



QUANTITATIVE-QUALITATIVE EVALUATION BETWEEN THE USE OF MACERATED AND 3D-PRINTED VERTEBRAE AS AN ALTERNATIVE TEACHING METHOD FOR BONE ANATOMY

AVALIAÇÃO QUANTITATIVA-QUALITATIVA ENTRE O USO DE VÉRTEBRAS MACERADAS E IMPRESSAS EM 3D COMO MÉTODO ALTERNATIVO DE ENSINO DE ANATOMIA ÓSSEA

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ABSTRACT

The discipline of anatomy has long employed the use of cadavers as a teaching method. However, synthetic materials with no animal origin have gradually replaced this approach. Thus, the present study evaluated whether 3D-printed lumbar vertebrae are equally effective in teaching bone anatomy compared to cadaveric ones in dogs. Additionally, the acceptance of these synthetic bones in teaching and clinical-surgical routines was assessed. Two lumbar vertebrae from a dog were obtained from a cadaver through biological maceration, along with four synthetic vertebrae, two made from ABS (Acrylonitrile Butadiene Styrene) filament and two from PLA (Polylactic Acid) filament, acquired through computed tomography of the macerated vertebrae. To compare these vertebral models, three questionnaires were developed related to identifying and recognizing anatomical structures in three different vertebrae (two in 3D model and one natural dissected), comparative analysis between the models, and acceptance of the prototyped models in teaching. In total, 500 questionnaires were answered by students, postgraduates, professors, and tutors. Statistical analyses were performed based on nominal variables. The synthetic vertebral models were anatomically similar to the natural vertebrae. They were well accepted as an alternative teaching method in veterinary medicine, mainly due to the ability to manufacture the same bone for all students, enabling standardization in teaching. Among the synthetic models, the one made from ABS was more accepted than the PLA model, indicating that 3D printing of synthetic bones is a promising alternative for teaching bone anatomy.

Keywords: 3D Printing; anatomy; dissection; teaching; veterinary medicine

RESUMO

A disciplina de anatomia há muito tempo utiliza cadáveres como método de ensino. No entanto, materiais sintéticos sem origem animal têm gradualmente substituído essa abordagem. Assim, o presente estudo avaliou se vértebras lombares impressas em 3D são igualmente eficazes no ensino da anatomia óssea quando comparadas às vértebras cadavéricas em cães. Além disso, foi avaliada a aceitação desses ossos sintéticos no ensino e nas rotinas clínico-cirúrgicas. Duas vértebras lombares de um cão foram obtidas de um cadáver por meio de maceração biológica, juntamente com quatro vértebras sintéticas, sendo duas feitas de filamento ABS (Acrilonitrila Butadieno Estireno) e duas de

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filamento PLA (Ácido Polilático), obtidas por meio de tomografia computadorizada das vértebras maceradas. Para comparar esses modelos vertebrais, foram elaborados três questionários relacionados à identificação e ao reconhecimento de estruturas anatômicas em três diferentes vértebras (duas em modelo 3D e uma natural dissecada), à análise comparativa entre os modelos e à aceitação dos modelos prototipados no ensino. No total, 500 questionários foram respondidos por estudantes, pós-graduandos, professores e tutores. As análises estatísticas foram realizadas com base em variáveis nominais. Os modelos vertebrais sintéticos apresentaram semelhança anatômica com as vértebras naturais. Eles foram bem aceitos como método alternativo de ensino na medicina veterinária, principalmente devido à possibilidade de fabricar o mesmo osso para todos os alunos, permitindo a padronização do ensino. Entre os modelos sintéticos, o confeccionado em ABS foi mais aceito do que o modelo em PLA, indicando que a impressão 3D de ossos sintéticos é uma alternativa promissora para o ensino da anatomia óssea.

Palavras-chave: Anatomia; Dissecção; Ensino; Impressão 3D; Medicina Veterinária.

