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The AR-G2 with Du Noüy Ring for Interfacial Rheometry

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INTRODUCTION

A number of methods have been developed over the years to investigate the surface and interfacial shear rheology [1]. Interfacial shear rheometry measures the mechanical strength or the shear viscosity of the interfacial layer i.e. the adsorbed monomolecular film at liquid-liquid or liquid-gas interfaces. Typical interfacial viscosities for insoluble monomolecular films are in the range from 10^{-5} to 10^{-2} N s/m. Assuming a film thickness if 1 nm, these viscosities are equivalent to bulk viscosities of 10^{4} to 10^{7} Pa s [2].

The Du Noüy ring, commonly used for interfacial tension measurements has been successfully applied to interfacial shear testing. Like the bi-cone, the ring is located at the interface of two liquids or a liquid and a gas. When the ring is subjected to an angular displacement, the surface between the ring and the circular wall of the vessel containing the liquids is sheared. The light construction of the ring permits the characterization of very fragile interfaces, without the inertia dominating the experiment. However, most bulk rheometers have too low sensitivity and too high inertia to use the Du Noüy ring in oscillation mode. Therefore specialized instruments with high sensitivity have been developed [3,4,7]. These instruments however have a limited operation range and cannot be used for rheological measurements other than interfacial shear experiments. The AR-G2 rotational rheometer with a torque range of almost nine decades and a sensitivity comparable to that of the specialized interfacial rheometers [5], has been successfully used in conjunction with a Du Noüy ring to perform dynamic interfacial rheology measurements over and beyond the range of specialized instruments.

AR-G2 AND DU NOÜY RING FOR INTERFACIAL SHEAR MEASUREMENTS

The Du Noüy ring can be used with the AR-G2 for sensitive interfacial measurements, because of its low inertia and its capability to control and apply tiny torques. The Du Noüy ring is attached to the stress motor, mounted on the slide of the rheometer. In the basic setup, a circular glass dish locates in the center on the Peltier plate and held in position

Figure 1: AR-G2 setup for interfacial measurements

with an annular Peltier cover as shown in figure 1.

For oscillation measurements, if the usually negligible contributions of the sample inertia and the instrument compliance are ignored, the torque balance of the rheometer provides:

$$\frac{M^*}{\phi^*} = \frac{(G_i^{'} + iG_i^{''})}{F_{ms}} - I\omega^2$$

with M^* the complex torque, ϕ^* the complex angular displacement, F_{ms} the geometry constant and $I\omega^2$ the contribution of the instrument inertia. G'_i and G''_i are the interfacial storage and loss modulus.

The geometry is treated as a two dimensional analogue of the concentric cylinder system. That means, the Du Noüy geometry constants are adopted from the concentric cylinder geometry by eliminating the bob length.

$$F_{ms} = \frac{1}{4\pi}\left(\frac{R_2^2 - R_1^2}{R_2^2 R_1^2}\right)$$

The Du Noüy ring used in this investigations is shown in figure 2 and has a radius of 10 mm, the thickness of the Pt-Ir wire being 0.36mm. Note that the annulus between inner and outer ring (R_2-R_1) is much wider, than what is typically used in bulk measurements. Possible errors due to the wide gap are not accounted for.

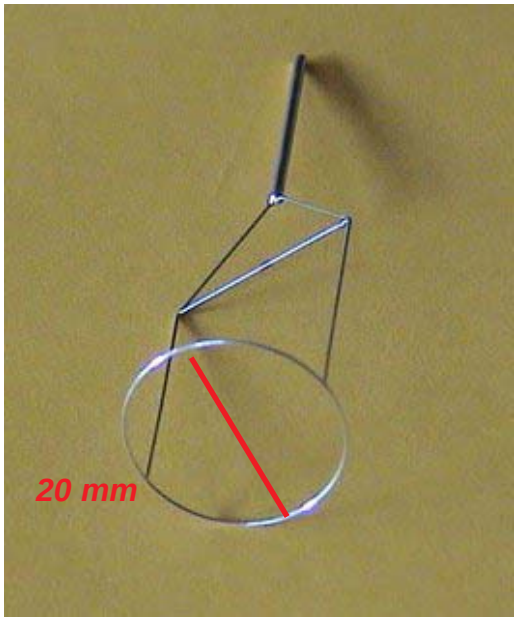


Figure 2: Du Noüy ring used for interfacial shear measurements. Ring diameter 20mm, Pt-Ir wire diameter 0.36mm

SAMPLE LOADING AND TEST SETUP

Interfacial measurements are very sensitive to contaminations. Before loading the sample, the Du Noüy ring has to be flamed in order to remove all sample residues. The circular glass vessel needs to be cleaned thoroughly also and rinsed with pure (de-ionized) water.

