

Non-Invasive Capillary Blood Flow Measurement: Laser Speckle and Laser Doppler

A.K.Jayanthy, N.Sujatha, M.Ramasubba Reddy

Abstract—Microcirculation is essential for the proper supply of oxygen and nutritive substances to the biological tissue and the removal of waste products of metabolism. The determination of blood flow in the capillaries is therefore of great interest to clinicians. A comparison has been carried out using the developed non-invasive, non-contact and whole field laser speckle contrast imaging (LSCI) based technique and as well as a commercially available laser Doppler blood flowmeter (LDF) to evaluate blood flow at the finger tip and elbow and is presented here. The LSCI technique gives more quantitative information on the velocity of blood when compared to the perfusion values obtained using the LDF. Measurement of blood flow in capillaries can be of great interest to clinicians in the diagnosis of vascular diseases of the upper extremities.

Keywords—Blood flow, Laser Doppler flowmeter, LSCI, speckle

I. INTRODUCTION

MEASUREMENT of tissue blood flow provides essential contribution to the diagnosis of diseases which causes an impaired blood flow. Many techniques are currently being used to measure blood flow. Some of the techniques are invasive such as the electromagnetic blood flowmeter and the thermal convection based blood flowmeter, while some use scanning techniques such as the ultrasound and color Doppler scanners. The ultrasound technique cannot be used for measuring blood flow in capillaries because of the very slow blood velocity in these vessels, which is typically of the order of 1mm/sec [1], [2].

When coherent light is scattered from a rough surface (roughness which is comparable to the wavelength of light being used) a random interference pattern known as a speckle pattern is formed. Its first prominence came into picture with the advent of the laser in 1960's [3], [4]. The speckle pattern was found to carry quantitative information about the object under illumination and applications were soon invented. The speckle pattern obtained from a biological specimen is termed as a bio-speckle pattern. The laser speckle contrast imaging [LSCI] technique overcomes the above disadvantages and

provides a non-invasive, whole field and real time image of microcirculation in tissues. In the past two decades a lot of activity has been going on around the world involving the laser speckle contrast imaging technique (LSCI) and has been used by many researchers for measurement of blood flow in microcirculation. In medicine, LSCI has been used to measure blood flow in the retina [5], cerebral blood flow [6], [7], human knee [8], characterization of atherosclerotic plaques [9], free flap measurements [10] and more recently in the non-invasive measurement of concentration of red blood cells in phantom body fluids [11], [12] etc.

The laser Doppler flowmetry [LDF] is also based on dynamic light scattering in tissue. The LDF analyses the frequency spectrum of the light intensity fluctuations observed when laser light is scattered from moving particles.

In this paper the LSCI technique and a commercially available laser Doppler blood flowmeter have been used to measure capillary blood flow in the finger tip and elbow of 10 normal subjects and the results have been compared.

II. THEORY

The LSCI technique is based on penetration of light in the skin. As the scattering and absorption effects decrease the intensity of light reaching the blood vessels, the mean penetration depth in skin is defined by that depth where the initial intensity is reduced to a fraction $1/e$ (37%) [13]. The mean penetration depth of He-Ne laser light with wavelength 632.8nm as cited in literature is about 0.5mm [13], [14] and since the thickness of the epidermis ranges from 38 μ m to about 370 μ m [13], [15] the light can just reach the blood vessels at the skin surface and is scattered by the red blood cells in the dermal layer [15]. The velocity of red blood cells in the capillaries is found to vary between 0 and approximately 2mm/sec [13], [16]. The light scattered from the moving red blood cells can therefore be utilized to obtain information about blood velocity in the capillaries [17].

A random intensity distribution called as a speckle pattern is formed when fairly coherent light is either reflected from a rough surface or propagates through a medium with random refractive index fluctuations [18]. The speckle pattern is random in nature and can only be explained by statistics. Goodman has developed a detailed theory and explained the statistics of speckle patterns [18]. The statistics of speckle patterns is conveniently divided into first and second order statistics. First order statistics are those that are concerned with the properties of the speckle pattern at a single point in space, while second order statistics as the name implies are concerned

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with the joint statistical properties of the speckle pattern at two or more points and gives measures of the granularity of the speckle patterns [19]. Speckle contrast is an example of a first order statistic while speckle size is an example of a second order statistic.

