Orientation of Methylguanidinium Ions at the Water–Air Interface

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Supporting Information

ABSTRACT: We use heterodyne-detected vibrational sum-frequency generation (HD-VSFG) to determine the orientation of the molecular plane of methylguanidinium ions at the surface of aqueous solutions. We measure the VSFG response of the symmetric and antisymmetric methyl stretch vibrations of the methylguanidinium ion with different polarization combinations. We find that for at least 50% of the methylguanidinium ions the molecular plane is at an angle >20° with respect to the surface plane. Hence, for only a minor fraction of the ions does the molecular plane have an orientation (near-)parallel to the surface plane, in contrast to the predictions of recent molecular dynamics simulation studies.

1. INTRODUCTION

The guanidinium cation (C(NH$_2$)$_3^+$ or Gdm$^+$) is one of the strongest and most widely used protein denaturants. The mechanism underlying its exceptional protein unfolding capacity has been the subject of many experimental and theoretical studies. The denaturation has been proposed to occur either (i) by indirect interactions mediated by ion-induced changes of the properties of the water solvent or (ii) by direct interaction between the Gdm$^+$ ion and charged or aromatic protein side groups.

The notion of an indirect mechanism strongly relies on the specific nature of the interaction between Gdm$^+$ and water, which has inspired a large number of studies on the structure of such solutions. In particular, molecular dynamics simulations have given much insight into the ion’s fascinating hydration properties. Because of its planar shape and strongly nonuniform charge distribution, the Gdm$^+$ ion exhibits a highly anisotropic hydration structure. The net positive charge of the ion makes the lone pairs of the nitrogen atoms poor hydrogen-bond acceptors. As a result, there are few hydrogen bonds formed pointing perpendicular to the molecular plane, essentially rendering the ion hydrophobic along this molecular axis. The main hydration interactions of the Gdm$^+$ cation are directional hydrogen bonds donated by the N–H groups oriented in the plane of the ion. Consequently, the water solvent is more structured around Gdm$^+$ than around most other ions of similar size.

It has been suggested by several authors that the strong anisotropy of the hydration structure and the amphiphilic nature of the Gdm$^+$ ion are essential for how the ion binds to protein surfaces. While the ion can bind to negatively charged regions by strong in-plane hydrogen bonds, the out-of-plane hydrophobic interaction is thought to align Gdm$^+$ parallel to hydrophobic and aromatic patches. The strong ion–protein interaction resulting from this alignment has been considered to be an important factor in guanidinium’s strong denaturation effect. The out-of-plane hydrophobicity is even so severe that the existence of Gdm$^+$–Gdm$^+$ planar co-ion pairs has been proposed. In both classical and ab initio MD simulation studies, Gdm$^+$ ions have indeed been found to stack. This finding agrees with neutron scattering data. This picture was later further experimentally supported by Shih et al., who showed that the red-shift of the nitrogen K-edge X-ray absorption spectroscopy (XAS) measurements could be theoretically reproduced from structures in which guanidinium ions form stacked co-ion pairs. Using quantum chemical calculations on the CCSD(T) level, Inagaki et al. found that such structures are stabilized through a subtle energy balance partly favorable because of π-stacking interactions and the reduction of hydrophobic effects, while partly unfavorable because of net repulsive Coulomb interactions. Gdm$^+$–Gdm$^+$ co-ion pairing has further been invoked to explain the stabilizing role of arginine–arginine interactions on many protein surfaces.

