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Two-Dimensional Correlation Infrared Spectroscopic Study of *N*-Methylacetamide as a Function of Temperature

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Generalized two-dimensional (2D) correlation vibrational spectroscopy has been used to study *N*-methylacetamide (NMA) as a function of temperature. It has been found that all the major bands of NMA are shifted in frequency as the temperature is raised. This gives rise to the appearance of new features in the 2D plots. These new features are not true bands, because they are not fundamental normal modes of vibration, but correspond to difference bands used to generate the correlation plots. Analysis of the amide I region of the spectrum suggests that transition dipole coupling plays a key role in the splitting of the amide I band in the liquid state.

Introduction

Generalized two-dimensional (2D) correlation spectroscopy, developed as a result of the seminal work of Noda,[1−3] has generated considerable interest and has been applied to a range of systems. The technique allows spectra that are obtained as a function of variables such as temperature and concentration to be spread over two frequency dimensions. As a result, it has been claimed that enhanced spectral resolution has been obtained and previously undetected bands revealed. Furthermore, the correlation spectra, most often reported as contour plots, have been interpreted in terms of a sequence of intensity change, which are then, in turn, related to structure or interactions. In studies of the self-association of *N*-methylacetamide (NMA) as a function of temperature,[4,5] for example, four amide I modes were detected in the asynchronous spectra and the sequence of intensity changes (1635 < 1650 < 1665 < 1685 cm^{-1}) was interpreted in terms of the presence of oligomers of various sizes, whose concentration changed systematically with temperature.

Work in our laboratory has for many years focused on infrared spectroscopic studies of hydrogen bonding, particularly in polymer blends, so we were intrigued by the potential power of this technique. However, we were particularly troubled by the interpretation of the data on NMA, as it neglects a broad range of studies that show that the splitting of the amide I mode in this material as well as polypeptides and nylons is a result of transition dipole coupling (see refs 6 and 7 and citations therein). Furthermore, as a result of a detailed examination of polymer blends obtained as a function of concentration,[8,9] we found that certain new bands revealed in previous studies[10−13] actually correspond to difference spectra bands that are generated by small bandwidth change and/or frequency shifts. In immiscible blends these changes can occur simply as a result of the problems associated with sample preparation (reproducibly casting films that are not uneven, which can lead to “wedge effect”, etc.). In studying materials such as NMA as a function of temperature, however, these problems do not occur, because the spectrum of the same sample is being obtained at different temperatures. Nevertheless, it is well-known that frequency shifts occur as a function of temperature. We will show here that it is these frequency shifts that give rise to difference spectra bands that become “new” features in asynchronous spectra. These asynchronous bands do not necessarily correspond to the fundament normal modes themselves.

Experimental Section

High purity (purity greater than 99.0%) NMA was purchased from Aldrich Chemical Co. and was used without further purification. The sample was prepared as a thin liquid film between two KBr windows, which are sealed with Teflon tape.

Infrared spectra were obtained on a Digilab FTS-60 Fourier transform infrared (FTIR) spectrometer using a minimum of 64 co-added scans at a resolution of 2 cm^{-1}. The film was sufficiently thin to be within the absorbance range where the Beer−Lambert law is obeyed.

IR spectra were baseline corrected. Standard two-dimensional correlation analysis was carried out with a Matlab software program written in this laboratory. An experimental matrix is formed from the original IR spectra ordered in the direction of decreasing temperature. The average (or mean) spectrum of the set is then subtracted from each of the original spectra to obtain a set of “dynamic” difference spectra. Synchronous and asynchronous correlation spectra are then calculated from these dynamic spectra using the Hilbert transform suggested by Noda.[2] The spectra shown on the top and at the side of the 2D correlation maps are the average spectrum and/or the difference spectrum obtained from the highest temperature spectrum in the set. In the 2D correlation plots, shaded regions indicate negative correlation intensities, while unshaded ones indicate positive correlations.

Results and Discussion

We will start by considering in detail the amide I region of the correlation spectra generated from the infrared spectra of NMA obtained as a function of temperature in the range 30−70 °C. Synchronous and asynchronous contour plots are shown in Figure 1. These are similar to the plots reported by Noda et