Laser speckle patterns obtained from illumination of most biological tissues are not “fully developed” in the sense that their brightness distribution does not follow the negative exponential relationship as derived by Goodman [20]. One of the properties of a fully developed (ideal) speckle pattern is that the standard deviation of the spatial intensity variations is equal to the mean intensity. For speckle patterns that are not fully developed as in the case of bio-speckles, the ratio of the standard deviation to the mean intensity can be used as a measure of the contrast of the speckle pattern [21]. This definition of speckle contrast is made use of in this paper.

The contrast C of a speckle pattern is defined as the ratio of the standard deviation (σ_s) of the intensity variations to the ensemble average of the intensity $\langle I \rangle$ and is expressed by equation (1) [18].

$$C = \frac{\text{Standard Deviation}}{\text{Mean Intensity}} = \frac{\sigma_s}{\langle I \rangle} \quad (1)$$

If the scattering particles producing the speckle pattern are in relative motion, the optical path differences in light travelling from the various particles to the image plane will be constantly changing and this results in a randomly fluctuating speckle pattern termed as “time varying speckle” [21]. When such a speckle pattern is captured by an imaging device that has a finite integration time, then some of the fluctuations will be averaged out and the resulting speckle pattern will display a lower contrast value [21]. The speckle contrast therefore depends on the integration time of the detector and on the velocity of the moving scatterers (moving red blood cells), and lies between the values of 0 and 1 [21].

Goodman related the variance $\sigma_s^2(T)$ of the spatial fluctuations to the time average of the autocovariance $C_t(\tau)$ of the intensity fluctuations by the relation given in equation (2),

$$\sigma_s^2(T) = \frac{1}{T} \int_0^T C_t(\tau) d\tau \quad (2)$$

where T is the integration time (exposure time of the CCD camera).

Assuming Lorentzian velocity distribution (because the capillary network is so convoluted) the speckle contrast C is given by equation (3) where τ_c is the correlation time of the intensity fluctuations (i.e. the time taken for the autocorrelation function of intensity to fall from a value of 1 to $1/e$).

$$C = \sqrt{\frac{\tau_c}{2T} \left\{ 1 - \exp\left(\frac{-2T}{\tau_c}\right) \right\}} \quad (3)$$

Having calculated the contrast value C from equation (1) the τ_c value can be calculated using equation (3). The next step is

to establish a relationship between the correlation time τ_c and the decorrelation velocity v_c .

Although the precise relationship between the velocity of the scatterers and τ_c is not known the simplest approach [22] defines the decorrelation velocity v_c as given by equation (4).

$$v_c = \frac{\lambda}{2\pi\tau_c} \mu\text{m/sec} \quad (4)$$

When He-Ne laser is used ($\lambda=632.8\text{nm}$) the equation (4) reduces to the form as given in equation (5).

$$v_c = \frac{0.1}{\tau_c} \mu\text{m/sec} \quad (5)$$

Another approximation for v_c was given by Bonner and Nossal [23] which took into account factors such as particle size and is given by equation (6).

$$v_c = \frac{3.5}{\tau_c} \mu\text{m/sec} \quad (6)$$

The moving scattering particles in blood namely the red blood cells (RBC) produce the time varying speckle in biological tissues. Any variation in the velocity of the blood causes a corresponding increase or decrease in the contrast value of the speckle pattern. As variations in contrast values of the speckle pattern contains the essential information and is considered for further analysis the technique is termed as Laser Speckle Contrast Imaging (LSCI).

The working principle (shown in Fig. 1) of the laser Doppler blood flowmeter is based on recording the Doppler shifts that the photons undergo when scattered by the moving red blood cells.

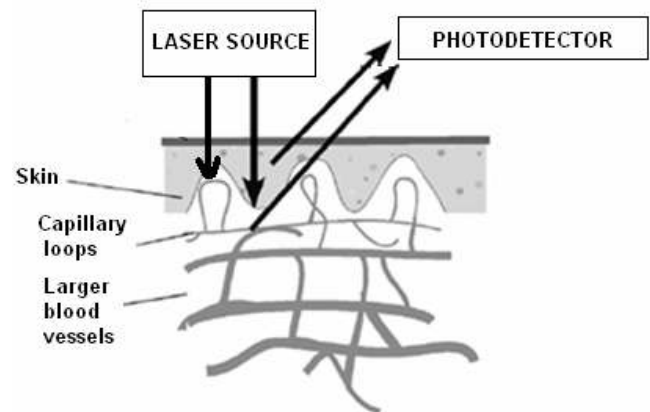


Fig. 1 Laser Doppler blood flowmeter

Laser light from a gas or a semiconductor laser illuminates the tissue through an optical fibre. The photons are reflected, scattered and absorbed in the tissue and those that hit the red blood cells become Doppler shifted, whereas those that are reflected in stationary structures are refracted without any change in frequency [24]. The pickup fibre conducts these photons to a detector where the shifted and the nonshifted photons are mixed at the surface of a square-law photodetector. The elementary wave mechanics says that this type of mixing results in a sum and difference frequency of which the difference wave is proportional to the average

velocity of the red blood cells and the resulting Doppler spectrum is usually in the range of 30 to 12,000Hz [24].

