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Microvascular blood flow monitoring with laser speckle contrast imaging using the generalized differences algorithm

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ABSTRACT

Laser speckle contrast imaging (LSCI) is a full-field optical technique to monitor microvascular blood flow with high spatial and temporal resolutions. It is used in many medical fields such as dermatology, vascular medicine, or neurosciences. However, LSCI leads to a large amount of data: image sampling frequency is often of several Hz and recordings usually last several minutes. Therefore, clinicians often perform regions of interest in which a spatial averaging of blood flow is performed and the result is followed with time. Unfortunately, this leads to a poor spatial resolution for the analyzed data. At the same time, a higher spatial resolution for the perfusion maps is wanted. To get over this dilemma we propose a new post-acquisition visual representation for LSCI perfusion data using the so-called generalized differences (GD) algorithm. From a stack of perfusion images, the procedure leads to a new *single image* with the same spatial resolution as the original images and this new image reflects perfusion changes. The algorithm is herein applied on simulated stacks of images and on experimental LSCI perfusion data acquired in three different situations with a commercialized laser speckle contrast imager. The results show that the GD algorithm provides a new way of visualizing LSCI perfusion data.

Introduction

The monitoring of microvascular blood flow can be performed with several optical techniques, among which laser Doppler flowmetry (LDF) and laser speckle contrast imaging (LSCI) are now currently used (Humeau-Heurtier et al., 2013a). In LDF, the tissue under study (skin for example) is illuminated with a low-power laser light. The backscattered light is transmitted to a photodetector. LDF relies on the Doppler frequency shift that appears when light is scattered by moving blood cells (mainly red blood cells). The LDF perfusion is defined from the first moment of the power spectrum of the photocurrent fluctuations (see, e.g., Stern, 1975; Humeau et al., 2007). LDF signals have been the subject of many works (see, e.g., Refs. Stefanovska et al., 1999; Bernjak et al., 2011), but one of the major drawbacks of LDF is that it is a single-point measurement technique. Due to the spatial variations of the microvascular blood flow (Wardell et al., 1994; Braverman, 2000), the reproducibility of the measure is poor (Roustit et al., 2010a,b). Laser Doppler imaging (LDI) has been designed to prevent this drawback (Wardell et al., 1993). However, for most of the imagers, the image is computed by scanning the area under study which entails long recording times and prevents the monitoring of rapid physiological phenomena. To overcome the latter point, full-field laser Doppler imagers have recently been proposed (Serov et al., 2002, 2005; Serov and Lasser, 2005; Draijer et al., 2009a; Leutenegger et al., 2011; He et al., 2012a, 2013; Harbi and Thacher, 2013), but require high-speed cameras.

LSCI is another full-field optical technique for the monitoring of microvascular blood flow. It is a real-time method that does not need any scan and uses a normal CCD or CMOS camera (Ponticorvo and Dunn, 2004). LSCI relies on the following principle (Fercher and Briers, 1981; Briers and Webster, 1996; Briers, 2001, 2007; Forrester et al., 2004; Draijer et al., 2009b; Boas and Dunn, 2010; Basak et al., 2012; Senarathna et al., 2013; Briers et al., 2013): when the tissue under study is illuminated by a laser with an expanded beam, the backscattered light forms an interference pattern on the detector (a camera). Due to the phase difference involved in the backscattered light, there are constructive and destructive interferences. The latter produce a pattern composed of bright and dark areas on the camera. This pattern is known as a speckle pattern. When particles move in the tissue under study, the speckle pattern changes. In LSCI, the changing speckle pattern is recorded with a camera that has an integration time in the millisecond range. Due to the long integration time compared to the typical decorrelation time of the speckle pattern, the speckle pattern is blurred in the image. The level of blurring is quantified by the speckle contrast K that is inversely related to blood flow (see an

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example in Fig. 1). In the laser speckle contrast imager used in our work, the perfusion is computed from $1/K - 1$ (Perimed Documentation, 2009). It has been shown that the power spectral density measurements of the light fluctuations derived using LSCI and LDF techniques are equivalent (Thompson and Andrews, 2010).

