Published in final edited form as:

Opt Lett. 2011 June 1; 36(11): 2143–2145.

Measurement of particle concentration in flow by statistical analyses of optical coherence tomography signals

Yi Wang and Ruikang K. Wang*

Department of Bioengineering, University of Washington, Seattle, WA 98195, USA

Abstract

We present a statistic method to measure the concentration of particles in flow by the use of optical coherence tomography (OCT). This method is based on the fact that the fluctuating numbers of particles contained within a small OCT detection volume caused by Brownian motion are governed by Poissonian distribution. Particle concentration is evaluated from the statistical analyses of the OCT intensity signals that are backscattered by the moving particles. The method is experimentally verified with flow phantoms of polymer microspheres. Experimental results are in good agreement with the theoretical ones for low concentrations while relatively large discrepancies emerge for high concentrations.

Over the past years, optical coherence tomography (OCT) has been developed as a powerful tool for depth-resolved measurements of blood flow in biological tissues. In this regard, the early developments were focused on quantifying of blood flow velocities based on the Doppler effect due to the back scattering from flowing blood cells, i.e. Doppler OCT [1–3]. Recent effort was paid to the use of OCT to visualize microvasculatures within scanned tissue volume without quantitative information [4–7]. Compared with visualization, quantitative information of blood flow is often required because such information is important for diagnostic and therapeutic purposes. In addition to flow velocity, another quantitative parameter of blood perfusion is the concentration of flowing particles, e.g. hematocrit. Compared with visualization, quantitative information of blood flow is often required because such information is important for diagnostic and therapeutic purposes in, for example, retinal diseases [8], neurological diseases [9], etc. However, in many cases, the concentration of particles in flow is often difficult to measure by noninvasive methods. In this letter, we present a noninvasive, quantitative approach for measuring absolute concentration of particles in flow by the statistical analyses of OCT signals. This method is based on the fact that the fluctuating numbers of particles contained within a small detection volume caused by Brownian motion are governed by Poissonian distribution [10,11]. When passing through the detection volume, particles cause relatively strong backscattered light and encode their moving information into OCT signals. Previously, we demonstrated an autocorrelation method for mapping transverse particle-flow velocities by estimating the dwell time of flowing particles crossing the probe beam [12]. In this letter, we focus on statistical analyses of the intensity of OCT signals, which directly reflects the fluctuations of the time-varying particle numbers contained within the detection volume. The fluctuations of OCT signals reflect the fluctuations of the particle numbers flowing through the detection volume. This method is analogous to fluorescence correlation spectroscopy (FCS) [10,11,13], which is widely used to deal with the intensity fluctuations of excited

fluorescence from fluorescent molecules. Chen et al. [14] demonstrated a photoacoustic counterpart of FSC using light-absorbing bead flow.

The detected temporal OCT interference fringe of an M-scan from a small detection volume at a depth of z is given by

$$I(z, t) = \sqrt{I_R I_S(z, t)} \cos(2knz - \varphi(z, t)) \quad (1)$$

where I_R and $I_S(z, t)$ denote the light intensities reflected from the reference mirror and back-scattered from the sample respectively, k is the wave number, n is the refractive index of the sample, z is the depth coordinate, and $\varphi(z, t)$ is the phase that depends on the path length difference between the reference and detection arms.

For non-flowing particle suspension, particles stochastically leave and enter the detection volume because of Brownian motion. The particle number within the detection volume at any time is governed by Poissonian distribution [10,11]. Therefore, the temporal average of the time-varying particle number $N(z, t)$ within the detection volume is given by:

$$\langle N(z, t) \rangle = \frac{\langle N(z, t) \rangle^2}{\langle (N(z, t) - \langle N(z, t) \rangle)^2 \rangle} \quad (2)$$

where $\langle \rangle$ denotes the temporal average. Suppose that I_0 is the average intensity of the light back-scattered by one particle within the detection volume, the time-varying light intensity $I_S(z, t)$ backscattered by the particles in the detection volume equals to $I_0 N(z, t)$. We arrive at,

$$\begin{aligned} \langle N(z, t) \rangle &= \frac{I_R^2 I_0^2 \langle N(z, t) \rangle^2}{I_R^2 I_0^2 \langle (N(z, t) - \langle N(z, t) \rangle)^2 \rangle} \\ &= \frac{\langle I_R I_S(z, t) \rangle^2}{\langle (I_R I_S(z, t) - \langle I_R I_S(z, t) \rangle)^2 \rangle} \end{aligned} \quad (3)$$

Equation (3) provides a straightforward evaluation of the particle number within the detection volume using the time-varying magnitudes of detected interference fringes, i.e. Eq. (1). Therefore, the particle concentration $C(z)$ equals $K \langle N(z, t) \rangle / V$, where V denotes the detection volume and K is a constant [11]. K/V can be experimentally determined by applying the present method to a particle flow with known concentration. Although deduced in non-flowing particle suspension, Eq. (3) is also suitable for flowing particle suspension because the temporal statistics of the light intensity fluctuations at one position are identical to the statistics of the spatial light intensity fluctuations [15]. This means that the average particle number calculated by Eq. (3) is independent of flow velocities.

