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Non-invasive monitoring of muscle blood perfusion by photoplethysmography: evaluation of a new application

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Abstract

Aim: To evaluate a specially developed photoplethysmographic (PPG) technique, using green and near-infrared light sources, for simultaneous non-invasive monitoring of skin and muscle perfusion.

Methods: Evaluation was based on assessments of changes in blood perfusion to various provocations, such as post-exercise hyperaemia and hyperaemia following the application of liniment. The deep penetrating feature of PPG was investigated by measurement of optical radiation inside the muscle. Simultaneous measurements using ultrasound Doppler and the new PPG application were performed to elucidate differences between the two methods. Specific problems related to the influence of skin temperature on blood flow were highlightened, as well.

Results: Following static and dynamic contractions an immediate increase in muscle perfusion was shown, without increase in skin perfusion. Liniment application to the skin induced a rapid increase in skin perfusion, but not in muscle. Both similarities and differences in blood flow measured by Ultrasound Doppler and PPG were demonstrated. The radiant power measured inside the muscle, by use of an optical fibre, showed that the near-infrared light penetrates down to the vascular depth inside the muscle.

Conclusions: The results of this study indicate the potentiality of the method for non-invasive measurement of local muscle perfusion, although some considerations still have to be accounted for, such as influence of temperature on blood perfusion.

Keywords blood flow, muscle blood perfusion, non-invasive, penetration depth, photoplethysmography, skin blood perfusion.

Blood flow monitoring may be performed on different types of human tissues and the techniques used are often related to the specific type of anatomy and vascularization. It has, until now, been difficult to measure local muscle blood perfusion directly in humans. Non-invasive techniques for total blood flow measurements include vs. occlusion plethysmography, which is based on determining the volume increase vs. time in a limb segment when venous outflow is arrested (McCully & Posner 1995), e.g. strain-gauge plethysmography. Blood flow in the limb can also be determined using ultrasound Doppler, which measures arterial inflow to a limb (Gill 1985). This technique is found applicably reliable on larger vessels, such as the femoral artery (Radegran 1997), but gives very limited information about local muscle blood perfusion. Laser-Doppler flowmetry (LDF) is mostly used for non-invasive measurement of the skin perfusion (Nilsson *et al.* 1980). However, further developments using the LDF technique has allowed blood perfusion to be measured invasively in local muscle tissue, as well. A fibre optic probe is inserted into the muscle and the blood flow is

expressed as the number of photons times the mean velocity of red blood cells (Salerud & Öberg 1987). Several studies have shown reasonable variations in muscle blood perfusion as a response to blood flow provocations (Kvernebo *et al.* 1990, Hoffmann *et al.* 1995, Jensen *et al.* 1995). Drawbacks of the method are, however, the trauma caused by the insertion of the optic fibre, which may affect the blood flow, and pain and discomfort experienced by the patient. The method is also prone to movement artefacts.

In photoplethysmography (PPG) light from a light emitting diode (LED) is directed toward the skin and this light is absorbed and scattered in the tissue. A small amount of this light is received by a photodetector placed, e.g. adjacent to the LED (reflection mode). Variations in the photodetector signal are related to changes in blood flow and blood volume in the underlying tissue (Challoner 1979, Kamal *et al.* 1989). The PPG signal consists of an AC and a DC component. The AC component is synchronous with the heart rate and depends both on the pulsatile pressure (Millasseau *et al.* 2000) and pulsatile blood volume changes (Babchenko *et al.* 2001). It has been suggested that the AC component is related both to pulsatile blood volume changes because of varying lumen of the vessel and pulsatile blood flow changes because of red cell orientation during each cardiac cycle (Graaf *et al.* 1993, Lindberg & Öberg 1993). The DC component of the signal varies slowly and reflects variations in the total blood volume of the examined tissue (Challoner 1979). Although it is generally accepted that PPG monitors changes in skin blood perfusion, there is no exact description of the relation between the signal and the blood flow. However, the commercial available PPG is inappropriate for monitoring deeper tissues, such as muscles. A method that non-invasively monitors variations in muscle blood perfusion without causing trauma or pain is preferable.

The aim of this study was to evaluate a specially developed PPG technique for simultaneous non-invasive monitoring of skin and muscle blood perfusion and to evaluate the ability of the method to discriminate between superficial and deep perfusion. There is no gold standard for measuring local muscle blood perfusion, and evaluation of the new technique was based on assessments of changes in blood perfusion in response to various blood flow provocations. In the present study the provocations included post-exercise hyperaemia and hyperaemia following the application of liniment. Simultaneous measurements using ultrasound Doppler and the new PPG application were performed, as well as experiments for demonstrating the deep monitoring feature of PPG. Moreover, specific problems related to the influence of skin temperature on blood perfusion were elucidated.

Subjects and methods

Subjects

Sixty-six healthy subjects were recruited to participate in the various trials. Details are presented below for each trial. All subjects were informed about the project and gave their informed consent. The ethical principles in the Declaration of Helsinki were followed and the local Ethics Committee approved the study.

