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1 **Paleoecological inferences from long bone microanatomical specializations**
2 **in Hippopotamoidea (Mammalia, Artiodactyla)**

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17 RH: Hippopotamoid long bone microanatomical specializations

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1 **Abstract**

2 Hippopotamoids are herbivorous mammals that originated in the late middle Eocene. This
3 taxon includes animals with a great variety of sizes and body proportions, from small and
4 gracile forms with slender limbs to heavy massive ones. Many hippopotamoids have
5 previously been considered semi-aquatic but recent studies have highlighted a diversity of
6 ecologies. This study focuses on bone microanatomy, one of the various proxies that enable
7 inferring the ecology of extinct taxa. The comparative analysis of the inner structure of the
8 stylopod bones in various hippopotamoids, based on both transverse and longitudinal virtual
9 sections, highlights a diversity of patterns and clarifies previously proposed hypotheses about
10 the ecology of the sampled hippopotamoids. The filling of the medullary area by spongy
11 deposits in the pygmy hippopotamus, *Choeropsis liberiensis*, appears associated with frequent
12 incursions into the water by an animal that essentially forages in forests. The common
13 hippopotamus, *Hippopotamus amphibius*, which spends most of the day submerged in water,
14 shows a greater filling of the medullary area by spongy bone and a thicker cortex. These
15 observations coupled with comparisons with diverse terrestrial and semi-aquatic mammals of
16 various sizes confirm that semi-aquatic lifestyle and heavy weight-bearing are associated with
17 similar microanatomical specializations causing an increase in bone mass. However, for a
18 given mass, comparisons enable determining if an additional increase in bone compactness
19 occurs, as in *Hippopotamus amphibius*, in which case a semi-aquatic lifestyle could be
20 inferred. Accordingly, this study suggests an essentially terrestrial lifestyle for *Microbunodon*
21 *minimum*, *Bothriodon velaunus*, *Elomeryx borbonicus*, *Merycopotamus medioximus*,
22 *Paenanthracotherium bergeri*, and probably also *Saotherium* cf. *S. mingoz*, a slight degree of
23 water dependence in *Brachyodus onoideus*, and a stronger one in *Libycosaurus bahri* and
24 *Hexaprotodon garyam*, though less intense than in *Hippopotamus amphibius*. Comparisons
25 with other large terrestrial and semi-aquatic taxa, and based on a large part of the diaphysis,
26 are required to better decipher the microanatomical changes associated with a semi-aquatic
27 lifestyle from those linked to loading in heavy quadrupedal mammals.

28
29 **Keywords:** Anthracotheriinae, bone microanatomy, Bothriodontinae, Hippopotamoidea,
30 Microbunodontinae, semi-aquatic lifestyle, weight-bearing.

1 **Introduction**

2 Hippopotamoids, including hippopotamids and anthracotheres, are herbivorous mammals that
3 emerged in the late middle Eocene (Soe et al. 2017). This clade is assumed to correspond to
4 the sister group of Cetacea based on both molecular (Irwin and Arnason 1994; Gatesy et al.
5 1996; Gatesy 1997; Montgelard et al. 1997; Ursing and Arnason 1998; Nikaido et al. 1999,
6 Arnason et al. 2002; Zhou et al. 2011) and morphological (Geisler and Uhen 2003; Boisserie
7 et al. 2005a; Geisler et al 2007; Gatesy et al. 2013) studies. Its abundant fossil record
8 illustrates a wide spatio-temporal distribution. Among hippopotamoids, anthracotheres lived
9 from the late middle Eocene to the early Pleistocene in Eurasia, northern America, and Africa
10 (Kron and Manning 1998; Dettel 2005; Lihoreau and Ducrocq 2007; Holroyd et al. 2010;
11 Tsubamoto 2010; Rincon et al. 2013; Lihoreau et al. 2016; Grandi and Bona 2017; Soe et al.
12 2017; Scherler et al. 2019; Lihoreau et al. 2019; Grossman et al. 2019). Anthracotheres are
13 assumed to correspond to stem Hippopotamidae (Boisserie et al. 2005a, 2010; Boisserie and
14 Lihoreau 2006; Orliac et al. 2010; Lihoreau et al. 2015). Currently 27 genera are recognized
15 and distributed in three subfamilies: the Anthracotheriinae, the Microbunodontinae, and the
16 Bothriodontinae (Lihoreau and Ducrocq 2007), including animals with a great variety of
17 sizes, from small (< 25 kg) to medium and large (> 1,000 kg) forms. Anthracotheriinae and
18 Bothriodontinae were medium to very large-sized animals, whereas Microbunodontinae were
19 exclusively small-sized anthracotheres with slender limbs. Anthracotheres have been
20 previously considered as essentially semi-aquatic hippo-like forms (e.g., Falconer and Cautley
21 1836; Rüttimeyer 1857). More recent studies have highlighted their great diversity of habitats
22 and diets (Lihoreau 2003) and that anthracotheres can be used as ecological markers to
23 reconstruct the paleoenvironmental history of the landmasses they occupied (Lihoreau and
24 Ducrocq 2007). Hippopotamids are first known from the early Miocene in Africa, and then
25 spread to Asia and Europe (Boisserie 2007; Orliac et al. 2010). These small-sized (~30 kg for
26 *Morotochoerus*; Orliac et al. 2010) to very large (mean weight of 1500 kg for the extant genus
27 *Hippopotamus*; Klingel 2013) animals are assumed to have adopted semi-aquatic habits in the
28 latest Miocene (Boisserie 2007; Boisserie et al. 2011).

29
30 Semi-aquatic forms occurred independently in various hippopotamoid lineages, so that
31 the question of the habitat of numerous fossil taxa, and notably their degree of water
32 dependence, remains unresolved (Lihoreau and Ducrocq 2007; Boisserie et al. 2011; Lihoreau
33 et al. 2014; Grandi and Bona 2017) and cannot be answered by sedimentological and

1 morphological data only. For this purpose, various approaches can be added, such as studies
2 on sensorial abilities (e.g., Orliac et al. 2014), on dental microwear and mesowear (e.g.,
3 Lihoreau 2003; Merceron et al. 2010; Boisserie and Merceron 2011; Lihoreau et al. 2014),
4 and isotopic investigations (e.g., Boisserie et al. 2005a; Nelson 2007; Clementz et al. 2008;
5 Lihoreau et al. 2014; Tütken and Absolon 2015). Morphofunctional studies on the postcranial
6 skeleton are also required. However, to date, such data relative to hippopotamoids are
7 extremely limited. They consist of limited studies on the skeleton of *Hippopotamus*
8 *amphibius* (Gratiolet 1867) and fossil taxa (Kowalesky 1873; Scott and Jepsen 1940; Dineur
9 1981; Pickford 2008), and on detailed descriptions of the forelimb and hind limb myology of
10 *Choeropsis liberiensis* and *H. amphibius*, respectively (Fisher et al. 2007; 2010). In addition,
11 there are data on swimming and aquatic bottom-walking abilities in *H. amphibius* (Coughlin
12 et al. 2009; Endo et al. 2019).

