DFT-Calculated IR Spectrum Amide I, II, and III Band Contributions of N-Methylacetamide Fine Components

Yan Ji,* Xiaoliang Yang, Zhi Ji, Linhui Zhu, Nana Ma, Dejun Chen, Xianbin Jia, Junming Tang, and Yilin Cao

ABSTRACT: The infrared spectrum (IR) characteristic peaks of amide I, amide II, and amide III bands are marked as amide or peptide characteristic peaks. Through the nuclear magnetic resonance study, N-methylacetamide has been determined to have six fine components, which include protonation, hydration, and hydroxy structures. Then the independent IR spectrum of every component in N-methylacetamide is calculated by using the density functional theory quantum chemistry method, and the contribution of each component to amide I, II, and III bands is analyzed. The results of this research can help to explain the formation of the amide infrared spectrum, which has positive significance in organic chemistry, analytical chemistry, and chemical biology.

INTRODUCTION

The amide is very important in organic chemistry and chemical biology.1 The structures of proteins, enzymes, polypeptides, and many other biological molecules all have peptide bond units, which are based on the amide bond.2 Many protein structural properties are associated with polypeptide bonds, such as functional execution, proton conduction, transcription, information reaction, and other activities related to peptide amide.3 Therefore, the study of amide-peptide bonds has aroused great interest in chemistry and biology.

A large number of studies that included theoretical calculations and spectral methods have found that amides have some resonance structures,4 for example, the form of enol. The absorption spectrum bands of amide infrared radiation (IR), amide I (1600–1800 cm⁻¹), amide II (1470–1570 cm⁻¹), amide III (1250–1350 cm⁻¹), and amide A (3300–3500 cm⁻¹), are peaks of infrared characterization of amide. A protein peptide bond is an amide group. A protein structure is determined by using amide I, II, III, and A IR absorption peaks.5 The infrared spectrum and other spectra are used to study the amide, forming the infrared spectrum of amide I, II, III, and A bands. The infrared spectrum multiband of amide indicates that there are many fine components in the amide, and the structures are reported in recent research.6 How the fine component of amide affects the biological activity of the peptide bond is being recognized.

N-Methylacetamide is a secondary amide similar to the protein peptide bond. Several fine components were found in pure N-methylacetamide. NMR (15N, 17O, 13C, 1H, and two-dimensional NMR in Figures S1–S10) confirmed the presence of six fine components in pure N-methylacetamide samples,7 which were protonated and hydrated, but it cannot be purified as an independent component.

Then the infrared spectra of the components were calculated by using quantum chemical theory, and the contribution of each component to the infrared spectra of amide I, II, and III was analyzed. Further analysis of the contribution of the fine component–effect relationship to amide I, II, and III IR spectra is possible in future protein imaging. Fine components A-1, A-2, A-3, Im-1, D-1, and O-1 are shown in Figure 1, where E-1 is the structure not found.

RESULTS AND DISCUSSION

Methods and Theory. ADF infrared spectroscopy can be used for molecular vibrations.8 In the Born–Oppenheimer and harmonic approximations, the vibrational frequencies are determined by the normal modes corresponding to the molecular electronic ground-state potential energy surface. Various methods and basis sets have been tried for infrared calculations. The amide IR calculation test methods are Local Density Approximation (LDA) (with function -Xonly and -VWN), Generalized Gradient Approximation (GGA) (func-
Generally speaking, we can say that SZ is a single-zeta basis set, DZ is a double-zeta basis set, DZP is a double-zeta polarized basis, TZP is a core double-zeta, valence triple-zeta, polarized basis set, and finally, TZ2P is a core double-zeta, valence triple-zeta, doubly polarized basis. This explains the more intuitive names that are given for the basis sets. The infrared peaks of various methods are very close. The frequencies and intensities generally agree quite well. Improving the integration accuracy improves the agreement. The analytical frequencies are slightly more stable to changes in integration accuracy than the numerical values.

