8.1 Relaxation in NMR Spectroscopy

An understanding of relaxation processes is important for the proper measurement and interpretation of NMR spectra. There are three important considerations.

1. The very small energy difference between $\alpha$ and $\beta$ states of a nuclear spin orientation in a magnetic field results in a very small excess population of nuclei in the ground vs the excited states. For many nuclei relaxation (i.e., return from excited to ground state) is a very slow process, with half-lives on the order of 0.1 to 100 seconds for a spin $\frac{1}{2}$ nucleus (compare this with micro-, pico- and femtoseconds for relaxation of electronic and vibrational transitions). It is thus very easy to saturate an NMR transition (equalize populations of excited and ground state), with the resultant loss in signal quality, and failure to obtain correct peak areas.

2. NMR lines are extraordinarily sharp, and extraordinarily close together (in energetic terms) compared to other spectroscopic methods. So much so that Heisenberg uncertainty broadening (which is a function of lifetime of a given energy state, and hence relaxation rates) is a dominant feature of many NMR spectra, and can limit our ability to measure and interpret spectra. When relaxation is very fast, NMR lines are broad, $J$-coupling may not be resolved or the signal may even be difficult or impossible to detect.

3. The success of many multipulse experiments, especially 2D and 3D spectra, depends crucially on proper consideration of relaxation times.

$T_1$ and $T_2$ Relaxation. We distinguish two types of relaxation, Spin-Lattice ($T_1$, also known as longitudinal relaxation, or relaxation in the z-direction) and Spin-Spin ($T_2$, also known as transverse relaxation, or relaxation in the x-y plane). $T_1$ relaxation corresponds to the process of establishing (or re-establishing) the normal Gaussian population distribution of $\alpha$ and $\beta$ spin states in the magnetic field. $T_2$ is loss of phase coherence among nuclei. $T_2$ is less than or equal to $T_1$ ($T_2$ relaxation is the same as or faster than $T_1$ relaxation, $R$ = relaxation rate, $R_2 = 1/T_2$, $R_2 \geq R_1$), since return of magnetization to the z-direction inherently causes loss of magnetization in the x-y plane. The line width of an NMR signal is determined by $T_2$ (short $T_2$ means broader lines, $\nu_{1/2} = 1/\pi T_2$, $\nu_{1/2}$ = width at half height), the maximum repetition rate during acquisition of an NMR signal is governed by $T_1$ (short $T_1$ means signal can be acquired faster).
8.1 Sources of Line Broadening in NMR

1. Instrumental problems - tuning, etc

2. Sample problems:
   - Sample inhomogeneity (poor mixing)
   - Temperature gradients across sample
   - Paramagnetic impurities

3. $T_1$ Relaxation (Spin-Lattice Relaxation): gain and loss of magnetization in the z-direction

Heisenberg Uncertainty Principle broadening due to inherent lifetime of spin states

$$\Delta E \cdot \Delta t = \frac{\hbar}{2\pi}$$

$$h\delta \nu \cdot \delta t = \frac{\hbar}{2\pi}$$

$$\nu_\frac{1}{2} = \frac{1}{2\pi T_1} \quad \text{(Half-width at half height)}$$

$$\nu_\frac{\pi}{4} = \frac{1}{\pi T_1} \quad \text{(Width at half height)}$$

Broadening due to paramagnetic impurities is a special case of $T_1$ broadening

Place unmagnetized sample in magnetic field: establishment of the normal Boltzmann equilibrium between the $\alpha$ and $\beta$ spin states has a first order rate constant of $k = 1/T_1$: $M_z = M_0 (e^{-t/T_1})$

Or apply a 90° pulse, and watch the magnetization return to the z-direction: $M_z = M_\infty (1 - e^{-t/T_1})$

The rate of spontaneous relaxation of nuclear spin orientations is almost zero. $T_1$ relaxation is caused by transient magnetic fields (usually due to molecular motion) at the Larmour precession frequency.

