CPQ Medicine (2019) 6:4
Research Article

Analysis of the Skin of Cats' Corpses Chemically Prepared with Ethylic Alcohol and Curing Salt Aiming Veterinary Surgical Practice - Chronic Effect on Biomechanics and Students´ Evaluation

Vinícius Camargo Fração, Raphael Chiarelo Zero, Alessandra Rodrigues, Beatriz Nogueira Ferreira, Alisson Diego Senna Fechis, Thiago André Salvitti de Sá Rocha, Maurício Theago Iozzi & Fabrício Singaretti de Oliveira*

Department of Animal Morphology and Physiology, São Paulo State University (UNESP), Brazil

***Correspondence to:** Dr. Fabrício Singaretti de Oliveira, Department of Animal Morphology and Physiology, São Paulo State University (UNESP), Brazil.

Copyright

Received: 11 April 2019

Published: 10 May 2019

Keywords: *Anatomy; Surgery; Teaching; Practice*

Abstract

Background

Various are the alternative methods that look for the animal's welfare in the veterinary surgical technique teaching, which aim to substitute the use of live animals, generating similar or superior learning to the students.

Aim

To determine, the maximum force and elongation for skin rupture in cats´ corpses chemically preserved with ethyl alcohol and curing salt, besides taking the evaluation of undergraduate students at a Veterinary School by performing incision and suture in those corpses skin.

Materials and Methods

For fixation, in eight cats´corpses, 96% ethyl alcohol solution containing 5% glycerin was infused via the common carotid artery. After, a solution constituted by curing salt (200g/l of sodium chloride, 10g/l of sodium nitrite and 10g/l of sodium nitrate) were injected the same way the alcohol. Corpses were kept under refrigeration (2° to 6°C) then. Control samples were collected before fixation and immediately submitted to biomechanical testing. In the second phase of this research, eight more cats were prepared using the same protocol of the first phase and undergraduate students of a Veterinary School performed incision and suture.

Results

The force and elongation necessary for skin rupture in control samples was 254.15±183.25N and 5.12±1.65mm, respectively. During conservation, they did not overcome 307.35±204.42N and 4.20±0.84mm, respectively. Statistics indicated difference among moments ($p = 0.0134$) but this difference occurred only among moments and not among moments and the control samples. Statistics did not point difference among scores in fresh and prepared corpses in incision ($p = 0.8055$) or even in suture ($p = 0.5022$).

Conclusions

There was a great score about tissue quality by students from the Veterinary Medicine Course as for using chemically prepared corpses for surgery practicing before performing surgical procedures on alive animals.

Introduction

Nowadays, various are the alternative methods that look for the animal's welfare in the veterinary surgical technique teaching, which aim to substitute the use of live animals, generating similar or superior learning to the students [1]. A negative emotional state may make it difficult more complex cognitive mechanisms, which means, disrupting a significant learning [2].

Cats´ corpses that were chemically prepared with ethyl alcohol and sodium chloride solution may be conserved until 8 months [3], besides being submitted to surgical training classes [4].

Several preservative solutions have been tested for surgical purpose. The Thiel solution (boric acid, ethylene glycol, potassium nitrate, (chloromethyl) phenol, sodium sulfate, and formaldehyde) [5], Klotz solution (sodium chloride, sodium bicarbonate, chloral hydrate, formaldehyde, and water), and Jores' solution (distilled water, formaldehyde, sodium sulfate, potassium sulfate, sodium chloride, sodium bicarbonate, glycerin, and sodium or potassium acetate) [6], are solutions that contain formaldehyde.

The use of satured sodium chloride solutions for preservation of an entire laboratory of animal anatomy was successfully evaluated [7] as well as for preservation of pericardium [8] and the diaphragm of dogs [9], both for surgical purposes.

Students´ acceptance was good even with solutions presenting formaldehyde during surgical training classes. The Laskowski´ solution was also recommended by students although corpses got so dark with it [10]. There was a great acceptance for first performing the training on chemically preserved cadavers and then on live animals, performing castration in population control programs of population control and 93.29% were in favor of this teaching method [11].

The use of cadavers for surgical purpose promotes greater learning and reduce costs, and allow for customization and repetitiveness of the exercise. The reduced stress during the practice surgical procedure benefits the whole process due increased student confidence and satisfaction, if the source of corpses is ethical [12].

There is a need to find an anatomical technique that preserves the body realistically for a long time, serving not only anatomical classes, but also surgical and clinical study, testing of new radiographic equipment and minimally invasive surgery [13].

