

Technical Bulletin #C0020: Direct Carbon Detection with CapNMR

This bulletin describes how to use the CapNMR probe for ^{13}C Direct Carbon on a Varian console/system.

Begin by loading an appropriate 'Proton' sequence. Make sure to use the appropriate power levels (see Figure 1). The solenoid microcoil in the CapNMR probe requires less transmitter power than traditional probes. The standard pulse width (pw90) is calibrated at installation for a transmitter power of 45 dB (tpwr=45).

Parameter	Varian Range	Bruker Range
Lock power	1 to 8	-55 to -45
Lock gain	34 to 48	110 to 130
Transmitter power level (dB)	40 to 45	15 to 20
90° Transmitter pulse width (μsec)	2.5 to 5	2.5 to 5
90° Decoupling pulse width (μsec)	5 to 12	7 to 12

Figure 1. Appropriate power levels for the CapNMR Probe

Once there is a good Proton spectrum, type 'Carbon' into the command prompt and this will load the Carbon pulse sequence (make sure tn='C13'). You can view the pulse sequence with a dps command. The sequence should look like the sequence in Figure 2.

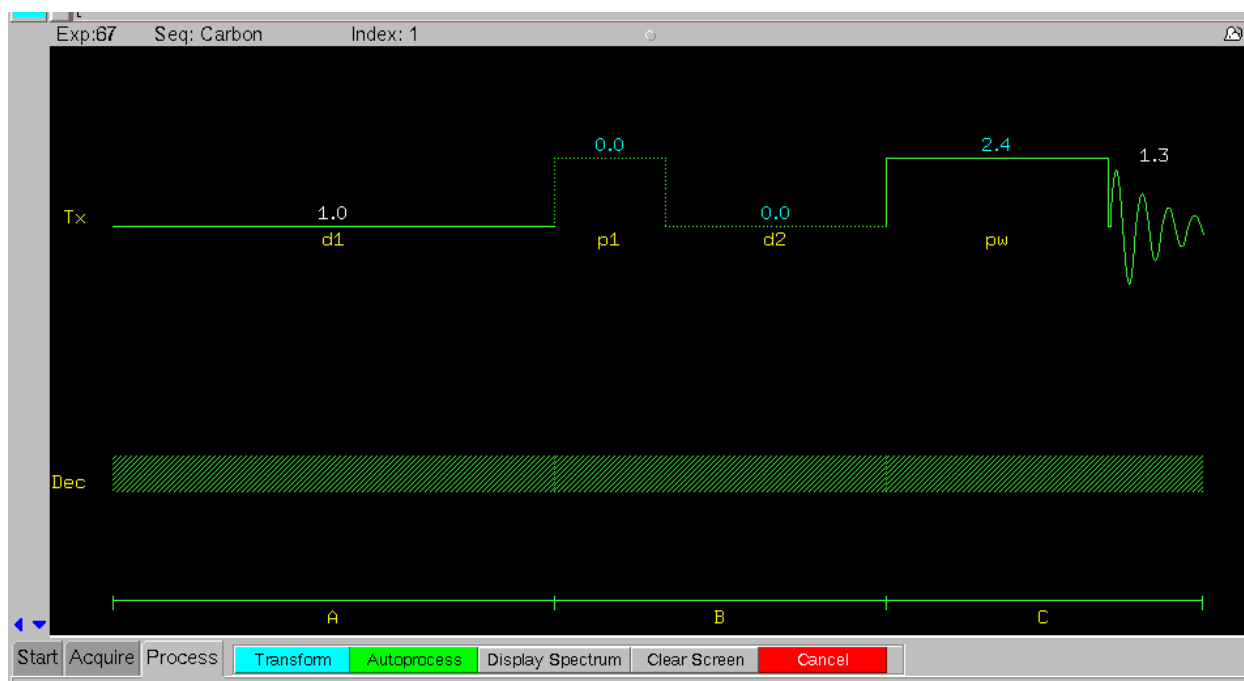


Figure 2. Direct Carbon Pulse Sequence

However, the pulse width is probe/console specific so your pulse width (pw) value should be different than the 2.4 value shown in Figure 2 and Figure 3. The pw90 for carbon was calibrated with a pwxcal sequence and for pwxlvl=45, the pw90 for this probe was 4.8 microseconds.

