REAL-TIME 3D SHAPE MEASUREMENT OF MICRO DROPLET USING DIGITAL HOLOGRAPHIC MICROSCOPY

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ABSTRACT

This paper reports the development of a microscopic 3D shape measuring method based on digital holographic microscopy, and its application to a microscopic droplet which changes in shape as time progresses. The use of an off-axis holographic/interferometric optical system enables measuring the geometry of microscopic objects in 3D and in real time. The measurement is performed in the volume of 176 µm x 176 µm x 100 µm at 60 fps with in-plane and out-of-plane resolutions of 1.1 µm and 62 nm respectively. We have successfully measured and visualize the 3D shape of a droplet and its dynamic change.

KEYWORDS

Digital holographic microscopy, droplet, visualization, real-time measurement, 3D measurement

INTRODUCTION

Microfluidic devices that utilize discrete flow format such as droplets or plugs to transport chemical reagents and sample materials have been widely used due to their advantage that the amount of the samples and their dead volume can be dramatically reduced [1]. The key issue to develop useful droplet-based devices is to get more understanding of the hydrodynamic phenomena found in the device and to feedback it to the designing of devices [2]. The fluid flow in the droplet-based microfluidic devices shows distinctive and complicated behaviors by the effect of viscous force and surface force. For example, in the process of droplet generation, the droplet takes unique 3D shape which changes periodically. The morphology of the droplet is really interesting from the view point of microfluidics and of great importance to control the size of droplets and the rate of their generation. In order to understand the droplet morphology, it is required to measure directly the 3D shape of a droplet and its dynamic change.

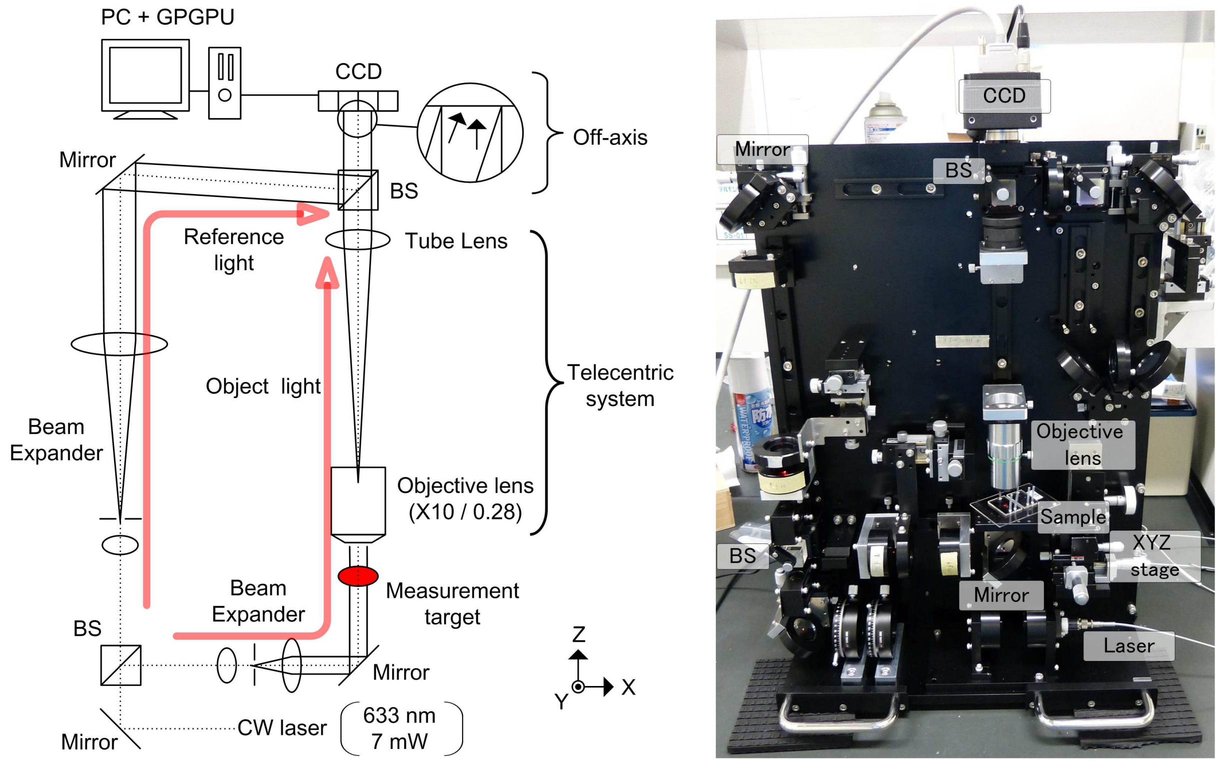
There have been already some commercial profilometers available for microscopic use. For example, the laser displacement sensors are quite practical and useful to measure the surface profile and its dynamic deformation with high temporal resolutions (up to 400 kHz). But, it takes a certain amount of time to scan a planar region even though the spatial scanning rate has been greatly improved. And also, the in-plane measurement resolution is limited by the size of laser spot (at least 500 µm). The confocal laser microscopy is also commercially available for 3D imaging of a microscopic object. However, the fluorescence labeling is always necessary for confocal imaging and the time required for in-plane and out-of-plane scanning is not negligible as well as the laser displacement sensor.