The objective of this research was to determine, in cat´s corpses, the maximum force and elongation for skin rupture in corpses of cats chemically preserved with ethyl alcohol and curing salt, besides taking the evaluation of undergraduate students at a Veterinary School by performing incision and suture in those corpses´ skin.

Materials and Methods

Twenty-four adults weighting 3.58±0.63kg, male and female corpses were used, from the Center of Zoonosis Control of Ribeirão Preto, São Paulo, Brazil, in a process previous approved by the Legal Department (process 02.2014.000027-1). The animals were frozen (freezer at -18°C) after death and then transported to the Laboratory of Surgical Anatomy of the UNESP Jaboticabal, SP, located 50km away.

For fixation, in eight cats, 96% ethyl alcohol solution containing 5% glycerin was infused via the common carotid artery, at a rate of 150ml per kg. After, 120ml per kg of a solution constituted by curing salt (200g/l of sodium chloride, 10g/l of sodium nitrite and 10g/l of sodium nitrate) were injected the same way the alcohol. Corpses were kept under refrigeration (2° to 6°C) then.

Considering as first day the moment of collection of control samples, biomechanical analyses were taken on days 1, 15, 30, 45, 60, 75 and 90. With a scalper and a 1 x 4cm stainless steel mold, skin samples were collected. The cadaver was placed in right lateral decubitus (left antimere, facing upwards) and three sequential samples were collected transverse to the cat's skin tension line [8], on the lateral side of the thorax, parallel to and 4cm from the median plane. Shaving had been performed throughout the thorax. Control samples were collected before fixation and immediately submitted to biomechanical testing.

To evaluate tissue resistance, a Universal Testing Machine (EMIC® - DL 2000) was used, with a 500 N load cell and electromechanical drive support, with a speed of 100mm/min. Traction claws were also used by manual compression, in the Laboratory of Surgical Anatomy of the Department of Animal Morphology and Physiology of São Paulo State University (UNESP), Jaboticabal, Brazil.

In the second phase of this research, eight more cats were prepared using the same protocol of the first phase and undergraduate students of the third and fourth year at a Veterinary School performed incision and suture on those corpses´ skin in the thorax and abdome area, giving scores from zero (really bad) to 10 (excellent). Eight fresh cat corpses (unfrozen in the same day) were put aside for comparison because tissue texture was considered ideal on them.

The Kruskal-Wallis, Dunn and Mann-Whitney tests were used in statistics.

Results

Mean and standard deviation of the maximum force and elongation for skin rupture of the control (day 1) and 15, 30, 45, 60, 75 and 90-day samples are on table 1 and 2.

Table 1: *Mean and standard deviation of the maximum force and elongation for skin rupture in corpses of cats subjected to ethylic alcohol and curing salt fixation.*

Days	Maximum force MÁXIMA (N)
1	254.19±183.25
15	220.0±202.93
30	240.24±126.64
45	203.12±124.22
60	307.35±204.42
75	209.38±165.77
90	234.68±108.17

*Day 1: fresh corpses (control samples)

Table 2: *Mean and standard deviation of elongation for skin rupture in corpses of cats subjected to ethylic alcohol and curing salt fixation.*

Days	Elongation (mm)
1	5.12±1.65
15	2.98±0.74
30	3.62±0.49
45	3.22±1.00
60	4.20±0.84
75	3.73±1.19
90	4.07±0.75

*Dia 1: fresh corpses (control samples)

Data from the maximum force and elongation were subjected to Shapiro-Wilk test and ($P< 0.05$) and the distribution was nonparametric. The Kruskal-Wallis test indicated significative difference among moments ($p = 0.0134$). However, Dunn test presented this difference and it occurred only among moments and not among moments and the control samples (Table 3).

Table 3: *P value in Dunn test of maximum force for skin rupture in corpses of cats subjected to ethylic alcohol and curing salt fixation.*

Analysis	P value
Day 1 (control) vs. Day 15	0.5005
Day 1 (control) vs. Day 30	>0.9999
Day 1 (control) vs. Day 45	>0.9999
Day 1 (control) vs. Day 60	>0.9999
Day 1 (control) vs. Day 75	0.8155
Day 1 (control) vs. Day 90	>0.9999

*Day 1: fresh corpses (control samples)

There was statistical difference in elongation ($p < 0.0001$) and the Dunn test indicated that significative differences occurred among control and samples from days 15, 30, 45 and 75 (Table 4).

