

Histomorphometric and fractal analysis of femoral, tibial, and metatarsal compact bone samples in sheep (*Ovis aries*), goat (*Capra hircus*), and roe deer (*Capreolus capreolus*)

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Abstract: Histological sections of femur, tibia, and metatarsals originating from sheep (*Ovis aries*), goat (*Capra hircus*), and roe deer (*Capreolus capreolus*) were examined in this study. Our research focused mainly on the quantitative characteristics of the microstructure of these bones. The areas, perimeters, minimum and maximum diameters of the secondary Haversian units, as well as structures of the primary osteonal units were assessed using comparative statistical methods. In addition to these parameters, the present paper investigated the usefulness of fractal analysis (fractal dimension and lacunarity), a new approach that quantifies the complexity and the emptiness pattern of biological structures. Moreover, our work uses pattern recognition methods as well as classical morphometric assessment methods, which appear to generate useful data for bone sample differentiation. Therefore, combining these methods should greatly facilitate further research in veterinary morphology, and forensic and legal veterinary medicine.

Key words: Osteon, fractals, sheep, goat, roe deer, histomorphometry, archaeozoology, legal veterinary medicine

1. Introduction

Quantitative and qualitative studies dealing with histomorphometry were considered pioneer work in the field of microscopic anatomy (1–6). Nowadays forensic studies, morphology investigations, and archaeozoological investigations deal with this kind of data regularly (7–18).

Fractal dimension measures the boundary irregularity of a structure by establishing a numeric value to its degree of complexity (19,20). Low values of fractal dimension indicate low complexity and high values show high complexity. Lacunarity is a measurement of the empty space distribution within a structure. Low values of lacunarity suggest homogeneity within the structure gap distribution and high values show a heterogeneous gap pattern (20). By corroborating the fractal dimension and lacunarity results with the morphometric ones, a more clear image of possible differences and similarities can be observed on a given structure. As far as we know, the attempt to describe bone patterns on manually ground bone slides is limited to a single study performed on humerus and metacarpal bone samples (21).

In the present study, our approach is represented by a morphometric comparative investigation of hind limb compact bone samples (femur, tibia, and metatarsal from

sheep, goat, and roe deer) and scale independent analyses (fractal dimension and lacunarity) that might offer a complementary approach for bone type differentiation. The discriminatory potential of two fractal parameters is also investigated, in order to establish if this scale-independent analysis can offer a complementary approach for bone type differentiation.

2. Materials and methods

Our biological study material was represented by bone samples originating from 6 sheep (*Ovis aries*), 4 goat (*Capra hircus*), and 5 roe deer individuals (*Capreolus capreolus*). All individuals were adult, with ages ranging from 2 to 8 years, and in good health condition. The exact number of bones used is shown in Table 1. The sheep bone samples were part of a collection owned by the Comparative Anatomy Department (Faculty of Veterinary Medicine Cluj-Napoca, Romania). The goat bone samples were collected over a period of 4 years (2010–2014) from local owners (Cluj county) that slaughtered mature individuals of a common unimproved breed for their own consumption. The roe deer bone samples were provided by the regional hunters association (Cluj county) after the annual population control measures in 2009, 2013, and 2014.

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Table 1. The number of bones sampled for the 3 studied species.

	<i>Capreolus capreolus</i>	<i>Capra hircus</i>	<i>Ovis aries</i>
Femur	4	2	4
Tibia	3	3	6
Metatarsus	5	5	4

The histological samples were obtained according to the method described by Maat using manual grinding procedures up to transparency (22). The samples of compact substance taken from the mid diaphyseal part of the bones were cut using a hand saw perpendicularly on the bone axis. The pieces were hand ground using grinding papers with increasing grits (1500/2000 and 3000). The specimens thus obtained were cleared in alcohol and then glued onto microscopic slides with a regular mounting medium and glass cover slips.