Methods

A PPG instrument and a PPG probe (Department of Biomedical Engineering, Linköping University, Linköping, Sweden) were applied to continuously record changes in skin and muscle blood perfusion. The technique is based on the following considerations according to Figure 1, which schematically demonstrates the photon distribution in tissue containing both superficial and muscle compartments. A short distance between a green LED and a photodetector correlates to a short penetration depth because of the high absorption in blood (haemoglobin). In contrast, a long distance between a near-infrared LED and a photodetector enables a deep penetration depth, because the photons are multiple scattered in tissue (dermis, fat, collagen) at this wavelength and undergoes a lower absorption in blood. Specially designed optical probes, according to Figure 2a,b, were used in this study. Principally, a probe consisted of three photodetectors, six LEDs (green) and two or three LEDs (near-infrared) placed in a special pattern and embedded in black silicone rubber.

The optical probe, containing several LEDs and photodetectors, was developed to meet the following specifications:

- the pulsatile signal, reflecting the blood perfusion, is measured from a large area on the skin surface, which gives an integrated PPG signal from a large vascular volume. This integration partly compensates for spatial variations in blood perfusion (Tenland *et al.* 1983, Lindberg & Öberg 1991);
- the blood perfusion is monitored both superficially and deeper in muscle tissue making use of a special optical geometry and two different optical wavelengths;
- 560 nm, green light, and a centre-to-centre distance of 3.5 mm between LED and photodetector for monitoring blood perfusion at a depth of approximately 1–2 mm;
- 880 nm, near-infrared light, and a centre-to-centre distance of 20 mm between LED and photodetec-

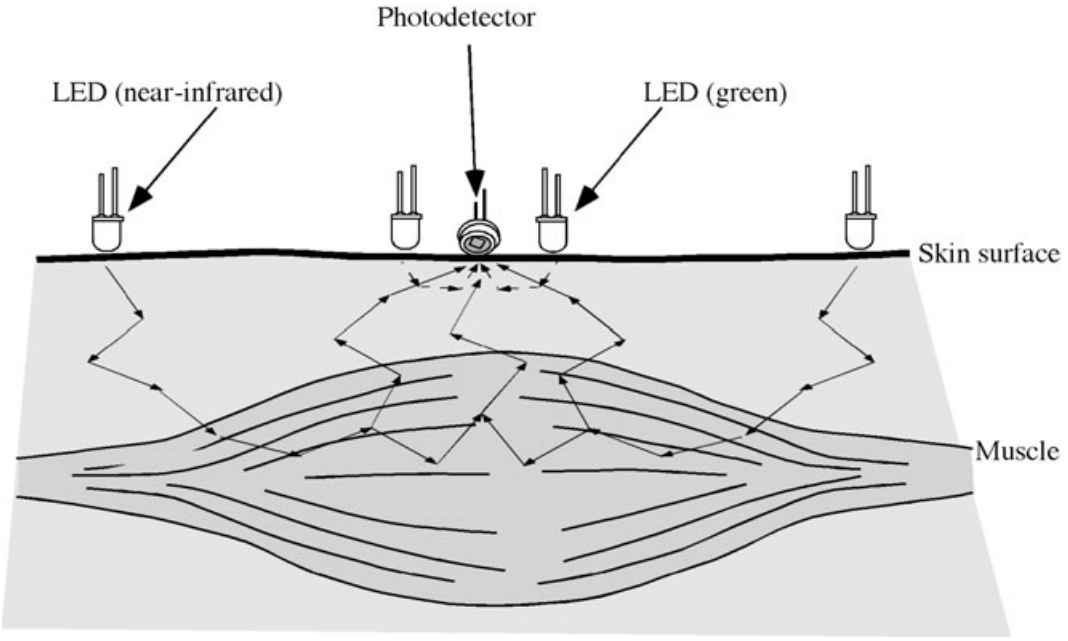

Figure 1 Schematic description of the photon propagation into tissue emerging from a green light and a near-infrared light source (LED) in combination with different distances between light source and photodetector.

