Summer 2014

Technion Magnetic Resonance Center
NMR Workbook

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Contents

NMR Samples

Solution	Chapter(s)
$CDCl_3$, 100%	1
or	
1-5% $CHCl_3$ in acetone-d_6	1
H_2O + D_2O, ~(2:1)	2
$CDCl_3$ + EtOH, ~(1:1)	3, 4
$CDCl_3$ + EtOH + *i*Pr, ~(2:1:1)	4, 5
DMF, 100%	6
MeOH, 100%	6
MeOH + D_2O, ~(1:5)	6

Abbreviations, physical constants, resources

0.1 Common abbreviations

NMR – Nuclear magnetic resonance

B – Magnetic flux density, magnetic induction, magnetic field strength (Tesla)

B^0– Static, external magnetic field strength

B^1– Transient, applied magnetic field strength

H – Magnetic field, magnetic field intensity (ampere/meter)

E – Energy

T – Tesla

I – Nuclear spin quantum number

J – Angular momentum

μ – Nuclear magnetic moment

γ – Nuclear gyromagnetic ratio

ν_L – Precession (Larmor) *frequency* of the nuclear spin in a magnetic field

σ – Shielding

rf – Radio frequency (MHz & kHz)

Hz – Hertz (1/seconds)

FID – Free induction decay

FT – Fourier transform

0.2 Physical constants (Bruker Almanac, 2007 p. 86)

k_B = Boltzmann's constant = 1.380 6505 (24) x 10^{-23} J/K

h = Planck's constant = 6.626 0693 (11) x 10^{-34} Js

μ_0 = Permeability of vacuum = 4π x 10^{-7} H/m

π = 3.14159

e = 2.71828

0.3 Fundamental relationships

$\Delta E = h\nu$	The Bohr frequency condition
$\nu_L = \mathbf{B}^0 \gamma / 2\pi$	The resonance condition
$\mathbf{B}^0 = \mu_0 \mathbf{H}$	Magnetic field

0.4 Resources . . . some starting points . . .

Bruker NMR guide: http://www.bruker.de/guide/

Technion NMR Facility, links: http://nmrlab.technion.ac.il

Encyclopedia of Nuclear Magnetic Resonance, Editors-in-chief, D.M. Grant, R. K. Harris
Chichester : Wiley, c199, **,REF 543.429 ENC** Chemistry and Biology Library

Bruker Almanac: copies available in the NMR lab

0.5 Aims

The aim of the following chapters is to provide some hands-on activities for the beginning user. Chapter 1 introduces the spectrometer. Ch. 2 focuses on shimming activities, ch. 3 on acquisition parameters, ch. 4 on resolution, ch. 5 on quantitative NMR, ch. 6 on heteronuclear experiments and finally ch. 7 deals with an introduction to the phenomenon of dynamic processes in NMR.

All of the topics broached during this short introduction can be described as the tip of the iceberg. As you delve deeper on your own into those ideas that most grab your attention, don't forget to have patience with yourself as you sift through the tremendous amount of information available on the internet, in the library, and from more experienced students, professors, and NMR personnel. Just as a teaser, consider: quantum computing, medical imaging, oil discovery, analysis of Antarctic ice cores using solely the earth's magnetic field, metabonomics, forensics, liquid crystals, non-crystalline solids, . . . the NMR Encyclopedia in the reference section of the ChemBio library spans 11 volumes. Good luck finding the balance between personal discovery using the spectrometers yourself and benefiting from the collective learning of more than six decades of NMR spectroscopists who have already paved some very clear and interesting paths into the previously unknown.

Chapter 1

Theoretical Background

1.1 Overview: magnets, rf, and computers

In general, there are three main components to a nuclear magnetic resonance (NMR) spectrometer:

1. *an unchanging, external magnetic field (B^0)*
2. *electronics, including a radio-frequency synthesizer, amplifiers, transmitters, a receiver, an analog-to digital converter, etc.*
3. *the user interface, i.e. a computer*

1.2 The magnetic field

A magnetic field naturally induces different energy levels in particles which have the quantum property named "spin." Many atomic nuclei have a non-zero magnetic moment, which means they have a non-zero quantum spin and can give rise to an NMR signal.

The magnet field of the earth is approximately 0.5 gauss, depending on your location. The earth's magnetic field is quite weak; it only induces an extremely small alignment of the nuclear magnetic moments.

i) Magnetic moments align in a magnetic field.

ii) The nuclear spin is closely related to the nuclear magnetic moment.

iii) One popular cartoon representation of single magnetic moment or spin, looks like this:

Activity 1.1

Knowing that 1 gauss = 10^{-4} Tesla (T), what is the earth's magnetic field in μT (microTesla = 10^{-6} T)?

Activity 1.2

Measure the strength of Earth's magnetic field with simple materials.

Reference:

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Measuring Earth's magnetic field simply
Gay B. Stewart, Phys. Teach. 38, 113 (2000)
*******************************************************************************************
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Compare your result with the calculated value of the Earth's magnetic field in Haifa at: http://www.ngdc.noaa.gov/geomag-web/#igrfwmm

You'll need to know **Haifa's latitude** and **longitude** for calculations. The following values came from http://www.getty.edu/vow/TGNFullDisplay (June 2004):

Coordinates:

Lat: 32 49 00 N *degrees minutes(*Lat: 32.8167 *decimal degrees)*

Long: 34 59 00 E *degrees minutes (*Long: 34.9833 *decimal degrees)*

The same site has this to say about ***Haifa***: Located on Mt. Carmel; conquered by Crusaders 1100; taken by Napoleon 1799; captured by Egypt 1839; surrendered to Turkey 1840; occupied by British 1918 & became part of Palestine mandate 1922; *[part of Israel since 1948;]* has steel mill, chemical plants & oil refinery.

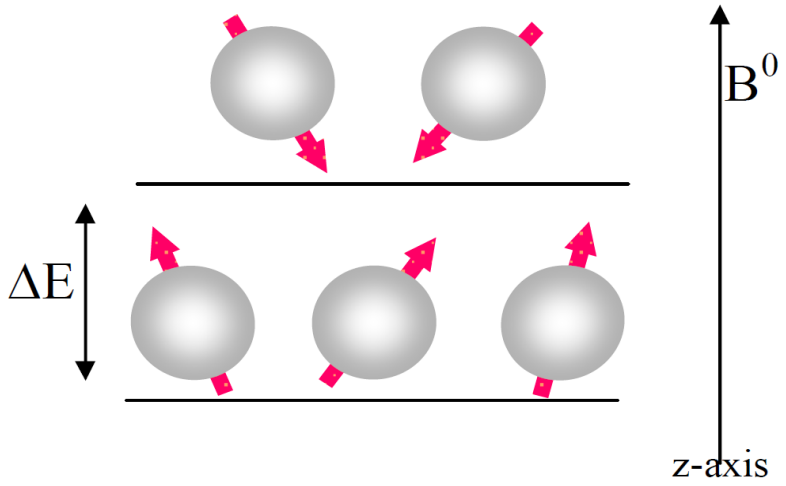
Activity 1.3

Check out an introductory NMR book and look up the relationship between the nuclear magnetic moment (μ) and spin angular momentum (J).

(hint: the library is on the 5th floor, the call numbers are in the 543.429 area)

Magnetic fields of up to 400,000 times the earth's magnetic field are currently available commercially. The impressively large superconducting magnet dominates most photographs of NMR spectrometers. The magnitude of B^0 determines the ultimate resolution and sensitivity of the NMR spectrometer.

When in a magnetic field the nuclear magnetic moments (spins) have different energy levels (called *Zeeman* energy levels) arising from different alignments in the magnetic field. The number of possible *Zeeman* energy levels is equal to $2I + 1$. The available energy levels are statistically populated by the nuclear spins in the magnetic field. It is helpful to have a pictorial representation of magnetic moments and populated energy levels (E) in a magnetic field (B^0). The following picture is for $I = 1/2$ particles where there are two Zeeman energy levels. The higher energy is populated by -1/2 spins, and the lower energy by +1/2 spins:



Activity 1.4

The ratio of the spin-state populations under steady–state conditions can be determined by the Boltzmann distribution:

$$\frac{N_-}{N_+} = e^{-\Delta E/k_B T} = e^{-\gamma \hbar B^0/k_B T} \qquad [1.1]$$

where $N_+ + N_- = N \equiv$ total population of particles with spin.