This section describes the general theory of the second analytical derivative of molecular energy where the derivative is about arbitrary parameters $X$ and $Y$. The theories described in this section also apply to the DFT and Hartree–Fock theories. In this section, we assume that the molecular energy $E$ is explicitly dependent on the orbital coefficient $c$ and parameters $X$ and $Y$, namely

$$E = E(c, X, Y)$$  

Although $E$ is clearly dependent on the orbital coefficient, which in turn depends on the coefficient of the $X$ and $Y$ parameters of the standard, throughout this article, $d/dX$ refers...
to X and is derived explicitly by the partial derivative, by taking the first derivative of total energy (eq 1) for the parameter X:

$$\frac{dE}{dX} = \sum_{\mu} \frac{\partial E}{\partial e_{\mu}} \frac{de_{\mu}}{dX} + \frac{\partial E}{\partial X}$$

(2)

Here, in the following discussion, the Greek characters are the index atomic basis functions, and Roman characters $i$ and $j$ are the indexes of the molecular orbit. It should also be noted that, in the instructions in this section, the first derivative of the energy is to the parameter $X$, and the second derivative is to the parameter $Y$.

However, for the derivatives of the available coefficients, other quantities of derivatives can also be calculated, such as the kernel derivatives of the density, $d/dR_A$, which can be used to calculate the dipole derivatives, such as:

$$\frac{d\mu_i}{dR_A} = -\int \frac{dp}{dR_A} + Z_A\delta_i$$

(3)

where $l$ is the dipole component, $I_{lex}$, and $Z_A$ is the nuclear charge of atom $A$, and the dipole derivative can be used to calculate the infrared absorption intensity and dipole strength. It is worth repeating that $dE/dY$ is not usually calculated explicitly but directly using $\mu_{ij}$ (eq 1).

This section discusses the basis of a second derivation theory for arbitrary parameters $X$ and $Y$, and the following sections discuss a second derivation theory for ADF whose parameters are nuclear coordinates.

Calculating the vibrational frequency of a molecule requires a second derivative of the molecular energy. They were also asked to calculate the hessian, from which a fixed point could be described as a maximum, minimum, or saddle point.

**Calculated IR Spectra.** Amide infrared radiation (IR) absorption spectrum amide I (1600–1800 cm$^{-1}$), II (1470–1570 cm$^{-1}$), III (1250–1350 cm$^{-1}$), and A (3300–3500 cm$^{-1}$) bands are the famous IR characterizations of amide. Protein peptide bonds are amide groups. The amide I, II, III, and A band IR absorption spectral peaks were used to determine the protein structures.$^{12,13}$ Fine components were protonation and hydration structures, and these were impossible to be purified to the independent component. Then quantum chemical software was used to calculate independently the IR spectrum of every component to analyze the contribution of every component to the IR spectra. Further analysis of the amide I, II, and III for the fine component—effect relationships’ contributions might be realized for protein imaging.$^{14}$ Fine components A-1, A-2, A-3, Im-1, D-1, and O-1, excluding E-1, calculated IR spectra are shown in Figures 2–7.

**IR Calculations.** The infrared spectrum is calculated by using the density functional theory (DFT) method of ADF$^{15}$ software. Based on NMR (Figures S1–S10) fine component data, the molecule structure of each fine component has been determined.$^6$ Structural optimization and frequency calculation were conducted to obtain the infrared spectrum of each fine component (Figures 2–7).

IR bands I, II, III, and A of N-methylacetamide were analyzed, and each fine component should contribute to the final IR spectrum.$^{16,17}$ Each chemical group has independent contributions to amide I, II, III, and A bands. The contributions of each component to amide I, II, III, and A bands were different. The infrared spectrum calculation results are based on chemical bond vibration information of chemical functional groups, so the calculation error can be received, and the results reflect the real information of each independent fine component. This means that determining the structure then determines the chemical groups and bonds and then the infrared spectrum vibrational position or wavenumber. Therefore, the ADF DFT method is accurate and feasible for infrared spectrum calculation.

Amide I band IR absorption ranges from 1600 to 1800 cm$^{-1}$. Based on the calculated IR spectra of fine components in N-methylacetamide, the A-1, A-2, and Im-1 have IR absorption peaks in the amide I band. A-1 has C=O and gave symmetry vibration, while molecular bond scissoring vibration produces IR absorption at 1630 cm$^{-1}$. A-2 has C–N symmetry vibration producing IR absorption at 1600 cm$^{-1}$. Im-1 has C=N symmetry vibration producing IR absorption at 1700 cm$^{-1}$. So, the fine components A-1, A-2, and Im-1 have contributions to the amide I band.

