# Polarized laser Doppler perfusion imaging—reduction of movement-induced artifacts

## M. G. D. Karlsson K. Wårdell

Linköping University Department of Biomedical Engineering and the Competence Centre for Non-invasive Medical Measurements (NIMED) Sweden Abstract. Laser Doppler perfusion imaging (LDPI) enables superficial tissue perfusion assessment, but is sensitive to tissue motion not related to blood cells. The aim was to investigate if a polarization technique could reduce movement-induced artifacts. A linearly polarized laser and a cross-polarized filter, placed in front of the detectors, were used to block specular reflection. Measurements were performed with, and without, the polarization filter, at a single site during horizontal and vertical movement of skin tissue (index finger, twelve subjects, n=112) and of a flow model (n=432), with varying surface structures. Measurements were repeated during different flow conditions and at increased skin specular reflection. Statistical analysis was performed using ANOVA models. The perfusion signal was lower (p < 0.001, skin and p < 0.05, flow model) using the polarization filter, due to movement artifact reduction. No significant influence from surface structure was found when using the polarization filter. Movement artifacts were lower (p < 0.05) in the vertical movement direction, however, depending on flow conditions for skin measurements. Increased skin specular reflection gave rise to large movement artifacts without the polarization filter. In conclusion, the polarized LDPI technique reduces movement artifacts and is particularly appropriate when assessing, e.g., ulcers and burns, where specular reflection is high. © 2005 Society of Photo-Optical Instrumentation Engineers. [DOI: 10.1117/1.2120467]

Keywords: Laser Doppler perfusion imaging; skin microcirculation; flow models; movement artifacts; polarized light; specular reflection.

Paper 05057R received Mar. 1, 2005; revised manuscript received May 24, 2005; accepted for publication Jun. 14, 2005; published online Nov. 2, 2005.

# 1 Introduction

Laser Doppler perfusion imaging (LDPI) is a bio-optical technique that can be used to generate two-dimensional maps of superficial tissue perfusion.<sup>1,2</sup> LDPI has successfully been used to investigate microvascular blood perfusion in many applications, e.g., assessment of irritant skin reactions,<sup>3</sup> for evaluating treatment of basal cell carcinoma,<sup>4</sup> and for investigating perfusion in burn wounds.<sup>5</sup> Although the technique has found widespread use as a research tool for physiological studies, it has not been frequently used in the clinical setting on a daily basis. Partly due to technical reasons such as lack of possibility to estimate absolute perfusion values, influence of tissue optical properties on the perfusion estimate,<sup>6</sup> and the sensitivity to tissue motion not related to blood flow,<sup>7</sup> the added clinical value has not yet proven substantial.

In order to achieve an accurate LDPI estimation of the microvascular perfusion it is not only assumed but also a prerequisite that the Doppler broadening of the backscattered laser light is caused by scattering in moving blood cells only. If other tissue motion is present, caused by, e.g., patient movements during skin perfusion assessment or by the movement of the heart during studies of myocardial perfusion, an overestimation of the true blood perfusion is inevitable. When the tissue being assessed is moving relative to the LDPI system, both specularly reflected and scattered photons will become Doppler shifted and erroneously contribute to the perfusion estimate. A possible hypothesis is that the perfusion estimate, derived from the LDPI system during tissue motion, is generated by Doppler broadening of: specularly reflected light (Doppler<sub>surface</sub>), light scattered in the tissue (Doppler<sub>tissue</sub>), and light scattered by moving blood cells (Doppler<sub>blood cells</sub>). During well-controlled measurement conditions, i.e., without any tissue motion, Doppler<sub>surface</sub> and Doppler<sub>tissue</sub> are zero. By eliminating either of Doppler<sub>surface</sub> or Doppler<sub>tissue</sub>, or both, movement artifacts may be reduced.

The use of polarized light in bio-optical research has attracted significant interest in recent years. Jacques et al. have developed a system for imaging superficial tissue layers based on linearly polarized light and proposed that the system may be used to, e.g., identify skin cancer margins.<sup>8,9</sup> Yaroslavsky and colleagues used polarized light combined with either a multispectral dye-enhanced technique<sup>10</sup> or fluorescence<sup>11</sup> to study the possibility of delineating margins of different types

All correspondence to: MG Daniel Karlsson, Department of Biomedical Engineering, Linköping University, University Hospital, SE-581 85 Linköping, Sweden. Tel: +46-(0)13-22 24 96, Fax: +46-(0)13-10 19 02, E-mail: dakar@imt.liu.se

<sup>1083-3668/2005/10(6)/064002/9/\$22.00 © 2005</sup> SPIE

of nonmelanoma skin cancers. The above examples use polarization techniques to enhance backscattered light that has retained the polarization of the incident light, in order to visualize superficial tissue layers only. However, it is also possible to use polarization techniques to block backscattered and reflected light, which has retained its original polarization, from being detected.

