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ORIGINAL ARTICLE

Elliptical Fourier descriptors of outline and morphological analysis in caudal view of foramen magnum of the tropical raccoon (*Procyon cancrivorus*) (Linnaeus, 1758)



Descripteurs elliptiques de Fourier du contour et de la morphologie en vue caudale du foramen magnum du raton-laveur tropical (Procyon cancrivorus) (Linnaeus, 1758)

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KEYWORDS

Elliptical Fourier analysis; Foramen analysis; Morphology; Procyon cancrivorus; Sexual dimorphism

Summary

Objectives. — To evaluate sexual-size dimorphism and attempt at categorization of interindividual shapes of foramen magnum outlines using Fourier descriptors which allow for shape outline evaluations with a resultant specimen character definition.

Materials and methods. — Individual characterization and quantification of foramen magnum shapes in direct caudal view based on elliptical Fourier technique was applied to 46 tropical raccoon skulls (26 females, 20 males).

Results. — Incremental number of harmonics demonstrates morphological contributions of such descriptors with their relations to specific anatomical constructions established. The initial harmonics (1st to 3rd) described the general foramen shapes while the second (4th to 12th) demonstrated fine morphological details. Sexual-size dimorphism was observed in females (87.1%) and 91.7% in males, normalization of size produces 75% in females and 83% in males. With respect to foramen magnum dimorphism analysis, the result obtained through elliptic Fourier analysis was comparatively better in detail information of outline contours than earlier classical methods. The first four effective principal components defined 70.63% of its shape properties while the rest (22.51%) constituted fine details of morphology.

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Conclusion. — Both size and shape seems important in sexual dimorphisms in this species, this investigation suggest clinical implications, taxonomic and anthropologic perspectives in foramen characterization magnum characterization and further postulates an increased possibility of volume reduction cerebellar protrusion, ontogenic magnum shape irregularities in the sample population with neurologic consequences especially among females.

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Résumé Ce travail vise à évaluer le dimorphisme sexuel de taille et à tenter de catégoriser les formes inter-individuelles du contour du foramen magnum en utilisant des descripteurs de Fourier qui permettent des évaluations des contours de forme avec une définition de caractère de l'échantillon résultant. La caractérisation et quantification individuelles des formes du foramen magnum en vue caudale directe basée sur la technique elliptique de Fourier ont été appliquées à 46 cranes de ratons-laveurs tropicaux (26 femelles, 20 mâles). Le nombre croissant d'harmoniques démontre les contributions morphologiques respectives de ces descripteurs et leurs relations avec des caractères anatomiques spécifiques. Les harmoniques initiaux (4e à 3e) ont décrit les formes générales tandis que les suivants (4e à 12e) ont démontré des détails morphologiques fins. Le dimorphisme sexuel de taille a été observé chez les femelles (87,1 %) et chez les mâles (91,7 %) ; la normalisation de la taille amène à 75 % chez les femelles et à 83 % chez les mâles. En ce qui concerne l'analyse du dimorphisme du foramen magnum, le résultat obtenu grâce à l'analyse de Fourier elliptique s'est avéré comparativement meilleur dans la caractérisation détaillée des contours que les méthodes classiques antérieures. Les quatre premiers composants principaux efficaces ont défini 70,63 % des caractéristiques de forme, tandis que le reste (22,51 %) offrait et révélait de nombreux détails de morphologie. La taille et la forme semblent importantes dans le cadre du dimorphisme sexuel de cette espèce. Nos résultats suggèrent des perspectives taxonomiques et anthropologiques dans l'analyse de la forme du foramen magnum et suggère la possibilité accrue de réduction de volume de la protubérance cérébelleuse, des irrégularités de forme d'origine ontogénétique, avec des conséquences neurologiques possibles, en particulier chez les femelles.