For imaging assessment purposes, normal light microscopy was used to examine the native specimens, without a staining method. The slides were used both for the qualitative and quantitative assessment (Olympus BX14 and Olympus UC30 digital camera) using Stream Basic software (Olympus Stream 2011 Basic). Photo acquisition was performed at the 10× magnification level in a TIFF image format, at a resolution of 2080 px/1544 px. In some cases, we used 20× for the clear separation of primary and secondary osteons.

An initial qualitative assessment of the acquired samples was performed based on the classification system of bone structures of de Ricqules (12,23–26).

For morphometric analysis the following measurements were taken using ImageJ (US National Institutes of Health, Bethesda, MD, USA) software, at the level of primary and secondary osteonal units:

For the secondary osteons:

- osteonal area
- osteonal perimeter
- minimal and maximal diameters

For the primary osteons:

- primary osteonal area
- primary osteonal perimeter
- primary osteonal minimal and maximal diameters.

According to the source literature available, a minimum of 100 primary osteonal units should be assessed in order to obtain comparable results (9).

The same digital images used for morphometric measurements (only a 10× magnification level) were also used to assess the fractal dimension and lacunarity. The 500 px/ 500 px regions of interest (ROIs) were cropped from each of the digital images using PhotoScape 3.62

(2001–2013 MOOII TECH). The cropped ROIs were then opened in ImageJ and using the FracLac plug-in (<http://rsb.info.nih.gov/ij/plugins/fractal/FLHelp/Introduction.htm>) and the box-counting method, the fractal dimension and lacunarity were computed.

The results were statistically assessed both interspecifically and intraspecifically (Figures 1 and 2).

Both morphometric and fractal results were processed using Microsoft Excel for data handling (Microsoft Corporation, Redmond, WA, USA) and GraphPad (GraphPad InStat3.05, San Diego, CA, USA) for descriptive and inferential statistical analysis. If the data passed the normal distribution test, a one-way ANOVA was performed, followed by Bonferroni multiple comparisons test if $P < 0.05$.

3. Results

3.1. Morphological qualitative assessment

As expected, the initial qualitative assessment showed a prevalence of plexiform bone tissue (as in most artiodactyls). The subtypes of lamellar and fibrolamellar (1a, 1c with 1c1, 1c3 subtypes) were clearly identified, as well as the fibrous 1f1, 1f2, and 1f3 subtypes (fibrolamellar complex). Areas with scattered, isolated Haversian systems

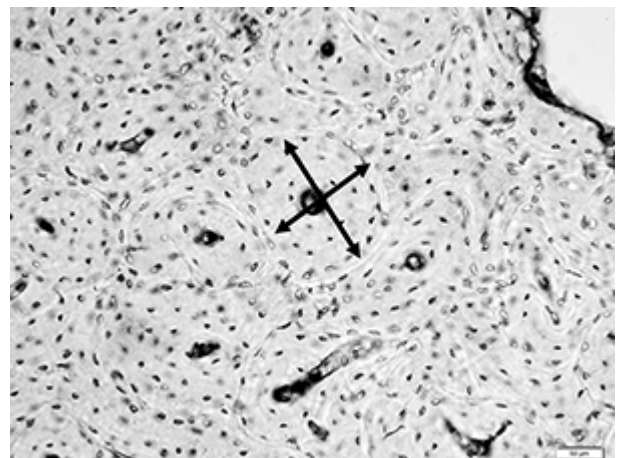


Figure 1. Histological image of the ground sample (*Capra hircus*). Measurements taken: area and maximal and minimal osteonal diameters.