Polarized light gradually loses its initial polarization state when being multiply scattered in tissue.<sup>12–15</sup> However, light being specularly reflected from the surface retains its original polarization state.<sup>16,17</sup> Inserting an analyzing polarization filter, with the polarization axis perpendicular to the polarization state of the incident laser light, in front of the detectors makes it possible to exclude the specularly reflected laser light from being detected, and still be able to detect photons scattered by moving blood cells.

The aim of this study was to investigate if the perpendicular polarization technique could be used to reduce the influence of movement-induced artifacts on the LDPI perfusion signal.

# 2 Material and Methods

## 2.1 Polarized Laser Doppler Perfusion Imager

The scanner head of an LDPI system<sup>2,18</sup> (PIM I, Lisca Development AB, Sweden) was modified in order to investigate possible differences in tissue motion influence, for polarized and nonpolarized detection. A linearly polarized He-Ne laser (C2-220T, 5 mW, 632.8 nm, Saven AB, Sweden) was used as a light source. An analyzing linear polarization filter (HN38, Neutral linear polarizer, Polaroid Corporation, Japan), with the polarization direction perpendicular to the polarization state of the laser light, was inserted in front of the detectors. The polarization filter could easily be removed from the scanner head, allowing measurements to be performed both with and without polarized detection.

## 2.1.1 LDPI signal generation and processing

Backscattered light impinging on the two photodetectors in the scanner head generated two signals: the photocurrent Doppler signal and the total backscattered light intensity signal (dc signal), formed by subtraction and addition of the two photocurrents, respectively.<sup>19</sup> The photocurrent Doppler signal was bandpass filtered (20 Hz–18 kHz, –3 dB), generating an ac signal. The ac and dc signals were sampled ( $5 \times 10^4$ samples s<sup>-1</sup>) and stored in a computer using a data acquisition board (DAQ-Card 6024E, National Instruments Corp., USA). Software developed in MATLAB<sup>®</sup> (MATLAB<sup>®</sup> 6.1, The MathWorks, Inc., USA) was used for off-line processing of the signals.

The discrete digital LDPI perfusion signal was calculated as the first-order moment of the ac signal power spectral density estimate, followed by noise compensation:<sup>2,20</sup>

Perfusion signal = 
$$\frac{1}{dc^2} \left( \sum_{\omega=F_1}^{F_2} \omega \cdot P[\omega] - V_{\text{Noise}}(dc) \right), \quad (1)$$

where  $P[\omega]$  is an estimate of the power spectral density, calculated from  $N_{\rm ac}$  points of the sampled ac signal,  $F_1$  and  $F_2$ are the lower and upper frequency limits, respectively, and  $V_{\rm Noise}({\rm dc})$  is a noise compensation function, describing the linear relationship between the detector noise and the dc signal level, established by calibration measurements on a stationary DELRIN<sup>®</sup> (polyacetal plastic) object. The dc<sup>2</sup> normalization and the noise compensation are performed using an average of the sampled dc signal from the  $N_{\rm ac}$  sample points.  $N_{\rm ac}$  was set to 512 samples during all analysis (giving one perfusion signal value approximately every 10<sup>th</sup> millisecond) and  $F_1$  and  $F_2$  were set to 100 Hz and 16 kHz, respectively.

## 2.2 Movement, Surface, and Flow Models

To generate a well-controlled movement, a setup consisting of a minishaker connected to an electrical power amplifier (Type 4810 and Type 2706, respectively, Brüel & Kjær, Copenhagen, Denmark) was used. The output from the electrical power amplifier (movement signal) controlled the position of the shaker. The movement signal was sampled ( $5 \times 10^4$ samples s<sup>-1</sup>) and stored simultaneously with the ac and dc signals, making it possible to correlate the LDPI signals to the movement of the shaker. By feeding the amplifier with a signal from a signal generator, the type of movement, the amplitude, and the frequency could easily be changed.