The dense phase (usually water or buffer solution) is loaded into the dish first. Next the ring has to be positioned into the plane of the liquid surface. Complete wetting of the ring is important. This step is critical and has to be done manually and by eye. Note: If a Langmiur trough is used instead of the glass dish, the Wilhelmy plate for monitoring the surface pressure can be used to position the ring at the surface. The surface active material such as a protein, a surfactant, etc., can be added to the water phase before loading or deposited on the surface afterwards. Finally, if required the lighter liquid phase is deposited on top of the dense liquid phase.

When the instrument is set up with a Langmuir trough, the surface pressure can be monitored with a Wilhelmy plate and increased during the experiment by compressing the interfacial area. The Langmuir trough has been modified for rheological testing. The center region is isolated with a circular boundary which defines the outer radius R_2 of the geometry. Channels in the circular barrier allow free diffusion of the surface active substances and as such changing the surface pressure without effecting the rheological measurement.

RESULTS

In a first experiment using the glass dish, the adsorption of lysozyme, a protein isolated from egg white was monitored at the water-decane interface. A 0.35µM solution of lysozyme was prepared in a phosphate buffer, pH 7.2. The decane was added after the Du Noüy ring was positioned at the surface. The full lines in figure 3 show the development of the interfacial storage G_i' and loss modulus G_i" with time, measured at a frequency of 0.05Hz and a strain amplitude of 2%. G_i' increases faster than G_i", which is due to the formation of a strong elastic gel at the interface. These results compare well with previous measurements done with the ISR400 rheometer developed by Gerry Fuller [6] (data points). The ISR400 rheometer measures the motion of a

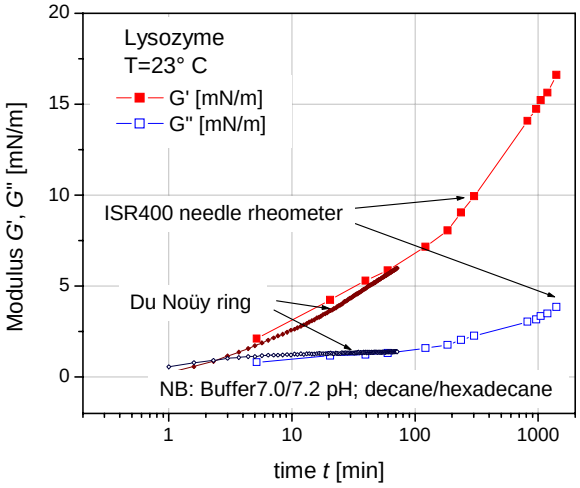
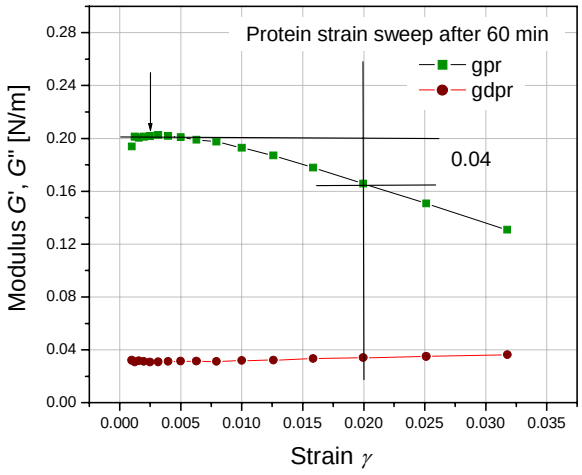


Figure 3: Adsorption of lysozyme at the water-decane interface, measured with the AR-G2 and the Du Noüy ring and the ISR400



gure 5: Strain sweep characterizing the interface layer of adsorbed Aß42 protein after 60 minutes

needle subjected to an oscillatory stress generated by a magnetic field. The measurements with the ISR400 were conducted over a much longer time period, showing that the modulus continues to increase indefinitely. The same test conditions were chosen for both experiments, except slight differences in the buffer pH (7.0 vs. 7.2) and the second liquid phase (decane vs. hexadecane)

After the adsorption of the native globular protein at the interface partially unfolding, exposing the hydrophobic groups, takes place. The proteins aggregate and form an elastic layer. The continuous increase of the interfacial shear modulus suggest that proteins continue to unfold and aggregate, forming multi-layers at the interface [6].

Additional experiments were carried out with a different protein, the prion Aß42. In contrast to the lysozyme, the Aß42 builds a monomolecular film at the surface and a constant modulus G'_i will eventually be reached. The adsorption of the protein to saturate the surface happens fast, the unfolding of the protein and the gelation at the interface is a much slower process.