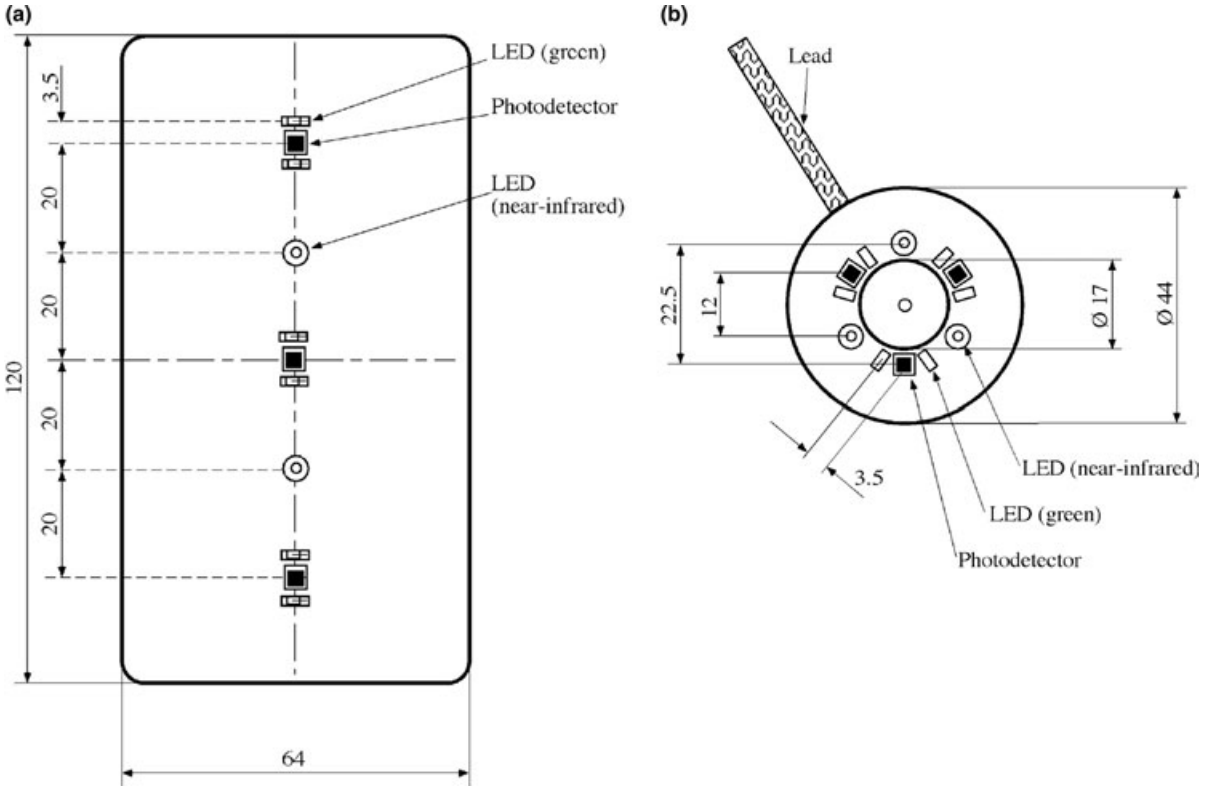

Figure 2 The rectangular PPG probe (a) and the circular PPG probe (b) used in this study.

tor (Fig. 2a), and 12 and 22.5 mm (Fig. 2b), respectively, for monitoring blood perfusion in muscle tissue.

Both signals from each wavelength are regarded as being simultaneously recorded because of a pulsed system. The green light source is on during approxi-

mately 0.2 ms followed by a light off for 1 ms, the near-infrared light illuminates then for 0.2 ms and is also off for 1 ms. This procedure is repeated 1000 times per second.

The signals from each wavelength were simultaneously processed in an amplifier and stored on a PC.

Provocation procedures

Different physiological procedures were applied in order to provoke the blood perfusion both superficially in the skin and deeper in the muscle tissue. Each procedure and corresponding aim is described in more detail below.

Static contraction. It is well known that the blood perfusion increases significantly in muscles after both static and dynamic exercise (Bangsbo & Hellsten 1998). A moderate static contraction of the muscle without a change in heart rate should result in a local enhancement of the muscle blood perfusion, but not a central one leading to changes in the skin perfusion too. To examine whether the new PPG application could detect changes in muscle perfusion in response to exercise, blood perfusion was measured in the biceps brachii muscle using PPG probe 2a in nine healthy males with a mean age of 26 years (range 20–32). The probe was attached to the skin above the muscle belly by adhesive tape, in close contact with the skin but without pressure. Each subject performed moderate static contraction in a sitting position with the forearm held horizontally with the elbow flexed 90° and supported by a hard pillow. After a resting period of 15 min in this position, the blood perfusion was measured for 60 s to establish the reference value. Thereafter, the subject performed a moderate static contraction in the biceps brachii muscle by holding a dumbbell (4 kg) in the hand for 60 s. Muscle blood perfusion was measured for 180 s directly after the contraction. The purpose of this experiment was to investigate the ability of the PPG instrument to separate changes in skin and muscle perfusion. During the post-exercise hyperaemia it was assumed that muscle blood perfusion would increase, but not skin perfusion.

Liniment application. Liniment (Transvasin®; Seton Products, Oldham, UK) was applied on the skin surface over the anterior tibial muscle in 16 healthy subjects (14 females, two males) with a mean age of 40 years (range 22–56) in order to increase the skin blood perfusion by vasodilator activity (Fulton *et al.* 1959). The active substances in Transvasin (tetrahydrofurfur salicylate, ethyl nicotinate and hexyl nicotinate) increase skin perfusion and has a dilating effect already after a few minutes. Probe 2b was adjusted for use of only one pair of LEDs (one LED of 560 and one of 880 nm and one photodetector). Blood perfusion was first measured for 60 s to establish the reference value. The probe was then removed and a minor amount of liniment (at the skin temperature of each subject) was quickly applied on the surface of the photodetector. After reapplication of the probe onto the same skin site, measurements of blood perfusion were continued for another 5 min. The purpose of this experiment was to investigate the ability of the PPG instrument to separate between skin and muscle blood perfusion by application of a substance known to induce skin vasodilatation.

In vivo determination of signal depth. The possibility that the photons may reach part of the muscle tissue is partly determined by the distance between the skin surface and the muscle location. This distance was measured at the middle of the leg, approximately 2 cm lateral to the anterior margin of the tibiae, in 43 subjects (30 females, 13 males) with the mean age of 45 years (range 21–65) using ultrasound Doppler.

