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COMPARISON OF LORENTZIAN AND GAUSSIAN BASED APPROACHES FOR LASER SPECKLE IMAGING OF BLOOD FLOW DYNAMICS

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ABSTRACT

Since blood flow is tightly coupled to the health status of biological tissue, several instruments have been developed to monitor blood flow and perfusion dynamics. One such instrument is laser speckle imaging (LSI). The objective of this work is to evaluate an LSI instrument employing two statistically based approaches to calculate the speckle flow index (SFI). To study the relation between SFI and the actual flow rate for the two statistical approaches, speckle images were acquired from a 0.5% blood filled tube embedded within a 5 mm thick agar gel. A syringe based infusion pump was used to inject the blood at flow rates between 0 and 5 mm/sec. We found a linear relationship between SFI and actual flow rate for both the Gaussian and Lorentzian based approaches. With the Gaussian based approach, the SFI dynamic range was up to six times larger than with the Lorentzian based approach. The Gaussian based approach is a good alternative for computation of SFI using LSI.

1. INTRODUCTION

Noninvasive monitoring of a therapeutic intervention is desired to provide the clinician or scientist with insight into the efficacy of the intervention. Since blood flow is tightly coupled into the health status of biological tissue, several instruments have been developed to monitor blood flow and perfusion dynamics, including laser Doppler flowmetry, Doppler ultrasound, Doppler optical coherence tomography, and laser speckle imaging (LSI).

LSI is a technique in which time-integrated speckle patterns generated by low power laser irradiation are imaged with a CCD camera. A single lowpass-filtering algorithm is used to convert raw speckle images to flow images. LSI has several advantages over existing methods, including simultaneous high spatial and temporal resolution, ease of implementation, and relatively low cost. LSI has been used to monitor noninvasively blood flow and perfusion dynamics in brain[1], retina[2], and skin[3,4]. Recently, we have employed LSI to monitor blood flow dynamics during photodynamic therapy[5] and have observed marked changes in the measured speckle flow index (SFI) values. The goal of this study was to evaluate an LSI instrument employing two statistically based approaches to calculate SFI. To achieve this goal, we employed an *in vitro* flow model.

2. METHODS

The LSI instrument consists of a 30 mW, 633 nm HeNe laser, plano convex lens, beam steering mirrors, digital CCD camera equipped with a macro lens, and desktop PC. The f-stop of the macro lens was set to ensure that the speckle size was equivalent to the pixel dimensions (6.45 x 6.45 μm^2) of the camera. To generate scattering agar gels, we heated a solution consisting of 100 mL deionized water and 10 mL glycerol to boiling. Glycerol was used to improve the mechanical integrity of the resultant gels. We added simultaneously 0.3 g TiO_2 and 2 g agar to the heated solution. The former was used to increase the scattering coefficient of the otherwise clear gels, and the mass added was deemed appropriate to simulate the reduced scattering coefficient of skin.

In specific gels, a ~550 μm inner diameter glass capillary tube was embedded into the mold prior to solidification. A syringe based infusion pump was used to inject fluid into the flow tube. Tygon tubing was used to deliver the fluid from the filled syringe mounted on the pump, to the tube embedded in the gel. 500 mL of indated whole blood was obtained from the San Diego Blood Bank and was used in this study. To achieve an experimental

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model similar to the rodent dorsal skinfold model used in the PDT study[5], we acquired images from a tube embedded within a ~5 mm thick agar gel. Since the vessels of interest in the PDT study are essentially at the surface of the skinfold, we positioned the tube so its upper wall was at the same level as the gel surface. The infusion pump was set to achieve flow rates between 0 and 5 mm/s. An integration time of 10 ms is similar to the 8 ms value used in the PDT study. In order to test both statistical approaches on an *in vivo* scenario, another set of experiments were developed on an *in vivo* chick chorioallantoic membrane (CAM) embryo .

The image processing algorithm has been described previously in detail[3]. Briefly, the recorded image sequence was converted to speckle contrast images by applying a 7 x 7 sliding window to each 1392 x 1040 image. At each window position, the mean graylevel intensity ($\langle I \rangle$) and standard deviation (σ) were determined, and the speckle contrast (K) of the center pixel in the window can be computed as[6]:

$$K = \frac{\sigma}{\langle I \rangle} \tag{1}$$

Assuming a Lorentzian profile:

$$\left|\gamma(t)\right| = e^{-t/2\tau} \tag{2}$$

or a Gaussian profile:

$$\left|\gamma(t)\right| = e^{-t^2/2\tau^2} \tag{3}$$

for the normalized autocorrelation function of the field, the correlation time (τ) of the intensity fluctuations can be calculated as:

$$K = \left[\frac{\tau}{T}\left(1 - e^{-T/\tau}\right)\right]^{1/2} \tag{4}$$

and

$$K = \left[\frac{\pi}{2}\frac{\tau}{T} erf\left(\frac{T}{\tau}\right)\right]^{1/2} \tag{5}$$

for the Lorentzian and Guassian based approaches, respectively; where T is the frame integration time and *erf* is the error function.

