Synthesis of 2´-*C*-β-difluoromethylribonucleosides and enzymatic incorporation into oligonucleotides

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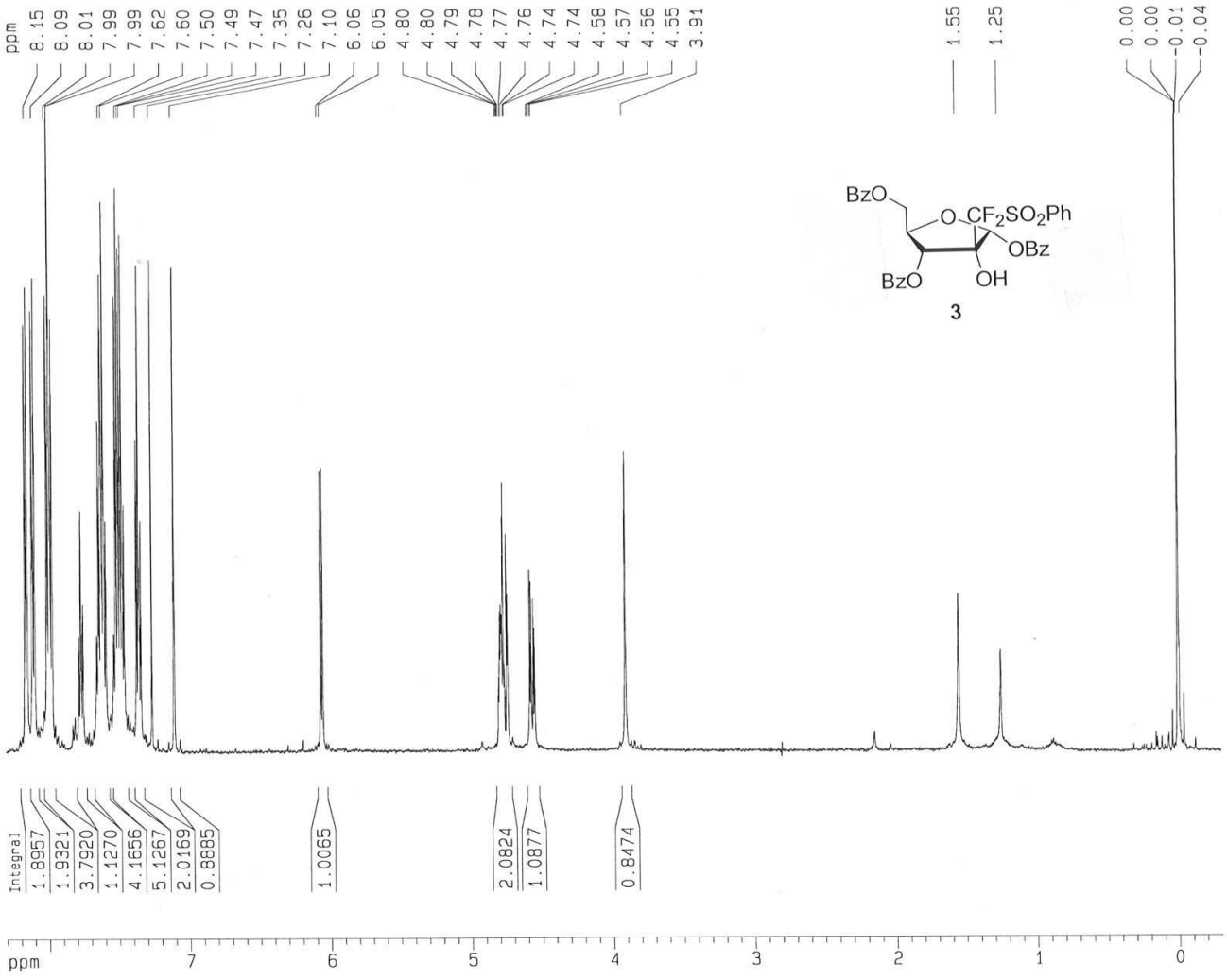


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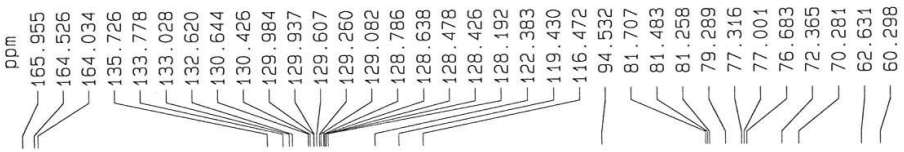

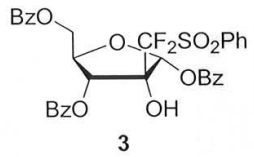

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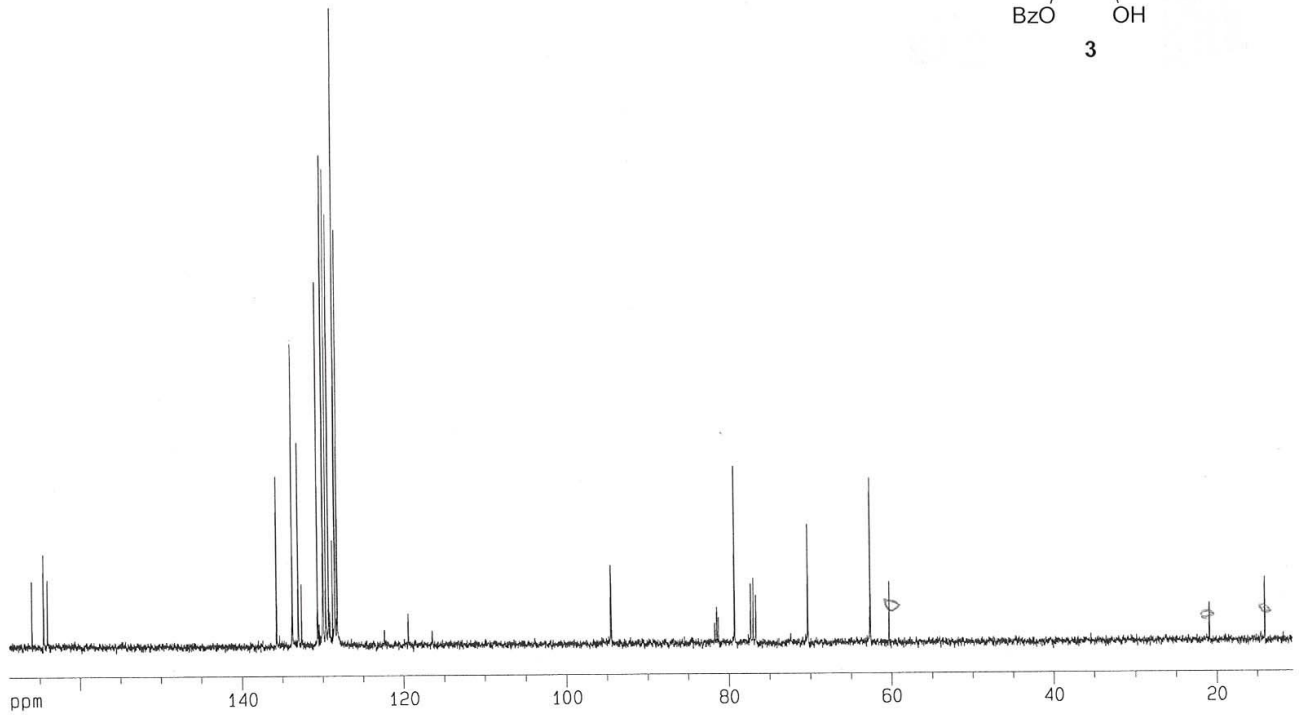


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============ CHANNEL f1 ==========
NUC1                     13C
P1                     11.00 usec
PL1                    -2.00 dB
SFO1              100.6254358 MHz

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CPDPRG2              waltz16
NUC2                     1H
PCPD2                101.00 usec
PL2                  120.00 dB
PL12                  19.00 dB
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PC                        1.00

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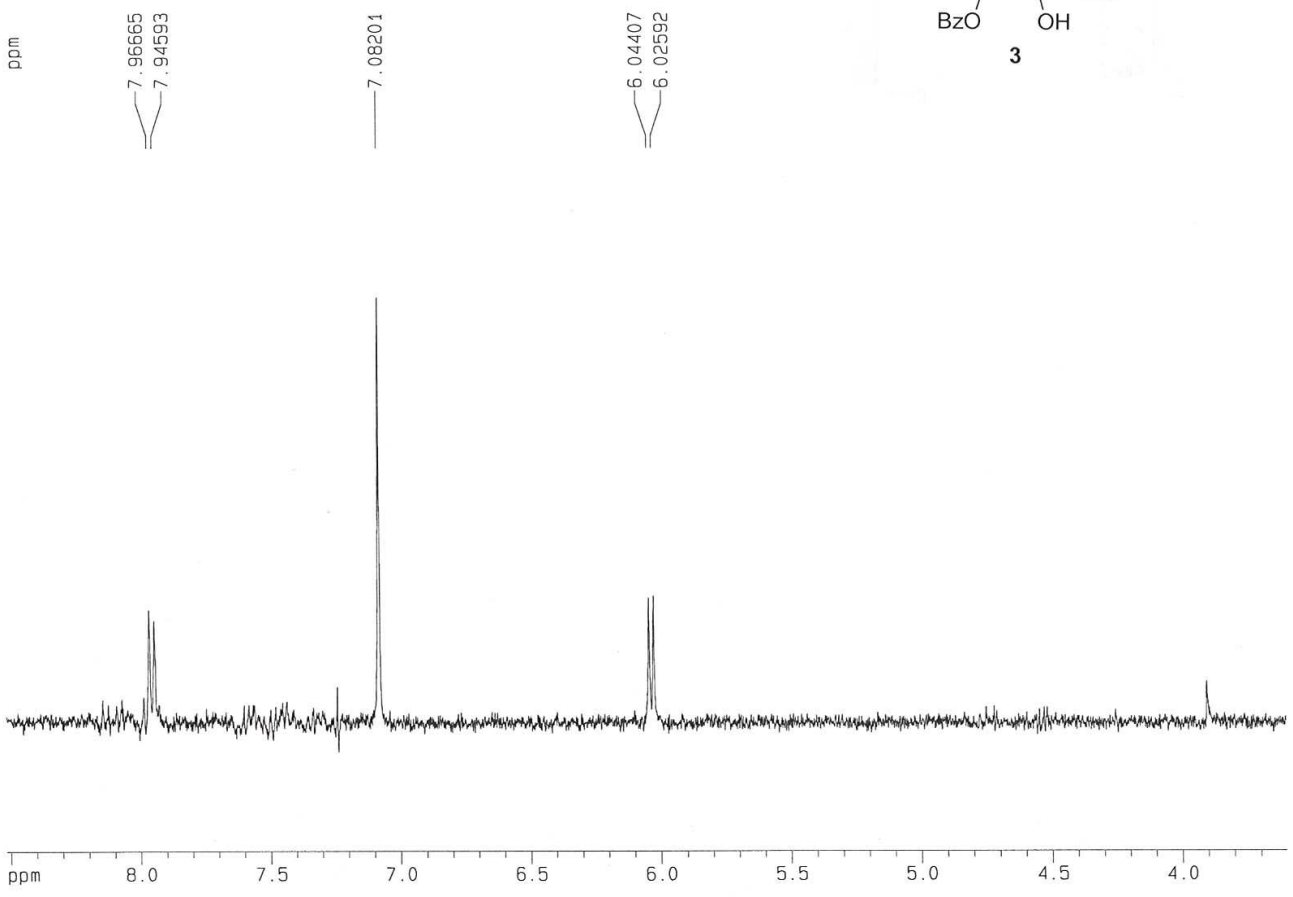

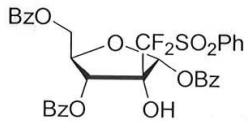

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PROCNO                      2

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======== CHANNEL f2 ========
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FG2LIST                 jyq.1
NUC2                       19F
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PL12                    24.00 dB
PL14                    54.00 dB
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GB                             0
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1D NMR plot parameters
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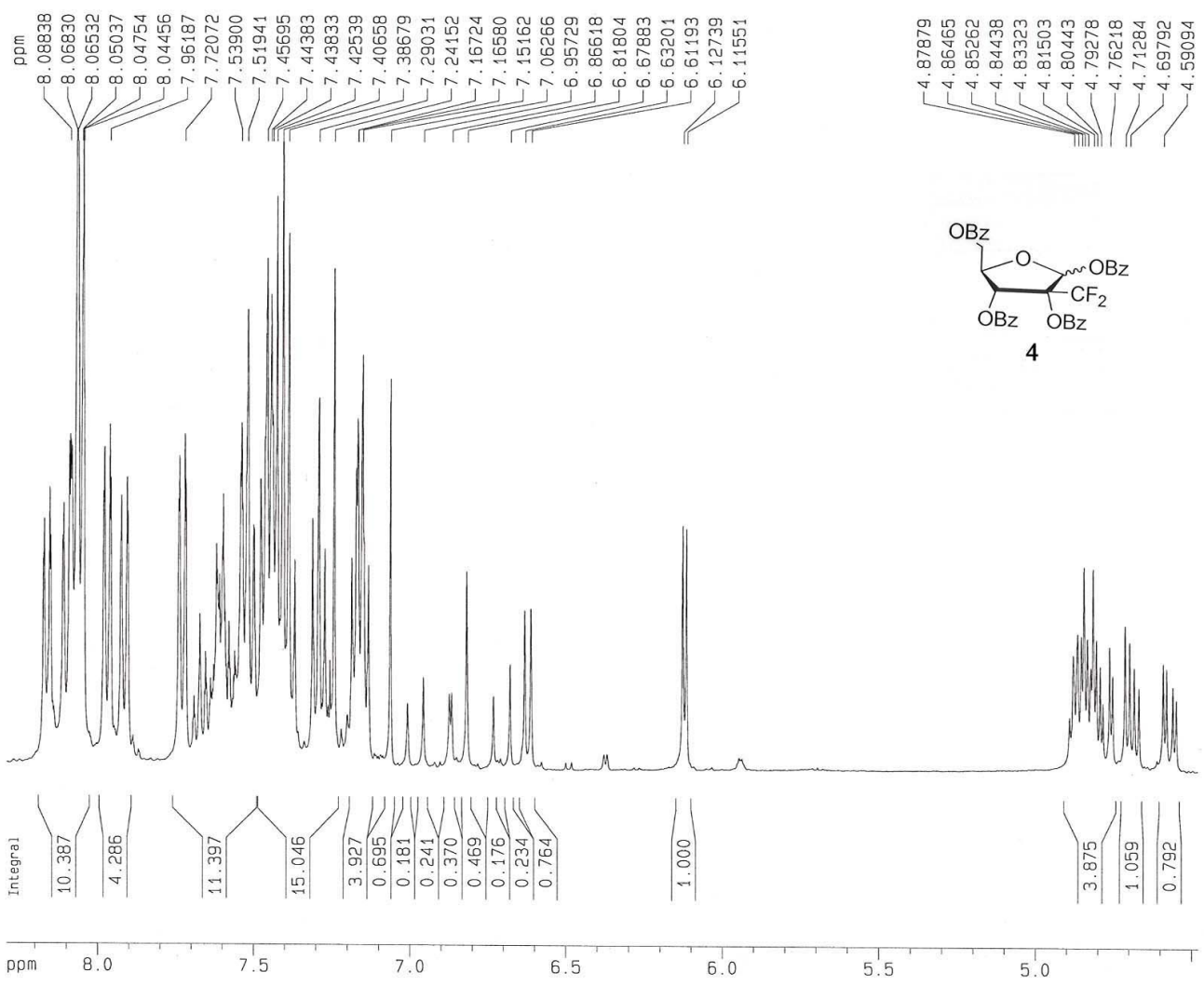

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FIDRES	0.128010 Hz
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RG	32
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TE	300.0 K
D1	2.00000000 sec

========== CHANNEL f1 ========

NUC1	1H
P1	10.50 usec
PL1	0.00 dB
SFO1	400.1319246 MHz

F2 - Processing parameters

SI	16384
SF	400.1300116 MHz
WDW	EM
SSB	0
LB	0.30 Hz
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PC	1.00

1D NMR plot parameters

CX	20.00 cm
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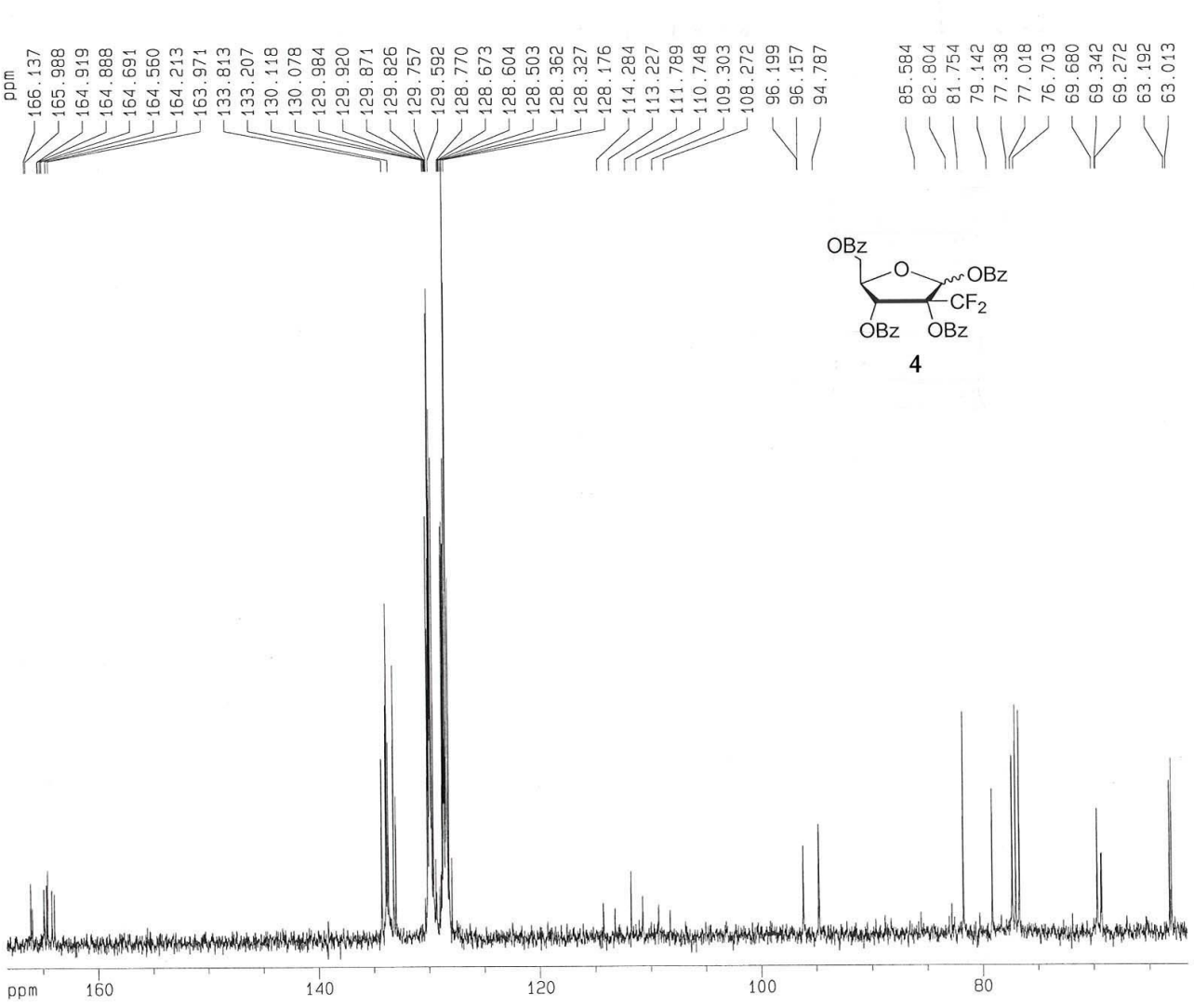


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EXPNO                      1
PROCNO                     1

F2 - Acquisition Parameters
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PULPROG                zgdc
TD                    32768
SOLVENT               CDCl3
NS                      428
DS                        0
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FIDRES             0.827035 Hz
AQ                 0.6046196 sec
RG                    1149.4
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DE                    7.50 usec
TE                   300.0 K
D1                3.00000000 sec
d11               0.03000000 sec

============ CHANNEL f1 ==========
NUC1                     13C
P1                     11.00 usec
PL1                    -2.00 dB
SFO1             100.6254358 MHz

============ CHANNEL f2 ==========
CPDPRG2              waltz16
NUC2                     1H
PCPD2                101.00 usec
PL2                    120.00 dB
PL12                    19.00 dB
SFO2             400.1324710 MHz

F2 - Processing parameters
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1D NMR plot parameters
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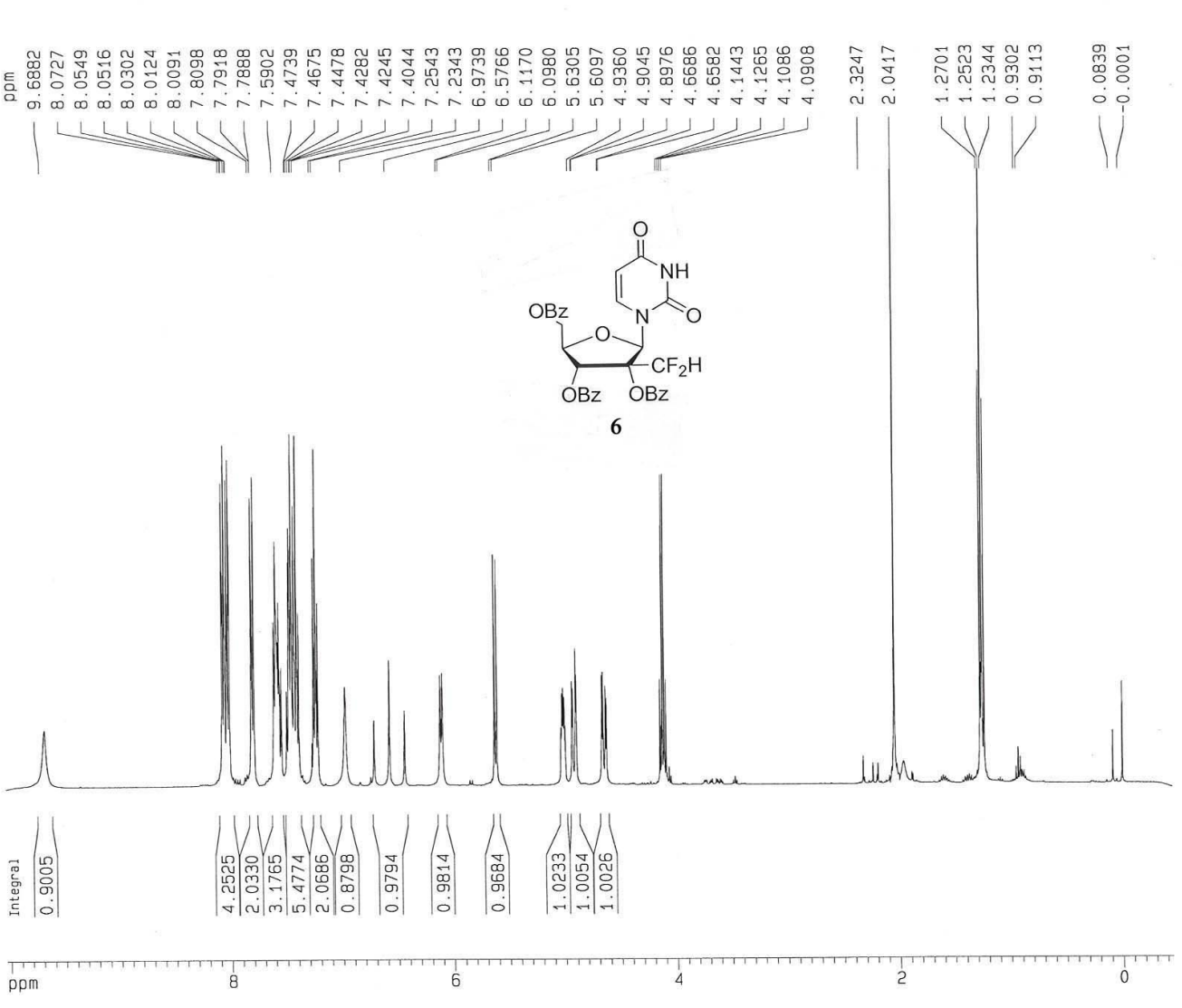

