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Propofol adsorption at the air/water interface: a combined vibrational sum frequency spectroscopy, nuclear magnetic resonance and neutron reflectometry study.

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Propofol is an amphiphilic small molecule that strongly influences the function of cell membranes, yet data regarding interfacial properties of propofol remain scarce. Here we consider propofol adsorption at the air/water interface as elucidated by means of vibrational sum frequency spectroscopy (VSFS), neutron reflectometry (NR), and surface tensiometry. VSFS data show that propofol adsorbed at the air/water interface interacts with water strongly in terms of hydrogen bonding and weakly in the proximity of the hydrocarbon parts of the molecule. In the concentration range studied there is almost no change in the orientation adopted at the interface. Data from NR show that propofol forms a dense monolayer with a thickness of 8.4 Å and a limiting area per molecule of 40 Å², close to the value extracted from surface tensiometry. The possibility that islands or multilayers of propofol form at the air/water interface is therefore excluded as long as the solubility limit is not exceeded. Additionally, nuclear magnetic resonance (NMR) measurements of the ¹H nuclear magnetic resonance chemical shifts demonstrate that propofol does not form dimers or multimers in bulk water up to the solubility limit.

Introduction

Alcohol/water mixtures are widely used as industrial solvents and chemical reagents, and the need of a better understanding of the interfacial behaviour of aqueous alcohol solutions at different interfaces has led to increasing interest in fundamental research.^{1, 2} The behaviour of alcohols at

solid interfaces plays a key role in applications like cleaning, etching, and electrochemical reactions.^{3, 4} Let alone the industrial applications, the properties of alcohols at buried interfaces have important implications in cell membrane function. The functioning of intrinsic membrane proteins can be altered by introduction of short chain alcohols (e.g. propofol) at the membrane surface.^{5, 6} As a result, propofol has, for example, been shown to induce a significant change in membrane permeability.⁷ This is potentially important since low molecular weight anaesthetics are used daily in hospitals, but yet the molecular mechanism of general anaesthesia remains to some extent ambiguous.⁸

In spite of the importance of interfacial properties of alcohol/water mixtures in a wide variety of applications^{9, 10} the study of such interfaces remain complex, mainly due to their dynamic nature and the lack of surface-specific experimental techniques. The development of nonlinear optical methods has contributed to overcoming some of these problems and facilitated better molecular understanding of such

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interfaces. Specifically, vibrational sum frequency spectroscopy (VSFS) has proven to be a powerful technique for studying a wide variety of aqueous interfaces¹¹ due to its capability of yielding surface-specific information at a molecular level.

Different types of alcohols have been studied using VSFS with respect to the stretching vibrations of the hydrocarbon chains,¹¹⁻¹⁴ the fingerprint region¹⁵ and the water of hydration.¹⁶⁻²⁰ It was found that for a series of increasing chain length alcohols (C1–C8), at the neat air/alcohol interface, the tails point out into the gas phase due to the amphiphilic character of alcohols, and that the presence of gauche defects is chain length dependent. At the same time a very well ordered interfacial hydrogen bonding network was detected.²¹ Short chain alcohols (C1–C3) do not affect the interfacial hydrogen bonding structure and orientation to a large extent, while long chain alcohols (C5–C12) make the interfacial water more strongly hydrogen bonded and reversely oriented.²⁰ In mixtures with water, glycerol was found to partition to the interface with air, whereby disturbing the topmost water layers.¹⁸ At a hydrophobic solid surface, methanol adsorbed from its mixture with water to the interface with the C–O bond aligned to the surface normal while the water band at 3200 cm⁻¹ weakened in strength with increasing methanol concentration.¹⁶

The knowledge of small hydrophobic but weakly amphiphilic drugs at the interface is rather limited compared with the more amphiphilic molecules. Amphiphilic molecules such as surfactants²², proteins²³, block co-polymers²⁴ and peptides²⁵ have been the subject of extensive investigations. By contrast there is a lack of body of work on the surface properties of small hydrophobic drugs. For example, for ibuprofen there has been a study on its interactions with lipid bilayers²⁶ and cholesterol²⁷ but to our knowledge not on its adsorption properties at the air/water interface. A recent study looked at the interactions of testosterone enanthate with surfactants at the air/water interface²⁸, but again to our knowledge the surface properties of the drug alone have not been examined explicitly. Further, for propofol the present authors have conducted a study on its interactions with phospholipids monolayers²⁹ but an analogous study of its adsorption properties at the air/water interface is missing. To address this shortcoming, we have decided to study its adsorption at the air/water interface.

