

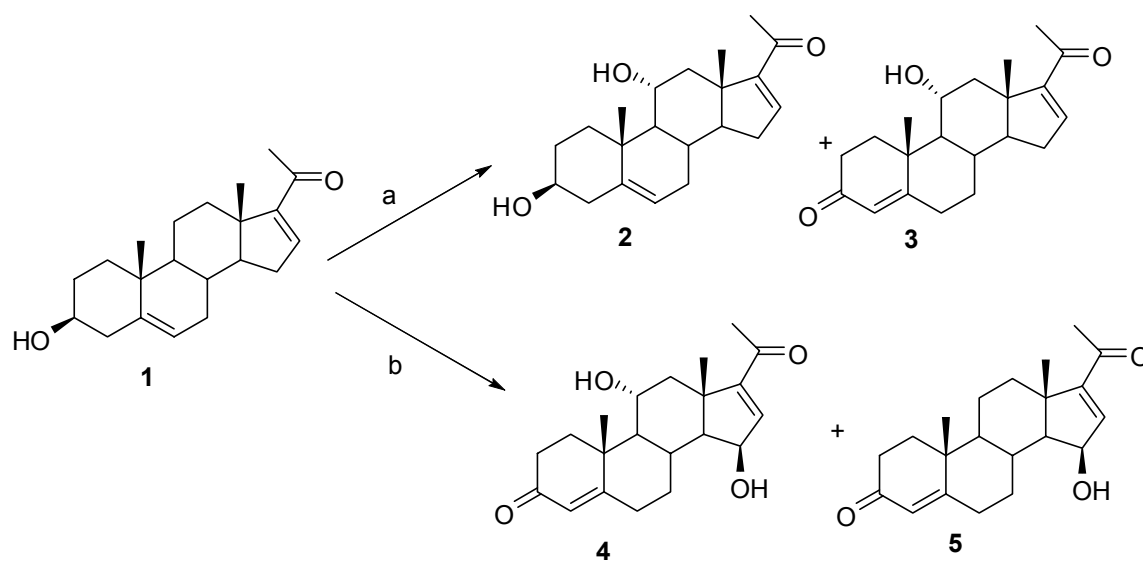
## Supporting Information

### Bio-transformation of FXR antagonist CDRI 80/574<sup>1</sup>

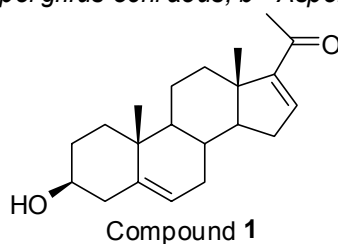
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Raja Roy, Anjani K. Mishra, and Ram Pratap\*

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*a=Aspergillus ochraeus, b= Aspergillus niger*



M.p. 205-207°C,  $M^+ + 1 = 315$ ,  $IR = 1735\text{cm}^{-1}$

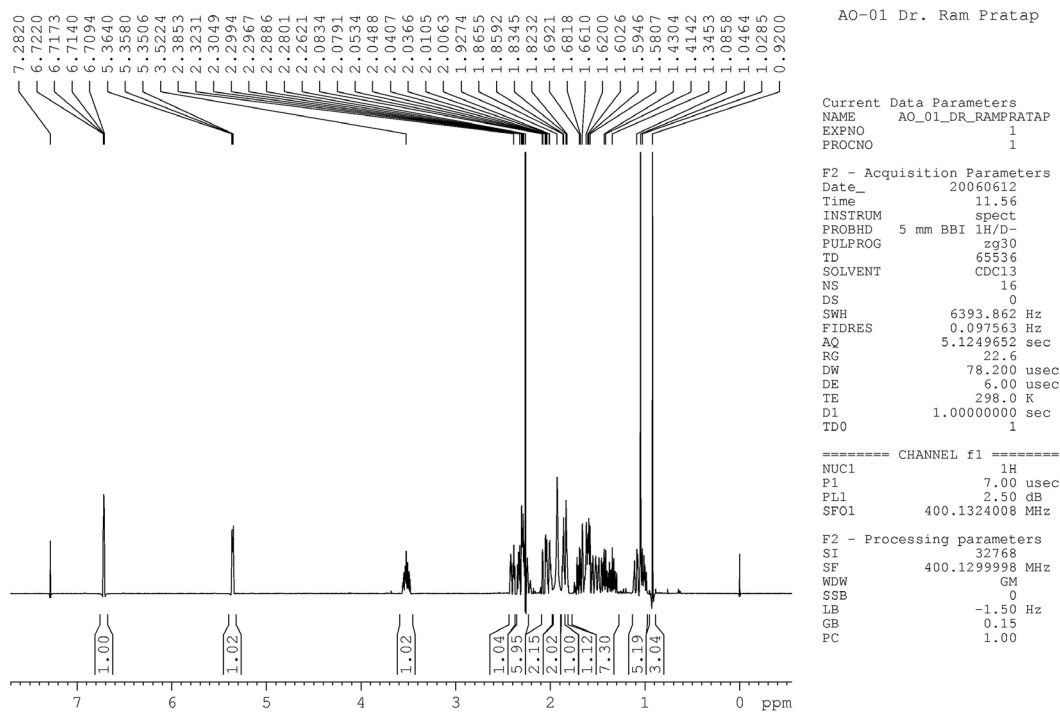
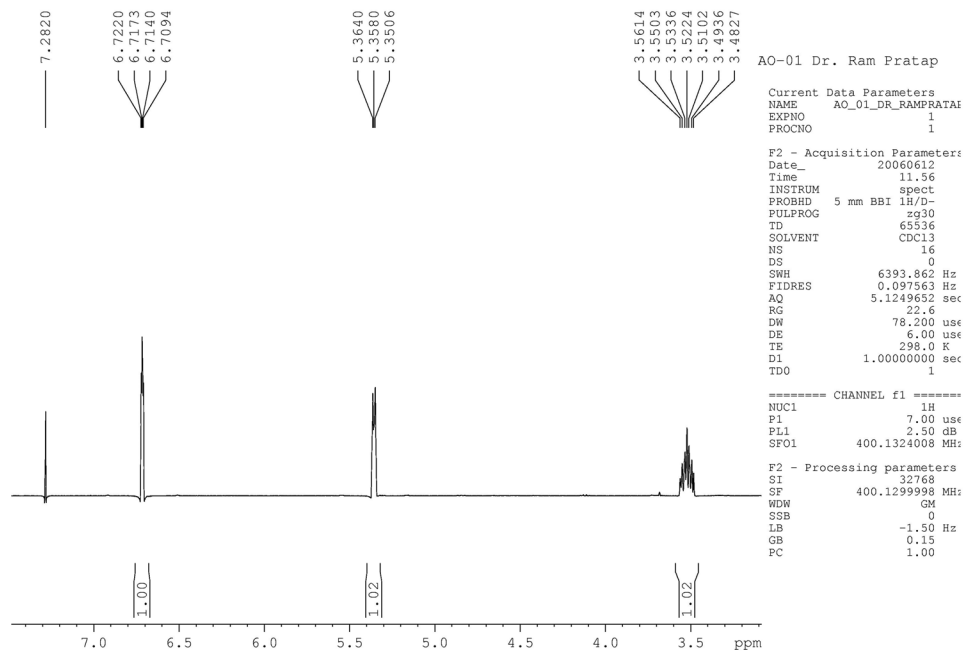
**Scheme**

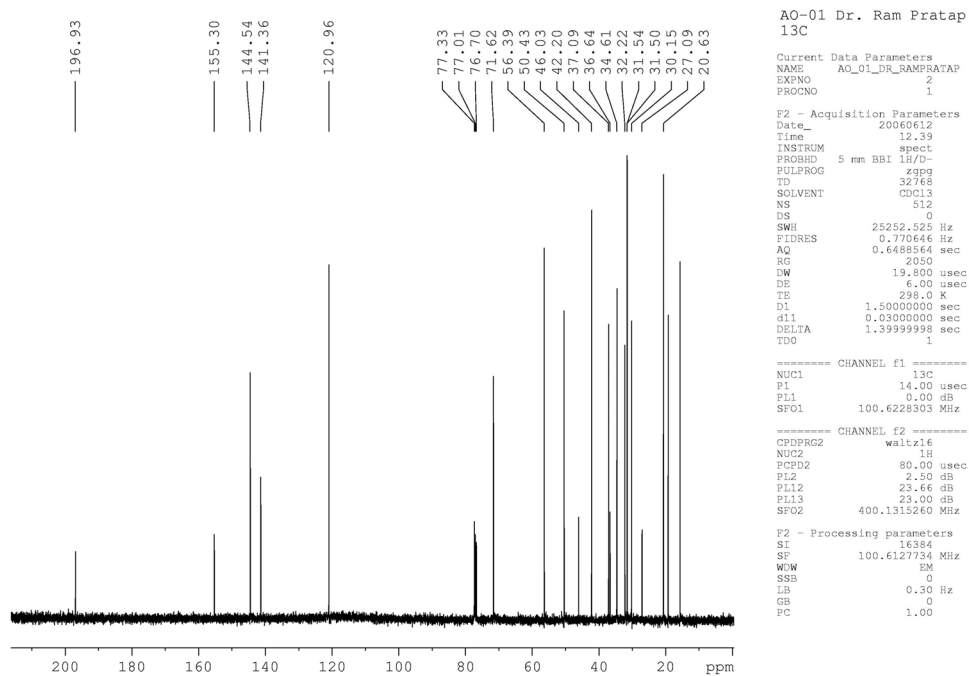
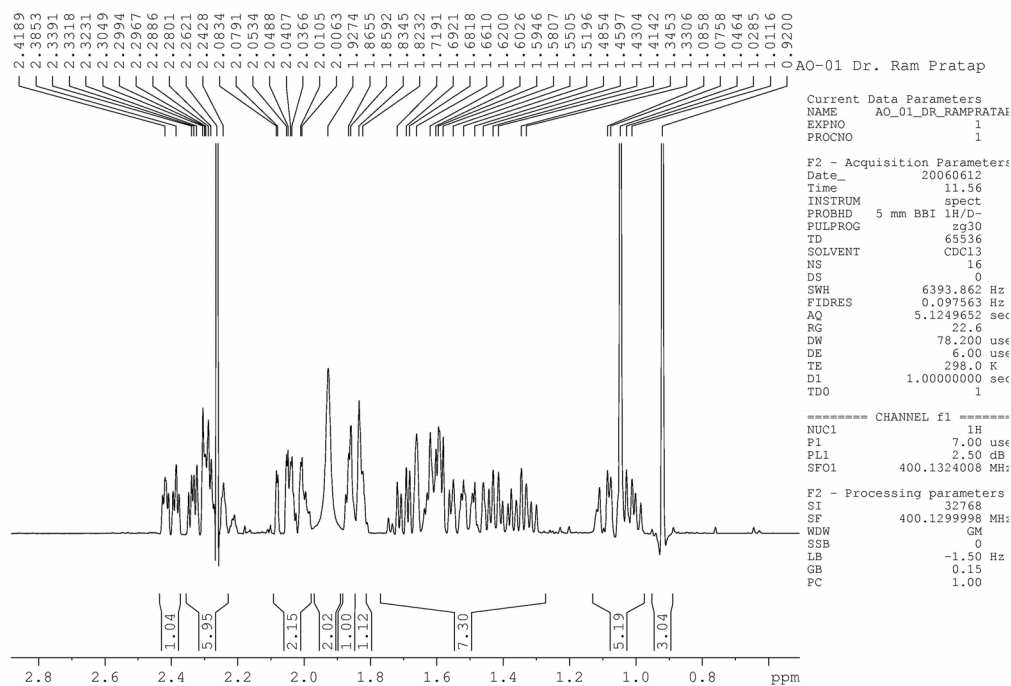
<sup>1</sup> CDRI Communication No. 7614.

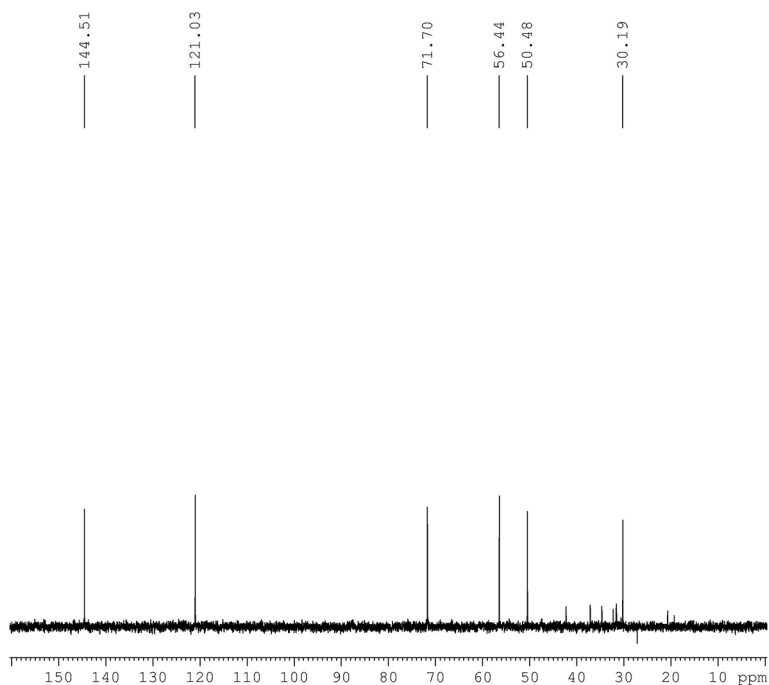
**Table 1.** Complete assignment of protons and carbons of 3 $\beta$ -hydroxy-5,16-dien-pregnane-20-one (CDRI/80-574, **1**) with help of HMBC and HSQC

|     | H    | J (Hz)           | <sup>13</sup> C |                 |
|-----|------|------------------|-----------------|-----------------|
| 1   | 1.08 | m <sup>e</sup>   | 37.1            | CH <sub>2</sub> |
| 1'  | 1.85 | m <sup>e</sup>   |                 |                 |
| 2   | 1.84 | m <sup>e</sup>   | 31.5            | CH <sub>2</sub> |
| 2'  | 1.5  | m <sup>e</sup>   |                 |                 |
| 3   | 3.52 | m <sup>e</sup>   | 71.7            | CH              |
| 4   | 2.27 | m <sup>e</sup>   | 42.2            | CH <sub>2</sub> |
| 5   | -    | -                | 141.4           | C               |
| 6   | 5.36 | d(5.6)           | 121             | CH              |
| 7   | 1.99 | m <sup>e</sup>   | 31.5            | CH <sub>2</sub> |
| 7'  | 1.6  | m <sup>e</sup>   |                 |                 |
| 8   | 1.7  | m <sup>e</sup>   | 30.2            | CH              |
| 9   | 1.01 | m <sup>e</sup>   | 50.4            | CH              |
| 10  | -    | -                | 36.6            | C               |
| 11  | 1.59 | m <sup>e</sup>   | 20.7            | CH <sub>2</sub> |
| 11' | 1.34 | m <sup>e</sup>   |                 |                 |
| 12  | 2.4  | td(3.1,<br>12.8) | 34.6            | CH <sub>2</sub> |
| 12' | 1.34 | m <sup>e</sup>   |                 |                 |
| 13  | -    | -                | 46              | C               |
| 14  | 1.42 | m <sup>e</sup>   | 56.4            | CH              |
| 15  | 2.32 | m <sup>e</sup>   | 32.2            | CH <sub>2</sub> |
| 15' | 2.05 | m <sup>e</sup>   |                 |                 |
| 16  | 6.72 | dd(1.8,3.2)      | 144.5           | CH              |
| 17  | -    | -                | 155.3           | C               |
| 18  | 0.92 | s                | 15.7            | CH <sub>3</sub> |
| 19  | 1.04 | s                | 19.3            | CH <sub>3</sub> |
| 20  | -    | -                | 196.9           | C               |
| 21  | 2.26 | s                | 27.1            | CH <sub>3</sub> |

m<sup>e</sup>: multiplet overlapped with other signals, s: singlet, brs: broad singlet, d: doublet, td: triplet of a doublet, dd: doublet of a doublet.

