WET Sequence and Multiple Solvent Suppression

This pulse sequence uses a shaped pulse to suppress one or more solvent signals. The option of carbon decoupling is available for suppression of solvent signals with large C13 satellites. This pulse sequence requires a waveform generator (not available on Mercury 300).

Ref: Smallcomb, S.H.; Patt, S.L.; Keifer, P.A.; "WET Solvent Suppression and Its Applications to LC NMR and High-Resolution NMR Spectroscopy", **Journal of Magnetic Resonance, Series A, 117**, 295-303 (1995).

BioPack Version (water only)
1. Set up **tof, sw**, and **pw**. Then run auto-calib. by **[SetUp] [Water] [AutoCalibration] [WET].**
Respond to questions and use the calibrated value of **pw90** instead of the standard parameters.

Manual Set-Up (multiple solvents or more selective on water)

1. Run 1-D proton spectrum. Optimize the window and tof. Find the pw90.

2. Expand the region that includes the peak(s) to suppress.

3. Select **[Pbox]**. (this is in the interactive display menu, use >**ds** to access) Choices for the type of shaped pulse will be in the menu bar (90, 180 etc). Select **[90].** The program will prompt the user to place the cursors around the peak to suppress. Place the cursors around the peak (or first peak).
5. Now select the type of pulse - there are many, many types. Suggested try: **[u-burp**] or select **[Other] [Other]** (prompt for a name:) >**seduce** (uburp tends to be better for 90% water or solvent and seduce better for multiple suppression)
6. At this point, place the cursors around another peak and select the pulse type again if desired.
7. After finishing selecting peak(s), choose **[Close]** and then **[Name].** Give the shaped pulse a name (i.e. seduce1 or sample1). Then select **[Close].** Enter the correct values for pw90 and tpwr for the 90. These values will be used to calculate the pulse. If they are entered incorrectly, the pulse will be wrong.
8. At this point, the machine will calculate the shaped pulse (should appear on screen). A file will be generated in the vnmrsys/shaplib called *pulsename*.RF (for whatever pulsename name it was given). All the info about the pulse is in this file including power and pulse length. Open a terminal window and find the pw and tpwr for this pulse by looking in the vnmrsys/shapelib and in the *pulsename*.RF file. It is in the top line and in the second panel.
9. Join a second experiment and move the parameters into this new experiment (i.e. **mp(1,2)**). Start the macro: >**Wet1d**
10. The wet1d parameters currently *do not* appear in the dg screen, but they are present. Enter the name of the pulse for the parameter **wetshape='*pulsename*'**. Enter the power

as **wetpwr=#** and the pulse length (in microsec.!) >**pwwet=#.** Check all the other proton parameters: **tof, sw, pw, d1** Often a short d1 is preferable. The proton parameters should match those used when calculating the pulse. Make sure **ss=2** minimum.
11. Check that >**c13wet='n'** This turns off the carbon decoupling. (check with >**dps**) Be sure to use more than one steady state scan. Use at least **ss=4**. Set **d1** appropriately.
12. To decouple carbon satellites, set >**c13wet='y'**. The parameters **dmfwet** and **dofwet** correspond to the offset and decoupler modulation frequency for the carbon decoupling and also need to be set for the decoupling.
13. >**go** Adjust gain accordingly.

Additional Hints:

- Optimize the delay parameter **dz** for better suppression. Shaped pulse widths (**pwwet**) can also be optimized. Optimized values tend to be lower than the calculated value. To optimize **pwwet**, set **nt=1, ss=-2, ssfilter='n'.**
- If the shaped pulse is not working, check it using pulsetool from a terminal. Be sure to check that the **pwwet** is in microseconds, not milliseconds.
- Do not use autolock (**alock**) in these experiments, it will not work with the shaped pulse.
- If the spectral width or **tof** is changed, the shaped pulse must re-calculated.
- If there is an error with referencing, **create('reffrq')** and **reffrq=sfrq**.

