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# Boundary flow of water on supported phospholipid films

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**Abstract.** – We report an investigation of the boundary flow of water on DPPC monolayers and bilayers deposited on Pyrex surfaces, using a dynamic surface force apparatus. We find that a no-slip boundary condition is verified at the water/bilayer interface within 2 nm. In contrast, the interfacial flow properties on monolayers evolve with the time of exposure to water. A significant slip of 10 nm is found on fresh monolayers, which are strongly hydrophobic. The hydrodynamic interaction force with a nearby surface is then significantly reduced over hundreds of nanometers. This slip effect disappears after one day of exposure to water, due to the increased hydrophilicity and roughness of hydrated monolayers.

*Introduction.* – Hydrodynamic interactions play an important role in a number of biological processes, such as the flow of red blood cells or the adhesion of monocytes to endothelial cells in blood vessels [1], boundary lubrication of articular cartilage [2], as well as in flow devices for biological measurements such as flux chambers [3] or microfluidic devices [4]. A key feature governing the hydrodynamic forces acting on an object moving in a liquid is the flow boundary condition on its surface, or equivalently the liquid-surface friction coefficient. If the no-slip boundary condition is usually assumed to provide a good description of liquid flows on most surfaces, recent results show that water can develop a significant slip on surfaces at the scale of some tens of nanometers, depending on the surface roughness and hydrophobicity [5, 6]. These results raise the question of small-scale hydrodynamics of water on surfaces of biological interest.

Phospholipid films have been widely used as model structures to study the properties of native biological cell membranes and to investigate biological processes [7]. As a model system, the morphology and stability of supported phospholipidic films has been well characterized [8, 9], and static interaction forces between phospholipid bilayers have been studied [10–13]. Recently, the stability of surfactant monolayers in water has been investigated by Surface Force Apparatus and atomic force microscopy (AFM) by Meyer *et al.* and Perkin *et al.* [14, 15]. Investigations at the scale of several tens of nanometers have been conducted with the RICM technique on hyaluronic-acid–coated lipid bilayers, and conclude to the validity of usual hydrodynamics on theses surfaces [16]. It was also observed that a significant slip exists for water flowing in nanopores covered by a surfactant fluid bilayer [17].

In this letter we report an experimental investigation of water boundary flow on supported monolayers and bilayers of DPPC phospholipids, resolved at the nanometer scale. We show that while the flow of water on dense DPPC bilayers is well described with a no-slip boundary

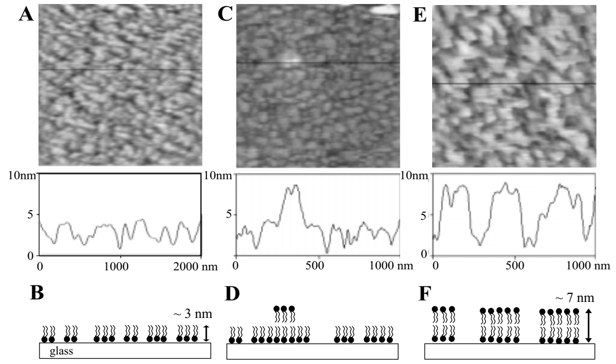

Fig. 1 – AFM image in contact mode of a DPPC monolayer immersed in water, and schematic arrangement of the lipids shown by the topography. A and B: after 2 hours in water; C and D: after 7 hours in water; E and F: after 1 day in water.

condition, significant slip arises on fresh DPPC monolayers, corresponding to a slip length of 10 nm. As a result, the hydrodynamic interaction with such a fresh monolayer is significantly reduced over hundreds of nanometers. This slip effect disappears on DPPC monolayers exposed several hours to water, as their roughness and hydrophilicity increases.

*Material.* – We study mono- and bilayers of DPPC (dipalmitoylphosphatidylcholine) (Aventi Polar Lipids) deposited on clean hydrophilic float Pyrex surfaces at high lateral pressure by the Langmuir-Blodgett method. For this purpose monolayers of DPPC are formed in a Langmuir trough by spreading a millimolar solution of DPPC in chloroform-ethanol (9:1 v/v) at the surface of ultrapure water (Millipore, MilliQ, 18.2 MΩ.cm). They are transferred on the Pyrex surfaces at surface pressure of 40 mN/m as previously described [18]. The monolayers obtained are stable in air and perfectly smooth: the roughness measured by AFM over a 1 $\mu\text{m}^2$ area is less than 2 Å r.m.s., which is comparable to the underlying surface roughness [19] and indicates a uniform surface coverage. Those monolayers are hydrophobic: the contact angle of water measured by the sessile drop method is 95° with a hysteresis of 10°.