 $^1\text{H}$  NMR Compound 1

 $^{13}\text{C}$  of Compound 1



AO-01  
C13DEPT90 CDC13 1

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EXPNO 71  
PROCNO 1

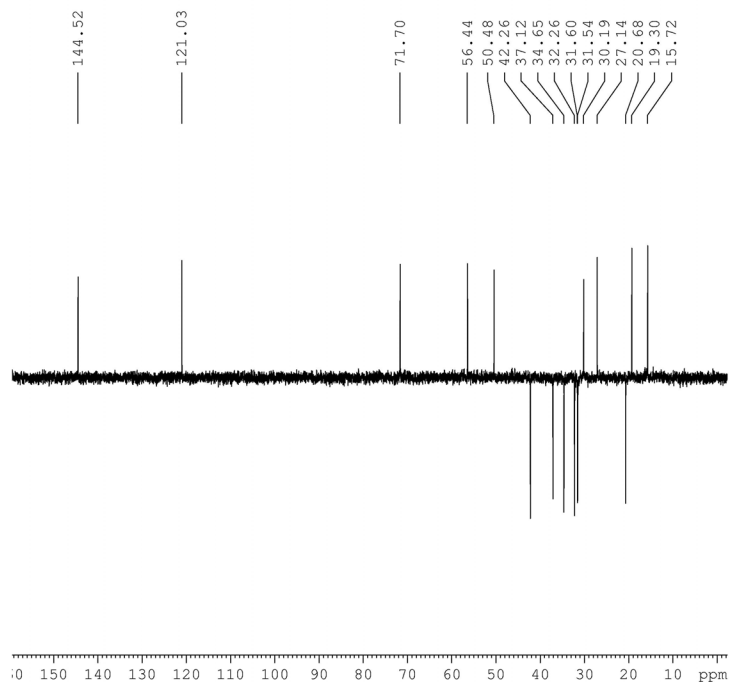
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SOLVENT CDC13  
NS 31  
DS 0  
SWH 18028.846 Hz  
FIDRES 0.275038 Hz  
AQ 1.8175818 sec  
RG 2050  
DW 27.733 usec  
DE 6.00 usec  
TE 298.7 K  
CNST2 145.000000  
D1 2.00000000 sec  
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DELTA 0.00001108 sec  
TDO 1

----- CHANNEL f1 -----  
NUC1 13C  
P1 8.70 usec  
P2 17.40 usec  
PL1 -3.00 dB  
SFO1 75.4752953 MHz

----- CHANNEL f2 -----  
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NUC2 1H  
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PL2 -1.00 dB  
PL12 17.00 dB  
SFO2 300.1312005 MHz

F2 - Processing parameters  
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Dept 90 of Compound 1



AO-01  
C13DEPT135 CDC13 1

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EXPNO 70  
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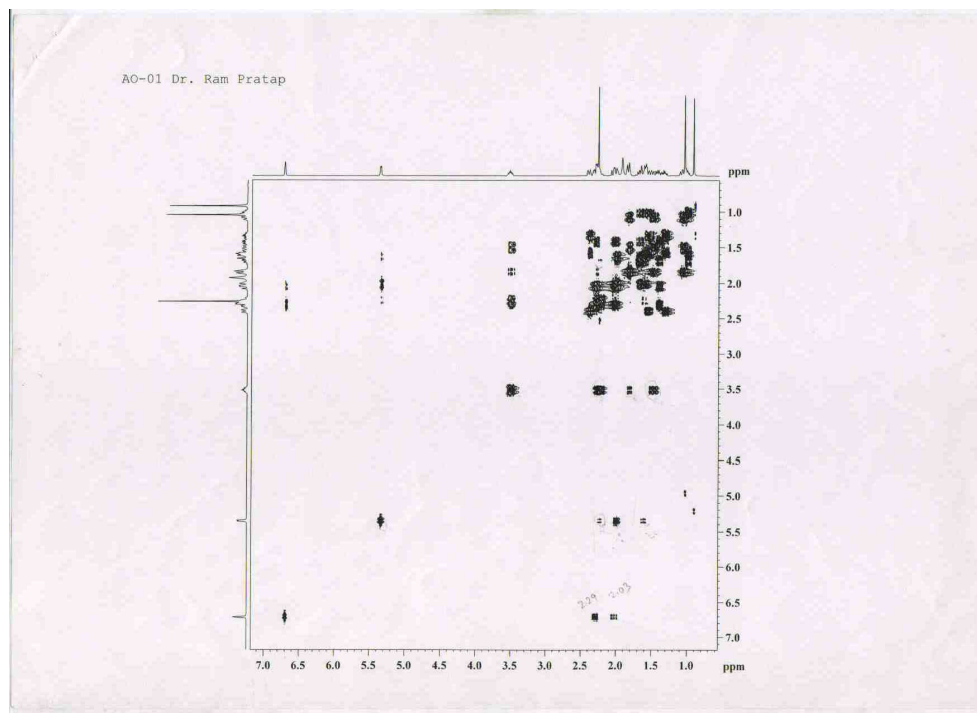
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TD 65536  
SOLVENT CDC13  
NS 107  
DS 0  
SWH 18028.846 Hz  
FIDRES 0.275038 Hz  
AQ 1.8175818 sec  
RG 2050  
DW 27.733 usec  
DE 6.00 usec  
TE 298.6 K  
CNST2 145.000000  
D1 2.00000000 sec  
d2 0.00344828 sec  
d12 0.00002000 sec  
DELTA 0.00001108 sec  
TDO 1

----- CHANNEL f1 -----  
NUC1 13C  
P1 8.70 usec  
P2 17.40 usec  
PL1 -3.00 dB  
SFO1 75.4752953 MHz

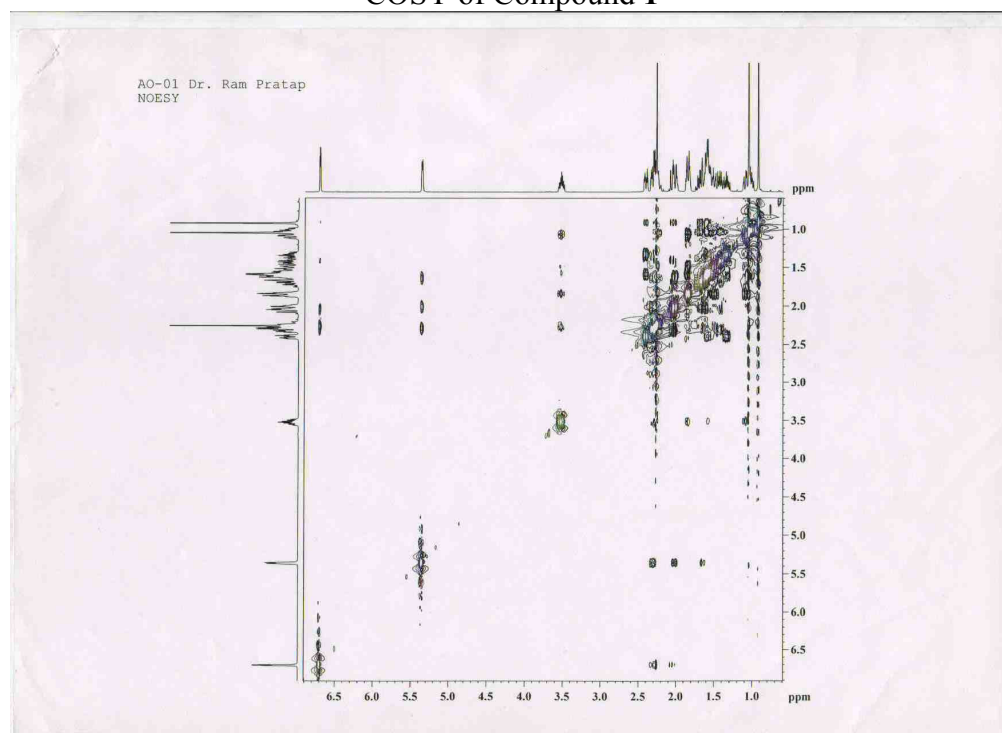
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P3 11.60 usec  
P4 23.20 usec  
PCPD2 80.00 usec  
PL2 -1.00 dB  
PL12 17.00 dB  
SFO2 300.1312005 MHz

F2 - Processing parameters  
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SF 75.4677490 MHz  
WDW EM  
SSB 0  
LB 1.00 Hz  
GB 0  
PC 1.40

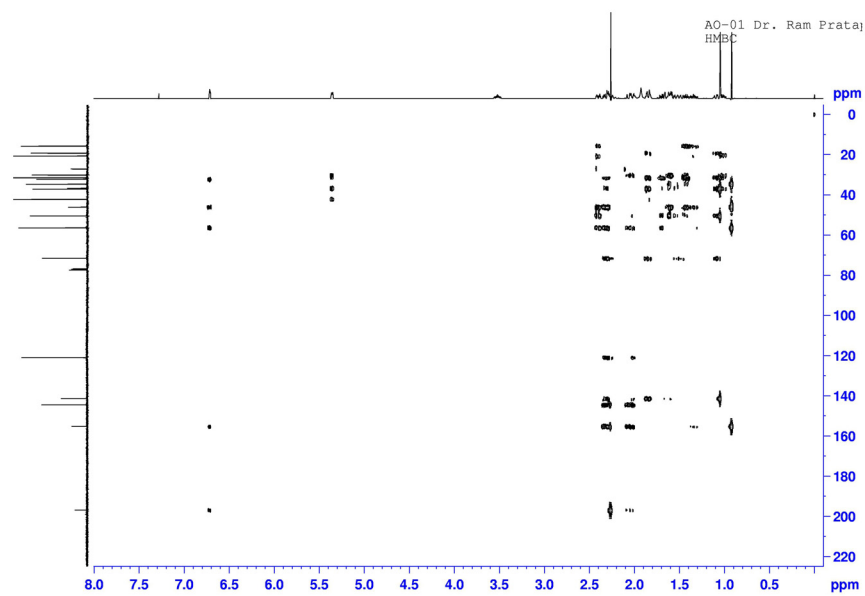
Dept 135 of Compound 1



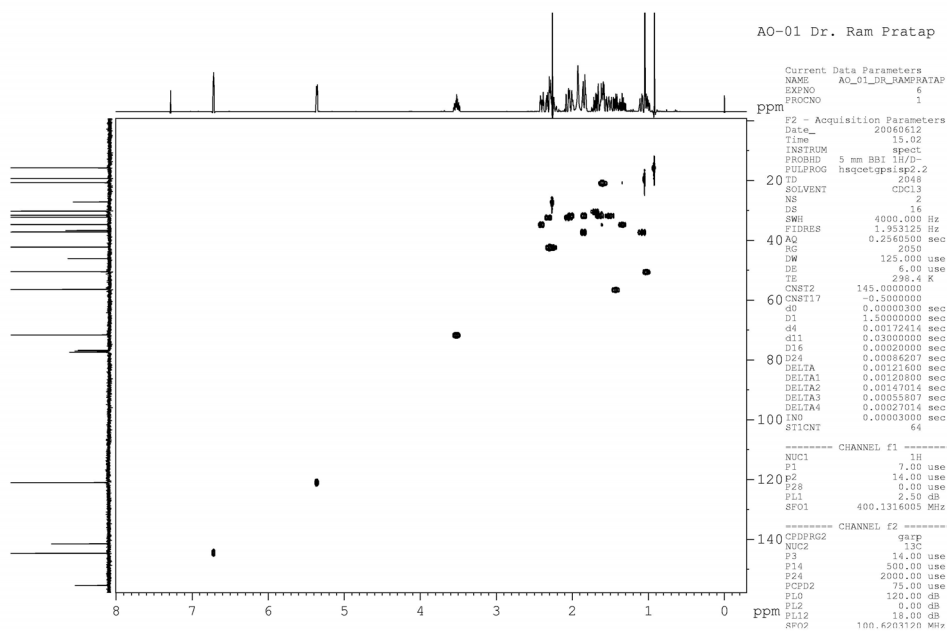
COSY of Compound 1



NOESY of Compound 1

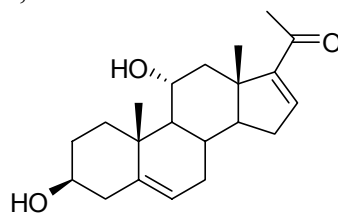


HMBC of Compound 1



HSQC of Compound 1

11 $\alpha$ -Hydroxy-4, 16-dien-pregnan-3, 20-dione **2**:



Compound **2**

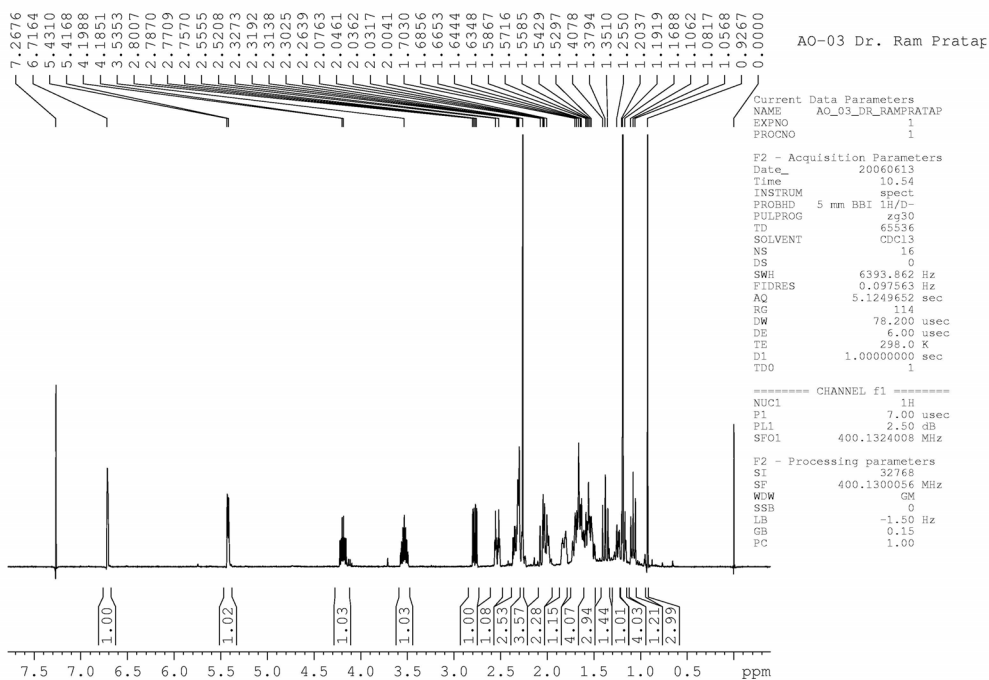
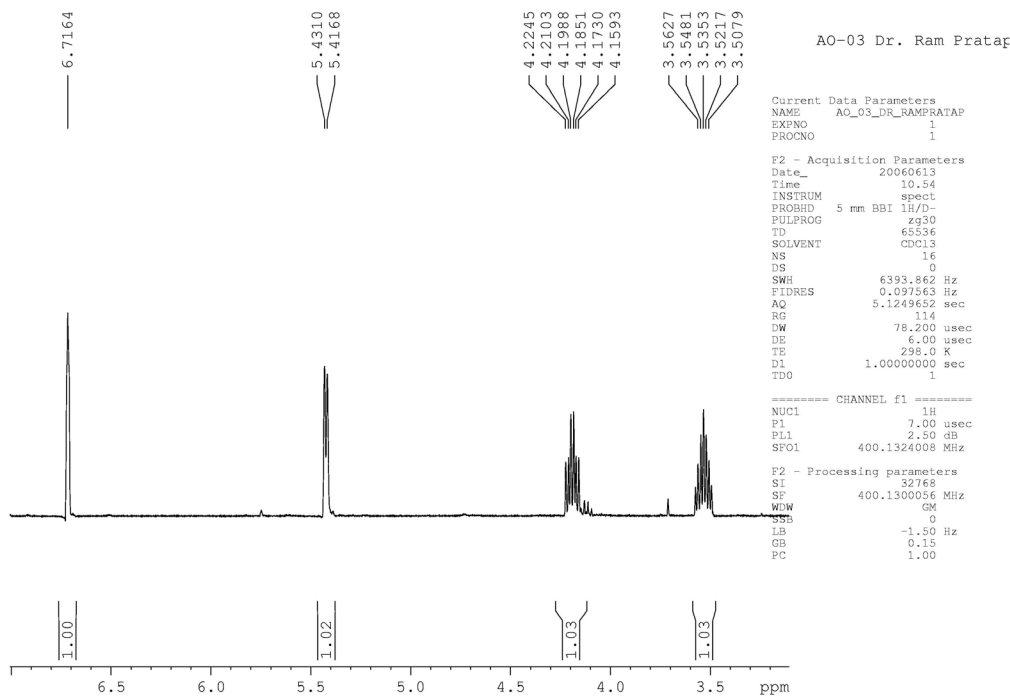
M.P. 162<sup>0</sup>C, M<sup>+</sup>+1(328); IR (KBr): 1661 cm<sup>-1</sup>;

