Application of a new laser Doppler imaging system in planning and monitoring of surgical flaps

Stefan Schlosser
University of Bern
Department of Plastic, Reconstructive and Hand Surgery
Inselspital
and
Department of Clinical Research
CH-3010 Bern, Switzerland

Raphael Wirth
Jan A. Plock
University of Bern
Department of Plastic, Reconstructive and Hand Surgery
Inselspital
CH-3010 Bern, Switzerland

Alexandre Serov
Central University Hospital of Lausanne (CHUV)
Service of Immunology and Allergy
CH-1011 Lausanne, Switzerland

Andrej Banic
Dominique Erni
University of Bern
Department of Plastic, Reconstructive and Hand Surgery
Inselspital
CH-3010 Bern, Switzerland

Abstract. There is a demand for technologies able to assess the perfusion of surgical flaps quantitatively and reliably to avoid ischemic complications. The aim of this study is to test a new high-speed high-definition laser Doppler imaging (LDI) system (FluxEXPLORER, Microvascular Imaging, Lausanne, Switzerland) in terms of preoperative mapping of the vascular supply (perforator vessels) and postoperative flow monitoring. The FluxEXPLORER performs perfusion mapping of an area 9×9 cm with a resolution of 256×256 pixels within 6 s in high-definition imaging mode. The sensitivity and predictability to localize perforators is expressed by the coincidence of preoperatively assessed LDI high flow spots with intraoperatively verified perforators in nine patients. 18 free flaps are monitored before, during, and after total ischemia. 63% of all verified perforators correspond to a high flow spot, and 38% of all high flow spots correspond to a verified perforator (positive predictive value). All perfused flaps reveal a value of above 221 perfusion units (PUs), and all values obtained in the ischemic flaps are beneath 187 PU. In summary, we conclude that the present LDI system can serve as a reliable, fast, and easy-to-handle tool to detect ischemia in free flaps, whereas perforator vessels cannot be detected appropriately. [DOI: 10.1117/1.3449598]

Keywords: microcirculatory blood flow; flap perfusion; ischemia; perforator vessel; free flaps.

Paper 09498RR received Nov. 17, 2009; revised manuscript received Apr. 24, 2010; accepted for publication Apr. 26, 2010; published online Jun. 15, 2010; corrected Aug. 25, 2010.

1 Introduction

Despite the latest technical improvements, the outcome of flap surgery can still be jeopardized by vascular complications, which are the most frequent cause of morbidity in reconstructive surgery. This includes the risk of partial failure, wound infection, and dehiscence due to ischemia in the remote areas of the flap or even total flap loss due to the occlusion of the arterial or venous anastomosis in microsurgical free flaps.[1–4] In both cases, vascular complication or its severity can be diminished if the preceding perfusion impairment was captured in time.[5–7]

Therefore, there is a need for technologies able to assess microcirculation in flap tissue quantitatively and reliably. This can be achieved by monitoring microcirculatory blood flow directly or flow-dependent variables, e.g., temperature, partial oxygen tension, oxygen saturation, pH, or oxidative energy metabolites before, during and after the operation.[5,8–11] All of these methods have their pros and cons; the criteria that make them eligible in clinical practice include high reliability of the data, low invasiveness, as well as easy handling and acceptable costs.

Obviously, the most conclusive data are obtained by direct measurement of microcirculatory blood flow. Among these methods, laser Doppler flowmetry plays a predominant role, because it fulfills the previously mentioned criteria most closely.[12–15] In laser Doppler flowmetry, a laser light is emitted toward a tissue surface.[16] The light penetrates the tissue at a depth of about 1 to 2 mm, where its wavelength is shifted by all particles moving within the sampled tissue volume, which is a few cubic millimeters in the case of conventional laser Doppler probes. The extent of the shift correlates with the moving particles, and the portion of shifted laser light corresponds to their amount. Out of these data, a value can be calculated that corresponds to volumetric blood flow, expressed in arbitrary units. The reliability of such measurements is complicated due to a high intersite, interindividual, and time-dependent variability.[17,18]

These drawbacks may be reduced by expanding the sample area from a few square millimeters to a larger surface. This is accomplished with laser Doppler imaging technology,[19,20] which provides more integrated microcirculatory blood flow data as well as a mapping of local flow irregularities that might possibly be related to the presence of underlying feeding vessels.[15]

The conventional laser Doppler imaging methodology consists of point-by-point sample assessments that are added to a