After 60 minutes, the time sweep experiment (Figure 4) was interrupted to conduct a series of additional tests. The results are shown in figure 5 and 6. Note that the modulus has not quite reached a constant value at this point. The strain sweep (figure 5) shows a linear region below 0.2% strain. The interfacial storage modulus decreases with increasing strain, while G" remains constant. The frequency response at 0.5% strain amplitude in figure 6 shows a typical solid like response with G" virtually independent of frequency and G‘ only

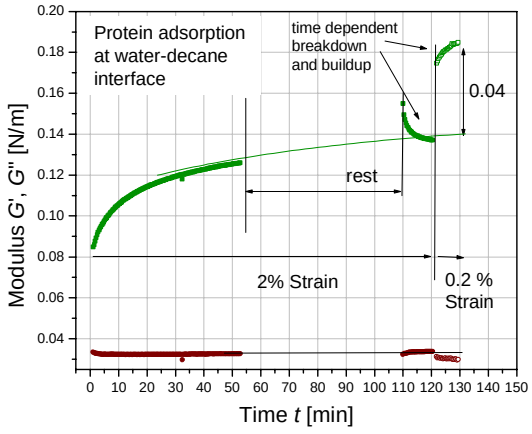


Figure 4: Adsorption of Aß42 at the water decane interface, monitored in a dynamic time sweep.

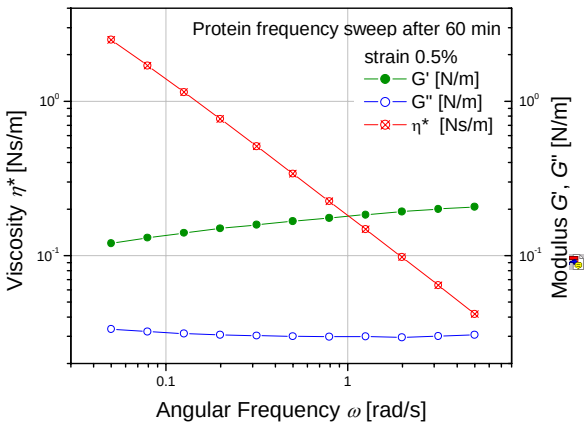


Figure 6: Frequency sweep characterizing the interface layer of adsorbed Aß42 protein after 60 minutes.

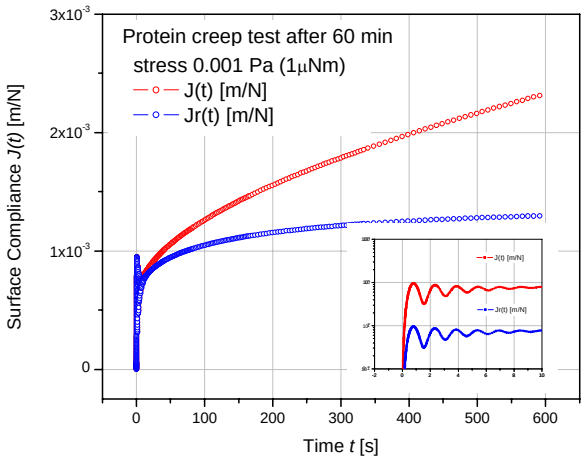


Figure 7: Ringing at the start up of the Creep and Recovery test of the interface layer of adsorbed Aß42 protein

slightly increasing with frequency.

The initial time sweep was performed at a frequency of 0.05 Hz and a strain amplitude of 2%. The onset of the non-linear region shows at a lower strain of 0.2% in figure 5. This explains the decrease of the storage modulus after the time sweep is resumed 110 minutes from the start with 2% strain (figure 4). By reducing the strain amplitude to 0.2%, the interfacial storage modulus increases by 0.04 N/m, which is consistent with the result from the strain sweep.

A creep revocery test was performed on the same sample, 1 hour after adding the protein Aß42 (Figure 7). At the start up of the creep and the recovery tests, pronounced ringing was observed. This is proof of the highly elastic nature of this protein layer at the interface.

The elasticity of the interfacial layer is due to the unfolding and aggregation of the protein at the interface. Most proteins have a globular three dimensional structure, held together by hydrogen bonds and Van der Waals forces. At the interface, the polar groups orient towards the water phase, the hydrophobic groups are exposed and oriented towards the non polar fluid phase. The bonds holding the three dimensional structure disappear and the protein unfolds at the surface to develop an energetically favorable configuration. Strong intermolecular forces are responsible for the aggregation and development of a “two dimensional” gel structure at the interface.

CONCLUSION

Due to the high torque sensitivity and the low system inertia, the AR-G2 equipped with the Du Noüy ring proves to be an excellent combination to characterize interfaces in both, transient and oscillation test modes. The Interfacial option is an inexpensive option for the AR-G2 and makes the AR-G2 an ideal rheometer for characterizing the bulk and interfacial rheological properties of colloid systems.

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