Propofol is a small alcohol that contains both hydrophobic and hydrophilic parts and has interesting biochemical properties, e.g., it is used commonly in

general anaesthesia to induce a state of reduced consciousness in patients during medical procedures.³⁰⁻³² Similar in structure to propofol, phenol has been briefly characterised by VSFS at the air/water interface,¹⁵ by focusing on only the carbon–oxygen stretching region of the spectrum. It was found that phenol is present at the interface at both low and high pH, and at high pH there are both phenol and phenolate ions. Surprisingly however, according to our knowledge, there are no detailed studies of propofol at the air/water or water/hydrophobic interface despite its fundamental interest as a small amphiphilic drug with important biomedical applications.

In the present work we evaluate the adsorption isotherm from surface tension data, and the adsorbed amount is also determined by neutron reflectivity (NR) measurements. NR and VSFS data are then combined to gain structural information on the adsorbed layer and its interaction with water. We also report a bulk study of the chemical shifts of hydrogen nuclei over the entire solubility range of propofol in water in order to detect possible multimeric structures. The results represent a necessary first step in resolving the driving forces for the interactions of propofol with interfaces, and they set the stage for future studies involving its interactions in more complex systems (including supported lipid bilayers^{33, 34} and membrane proteins^{35, 36}) that are closer in nature to those in practical applications of the drug.

Materials and Methods

Materials

Propofol (European Pharmacopoeia) with purity higher than 99% was bought from Sigma Aldrich. Its chemical structure is presented in Figure 1, where also the NMR chemical shifts are listed. A Millipore Milli-Q Plus system was used as water supply, providing purified water with a resistivity of 18.2 MΩ cm.

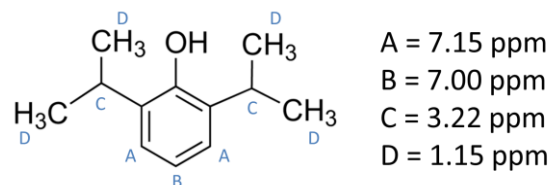


Figure 1. The chemical structure of propofol together with the ¹H NMR peak assignments.

Nuclear Magnetic Resonance

^1H NMR spectra of propofol dissolved in D_2O were obtained for a concentration series on a Bruker Advance III 500 MHz spectrometer using a 5 mm Bruker DIFF 30 probe. All experiments were performed at 293 K and the chemical shifts of the different propofol peaks were recorded; as spectral reference, we used the ^1HDO signal (set to 4.75 ppm).

Surface Tensiometry

A Du Noüy ring instrument (Krüss) was used to determine the surface tension of aqueous propofol solutions. An independent solution was prepared for each concentration. To estimate the surface excess from the surface tension measurements, we used the Gibbs equation assuming only a single adsorbing neutral species, as shown in the equation below.

$$\Gamma_{\text{ST}} = -\frac{1}{RT} \frac{d\gamma}{d\ln c} \quad (1)$$

where Γ_{ST} is the surface excess as measured by the surface tension method, γ is the surface tension, c is the bulk concentration, R is the gas constant, and T is the absolute temperature. Since the NMR data reported below do not show any evidence for dimerisation, eq. 1, where the concentration is used instead of the activity, is judged to be a good approximation up to the solubility limit.

The solutions were prepared by dropwise adding liquid propofol to water, then gently shaking and hand warming for a few minutes until the propofol was dissolved. At least 10 min was allowed before the measurements were recorded. For higher concentrations it took longer time for propofol to completely dissolve.

Neutron Reflectometry (NR)

NR measurements were performed using the time-of-flight reflectometer FIGARO at the Institut Laue-Langevin (Grenoble, France)³⁷. The neutron reflectometry determines the ratio of the number of neutrons in the specular reflection to those in the incident beam with respect to the momentum transfer, q_z ,

$$q_z = \frac{4\pi \sin \theta}{\lambda} \quad (2)$$

where λ is the neutron wavelength and θ is the angle of incidence³⁸. Data were recorded with 7 % $\delta\lambda/\lambda$

using a frame overlap mirror of 16 Å. The surface excess of propofol at the air/water interface at six different bulk concentrations was recorded at $\theta = 0.623^\circ$ for 15 min, where the solvent was air contrast matched water (ACMW), which is 8.1 % by volume D_2O in H_2O . The surface excess, Γ_{NR} (in mol m^{-2}), as determined by neutron reflection was calculated using:

$$\Gamma_{\text{NR}} = \frac{\rho \cdot d}{N_A \cdot \sum b_i} \quad (3)$$

where ρ is the scattering length density of propofol, d the fitted layer thickness, N_A is Avogadro's number and $\sum b_i$ is the scattering length of propofol. Note that the scattering length density of propofol was calculated as $6.2 \times 10^{-7} \text{ Å}^{-2}$ according to its molecular volume of 285 Å^3 (which is calculated from bulk density).

