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Deep learning-based enhancement of digital holographic particle tracking velocimetry

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Abstract

Digital holographic PTV (DHPTV) can provide time-resolved 3D positional information of micro-scale particles from recorded holograms. Thus, it has been employed to analyze various micro-scale flows. However, numerical image reconstruction and auto-focusing processes are essentially required to extract 3D volumetric information of micro particles. These processes are computationally expensive and time-consuming. In this study, we developed a new method for rapidly obtaining 3D positional information of particles using DHPTV combined with deep learning algorithms. The developed method was applied to investigate micro-scale pipe flow for examining its performance. 3D trajectories of particles were consecutively tracked and 3D velocity field information was obtained. Experimental results demonstrate that the adoption of deep learnings on DHPTV makes it possible to analyze micro-scale flow without reconstructing and auto-focusing with dramatically reduced computation time.

1 Introduction

Digital holographic microscopy (DHM) can record interference patterns (hologram) of a sample. Since the hologram encrypts 3D information, digital holographic particle tracking velocimetry (DHPTV) has been widely utilized to analyze the dynamic behaviors of various micro-scale objects or to investigate microscale flows (Choi et al. 2012; Katz and Sheng 2010; Memmolo et al. 2015). DHPTV can simultaneously track hundreds of samples in a volume with deep observable depth without calibration or depthwise scanning procedures.

However, additional post image processes are necessary to obtain 3D positional information from the captured hologram. Each recorded hologram is numerically reconstructed and the image sharpness of all reconstructed images within a range of estimated distance is then calculated by using a proper autofocusing function (Choi and Lee 2009; Choi et al. 2012; Lee et al. 2011; Memmolo et al. 2015). Furthermore, several physical parameters, such as camera resolution, spatial resolution in the holographic image plane, wavelength of the light source, reconstruction depth, size of image segmentation for autofocusing, should be manually determined.

Recently, deep learning has been getting large attention, because of its usefulness and effectiveness in various fields (LeCun et al. 2015). Deep learning is a multi-layered artificial neural network and it has been successfully applied in various areas where numerous data are available, such as computer vision, medical image analysis, image resolution enhancement, and noise reduction.

In the present study, we proposed a new method for extracting 3D positional information of numerous objects without time numerical reconstruction and autofocusing procedures by applying deep learnings to DHPTV technique. Holograms of transparent polystyrene microbeads located at various depth positions were used as training data sets. To determine in-plane positions of the microparticles, SegNet and circular Hough transform (CHT) were employed (Badrinarayanan et al. 2015; Cheong et al. 2009). The trained SegNet provided a binary image from which the background and the particles were separated. Then CHT extracted the centers of individual particles from the binary image. After this in-plane positioning procedure, the holograms of particles were segmented and then used for training deep convolutional neural network (CNN) (Vedaldi and Lenc 2015). This trained network was used to identify the depth positions of the particles from these segmented holograms. The performance of the proposed 3D position determination method was verified by conducting a planar surface experiment and tracking the particles moving in a microtube. Compared to conventional methods, the proposed method was found to obtain 3D positional information of particles more accurately and the processing time was greatly reduced.

2 Material and methods

2.1 Experimental set-up

A single-beam in-line DHM system was employed to measure the 3D positional and motional information of 10 μm polystyrene particles. Fig. 1(a) shows the experimental set-up of the in-line DHM system. It consists of a continuous diode-pumped solid state laser (λ = 532 nm, 100 mW, Crystal Laser, USA), a spatial filter, a collimating lens, a water immersion objective lens (40×, Nikon, Japan), and a high-speed CMOS camera (FASTCAM Mini UX100, Photron, Japan). The laser beam was spatially filtered and collimated. The collimated laser beam illuminated particles suspended in a NaCl solution (n = ∼ 1.333). The 40X water immersion objective lens magnified holograms of particles. The magnified holograms were recorded by the high-speed CMOS camera with 1280 × 1024 pixels. The spatial resolution is 0.25 μm/pixel.