Address all corresponding to: Dominique Erni, Department of Plastic, Reconstructive and Hand Surgery. Inselspital, University of Bern, CH-3010 Bern, Switzerland. Tel: +41 31 632 81 35; Fax: +41 31 382 44 96; E-mail address: Dominique.Erni@insel.ch

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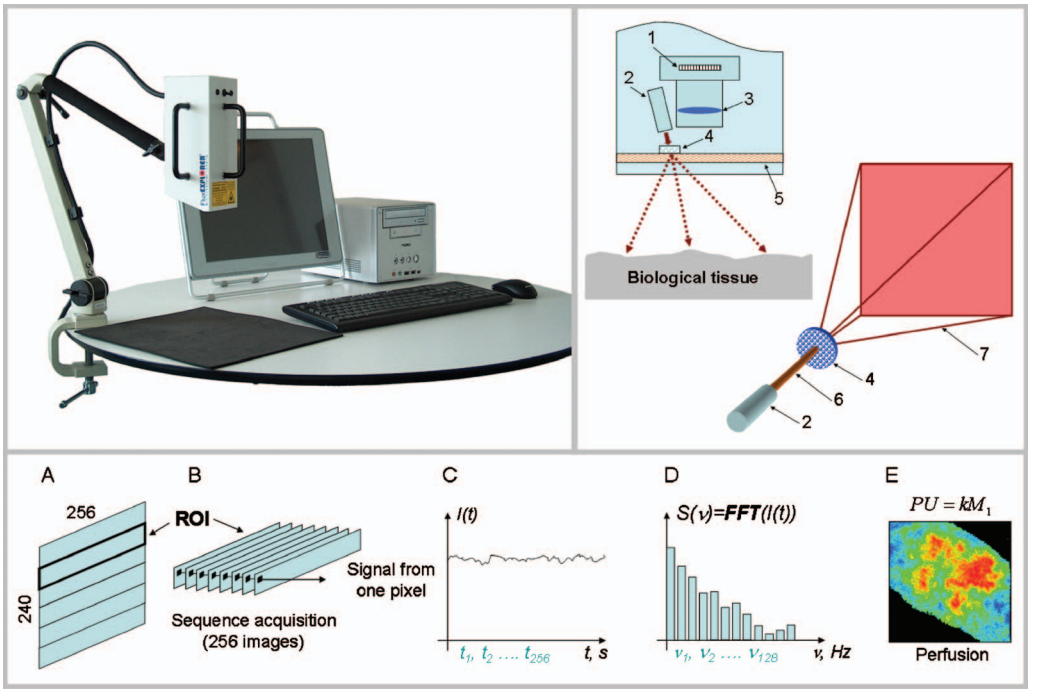

Fig. 1 Top left: Laser Doppler imaging system (FluxEXPLORER, Microvascular Imaging, Lausanne, Switzerland) consists of a CMOS camera carried by a flexible arm that can be fixed on a table, data acquisition and processing software, computer, and monitor. Besides the LDI values, the camera is able to capture black and white photographs of the same area. Top right: imaging head stuffing: 1 CMOS image sensor; 2 near-infrared laser module; 3 objective lens; 4 optical diffuser; 5 infrared filter; 6 collimated laser beam emitted by the laser module; and 7 diffused divergent laser beam after the optical diffuser. Bottom: data acquisition and processing. A CMOS image sensor; B ROI sampling; C intensity fluctuations at on pixel; D power density spectrum of the signal fluctuations; and E perfusion image.

mosaic, and that require a few minutes to scan a surface to obtain a flow map of about 300×300-pixel resolution. This technology is prone to movement artifacts, and data acquisition is time consuming.[21,22]

In the present study, we intended to test the performance and applicability of a new high-speed high-definition laser Doppler imaging (LDI) system (FluxEXPLORER) in reconstructive flap surgery. This system is based on a fast frame rate complementary metal oxide semiconductor (CMOS) image sensor, an infrared 808-nm laser source, diffractive optics for shaping the illuminating beam, and a new algorithm for obtaining high-definition images. Specifically, we aimed to: 1. testing the option of using this LDI system in the preoperative planning of flap design in terms of detecting perforator vessels that can be used as vascular pedicles nourishing the flap; and 2. establishing a threshold of the LDI signal that would most reliably predict ischemia.

