

Does Glue Contaminate the Surface Forces Apparatus?

Frédéric Pincet,* Eric Perez, and Joe Wolfe†

Laboratoire de Physique Statistique de l'École Normale Supérieure, associé aux Universités Paris 6 et Paris 7, 24 rue Lhomond, Paris 75005, and School of Physics, University of New South Wales, Sydney 2052, Australia

Received July 22, 1994. In Final Form: October 7, 1994

A surface forces apparatus (SFA)¹ is widely used to measure forces (precision typically $\pm 10^{-7}$ N) between two curved surfaces as a function of their separation (± 0.02 – 0.1 nm).² To achieve a geometrical configuration which allows local close approach of regular surfaces, two thin mica sheets, cleaved to yield atomically smooth surfaces, are glued onto two cylindrical glass lenses with a thermosetting glue, usually Epikote 1004 (from Shell). It has been reported that this glue contains some surface-active components which appear at air-water interfaces when sufficiently large areas of the glue are exposed to aqueous solutions.³ Most experiments using the SFA involve the exposure of an area of glue, albeit small, to the solution used. It is therefore important to measure or to place an upper limit on the effects of surface-active compounds which may be released into aqueous solutions under conditions similar to those in an experiment with the SFA.

We have used the SFA to study the forces between lipid bilayers in aqueous solutions of a variety of solutes.⁴ We therefore conducted two series of experiments using surface tension measurements to determine the extent of surfactant contamination from the glue. The first series sought the effects of contaminant release over several days. The second was more sensitive and sought to discover the quantity of glue required to give a measurable effect. The solutions used were those used in our bilayer study: pure water, a salt solution (NaCl), a sugar solution (sorbitol), and a mixture of water and the solvent dimethyl sulfoxide, hereinafter called DMSO.

In the first series of experiments, thin silvered pieces of mica, about 7 mm square, were glued to small glass plates in the same manner as that used to glue mica to the lenses of the apparatus. Each plate was placed in a crystallization dish. All glassware was cleaned with chromosulfuric acid and copiously rinsed in water purified by an Elga UHQ system. For each of the solutions there was also a control which contained a glass plate without mica or glue. To each pair of dishes, experiment and control, was added 50 mL of fluid. The fluids used were pure water, water plus DMSO (3:1 v:v), 1 kmol·m⁻³ sorbitol solution, and 150 mol·m⁻³ NaCl. A further dish contained pure water and a glass plate upon which a small quantity of glue (ca. 0.8 mg) had been melted and allowed to cool to form a patch about 8 mm in diameter. The surface tension of each solution was measured immediately after the addition of the solution, 1 day later, 2 days later, and 1 week later. The measurement technique was a variant of the Wilhelmy plate method⁵ with an absolute precision of 0.5 mN·m⁻¹.

When measured immediately following preparation, all samples gave surface tensions that differed from those of the controls by less than 0.5 mN·m⁻¹. One and 2 days after preparation, the surface tensions of all samples had changed by less than 0.5 mN·m⁻¹. After 1 week the surface tension of all samples had changed by less than 0.5 mN·m⁻¹, with two exceptions: that of the mica glued to glass in the salt solution fell by 0.8 ± 0.5 mN·m⁻¹, and that of the sorbitol control sample (i.e. glass without glue or mica in sorbitol solution) fell by 0.8 ± 0.5 mN·m⁻¹.

In the second series of measurements, the surface tensions of fluids were measured in a Teflon container (surface area 20 cm²). A Teflon sweeper allowed compression of the surface to 7 cm², this limit being imposed by the measurement of surface tension which requires a region in which the meniscus is sensibly flat. The first control contained only pure water; the second control contained pure water and a clean glass plate. In the first experiment, a piece of silvered mica was glued to a glass plate, as described above, and placed in clean water. In the second experiment, a patch of glue (ca. 0.8 mg and 8 mm in diameter) was melted onto a clean glass plate and cooled and the plate placed in clean water. In the third experiment, about 1 mg of glue (comparable to the quantity used for SFA experiments) was deposited onto the surface with a clean spatula. In the fourth experiment, 15 mg of powdered glue was deposited onto the surface. The reason for using powdered glue was to simulate the experiments reported by Parsegian and Gershfeld.³