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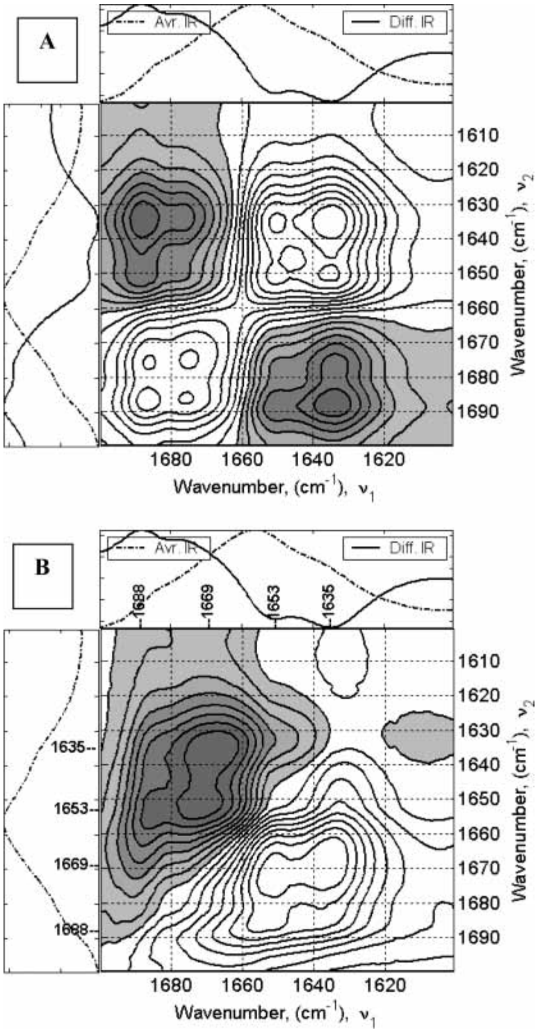

Figure 1. Synchronous (A) and asynchronous (B) contour plots in the region 1700−1600 cm^{-1} calculated from NMA spectra obtained as a function of temperature (30 °C− 70 °C).

al.[4] and Ozaki et al.,[5] but there are also some important differences. First, the synchronous contour plot has negative (shaded) cross-peaks, indicating that the intensities of the corresponding bands are changing in the opposite direction with temperature. In the work cited above, the intensities of all the bands in the original infrared spectra decreased with increasing temperature, so negative cross-peaks were not observed. This is simply a sample preparation problem, however. NMA absorbs strongly in the infrared, so in order to obtain spectra that are within the Beer−Lambert law range one cannot use commercially available sealed cells, as they are too thick. Instead a thin film of liquid squeezed between salt windows is analyzed. Unfortunately, we found that under these conditions the sample started to evaporate as the temperature was raised. We managed to limit this by wrapping Teflon tape around the edges of the window, but some evaporation still occurred. Nevertheless, if one carefully examines the infrared spectra in the amide I region, shown in Figure 2, it can be seen that the bands near 1656 and 1635 cm^{-1} that can be assigned to hydrogen-bonded groups decrease with increasing temperature, while the band due to what we will call end-group free carbonyl groups near 1680 cm^{-1} increases slightly with increasing temperature, as one would expect (band assignments will be discussed in detail later in this paper). This increase will no doubt be more pronounced if we could completely prevent evaporation, but it is nevertheless sufficient for cross-peaks to be observed.

Turning now to the asynchronous contour plot, the "standard" analysis of the results shown in Figure 1 would lead one to conclude that there are four bands present, near 1688, 1669, 1653, and 1635 cm^{-1}, respectively. A similar but not identical pattern was reported by Noda et al.[4] and Ozaki et al.,[5] with four bands being identified, but at somewhat different frequencies. This difference is presumably due to the fact that we were able to limit the evaporation of sample to a greater extent than these authors and our upper temperature was higher, but it will not be important in what follows. What is important and immediately striking is that none of these bands corresponds in frequency to the position of the main band near 1656 cm^{-1}. Certainly two strongly overlapping bands of equal intensity can result in the appearance of a single mode centered somewhere between the two, but they have to be of equal intensity, otherwise asymmetric bands would be observed. A close examination of the parent spectra and the difference spectra used to generate the contour plots suggests that is not occurring here.

It is clear from Figure 2 that the amide I and II bands shift as a function of temperature, the amide I bands to higher frequency and the amide II band to lower frequency. This is well understood. As the temperature increases, the hydrogen bond weakens and lengthens, resulting in the observed changes. This frequency shift, although small, has a pronounced effect on the asynchronous spectrum. This is because it is not the parent spectra themselves that are used to generate these plots, but difference spectra obtained by subtracting a mean spectrum from each of these. For example, the difference spectrum obtained by subtracting the mean spectrum from the one obtained at 70 °C is shown in Figure 3. Subtracting two single bands that are shifted in frequency from one another results in