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Introduction

The foramen magnum by its diverse morphology presents some peculiar challenges in attempts at its characterization, the caudal view shape of the foramen is important in both neurological and anthropological studies [1,2]. Classical metric quantification methods in assessment of the foramen magnum scarcely applied with reference to limited and subjective available information for cephalometric evaluations [3]. Difficulties in precise homologous point location for repeatability and biological shape pattern recognition and discrimination interpretations could explain this difficulty. In anthropological investigations, accurate description of individuals is necessary for forensic studies on species from different ecological populations, skeletal remains, taxonomic and in evolutionary processes investigations [4–7].

Size, shape and architecture of foramen magnum present important race and individual variations [2] and as seen in raccoons (*Procyon cancrivorus*); indices among subspecies peak within juveniles' age group especially in the smaller subspecies [8]. Morphology of the foramen outline in the present species remains poorly described. The most caudoventral portion morphology of this structure also offers variations, which may be of further relevance in classification of the family [9].

Females, in this regard have about 15% attenuated size development of the skull bones as was confirmed by Watson et al. [10]. As in canids, the possibility of cerebellar

protrusion occasioned by volume reduction of the posterior fossa, syringomyelia and neurological disorders [7,9] remains potent with observation of open dorsal notches, which has been frequently reported associated with captive breeding and domestication attempts [11].

Advances in morphometry especially through computational biology introduced approaches in which homology of landmark points are not important while details of shape outlines can be explained geometrically, Elliptical Fourier method has been utilized by Dixon et al. [12] and Urbanová [13] after being initiated by Kuhl and Giardina [14] in the description of complex outlines but the methodology still remains largely unknown [15,16].

The purpose of this study is to morphologically analyze the elliptical Fourier descriptors of the foramen magnum outlines using variation—covariation by stepwise reconstruction in separate gender classes with contributions of the principal components of the coefficients as developed by Iwata and Ukai [17] to assign quantities, compare interindividual variability and evaluate dimorphisms.

Materials and methods

Osteological materials generation and categorization

This in vitro experiments involved 46 adult tropical raccoons (*P. cancrivorus*) comprising of 26 females and 20 males

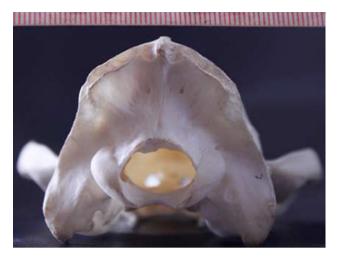


Figure 1 Picture of *Procyon cancrivorus* showing Foramen magnum in caudal view.

obtained from the wild, the heads were taken over a time space of two years and skulls macerated. The material was obtained with permission from the Department of Veterinary Anatomy, Faculty of Veterinary Medicine, and University of Ibadan, UI-ACUREC/App/2015/055.

Only adult skulls with fully erupted molars were selected for the study, gender categorization was then carried out. Transparent tracing paper with Cartesian co-ordinate was placed on these images represented on the x, y co-ordinate (described below) with a dorsoventral illumination of the foramen.

Foramen magnum outlines extraction

Photographic images of the caudal aspect of the specimen skulls (Fig. 1) were taken with digital camera Canon EOS 1200D equipped with EF-S 18-55 IS 11 kit, with Hamas tripod plumb and stabilizer, in a direct horizontal focal axis of the central point of foramen Images were taken at constant distance of 47 cm between camera and foramen magnum condyle in auto mode without mandibles in place. The outer rim of this foramen was screened off with polysiloxane putty. Images obtained were processed as 2179×430 pixels, 24-bit depth BMP pictures with Microsoft paint format as appropriate for SHAPE chain coder program software package for 2-dimensional quantification and evaluation of biological shapes recognition and visualization. Chain code is a coding system for describing geometrical information about contours in numbers 0-7 before binarizing a full color picture to black and white. Scanned outlines (in y to x direction) of the tracings were digitally processed as earlier explained in a clockwise direction and data on contours foramen magnum outlines was stored as a pair of Cartesian co-ordinates (x, y) on a scale of 50 mm [12-14,17].