## **2.2.1** Flow model with varying surface structures

A flow model with varying surface structures and reflecting properties was created using a piece of DELRIN<sup>®</sup>. One half of it was prepared with sandpaper (granularity 80) and the other half with plexiglas polish (Fakopol, Röhm & Hass GmBH, Germany) in order to get a rough surface structure (denoted R) and a smooth surface structure (denoted S), respectively. The smooth surface gave rise to a large amount of specular reflection, compared to the rough one. Adding one layer of semitransparent tape (Scotch<sup>®</sup> Magic<sup>TM</sup> tape 810, 3M, USA) to the smooth surface created a smooth surface structure with a lower amount of specular reflection (denoted T).

Flow channels, parallel to the surface, were created by drilling a hole, 1.3 mm below each of the two surfaces (R and S/T), and inserting a tube (Polyethylene tube, ClayAdams, Division of Becton, Dickingson & Company, USA,  $\emptyset = 2 \text{ mm}$  and 1.6 mm, outer and inner diameter, respectively) through the holes [Fig. 1(a)]. Flow measurements were enabled by connecting one end of the tube to a syringe pump (M362, SAGE<sup>TM</sup>, Orion Research, Inc., USA), with a well-controlled flow. The DELRIN<sup>®</sup> piece could be attached to the minishaker in a way that enabled movement in two directions; horizontal and vertical (Fig. 1).

## 2.3 Flow Model and Skin Measurements

The scanner head was placed 15 cm above the measurement object and each measurement procedure, performed in darkness, lasted for 15 s. A sinusoidal shaker movement (amplitude 3.2 mm) was used and measurements were performed at four different shaker frequencies; 0, 1, 2, and 3 Hz, corresponding to an approximate maximum velocity of 0, 1, 2, and 3 cm s<sup>-1</sup>, respectively. All measurements were performed both with and without the polarization filter in front of the detectors and both in the horizontal and the vertical direction (Fig. 1).



Fig. 1 Measurement setup, showing an example of a flow model measurement during horizontal shaker movement (a) and a skin measurement during vertical movement (b). Skin and flow model measurements were performed both during vertical and horizontal movement.

# 2.3.1 Flow model measurements

Measurements on the flow model were performed for all three surface structures. Milk with 3% fat concentration was diluted (1:500) in water at room temperature, and perfused through the tube at nine different flow velocities ranging from approximately 0.6 to 6 mm s<sup>-1</sup> ( $v_1$ – $v_9$ ). The total number of measurements was 432 (three surfaces, with and without polarization filter, horizontal and vertical movement, four shaker frequencies, and nine flow velocities).

#### **2.3.2** *Skin measurements*

Twelve healthy Caucasian subjects (six male and six female, mean age 30, range 24–48), denoted A–L, were recruited for this study, which was approved by the regional Human Ethics Committee (Number 15-04). All subjects were nonsmokers and nondiabetic and were not allowed to drink beverages containing caffeine during the two hours immediately prior to the measurements. Before measurements were performed at the dorsal sides of the index fingers, proximal to the fingernail [Fig. 1(b)], these sites were inspected in order to ensure that no wounds were present. All subjects gave informed, written consent.

Before the initial measurements, the subject was acclimatized to the room temperature  $(24-25 \,^{\circ} C)$  during 15 min. To achieve maximal support and stability, a vacuum pillow was positioned around the lower arm of the subject. Measurements were performed both during a normal (unprovoked) blood flow state and, thereafter, at an occluded, blood-emptied state. The latter was accomplished by rolling several elastic rubber rings in proximal direction, from the distal end of the fingertip. Measurements were also performed after applying nondrying immersion oil (Type A, Nikon, Japan) to the skin, in order to mimic tissue surfaces with high specular reflection, e.g., ulcers and skin burns. Initial shaker movement direction and finger (right/left) for oil measurements were randomly chosen for each subject and kept constant throughout the experiment.

For normal blood flow, three measurements were carried out at each shaker frequency (1-3 Hz), both with and without the polarization filter. Initially a baseline recording without tissue motion was performed (I), thereafter a measurement during shaker movement (II), and finally a baseline measure-



**Fig. 2** Example of a skin recording, showing the mean perfusion signal level extending over a complete sinusoidal shaker movement period. This was formed by using the movement signal as a trigger and averaging the perfusion signal for all movement periods recorded during a 15-s measurement sequence. Corresponding tissue velocity is presented below. The arrows indicate the two maximum perfusion signal values during the reciprocating motion (one in each direction) that were averaged and used for statistical analysis, both with and without the polarization filter.

ment without shaker movement (III). Before changing shaker frequency and repeating measurements (I–III), there was a resting period of 2 min.

