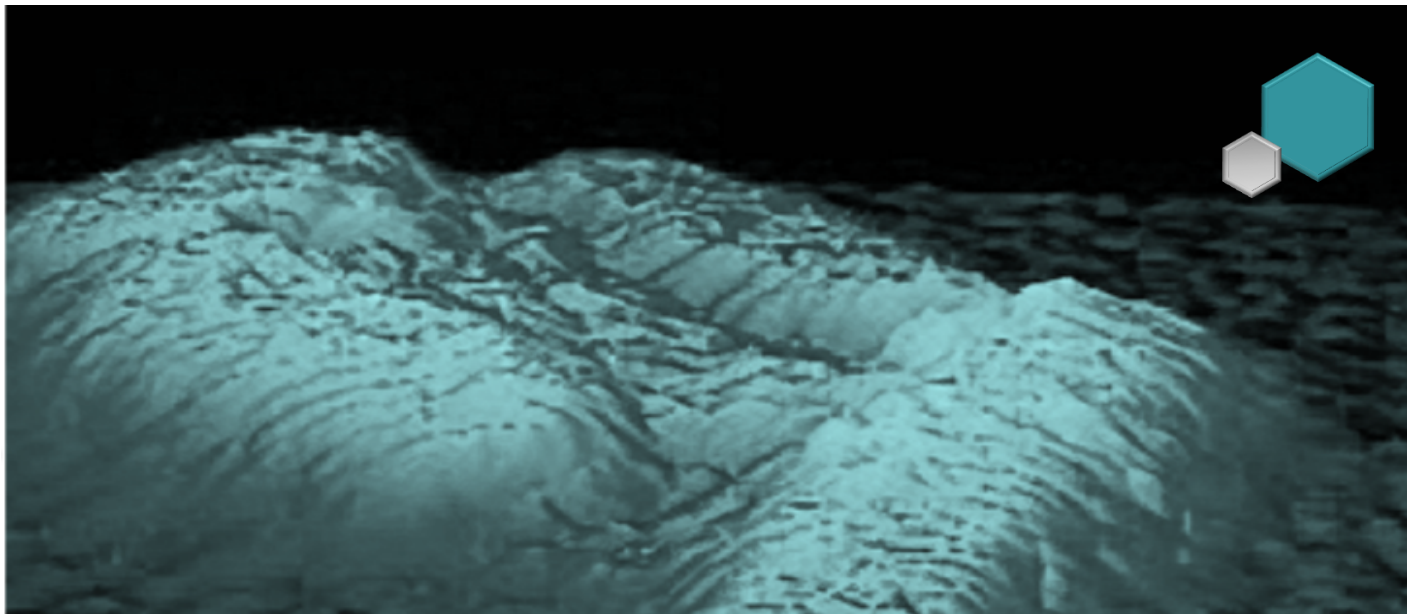


PHYSICS: Atomic Force Microscopes

How does an Atomic Force Microscope work?



PARTS 4: Instrumentation Design: Statistical Analysis with Instrumentation

*Image Credit: SA*Atomic Force Microscope Image, *Escherichia coli*--. C.J. Kazilek, Arizona State University. (2001).
https://www.nsf.gov/news/mmg/mmg_disp.jsp?med_id=51835&from=search_list

Files Included for this Activity	5E Lesson Plans	Activity Lab Sheets*
PART 4: Instrumentation Design: Statistical Analysis with Instrumentation	✓	✓

*The activity Lab sheets are located at the end of this document.

Additional Files Available for this Series	5E Lesson Plans	Activity Lab Sheets*
PART 1: Understanding How Forces Impact Cantilevers & Springs	✓	✓
PART 2: Hooke's Law: Calculating the Relationship Between Deflection & Applied Force in Cantilevers	✓	✓
PART 3: Instrument Design: Understanding How AFMs Use Optics & Reflected Light to Determine Sample Properties	✓	✓

This module was developed by Professors Vernita Gordon and Alexandra Eusebi, with assistance from research & UTeach intern Khusbu R. Dalal, at the University of Texas, Austin, with funding provided by the National Science Foundation, Division of Civil, Mechanical, and Manufacturing Innovation, award numbers 1727544 and 2150878, to Vernita Gordon.

If you use any part of this module, please send an email describing your experience to Professor Gordon, gordon@chaos.utexas.edu. Please include the approximate number of students taught. Documenting this module's use and effectiveness will help us obtain more funding for outreach and education in the future.

LESSON PLAN:

Atomic Force Microscopes

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Driving questions for lesson:

What are AFMs and how do they use forces to image surfaces on the nanometer scale?

PART 1

Understanding How Forces Impact Cantilevers & Springs

- How do Atomic Force Microscopes use forces to map surfaces?
- How do forces impact a cantilever's movements?

PART 2

Exploring Hooke's Law with the Deflection of Cantilevers

- How can we describe a cantilever system with Hooke's law?
- How can we use mathematical models to describe a cantilever?

PART 3

Instrumentation Design: How AFMs Use Incident & Reflected Light

- How are angle of incidence and angle of reflection used to map a cantilever's deflection
- Extension - How can we use statistical analysis to determine the effectiveness of a given experimental setup?

PART 4

Instrumentation Design: Statistical Analysis with Instrumentation

- How can we use statistical analysis to determine the effectiveness of a given experimental setup?
- How do statistical tests help scientists determine the validity of an experiment and its results?

LESSON PLAN:

Atomic Force Microscopes

Texas Essential Knowledge and Skills (TEKS)

This lesson was developed as cross-curricular, supporting Physics and Mathematics TEKS.