Size normalization of foramen magnum outlines coefficients

The foramen magnum outlines coefficients were also recalculated after size normalization of descriptors to be invariant of size and was based on the first harmonic (first ellipse) using Chc2Nef SHAPE ver. 1.3 for obtaining elliptic Fourier descriptors [17] values of the enclosed area for all samples.

The elliptic Fourier sequences

The elliptic Fourier sequences of co-ordinates descriptors of contours (x and y) as described by Dixon et al. [12] are expanded thus Fig. 1 (supplementary material) which allows the description of an outline by ordered series of harmonics, the geometry of each corresponds to an ellipse.

For the present purpose, four new descriptors were defined for each harmonic:

- major axis length/2;
- minor axis length/2;
- orientation of the major axis;
- angle (θ) of phase corresponding to the position of the first point on the ellipse.

Buck [18], each harmonic ellipse was also evaluated by its size (Major axis length/ $2 \times \text{Minor axis length/}2$) as well by its peculiar anisotropy.

Where A_n , B_n , C_n , D_n are the descriptors for n number of harmonics.

Elliptical Fourier reconstructions

Stepwise reconstructions of foramen magnum outlines were performed by increasing the number of harmonics using descriptors derived from elliptical Fourier analysis.

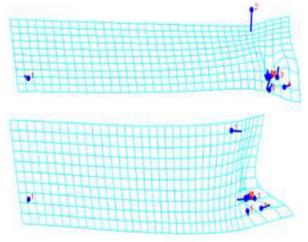
Principal component analysis

Based on variance—covariance, matrix of normalized coefficients in shape outline descriptors conducted by 'PrinComp' because coefficients with small variance and covariance values are generally not significant in explaining morphological variations are calculated and now employed in a derivation of principal components containing all information regarding the contour shapes in the first 12 harmonics as described by [19]. This was also visualized after analysis with MorphoJ software version 1.60b [20] using Grid wire depiction of components of shape in the foramen magnum of *P. cancrivorus* (Fig. 2).

Statistical analysis

The data of this elliptical Fourier analysis were analyzed with Paleontological statistics software (PAST) version 3.0 [21] package utilizing summary statistics (mean \pm standard deviations) for the descriptors. A gender based discriminant analysis with an increment in elliptical Fourier descriptor numbers was performed, the significance of the discriminant function was assessed by Bonferroni post-tests statistics with level of significance set at 0.05.





PC 2



PC 3

Figure 2 Grid representations of contributions of principal components of foramen magnum outline shape contour in *Procyon cancrivorus* for the first 18 harmonics aligned by principal axis.

Results

Quantitative analysis of the elliptical Fourier descriptors

Summary statistics of the descriptors of first 12 harmonics of foramen magnum outlines in *P. cancrivorus* studied (26 females and 20 males) for size-normalized outlines and magnitude with anisotropy of their ellipses are shown in Tables 1—3. Covariance analysis number of analyzed harmonics = 20, Normalized elliptical Fourier coefficient a1, b1 and c1 and d1 are the constant descriptor coefficients. Visual representations of the first 12 harmonics are given on Fig. 2.

The 1st harmonic demonstrated highest values of the elliptical descriptors in terms of their major and minor axis/2 as well as the magnitude of their ellipses irrespective of gender, and followed a gradient decline in values as the harmonics increased (Tables 1–3) especially among males. Female samples in that regards did not follow a similar pattern; but increase in value from the 2nd harmonic through the 7th followed by a gradual decline to the 12th harmonic. From the 5th harmonic (males), these descriptors presented values which were inferior or equal to 15% of the 1st harmonic value, and became minute after the 12th harmonic but the higher orders were retained in the tables for fine details.

The orientations of the major axis (between 0.88 to 103) (Tables 1 and 2) corresponding to the third elliptical descriptor is associated with no particular harmonic order but characteristic of each.