The signal depth is defined as the depth from which some of the photons originate, and contribute to the PPG signal and this depth is determined as follows: after a local anaesthetic injection (1–2 mL of 1% lidocain) in the skin a catheter (32 × 1.2 mm) was inserted into the anterior tibial muscle at an angle to the skin surface of about 30° in three subjects. It was further advanced parallel to the muscle fibres as described elsewhere (Styf & Körner 1986). An optical fibre was then inserted into the plastic tube and connected to a Power Optical Meter for recording the radiant power in the muscle. The tip of the fibre was first positioned approximately in the middle of the muscle and then also closer to the skin surface. Exact location of the fibre tip was determined using Ultrasonics, by which the distance from the skin surface to muscle fascia and to fibre tip, respectively, was measured. The PPG probe 2a was placed over the anterior tibial muscle above the fibre tip and the multiple scattered photons from deep inside the muscle were measured using the Power Optical Meter.

Temperature dependence during long time recording. Skin and muscle blood perfusion was measured using probe 2b in 13 healthy supine females with a mean age of 39 years (range 21–55 years). The aim was to investigate the influence of temperature changes, originating from changes in the local environment as well as the local temperature regulation, on blood perfusion. After application of the probe over the anterior aspect of the tibia, measurements of blood perfusion were performed for 30 min and recordings of both skin and muscle blood perfusion were stored and analysed afterwards. A temperature sensor was positioned between the skin surface and the PPG probe. In a second step, four of the subjects participated in three

experiments where only one of the LEDs at a time was applied, either the green or near-infrared ones, or only the probe without any LEDs turned on.

Perfusion changes in the tibialis anterior muscle – a comparison with pulsed Doppler velocimetry. Muscle blood perfusion was measured in the tibialis anterior muscle, utilizing probe 2a in 10 subjects (five males, five females) with the mean age of 35 years (range 23–52 years). As a reference, peak femoral blood flow (FBF) was monitored at specific time points using pulsed Doppler velocimetry (ATL, HDI 5000, L 12–5; Philips, Bothel, WA, USA). Peak FBF was determined from the spectra of the pulsed Doppler ultrasound signal. A flat probe (operating frequency 4 MHz) was fixed to the skin over the vessel 2–3 cm distal to the inguinal ligament. Peak femoral flow velocity and artery diameter were measured after 1 min of full range- of motion dorsi- and plantar flexion of the ankle (i.e. dynamic contractions) in order to follow the post-exercise hyperaemic response for 5 min. From the flow velocity and artery diameter measures a blood flow value in mL s^{-1} was calculated.

Skin and muscle blood perfusion were simultaneously measured by application of the PPG probe over the anterior tibial muscle. This enabled a broad blood flow range in order to assess the relationship between blood flow measured using PPG and ultrasound Doppler (reference blood flow). The experiment was performed to elucidate specific differences and similarities between the two techniques.

Statistical analyses

Results are given as the mean (SE). The Wilcoxon signed rank non-parametric test was used for the statistical analyses. The mean values of blood flow changes are expressed as a percentage of initial 60s of resting values (0%). A *P*-value of <0.05 was considered significant.

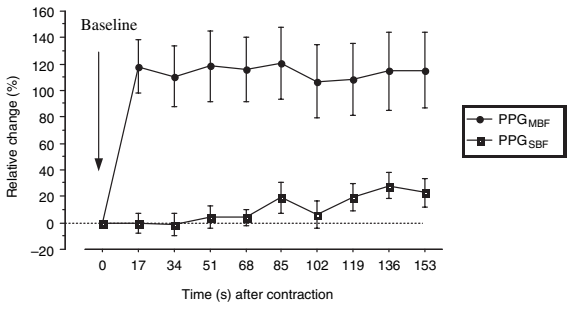

Figure 3 Relative changes in PPG_{SBP} (skin blood perfusion) and PPG_{MBP} (muscle blood perfusion) intermittently recorded for 60 s after static contraction. Values (mean ± SE) are given as the percentage of the initial resting value (0%).

Results

Static contraction of the biceps brachii muscle

Figure 3 shows relative changes in skin and muscle blood perfusion vs. time measured by PPG_{SBP} (skin blood perfusion) and PPG_{MBP} (muscle blood perfusion) after moderate static contraction in nine subjects. Muscle blood perfusion increased significantly ($P = 0.012$) using PPG_{MBP}. Skin perfusion, using PPG_{SBP}, did not change significantly ($P = 0.238$), although there was a small increase in skin blood flow at the end of the measuring period.

Liniment application

Figure 4 shows the relative change in PPG_{SBP} and PPG_{MBP} during 5 min after the application of liniment. During the first 2.5–3.1 min (time point 1) PPG_{SBP} increased significantly ($P = 0.001$) while PPG_{MBP} remained constant ($P = 0.281$). At the end of the 5-min period (time point 2) PPG_{SBP} had increased further, while only a minor increase was found in PPG_{MBP}.

