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Measurement of steep edges and undercuts in confocal microscopy



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ABSTRACT

Confocal microscopy is widely used to measure the surface topography of specimen with a precision in the micrometer range. The measurement uncertainty and quality of the acquired data of confocal microscopy depends on various effects, such as optical aberrations, vibrations of the measurement setup and variations in the surface reflectivity. In this article, the influence of steep edges and undercuts on measurement results is examined. Steep edges on the specimen's surface lead to a reduced detector signal which influences the measurement accuracy and undercuts cause surface regions, which cannot be captured in a measurement. The article describes a method to overcome the negative effects of steep edges and undercuts by capturing several measurements of the surface with different angles between the surface and the optical axis of the objective. An algorithm is introduced which stitches different angle measurements together without knowledge of the exact position and orientation of the rotation axis. Thus, the measurement uncertainty due to steep edges and undercuts can be avoided without expensive high-precision rotation stages and time consuming adjustment of the measurement setup.

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1. Introduction

Confocal microscopy is a popular technique for the inspection of specimen in the micrometer range. Modern confocal microscopes allow to capture high-resolution images as well as the specimen topography within seconds. The popularity of confocal microscopy results from its many advantages, for example a high variability in usage which results, among other reasons, from the variety of available objective lenses. Thus the resolution, measurement range and duration can easily be adapted. Due to the high variability of confocal microscopes, this technique is frequently used in different fields including physics, biology, functional surface development, aviation and automobile industries, and optics.

All applications in these fields have in common, that not only a magnified image of the specimen should be captured but also the specimen topography with a precision in the micrometer range. The surface topography of specimen is important for the functionality of many components or processes (de Chiffre et al., 2003; Bruzzone et al., 2008). For example, engineered surfaces with high aspect ratio are frequently used in chemical and biomedical applications to achieve a higher active surface area per unit. Microstructured surfaces are also used to influence the interactions of cells with

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http://dx.doi.org/10.1016/j.micron.2016.03.001 0968-4328/© 2016 Elsevier Ltd. All rights reserved. the surface or can be used as microprobe arrays. The application of micro- and nanotechnology in biomedical systems is commonly known as BioMEMS (Folch, 2013; Bashir, 2004). Special engineered microstructured surfaces can be used in BioMEMS for example as biochips, for drug-targeted delivery or to influence molecule surface reactions. A practical example of the use of microstructured surfaces are micro cantilevers which can be used for analyzing biomolecules such as DNA and proteins (Yue et al., 2004). Yue et al. describe a chip with a cantilever which has a length of 200 µm and a thickness of 0.5 µm. The upper surfaces of the cantilevers are covered with probe molecules, which interact with target molecules. When the probe molecule interacts with the target molecule, the microcantilever deflects due to surface stress. The basic structure of a microcantilever for biomolecule analysis is shown in Fig. 1. A laser beam and an optical system can be used to measure the deflection of the cantilever. The deflection of the cantilever depends on the interaction of the probe and target molecule, but also depends on the geometric properties of the cantilever. In Bashir (2004) the deflection Δz of the cantilever is given by

$$\Delta z = f \frac{L^2}{t^2} \frac{1 - \nu}{E} (\Delta \sigma) \tag{1}$$

L is the cantilever length, *t* is the cantilever thickness, ν is the Poisson's ration, *E* is the Young's modulus and $\Delta\sigma$ is the change in surface stress. According to Eq. (1), it is important to know the dimensions of the cantilever for a detailed analysis. Confocal microscopy is well suited for the measurement of typical cantilever





Fig. 1. Microcantilever for biomolecule analysis. For better illustration, the cantilever and the target molecules are not drawn in the same dimensional scale.



