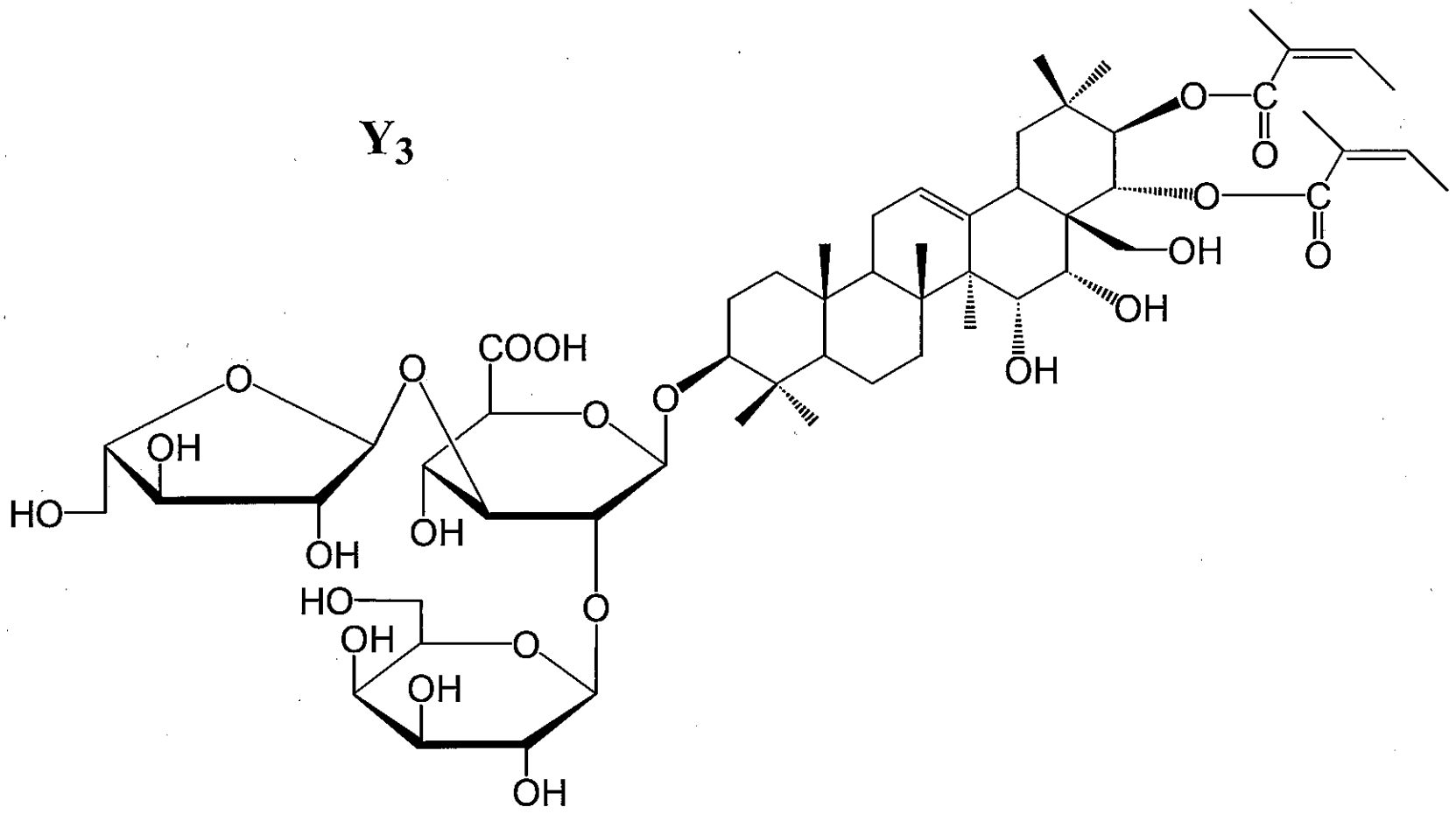


Figure 17

Chemical Structure of compound Y (Y3)



~~20/77~~ 21/66

Figure 18

Proton NMR of Y

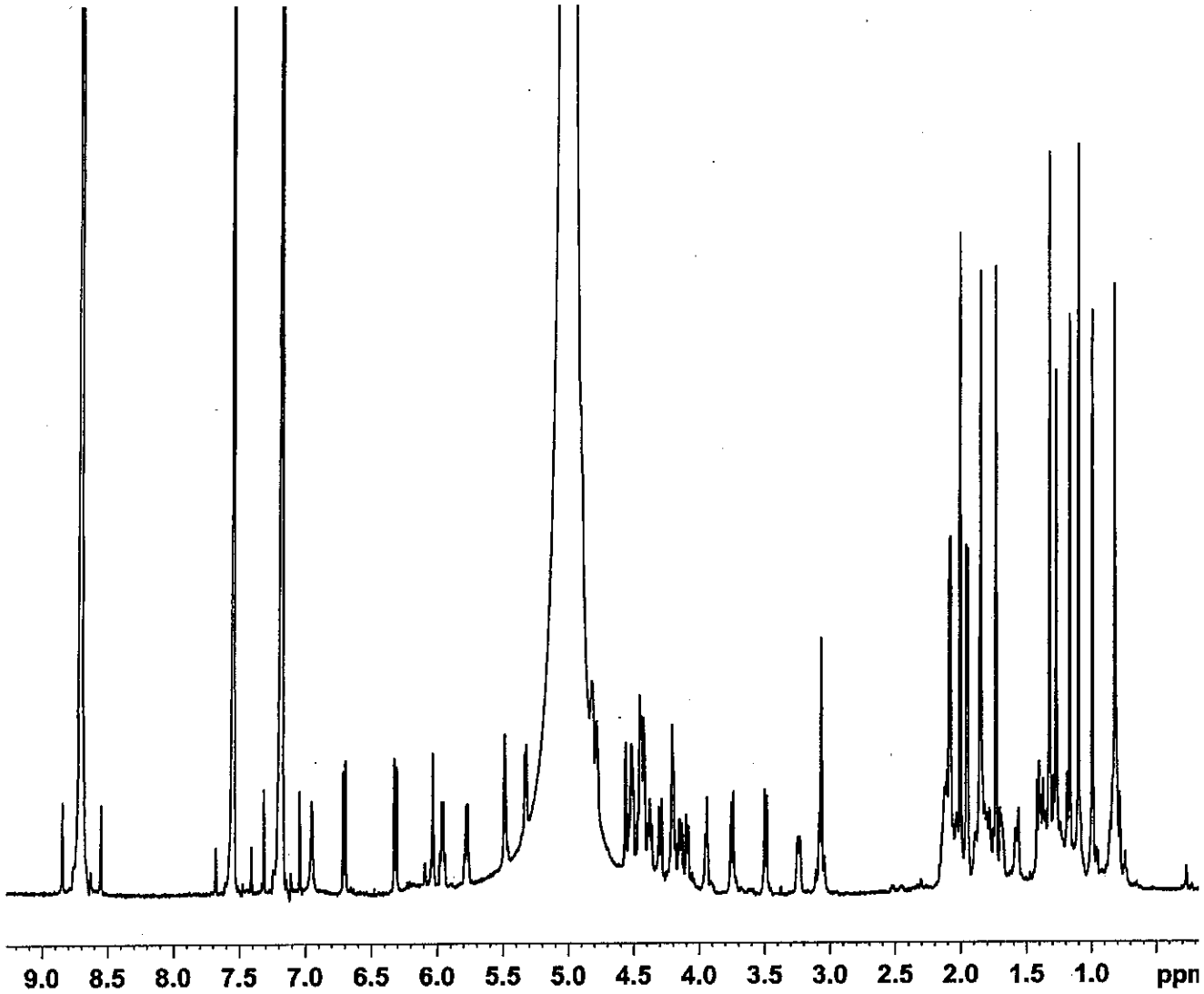


Figure 19

2D NMR (HMQC) of Y

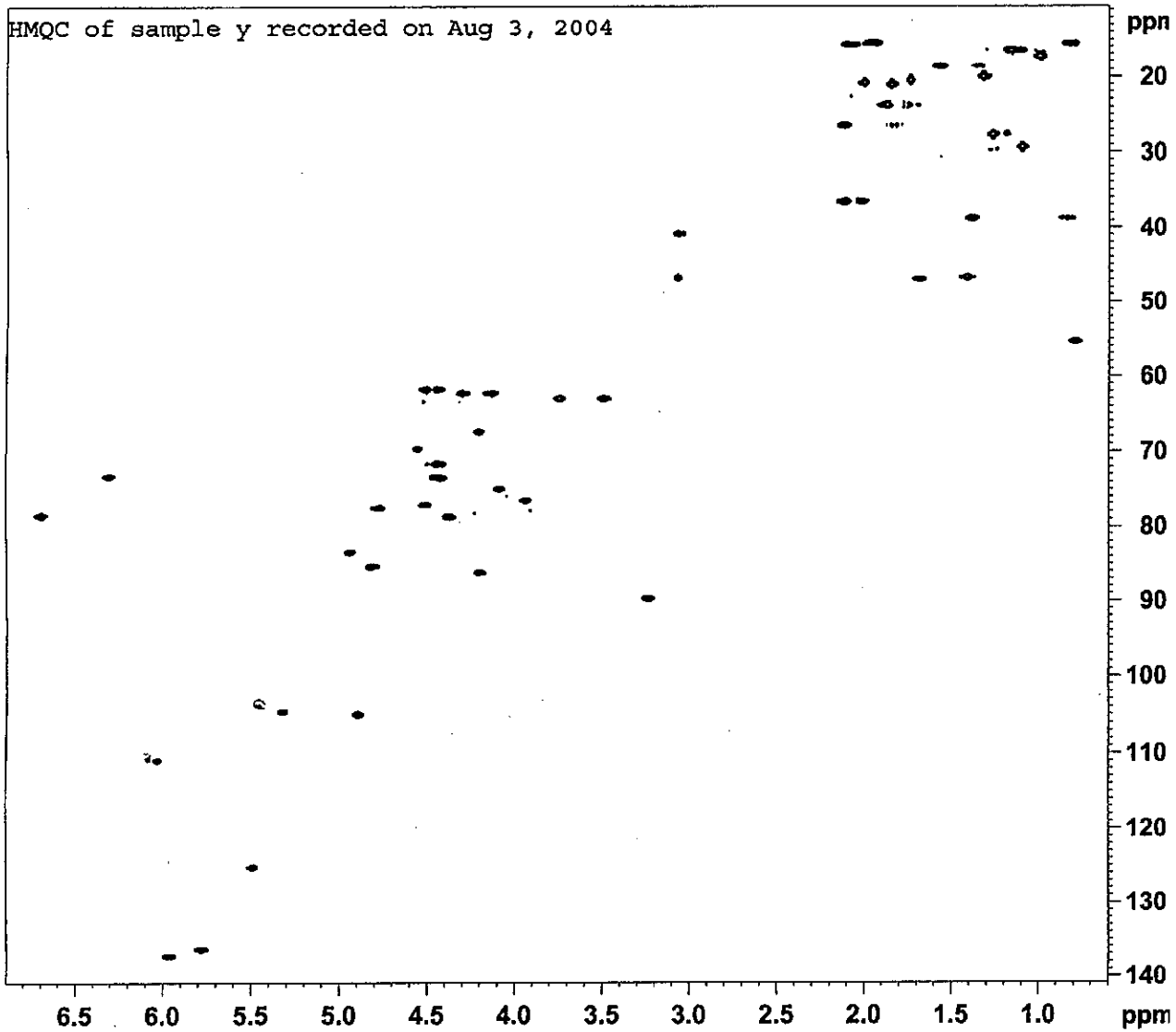


Figure 20
HMBC of Y

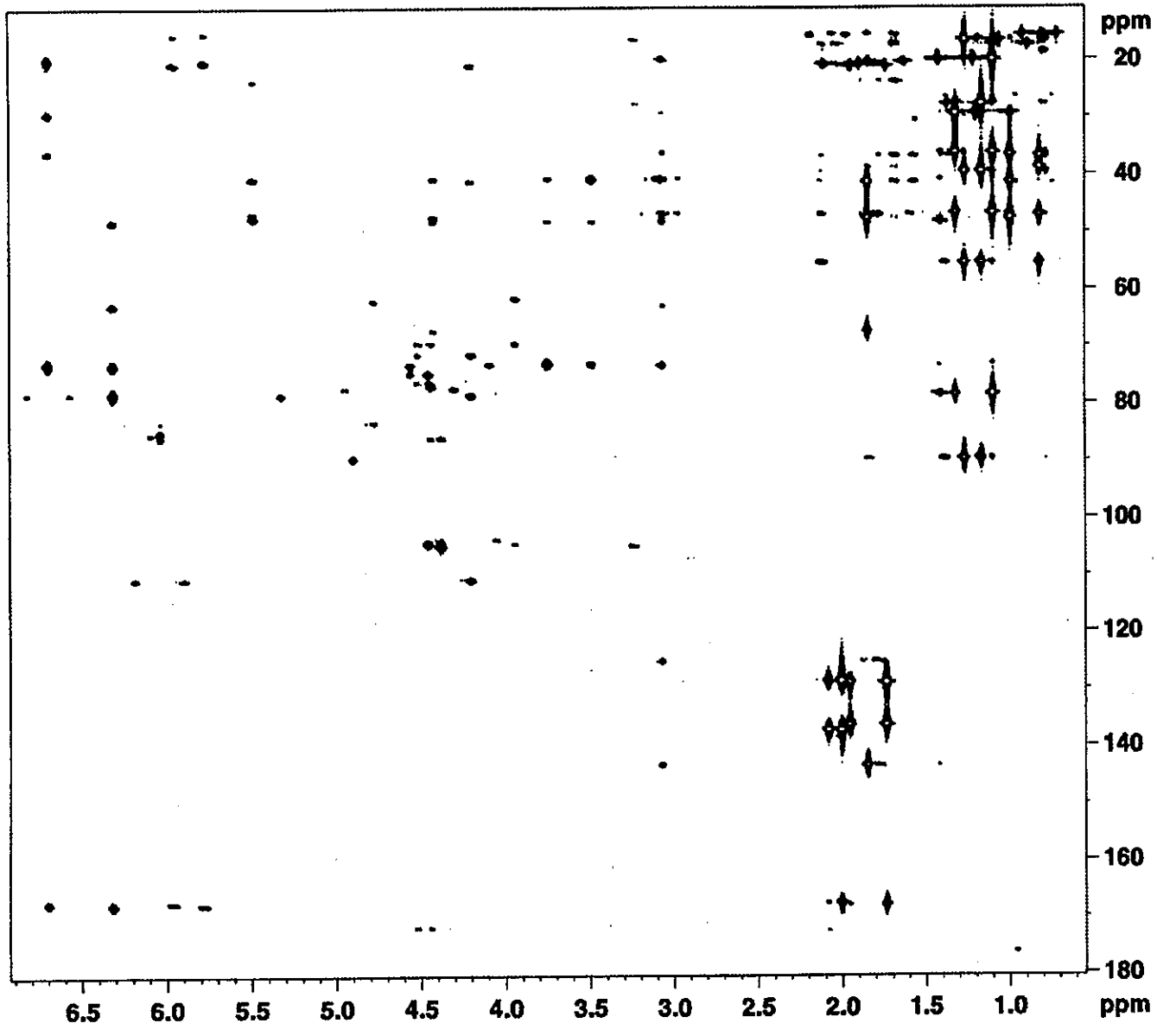
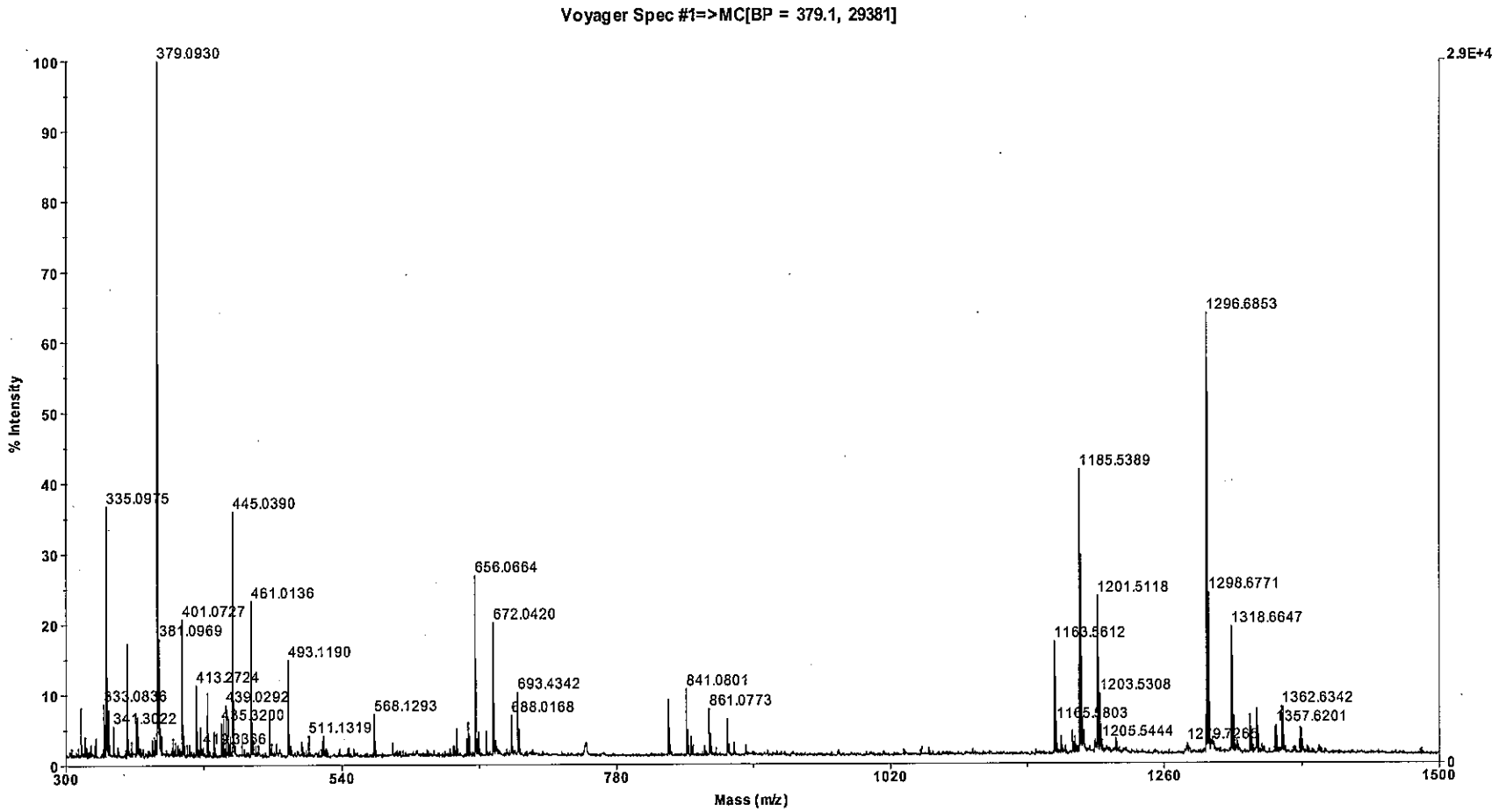


Figure 21

MS (MALDI-TOF) of Y

Y + Matrix (CHCA) + Angiotensin 1 "two point Calibration"



~~24/77~~ 25/66

Figure 22
MS-ESI of Y

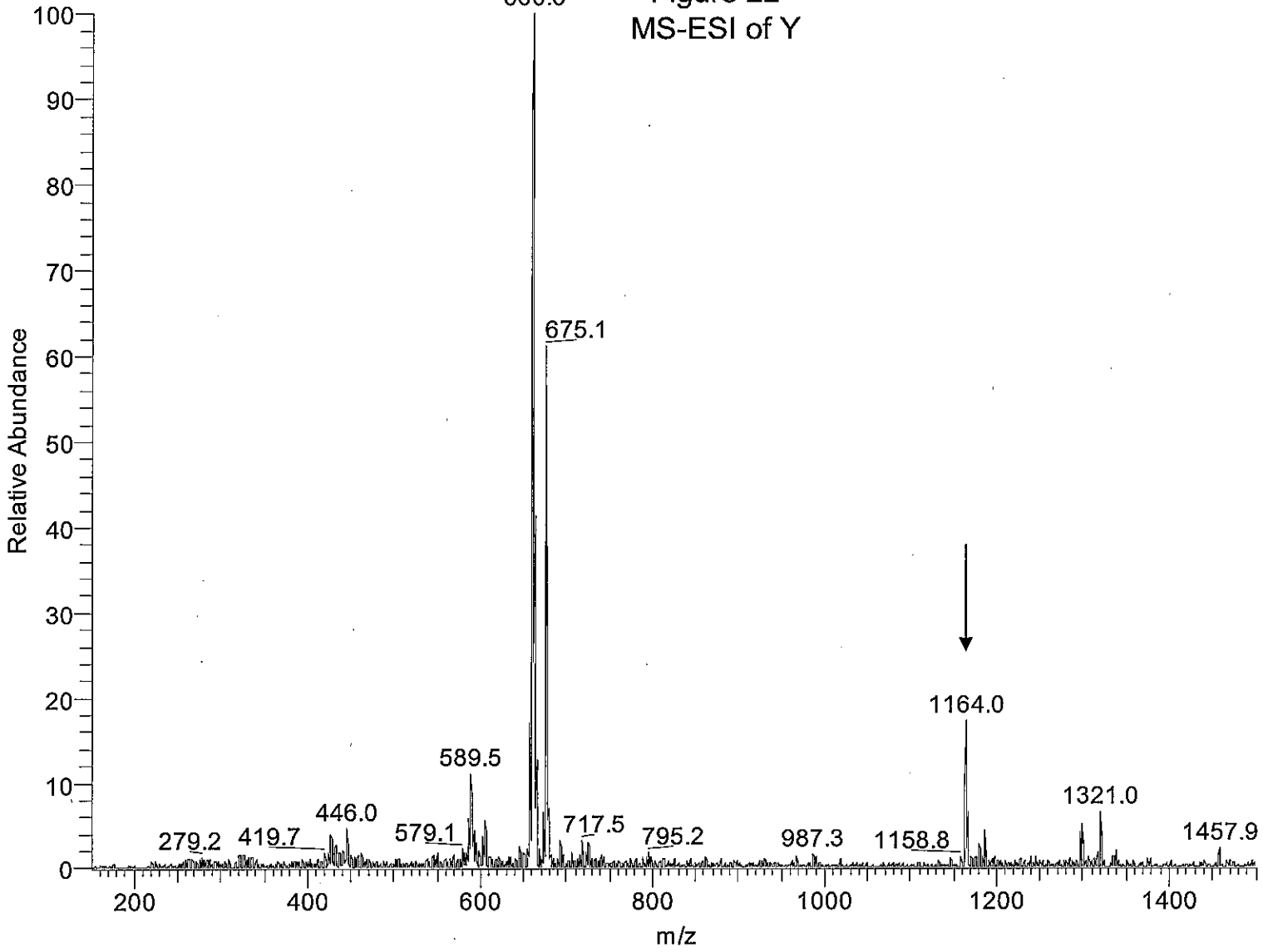
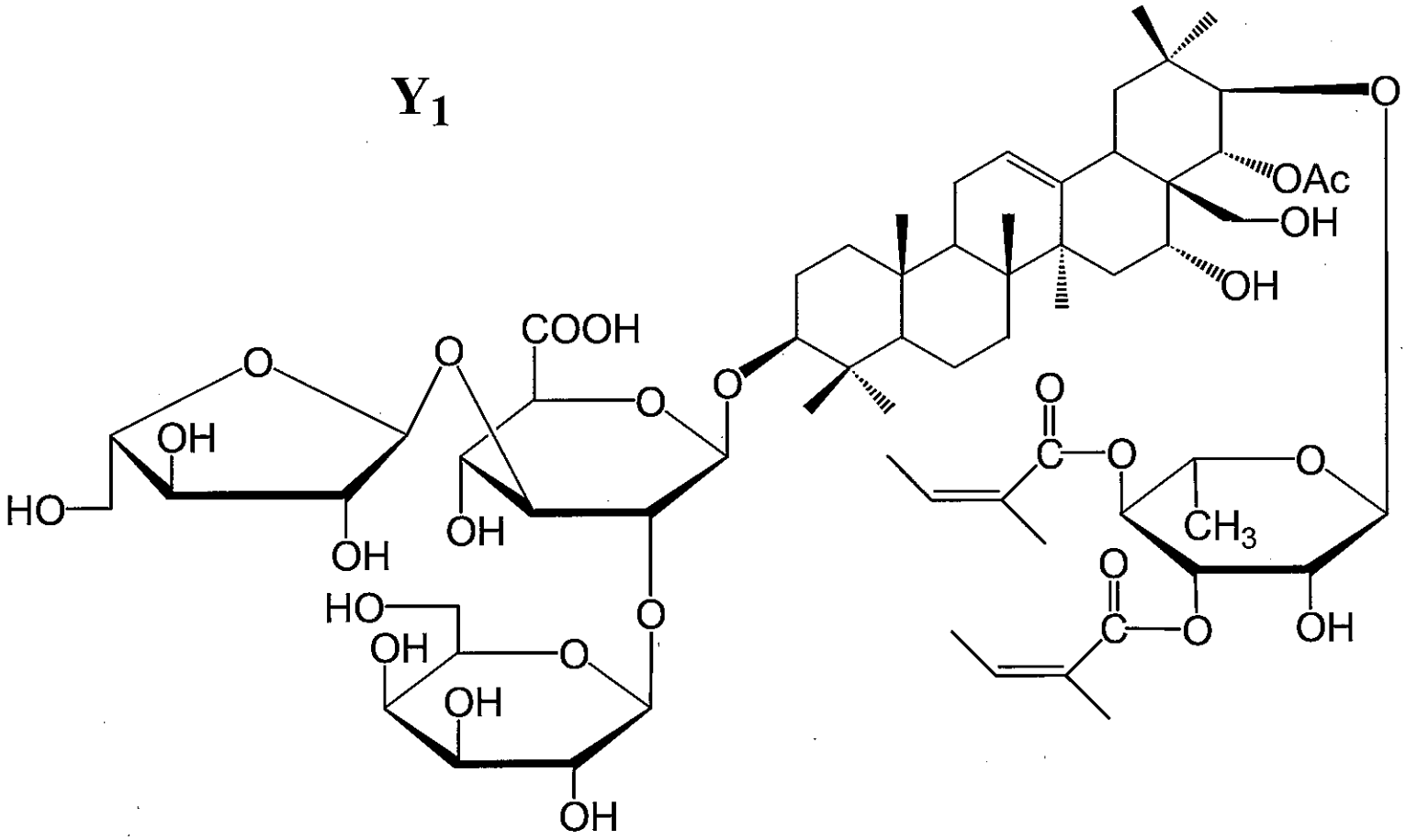


