ORGANIC LETTERS

2009
Vol. 11, No. 6
1349−1352

General Chromatographic NMR Method in Liquid State for Synthetic Chemistry: Polyvinylpyrrolidone Assisted DOSY Experiments

Jari S. Kavakka, Ilkka Kilpeläinen, and Sami Heikkinen*

Laboratory of Organic Chemistry, Department of Chemistry, University of Helsinki, P.O. Box 55, FIN-00014, University of Helsinki, Finland

Sami.Heikkinen@Helsinki.fi

Received January 22, 2009

ABSTRACT

A polymer assisted liquid state DOSY NMR technique, to extract the spectra of the individual components from mixtures in $CDCl_3$, is demonstrated. The enhancement of diffusion coefficient differences is achieved using a soluble polymer, polyvinylpyrrolidone, as the "stationary phase". The "separation" of analytes resembles normal-phase chromatography, for example, TLC. This method provides a fast, cheap, and simple technique to resolve the NMR spectra of complex mixtures.

NMR is an essential analytical tool in synthetic organic chemistry. Typically, several spectra are needed during the course of a synthesis: spectra from starting materials, intermediates and final products. As the NMR spectrum of a mixture can be very complex, a purification step is usually needed before the analysis. However, the deficiencies of NMR in the analysis of certain mixtures has led to the development of a "hyphenated technique": LC-NMR.[1] Unfortunately, this technique necessitates dedicated hardware that is not commonly available in most synthetic laboratories. In addition, the setup of LC-NMR runs can be quite tedious.

In feasible cases, LC-NMR-type data can be obtained without actual physical separation of compounds using an NMR technique called diffusion ordered spectroscopy (DOSY), where the spectra of the individual compounds in a mixture are separated according to their diffusion rates.[2−6] To perform this "chromatographic NMR" experiment, the spectrometer must be equipped with pulsed field gradient capabilities, hardware that is common for modern NMR spectrometers.

The diffusion NMR experiment measures molecular mobility, that is, the diffusion rate (diffusion coefficient, *D*). The diffusion coefficient is a property of a molecule as a

(1) Albert, K. *On-Line LC-NMR and Related Techniques*; Albert, K., Ed.; Wiley: Chichester, Sussex, 2002.

(2) Stejskal, E. O.; Tanner, J. E. *J. Chem. Phys.* **1965**, *42*, 288–292.
(3) Morris, K. F.; Johnson, C. S., Jr. *J. Am. Chem. Soc.* **1993**, *115*, 4291–4299.
(4) Chen, A.; Wu, D. H.; Jonson, C. S. *J. Am. Chem. Soc.* **1995**, *117*, 7965–7970.
(5) Morris, K. F.; Johnson, C. S., Jr. *J. Am. Chem. Soc.* **1992**, *114*, 3139–3141.
(6) Gibbs, S. J.; Johnson, C. S., Jr. *J. Magn. Reson.* **1991**, *93*, 395–402.

10.1021/ol9001398 CCC: $40.75
Published on Web 02/20/2009

whole and it is proportional to the molecular size (or more precisely, to the hydrodynamic volume). With a suitable measurement setup and processing (monoexponential fit,[7] inverse Laplace transformation,[3,8,9] DECRA[10,11]), the spectra of individual mixture components can be separated, provided the mobilities differ sufficiently and/or the spectral resolution is sufficient. Frequently, however, the mobility differences between compounds are not large enough for sufficient separation in the diffusion coefficient domain. Therefore, it has been suggested that a solid chromatography stationary phase material, like silica, could be added to the mixture sample to reinforce mobility differences via solute-stationary phase interactions.[12] As solid particles within a liquid NMR sample severely degrade spectral quality, magic angle spinning (MAS) can be applied to retain acceptable line widths. This, however, requires a spectrometer with solid-state capabilities. It is also possible to use a normal liquid state NMR probe if low concentrations of small particle sized silica are used and solvent susceptibility is matched to silica using special solvent mixtures.[13] In this approach, measurement times are somewhat restricted because silica will eventually precipitate. Pure liquid state DOSY experiments with narrow line widths using diffusion separation enhancing additives have also been described. Previously, Morris et al. have enhanced DOSY separation by analyte−micelle interactions in water solution.[14,15] Hodge et al.[16] utilized acid−base interactions when binding amine/carboxylic acid onto methanol soluble polymer-supported carboxylic acid/amine. Still, there is a need for an all-round method enabling LC-NMR-type data accumulation in the pure liquid state using a typical NMR spectrometer setup. Such a method would be a valuable everyday tool in synthetic organic chemistry.