The particle suspension was injected into a transparent FEP (fluorinated ethylene propylene, n = 1.338) microtube of 300 μm in diameter, which was immersed in water. NaCl solution was used as the working fluid to match the densities of the solution and the polystyrene particle (ρ = 1.05 g/mL). Flow rate was adjusted to maintain the Reynolds number of 1.5. The focal plane of the objective lens was adjusted using the z-axis translation stage of the microscope. With the help of this simple experimental set-up, a number of holograms of particles located at various depths (z) ranged from z = 0 to z = 500 μm could be simultaneously obtained to collect data sets.

2.2 Deep learning-based DHPTV

The overall procedure for obtaining 3D motion of microparticles using the DHPTV and deep learnings is illustrated in Fig 1. Holograms of microparticles were recorded by using the in-line DHM system [Fig. 1a].

After recording holograms, the in-plane positional information of particles was acquired by applying the SegNet and CHT [Fig. 1b]. The SegNet is a deep fully convolutional neural network architecture for semantic pixel-wise segmentation (Badrinarayanan et al. 2015). The SegNet was trained with 30 pairs of raw holographic images and their corresponding binary images. After training, the SegNet could rapidly create binary images to separate particles from the background.

The CHT is one of the modified versions of the Hough transform, which has been widely used to detect boundaries in an image (Cheong et al. 2009). The CHT focuses on finding of circular patterns in an image. The centers of individual particles were detected by employing the CHT.

After detecting in-plane positions of particles, depth-positional information of particles was determined by the CNN [Fig. 1c]. The area around the center of each particle was segmented with a square box of 30 × 30 μm^2. The CNN was trained with these segmented holograms of particles at various depths (z = 0 – 500 μm with intervals of 1 μm) and then the automatic classification of the recorded holograms of particles into the pretrained 501 classes was conducted. The depthwise positions of individual particles were accurately acquired by the trained CNN.

After extracting the 3D positional information of individual particles, the two-frame particle tracking velocimetry (PTV) algorithm based on matching probability was applied to obtain the 3D motions of particles inside the microtube [Fig. 1c] (Baek and Lee 1996).

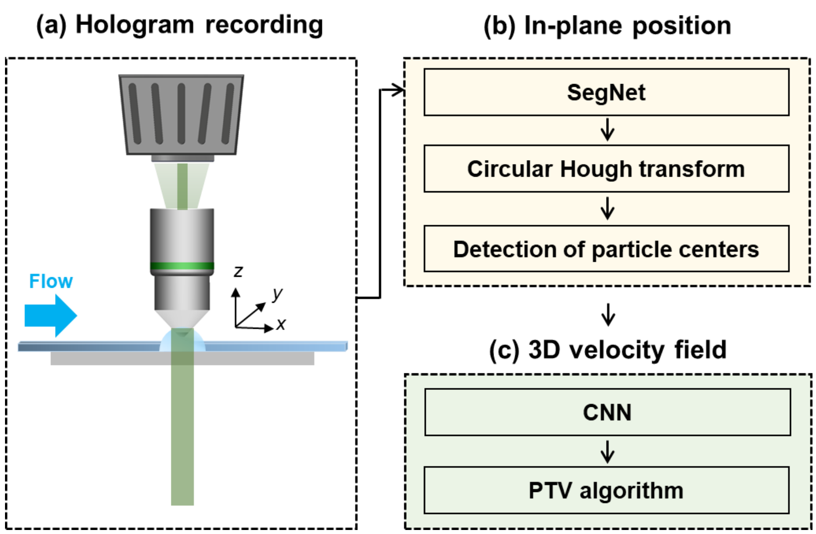

Figure 1: Overall procedure of DHPTV enhanced by deep learnings

3 Results and discussion

3.1 Extraction of in-plane position

To determine the in-plane positions of particles accurately and rapidly, the SegNet and CHT were adopted in this study. We detected the center positions of particles by adopting CHT after converting raw holographic images into the binary images by employing the SegNet, for which the boundaries between the particles and the background are clear and noiseless. A total of 30 pairs of raw holograms and corresponding binary images were utilized as training data sets of the SegNet [Fig. 2a]. After the training stage, the SegNet converted raw holographic images into the binary images to separate background and particles [Fig. 2b]. In the converted binary image, the centers of particles were detected by applying the CHT [Fig. 2c]. These processes used for obtaining in-plane positions of particles do not include procedures of numerical reconstruction and projection of reconstructed images. Thus, the computational time is greatly reduced.

