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Fast analysis of multiple exposure speckle data to provide relative blood flow maps using Convolutional Neural Networks

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ABSTRACT

Laser Speckle Contrast Imaging is a well-established technique able to produce relative blood flow maps contactless and without using dyes. It relies on the statistical analysis of dynamic speckle images, observed when a coherent light is used to illuminate a medium that contains moving scatterers. The local speckle contrast is related to the movements of the scatterers. Multiple exposure speckle imaging (MESI) is a variant of the technique that takes advantage of multiple exposure data to retrieve more quantitative flow maps by accounting for the unwanted and superimposed contribution of static scatterers. Yet, in MESI, a model is adjusted pixelwise to the experimental data requiring long computation times and an a priori guess on the flow regimes. These issues hindered so far, the translation of MESI to clinical applications though some studies have already demonstrated its potential. Here we propose an alternative method based on Convolutional Neural Networks to analyze MESI data. The proposed CNN architecture has been trained and validated using experimental data acquired on calibrated microfluidics flow phantoms. Then, the trained network was applied to analyze MESI data acquired in vivo in mice brain. In addition to be model-bias-free, we have found that the CNN approach infers flow maps much faster than the classical pixelwise regression approach. This new approach is promising for the clinical translation of MESI.

Keywords: Laser Speckle Contrast Imaging, blood flow, multiple exposure, Convolutional Neural Network.

1. INTRODUCTION

Laser speckle imaging (LSCI) has been used in the past decades to image the blood flow at the surface vessels of tissues either in small animal models or in clinical applications¹. Coherent light is used to illuminate biological tissues and the backscattered light produces an interference pattern, which is called speckle pattern. For living tissues, this pattern varies with time as some of the scatterers inside the tissue are moving (e.g. red blood cells in blood vessels). The method relies on the computation of the speckle contrast K, which was is related to the decorrelation time τ_c of the moving scatterers². Multiple exposure speckle imaging (MESI) was demonstrated to better account for the static scatterers contribution thus increasing the ability of the technique to estimate accurate changes of the flow over a wider range of flows³. However, this improvement of the initial LSCI technique came with a more complex instrumentation required to modulate the laser in both time and amplitude and the need to implement pixelwise non-linear fitting of the MESI-model to the data. This resulted in complex acquisition schemes and long post processing times to obtain the relative blood flow maps and ultimately hindered "real-time" imaging using MESI⁴. With the aim of producing faster acquisition and analysis of MESI data, several approaches have been implemented in the last decade. On the instrumental side, FPGA processing was used to acquire multiple exposure data with high temporal resolution⁵. Implementation of MESI set-up based on synthetic exposure without laser modulation was further demonstrated using a sCMOS camera and careful noise corrections⁶. Once the speckle contrast K images are obtained at several exposures T, the flow-related decorrelation time τ_c of the scatterers is obtained by fitting a mathematical model of K (T, τ_c). There are several issues with this approach: (i) the time need to process a single frame prevents "real time" imaging (ii) the MESI model usually relies on the hypothesis of a flow regime identical over the whole field of view, (iii) a limited robustness to noises. As an alternative, the use of machine learning has been proposed for MESI data analysis. The rationale is to bypass the non-linear fitting by using a neural network to directly infer the relative flow maps from MESI data. A first implementation using ANN trained with simulated data showed the validity of the approach and its in vivo validation^{7,8}. In the present study, we have implemented convolutional neural networks with architecture dedicated to the processing of MESI images. To better account for acquisition related parameters that are difficult to simulate, we have chosen to train the network using experimental data, acquired from calibrated microfluidic phantoms.

2. METHODS

MESI imager and microfluidic data

The MESI imaging system is composed of a 634 nm laser diode (Shangai laser, China) with a maximum output power of 400 mW and sCMOS camera (Orca Flash Hamamatsu, Japan). MESI data are acquired in the synthetic exposure approach with careful subtraction of noise contributions⁶. The microfluidic channels and set-up were described in details previously⁴. Speckle images used to train the network were recorded for microfluidic channels with a rectangular section of 40,60,80,100, 300 and 500 μ m of widths and 75 μ m of length⁹. We used 2% intralipid as the flowing media. Flows from 1 μ l.min-1 up to 7 μ l.min-1 (corresponding to velocities between 0.05 and 20mm.s⁻¹) were controlled using a pressure controller with inline flow feedback (Fluigent, France).

Convolutional Neural Network implementation

A residual 3D residual neural network (Res3D-Net) was implemented. The network model focuses on spatial features and the correlation information between MESI images of the same object acquired at different exposure times. The input is a stack of speckle contrast images at increasing exposure times. Images are further divided into patches (100 x 100 pixels) before processing by the network. Res3D-Net relies on 3D residual blocks to learn the spatial- exposure features. The first 3D residual blocks allow to extract features across multiple exposure speckle contrast images. At the end of the feature extractor, the generated features are then fed into three spatial-exposure residual blocks. We use a no padding strategy along the exposure time dimension to allow for a gradual extraction of neighboring exposure features from local to global information while preserving input distribution along the borders of the exposure-time dimension. The gold standard non-linear fit analysis was implemented as detailed previously⁴. It provided the decorrelation time τ_c for each pixel. As a metric of the relative flow changes, we used the inverse of τ_c usually coined ICT for inverse correlation time. For the analysis of *in vivo* data both techniques (CNN method and non-linear fitting method to the MESI model⁴) were compared on Python 3.9 using Intel Xeon Gold 6244 CPU @ 3.60 GHz 16/32 physical/logical cores.

In vivo experiments

All experiments were performed in accordance with the European Directive 2010/63/UE (Ethical project #CEB-03-2018) regarding the care and use of laboratory animals. MESI recordings were carried out in anesthetized mice (male, C57BL6, mean weight = 48 g) after a craniotomy was performed over the barrel cortex. A unilateral mechanical stimulation of whiskers protocol at 3 Hz was applied to visualize the blood flow change in the medial cerebral artery (MCA). A trial recording consisted of a baseline phase (10 s) followed by mechanical stimulation of the whiskers phase (30 s) and finally a rest phase (30 s). MCA images were acquired on the contralateral side of whiskers being stimulated.

3. RESULTS

We have implemented the CNN analysis of the MESI data and compared it with the gold standard non-linear pixelwise fit of the MESI model to the data. Figure 1 shows the ability of the CNN approach to provide blood flow maps for the microcirculation at the surface of the exposed brain in mice. Such map was obtained in less than one second, a processing time several order of magnitudes faster than that of the non-linear fit approach for the same data. On figure 2, we show the comparison of the analysis of CNN and non-linear fit data obtained during functional stimulation of the barrel cortex. Figure 2A shows the ROI drawn over the vessel. This was used to calculate the mean value for each frame for ICT or velocities computed by each method and showed on figure 2B and D respectively. Both approaches show (i) a moderate increase of blood flow after whiskers stimulation onset and then return to baseline after stimulation ceases (ii) similar temporal dynamics (iii) lower amplitude of the changes observed after CNN analysis compared to non-linear fit.



Figure 1. Blood flood map obtained at rest using the MESI-CNN analysis.



Figure 2. Comparison of non-linear and CNN approach analysis do MESI data acquired during functional stimulation of the barrel cortex. A) anatomical image of the vasculature obtained under green illumination and ROI used to produce B) and C. B) Time course of relative changes in ICT during C) Time course of relative changes of velocities

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