The aim of the present study was to develop a general and simple method that would result in separation data resembling normal phase thin layer chromatography (TLC) for a large variety of functionalities. Deuterated chloroform ($CDCl_3$) was selected as a solvent because it is the most widely used NMR solvent in synthetic organic chemistry. To avoid any line shape problems originating from susceptibility differences, the "stationary phase" should also be soluble. We chose to use a polymer with a large molecular size as the "stationary phase" because a low polymer mobility would lead to a measurable change in the analyte's mobility, even with very weak polymer−analyte interactions.

It was concluded that a suitable polymer should contain polar groups to interact with and slow down the diffusion rate of polar molecules, more than less-polar ones. In addition, the NMR spectrum of the polymer should preferably be simple to avoid possible overlap with analyte signals. Otherwise, suppression of polymer signals must be applied, for example using presaturation or T_2-filtering. To maximize the intermolecular interactions between "stationary phase" and analytes, a relatively high concentration of polymer is desirable. This, in turn, must not cause too high sample viscosity, leading to extensive line broadening. After extensive testing with different polymers, an inexpensive commercially available polyvinylpyrrolidone (PVP) was found to be suitable as the "stationary phase" due to its high solubility in $CDCl_3$ and known ability to bind to a variety of organic molecules.[17] Commercial PVP (purchased from Sigma-Aldrich, MW 10000) was further purified to remove monomers and other small molecules: 4 g of PVP was dissolved in 170 mL of chloroform and the resulting solution added dropwise to 1 L of pentane, with vigorous stirring. Precipitated PVP was filtered and the procedure was repeated. The filtrate was dried under high vacuum for 18 h. A $CDCl_3$ solution of PVP remains low-viscous, even at high polymer concentrations (>200 mg/0.6 mL). Since the changes in viscosity alter the analyte's mobility, standardized conditions for all samples were chosen to allow comparison of compounds with different functional groups: 3 mg of analyte and 50 mg of PVP in 0.6 mL of $CDCl_3$. This PVP concentration is adequate for effective analyte-polymer interactions and still allowing for relatively narrow line widths (TMS line width ~1.2 Hz). A further increase in PVP concentrations resulted in only minor enhancements in the diffusion coefficient changes. For routine application of the proposed method, precise concentrations of PVP and analytes are not necessary. The PVP/analyte weight ratio should be high enough for sufficient polymer−analyte interactions.

To study changes in the analyte's diffusion coefficients ($D_{analyte}$), resulting from interactions with PVP, the effect of viscosity ($D_{analyte}$ in $CDCl_3$ vs $D_{analyte}$ in PVP/$CDCl_3$) was corrected for, as described earlier.[16] Interaction of the solvent with PVP was assumed to be negligible and therefore the viscosity corrected $D_{analyte}$ in $CDCl_3$/PVP was calculated, as ($D_{analyte}$ in PVP/$CDCl_3$) × ($D_{CHCl3\ without\ PVP}/D_{CHCl3\ with\ PVP}$). This viscosity correction procedure was performed only to evaluate interactions between PVP and different functionalities, i.e. to monitor if PVP behaves similar to silica in normal phase TLC. It should be emphasized, however, that an actual application of the proposed method to separate the spectra of analytes, using DOSY measurements, does not necessitate any viscosity corrections. DOSY data sets were recorded using 20 or 40 diffusion gradient strengths ranging from 0.5 to 20 G/cm. Because PVP gives rise to ^{1}H NMR signals at 1.5−2.5 ppm and 3.0−4.0 ppm, the polymer signals were suppressed by applying T_2-filtering (a 175 ms Carr-Purcell-Meiboom-Gill spin echo train, CPMG,[18,19]

(7) Press, W. H.; Teukolsky, S. A.; Vetterling, W. T. *Numerical Recipies in FORTRAN*, 2nd ed.; Press, W. H., Teukolsky, S. A., Vetterling, W. T., Flannery, B., Eds.; Cambridge University Press: New York, 1992; Vol. 1.