Part 1	Part 2	Part 3	Part 4	§112.39. Physics
				(2) Scientific processes. The student uses a systematic approach to answer scientific laboratory and field investigative questions. The student is expected to:
√	√	√	√	(D) design and implement investigative procedures, including making observations, asking well defined questions, formulating testable hypotheses, identifying variables, selecting appropriate equipment and technology, evaluating numerical answers for reasonableness, and identifying causes and effects of uncertainties in measured data;
	√	√	√	(E) demonstrate the use of course apparatus, equipment, techniques, and procedures, including multimeters (current, voltage, resistance), balances, batteries, dynamics demonstration equipment, collision apparatus, lab masses, magnets, plane mirrors, convex lenses, stopwatches, trajectory apparatus, graph paper, magnetic compasses, protractors, metric rulers, spring scales, thermometers, slinky springs, and/or other equipment and materials that will produce the same results;
	√	√	√	(F) use a wide variety of additional course apparatus, equipment, techniques, materials, and procedures as appropriate such as ripple tank with wave generator, wave motion rope, tuning forks, hand-held visual spectrosopes, discharge tubes with power supply (H, He, Ne, Ar), electromagnetic spectrum charts, laser pointers, micrometer, caliper, computer, data acquisition probes, scientific calculators, graphing technology, electrostatic kits, electroscope, inclined plane, optics bench, optics kit, polarized film, prisms, pulley with table clamp, motion detectors, photogates, friction blocks, ballistic carts or equivalent, resonance tube, stroboscope, resistors, copper wire, switches, iron filings, and/or other equipment and materials that will produce the same results;
	√	√	√	(G) make measurements with accuracy and precision and record data using scientific notation and International System (SI) units;
	√	√	√	(H) organize, evaluate, and make inferences from data, including the use of tables, charts, and graphs;
√	√	√	√	(I) communicate valid conclusions supported by the data through various methods such as lab reports, labeled drawings, graphic organizers, journals, summaries, oral reports, and technology-based reports; and
	√	√	√	(J) express relationships among physical variables quantitatively, including the use of graphs, charts, and equations.
				(3) Scientific processes. The student uses critical thinking, scientific reasoning, and problem solving to make informed decisions within and outside the classroom. The student is expected to:
√	√	√	√	(A) analyze, evaluate, and critique scientific explanations by using empirical evidence, logical reasoning, and experimental and observational testing, so as to encourage critical thinking by the student;
				(4) Science concepts. The student knows and applies the laws governing motion in a variety of situations. The student is expected to:
	√	√		(D) calculate the effect of forces on objects, including the law of inertia, the relationship between force and acceleration, and the nature of force pairs between objects using methods, including free body force diagrams.

LESSON PLAN:

Atomic Force Microscopes

Part 1	Part 2	Part 3	Part 4	§111.39. Algebra I
				(1) Mathematical process standards. The student uses mathematical processes to acquire and demonstrate mathematical understanding. The student is expected to:
	√	√	√	(B) use a problem-solving model that incorporates analyzing given information, formulating a plan or strategy, determining a solution, justifying the solution, and evaluating the problem-solving process and the reasonableness of the solution;
	√	√	√	(C) select tools, including real objects, manipulatives, paper and pencil, and technology as appropriate, and techniques, including mental math, estimation, and number sense as appropriate, to solve problems;
	√	√	√	(D) communicate mathematical ideas, reasoning, and their implications using multiple representations, including symbols, diagrams, graphs, and language as appropriate;
	√	√	√	(E) create and use representations to organize, record, and communicate mathematical ideas;
	√	√	√	(F) analyze mathematical relationships to connect and communicate mathematical ideas; and
	√	√	√	(G) display, explain, and justify mathematical ideas and arguments using precise mathematical language in written or oral communication.
				(2) Linear functions, equations, and inequalities. The student applies the mathematical process standards when using properties of linear functions to write and represent in multiple ways, with and without technology, linear equations, inequalities, and systems of equations. The student is expected to:
	√	√	√	(B) write linear equations in two variables in various forms, including $y = mx + b$, $Ax + By = C$, and $y - y_1 = m(x - x_1)$, given one point and the slope and given two points;
	√	√	√	(C) write linear equations in two variables given a table of values, a graph, and a verbal description;
				(3) Linear functions, equations, and inequalities. The student applies the mathematical process standards when using graphs of linear functions, key features, and related transformations to represent in multiple ways and solve, with and without technology, equations, inequalities, and systems of equations. The student is expected to:
	√	√	√	(A) determine the slope of a line given a table of values, a graph, two points on the line, and an equation written in various forms, including $y = mx + b$, $Ax + By = C$, and $y - y_1 = m(x - x_1)$;
	√	√	√	(C) graph linear functions on the coordinate plane and identify key features, including x-intercept, y-intercept, zeros, and slope, in mathematical and real-world problems;
				(4) Linear functions, equations, and inequalities. The student applies the mathematical process standards to formulate statistical relationships and evaluate their reasonableness based on real-world data. The student is expected to:
			√	(A) calculate, using technology, the correlation coefficient between two quantitative variables and interpret this quantity as a measure of the strength of the linear association;
	√	√	√	(C) write, with and without technology, linear functions that provide a reasonable fit to data to estimate solutions and make predictions for real-world problems.
	√	√	√	(F) analyze mathematical relationships to connect and communicate mathematical ideas; and
	√	√	√	(G) display, explain, and justify mathematical ideas and arguments using precise mathematical language in written or oral communication.

