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Recognizing and Avoiding Artifacts in AFM Imaging

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1. Introduction

Images taken with the atomic force microscope (AFM) originate in physical interactions that are totally different from those used for image formation in conventional light and electron microscopy. One of the effects is that a new series of artifacts can appear in images that may not be readily recognized by users accustomed to conventional microscopy. Because we are addressing ourselves to novices in this field, we would like to give an idea of what can happen while taking images with the AFM, how one can recognize the source of the artifact, and then try to avoid it or minimize it. Essentially, one can identify the following sources of artifacts in AFM images: the tip, the scanner, vibrations, the feedback circuit, and image-processing software.

2. Tip Artifacts

The geometrical shape of the tip being used will always affect the AFM images taken with it. Quite intuitively, as long as the tip is much sharper than the feature under observation, the profile will resemble closely its true shape. Depending on the lateral size and height of the feature to be imaged, both the sharpness of the apex and the sidewall angle of the tip will become important. In general, the height of the features is not affected by the tip shape and is reproduced accurately, whereas the greatest artifacts are evident on the lateral geometry of objects, especially if they have steep sides.

Avoiding artifacts from tips is achieved by using the optimal probe for the application: the smaller the size of the object, the sharper the tip. A notable exception arises in the case of high-resolution imaging on ordered crystals, where often better images are obtained with standard tips. This can be explained by realizing that at this dimensional scale the measurable radius of curvature of the tip is not in fact involved in the imaging process, but instead smaller local

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protrusions on the apex of the probe will be the real tip (or tips) effectively taking the image.

Further understanding of AFM tip properties and related artifacts can be gathered from the vast literature on the subject, together with a variety of methods for their correction ***(1–9)***. Specific artifacts, depending on the mode of operation, have been investigated and explanations have been proposed ***(10–14)***.

Because we are now interested in showing a general overview of the subject for beginners in the field, we shall have a look at the main tip artifacts in a very simple way.

2.1. Features Protruding on the Surface Appear Larger Than Expected

In **Fig. 1**, the different profiles were obtained using a dull or a sharp tip when scanning a surface feature. In addition to sharpness, the geometrical shape also is important: a conical tip will affect the lateral shape of the feature less than a pyramidal one. Very small features, such as nanoparticles, nanotubes, globular proteins, and DNA strands, will always be subject to image broadening, so that the measured lateral size should be taken as an upper limit for the true size. Note that in all these cases the height of the sample will be reported accurately.

2.2. Repetitive Abnormal Patterns in an Image

When the size of the features on a flat surface is significantly smaller than the tip, repetitive patterns may appear in an image. Spherical nanoparticles or small proteins may assume an elongated or triangular shape reflecting the geometry of the apex of the tip. Sometimes a so-called "double image" will appear along the fast scanning direction as a result of the presence on the tip of more than one protrusion slightly separated from one another and making contact with the sample (**Fig. 2**).

2.3. Pits and Holes in the Image Appear Smaller and Shallower

When the tip has to go into a feature that is below the surface, such as a hole, the lateral size and depth can appear too small and the tip may not reach the bottom. The geometry of the probe will dominate the geometry of the sample as is apparent from the line profile shown in **Fig. 3**. However, it is still possible to measure the opening of the hole from this type of image. Also, the pitch of repeating patterns can be accurately measured with probes that do not reach the bottom of the features being imaged.

2.4. Damaged or Contaminated Tips

If the probe is badly damaged or has been contaminated by debris from a less-than-clean sample surface, strangely shaped objects may be observed in

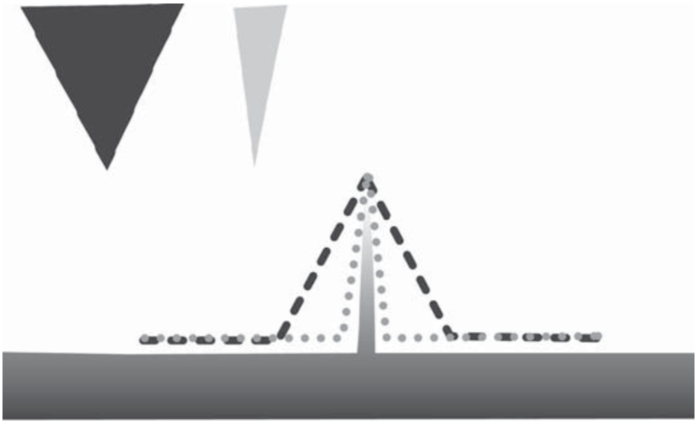

Fig. 1. Traces followed by a dull and a sharp probe as they go over a protruding feature. In such a measurement, the side of the tip will cause a broadening of objects in the image.

