How to perform 2D NMR Experiments on the Varian/Agilent VNMRS 500 when using the Chempack interface

June 3, 2014

To start:

1. Insert your sample

2. If running **any 2D** spectrum or even just a 1D ^{13}**C spectrum on the 500, tune the probe !!!** (see last page).

For ALL experiments:

1. Click on **New Study** (bottom left corner of the VnmrJ screen).

2. **Enter a Sample Name.** Your data will be stored as /home/*yourloginname*/vnmrsys/data/samplename_experiment_version, e.g. /home/morin/vnmrsys/data/ethylindanone_PROTON_01.

3. **Select the solvent** from the drop down Solvent menu (or click on the supplied buttons).

4. **Select the experiments** you wish to perform. If you select any ^{1}H-^{1}H 2D experiment, a proton experiment will <u>automatically</u> be inserted first. If you are running any ^{13}C-^{1}H 2D experiment you have to add a Carbon experiment <u>yourself</u>. See pages 2-3 for instructions about the 2D experiments.)

5. **To change any parameters** from their default values double click on the time field (or "Node") in the Study queue. Change, e.g. the number of scans then click on **Save**.

6. Click on **Submit**.

The instrument will lock, gradient shim and run each of the experiments. You cannot process your 2D data so see what it looks like until the acquisition is completed.

To leave the instrument: **Click on Logout on the Start tab, Standard page**. Answer **OK** if prompted. Then select **Log Out** from the **System** menu.

Common 2D Experiments for use on the VNMRS 500

<u>**COSY**</u> Procedure

Select the **(HH)Homo2D tab** then **gCOSY**

- If the sample is dilute increase **Scans per t1 Increment** on the **Acquire tab/Defaults** page. Use a multiple of 1. Leave the **t1 increments** parameter at the default value.
- A proton experiment will automatically be submitted also.
- You can use the **Show time** button to see how long the experiment will take.

<u>**HSQC**</u> procedure (to observe correlations of carbons with their directly attached protons)

Select the **Crisis2 tab** then **gc2hsqcse** (right hand column, 2nd from the top)

- If the sample is dilute increase **Scans per t1 Increment** on the **Acquire tab/Defaults** page. Use a multiple of 4. Leave the **t1 increments** parameter at the default value.
- A proton experiment will automatically be submitted also but you will have to add the ^{13}**C experiment yourself.**
- You can use the **Show time** button to see how long the experiment will take.

Common 2D Experiments for use on the VNMRS 500 (continued)

HMBC procedure (to observe correlations of carbons with protons that are 2 or 3 bonds away allowing you to observe the nonprotonated carbons that can't be observed with an HSQC-type experiment.)

Select the **Crisis2 tab** then **gc2hmbc** (left hand column, 3rd from the top)

- If the sample is dilute increase **Scans per t1 Increment** on the **Acquire tab/Defaults** page. Use a multiple of 4. Leave the **t1 increments** parameter at the default value.
- A proton experiment will automatically be submitted also but you will have to add the ^{13}**C experiment yourself.**
- You can use the **Show time** button to see how long the experiment will take.
- Don't forget to add the ^{13}**C experiment**

The pulse sequence is optimized for $^{2,3}J_{CH}$ = 8 Hz. If there is a coupling constant that is very different from that value, you have two choices: (a) run the HMBC experiment again with a larger or smaller coupling constant (found either on the **Acquire tab/Default or Pulse Sequence pages**), or (b) try the **CIGARAD** experiment under **the Jn(CH)corr tab**. The CIGARAD experiment allows for the specification of a range of jnxh as it is called, accessible again from the **Acquire tab/Default or Pulse Sequence pages). However**, the CIGARAD experiment I significantly less sensitive than the HMBC-type experiments so it will take longer to run, and some correlations that were observed in a **gc2hmbc** experiment may not appear in the CIGARAD experiment.

There is also another useful variant of the HMBC experiment available on the Varian 500 called **gc2hmbcme**. This is a multiplicity-edited version. You would select it from the **Crisis2 tab** (right hand column at the bottom) and run it as you would the **gc2hmbc** experiment. After the acquisition is complete, on the **Process tab/Default tab** you will see three buttons: "**Full 2D FT – No Editing**" which will give you the normal HMBC data displayed, "**Full 2D FT – C & CH2**" which will only show C and CH2 carbons and "**Full 2D FT – CH & CH3**" which will only show CH and CH3 carbons. The utility of this is obvious. Again, this sequence is optimized for 8 Hz.

NOESY or **ROESY** procedure

Select the **(HH)Homo2D tab** then **NOESY** or **ROESY**

- Regulate the temperature at e.g. 24C or 25C (go to the Spin/Temp page, move the sliding bar and click on "Regulate temperature"). Wait until the temperature stabilizes.
- If the sample is dilute increase **Scans per t1 Increment** on the **Acquire tab/Defaults** page. Use a multiple of 8. Leave the **t1 increments** parameter at the default value.
- A proton experiment will automatically be submitted also.
- You can use the **Show time** button to see how long the experiment will take.
- If you fail to see correlations that you believe should be there: in the case of NOESY, increase the "NOE Mixing time" to 700 or, in the case of ROESY, increase the "Spinlock Mixing time" to 200.

Processing

You can retrieve your spectra as usual using Open …

Expand in the usual ways as much as you wish.

Under the Process tab / Plot page click on **Automatic Plot** to obtain a plot of the 2D spectrum with the 1D proton and 1D ^{13}C spectra along the sides.

The software is smart enough to use the 1D ^{1}H and 1D ^{13}C spectra that were obtained during the Study. If you want to use different 1D spectra you may select them, as indicated below (excuse the low resolution of the figure), specifying a filename or different workspaces that contains the spectra. In the latter case you can scale the 1D spectrum in the other workspaces to the desired height and then deselect the "Auto Vert Scale" box.

Tuning procedure for 500 MHz NMR probe:

Remember that when you are running just a ^{13}C 1D spectrum you must tune **both** the ^{13}C and ^{1}H channels.

1. Insert your sample.

2. Enter "**mtune**", the parameter panel becomes the tuning panel.

3. Select channel 1 (= H1).

4. Click "**Start probe tune**", wait for a few seconds and then click "**Autoscale**".

5. You want the dip positioned at the vertical blue line and as close to the baseline as possible. If it is go to step 7.

6. If adjustments need to be made:

   - to make it easier to see the blue line from the magnet, place the red cursor on top of the blue line,
   - go to the magnet, at the bottom of which there are two capacitors: one for H1 and one for C13. Turning the shaft of the capacitor shifts the dip left and right; turning the bottom knob of the capacitor moves the dip up and down as well as a bit sideways.

7. Click on "**Stop probe tune**". Wait a few seconds.

8. Select channel 2 (=C13) if you are doing any ^{13}C-^{1}H correlation experiment or running even a simple 1D ^{13}C spectrum.

9. Click "**Start probe tune**", wait for a few seconds and then click "**Autoscale**".

10. Tune the C13 channel with the same object in mind (step 5).

11. Click on "**Stop probe tune**". Wait a few seconds.

12. Click on "**Quit**" to exit the Tuning routine.