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Effect of Surface Morphology on Adsorption-Induced Bending of Microcantilevers

Ramya Desikan
University of Tennessee - Knoxville

To the Graduate Council:

I am submitting herewith a thesis written by Ramya Desikan entitled "Effect of Surface Morphology on Adsorption-Induced Bending of Microcantilevers." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Physics.

Thomas George Thundat, Major Professor

We have read this thesis and recommend its acceptance:

Ida Lee, Marianne Breinig

Accepted for the Council:
Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

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EFFECT OF SURFACE MORPHOLOGY ON ADSORPTION-INDUCED BENDING OF MICROCANTILEVERS

A Thesis Presented for the
Master of Science
Degree
The University of Tennessee, Knoxville

Ramya Desikan

December 2005

Dedication

This thesis is dedicated to my favorite god "**GANESHA**", without whom none of this would have been even possible.

Acknowledgements

Many people have been a part of my graduate education, as friends, teachers, and colleagues. I would like to especially thank Dr. Thomas George Thundat without whom this thesis would not exist. I also gratefully acknowledge the enthusiastic supervision Dr. Ida lee during my thesis work. Dr. Marianne Breinig suggestions and comments for the technical documentation are highly appreciated. The research community in Oak ridge National laboratory has been extremely supportive, thanks to you all. Finally, I am forever indebted to my parents R. V. Desikan and Usha Desikan for their understanding, endless patience and encouragement when it was most required. I wish to thank my entire family and friends for providing me a loving environment.

Abstract

Microcantilevers undergo bending due to molecular adsorption when adsorption is confined to a single surface. The origin of the adsorption-induced force is assumed to be surface stress variation due to molecular adsorption. Single crystal silicon cantilevers were etched for a series of different time periods using two different types of Potassium Hydroxide solutions in order to obtain a rough and a smooth finish on the cantilever surface. Cantilevers that approximately had the same resonance frequency in the rough and smooth etched categories were chosen for comparison in the experiment. Liquid phase adsorption of 1-Do-decan-thiol on the cantilevers having a thin gold receptor was investigated with optical beam deflection method. The surface roughness of the cantilevers was quantified using atomic force microscopy imaging of the cantilever. Our results indicate that an increase in surface area does not increase the bending of a microcantilever, a smoother surface provides a better platform for the formation of a Self Assembled Monolayer. The un-etched cantilevers were used as the control and had the least deflection. . Self assembly of alkanethiols closely follows Langmuir type kinetics up to a single monolayer assembly. My results demonstrate that surface stress and adsorption kinetics of alkanethiols on the gold layer is considerably affected by its structural conformation.

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Chapter 1

Introduction

Microcantilevers can be used not only for imaging in scanning force microscopy, but also as an important tool to explore the physics and chemistry of the nanometer world. A thin flexible beam made of silicon coated with a sensor layer serves as a sensor. Nanomechanical cantilevers are very small and extremely sensitive to force and mass adsorbed. By coating its surface with a material that selectively adsorbs to a given target molecules (as shown in figure I-a), a cantilever can be converted into a highly sensitive and selective chemical or biochemical sensor. When the cantilever comes in contact with the target substance, it reacts with a mechanical response: the cantilever bends, and its resonance frequency varies. Both signals are measured with extremely high accuracy allowing identification and quantitative detection of the target substance.

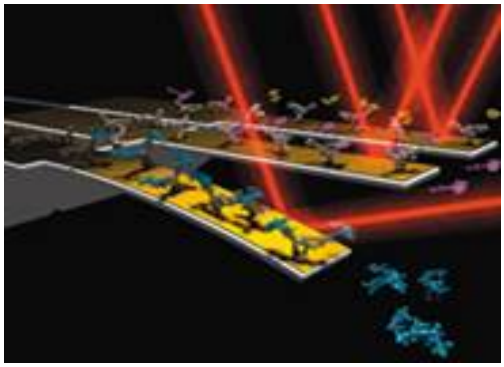

Figure I-a. Shows a Cartoon of a Microcantilever Array Immobilized with Antibodies

Microcantilevers designed for atomic force microscopy have been successfully used as extremely sensitive chemical, physical and biological sensors. Cantilevers transform a chemical reaction into a mechanical motion on the nanometer scale, which can be measured directly, e.g. by the deflection of a light beam from the cantilever surface. Therefore, no fluorescent or radioactive labels are necessary for the detection of biomolecules like DNA or proteins. This significantly reduces the number of steps needed for the detection of biomolecules and eliminates the influence of labels on the molecules.

The motivation behind my thesis work is based on the results published with reference to the effect of surface stress and deflections on microcantilever sensors with different surface morphology. Results by N. Lavrik et.al[19] claim that in rougher surfaces deflections are several orders of magnitude larger than conventional smooth surfaces.

In this study the microcantilevers, were used to investigate the effect of surface morphology on surface stress and adsorption kinetics, of alkanethiol self-assembled monolayers The origin of the adsorption-induced force is not understood, but assumed to be due to surface stress variation as a consequence of molecular adsorption. I discuss in detail the experiments on the liquid phase adsorption of alkanethiol on the cantilevers with different surface topology having a thin gold receptor was investigated with optical beam deflection method.

The cantilevers used for this study were the single crystal silicon Nanosensors manufactured by Nanosensors, Germany (See Figure I-b) and Mikromacsh manufactured by SPM tips, Estonia. The Nanosensor cantilevers are made of single crystal silicon of length of the cantilever 450 ± 5 μm; width of 50 ± 5 μm and the thickness of the cantilever is 2 μm. The Mikromacsh Cantilevers are polycrystalline and 300 μm long, 35 μm wide and the thickness of these cantilevers is 1.3 μm.

In order to increase the surface area, the single crystal silicon cantilevers were etched in pure 30% Potassium Hydroxide solution which results in the formation of micro-pyramidal structures on the silicon surface there by giving a rough finish. In order to obtain a smooth finish (i.e. without micro-pyramidal hillocks) cantilevers surface, the cantilevers were etched in a solution mixture of KOH and isopropyl alcohol which constituted 1% of the total volume. It is essential to clean the surface of the Microcantilever for better results before etching. After extensive cleaning they were etched (smooth and rough etched) accordingly. The cantilevers were then evaporated with 3.5 Nm Ti and 35 Nm Au, in an evaporator. The gold-coated cantilevers with a thin Au receptor layer on one side, serves as sensors for liquid phase adsorption of 1-Do decan thiol. Tapping mode AFM was used to quantify the surface of the cantilevers.

My results comply with the results produced by Michel Godin, L.Y. Beaulieu1 et.al. who have showed that show that both the kinetics of SAM formation and the

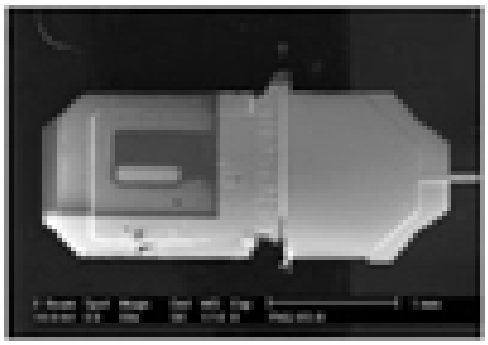

Figure I-b. Shows a Picture of a Nanosensor

resulting SAM structure are strongly influenced both by the surface structure of the underlying receptor layer. The Experimental procedure and results will be discussed in detail.

Chapter 2

Overview of Microcantilevers

The past decade has seen the beginnings of an exciting new field: that of micro-electromechanical sensors (MEMS). These devices promise to revolutionize many aspects of science and scientific applications. For example, researchers are developing microsensors for detecting drugs and explosives that will render canine detection obsolete. Microcantilevers are in many ways at the forefront of this emerging area of technology.

In its most basic form, a microcantilever is a micron-scale miniature diving board used for sensing and measurement application. (See Figure II-a for an image of a microcantilever.) Variances in microcantilever characteristic responses can be measured, and used as indicators of changes in environmental conditions. The common responses of interest are changes in deflection (that is bending), the resonance frequency, the Q-factor (damping rate), and the oscillation amplitude of a cantilever. There are some simple examples of how each of theses responses works. Suppose a microcantilever is coated with gold on its upper surface and then exposed to thiol vapor.

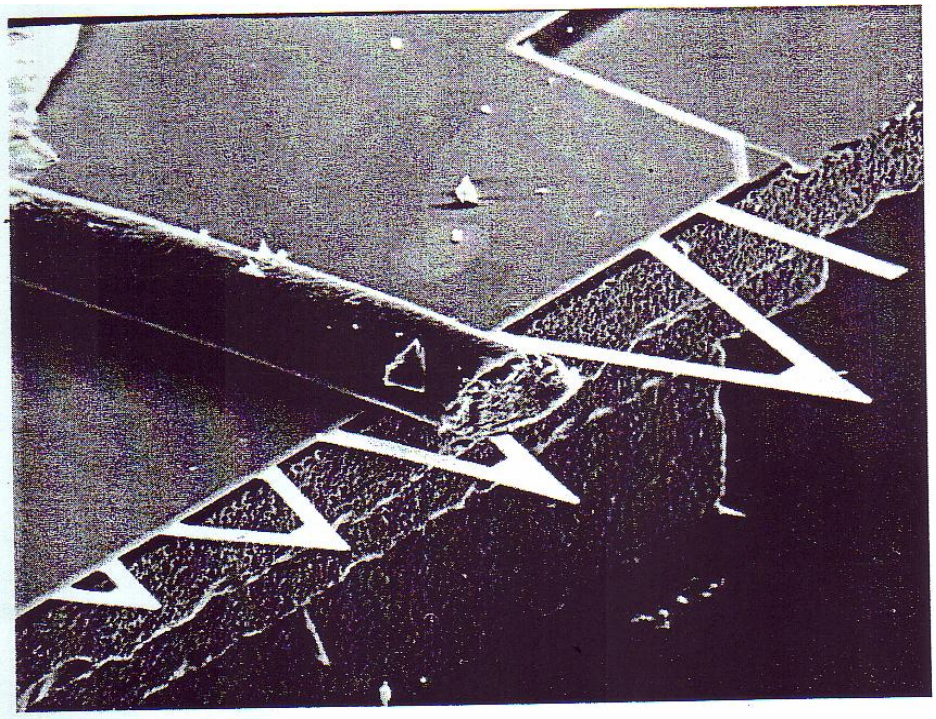

Figure II- a. Microcantilevers and Human Hair

A group of microcantilevers attached to a substrate wafer shown with a human hair for size comparison. The image was taken with and SEM. Photo Courtesy of Dr. Bruce Warmack, ORNL.

Gold has an excellent affinity for adsorbing thiol. As thiol molecules bind to the gold and form a strong bond, the surface stress on the upper surface of the cantilever changes and the cantilever bends. This bending can be used as an indicator of the thiol concentration in air. The resonance frequency of a cantilever can change for a number of different reasons, one of the most common being mass loading caused by adsorption of a target molecule onto the cantilever (such as in the gold thiol example). Generally this occurs in conjunction with changes in the Young's modulus of a material in the cantilever as the molecules diffuse into it (this also acts to change the natural resonance frequency). Another reason for a change in cantilever's resonance frequency might be the presence of an electric field gradient. This has the effect of reducing the effective spring constant of the cantilever, causing a change in resonance frequency. Lastly, a cantilever's Q-factor will change if the viscosity of the surrounding medium changes.

The sensitivity of microcantilevers is extraordinary. Originally microcantilevers were used almost exclusively in Atomic Force Microscopes (AFMs) for surface imaging. As a microcantilever rose and fell with a surface morphology, deflections of an Angstrom could be easily measured using a laser or other position detection system, enabling imaging of individual atoms. In 1993, researchers reported observed changes in cantilever behavior, specifically frequency, and bending due to variations in humidity. They realized that microcantilevers could be used for sensing and measurement in a whole host of applications. Since then, microcantilever measurement and sensor systems have been demonstrated in many

physical, chemical and biological areas. Sensitivities of parts per trillion for chemicals in air, picograms for cantilever mass change due to molecular adsorption, and femtojoules in calorimetry have all been reported.

The number of researchers working in the microcantilever field has been steadily increasing and several companies have been formed specifically to develop microcantilever-based products. The ultimate potential of the micro cantilever sensor is as yet unknown, but the future looks promising.

Chapter 3

Atomic Force Microscope

The Atomic Force Microscope (AFM) is being used to solve processing and materials problems in a wide range of technologies affecting the electronics, telecommunications, biological, chemical, automotive, aerospace, and energy industries. The materials being investigating include thin and thick film coatings, ceramics, composites, glasses, synthetic and biological membranes, metals, polymers, and semiconductors. The AFM is being applied to studies of phenomena such as abrasion, adhesion, cleaning, corrosion, etching, friction, lubrication, plating, and polishing. By using the AFM one can not only image the surface in atomic resolution but also measure the force at nano-newton scale. The publications related to the AFM are growing speedily since its birth.

The first AFM was made by meticulously gluing a tiny shard of diamond onto one end of a tiny strip of gold foil. In the fall of 1985 Gerd Binnig and Christoph Gerber used the cantilever to examine insulating surfaces. (See Figure III –a)

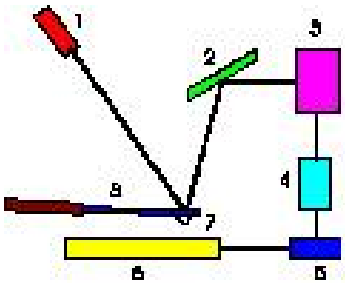

Figure III-a. Layout of the AFM System

Today the tip-cantilever assembly typically is micro fabricated from Si or Si_3N_4. The era of AFM came finally when the Zurich group released the image of a silicon (111) 7X7 pattern. After several years the microcantilevers have been perfected, and the instrument has been embraced by scientists and technologists.