Amide II band IR absorption ranges from 1470 to 1570 cm$^{-1}$. The fine component A-1 has C=O connected carbon giving symmetry vibration that produces IR absorption at the 1518 cm$^{-1}$ peak. A-2 C–N–C=O has symmetry vibration that
produces IR absorption at the 1499 cm\(^{-1}\) peak. **A-2** C–H in-plane bending vibration produces IR absorption at the 1409 cm\(^{-1}\) peak. **A-3** C–C symmetry vibration produces IR absorption at the 1390 cm\(^{-1}\) peak. **O-1** CH\(_3\) hydroxygen has out-of-plane bending vibration that produces IR absorption at the 1444 cm\(^{-1}\) peak. **Im-1** C-H3 symmetry vibration produces IR absorption at the 1395 cm\(^{-1}\) peak. So, the **A-1**, **A-2**, **A-3**, **O-1**, and **Im-1** have contributions to the amide III band.

The amide III band has a range of 1250–1350 cm\(^{-1}\). **A-2** C–H has in-plane bending vibration that produces IR absorption at the 1388 cm\(^{-1}\) peak. **A-3** C–N symmetry vibration produces IR absorption at the 1215 cm\(^{-1}\) peak. The **D-1** molecular bond has symmetry vibration that produces IR absorption at the 1210 cm\(^{-1}\) peak. **O-1** CH\(_3\) hydroxygen has out-of-plane bending vibration that produces IR absorption at the 1082 cm\(^{-1}\) peak. **Im-1** C-H3 symmetry vibration produces IR absorption at the 1109 cm\(^{-1}\) peak. So, the fine components **A-2**, **A-3**, **O-1**, and **Im-1** have contributions to the amide III band.

The amide A band ranges 3300–3500 cm\(^{-1}\). **A-2** N–H2 has symmetry vibration that produces IR absorption at the 4076 cm\(^{-1}\) peak. **A-3** O–H symmetry vibration produces IR absorption at the 2795 cm\(^{-1}\) peak. **O-1** CH\(_3\) hydroxygen has out-of-plane bending vibration that produces IR absorption at the 2676 cm\(^{-1}\) peak. So, the fine components **A-2**, **A-3**, and **O-1** have contributions to the amide A band. The other IR absorption peaks and the fine component calculated IR absorption peaks that contributed to the amide I, II, III, and A bands are listed in Table 1.

A summary is shown in Figures 2–8. The **A-1**, **Im-1**, and **A-2** have contributions to the amide I band. The amide I was from the double bond (C=O, C=N) stretch vibration. The components **A-1** and **A-2** have carbonyl bond N–(C=O)–CH\(_3\) stretch vibration, which shows that the amide II was from the methyl-related C--
HorC−C stretch vibrations. Amide A was about 3500 cm\(^{-1}\), mainly from the H-N stretch/bend vibration or hydroxyl H-O-stretch/bend vibration.

The final IR spectra might be the comprehensive static results of the multicomponent, and the amide I and II changed with the ratios changing of fine components in an amide. The fine components may be transformed into others, which might be based on some changing environmental conditions (pH, temperature, metal ions, anions, light, heat, pressure, collision, solvents, air, wet, and so on). So, the amide final IR spectra have different states based on the ratios of fine components. These amide fine components are connected with the peptide bond IR spectra.\(^{18,19}\)

**IR Spectra.** To compare the calculated results, the infrared absorption spectrum of pure N-methylacetamide was recorded. The results are shown in Figure 9. It is generally believed that the peak of 3500 cm\(^{-1}\) in Figure 9 is from the infrared absorption peak of water, namely, the hydroxyl peak. In the fine component, the hydroxyl structure exists in components Im-1, O-1, D-1, and A-3. The hydroxyl \(–\text{OH}\) elastic vibration peak of IR is around 3000–4000 cm\(^{-1}\). This means that the hydroxyl peaks of amide 3000–4000 cm\(^{-1}\) in Figure 9 come from the \(–\text{OH}\) groups or \(–\text{CH}_3\) oscillations of the amide fine peak of IR is around 3000–4000 cm\(^{-1}\). This means that the hydroxyl peaks of amide 3000–4000 cm\(^{-1}\) in Figure 9 come from the \(–\text{OH}\) groups or \(–\text{CH}_3\) oscillations of the amide fine

Figure 8. Calculated IR spectrum of 1-(methyl-amino)ethan-1-ol (E-1) component by DFT methods. This E-1 was a reference component not found in N-methylacetamide. 2838 cm\(^{-1}\) (CH3-ν-s). 1390 cm\(^{-1}\) (C−C ν-s with C−H β). 1151 cm\(^{-1}\) (C−N ν-s) 1019 cm\(^{-1}\) (O−H β) 786 cm\(^{-1}\) (C−O ν-s), 599 cm\(^{-1}\) (N−H γ). 328 cm\(^{-1}\) (O−H γ).