Calculate how many of the particles ($I = ½$) in a 7.0 T field are aligned "with the field" and how many are aligned "against the field."

Repeat the calculations for spin alignment in the earth's magnetic field.

The frequency condition, $\Delta E = h\nu$, tells us that *energy differences* and *frequencies* are equivalent. The differences in Zeeman energy levels (ΔE) depend on the strength of the magnetic field, according to the relationship:

$$\nu_L = B^0 \gamma / 2\pi \qquad [1.2]$$

Where γ (gamma), also called the nuclear magnetogyric ratio, is a fundamental physical property of atomic nuclei. Since $\gamma/2\pi$ is a constant (the same value) under all external conditions, the crux of the relationship is the direct proportionality between ν_L and B^0. The symbol ν_L represents the precession *frequency* of the nuclear spin in a magnetic field, also known as the Larmor frequency.

Activity 1.5

Fill in the following table. Use information available on the world-wide web (http://www-usr.rider.edu/~grushow/nmr/NMR_tutor/periodic_table/nmr_pt_frameset.html), in a Bruker Almanac, or a number of other reference books.

Isotope	γ	I	Natural abundance	4.7 T	21 T
^{1}H	28.2105 x 10^{27} J/T	½	99.984%	200 MHz	900 MHz
^{2}H					
^{12}C					
^{13}C					
^{15}N					
^{17}O					
^{18}O					
^{29}Si					

1.3 Radio frequency pulses

At the magnetic field strengths used for NMR spectroscopy the Larmor frequencies of atomic nuclei are in the MHz range. This is the same frequency range at which radio signals get transmitted. This is where the “resonance” in NMR comes to play. Resonance refers to an amplification response that occurs when frequencies are matched. Examples familiar to us include swinging on playground swings or pairs of tuning forks, where one can be made to ring by striking the second even though they are not in direct physical contact.

The electronics of the spectrometer, under control of the operator through the computer, transmit (send out) an rf-pulse synthesized to be at the same frequency as the calculated Larmor frequency of the sample (which is sitting in a *coil* in the B^0 field). This burst of electromagnetic irradiation (pulse) creates a temporary magnetic field (B^1) oriented perpendicular to B^0. The nuclear magnetic moments now change their orientation to be aligned along the new (effective) magnetic field axis (if we match our principle axis system to the Larmor frequency of our spin, we call it "being on resonance in the rotating frame." Within this mathematical framework we can ignore B^0).

When the B^1 field is turned off, the nuclear magnetic moments yet again reorient themselves, this time back to the original alignment and population distribution found in the B^0 field at equilibrium. The measured signal intensity acquired through the resonance phenomenon is detected through the same coil surrounding the sample. The received signal (emf; induced voltage in the coil) is amplified, digitized, and displayed on the computer screen for further mathematical processing.

1.4 The computer

The computer allows the user to control the timing of the pulses and delays (short breaks between pulses). There are five aspects of the B^1 field, induced by the rf-pulses, which are all under the control of the operator:

i) *pulse duration - μs to seconds (for a pulse train), this will also determine the bandwidth of the pulse*

ii) *pulse power – usually given in kHz ($\gamma B^1/ 2\pi$), although the amplifier puts out power in watts and the spectrometer usually requests dB (or how much to attenuate (reduce) the maximum amplifier output)*

iii) *pulse phase – in the sense that sin and cos waves are 90^o out of phase*

iv) *frequency – this is the center frequency of the pulse and also determines the middle of the spectral window (frequency domain)*

v) *shape – the basic pulse-acquire sequence most popularly uses a rectangular pulse*

The intensity (amplitude) of the received signal will depend on the number of resonating nuclei, the γ–value (a physical constant), and the duration, power, and relative phases of the B^1 field applied to induce resonance prior to detection. The degree of amplification of the observed signal is also under control of the operator, by means of the computer. As are the length of time that the detector is left on and the digitization rate of the signal.

The observed signal intensity is damped (decays) as the nuclear spins return to the B^0 field steady-state. This damped signal under observation is called a free-induction decay (FID) – and is the result of a pulsed-NMR measurement (as opposed to the continuous wave NMR experiments, which will not be covered in this workbook).

All the processing of the received signal (Fourier transform, phase corrections, baseline corrections, signal integrations, simulations, and plotting) are carried out using the computer.

Activity 1.6

Using a graphing program (or an internet based tutorial) generate two exponential decay curves with two different decay rates. Fourier transform the exponential decay curves. What is the correlation between line width of the transformed spectrum and the decay rate of the exponential curve?

1.5 Summary

In summary, the transmitted rf-pulses, when in resonance with the nuclear Larmor frequencies, induce changes in the nuclear spin states (spin gymnastics). The received signal follows the free decay (or relaxation) of the nuclear spins back to their equilibrium conditions. This detected signal is called a free induction decay (FID) and is plotted on the computer screen as intensity *versus* time. The FID is mathematically manipulated by a Fourier Transform to get an NMR spectrum, which is a plot of intensity *versus* frequency.

The details recorded in the NMR spectrum can be used to elucidate molecular structures, dynamics and interactions. It can also provide information about the relative concentrations of different molecules present in the same sample. Measurements of isotopes, such as ^{2}H, can provide information about the geographical origin of natural products.

Chapter 2

Homogeneous Magnetic Fields

2.1 Sample preparation

For the activities below, you will need to prepare two NMR samples:

1. D_2O + tap water (~2:1)
2. 1-5% $CHCl_3$ in acetone-d_6 (or just 100% $CDCl_3$ as an alternative)

To keep cost down, you can re-use a “thrift” or an “economy” NMR tube, just be sure it isn’t scratched or warped. For samples where maximum resolution at the given field is important, you should use high quality tubes. In general, shimming is easier at *lower fields* where slightly greater spatial variations of the external, magnetic field (B^0) can be tolerated. Easier refers both to the quality of the final result and the time needed to achieve it.

To prepare a sample in a 5 mm (outer diameter) NMR tube, first rinse the tube with the deuterated solvent you will be using. Then fill to a height of about three-fingers (~ 3 cm or ~ 600 μl) with your solution. Make sure that the outside of the NMR tube is clean and dry. Label and cap (or seal) the tube.

2.2 Shim gradients

The two types of magnetic resonance (MR) commonly encountered are *imaging* and *spectroscopy*. In basic MR imaging, a single chemical species (^1H_2O) is monitored as a function of position. In basic NMR spectroscopy, different ^1H species are separated as a function of effective shielding by local chemical groups, irregardless of position in the NMR tube. To understand the differences one must consider the affects of variations of the magnetic field in space (position) and the relationship between the Larmor frequency of the nucleus and the magnetic field:

$$\nu_L = \gamma B^0 / 2\pi$$

Imagine a water sample (every molecule having the same chemical environment). According to the relation above, we expect that all the hydrogen nuclei (protons) will resonate at the same frequency. But what if the magnetic field varies over space (ΔB^0)? Then each proton will resonate at a different frequency ($\Delta \nu_L$) as a function of its position (imaging). Which part of the body would you like to see imaged in this way? Check the web, there may be an example from MRI already on-line for viewing.

Now, what if the magnet field gradient is non-deliberate? The spectroscopist will get a range of resonant frequencies for each variation in the magnetic field, *i.e.* broad spectral lines. To get the desired narrow spectral lines, the magnetic field must have high *homogeneity* (no variation in the field within the sample space).

The two biggest technical challenges to overcome in NMR spectroscopy are ***resolution*** and ***sensitivity***. Broad lines negatively affect both aspects of NMR:

1. Resolution (the separation between different NMR signal lines arising from different chemical species) is reduced when each individual line is broad (chemical shifts and shielding will be covered in chapter 4).
2. Nuclear spin transitions are of relatively low energy, making NMR an insensitive method in terms of signal-to-noise ratios (SNR). Assuming the number of nuclei in the NMR tube is constant, broader line widths correspond to reduced signal intensities, further lowering the SNR.

