Molecular Imaging

Applications of MAC Mode AFM

in Biology, Pharmaceutical, and Other Bio-Related Industries

Application Note

Judy Y. Zhu

Ever since its invention, atomic force microscopy (AFM) has revolutionized the field of interfacial surface science by enabling direct, high-resolution visualization of surface morphology in various solutions and gas environments. With the advent of new technologies that simplify the preparation and handling of samples, the use of AFM is becoming increasingly widespread in biological research. The potential of Agilent's MAC Mode AFM for accelerating drug development is also tremendous.

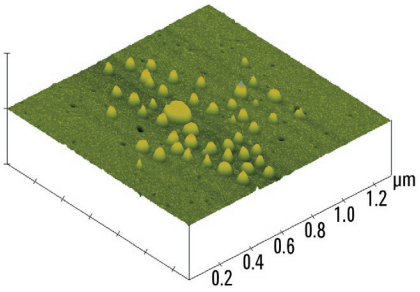

Figure 1. MAC Mode image (scan size = 1.15 µm) of dimerystic phosphotydal-choline (DMPC) liposomes in phosphate buffer. The liposomes diameters range from 50 to 200 nm.

Highlighted below are a few examples of the many MAC Mode applications performed under ambient conditions, in solvents, or under other controlled conditions (e.g., 37 °C) using Agilent atomic force microscopes.

Liposomes and Other Drug Carriers

Liposomes are widely used protein, DNA, or drug carriers. Liposome structure is crucial to function; this is often measured using light-scattering techniques, an approach that only yields size information. When liposomes are deposited on a suitable substrate in buffer, the size and shape can be directly visualized with AFM. The surface structures of other drug carriers, such as lactose crystals, can also be studied with AFM under various conditions. AFM, therefore, can greatly aid research in the pharmaceutical industry.

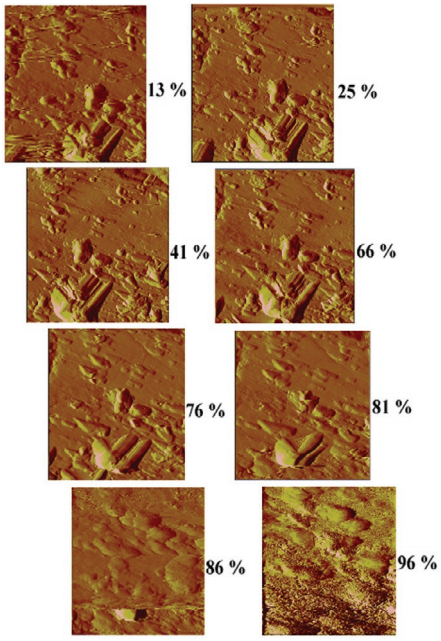

Figure 2. A series of AFM images (scan size = 5 µm) of a lactose crystal's humidity, ranging from 13 to 96%. The crystals are often used as an inhaled drug carrier. Surface structures appear to "melt" at about 80% humidity. (Sample courtesy of Drs. Gary Ward and Mike Maniaci of Dura Pharmaceuticals.)

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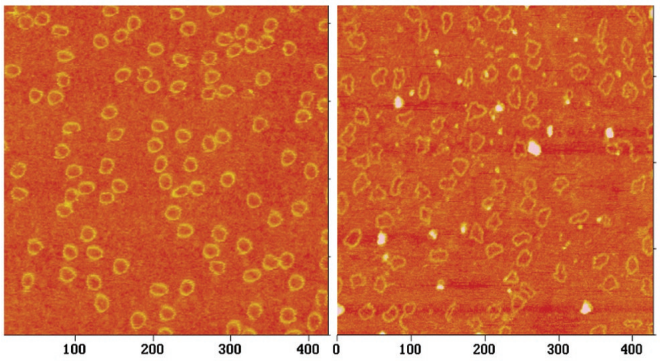

Figure 3. MAC Mode images (scan size = 425 nm) of 168-bp DNA minicircles in H2O containing MgCl2 (left) and ZnBr2 (right). The average width of the double helix measured is about 3.5 nm. In MgCl2, the DNA looks circular, whereas in ZnBr2, the DNA is kinked. Although DNA kinking *in vivo* as a function of ionic conditions has been suggested by other techniques, but MAC Mode images provide direct evidence for this behavior.

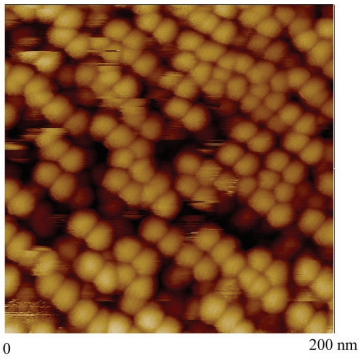

Figure 4. A MAC Mode image (scan size = 200 nm) of ferritin, an iron-storage protein, in H_2O. The size of the ferritin was measured to be 10 nm, demonstrating a resolution of about 1 nm.

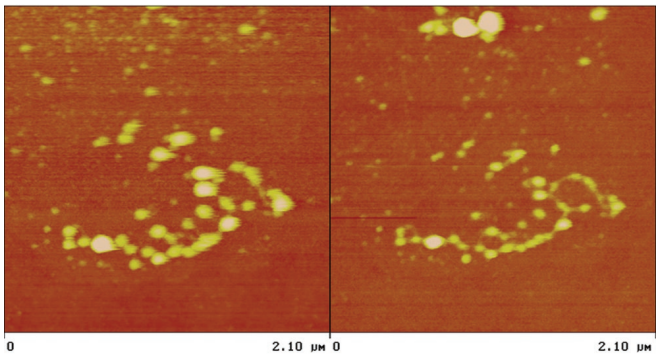

Figure 5. Mac Mode images of chicken chromatin in buffer without (left) and with (right) Mg2+. A flow-through liquid cell was used to change the buffer. Upon the addition of Mg2+, the nucleosomes condensed and their height was reduced by approximately 25%.