Figure 9. Experimental test IR absorption spectrum of pure N-methylacetamide liquid sample (Shanghai Aladdin Reagent. Melting point 29–31 °C). (1656, 1564, 1375, 1303, 1164, 593, 3313 cm\(^{-1}\)).

Figure 10. Weight addition of the calculated IR absorption spectra (Figures 2–7) of fine components in N-methylacetamide. Compared with Figure 9, the real IR spectrum. (1630, 1518, 546 cm\(^{-1}\)).

Figure 11. Unity IR spectra with the calculation of all the calculated IR spectra contributions to the amide I, II, and III bands.

Figure 12. Comparison of the real IR spectrum with the calculation IR spectra.
resonance structure rather than from the IR absorption peaks of unpurified water. These results may correct the misunderstanding that the 3000–4000 cm⁻¹ is not from unpurified water; it should be from the amide several fine components.

Different components in the sample act together on the final infrared spectrum. Each component has a weight on the spectrum. Based on the integral area data of NMR, Figure 10 shows the weight addition for the IR spectra of the six components, which are compared with the actual IR spectrum in Figure 9. The components based on NMR integral area added weights to the infrared spectral contribution, and there is a big difference between Figure 10 and Figure 9. This means that the contribution of the fine component to the infrared spectrum may be a nonlinear weight ratio, which is subject to some mathematical functional relations and needs to be determined by mathematical methods. The result is close to but not equal to the real infrared spectrum. The comparison results are shown in Figure 12. The contribution of each component to the infrared spectra of amide I, II, and III bands is shown in Figure 11.

Figure 11 shows the fine components of amide I, II, and III contributions to the infrared spectrum. The contribution of each component to the infrared spectrum is seen. So, amide I has a contribution from A-1, A-2, and Im-1. The amide II band has contributions from all components. D-1 mainly contributes to amide III, and other fine components also contribute to the amide III band.

Figure 12 shows the comparison between the actual IR spectrum of N-methylacetamide and the calculated IR spectra of the fine components. The detailed properties of amide I, II, and III were defined by the analysis of each fine component. Therefore, it is helpful to find the relationship between the amide property and the structure. The structural effect of the amide or protein peptide bond might be determined by analyzing the infrared spectra of fine components.

## CONCLUSIONS

Based on 1H, 13C, 15 N, 17O, and two-dimensional NMR, it has been found that N-methylacetamide has several fine components, which include protonation, hydration, and hydroxyl structures. For several fine components of pure N-methylacetamide, the infrared spectrum of every independent component is calculated by the DFT method. The results show that the IR spectra of N-methylacetamide only by single structural analysis may interfere, and it is difficult to get accurate analysis results of the peptide or protein. Therefore, the infrared spectrum or other spectra of N-methylacetamide should be more clear and accurate by basing on these amide fine components. The detailed composition makes the structure–effect relationships easier to determine than they used to be in a confined structure. The results of infrared spectrum calculation of these fine components will be helpful to the study of amide I, II, III, and A and to even clarify some experiments and theories of arguing. The significance of these results is helpful for better analysis of amide I, II, III, and A bands in different microstructure ratios. These results can enhance the characterization of protein and help to discover the contribution of amide fine components to the structure–effect relationship of the peptide and protein.

The application of the two-dimensional vibration spectrum will be the next step in the restudy of amides. The vibration spectrum is a tool widely used in the study of molecular structures and surfaces. These one-dimensional techniques include infrared reflection absorption spectroscopy, Attenuated Total Reflection (ATR) spectroscopy, and sum frequency generation spectroscopy. More recently, multiple efforts have been made to extend the monolayer sensitivity of these techniques to vibrational spectroscopies of higher dimensionality. Two-dimensional infrared (2D IR) and other multidimensional techniques contain additional features that are not easily equated between the one-dimensional spectral peaks, such as the cross-coupled oscillator and the experimental separation ability of the isomorphic inhomogeneous donor and line width. These spectra provide a wealth of information on the structure and dynamics of samples and have been proven useful in bulk system studies in chemistry and biology. These spectral methods can help in amide component research in the future.

## ASSOCIATED CONTENT

**Supporting Information**

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.9b04421.