In Table 3, the elliptical anisotropy (1.05 to 3.21) was consistent with each harmonic but presented no harmonic order relationship.

Results of size normalization showed a marked reduction of descriptor values related to amplitude according to harmonics (factor around 1/17 to 1/40) in accord with harmonics, the angular orientation of the elliptical major axis was also markedly altered (1/2).

Morphological analysis of the stepwise reconstructions

Step by step reconstructions of the foramen magnum outline in *P. cancrivorus* from the 1st to the 12th harmonics are represented on Fig. 3 which demonstrated more accuracy with increasing number of harmonics used and displayed visually by a representation of the fit index (reconstructed vs. original values); this accuracy by using the first 12 harmonics was in the range of 97%.

The geometric contributions of each principal components of elliptical Fourier descriptor and their morphological analysis as exposed by the stepwise reconstructions shown on Fig. 3 using the first 7 harmonics finer features of the foramen were described by harmonics of higher order.

Out of 76 (seventy-six) principal components coefficients with 12 analyzed harmonics and a total variance of 4.10859E-001, the first three components effectively described the foramen magnum.

Major morphological characteristics of the foramen magnum in the studied samples were fully described by the 4th harmonic and elucidated perfectly by the 12th, the right and left dorsolateral rims in a dorsal direction at caudal view from a graphic central point was the first to be described in relation to the central point with the use of the first harmonic whereas the direct dorsal limit and notch was precisely explained by the 2nd principal component while PC3 described the ventral limit at the convergence of the condylar rims (Figs. 2 and 3) the 3rd harmonic demonstrated some variability in this respect. The asymmetric components appeared correctly between 6th to 12th harmonics. The last portion to be reconstructed was the ventral notch (8th-12th), it was completely described by the 12th. Vector analysis of this shape outline (below) (Fig. 2) further visualized the direction of each contribution.

Sexual dimorphism analysis

Significant sexual variation in shape of foramen magnum outlines between females and males were shown by the discriminant elliptical Fourier descriptor analysis and by the Mahalanobis distance between the group centroids (5.25) and significant at (P < 0.01). Significant but lesser Mahalanobis distance differences still existed after size normalization of the foramen magnum outlines (3.41; P < 0.02), principal component analysis for the first 4 harmonics for the descriptors coefficients and size-normalized outline after

Table 1 Sex based *P. cancrivorus* foramen magnum outlines in caudal view Elliptical descriptors for the first Fourier 10 harmonics in summary (26 females, 20 males).

	Females			Males			
	Major axis length/2	Minor axis length/2	Orientation of major axis (°)	Major axis length/2	Minor axis length/2	Orientation of major axis (°)	
Harmonics	$Mean \pm SD$			$\mathrm{Mean}\pm\mathrm{SD}$			
1	89.40 ± 9.20	67.83 ± 4.77	29.67 ± 3.44	89.70 ± 16.55	84.95 ± 22.32	28.87 ± 5.44	
2	70.21 ± 8.90	66.65 ± 5.86	103.19 ± 4.47	85.77 ± 21.93	78.71 ± 26.23	27.68 ± 7.64	
3	77.57 ± 15.44	69.32 ± 12.85	36.91 ± 3.10	87.62 ± 25.11	69.06 ± 35.99	27.50 ± 4.96	
4	82.17 ± 10.20	72.17 ± 13.55	28.13 ± 2.21	79.46 ± 30.10	53.73 ± 30.15	26.29 ± 12.72	
5	85.46 ± 10.17	72.15 ± 14.09	30.94 ± 3.05	52.43 ± 35.40	34.47 ± 31.66	21.76 ± 13.81	
6	87.45 ± 12.09	67.27 ± 17.88	36.30 ± 9.85	50.31 ± 36.07	32.69 ± 31.44	18.97 ± 16.89	
7	88.64 ± 20.20	76.5 ± 25.41	27.11 ± 1.40	39.87 ± 31.77	19.17 ± 24.02	18.87 ± 14.88	
8	82.28 ± 21.8	64.71 ± 26.98	30.53 ± 6.59	35.21 ± 32.16	30.00 ± 28.07	16.17 ± 14.12	
9	67.47 ± 28.96	50.75 ± 38.75	24.40 ± 14.60	32.47 ± 31.26	17.00 ± 22.88	11.28 ± 12.71	
10	52.67 ± 30.61	39.60 ± 28.98	25.15 ± 10.48	30.23 ± 35.00	25.46 ± 31.68	13.51 ± 14.70	
11	28.24 ± 10.21	13.68 ± 21.55	9.76 ± 10.812	14.21 ± 21.01	10.76 ± 22.00	10.59 ± 10.66	
12	18.54 ± 6.00	8.36 ± 6.88	7.09 ± 10.77	9.51 ± 20.00	6.91 ± 18.02	$\textbf{7.95} \pm \textbf{9.07}$	

