Post-operative monitoring of free muscle transfers by laser Doppler imaging

Type of study Original Article

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Funding This work was supported by KTI/CTI grant 12976.2.

Running head LDI for muscle flap monitoring

Post-operative monitoring of free muscle transfers by laser Doppler imaging: a prospective study

Abstract

PURPOSE: Despite different existing methods, monitoring of free muscle transfer is still challenging. In the current study we evaluated our clinical setting regarding monitoring of such tissues, using a recent microcirculation-imaging camera (EasyLDI) as an additional tool for detection of perfusion incompetency.

PATIENTS AND METHODS: This study was performed on 7 patients with soft tissue defect, who underwent reconstruction with free gracilis muscle. Beside standard monitoring protocol (clinical assessment, temperature strips and surface Doppler), hourly EasyLDI monitoring was performed for 48 hours. Thereby a baseline value (raised flap but connected to its vascular bundle) and an ischaemia perfusion value (completely resected flap) were measured at the same point.

RESULTS: The mean age of the patients, mean baseline value, ischaemia value perfusion were 48.00± 13.42 years, 49.31± 17.33 Arbitrary Perfusion Units (APU), 9.87± 4.22 APU, respectively. The LDI measured values in 6 free muscle transfers were compatible with hourly standard monitoring protocol, and Normalized LDI values significantly increased during time ($p < 0.001$, r = 0.412). One of the flaps required a return to theatre 17 hours after the operation, where an unsalvageable flap loss was detected. All Normalized LDI values of this flap were under the ischaemia perfusion level and the trend was significantly descending during time ($p< 0.001$, r =- 0.870).

CONCLUSION: Due to the capability of early detection of perfusion incompetency, LDI may be recommended as an additional post-operative monitoring device for free muscle flaps, for early detection of suspected failing flaps and for validation of other methods.

Key Words Blood perfusion; Laser Doppler Imaging; Microcirculation; Muscle flap monitoring

Introduction

Over the past decades free tissue transfers have been used successfully for reconstruction of tissue defects after trauma or oncological ablative surgeries.[1,2] The success of free muscle transfer surgeries depends on the continuous arterial inflow and venous outflow through patent microvascular anastomoses till establishment of neovascularization by peripheral growth of vessels.[3]

Muscle has a high metabolic rate that makes a muscle transfer more susceptible to ischemia than other free tissue transfers. As a result a possible delay in the detection of ischemia is more important to a muscle transfer and therefore early detection of vascular compromise in the immediate postoperative period is essential[4,5]. Bare muscle is more challenging to monitor clinically than a muscle transfer with a skin paddle attached, even for experienced staff.[1,2,4] Hence there is a need for methods able to assess microcirculation in free muscle transfers quantitatively and reliably.[6] Various strategies have been developed to monitor such flaps postoperatively to detect flap complications.[1,7,8] The most widely used monitoring method of transferred muscle perfusion is direct clinical assessment, which can be done according to clinical characteristics of the muscle flap by an experienced clinician.[1,3] Numerous flap monitoring methods have been developed up to now, which differ in complexity, invasiveness and efficacy.[1] These methods include temperature monitoring, Doppler ultrasonography, electrical impedance plethysmography, photoplethysmography, intravenous fluorescein, transcutaneous oxygen monitoring, radionuclide imaging techniques, tissue pH, implantable Doppler monitoring, near infrared spectroscopy, microdialysis and muscle contractility testing.[3,9-11] However, there is still no consensus on which of the above mentioned techniques has the ability to become the standard accepted method for monitoring free muscle flaps.[1] Obviously, the most conclusive data are obtained by direct measurement of microcirculatory blood flow. Among these methods, laser Doppler perfusion imaging [12-14] and related techniques like laser speckle contrast analysis and laser speckle imaging [13,15,16] gained much

interest and in recent systemic reviews laser Doppler perfusion imaging has been described as most promising and as best monitoring device in free flap monitoring [3,16]. The aim of this study was to describe our clinical setting regarding surveillance of free muscle transfers using a new laser Doppler imaging technology (EasyLDI), as an additional monitoring method for early detection of vascular complications in free muscle flaps.

