

Observing Proton Exchange in Aqueous Ethanol with a 60 MHz FT-NMR Spectrometer

Gregory A. Manley, David Rovnyak

Department of Chemistry, Bucknell University, Lewisburg PA 17837, drovnyak@bucknell.edu. (© G. Manley, D. Rovnyak, 2006)

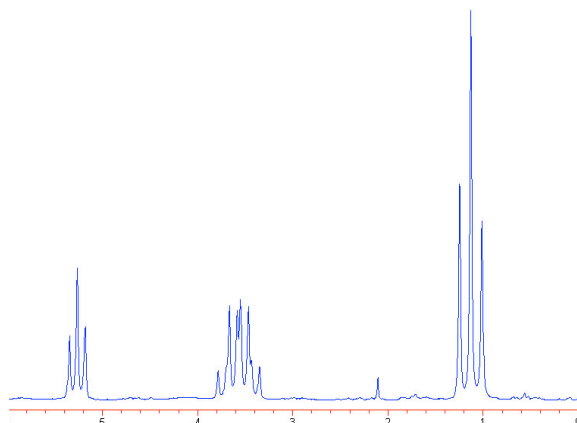
Introduction

We have implemented a classic experiment [1] for observing and characterizing proton exchange between ethanol and water on a permanent magnet pulsed FT-NMR spectrometer operating at 60 MHz for ^1H observation (EFT-60, Anasazi Instruments, Inc., Indianapolis, IN). Higher field spectrometers (e.g. 300 MHz) are not well suited to observe this process in ethanol/water solutions. Indeed, we have followed a study that utilized 40 MHz ^1H observation. [1,2]

This experiment should help meet two pedagogical goals. The first is to demonstrate in detail the exchangeability of alcohol protons in aqueous solution. This experiment challenges and clarifies the dangerously general assumption that alcohol protons are unobservable in aqueous solution. The second goal is to illustrate two separate spectral effects for exchange in ethanol:water solutions, termed coalescence and crossover, with which the exchange rate may be easily estimated.

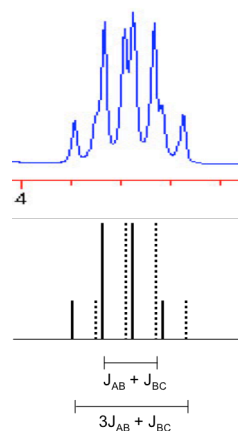
Results

A ^1H NMR spectrum of neat ethanol (>99.99% purity) is shown in Figure 1, where the narrow, well-resolved lines clearly show that the alcohol proton is not exchangeable.



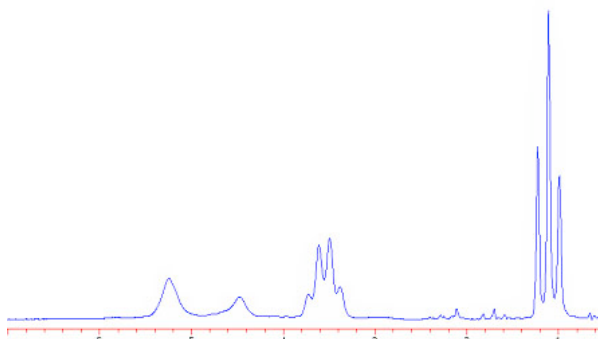
(Figure 1. ^1H NMR spectrum of 99.99% Ethanol)

The alcohol proton is a triplet at ~ 5.3 ppm due to scalar coupling with the two equivalent methylene protons. This is an important observation since it demonstrates that non-exchanging protons are always observed and subject to J-splittings. The methyl protons located at ~ 1.1 ppm are also a triplet. Interestingly, the methylene protons at ~ 3.6 ppm form a more complicated multiplet due to scalar coupling to both the methyl and hydroxyl protons. The three methyl protons lead to a quartet, which is then doubled by the scalar coupling to the hydroxyl proton as shown in Figure 2.



(Figure 2. An expanded view of the methylene multiplet observed in neat ethanol is shown in the top panel; in the bottom panel the doublet of quartets is schematically represented in which $J_{AB} > J_{BC}$; we identify A with the methyl protons, B with the methylene protons, and C with the hydroxyl proton)

In Figure 2 there are many ways to extract the individual scalar couplings, such as by examining the peak distances indicated in the bottom panel. In this case we find $J_{\text{H}^{\text{CH}_2}\text{H}^{\text{CH}_3}} = 6.81 \pm 0.12$ Hz and $J_{\text{H}^{\text{CH}_2}\text{H}^{\text{OH}}} = 5.37 \pm 0.18$ Hz.