INTRODUCTION

The gold standard for anatomical teaching remains, in many countries, the dissection of cadavers in both medicine and veterinary medicine. It is considered the first connection students have with their patients. However, there are disadvantages to this method, such as the ethical acquisition of cadavers, preservation methods, and the need for appropriate storage locations, as well as the maceration of bone tissues, which leads to unpleasant odors and potential contaminants (Silveira et al., 2008; Rodrigues et al., 2012). Consequently, various universities have sought alternative methods to reduce using *ex-vivo* materials as teaching tools.

Among the available options, producing three-dimensional anatomical models made through additive layer manufacturing, also known as 3D printing or rapid prototyping (Valliyate et al., 2012), is the best substitute for using cadavers and their derivatives.

Originally developed to enhance and accelerate the production of objects by industry, rapid prototyping has been explored in medicine. This method, where computer-generated three-dimensional images are printed into physical models, has proven helpful for creating anatomical models from medical imaging examinations. It is effective for understanding the anatomy of surgical cases and for teaching students. It can produce precise and detailed





anatomical structures similar to natural models. Furthermore, while providing a tactile experience absent in virtual models, it avoids all the disadvantages associated with the use of cadavers (Hart et al., 2005; Ozkadif e Eken., 2012; Valliyate et al., 2012; Fredrieu et al., 2015; Li et al., 2018; Merino et al., 2018). Additionally, they do not emit unpleasant odors and, being non-animal-based, do not evoke negative feelings in students (Valliyate et al., 2012; Yushchenko et al., 2012). Thus, synthetic models currently represent, according to some authors, the best means for replacing the study of cadavers and ex-vivo materials, and they do not require special storage, facilitating the acquisition of a large stock of identical anatomical pieces (Abouhashem et al., 2015; Fredrieu et al., 2015; Li et al., 2018; Merino et al., 2018).

To obtain a printed anatomical model, it is initially necessary to acquire images through sensitive diagnostic modalities such as computed tomography (CT) and magnetic resonance imaging (MRI) (White et al., 2008). This study aimed to evaluate the effectiveness of manufactured (prototyped – 3D) vertebrae made from layers of ABS (Acrylonitrile Butadiene Styrene) and PLA (Polylactic Acid) as a method for understanding anatomy compared to natural vertebrae obtained through cadaver maceration.

MATERIAL AND METHODS

Ethics Approval

The project was approved by the Ethics Committee on Animal Use of the Faculty of Agricultural and Veterinary Sciences (FCAV, from Portuguese Faculdade de Ciências Agrárias e Veterinárias) at São Paulo State University (UNESP), Jaboticabal Campus, SP (protocol 017560/18), and the University of Franca (UNIFRAN), Franca, SP. As the study involved people's opinions, resolution 196/96 Brazilian National Health Council recommendations were followed. To this end, an informed consent form was created detailing the methodology and objectives of the study. This form was submitted to the Research Ethics Committee of





each university involved in the study and given to each participant, who only took part in the research after agreeing and signing the form.

Natural bones

Natural bones were obtained using the biological maceration process employed in the Department of Animal Morphology and Physiology at FCAV/UNESP. First, the soft tissues surrounding the lumbar vertebrae (L4 and L5) were dissected. Subsequently, these bones were moistened and stored in an airtight container for 30 days to allow microorganisms to proliferate and complete the decomposition of the remaining soft tissues. After this period, the bone specimens were boiled for five minutes in 10-volume hydrogen peroxide, diluted in water, and then exposed to the sun for 48 hours for drying, facilitating bone whitening and the highlighting of bone landmarks.

Synthetic bones

The natural vertebrae underwent CT examination (Toshiba Asteion Super 4 spiral) and subsequent 3D digital reconstruction using the 3D Slicer software. Printing was done using a Prusa I3 printer with extruded filaments of colorless ABS and PLA to match the color of the natural bones. The PLA had a pearlescent color and was less opaque, while the ABS had a beige tone. Four synthetic vertebrae were produced: two of the fourth lumbar vertebra and two of the fifth lumbar vertebra, each being made in both ABS and PLA (Figure 1). Two synthetic vertebrae, one of each material and one natural vertebra, were sent to UNIFRAN, while the remaining were sent to FCAV/UNESP for evaluation via questionnaires.