Rapid compression of the surface from 20 to 7 cm² did not change the surface tension for the controls, for the glass with glued mica, or for 1 mg glue. For the plate with only melted glue, the surface tension fell by 0.5 ± 0.5 mN·m⁻¹ upon compression. In all these cases, the result was unchanged when the measurements were repeated 1 h after immersion. For the experiment with 15 mg of glue powder, however, the surface tension fell by about 10 mN·m⁻¹ upon rapid compression. This last measurement, using a large surface density of finely divided glue, is consistent with the observations of Parsegian and Gershfeld (personal communication).

We note that the size of the effect depends not only on the volume of the glue used but also upon the surface area exposed. This may explain why all the experiments involving mica glued to glass gave null results. The area covered by the mica is always greater than that covered by the glue beneath it, so the area of glue exposed to the aqueous phase is at the perimeter of the glued area. An upper estimate of the area of glue exposed to the solution is obtained from the quantity of glue used (typically several hundred micrograms) by assuming that the glue forms a layer of uniform thickness between mica and glass. This gives a value of about 0.5 mm². This is an overestimate because the glue layer is actually thinnest at the perimeter of the glued area. This contrasts with a surface area of 20 mm² for 1 mg of finely divided glue, estimated from measurements of the sizes of the particles. This observation explains the contradictory reports of the extent of contamination by glue: in order to give a level of contamination that is readily measurable, the glue must have a surface area about 2 orders of magnitude greater than is usually the case in experiments using the SFA.

Following the suggestion of Parsegian (personal communication), who read a draft of this note, we also conducted the following experiment. One mica square was glued to glass as described above and placed in a beaker with water from the Elga system. A control mica

† University of South Wales.

(1) Israelachvili, J. N.; Adams, G. E. Measurements of forces between two mica surfaces in aqueous electrolyte solutions in the range 0–100 nm. *J. Chem. Soc. Faraday Trans. 1* **1978**, *74*, 975–1001.

(2) Israelachvili, J. N. *Intermolecular and Surface Forces*, 2nd ed., Academic Press: London, 1992.

(3) Parsegian, V. A.; Gershfeld, N. L. Inert glue in the surface force apparatus—where are the controls? *Biophys. J.* **1993**, *64*, 222A.

(4) Pincet, F.; Perez, E.; Wolfe, J. Do trehalose and dimethylsulphoxide affect inter-membrane forces? *Cryobiology* **1994**, *31*, 531–539.

(5) Mann, E. Doctoral thesis, École Normale Supérieure, Paris, 1993.

square and glass plate were placed in water without glue. After 20 h, both pieces of mica were removed and dried in a jet of nitrogen. A third piece of mica was freshly cleaved. A small drop of water from the Elga system was placed on each and the contact angle behavior observed. In both experiments and control, the drop spread rapidly to form a large thin film on the mica, and no differences between the spreading behaviors were observed. We also conducted an experiment to look for contamination of the mica *in situ* in the solution by bringing an air bubble into the vicinity of mica surfaces. The behaviors of mica glued to glass and unglued mica were the same. In neither case was there any indication of a tendency of the air bubble to adhere to the mica.

The following question remains: even if contamination is not readily measurable using surface tension techniques or spreading behavior, could there still be sufficient contamination to invalidate the applicability of results made with the SFA? The answer to this question depends

on the circumstances of the use of the SFA, the degree of the putative contamination, and the sensitivity of the results to trace contamination.

In the case of bilayer studies, the surfaces between which forces are measured are mica upon which lipid bilayers have been deposited at surface pressures of typically $40 \text{ mN}\cdot\text{m}^{-1}$. It seems unlikely that a contaminant that produces no measurable surface pressure in a pure water interface will partition in substantial quantity into a bilayer deposited at high surface pressure.

Our conclusion from the observations involving glue is that, in the manner in which the surface forces apparatus is usually used, and over a period of up to 2 days, glue is unlikely to change the surface tension of the solution in the apparatus by $0.5 \text{ mN}\cdot\text{m}^{-1}$ and unlikely to affect the results of experiments using deposited bilayers.

LA9405814