SOLIDWORKS DESIGN OF A TIP-ENHANCED NEAR-FIELD OPTICAL MICROSCOPE

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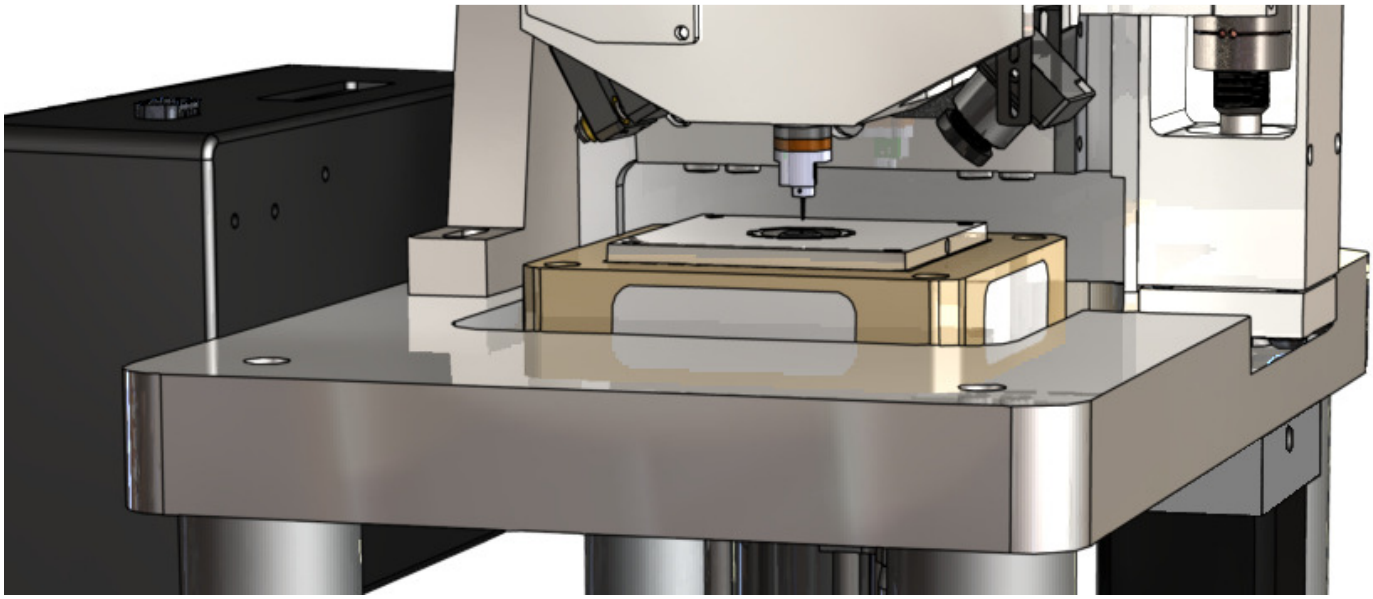
Overview

Optical microscopy is favored by many scientists due to its simplicity and versatility. Nearly any sample may be imaged using a wide variety of optical techniques. The resolution, however, is fundamentally limited by the diffraction of light. When imaging with visible light, this restricts imaging to features larger than about 200 nm; when contrasted with more complicated techniques such as electron microscopy and scanning probe microscopy, this resolution limit represents a significant drawback to optical microscopy. However, the versatility and non-destructive nature of optical imaging still make it a viable option for many applications.

Fortunately, the diffraction limit is not an absolute barrier, and several techniques have been developed to circumvent this limit. One such technique is known as tip-enhanced near-field optical microscopy (TENOM), in which the imaging light is confined and enhanced by the presence of a metal tip near the sample.

In the process of creating a TENOM system, the mechanical requirements necessary for both optical and atomic force imaging became too complex to visualize mentally, and the determination was made that the design phase should occur virtually in a 3D CAD environment. SolidWorks was selected due to its ease of use, intuitive drafting features, support of complex assemblies, exportation of files to CNC machines, and the availability of SolidWorks parts files from commercial vendors. This greatly simplified the design process, resulting in a versatile instrument in which compatibility and alignment of the various components was guaranteed.



SOLIDWORKS

1. Beating the diffraction limit of optical microscopy

Optical microscopy is fundamentally limited by diffraction. When a lens is used to focus light, the size of the focused spot corresponds to the maximum obtainable resolution. Its value is given by[1]

$$d = \frac{\lambda}{2(n\sin\alpha)},$$

where λ is the wavelength of the light, n is the refractive index (a measure of the effect of the lens material on the speed of light), and α is the half-angle of the cone of light that propagates through the lens.