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PROCNO                  1

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PULPROG                zg
TD                  32768
SOLVENT             CDCl3
NS                      4
DS                      0
SWH            4194.631 Hz
FIDRES         0.128010 Hz
AQ             3.9059956 sec
RG                   22.6
DW                119.200 usec
DE                  4.50 usec
TE                 300.0 K
D1             2.00000000 sec

============ CHANNEL f1 =========
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P1                 10.50 usec
PL1                 0.00 dB
SFO1          400.1319246 MHz

F2 - Processing parameters
SI                     16384
SF            400.1300057 MHz
WDW                    EM
SSB                     0
LB                   0.30 Hz
GB                      0
PC                   1.00

1D NMR plot parameters
CX                  20.00 cm
F1P                 10.037 ppm
F1                 4016.18 Hz
F2P                 -0.446 ppm
F2                 -178.45 Hz
PPMCM              0.52416 ppm/cm
HZCM             209.73154 Hz/cm
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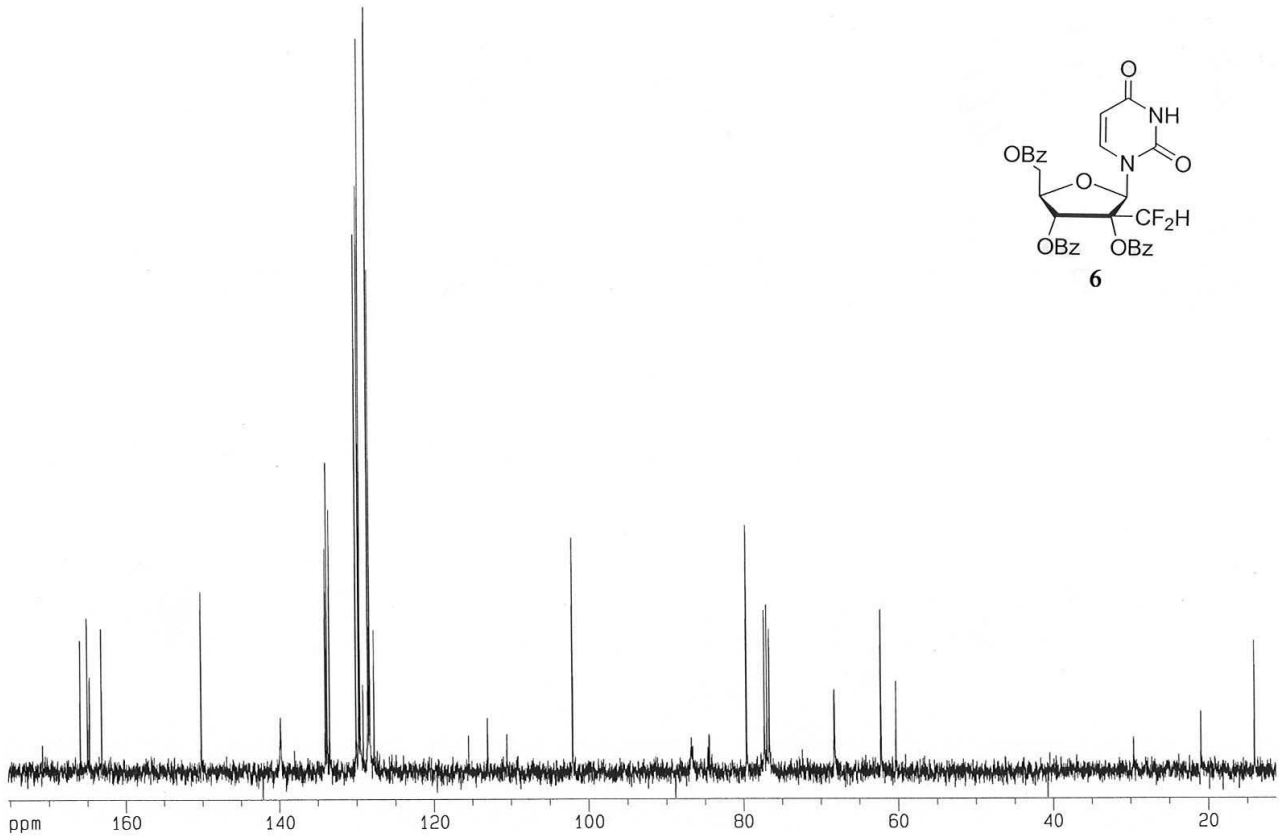

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Current Data Parameters
NAME                  B19_c
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
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PULPROG               zgdc
TD                   32768
SOLVENT              CDCl3
NS                     158
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FIDRES            0.827035 Hz
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RG                   1149.4
DW                   18.450 usec
DE                    7.50 usec
TE                   300.0 K
D1             3.00000000 sec
d11            0.03000000 sec

============ CHANNEL f1 ==========
NUC1                   13C
P1                   11.00 usec
PL1                  -2.00 dB
SFO1           100.6254358 MHz

============ CHANNEL f2 ==========
CPDPRG2            waltz16
NUC2                   1H
PCPD2               101.00 usec
PL2                  120.00 dB
PL12                  19.00 dB
SFO2           400.1324710 MHz

F2 - Processing parameters
SI                    16384
SF             100.6127793 MHz
WDW                      EM
SSB                       0
LB                     1.00 Hz
GB                        0
PC                     1.00

1D NMR plot parameters
CX                    20.00 cm
F1P                  175.273 ppm
F1                 17634.66 Hz
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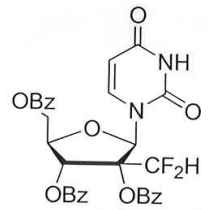

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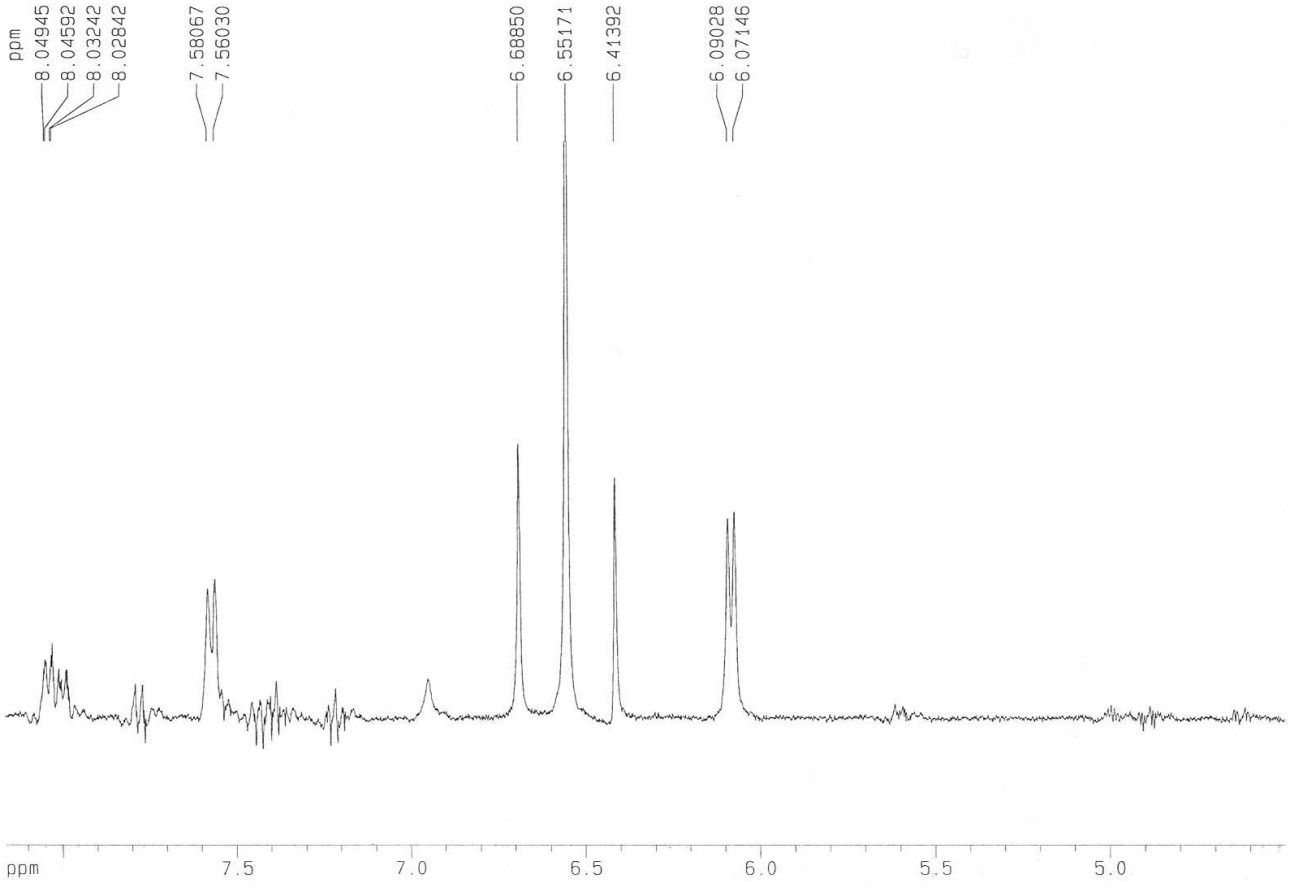


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PROCNO                       2

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TD                        4698
SOLVENT                   CDCl3
NS                           80
DS                            4
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FIDRES                0.343761 Hz
AQ                    1.4545507 sec
RG                         71.8
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DE                        4.50 usec
TE                       300.0 K
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d12                   0.00002000 sec
D20                   3.00000000 sec
L4                           1

======== CHANNEL f1 ========
NUC1                        1H
P1                       10.50 usec
PL1                       0.00 dB
SF01                400.1325855 MHz

======== CHANNEL f2 ========
CPDPRG2                 mlev16
FQ1LIST                 jyq.1
FQ2LIST                 jyq.1
NUC2                        19F
PCPD2                   100.00 usec
PL2                     120.00 dB
PL12                     24.00 dB
PL14                     60.00 dB
SF02                376.4120598 MHz

F2 - Processing parameters
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SF                  400.1300177 MHz
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SSB                           0
LB                         0.30 Hz
GB                            0
PC                         1.00

1D NMR plot parameters
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F2P                       4.500 ppm
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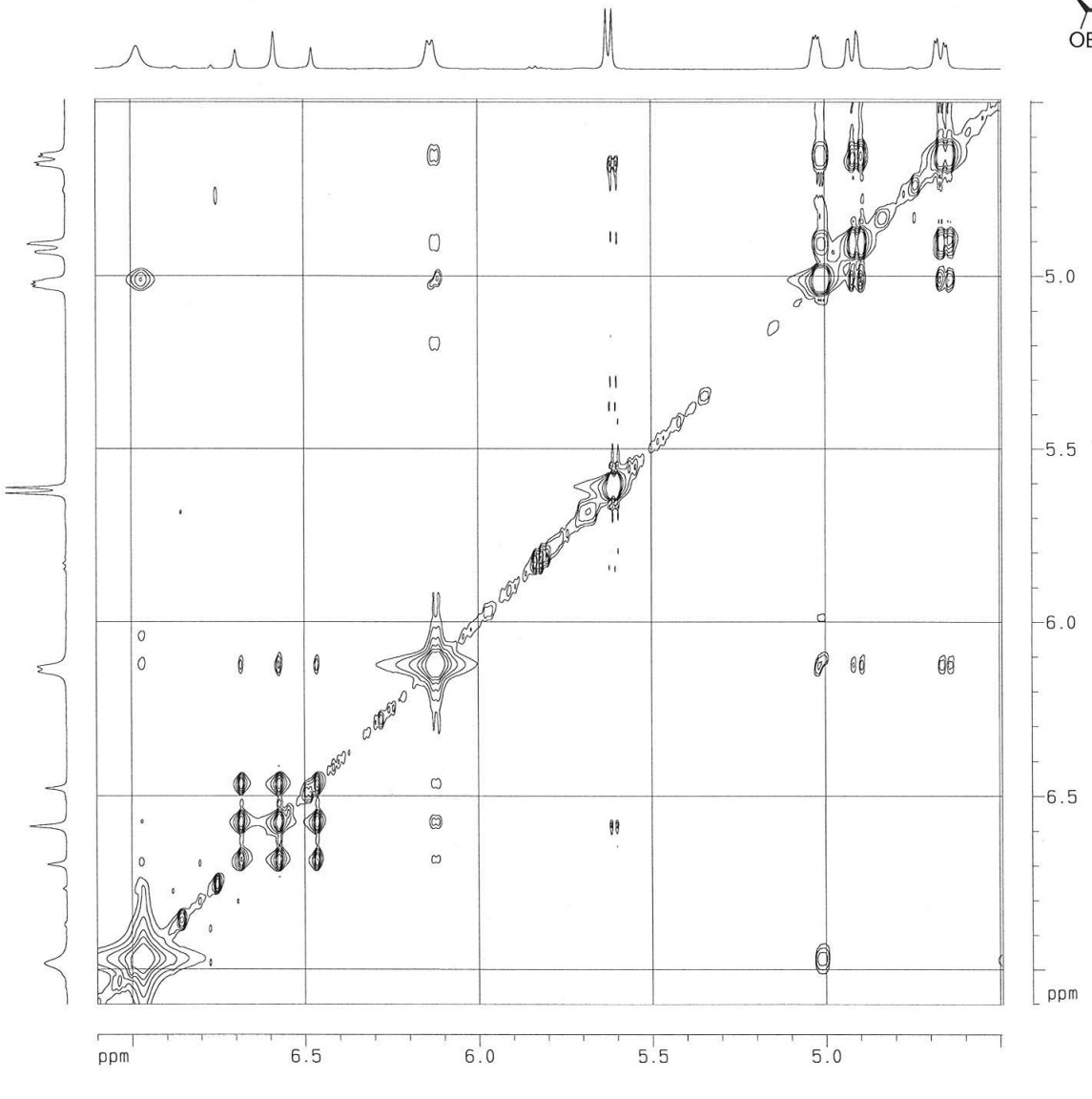

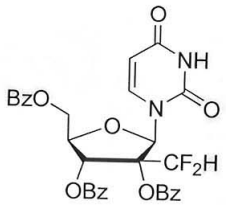

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B23_noesy

Current Data Parameters

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PROCNO	1

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PULPROG	noesytp
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NS	16
DS	16
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========== CHANNEL f1 ==========

NUC1	1H
P1	6.00 usec
PL1	0.00 dB
SFO1	500.1332065 MHz

F1 - Acquisition parameters

ND0	2
TD	256
SFO1	500.1332 MHz
FIDRES	7.398201 Hz
SW	3.787 ppm

F2 - Processing parameters

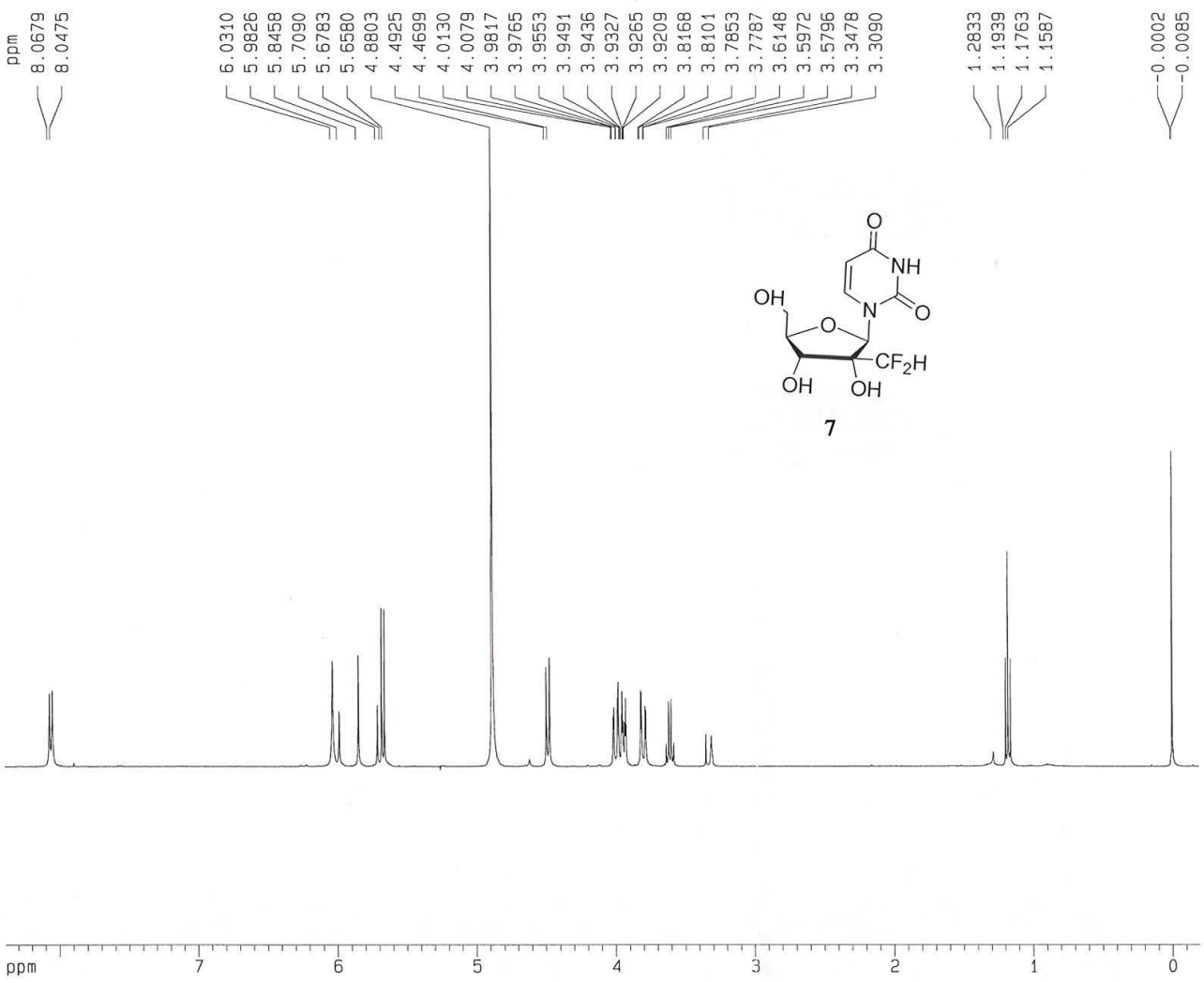
SI	2048
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

SI	1024
MC2	TPPI
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0

2D NMR plot parameters

CX2	15.00 cm
CX1	15.00 cm
F2PLO	7.100 ppm
F2LO	3551.11 Hz
F2PHI	4.493 ppm
F2HI	2247.18 Hz
F1PLO	7.102 ppm
F1LO	3552.04 Hz
F1PHI	4.491 ppm
F1HI	2246.25 Hz
F2PPMCM	0.17381 ppm/cm
F2HZCM	86.92886 Hz/cm
F1PPMCM	0.17406 ppm/cm
F1HZCM	87.05216 Hz/cm



Current Data Parameters

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EXPNO	1
PROCNO	1

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INSTRUM	spect
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PULPROG	zg
TD	39046
SOLVENT	MeOH
NS	8
DS	0
SWH	4807.692 Hz
FIDRES	0.123129 Hz
AQ	4.0608339 sec
RG	2.2
DW	104.000 usec
DE	7.00 usec
TE	300.0 K
D1	2.00000000 sec
P1	7.70 usec
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NUC1	1H
PL1	-6.00 dB

F2 - Processing parameters

SI	32768
SF	400.1300073 MHz
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SSB	0
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1D NMR plot parameters

CX	20.00 cm
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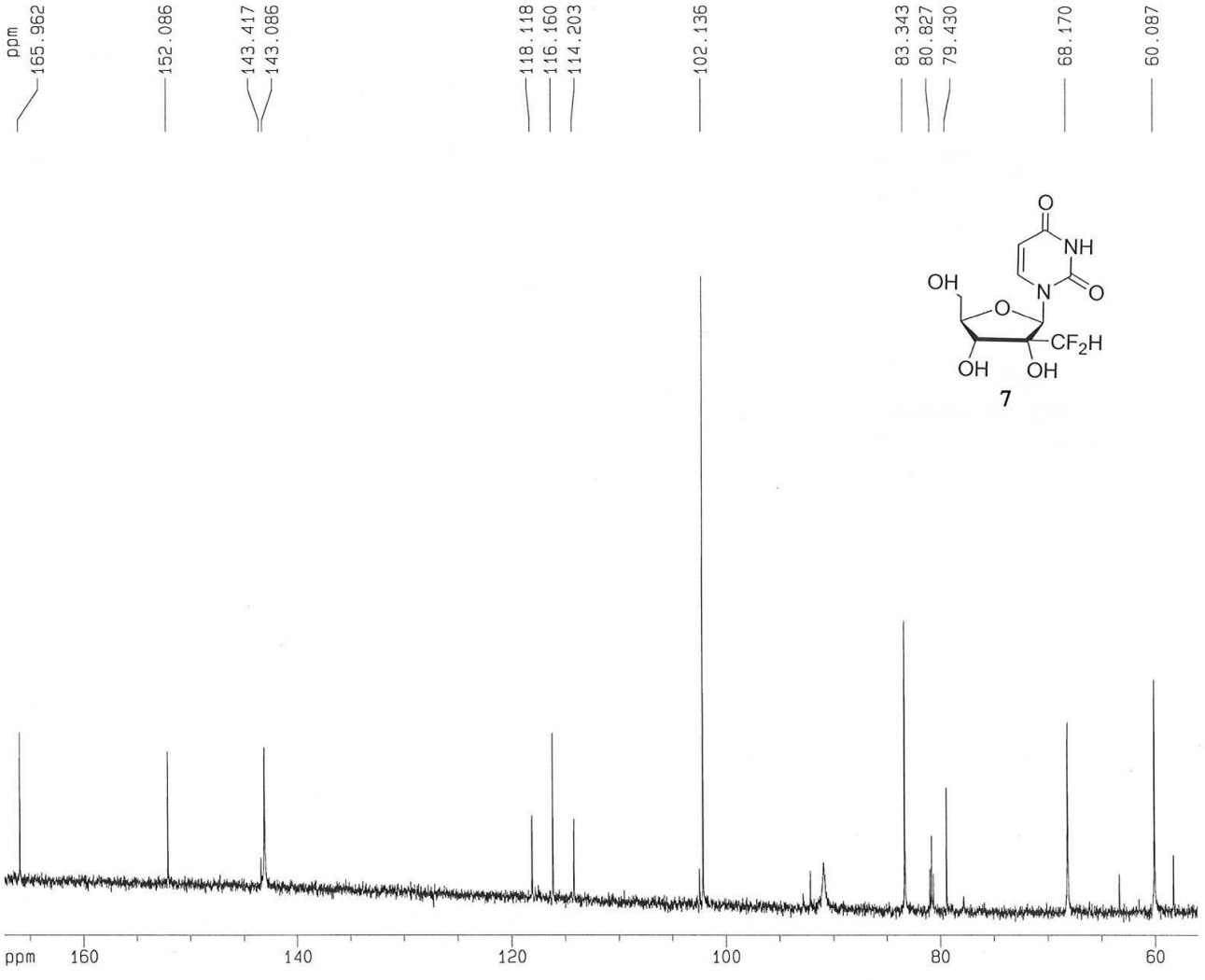
97/10/17, #5, 122.5 mg



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EXPNO                      1
PROCNO                     1

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PULPROG                zgdc
TD                    65536
SOLVENT               Aceton
NS                       205
DS                         0
SWH               39682.539 Hz
FIDRES             0.605507 Hz
AQ                 0.8258036 sec
RG                       512
DW                    12.600 usec
DE                     7.50 usec
TE                    300.0 K
D1                 3.00000000 sec
d11                0.03000000 sec

============ CHANNEL f1 ==========
NUC1                      13C
P1                     8.20 usec
PL1                    0.00 dB
SFO1              125.7736214 MHz

============ CHANNEL f2 ==========
CPDPRG2              waltz16
NUC2                      1H
PCPD2                 90.00 usec
PL2                  120.00 dB
PL12                  19.00 dB
SFO2              500.1338000 MHz

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SF                125.7576186 MHz
WDW                        EM
SSB                        0
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PC                     1.40

