

Artigo Original de Investigação

# Um método semiautomático para a segmentação e reconstrução 3D de dados de tomografia computadorizada do cérebro: uma ferramenta de radiologia veterinária experimental – Primeiros resultados

A semi-automatic method for segmentation and 3D reconstruction of brain computed tomography data: an experimental veterinary radiology tool – First results

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O cérebro é um órgão fundamental e complexo, cujo estudo pode ser aprimorado pela definição nas imagens dos contornos das diferentes estruturas anatómicas da cabeça, para posterior processamento (segmentação). Inicialmente, os métodos aplicados baseavam-se na técnica de segmentação manual de imagens, continuando estes a evoluir para a automatização. Seguindo esta tendência, desenvolvemos um método semiautomático e aplicado na área da Medicina Veterinária. Dessa forma, o foco foi direcionado para acelerar o processo de segmentação, para diminuir a variabilidade manual interoperador e para auxiliar a análise avançada de dados anatómicos, com o intuito de desenvolver futuros métodos de diagnóstico auxiliados por computador.

Para testar o método, foram realizadas aquisições de tomografia computadorizada do crânio em três cães, cujas imagens foram processadas e analisadas por um programa Java - SAEM, Imagej e 3DSlicer.

Após a aplicação de diferentes técnicas de segmentação, o cérebro canino foi isolado de estruturas como osso e tecidos moles e, finalmente, cortes de tomografia computadorizada (TC) foram usados para produzir uma reconstrução 3D. Para comparar os três métodos foi calculada a média de diferença entre os três métodos, normalizada à média de volume cerebral.

Esta experiência oferece um método semiautomático para segmentação do cérebro usando imagens axiais de TC da cabeça. No entanto, algum ruído indesejado ainda permaneceu após a implementação da nossa abordagem.

*The brain is a fundamental and complex organ, whose study can be improved by defining the contours of the anatomical structures of the head images (segmentation), for further processing. Initially, the basic imaging methods were based on imaging manual segmentation technique, continuing to evolve towards automation. Following this tendency, we have developed a semi-automatic method applied in the field of Veterinary Medicine. Thus, the focus was on accelerating the expansion process, decreasing inter-operator manual variability, and aiding advanced analysis of anatomical data, with the aim of developing future computer-aided diagnostic methods.*

*In order to test the method, brain computed tomography acquisitions were performed in three dogs, whose images were processed and analyzed by a Java program - SAEM, Imagej and 3DSlicer.*

*After applying different application techniques, the canine brain was isolated from structures such as bone and soft tissue, and finally computed tomography (CT) slices were used to produce a 3D reconstruction. To compare the three methods, the mean difference between the three methods was calculated, and normalized to the mean brain volume.*

*This experiment offers a semi-automatic method to transform the brain using axial CT images of the head. However, some unwanted noise still remained after implementing our approach.*

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**PALAVRAS-CHAVE:** Cérebro; segmentação; reconstrução em 3D; programação Java.

**KEY WORDS:** Brain; segmentation; 3D reconstruction; Java programming.

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## INTRODUCTION

Medical image analysis is receiving more and more importance during the last years enhancing the traditional visual analysis. Biomedical digital images contain underlying information that can be used for hypothesis generation, testing or both. In fact, after

quantitative features (voxel/pixel intensity, shape, size or volume and texture) extraction, the data generated can be used for evidence-based clinical decision namely for diagnostic, prognostic and predictive accuracy. The end goal is to gather all the extracted information inside a database and combine it with patient characteristics and finally allowing biomarkers development<sup>1-5</sup>.

In general, magnetic resonance imaging (MRI) is the imaging modality of choice for brain study, thanks to higher tissue contrast and, for that reason, most of research concerning brain segmentation is based on MRI. However, in some cases MRI is contraindicated (claustrophobic patients, patients with metallic or electrical implants), and alternatively a computed tomography (CT) scan is necessary. Some of the CT scan advantages for brain studies include good detection of tumors, hemorrhage and bony detail. Because of that, we consider extremely important further analysis and CT image processing as seen in MRI, where techniques such as Voxel-Based Morphometry have provided valuable additional information<sup>6, 7</sup>.