The Doppler frequency recorded [25] is given by equation (7) where $\delta\nu$ is the Doppler frequency, c is the velocity of the wave, ν its frequency and u is the velocity of the moving particle. As both c and ν are known the velocity u can be calculated.

$$\delta\nu = \nu \frac{2u}{c} \quad (7)$$

As mentioned earlier coherent light scattered from moving particles produces intensity fluctuations that can be used to measure the velocity of the moving scatterers. Both the approaches namely LSCI and LDF can be used to analyse these fluctuations. It has been established [25] that the two techniques are effectively identical. The aim of this work is to establish that LSCI has the added advantage of giving more quantitative information when compared to LDF.

III. MATERIALS AND METHODS

The schematic of the experimental setup used for the LSCI analysis is shown in Fig. 2. A laser source (632.8 He-Ne laser) illuminates the object of interest and the resulting bio-speckle pattern is imaged by a CCD camera. The speckle pattern is captured by the frame grabber card and processed on the computer using the specially developed software and the corresponding false color contrast map is observed on the monitor.

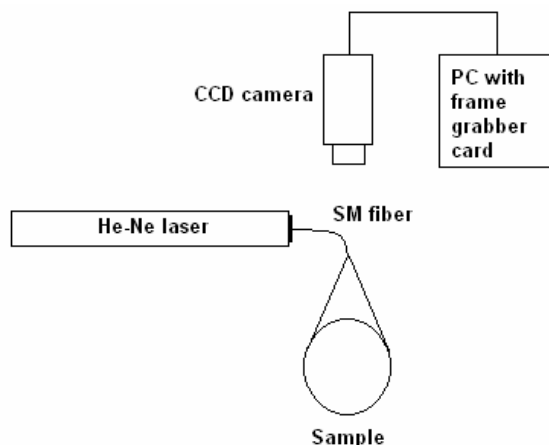


Fig. 2 Schematic of the experimental setup for LSCI

The number of pixels used to compute the local speckle contrast and the scaling of the contrast map can be selected by the user. A choice of a low number of pixels reduces the validity of the statistics whereas the choice of a higher number limits the spatial resolution of the technique [21]. The speckle contrast defined by equation (1) was computed using 5x5 areas of pixels for the study in this paper.

The contrast values thus obtained are then assigned a false color value according to the false color scale in use and the resulting false color contrast map is displayed on the monitor. The false color scale is linear in equal steps of contrast. In general, the contrast range obtained in an image is much

smaller than the theoretically possible range of 0 to 1 [21]. In order to make full use of the false-color scale, the user can also have the option of applying a scaling factor to stretch the contrast range to fill the scale.

The ML-191 model blood flowmeter (schematic shown in Fig. 3) has been used to measure the blood perfusion in the finger tip and well as in the elbow using a surface probe with a self adhesive ring. The LDF consists of an 830nm±10nm semiconductor laser diode and is designed to be used with the AD instruments PowerLab system (Data Acquisition System) and LabChart software. The LDF output produces 1mV for each blood perfusion unit and the maximum output of the unit is 5000 perfusion units.

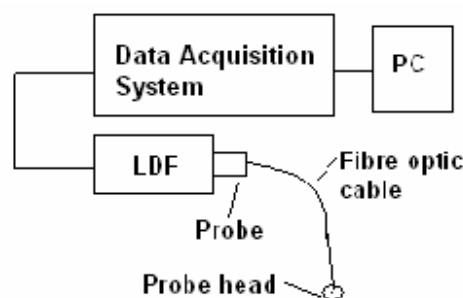


Fig. 3 Laser Doppler blood flowmeter

IV. RESULTS AND DISCUSSION

Initial experiments have been carried out to demonstrate the validity of the LSCI technique in monitoring the capillary blood flow in the finger tip and elbow of 10 normal subjects and the results have been compared with the corresponding readings obtained using a LDF (Model ML-191 Blood FlowMeter/AD Instruments) and the results are encouraging.

