



Development of cadaver perfusion models for surgical training: an experimental study

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Abstract

Purpose Perfusion techniques on cadavers are heterogeneous and imperfect. The objective of this study was to improve the existing circulation model for surgical simulation on cadavers.

Methods We used a three-step experimental approach. The first part of the experiment tested two variables: the type of circuit and the use of a heater for perfusion. The second approach evaluated two parameters: the injection fluid and the type of body conditioning (embalmed or freshly dead prepared using different washing techniques). The third one was an improvement on the best circulation obtained, which focused on the injection fluid. To compare the realism of these different techniques, we constructed a score with realism parameters: the volume of return flow, the presence of peripheral venous return and the perfusion of abdominal arteries.

Results We found that the use of a heater seemed to improve the perfusion, while performing an arteriovenous bypass did not seem very effective. A correlation rate of 0.84 was found between the realism score and the injected fluid chosen. The best score (4/6) was found for a non-embalmed body with a low-pressure washing technique using a gelatin-based liquid at a concentration of 4 g/L for circulation. Scores obtained using embalmed bodies for both injection fluids for high-pressure washing or for 8-g/L gelatin injection fluid did not exceed 3/6.

Conclusions We showed that using a non-embalmed body with low-pressure washing and a 4-g/L gelatin-based fluid was the most effective technique for cadaver perfusion.

Keywords Perfusion · Cadavers · Surgical training · Simulation · Medical education

Introduction

Nowadays, surgical training makes use of a variety of tools, including medical simulation, which has become increasingly the norm [1]. Two types of surgical simulation techniques are currently used: organic and inorganic simulators [2]. Organic simulators group animal models and cadavers

from body donations, which are the most realistic [3]. Inorganic simulators group virtual reality models and synthetic dummies. Their realism is increasing and they tend to equalize animal models while simplifying them [4]. However, they are limited in that they do not represent fascia and fibrous spaces. Despite the progress of the few other simulation techniques including numeric models, the anatomic cadaver model remains an essential way to learn surgical procedures [5–9] during a surgeon's education, continuing education or when testing surgical devices.

The cadaveric model is limited in that it remains static. Therefore, over the past few years, many studies have examined this problem and the possibility of creating a vascularised and ventilated cadaveric model for surgical training, closer to the conditions encountered in the operating theatre. A literature review [10] presented the 12 main revascularization techniques: 58% of the models use coloured tap water with sodium chloride as a blood substitute. They often use freshly dead cadavers, often less

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than 24 h since death, a pulsatile flow and a single pump. However, the literature rarely describes the type of circuit used and the details of the technique used. Currently, the most advanced models are from Delpech et al. [11, 12] and Aboud et al. [13] and show good results concerning realism criteria: pulsatile flow, liquid colouration, body temperature and arterial pressure. In other words, these models' realism is based on a realistic colouration of the blood substitute that perfuses the trunk with a pulsatile pressure, artificial ventilation and a realistic body temperature. However, these two studies provide different models based on opposite techniques and today it seems difficult to objectively prefer one technique over the other. Furthermore, these models' complexity makes them difficult to reproduce, because they depend on a permanent simulation device and require many vascular surgical approaches. They often cause abdominal swelling and skin oedema [14, 15]. Finally, a lack of realism could be explained by the low viscosity of tap water used as a blood substitute compared to real blood [14]. In fact, there are presently no detailed data on the blood substitutes used for cadaver vascularization. We noted two liquids that are used or were used as a plasma substitute for filling during haemorrhagic shock, which also had a low manufacturing cost. The first one, made of denatured alcohol, sugar and sodium chloride, is inspired by a literature review by Kiel [16]. The second one, made of animal gelatin and sodium chloride, is inspired by a colloidal plasma substitute composition [17].

The purpose of this study was to develop a simplified revascularization model for cadavers, based on a review of the existing models, using an experimental approach with five parameters: the type of circulation, injection pressure, injection liquid, use of a heater and type of body (embalmed or non-embalmed).

Materials and methods

Study design

This study took place in three steps: the first one tested two variables, namely the type of circuit and the use of a heater for perfusion. The second step examined three other variables, namely the injected liquid, the pressure of injection and the type of body. The experimental design used a factorial experiment with these variables. A score was established at the end, to compare the efficiency of the circulation according to these parameters. Thirdly, according to previous results, the study focused on the best tested circulation to improve its efficiency. Only one variable was tested, the injected liquid.

