

Repetition Rates in NMR: Measurement of T_1 Values Selective 1D: Tocsy1D

Use Persephone (Av-500) for this week's HW. Use the same sample as in HW#8-9.

Reading – Spin Relaxation in Claridge section 2.4 (esp. 2.4.1 & 2.4.2)
– Tocsy1d in Claridge section 5.7 (esp. 5.7.4)

Goals – Understand how to estimate/measure ^1H T_1 and utilize that for setting $d1$ / repetition rates¹ in a variety of NMR experiments.
– Utilize Tocsy-1d as an alternative to the 2D analog.

I. T_1 estimates in TopSpin

☞ Use Estimating T_1 in TopSpin in the User Guides → Avance section (item 6) as a guide for measuring $d7_{\text{null}}$ values for your sample. From these measurements, estimate:

$T_1(\text{shortest of interest})$ and $T_1(\text{longest of interest})$, where

$$T_1 = d7_{\text{null}} / \ln 2 \cong d7_{\text{null}} \times 1.4$$

1☞ Plot the two spectra that are closest to $T_1(\text{shortest of interest})$ and $T_1(\text{longest of interest})$ in a stack that includes the ^1H 1D spectrum. Upload as usual.

II. T_1 measurements using IconNMR

☞ Use Accurate T_1 measurements using IconNMR in the Avance User Guides → Avance section (item 7) as a guide for obtaining a pseudo 2D (stacked spectra) T_1 dataset for your sample.

This data can be obtained on Persephone using IconNMR *while staying in your TopSpin session*.

☞ See Running IconNMR in Individual TopSpin Sessions in the Avance User Guides → Avance section (item 14) as a guide for how to do this.

Open the spectrum in MNova, and estimate the same $T_1(\text{shortest of interest})$ and $T_1(\text{longest of interest})$ for your sample.

2☞ Plot the IconNMR data as a stack plot.² Use the Text Annotation feature to point out the key areas in the stack that you used to estimate the T_1 values. Upload as normal.

¹ $d1$ is the parameter most often used to vary the *repetition rate* (t_{rr}) of an experiment. It is important to keep in mind, however, that t_{rr} is the time in the experiment from one scan to the next. Usually $t_{rr} \approx (d1+aq)$, but it depends on the experiment. A noesy experiment, as one example, often has a mix time $\equiv d8$ that is significant: now $t_{rr} \approx (d1+aq+d8)$.

² Remember that you can phase the stack plot by choosing one spectrum (usually the first, smallest vd spectrum) and phasing it (negative). The other spectra will take this phase.

- Q1** After taking both sets of data, describe at least one advantage and one disadvantage for each method (estimating T_1 values using TopSpin vs IconNMR).

III. Tocsy1D data

Use Selective-1D experiments in the User Guides → Avance section (item 10) as a guide for obtaining a pseudo 2D (stacked spectra) T_1 dataset for your sample. Open the spectrum in MNova, and again estimate T_1 (shortest of interest) and T_1 (longest of interest) for your sample.

Selective-1D experiments can provide significant advantages to the corresponding 2D experiments: (1) they are much faster to acquire; (2) they often provide superior data quality, (e.g., unambiguous identification of much smaller NOEs); (3) they provide significantly better resolution; and (4) they often pull high-resolution multiplets cleanly out of overlapped regions of the spectrum. Most selective-1D experiments require resolved peaks/multiplets for the selection. Many small molecules fit this criterion, and noesy1d and tocsy1d experiments (in particular) have become mainstays of research performed in the Chemistry NMR facility, due to the advantages stated above.

- Acquire TOCSY-1D spectra of two (or more) multiplets of interest in your compound.³ Use the 2D TOCSY or COSY spectra acquired earlier as guides if you wish.

– In tocsy1d, $d1 = 1.5 \times T_1(\text{longest of interest})$ is sufficient, and

$d9[\text{mix}] = 0$	should show <i>only</i> the selected multiple inverted
$= 0.015 \text{ s}$	gives cosy-like spectra (2- and 3- bond connectivity only)
$= 0.03 \text{ s}$	is a common small value (2,3-bond connections)
$= 0.05 \text{ s}$	good intermediate value
$= 0.08 \text{ s}$	is a common long value (full connectivity through spin system)
$= 0.12 \text{ s}$	is a common longest value (if 80ms proves insufficient)
$= 0.20 \text{ s}$	is the maximum value that should every be used for the spin-lock

Acquire each TOCSY1D set with ≥ 4 mix times (thus, ≥ 8 spectra for the two multiplets).

For TOCSY1D, always acquire the $d9=0$ tocsy1d spectrum. It is very hard to insure that the selective pulse has been setup correctly without doing this initial experiment.

- Publication of TOCSY (and ROESY) data should include the “spin-lock field strength.” The way this value is calculated depends on the specific sequence being used; see NMR staff if you have questions regarding how to publish this value.

- 3+4☞ Turn in a stack plot for each multiplet showing all mix times (including $d9=0$) plus the ^1H 1D spectrum. These spectra should confirm or extend the assignments you have made previously.

Turn in 4 spectra (all stack plots) plus the answer to 1 question.

³ Do not choose singlets. Remember that TOCSY spectra will correlate only those protons with $J_{\text{HH}} \geq 2\text{-}3 \text{ Hz}$.