2 Patients and Methods

With the approval of the local ethics committee, 19 consecutive patients scheduled for reconstructive flap surgery at our unit between November 2007 and July 2008 were included in this study. All measurements were taken by Schlosser. The inclusion criteria were flaps that are knowingly perfused by perforator vessels, free flaps, and combinations of both. Exclusion criteria were patients under 18 years, emergency cases (flap reconstruction within 3 days after injury), and mental or legal incompetence.

2.1 *Laser Doppler Imaging: FluxEXPLORER*

The LDI system (FluxEXPLORER, Microvascular Imaging, Lausanne, Switzerland) was developed on the basis of the predecessor system described by Serov and Lasser.[23] The new system was improved by utilizing an infrared 808-nm laser source, diffractive optics for shaping the illuminating beam, and a new algorithm for data processing (Fig. 1).

2.1.1 *System design and imaging principle*

Figure 1 on the right schematically shows the imager head stuffing. A collimated laser beam emitted by a laser diode of 808 nm is diffused with a diffractive optical element (DOE). Beyond the DOE, the laser beam is diverged. Projected on the area of interest, the beam forms a square shaped pattern with a homogeneous intensity profile. Photons reemitted from the illuminated tissue are collected with an objective optics and guided to light-sensitive pixels of the CMOS image sensor. For measuring the intensity fluctuations generated by Doppler-shifted photons, each pixel is sampled at a frequency of a few kilohertz. The intensity variations are recorded for each pixel, converted in digital format, and transmitted to the host PC via fast data transmission interface. After the data are acquired, an algorithm is applied to calculate the flow values.

2.1.2 *Data acquisition and processing algorithm*

The data processing is based on the classical laser Doppler power spectrum momentum analysis. To obtain one flow map over a region of interest (ROI) 256×240 pixels, the ROI was

subdivided into ten smaller regions of 256×24 pixels to achieve a faster sampling rate per pixel. For each subframe, 256 frames were acquired to record the time-domain intensity variations at each pixel. Effectively, the intensity fluctuation history was recorded for each pixel of the ROI at the sampling rate of 5 kHz per pixel. With this described procedure, the data acquisition, processing, and building up of one 256 ×240-pixel flow map takes about 1 s.

The first moment (M_1) of the power density spectrum $S(v)$ of the intensity fluctuations $I(t)$ for each pixel was calculated. The first moment M_1 is proportional to the root-mean-square (rms) speed of moving particles times the moving particle concentration in the volume sampled by the pixel. We measured the perfusion in arbitrary perfusion units (PUs), defined here as:

$$\mathrm{PU} = kM_1,$$

$$M_1 = \int_{v_1}^{v_2} vS(v)dv,$$

$$S(v) = \left| \frac{1}{\langle I \rangle} \int_0^T I(t)\exp(-i2\pi vt)dt \right|^2 - S_n(v,\langle I \rangle). \quad (1)$$

Here, k is an instrumental constant, v_1 and v_2 are low and high acquisition cutoff frequencies, accordingly; $\langle I \rangle$ is mean intensity; T is data acquisition time for one pixel; and $S_n(v)$ is the power density spectrum of the noise signal. The noise spectrum is taken from the look-up table, known after the instrument calibration performed once on a nonmoving skin-optical-properties-alike phantom. For spectrum calculation, a fast Fourier transform (FFT) algorithm is used. With as Intel 2.6 GHz Core2Duo, this LDI system provided microcirculatory blood flow mappings in an area of 9×9 cm with a resolution of one 256×240-pixel flow map within about 1 s in standard imaging mode, and in less then 10 s in the high-definition (HD) imaging mode.

2.1.3 *High-Definition imaging mode*

To reduce the influence of the heart beat or stochastic temporary flow changes, we implemented an image processing algorithm that calculates the mean value of perfusion measured by the pixel. In this HD mode, ten sequential flow values are measured for each pixel over the observation period of 6 s (Fig. 2). The final signal output reveals the average of each measurement at each pixel, thus maximizing the measurement accuracy of the data and minimizing the contribution of random signal variations. From the perfusion map quality point of view, the averaged image has less image noise, and better image contrast and clarity allowing clinicians to diagnose with more confidence.

Data processing was made with FluxEXPLORER Acquisition and Analysis software (FluxEXPLORER™, Microvascular Imaging, Lausanne, Switzerland), which provides the LDI value of a chosen area and a corresponding color map. (Fig. 3). Room temperature was kept within 22 and 26 °C for all measurements. The clinical use of the prototype LDI device was approved by the Swiss Agency for Therapeutic Products (Swissmedic, Bern, Switzerland).