In most situations $T_1$ relaxation is optimal if the average rate of molecular motion is at the Larmour precession frequency

$\tau = \text{Molecular correlation time: the time it takes the average molecule to rotate one radian}$
4. $T_2$ Relaxation (Spin-Spin Relaxation):
Heisenberg Uncertainty Principle broadening due to lifetime of spin coherence - gain and loss of magnetization in the x,y-direction

$$\nu_{1/2} = \frac{1}{\pi T_2} \quad T_2 \leq T_1$$

For protons, $T_2$ is usually between 1 and 10 seconds

- $T_2 = 1$ sec, $\nu_{1/2} = \frac{1}{\pi} = 0.3$ Hz
- $T_2 = 10$ sec, $\nu_{1/2} = \frac{1}{10\pi} = 0.03$ Hz

$T_2$ relaxation is caused by transient magnetic fields (usually due to molecular motion) at any frequency.

$T_2$ relaxation is also caused by swapping of chemical shifts or coupling constants (chemical exchange). Line broadening due to chemical exchange provides an important tool for measurement of the rates (see Sect 8-MULTI-3).
Spin ½ nuclei in magnetic field $B_0$

Top: Before the pulse we have more nuclei oriented in the up than in the down direction (net magnetization in $z$ direction)

Bottom: After the pulse we have more nuclei oriented to the right than to the left (net magnetization is now in the $x'$ direction)

Net Magnetization Vector

This magnetization is rotating at the Larmor precession frequency, and it is this oscillating magnetic field that is detected by the NMR spectrometer receiver coils.
Measurement of $T_1$ by Inversion-Recovery

Measurement of $T_1$. One of the methods for measuring $T_1$ is the $5T_1-\pi-\tau-\pi/2$-FID pulse sequence, in which a series of NMR spectra are measured in which $\tau$ is incremented, and the degree to which the signal has returned to its equilibrium value during the waiting period $\tau$ for each sequence is measured.
Relaxation Mechanisms. Spontaneous $T_1$ relaxation of spin ½ nuclei, i.e., relaxation in the absence of external influences, is essentially absent. For $T_1$ relaxation to occur there must be magnetic field fluctuations in the x,y direction. Such fluctuations are most effective when they occur at the Larmor precession frequency ($\nu_0$). $T_1$ relaxation is thus field dependent, since $\nu_0$ varies with the field.

$T_2$ relaxation is caused by fluctuations in any direction.

The principal source of fluctuating magnetic fields in most molecules is molecular motion. We can define a correlation time $\tau_c$ for a molecule (assuming it behaves more or less spherically). This is the average time it takes the molecule to rotate through one radian. The correlation time for small molecules is of the order of $10^{-12}$ sec in solution (longer in viscous solvents). Since for a proton at 300 MHz $\nu_0 \approx 10^8$, most molecules below molecular weight of 1000 are moving too rapidly for effective relaxation. There are several mechanisms by which molecular motions can influence nuclear relaxation: direct interactions with nearby magnetic nuclei (DD), chemical shift effects (CSA), quadrupole-electric field gradient interaction (QR) and rapid modulation of $J$-coupling (SC). In addition to molecular motion, rotational transitions can also be the source of fluctuating magnetic fields (SR).

Below are summarized the relaxation mechanisms.

1. Spin Rotation (SR). A local magnetic field is generated by the circular motion of electrons in a rapidly rotating molecule (or part of a molecule, such as a methyl group).

```
H
\( \text{B changes as the rate of rotation changes} \)
```

The magnitude of this field changes when the rotational energy levels change as a result of molecular collisions. These changes, if they occur at the Larmor precession frequency, can cause relaxation of nearby nuclei. The correlation time for SR relaxation is not molecular motion, but the lifetime of rotational quantum levels.

Characteristic features:

- Small molecules, or freely spinning portions of larger molecules without other efficient relaxation mechanisms.
- Better relaxation (shorter $T_1$) at higher temperatures.
- The SR mechanism has been detected for methyl groups, and for quaternary carbons of small molecules which do not have other effective relaxation mechanisms (e.g. CS$_2$).
- Operation of this mechanism causes reduction or loss of NOE effects.
2. Chemical Shift Anisotropy (CSA). The chemical shift of a nucleus is a function of the orientation of the molecule in the magnetic field (i.e. it is a tensor quantity), provided the nucleus is not at the center of tetrahedral or octahedral symmetry). For ethylene ($^{13}$C) and nitrobenzene ($^{15}$N), the three limiting chemical shifts are given below:

$$
\delta_C = \begin{cases} 
24 & \text{for } H \equiv C \equiv H \\
234 & \text{for } H \equiv C \equiv H \\
120 & \text{for } H \equiv C \equiv H 
\end{cases} 
$$