The study was performed on 10 healthy non-smoking male and female volunteers. The mean age was 27.1±5.93 (SD) years (range 21-37 years) and the mean body mass index (BMI) was 21.93±2.01 (SD) (range 19.49 – 23.89). Informed consent was obtained from all volunteers, and the study was approved by the Institutional Ethics Committee of Indian Institute of Technology, Madras.

The finger tip and elbow region were illuminated with the He-Ne laser and the corresponding speckle images were recorded using the experimental setup shown in Fig. 2. Using the developed software the contrast values were calculated and the false colour contrast images were also obtained. In the obtained false colour contrast images an area of interest was fixed as 50x50 pixels on the elbow region and as well as at the finger tip and the average contrast value and the average velocity in these areas were calculated and tabulated in Table I.

Using the LDF (Model-ML-191 Laser Doppler Blood FlowMeter/AD Instruments) the blood perfusion values were recorded for duration of 1minute on the same area of interest in the elbow and the finger tip and the average perfusion value (PU) was calculated. The readings are tabulated in Table I.

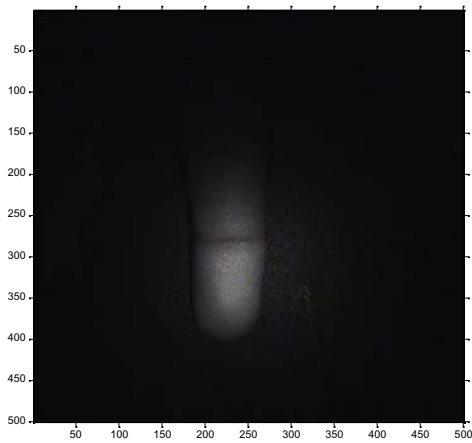


Fig. 4(a) Raw speckle image of finger tip

The raw speckle image and the corresponding false colour contrast map image computed using equation (1) of the finger tip of one of the subjects are illustrated in Fig. 4(a) and Fig. 4(b). The speckles are clearly visible in the raw image. A closer inspection shows that blurring has occurred in areas of higher blood flow which can be clearly seen in the false colour speckle contrast map (Fig. 4(b)).

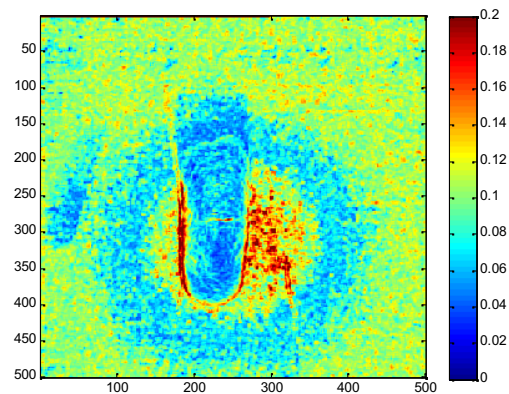


Fig. 4(b) False colour contrast map of image in Fig. 4(a)

The raw speckle image and the corresponding false colour contrast image computed using equation (1) of the elbow region of one of the subjects are illustrated in Fig. 5(a) and Fig. 5(b).

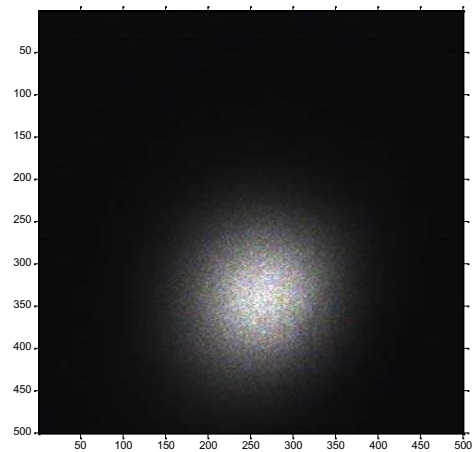


Fig. 5(a) Raw speckle image of elbow region

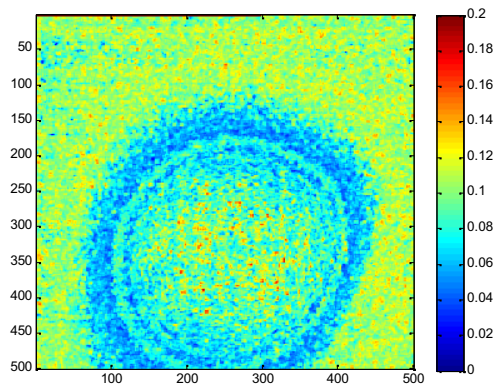


Fig. 5(b) False colour contrast map of image in Fig. 5(a)

The LDF recording at the finger tip of one of the subjects is shown in Fig. 6 and the LDF recording at the elbow region of one of the subjects is shown in Fig. 7.