CT consists in an imaging modality that allows sectional imaging, with almost no overlap of organs or anatomical structures. It uses X-ray radiation in order to obtain 3D images about the object under study. For that, a high-powered X-ray tube is applied around a slice of the patient's anatomy. X-rays penetrate tissues through different ways depending on photons energy and the type of tissue. For example, bone tissue (high density) appears white and air appears black (low density).

After X-rays emission and interaction with patient's anatomy, they reach the detectors (transforming radiation in an electrical signal). The resulted signal is proportional to the penetrated tissue density, which then generates a matrix image<sup>8-10</sup>.

For image processing and analysis, a public domain Java program, imagej (<https://imagej.nih.gov/ij/>) was applied during our segmentation approach. Some of the related capabilities include display, edit, analyses about 8-bit, 16-bit and 32-bit images. Besides that, the used program is able to read different image formats (TIFF, JPEG, DICOM, etc.), also supporting "stacks", or in other words a series of images that share a single window. The program possesses an open architecture, providing Java plugins or macros, capable to solve image processing and analysis challenges. The first option is flexible and faster than macros, however harder to compose, written in Java programming language and compiled to class files. In the other hand, macros represent the easiest way to perform a series

of commands using a Java-like language, which code is stored in text files (.text and .ijm extensions)<sup>11</sup>.

In Human Medicine, medical image processing using segmentation based on textural feature techniques is the chosen approach, however this kind of analysis is complex and challenging. It can be defined as a tool that considers related elements, which starts with a spatial arrangement of texture element, where texture primitive is a group of pixels. A texture may present different characteristics such like being fine, coarse, smooth or grained, depending on pixels intensities properties and spatial relationship between them<sup>12</sup>.

Thresholding is a fundamental approach for segmentation that enjoys a significant degree of popularity, especially in applications where speed is an important factor. In fact, it plays an important role for image segmentation, thanks to intuitive properties and easy implementation. Supposing that the image under analysis is composed by white regions on dark background, thresholding basically groups into fields the pixels intensities. Based on the pixel intensity values, it is possible to extract a white object (higher pixel intensity) in relation to the dark background of the images (lower pixel intensity), applying a specific threshold of pixel intensity values that allows for separation of the denser object in relation to a less dense background. Threshold can also be applied in color images, through a specific function called Color Threshold<sup>13</sup>.

The literature on humans and brain segmentation using CT images is quite short, and even more limited in the Veterinary Medicine field. In the first case, it is possible to find some recent published studies, which have actually achieved remarkable and promising results. Torres et al. proposed a brain segmentation method which combines edge-and region-based information with spectral techniques through the morphological algorithm of watersheds<sup>14</sup>. Sharma et al. proposed a method for brain segmentation for identifying and extracting hemorrhage from 2D CT scan<sup>15</sup>. Lauric et al. compared three soft segmentations applied to CT image of the brain. These methods have been used on MRI data with good results and are representative of the class of soft segmentation

algorithms<sup>6</sup>.

Regarding brain segmentation using CT as imaging modality and focusing in Veterinary Medicine, in fact we were unable to find any published study. In the other hand, there are some interesting articles involving brain segmentation and corpus callosum, but using MRI<sup>16</sup>. For several centuries that domestic animals have been used for neuroscience investigation and in translational neurology. For example, it is known that dogs suffer from age-related cognitive dysfunction similar to human Alzheimer's disease<sup>17</sup>. CT scan is capable to provide relevant information in shorter time than MRI. Depending on clinical information, in some situations it represents the most appropriate diagnostic imaging technology, reason why we consider fundamental further brain CT image analysis.

## MATERIALS AND METHODS

### Dataset and participants

CT data were obtained from the Veterinary Imaging Center, Portugal (<https://www.imagiologiaveterinaria.com/>). Whole-brain CT scan from three dogs (Age in years: Dog 1=12; Dog 2=10; Dog 3=9; mean: 10,33 and SD: 1,25) were used as a dataset for this pilot test. The acquired exams were all performed in a clinical context, the animals were not exposed on purpose to perform these measurements.