Maple™ software (Maplesoft, Ontario, Canada) was used to obtain an analytic series expansion for τ as a function of K for the Lorentzian based approach and Origin™ software (OriginLab Corp, Northampton MA, USA) was used to obtain a polynomial approximation for τ as a function of K for the Gaussian based approach. Relative flow images were obtained by calculating $1/\tau$ at each image pixel; a higher pixel value is assumed to be analogous to faster blood flow.

Since we were not concerned with real time speckle flow imaging in this study, we performed the image processing offline. We selected a central 260x260 μm^2 (i.e. 40 x 40 pixels) region of interest and computed the mean SFI.

3. RESULTS

Figure 1 shows the results of SFI vs actual flow rate over a physiologically relevant 0 to 5 mm/s range for the *in Vitro* experiment described above, employing whole blood as the flow fluid and an integration time of 10 ms.

From Figure 1, we can observe a linear relationship between the SFI and the actual flow rate for both the Lorentzian (R= 0.99) and Gaussian (R=0.98) based approaches, Moreover, with the Gaussian based approach, the SFI dynamic range was up to six times larger than with the Lorentzian based approach. Thus, our results suggest that the

Gaussian based approach for LSI image processing is superior to the standard Lorentzian based approach, for the flow rates used in this study.

Figure 2 shows the SFI results obtained from an *in vivo* CAM employing both statistical approaches with an integration time of 20 ms. With the Lorentzian based approach, it is difficult to distinguish the relative velocity between individual blood vessels; moreover, it is difficult to identify small vessels due to background noise generated by Brownian movement and/or embryo movement. However, with the Gaussian based approach, due to a larger dynamic range of the SFI, it is possible to identify clearly small and large blood vessels versus the background.

We believe that the differences between both statistical approaches may originate from:

1.- The model assumed for the velocity distribution.

2.- The somewhat arbitrary definition of τ (correlation time) for the Lorentzian and Gaussian approaches, and the inherent uncertainly generated from truncation of the series expansion (for the Lorentzian) and the polynomial fit (for the Gaussian) approximations employed to solve Eqs. 4 and 5 respectively.

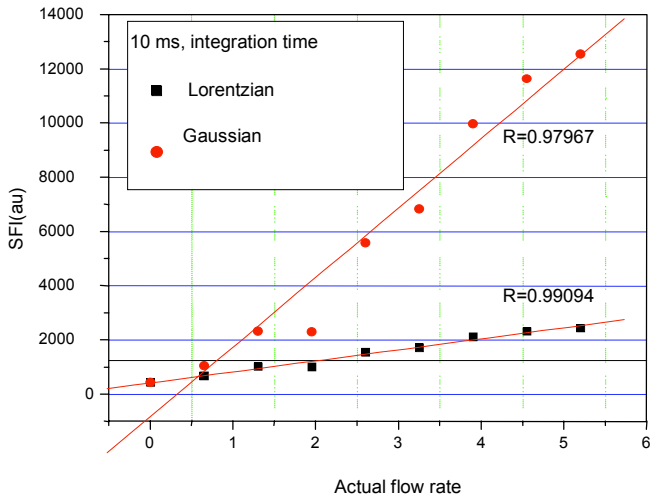
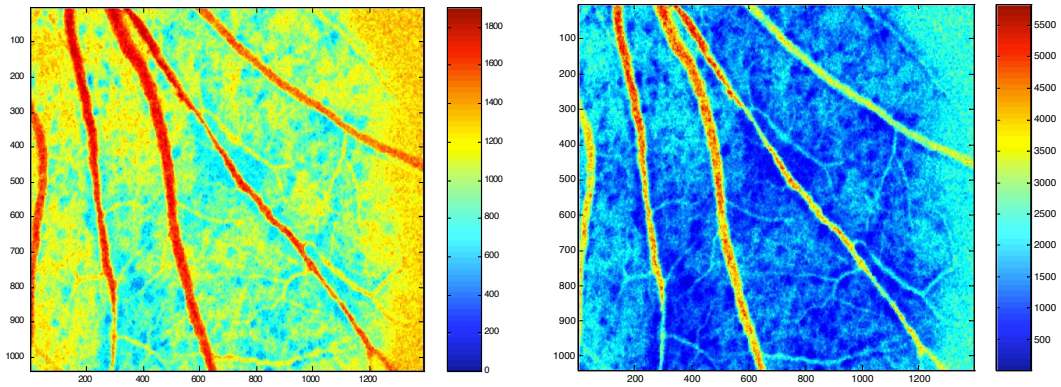


Figure 1. SFI versus actual flow rate over physiologically relevant flow values for Lorentzian and Gaussian based approaches.



(a) **(b)**

Figure 2. LSI images (8.7 X 9.3 mm^2) displaying vascular perfusion in an *in vivo* CAM model, processed with either the a) Lorentzian or b) Gaussian based approach.

4. CONCLUSIONS

Our results suggest that SFI in LSI images of superficial blood vessels (for example, in the rodent dorsal skinfold model and probably other *in vivo* vascular models), are linearly related to one another. Our results suggest that the Gaussian based approach for LSI image processing is superior to the standard Lorentzian based approach, for the flow rates used in this study.

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