LESSON PLAN:

Atomic Force Microscopes

Part 1	Part 2	Part 3	Part 4	§111.4. Geometry
				(1) Mathematical process standards. The student uses mathematical processes to acquire and demonstrate mathematical understanding. The student is expected to:
√	√	√	√	(D) communicate mathematical ideas, reasoning, and their implications using multiple representations, including symbols, diagrams, graphs, and language as appropriate;
√	√	√	√	(E) create and use representations to organize, record, and communicate mathematical ideas;
√	√	√	√	(F) analyze mathematical relationships to connect and communicate mathematical ideas; and
√	√	√	√	(G) display, explain, and justify mathematical ideas and arguments using precise mathematical language in written or oral communication.
				(7) Similarity, proof, and trigonometry. The student uses the process skills in applying similarity to solve problems. The student is expected to:
		√	√	(B) apply the Angle-Angle criterion to verify similar triangles and apply the proportionality of the corresponding sides to solve problems.
				(9) Similarity, proof, and trigonometry. The student uses the process skills to understand and apply relationships in right triangles. The student is expected to:
		√	√	(A) determine the lengths of sides and measures of angles in a right triangle by applying the trigonometric ratios sine, cosine, and tangent to solve problems

LESSON PLAN:

Atomic Force Microscopes

Objective/s- Write objective/s The student should be able to:	Assessment: What will you accept as evidence of student progress toward your lesson objective?
<p>PART 1</p> <ul style="list-style-type: none">• Identify the forces acting upon a cantilever.• Demonstrate and describe qualitatively how force causes deflection of a cantilever.• Describe qualitatively how a material's adhesive properties impact the force required to detach it from a surface.	<p>Part 1</p> <ul style="list-style-type: none">• Students will articulate how the lab setup models the real-life scenario of AFMs and the mechanical force/action required to free the cantilever. Students will explain how the movement of their arm exerts a force on the model. Students will also be able to qualitatively describe and order the force required to disturb various tape samples and relate these forces to the cantilever's deflection.• Formative Assessment: The teacher will formatively assess student understanding as they walk around and ask questions to students/groups as they are working through the activity or handout.
<p>PART 2</p> <ul style="list-style-type: none">• Identify and illustrate the forces acting upon a mass suspended from a cantilever by drawing and labeling a free body diagram for the system in equilibrium.• Graphically represent the relationship between deflection and mass added to the free end of a cantilever.• Mathematically model the behavior of a cantilever system with an equation and graph, developing Hooke's law and recognizing the spring constant.	<p>PART 2</p> <ul style="list-style-type: none">• Students will be able to quantitatively describe how a cantilever's deflection relates to the amount of force exerted with the use of Hooke's law.• Students will calculate the spring constant for the given cantilever system.• Students will algebraically manipulate (rearrange) the equation for the spring constant.• Students should be able to draw a free body diagram for the mass suspended from a cantilever.• Formative Assessment will be happening as the teachers walk around and ask questions to students/groups as they are working through the activity or handout.

LESSON PLAN:

Atomic Force Microscopes

Objective/s- Write objective/s The student should be able to:	Assessment: What will you accept as evidence of student progress toward your lesson objective?
<p>PART 3</p> <ul style="list-style-type: none">● Identify and illustrate the path and angle of the reflected incident light.● Graphically represent the relationship between the change in the angle of the incident light and the mass added to the free end of a cantilever.● Graphically represent the relationship between the change in the angle of the incident light and deflection when mass is added to the free end of a cantilever.	<p>PART 3</p> <ul style="list-style-type: none">● Students will effectively articulate how instrumentation with angle of reflection and angle of incidence can be used to map surfaces with AFMs.● Formative Assessment will be happening as the teachers walk around and ask questions to students/groups as they are working through activity or handout.
<p>PART 4</p> <ul style="list-style-type: none">● Identify and illustrate the path and angle of the reflected incident light.● Graphically represent the relationship between the change in the angle of the incident light and the mass added to the free end of a cantilever.● Graphically represent the relationship between the change in the angle of the incident light and deflection when mass is added to the free end of a cantilever.● Statistically analyze discrepancies between experimental values and theoretical values using a chi-squared test.● Draw a conclusion about the effectiveness of this experiment with statistical tests.	<p>PART 4</p> <ul style="list-style-type: none">● Students will be able to determine the effectiveness of this instrumentation through the use of statistical analysis (Pearson's Chi-Squared Test).● Students will calculate the p-value for Pearson's Chi-Squared Test through the use of excel or other computer software and draw a conclusion based on that finding.● Formative Assessment will be happening as the teachers walk around and ask questions to students/groups as they are working through activity or handout.

PART 4: Instrumentation Design: Statistical Analysis with Instrumentation

<p>ENGAGEMENT</p> <p>Estimated Time: 10 minutes</p> <p>Overview of Activity:</p>		
<p>Resources Needed: whiteboard & dry erase markers or poster paper & markers to record student comments</p> <p>Safety Considerations: None.</p>		
<p>What the teacher is doing:</p>	<p>What the student does:</p>	<p>Possible questions to ask students and anticipated student responses:</p>
<p>The teacher introduces nanoscience - what it is, why it’s important, and how AFMs play a role.</p> <p>The teacher will then discuss the main mechanics of an AFM and how it is used to quantify substances on the nanoscale. The main point will be to connect the use of optics in instrumentation as an introduction to the lab activity and make it relevant to how optics are used in the real world.</p> <p>Teacher shares ideas on nanoscale and nanotechnology.</p> <ul style="list-style-type: none"> • https://www.science.org.au/curious/nanoscience#:~:text=Nanoscience%20has%20the%20potential%20to,from%20manufacturing%20to%20health%20care (There is a helpful graphic on this page that helps students better visualize what a nanometer is.) <p>Note: students may struggle with visualizing a nanometer and may</p>	<p>Students share their responses.</p> <p>If desired, or if prompting is needed, students can do this think-pair-share style: students think about the question independently, discuss possible answers with a partner, and then share with the class.</p>	<p>Have you ever heard of a nanometer? What is it?</p> <ul style="list-style-type: none"> • <i>A ninth of a meter</i> (incorrect assumption) • $1 \times 10^{-9}m$ <p>Just how small do you think a nanometer is? What objects would be measured in nanometers?</p> <ul style="list-style-type: none"> • <i>They are too small to see.</i> • <i>Size of a molecule</i> <p>Why is it helpful to study substances on the nanoscale?</p> <ul style="list-style-type: none"> • <i>To understand how something works</i> • <i>Understanding something at the nanoscale could help us lead to breakthroughs in manufacturing or health care.</i> <p>Knowing what an AFM is capable of, in what ways can you think it could be used?</p>