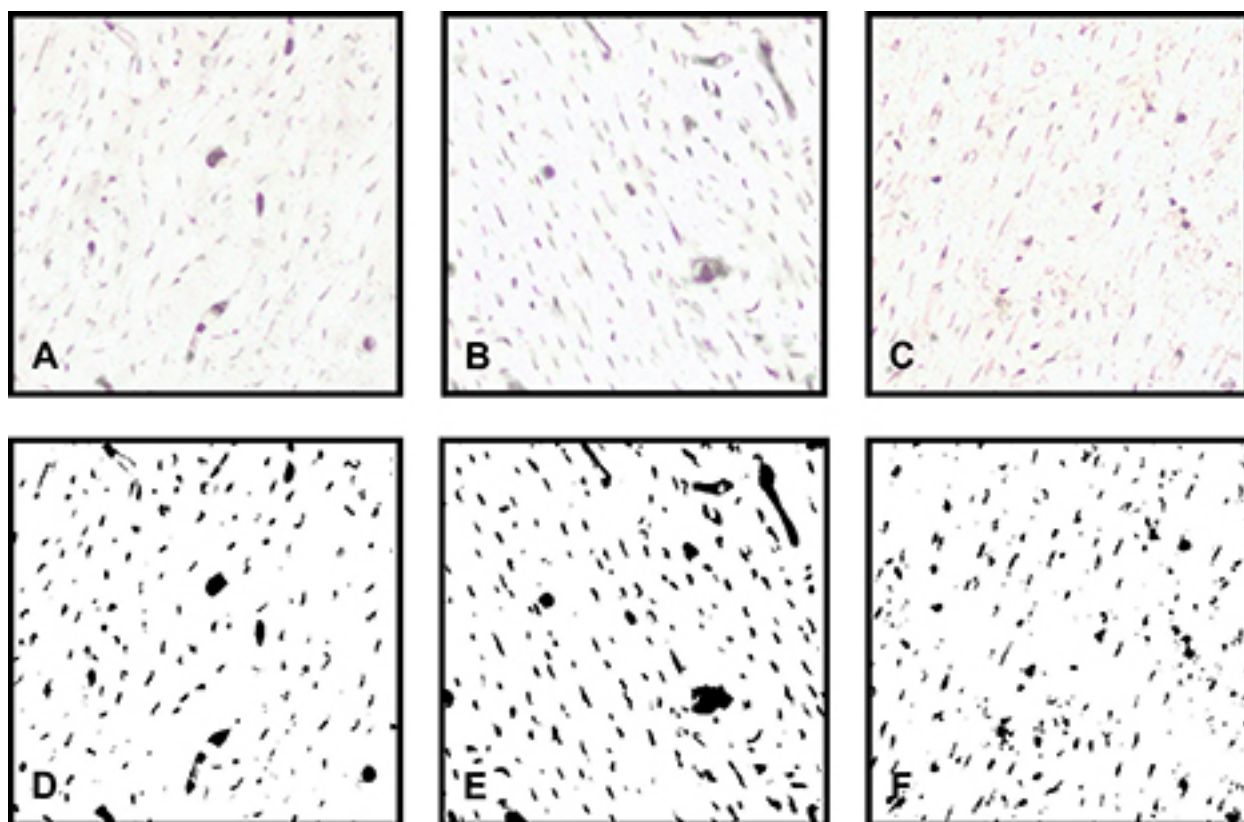


Figure 2. Femur. Sample ROIs and binarization. A, D- *Capreolus capreolus*; B, E- *Capra hircus*; C, F- *Ovis aries*.

(2a1a subtype mainly) and some isolated secondary osteons arranged in rows (osteon banding) were observed. These features were noticed in all three species, with no clear differences among them.

3.2. Quantitative assessment-morphometry

Our quantitative assessment refers to 378 secondary osteons and 514 primary osteonal units. The data from the vascular canalicular elements were not computed nor displayed in this paper due to space limitations.

The data obtained for the area of secondary osteons show a large array of values, with a higher amplitude in sheep (*Ovis aries*), and a much lower amplitude in goat (*Capra hircus*) and roe deer (*Capreolus capreolus*). As far as the perimeter is concerned, the values for the secondary osteons seem to be concentrated in a relatively narrow interval in all species, while those for the primary osteons show a large range of values in terms of species and anatomic segment. The maximal and minimal diameters of the secondary osteons show relatively constant amplitudes in domestic species and a higher one in roe deer (*Capreolus capreolus*), but similar average values (Table 2). The data were analyzed on species and then on bone-related series (femur, tibia, and metatarsus).