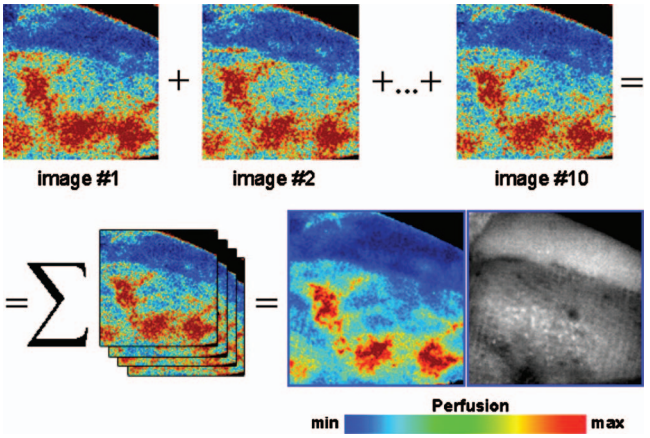

Fig. 2 High-definition imaging algorithm principle. The system obtains ten measurements sequentially within 6 s. The mean value is calculated for each pixel, thus resulting in the final perfusion map. This approach leads to more accurate measurement and detailed appearance of the flow maps.

2.2 *Perforator Vessels*

Nine patients qualified for assessment of perforator localization. LDI mappings were taken from the flap donor sites preoperatively (Table 1). The areas revealing an LDI signal that exceeded the average of the sample area by 20% were considered "hot spots." The LDI data were blinded to the surgeon, who identified the perforators intraoperatively and

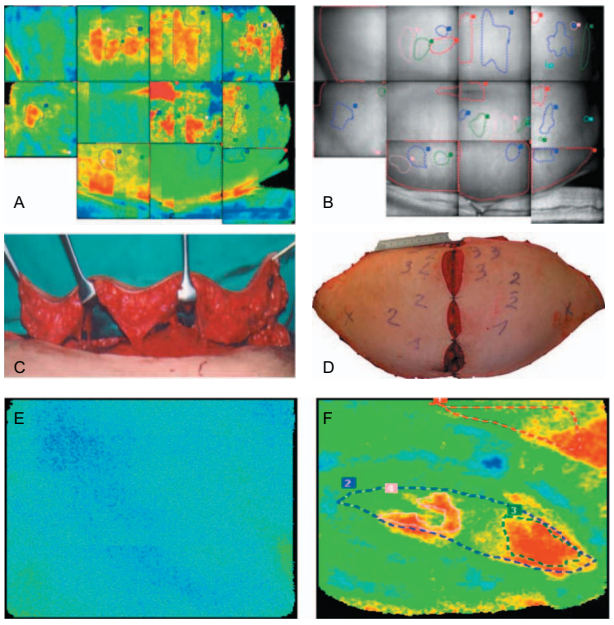

Fig. 3 (a) Color mapping of the laser Doppler imaging values obtained in abdominal skin. (b) The areas of increased blood were transferred onto a black and white photograph of the same area, on which anatomical landmarks (umbilicus, superior anterior iliac spine) can be identified. (c) The true perforator vessels were identified during flap dissection and (d) their localization was marked on the flap skin, together with the anatomical landmarks. (e) LDI image of the flap during ischemia and (f) after revascularization of the flap with microsurgical anastomoses.

Table 1 Patients data of the subset examined for detecting perforator vessels with LDI.

Patient number	Age (years)	Sex	Weight (kg)	Height (cm)	BMI (kg/m^2)	Site of examination	LDI hot spots	True perforators
1	46	F	91	160	35	abdomen	21	12
2	83	F	58	158	23	thigh	6	2
3	74	M	109	180	33	flank	15	5
4	68	F	59	154	24	abdomen	5	5
5	65	F	76	157	31	abdomen	3	1
6	50	M	56	173	18	calf	2	2
7	49	M	65	168	23	thigh	10	5
8	68	F	62	160	24	abdomen	9	6
9	54	F	60	162	22	abdomen	6	8

marked their localizations on the overlying skin, which was photographed. The coincidence of these marks and the LDI "hot spots" was evaluated by using anatomic landmarks as references, and the distances between the marks and the hot spot epicenters were measured. Thresholds of 1- and 2-cm distances were set to determine the accordance between hot spot epicenter and true perforator localization. The sensitivity and positive predictive value (PPV) of the LDI system were calculated for both thresholds. Sensitivity was defined as the percentage of true perforator localizations that matched with the hot spots, and PPV was determined as the percentage of hot spots that matched with the localizations of true perforators.

