IMPACT OF ACTIVE THERMOREGULATION ON THE MICROCIRCULATION OF FREE FLAPS

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Background: While it is a known fact that warming increases blood flow in healthy tissue, little is known about the impact of active thermoregulation on the altered microcirculation of free flaps. The objective of the study was to identify the impact of postoperative active thermoregulation on free flap microcirculation. *Methods:* Tissue temperature was assessed in 25 free perforator flaps using an implanted probe. Active thermoregulation was achieved using a water circulation based system. Changes in microcirculation were evaluated at the day of surgery and throughout the first three postoperative days after passive cooling (room temperature), passive warming (wound dressing), active warming (38°C) and active cooling (15°C) using laser Doppler flowmetry and remission spectroscopy. *Results:* Active warming increased flap temperature by 7.7% to 36.4°C ± 0.5°C in comparison to the initial values of flaps without dressing ($P < 0.001$). As a result, the blood flow increased by 77.7% of the base value ($P < 0.001$). A significant correlation between all microcirculation parameters and tissue temperature was observed with a 5.52 AU blood flow increase per degree temperature increase ($r = 0.7$; $P < 0.001$). All microcirculation parameters showed a statistically significant increase after both passive and active warming, whereby active warming showed significantly higher values than passive warming. *Conclusions:* Active thermoregulation using water-based circulation is an effective and safe procedure to improve microcirculation in free flaps and is superior to conventional passive warming strategies. © 2015 Wiley Periodicals, Inc. Microsurgery 00:000–000, 2015.

In the past decades, free flaps have revolutionized reconstructive surgery. Technical advances and the growing experience in planning and performing microvascular free flap transfers have helped to increase the safety of this technique gradually. Most strategies to reduce the risk of complications are aimed at avoiding *macro*circulation disruptions caused by thrombosis at the site of the anastomosis, which can lead to total flap failure if left untreated.[1–9] Disruptions in the *micro*circulation system, on the other hand, are commonly associated with partial flap failure. Scientific research in this area is scarce, although the significance of microcirculation disruptions should not be underestimated: partial flap necrosis often requires revision surgery with all associated risks for the patient and poses a considerable economic burden. Depending on location and severity, a decrease in microcirculation may jeopardize the whole reconstructive procedure.

An increase in temperature leads to an increased blood flow in living tissues. Although most microsurgeons would agree that an increase in temperature also has a positive impact on free flap perfusion, this assumption has never been properly investigated. So far, the clinical practice of thermoregulation is limited to applying wound dressings on free flaps to prevent cooling. There is no mention of postoperative active flap warming in standard postoperative protocols,[1,2,10,11] although Awwad et al.[12] demonstrated a significant increase of blood flow in the macrocirulation system of skin flaps back in 1983. The observed changes in the macrocirculation following active thermoregulation, however, cannot be directly applied to the hemodynamics of the equally important nutritive microcirculation system. Moreover, the hemodynamics of both macro- and microcirculatory vessels cannot be compared with healthy tissue. Micro- and macrocirculation change severely after flap raising and restoring of the perfusion following flap transfer,[13–15] which is mainly ascribed to postischemic metabolic changes and denervation (sympathectomy) of the flap tissue.[16,17] If, and to what extend active postoperative tissue warming has an influence on free flap microcirculation remains unknown and is the object of this study.

PATIENTS AND METHODS

This prospective, clinical trial was conducted from November 2013 to April 2014. The ethic committee of the Bayerische Landesärztekammer approved the consent procedure and the whole trial (No. 11038). All patients gave written informed consent. The study was planned and conducted in accordance with the World Medical Association Declaration of Helsinki (June 1964) and subsequent amendments. This study is registered with

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For this study, patients between 18 and 50 years of age who needed soft tissue reconstruction by means of a free perforator flap were recruited. Defect location was irrelevant. Patients with (1) a history of cardiac disease, (2) arterial hypo- or hypertension, (3) intake of vasoactive drugs, (4) peripheral arterial occlusive disease and (5) chronic kidney and liver disease, (6) vasculitis, and (7) incompliance were excluded from the study.