Figure 1 - (A) 3D reconstruction images of lumbar vertebrae L5 (left) and L4 (right) obtained from CT scans of the corresponding macerated vertebrae. (B) Photographic images of L4 vertebral models in PLA (left), macerated (center), and ABS (right). (C) Photographic images of L5 vertebral models in PLA (left), macerated (center), and ABS (right).

Questionnaires

Three distinct questionnaires were developed. The first contained basic technical questions and was answered by 200 undergraduate Veterinary Medicine students from UNESP and 200 from UNIFRAN (Students Group or SG). Questionnaire 2 consisted of opinion and technical questions related to using bone models in surgical procedures and was aimed at 50 postgraduates (residents, master's, and doctoral students) and faculty members from both universities (PGG). Questionnaire 3 included opinion and comparative questions between natural and prototyped bone models, intending to assess the impact of these models on 50 dog owners (TG). All the questionnaires assessed and compared precisely seven different structures: the vertebral body and the spinous, mammillary, transverse, accessory, cranial articular, and caudal articular processes.

The questionnaires can be accessed and viewed through the following QR Code:





Groups Description

The SG and PG groups were further subdivided to enhance the evaluation of results. The SG group had five subdivisions, each corresponding to a year of the undergraduate program: SG1 – responses from first-year veterinary students; SG2 – responses from second-year students; and so on up to SG5. The PG group was divided into four subdivisions based on the academic level of the participants: PGR – consisting of surgical residents from both universities; PGM – master's students; PGDT – doctoral students; and PGDO – responses from faculty members in the surgery departments at UNESP (Jaboticabal) and UNIFRAN.

Statistical Analysis

Nominal qualitative variables characterize the research data. Thus, the percentage of “yes” and “no” responses was calculated for presentation and comparison through graphs for each group and subgroup.

RESULTS

Regarding the undergraduate students participating in this study, 99.50% agreed with the use of cadavers in veterinary anatomy teaching, and 95.00% believe that this practice does not go against animal rights, as educational institutions regulate it. This high acceptance of the traditional teaching technique, combined with the limited alternative methods described in scientific literature, is reflected in the fact that only 54.25% of students believe universities should seek replacement methods. In this regard, analyzing the categories, it was observed that most first- and second-year students opposed the adoption of alternative teaching methods, representing 56.82% and 69.23%, respectively, differing from the other years, where most favored replacement.





Despite these results, 14.50% of students are afraid of contracting a disease when handling materials from cadavers, and 10.75% experience negative feelings when handling cadaveric specimens. Thus, when presented with synthetic vertebral models, 74.25% stated that they felt more comfortable knowing that no animal had died previously to obtain the prototypes, and 94.75% felt safer regarding their health when handling synthetic material. Additionally, 53% of students believed that synthetic models have a better odor than natural vertebrae; 69.50% thought that synthetic vertebrae could replace cadaveric specimens in teaching the skeletal system of veterinary anatomy, and 95% believed that 3D printing would enhance anatomical learning by providing a sufficient quantity of standardized pieces for all students and enabling the creation of prototypes for different species and breeds.

When conducting a thorough inspection of the synthetic and macerated vertebrae, each vertebral structure could be identified and compared between the models, aiming to evaluate the degree of accuracy and detail of the 3D printing and whether this production method would impair the identification and fidelity of bone landmarks. All seven structures were evaluated with at least 74.25% similarity between synthetic and natural models, with the caudal articular process being considered the least similar (74.25%) and the spinous process the most equivalent (83.75%) to its counterpart in the macerated vertebra. At the end of this comparative analysis, 70.75% of students believed that 3D printing of dog lumbar vertebrae would not impair learning.