The diffraction of light limits resolution in optical microscopy to roughly λ/2.

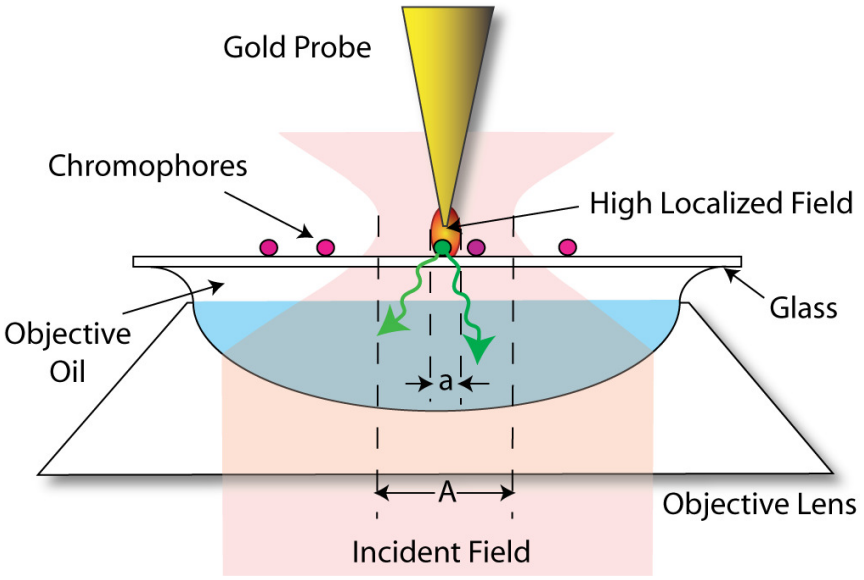
The quantity (n sin α) is known as the numerical aperture and generally serves as a good indicator of the resolving power of a lens. Typical values of numerical aperture for objective lenses range from 0.1 to 1.4, corresponding to a maximum resolution of ≈λ/2, or roughly 250 nm, in optical microscopy.

In 1928, a technique was proposed[2] to beat the diffraction limit by point scanning through a sufficiently small aperture such that light would not propagate. Within one wavelength of the source, there exists an evanescent (that is, stagnant) field, the so-called "near-field", which is non-propagating and thus not subject to diffraction.

This technique remained experimentally impossible until the invention of atomic force microscopy (AFM)[3]. Replacement of the AFM probe tip with a tapered optical fiber[4,5] resulted in a resolution of 25 nm, or ≈λ/20. This technique is known as near-field scanning optical microscopy (NSOM).

In 1985, a technique was proposed[6] to further improve resolution while simultaneously eliminating the need for an aperture probe. By illuminating a sharp metallic tip with an external laser, an enhanced field is generated at the end of the probe. As the tip scans the sample surface, the enhanced field generates a spectroscopic response from the sample. This technique is known as tip-enhanced near-field optical microscopy (TENOM) and has shown spatial resolution on the order of 20 nm[7].

Resolution in near-field techniques is limited only by the geometry of the probe.



Conceptual drawing of TENOM probe. "A" is the diffraction limited illumination source and "a" is the effective imaging probe[8].

2. Motivation

This design was originally conceived as a replacement for a heavily modified commercial microscope being converted into a TENOM system. As the project continued to progress and further modification became necessary, it grew increasingly apparent that adaptation of an existing system would not afford the necessary flexibility. It was therefore decided to design a system from the ground up with an emphasis on stability, simplicity, and versatility.