2.3 *Ischemia Threshold*

LDI images were taken from the 18 free flaps (17 patients) preoperatively (baseline) and intraoperatively before and after transection of the nourishing pedicle. Eight patients were male and nine were female. The age of the patients was 60 ± 13 years (min 39, max 83, median 58), and their body mass index was 26 ± 5 kg/m^2 (min 18, max 35, median 24). The perfusion was allowed to stabilize for 5 min before each measurement. Further time points were 1 h and every 24 h after completion of surgery until postoperative day 5. The mean value of the flap was taken and averaged for three consecutive measurements. A LDI threshold value was opted for that was supposed to provide the highest possible accuracy in predicting flap ischemia. This was achieved by validating all the LDI values against the fact whether the tissue was perfused or ischemic when the measurements were taken. The flap tissue was considered ischemic between transection of the nourishing pedicle and revascularization on the recipient site, whereas it was considered perfused at all other time points, provided there were no vascular complications.

2.4 *Statistics*

InStat version 3.0 software (Graph Pad Software, San Diego, California) was used for statistical analysis. Unless otherwise indicated, all data were presented as mean $\pm$ standard deviation. Paired ANOVA was used for assessing time-related differences. A p value of <0.05 (two-tailed) was considered statistically significant.

3 Results

The preoperative baseline flow ranged between 226 and 371 perfusion units (PU, 309 ± 37).

3.1 *Perforator Vessels*

77 hot spots revealing values of 370 ± 21 PU were found in the preoperative LDI measurements. Their LDI signal exceeded the average of the corresponding area by $37 \pm 15\%$. 46 perforators were identified during flap dissection. The distance between the location of these perforators to the next hot spot epicenter was 0.6 ± 0.5 cm. The sensitivity and PPV were 63 and 39%, respectively, for the 2-cm margin, and 38 and 24% for the 1-cm margin (Table 2).

3.2 *Ischemia Threshold*

All free flaps survived without any vascular complication. Values close to baseline were found intraoperatively after dissection of the flap, whereas flow decreased to $47 \pm 8\%$ after disconnecting the flaps (Fig. 4). Slight, but nonsignificant hyperemia was found on the first days after flap revascularization. All values obtained in the perfused flaps were above 221

Table 2 Sensitivity and positive predictive value (PPV) of LDI to detect perforator vessels accepting 1- or 2-cm margins between the LDI hot spot and true perforator.

	2-cm margin	1-cm margin
Sensitivity (%)	63	38
PPV (%)	39	24

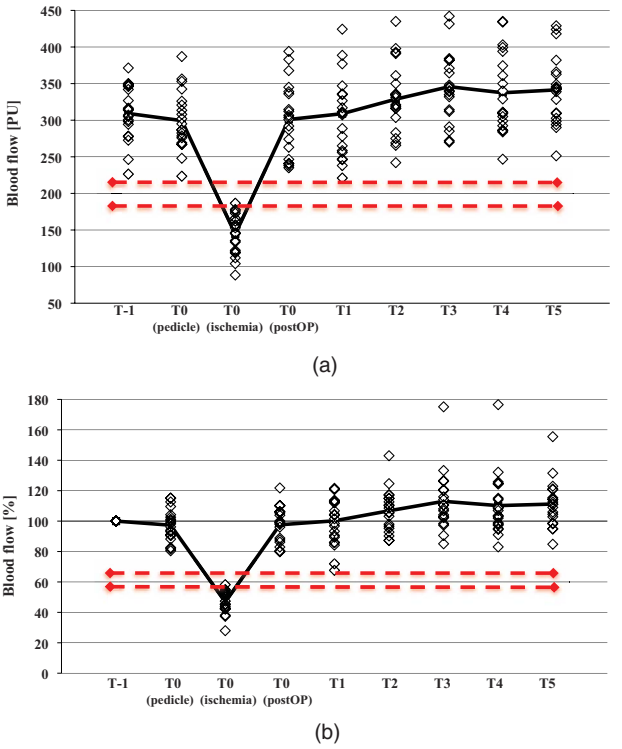

Fig. 4 Scatter plots of the averaged laser Doppler imaging values of 18 free flaps before flap dissection (T-1), after being pedicled on the feeding vessels (T0 pedicle), after being detached from circulation (T ischemia), immediately after the operation (T0 postop), and 1 to 5 days postoperatively (T1 to T5). The values are given in (a) perfusion units and (b) in percentages of preoperative baseline. The continuous line represents the mean value for each time point, and the dotted lines represent the lowest and highest values obtained in the perfused tissues and flaps, respectively.