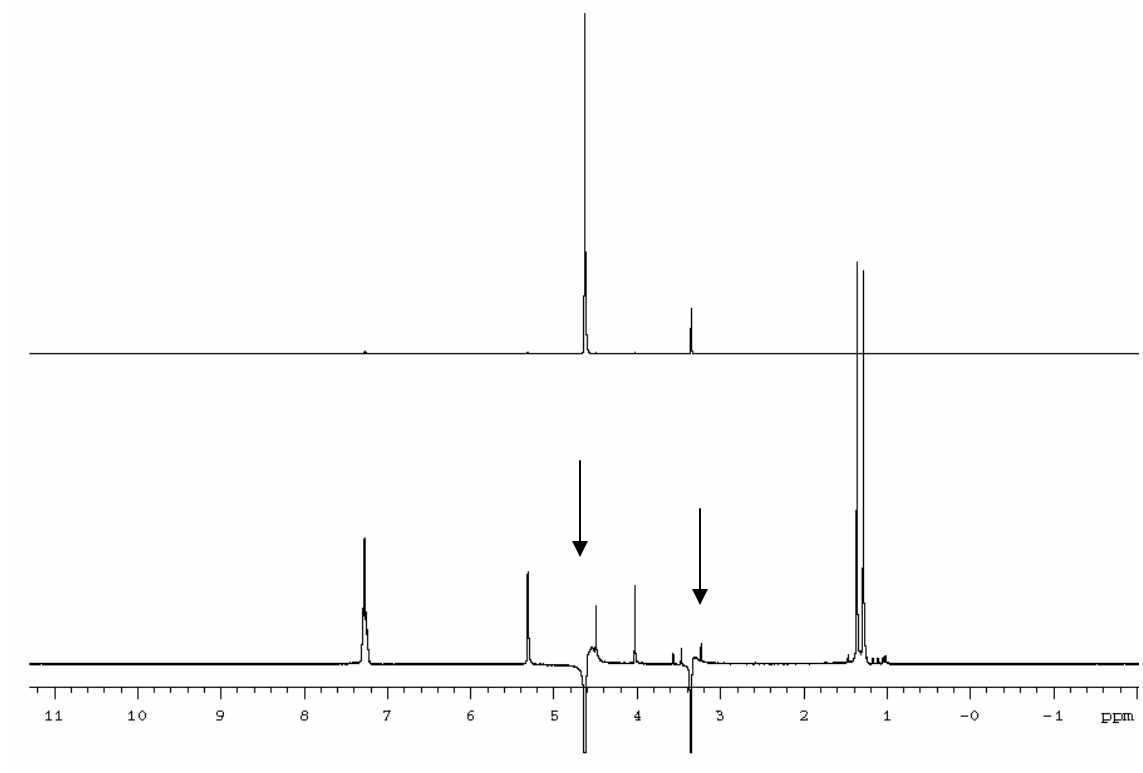


Figure 25: Double solvent suppression using a 1-D WET sequence allows the receiver **gain** to be increased and gives better signal-to-noise for observing the sample peaks.

WET Sequence and Multiple Solvent Suppression

(VNMRj 1.1D new interface instructions)