LSCI data have shown to have excellent reproducibility (Roustit et al., 2010b; Rousseau et al., 2011; Puissant et al., 2013; Humeau-Heurtier et al., 2014). The rapid adoption of LSCI in clinical research is also probably due to the relative ease and low cost of building an instrument, compared to other techniques such as MRI or CT (Ponticorvo and Dunn, 2004; Mehrabian et al., 2013). The developments of the technique are again the subject of many studies (see, e.g., Duncan and Kirkpatrick, 2008a; Parthasarathy et al., 2008; Miao et al., 2009; Duncan and Kirkpatrick, 2008b; Duncan et al., 2008; Domoki et al., 2012; Yang and Choi, 2012; He et al., 2012b; Wang et al., 2013; Binzoni et al., 2013; Richards et al., 2013; Humeau-Heurtier et al., 2013b; Kazmi et al., 2014). LSCI is now used in many medical fields such as dermatology, cardiology, vascular medicine, diabetology, neuroscience, ophthalmology (Boas and Dunn, 2010), among others. In cardiovascular studies, LSCI can be used to analyze the impairment of tissue blood supply provoked by pathologies such as diabetes, Raynaud's phenomenon, or peripheral vascular diseases. Monitoring blood flow with LSCI can therefore allow early diagnoses or an evaluation of the evolution for such diseases. However, LSCI (as LDF and LDI) gives blood flow values in arbitrary units: no absolute values as ml $g^{-1}\cdot min^{-1}$ tissue are possible (Briers, 2001; Oberg, 1990), as again pointed out recently (see, e.g., Briers et al., 2013). Another drawback of the LSCI technique is that it leads to a large amount of data: the frequency sampling of the images can be of several Hz (depending on the application) and the recordings usually last several minutes. In order to monitor the blood flow, the clinicians have therefore a stack containing many images (perfusion maps) reflecting the temporal evolution of the blood flow on the area under study. To analyze the data easily and rapidly, one or several regions of interest (ROIs) are often chosen on the first image of the image stack, and an average of the blood flow is performed in this ROI. The same ROI is chosen on the other images of the stack and thus an average of the blood flow can be studied in this ROI in time, at the rate of the image frequency acquisition (Boas and Dunn, 2010; Rousseau et al., 2011). This leads to a 1D-signal reflecting the time evolution of the average value for the blood flow in the ROI (see an example in Fig. 2). In the same time, algorithms or methods to obtain a higher spatial resolution for the images are still proposed (see, e.g., Cheng et al., 2003; Yu et al., 2008; Liu et al., 2008; Miao et al., 2010). However, this spatial resolution is "lost" by the averaging procedure performed in the ROIs, in clinical routine, to evaluate the evolution of the blood flow in a simple and rapid way. To get over this dilemma we propose herein a new way of visualizing LSCI perfusion data from commercialized imagers. Our procedure uses the so-called generalized differences (GD) algorithm. This leads to the computation of a new *single perfusion image* reflecting by itself the *variations* of the blood flow on the whole images of the stack. Moreover, this new image has the advantage of possessing *the same spatial resolution as the original images*. Our work therefore corresponds to a post-acquisition perfusion image processing procedure. Our goal is not to propose an imaging pre-processing method on raw speckle images. The problem raised by large volumes of data when imaging blood flow is of importance. Moreover, the visualization of the blood flow data remains a challenging task (see, e.g., Borkin et al., 2011; Ropinski et al., 2009).

Among the algorithms that are often used in the evaluation of the activity for laser speckle images we find the Fujii's method (Fujii et al., 1985, 1987) and the temporal difference method (Tekalp, 1995). However, unlike the GD method (see below), the Fujii's algorithm does not take into account the difference between nonconsecutive images. This presents a drawback in the case of laser speckle perfusion evaluation as variations of perfusion are slow compared to the frequency of the image acquisition; the variations are therefore low between two consecutive images. Comparisons between perfusion images differing by short times are not reliable because this prevents a salient analysis of slow phenomena when a relatively short duration is studied (a few seconds). Moreover, as for the Fujii's method, the temporal difference method is not appropriated for microvascular blood flow characterization with laser speckle perfusion data because the differences between nonconsecutive perfusion images are not taken into account. Moreover, the temporal difference method leads to a stack of $N - 1$ difference images which does not correspond to our goal to obtain a new single image. These two methods have therefore been discarded from our work.

In what follows, we first present the GD algorithm. Afterwards, the proof of concept of our work and the measurement procedure used to acquire experimental laser speckle contrast images in different situations are described. The results obtained on simulated and experimental

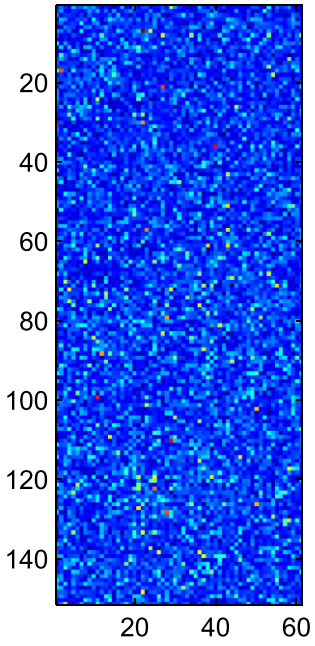

Fig. 1. Perfusion image (151 rows and 61 columns) from LSCI technique of a zone on the forearm of a healthy subject.

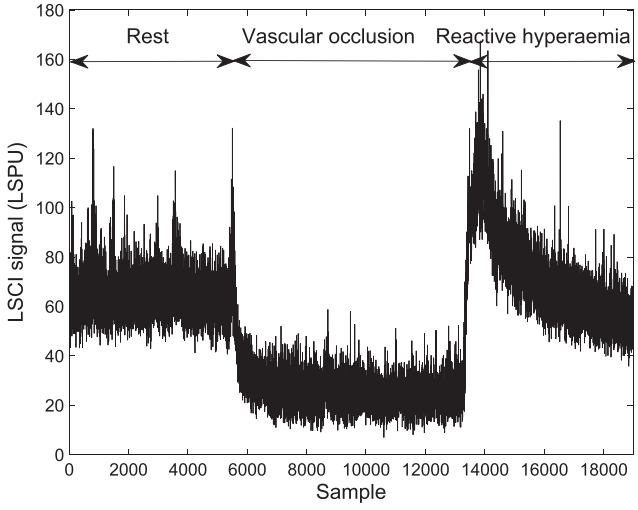

Fig. 2. Laser speckle contrast signal (perfusion signal) of a zone on the forearm of a healthy subject, at rest, during vascular occlusion and post-occlusive reactive hyperemia, computed from spatial averaging on a region of interest of 7×7 pixels^2 in laser speckle contrast images (perfusion images).