Patients and Methods

In a prospective study between February and August 2014, 7 patients with soft tissue defects on extremities after trauma, burn, tumor excision and infection who underwent reconstruction with free gracilis muscle in Bern Inselspital, Bern, Switzerland were included in the study. Exclusion criteria included, smoking, known microcirculatory disorders, rheumatologic disorder, asthma, peripheral vascular disease and diabetes mellitus. All procedures of this study were in accordance with the ethical standards of the Ethical Committee of the Canton of Bern (KEK) on human experimentation (No.157/12) and with the Helsinki Declaration of 1975, as revised in 1983. Each volunteer was also required to sign the informed consent and received a code number so that data could be anonymized.

The gracilis muscle was harvested from the thigh in an extra fascial manner with an adequate length of the neurovascular pedicle as described before.[17] The free gracilis was then transferred to the target for the vascular anastomosis.

In our centre the muscle flaps are routinely monitored by clinical observation, surface temperature measurements and surface Doppler monitoring. LDI measurement was done parallel to the standard methods by an independent person outside the clinical team. EasyLDI-Image analyses were done after the 48h of monitoring and had no impact in the decision making regarding flap revision. These decisions were made only based on the standard methods mentioned above.

Standard monitoring

In the first 48 hours an experienced and trained nurse was monitoring every hour the external appearance of free muscle transfers such as color (normal, dark and pale),[18] swelling or increased bleeding for signs of vascular compromise.

During our clinical observation the temperature strips (Sharn Anesthesia, Inc., Tampa, Florida) were also recorded for the patients.[18] The surgeon put the probe on two points (reference point: near the muscle and flap point: on the muscle) after the operation. In the first 48 hours an experienced nurse was measuring and recording the surface temperature from the previously marked points every hour. Temperature difference ΔT was calculated as $\Delta T = T_{Reference} - T_{Flap}$. A $\Delta T > 2°C$ was considered significant for ischemia.[18]

Furthermore, regarding the anastomosed site, a point was marked on the muscle flap after the operation. In the first 48 hours an experienced nurse was monitoring the characteristic arterial and venous flow pulsation using a low-frequency continuous-wave 8- MHz Doppler probe.

EasyLDI monitoring

The first image from the muscle was taken using the LDI device during the operation (baseline value image) when the muscle flap had been raised but was still connected to its vascular bundle (Fig.1). The second image was taken from the same point on the muscle when it had been resected completely and had been on a sterile table (ischaemia perfusion value image) (Fig.2). One hour after vascular anastomoses experienced staff members who did not participate in the clinical evaluation continued taking the images from the marked point hourly up to 48 hours. The time to reach baseline perfusion was recorded.

EasyLDI Device Measurements and Image analysis

At each of these locations, a 10-s video of perfusion was saved along with the corresponding colour photograph for future location identification. This resulted in 130 absolute perfusion

measurements for each site. For each video sequence, a region-of-interest (ROI) was manually defined and applied to the same location for all images in the video sequence. Next, the average perfusion unit per unit area of the ROI was plotted versus time. The recordings for each measurement were given in Arbitrary Perfusion Units (APU).

Laser Doppler Imaging was performed using a commercially-available microcirculation-imaging camera (EasyLDI; Aimago SA, Lausanne, Switzerland). The field-of-view (FOV) of the LDI camera is 7×7 cm with 150×150 μm pixel size. The frame rate of the perfusion images is 13 Hz. The instrument also provides simultaneously a regular color image of the FOV. An LDI instrument consists of a light source (a monochromatic laser with a long wavelength), a fast detector and a unit for processing and recording the detected signals. When the laser illuminates the tissue, part of the light is shifted in wavelength by interaction with moving red blood cells due to the Doppler effect. After detection of the coherently mixed static and dynamic light fields, the interference of both fields makes a detectable beating of the light intensity that is recorded by the device.[19-21]

