PicoLE AC Mode
User's Manual

v1.1

Molecular Imaging

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System Overview

ACAFM is an intermittent contact mode of atomic force microscopy (AFM) in which the cantilever is driven by an alternating current (AC). The AC Mode option, added to the PicoLE system, allows the user to image softer or more fragile samples than is possible with normal contact mode AFM. The Molecular Imaging MAC Mode Controller can operate in two different modes; magnetic AC (MAC) and acoustic AC (AAC). In MAC mode, the alternating current running through a coil creates an alternating magnetic field. The field drives the cantilever via a magnetic coating on the back of the cantilever. Typical MAC frequencies are on the order of 80kHz. In AAC mode, a voltage that oscillates on the order of 300kHz drives a piezo crystal in the scanner nose. The drive frequency in AAC mode is based on the natural resonant frequency of the cantilever and, as such, may vary from cantilever to cantilever. In MAC Mode, the cantilever can be used at frequencies below that of resonance. At the drive frequency, the tip moves up and down with a measurable amplitude when it is unaffected by (distant from) the sample surface. The system detects changes in the amplitude first, as the tip approaches the surface and then, with greater pressure, as the tip reaches the surface and atomic repulsion effects become dominant. An intermolecular force versus distance curve is shown in Figure 0.1 below.

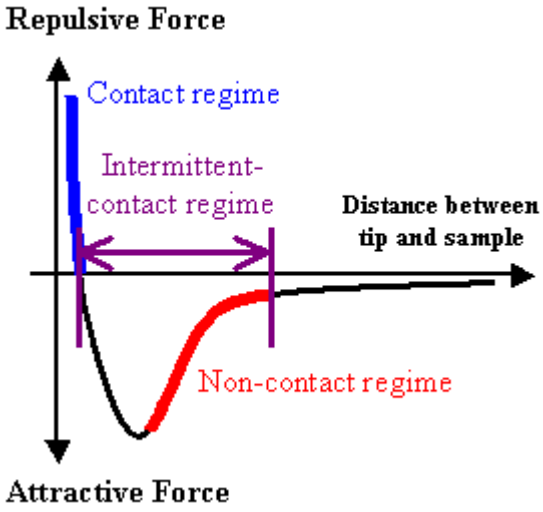

Figure 0.1 (Intermolecular force-distance curve)

When the tip-sample force is attractive, the amplitude increases and when the tip-sample force is repulsive, the amplitude is decreased. The Z-axis piezo adjusts to maintain the setpoint amplitude and, thus, generates the topography data as the cantilever is scanned across the sample surface.

Chapter 1: Initial Setup

Note: The use of the term AC Mode refers to either MAC Mode® or Acoustic AC® Mode. The terms MAC Mode and Acoustic AC are used only when there is a need to differentiate between the two modes of imaging.

List of MAC Mode Components

- MAC Mode controller
- Acoustic AC Nose Assembly
- MACleversTM
- AAC Probes
- MAC Mode Sample stage
- DB25 to DB25 – connects AC Mode Controller to Head Electronics Box.
- BNC to BNC – connects AC Mode Controller to Head Electronics Box.
- Power Cable – connects AC Mode controller to power source.
- RS-232 (serial) cable – connects AC Mode controller to computer.
- MAC/EC cable – connects MAC Mode sample stage to microscope base.

Please contact MOLECULAR IMAGING if any of these items are missing.

Connections

See Figure 2.2 in the next chapter to reference the connections described below. The terms "male" and "female" refer to the connector fittings, and not to the pins within the fittings.

Power Cord

Insert the power cord supplied with the AC Mode package into the back of the AC Mode controller. Do not power on the controller at this time.

PicoLE Head Electronics Box Connection

Connect the female end of one of the DB25 cables to the bottom PICO SPM I connection on the AC Mode controller. Be sure the cable fits snugly and is not loose; if the connection is loose this may cause serious problems while imaging.

After connecting the cable to the AC Mode controller, connect the other (male) end to the CONTROLLER connector on the PicoLE Head Electronics box.

Connect the BNC cable to the MAC port on the AC Mode Controller, making sure the connector twists and locks into place, then connect the other BNC end of the cable to the MAC connector on the PicoLE Head Electronics box. Mention AAC to AAC BNC connections for AAC Mode operation.

If imaging in MAC Mode, the microscope must be connected to the sample plate using the MAC/EC cable. Plug the round jack into the underside of the microscope, and the 6 pin connector into the sample plate. If using Acoustic AC, the signal travels through the normal connections and this connection is not necessary.

Serial Port Connection

For most users, the AC Mode controller will be controlled using a standard RS-232 serial link. Connect to the computer COM1, COM2, or COM3. The default is COM1, but this setting can be changed in the PicoScan software. See **Advanced** under **Chapter 3: AC Mode Software Controls** below for more information.

PicoScan Controller Connection

Connect the male end of the other DB25 cable to the top PICO SPM I connector on the AC Mode controller. Be sure the cable fits snugly and that it is not loose; this too can result in serious malfunctions while imaging.

After connecting the cable to the AC Mode controller, connect the other (female) end of the DB25 cable to the PICOSPM connector on the PicoScan™ 2100 controller. If using a PicoScan 2500 or 3000 controller, use the connection labeled PICOSPM I.

Note: It is not possible to do both MAC Mode and Acoustic AC Mode imaging at the same time.

Chapter 2: The AC Mode Controller

The AC Mode controller is the driving force behind AC Mode. It is here that the drive signal is produced and controlled. After preparing and mounting the sample it is now time to turn our attention to the AC Mode controller. If using an older AC Mode box (with only 25-pin connectors), refer to the original manual. Note that the AC Mode controller must be plugged into the wall.

Front Panel

Figure 2.1 (Front panel of the MAC Mode Controller)

The meter on the front panel displays the amplitude of the cantilever in volts. To convert it to actual amplitude, use this equation:

$$Amplitude(nm) = \frac{MeterVoltage(V)}{20 \cdot InputGain} \cdot 4.3 \cdot Sensitivity(\frac{nm}{V})$$

- ***MeterVoltage***: The number displayed on the meter.
- ***InputGain***: The input gain setting in the **AC Mode Controls** window in PicoScan. See **Main** under **Chapter 3: AC Mode Software Controls** below for more information.
- ***Sensitivity***: The PicoScan controller pre-amp conversion coefficient. This is calibrated in PicoScan by taking the inverse of the Force vs. Distance contact region slope in contact AFM mode. If amplitude data are crucial, please note that the sensitivity should be recalibrated each time the laser spot is adjusted on the cantilever. See the PicoScan manual for more information.

To calibrate amplitude based on the signal displayed in the software (on the amplitude monitor), use this equation:

$$Amplitude(nm) = -\left[\frac{(MonitorVoltage(V) - 4.3xS.P.(V))}{20 \cdot InputGain} \cdot Sensitivity(\frac{nm}{V})\right]$$

- ***MonitorVoltage***: The value displayed on the amplitude monitor in PicoScan.
- ***S.P.***: The amplitude setpoint in the **Servo Controls** window in PicoScan.

The detector electronics act as a low pass filter with a 3dB point of 320 kHz. Take this into account when calculating high-frequency amplitudes.

Rear Panel

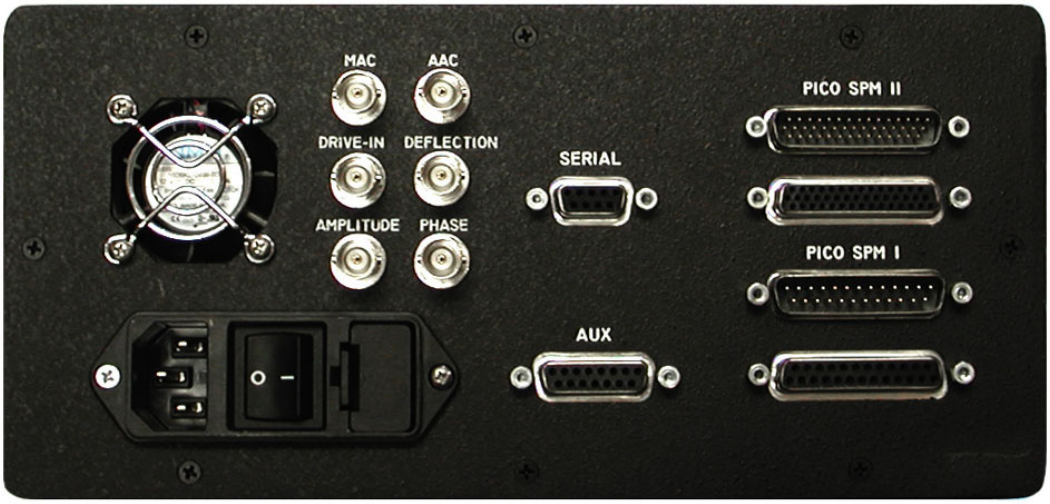

Figure 2.2 (Rear panel of the MAC Mode Controller)

MAC

Drive output for MAC Mode. Connect to the BNC cable that plugs into the MAC connection on the PicoLE Head Electronics box.

AAC

Drive output for Acoustic AC. Connect to the BNC cable that plugs into the AAC connection on the PicoLE Head Electronics box.

Drive In

This is for an auxiliary input signal to be summed into the AC Mode drive signal.

Deflection

Average position of the laser spot on the photodiode. The AC Mode box generates this signal by putting the raw input from the photodiode through a low pass filter.

Amplitude

This is the output from the lock-in amplifier representing amplitude. The signal is the same as the meter but the sign is reversed.

Phase

Phase shift of the center frequency component of the input signal. The conversion factor is 9 degrees per volt.

The **Deflection**, **Amplitude** and **Phase** BNC connections are outputs that can be read through an oscilloscope, or through the software by plugging them into the auxiliary input (**AuxIn**) on the PicoScan Controller.

Serial Port

Connects the AC Mode box to the computer.

Aux

This connector is not currently in use.

Pico SPM II / Pico SPM I

These DB44 or DB25 connectors go to the controller and microscope. The PicoLE microscope uses the DB25 (PICO SPM I) connectors.

Chapter 3: AC Mode Software Controls

Open the **AC Mode Controls** window from the **View** menu in PicoScan. If the **AC Mode Controls** selection is grayed out, enable it from the **Misc. Options** tab of the **Setup (Scanner, Controller etc.)** window. This window can control any PicoSPM I or PicoSPM II AC Mode Controller. If using an older AC Mode Controller, some controls will not appear in the window, as they are controlled on the box itself.

Main

The **Main** window contains the most-used AC controls. All values in this window may be changed using the text box or the up and down arrows on the keyboard.

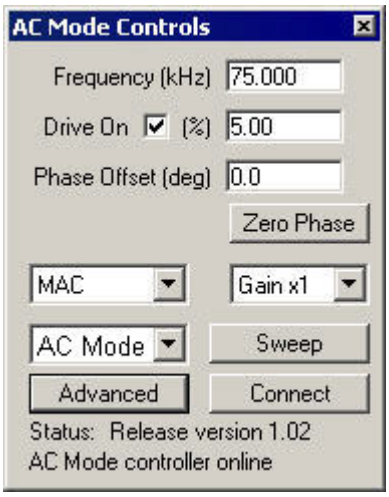

Figure 3.1 (The Main AC Mode Controls window)

- **Frequency (kHz)**: Displays the current drive frequency.
- **Drive (%)**: The amplitude of the drive force as a percentage of maximum available. The **On** checkbox denotes whether the drive is turned on or off (for older controllers, this control is on the box itself as well). The checkbox defaults to checked when PicoScan starts.
- **Phase Offset (deg)**: Allows you to change phase offset.
- **Zero Phase**: This button zeroes the phase offset and sets the phase signal to zero.
- **MAC / AAC**: This pulldown menu allows the user to designate which mode (MAC Mode or Acoustic AC) is being used. This menu will only appear if using a PicoPlus AC Mode Controller. This menu defaults to MAC when PicoScan starts.
- **Input Gain**: The signal into the MAC Mode Controller is multiplied by the factor designated in this pulldown menu. Selecting a higher gain can result in a higher signal to noise ratio for a small signal. Do not use a high gain setting with a large amplitude, because any signal outside the range ±10 V is clipped.
- **Contact / AC Mode**: This pulldown menu allows the user to designate contact AFM or ACAFM mode. This menu only appears if a PicoPlus AC Mode Controller is being used. This menu has the same function as the **AC Mode** button on old AC Mode boxes. This menu defaults to **Contact** when PicoScan starts in AFM Mode, and **AC Mode** when PicoScan starts in AC AFM mode.

- **Sweep**: Brings up the **Amplitude vs. Frequency Spectroscopy** dialog.
- **Connect**: When this button is clicked, PicoScan will attempt to connect to the MAC Mode Controller. Use this button to reinitialize the MAC Mode Controller after powering off.
- **Advanced**: Toggles the window to the **Advanced** functions.

Advanced

The **Advanced** window contains optional and advanced AC mode settings.

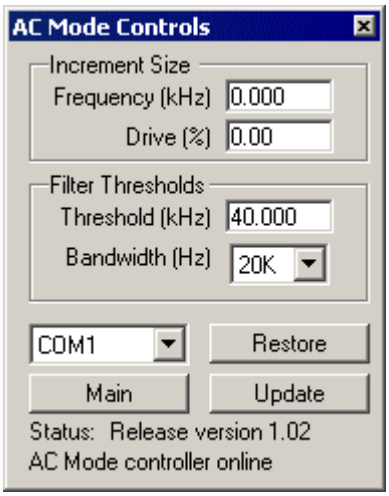

Figure 3.2 (The Advanced AC Mode Controls window)

- **Increment Size**: These numbers determine the increment size when the up and down arrows on the keyboard are used to change **Frequency** and **Drive**.
- **Filter Thresholds**: These controls set the band pass filter of the lock-in amplifier of the MAC Mode Controller for different frequencies. The active band pass filter is the one with the highest threshold below the current drive frequency. It is recommended that the user not change these unless they thoroughly understand the theory behind lock-in amplifiers. Any changes may adversely affect the performance of the system.
- **Restore**: Restores all default settings.
- **COM Port Selector**: This pulldown menu allows the user to designate into which COM port on the computer the MAC Mode Controller is plugged.
- **Update**: Updates the firmware in the MAC Mode Controller.
- **Main**: Toggles the window to the **Main** functions.

Chapter 4: MACAFM Tutorial

A strong point of the PicoSPM system is its AC mode capability. With this mode, one can obtain high-quality images of soft samples in-situ. AC mode means that the cantilever oscillates at a controlled frequency, and PicoScan measures the resulting amplitude and phase changes as the tip of the cantilever interacts with the surface being imaged. Either an oscillating magnetic field or a vibrating piezo crystal induces the cantilever oscillations. In the case of magnetic excitation (MAC Mode), the cantilever is coated with magnetic material, which allows the magnetic field to drive the cantilever. This tutorial will use MAC Mode; however, all the principles are the same for Acoustic AC Mode. This tutorial will use a Large scanner and a Type II MAClever ($C \approx 3$ N/m) to image a gold-on-mica substrate. This tutorial should only be done after completion of the AFM tutorial in the PicoScan software manual. If the images obtained are of unsatisfactory quality, try using the PicoIC™ isolation chamber. This tutorial is written for a system with a PicoLE microscope and a MAC Mode Controller.

Hardware

- AC Mode controller
- MAC Mode Sample plate
- AFM scanner
- MAC Mode AFM cantilever
- Tweezers
- Sample
- Two-sided tape
- Photodiode detector

System Setup

Hardware Setup

Set up the microscope as detailed in the **PicoLE System module**. Make the hardware connections that are described in **Chapter 1: Initial Setup** (page 3) of this module. Note the MAC/EC cable must connect the sample plate to the microscope in order to use MAC Mode. The **Contact/AC Mode/CS AFM Switch** on the **PicoLE Head Electronics Box** should be turned to AC Mode. Be sure to use a MAC sample plate, as the magnetic core of the standard sample plate will interfere with the MAC Mode drive field. See Figure 4.1 below.