Bilayers are obtained by dipping into the trough the substrates already covered by a monolayer. DPPC bilayers are in the gel phase at room temperature. They are hydrophilic and not stable in air. When imaged with contact mode AFM in water, bilayers appear smooth (less than 2 Å r.m.s.) and do not reveal defects such as holes or protrusions as previously found [18]. This is in agreement with numerous studies [9, 20, 21] showing that defects are present only when the second lipid layer is transferred at a lower surface pressure. It can therefore be assumed that the DPPC bilayer covers uniformly the Pyrex substrate.

In order to study the flow of water on monolayers, these monolayers have to be immersed in water. The structure of DPPC monolayers in water evolves slowly in time due to lipids reorganization [22]. Such behaviour has already been reported in the case of DPPE on mica surfaces [9] and for surfactant monolayers in water [14,15]. Just after immersion the monolayer roughness increases compared to its value in air (fig. 1A). The height of holes, about 3±1 nm, is half a DPPC bilayer thickness [18,20] and indicates a partial desorption of the lipids (fig. 1A). Of course such a topographical image itself does not guarantee that the bottom of the holes is the bare glass. It might be a first lipid monolayer covered by a second one (*i.e.* a bilayer presenting defects in its upper leaflet as found in [9]). It might be also a first lipid bilayer covered by a lipid monolayer with defects (*i.e.* a trilayer as a whole). However, we deposited initially just a monolayer with no lipid excess. As the sample seems homogeneous, it is very

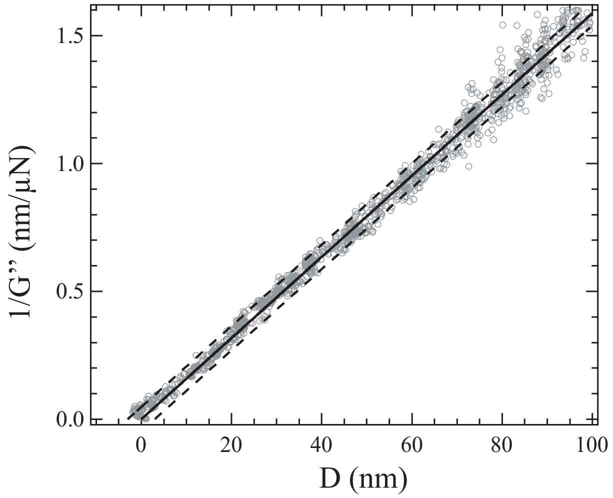

Fig. 2 – Inverse of the damping $1/G''$ measured between a Pyrex sphere and a plane supporting a dense DPPC bilayer, as a function of their distance $D$. The experiment is performed at a frequency $\omega/2\pi = 69\,\mathrm{Hz}$ with an oscillating amplitude of 7 Å. The sphere radius is $R = 2.9 \pm 0.1\,\mathrm{mm}$. The solid line is the best adjustment of the data with a straight line intersecting the origin, which corresponds to the Reynolds damping with a no-slip boundary condition. The dotted lines show the far-field damping that would be obtained for a no-slip plane located at ± 3 nm from the origin.

unlikely that the only desorbed lipids created supplementary lipid layers. It seems therefore that the surface keeps a monolayer structure (fig. 1B). At longer time scales (several hours) the roughness of the film increases up to several nanometers and reveals self-assembled lipid structures on the top of the monolayer (figs. 1C-D). After one day and more in solution, a significant part of the surface seems covered by a reconstructed bilayer with holes of 6 nm height exposing the bare Pyrex to water (figs. 1E-F). Again, here the absence of lipid excess makes any other conclusion very unlikely. This structural evolution is associated to changes in the macroscopic contact angle of water on samples withdrawn from bath. The advancing contact angle decreases with the time of exposure to water, while the contact angle hysteresis increases, as expected for a film with increasing roughness and portions of hydrophilic area [22].