LESSON PLAN:

Atomic Force Microscopes

not have a good grasp of scientific notation. This resource provides a good comparison:

<https://www.nano.gov/nanotech-101/what/nano-size#:~:text=A%20human%20hair%20is%20approximately,fingernail%20grows%20in%20on%20second>

"A nanoparticle to an ant, is the same ratio as an ant to a race track".

The teacher facilitates students' discussion and invites groups to share their answers.

- *characterizing bacteria and cells*
- *analyzing DNA molecules*
- *studying proteins in real-time*
- *imaging molecules down to sub-atomic resolution.*

PART 4: Instrumentation Design: Statistical Analysis with Instrumentation

Note: Part 4 of this series provides an alternate approach to exploring optic use within an AFM. While it can stand alone, it could also be used as an extension of Part 3 Instrument Design: Understanding How AFMs Use Optics & Reflected Light to Determine Sample Properties.

EXPLORATION

Estimated Time: 25 minutes

Overview of Activity: Students will explore angle of incidence and angle of reflection through instrumentation of a cantilever and laser system.

Resources Needed:

- 2 wooden meter sticks, 1 metric ruler, 1" x 1" mirror, 2 C-clamps, laser pointer (red or green), ring-stand with 2 clamps (to hold the laser pointer and one of the meter sticks), weights (ranging in mass: 0 g, 50g, 100 g, 150g, 200 g, 250g, 300 g), weight holder or string, two pieces of printer paper, Tape, colored pencils & highlighters, Protractor, laptop or calculator (to do the χ^2 test and look up the χ^2 table for p-values).
- Per student: "AFM Activity 3 Lab Sheet – Incident Light" handout, writing utensils
- Note: You may also want to test the equipment prior to completing the activity with students to ensure you are not asking students to add more mass to the system than the meter stick can hold.

Safety Considerations:

- Mirrors are generally made of glass, so handle them carefully.
- Do not look directly into the laser; the light can damage your eye.
- Do not aim the laser at anyone's face.
- Keep the laser turned off when not taking measurements.
- Be mindful of your surroundings when using a meter stick to avoid collisions.
- Avoid putting excessive force on the meter stick and breaking it. If the meter stick breaks, be careful of the sharp end and dispose of it immediately.
- Be cautious when putting weights on the meter stick. Heavy weights can cause injury when dropped. Avoid standing or putting your hands in locations in which weights could fall.
- Be cautious when using the clamps so as not to pinch fingers.

LESSON PLAN:

Atomic Force Microscopes

What the teacher is doing:	What the student does:	Possible questions to ask students and anticipated student response:
<p>Throughout the activity, the teacher takes the role of facilitator and "lead-learner."</p> <p>Pass out materials to each student group or have a predetermined team equipment manager pick them up from a designated location.</p> <p>Pass out the "AFM Activity 4 Lab Sheet – Statistical Analysis of Incident Light" handout to each student.</p> <p>After independently reading through the handout, have students discuss the activity with their group and develop a short list of what they know and wonder. Have them think through their questions and try to reason through them before sharing them with you. Doing so will identify student groups uncertain of the directions and promote cooperation skills while encouraging autonomy.</p> <p>Note: It may be necessary to model how to set up the experiment.</p> <p>As students work in their groups,</p> <ul style="list-style-type: none"> • Make sure each group has set up the experiment correctly. • Ensure each student is participating. • Rotate between groups and ask guiding questions to 	<p>In student groups of 3-4:</p> <p>Students read through the "Procedure" section of the "AFM Activity 4 Lab Sheet – Statistical Analysis of Incident Light" handout.</p> <p>Students will discuss the activity and observation questions with their group, recording what they know and wonder about and discussing any potential questions or concerns.</p> <p>Students will follow the directions on the handout to set up the experiment before proceeding to conducting the experiment and recording their findings.</p> <p>Once completed, students will disassemble the experiment and return the materials to the proper location.</p>	<p>What are some of your controlled variables?</p> <ul style="list-style-type: none"> • <i>Position of the laser (angle of the laser relative to the mirror)</i> • <i>Placement of mirror, masses, etc.</i> <p>How does the horizontal displacement look? Does the trend match what we did last time?</p> <ul style="list-style-type: none"> • <i>The horizontal displacement looks equally spaced out (linear). The trend matches the results from the last experiment.</i> <p>Should the vertical deflection match the same trend? Why or why not?</p> <ul style="list-style-type: none"> • <i>The vertical deflection should match the linear trend from the last lesson since it follows Hooke's law.</i> <p>Would this setup work if the laser was angled? Why or why not?</p> <ul style="list-style-type: none"> • <i>The setup would work if the laser was angled, but you would need to measure the angle of the incident ray relative to the normal line of the mirror's center each time.</i>

LESSON PLAN:

Atomic Force Microscopes

<p>individual students within each group:</p> <ul style="list-style-type: none">○ Goal: provide students time to process the different aspects of the system and how one may impact the other and assess student understanding as they work through the activity.○ Use open-ended and guiding questions to encourage critical thinking while subsequently holding students accountable for their learning.		<p>How do the angles you are measuring differ between masses?</p> <ul style="list-style-type: none">● <i>Theoretically, the angle will get larger, but due to the instrumentation, the change in angle will not be very big.</i> <p>Does the vertical deflection you calculated with the measured angles make sense?</p> <ul style="list-style-type: none">● <i>The vertical deflections do not make sense. They are not following the linear trend.</i> <p>How does your p-value compare to the significance level, and what does this mean?</p> <ul style="list-style-type: none">● <i>The p-value is less than the significance level; therefore, we can reject the null hypothesis.</i>● <i>The p-value is greater than the significance level; therefore, we cannot reject the null hypothesis.</i>● Note: it is important to mention that not rejecting the null hypothesis does not imply that the null hypothesis is true.
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LESSON PLAN:

Atomic Force Microscopes

PART 4: Instrumentation Design: Statistical Analysis with Instrumentation

EXPLANATION

Estimated Time: 15 minutes

Overview of Activity: Students will mathematically model the behavior of a cantilever system with an equation and graph. Students will also statistically analyze the significance of the instrumentation with Pearson's Chi-Squared Test.

Resources Needed: whiteboard & dry erase markers or poster paper & markers to record student comments, document camera.

Safety Considerations: None.

What the teacher is doing:	What the student does:	Possible questions to ask students and anticipated student response:
<p>The teacher guides a whole-group discussion over the students' experiences during the activity and their observations.</p> <p>The teacher asks students to summarize the activity and their observations. The teacher acts as the facilitator and provides prompting, guiding questions, and open-ended questions.</p> <p>The teacher records student comments on the whiteboard or poster paper.</p>	<p>Students follow the teacher's lead in a whole group discussion.</p> <p>Students discuss:</p> <p>Another option would be to have each group create a poster summarizing the experiment and the group's observations. Student groups would present their posters and answer guiding questions provided by the teacher.</p>	<p>Why did our angles not match the expected values?</p> <ul style="list-style-type: none"> • <i>Random error: each will be slightly off since we did each measurement by hand.</i> • <i>Systematic error: There could be an offset with the way the string is placed.</i> <p>What was the conclusion from the statistical analysis?</p> <ul style="list-style-type: none"> • <i>Failed to reject the null hypothesis.</i> • <i>Rejected the null hypothesis.</i> <p>How could using statistical tests help determine the effectiveness of an experiment?</p> <ul style="list-style-type: none"> • <i>We can determine if there is statistical significance.</i>

LESSON PLAN:

Atomic Force Microscopes

PART 4: Instrumentation Design: Statistical Analysis with Instrumentation

ELABORATION

Estimated Time: 15 minutes

Overview of Activity: Provides students an opportunity to apply or extend the new ideas and information on the instrumentation of a cantilever system through statistical analysis.

Resources Needed: Dependent on activity.

Safety Considerations: Dependent on activity.

What the teacher is doing:	What the student does:	Possible questions to ask students and anticipated student response:
<p>The teacher facilitates the discussion by providing guiding questions geared to help students apply or expand upon the day's lessons over instrumentation with angle of incidence and angle of reflection.</p>	<p>Students take part in active discussion and subsequent activity.</p> <p>If desired, or if prompting is needed, students can do this as a group.</p>	<p>Why should we consider the effectiveness of instrumentation?</p> <ul style="list-style-type: none"><i>To ensure the data collection represents the experiment accurately.</i> <p>Are there other ways to evaluate if this is a good model?</p> <ul style="list-style-type: none"><i>We can use a t-test, look at linear regressions and evaluate the R^2 values.</i> <p>What are some ethical problems that might arise with statistical analysis?</p> <ul style="list-style-type: none"><i>People can change the conclusion by changing the significance level.</i><i>People can also alter the p-value by the way they analyze their data.</i>

LESSON PLAN:

Atomic Force Microscopes

PART 4: Instrumentation Design: Statistical Analysis with Instrumentation

EVALUATION

Estimated Time: Embedded throughout the lesson. Account for time required for pre-assessments and post-assessments (if necessary)

Overview of Activity: Formative and summative assessment of student understanding and effectiveness of the lesson.

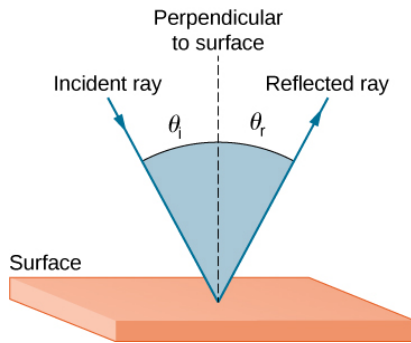
Resources Needed: varies

Safety Considerations: None.