A total of 25 patients (10 female, 15 male) who had received a free perforator flap between November 2013 and April 2014 were included into the study. The mean age of the patients was 49 ± 13 years. Reconstructive Surgery was performed because of traumatic soft tissue injury, open fractures, amputation injuries, soft tissue malignant neoplasm, scar contractures, unstable scars, and for breast reconstruction after breast cancer. Anterolateral thigh perforator flaps were used in 44% of the cases, followed by epigastric perforator flaps in 28% of the cases, groin flaps (16%), parascapular flaps (8%), and extended lateral arm flaps (4%). Recipient sites were head (12%), upper (12%) and lower extremities (48%), and breast (28%).

Measuring Methods

Measurements were conducted at the day of the operation as well as the following 3 days after exposing the tissues to a particular temperature for 1 hour. Changes in microcirculation were evaluated each day after passive cooling (without dressing at room temperature ~24°C), passive warming (conventional wound dressing), active warming (38°C), and active cooling (15°C) (Table 1). Passive warming was achieved with a standard wound dressing composed of three layers of cotton gauze at room temperature. Active warming and cooling were enabled using a water circulation based system (Hilotherm Clinic®, Hilotherm GmbH, Argenbuhl-Eisenharz, Germany). This system allows temperature transmission by a water suffused polyurethane cuff that is placed on the skin island of the flap. Subsequently, the flap was covered with a conventional wound dressing comprising of three layers of cotton gauze (Fig. 1).

To assess tissue temperature most accurately, an invasive temperature measurement probe (Licox® IMC Temperature Micro Probe, Integra LifeSciences Corporation, Plainsboro, NJ) was placed 8 mm underneath the skin.

The impact of active thermoregulation on the microcirculation of free perforator flaps was assessed using combined laser Doppler flowmetry and remission spectroscopy (Oxygen to See, O2C®, Lea Medizintechnik, Giessen, Germany). The functional principle and the application of the O2C® device as a monitoring tool in free flap surgery has been described elsewhere in detail.[18,19] It allows non-invasive measurement of blood flow (arbitrary units, AU), capillary venous oxygen saturation (%) and relative postcapillary filling pressure (AU) using a fiber optic probe. The probe was placed and fixed in position in the center of the perforator flap skin island, whereby measurements were taken at a level of the temperature probe, 8 mm below the skin. The primary outcome measure was the blood flow of the microcirculation. Secondary outcome measures were tissue temperature, capillary venous oxygen saturation, and the relative postcapillary filling pressure of the free flap.

Table 1. Experimental Protocol

Measurement	Thermoregulation	Applied temperature (60 min)	Description
1	Passive cooling	~24°C	Exposure to room temperature, no dressing
2	Passive warming	N/A	Wound dressing only
3	Active warming	38°C	Wound dressing and Hilotherm
4	Active cooling	15°C	Wound dressing and Hilotherm

Measurements 1–4 are repeated daily at the day of surgery and throughout the first three postoperative days

Abbreviations: N/A, not applicable. Due to the experimental setup, no predefined temperature value can be provided for "passive warming."

The objective of this study was to assess the effect of different thermoregulation methods on tissue temperature after free flap transfer and to analyze the impact of temperature change on microcirculation parameters. The described experimental design allowed standardized measurements of microcirculation changes at different temperatures without the need to manipulate the wound dressing. Furthermore, motion and pressure artifacts could be avoided, thereby eliminating these common sources of error when using the sensible O2C probe.

Statistics

Continuous measurements are presented as mean ± standard deviation or median (min–max) for skewed distributions and categorical values as absolute and relative frequencies. In order to account for repeated measurements, mean values over time (days 1–4) or over method (without dressing, with dressing, active warming, active cooling) were computed and used for analysis where appropriate. Repeated measurements ANOVA and paired *t*-tests were used to investigate differences in measurements between days or methods. Strength of association between two variables was assessed using between and within subject correlations while linear mixed models with subject and day of measurement as random effects