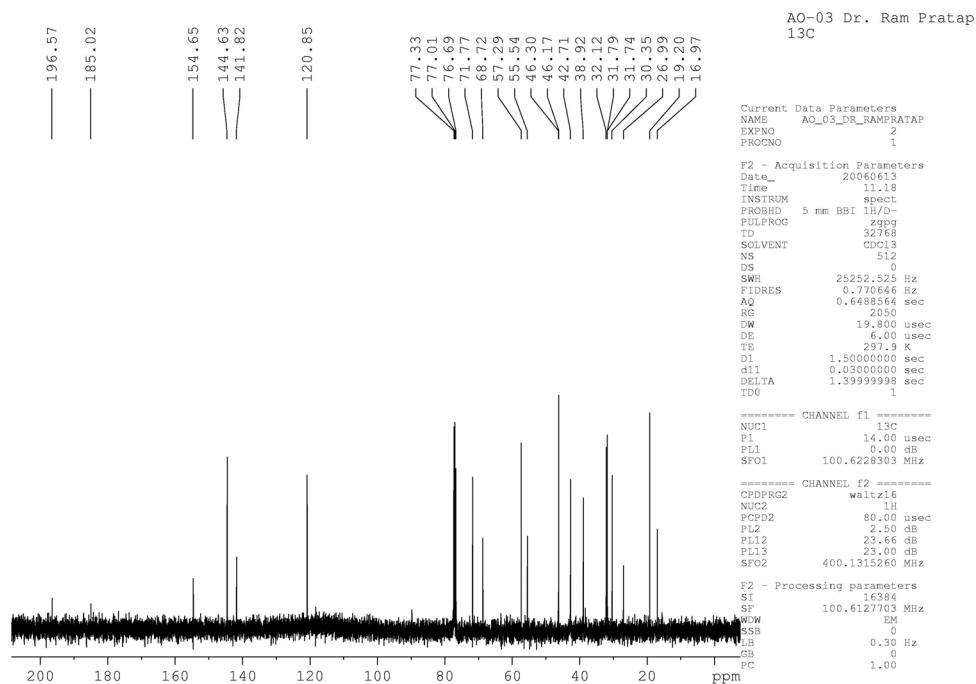
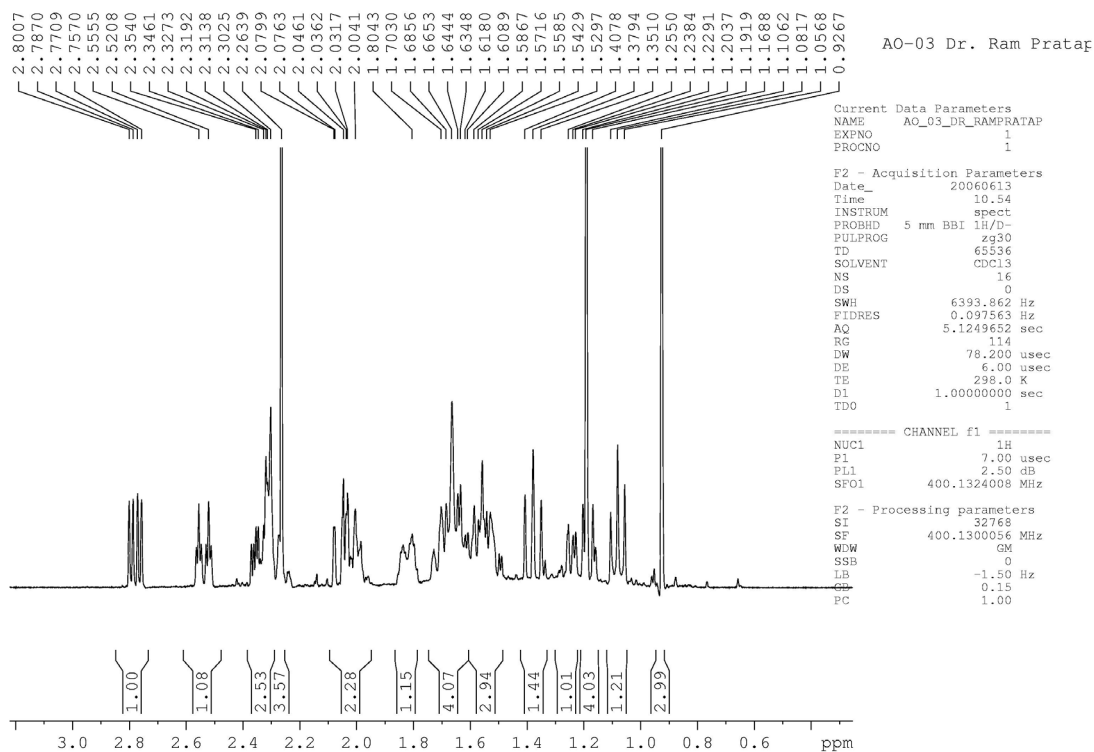
**Table 2.** Complete assignment of proton and carbon signals of 3 $\beta$ ,11 $\alpha$ -dihydroxy-5,6-dien-pregnane-20-one **2** with help of HSQC and HMBC

|     | H    | J (Hz)         | <sup>13</sup> C |                 |
|-----|------|----------------|-----------------|-----------------|
| 1   | 2.54 | td (3.5, 13.6) | 38.9            | CH <sub>2</sub> |
| 1'  | 1.19 | m <sup>e</sup> |                 |                 |
| 2   | 1.82 | m <sup>e</sup> | 31.7            | CH <sub>2</sub> |
| 2'  | 1.54 | m <sup>e</sup> |                 |                 |
| 3   | 3.53 | m <sup>e</sup> | 71.8            | CH              |
| 4   | 2.29 | m <sup>e</sup> | 42.7            | CH <sub>2</sub> |
| 5   | -    | -              | 141.8           | C               |
| 6   | 5.43 | d(5.6)         | 120.9           | CH              |
| 7   | 2.02 | m <sup>e</sup> | 31.7            | CH <sub>2</sub> |
| 7'  | 1.7  | m <sup>e</sup> |                 |                 |
| 8   | 1.65 | m <sup>e</sup> | 30.3            | CH              |
| 9   | 1.08 | t (9.8)        | 57.3            | CH              |
| 10  | -    | -              | 38.3            | C               |
| 11  | 4.18 | td( 5.7, 10.3) | 68.7            | CH <sub>2</sub> |
| 12  | 2.78 | dd(5.5, 11.9)  | 46.1            | CH <sub>2</sub> |
| 12' | 1.38 | t( 11.3)       |                 |                 |
| 13  | -    | -              | 46.3            | C               |
| 14  | 1.55 | m <sup>e</sup> | 55.5            | CH              |
| 15  | 2.33 | m <sup>e</sup> | 32.1            | CH <sub>2</sub> |
| 15' | 2.05 | m <sup>e</sup> |                 |                 |
| 16  | 6.72 | dd(1.8,3.2)    | 144.6           | CH              |
| 17  | -    | -              | 154.6           | C               |
| 18  | 0.93 | s              | 16.9            | CH <sub>3</sub> |
| 19  | 1.19 | s              | 19.2            | CH <sub>3</sub> |
| 20  | -    | -              | 196.6           | C               |
| 21  | 2.26 | s              | 26.9            | CH <sub>3</sub> |

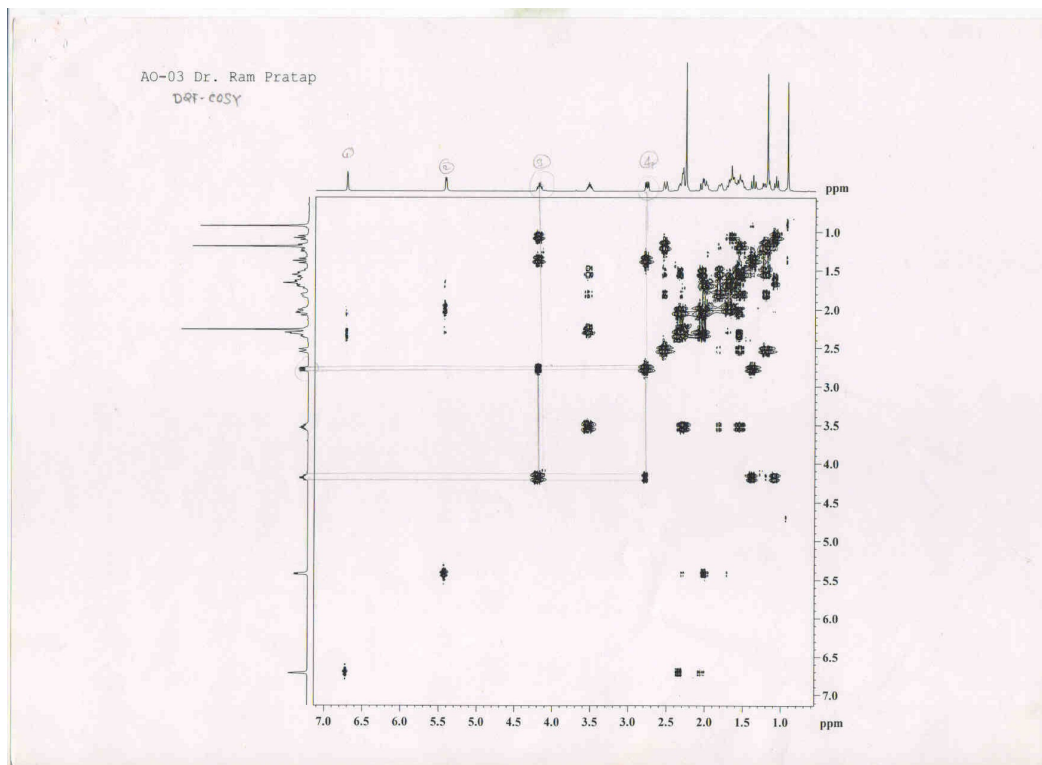
m<sup>e</sup>: multiplet overlapped with other signals, s: singlet, brs: broad singlet, d: doublet, td: triplet of a doublet, dd: doublet of a doublet.