Two different data acquisition approaches were used to determine: (1) the adsorption isotherm and (2) the interfacial structure of the adsorbed molecules at the air/water interface, following the methodology adopted in ref.³⁹ In the first case, measurements were made only in ACMW, and the data were reduced only over 4.5–12 Å to restrict the q_z -range to 0.01–0.03 Å^{-1} . This approach reduces the sensitivity of the analysis to details of the structure at the interface.⁴⁰ Note that the surface excess of propofol could be resolved accurately even though it was used in its normal, non-deuterated form thanks to the high flux at low- q_z of FIGARO. The interfacial roughness values were fixed to 3.1 Å, consistent with the outcome of the structural analysis (note that for this low- q_z analysis even neglect of the roughness values resulted in a change in surface excess of < 0.5 %). The background was not subtracted from these data in this analysis, but its accurate determination is essential to the precise quantification of the scattering excess: its value was iterated from the lowest value possible, where a layer of zero surface excess could be fitted to a measurement of pure ACMW.⁴¹

In the second case, for the structural analysis, data were recorded at a propofol concentration of 0.89 mM over a broader q_z -range with measurements at $\theta = 0.623^\circ$ and $\theta = 3.78^\circ$ both in ACMW and D_2O . The background was subtracted from these data with the use of the 2D detector on the instrument. A single-layer structural model adequately described the data. In this case the fitting parameters were the thickness of the layer and its volume fraction. The roughness values of the air/propofol and propofol/water interfaces were constrained to be equal to each other

and consistent with capillary wave theory, following an approach described recently.⁴² The data analysis was performed exclusively using Motofit.⁴³

Vibrational Sum Frequency Spectroscopy

The experimental VSF spectrometer has been described previously⁴⁴. A 1064 nm laser beam generated from a Nd:YAG laser system (PL-2251A-20, Ekspla) is used to pump the optical parameter generator/optical parameter amplifier OPG/OPA (LaserVision) system, which produces a fixed visible beam (532 nm) and a tuneable IR beam (1000–4000 cm⁻¹). These two laser beams overlap at the sample surface with incident angles of 55° and 63° from the surface normal for the visible and IR beams, respectively. The VSF beam is collected and filtered both optically and spatially before being sent to a photomultiplier tube (Jobin Yvon). The signal is integrated in a boxcar and finally processed by a computer program.

The theory behind VSFS is well described in the literature.⁴⁵⁻⁴⁹ It is a nonlinear optical technique, which is able to provide surface specific information such as the nature and orientation of the species present at an interface. The intensity of the collected beam is proportional to the intensities of the incoming visible and infrared beams and the square of the effective second order nonlinear susceptibility $\chi^{(2)}_{\text{eff}}$. $\chi^{(2)}_{\text{eff}}$ is in turn proportional to the number density multiplied by the orientationally averaged molecular hyperpolarisability $\langle\beta^{(2)}\rangle$ of the probed species. We note that in order for a molecule or part of a molecule to be VSF active it has to be both IR and Raman active, according to Eq. (4):

$$\beta_{\alpha\beta\gamma}^{(2)} = \frac{\alpha_{\alpha\beta}\mu_{\gamma}}{\omega_n - \omega_{\text{IR}} - i\Gamma_n} \quad (4)$$

where α , β , and γ are the molecular coordinates, $\alpha_{\alpha\beta}$ is the Raman polarisability tensor, μ_{γ} the transition IR dipole moment, ω_n the peak position frequency, ω_{IR} the irradiating infrared frequency, Γ_n the damping constant of the n^{th} resonant mode and i the imaginary unit. The transformation of the molecular hyperpolarisability coordinates into laboratory coordinates is done using an Euler transformation matrix.⁵⁰ The obtained VSF spectra were fitted using a Lorentzian line profile using an IgorPro (WaveMetrics) program:

$$I_{\text{VSF}} = \left| A_{\text{NR}} + \sum_n \frac{A_n}{\omega_n - \omega_{\text{IR}} - i\Gamma_n} \right|^2 \quad (5)$$

where I_{VSF} is the recorded VSF signal, A_{NR} is the non-resonant amplitude of the VSF signal and A_n is the amplitude (oscillator strength) of the n^{th} resonant mode.

Orientational information about the probed molecular species is given by elements of the second order nonlinear susceptibility tensor, specifically by using different polarisation combinations. In this study we have used SSP, PPP, and SPS, where P refers to light polarised parallel to the plane of incidence and S refers to light polarised perpendicular to the plane of incidence. The first letter corresponds to the polarisation of the sum frequency beam, the second to that of the visible beam, and the third to the infrared beam.

