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Comparison of the Internal Structures of Bones by Microtomography

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Abstract: The use of computed microtomography (μCT) has revolutionized many areas of research, such as noninvasive, fast and high precision techniques, which allows immediate visualization of internal structures without any risk to the object of study. μCT is widely accepted for medical diagnostics, is also important for purposes of zoological research and paleontological. In this work, we used the μCT to investigate the internal structure of bones from mammalian and poultry. We studied the bones of rats and quail. Through microtomography images, we observed that the bones of the poultry have a bony structure in the form of a trellis that is not present in bones of mammals. These trellises bony is an evolutionary adaptation that allowed the bones of the birds to become longer and lighter maintaining its strength. It was also observed that the percentage of the trabecular area in poultry is almost half of that observed in mammals. The results obtained validate the use of μCT as a technique that allows the study of bone structures in small samples, enabling to explore the morphological differences between the bones of those species.

Key words: Microtomography, bones, X-ray.

1. Introduction

The computed microtomography (μCT) is a noninvasive method, which allows the study of the complex internal structures of an object. This technique has been used in various areas of knowledge. However, the biomedical applications are the most common [1]. The μCT is being used in many medical studies as osteoporosis [2], osteoarthritis [3], pseudoarthrosis [4], fragility and fracture risk [5] and the differences between healthy bones and unhealthy bones [6]. These studies use μCT to investigate the internal structures of osseous tissue.

In microtomography samples are explored using some kind of radiation to produce detailed images of axial body. μCT can be analyzed as a two-step process: scanning of samples and image reconstruction. During scanning, the bodies of the samples under study are radiographed at different angles generating a set of radiographs. These radiographs are digitally processed in tomographic image reconstruction process to generate a three-dimensional image of the interior of the object under study from a series of two-dimensional images.

In this work, we use the μCT to compare the internal structure of mammalian bones with the poultry bones, using rat bones (Rattus Norvegicu) and quail bones (Coturnix Coturnix).

The main motivation of this work is to explore the internal morphological structures of pneumatic bones of birds and compare them with mammals. The pneumatization of bird bones is linked to its ability to fly, to provide strength and lightness of his bone structure.

2. Experimental Setup

The authors analyzed four samples, two bones of rats and two bones of poultries. Fig. 1 is a 3D rendering of the samples used.

To explore the samples of rat bones we use the scanner Skyscan Model 1174, Fig. 2, operated at 50 kV, with aluminum filter of 0.75 mm, and pixel size

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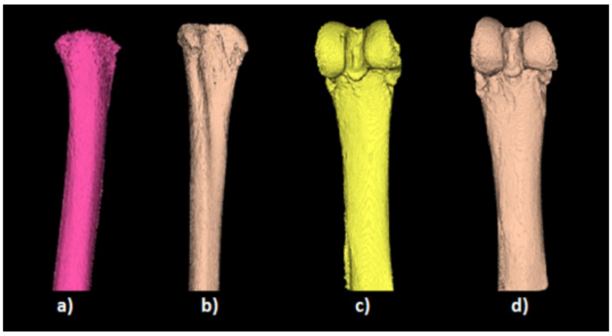

Fig. 1 3D Rendering of samples: quail bones a and b, rat bones c and d.

Fig. 2 Scanner Skyscan Model 1174.

Table 1 Dimensions of the samples.

Sample	Number of slices	Pixel size (μm)	Size in mm
Poultry 1	2,020	22.08	44.61
Poultry 2	2,033	22.08	44.90
Rat 1	1,323	33.29	44.04
Rat 2	1,149	33.29	38.25

of 33.29 μm. A scanner Skyscan model 1173, operated at 50 kV, with aluminum filter of 1.00 mm and pixel size of 22.08 μm, produced the radiographs of poultry bones.

Table 1 summarizes the dimensions of the samples and the number of horizontal slices reconstructed from radiographs obtained in the exploration of samples.