The force between the tip and the sample surface is very small, usually less than 10^{-9} N. How to monitor such small forces is another story. The detection system does not measure force directly. It senses the deflection of the microcantilever. The detecting systems for monitoring the deflection fall into several categories.

The first device introduced by Binning was a tunneling tip placed above the metallized surface of the cantilever. This is a sensitive system where a change in spacing of 1 Å between tip and cantilever changes the tunneling current by an order of magnitude. It is straightforward to measure deflections smaller than 0.01 Å. Subsequent systems were based on the optical techniques. The interferometer is the most sensitive of the optical methods, but it is somewhat more complicated than the beam-bounce method, which was introduced, by Meyer and Amer. The beam-bounce method is now widely used as a result of the excellent work by Alexander and colleagues. In this system an optical beam is reflected from the mirrored surface on the backside of the cantilever onto a position-sensitive photo detector. In this arrangement a small deflection of the cantilever will tilt the reflected beam and change the position of beam on the photo detector. A third optical system introduced by Sarid uses the cantilever as one of the mirrors in the cavity of a diode

laser. Motion of the cantilever has a strong effect on the laser output, and this is exploited as a motion detector.

According to the interaction of the tip and the sample surface, the AFM can be classified as repulsive or Contact mode and attractive or Non-Contact mode. Tapping mode now shows a prosperous future to image the micro-world.

Operation

The principles on how the AFM works are very simple. An atomically sharp tip is scanned over a surface with feedback mechanisms that enable the piezo-electric scanners to maintain the tip at a constant force (to obtain height information), or height (to obtain force information) above the sample surface. Tips are typically made from Si_3N_4 or Si, and extended down from the end of a cantilever. The Nanoscope AFM head employs an optical detection system in which the tip is attached to the underside of a reflective cantilever. A diode laser is focused onto the back of a reflective cantilever. As the tip scans the surface of the sample, moving up and down with the contour of the surface, the laser beam is deflected off the attached cantilever into a dual element photodiode. The photo detector measures the difference in light intensities between the upper and lower photo detectors, and then converts it to voltage. Feedback from the photodiode difference signal, through software control from the computer, enables the tip to maintain either a constant force or constant height above the sample. In the constant force mode the piezo-electric transducer monitors real time height deviation. In the constant height mode

the deflection force on the sample is recorded. The latter mode of operation requires calibration parameters of the scanning tip to be inserted in the sensitivity of the AFM head during force calibration of the microscope.

Some AFM's can accept full 200 mm wafers. The primary purpose of these instruments is to quantitatively measure surface roughness with a nominal 5 nm lateral and 0.01nm vertical resolution on all types of samples. Depending on the AFM design, scanners are used to translate either the sample under the cantilever or the cantilever over the sample.

By scanning in either way, the local height of the sample is measured. Three dimensional topographical maps of the surface are then constructed by plotting the local sample height versus (See figure III- b) horizontal probe tip position.

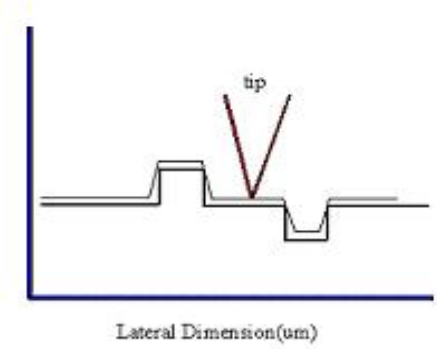

Figure III- b. Horizontal Probe Tip Position

AFM Resolution

The concept of resolution in AFM is different from radiation-based microscopies because AFM imaging is a three-dimensional imaging technique. The ability to distinguish two separate points on an image is the standard by which lateral resolution is usually defined. There is clearly an important distinction between images resolved by wave optics and scanning probe techniques. The former is limited by diffraction, and later primarily by apical probe geometry and sample geometry.

Indeed, many authors have seen that it is the radius of curvature that significantly influences the resolving ability of the AFM. Images of DNA made by the sharper tip have shown dramatic improvements in resolution widths. Even greater improvements in resolution have been attained with tapping mode but contact imaging still is capable of high-resolution imaging.

The Common AFM Modes

Many modes have appeared for special purpose while the technique of AFM is becoming mature. Here I only specify the three commonly used techniques: Contact Mode, Non Contact mode and Tapping Mode. See Figure III- c respectively.

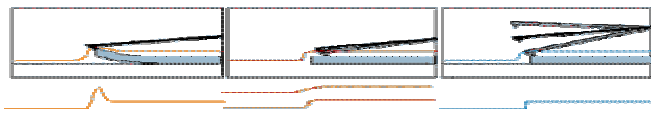

Figure III- c. The Common AFM Modes

Contact Mode

The contact mode where the tip scans the sample in close contact with the surface is the common mode used in the force microscope. The force on the tip is repulsive with a mean value of 10^{-9} N. This force is set by pushing the cantilever against the sample surface with a piezoelectric positioning element. In contact mode AFM the deflection of the cantilever is sensed and compared in a DC feedback amplifier to some desired value of deflection. If the measured deflection is different from the desired value the feedback amplifier applies a voltage to the piezo to raise or lower the sample relative to the cantilever to restore the desired value of deflection. The voltage that the feedback amplifier applies to the piezo is a measure of the height of features on the sample surface. It is displayed as a function of the lateral position of the sample. A few instruments operate in UHV but the majority operates in ambient atmosphere, or in liquids. Problems with contact mode are caused by excessive tracking forces applied by the probe to the sample. The effects can be reduced by minimizing tracking force of the probe on the sample, but there are practical limits to the magnitude of the force that can be controlled by the user during operation in ambient environments. Under ambient conditions, sample surfaces are covered by a layer of adsorbed gases consisting primarily of water vapor and nitrogen, which is 10-30 monolayers thick. When the probe touches this contaminant layer, a meniscus forms and the cantilever is pulled by surface tension toward the sample surface. The magnitude of the force depends on the details of the probe geometry, but is typically on the order of 100 nano Newtons. Operating with the probe may neutralize this

meniscus force and other attractive forces and part or the entire sample totally immersed in liquid. There are many advantages to operate AFM with the sample and cantilever immersed in a fluid. These advantages include the elimination of capillary forces, the reduction of Van der Waals' forces and the ability to study technologically or biologically important processes at liquid solid interfaces. However there are also some disadvantages involved in working in liquids. These range from nuisances such as leaks to more fundamental problems such as sample damage on hydrated and vulnerable biological samples.

In addition, a large class of samples, including semiconductors and insulators, can trap electrostatic charge (partially dissipated and screened in liquid). This charge can contribute to additional substantial attractive forces between the probe and sample. All of these forces combine to define a minimum normal force that can be controllably applied by the probe to the sample. This normal force creates a substantial frictional force as the probe scans over the sample. In practice, it appears that these frictional forces are far more destructive than the normal force and can damage the sample, dull the cantilever probe and distort the resulting data. Also many samples such as semiconductor wafers can not practically be immersed in liquid. An attempt to avoid these problems is the Non-contact Mode.

Non-Contact Mode

A new era in imaging was opened when microscopists introduced a system for implementing the non-contact mode, which is used in situations where tip contact

might alter the sample in subtle ways. In this mode the tip hovers 50 - 150 Angstrom above the sample surface. Attractive Van der Waals forces acting between the tip and the sample are detected, and topographic images are constructed by scanning the tip above the surface. Unfortunately the attractive forces from the sample are substantially weaker than the forces used by contact mode. Therefore the tip must be given a small oscillation so that AC detection methods can be used to detect the small forces between the tip and the sample by measuring the change in amplitude, phase, or frequency of the oscillating cantilever in response to force gradients from the sample. For highest resolution, it is necessary to measure force gradients from Van der Waals forces, which may extend only a nanometer from the sample surface. In general, the fluid contaminant layer is substantially thicker than the range of the Van der Waals force gradient and therefore, attempts to image the true surface with non-contact AFM fail as the oscillating probe becomes trapped in the fluid layer or hovers beyond the effective range of the forces it attempts to measure.

Tapping Mode

Tapping mode is a key advance in AFM. This potent technique allows high resolution topographic imaging of sample surfaces that are easily damaged, loosely hold to their substrate, or difficult to image by other AFM techniques. Tapping mode overcomes problems associated with friction, adhesion, electrostatic forces, and other difficulties that plague conventional AFM scanning methods by alternately placing the tip in contact with the surface to provide high resolution and

then lifting the tip off the surface to avoid dragging the tip across the surface. Tapping mode imaging is implemented in ambient air by oscillating the cantilever assembly at or near the cantilever's resonant frequency using a piezoelectric crystal. The piezo motion causes the cantilever to oscillate with a high amplitude (typically greater than 20nm) when the tip is not in contact with the surface. The oscillating tip is then moved toward the surface until it begins to lightly touch, or tap the surface. During scanning, the vertically oscillating tip alternately contacts the surface and lifts off, generally at a frequency of 50,000 to 500,000 cycles per second. As the oscillating cantilever begins to intermittently contact the surface, the cantilever oscillation is necessarily reduced due to energy loss caused by the tip contacting the surface. The reduction in oscillation amplitude is used to identify and measure surface features.

During tapping mode operation, the cantilever oscillation amplitude is maintained constant by a feedback loop. Selection of the optimal oscillation frequency is software-assisted and the force on the sample is automatically set and maintained at the lowest possible level. When the tip passes over a bump in the surface, the cantilever has less room to oscillate and the amplitude of oscillation decreases. Conversely, when the tip passes over a depression, the cantilever has more room to oscillate and the amplitude increases (approaching the maximum free air amplitude). The oscillation amplitude of the tip is measured by the detector and input to the Nanoscope III controller electronics. The digital feedback loop then

adjusts the tip-sample separation to maintain constant amplitude and force on the sample.

When the tip contacts the surface, the high frequency (50k - 500k Hz) makes the surfaces stiff (viscoelastic) and the tip-sample adhesion force is greatly reduced. Tapping Mode inherently prevents the tip from sticking to the surface and causing damage during scanning. Unlike contact and non-contact modes, when the tip contacts the surface, it has sufficient oscillation amplitude to overcome the tip-sample adhesion forces. Also, the surface material is not pulled sideways by shear forces since the applied force is always vertical. Another advantage of the Tapping Mode technique is its large, linear operating range. This makes the vertical feedback system highly stable, allowing routine reproducible sample measurements.

Tapping mode operation in fluid has the same advantages as in the air or vacuum. However imaging in a fluid medium tends to damp the cantilever's normal resonant frequency. In this case, the entire fluid cell can be oscillated to drive the cantilever into oscillation. This is different from the tapping or non-contact operation in air or vacuum where the cantilever itself is oscillating. When an appropriate frequency is selected (usually in the range of 5,000 to 40,000 cycles per second), the amplitude of the cantilever will decrease when the tip begins to tap the sample, similar to Tapping Mode operation in air. Alternatively, the very soft cantilevers can be used to get the good results in fluid. The spring constant is typically 0.1 N/m compared to the tapping mode in air where the cantilever may be in the range of 1-100 N/m.

Chapter 4

Microcantilever Theory

Microcantilever Composition and Geometry

Microcantilevers usually come in one of two distinct shapes. The first is a bar or a diving board shape, while the second is a hollow centered isosceles triangle shape in which the mounting base forms the third and shortest side. (See figure IV-a for an image of a bar cantilever and figure IV-b for an image of a triangular cantilever) Each shape has distinct advantages and disadvantages. Triangular cantilevers have a larger surface area than the bar version. They can be easily heated by passing a current through the legs (the base of one leg is positive, the base of the other is negative). The primary disadvantage of triangular cantilever is the presence of torsional vibration modes (side to side, rather than up and down), which can complicate cantilever resonance at certain frequencies. The disadvantages of bar cantilever are small surface area and its inability for easy heating.

Microcantilevers used in AFM have a tip, a narrow cone-like structure projecting from the underside of the cantilever near its apex. (See figure IV-c for a picture of a tip.) The tip is typically very sharp, although manufacturing

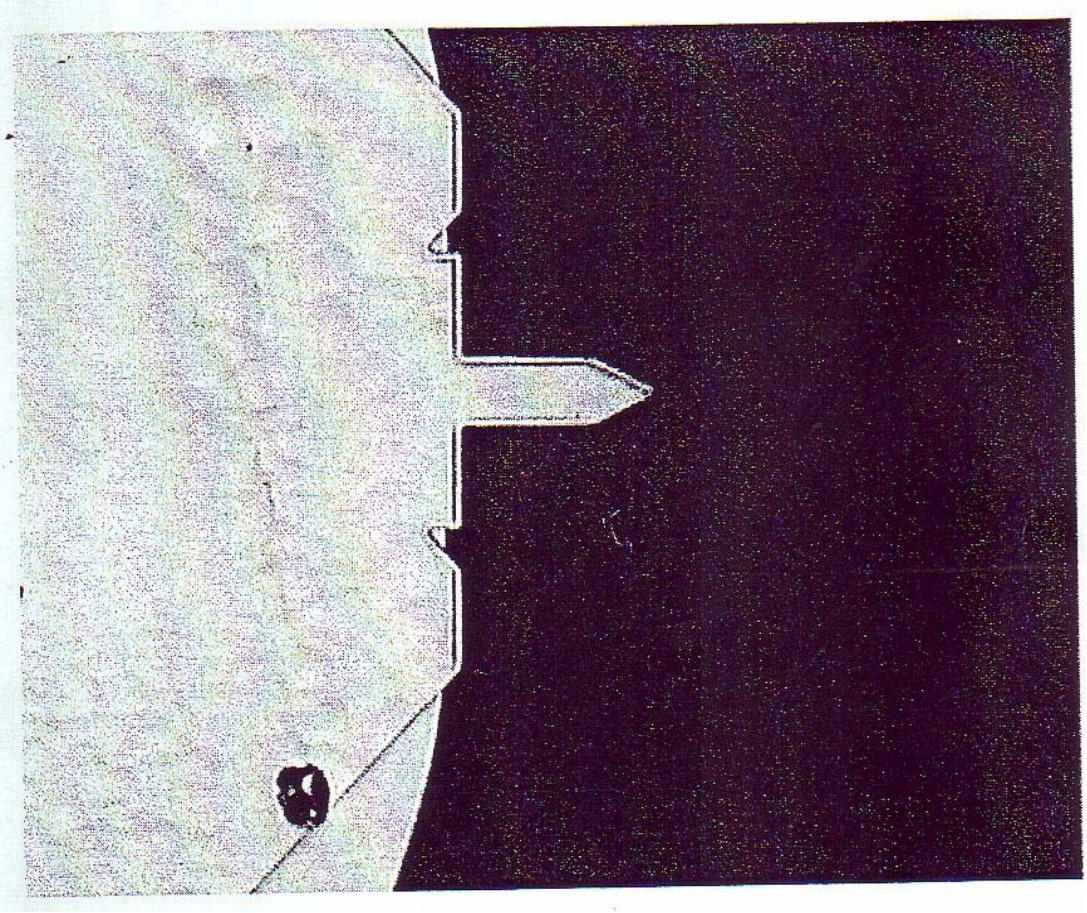

Figure IV-a. Bar Cantilever

Bar cantilever come as purely rectangular shapes, or as in the case, with pointed ends.