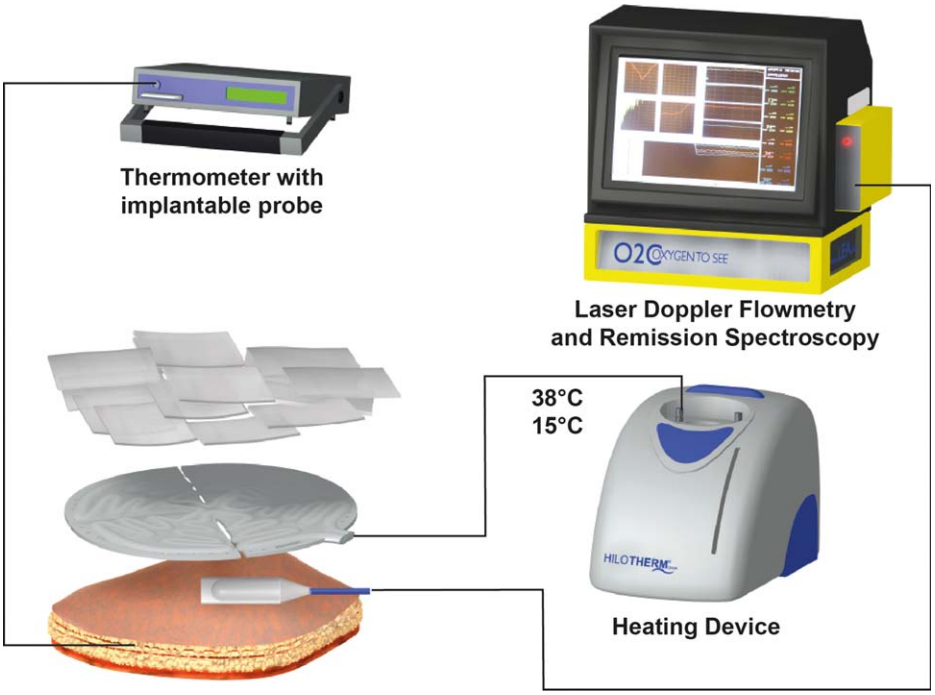

Figure 1. Experimental setup. Active warming and active cooling was achieved using a water based heating device connected to a water suffused polyurethane cuff which was placed directly on the flap. Tissue temperature was assessed using an invasive probe placed 8 mm underneath the skin. Measurement of microcirculation parameters was achieved using combined laser Doppler flowmetry and remission spectroscopy (O2C device).

were used to describe the influence of temperature on variables.

All reported P values are two-sided and have not been adjusted for multiple testing. All analyses were conducted with the use of R software, version 3.1.0.

RESULTS

In all cases, the postoperative course was uneventful, no adverse effects to any thermoregulation method used in this study was observed. All flaps showed no clinical evidence of circulatory disturbances and healed in without complications (follow-up time 1 year, no patient was lost to follow-up).

Tissue Temperature

The mean tissue temperature of the flaps was 33.8°C ± 1.1°C without dressing. All thermoregulatory methods (passive and active) achieved statistically significant changes of the tissue temperature ($P < 0.001$). If the flaps were covered with a wound dressing, an increase in temperature to 34.8°C ± 0.9°C was achieved. Active warming further increased the tissue temperature to 36.4°C ± 0.5°C. After active cooling, tissue temperature dropped to 34.5°C ± 0.9°C (Fig. 2). This trend could be observed at all four days (Fig. 3). Moreover, mean tissue temperature differed significantly between days ($P = 0.002$), with values at day 1 being significantly lower compared with the following days [Δ day 1–day 2: 0.84 (95% CI: 0.38–1.30); Δ day 1–day 3: 0.74 (95% CI: 0.21–1.28); Δ day 1–day 4: 0.5 (95% CI: 0.14–0.86), see Fig. 3].

Microcirculation Parameters

Blood flow. In comparison with the base values (no dressing), all thermoregulatory methods had a statistically significant impact on blood flow ($P < 0.001$). Furthermore, active warming yielded significantly higher flower rates compared with all other thermoregulatory methods (passive warming, active cooling) ($P < 0.001$). Mean increase of the blood flow was 21.9% after applying a wound dressing. Active warming further increased mean blood flow by 55.8%, resulting in an overall increase of 77.7% in comparison with the base values (no dressing) (Fig. 4). Active cooling of the free flaps, on the other hand, resulted in a decrease of blood flow by 23.9%. This trend could be observed at all 4 days (Fig. 3). Throughout the investigation period, a significant correlation between blood flow and tissue temperature was observed ($r = 0.7$; $P < 0.001$ within subject correlation). The investigation of a functional relationship between these two parameters showed a significant influence of temperature on the blood flow ($P < 0.001$) with a 5.52 AU blood flow increase per degree temperature increase.