Note, most Carbon sequences will use a 45 degree pulse and set pw to half of pw90.

Here is an example of the parameter set used for the Carbon pulse sequence. In order to view this window, click on Process and select Text Output and then type 'dg'.

	ACQUISITION	TRANSMITTER	SPECIAL	PROCESSING			
seqfil	s2pul	tn	C13	temp	not used	lb	2.00
sw	36199.1	sfrq	150.826	spin	not used	sb	not used
at	1.300	tof	2339.0	gain	30	gf	not used
np	94152	tpwr	45	hst	0.008	awc	not used
fb	20000	pw	2.400	pw90	4.800	lsfid	not used
bs	64	DECOUPLER	alfa	10.000	fn	not used	
dl	1.000	dn	H1	PRESATURATION		FLAGS	
nt	3116	dof	0	satmode	n	il	n
ct	448	dm	yyy	satfrq	0	in	n
	SAMPLE	dmm	w	satdly	0	dp	y
date	Sep 15 2006	dpwr	23	satpwr	0	hs	nn
solvent	CDC13	dmf	200				
sample							

Figure 3. Direct Carbon Parameters for the CapNMR Probe

Note for older Inova consoles at 600 MHz:

Some older Inova consoles at 600 MHz should be re-cabled in order to detect direct carbon. If you have multiple relays in the back of your preamp tower, this might take care of the electronic pathway. **However, ensure that your proton channel either goes through a high band pass filter or is appropriate for constant proton decoupling. If you are not sure of the proper cabling configuration, Varian tech support should be able to confirm the correct cabling scheme for your specific Varian console.** [Varian technical support center (800) 356-4437]

Sample and Data Set:

The sample used was 10% (v/v) 2-ethyl-1-indanone in CDCl₃. Pure 2-ethyl-1-indanone is a clear, brown liquid with a T1 relaxation of approximately 0.8 seconds. This data set was taken on a **5- μ L of 10% 2-ethyl-1-indanone (519 μ g; 3.2 μ mol; v/v in CDCl₃)**, which was injected and then pushed into a 5 microliter flowcell with more CDCl₃ solvent. This data was taken on a Varian Inova at 600 MHz with a TXI (Triple Inverse ¹H, ²H, ¹³C, ¹⁵N) CapNMR probe with a 5 microliter enhanced flowcell in 2 hours.