At the blood-emptied state, measurements were performed sequentially, without interruption according to: a baseline measurement without tissue motion, measurements during shaker movement (1-3 Hz), and finally a baseline measurement without tissue motion.

The total number of measurements for each subject was 112 (normal blood flow—blood-emptying, with and without polarization filter and immersion oil, horizontal and vertical movement direction, three shaker frequencies, movement and baseline measurements).

## 2.4 Data Analysis

An averaged perfusion signal value [mean and standard deviation  $(m\pm s.d.)$ ] was calculated using the entire measurement sequence of 15 s. Further, an averaged perfusion signal extending over a complete sinusoidal movement period was formed. This was achieved by using the movement signal as a trigger and averaging the perfusion signal for all movement periods recorded during the 15-s measurement sequence. Thereby, it was possible to relate the perfusion signal to the velocity of the measurement object (Fig. 2). The velocity of the measurement object during a movement period was calculated by differentiation of the movement signal.

Optical effects, caused by the insertion of the polarization filter in front of the detectors, changed the perfusion signal amplification. Therefore, a normalization factor ( $Perf_{polarization}/Perf_{no polarization}$ ) was calculated for each combination of the measurement conditions (oil, flow, surface) for each subject and in the flow model. This was performed by using all baseline perfusion signals available, without shaker movement. All perfusion signals recorded using the polariza-

tion filter were normalized with the corresponding normalization factor.

The averaged perfusion signal extending over a complete movement period was investigated at the highest shaker frequency (3 Hz), where the influence from tissue motion is at a maximum. The two maximum perfusion signal values during the reciprocating motion (one in each direction), without the polarization filter, were identified and averaged, resulting in one value for each measurement sequence of 15 s (Fig. 2). This value, together with the corresponding value recorded using the polarization filter calculated at identical tissue velocity, was used as an input to the statistical analysis.

## **2.4.1** *Statistical analysis*

The statistical analysis was performed according to full factorial ANOVA models, using MINITAB<sup>TM</sup> v. 13.32 (Minitab, Inc., USA) and Statistica v. 6.0 (StatSoft, Inc., USA). Flow model data was analyzed using the factors polarization (with and without the polarization filter), movement direction (horizontal or vertical) and surface structure (R, S, and T), with tube flow (v1–v9) as a block. For skin measurements, the factors polarization, movement direction, and flow (normal or blood-emptied), with an additive random effect for subject (A–L), were used.

Homogeneity of variances and normality was tested using the Brown-Forsythe modification of Levene's test,<sup>21</sup> considering the distances of the observations from their sample median rather than their sample mean, and the Ryan-Joiner test,<sup>22</sup> respectively. Residuals were plotted against all factors, variables, and fitted values, and were visually studied to reveal any patterns and relations.<sup>23</sup> Transformation of data was applied for the analysis of the flow model measurements, in order to fulfill ANOVA assumptions. A p-level <0.05 was regarded as significant. Pairwise comparison of group means was performed using 95% confidence intervals (double-sided, *t* distribution) with Bonferroni correction for multiple comparisons.

Oil measurements performed on the skin were excluded from the statistical analysis because this factor was difficult to control; a large amount of specularly reflected light hit the detector in some measurements, but not in others, leading to large variances in the measurements.

## **3 Results**

## 3.1 Flow Model Measurements

Flow model recordings showed significant main effects for all three factors and a significant (p < 0.001) three-factor interaction effect between polarization, movement direction, and surface structure.

Measurements with the polarization filter resulted in a lower (p < 0.05) perfusion signal. This was independent of surface structure and movement direction. Examples of perfusion signals, with and without polarization filter, for the three surface structures during horizontal movement, are presented in Fig. 3.

The three surface structures gave rise to different (p < 0.05) perfusion signal levels ( $Perf_S > Perf_R > Perf_T$ ) without the polarization filter, in the horizontal movement direction. With the polarization filter, however, no significant dif-



**Fig. 3** Influence of surface structure and reflecting properties on the averaged (all complete movement cycles during the 15-s measurement) perfusion signal, during horizontal movement of the flow model, at a tube flow velocity of  $2.7 \text{ mm s}^{-1}$ . The rough (R), smooth (S), and tape (T) surface structures are presented in (a), (b), and (c), respectively. Note the large influence of motion in measurements without the polarization filter (dependent on surface structure) compared to measurements with the polarization filter (independent of surface structure).