1D NMR plot parameters
CX                    20.00 cm
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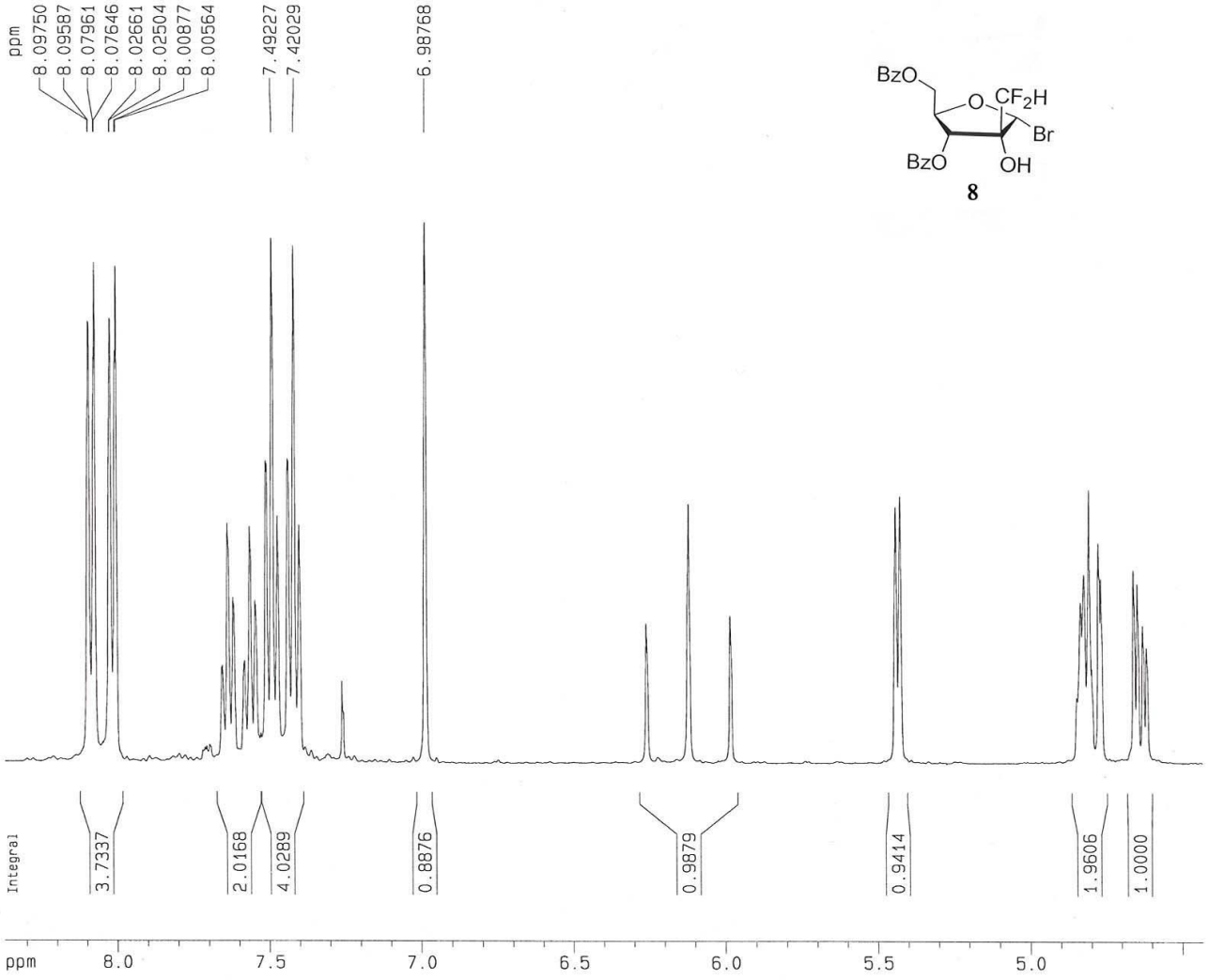

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Current Data Parameters

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PROCNO	1

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PULPROG	zg
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NS	8
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FIDRES	0.123129 Hz
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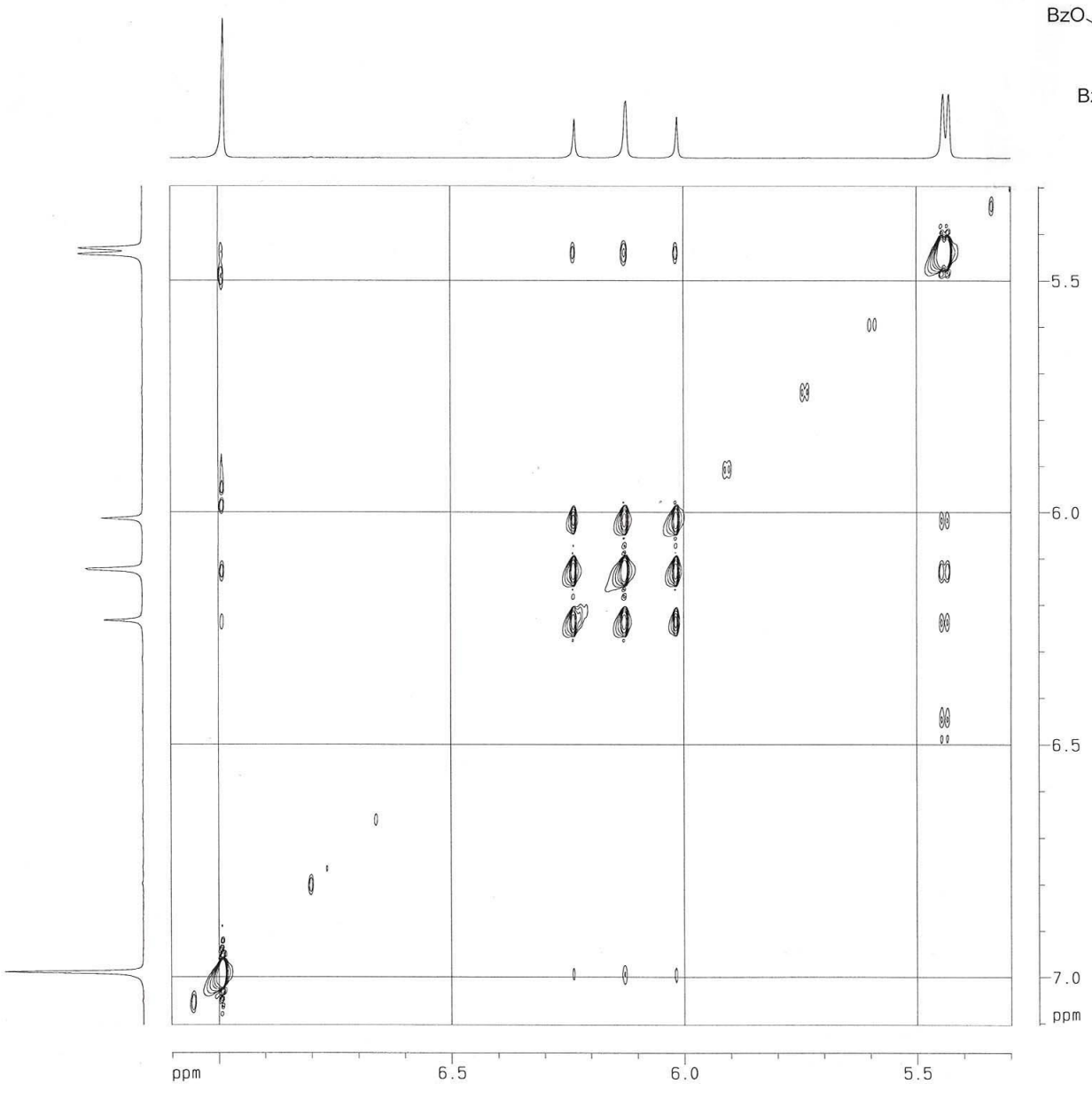
DE	7.00 usec
TE	300.0 K
D1	2.00000000 sec
P1	7.70 usec
SFO1	400.1317512 MHz
NUC1	1H
PL1	-6.00 dB

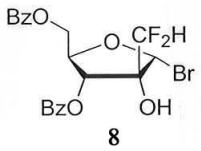
F2 - Processing parameters

SI	32768
SF	400.1300089 MHz
WDW	EM
SSB	0
LB	0.30 Hz
GB	0
PC	1.00

1D NMR plot parameters

CX	20.00 cm
F1P	8.373 ppm
F1	3350.16 Hz
F2P	4.434 ppm
F2	1774.13 Hz
PPMCM	0.19694 ppm/cm
HZCM	78.80158 Hz/cm





8

Current Data Parameters

NAME	B44
EXPNO	3
PROCNO	1

F2 - Acquisition Parameters

Date_	20020805
Time	23.26
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	noesytp
TD	2048
SOLVENT	CDCl3
NS	32
DS	16
SWH	2042.484 Hz
FIDRES	0.997306 Hz
AQ	0.5014004 sec
RG	20
DW	244.800 usec
DE	4.50 usec
TE	300.0 K
d0	0.00000300 sec
D1	2.00000000 sec
DB	0.75000000 sec
INO	0.00024480 sec

========== CHANNEL f1 ==========

NUC1	1H
P1	6.00 usec
PL1	0.00 dB
SFO1	500.1332173 MHz

F1 - Acquisition parameters

NDO	2
TD	256
SFO1	500.1332 MHz
FIDRES	7.978452 Hz
SW	4.084 ppm

F2 - Processing parameters

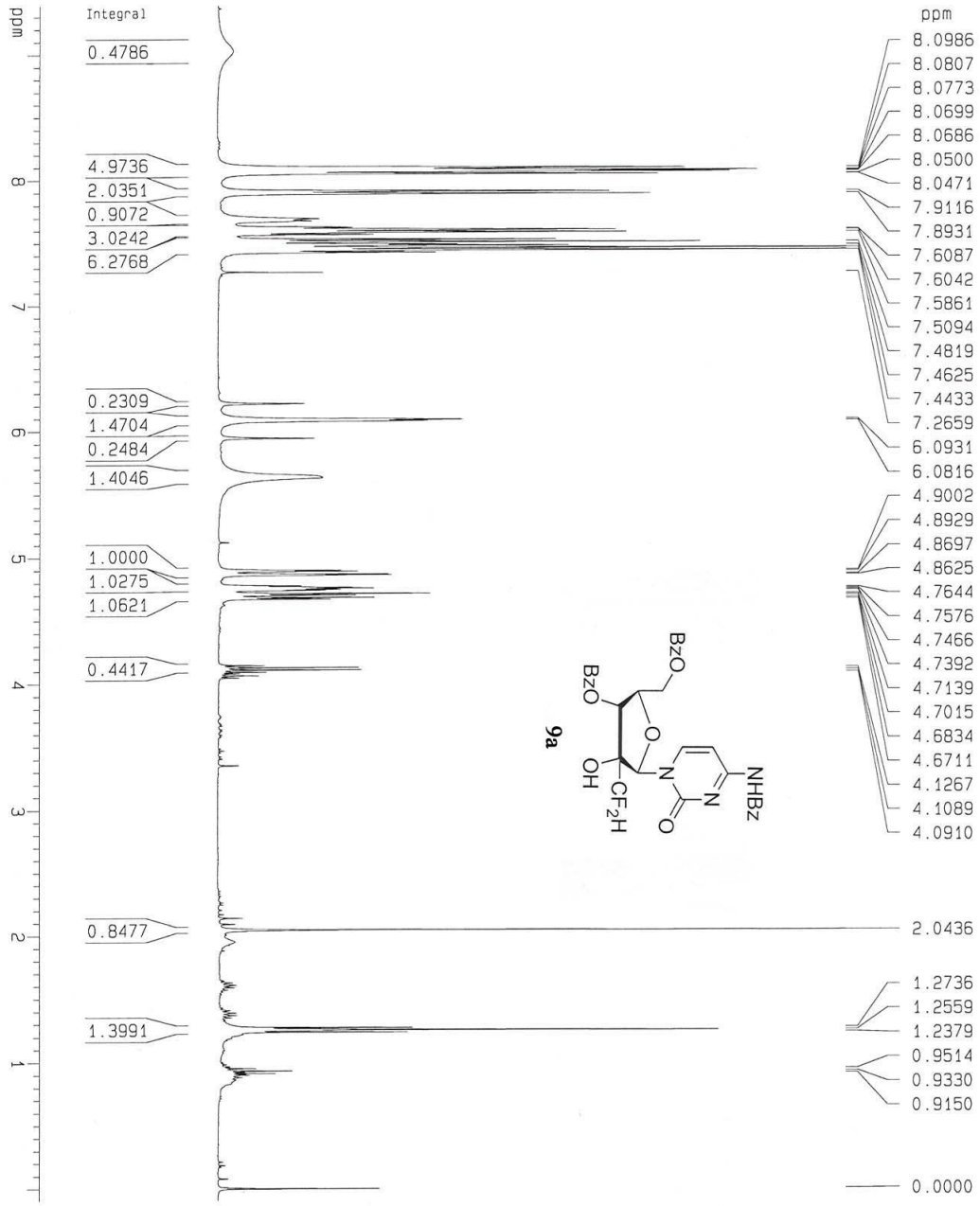
SI	2048
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

SI	1024
MC2	TPPI
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0

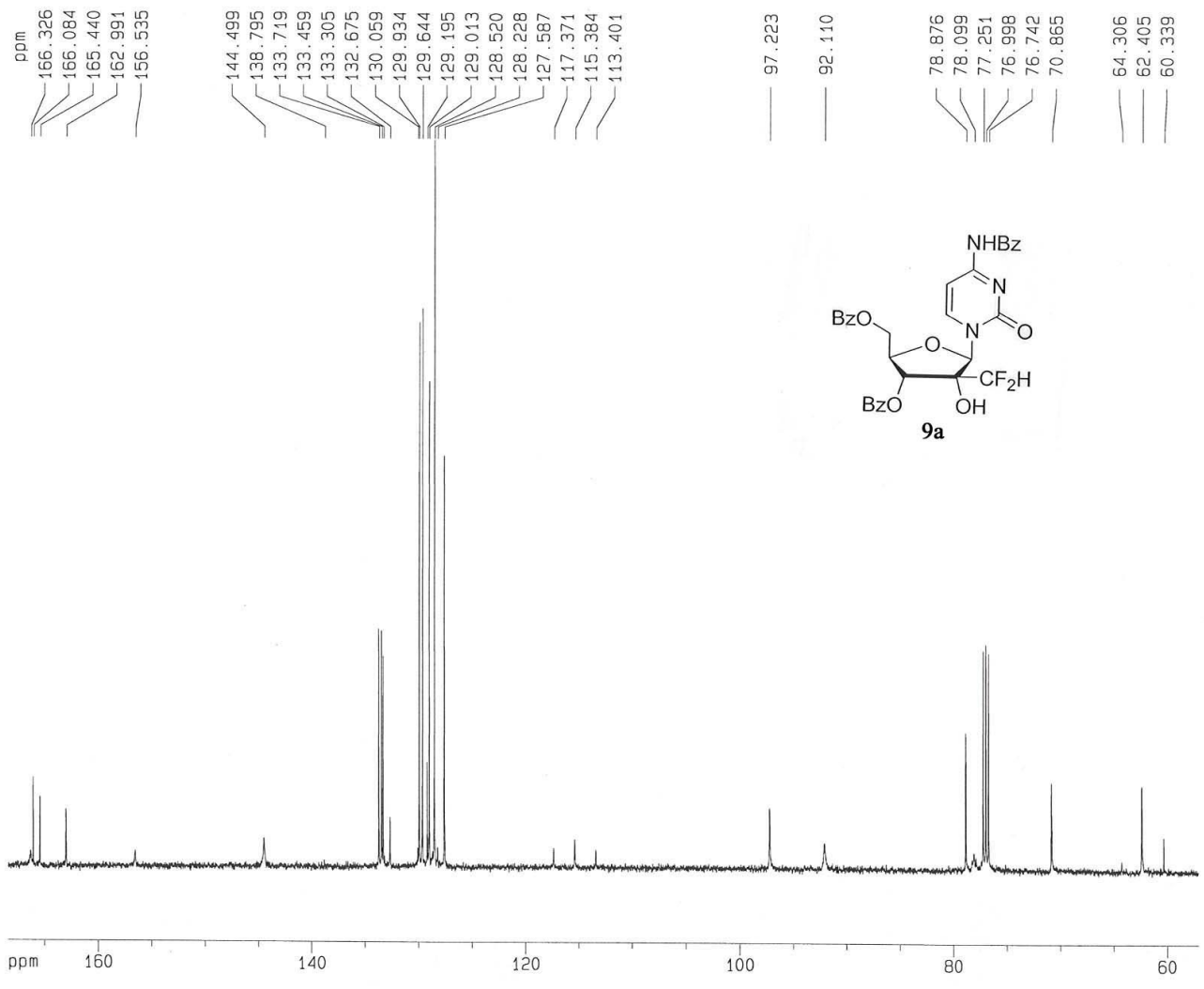
2D NMR plot parameters

CX2	15.00 cm
CX1	15.00 cm
F2PLO	7.102 ppm
F2LO	3551.90 Hz
F2PHI	5.299 ppm
F2HI	2650.34 Hz
F1PLO	7.104 ppm
F1LO	3552.90 Hz
F1PHI	5.297 ppm
F1HI	2649.34 Hz
F2PPMCM	0.12018 ppm/cm
F2HZCM	60.10434 Hz/cm
F1PPMCM	0.12044 ppm/cm
F1HZCM	60.23732 Hz/cm



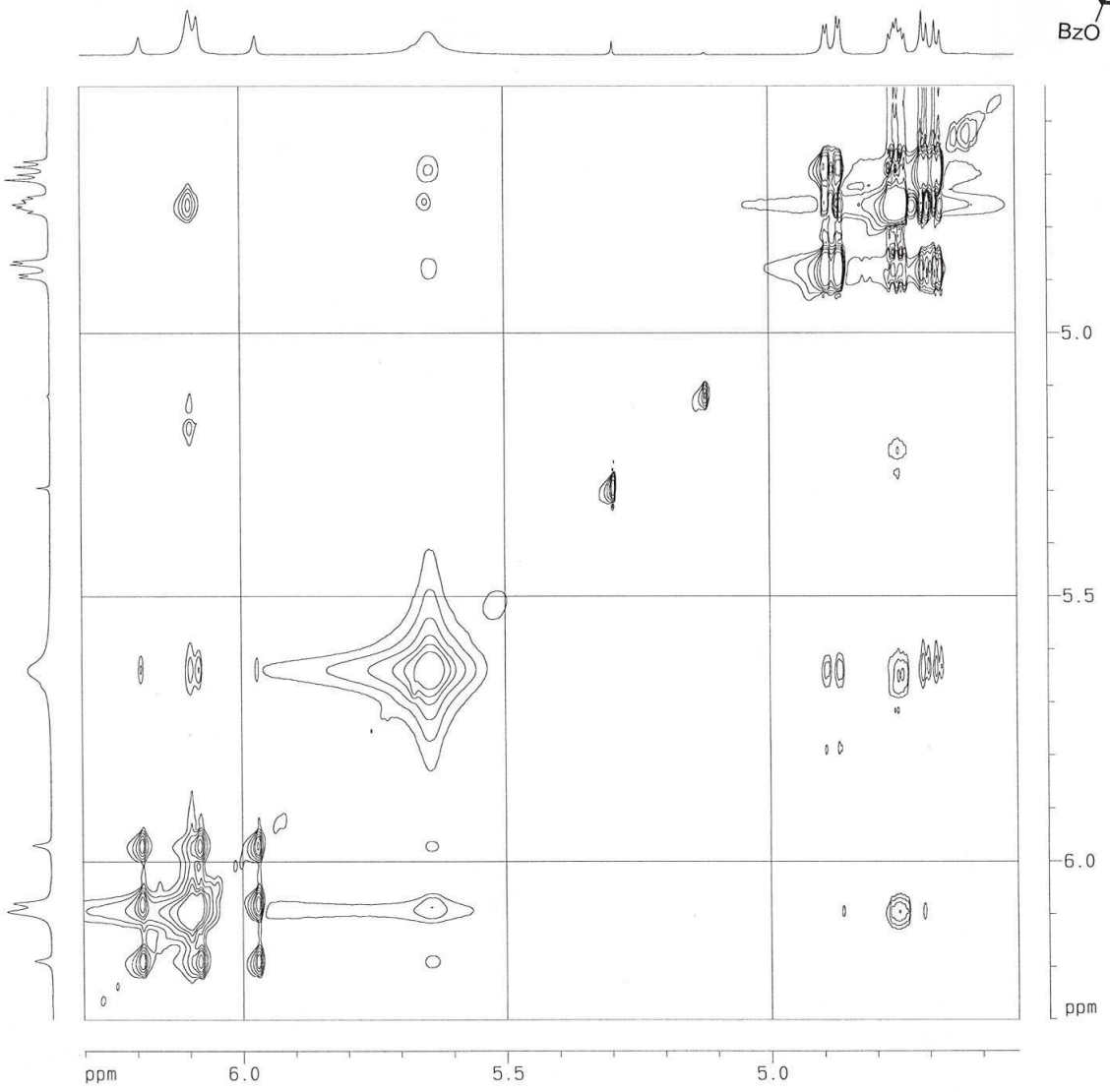

```
Current Data Parameters
NAME                  c20b
EXPNO                    1
PROCNO                   1

F2 - Acquisition Parameters
Date_              20030224
Time                  14.30
INSTRUM                spect
PROBHD     5 mm QNP 1H
PULPROG                zg
TD                    32768
SOLVENT               CDCl3
NS                        8
DS                        0
SWH               4807.692 Hz
FIDRES            0.146719 Hz
AQ                3.4079220 sec
RG                       32
DW                104.000 usec
DE                  4.50 usec
TE                  300.0 K
D1                2.00000000 sec

======== CHANNEL f1 ========
NUC1                     1H
P1                   10.50 usec
PL1                   0.00 dB
SFO1           400.1319246 MHz

F2 - Processing parameters
SI                      16384
SF            400.1300072 MHz
WDW                        EM
SSB                         0
LB                   0.30 Hz
GB                          0
PC                       1.00

1D NMR plot parameters
CX                      20.00 cm
F1P                   9.389 ppm
F1                 3756.79 Hz
F2P                 -0.093 ppm
F2                  -37.11 Hz
PPMCM              0.47408 ppm/cm
HZCM             189.69481 Hz/cm
```

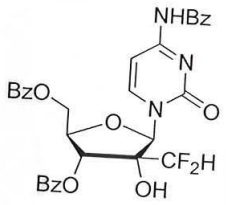
97/10/17, #5, 122.5 mg



9a

```
Current Data Parameters
NAME                  c20b_C
EXPNO                      1
PROCNO                     1

F2 - Acquisition Parameters
Date_              20030314
Time                  18.20
INSTRUM               spect
PROBHD      5 mm GNP  1H
PULPROG               zgdc
TD                    65536
SOLVENT               CDCl3
NS                      468
DS                        0
SWH               39682.539 Hz
FIDRES             0.605507 Hz
AQ                 0.8258036 sec
RG                      512
DW                   12.600 usec
DE                    7.50 usec
TE                   300.0 K
D1                3.00000000 sec
d11               0.03000000 sec

============ CHANNEL f1 ==========
NUC1                    13C
P1                     8.20 usec
PL1                    0.00 dB
SFO1             125.7736214 MHz

============ CHANNEL f2 ==========
CPDPRG2              waltz16
NUC2                    1H
PCPD2                90.00 usec
PL2                  120.00 dB
PL12                  19.00 dB
SFO2             500.1338000 MHz

F2 - Processing parameters
SI                      32768
SF               125.7577982 MHz
WDW                        EM
SSB                        0
LB                     1.00 Hz
GB                         0
PC                      1.40

1D NMR plot parameters
CX                    20.00 cm
F1P                  168.443 ppm
F1                 21183.00 Hz
F2P                   57.086 ppm
F2                 7178.98 Hz
PPMCM                5.56785 ppm/cm
HZCM               700.20074 Hz/cm
```





9a

Current Data Parameters

NAME	c20_2d
EXPNO	3
PROCNO	1

F2 - Acquisition Parameters

Date_	20030416
Time	23.27
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	noesytp
TD	2048
SOLVENT	CDCl3
NS	32
DS	16
SWH	1860.119 Hz
FIDRES	0.908261 Hz
AQ	0.5505524 sec
RG	20
DW	268.800 usec
DE	4.50 usec
TE	300.0 K
d0	0.00000300 sec
D1	2.00000000 sec
DB	0.75000000 sec
IN0	0.00026880 sec