Results and discussion

Propofol in Aqueous Bulk Solution

The ^1H NMR spectrum of propofol has been published before,^{51, 52} and therefore it is not shown here. As is clear from Figure 2, the chemical shifts of the hydrogen atoms in propofol remain constant in the concentration range 0.02–0.89 mM. Since propofol contains an aromatic moiety with large resulting “ring current” effects, any aggregation should have a large effect on the observed shifts.^{53, 54}

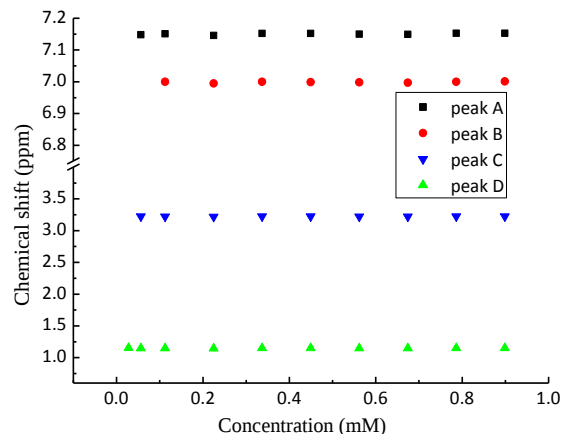


Figure 2. The chemical shifts (see Figure 1 for peak assignment) recorded in solution of propofol at different concentrations between 0.02 mM and 0.89 mM. A – aromatic H3 and H5, B – aromatic H4, C – CH(CH₃)₂, and D – CH₃.

The absence of such effects points to the absence of any significant aggregation/dimerisation phenomena. This is in contradiction to what has been inferred in gas-phase where “nano-micelles” containing several multimers of propofol were found.⁵⁵ In addition, the NMR intensity was proportional to the concentration in the whole range

which also excludes the presence of any large aggregates (for which the NMR signal could possibly be lost).

Propofol Adsorption Isotherm

In order to gain insight into the adsorption process of propofol at the air/water interface we have combined information from surface tensiometry, NR and VSFS.

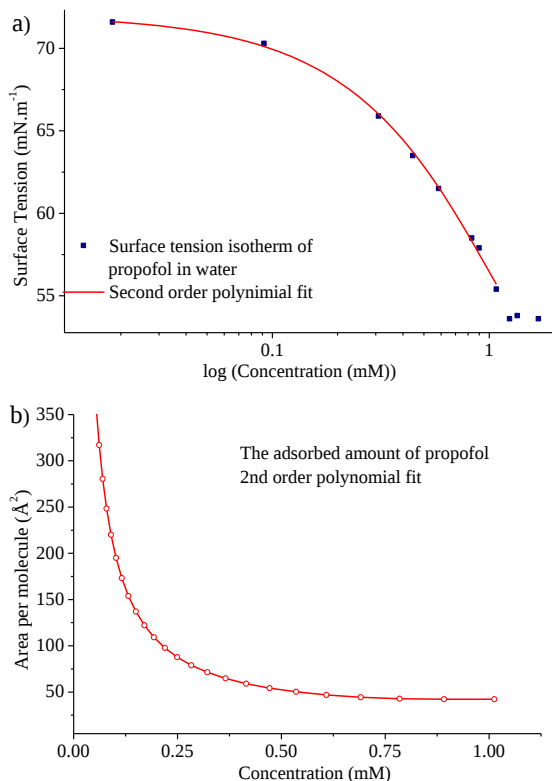


Figure 3. a) Surface tension isotherm of propofol and a second order polynomial fit to the data (line), and b) area per molecule calculated using the second order polynomial fit and eq. 1.

Figure 3a shows the surface tension isotherm of aqueous propofol solutions. The decrease in the surface tension is due to adsorption of propofol at the air/water interface. The surface tension drops smoothly with concentration up to a concentration of about 1 mM, above which it remains constant. The limiting area per molecule is about 42 Å², as seen in Figure 3b. A discussion regarding the structure of the adsorbed propofol layer will follow in the light of the NR results presented next. Details on area per molecules calculation are given in Supporting Information.

The surface excess was measured directly at six different bulk concentrations at low q_z values in

ACMW, as shown in Figure 4a. Also shown is the measurement of pure ACMW used to determine the background level. The resulting adsorption isotherm is shown in Figure 4b, where it is compared to the surface excess obtained from surface tension measurements. We regard the agreement as satisfactory, particularly at high propofol concentrations, considering the different evaluation methods.