In vivo determination of signal depth

The distance between the skin surface and the muscle fascia was in mean 5.4 mm (range 2.1–13.8). After insertion of an optical fibre into the muscle in three subjects, the radiant power was measured just underneath the PPG probe at two different fibre depths. Table 1 shows the recorded radiant power and the distances between the skin surface and the fascia and the fibre tip, respectively. When the LED in the probe was turned off the Power Optical Meter showed zero radiant power. Subject no. 3 also performed dynamic

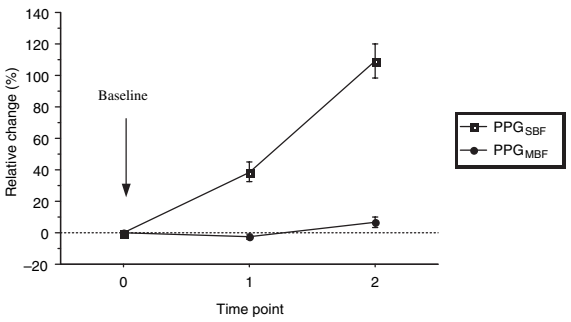

Figure 4 Relative changes in PPG_{SBP} (skin blood perfusion) and PPG_{MBP} (muscle blood perfusion) after liniment application. Time point 0 refers to rest before liniment application, time point 1 to a time range of 2.5–3.1 s after application, and time point 2 to approximately 5 min after application. Values (mean ± SE) are given as the percentage of the initial resting value (0%).

Table 1 Radiant power and distances between the skin surface and the fascia and fibre tip, respectively, in three subjects

Subject number	Radiant power (nW) measured in muscle	Distance (mm)	
		Surface-fascia	Surface-fibre tip
1	0.26	3.9	10.4
1	0.44	3.9	8.0
2	0.16	5.5	13.0
2	1.35	5.5	8.0
3	0.20	7.9	10.7

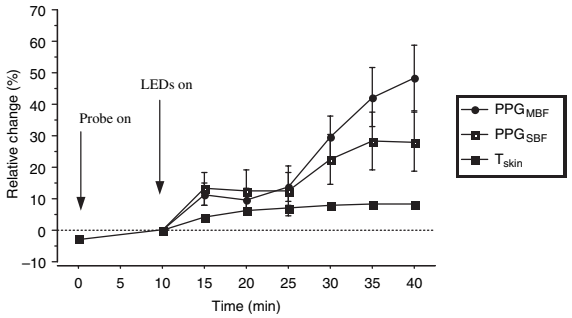

Figure 5 Relative changes in PPG_{SBP} (skin blood perfusion), PPG_{MBP} (muscle blood perfusion) and T_{skin} (skin temperature) in 13 subjects at rest during 40 min. Values (mean ± SE) are given as the percentage of the value at start of the recordings when the LEDs were turned on (baseline = 0%).

contractions for 60 s and the radiant power was then measured for 60 s after the release in order to follow the post-exercise response. The radiant power decreased by 12%, which indicates an increased blood volume in the muscle.

Temperature dependence during long time recordings

Figure 5 shows changes in percentage in skin and muscle blood perfusion and temperature, measured by PPG_{SBP}, PPG_{MBP} and T_{skin} during a resting situation in 13 healthy subjects. Application of the probe only gave rise to an increase in skin temperature during the initial 10 min. When the LEDs were turned on the temperature slope was further enhanced and both PPG_{SBP} and PPG_{MBP} exhibited an increase during 5 min. At approximately 25 min PPG_{SBP} and PPG_{MBP} started to increase further, whereas the temperature, T_{skin} flattened out more and more. This was also accomplished by a saturated level in PPG_{SBP}, but not in PPG_{MBP}.

In order to distinguish between the effects of 560 and 880 nm, experiments were repeated in four subjects by turning on the two types of LEDs separately. An increase was obtained in PPG_{SBP} when the green LEDs were turned on (Fig. 6a) and in PPG_{MBP} a decrease was shown when only the near-infrared LEDs were turned

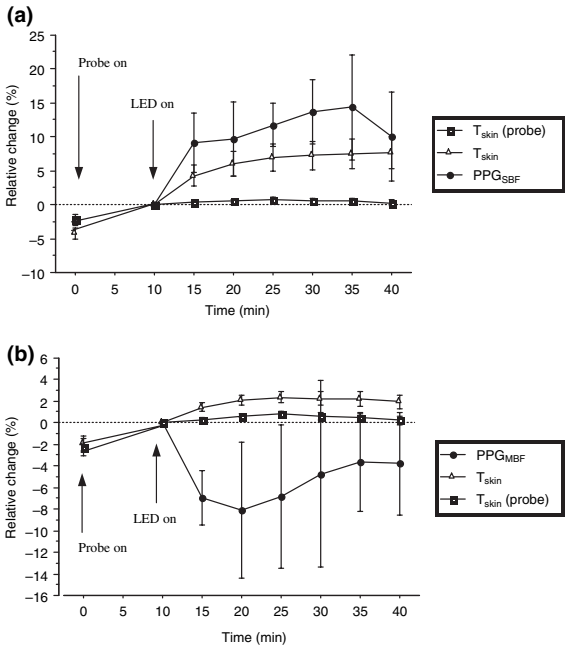

Figure 6 Relative changes in PPG_{SBP} (skin blood perfusion) and T_{skin} (skin temperature) when activating the green LEDs (a), and in PPG_{MBP} (muscle blood perfusion) and T_{skin} when activating the near-infrared LEDs (b) in four subjects. T_{skin} (probe) corresponds to skin temperature and application of the probe only, without activated LEDs (a and b). Values (mean SE) are given as the percentage of the value at start of the recordings when the LEDs were turned on (baseline = 0%).

on (Fig. 6b). The skin temperature rise, T_{skin}, and skin and muscle blood perfusion were more influenced by the green LEDs than the near-infrared ones. The application of the probe only, with no LEDs turned on, increased skin temperature T_{skin} (probe) initially, but from 10 min and onwards there was only a minor increase in skin temperature (Fig. 6a,b).