========== CHANNEL f1 ==========

NUC1	1H
P1	6.00 usec
PL1	0.00 dB
SF01	500.1332083 MHz

F1 - Acquisition parameters

ND0	2
TD	256
SF01	500.1332 MHz
FIDRES	7.266090 Hz
SW	3.719 ppm

F2 - Processing parameters

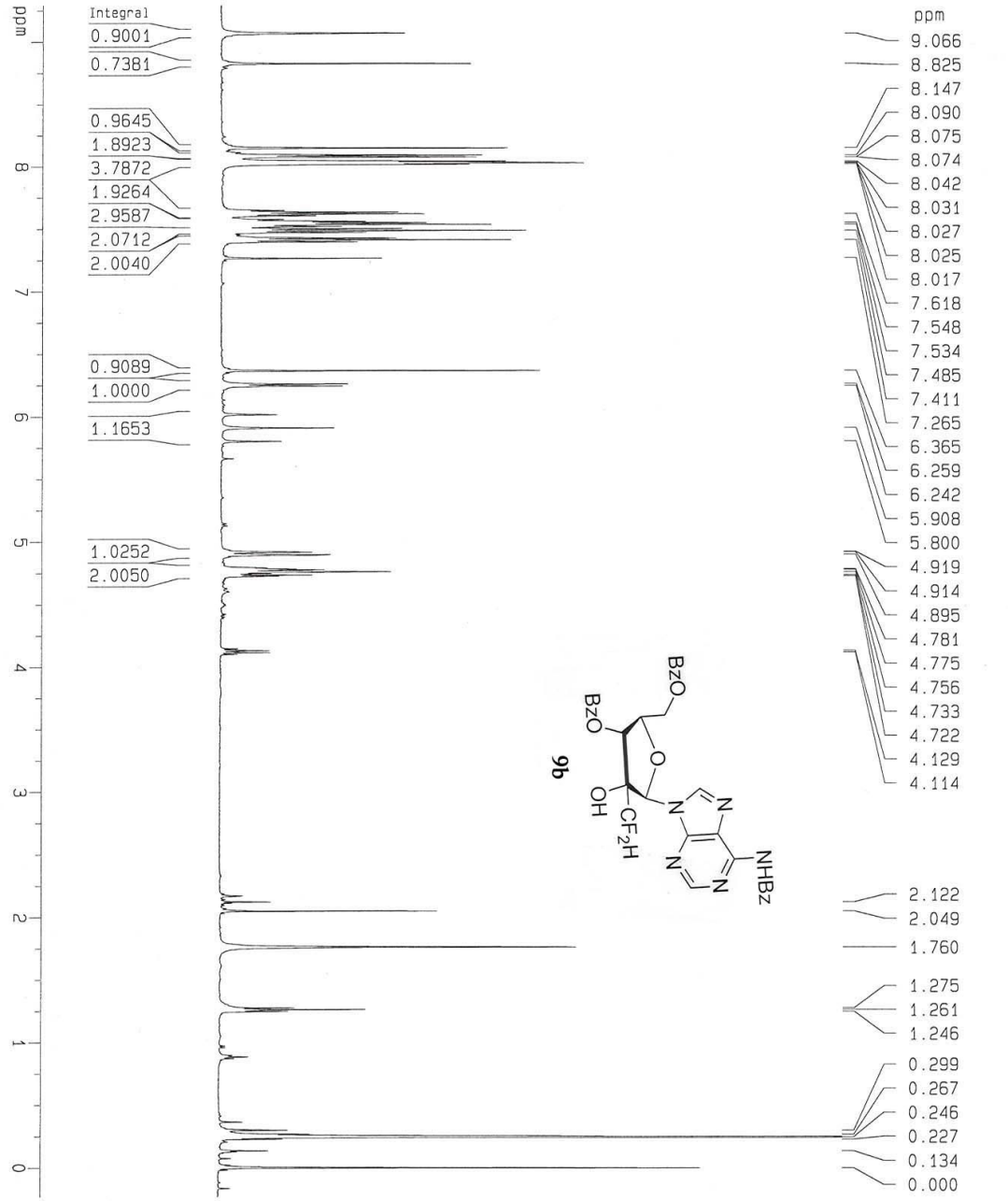
SI	2048
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

SI	1024
MC2	TPPI
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0

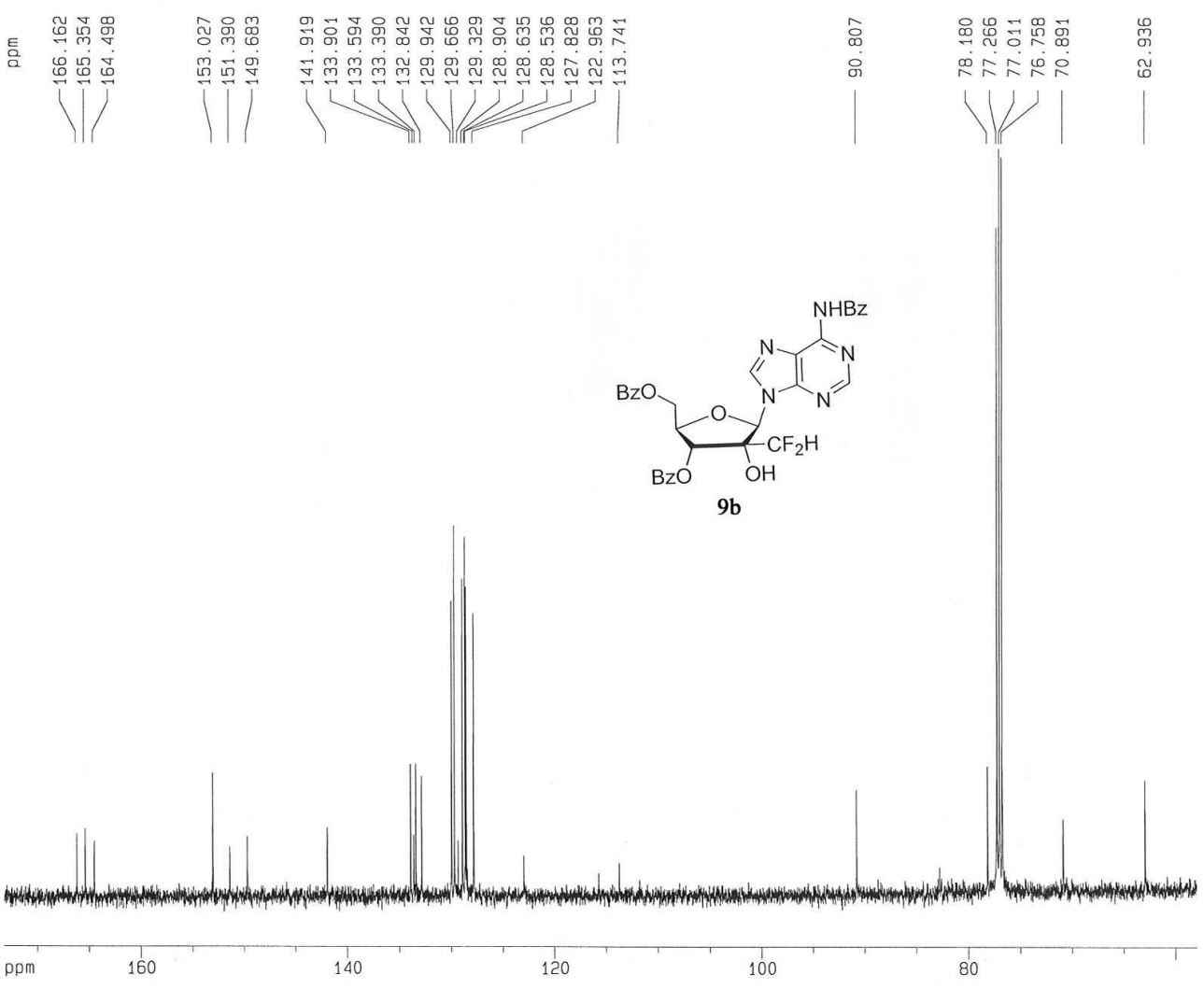
2D NMR plot parameters

CX2	15.00 cm
CX1	15.00 cm
F2PLO	6.301 ppm
F2LO	3151.49 Hz
F2PHI	4.533 ppm
F2HI	2266.84 Hz
F1PLO	6.301 ppm
F1LO	3151.49 Hz
F1PHI	4.533 ppm
F1HI	2266.84 Hz
F2PPMCM	0.11792 ppm/cm
F2HZCM	58.97644 Hz/cm
F1PPMCM	0.11792 ppm/cm
F1HZCM	58.97644 Hz/cm

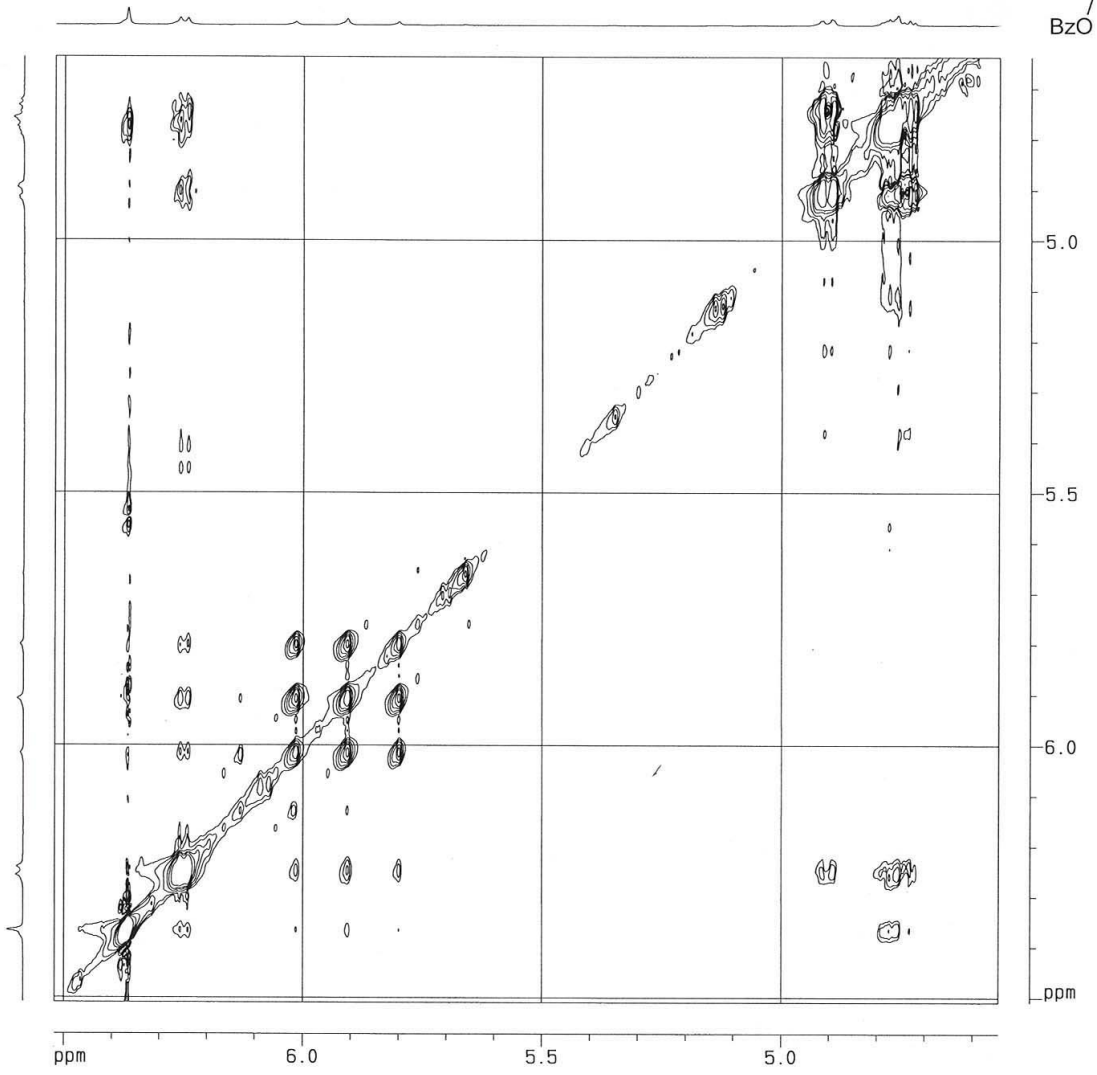


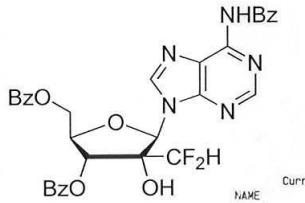

```
Current Data Parameters
NAME                  D23a7
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
Date_              20030915
Time                  21.25
INSTRUM               spect
PROBHD     5 mm QNP 1H
PULPROG                zg
TD                    32768
SOLVENT               CDCl3
NS                        8
DS                        0
SWH              6009.615 Hz
FIDRES           0.183399 Hz
AQ               2.7263477 sec
RG                       128
DW                  83.200 usec
DE                   4.50 usec
TE                  300.0 K
D1              2.00000000 sec

======== CHANNEL f1 ========
NUC1                   1H
P1                 10.00 usec
PL1                 0.00 dB
SFO1         500.1320118 MHz

F2 - Processing parameters
SI                    16384
SF           500.1300113 MHz
WDW                      EM
SSB                       0
LB                  0.30 Hz
GB                        0
PC                     1.00

1D NMR plot parameters
CX                   20.00 cm
F1P                 9.292 ppm
F1                4647.13 Hz
F2P                -0.231 ppm
F2                 -115.45 Hz
PPMCM             0.47613 ppm/cm
HZCM           238.12936 Hz/cm
```

97/10/17, #5, 122.5 mg




```
Current Data Parameters
NAME                 D23a7c
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
Date_              20030915
Time                 21.48
INSTRUM              spect
PROBHD     5 mm QNP  1H
PULPROG              zgdc
TD                   65536
SOLVENT              Aceton
NS                     510
DS                       0
SWH              39682.539 Hz
FIDRES           0.605507 Hz
AQ               0.8258036 sec
RG                    2048
DW                 12.600 usec
DE                  7.50 usec
TE                 300.0 K
D1               3.00000000 sec
d11              0.03000000 sec

============ CHANNEL f1 ==========
NUC1                  13C
P1                  8.20 usec
PL1                 0.00 dB
SFO1           125.7736214 MHz

============ CHANNEL f2 ==========
CPDPRG2           waltz16
NUC2                  1H
PCPD2               90.00 usec
PL2                120.00 dB
PL12                19.00 dB
SFO2           500.1338000 MHz

F2 - Processing parameters
SI                    32768
SF             125.7577934 MHz
WDW                   EM
SSB                      0
LB                  1.00 Hz
GB                       0
PC                  1.40

1D NMR plot parameters
CX                  20.00 cm
F1P                173.228 ppm
F1                21784.80 Hz
F2P                 57.946 ppm
F2                 7287.10 Hz
PPMCM              5.76414 ppm/cm
HZCM             724.88531 Hz/cm
```





9b

Current Data Parameters

NAME	D23_2d
EXPNO	3
PROCNO	1

F2 - Acquisition Parameters

Date_	20030918
Time	21.32
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	noesytp
TD	2048
SOLVENT	CDCl3
NS	40
DS	16
SWH	1627.604 Hz
FIDRES	0.794729 Hz
AQ	0.6291956 sec
RG	20
DW	307.200 usec
DE	4.50 usec
TE	300.0 K
d0	0.00000300 sec
D1	2.00000000 sec
D8	0.75000000 sec
IN0	0.00030720 sec

========== CHANNEL f1 ==========

NUC1	1H
P1	6.00 usec
PL1	0.00 dB
SF01	500.1330971 MHz

F1 - Acquisition parameters

ND0	2
TD	256
SF01	500.1331 MHz
FIDRES	6.357829 Hz
SW	3.254 ppm

F2 - Processing parameters

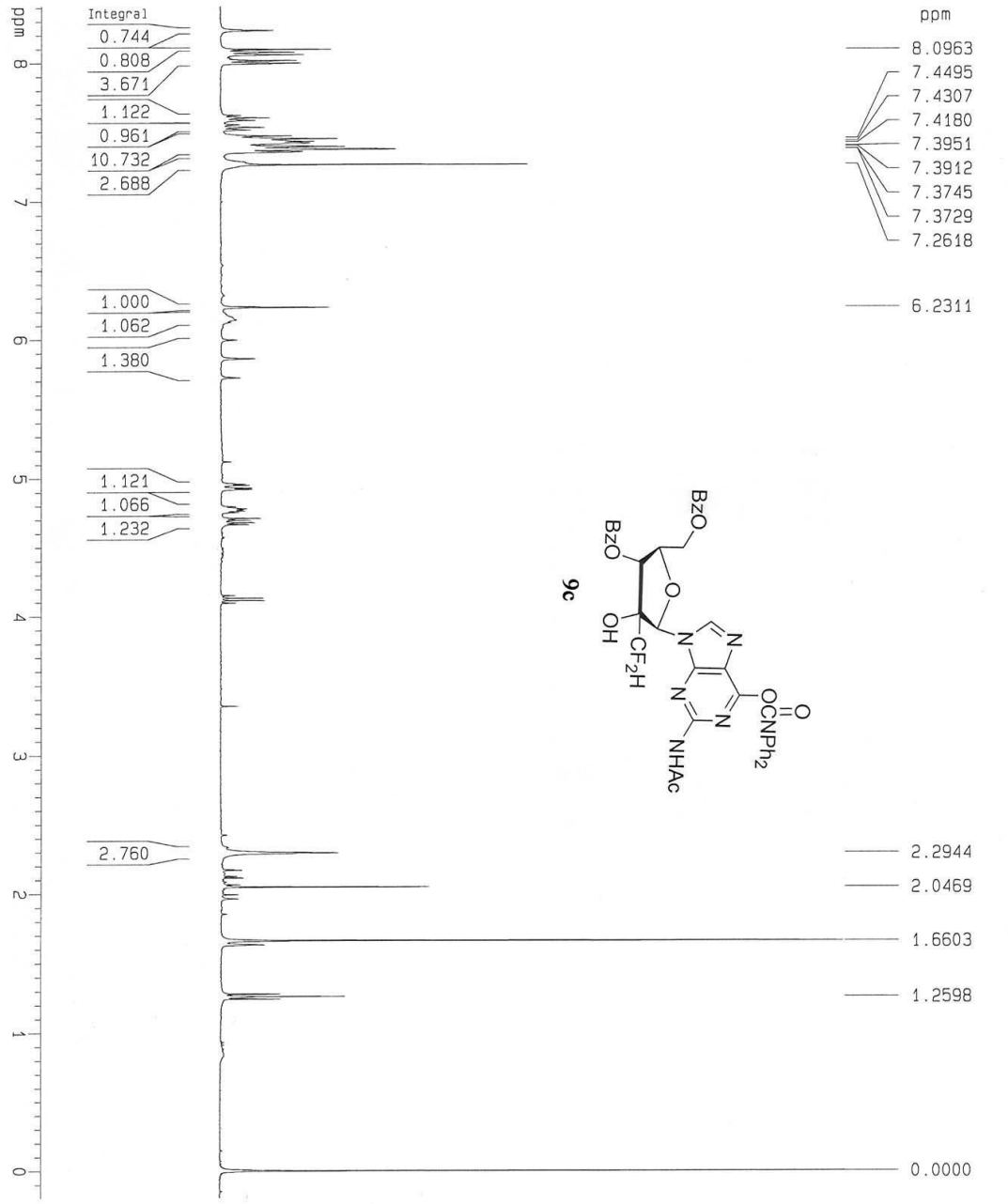
SI	2048
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

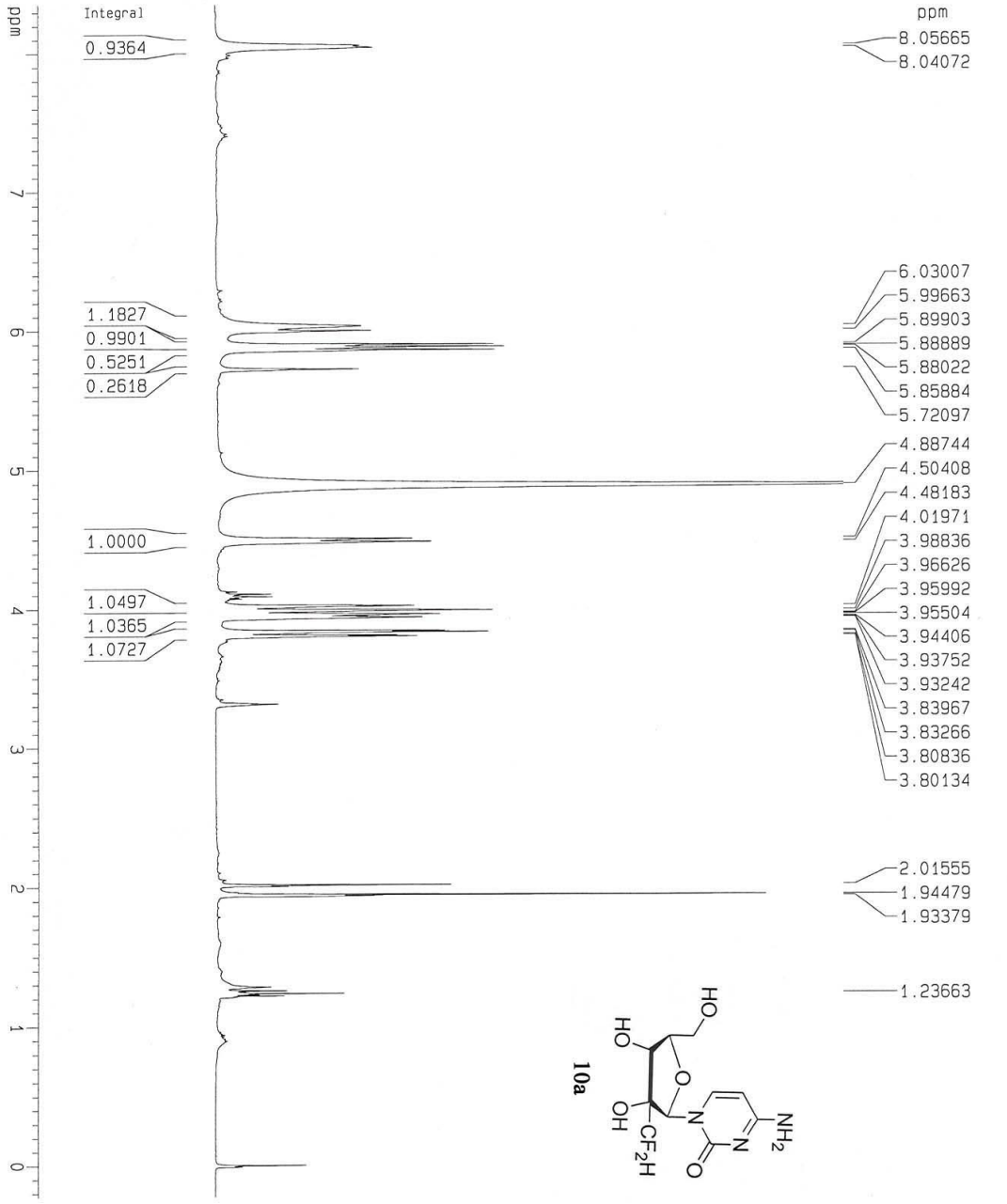
SI	1024
MC2	TPPI
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0

2D NMR plot parameters

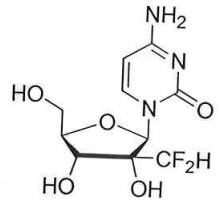
CX2	15.00 cm
CX1	15.00 cm
F2PLO	6.519 ppm
F2LO	3260.50 Hz
F2PHI	4.546 ppm
F2HI	2273.45 Hz
F1PLO	6.510 ppm
F1LO	3255.73 Hz
F1PHI	4.638 ppm
F1HI	2319.54 Hz
F2PPMCM	0.13157 ppm/cm
F2HZCM	65.80353 Hz/cm
F1PPMCM	0.12479 ppm/cm
F1HZCM	62.41269 Hz/cm