Figure 23
Chemical Structure of Y1



~~26/76~~ 27/66

Figure 24

Proton NMR of Y1

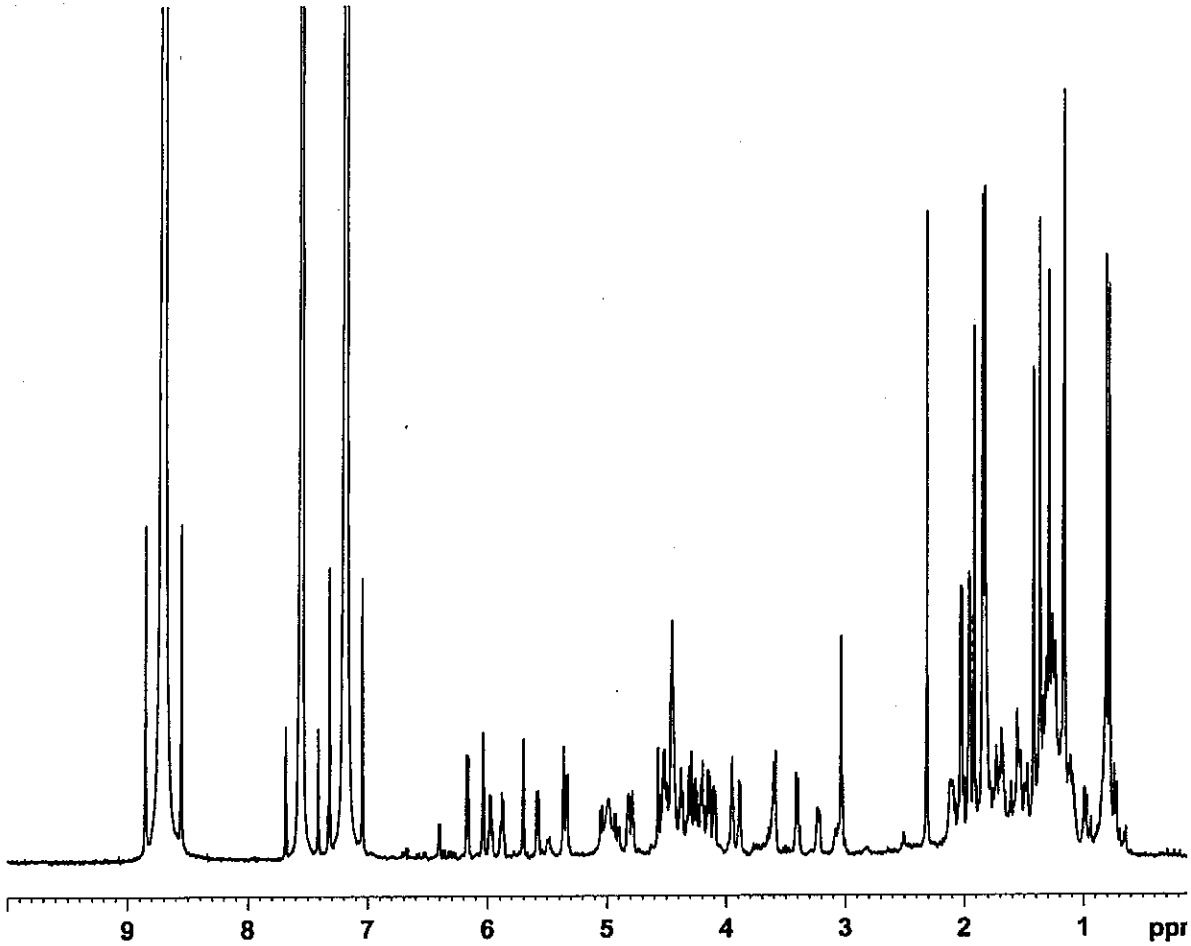
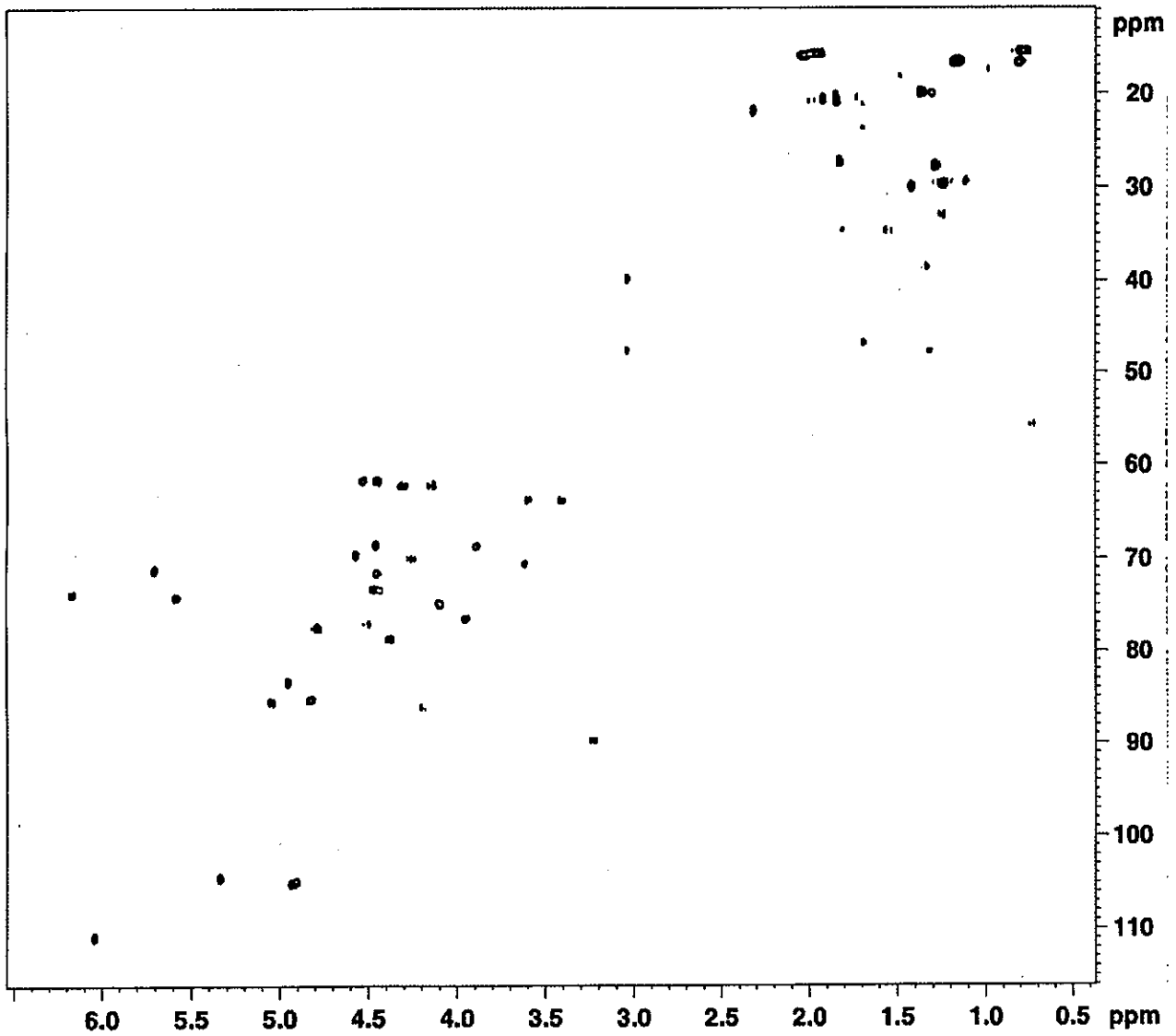