Images were transferred to the EasyLDI Studio software where the average perfusion per unit per area of ROI was then extracted for each measurement of each patient.[21,22] The analysis made by a blinded person was done after the 48 hours of monitoring. In order to have more precise values we set our zero and exclude any interference from the device. Ischaemia perfusion value in each patient was considered as Zero APU and subsequent LDI measured values in each patient were adjusted according to the following formulas:

$$Adjusted\ LDI\ baseline\ value = Measured\ baseline\ level\ LDI\ value - \ ischaemia\ perfusion\ value$$

$$Adjusted\ LDI\ value = Measured\ LDI\ value - ischaemia\ perfusion\ value$$

Since vascular anatomy may be different among individuals and even between opposite sides in the same individual,[19,23] it is necessary to omit the possible intra-individual variability to

have the opportunity to compare this data between the patients. So we used the Adjusted LDI baseline value of each patient as the reference number for each person (i.e., corresponding to Normalized LDI value of 1) and the other Adjusted LDI values were normalized as following:

$$Normalized\ LDI\ percentage = \frac{Adjusted\ LDI\ value}{Adjusted\ LDI\ baseline\ value} * 100$$

Statistical analysis

All analyses were conducted using the Statistics Package for the Social Sciences (SPSS Inc., Chicago, IL, USA) version 21.0. Descriptive statistics were presented for the patients in mean ± SD. In order to validate the relationship between Normalized LDI percents of the free muscle transfers and other values (time, temperature, blood pressure and pulse rate), Pearson normalized correlation coefficient was computed across subjects.

In all cases significant results were considered for p-values less than 0.05. Due to multiple comparisons we also corrected the p-values for the correlation coefficients with the number of measurements (i.e., the corrected α-level corresponded to 0.001).

Furthermore a scatterplot was provided according to all relative measured values during the time and a straight interpolation line was fitted to the points.

Results

A total of 18 patients fulfilled the inclusion criteria. 11 cases were excluded due to the excluding criterias (smoking, known microcirculatory disorders, rheumatologic disorder, asthma, peripheral vascular disease and diabetes mellitus). We included 7 patients in our study (6 male, 1 female) aged between 24 and 69 years (mean 48.00± 13.42). All of the free gracilis transfers survived except one (14.28 %), which required a return to the operating theatre after a change detected by clinical assessment 17 hours after the operation. In the theatre a complete flap loss was detected which was not salvageable (Table1).

The first EasyLDI perfusion measurement (baseline value) ranged between 30.67 and 70.12 APU (mean 49.31± 17.33, median 40.05) and the second EasyLDI perfusion measurements (ischaemia perfusion value) ranged between 5.83 and 18.15 APU (mean 9.87± 4.22, median 9.75) (Table 2).

The muscle perfusion recovery time ranged between 60 and 180 minutes (mean 110.00± 45.16, median 119.00). In the hourly EasyLDI perfusion measurements mean Normalized LDI percent was 137%± 90 % (median 137 %).

6 free muscle transfers had shown normal results in the 48 hours of monitoring in clinical evaluations and all other used modalities. Normalized LDI percent ranges for the successful flaps and the failed flap were 10% to 384% (mean149%± 74%, median 142%), and -92% to -33 % (mean -61%± 19%, median -71 %), respectively. The Pearson correlation test showed significant positive correlation between Normalized LDI and time in the successful flaps ($p < 0.001$, r = 0.412) (Table 2, Fig.3-5).

During the first 16 hours the clinical evaluation and all other modalities of the fourth muscle flap showed the competency of the flap. The temperature difference between the flap and the reference ranged between 0,2 to 1.5 °C (mean 1.11°C). 17 hours after vascular anastomosis the clinical assessment and the surface Doppler monitoring showed no signs of perfusion. On the other hand the EasyLDI monitoring of this muscle flap did not detect any rise of the perfusion values after the anastomoses. They stayed below the ischaemia perfusion value and the trend was significantly descending ($p < 0.001$, r =- 0.870) (Table 2- Fig. 3,5).

