

# Analysis of images from optical coherence tomography for the nanoparticle transport

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## Abstract

This paper describes an analysis of contrast image sequences from optical coherence tomography. The application of bare nanoparticles in unique single fibre flow phantom is performed in order to examine their spatial and temporal behaviour inside the fibre and the wall of the fibre. The speckle variance method is used to show its usefulness in analysis of contrast OCT sequences.

## 1 Introduction

Optical coherence tomography (OCT) is relatively new imaging modality, which combines microseconds temporal resolution and micron spatial resolution. It enables to study optical properties (local reflectivity) of tissues and other samples with high dynamic range [3]. OCT was introduced in 1991 and since then a number of enhancements, modifications and applications have been presented [2]. The applications cover different fields of clinical, preclinical and experimental research ranging from ophthalmology, intracoronary imaging small animal experiments and nanoparticle research [1, 7]. OCT technique enables to evaluate both, the morphological and functional properties of examined samples/tissue. The functional OCT (fOCT) imaging is an important tool for investigating the processes in tissues of in vivo and in vitro systems. It includes different aspects of tissue/organ function, which can be divided into the contrast imaging approaches using bare nanoparticles, molecular imaging using target nanoparticles and hemodynamic or flow imaging. These categories are of course closely connected.

In this paper, we examine the behaviour of nanoparticles in capillary-simulating phantom via image processing methods. We describe application of nanoparticles for flow visualization and for their transport through semi-permeable membrane in custom-made single-fibre flow phantom.

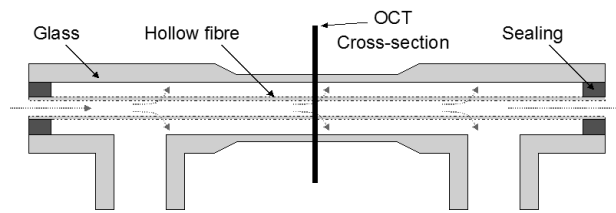


Figure 1: Scheme of the single-fibre flow phantom.

## 2 Method

### 2.1 Data acquisition on single-fibre phantom

In 2013 we presented the construction of a perfusion flow phantom for OCT [6]. Advanced construction of this single fibre phantom has been used for the purpose of this experiment (Figure 1). The polypropylene hollow fibres with pore sizes over  $200\ \mu\text{m}$  has been used ([www.zena-membranes.cz](http://www.zena-membranes.cz)). The fibre was placed in glass capillary with four openings. Two openings were used for setting the inner flow and other two openings for filling the glass capillary with water to ensure optical path without strong reflections. Two syringe pumps (NE-1010, New Era, USA) were used - the first pump (Pump 1) for setting the water flow rate (FR1) through the fibre and the second pump (Pump 2) for constant infusion of the nanoparticles by defined flow rate (FR2). The flow rate of Pump 1 has been constant ( $100\ \mu\text{l}/\text{min}$ ) during the whole experiment. The flow rate of Pump 2 was changing from  $100$  to  $500\ \mu\text{l}/\text{min}$  with  $100\ \mu\text{l}/\text{min}$  step. Therefore, with increasing FR2 the concentration of nanoparticles increases with ratio given by  $FR2/FR1$ . For each setting, one OCT sequence has been recorded with framerate 35 fps. The acquisition time was around 10s to ensure that steady state of nanoparticles flow occurs.

In this experiment, gold nanorods (Nanopartz Inc., A12N-25-1400) were used. They have plasmon-resonant peak matching the wavelength of our OCT system (Thorlabs, Swept Source OCT system, OCS1300SS, center wavelength 1325 nm). The diameter of these nanorods is  $25\ \text{nm}$  and length  $256\ \text{nm}$ . The concentration is  $0.05\ \text{mg}/\text{ml}$ .

### 2.2 Data analysis

The data were analysed in three ways:

1. We examined whether the spatial distribution of nanoparticles is homogeneous through the fibre cross-section or not. (Task 1)
2. We analysed whether there is some relation between the amount (concentration) of nanoparticles and image intensity or not. (Task 2)
3. We examined whether the nanoparticles penetrate through the fiber's wall or not. (Task 3)

For this purpose, we used analysis with speckle variance approach, which has been described in [4, 5]. Alternatively, we also examined the temporal changes of image intensity. We also compare these two approaches (speckle based and intensity based).