To optimize resolution and sensitivity (SNR) at any particular field strength, current NMR spectrometers allow users to locally sculpt corrections to the magnetic field using coils and electric currents, called shim gradients for historical reasons. Currently automatic shim optimization routines are quite fast and accurate, moving the art of shimming into obsolescence – however, where automation is not available, it is still essential to learn to shim well to get good NMR results.

Recommended reading: Chmurny, G.N. and Hoult, D.I., "The ancient and honourable art of shimming," Concepts Magn. Reson., 1990, 2, 131-140.
also:http://nmrlab.technion.ac.il/NMR.htm#shim

2.3 Shimming on the solvent ^{2}H signal

The most common shimming method is the use the ^{2}H lock signal to monitor the local magnetic field over the effective sample volume. It is also possible to get a crude sense of the field homogeneity from the unlocked signal.

Activity 2.1

Shimming on the solvent ^{2}H signal is the method introduced when first learning to use the NMR spectrometer. This activity aims to help you look at the signal before being locked.

1) Place your D_2O + tap water sample into a spinner at the proper depth.
2) Place the NMR tube + spinner into the magnet using the lift air.
3) Display the swept ^{2}H signal on the computer screen (make sure the "sweep" button on the BSMS key pad is lit and open the lock display (lockdisp). There is one ^{2}H signal, due to the single ^{2}H species (D_2O) present in the tube. The signal "ringing" to the side of the resonance signal should also be apparent. The stronger and longer the ringing, the better the magnetic field homogeneity.
4) Read in a good shim file and note the effects on the ^{2}H signal.
5) Read in a poor shim file (or set all shim values to zero), note the changes in the signal.
6) Starting from the poor shim file (inhomogeneous magnetic field), change the z-shim and note the effect on the ringing. Can you find an optimal z-shim value?

Once the signal is locked the sweep is off, the intensity of the ^{2}H signal as a function of time appears in the lock window. As the integral of the NMR signal corresponds to the number of nuclei detected in the coil, the higher the lock level the narrower the line widths and the higher the spectral resolution and SNR. Most routine shimming relies on the ^{2}H lock signal due to its relative speed and convenience.

In the recommended readings, the rationale of gradient shims is explained in detail. This section ends with one of the many quotable thoughts to be found in the Churny and Hoult article:

> . . . to put our shimming task in mathematical terms, we must search for the global maximum on the error surface in the shim set hyperspace. In other words, we are searching for a maximum lock signal, with, say, 18 variables to adjust, in the possible presence of many inferior, false maxima. Most readers will need little reminding that such a process is tedious, difficult, and frustrating . . .

2.4 Shimming on the ^{1}H FID

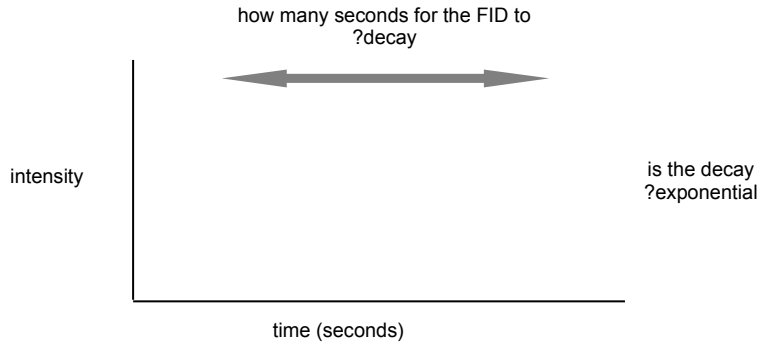
An FID of a single spin type (water protons, for example) should show an exponential decay provided a homogeneous magnetic field. Variations from ideality have characteristics that are well correlated with specific shim gradients (excellent examples under recommended reading, Section 2.2).

The biggest advantage to shimming on the FID is the additional information (compared to the ^{2}H lock signal intensity) about which particular shim gradient needs adjusting to improve the field homogeneity. Also, field adjustments are evaluated directly on the nucleus of interest.

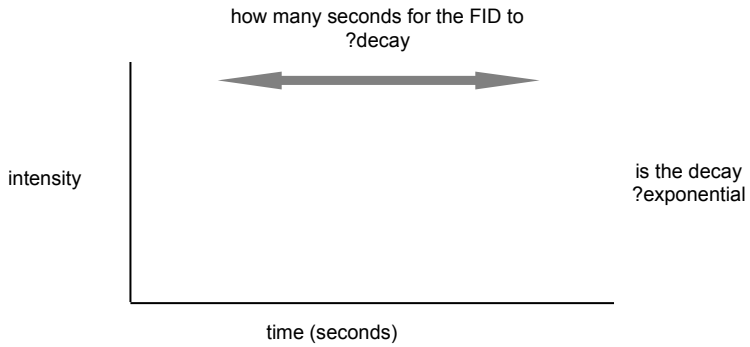
The major drawback to this approach to shimming is the often increased time needed. Under ideal working conditions the sample would relax back to equilibrium quickly allowing for real time adjustments while observing the FID. However, the mathematics of the Fourier Transform tells us as a certainty that narrow spectral lines, ideal for optimal resolution and sensitivity, must necessarily have long relaxation times (more difficult *working* conditions). Since the tolerable amount of variation in the external magnetic field B^{0} is always a fraction of the narrowest spectral line width, the samples needing the most exact shimming will have the longest relaxation times and be least practical for real-time shimming on the FID. The best way to understand these relations is through a hands-on approach.

Activity 2.2

1) Use your D_2O + tap water sample (it will have a relatively fast relaxation rate).
2) Read in a parameter set for a ^1H NMR measurement and a good shim file.
3) Reduce the pulse width by a factor of 10. (p1)
4) Reduce the first delay to 100 ms. (d1)
5) Go into go-setup mode (gs)
   the spectrometer will continuously pulse and acquire.
6) Sketch what you observe:



7) Coarsely (make sure the “fine” button on the BSMS keypad is **not** lit). Make a large change in the z-shim. Sketch what you observe:



Activity 2.2 (continued)

8) Fill in the following table:

SHIM gradient	SPIN? (Y/N)	FID decay time (s)	FID features (sketch)

9) Try other shims and spinning conditions. Record your observations in your notebook. Compare your observations with figures in the recommended reading (Section 2.2).

10) Repeat all of the above with the chloroform sample, which should have narrower line widths (longer relaxation times).

2.5 Shimming on the ^{1}H spectral line

The shapes and widths of the NMR lines themselves are always the final test of whether or not the magnetic field was sufficiently shimmed to homogeneity. In addition to shimming on the ^{2}H lock signal or the FID, you can also use the NMR spectral lines for shimming.

If relaxation of the spins is fast enough, you can shim while observing the spectral lines in gs mode. Otherwise it is necessary to work iteratively: *i)* acquire a spectrum, *ii)* evaluate the line shapes and widths, *iii)* apply shim corrections based on suggestions in the literature (Section 2.2), *iv)* repeat steps *i* - *iii* until satisfied with the magnetic field homogeneity.

Activity 2.3

Understanding the effect of each shim gradient on the spectrum helps "in the field" when trying to improve a poor shim. The recommended reading is an excellent supplement.

1) Use your chloroform sample (it will have a relatively long relaxation rate).

2) Read in a parameter set for a ^{1}H NMR measurement and a good shim file.

3) Acquire a single scan, Fourier Transform and phase.