The Pearson correlation test showed a significant positive correlation between Normalized LDI percent and muscle flap temperature ($p < 0.001$, r =0.374). Normalized LDI percent were not significantly correlated with pulse rate, diastolic and systolic blood pressure (p= 0.757, r = - 0.038; p= 0.454, r = 0.088; p= 0.430, r = -0.092 respectively).

Discussion

To the best of our knowledge we evaluated a new microcirculation-imaging camera (EasyLDI, Aimago SA) for the first time as a non-invasive additional technology for free muscle flap monitoring. With it we documented a vascular incompetency 17 hours before clinical assessment and other applied modalities. There are many techniques suitable for measurement of cutaneous microvascular perfusion, no technology has been adopted as a standard for perfusion monitoring of free muscle flaps.[2,22] Each reconstructive surgeon chooses the postoperative flap monitoring technique based on cost, availability and experiences.[18] Creech and Miller defined in 1975 the ideal monitoring device as being: Harmless to the patient, harmless to the flap, accurate and reliable, rapid, simple and inexpensive, applicable to all types of flaps, repeatable, objective, recordable, capable of prolonged constant monitoring, rapidly responsive to circulatory change, and should be equipped with a simple display that could alert relatively inexperienced personnel to development of compromised circulation.

Nowadays in most of the reconstruction clinics free tissue transfers are monitored by non-invasive conventional methods such as direct clinical and visual inspection for signs of vascular compromise, surface temperature and surface Doppler ultrasound monitoring.[2,3,11,22,24] Clinical monitoring is still widely used as the gold standard today [18,25] but demands an experienced staff for interpretation and the same staff to repeat the examination to avoid inter-observer variability.[18] Thus, less experienced staff can provide inaccurate monitoring that can result in a misinterpretation of flap status.[18,26] Additionally clinical assessment is usually subjective, and largely depending on personal experiences, thus minor, but significant changes can be missed.[18,26] The muscle colour interpretation is subjective and influenced by environmental lighting conditions.[18] Although differential surface temperature monitoring has been described for clinical monitoring experimental data does not substantiate its sensitivity because it might be influenced by environmental factors, core temperature and dressings.[3,25,27] This correlates with our data with temperature

differences less than 1.6°C between flap and reference temperature measured on a muscle flap with documented vascular compromise. Kaufman et al. assessed the effects of environmental factors on experimental muscle flap temperature as a modality for assessing vascular patency. They concluded that temperature is an unreliable, non-reproducible method to monitor the vascular status unless all environmental variables are carefully controlled,[27] because the skin temperature is not only influenced by blood flow but also by ambient temperature, by the core temperature during fever and by light. Doppler surface monitoring has several limitations as well. It is considerably more difficult to identify the weaker venous than the arterial signal by Doppler surface monitoring. There is the possibility to be deceived by a continuing Doppler arterial sound despite complete occlusion of the venous anastomosis. And it can also be challenging to differentiate between the recipient vessels and the flap's pedicle.[3]

The downsides in the current monitoring methods and the interesting recent advances in microvascular surgery have raised the need for additional methods to monitor blood perfusion of flaps to provide objective and easily communicated data . Especially free microvascular tissue transfer without skin to monitor the perfusion.[3,11,24] Laser Doppler perfusion imaging (EasyLDI) which is a recently developed imaging counterpart of Laser Doppler Flowmetry might be a good candidate. LDI eliminates the need of a probe affixed to the surface and allows measurement from a distance of a much larger area by using the Doppler shift of laser light.[28,29] The frequency shift between the transmitted and reflected light is directly proportional to the velocity of capillary blood flow. The total intensity of reflected light is inversely proportional to the amount of blood contained within the tissue.[3]

Due to the complex structure of tissue and the complexity of laser-tissue interactions blood flow parameters generated by laser Doppler systems are unable to provide an absolute measurement.[21,30] Therefore we used EasyLDI for the relative measurement of blood perfusion as the product of concentration and average velocity of red blood cells within a volume of muscle. As shown before the Normalized LDI values were widespread individually

and interindividually. Reasons for this wide range are the multiple factors which influence the perfusion of the flap tissue such as blood pressure, core temperature, haematocrit, dressings and others at the time of measurement. Therefore the trend of perfusion values should be considered more important than the absolute values, especially in the cases of venous occlusion in which there is a less abrupt decline in flow values.[40]