13 Beyond studies on bone shape and muscle insertions, the analysis of bone
14 microanatomical adaptations can also be very useful for paleobiological inferences. Indeed,
15 bone inner structure reflects the functional constraints imposed on the skeleton and thus the
16 functional requirements of the organism (e.g., Ruff 1992; Ruimerman et al. 2005; Volpato et
17 al. 2008; Mielke et al. 2018). The form-function relationships of bone microanatomy have
18 been widely used in the context of adaptation to an aquatic lifestyle, focusing on both aquatic
19 and semi-aquatic forms (e.g., Canoville and Laurin 2010; Dumont et al. 2013; Amson et al.
20 2014; Diaz-Berenguer et al. 2019; Klein et al. 2019). Yet, hippopotamoids also include large
21 quadrupedal taxa. Adaptations to heavy weight bearing are also observed in the bone
22 microanatomy of large quadrupeds (Wall, 1983; Houssaye et al. 2016a). However, they can
23 be hard to decipher from those linked to a semi-aquatic lifestyle, as both are characterized by
24 an increase in bone compactness (Cooper et al. 2016; Houssaye et al. 2016b). Indeed, both
25 semi-aquatic taxa swimming or walking on the bottom at shallow depths and heavy terrestrial
26 quadrupeds generally exhibit an increased compactness characterized by a more or less
27 extensive thickening of the cortex and filling of the medullary cavity by cancellous bone
28 (Nakajima and Endo 2013; Houssaye et al. 2016a,c; Houssaye and Botton-Divet 2018). The
29 increased bone mass in semi-aquatic taxa is assumed to hydrostatically counter buoyancy to
30 dive and stay at shallow depths but also to counter waves to improve stability in rough waters
31 (Wall 1983; Taylor 2000). Conversely, in heavy animals, such a structure is believed to
32 increase bone strength and improve the absorption of impact energy (Oxnard 1990; Augat and
33 Schorlemmer 2006). Investigations on phylogenetically close taxa showing light and massive
34 terrestrial and semi-aquatic forms thus constitute a great opportunity to better characterize

1 these microanatomical specializations that have convergently evolved multiple times in heavy
2 and/or semi-aquatic (and aquatic) amniotes (Houssaye et al. 2016a,c). Only limited
3 microanatomical data are available for hippopotamoids beyond the mid-diaphyseal sections of
4 modern hippopotamids (Cooper et al. 2016; Houssaye et al. 2016a). Cooper et al. (2016)
5 investigated the microanatomical features of three extinct hippopotamoid species from two
6 genera, the microbunodontine *Microbunodon silistrense* and the bothriodontines
7 *Merycopotamus dissimilis* and *Merycopotamus medioximus*, in order to infer their
8 paleoecology, suggesting a terrestrial lifestyle for *Mi. silistrense* and a semi-aquatic one for
9 the two *Merycopotamus* species.

10
11 The objectives of the present study are to investigate the microanatomical adaptive
12 features of hippopotamoid bones by analyzing the inner structure of limb long bones of
13 diverse hippopotamoids supposed to illustrate various degrees of water dependency, taking
14 into account their body proportions, and to discuss inferences related to the paleoecology of
15 the sampled fossil forms.

16 It is predicted that 1) the bones of terrestrial forms will show a tubular organization,
17 i.e., a compact cortex surrounding an empty medullary cavity, typical of terrestrial amniotes
18 (Laurin et al. 2011; Houssaye et al. 2018), 2) the bones will be more compact in the largest
19 terrestrial species, with a thicker cortex and some trabeculae in the medullary area, 3) the
20 species most dependent on the aquatic environment will also show more compact bones, and
21 4) the species that are both large and highly dependent on the aquatic environment will be
22 much more compact.

23 24 25 **Material and Methods**

26 **Material**

27 The material examined consists of the humeri and femora of various hippopotamoids.

28 Stylopod bones are assumed to bear a strong morphofunctional signal in their microanatomy
29 (e.g., Quemeneur et al. 2013). They were therefore selected in order to investigate the long
30 bone microanatomical adaptations related to the posture and lifestyle of these extinct taxa.

31 Moreover, most of the comparative data for ungulates are available only for these bones.

32 Fossil material includes the bones of four hippopotamoid subfamilies (Tables 1, 2).

1 *Microbunodon minimum* (Cuvier 1822) is a microbunodontine from the late Oligocene
2 of Europe. The sampled specimens are from the collections from La Milloque, France
3 (Lihoreau et al. 2004). This small-sized species (around 20 kg) is lightly built with long and
4 slender limbs, the hind limb being longer and more robust than the forelimb (Cabard 1976;
5 Lihoreau 2003; Lihoreau et al. 2004; Lihoreau and Ducrocq 2007). *Microbunodon* is assumed
6 to be terrestrial and to have lived in closed habitats (dense forest) based on its morphology
7 (e.g., low orbits, no specialization for subaquatic hearing, autopod showing a digitigrade
8 stance, weakly elongated metapodials; Cabard 1976; Boisserie 2002; Lihoreau 2003;
9 Lihoreau and Ducrocq 2007), which is corroborated at the generic level by micro- and
10 mesowear data, and isotopic studies (Lihoreau 2003; Lihoreau and Ducrocq 2007; Nelson
11 2007).

12 The sampled anthracotheriine is *Paenanthracotherium bergeri*, from the Oligocene of
13 La Bénissons-Dieu, France. It is large (< 950 kg; estimation based on talus measurement
14 following Martinez and Sudre [1995]) and heavily-built with short and robust limb bones
15 (Roman and Boucher 1936) and may have lived in swamps as was historically supposed for
16 the closely related *Anthracotherium* (Cuvier 1822). However, the low position of its orbits on
17 the skull (see plates in Roman and Boucher 1936; Scherler et al. 2019), though not
18 incompatible with a semi-aquatic lifestyle, does not indicate a hippo-like lifestyle at the
19 interface between air and water.

20 Within Bothriodontinae, *Bothriodon velaunus*, from the early Oligocene of Ronzon,
21 France (Lihoreau and Ducrocq 2007) is a medium sized animal (< 150 kg; estimation based
22 on talus measurement following Martinez and Sudre [1995]) displaying a lightly-built
23 skeleton with gracile legs (Filhol 1881; Lihoreau and Ducrocq 2007). Though little is known
24 about its ecology, its diet is supposed to have been predominantly folivorous, based on cusp
25 morphology (Lihoreau and Ducrocq 2007). It is similar to *Elomeryx borbonicus* (<130 kg;
26 estimated from Martinez and Sudre [1995]), from the late Oligocene of Saint-Henri/Saint-
27 André, Marseille, France (Lihoreau and Ducrocq 2007). The hind limbs of *Elomeryx*
28 *borbonicus* are longer and more robust than its forelimbs (Geais 1934). If the proposed
29 occurrence of webbed feet (Geais 1934) suggests a semi-aquatic lifestyle, supposedly in
30 marshy habitats, the position of the superior border of the orbit and the morphology of the
31 tympanic bulla and of the auditory tube are typical of terrestrial animals (Lihoreau 2003).
32 *Libycosaurus bahri*, from the late Miocene of Toros-Ménalla, Chad (Vignaud et al. 2002),
33 displays a heavy (< 1,600 kg) hippo-like morphology with short and stocky limbs, although
34 being more elongated and slender than those of the common hippopotamus (Lihoreau et al.

