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A new model for thoracic radiology teaching and research in cadavers of embalmed dogs

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ABSTRACT: Nowadays, alternative methods that do not use formaldehyde to preserve cadavers must be used due to this substance's toxicity. Synthetic models are a training option for teaching, but cost makes their use unviable in most underdeveloped countries. The present study's main objective was to develop a model for education and researching thorax radiology in cadavers of chemically prepared dogs. Megaesophagus, pleural effusion, pneumothorax, and bronchography, as well as pulmonary insufflation, were simulated in 32 dogs cadavers, which received 150 mL/kg of pure ethyl alcohol solution with 5% glycerin followed by injection of 120 mL/kg of a solution containing 20% sodium chloride, 1% sodium nitrite and 1% sodium nitrate; they were kept refrigerated between 2 to 6 °C, for 30, 60, 90 or 120 days (G30, G60, G90, G120). There was no contamination, putrid odor, or subcutaneous emphysema. The pulmonary insufflation was kept, and the color and the consistency were similar to a fresh corpse after 120 days of conservation. It was possible to perform radiographic procedures, and almost all affections could be greatly mimicked. Megaesophagus and bronchography were easily simulated. Pneumothorax was the most challenging condition to be reproduced, especially in cadavers with some liquid in the thorax. The alcoholic and curing salt solutions are an embalming alternative with low financial and environmental costs and proved to preserve corpses.
Key words: anatomy, preservative solutions, radiography, training.

Um novo modelo visando ao ensino e pesquisa da radiologia torácica em cadáveres de cães quimicamente preparados

RESUMO: Nos dias atuais, métodos alternativos e que não utilizem o formaldeído para conservação de cadáveres devem ser empregados, devido à toxicidade desse agente. Modelos sintéticos são opção de treinamento para o ensino, mas geralmente o preço inviabiliza seu uso na maioria dos países subdesenvolvidos. O objetivo do presente trabalho foi desenvolver um modelo visando ao ensino e pesquisa da radiologia torácica em cadáveres de cães quimicamente preparados. Foram simulados megaesôfago, efusão pleural, pneumotórax e broncografia, além de insuflação pulmonar, em 32 cadáveres de cães, que receberam 150 mL/kg de solução de álcool etílico puro com 5% de glicerina seguido de injeção de 120 mL/kg de solução contendo 20% de cloreto de sódio, 1% de nitrito de sódio e 1% de nitrato de sódio, mantidos sob refrigeração entre 2 e 6 graus, por 30, 60, 90 ou 120 dias (G30, G60, G90, G120). Não houve contaminação, odor pútrido ou enfisema subcutâneo. A insuflação pulmonar foi mantida, e a cor e a consistência foram semelhantes a um cadáver fresco após 120 dias de conservação. Em todos os grupos foi possível realizar os procedimentos radiográficos e quase todas afecções puderam ser grandemente mimetizadas. O megaesôfago e a broncografia foram facilmente simuladas. O pneumotórax foi a afecção mais difícil de ser simulada principalmente nos cadáveres com um pouco de líquido na cavidade torácica. A solução alcoólica e de sal de cura são uma alternativa de embalsamamento com baixo custo financeiro e ambiental e comprovadamente conservam cadáveres.
Palavras-chave: anatomia, radiografia, soluções conservantes, treinamento.

INTRODUCTION

The use of cadavers is essential for teaching, and the concern regarding the conservation of anatomical specimens has existed for more than 5,000 years (CURY et al., 2013). Formaldehyde should be avoided because of the risk of developing cancer, allergic reactions, and teratogenesis (IARC, 2005).

At the University of Berlin, a solution with 23% curing salt, 30% ethanol, 20% Pluriol E400 (a mixture of polyethylene glycols), and 0.1% oregano oil were used as an alternative to the use of formaldehyde in the preservation of anatomical specimens (JANCZYK et al., 2010). The University of Compostela used a 0.9% saline solution through the common carotid artery and drained by caudal vena cava, and then conservation was performed in a vat with saline solution. The specimens could be preserved for an extended period, and they maintained a similar appearance and consistency compared to the fresh ones (LOMBARDERO et al., 2017).