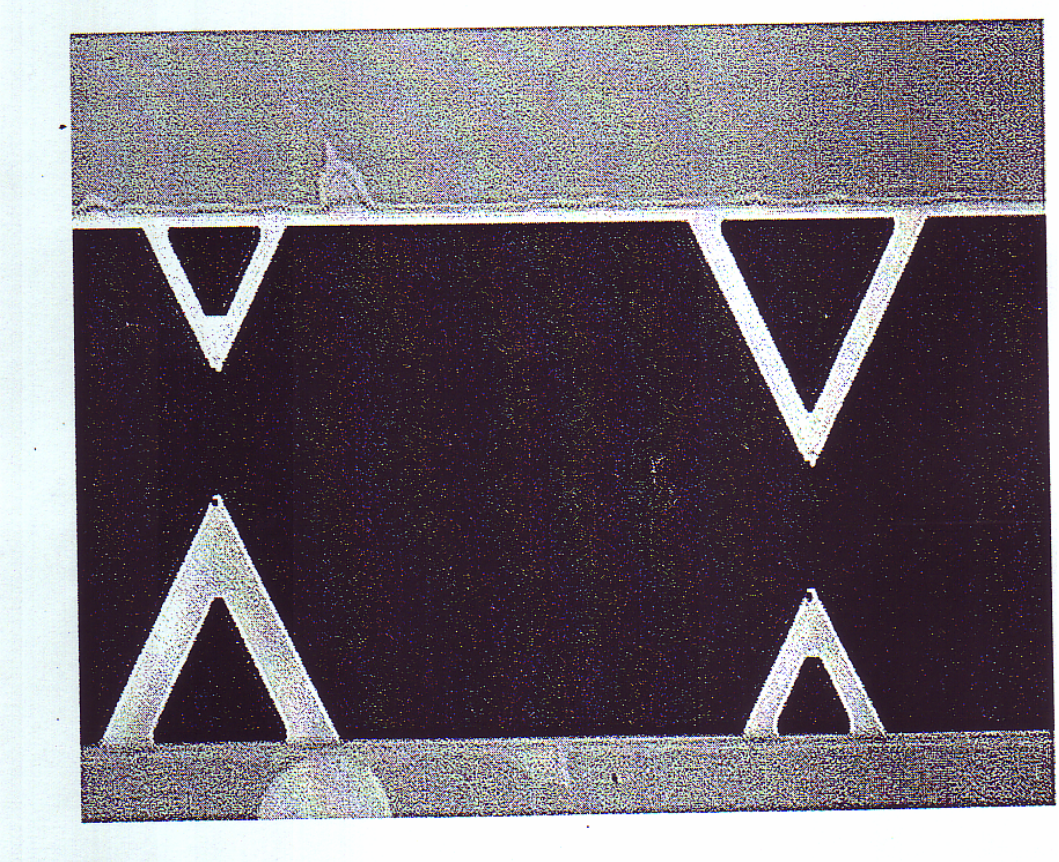

Figure IV –b. Triangular Cantilevers

Cantilevers of different lengths and width are shown. The areas that appear to be holes are actually indentations in the upper surface of the cantilever above the tip.

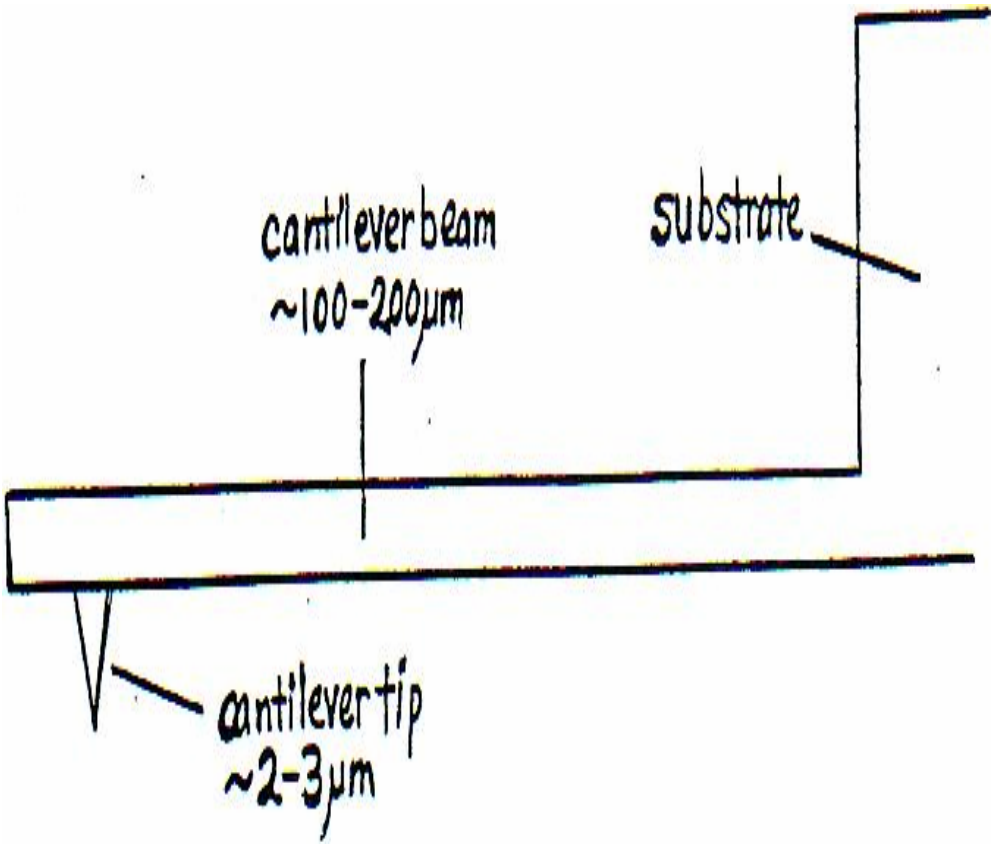

Figure IV-c. Microcantilever Tip (Side View)

The arrangement of a cantilever tip relative to a microcantilever beam.

limitations dictate that the apex of the tip be spherical in shape. The radius of curvature of the apex of the tip is generally on the order of 15-50 nm and the total tip length is two or three microns. The presence of a tip is critical for microscopy application as it allows for a very small area of contact between the microcantilever and the surface being probed, thus maximizing resolution. The main portion of the cantilever is generally referred to as the beam when it is necessary to avoid confusing it with the tip.

Commercially available microcantilevers come in a variety of sizes with typical lengths of 100-800μm, widths (per leg) of 20-50μm, and thickness of 0.3-2μm. With these large variations in dimension, cantilevers vary substantially in harmonic frequency, stiffness, damping rates, and other characteristics.

In theory, microcantilevers can be constructed from a vast range of materials. In practice, they are usually made from semiconductors as existing semiconductor manufacturing technology makes it relatively easy to make such small devices cheaply and precisely compared to construction out of other materials. Specific materials used to date are silicon (Si), silicon nitride ($Si_3 N_4$), and gallium arsenide (GaAs). All cantilevers used in this work were single crystal silicon cantilevers.

Basic Mechanical Characteristics of Microcantilevers

As alluded to earlier, microcantilevers, being rather like miniature diving boards, exhibit similar kinds of mechanical characteristics; they can be bent if a

force is applied to them and, if excited, they tend to vibrate at particular frequencies. (See Figure IV-d for an illustration of the basic mechanical characteristics of microcantilevers.) The parameters for these kinds of motions (spring constant and resonance frequency) can be calculated if basic structural information is known. This will now be discussed and explained. The following equations for f, k, and I and accompanying discussions are based on Dror Sarid's book.[5]

The motion of a cantilever tip approximates a simple spring quiet well. Cantilevers have distinct frequencies of vibrations, spring constant, and damping rates. The resonance frequency, f, of an oscillating cantilever can be expressed as

$$f = \frac{1}{2\pi}\sqrt{\frac{k}{m^*}}$$

where k is the spring constant and m^* is the effective mass of the cantilever. The effective mass of a cantilever is related to the actual mass of the bar, m_b, by a geometric parameter n where, $m^* = n * m_b$ The value of n in a bar cantilever is 0.24, while triangular cantilevers typically have values between 0.143 to 0.18, depending on their exact geometry. Frequency can shift as a result of changes in either mass or spring constant.

Shifts in spring constants are generally the result of changes in either the surface stress or the Young's modulus of the cantilever. This can be problematic as in some cases; changes in mass and spring constant upon exposure to the influence of interest can offset each other by producing no net effect. If this problem

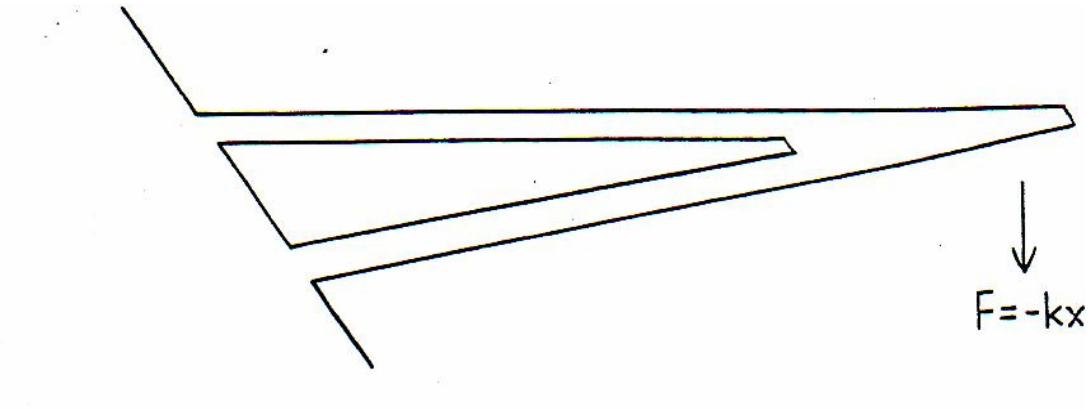

Figure IV-d. Basic Mechanical Characteristics of Microcantilevers

A cantilever has some spring constant k that determines how much it bends when a force is applied to it. It also has some natural rate of oscillation,ω, when excited.

occurs, it can usually be mitigated in one of several ways. One such method is to confine the adsorption area to the terminal end of the cantilever (end loading), thus minimizing differential stress and ensuring that changes in resonance frequency can be wholly attributed to changes in mass loading.

Determining k for a bar cantilever of uniform composition is quiet simple

$$k = \frac{3EI}{l^3}$$

where E is the Young's modulus of elasticity for the cantilever material, I is the moment of inertia, and *l* is the length of the lever.

Since inertia for a rectangular lever is

$$I = \frac{wt^3}{12},$$

where w is the width and t is the thickness of the cantilever, then

$$k = \frac{Ewt^3}{4l^3}$$

In most general cases approximations of *k* are used. One common approximation for a triangular cantilever of uniform composition consists of two cantilevers lying side by side, where each cantilever represents one leg of the triangular cantilever. The lengths are taken as the maximum length of a leg in the original cantilever. The

two cantilevers side by side are then treated as a single bar cantilever. This method is generally accurate with in 10%. An actual calculation of k (non approximation) involves integrating over the area of the cantilever to find the moment of inertia. There is no simple equation as geometries vary substantially. Similarly, when cantilevers are inhomogeneous, i.e. have multiple layers, calculating k becomes increasingly difficult. In such cases spring constant can be determined experimentally by applying a force on the cantilever and measuring the bending response.

Having shown how to calculate frequency and the spring constant for a bar cantilever of uniform composition; other geometries and cases involving multiple layers will not be discussed. As mentioned earlier these cases quickly get very complex. In experiments values were experimentally determined as needed or taken from the manufacturer's data if deemed sufficiently accurate.

Force Fields and Microcantilevers

When in the presence of a force with non-zero derivative over space, microcantilevers will exhibit a change in the resonance frequency. Forces with derivatives equal to zero will simply deflect the cantilever and the lever will resonate about this new position with an unchanged frequency. (This assumes that the amount of deflection is not sufficient to move it out of the area of purely elastic material response.) This effect is very important to microcantilevers sensors and Atomic force Microscopes in general and to our application in particular. Forces

with non-zero derivatives are found near the surfaces and in most electric fields, to give two pertinent examples. The following derivation follows that of Dror Sarid. Fields with non –zero derivatives cause frequency shifts in microcantilevers by changing the effective spring constant. It is simple to determine this mathematically. Spring constant can be defined in several possible ways. Besides the usual force per distance method, spring constant can be written as a function of potential energy, W, of a deflected cantilever, which is

$$W = \frac{1}{2} k z^2$$

Taking the second derivative of energy W with respect to z,

$$k = -\frac{\partial^2 W}{\partial z^2}$$

If a force *F(z)* is present with a derivative in the direction of the deflection of the cantilever, we can add this second force to the restoring force to get the total force,

$$F(z) = F(z_0) + \frac{\partial F(z_0)}{\partial z}$$

Applying the deflection of k to the definition of W (which now includes the second force term), gives us the effective spring constant, $k_{eff} = k - F_1$, where

$F_1 = -\frac{\partial F}{\partial z}$.