The results of the horizontal statistical procedure (a comparison of the same bones in species-series) are listed in Table 3.

Regarding the area of secondary osteonal units, it can be noted that there is only a low degree of differentiation for the *Capra-Ovis* pair, as the rest of the pairs seem to be undifferentiated. The same applies for the perimeter and the minor and major axes of all three bones studied. For the items of the primary osteonal units, the pair *Capra* vs. *Ovis* seems to be the most frequent pair with a high degree of differentiation.

The results of the vertical statistical comparison are listed in Table 4. A constantly present pair with a high degree of differentiation is the pair femur-tibia in sheep (*Ovis aries*), but for the other species this pair is not identified as differentiated. Moreover, in the case of goat (*Capra hircus*) and roe deer (*Capreolus capreolus*) specimens, there are no significant differences among bones within the species as far as secondary osteonal units are concerned.

3.3. Fractal analysis

The results obtained from the fractal dimension (Table 5) show that structure complexity varies significantly among the three species assessed in our study.

Table 2. Descriptive statistics for the osteonal unit morphometry (mean ± standard deviation).

Measured structures	Species	Bone	Area (µm ²)	Perimeter (µm)	Max diameter (µm)	Min diameter (µm)
Secondary osteons	<i>Ovis aries</i> (n = 152)	Femur	12,055 ± 2940	392 ± 53.1	170.0 ± 14	141.2 ± 23
		Tibia	25,098 ± 9970	558 ± 111.4	197.5 ± 41	155.8 ± 30.9
		Metatarsal	17,486 ± 5415	471 ± 78.3	170.8 ± 29.4	127.2 ± 22.73
	<i>Capra hircus</i> (n = 120)	Femur	17,094 ± 624.7	466.06 ± 82	169.9 ± 30.8	124.8 ± 23.5
		Tibia	18,982 ± 8383	474.1 ± 149	165.4 ± 52	133.6 ± 43.1
		Metatarsal	22,089 ± 4091	535 ± 79.7	184.3 ± 24	151.6 ± 11.1
	<i>Capreolus capreolus</i> (n = 106)	Femur	13,868 ± 3922	417.5 ± 57	143.4 ± 23	121.7 ± 15.8
		Tibia	17,274 ± 3965	469 ± 59.3	162.2 ± 26.5	134.5 ± 12.4
		Metatarsal	20,577 ± 7127	510.1 ± 90.79	175.8 ± 36	145.5 ± 23.6
Primary osteons	<i>Ovis aries</i> (n = 241)	Femur	5293.8 ± 3229	256.5 ± 78	94.4 ± 28	65.8 ± 19
		Tibia	15,099 ± 5853	428.1 ± 114	149 ± 40	120.6 ± 32
		Metatarsal	7861.7 ± 3417.7	311.5 ± 67.6	114.6 ± 24	86.7 ± 19
	<i>Capra hircus</i> (n = 150)	Femur	9175 ± 3766	346.4 ± 70	130.5 ± 27	86.4 ± 20
		Tibia	11,668 ± 3697	383.29 ± 58	134.6 ± 22	108.5 ± 17.2
		Metatarsal	12,939 ± 4840	406.26 ± 73	141.9 ± 30	112.5 ± 20.1
	<i>Capreolus capreolus</i> (n = 123)	Femur	8253 ± 2803	320.0 ± 52	110.5 ± 20.7	93.1 ± 14
		Tibia	5579 ± 625	709 ± 473	250.31 ± 168	198 ± 133
		Metatarsal	11,480 ± 4988	378.5 ± 66	131.7 ± 22.9	107.8 ± 21

The highest fractal dimension value was found in roe deer (*Capreolus capreolus*) metatarsus and the lowest in sheep (*Ovis aries*) femur. For lacunarity, the highest value was found in goat (*Capra hircus*) tibia and sheep (*Ovis aries*) femur, and the lowest one in goat (*Capra hircus*) femur and sheep (*Ovis aries*) tibia (Table 5).