1 2014). This sexually body-mass-dimorphic taxon appears to be semi-aquatic, based among
2 others on its highly elevated orbits and external nares, the morphology of the tympanic bulla,
3 and on isotopic studies (Lihoreau et al. 2014, 2006). Finally, *Brachyodus onoideus*, from the
4 early Miocene of Nancray and Neuville, near Orléans, France (Lihoreau and Ducrocq 2007),
5 is much larger (< 2,350 kg; estimate based on talus measurement following Martinez and
6 Sudre [1995]). This large-sized species displays strong limb bones albeit being more
7 elongated and slender than those of the common hippo (Dineur 1981). A terrestrial mode of
8 life has been suggested for this species based on the conformation of its limbs (Dineur 1981)
9 but the amphibious position of the sensory organs on the head along with the morphology of
10 the tympanic bulla potentially enabling underwater directional hearing rather suggest
11 hydrophilic and water immersive habits (Orliac et al. 2013). Trackways from the lower
12 Miocene of Spain possibly attributable to this species seem to indicate a gregarious behavior
13 close to that of hippos with trails in palustrine environments (Diaz Martinez et al. 2020).

14 Within hippopotamines, *Saotherium* cf. *S. mingozi* (Boisserie et al. 2003), from the
15 early Pliocene of Kossom Bougoudi, Chad (Boisserie 2007; Weston and Boisserie 2010), is of
16 intermediate size (<1,000 kg; estimated from Martinez and Sudre [1995]) between the extant
17 *C. liberiensis* (< 275 kg) and *H. amphibius* (< 2,000 kg). It does not show adaptations to a
18 semi-aquatic lifestyle (Boisserie et al. 2003; Boisserie 2005, 2007; Weston and Boisserie
19 2010) but isotopic data suggest water dependence (Zazzo et al. 2000). As for *Hexaprotodon*
20 *garyam*, from the late Miocene of Toros-Ménalla, Chad (Boisserie 2007; Vignaud et al. 2002;
21 Weston and Boisserie 2010), weighting up to 2,200 kg (Lihoreau et al. 2014; estimated from
22 Martinez and Sudre [1995]), it displays a set of morphological (Boisserie 2002) and isotopic
23 features (Jacques 2007) that might indicate a semi-aquatic lifestyle (Boisserie 2002; Boisserie
24 et al. 2005b; Lihoreau et al. 2014). The extant hippopotamine, *C. liberiensis*, about six times
25 lighter than *H. amphibius*, is more gracile with slender elongated limbs (Eltringham 1999;
26 Nowak and Paradiso 1983; Robinson, 2013), whereas *H. amphibius* displays a massive barrel-
27 shaped body supported by a heavily-built skeleton with short and stocky limbs (Eltringham
28 1999; Klingel 2013; Nowak and Paradiso 1983). Common hippopotamuses inhabit wetlands
29 with permanent water bodies deep enough to enable immersion, such as lakes, rivers, pools,
30 swamps, marshes, and ponds, in the vicinity of reed beds and grasslands (Eltringham 1999;
31 Klingel 2013; Nowak and Paradiso 1983). *Choeropsis* inhabits wetter, closer, and denser
32 environments than the common hippopotamus, which is consistent with its less webbed toes
33 and its nocturnal habits. It lives close to streams, rivers, or swamps, and can dive, thus
34 showing a lifestyle close to that of extant forest tapirs (Eltringham 1999; Nowak and Paradiso

1 1983; Robinson 2013), despite additional adaptations for a semi-aquatic lifestyle, such as the
2 ability to occlude ears and nostrils when diving.

3
4 The comparative material includes terrestrial and semi-aquatic ungulates covering and
5 extending the possible ecologies as well as the size range of the hippopotamoid sample (see
6 Table 1; Fig. 1). It includes the basal cetancodont *Indohyus indirae* and archaeocetes belonging
7 to the Pakicetidae, Remingtonocetidae, and Protocetidae, in order to illustrate various
8 ecological steps in the progressive adaptation to an aquatic lifestyle in the cetacean lineage. It
9 also includes small to medium-sized even-toed ungulates with long and slender legs (e.g.,
10 *Hyemoschus aquaticus*, *Potamochoerus porcus*), but also medium- to large-sized even-toed and
11 odd-toed ungulates, from gracile (e.g., *Giraffa Camelopardalis*, *Alces alces*) to heavily-built
12 (e.g., *Syncerus caffer*, *Rhinoceros unicornis*) animals (Table 1).

13 14 **Methods**

15 *Data acquisition*

16 Bones were scanned using high-resolution computed tomography at: 1) the IC2MP,
17 University of Poitiers (UMR 7285; EasyTom XL duo, RX Solutions), 2) the ISEM,
18 University of Montpellier (UMR 5554; EasyTom 150, RX Solutions) 3) the Steinmann-
19 Institut, University of Bonn (Germany; GE phoenix|X-ray v|tome|xs 240), 4) the AST-RX
20 platform of the Muséum National d'Histoire Naturelle, Paris (UMS 2700; GE phoenix|X-ray
21 v|tome|xs 240), and 5) the Equine Diagnostic Imaging Centre at the Royal Veterinary
22 College, London (GE Lightspeed), with reconstructions performed using X-Act (RX
23 Solutions), DATOX/RES, phoenix datos|x or MEDVIEW software (MedImage). Voxel size
24 naturally varies pending on specimen size (Table 1).

25 Transverse virtual thin-sections were made for each bone at the plane assumed to cross
26 the growth center (where the cortex is the thickest in the case of a non-uniform thickness, see
27 Houssaye et al. 2018, and at the intersection of the nutrient arteries the central axis of the
28 medullary cavity, see Houssaye and PrévotEAU 2020). Sagittal and coronal virtual sections
29 were made at the planes crossing the core of the transverse section. Image visualization and
30 virtual sections were performed using VGSTUDIOMAX, versions 2.2 (Volume Graphics
31 Inc.).

32 33 *Data analysis*

1 Quantitative parameters were used to describe the transverse sections, except those of extinct
2 taxa showing an insufficient contrast between bone and the infilling sediment (see Table 1).
3 The choice of parameters follows Houssaye et al. (2018), using the BoneJ plugin (Doube et al.
4 2010) of ImageJ (Wayne Rasband National Institutes of Health, USA) and the software Bone
5 Profiler (Girondot and Laurin, 2003): A) CSA_b (Cross-Sectional Area of bone), representing
6 the surface occupied by bony tissue; B) CSA_t (total Cross-Sectional Area), as the surface of
7 the whole transverse section; C) C (Compactness), as the ratio of CSA_b over CSA_t; D) R, as
8 the radius of the section approximated as a circle calculated based on the perimeter/ 2π , and
9 used as a proxy of size; E) CSS (Cross-Sectional Shape), as the ratio between the maximal
10 and minimal second moments of area (I_{max}/I_{min}); F) Z_{pol} (Polar Section Modulus)
11 representing the resistance of a section to torsion and bending (see Ruff 2002; Ksepka et al.
12 2015); G) R_{MeanT}, as the relative mean thickness of cortical bone (after separation by
13 segmentation of the cortical and medullary areas), calculated as the absolute mean thickness
14 of cortical bone divided by R; H) R_{SDT}, as the relative standard deviation of cortical bone
15 thickness; I) P: the extent of the medullary cavity as measured by the relative distance from
16 the center of the section to the point where the most abrupt change in compactness occurs;
17 and J) S: the width of the transitional zone between the compact cortex and the medullary
18 cavity as measured by the reciprocal of the slope of the compactness profile at the inflection
19 point.

