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Skull morphometry and vault sutures of *Myrmecophaga tridactyla* and *Tamandua tetradactyla*

Camila M. de S. Hossotani, José Ragusa-Netto & Helder S. e Luna

Programa de Pós-Graduação em Biologia Animal Universidade Federal do Mato Grosso do Sul-Campo Grande MS/Campus II de Três Lagoas, Av. Ranulpho Marques Leal, 3484 – Distrito Industrial, 79610-100 Três Lagoas, MS (chossotani.bio@gmail.com)

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ABSTRACT. This study aimed to examine the relationship between skull size and the level of cranial vault suture closure. A total of 50 *Myrmecophaga tridactyla* Linnaeus, 1758 and 178 *Tamandua tetradactyla* Linnaeus, 1758 skulls were analyzed in relation to 18 skull dimensions. The skulls were grouped into three levels of suture closure: no sutures closed (level 0), one or all the fallowing sutures closed: interfrontalis, sagitalis and coronalis (level 1) and all sutures closed (level 2). The results indicated that among the 18 variables measured, 17 showed significant differences ($p \leq 0.01$) between level 0 and level 1 skulls of *T. tetradactyla*; as well as between level 0 and level 1, and level 0 and level 2 skulls of *M. tridactyla*. *M. tridactyla* level 1 and level 2 had no significant difference among any of the 18 dimensions. The foramen magnum height in both species showed no significant difference ($p > 0.05$) among any suture categories. In principle, suture closure level and cranial dimensions are related. The specimens with larger cranial dimensions showed greater number of cranial vault sutures closed for both species of anteaters. *Tamandua tetradactyla* and *M. tridactyla* specimens with none of the cranial vault suture closed have a foramen magnum height similar to those with cranial vault suture closed.

KEYWORDS. Age, anteater, cranial morphology, xenarthra.

RESUMO. Morfometria craniana e suturas da abóboda de *Myrmecophaga tridactyla* e *Tamandua tetradactyla*. O presente estudo teve como objetivo examinar a relação das dimensões do crânio e o grau de fechamento das suturas da abóboda craniana. Foram analisados 50 crânios de *Myrmecophaga tridactyla* Linnaeus, 1758 e 178 de *Tamandua tetradactyla* Linnaeus, 1758 em relação a 18 dimensões do crânio. Os crânios foram divididos em três grupos quanto ao grau de fechamento da sutura: nenhuma sutura fechada (grau 0), uma ou todas das seguintes suturas fechadas: interfrontalis, sagitalis e coronalis (grau 1) e todas as suturas fechadas (grau 2). Os resultados indicaram que das 18 dimensões 17 apresentaram diferença significativa ($p \leq 0{,}01$) entre grau 0 e grau 1 de *T. tetradactyla*; entre grau 0 e grau 1, e grau 0 e grau 2 de *M. tridactyla*. Indivíduos grau 1 e grau 2 de *M. tridactyla* não apresentaram diferença significativa em nenhuma das 18 dimensões. A altura do forame magno nas duas espécies não apresentou diferença significativa ($p > 0{,}05$) entre nenhuma das categorias de sutura (grau 0, grau 1 e grau 2). Em princípio, o nível de fechamento da sutura e as dimensões cranianas estão relacionados. Os espécimes com dimensões cranianas maiores apresentaram maior número de suturas de abóbada craniana fechadas para ambas as espécies de tamanduás. Os espécimes de *T. tetradactyla* e *M. tridactyla* sem sutura da abóbada craniana fechada possuem altura do forame magno semelhante àquelas com sutura da abóbada craniana fechada.

PALAVRAS-CHAVE. Idade, morfologia craniana, tamanduá, xenartra.

The Giant anteater (*Myrmecophaga tridactyla* Linnaeus, 1758) is the largest anteater of the Myrmecophagidae (Pilosa); it has a wide Neotropical distribution, occurring from south Belize and Guatemala to the north extreme of Argentina (Eisenberg & Redford, 1992). *Tamandua tetradactyla* Linnaeus, 1758, commonly called the lesser anteater, also belongs to Myrmecophagidae and is widespread in South America (Miranda *et al*., 2014). It is medium-sized with arboreal and terrestrial habits. These two anteater species have body and skull characteristics that reflect their habits of feeding on ants and termites (Eisenberg & Redford, 1992; Miranda, 2012).

The skull of *M. tridactyla* has a long, cylindrical rostrum region with a small oral opening, an incomplete zygomatic arch, and no fused jaws (Naples, 1999). *Tamandua tetradactyla* also has these skulls traits, although the rostrum region is proportionally smaller. In particular, the end of the snout has no fused structure, called the pre-maxilla, which is absent in *M. tridactyla* (Miranda, 2012). Both anteater species have the pterygoids expanded and united in the middle, with no interpterygoid vacuity, in which a soft palate is often present (Reiss, 2000). They have an elongated hard palate, which allows the housing of a long tongue without interference with breathing (Naples, 1999; Miranda, 2012).