The experimental setup used in this work is similar to that used in our previous work, which was a spectral domain OCT system [9]. Briefly, a superluminescent diode was used as the light source that has a central wavelength of 1310 nm and a bandwidth of 56nm, giving a $\sim 10\mu\text{m}$ axial resolution in tissue ($n=1.35$). In the detection arm, we used an objective lens with a focal length of 50mm to achieve a $\sim 16\mu\text{m}$ lateral resolution. A home-built spectrometer was employed to record interference fringes. The frame rate of the line scan camera was 47KHz. At each spatial location, we captured 4000 axial scans (A-lines) over ~ 85 milliseconds that represent an M-scan. We used particle flow phantoms of polymer microspheres (Latex Microsphere Suspensions, $0.3\mu\text{m}$, Duke Science) within a plastic tube (inner diameter ~ 1.2 mm) to test the present method. The flow was precisely controlled by a

syringe pump. The starting relative particle concentration in flow (0.5‰(w/w)) was set as 1, and it was then serially two-fold diluted to 1/2, 1/4, 1/8, 1/16, 1/32, 1/64, and 1/128, respectively. The tube was adjusted to be perpendicular to the detection beam. Eq. (3) was used to calculate the particle concentrations in flow phantoms.

Figure 1(a) shows the magnitude of the detected OCT signals of an M-scan from the fluid particle flow with an average flow velocity of 14.2mm/s (thin solid line), non-flowing particle suspension (dotted line), and pure water (thick solid line). It can be seen that the OCT signals from particle flow mainly depend on the light backscattered by particles, and flowing particles cause the stochastic fluctuations of OCT signals. Shown in Fig. 1(b) are the histograms of the detected OCT signals of the M-scan from a single position near the center of the tube with different relative concentrations of 1, 1/8, and 1/64 and velocities at 0 (i.e., non-flow), 3.55, 7.1 and 14.2mm/s, respectively. This result shows that the statistical distributions of the magnitudes of OCT signals are determined by particle concentrations, revealing the statistical characteristic of OCT signals from particle flow. In addition, it also shows that the statistical distributions are independent of flowing velocities. Consequently, the results from Eq. (3) are independent of flowing velocities. This agrees with the previous analysis.

The measured results of an M-scan with average velocities of 14.2mm/s, 7.1mm/s and 3.55mm are shown in Fig. 2, where the x-axis denotes the depth along the detection beam. From top to bottom, the theoretical relative concentrations are 1, 1/2, 1/4, 1/8, 1/16, 1/32, 1/64, and 1/128, respectively. The measured concentrations at different flow velocities are generally in agreement with each other along all the measured depth, especially for the concentrations below 1/8. This further indicates that the measured results are independent of flow velocities.

Data at the depth of 1.4 mm, as shown in Fig. 2 are plotted in Fig. 3. The dash line shows the theoretical relative concentration, i.e. $y = 2^x$. The experimental results are in agreement with the theoretical concentrations for low concentrations while relatively large discrepancies emerge for high concentrations. This discrepancy may be caused by shadowing or particle aggregation effect for high concentrations, which leads to the statistical divergence from Poissonian distribution. Therefore, Eq. (3) needs to be modified if high particle concentrations are presented in the flow. Not surprisingly, this limitation is similar to that in FCS [10]. This situation can be improved by the use of a lens with a high numerical aperture at the detection arm, which produces a relatively smaller detection volume. Although relatively large discrepancies between the experimental and theoretical concentrations emerge for higher concentrations, the results for higher concentrations are similarly determined by particle concentrations and are independent of flow velocities, as shown in Figures 1(b), 2 and 3. This indicates a modified Poissonian model could be employed for concentration measurements in particle flow with high concentrations to overcome the shadow effect and aggregation effect.

In conclusion, we have shown the feasibility of obtaining quantitative information as to the particle concentration by statistically analyzing the OCT signals. We demonstrated the statistics characteristic of the intensity fluctuations of OCT signals backscattered from particles in flow and proposed a statistical method for noninvasive concentration measurements based on the fact that the fluctuating numbers of particles within a small detection volume caused by Brownian motion are governed by Poissonian distribution. The method has been experimentally verified with polymer microsphere scattering flow, the results of which are in agreement with the theoretical concentrations for low concentrations while relatively large discrepancies emerging for high concentrations. Further study is

needed to modify the statistical divergence form Poissonian distribution for high concentrations.