Perfusion changes in the tibialis anterior muscle – a comparison with ultrasound

Figure 7 shows relative changes in PPG_{SBP}, PPG_{MBP} and $Doppler_{FBF}$ (peak FBF) at specific time points directly after dynamic contractions and during the following 5 min. Similarly, PPG_{MBP} and $Doppler_{FBF}$ show a hyperaemic response at 5 and 25 s. Thereafter, $Doppler_{FBF}$ approaches the resting value, whereas PPG_{MBF} remains at a high level (~70%) compared with the resting value. PPG_{SBP} exhibits a decrease directly after the contractions and stays close to the resting value thereafter.

Discussion

This paper presents and evaluates a non-invasive technique for monitoring changes in pulsatile blood

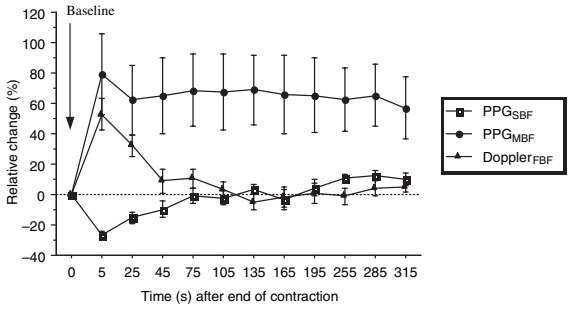

Figure 7 Relative changes in PPG_{SBP} (skin blood perfusion), PPG_{MBP} (muscle blood perfusion) and peak $Doppler_{FBF}$ (femoral blood flow) at specific time points for 5 min immediately after dynamic contractions. Values (mean ± SE) are given as the percentage of the initial resting value (0%).

perfusion in human muscle tissue utilizing PPG. The results indicate the potentiality of the method for monitoring local muscle blood perfusion, although some considerations still have to be accounted for and will be discussed below.

According to Figure 3, a post-contraction hyperaemic blood flow response after static contraction was measured as an increase in PPG_{MBP} but not in PPG_{SBP}. A moderate static contraction was chosen to reduce the influence of variations in heart rate and therefore the central circulation. These experiments clearly show this method's ability to discriminate the depth of blood perfusion when there is no change in the superficial blood perfusion. However, when there are also changes in the skin perfusion this will, to some unknown extent, also influence the measurement of deep blood perfusion.

The application of liniment on skin increased skin blood perfusion (PPG_{SBP}) almost immediately, while PPG_{MBP} remained unchanged (Fig. 3). The minor increase in PPG_{MBP} at the end of the measuring period is probably because of an influence of superficial blood perfusion on deep perfusion when PPG_{SBP} exhibits a substantial increase (here 100%).

The application of the probe enhances the temperature measured between the skin surface and the probe. This is probably explained by the inhibited convection, which makes transport of heat from the skin surface impossible. When the LEDs are turned on, the distributed heat gives rise to vessel dilatation and a corresponding increase in blood flow. The substantial increase in skin and muscle perfusion after 25 min is because of a temperature regulation effect (Svanes 1980), which is followed by a flattened temperature curve (Fig. 5).

Figure 6a demonstrates an increase in skin blood perfusion measured by PPG_{SBP}, when only the green light is on, which is a response to the increased heat at the skin surface. The turn on of only near-infrared light is followed by a decrease in muscle blood perfusion measured by PPG_{MBP} (Fig. 6b). This may be explained by the same heat increase in the superficial tissue mentioned above, but which gives rise to a local re-distribution of blood from the muscle tissue up towards the skin. These results also emphasize the sensitivity of the temperature regulation system, which in these tests enhances the muscle blood perfusion by approximately at first 10% as a response to impeded convection by the probe and later up to approximately 40% as a response to heat generation induced by the LEDs. This fact emphasizes the importance of reducing the generated heat by mainly the green light source. Technically, this is achieved by further decreasing the time the green light source is on in the pulsed system described in the Methods section. At present the heating influence is a limitation of the technique in long-term monitoring, extending 15 min, but not during short-term monitoring (see Figs 3, 4 and 7). Overall, a resting period of approximately 15 min after probe application and power on seems to be a good procedure before experiment starts.

As shown in Table 1 the radiant power measured in the muscle tissue indicates that the near-infrared light penetrates down to at least a vascular depth of 13.0 mm from the skin surface in these three subjects. This was demonstrated by zero indication on the Power Optical Meter with the LEDs turned off, but by indication (Table 1) with the LEDs turned on. Feng *et al.* (2001) also discussed the limitation of light penetration by the influence of overlying tissue on the signal quality in pulse oximetry, which also utilizes PPG.

A similar hyperaemic response directly after dynamic contractions was measured both by PPG_{MBF} and ultrasound Doppler (Fig. 7). However, thereafter the relatively high level in PPG_{MBF} demonstrates an enhancement of local muscle blood perfusion, which is not monitored by the Doppler technique in the femoral artery.