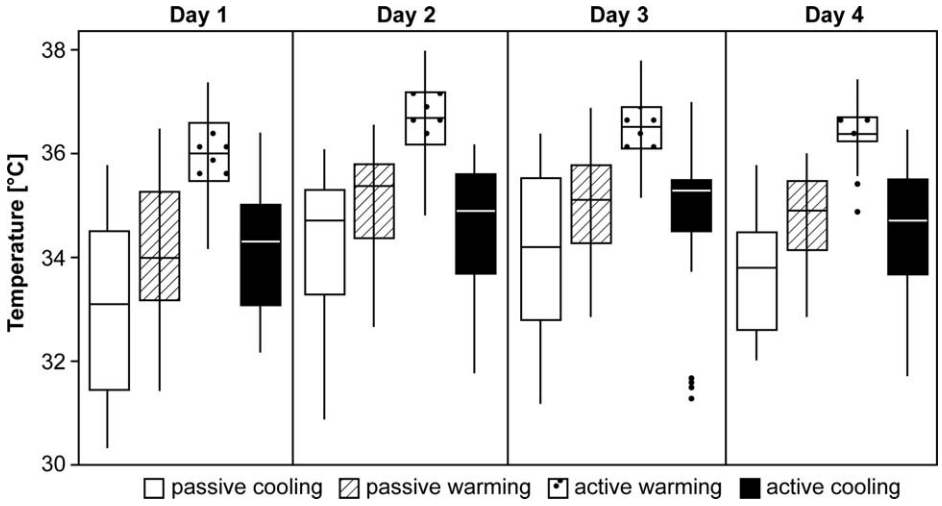

Figure 2. Thermoregulation induced temperature changes. Boxplot analysis of temperature changes after passive cooling (exposure to room temperature), passive warming (wound dressing), active warming and active cooling (water based heating/cooling device) at days 1 (day of the operation) through 4 (postoperative day 3).