20
21 We tested the influence of size on all parameters by performing linear regressions of
22 each parameter to R (lm function). We comparatively analyzed the values of each parameter
23 for the hippopotamoids separately, all rhinos, the (dominantly) terrestrial and the
24 (dominantly) aquatic ungulates (Table 1; Figs S2; S4).

25 We tested the phylogenetic signal for each parameter on the raw data. For that we
26 averaged the values obtained for each species when several specimens from the same species
27 were available. Then we calculated the K-statistic following Blomberg et al. (2003) for each
28 parameter and performed randomization tests. The K-statistic compares the observed
29 phylogenetic signal in a trait with the signal under a Brownian motion model of trait
30 evolution. A K-value > 1 implies more similarity between relatives than expected under
31 Brownian motion; $K < 1$ highlights convergences.

32 For this testing, we used two different consensus trees (Fig. 1) based on phylogenies
33 from Lihoreau et al. (2015, 2019), Boisserie et al. (2011, 2017), and Gomes Rodrigues (2019)
34 for the relationships within Hippopotamoidea, the proposition of Thewissen et al. (2007) and

1 Vautrin et al. (2020) for the position of *Indohyus* and Cetacea, and recent phylogenetic
2 hypotheses based on molecular data for extant representatives of modern clades of
3 laurasiatherian mammals (Gilbert et al. 2006; Bibi 2013; Foley et al. 2016; Springer et al.
4 2019). These phylogenies differ by the relationships observed within Hippopotamoidea. The
5 first hypothesis links Hippopotamidae to archaic bothriodontines, i.e., the African Paleogene
6 clade including *Bothriogenys* and *Brachyodus*. The second hypothesis proposes the
7 Hippopotamidae as sister-group of the whole Bothriodontinae.

8 In order to analyze quantitatively the distribution of the different specimens in the
9 microanatomical morphospace, but also for the purpose of evaluating how the different
10 microanatomical and cross-sectional parameters explain the variations observable in our
11 sample, we conducted normalized PCAs (David and Jacobs 2014).

12 All statistical analyses were performed using the statistical software R (R Core Team
13 2014).

15 Results

17 Humerus

18 *Qualitative description*

19 Humeral sections of *Choeropsis* do not show a particular thickening of the cortex (Figs. 2A-
20 B, 3A), as compared to most quadrupedal mammals (see Houssaye et al. 2018; Canoville and
21 Laurin 2010; Fig. S1). However, whereas these taxa show a generally tubular diaphysis with a
22 compact cortex surrounding an empty medullary cavity, the humerus medullary space of
23 *Choeropsis* is filled with a spongy tissue. The thickness of the cortex increases distal to the
24 deltoid tuberosity but is then rather homogeneous in the rest of the diaphysis. The
25 organization appears rather similar in *Saotherium* (Figs. 2C, 3B), *Libycosaurus* (Figs. 2J, 3H)
26 and in one specimen of *Hexaprotodon* (TM 55-XX-05; Figs. 2F, 3D), but also in *Brachyodus*
27 (Figs. 2H, 3F), though its spongiosa is poorly preserved. The spongiosa is also poorly
28 preserved in *Bothriodon* whose cortex is thinner than in the other taxa (Figs. 2I, 3G). In
29 *Hippopotamus* the thickness of the cortex varies along the diaphysis distal to the deltoid
30 tuberosity, with a pronounced thickening around the growth center (Figs. 2D-E, 3C). As in
31 *Choeropsis* a spongiosa fills the medullary area. Such a thickening of the cortex is also
32 observed in one specimen of *Hexaprotodon garyam* (TM 258-01-27; Figs. 2G, 3E) and, to a
33 lesser extent, in *Paenanthracotherium* (Figs. 2M, 3K). The spongiosa in the medullary cavity

1 of the later is not preserved or was absent. As for *Elomeryx* and *Microbunodon*, they show a
2 tubular organization (Figs. 2K-L, 3I-J). As for *Merycopotamus*, sections of *Me. medioximus*
3 (Lihoreau et al. 2004) from Cooper et al. (2016) show an organization close to that of
4 *Elomeryx* and *Microbunodon* (Fig. S1A) but that of *Me. dissimilis* (Falconer and Cautley
5 1836) is filled with some trabeculae (Fig. S1B).

6 7 *Quantitative comparisons*

8 Compactness (C) is the highest in *Hippopotamus*. It is higher than in rhinos and in
9 *Hexaprotodon* (TM 258-01-27), which show higher values than the other taxa (even than
10 most aquatic ones; Fig. S2A). *Choeropsis* globally displays a higher compactness than the
11 other terrestrial ungulates, but it is much lower than in rhinos. The other hippopotamoids
12 show compactness values slightly (especially *Brachyodus*) lower than those of *Choeropsis*.
13 The cross-sectional shape (CSS) is strongly higher in aquatic taxa (Fig. S2B); as for the
14 others, it is maximal in the comparative terrestrial taxa and globally much higher in rhinos,
15 *Hippopotamus*, *Microbunodon*, and *Bothriodon* than in *Choeropsis*, *Elomeryx*, *Brachyodus*,
16 and *Hexaprotodon*, which thus present more rounded sections (Fig. S2C). The relative mean
17 cortical thickness (RMeanT) is the highest in *Hippopotamus* and *Hexaprotodon* (TM 258-01-
18 27). It varies strongly within rhinos where it is generally higher than in other terrestrial
19 ungulates. RMeanT is much higher in *Brachyodus* than in *Choeropsis*, *Elomeryx*, *Bothriodon*,
20 *Microbunodon*, and the comparative terrestrial and aquatic taxa (Fig. S2D). The cortical
21 thickness is rather homogeneous in most terrestrial and in aquatic taxa, which show the lowest
22 RSDT values (Fig. S2E). This is also the case for *Microbunodon*, *Elomeryx*, and *Bothriodon*.
23 However it is slightly higher in *Choeropsis* and much higher in *Brachyodus* and even more in
24 *Hippopotamus* and *Hexaprotodon garyam* (TM 258-01-27). RSDT varies strongly among
25 rhinos, from values close to those of *Brachyodus* to values higher to those of *Hippopotamus*.
26 The extent of the medullary cavity (P) is the highest in *Bothriodon*, which shows values
27 similar to most terrestrial forms (Fig. S2F). P is rather similar in *Microbunodon*, *Elomeryx*,
28 *Choeropsis*, and *Brachyodus*, being slightly lower than in most terrestrial taxa. It is lower in
29 rhinos and *Hexaprotodon*, and clearly low in *Hippopotamus*. Values vary strongly in aquatic
30 taxa. S, the transition zone (between the compact cortex and the medullary cavity), is rather
31 similar in *Choeropsis*, *Microbunodon*, *Elomeryx*, and *Hexaprotodon* compared to most other
32 terrestrial ungulates (Fig. S2G). It is higher in rhinos and much higher in *Hippopotamus*, and
33 intermediate between *Choeropsis* and rhinos for *Bothriodon* and *Brachyodus*. It varies a lot
34 among aquatic taxa. As for the polar section modulus (Zpol), it varies a lot within rhinos

1 where it is the highest but is extremely constant in *Hippopotamus* and *Choeropsis*. It is high
2 in *Hippopotamus*, *Brachyodus*, and *Hexaprotodon* and very high in rhinos (Fig. S2H).

3 4 *Quantitative analyses*

5 All parameters, except Zpol (in mm³), are dimensionless ratios. All parameters except CSS
6 are correlated with size (Table 3). This shows, beyond a size effect, a strong allometry in the
7 data, except for the shape of the diaphyseal sections (CSS).