### Anatomical CT data acquisition

The anatomical images were acquired using a CT scan (Emotion Duo Siemens). Before the beginning of the examination, animals were anesthetized and then placed in head-first and ventral position. The acquisition protocol used in this study was: 131 mA; 120 Kv; slice thickness 1.3 mm; slice interval 0.6 mm.

### Quality assessment

To assess the quality of segmentation and volumetry, we performed two manual segmentations and volumetry, using the imagej (<https://imagej.nih.gov/ij/>) and 3DSlicer (<https://www.slicer.org/>) software. These softwares provide a series of standard solutions that allow

manual segmentation to be performed, however this procedure is time consuming. For control and comparison purposes, a CT acquisition of a 1.5-liter phantom was performed comparing volumetry methods.

### Manual segmentation – imagej

Imaging data was initially loaded using the option import image sequence. After applying a threshold, a region of interest was drawn around the brain tissue and the voxel counter tool was used to measure the number of voxels and the thresholded volume corresponding to the brain tissue.

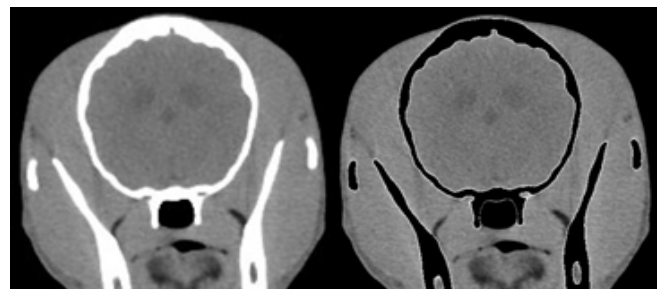
### Manual segmentation – 3DSlicer

The segmentation of the data using the 3D Slicer was performed using the tools of the editor module, namely the level tracing effect option, applied to each tomographic cut to identify and segment the brain tissue. The volume calculation was performed using the quantification module, option label statistics.

### The semi-automatic experimental method – SAEM

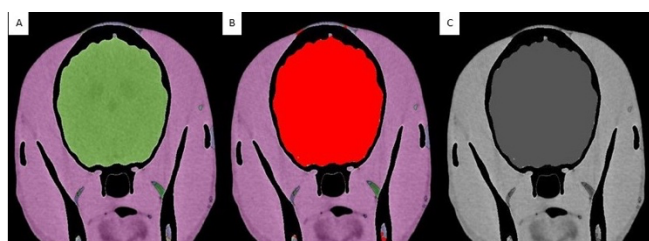
Initially a Java software was executed, after what canine brain CT images (Fig. 1 left image) were imported and opened. The following step required was bone extraction, where a simple Macro developed by our team was applied (Fig. 1 right image). Basically, this Macro defines the lower and upper threshold level, creates a selection involving the threshold area and removes information inside that selection. In fact, for bone segmentation using CT data, threshold application is sufficient, thanks to the high contrast existent between bone and surrounding tissues. At last, segmented images were saved in a new folder.

**Figure 1 – Bone extraction.** Canine brain CT slice showing before (left image) and after skull extraction (right image).



In order to proceed with brain segmentation, a manual step is required where CT images generated without bone tissue are subsequently imported and submitted to a morphological segmentation plugin (Fig. 2A). It contains a graphical user interface to segment 2D/3D images based on morphological operations such as minima and morphological gradient, with watershed flooding algorithms to segment grayscale image of any type (8, 16 and 32-bit) in 2D and 3D. The first step required is an input image, after what two options can be seen: border image and object image. Second option was selected (if borders of the object do not have high intensity values than the rest of voxels in the image). All the remaining options are kept as default and Run option is executed. In the end of the process a new pile of images was created and saved.

**Figure 2 – Brain segmentation.** Morphological segmentation plugin application (A), Color Threshold (B) and 8-bit image conversion (C), respectively in all images contained inside the stack under analysis.



