Preservation of Cadavers for Surgical Technique Training

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Objective—To evaluate a technique for preservation of organoleptic tissue characteristics (color, odor, texture, and flexibility) in cadavers used for surgical instruction.

Study Design—Experimental study.

Animals—Forty-three canine cadavers.

Methods—Cadavers were preserved with a modified Larssen solution of the Hospital Cochim, Paris and cryopreservation. Tissue handling qualities were evaluated in surgical laboratory sessions. **Results**—All cadavers kept texture and tissues consistency, especially skin and muscle, similar to those of live animals. Some skin desquamation and pallor of the mucous membranes occurred with repetitive freeze–thaw cycles.

Conclusions—This preservation technique provides acceptable cadaver quality and tissue handling for use in surgical instruction.

Clinical Relevance—Preparation of patient cadavers by intravascular injection of modified Larssen solution yielded suitable instructional models for surgical training.

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Key words: cadaver, dog, embalming, Larssen solution, surgical instruction.

INTRODUCTION

I N MANY VETERINARY schools, fresh or preserved dog cadavers are used for surgical teaching. No significant differences in surgical performance have been identified between veterinary students who worked with live, anesthetized animals and those that worked with cadavers.¹ At the University of São Paulo, chemically preserved dog cadavers are used instead of live dogs for training in surgical technique. For training to be effective, it is desirable that cadavers simulate as closely as possible the tissue characteristics of live animals.

In teaching surgical techniques, satisfactorily preserved cadavers that most closely resemble living animals in their physical characteristics provide greater acceptance and improved learning. Our objective was to evaluate a technique for cadaver preservation that could be used for surgical instruction of veterinary students, that would retain organoleptic characteristics of live animals (color, odor, texture, and flexibility) and maximize repetitive use of cadavers for instruction.

MATERIALS AND METHODS

Several breeds of male and female dogs, of differing weights, that died in our veterinary hospital of causes except infectious disease or zoonoses were preserved.

To preserve the cadavers we used a modified Larssen solution. Sampaio² reported use of the original formula of Liquid of Larssen from the Hospital Cochim, Paris. This solution was composed of 500 g sodium chloride, 900 g sodium bicarbonate, 1000 g chloral hydrate, 1100 g sodium sulfate, and 500 mL of a solution of 10% formalin and 1 L distilled water. Sampaio used 1 part of this solution with 5 parts of distilled water.² Our modification was prepared by mixing 100 mL 10% formalin, 400 mL glycerol, 200 g chloral hydrate, 200 g sodium sulfate, 200 g sodium bicarbonate, 180 g sodium chloride, and 2 L distilled water. The working solution was made from 1 part concentrate and 3 parts distilled water, mixed at

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room temperature with a blender, and then stored in 11 L plastic containers.

Before fixation, cadavers were sanitized by removal of ectoparasites and dirt from the hair and skin, washing of natural orifices, and facilitated emptying of the intestinal tract by rectal enema with a jet of water. With the cadaver in a dorsal recumbency, an incision was made in the ventral neck region to expose the carotid artery and jugular veins, which were cannulated. Warmed (25–30°C) saline (0.9% NaCl) solution (corresponding to 10% of body weight) and modified Larssen solution were injected through the carotid cannula using gravity flow (fluid container 1 m above the cadaver and a flow of 10–15 mL fixative/min). The jugular cannula was used for egress of blood and preserving solution.

Immediately after saline perfusion, the vascular system was cleansed with modified Larssen solution (volume equal to about 5% of cadaver weight). The plastic container of modified Larssen solution was shaken beforehand so that any precipitate was re-dissolved before administration. Perfusion was maintained for about 1 hour after the solution initially exited from the jugular cannula. Then the jugular vein was ligated and a volume of modified Larssen solution corresponding to 10% of cadaver weight was perfused by gravity flow (10–15 mL/min) to preserve the cadaver; the carotid artery was ligated at completion.

After fixation and between each class use, each cadaver was stored in a plastic bag that was hung from iron S-hooks, in a walk-in freezer (-16° C to -20° C). The average storage was 4 months.

Cadaver Use in Surgical Technique Instruction

Cadavers were removed from the freezer 24 hours before class, thawed in tanks of water kept at ambient temperature $(11.3-34.7^{\circ}C)$.

Procedures performed by students were skin surgery (tension-relieving techniques and transposition flaps), ear surgery (aural hematoma, lateral resection of the vertical canal), oral cavity surgery (treatment of incomplete closure of the palate, mandibular fracture), cervical esophagotomy, resection of the mandibular/sublingual gland, tracheostomy, surgery of the eye and adnexa (enucleation, third eyelid surgery, entropion), and orchiectomy. Students also had the opportunity to practice aseptic patient and surgeon preparation.

Evaluation of cadavers and their tissue characteristics was done by observation and description during the training laboratories. Odor perception was evaluated by student and instructor comments.

RESULTS

Each cadaver was used 4 times, once for each surgical training laboratory, without emitting decomposition odors. Cadavers kept texture, color, and consistency of tissues like skin and muscles, similar to those found in live animals. The oral mucosa became paler with repeated freeze-thaw cycles.