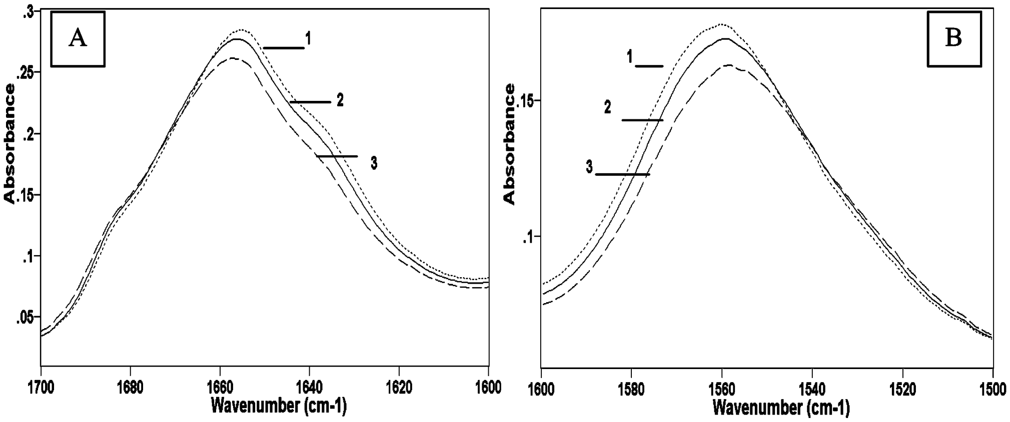

Figure 2. Infrared spectra of NMA in the region 1700−1500 cm^{-1} obtained at three different temperatures: (1) 30 °C, (2) 50 °C, and (3) 70 °C.

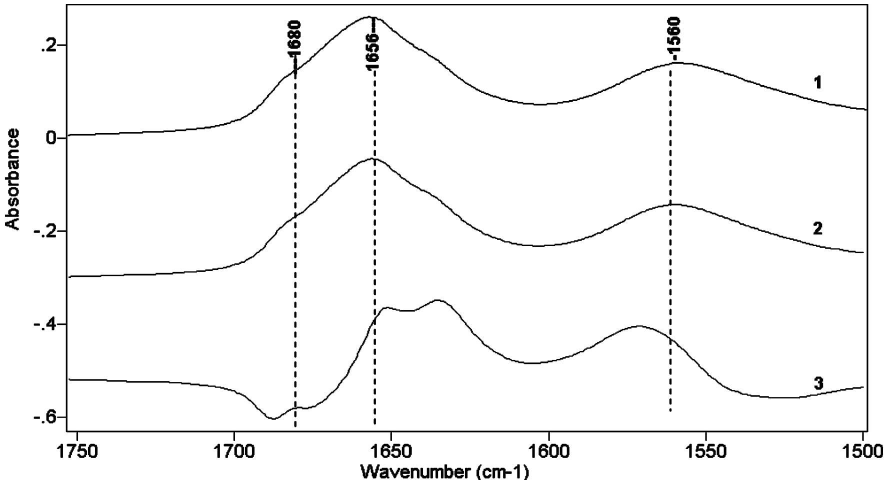

Figure 3. Infrared spectrum of NMA in the region 1750−1500 cm^{-1} taken at 70 °C(1), mean spectrum of the spectra obtained at 30, 50, and 70 °C (2), and the difference spectrum obtained by subtracting the mean spectrum from the one obtained at 70 °C (3).

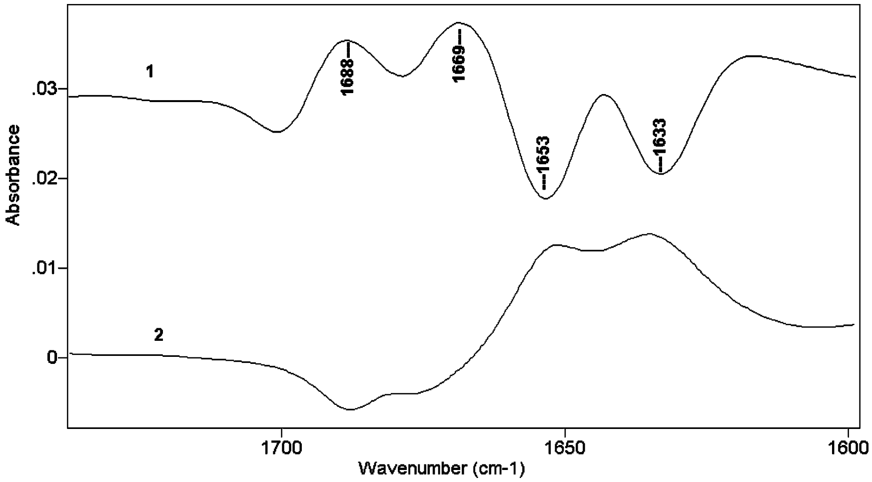

Figure 4. Difference spectrum (amide I mode) in the 1730−1600 cm^{-1} range obtained by subtracting the mean spectrum from the 70 °C spectrum (2), and the second derivative of this spectrum (1).

positive and negative difference bands that give a characteristic pattern in the asynchronous spectrum, as discussed in our recent work[9] and demonstrated in studies by other workers that preceded this.[14−18] In the amide I region the situation is more complicated, because we are subtracting two profiles made up of overlapping bands that are shifted in frequency. Nevertheless, the difference spectrum, shown in Figure 3, displays features, both positive and negative, that clearly do not correspond to the original bands, but are generated by the subtraction of one shifted profile from another. Figure 4 shows a scale-expanded section (amide I mode) of the difference spectrum and a second derivative of this spectrum to indicate the approximate position of these positive and negative features. It is these difference bands that generate the asynchronous bands, not the normal modes themselves.

