Two-dimensional NMR spectroscopy was first proposed in 1971, but its full power was not realized until 1980.

2D pulse sequences consist of a preparation period (frequently a 90° pulse), an evolution period ($t_1$) during which some modulation occurs due to the interaction of interest, followed by a detection period ($t_2$).

A series of FIDs are recorded while the value of $t_1$ is incremented in a regular manner from one experiment to another (i.e., FIDs $Z(t_1,t_2)$ are acquired). Fourier transformation of the $t_2$ time domain gives us a set of spectra as a function of the $t_1$ step size. A second Fourier transformation of $t_1$ time domain gives a 2D spectrum revealing the frequencies of the modulation that were occurring during the $t_1$ evolution period.

2D NMR spectroscopy can be used to probe specific NMR interactions selectively. Spreading out the spectrum in two dimensions has the advantage for large molecules that it removes much peak overlap. While the simple vector model may be used to explain some 2D experiments, it is unable to explain 2D techniques.

**NOESY**

One of the simplest 2D experiments

Can be “pictured” in terms of the vector model.
The amplitude of the signal in the $Z(t_1, F_2)$ spectra (after the first Fourier transformation of $Z(t_1, t_2)$) varies regularly as a cosine function of $t_1$. Why?

Consider a two-spin system $I,S$ (both spin-1/2), $J_{IS} = 0$.

The experiment is designed to record only the $z$ components of the magnetization evolved during the mixing period $\tau_m$. The $x$ component is rejected using a suitable phase-cycling scheme. A third $90^\circ_x$ pulse converts $z$-components into transverse signal for detection. The $z$ components are said to be frequency labelled.
\( \tau_m = 0 \): a second FT with respect to \( t_1 \) extracts the frequency of the amplitude modulation of \( Z(t_1, F_2) \):

\[ \omega \]

\( \tau_m \neq 0 \): useful information can be obtained if the two spins exchange magnetization during \( \tau_m \), either by cross-relaxation or by chemical exchange.

When \( t_1 \) is such that \( \omega_1 t_1 = \pi/2 \), the \( z \) component of \( I \) has zero intensity at the start of \( \tau_m \), while the corresponding \( S \) vector is still largely inverted.

This is similar to the situation at the start of the \( \tau \) in a normal 1D transient NOE experiment (one spin is completely inverted, while the other is at equilibrium). Cross-relaxation during \( \tau_m \) results in a transient NOE enhancement at \( I \), similar to that expected in the 1D experiment. The size and direction of these enhancements depend on the difference in the values of the \( z \) components of \( I \) and \( S \) at the start of \( \tau_m \). These values in turn depend on the extent of precession that \( I \) and \( S \) underwent during \( t_1 \).
By the end of the mixing time $\tau_m$, the intensity of the $I$ vector, which was initially given by $-\cos \omega_I t_1$, acquires an additional dependence on $\cos \omega_S t_1$. This results in a major peak (diagonal peak) at $F_1 = \omega_I$ and a minor peak (cross-peak) at $F_1 = \omega_S$ on Fourier transformation with respect to $t_1$. The presence of cross-peaks in NOESY spectra indicates that the proton spin sites are close in space ($< 5$ Å).

Other 2D Spectra

COSY - Homonuclear Correlation Spectroscopy is based on the pulse sequence:

$90^\circ_x - t_1 - 90^\circ_x -$ acquire ($t_2$)

Double Fourier transformation gives a 2D spectrum with the normal one-dimensional spectrum along the diagonal, while cross-peaks indicate $J$-couplings between nuclei.

There are many variants of the COSY sequence. For instance, the pulse sequence can be modified to emphasize long-range $J$-couplings. Identifying cross-peaks close to the diagonal can be a major problem, but methods exist to reduce (or eliminate) diagonal peak intensity (e.g. Double-Quantum Filtered COSY).
HETCOR - HETeronuclear CORrelation coupling experiments are also straightforward. In the case of $^{13}$C/$^1$H correlation, the cross-sections through the two axes reveal the normal one-dimensional $^{13}$C and $^1$H spectra, while the 2D peaks reveal which carbons are bonded to which hydrogens. This simplifies considerably assignment of peaks.

\[
\begin{array}{c}
\text{CH}_3\text{-CH}_2\text{-CH(OH)}\text{-CH}_3 \\
\end{array}
\]

INADEQUATE - correlates through the coupling between dilute nuclei. This enables, for example, the entire carbon skeleton to be determined in those cases where $^1$H and $^{13}$C-$^1$H $J$-couplings are not sufficient to solve the problem. It is a powerful technique, but time-consuming.

\[
\begin{array}{c}
\text{CH}_3\text{-CH}_2\text{-CH}_2\text{-CH}_2\text{-OH} \\
\end{array}
\]
**J-resolved Spectroscopy** - both homonuclear and heteronuclear versions exist. The pulse sequence is based on a simple spin-echo type experiment. One dimension gives the spectrum with chemical shift information only (i.e., all $J$-couplings removed), while the other dimension shows only $J$-coupling information.

**CH$_3$-CH$_2$-CH(OH)-CH$_3$**

$\delta_C / ppm$

$1_J^{CH}$

$2D$ EXSY - gives cross-peaks between sites that are in slow chemical exchange; this enables dynamic information to be obtained when in the slow-exchange limit on the chemical shift timescale.

2D $^{13}$C-$^{13}$C EXSY
Methylcyclohexane, C$_6$H$_{11}$CH$_3$,
in CDCl$_3$/CFCl$_3$ (1:3 v/v) at 200 K.

The observed cross-peaks are due to slow exchange between two chair conformers of C$_6$H$_{11}$CH$_3$ with axial (population ~1%) and equatorial (~99%) orientations of the methyl group.