In free muscular flaps blood flow increases after anastomosis, however, this increase in blood flow seems not to take place immediately after transplantation.[31,32] The recovery in perfusion (110.00± 45.16 minutes in our study) within a certain period of time guarantees the survival of the muscle graft.[33] Interestingly, a relationship between ischaemic time and the occurrence of fat necrosis has recently been described.[37] A threshold value was found whereby patients with an ischaemic time longer than 99.5 min appeared to experience a significantly higher fat necrosis rate than patients with shorter times.

In the hourly monitoring of the current study EasyLDI measured values of all free gracilis, except one, were compatible with clinical evaluations done by experienced staff and other applied standard modalities. The Pearson correlation test showed that Normalized LDI values had a significant positive correlation with time ($p < 0.001$, $r = 0.412$). In one of the muscle flaps the clinical evaluation was not able to detect the malperfusion and the flap couldn't be salvaged in a return to theatre resulting in complete flap loss. In this case the patient had to undergo another reconstruction surgery which has extended his hospitalization duration and delayed his rehabilitation and functional recovery. However, the serial EasyLDI measured values showed a significantly ($p< 0.001$, $r = -0.870$) descending perfusion pattern below ischaemia perfusion value after the anastomosis and found a vascular incompetency in the muscle flap 17 hours earlier than other modalities. In a similar study by Yuen and Feng on different types of microvascular tissue transfers but using a laser Doppler flowmeter, they detected vascular compromise in the cases of suspected failing free flaps with no false positives or negatives.[34]

Moreover, the Pearson correlation test showed a significant positive correlation between Normalized LDI values and muscle flap temperature ($p < 0.001$, $r = 0.374$).While, Normalized LDI values were not significantly correlated with pulse rate, diastolic and systolic blood pressure. It should be noted that the absence of significant correlation between Normalized LDI values in muscular free flaps and other measured values might be due to denervation of the muscle flaps with loss of systemic neurovegetative regulations.

The primary limitation of this study was mainly the low number of cases. Among other things one of the reasons were the strict excluding criteria (smoking, known microcirculatory disorders, rheumatologic disorder, asthma, peripheral vascular disease and diabetes mellitus). Therefore it was not easy to reach a statistical significance from a pilot study with low patients numbers. However the results of our current study showed that LDI has the capability of early detection of perfusion incompetency and might be recommended as an additional post-operative monitoring device in free muscle flaps and for validation of other methods. Despite all its limitations our study showed promising results and it may provoke us and other centers to set up new trials regarding this monitoring method.

Acknowledgments

This work was also supported in part by the KTI/CTI (under grant 12976.2).

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Tables:

Table 1-clinical data of the muscle flaps

ID	Age (Years)	Sex	Primary Diagnosis	Flap details	Anastomose details						complications
					Artery			Vein			
					Name	No.	Anastomosis type	Name	No.	Anastomosis type	
1	51	m	Infection (defect hand right dorsal)	Right Gracilis	Right radial artery	**1**	end-to-end	Right concomitant radial vein	**1**	end-to-end	None
2	42	m	Trauma/infection (right open subtalar fracture)	Left Gracilis	Right posterior tibial artery	**1**	end-to-end	Right posterior tibial vein	**1**	end-to-end	None
3	52	m	Trauma/Burn (high voltage burn, blunt trauma in right lower leg)	Right Gracilis	Right anterior tibial artery	**1**	end-to-end	Right anterior tibial vein	**1**	end-to-end	Minor distal flap necrosis, spontaneous healing
4	69	m	Tumor (leiomyosarcoma in left forearm)	Left Gracilis	Left radial artery	**1**	end-to-end	Left concomitant radial vein	**1**	end-to-end	Complete flap failure
5	50	m	Trauma (blunt trauma cuboid/MT4+5 fracture)	Left Gracilis	Left anterior tibial artery	**1**	end-to-end	Left anterior tibial vein	**1**	end-to-end	None
6	48	m	Infection (chronic osteomyelitis after right lower limb fracture)	Right Gracilis	Right posterior tibial artery	**1**	end-to-end	Right posterior tibial vein	**1**	end-to-end	None
7	24	f	Trauma (burst calcaneus fracture)	Left Gracilis	Left dorsal artery	**1**	end-to-end	Left dorsal vein	**1**	end-to-end	None