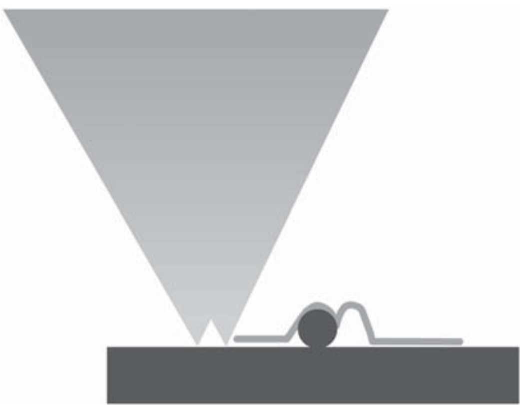

Fig. 2. A double tip will cause a shadow or double image along the scanning direction

the image and difficult to explain. For example, a damaged tip following the geometry of a regular test pattern (as in **Fig. 4**) will produce an asymmetric profile. In the case of contaminants, one often notices an abrupt change of detail contrast during scanning and a blurring of the image. Sometimes the debris particle may partially detach and is dragged along during scanning, leaving a diagonal track on the image that could be erroneously interpreted as a

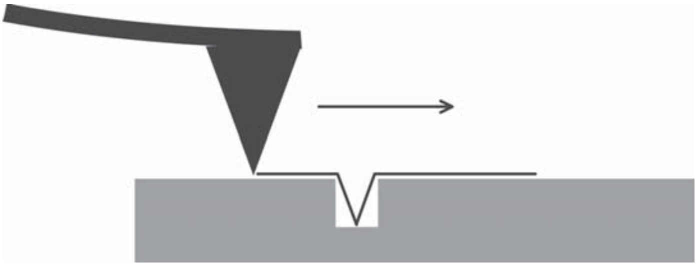

Fig. 3. Because of the width of the tip, the hole will not be faithfully reproduced.

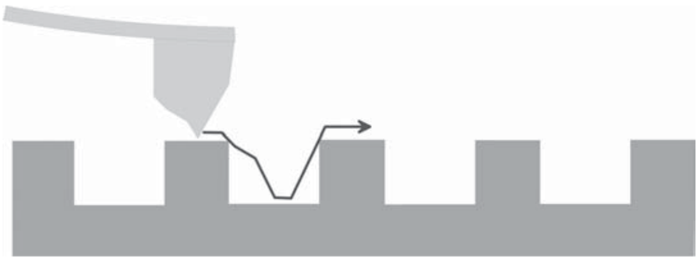

Fig. 4. A badly damaged tip creates artifacts while scanning a regular test pattern.

surface feature. Telltale signs in this case are the instabilities and glitches in the feedback signal that occur each time the particle is dragged along.

3. Scanner Artifacts

Piezoelectric ceramic scanners were one of the breakthroughs that made AFM possible. Their design has been constantly improved, but a number of artifacts still arise from their physical and mechanical properties. One point that must not be neglected is that scanner properties change with time and use. In fact, the piezoelectric material will change its sensitivity to driving signals if it is often used (it will become slightly more sensitive) or if it is left idle (it will depolarize and become less sensitive). The best thing to do is to periodically calibrate the scanner following the manufacturer's instructions.