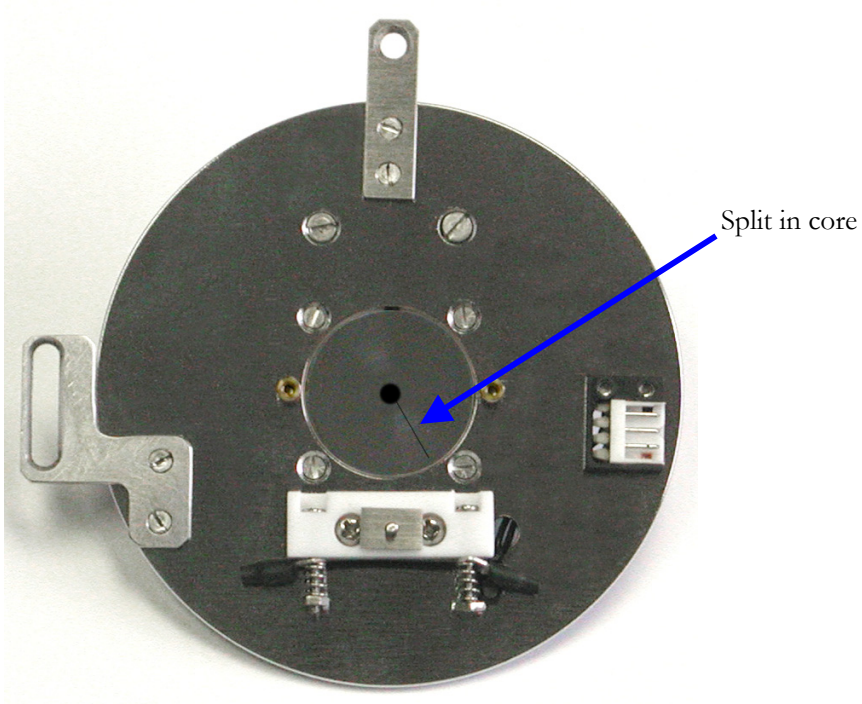

Figure 4.1 (The MAC Mode sample plate has a split in the core that prevents the formation of an induced magnetic counter-field)

Software Setup

PicoScan software should have already been installed on the computer. Use the following steps as a guide to become familiar with the system settings required to make good images.

1. Click **View** on the main menu bar and select **Setup (Scanner, Controller etc.)**.

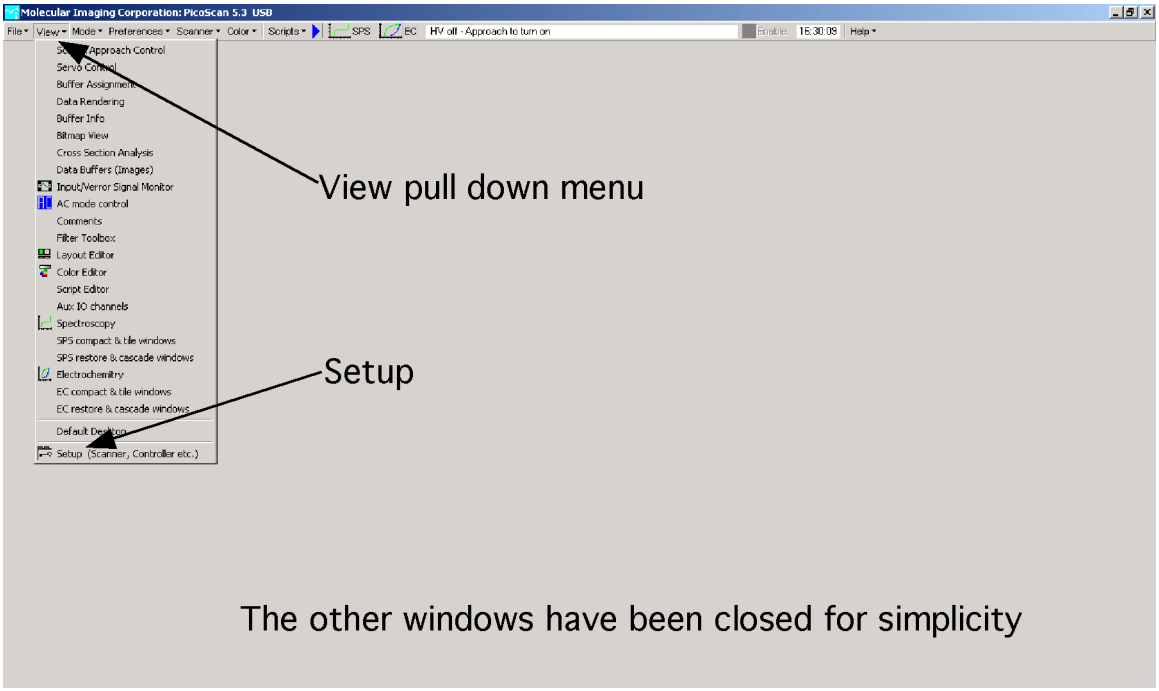

Figure 4.2

The setup dialog will open to the **Scanner & Preamp Calibration** tab

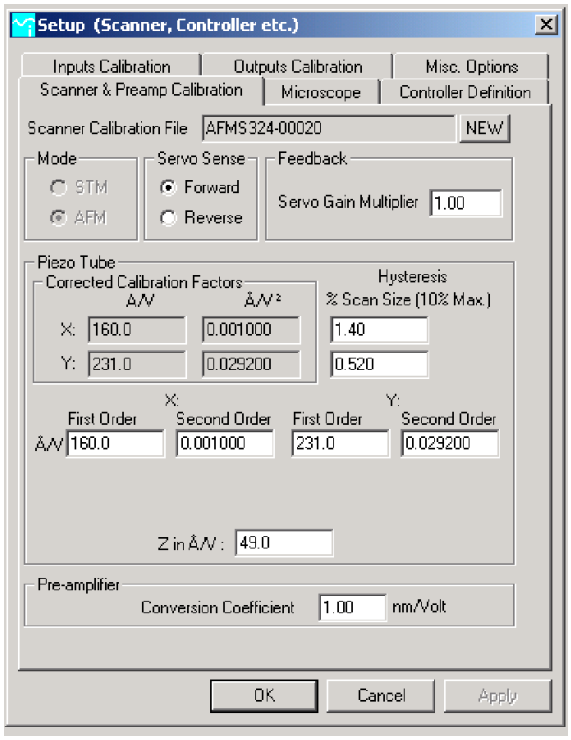

Figure 4.3

2. Click on the **Microscope** tab.

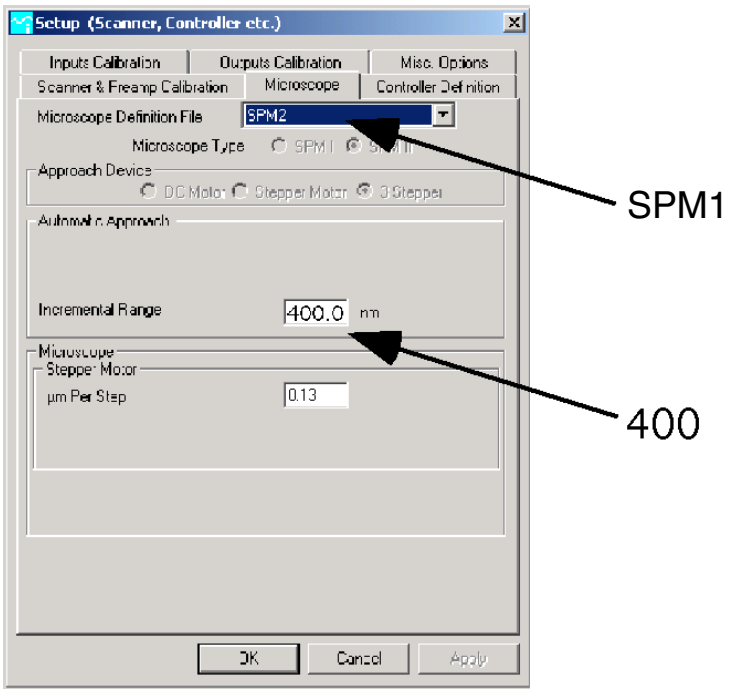

Figure 4.4 (Make sure SPM1 is selected in order for the system to communicate properly with the PicoLE microscope)

3. Select SPM1 as the **Microscope Definition File**. Enter a default value of '400' into the Incremental Range.
4. Select the **Misc. Options** tab. Then select the **AC Mode Controller Supported** box. Click **OK** to accept the changes and close this dialogue.

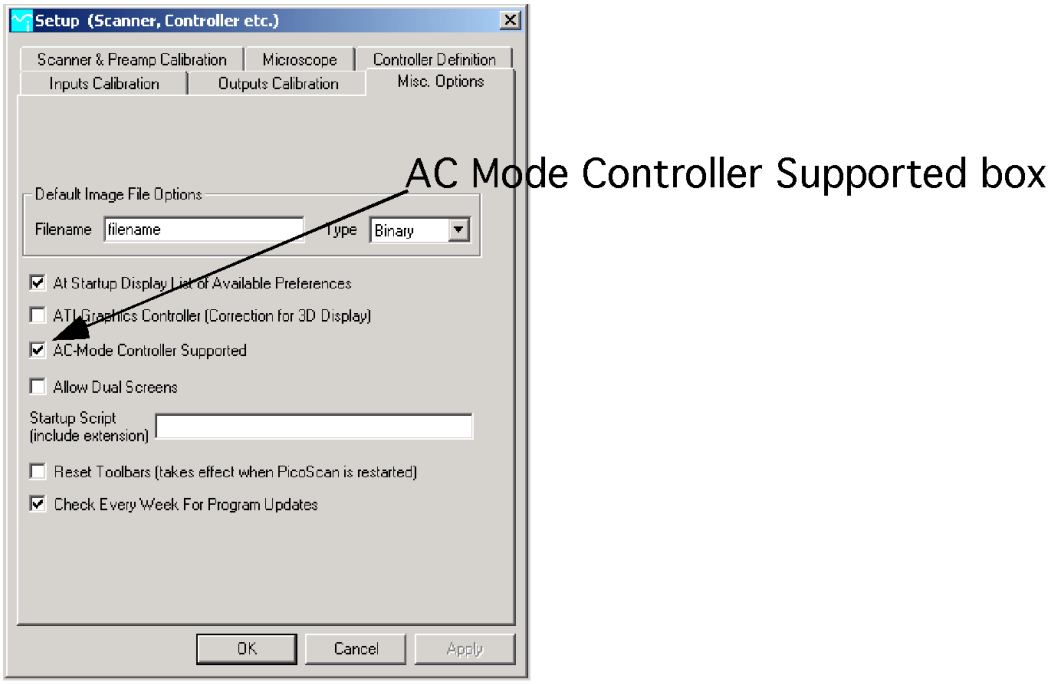

Figure 4.5

5. Select **Live Scan** from the **File** drop-down menu on the **Main Toolbar**.

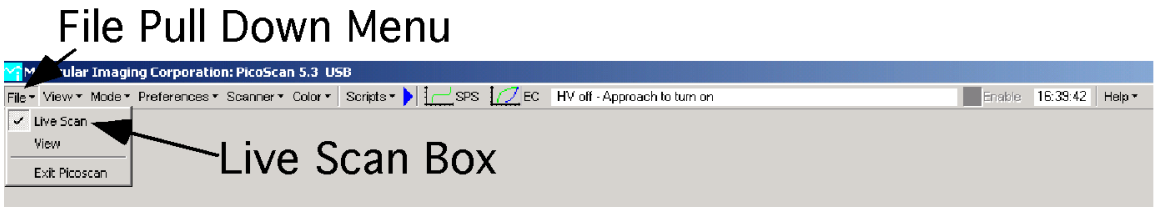

Figure 4.6

6. Under **Mode** on the main menu, select **AC AFM**.

Figure 4.7

7. Under **Scanner** on the main menu select the scanner calibration file that matches the serial number of the scanner being used. See the diagram below for the location of the serial number.

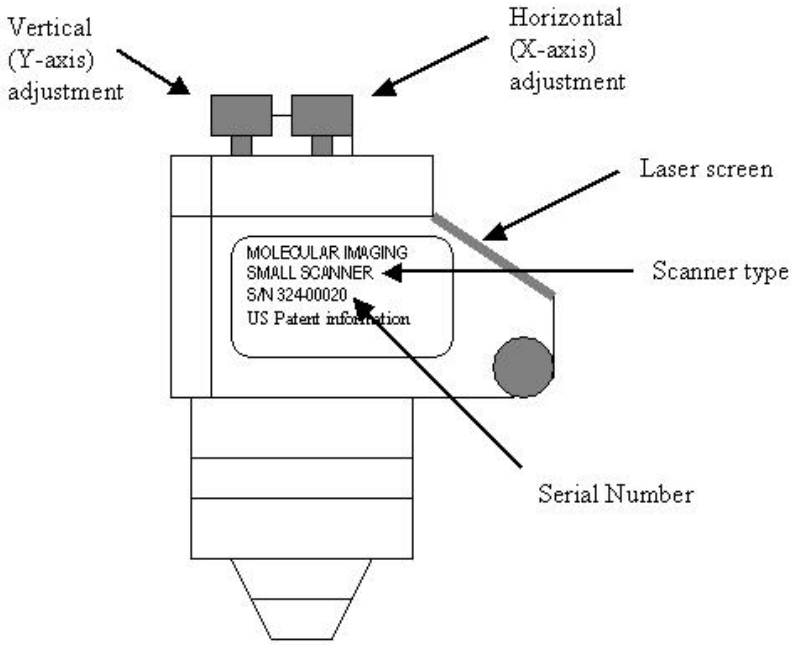

Figure 4.8

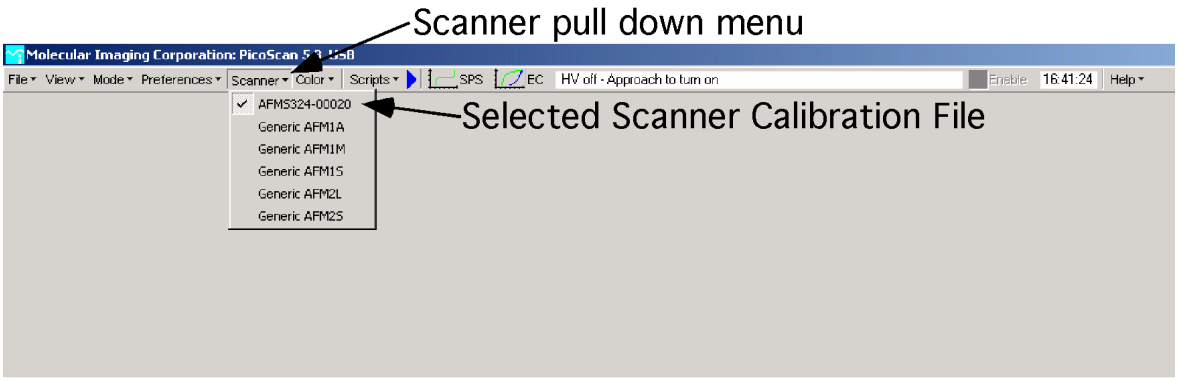

Figure 4.9 (Select the calibration file that matches the serial number of the scanner)

Tuning the Tip

8. Under **View** on the main menu, select **AC Mode Controls**, then perform the following;

- Select **MAC** (as indicated in Figure 4.10 below) to perform a Magnetic AC scan using a MAC tip
- Select **Drive On.**
- Enter 5 for the drive %.
- Set the **Gain** to 1x.

