Int. J. Morphol.,
37(3):838-844, 2019.

Volumetric Computed Tomography Reconstruction, Rapid Prototyping and 3D Printing of Opossum Head (*Didelphis albiventris*)

Reconstrucción Volumétrica por Tomografia Computarizada, Prototipado Rápido e Impresión 3D de Cabeza de Zarigüeya (*Didelphis albiventris*)

Catia Helena de Almeida Lima Massari[1]; Ana Carolina Brandão de Campos Fonseca Pinto[2]; Yuri Karaccas de Carvalho[3]; Adriano Ferreira Silva[1] & Maria Angélica Miglino[2]

MASSARI, C. H. A. L.; PINTO, A. C. B. C. F.; DE CARVALHO, Y. K.; SILVA, A. F. & MIGLINO, M. A. Volumetric computed tomography reconstruction, rapid prototyping and 3D printing of opossum head (*Didelphis albiventris*). *Int. J. Morphol., 37(3)*:838-844, 2019.

SUMMARY: Natural anatomical pieces of wild animals are rare and teachers seek alternatives in satisfactory quantity and quality to inform their students. This article aims to describe the use of multiplanar reconstructions and 3D volume rendering computed tomography (CT) images, rapid prototyping and 3D printing of opossum head to create a biomodel to veterinary education in descriptive anatomy of wild animals. A six-step method study was conducted to construct the biomodel: (1) selection of opossum head from museum; (2) CT scanning of bones structures in veterinary hospital; (3) DICOM visualization medical images in multiplanar reconstructions and 3D volume rendering; (4) .dicom file conversion to .stl; (5) 3D printing of opossum head by rapid prototyping; (6) comparison of 3D model printed with the original anatomical piece. The use of CT images with their different forms of reconstruction can provide a more comprehensive 3D view of opossum craniofacial region and allow a better understanding of head anatomy of this species. The 3D printed biomodel can be a viable alternative to original bone specimens when used in anatomy education. However, further studies must be continued to validate the method in Veterinary Medicine courses.

KEY WORDS: Veterinary Education; Anatomy; Biomodel; 3D Printing; Opossum.

INTRODUCTION

The white-eared opossum, *Didelphis albiventris*, can be considered an excellent model for comparative embryology in developmental biology studies. The group Metatheria (Didelphimorphia) has relevance among mammals, particularly related to reproduction aspects, as they represent an important transition link between Prototheria and Eutheria groups.

But it is not so easy to study the opossums today because of the difficulty to obtain animal corpses in many veterinary universities. Most natural anatomical pieces of wild animals are rare and teachers seek alternatives in satisfactory number and gross peculiarity to inform their students. Fortunately, the traditional method of teaching has undergone innovations time after time. The use of diagnostic imaging in order to develop virtual anatomical models and the use of 3D printing to objectify through rapid prototyping these scanned models is a trend in veterinary anatomy teaching. This didactic alternative is not substitutive for the dissection based teaching but a complement to the traditional method of teaching-learning in anatomy (Massari *et al*., 2018).

The available literature suggests that both digital and physical scale models of animal skeletal components can be rapidly produced by using 3D printing technology (Li *et al*., 2018).

Understanding the three-dimensional nature of wild animal form is imperative for an effective medical practice

[1] PhD Student in Anatomy of Domestic and Wild Animals, School of Veterinary Medicine and Animal Science, University of São Paulo (FMVZ/USP), Brazil.
[2] Surgery Department, School of Veterinary Medicine and Animal Science, University of São Paulo (FMVZ/USP), Brazil.
[3] Laboratory of Veterinary Anatomy - 3D Educational Technologies. Federal University of Acre (UFAC), Rio Branco, Acre, Brazil.

especially in descriptive anatomy where morphology is compared between different species of domestic and wild animals. Literature affirms that the emergence of 3D printing can provide numerous opportunities to enhance aspects of medical and healthcare training. These plastic models can be used in anatomy education as a teaching tool as well as a method for increasing the curriculum and innovating established learning modalities, integrating anatomy to radiology (Smith *et al*., 2017).