The effect of frequency shifts can be more clearly seen in other regions of the spectrum that contain fewer overlapping bands. A simple examination by eye of the amide II region (see Figure 2) would suggest that this profile consists of a single broad band centered near 1560 cm^{-1}, corresponding to observations we have made in studies of various nylons.[19−21] The asynchronous spectrum, shown in Figure 5A, suggests the presence of two bands near 1570 and 1535 cm^{-1}, however. Again, features in the difference spectra, shown in Figure 6, are responsible for these contours. To reiterate, these difference bands do not correspond directly to frequencies of normal modes, but are the result of subtracting one frequency-shifted band or band profile from another. This effect is even more pronounced in the N−H stretching region, dominated by an intense band near 3304 cm^{-1} in the 70 °C spectrum (Figure 7). Again, because this mode shifts to higher frequency with temperature, the difference spectrum obtained by subtracting the mean spectrum shows two features near 3345 and 3276 cm^{-1}, as shown in Figure 7, that give rise to the contour peaks in the asynchronous spectrum shown in Figure 5B.

A simple examination by eye of the amide I and II modes, supported by the second derivative of these spectra, shown in Figure 8, suggests that there are three modes in the amide I region and one in the amide II region, as opposed to the total of six bands suggested by 2D correlation analysis. Using just four bands we indeed obtain a very good fit, as also shown in Figure 8 for the spectrum obtained at 30 °C. Although the primary goal of this paper is to demonstrate that infrared correlation spectra of materials such as NMA, where bands shift as a function of temperature, generate artifacts that have been interpreted as real infrared bands, it is also important to clarify band assignments, as there remain some conflicting assignments, as recently pointed out by Herrebout et al.[22] in this journal.

First, we should note in passing that *N*-methylacetamide is highly hygroscopic; even when great care is taken in sample preparation, trace amounts of water are almost inevitably present, as Herrebout et al.[22] pointed out. However, our spectra are

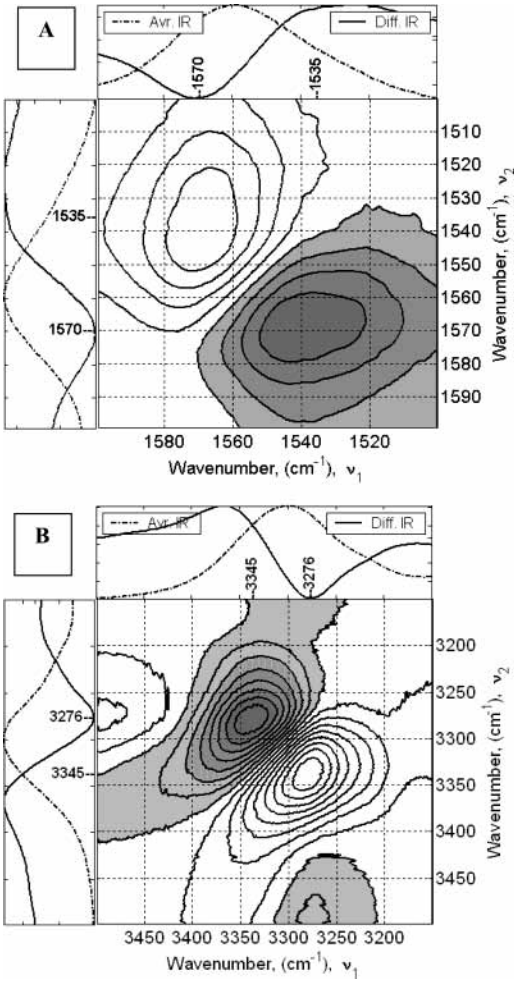

Figure 5. Asynchronous contour plots in the region of 1600−1500 cm^{-1} (A) and 3500−3150 cm^{-1} (B) of the data obtained from spectra taken at three different temperatures.

essentially the same as those reported by other authors,[22,23] leading us to conclude that water does not significantly affect our results and the interpretations that follow.

The recent correlation spectroscopy results were interpreted in terms of hydrogen-bonded oligomers of different lengths. Similarly, a study by Schweitzer-Stenner et al.[23] assigned a band near 1680 cm^{-1} (see Figure 8) to the presence of monomers, a band near 1658 cm^{-1} to dimers or oligomers, while a band near 1635 cm^{-1} was assigned to longer oligomers. We were perplexed by these assignments, because if there is a broad distribution of oligomeric species, and there undoubtedly is, then if oligomers of different lengths were each characterized by specific amide I bands, we would expect to see a single broad asymmetric band (it is easy to show that the statistics describing the distribution of hydrogen-bonded chain lengths is the same as the statistics of linear polycondensation, so the shape of this distribution is predictable). Accordingly, we would like to suggest a different interpretation.