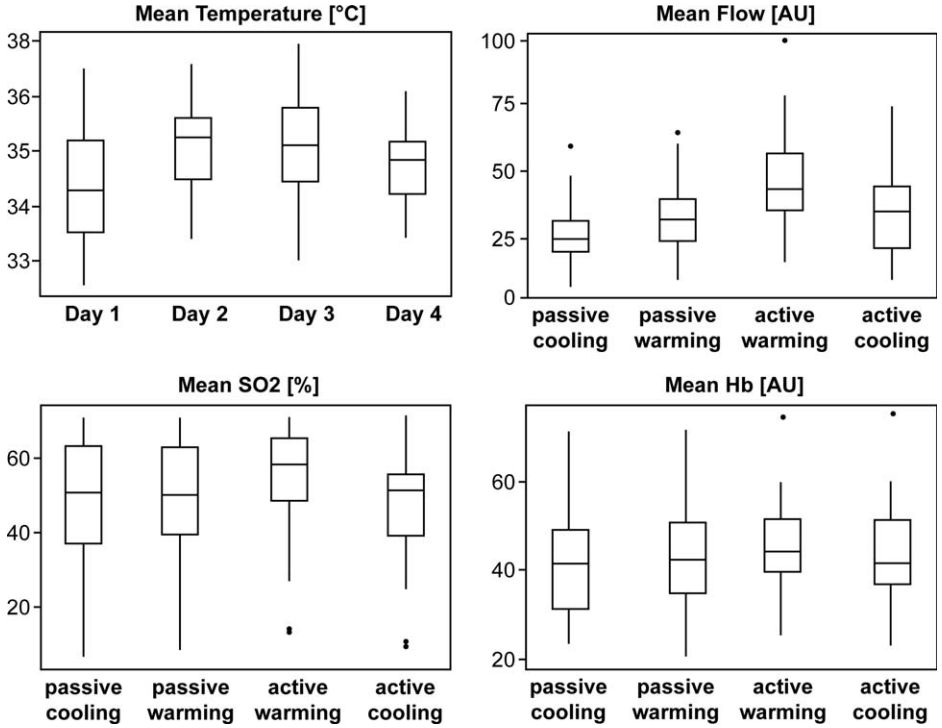

Figure 3. Impact of thermoregulation on microcirculation parameters. (Upper left) Boxplot analysis of mean temperature changes (all groups) at days 1 (day of the operation) through 4 (postoperative day 3). Boxplot analyses of mean blood flow (upper right, arbitrary units, AU), mean tissue oxygenation (SO2 in percent, lower left) and mean postcapillary venous filling pressure (Hb, lower right) after passive cooling (exposure to room temperature), passive warming (wound dressing), active warming and active cooling (water based heating/cooling device) at all time points.

Oxygen saturation. Oxygen saturation differed significantly after active warming in comparison with all other groups [$P = 0.003$ (with dressing); $P = 0.008$ (no dressing); $P = 0.005$ (active cooling)]. Active warming led to an increase of the mean oxygen saturation of 13.5% in comparison with passive warming (dressing only) and an overall increase of 17% in comparison with the base values (no dressing). Active cooling after initial warming of the flap led to a decrease of the mean oxygenation of 12.3% (Fig. 5). This observation could be

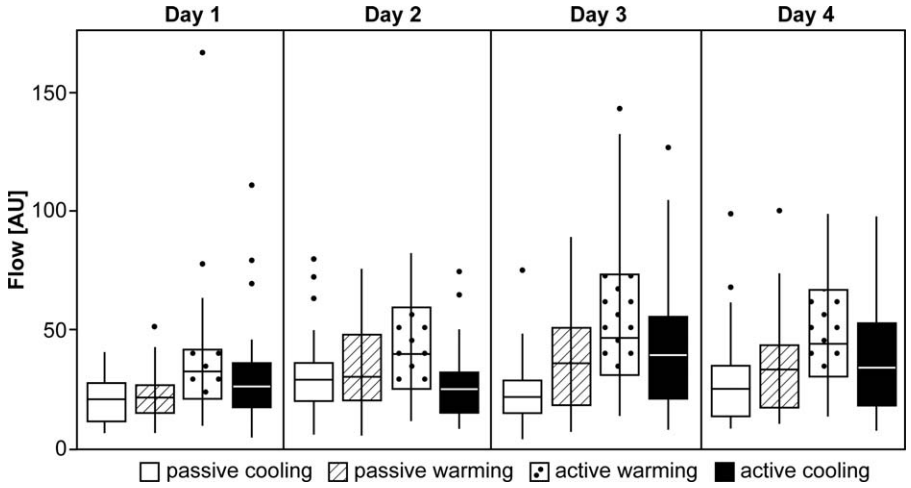

Figure 4. Impact of thermoregulation on blood flow. Boxplot analysis of changes in blood flow (arbitrary units, AU) after passive cooling (exposure to room temperature), passive warming (wound dressing), active warming and active cooling (water based heating/cooling device) at days 1 (day of the operation) through 4 (postoperative day 3).