Figure 25
HMQC of Y1



~~28/77~~ 29/66

Figure 26

HMBC of Y1

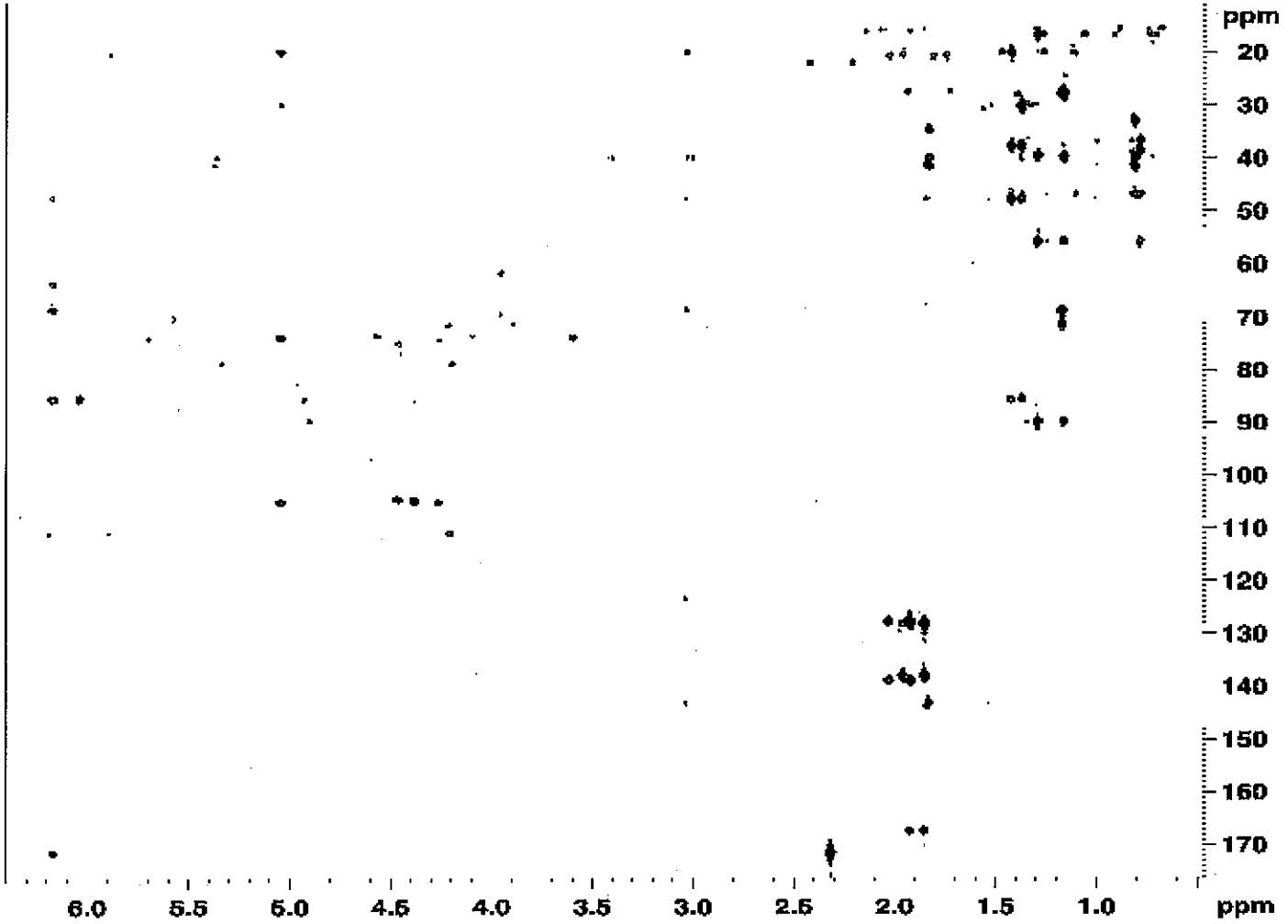


Figure 27
2D NMR (COSY) of Y1

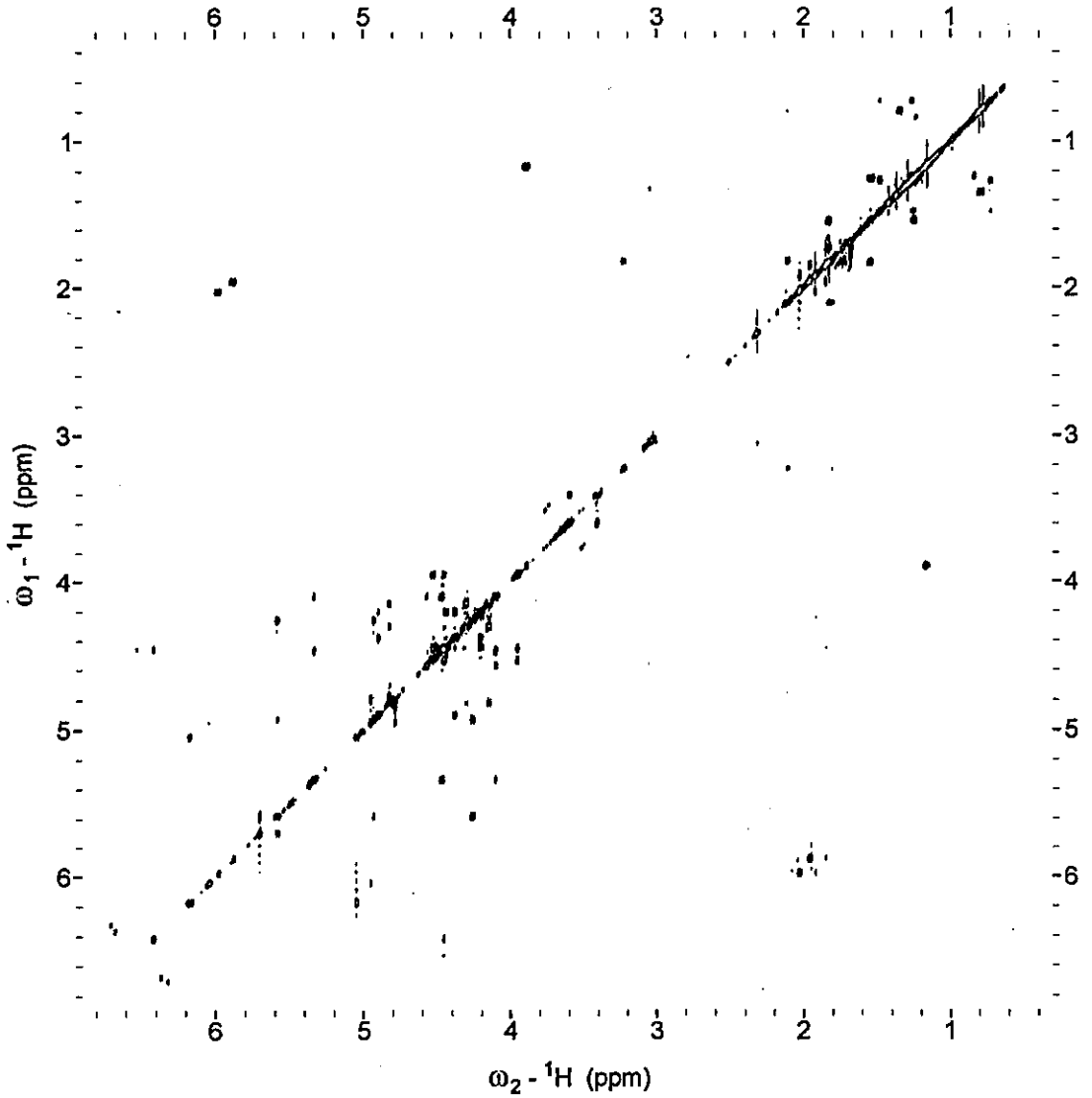


Figure 28. Chemical Structure of Y2

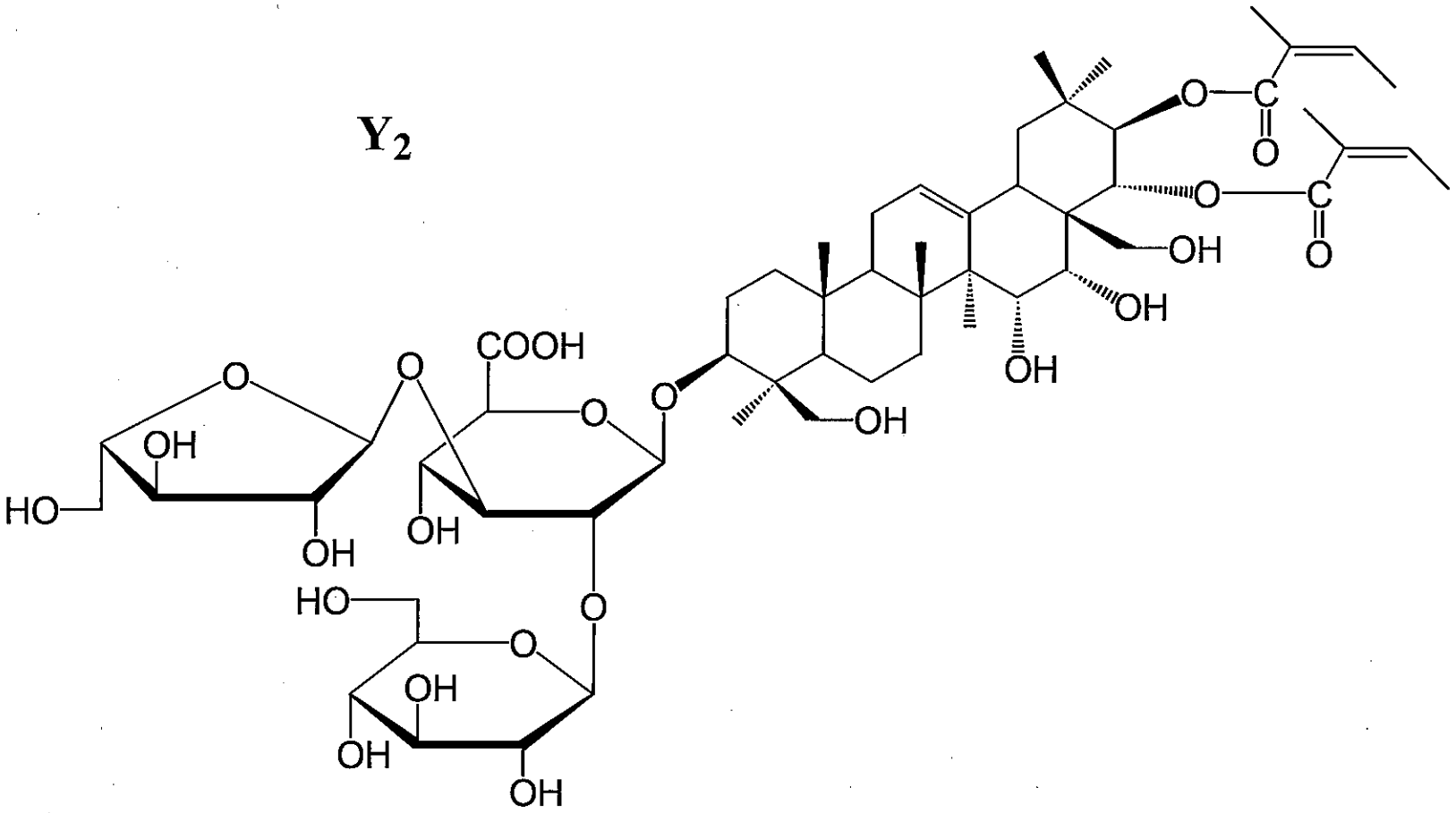


Figure 29
H-NMR Spectrum of Y2

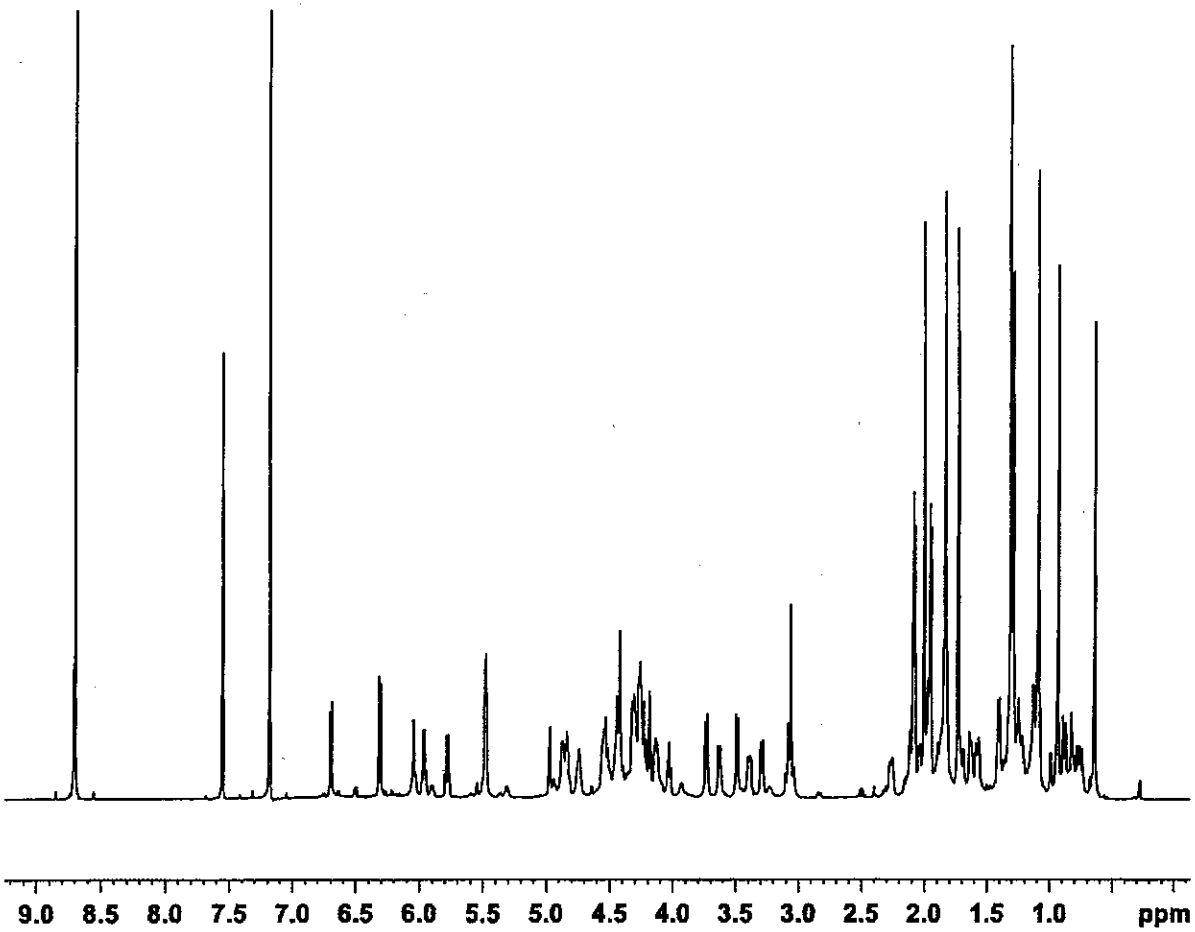


Figure 30
Y2 HMQC-level-1

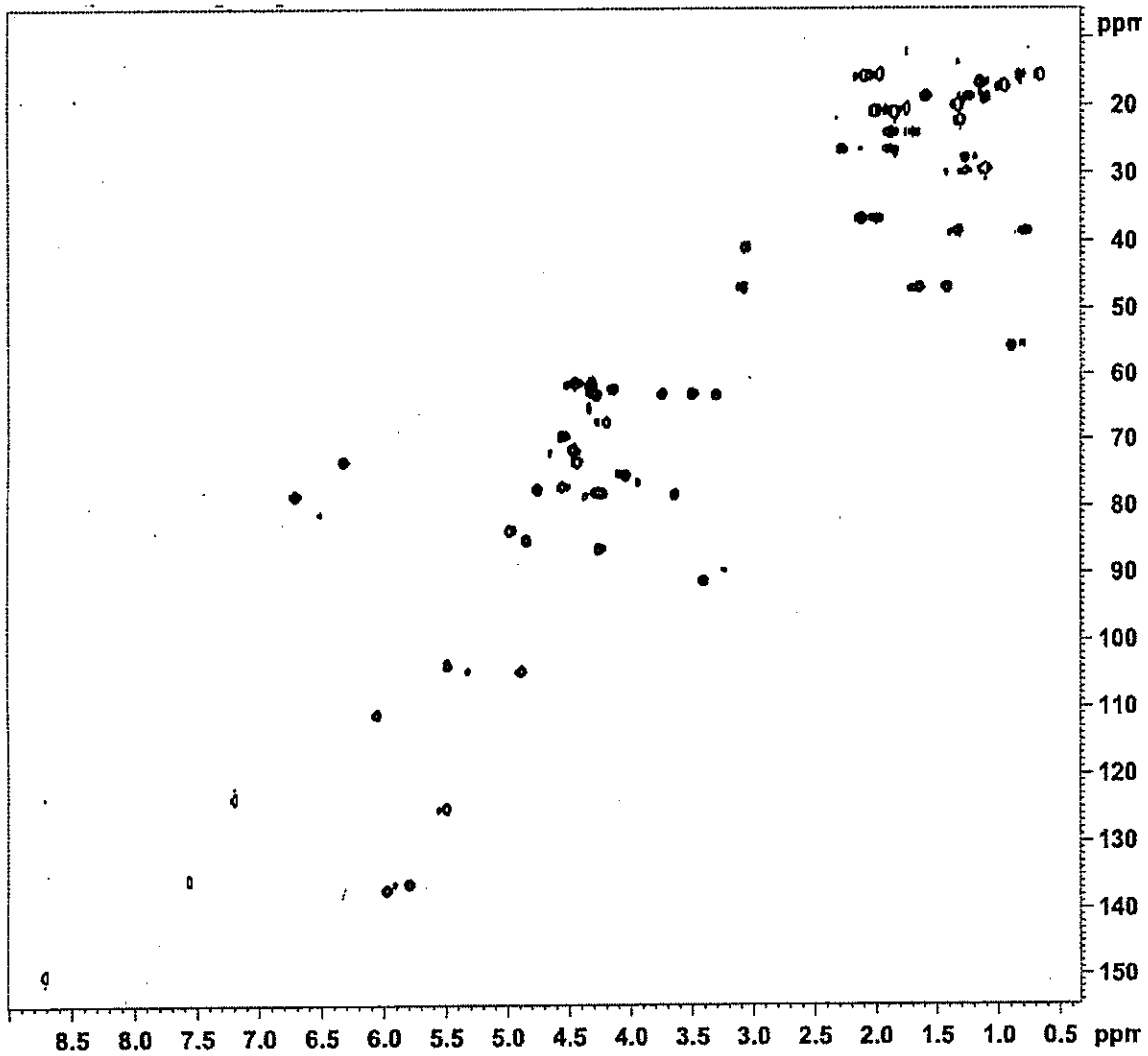


Figure 31

Y2 C13 H-NMR spectrum

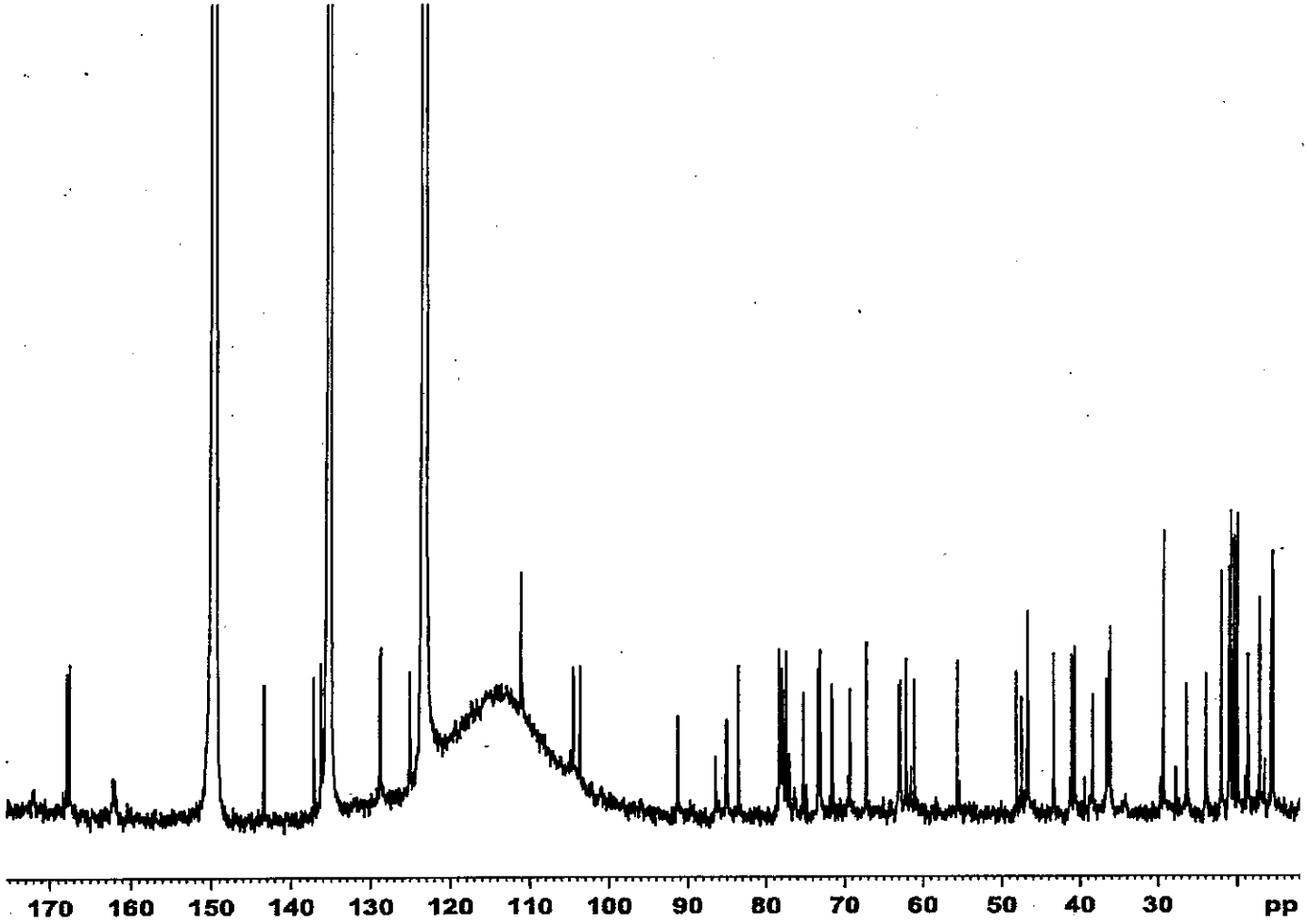


Figure 32
Y2-HMBC-level 1

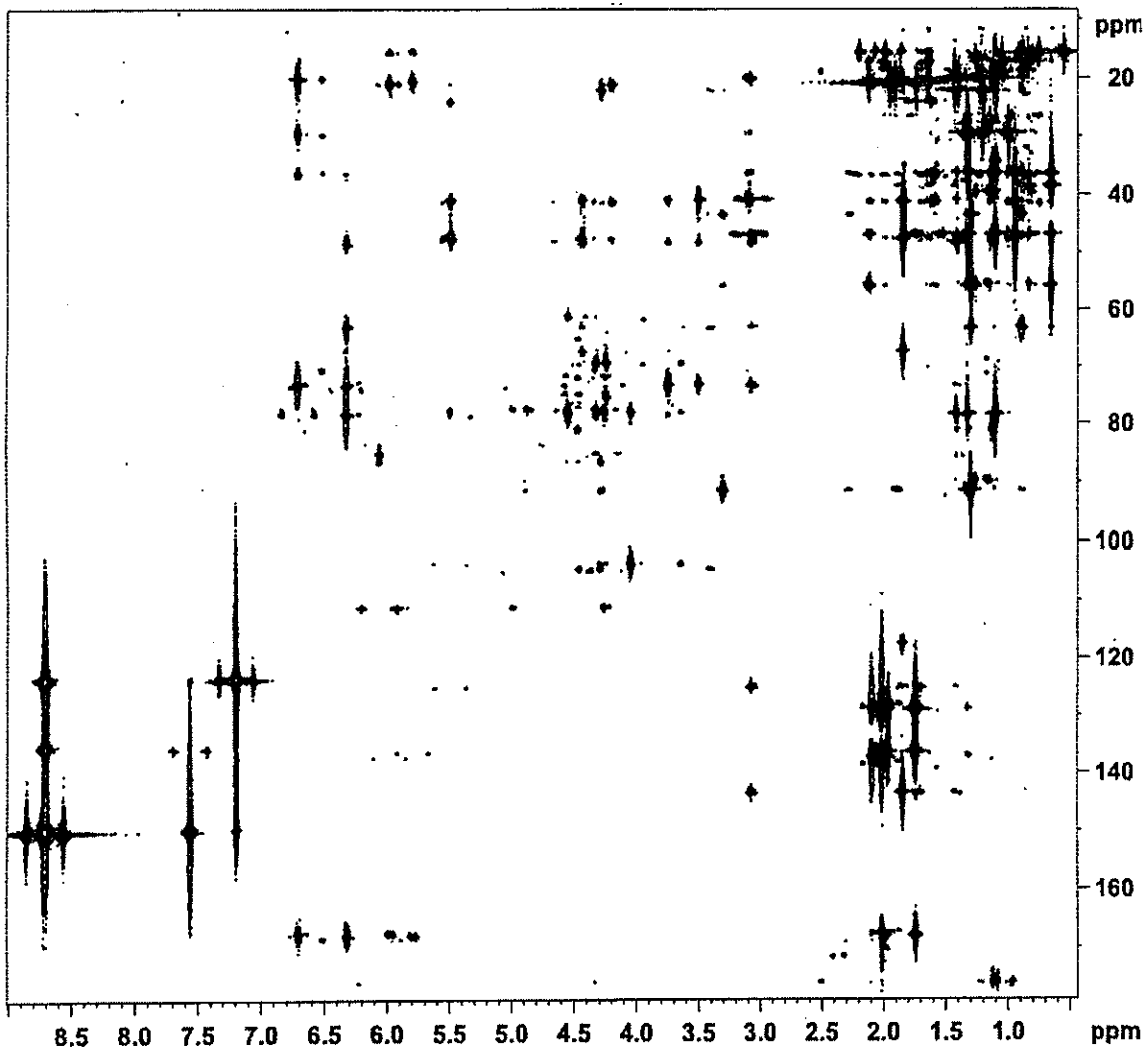


Figure 33

Y2-HOHAHA-level-1

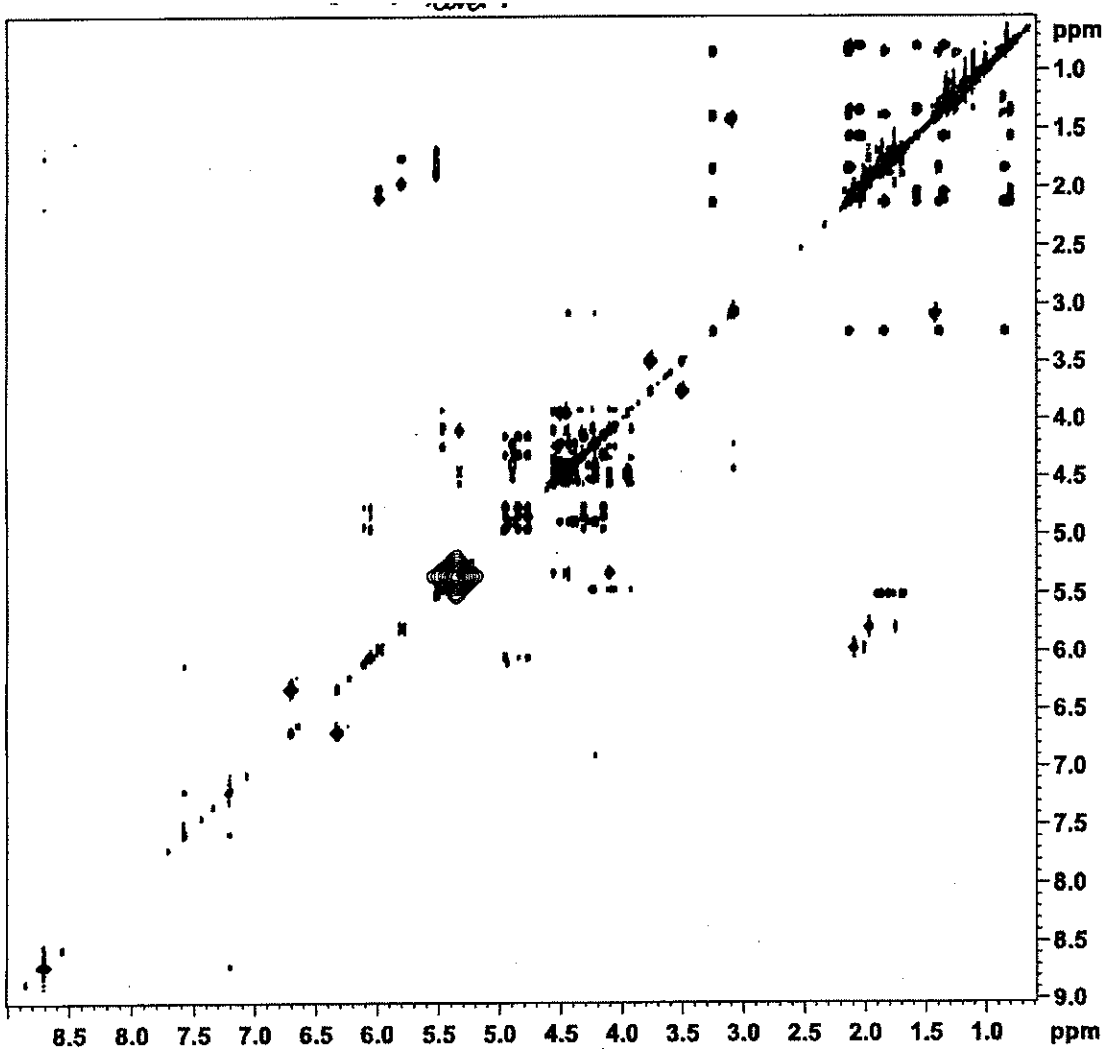


Figure 34

Mass spectrum of Y2 + Matrix + Standards

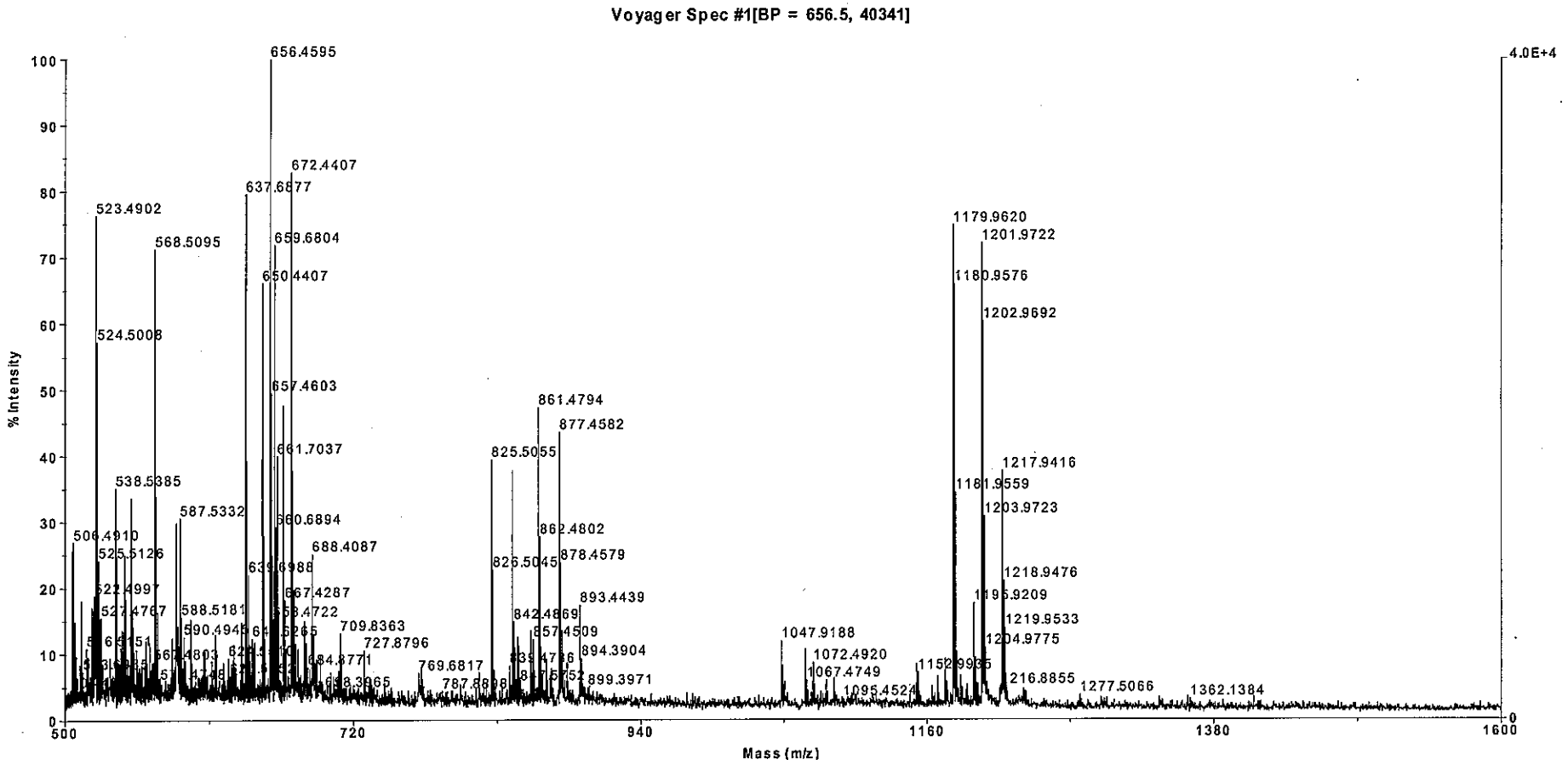
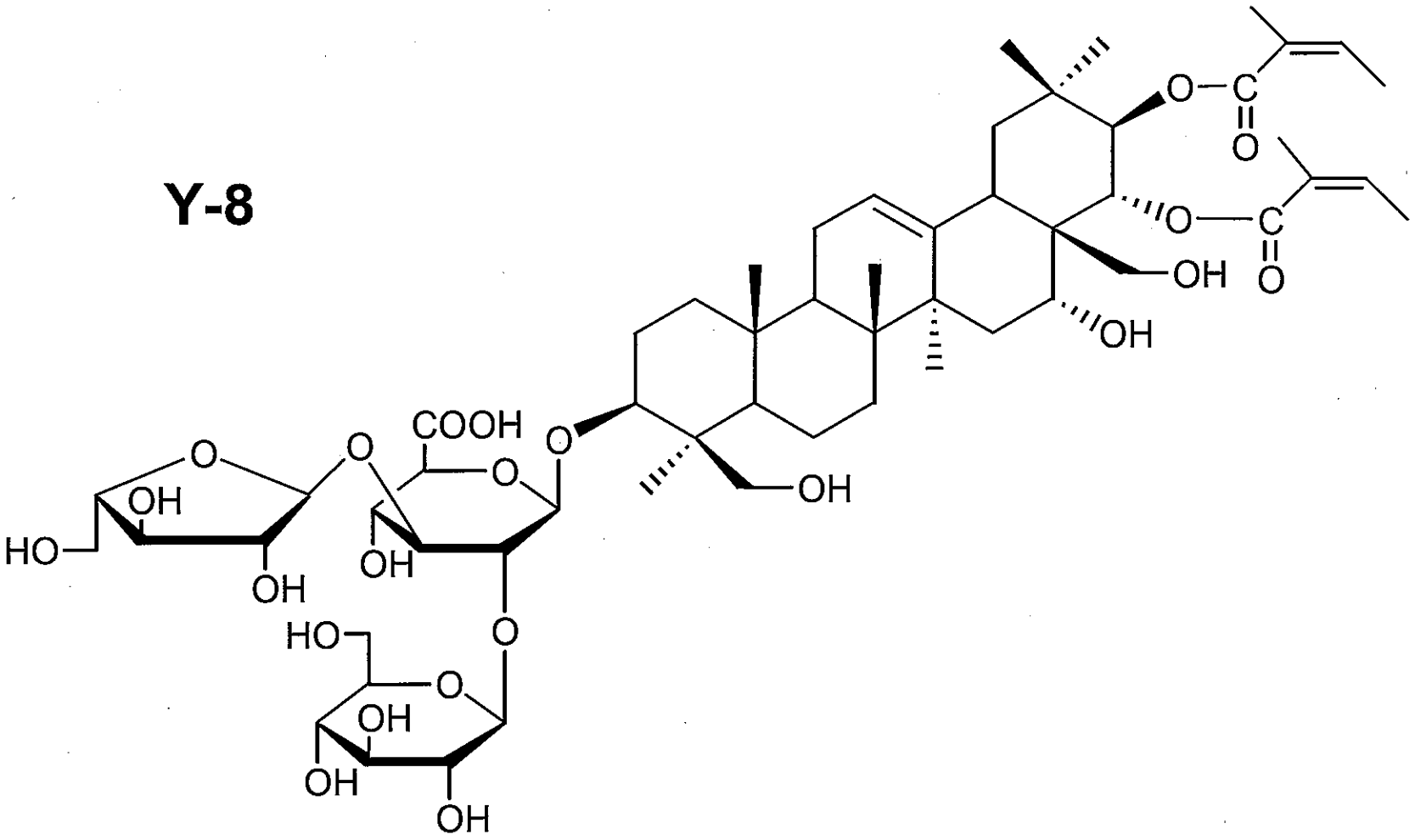


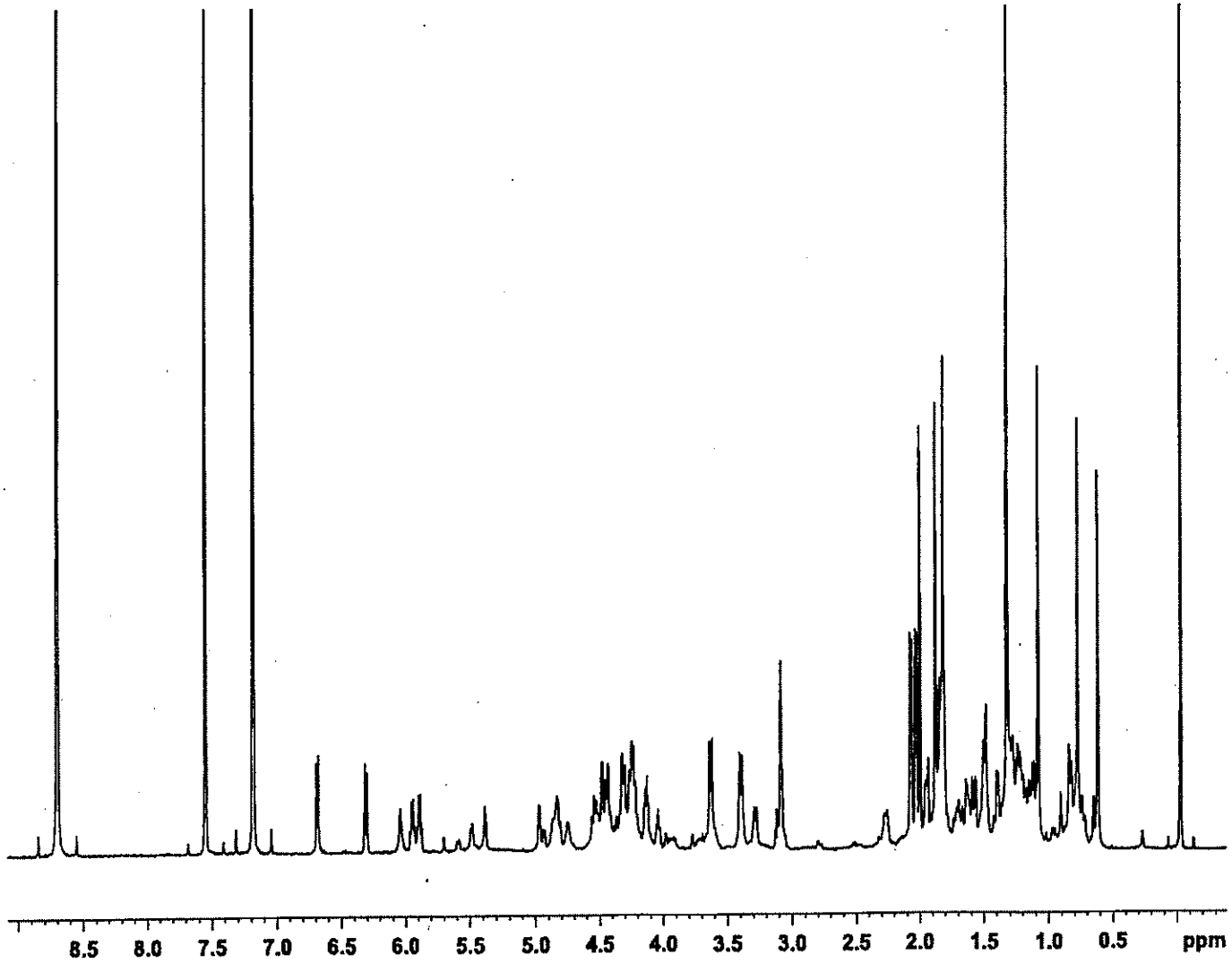
Figure 35
Structure of Y8



~~38/77~~ 39/66

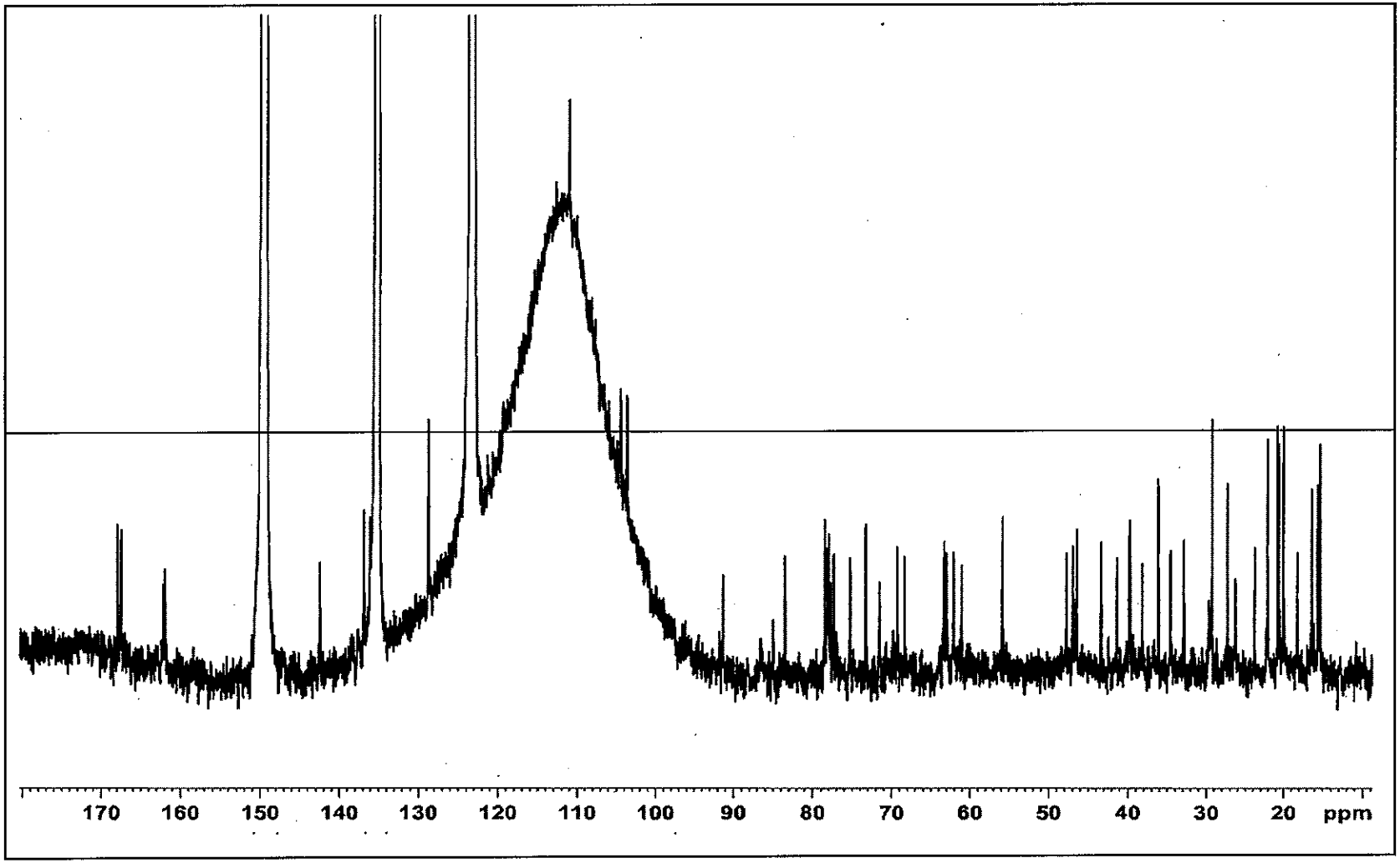
Figure 36

Y8-H-NMR-full



~~Figure 37~~

~~Y8-C13 NMR~~



40/77 66

Figure 38 7

Y8-HMQC-level-1

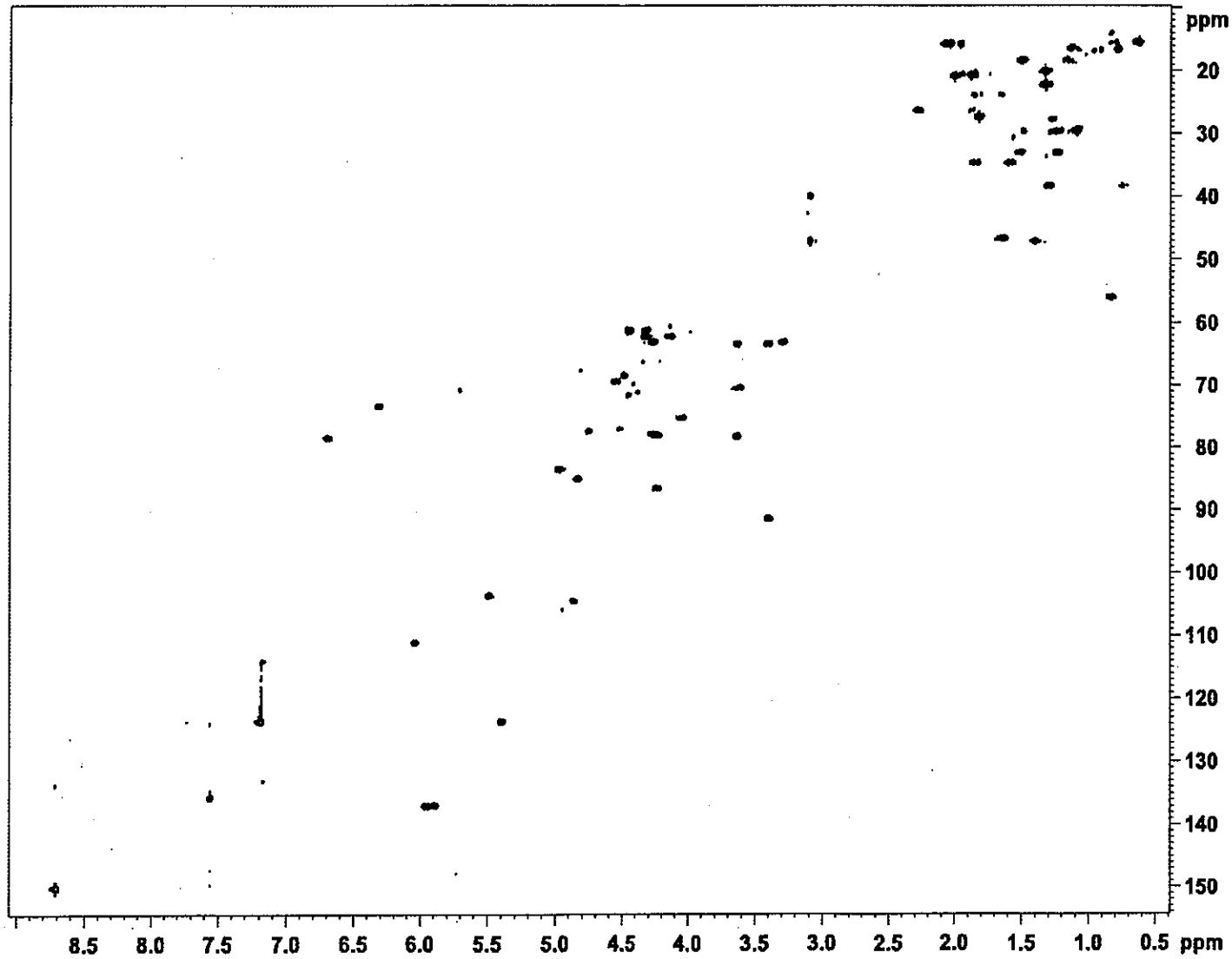


Figure 38

Y8-C13 NMR (or HMQC-level 2)