In medical science there is a need to monitor different physiological parameters from deeper laying tissues and vessels. Near-infrared spectroscopy (NIRS) is used, e.g. to extract information about tissue oxygenation from deeper vascular compartments in both cerebral tissue (Owen-Reece *et al.* 1999) and muscle tissue (Boushel & Piantadosi 2000). Recently, a study presented the first attempt to measure foetal arterial blood oxygenation using pulse oximetry (Zourabian *et al.* 2000). The preliminary results showed that the foetal pulse could be discriminated from the maternal pulse assuming a distance between the skin surface of the mother and the foetal head of 15–30 mm. As this method also utilizes arterial pulsations from deeper laying tissue it shows similarity with the method described in this paper.

Loukogeorkakis *et al.* (2002) have during pulse wave velocity measurements demonstrated the ability to

obtain PPG signals (wavelength 880 nm) from larger arteries, such as the radial artery. Also, a new PPG probe (wavelength 880 nm) designed for measurement of ankle systolic blood pressure was based on blood flow related signals from the anterior and posterior tibial arteries (Jönsson *et al.* 2005).

It has earlier been demonstrated (Fridolin & Lindberg 2000) that the possibility to image veins at different depths in human skin depends on the distance between the light source and photodetector. At a distance of 6 mm, veins located at 3 mm depth were imaged during a scanning procedure and using near-infrared light of 880 nm. Optical radiation in the green wavelength range, here 560 nm, is strongly absorbed in skin (pigment) and blood (e.g. haemoglobin). This makes it possible to utilize 560 nm for measuring skin blood perfusion (Lindberg & Öberg 1991, Hales *et al.* 1993, Futran *et al.* 2000). Based on their results, Hales *et al.* (1993) discussed the possibility to discriminate between different vascular beds by utilizing different optical wavelengths. They suggested that PPG (using a wavelength of 845 nm) and LDF (810 nm) should be used to measure local blood flow including arteriovenous anastomoses, and that light of a much shorter wavelength, probably green light, should be used to mirror capillary perfusion. Niklas *et al.* (1998) performed measurements with PPG in reflection mode on an *in vitro* model, containing a flexible tube and different surrounding materials such as water, agar–agar and muscle tissue. The centre-to-centre distance between the light source and the photodetector was 5 mm. They concluded that their equipment (wavelength 880 nm) measured blood perfusion mainly in the subpapillary plexus (3–5 mm) and that muscle perfusion could not be measured using their instrumentation. However, it must be stressed that they did not mention which muscle depths they were referring to and their conclusion about monitoring depths was based on *in vitro* experiments.

There is a great variability among subjects regarding distance from the skin to the muscle fascia. Skin and adipose tissue is considered as overlying tissue relative to the muscle. At the near-infrared wavelength used in this study (880 nm), the scattering is approximately the same for adipose and muscle tissue. However, the adipose is a weak absorption medium, while the muscle has strong absorption at 880 nm (Cheong *et al.* 1990, Graaf *et al.* 1993), partly because of higher blood vessel density in the muscle. This means that the light penetrating into the adipose undergoes a minor attenuation, which facilitates the possibility for optical radiation to reach the muscle belly. It has also been shown that increasing the distance between the light source and the detector improves the sensitivity for monitoring optical changes from the muscle in reflection mode, while increasing the adipose thickness decreases this sensitivity (Feng *et al.* 2001). This indicates that in order to monitor muscle blood perfusion such as in the anterior tibialis, the biceps brachhii, the triceps and the trapezius muscles one must find the best optical geometry for different tissue locations.

Monitoring of changes in muscle blood perfusion will enable new applications and possibilities in clinical diagnostics and also in therapy. Some of these new possibilities include:

- monitoring of muscle blood perfusion directly before and after different types of provocations for measuring the circulatory capability in the muscles;
- assessing the significance of muscle blood perfusion during increased intramuscular pressure, as in compartment syndromes;
- as a complement in the diagnostics of certain muscle pain conditions;
- assessing the effects of different treatment modalities aimed at increasing muscle blood perfusion.

In conclusion, the results of this study indicate the potential of PPG to non-invasively monitor intramuscular blood perfusion. Continuous monitoring for 10 min offers valid measurements, whereas continuous long-term monitoring (>15 min) is influenced by a heating effect from mainly the green LEDs. Technical improvements in order to reduce this influence are under way.

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References

Babchenko, A., Davidson, E., Ginosar, Y. *et al.* 2001. Photoplethysmographic measurement of changes in total and pulsatile tissue blood volume, following sympathetic blockade. *Physiol Meas* **22**, 389–396.

Bangsbo, J. & Hellsten, Y. 1998. Muscle blood flow and oxygen uptake in recovery from exercise. *Acta Physiol Scand* **162**, 305–312.

Boushel, R. & Piantadosi, C. 2000. Near-infrared spectroscopy for monitoring muscle oxygenation. *Acta Physiol Scand* **168**, 615–622.

Challoner, A. 1979. Photoelectric plethysmography for estimating cutaneous blood flow. In: P. Rolfe (ed.) *Non-Invasive Physiological Measurements: 1*, pp. 125–151. Academic Press, London.

Cheong, W., Prahl, S. & Welch, A. 1990. A review of the optical properties of biological tissue. *IEEE J Quantum Electron* **26**, 2166–2185.