**Fig. 4** Perfusion signals recorded at the smooth surface (S) in the flow model, averaged during the entire measurement (15 s) at shaker frequencies 0 to 3 Hz, for nine different tube flow velocities (0.6 to 6 mm s<sup>-1</sup>). Measurements were performed during horizontal movement, (a) without the polarization filter and (b) with, and during vertical movement, (c) without the polarization filter and (d) with. Note the large influence of horizontal movement without the polarization filter and how movement artifacts are reduced by the polarization filter. Low influence of vertical movement was found.

ference was found (Fig. 3). In the vertical movement direction, no significant difference between any of the three surfaces was found. This was independent of the use of the polarization filter or not.

Vertical movement resulted in lower (p < 0.05) perfusion signal levels, compared to horizontal. This was independent of polarization and surface structure. Perfusion signals for surface structure S, calculated as an average of the entire measurement period (15 s), are presented in Figs. 4(a)–4(d). It includes measurements with and without the polarization filter at both horizontal and vertical movement (0 to 3 Hz) for different tube flow velocities. The figure illustrates the lower movement influence in the vertical direction, particularly evident without the polarization filter [Figs. 4(a) and 4(c)]. It also shows the effects of the polarization filter, reducing the movement influence, noticeable in the horizontal movement direction [Figs. 4(a)–4(b)].

## 3.2 Skin Measurements

Skin perfusion measurements showed significant main effects for all factors (polarization, movement direction, and flow) and a significant two-factor interaction effect between flow and movement direction.

Measurements with the polarization filter resulted in a lower (p < 0.001) perfusion signal level (7192±2272 a.u.) compared to without (8926±2983 a.u.), 95% confidence interval of the difference (a.u.): 955  $\leq$  Perf<sub>no polarization</sub> – Perf<sub>polarization</sub>  $\leq$  2513, because movement artifacts were reduced. This is exemplified in Fig. 5, showing a skin measurement performed with and without the polarization filter, during horizontal movement and normal blood flow, at three different shaker frequencies.

Vertical tissue movement resulted in a lower (p < 0.05)



**Fig. 5** Influence of skin tissue motion on the averaged (from all complete movement cycles during the 15-s measurement) perfusion signal, with and without the use of a polarization filter in front of the detectors. In this example, the perfusion signal was recorded during horizontal tissue motion at three different shaker frequencies, (a) 3 Hz, (b) 2 Hz, and (c) 1 Hz, at normal blood flow conditions.

perfusion signal compared to horizontal, 95% confidence interval of the difference (a.u.):

 $808 \le \text{Perf}_{\text{horizontal}} - \text{Perf}_{\text{vertical}} \le 3339$ . However, this was valid only in the blood-emptied case.

Blood-emptying resulted in a significantly lower signal compared to normal during vertical movement, 95% confidence interval of the difference (a.u.):

 $1587 \leq \text{Perf}_{\text{normal}} - \text{Perf}_{\text{blood-emptied}} \leq 4117$ . During horizontal movement, however, no significant difference was found. This can be explained by the large influence of tissue motion compared to blood flow on the perfusion signal [Fig. 5(a)].

Application of the immersion oil caused a large amount of light being specularly reflected backward to the scanner head. During movement, the perfusion signal level was substantially reduced by the polarization filter in cases when the specular reflection hit the detector [Fig. 6(a)], otherwise no effects were seen [Fig. 6(b)].

When specular reflection hit the detector, oil measurements gave rise to large movement artifacts compared to normal skin measurements.

## 4 Discussion

In this study, a possibility to reduce movement-induced artifacts in the LDPI perfusion signal by blocking specularly reflected light, utilizing a polarization technique, has been investigated. Measurements on the skin and in a flow model were performed during well-controlled movement of the measurement objects. The polarized LDPI technique reduced the perfusion signal levels significantly. At a tissue velocity of 3 cm s<sup>-1</sup>, the perfusion signal was strongly influenced by motion not related to blood flow [Fig. 5(a) and Fig. 6(a)]. Therefore, the lower signal levels found when using the polarization filter could be explained by reduced movement artifacts. This was supported by the similar results from the flow model measurements.