Table 2 Sex- based size-normalized *P. cancrivorus* foramen magnum outlines in caudal view showing elliptical descriptors of first 10 Fourier harmonics in summary (26 females, 20 males).

	Females			Males			
	Major axis length/2	Minor axis length/2	Orientation of major axis (°)	Major axis length/2	Minor axis length/2	Orientation of major axis (°)	
Harmonics	${\sf Mean}\pm{\sf SD}$			${\sf Mean}\pm{\sf SD}$			
1	5.18 ± 3.00	1.61 ± 3.00	5.65 ± 7.58	6.40 ± 1.68	6.07 ± 9.83	0.08 ± 0.24	
2	5.01 ± 0.01	4.76 ± 0.03	4.45 ± 11.45	6.21 ± 0.16	5.62 ± 0.04	7.14 ± 12.50	
3	$\textbf{5.54} \pm \textbf{0.01}$	4.95 ± 0.03	4.06 ± 13.43	6.25 ± 1.27	4.93 ± 0.06	10.26 ± 12.48	
4	5.87 ± 0.00	$\textbf{5.15} \pm \textbf{0.01}$	11.91 ± 16.84	5.67 ± 0.03	$\textbf{3.84} \pm \textbf{0.01}$	12.45 ± 16.45	
5	6.10 ± 0.01	$\textbf{5.15} \pm \textbf{0.01}$	12.09 ± 19.05	3.74 ± 0.01	$\textbf{2.46} \pm \textbf{0.01}$	14.62 ± 12.61	
6	6.24 ± 0.01	4.80 ± 0.01	13.88 ± 23.46	3.59 ± 0.01	$\textbf{2.34} \pm \textbf{0.01}$	10.29 ± 22.18	
7	6.33 ± 0.00	$\textbf{5.46} \pm \textbf{0.00}$	$\textbf{9.83} \pm \textbf{20.44}$	$\textbf{2.84} \pm \textbf{0.01}$	$\textbf{1.36} \pm \textbf{0.01}$	6.48 ± 25.19	
8	6.31 ± 0.00	4.62 ± 0.00	9.16 ± 24.02	2.51 ± 0.01	$\textbf{2.14} \pm \textbf{0.00}$	1.98 ± 15.33	
9	4.78 ± 0.00	$\textbf{3.62} \pm \textbf{0.00}$	8.52 ± 20.54	2.31 ± 0.01	$\textbf{1.21} \pm \textbf{0.00}$	1.02 ± 32.32	
10	3.17 ± 0.00	2.83 ± 0.00	$\textbf{5.60} \pm \textbf{20.38}$	$\textbf{2.14} \pm \textbf{0.01}$	$\textbf{1.78} \pm \textbf{0.00}$	0.93 ± 14.04	
11	1.01 ± 0.00	1.00 ± 0.00	2.97 ± 15.09	2.51 ± 0.01	1.07 ± 0.00	0.21 ± 12.59	
12	$\textbf{0.47} \pm \textbf{0.00}$	0.06 ± 0.00	$\textbf{0.96} \pm \textbf{10.71}$	2.03 ± 0.01	$\textbf{0.88} \pm \textbf{0.00}$	$\textbf{0.13} \pm \textbf{9.88}$	
P. cancrivorus: Procyon cancrivorus.							