What the teacher is doing:	What the student does:	Possible questions to ask students and anticipated student response:
<p>Throughout the lesson and activity, the teacher uses formative assessments to determine how much the students understand and if they are grasping the new concepts, as well as to evaluate the quality of the lesson.</p> <p>Formative assessment occurs during the learning process, provides an opportunity to identify the need for flexibility in instruction, identifies areas for improvement (both for student and for the lesson), and allows students a chance to implement feedback. Examples of formative assessment include hand signals, direct or indirect questioning, quizzes, observations, homework, and classwork.</p> <p>Summative assessment is completed at the end of the learning process and provides an evaluation of student concept knowledge. Summative assessment will often include evaluating the student work products, such as responses on the guided notes, handouts, posters, teacher-created exit slips, or unit tests.</p>	<p>Throughout the lesson, students work in groups to answer questions regarding the experiment and observations.</p> <p>Students will also answer questions independently when prompted by the teacher during the lesson phases or through teacher-prepared pre-assessments, post-assessments, or exit slips.</p>	<p><i>See questions embedded in the sections above. Also, see accompanying Part 3 handout.</i></p>

Atomic Force Microscopes (AFMs)

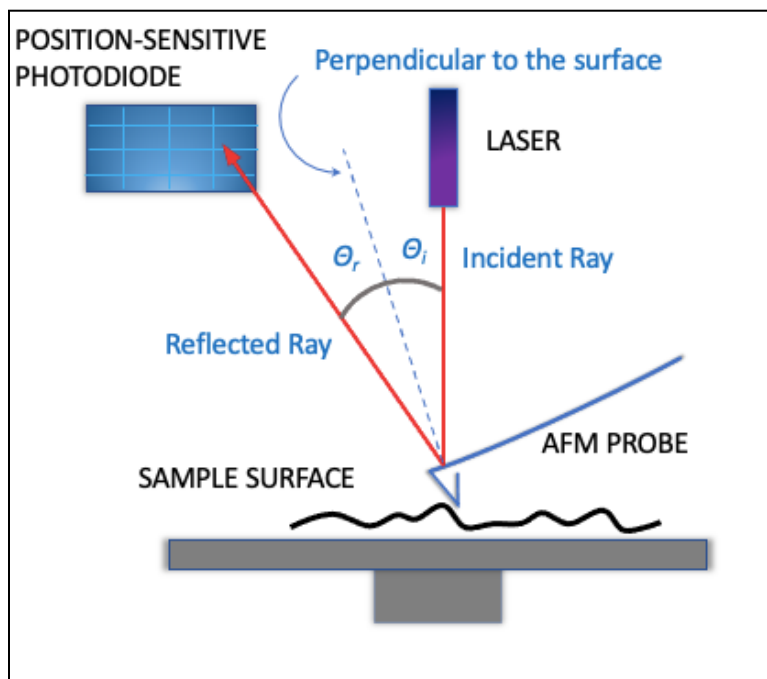
PART 4: Statistical Analysis: Understanding How AFMs Use Optics & Reflected Light to Determine Sample Properties



[Image Source](#)

Introduction

Atomic force microscopes (AFM) allow us to study samples at the nanoscale to measure and localize many forces, including adhesion, magnetic forces, and mechanical properties, to create a 3D surface profile of the sample. To do this, the AFM uses a cantilever system with a very sharp tip to traverse the surface of a material. Laser light is directed at a small mirror at the end of the cantilever. The **incident ray** then reflects onto a **position-sensitive photodiode (PSPD)**. As the cantilever deflects from forces exerted on the tip, slight changes occur in the **reflected ray**. The PSPD registers any fluctuations in the **angle of incidence θ_i** and **angle of reflection θ_r** . Thus, tracking the changes. As the AFM passes over a raised surface, the PSPD records the cantilever deflection and the resulting change in the angle of incidence.



Figures 1: Diagram of the optics system of an AFM.

From the equations derived in the last lesson, we can calculate (as a function of added mass) the expected values of horizontal deflection, vertical deflection, or angle of incidence. Then we can use statistical analysis to determine the effectiveness and significance of our instrumentation for mapping nanometer-sized surfaces.

In this lab, you will learn how to:

- identify and illustrate the path and angle of the reflected incident light.
- graphically represent the relationship between the change in the angle of the incident light and the mass added to the free end of a cantilever.
- graphically represent the relationship between the change in the angle of the incident light and deflection when mass is added to the free end of a cantilever.
- statistically analyze discrepancies between experimental values and theoretical values using a chi-squared test.
- draw a conclusion about the effectiveness of this experiment with statistical tests.

Activity: Calculating the Relationship Between Deflection & Applied Force in Cantilevers

Safety Precautions:

- Mirrors are generally made of glass, so handle them carefully.
- Do not look directly into the laser; the light can damage your eye.
- Do not aim the laser at anyone's face.
- Keep the laser turned off when not taking measurements.
- Be mindful of your surroundings when using a meter stick to avoid collisions.
- Avoid putting excessive force on the meter stick and breaking it. If the meter stick breaks, be careful of the sharp end and dispose of it immediately.
- Be cautious when putting masses on the meter stick. Heavy masses can cause injury when dropped. Avoid standing or putting your hands in locations in which masses could fall.
- Be cautious when using the clamps so as not to pinch fingers.

For this activity, you will need the following:

- 2 wooden meter sticks
- 1 metric ruler
- 1" x 1" mirror
- 2 C-clamps
- laser pointer (red or green)
- ring-stand with 2 clamps (to hold the laser pointer and one of the meter sticks).
- weights (ranging in mass: 0 g, 50g, 100 g, 150g, 200 g, 250g, 300 g)
- weight holder or string
- two pieces of printer paper

4. What are the intended takeaways from this activity?

Procedure

Step 1: Experimental Setup

Cantilever: Using the C-clamps, attach the meter stick horizontally to a table or desk surface with 70 cm of the meter stick hanging over the edge of the surface, as shown in Figure 2. Attach the mirror to the free end of the meter stick with 5 cm of free space at the end. Tie a length of string to the meter stick's free end. You can secure the string by wrapping it several times around the end of the meter stick and tapping it. Tape a piece of string of length 1 m to the top of the free end. Make sure the string aligns with the point where the laser hits the mirror.