To evaluate the performance of the proposed method, the degree of deviations (d) from the real in-plane positions were calculated. In addition, the corresponding RMS in-plane positioning errors of the proposed method and the conventional method were compared [Fig. 2d]. The deviations at a projection (r) were approximately 0.40 ± 0.43 μm and 0.68 ± 0.3 μm for the SegNet with CHT and the conventional local intensity peak searching method, respectively. The RMS in-plane positioning error for the proposed method was decreased by 24%, compared to the conventional method.

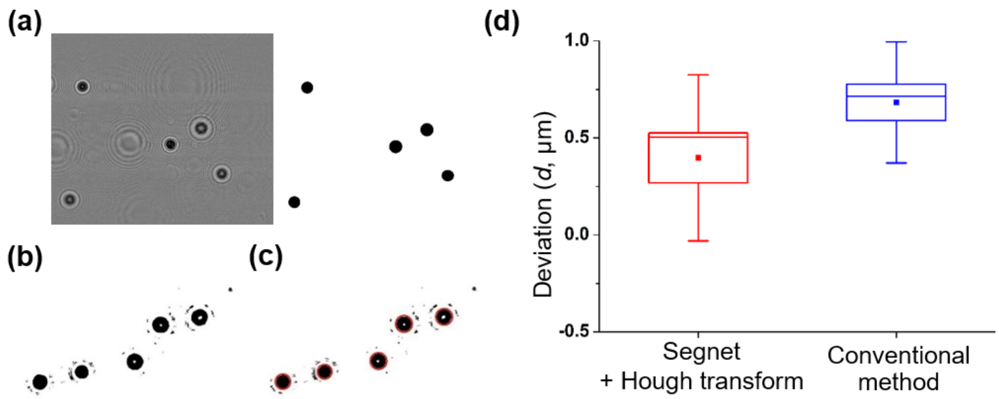

Figure 2: Extraction of in-plane positions of microparticles

3.2 Extraction of depthwise position

To determine the depth positions of particles, CNN was employed as a deep learning network in the present study (Vedaldi and Lenc 2015). To collect training data sets based on the determined in-plane positions, the areas around the extracted in-plane positions were segmented into cross-sections of 30 × 30 μm^2. These cropped images were used as training data sets for the CNN [Fig. 3a]. 136 holograms at various depths ranged from z = 0 to 500 μm were utilized as training data sets. The CNN differentiated the undefined input holograms and provided depth positional information of particles.

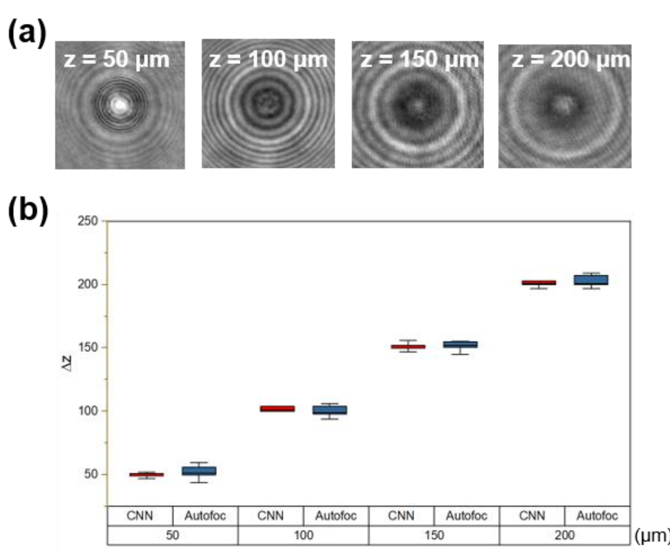

Figure 3: Extraction of depthwise positions of microparticles

The performance of the established depth position measurement technique was evaluated by using a planar test target. The test target was precisely positioned at four different depths (50, 100, 150, and 200 μm) apart from the focal plane. Through this planar surface experiment, the uncertainty in the depth position measurement could be analyzed.