Thus, a tip-sample attractive force with a positive derivative will decrease the resonance frequency of the cantilever. For tip-sample attractive forces, the

derivative will normally be equal to or greater than zero. If the derivative is negative, the tip-sample attractive force will decrease as the tip moves closer to the sample. Such a situation does occur very close to the surfaces. It arises when the repulsive (contact) force begins to rise more quickly (in normal terms) than the (attractive) van der Waals force. This occurs at approximately one angstrom and is thus irrelevant to us. Tip-sample repulsive forces will normally increase the resonance frequency of the cantilever. As in the case of the attractive force, a reversal of the sign of the force derivative (in this case from the negative to positive) is unusual and is not important to us. Note that the reversal of the sign will cause the cantilever's frequency to decrease instead of increase in repulsive forces, or increase instead of decrease for attractive forces.

It will be remembered that the frequency can be written as a function of the spring constant, k, and the effective mass, m*. In the fields of the microcantilever sensors and the AFMs (Atomic Force Microscopes), the more common convention is to use ω, as in

$$\omega = \sqrt{\frac{k}{m^*}}$$

Having found the effective spring constant under the conditions of a force with a non-derivative, we can now write the new equation for oscillation rate, namely,

$$f = \frac{1}{2\pi}\sqrt{k - \frac{F_1}{m^*}} \text{ or } \omega = \sqrt{k - \frac{F_1}{m^*}}$$

Or, alternatively, we can substitute k_{eff} for the t_{rms} *k-Ft,* this, like the use of *f* or ω, is purely a matter of personal preference.

Electric Fields and Microcantilevers

If a microcantilever is near a surface, a difference in potential between the cantilever and the surface will result in electrostatic force acting between the cantilever and the surface. Derivation of the theoretical model for the force has been done using approximations to the shape of the tip. Empirical results support this models.[7]

The electrostatic force between the cantilever and the surface is dependent on a number of different parameters, in particular: *d,* the tip-to-sample separation; *R*, the tip radius; Θ, the tip cone angle; *L*, the tip length; and *U,* the potential difference between the tip and the surface (in volts). For the tipped cantilever in the near vicinity of a surface, it is sufficient to just consider the tip because the distance between the surface and the rest of the cantilever results in the surface-beam force being small, the electrostatic force being $1/r^2$ force. Using this fact, several different geometrical representation approximations have been made for cantilever tips: the plane surface model (a circular area), the sphere model, and the charged line model (the equipotential surface from a uniformed charge line is a good approximation to a conical tip). It has been found that the sphere model works well for d<R, while the charge line model is superior when R<d<L [7,8.] Given below are the formulae for F, the tip force and F' the tip force gradient for the plane surface model, the sphere model and the van der Waal force model. Note that the electrostatic force equations are valid for d<R, zero contact potential and an electrically grounded tip. As mentioned above, the charge line model works reasonably well for R<d, even though the equation is not technically valid in that case. The van der Waals force is

dominant at low voltages while the electrostatic force is most important above several volts.[9]

Electrostatic- Sphere [6]

$$F = -\frac{\pi\varepsilon_0 R U^2}{d} \qquad F' = \frac{\pi\varepsilon_0 R U^2}{d}$$

Electrostatic - Charged Line[7]

$$F = \left(\frac{-\alpha}{4\pi\varepsilon_O}\right)\ln\left(\frac{L}{4d}\right) \mathrm{U}^2 \text{ or } F' = \frac{\alpha^2 U^2}{4\pi\varepsilon_O d}$$

$$\text{Where } \alpha = \frac{2\pi\varepsilon_o}{\sinh^{-1}\left(\frac{1}{\tan\left(\frac{\Theta}{2}\right)}\right)}$$

Van Del Waals (sphere)[10]

$$F = -\frac{HR}{6d^2} \quad F' = -\frac{HR}{6d^3}$$

Calculating the deflection of the microcantilever exposed to a force field is straightforward. Recalling that the cantilever behaves in a highly similar manner to the spring, one can simply use and rearrange Hooke's law which yields

$$\mathrm{x} = -\frac{F}{k}$$

Where x, is the distance of deflection, the minus sign is an artifact of mathematics and can be easily ignored. Note that for force fields with non-zero gradients deflection of a cantilever may cause that cantilever to move into an area of different field strength, reducing the bend below what was expected the motion is in to an

area of lesser field strength. Likewise, motion in to an area of greater field strength will make the bending greater than what was initially predicted. This effect can complicate matters. Most of the time though, for small deflection this is a minor issue. Besides causing deflection of the cantilever a tip sample force will cause a shift in resonance frequency. If the force gradient is approximately constant over the range of tip motion, the relationship between the force gradient F=∂F/∂d, and the shift in resonance frequency is Δf is

$$F = -2k\frac{\Delta f}{f_{res}}$$

Where, k is the spring constant of the cantilever, and f_{res} is the resonance frequency of the cantilever when no external force is acting upon it. [11]Based on the information present in this section, it can be seen that cantilevers with low spring constant are rather more sensitive to forces in general and electrostatic forces in particular when compared to cantilevers of higher spring constants. The lower the cantilever's spring constant, the more it will deflect if a given force is applied to it. Likewise, it can be seen from the last equation that for a given force, the change in resonance frequency will increase with a decreasing spring constant. Both of these observations make sense intuitively.

Cantilever damping rates are more complex than other parameters discussed, in large measure because of their tendency to drift substantially over time. Due to various natural processes such as changes in relative humidity. These natural drifts

are quiet large relative to the shifts they undergo due to the effect of interest. This means that they may be useful for experimentation, but do not make a very practical basis for a field detector. Some general statements can be made without entering into complex details. For a cantilever, that shows an increase in frequency as a result of exposure to a field, damping will decrease. Conversely, a cantilever that shows a decrease in frequency will damp out more quickly.

Chapter 5

Cantilever Control Systems

Introduction

By itself, a microcantilever is a very small object. Under good conditions it may be possible for someone with sharp eyesight to barely make it out. Dealing with a microcantilever – moving it around, measuring its response- is not a very straightforward process. Certain things can be done to facilitate handling and control of a microcantilever. As and example, cantilevers are attached to a small substrate wafer that can be moved around with hand tweezers. (See Figure V-a)

In this section, some important practical aspects of using microcantilevers will be discussed. The way in which a cantilever is physically dealt with (being so small) and how its mechanical response is measured will be covered. Several different methods for detection of deflection will be discussed as will the use of a cantilever when a surface is present, as well as when one is not present. Control electronics, being either an AFM (Atomic Force Microscope) controller or a direct interface using an oscilloscope and, if desired, a driving function generator, will be explained, as will different issues and problems that can arise, particularly with respect to different methods of control.

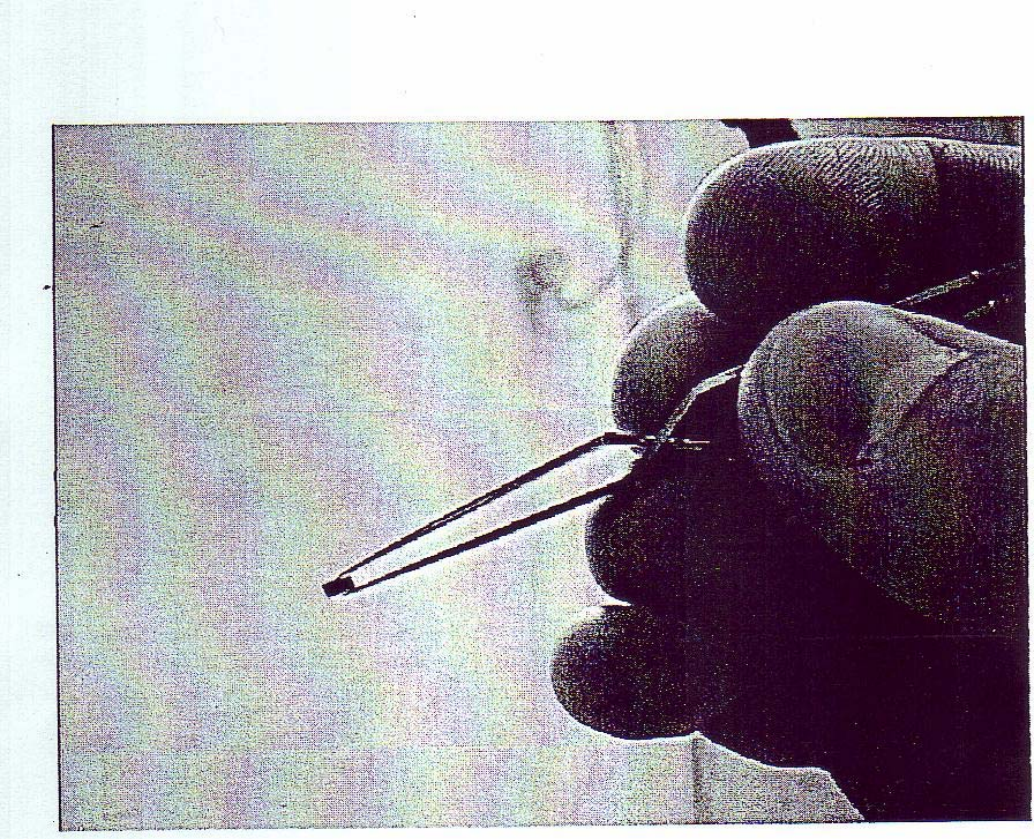

Figure V-a. Handling Microcantilevers

One or more cantilevers are attached to a substrate that is large enough to be manipulated by hand, usually with tweezers. A typical substrate is shown above held in the tweezers.

Optical Cantilever Control Using a Head

There are several different ways of controlling a cantilever. Typically, cantilevers are mounted in a head that can either be a part of a complete system assembly or run independently with some simple equipment. A head is essentially a large mounting bracket that contains equipment for moving and exciting cantilevers and for measuring cantilever deflection. (Other information, such as the vibration frequency, is extracted from the deflection data.) The cantilever itself sits in a cantilever mount that in turn fits into the head. (See Figure V-b for a diagram of a cantilever mount.) The cantilever mount serves several functions such as holding the cantilever in approximately the correct position relative to the rest of the head. (Some variation in position does occur; thus the laser used for deflection detection can be shifted slightly to allow correct focusing on the cantilever, as will be discussed shortly.) Applying an AC signal of variable frequency to a piezoelectric crystal residing with in the cantilever mount produces excitation of the cantilever. These crystals allow the microcantilever to be vibrated at a particular frequency and amplitude.

While deflection data can be generated using piezo crystals, the deflection measurement system that is typically used is a bit different. A laser is aimed at the end of a cantilever and bounces off. A Position Sensitive Detector (PSD) is positioned in the path of the deflected beam. When a cantilever bends, the angle

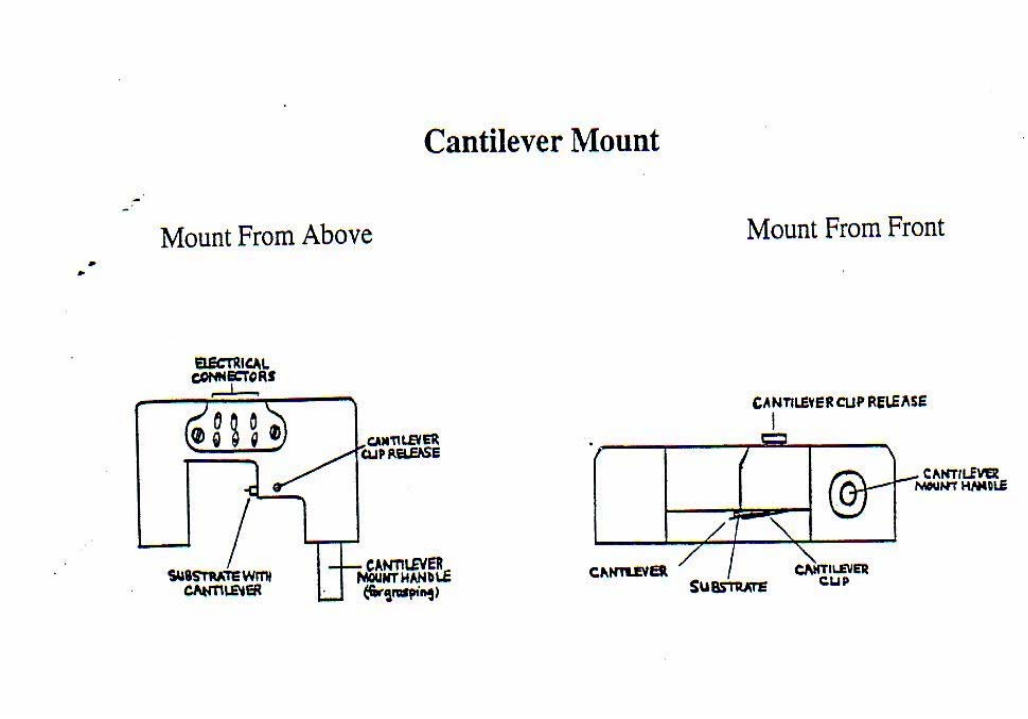

Figure V-b. Cantilever Mount

A substrate with attached cantilevers is inserted into a cantilever mount, which holds the substrate, and the cantilevers steady and can vibrate them at a particular frequency and amplitude if desired, using a built-in piezo crystal. The cantilever mount is inserted into an AFM head.

between the surface and the incoming laser beam changes causing the angle of the deflection beam to shift.