While the somewhat unintuitive Gdm$^+$ co-ion pairing is becoming more and more accepted, this pairing is not supported by dielectric relaxation spectroscopy (DRS) and conductivity measurements. The anisotropic hydration properties of Gdm$^+$ lead to quite peculiar behavior of the ion near water–air interfaces. Overall, the water surface region shows a net depletion of guanidinium ions, resulting in an increased surface tension. However, recent liquid-jet photoelectron spectroscopy experiments by Werner et al. showed that the concentration of guanidinium ions is enhanced at the surface itself, in the top molecular layer.
layer. MD simulation studies showed that surface-bound Gdm+ ions show a strong preferential orientation parallel to the water surface, allowing for the formation of in-plane hydrogen bonds, while desolvating one of the hydrophobic faces. Similar observations were made in MD simulations where slabs of GdmCl electrolytes were placed between either hydrophobic or hydrophilic plates. The authors found that hydrophobic surfaces induce a stronger orientational parallel ordering of Gdm+ ions than hydrophilic surfaces, thus further illustrating how the behavior of the ion at interfaces is driven by its anisotropic amphiphilic nature.

Recently, in a MD simulation study by Ou et al. the surface hydration properties of guanidinium were compared to those of methylguanimidium (M-Gdm+, a derivative of Gdm+). In particular, the authors studied to what extent solute-induced interfacial solvent density fluctuations can explain the surface propensity of various orientational configurations of the solute, a topic which recently has attained significant attention for rationalizing the varying surface propensity of simple ions. The net surface activity of M-Gdm+ and Gdm+ was somewhat different, they found that both ions have primarily parallel-oriented configurations at the water surface.

In this work, we use heterodyne-detected vibrational sum frequency generation (HD-VSFG) to study the orientation of M-Gdm+ ions at the water–air interface. HD-VSFG allows for the measurement of the absolute orientation of molecules at interfaces. The point group symmetry of the Gdm+ ion is D3h, which implies that the ion possesses degenerate N–H vibrational modes of $E'$ symmetry that should be VSFG active. However, the N–H stretch vibrations spectrally overlap with the relatively strong signal of the O–H stretch vibrations of water, thus making an analysis of these modes difficult. For M-Gdm+, the methyl stretch vibrations can be clearly distinguished from the response of the water solvent, thus making this ion a much more suitable system to determine its orientation at the surface. By measuring the HD-VSFG response of the symmetric and antisymmetric stretch vibrations of the methyl group of M-Gdm+ with different polarization combinations, we determine the orientation of the methyl group and thereby of the M-Gdm+ ion at the water–air interface.

2. EXPERIMENT

The details of the HD-VSFG setup have been reported before. Briefly, part of the pulses produced by a Ti:sapphire regeneratively amplified laser system (Coherent Legend, 1 kHz, ∼35 fs, ∼3.5 mJ) is used to produce broadband infrared (IR) pulses (tunable from 2 to 10 μm) with a home-built optical parametric amplifier. The remaining part of the 800 nm fundamental is spectrally narrowed using an etalon (bandwidth ∼15–20 cm−1) and is spatially and temporally overlapped with the broadband IR pulse to generate sum-frequency light from the surface of a local oscillator, in this case a gold mirror (Thorlabs model PF 10-03-M01). The generated LO-SFG light is then delayed in time using a silica plate and is overlapped with the broadband IR pulse to generate sum-frequency second harmonic generation (SFG) light from the surface of a local oscillator, in this case quartz in the 3000 cm−1 region. It is also crucial that the HD-VSFG signal obtained from z-cut quartz is generated at the same height as the HD-VSFG signal from the sample, as height differences would introduce phase errors. The height can be controlled by monitoring the location of the signal on the CCD camera, leading to an estimated phase uncertainty of ∼π/10 (~2°).

The samples (3 M methylguanidine hydrochloride) were prepared from mixing D2O (99.96%, Cambridge Isotope Laboratories) and methylguanidine hydrochloride (98%, Sigma-Aldrich).

3. THEORY: VSFG SIGNAL OF A METHYL GROUP

3.1. Relation between the VSFG Signal and the Molecular Hyperpolarizability. To determine the orientation of molecular groups at interfaces with VSFG, we need to relate the effective nonlinear susceptibility $χ^{(2)}_{\text{eff}}$ to the microscopic molecular hyperpolarizability $\beta_{\chi/X}$ tensor elements. This relation can be found in the literature. Here we present this relation for the $C_3$ symmetry molecular group at rotationally isotropic achiral interfaces ($C_{\infty v}$).