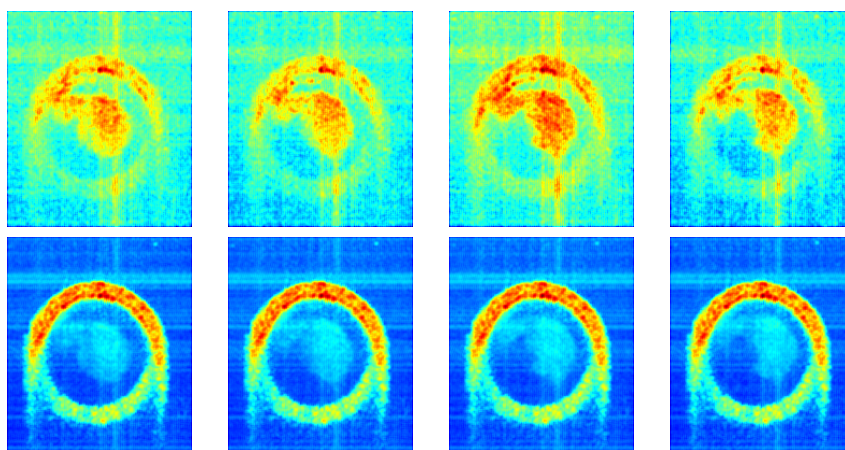


Figure 2: Four SV images (top row) and corresponding intensity images (bottom row) computed from the steady state for  $FR2 = 100 \mu l/min$  in different time intervals. Increased intensity inside the fibre shows presence of nanoparticles.

### 2.3 Speckle variance method

Here we briefly describe principle of the speckle variance (SV) optical coherence tomography method and its adaptation for our analysis. Let assume a sequence of OCT B-mode image  $I_i(j,k)$ , where  $j$  and  $k$  are spatial indexes in image matrix and  $i$  is temporal index. Inter-frame speckle variance between  $N$  images is computed as:

$$SV(j,k) = \frac{1}{N} \sum_{i=1}^N (I_i(j,k) - I_{mean}(j,k))^2, \quad (1)$$

where  $I_{mean}$  is an averaged image obtained from  $N$  frames. If we examine the flow or movement of some particles, the speckles differ from frame to frame and SV image thus represents this variations. It is assumed that there is no other movement (e.g. tissue or OCT probe) in examined scene or it is supposed that these tissue displacements between frames are less than the OCT beam radius. It is also supposed that other sources of noise are negligible (e.g. shot noise, electronic noise). The gate length  $N$  plays an important role. Authors of [5] used higher values ( $N=4$  or  $8$ ) for static scene and the minimum acceptable value ( $N=2$ ) for scene with possible movements. This value of course depends on current framerate. It is also suggested to averaged  $M$  consecutive SV images to improve image quality [5].  $M$  value depends on the properties of the scene. For static scene  $M$  value can be large and for dynamic scene smaller or even  $M = 1$ .

We changed the SV definition above by replacing the quadratic term by absolute value, in order to be able to represent the SV image in intensity levels. We also applied a spatial averaging filter ( $3 \times 3$  kernel) for smoothing the final SV image when examining the uniformity of nanoparticles distribution. This helps suppress noisy pixels and also slightly increases the contrast.

## 3 Results and Discussion

### Task 1

Several SV images computed at steady flow rate in different intervals (1 second) are shown

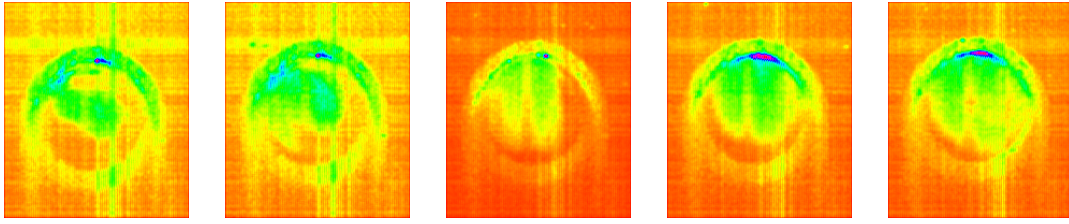


Figure 3: Five SV images computed from the steady state for (from left to right):  $FR2 = 100 \mu l/min$ ,  $FR2 = 200 \mu l/min$ ,  $FR2 = 300 \mu l/min$ ,  $FR2 = 400 \mu l/min$  and  $FR2 = 500 \mu l/min$ . Increased intensity inside the fibre shows occurrence of nanoparticles during 3 seconds long interval.

in Figure 2 (top row) for  $FR2 = 100 \mu l/min$ . Each image has been created from 16 frames (four frames were used for creating particular SV image and four SV images were averaged). Considering the temporal sampling ( $28ms$ ), each image can be examined as a cross-sectional flow during  $448ms$  interval. The same temporal window was used for creating the averaged intensity sequence (every 16 frames were averaged to create new image), see Figure 2 (bottom row). Highly non-uniform spatial distribution of nanoparticles can be seen in both sequences. It can be also seen that the contrast in the SV sequence was higher than in the average intensity sequence. Similar images can be obtained also for other FR2 values. This is shown in Fig. 3. Each figure represents mean speckle variance computed with  $N = 5$  and  $M = 21$ , which corresponds to 3 seconds interval. It can be observed that as FR2 increases (amount of nanoparticles is increasing and also the total flow rate increases), the nanoparticles flow became more spatially homogeneous.