Structural and Molecular Biology

Due to its high resolution and ability to image in fluid, MAC Mode with AFM has gained acceptance in the fields of biophysics, structural biology and molecular biology. AFM can give detailed structural information about DNA, RNA, proteins, lipids, live cells and sub-cellular structures in native environments. When combined with electrochemistry AFM is a powerful tool for studying membrane proteins and ion channels.

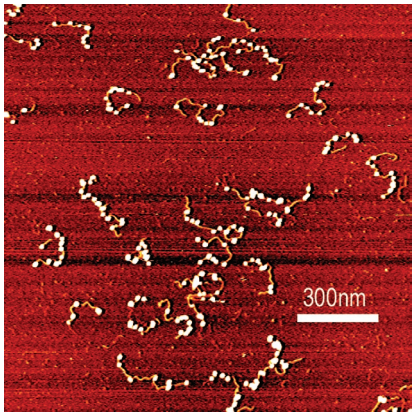

Figure 6. A MAC Mode AFM image of Mouse Mammary Tumor Virus (MMTV) promoter chromatin. The image shows the typical "beads on a string" pattern for chromatin. Individual DNA and histone protein clusters are clearly resolved in the image. The sample was prepared by covalently attaching the histone proteins to chemically modified mica. Imaged in PBS buffer. Scan size 1.5 µm x 1.5 µm. (Hongda Wang, ASU)

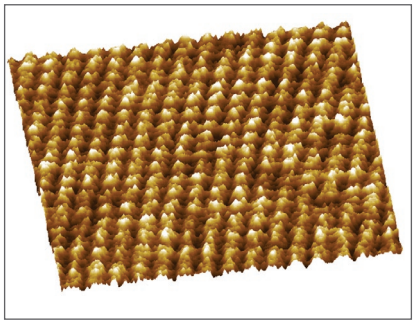

Figure 7. A MAC Mode AFM image of bacterial cell S-layer proteins. Imaging was performed in a PBS buffer solution using the liquid cell. Scan size 250 nm x 250 nm

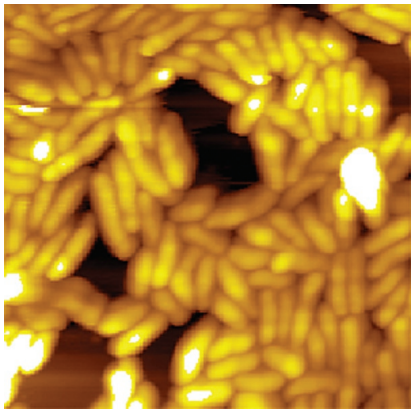

Figure 8. Mac Mode image of Rhodopseudomonas palustris. The results demonstrate the power and utility of AFM in the characterization of microbiological specimens, including its potential for detection and diagnosis of pathological conditions in living cells. Scan size 11 µm x 11 µm.

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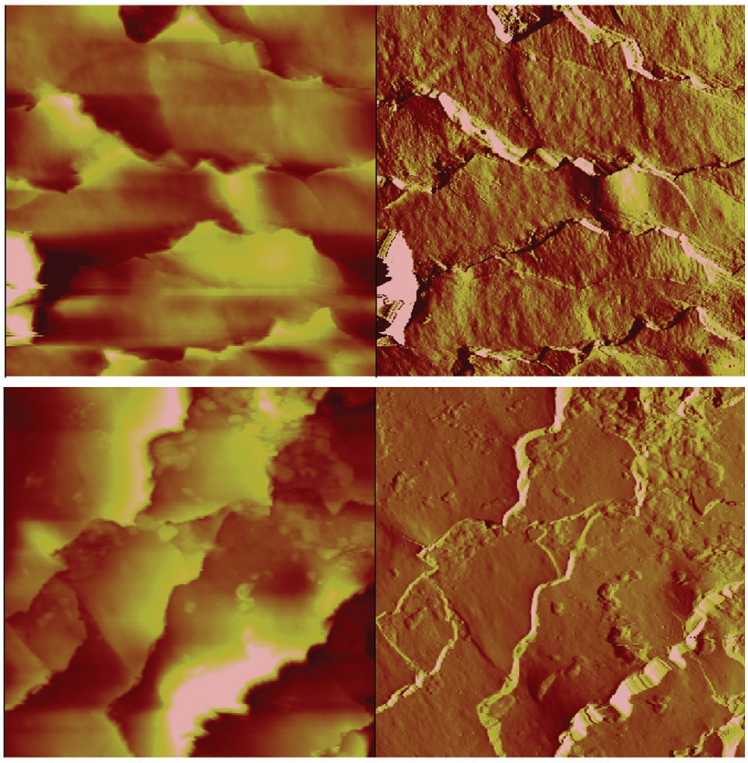

Figure 9. AFM images (scan size = 22 µm) of human hair before (left) and after (right) treatment with shampoo. Studies of this type can help design better hair products.

Cosmetics Industry

In the cosmetics and hygiene industries, material surfaces (e.g., fabric and human hair) are routinely studied before and after applying a cleaning agent (e.g., shampoo). AFM provides a quick way to obtain high-resolution images of material surfaces with less sample preparation.

MAC Mode application examples.

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