8. OPTIMIZATION OF THE ACQUISITION PARAMETERS

Congratulations – at the end of the last video you should have acquired your first NMR signal – the free induction decay. It's stored here in the Topspin interface under the “FID” tab, although you will also see it in the acquisition window. Let's take a closer look at the proton FID of this sample.

On the y-axis we have intensity. This physically corresponds to the voltage induced in the receiver coil by the precession of the net magnetization of the spin distribution that was temporarily reoriented by the transiently applied rf-pulse. Once “freed” from the influence of the pulse, the observable signal will die off due to relaxation processes. That's this exponential decay of the FID that we observe as a function of time. Time, in seconds, is our x-axis. And we know, from our acquisition parameters, as seen in the last video, that aq, the acquisition time, was set to 2 seconds.

The FID is an interferogram, which reflects the sum of all the peaks and all the troughs of all the individual proton signals that were recorded. Every set of protons with the same chemical environment will have the same precession frequency. And likewise, every set of protons with different chemical environments will have a different precession frequency. Since we expect, based on the label on the NMR tube of this sample, that the clear liquid inside the tube is a mixture of deuterated and protonated chloroform ($CDCl_3$ and $CHCl_3$, respectively), there should only be one proton signal. Its resonance or Larmor frequency corresponds to the effective magnetic field felt by the nucleus. The effective magnetic field is a combination of the static, external, homogeneous magnetic field of 4.7 T and shielding from the magnetic component of the electron density around the $CHCl_3$ hydrogen nucleus.

So, here we should have one frequency that decays exponentially. Let's look. Is it decaying exponentially? If yes, good, we're done. If no, if there's a step function that looks like a “box” then we probably forgot the Topspin command “rga” to automatically set the receiver gain.

Next, we're going to ask, has the signal completely relaxed before we turned off the detector? If not, we're probably truncating the FID. That will also give us a step function that will lead to an undesirable sinc function in our spectrum. A good “rule of thumb” is to have about half of the acquisition time recording noise. This is a compromise where we can quickly estimate an acquisition time, avoid truncation of the FID, to have good digital resolution, and not record too much noise that will be folded back into the spectrum. Don't change the acquisition time yet, first let's check the shimming.

For those of you following along the basic instructions, we're still on step number 7d). The “time domain” of the FID is a plot of intensity vs. time, how do we get to our NMR spectrum, or the “frequency domain” which plots intensity vs. frequency? We'll request the computer to perform a Fourier transform. We'll go over NMR processing in detail in the next video. Here our primary goal and interest is in optimizing the acquisition parameters.

Ta-da – the proton NMR spectrum of chloroform. What do we see? Two peaks! And our x-axis, which physically is energy, and then we divide by Planck's constant to express it as a frequency, appears as ppm. What's ppm? It's a way to normalize the chemical shifts given as an offset from a standard, defined as TMS = zero ppm, so that the value will be the same for every spectrometer, regardless of the magnetic field strength. So, here we see on signal around 7.25 ppm and the other ~1.5 ppm. What are they? Well, we have a very concise and useful NMR article here from the Journal of Organic Chemistry by Hugo Gottlieb et al, entitled”NMR chemical shifts of common laboratory solvents as trace impurities,” where Table 1 lists water in $CHCl_3$ at 1.5 ppm, consistent with a trace amount of water in the NMR sample.

Next, let's take a look at the line shape and ask ourselves, “how is the shimming?” This looks fine, but let's say for the sake of example, that every peak in the spectrum showed the same “shoulder” to the same side of the peak. Or in other words, let's see what bad shimming might look like. Remember from our previous video on shimming, that shimming corrects for external magnetic field inhomogeneities, and that this will affect every peak in the spectrum in the same way - as a consistent deviation from the expected Lorentzian line shapes across all the lines in the spectrum.

To see the effect of the shim gradients on the proton spectrum in real time, we're going to enter “go-setup” mode. First we're going to change d1 to 0 seconds. In gs mode, every aq + d1, which is 2 seconds total, we see a new FID, or if I toggle with this button here, a new spectrum. This is in real time. No data is being stored. Let's look at the lock display window at the same time. What happens when the shim is worse – like so? Now I'll undo the changes I made on the Z shim gradient by pressing the lit Z button. There we go. To leave gs mode I'll click stop. Nothing is saved and there's been no change to my data.

On our instructions we're still following step number 7d). Now we're going to define in our acquisition parameters the region of interest for our NMR spectrum. Our default parameters we used were sw = 20 ppm and o1p = 5 ppm, which matches our current spectral range from -5 to +15 ppm, check. The default parameters set a large sw to catch any surprises outside the ppm range that I expect. Now that I see what's in my sample, I want to set a more tailored region of interest. For the sake of example, I'll choose from 0 to 14 ppm. What's my sw? The difference between the limits 0 and 14, which is 14 minus 0, which is 14 ppm. I'll change sw = 14 ppm now.

What's the center frequency of the spectrum? Let's figure out the halfway point between our new limits of 0 and 14 ppm: half of 14 ppm is 7 ppm. So from each of the spectral limits, fourteen and zero, the center should be exactly at half sw. Or with our current numbers: fourteen on the left minus half-sw which is seven equals 7. And zero on the right+ half-sw, or 7 also equals 7. And there we go, seven is exactly half-way between zero and fourteen. Now we're experts at sw and o1p, and I'll set o1p to 7 ppm. The default sw was 20 and now it's fourteen. Since sw is smaller and dw is automatically changed by Topspin to one over twice sw, it is now larger. Aq is td times dw, so aq has also become larger. I want a long enough aq so that the FID has decayed down to the level of the noise at approximately half the acquisition time. I will adjust td, which is just a number, to get an appropriate acquisition time for this sample. While I'm thinking about parameters, I'll set one processing parameter now, although I can do it later, and that's si. I want to set si to twice td, for what is called “zero filling.”

The next parameter to optimize is ns. The signal to noise ratio of my spectrum will increase with the square root of the number of shots. So, to double the signal-to-noise ratio of my scout shot, I'll set ns to 4. This increases the signal intensity by 4 and the noise by 2, thus doubling the SNR.

One last thing that I want to point is the relationship between the oscillations we see in the FID and the precession frequencies of the nuclei. Here's our previously stored FID of this sample. The o1p was 5 ppm and our chloroform signal was at 7.2, an offset of 2.2 ppm or 440 Hz. So, here on the time axis, the inverse of 440 Hz is one waver per 2.3 ms. Now we've changed the center frequency to 7 ppm, what will happen? Let's start the acquisition – here are our 4 shots. Here's our new acquisition time on the x-axis. Here's the time remaining, Here's a zoom to see the oscillation frequency, which is the difference from the center frequency. This difference is taken AFTER the signal is detected but before it is digitized. The offset of the chloroform sample at 7.2 ppm from the center frequency at 7 ppm, is only 0.2 ppm or 40 Hz or one wave every 25 ms.

In the next video we'll go over these Topspin buttons up here and learn about spectral processing.