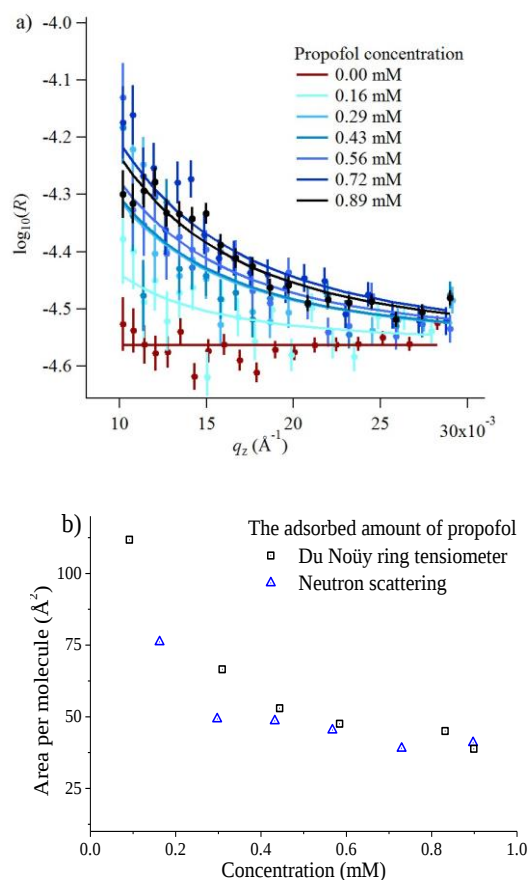


Figure 4. a) Neutron reflectometry data, R , and model fits for propofol solutions at the air/water interface, where the six bulk concentrations indicated are drawn progressively darker in colour; the pure ACMW background data are shown in red, and model fits are shown as lines; b) a comparison of the area per molecule obtained from surface tension measurements and NR.

Propofol Interfacial Organisation

In Figure 5, the NR data and model fits for a 0.89 mM propofol solution at the air/water interface are presented. The inter-layer roughness values were again constrained to the capillary wave value of 3.1 Å, the residual background was fitted to 2×10^{-7} (a.u.), and the thickness of the propofol monolayer and its volume fraction were fitted. The fit result using a generic algorithm converged to a dense propofol layer

of 8.4 Å thicknesses with a volume fraction of 0.86. These demonstrate that propofol forms a uniform fluid monolayer of high coverage and that propofol does not form islands at the interface.

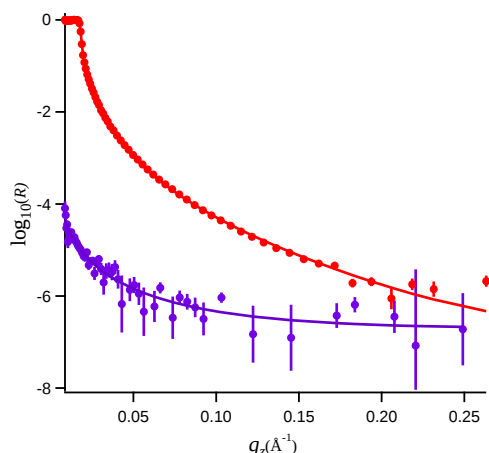


Figure 5. Neutron reflectometry data, R , and model fits for propofol solutions at the air/water interface recorded in D_2O (red, top) and air contrast matched water (purple, bottom).

Propofol is considered to be of cylinder-like shape. The total volume of such cylinder, calculated from the inverse bulk density, is $285\text{\AA}^3$. Close packing of such cylinder-like shapes accounts for the van der Waals volume of propofol⁵⁶ which is $198\text{\AA}^3$, 11% water (from the NR analysis) and about 22% air of the total volume. Let's consider the two extreme cases. If the molecules (thought as cylinders) are considered to stand on their ends, and if the cylinder is considered to have a cross section area of $25\text{\AA}^2$, slightly larger than for straight chain alkanes, then its length would be about 11 Å. The other extreme case would be a cylinder lying down giving an area per molecule of about $60\text{\AA}^2$ and a thickness of about 5-6 Å. Clearly, NR and tensiometry data are consistent with an intermediate situation (area per molecule $40\text{-}42\text{\AA}^2$, thickness 8.4 Å), suggesting a tilted orientation of propofol with the OH-group towards the aqueous phase (the latter supported by the VSFS data discussed next). We are reluctant to go further and report a mean tilt angle as it would require a specific assumption about the packing of the molecules, which is not known a priori.

VSF Spectra: CH region

The VSF data in Figure 6a show well defined peaks, which testify that propofol adsorbs at the air/water interface in a non-random fashion. In Figure 6b the attenuated total reflection (ATR)-IR spectrum of

propofol in the CH region is presented, showing that the bulk IR peaks are reproduced in the surface VSF spectra. The assignments are based on published IR data^{31, 57} and VSF spectroscopy work on similar molecules.^{13, 58-62} It is clear from Figure 6a that the SSP polarisation combination exhibits most of the vibrational features. The peak centred at 2873 cm^{-1} is assigned to the symmetric CH_3 stretch (sCH_3), and is similar to the sCH_3 peak found for alkyl chains. The small peak at 2910 cm^{-1} , which is present only at high concentrations, is assigned to the CH of the isopropyl unit. The peak at 2965 cm^{-1} dominates the entire SSP spectrum and is assigned to the antisymmetric CH_3 stretch (aCH_3). This peak is also observed in the PPP polarisation, but barely distinguishable in the SPS polarisation. There are two additional peaks in the SSP polarisation: the aromatic CH stretches at 3035 and 3071 cm^{-1} , which are clearly distinguished at high concentrations.