The results of the multiple comparison assessment of fractal dimension among the three studied species, for each bone type, show extremely significant differences for most of the pairs analyzed, except for the tibial values in the roe deer (*Capreolus capreolus*) vs. goat (*Capra hircus*) pair.

For lacunarity results, the same statistical assessment shows similar findings, except for the roe deer (*Capreolus capreolus*) vs. sheep (*Ovis aries*) pair (femoral and metatarsal values) and roe deer (*Capreolus capreolus*) vs. goat (*Capra hircus*) pair (tibial values) (Table 6).

When the fractal dimension values for the three studied bones were compared within each species, only the difference between the femoral and tibial bones in *Capreolus capreolus* species was not statistically significant.

Regarding the lacunarity values, no statistical significant differences were found between the femoral and tibial bones in *Capreolus capreolus* species and between the tibial and metatarsal bones in the *Capra hircus* species (Table 7).

4. Discussion

4.1. Morphometric data and their relevance

According to literature data, there are only a few studies using osteonal morphometric data in capriovids (7–10,25,27). In fact, when referring to hind limb bones, data are available only for sheep femur (8), as the authors dealt with a related topic, focusing mainly on the qualitative data and little on the quantitative data while Giua et al. (25) dealt with histometric data mainly.

As far as the femur is concerned, our study revealed values for the secondary osteonal area that were smaller than the ones presented by Martiniakova et al. (28), with the maximal value around 17,000 µm². The average for our samples showed a value of 12,055 µm², which is not close to the one presented by the earlier-mentioned source (with

Table 3. Multiple comparison test results of the morphometric parameters' values, for each bone type, of the three studied species (horizontal comparison).

Bone	Femur	Tibia	Metatarsal
Area- secondary osteon	^{ns} CC vs. CH	^{ns} CC vs. CH	^{ns} CC vs. CH
	^{ns} CC vs. OA	^{ns} CC vs. OA	^{ns} CC vs. OA
	*CH vs. OA	*CH vs. OA	^{ns} CH vs. OA
Perimeter- secondary osteon	^{ns} CC vs. CH	^{ns} CC vs. CH	^{ns} CC vs. CH
	^{ns} CC vs. OA	^{ns} CC vs. OA	^{ns} CC vs. OA
	*CH vs. OA	*CH vs. OA	^{ns} CH vs. OA
Major axis- secondary osteon	^{ns} CC vs. CH	^{ns} CC vs. CH	^{ns} CC vs. CH
	^{ns} CC vs. OA	^{ns} CC vs. OA	^{ns} CC vs. OA
	*CH vs. OA	*CH vs. OA	^{ns} CH vs. OA
Minor axis- secondary osteon	^{ns} CC vs. CH	^{ns} CC vs. CH	^{ns} CC vs. CH
	^{ns} CC vs. OA	^{ns} CC vs. OA	**CH vs. OA
	^{ns} CH vs. OA	*CH vs. OA	**CC vs. OA
Area- primary osteon	^{ns} CC vs. CH	***CC vs. CH	^{ns} CC vs. CH
	***CH vs. OA	***CC vs. OA	***CC vs. OA
	*CC vs. OA	^{ns} CH vs. OA	***CH vs. OA
Perimeter- primary osteon	^{ns} CC vs. CH	***CC vs. CH	^{ns} CC vs. CH
	***CH vs. OA	***CC vs. OA	***CC vs. OA
	CC vs. OA	^{ns} CH vs. OA	*CH vs. OA
Major axis- primary osteon	^{ns} CC vs. OA	***CC vs. CH	^{ns} CC vs. CH
	***CH vs. OA	***CC vs. OA	***CC vs. OA
	CC vs. CH	^{ns} CH vs. OA	*CH vs. OA
Minor axis- primary osteon	^{ns} CC vs. CH	***CC vs. CH	^{ns} CC vs. CH
	***CH vs. OA	***CC vs. OA	***CC vs. OA
	***CC vs. OA	^{ns} CH vs. OA	***CH vs. OA