Experimental protocol

During the first part of the study, a feasibility study, we experimented on a smaller scale, with non-embalmed upper limbs, in order to improve our perfusion technique and to ultimately apply them to the full body. Then, we experimented different vascularization circuits: arterio-venous ($n=4$), arterio-arterial ($n=3$) and veno-venous ($n=2$). The severity of the oedema was graded in 1, 2 or 3: respectively, no oedema, pitting oedema and non-pitting oedema with tensed skin. The reperfusion was stopped when the oedema was at the final grade or when there was no return flow for 5 min. Regarding setting the circuits, for each limb studied the first step of the experiment consisted in a careful dissection of the vascular pedicles of interest. As many vessels as possible were identified in order to cannulate those participating in the circuit, and to clamp those which were not. The arterial bed was systematically verified by inserting a Fogarty (Edward Lifesciences®) probe in the vessels of interest, allowing the disobliteration of the vascular lumen that could have been blocked by blood clots. For each circuit, two vessels were cannulated; the liquid was injected in one of them and was then collected at the end of the other one. Several cannulas with different diameters adapted to the size of the vessel were used to connect those vessels to a pump. During the experiments, the circuit evolved from a manual pump to an extra-corporeal pump (Medtronic® Bio-Medicus® 550 Bio-Console) set at 2000 rpm. A perfusion warmer was also added (Lotus Medical Equipment Limited® BFW-1000) set at 37 °C, making the model more realistic. For each type of circuit, the heater was used twice for the arterio-venous loop and once for the arterio-arterial and veno-venous loops. The injected liquid was tap water.

After identifying the best type of circuit and the utility of the heater in this first part, the study focused on other variables of interest. During the second part of the study, the two liquids were tested on complete cadavers: the first one was composed of powdered porcine gelatin (4 g/L) and sodium chloride (15 g/L), based on a colloidal plasma substitute composition [17]. The second one contained 90% denatured alcohol (100 mL/L), sucrose (white sugar, 150 g/L) and sodium chloride (15 g/L). Indeed, the use of a hypertonic solution limits the extravasation of the injected liquid based on a literature review by Kiel [16]. The liquid was also stained with methylene blue. For this part, we worked on non-embalmed bodies as well as embalmed bodies to assess the impact of body type. Three types of conditioning were used: non-embalmed bodies having undergone low-pressure (≤ 0.2 bar) washing before circulation, non-embalmed bodies having undergone high-pressure (≤ 1.5 bar) washing and embalmed

bodies that had undergone a washing technique performed by the anatomy laboratory's embalmer. We used two levels of pressure. These levels come from the two best perfusion models according to the systematic review [10]: a low-pressure model [12] and a high-pressure model [13]. For embalmed bodies, washing was performed before the experiment at a pressure of 0.5 bar. Their blood mass was extracted and replaced with a formaldehyde-based preservative (ARTHYL 26 HYGECO®). They were then kept at low temperature (± 4 °C) until they were used for circulation. For each dissection, four bilateral vascular surgical approaches were undertaken, at the femoral triangle in addition to the common carotid arteries and the common jugular veins. Endotracheal intubation was systematically tried. In case of failure, a tracheostomy tube was placed between the second and third tracheal rings and connected to an insufflation balloon in order to improve pulmonary perfusion [12]. Four tourniquets were placed at the humeral and femoral distal thirds to prevent perfusion of the limbs and to allow better control of thoracic and abdominal perfusion. The left common carotid artery was cannulated and connected to the pump. Given that the left common carotid artery is a direct aortic branch, it was selected to provide direct access to the systemic arterial circulation. The pump (Inotech®) was tuned to insufflate air; we also added a manometer to control the injection pressure between the pump and the injection jar.

For the non-embalmed body with a low-pressure wash (Fig. 1), washing was performed by injecting a large volume of liquid (± 10 L) into the left common carotid artery, at a low pressure (≤ 0.2 bar). The liquid used was water with

sodium chloride (15 g/L) at 38 °C. The fluid had been previously heated to 38 °C. The right and bilateral femoral arteries as well as the bilateral femoral veins and the right jugular vein were approached and then clamped. The left jugular vein was approached and incised to allow venous outflow. The stopping criteria during washing were a return of fluid observed at the three arteries incised, a 10-cm increase in the abdominal perimeter or an abundant flow of fluid through the upper airways.