For alkyl chains, the aCH_3 peak is normally significantly weaker than the sCH_3 peak in the SSP polarisation combination due to the chain orientation.⁶³⁻⁶⁵ However, for propofol the opposite is found. The reason is that the total VSF signal has contributions from all four closely spaced methyl groups (see Figure 1), and due to different directions of the transition dipole moments for the symmetric and antisymmetric methyl stretches, their signal strength will be differently enhanced. The same argument rationalises that the antisymmetric methyl stretch is stronger in SSP than PPP, which normally is not observed for alkyl chains. It should be pointed out that in different environments (e.g. without water as solvent) the propofol SSP spectrum (not shown) resembles a typical spectrum from an alkyl chain, which emphasises that the unusual SSP spectrum for propofol at the solution-air interface is due to the propofol orientation.

All VSF peaks discussed above are also present in the IR spectrum shown in Figure 6b. This includes the isopropyl CH peak at 2910 cm^{-1} , which is observed as a small bump. Additionally, the IR spectrum shows the Fermi Resonance CH_3 at 2935 cm^{-1} which is not clearly observed in the VSF spectra. The fitted amplitudes of the VSF peaks are provided in the Supporting Information. In Figure 6c the fitted amplitudes of the sCH_3 and the aCH_3 stretches are presented as a function of propofol concentration. Above 0.2 mM the fitted amplitudes of both peaks approach a plateau, a result that is consistent with the