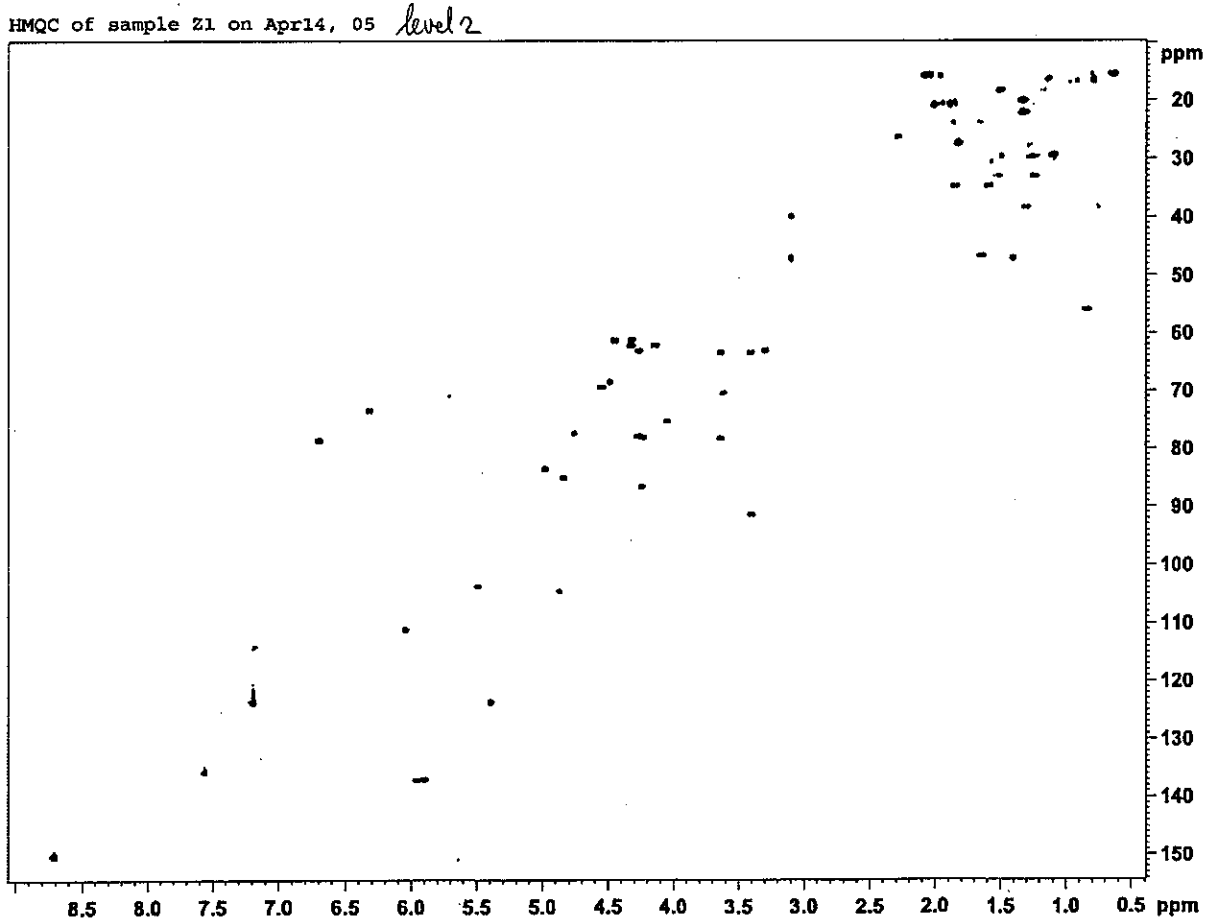


Figure 39
Structure of Y9

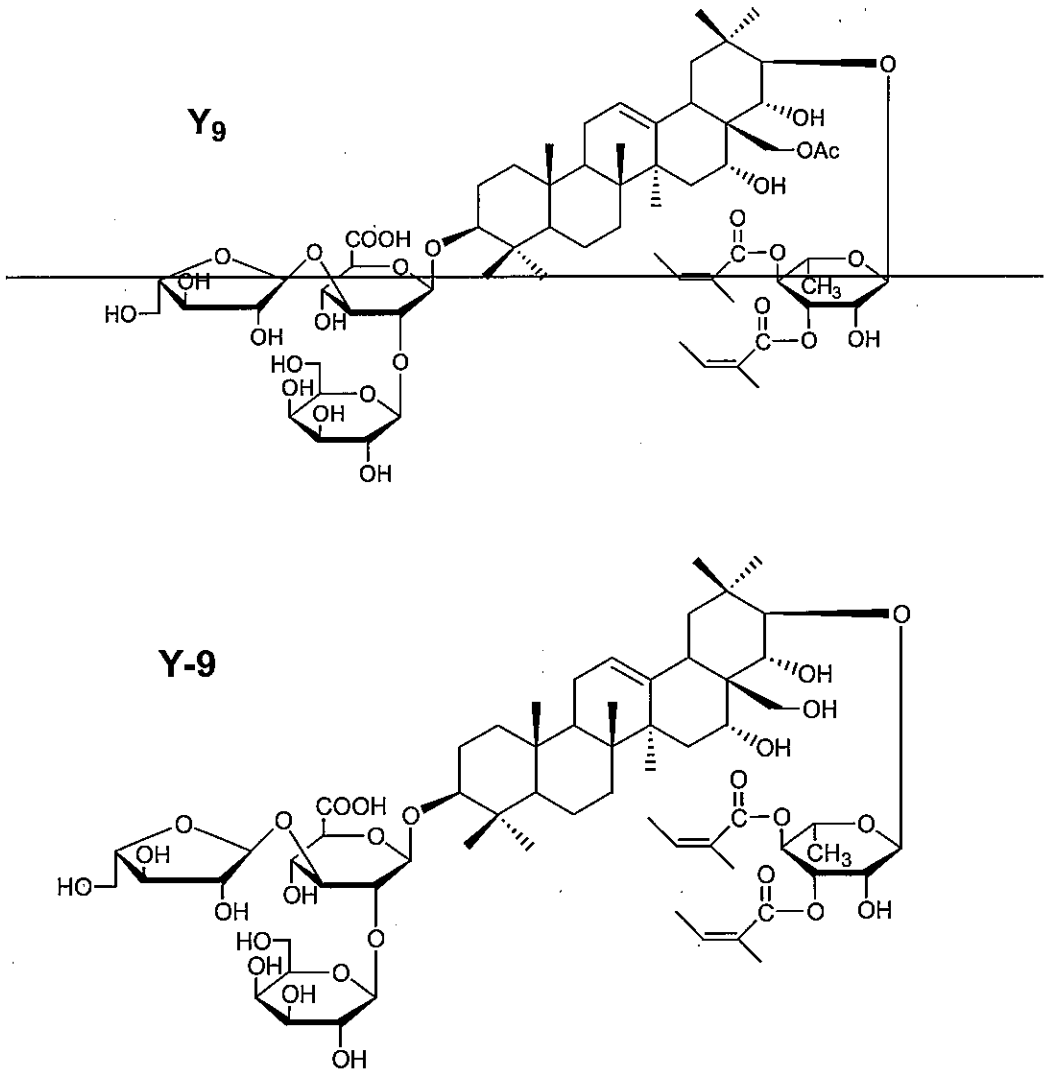


Figure 40

Y9-H-NMR

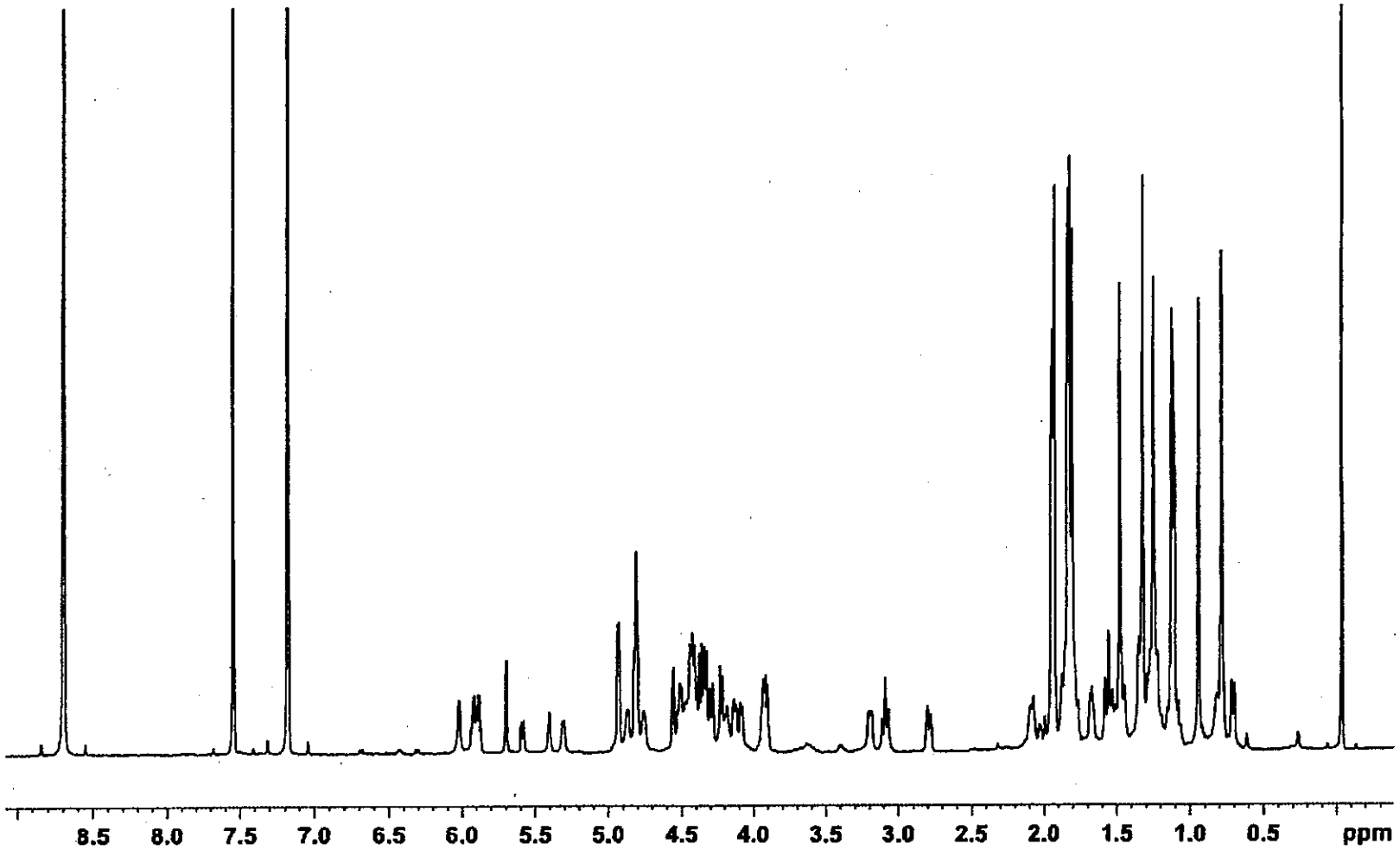


Figure 41
Y9-HMQC-level-1

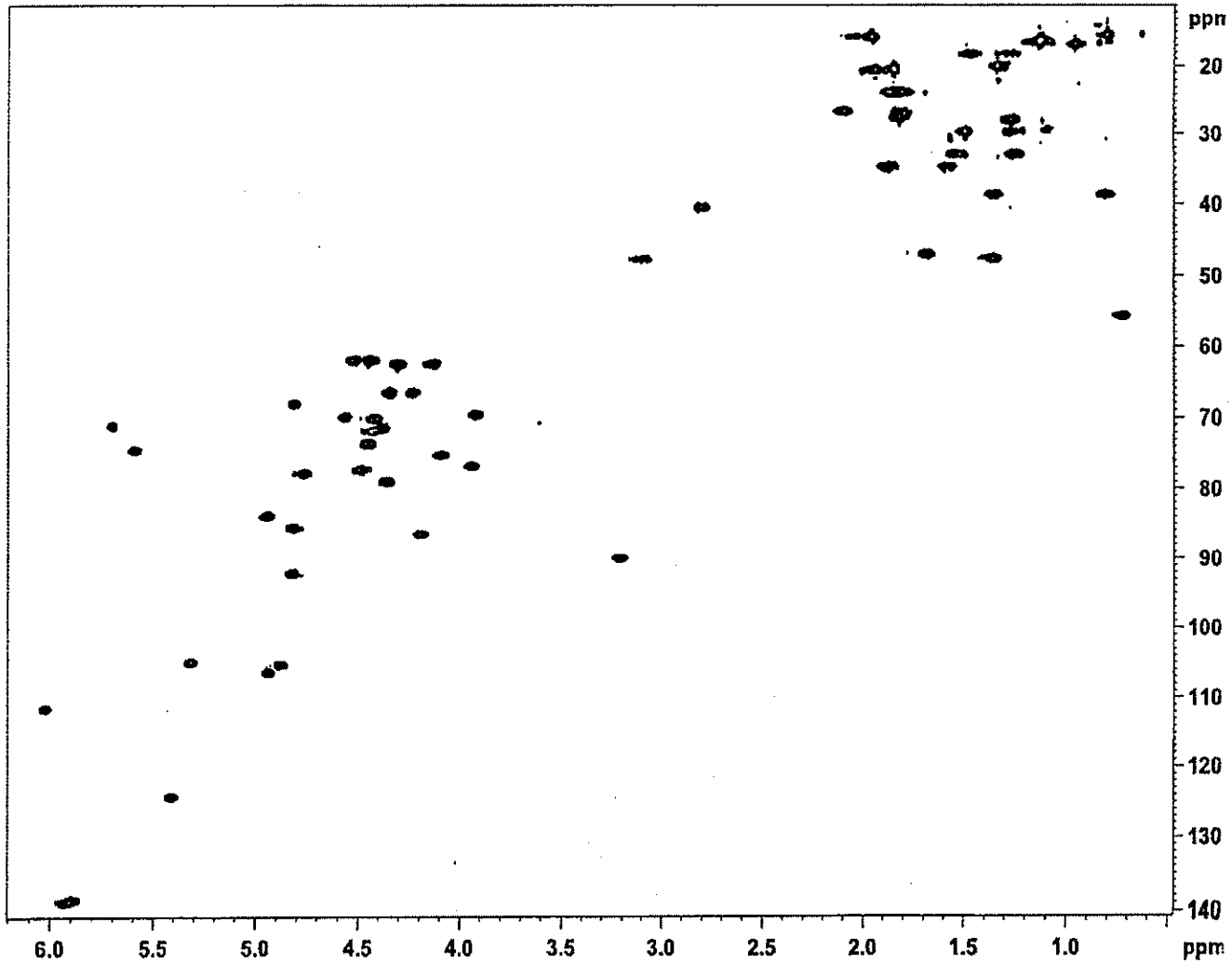
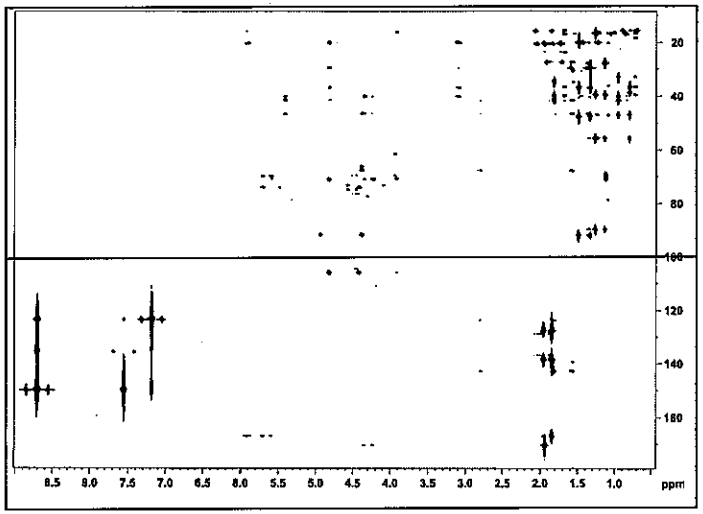


Figure 42

Y9 2D NMR (~~HMBC~~) (HMQC) level 2



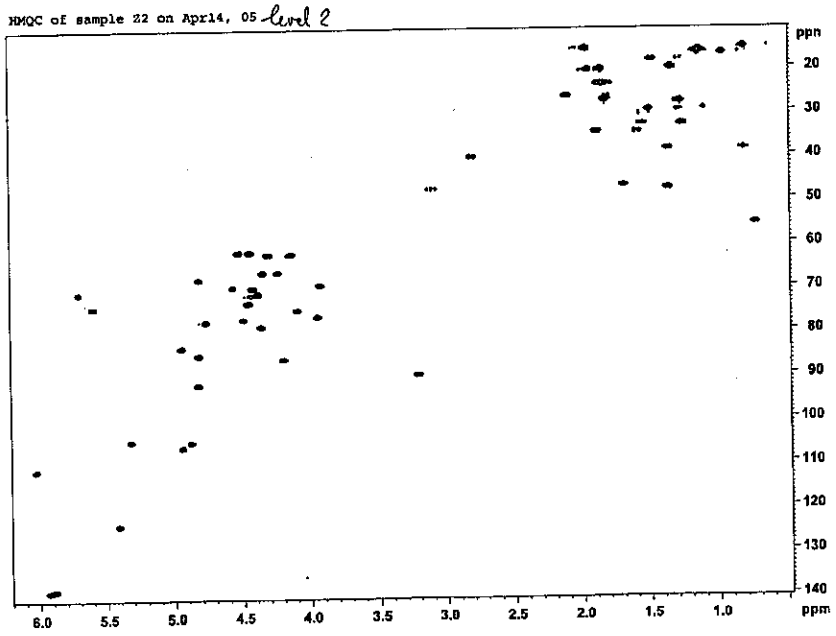


Figure 43
Structure of Y10

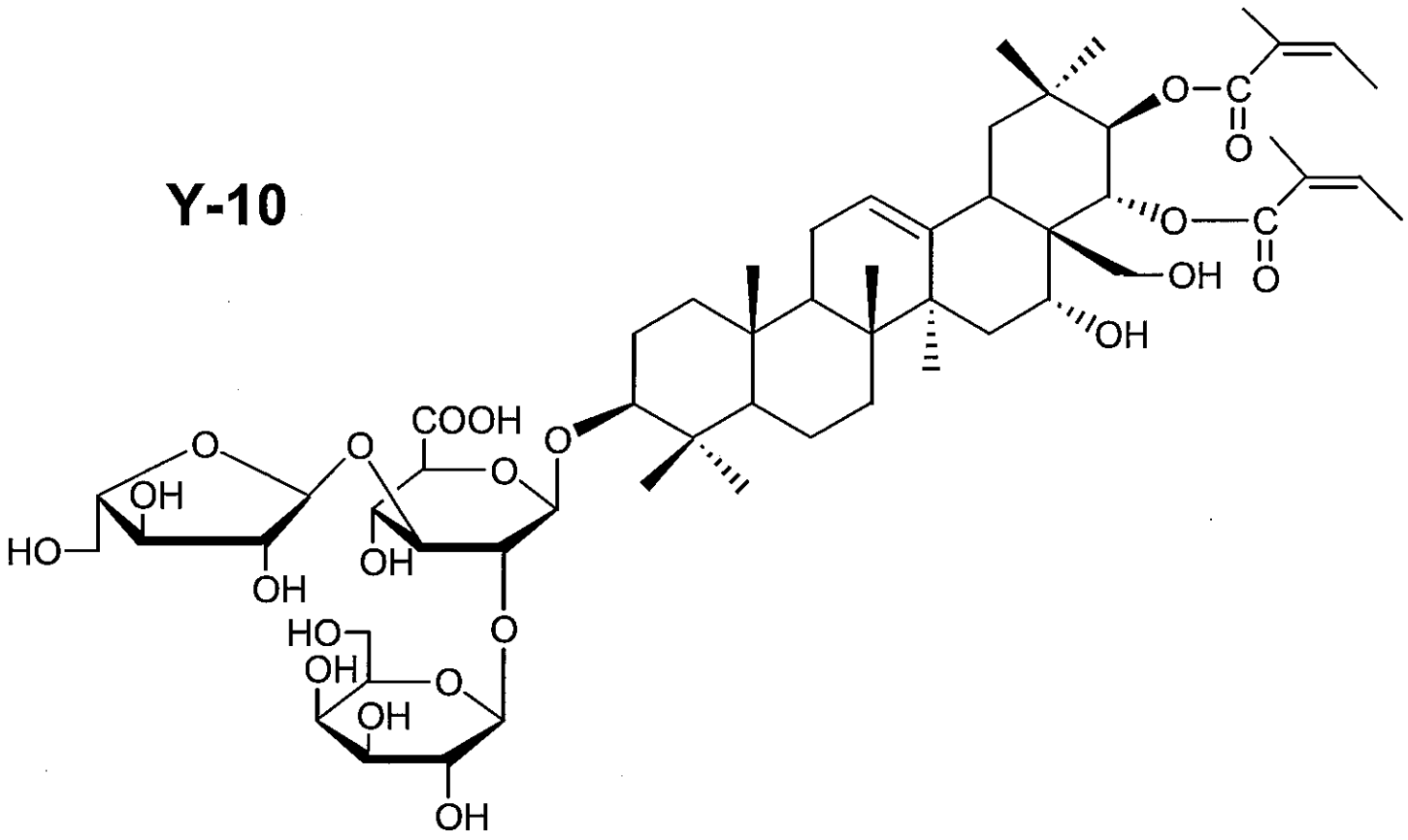
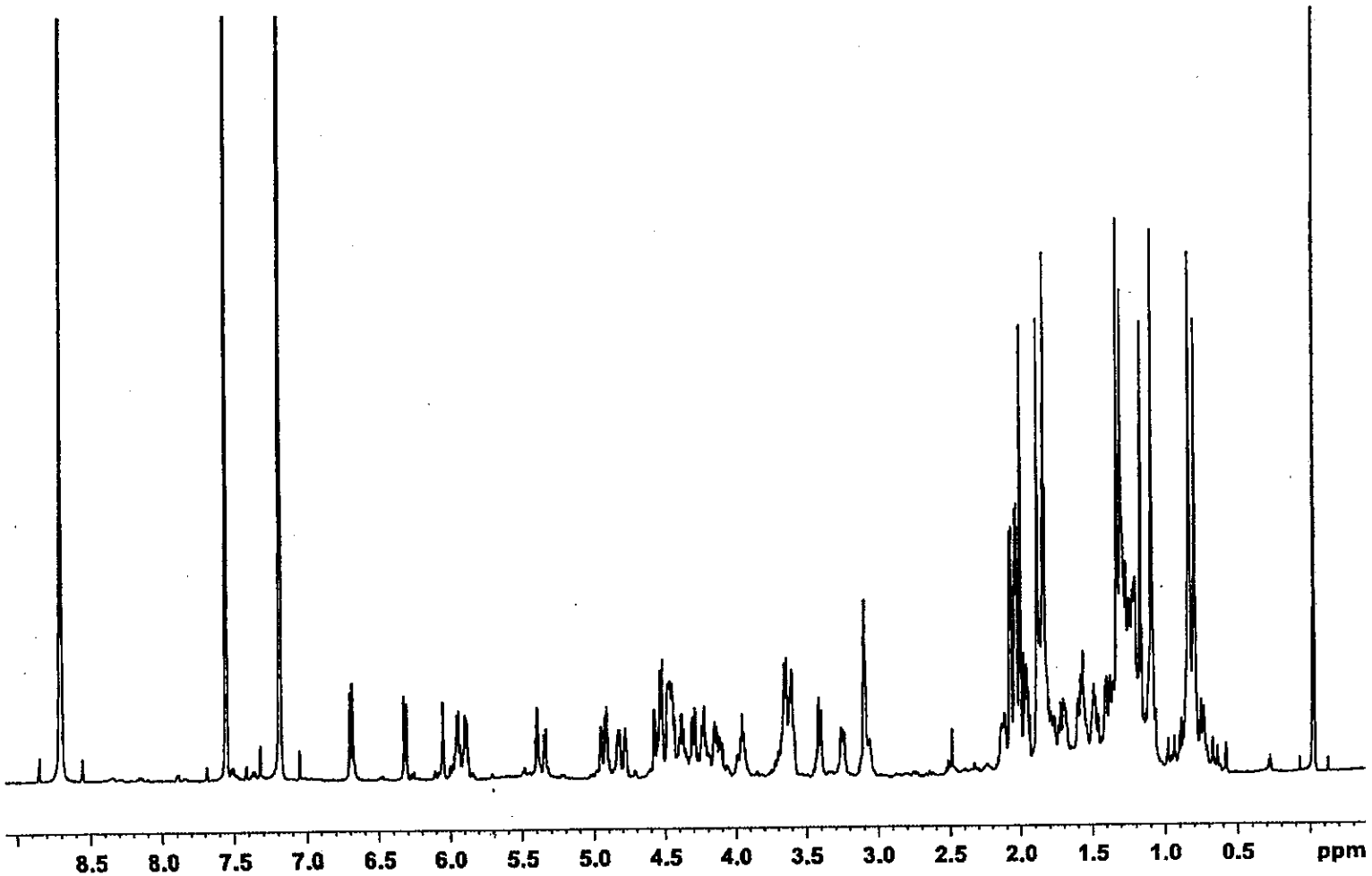
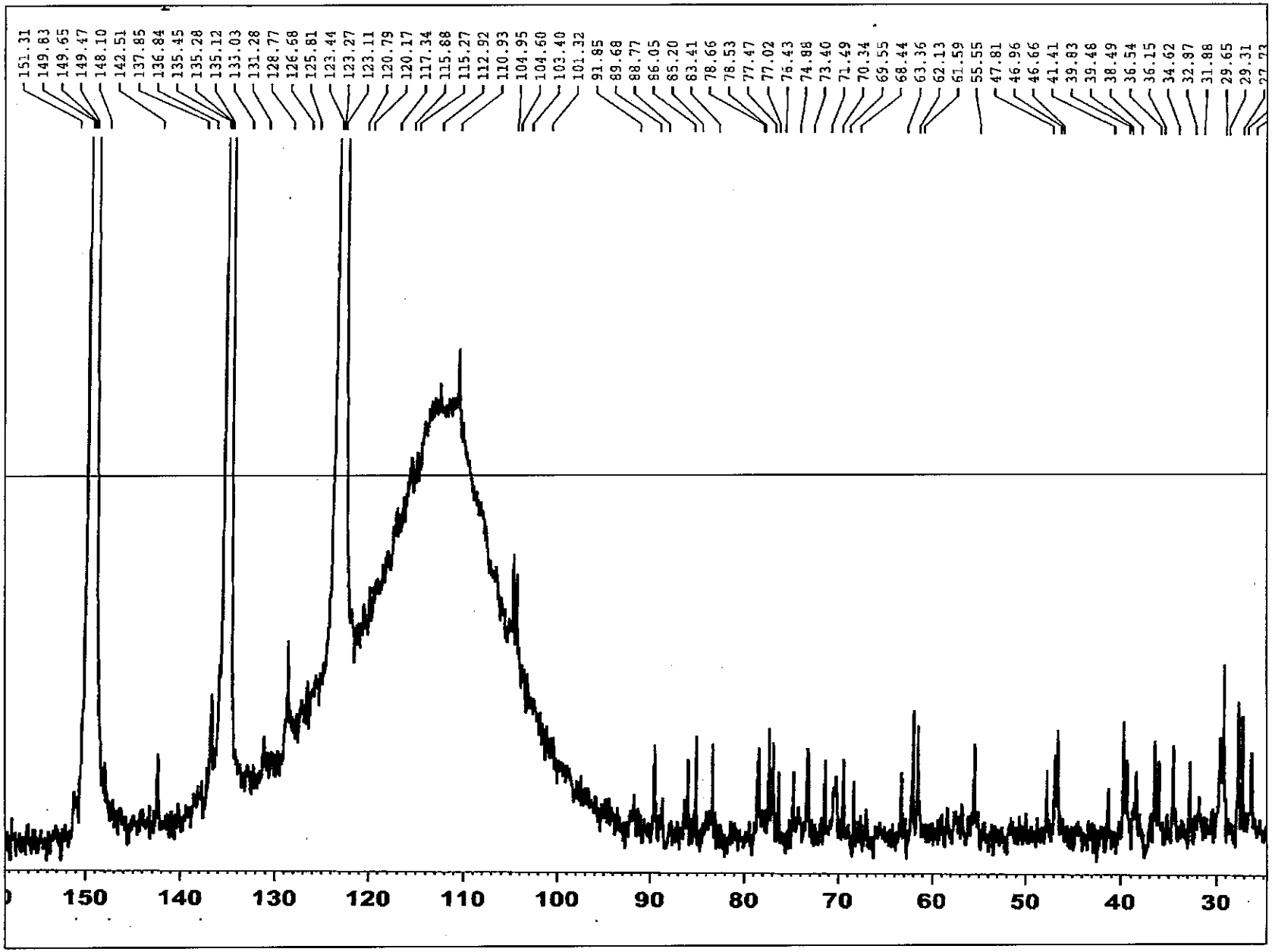