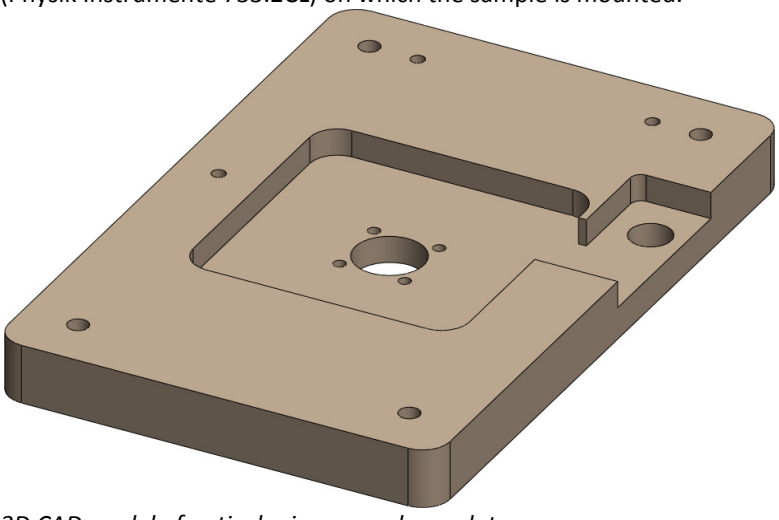
It soon became apparent that, despite the intended simplicity, it would be nearly impossible to mentally visualize each of the various components of such a complex instrument and have any confidence in their placement, compatibility, or alignment. As an alternative, it was decided that the microscope should be constructed in a virtual environment using SolidWorks. Each individual component was drafted and then brought together into one comprehensive assembly, providing a fast and free design phase in which all of the parts were guaranteed to fit together exactly as they were intended to.

The design phase was undertaken entirely in a virtual 3D CAD environment using SolidWorks.

3. Design of optical microscope

The critical components of an optical microscope are the illumination source, the condenser lens which focuses the illumination light, the objective lens which magnifies the image, and the detector. In near-field microscopy, the most practical arrangement is an inverted microscope with epi-illumination, allowing the objective lens to double as the condenser. The physical design of the optical microscope provides the optical pathways necessary for excitation and collection in this system, emulating the function of a commercial system at a fraction of the price.

The CAD phase was greatly simplified due to the availability of SolidWorks parts files from multiple vendors. These files were downloaded and mated with custom parts. The first step was construction of a stable base. Four 1.5" diameter posts (Thorlabs P8) were mounted to an optical breadboard (Thorlabs MB1824) to support the custom designed base plate. The base plate supports the scan bed, a two axis nanopositioning stage (Physik Instrumente 733.2CL) on which the sample is mounted.

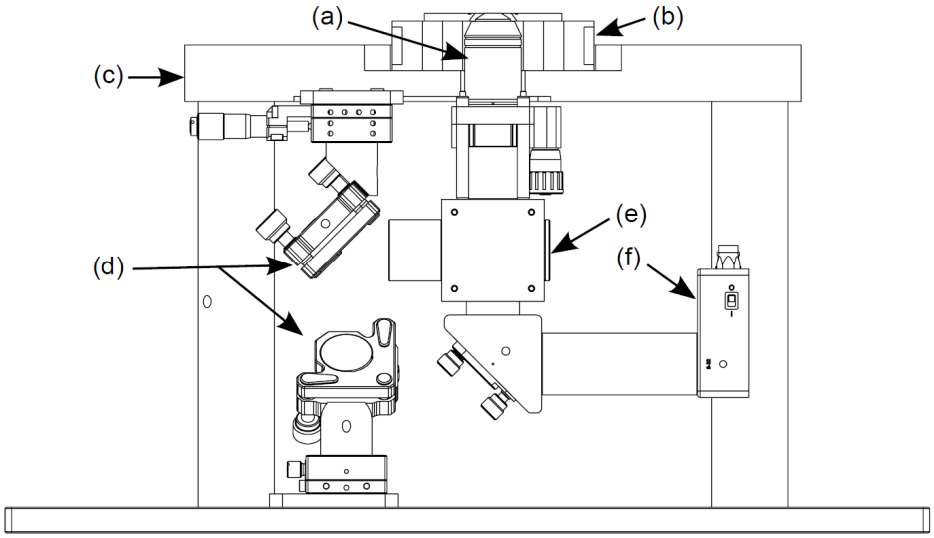


3D CAD model of optical microscope base plate

The part file for the base plate was converted to .dxf format for direct upload into a CNC mill.