A possible explanation for the reduction of the movement artifacts is that photons reflected at the skin surface retain their original polarization,<sup>16,17</sup> thus being effectively blocked by the polarization filter. This prevents the formation of unwanted beat notes from appearing on the detector surface, because the Doppler-shifted photons carrying no blood flow-



**Fig. 6** Two typical examples (from different subjects) of vertical tissue motion influence on the averaged (from all complete movement cycles during the 15-s measurement) perfusion signal when immersion oil had been applied to the skin to increase specular reflection. (a) When the specularly reflected light hit the detector, a large influence from movement was registered. By inserting an analyzing polarization filter in front of the detectors (polarization perpendicular to incident laser light), movement influence was substantially reduced. (b) When specularly reflected light did not impinge on the detector, movement influence was substantially lower and similar both with and without the polarization filter.

related information cannot mix with randomly polarized backscattered photons. Hence, no influence on the detected photocurrent Doppler signal is obtained. These results support the hypothesis that movement artifacts in the perfusion signal can be reduced by blocking specularly reflected light from being detected.

Flow model measurements, without the polarization filter, confirmed previous findings of different magnitudes of movement-induced artifacts from varying surface structures.<sup>7,24</sup> Since the polarization filter blocks the specularly reflected light, the perfusion signal is expected to become unaffected by the surface structure and its reflecting properties. This was also confirmed in the flow model measurements.

Immersion oil was applied to the skin in order to mimic a shiny, smooth surface structure, found at, e.g., skin burns, ulcers, and internal organs. During tissue motion, the glare generated by the specularly reflected light gave rise to large movement artifacts when the reflection hit the detectors [Fig. 6(a)]. In these cases, the light intensity at the tissue surface was large at the site of the incident laser beam, as seen from the detectors, caused by the specular reflection. Thus, the light spot generated by the reflected and backscattered light was considerably different compared to normal skin. The number of speckle areas on the detector is dependent on the spot size;<sup>2,25</sup> smaller spot size means fewer speckle areas and larger amplification of the perfusion signal, possibly explaining the large signals registered. Similar results were found in the flow model measurements during horizontal movement, where the movement influence on the perfusion signals was largest for the smooth (S) surface structure, which gave rise to a large amount of specular reflection impinging on the detectors. Applying the polarization filter in front of the detectors, however, resulted in markedly reduced movement artifacts [Fig. 3(b) and Fig. 6(a)].

Even without tissue motion, the surface structure can influence the LDPI perfusion estimate due to the amplification effect of specular reflection. Hence, different readings for the same tissue blood perfusion could be recorded. Further, specular reflection has been reported to give rise to overestimated perfusion values during measurements on burn scars, caused by saturated total backscattered light intensity (dc signal) levels.<sup>26</sup> These false perfusion readings can be avoided by using polarized LDPI, an additional advantage besides reducing movement artifacts, thus increasing system usability and making LDPI more useful in the clinical setting.

However, by using the polarization filter not only specularly reflected light is blocked from being detected, but also approximately 50% of the randomly polarized, multiply scattered light. This results in a decreased signal-to-noise ratio if laser light intensity is not increased. The use of polarized light and a perpendicular analyzing polarization filter also affects the amplitude and interpretation of the perfusion signal, due to optical effects. Besides a changed tissue spot size, as seen from the detectors, the relation between homodyne and heterodyne contribution<sup>19</sup> is affected, since static specularly reflected and weakly scattered light are blocked from being detected.<sup>16,17</sup> Additionally, polarization may also affect the sampling volume.<sup>13–15,27</sup> Hence, perfusion readings from an ordinary LDPI system cannot be directly compared to readings from a polarized LDPI system. In this study, a normalization factor, dependent on the measurement object and measurement conditions, was used to compensate for these effects.

Horizontal and vertical movement is expected to influence the LDPI signal differently.<sup>7</sup> The size of the Doppler shift generated by a moving object equals  $\mathbf{q} \cdot \mathbf{v}$ , where  $\mathbf{v}$  is the scattering object velocity and  $\mathbf{q}$  is the scattering vector, the latter defined as the difference between the incident ( $\mathbf{k}_i$ ) and the scattered ( $\mathbf{k}_s$ ) light wave direction.<sup>28</sup> Assuming tissue movement at a single velocity in the vertical direction, when mixing of the light occurs at the surface of the square-law detector, no reference beam (unshifted light) is present.