This change in angle results in the shifting of the laser across the PSD. The movement is detected by the PDS and the output signal is adjusted accordingly (the level of the output signal varies approximately linearly with the deflection of the cantilever). (See Figure V-c for the lay out of the optical detection system.) In order for this system to work, the laser must be aimed sufficiently well such that approximately half of the laser light is incident on an upper detector and half on a lower detector. (If too much laser light strikes one side of the PSD relative to the other, the relationship between deflection and change in signal breaks down.) A digital voltmeter on the AFM head registers the sum of the light intensity falling on both sides of the PSD – A+B – and measure of the light intensity on the two sides – (A-B)/(A+B)- that is used for determining cantilever deflection. (See Figure V-d for a diagram of the head and Figure V-e for a picture of one.) In the preceding equations, A and B are the intensities of the light falling on the two PSD plates. The two signals are used during set up respectively for positioning the laser beam on the cantilever beam and aiming it at the center of the PSD.

The deflection measurement system is not perfect. A more accurate way of putting this would be to say that the system measures the change in the angle of the cantilever at the point where the laser strikes it.

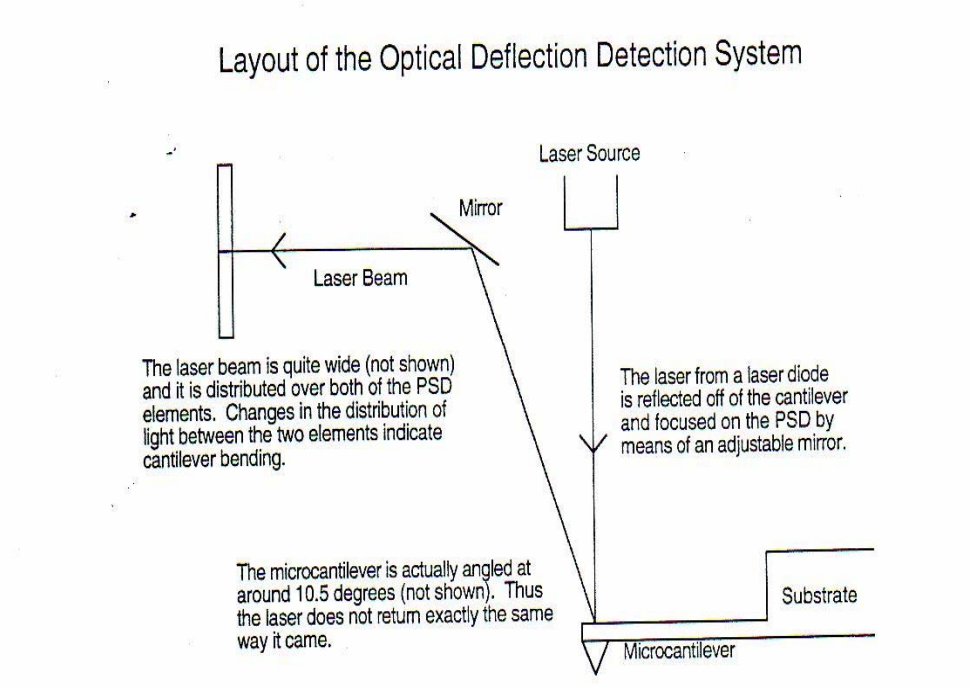

Figure V-c. Optical Deflection Detection System Layout

The most common method of detecting the cantilever motion is the optical system, which involves bouncing a laser off the end of the microcantilever. This is a convenient method because AFM head already contains most of the electronics and control hardware that is needed.

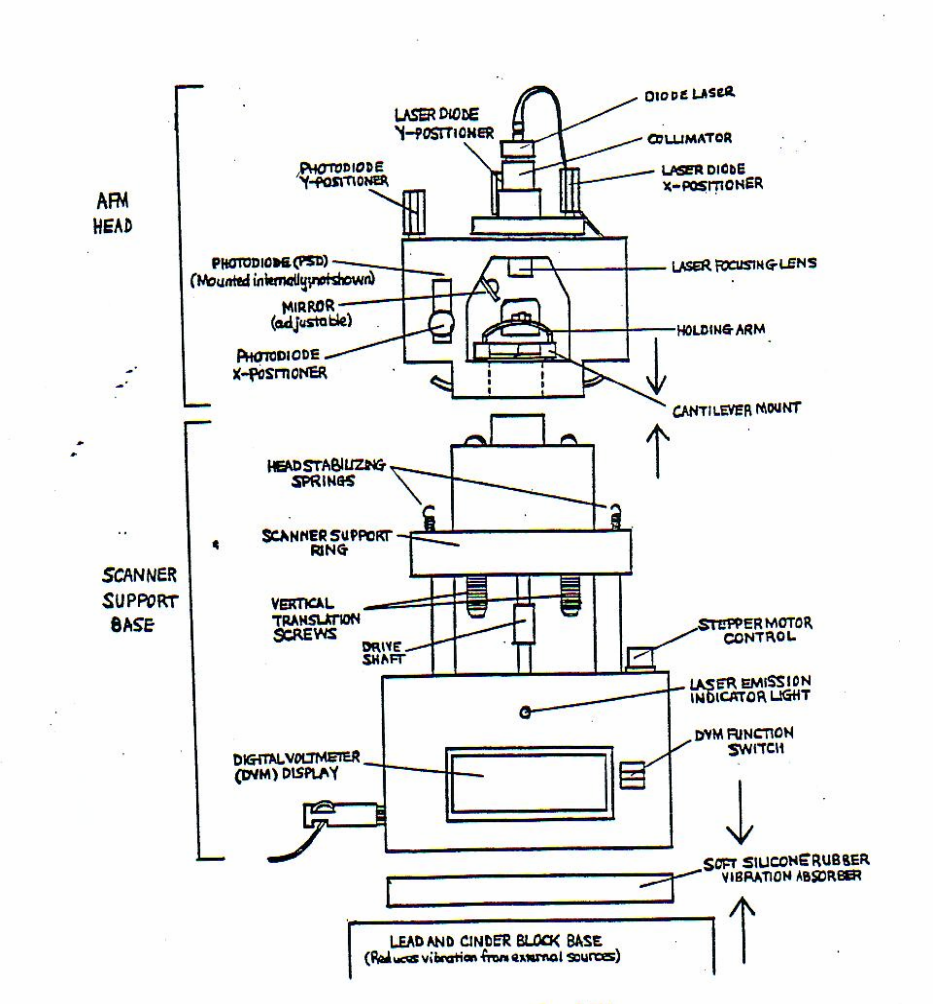

Figure V-d. AFM Head Diagram

The positioners on top allow that the laser be aimed at the cantilever tip and the reflected beam to be aimed at the center of the PSD (Position Sensitive Detector). The digital Voltmeter (DVM) display switches between two readings, one being the sum of the intensity of the beam falling on the two plates and the other being a measure of the skewness of the laser beam towards one side of the PSD or the other.

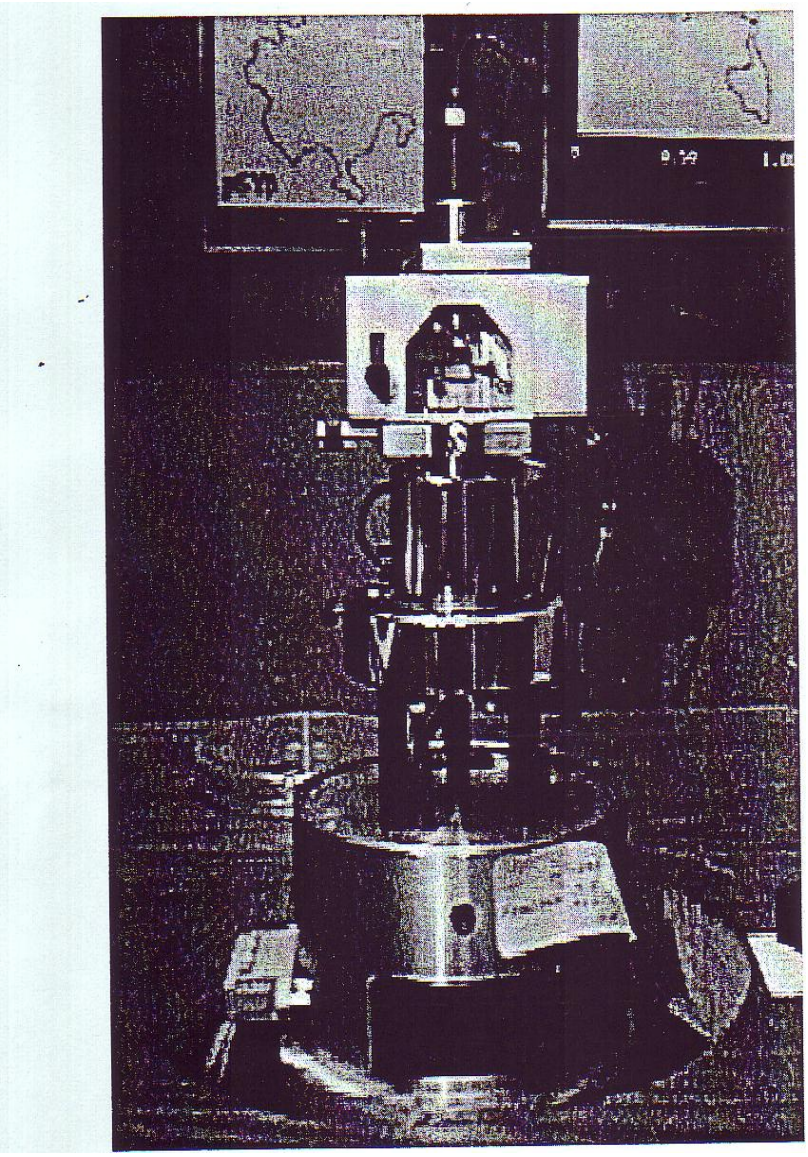

Figure V-e. Photograph of an AFM Head

Most of the items shown in the diagram (Figure V-d) can be distinguished in this photograph.

It is assumed that the change in angle is proportional to the change in the deflection. In general this is true, although the proportionality constant can change depending on the geometry of the cantilever, the angle at which the cantilever is mounted, and the point on the cantilever where the laser beam strikes it. In order to get an accurate determination of the value of dx/dV (that is, the change in cantilever deflection per unit change in output signal) it is necessary to measure a force calibration curve.

Other Methods of Deflection Detection

There are other methods available for measuring cantilever deflection. The second most common technique is the piezoresistive method in which a cantilever is made in part or in whole of a piezo resistive material. The deflection of a cantilever compresses or stretches the piezo crystal producing a voltage across it, which can be measured. For fairly small deflections the voltage produced is proportional to the bending of the lever. One disadvantage is that such cantilevers must be least 10μm thick and thus have rather high spring constants. Another is that thick cantilevers tend to be insensitive. Beyond the optical and piezoresistive methods, the most deflection methods are fairly obscure. The capacitive method was investigated by several different groups but has been largely discarded, although the Molecular Imaging Group (MIG) at Oak Ridge National Laboratory (ORNL) is working on an array of cantilevers that will use the capacitive method. This technique works by measuring the capacitance between two plates, one being the cantilever itself and the other being a small conductive area set a short distance

below the cantilever. As the distance between the plates changes due to the bending of the cantilever, the capacitance of the system changes. A big disadvantage of this system is that charge on the cantilever can change due to some external effect (such as a radiation interaction) and the system can be fooled into reporting a deflection that did not in fact occur. There are a couple of other methods (interference, STM) that are not widely used at this time.

AFM Systems

AFM controllers are often used in cantilever sensing experiments. (See Figure V-f for a simplified diagram of an AFM control system lay out.) AFM systems incorporate hardware and software for all kinds of imaging applications.

Many of these features are also useful for experiments in cantilever physics and in sensing the detection. For example, the Nanoscope series of AFM systems (the type I used in my research) are set up to easily "tune" cantilevers that are to determine their harmonic frequencies by exciting them at different frequencies and finding the frequency at which the greatest amplitude occurs. This feature is very useful in experiments in which a frequency shift is used to detect or measure something. AFM control systems are fairly complex pieces of equipment, but their basic principles of operation are fairly straightforward. A cantilever head contains a cantilever held in place above the sample surface. The system can move the sample surface closer or farther away from the cantilever tip and can vibrate the cantilever

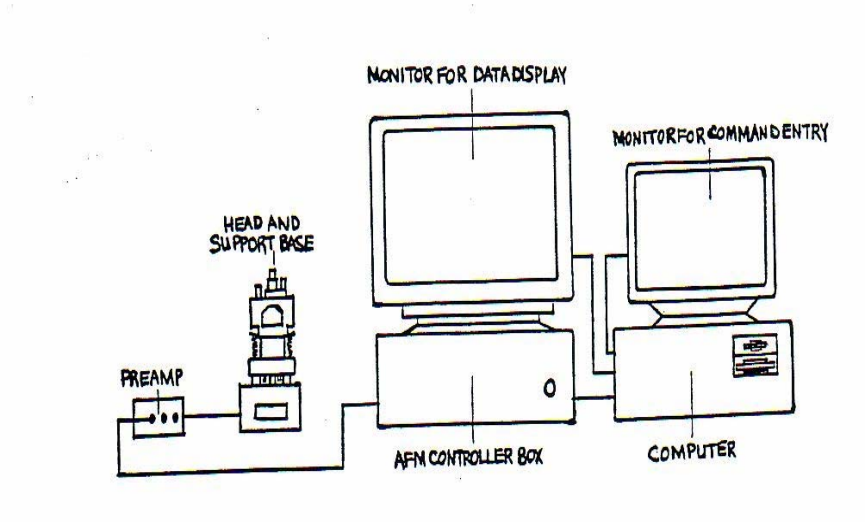

Figure V-f. Simplified AFM Control System Layout

at a frequency and strength specified by using piezoelectric crystals mounted under the sample and in the cantilever mount. Large motions can be accomplished using a combination of computer controlled and user-operated screws attached to a vertical translator. As discussed earlier, deflection is most often detected and measured using a laser-based optical system. Normally, the item or surface of interest is placed on the sample mount and the cantilever is brought into contact with it. The sample being investigated does not necessarily have to be a solid. The user might want to investigate cantilever behavior in a liquid. In this case, the cantilever might be lowered into a small puddle (formed by several drops of water) or open topped container of liquid sitting on the top of the sample mount. Cantilevers do not necessarily have to be operated near a surface using a controller. They may be operated free in the air. This permits investigation of cantilever physics and the effects of different gases on cantilever behavior. When no surface is present, the controller can do all the things that it can normally do to the cantilever except for those requiring the presence of a surface (such as a force calibration curve).