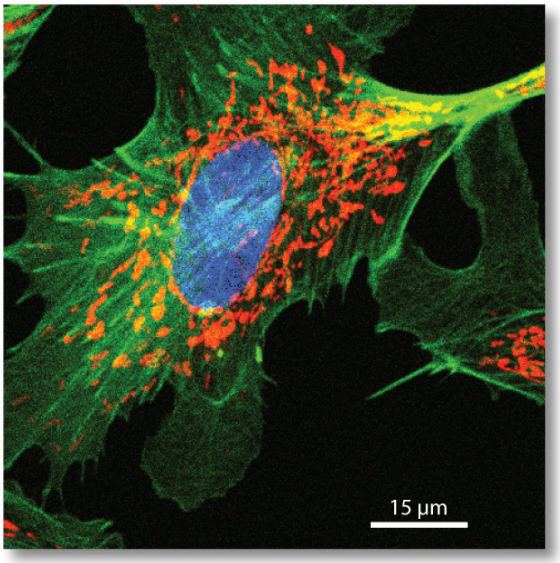
The illumination source, an external laser, is brought into a cube-mounted beam splitter (Thorlabs CM1-BS013) by a periscope consisting of two kinematic mirror mounts (Thorlabs KM100) attached to single axis translation stages (Melles Griot 148-103) mounted to the optical breadboard and the bottom of the base plate.

The beam splitter reflects the light to the objective lens (Olympus 1.4 NA, 60x), which is mounted in a z translation stage (Thorlabs SM1Z). This translator controls the distance between the objective and the sample, providing focusing capabilities. The light is reflected back down through the objective and beam splitter to a 90° kinematic mirror mount (Thorlabs KCB1), which redirects the light to the detector (Thorlabs APD110A).



Two-dimensional cross section view of optical pathways in inverted optical microscope design. a) Objective lens b) X-Y scanner c) Base plate d) Periscope assembly e) Beam splitter f) Avalanche photodiode

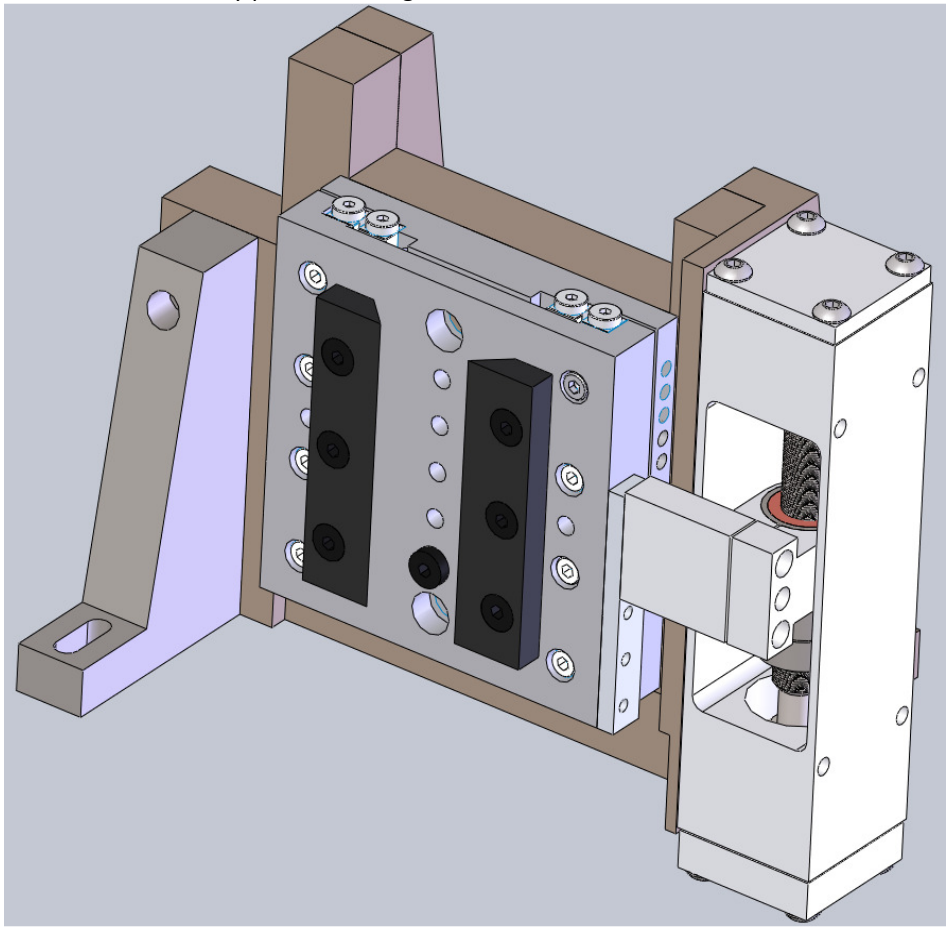
The implementation of point-scanning capabilities allows imaging of a much larger area at increased resolution by measuring the light intensity at each pixel. The algorithms necessary for scanning and collection are written in LabVIEW and controlled through a National Instruments data acquisition (DAQ) card (PCIe-7852R) with a field programmable gate array (FPGA) microprocessor.