Due to the unique skulls of anteaters, many studies have been developed on the morphology and phylogeny of this mammal group, although few investigations have focused on the timing of cranial suture (Rager *et al*., 2014). Nevertheless, there are some studies on xenarthrans, which used suture closure level as a parameter to estimate age (Naples, 1982; Squarcia *et al*., 2009), and this has also been shown in studies of other mammals (Schweikher, 1930; Cheverud, 1996; Cray *et al*., 2008). The focus on suture closure in cranial studies is common, mainly due to sutures being located between two developing bones of a skull increasing in size (Baer, 1954; Moss, 1954). Indeed, almost all mammal sutures are visible in young individuals, but may be too closed and almost imperceptive in adults. Thus, suture closure level has been assumed a valid tool to estimate age in skulls of mammals (Elbroch, 2006).

Knowledge of skulls is important for studies with museum specimens to avoid possible age effect on data (Loy, 2007). Thus, in addition to skull suture closure level, some studies on mammals have also utilized tooth analysis to determine an approximate age in skulls (Cheverud, 1996; Brunner *et al*., 2004). However, anteaters are toothless (Wetzel, 1975). Thus, due to the scarce information about the timing of suture closure in anteaters, and the absence of other cranial age parameters, the present study aimed to assess the relationship between ectocranial suture closure level and the skull dimensions of both *Myrmecophaga tridactyla* and *Tamandua tetradactyla*.

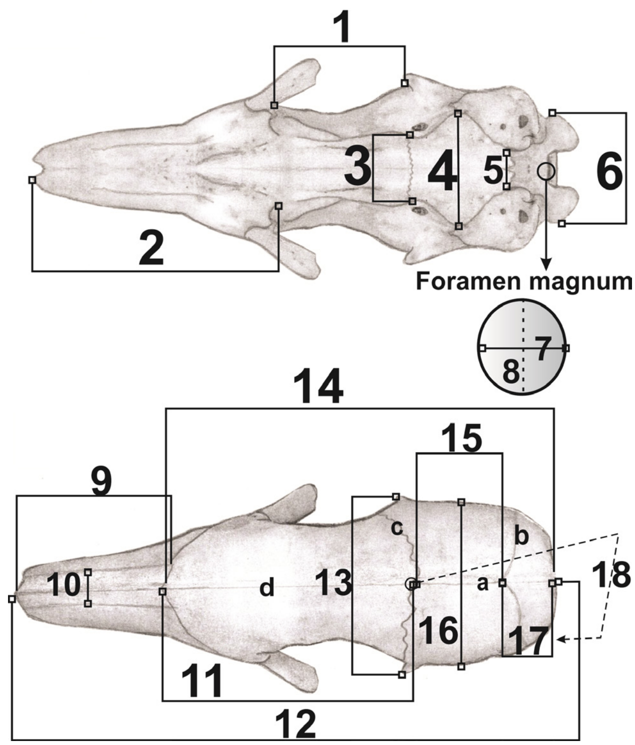

Fig. 1. Ventral and dorsal views of skull measurements of *Tamandua tetradactyla* (MZUSP 2463 ♀) used in morphometric analyses. The same procedure was used for *Myrmecophaga tridactyla* skulls. Cranial measurements used in this study: 1, orbital length (OL); 2, maxilla length (ML); 3, palatal width (PW); 4, pterygoids breadth (PtB); 5, inner nostril breadth (INB); 6, occipital condyle breadth (OCB); 7, foramen magnum diameter (FMD); 8, foramen magnum height (FH); 9, nasal length (NL); 10, nasal breadth (NB); 11, frontal length (FL); 12, skull length (SL); 13, posterior zygomatic arch breadth (ZB); 14, neurocranium length (NC); 15, parietal length (PL); 16, cranial breadth (CB); 17, Supraoccipital length (SOL); 18, cranial height (CH). Sutures analyzed: a, sagitalis; b, lambdoidea; c, coronalis; d, interfrontalis.

MATERIAL AND METHODS

A total of 228 skulls, 50 *Myrmecophaga tridactyla* and 178 *Tamandua tetradactyla,* were analyzed from the collections of Museu Nacional, Universidade Federal do Rio de Janeiro (MNRJ), Museu de Zoologia da Universidade de São Paulo (MZUSP) and Laboratório de Morfologia e Reprodução Animal da Universidade Federal do Mato Grosso do Sul campus II de Três Lagoas (LAMORA).

To classify skull vault suture closure level, a scale ranging from 0 to 2 was utilized as follows: level 0 – no sutures of the skull vault closed; level 1 – one or all of the fallowing sutures closed: interfrontalis, coronalis and sagitalis; and level 2 – all sutures of the skull vault closed. This method is similar to those used in other studies with mammals (Schweikher, 1930; Brunner *et al*., 2004; Cray *et al*., 2008) and anteaters (Wetzel, 1975).