4) Fill in the following table:

SHIM gradient	SPIN? (Y/N)	FID decay time (s)	Spectral features (sketch)

5) Try other shims and spinning conditions. Record your observations in your notebook. Compare your observations with figures in the recommended reading (Section 2.2)

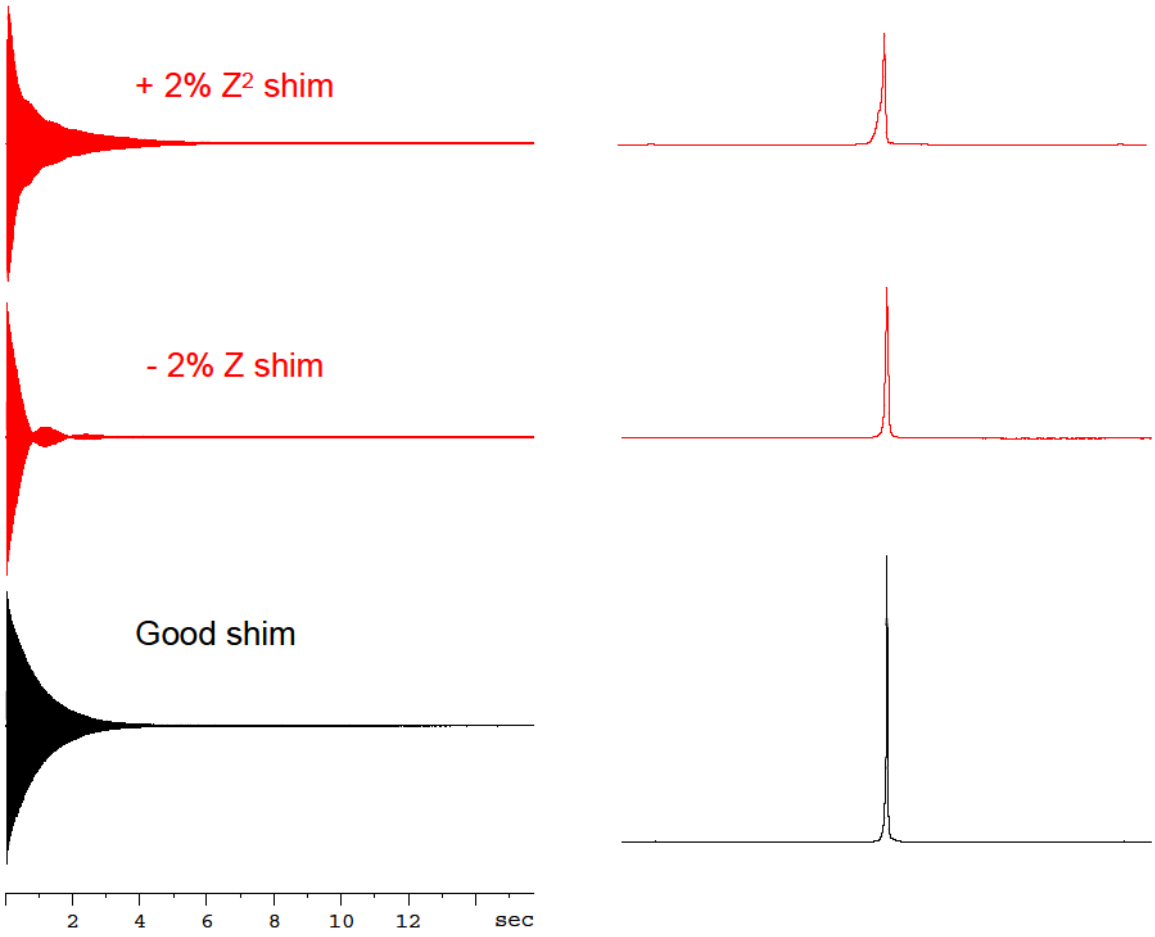


Figure 2.1. Effect of shim current changes on FIDs and spectral lines. Data acquired on a Bruker Avance 500 Spectrometer. ^{1}H NMR of 3% $CHCl_3$ in acetone-d6.

2.6 Automatic shimming

Various routines for automatic shimming have been developed over the years. Most automation routines typically took longer and gave poorer results when compared to a trained user. However, Bruker's “topshim” routine bucked the trend and actually shims very quickly with excellent results.

2.7 Summary

The maximum resolution and sensitivity that you can achieve at a particular magnetic field depends on the quality of the shimming. The lineshape is the final measure of the shim quality. The more you shim, the better you’ll get at it.

Chapter 3

Controlling pulses and delays

3.1 Sample preparation

In addition to the water/D_2O sample prepared in chapter 2, you will also need to prepare a roughly 1:1 $CDCl_3$ + ethanol sample.
More detailed instructions for sample preparations can be found in Section 2.1 or: http://www-usr.rider.edu/~grushow/nmr/NMR_tutor/pages/plan/plan_levela1.html

3.2 NMR spectrometer variables

Application of radio-frequency (rf) radiation can produce observable NMR signals, which are themselves electromagnetic waves picked up by the NMR receiver. If Activity 2.2 succeeded, you should be familiar with the NMR signal recorded in the time domain (the free-induction decay, FID). Ideally you are quite comfortable looking at the FID. The goal of this chapter is to familiarize you with the four properties of electromagnetic waves: frequency, phase, amplitude, and duration. In addition, the *delays* when the spectrometer is waiting and the *acquisition* when the FID is being detected and digitized, are two more parameters having duration.

3.3 Frequency of radiation

The centrality of the relationship $\nu_L = \gamma B^0 / 2\pi$ to magnetic resonance should be becoming obvious, simply by the number of times it appears. Resonance occurs and a signal is generated when the applied radio frequency ($\gamma B^1 / 2\pi$) matches the nuclear precession frequency (ν_L) in the B^0 magnetic field (Section 1.3).

Why not do one NMR experiment and observe multiple nuclei? If one could generate a large enough excitation and observation bandwidth (range of

frequencies) and digitize signals of very different intensities with a high digital resolution, then such an experiment should be possible.

Recommended reading: Cano, K.E., Smith, M.A., Shaka, A.J., "Adjustable, broadband, selective excitation with uniform phase," J. Magn. Reson., 2002, 155 , 131-139.

Standard methods (2014) limit spectral bandwidths to a few hundred kilohertz. Within these limitations, the NMR spectroscopist has control of the frequency range to be observed (for Bruker, the sw parameter defines the spectral width). The excitation bandwidth of the pulse will be inversely proportional to its duration. Thus short ("hard") pulses have a broad excitation profile and long ("soft") pulses can be used for selective excitation. Such control becomes important in advanced NMR experiments.

Activity 3.1

This activity aims to help you discover the meaning of "on" and "off" resonance. The tasks are to put an NMR signal "on-resonance" and to understand the effects of offset frequency on the FID.

Start with the water sample; it will be simplest since it only has a single resonance. To avoid truncation, reduce the pulse duration by a factor of ~ 10 (p1). After acquiring a single shot using typical parameters, determine *if* there is an off-resonance offset of the water signal: Is the water signal in the *exact center* of the spectrum?

Yes means that it is on-resonance.

No means that there is an off-resonance offset.

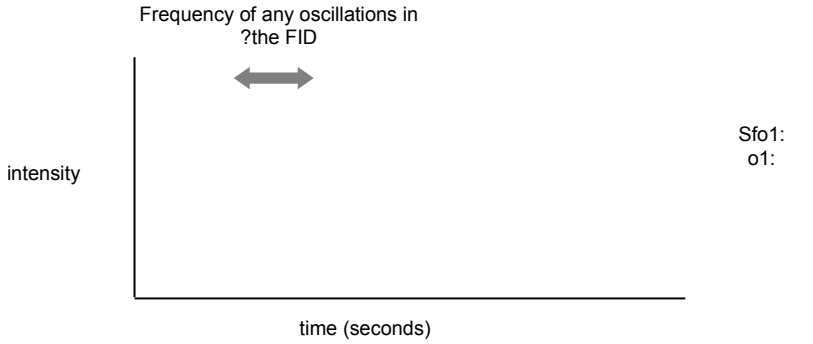
The current "carrier" frequency (center of the applied rf) should be listed in the acquisition parameters (AcquPar tab, or type: ased). List your current values of: BF, SFo1, o1 and o1p. Note the *units* of the different parameters.

Notice which values are included by default on your spectrum when you print a hard copy with parameters.

Activity 3.1 (continued)

Put the water signal on-resonance. There are several different possible methods. For example, set the parameter o1p equal to the chemical shift of the water signal. Alternatively, use the red lightning bolt icon (.setto123). Or, in gs-mode change sfo1 until the oscillations in the FID are gone.

Hopefully you now see the characteristic FID for an on-resonance signal. Sketch what you see and fill in the value of the carrier frequency being used (sfo1).