Table 2-Free muscle transfers monitoring

ID	Monitoring modalities' results							
	Clinical evaluation	Flap temperature (°C)	References temperature (°C) †	ΔT± (°C)	Surface Doppler	LDI		
						Baseline level (apu)	Critical perfusion level (apu)	Normalized hourly values (%)
1	NL in 48 h	34.0± 1.2	32.8± 1.3	-1.2± 2.0	D in 48 h	67.92	5.86	62± 18
2	NL in 48 h	35.7± 0.6	35.8± 0.5	0.1± 0.7	D in 48 h	70.12	7.86	156± 47
3	NL in 48 h	34.1± 1.4	34.8± 1.5	0.7± 0.9	D in 48 h	37.93	9.75	164± 87
4	NL till 17 h	31.7± 0.4	33.0± 0.6	1.3± 0.5	D till 17 h	34.07	18.15	-062± 19
5	NL in 48 h	35.3± 0.8	36.1± 0.4	0.8± 0.6	D in 48 h	30.67	5.83	230± 78
6	NL in 48 h	34.3± 0.9	34.1± 1.3	-0.2± 1.1	D in 48 h	40.05	11.08	145± 49
7	NL in 48 h	34.7± 0.6	35.2± 0.7	0.5± 0.5	D in 48 h	64.41	10.54	141± 42

* *NL: Normal, h: Hours, D: Detectable, C: Centigrade*

Figure :

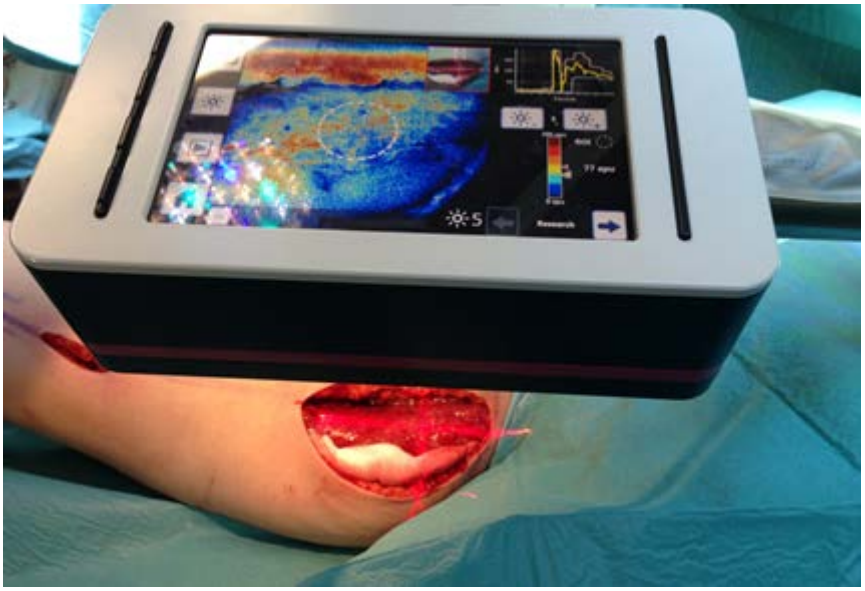

Figure 1- Baseline value image

The muscle flap had been raised but was still connected to its vascular bundle.