Figure 44
Y10-H-NMR



~~Figure 45~~

~~C13 NMR of Y10~~



48/77 66

Figure ~~46~~ 45

Y10-HMQC-level-1

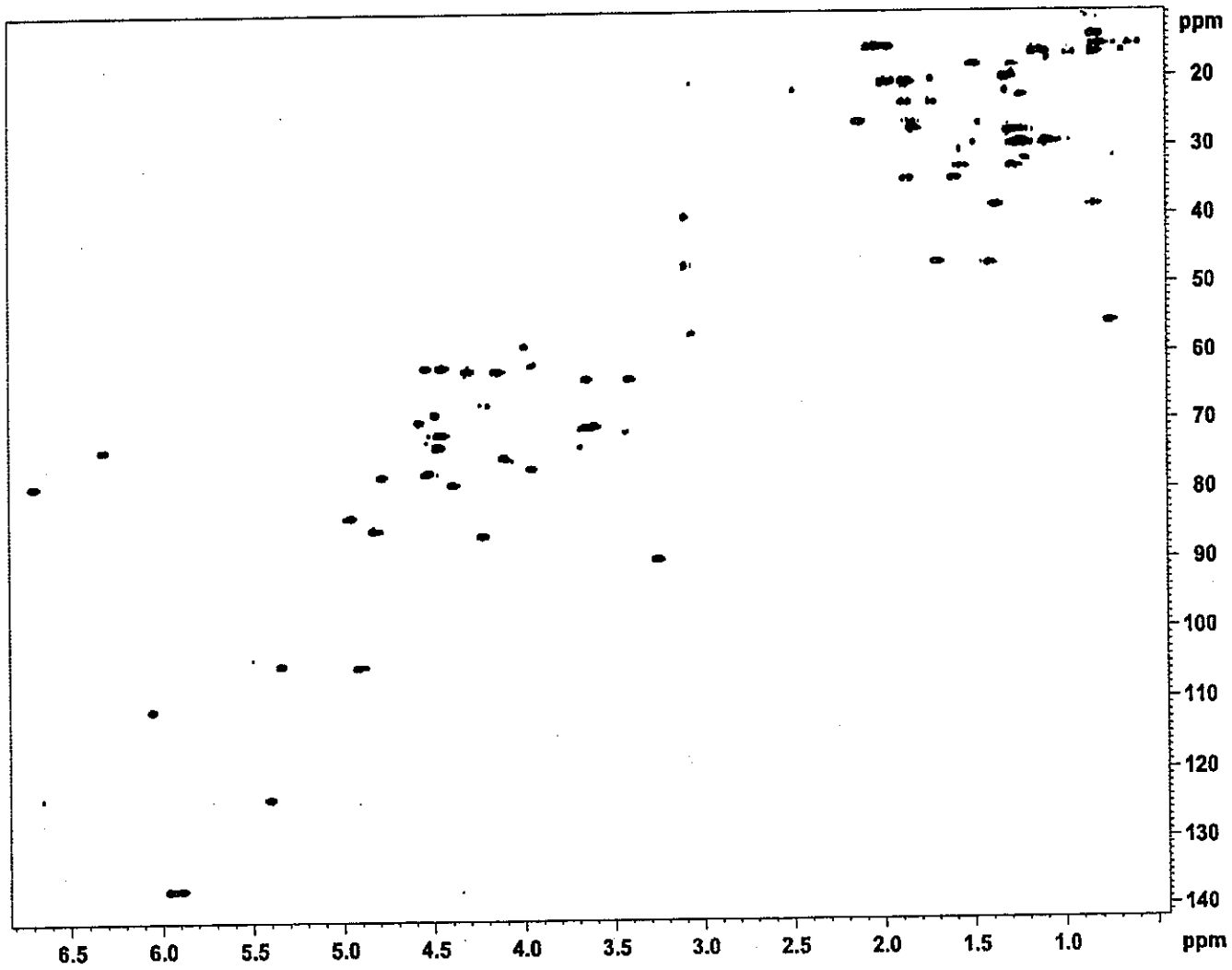


Figure 46

Y10-HMQC-level-2

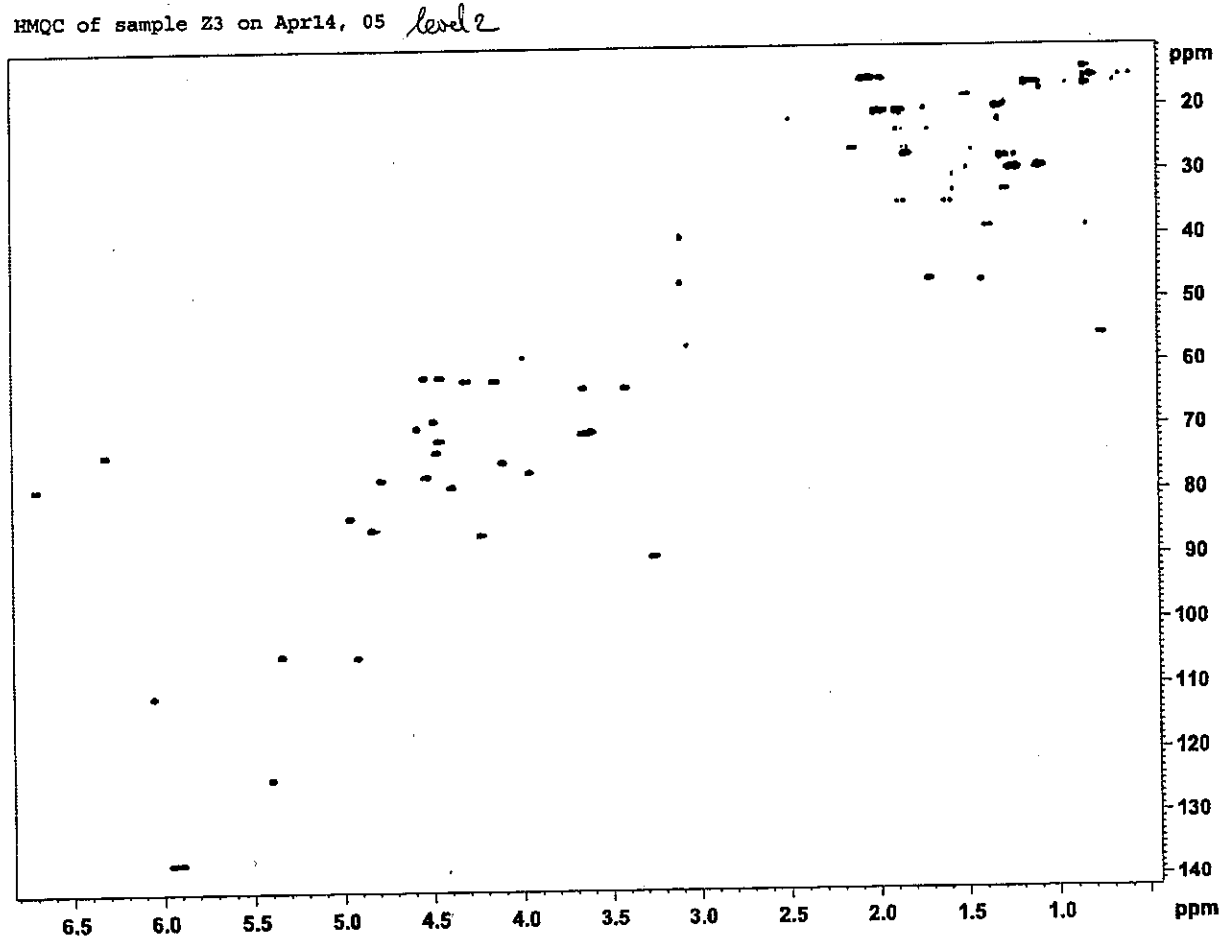
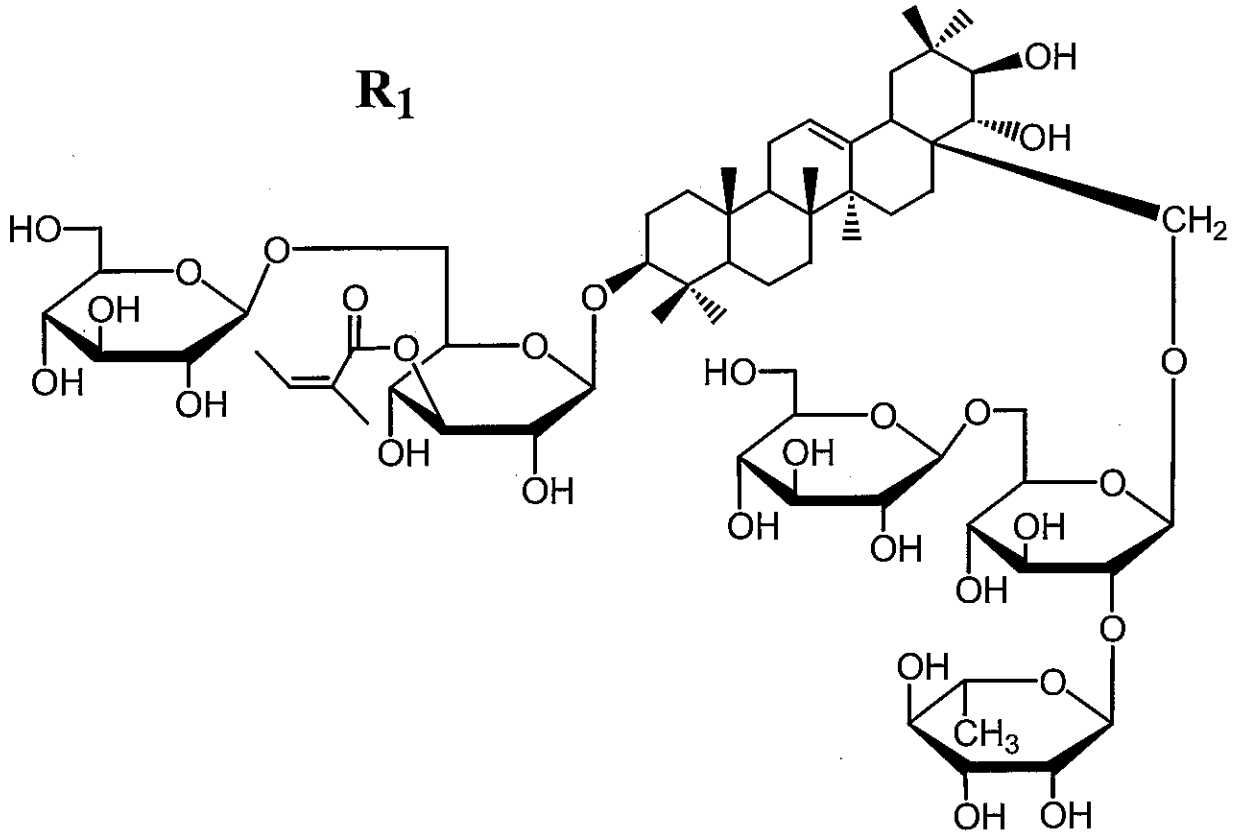


Figure 47
R1-Structure

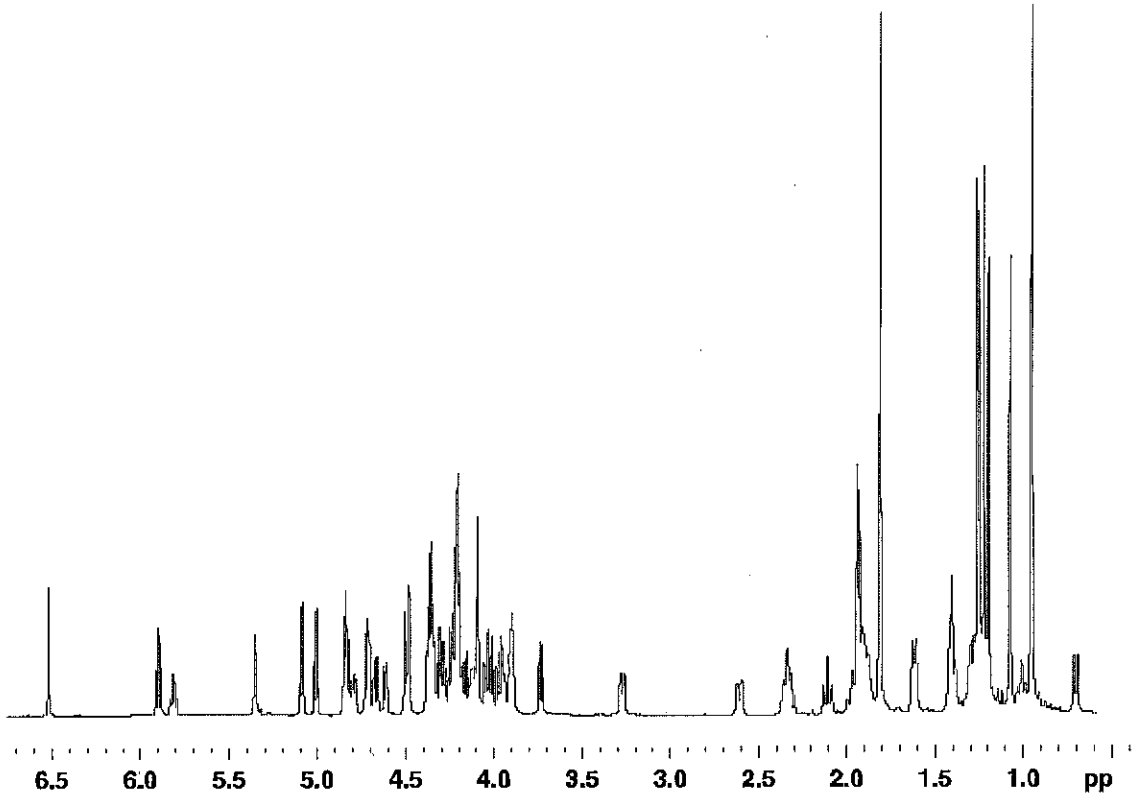


R1: 3-*O*-[angeloyl-(1→3)-*β*-D-glucopyranosyl-(1→6)]-*β*-D-glucopyranosyl-28-*O*-[*α*-L-rhamnopyranosyl-(1→2)-*β*-D-glucopyranosyl-(1→6)-*β*-D-glucopyranosyl-3*β*, 21*β*, 22*α*, 28-tetrahydroxyolean-12-ene