The following sutures were examined: coronalis, sagitalis, lambdoidea and interfrontalis. The interfrontalis suture is between the frontal bones, the sagittalis suture is between the parietal bones, the coronalis suture is between the frontal and parietal bones, and the lambdoidea is the suture between the parietal and supraoccipital bones (Gardner *et al*., 1971). These four sutures of the cranial vault were the most preserved in the samples, the palatal region, for example, in many samples was very fragile or dismantling making it impossible to observe the sutures closure level with accuracy. Eighteen measurements (Fig. 1) were taken, with 150 mm ZAAS digital calipers to 0.01mm/0.0005" precision, a metal ruler and measuring tape.

In order to eliminate possible sexual dimorphism influence the presence of sexual dimorphism was verified for *T. tetradactyla* data. However, for the *M. tridactyla* data, it was not possible to verify the presence of sexual dimorphism because the collections visited had no gender information for all samples. Univariate tests were chosen according to normal distribution of the data. For *T. tetradactyla* data, the Shapiro-Wilk W (Shapiro & Wilk, 1965) normality test was employed and indicated non-normal distribution. Hence, a non-parametric Wilcoxon-Mann-Whitney test (Mann-Whitney) was carried out to assess differences among measurement medians between all skulls classified as level 0 and level 1 as well as between females and males of level 0 and level 1 for sexual dimorphism.

The Shapiro-Wilk W test (Shapiro & Wilk, 1965) was also employed on *M. tridactyla* data, and normal distribution was found. Therefore, an ANOVA test (analysis of variance) was employed to compare mean differences of skull measurements between skulls classified as level 0, level 1 and level 2. Posteriorly, a Tukey test was used for pairwise comparison among the three suture closure levels.

In addition to univariate tests, multivariate tests were also used. For the multivariate tests, specimens with missing measurement data were removed from the analyses. For both species, the multivariate test utilized was the NPMANOVA (non-parametric MANOVA). A principal components analysis (PCA) of the 18 variables (skull measurements) was carried out to inspect loadings values of variables important for group ordination. According to this, a MANOVA/CVC test for *M. tridactyla* and a discriminant/hotelling for *T. tetradctyla* data were carried out to compare groups. In the MANOVA/CVC test for *M. tridactyla*, the matrix values were log-transformed before statistical analyses.

Specimens examined. *Myrmecophaga tridactyla*: Amazonas state, MZUSP 5273♀; Pará state, MZUSP 19958♀, 5454♀; Goiás state, MNRJ 37561, 68176, 68177; São Paulo state, MZUSP 1170♂, 1172; Espírito Santo state, MZUSP 2415; Mato Grosso do Sul state, MNRJ 5073♀, 24828♀, MZUSP 34773(-), 7789♀, LAMORA 39.1♀, 30.1, 26.1, 34.1, 32.1 , 9.1, 33.1, 29.1, 11.1, 17.1, 1.1, 25.1, 22.1; Mato Grosso state, MNRJ 2332♂, 23945,64003, 64154, 64733, 71089, 71061, MZUSP 3727 ♀; Rio Grande do Sul state, MZUSP 1771. Unknown locality: MNRJ 1636, 1635, 1671, 3346, 23935, 23937, 23946, 23947, 23948, 24098, 32694, MZUSP 7480, 3683, 32354, TX080035. *Tamandua tetradactyla*: Roraima state, MZUSP 9693♂; Amapá state, MNRJ 24849♀, 23951♀; Amazonas state, MZUSP 4991♀, 5443♂, 928♀, 5439♀, 19949♀, 5139♀, 4993♀, 5136♀, 5137♀, 4989♂, 5455♂, 5138♂, 10483♂, MNRJ 24832♀, 2341♂, 5967; Pará state, MZUSP 19954♀, 23593, 23594, 19953♀, 3652♀, 20002♀, 5453♀, 5440♀, 19961♀, 19951♂, 19959♂, 5456♂, 5451♂, 5448♂, 19986, 13484, 19952♀, 5454♀, 19972♀, 19960♀, 19962♀, 10708♀, 5236♀, 4978♀, 5237♀, 5239♀, 4979♀, 5240♀, 5238♀, 5442 ♀, 10484♀, 8999♂, 5449♂, 5234♂, 21239♂, MNRJ 5057♀, 5063♀, 5064♀, 5065♀, 5066♀, 5058♀, 5067♀, 5071♀, 5052♀, 5060♀, 5050♀, 5053♀, 5070♀, 5072♀, 5055♀, 5510♀, 5638, 5726♀, 5743♀, 23952♀, 4536♂, 5051♂; Acre state, MZUSP 7336♀, 19950♀; Tocantins state, MNRJ 75094; Piauí state, MNRJ 63520; Pernambuco state, MZUSP 19955♀, 19956♀; Alagoas state, MZUSP 7367♀, 7368♀, 7526♀, MNRJ 23950♀; Bahia state, MNRJ 10983, 2652, 9677♀, 10985♀, 9678♂, 67898; Espírito Santo state, MNRJ 24097♀, 5883♀, 5634♀, 5515♂; Minas Gerais state, MZUSP 3065♀, 3112♂, MNRJ 7620♀, 79028, 79027, 73484, 24850, 24852, 24855, 2335; Goiás state, MNRJ 5069♂, 3846♂, 5059♂, 5054♀, 5062♀, 24854, 4538♀, 5068♂, 4613, MZUSP 10641♀; Mato Grosso state, MZUSP 7038♀, 7036♀, 6334♀, 6335♀, 7037♀, 35104, MNRJ 64434; São Paulo state, MZUSP 836♂, 1905♂, 2838♀, 8471♂, 8305♂, 4088, 2997♀, 1862♀, 2837♀, 1861♂, 1813♂, 19964♂, 5763♂, 1927, 31990, 1283, 1769, MNRJ 11607♂; Mato Grosso do Sul state, MZUSP 7788♀, 20000, 4297, MNRJ 5056♀, 5061♂; Paraná state, MZUSP 2463♀; Santa Catarina state, MZUSP 1685A, 1762, 1685B, 1763; Rio Grande do Sul state, MZUSP 1442♀, 1770, 1441, 1771; Unknown locality, MZUSP 32620♂, MNRJ 43, 23965, 23956♀, 2334, 22397, 19987, 17507♀, 23963♀, 24851♀, 2685, 2327, 23953, 23966, 44, 79118, 43804, 24857, 24856, 24853, 72776, 41, 8343.