Controllers provide an interface between the AFM software on a computer and the AFM head (containing the microcantilever). Controllers take the commands from the software and the information coming in from the head , process it , and send out the necessary signals to the head to accomplish the desired result (such as moving the cantilever or vibrating it at a particular frequency and amplitude). Some functions, such as exciting the cantilever via the piezo in the sample mount, can be

accomplished without the use of the controller box. Other functions, such as moving the sample mount relative to the cantilever, cannot be done without the controller and software (i.e. a complete AFM system). Of course, the cantilever mount can be moved up and down relative to the sample (surface) mount in coarse increments (microns), but it cannot approach the angstrom level movements when using the controller and software. This makes surface or near surface work impossible without the controller.

Generally, controllers are used for surface or near surface work. The user manually positions the cantilever close to the surface (the surface mount can be raised and lowered by hand) and the system controls the final approach by the cantilever to the surface. Contact is assumed when the cantilever deflection signal increases beyond a certain level. The deflection comes as a result of the cantilever being pushed against the surface; the tip either no longer moves (for rigid samples) or does not move very much (for softer samples) when the base is moved (as the tip is now held in place by pressure and friction). This determination is made by setting a specific amount of cantilever deflection as indicated by the PSD to be a trigger to indicate that tip motion has stopped or almost stopped and the movement of the base of the cantilever is producing deflection. Upon making contact with a surface, a cantilever can then be retracted to a user – specified distance from the surface if desired. This is useful for studying electric fields around an object, for example. False contacts can be major problems when using particularly reflective samples, especially if they are flat. The laser beam in the Nanoscope's optical detection and measurement

system is not very well focused. Invariably, a significant portion of the laser footprint misses the cantilever and strikes the surface below. Generally this secondary beam is reflected in approximately the same direction as the main beam from the cantilever. (In some circumstances this beam can go directly into the PSD.) As the cantilever is moved, the signal beam from the cantilever and the secondary beam from the surface can interact. Going back and forth between constructive and destructive interference, causing the optical signal intensity to fluctuate. Sufficiently large reductions in the intensity can fool the system into registering large deflections (substantial deflection of the cantilever leads to a reduction in the signal as the laser's footprint begins to move off the PSD) and thus reporting contact with the surface. This can be a very serious problem in certain situations. Since the system ceases to move the cantilever closer to the sample when it registers contact, this can lead to necessity of repeatedly attempting to engage the surface and resetting the deflection measurement system each time. Several options for mitigating this effect exist. If the surface is flat, it can be placed at a shallow angle so that the surface-deflected beam will not interact with the signal beam as much. Another possibility is to coat the surface with a non-reflective paint –on or spray-on polymer such as Aerodag G.

Direct Interface

Cantilevers can be controlled by direct interface with the head. For example, one method is to use an output from a signal generator to drive the piezo crystal, allowing the cantilever to be vibrated. The signal output representing deflection

might be fed into a digitizing oscilloscope to monitor the micro cantilever's motion over time or a lock in amplifier to find the frequency and measure any shifts (See Figure V-g for a direct of direct head interface.) Positioning the Cantilever accurately near a surface is not really possible with this system, so a controller must be used to put the cantilever in place. Once it is in the place, control of the system can be switched from the AFM controller to the signal generator or other source of a driving function. It is not really possible to move the cantilever closer to or farther away from the surface at this point unless control is switched back to the controller, which can be a problem if an experiment, is running. This system does have several important advantages over using an AFM controller in many situations, however. One is the ability to apply a wide variety of driving functions to a cantilever that an AFM controller is not designed to provide. Another is that the direct readout can give a clearer picture of what is actually going on as AFM systems typically present data in a processed from that is dependent on certain assumptions that are made being true.

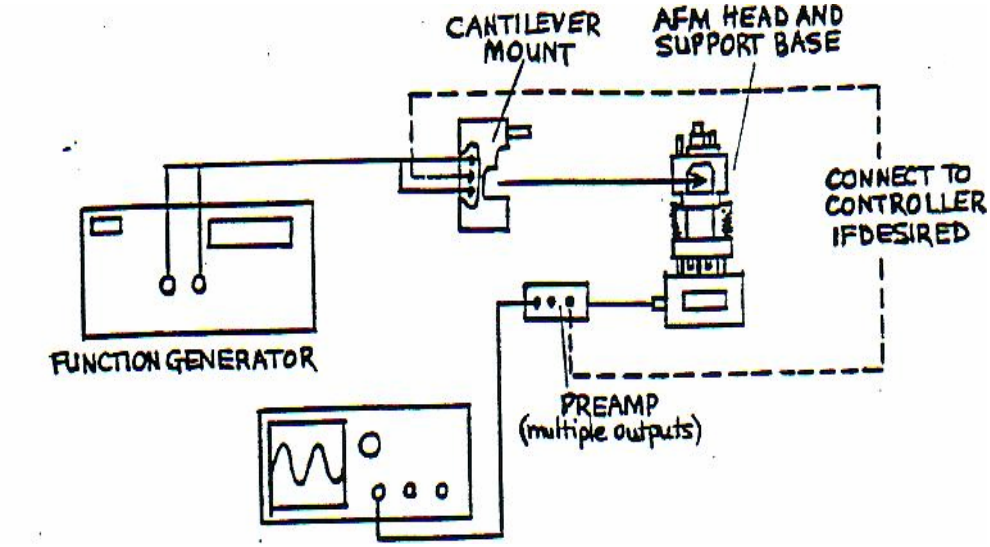

Figure V-g. Direct AFM Head Interface

This is carried out using a driving function source (such as a function generator) and some kind of readout electronics (such as a digitizing oscilloscope). Depending on what is connected directly and how, the AFM controller box and associated equipment can usually be connected if desired in order to accomplish such things as changing the separation distance between the cantilever and an adjacent surface.

Chapter 6

Experimental Procedure

The cantilevers used for this study were the Nanosensor cantilevers manufactured by Nanosensors, Germany and Mikromacsh cantilevers manufactured by SPM tips, Estonia. The Nanosensor cantilevers are made of single crystal silicon with plane orientation 100 and resistivity in the range of 0.01-0.02 Ωcm. The length of each cantilever is 450 ± 5 μm; the width of the cantilever is 50 ± 5 μm and the height of the cantilever is 10 to 15 μm. The resonance frequency of the cantilevers is 10- 17 kHz and the spring constant lies between 0.07 and 0.4 N/m. Cantilevers from the same batch were used for the experiment. The Mikromacsh Cantilevers are polycrystalline and 300 μm long, 35 μm wide and the thickness of these cantilevers is 1.3 μm.

A batch of 45 cantilevers was cleaned simultaneously using small glass sleeves. The Nanosensor cantilevers were sequentially cleaned with Acetone, Isopropyl Alcohol and Methanol individually for 10 minutes. They were then cleaned in UV for 30 minutes. After cleaning they were etched in two different (smooth and rough etched) styles accordingly. Etching is done in order to increase the surface area. When the silicon cantilever is etched, the thickness of the cantilever changes. Rough etching was done using 30% Potassium Hydroxide solution. Pure KOH solution attacks the silicon in the 100 plane, producing characteristic anisotropic V-etch with side walls that form a 54.7º with the surface, resulting in the formation of

micro-pyramids, which enhances the surface area. The arrangement of the rectangular based micro-pyramids on the {100} etch bottom depends on the etching time, etching position of the chip (vertical or horizontal) and on the oxygen content in connection with the thermal history of the wafer material. In order to obtain a smooth finish on the cantilevers surface, the Potassium Hydroxide solution was prepared by weighing 1 part KOH pellets (by weight) into a plastic beaker and adding 2 parts DI water. 100 g KOH with 200 ml water was used and mixed on a warm surface until the KOH has completely dissolved, 1% by volume of the Potassium Hydroxide solution was isopropyl alcohol, and this was achieved by adding 40 ml of isopropyl alcohol to the Potassium Hydroxide solution. The reason for smooth etching the cantilevers was to get a good comparison of deflection differences between cantilevers from both categories that had a comparable resonance frequency. The surface roughness of the cantilevers was quantified using atomic force microscopy imaging of the cantilever.

In order to obtain the right parameter for etching, the cantilevers were placed on small thin strips of gel packs. They were then inverted on to a plastic container with the rough etch KOH solution and similarly another batch of smooth etched cantilevers were immersed in smooth etch KOH solution, for a series of different time periods of 5, 10, 15, 20 and 25 minutes. The cantilevers were then mounted on to the flow cell of the single cantilever system and the resonance frequency was then measured for each cantilever. Titanium and gold were evaporated on the cantilevers. The electron beam evaporator is used to deposit thin films of Titanium

Gold onto substrates. A rotating motor that would hold the target cantilevers was mounted inside the evaporator. The motor was rotated at a constant speed and was mounted at 35º angle in order to maintain the roughness in the rough etched cantilevers on evaporation of Gold. Evaporation is done under a high vacuum in a bell jar chamber. Evaporation is achieved by heating a source with an electron beam. As the source material evaporates, it forms a thin film of gold is deposited on the cantilever surface. Titanium was used as an adhesion layer, between Gold and Silicon. 3.5 Nm of Ti and 35 Nm of Au were deposited on the Nanosensor Cantilevers.

Deflection Experiment

The most common read out technique for cantilever motion is the optical beam deflection technique. A light beam from the laser is focused at the end of the cantilever, and reflected onto a PSD (Sarid 1994). The bending of the cantilever changes the radius of curvature of the cantilever, resulting in a large change in the direction of the reflected beam. The dc signal provides the cantilever bending, while the ac signal yields the resonance frequency and the Q-factor. The flow cell and the flow system comprising of the syringe pump and the valves were cleaned several times and were rinsed with ethanol. Cantilevers are mounted onto the flow cell at a 12.5º angle with the Silicon side of the cantilever facing the laser. The buffer used is Ethyl Alcohol (absolute 200 proof) and the flow rate is 4 ml/hr. The in flow, the out flow and the O-ring cap is filled with the buffer. The flow cell is mounted onto the AFM head and the laser is focused on to the tip of the Silicon side of the

cantilever. The solution used for injection is 1-Do-decan thiol at a concentration 8.4mM. Alkane-thiol was the choice as it forms a very strong bond with gold and hence when thiol is injected into the flow system it would result in the bending of the cantilever as a consequence of adsorbing to the gold receptor layer. The Alkane thiol solution is introduced into the system only after a steady base line is obtained. Software packages used for data analysis are Agilent BenchLink Data Logger and Slant Nano. Agilent BenchLink Data Logger is designed to make it easy to use the Agilent 34970A Data Acquisition/Switch Unit with the PC for gathering and analyzing measurements. Slant Nano is written for fitting a Lorentzian Function to a given X-Y data set downloaded form the spectrum analyzer -SRS-760. The deflection experiment for each type of cantilever (i.e.) rough etched, smooth etched and un-etched Cantilevers was carried out and the bending deflection on thiol immobilization was measured.

AFM Imaging

After the deflection experiment, the cantilever surface was imaged using tapping mode AFM. A cantilever with attached tip is oscillated at its resonant frequency and scanned across the sample surface. Constant oscillation amplitude (and thus a constant tip-sample interaction) is maintained during scanning. Typical amplitudes are 20-100nm. The amplitude of the oscillations changes when the tip scans over bumps or depressions on a surface. Tapping mode AFM is used to scan the surface of the rough etched, smooth etched and UN- etched cantilevers.

Chapter 7

Results and Discussion

In general the sensitivity and specificity of microcantilever sensors can be optimized by careful geometric design of the cantilever, its surface topology and its coatings. For example, the mass sensitivity of a cantilever is proportional to $(\rho d)^{-1}$, where ρ is the density of the cantilever material and d is the thickness of the cantilever. Therefore by reducing the thickness of the cantilever, mass sensitivity can be improved by several orders of magnitude. The cantilever deflection approach requires long cantilevers with smaller spring constants. In order to establish an optimum standard for comparing two cantilevers etched in different solutions of KOH, cantilevers with close values of resonance frequencies in both air (refer **Table (i)**) and in liquid (refer **Table (ii)**) were chosen for the optical deflection method. Cantilevers that were etched for 10 minutes in the pure 30% KOH solution were compared with cantilevers that were treated in (30%) KOH + Isopropyl alcohol solution for 20 minutes, as they had very close, comparable resonance frequencies (refer **Table (iii)**). Thickness is calculated from the from the resonance frequency measurements using the formula

$$\frac{f+\Delta f_1}{f+\Delta f_2} = 1 - \frac{\Delta t^2}{t^2}$$

Where Δf_1 is the shift in the resonance frequency measurement in air and Δf_2 is the shift in the resonance frequency measurement in liquid.