As shown in Fig. 3b, the depth locations acquired from the CNN are well matched with the target locations. The conventional method also provides acceptable results in depth position. The CNN method provides better results, compared to conventional autofocusing method. Compared to the conventional method, S.D. and corresponding RMS errors are largely reduced.

To measure accurate depth position accurately using the conventional method, several requirements have to be satisfied. The type of autofocus function and segmentation size should be carefully determined by a human, because it is closely related with the depth (z) positioning error. In addition, the segmentation size should be determined adequately to extract the depth positions of microparticles accurately. Compared to the conventional method, the proposed method using the CNN helps to minimize human interpretation in error.

3.3 3D motion of microparticles flowing in a microchannel

The proposed method was applied to track 3D motions of microparticles and obtain velocity field of particles moving in a circular microtube flow. After capturing more than 1000 consecutive hologram images, 3D positional information of microparticles were extracted. Then a PTV algorithm was adopted to trace the particles in 3D space unambiguously. The 4D (3D space and time) tracking results of moving particles are shown in Fig. 4a. The linear movements of microparticles are clearly observed. The time-averaged velocity profile is shown in Fig. 4b. The velocity profile is in a good agreement with the theoretical profile of a Hagen-Poiseulle flow. Although there are some time varying velocity fluctuations due to 3D positioning errors, 3D motions of particles are accurately tracked within an acceptable level of uncertainty.

The time required for obtaining 3D motions of particles flowing in the microtube flow was also evaluated. Totally 24,230 holograms of microparticles in 2,000 consecutive holographic images were analyzed to investigate the microtube flow. The size of each hologram image was 1280 × 700 pixels. Image analysis was carried out using a desktop computer. The processing time for 3D tracking is largely decreased by more than 98%.

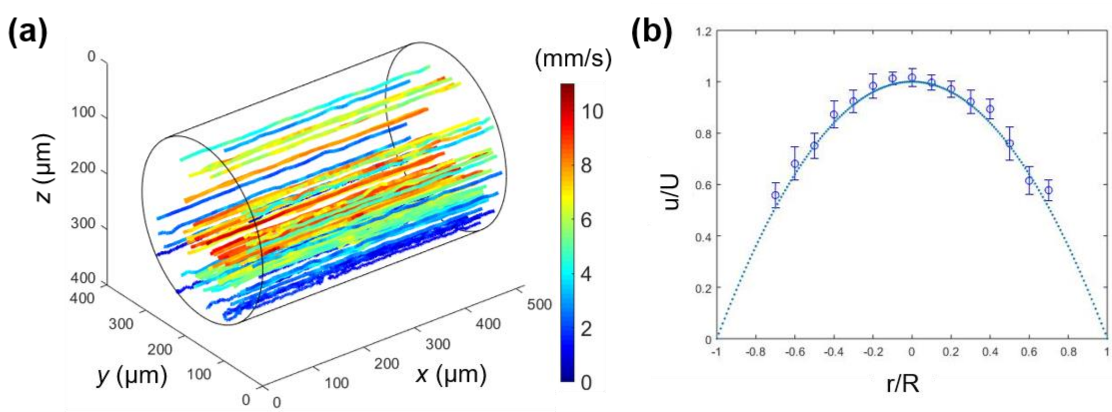

Figure 4: 3D motions of microparticles flowing in a microtube

4 Conclusion

In this study, a new framework for 3D positioning of microparticles by combining DHPTV technique and deep learnings and was proposed. To determine the in-plane positions of particles, SegNet and CHT were adopted. In addition, the CNN was trained to determine the depth positions of particles for minimizing human interpretation. The proposed deep learning enhanced DHPTV technique would be useful for accurate and rapid analysis of 3D dynamic motion of particles or cells.

Acknowledgements

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