Fill in the following table to help yourself discover the correspondences between resonance offset and the appearance of the FID (oscillations).

Carrier frequency o1 (Hz)	Offset of spectral center from water shift	Oscillation frequency of FID	FID features (sketch)
	0		
	+100		
	-100		
	1000		
	-1000		

3.4 Phase of radiation

Dividing a 360° rotation into four gives the most common phases in use: $\pi/2$, π, $3\pi/2$, 2π (equivalent to 90°, 180°, 270°, 360°; also represented as 0, 1, 2, 3 and x, y, -x, -y).

Since both the applied rf radiation and the received electromagnetic radiation are sinusoidal waves they will have a phase, defined by the spectrometer relative to a reference signal. With modern spectrometers, practically any angle can be defined.

Activity 3.2

The phase of the signal after Fourier transform, without additional phase corrections, depends solely on the position of the first point of the FID. The amount of zero order phase correction (phc0) or first order phase correction (phc1) used can be found in the ProcPar tab (or type: edp).

To observe the first point of the FID, first right-click and choose Spectral Display Properties . . . and check “show data points”, then zoom in on the first portion of the FID the same way you would do a zoom in the frequency domain. It will be easier to see if you separate on the display the “real” and “imaginary” components of the signal ($\cos x + i \sin x$ = Re + Im).

1) Add a column in the margin to the table in Activity 3.1 called “zero-order phase correction”. For each entry write in the value of the zero-order phase correction needed to get a spectral line that is correctly phased as a positive absorption signal (phc0).

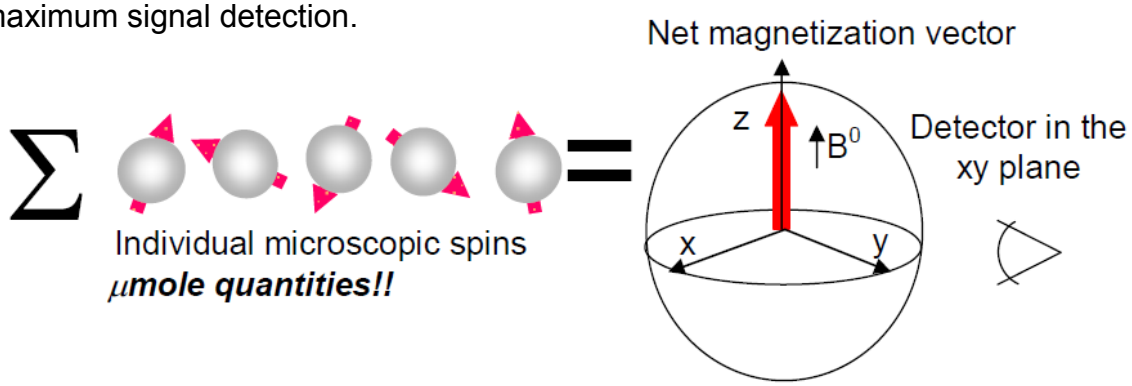
During standard signal acquisition, "phase cycling" is used in order to correct for pulse imperfections and DC offsets. Phase cycling deliberately alters the phase of the transmitted pulse, usually through a pattern of four or eight different settings, for each separate shot of signal averaging (ns 8, for example). The phase cycling is often easiest to observe when the signal is on-resonance.

2) Place the signal on resonance. Be sure the signal intensity is not being truncated by the filters (reduce the pulse duration, p1, if necessary). Set ns to 8 scans and type zg <enter>. Is the phase of the FID varying with each scan?

3.5 Amplitude and pulse duration

How much of the signal is detected by the receiver can be intuitively contemplated using the model of the net magnetization vector.

The tip angle (α) of the net magnetization vector depends on the duration (τ_p) and the amplitude (γB^1) of the resonant rf radiation: $\alpha = (\tau_p)(\gamma B^1)$. A 90^0 pulse (or $\alpha = \pi/2$ pulse) rotates the net magnetization vector completely away from its equilibrium position (parallel to the static magnetic field) into the xy plane for maximum signal detection.



Activity 3.3 will be an exercise in calibrating the tip angle for a constant rf amplitude (*i.e.* power, γB^1) by varying the pulse duration. This is called a "nutation" experiment. Advanced NMR experiments often call for pulses of a specific tip angle; a quick calibration can often be done in a matter of minutes. The addition of adding shapes to the pulse envelope is another versatile and powerful, but more advanced NMR tool.

Activity 3.3

Perform a pulse nutation experiment to calibrate the tip angle of the observed NMR signal. Use the $CDCl_3$/EtOH sample. We'll focus on the chloroform peak.

1) Acquire a spectrum with the residual chloroform ^1H signal *on resonance*. A single scan is sufficient. Use the same phase parameters for each Fourier transform (***pk*** and ***not*** *apk*).

Activity 3.3 (continued)

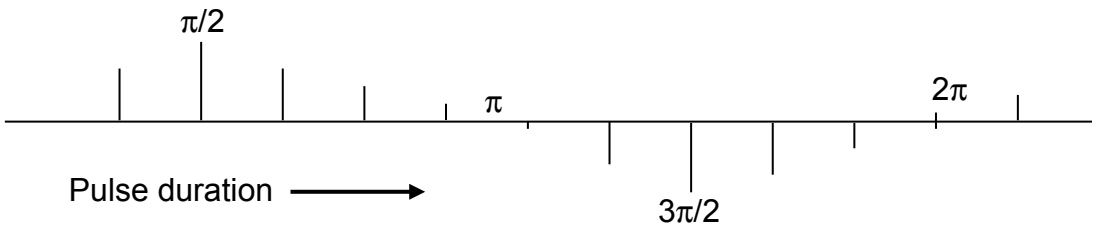
2) The default parameters are usually set to approximately a 90° pulse; record the current p1 for the given pl1.

3) Double the pulse duration (in μs). What happens to the signal?

4) Double the pulse duration again. What happens to the signal?

5) Make a plot of signal intensity versus pulse duration:

Each line represents the *single* chloroform signal. Each signal was acquired using a *different* pulse duration (x-axis), and plotted according to its absolute y-intensity (y-axis). Find and record the pulse durations (μs) required to see a 90°, 180°, 270°, and 360° signal:



3.6 Delay times

During rf pulses, perturbation of the quantum mechanical spins away from equilibrium is induced. There are simple "hand-waving" pictures to get a basic, intuitive appreciation of this phenomenon (such as the net magnetization vector model). Such models are extremely important, allowing spectroscopists to speak a common language, essential for effective communication. The more sophisticated models (density matrix theory, average Hamiltonian theory, product operator theory, Floquet theory, . . .) offer physical insights into the observed process.

During delays (referring to those times within the NMR pulse sequence when there is no externally applied rf field), evolution of various spin-spin interactions and relaxation processes occur.

In this section we will focus on relaxation effects (T_1) that occur during the repetition delay (a delay separating multiple repetitions of the same NMR experiment applied in order to increase the experimental signal-to-noise ratio).

Activity 3.4

A technique used to eliminate an NMR signal is to saturate it by continuous irradiation. By preventing relaxation, the NMR transition is no longer observable. Similarly, if the time between pulses is shorter than the time needed to relax back to equilibrium, the NMR signal will become saturated and disappear from the spectrum.

How much time should be allowed for relaxation between repetitions of a single pulse experiment to avoid signal saturation? One should wait five times the spin-lattice relaxation time constant (T_1) to allow for full (>99%) relaxation.

Saturate the NMR signals of the $CDCl_3$/EtOH sample by using too short a repetition delay.

1) Acquire a normal spectrum and be sure that all signals are observable.
2) Set the number of scans (ns) to 8 and dummy scans (ds) to 0.
3) Reduce the repetition delay (d1) to a fraction of a second.
4) Acquire while observing the FID.

What happens to the signal-to-noise ratio as the number of scans increases during signal averaging? This problem, in practice, is most acute when trying to directly acquire signals from low-γ nuclei in a rigid conformation, common to ^{13}C=O groups or ^{29}Si signals.