Figures S1 to S10, NMR spectra (pages S6–S15), Figure S11, electronic clouds of fine components (page S16), Figures S12–S18 (G09_MP2), Figure S19–S25 (G09_DFT), the calculated IR data of fine components (page S17–S30) (PDF)

## AUTHOR INFORMATION

**Corresponding Author**

Yan Ji – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China; orcid.org/0000-0003-3299-636X; Email: jyjian@htu.edu.cn

**Authors**

Xiaoliang Yang – School of Chemistry and Chemical Engineering, Nanjing University, Nanjing 210000, China
Zhi Ji – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China
Linhuizh Hu – College of Chemical and Environmental Engineering, Shandong University of Science and Technology, Qingdao 266510, China
Nana Ma – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China; orcid.org/0000-0003-3225-9554
Dejun Chen – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China
Xianbin Jia – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China
Junming Tang – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China
Yilin Cao – School of Chemistry and Chemical Engineering, Henan Normal University, Xinxiang 453007, China

Complete contact information is available at: https://pubs.acs.org/10.1021/acsomega.9b04421

**Author Contributions**

Y.J. conceived, designed, and carried out the experiments, analyze data, and wrote the paper. X.Y. and J.T. performed NMR and structure analysis. Z.J. and L.Z. performed calculations. N.M. gave support software. D.C. and X.J. took the spectra. Y.C. directed calculations and support devices.
Notes
The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

Henan Normal University Doctor Project Startup Science Foundations (QD15114); Henan Normal University National Project Cultivation Fund Funded Projects (2017PL05).

■ REFERENCES

(1) (a) Pauling, L. The Nature of the Chemical Bond; Cornell University Press: Ithaca, NY, 1960. (b) Wheland, G. W. Resonance in Organic Chemistry; Wiley: New York, 1955. (c) Carey, F. A. Organic Chemistry; 6th ed.; McGraw Hill: New York, 2006. (d) Wade, L. G. Organic Chemistry; 6th ed.; Pearson Prentice Hall: Upper Saddle River, NJ, 2006. (e) McMurry, J. Organic Chemistry; 6th ed.; Brooks/Cole-Thomson Learning: Belmont, CA, 2004.

(2) Peter, M.; Walker, B. (1990 ed.). Cambridge Dictionary of Science and Technology; reprint ed.). Press Syndicate of the University of Cambridge: Edinburgh, 1988, p 658.

(3) Nakamura, A.; Ishida, T.; Kusaka, K.; Yamada, T.; Fushinobu, S.; Tanaka, I.; Kaneko, S.; Ohta, K.; Tanaka, H.; Inaka, K.; Higuchi, Y.; Niihara, N.; Samejima, M.; Igarashi, K. “Newton’s cradle” proton relay with amide-imide acid tautomerization in inverting cellulose visualized by neutron crystallography. Sci. Adv. 2015, 1, e1500263.

(4) Kemnitz, C. R.; Loewen, M. J. “Amide resonance” correlates with a breath of C-N rotation barriers. J. Am. Chem. Soc. 2007, 129, 2521–2528.

(5) Ye, S.; Li, H.; Yang, W.; Luo, Y. Accurate determination of interfacial protein secondary structure by combining interfacial-sensitive amide I and amide II spectral signals. J. Am. Chem. Soc. 2014, 136, 1206–1209.

(6) Ji, Y.; Yang, X. L.; Ji, Z.; Zhu, L. H.; Tang, J. M. N-methylacetamide Fine Components by NMR to Predict Protein Code Units, under review for publishing.

(7) Fan, L.; Ziegler, T. Application of density functional theory to infrared absorption intensity calculations on transition-metal carbonyls. J. Phys. Chem. 1992, 96, 6937–6941.

(8) Fan, L.; Ziegler, T. Nonlocal density functional theory as a practical tool in calculations on transitionstates and activation energies. Applications to elementary reaction steps in organic chemistry. J. Am. Chem. Soc. 1992, 114, 10890–10897.

(9) Bérces, A.; Dickson, R. M.; Fan, L.; Jacobsen, H.; Swerhone, D.; Ziegler, T. An implementation of the coupled perturbed Kohn-Sham equations: perturbation due to nuclear displacements. Comput. Phys. Commun. 1997, 100, 247–262.

(10) Jacobsen, H.; Bérces, A.; Swerhone, D. P.; Ziegler, T. Analytic second derivatives of molecular energies: a density functional implementation. Comput. Phys. Commun. 1997, 100, 263–276.