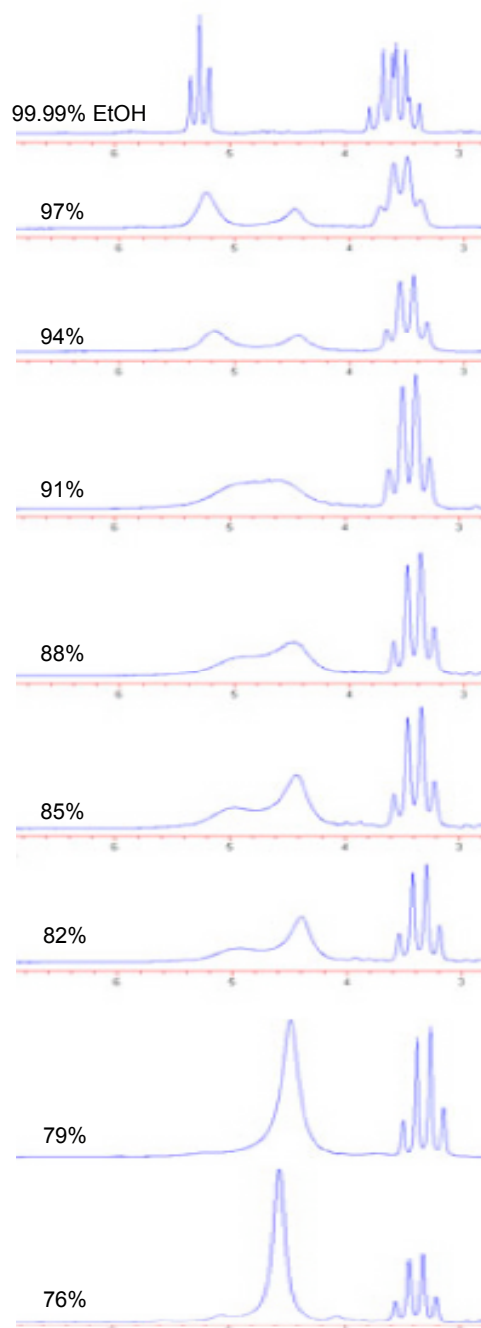
(Figure 3. Aqueous solution of 97% EtOH)

Proton exchange may be initiated by making an aqueous solution, such as the 97% (v/v) EtOH:H₂O solution whose ¹H 60 MHz NMR spectrum is shown in Figure 3. There is prominent broadening of the alcohol proton signal at ~5.3 ppm, and a new signal due to H₂O at ~4.5 ppm. The methylene signal is beginning to approximate a pure quartet since exchange diminishes the hydroxyl scalar coupling. Comparison of Figures 1 and 3 clearly demonstrates these typical spectral effects for exchange processes in NMR spectra.

What determines the time scale in which these exchange effects are observable in NMR spectra? To address this question, we examine a series of spectra in which the proton exchange rate is gradually increased with the concentration of water in solution (Figure 4) using the EFT-60 Anasazi spectrometer. These experiments were carried out at 27 °C, as determined using a neat methanol standard and the relation $T = -23.832\Delta^2 - 29.46\Delta + 403.0$. [3] Nine samples were prepared using 3% volume decrements of ethanol per sample (100%, 97%, 94%, 91%, 88%, 85%, 82%, 79%, 76% v/v EtOH:H₂O). As seen in Figure 3, the triplet corresponding to the alcohol proton collapses to a single, broad line, while two resonances emerge and then coalesce to a single line. The methylene multiplet changes to a quartet as the concentration of water increases. [1] The spectrum of the 97% ethanol sample yields approximately the separate ethanol and water chemical shifts when the exchange rate is slow. These will be the basis for estimating the exchange rate.

We first describe two limits on the observability of exchange phenomena. First, since the time-domain NMR signal is digitized, the time between the acquisition of individual data points, the dwell time, imposes a constraint on the short time resolution of the NMR technique. If rapid exchange takes place during the dwell time, only one signal corresponding to the average chemical environment of the rapidly exchanging proton will be observed. This could be an exchange timescale of up to 1-2 ms for ¹H NMR at 60 MHz, and this limit is being approached by the 79% and 76% EtOH spectra in Figure 4. Next, the role of the total acquisition time, which is the product of the dwell time and the number of data points

collected, should be considered. If during the acquisition period the exchange rate is so slow that no appreciable exchange of protons between *n* environments occurs, then *n* individual signals will be observed in the NMR experiment. This is almost the case for the spectrum of 97% v/v EtOH spectrum that is second from the top of Figure 4.