3.1. Effects of Intrinsic Nonlinearity

If the extension of the scanner in any one direction is plotted as a function of the driving signal, the plot will not be a straight line but a curve similar to the one shown in **Fig. 5**. The nonlinearity may be expressed as a percentage (describing the deviation from linear behavior), and it typically ranges from 2–

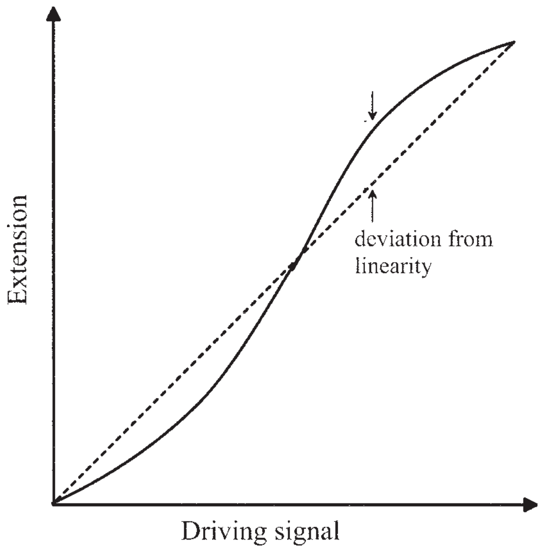

Fig. 5. Plot of the scanner extension vs driving signal. Notice the large deviation from linearity.

25%, depending on the driving signal applied and scanner construction. The effects will be present both in the plane and in the vertical direction.

3.1.1. In the Plane

An AFM image of a calibration grid with periodic structures such as squares will appear severely distorted, with nonuniform spacing and curvature of features, typically appearing smaller on one side of the image than on the other (**Fig. 6**). On a generic sample with no regular pattern the distortion may not be recognizable, but it will be certainly present. Once the scanner is properly linearized, it is also critical that the scanner be calibrated. For example, it is possible for the scanner to be linear but not calibrated. If the calibration is incorrect, then the x and y values measured from line profiles will be incorrect.

3.1.1.1. In Plane Linearization

There are essentially two methods to linearize a scanner in the x and y directions: by software or hardware. Software correction is performed by mathematically modeling the nonlinear behavior of the scanner, finding the parameters for a correction algorithm imaging a known grid, and then applying the algorithm during scanning using the parameters stored in a look-up table. The limits of this method lie in the fact that unfortunately the corrections strongly

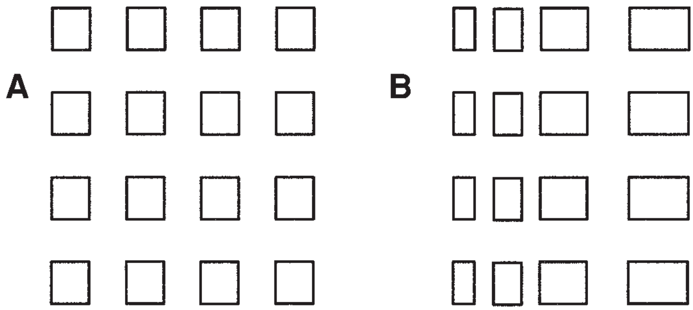

Fig. 6. Distortion of a test pattern caused by scanner nonlinearity.

depend upon the scan speed, scan direction, and offset that have been used during the calibration procedure. When images in normal use are taken under conditions similar to the calibration, the correction will be accurate; otherwise, nonlinearities will be again present. More recently hardware correction for large scanners has become popular ***(15)*** because it gives better results. In this case, the true position of the scanner in the *x* and *y* directions is measured by a sensor during scanning and compared with the intended scanner position. A feedback circuit applies an appropriate driving signal to the scanner in order to attain the desired position.

3.1.2. In Height Measurements

Because the height range of scanners is usually an order of magnitude smaller than the range in the scanning plane, effects of nonlinearity are less severe but still present. To make accurate height measurements with an AFM, it is necessary to calibrate the scanner in the *z*-axis. Often the microscope is calibrated at only one height. This means that if the relationship between the measured *z* height and the actual *z* height is not linear, then the height measurements will not be correct unless the feature being observed has a height close to the calibration measurement (**Fig. 7**). It is also to be noted that although calibration gratings are reasonably easy to make by lithographic techniques, step–height calibration standards are more difficult to obtain, especially for very high-resolution work. Often researchers make their one reproducible height standards for accurate measurements in this range from crystals that have known height steps.