3.7 Acquisition times

Once the final rf pulse has been turned off, those spins induced away from their equilibrium condition are free to decay back to their original, unperturbed states. Relaxation via spin-lattice (T_1) interactions was discussed in the previous section. A second process known as spin-spin relaxation or the transverse relaxation rate constant (T_2) is occurring simultaneously and independently. The

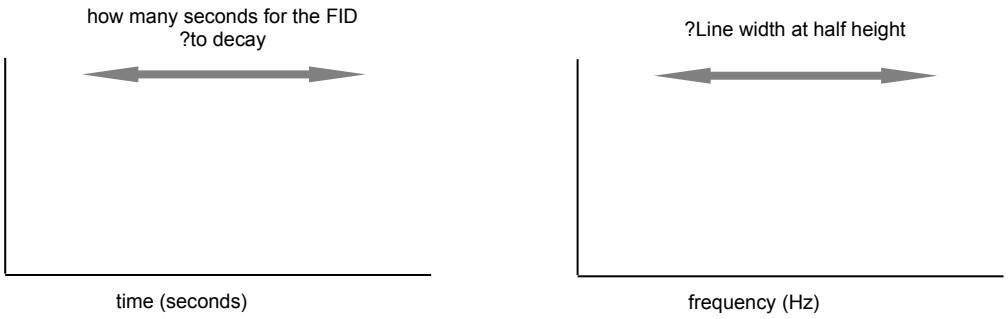
single restriction being that T_2 is always less than or equal to T_1. The time scales of the two processes are usually similar in the liquid phase, whereas in solids T_2 and T_1 can have order(s) of magnitude differences.

The main associations with T_2 relaxation include dephasing of the coherent signal and the relationship between spectral line width and decay time (the faster the decay, the broader the line width at half-height after Fourier transformation).

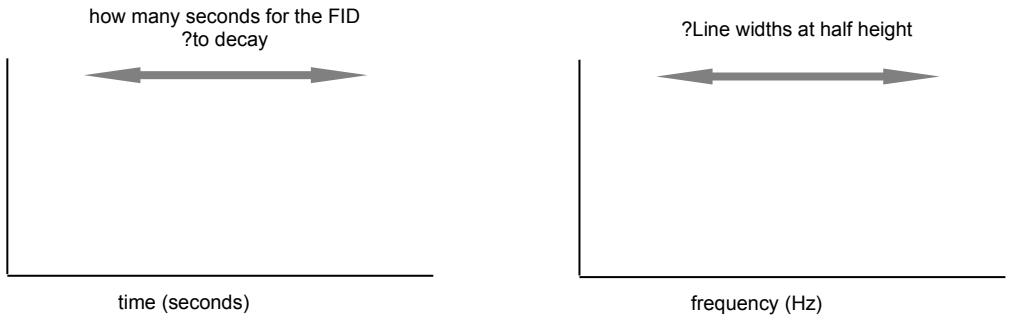
Activity 3.5

Compare the decay times and the spectral line widths of the two NMR samples.

1) D_2O/Water sample:



2) $CDCl_3$/EtOH sample:



3.8 Quantitative NMR: Single pulse excitation

It is explained in all basic NMR texts that the integral of the NMR signal is directly proportional to the number of spins present in the sample. This is true *provided* that all the acquisition parameters have been correctly set *and* the system is not undergoing significant dynamic processes (to be covered in Chapter 5).

What needs to be set to be sure that the NMR spectrum will be quantitative?

1) All frequencies are equally excited. It would not be quantitative to compare an on-resonance signal that experienced a 90° pulse with an off-resonance peak that was only rotated a few degrees.
2) The time permitted for spin-lattice relaxation between pulses is 5 * T_1 for the signal taking the *longest* time to return to equilibrium. Although this does not necessarily need to include the solvent peak, comparing a fully represented signal to a partially saturated signal would give a quantitative comparison of the respective number of atoms present.
3) The detector is on long enough; the FID should decay by the first third of the screen. This is to avoid truncation of the signal in the time decay. Fourier transform of a truncated signal leads to wiggles in the base line near the resonance peak which will distort the determination of the area under the curve. If the acquisition time is too long, unnecessary noise is being folded into the spectrum.
4) Signal-to-noise ratios are sufficiently high for accurate integration.
5) The baseline is straight and smooth, allowing for accurate integration.
6) Correctly set bias and slope of all integrands.
7) The signals to be compared are either fully resolved or analyzed using a reliable deconvolution routine.

Activity 3.6

Each of the parameters below can be matched with a number in the list given above. Fill in the appropriate number next to the parameter:

|__ ns |__ sw |__ p1 |__ d1 |__ d1 + aq |__ aq |__ td |__ o1 |__ SF |__ SFO1

The two most expected questions at this point are:

i) How do I correctly set the parameters to do a quantitative measurement?

ii) The numbers I get are good enough – what's all the fuss and bother about?

How meticulous you are in checking your acquisition parameters depends on the question that you are asking. If you are simply checking your crude product

to determine whether or not novel signals can be detected, then working with default values should be good enough (although it is important to know about the possibilities of rf saturation or exchange dynamics to avoid being fooled into accepting a false negative). On the other hand, if you need an accurate measure of an impurity within a mixture, then not only is it essential to take care with all the parameter settings, it is also important to determine the precision (reproducibility) of your measured values through multiple measurements.

3.9 Summary

A basic understanding of how the spectrometer works and how the parameters affect the NMR results is important for obtaining data that best represents the sample under analysis.

Chapter 4

Spectral Interpretation

4.1 Samples

We will continue to use the $CDCl_3$ + ethanol sample plus a new sample:

$CDCl_3$ + ethanol + isopropanol ~(2:1:1)

4.2 Chemical shielding

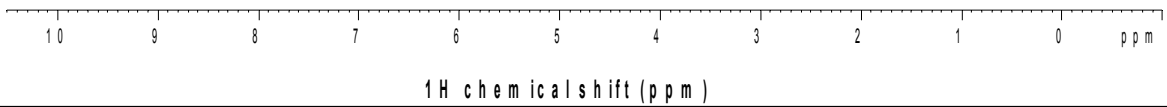
Recall the relationship between nuclear energy (ν_L) and magnetic field (B^0). The power of NMR spectroscopy to solve chemical problems comes from a very small additional shielding term (σ), giving:

$$\nu_L = (\gamma / 2\pi) * B^0 (1 - \sigma).$$

In molecules, the electrons circulating around the nuclei generate their own electromagnetic fields. A bare nucleus in a magnetic field would have a much higher frequency (less shielded) than a (more shielded) nucleus within a molecule, where each nucleus experiences a unique, *effective* magnetic field depending on the extent of shielding from the *local* electronic environment.

Activity 4.1

Where would you expect the following ^1H species to appear? CH_3, CH_2, CH, OCH_3, $Si(CH_3)_4$, C_6H_6, $H_2C{=}CH_2$, COOH, OH, NH - plot their positions on the axis below:



As the external magnetic field (B^0) is increased, the resolution between the different Larmor frequencies (ν_L) is improved.

Activity 4.2

With the $CDCl_3$ + EtOH + iPro sample, compare the resolution between the two CH_3 resonances at two different fields (*i.e.* 200 MHz *vs.* 400 MHz). Make the digital resolution (Hz / point) at the two fields comparable by adjusting sw and td.

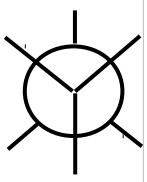
4.3 Scalar coupling

Further power of NMR spectroscopy comes from resolution of spin-spin interaction energies, called scalar or J-couplings; mediated through chemical bonds and extremely sensitive to geometry and substituent groups.

Activity 4.3

In samples where there is a large difference between the chemical shift values ($\Delta\sigma$) and the J-couplings ($^3J_{HH}$), the spectrum is considered "first order" and interpretation follows the introductory explanations.

1) Draw the Newman projection of EtOH and mark down the expected J-couplings: $^3J_{HH}^{expected}$ = _____Hz



2) Acquire a spectrum of the $CDCl_3$ + EtOH sample and print an expansion plot of the alcohol peaks. The plot should include "peak picking" (the chemical shifts (also record the chemical shift for each multiplet in Hz (according to the center position of the multiplet). How close is the experimental splitting value for the CH_2 and CH_3 groups (Hz) to the expected value? How can you account for any variations?