3. Experimental Results

Fig. 3 shows the poultry bones and their slices. It is possible to observe the bone of the bird at its extremity, which is formed by a trabecular bony structure, followed by a trellis section followed by a cortical section. Fig. 4 shows the rat bones and its slices, the authors can see that the bone also begins with a trabecular bone structure, but followed by cortical section of bone, not presenting in its interior a trellis structure.

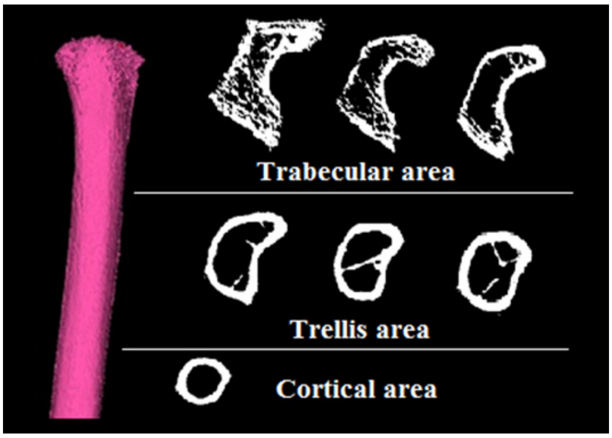

Fig. 3 Poultry bone slices examples.

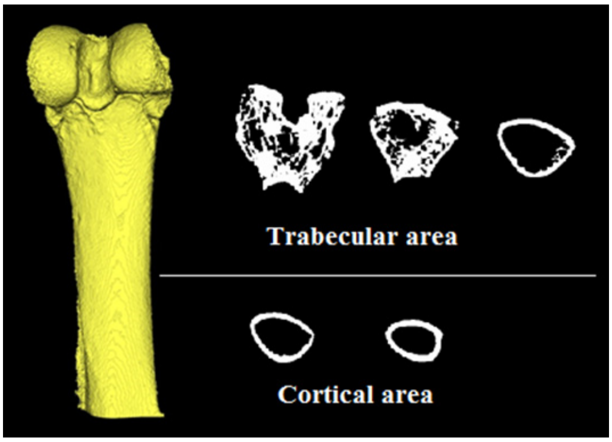

Fig. 4 Examples of rat bone slices.

As seen in Table 2, there is a significant decrease in the area of trabecular bone of the poultry, this area is replaced by bone trellises. Those trellises are displayed in 3D, from different angles for better viewing in Fig. 5.

For a better view of the trellis region was chosen and separate a section of quail 1, composed of 40 slices, displayed in Fig. 6. The selected area corresponds to a section of ~ 0.883 mm or ~ 2% of the sample area and ~ 6.41% of the trellis region in the sample.

Table 2 Percentage of different types of internal bone structures.

Sample	Trabecular area %	Trellis area %	Cortical area %
Poultry 1	27.48	31.63	40.89
Poultry 2	32.86	24.5	42.65
Rat 1	51.17	0	48.83
Rat 2	56.22	0	43.86

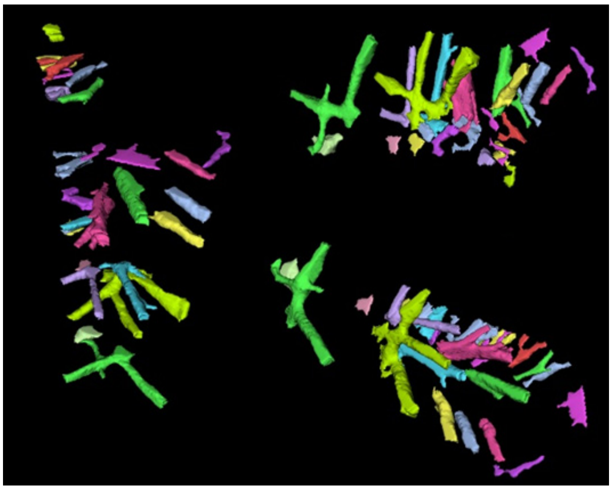

Fig. 5 Rendering 3D bone trellis at different angles.