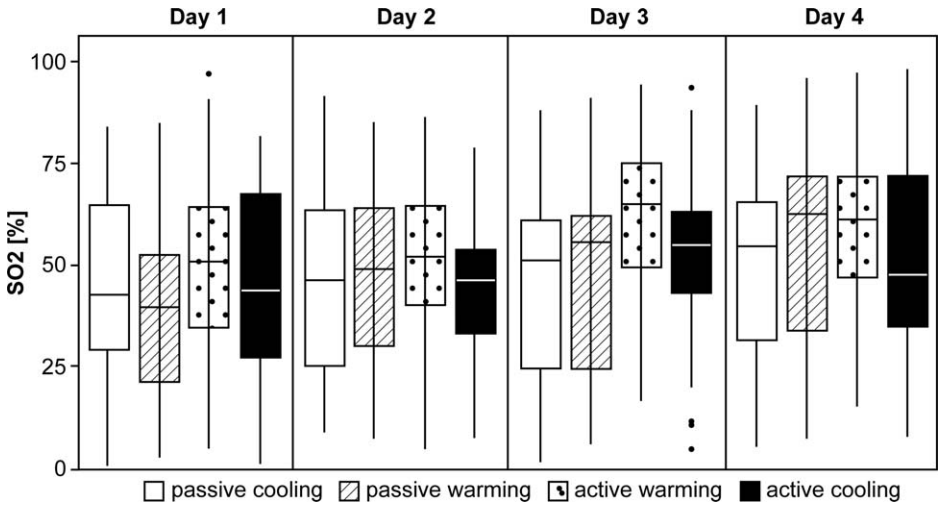

Figure 5. Impact of thermoregulation on tissue oxygenation. Boxplot analysis of changes in oxygen saturation (SO2 in percent) after passive cooling (exposure to room temperature), passive warming (wound dressing), active warming and active cooling (water based heating/cooling device) at days 1 (day of the operation) through 4 (postoperative day 3).

made at all four days (Fig. 3). Covering the flap with a wound dressing (passive warming) led to an increase of only 3% ($P > 0.05$) when compared with the baseline values (no dressing). Oxygen saturation and tissue temperature correlated significantly over the entire investigation period ($r = 0.43$; $P < 0.001$). Temperature change had a significant impact on oxygenation ($P < 0.001$) with a 2.19% increase of oxygen saturation per degree temperature increase.

Postcapillary Venous Filling Pressure

Again, active warming differed statistically significantly from all other study groups with regard to postcapillary venous filling pressure [$P = 0.002$ (dressing only)/$P < 0.001$ (no dressing)/$P = 0.002$ (active cooling)]. In comparison with passive warming (wound dressing) and passive cooling (no wound dressing), active warming led to an increase of the postcapillary filling pressure of 5% and 8%, respectively. Active cooling, on the other hand, was followed by a decrease of the mean postcapillary filling pressure by 4% (Fig. 6). The comparison between passive cooling and passive warming reached no statistical significance (2.6% increase after passive warming, $P = 0.06$). These trends were observed at all 4 days (Fig. 4). Postcapillary venous filling pressure and tissue temperature correlated significantly over the entire investigation period ($r = 0.53$; $P < 0.001$). Temperature changes had a significant impact on the postcapillary

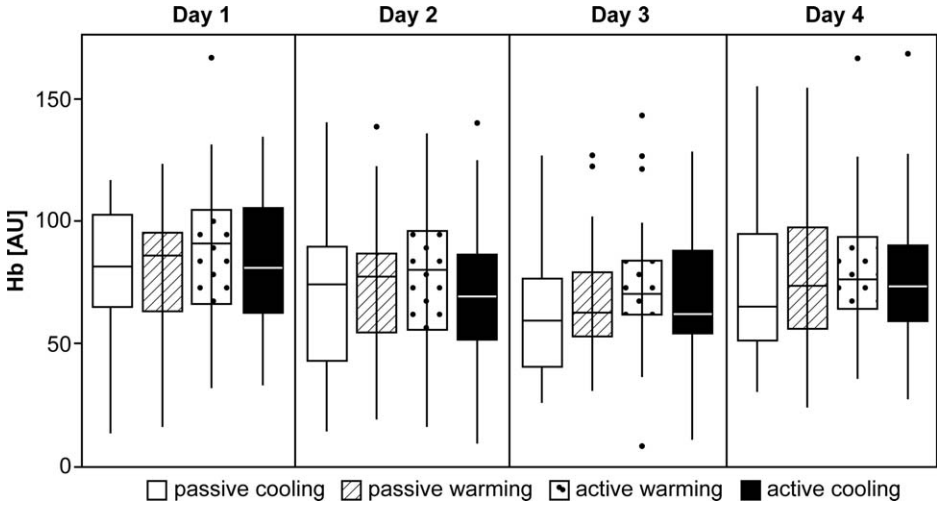

Figure 6. Impact of thermoregulation on the postcapillary venous filling pressure. Boxplot analysis of changes of the postcapillary venous filling pressure (Hb in arbitrary units, AU) after passive cooling (exposure to room temperature), passive warming (wound dressing), active warming and active cooling (water based heating/cooling device) at days 1 (day of the operation) through 4 (postoperative day 3).

venous filling pressure ($P < 0.001$) with a 0.94 AU increase of the postcapillary venous filling pressure per degree temperature increase.