3) How do the chemical shift values and the J-couplings for the alcohol compare at two different fields (*i.e.* 200 MHz *vs.* 400 MHz)?

B^0 (MHz, ^1H)	δ_{OH} (ppm / Hz)	δ_{CH2} (ppm / Hz)	δ_{CH3} (ppm / Hz)	$^3J_{(CH2\text{-}CH3)}$ (Hz)

When coupled nuclei have similar, but non-identical, chemical shifts, "higher order" effects appear in the spectrum, for example the familiar "roof effect."

Activity 4.4

When dealing with higher order spectra, it is often desirable to tease out the components giving rise to the observed spectrum. This can be accomplished using simulations.

1) Download the simulation package Spinworks 2.5 by Kirk Marat: ftp://davinci.chem.umanitoba.ca/pub/marat/SpinWorks

2) Open Spinworks → Help → Contents → Simulation tutorial → ABX spectrum and follow the tutorial to learn how to run NUMMRIT.

3) Vary the relative positions between two coupled protons: start with 2 distinct doublets and end with a pronounced roof effect. You may want to experiment with the spectral parameters given by Keeler (chapter 2, page 15) in your simulations.

4.5 Summary

NMR measures chemical shifts, J-couplings, and peak areas. It may not sound like much at first, but it actual gives a lot of information about which molecules are in the solution, how many are there, and what are their structures.

Chapter 5

Double Resonance

5.1 Sample

You will only need the $CDCl_3$ + EtOH + iPro sample used in the last chapter.

5.2 Heteronuclear scalar couplings

In Section 4.3 J-couplings were first introduced. These through-bond interactions only require that the two interacting nuclei have spin (I) that is non-zero. As you learned in Activity 1.5, ^{12}C (98.9% natural abundance) does not have a magnetic moment (I = 0), however ^{13}C does (I = 1/2 ; 1.1% natural abundance).
We use the relation $2nI + 1$ to determine the multiplet pattern that will result from through-bond spin-spin coupling, where *n* is the number of neighboring nuclei and I is the quantum spin number.

Activity 5.1

Collect a high signal-to-noise ratio ^1H spectrum of the $CDCl_3$ + EtOH + iPro sample. Expand the y-axis until it is scaled up enough to view the ^{13}C satellites. Since each group of ^1H's sees only one ^{13}C neighbor, *n* = 1 and I = 1/2. A ^1H-^{13}C doublet centered around the ^1H-^{12}C singlet should be resolvable at 0.5% the singlet's height. What is the separation between the doublet signals in Hz?

$^1J_{CH}$ = ___ Hz

Activity 5.1 (continued)

Use the same $CDCl_3$ + EtOH + iPro sample. Change the irradiation frequency to ^{13}C and acquire a ^{13}C spectrum (check the probes power levels!).

The lower frequency tells you the energy transition is smaller and the observed signal will be weaker. When you acquire a carbon spectrum, you will only detect signal from the 1% of the carbon nuclei that have a non-zero spin. You will need to collect significantly more scans compared to the ^{1}H spectrum.

What was your final ns = ___?

All the hydrogen nuclei attached to ^{13}C are spin-1/2, n = 2 for the CH_2 groups and n = 3 for methyl groups. What are the expected multiplicities of the ^{13}C signals coupled to ^{1}H?

^{13}C data	$^{1}J_{CH2\text{-}EtOH}$ (Hz)	$^{1}J_{CH3\text{-}EtOH}$ (Hz)	$^{1}J_{CH2\text{-}iPro}$ (Hz)	$^{1}J_{CH3\text{-}iPro}$ (Hz)
Multiplicity?				
Experimental value?				

5.3 Decoupling

When you look at the cables connecting the rf electronics to the probe head in the magnet, you should see at least three cables: ^{2}H (lock), ^{1}H and X. The X stands for a non-hydrogen nucleus and usually includes ^{13}C NMR.

"Gating" the rf is to turn on the irradiation. Since ^{1}H and ^{13}C have separate rf cables, it is possible to irradiate the sample simultaneously at both frequencies. For instance, on the ^{13}C channel we can apply a hard pulse and acquire at the carbon frequency. Simultaneously, ^{1}H rf can be turned on or off at different times giving different spectral results. In Bruker's Topspin program, you can see a graphical display of the pulse sequence by typing: showpp <enter>.

If the proton rf is gated on during X acquisition, any multiplets in the X spectrum due to J-coupling with a neighboring spin-1/2 proton will be collapsed into a singlet, giving improved resolution and SNR. The proton rf saturates the ^{1}H

transitions killing the scalar spin-spin coupling. Gated proton rf, while the X-spins are at equilibrium, transfers magnetization from nearby protons via the NOE effect.

Activity 5.2

For X-acquisition with ^{1}H decoupling, the pulse program is called "invgate" and the file name is “zgig”. Draw the pulse sequence (time on x-axis, power on y-axis, a separate line for each frequency channel).

^{1}H _____

^{13}C _____

Read in the standard parameters (13C_1Hdec) and acquire a spectrum.
How many scans were needed to get a decent ^{13}C spectrum with ^{1}H decoupling?

ns = ___

5.4 Nuclear Overhauser effect

It still requires significantly more time to acquire a ^{13}C spectrum relative to the ^{1}H spectrum. Resolution and signal-to-noise ratio enhancements achieved by ^{1}H decoupling (collapsing multiplets into singlets) raises the question of what other tricks can be played to get more information faster. A significant amount of research, thought, and energy is dedicated to improving NMR by finding methods to enhance the small signal. What if NMR could be done on significantly smaller amounts of material in a significantly faster time? The physical limitation originates with the small amount of polarization in the magnetic field according to the Boltzmann relation explored in Activity 1.4.

Albert W. Overhauser proposed in the 50’s a method for transferring polarization between electrons and nuclei to derive an enhancement factor of >600.

Today, the Overhauser effect more generally refers to driving a change in polarization between two coupled nuclear spins.

The nuclear Overhauser effect (NOE) is used ubiquitously in NMR structural studies. It is a useful method for determining through-space ^{1}H-^{1}H proximities. It is also routinely used in heteronuclear NMR experiments in order to enhance the ^{13}C signal intensity via the higher ^{1}H polarization.

Activity 5.3

For X-acquisition with ^{1}H decoupling, the pulse program is called "powgate" and the file name is “zgpg”. Draw the pulse sequence (time on x-axis, power on y-axis, a separate line for each frequency channel).

^{1}H _____

^{13}C _____

Read in the standard parameters (13C_1Hnoedec) and acquire a spectrum.
How many scans were needed to get a decent ^{13}C spectrum with ^{1}H decoupling?

ns = ___

5.5 Polarization transfer

The proton signal has the largest intensity since it has the largest gyromagnetic ratio (γ), excluding tritium. One way to enhance the signal intensity of a lower-γ nucleus is to use a spin-spin interaction to transfer the larger proton polarization difference (Boltzmann) to the coupled nucleus.

For example, the NOE can transfer polarization through-space. However, the NOE would not be appropriate for enhancing ^{15}N or ^{29}Si signals since those nuclei have a negative γ and hence the effect is negative (“dehancement”).

Another example is to transfer polarization through-bonds using the J-coupling interaction. The Hz units remind us that *i)* it is an interaction *energy* since E = hν, and *ii)* in the time domain it corresponds to a nuclear precession frequency.

5.6 Spectral editing

The number of protons attached to an X-nucleus affects the magnitude of the J-coupling and the multiplicity. Creative uses of rf irradiation and delays have been developed to tease out these differences in a robust and graphical way. For instance, edited ^{13}C spectra usually show 180^0 phase differences between methyl and methylene carbons making assignment quite straightforward.

Activity 5.4

You may want to set up an automation program to run one experiment right after the other for comparison. Be sure to make all the acquisition and processing parameters as similar as possible for comparison (ns, rg, total experimental time, sw, td, pulse calibrations, aq, etc.)

Signal-to-noise (SNR) ratios can be measured by integrating over a region of noise compared to a selected signal.