* = significant, P < 0.05; ** = very significant, P < 0.01; *** = extremely significant, P < 0.001; ^{ns} = not significant, P > 0.05.

an average value of 20,000² μm) but a little closer to data presented by Giua et al. (25) (an average of 17,300 μm²). The values computed for the secondary osteonal perimeter are, in fact, close to the ones presented by our source (390 vs. 400–410/420 μm (28) and 470 μm (25)). The situation seems similar in terms of the maximal diameter (170 μm vs 200 μm for data provided by Martiniakova et al. (8)) but comparable to those of Giua et al. (25), while in terms of the minimal diameter the situation is different, with values almost two times higher found in Martiniakova et al.'s investigation (8), but similar to those of Giua et al. (25). So, to conclude in this case, further data are necessary in order to state that a certain value is characteristic for this species (due to the limited choice of sampling and the relatively low number of items used for calculations).

The values computed for the primary osteonal data in sheep (*Ovis aries*) show values ranging from 5300 μm² to 15,000 μm² (as averages for the 3 bones). Post hoc tests

indicate that there is a significant difference, mainly for the femur vs. tibia and tibia vs. metatarsus pairs, in all the elements that we took into consideration (area, perimeter, and maximal and minimal diameters).

The secondary osteonal values (area) in goat (*Capra hircus*) range from 17,000 μm² to 22,000 μm². The perimeter values range from 256 to 428 μm, while the maximal diameter values range from 94 to 149 μm. Although the values might seem different, post hoc tests show no significant differences among the series.

Primary osteonal values in goat (*Capra hircus*) range from 9100 μm² to 12,900 μm² for the area, 346 to 406 μm for the perimeter, 130 to 140 μm for the maximal diameter, and 86 to 112 μm for the minimal diameter. Even though the values seemed quite similar, post hoc tests showed significant differences for the femur vs. metatarsus pair.

The secondary osteons in roe deer (*Capreolus capreolus*) range from 20,000 μm² to 14,000 μm² (area), 420 to 510 μm

Table 4. Multiple comparison test results of the morphometric parameters' values, for each species, among the three bone types (vertical comparison).

Bone	<i>Ovis aries</i>	<i>Capra hircus</i>	<i>Capreolus capreolus</i>
Area- secondary osteon	*** Fem vs. Tib	^{ns} Fem vs. Tib	^{ns} Fem vs. Tib
	*** Tib vs. Mt	^{ns} Tib vs. Mt	^{ns} Tib vs. Mt
	* Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Perimeter- secondary osteon	*** Fem vs. Tib	^{ns} Fem vs. Tib	^{ns} Fem vs. Tib
	*** Tib vs. Mt	^{ns} Tib vs. Mt	^{ns} Tib vs. Mt
	* Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Major axis- secondary osteon	***Fem vs. Tib	^{ns} Fem vs. Tib	^{ns} Fem vs. Tib
	* Tib vs. Mt	^{ns} Tib vs. Mt	^{ns} Tib vs. Mt
	** Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Minor axis- secondary osteon	***Fem vs. Tib	^{ns} Fem vs. Tib	^{ns} Fem vs. Tib
	***Tib vs. Mt	^{ns} Tib vs. Mt	^{ns} Tib vs. Mt
	* Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Area- primary osteon	***Fem vs. Tib	***Fem vs. Mt	***Fem vs. Tib
	***Tib vs. Mt	*Fem vs. Tib	***Tib vs. Mt
	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Perimeter- primary osteon	***Fem vs. Tib	***Fem vs. Mt	***Fem vs. Tib
	***Tib vs. Mt	*Fem vs. Tib	***Tib vs. Mt
	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Major axis- primary osteon	***Fem vs. Tib	^{ns} Fem vs. Tib	***Fem vs. Tib
	***Tib vs. Mt	^{ns} Tib vs. Mt	***Tib vs. Mt
	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt	^{ns} Fem vs. Mt
Minor axis- primary osteon	***Fem vs. Tib	***Fem vs. Mt	***Fem vs. Tib
	*** Tib vs. Mt	^{ns} Fem vs. Tib	***Tib vs. Mt
	*Fem vs. Mt	^{ns} Tib vs. Mt	^{ns} Fem vs. Mt

* = significant, P < 0.05; ** = very significant, P < 0.01; *** = extremely significant, P < 0.001; ^{ns} = not significant, P > 0.05.