For a non-embalmed body with high-pressure washing (Fig. 1), washing was performed at a pressure of 1.5 bar, in the arterial bed, then in the venous bed. At first, the arterial wash was done by a left common carotid artery injection of one of the injection fluids to be used for the circulation. The incision of the bilateral femoral arteries and the right common carotid artery allowed the evacuation of the blood present in the arterial bed. The arteries of the different approaches were gradually clamped as soon as a return of liquid was observed, which ensured more thorough washing. The stopping criteria of arterial washing were the same as before. Venous washing was done by injection into the left femoral vein. We chose a femoral vein to avoid flow resistance due to venous valves in the inferior vena cava. The right femoral vein and the bilateral jugular veins were incised to allow the evacuation of the blood present in the venous system. Finally, the circulation step was performed at pressures up to 1.5 bar. This was achieved by an injection in the left common carotid artery, with the other arteries approached clamped. The left jugular vein was used in discharge for the venous return and the others were clamped. During this stage, we used a laparotomy to examine the intestinal loop

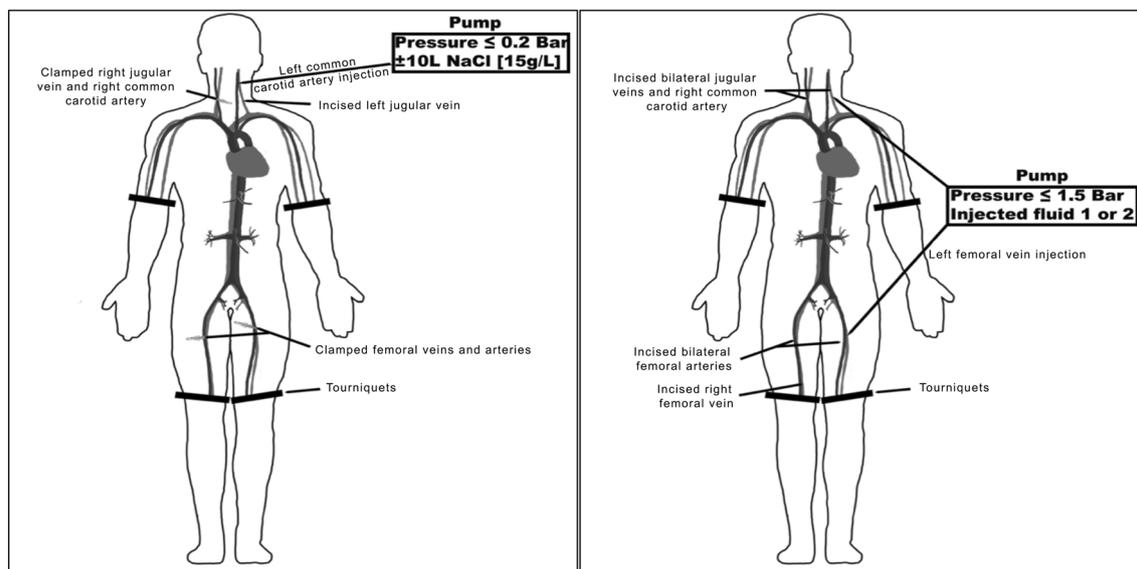


Fig. 1 Experimental designs of revascularization system. **a** Non-embalmed low-pressure washing. **b** Non-embalmed high-pressure washing. *art.* artery

colour and then intestinal arteries were sectioned to assess the perfusion quality.

For the third part of the study, we only used non-embalmed bodies with low-pressure washing, identical to the one described above, based on the first analysis. The circulation step was also identical, except for the injected liquid, which was composed of powdered porcine gelatin (8 g/L) and sodium chloride (15 g/L). This fluid was twice as concentrated in gelatin as our best fluid obtained in the first part.

Evaluation criteria

For the feasibility study in the first part, we assessed the quality of each circuit based on three criteria: the percentage of injected liquid that was collected at the end of the circuit, the presence or absence of subcutaneous oedema and the duration of effective reperfusion.

For the next steps in the full body, the effectiveness of cadaveric perfusion was evaluated according to a combined main criterion made up of four realism criteria:

- The collected liquid volume relative to the injected liquid volume. The volume injected was the whole liquid injected into the left common carotid artery during washing and circulation. The volume of liquid collected corresponded to the liquid collected by the outflow on the dissection table, added to the liquid collected by the suction pump used during the laparotomy.
- The peripheral venous return corresponded to the presence of an outflow, similar in colour to the injected fluid, by either the jugular veins or the femoral veins.
- The return from the branches of the superior mesenteric vein or artery corresponded to the presence of liquid outflow, similar in colour to the injected liquid, by the

branches of the superior mesenteric artery or vein (in particular, the jejunal and ileal arteries and veins).