8 All parameters show a significant phylogenetic signal (at 5%; Table 3), whatever the
9 chosen phylogeny (Fig. 1), with K values however always below 1. This signal, as significant
10 in R, could be partly associated with the allometry, but not only, as CSS is not significantly
11 correlated with size.

12
13 A first PCA was performed on the humerus data but as the specimen of *Maiacetus* was
14 driving too much the variation along PC2, another one was conducted without this specimen.
15 The two first axes of the PCA on humerus data represent 78.30% of the variance with 64.55
16 for the first axis and 13.75% for the second one. They are both correlated with size (Table 3)
17 and commented hereafter. The first axis rather clearly discriminates most terrestrial ungulates
18 (negative values) from semi-aquatic and heavy ones (Fig. 4). Specimens of *Hippopotamus*
19 group together on the positive part. Specimens of *Choeropsis* are rather intermediary between
20 most terrestrial ungulates and the heavy and semi-aquatic ones. Both specimens of
21 *Microbunodon* and the specimen of *Bothriodon* clearly group with most terrestrial ungulates;
22 it is the same for both specimens of *Elomeryx*, which are close to *Choeropsis*'. Conversely,
23 the two specimens of *Brachyodus* are between *Choeropsis* and rhinos. The specimen of
24 *Hexaprotodon* TM 258-01-27 is very close to *Hippopotamus*. The first axis is essentially
25 driven by RSDT, P, C, and Zpol (RMeanT and S to a slightly lower extent), which all show
26 allometry. Zpol and S covary, and P varies antagonistically with C. This axis shows that
27 larger humeri show a higher compactness (C), a reduced medullary area (P, RMeanT),
28 resulting in a higher resistance to bending and torsion (Zpol), but also a wider transition zone
29 between the compact cortex and the medullary cavity (S), associated with the filling of the
30 medullary cavity by a spongiosa, and also a higher variation of the cortical thickness in the
31 transverse section (RSDT), which means a more heterogeneous growth in width of the bone.
32 The second axis poorly discriminates groups. This axis is essentially driven by the cross-
33 sectional geometry (CSS), and to a lesser extent by the transition zone (S) and Zpol.

34

1 **Femur**

2 *Qualitative analyses*

3 *Choeropsis* shows a femoral inner organization similar to that of the humerus, except that the
4 trabecular network appears much looser in most of the diaphysis (Fig. 5A-B). In
5 *Hippopotamus*, the cortex is thicker than in *Choeropsis* but its thickness is rather
6 homogeneous along the diaphysis (Fig. 5C-D). Both taxa show a medullary space partially
7 filled by a loose spongiosa. Conversely, in all the other hippopotamoids sampled, the
8 medullary space appears empty (Figs. 5E-J, 6C-H) so that their organization is tubular. Only
9 the thickness of the cortical layer varies, being the lowest in *Elomeryx*, whose section is
10 similar to those of *Me. medioximus* and *Mi. silistrense* (Cooper et al. 2006).

11 12 *Quantitative comparisons*

13 Femoral compactness in transverse sections of *Elomeryx* is rather similar to that of most
14 terrestrial ungulates (Fig. S3). However, it is progressively higher in *Choeropsis*,
15 *Hexaprotodon*, rhinos, *Brachyodus*, *Libycosaurus*, and *Hippopotamus* that show the highest
16 compactness values, with aquatic ungulates (Fig. S4). CSS values are much higher in rhinos
17 than in all other taxa; hippopotamoids show values similar to (e.g., *Elomeryx*) or lower
18 (especially *Choeropsis* and *Hippopotamus*) than those of the terrestrial ungulates. *Elomeryx*,
19 like rhinos and most terrestrial ungulates, has rather low values of relative cortical thickness
20 (RMeanT), which are minimal in the aquatic ungulates. Values are higher in *Choeropsis* and
21 *Libycosaurus*, and much higher in *Hippopotamus*, *Brachyodus*, and *Hexaprotodon*. RSDT is
22 rather low in *Elomeryx*, most terrestrial and aquatic ungulates. It is slightly higher in
23 *Choeropsis* and progressively much higher in *Hexaprotodon*, *Libycosaurus*, and rhinos,
24 *Hippopotamus*, and *Brachyodus*. The size of the medullary cavity in *Elomeryx* is like in most
25 terrestrial ungulates, whereas it is smaller in rhinos and *Choeropsis*, and much smaller in
26 *Brachyodus*, *Hexaprotodon*, *Libycosaurus*, and *Hippopotamus*. It is minimal in aquatic taxa.
27 The transition zone (S) is much wider in rhinos than in all other taxa and minimal for most
28 terrestrial ungulates. As for the polar section modulus (Z_{pol}), it is similar as in most terrestrial
29 ungulates in *Choeropsis*, *Elomeryx*, and aquatic ungulates, but higher in *Hippopotamus* and
30 *Libycosaurus*, much higher in rhinos and *Hexaprotodon*, and very high in *Brachyodus*.

31 32 *Quantitative analyses*

33 In the femora, all variables, except CSS, are significantly correlated with size (Table 4).

1 All parameters show a significant phylogenetic signal, for both phylogenies (Table 4), with
2 high K values for C and, to a lesser extent, P. Again, the signal could be partly associated with
3 the allometry.

4
5 The first two axes represent 72.93% (54.07, and 18.86%, respectively) of the variance
6 for the femur data. Only PC1 is correlated with size (Table 4). Again, C and RMeanT vary
7 antagonistically to P. Zpol covaries with RSDT. As for the humerus, PC1 distinguishes most
8 terrestrial ungulates from rhinos and massive hippopotamoids (Fig. 7). *Choeropsis* specimens,
9 like those of *Tapirus terrestris* and some large terrestrial ungulates (e.g., *Alces americanus*,
10 *Bubalus bubalis*, *Syncerus caffer*), are intermediate between most terrestrial ungulates and
11 rhinos and large hippopotamoids. *Elomeryx* groups with most terrestrial ungulates, whereas
12 *Libycosaurus* and *Hexaprotodon* are between *Choeropsis* and *Hippopotamus*, and *Brachyodus*
13 gathers with *Hippopotamus*. The first axis essentially discriminates based on RSDT, C, P,
14 Zpol, and RMeanT. Bones on the positive part of the axis show a higher compactness, a
15 reduced medullary cavity, and a wider transition zone, with a cortical thickness being more
16 heterogeneous along the transverse section. As it is correlated with size (but not only), this
17 trend can be generalized to femora when they become larger in our sample. Along the second
18 axis, rhinos are clearly distinct from hippopotamoids and from most terrestrial ungulates.
19 They group with tapirs and the aquatic *Remingtonocetus*. The second axis is essentially driven
20 by CSS and S and, to a lesser extent, by RMeanT.