DISCUSSION

Instead of supporting and optimizing free flap perfusion, postoperative treatment is focused on the early detection of macrocirculation disturbances using more and more technologized equipment.[1,2,10,11] Postoperative supportive measures are usually limited to the controversially discussed administration of anticoagulants aimed at improving blood rheology and avoiding early thrombosis.[1,2,10,11] Alternative concepts to influence flap perfusion are rarely considered. Despite a body of evidence indicating that warming increases blood flow in healthy tissues,[20] postoperative measures of thermoregulation after free flap transfer are usually limited to avoid cooling by dressing the wound. Active warming is even discouraged by some for fear of dermal burn injuries to the denervated skin island.[21] The current low standing of postoperative thermoregulation in free flap transfer may be explained by the limited data available on this subject. While the influence of temperature on blood circulation in healthy skin has been well studied,[20] this knowledge cannot be applied directly to free flap transfer because of the substantially altered physiology of free flaps. In contrast to intact skin, free flaps have been deprived of all afferent and efferent nerve connections resulting in a hemodynamically relevant sympathectomy.[13,22,23] Thus, free flaps are completely cut off from the dominating central thermoregulation and need to react to temperature changes autonomously by local mechanisms. Hemodynamics are additionally affected by metabolic factors following ischemia-reperfusion.[17,24] Despite these changes, blood flow can be influenced by thermal stimuli from the outside: Awwad et al.[12] showed a significant increase in blood flow in the macrocirulation after warming skin flaps. A sufficiently high blood flow in the supplying blood vessels is mandatory in free flap transfer to avoid thrombosis at the site of the anastomosis.[25,26] Despite being of equal importance for the survival of the supplied tissues, blood flow in the microcirculation of the capillary system cannot be directly compared with the macrocirculation in the large supplying vessels. This becomes particularly obvious if one looks at the opposed hemodynamic changes in the two vascular systems after flap raising and microvascular anastomosis:

Followed by the above mentioned changes in blood flow regulation, a reactive increase in blood flow of the macrocirculation system can be observed, while blood flow in the microcirculation system decreases.[14,15,17,27] Arteriovenous shunts, so-called "choke vessels," seem to have a significant influence on these opposing changes. Up to 85% of total blood flow can be shunted by these vessels following flap raising, draining off the corresponding blood volume from the microcirculation system.[28–33] Considering the fact that shunt vessels also react to thermal stimuli,[34] it is safe to assume that the increase in blood flow Awwad et al.[12] observed in the macrocirculation system after active warming can at least partly be attributed to an increased shunt volume. This in turn would mean that active warming of free flaps would in fact trigger a decrease in blood flow in the microcirculation system.

Our data, however, suggest that active warming of free flaps increases blood flow even in the microcirculation system. What is more, it is becoming clear that even

small temperature changes of a single degree Celsius can be sufficient to increase blood flow in the microcirculation system by 20.7%. The mean flap temperature (with wound dressing but without active warming) was 35°C, which is comparable with measurements of other authors.[35] This provides adequate scope for temperature increase without risking cell damage (>40°C).[36] The thermoregulation device used in our study was regulated to a maximum temperature of 38°C. The incoming water suffused tube and the isolating air layer between heating cuff and flap surface resulted in a certain heat loss which prevented the achievement of a targeted tissue temperature of 38°C (mean temperature after active heating was 36.4°C ± 0.5°C). It seems plausible, that an additional temperature increase may further increase blood flow in the microcirculation system, since the maximum vasodilation is observed at 42°C in healthy skin.[37–39]