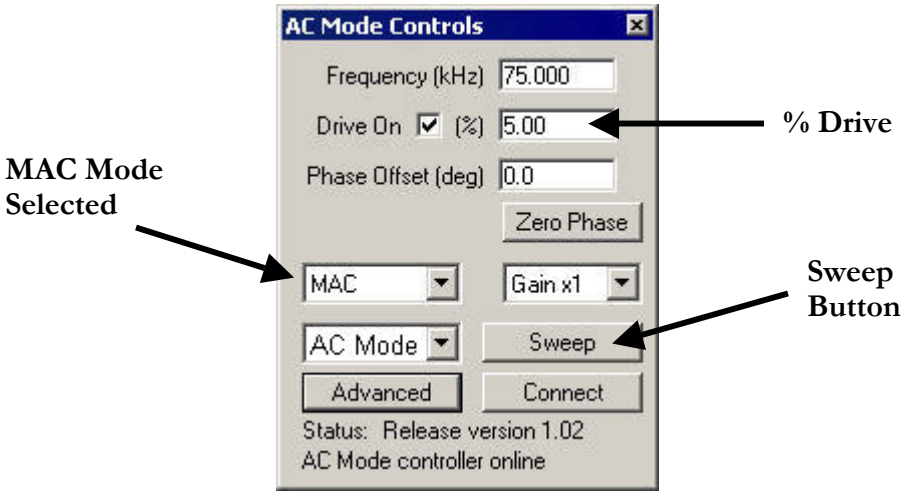

Figure 4.10

9. Click on the **Sweep** button. The AFM AC mode Frequency Plot will now appear.

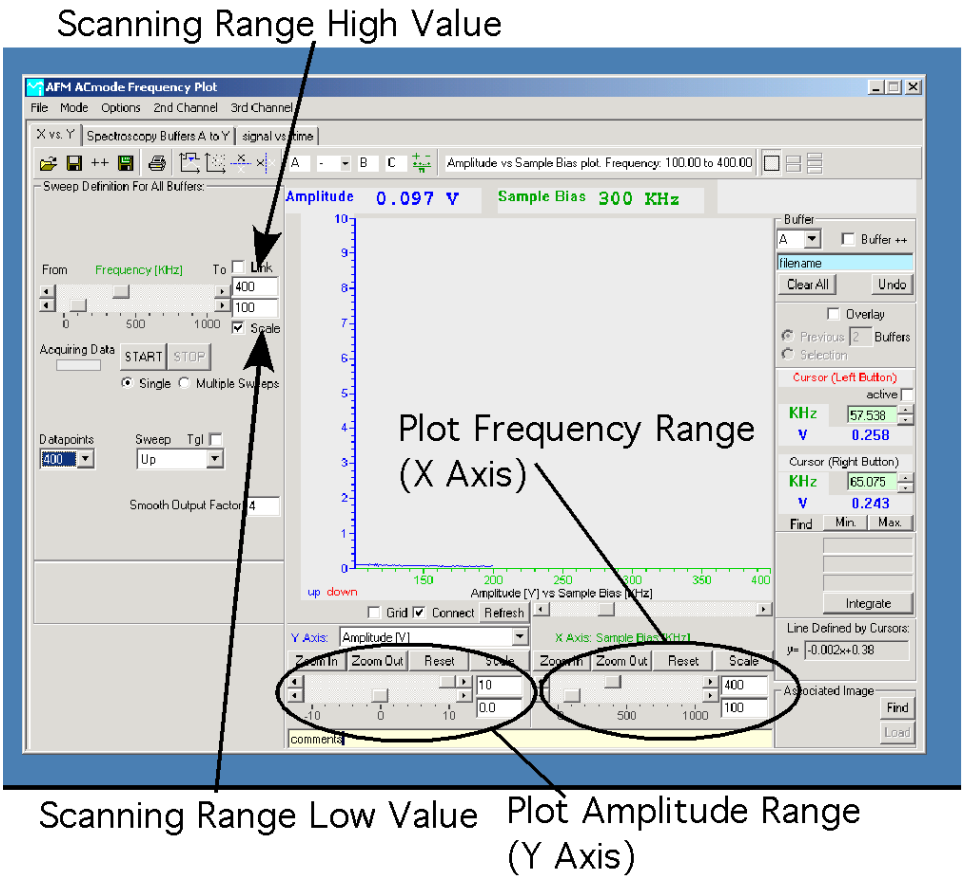

Figure 4.11

10. The resonance frequency of the cantilever must be empirically determined by performing a full-scale frequency sweep.

- Select the scale check box directly under the Frequency Range values.

- Drag the top Frequency slider completely to the right and the bottom Frequency slider completely to the left.
- Click the START button.
- The Resonance peak indicates the resonant frequency of the cantilever (shown in Figure 4.12 below).

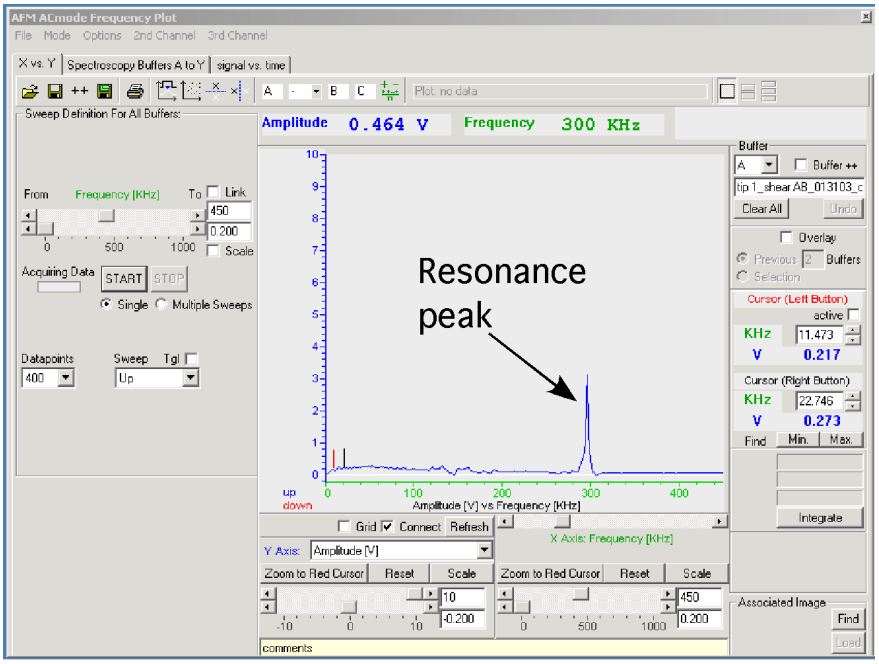

Figure 4.12

- Zoom in on the resonance peak displayed on the frequency plot by adjusting the two Frequency sliders until the resonance peak is centered in the plot.
- Click on the START button in order to initiate another frequency sweep.
- Repeat the above procedure until the frequency range of the sweep is less than 10kHz.
- If off scale, use the software controls to reset the drive to a lower value. If the amplitude is greater than 10 volts, the electronics won't work correctly.

11. Check the **Active** check box in the Frequency plot window. See Figure 4.13 below.
12. Update the drive frequency of the AC Mode control by left clicking slightly to the left of the resonant peak maximum.

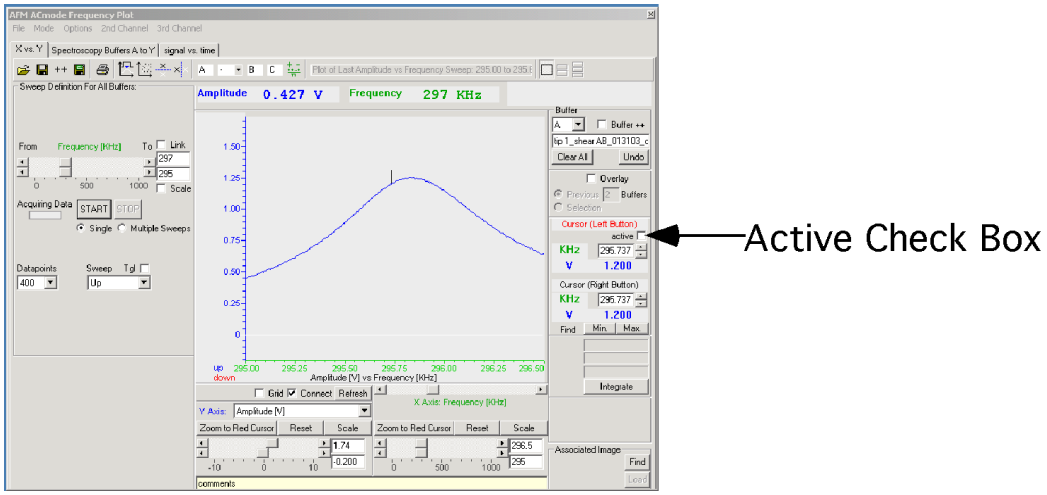

Figure 4.13

13. After the drive frequency has been updated, adjust the drive percentage so that the amplitude, displayed on the front of the AC Mode controller box (the hardware unit, not to be confused with the **AC Mode Controls** window in the software), is 5 ±1 volts.
14. In the **Buffer Assignment** window, add a buffer by clicking the **+** button as needed to get three buffers.

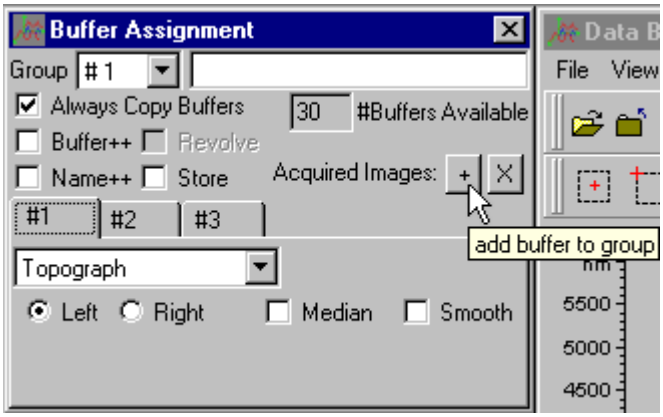

Figure 4.14 (Adding buffers to the group)

Set the three buffers to **Topograph**, **Amplitude**, and **Phase** by selecting each from the drop-down menu on the tab for each buffer. **Topograph** displays the topography of the sample. **Amplitude** displays the amplitude of the cantilever oscillation at each point. The microscope tries to keep the amplitude constant, so this image shows changes in the sample surface. **Phase** displays the phase between the cantilever oscillation and the drive signal.

15. In the **View** menu, select **Layout Editor**.

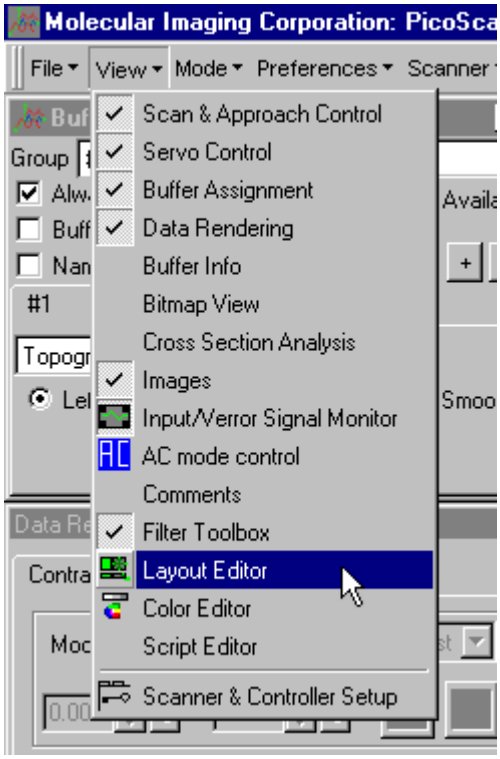

Figure 4.15 (Opening the Layout Editor)

Select the option to display three buffers and click **OK**.

16. Align the photodiode detector such that the LFM and deflection signals on the microscope display are as close to zero as possible. In other words, center the laser beam on the detector. See **Aligning the Photodiode Detector** in Chapter 3 of the **PicoLE System module** for more information.

Engaging the Sample

17. Set the integral (**I**) gain % to 1.000 and the proportional (**P**) gain to 0.5 in the **Servo** tab of the **Servo Control** window.
18. In this same tab, make sure the **Servo Range** is set to its maximum value.

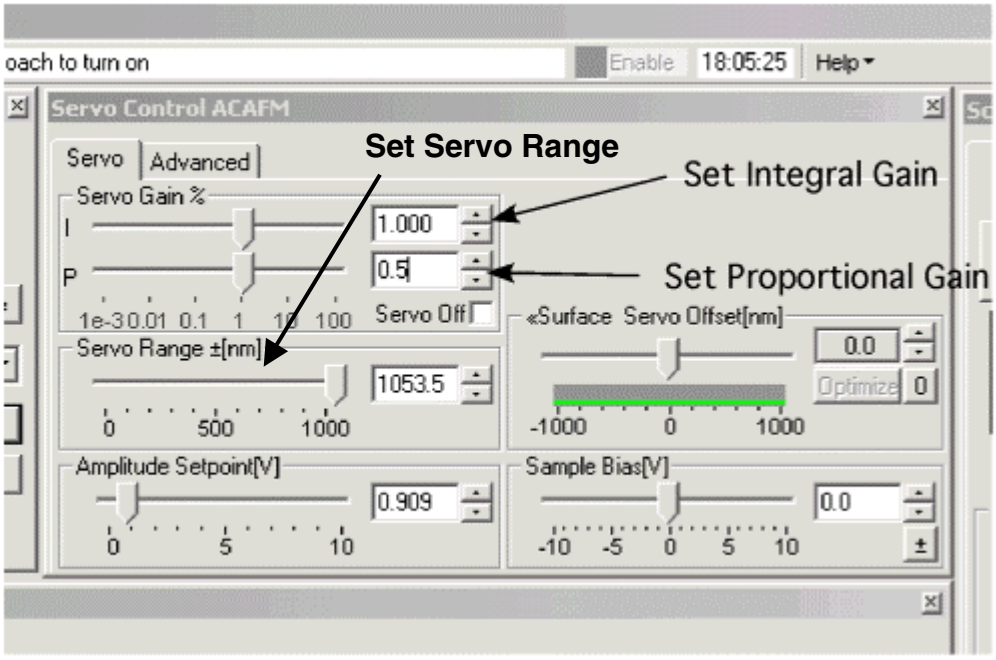

Figure 4.16 (Servo Control window)

19. Set the **Approach Motor Speed** to 5 and the stop at value to 0.9 in the **Approach** tab of the **Scan and Approach Control** window.

