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High-resolution *in-situ* LDV monitoring system for measuring velocity distribution in blood vessel

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ABSTRACT

We herein describe a cross-sectional multiple-point laser Doppler velocimetry (CS-MLDV) system for monitoring blood vessels that are sutured and connected during an operation. In order to observe the condition of a blood vessel during an operation, the previously developed linear MLDV (L-MLDV) system can realize velocity distribution imaging of the carotid artery in a living mouse by means of traverse laser light. We subsequently developed a CS-MLDV system, which can measure the instantaneous two-dimensional (2D) flow velocity, by upgrading the optical components and signal processing used in L-MLDV. The validity of the CS-MLDV results was verified through comparison with the results of a computational fluid dynamics (CFD) analysis. The results of the CFD analysis were similar to the experimental results obtained under the same flow field condition. Moreover, an instantaneous 2D velocity distribution can be obtained even for the case of flowing blood. Finally, we carried out *in-vivo* measurement in a mesenteric vessel of a mouse in order to demonstrate the potential of the CS-MLDV for use in surgery.

1. Introduction

Recently, *in-vivo* blood flow measurements, referred to as blood flow imaging techniques, have been useful in the diagnosis and monitoring of disease, as well as medical research [1,2]. In particular, techniques for measuring blood flow are important in surgeries that involve vascular suturing [3,4]. Furthermore, visualization of the blood flow distribution in a sutured vessel can be used to confirm the effectiveness of the suture during surgery if the visualization results are immediately available (instantaneous imaging is ideal).

Diagnosis of the blood flow condition during abdominal surgery requires live images of the blood flow velocity distribution with both high spatial resolution and high time resolution [5]. However, the spatial resolution of conventional clinical methods, such as ultrasonography [6] and X-ray radiography [7,8], is insufficient for this purpose. Ultrasonography provides relative information on the blood flow rate, and X-ray radiography can provide high spatial resolution, but requires the patient to be injected with a contrast agent.

Laser Doppler velocimetry (LDV) is widely used in blood flow velocity measurements and is well established as a method for analyzing particle movements at a single point in a fluid based on the Doppler effect [9–14]. However, LDV requires a significant measurement time in order to delineate a two-dimensional (2D) or three-dimensional (3D) flow field. Although this technique can obtain blood flow images in blood vessels by means of a scanning laser beam, the image output remains slow.

In particle image velocimetry (PIV), velocity information can be obtained by analyzing images of tracer particles dispersed in a working flow captured by a camera [15–20]. This commonly involves the use of an imaging sensor, an irradiation apparatus, and a computer for data analysis. Particle image velocimetry concurrently acquires 2D velocity information across a measurement plane making it possible to detect in-plane 2D blood flow structures with excellent spatial resolution. Hybrid PIV-particle tracking velocimetry (PTV), which has a high spatial resolution, can be used to measure *in-vivo* blood flow [21]. However, in order to measure fluctuations in the velocity profile, both the blood cells and the fluorescent particles must act as seed particles. Hence, it is necessary to inject exogenous fluorescent particles.

Moreover, by using an intravital microscope with a water

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immersion objective lens and metal-halide back illumination in conjunction with a high-speed digital video system, an *in-vivo* PIV method having high spatial resolution (0.8 μm × 0.8 μm) and high temporal resolution (1000 frames/s) was applied to images of the arteriole in the rat mesentery [22]. This PIV technique was improved to increase the accuracy of red blood cell velocity measurements by tracking the mesentery motion. However, in this system, a sufficient working distance cannot be secured, because the measurement subject is in contact with the lens via the immersion liquid. As a problem endemic to PIV, the processing load for the time-series pulsatile blood flow velocity data is excessively high, even using the most recent data processing method [23].

In LDV, only blood cells can be used as self-seeding particles, and neither fluorescent particles nor a contrast medium can be injected into the blood vessel in order to measure the *in-vivo* blood flow. The novel contribution of the present study is the development of an apparatus that can provide allow instantaneous blood flow imaging in a vessel. In order to obtain instantaneous velocities, we have proposed two LDV methods: (1) simultaneous measurement using multiple channels with a linear arrangement, referred to as L-MLDV [24–26], and (2) simultaneous measurement using multiple channels arranged in a grid pattern, referred to as cross-sectional multiple-point LDV (CS-MLDV) [27]. Cross-sectional multiple-point LDV is effective for ensuring a working distance of 280 mm between the measurement area and the optical fiber array. The working distance is determined based on the focal length of the achromatic lenses along the received light line. In a previous study, we concentrated on resolving technical issues involved in obtaining the flow velocity distribution. In the present study, the validity of the measurement results obtained by CS-MLDV was confirmed through comparison with the results of a computational fluid dynamics (CFD) analysis. Furthermore, we attempted to perform 2D blood flow imaging of a blood vessel passing along a membrane in a mouse (*i.e.*, a mesenteric vessel).