This research developed a chemically prepared dog cadaver model for thoracic radiology,

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simulating thoracic conditions, bronchography, and lung insufflation, for education and research purposes.

MATERIALS AND METHODS

Thirty-two cadavers of dogs, male and female, adults, with body weight between 5 to 15 kg, non-obese, died from causes that did not involve evident morphological alteration, such as tumor masses or bone fractures, were used. The animals were frozen (freezer at -18 °C) after death and then transported to the Laboratory of Surgical Animal at UNESP Jaboticabal, SP, located 50 km away. They were defrosted in a horizontal refrigerator with a temperature (4-6 ºC) for 72 hours, shaved to avoid hair interference on thoracic X-rays, and then prepared by syringe infusion with 150 mL/kg of pure ethyl alcohol with 5 % glycerin and 150 mL/kg of a solution containing 20% sodium chloride, 1% nitrite and 1% sodium nitrate, via dissection and cannulation of the common carotid artery. The corpses were kept at 2 to 6 °C in a horizontal refrigerator, individually packed in transparent plastic bags, for periods of 30 days (G30), 60 days (G60), 90 days (G90), and 120 days (G120).

After the end of the conservation period of each group (30, 60, 90, or 120 days), the radiographic images were taken at the Veterinary Hospital of the São Paulo State University (Unesp), Jaboticabal, SP, Brazil, using a radiographic device (Tridoros 812 -E 800 mA, Siemens, Berlin), digitalized on a computerized device (CR 30-X, Agfa, Belgium) and then evaluated on a diagnostic monitor (MDRC-1119, Barco, United States). According to the animal size, the radiographic technique used in the radiographs ranged from 51 to 60Kv, ten milliamperes, and 3.2 seconds. The corpses were positioned in the lateral right and dorsal decubitus.

A simple radiograph was performed on all the cadavers before the simulation of the conditions and bronchography. The corpse's pulmonary insufflation was performed with an Ambu connected to the endotracheal tube, generating positive pressure to eliminate residual air through the chest tube. After pulmonary insufflation, the endotracheal tube was obliterated with hemostatic forceps to maintain positive pulmonary pressure. In sequence, to establish the negative pressure, the air was drained from the thoracic cavity using a 60 mL syringe connected to the thoracic tube. At the end of the drainage process, the tube was obliterated with hemostatic forceps to maintain negative pressure.

The first simulated condition was pneumothorax. For this, intercostal thoracotomy and insertion of the thoracic tube were performed. The corpses were positioned in the right lateral decubitus, palpation of the last rib was performed and, the 10th or 11th intercostal space was identified. A small incision was made in the skin on the dorsal third of the thoracic wall by introducing a number 14 Levine tube with a hemostatic clamp. A subcutaneous tunnel was created up to the cranial border of the 7th rib, aiming to promote negative pressure, mimicking the thorax of a living dog (KAHN, 2007). This technique was also used to simulate pleural effusion by the injection of a saline solution. A 300 mL of air was added to the thoracic cavity through the chest tube, using a 60 mL syringe. Subsequently, the radiographic examination and air removal with a syringe via the chest tube were performed to prepare for the second condition.

For the megaesophagus simulation, the dog corpse was positioned in the dorsal decubitus position, and an esophagotomy was performed. With the scalp, an incision of the ventral cervical midline was made in the raphe of the sternohyoid muscle, retracting and displacing the trachea to the right side, followed by esophageal location and dissection (KYLES, 2012). An esophageal incision was made to allocate a straw-type inflatable rubber balloon, involving a number 6 urethral tube and sealing this interface with adhesive tape to prevent air from escaping (Figure 1).

To simulate the megaesophagus, the rubber balloon was allocated inside the esophagus and insufflated using a syringe (approximately 120 mL), dilating the esophagus. After this procedure, a chest X-ray was performed.