Fluorescence image of bovine pulmonary artery endothelial (BPAE) cells[8]

4. Atomic Force Microscopy

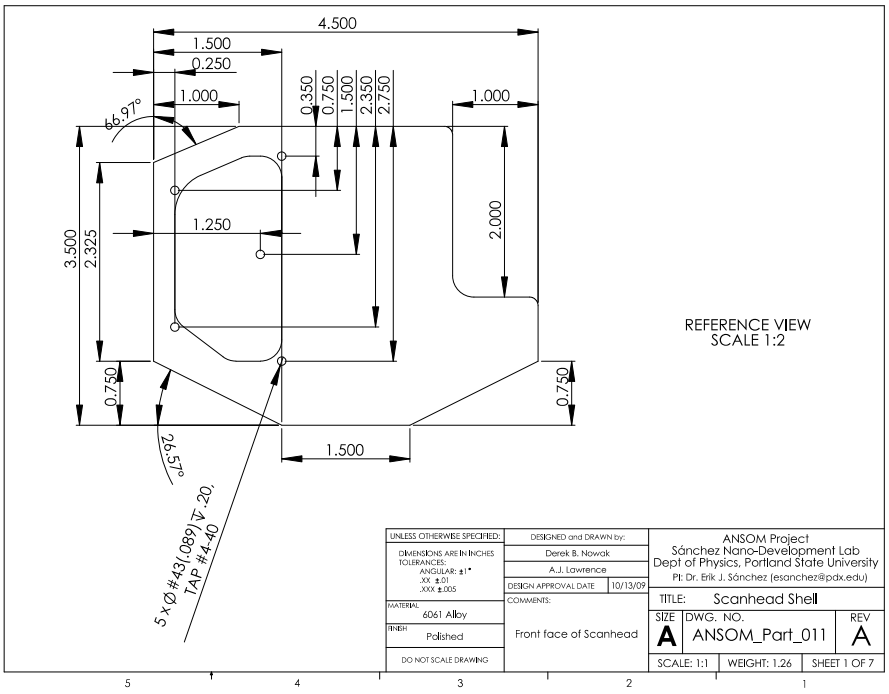
The optical microscope was designed with forethought given to the necessity of integration of AFM capabilities. The custom base plate, which serves as the ceiling and sample holder of the optical microscope, functions as the base of the AFM, leaving the two systems integrated yet independent. A pocket milled into the base plate accommodates the x-y piezo scan stage.



3D CAD model of AFM backplane

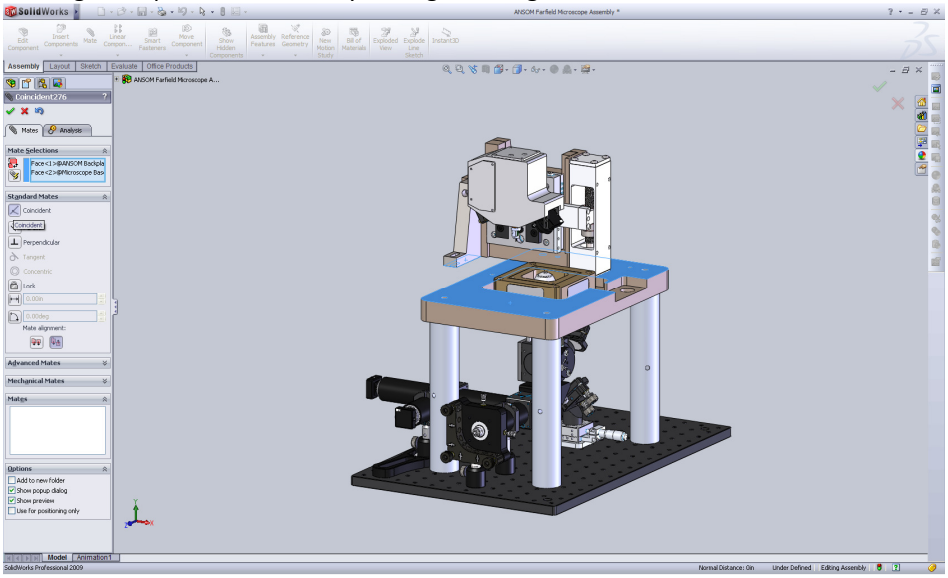
Mounted on the base plate is a stable backplane assembly, consisting of a thick plate and three braces, all custom designed in SolidWorks and machined from cast iron. The sole function of the backplane assembly is to position the AFM scan head such that the tip is suspended over the sample surface. Mounted to the back of the scan head is a dovetail (Thorlabs XT66D3) which nests in custom rails mounted to a 50 mm translation stage (Thorlabs LNR50M with drive removed). The translation stage is mounted to the backplane, and the z height is controlled by a stepper motor (Oriental Motors PK243B1A-SG36) and lead screw (Universal Thread) assembly.