**Fig. 7** (a) During tissue motion at a single velocity, incident light (**k**<sub>i</sub>) and backscattered light (**k**<sub>s</sub>), the latter impinging on the detector, define a scattering vector (**q**) distribution, which gives rise to a detected Doppler broadening. (b) Most **q** will have approximately the same direction as the incident light ( $\theta \approx 0$ ) due to the declining backscattered light intensity. During vertical tissue movement (**v**<sub>v</sub>) the detected Doppler broadening becomes smaller compared to during horizontal movement (**v**<sub>h</sub>), since  $\Delta$ (**q** · **v**<sub>v</sub>) <  $\Delta$ (**q** · **v**<sub>h</sub>) for small  $\theta$ , illustrated by the vertical and horizontal bars.

Rather, all light mixed at the detector has been Doppler shifted by the moving tissue, which means shifting all light slightly in frequency. This effect is cancelled out by the square-law detector, which explains why the small tube flow velocity variations can be detected despite the large shaker movement, resulting in a linear response for lower tube flow velocities (Fig. 4). If a static beam reference (632.8 nm) is introduced at the detector in this case, large movement artifacts would be registered.

A possible explanation to the different influence of horizontal and vertical movement is illustrated in Fig. 7. Differences in the direction of incident light  $(\mathbf{k}_i)$  and backscattered light  $(\mathbf{k}_s)$ , impinging on the detector, define a **q**-distribution, which gives rise to a detected Doppler broadening. The q-distribution depends on, e.g., tissue spot size, detector size, and the tissue-detector distance. Therefore, in a typical LDPI setup most scattering vectors are expected to have approximately the same direction as the incident light. During vertical movement  $(\mathbf{v}_{v})$ , the detected Doppler broadening becomes smaller compared to during horizontal movement  $(\mathbf{v}_{\rm h})$ , due to smaller variations of the projection of the different  $\mathbf{q}$  on the corresponding v [Fig. 7(b)]. Additionally, variations in surface structure at the site of the impinging laser light will give rise to a larger Doppler broadening during horizontal movement, making the difference even larger.

The perfusion signal in the flow model measurements was significantly lower during vertical, compared to horizontal, movement in all cases. For skin measurements, however, this result holds true only in the blood-emptied case. One explanation to this discrepancy may be that the tissue of the finger is compressible, unlike the DELRIN<sup>®</sup> piece. Therefore, in our movement model, the compression of the tissue may cause redistribution of blood cells influencing the blood flow at the measurement site.

## 5 Conclusions

This study has shown the possibility to reduce movementinduced artifacts in the perfusion signal by blocking specularly reflected light, utilizing a polarization technique. Polarized LDPI is particularly appropriate when applied to shiny tissues, such as internal organs, ulcers, and skin burns.

## Acknowledgments

This study was supported by the Competence Centre for Noninvasive Medical Measurements (NIMED). The authors would like to thank Eva Enqvist, Senior Lecturer, Department of Mathematics, Linköping University, for invaluable help and fruitful discussions regarding the statistical analysis and Håkan Rohman, Department of Biomedical Engineering, Linköping University, for skilful help when building the flow model.