We will first consider where “free” or non-hydrogen-bonded groups absorb. In studies of both amorphous and crystallizable nylons as a function of temperature,[19,20] we have shown that there is a free carbonyl band near 1680 cm^{-1}. This is an “end-group” free band, illustrated in Figure 9. As mentioned above, Schweitzer-Stenner at al[23] assigned this band to monomers. However, this assignment was based on the appearance of a band near 1675 cm^{-1} in the spectrum of dilute solutions of NMA in acetonitrile. In such solutions, the N−H group of NMA hydrogen bonds to the nitrogen of CH_3CN. This perturbs the electron density of the amide group, shifting the carbonyl band to lower frequency. This can be clearly seen if we compare the infrared spectra of dilute solutions of NMA in acetonitrile and cyclohexane, shown in Figure 10. In the latter solvent, which is not capable of hydrogen bonding, the “free” amide I band can be observed at 1703 cm^{-1}, and it is this band that should be assigned to non-hydrogen-bonded monomers.

Although it is interesting that electron delocalization through the amide group is such that hydrogen bonding the N−H group shifts the free carbonyl band about 23 cm^{-1}, the crucial point is that no “totally free” or monomer bands near 1703 cm^{-1} are observed, only end-group free vibrations, indicating that hydrogen bonding is quite extensive. Indeed, the number average length of the hydrogen-bonded chains is simply equal to the reciprocal of the fraction of free groups, which we estimate to be approximately 0.14, giving a number average length of 7 amide units at 30 °C. (This calculation is based on previous work on polyamides,[19−21] where we found that the ratio of absorption coefficients of bonded to free bands is 1.2. We assumed that both the 1635 and 1656 cm^{-1} modes corresponded to hydrogen-bonded groups—more on this shortly). This is important because Herrebout at al.[22] calculated the effect of cooperativity on the amide I band of NMA and found that the frequency of this mode converges to a constant value once the

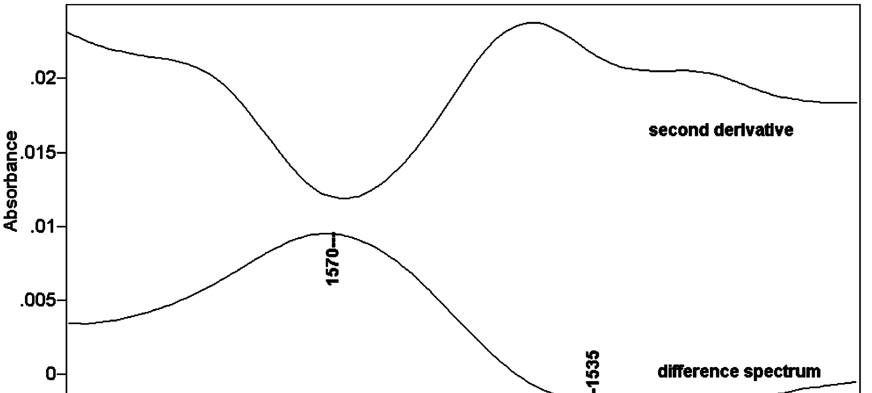

Figure 6. Difference spectrum (amide II mode) between 1600 and 1500 cm^{-1} obtained by subtracting the mean spectrum from the 70 °C spectrum (bottom), and the second derivative of this spectrum (top).