Comparing the two synthetic models, 94% of students found a similarity between the ABS and natural vertebra, while only 49% found the PLA model similar to the natural vertebra. In this context, 73% of the SG found differences between the two synthetic models during the identification of vertebral structures, with the ABS vertebra being superior to the PLA in the quality of the three-dimensional printing of all seven evaluated structures.

For veterinarians, 62% believed that using vertebrae from cadavers is essential for teaching veterinary anatomy, unlike faculty members, where 62.50% consider other teaching methods equally effective. Among this group, 28% of PGG members are afraid of contracting a disease, and 6% have negative feelings associated with handling cadaveric materials.





Despite these results, 80% of veterinarians believe that using standardized anatomical pieces for all students would provide uniformity in teaching. Thus, 82% thought synthetic models could replace cadaveric bone structures in veterinary anatomy teaching, with 87.50% of faculty members and 75% of doctoral students supporting this replacement.

Additionally, 51% of the PGG believed that prototyped models have a better odor; 95% feel safer regarding their health when handling synthetic pieces, and 72% feel more comfortable knowing that no animal had died before obtaining these anatomical structures. A variation between 58% and 78% was similar to the macerated vertebra, with the cranial articular process being the least similar and the transverse process the most equivalent. At the end of this analysis, 78% of veterinarians, including 100% of faculty members, believe that using synthetic vertebrae would not impair learning.

When comparing the two synthetic models, 86% of the PGG found differences when identifying vertebral structures. 94% found the ABS model similar to the natural vertebra, while only 38% found a similarity between the PLA and natural models. Therefore, the ABS prototyped vertebra was superior to the PLA in all structural evaluations.

The results also revealed that 80% of the PGG would feel more comfortable using synthetic vertebrae instead of those from cadavers during consultations with owners; 96% believe that the synthetic model would help plan neurosurgeries and could minimize transoperative errors; 94% thought that these models could be used for surgical training, and 100% believed in the feasibility of using 3D printing to create drilling guides.

According to the owners, 98% agreed with the use of cadavers in veterinary anatomy teaching; 86% did not experience any negative feelings when handling cadaveric pieces, and 96% responded that this practice does not go against animal rights, as it is regulated. However, 64% stated that universities should seek new teaching alternatives. Furthermore, 44% responded that prototyped vertebrae are safer for health, and 41% noticed that they have a better odor than the macerated model. Only 70% of the TG group saw a similarity between the PLA and natural models, while 94% found the ABS model similar to the macerated vertebra. This resulted in 70% of the owners not noticing significant anatomical differences





between the models that could impair veterinary teaching. Finally, 58% would approve of receiving explanations about spinal conditions with the help of a macerated vertebra, but this acceptance increased to 66% when involving a synthetic vertebral model.

DISCUSSION

Traditionally, the teaching of veterinary anatomy has relied on the use of cadavers (Albanese, 2010; Lombardi et al., 2014), and this practice, considered an educational cornerstone (Kot et al., 2013; Fredrieu et al., 2015), continues to be accepted both within and outside the academic environment. This is supported by the data from this study, where 99.50% of undergraduate students and 98% of dog owners agreed with the use of cadavers in teaching, and 95% of students and 96% of owners believed that this practice does not violate animal rights, as it is conducted in a regulated manner.

In 1959, William Russel and Rex Burch proposed that if animals were used in experiments, every effort should be made to replace them with non-sentient alternatives, minimize their use, and refine experiments to ensure minimal pain and distress. These principles, known as the “3Rs” (Replacement, Reduction, and Refinement), initially received little attention. However, over time, there has been a shift in mindset, and these principles have become established not only in animal research but also in various legislations, ethics committees, and public opinion (Flecknell, 2002). This ideological shift has led to a growing repulsion toward using animals in teaching and research, contributing to a negative sentiment associated with this practice, as observed in this study. Furthermore, more than half of both groups in this study supported adopting alternative teaching methods at universities.

