ASL – Perfect spectra from poor data

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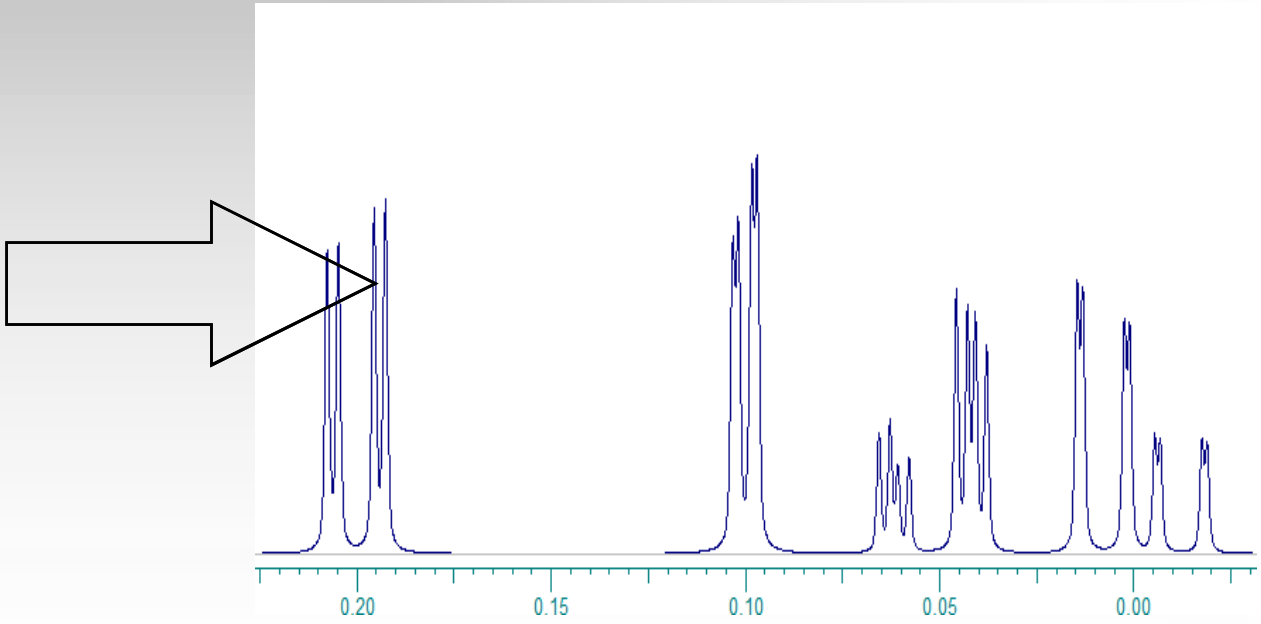
ASL

ν_A, ν_B, ν_C, ν_X

J_{AB}, J_{AC}, J_{AX}, J_{BC},

J_{BX}, J_{CX}



ASL (Adaptive Spectral Libraries)