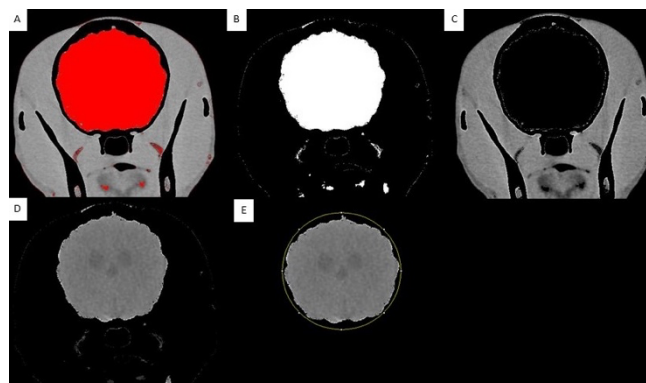
All the subsequent steps were performed by an automatic method (Macro application), which will be discussed. Images analyzed through morphological segmentation plugin were exposed to the Color Threshold function allowing brain tissue selection (Fig. 2B). The stack generated was saved and imported, in order to convert them in 8-bit images (converts to 8-bit grayscale by linearly scaling from min-max to 0-255, where min and max are these two values as display range), which are ultimately also saved (Fig. 2C). Then, Threshold function was applied allowing once more brain tissue selection (Fig. 3A). Consequently, a binary mask (a new 8-bit image whose pixels have a value of 255 outside the selection and 0 inside) is created for each image inside the CT stack (Fig. 3B) and those masks are then used to subtract the initial bone

segmented images (Fig. 3C). Finally, Remove Outliers (replaces a pixel by the median of the pixels in the surrounding area) function is applied in the final stack (Fig. 3D), erasing some undesired pixels surrounding brain tissue (Fig. 3E and Fig. 4).

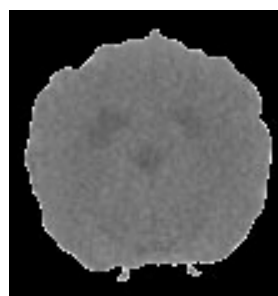
### Volume measurement and statistics

The plugin voxel count was used and adapted to measure a series of parameters, namely: the thresholded voxels in a stack, the average voxels count per slice, total ROI voxels, voxels in stacks, voxel size, thresholded volume, average volume per slice, total ROI volume and volume of stack. In this case, thresholded volume was used to extract the brain volume after the segmentation process.

**Figure 3 – Brain extraction.** Application of Threshold (A), binary mask creation (B), image calculator (C), image subtracted (D) and elliptical selection use as well as clear outside option (E).



**Figure 4 – Brain slice.** Canine brain CT slice (ventricles level) illustrating only cerebral tissue.





## RESULTS

The volume obtained with imagej for brain tissue were Brain1 = 41235.13 mm<sup>3</sup>, brain2 = 43398.95 mm<sup>3</sup> and Brain3 = 50992.60 mm<sup>3</sup>; with 3DSlicer the values were Brain1 = 47418.53 mm<sup>3</sup>, Brain2 = 49906.82 mm<sup>3</sup> and Brain3 = 58639.17mm<sup>3</sup>; with SAEM the values were Brain1 = 52062.70 mm<sup>3</sup>, brain2 = 54794.70 mm<sup>3</sup> and Brain3 = 64382.30 mm<sup>3</sup>, respectively (Fig. 1).

The mean of differences between the three methods was calculated and normalized to the mean of the three results for each brain:  $\text{abs}(\text{Vol\_ImageJ-Vol\_3DSlicer}/(\text{mean of the 3 methods}))$  with a consistent difference of 0.13 for all brains;  $\text{abs}(\text{Vol\_ImageJ-Vol\_SAEM})/(\text{mean of the 3 methods})$  with a consistent difference of 0.23 for all brains, and  $\text{abs}(\text{Vol\_3DSlicer-Vol SAEM})/(\text{mean of the 3 methods})$ , with a consistent difference of 0.01 for all brains.