RESULTS

Tamandua tetradactyla sexual dimorphism. The results indicated no sexual dimorphism for all measurements between males and females skulls classified as level 1 (Tab. I). Skulls classified as level 0 had significant difference between males and females only for frontal length measurement (Tab. II).

Tamandua tetradactyla suture closure level. Only two specimens of *T. tetradactyla* were classified as level 2 (all sutures closed). Thus, level 2 was excluded from the statistical analysis for this species, which included only specimens classified as level 0 and level 1. Seventeen measurements had significant differences (Tab. III) between specimens classified as level 0 and level 1: foramen magnum diameter; occipital condyle breadth; cranial height; cranial breadth; orbital length; nasal breadth; posterior zygomatic arch breadth; pterygoids breadth; supraoccipital length; parietal length; inner nostril breadth; nasal length; palatal width; neurocranium length; frontal length; skull length and maxilla length. The foramen magnum height was the only measurement that did not show a significant difference ($p > 0.05$) (Tab. III). The level 0 specimens ($n = 53$) were visibly smaller than level 1 and some skulls still had the fontanel. The level 1 specimens were plentiful ($n = 125$) and showed, apparently, thicker bones than the immature specimens (Tab. III).

In addition, a non-parametric Wilcoxon-Mann-Whitney test (Mann-Whitney) was carried out to assess differences among frontal length medians between females classified as level 1 and level 0 (Tab. IV), as well as between males classified as level 1 and level 0 (Tab. V). The results corroborate with the suture closure level tests previously performed and indicated that even though there is a difference in size of frontal bone between females and males of level 0, this does not seem to be significant in relation to the suture closure level.

Tab. I. Results of Mann-Whitney test for sexual dimorphism of *Tamandua tetradactyla* Linnaus, 1758 skull measurements (in mm) mean and standard deviation (±) of specimens classified as level 1 (P, test significance; *n*, sample size; FMD, foramen magnum diameter; OCB, occipital condyle breadth; FH, foramen magnum height; CH, cranial height; CB, cranial breadth; OL, orbital length; NB, nasal breadth; ZB, posterior zygomatic arch breadth; PtB, pterygoids breadth; SOL, supraoccipital length; PL, parietal length; INB, inner nostril breadth; NL, nasal length; PW, palatal width; NC, neurocranium length; FL, frontal length; SL, skull length; ML, maxilla length).

Measurements	Suture closure level 1		P
	Female (*n* = 55)	Male (*n* = 28)	
FMD	12.9 ±1	12.8 ±1	0.5
OCB	23.7 ±1	23.6 ±2.5	0.8
FH	11.5 ±5.8	10.8 ±1.2	0.6
CH	32.4 ±1.6	31.9 ±1.4	0.1
CB	41.5 ±2.3	41.8 ±2.1	0.6
OL	32.9 ±2.4	33.0 ±2.7	0.6
NB	7.8 ±1.3	7.6 ±1.24	0.7
ZB	42.5 ±2.2	42.09 ±2.4	0.7
PtB	28.7 ±1.9	28.01 ±3.0	0.3
SOL	16.0 ±1.6	15.9 ±1.78	0.7
PL	21.4 ±1.9	24.25 ±10.65	0.5
INB	9.85 ±1.1	9.6 ±0.7	0.2
NL	43.8 ±2.6	43.5 ±4.6	0.9
PW	21.3 ±4	21.0 ±2.8	0.5
NC	87.07 ±4.5	85.5 ±8.8	0.9
FL	50.7 ±6.9	50.55 ±4.2	0.7
SL	128.9 ±5.6	128.1 ±7.4	0.5
ML	56.7 ±5.9	56.08 ±8.2	0.6