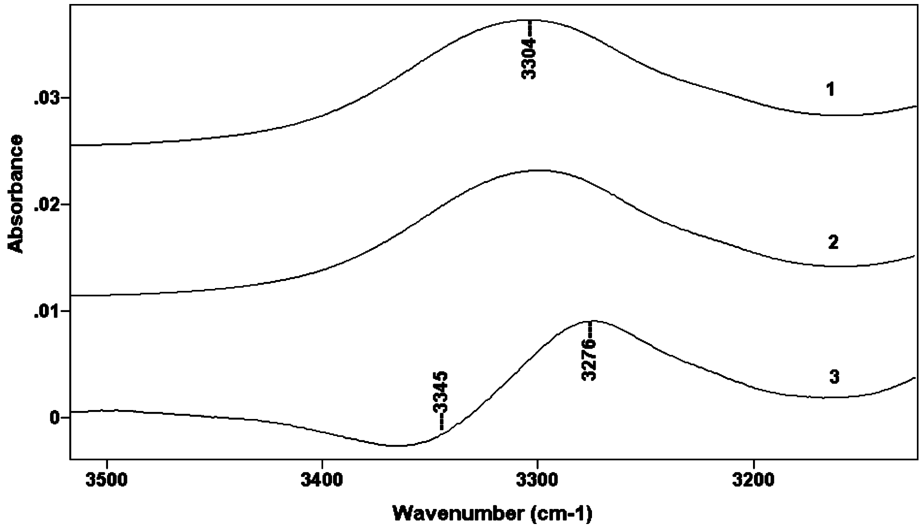

Figure 7. Infrared spectrum of NMA in the region 3500−3150 cm^{-1} taken at 70 °C (1), mean spectrum of the spectra set at 30, 50, and 70 °C (2), and the difference spectrum obtained by subtracting the mean spectrum from the one obtained at 70 °C (3).

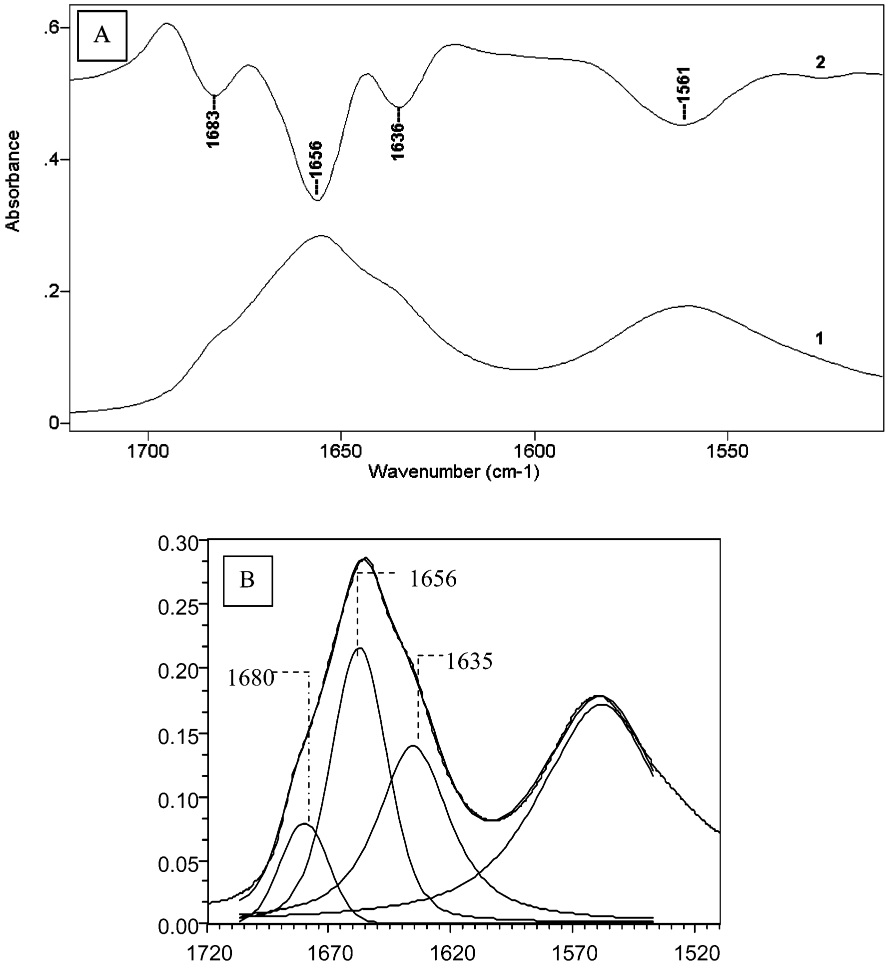

Figure 8. Infrared spectrum of NMA in the region 1750−1500 cm^{-1} taken at 30 °C (A-1) and its second derivative (A-2), and a schematic of the curve fitting (B).

number of molecules in the multimer reaches a value of about 5 or 6. Accordingly, we would suggest that the bands near 1635 and 1656 cm^{-1} have their origin in interactions between adjacent hydrogen-bonded amide groups in oligomeric chains. Adding support to this conclusion is the fact that these bands correspond to those observed in the disordered crystal form of NMA. In the ordered crystal form, found at temperatures below 10 °C, the hydrogen-bonded molecules are related by a 2-fold screw axis, while in the disordered crystal form, found at temperatures between 10 and 28 °C, these molecules can randomly have this orientation or one where their orientation is the same.[24] On going from the disordered crystalline state to the liquid form, the same bands persist in both the Raman and infrared spectra, with just the intensities of the Raman lines near 1656 and 1635 cm^{-1} changing.[22,23] This suggests that the hydrogen-bonded bands in these spectra reflect the same interactions between the amide