23 Discussion

25 Hippopotamoid microanatomical features

26 Humerus

27 This study reveals a high variability in humerus microanatomy within hippopotamoids. It
28 clearly shows for the first time, thanks to longitudinal sections, that the medullary area of
29 *Choeropsis liberiensis* is completely filled by a spongy tissue. This is also the case for
30 *Hippopotamus amphibius*, as previously shown in Wall (1983) and in other hippopotamoids.
31 It is clear in *Saotherium* cf. *S. mingoz*, *Hexaprotodon garyam*, and *Libycosaurus bahri*, likely
32 in *Brachyodus onoideus*, but it is unclear for *Bothriodon velaunus* and *Paenanthracotherium*
33 *bergeri*. A spongy medullary area occurs in only a few terrestrial mammals with most of

1 them showing a tubular organization (Canoville and Laurin 2010; Laurin et al. 2011;
2 Houssaye et al. 2018; Fig. S1). The spongiosa is tighter and more extended along the whole
3 diaphysis in these hippopotamoids than in some sloths, anteaters and armadillos (Montanez-
4 Rivera et al. 2018) as well as some otters (Houssaye and Botton-Divet 2018), but rather close
5 to what is observable in tapirs (AH, pers. obs.), the giant anteater, large ground sloths (Amson
6 and Nyakatura 2018), and large otters (Houssaye and Botton-Divet 2018), whereas it is tighter
7 in rhinos (AH, pers. obs.). The thickening of the cortex near the growth center in the humerus
8 of *Hippopotamus* and one specimen of *Hexaprotodon garyam* is similar to what is observed in
9 the sea otter (Houssaye and Botton-Divet 2018), some rhinos (Fig. S1X; AH, pers. obs.), and
10 maybe also in the ground sloth *Hapalops* (Amson and Nyakatura, 2018). It evokes what is
11 observed in the astrapothere *Parastrapotherium* sp., though the cortex of the latter is less
12 thick – approximating the condition in *Libycosaurus bahri* and *Brachyodus onoideus* – and in
13 the notungulate *Nesodon imbricatus*, which nevertheless shows an empty medullary cavity
14 and is maybe closer to *Paenanthracotherium bergeri* (Houssaye et al. 2016b). The
15 organization in *Choeropsis* appears close to that of *Tapirus terrestris* (Fig. S1U). *Elomeryx*
16 *borbonicus* and *Mi. minimum*, like most specimens of *Merycopotamus* (except that of *Me.*
17 *dissimilis*; Cooper et al. 2016; Fig. S1A-B), display a tubular inner structure rather close to
18 that of large terrestrial mammals, like *Alces* and *Bubalus bubalus*, but also of the small semi-
19 aquatic *Hyemoschus aquaticus* and *Indohyus indirae*.

20 A high compactness, as compared to most terrestrial ungulates, is observed in all
21 hippopotamoids except *Elomeryx*, *Microbunodon*, and *Me. medioximus*, and it is maximal in
22 *Hippopotamus* and *Hexaprotodon*, which show a much thicker cortex around the growth
23 center. The more massive taxa (*Hippopotamus*, *Hexaprotodon*, *Brachyodus*, and rhinos) show
24 the most heterogeneous distribution of cortical bone along the transverse section, and thus a
25 heterogeneous bone growth in diameter. Their bone structure suggests a high resistance to
26 torsion and bending (though less than for rhinos) as indicated by their high polar section
27 modulus values.

28 29 *Femur*

30 A spongy organization is also observed in the femur of *Choeropsis* and *Hippopotamus*. In
31 *Choeropsis*, it evokes what is observed in some large otters (*Lontra*) and *Tapirus terrestris*
32 with an empty core of the medullary area (Houssaye and Botton-Divet 2018; Fig. S3T). The
33 spongiosa is also less tight than in *Hippopotamus*, where it is less tight than in rhinos (Fig.
34 S3V-Y). *Hippopotamus* does not show a thickening of the cortex around the growth center as

1 in the humerus though its cortex is thicker than in *Choeropsis*. All other hippopotamoids
2 sampled display a medullary area with as much spongy deposits as in *Choeropsis*, or less.
3 There is less variability in microanatomical features among hippopotamoid femora. Most,
4 except *Elomeryx borbonicus*, show a cortex thicker than in the biggest terrestrial ungulates,
5 like *Syncerus caffer*, or than the putative semi-aquatic *Indohyus indirae* (Fig. S3B,J), and
6 rather evoking *Pteronura brasiliensis* (Houssaye and Botton-Divet, 2018). *Elomeryx*
7 *borbonicus* sections are similar to those of *Hyemoschus*, *Bubalus*, and *Alces*. As for *Tapirus*
8 *terrestris*, *Parastrapotherium* sp., and *Nesodon imbricatus*, they display a relatively thinner
9 cortex with a spongy transition zone between the compact cortex and the medullary cavity
10 and an open medullary cavity (Fig. S3T; Houssaye et al. 2016b), which thus differs from the
11 patterns observed in the sampled hippopotamoids, though only slightly to *Choeropsis*'. The
12 organizations observed in most hippopotamoids sampled (i.e., except *Choeropsis*,
13 *Hippopotamus*, and *Elomeryx*) appear thus only similar to what is observed in various otter
14 femora (Houssaye and Botton-Divet 2018) and *Syncerus caffer*, based on the comparative
15 material available to date.

16 We generally observe the same interspecific variations in compactness in the femur
17 than in the humerus. All hippopotamoids except *Elomeryx* show a high compactness as
18 compared to terrestrial ungulates, especially *Hippopotamus*, though to a much lesser extent
19 than in the humerus (like for one specimen of *Hexaprotodon*). The cortex is proportionally
20 thicker in *Hippopotamus*, *Brachyodus*, and *Hexaprotodon*. As for the humerus, the more
21 massive taxa show the most heterogeneous organization along the section. Contrary to what is
22 observed for the humerus, the cross-sectional shape (as indicated by CSS) differs significantly
23 between hippos (whose femoral diaphysis is more cylindrical) and rhinos, which also show a
24 larger transition zone linked to their tighter spongiosa. The bone structure of *Brachyodus* and
25 *Hexaprotodon* (and of rhinos) suggests a higher resistance to torsion and bending than in the
26 other taxa.

29 **Microanatomical features linked to size**

30 There is a phylogenetic signal in all parameters analyzed. However, as size also follows the
31 phylogeny (whatever the phylogeny chosen), and as all parameters (except CSS, for both
32 bones) are correlated with size, it is difficult to distinguish the phylogenetic signal from the
33 allometry. In the quantitative analyses, the impact of size appears to be the main driver of
34 variation. The analyses have shown that in both humeri and femora an increase in bone

1 absolute width is associated with a relative increase in compactness, a more heterogeneous
2 growth in width of the bone, a wider transition zone between the compact cortex and the open
3 medullary cavity, a more reduced medullary area, and the filling of the medullary cavity by a
4 spongiosa. This confers to the bone a higher resistance to bending and torsion. This is
5 consistent with previous studies including heavy taxa (Houssaye et al. 2016a,b). The intensity
6 of the observed microanatomical specializations - increase in cortical thickness and spongiosa
7 in the medullary area - are more intense in humeri than in femora in many hippopotamoids
8 (see below).

9 If the size parameter (radius of the section approximated as a circle), which reflects
10 animal weight because long bone diameter is known to be correlated with body mass
11 (Anderson et al. 1985), appears as the main driver distinguishing hippopotamoids and rhinos
12 from other ungulates in the quantitative analyses, the qualitative observations allow to better
13 distinguish and characterize the microanatomical specializations of the various taxa analyzed.
14 Indeed, qualitative observations are also based on longitudinal sections and not only
15 transverse ones and thus cover a much larger proportion of the bone. Moreover, the
16 quantitative parameters do not include enough information about the trabecular bone that
17 would allow to better decipher between taxa, notably between rhinos and hippos in our case.
18

19 **Ecological inferences**

20 In *Choeropsis*, humeri and femora display rather similar microanatomical organizations,
21 characteristically differing from most terrestrial ungulates by showing a filling of the
22 medullary area by spongy bone. Both bones show microanatomical features very similar to
23 those of *Tapirus terrestris* (despite a slightly thinner cortex in the latter). Both animals are of
24 approximately the same weight and forage in similar habitats, i.e., lowland forests close to
25 streams, rivers, or swamps (Medici et al. 2001). The filling of the medullary area is also
26 observed in other semi-aquatic mammals (e.g., large otters [Houssaye and Botton-Divet
27 2018]) and is probably associated with their frequent forays into the water.

