VIII
CHEMICAL EXCHANGE

Lecture notes by Assaf Tal

Chemical exchange has surprising ties with relaxation as we shall see. Understanding exchange lets us understand phenomena, some of which at first glance seems totally unrelated:

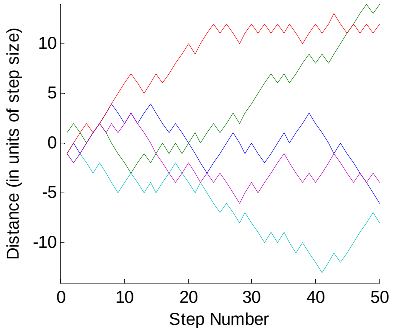
- Why do some peaks disappear when certain solvents are used?
- How is T_2 related to the fluctuating fields?
- Why don't we see any effect of the electron spin on the nuclear spectrum (in diamagnetic samples)?
- Why and when does motional narrowing occur?

At the heart of exchange phenomena lies a very simple model: as a spin "jumps" from one molecule to the next, it retains its magnetization, but its precession frequency changes. Thus, we can model chemical exchange as a single spin with a time varying resonant frequency, ω(t).

1. A TOY MODEL

1.1 RANDOM WALKS

Imagine a drunk person starting at x=0 and taking a step of size *d* either left or right with equal probability in each time unit. We call this a random walk. If we plot the position of an ensemble of, say, 5 drunkards as a function of time (number of steps, to be precise) for 50 steps, we'll get something that looks like this:



We see that while averaged over all drunks the distance from the origin is 0 (as expected from an equal-probability walk), the drunks seem to be getting farther from the origin. In fact, the average absolute distance from the origin (the root mean square, to be exact) is

$$\Delta x_{rms} = d\sqrt{N} \ .$$

If the time between steps is τ, then, after a time t, during which we have taken N=t/τ steps, the drunks' average RMS is

$$\Delta x_{rms}\left(t\right) = d\sqrt{N} = \sqrt{\tfrac{d^2}{\tau}t} \ .$$

in the limit τ→0 and d→0 (with $\frac{d^2}{\tau}$ constant) we get a classical diffusion problem which can be used to accurately describe the diffusion of, say, molecules in water. The quantity

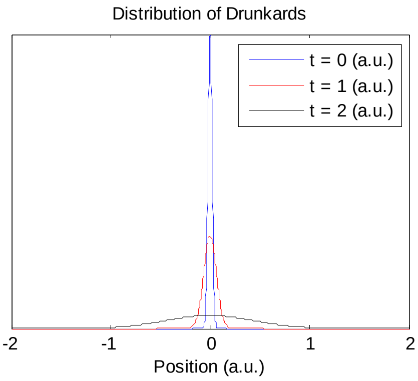
$$D = \tfrac{d^2}{2\tau}$$

is called the diffusion coefficient (for water molecules in a glass of water, it is about 2 $\mu m^2/ms$), and we can write

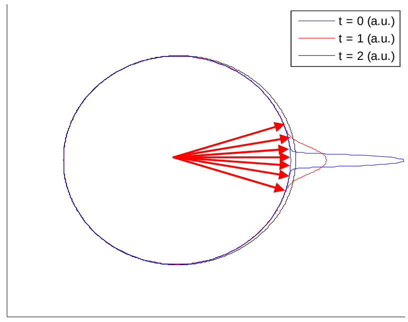
$$\Delta x_{rms}\left(t\right) = \sqrt{2Dt} \ .$$

This can be used to measure microscopic properties of molecular collisions simply by looking at their diffusion coefficients (something we won't be doing here).

When looking at a large number of drunkards it makes sense to discuss the **probability of finding a drunk** or the **spatial distribution of drunks**. If all the drunkards begin at the origin (x=0), then this distribution will look like this:



What would happen if we put the drunkards on a circle and have them all start out from θ=0? Then they would diffuse in a similar manner, until eventually their density becomes constant all along the circle (the red circles show schematically the positions of the drunks at time t=1):