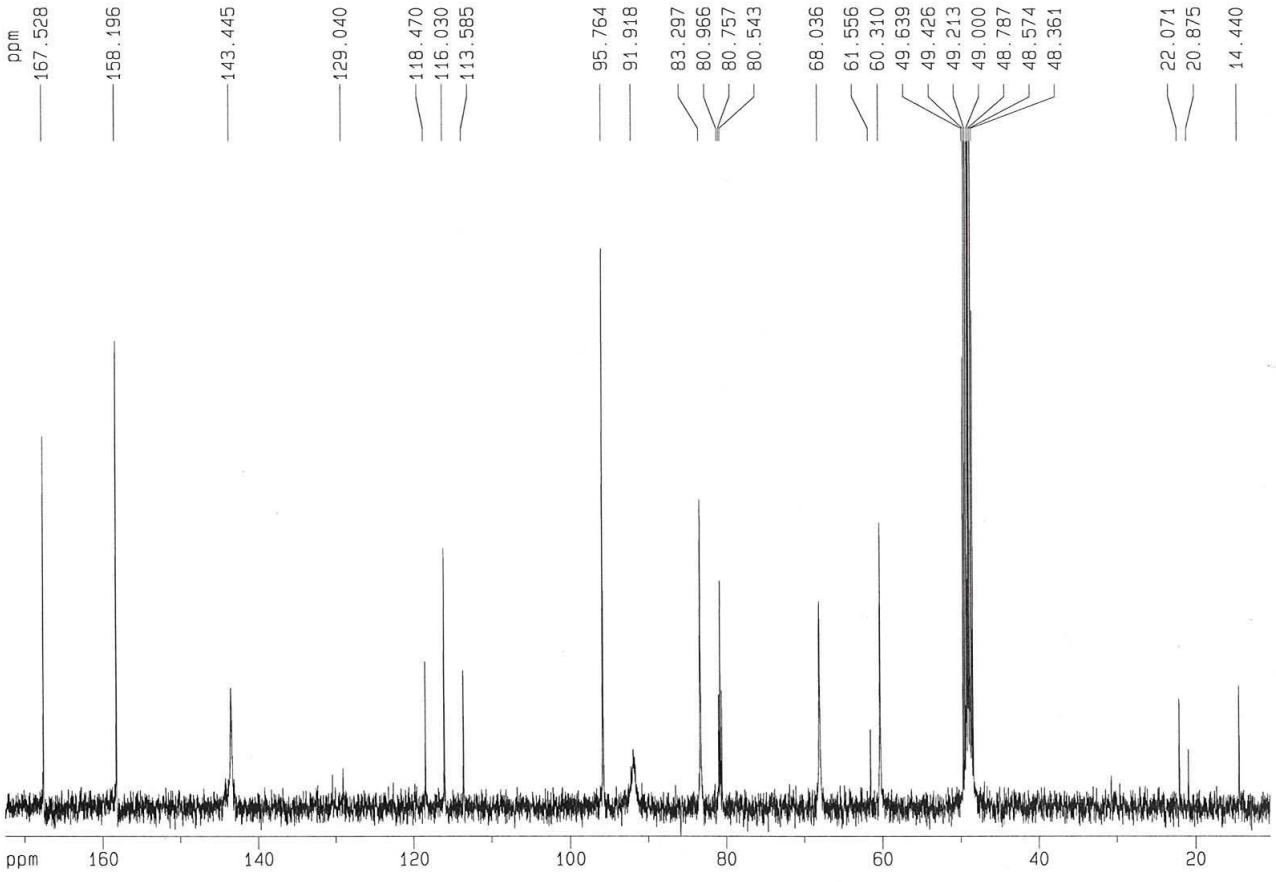

```
Current Data Parameters
NAME                  c59_4
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
Date_              20030416
Time                   19.29
INSTRUM                spect
PROBHD     5 mm QNP  1H
PULPROG                zg
TD                     16384
SOLVENT                CDCl3
NS                         8
DS                         0
SWH                4807.692 Hz
FIDRES             0.293438 Hz
AQ                 1.7039860 sec
RG                      25.4
DW                 104.000 usec
DE                      6.00 usec
TE                     300.0 K
D1                 2.00000000 sec

======== CHANNEL f1 ========
NUC1                       1H
P1                      10.50 usec
PL1                      0.00 dB
SFO1               400.1320007 MHz

F2 - Processing parameters
SI                        32768
SF                 400.1300008 MHz
WDW                       EM
SSB                        0
LB                      0.30 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                     20.00 cm
F1P                     8.413 ppm
F1                   3366.24 Hz
F2P                    -0.194 ppm
F2                     -77.68 Hz
PPMCM                  0.43035 ppm/cm
HZCM                 172.19603 Hz/cm
```



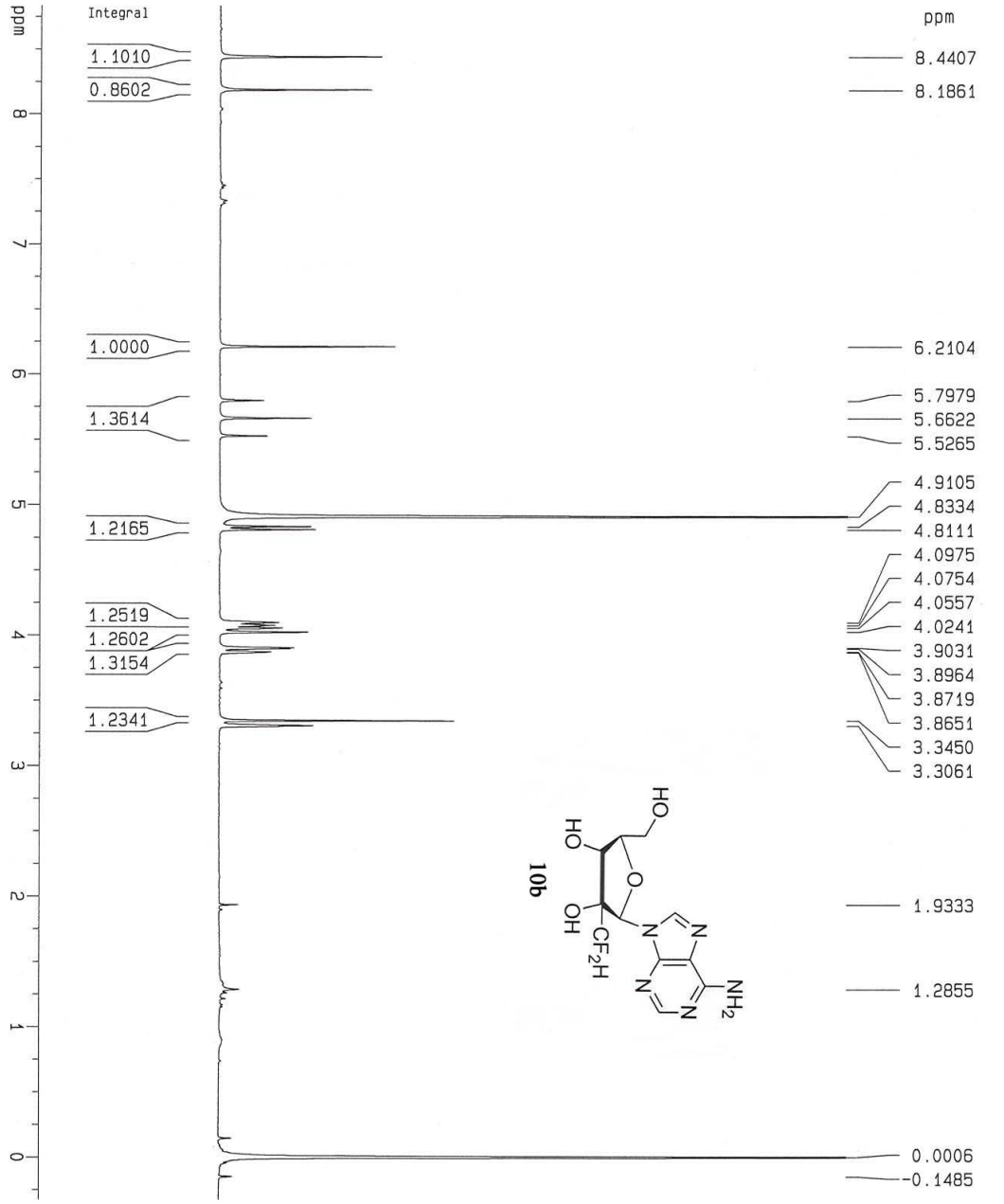

```
Current Data Parameters
NAME                  c30
EXPNO                   1
PROCNO                  1

F2 - Acquisition Parameters
Date_              20030314
Time                 18.01
INSTRUM              spect
PROBHD     5 mm QNP 1H
PULPROG                zg
TD                    16384
SOLVENT               MeOH
NS                       8
DS                       0
SWH               4807.692 Hz
FIDRES            0.293438 Hz
AQ                1.7039860 sec
RG                      25.4
DW                 104.000 usec
DE                   6.00 usec
TE                 300.0 K
D1                 2.00000000 sec

======== CHANNEL f1 ========
NUC1                     1H
P1                   10.50 usec
PL1                   0.00 dB
SFO1          400.1320007 MHz

F2 - Processing parameters
SI                      32768
SF            400.1300063 MHz
WDW                        EM
SSB                         0
LB                   0.30 Hz
GB                          0
PC                   1.00

1D NMR plot parameters
CX                  20.00 cm
F1P                 8.354 ppm
F1                3342.60 Hz
F2P                -0.227 ppm
F2                 -90.72 Hz
PPMCM             0.42903 ppm/cm
HZCM              171.66595 Hz/cm
```



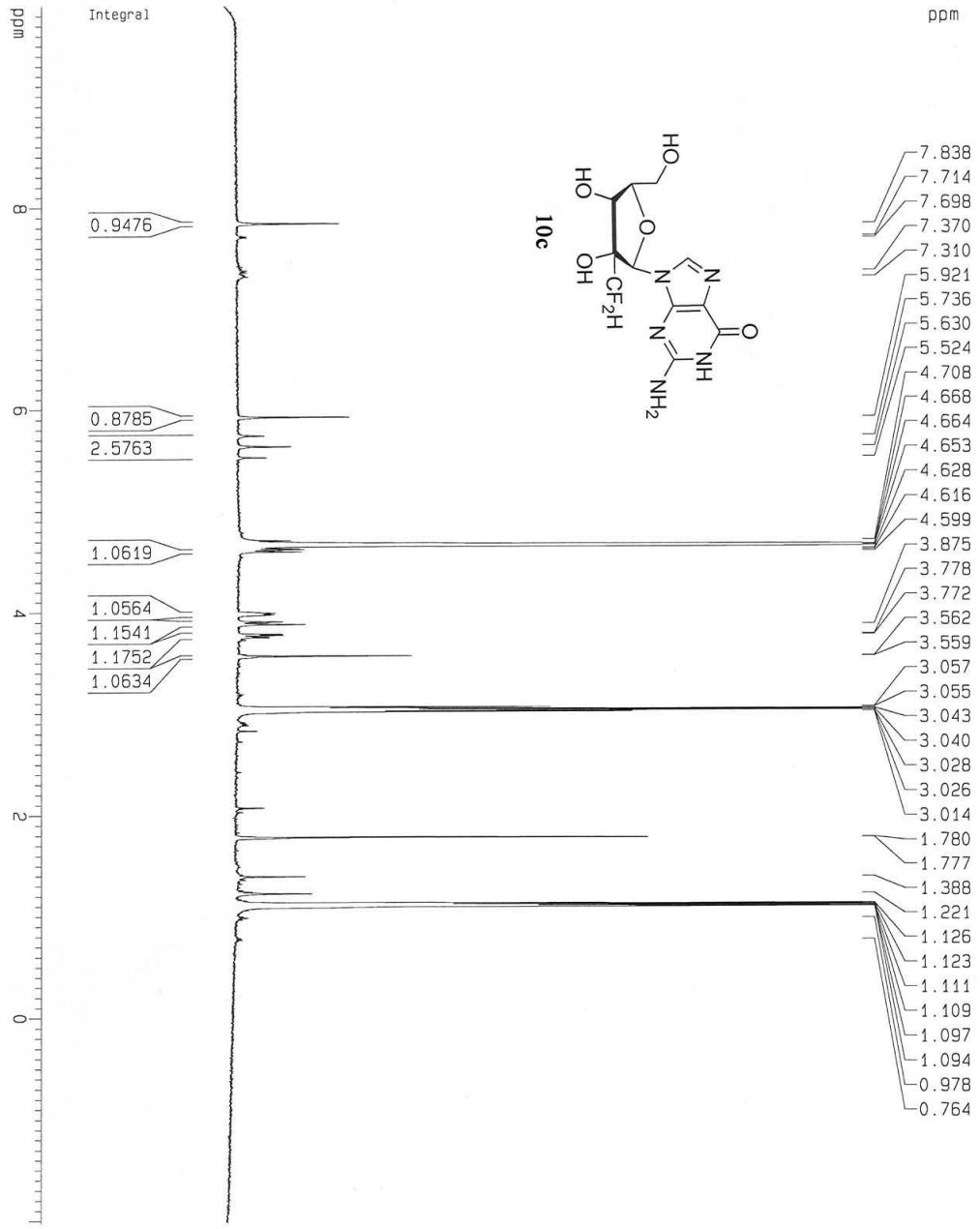
10a



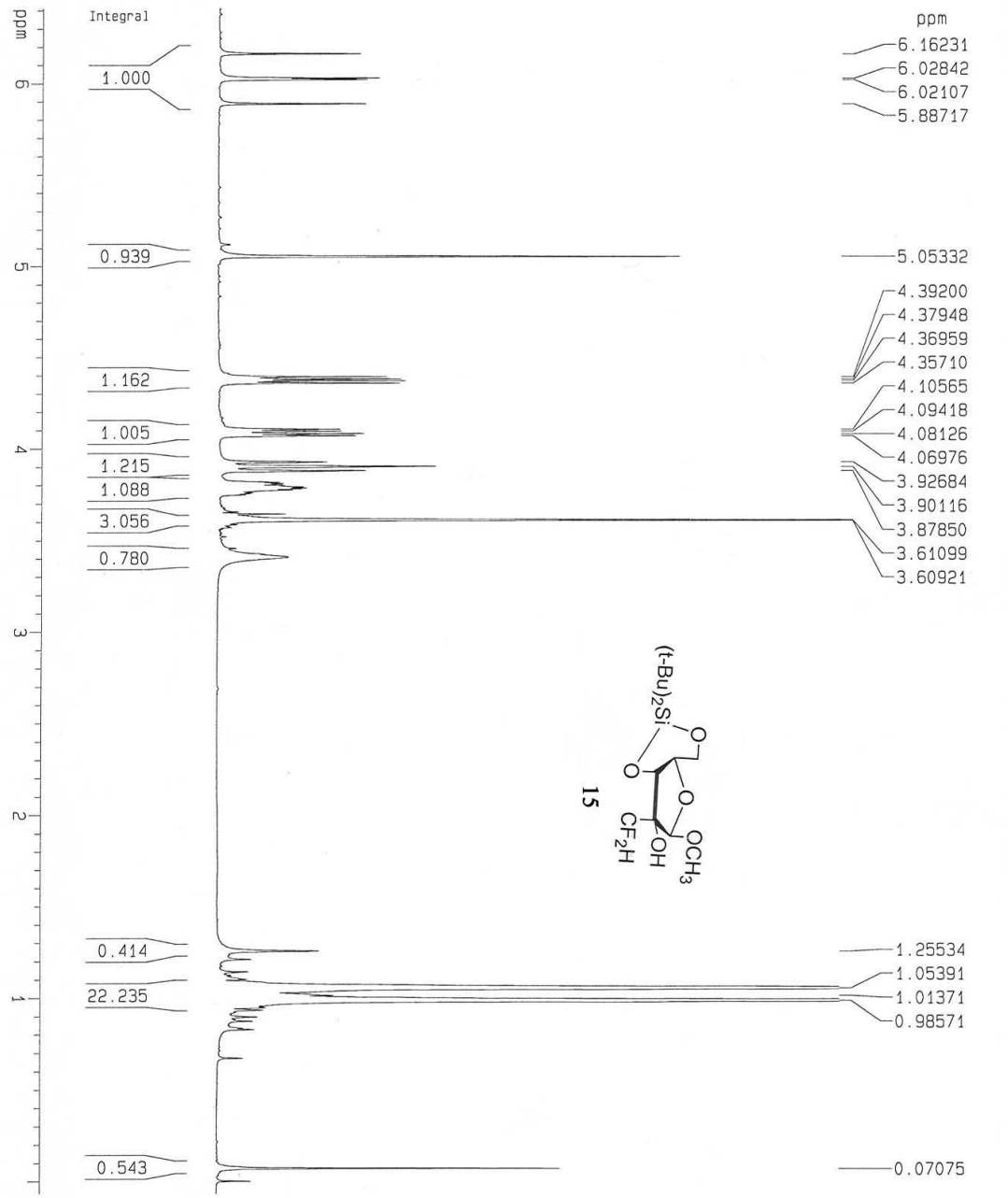

```
Current Data Parameters
NAME                  c30C
EXPNO                    1
PROCNO                   1

F2 - Acquisition Parameters
Date_              20030314
Time                 18.10
INSTRUM              spect
PROBHD     5 mm QNP  1H
PULPROG              zgdc
TD                 162598
SOLVENT              CDCl3
NS                     242
DS                       0
SWH              25062.656 Hz
FIDRES           0.154139 Hz
AQ               3.2438800 sec
RG                  3649.1
DW                  19.950 usec
DE                   7.50 usec
TE                  300.0 K
D1               4.00000000 sec
d11              0.03000000 sec

============ CHANNEL f1 ==========
NUC1                   13C
P1                   11.00 usec
PL1                  -2.00 dB
SFO1           100.6227903 MHz

============ CHANNEL f2 ==========
CPDPRG2            waltz16
NUC2                   1H
PCPD2               101.00 usec
PL2                 120.00 dB
PL12                 19.00 dB
SFO2           400.1324710 MHz

F2 - Processing parameters
SI                    16384
SF             100.6126333 MHz
WDW                      EM
SSB                      0
LB                    1.00 Hz
GB                       0
PC                    1.00

1D NMR plot parameters
CX                   20.00 cm
F1P                 172.433 ppm
F1                17348.90 Hz
F2P                  10.517 ppm
F2                 1058.18 Hz
PPMCM              8.09577 ppm/cm
HZCM             814.53619 Hz/cm
```



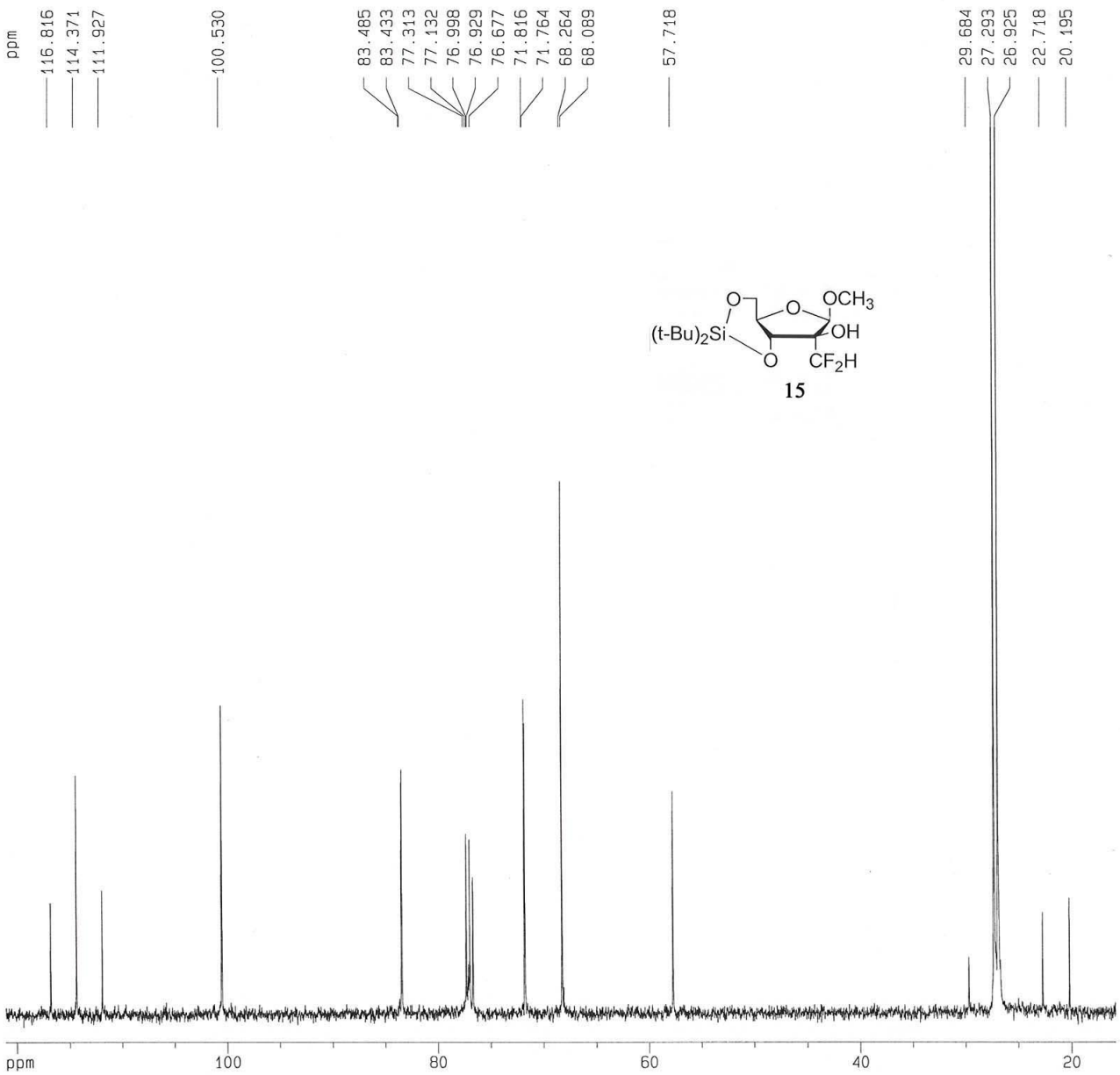

```
Current Data Parameters
NAME                  D31
EXPNO                   1
PROCNO                  1

F2 - Acquisition Parameters
Date_              20030922
Time                  17.39
INSTRUM                spect
PROBHD     5 mm QNP 1H
PULPROG                zg
TD                    16384
SOLVENT               MeOH
NS                        8
DS                        0
SWH               5580.357 Hz
FIDRES               0.340598 Hz
AQ                1.4680564 sec
RG                      161.3
DW                   89.600 usec
DE                    6.00 usec
TE                   300.0 K
D1                2.00000000 sec

======== CHANNEL f1 ========
NUC1                      1H
P1                     10.50 usec
PL1                     0.00 dB
SFO1           400.1318006 MHz

F2 - Processing parameters
SI                        32768
SF             400.1300089 MHz
WDW                       EM
SSB                        0
LB                      0.30 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                     20.00 cm
F1P                     8.834 ppm
F1                   3534.81 Hz
F2P                    -0.325 ppm
F2                   -130.00 Hz
PPMCM                 0.45795 ppm/cm
HZCM               183.24020 Hz/cm
```




```
Current Data Parameters
NAME                  B148_2
EXPNO                      1
PROCNO                     1

F2 - Acquisition Parameters
Date_              20021216
Time                  12.22
INSTRUM                spect
PROBHD     5 mm QNP  1H
PULPROG                zg
TD                     32768
SOLVENT                CDCl3
NS                        64
DS                         0
SWH                6009.615 Hz
FIDRES             0.183399 Hz
AQ                 2.7263477 sec
RG                       256
DW                  83.200 usec
DE                   4.50 usec
TE                   300.0 K
D1                 2.00000000 sec

========== CHANNEL f1 ========
NUC1                      1H
P1                    10.00 usec
PL1                    0.00 dB
SFO1           500.1320118 MHz

F2 - Processing parameters
SI                        16384
SF             500.1300181 MHz
WDW                       EM
SSB                        0
LB                    0.30 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                    20.00 cm
F1P                    9.994 ppm
F1                   4998.53 Hz
F2P                   -2.022 ppm
F2                   -1011.08 Hz
PPMCM                 0.60081 ppm/cm
HZCM                300.48077 Hz/cm
```




```
Current Data Parameters
NAME                  XM81
EXPNO                    1
PROCNO                   1

F2 - Acquisition Parameters
Date_              20021107
Time                 16.59
INSTRUM              spect
PROBHD     5 mm QNP 1H
PULPROG                 zg
TD                   16384
SOLVENT              CDCl3
NS                       8
DS                       0
SWH                4807.692 Hz
FIDRES             0.293438 Hz
AQ                 1.7039860 sec
RG                      12.7
DW                 104.000 usec
DE                   6.00 usec
TE                   300.0 K
D1                 2.00000000 sec

======== CHANNEL f1 ========
NUC1                     1H
P1                   10.50 usec
PL1                   0.00 dB
SFO1           400.1320007 MHz

F2 - Processing parameters
SI                      32768
SF             400.1300050 MHz
WDW                        EM
SSB                         0
LB                   0.30 Hz
GB                          0
PC                       1.00

1D NMR plot parameters
CX                   20.00 cm
F1P                   6.411 ppm
F1                 2565.28 Hz
F2P                  -0.067 ppm
F2                   -26.69 Hz
PPMCM              0.32389 ppm/cm
HZCM               129.59866 Hz/cm
```