- The intra-abdominal outflow was studied by the presence of liquid in the intra-peritoneal or retro-peritoneal cavities.

This experiment sought to test the hypothesis that the body type and the injection fluid had an impact on the quality of the circulation. We developed a score based on our evaluation criteria (Table 1), the goal being to compare these different circulations with each other. The evaluation criteria were considered positive when they favoured efficient circulation. They were considered negative when they indicated an outflow of injected fluid outside the arterio-venous network. The allocation of points was weighted according to the quality of the perfusion they represented. This score is composed of four criteria whose sum varies from −2 points to +6 points.

Statistics

We calculated the position and dispersion indicators of the descriptive variables. We measured the association between several variables if necessary with the Wilcoxon test. We also performed an analysis of the nominal and ordinal multimodal qualitative variables using a focused principal component analysis for exploratory purposes. Morphometric variables were also included in the analysis, such as weight, height and age, to look for predictors of successful circulation. All the statistical analyses were carried out using the RStudio software (version 1.0.143—© 2009–2016 RStudio, Inc.).

Table 1 Main characteristics of perfusions performed

	Model 1 (first part)	Model 2 (second and third parts)
Perfusion area	10 upper limbs	8 trunks (complete body)
Type of circulation	4 with arterio-venous circuit 3 with arterio-arterial circuit 2 with veno-venous circuit	8 with arterio-venous circuit
Heater	10 cadavers	8 cadavers
Injected fluid	9 with tap water 1 with Glycerin (63%) + NaCl (15 g/L) 1 with Gelatin (4 g/L) + NaCl (15 g/L)	3 with Alcohol (100 ml/L) + Sucrose (150 g/l) + NaCl (15 g/L) 3 with NaCl (15 g/L) + Gelatin (4 g/L) 2 with NaCl (15 g/L) + Gelatin (8 g/L)
Pressure	10 with high pressure	4 with high-pressure
Type of body	9 non-embalmed cadavers 1 embalmed cadavers	6 non-embalmed cadavers 2 embalmed cadavers

Legislation

The current funeral legislation governing the donation of the body was respected in this study.

Results

Population

The inclusion criteria of this study included bodies from the body donations that arrived at the anatomy laboratory of the Grenoble Medical School between August 2017 and January 2018. The non-integrity of the upper limb or of the trunk and the presence of scars were exclusion criteria.

We included 21 cadavers and excluded three because of the presence of scars. We performed 18 dissections: ten upper limbs and eight full bodies. There were a majority of females (66%). The median age was 83 years (± 6) and the median height was 154 cm (± 6).

First part

We calculated the association between the perfusion time and the presence of a heater with the Wilcoxon test that reported a *p* value of 0.046. Moreover, we calculated the median values of recovered liquid, oedema and perfusion duration for the arterio-venous, arterio-arterial and veno-venous loop circuits using the results obtained in experiments 1–10 (Table 2). The highest percentage of liquid return was observed for the veno-venous circuits (33%) followed by the arterio-arterial and arterio-venous circuits (6.25% and 5.2%, respectively). The oedema was the greatest in the veno-venous circuit and the lowest in the arterio-venous loop. The duration of reperfusion was the highest for the veno-venous circuit (both a median of 15 min) and the lowest for the arterio-venous circuit (7.5 min).

Second part

With regard to embalmed body circulation, the score showed no difference between the use of an alcohol-based

Table 3 Results obtained according to the different types of circuit in the first part of the study

Mean	Arterio-venous (<i>n</i> =4)	Arterio-arterial (<i>n</i> =4)	Veno-venous (<i>n</i> =2)
Liquid recovered	5.2%	6.25%	33%
Median oedema	2.5	2.75	3
Median perfusion time (min)	7.5	10	15.5

fluid and the use of a gelatin-based fluid. The two scores were equivalent and null. These results indicate that embalmed body circulation is not sufficiently effective (Tables 3, 4).

During circulation in non-embalmed bodies with high-pressure washing, the use of an alcohol-based fluid resulted in the lowest score, – 1. On the other hand, using the gelatin-based fluid, the score was 2.

Finally, using non-embalmed bodies with low-pressure washing, the use of an alcohol-based fluid gave a score equal to 1. The gelatin-based fluid gave the highest score (4).