$^{13}$C chemical shifts tensors
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$$
\delta = \begin{cases} 
57 \text{ ppm} & \text{for } Ph - N - O - O \\
-179 \text{ ppm} & \text{for } Ph - N - O - O \\
126 \text{ ppm} & \text{for } Ph - N - O - O 
\end{cases} 
$$

$^{15}$N chemical shifts tensors

As the molecule tumbles in solution, the chemical shift (and hence the magnetic field at the nucleus) is constantly changing, and this can cause relaxation of the nucleus. The relaxation rate is proportional to the square of the gyromagnetic ratio and of the magnetic field strength, as well to the chemical shift anisotropy. The correlation time is molecular motion. The equation below applies to cylindrically symmetric molecules (one symmetry element):

$$
R_{1(CSA)} = \frac{1}{T_{1(CSA)}} = \frac{2}{15} \gamma^2 B_0^2 (\sigma_x - \sigma_y)^2 \tau_c
$$

Chemical shift anisotropy
(if symmetrical, no CSA relaxation)

- Magnetic field
- Magnetogyric ratio
- Correlation time
- usually molecular motion

Characteristic features:

- The rate of relaxation is proportional to the square of the field strength, and thus becomes much more important at high field strengths. For some spin ½ nuclei with large chemical shift ranges, lines become sufficiently broadened by CSA relaxation at high field to cause loss of coupling information.

- Generally dominant in nuclei with large chemical shift ranges, and in bonding situations which produce large magnetic anisotropies. Molecules in which the nucleus is at the center of tetrahedral, octahedral, or higher symmetry ($^{119}$Sn in Me$_4$Sn. $^{29}$Si in Me$_3$Si) have no chemical shift anisotropy and so do not relax by this mechanism.

- The mechanism is never seen for protons, and is seen for carbon only when there are no attached protons (e.g., carbonyl compounds other than aldehydes and formates), but nuclei such as $^{199}$Hg, $^{119}$Sn and $^{77}$Se with large chemical shift ranges are relaxed largely or entirely by the CSA mechanism.

- Operation of this mechanism causes reduction or loss of NOE effects.
3. Quadrupolar Relaxation (QR). This mechanism operates for spin >½ nuclei only, and only for nuclei which are not at the center of tetrahedral or octahedral symmetry. Spin ½ nuclei can be considered to have spherical charge distributions, but for spin >½ nuclei the charge distribution has the shape of an oblate or prolate spheroid. Electric field gradients in such molecules exert a torque on the quadrupolar nuclei. Tumbling of the molecule can then initiate transitions among the spin states (there is "friction" between the nucleus and the surrounding electrons, the "quadrupole coupling constant" \(e^2Qqz/h\). The effectiveness of this relaxation mechanism is critically dependent on this coupling. If \(Q\) is small (as for \(^2\text{H}\) and \(^6\text{Li}\)) the nucleus behaves like a spin ½ nucleus, if it is large the nucleus can have very short \(T_1\), and observation can be very difficult.

\[
R_{QR} = \frac{1}{T_1(QR)} = \frac{3}{10} \frac{\nu^2}{\eta^2} \frac{2I + 3}{(2I - 1)} \left(1 + \frac{\eta^2}{3}\right) \left(\frac{e^2Qq_z}{h}\right)^2 \tau_c
\]

\(\nu = (q_{yy} - q_{xx})/q_{zz}\) (asymmetry parameter)

\(q\) = electric field gradient

\(Q\) = electric quadrupole moment

\(\tau_c\) = molecular correlation time (molecular or segmental rotation)

\(I\) = nuclear spin

In molecules with tetrahedral or octahedral symmetry the electric field gradient is small or zero, and \(T_1\) values can be long enough to obtain acceptable spectra (e.g., \(\text{ClO}_4^-\) for \(^35\text{Cl}\), \(\text{SO}_4^{2-}\) or \(\text{R}_2\text{SO}_2\) for \(^33\text{S}\)).