3.2. Effects of Hysteresis

All piezoelectric ceramics display hysteretic behavior, that is, if slowly scanned back and forth cyclically, to the same driving signal does not correspond the same position in the two scanning directions. This can be easily

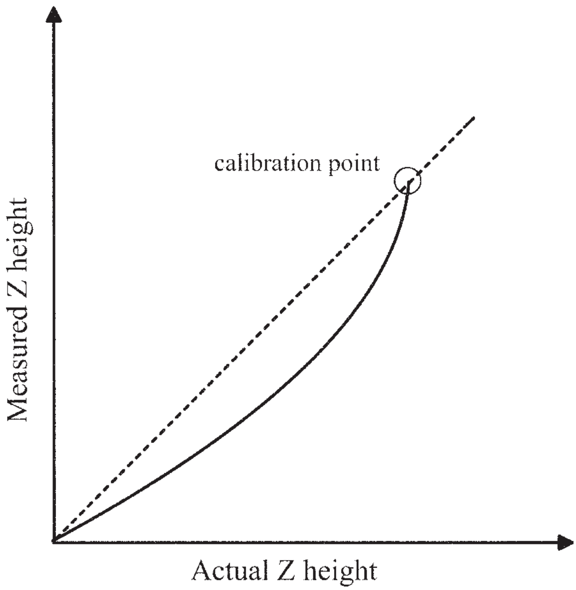

Fig. 7. Quite often, the *z* height response of the scanner is calibrated in only one point. The plot represents the deviation from the true value for measurement of heights that differ from the one at which the scanner has been calibrated.

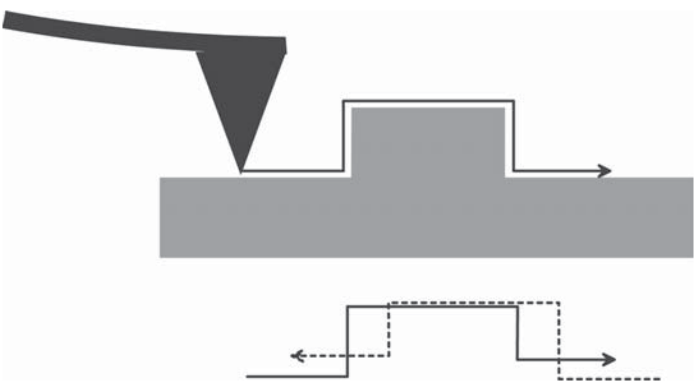

Fig. 8. Effect of scanner hysteresis on a scan (trace and retrace) of a step.

observed by comparing the profiles taken from left to right and in the opposite direction on a feature on the surface of a sample. The result would be like **Fig. 8**, where there is a lateral shift between the two profiles. Notice that an effect is also present in the vertical direction because the contraction and extension response of the scanner to the driving signal will be different, giving rise to an asymmetric step height.

3.3. Effects of Creep

When the scanner is subjected to a very fast variation in the driving voltage, it does not change its position all at once. The dimensional change occurs in two steps: the first step takes place in less than a millisecond, the second on a much longer time scale. The second slower movement is called creep. This causes several effects. Scans taken at different scan rates will have slightly different magnification. If one tries to zoom-up onto a feature, making a smaller scan just after a larger scan, the feature will not be centered and may be distorted in the second image because of creep. On a structure made of parallel lines the effect will be a bending of the lines in the first portion of the scanned image (**Fig. 9**). This is often also called drift, but must not be confused with thermal drift, which is different.

In the vertical direction, creep becomes apparent as an overshoot of the scanner position at the leading and trailing edge of features that have steep sides (**Fig. 10**). This can be often found as a lateral "shading" of protruding features on flat substrates in top view topographical images.