PU (66% of baseline), and all values received in the ischemic flaps were beneath 187 PU (58% of baseline).

4 Discussion

The principal findings of this study were that the predictability and sensitivity of the LDI in terms of localizing a perforator vessel were unacceptably low, whereas a threshold could be defined that revealed sensitivity and specificity of 100% each to detect total ischemia.

The rationale of using the LDI system to localize the presence of perforator vessels is based on the assumption that microcirculatory blood flow in the skin overlying such a perforator would be increased. This hypothesis was supported by both experimental[11] and clinical studies[24–26] with infrared light-based thermography, and by a clinical study using LDI.[15] Whereas a temperature stress is required to obtain a sufficient contrast with thermography mapping, clearly demarcated zones of increased perfusion can be perceived very easily with LDI. However, unlike in the previously mentioned studies, our data revealed that the accordance of the perfusion hot spots with the underlying perforator vessels was not within a range that was acceptable for clinical use in terms of utilizing LDI to outline the flap design.

The explanations for these discrepancies are many. It is very possible that in rats, subcutaneous arteries may raise the temperature in the overlying thin skin without increasing microcirculatory blood flow there, whereas the penetration depth of LDI is not sufficient to detect deeper vascular structures in humans. In the clinical study, however, the method of validation of the LDI system was neither defined nor quantified. Another reason may be that the course of the perforator vessels may not be perpendicular to the skin,[27,28] thus causing a shift between the area where these vessels enter the skin and the area where they exit the fascia, which are both essential in flap design. Finally, our results suggest that the pattern of cutaneous microcirculation is driven by local regulations of the vascular tone in a random manner, rather than by the nearby presence of vascular in- and outflow, possibly in terms of flow motions with fluctuation periods exceeding the LDI sampling time by a higher magnitude.[29,30]

Unlike in detecting perforators, the LDI system proved to be highly accurate in discerning between normal blood flow and ischemia. Although the flaps were completely deprived of any blood flow during ischemia, the LDI signal did not decline to zero. This phenomenon has been defined as "biological zero" and explained by fluctuations of the red blood cell column due to vasomotion, and the Brownian motion of macromolecules arising from the interstitial compartment.[31,32] Therefore, it was our initial goal to outline a LDI threshold value that could be used to predict ischemia with the highest possible reliability. This expectation was exceeded because instead of a threshold value with limited accuracy, we obtained a margin of approximately 30 PU (8% of baseline) that separated LDI values measured in perfused tissue from ischemia values completely. In other words, our data suggest that all LDI values below 200 PU or 62% of baseline indicate ischemia most reliably and accurately.

High sensitivity and specificity are indispensible in the use of LDI as a monitoring device of free flaps to detect occlusion of the microvascular anastomoses. To date, the rate of such complications is reported to range between 7 and 13% in the hands of experienced microsurgeons.[1–3,33] Half of these flaps can be saved, which relies on early detection of the vascular complication, whereas total flap loss results from undetected occlusion or unsuccessful revision, thus causing major morbidity. Although the most reliable method to monitor a free flap is clinical examination by an experienced surgeon, this cannot be provided continuously due to logistic reasons, which in turn is the rationale of technical instrumentation. The decisive criteria in the choice of such a technology are the costs, and the ease of handling the device and interpretability of the produced data, as well as their reliability. The currently available techniques used for this purpose include conventional Doppler sonography and laser Doppler flowmetry, the monitoring of flow-related variables such as temperature, partial oxygen tension, oxygen saturation, oxidative energy metabolites, or the distribution of injected fluorescent dyes (indacyanine green).[9,10,34–37] Taking the previously mentioned criteria into account, our data suggest that the present LDI system may provide distinct advantages compared to these techniques. However, because no vascular occlusion was encountered in the present series of patients, the usefulness of LDI in detecting such a complication could not be proven in the present series of patients, and therefore requires confirmation in a larger clinical study.

In conclusion, our study provided encouraging data that promote the potential use of the tested LDI to monitor free flaps to prevent them from total failure, whereas probably due to anatomical and physiological reasons, this system is not able to predict the localization of perforator vessels in the course of preoperative flap design.

Acknowledgments

This study was supported by the Gebert Rüf Stiftung organization, grant number GRS-052/06.

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