In VSFG spectroscopy the generated electric field at the sum-frequency is proportional to the electric fields of the incident IR and VIS beams and the effective second-order nonlinear susceptibility $χ^{(2)}_{\text{eff}}$:

$$E_{\text{SFG}}(\omega) = \frac{\chi^{(2)}_{\text{eff}}(\omega)E_{\text{VIS}}(\omega_1)E_{\text{IR}}(\omega_2)}{E_{\text{VIS}}(\omega_1) + E_{\text{IR}}(\omega_2)}$$

where $\omega_1$, $\omega_2$, and $\omega_3$ are the frequencies of sum-frequency, VIS, and IR, respectively. $E_{\text{SFG}}(\omega)$, $E_{\text{VIS}}(\omega_1)$, and $E_{\text{IR}}(\omega_2)$ are the strengths of those respective electric fields. We define the laboratory coordinates with the z-axis as the surface normal and with x and y forming the rotationally symmetric surface plane. All beams propagate in the $x,z$ plane; thus, p-polarization denotes the polarization of the optical field in the $x,z$ plane, while s-polarization is along the y-axis, perpendicular to the $x,z$ plane.

In total, the $χ^{(2)}_{\text{eff}}$ tensor can have 27 elements, but because of symmetry considerations interfaces with $C_{\infty v}$ symmetry (achiral rotationally isotropic) have only 7 nonzero tensor elements, of which only 4 are independent; $χ^{(2)}_{\text{xxz}} = χ^{(2)}_{\text{yyz}}$ and $χ^{(2)}_{\text{zxy}} = χ^{(2)}_{\text{zyx}}$. These four components can be deduced by measuring SFG intensities with different input and output polarization combinations, such as SSP, SPS, and PPP, where the

DOI: 10.1021/acs.jpcc.7b03752

J. Phys. Chem. C 2017, 121, 23398–23405
respective positions refer to the polarization of the contributing SFG, VIS, and IR beams, respectively. SSG polarization probes only one tensor element $\chi^{(2)}_{ss}$, whereas PPP probes combination of all four independent tensor elements. $\chi^{(2)}_{kk}$ for those four polarization combinations are related to all the nonzero seven $\chi^{(2)}_{jk}$ tensor elements:

$$\chi^{(2)}_{kk} = L_{yy}(a)L_{yy}(w)L_{zz}(w)\sin\eta \gamma^{(2)}$$

where $\omega$ and $Q_n$ are the resonant vibrational frequency and the normal coordinates of $q$th vibrational mode and $\epsilon_0$ denotes vacuum permittivity. The macroscopic susceptibility $\chi^{(2)}_{kk}$ is an ensemble average of the microscopic hyperpolarizabilities and can be written as

$$\chi^{(2)}_{kk} = \frac{N_s}{\epsilon_0} \sum_{i j k} \langle R_{ik} R_{jk} \rangle \beta^{(2)}_{i j k}$$

where $N_s$ is the number density of the molecules probed at the interface. $R_{ij}$ is the element of the rotational transformation matrix used to convert from the coordinates $(a, b, c)$ to surface bound coordinates $(x, y, z)$. The $\chi^{(2)}_{kk}$ and $\beta^{(2)}_{i j k}$ can be connected using the symmetry of the molecular vibration and performing an Euler transformation for all three angles $(\theta, \phi, \psi)$. For the methyl group with $C_3v$ symmetry, we naturally choose the $c$-axis along the symmetry axis of the $C_3$ methyl group, and the $a$ and $b$ axes form a plane perpendicular to the $c$-axis. The $a$-axis is along one of the C–H bonds (see Figure 1b). There are 11 nonzero molecular hyperpolarizability elements for the methyl group (of which 4 are independent); 3 for the symmetric vibration $(\beta_{aa} = \beta_{bb} = \beta_{cc})$ and 8 for the antisymmetric vibration $(\beta_{aa} = \beta_{bb} = \beta_{cc} = -\beta_{ba} = -\beta_{bb} = -\beta_{cc})$.39 We have for the symmetric vibration