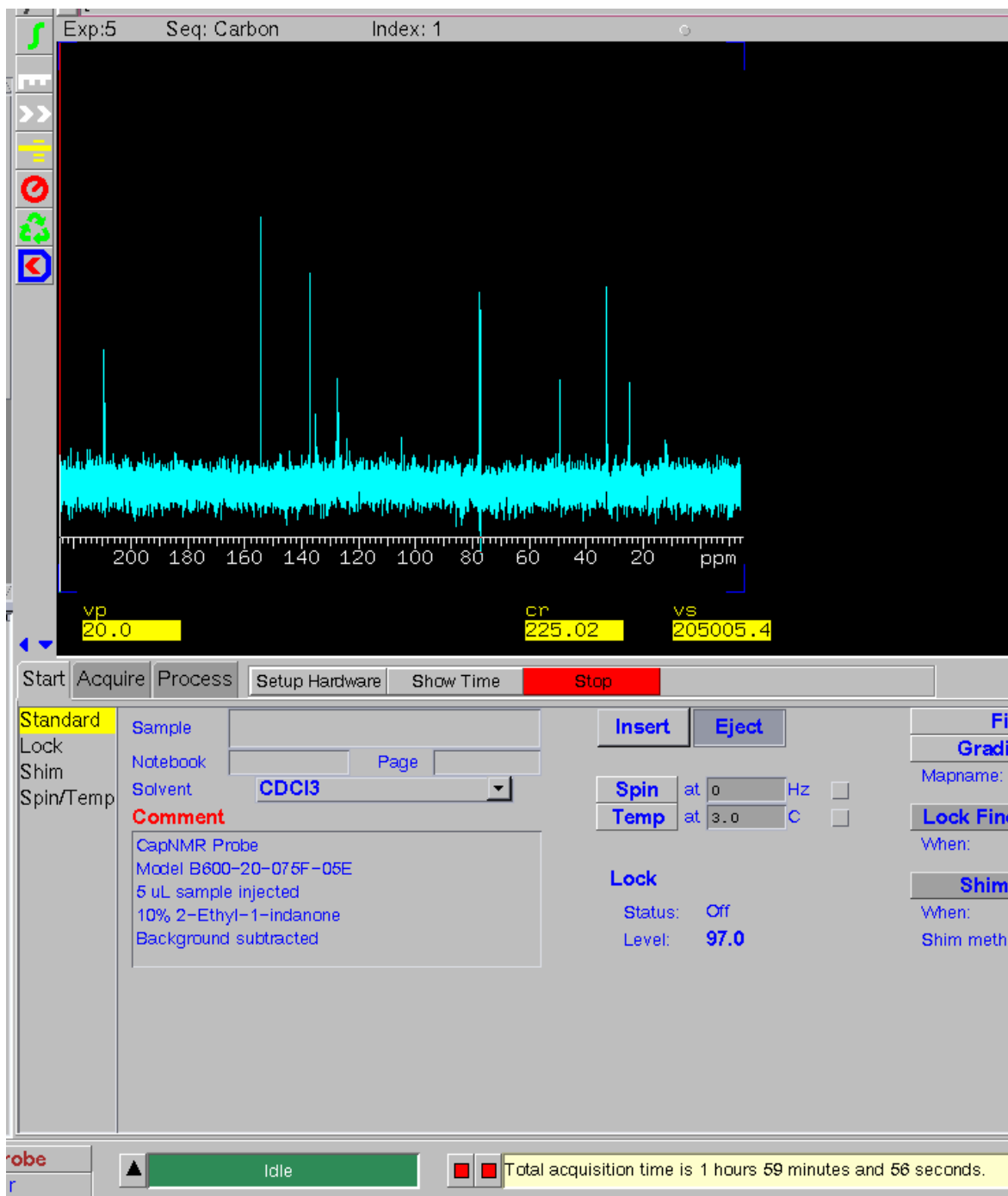


Figure 4. Direct Carbon Spectrum (Background Subtracted) from 10% 2-ethyl-1-indanone

Background Subtraction:

The CapNMR probe has some natural carbon background peaks that occur around 90 and 110 ppm and represent fabrication materials in the CapNMR probe. Varian software makes background subtraction very easy with the 'spadd' and 'spsub' macros. (See 'Tech Bulletin L0415 – Background Subtraction' or 'Varian Commands and Parameters Manual')