The creation of new rapid prototyping techniques, the low cost of 3D printers as well as the emergence of new software for image analysis have allowed the creation of 3D models of bones. Truly, its application is possible in the field of anatomy teaching in many universities of animal sciences. The deterioration of bone pieces by continuous use, the difficulty in obtaining a good number of anatomical pieces for all students as well as the coming up of the 3D printers at an affordable price for the manufacturing of 3D bones models at full scale have led to the introduction of a new way of studying anatomy. The advent of 3D printers has allowed the creation of biomodels by rapid prototyping, providing production of three-dimensional structures starting from a computer graphic modeling and, then, an additive manufacturing process (Lozano *et al*., 2017).

Technological resources can contribute to veterinary anatomy teaching, making subjects related to this area which are essential for the training of Veterinary Medicine students, increasingly updated in the face of new technologies and new generations of students. Specific in osteology, today there are biomodels of canine and equine skeletons reporting the use of 3D printing to produce them (dos Reis *et al*., 2017).

This article aims to describe the use of multiplanar reconstruction, volumetric computed tomography reconstruction, rapid prototyping and 3D printing of opossum head to create a biomodel to veterinary education in descriptive anatomy of wild animals. The academics need to know gross anatomy of wild animals but unfortunately, they often do not have an available laboratory with a satisfactory collection of anatomical pieces. Currently, the main macroscopic study tool is by comparative anatomy investigating the shape and development of the animal body through the analogy between different species.

MATERIAL AND METHOD

This study was developed in the University of São Paulo (USP), Brazil, comprising six steps as shown by the flowchart below (Fig. 1).

The first step was borrowing the opossum head from the collection of the University's Veterinary Anatomy Museum (MAV/USP), which made possible to study the bones in situ to know this topographic region. The second stage comprised a computerized tomography examination by the Diagnostic Imaging Service- Surgery Department – School of Veterinary Medicine and Animal Science; the computed tomographic images were acquired with a Philips