*Hydrodynamic measurements.* – The boundary flow of water on the supported films is investigated with a dynamic Surface Force Apparatus [23] specifically designed to measure hydrodynamic interactions between surfaces separated by a liquid layer. This device measures directly the hydrodynamic force between the supported film and a sphere of millimetric radius $R$, oscillating at very small amplitude in a direction normal to the film. In the case of a no-slip boundary condition, the viscous damping induced by the flow, defined as the dissipative force divided by the oscillation amplitude, is given by the Reynolds force: $G(\omega)'' = 6\pi\omega\eta R^2/D$. Here $\omega/2\pi$ is the oscillating frequency, $\eta$ the fluid viscosity, $R$ the radius of the sphere and $D \ll R$ the sphere-layer distance. Figure 2 shows the inverse of the damping $1/G''(\omega)$ as a function of the distance $D$ obtained for a DPPC bilayer in water. The sphere is made of smooth plain Pyrex, on which a no-slip boundary condition is obtained for water flow [6]. The damping in fig. 2 does not depend on the shear rate of the oscillating motion, up to the maximum range investigated, approximately $10^3\,\mathrm{s}^{-1}$. The plot is in very good agreement with the Reynolds force, which predicts a linear variation of $1/G''(\omega)$ as a function of $D$. From the slope of the plot, we derive the viscosity of the confined water: $0.92 \pm 0.07\,\mathrm{mPa\,s}$ (the error

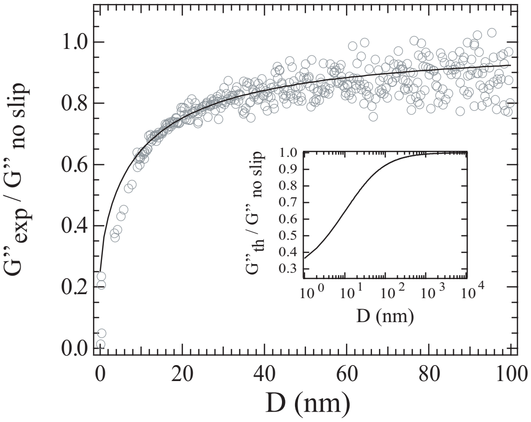

Fig. 3 – Ratio of the damping $G''_{exp}$ measured on the fresh supported DPPC monolayer in water, to the damping expected in the case of a no-slip boundary condition, as a fonction of the sphere-layer distance $D$. The solid line is the theoretical ratio for a slip length of 10 nm at the water-monolayer interface. The inset is this theoretical ratio plotted on a larger distance scale.

bars for the viscosity come from the error bars in the determinaion of the radius of the sphere), in good agreement with the viscosity of water at 25 °C: 0.89 mPa s. Therefore the confinement of water on a DPPC bilayer has no measurable effect on its viscosity down to confinement of a few nanometers, in good agreement with results obtained on other surfaces [6, 24, 25]. This plot also shows that the no-slip boundary condition holds on a DPPC bilayer, since the best fit of the data with a straight line intercepts the $x$-axis at the origin. More precisely, one can define a "no-slip plane" from the extrapolation on the $x$-axis of the linear variation of $1/G''$ with the distance. Taking into account the scatter of the data, the position of this no-slip plane is obtained with a ± 2 nm resolution. This position corresponds to the position of the water/bilayer interface, taken as the origin for the distance $D$. The latter is determined with an error bar of 2 nm [6] from the point where the quasi-static interaction force becomes strongly repulsive. The typical forces acting on the bilayer during the contact, tens of mN/m, are not strong enough to eject the bilayer from the contact area [11]. We conclude from these findings, that the hydrodynamic force exerted by a flow on a supported bilayer can be described by a no-slip boundary condition applied on the water/upper leaflet interface with a resolution of 2 nm.

Figure 3 presents the ratio of the damping obtained for water flowing on a fresh DPPC monolayer, exposed to water for less than 1 hour, to the damping expected in case of a no-slip boundary condition at the monolayer/water interface. This ratio is less than 1 over the range of distance investigated, extending up to 100 nm, which shows that the hydrodynamic interaction of the sphere with the fresh monolayer is significantly reduced over that range. We attribute this reduction to the boundary slip of water on the monolayer. This slippage effect is more accurately described by the slip length $b$ which relates locally the boundary velocity of water $V_s$ to the shear rate at the wall $\dot{\gamma}_s$: $V_s = b\dot{\gamma}_s$. A determination of $b$ is made by adjusting an analytic expression with the experimental curve. The analytic expression used corresponds to the viscous force between a non-slipping sphere and a slipping plane [26]:

$$G''(\omega) = \frac{6\pi\omega\eta R^2}{D} f^*\left(\frac{b}{D}\right), \qquad f^*\left(\frac{b}{D}\right) = \frac{1}{4}\left(1 + \frac{6D}{4b}\left[\left(1 + \frac{D}{4b}\right)\ln\left(1 + \frac{4b}{D}\right) - 1\right]\right). \quad (1)$$