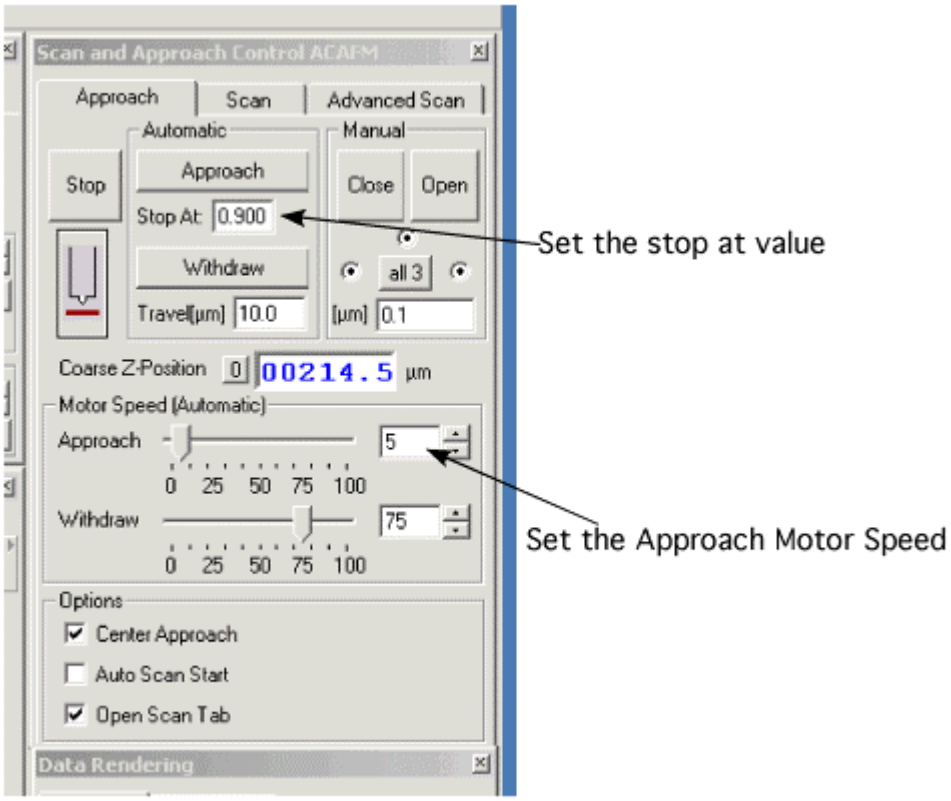

Figure 4.17

20. Press the **Approach** button. When the tip reaches an appropriate imaging distance from the surface of the sample, the following message appears:

Figure 4.18

The microscope is now engaged!

21. Click OK.

Imaging

22. The **Scan and approach Control** window should appear with the **Scan** tab displayed. If not, click the **Scan** tab to bring it to the front.
23. Set the desired X and Y scan dimensions then press the **Start** button.
24. An image should slowly appear in the **Data Buffers** window.

Note: If a blank image appears, the system may have experienced a false engagement, meaning the tip was sufficiently deflected to cause the microscope system to begin acquiring data without actually reaching the sample. This can happen with softer tips and with samples in liquid.

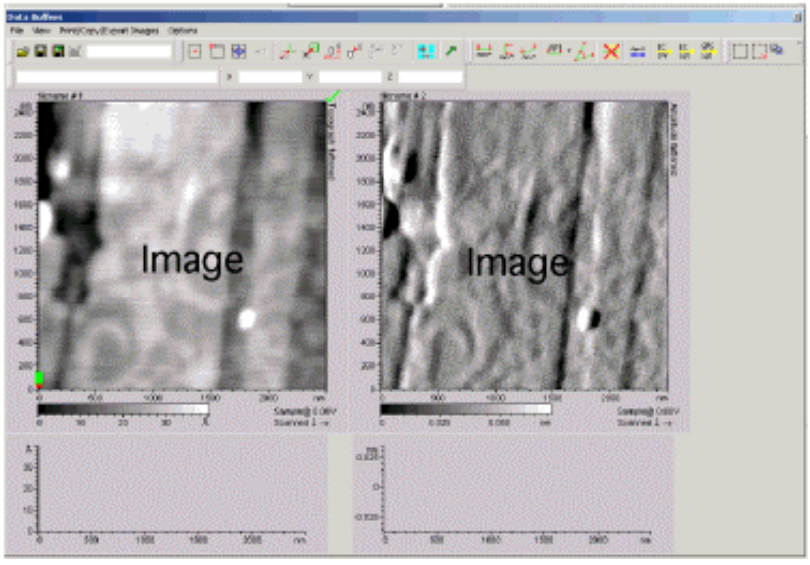

Figure 4.19

Clicking the **Adjust** box (with the green border) in the **Optimizing** tab of the **Data Rendering** window will auto-scale the image displayed in the **Data Buffers** window.

To get continuous scans, uncheck the **Single Scan** box in the **Scan** tab of the **Scan and Approach Control** window. From this tab, the system can be told to always scan up, always scan down or to alternate (Toggle) the scan direction.

Image Processing

The following parameters can be adjusted to maximize image quality:

1. **Mode** (Flattened, Equalized etc., in the Optimizing tab of the Data Rendering window)
2. **Z-Scale**
3. **Integral Gain**
4. **Proportional Gain**
5. **Set point**

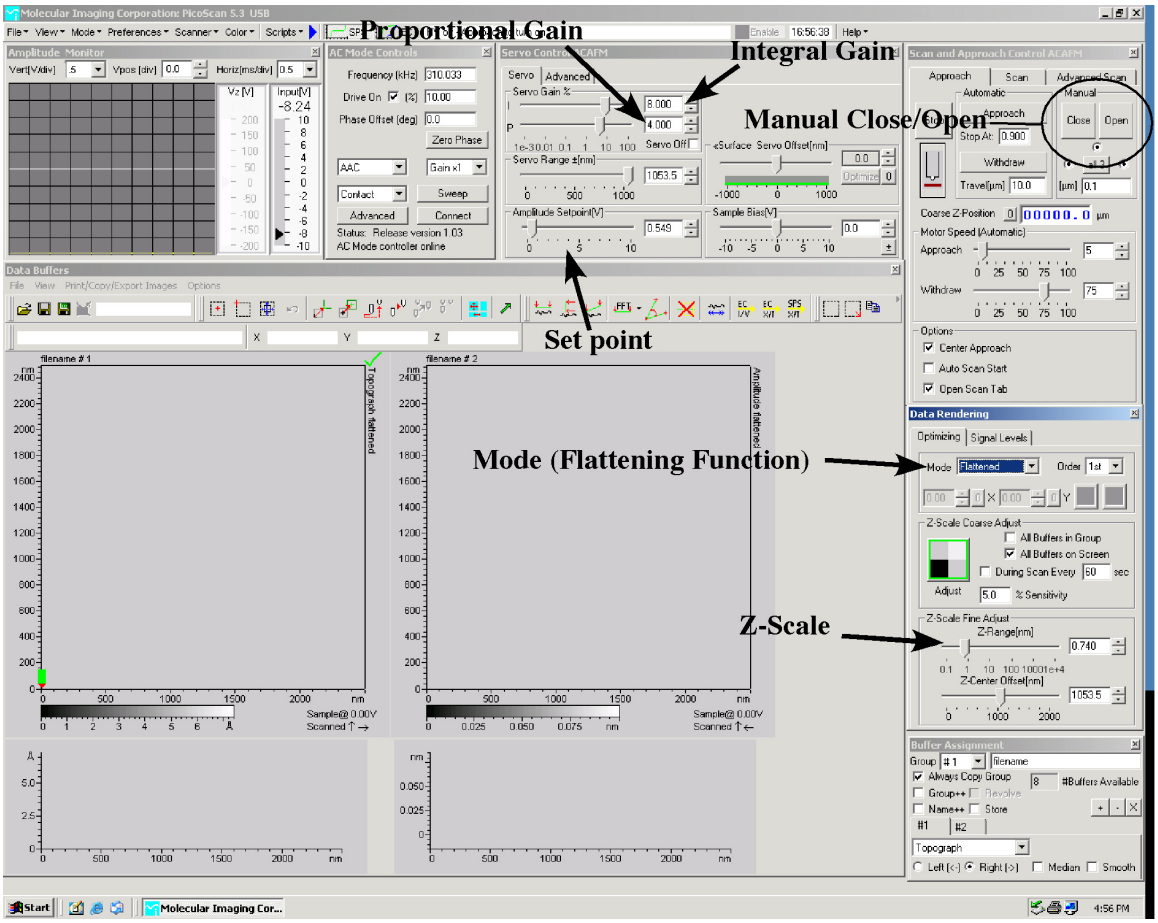

Figure 4.20

For the **Mode** setting, Flattened first order is the best starting choice. Sometimes second order can remove distortions caused by sample bowing.

The **Z-Scale** adjustment can be set automatically by pressing on the adjustment tab. A good value for the % sensitivity is 5. Fine adjustments to the Z range can be made either by entering new values or by moving the slider below the adjustment tab.

The integral and proportional gain can be used after initial Mode and Z-scale adjustments. Increasing these parameters can sharpen an image. Decreasing these parameters can remove noise-like distortion from an image. See the **PicoScan Software Manual** for more details.

The **Set Point** can be adjusted to remove image distortion that results from the tip location relative to the surface. The tip being too high can result in blurry and physically distorted sample. The tip being too low can result in horizontal (scratch like) lines.
Note: This parameter should be carefully adjusted in small increments as improper adjustments can crash the tip!

Other parameters that can be considered for improving image quality:

1. **Scan speed**: Sometimes a sample needs to be scanned at a slow rate. Usually samples can be scanned at 2 lines/second.

2. **Rotate adjustment**: A large feature off the screen can sometimes distort a sample. Rotating the scan direction can sometimes solve this problem. This value is in degrees.

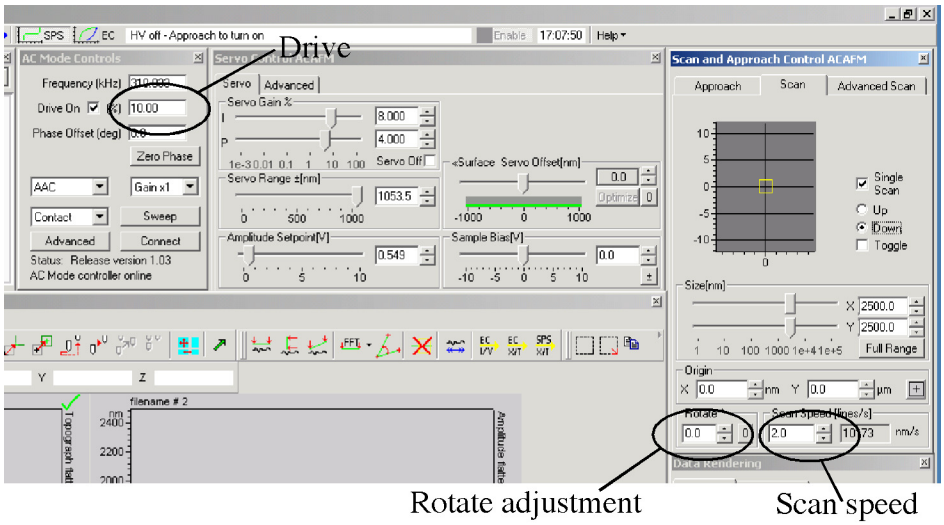

Figure 4.21

Remember to save images with the **Save** function on the files menu in the data buffers box.

File Management

THE PICOSCAN SOFTWARE USES STANDARD WINDOWS FILE MANAGEMENT DIALOGS.

To save an image, simply click the **Save** button in the toolbar of the **Data Buffers** window or of the **Spectroscopy** or **Potentiostat** windows. **Save** can also be selected from the **File** drop-down menu in any of these windows. The file name will be what is entered in the filename portion of the **Buffer Assignment** window. By having the **Name++** box checked, the file names will automatically increment (filename01, filename02, etc.), placing each saved set of data in a separate file. If this box is not checked, the previous data will continue to be overwritten to the same file.

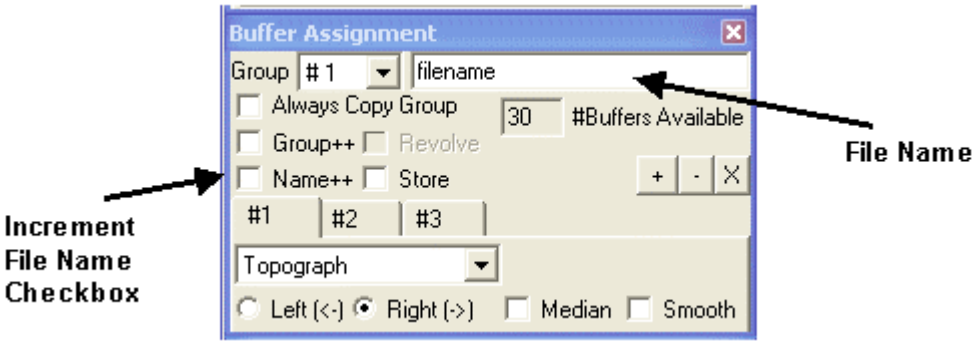

Figure 4.22

If **Store** is selected, PicoScan will save the buffer data after each scan is done.

Save Data As… is also a choice in any of the above listed locations. This selection allows you to designate a file name for the data.

To retrieve an already saved image, simply open the file by clicking the **Open** icon or by selecting **Open** from the **File** drop-down menu.
Note: Loading saved data automatically places PicoScan into **View** mode. You will have to make sure the system is in **Live Scan** mode before acquiring any new data.

Chapter 5: AACAFM Tutorial

A strong point of the PicoSPM system is its AC mode capability. With this mode, one can obtain high-quality images of soft samples in-situ. AC mode means that the cantilever oscillates at a controlled frequency, and PicoScan measures the resulting amplitude and phase changes as the tip of the cantilever interacts with the surface being imaged. A vibrating piezo crystal in the nose assembly induces the cantilever oscillations. This tutorial will use the PicoLE system in AAC Mode. A Large scanner and an NCH (non-contact high-frequency) cantilever (C ≈ 40 N/m) will be used to image a gold-on-mica substrate. This tutorial should only be done after completion of the AFM tutorial in the **PicoLE System module**. If the images obtained are of unsatisfactory quality, try using the PicoIC™ isolation chamber. This tutorial is written for a system with a PicoLE microscope and an AC Mode Controller. For a different system configuration, refer to the original AC Mode manual for details.

Hardware

- AC Mode controller
- AFM scanner
- Acoustic AC Nose Assembly
- AAC Cantilever
- Tweezers
- Sample
- Two-sided tape
- Photodiode detector

System Setup

Hardware Setup

Set up the microscope as detailed in the **PicoLE System module**. Make the hardware connections that are described in **Chapter 1: Initial Setup** (page 3) of this module. The **Contact/AC Mode/CS AFM Switch** on the **PicoLE Head Electronics Box** should be turned to AC Mode. Make sure that an AAC nose is placed in the scanner, see Figure 5.1 below. See the **AFM Nose Assembly** section in Chapter 3 of the **PicoLE System module** for instruction on placing the nose into the scanner.

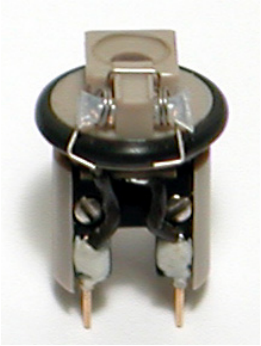

Figure 5.1 (AAC nose)

Software Setup

PicoScan software should have already been installed on the computer. Use the following steps as a guide to become familiar with the system settings required to make good images.