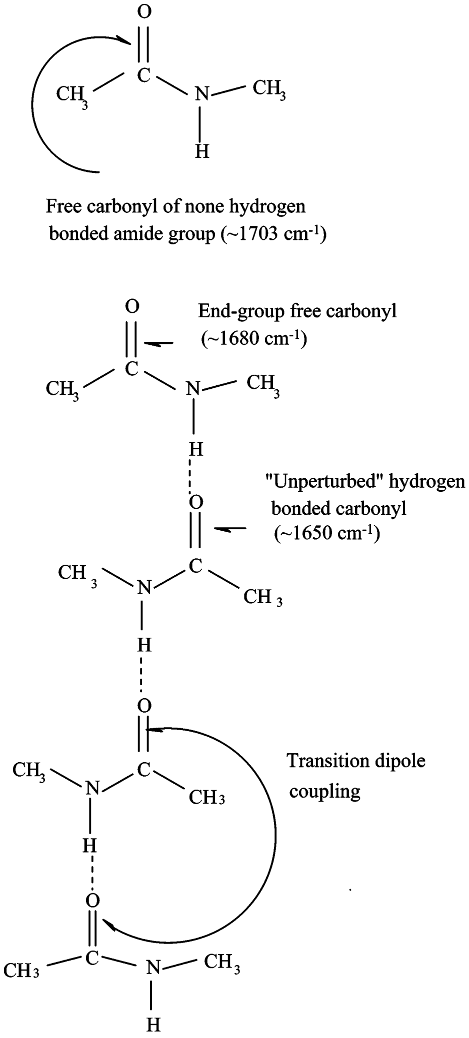

Figure 9. Schematic of free, end-group free, and “unperturbed” hydrogen-bonded carbonyl bands.

units, which Krimm and co-workers[6,7,23] have shown to be transition dipole coupling.

In the crystalline state, transition dipole coupling leads to some complexity,[22,23] but for essentially one-dimensional hydrogen-bonded chains the situation is simpler, if we assume that these oligomeric chains are arranged randomly with respect to one another such that interactions between chains cancel one another. In a long, ordered, one-dimensional chain the frequency of the amide I mode would then be given by

$$\nu = \nu_o + D\cos\delta \qquad (1)$$

where ν is the observed frequency, ν_o is the unperturbed (by transition dipole coupling) frequency of the hydrogen-bonded band, D is an interaction parameter, and δ is a phase angle between vibrations in adjacent amide groups. For molecules related by a 2-fold screw axis in an ordered hydrogen-bonded single chain, corresponding to the arrangement found in the crystalline state, two vibrational modes, corresponding to $\delta = 0$ and $\delta = \pi$, should be observed. In the liquid state, the arrangement of adjacent hydrogen-bonded groups in oligomeric chains is presumably less well defined, but for the totally symmetric vibration, $\delta = 0$, so that a random arrangement of adjacent hydrogen-bonded molecules, would not affect the frequency shift from the unperturbed (ν_o) value. A Raman line near 1635 cm^{-1}, observed in NMA liquids and the disordered crystalline form, can be assigned to this symmetric mode, which also appears as a shoulder in the infrared spectrum. For the nonsymmetric vibrational mode of ordered chains where the molecules are related by a 2-fold screw axis, the value of δ is π, whereas molecules with identical orientations have a phase angle of zero. For random sequences, which would seem to be likely in the liquid state, it would then seem reasonable to assume that $\cos\delta$ has the mean value of zero, so that the observed strongest infrared band at 1656 cm^{-1} would be observed near the unperturbed hydrogen-bonded frequency. This is very close to the frequency of the hydrogen-bonded carbonyl band observed in amorphous nylons[19] (1654 cm^{-1}), where similar arguments should apply.

Accordingly, even though there is undoubtedly a broad distribution of hydrogen-bonded chains in the liquid state, the results of Herrebout et al.[22] indicating that the observed frequencies converge to constant values at very short oligomeric chain lengths, together with a simple (if not simplistic) consideration of transition dipole coupling between adjacent groups in the hydrogen-bonded chains, would suggest that the observed amide I hydrogen-bonded bands are not from oligomeric species of different lengths, but are an average over the sample. This interpretation also makes sense in terms of the observed spectra. If chains of different lengths were each characterized by an individual amide I frequency, we would expect to see a broad, asymmetric band, not the observed, easily resolved pair of hydrogen-bonded bands.

