

Resonance Reflectivity from Langmuir Monolayers of Fatty Acids with Associated Counterions

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Introduction

Resonant x-ray reflectivity can simultaneously determine the overall electron density distribution of a monolayer and the precise location of resonant atoms within that layer, with sub-angstrom resolution [1]. This makes it a valuable tool for studying proteins whose function can often depend both on the presence of a single metal atom bound to a specific site within the macromolecule and on their incorporation into a membrane [2]. The role of Langmuir monolayers as reasonable model membrane systems makes it desirable to adapt resonant x-ray reflectivity to the air-water interface [3]. The technique makes use of the change in the scattering factor of atoms in the vicinity of their absorption edge — $f(E) = f'(E) + if''(E)$ — and compares reflectivity data collected at the absorption edge and data collected at energies above and below the edge. It requires a liquid surface spectrometer (LSS) and constant exit-height monochromator so that the alignment of the spectrometer is maintained as the energy is changed. As a test case, we studied a fatty acid monolayer spread on a subphase containing a zinc salt by using x-rays at and near the K absorption edge of Zn.

Methods and Materials

The fatty acid $\text{CH}_3(\text{CH}_2)_{18}\text{COOH}$, called arachidic acid and abbreviated AA, was purchased from Sigma (St. Louis, MO), dissolved in high-pressure liquid chromatograph (HPLC)-grade chloroform at 1 mg/mL, and spread onto aqueous subphases (pH 8.3) that contained 10 mM of ZnCl_2 (the ZnAA monolayer) or 10 mM of BaCl_2 (BaAA) as a control. Working at 22°C, the monolayers were compressed to 5 mN/m and maintained at that surface pressure during the reflectivity measurements. The first harmonic of the undulator was used to obtain photon energies of $K = 9659$ eV, calibrated

TABLE 1. Real (f') and imaginary (f'') parts of the scattering factor of Zn in the vicinity of its K absorption edge, 9659 eV

E (eV)	f' (e^-)	f'' (e^-)
9559	25.58	0.5123
9659	18.43	3.892
9759	25.93	3.832

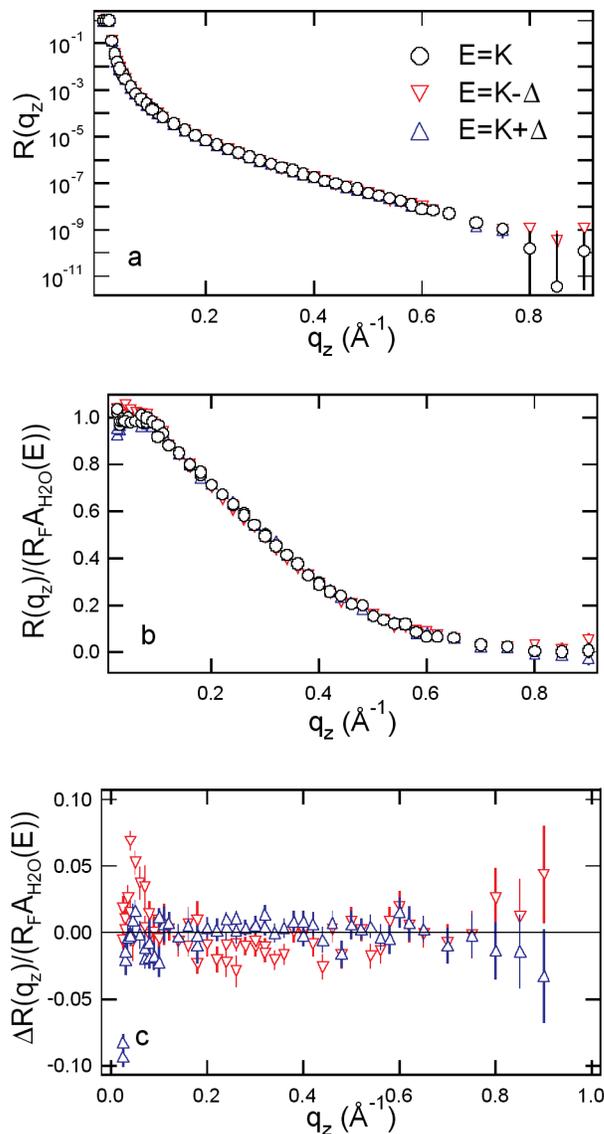


FIG. 1. Top graph a: X-ray reflectivity data from a clean water surface collected with the incident photon energies indicated, where $K = K_{\text{Zn}} = 9659$ eV and $\Delta = 100$ eV. Middle graph b: The same data after Fresnel- and energy-normalization. Bottom graph c: Differences between off-edge data sets and the data set collected at the edge energy

by the absorption from Zn foil. Reflectivity was recorded at energies of K , $K + \Delta$, $K - \Delta$, K , where $\Delta = 100$ eV. At each energy, a complete scan for $0.010 \leq q_z \leq 0.7 \text{ \AA}^{-1}$ required about 70 minutes. The trough was translated 5 mm transverse to the beam between scans. The expected changes in the scattering factor of Zn are shown in Table 1 [4].

Results

Measuring the reflectivity from water, which is well understood, allowed us to verify that the LSS alignment could be maintained as energy was changed. Figure 1 demonstrates that we achieved adequate control of the LSS. The middle graph (Fig. 1b) shows that the Fresnel-normalized reflectivity, which for a clean interface is well described by a Gaussian, could be superimposed after normalization by a factor accounting for small systematic energy-dependent effects, $A_{\text{H}_2\text{O}}(E) \leq 1.1$.