~~50/77~~ 51/66

Figure 48

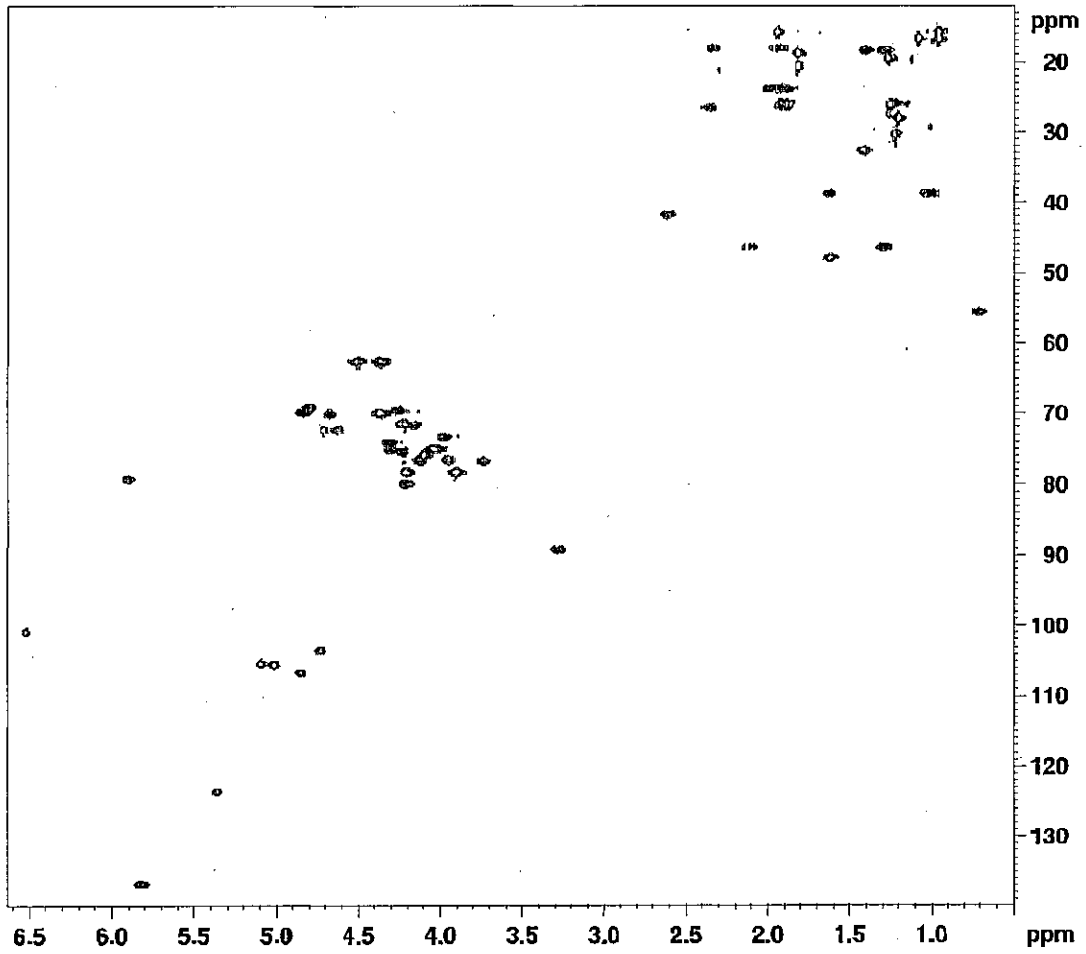
R1-H-NMR



51/77 52/66

Figure 49

R1-HMQC



~~52/77~~ 53/66

Figure 50

R1-HMBC-level 1

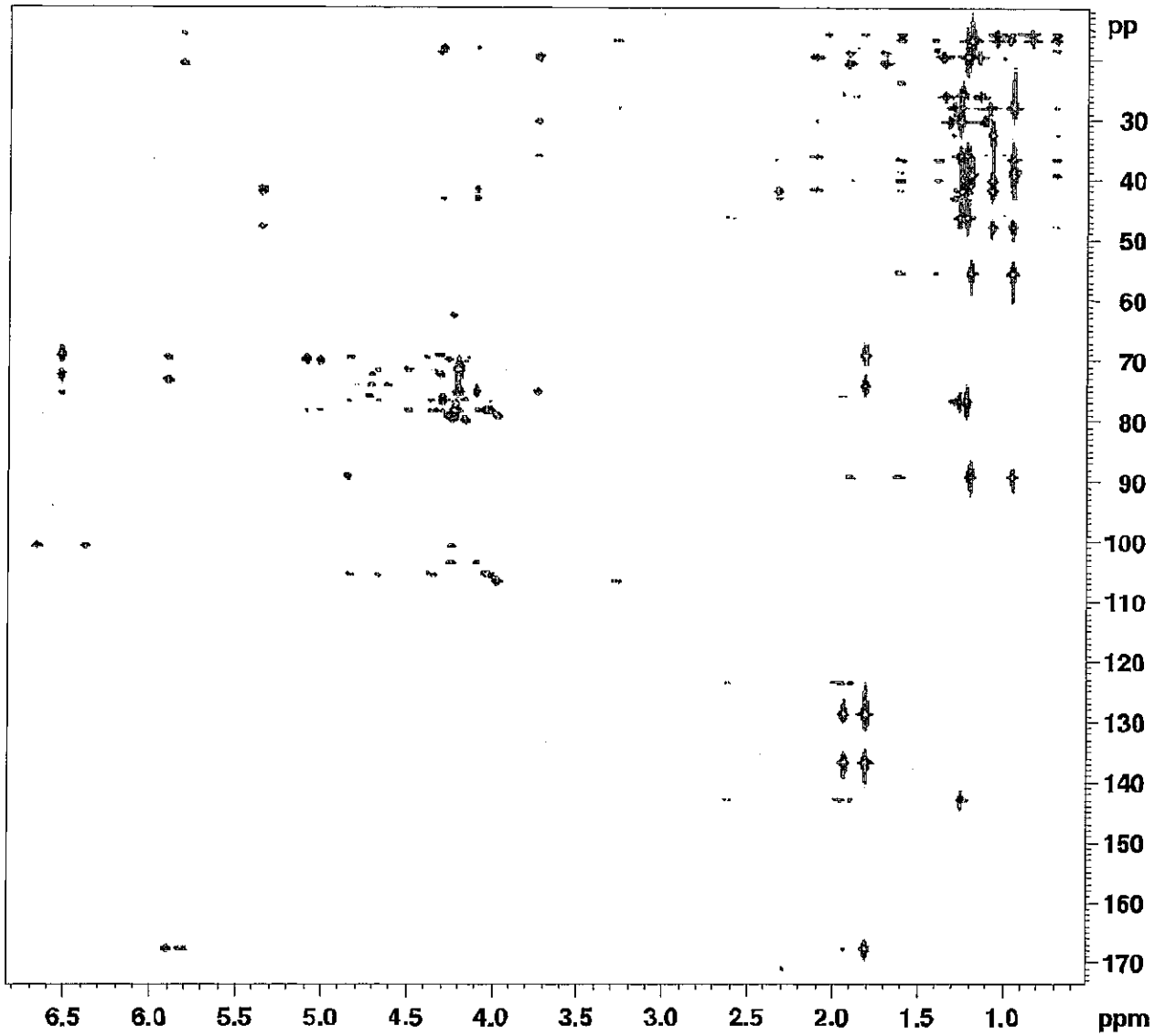


Figure 51
R1-COSY

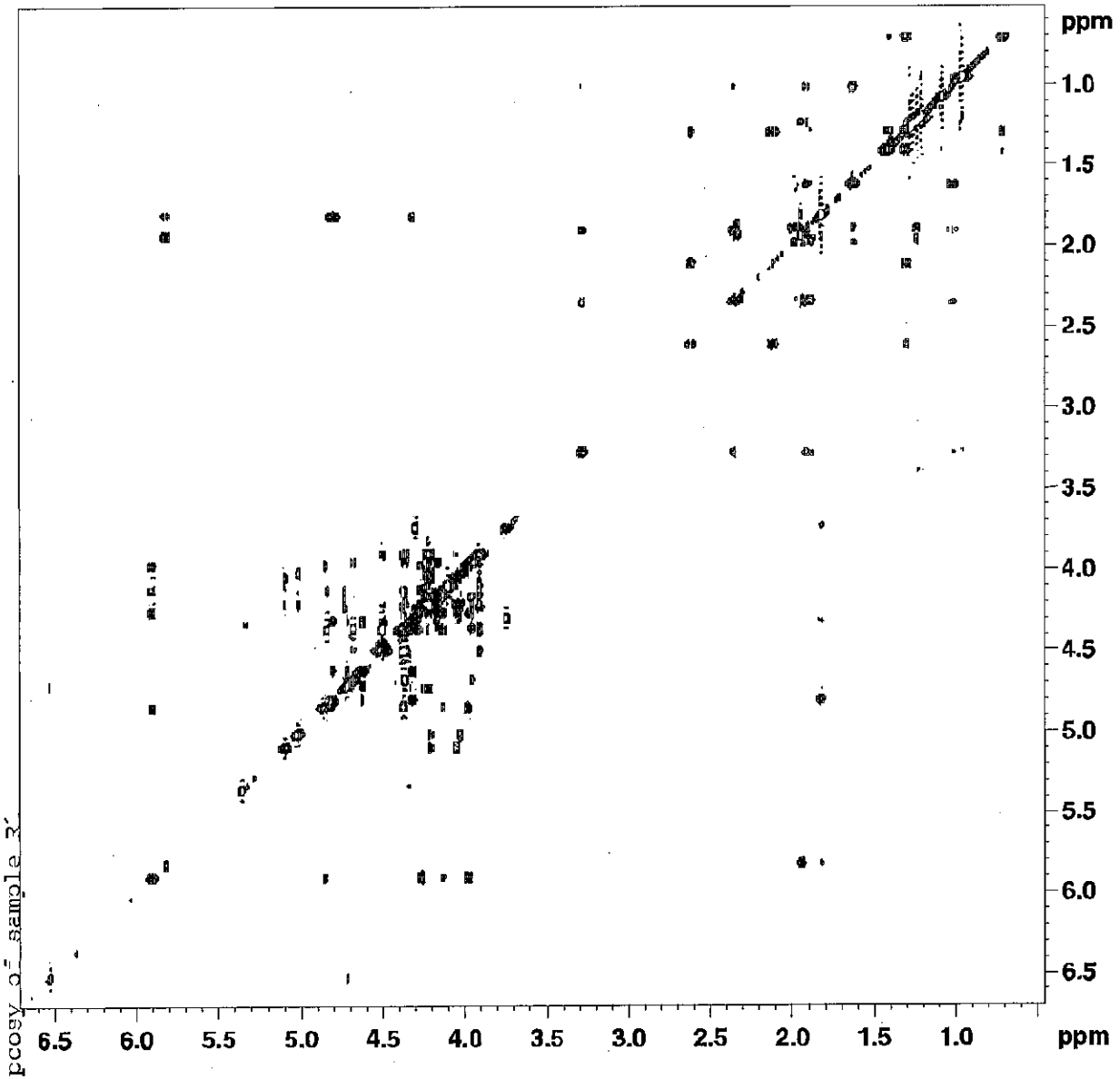


Figure 52
R1-C13-1

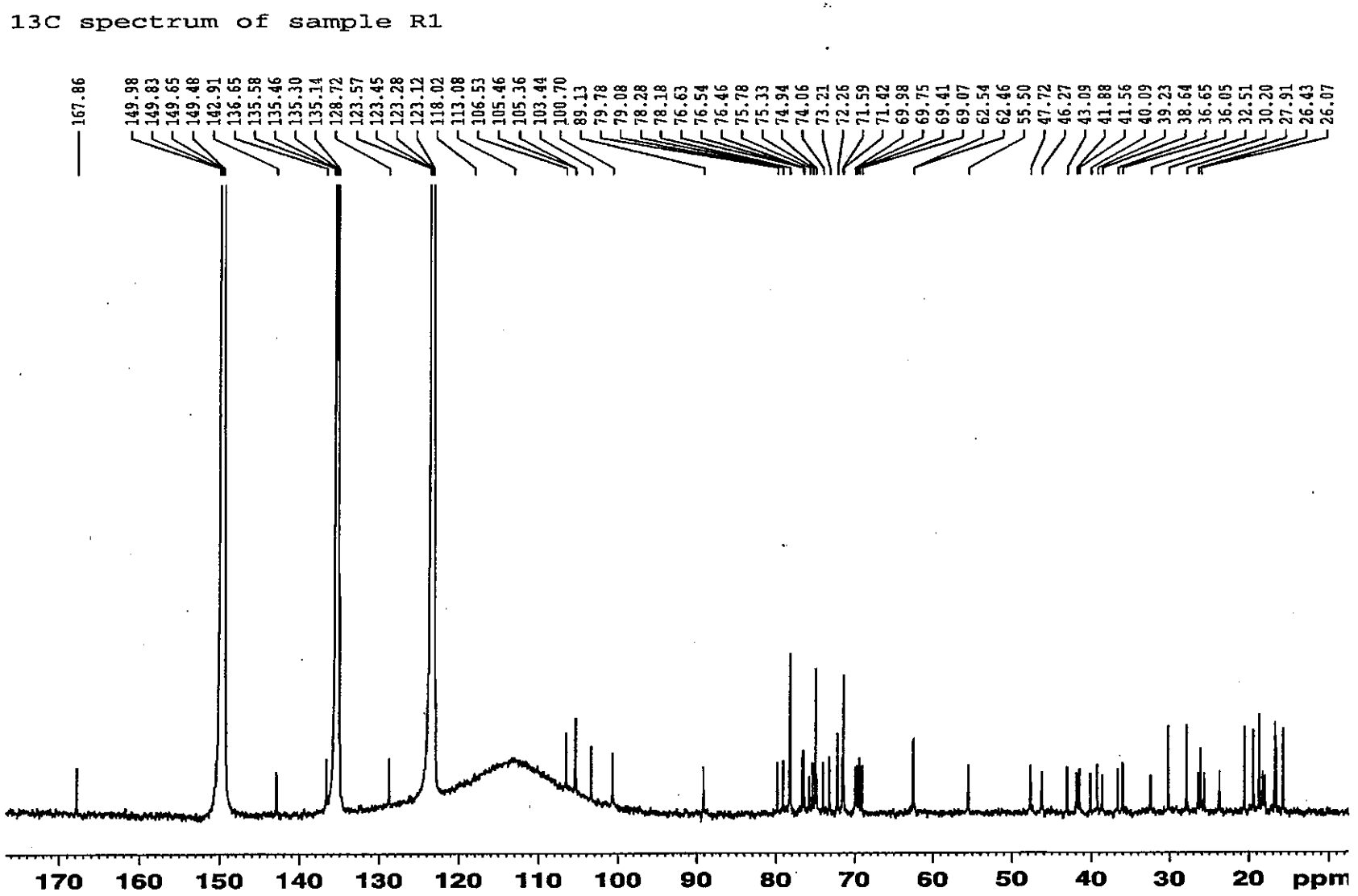
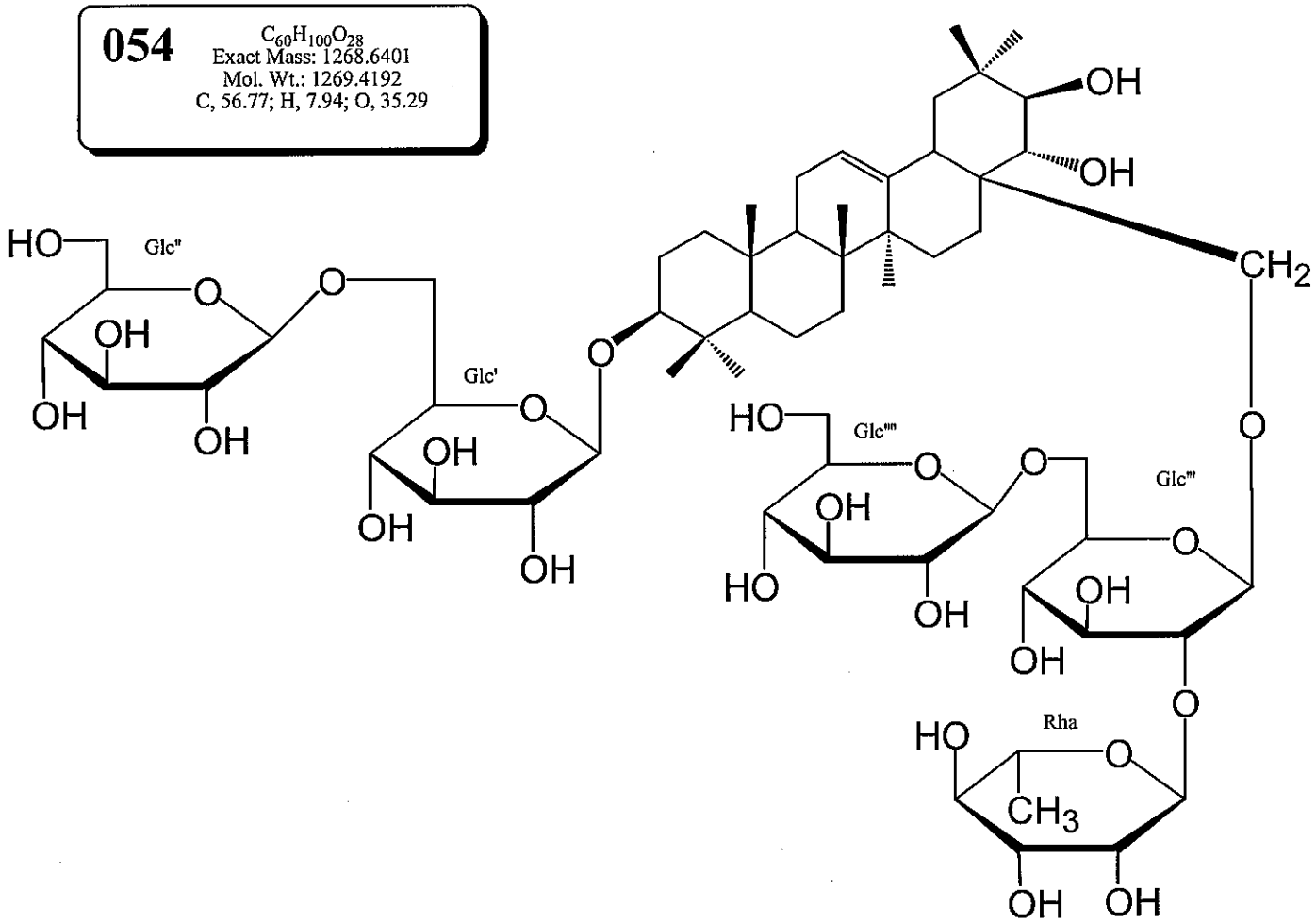


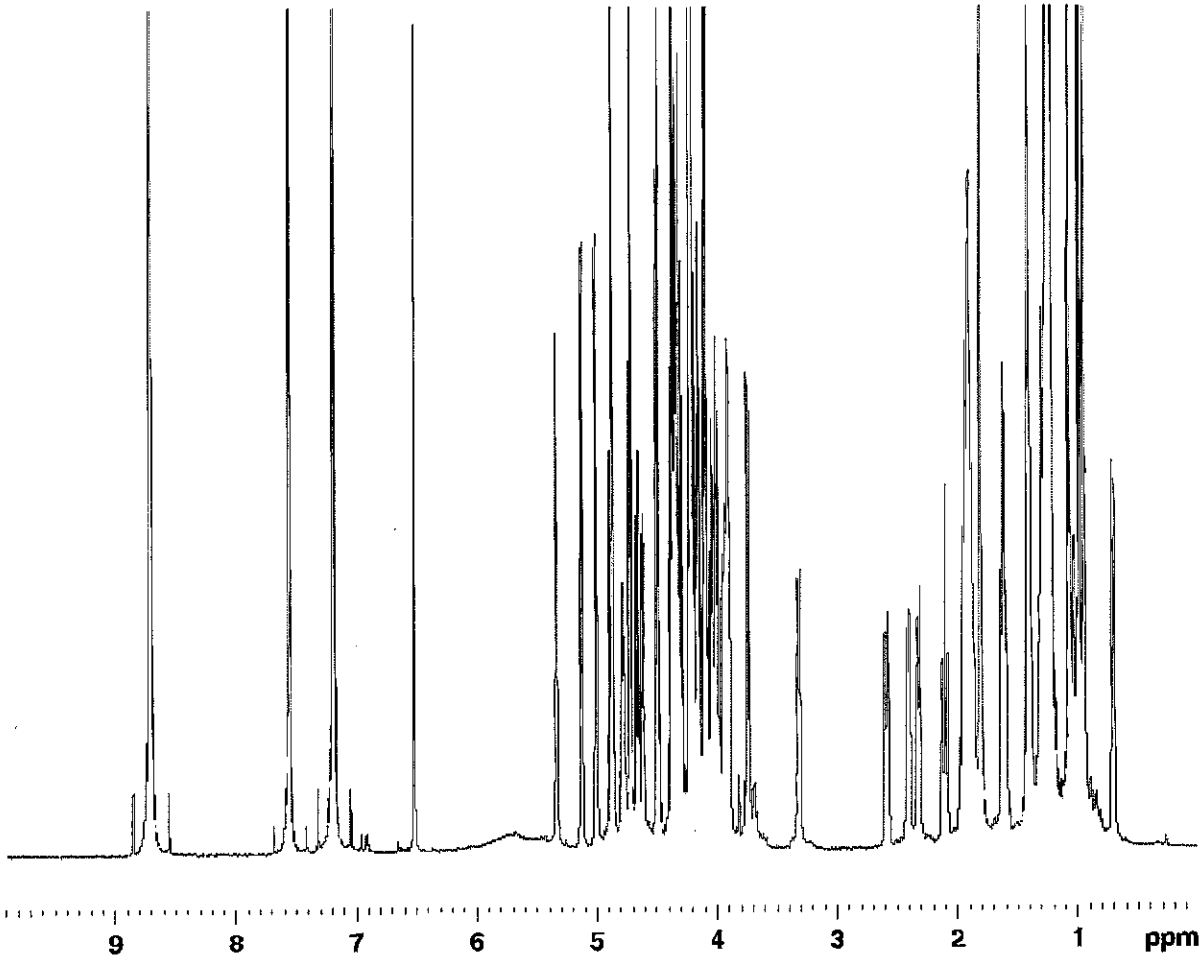
Figure 53
Structure of Compound O54



~~56/77~~ 57/66

Figure 54

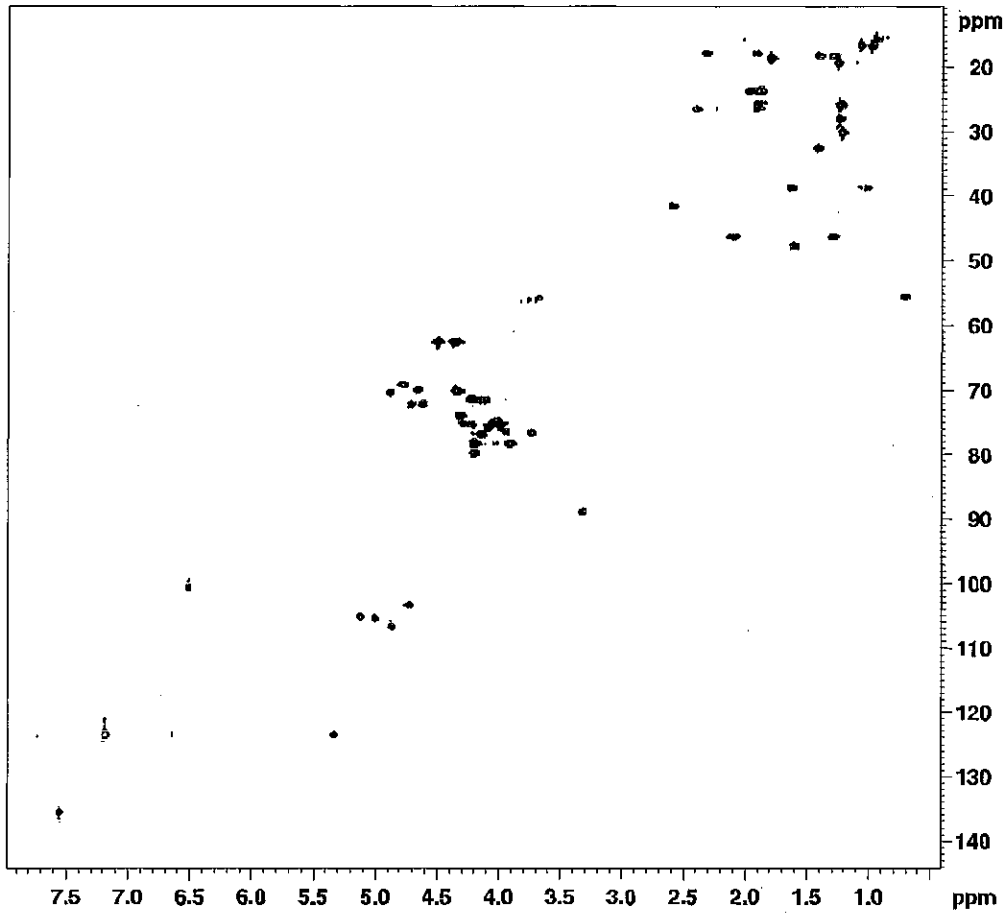
O54-H-NMR



~~57/77~~ 58/66

Figure 55

O54-HMQC



~~58/77~~ 59/66
Figure 56
O54-HMBC

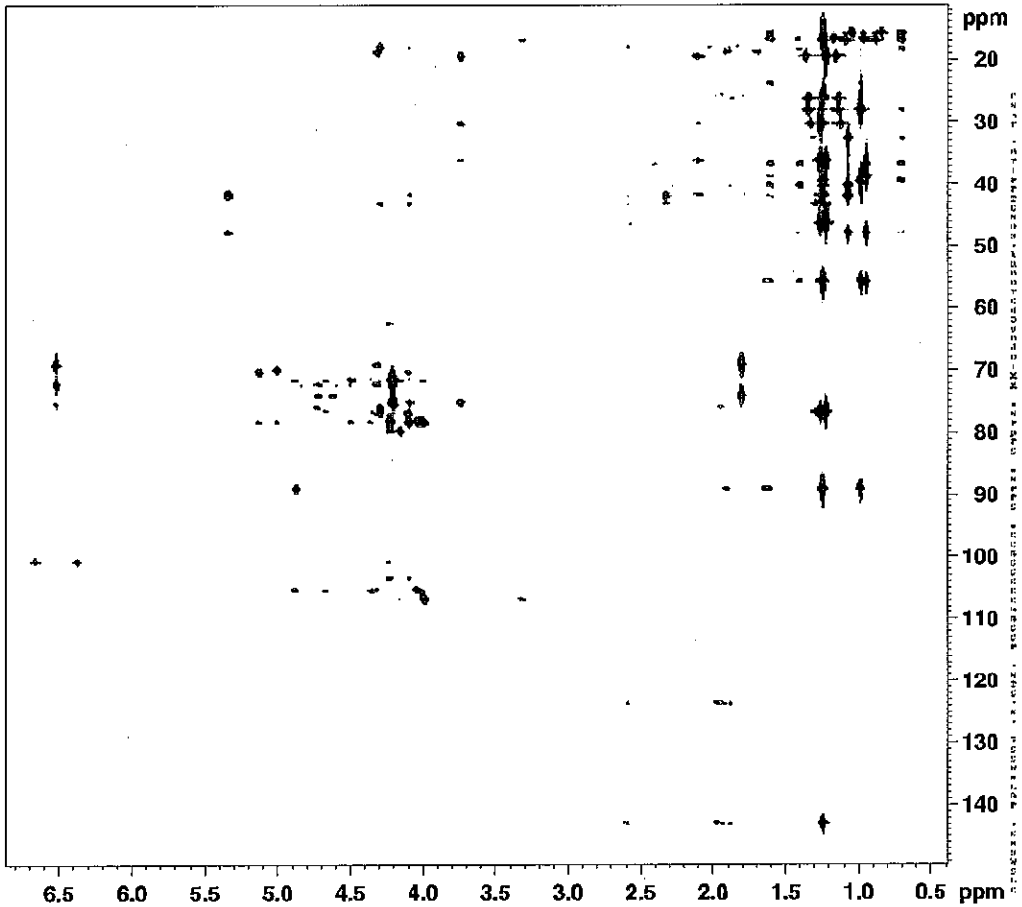
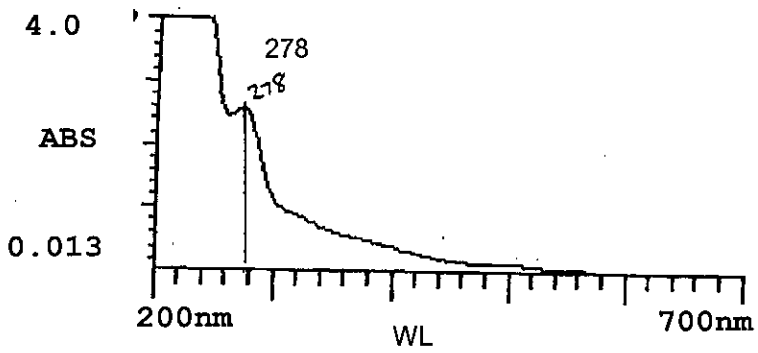
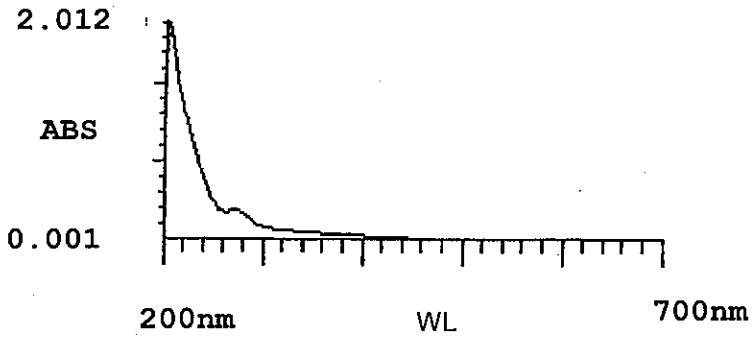
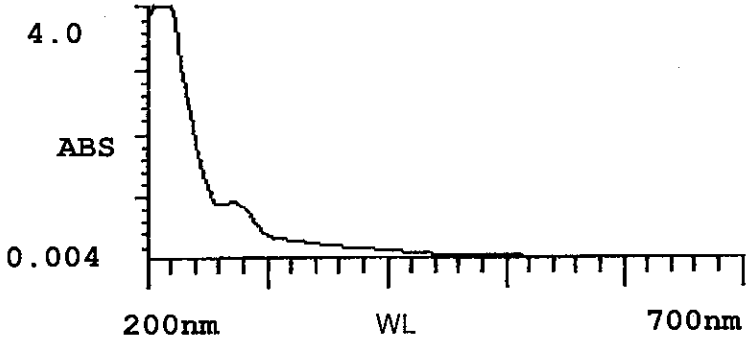
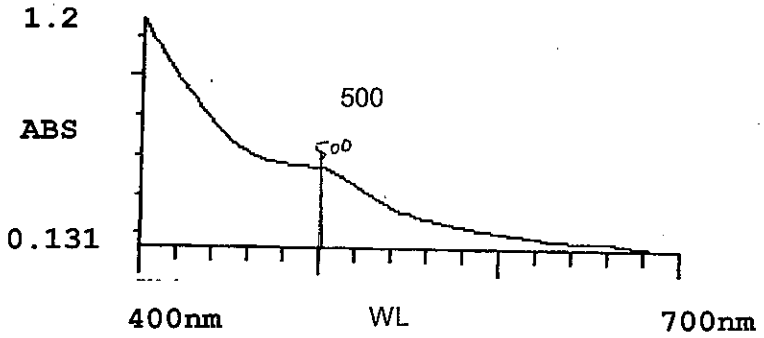


Figure 57


4.0
ABS
0.013
278
278
200nm
WL
700nm


2.012
ABS
0.001
200nm
WL
700nm


4.0
ABS
0.004
200nm
WL
700nm


1.2
ABS
0.131
500
500
400nm
WL
700nm

~~60/77~~ 61/66

Figure 58

H-NMR of Y4

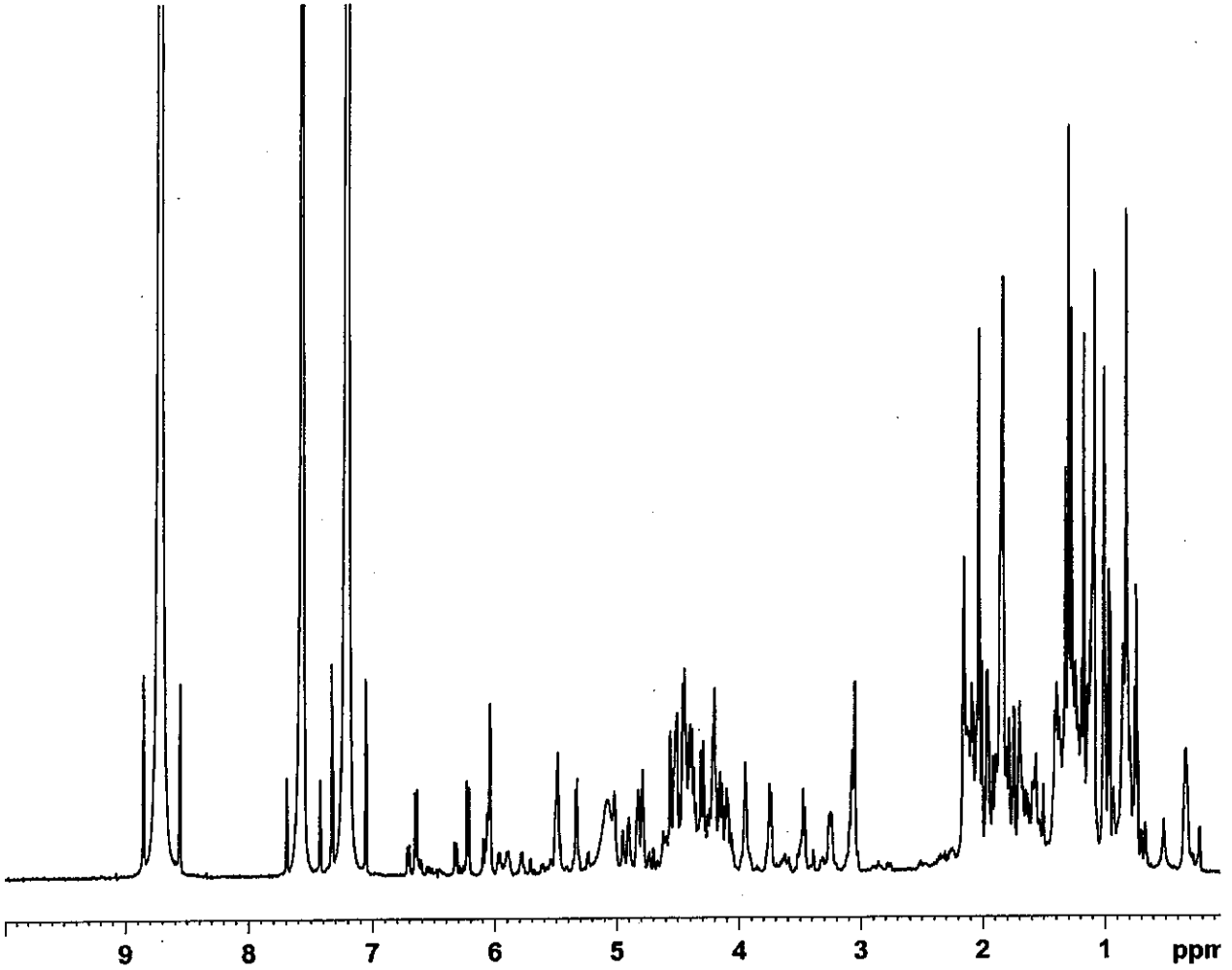


Figure 59
HMQC of Y4

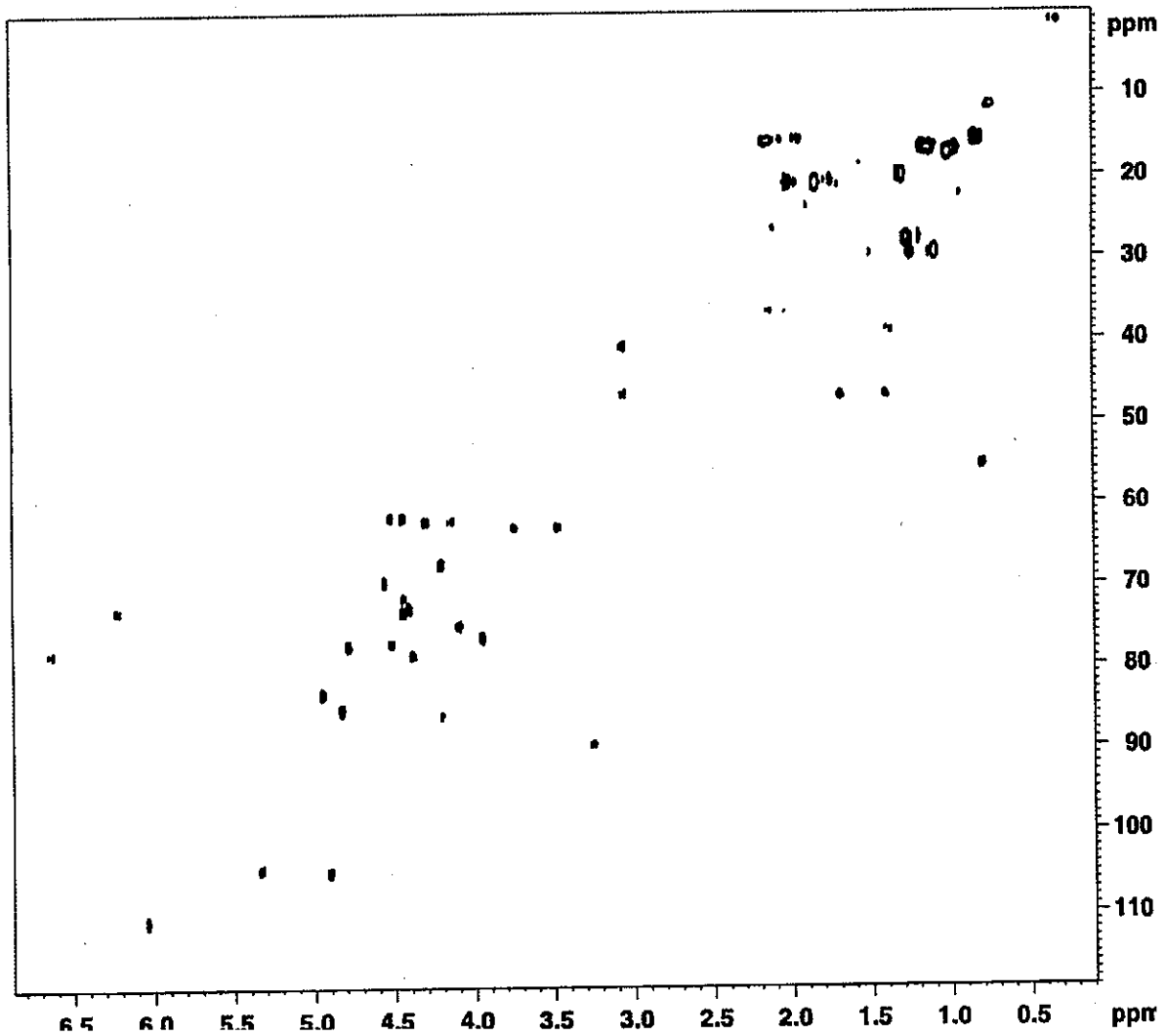
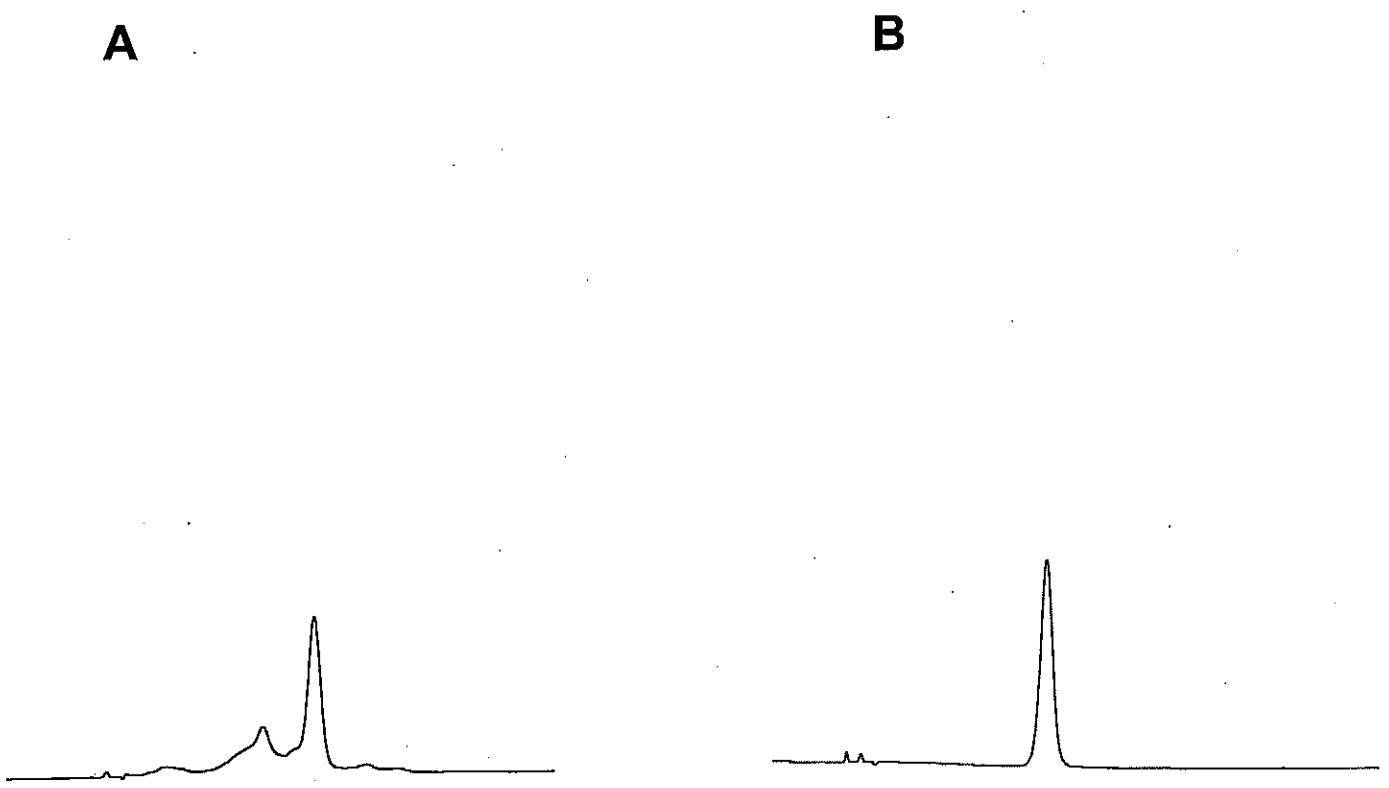
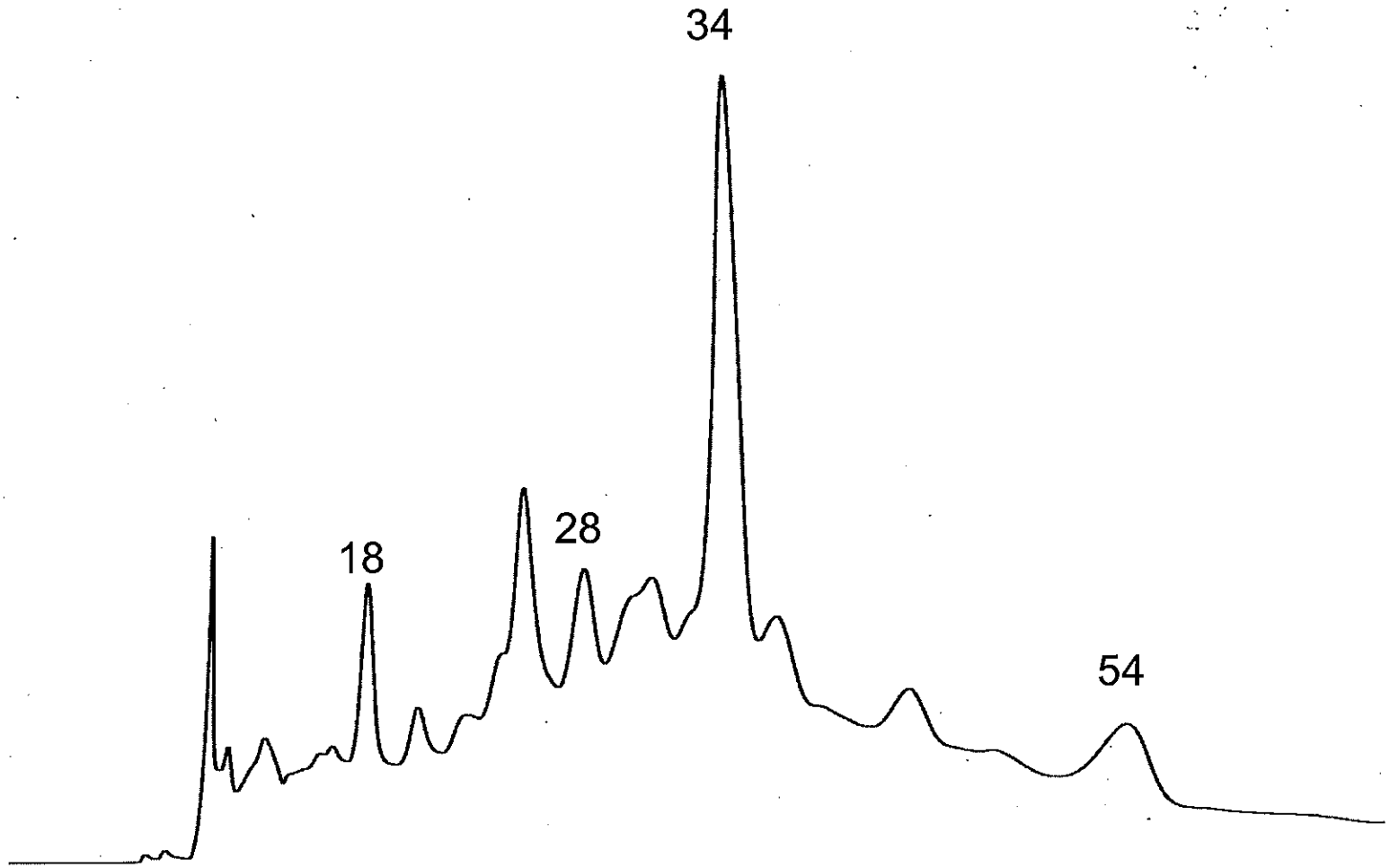