15

Current Data Parameters

NAME	XM81_C
EXPNO	1
PROCNO	1

F2 - Acquisition Parameters

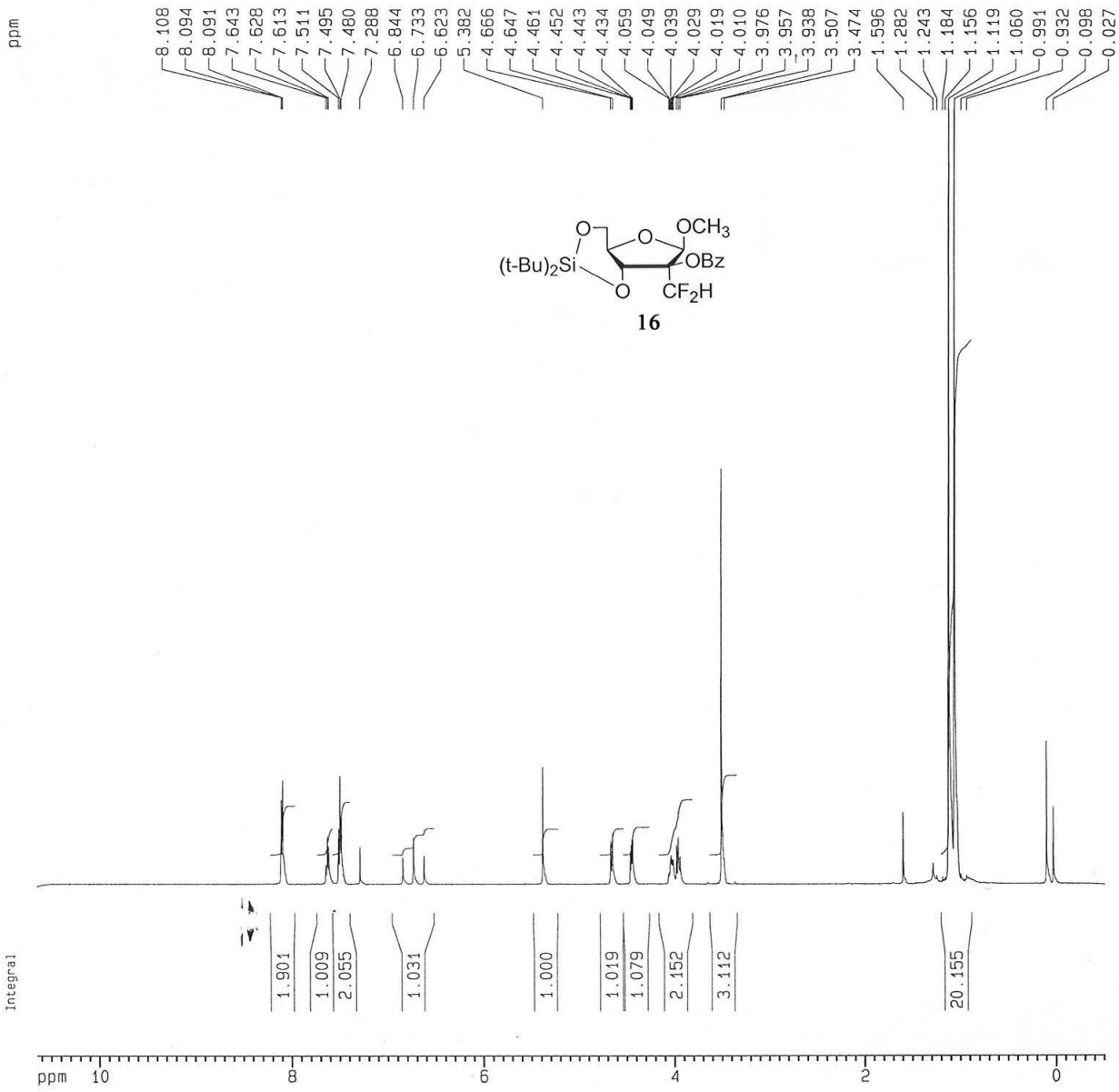
Date_	20021108
Time	19.32
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	zgdc
TD	32768
SOLVENT	CDCl3
NS	234
DS	0
SWH	30303.031 Hz
FIDRES	0.924775 Hz
AQ	0.5407220 sec
RG	574.7
DW	16.500 usec
DE	7.50 usec
TE	300.0 K
d11	0.03000000 sec
PL12	20.00 dB
CPDPRG2	waltz16
PCPD2	105.00 usec
SFO2	400.1329209 MHz
NUC2	1H
PL2	120.00 dB
D1	1.50000000 sec
P1	6.80 usec
SFO1	100.6223610 MHz
NUC1	13C
PL1	-6.00 dB

F2 - Processing parameters

SI	16384
SF	100.6127717 MHz
WDW	EM
SSB	0
LB	1.00 Hz
GB	0
PC	1.00

1D NMR plot parameters

CX	20.00 cm
F1P	121.081 ppm
F1	12182.28 Hz
F2P	15.821 ppm
F2	1591.84 Hz
PPMCM	5.26297 ppm/cm
HZCM	529.52197 Hz/cm



16

Current Data Parameters

NAME	1124a
EXPNO	1
PROCNO	1

F2 - Acquisition Parameters

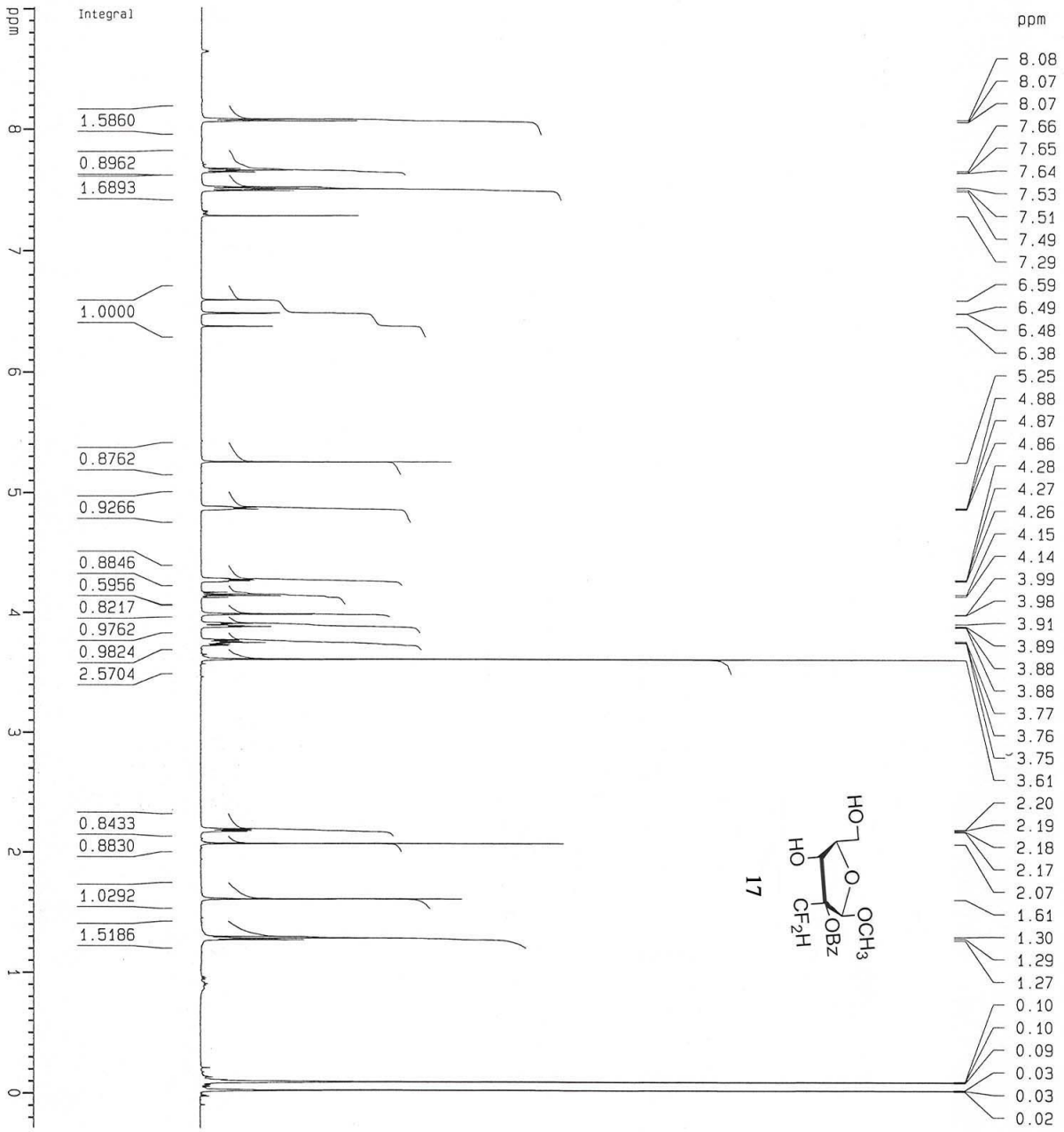
Date_	971124
Time	12.59
INSTRUM	spect
PROBHD	5 mm QNP 1H
PULPROG	zg
TD	32768
SOLVENT	CDCl3
NS	8
DS	0
SWH	5580.357 Hz
FIDRES	0.170299 Hz
AQ	2.9360628 sec
RG	64
DW	89.600 usec
DE	4.50 usec
TE	300.0 K
D1	1.00000000 sec
P1	8.00 usec
DE	4.50 usec
SFO1	500.1325364 MHz
NUC1	1H
PL1	0.00 dB

F2 - Processing parameters

SI	16384
SF	500.1300000 MHz
WDW	EM
SSB	0
LB	0.30 Hz
GB	0
PC	1.00

1D NMR plot parameters

CX	20.00 cm
F1P	11.000 ppm
F1	5501.43 Hz
F2P	-1.000 ppm
F2	-500.13 Hz
PPMCM	0.60000 ppm/cm
HZCM	300.07800 Hz/cm

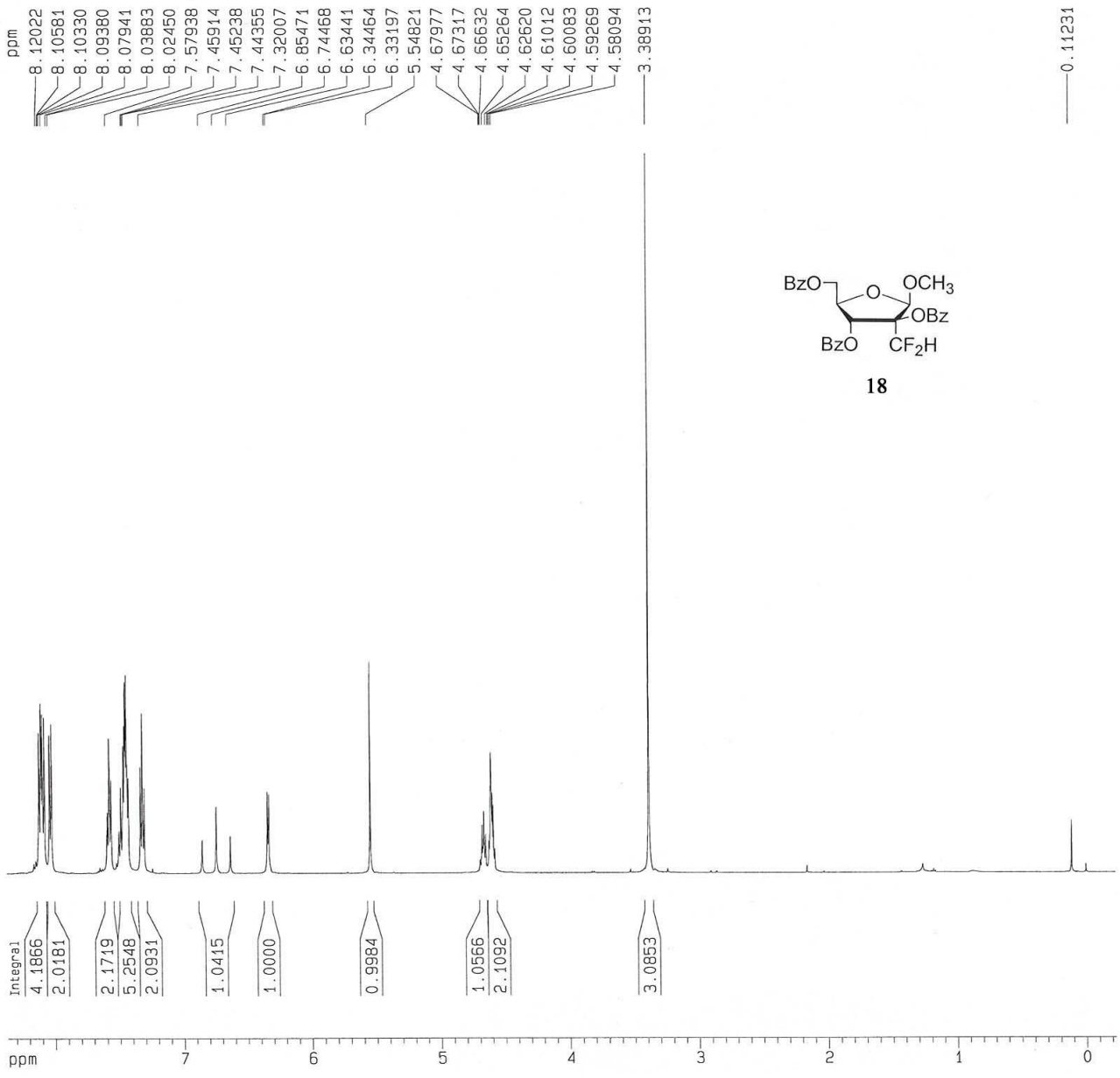


Current Data Parameters
NAME 062998a
EXPNO 1
PROCNO 1

F2 - Acquisition Parameters
Date_ 980629
Time 19.21
INSTRUM spect
PROBHD 5 mm QNP 1H
PULPROG zg
TD 32768
SOLVENT CDCl3
NS 8
DS 0
SWH 5580.357 Hz
FIDRES 0.170299 Hz
AQ 2.9360628 sec
RG 128
DW 89.600 usec
DE 4.50 usec
TE 300.0 K
D1 1.00000000 sec
P1 8.00 usec
DE 4.50 usec
SFO1 500.1325364 MHz
NUC1 1H
PL1 0.00 dB

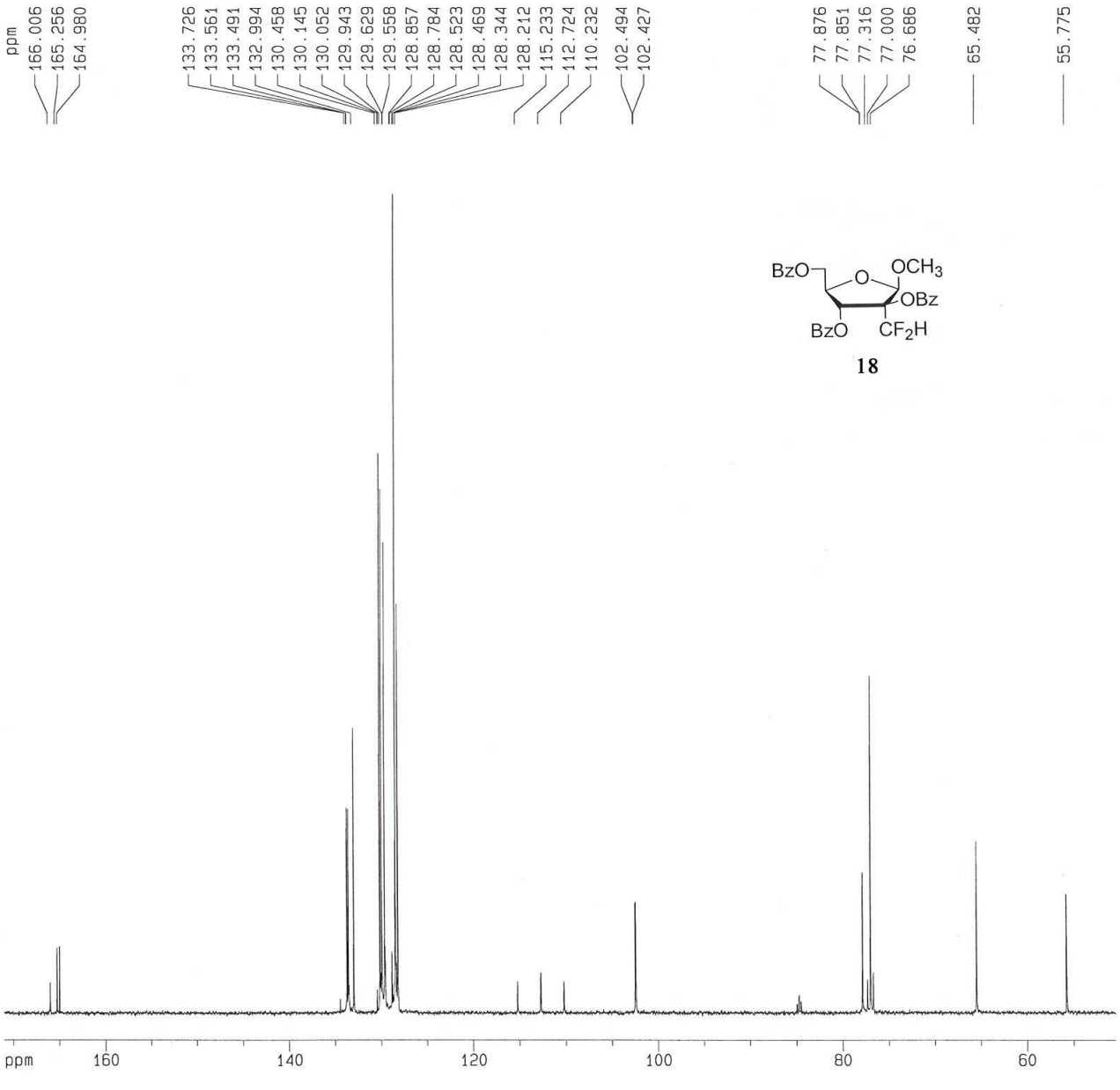
F2 - Processing parameters
SI 16384
SF 500.1300000 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

1D NMR plot parameters
CX 20.00 cm
F1P 9.012 ppm
F1 4507.38 Hz
F2P -0.286 ppm
F2 -142.92 Hz
PPMCM 0.46491 ppm/cm
HZCM 232.51491 Hz/cm



18

```
Current Data Parameters
NAME                  XM84
EXPNO                    1
PROCNO                   1

F2 - Acquisition Parameters
Date_              20021108
Time                 19.19
INSTRUM              spect
PROBHD     5 mm Multinu
PULPROG                zg
TD                  32768
SOLVENT             CDCl3
NS                       8
DS                       0
SWH               6009.615 Hz
FIDRES            0.183399 Hz
AQ                2.7263477 sec
RG                       2
DW                  83.200 usec
DE                   4.50 usec
TE                  300.0 K
D1                3.00000000 sec

============ CHANNEL f1 ========
NUC1                    1H
P1                   5.60 usec
PL1                  0.00 dB
SFO1          500.1322000 MHz

F2 - Processing parameters
SI                   16384
SF            500.1300234 MHz
WDW                     EM
SSB                      0
LB                   0.30 Hz
GB                       0
PC                    1.00

1D NMR plot parameters
CX                   20.00 cm
F1P                  8.371 ppm
F1                 4186.42 Hz
F2P                 -0.224 ppm
F2                 -111.85 Hz
PPMCM              0.42972 ppm/cm
HZCM             214.91341 Hz/cm
```



Current Data Parameters

NAME	XM84_C
EXPNO	1
PROCNO	1

F2 - Acquisition Parameters

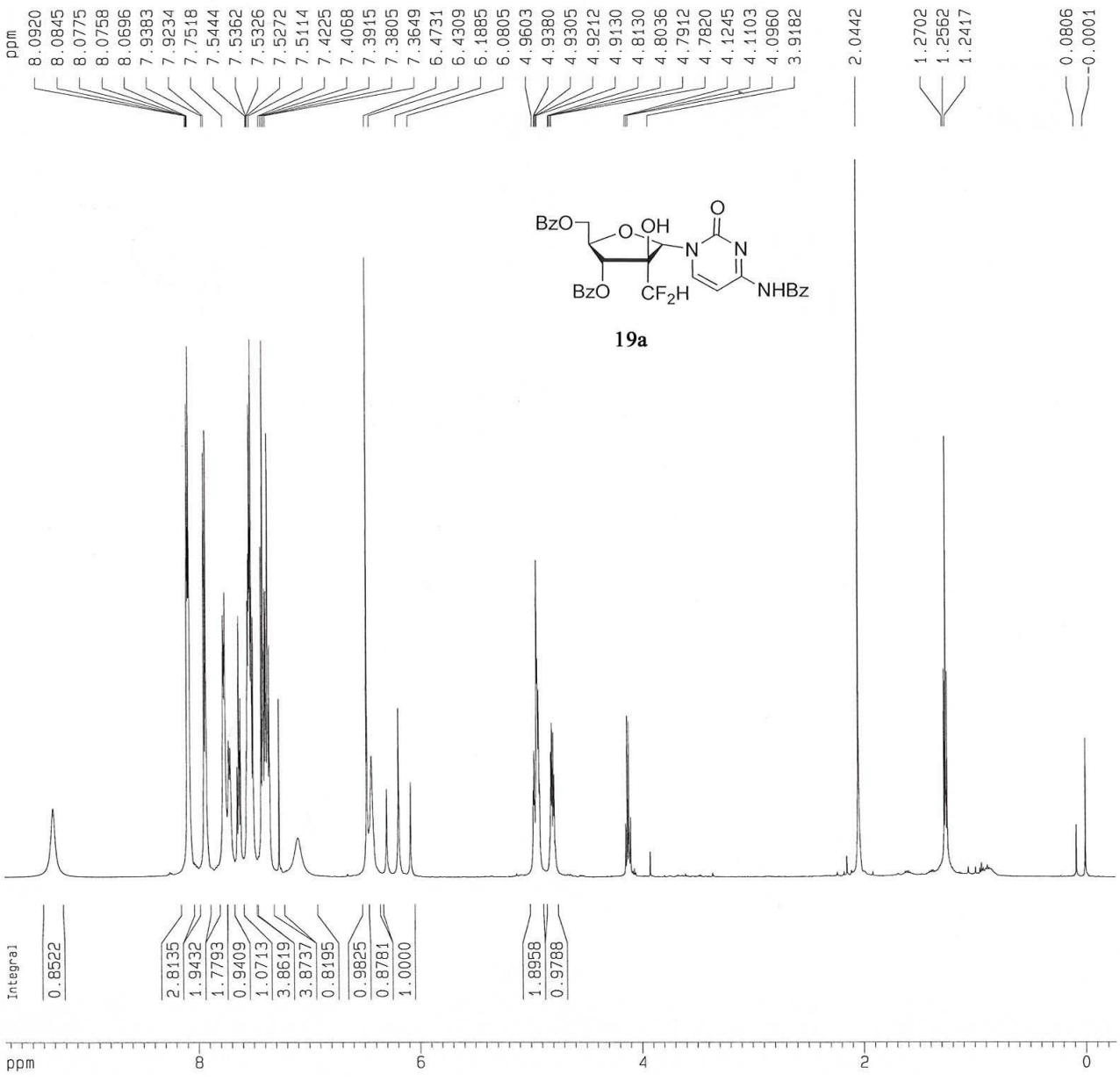
Date_	20021108
Time	19.41
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	zgdc
TD	32768
SOLVENT	CDCl3
NS	135
DS	0
SWH	30303.031 Hz
FIDRES	0.924775 Hz
AQ	0.5407220 sec
RG	574.7
DW	16.500 usec
DE	7.50 usec
TE	300.0 K
d11	0.03000000 sec
PL12	20.00 dB
CPDPRG2	waltz16
PCPD2	105.00 usec
SFO2	400.1329209 MHz
NUC2	1H
PL2	120.00 dB
D1	1.50000000 sec
P1	6.80 usec
SFO1	100.6223610 MHz
NUC1	13C
PL1	-6.00 dB