(8) Delsuc, M. A.; Malliavin, T. E. *Anal. Chem.* **1998**, *70*, 2146–2148.

(9) Malliavin, T. E.; Louis, V.; Delsuc, M. A. *J. Chim. Phys.* **1998**, *95*, 178–186.

(10) Windig, W.; Antalek, B. *J. Am. Chem. Soc.* **1996**, *118*, 10331–10332.

(11) Antalek, B.; Windig, W. *J. Chemom. Intell. Lab. Syst.* **1997**, *37*, 241–254.

(12) Viel, S.; Ziarelli, F.; Caldarelli, S. *Proc. Natl. Acad. Sci. U.S.A.* **2003**, *100*, 9696–9698.

(13) Hoffmann, R. E.; Arzuan, H.; Pemberton, C.; Aserin, A.; Garti, N. *J. Magn. Reson.* **2008**, *194*, 295–299.

(14) Morris, K. F.; Stilbs, P.; Johnson, C. S., Jr. *Anal. Chem.* **1994**, *66*, 211–215.

(15) Zielinski, M. E.; Morris, K. F. *Magn. Reson. Chem.* **2009**, *47*, 53–56.

(16) Hodge, P.; Monvisade, P.; Morris, G. A.; Preece, I. *Chem. Commun.* **2001**, 239–240.

(17) Köhler, J. *Chromatographia* **1986**, *21*, 573–582.

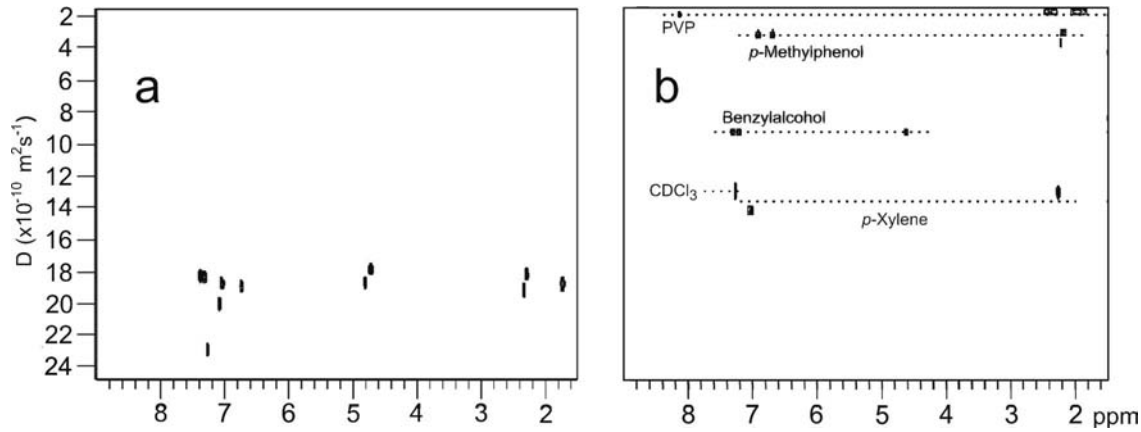

Figure 1. ^{1}H DOSY spectra (500 MHz) of benzylalcohol, *p*-methylphenol, and *p*-xylene mixture before and after addition of 50 mg of PVP. Three milligrams of analyte (each) in 0.6 mL of $CDCl_3$. DOSY spectra were processed with Varian VNMR 6.1C spectrometer operating software. Exchangeable proton signal at 4.8 ppm (a) disappears after the addition of PVP (b, see text for details).

implemented in the Bipolar Pulse Pair Stimulated Echo pulse sequence, BPPSTE,[20] prior to the acquisition period led to sufficient PVP signal suppression).