which the multivariate analysis was no longer necessary (Wilks' lambda values P > 0.5) after these harmonics. Table 4 shows the principal component contributions of the foramen being studied and for the first 10 reconstructions (Fig. 3) the principal component scores between the genders. Variance-covariance matrix showed a higher variance (0.024) in males and (0.0086) in females. A step by step discriminant analysis of harmonic increment and Mahalanobis distance revealed a gradual increase in the difference between both sexes (Figs. 4—6), the analysis became insignificant after the 12th harmonic.

The proportion of individuals presenting a significant sexual dimorphism of the foramen magnum outline was 87.1% in females and 91.7% in males (Table 5), 75% and 83% in females and males respectively after size normalization (Table 5). Size factor elimination using size normalization yielded a 12.1% and 8.7% accuracy decline in females and males respectively, the non-occurrence of significant sexual dimorphism was observed fairly less frequently in males (8.3%) and (16.7%) in females; after normalization of size this was also the case but with a reduced difference; 16.7% and (33.3%) in males and females respectively as shown in Table 5, these tables displays a graphical pattern of discriminant scores distribution for foramen magnum outlines and for size-normalized outlines respectively showing patterned anatomical variation between sexes. Gender discrimination was reduced in size-normalized outlines while overlap was pronounced.

Table 3	Sex based size-normalized P. cancrivorus foramen magnum outlines in caudal view showing magnitude and anisotropy
of the ell	lipses of the first 10 Fourier harmonics in summary (26 females, 20 males).

	Females		Males		
	Elliptical magnitude	Elliptical anisotropy	Elliptical magnitude	Elliptical anisotropy	
Harmonics	Mean \pm SD		Mean \pm SD		
1	8.34 ± 0.20	3.21	38.84 ± 0.07	1.05	
2	23.39 ± 0.63	1.07	34.90 ± 0.05	1.11	
3	27.42 ± 0.76	1.12	30.81 ± 0.07	1.27	
4	30.23 ± 0.23	1.14	21.77 ± 0.01	1.48	
5	31.41 ± 0.72	1.18	9.20 ± 2.29	1.52	
6	29.95 ± 0.63	1.30	8.40 ± 0.00	1.53	
7	34.56 ± 0.56	1.16	3.86 ± 7.93	2.08	
8	29.15 ± 0.11	1.37	5.37 ± 9.83	1.17	
9	17.30 ± 0.09	1.32	2.79 ± 4.33	1.91	
10	8.97 ± 0.08	1.12	3.81 ± 3.57	1.20	
11	1.01 ± 0.07	1.01	2.69 ± 2.81	2.35	
12	0.03 ± 0.88	7.93	1.79 ± 3.00	2.31	

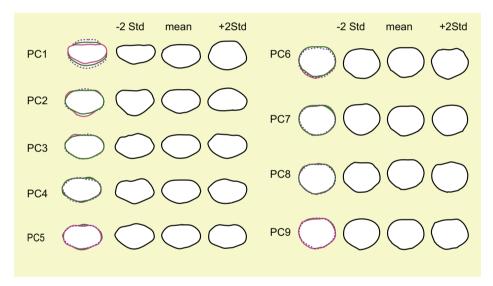


Figure 3 Contour reconstructions of the foramen magnum in *Procyon cancrivorus* for the first 12 harmonics [Overlapped reconstructions- (-2std = purple; mean = green; +std = pink)].