In 2002 and 2003, we used 97 cadavers for surgical technique training. Three cadavers were pale and 1 had intense red tissue coloration at initial use. Five cadavers were pale at the 4th use. None had loss of joint movement and the limbs were easily manipulated during surgical exercises. By the 2nd week, some cadavers had desquamation in the abdominal and inguinal regions. This was observed in 9 cadavers during the 2nd week, in 12 cadavers during the 3rd week, and in 14 cadavers during the 4th week. However, because these regions were not used it did not affect the planned teaching exercises.

DISCUSSION

With increased moral and ethical concerns about the euthanasia of animals for instruction, various veterinary teaching centers have resorted to use of patient cadavers from veterinary hospitals. Successfully implemented, this has contributed much to minimizing euthanasia of otherwise healthy animals for didactic purposes and is more readily accepted by students as an appropriate animal or tissue source for their learning.³

Many veterinary students are sufficiently concerned about this issue that if the source of the animal and its cause of death is known, learning is less likely to be diminished by anxiety about these questions.⁴ In providing animals for instruction it is also important to consider the likelihood of diseases that might present health risks to students and instructors.⁵

In our instructional program, we use cadavers from dogs that died in our veterinary hospital from different causes but were free of contagion or zoonoses. Adequate preparation of cadavers that simulates living tissue was our goal because it permits intensive training in surgical techniques, creates a better learning environment, and teaches respect for life.

Cadaver preparation alternatives for instruction include fresh, fresh frozen, or embalmed, usually with formalin. Obtaining sufficient fresh cadavers for instructional use on a given day is problematic. The other traditional preparation techniques do not provide desirable tissue characteristics for surgical training. Thawed fresh frozen cadavers typically have increased tissue friability especially with repeated freeze-thaw cycles. Delicate tissues like intestinal and urogenital mucosa are typically not suitable for surgical training in thawed cadavers. Formalin-fixed cadavers have firm pallid tissues, are not pliable, and do not reflect normal tissue handling characteristics, but can be useful for surgical anatomy instruction.

For our surgical instruction classes, 40 cadavers are needed for each lesson. The high ambient temperatures of our tropical climate make use of fresh or refrigerated cadavers impractical. Chemical preservation by modified Larssen solution and freezing as a method of storage allowed cadavers to be used more than once. Repetitive use of these cadavers substantially reduced the number of the animals needed for teaching.

Larssen solution contains substances of sodium sulfate and chloral hydrate that are capable of dissolving and removing blood coagulates²; therefore, it is effective in cleaning debris from vessels. The carotid artery is the most effective peripheral access point for injecting liquid preservative.⁶

Formalin is a fixative that allows cadaver use for extended time without deterioration.⁴ Preservation of cadavers with formalin has been the classic approach for repetitive anatomic instruction in veterinary medicine. Besides an odor that many persons find objectionable and concerns about environmental pollution and personal health risk, formalin preservation alters tissue characteristics, flexibility, and coloration.⁷ Modifications we made in the preservative solution were the inclusion of glycerol for joint flexibility, and alterations in the ratio of substances and in the dilution of the working solution. The reduced formalin concentration in combination with other substances in this variant of Larssen solution allowed preservation and maintenance of organoleptic characteristics important for the practice of surgical procedures. We found these chemically preserved cadavers suitable for surgical technique training.

Embalmed cadavers can be stored at room temperature,³ but last longer when refrigerated ($\leq 4^{\circ}$ C). Recommendations for preservation at room temperature were developed in cold climates. Because we live in a tropical climate, we chose to keep cadavers cryopreserved at temperatures from -20° C to -16° C between each use, to retard deterioration. Maintenance of cadavers without preservation at low freezer temperatures is a traditional method for preservation and later dissection.⁸

Although the temperature of refrigeration $(2-4^{\circ}C)$ prevents ice crystal formation, it only preserves the most superficial structures and not the deeper muscular tissue and structures of both thoracic and pelvic limbs. The freeze-thaw cycles we used did not result in appreciable deterioration of gross structures, which remained suitable for surgical training.

One solution used to preserve tissue color and pliability is Klotz solution. Components common to Klotz and modified Larssen solution are formalin, sodium chloride, sodium bicarbonate, and chloral hydrate. A major difference is the concentration of formalin. Our modified Larssen solution contained 10% formalin whereas Klotz solution has 40% formalin. Modified Larssen solution includes sodium sulfate, which preserves tissue coloration, and glycerol, which is responsible for joint flexibility. Another difference in use of these 2 solutions is the fixation period. Specimens fixed by Klotz solution are typically immersed for 3–10 days, followed by 24 hours of rinsing in water then re-immersion in Klotz solution.⁵ By vascular perfusion of modified Larssen solution we were able to achieve more rapid preservation than can be achieved by immersion.

Esthetic preparation of a functional cadaver for surgical instruction is important to optimize the learning environment. We achieved our instructional objectives by using patient cadavers provided by clients of our teaching hospital. Removal of dirt and ectoparasites, and evacuation of the caudal digestive tract improved the esthetic acceptance of the cadavers by the students. Retaining organoleptic characteristics without any undesirable odor made achieving our surgical training objectives possible. We have used this preservation approach for 4 years in our curriculum and recommend the technique to others.

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