Kraemer et al.,[35,40] also demonstrated a correlation between free flap temperature and microcirculation parameters. The basic idea behind their study was the assumption that microcirculatory disruptions in critical flaps are followed by a passive decrease in tissue temperature. Therefore, measurement of flap temperature could be used as a postoperative monitoring tool to assess flap viability. Our study investigated the reverse perspective. The effect of active postoperative warming on the microcirculation of free flaps has not been systematically studied before. To the best of our knowledge, our study is the first to demonstrate a significant impact of an *active* thermoregulation on *microcirculation* parameters of free flaps. Providing a sufficient microcirculation inside the flap tissue is essential to avoid flap necrosis. Experimental ischemia models could demonstrate that the development of necrosis in the critical postcapillary areas of the flap can be explained by the perfusion pressure which decreases to the periphery on the one hand, and a considerably higher resistance of peripheral small vessels on the other hand.[30,33] Tissue warming can lead to a reduction in vascular resistance via locally induced vessel dilatation.[20] In conjunction with the Venturi effect, the resulting increase of the vessel diameter should lead to a slowdown in blood flow, in principle. Instead, our results indicate an increase in blood flow, which we explain with a simultaneous increase of the perfusion pressure. The observed increase of the postcapillary venous filling pressure after active warming supports this theory. Both effects, the reduction of the vessel resistance, as well as the increase of the postcapillary venous filling pressure counteract pathophysiological mechanisms that might otherwise lead to partial necrosis.

Despite this positive influence, it needs to be addressed that tissue warming also increases cell metabolism, marked by the fact that changes in oxygenation after active warming was disproportionate to blood flow in our study. Especially against the background of an increased cell metabolism after active warming, the observed increase of oxygen saturation indicates that the increased blood supply more than compensated for an increased oxygen demand. A general temperature increase observed between day 1 and the following days may be contributed to adaptation of the hemodynamic system to the altered blood flow situation after flap raising. This is in line with results from Hanasono et al.,[41] who described an increase of the blood flow of the large supplying vessels in the first 3 days following surgery. Significantly, these hemodynamic reorganizations in the first 3 days coincide with the highest risk of thrombosis.[42,43] Even though technical errors are the most common cause for vessel occlusions,[44,45] the low initial perfusion post-surgery increases the risk of thrombosis.[25,26] In the attempt to ensure patency of the anastomoses after free flap transfer, all steps necessary should be taken to establish high perfusion levels in the early postoperative phase. Active temperature regulation poses the ideal instrument to achieve just that. As our results have shown, the heating device used in our study achieved a more efficient warming of the flap tissue than conventional measures (wound dressing). Moreover, conventional wound dressings can even have the opposite effect: dressings are intended to absorb wound exsudate or leaking blood and may cause evaporation cooling. Cooling free flaps to the temperature level before active warming lead to a decrease of microcirculation parameters in our study. This provides additional prove that the changes observed in microcirculation can in fact be attributed to the induced changes in tissue temperature.

Prerequisites for the routine clinical application of active thermoregulation are a high level of security with regard to the prevention of burn injuries, as well as a certain comfort of use for both patients and nursing staff. Existing devices commonly used for tissue warming like hot-water bottles or infrared lamps lack sufficient control of heat development and distribution, which is why these procedures are often associated with dermal burn injuries in the literature.[21,46] Hot-air blowers have the drawbacks of a high noise level and a non-selective warming area. The Hilotherm® device used in the presented study, on the other hand, provides low-noise, controlled, safe, and locally restricted warming of selected skin areas and has proven to be a novel and suitable instrument for the thermoregulation of microvascular free flaps superior to conventional methods.

The statements of the presented study are limited by a relatively small sample size ($n = 25$). In future studies, the effects of a temperature increase should be pursued over a longer time span. Also, hemodynamic values may vary in different flaps and different locations of the flaps. Therefore, it should be further investigated if and to

what extent these differences have an influence on the effect of active postoperative flap warming.

CONCLUSIONS

Our data show that active thermoregulation during the first 3 days after microvascular free flap transfer is an effective, safe, and convenient procedure to optimize microcirculation in free flaps. In our opinion, active thermoregulation should be considered as an integral part of the postoperative treatment concept after free flap transfer. Future studies will need to address the question whether or not the observed positive effects on microcirculation have a significant impact on the reduction of partial necrosis caused by territorial microcirculation disturbances.

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