How long will it take for the drunks to fan out completely across the circle? Here,

$$\Delta\theta_{rms}(t)=\sqrt{2Dt}, \qquad D=\frac{(\Delta\phi)^2}{2\tau}$$

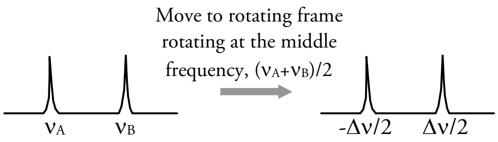
where Δϕ is the angular step size in each step. The drunks will “fan out” when

$$\Delta\theta_{rms}\sim 2\pi \qquad \Rightarrow \qquad t_{2\pi}=\frac{(2\pi)^2}{2D}=\left(\frac{2\pi}{\Delta\phi}\right)^2\tau\ .$$

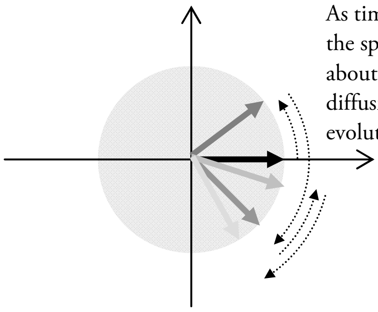
1.2 Exchange is a Random Walk

We begin by analyzing the simple pulse-acquire 1D experiment in the presence of chemical exchange. As a proton, X, "hops" between different chemical sites, its resonant frequency changes. If the hopping is much slower than T_2 then the proton will precess with a well defined frequency throughout the experiment.

With frequency jumps on the timescales of T_2 or faster, the proton will change its precession frequency throughout the evolution. Suppose, for simplicity, only 2 frequencies exist, ν_A and ν_B, that all other properties (T_1, T_2) are equal between the two sites, and that the spin "hops" back and forth between the two frequencies at a time scale τ_{ex}. This problem is easiest to analyze in a rotating frame in which the frequencies are symmetrical about zero:



where $\Delta\nu=\nu_B-\nu_A$. Considering the evolution of a spin in the transverse plane, starting out from the x-axis. At any given time it will precess with one of two possible frequencies, $\pm\Delta\nu/2$, stochastically alternating between them roughly every τ_{ex} seconds. Thus, it will accumulate a phase, ϕ(t), in analogy to a "drunk" executing a random walk:



As time advances the spins "fan out" about 0 due to their diffusion like time evolution.

The hopping of the spin between two sites can be described by a simple first order chemical exchange equation as is done in basic chemistry, with a rate constant k in Hz (k is the number of time the spin hops from A to B or from B to A in a second, assuming both transitions are equal). The time τ_{ex} is the inverse of the rate constant:

$$\tau_{ex}=\frac{1}{k}\ .$$

We now examine two limits: "fast" and "slow" exchange.

1.2 FAST EXCHANGE ($\tau_{EX} \ll \Delta\nu^{-1}$)

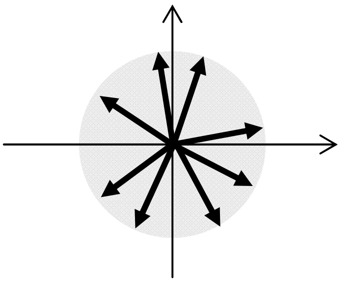
In this limit the spin precesses by a small amount before jumping (since $\Delta\nu\cdot\tau_{ex} \ll 1$). In this limit of small time steps, the spin's motion can be modeled as a circular diffusion process with diffusion coefficient $D=(\Delta\phi)^2/(2\tau_{ex})$, where $\Delta\phi$ is the typical phase the spin accumulates before flipping its precession direction. Since $\Delta\phi \sim 2\pi\cdot\Delta\nu\cdot\tau_{ex}$, we have $D=(\Delta\phi)^2/(2\tau_{ex})=(2\pi)^2\cdot\Delta\nu^2\cdot\tau_{ex}/2$. The average "distance" (in phase) the spin accumulates after a time t is then given by

$$\Delta\phi_{rms}(t) \sim \sqrt{2Dt} = \sqrt{(2\pi\cdot\Delta\nu)^2\cdot\tau_{ex}\cdot t}$$