The plot of the inverse of the damping $1/G''$ *vs.* $D$ is no longer a straight line. However, in the limit $D \gg b$, the following expression for the asymptote is obtained: $G''(\omega) = \frac{6\pi\omega\eta R^2}{D+b}$.

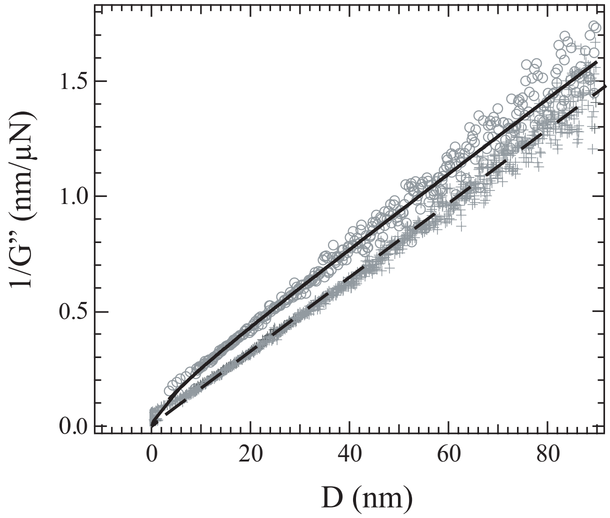

Fig. 4 – Inverse of the damping $1/G''$ measured on DPPC monolayers in water as a function of the sphere-layer distance $D$. The experiments are performed at a frequency $\omega/2\pi = 69\,\mathrm{Hz}$ with an oscillating amplitude of 8 Å. The sphere radius is $R = 2.8 \pm 0.1\,\mathrm{mm}$. (○) freshly hydrated monolayer after less than 2 hours in water ; (+) same monolayer after one day in water. The dashed line is the fit with a Reynolds damping for a no-slip hydrodynamic boundary condition. The solid line is the fit with the Reynolds damping for a 10 nm slip length.

Hence, for distances $D \gg b$, the plot of $1/G''(D)$ as a function of $D$ is translated by a distance $b$ to the left compared to the no-slip case.

In the fresh DPPC monolayer case a good agreement is found with a slip length value of $b = 10 \pm 2\,\mathrm{nm}$. One can see on the inset of fig. 3 that the damping expected with this slip length value remains significantly lower than the damping for a no-slip condition, up to distances of $1\,\mu\mathrm{m}$. This effect is due to the long-range nature of hydrodynamic forces, and reinforce the importance of a good description of the boundary condition for estimating the hydrodynamic force acting on the lipidic film.

Finally, fig. 4 compares the inverse of the damping $1/G''(\omega)$ obtained for a fresh monolayer and a monolayer aged one day in water. On the strongly hydrated monolayer the linear variation of $1/G''$ with the distance intersects the $x$-axis at the origin, *i.e.* the no-slip boundary condition is recovered. This loss of slipping property is in agreement with the actual understanding of interfacial hydrodynamics: both roughness and hydrophilicity, which are increased after one day in water (fig. 1), are expected to decrease boundary slip effects [27–29]. From the slopes of the plots, we derive the viscosity of the confined water: $0.96 \pm 0.07\,\mathrm{mPa\,s}$ (the error bars for the viscosity come again from the error bars in the determination of the radius of the sphere), in good agreement with the viscosity of water at 25 °C: $0.89\,\mathrm{mPa\,s}$.

One also notices that at distances less than 10 nm the data for the hydrated monolayers deviates significantly from the fit with the Reynolds damping. This may reflect the spatial non-uniformity of the hydrated monolayers. Work is in progress to study this specific aspect.