3.4. Effects of Cross Coupling and Sample Tilting

Usually scanners are assembled in the AFM having a free end that is scanned (to which either the cantilever or the sample is attached) and the other end is attached to the microscope body. For this reason the motion of the scanner will follow an arc (spherical or parabolic depending on the type of scanner) and not a plane (**Fig. 11**). The affected images will show a bow, which is especially evident in large scans. This artifact can easily be subtracted by image processing. When very small features have to be detected on flat surfaces, the bow will not allow them to be seen during scanning as the vertical scale of the image would have to adjust to accommodate it: for this reason, the AFM often has the option to subtract the appropriate curve from each line during acquisition, allowing small features to become immediately evident. When there is mechanical or electronic cross coupling between the x and y direction elements of the scanner, this will become apparent in the image of test structures, where the angles between features in the x and y plane will be modified. Mechanical coupling between the piezoelectric ceramics that move the probe in the x or y directions and in the z direction can cause substantial errors when measuring sidewall angles.

Another source of cross coupling arises when the scan direction is not parallel to one of the piezoelectric elements that constitute the scanner. Rotated scans are obtained by sending appropriately mixed driving signals to both the x

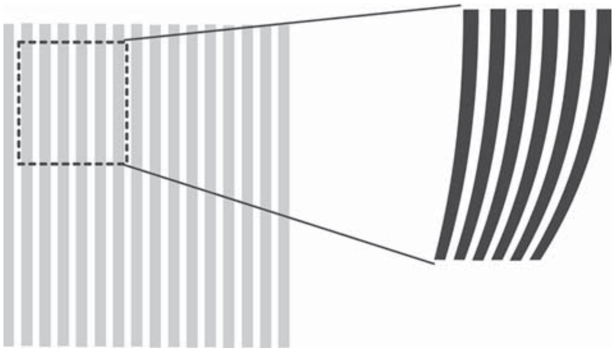

Fig. 9. Effect of creep on a scan performed zooming up onto a detail in a larger image.

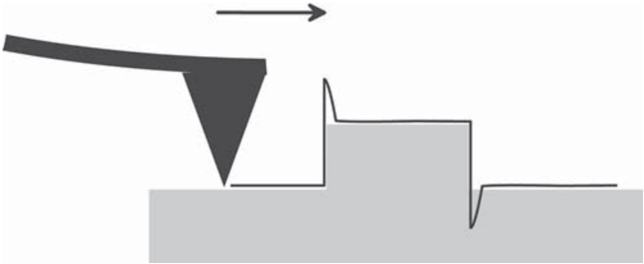

Fig. 10. Effect of creep in the vertical direction: overshooting at the edges of the step.

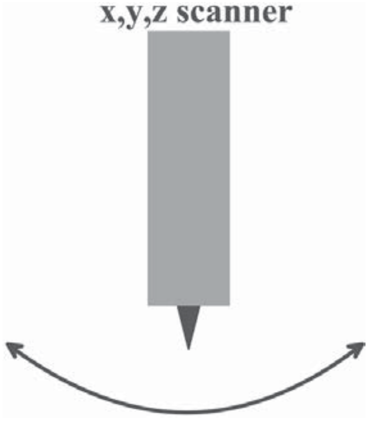

Fig. 11. The free end of the scanner will follow an arc during scanning, creating a bowl-like image. This effect is especially evident on large scans of flat surfaces.

and *y* piezoelectric elements: if they are not both accurately calibrated the image will be affected by a geometrical distortion.

It is useful to add that quite often (in fact, always) the sample will have a plane tilt relative to the motion of the scanner. Although all acquisition software allows for subtracting the tilt during scanning, it is good practice to try and mount the sample as planar as possible so that the piezoelectric element responsible for the vertical movement will operate across a smaller range and hence behaving linearly.

3.5. Thermal Drift

External temperature changes or gradients will affect the AFM and its scanner depending on their mechanical properties. AFMs are built in such a way as to minimize this phenomenon by using special materials and appropriate design, but nevertheless thermal drift can be present. In the case of very high-resolution imaging of atomic structures, it is often necessary to wait some time during scanning before the system will stabilize and stop drifting. Also, electronics and the laser spot on the cantilever can induce drift in measurement settings that need to stabilize in time. In the case of AFMs mounted onto inverted microscopes and eventually equipped with a heated stage, special care has to be taken. When a liquid cell is used and reagents are flowed in during an experiment, the problem becomes acute as temperature changes of even a fraction of a degree can cause large bending of the cantilever. Often, a good ambient air conditioning system can be useful in reducing thermal effects.

