Three Dimensional Microscopy - Imaging 3D Microstructures by Freeform Microoptics

A traditional microscope looks at a sample from one specific direction and acquires a two-dimensional (2D) projection of the sample in that direction. Although the information collected by a conventional microscope is very helpful in understanding the microstructures of the sample, under many circumstances 2D information alone is not enough or even confusing. In recent decades there is a trend to develop three-dimensional (3D) microscopes in order to obtain 3D microstructures. To this end, several 3D microscope designs have been studied and many of the new 3D microscopes are based on Z-stack scanning.

3D Microscope Designs

In a microscope based on Z-stack scanning, the observer is still looking in one direction. By utilizing a one-dimensional (1D) scanner, usually a piezoelectric scanner, the focal plane of the microscope is continuously adjusted and a series of 2D slices are obtained. The 3D model of the sample is then reconstructed by combining the 2D slices. The issues of this method are: 1) mainly limited to transparent samples such as cells; 2) the scanning ranges are limited by the scanner and thus are mainly used for very small samples; 3) the scanners are expensive; 4) for better resolution, more expensive setup or software are required, such as a confocal setup or a deconvolution program. This method is costly and not suitable for non-transparent, large-size samples.

Another method for 3D microscopy is based on modifying the viewing direction such as the commercial products listed in references [1, 2]. However, the devices needed for changing viewing direction increase the complexity of the system and therefore can result in alignment errors when the individual views are combined. Moreover, a new set of microscope is required therefore can be costly. One of the main goals of the current research is to develop a new method as well as a new device for low cost 3D microscopy. The new device needs to be integrated with conventional microscopes with minimum additional setup.

Freeform Microoptics
Figure 1 shows the principle of the 3D microscope based on freeform microoptics.

This 3D microscope demo is a combination of a regular machine vision microscope (VZMTM 450i, Edmund Optics, or any similar microscope) and a freeform prism array that is fabricated in the Precision Engineering Research Laboratory at The Ohio State University.

The idea of this 3D microscope is to view the objects from several directions simultaneously by using a freeform prism array. The freeform prism array in this design has nine facets, i.e., one in the center and eight at the outer side, thus can cover the 360º space in the X-Y plane. The eight facets were tilted at approximately 37º and thus resulting in an approximately 20º tilt in the optical axis. The viewing angle of the device in the X-Z plane is 0º and ±20º.

To compensate the aberration caused by the prism, a freeform surface design was used. By using the freeform design, the light rays from the object that are refracted by the prism can be traced back to a virtual object point. The virtual object points formed by the nine prisms (including the prism in the center) were designed to be on the same plane normal to the Z axis and thus nine clear images can be obtained simultaneously without further focal length adjustments (only three virtual object points V1, V2, and V3 were shown in fig. 1). The freeform prism array was fabricated (fig. 2) using a high precision freeform optical fabrication method, i.e., the ultraprecision diamond machining and broaching process (350 FG ultraprecision machine by Moore Nanotechnology System, Keen NH). The details of the design and the fabrication process can be found in a recent publication [3].

Figure 3 shows three examples that were taken by using the microscope with the 3D freeform prism array. Figure 3a is the head of a small screw. Figure 3b is a piece of small stone, and figure 3c is bat teeth. It can be concluded from figure 3 that it is much easier to observe the real shape of the objects with complex 3D structures with the help of images taken in different directions. Furthermore a 3D
model of the object can be reconstructed with the information obtained using the 3D microscope.

**3D Microscopy at Low Cost**

In this article, we exhibited a new method to obtain 3D microscopy at low cost and also demonstrated the preliminary results of the newly developed device. Compared to other 3D microscopy technology, the new method and the associated device do not need scanning or adjustment during operation and can obtain images in multiple directions of the same object simultaneously. This new method and device are the results of the development of freeform optics in recent years. Freeform in this research is loosely defined as the optical surfaces that do not have regular shapes such as planar, spherical or aspherical surfaces.

With freeform optics, new devices have been developed such as heads on displays, light concentrators, beam-shaping optics for light-emitting diodes (LED), and f-theta lenses in laser scanners. Freeform optical elements allow significantly reduction in the number of elements in an optoelectronic system and simplify aberration correction and permit increased applications in many fields that require special design goals. The rapid development of freeform optics owns its success largely to the advances in high precision freeform surface fabrication technology, specifically, ultraprecision diamond machining with slow tool servo or fast tool servo. Moreover, by combining ultraprecision diamond machining and injection molding process, high quality and low cost freeform optics can be routinely manufactured.

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