After making simple radiographs, in the megaesophagus simulation, the contrasted radiography (esophagogram) was performed. For

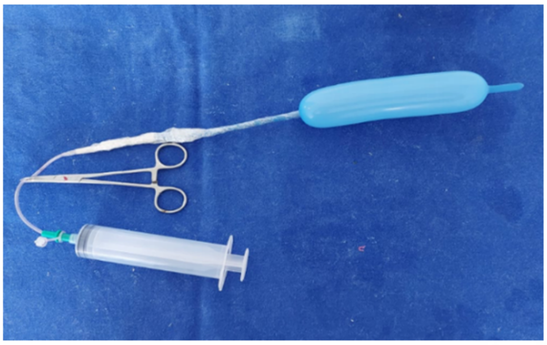

Figure 1 - Inflatable straw-type rubber balloon with air connected to a number 6 urethral probe and sealed with tape and hemostatic forceps to prevent air outflow.

this purpose, the air was removed from the inflatable balloon, and 120 mL of saline solution was injected with barium solution (100 mL of saline solution and 20 mL of Bariogel®), using a 60 mL syringe and a number six urethral probe. The pilot study observed that this proportion of 20 mL of contrast to 100 mL of saline solution (1: 5 ratio) was ideal after carrying out tests with the proportions of 1: 1, 1: 3, and 1:7.

Subsequently, the inflatable balloon was removed from the inside of the esophagus to simulate pleural effusion. With a 60 mL syringe, 300 mL of saline solution was injected into the thoracic cavity through the thoracic tube, and the radiographic image was performed.

After that, the bronchography was simulated. The lungs were inflated using an Ambu, and a tracheal tube was introduced by tracheotomy. The cadavers were positioned in the dorsal decubitus and, with a scalp, a median ventral cervical access was performed, between the fifth and sixth tracheal ring, a transverse incision through the annular ligament for allocation of an endotracheal tube, compatible with the size of the animal (numbering ranging from 3.5 to 5.5) and cuff insufflation (MACPHAIL, 2015). The 300 mL of saline solution previously injected into the thoracic cavity was removed using a 60 mL syringe connected to the thoracic tube. Through the endotracheal tube, 120 mL of barium solution was injected with the animal tilted upside down, followed by the radiographic images.

RESULTS

The alcohol and curing salt solutions effectively preserved the dogs' cadavers for up to four months under refrigeration. No apparent contamination, putrid odors, or subcutaneous emphysema were observed, which could suggest bad conservation.

On the 120th day of preparation, pulmonary insufflation was maintained and considered to be very good. The color and the lungs' consistency were similar to a live animal (Figure 2). Regardless of the preservation time (30, 60, 90, or 120 days), simulations of thoracic disorders and bronchography could be performed effectively on all cadavers.

Before simulating a thoracic disorder or a bronchography, negative pressure was applied, using thoracic or endotracheal tubes to inflate the lungs.

In animals with a more significant layer of subcutaneous adipose tissue, the lung tended to become more radiopaque due to the overlap of fat, as well as in cadavers with a little fluid in the lung.

Pleural effusion could not be faithfully simulated because, in live animals, the lung is radiolucent when that occurs, which cannot be seen in the simulations. However, it is possible to observe, radiographically, an increase in radiopacity due to the liquid in the thoracic cavity, with loss of visualization of the cardiac silhouette, as in live animals (Figure 3).

The Bronchography was also successfully performed. In only one corpse, the ventrodorsal radiographic exam allowed to view of the right hemithorax's pulmonary bronchi since the endotracheal tube was displaced caudally to the carina (Figure 4).

The megaesophagus was easily simulated. On a simple radiographic examination, the esophagus's dilation by radiolucent content (gas) was observed due to the air inside the balloon. In the contrasted investigation, it was possible to see a dilated esophagus due to radiopaque content (barium solution) (Figure 5).

DISCUSSION

This research developed a model for teaching and researching thoracic radiology in embalmed dogs.