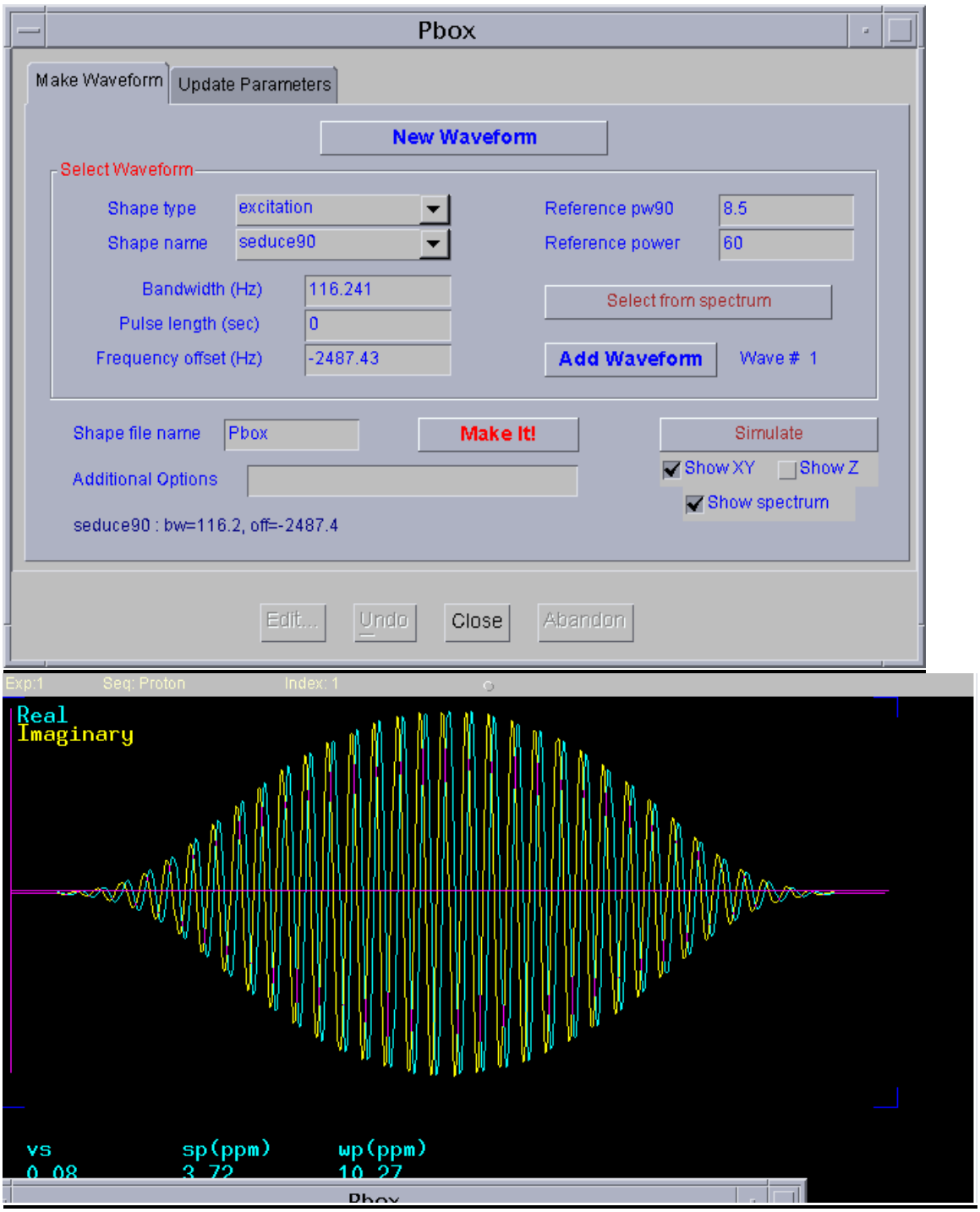
This pulse sequence uses a shaped pulse to suppress one or more solvent signals. The option of carbon decoupling is available for suppression of solvent signals with large C13 satellites. This pulse sequence requires a waveform generator (not available on Mercury 300).

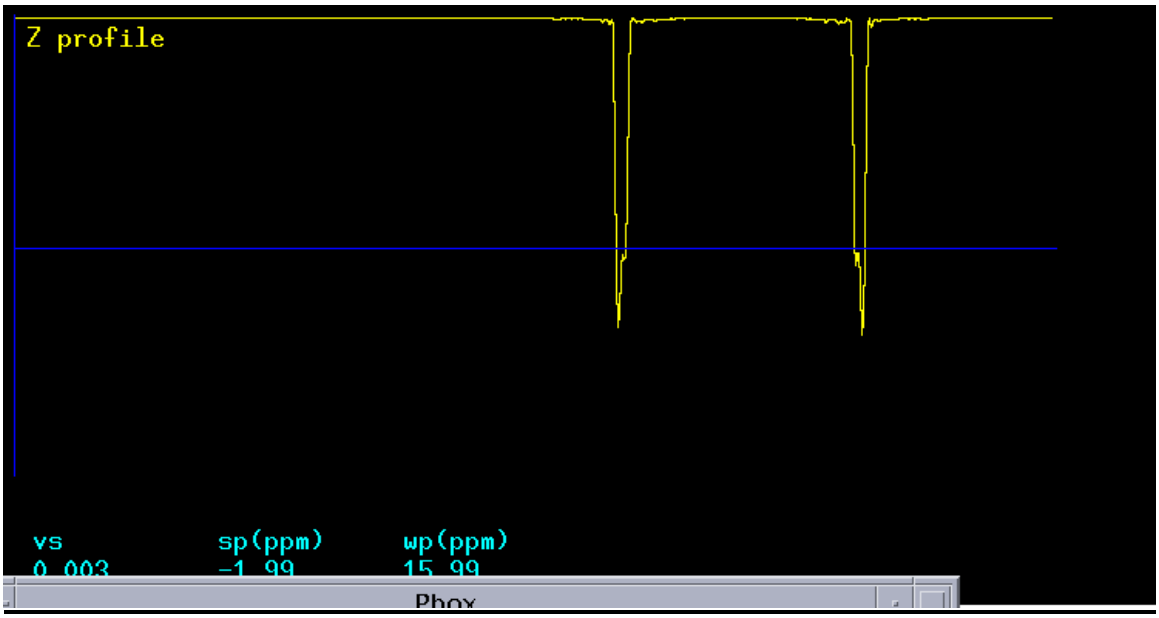
Ref: Smallcomb, S.H.; Patt, S.L.; Keifer, P.A.; "WET Solvent Suppression and Its Applications to LC NMR and High-Resolution NMR Spectroscopy", **Journal of Magnetic Resonance, Series A, 117**, 295-303 (1995).