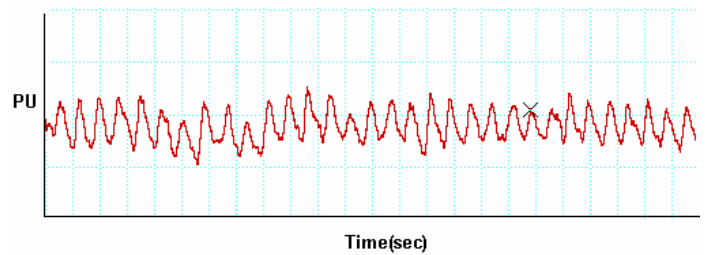


Fig. 6 LDF recording at the finger tip

TABLE I
LSCI AND LDF VALUES AT FINGER TIP AND ELBOW REGION FOR DIFFERENT SUBJECTS

Subject No.	Sex	Age	Height	Weight	BMI	LDF Elbow (PU)	Speckle Contrast(C) Elbow	Velocity Elbow(v _c) (µm/sec)	LDF Finger Tip (PU)	Speckle Contrast(C) Finger tip	Velocity Finger Tip(v _c) (µm/sec)
1.	Female	22	163	55	20.7008	27.4349	0.1027	8,296	451.8737	0.0770	14,758
2.	Male	27	167	70	25.0995	41.3426	0.1108	7,127	477.0368	0.0556	28,305
3.	Female	33	165	55	20.2020	59.8742	0.1084	7,446	493.9649	0.0458	41,714
4.	Male	23	171	62	20.7157	26.7309	0.1094	7,311	265.265	0.0650	20,710
5.	Male	27	176	74	23.8895	22.4430	0.0863	11,749	485.6186	0.0486	37,045
6.	Male	25	166	54	19.5965	45.3909	0.1045	8,013	371.901	0.0555	28,407
7.	Male	21	180	75	23.1482	19.1242	0.1231	5,774	196.3716	0.0751	15,514
8.	Male	21	174	59	19.4874	32.7487	0.0980	9,111	277.2915	0.0645	21,032
9.	Female	37	171	67	22.9130	29.4101	0.1246	5,636	572.672	0.1069	7,657
10.	Male	35	170	68	23.5294	29.2866	0.1161	6,491	780.3032	0.0571	26,837

PU = perfusion units, µm/sec = micrometer/sec

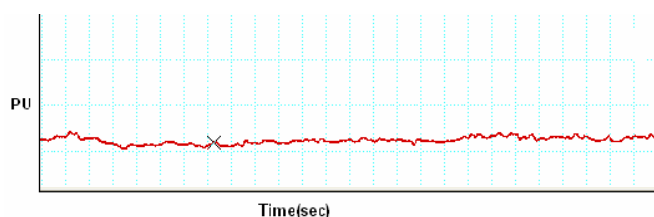


Fig. 7 LDF recording at the elbow area

TABLE I shows that there is an appreciable difference in the finger tip and elbow region blood flow values as obtained using both LSCI and LDF techniques. The LDF output is in perfusion units whereas the LSCI gives a more usable output in terms of quantitative values. The skin thickness is less in the finger tip when compared to that at the elbow and hence more blood flow is reflected at the finger tip. This difference in blood flow at the finger tip and the elbow region which is sensed by both the techniques can be put to good use in the follow-up of the treatment of certain diseases which may cause decreased blood flow in the body extremities such as the finger tips as in the case of Raynaud's syndrome.

V. CONCLUSION

Both the techniques namely LSCI and LDF are able to pick up changes in blood flow which may arise due to various physiological conditions. But the LDF output is in perfusion units which is an arbitrary unit while the LSCI is able to detect changes and give a more useful quantitative output.

The LSCI technique can be used to compare differences in blood flow values at the finger tip and elbow region in normal subjects and in diseased subjects as in the case of Raynaud's syndrome where the fingers or a particular finger is affected by reduced blood flow.

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