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Monitoring of partial and full venous outflow obstruction in a porcine flap model using laser speckle contrast imaging

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Summary *Background:* In microsurgery, there is a demand for more reliable methods of post-operative monitoring of free flaps, especially with regard to tissue-threatening obstructions of the feeding arteries and draining veins. In this study, we evaluated laser speckle contrast imaging (LSCI) and laser Doppler flowmetry (LDF) to assess their possibilities to detect partial and full venous outflow obstruction, as well as full arterial occlusion, in a porcine flap model.
Methods: Cranial gluteal artery perforator flaps (CGAPs) were raised, and arterial and venous blood flow to and from the flaps was monitored using ultrasonic flow probes. The venous flow was altered with an inflatable cuff to simulate partial and full (50% and 100%) venous obstruction, and arterial flow was completely obstructed using clamps. The flap microcirculation was monitored using LSCI and LDF.
Results: Both LDF and the LSCI detected significant changes in flap perfusion. After partial (50%) venous occlusion, perfusion decreased from baseline, LSCI: 63.5 $\pm$ 12.9 PU ($p = 0.01$), LDF 31.3 $\pm$ 15.7 ($p = 0.64$). After 100% venous occlusion, a further decrease in perfusion was observed: LSCI 54.6 $\pm$ 14.2 PU ($p < 0.001$) and LDF 16.7 $\pm$ 12.8 PU ($p < 0.001$). After release of the venous cuff, LSCI detected a return of the perfusion to a level slightly, but not significantly, below the baseline level 70.1 $\pm$ 11.5 PU ($p = 0.39$), while the LDF signal returned to a level not significant from the baseline 36.1 $\pm$ 17.9 PU ($p > 0.99$). Perfusion during 100% arterial occlusion decreased significantly as measured with both methods, LSCI: 48.3 $\pm$ 7.7 (PU, $p < 0.001$) and LDF: 8.5 $\pm$ 4.0 PU ($p < 0.001$). During 50% and 100% venous occlusion, LSCI showed a 20% and 26% inter-subject variability (CV%), respectively, compared to 50% and 77% for LDF.

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Conclusions: LSCI offers sensitive and reproducible measurements of flap microcirculation and seems more reliable in detecting decreases in blood perfusion caused by venous obstruction. It also allows for perfusion measurements in a relatively large area of flap tissue. This may be useful in identifying areas of the flap with compromised microcirculation during and after surgery.

Introduction

Microsurgical free-tissue transfer is a well-established technique to cover defects after trauma and cancer surgery. The major cause of flap failure is venous thrombosis, which leads to a decrease of the blood drainage from the flap and a stasis and subsequent congestion of blood in the tissue. This mostly occurs within the first 2 days postoperatively, during which the viability of the flap must be regularly checked by the surgeon and other health staff.[1,2]

The success rate for most types of microsurgical free tissue transfers is very high. However, follow-up studies necessitate reoperation in some patients due to compromised flap viability and also show the loss of that a few percent of the flaps due to flap failure.[1,3,4]

One challenge has therefore been to find a reliable way to monitor the viability of the flap with a high degree of sensitivity and specificity. Surgeons still much rely on clinical controls such as colour changes of the skin, changes in temperature and capillary blink. However, a number of different methods have evolved over the years to allow for more reliable and user-independent surveillance of flap viability.[5]

One of the most commonly used techniques for monitoring blood flow in microvascular flaps is laser Doppler flowmetry (LDF), which measures the microvascular perfusion by combining the concentration and velocity of red blood cells in the tissue. This method is limited by its small measurement volume, and the measured perfusion value is therefore subject to the large inherent heterogeneity of the microcirculation of the flap tissue, which in turn can produce false high or low perfusion values.[6–11]

Laser speckle contrast imaging (LSCI) is a camera-based technique that illuminates an area of tissue with divergent 785-nm laser light and analyses the interference pattern of the light that is scattered from the tissue. This makes it possible to measure perfusion in an area of tissue. The method has been used both in experimental settings to measure blood perfusion after controlled tissue provocations, including post-occlusive reactive hyperaemia, and clinically to assess perfusion in scald burns.[12–15]

Venous congestion has been shown to occur more frequently and to cause more deleterious effects on the tissue than arterial ischaemia.[16] However, most studies on flap perfusion have been focussing on arterial obstruction, rather than venous obstruction. As vessel thrombosis is likely to progress from partial to full occlusion, we designed this study to address partial as well as full venous obstruction. We used a porcine flap model that allows for controlled venous occlusion with the aim to compare LSCI measurements to single-point measurements with LDF.[17,18] The CGAP flap was designed to consistently include an area of compromised perfusion, in order to assess if LSCI could detect spatial variations in perfusion depending on the distance from the pedicle.

Materials and methods

Animals

Five mixed-breed pigs (mean age: 4 months, 45 kg, Swedish Landrace pigs) were used for the study. They were pre-anaesthetized with Dexdomitor (0.1 mg/kg), Zoletil (5 mg/kg) and atropine 0.05 mg/kg. Anaesthesia was withheld with pentobarbital sodium (8 mg/kg/h) and fentanyl (0.5 μg/kg/h) dissolved in Ringer's acetate given continuously intravenously with motorized infuser along with crystalloid fluids (Ringer's acetate). Body temperature, blood pressure, heart rate and oxygen saturation were monitored during the whole procedure. After the interventions and measurements had been performed on one side, the animal was turned and the procedure was repeated on the contralateral side. The pigs were finally euthanized with a mix of pentobarbital sodium and 70% ethanol without regaining consciousness.

Surgery

A 12 × 15-cm fasciocutaneous island flap based on the cranial gluteal artery perforator (CGAP) was raised from the buttocks of each pig (Figure 1). The flap was dissected along the surface of the muscle, including skin, subcutaneous tissue and muscle fascia in the flap. The pedicle of the flap, containing the perforator artery along with comitant veins, was carefully isolated and the sensory nerve cut. The comitant veins were separated from the artery and one of them was ligated. Then, perivascular flow probes (Transonic Precision Perivascular Flow Probe MA-2PSB Transonic Systems Inc. 34 Dutch Mill Road, Ithaca, NY 14850 USA) were placed around the vessels, after which the corners of the flap were sutured to the corners of the defect and wet swabs were placed on uncovered wound surfaces to keep them moist.

An inflatable vascular occluder (Norfolk Medical Products Inc., 7350 N. Ridgeway, Skokie, IL 60076 USA) was placed around the vein, distal to the flow probe, and fixated to the nearby tissue with sutures. A 2-ml syringe was used to inflate the cuff with saline to control the amount of occlusion applied to the vein.