4. Vibrations

Because the AFM operates thanks to its very high sensitivity to the small deflections of the cantilever assembly, it is evident that if external vibrations affect the cantilever these will create artifacts in the images. Typically, the artifacts will appear as oscillations. Both acoustic and floor vibrations can excite vibrational modes in an AFM and cause artifacts.

The floor in a building can vibrate vertically several micrometers at frequencies below 5 Hz. The floor vibrations, if not properly filtered, can cause periodic structure in an image. This type of artifact is most often noticed when imaging very flat samples. Sometimes the vibrations can be started by an external event such as an elevator in motion, a train going by, or even people walking in a hallway. A special air table or bungee cords must be used to isolate the AFM from these vibrations. A good idea is also to install the instrument near a corner of the laboratory instead of at the center of a room, choosing if possible the lowest floor in the building.

A person speaking in the same room as the microscope, music, a door that shuts, an airplane going over the building can generate sound waves that will generate artifacts in the AFM images. Some instruments have as an option an acoustic hood or enclosure to isolate the AFM from external noise.

5. Effects of Feedback and Other Parameter Settings

Depending on the mode of operation, several parameters have to be set by the user to obtain the best images. Among these, one can find deflection set point (in contact mode), oscillation amplitude and dampening (in AC modes), feedback gain (sometimes separated into a proportional gain setting and integral-derivative setting), low pass filters, scan speed, and so on.

The setting of these parameters is a trial-and-error process. Each time a new sample is put into the microscope, the best values must be searched and during the process many artifacts can be produced in images. Soft samples generally must be imaged at low scan speeds and low interaction forces, otherwise glitches in the scan direction or even sample deformation may occur. Rough samples again need to be imaged slowly, but larger amplitude or deflection might be needed to keep track of the surface. Especially in AC imaging modes (but also in DC mode) special care must be taken in tuning the gain parameters of the feedback. If the feedback loop of a scanning probe microscope is not optimized, the image can be affected. When feedback gains are too high, the system can oscillate, generating high-frequency periodic noise in the image. This may occur throughout the image or be localized to features with steep slopes. However, when feedback gains are too low, the tip cannot track the surface, and features will be distorted and smeared out. On large objects with sharp slopes, an overshoot can appear in the image as the tip travels up the slope, and an undershoot can appear as the tip travels down the slope. Taking a force-vs-distance curve to ascertain the presence of adhesion forces or other effects can help to guide the choice of imaging parameters.

6. Image Processing

Image processing is readily available in AFM as the data is stored digitally on a computer disk. One can easily access routines for flattening, polynomial-line or surface subtraction, removal of bad data, matrix filtering, and three-dimensional representation with sophisticated rendering. Often some kind of processing will be necessary to analyze data and compare it with other results, but care must be taken to avoid introducing artifacts. The most common ones stem from careless use of the powerful image processing tools available. For example, as we have seen in **Subheading 3.4.**, nearly all images are affected by a tilt and by a bow introduced by the scanner geometry. If the wrong curve fit is applied or if large features are not excluded from the surface subtraction parameter computation (all image analysis software allow to include or exclude surface area portions from the computation), distortions will be introduced. This is particularly true with line-by-line curve fit and subtraction.

Low-pass filters, although capable of reducing noise in the data, will introduce smoothing of sharp features and, in the worst cases, delete smaller details. Fourier transform and power spectrum filtering if misused can create periodic features that may seem to be atomic structures, wheres in reality they are only noise.

7. Some Guidelines for Artifact Testing

If during a measurement you get suspicious that an image may contain artifacts, here are some things you can do to be sure whether or not they are present:

- Take more that one image of the same area or the same line to ensure that it looks the same. When looking at a single scan line profile during acquisition, look if the traces are identical and stable in time.
- Try changing the scan direction and take a new image. You can do this also on a single scan line looking at the profile and observing directly the difference between the trace and retrace plots.
- Change the scan size and take an image to ensure that the features scale properly.
- Rotate the sample and take an image to identify artifacts induced by the shape of the tip.
- Change the scan speed and take another image (especially when suspicious periodic or quasiperiodic features are present). If they scale, you are looking at periodical noise.

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