Table 4: *P value in Dunn test of maximum force for skin rupture in corpses of cats subjected to ethylic alcohol and curing salt fixation.*

Analysis	P value
Day 1 (control) vs. Day 15	<0.0001
Day 1 (control) vs. Day 30	0.0015
Day 1 (control) vs. Day 45	<0.0001
Day 1 (control) vs. Day 60	0.8477
Day 1 (control) vs. Day 75	0.0007
Day 1 (control) vs. Day 90	0.3904

*Day 1: fresh corpses (control samples)

On second phase of the research, cats´ corpses were chemically prepared and subjected to undergraduate students. Mean and standard deviation of scores in incision and suture are shown in tables 5 and 6.

Table 5: *Mean and standard deviation of incision scores.*

Corpses	Scores
Fresh	8.60±1.22
Chemically Prepared	8.72±1.10

Table 6: *Mean and standard deviation of suture scores*

Corpses	Scores
Fresh	8.88±1.24
Chemically Prepared	8.76±1.01

The Mann-Whitney test was used to compare the cadavers' evaluation and did not point statistical difference among scores in fresh and prepared corpses in incision ($p = 0.8055$) or even in suture ($p = 0.5022$).

Discussions

The preservation technique of this research has satisfied the desired requirements [9] for up to 90 days, in a realistic manner, for use in skin surgical training, similar to researches about biomechanical effects on skin and intestines [3], common carotid artery [14] and external jugular vein [15] of dogs' corpses during 120 days.

There was no skin hardening or great color change during the conservation, differently from what occurs when formaldehyde was used as fixative agent [7], and there was good tissue quality, like the report about human cadavers' fixation with alcohols up to one year [16].

In dogs weighting 7.6±2.7kg and prepared using ethyl alcohol in fixation for one to four months and sodium chloride solution for conservation during four months, the maximum force required to rupture the skin ranged from 106.7 N to 177.5 N (mean 142.1 N) [3]. In our research, in cats weighting 3.58±0.63kg, the force to rupture skin was greater and ranged from 254.19±183.25 (fresh/control samples) to 234.68±108.17 (after 90 days of conservation). This demonstrated the higher resistance do rupture of the cat´skin when compared to the dog´s weighting twice and it is related to elastic properties in the formers.

Our results in skin incision and suture were as good as the reported in surgical training in dogs 'corpses prepared using ethyl alcohol for fixation and tanks with the same substance for 30 days, followed by conservation with sodium chloride solution, in which students assigned a score of 7.32 ± 1.63 for tissue mailability and resistance for suture/incision [4]. Most of students approve the use of chemically preserved cadavers in the teaching of surgery, followed by practice on animals submitted for elective surgery at a Veterinary Hospital [3,10].

Formaldehyde limits the use of cadavers for surgical practice because it profoundly changes tissue resistance. Fresh cadavers cannot be used several times [5] but, in this research, each cadaver, chemically prepared with

no formaldehyde, could be preserved throughout 90 days, and be used for surgical training with good softness and tissue malleability. The conservation solution decreased skin elongation when compared to fresh samples but values were only 20% lower after 90 days, what is excellent for surgical practice.

Surgical training with chemically prepared cadavers is a worldwide trend, and almost all universities in Canada and the United States use alternative teaching methods to minimize the use of live animals for this purpose, and thereby preventing thousands of dogs from being euthanized [17].

Conclusions

There was biomechanical similarity in the skin of cats subjected to chemical preparation when compared to fresh samples after 90 days. Besides, a great score about tissue quality was pointed by students from a Veterinary Medicine Course in surgery practicing, suggesting that the anatomical technique used in this research was efficient.

Acknowledgement

The authors wish to thanks FAPESP, processes 2015/08259-9; 2017/17886-2 and Usina São João, Araras, SP, Brazil

Bibliography

1. Silva, R. M. G., Matera, J. M., Ribeiro, A. A. C. M. (2007). New Alternative Methods to Teach Surgical techniques for Veterinary Medicine Students despite the Absence of Living Animals. Is that an Academic Paradox? *Anat Hist Embryol., 36*(3), 220-224.

2. Paixão, R. L. (2008). Métodos substitutivos ao uso de animais vivos no ensino - Repensando o que aprendemos com os animais no ensino. *Ciência Veterinária nos Trópicos, 11*(Suppl 1), 88-91.