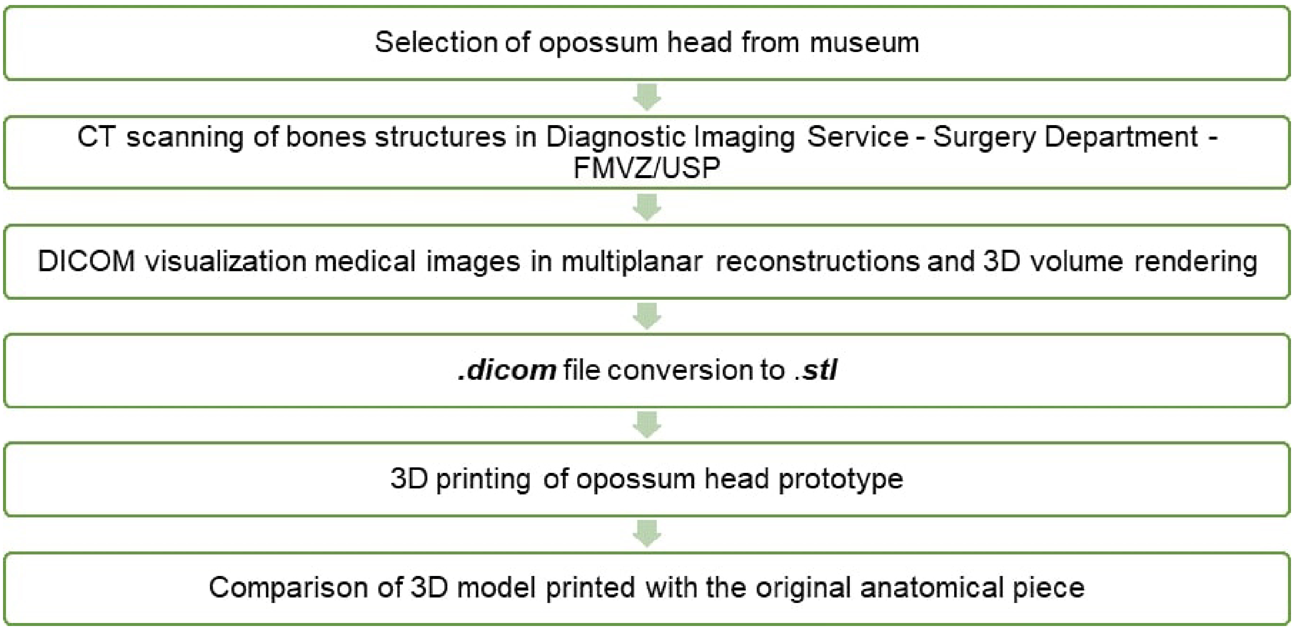

Fig. 1. Flowchart for design of 3D anatomical model. Sequence of six steps to creation of 3D prototyping of opossum head.

CT Scanner MX 8000 IDT® 16 data channels in DICOM (Digital Imaging and Communications in Medicine) file format. It was produced in 271 slices thickness of 0,7 mm. The third step involved the files analysis in Osirix® and Radiant® DICOM viewers, which allowed to compare different sequences: the multiplanar reconstructions in orthogonal planes (transverse, sagittal and dorsal planes) and the 3D volume rendering that allows the view of a large volume of data generated by CT scanner in three-dimensional space and interactively explored in different aspects. The fourth step involved the conversion of DICOM file to STL through the software InVesalius® for the creation of the opossum head prototype. The fifth step was to print the opossum head prototype through the 3D printer model Flexprinter® 2025 (available in www.flexbras.com.br), according to the following specifications: layer thickness 0.2 mm, wall thickness 1.2 mm, fill 20 %, material ABS color ivory. These were used to produce a 1:1 scale physical model with the fused deposition modeling (FDM) 3D printer to produce highly accurate opossum head model using the software Cura®. And finally, the sixth stage comprised the comparison of the 3D model with the original anatomical piece of the museum, evaluating pros and cons of the biomodel created.

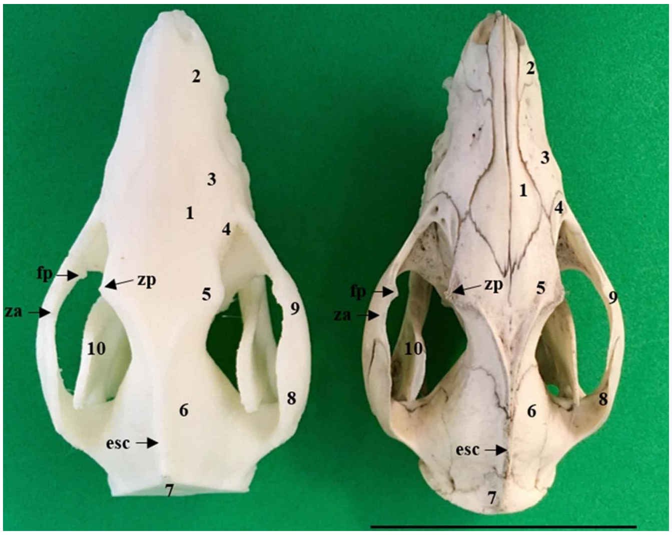

Fig. 2. Dorsal view of natural bones (left) and 3D-printed bones (right) that compose the head of opossum. 1, nasal; 2, incisive; 3, maxilla; 4, lacrimal; 5, frontal; 6, parietal; 7, occipital; 8, temporal; 9, zygomatic; 10, mandibula. Note also: esc, external sagittal crest; za, zygomatic arch; zp, zygomatic process of the frontal bone; fp, frontal process of the zygomatic bone. Bar = 5 cm.