\(^{11}\text{B} \quad I = 3/2 \quad Q = 0.036\)

\(\text{Na}^+ \text{BH}_4^- \quad T_1 = 11\) sec

\(\text{BET}_3 \quad T_1 = 0.01\) sec

\(^{14}\text{N} \quad I = 1 \quad Q = 0.016\)

\(\text{NH}_4^+ \text{Cl}^- \) in \(\text{H}_2\text{O} \quad T_1 = 50\) sec

\(\text{CH}_3-\text{C}≡\text{N} \) in \(\text{H}_2\text{O} \quad T_1 = 0.022\) sec

However when \(Q\) is large enough, even nuclei in a formally symmetric environment can show fast QR. For example the spherically symmetrical iodide ion \((I = 5/2)\) has such a large quadrupole moment \((Q = 0.79)\) that line widths of from 1200 to >50,000 Hz are seen in various solvents due to transient asymmetric solvation.

Quadrupolar relaxation of a nucleus can also have effects on nearby magnetic nuclei, since rapid relaxation can either broaden or entirely remove \(J\)-coupling between the two nuclei. This effect is especially common for \(^{14}\text{N}-\text{X}\) and \(^{11}\text{B}-\text{X}\) groups. Such broadening is actually a \(T_2\) effect at the \(X\) nucleus (interchange of spin states). It is often difficult to observe carbons or protons directly bonded to boron because of this kind of \(T_2\) broadening. Rapid quadrupolar relaxation of Cl, Br and I nuclei is the reason we don't normally see any indication of coupling between these nuclei and adjacent carbons or protons (self-decoupling).

**Characteristic features:**

- only for nuclei with \(I > 1/2\).
- faster relaxation at lower temperatures in solution.
- there must be a permanent (or transient) electric field gradient across the nucleus (i.e., QR is much less effective in molecules in which the nucleus is at a center of tetrahedral or octahedral symmetry).
- for nuclei with a large electric quadrupole moment, this mechanism is so effective that only kHz wide lines can be observed, and no \(J\) coupling to other nuclei can be detected.
4. **Scalar Relaxation.** Scalar ($J$) coupling of a nucleus $Y$ to a second quadrupolar nucleus $X$ can provide a relaxation mechanism for $Y$ if $X$ is undergoing very rapid $T_1$ relaxation. Under these conditions $Y$ is subject to a fluctuating magnetic field because of the rapid spin reorientation of $X$. For this mechanism to be very effective, the Larmor precession frequencies of $X$ and $Y$ must be very close together (i.e., $\omega_X - \omega_Y$ must be small).

$$R_{1(SC)} = \frac{1}{T_{1(SC)}} = \frac{8\pi^2 J^2}{3} \frac{\tau_{SC}}{1 + (\omega_X - \omega_Y)^2} \frac{l(l + 1)}{\tau_{SC}^2}$$

- $R_{SC}$ = Scalar relaxation rate of $Y$ (spin 1/2) caused by $X$ (quadrupolar nucleus)
- $J$ = coupling constant between $X$ and $Y$
- $\tau_{SC}$ = $T_{1(QR)}$ (1/rate of quadrupolar relaxation of $X$)
- $\omega_X, \omega_Y$ = Larmor precession frequencies of $X$ and $Y$
- $l$ = spin of $X$

The unusually short $T_1$ values for bromine-bearing carbons are believed to be caused by SC relaxation. On a 200 MHz spectrometer, the Larmor precession frequency of $^{13}\text{C}$ is 50.28 MHz, that of $^{79}\text{Br}$ ($I = 3/2$), 50.18 MHz.

- $T_1$ of CHCl$_3$ = 32.4 sec
- $T_1$ of CHBr$_3$ = 1.65 sec

Note that the SC $T_1$ relaxation process is quite distinct from the broadening of $Y$ caused by partially coalesced $J$ coupling between $Y$ and $X$ that occurs when $T_{1(X)}$ has values in the range of $1/\langle \pi J_{C-X} \rangle$ (as is often seen for spin $1/2$ nuclei attached to $^{14}\text{N}$ and $^{11}\text{B}$). Such broadening is a $T_2$ process in $Y$ (swapping of spin states).
5. Dipole-Dipole Relaxation (DD). Coupling between magnetic nuclei is of two types: the scalar ($J$) coupling is the result of polarization of the electrons by the nuclear spins. The direct dipole-dipole coupling interaction is very large (often kilohertz) and depends principally on the distance between nuclei and the angular relationship between the magnetic field and the internuclear vectors. This coupling is not seen in mobile solutions because it is averaged to zero by tumbling of the molecule. However, as the molecule tumbles in solution the dipole-dipole coupling is constantly changing as the vector relationships change. This creates a fluctuating magnetic field at each nucleus. To the extent that these fluctuations occur at the Larmor precession frequency, they can cause nuclear relaxation. Since the proton has the highest magnetic dipole of common nuclei, it is the most effective nucleus for causing DD relaxation. DD relaxation is the principal relaxation pathway for protons in molecules containing contiguous protons, and for carbons with directly attached protons.