Tab. II. Results of Mann-Whitney test for sexual dimorphism of *Tamandua tetradactyla* Linnaeus, 1758 skull measurements (in mm) mean and standard deviation (±) of specimens classified as level 0 (P, test significance; *n*, sample size; FMD, foramen magnum diameter; OCB, occipital condyle breadth; FH, foramen magnum height; CH, cranial height; CB, cranial breadth; OL, orbital length; NB, nasal breadth; ZB, posterior zygomatic arch breadth; PtB, pterygoids breadth; SOL, supraoccipital length; PL, parietal length; INB, inner nostril breadth; NL, nasal length; PW, palatal width; NC, neurocranium length; FL, frontal length; SL, skull length; ML, maxilla length).

Measurements	Suture closure level 0		P
	Female (*n* = 33)	Male (*n* = 8)	
FMD	12.83 ±2.3	12.4 ±1.4	0.2
OCB	22.3 ±2.23	22.7 ±1.4	1
FH	10.6 ±0.7	11.3 ±0.9	0.1
CH	29.9 ±2.9	31.2 ±1.2	0.1
CB	38.3 ±3.6	39.4 ±1.59	0.4
OL	28.9 ±4.3	30.3 ±3.18	0.4
NB	7.1 ±1.2	7.1 ±0.7	0.9
ZB	37.5 ±3.8	38.5 ±2.4	0.3
PtB	25.8 ±3.6	27.8 ±2.1	0.1
SOL	14.8 ±2.2	14.8 ±1.46	0.8
PL	20.4 ±4.8	20.8 ±1.9	0.4
INB	8.9 ±1	9.12 ±0.6	0.4
NL	33.8 ±8.8	37.3 ±4.6	0.2
PW	18.8 ±3.1	19.4 ±2.17	0.8
NC	76 ±10	81.5 ±7.1	0.1
FL	41.66 ±7.1	46.8 ±6.3	0.04
SL	106.9 ±17.2	117.2 ±9.7	0.1
ML	45.7 ±9.8	50.9 ±5.2	0.1

Tab. III. Results of Mann-Whitney test of *Tamandua tetradactyla* skull measurements (in mm) mean and standard deviation (±) (P, test significance; *n*, sample size; FMD, foramen magnum diameter; OCB, occipital condyle breadth; FH, foramen magnum height; CH, cranial height; CB, cranial breadth; OL, orbital length; NB, nasal breadth; ZB, posterior zygomatic arch breadth; PtB, pterygoids breadth; SOL, supraoccipital length; PL, parietal length; INB, inner nostril breadth; NL, nasal length; PW, palatal width; NC, neurocranium length; FL, frontal length; SL, skull length; ML, maxilla length). Two asterisk (**) denotes P value ≤ 0.001. See figure 1A and 1B for the skull measurement acronyms and definitions.

Measure-ments	Samples		P
	Level 0 (*n* = 53)	Level 1 (*n* = 125)	
FMD	12.58 ±2.0	12.94 ±1.20	0.015
OCB	21.84 ±3.27	23.86 ±1.71	(**)
FH	10.70 ±0.93	11.11 ±3.98	0.838
CH	30.15 ±2.75	32.44 ±1.68	(**)
CB	38.38 ±3.46	41.73 ±2.34	(**)
OL	29.08 ±4.17	32.77 ±2.6	(**)
NB	7.17 ±1.08	7.83 ±1.44	0.013
ZB	37.42 ±3.8	42.46 ±2.5	(**)
PtB	25.64 ±3.9	28.39 ±2.34	(**)
SOL	14.95 ±2.1	16.15 ±1.71	(**)
PL	20.29 ±3.9	22.01 ±5.4	(**)
INB	9.01 ±0.95	9.76 ±1.05	(**)
NL	33.72 ±8.18	43.75 ±4.5	(**)
PW	18.94 ±3.2	21.07 ±3.3	(**)
NC	76.53 ±9.6	86.61 ±6.0	(**)
FL	42.37 ±7.0	50.55 ±5.7	(**)
SL	108.33 ±16.6	128.44 ±7.3	(**)
ML	45.78 ±9.6	56.55 ±6.3	(**)

The principal components analysis showed two separated groups by the first principal component (PC1) (Fig. 2). The first principal component (PC1) explained 42.32% of the variability observed, and all variables contributed positively. However, only four variables had a coefficient loading (lo) higher than 0.25 and were assumed as important for the observed ordination: maxilla length (lo = 0.41), skull length (lo = 0.65), frontal length (lo = 0.39) and neurocranium (lo = 0.48). The second principal component explained 12.37% of the variation observed, and the following measurements contributed to ordinate specimens: frontal length (lo = 0.31), Neurocranium (lo = 0.33), and palatal width (lo = 0.52). The discriminant/hotelling t^2 test (t^2 = 412.47, F = 19.2, $p < 0.0001$), performed on the above variables, indicated two distinct groups. The NPMANOVA test showed significant differences (F = 12.02, $p < 0.01$) between immature and sub-adult skull measurements.