Figure 60



~~63/77~~ 64/66

Figure 61
(iso-20)



~~64/77~~ 65/66

Figure 62

Re-chromatography of ~~#54~~ #28 and #34 (from iso-20)

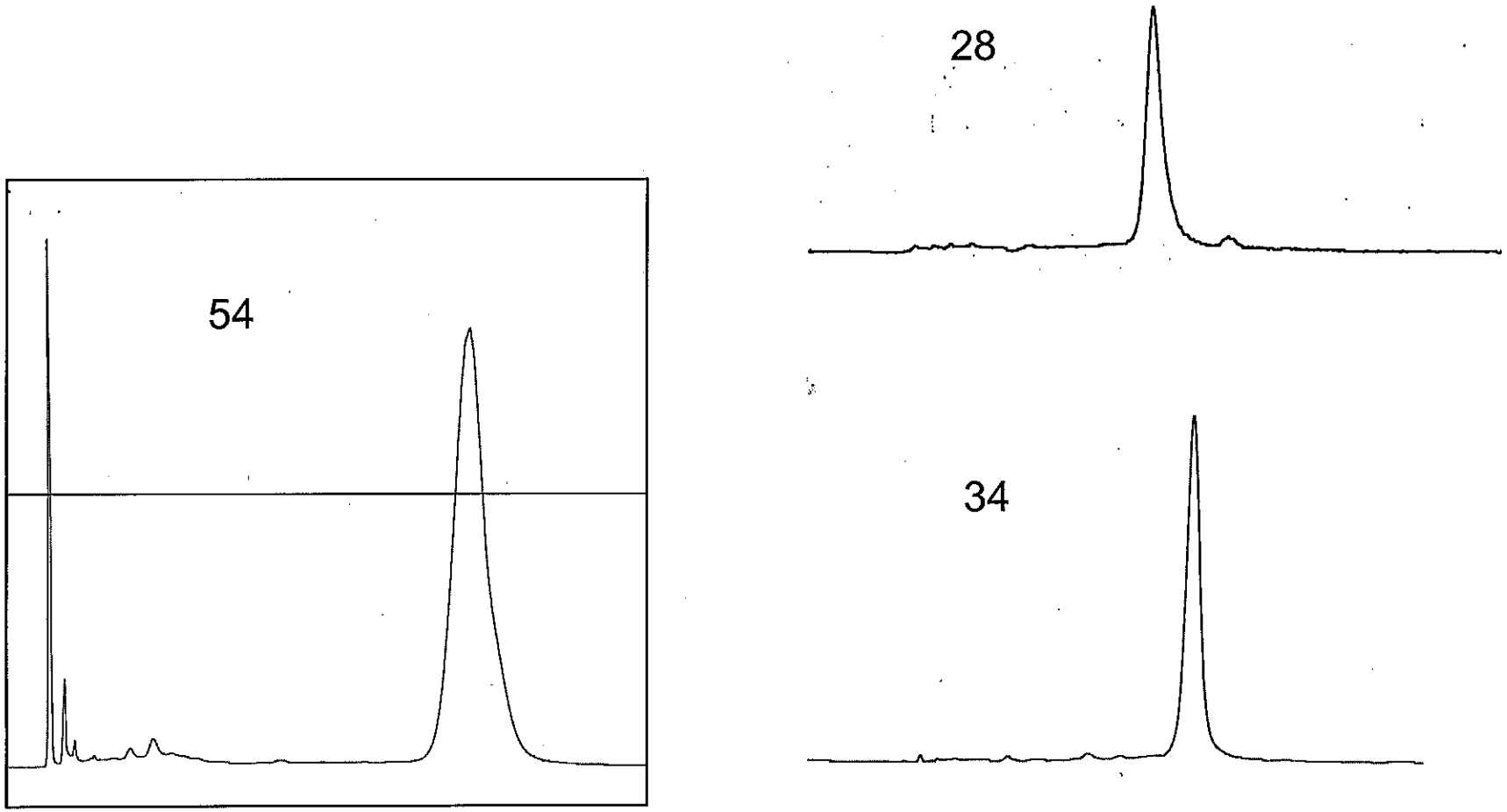


Figure 63
Re-chromatography of #54 (from iso-20)