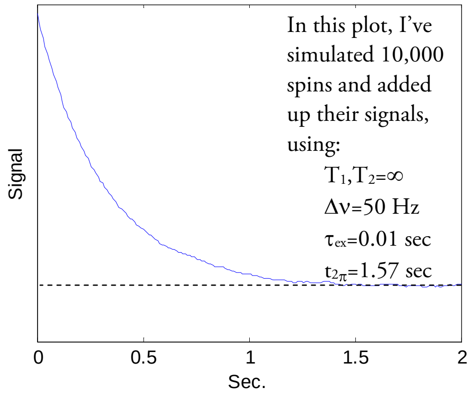
This is precisely what's described in the circular diffusion of drunkards in the previous section. A large number of spins executing this diffusion very rapidly will eventually "fan out" over the entire xy-plane, and their vector sum will add up to 0. This happens at a time $\Delta\phi_{rms}(t_{2\pi}) = 2\pi$, or (solving for $t_{2\pi}$):

$$t_{2\pi} = \frac{(2\pi)^2}{2D} = \frac{1}{\Delta\nu^2\cdot\tau_{ex}}.$$

At this point the spins will go out of phase and our signal will decay to about 0:



So, if we measure our signal as a function of time, neglecting T_2 relaxation, it will still decay due to this diffusion process:



In this plot, I've simulated 10,000 spins and added up their signals, using:

$T_1, T_2=\infty$

$\Delta\nu=50$ Hz

$\tau_{ex}=0.01$ sec

$t_{2\pi}=1.57$ sec

The resulting lineshape will have a width ~ $1/t_{2\pi}$ (up to some constant factor). Thus we obtain the following two insights:

- On average, the spins will remain at $\phi=0$, leading to an apparent frequency of zero and **a single line** at $\nu=0$, while "fanning out" and go out of phase.
- Neglecting T_2 relaxation, the width of the peak will be given by $t_{2\pi}^{-1}=\Delta\nu^2\cdot\tau_{ex}$ (in the absence of other relaxation mechanisms). As the hopping becomes more frequent, $\tau_{ex}\rightarrow 0$, the linewidth will become narrower. This is sometimes termed **motional narrowing**.

Motional narrowing associated with exchange of course can't make our lines infinitely narrow. At some point as $\tau_{ex}\rightarrow 0$ we're going to hit a "brick wall" because of the microscopic T_2 processes occurring at each site, a limit known as, the **very fast exchange** limit. What will our "ultimate linewidth" become? If we assume two sites,

$$1 \underset{k_{21}}{\overset{k_{12}}{\rightleftarrows}} 2,$$

then the spins' relative populations will be $p_1=k_{21}/(k_{12}+k_{21})$ in site 1 and $p_2=k_{12}/(k_{12}+k_{21})$ in site 2. Hence its transverse relaxation will be the average of both, $R_2 = p_1R_2^{(\text{site 1})} + p_2R_2^{(\text{site 2})}$. This will be the "intrinsic" linewidth in the limit of extremely fast exchange, when motional narrowing becomes negligible. Our *effective* T_2 that will determine our line's width will be:

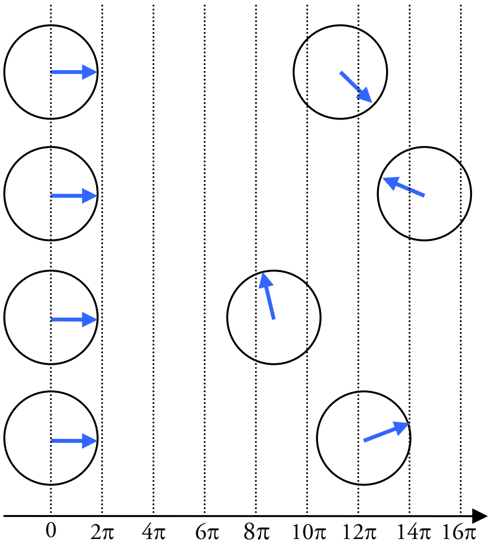
$$\frac{1}{T_2^{eff}} = \frac{p_1}{T_2^{(1)}} + \frac{p_2}{T_2^{(2)}} + \frac{1}{t_{2\pi}} = \frac{p_1}{T_2^{(1)}} + \frac{p_2}{T_2^{(2)}} + (\Delta\nu)^2\,\tau_{ex}.$$

