High-Resolution Two-Field Nuclear Magnetic Resonance Spectroscopy at 0.33 and 14.1 T

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High magnetic fields enhance the sensitivity and resolution of Nuclear Magnetic Resonance experiments. Therefore, a significant effort is being focused on the development of NMR spectrometers with the highest magnetic field possible. However, some nuclear properties become less favorable at high fields than at low fields. (i) The range of NMR frequencies may become too large to obtain effective irradiation by limited radiofrequency fields, (ii) the transverse relaxation of nuclei with high chemical shift anisotropy can become too fast at high fields, (iii) chemical exchange may lead to severe signal broadening even beyond the detection limit.

Here, we present a unique NMR spectrometer that allows excitation of ^{1}H, ^{13}C and ^{15}N spins and their signal observation at two magnetic fields (Fig. 1). A standard NMR spectrometer (at 14.1 T) is augmented by a second homogenous magnetic field center (0.33 T) in the stray field of the superconducting magnet. The low-field center is equipped with a triple resonance Z-gradient probe and the sample is transferred between the high field and the low field position in ~100 ms by a pneumatic shuttling system.

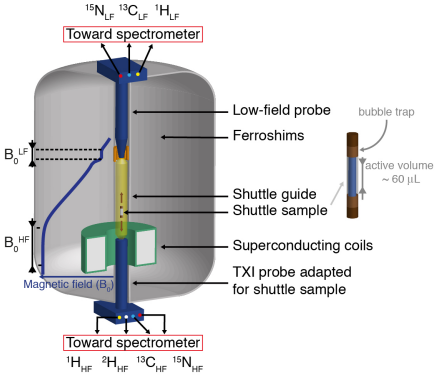

Figure 1. Schematic description of the main components of the two-field NMR spectrometer.

The advantage of the two-field system has been demonstrated by a study of a system in chemical exchange. In 4-methoxy-*N*,*N*-dimethylbenzotriazene (Fig. 2a), two methyl groups undergo chemical exchange in the intermediate regime at 14.1 T. The broadening of their carbon signal in a classical HSQC is more pronounced as the coalescence temperature (~ 35° C) is approached (Fig. 2b-d). We designed a new two- field NMR experiment 2F-HZQC in which the evolution in the indirect dimension takes place at the low field position, while the signal in the direct dimension is detected at the high field position. The evolution of zero-quantum coherence in the indirect dimension is used in order to suppress the effect of remaining inhomogenities of the magnetic field. Then, the carbon chemical shift is re-introduced using a shearing transformation during the processing. The signal of the exchanging carbons is obtained in 2F-HZQC as the exchange at low field is in the fast exchange regime.

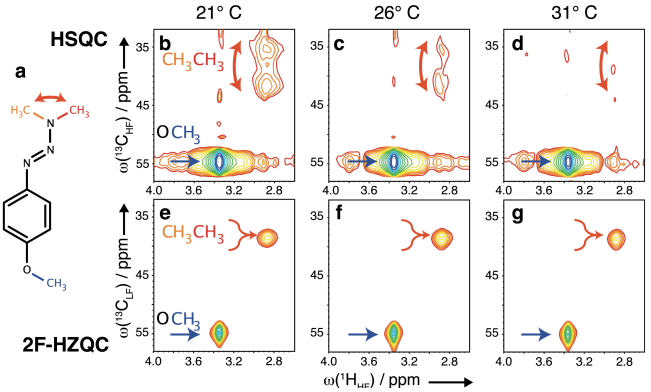

Figure 2. (a) The triazene compound. The two methyl groups highlighted in orange and red undergo chemical exchange, the methoxy group in blue provides a reference signal. Upper boxes (b-d) correspond to the high-field HSQC. Lower boxes (e-g) correspond to the two-field HZQC spectra.

^{13}C chemical shifts span a wide range so that broadband rf pulses are difficult to obtain at high field. In particular, ^{13}C TOCSY experiments are very challenging in high-field NMR. The two-field NMR spectrometer was used for a ^{13}C TOCSY experiment. The sample was transferred to the low-field position for the TOCSY irradiation period to profit from the small range of ^{13}C resonance frequencies at 0.33 T, while the sample polarization and the signal detection occurs at the high field position. Correlations between all carbons in ^{13}C labeled leucine and phenylalanine were detected by the two-field TOCSY experiment (Fig 3a) in contrast to the conventional high field TOCSY experiment (Fig. 3b). The two-field TOCSY experiment provides correlations even between carbonyl and methyl ^{13}C in leucine (over 150 ppm difference in chemical shifts) with a weak FLOPSY isotropic mixing irradiation ($\omega_1/2\pi$ = 1.5 kHz).

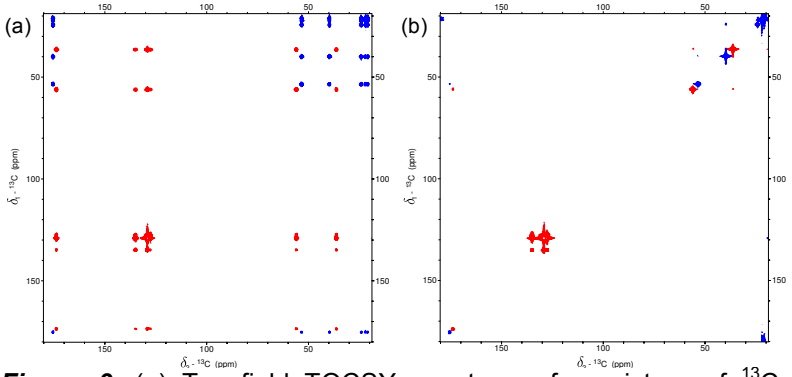

Figure 3. (a) Two-field TOCSY spectrum of a mixture of ^{13}C labeled phenylalainine and leucine. (b) High-field TOCSY spectrum acquired at single field (14.1 T). The peaks of leucine (blue) and phenylalanine (red) are distinguished by color.