In some cases, parts had to be reverse engineered from two-dimensional drawings provided by the manufacturer. This was the case with the lead screw.



Engineering drawing of scan head shell

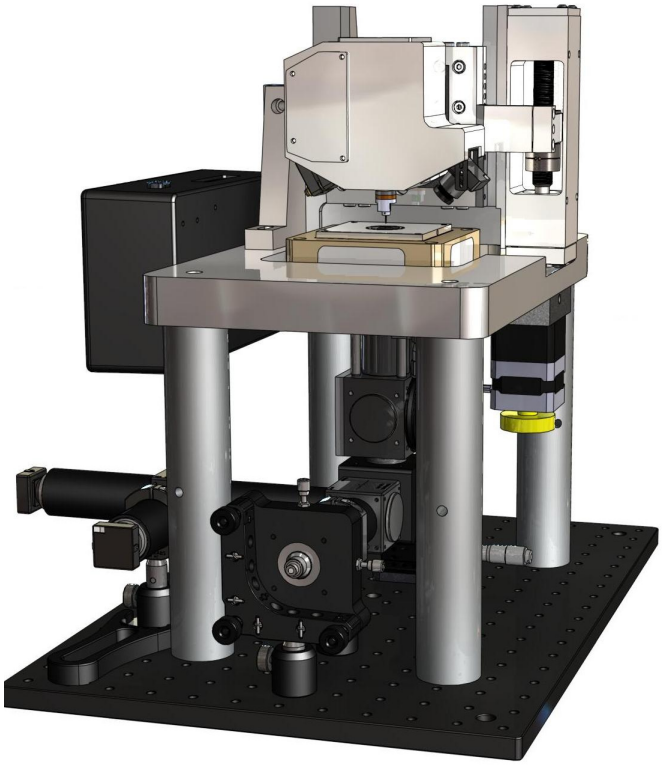
The primary design complication of the backplane was the three-dimensional positioning of the AFM tip. The x and y dimensions were constrained by the necessity for the tip, and therefore the center of the scan head, to be perfectly centered over the objective lens, thereby making simultaneous optical and AFM imaging possible. The z dimension was less critical due to the inherent flexibility afforded by the 50 mm translation stage, but in general, it was desirable that the tip be near the surface when the stage was positioned midway through its range of travel.



Mates between subassemblies ensured compatibility and alignment of components.

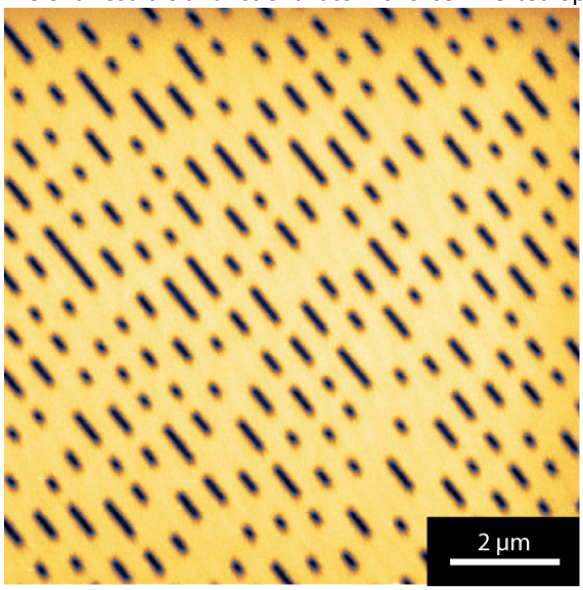
The realization of these requirements was greatly simplified by the CAD design process undertaken in SolidWorks. Separate subassemblies were constructed for the scan head, backplane, and optical microscope. By making the necessary mates within the assembly, such as constraining the dovetail to the dovetail rails, the bottom of the backplane to the top surface of the base plate, etc., all but three degrees of freedom were

eliminated---the x and y positions of the backplane and the z position of the scan head. The subassemblies were then moved to appropriate locations, revealing the optimal positions for the mounting holes. For added flexibility, slots were machined in the backplane braces, allowing 1/4" of travel in the y position of the backplane.