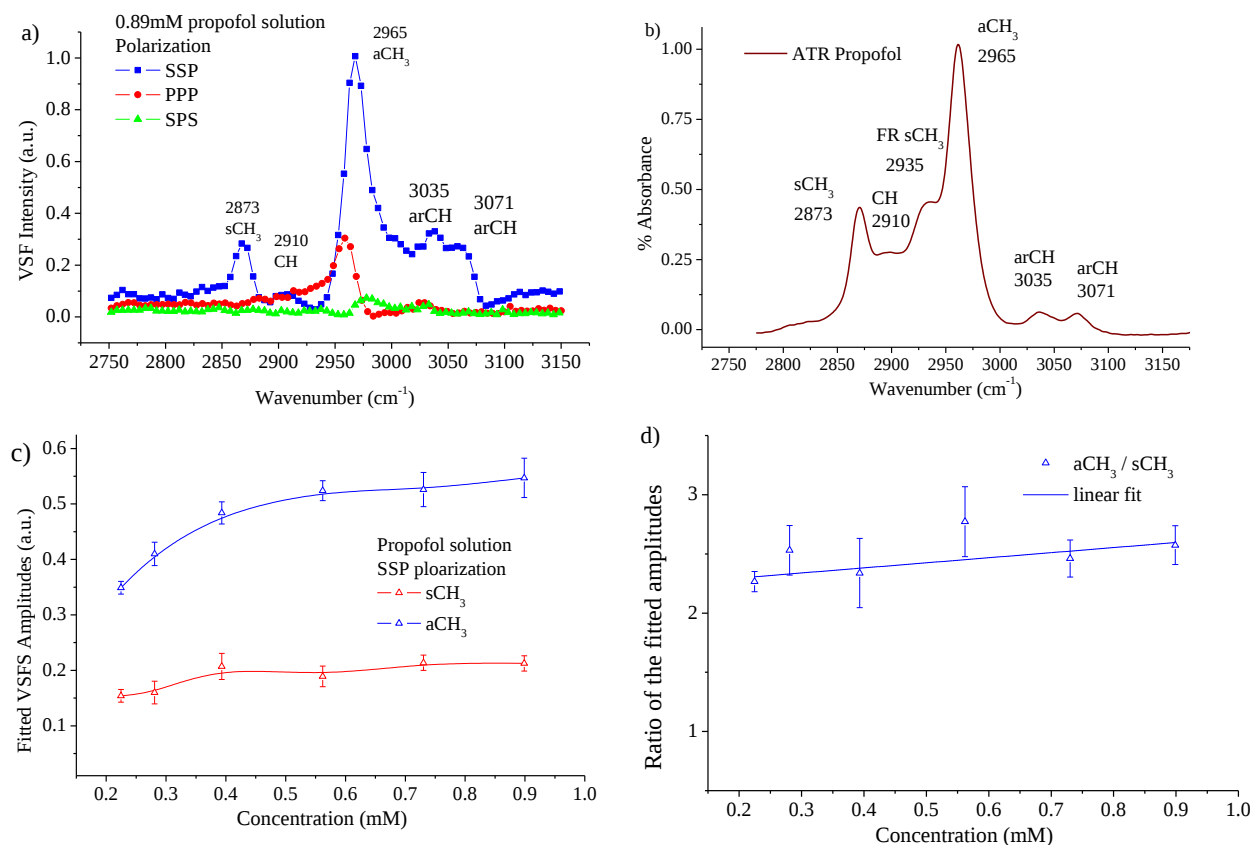


Figure 6. a) VSF spectra of a 0.89 mM propofol solution in the SSP, PPP, and SPS polarisations. All peaks are normalised to the highest intensity peak – SSP aCH₃; b) ATR-IR spectrum of pure propofol in the CH stretching region; c) fitted amplitudes for the sCH₃ stretch and the aCH₃ stretch as a function of propofol concentration in the SSP polarisation. d) the ratio of the aCH₃ to sCH₃ amplitudes as a function of concentration. The line is a linear fit to the data.

findings from surface tension isotherms, as discussed earlier (Figure 3). The sCH₃/aCH₃ fitted amplitude ratio as a function of concentration is presented in Figure 6d. The fitted amplitude depends both on the average orientation and the number of molecules probed, whereas the sCH₃/aCH₃ ratio is only affected by the orientation, since the number density is cancelled by taking this ratio. We note that this ratio is almost constant over the concentration range 0.2 – 0.89 mM, meaning that the average orientation of the adsorbed propofol is almost constant. The SPS polarisation did not show any CH feature that could be used reliably in the fitting procedure. This, and the fact that there are four methyl groups pointing in different directions that contribute to that signal, prohibits a more detailed orientational analysis. The assignments of vibrational features of propofol in the CH stretching region are presented in Table 1.

VSF Spectra: Water Region

In order to have a reference between different measurement sessions, spectra of the free OH vibration of the pure water surface were recorded

before all measurements. In Figure 7 spectra of a 0.89 mM propofol solution together with the pure water spectra in the SSP and PPP polarisations are shown.

Both the SSP and the PPP spectra of water agree with the spectra recorded before,^{17, 63, 68} where the most prominent features exhibited in the SSP polarisation are the broad band spanning over several hundred wavenumbers and centred at around 3300 cm⁻¹ as well as the sharp peak at 3704 cm⁻¹.

The latter peak, which is present in both the SSP and PPP polarisation, is assigned to the OH stretching mode of the topmost surface OH bonds of water molecules protruding into air and vibrating free from hydrogen bonding.⁶⁹ The assignment of the broad band was for a long time a source of debate.⁷⁰⁻⁷² However, it is broadly agreed now that the broad band from 3 150 to 3 450 cm⁻¹ is assigned to hydrogen bonded water molecules with varying strength and coordination meaning that the OH signal is a signature of a collective vibration of several water molecules.⁷³⁻⁷⁶

Table 1. Assignments of vibrational features of propofol in the CH stretching region.

Peak(cm^{-1})	Polarisation	Observed	Assignments	Refs.
2873	SSP	VSF/IR	sCH ₃	13, 57, 66
2910	SSP, PPP	VSF/IR	CH isopropyl	67
2965	SSP, PPP, SPS	VSF/IR	aCH ₃	13, 57, 61, 62
3035	SSP, PPP	VSF/IR	arCH	13, 57
3071	SSP	VSF/IR	arCH	13, 57

The SSP polarisation spectrum of a 0.89 mM propofol solution, shown in Figure 7a, is considerably different from the pure water spectrum. At this propofol concentration, the free OH peak at 3704 cm^{-1} has essentially vanished, suggesting that none or very few free OH bonds remain. However, a new broad band, centred around 3670 cm^{-1} , appears. A similar band has been observed for several amphiphilic molecules (e.g., decanol, sugar surfactants and alkyl polyethylene glycol surfactants) at the air/water interface, and has been assigned to the OH stretch of ordered water molecules weakly interacting with hydrocarbon moieties.¹⁷ This is consistent with the relatively large hydrophobic part of propofol, and suggests direct contact between water and this region of propofol. The small red shift of around 30 cm^{-1} compared to the free OH vibration indicates that the interaction is weak. We note that since propofol is non-ionic, any significant enhancement of the water signal as observed for ionic amphiphiles^{77, 78} is neither expected nor seen.

In the PPP spectrum in Figure 7b a band covering nearly the whole OH stretching region is observed, with the maximum intensity around 3600 cm^{-1} . Thus, the maximum is red shifted with around 70 cm^{-1} in comparison with the SSP spectrum, which exhibits nearly zero intensity at 3600 cm^{-1} . The OH bond populations responsible for the peaks at 3600 cm^{-1} and 3670 cm^{-1} possess weak interactions, and are thus excluded from hydrogen bonding due to proximity to the hydrophobic moieties of propofol. However, since the two peaks have their maximum at different wavenumbers, the two populations experience dissimilar environments.