In Figure 1, the strength of the method in enhancing the performance of DOSY is demonstrated: three compounds of similar sizes were mixed and a DOSY spectrum was measured. Since the compounds have approximately similar mobilities in $CDCl_3$, the diffusion coefficients are very close to each other and a sufficient separation of individual spectra is not achieved under standard DOSY conditions (Figure 1a). After the addition of PVP, DOSY clearly resolves the spectra of the compounds (Figure 1b). Naturally, small changes in chemical shifts can occur for compounds that strongly interact with PVP. Especially, the line widths and/or chemical shifts of exchangeable protons can be affected, due to changes in exchange rates or from interactions with the polymer. Notably, the aromatic and methyl signals of *p*-xylene appear at slightly different D-values (Figure 1b). This is due to the partial overlap of the *p*-xylene methyl with the PVP signals and subsequenly the inability of the simple DOSY algorithm to treat multiexponential decays. The use of more sophisticated analysis programs (vide supra) would solve this ambiguity.

Figure 2 demonstrates the effect of PVP addition on the diffusion coefficients for six compounds, bearing different functionalities. A larger set of diffusion coefficients for 29 compounds with different functional groups can found in the Supporting Information. As expected, the interaction of the analytes with PVP essentially follows normal-phase chromatography rules, that is, the interaction of the stationary phase with polar molecules is stronger than with nonpolar ones.

In conclusion, we have demonstrated a generally applicable, polymer assisted, purely liquid state DOSY technique which can be easily adopted as a routine analysis tool for synthetic organic chemistry. The presented method significantly improves the performance of DOSY in crude mixture analyses by enhancing resolving power in the diffusion coefficient domain.

The use of commercially available and inexpensive PVP as "stationary phase" in $CDCl_3$ allows for significantly

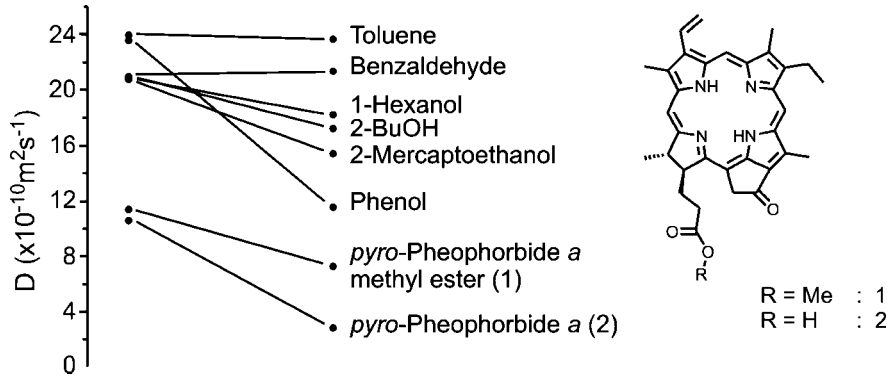

Figure 2. Measured diffusion coefficients (*D*) before (left) and after addition of PVP (right, viscosity corrected).

increased spectral separation of compound mixtures with the DOSY experiment.

Because PVP interacts with a broad spectrum of functionalities, the technique described herein is not functional group restricted. Obviously, if the ^{1}H spectrum alone does not provide sufficient spectral resolution for successful application of basic DOSY, it is expected that more sophisticated DOSY variants (DOSY-INEPT,[21] DOSY-HMQC,[22] DOSY-COSY,[23] etc.) with advanced processing algorithms, such as inverse Laplace transformation,[3,8,9] may be used. We are currently adapting the proposed technique to functional group specific and reversed-phase systems.

Supporting Information Available: PVP purification procedure, list of all analytes, detailed NMR measurement and processing parameters, as well as diffusion coefficient data for all studied analytes. This material is available free of charge via the Internet at http://pubs.acs.org.

OL9001398

(18) Carr, H. Y.; Purcell, E. M. *Phys. Rev.* **1954**, *94*, 630–638.
(19) Meiboom, S.; Gill, D. *Rev. Sci. Instrum.* **1958**, *29*, 688–691.
(20) Pelta, M. D.; Barjat, H.; Morris, G. A.; Davis, A. L.; Hammond, S. *J. Magn. Reson. Chem.* **1998**, *36*, 706–714.
(21) Wu, D.; Chen, A.; Johnson, C. S., Jr. *J. Magn. Reson. A* **1996**, *123*, 215–218.
(22) Barjat, H.; Morris, G. A.; Swanson, A. G. *J. Magn. Reson.* **1998**, *131*, 131–138.
(23) Wu, D.; Chen, A.; Johnson, C. S., Jr. *J. Magn. Reson. A* **1996**, *121*, 88–91.