Myrmecophaga tridactyla. Exhibited similar results to *T. tetradactyla*, although all three suture closure levels were analyzed. The three collections consulted had a small number of skulls and many of them were broken. In the LAMORA (n = 13), specimens were from roadkill. In this case, measurements were from undamaged bones. As in the case of *T. tetradactyla*, *M. tridactyla* skulls classified as level 1 and level 2 had larger measurements than those of level 0, except for the foramen magnum height (Tab. VI). The ANOVA test showed a significant difference (F = 7.2, $p \leq 0.05$) among the three categories of suture closure level. However, the pairwise

Tab. IV. Result of Mann-Whitney test for frontal length (FL) measurement of *Tamandua tetradactyla* Linnaeus, 1758 among females classified as level 0 and level 1. Frontal length (FL) (in mm) mean and standard deviation (±) (P, test significance; *n*, sample size).

Measurement	Level 0 female (*n* = 32)	Level 1 female (*n* = 53)	P
FL	41.66 ±7.1	50.7 ±6.9	>0.001

Tab. V. Result of Mann-Whitney test for frontal length (FL) measurement of *Tamandua tetradactyla* Linnaeus, 1758 among males classified as level 0 and level 1. Frontal length (FL) (in mm) mean and standard deviation (±) (P, test significance; *n*, sample size).

Measurement	Level 0 male (*n* = 8)	Level 1 male (*n* = 28)	P
FL	46.8 ±6.3	50.55 ±4.2	0.04

Tukey test (Tab. VII) revealed no significant differences ($p > 0.05$) between level 1 and level 2 samples.

The principal components analysis separated the three suture closure categories into two groups: the level 0 samples in one group and level 1 and level 2 in another. The first principal component explained 61.01% of the observed variation and the second principal component explained 12.94%. All 18 variables contributed positively to the first principal component, and the following measurements, which most contributed to the ordination, demonstrated coefficient loadings (lo) higher than 0.20: maxilla length (lo = 0.50), neurocranium (lo = 0.47), frontal length (lo = 0.23), nasal

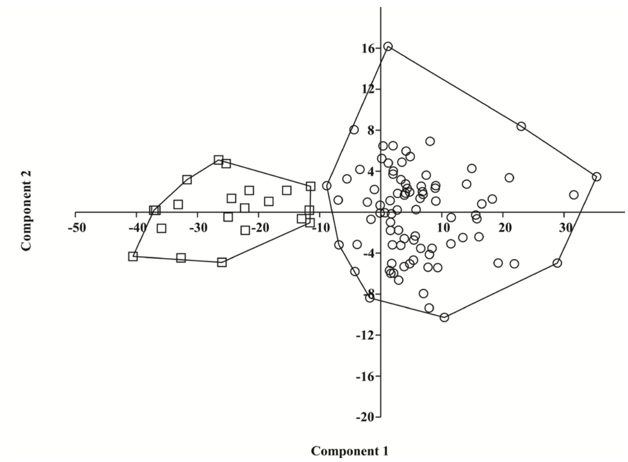

Fig. 2. Results of *Tamandua tetradactyla* Linnaeus, 1758 principal component analysis: plot of the scores of principal component 1 and 2 extracted from variance-covariance matrix of all skull measurements of samples classified as suture closure level 0 (square) and level 1(circle).

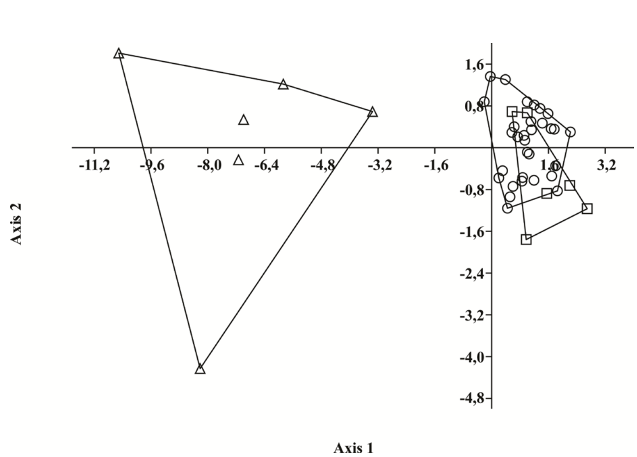

Fig. 3. Results of *MANOVA/CVC* test of *Myrmecophaga tridactyla* Linnaeus, 1758 skull measurements. Samples classified as suture closure level 0 (triangle), level 1 (circle) and level 2 (square).

