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Willaert, Wouter; De Somer, Filip; Grabherr, Silke; D'Herde, Katharina; Pattyn, Piet

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Post-mortem Reperfusion of a Pig: a First Step to a New Surgical Training Model?

Wouter Willaert · Filip De Somer · Silke Grabherr · Katharina D'Herde · Piet Pattyn

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Abstract The purpose of this experimental study was to establish a short-term post-mortem circulation in a pig model using liquid paraffin. This study also investigated the quality of vascular perfusion in the peripheral tissues. This is the first step in the development of a new revascularized human surgical training model. This first experience was performed on the hind leg of a pig. Initial cannulation of the external iliac artery and vein was followed by connection of the arterial inflow to a heart–lung machine and using the venous outflow to flush post-mortem clots and blood. Subsequently, after connecting the venous outflow to the heart–lung machine, circulation was initiated. Circulation was established during 27 min, during which the flow was constantly 130 mL/min. A steady increase in inlet pressure was observed during the experiment, which finally reached a minimum value of 124 mmHg. Perfusion was interrupted early due to an uncontrollable fluid leak. Afterwards, the distal hind leg was incised showing an equal distribution of paraffin. A short-term revascularization was successfully re-established under excellent conditions. Although the results are promising, further

experiments are necessary to eventually perform a wide range of surgical procedures on revascularized human cadavers.

Keywords Heart–lung machine · Paraffin · Perfusion · Swine · Post-mortem

Introduction

In recent decades, there has been a notable trend towards minimally invasive surgical techniques, which require a long learning curve. Therefore, a wide range of learning models have been developed as an alternative to training on patients. These models include laparoscopic video-box trainers, virtual reality simulation, anesthetized animals and human cadavers [1–4]. However, surgical procedures performed on human corpses undoubtedly simulate best *in vivo* conditions [4].

In 2001, the first human cadaveric circulation model was reported. Although this model has its merits, only flow in the arterial anatomy was established [5]. Recently, a more realistic training model has been developed, using coloured fluid under static and pulsating pressure for arteries and static pressure alone for veins. A drawback of this model is the separation of both circulations [6].

Despite the usefulness of these circulation models, a realistic human training model must have a lifelike continuous flow in both the arterial and venous system. Grabherr has successfully re-established the total vascular circulation during a short period in deceased patients using a modified heart–lung machine. However, this technique has only been performed for experimental and diagnostic purposes in forensic medicine [7–9]. Previously, the same author effected a long-term post-mortem circulation in a cat and two dogs with odouriferous diesel oil [8]. Therefore, the aim of the current study was to establish a lifelike short closed post-mortem circulation in the hind leg and hemipelvis of a pig using odourless paraffin oil.

W. Willaert (✉) · P. Pattyn
Department of Gastrointestinal Surgery, Ghent University Hospital,
De Pintelaan 185, 9000 Ghent, Belgium
e-mail: wouter.willaert@ugent.be

F. De Somer
Department of Cardiac Surgery, Ghent University Hospital, De
Pintelaan 185, 9000 Ghent, Belgium

S. Grabherr
University Centre of Legal Medicine Lausanne-Geneva, University
of Lausanne, Rue du Bugnon 21, CH-1011 Lausanne, Switzerland

K. D'Herde
Department of Anatomy, Embryology, Histology and Medical
Physics, Ghent University Hospital, De Pintelaan 185, 9000 Ghent,
Belgium

Materials and Methods

Subject

This experiment was approved by the local ethics committee. We used the right hind leg of an adult pig connected to the hemipelvis. This specimen weighed 13.9 kg and was detached of the body in the slaughterhouse 3 h before by roughly cutting through the skin, pelvic bones and muscles, and iliac vessels. Initially, the circumference of the ankle was measured. Afterwards, the external iliac artery and vein were prepared and cannulated, while the tubes were affixed with Silkam 0 sutures (B. Braun, Tuttlingen, Germany) (Fig. 1).

Establishing a Post-mortem Circulation

This was followed by priming the circuit with paraffinum perliquidum (PP) (Sigma-Aldrich, Bornem, Belgium). Subsequently, the arterial tube was connected to a heart–lung machine (Cobe, Sorin Group, Mirandola, Italy). Next, a board-certified clinical perfusion scientist started the perfusion via the arterial tube. The presence of perfusate containing post-mortem clots and blood emerging from the cannulated iliac vein was considered to indicate a successful perfusion. Any leak from small vessels was adequately clipped or sutured. The venous tube was then connected to the heart–lung machine when the perfusate was free of clots and blood.

Afterwards, post-mortem circulation was established, during which leg circumference at the ankle (in centimetre), perfusion time (in minute), perfusion inlet pressure (in millimetre of mercury), flow rate (in millilitre per minute) and perfusion volume (in millilitre) were measured every 3 min. This circulation was successful if the perfusion parameters (inlet pressure and flow rate) reached a steady state.