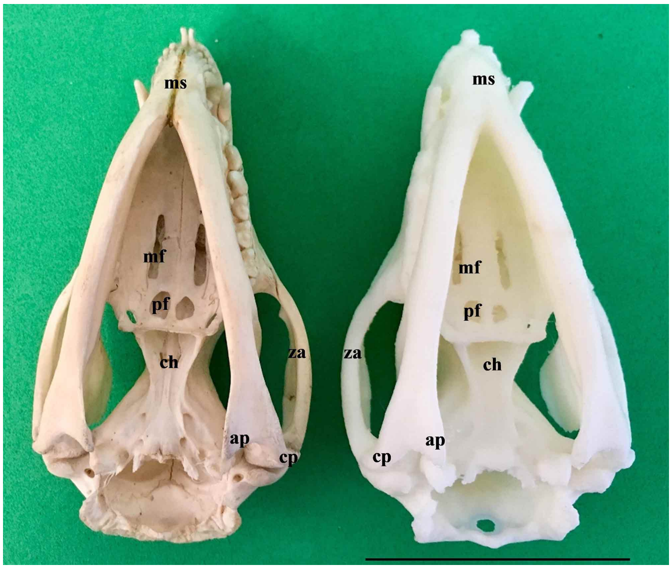

Fig. 3. Ventral view of natural bones (left) and 3D-printed bones (right) that compose the head of opossum. Note also: ms, mandibular symphysis; za, zygomatic arch; mf, maxillopalatine fenestra; pf, palatine fenestra; ch, choana; ap, angular process of mandibula; cp, condylar process of mandibula. Bar = 5 cm.

RESULTS AND DISCUSSION

Particularly the skull is made of many fused bones as the animal becomes skeletally mature. CT is an excellent modality to depict and study the complex anatomy of the skull and the jaws using multi-planar 2D images as well as 3D rendering images. The skull is generally symmetric along sagittal plane which can be used for comparisons of paired structures during interpretation (Wisner & Zwingenberger, 2015).

The opossum skull is relatively small and narrow. It can be divided into two parts: facial or viscerocranium, which comprises a pair of mandibles and bones that circumscribe the initial part of the digestive and respiratory systems, and neurocranium which houses the encephalic organs from central nervous system. Figures 2, 3 and 4 show the gross anatomy of bones that compose the head of an opossum.

This research proves that the parts of maxilla and premaxilla palate are fenestrated by a series of paired apertures. The external sagittal crest, the most prominent structure in the dorsal view of the skull, can also be palpated in a living animal during semiology examination.

The mandible is definitely the largest bone in the head. However, in this work it was not possible to separate the mandible from the skull since it was already found artificially glued because it is a very fragile piece exposed in the Anatomy Veterinary Museum.

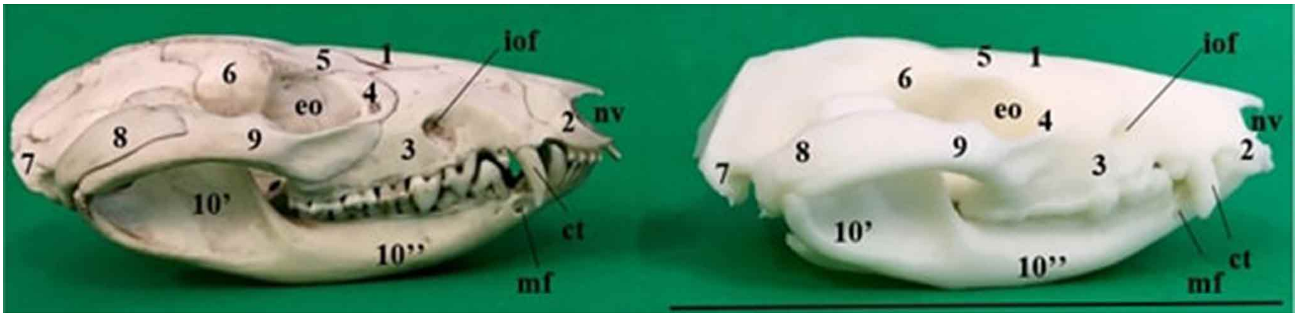

Fig. 4. Left lateral view of natural bones (left) and 3D-printed bones (right) that compose the head of opossum. 1, nasal; 2, incisive; 3, maxilla; 4, lacrimal; 5, frontal; 6, parietal; 7, occipital; 8, temporal; 9, zygomatic; 10', mandibular ramus; 10'', mandibular body. Note also: nv, nasal vestibule; iof, infraorbital foramen; ct, canine tooth; mf, mentalis foramen; eo, eye orbit. Bar = 9.5 cm.