1. Click **View** on the main menu bar and select **Setup (Scanner, Controller etc.)**.

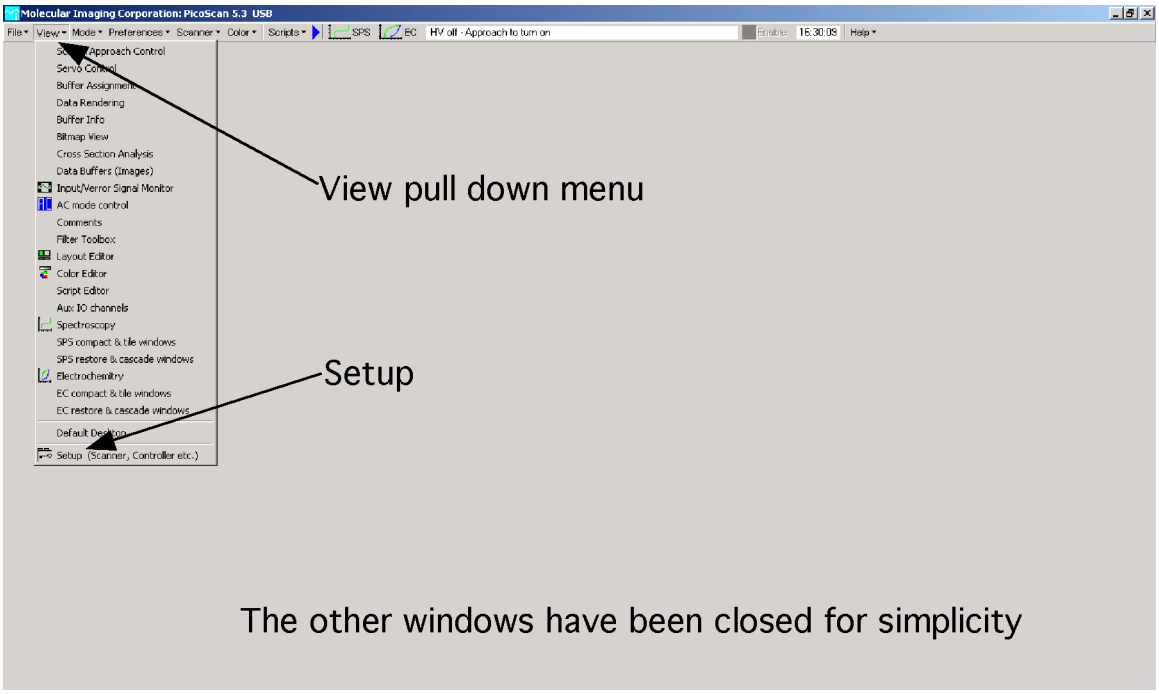

Figure 5.2

The setup dialog will open to the **Scanner & Preamp Calibration** tab

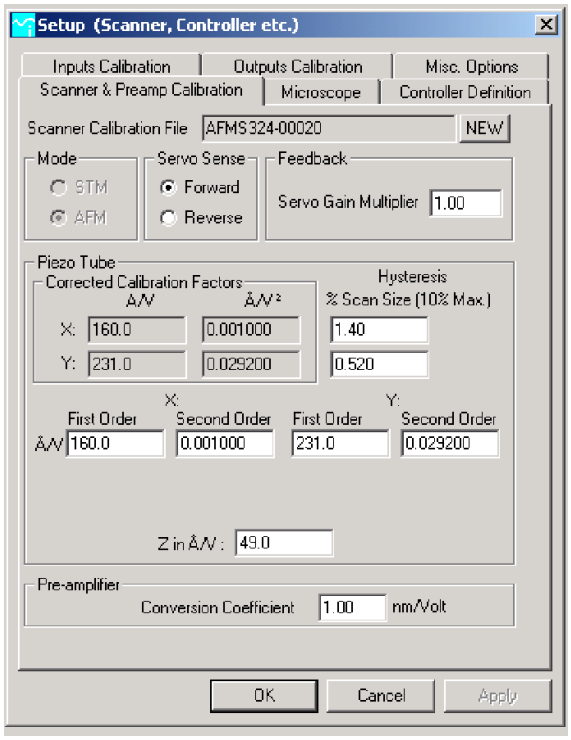

Figure 5.3

2. Click on the **Microscope** tab.

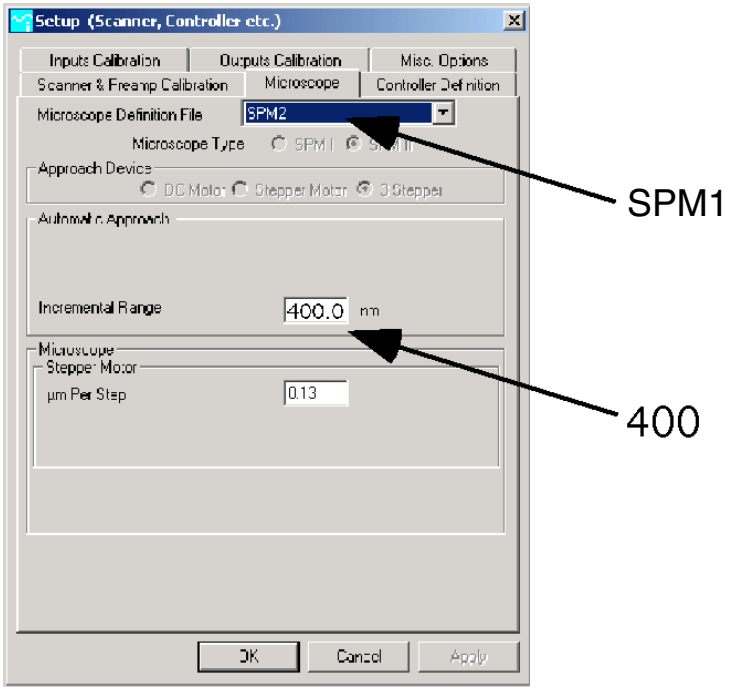

Figure 5.4 (Make sure SPM1 is selected in order for the system to communicate properly with the PicoLE microscope)

3. Select SPM1 as the **Microscope Definition File**. Enter a default value of '400' into the Incremental Range.

4. Select the **Misc. Options** tab. Then select the **AC Mode Controller Supported** box. Click **OK** to accept the changes and close this dialogue.

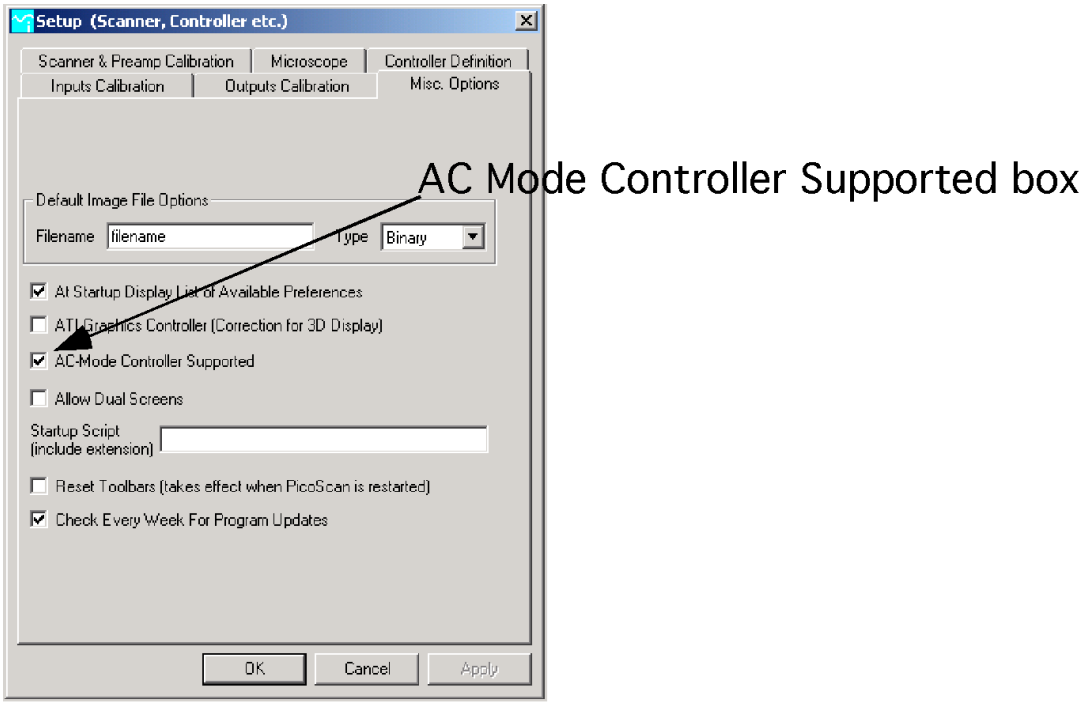

Figure 5.5

5. Select **Live Scan** from the **File** drop-down menu on the **Main Toolbar**.

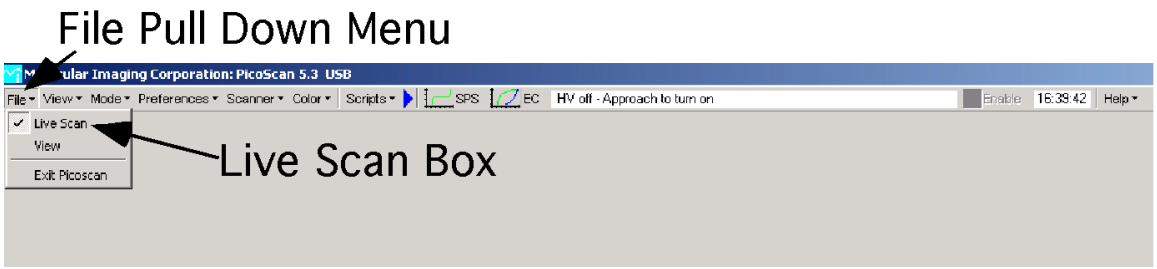

Figure 5.6

6. Under **Mode** on the main menu, select **AC AFM**.

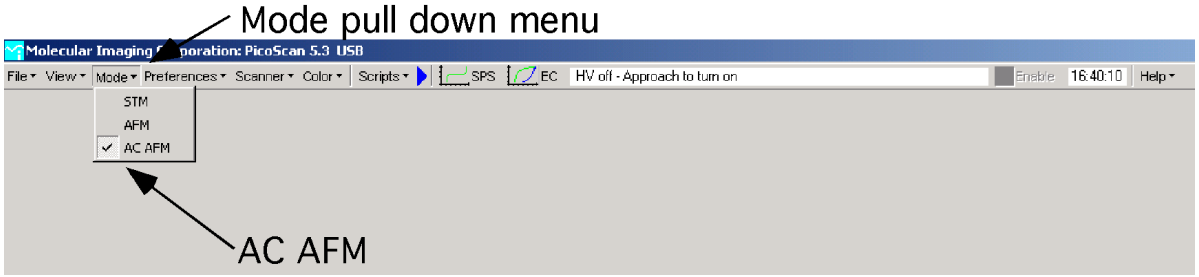

Figure 5.7

7. Under **Scanner** on the main menu select the scanner calibration file that matches the serial number of the scanner being used. See Figure 5.8 below for the location of the serial number.

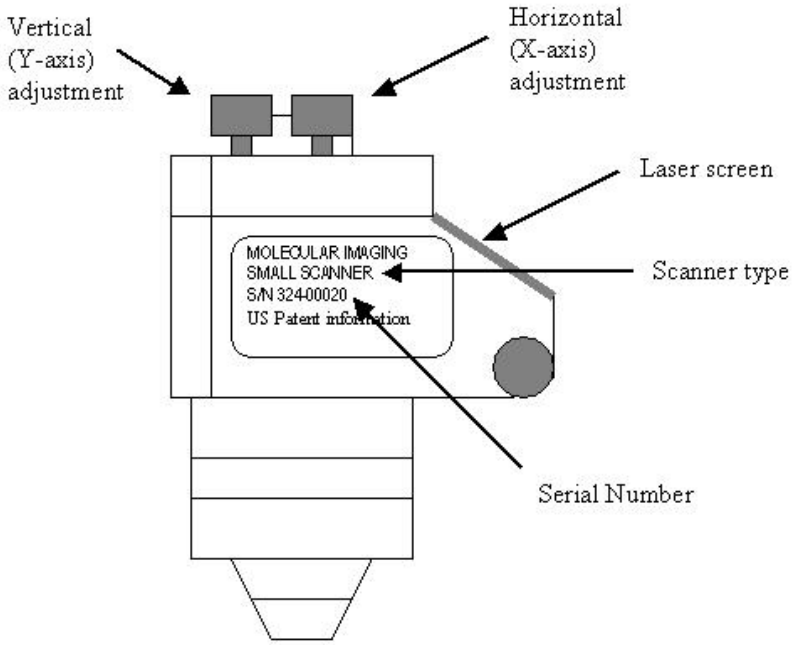

Figure 5.8

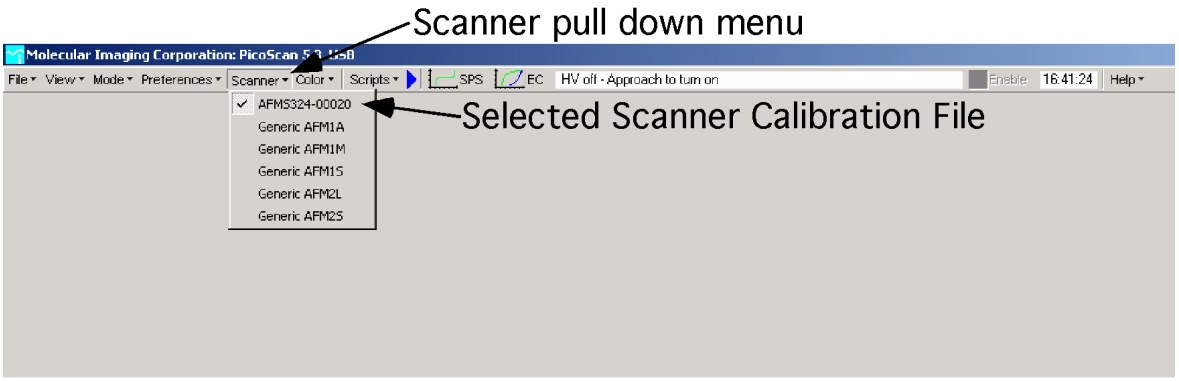

Figure 5.9 (Select the calibration file that matches the serial number of the scanner)

Tuning the Tip

8. Under **View** on the main menu, select **AC Mode Controls**, then perform the following;

- Select **AAC** (as indicated in Figure 4.10 below) to perform a Acoustic AC scan using an AAC tip
- Select **Drive On.**
- Enter 5 for the drive %.
- Set the **Gain** to 1x.