(Figure 4. ¹H 60 MHz NMR spectra of aqueous solutions of varying concentration of ethanol in water (v/v), as indicated in the figure; Spectra were obtained at 27 degrees Celsius).

Between these two extreme regimes, how does the exchange rate relate to observable changes in the spectra such as the *coalescence* and *crossover* points? Exchange effects on NMR spectra depend on how the exchange rate compares to the inverse of the peak separation. A good description of these processes is given by Levitt.[4]

The *crossover point* is the first appearance of a single line for the exchanging species. At the crossover point :

$$\frac{1}{\tau} \cong \pi(\nu_A - \nu_B) \quad (1)$$

where τ is the average lifetime of a hydroxyl proton between exchange events, and ν_A, ν_B are the peak positions in the slow exchange limit in units of Hz.

The spectrum of 79% (v/v) EtOH (note this is 75 wt% EtOH) in Figure 4 most closely resembles the criteria for the crossover point. Note also that at the crossover point, the single line occurs at a population weighted intermediate chemical shift of the two sites.

The *coalescence point* is perhaps more commonly employed in qualitative analyses of the type shown here, although it may not be any easier to determine unambiguously. At the coalescence point:

$$\frac{1}{\tau} \cong \frac{\pi}{\sqrt{2}}(\nu_A - \nu_B) \quad (2)$$

where the symbols have the same meaning as in Equation 1. Coalescence is recognized as the onset of the merging of the two lines. Levitt points out that the strict definition is that the second derivative of the frequency spectrum at the position of the average of the two frequencies vanishes.[4] The coalescence point will occur at a slower exchange rate than the crossover point, and the spectrum of 91% (v/v) EtOH (note this is 89 wt%) in Figure 4 appears to most closely fit the criteria for coalescence.

Identifying 91% (v/v) and 79% (v/v) as the points of coalescence and crossover respectively for aqueous ethanol solutions at 27 °C, we find:

	τ
Coalescence (91% v/v EtOH)	~9.8 ms
Crossover (79% v/v EtOH)	~7.0 ms

Discussion

This experiment should be appropriate for use in instruction since the total preparation time for the samples is considerably less than an hour, and all spectra were recorded on the EFT-60 spectrometer in well under one hour. Neat ethanol was inexpensively purchased at 99.99% purity and used without further distillation or drying. We believe this experiment could be effectively carried out by groups of up to 5-6 students. The experiment was originally devised as an independent study. The choice of 3% decrements was somewhat arbitrary and finer grained series could be studied with more students.

There are several potential sources for errors or inconsistencies in performing this experiment. Proton exchange is well known to be both acid and base catalyzed and will be sensitive to the quality of the dd-H₂O used to prepare samples. Recognizing the coalescence and crossover points is somewhat subjective and may not be unambiguous; indeed our own determinations are strictly qualitative. The growing intensity of the water resonance as EtOH content decreases causes distortions in the line shape during coalescence and crossover as can be seen in Figure 4. This also should not be mistaken for asymmetric exchange. Finally, the determination of $(\nu_A - \nu_B)$ would be better obtained from not less than a 99% EtOH solution.

Our results compare well to the observations of Weinberg and Zimmerman who do not report the temperature at which their experiments are performed.[1] They appeared to observe coalescence and crossover at 80 wt% and 70 wt% EtOH respectively and find a 15 ms average hydroxyl proton lifetime at 75 wt% EtOH.[1,2] We observe coalescence and crossover at 89 wt% and 75 wt% and find a 7 ms average hydroxyl proton lifetime at 75 wt%. While our observations are very close, we suspect that Weinberg and

Zimmerman were operating closer to standard temperature conditions (e.g. 20 °C), which would lead to a longer proton lifetime relative to the 27 °C temperature used in this study. Any acid or base contaminants in our samples could also have led to our observation of a slightly shorter proton lifetime.

Finally we note the inverse dependence between τ and $(\nu_A - \nu_B)$ as shown in Equations (1) and (2) which indicates that slower exchange processes will be better observed at low static fields such that $(\nu_A - \nu_B)$ is small. We were unable to observe coalescence in 300 MHz ^1H NMR spectra under a variety of temperature and concentration limits since the exchange is too slow to be easily observed at this higher field.

Summary

The proton exchange rate for the alcohol proton of aqueous solutions of ethanol may be estimated at two compositions which display the characteristic spectral features for coalescence and crossover points. This straightforward exercise illustrates dynamic processes in a binary solution and permits the characterization of the timescale of these processes.

References

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