When comparing the two pieces (in natura and artificial), the printed template has estimated weight of 21.47 grams while the original piece has 23.10 g. The area measures are similar presenting 9.5 cm of height and 5.0 of width.

The qualitative aspects of rapid prototyping and 3D printing regarding structures that can be seen and structures that unfortunately cannot be seen when compared to the piece in natura are presented in the Table I.

About dental arch defect observed in biomodel, it is believed that the problem actually occurred during 3D printing process since the Figure 5 proves that the dental roots were correctly scanned by CT. The maximum intensity projection (MIP), projecting the voxel with the highest attenuation value on every view throughout the volume onto a 2D image makes possible to analyze with a close definition not only the tooth but also the gomphosis or the dentoalveolar syndesmosis.

Table I. Analysis of qualitative aspects of the prototyped piece compared to the natural one. Images from authors.

Fig. 5. Maximum intensity projection (MIP) is a CT method analysis for opossum dentistry evaluation.

This kind of defect is not rare when using a 3D printer; biomodel presents this dentistry identification problem when compared to in natura piece or DICOM file images from CT. In the original one, it is possible to delimit clearly the dental roots of some teeth, mainly of the premolars, whereas in the biomodels the region of dental arch is not so well identifiable. One of the possibilities for correction of this defect can be by applying Meshmixer®, a free software to clean the 3D scan before printing.

Opossums have the following dental formula: I 5/4, C 1/1, PM 3/3, M 4/4, totalizing 50 teeth, according to Silva *et al*. (2006). They have more incisors teeth in inferior dental arch (mandibula) than in superior dental arch. Marsupials have absence of deciduous dentition; its only deciduous tooth is the third premolar. In this study it was possible to observe that the superior canine teeth are extremely well developed for hunting.

The opossum skeleton basically follows the pattern of all other mammals. However, the literature points at distinguishing characteristics related to the locomotor system like the presence of epipubic bones (Silva *et al*.) and the absence of a true patella. After all, there was no significant difference between bones found in head of the white-eared opossum when compared to those described for dogs.

In this study, CT proved to be an important teaching-learning resource as it becomes more advanced for animal patients and available for students of Veterinary Medicine. It can also dissect virtually some extremely fragile or hard-to-reach regions in anatomical pieces and to improve teaching on complex spatial interactions. The multiplanar and 3D postprocessing techniques demonstrated the feasibility of generating virtual models, being a teaching-learning tool that offers an opportunity to study anatomical pieces that are often rare in veterinary anatomy laboratories.

The use of CT images with their different forms of reconstruction can provide students with a more comprehensive 3D view of opossum craniofacial region, allow greater understanding of head anatomy and awareness for the preservation of this important species. And the 3D model of skull obtained using the addition manufacturing technique in 3D printing with FDM technology reproduces the anatomical details that the students of anatomy must know, with great accuracy and precision. The rapid advancement in the design of increasingly fast and accurate 3D printers as well as the appropriated software that facilitates their use suggests the growing implementation of this technology in the field of veterinary education.

In relation to the aspects of the biomodel creation itself, the value of US$ 17.00 was spent for the production of one anatomical piece. The time of creation was of 1h 30 min for conversion of the CT image in stereolithography file format and the time for the actual printing was of 3h. The quality of slight loss in the printed model was due to the inexperience of the authors about handling the 3D printed biomodel, being recommended the standardization of this new 3D printer for future works with bones scanned by TC.

The results present so many advantages of biomodel when comparing to the preparation of a natural anatomical piece through traditional anatomical technique. Firstly, all bones used in animal anatomy studies must be obtained legally and it is unquestionable the difficulties today in obtaining corpses of wild animals due to ethical obstacles and by government agencies regarding the collection of dead wild animals in forests or roads. Secondly, the anatomical osteotechniques involves bones maceration, drying, assembly of skeletons and application of varnishes to preserve them. These steps must occur in a preparation room of anatomy laboratory and involve a correct environmental disposal of biological material such as removing soft tissues. All of this certainly involves higher financial investments with specialized technical manpower, chemical products, surgical instruments and appropriate facilities. In addition to this the storage of the natural parts needs to be in an appropriate place to avoid fungus deterioration. In face of all this, the biomodeling of synthetic bones is less expensive, faster and requires much less care for the storage of the parts made in polymerizable resins.