*Conclusion.* – The boundary flow of water on phospholipidic films is an important parameter which determines the viscous forces and hydrodynamic interactions involving these films. This investigation at a nanometer scale shows that a no-slip condition holds within 2 nm on DPPC bilayers in the gel phase. This result confirms the validity of usual hydrodynamics for describing flow on these systems. By extension we expect also usual hydrodynamics to hold at a small scale on more complex lipid membranes, since surface roughness and heterogeneity usually favor higher liquid friction at the interface. A no-slip boundary condition is also observed on fully hydrated monolayers, and is attributed to their heterogeneous structure. In contrast, substantial slip is observed on fresh smooth DPPC monolayers. The slip length of 10 nm corresponds to a water/monolayer friction coefficient of $10^5\,\mathrm{N\,s\,m^{-3}}$. Work is in progress to study the inter-monolayer friction within fluid bilayers.

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## REFERENCES

[1] Lawrence M. B. and Springer T. A., *Cell*, **65** (1991) 859.
[2] Benz M., Chen N., and Israelachvili J., *J. Biomed. Mater. Res. A*, **71** (2004) 6.
[3] Pierres A., Benoliel A.-M., Zhu C. and Bongrand P., *Biophys J.*, **81** (2001) 25.
[4] Whitesides G. M., and Stroock. A. D., *Phys. Today*, **54** (2001) 42.
[5] Lauga E., Brenner M. P. and Stone H. A., *Handbook of Experimental Fluid Dynamics*, edited by Foss J., Tropea C. and Yarin A. (Springer, New York) 2005, Chapt. 15.
[6] Cottin-Bizonne C., Cross B., Steinberger A. and Charlaix E., *Phys. Rev. Lett.*, **94** (2005) 056102.
[7] Sackmann E., *Science*, **271** (1996) 43.
[8] Dufrêne Y. F. and Lee G. U., *Biochim. Biophys. Acta*, **1509** (2000) 14.
[9] Benz M., Gutsmann T., Chen N. H., Tadmor R. and Israelachvili J., *Biophys. J.*, **86** (2004) 870.
[10] Marra J. and Israelachvili J., *Biochemistry*, **24** (1985) 4608.
[11] Wong J. Y., Park C. K., Seitz M. and Israelachvili J., *Biophys J.*, **77** (1999) 1458.
[12] Leckband D., *Nature*, **376** (1995) 617.
[13] Pincet F., Perez E., Bryant G., Lebeau L. and Mioskowsk C., *Phys. Rev. Lett.*, **73** (1994) 2780.
[14] Meyer E. E., Lin Q., Hassenkam T., Oroudjev E. and Israelachvili J., *Proc. Natl. Acad. Sci. U.S.A.*, **102** (2005) 6839.
[15] Perkin S., Kampf N. and Klein J., *J. Phys. Chem. B*, **109** (2005) 3832.
[16] Shilling J., Sengupta K., Goennenwein S., Baush A. R. and Sackmann E., *Phys. Rev. E*, **69** (2004) 021901.
[17] Cheikh C. and Koper G., *Phys. Rev. Lett.*, **91** (2003) 156102.
[18] Cross B., Ronzon F., Roux B. and Rieu J.-P., *Langmuir*, **21** (2005) 5149.
[19] Rieu J.-P., Ronzon F. and Roux B., *Thin Solid Films*, **406** (2002) 241.
[20] Mou J. X., Yang J. and Shao Z. F., *J. Mol. Biol.*, **248** (1995) 507.
[21] Bassereau P. and Pincet F., *Langmuir*, **13** (1997) 7003.
[22] Cross B., Steinberger A., Cottin-Bizonne C., Charlaix E. and Rieu J.-P., *Aging of hydrated phospholipid layers: structural forces and hydrodynamic boundary condition*, in preparation.
[23] Restagno F., Crassous J., Charlaix E., Cottin-Bizonne C. and Monchanin M., *Rev. Sci. Instrum.*, **73** (2002) 2292.
[24] Israelachvili J. N., *J. Colloid Interface Sci.*, **110** (1986) 263.
[25] Raviv U., Giasson S., Frey J. and Klein J., *J. Phys. Condens. Matter*, **14** (2002) 9275.
[26] Vinogradova O. I., *Langmuir*, **11** (1995) 2213.
[27] Barrat J. L. and Bocquet L., *Phys. Rev. Lett.*, **82** (1999) 4671.
[28] Richardson, *J. Fluid Mech.*, **59** (1973) 707.
[29] Schmatko T., Hervet H. and Leger L., *Phys. Rev. Lett.*, **94** (2005) 244501.