Discussion

Interests of Elliptical Fourier methods in Foramen magnum morphological analysis

Reproducibility of experimental procedures through automatization of image analysis with personal software use in microcomputers has enhanced its usefulness among less dexterous users. Irrespective of its morphological complexity, the present method permits the quantification of the shape of an outline with precise individual variations or signatures, which favors polymorphism investigations. Elliptical Fourier analysis introduces scientific perspectives with ecological and anthropological interests in

characterization of foramen magnum outlines. According to Urbanová [13], the contributions of the elliptical Fourier descriptors can be associated with geometrical interpretations more easily than classical Fourier coefficients and precise anatomic architecture deduced. If the foramen magnum is described by morphometric methods based on landmarks, its curved shape mainly provides type-2 landmarks, which are points of maximum curvature [22]. Such landmarks are likely to be sensitive to measurement error. Elliptic Fourier descriptors allow an outline analyses with a low loss of contour information.

The first harmonics (1st to 4th) described the general shape of the foramen magnum outline in the present species while the later harmonics (5th to 12th) elucidates finer and other peculiar details of this structure (Fig. 3). Visual

Table 4 Eigen value and proportions showing coefficients whose percentage proportion values are greater than 1.

Eigen value	Proportion (%)	Cumulative (%)	> 1/76
PC1	1.656150E-002	27.00	27.00 ^a
PC2	1.230742E-002	20.07	47.07 ^a
PC3	7.947657E-003	12.96	60.03 ^a
PC4	6.497890E-003	10.60	70.62 ^a
PC5	4.464331E-003	7.28	77.90 ^a
PC6	3.518633E-003	5.74	83.64 ^a
PC7	2.029116E-003	3.31	86.95 ^a
PC8	1.589538E-003	2.60	89.54 ^a
PC9	1.028969E-003	1.68	91.22ª
PC10	8.684068E-004	1.42	92.64 ^a

^a indicates coefficient values which are both greater than 1 and significant.

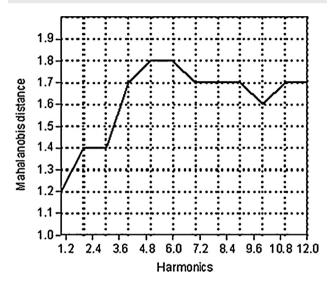


Figure 4 Analysis of sexual dimorphism of the foramen magnum outlines in caudal view. Fit diagram of increment in Elliptical Fourier harmonics calculated from discriminant analysis and Mahalanobis distance.

observation of the convergence between the reconstructed and the original outline can be appreciated with fit index quantification. An accurate and detailed structural data of the foramen magnum outline is made available by the use of a maximum of only 12 harmonics where similarity with the original is demonstrated, this method therefore serves a viable means of data compression.

Ontogeny

The development of the foramen magnum depends largely on genetic composition, ecology and diet as suggested by [8,23], in comparing skull dimensions in N.p. viverrinus and N.p. Usuriensis, N.p. lotor differs with such diet of the cancrivorus subspecies [24] consisting of 40% invertebrates, 33% plant materials, 27% vertebrates [8]. This carnivoreomnivore thus possesses a peculiar occipital morphology relative to similar sized carnivores [25,26]. Morphological variations observed in individuals has however not been ascertained to be related any pathologic manifestations.

Assessment method comparison

Available literary information on quantitative or metric methods of investigating sexual dimorphic characters of the foramen magnum in some domestic species included Manoel et al. [11] and Onar et al. [27]. Watson et al. [28] and Janeczek et al. [3] studied American Staffordshire terriers utilizing conventional means and revealed a higher foramen magnum index in male dogs. Dimorphic non-metric traits of the structure under current investigation [27,29] confirmed a lighter variation in size between the sexes though unconfirmed studies suggests environmental inputs aside genetic considerations of recruitment pathways (Co-option) of existing genes for new functions as a fundamental mechanism for creating new phenotypes [30].

Elliptical Fourier technique as used in this evaluation affords an illustration of clearer principal components of dimorphism in shape observed by other earlier methodologies (Table 5). Information made available on the foramen magnum outline over 840 sampled points may be postulated to give better and precise dimorphic shape differences in contrast to fewer reference points assessed by conventional means which provide only debatable morphological information.