Table (i) Resonance Frequency Measurements in Air

# of Cantilevers	Rough Etched Cantilever (Etch time = 10 minutes)	Smooth Etched Cantilever (Etch time = 20 minutes)	Un-etched Cantilever
1	17.368	18.763	23.431
2	17.669	18.985	22.897
3	18.327	18.541	22.564
4	18.615	19.219	23.392
Average	17.994	18.877	23.071

Table (ii) Resonance Frequency Measurements in liquid

# of Cantilevers	Rough Etched Cantilever (Etch time = 10 minutes)	Smooth Etched Cantilever (Etch time = 20 minutes)	Un-etched Cantilever
1	10.617	9.732	12.165
2	11.365	10.089	12.072
3	10.167	9.895	11.271
4	10.439	10.061	11.983
Average	10.647	9.944	11.873

Table (iii): Etch Time Vs Resonance frequency in Fluid (Average)

Time	Resonance Frequency Rough Etched	Resonance Frequency Smooth Etched
10 Minutes	10.647 kHz	7.967 kHz
20 Minutes	14.611 kHz	9.944 kHz
30 Minutes	Cantilever Curls up	11.0193 kHz

Based on the resonance frequency results the smooth etched cantilevers that had a resonance frequency close to the rough etched ones were used for deflection experiments. The reason being cantilevers with comparable resonance frequencies have a comparable thickness. Based on the resonance frequency measurements the average values of thickness of the rough etched cantilever is determined to be 1.2925 ± 8.83% microns and thickness of the smooth etched cantilever is 1.207 ± 5.96% microns.

The origin of the adsorption induced force is assumed to be surface stress variation due to molecular adsorption. .The Cantilever that was smoothly etched (1.32 μm-thickness) had the largest deflection (See Figure VII-a). While the rough etched cantilever (1.27 μm-thicknesses) showed only a considerable increase in bending deflection but were lesser when compared to the smoothly etched cantilevers in spite of their larger surface area. **Table (iv)** gives details of the cantilever parameters of the cantilevers compared in this study.

Table (iv) –Cantilever Parameters

Cantilever Type	Length	Width	Thickness
Unetched Cantilever	450 ± 5 μm	50 ± 5 μm	2 μm
Rough-etched Cantilever	450 ± 5 μm	50 ± 5 μm	1.27 μm
Smooth-etched Cantilever	450 ± 5 μm	50 ± 5 μm	1.30 μm
Mikromacsh Cantilever	300 ± 5 μm	35 ± 3 μm	1.3 μm

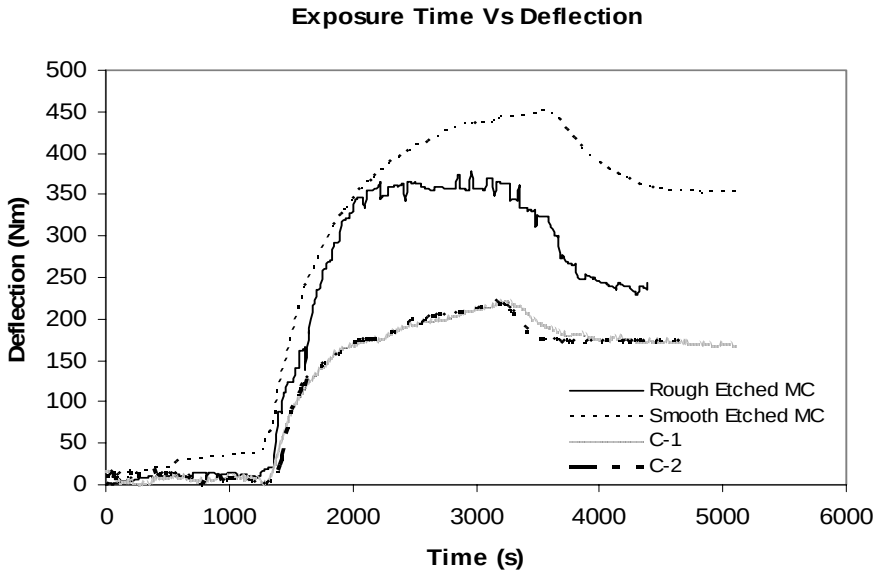

Figure VII-a. Time Vs Deflection

Shows the deflection details of the various cantilevers compared

The Un-etched cantilevers (2.0μm-thicknesses) and Mikromacsh- microcantilevers (1.3 μm-thicknesses) were used as the control and had the least deflection.

My results indicate that an increase in surface area does not increase the bending capabilities of a microcantilever; a smoother surface provides a better platform for the formation of a Self Assembled Monolayer. In terms of surface stress, the Unetched Nanosensor microcantilevers and the Micromacsh microcantilevers had higher values when compared to the rough etched and smooth etched cantilevers indicating that surface stress is largely influenced by the surface morphology of the cantilevers receptive layer (See Figure VII-b).

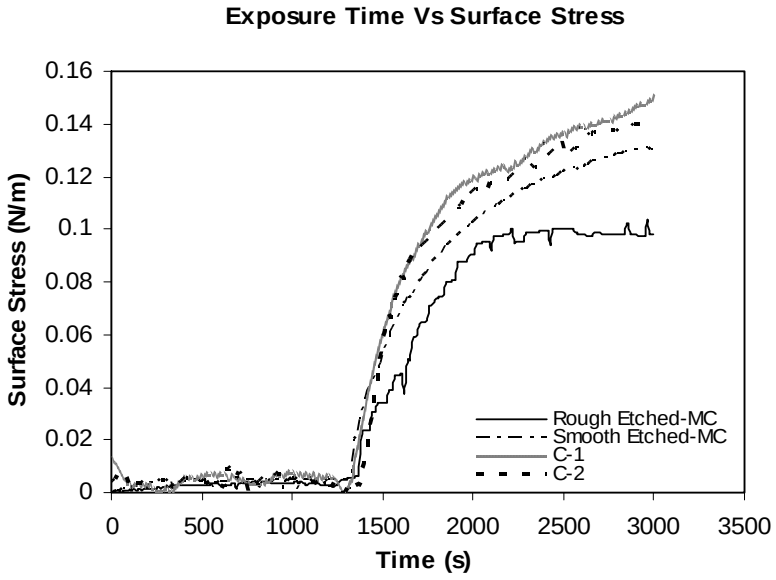

Figure VII-b. Time Vs Surface Stress

Points out that surface stress is the higher for cantilevers with smoother surfaces indicating that SAM formation is affected by the surface features of the cantilever.

This suggests that substrate morphology influences the Self assembled monolayer structure and the kinetics of SAM formation. The surface features of the cantilevers used in this experiment were quantified using tapping mode AFM. Based RMS roughness values the Rough etched Nanosensors (See Figure VII-c) had the highest RMS roughness value of 12.789Nm. The commercially available cantilevers have the lowest values for RMS roughness and highest surface stress. The Surface features of the smooth and commercial cantilevers too were monitored using tapping mode AFM (See Figure VII-d, VII-e). Despite its larger surface area the rough etched cantilever showed lower surface stress values as set against to the smooth etched cantilevers and commercial cantilevers.

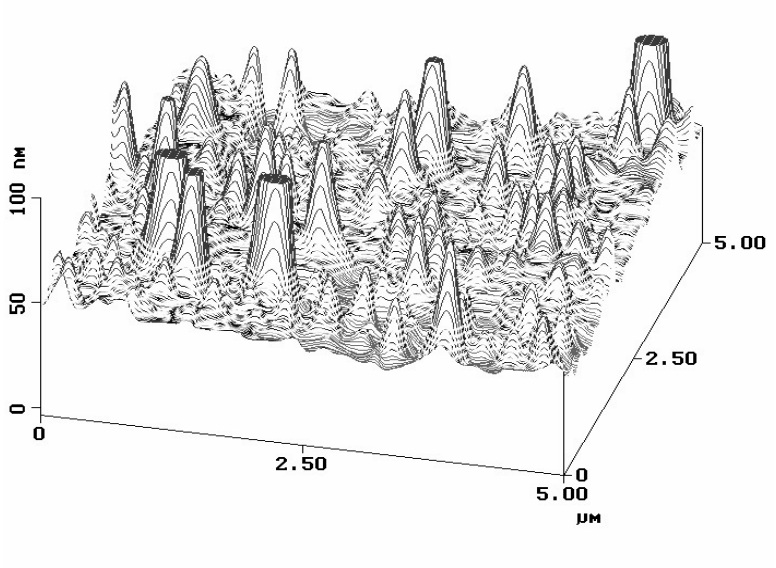

Figure VII-c. Surface Image of a Rough Etched Cantilever

RMS Roughness =12.789Nm

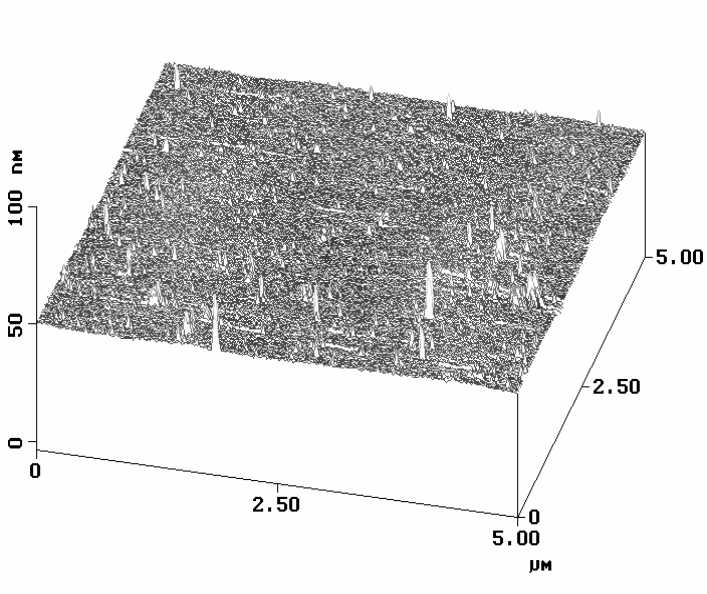

Figure VII-d. Surface Image of a Smooth Etched Cantilever

RMS Roughness =2.362 Nm

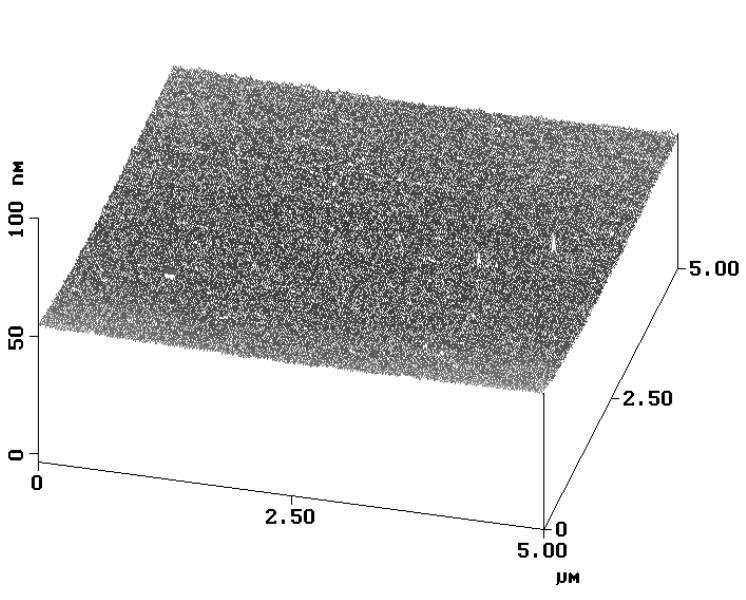

Figure VII-e: Surface Image of Commercial Cantilever (C1)

RMS Roughness =0.895 Nm

According to the Langmuir adsorption model, the Alkane coverage θ of the cantilever can be described by $\theta \propto$ 1- exp ($^{-k_{obs}t}$) where t is the adsorption time. The observed rate constant k_{obs} is a result of the rate constant for Alkanethiol adsorption k_{ads} as well as Alkanethiol desorption k_{des} and is given by $k_{obs} = k_{ads}\, c + k_{des}$; where c is the molar concentration of the Alkanethiol solution. Since the thiol-gold binding is very strong, the desorption rate constant is expected to be low. The equilibrium binding constant of the adsorption of Alkanethiol on gold can be calculated as $k_e = k_{ads} / k_{des}$ and the corresponding free energy change of the process is $\Delta G = -RT \ln(k_e)$; where R is the gas constant and T is the ambient temperature (T = 293 K). We see that in the case of the rough etched cantilever the Langmuir

model does not fit well with the stress curve unlike the other cases. These results show that both the kinetics of SAM formation and the resulting SAM structure are strongly influenced both by the surface structure of the underlying substrate. The adsorption fit (Figures-VII-f, VII-g, VII-h VII-i) are well defined by 1st order Langmuir equation. The k_{ads} values for the smooth etched and the two commercial cantilevers (Unetched Nanosensor and Mikromacsh) lie close to one another and are 0.257 $M^{-1}s^{-1}$, 0.259 $M^{-1}s^{-1}$, 0.271 $M^{-1}s^{-1}$ respectively , while the k_{ads} for a rough etched cantilever is 0.346 $M^{-1}s^{-1}$ which is slightly on the higher side. Similarly the k_{des} values for the smooth etched and the two commercial cantilevers are 0.0006029 s^{-1} , 0.0004564 s^{-1} and 0.0005856 s^{-1} respectively, while the k_{des} for the cantilever with a rough surface is 0.001567 s^{-1} indicating that the adsorption and desorption rates are higher for a rough etched cantilever.