Table 5. Descriptive statistics for fractal dimension and lacunarity results (mean ± standard deviation).

Species	Bone	Fractal dimension	Lacunarity
<i>Capreolus capreolus</i>	Femur (219 ROIs)	1.48 ± 0.04	0.78 ± 0.14
	Tibia (216 ROIs)	1.47 ± 0.05	0.76 ± 0.15
	Metatarsus (165 ROIs)	1.62 ± 0.04	0.67 ± 0.14
<i>Capra hircus</i>	Femur (185 ROIs)	1.59 ± 0.04	0.59 ± 0.12
	Tibia (103 ROIs)	1.45 ± 0.04	0.81 ± 0.12
	Metatarsus (203 ROIs)	1.49 ± 0.03	0.78 ± 0.12
<i>Ovis aries</i>	Femur (106 ROIs)	1.41 ± 0.08	0.81 ± 0.13
	Tibia (74 ROIs)	1.57 ± 0.04	0.59 ± 0.15
	Metatarsus (99 ROIs)	1.46 ± 0.06	0.71 ± 0.13

Table 6. Multiple comparison test results of the fractal dimension and lacunarity values, for each bone type, among the three studied species (horizontal comparison).

Parameter	Bone		
	Femur	Tibia	Metatarsus
Fractal dimension	***CC vs. CH	^{ns} CC vs. CH	***CC vs. CH
	***CC vs. OA	***CC vs. OA	***CC vs. OA
	***CH vs. OA	***CH vs. OA	***CH vs. OA
Lacunarity	***CC vs. CH	^{ns} CC vs. CH	***CC vs. CH
	^{ns} CC vs. OA	***CC vs. OA	^{ns} CC vs. OA
	***CH vs. OA	***CH vs. OA	**CH vs. OA

* = significant, P < 0.05; ** = very significant, P < 0.01; *** = extremely significant, P < 0.001; ^{ns} = not significant, P > 0.05.

Table 7. Multiple comparison test results of the fractal dimension and lacunarity values, for each species, among the three bone types (vertical comparison).

Parameter	Species		
	<i>Capreolus capreolus</i>	<i>Capra hircus</i>	<i>Ovis aries</i>
Fractal dimension	^{ns} Femur vs. Tibia	***Femur vs. Tibia	***Femur vs. Tibia
	***Femur vs. Metatarsus	***Femur vs. Metatarsus	**Femur vs. Metatarsus
	***Tibia vs. Metatarsus	***Tibia vs. Metatarsus	***Tibia vs. Metatarsus
Lacunarity	^{ns} Femur vs. Tibia	***Femur vs. Tibia	***Femur vs. Tibia
	***Femur vs. Metatarsus	***Femur vs. Metatarsus	**Femur vs. Metatarsus
	***Tibia vs. Metatarsus	^{ns} Tibia vs. Metatarsus	***Tibia vs. Metatarsus

* = significant, P < 0.05; ** = very significant, P < 0.01; *** = extremely significant, P < 0.001; ^{ns} = not significant, P > 0.05.

in perimeter, 143 to 175 µm for the maximal diameter, and 120 to 145 µm for the minimal diameter.

As far as the area is concerned, primary osteons range between 5500 and 11,480 µm². Perimeters range from 709 to 320 µm. The maximal diameters range from 110 to 250 µm, while the minimal diameters range between 93 and 198 µm. Post hoc tests show a significant difference for the values of the femur vs. tibia, as well as for the values of the tibia vs. metatarsus.