This study focuses on the digital holographic microscopy (DHM) [3], which is a kind of interferometry, as a key technology to solve the problems associated with scanning time and fluorescence labeling. The DHM technique enables us to obtain a digital hologram which contains 3D shape information of a non-fluorescently labeled target without spatial scanning. By analyzing the obtained hologram, we can reconstruct the 3D geometry digitally and measure the shape of the target quantitatively. In this study, we aim to materialize a true real-time 3D shape measurement in microscale using the DHM, and apply it to the measurement of 3D shape of a microscopic droplet.

MEASURING SYSTEM

We have developed the DHM system which is available for the shape measurement of microscopic droplets. Figure 1 shows the schematic illustration and the photograph of the developed DHM system, which consists of an off-axis holographic/interferometric optical system, a He-Ne laser unit (1137P, JDS Uniphase Corporation, USA), a CCD camera (avA1000-120km, BASLER, Germany), a mechanical stage, and a powerful desktop PC with GPGPU (General-Purpose computing on GPU). The image sensor of the CCD camera has the capability to record holograms with 1024 pixel x 1024 pixel resolution at 60 fps. The He-Ne laser emits a continuous laser beam with a wavelength of 633 nm at 7 mW laser power, which is used as both the illumination light and the reference light for holography. When the objective lens with 10x magnifications (M Plan Apo 10X, Mitutoyo Corporation, Japan) is installed in the optical system, the system provides the theoretical measurement volume of 563 µm x 563 µm x 986 µm and the in-plane and out-of-plane measurement resolutions of 1.1 µm and 62 nm respectively. We have also developed the original software for hologram analysis using CUDA which is an IDE developed by NVIDIA Corporation for graphics processing. Our software can show the user the reconstructed intensity images and phase images side-by-side on the same window in real time.

The developed DHM system has three important features. The first one is the optical system based on the off-axis method, which enables the time-resolved measurement at the same speed as the frame rate of the CCD camera. This is because the off-axis optical system allows us to obtain the phase shift data required for interferometry spatially instead of temporally [4]. Secondly, the microscope setup used here is free from phase distortion due to the use of telecentric optical system [5]. The other feature is the capability to display the analysis results on the screen of PC in real time (more than 30 fps at full frame). Since the use of Fourier transform method and GPGPU speeds up the calculation of the optical diffraction [6], the system can reconstruct the intensity image and the phase image in a moment, and show us them in real time.



(a) Schematic of the optical system (b) Photograph of the original microscope

Figure 1. The developed DHM system

3D SHAPE MEASUREMENT

Figure 2 shows the experimental setup for droplet formation. A T-shaped microchannel having the depth of 93 µm has been prepared for this experiment. Silicone oil (KF-6001, Shin-Etsu Chemical Co., Ltd., Japan) is streaming in the main channel (400 µm wide) and water is flowing from the branched channel (50 µm wide) into the main channel at a T-shaped intersection, where small water droplets are generated repeatedly. We measured the surface profile of the water which was changing into a droplet. Since the channel is made of PDMS, the surface of the channel is hydrophobic and the refractive index of the channel is 1.412 (25°C). And also, the silicone oil has the refractive index of 1.412 (25°C), which is the same as that of PDMS. On the other hand, the refractive index of water is 1.333 (25°C). The DHM can detect the liquid interface between the water and the silicone oil thanks to the presence of the difference of refractive index. Both flow rates of the silicone oil flow and the water flow are kept at the flow rate of 3.0 µL/min by using mechanical syringe pumps. As shown in Fig. 2, the measurement was performed in the area of 176 µm x 176 µm at 60 fps in this experiment although the developed system has the capability to measure the area of 563 µm x 563 µm potentially.

The analytical flow chart including the images at each step is shown in Fig. 3. First, a raw image, which is a hologram, is captured by using the developed DHM system. By analyzing the raw image, we can digitally reconstruct the intensity image and the phase image. Then, the height information of the water-oil interface is extracted from the phase image. At that time, we use the intensity image as a mask in order to calculate only the required region and reduce the computational load. The time-series height information is obtained by applying these processes to sequential images. In this way, we can measure the 3D profile of the water surface which deforms dynamically as time progresses.

Figure 4 shows the 3D shape visualizations of the droplet which is formed at a T-shaped microchannel junction. Since we captured the holographic images at 60 fps, the surface profile of the water was obtained at the time intervals of 16.7 msec. This result clearly shows that the water flow becomes thin and elongated before forming a droplet. This means we could successfully measure the 3D profile of the droplet surface sequentially and quantitatively by using the developed system and method.