Laser: Attach the laser pointer to the ring stand with a clamp, so the ray points vertically upward. *Use caution not to shine the laser at anyone or to look at it.* Place the ring stand directly under the mirror. Tape two pieces of paper together on the floor with the edge of one piece under the ring stand. You will record the position of the reflected beam on the paper as the lab progresses.

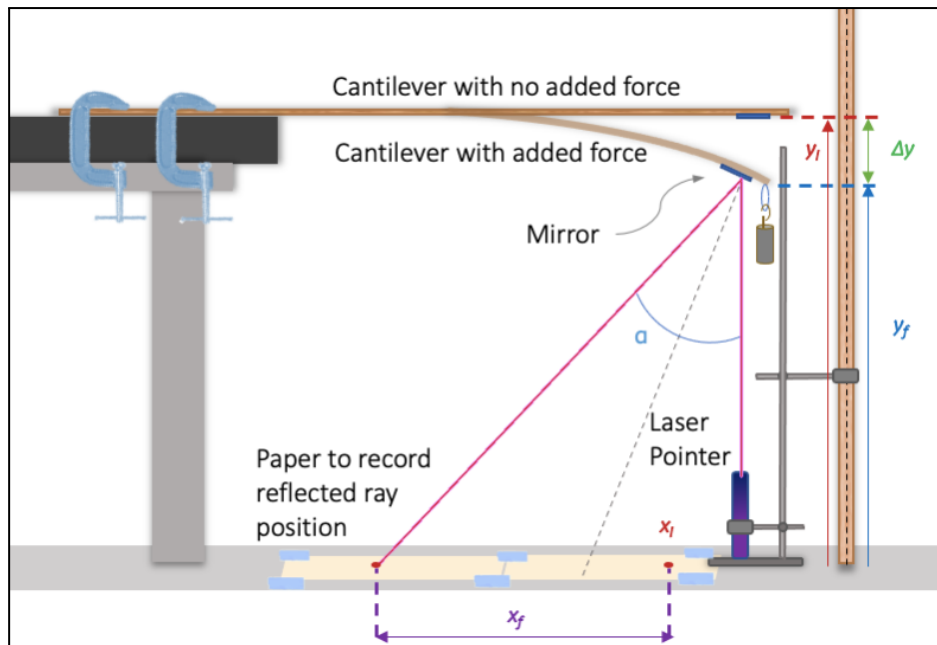


Figure 2: Experimental setup

Step 2: Take initial measurements

Cantilever height: Attach the second meter stick to the ring stand (perpendicular to the floor with the 0 cm at the floor.) Place the ring stand near the free end of the cantilever. Measure the height of the free end of the meter stick in centimeters to the closest millimeter. Record the initial height value in Table 1 as x_i (corresponding to 0 g of added mass).

Reflected light: With the laser pointer turned on, carefully mark where the reflected beam hits the paper. Label this point y_i . We will measure the distance the reflected beam hits the floor from this position as mass is added to the cantilever.

Step 3: Record the location of reflected light

Mark the location of the reflected ray on the paper. Measure the horizontal distance of the new reflected ray from the initial position x_i to the nearest millimeter and record it in centimeters in Table 1.

Step 4: Exert force on the cantilever, record new height & angle

Attach the 50 g mass to the string. Place the center of the protractor on the point y_i . Using the string, measure the angle α (where $\alpha = \Theta_i + \Theta_r$). Record the mass (in grams) and corresponding angle as α in Table 1. Measure the height of the free end of the meter stick, measuring upward from the floor. Record the mass (in grams) in Table 1.

Step 5: Repeat Steps 3 and 4 using 100g, 150g, 200g, 250g, and 300g.

Data, Observations, & Analysis

1. Estimate how much the cantilever deflected with no added mass. How about 100g of added mass?
2. As you work through the lab, for each trial:
 - a. Record the mass m and the resulting height of the cantilever x_i in Table 1. Calculate the deflection Δy ($\Delta y = y_i - y_f$).
 - b. Record the horizontal distance of the reflected ray relative to the initial position x_i .
 - c. Calculate the ratio of the horizontal to the vertical position using x_i/y_i .

3. Using your string and protractor, measure angle α in degrees and record it in Table 1.

Table 1: Data Collection & Analysis

Mass of Attached Weight, m (g)	Horizontal Position of the Reflected Ray, x_f (cm)*	Vertical Height, y_f (cm)	Angle of Incidence, θ_i (degrees)
0	$x_i = 0$	$y_i =$	90°
50			
100			
150			
200			
250			
300			

*For $m = 0$ g, $x_i = x_f$ and $\theta_i = 90^\circ$. All other values of x_f are measured as the distance from x_i and θ_i is measured each time.

4. Calculate the new height for each added mass using trig functions. Then find the deflection value by subtracting y_f from the initial height. Record your values in the following table:

Mass of Attached Weight, m (g)	Angle of Incidence, θ_i (degrees)	Vertical Height Δy (cm)	Deflection $\Delta y = y_i - y_f$ (cm)
0			
50			
100			
150			
200			
250			
300			

5. Do the vertical deflection values make intuitive sense? Explain your reasoning.

6. On the provided graph (Figure 3), label the horizontal axis as "Force (N)" and the vertical axis as "Vertical Deflection (cm)." Plot the angle of incidence as a function of added mass.