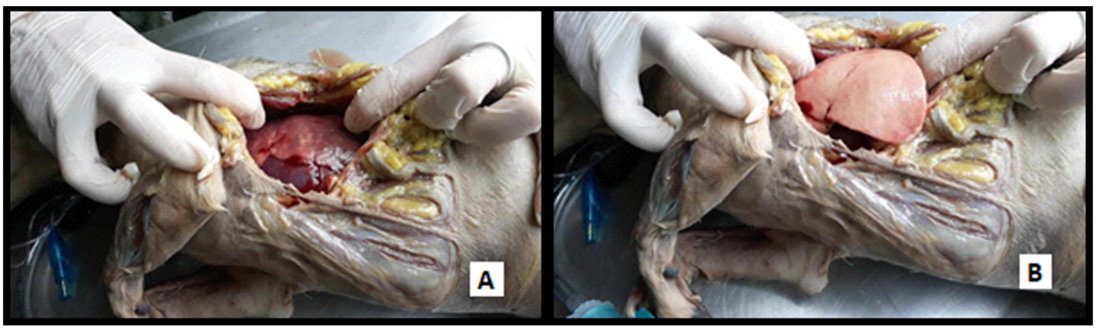

Figure 2 - Embalmed dog. Open thoracic cavity with uninflated (A) and inflated (B) lungs, after 120 days of storage in temperature of 2-6 ºC.

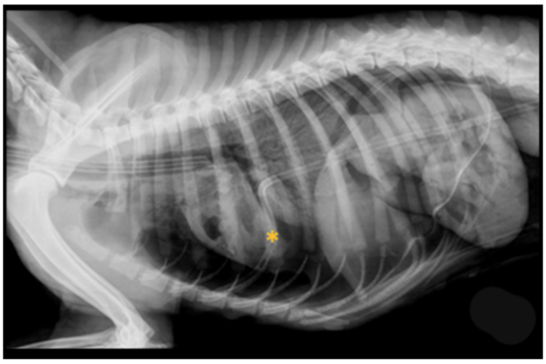

Figure 3 - Thoracic radiographic image of embalmed dog after 120 days, in right laterolateral projection, simulating pneumothorax by injection of air through a thoracic tube (yellow asterisk).

This method used ethyl alcohol with glycerin, followed by injection of a saturated solution of curing salt, which has been used for a long time in preserving meat because they delay the microbial or enzymatic action, preventing deterioration (PARDI, 1996).

The anatomical technique used proved to be efficient and present no risk to human health, such as formaldehyde would, and provided good practice with ethical corpses (CURY et al., 2013). It was an excellent alternative embalming method with low financial and environmental costs (JANCZYK, 2011) using alcoholic and curing salt solutions, which provided exemplary conservation and avoided deterioration (RODRIGUES, 2010; BALTA et al., 2015). Alcohol has been used in the preservation of dogs' and cats' corpses for up to four months, aiming the surgical training with excellent quality of the biomechanical characteristics of the skin and intestine (ROCHA et al., 2018; ZERO et al ., 2020), of the common carotid artery (CERQUEIRA et al., 2017) and the external jugular vein (PELOGIA et al., 2018).

The corpses could be kept for 120 days, a shorter but very satisfactory period of time for teaching and research, such as the 12 months presented by the anatomical technique at the University of Berlin (JANCZYK et al., 2010) and the five years at the University of Compostela (LOMBARDERO et al., 2017).

The pulmonary insufflation, megaesophagus, pneumothorax, pleural effusion, and bronchography could be simulated in the embalmed dogs for up to 120 days under refrigeration. It was essential for the training and veterinary teaching of thoracic radiology in small animals. The use of alternative methods for veterinary medicine study is currently recommended, as described on chemically preserved cadavers provided for surgical training (SILVA et al., 2003; HAYASHI et al., 2016).

Pulmonary insufflation in cadavers preserved for a long time has never been described in the literature, perhaps because most fixatives contain formaldehyde in their formulation, which causes tissue hardening (TOLHURST & HART, 1990) and affects the quality of soft tissues (WILKE et al., 2011), preventing the lings for insufflation. This research presents something new and essential to assist students and researchers, endotracheal intubation, and ventilation mechanisms in the lungs, which are necessary for residents or professionals in the practice of emergency medicine (SAGARIN et al., 2005). In veterinary medicine, establishing a safe airway via endotracheal intubation is the standard procedure for animals undergoing sedation, anesthesia, or cardiac-pulmonary-circulatory resuscitation (BRAINARD & HOFMEISTER, 2012).