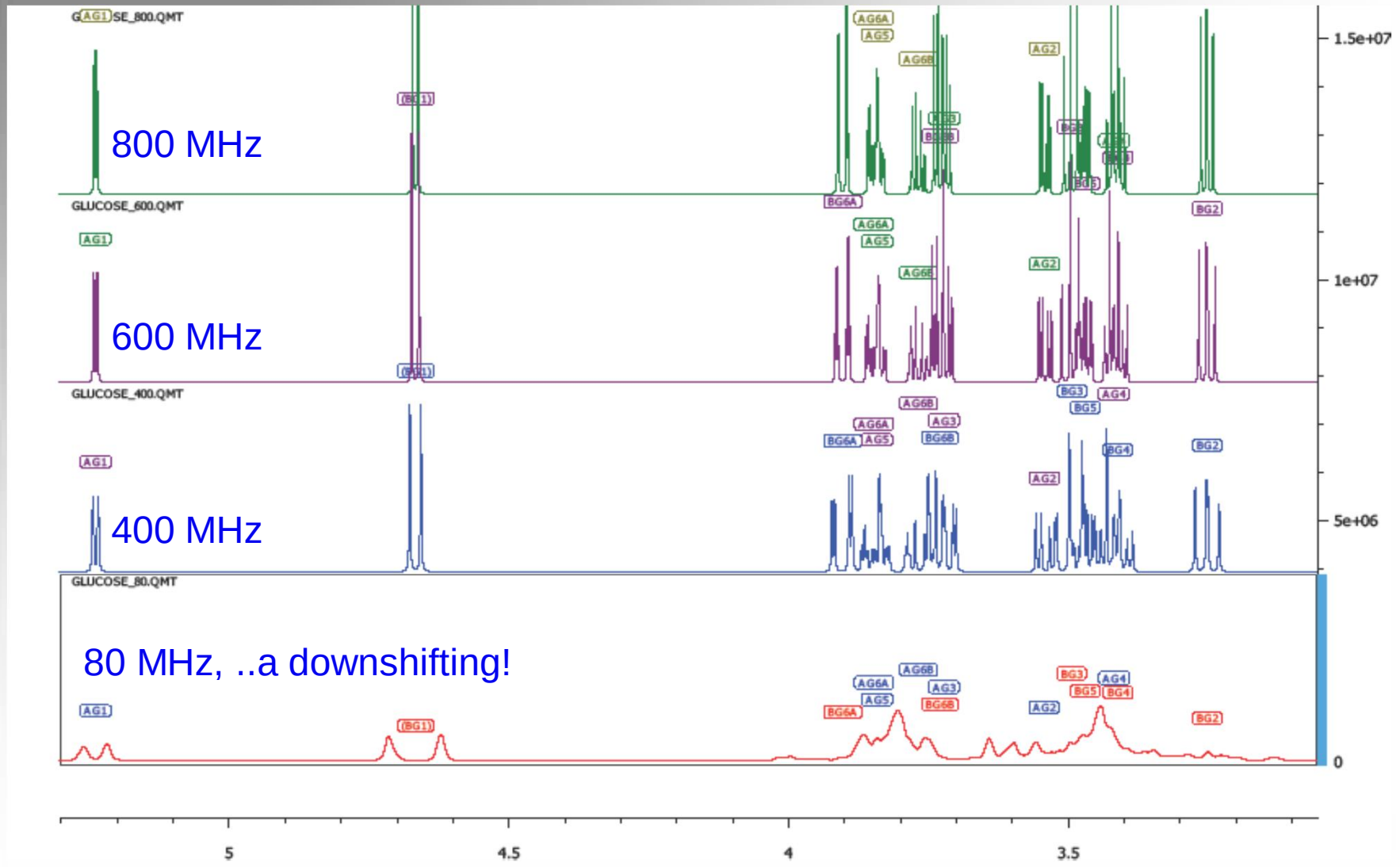
ASL spectra:

Perfect spectra, one spectrum – any field and line-shape – with no artefacts, from poor data with 90% data compression !

- *The essential information about a spin-system can be packed into one txt-file !*
- *The information can be used to simulate the spectrum with any shifts and line-shape, at any field - in a few seconds !*