Based on this set of slices the 3D volume was rebuilt and is represented in Fig. 7. In this image can be observed clearly the structural nature of the bone trellis.

Comparing the trellis region with the trabecular region of the quail bone, a huge difference between the two regions is observed. The trabecular region of the bone, by its definition, has a spongy structure without defining pattern filling the entire inside of the bone. The trellis region has a large empty area with several narrow and straight structures in the form of pillars which support between themselves and inner walls of the bone in a triangular arrangement. This kind of structure reduces the amount of bone volume, making the bone bird lighter.

In Table 3 the values of five morphometric parameters used in bone assessment studies are presented: TV (total volume), BV (bone volume), BV/TV (bone volume to tissue volume), BS (bone surface) and BS/BV (bone surface to bone volume). These values were calculated by CTAn® software [7] that composes the software package of Skyscan scanner used at work.

The TV is the total volume 3D VOI (volume-of-interest) which is measured based on the marching cubes of the model. The BV is the total volume of binarised objects within the VOI. The BV/TV is the proportion of the VOI occupied by binarised solid objects. The BS is the surface area of all the solid objects within the VOI and the BS/BV is the surface to volume ratio or “specific surface” is a useful basic parameter for characterizing the thickness

Fig. 6 Set of slices of trellis area of Poultry 1.

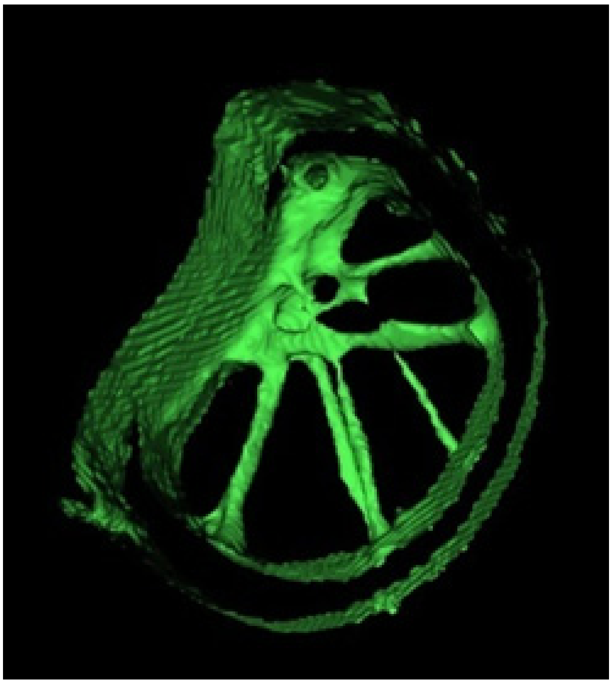

Fig. 7 3D volume based on a set of slices of trellis area.

Table 3 Morphometric parameters of the samples.

	Poultry 1	Poultry 2	Rat 1	Rat 2
TV (μm^3)	4.01E+12	4.11E+12	4.74E+12	3.56E+12
BV (μm^3)	1.68E+11	1.45E+11	2.36E+11	2.77E+11
BV/TV	4.1850	3.5274	4.9816	7.7837
BS (μm^2)	1.84E+09	2.09E+09	1.97E+09	2.07E+09
BS/BV (μm^{-1})	0.01099	0.0144	0.00833	0.007443

and complexity of structures. In bone biology, this measure is used quite often, since it gives a measure on how many bone lining cells cover a given volume of bone. The authors can see from Table 3 that the BS/BV values are significantly higher in the poultries samples. These values indicate a trabecular structure with less complexity and with greater separation.

4. Conclusions

These results show that by using μCT images you can view and separate the existing bone trellises inside the long bones of poultries.

With the segmentation of these structures will be possible, using computational tools to study the load capacity and resistance they provide to the bone. This information may be used to compare with the bones of other birds and so describe the different adaptations in bone structures that contribute to their flight.

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