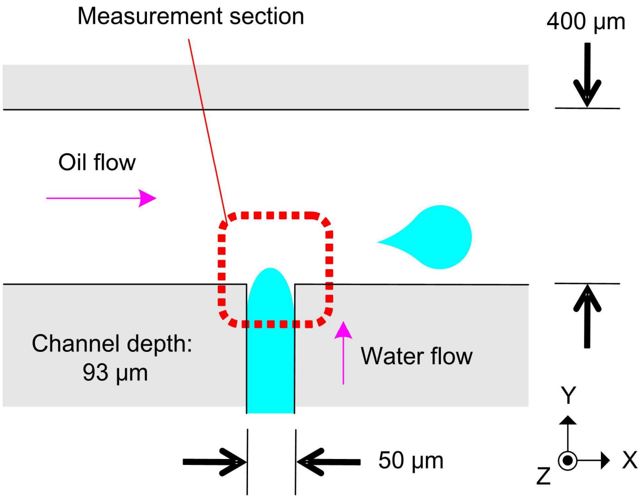


Figure 2. Schematic illustration of the T-shaped microchannel for droplet formation

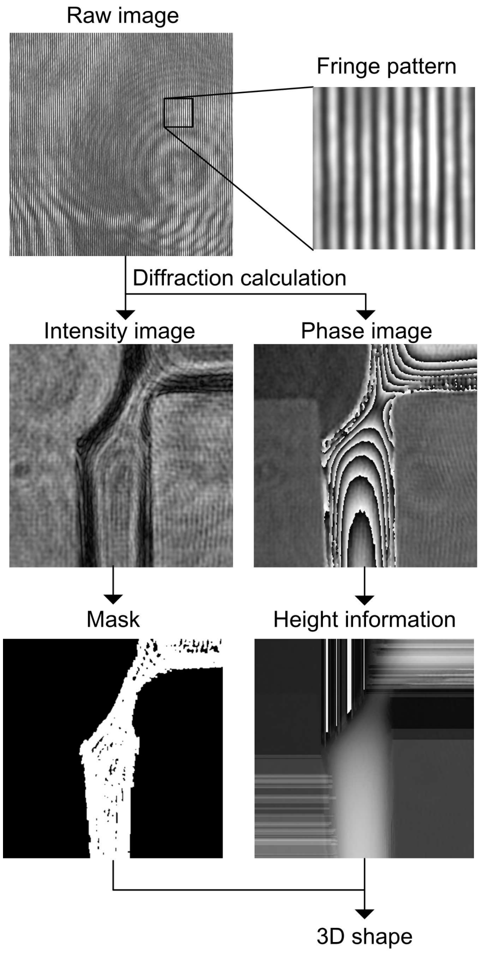


Figure 3. Analytical flow chart

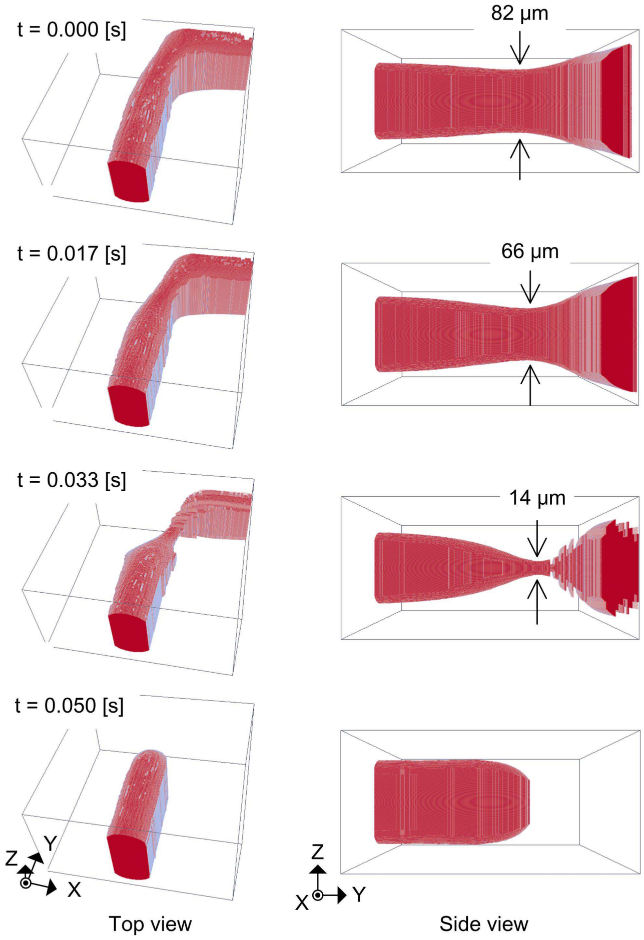


Figure 4. Time-series 3D visualizations of the droplet formation

CONCLUSIONS

We have developed a new microscopic 3D shape measuring system based on DHM. The developed system enables measuring the geometry of a microscopic object or surface in 3D and in real time with both high spatial and time resolution. We have applied the developed DHM-based system to the measurement of 3D shape of a droplet which is formed at a T-shaped microchannel junction. As a result, we have succeeded in quantitative measurement of the 3D geometry of the droplet and its dynamic change in a microchannel.

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