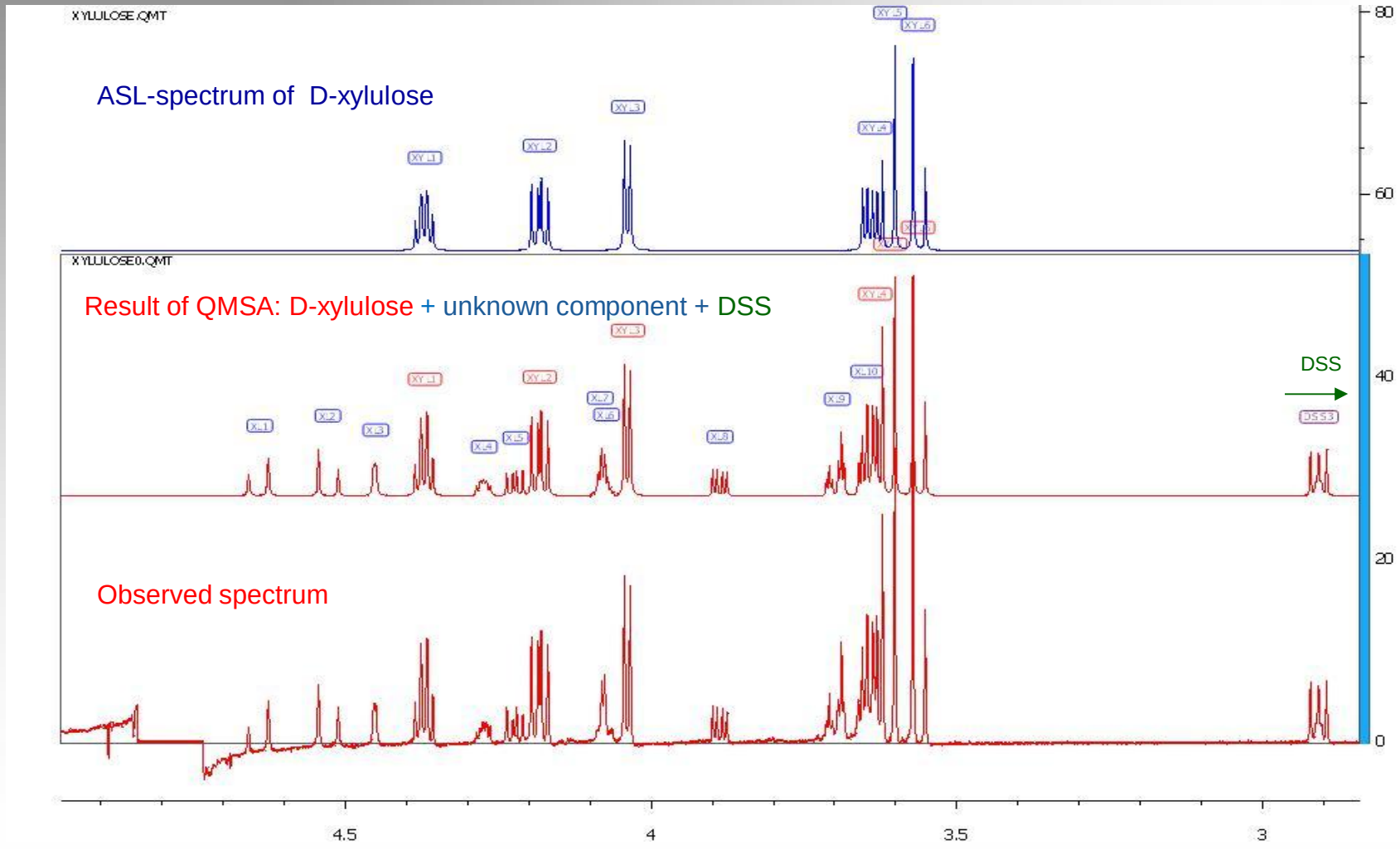
Adaptive Spectrum:

Glucose, simulated on the basis of the 500 MHz spectrum analysis.
Line-width was 1.0 Hz in all the spectra.



From poor data to perfect spectra

Spectrum of D-xylulose from HMDB (HMDB01644): from original spectrum to ASL-spectrum:



ASL & PMR FORMAT (PMR for general ChemAdder file) file contains the essential information about the spectrum, to be used to simulate the spectrum at any field, with any line-width and line-shape or to build up models (recipes, see below) for biofluids

