

1 Detail of the optical train of the MEMS based fluorescence laser scanning microscope.

2 Dimensional comparison of various micro-scanning mirrors.

MEMS BASED CONFOCAL FLUORESCENCE MICROSCOPE

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Motivation

It was around 30 years ago that Marvin Minsky sent in a patent application for what have now become established and widely used research instruments – confocal microscopes. This is especially due to their capability to selectively survey cross-sections of individual layers from ‘thick’ samples. This is a decisive advantage particularly in regard to topographical and biological specimens. The markedly improved scattered light suppression combined with a likewise high resolution in both the axial and lateral directions are reasons for their application versatility in the Life Sciences.

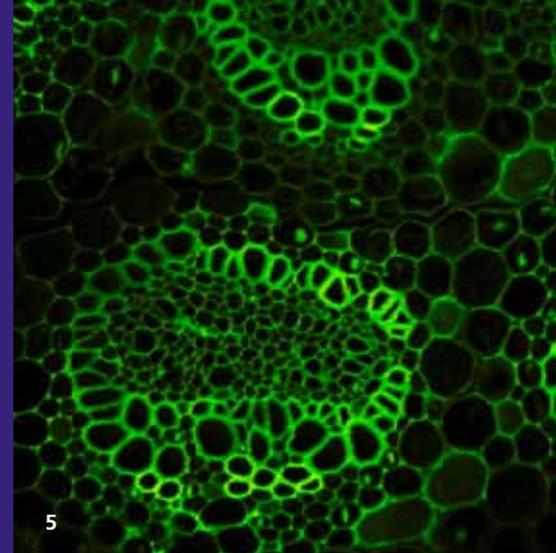
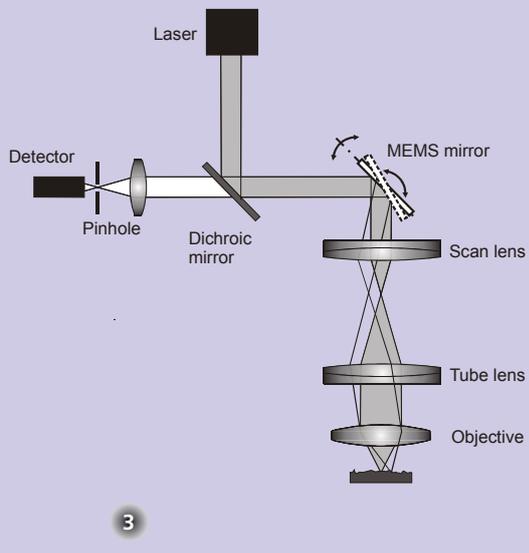
With a confocal fluorescence laser microscope, the sample is irradiated pointwise, and the fluorescent radiation that is stimulated is then measured. Alongside the imaging of horizontal cross-sections,

this technology makes creating 3D models of structured surfaces and fluorescent specimens possible. Some of the main applications are found in the areas of biological and medical research, as well as in industrial quality control.

Due to the main application area in research, the instruments dealt with here are generally complex, stationary units and are accordingly cost-intensive.

Goal

It is for this reason that Fraunhofer IPMS has developed a robust and portable MEMS based fluorescence laser microscope using standard optics. This has been made possible by the integration of a 2D micro-scanner mirror (ref. fig. 2) developed at Fraunhofer IPMS.



Mode of Operation

This microscope works along the same basic principles as a traditional confocal fluorescence laser scanning microscope, yet it features a micro-scanning mirror developed at Fraunhofer IPMS as a core component. The optical train of the microscope is illustrated in fig. 3. The collimated light of a 488 nm diode laser initially passes through a dichroic beam splitter and is then directed onto the micro-scanning mirror. The laser beam is subsequently expanded by means of a telescope and focussed on the sample by a microscope objective. Fluorescence is thus selectively stimulated in the specimen. The fluorescent light emitted from the specimen passes through the objective in the opposite direction and is then separated from the excitation light by a dichroic beam splitter. Finally, the light is focussed onto a pinhole by an additional lens, behind which a detector is situated. Light from layers other than the focal layer of the sample are not focussed on the pinhole. An image of the sample can then be reconstructed with software using the digitalized output signals of the receiver and the knowledge of the current position of the mirror.

Technology

The central element of the confocal fluorescence laser scanning microscope is a micro-scanning mirror developed and manufactured at Fraunhofer IPMS. This silicon MEMS component facilitates the control of the laser beam that is necessary to scan the sample. The miniscule dimensions of the scanning mirror is what makes the compact design of the microscope possible. The electrostatically driven micro-scanning mirror employed has a mirror surface area with a diameter of $D = 2 \text{ mm}$ and oscillates in two orthogonal directions with 190 and 1,290 Hz respectively. The sample is scanned pointwise with a focussed laser spot in the form of a Lissajous pattern in this manner. The image of the object under examination can then be reconstructed with a special deconvolution algorithm.

Focus

- Portable microscope
- MEMS based, robust construction
- Expansion options for 3D images available
- Measurement range $(480 \times 480 \times 100) \mu\text{m}^3$
- Axial and lateral resolution $2 \mu\text{m}$

Fields of Application

- Dermatology / Skin analysis
- Biotechnology
- Industrial quality control
- Non-destructive testing, NDT
- Fluorescent labeled samples

This project was developed in cooperation with TU Dresden and supported by the European Social Fund.

3 Schematic optical train of the fluorescence laser scanning microscope.

4 Specimen holder on the microscope.

5 Fluorescent image of *Convallaria* (Lily of the Valley).