An interesting observation is that the right hand side of the broad band, towards 3400 cm^{-1} is almost completely suppressed in the SSP polarisation combination (Figure 7a). Instead, a new band centred at about 3175 cm^{-1} clearly shows up in the spectrum. The fact that this band appears at such low frequency, close to the centre frequency for ice,^{79, 80} suggests that it is associated with interfacial water that experience

strong interactions with the hydroxyl part of propofol, similar to what was found for long chain alcohols.²⁰ A similar band, centred at 3150 cm^{-1} has further been observed for interfacial sugar surfactants possessing –OH groups (C10 maltoside and C10 glucoside) and assigned to OH stretching vibrations of the hydroxyl groups in the sugar rings and their hydration shells.¹⁷ In contrast, 1-decanol, which, like propofol, contains a single –OH group and a large hydrophobic moiety, exhibited a band centred at 3250 cm^{-1} , thus indicating relatively weaker hydrogen bonds.

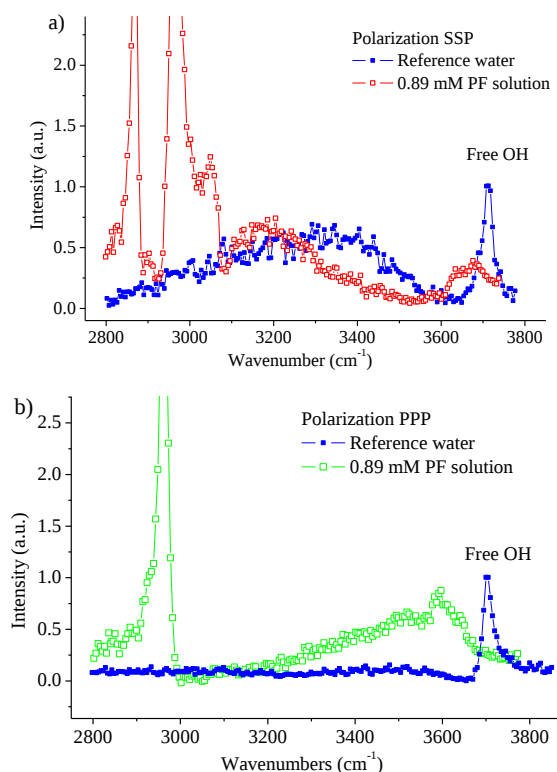


Figure 7. Water region spectra of a 0.89 mM propofol solution together with a pure water spectrum in a) SSP, and b) PPP polarisation combination. These two spectra are normalised to the intensity of the free OH vibration at the pure water surface.

Moreover, the PPP spectrum of 1-decanol showed a broad band extending over the region 3000 – 3800 cm⁻¹, obviously different from that obtained in presence of propofol. Accordingly, the strength of the hydrogen bonds of water that are hydrating propofol (SSP spectra) is in between those observed with the sugar surfactants and decanol, which indicates that the hydrogen bond strength depends not only on the hydrophilic group, which is the same for decanol and propofol, but also the surrounding hydrophobic environment.²⁰

The freedom of the molecules to rotate or move around may also influence the strength of the hydrogen bond. Propofol has a larger area per molecule than decanol¹⁷ and therefore has less constraints in adopting a position that allows for stronger hydrogen bonds to be formed.

Summary and Conclusion

Propofol does not form dimers or multimers in bulk solution up to the solubility limit (0.89 mM). It adsorbs at the air/water interface forming a dense (volume fraction 0.86) uniform film (area/molecule $\approx$ 40-42 Å², thickness $\approx$ 8.4 Å) close to the solubility limit. The propofol molecule is tilted relative the surface normal and oriented with the OH-group towards water. Its orientation at the interface is almost constant in the concentration range 0.2 – 0.89 mM.

We have identified different water populations that hydrate propofol at the air/aqueous solution interface. Strong hydrogen bonds, similar to those found for the tetrahedrally-coordinated hydrogen bonding in ice, is formed between water and the OH-group of propofol. These hydrogen bonds are stronger than those found between water and decanol, but weaker than those found next to sugar surfactants. Thus, the hydrogen bond strength does not only depend on the hydrophilic group, which is the same for decanol and propofol, but also on the surrounding hydrophobic environment and the ability to adopt conformations that allow formation of strong hydrogen bonds.

Effective molecular information regarding the arrangement and hydration of propofol at the air/water interface opens the door for a more extensive examination of the interfacial properties of propofol, especially at the buried membrane/water interfaces where this may have important implications for understanding the driving forces for the underlying interactions of drugs in model systems, and they set the stage for us and others to proceed with studies of

the interactions of propofol and other small model drugs with more complex interfacial morphologies.

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Conflicts of interest

There are no conflicts to declare

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