Over the years, alternative methods to using cadavers in veterinary anatomy teaching have been developed and implemented. Initially, anatomical models were created, once simplistic representations made of wood, ivory, papier-mâché, plaster, or wax (Fredrieu et al., 2015). However, the lack of detail and similarity to cadaveric models did not encourage the adoption of these methods in teaching. Thus, until now, alternative approaches to using





cadavers in teaching anatomy have not been widely accepted by the academic community. This may explain why only 54.25% of undergraduate students in this study supported alternative means to replace cadaveric specimens and why most postgraduate students (residents, master's, and doctoral students) believe that using cadaveric bones is essential for teaching anatomy. In contrast, faculty members presented a broader, more versatile, and up-to-date perspective on learning, resulting in a majority favoring the use of new teaching methods, possibly due to their experiences in classrooms and research projects.

Computational methods involving virtual dissection software have been widely used by various institutions (Drake et al., 2009; Sugand et al., 2010; Khot et al., 2013), requiring minimal maintenance and occupational infrastructure, being accessible, hygienic, and meeting the needs of each student at any time (Pandey et al., 2006; Albanese, 2010; Khot et al., 2013). In recent years, however, three-dimensional anatomical models produced through rapid prototyping have gained more prominence. For some authors, this has proven to be the best replacement for cadavers in teaching (Fredrieu et al., 2015; Li et al., 2018; Merino et al., 2018), and this study also revealed such a trend. After being introduced to synthetic vertebral models and comparing them with natural ones, most interviewed groups believed that prototyped models could replace cadaveric anatomical structures without impairing learning. Additionally, most of each group felt safer regarding their health and more comfortable knowing that no animal had died before obtaining these models.

Different 3D printing methods have been developed, and various materials can be used to construct three-dimensional anatomical models. The choice of material should be based on aesthetic, structural, and practical parameters. Therefore, resolution, strength, color, flexibility, opacity, stability, and cost should be considered when constructing medical education specimens (Fredrieu et al., 2015). According to the results of this study, almost all interviewed individuals found a similarity between the ABS models and the natural (macerated) vertebra. In contrast, fewer individuals saw the same similarity between the PLA models and the macerated vertebra. This discrepancy was likely associated with the color of the materials used in 3D printing: the PLA used was bright. At the same time, the ABS was





beige and more similar in appearance to a natural vertebra. Thus, the aesthetic parameters directly influenced the results, with ABS being better accepted by the respondents.

Additionally, it was found that some anatomical structures of the prototyped vertebrae had less definition than natural bones, with the caudal articular process being the least similar, according to the students. This reduced definition of small anatomical structures in the vertebrae is likely related to the slice thickness used in the computed tomography (CT) examination (Hespel et al., 2014; Mulford et al., 2016; Gümperlein et al., 2018; Hamilton-Bennett et al., 2018; Giannatsis and Dedoussis, 2009). The 2 mm slice thickness may have been insufficient for defining small anatomical details, such as the caudal articular process. However, it was adequate for generating good definitions for more considerable anatomical information.

In addition to serving as substitutes for cadavers in anatomy teaching, prototyped anatomical models can also be used in surgical practice to plan and train techniques (Anderson et al., 2016; Xu et al., 2016). In this context, almost all veterinarians interviewed in this study are aware of and value the use of this new technology in the surgical field, aiming to use all available methods to enhance further their knowledge and practices for the benefit of their patients.

CONCLUSION

The traditional use of dog cadavers in anatomy teaching remains an accepted practice both within and outside academic settings. However, synthetic anatomical models manufactured through rapid prototyping are promising alternatives to this practice. This study revealed that 3D printed bones were well accepted as an alternative teaching method in veterinary medicine. Among the synthetic models, the ABS model was more favored than the PLA model.





Statements and Declarations

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Conflict of Interests

The authors had no conflicts of interest.

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