2. Results and discussion

2.1. Results of pulsatile L-MLDV in the carotid artery

In a previous study, we demonstrated that an L-MLDV system could be used to visualize the blood flow in the capillaries of a mouse [28–30]. However, the values in our velocity images were averaged over the measurement period (1.0 s), and the possibility of simultaneous blood flow measurements at all points in the imaged area was not discussed. Therefore, in the present study, we first performed simultaneous blood flow measurements in the mouse blood vessel using multiple-point LDV (MLDV). The carotid artery is the optimum measurement target because it is clearly visible.

Fig. 1 shows a schematic diagram of the operating principle for the previously developed L-MLDV. The optical components are described in detail in Refs. [24–26]. Two laser light sheets emitted from the rod lenses are made to intersect along a straight line at the focal point of the imaging system, and Doppler signals from different points on this line are detected using a linear array of optical fibers. Individual velocity information for multiple points along a straight line can be obtained simultaneously.

In LDV, which detects the Doppler shift frequency in a single laser beam, blood flow parameters are estimated by integrating the spectrum of the optical signal [31,32]. The number of moving red cells (N) in the measurement volume is proportional to the integral of the Doppler power spectrum [$P(f)$], which can be obtained as follows:

$$N \propto \frac{1}{I^2}\int_0^\infty P(f)df \tag{1}$$

where I is the laser light intensity. If the measurement volume can be determined, Eq. (1) can be expressed in terms of the red blood cell concentration. The perfusion [*i.e.*, the product of N and velocity (v)] is proportional to the integral of the frequency-weighted Doppler power spectrum, which can be expressed as follows:

$$\text{Perfusion} \propto \frac{1}{I^2}\int_0^\infty fP(f)df \tag{2}$$

Therefore, the average velocity can be written as follows:

$$v = d\frac{\int_0^\infty fP(f)df}{\int_0^\infty P(f)df},\quad d = \frac{\lambda}{2n\ \sin\ \theta} \tag{3}$$

where d is the fringe width in the interference pattern, 2θ is the intersection angle between the two laser beams, and n is the refractive index of the flowing fluid.

Fig. 2(a) shows the mouse carotid artery that was visualized in the present study. The blood flow velocity in the artery was measured using L-MLDV at the location indicated by the asterisk. Blood flow velocity mapping was performed for the area indicated by the dashed line (2.5 mm × 5.0 mm). Fig. 2(b) shows the temporal change in blood perfusion estimated using Eq. (2), and Fig. 2(c) shows the blood velocity at the same position estimated using

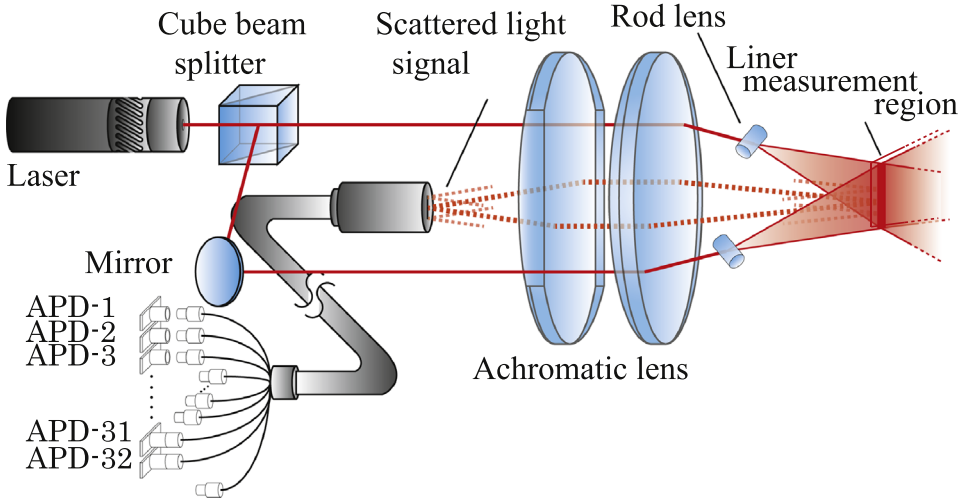

Fig. 1. Schematic diagram of L-MLDV system.