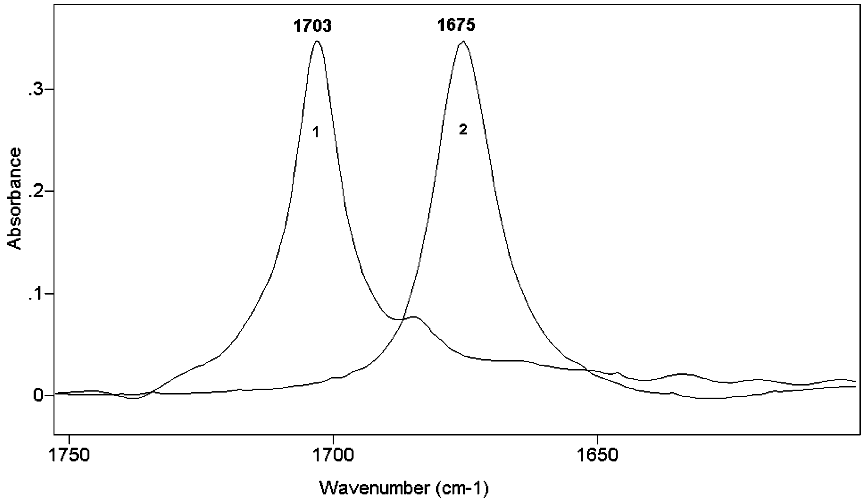

Figure 10. NMA spectra in the region of 1750−1600 cm^{-1} of dilute solutions of NMA in cyclohexane (1), and acetonitrile (2).

Conclusions

Many of the “new” features revealed in the asynchronous spectra of *N*-methylacetamide obtained as a function of temperature correspond to difference bands that are a result of frequency shifts. They are not fundamental normal modes of vibration. The observed amide I bands correspond to hydrogen-bonded and (end-group) “free” carbonyls, respectively. We suggest that the former are split as a result of transition dipole coupling.

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References and Notes

(1) Noda, I. *Appl. Spectrosc.* **1993**, *47*, 1329.
(2) Noda, I. *Appl. Spectrosc.* **2000**, *54*, 994.
(3) Noda, I.; Dowrey, A. E.; Marcott, C.; Story, G. M.; Ozaki, Y. *Appl. Spectrosc.* **2000**, *54*, 236A.
(4) N.; Noda, I.; Liu, Y.; Ozaki, Y. *J. Phys. Chem. B* **1996**, *100*, 8665.
(5) Ozaki, Y.; Liu, Y.; Noda, I. *Appl. Spectrosc.* **1997**, *51*, 526.
(6) Moore, W. H; Krimm S. *Biopolymers* **1976**, *15*, 2439.
(7) Moore, W. H; Krimm, S. *PNAS* **1975**, *72*, 4933.
(8) Huang, H.; Malkov, S.; Coleman, M. M.; Painter, P. C. *Macromolecules*, in press.
(9) Huang, H.; Malkov, S.; Coleman, M. M.; Painter, P. C. *Macromolecules*, in press.
(10) Nakashima, K.; Ren, Y.; Nishioka, T.; Tsubahara, N.; Noda, I.; Ozaki, Y. *J. Phys. Chem. B* **1999**, *103*, 6704.
(11) Matsushita, A.; Ren, Y.; Matzukawa, Y.; Inoue, H.; Minami, Y.; Noda, I.; Ozaki, Y. *Vib. Spectrosc.* **2000**, *24*, 171.
(12) Ren, Y.; Matsushita, A.; Matsukawa, K.; Inoue, H.; Minami, Y.; Noda, I.; Ozaki, Y. *Vib. Spectrosc.* **2000**, *23*, 207.
(13) Ren, Y.; Murakami, T.; Nishioka, T.; Nakashima, K.; Noda, I.; Ozaki, I. *J. Phys. Chem. B* **2000**, *104*, 679.
(14) Greike, A., et al. *Biospectroscopy* **1996**, *2*, 341.
(15) Czarnacki, M. A. *Appl. Spectrosc.* **1998**, *52*, 1883.
(16) Czarnacki, M. A. *Appl. Spectrosc.* **2000**, *54*, 986.
(17) Elmore, D. L., et al. *Appl. Spectrosc.* **2000**, *54*, 956.
(18) Morita, S., et al. *Appl. Spectrosc.* **2002**, *56*, 502.
(19) Skrovanek, D. J.; Howse, S. E.; Painter, P. C.; Coleman, M. M. *Macromolecules* **1985**, *18*, 1676.
(20) Coleman, M. M.; Skrovanek, D. J.; Painter, P. C. *Macromol. Chem., Macromol. Symp.* **1986**, *5*, 21.
(21) Bhagwagar, D. E.; Painter, P. C.; Coleman, M. M.; Krizan, T. D. *J. Polym. Sci., Part B, Polym. Phys* **1991**, *29*, 1547.
(22) Herrebout, W. A.; Clou, K.; Desseyn, H. O. *J. Phys. Chem. A* **2001**, *105*, 4865.
(23) Schweitzer-Stenner, R.; Sieler, G.; Mirtein, N. G.; Krimm, S. *J. Phys. Chem. A* **1998**, *102*, 118.
(24) Katz, J. H.; Post, B. *Acta Crystallogr.* **1960**, *13*, 624.