5.6 Summary

Although the majority of NMR measurements are still proton spectra, many other NMR sensitive nuclei are available for investigation. Knowing when to leave things coupled, add decoupling, and use enhancements increases the usefulness of multinuclear NMR studies.

Chapter 6

Chemical Exchange

6.1 Samples

i) *neat* MeOH (dry)

ii) MeOH + D_2O ~(1:10)

iii) *neat* DMF

6.2 Dynamic NMR

X-ray crystallography beautifully provides the conformation of a molecule with fixed positions in space. Once in solution dissolved molecules are disordered, they flow, they are flexible. In solution NMR the picture captured by the spectrometer depends on the “shutter speed” relative to the rate at which the molecules are dancing.

When the molecular conformation is fixed relative to the NMR timescale, sharp spectral lines are expected for each resonating nucleus in a unique chemical environment. When a particular nucleus jumps or moves around between several different electron environments quickly relative to the NMR timescale, sharp spectral lines are expected corresponding to the average local magnetic field experienced by the resonating nuclei. And when the motion is neither fast nor slow relative to the NMR timescale, but intermediate, a “fuzzy” picture, where the lines are “smeared” results. Better stated, intermediate motions on the NMR timescale result in broad NMR spectral signals.

Activity 6.1

Figure out whether broad lines can be modeled solely by two different alternating frequencies in the time domain, corresponding to a nucleus experiencing two different effective magnetic fields during the NMR acquisition.

See, for example, Malcolm H. Levitt, Spin dynamics: Basics of nuclear magnetic resonance, Chichester : Wiley, c2001 **543.429 LEV**

6.3 Hydrogen bonds

As hydrogen bonds break and reform the electron density at the hydrogen atom's nucleus changes, changing the effective magnetic field felt by the proton, changing its resonance frequency. Water resonates at 1.56 ppm in $CDCl_3$ and at 4.87 ppm in CD_3OD (Gottlieb, *et al*, *J. Org. Chem*. **1997**, *62,* 7512-7515). This tells us that a hydrogen nucleus participating in a hydrogen bond is deshielded and resonates at a higher frequency.

Activity 6.2

Sketch your prediction of the ^1H NMR spectrum of methanol. The methyl protons resonate at 3.31 ppm. $^3J_{HH}$ between the OH and the CH_3 protons is 4.6 Hz (**SDBS No.:** 3302).

_____

δ_H (ppm)

Acquire a ^1H NMR spectrum of neat methanol at room temperature. Sketch your results.

_____

δ_H (ppm)

What happened to the J-coupling?

6.4 Variable temperature NMR

The NMR timescale is determined by B^0. The rate of chemical exchange may depend on pH, concentration, contaminants, salt content, temperature and pressure. Experimentally, it is relatively easy to change the solution temperature while keeping the other parameters constant.

Activity 6.3

Observe the effect of temperature on chemical exchange using the neat methanol sample. Try the following temperatures: -65°, -10°, +10°, and +37°C, which are shown in Figure 9.2 of Harald Günther's book NMR spectroscopy: Basic principles, concepts, and applications in chemistry, Chichester: Wiley, c1995, chapter 9, **543.429 GUN**.

Methanol is a standard NMR thermometer used for -98° – +57°C (Van Geet, *Anal. Chem.* **1968,** *42,* 2227). The chemical shift of the methyl group is insensitive to temperature. The hydroxyl group resonance frequency decreases as the temperature increases. At higher temperatures the average hydrogen bond length increases resulting in a deshielding of the hydroxyl proton. A calibrated temperature is calculated based on the chemical shift difference between the two resonance signals in the proton NMR spectrum of methanol. Water in the methanol sample will change the chemical shift of the OH group and give a false temperature reading. Similarly, for higher temperatures, ethylene glycol can be used as a NMR thermometer.

Activity 6.4

Graph the calibrated temperature as a function of the nominal temperature (the temperature read off the spectrometer's VT unit) and use a linear fit to be able to interpolate for any temperature. Instructions for determining the calibrated temperature can be found in Van Geet's article, referenced above, in Harald Günther's book, referenced in the previous activity, or on-line at http://nmrlab.technion.ac.il/pdfs/Avance/TempCal.pdf

6.5 Chemical exchange

One way to begin appreciating the many beautiful examples of chemical exchange in solution is with Alex Bain's introduction at http://www.chemistry.mcmaster.ca/bain/Intro.pdf. His review article (*Prog. Nucl. Magn. Reson. Spectrosc.* 2003, *43,* 63-103) begins, "Almost everyone who has done NMR has seen some evidence of chemical exchange." If you do Activity 6.5 you will squarely be on the list of those who have. In general, adding D_2O to an NMR sample can often be used to identify exchanging protons in a proton NMR spectrum.

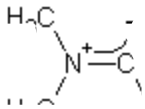
Activity 6.5

First, acquire a proton NMR spectrum of methanol. Second, acquire a proton NMR spectrum of methanol diluted in D_2O (approximately 10% MeOH).

What happened to the OH peak that could be seen in the first sample?

6.6 Hindered rotation

Let's follow Harald Günther's example and begin by considering *N,N-dimethylformamide* (DMF).



The partial double bond between carbon and nitrogen restricts the rotation around the N-CO bond. The asymmetry of the hindered molecule means that when rotation around the N-CO bond is slow on the NMR time scale the two different methyl groups have different resonance frequencies. When the methyls undergo fast rotation on the NMR time scale, then a single methyl peak at the average chemical shift is observed.

It is pointed out on page. 345 of Günther's book that

> Between 1956 and 1969 the values determined for the energy of activation for the internal rotation of dimethylformamide . . . rose from 29 ± 12 kJ mol^{-1} (7 ± 3 kcal mol^{-1}) to 118 ± 8 kJ mol^{-1} (28.2 ± 2 kcal mol^{-1}).

Activity 6.6

Use a neat DMF sample to measure the activation energy for the internal rotation around the N-CO bond.

Use Spinworks to simulate the data (ftp://davinci.chem.umanitoba.ca/pub/marat/SpinWorks/ this is the same program used in Activity 4.4, you only need to install it one time). Instructions for determining the activation energy can be found in the Help menu of Spinworks (dynamic NMR tutorial).

How close do you get to the accepted value of 85.8 ± 0.8 kJ mol^{-1} (20.5 ± 0.2 kcal mol^{-1})?

6.7 Kinetics and thermodynamics

As Alex Bain writes in his review, cited in section 6.5:

> Exchange involves the passing of the molecule through a transition state, at the top of a kinetic barrier. Measurements of the rate as a function of temperature . . . are one of the few ways of experimentally determining the height of the barrier.

We should keep in mind the warnings issued by Binsch and Kessler in their highly sited article (*Angew. Chem.* **1980,** *92,* 445-463):

> Such care [in temperature measurements] is mandatory, to be sure! If one is not willing to inform oneself about the possibilities of error and to invest the minimum expenditure of effort, it is preferable to abstain altogether from any attempt at evaluating the temperature dependence of *k* [chemical exchange rate] quantitatively; expecially [*sic*] with regard to activation entropies the pertinent literature is already burdened with such a heavy load of ballast that we can easily do without more of the same.

6.8 Summary

The dynamic nature of molecules in solution affects the NMR results. The spectral lines of any molecules undergoing chemical exchange need to be considered with respect to the NMR time scale at which they were measured.

Postscript

Further Reading

As was the stated goal at the outset, we have scratched the surface of NMR. For those of you who learn best by doing, this workbook will have achieved its goal if you have managed to overcome your own personal activation energies to get a sample made and into the magnet. Murphy's law and a workbook of ideas to guide your path will, I hope, have taken care of the rest.

I'll close by strongly recommending further reading for more "hands-on" activities:

Stefan Berger and Siegmar Braun, 200 and more basic NMR experiments :a practical course, Weinheim: Wiley-VCH, c2004 **543.429 BER**

or either of its earlier incarnations:

S. Braun, H.-O. Kalinowski, S. Berger, 100 and more basic NMR experiments :a practical course, Weinheim : VCH, c1996

S. Braun, H.-O. Kalinowski, S. Berger, 150 and more basic NMR experiments :a practical course, Weinheim : Wiley-VCH, c1998