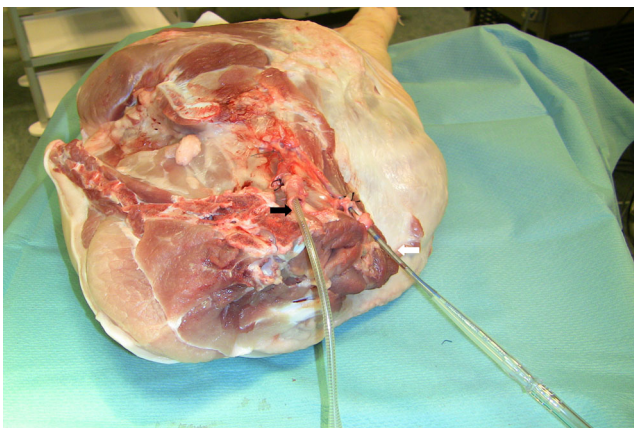


Fig. 1 The right hind leg of an adult pig connected to the hemipelvis. The tubes are inserted in the external iliac artery (*white arrow*) and vein (*black arrow*)

Macroscopic Inspection

Finally, after terminating the procedure, the skin and subcutis at the ankle were incised to evaluate the presence of PP.

Results and Analysis

Establishing a Post-mortem Circulation

Half a litre of PP adequately flushed most remaining blood and post-mortem clots. However, PP diffusely leaked from the raw muscle surface and sawn sacral bone marrow. This leak was uncontrollable but moderate and diminished after initiating circulation, which was effected during 27 min at a constant flow rate of 130 mL/min (Fig. 2). This circulation was interrupted early due to the persistent leak, causing a considerable reduction in circulating volume. The results of the perfusion parameters during this procedure are presented in Figs. 3 and 4.

The circumference of the ankle did not change. Moreover, the appearance of the external iliac vein remained physiological without signs of high wall tension (Fig. 2).

Macroscopic Inspection

Finally, after terminating the perfusion procedure, the distal hind leg was incised, showing equal distribution of PP in the tissues and some small amounts of remaining blood (Fig. 5).

Discussion

The purpose of this experiment was the establishment of a continuous lifelike closed post-mortem circulation in the right

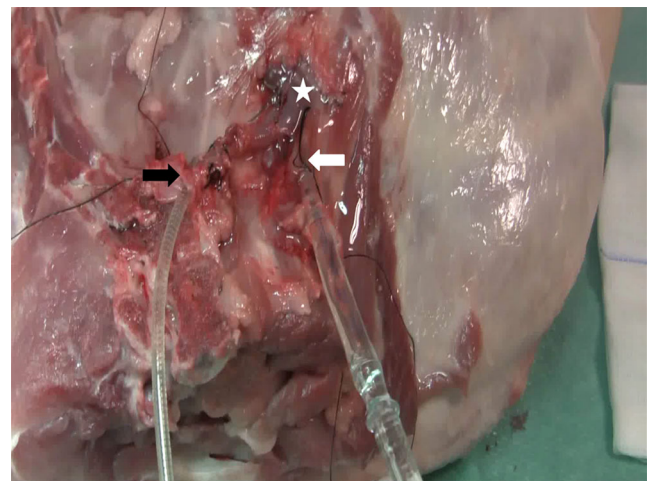


Fig. 2 The glossy raw surface of the muscles during the circulation due to diffuse leak of PP. The tubes are inserted in the external iliac artery (*white arrow*) and vein (*black arrow*). The asterisk shows physiological swelling of the external iliac vein

Pressure-Time curve

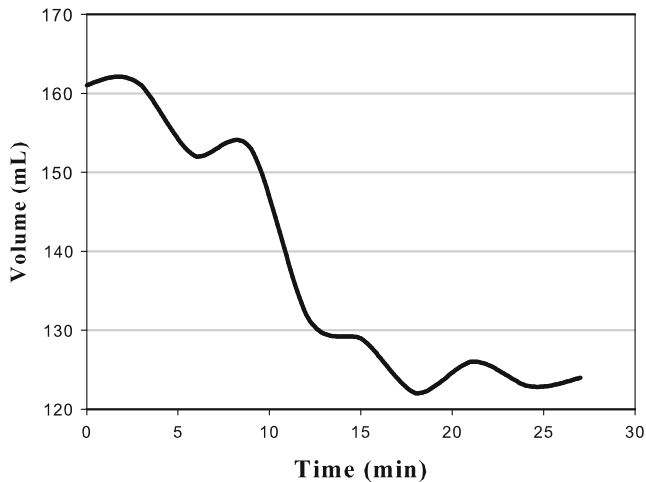


Fig. 3 A diminishment of the perfusion pressure from 161 mmHg at the beginning of the circulation to a steady state of 124 mmHg at the end. mmHg=millimetre of mercury

hind leg and hemipelvis of a pig. The hind leg model was chosen because its supplying iliac vessels have a considerable lumen, making an adequate flushing and subsequent circulation possible.

A persistent perfusion during 27 min was successfully effected. To our knowledge, this is the first report describing short-term pump-driven reperfusion of the total vascular system with PP. Similarly, diesel oil has been used to establish long-term post-mortem circulation, but its strong odour makes it unusable for surgical training models. Moreover, perfusion parameters are not reported in this experiment [8].