Acknowledgments

This work was supported in part by research grants from the National Heart, Lung, and Blood Institute (R01 HL093140), National Institute of Biomedical Imaging and Bioengineering (R01 EB009682), and the American Heart Association (0855733G).

References

1. Yazdanfar S, Kulkarni MD, Izatt JA. High resolution imaging of in vivo cardiac dynamics using color Doppler optical coherence tomography. *Optics Express*. 1997; 1(13):424–431. [PubMed: 19377566]
2. Chen ZP, Zhao YH, Srinivas SM, Nelson JS, Prakash N, Frostig RD. Optical Doppler Tomography. *IEEE Journal of Selected Topics in Quantum Electronics*. 1999; 5(4):1134–1142.
3. Yang VXD, Gordon ML, Qi B, Pekar J, Lo S, Seng-Yue E, Mok A, Wilson BC, Vitkin IA. High speed, wide velocity dynamic range Doppler optical coherence tomography (Part I): System design, signal processing, and performance. *Optics Express*. 2003; 11(7):794–809. [PubMed: 19461792]
4. Wang RK, Hurst S. Mapping of cerebrovascular blood perfusion in mice with skin and cranium intact by Optical Micro-AngioGraphy at 1300nm wavelength. *Optics Express*. 2007; 15(18):11402–11412. [PubMed: 19547498]
5. Mariampillai A, Standish BA, Moriyama EH, Khurana M, Munce NR, Leung MKK, Jiang J, Cable A, Wilson BC, Vitkin IA, Yang VXD. Speckle variance detection of microvasculature using swept-source optical coherence tomography. *Optics Letters*. 2008; 33(13):1530–1532. [PubMed: 18594688]
6. Vakoc BJ, Lanning RM, Tyrrell JA, Padera TP, Bartlett LA, Stylianopoulos T, Munn LL, Tearney GJ, Fukumura D, Jain RK, Bouma BE. Three-dimensional microscopy of the tumor microenvironment in vivo using optical frequency domain imaging. *Nature Medicine*. 2009; 15(10):1219–1223.
7. Wang RK, An L. Doppler optical micro-angiography for volumetric imaging of vascular perfusion in vivo. *Optics Express*. 2009; 17(11):8926–8940. [PubMed: 19466142]
8. Aizu Y, Asakura T. Coherent Optical Techniques for Diagnostics of Retinal Blood Flow. *J. Biomed. Optics*. 1999; 4:61–75.
9. Calderon-Arnulphi M, Alaraj A, Slavin KV. Near infrared technology in neuroscience: past, present and future. *Neurol Res*. 2009; 31:605–614. [PubMed: 19660190]
10. Oleg Krichevsky and Grégoire Bonnet, Fluorescence correlation spectroscopy: the technique and its applications. *Reports on Progress in Physics*. 2002; 65:251–297.
11. Chen A, Hao J, Zhou Z, He K. Particulate concentration measured from scattered light fluctuations. *Optics Letters*. 2000; 25(10):689–691. [PubMed: 18064152]
12. Wang Y, Wang RK. Autocorrelation optical coherence tomography for mapping transverse particle-flow velocity. *Optics Letters*. 2010; 35(21):3538–3540. [PubMed: 21042342]
13. Rigler R, Mets Ü, Widengren J, Kask P. Fluorescence correlation spectroscopy with high count rate and low background: analysis of translational diffusion. *European Biophysics Journal*. 1993; 22(3):169–175.
14. Chen SL, Ling T, Huang SW, Baac HW, Guo LJ. Photoacoustic correlation spectroscopy and its application to low-speed flow measurement. *Optics Letters*. 2010; 35(8):1200–1202. [PubMed: 20410966]
15. Briers JD. Laser Doppler and time-varying speckle: a reconciliation. *J. Opt. Soc. Am. A*. 1996; 13(2):345–350.

Fig. 1.

(a) The magnitudes of the detected OCT signals from fluid particle flow with an average flow velocity of 14.2mm/, non-flowing particle suspension, and pure water. (b) The histograms of the detected OCT signals from the particle flow with different relative concentrations and different flowing velocities.

Fig. 2.

The measured relative concentrations of particle flow with the velocities of 14.2mm/s, 7.1mm/s, and 3.55mm/s. From top to bottom, the theoretical relative concentrations are 1, 1/2, 1/4, 1/8, 1/16, 1/32, 1/64, and 1/128, respectively.

Fig. 3.

The measured concentrations at the depth of 1.4mm, and the dash line shows the theoretical relative concentration, i.e. $y = 2^x$.