3. Rocha, T. A. S. S., Yanagihara, G. R., Shimano, A. C., Rolim, G. S., Santos, C. C. C., Fechis, A. D. S. & Oliveira, F. S. (2018). Biomechanical analysis of the skin and jejunum of dog cadavers subjected to a new anatomical preservation technique for surgical teaching. *The Journal of Plastination, 30*(1), 16-23.

4. Rocha, T. A. S. S., Santos, C. C. C., Iozzi, M. T., Dias, R. S., Zero, R. C., Cardozo, M. V. & Oliveira, F. S. (2019). Chemically prepared dog cadavers in teaching of surgical technique - evaluation by students of a veterinary medicine course. *Acta Scientiae Anatomica, 1*(2).

5. Groscurth, P., Eggl, P., Kapfhammer, J., Rager, G. J., Hornung, P. & Fasel, J. D. H. (2001). Gross Anatomy in the Surgical Curriculum in Switzerland: Improved Cadaver Preservation, Anatomical Models, and Course Development. *The Anatomical Record (New Anat)., 265*(6), 254-256.

6. Rodrigues H. (2010). Técnicas Anatômicas. Vitória-ES, Brazil. GM Gráfica & Editora.

7. Oliveira, F. S. (2014). Assessing the effectiveness of 30% sodium chloride aqueous solution for the preservation of fixed anatomical specimens: a 5-year follow-up study. *Journal of Anatomy, 225*(1), 118-121.

8. Brun, M. V., Pippi, N. L., Dreimeier, D., Contesini, E. A., Beck, C. A. C., Cunha, O., *et al.* (2002). Solução hipersaturada de sal como conservante de pericárdio canino utilizado na reparação do músculo reto abdominal de ratos Wistar. *Ciência Rural, 32*(6), 1019-1025.

9. Brun, M. V., Pippi, N. L., Driemeier, D., Contesini, E. A., Beck, C. A. C., Cunha, O., *et al.* (2004). Solução hipersaturada de sal ou de glicerina a 98% como conservantes de centros frênicos caninos utilizados na reparação de defeitos musculares em ratos Wistar. *Ciência Rural, 34*(1), 147-153.

10. Silva, R. M. G., Matera, J. M., Ribeiro, A. A. C. M. (2004). Preservation of cadavers for surgical technique training. *Veterinary Surgery, 33*(6), 606-608.

11. Silva, R. M. G., Matera, J. M. & Ribeiro, A. A. C. M. (2003). Avaliação de ensino da técnica cirúrgica utilizando cadáveres quimicamente preservados. *Revista de Educação Continuada do CRMV-SP, 6*(1), 1-3.

12. Knight, A. (2007). The Effectiveness of Humane Teaching Methods in Veterinary Education. *Alternatives to Animal Experimentation, 24*(2), 91-109. Kirpensteijn J., Haar G. (2013). Reconstructive Surgery and Wound Management of the Dog and Cat. Manson Publishing Ltd. London UK. (p.12).

13. Balta, J. Y., Cronin, M., Cryan, J. F. & O´Mahony, S. M. (2015). Human Preservation Techniques in Anatomy: A 21st Century Medical Education Perspective. *Clinical Anatomy, 28*(6), 725-734.

14. Cerqueira, E. S. F., Pelogia, M. E. S., Silveira C. P. B., Fechis A. D. S., Rocha T. A. S. S., Laus J. L. & Oliveira, F. S. (2017). Suture analysis and arterial traction test in dogs fixed on alcohol and preserved on saline solution aiming surgical practice. *Global Adv Res J Med Medical Sci., 6*(11), 292-295.

15. Pelogia, M. E. S., Cerqueira, E. S. F., Silveira, C. P. B., Rolim, G. S., Fechis, A. D. S., Rocha, T. A. S. S., Laus, J. L. & Oliveira, F. S. (2018). Suture and venous traction test analysis in dogs fixed in alcohol and preserved in saline solution. *Pesq Vet Bras., 38*(9), 1834-1837.

16. Goyri-o'Neill, J., Pais, D., Freire, F. A., Ribeiro, P., Belo, A., O'Neill, A., Ramos, S. & Neves, C. M. (2013). Improvement of the embalming perfusion method: The innovation and the results by light and scanning electron microscopy. *Acta Med Port., 26*(3), 188-194.

17. Balcombe, J. (2000). *The use of animals in higher education: problems, alternatives and recommendations.* Washington, DC: The Humane Society Press.