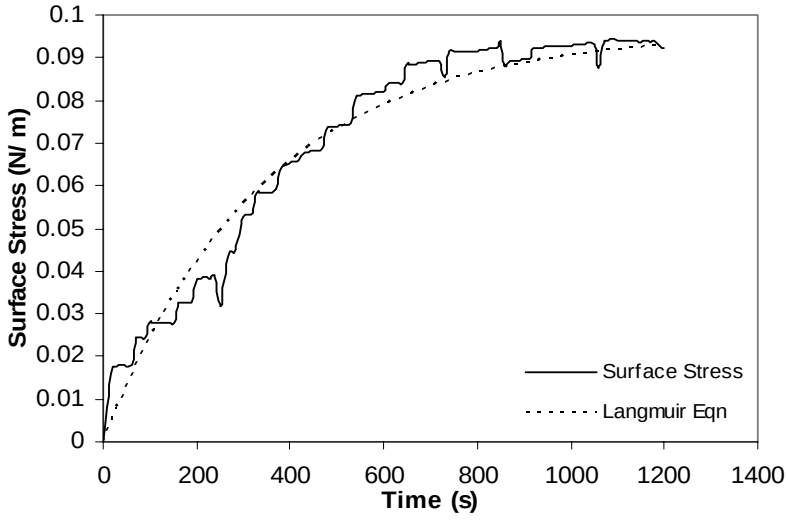

Figure VII-f. Adsorption Fit - Rough Etched Cantilever

Adsorption fit using 1st order Langmuir equation for a rough etched cantilever

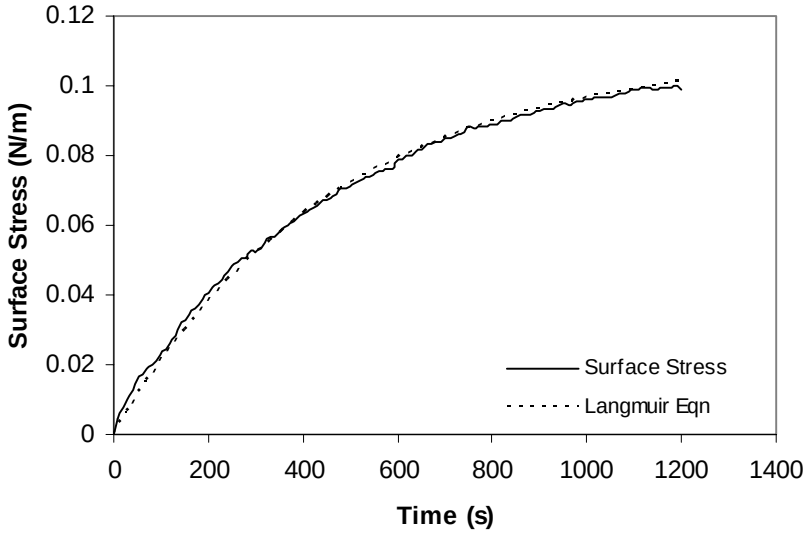

Figure VII-g. Adsorption Fit - Smooth Etched Cantilever

Adsorption fit using 1st order Langmuir equation for a smooth etched cantilever

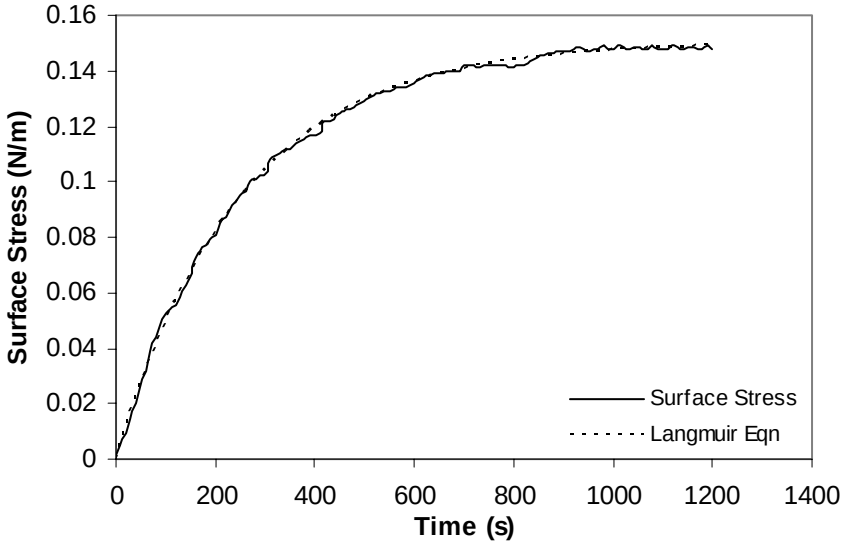

Figure VII-h. Adsorption Fit - Commercial Cantilever (C1)

Adsorption fit using 1st order Langmuir equation for an Un-etched cantilever

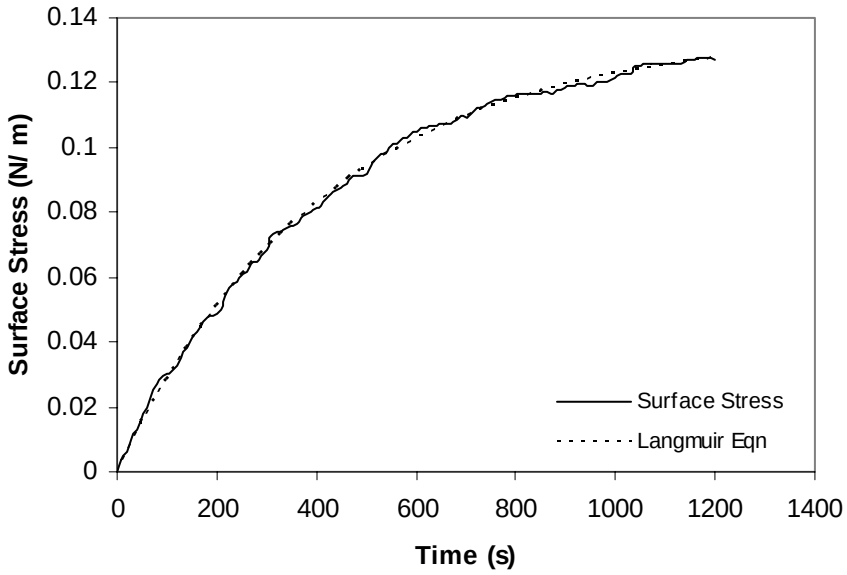

Figure VII-i. Adsorption Fit - Commercial Cantilever (C2)

Adsorption fit using 1st order Langmuir equation for the polycrystalline commercially available cantilever.

The free energy change associated with this process for the four types of cantilevers compared (Smooth etched, Unetched Nanosensor, Mikromacsh and Rough etched) in this study are -14.751 kJ M^{-1} , -15.556 kJ M^{-1}, -14.948 kJ M^{-1} and -13.230 kJ M^{-1}.

The immobilization curve has been fitted with the Langmuir isotherm (Figures VII-j, VII-k, VII-l, VII-m), which reflects the adsorption of Alkanethiols.

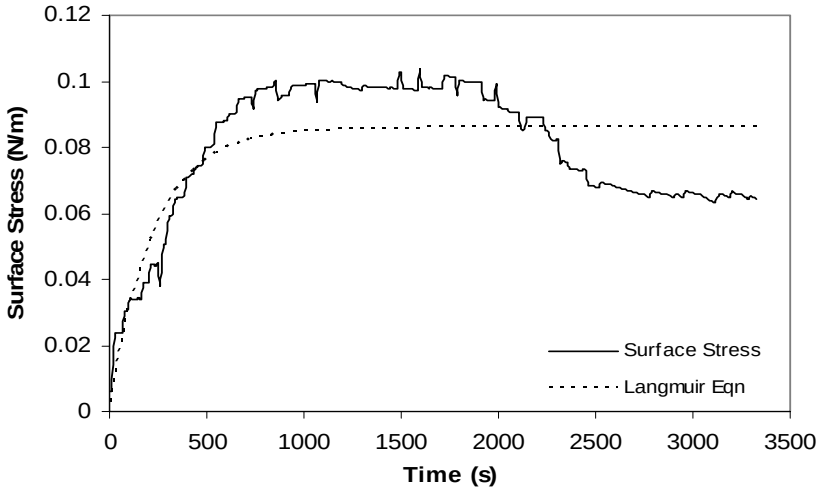

Figure VII-j. Langmuir Isotherm - Rough Etched Cantilever

Shows that a rough etched cantilever is not very well described by the Langmuir isotherm suggesting the influence of surface morphology on adsorption kinetics of 1-dodecanthiol SAM formation.

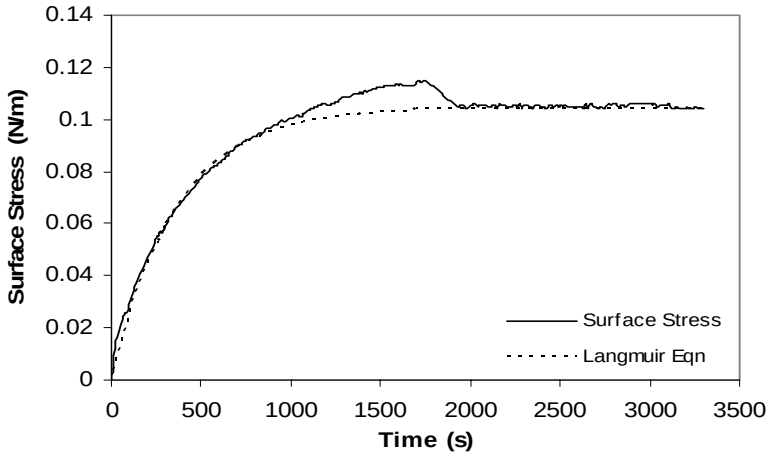

Figure VII-k. Langmuir Isotherm - Smooth Etched Cantilever

Shows that the Langmuir isotherm for a Smooth etched cantilever, it describes well the first phase and the last part of the immobilization curve.

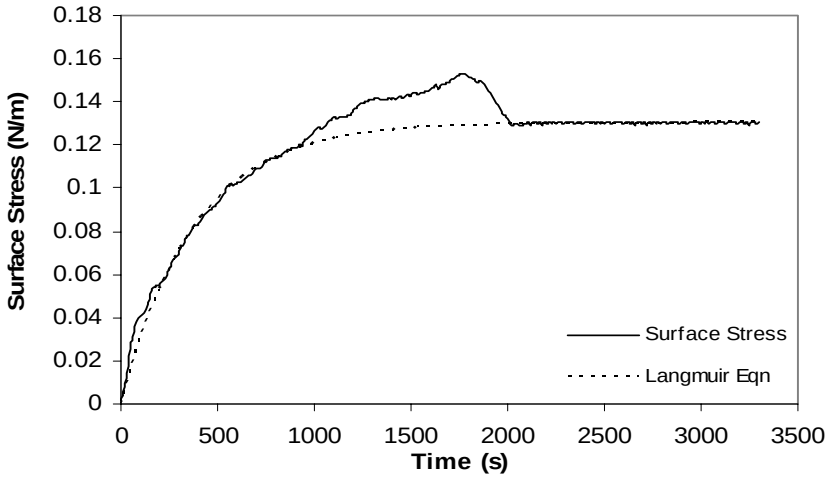

Figure VII-l. Langmuir Isotherm - Commercial Cantilever (C1)

For an Un-etched cantilever- (C1) the Langmuir equation describes well the first and last part of the stress curve and is similar to the Smooth –etched cantilever.

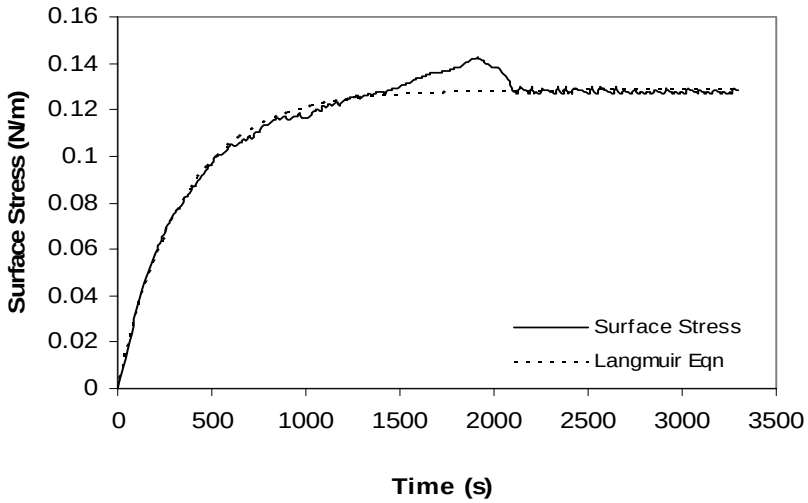

Figure VII-m. Langmuir Isotherm - Commercial Cantilever (C2)

1st order Langmuir equation fit for the commercial cantilever (C2), shows a good fit of the stress curve and also fits well in the last part of the stress curve where the loosely bound thiol molecules are washed of by ethanol

As the stress curve follows Langmuir model characteristics it implies that the surface stress is proportional to the number of adsorbed molecules. We see that in the case of the rough etched cantilever the Langmuir model does not fit well with the stress curve unlike the other cases. These results show that both the kinetics of SAM formation and the resulting SAM structure are strongly influenced both by the surface structure of the underlying substrate.

Conclusions

Microcantilevers undergo bending due to molecular adsorption when adsorption is confined to a single receptive layer. The origin of the adsorption-induced force is assumed to be surface stress variation due to molecular adsorption. Unlike earlier results obtained by P G. Datskos et.al. , which mention that an increase in surface area drastically increases the bending and in surface stress, I observed that cantilevers that were smoothly etched were the ones that had the largest deflection and higher values of surface stress, in this respect my results agree with results produced by Michael Godin et.al, where in the cantilevers with flat gold surfaces have larger surface stress when compared to cantilevers with grainy gold surfaces. Thus the surface stress is significantly influenced by the topology of the cantilever's active receptor layer. Our results indicate that an increase in surface area does not increase the bending of a microcantilever; a smoother surface provides a better platform for the formation of a Self Assembled Monolayer.

These experiments offer the opportunity for monitoring the immobilization processes and yield a direct real-time measure of the stress in the formed Alkanethiol monolayer. It has been recently reported that a maximum surface-stress change is seen on flat gold surfaces. The stress change is approximately 1000 times higher than as observed here. It is difficult to find a single explanation for the large difference in observed surface-stress signals; since the experiment here is done in liquid phase and cantilever's configuration differ. However, we find it likely that the difference in stress signals is related to variations in the adsorption properties of the used cantilever surfaces. Thus cantilever-based sensor can be used to obtain new knowledge on the stress formation and the kinetics related to immobilization processes. The sensor is thus not limited to DNA or protein detection but can be used as a technology platform for understanding kinetics involved in the interactions between a diverse array of biological and chemical substances.

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