Computed tomography images of a phantom with a previously known volume, equal to 1500000 mm<sup>3</sup> or 1.5 L, were acquired to serve as a gold standard for the

calculation of volumes. The value for the phantom volumetry using different methods was imagej = 1584048.16 mm<sup>3</sup>, 3DSlicer = 1517600.00 mm<sup>3</sup> and SAEM = 1589320.07 mm<sup>3</sup>.

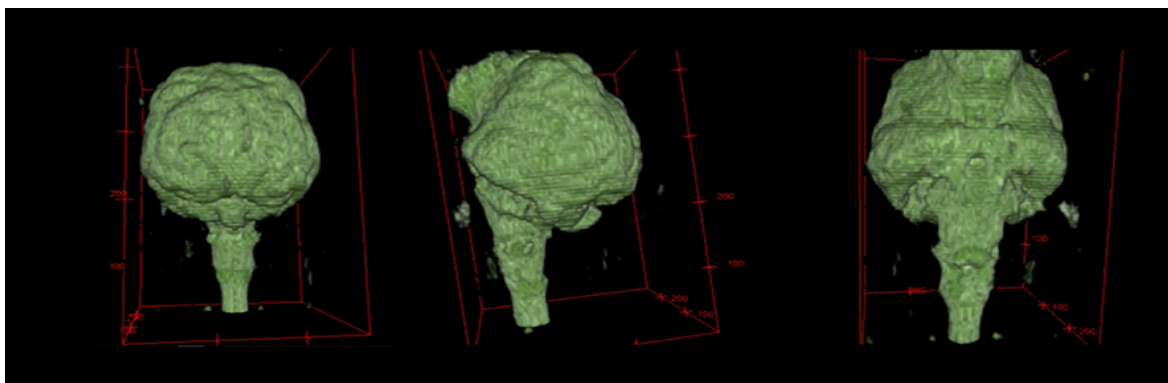
The mean of the differences between the three methods was calculated and normalized, in this case, to the already known phantom value (1500000 mm<sup>3</sup>):  $\text{abs}(\text{Vol\_ImageJ-Vol\_3DSlicer})/(1500000 \text{ mm}^3)$  with a value of 0.04;  $\text{abs}(\text{Vol\_ImageJ-Vol\_SAEM})/(1500000 \text{ mm}^3)$  with a value of 0.00, and  $\text{abs}(\text{Vol\_3DSlicer-Vol SAEM})/(1500000 \text{ mm}^3)$  with a difference of 0.05.

No advanced statistical analyses were performed due to dataset size limitations, though future work may address this. One important plug-in used during our analysis was 3D Viewer, allowing a powerful and fast 3D visualization method, where stacks are displayed as texture-based volume renderings, surfaces or orthoslices (Fig. 5), and the whole-Brain Volume was calculated using the voxels per slices images (Fig.6).

**Table 1** – Brain volumetry.

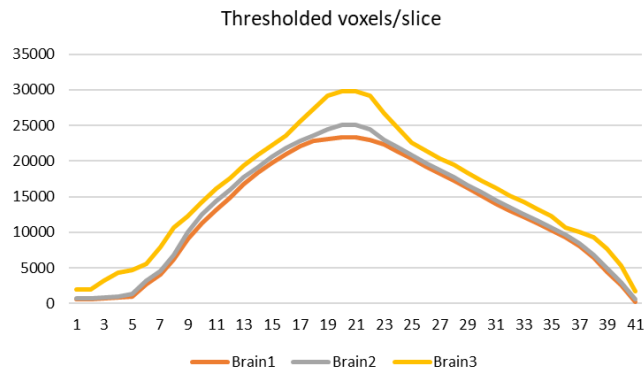
	Age (years)	Volume (mm3): imagej	Volume (mm3): 3Dslicer	Thresholded voxels: SAEM
Brain 1	12	41235.13	47418.53	52062.7
Brain 2	10	43398.95	49906.82	54794.7
Brain 3	9	50992.60	58639.17	64382.3

**Figure 5** – Whole-Brain 3D reconstruction. Canine brain CT 3D reconstruction showing the segmented cerebral tissue.



**Figure 6** – Whole-Brain Volume analysis. Distribution of voxels per CT slices of the three segmented canine brains used for

morphometry analysis.