ASL-file for Leucine, with less essential information removed:

```
&CASEINFO: %P1=human, %P2=male, %P3=50, %P4=
&DEFAULTS:
        SPECTRUM = C:\CHEMADDER\URINE\LEUCINE.QMT
       REFERENCE = TSP        ; TMS, TSP, DSS, REF
         SOLVENT = D2O        ; CDCL3, DMSO, ACD6, CD3CN, CD3OD, D2O, CD2Cl2, ..ND
  REFERENCE MMOL = 9.292      ; QUANTITATIVE REFERENCE (ND=1.0)
        GAUSSIAN = 0.000      ; GAUSSIAN % IN LINE-SHAPE (CAN BE >100%)

&CHEMICAL SHIFTS(PPM):
LEUCINE  2*SPIN= 1 SPECIES=1H POPULATION(Y)=  1.000000[OBS=  1.000000] MWGT=  131.180 SLOPE=  1.0000
 LEU1    3.7381  1*1*1  STAT=Y  PRED= 3.738 RANGE(0)= 0.028 WIDTH(Y)= 1.000 RESP(N)= 1.0000 TYPE= 100q
 LEU5    1.7482  1*1*1  STAT=Y  PRED= 1.748 RANGE(0)= 0.018 WIDTH(Y)= 1.000 RESP(N)= 1.0000 TYPE= 100o
 LEU6    1.6913  1*1*1  STAT=Y  PRED=
 LEU7    1.7177  1*1*1  STAT=Y  PRED=
 LEU8    0.9596  1*1*3  STAT=Y  PRED=
 LEU9    0.9718  1*1*3  STAT=Y  PRED=
&COUPLING CONSTANTS:
 LEU15   5.330  J LEU1  LEU5  STAT=N  PRED=  5.33 RANGE= 0.14
 LEU16   8.770  J LEU1  LEU6  STAT=N  PRED=  8.77 RANGE= 0.18
 LEU56 -14.410  J LEU5  LEU6  STAT=N  PRED=-14.41 RANGE= 0.24
 LEU57   8.870  J LEU5  LEU7  STAT=N
 LEU67   6.150  J LEU6  LEU7  STAT=N
 LEU78   6.530  J LEU7  LEU8  STAT=N
 LEU79   6.510  J LEU7  LEU9  STAT=N  PRED=  6.51 RANGE= 0.16

&BARTLETTS  38    15.010  5703.827     (= N & BROADENING & 1ST)
 460 7 461 25 462 35 463 23 464 6 618 1 619 6 620 16 621 43 623 50 624 49 625 43 626 32 627 17 628 ..

&ASL TEMPLATES AT:  600.402822 MHZ
      1  LEUCINE
      1   2253.033447      0.765327    1    1     1.000
      2   2252.666992      1.065659    1    1     1.000
      3   2251.659180      4.946416    1    1     1.000
      ...
&END of FILE
```

&CASEINFO gives external information about the sample, to be used at the last phase of the analysis to classify the samples using non-linear methods …under work, not relevant with leucine.

&DEFAULTS defines the defaults, like the reference concentration.

MWGT = Molecular weight

STAT=Y/N shift optimizable/fixed, PRED= default, RANGE(i)=range(symmetry), WIDTH(Y/N)= linewidth (optimizable/fixed), RESP(Y/N)=Response factor (optimizable/fixed), TYPE= 1H type (for HOLISTICS). If shifts (the 2nd column) and PRED are same for shifts, the shifts are kept equal.

STAT=Y/N if coupling is optimizable/fixed (N is default in metabolomic analyses). If couplings have the same name (the 1st column) they are kept equal.

&BARTLETT integrals and &ASL TEMPLATES (a packed peak-list) give the spectrum in an ultrapacked form, for fast metabolite search & profiling.