F2 - Processing parameters

SI	16384
SF	100.6127828 MHz
WDW	EM
SSB	0
LB	1.00 Hz
GB	0
PC	1.00

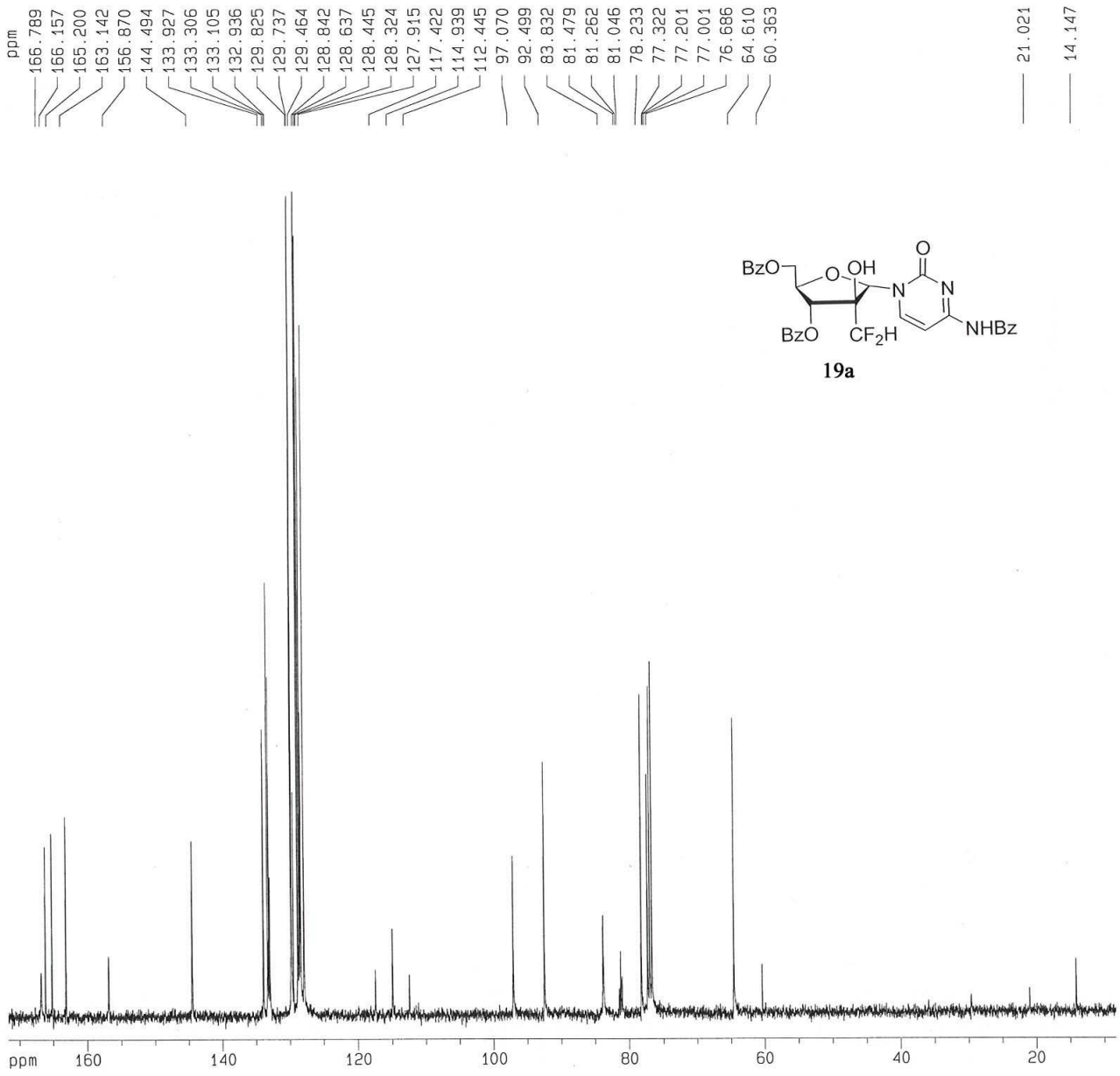
1D NMR plot parameters

CX	20.00 cm
F1P	170.961 ppm
F1	17200.86 Hz
F2P	50.487 ppm
F2	5079.65 Hz
PPMCM	6.02369 ppm/cm
HZCM	606.06061 Hz/cm



19a

```
Current Data Parameters
NAME                  XM85a1_3
EXPNO                        1
PROCNO                       1

F2 - Acquisition Parameters
Date_                 20021108
Time                     18.25
INSTRUM                  spect
PROBHD       5 mm Multinu
PULPROG                    zg
TD                      32768
SOLVENT                 CDCl3
NS                           8
DS                           0
SWH                 6009.615 Hz
FIDRES              0.183399 Hz
AQ                  2.7263477 sec
RG                          32
DW                     83.200 usec
DE                      4.50 usec
TE                      300.0 K
D1                  3.00000000 sec

============ CHANNEL f1 =========
NUC1                        1H
P1                       5.60 usec
PL1                      0.00 dB
SFO1             500.1322000 MHz

F2 - Processing parameters
SI                       16384
SF                500.1300124 MHz
WDW                         EM
SSB                          0
LB                       0.30 Hz
GB                           0
PC                       1.00

1D NMR plot parameters
CX                      20.00 cm
F1P                      9.745 ppm
F1                     4874.00 Hz
F2P                     -0.268 ppm
F2                     -134.01 Hz
PPMCM                 0.50067 ppm/cm
HZCM                250.40063 Hz/cm
```



Current Data Parameters

NAME	XM85a_C
EXPNO	1
PROCNO	1

F2 - Acquisition Parameters

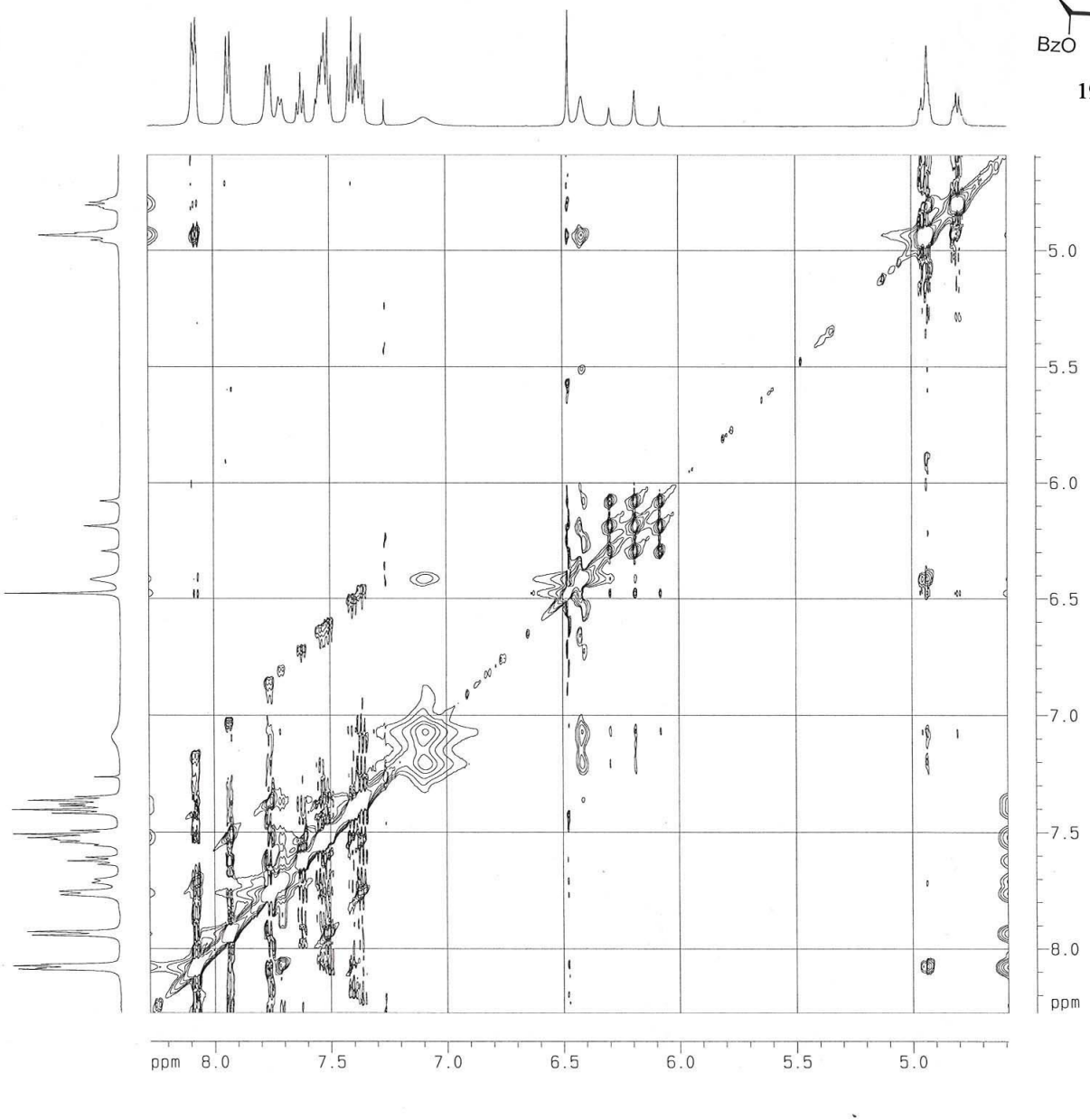
Date_	20021108
Time	18.37
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	zgdc
TD	32768
SOLVENT	CDCl3
NS	1154
DS	0
SWH	30303.031 Hz
FIDRES	0.924775 Hz
AQ	0.5407220 sec
RG	574.7
DW	16.500 usec
DE	7.50 usec
TE	300.0 K
d11	0.03000000 sec
PL12	20.00 dB
CPDPRG2	waltz16
PCPD2	105.00 usec
SFO2	400.1329209 MHz
NUC2	1H
PL2	120.00 dB
D1	1.50000000 sec
P1	6.80 usec
SFO1	100.6223610 MHz
NUC1	13C
PL1	-6.00 dB

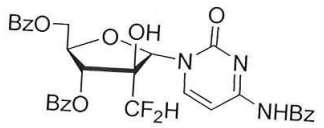
F2 - Processing parameters

SI	16384
SF	100.6127754 MHz
WDW	EM
SSB	0
LB	1.00 Hz
GB	0
PC	1.00

1D NMR plot parameters

CX	20.00 cm
F1P	171.655 ppm
F1	17270.74 Hz
F2P	8.333 ppm
F2	838.37 Hz
PPMCM	8.16614 ppm/cm
HZCM	821.61835 Hz/cm





19a

Current Data Parameters

NAME	XM85a_2d
EXPNO	3
PROCNO	1

F2 - Acquisition Parameters

Date_	20021113
Time	10.12
INSTRUM	spect
PROBHD	5 mm Multinu
PULPROG	noesytp
TD	2048
SOLVENT	CDCl3
NS	16
DS	16
SWH	1843.658 Hz
FIDRES	0.900224 Hz
AQ	0.5554676 sec
RG	20
DW	271.200 usec
DE	4.50 usec
TE	300.0 K
d0	0.00000300 sec
D1	2.00000000 sec
DB	0.75000000 sec
IN0	0.00027120 sec

CHANNEL f1

NUC1	1H
P1	6.00 usec
PL1	0.00 dB
SFO1	500.1332296 MHz

F1 - Acquisition parameters

ND0	2
TD	256
SFO1	500.1332 MHz
FIDRES	7.201788 Hz
SW	3.686 ppm

F2 - Processing parameters

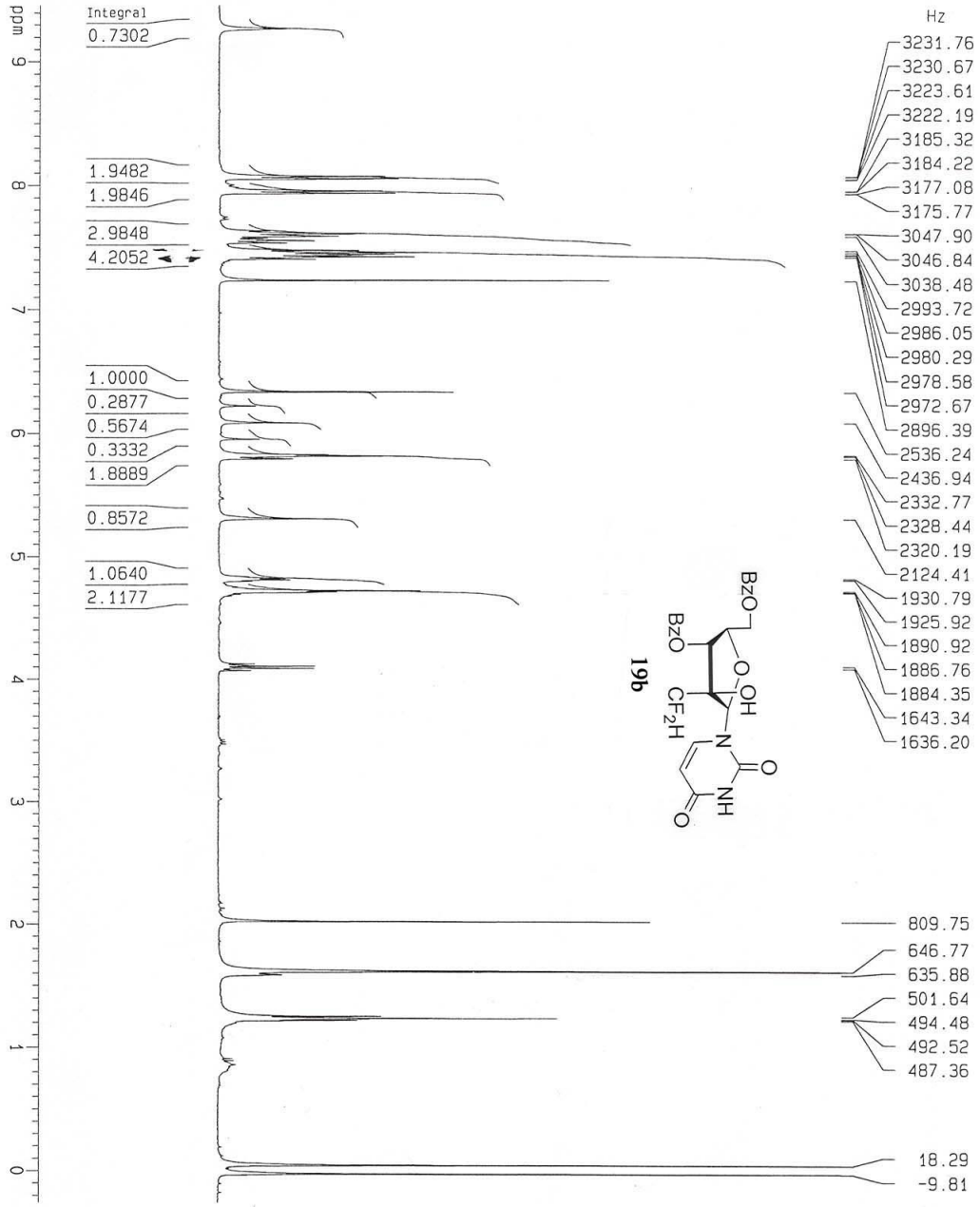
SI	2048
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0
PC	1.00

F1 - Processing parameters

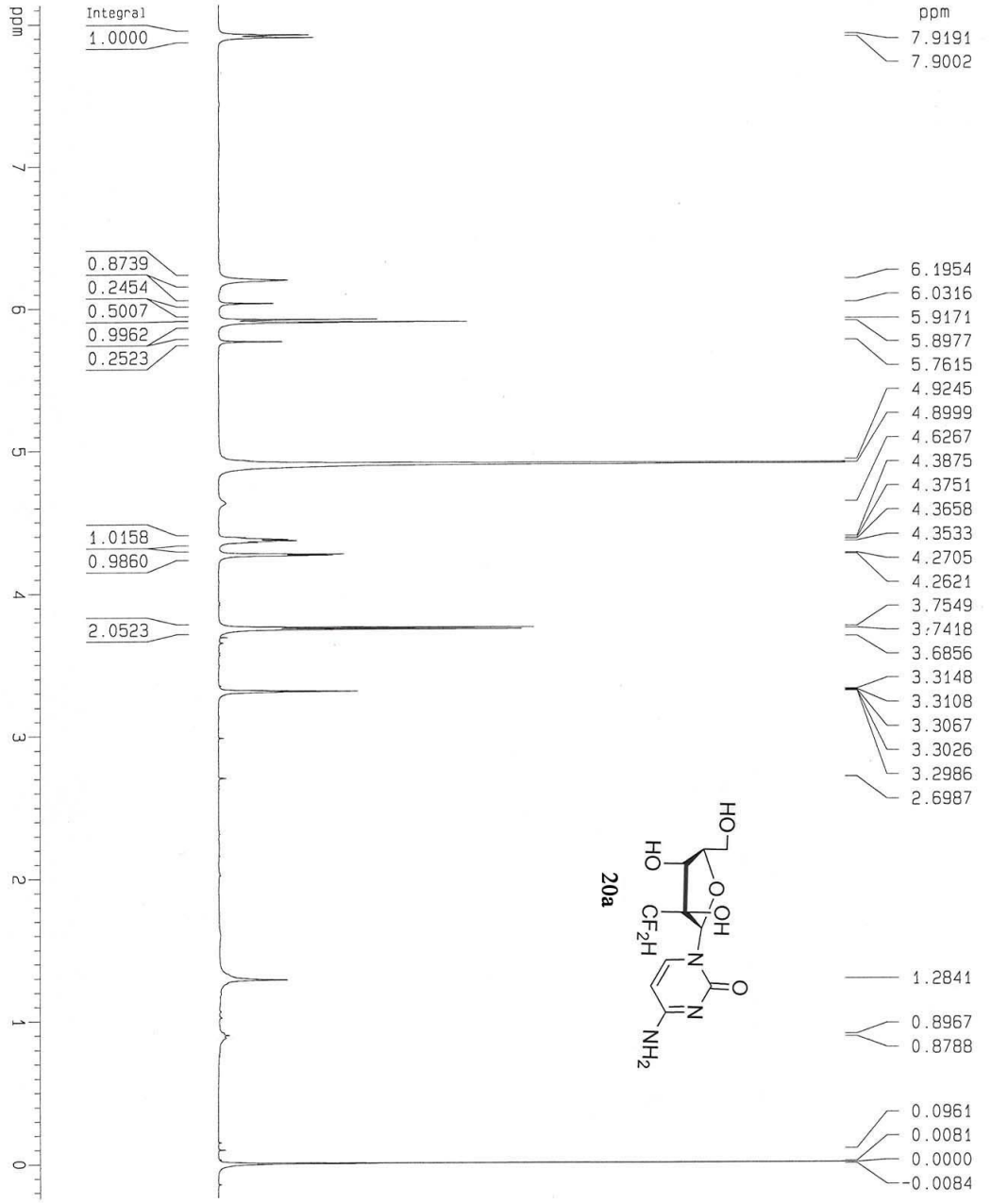
SI	1024
MC2	TPPI
SF	500.1300114 MHz
WDW	QSINE
SSB	2
LB	0.00 Hz
GB	0

2D NMR plot parameters

CX2	15.00 cm
CX1	15.00 cm
F2PLO	8.278 ppm
F2LO	4139.99 Hz
F2PHI	4.591 ppm
F2HI	2296.33 Hz
F1PLO	8.278 ppm
F1LO	4139.99 Hz
F1PHI	4.591 ppm
F1HI	2296.33 Hz
F2PPMCM	0.24576 ppm/cm
F2HZCM	122.91051 Hz/cm
F1PPMCM	0.24576 ppm/cm
F1HZCM	122.91051 Hz/cm




```
Current Data Parameters
NAME                  081898b
EXPNO                       1
PROCNO                      1

F2 - Acquisition Parameters
Date_               980818
Time                     16.46
INSTRUM                  spect
PROBHD     5 mm QNP 1H
PULPROG                  zg
TD                       32768
SOLVENT                  CDCl3
NS                       16
DS                        0
SWH                 4194.631 Hz
FIDRES              0.128010 Hz
AQ                  3.9059956 sec
RG                       228.1
DW                  119.200 usec
DE                   4.50 usec
TE                  300.0 K
D1                  1.00000000 sec
P1                       8.50 usec
SFO1          400.1319246 MHz
NUC1                     1H
PL1                 0.00 dB

F2 - Processing parameters
SI                       16384
SF            400.1300177 MHz
WDW                      EM
SSB                       0
LB                  0.30 Hz
GB                        0
PC                  1.00

1D NMR plot parameters
CX                  20.00 cm
F1P                      9.463 ppm
F1                  3786.58 Hz
F2P                      -0.256 ppm
F2                  -102.48 Hz
PPMCM               0.46597 ppm/cm
HZCM                194.45308 Hz/cm
```

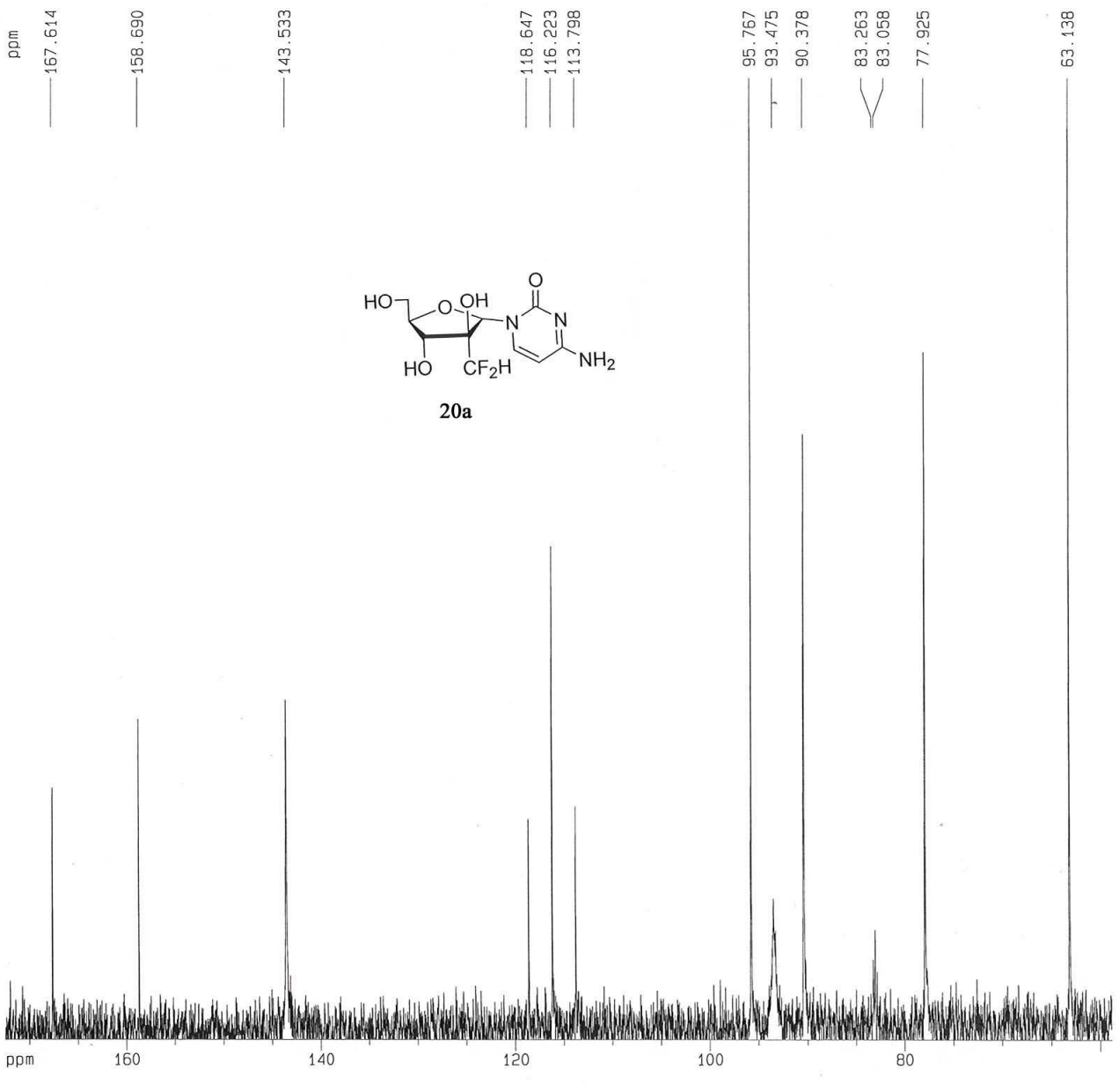


Current Data Parameters
NAME XM86a
EXPNO 1
PROCNO 1

F2 - Acquisition Parameters
Date_ 20021108
Time 19.50
INSTRUM spect
PROBHD 5 mm Multinu
PULPROG zg
TD 39046
SOLVENT C6D6
NS 8
DS 0
SWH 4807.692 Hz
FIDRES 0.123129 Hz
AQ 4.0608339 sec
RG 35.9
DW 104.000 usec
DE 7.00 usec
TE 300.0 K
D1 2.00000000 sec
P1 7.70 usec
SFO1 400.1317512 MHz
NUC1 1H
PL1 -6.00 dB