1.3 Slow Exchange (τ_{ex}>>$\Delta\nu^{-1}$)

When the spins precess >> 2π before hopping to a different frequency - that is, when $\Delta\nu\cdot\tau_{ex}$>>2π - a different situation emerges. Imagine for a moment that all spins begin at frequency ν_A; that is, at -$\Delta\nu/2$. They will all precess coherently as a unit and, around τ_{ex}, jump over to +$\Delta\nu/2$. However, **by the time they jump, their relative phases will be completely non-coherent** since the can be anywhere in the transverse plane when the jump will occur. *Even when they hop back to -$\Delta\nu/2$, they will still be out of phase.* This means that the entire signal will decay after about a time τ_{ex} and **never return**. Thus we can summarize:

- We will see the spins precess at -$\Delta\nu/2$.
- After τ_{ex} the signal will decay to zero, leading a linewidth of ~ $1/\tau_{ex}$.

Thus we will observe a single line at -$\Delta\nu/2$ with linewidth $1/\tau_{ex}$ (neglect once again all other relaxation mechanisms). Of course, we have an equal population starting out at +$\Delta\nu/2$ and decaying the same way, so overall we will end up seeing two distinct lines at ±$\Delta\nu/2$ with linewidths governed by $1/\tau_{ex}$.



Accumulated phase ∝ time before jump

If we now add T_2 relaxation then our linewidth becomes:

$$\frac{1}{T_2^{eff}}=\frac{1}{T_2}+\frac{1}{\tau_{ex}}.$$

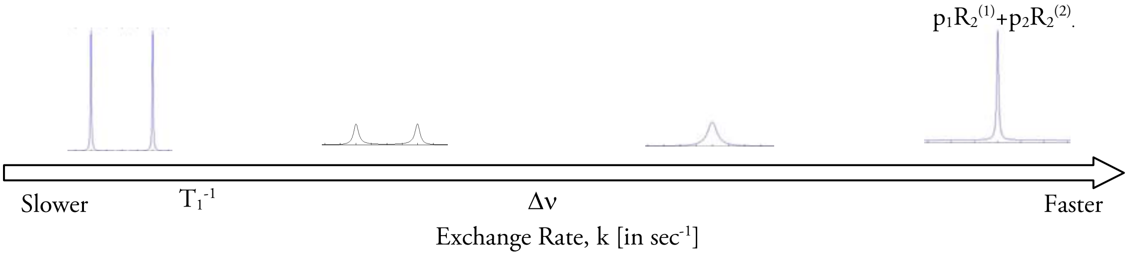
Summary: Exchange between two equally populated sites with equal bidirectional rate constants

Exchange has no effect

Slow Exchange: Two wide peaks, with width ~ $1/T_2 + 1/\tau_{ex}$.

Fast Exchange: Single broad peak, with width ~ $\Delta\nu^2\tau_{ex}$.

Very Fast Exchange: Single sharp peak, with width ~ $p_1R_2^{(1)}+p_2R_2^{(2)}$.



1.4 Unequal Size Peaks

So far we've assumed the two pools of spins have equal concentrations and rate constants. It makes absolutely no difference if the magnetization of (A) and (B) differ initially (for example, if one peak is intense and the other isn't): each peak can be thought of independently. As spins starting out at (A) hop to (B) and back to (A) and so forth, they will perform the same kind of diffusion processes described above.

What **do** matter are the rate constants and the **concentrations**. The concentration has two effects. Think of a dilute solute in a solvent. The high solvent concentration will lead to a large solvent peak, which we just argued is meaningless. However, the low solute concentration means that once a proton jumps from the solute to the solvent, it's probably not going to see another solute molecule before decaying back to thermal equilibrium, meaning the *effective rate constant* from solvent to solute is almost zero. The effective rates k_{12}, k_{21} in the $A \underset{k_{BA}}{\overset{k_{AB}}{\rightleftarrows}} B$ system are therefore proportional to not just the true rates, but also to the corresponding fractional concentrations:

$$k_{AB}^{(\mathrm{eff})} = k_{AB} \cdot \tfrac{C_B}{C_A + C_B}$$
$$k_{BA}^{(\mathrm{eff})} = k_{BA} \cdot \tfrac{C_A}{C_A + C_B}$$

This brings us to the problem of ***uneven rate constants***. These effects can be incorporated into our toy model by assuming the time a spin spends at a site is proportional 1/(rate constant). We will not repeat the math, and merely quote the changes:

1. In the fast exchange limit, the coalesced peak will resonate at the average frequency $p_A\nu_A + p_B\nu_B$, and have an average linewidth given by $p_A R_2^{(A)} + p_B R_2^{(B)}$, where now

$$p_A = \frac{k_{AB}^{(\mathrm{eff})}}{k_{AB}^{(eff)} + k_{BA}^{(eff)}} = \frac{k_{AB} C_B}{k_{AB} C_B + k_{BA} C_A}$$
$$p_B = \frac{k_{BA}^{(\mathrm{eff})}}{k_{AB}^{(eff)} + k_{BA}^{(eff)}} = \frac{k_{BA} C_A}{k_{AB} C_B + k_{BA} C_A}$$

2. In the slow exchange limit, the peaks will resonate at their respective frequencies. Broadening will occur as before, since in the slow exchange case once a spin "hops" it loses its phase coherence irrespective of the size of the pool it hops to. Thus, the exchange broadening will be $\sim 1/\tau_{ex}$ as before, and will be appreciable only if it is wider than T_2 for both sites. That is, the linewidth will be given by $\sim 1/T_2 + 1/\tau_{ex}$. Since the exchange rate A→B and B→A are different, we should write that the broadening for each site should be:

$$\tfrac{1}{T_2^{(A)}} \to \tfrac{1}{T_2^{(A)}} + k_{AB}^{(\mathrm{eff})}$$
$$\tfrac{1}{T_2^{(B)}} \to \tfrac{1}{T_2^{(B)}} + k_{BA}^{(\mathrm{eff})}$$

Of course one could have a "fast" coefficient in one direction and a "slow" one in the other direction. What do you think would happen then?

1.5 Modified Bloch Equations

Exchange can be described by incorporating exchange terms in the Bloch equation. We are aided by the fact that when a nucleus “jumps” to another molecule, it retains its magnetization.

A simple first order exchange system is:

$$\frac{dA}{dt} = -k_{AB}^{(\mathrm{eff})} A + k_{BA}^{(\mathrm{eff})} B$$
$$\frac{dB}{dt} = k_{AB}^{(\mathrm{eff})} A - k_{BA}^{(\mathrm{eff})} B$$

If we have two magnetic sites without exchange, each would simply evolve with its own Bloch equation:

$$\frac{d\mathbf{M}_A}{dt} = \gamma \mathbf{M} \times \mathbf{B}^{(A)}(t) + (\mathrm{relax})$$
$$\frac{d\mathbf{M}_B}{dt} = \gamma \mathbf{M} \times \mathbf{B}^{(B)}(t) + (\mathrm{relax})$$

Adding exchange now is a simple matter of adding the relevant exchange terms, since the two phenomena (Larmor precession and exchange) do not interfere with each other (i.e. there is no “cross-talk” term to worry about):

$$\frac{d\mathbf{M}_A}{dt} = \gamma\mathbf{M}\times\mathbf{B}^{(A)}(t) + (\text{relax}) - k_{AB}^{(\text{eff})}\mathbf{M}_A + k_{BA}^{(\text{eff})}\mathbf{M}_B$$
$$\frac{d\mathbf{M}_B}{dt} = \gamma\mathbf{M}\times\mathbf{B}^{(B)}(t) + (\text{relax}) + k_{AB}^{(\text{eff})}\mathbf{M}_A - k_{BA}^{(\text{eff})}\mathbf{M}_B$$

We will not solve these equations here since we've already obtained useful insight by considering a toy model, but the above equations provide a more rigorous framework for treating exchange for those interested.

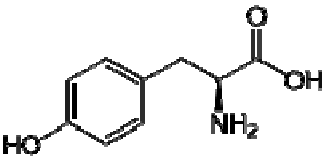
2. UNDERSTANDING EXCHANGE PHENOMENA

2.1 SOLVENT EXCHANGE

Some NMR peaks tend to disappear depending on the solvent used. For example, hydrogens in OH and NH bonds tend to exchange with hydrogens in water. The following data refer to some amino acids at a pH of 7.0 and a temperature of 37° C:

Tyrosine OH	5-15 kHz
Arginine NH	1.2 kHz
Arginine NH_2	0.7 kHz
Histidine NH	1.7 kHz