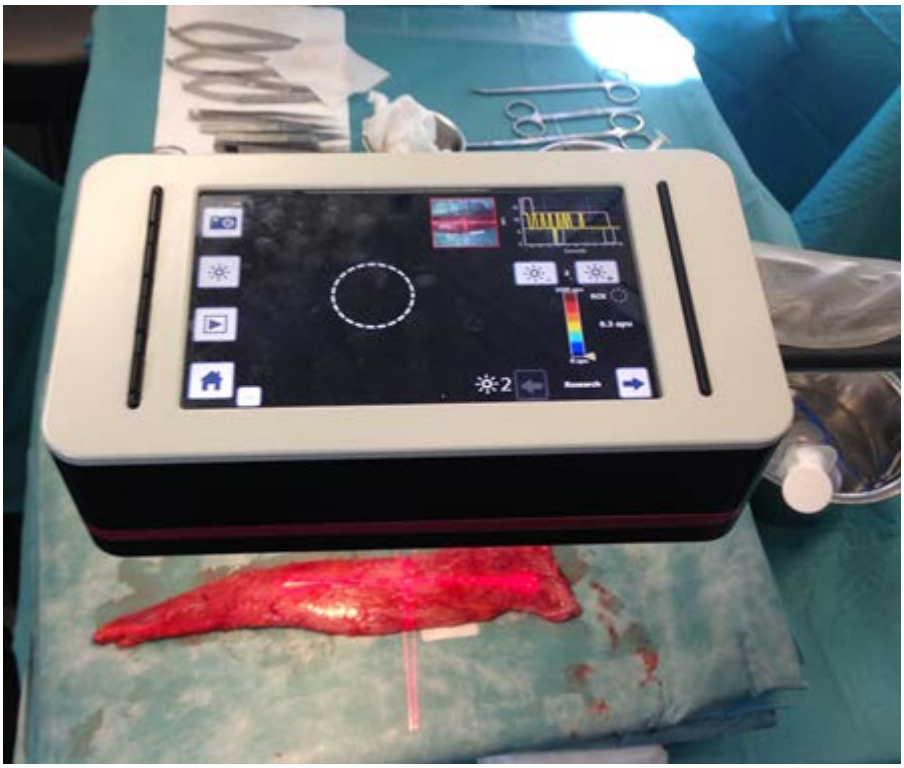

Figure 2- Ischaemia perfusion value image

The muscle was resected completely and had been on a sterile table.

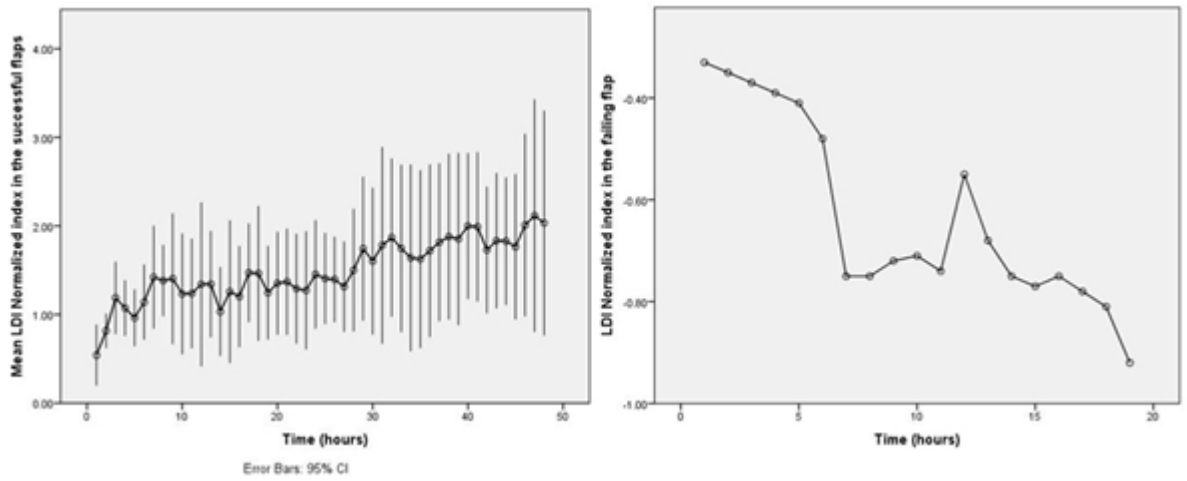

Figure 3- LDI Normalized values during the follow up time

Left diagram showed the mean LDI Normalized values of the successful flaps during the follow up time. All of the perfusion values were above ischaemia perfusion value and the trend was significantly ascending during time ($p < 0.001$, r =0.412).