$$\chi^{(2),as}_{xx} = \frac{1}{2} N \beta_{xx} \langle \cos \theta \rangle [(1 + R) - (1 - R)D]$$

and for the antisymmetric vibration

$$\chi^{(2),as}_{xx} = N \beta_{xx} \langle \cos \theta \rangle [R - (1 - R)D]$$

where $\theta$ is the tilt angle between the methyl $C_3$ axis and the surface normal $z$ and $R$ is the ratio of the hyperpolarizability elements, $R = \beta_{aa}/\beta_{cc}$. $D$ is an orientational parameter, defined as

$$D = \frac{\langle \cos^2 \theta \rangle}{\langle \cos \theta \rangle}$$

The second-order susceptibilities are determined by the microscopic hyperpolarizabilities $\beta^{(2)}_{i j k}$ $(i'j'k = a, b, c$; molecular coordinates, see Figure 1b). From a quantum mechanical treatment, using perturbation theory of the electron wave function, it can be shown that hyperpolarizability tensor elements of a particular vibrational mode are proportional to the Raman polarizability $\alpha_{i j}$ and dipole moment $\mu_L$ derivative tensor elements:

$$\beta^{(2)}_{i j k} = -\frac{1}{2\epsilon_0 a^2} \frac{d\alpha_{i j}}{dQ_{i}^q} \frac{d\mu_L}{dQ_{q}}$$

Figure 1. (a) Geometry of our VSFG experiment in a Cartesian lab coordinate system. The incident and emitted beams propagate in the $x_z$ plane. (b) Molecular coordinates $(a, b, c)$ and laboratory coordinates $(x, y, z)$, relevant for the methyl group at water surfaces. The angle $\theta$ denotes the tilt angle between the methyl $C_3$ main symmetry axis $(c$-axis) and the surface normal $z$. 

DOI: 10.1021/acs.jpcc.7b03752

J. Phys. Chem. C 2017, 121, 23398–23405

23400
If one assumes that the orientation distribution function is a δ-function, the tilt angle (θ₀) is

\[ \theta_0 = \arccos(\sqrt{D}) \]  

(9)

It follows from from eqs 6 and 7 that the ratios of the signals measured for different vibrations, for example the symmetric and the antisymmetric vibration, depend on only θ, via D. Hence, D is the crucial parameter to be obtained from the VSFG measurements to determine the molecular orientation of the methyl group at the water surface.

3.2. Dependence of the VSFG Signal on the Orientation Angle. The formalism presented in this section provides the relation between the effective second-order susceptibilities \( \chi''(2) \) measured with HD-VSFG with different polarization combinations and the orientation of the CH₃ group relative to the water surface. To make such a comparison several optical parameters of the system need to be determined.

The Fresnel coefficients depend on the refractive indices of the sum-frequency, the 800 nm and the mid-infrared light at ∼2900 cm⁻¹. The refractive indices of the 800 nm and the sum-frequency (∼645 nm) beams show very little change with frequency and solute concentration, as neither D₂O nor methylguanidinium possesses resonances in the visible. For both the 800 nm beam and the sum-frequency beam we use a refractive index of 1.47, as reported by Sigma Aldrich for the refractive index of 1.48 at the mid-infrared frequency around 2900 cm⁻¹. We neglect the dispersion of the refractive indexes and use the same values for all frequencies within the VSFG spectrum.

Hyperpolarizability \( \beta_{ffe} \) tensor elements \( \beta_{aat} \beta_{ait} \beta_{cct} \) can be calculated by the so-called bond additivity method, also called the bond polarizability derivative method, which was first derived by Hirose et al.⁴²,⁴³ From this formalism follows that 4 > R > 1. The exact value of R depends on the molecule to which the methyl group belongs. More recently, ab initio calculations have been employed to determine hyperpolarizability tensor elements.⁴⁴ We determine R from the fit of the experimental data.