Thoracic disorders such as megaesophagus, pneumothorax, and pleural effusion are common in the small animal routine and could be simulated in this study. Pneumothorax could be simulated. Radiographically, the loss of contact between the cardiac silhouette and the sternum and increased radiolucency of the thoracic cavity could be observed, as described in the literature (THRALL, 2013). However, it was more challenging to be performed on cadavers that presented liquid in the thoracic cavity.

Bronchography was the most uncomplicated technique to be performed with

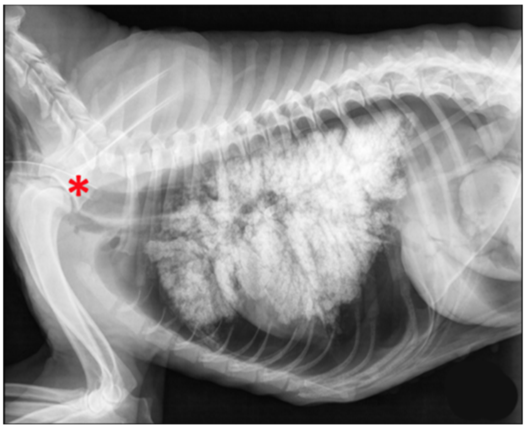

Figure 4 - Thoracic radiographic images of a embalmed dog after 120 days, in right laterolateral projection, simulating bronchography after injection, through the endotracheal tube (red asterisk), of a solution with barium.

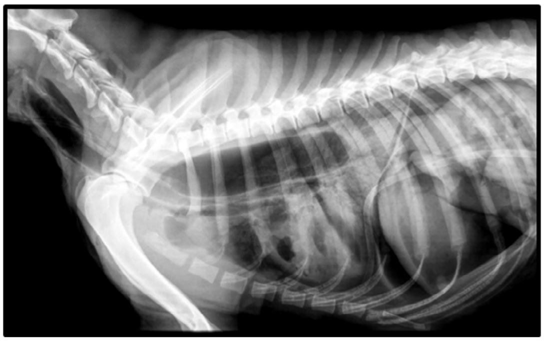

Figure 5 - Thoracic radiographic image of a embalmed dog after 120 days of conservation, in right laterolateral projection simulating megaesophagus. A number 6 urethral probe was inserted via cervical esophagotomy into a straw-type inflatable rubber balloon.

high quality due to the excellent lung expansion allowed with this conservation method. The bronchial tree could be studied as did in live animals (KILLNER, 1956). However, there are not many similar studies due to radiographic contrast, which can cause harmful effects on the organism. Currently, computed tomography and endoscopy are recommended in these cases (KEALY & MCALLISTER, 2012).

Among the thoracic disorders, the megaesophagus is the least common; the esophagus's dilation characterizes it by gas, food, or liquid (THRALL, 2013). When it is not altered, it is not visible radiographically. In this research, it was possible to dilate the esophagus with gas using a straw-type inflatable balloon. This technique was easily employed and low-cost and easy acquisition of materials.

CONCLUSION

The anatomical technique employed allowed the development of a teaching and researching model for thoracic radiology in chemically prepared dogs' cadavers, for up to 120 days, without using any toxic substances, making it possible to perform pulmonary insufflation and simulate pneumothorax, pleural effusion, megaesophagus, and bronchography in a very similar way to live animal.

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DECLARATION OF CONFLICT OF INTEREST

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

AUTHORS' CONTRIBUTIONS

All authors contributed equally to the conception and writing of the manuscript. All authors critically revised the manuscript and approved the final version.

BIOETHICS AND BIOSECURITY COMMITTEE APPROVAL

This study was approved by the Animal Ethics and Use Committee of the Faculdade de Ciências Agrárias e Veterinárias da Universidade Estadual Paulista (CEUA-FCAV UNESP protocol no. 004593/19). The study was carried out in accordance with the animal experimentation guidelines of CONCEA.

The corpses came from the Zoonoses Control Center of Ribeirão Preto, S.P., in a process already approved by the Municipal Legal Department (process: 02.2014.000027-1) and by the Institution's Ethics Committee (process: 004593/19).

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