TARGETED RECIPE (PMR file in include format) FOR URINE

The metabolites are given in order of typical abundances and finding probabilities (here from Bouatra, S. et al. *The human urine metabolome*. PLoS ONE 8, e73076, 2013):

```
&CASEINFO: %P1=human, %P2=male, %age=50, &type=5, &class=12
&DEFAULTS:                                          (Less essential information removed)
        ORIGINAL = C:\URINE\1_URINE220218.JDX
       BATCHFILE = C:\CHEMADDER\SCRIPTS\URINE.BAT                  = The MENU (SCRIPT file) for analysis
         PROFILE = C:\CHEMADDER\PROFILE\SEARCHPROFILE.TXT          = Defaults for iteration
  REFERENCE MMOL = 9.292

&INC C:\CHEMADDER\URINE\HIPPURATE.ASL                POPULATION=  0.82  ***  = 100% probability
&INC C:\CHEMADDER\URINE\CITRATE.ASL                  POPULATION=  0.72  ***
&INC C:\CHEMADDER\URINE\GLYCINE.ASL                  POPULATION=  0.38  ***
&INC C:\CHEMADDER\URINE\ME3-AMINE-OXIDE.ASL          POPULATION=  0.32  ***
&INC C:\CHEMADDER\URINE\TAURINE.ASL                  POPULATION=  0.29  ***
&INC C:\CHEMADDER\URINE\CYSTEINE.ASL                 POPULATION=  0.23  ***
&INC C:\CHEMADDER\URINE\CREATINE.ASL                 POPULATION=  0.16  ***
&INC C:\CHEMADDER\URINE\HISTIDINE.ASL                POPULATION=  0.15  ***
&INC C:\CHEMADDER\URINE\GLYCOLATE.ASL                POPULATION=  0.15  ***
&INC C:\CHEMADDER\URINE\ISOCITRATE.ASL               POPULATION=  0.17       < 50% probability
&INC C:\CHEMADDER\URINE\GLUCOSE.ASL                  POPULATION=  0.13  ***
&INC C:\CHEMADDER\URINE\GLUTAMINE.ASL                POPULATION=  0.13  ***
&INC C:\CHEMADDER\URINE\ETHANOLAMINE.ASL             POPULATION=  0.13  ***
&INC C:\CHEMADDER\URINE\ARABINITOL.ASL               POPULATION=  0.11  **
&INC C:\CHEMADDER\URINE\MANNITOL.ASL                 POPULATION=  0.11
&INC C:\CHEMADDER\URINE\TRIGONELLINE.ASL             POPULATION=  0.11  ***
...
&INC C:\CHEMADDER\URINE\ALPHA-KETOISOVALERATE.ASL    POPULATION=  0.01  *
&INC C:\CHEMADDER\URINE\SYRINGATE.ASL                POPULATION=  0.01
&INC C:\CHEMADDER\URINE\N-ACETYLPUTRESCINE.ASL       POPULATION=  0.01
&INC C:\CHEMADDER\URINE\13-DINH2-PROPANE.ASL         POPULATION=  0.01
&INC C:\CHEMADDER\URINE\TRANS-FERULATE.ASL           POPULATION=  0.01
&INC C:\CHEMADDER\URINE\MALEATE.ASL                  POPULATION=  0.01

&END of FILE                                                          Totally >200 metabolites
```

ASL-file for ALANINE ^{13}C ISOTOPOMERS

```
SpinAdder2018.01 ..\NMRDATA\FLUX\ALIPHATIC\ALANINE.ASL

&DEFAULTS:
        SPECTRUM = C:\NMRDATA\FLUX\ALIPHATIC\ALANINEC.QMT ; ND => (RE)READ ORIGINAL!
         PROFILE = C:\CHEMADDER\PROFILE\HSQC_PROFILE.TXT ; OPTIONS/ADDER PROFILE
       STRUCTURE = C:\NMRDATA\FLUX\MMLIB\ALANINE.MMS ; MMS/SDF/MOL-FILE/ND
      LINE WIDTH = 4.938           ; 0.0 = USE SPECIES DEFAULT (HZ)
        GAUSSIAN = 75.000          ; GAUSSIAN % IN LINE-SHAPE (CAN BE >100%)
        QM LINES = 14              ; NO. OF QM LINES
            RRMS = 0.9549          ; 100 * RRMS
```

Shifts and isotopomers (7)

```
&CHEMICAL SHIFTS(PPM):
ALA2  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.059907[OBS=  0.059907] MWGT=   89.090 SLOPE=  1.0000 ROI= A2
 A2_0         51.556  1*1*1  STAT=Y  PRED= 51.556 RANGE(0)= 0.100 WIDTH(Y)= 7.222 RESP(N)= 1.0000 TYPE= ENOs
ALA2_M  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.035861[OBS=  0.035861] MWGT=   89.090 SLOPE=  1.0000 ROI= A2
 A2_M         51.553  1*1*1  STAT=Y  PRED= 51.553 RANGE(0)= 0.100 WIDTH(Y)= 7.487 RESP(N)= 1.0000 TYPE= ENOd
 AM          -50.000  1*1*1  STAT=N
ALA2_1  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.023742[OBS=  0.023742] MWGT=   89.090 SLOPE=  1.0000 ROI= A2
 A2_1         51.547  1*1*1  STAT=Y  PRED= 51.547 RANGE(0)= 0.100 WIDTH(Y)= 7.487 RESP(N)= 1.0000 TYPE= ENOd
 A1          150.000  1*1*1  STAT=N
ALA2_1M  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.125601[OBS=  0.125601] MWGT=   89.090 SLOPE=  1.0000 ROI= A2
 A2_1M        51.543  1*1*1  STAT=Y  PRED= 51.543 RANGE(0)= 0.100 WIDTH(Y)= 7.262 RESP(N)= 1.0000 TYPE= ENOq
 AM          -50.000  1*1*1  STAT=N
 A1          150.000  1*1*1  STAT=N

ALAM  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.253036[OBS=  0.253036] MWGT=   89.090 SLOPE=  1.0000 ROI= AM
 AM_0         55.366  1*1*1  STAT=Y  PRED= 55.366 RANGE(0)= 0.100 WIDTH(Y)= 4.824 RESP(N)= 1.0000 TYPE= ENOs
ALAM_1  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.004912[OBS=  0.004912] MWGT=   89.090 SLOPE=  1.0000 ROI= AM
 AM_1         55.357  1*1*1  STAT=Y  PRED= 55.357 RANGE(0)= 0.100 WIDTH(N)= 5.103 RESP(N)= 1.0000 TYPE= ENOd
 A1          150.000  1*1*1  STAT=N
ALAM_2  2*SPIN= 1 SPECIES=13C POPULATION(Y)=  0.496941[OBS=  0.496941] MWGT=   89.090 SLOPE=  1.0000 ROI= AM
 AM_2         55.353  1*1*1  STAT=Y  PRED= 55.353 RANGE(0)= 0.100 WIDTH(Y)= 4.940 RESP(N)= 1.0000 TYPE= ENOd
 A2          100.000  1*1*1  STAT=N
```