Figure 2 shows calculated \( \chi''(2) \) values for the symmetric (SS) and antisymmetric (AS) methyl stretch vibrations in both SSP and PPP polarization combinations, plotted as a function of the angle θ (see Figure S1 in Supporting Information for SPS polarization combination) assuming a δ-distribution function for θ. It is important to note that for the SS mode the calculated intensity has \( \beta_{cct} \) as a unit, and the AS mode has \( \beta_{aat} \) as a unit. Hence, the scales of graphs in Figure 2a,b cannot be compared directly, unless the absolute values of hyperpolarizability tensor elements are known. The ratio \( \beta_{aat}/\beta_{cct} \) can acquire quite different values depending on the system studied. For instance, \( \beta_{aat}/\beta_{cct} \) was found to be 0.4 for methanol and 4.5 for ethanol.⁴⁴ Hence, we will use this ratio as a fit parameter S (=\( \beta_{aat}/\beta_{cct} \)) in describing the experimental spectra.

It follows from Figure 2 that for the symmetric mode, the imaginary \( \chi''(2) \) measured with SSP is always at least ~1.5 times higher than the imaginary \( \chi''(2) \) measured with PPP and that the sign is opposite for the two polarization combinations. For the antisymmetric mode the intensity is larger in PPP than in SSP (~2 times) and the sign is also opposite in the two polarization combinations.

4. RESULTS AND DISCUSSION

In Figure 3 we show imaginary \( \chi''(2) \) spectra of 3 M methylguanidinium hydrochloride solutions measured with SSP and PPP polarization combinations. As M-Gdm⁺ possesses a single methyl group, there will be three modes contributing to the signal in this frequency region: the symmetric stretch vibration (SS), the Fermi resonance (FR), and the antisymmetric stretch vibration (AS). The Fermi resonance results from the interaction between the symmetric methyl vibration and the overtone of the bending mode (∼1460 cm⁻¹). Following previous work, we label the band at 2922 cm⁻¹ as...
the Fermi resonance (FR) band. However, both bands (SS and FR) contain symmetric stretch vibrational character, and this character determines the VSFG cross section of these bands. The band at 2863 cm\(^{-1}\) is the lower-frequency band of the Fermi split symmetric stretch, and the band at 2922 cm\(^{-1}\) is the higher-frequency band of the Fermi split symmetric stretch. It follows that both bands have the same symmetry and that the total amplitude of the symmetric methyl stretch vibration is represented by the sum of the amplitudes of the two bands. The sign of the SS and the FR bands thus shows the same dependence on the polarization combination. The peak at 2863 cm\(^{-1}\) shows opposite behavior, and we assign this band to the AS mode.

Using the dependence of the VSFG signal of the different vibrations on the orientational angle (illustrated in Figure 2), we can now fit the experimental data (Figure 3). In this fit we determine the center frequencies and widths of the AS, FR, and SS peaks, the \( R \) and \( S \) parameters describing the hyperpolarizability ratios \( R = \beta_{\text{as}} / \beta_{\text{is}}, S = \beta_{\text{as}} / \beta_{\text{is}} \), the parameter \( D \) that describes the orientation of the methyl group, a parameter that defines the ratio of the FR and SS peaks and an overall scaling parameter. The relative peak intensities of the SS and AS bands in the two polarization combinations are described by the \( R \) and \( D \) parameters. The FR bands have the same ratio as the SS bands in the two polarization combinations. The relative peak intensities of the AS bands in the two polarization combinations is determined only by the \( D \) parameter. Within each polarization combination, the ratio of SS and FR with respect to AS is determined by the \( S \) parameter.