Tab. VI. Results of ANOVA test of *Myrmecophaga tridactyla* Linnaeus, 1758 skull measurements (in mm) mean and standard deviation (±) (P, test significance; *n*, sample size; FMD, foramen magnum diameter; OCB, occipital condyle breadth; FH, foramen magnum height; CH, cranial height; CB, cranial breadth; OL, orbital length; NB, nasal breadth; ZB, posterior zygomatic arch breadth; PtB, pterygoids breadth; SOL, supraoccipital length; PL, parietal length; INB, inner nostril breadth; NL, nasal length; PW, palatal width; NC, neurocranium length; FL, frontal length; SL, skull length; ML, maxilla length). Two asterisk (**) denotes P value ≤ 0.001.

Measurements	Samples			P
	Level 0 (*n* = 7)	Level 1 (*n* = 37)	Level 2 (*n* = 6)	
FMD	15.68±0.5	18.55±1.5	19.16±2.0	(**)
OCB	35.61±2.3	40.47±2.4	41.93±1.0	(**)
FH	15.93±1.2	19.04±4.5	18.31±1.5	0.226
CH	42.18±4.2	51.30±2.4	52.46±2.4	(**)
CB	53.46±3.7	62.25±2.0	63.79±1.7	(**)
OL	42.92±7.1	59.06±7.1	62.13±6.1	(**)
NB	10.64±0.4	14.60±1.3	14.94±1.1	(**)
ZB	51.46±5.6	65.57±3.6	68.97±3.3	(**)
PtB	33.82±3.7	40.72±2.3	40.74±1.4	(**)
SOL	21.52±4.6	28.71±2.9	28.85±2.4	(**)
PL	23.90±2.3	29.50±4.3	31.38±3.0	0.002
INB	15.50±2.1	17.96±1.3	18.59±1.3	(**)
NL	89.83±21.0	170.78±10.5	174.58±10.4	(**)
PW	15.47±2.5	19.37±1.8	19.38±1.1	(**)
NC	137.63±24.01	184.61±15.8	191.91±17.3	(**)
FL	79.63±20	110.62±21.7	116.18±5.9	0.001
SL	248.4±58	348.05±17.5	362±28.5	0.001
ML	118±31.7	214.9±16.5	220±17.5	(**)

Tab. VII. Results of Tukey Pairwise test of *Myrmecophaga tridactyla* Linnaeus, 1758 skull measurements among samples classified as immature (level 0), sub-adult (level 1) and adult (level 2) (X, pairwise between level 0 and level 1; Y, pairwise between level 0 and level 2; Z, pairwise between level 1 and level 2; FMD, foramen magnum diameter; OCB, occipital condyle breadth; FH, foramen magnum height; CH, cranial height; CB, cranial breadth; OL, orbital length; NB, nasal breadth; ZB, posterior zygomatic arch breadth; PtB, pterygoids breadth; SOL, supraoccipital length; PL, parietal length; INB, inner nostril breadth; NL, nasal length; PW, palatal width; NC, neurocranium length; FL, frontal length; SL, skull length; ML, maxilla length).

Measurements	X	Y	Z
FMD	0.001	0.00018	0.69
OCB	0.00036	0.00013	0.40
FH	0.269	0.458	0.92
CH	0.00012	0.00012	0.64
CB	0.00012	0.00012	0.35
OL	0.00016	0.00012	0.63
NB	0.00012	0.00012	0.86
ZB	0.00012	0.00012	0.18
PtB	0.00012	0.00012	1
SOL	0.00018	0.00018	0.99
PL	0.01406	0.00094	0.58
INB	0.03365	0.00032	0.65
NL	0.00012	0.00012	0.81
PW	0.00036	0.00036	1
NC	0.00012	0.00011	0.66
FL	0.00716	0.00147	0.83
SL	0.00012	0.00012	0.85
ML	0.00012	0.00012	0.67

length (lo = 0.36) and skull length (lo = 0.55). For the second principal component, the neurocranium (NC) was the only variable that contributed substantially (lo = 0.70), and the other variables demonstrated loadings lower than 0.20. Skull length (lo = -0.45) and maxilla length (lo = -0.26) contributed most negatively. The MANOVA/CVC test ($F = 5.78$; $p < 0.000001$) also indicated two groups: level 0 in one group, and level 1 and level 2 in another (Fig. 3). The same results were observed with the NPMANOVA test ($p < 0.01$), which showed significant differences between level 0 and level 1 ($p < 0.001$), level 0 and level 2 ($p < 0.001$), but not between level 1 and level 2 ($p > 0.05$, $p = 0.38$).