Feng, W., Haishu, D., Fenghua, T., Jun, Z., Qing, X. & Xianwu, T. 2001. Influence of overlying tissue and probe geometry on the sensitivity of a near-infrared tissue oximeter. *Physiol Meas* **22**, 201–208.

Fridolin, I. & Lindberg, L.-G. 2000. Optical non-invasive technique for vessel imaging and monitoring I – experimental results. *Phys Med Biol* **45**, 3765–3778.

Fulton, G., Farber, E. & Moreci, A. 1959. The mechanism of action of rubefacients. *J Invest Dermatol* **33**, 317–325.

Futran, N., Stack, B. Jr, Hollenbeak, C. & Scharf, J. 2000. Green light photoplethysmography monitoring of free flaps. *Arch Otolaryngol Head Neck Surg* **126**, 659–662.

Gill, R. 1985. Measurement of blood flow by ultrasound: accuracy and sources of error. *Ultrasound Med Biol* **11**, 625–641.

Graaf, R., Dassel, A., Koeölink, M., de Mul, F., Aarnoudse, J. & Zilstra, W. 1993. Optical properties of human dermis in vitro and in vivo. *Appl Opt* **32**, 435–447.

Hales, J., Roberts, R., Westerman, R., Stephens, F. & Fawcett, A. 1993. Evidence for skin microvascular compartmentalization by laser-Doppler and photopletysmographic techniques. *Int J Microcirc: Clin Exp* **12**, 9–104.

Hoffmann, U., Uckay, I., Fischer, M., Wen, S., Franzeck, U. & Bollinger, A. 1995. Simulataneous assessment of muscle and skin blood fluxes with the laser-Doppler technique. *Int J Microcirc Clin Exp* **15**, 53–59.

Jensen, B., Sjogaard, G., Bornmyr, S., Arborelius, M. & Jorgensen, K. 1995. Intramuscular laser-Doppler flowmetry in the supraspinatus muscle during isometric contractions. *Eur J Appl Physiol Occup physiol* **71**, 373–378.

Jönsson, B., Laurent, C., Skau, T. & Lindberg, L.-G. 2005. A new probe for ankle systolic pressure measurement using photoplethysmography (PPG). *Biomed Eng*, Accepted for publication.

Kamal, A., Harness, J., Irving, G. & Mearns, A. 1989. Skin photoplethysmography – a review. *Comput Meth Prog Biomed* **28**, 257–269.

Kvernebo, K., Staxrud, L. & Salerud, E. 1990. Assessment of human muscle blood flow perfusion with single-fiber laser Doppler flowmetry. *Microvasc Res* **39**, 376–385.

Lindberg, L.-G. & Öberg, P.Å. 1991. Photoplethysmography II. Influence of light source wavelength. *Med Biolog Eng Comput* **29**, 48–54.

Lindberg, L.-G. & Öberg, P.Å. 1993. Optical properties of blood in motion. *Opt Eng* **32**, 253–258.

Loukogeorkakis, S., Dawson, R., Phillips, N., Martin, C.N. & Greenwald, S.E. 2002. Validation of a device to measure arterial pulse wave velocity by a photoplethysmographic method. *Physiol Meas* **23**, 581–596.

McCully, K. & Posner, J. 1995. The application of blood flow measurements to the study of aging muscle. *J Geront A Biol Sci Med Sci* **50**, 130–136.

Millasseau, S.C., Guigui, F.G., Kelly, R.P. *et al.* 2000. Noninvasive assessment of the digital volume pulse. Comparion with the peripheral pressure pulse. *Hypertension* **36**, 952–956.

Niklas, M., Moser, U., Buehrer, A. *et al.* 1998. Attenuation of the near-infrared and red photoplethysmographic signal by different depth of tissues. *Eur J Med Res* **3**, 241–248.

Nilsson, G., Tenland, T. & Öberg, P. 1980. Evaluation of a laser Doppler flowmeter for measurement of tissue blood flow. *IEEE Trans Biomed Eng* **27**, 597–604.

Owen-Reece, H., Smith, M., Elwell, C. & Goldstone, J. 1999. Near infrared spectroscopy. *Br J Anaesth* **82**, 418–426.

Radegran, G. 1997. Ultrasound Doppler estimates of femoral artery blood flow during dynamic knee extensor exercise in humans. *J Appl Physiol* **83**, 1383–1388.

Salerud, E. & Öberg, P. 1987. Single-fiber laser Doppler flowmetry. A method for deep tissue perfusion measurements. *Med Biol Eng Comput* **25**, 329–334.

Styf, J. & Körner, L. 1986. Chronic anterior-compartment syndrome of the leg. Results of treatment by fasciotomy. *J Bone Joint Surg Am* **68**, 1338–1347.

Svanes, K. 1980. Effects of temperature on blood flow. In: G. Kaley & B. Altura (eds) *Microcirculation*, pp. 21–42. University Park Press, Baltimore.

Tenland, T., Salerud, E.G. & Öberg, P.A. 1983. Spatial and temporal variations in human skin blood flow. *Int J Microcirc Clin Exp* **2**, 81–90.

Zourabian, A., Chance, B., Rode, M. & Boas, D. 2000. Transabdominal monitoring of fetal arterial blood oxygenation using pulse oximetry. *J Biomed Optics* **5**, 391–405.