The correlation time for DD relaxation is molecular (or, more accurately, segmental) motion. The equation governing DD relaxation of an X nucleus by nearby protons in the region of fast molecular motion (extreme narrowing region):

$$R_{1(DD)} = \frac{1}{T_{1(DD)}} = \gamma_X^2 \gamma_H^2 \left( \frac{h}{2 \pi} \right)^2 \tau_c \sum \frac{1}{r_{HX}^6}$$

$\tau_c$ the correlation time (average time for the H-X vector to rotate 1 radian)
$\gamma_X, \gamma_H$ gyromagnetic ratios
$r_{HX}$ distance between H and the X nucleus being relaxed.

DD relaxation shows a very strong distance dependence, and operates most effectively between directly bonded nuclei. At the usual spectrometer frequencies, small molecules (MW <1000) are tumbling too fast for the most effective relaxation ($\tau_c$ is too short). Thus the more rapidly a molecule or part of a molecule tumbles, the less effective DD relaxation is, and the longer $T_1$ becomes. Large molecules (e.g. proteins) are usually moving too slowly ($\tau_c$ is too long), and they have the opposite relationship between molecular motion and $T_1$ (i.e., relaxation is more effective when the molecule moves faster).

For $^{13}$C NMR, dipole relaxation by directly attached protons (if any are present) is the principal relaxation mechanism. Thus quaternary carbons will have long relaxation times (hence their low intensity under normal conditions of spectrum acquisition), CH groups will have shorter, and CH$_2$ groups still shorter, by approximately a factor of two. CH$_3$ groups show highly variable $T_1$ values. They usually move much more rapidly than the rest of the molecule, and thus undergo less effective DD relaxation, but this may be complicated by SR relaxation.

Anisotropic Motion: Long thin molecules do not move isotropically in solution (i.e., $\tau_c$ will be different for rotation around different axes of the molecule), so that relaxation times can be quite different for carbons which are on the long axis, compared to those off it. In the example of diphenyldiacetylene, the para carbons have relatively short relaxation time (1.1 s) whereas for the ortho and meta carbons $T_1$ is about five times as long. This is because rotation around the long axis is very fast, and causes inefficient relaxation for the off-axis carbons. The para carbons change their relationship with the attached hydrogen only when rotating around an axis perpendicular to the acetylenes, a much slower process. This effect can sometimes be used to assign carbon signals.
**Segmental Motion:** Different parts of molecules can have different correlation times, and consequently $T_1$ values. For 1-bromodecane, the $T_1$ values are uniform down the chain except near the ends, where local conformational motions are somewhat faster. On the other hand, in 1-decanol the $T_1$ values become progressively shorter closer to the OH group, probably a consequence of “anchoring” of the OH end by intermolecular hydrogen bonding. You can also see the effects of segmental motion in the cholesteryl chloride $T_{1}$ values - *all of ring CH are ca 0.5 sec, the CH$_2$ 0.25 sec - that part of the molecule moves as a unit.* However, *in the side chain the $T_1$ values become longer, up to 0.67 sec for the last CH$_2$ and 1.8 sec for the last CH, because the side chain has additional flexibility, and thus a shorter correlation time.*

![Effect of DD relaxation by oxygen](image1)

**Effect of DD relaxation by oxygen**

Oxygen is paramagnetic and is capable of causing proton and carbon relaxation by a dipolar interaction with the unpaired spins on O$_2$. The effect is not large, but can be easily seen for the quaternary carbons of phenylacetylene - e.g. the ipso carbon $T_1$ increases from 56 to 107 sec if the dissolved oxygen is removed. This can be done by vacuum techniques, or more simply by bubbling N$_2$ or argon through the sample for a minute or two. It is important to remove dissolved oxygen from samples where long $T_1$ values are important, or where proton-proton DD relaxation must be maximized (e.g. in the NOE experiment discussed in Section 8.2).

![Effect of Segmental motion](image2)