The ANOVA test of morphometric data showed that, unlike the other 17 dimensions measured, the foramen magnum height of *M. tridactyla* did not differ ($p > 0.05$) among the three categories analyzed. Level 0 samples had mean ($\bar{X}$) and standard deviation (±) of foramen magnum height ($\bar{X}$= x15.93 ± 1.2 mm) statistically similar to the level 1 ($\bar{X}$= 19.04 ± 4.5 mm) and level 2 ($\bar{X}$= 18.31 ± 1.5 mm). On the other hand, the foramen magnum diameter, a horizontal measurement, showed significant differences between level 0 and level 1 ($p = 0.001$) and between level 0 and level 2 ($p = 0.00018$).

A Mann-Whitney test of *Tamandua tetradactyla* data showed similar results: there was no significant difference ($p > 0.05$) on foramen magnum height between level 0 ($\bar{X}$= 10.7 ± 0.9 mm) and level 1 ($\bar{X}$= 11.1 ± 3.9 mm). However, for the foramen magnum diameter, there was a significant difference ($p = 0.01$). Therefore, the results indicated that *M. tridactyla* and *T. tetradactyla* level 0 specimens have a foramen magnum height of a similar approximate size as the level 1 and level 2, but not diameter.

The results of suture closure level analyses also indicated that the interfrontalis suture of *M. tridactyla* and *T. tetradactyla* is the first suture of the cranial vault to be closed and the lambdoidea the last one. The sagitalis suture is the second and coronalis the third to be closed in *M. tridactyla* skulls, but it was not possible to affirm the same for *T. tetradactyla* skulls.

DISCUSSION

Based in our results of the foramen magnum height (FH) between specimens classified as level 0, level 1 and level 2 we believe that both *M. tridactyla* and *T. tetradactyla* specimens with small skull dimensions, which we consider to be young, present FH sizes similar to those with larger measurements (level 1 and 2) which are possibly older. The no significant differences of FH between the three suture closures may be a consequence of the early development of the medulla oblongata in anteaters, based on the assumption that is through the foramen magnum that passes the medulla oblongata. Which is a part of the encephalon continues with the spinal cord, vertebral arteries and nerves (TORTORA, 2000).

The specimens classified as level 1 and level 2 of *M. tridactyla* the presented no significant differences in the sizes of skulls. It is possible that the same occurs for *T. tetradactyla.* However, a small sample size of skulls classed as level 2 (n = 2) made further comparisons of skull size inadequate. ELBROCH (2006) reported that almost all sutures are visible in immature vertebrates, and as age advances, sutures gets so closed that they become hardly visible. Thus, suture closure level may be assumed to be a useful tool to estimate mammal age, but in the present paper we can only affirms that the cranial vault sutures observed have relations to the skull dimensions.

According to some studies, sutures are bone growth sites stimulated by the expansion of the brain (BAER, 1954; MOSS, 1954). As the brain expands, an intramembranous ossification process occurs in the suture region, particularly in the front edges of skull bones, to accommodate the largest brain (OPPERMAN, 2000). Therefore, it is possible that as the brain expansion of *M. tridactyla* decreases or ceases, bones of the vault and other skull bones also reduce or stop growth, and then the sutures become permanently closed. With the decline of bone growth, there will be a few bone size differences between animals classified as level 1 and level 2, since the animals classified as level 1 have one (interfrontalis) to three cranial vault sutures closed.

KEY *et al.* (1994) tested three methods to determine the age of death in humans by examining the degree of obliteration of the sutures of the cranial vault. They found that age estimation by sutures is only valid to infer an age group and not the precise age of death. In this study, similar results were obtained, since the animals classified as level 0 had smaller cranial measurements than those classified as level 1 and 2. It is known that immature individuals of both giant and lesser anteaters have smaller body dimensions than sub-adults and adults (NOWAK, 1999; RICHARD-HANSEN *et al.*, 1999; HAYSSEN, 2011). Then, the age classification by assessing the level of obliteration of the sutures may be a valid technique to estimate an age group for these species. However, it should be noted that we used specimens with unknown ages and therefore the results indicate only the relation between the sutures closed and skull dimensions and not the precise age of death.

Age analysis by suture closure level has been used in other studies that focused on the mammal skull (SCHWEIKHER, 1930; CHEVERUD, 1996; BRUNNER *et al.*, 2004; WESTGATE, 2007). In these studies, a classification technique using suture closure level was used, similar to that used in this study. Therefore, the present study indicates that the analysis of ectocranial suture closure level of the skull vault of *M. tridactyla* and *T. tetradactyla* has relationships with the dimensions of the skull. Open sutures were highly present in skulls with smaller dimensions, while larger skulls often had at least one or three closed sutures. Importantly, the foramen magnum height (FH) of *M. tridactyla* and *T. tetradactyla* was not different in size among level 0, level 1 and level 2 samples. Based on our results and considering that anteaters are toothless, we concluded that the analysis of cranial vault suture closure level have relation to skull dimensions and could be a potential tool if associated with other technics. Furthermore, this could contribute to further studies on taxonomy, sexual dimorphism and phylogeny in which skull age influence is important.

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