1. Collect a 1-D proton spectrum. Optimize the window and offset and find the pw90.
2. Display a 1-D proton spectrum of the sample in the window and open the Pbox using **Process->Pbox** from the top menu. See Pbox figures below.
3. In the Pbox window, select **[New Waveform]**. Then select the pulse type as **[Excitation]** and the shape name **[seduce90]** (suggested shape names: seduce90 for multiple suppression, u-burp for 90% water)
4. Enter the reference pw90 and reference power based on the proton 90 degree pulse calibration of the sample. Enter the shape name in the bottom part of the Pbox window (this will be the name of the .RF file generated).
5. Place cursors in the spectrum around the peak to be suppressed. Select **[Select from spectrum]** in the Pbox window. Then **[Add waveform]**. Repeat this step if another peak is to be suppressed.
6. Then in the bottom of the Pbox window select [**Make it]**. This generates the pulse and the pulse will appear in the display screen.
7. To check the pulse, use the **[Simulate]** button. Close the Pbox.
8. Join another experiment. Select from the top menu **Experiments->Wet1d**
9. Check the proton parameters: **sw, tof, pw, tpwr, d1**.
10. Find the shape pulse parameters by opening a terminal and viewing the .RF file in the vnmrsys/shapelib. In this file will be values for pw and tpwr for this pulse. Enter them at the command line as **pwwet=#** (in microseconds!) and **wetpwr=#** (dB). Enter the shape pulse name as **wetshape=*pulsename*** Use >**dps** to view the pulse sequence. Unfortunately, these parameters do not appear currently in the **dg** screen, but they are present.
11. Adjust the **nt** and make sure **ss=2** (minimum). Adjust the **gain**. Use >**ga** to acquire the spectrum.

Additional Hints:

- Optimize the delay parameter **dz** for better suppression. Shaped pulse widths (**pwwet**) can also be optimized. Optimized values tend to be lower than the calculated value. To optimize **pwwet**, set **nt=1, ss=-2, ssfilter='n'.**
- If the shaped pulse is not working, check it using pulsetool from a terminal. Be sure to check that the **pwwet** is in microseconds, not milliseconds.
- Do not use autolock (**alock**) in these experiments, it will not work with the shaped pulse.


Pbox
Make Waveform
Update Parameters
New Waveform
Select Waveform
Shape type
excitation
Shape name
seduce90
Bandwidth (Hz)
116.241
Pulse length (sec)
0
Frequency offset (Hz)
-2487.43
Reference pw90
8.5
Reference power
60
Select from spectrum
Add Waveform
Wave # 1
Shape file name
Pbox
Make It!
Simulate
Additional Options
Show XY
Show Z
Show spectrum
seduce90 : bw=116.2, off=-2487.4
Edit...
Undo
Close
Abandon
Exp:1
Seq: Proton
Index: 1
Real
Imaginary
vs
sp(ppm)
wp(ppm)
0.08
3.72
10.27
Pbox


Z profile
vs
sp(ppm)
wp(ppm)
0.003
-1.99
15.99
Pbox