Low concentrations of ZnCl_2 or BaCl_2 in the subphase affect the isotherm of AA monolayers significantly (Fig. 2), while higher concentrations make the monolayers very rigid and hard to compress. We chose to work with subphases containing either $10 \mu\text{M}$ of ZnCl_2 or BaCl_2 (as a control).

Reflectivity data from the ZnAA monolayer showed that the structure was not stable with time but continued to evolve, even after several hours. The BaAA monolayer was more stable, allowing us to record consistent data at different energies (Fig. 3).

Discussion

The instability of the ZnAA monolayer prevented us from observing energy-dependent changes in the reflectivity data that we could unambiguously attribute to resonant effects. This underscores the importance of a data collection protocol that cycles through the energies of interest and verifies that the structure remains unchanged during the measurement. Results

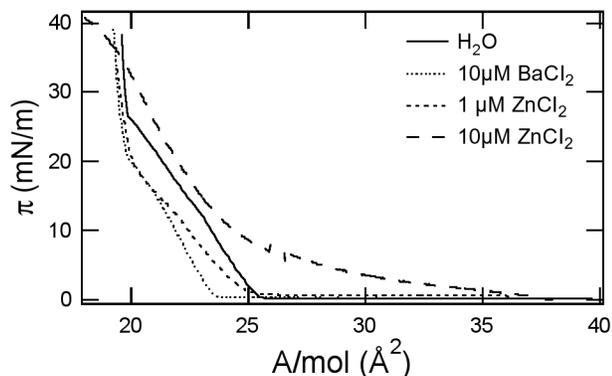


FIG. 2. Isotherms of AA monolayers collected at 22°C with the subphase conditions indicated.

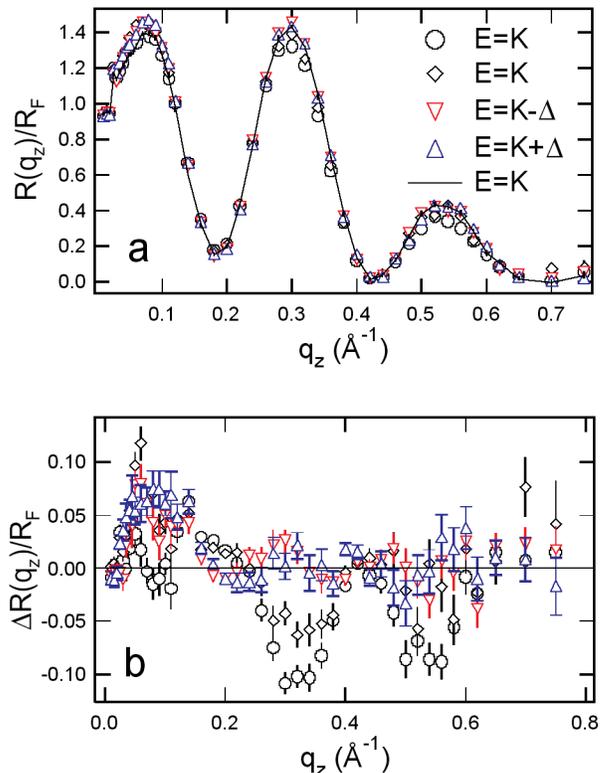


FIG. 3. Top graph a: Fresnel-normalized reflectivity from the control BaAA monolayer collected at different energies. Bottom graph b: Differences between the data sets relative to the last data set collected at the K edge (solid curve in Fig. 3a) show a sensitivity similar to that obtained for a clean water surface (Fig. 1b) when only the off-edge data sets (down-pointing and up-pointing triangles) are considered. Note, however, that the differences between the last scan at the edge energy and the first two scans at the edge energy (circles and diamonds) are more significant, indicating that the monolayer structure changed during the measurement.

from the clean water surface and the control BaAA monolayer demonstrate that the LSS can be kept in alignment as energy is changed. With a more stable monolayer system, we should be able to observe resonant effects in reflectivity from monolayers at the air-water interface (see our other activity report).

Acknowledgments

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References

- [1] J.M. Pachence, R.F. Fischetti, and J.K. Blasie, *Biophys. J.* **56**, 327-337 (1989).
- [2] J.K. Blasie and J. Stamatoff, *Annu. Rev. Biophys. Bioeng.* **10**, 451-458 (1981).
- [3] D. Vaknin, P. Krüger, and M. Lösche, *Phys. Rev. Lett.* **90**, 178102 (2003).
- [4] B.L. Henke, E.M. Gullikson, and J.C. Davis, *X-ray Interactions: Photoabsorption, Scattering, Transmission, and Reflection at $E = 50\text{-}30000$ eV, $Z = 1\text{-}92$* , Atomic

Data and Nuclear Data Tables **54**(2), 181-342 (July 1993), http://www-cxro.lbl.gov/optical_constants/pert_form.html.

- [5] J. Strzalka, E. DiMasi, I. Kuzmenko, T. Gog, and J.K. Blasie, "Resonance reflectivity from Langmuir monolayers of fatty acids incorporating a covalently bound resonant species," STRZALK2 in Advanced Photon Source Activity Report 2002 Database, http://www.aps.anl.gov/aps/frame_home.html.