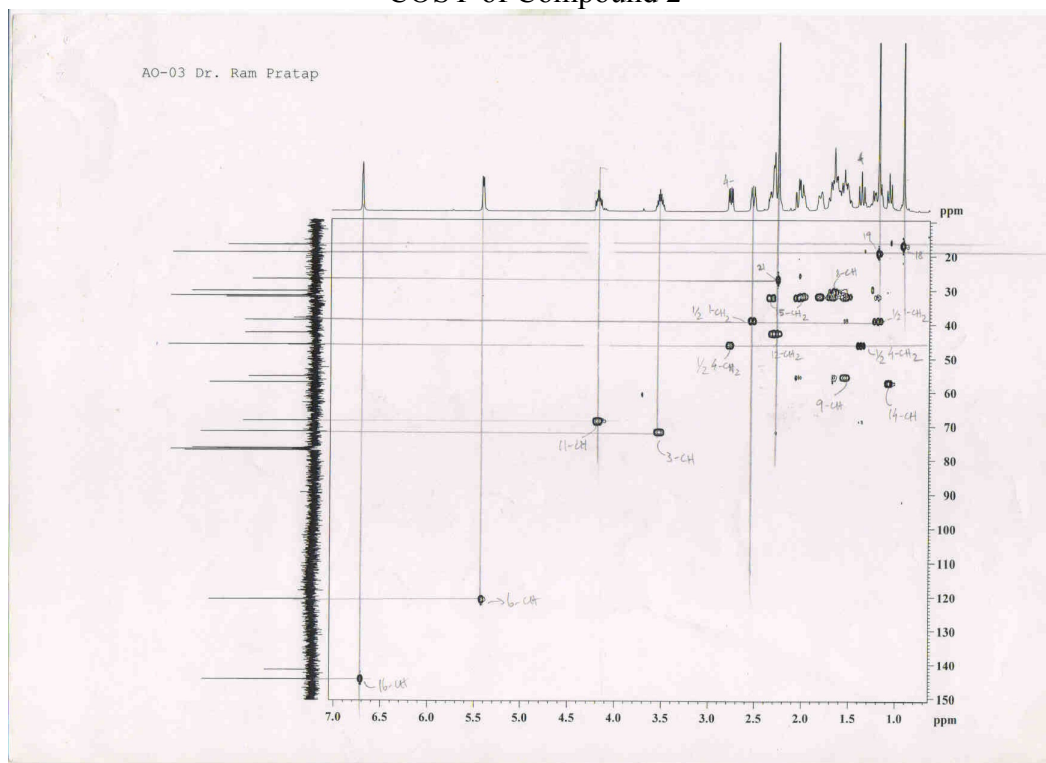
**<sup>1</sup>H NMR Compound 2**



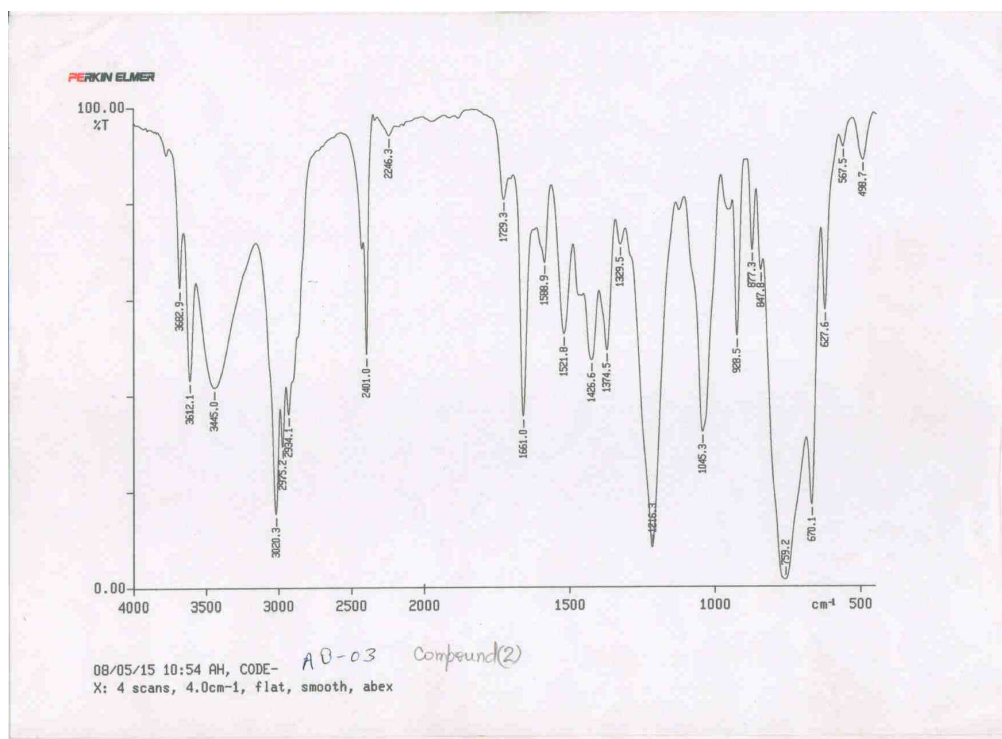
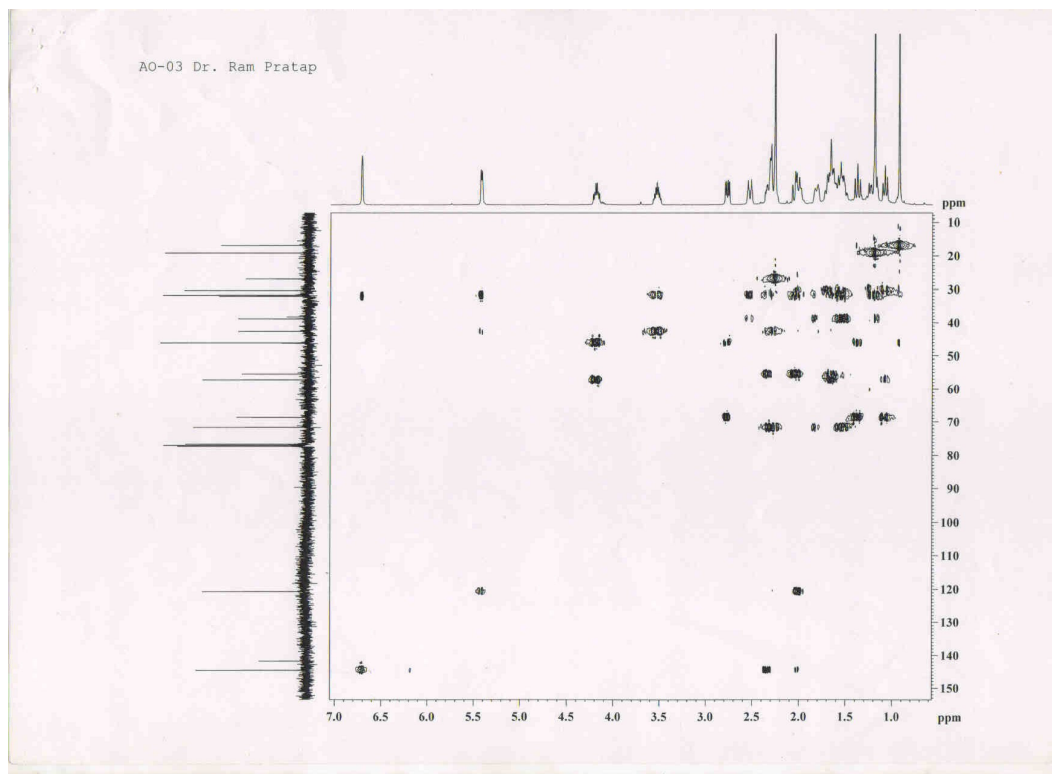
<sup>13</sup>C of Compound 2

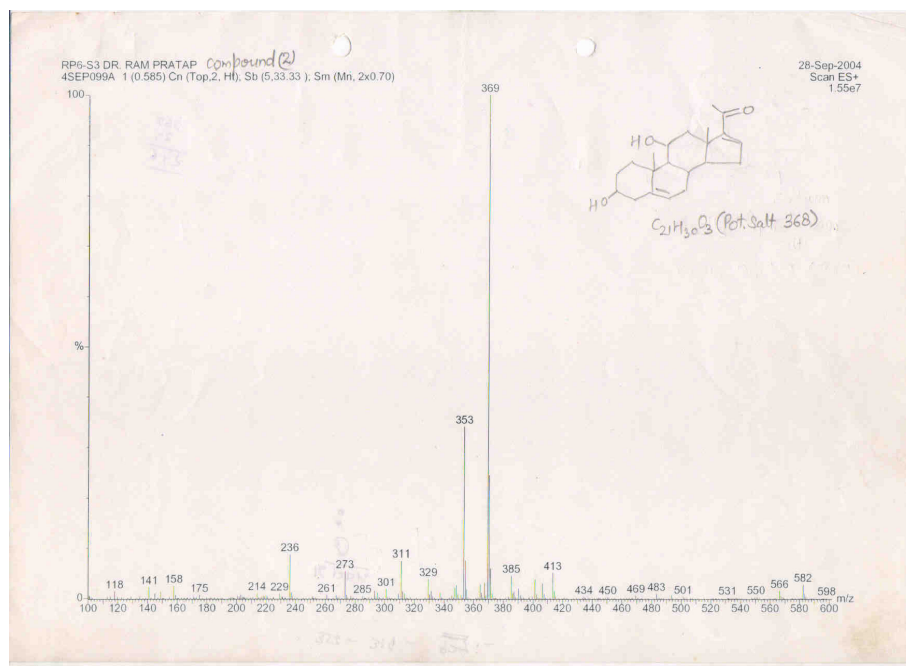


COSY of Compound 2



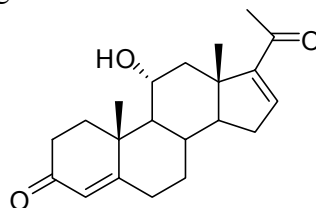
HMBC of Compound 2





Mass spectrum of Compound 2

3 $\beta$ , 11 $\alpha$ -Dihydroxy-5, 16-dien-pregnane-20-one 3:



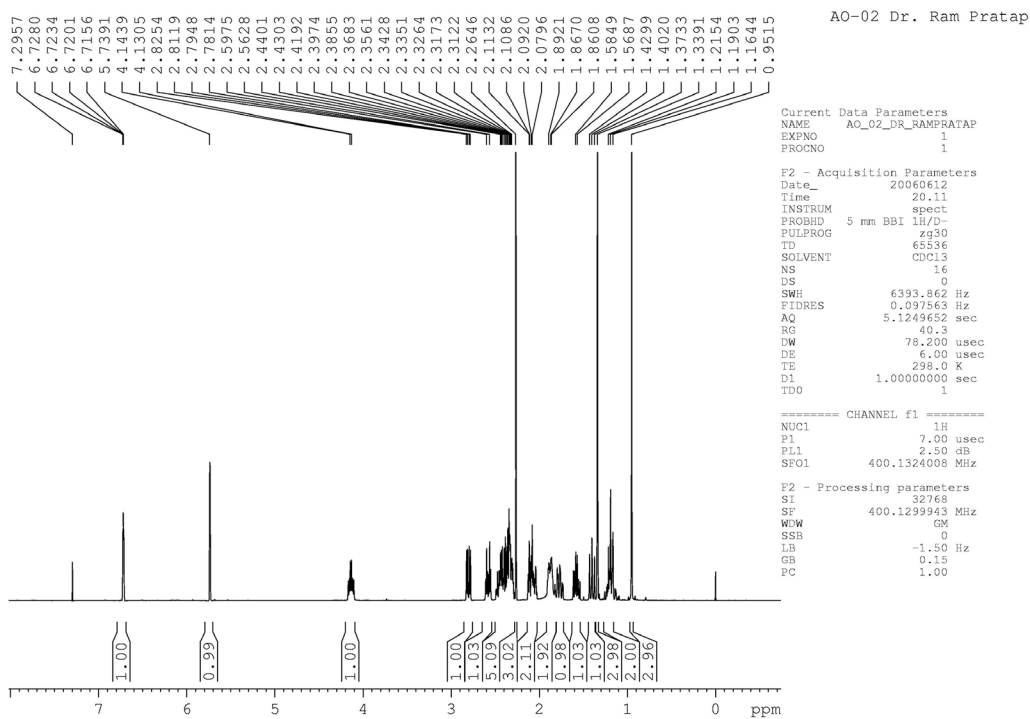
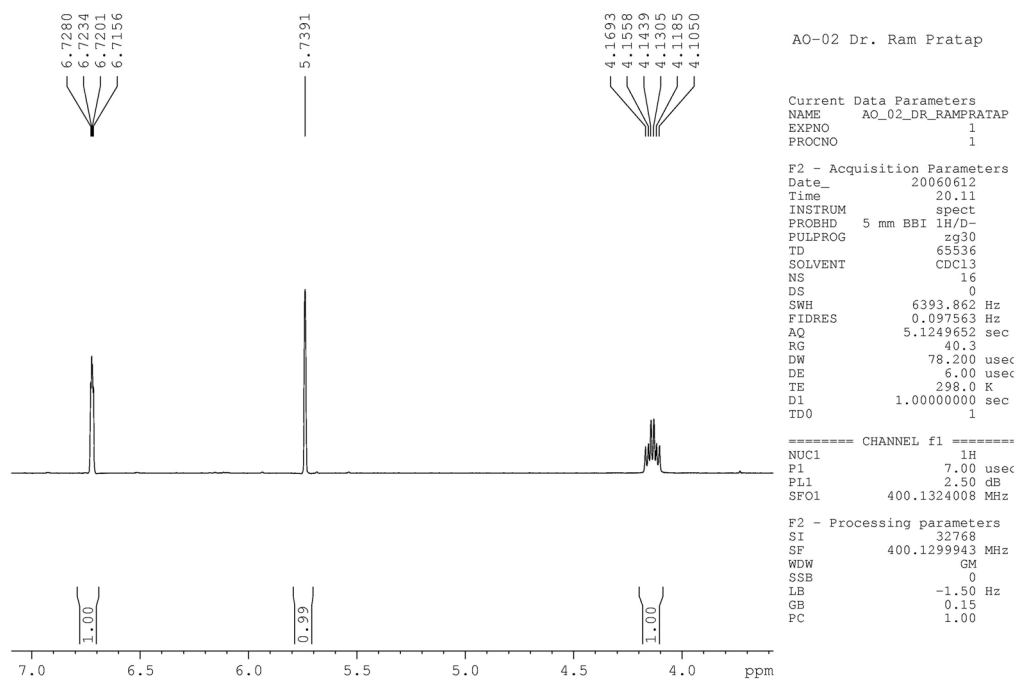
Compound 3

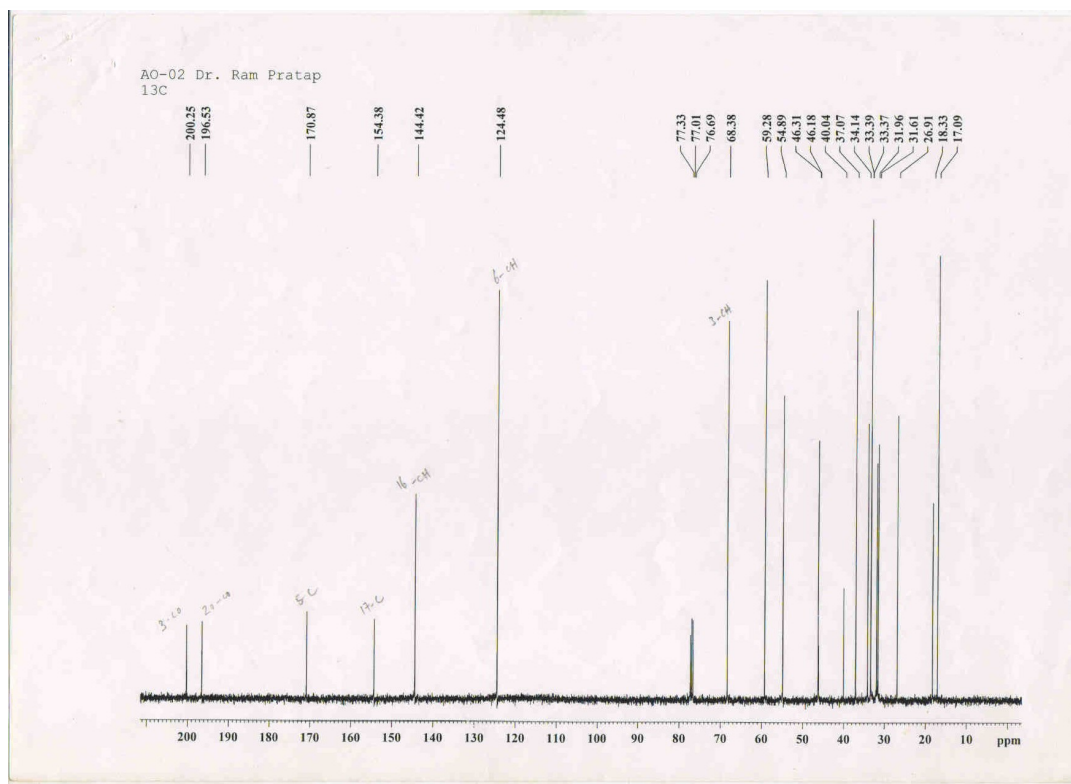
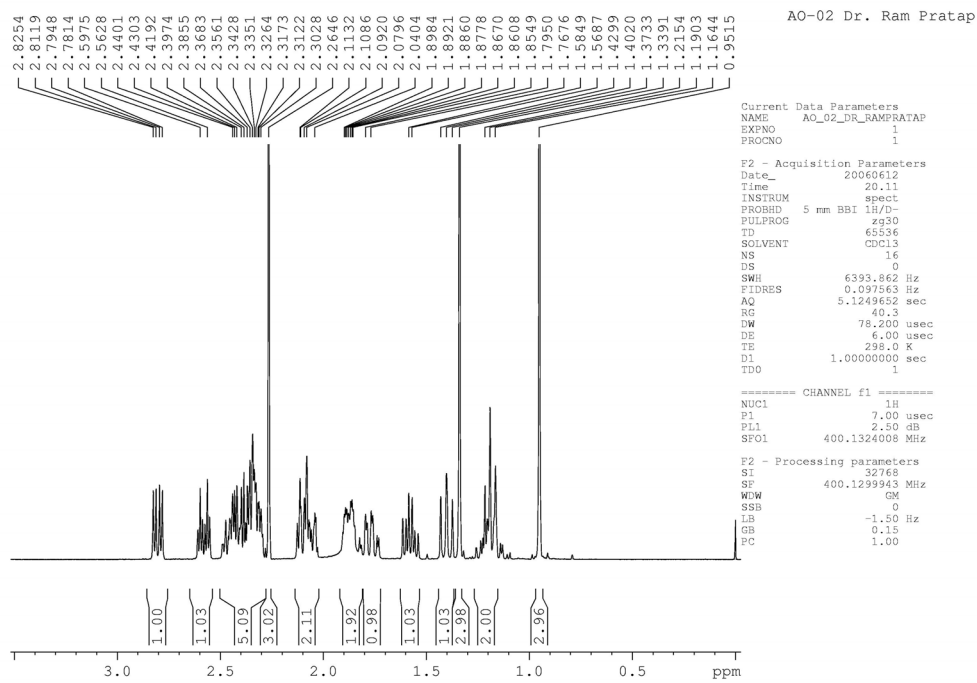
M.P. 205<sup>0</sup>C; M<sup>+</sup>+1(330); IR (KBr): 1652 cm<sup>-1</sup>; NMR:

**Table 3.** Complete assignment of proton and carbon signals of 11 $\alpha$ -hydroxy-4,16-dien-pregnan-3,20-dione **3** with help of HSQC and HMBC

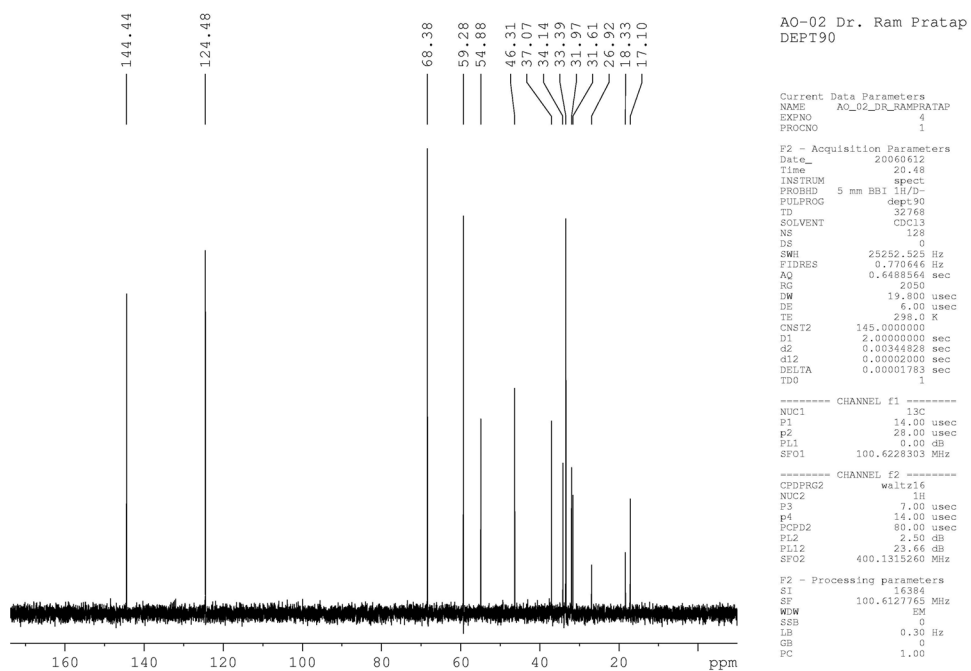
|     | H    | J (Hz)         | <sup>13</sup> C |                 |
|-----|------|----------------|-----------------|-----------------|
| 1   | 2.59 | td (4.5, 14.0) | 37.1            | CH <sub>2</sub> |
| 1'  | 2.09 | m <sup>e</sup> |                 |                 |
| 2   | 2.43 | m <sup>e</sup> | 34.1            | CH <sub>2</sub> |
| 2'  | 2.34 | m <sup>e</sup> |                 |                 |
| 3   | -    |                | 200.3           | C               |
| 4   | 5.74 | brs            | 124.5           | CH              |
| 5   | -    | -              | 170.9           | C               |
| 6   | 2.44 | m <sup>e</sup> | 33.4            | CH <sub>2</sub> |
| 6'  | 2.34 | m <sup>e</sup> |                 |                 |
| 7   | 1.88 | m <sup>e</sup> | 31.6            | CH <sub>2</sub> |
| 7'  | 1.18 | m <sup>e</sup> |                 |                 |
| 8   | 1.78 | qd (3.1, 11.5) | 33.4            | CH              |
| 9   | 1.19 | -              | 59.3            | CH              |
| 10  | -    | -              | 40.0            | C               |
| 11  | 4.14 | td (5.4, 10.3) | 68.4            | CH              |
| 12  | 2.8  | dd (5.5,12.1)  | 46.3            | CH <sub>2</sub> |
| 12' | 1.29 | m <sup>e</sup> |                 |                 |
| 13  | -    |                | 46.2            | C               |
| 14  | 1.57 | td (6.4, 11.7) | 54.9            | CH              |
| 15  | 2.35 | dd (2.9,5.3)   | 31.9            | CH <sub>2</sub> |
| 15' |      | m <sup>e</sup> |                 |                 |
| 16  | 6.72 | dd (1.8, 3.2)  | 144.5           | CH              |
| 17  | -    | -              | 154.4           | C               |
| 18  | 0.95 | s              | 17.1            | CH <sub>3</sub> |
| 19  | 1.34 | s              | 18.3            | CH <sub>3</sub> |
| 20  | -    | -              | 196.7           | C               |
| 21  | 2.27 | s              | 26.9            | CH <sub>3</sub> |

m<sup>e</sup>: multiplet overlapped with other signals, s: singlet, brs: broad singlet, d: doublet, td: triplet of a doublet, dd: doublet of a doublet.