Relationship Between Force and Vertical Deflection

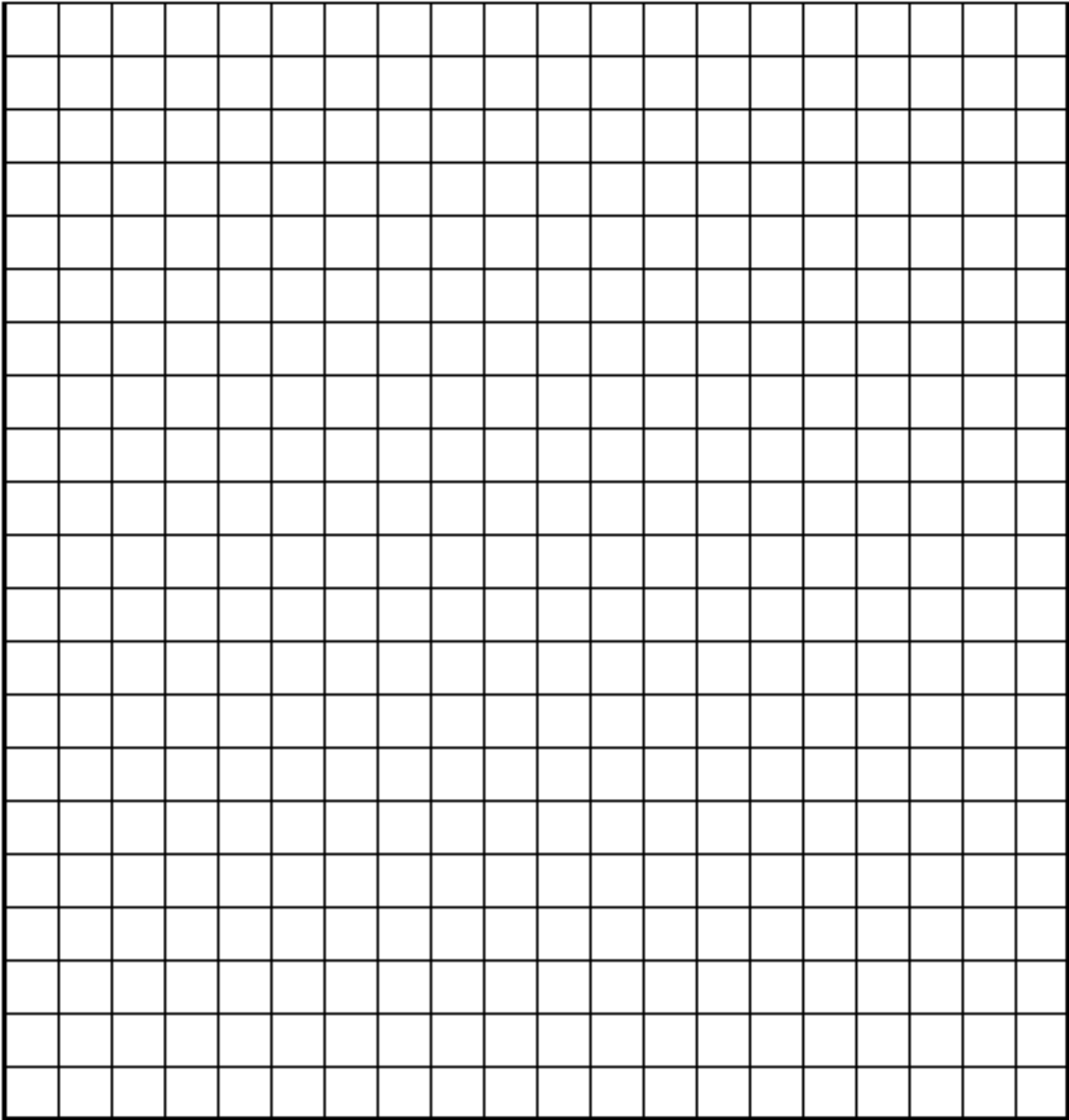


Figure 3. Force versus Vertical Deflection.

7. On the same graph (Figure 3), use a different color to plot the expected vertical deflections for a given force. How do the expected values compare with the observed values? Do your numbers make sense? Why or why not?

8. Using your trig equation from the previous lesson, find the expected angles and record them in the following table with the corresponding mass.

Mass of Attached Weight, m (g)	Expected Angle of Incidence, θ_{Ei} (degrees)
0	
50	
100	
150	
200	
250	
300	

9. Compare your expected angles with your measured angles. Do they seem similar, or are they different? If they were different, how could this impact the calculations for vertical deflection?

Statistical Analysis

We can use Person's Chi-Squared Test to determine if there is a statistically significant difference between the expected and observed values in one or more categories of a contingency table. To perform Person's Chi-Squared Test, we need the degrees of freedom. The degree of freedom is the least number of values you can know to solve for the Chi-Squared value. In this case, degrees of freedom, denoted as df , is equal to the sample space (denoted as n) minus one.

$$df = n - 1$$

A contingency table is a matrix that displays the multivariate frequency distribution of the variables. The following formula determines Person's Chi-Squared value:

$$\chi^2 = \sum_{i=1}^n \frac{(O_i - E_i)^2}{E_i}$$

10. In the following consistency tables, copy your expected and observed values.

Mass of Attached Weight, m (g)	Angle of Incidence, θ_i (degrees)	Expected Angle of Incidence, θ_{Ei} (degrees)
0		
50		
100		
150		
200		
250		
300		

11. Write your χ^2 value and degrees of freedom:

a. $\chi^2 =$

b. $df =$

Assessment

1. Using a Chi-Squared Table or Chi-Squared calculator, state your p-value and significance level. What does your p-value tell you?
2. State your conclusion based on the results from the Chi-Squared Test.
3. According to your conclusion, would you recommend this instrumentation method for finding vertical deflection? Why or why not?
4. What changes could you make to better the experimental setup? What statistical tests could you run to make sure your experimental setup is an improvement?