3D CAD model of inverted optical microscope with AFM capabilities

The custom parts were outlined in SolidWorks drawing files and fabricated from cast iron, aluminum, and MACOR. Once assembled, the microscope was placed in a light proof enclosure on a floating optical table to shield it from external fields and vibration. The end result is a functional atomic force inverted optical microscope.

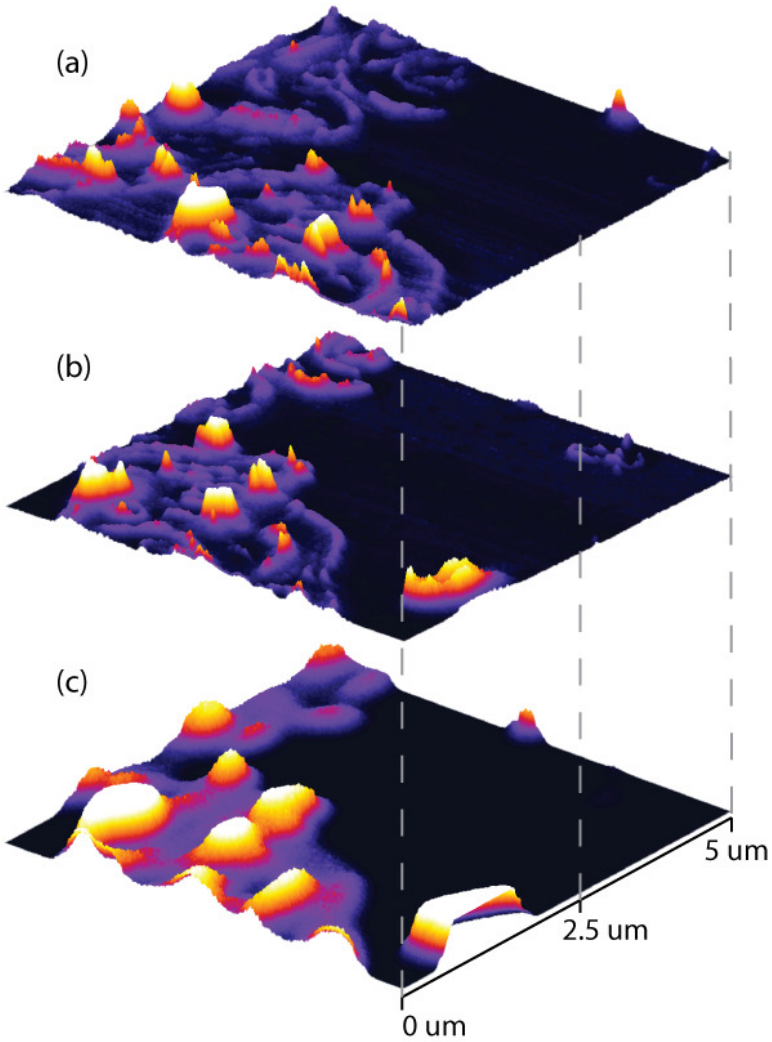


Topographic AFM image of pits on the surface of a DVD[8]

The final design is a highly adaptable, low-cost instrument with potential capabilities beyond those of any commercially available system.

5. Tip-Enhanced Near-Field Optical Microscopy

With the optical and AFM systems functional, integration of a specialized probe tip, created via electrochemical etching and focused ion beam milling, added near-field imaging capabilities to the system[9,10].



Three images of same region of interest using different optical systems. a) Near field two-photon fluorescence image obtained with low cost near-IR diode laser. b) Near field two-photon fluorescence image obtained with a much more expensive pulsed laser system c) Diffraction-limited two-photon fluorescence image.

SolidWorks proved to be an invaluable tool in the development of this instrument. Without the support for complex assemblies and the intuitive mating capabilities the software affords, the design phase of this project would have proven unnecessarily complicated.

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This project is being disseminated an as open system design. For more information and relevant documentation, please visit the ANSOM Project webpage at ansom.research.pdx.edu.

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