Another great advantage lies in the provision of a large number of biomodels of different species, often not present in that geographic region, for an infinite number of students in short time production. It also allows to so many universities, including those addressed far away from the habitat of this species, to access the biomodel.

For learning and teaching gross anatomy practically, a variety of specimen is necessary in quantity that meets all the academic. Hagebeuker *et al*. (2018) related to the importance in producing syntactic didactic material for classes. They demonstrated that anatomically correct valuable equine skulls of horses at different ages in different sizes can easily be produced with the help of 3D printing in any requested number.

Access to adequate anatomical specimens can be an important aspect in learning the anatomy of wild animals. Moreover, the 3D printed biomodel can be a viable alternative to original bone specimens when used in anatomy education. This study demonstrated an important example of reproducing bone models to be used in anatomy veterinary education. Schimming *et al*. (2016) related that knowledge of the skull morphology in mammals is very important, because it provides baseline anatomic information and can improve veterinarian medical and surgical clinics knowledge.

Due to the existence of few published works using 3D manufacture in the area of teaching of the anatomy of wild animals, certainly this work has an important role in the disclosure of biomodels as a didactic alternative to veterinary osteology teaching. However, further studies must be continued to validate the method in the teaching-learning relationship of academics in veterinary medicine courses. So, an investigation into the use of 3D-printed anatomical models in undergraduate anatomy education is suggested in future research certainly because the progress in the creation of new biomodels will be the greatest achievement in veterinary education development.

In Brazil, according to Federal Council of Veterinary Medicine (Conselho Federal de Medicina Veterinária, 2018), there are more than 300 undergraduate courses in veterinary medicine today besides other related courses such as animal sciences (zootechnic courses) and biological sciences (Conselho Federal de Medicina Veterinária). So, 3D printing can overcome problems associated with the poor infrastructure of anatomy laboratories.

This work also presented a multidisciplinary aspect showing that it is possible to integrate disciplines such as anatomy, diagnostic imaging and surgery (producing specific biomodels for surgical training). Nowadays, we are experiencing a badly reduction in the workload of the anatomy class in order to increase the workload of disciplines linked to the medical clinic. Under this point of view, this work can provide a new educational perspective by changing the traditional teaching method for the integration of contents.

CONCLUSION

It is concluded that it is possible to mitigate the problems caused by the restriction of the use of animals or their bodies in educational institutions, creating biomodels to veterinary education. This work certainly reduces the academic gap between students from universities with large natural collections and students from beginners' institutions especially in the Brazilian private higher education network.

ACKNOWLEDGEMENTS

The authors thank the University of São Paulo Museum of Veterinary Anatomy (http://mav.fmvz.usp.br) for lending its collection and Flexbras.
(http://impressora3dflexbras.com.br) for the development of Brazilian 3D printing technology.

MASSARI, C. H. A. L.; PINTO, A. C. B. C. F.; DE CARVALHO, Y. K.; SILVA, A. F. & MIGLINO, M. A. Reconstrucción volumétrica por tomografía computarizada, prototipado rápido e impresión 3D de cabeza de zarigüeya (*Didelphis albiventris*). *Int. J. Morphol., 37(3)*:838-844, 2019.