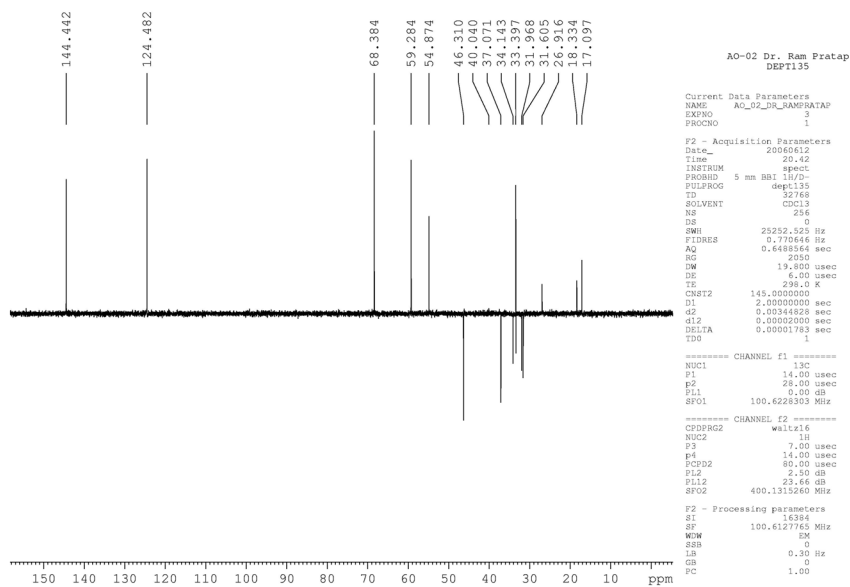
 $^1\text{H}$  NMR of Compound 3

 $^{13}\text{C}$  of Compound 3

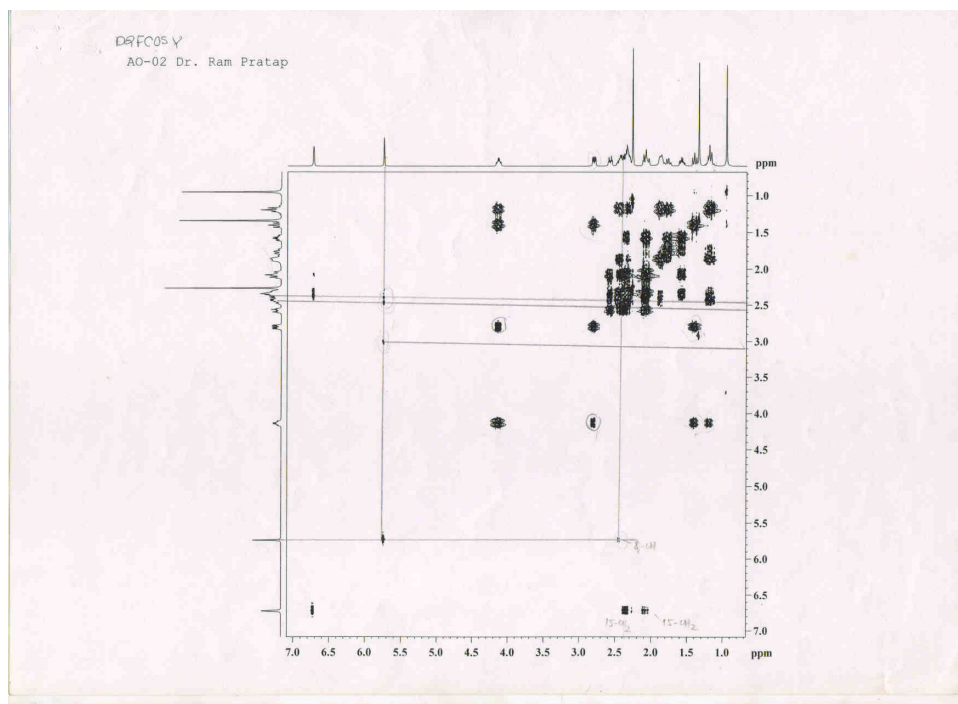




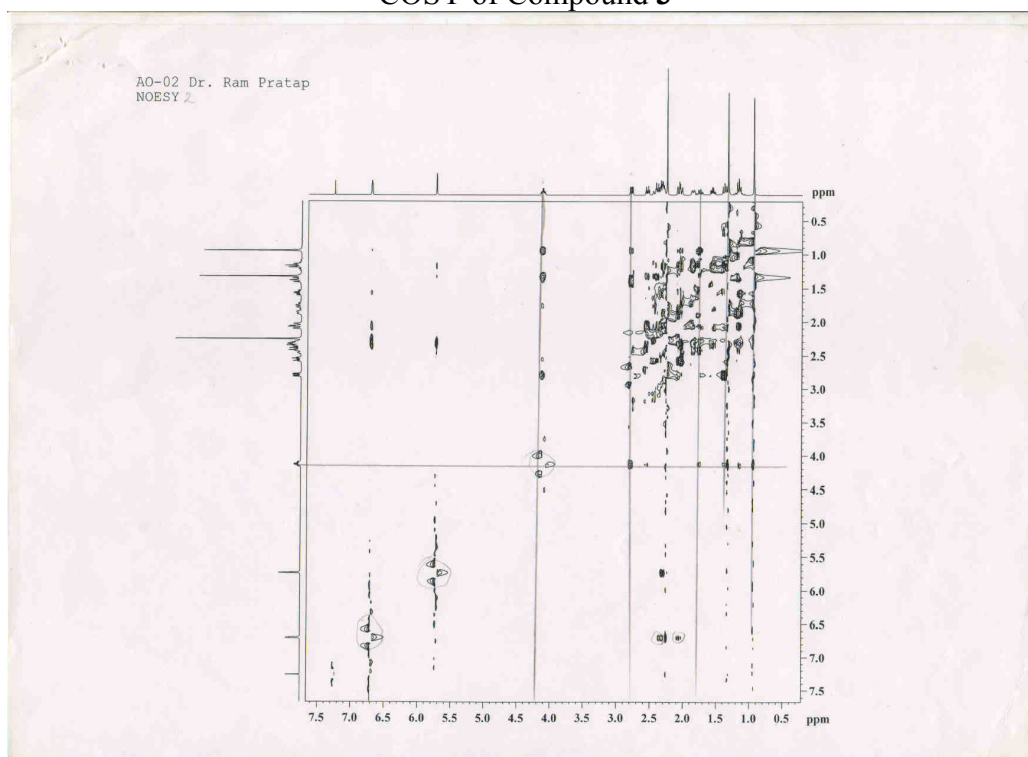
Dept 90 of Compound 3



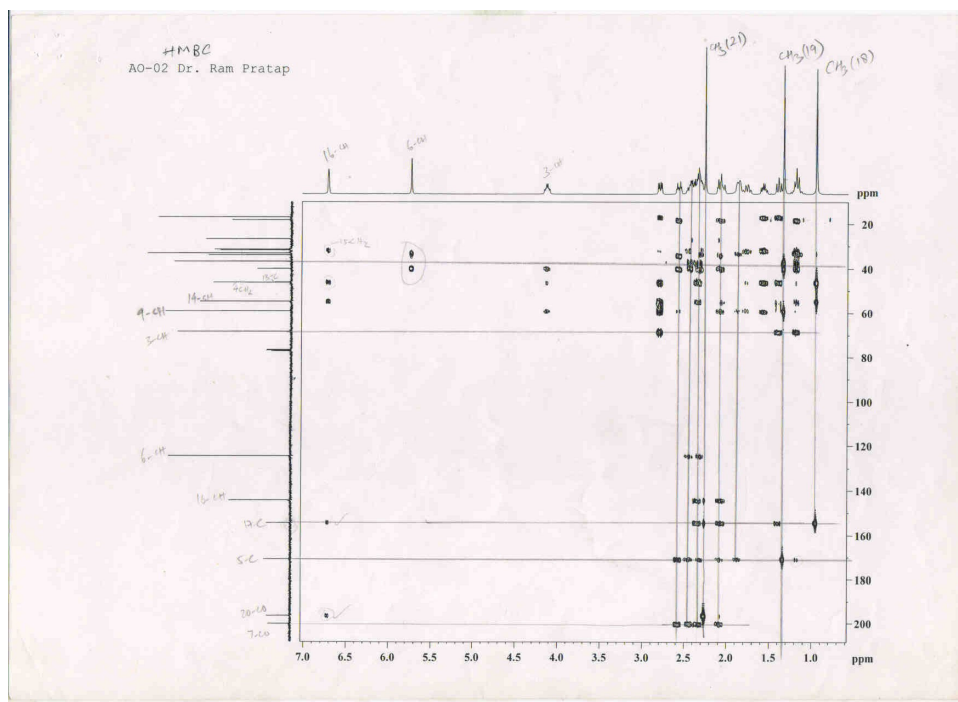
Dept 135 of Compound 3



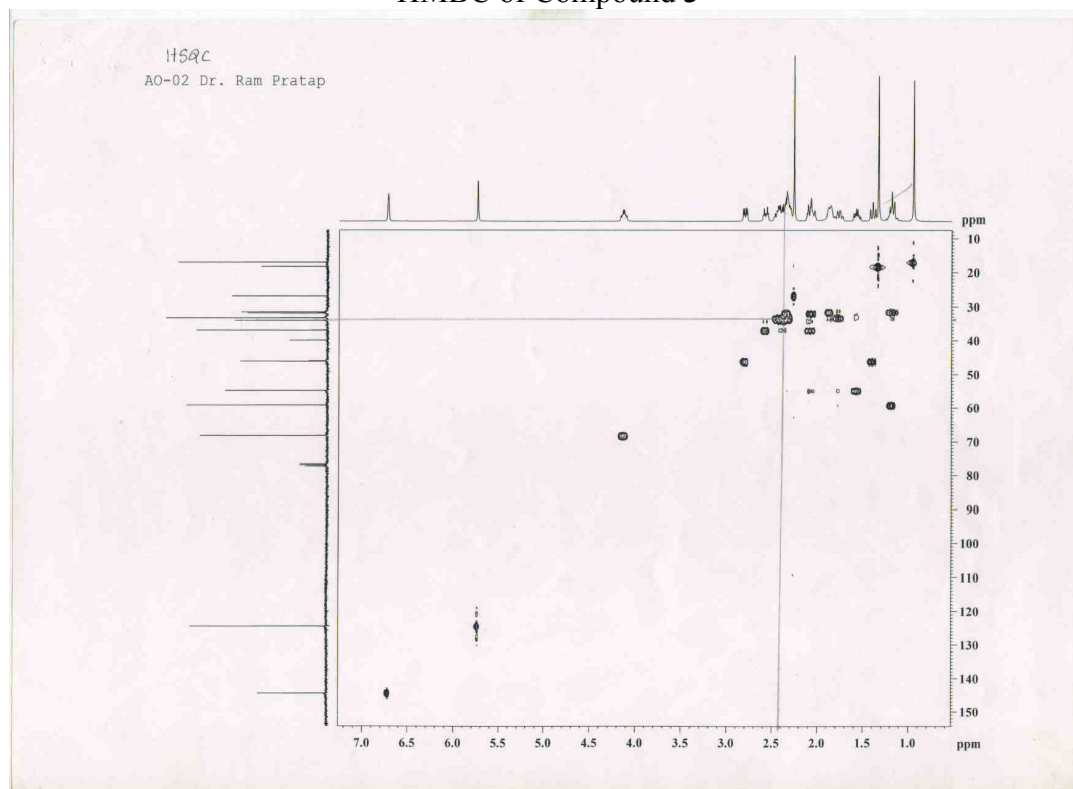
COSY of Compound 3



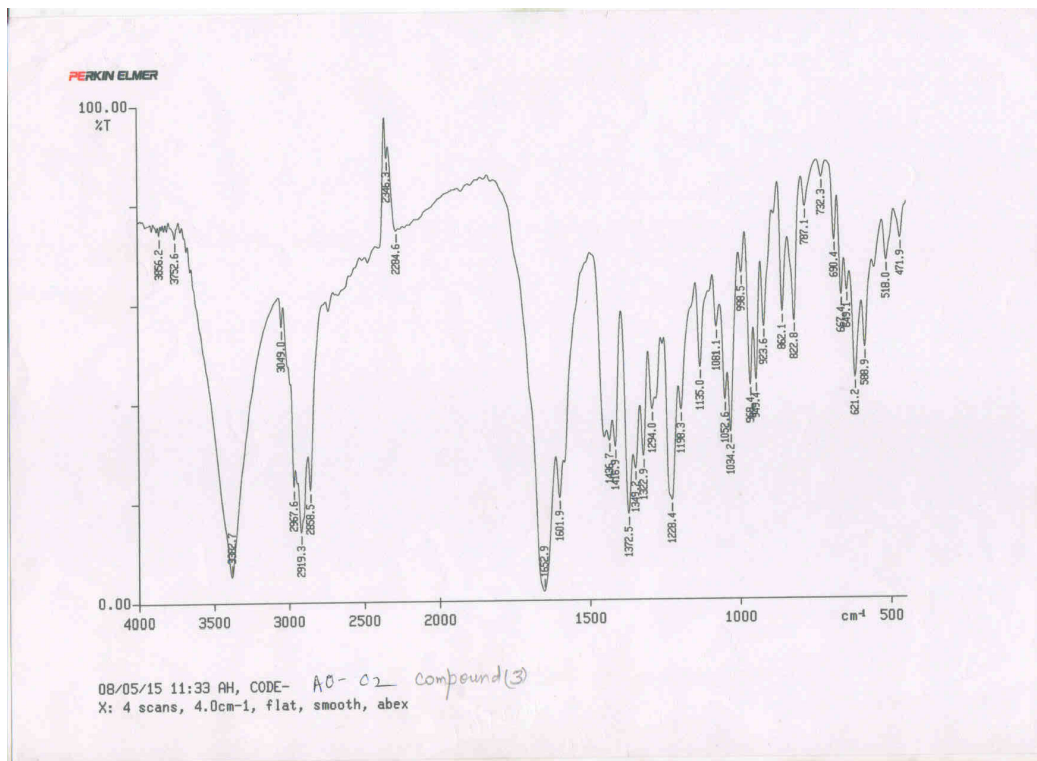
NOESY of Compound 3



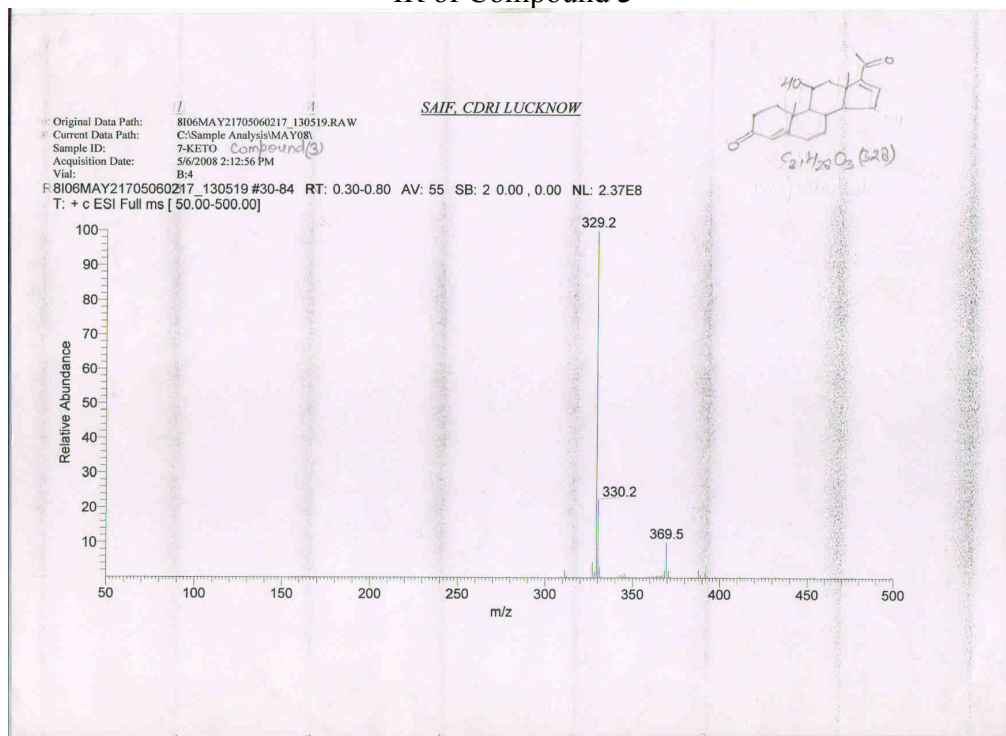
HMBC of Compound 3



HSQC of Compound 3

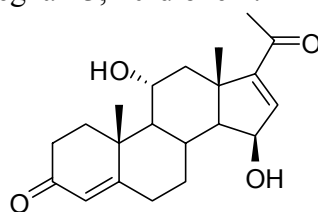


IR of Compound 3



Mass Spectrum of Compound 3

11 $\alpha$ , 15 $\beta$ -Dihydroxy-4, 16-dien-pregnan-3, 20-dione **4**:



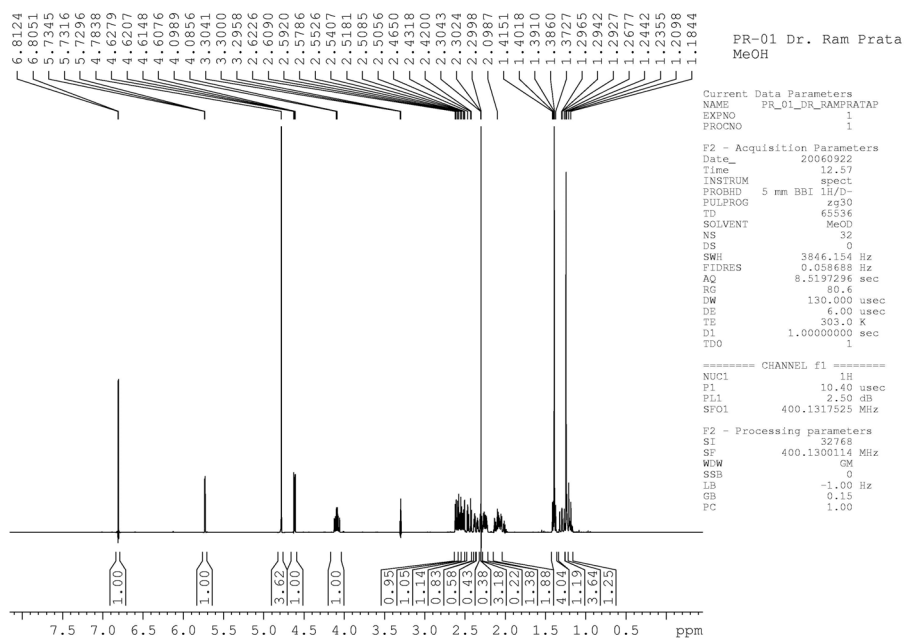
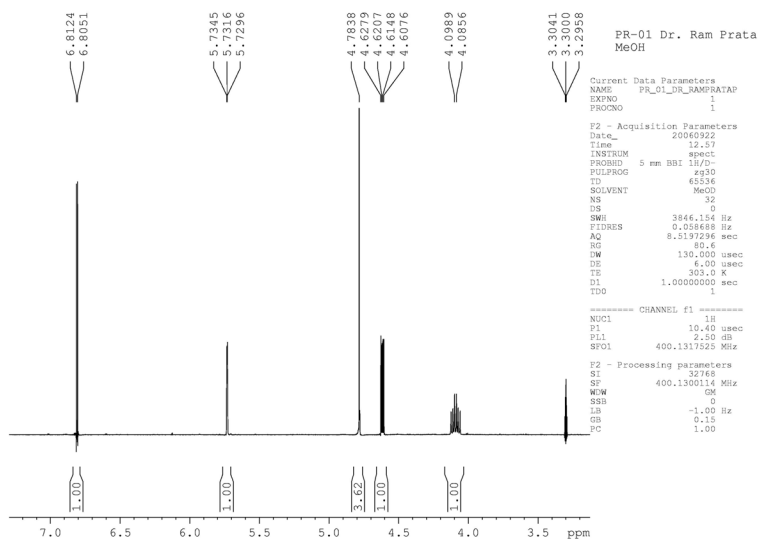
Compound **4**

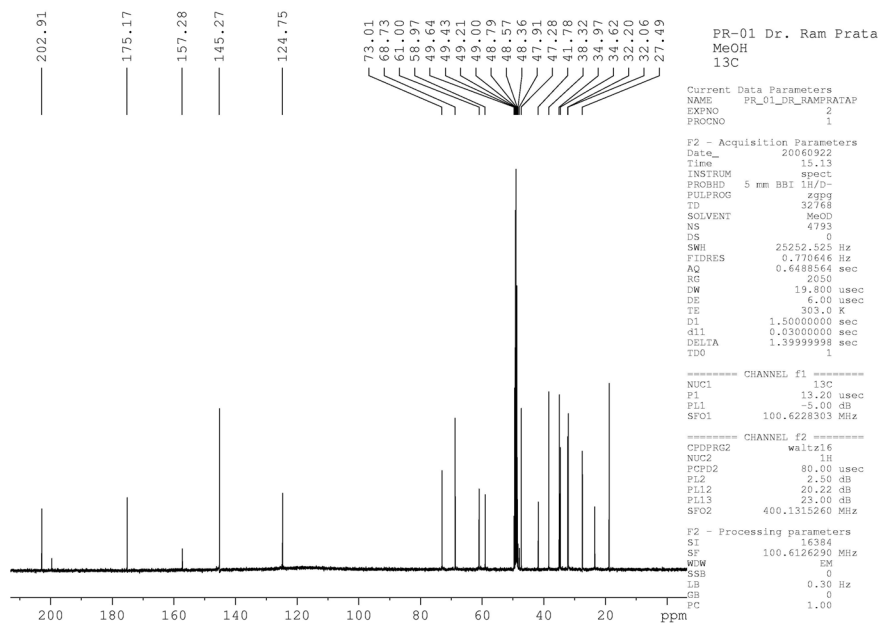
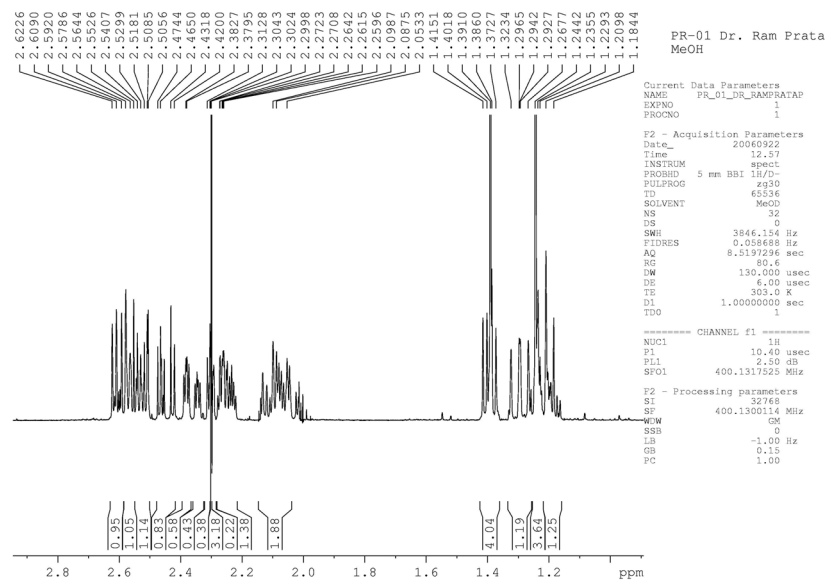
M.P. 200 °C;  $M^{+}+1$  (345); IR (KBr): 1657  $\text{cm}^{-1}$ ; NMR:

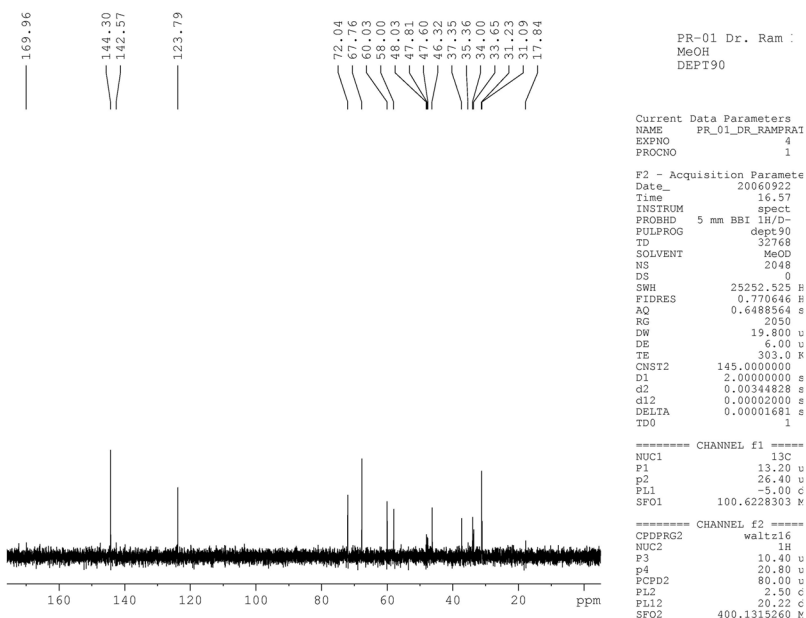
**Table 3.** Complete assignment of proton and carbon signals of 11 $\alpha$ , 15 $\beta$ -dihydroxy-4,16-dien-pregnan-3,20-dione **4** with help of HSQC and HMBC

|     | H    | J (Hz)            | $^{13}\text{C}$ |                 |
|-----|------|-------------------|-----------------|-----------------|
| 1   | 2.52 | $m^e$             | 38.2            | CH <sub>2</sub> |
| 1'  | 2.09 | $m^e$             |                 |                 |
| 2   | 2.44 | $m^e$             | 35              | CH <sub>2</sub> |
| 2'  | 2.27 | $m^e$             |                 |                 |
| 3   | -    |                   | 202.9           | CH              |
| 4   | 5.73 | brs               | 124.7           | CH <sub>2</sub> |
| 5   | -    | -                 | 175.2           | C               |
| 6   | 2.56 | $m^e$             | 34.6            | CH              |
| 6'  | 2.36 | ddd(14.6,2.5,3.9) | 32.1            | CH <sub>2</sub> |
| 7   | 2.24 | $m^e$             |                 |                 |
| 7'  | 1.21 | $m^e$             |                 | CH              |
| 8   | 2.05 | $m^e$             | 32.2            | CH              |
| 9   | 1.2  | $m^e$             | 61              | C               |
| 10  | -    |                   | 41.8            | CH <sub>2</sub> |
| 11  | 4.09 | td(5.5, 10.0)     | 68.7            | CH <sub>2</sub> |
| 12  | 2.59 | $m^e$             |                 |                 |
| 12' | 1.29 | $m^e$             | 47.3            | C               |
| 13  | -    |                   | 47.9            | CH              |
| 14  | 1.39 |                   | 59, 73          | CH <sub>2</sub> |
| 15  | 4.62 | dd(2.9,5.3)       |                 |                 |
| 16  | 6.86 | d(2.9)            | 145.3           | CH              |
| 17  | -    | -                 | 157.3           | C               |
| 18  | 1.24 | s                 | 23.5            | CH <sub>3</sub> |
| 19  | 1.39 | s                 | 18.8            | CH <sub>3</sub> |
| 20  | -    | -                 | 199.7           | C               |
| 21  | 2.29 | s                 | 27.5            | CH <sub>3</sub> |