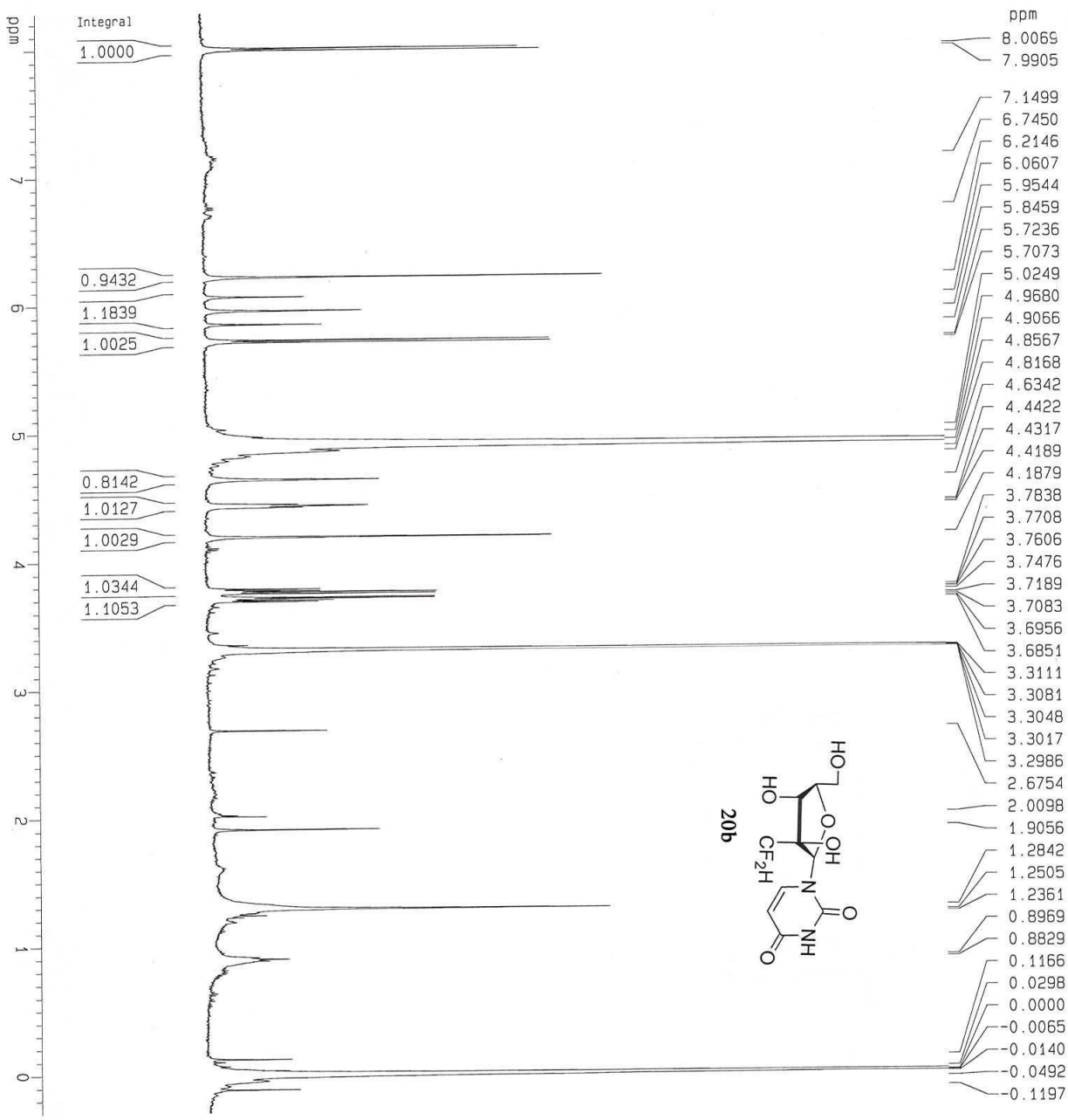
F2 - Processing parameters
SI 32768
SF 400.1300085 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

1D NMR plot parameters
CX 20.00 cm
F1P 8.133 ppm
F1 3254.43 Hz
F2P -0.240 ppm
F2 -96.09 Hz
PPMCM 0.41868 ppm/cm
HZCM 167.52579 Hz/cm

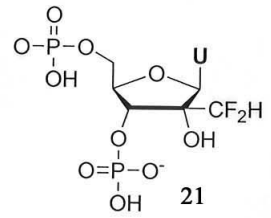


20a

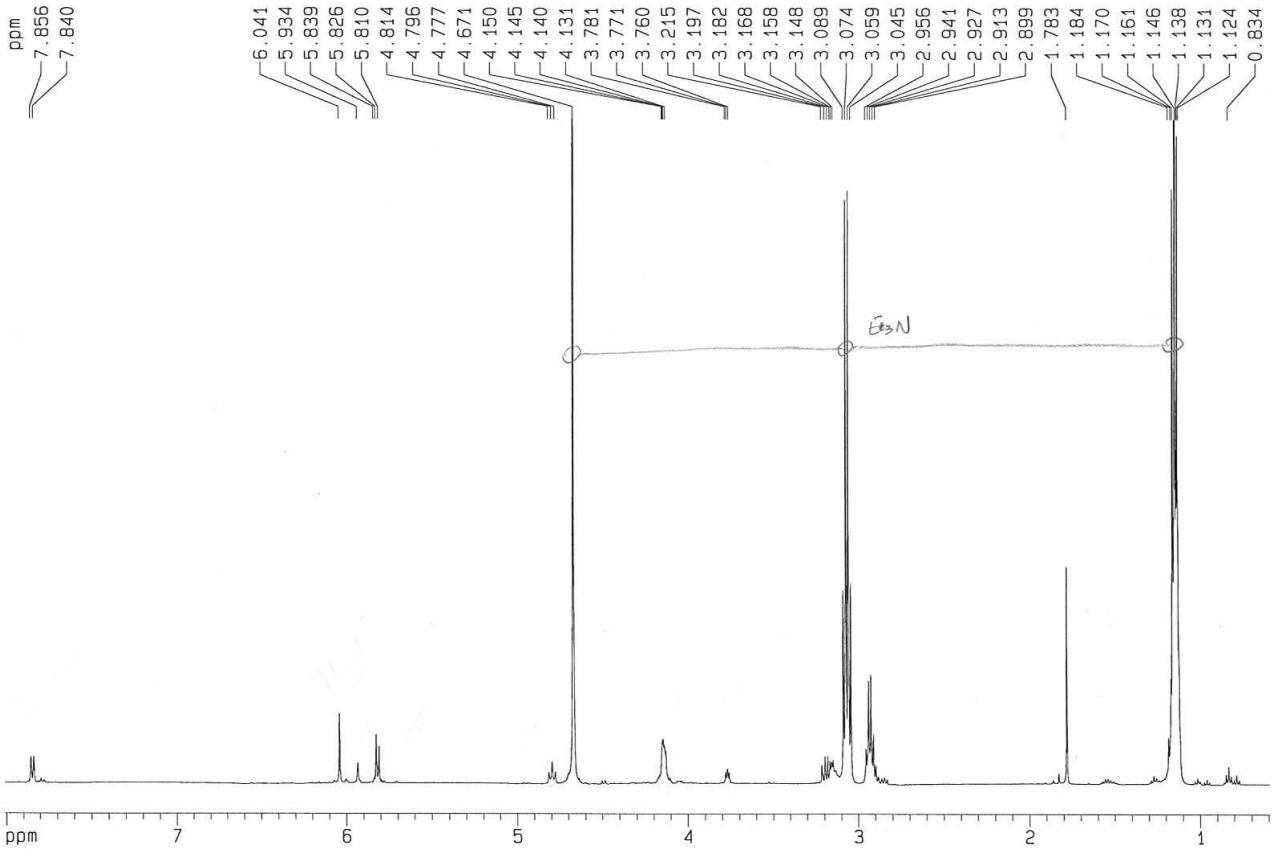
```
Current Data Parameters
NAME                 XM86a_C
EXPNO                      1
PROCNO                     1

F2 - Acquisition Parameters
Date_               20021108
Time                   19.59
INSTRUM                spect
PROBHD      5 mm Multinu
PULPROG                zgdc
TD                    32768
SOLVENT                C6D6
NS                     1155
DS                        0
SWH              30303.031 Hz
FIDRES            0.924775 Hz
AQ                0.5407220 sec
RG                    574.7
DW                   16.500 usec
DE                    7.50 usec
TE                   300.0 K
d11              0.03000000 sec
PL12                  20.00 dB
CPDPRG2              waltz16
PCPD2                105.00 usec
SFO2             400.1329209 MHz
NUC2                      1H
PL2                   120.00 dB
D1               1.50000000 sec
P1                    6.80 usec
SFO1             100.6223610 MHz
NUC1                      13C
PL1                   -6.00 dB

F2 - Processing parameters
SI                     16384
SF               100.6126296 MHz
WDW                       EM
SSB                        0
LB                      1.00 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                     20.00 cm
F1P                   172.484 ppm
F1                  17354.07 Hz
F2P                    58.841 ppm
F2                   5920.14 Hz
PPMCM                 5.68215 ppm/cm
HZCM                571.69635 Hz/cm
```



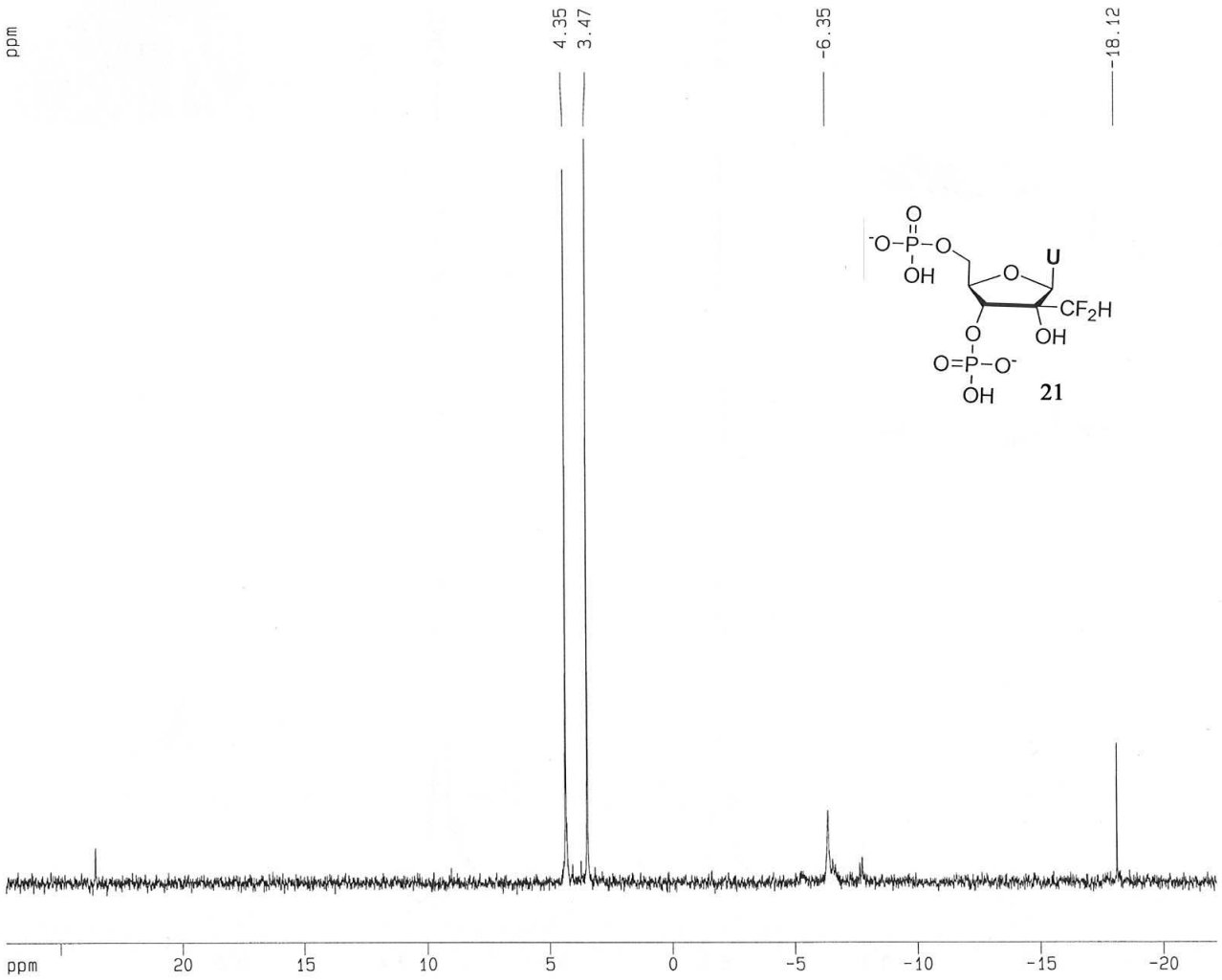

```
Current Data Parameters
NAME                  XM66b
EXPNO                     1
PROCNO                    1

F2 - Acquisition Parameters
Date_              20021108
Time                 20.02
INSTRUM                spect
PROBHD     5 mm Multinu
PULPROG                 zg
TD                   32768
SOLVENT               MeOH
NS                       8
DS                       0
SWH               6009.615 Hz
FIDRES             0.183399 Hz
AQ                2.7263477 sec
RG                      128
DW                 83.200 usec
DE                  4.50 usec
TE                  300.0 K
D1                3.00000000 sec

========== CHANNEL f1 ========
NUC1                    1H
P1                    5.60 usec
PL1                   0.00 dB
SFO1          500.1322000 MHz

F2 - Processing parameters
SI                     16384
SF          500.1300139 MHz
WDW                       EM
SSB                        0
LB                   0.30 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                   20.00 cm
F1P                   8.284 ppm
F1                 4142.89 Hz
F2P                  -0.297 ppm
F2                 -148.75 Hz
PPMCM              0.42905 ppm/cm
HZCM              214.58174 Hz/cm
```



21

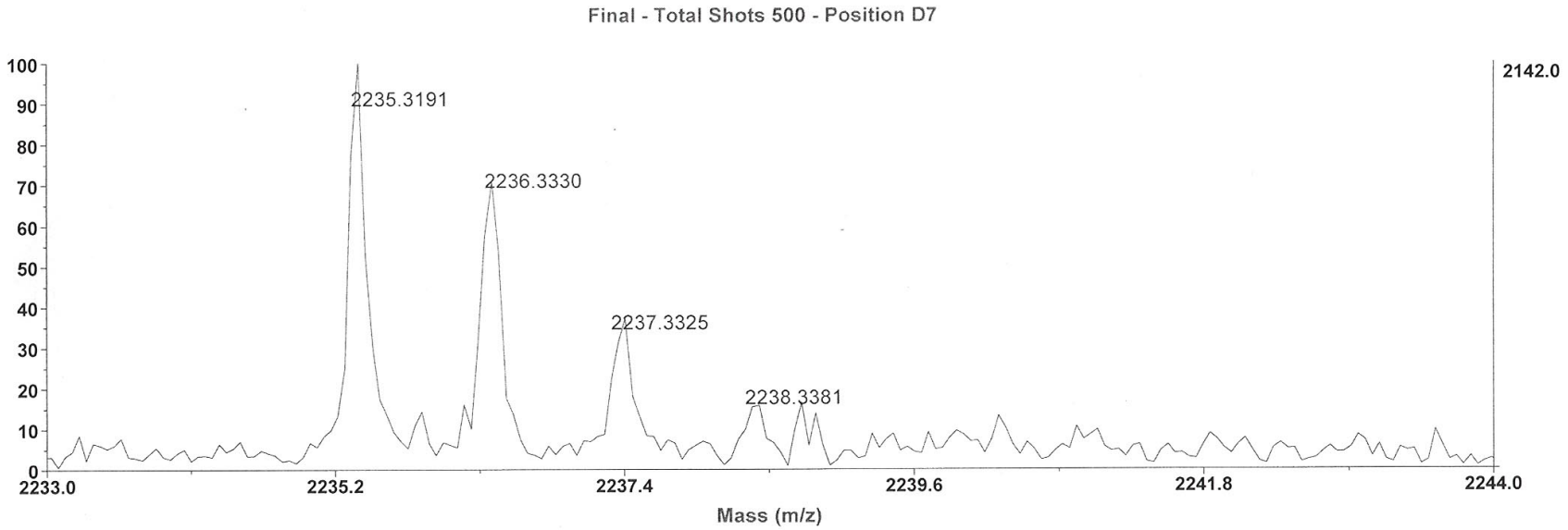



```
Current Data Parameters
NAME                  A129_H
EXPNO                      1
PROCNO                     1

F2 - Acquisition Parameters
Date_              20020516
Time                  15.32
INSTRUM               spect
PROBHD     5 mm QNP  1H
PULPROG                  zg
TD                    32768
SOLVENT               CDCl3
NS                         8
DS                         0
SWH                5208.333 Hz
FIDRES            0.158946 Hz
AQ                3.1457779 sec
RG                       256
DW                  96.000 usec
DE                   4.50 usec
TE                   300.0 K
D1                2.00000000 sec

============ CHANNEL f1 =========
NUC1                      1H
P1                    10.00 usec
PL1                    0.00 dB
SFO1           500.1320118 MHz

F2 - Processing parameters
SI                        16384
SF             500.1300181 MHz
WDW                       EM
SSB                        0
LB                     0.30 Hz
GB                         0
PC                      1.00

1D NMR plot parameters
CX                    20.00 cm
F1P                   8.009 ppm
F1                  4005.78 Hz
F2P                   0.596 ppm
F2                   297.86 Hz
PPMCM              0.37070 ppm/cm
HZCM             185.39598 Hz/cm
```




```
Current Data Parameters
NAME                  A129_P_2
EXPNO                        1
PROCNO                       1

F2 - Acquisition Parameters
Date_                 20020517
Time                     11.43
INSTRUM                  spect
PROBHD       5 mm QNP  1H
PULPROG                  zgdc
TD                      65536
SOLVENT                   D2O
NS                         16
DS                          1
SWH                60790.273 Hz
FIDRES             0.927586 Hz
AQ                 0.5390836 sec
RG                      16384
DW                      8.225 usec
DE                     11.75 usec
TE                     300.0 K
D1                1.50000000 sec
d11               0.03000000 sec

============ CHANNEL f1 ==========
NUC1                       31P
P1                        5.00 usec
PL1                       0.00 dB
SFO1              202.4560850 MHz

============ CHANNEL f2 ==========
CPDPRG2                waltz16
NUC2                        1H
PCPD2                   100.00 usec
PL2                     120.00 dB
PL12                     23.00 dB
SFO2              500.1320005 MHz

F2 - Processing parameters
SI                        32768
SF                202.4556064 MHz
WDW                         EM
SSB                          0
LB                        1.00 Hz
GB                           0
PC                        1.40

1D NMR plot parameters
CX                       20.00 cm
F1P                      27.221 ppm
F1                     5510.94 Hz
F2P                     -22.161 ppm
F2                    -4486.57 Hz
PPMCM                  2.46906 ppm/cm
HZCM                 499.87589 Hz/cm
```

Spectrum Report



Summary

Serial Number	347007026
Instrument Name	AB 347007026

Spot

Plate Name	00330005386
Plate Barcode	00330005386
Spot Set Name	**MSMS I - 5386 - 50607**
Job Run Comments	50613 - Jingdong Ye - 7mer - with and without amm dihydrogen phos 76.8 F
Spot Position	**D7**
Spot Name	50613 - Jingdong Ye 7mer
Spot Type	Unknown
Location X Min	16629.000
Location X Max	17364.000
Location Y Min	29056.000
Location Y Max	29621.000
Spot Run #	**4**

Spectrum

Rejected Sub-spectra	0
Calibration Type	None
Rejected Shots	0
Update Default Calibration	Disabled
Stop Reason	Specified signal to noise ratio met
Total Accumulations	4
Total Ion Count	9.863878e+006
Total Shots	500
Skip Job on Plate Calibration Failure	Disabled
Job-Wide Interpretation	Disabled
Configured for LCMS Experiments	Disabled

Acquisition

Method Name	\Refl (-) 500 5K 2kfm v3000s
Mass Analysis Type	MS Reflector Negative Ion Mode
CID	Off
CID Gas Type	Atmosphere
CID Gas Pressure	Medium
Metastable Suppressor	Off
Start Time	06/13/05 15:11:37
Laser Intensity Range	2950 To 2950

Processing

Processing Method Name	\Refl Neg SN=5
Calibration Type	Default
Peak Count	116
Tallest Peak Height	1.034536e+004
Tallest Peak S/N	279.4777
Tallest Peak Mass	857.0168
Tallest Peak Resolution	8069.7490
Calibration Coefficients	
	1.250903e+002
	3.187997e-007
	2.273230e-006

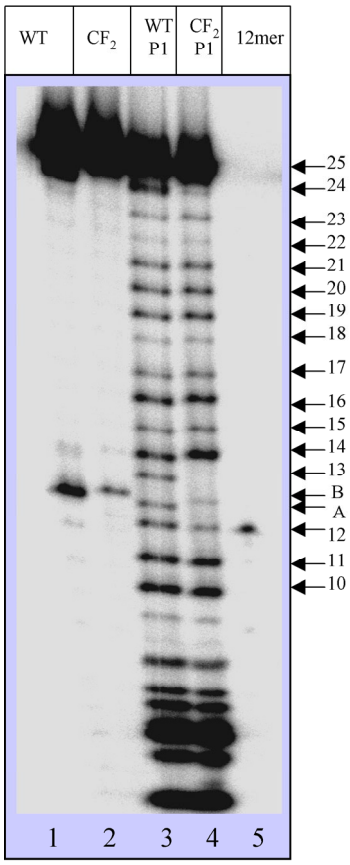
Interpretation

Interpretation Method Name	<<none>>

Pressures

Source 2	1.8e-008
TC Turbo	1.9e-002
Reflector	2.8e-008
Source 1	3.5e-008

P1 nuclease digestion. *p$(dN)_{12}rU_{2'CF2H}(dN)_{12}$ (or *p$(dN)_{12}rU(dN)_{12}$) was incubated in P1 nuclease buffer in the presence of 0.01 mg/ml P1 nuclease (USB) for 10 min at 37 ^{o}C. The mixture was then heated at 90 ^{o}C for 1 minute, combined with gel loading buffer (final concentration: 50 mM Tris-HCl, pH 8, 3.5 M urea and 0.0025% (w/v) each of bromophenol blue and xylene cyanol) and separated by 20% sequencing PAGE. Figure S1 shows the digestion pattern. The shorter P1 digestion products of the CF_2 oligonucleotide, arising from cleavage upstream of the modification (12 nucleotides and less), comigrate with their wild-type counterparts during PAGE. This is expected, as these products lack the modification and therefore are identical to those produced from wild-type oligonuleotide. The 12mer was assigned using the oligonucleotide from which the modified CF_2 oligonucleotide was generated (Figure S1, lane 5). We observe no product corresponding to P1 cleavage at the site bearing the difluoromethyl group (13mer), suggesting that the modification hinders P1 activity. The longer P1 digestion products of the CF_2 oligonucleotide, arising from cleavage downstream of the modification (14 nucleotides and greater), migrate modestly slower than their wild-type counterparts, presumably due to the presence of the difluoromethyl modification. The species labeled as A and B in lanes 3 and 4 respectively, are present in the undigested controls (lanes 1 and 2, respectively) and most likely correspond to cleavage products arising from internal transphosphorylation at the ribose linkage.



1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25

*pdCTGTCACCGAAAU$_x$ACACGCAAGATG

U_x = U or $U_{2'CF_2H}$

Figure S1. Nuclease P1 digestion of chimeric oligonucleotides containing uridine or 2′-*C*-β-difluoromethyluridine. Lane 1, WT, *pd(CTGTCACCGAAA)rUd(ACACGCAAGATG), input; Lane 2, CF_2, *pd(CTGTCACCGAAA)r$U_{2'CF_2H}$d(ACACGCAAGATG), input; Lane 3, WT, with P1 nuclease; Lane 4, CF_2, with P1 nuclease; Lane 5, 12mer, *pd(CTGTCACCGAAA).