Supporting Information

Mapping Molecular Perturbations by a New Form of Two-Dimensional Spectroscopy.

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Figure 1S shows the lineshape transformation in a simulated two-dimensional TIPSY spectrum upon sine-bell window multiplication. Following the Radon transform and shearing of the spectrum shown in Fig 1 (see the main text) the cross-peaks show the characteristic 'butterfly' lineshape (Fig. 1S, the bottom panel). The top panel of Fig. 1S shows the effect of sine-bell window multiplication applied to the directly detected dimension. It largely suppresses the broad features in the spectrum leaving star-shaped cross-peaks that are similar to the lineshapes familiar in conventional two-dimensional NMR spectroscopy.

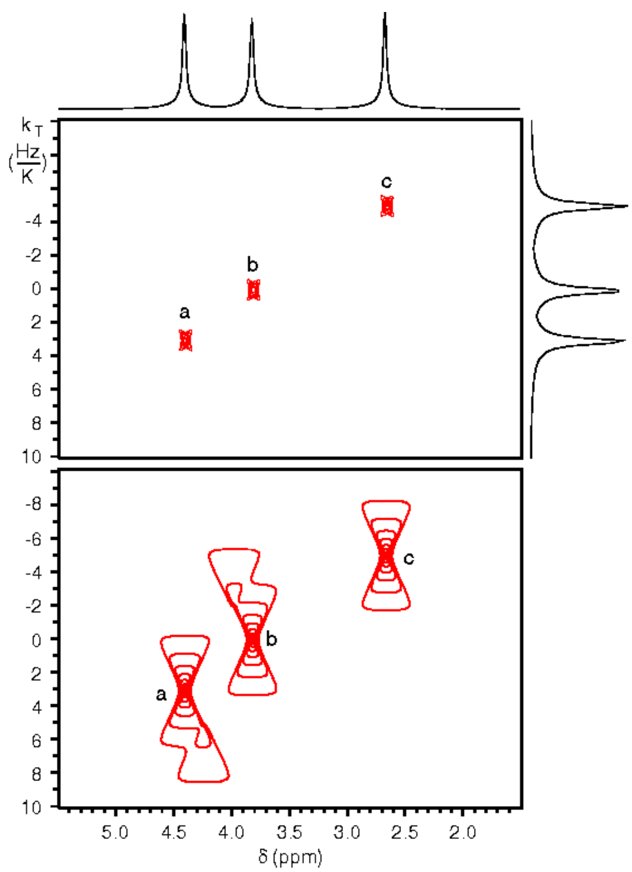

Figure 1S. Lineshape transformation following sine-bell multiplication of the FID-s in a two-dimensional TIPSY spectrum shown in absolute value mode. The bottom panel shows the 'natural', butterfly lineshapes and the associated artefacts due to the overlap of their pedestals (peaks a and b). The top panel shows the star-shaped lines and their 'skyline' projections after window multiplication. For further details see Fig. 1 in the main text.

Figure 2S shows the effect of temperature dependence, J_T of the *J*-coupling on doublets in the TIPSY spectra. Independently of the position in the spectrum, the doublet appears tilted anti-clockwise if the *J*-coupling increases with temperature ($J_T > 0$) and clockwise if the *J*-coupling decreases with temperature. The magnitude of the temperature effect can be measured directly from the vertical displacement between the two doublet components in the TIPSY spectra. The effects of both homo-nuclear and hetero-nuclear J-couplings are expected to influence the TIPSY spectra similarly (neglecting any higher-order effects).

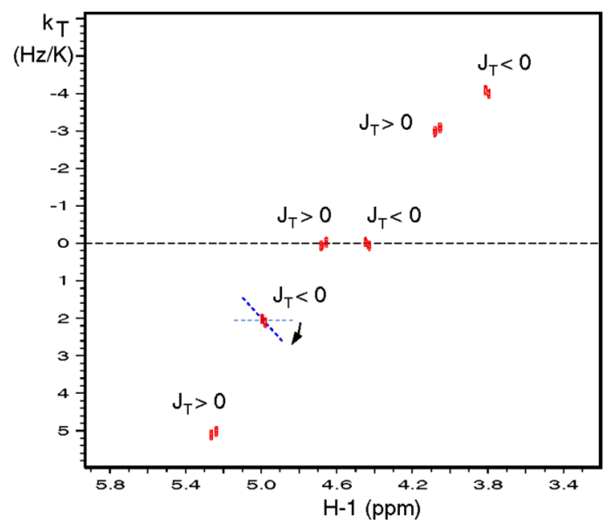

Figure 2S. The effect of J-coupling temperature dependence on the appearance of cross-peaks in two-dimensional TIPSY spectra. All J couplings were assumed to be 12 Hz and their temperature coefficients, J_T were assumed to be either +0.1 Hz/K or –0.1 Hz/K.

Since the Radon transform is relatively new to NMR spectroscopy, we deliberately chose to outline the method in the simplest, most transparent form, where the directly detected dimension is processed first. In actual practice processing of the evolution dimension is more conveniently implemented while the data is still in the time domain This comprised projection at an angle α, incrementation of α over a suitable range, storage of these projections in a two-dimensional matrix, and finally the shearing operation.[13] Fourier transformation of the direct detection dimension was in fact the last stage.

The sensitivity of TIPSY to small perturbations clearly depends on the resolution in the evolution (tanα) dimension, determined first of all by the extent of the evolution dimension, by the linewidth in the directly detected dimension, and the fineness of the steps in α. Line broadening can be reduced to a limited extent by application of a windowing function, or by other processing techniques (currently under investigation). We have chosen to show TIPSY peaks in the absolute-value mode, but phase-sensitive responses can be displayed if required. Indeed, once the baseline 'butterfly' effect has been corrected, the general form of TIPSY peaks closely resembles that seen in conventional two-dimensional NMR (see Fig 1S, top panel).

For simplicity of presentation we assumed that temperature-dependent shifts were linear, but this is not a *necessary* condition. Small deviations from linearity sacrifice some sensitivity because the integrated intensities are diminished. Non-linearities are also reflected in the lineshape of the correlation peaks, as can be seen for the NH peak in Figure 3. This indicates that lineshapes provide further detail about the character of the perturbations. Further investigation of this and other phenomena is currently underway and will be described in detail elsewhere.

References

13) Ernst R. R.; Bodenhausen G.; Wokaun, A. *Principles of Nuclear Magnetic Resonance in One and Two Dimensions*, Oxford University Press, **1987**, Ch 6, 336-338.