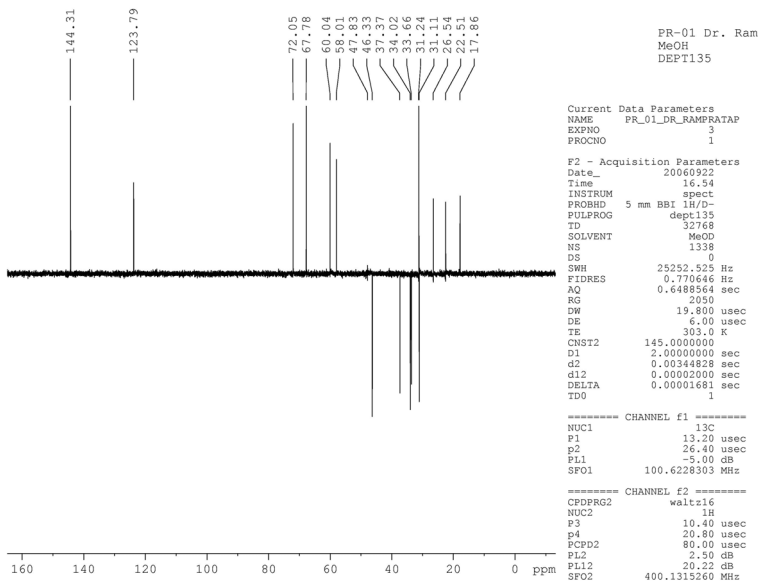
$m^e$ : multiplet overlapped with other signals, s: singlet, brs: broad singlet, d: doublet, td: triplet of a doublet, dd: doublet of a doublet, doublet of a double doublet.

 $^1\text{H}$  NMR of Compound 4

<sup>13</sup>C of Compound 4

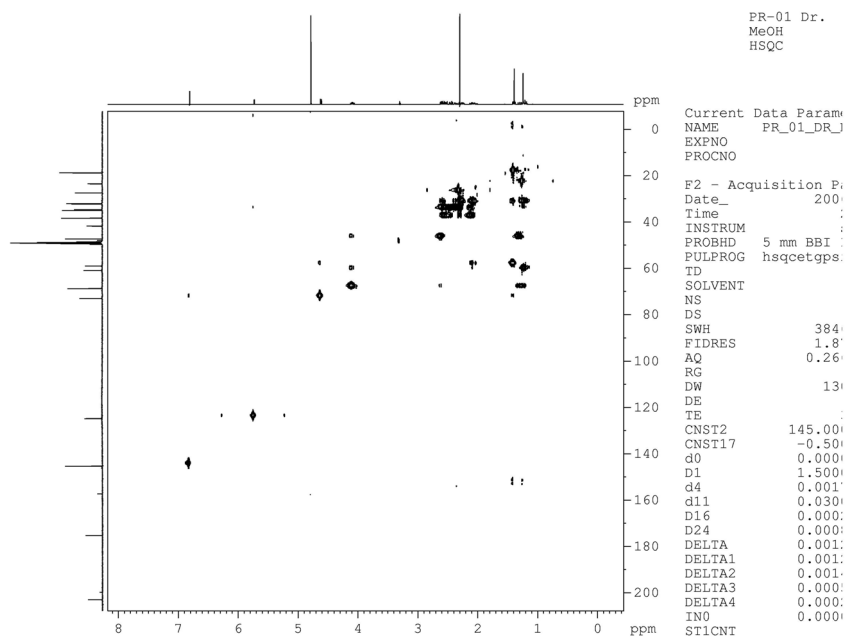


Dept 90 of Compound 4

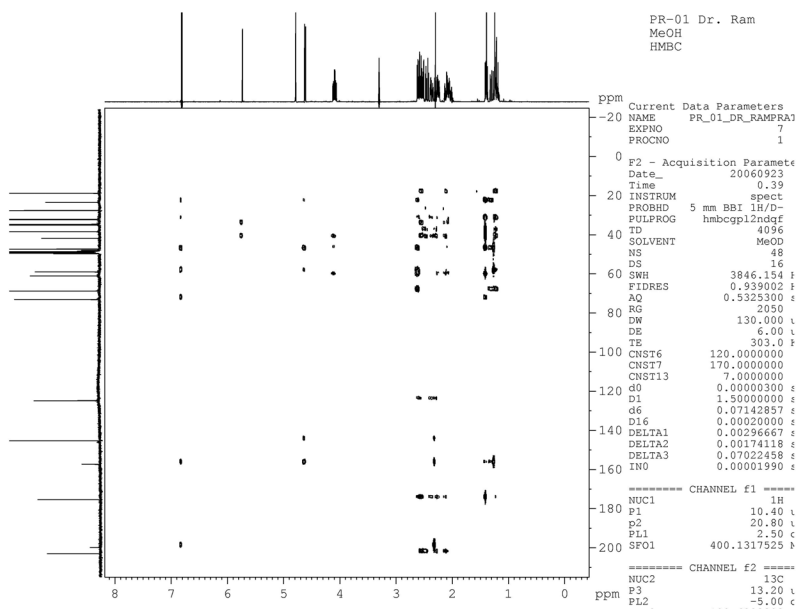


Dept 135 of Compound 4

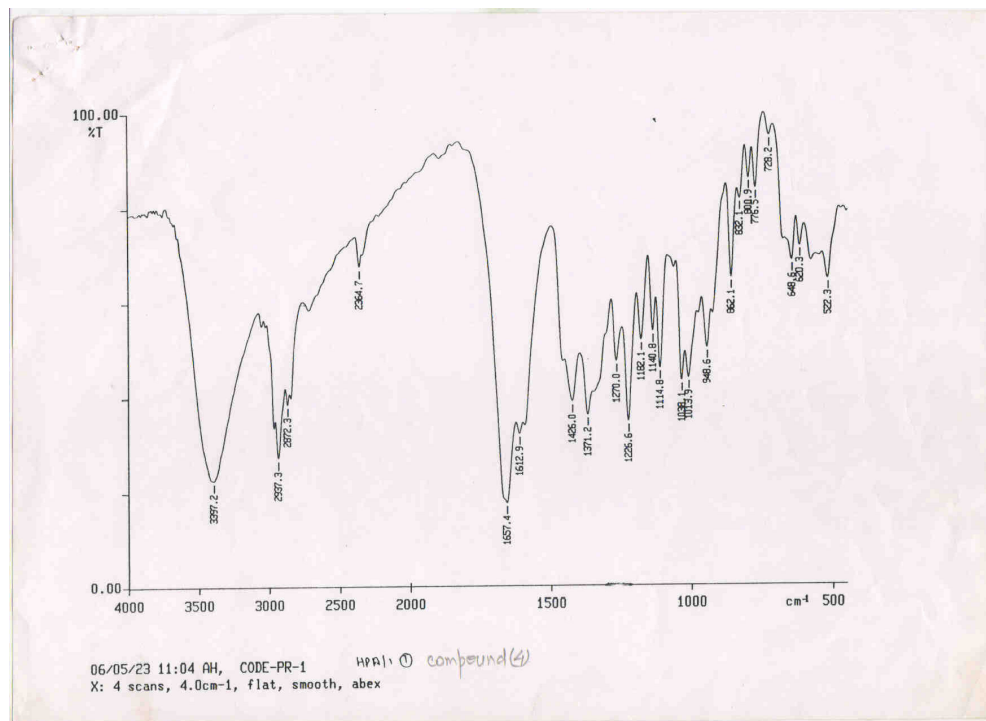




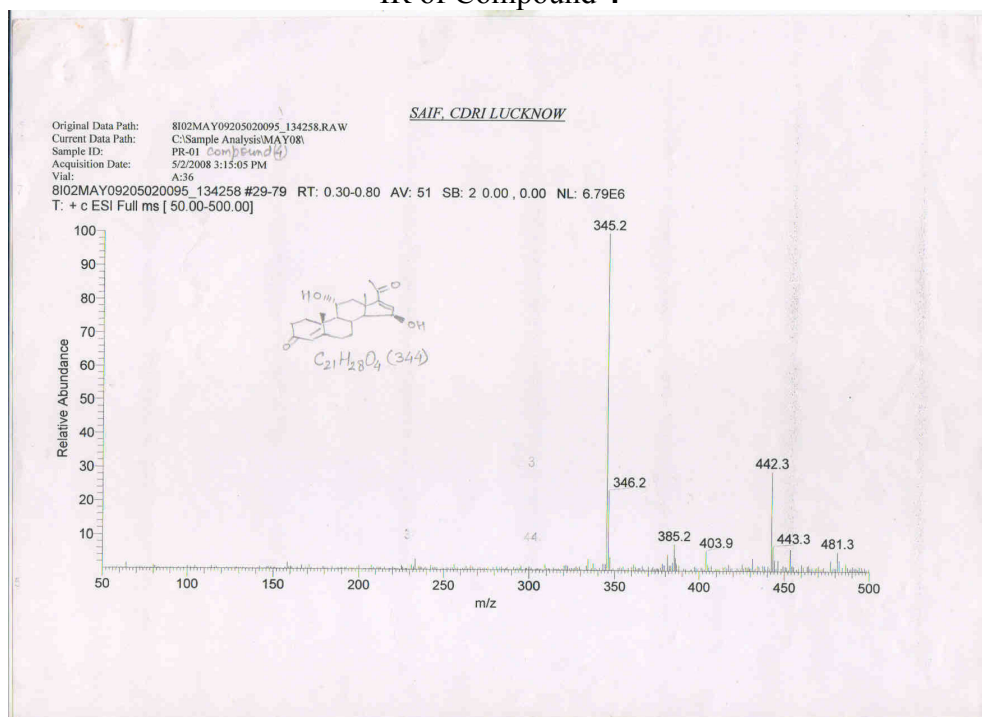
HSQC of Compound 4



HMBC of Compound 4

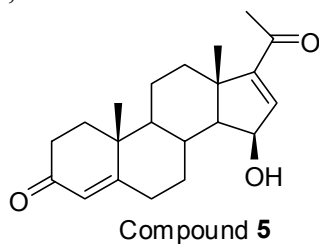


IR of Compound 4



Mass Spectrum of Compound 4

15 $\beta$ -Hydroxy-4, 16-dien-pregnan-3, 20-dione **5**:



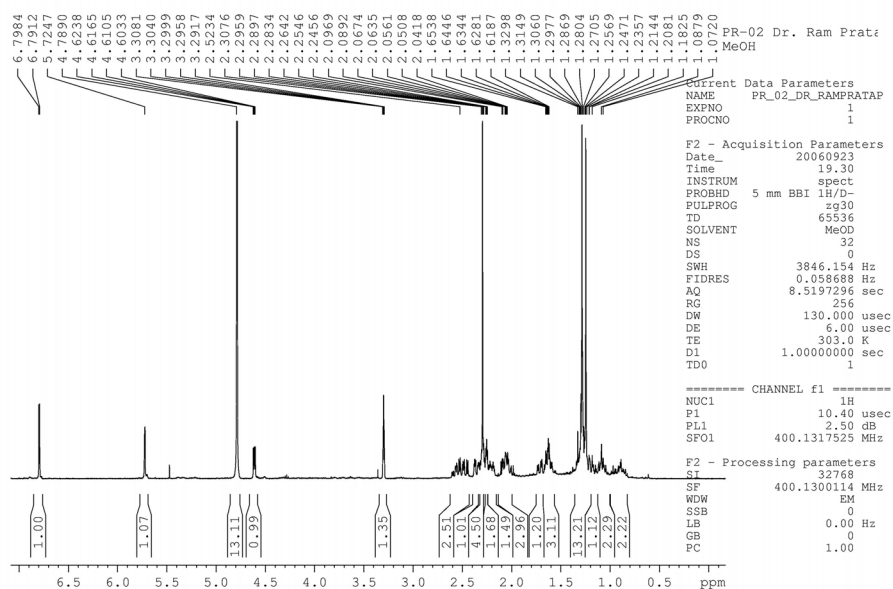
M.P. 210 °C;  $M^{+}+1$  (329); IR (KBr): 1596  $\text{cm}^{-1}$ ;

NMR: Complete assignment of proton and carbon signals of 15 $\beta$ -Hydroxy-4, 16-dien-pregnan-3, 20-dione **5** with help of HSQC and HMBC