28 *Hippopotamus* displays a thickening of the cortex - stronger in the humerus than in the
29 femur, especially near the growth center - and the filling of the medullary area by spongy
30 bone. This is consistent with *Hippopotamus* being more specialized than *Choeropsis* for an
31 aquatic lifestyle as it spends most of the day submerged in water. Its nevertheless still active
32 locomotion on land, impeding more drastic functional aquatic adaptations (Fisher et al. 2007;
33 2010), is consistent with the absence of osteosclerosis (strong increase in whole bone
34 compactness; see Houssaye 2009).

1 Both extant hippopotamids show muscular adaptations associated with body weight
2 support and increased resistance against water, especially in the forelimb to propel the trunk
3 forward and in the autopod loading for stability on wet ground (Fisher et al. 2007).

4 Modifications of the pectoral muscles might explain the stronger compactness observed in
5 hippo humeri than femora.

6 The humeral sections of *Hippopotamus* evoke what is observed in some rhinos.
7 However, *Hippopotamus* femora differ from those of rhinos, whose diaphyses show a
8 heterogeneous (proximo-distal) organization along the diaphysis (Wall, 1983; AH pers. obs.)
9 whereas it is homogeneous in *Hippopotamus*. Longitudinal sections of the long bones of
10 diverse extant rhino species would be required in order to better analyze the variation
11 observed among rhino transverse sections (Fig. S1W-A') considering the strong changes
12 occurring along the diaphysis (Wall, 1983; AH, pers. obs.). This would also allow to
13 determine more precisely to which rhinos *Hippopotamus* is the closest, some rhinos being
14 exclusively terrestrial whereas others spend most of their time in shallow waters (Dinerstein
15 2011), and to link more precisely the microanatomical features observed with the habitat and
16 mass.

17 *Saotherium* cf. *S. mingoz* has a humerus inner structure very close to that of
18 *Choeropsis*, despite being much larger, so that this filling could be only related to mass and
19 not necessarily to a semi-aquatic lifestyle. Femur data were unfortunately not available to
20 verify this hypothesis.

21 Humeri in *Mi. minimum* and humeri and femora in *Elomeryx borbonicus*, as for *Me.*
22 *medioximus* (Cooper et al. 2016; Fig.S1A), have a tubular inner structure close to that of large
23 terrestrial ungulates, like *Alces* and *Bubalus* but also to the small *Hyemoschus*, and *Indohyus*
24 for the humerus. The cortical layer is slightly thicker than in ungulates of similar sizes. This
25 suggests a lifestyle possibly similar to that of *Hyemoschus*, which is essentially terrestrial,
26 living in closed habitats, but foraging along the banks of rivers at night, which is in agreement
27 with previous ecological reconstructions (Lihoreau 2003; Lihoreau and Ducrocq 2007; Table
28 2). No femur is available for *Microbunodon*. The higher morphological robustness of the hind
29 limb in *Elomeryx borbonicus* is not reflected in the microanatomy of its stylopod bone.

30 The humerus of *Bothriodon velaunus* is close to those of *Microbudondon* and
31 *Elomeryx*, though it shows a thinner cortex and a spongy transition zone between a
32 compact cortex and a probably empty core (at least) of the medullary area, with a transverse
33 section close to that of *Tapirus terrestris*. This taxon, very close in size (< 150 kg) and
34 morphology to *Elomeryx borbonicus*, probably also shared a similar lifestyle (Table 2).

1 The humerus of *Paenanthracotherium bergeri* shows a rather thick cortex, slightly
2 thicker than in *Bubalus bubalis* for a similar diameter, and the medullary area appears
3 essentially void of trabeculae (if it is not a preservation artefact), except a few at the transition
4 zone. This organization is rather similar to that of the notungulate *Nesodon imbricatus* (< 500
5 kg; Cassini et al. 2012; Forasiepi et al. 2015; Houssaye et al. 2016b), which suggests an
6 essentially terrestrial lifestyle for this rather massive (< 950 kg) and heavily built animal, in
7 accordance with Scherler et al. (2019; Table 2). This could be compatible with walking, but
8 not underwater swimming or bottom walking in swamps. Unfortunately, no femur was
9 available to further explore this comparison.

10 The humerus of *Brachyodus onoideus* resembles that of *Choeropsis*. The bad
11 preservation of the core of the bone prevents us from knowing to which degree the medullary
12 cavity is filled with a spongiosa and thus to state if it was more similar to *Choeropsis*, to some
13 rhinos, or to the astrapothere *Parastrapotherium* (Houssaye et al. 2016b). Its femur seems
14 clearly void of trabeculae in the medullary area and thus differs from *Choeropsis*,
15 *Hippopotamus*, and rhinos. It also differs from *Parastrapotherium* whose cortex is thinner and
16 the transition zone large (Houssaye et al. 2016b). In *Brachyodus onoideus*, humerus and
17 femur, like in *Choeropsis*, present similar microanatomical organizations, which might
18 suggest comparable relative use of the forelimbs and hind limbs in these two taxa. More
19 specimens would naturally be required to confirm this hypothesis. The relatively thick cortex
20 is consistent with its heavy weight (< 2,350 kg), the spongy tissue in the medullary area of
21 the humerus with a semi-aquatic lifestyle, but animals with aquatic affinities of similar size,
22 i.e., *Hippopotamus* and the Asiatic rhinos combine a thick cortex and spongy tissue in the
23 medullary cavity. *Brachyodus onoideus*, if semi-aquatic (Orliac et al. 2013), would probably
24 have had reduced immersive habits as compared to these species.

25 The humerus of *Libycosaurus bahri* is close to those of *Choeropsis* and *Brachyodus*
26 *onoideus*. As opposed to in *Brachyodus onoideus*, the spongiosa is clearly well developed and
27 extended in the medullary area. As for the femur, it is also void of trabeculae but with a
28 spongy transition zone. *Libycosaurus bahri* is a heavy (< 1,600 kg) animal with a hippo-
29 like morphology. Its microanatomical features are consistent with a semi-aquatic lifestyle as
30 suggested by previous works (Lihoreau et al. 2006, 2014; Table 2).

31 The specimens of *Hexaprotodon garyam* differ in their microanatomical
32 specialization, despite a similar size. One humerus (TM 115-06-01) is close to that of
33 *Saotherium* cf. *S. mingoz*, whereas the other (TM 258-01-31) has a much thicker cortex,
34 especially near the growth center. This difference in cortical thickness, and thus of

1 compactness, is also observed, though to a lesser extent, between the two femora. Both
2 humeri show spongy bone extending in the medullary area. Like for *Brachyodus onoideus*
3 and *Libycosaurus bahri*, the femora show a relatively thick cortex but an open medullary
4 cavity. Both microanatomical patterns are compatible with a semi-aquatic lifestyle for this
5 heavy taxon (<2,200 kg), as suggested by previous studies (Boisserie 2002; Boisserie et al.
6 2005b; Lihoreau et al. 2014; Table 2). However, if one specimen (TM 115-06-01) is more
7 compatible with the lifestyle of *Choeropsis*, the other (TM 258-01-31) is more similar to
8 *Hippopotamus*.