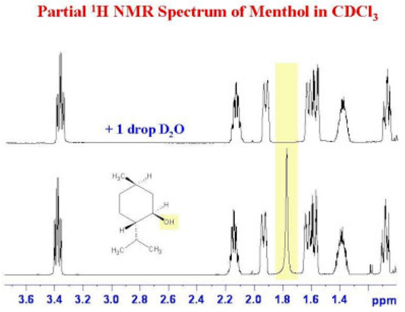
Water resonates at 4.7 ppm, while the phenolic OH protons of Tyrosine resonate much higher at around 9 ppm:



At 500 MHz, the difference between the two is about 2 kHz, implying the exchange is fast. Thus, if put in H_2O the OH and H_2O peaks will coalesce into one peak. Since H_2O is so much more abundant, the coalesced peak will appear pretty much at 4.7 ppm, meaning the OH will "disappear" and only the water will show up.

2.2 D/H STRUCTURAL INFORMATION

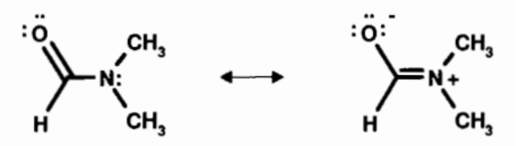
A nifty experiment called the "D_2O shake" goes like this: first, run an experiment in a regular, non-exchanging solvent such as $CDCl_3$. Add a drop of D_2O, shake, and let the D_2O molecules separate and form their own phase at the top outside the receiver coil. What will happen is that all of the OH molecules will exchange with the D_2O and then coalesce outside the sample, in effect identifying all OH (and other quickly exchanging) resonances. The following example shows menthol with (top) and without (bottom) the extra D_2O:


Partial ^1H NMR Spectrum of Menthol in $CDCl_3$

If the exchange is very slow, the exchange rates can be quantified by recording a series of 1D spectra and fitting the peaks with a decaying exponential as a function of time over several seconds/minutes/hours. For example, when studying proteins in H_2O, some D_2O can be added to test tube. The amide (NH) protons in the protein's backbone will then exchange with the D_2O and their magnitude as a function of time will yield their exchange constants. This also lets one obtain crude information about protein structure, since amide protons which are closed off from the solvent due to the protein's folding will not exchange or exchange very slowly. Sometimes 2D HSQC and not 1D spectra are recorded, in which case the exchange must be very, very slow (slower than the time it takes to run a 2D experiment) for us to use this method.

2.3 MOLECULAR INTERNAL ROTATIONS

Let's look at a simple molecule, N,N-dimethylformamide, at room temperature. The molecule displays a resonance by the electron shared between the NCO group:



There is no internal rotation of the molecule at room temperature, since molecules don't tend to internally rotate about double bonds, or do so very slowly. Each of the methyl groups appears at a distinct resonance in the ^{1}H NMR spectrum. As temperature is increased, however, the two methyls rotate about the C=N bond faster and faster, and at 130° C the two methyl resonance coalesce into a single line.

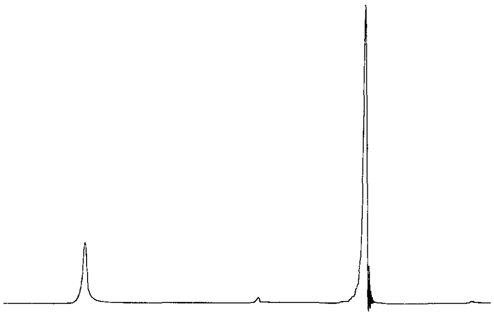
2.4 Decoupling

Even decoupling can be (partially) understood in terms of an exchange. As we irradiate the carbon in a C-H pair, we're rotating C and changing the field H feels. Here, H "jumps" from one frequency ($\nu+J/2$) when C points in one direction, to another ($\nu-J/2$) when C points in a different direction. Think of the Hamiltonian of a two-spin system:

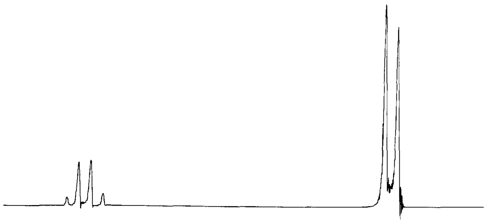
$$H = \omega_1 I_{1z} + \omega_2 I_{2z} + 2\pi J I_{1z} I_{2z} .$$

If we think of I_{1z} semiclassically as a "coefficient" of I_{2z} and modulate it as a function of time, it is similar to the chemical exchange problem. If we pulse on the C at a rate $\tau^{-1}>>J$, we can average out the effects of J-coupling, where τ is the spatial spacing between the π-pulses.