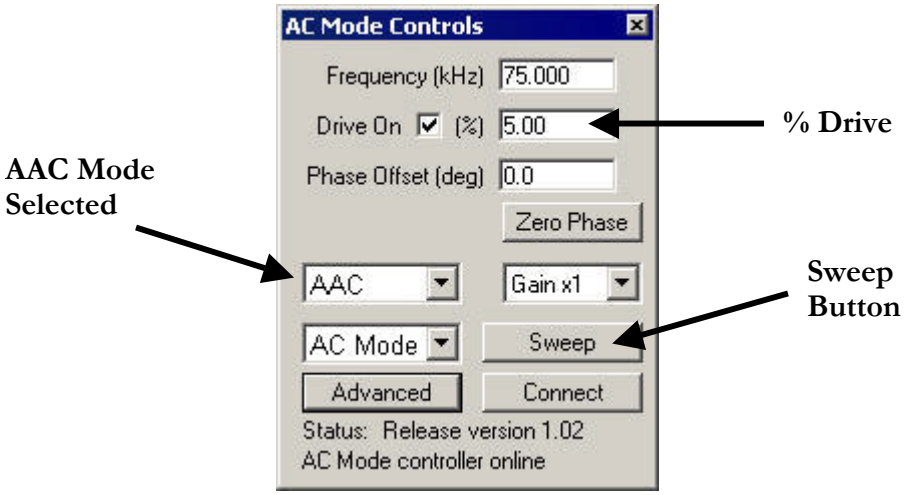

Figure 5.10

9. Click on the **Sweep** button. The AFM AC mode Frequency Plot will now appear.

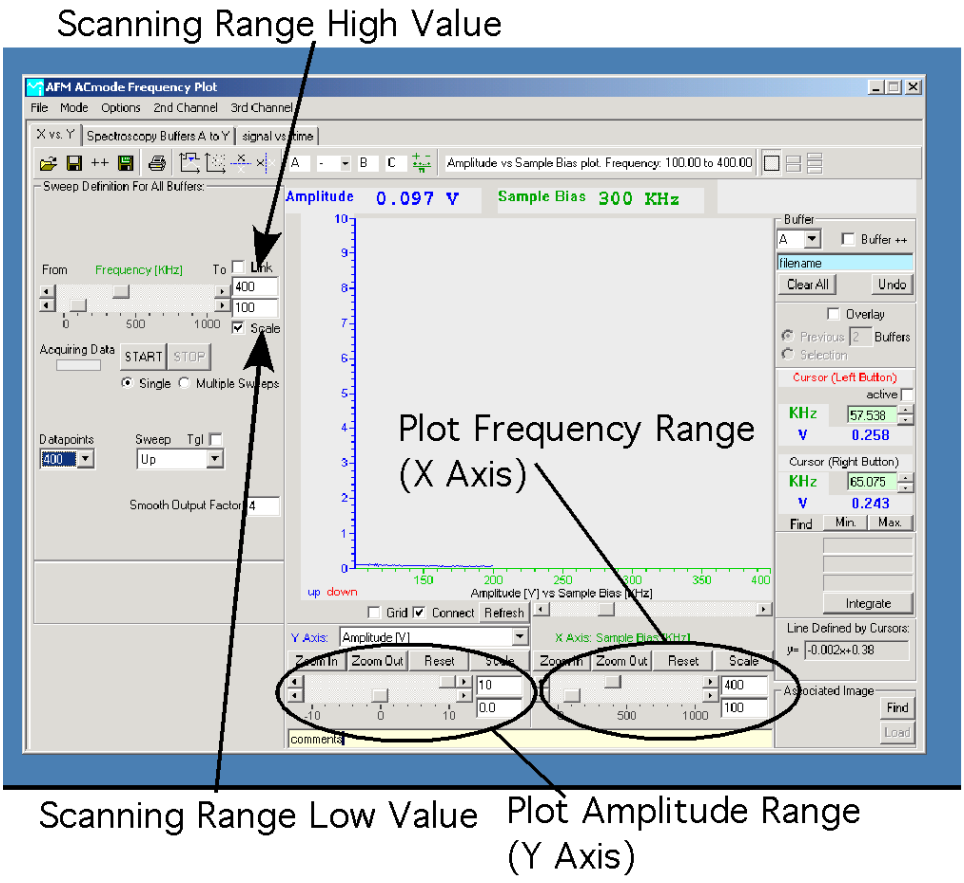

Figure 5.11

10. The resonance frequency of the cantilever must be empirically determined by performing a full-scale frequency sweep.

- Select the scale check box directly under the Frequency Range values.

- Drag the top Frequency slider completely to the right and the bottom Frequency slider completely to the left.
- Click the START button.
- The Resonance peak indicates the resonant frequency of the cantilever (shown in Figure 5.12 below).

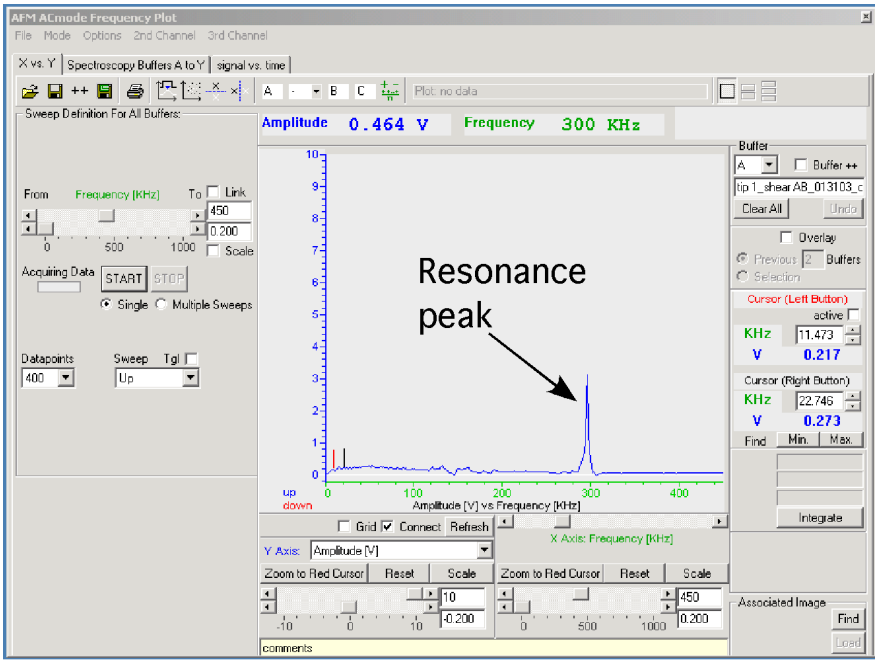

Figure 5.12

- Zoom in on the resonance peak displayed on the frequency plot by adjusting the two Frequency sliders until the resonance peak is centered in the plot.
- Click on the START button in order to initiate another frequency sweep.
- Repeat the above procedure until the frequency range of the sweep is less than 10kHz.
- If off scale, use the software controls to reset the drive to a lower value. If the amplitude is greater than 10 volts, the electronics won't work correctly.

19. Check the **Active** check box in the Frequency plot window. See Figure 5.13 below.
20. Update the drive frequency of the AC Mode control by left clicking slightly to the left of the resonant peak maximum.

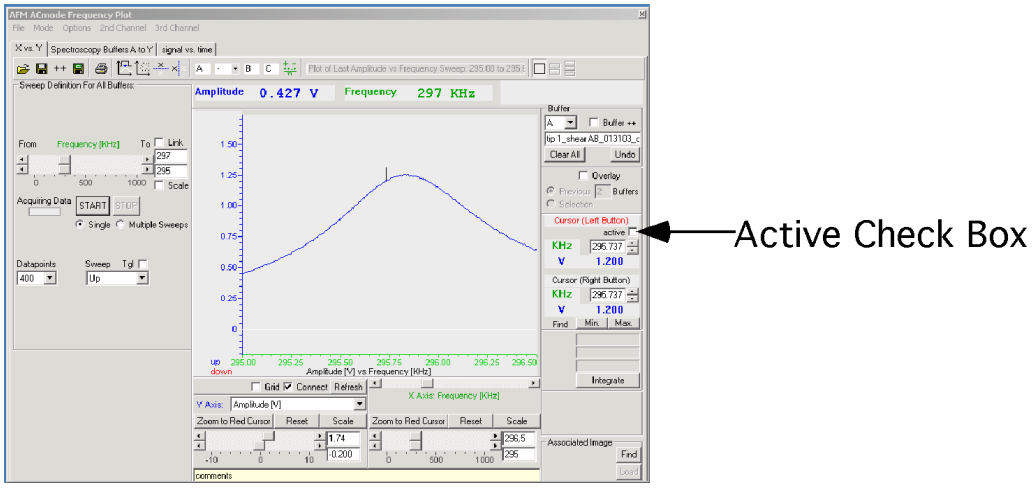

Figure 5.13

21. After the drive frequency has been updated, adjust the drive percentage so that the amplitude, displayed on the front of the AC Mode controller box (the hardware unit, not to be confused with the **AC Mode Controls** window in the software), is 5 ±1 volts.
22. In the **Buffer Assignment** window, add a buffer by clicking the **+** button as needed to get three buffers.

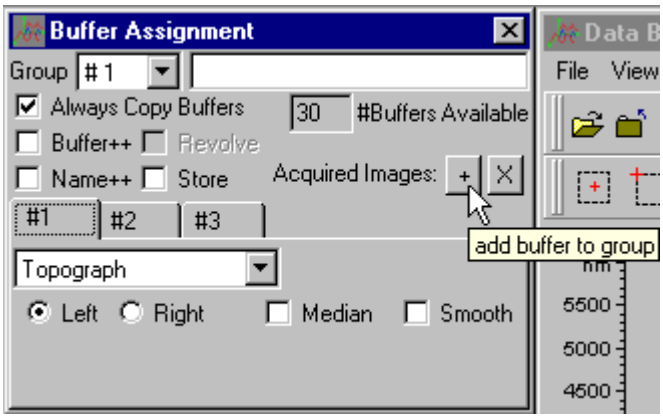

Figure 5.14 (Adding buffers to the group)

Set the three buffers to **Topograph**, **Amplitude**, and **Phase** by selecting each from the drop-down menu on the tab for each buffer. **Topograph** displays the topography of the sample. **Amplitude** displays the amplitude of the cantilever oscillation at each point. The microscope tries to keep the amplitude constant, so this image shows changes in the sample surface. **Phase** displays the phase between the cantilever oscillation and the drive signal.

23. In the **View** menu, select **Layout Editor**.

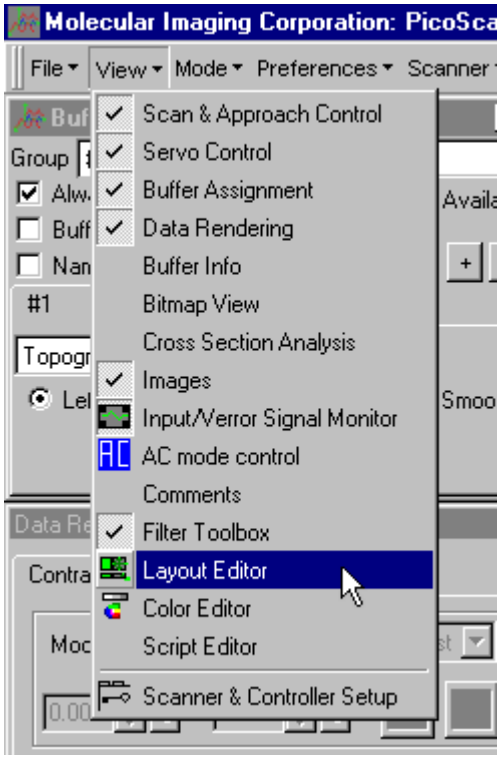

Figure 5.15 (Opening the Layout Editor)

Select the option to display three buffers and click **OK**.

24. Align the photodiode detector such that the LFM and deflection signals on the microscope display are as close to zero as possible. In other words, center the laser beam on the detector. See **Aligning the Photodiode Detector** in Chapter 3 of the **PicoLE System module** for more information.

Engaging the Sample

25. Set the integral (**I**) gain % to 1.000 and the proportional (**P**) gain to 0.5 in the **Servo** tab of the **Servo Control** window.
26. In this same tab, make sure the **Servo Range** is set to its maximum value.

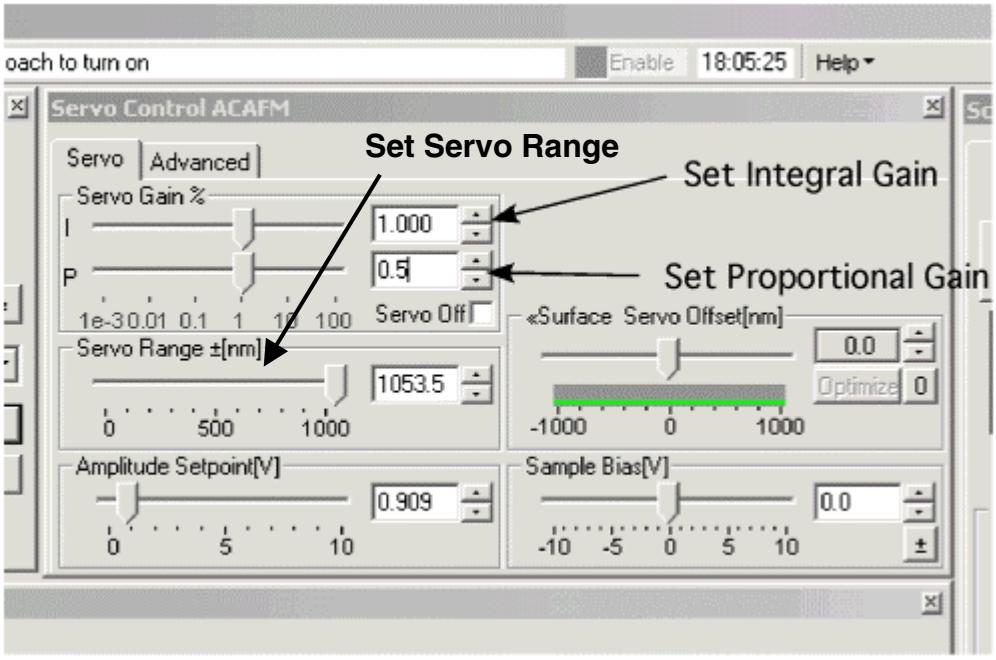

Figure 5.16 (Servo Control window)

19. Set the **Approach Motor Speed** to 5 and the stop at value to 0.9 in the **Approach** tab of the **Scan and Approach Control** window.

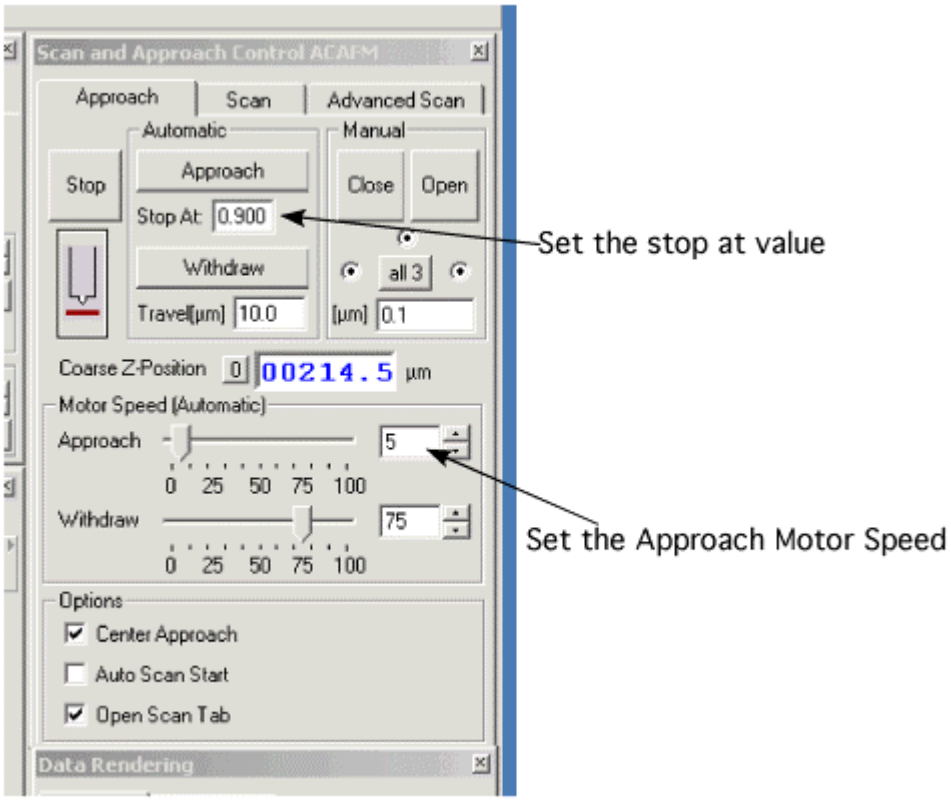

Figure 5.17

25. Press the **Approach** button. When the tip reaches an appropriate imaging distance from the surface of the sample, the following message appears:

Figure 5.18

The microscope is now engaged!

26. Click OK.

Imaging

27. The **Scan and approach Control** window should appear with the **Scan** tab displayed. If not, click the **Scan** tab to bring it to the front.
28. Set the desired X and Y scan dimensions then press the **Start** button.
29. An image should slowly appear in the **Data Buffers** window.

Note: If a blank image appears, the system may have experienced a false engagement, meaning the tip was sufficiently deflected to cause the microscope system to begin acquiring data without actually reaching the sample. This can happen with softer tips and with samples in liquid.