## DISCUSSION

The general objective of segmentation is to define the contours of the different structures in the images, for further processing. Segmentation represents an important step for image analysis, working like a method that divides an image in several smaller regions, according to some characteristics, such as color, texture, gray level and contrast. In this case, the process finishes when the object of interest is isolated, however this technique is one of the hardest ways for image processing. Segmentation precision is crucial for analysis success or failure of diagnostic methods. Even in nuclear medicine field, particularly in positron emission tomography (PET), segmentation is extremely relevant for quantification purposes, disease diagnosis and staging, which is currently highly used, for example in lung analysis. Focusing in brain and using CT scan, some inherent challenges involve partial volume effects that produce low brain tissue contrast and also fuse objects in the same intensity range. The ideal segmentation techniques depend on image acquisition modality, as well as on the tissue of interest<sup>18-20</sup>.

Image segmentation algorithms generally are based on one of two basic properties of intensity values: discontinuity and similarity. In the first category, the approach is to partition an image based abrupt changes in intensity, such as edges in an image. The principal approaches in the second category are based on partitioning an image into regions that are similar according to a set of predefined criteria<sup>21</sup>.

This experiment offers a semi-automated method for brain segmentation using axial head CT images. Our results also support that brain segmentation can be achieved by using CT data and will certainly increase the understanding about this area, currently with limited research.

When the methods were tested to perform segmentation and volumetry based on the voxel of the brain imaging data of three dogs, we found that the mean of differences between the three methods, normalized to the mean of the brain volume result, was smaller for  $\text{abs}(\text{Vol\_3DSlicer} - \text{Vol SAEM}) / (\text{mean of the 3 methods})$ . The 3DSlicer has more user-friendly segmentation tools, which can explain its performance when compared to the other methods.

These results encourage us to continue this project to fully automate this method. Bearing in mind that the life expectancy of dogs is increasingly long and that, for this reason, the occurrence of cognitive alterations in dogs is already evident, namely canine cognitive dysfunction (CCD)<sup>22</sup>, consequently it is essential to have analysis tools that allow the detection of morphometric changes at the brain level of these animals for clinical and research application.

Cerebral morphometry applied to veterinary medicine will permit comparative studies<sup>23</sup> to be carried out. The brain of dogs can thus serve as a natural model of the disease for the study of neurodegenerative diseases (Alzheimer's disease in humans and CCD in dogs). These studies are essential for the development of imaging biomarkers. In this case, brain morphometry research can benefit from the use of imaging databases.

## CONCLUSIONS

These developments aim to help veterinarians and researchers to perform additional studies, likewise brain volumetric analysis for future comparisons. In fact, by assessing for example the intracranial volume, it is possible to calculate brain atrophy rate. Modern imaging techniques, such as CT have provided the opportunity for non-invasive volumetric quantification of intracranial structures. However, further work focusing the complete automation process is

required, in order to speed up our segmentation method. Another necessary improvement involves full brain segmentation, by removing all undesired pixels nearby the brain model under study. In future works, we intend to study cerebral structures and isolate them from brain tissue, by adopting a similar strategy described in this paper. In order to improve this project, it will be necessary to work on correctly organized and described data so that it can become information useful for knowledge creation.

Figueiredo and Pereira defined knowledge as information combined with experience, context, interpretation and reflexion<sup>24</sup>. In order to produce knowledge, there are three stages preceding that goal: data gathering, its organization into structured information, and then its transformation into knowledge. Data collection and its structure are possible by the use of databases. A database is a collection of stored data and it can simply be a computer spreadsheet as to a dedicated tool designed with all the features necessary to collect, describe and search data. A database's complexity is determined by information requirements and the capacity to invest in a technological solution. Data and information's exponential growth combined with the need of obtaining and using it promptly and anywhere makes online databases an indispensable tool for scientific research. Future works in this field will benefit from the existence of well-structured databases.

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