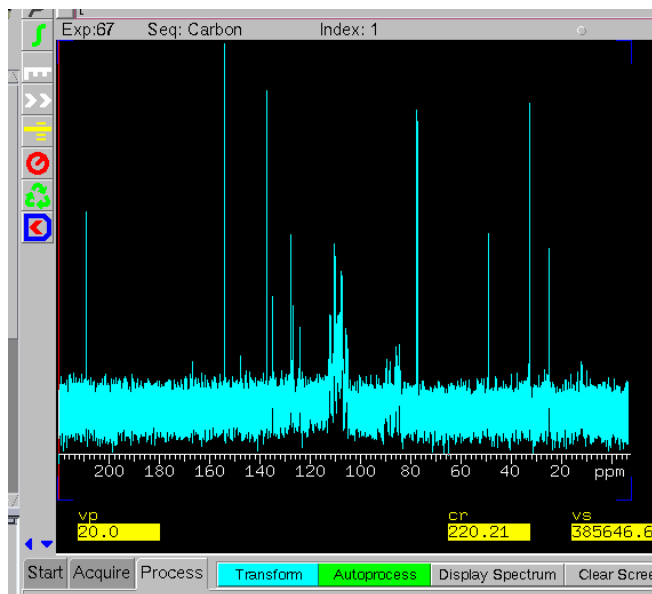


Figure 5. Raw Direct Carbon Spectrum

Figure 5 shows the full spectrum before any processing. Figure 6 shows the region of the spectrum in which the background occurs while Figure 7 shows the same region of data after the blank solvent subtraction using the spadd and spsub Varian macros.

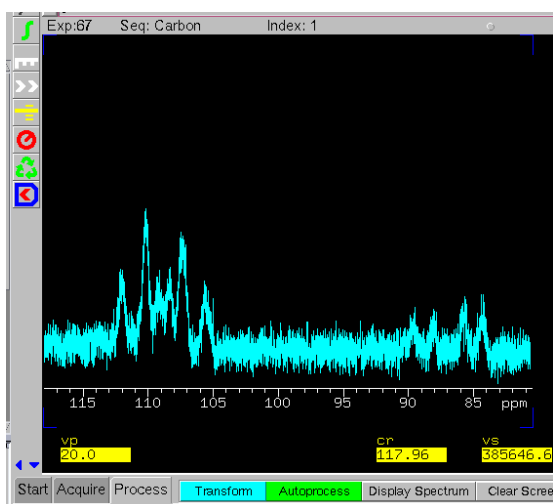


Figure 6. Background signal region before subtraction

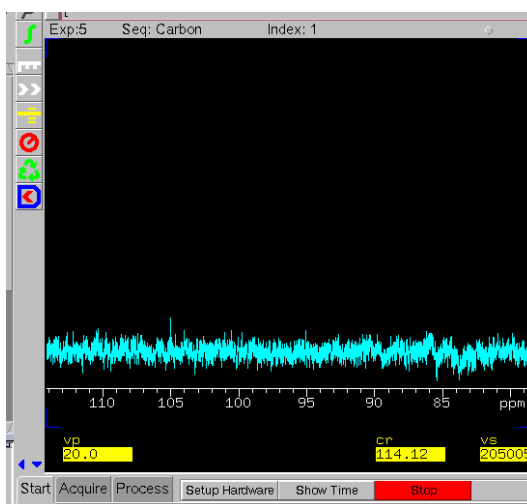


Figure 7. Background signal region after subtraction

For the blank solvent spectrum, another dataset was acquired on just CDCl₃ solvent with the same exact acquisition parameters, data points, and spectral window except at 1/6th the time ($nt=nt/6$) which is the 'blank'. This blank does not have to be same time as the original data set as the `spsub` macro can use a multiplier so the blank won't take as much time to acquire. (*For more details, see the 'Varian Commands and Parameters Manual' under `spadd` and `spsub`.*)

These natural carbon background peaks are very broad and are easily distinguished from the actual sharp sample peaks. However, a clean, background-subtracted, properly-processed spectrum is always desired. *See the final background-subtracted spectrum in Figure 4.*

Injection Method

A Hamilton Gastight 25- μ L syringe was filled with clean CDCl₃ solvent and used to rinse the probe. Then, 5- μ L of 10% 2-ethyl-1-indanone (519 μ g; 3.2 μ mol; v/v in CDCl₃) was drawn into the syringe and injected into the probe. Using the same syringe, 12 μ L of clean CDCl₃ was picked up and injected into the probe to push the 5- μ L sample into the NMR flow cell of the CapNMR probe. The 12- μ L Push Volume was calibrated in advance of sample injection.

Screenshots

All of the screenshots shown in this Technical Bulletin are from a Varian Inova console running Varian VnmrJ 2.1B software on a Unix workstation. The magnet was an Oxford 600 MHz magnet. (For more information on the Varian VnmrJ software, see <http://www.varianinc.com/cgi-bin/nav?products/nmr/>)