(11) Wolfs, S. K. Analytical second derivatives in the Amsterdam density functional package. Int. J. Quantum Chem. 2005, 104, 645–659.

(12) Keiderling, T. A. Vibrational CD of biopolymers. Nature 1986, 322, 85–852.

(13) Hilario, J.; Kubelka, J.; Keiderling, T. A. Optical spectroscopic investigations of model β-sheet hairpins in aqueous solution. J. Am. Chem. Soc. 2003, 125, 7562–7574.

(14) Roitberg, A.; Gerber, R. B.; Elber, R.; Ratner, M. A. Anharmonic wave functions of proteins: quantum self-consistent field calculations of BPTI. Science 1995, 268, 1319–1322.

(15) (a) ADF2016.106; SCM, Theoretical Chemistry, Vrije Universiteit, Amsterdam, The Netherlands, https://www.scm.com; (b) Te Velde, G.; Bickelhaupt, F. M.; Baerends, E. J.; Fonseca Guerra, C.; van Gisbergen, S. J. A.; Snijders, J. G.; Ziegler, T. Chemistry with ADF. J. Comput. Chem. 2001, 22, 931–967. (c) Fonseca Guerra, C.; Snijders, J. G.; Te Velde, G.; Baerends, E. J. Towards an order-N DFT method. Theor. Chem. Acc. 1998, 99, 391–403.

(16) Hayashi, T.; Mukamel, S. Vibrational– Exciton couplings for the amide I, II, III, and A modes of peptides. J. Phys. Chem. B 2007, 111, 11032–11046.

(17) DeFlores, L. P.; Ganim, Z.; Ackley, S. F.; Chung, H. S.; Tokmakoff, A. The anharmonic vibrational potential and relaxation pathways of the amide I and II modes of N-methylacetamide. J. Phys. Chem. B 2006, 110, 18973–18980.

(18) Mikhonin, A. V.; Asher, S. A. Uncoupled Peptide Bond Vibrations in α-Helical and Polyproline II Conformations of Polyanaline Peptides. J. Phys. Chem. B 2005, 109, 3047–3052.

(19) Moran, A.; Mukamel, S. The origin of vibrational mode couplings in various secondary structural motifs of polypeptides. Proc. Natl. Acad. Sci. U. S. A. 2004, 101, 506–510.

(20) Martin, A. H.; Meinders, M. B. J.; Bos, M. A.; Cohen Stuart, M. A.; van Vliet, T. Conformational Aspects of Proteins at the Air/Water Interface Studied by Infrared Reflection–Absorption Spectroscopy. Langmuir 2003, 19, 2922–2928.

(21) Vigano, C.; Mancini, L.; Buyse, F.; Goormaghtigh, E.; Ruysschaert, J. M. Attenuated total reflection IR spectroscopy as a tool to investigate the structure, orientation and tertiary structure changes in peptides and membrane proteins. Biopolymers 2000, 56, 373–380.

(22) (a) Stiopkin, I. V.; Weeraman, C.; Pleniazek, P. A.; Shalhout, F. Y.; Skinner, J. L.; Benderski, A. V. Hydrogen bonding at the water surface revealed by isotopic dilution spectroscopy. Nature 2011, 474, 192–195. (b) Wang, Z.; Carter, J. A.; Lagutchev, A.; Koh, Y. K.; Seong, N.-H.; Cahill, D. G.; Dlott, D. D. Ultrashort Flash Thermal Conductance of Molecular Chains. Science 2007, 317, 787–790.

(23) (a) Ham, P.; Zanni, M. T. Concepts and Methods of 2D Infrared Spectroscopy; Cambridge University Press: NY, 2011; (b) Mukamel, S. Principles of Nonlinear Optical Spectroscopy; Oxford University Press: NY, 1995; (c) Cho, M. Two-Dimensional Optical Spectroscopy; Taylor & Francis Group: Boca Raton, 2009.

(24) Petti, M. K.; Ostrander, J. S.; Saravas, V.; Birdsall, E. R.; Rich, K. L.; Lomont, J. P.; Arnold, M. S.; Zanni, M. T. Enhancing the signal strength of surface sensitive 2D IR spectroscopy. J. Chem. Phys. 2019, 150, No. 024707.

(25) Ghosh, A.; Ostrander, J. S.; Zanni, M. T. Watching Proteins Wiggle: Mapping Structures with Two-Dimensional Infrared Spectroscopy. Chem. Rev. 2017, 117, 10726–10759.