RESUMEN: Las piezas anatómicas naturales de animales salvajes son raras y los profesores buscan alternativas satisfactorias, en cantidad y calidad, para enseñar a sus alumnos. Este artículo tuvo como objetivo describir el uso de la reconstrucción volumétrica por tomografía computarizada, la creación rápida de prototipos y la impresión 3D de la cabeza de zarigüeya para obtener un biomodelo en anatomía descriptiva de animales salvajes para educación veterinaria. Se realizó un estudio en seis pasos para construir el biomodelo: (1) selección de cabeza de zarigüeya del museo; (2) tomografía computarizada de estructuras óseas en hospital veterinario; (3) visualización de las imágenes médicas en DICOM por reconstrucciones multiplanares y renderización de volumen 3D; (4) conversión de archivos .dicom a .stl; (5) impresión 3D de cabeza de zarigüeya mediante prototipado rápido; (6) comparación del modelo 3D impreso con la pieza anatómica original. El uso de imágenes de tomografía computarizada, con sus diferentes formas de reconstrucción, puede proporcionar una vista 3D más completa de la región craneofacial de zarigüeya y permitir una mejor comprensión de la anatomía de la cabeza de esta especie. El biomodelo 3D impreso puede ser una alternativa viable a las muestras óseas originales cuando se utiliza en la educación de la anatomía. Sin embargo, se deben continuar los estudios para validar el método en los cursos de Medicina Veterinaria.

PALABRAS CLAVE: Educación Veterinaria; Anatomía; Biomodelo; Impresión 3D; Zarigüeya.

REFERENCES

Conselho Federal de Medicina Veterinária (CFMV). Website. Brasilia, Conselho Federal de Medicina Veterinária, 2018. Available in: http://portal.cfmv.gov.br

dos Reis, D. A. L.; Gouveia, B. L. R.; Alcântara, B. M.; Saragiotto, B. P.; Baumel, É. E. D.; Ferreira, J. S.; Rosa Júnior, J. C.; Oliveira, F. D.; Santos, P. R. S. & Assis Neto, A. C. Biomodelos ósseos produzidos por intermédio da impressão 3D: uma alternativa metodológica no ensino da anatomia veterinária. *Rev. Grad. U. S. P., 2(3)*:47-53, 2017.

Hagebeuker, K.; Zandt, E.; Wölfel, I.; Weber, S. & Poulsen Nautrup, C. *Different forms of equine scульls produced with a 3D printer.* Hannover, 32nd Congress of the European Association of Veterinary Anatomists, 2018.

Li, F.; Liu, C.; Song, X.; Huan, Y.; Gao, S. & Jiang, Z. Production of accurate skeletal models of domestic animals using three-dimensional scanning and printing technology. *Anat. Sci. Educ., 11(1)*:73-80, 2018.

Lozano, M. T. U.; Haro, F. B.; Diaz, C. M.; Manzoor, S.; Ugidos, G. F. & Mendez, J. A. J. 3D digitization and prototyping of the skull for practical use in the teaching of human anatomy. *J. Med. Syst., 41(5)*:83, 2017.

Massari, C. H. A. L.; Schoenau, L. S. F.; Cereta, A. D. & Miglino, M. A. Tendências do ensino de anatomia animal na graduação de medicina veterinária. *Rev. Grad. U. S. P., 3(2)*:25-32, 2018.

Schimming, B. C.; Reiter, L. F. F.; Sandoval, L. M.; Filadelpho, A. L.; Inamassu, L. R. & Mamprim, M. J. Anatomical and radiographic study of the white-eared opossum (*Didelphis albiventris*) skull. *Pesq. Vet. Bras., 36(11)*:1132-8, 2016.

Silva, J. C. R.; Dias, J. L. C. & Cubas, Z. S. *Tratado de Animais Selvagens*. São Paulo, Roca, 2006.

Smith, C. F.; Tollemache, N.; Covill, D. & Johnston, M. Take away body parts! An investigation into the use of 3D-printed anatomical models in undergraduate anatomy education. *Anat. Sci. Educ., 11(1)*:44-53, 2018.

Wisner, E. R. & Zwingenberger, A. L. *Atlas of Small Animal CT and MRI.* Ames, Wiley Blackwell, 2015.

Corresponding author:
Catia Helena de Almeida Lima Massari
University of São Paulo
School of Veterinary Medicine and Animal Science
Surgery Department
Postgraduate Program in Anatomy of Domestic and Wild Animals
Av. Prof. Orlando Marques de Paiva, 87
Cidade Universitária
São Paulo-SP 05508-010
BRAZIL

Email: catia.massari@usp.br

Received: 12-12-2018
Accepted: 01-02-2019