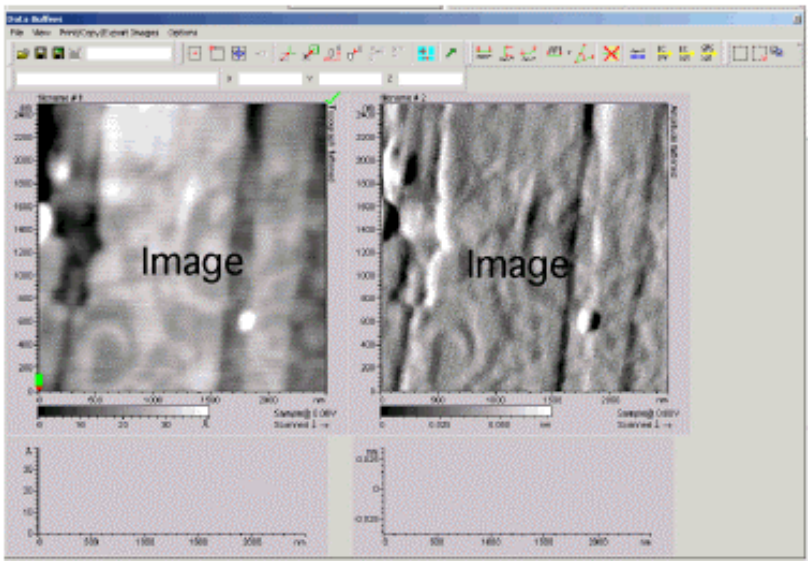

Figure 5.19

Clicking the **Adjust** box (with the green border) in the **Optimizing** tab of the **Data Rendering** window will auto-scale the image displayed in the **Data Buffers** window.

To get continuous scans, uncheck the **Single Scan** box in the **Scan** tab of the **Scan and Approach Control** window. From this tab, the system can be told to always scan up, always scan down or to alternate (Toggle) the scan direction.

Image Processing

The following parameters can be adjusted to maximize image quality:

6. **Mode** (Flattened, Equalized etc., in the Optimizing tab of the Data Rendering window)
7. **Z-Scale**
8. **Integral Gain**
9. **Proportional Gain**
10. **Set point**

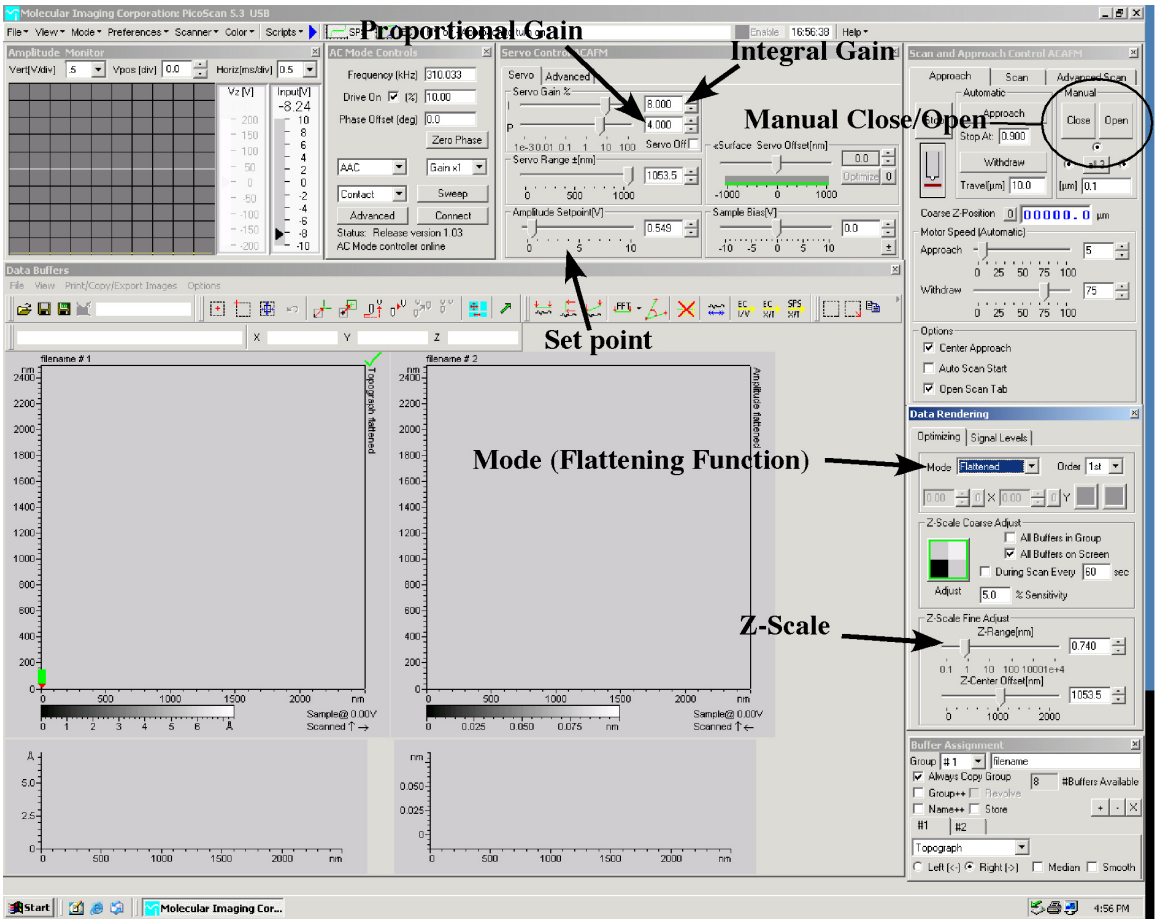

Figure 5.20

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The **Set Point** can be adjusted to remove image distortion that results from the tip location relative to the surface. The tip being too high can result in blurry and physically distorted sample. The tip being too low can result in horizontal (scratch like) lines.
Note: This parameter should be carefully adjusted in small increments as improper adjustments can crash the tip!

Other parameters that can be considered for improving image quality:

1. **Scan speed**: Sometimes a sample needs to be scanned at a slow rate. Usually samples can be scanned at 2 lines/second.

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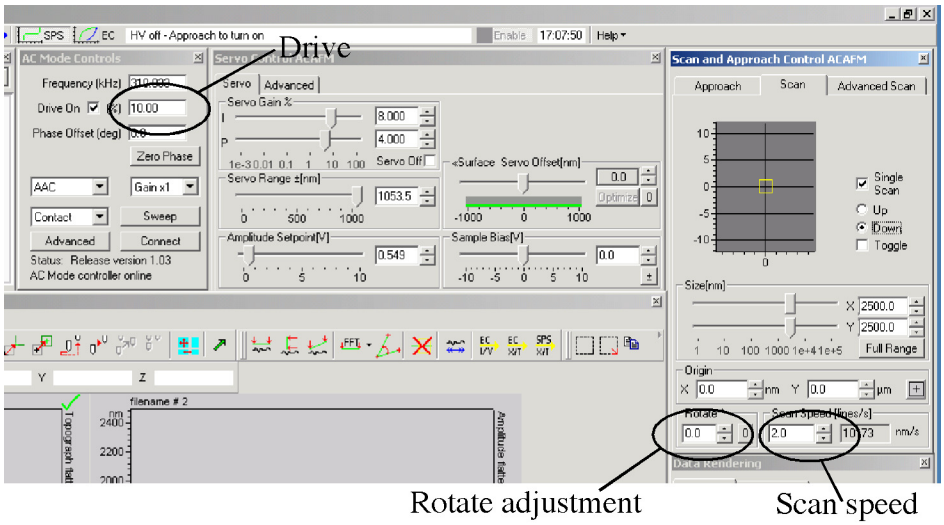

Figure 5.21

Remember to save images with the **Save** function on the files menu in the data buffers box.

File Management

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To save an image, simply click the **Save** button in the toolbar of the **Data Buffers** window or of the **Spectroscopy** or **Potentiostat** windows. **Save** can also be selected from the **File** drop-down menu in any of these windows. The file name will be what is entered in the filename portion of the **Buffer Assignment** window. By having the **Name++** box checked, the file names will automatically increment (filename01, filename02, etc.), placing each saved set of data in a separate file. If this box is not checked, the previous data will continue to be overwritten to the same file.

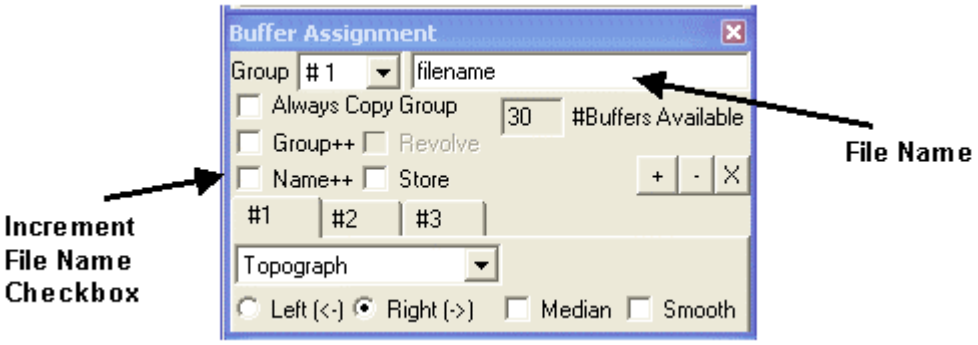


Figure 5.22

If **Store** is selected, PicoScan will save the buffer data after each scan is done.

Save Data As… is also a choice in any of the above listed locations. This selection allows you to designate a file name for the data.

To retrieve an already saved image, simply open the file by clicking the **Open** icon or by selecting **Open** from the **File** drop-down menu.
Note: Loading saved data automatically places PicoScan into **View** mode. You will have to make sure the system is in **Live Scan** mode before acquiring any new data.

Chapter 6: ACAFM Procedure

Selecting and Mounting a Cantilever

Choice of an appropriate AFM cantilever is critical to the successful operation of AC Mode AFM. Typically cantilevers with spring constant on the order of 1 N/m or higher are used. Most vendors will specify force constant and resonance frequency of their cantilevers; make note of them. The actual values of a cantilever's force constant and resonant frequency typically vary by 10-15% of the published values.

Mounting the Cantilever

In ACAFM, mounting of the cantilever is the same as that of contact AFM.

Aligning and Tuning the Cantilever

As previously noted, AC Mode provides the best signal to noise response when the cantilever is oscillated at or near its resonance frequency. It is, therefore, important to become skilled in properly aligning the cantilever and identifying the cantilever resonance frequency. Refer to the tutorial on page **9** if necessary for more information. If using an older MAC Mode Controller, refer to its original manual. The only difference is the software control.

System Setup

1. Attach all cables as described earlier in this manual.
2. The AC Mode electronics must be switched on prior to operation. The power switch can be found on the rear panel alongside the power cable.
3. Ensure that **AC Mode** and **Drive** are turned off in PicoScan. To turn off AC Mode, make sure the **Contact / AC Mode** pulldown menu has **Contact** selected.
4. In PicoScan, adjust the Amplitude Setpoint to zero. The control can be found in the **Servo Control** window of PicoScan.
5. Using the appropriate AC Mode equipment, set up the microscope as if for a standard AFM experiment and align the laser on the apex of the cantilever. This procedure is described in the AFM chapter of the **PicoLE System module**.
6. Align the detector in the microscope such that the deflection and LFM signals on the microscope electronics box are close to zero. This procedure is described in the AFM chapter of the **PicoLE System module**.

Tuning the Cantilever

"Tuning" is a commonly used term referring to the systematic location of the cantilever's natural resonance frequency and the selection of an appropriate driving signal to establish an appropriate oscillation amplitude.

7. The **AC Mode Controls** window is shown in the tutorial in the preceding chapter. Set **Input Gain** to x1. Set **Drive** to 5%. Click on the **Sweep** button. If imaging in MAC Mode, ensure that the MAC Mode sample stage is positioned such that the ferrite core is directly under the cantilever and as close as possible without interfering with the free oscillation of the cantilever.
8. Select an appropriate frequency sweep range for the installed cantilever. Select **Single Sweep** and set the **# Data Points** (samples) setting. The frequency range divided by **# Data Points** is the resolution.

9. Press the **START** button in the **AFM ACMode Frequency Plot** window. The software will perform a simple sampling procedure measuring the oscillation amplitude of the cantilever while stepping the frequency in the specified range. The sweep will be displayed in the window.
10. The resonance response curve, also called the tuning curve, should show a single, sharp peak that stands several times taller than the baseline noise. The height of the peak can be changed by adjusting the **Drive** in the **AC Mode Controls** window, and the frequency axis can be changed using the From/To Frequency controls in the spectroscopy window. Using these controls and, if necessary, increasing **Nr Data Points**, locate the frequency at which the curve reaches its maximum value. This is the natural resonance frequency of the cantilever. The ideal imaging frequency is at, or just below, the free resonant frequency of the cantilever. For the best image quality, select either the peak value by clicking the **Find: Max** button just below the **active** checkbox or manually select a drive frequency from the steep slope on the low (left) side of the peak in the frequency sweep. To select a drive frequency, check the **active** checkbox for the red cursor in the **AFM AC Mode Frequency Plot** window and then click the desired location on the plot. The resonant frequency may decrease slightly when the sample is engaged.
11. The cantilever's amplitude is positive and always shown on the AC Mode Controller's meter. Switch the pulldown menu from **Contact** to **AC Mode** to turn on AC Mode. Adjust **Drive** in the **AC Mode Controls** window until the display on the MAC box reads between 2.0 and 5.0. The **Amplitude Setpoint** is the amplitude the servo will maintain when engaged.
12. In the **Scan and Approach Control** window, make sure that **Stop At:** is set to 0.9 V. The system is now ready to approach and image the sample.

Obtaining an Image with AC Mode

- For softer samples, the setpoint will need to be adjusted as necessary to minimize the tip-sample interactions. If the difference between the setpoint and free amplitudes is too great, there is a potential for degrading the sample quality (and damaging the tip).
- Approach the probe to the sample surface in the same way as in contact mode AFM and start to image after the probe is in range.
- It is important to note that the controller approaches the tip to the sample until the amplitude of the cantilever in volts has been decreased to the Amplitude Setpoint voltage.
- Using the amplitude-distance curve to adjust the set point is a good way to optimize imaging conditions. The amplitude is represented by a positive voltage, thus free amplitude is at the top of the plot and zero amplitude is at the bottom. Under normal conditions you should adjust the force set point to about 80% of free amplitude. Again with softer samples this value may need to be adjusted to a higher percentage of the free amplitude.
- Typically in AC Mode AFM, it is desirable to use lower gain settings (I, P = 0.3, 0.3) and a slower scan rate (2-3 Hz). Scan rates vary with the stiffness of the cantilever. The stiffer the cantilever, the more quickly the Z-piezo can respond, which allows good image resolution at higher scan rates.

Additional Tips for Magnetic AC Imaging

If unable to get a good image in MAC Mode, these tips may help:

- Place the microscope in a PicoIC with the door closed. Make sure that there are no significant sources of noise in the area.
- Separate all relevant cables so that their magnetic fields will not interact to produce noise. Also keep the cables away from any other power cords or electric devices.
- Handling the microscope can cause thermal instabilities in the system. Try leaving the microscope alone for at least an hour after setting it up to allow the system to reach thermal equilibrium before imaging.

- If unable to find a good resonance peak with the frequency sweep, or if the cantilever drifts away from the laser beam, try gluing or otherwise securely fastening the cantilever to the cantilever holder.

Acoustic AC imaging is discussed in-depth in **Chapter 8: Advanced Theory**.