An interesting application of this can be seen in methanol (CH_3OH). In the presence of impurities or at 50°C temperature, methanol contains two peaks:



This is due to the H in the OH detaching and re-attaching to the molecule. At low enough temperatures (-30°C) or pure enough methanol samples, however, one observes:



As one would expect, the H is split thrice into a quartet and the methyl (CH_3) is split once by the OH due to J-coupling (they are an AX_3) system. What is happening here? As the H in OH detaches and re-attaches fast enough, it "scrambles" the phase of the J evolution.

2.5 Why Don't We See Non-Isotropic Interactions in The Liquid State?

Why are liquid state spectra so simple? Because many interactions get averaged out. This vague statement can now be made more exact: as the molecule changes its rotational state many non-isotropic interactions will cause the nuclear spins' larmor frequency to change as a function of time. If the rotational correlation time, τ_{rot}, is shorter than the range of frequencies of the interaction (i.e., the "size" of the interaction), then the interaction will get averaged out.

For example, consider dipolar coupling between two spins ~ 1 nm. The dipolar field of one spin at the position of the other is given by:

$$\mathbf{B}(\mathbf{r}) = \frac{\mu_0}{4\pi}\frac{3(\mathbf{m}\cdot\mathbf{r})\mathbf{r}-\mathbf{m}r^2}{r^5}.$$

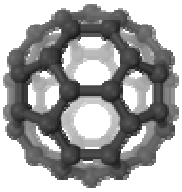
The interaction is maximal on the axis, where, for a proton magnetic moment,

$$\mathbf{B} = \frac{m\mu_0}{2\pi r^3} \sim 10^{-5}\ \mathrm{T}.$$

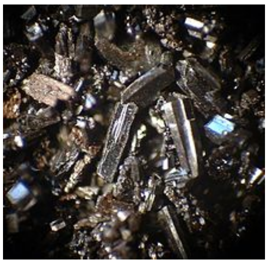
This translates to a frequency shift of about $\gamma B \sim 1$ kHz for protons. Of course, since $\mathbf{B} \sim r^{-3}$, this can get much larger (~10 kHz) for closer proton spin pairs. The dipolar interaction will assume many values as the molecule rotates, but its magnitude will be insignificant when compared to the *frequency* at which the molecule rotates. The rotational correlation time for small molecules in liquids is about 1 ps in liquids, and its reciprocal is 10^{12} Hz, much much larger than the dipolar interaction. We are thus in the "fast exchange" limit in which all possible frequencies coalesce into a single frequency, which we observe in liquid state NMR.

2.6 A "DISORDERED SOLID"

A C_{60} molecule is a ball made out of 60 carbon atoms:

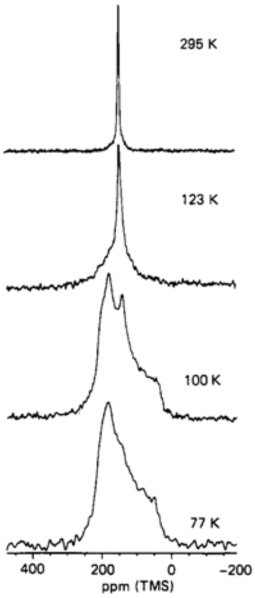


These molecules, also called Buckminsterfullerene (after the inventor who built the first geodesic dome, which assumed a similar shape), have many interesting applications in chemistry. Solids of C_{60} molecules can be created (this is a famous 1990 Nature paper), in which the molecules assume a somewhat disordered close packing:



Solid C_{60} at room temperature.

One can measure a ^{13}C spectrum of these solids at different temperatures:



From: *Yannoni et. al.*
J. Phys. Chem. 95:9-10 (1991)

We see that at room temperature we get a sharp line, giving us a strong indication that the molecule is rotating isotropically and rapidly (faster than the solid state dipolar interactions and mostly the chemical shift anisotropy). As temperature drops the molecular rotational motion slows down and we start seeing solid effects of chemical shift anisotropy.