Right diagram showed the mean LDI Normalized values of the failed flap (fourth patient) during the follow up time. All of the perfusion values were below the ischaemia perfusion value and the trend was significantly descending during time ($p < 0.001$, r =- 0.870).

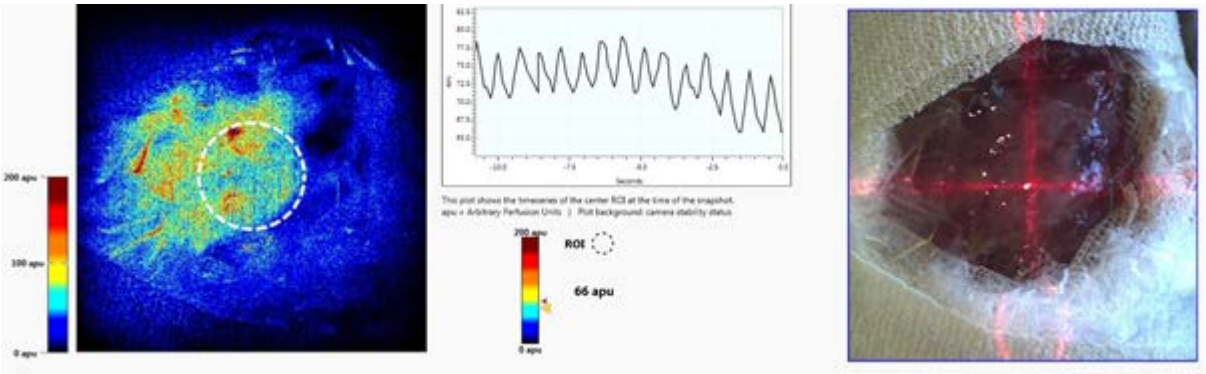

Figure 3- LDI imaging in a successful flap

Left image was a LDI image of a free muscle transfer, and right image showed the normal image of the same area. A region of interest (ROI) could be seen as a circle in LDI image, and the chart showed a 10 seconds perfusion monitoring of that free muscle transfer.

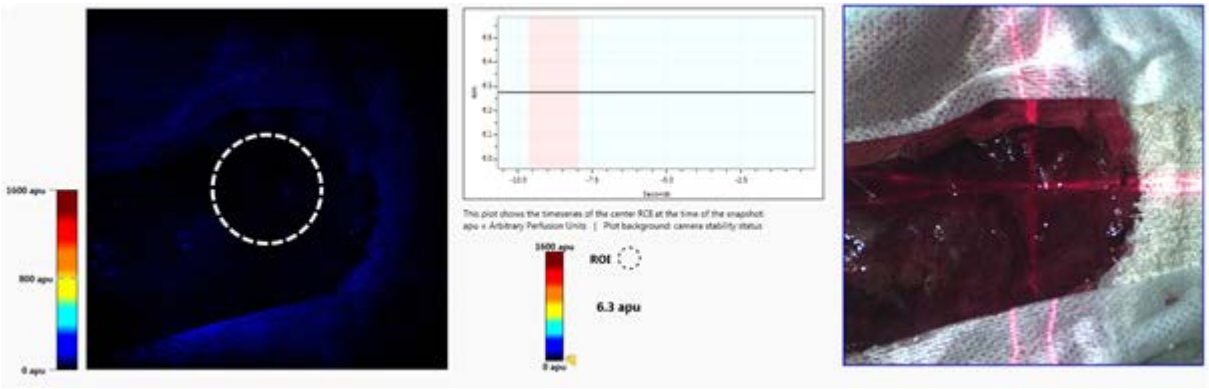

Figure 4- LDI imaging in a failed flap

Left image was a LDI image of free muscle transfer, and right image showed the normal image of the same area. A region of interest (ROI) could be seen as a circle in LDI image, and the chart showed a 10 seconds perfusion monitoring of that free muscle transfer.