The resulting fit parameters are given in Table 1, and the result of the fit is plotted together with the experimental data in Figure 3. For \( R \) we obtain a value of 1.0 ± 0.1, and for \( S \) a value of 1.1 ± 0.1, which implies that the three hyperpolarizability tensor elements \( \beta_{\text{as}}, \beta_{\text{is}}, \) and \( \beta_{\text{is}} \) would be quite similar in size for the M-Gdm\(^+\) ion. For \( D \) we find a value of 0.5 ± 0.06.

The orientation dependence of the VSFG signals of the three different modes in the SSP and PPP experiments is expressed in the value of \( D \). It should be realized that the same value of \( D \) can result from different angular distributions. To explore the range of possible angular distributions, we define the distribution as a Gaussian function with central angle \( \theta_c \) which has zero amplitude for \( \theta > 90^\circ \). This distribution can be rationalized by considering the hydrophobic nature of the methyl group, making it unlikely that surface-bound M-Gdm\(^+\) would orient such that the methyl group would be pointing toward the bulk aqueous phase, i.e. \( \theta > 90^\circ \). We will refer to such a distribution as a partial-Gaussian, defined as

\[
F_{\text{PG}}(\theta) = \begin{cases} 
\exp[-4\ln 2(\theta - \theta_c)^2 / \sigma^2] & \text{if } 0^\circ < \theta < 90^\circ \\
0 & \text{if } 90^\circ < \theta < 180^\circ 
\end{cases} 
\]

\( D \) can be evaluated for this distribution using

\[
D = \frac{\int_0^{90} \cos \theta \sin \theta F_{\text{PG}}(\theta) d\theta}{\int_0^{90} \cos \theta \sin \theta F_{\text{PG}}(\theta) d\theta} 
\]

In Figure 4a the orientational parameter \( D \) (calculated from eq 11) is plotted as a function of the center angle \( \theta_c \) for three different widths \( \sigma \) (20\(^\circ\), 50\(^\circ\), and 90\(^\circ\)) of partial-Gaussian distribution function. In Figure 4b, \( D \) is shown as a function of the width \( \sigma \) for \( \theta_c \) equal to 45\(^\circ\) and 90\(^\circ\). In case the angular distribution would be a \( \delta \)-function (zero width), the \( D \) value of 0.5 ± 0.06 would correspond to a molecular tilt angle \( \theta_c \) of 45\(^\circ\) ± 2\(^\circ\). For \( \theta_c = 90^\circ \), a minimum width of 150\(^\circ\) is required to obtain the experimentally determined \( D \) value of 0.5 ± 0.06. Figure 4c illustrates the distribution with \( \theta_c = 90^\circ \), a width of 150\(^\circ\), and a \( \delta \)-distribution at \( \theta = 45^\circ \). In the Supporting Information we compare calculations for a partial-Gaussian distribution with a full-Gaussian distribution (no truncation at \( \theta = 90^\circ \) in eq 10; see Figure S3).

**Table 1. Parameters Determined from a Global Fit of the SSP and PPP Spectra of M-Gdm\(^+\)**

| Parameter | Value                  |
|-----------|------------------------|
| \( R \)   | 1.0 ± 0.1              |
| \( S \)   | 1.1 ± 0.1              |
| \( D \)   | 0.5 ± 0.06             |
| SS        | \( \omega_{\text{SS}} \)   | 2863 cm\(^{-1}\) |
|           | \( \sigma_{\text{SS}} \) | 23 cm\(^{-1}\)   |
| FR        | \( \omega_{\text{FR}} \) | 2922 cm\(^{-1}\) |
|           | \( \sigma_{\text{FR}} \) | 23 cm\(^{-1}\)   |
| AS        | \( \omega_{\text{AS}} \) | 2950 cm\(^{-1}\) |
|           | \( \sigma_{\text{AS}} \) | 21 cm\(^{-1}\)   |
The orientation of Gdm$^+$ and M-Gdm$^+$ cations at the water−air interface has been investigated by Ou et al. using MD simulations.24,27 These MD simulations find the most probable surface-bound configuration to be parallel to the surface flat, which can be described with a partial-Gaussian with center angle $\theta_c = 90^\circ$. Interestingly, further toward the vapor phase, the simulations of M-Gdm$^+$ show that the ion has a tendency to orient with its methyl group pointing away from the liquid phase, corresponding to configurations with tilt angles $\theta$ close to $0^\circ$. The density profiles reported by Ou et al. further suggest that the relative contribution from M-Gdm$^+$ in the vapor phase is rather small,27 which corresponds to a partial-Gaussian with center angle $\theta_c = 90^\circ$ and a relatively narrow width.