Remarkably, this study approaches unique lifelike conditions as the perfusion pressure, after an initial increase, diminishes during the circulation to reach a steady state of 124 mmHg. In accordance to the literature, this value will not damage the wall of the perfused vessels [10]. This finding

Volume-Time curve

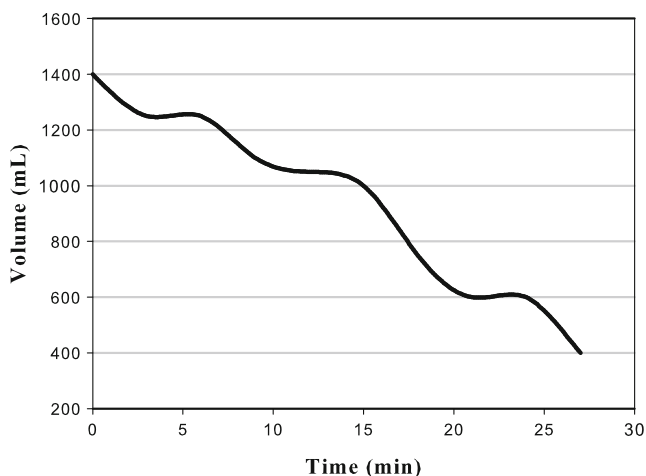


Fig. 4 A loss of 1,000-mL perfusion volume during the circulation. mL=millilitre

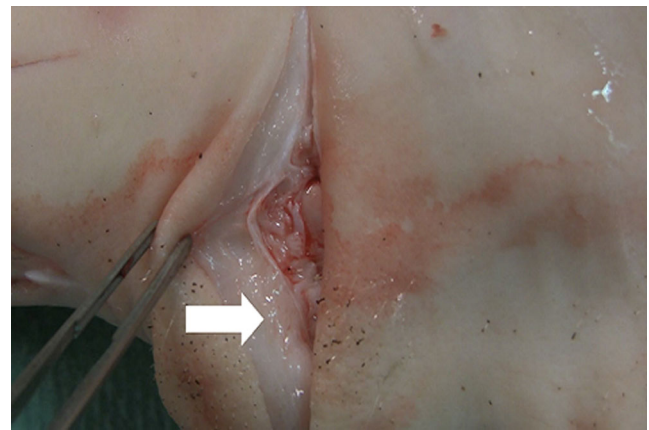


Fig. 5 PP and some blood at the distal hind leg. PP has a glossy appearance (white arrow). Oedema is absent

is affirmed by the physiological appearance of the external iliac vein and the absence of oedema at the ankle.

Not surprisingly, the constant flow rate of 130 mL/min during the circulation is lower than in human beings. Interestingly, the course of the perfusate through the tubes is clearly visible and fast, which is ideal for a future surgical training model.

The presence of PP at the ankle demonstrates its important properties. Nevertheless, some small vessels contain blood, which can be tackled by using less viscous perfusate. The viscosity of PP, which is 31 mPa·s (millipascal second, SI unit of viscosity), can be diminished by adding an alkane, resulting in perfusion of smaller vessels. Note that a perfusate with a viscosity lower than 18 mPa·s needs to be avoided because perfusion of the fenestrated capillary system results in oedema [11]. Establishing a circulation using a more viscous perfusate like paraffinum liquidum, which we previously tested in a pig model, is not a valuable alternative for PP because it necessitated too high inlet pressure resulting in low flow rates (data not published).

Unfortunately, the circulation was interrupted early due to loss of perfusion volume. The rate of this loss was lower during circulation compared with the flushing period, due to pump-induced suction in the lumen of the venous tube. This loss can have several causes, firstly, but probably least importantly, owing to recruitment of extra vessels during circulation. This phenomenon might play a role at the beginning but may be limited because it mainly involves small blood vessels with a higher opening pressure. Secondly, rupture of the vessels can cause leaks, which is unlikely because of the physiological appearance of the external iliac vein and the absence of oedema at the ankle. Thirdly, leaks at the level of the cutting edges of the muscle surface and the sawn sacrum are considered the main cause of the observed volume loss, which can be easily tackled by using a total body.

It should be borne in mind that PP is colourless, which is a disadvantage. Adding a fat-soluble red dye can colour PP to mimic blood [9].

A possible limitation of this experimental study is the use of only one specimen. However, post-mortem revascularization of

animals and full human bodies for a few minutes for diagnostic purposes has been reported in large series in the literature [7, 8]. Thus, the authors mean that successfully establishing a longer reperfusion in a small part of one body proves the possibilities of this experimental model making the use of a series of hind legs superfluous. Nevertheless, further research is necessary to investigate if PP circulates under the same conditions in an organ or the total body. Moreover, the microscopic circulation and possible adverse effects of prolonged reperfusion need to be evaluated.

In conclusion, PP enables a short-term reperfusion of the vascular system under ideal conditions. Both the heart–lung machine and PP can form a unique combination in the future development of revascularized human surgical training models enabling to make vascular reconstructions and to handle vessels during minimally invasive procedures in the thorax and abdomen.

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Conflict of Interest The authors declare no conflict of interest.

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