Couplings

```
&COUPLING CONSTANTS:
ALA2_M
 J_A2M      34.125    J A2_M     AM         STAT=N  PRED= 34.12 RANGE= 1.00
ALA2_1
 J_A12      59.390    J A2_1     A1         STAT=N  PRED= 59.39 RANGE= 1.00
ALA2_1M
 J_A2M      34.125    J A2_1M    AM         STAT=N  PRED= 34.12 RANGE= 1.00
 J_A12      59.390    J A2_1M    A1         STAT=N  PRED= 59.39 RANGE= 1.00
*
ALAM_1
 J_A1M      16.355    J AM_1     A1         STAT=N  PRED= 16.35 RANGE= 1.00
ALAM_2
 J_A2M      34.125    J AM_2     A2         STAT=N  PRED= 34.12 RANGE= 1.00
```

...Continued on next slide

Continued...

"**GLOBAL: COUPLINGS**" means that couplings with same name are kept equal

ROI = Region of Interest in 2D spectrum

```
&CONSTRAINTS
 GLOBAL: COUPLINGS
 IGNORE(PPM): 62.81548 to 56.10651
 IGNORE(PPM): 54.60394 to 52.29091
 IGNORE(PPM): 50.79745 to 25.51133
 ROI=AM    1.5920    0.1000   55.3600    1.5000  VOL= 0.237E+11  TYPE=HSQC
 ROI=A2    4.1850    0.1000   51.5500    1.5000  VOL= 0.854E+10  TYPE=HSQC
 WEIGHT= 1.000
 POPULATION  ALA2_M + ALA2_1M - ALAM_2 = 0.0000  [CALC=0.0003]
 POPULATION  ALA2_1 + ALA2 - ALAM - ALAM_1 = 0.0000  [CALC=-0.0005]
```

POPULATION: linear constraints for populations, here based on isotopomer ratio rules

```
&ASL TEMPLATES AT:  150.854006 MHZ
      1  ALA2
      1   7777.019531     13.847084    1    1     7.222
      2  ALA2_M
      2   7793.504395      6.663256    1    2     7.465
      3   7760.534180      6.692832    1    2     7.465
      3  ALA2_1
      4   7805.868164      6.705251    1    4     7.479
      5   7746.796875      6.651945    1    4     7.479
      4  ALA2_1M
      6   7822.353027      3.440226    1    6     7.235
      7   7788.009277      3.472610    1    6     7.199
      8   7763.281738      3.430198    1    6     7.198
      9   7728.938477      3.428029    1    6     7.235
      5  ALAM
     10   8352.618164     20.731327    1    9     4.824
      6  ALAM_1
     11   8359.486328      9.809249    1   10     5.073
     12   8343.001953      9.787066    1   10     5.073
      7  ALAM_2
     13   8367.729492     10.173408    1   12     4.933
     14   8333.385742     10.071101    1   12     4.933

&END of FILE
```