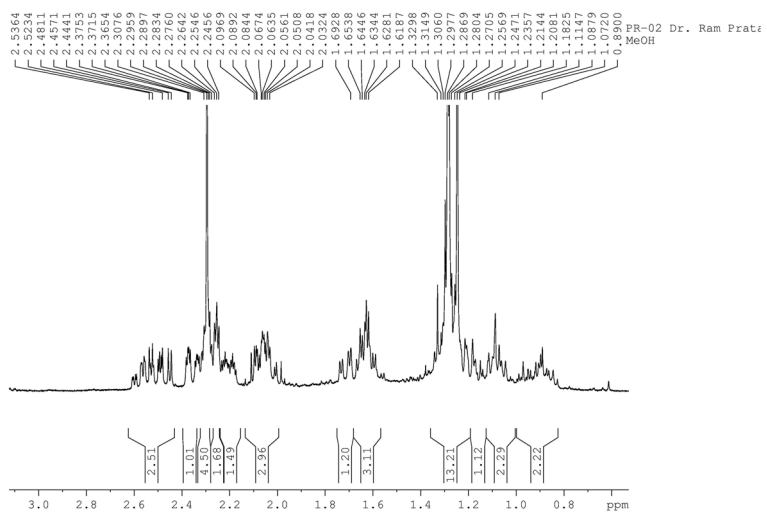
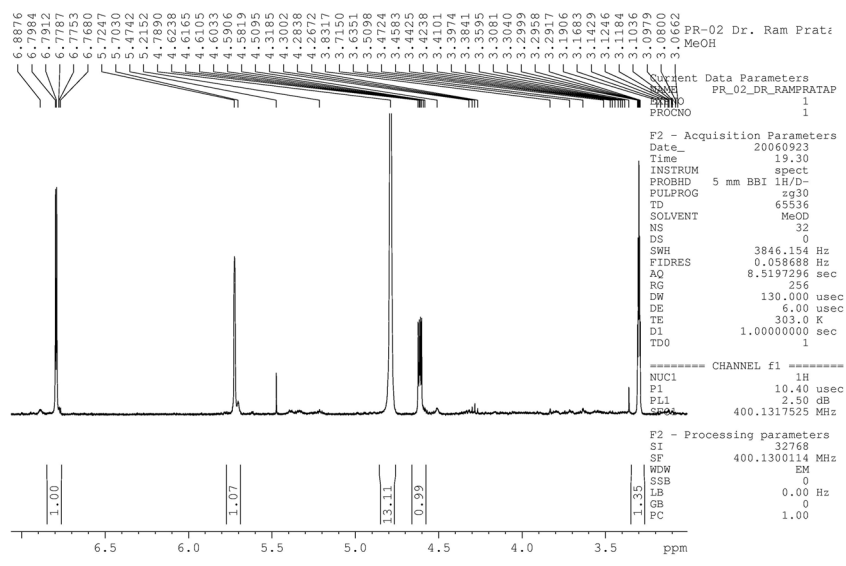
| H   | $\delta$ | J (in Hz)           | $\delta$ | Type of carbon |
|-----|----------|---------------------|----------|----------------|
| 1   | 1.7      | $m^e$               | 36.7     | $\text{CH}_2$  |
| 1'  | 2.07     | $m^e$               |          |                |
| 2   | 2.49     | $m^e$               | 34.8     | $\text{CH}_2$  |
| 2'  | 2.29     | $m^e$               |          |                |
| 3   | -        | -                   | 202.4    | CH             |
| 4   | 5.72     | brs                 | 124.3    | $\text{CH}_2$  |
| 5   | -        | -                   | 175.2    | C              |
| 6   | 2.58     | ddd (1.8,5.3, 14.7) | 33.9     | $\text{CH}_2$  |
| 6'  | 2.35     | ddd (2.6,4.1,14.7)  |          |                |
| 7   | 2.21     | $m^e$               | 32.3     | $\text{CH}_2$  |
| 7'  | 1.19     | $m^e$               |          |                |
| 8   | 2.05     | $m^e$               | 32.4     | CH             |
| 9   | 1.08     | $m^e$               | 56.1     | C              |
| 10  | -        | -                   | 40.3     | $\text{CH}_2$  |
| 11  | 1.63     | $m^e$               | 21.7     | $\text{CH}_2$  |
| 11' | -        | -                   |          |                |
| 12  | 2.27     | $m^e$               | 36.0     | $\text{CH}_2$  |

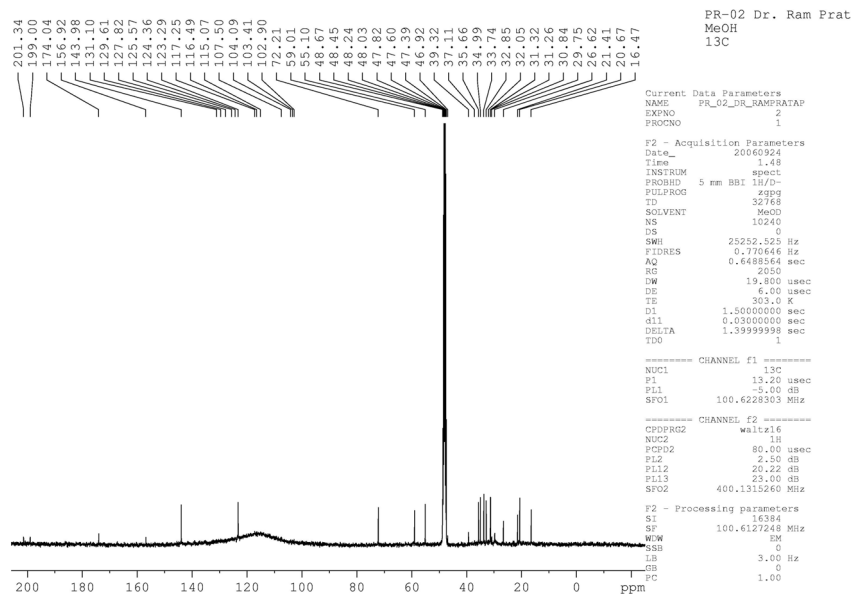
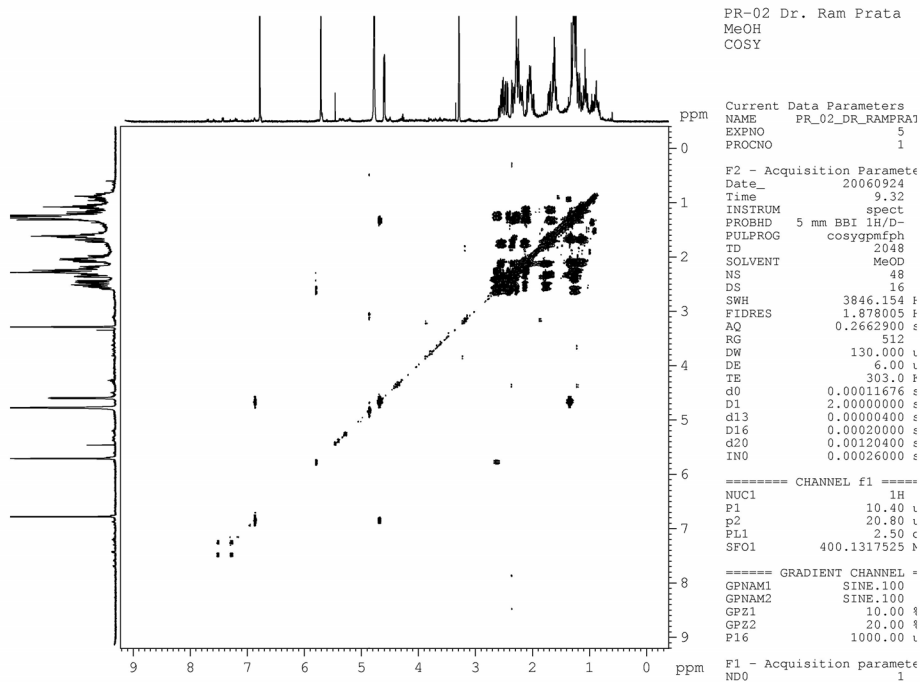
|     |      |                |       |                 |
|-----|------|----------------|-------|-----------------|
| 12' | 1.27 | m <sup>e</sup> |       |                 |
| 13  | -    | -              | 47.9  | C               |
| 14  | 1.28 | m <sup>e</sup> | 60.0  | CH              |
| 15  | 4.61 | dd (2.9,5.3)   | 73.2  | CH <sub>2</sub> |
| 15' | -    | -              |       |                 |
| 16  | 6.8  | d (2.9)        | 144.9 | CH              |
| 17  | -    | -              | 157.9 | C               |
| 18  | 1.25 | s              | 22.4  | CH <sub>3</sub> |
| 19  | 1.29 | s              | 17.5  | CH <sub>3</sub> |
| 20  | -    | -              | 200.0 | C               |
| 21  | 2.30 | s              | 27.6  | CH <sub>3</sub> |

m<sup>e</sup>: multiplet overlapped with other signals, s: singlet, brs: broad singlet, d: doublet, td: triplet of a doublet, dd: doublet of a doublet, doublet of a double doublet.

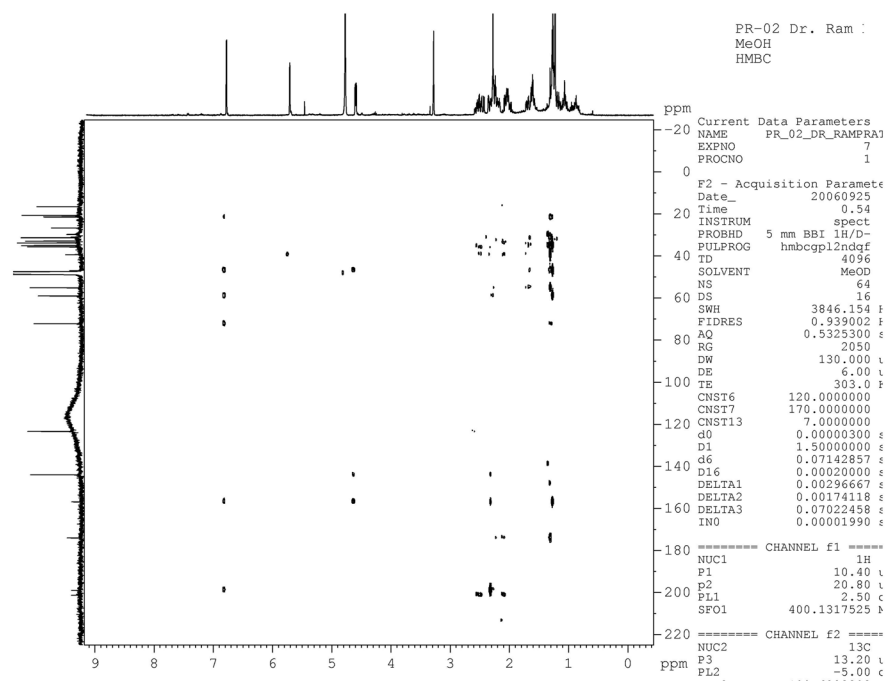


<sup>1</sup>H NMR of Compound 5

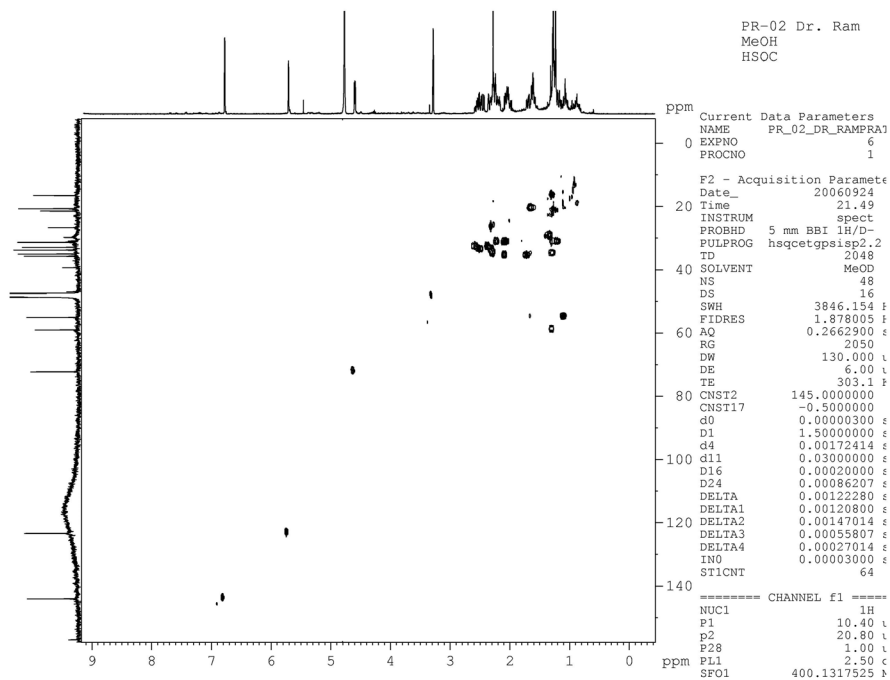


 $^{13}\text{C}$  of Compound 5

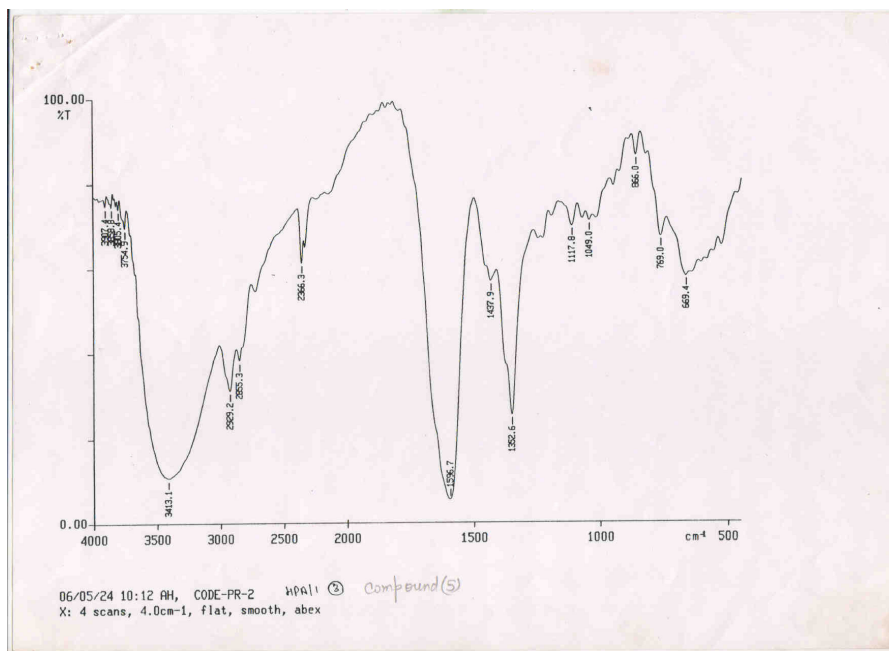
COSY of Compound 5



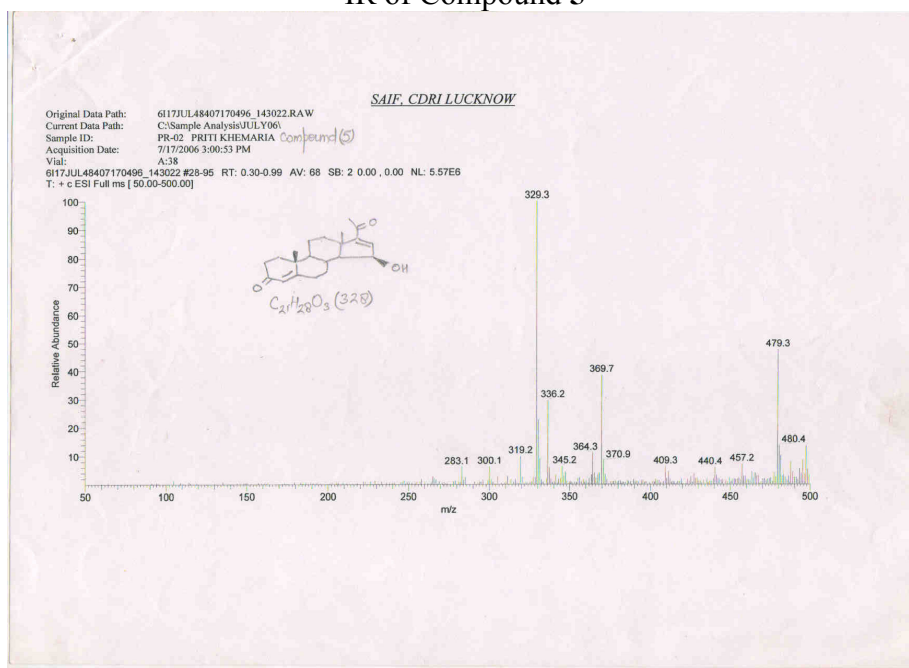
HMBC of Compound 5



HSQC of Compound 5



IR of Compound 5



Mass Spectrum of Compound 5