Female—male differences in foramen magnum morphology

Male—female differences in skeletal attributes have been evaluated extensively in most species; [10,31,32] and itself a precondition in sex discrimination for forensic anatomy [33]. However, studies on the foramen magnum outline are less abundant in literature in contrast to other bony structures. Similar to existing works on other structures, observations of non-significant sexual dimorphism as explained by Iscan and Steyn [31] and Nowicki et al. [34] shown in (Table 5); there is a reduction of such population in females while an increase in proportion among males in this study which can be attributed to sampling error, non-adequate performance or an affirmation of minimal sexual-size non-similarity in this species reported from previous works [8,25]. Further investigation may be necessary to determine precisely if this observation is biologically viable.

Aspects of size and shape components in foramen magnum morphology

Sexual dimorphism in foramen magnum morphology is based on these two components but the size component contributes more and considered more important in this respect [27,33], in most species, male bony structures are larger and more robust than those of females [33] with size component making up about 70% of sexual dimorphic differences and excluded the probability of an accurate discriminant function construction if size factor was excluded, shape component constituted less than 23% of sexual variations. Elliptic Fourier method, in absence of size normalization takes size and shape components of the foramen magnum into consideration whereas after size normalization where only accounts for the shape component in foramen

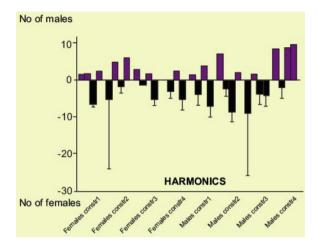


Figure 5 Analysis of sexual dimorphism of the foramen magnum outlines in caudal view. Distribution of the discriminant scores resulting from discriminant analysis of the elliptical Fourier descriptors (26 females and 20 males).

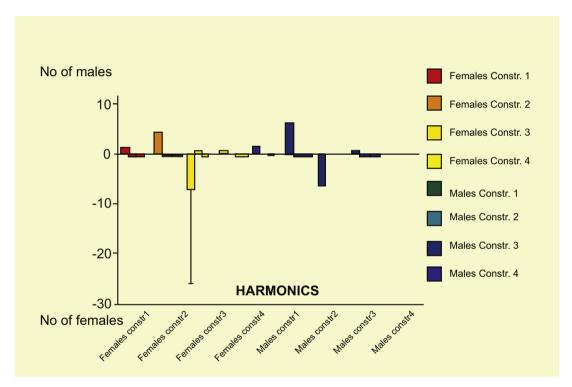


Figure 6 Analysis of sexual dimorphism of the size-normalized foramen magnum outlines in caudal view. Distribution of the discriminant scores from discriminant analysis of the Elliptical Fourier descriptors (26 females and 20 males).

magnum outlines is given (eliminating size component), for the former, 87.1% of sexual dimorphism was observed in females, 91.7% in males (Table 5) while for the former 75% and 83% in females and males respectively (Table 5) corresponding to 12.1% and 8.7% decrease accuracy in females and males. Conventional method of circular shape measurement could not distinguish between size and shape elements as presently demonstrated. Within a safe margin

of accuracy, we postulate a more pronounced importance of shape expression in sexual dimorphism in *P. cancrivorus* species.

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Sex group	No. individuals	Individuals classified as females		Individuals classified as males	
		No.	%	No.	%
Females	26	23	87.1	3	12.9
Males	20	2	8.3	18	91.7
Females	26	20	75	4	15
Males	20	3	17	17	83

Table 5 Analysis of sexual dimorphism classifications procedure and size-normalized of foramen magnum outline performed from the Elliptical Fourier descriptors (26 females and 20 males).

Disclosure of interest

No.: number.

The authors declare that they have no competing interest.

Appendix A. Supplementary data

Supplementary material associated with this article can be found in the online version available at https://doi.org/10.1016/j.morpho.2017.06.001.

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