Fig. 2. Topography of a riblet-structured surface.

dimensions in the micrometer range. In Silk et al. (2006) cubic fins with an edge length of 1 mm were manufactured on a component surface to alter the thermal properties of the component. In that application, the dimensions of the surface structures are much larger than the dimensions of the microcantilevers, but confocal microscopy is also well suited for the inspection of structures up to the millimeter range. Another example for the importance of microstructured surfaces can be found in the aviation industry. Sophisticated microstructured surfaces on the outer skin or on the compressor blades of an airplane can be used to reduce the air drag of the aircraft which results in a reduced fuel consumption and a reduced pollutant emission. These microstructures are called riblets and consist of periodic, trapezoidal elevations of the component. The dimensions and period of the riblets depend on the flow conditions and are typically in the range of $20-200 \,\mu$ m. Fig. 2 shows the basic riblet structure. Riblets were first described 30 years ago (Walsh, 1982), but are still rarely used, because fast and accurate manufacturing techniques are still in research. In laboratory experiments, the wall friction of components was reduced by approximately 10% (Bechert et al., 1997). Microstructured surfaces in aviation industry can be challenging because large areas, e.g. the outer skin of an aircraft, needs to be covered with these structures in order to achieve a noticeable effect. Confocal microscopy can be used to measure the structure height, period length and the tip radius of the riblets. These are the most important parameters for the functionality of the riblets. However, the edges of the riblets are difficult to measure using confocal microscopy (Lietmeyer et al., 2013). The edges of the riblets have a flank angle in the range of 45–60°. Therefore, the light intensity which is collected by the objective lens of the confocal microscope is very low. This leads to a low signal-to-noise ratio for the riblet's edges.

The given examples demonstrate the importance of microstructured surfaces. In the research, development and manufacturing process of the microstructured surfaces, it is important to be able to inspect these surfaces. It is desirable to capture high-resolution magnified images of the surface and it is also very desirable to capture the surface's topography. As mentioned above, confocal microscopy is suited for most of the measurement tasks which are required for the inspection of microstructured surfaces. Confocal microscopy was invented by Minsky and patented in 1957 (Minsky, 1961). In the following years, many further developments



Fig. 3. Basic setup of a confocal microscope.

and improvements on confocal microscopy were published. Some notable examples are the first imaging confocal microscope (Petrán et al., 1968), the confocal laser scanning microscope (Åslund et al., 1987) and the two photon fluorescence microscopy (Denk et al., 1990) which is particularly important for the inspection of semitransparent specimen like living cells. A good overview of the history of confocal microscopy can be found in Pawley (2006).

In spite of the variety of available confocal microscopes, most of them have a similar principal design (Leach, 2011). Fig. 3 shows the basic design of a confocal microscope. As a light source a laser or a halogen lamp is commonly used. For image generation, an adjustable mirror system or a rotating Nipkow disc can be utilized. Photomultiplier tubes or CCD sensors are commonly used as detectors. Some commercially available microscope systems even contain several detectors and light sources. For example a photomultiplier tube with laser illumination for the topography measurement of the specimen and a CCD sensor with white light illumination for high-resolution images of the specimen. Most confocal microscopes contain an objective revolver to allow a quick change of the objective and thereby a fast change of measurement parameters, e.g. the magnification factor, lateral and axial resolution and field of view. For axial scanning, the objective is mounted on an axial scanning system. The axial scanning system can include linear and piezo stages for a precise and repeatable displacement of the objective. Most significant for the functionality of a confocal microscope is the pinhole in front of the detector. This pinhole allows light to pass through to the detector, when the specimen is in the focal plane of the objective. When the specimen is not in the focal plane, the pinhole efficiently blocks light and no or very low light intensity is measured by the detector. During a measurement, the distance shift of the objective is carried out by the axial scanning system. The surface topography can be calculated by searching axial position of the objective which has the highest light intensity on the detector. Since the number of axial scanning steps is limited, interpolation techniques are frequently used to precisely determine the position with the highest light intensity.

As mentioned earlier, most confocal microscopes can work with a set of exchangeable objectives. However, the calibration of the microscope must be conducted for each objective differently in order to receive x, y, z measurement data in metric units or, to compensate objective aberrations for example. Objectives for commercially available confocal microscopes are often plan apochromatic objectives with a very high aberration correction. Common magnifications for these objectives range from $1 \times to 200 \times$ and the numerical apertures range from 0.045 to 0.95 (Leach, 2011). Table 1 compares five different objectives.

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