The experimentally observed $D$ can be reproduced with a partial-Gaussian distribution with center angle $\theta_c = 90^\circ$ in case this distribution has a large width of $\sim 150^\circ$. This width implies that $\sim 80\%$ of the M-Gdm$^+$ ions are oriented at an angle $>20^\circ$ with respect to the surface plane. For all possible center angle $\theta_c$ and width $\sigma$ combinations yielding $D = 0.5$, the fraction of molecules that are at an angle $>20^\circ$ with respect to the surface plane is at least $\sim 50\%$. This notion applies both to the partial-Gaussian and a full-Gaussian distributions (see the Supporting Information). Hence, the VSFG results show that M-Gdm$^+$ ions at the water−air interface are oriented less parallel to the water−air interface than has been predicted by MD simulations.27

5. CONCLUSIONS

We presented a heterodyne-detected vibrational surface sum-frequency generation study of the orientation of methylguanidinium ions at the water−air interface. To this purpose, we measured HD-VSFG spectra of the symmetric and antisymmetric methyl stretch vibrations of the ion in SSP and PPP polarization combinations. From these spectra we obtained the ratios of the second-order susceptibility tensor elements that provide information on the molecular orientation of the probed methyl group. Assuming a $\delta$-distribution for the orientation angle, we find that the observed spectra can be well explained if the $C_3$ axis of the methyl group of methylguanidinium is at an angle of $\sim 45^\circ$ with respect to the surface normal. Assuming a partial-Gaussian orientational distribution with its maximum at $90^\circ$ (=parallel to the surface plane), we find that the spectra can be explained only if this distribution has a width of $\sim 150^\circ$. From this, we conclude that for $\sim 80\%$ of the methylguanidinium ions the molecular plane is at an angle $>20^\circ$ with respect to the surface plane, which implies that only a minor fraction of the ions have an orientation (near-)parallel to the water surface. We find that at least $50\%$ of methylguanidinium ions are oriented at an angle $>20^\circ$ with respect to the surface plane.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.jpcc.7b03752. Details of fitting SSP and PPP spectra of methylguanidinium; calculated $\chi^{(2)}$ versus orientational angle $\theta$ for symmetric and antisymmetric methyl vibration in SSP, SPS, and PPP polarization combinations (Figure S1); deviations of the fitted SSP and PPP spectra if $R$ or $D$ changes within the range of errors, determined from fitting (Figure S2); comparison of partial and full-
Gaussian distributions for dependence of $D$ on the angle $\theta$, of the maximum of the distribution for three different widths, width of the distribution required to get a $D$ value of 0.5 as a function of $\theta$, and the fraction of molecules with their main axis at an angle $\theta > 20^\circ$ with respect to the surface plane for different values of $\theta_c$ (Figure S3)(PDF)

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**Notes**

The authors declare no competing financial interest.

**ACKNOWLEDGMENTS**

This work is part of the research program of the “Stichting voor Fundamenteel Onderzoek der Materie (FOM)”, which is financially supported by the “Nederlandse organisatie voor Wetenschappelijk Onderzoek (NWO)”. The work was performed within the framework of a FOM Industrial Partnership Program with Top-institute Wetsus for water research and is also financially supported by Wetsus.

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