#### Task 2

In the next step, we analysed relation between the amount of nanoparticles and image intensity for each FR2 values. Firstly we computed mean intensity inside the fibre using manually defined circular mask. The mean value has been computed in both, spatial and temporal domain to obtain one value representing the mean intensity (for intensity sequence) or mean SV value (for SV sequence). To analyse the influence of  $N$  and  $M$  parameters (see Section 2.2), the SV sequence has been computed for different values of these parameters. Figure 4 shows our results. We can see similar increasing trends for both, intensity and SV sequence. Saturation for high FR2 is clearly seen, which probably means that the concentration of nanoparticles is high and attenuation dominates over scattering. Figure 4b also shows influence of  $N$  and  $M$  on the result mean SV intensity. We can see that increasing  $N$  value increases also SV mean intensity, but the difference became low for  $N = 6$  and  $8$ . The multiple colour lines represents different numbers of averaged SV images used for creating the final SV image. We can conclude that this number doesn't significantly influence the SV values. We also computed correlation coefficient between FR2 and intensity or SV values, respectively for FR2 range  $100 - 400 \mu l/min$ . All values were above 0.95, which signifies high linear relation between these variables.

#### Task 3

In the last step of our analysis we evaluated the transport of nanoparticles through semipermeable membrane of the fibre. We evaluated the time course of the intensity and SV value in the whole sequence inside the fibre and within the wall of the fibre in a case of fast application of bolus of nanoparticles. The additional binary annular masks for this analysis have been defined manually.

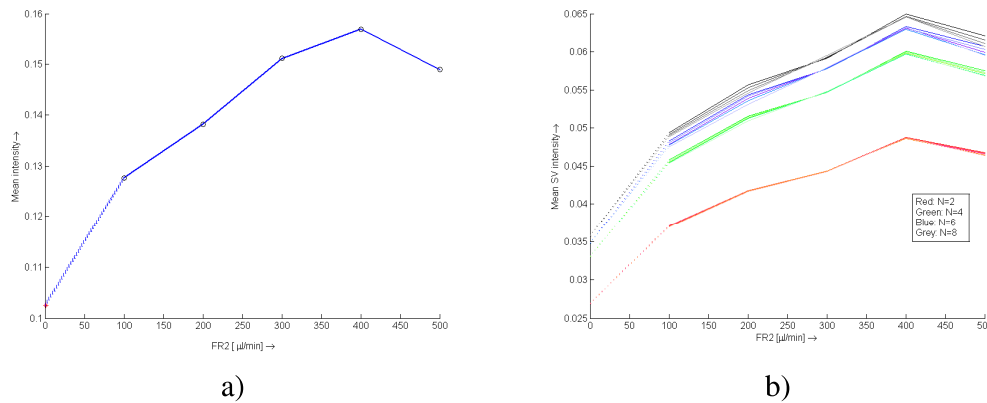


Figure 4: Curves showing the spatial and temporal mean intensity (a) and mean SV intensity (b) computed inside the fibre as a function of  $FR2$ . Value for  $FR2 = 0$  represents measurement only with water. The x-axis also represents concentration of nanoparticles as described in Section 2.1.