As far as the horizontal assessment of the femur is concerned, the goat (*Capra hircus*) vs. sheep (*Ovis aries*) pair appears to be the most differentiated throughout the series of values for the primary and secondary osteonal series (except for the minor axis of the primary osteons).

For tibia, in the case of the secondary osteonal unit, the goat (*Capra hircus*) vs. sheep (*Ovis aries*) pair appears again to be the most differentiated. For the primary osteonal units the situation is not as clear, and the roe deer (*Capreolus capreolus*) vs. goat (*Capra hircus*) and roe

deer (*Capreolus capreolus*) vs. sheep (*Ovis aries*) pairs (in terms of area and perimeter) are pointed out as the most unlikely series, while in terms of the major and minor axis the situation is uncertain.

For the metatarsus, it seems that there are no significant differences among the compared series, mainly in terms of secondary osteonal data. For the primary osteonal units, the difference appears constantly between the same two series, *Capreolus capreolus* vs. *Ovis aries* and *Capra hircus* vs. *Ovis aries*.

4.2. Fractal data and their relevance

Due to a lack of literature data in the area of the current study, no comparisons with values taken from other literature sources are possible. In the present study, in almost all cases, the fractal dimension was able to distinguish clearly between the bone samples, while lacunarity provided complementary discriminatory values that made the differentiation possible.

Interspecifically, for the *Capreolus capreolus* vs. *Capra hircus* pair, the tibial bone pattern seems to be alike. Intraspecifically, the femoral and tibial bone pattern of *Capreolus capreolus* showed close values for fractal dimension and lacunarity.

4.3. Concluding remarks

This study brings to light some new morphometric data for the species discussed, data that have never been published, to the best of our knowledge.

As far as morphometric data is concerned, the area of the secondary osteons does not appear to be the most reliable item that one can use for interspecific differentiation (at least for these skeletal segments), although some earlier papers suggest that (9). Our data showed some reliable differences for *Ovis aries* vs. *Capra hircus* in the case of the femur and tibia, but failed to show such a difference in the case of the metatarsals. The same applies to the perimeter of the secondary osteons: the *Ovis aries* vs. *Capra hircus* pair was the reliable pair in the case of the femur and tibia. However, in the case of the metatarsals, our data show no reliable differences. A similar tendency is displayed by the data of the major axis of the secondary osteon for the *Capra hircus* vs. *Ovis aries* pair. The other items associated with the structures of the secondary osteons (maximal diameter) and primary osteons (area, perimeter, axis) show no reliable differences, as each of the elements shows different sets of combinations that seem to be involved in the differentiation.

Fractal dimension and lacunarity data show that *Capreolus capreolus* bones have similar pattern features and complementary evaluation is needed to determine the differences between them. One such parameter might be succolarity, another fractal parameter that can quantify the texture by taking into consideration how a fluid would flow through certain "obstacles" on the studied structure (29).

For a comparison among species (interspecific), the values of the morphometric data are less powerful when compared with the fractal data. This is a significant conclusion, as the morphometric data failed to show such a difference (with respect to secondary osteonal data

as well as primary osteonal metric data). Even though morphometry showed differences within species in regards to different hind limb bones, the fractal dimension showed much clearer and more powerful differences among the series, with the exception of a single bone (tibia). Although the lacunarity data alone were not able to distinguish bone samples at the same level as fractal dimension, when taken together, these two fractal parameters show great potential.

As far as intraspecific differences are concerned, the morphometry data showed some constant differences for at least one pair (secondary and some of the primary osteonal area in *Capra* vs. *Ovis*), but failed to demonstrate the constantly present differences for the other species pairs under study. The fractal dimension works much better in this respect, pointing out an almost complete series of 3 differentiated bones within the species (with the exception of the bones of *Capreolus capreolus* that show only 2 differentiated species).

In our study the utility of fractal analysis, as an analytical tool alongside the morphological assessment (both qualitative and quantitative), shows a promising potential for differentiating bone types between species and/or within a species. Further in-depth studies are needed to establish a clear pattern-map, with practical application in the specific morphological identification of bone fragments.

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