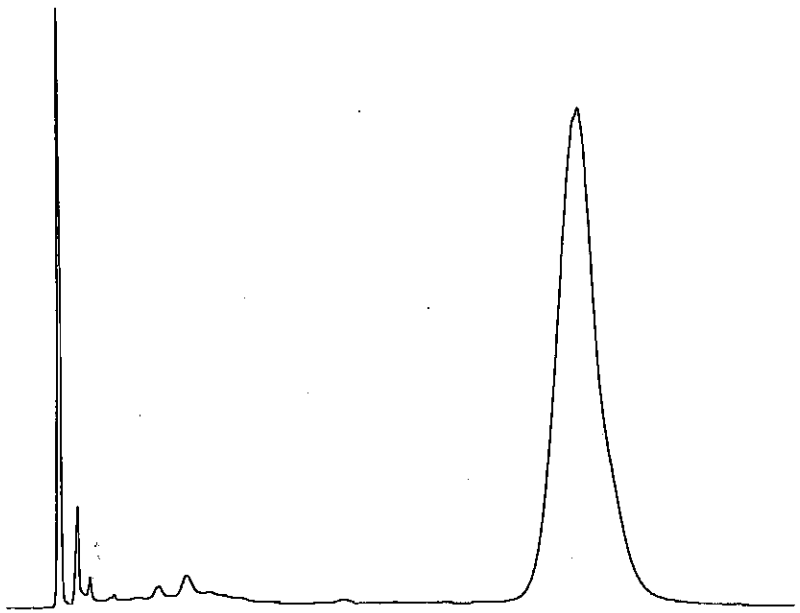


Figure 64

HPLC (iso-45) profile of FPLC fractions #5657

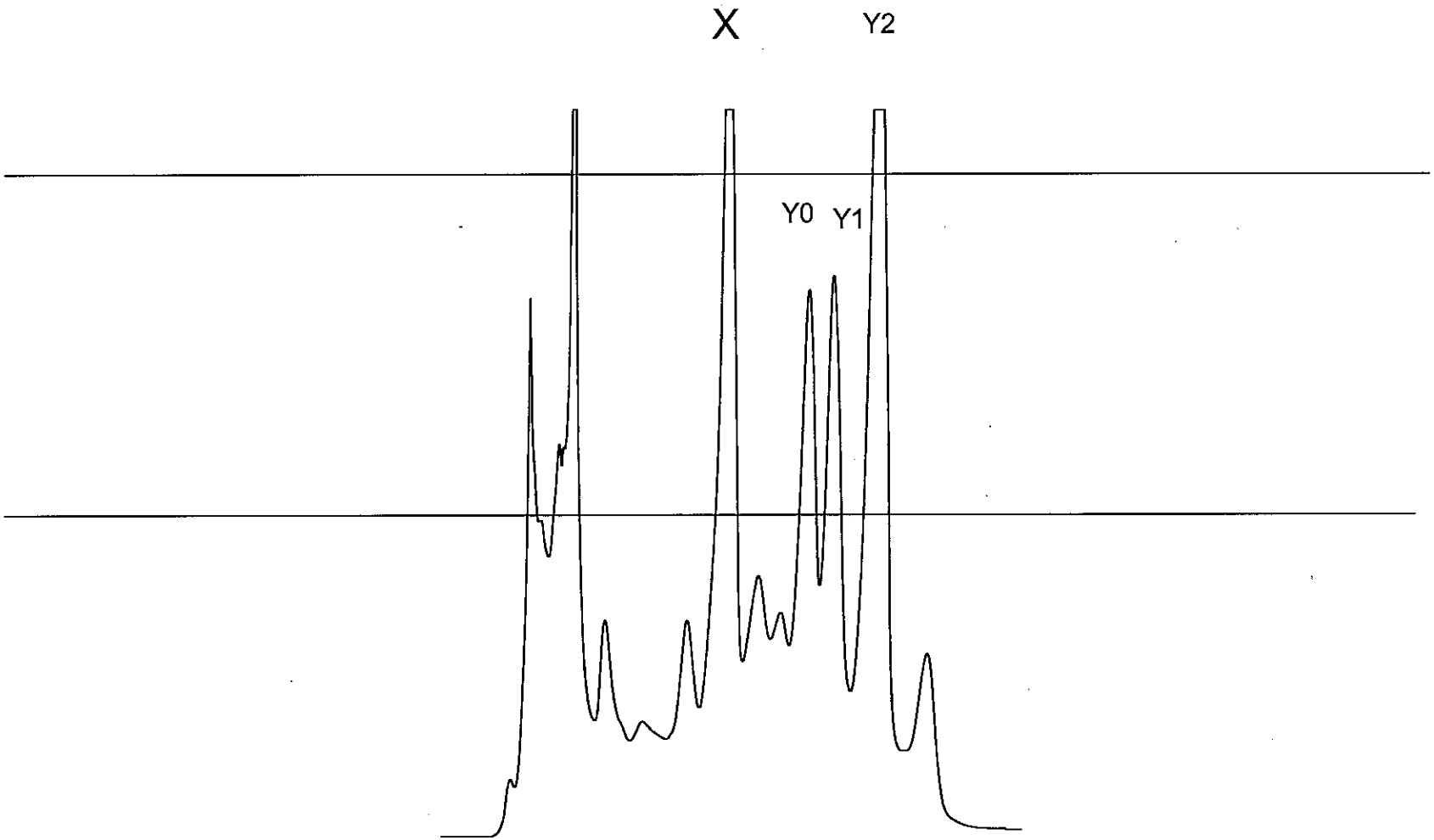


Figure 65

Effect of Compound X on the growth of OCAR-3 cells

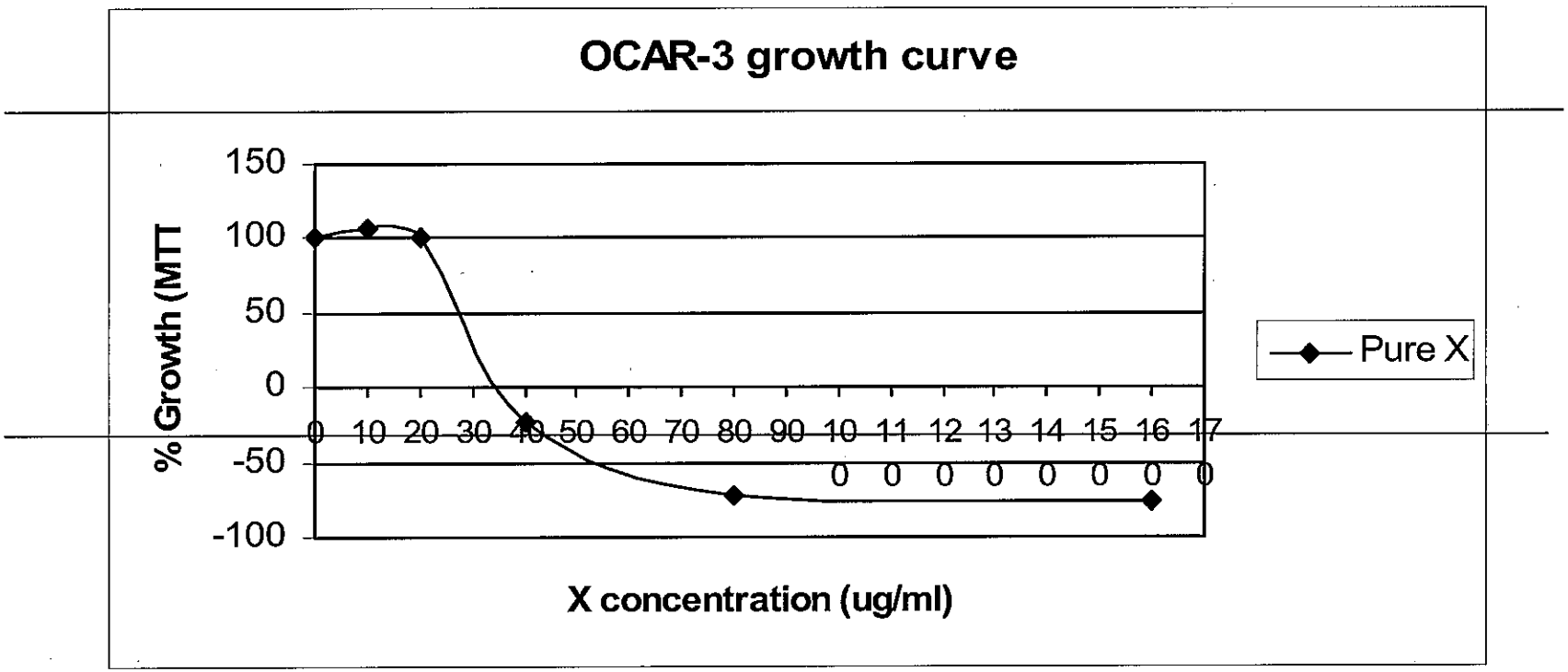


Figure 66
H-NMR of Compound X

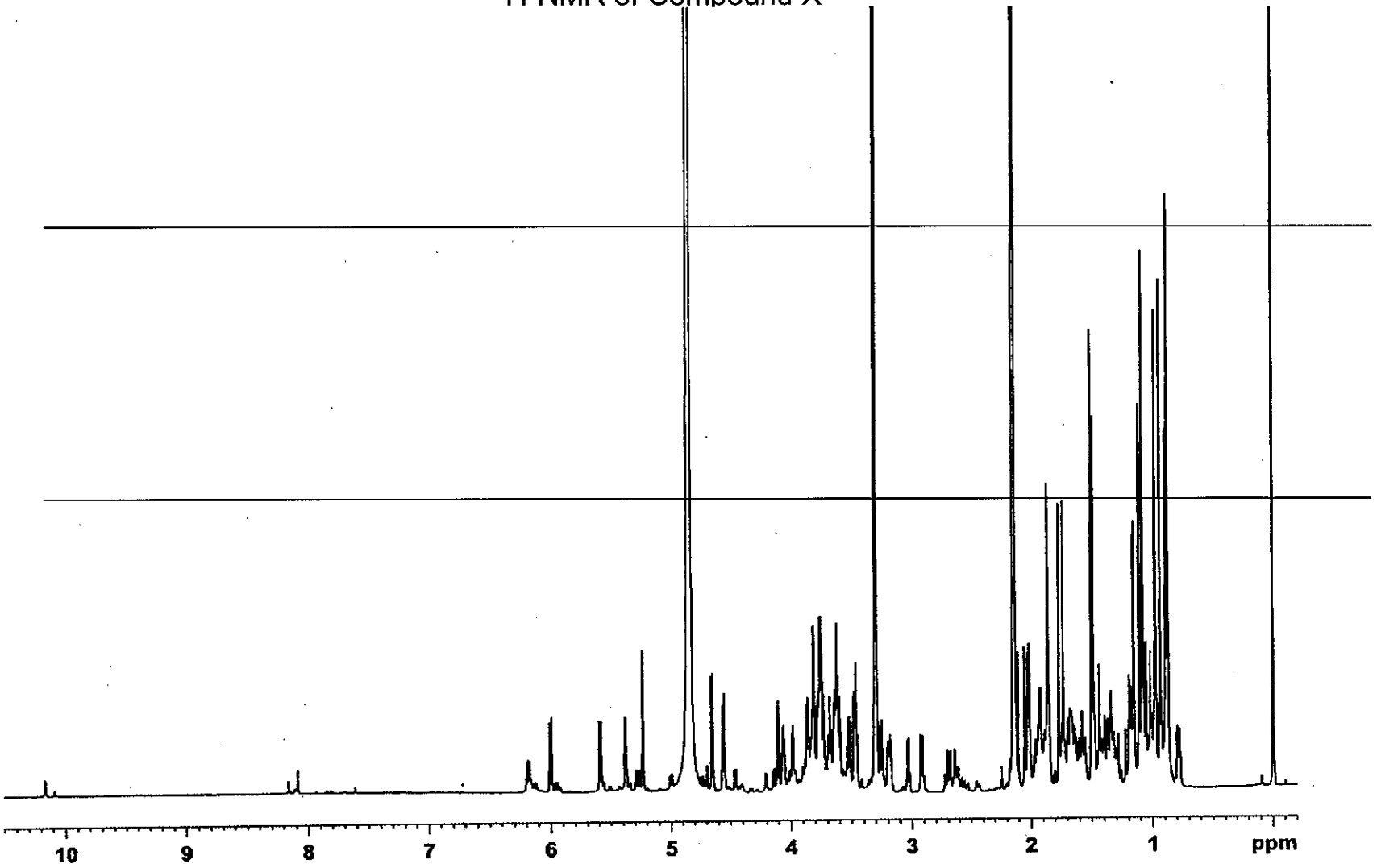


Figure 67
2D NMR (HMQC) of Compound X

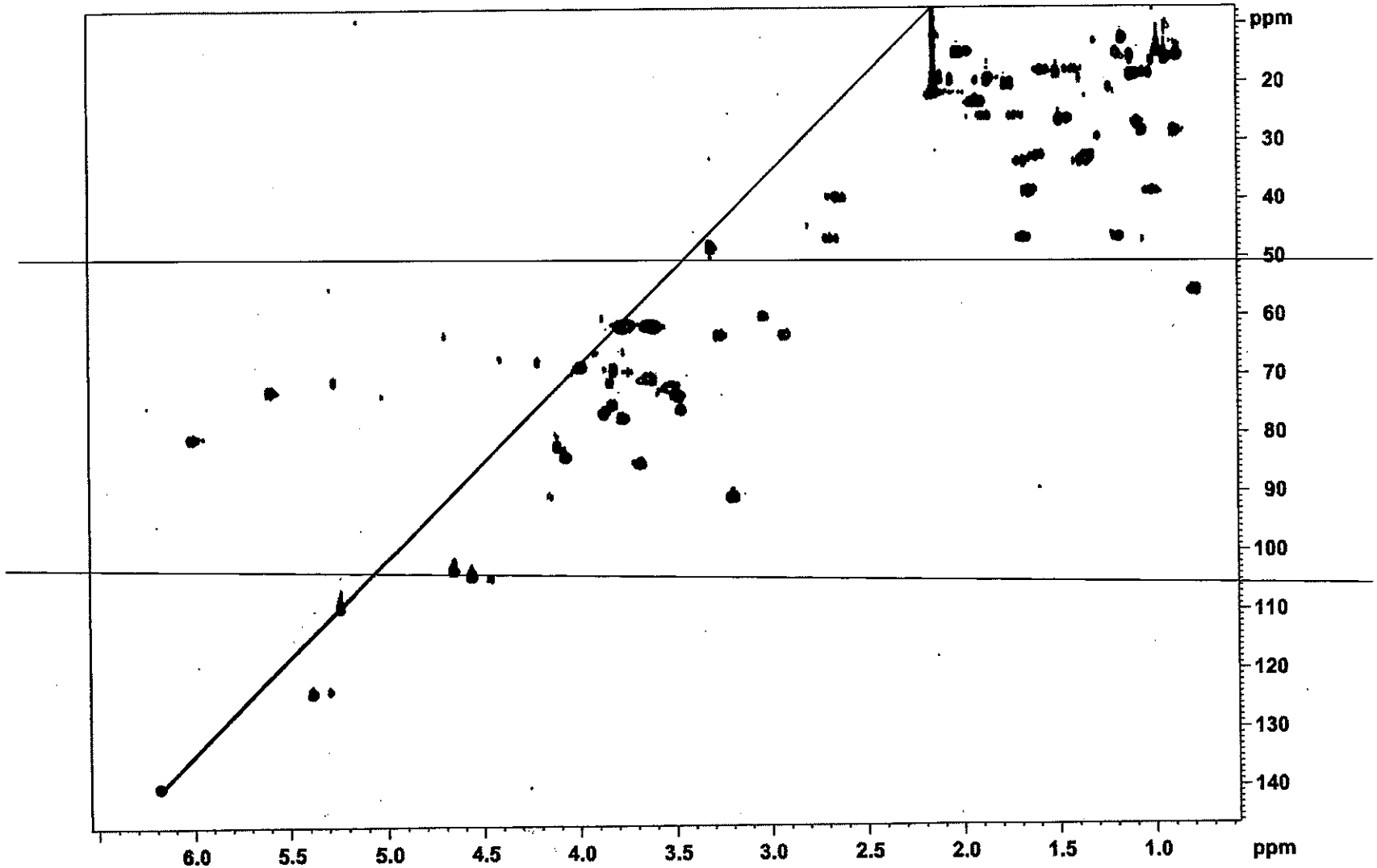


Figure 68
2D NMR (HMBC) of Compound X

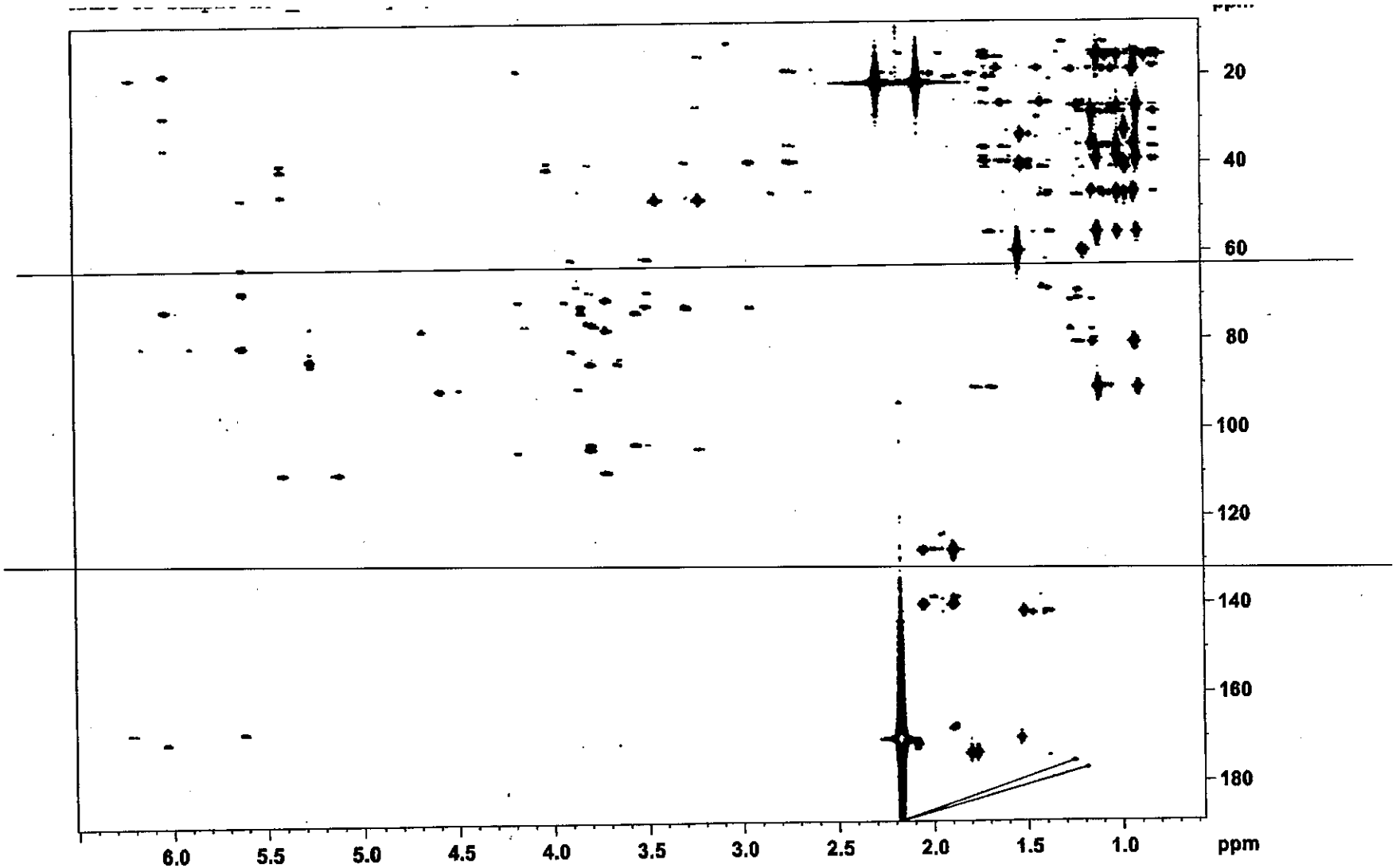


Figure 69
C13-NMR of Compound X

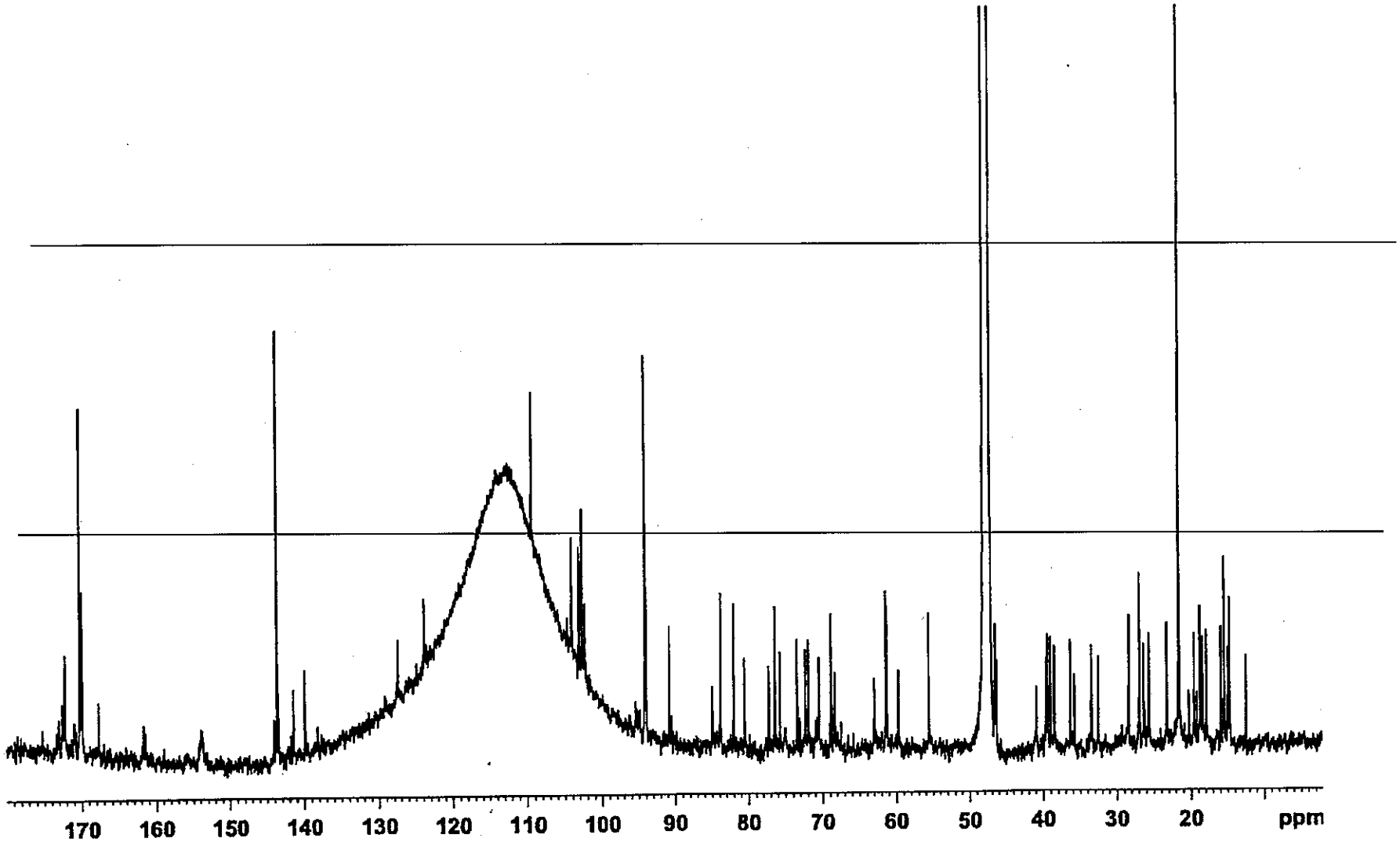


Figure 70
Chemical Structure of Compound X

$C_{58}H_{92}O_{22}$
Exact Mass: 1140.6080
Mol. Wt.: 1141.3379
C, 61.04; H, 8.12; O, 30.84

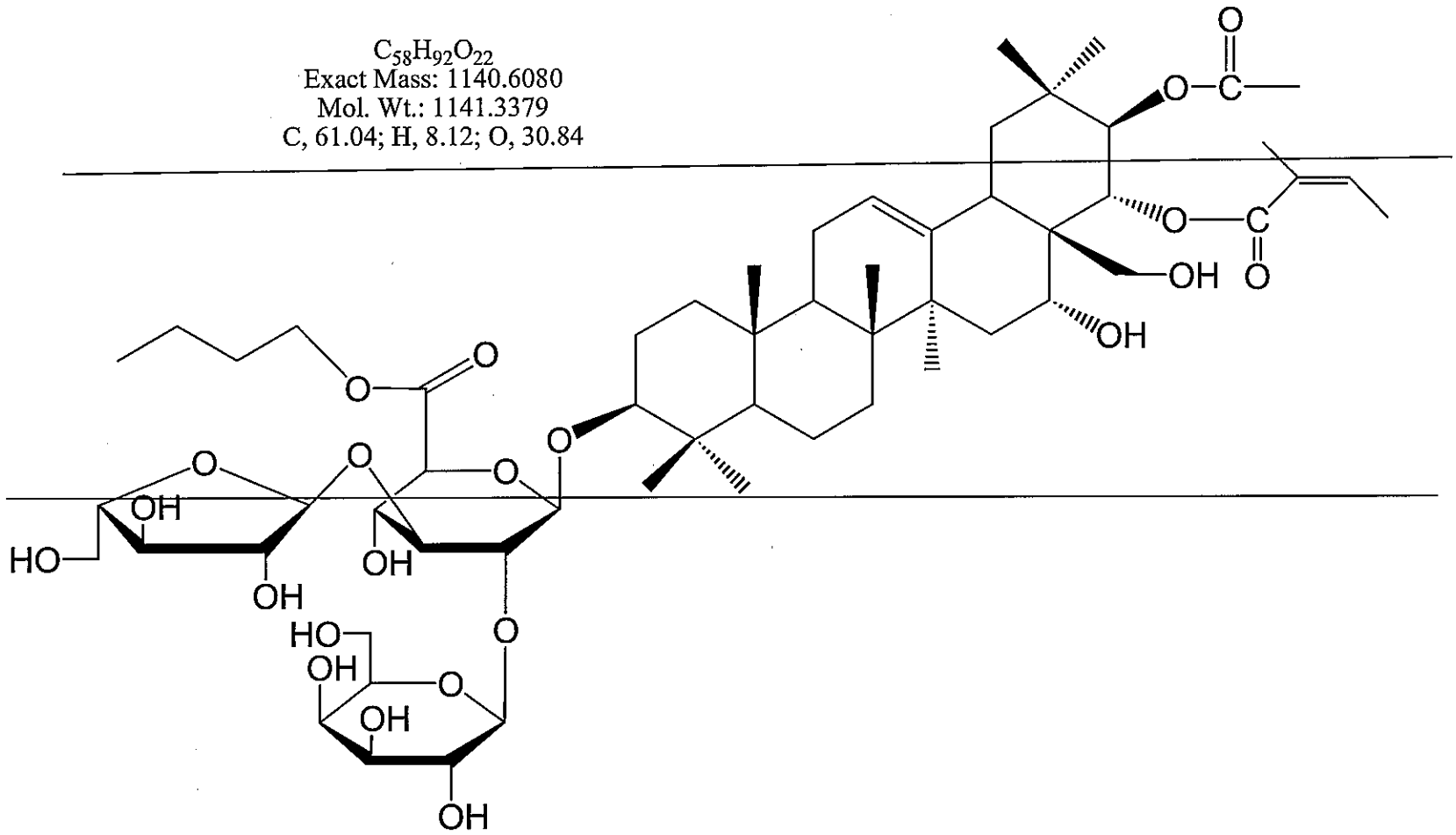


Figure 71

Mass spectrum (WALDI-TOF) of Compound X

X+matrix(CHCA) without calibration

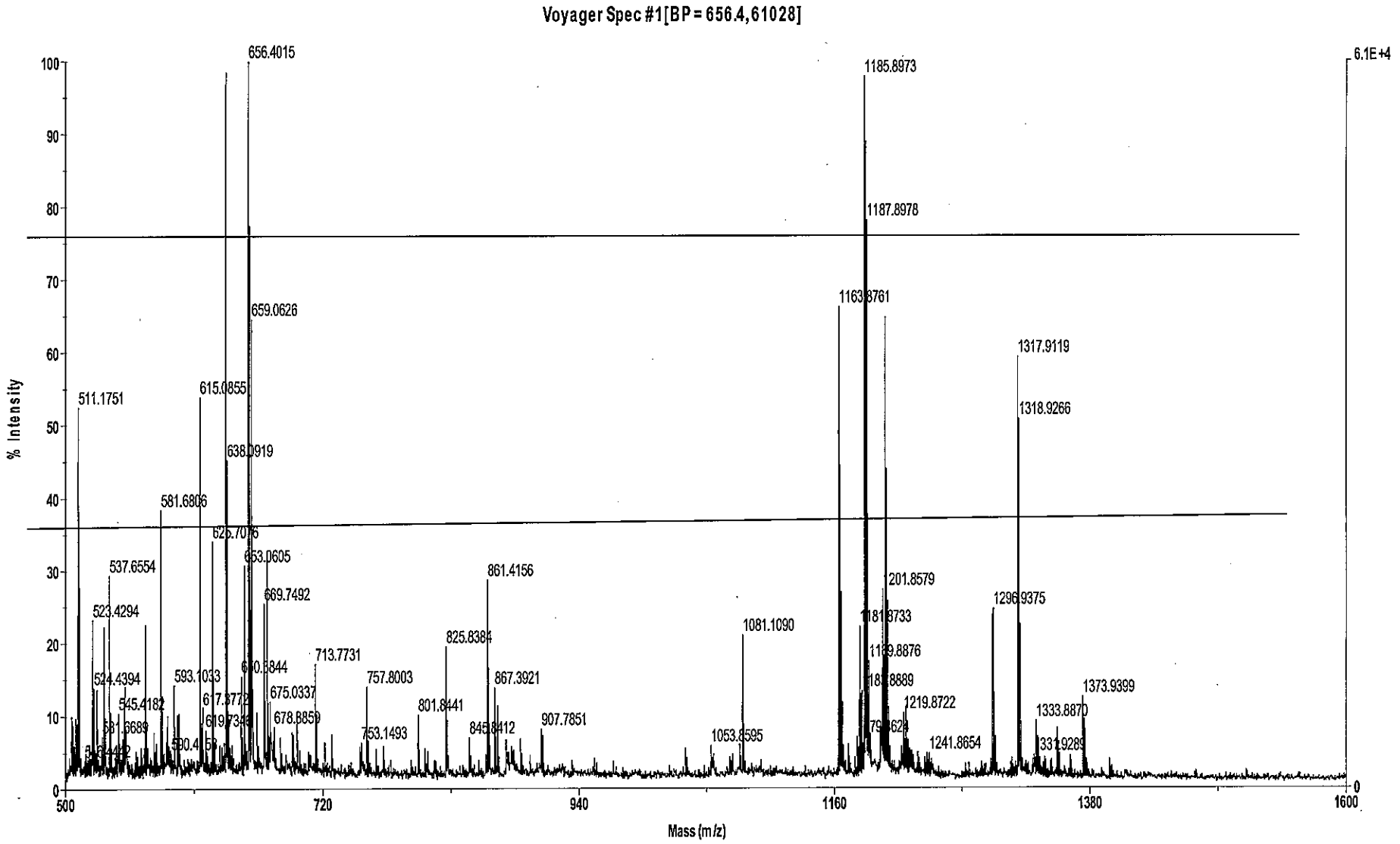
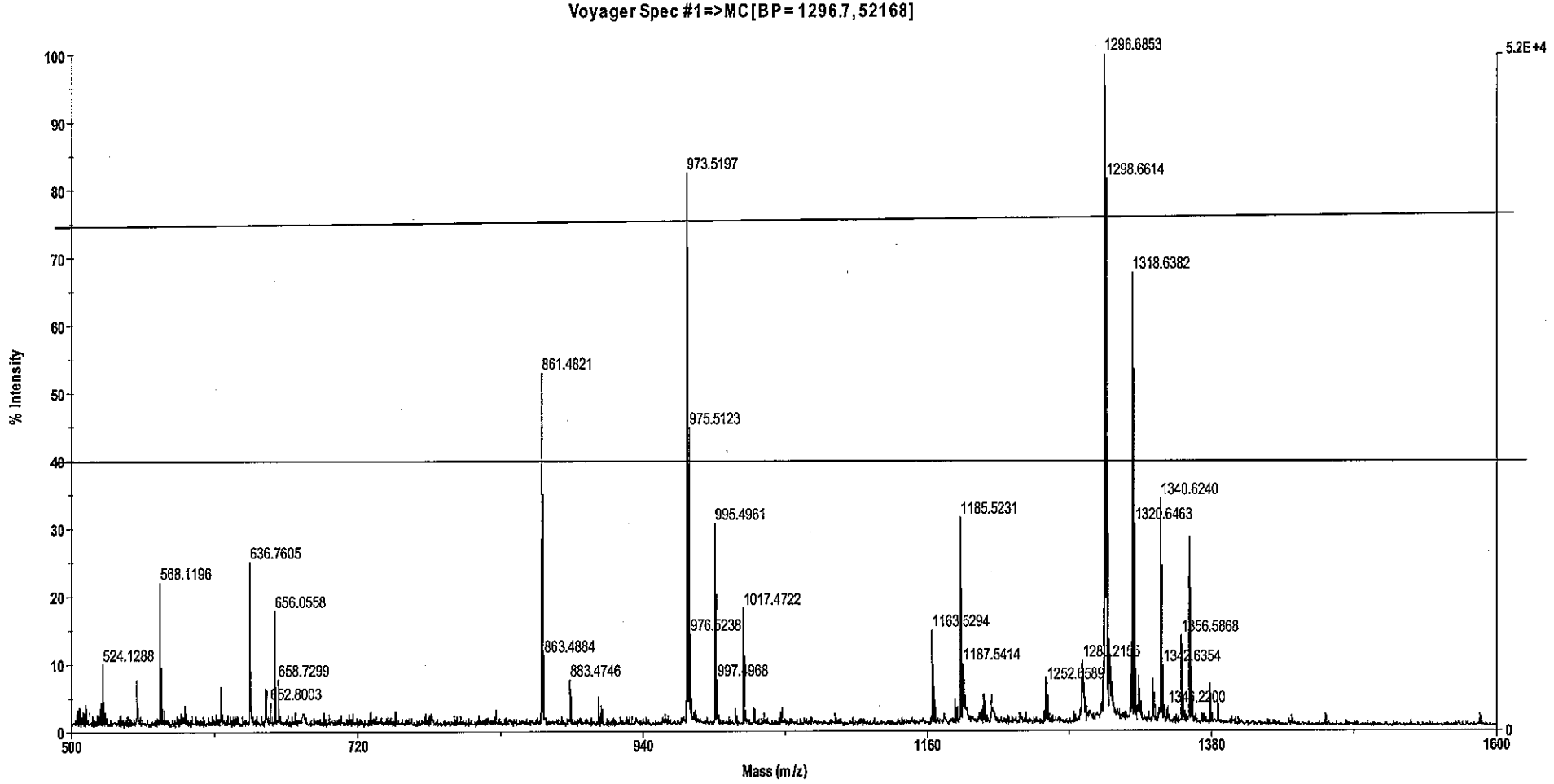


Figure 72

Mass spectrum (WALDI-TOF) of Compound X

X+matrix(CHCA)+Angiotensin after calibration



The mass of the major peak is (a) 1185.5231 and (b) 1163.5294

The theoretical mass of X is 1140.608

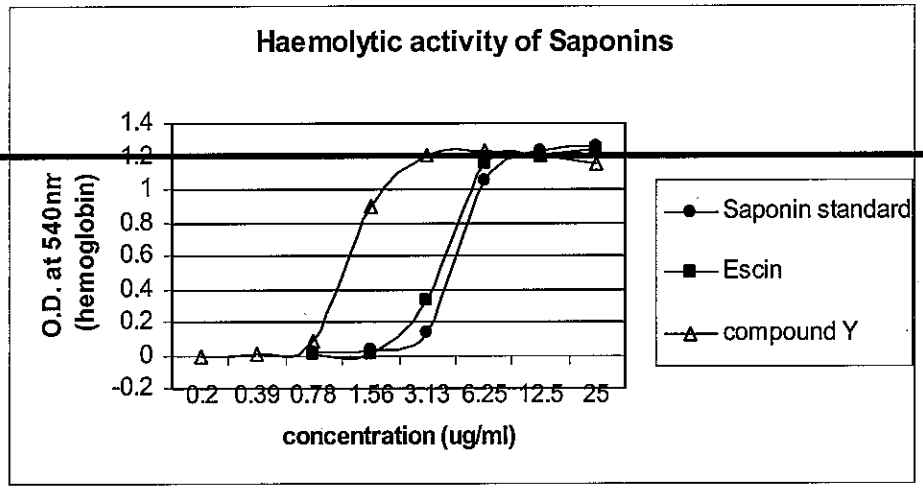
(a) X + 2Na – H =1140.608 + 45.9796 – 1= 1185.5876

(b) X + Na = 1140.608 + 22.9898 = 1163.5978

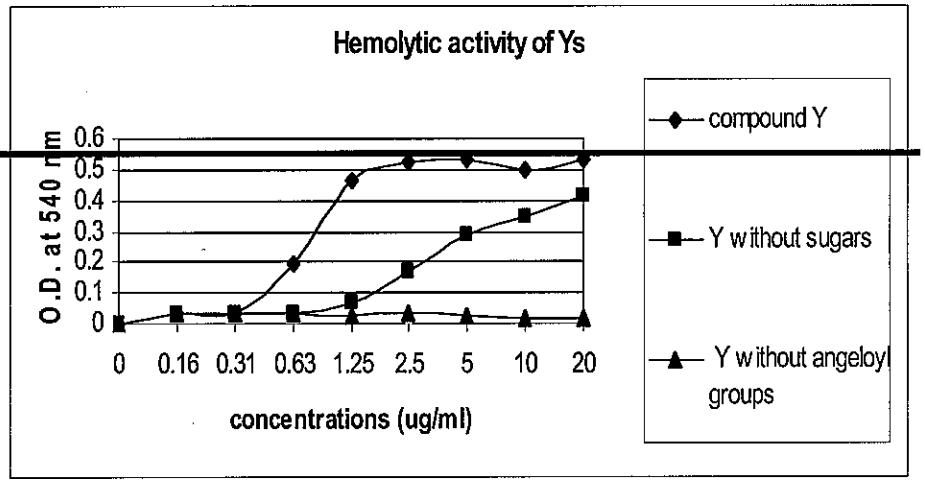
Figure 73

Haemolytic and Mtt activities of Compound Y

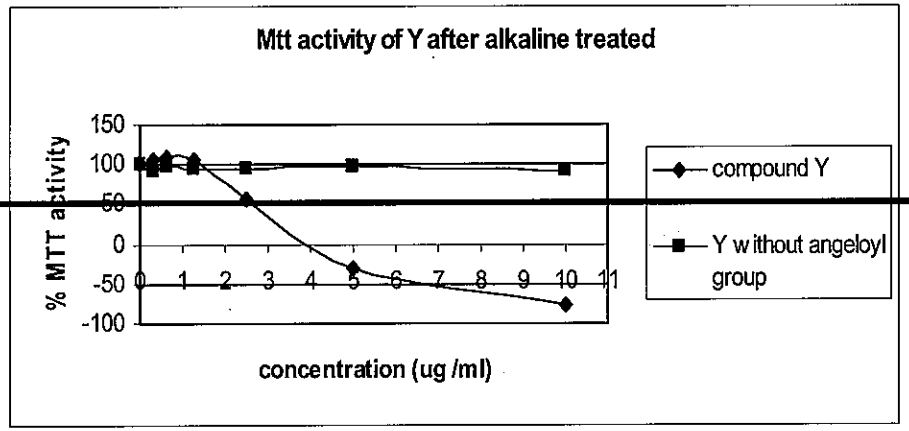
A



B



C



D

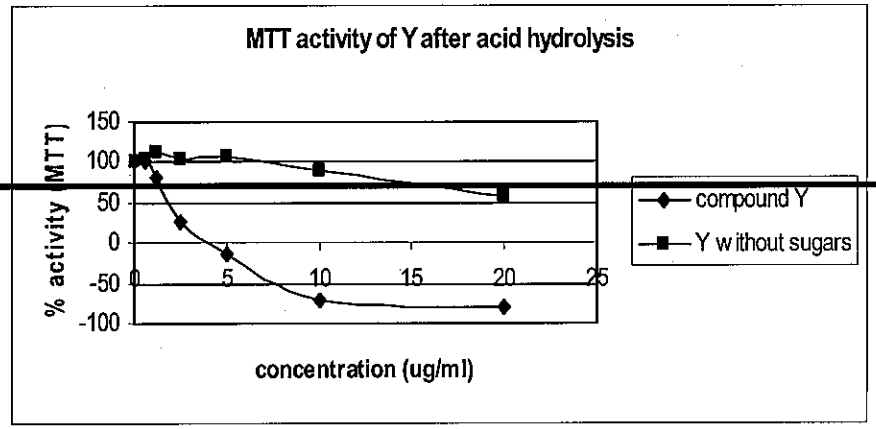


Figure 74

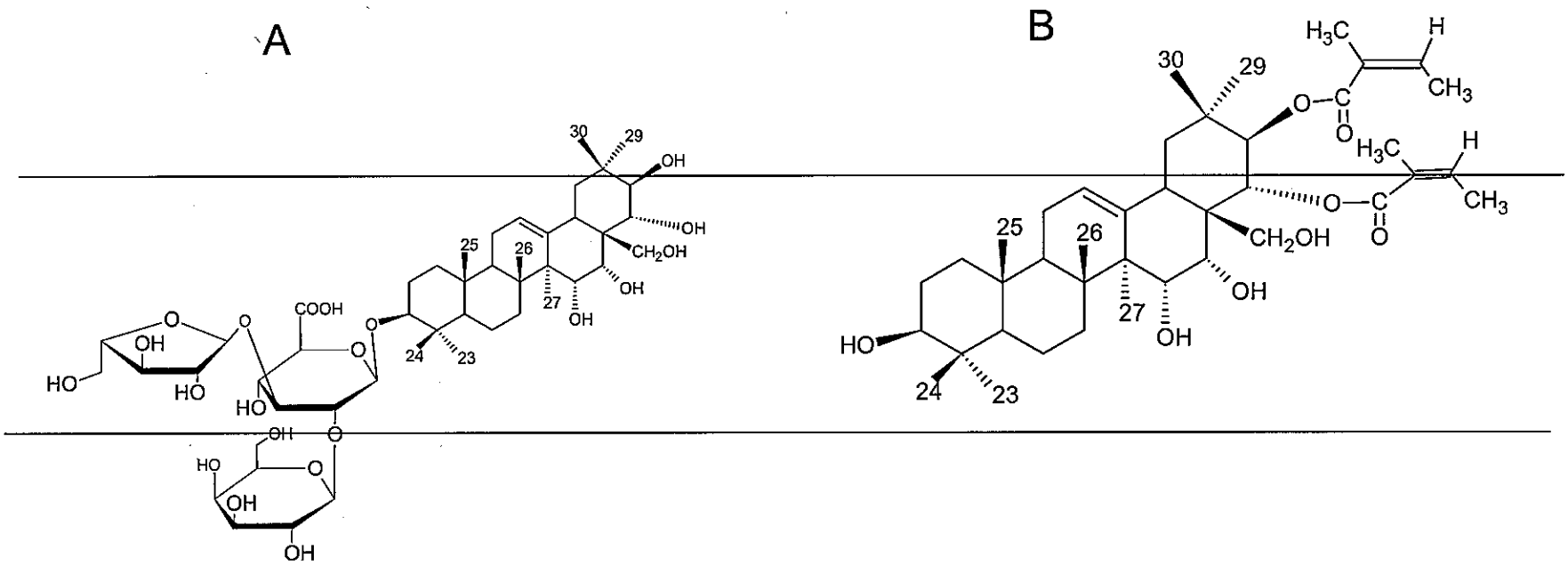
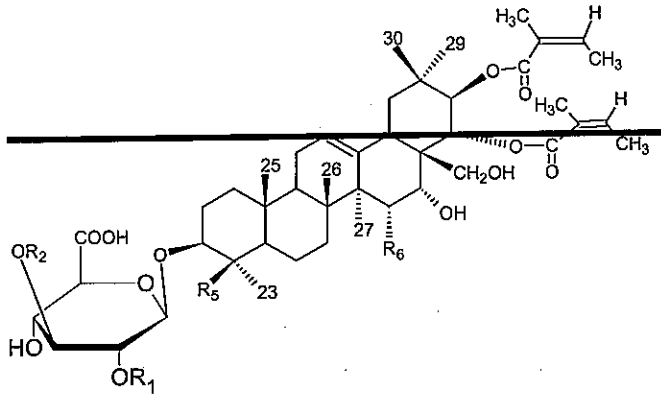
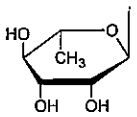
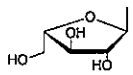
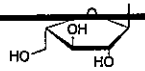
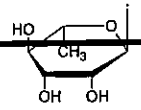
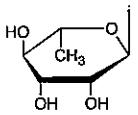
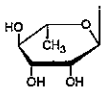
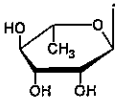
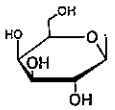
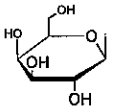
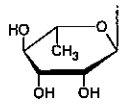
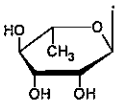
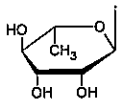

A
B
30
29
OH
OH
CH2OH
OH
OH
25
26
27
24
23
COOH
O
O
O
O
OH
HO
HO
HO
OH
O
HO
O
OH
OH
H3C
H
CH3
O
C
O
H3C
H
CH3
O
C
O
CH2OH
OH
OH
HO
25
26
27
24
23

Figure 75

Structures of Compounds



	R1	R2
9		
10		
11		
12		
13		
14		H
15	H	

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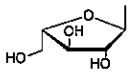
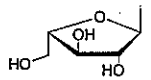
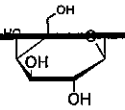
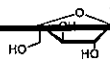
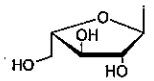
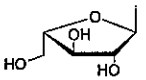
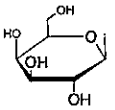
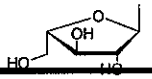
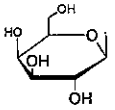
1		
2		
3	H	
4		
5		H
6		
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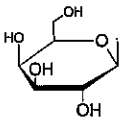
8	H	

R6=H or OH

R5=CH3 or CH2OH

R1= D- glucose or D-galactose or L-rhamnose or L-arabinose or, D-xylose or alduronic acid or D- glucuronic acid or D-galacturonic acid

R2=D- glucose or D-galactose or L-rhamnose or L-arabinose or, D-xylose or alduronic acid or D- glucuronic acid or D-galacturonic acid