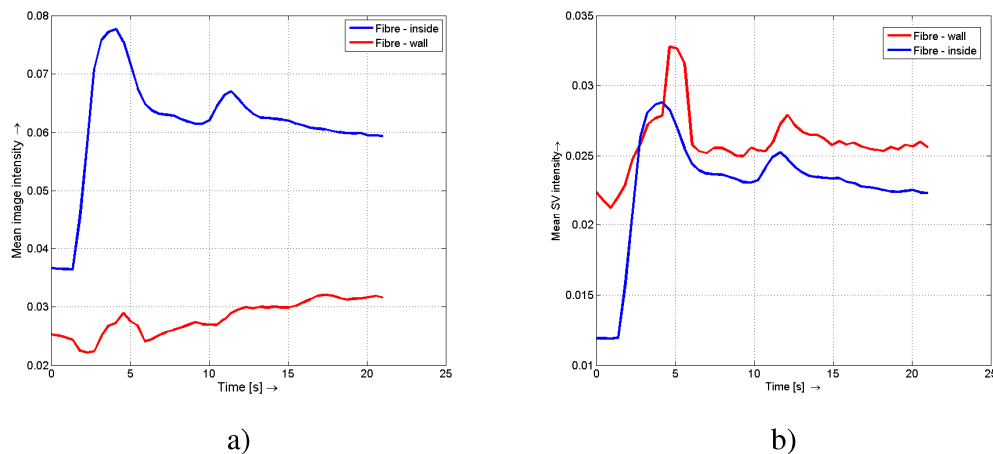


Figure 5: Time curves from inside (blue) and from the wall (red) of the fibre in fast bolus experiment for a) image intensity sequence and b) SV sequence.

Figure 5 shows these curves for mean intensity and mean SV sequences. Blue curves (with higher dynamic range) represents change in image intensity or SV intensity, respectively, inside the fibre during and after bolus administration. The first peak represents the maximum concentration achieved during administration of the bolus and the second peak probably represents the end of bolus as there is some pressure distortion in the circuit. It can be seen that the intensity and SV curves extracted from the wall of the fibre are different. The SV curve follows the shape of the curve from inside of the fibre with small delay (around 0.5 second) as can be expected. In the intensity curve, only the first delayed peak can be identified. The dynamic range of this curve is also smaller, which show limited sensitivity. The intensity changes in the fibre is very low, because the light reflections from this environment is high and superimpose the small variations of intensity. It must be noted that the values of this curve has been decreased (because the image intensity inside the wall is significantly higher) to be able to compare both curves.



## 4 Conclusion

We have shown the usability of speckle variance method for analysis of OCT sequences of scene containing nanoparticles. The SV approach achieved higher contrast in resulting images in comparison to intensity analysis (Task 1). SV is also useful for evaluation of the nanoparticle's movement through reflective environment (Task 3). We also observed a linear relation between concentration of nanoparticles in defined range, represented by FR2 (Task 2). It must be noted that SV method requires static scene, which can be difficult to achieve in some applications. However, frame-to-frame registration can be used for sequence stabilization. Another phenomena is signal attenuation, which decreases the intensity from lower depth and affects particularly the intensity based approaches of analysis. Nevertheless, several papers on this topic have been published, e.g. [8].

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## References

- [1] Lev Dykman and Nikolai Khlebtsov. Gold nanoparticles in biomedical applications: recent advances and perspectives. *Chemical Society reviews*, 41(6):2256–82, March 2012.
- [2] Liu Gangjung. *Selected Topics in Optical Coherence Tomography*. InTech, 2012.
- [3] Kirill V Larin, Joel D Morrisett, Mohamad G Ghosn, and Valery V Tuchin. Noninvasive functional imaging of tissue abnormalities using optical coherence tomography. In *2010 IEEE Sensors*, pages 2283–2286, November 2010.
- [4] Adrian Mariampillai, Beau a Standish, Eduardo H Moriyama, Mamta Khurana, Nigel R Munce, Michael K Leung, James Jiang, Alex Cable, Brian C Wilson, I Alex Vitkin, and Victor X D Yang. Speckle variance detection of microvasculature using swept-source optical coherence tomography. *Optics letters*, 33(13):1530–2, July 2008.
- [5] Adrian Mariampillai, Michael K K Leung, Mark Jarvi, Beau A Standish, Kenneth Lee, Brian C Wilson, Alex Vitkin, and Victor X D Yang. Optimized speckle variance OCT imaging of microvasculature. *Optics letters*, 35(8):1257–9, April 2010.
- [6] Petra Podlipná and Radim Kolář. Single fiber perfusion phantom for optical coherence tomography. In *European Conferences on Biomedical Optics*, page 88020Q. International Society for Optics and Photonics, June 2013.
- [7] Y. Ponce de León, J. L. Pichardo-Molina, N. Alcalá Ochoa, and D. Luna-Moreno. Contrast Enhancement of Optical Coherence Tomography Images Using Branched Gold Nanoparticles. *Journal of Nanomaterials*, pages 1–9, 2012.
- [8] K A Vermeer, J Mo, J J A Weda, H G Lemij, and J F de Boer. Depth-resolved model-based reconstruction of attenuation coefficients in optical coherence tomography. *Biomedical optics express*, 5(1):322–37, December 2013.