Chapter 7: Troubleshooting

Getting a clear, stable image depends on many parameters. Having a clean, properly prepared sample and a good cantilever are essential to attaining quality images. There are other factors that play a role in the acquisition of quality images. The following tips offer helps in resolving some of the problems encountered in MAC mode imaging.

Unable to get clear images after placing a good cantilever

The optimized operation force set point is with in the range ~0.7 to 0.9 of the free amplitude before the tip touches the surface of the sample. When the set point is above the shoulder, as shown in Figure 7.1, the image will be blurry. To resolve this problem, gradually lower the set point to bring it below the shoulder. Or, as a last resource, an Amplitude vs. Distance curve can be run in order to adjust the set point.

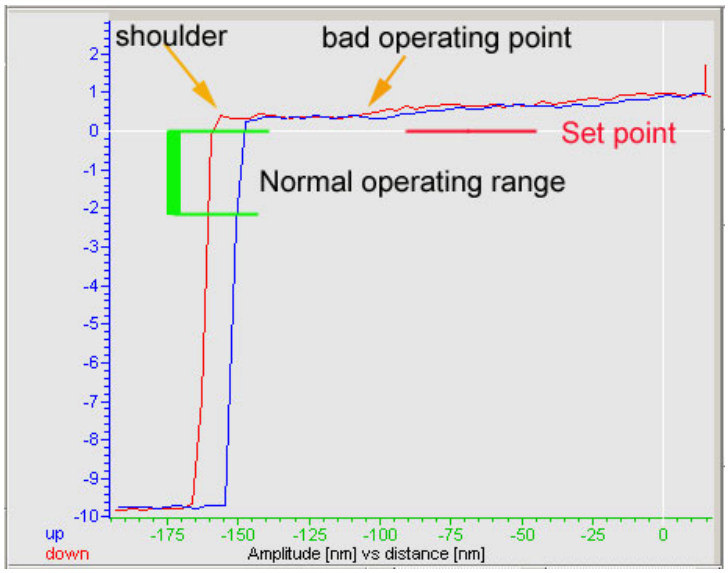


Figure 7.1 (Amplitude vs. Distance curve)

Noisy Amplitude vs. Frequency curve

Occasionally, the Amplitude vs. Frequency curve becomes noisy as shown in the left side of Figure 7.2 below. A readjustment of the deflected laser spot will often solve this problem, as shown in the right side of Figure 7.2.

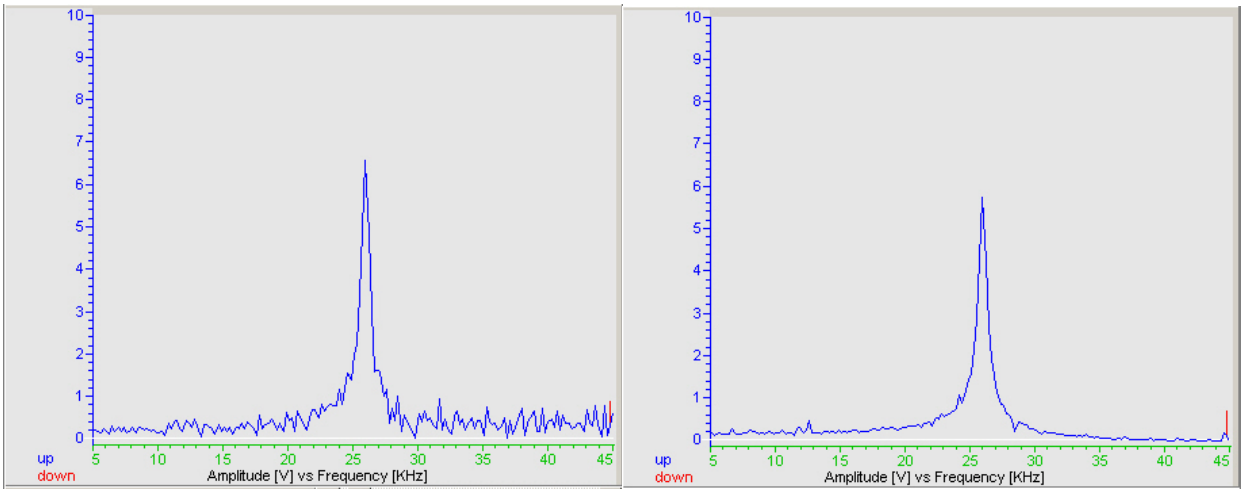


Figure 7.2 (left: Noisy and right: Clean Amplitude vs. Frequency curves)

Chapter 8: Advanced Theory

The following document was written by Chad Rigetti concerning Acoustic Mode ACAFM. Most of the concepts also apply to MAC Mode. The document contains a wealth of information on the finer points of AC Mode imaging. It is presented as-is on the following pages. Note that this document was written for the PicoSPM I microscope.

Optimizing Acoustic Mode Image Quality with Molecular Imaging Corp.'s PicoSPM

Introduction

The PicoSPM is an all-purpose scanning probe microscope. Its capabilities include acoustic mode atomic force microscopy (AFM), MacMode AFM, and scanning tunneling microscopy (STM). While this paper is intended to be a guide to acoustic mode imaging in air, much of the information will be applicable in MacMode as well. No prior knowledge of AFM has been assumed, so the paper can act as a self-contained guide for new users.

Physics of Acoustic Mode Atomic Force Microscopy

When playing a guitar, we strum the strings to create standing mechanical waves, which in turn produce acoustic waves. Acoustic excitation of a cantilever works in the opposite way – acoustic waves are used to induce mechanical vibrations. We can flex the cantilever up and down by setting the drive frequency to one of the cantilever's *bending modes* (which we call its resonant frequency). In the most general case, the amplitude and phase of the induced oscillations as a function of drive frequency are described by (1) and (2),

$$A(\omega) = \frac{F_0 / m}{\left[\left(\omega_o^2 - \omega^2\right)^2 + \frac{\omega_o^{\,2}\omega^2}{Q^2}\right]^{1/2}} \qquad (1)$$

$$\tan\varphi = \frac{2\omega_o\omega}{Q\left(\omega_o^2 - \omega^2\right)} \qquad (2)$$

where ω_o is the cantilever's resonant frequency, ω is the drive frequency, F_o is the drive amplitude and m is the effective mass of the cantilever. Q is the *quality factor*; it is a measure of the peak's sharpness, as well as the energy dissipated per oscillation. These equations describe the free oscillation of a cantilever (though there are other factors that must be included in a rigorous analysis, such as the bending of the cantilever holder and the chip itself[1]).

Acoustic mode imaging relies on an interaction between the oscillating cantilever/tip and the sample surface. The interaction introduces a net force on the cantilever according to the Lennard-Jones force curve. Depending on the mean tip-sample separation, the net force on the cantilever over one cycle of motion can be either attractive or repulsive. The interaction force is illustrated in figure 1. Incorporating the effects of this force on the cantilever's oscillation is non-trivial, primarily due to its non-linearity over the course of a single oscillation. Indeed, for large amplitudes (10 – 100 nm), where the tip spends time in both the attractive and repulsive regimes, the system dynamics are not amenable

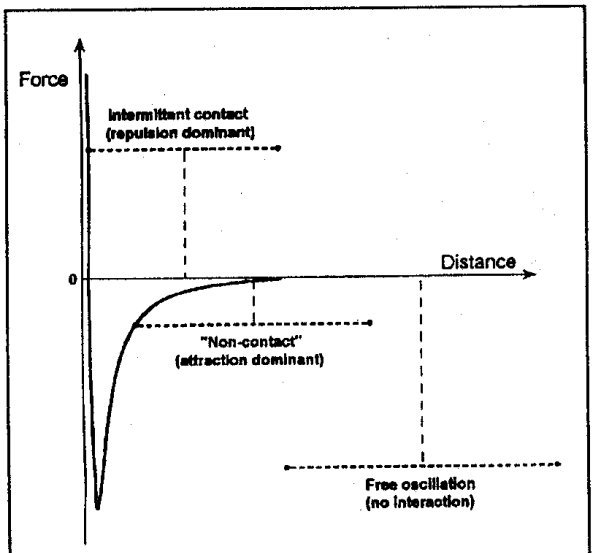


Fig. 1. Schematic illustration of three distance-dependent regimes of interaction between an oscillating cantilever/tip and a sample surface. Dashed horizontal line segments denote distance intervals explored by the tip in each regime, at three values of mean distance (vertical dashed lines). The interaction regimes are labeled free oscillation (no interaction, bottom), non-contact (attraction dominant, middle), and intermittent contact (repulsion dominant, top).

to analytical techniques[2]. However, the system can be solved numerically by first writing the differential equation of motion as:

$$m\ddot{z} + c\dot{z} + kz + F(z) = F_o \sin \omega t \qquad (3)$$

where m is the effective cantilever/tip mass, c is the damping constant, k is the cantilever's spring constant, F_o is the drive amplitude and ω is the drive frequency. The $F(z)$ term is the non-linear force of interaction with the sample surface[3].

The behavior of the system is revealed by the solutions to this equation. If the drive frequency is set at ω_o, the free resonance of the cantilever/tip, we will see the amplitude and phase behavior described in b) of the figure below. Note the small jump in amplitude (on the linear portion of the graph) as the mean tip-sample separation is reduced. This is accompanied by a corresponding phase discontinuity; it marks the transition from net attraction (non-contact) to net repulsion (intermittent contact). As the separation is reduced further, a second discontinuity denotes a return to net attraction.

If the drive frequency is set below ω_o, these jumps in amplitude will not appear. There will, however, be a sharp peak at the top of the linear portion, as seen in part a). In the case that the drive is higher than ω_o, we see a combination of these two behaviors, as shown in c).

This behavior also implies a shift in the resonance peak as a result of the tip-sample interaction. In the attractive regime, the location of the resonance is shifted *downward* relative to the free resonance, while in the repulsive regime it is shifted *upward.* At the interface between the two regimes, the tip begins 'tapping' the surface at the peak of its motion. Ostensibly, this should lead to a *decerease* in amplitude, because the interaction is surely dissipative. A more thorough examination, however, reveals that this tapping causes an abrupt shift in the location and width of the resonant peak, and thus gives rise to the phase and amplitude discontinuities discussed above. The amplitude actually *increases* because the net effect of the shift is to move the resonance *nearer* to the drive frequency.

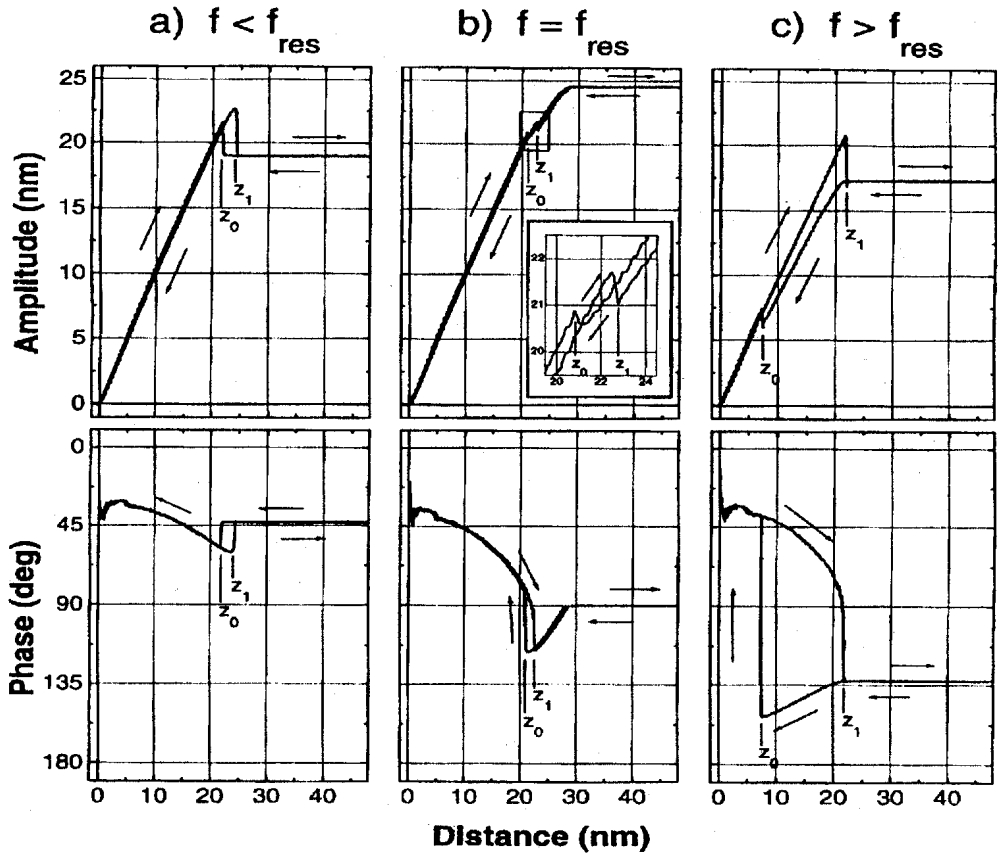
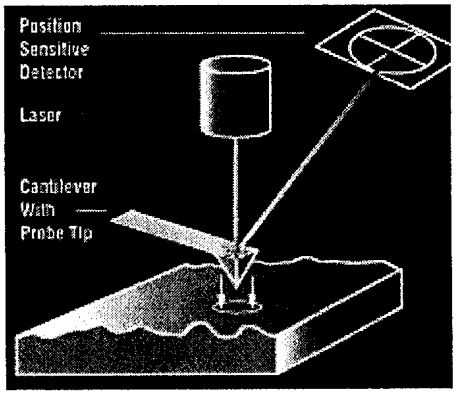


Fig. 1. Experimental DFS curves as obtained by measuring the oscillation amplitude and phase shift as a function of the average tip–sample distance. The dynamic system was driven (a) below ($f = 299.45$ kHz), (b) at ($f = 299.95$ kHz), and (c) above ($f = 300.45$ kHz) its resonance frequency f_{res}. The DFS curves were probed by a silicon tip on a silicon wafer.

Why do we care about all this? Because if we know precisely how an oscillating cantilever is affected by the forces arising from the sample surface, then we can translate the cantilever's dynamics into information about the surface itself. That, essentially, is the general principle of atomic force microscopy.



Exactly how the information is translated is very complex. The first step is to gather the required data about the cantilever's position and amplitude. The amplitude is measured by reflecting a laser off the end of the cantilever to a *segmented photodiode.* A schematic is shown at left. The photodiode is sensitive to the position at which the laser beam is incident. An oscillating cantilever will result in an ac output (though it is ultimately converted to a voltage for the servo) that contains the amplitude information. Now, we know from figure 4 that the amplitude is a function of tip-sample separation. So the instrument uses a servomechanism to maintain a *constant* cantilever amplitude by moderating this z-distance. The z-modulation is

effected by a piezoelectric column. As the tip is scanned over the surface (also effected by the piezoelectric column), the voltage applied to the z-piezo as a function of position provides the raw topographical data. The cantilever's phase relative to the drive is also contained in the signal from the photodiode, which allows the simultaneous formation of a phase image. The phase data can be interpreted as a measure of *energy dissipation* in the tip-sample interaction[4].

It is also typical to save the cantilever amplitude data itself. Even though the servo attempts to maintain constant amplitude, it can do so perfectly only in an idealized situation. The amplitude data is essentially the *error signal*, or the error in the topography image.

Keep in mind that all of this signal processing requires some serious circuitry. This is very much a trivialized overview of an extremely complex electronic process.

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[1] S.M. Lindsay, *The Scanning Probe Microscope In Biology, 305-307 (1999).*
[2] B. Anczykowski, D. Kruger, K.L. Babcock, and H. Fuchs, *Ultramicroscopy* **66**, *251-259 (1996).*
[3] Ibid.
[4] G Haugstad and R. R. Jones, *Ultramicroscopy,* **76**, *77-86 (1999).*