Third part

With the third fluid (which was twice as concentrated in gelatin as liquid 2), we obtained the same score (3) for the two bodies we dissected (Table 3).

Synthesis

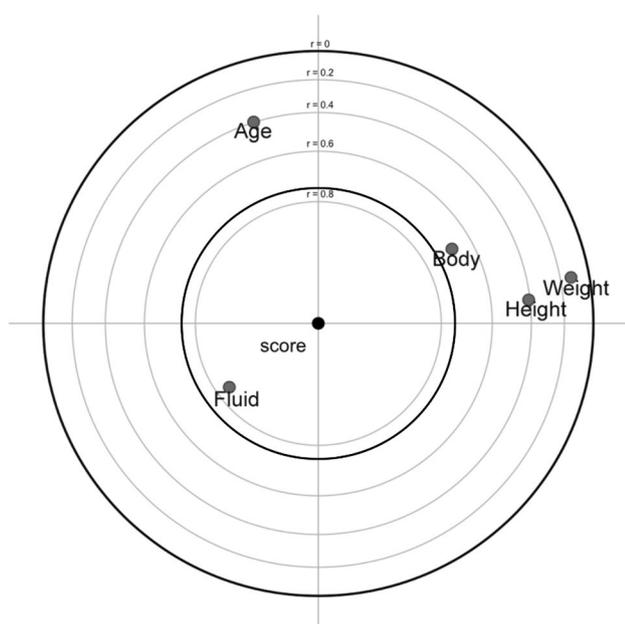
The principal component analysis, focusing on the score (Fig. 2), showed a significant correlation, at the threshold of 5%, between the score and the fluid injected (0.84). On the other hand, this analysis showed a correlation between the score and the type of body used (0.69), which means that the quality of the circulation is related to the fluid injected and the type of body. A relation was also established between the score and height (0.41), age (0.40) and weight (0.13), respectively.

Table 2 Score comparing the different models in the second and third parts of the study

Positive criteria	0 points	1 point	2 points
Volume of liquid collected	< 25%	25–50%	> 50%
Peripheral venous return	Absent	Different colour from injected fluid	Same colour as injected fluid
Branches of the superior mesenteric artery and vein return	Absent	Different colour from injected fluid	Same colour as injected fluid
Negative criteria	0 points	–1 point	–2 points
Intra-abdominal outflow	Absent	Different colour from injected fluid	Same colour as injected fluid

Table 4 Scores for the second and third parts of the study (liquid 1 corresponds to the alcohol-based fluid, 2 the gelatin-based fluid and liquid 3 to the gelatin-based fluid with double the concentration of liquid 2)

Body type	Fluid injected	Score				Score results
		Volume of liquid collected	Peripheral venous return	Return from the branches of the superior mesenteric vein or artery	Intra-abdominal outflow	
Non-embalmed with high-pressure wash	1	0	0	0	1	-1
Non-embalmed with high-pressure wash	2	1	2	0	1	2
Embalmed	2	2	0	0	2	0
Embalmed	1	2	0	0	2	0
Non-embalmed with low-pressure wash	2	1	2	2	1	4
Non-embalmed with low-pressure wash	1	0	1	2	2	1
Non-embalmed with low-pressure wash	3	1	1	2	1	3
Non-embalmed with low-pressure wash	3	1	1	2	1	3

**Fig. 2** Principal component analysis focused on the score

Discussion

The goal was to determine the best model for cadaveric perfusion for surgical simulation purposes because the models reported in the literature are heterogeneous and discordant. In response to this aim, the first part of the study showed that it was difficult to consider arterio-venous circulation as the most effective with the lowest perfusion time and lowest venous return. However, the association of the reperfusion time is significant with the heater ($p = 0.046$). In the second part, we showed that low-pressure washing in a non-embalmed body, using a

gelatin-based liquid at a concentration of 4 g/L for circulation, provides the best quality and realism for circulation (score 4/6). The correlation with the injected fluid is the most significant (0.84), but the increase in liquid viscosity, by increasing the gelatin concentration, does not improve the results (at 8 g L⁻¹ the score was 3/6). The lowest scores found were for the experiment on embalmed bodies (score 0) and on the high-pressure washed non-embalmed body (score -1).