#### References

- T. J. Essex and P. O. Byrne, "A laser Doppler scanner for imaging blood flow in skin," J. Biomed. Eng. 13 (3), 189–194 (1991).
- K. Wårdell, A. Jakobsson, and G. E. Nilsson, "Laser Doppler perfusion imaging by dynamic light scattering," *IEEE Trans. Biomed. Eng.* 40 (4), 309–316 (1993).
- A. Fullerton, B. Rode, and J. Serup, "Skin irritation typing and grading based on laser Doppler perfusion imaging," *Skin Res. Technol.* 8 (1), 23–31 (2002).
- A. M. Enejder, C. af Klinteberg, I. Wang, S. Andersson-Engels, N. Bendsoe, S. Svanberg, and K. Svanberg, "Blood perfusion studies on basal cell carcinomas in conjunction with photodynamic therapy and cryotherapy employing laser-Doppler perfusion imaging," *Acta Derm Venereol* 80 (1), 19–23 (2000).
- F. W. Kloppenberg, G. I. Beerthuizen, and H. J. ten Duis, "Perfusion of burn wounds assessed by laser Doppler imaging is related to burn depth and healing time," *Burns* 27 (4), 359–363 (2001).
- M. Larsson, W. Steenbergen, and T. Strömberg, "Influence of optical properties and fiber separation on laser Doppler flowmetry," *J. Biomed. Opt.* 7 (2), 236–243 (2002).
- M. G. D. Karlsson, M. Larsson, T. Strömberg, and K. Wårdell, "Influence of tissue movements on laser Doppler perfusion imaging," *Proc. SPIE* 4624, 106–114 (2002).
- S. L. Jacques, J. R. Roman, and K. Lee, "Imaging superficial tissues with polarized light," *Lasers Surg. Med.* 26 (2), 119–129 (2000).
- S. L. Jacques, J. C. Ramella-Roman, and K. Lee, "Imaging skin pathology with polarized light," J. Biomed. Opt. 7 (3), 329–340 (2002).
- A. N. Yaroslavsky, V. Neel, and R. R. Anderson, "Demarcation of nonmelanoma skin cancer margins in thick excisions using multispectral polarized light imaging," *J. Invest. Dermatol.* **121** (2), 259– 266 (2003).
- A. N. Yaroslavsky, V. Neel, and R. R. Anderson, "Fluorescence polarization imaging for delineating nonmelanoma skin cancers," *Opt. Lett.* 29 (17), 2010–2012 (2004).
- F. C. MacKintosh, J. X. Zhu, D. J. Pine, and D. A. Weitz, "Polarization memory of multiply scattered light," *Phys. Rev. B* 40 (13), 9342– 9345 (1989).
- J. M. Schmitt, A. H. Gandjbakhche, and R. F. Bonner, "Use of polarized light to discriminate short-path photons in a multiply scattering medium," *Appl. Opt.* **31** (30), 6535–6546 (1992).
- V. Sankaran, K. Schönenberger, J. T. Walsh, Jr., and D. J. Maitland, "Polarization discrimination of coherently propagating light in turbid media," *Appl. Opt.* 38 (19), 4252–4261 (1999).
- V. Sankaran, J. T. Walsh, Jr., and D. J. Maitland, "Comparative study of polarized light propagation in biologic tissues," *J. Biomed. Opt.* 7 (3), 300–306 (2002).
- R. R. Anderson, "Polarized light examination and photography of the skin," Arch. Dermatol. 127 (7), 1000–1005 (1991).
- S. P. Morgan and I. M. Stockford, "Surface-reflection elimination in polarization imaging of superficial tissue," *Opt. Lett.* 28 (2), 114–116 (2003).
- G. E. Nilsson, A. Jakobsson, and K. Wårdell, "Tissue perfusion monitoring and imaging by coherent light scattering," *Proc. SPIE* 1524, 90–109 (1991).
- G. E. Nilsson, T. Tenland, and P. Å. Öberg, "A new instrument for continuous measurement of tissue blood flow by light beating spectroscopy," *IEEE Trans. Biomed. Eng.* 27 (1), 12–19 (1980).

- G. E. Nilsson, T. Tenland, and P. Å. Öberg, "Evaluation of a laser Doppler flowmeter for measurement of tissue blood flow," *IEEE Trans. Biomed. Eng.* 27 (10), 597–604 (1980).
- M. B. Brown and A. B. Forsythe, "Robust tests for the equality of variances," J. Am. Stat. Assoc. 69 (346), 364–367 (1974).
- 22. J. T. A. Ryan and B. L. Joiner, "Normal probability plots and tests for normality," *Technical Report, Statistics Department, The Pennsylvania State University* (1976).
- 23. D. C. Montgomery, *Design and Analysis of Experiments*, 4th ed., John Wiley & Sons, New York (1997).
- M. Arildsson, G. E. Nilsson, and K. Wårdell, "Critical design parameters in laser Doppler perfusion imaging," *Proc. SPIE* 2678, 401–408 (1996).
- 25. A. Serov, W. Steenbergen, and F. de Mul, "Dynamic speckle patterns formed by a mixture of moving and stationary scatterers: theory and experiment," *Proc. SPIE* **3915**, 158–169 (2000).
- R. Bray, K. Forrester, C. Leonard, R. McArthur, J. Tulip, and R. Lindsay, "Laser Doppler imaging of burn scars: a comparison of wavelength and scanning methods," *Burns* 29 (3), 199–206 (2003).
  I. M. Stockford, S. P. Morgan, P. C. Chang, and J. G. Walker, "Analy-
- I. M. Stockford, S. P. Morgan, P. C. Chang, and J. G. Walker, "Analysis of the spatial distribution of polarized light backscattered from layered scattering media," *J. Biomed. Opt.* 7 (3), 313–320 (2002).
- G. E. Nilsson, E. G. Salerud, N. O. T. Strömberg, and K. Wårdell, "Laser Doppler perfusion monitoring and imaging," in *Biomedical Photonics Handbook*, T. Vo-Dinh, Ed., pp. 15:1–24, CRC Press, Boca Raton, FL (2003).