The heater makes it possible to increase realism in the simulation, reduce extravasation and improve the return of liquid. To our knowledge, there are no data to explain this effect on the cadaver, but as in patients with hypothermia, the warming of the body seems to allow better perfusion. Furthermore, the type of circuit used is rarely precisely described in the literature, but two pumps are used in only 33% of cases, suggesting that in two-thirds of the cases an arterio-venous circuit is used [10]. This is why we took the arterio-venous model as a reference during the experiments, even if the first part of the study suggests that the circulation of liquid in the capillaries in post-mortem is difficult. It seems better to use a model that shunts the capillaries, by either performing a double-circuit (arterio-arterial and veno-venous loop), as done in the first part of the study, or creating an external shunt to reproduce a more physiological model replicating the increase in the resistance to the circulation of liquid in the capillaries. However, regardless of the type of circuit used, the liquid recovered did not exceed 33%.

The washing technique and the type of circulation fluid seem to be key values to obtain good-quality circulation, with the greatest correlation in the second part of the study. Indeed, the fluid viscosity, i.e. the fluid oncotic pressure, and ionic composition are the two variables that, according to the Starling laws, are important to control so as to avoid extravascular fluid leakage [18]. The injection fluid used in 58% of the studies was tap water and in 42% saline solution

[10]. The two most realistic studies according to the systematic review criteria used tap water (Delpech et al. [11, 12]) and saline solution (Aboud et al. [13]). The present study introduces a new liquid with higher oncotic pressure using macromolecules such as gelatin or glucose to prevent extravasation and oedema. In the first part of this study, tap water did not seem to be the optimal liquid. As for the perfusion pressure during the washing step, we also noted variability between the different studies in the literature, with low-pressure washing in the Delpech et al. model [11, 12] (0.4 bar) and high-pressure washing in the Aboud et al. [13] model (2 bar). The present study tends to show that low pressure provides greater realism. It is possible that low pressure preserves the capillaries better.

According to these results, it does not seem possible to use embalmed bodies with a conservation protocol. This protocol uses an excessively high injection pressure, which probably injures the microcirculation, creating extravasation and oedema, leading to abdominal leakage during the perfusion step. Moreover, the first embalmed corpse we perfused had been washed of its blood with an intra-cardiac puncture for aspiration of the blood mass, and this lesion caused substantial thoracic leakage during the perfusion step. To avoid this, the second embalmed corpse used in the study was prepared using femoral drainage, which avoided thoracic fluid leakage but did not limit abdominal fluid leakage. Nevertheless, we found a null score with these embalmed corpses, and it is important to note that the score is overestimated in these cases because it is calculated using the volume of liquid collected under the operating table and this liquid does not flow out of the vascular compartment, but rather the abdominal cavity. These results with embalmed corpses seem paradoxical, because 41% of the previous studies worked on embalmed cadavers, without reporting such issues [10, 12].

However, our best result is in line with the other articles on cadaver perfusion. As in the literature review by Bellier et al., the majority of laboratories use fresh cadavers for perfusion; the best score we obtained was with a non-embalmed body.

A strong point of this study is the comparison of different models with different circuit models, different types of injection liquid and different body preparation techniques in the same experiment. Furthermore, the score obtained contains very specific and objective variables, easily measured, which are intermediary criteria to assess the realism of the model. In the current state of knowledge, there is no score to assess the realism of a cadaveric model for surgical simulation and learning, which is why the score used herein gathers different realism criteria. The main weakness of the study stems from the fact that it is an experimental study with a small number of corpses. It forced us to use bodies that were not always fresh cadavers. The study did not overcome

inter-individual variability because it was not possible to do more than two experiments for each technique. We evaluated intermediary realism criteria, which do not directly represent the main objective of a simulation model, i.e. improving the learning of surgical techniques.

This study sought to improve the perfusion model in cadavers for surgical simulation. We showed that using a non-embalmed body with a low-pressure washing technique and a gelatin-based fluid is the most efficient technique for cadaver perfusion. The injected fluid is the parameter which correlates best with perfusion efficacy. The use of a heater also seemed to be advantageous, while the use of an arterio-venous circuit should be avoided in our opinion. Moreover, no studies, including ours, have evaluated the impact of the technique on surgical learning time by collecting feedback from surgical students. Extravasation on a cadaver has not been studied to our knowledge. Further studies could improve these shortcomings.

Authors' contribution AB: protocol/project development, data analysis, manuscript writing. JF: data collection or management, manuscript writing. QF: data collection or management, manuscript writing. SS: data collection or management, manuscript writing. CM: data collection or management, manuscript writing. PC: protocol/project development, manuscript editing.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflicts of interest.

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