9 *Hexaprotodon garyam* and *Libycosaurus bahri*, like *Hippopotamus amphibius* and
10 contrary to *Choeropsis* and *Brachyodus onoideus*, display a stronger compactness in the
11 humerus than in the femur. *Libycosaurus bahri* and the most compact *Hexaprotodon garyam*
12 specimens evoke what is observed in *Parastrapotherium*. This taxon, supposedly with some
13 species as heavy as *Hippopotamus*, was assumed to be a megaherbivore frequently foraging in
14 aquatic environments (Avilla and Vizcaino, 2005; Flynn et al. 2012). *Parastrapotherium*
15 nevertheless shows a lower increase in compactness and extension of the spongiosa than
16 *Hippopotamus*. *Hippopotamus* is so far the hippopotamoid displaying the strongest increase in
17 bone compactness. *Hippopotamus* is among the heaviest hippopotamoids but not strongly
18 heavier than some others (e.g., *Hexaprotodon* and *Brachyodus*). This taxon might thus
19 illustrate the strongest degree of adaptation to a semi-aquatic lifestyle among hippopotamoids.
20 Only *Pyrotherium romeroi* (of about 3,500 kg; Shockey and Daza, 2004), considered showing
21 extreme microanatomical adaptation to heavy-weight support, exhibits more compact
22 stylopod bones than *Hippopotamus*, at least near the growth center (no longitudinal section is
23 available for *Pyrotherium romeroi*). This extreme increase in bone mass, if indeed not
24 associated with a semi-aquatic lifestyle, remains poorly misunderstood and requires further
25 investigation. Indeed, heavier terrestrial taxa do not show such a high inner compactness
26 (Houssaye et al. 2016a). Only the sea otter shows a similarly high compactness near the
27 growth center (Houssaye and Botton-Divet, 2018) but this specialization is linked to its
28 almost exclusively aquatic lifestyle and not to loading. Comparisons with other large
29 terrestrial and semi-aquatic taxa are required to better decipher the microanatomical changes
30 associated with a semi-aquatic lifestyle from those linked to loading in heavy quadrupedal
31 mammals.

32 33 **Conclusion**

1 This microanatomical study of the stylopod bones of various hippopotamoids and
2 comparisons with diverse terrestrial and semi-aquatic quadrupedal ungulates highlight
3 converging specializations for a semi-aquatic lifestyle and loading. This consists of an
4 increase in bone compactness by filling the medullary area with cancellous bone and a
5 thickening of the cortex. However, comparisons between animals of similar weight suggest,
6 in the case of excessive compactness, a semi-aquatic lifestyle. In this context, the
7 microstructure of *Choeropsis liberiensis* appears to be consistent with an animal that is
8 mainly foraging in forests but frequently invades the water, whereas that of *Hippopotamus*
9 *amphibius* shows a stronger compactness consistent with an animal that spends a large part of
10 its time standing in the water or walking/swimming on the bottom. Accordingly, on the
11 ground of microanatomical features, the extinct taxa *Microbunodon minimum*, *Bothriodon*
12 *velaunus*, *Elomeryx borbonicus*, *Merycopotamus medioximus*, *Paenanthracotherium bergeri*,
13 and probably also *Saotherium* cf. *S. mingoz* are inferred as essentially terrestrial animals,
14 *Brachyodus onoideus* as slightly water-dependent, and *Libycosaurus bahri* and *Hexaprotodon*
15 *garyam* as clearly semi-aquatic, although less specialized for this ecology than *Hippopotamus*
16 *amphibius*. In order to better decipher the microanatomical changes associated with a semi-
17 aquatic lifestyle in relation to those associated with loading in heavy quadrupedal mammals,
18 microanatomical investigations, preferentially including a large part of the diaphysis, and
19 functional analyses on limb bones of other large terrestrial and semi-aquatic taxa are strongly
20 needed.

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12 **Declarations**

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15 **Conflicts of interest/Competing interests** None

16 **Ethics approval** Not applicable

17 **Consent to participate** Not applicable

18 **Consent for publication** Not applicable

19 **Availability of data and material** Microtomographic scans will be available under request to
20 the authors.

21 3D image data (.tif format) obtained by micro-tomography of MNHN specimens will be
22 permanently saved by the MNHN DSI and identified by the inventory number of the
23 specimen. They will be visible as work carried out on the interface <https://3dtheque.mnhn.fr/>
24 and made available via the interface <http://colhelper.mnhn.fr/>. All 3D image data obtained by
25 micro-tomography at the IC2MP (University of Poitiers) are permanently saved by
26 PALEVOPRIM under the responsibility of the director of the research unit (currently: JRB).
27 Inventory will be communicated upon request, and data will be made available depending on
28 the policy of the original specimen repository institution.

29 **Code availability** Not applicable

1

2

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11

12 **Tables**

13
14 **Table 1. List of the material analyzed in this study.** H: humerus; F: femur. Abb:
15 abbreviations, as in Figs 4,7). Mass estimates from Bongianni et al. (1988), Cooper et al.
16 (2011), and Dinerstein (2011), in kilograms. Resol: resolution of the microtomographic scans,
17 in micrometers. B: Bonn; L: London; M: Montpellier; P: Poitiers; Pa: Paris; °: taxa considered
18 as aquatic ungulates in the comparative analyses (Figs. S2, S4). Institutional abbreviations: H-
19 GSP: Howard University-Geological Survey of Pakistan, Quetta, Pakistan; GSP-UM:
20 Geological Survey of Pakistan-University of Michigan, specimens archived in Quetta,
21 Pakistan; JRHRVC: uncatalogued research collection of John R. Hutchinson at The Royal
22 Veterinary College, Hatfield, United Kingdom; KB: Kossom Bougoudi, Centre National de
23 Recherche pour le Développement, N'Djamena, Chad; MHNL: Musée des Confluences,
24 Lyon, France; MNHN: Muséum National d'Histoire Naturelle, Paris, France; NHMUK,
25 Natural History Museum, London, UK; NMB: Naturhistorisches Museum Basel, Switzerland;
26 RON: Ronzon, Musée Crozatier, Le Puy-en-Velay, France; RR: A. Ranga Rao Collection of
27 India currently housed at the Department of Anatomy and Neurobiology at the Northeastern
28 Ohio Universities College of Medicine, Ohio; STIPB: Steinmann-Institut, Universität Bonn,
29 Germany; TM: Toros-Ménalla, Centre National de Recherche pour le Développement,
30 N'Djamena, Chad; UCBL-FSL: Faculté des Sciences de la Terre, Université Claude Bernard
31 Lyon 1, France; UMZC: University Museum of Zoology, Cambridge, United Kingdom; UP:
32 Université de Poitiers, France; ZFMK: Zoologisches Forschungsmuseum Alexander Koenig,
33 Bonn, Germany. *: sections not used in quantitative analyses

(sub)Family	Species	Abb.	Mass	Inventory number	Bone	Resol
Equidae	<i>Equus caballus</i>	Ec	380-600	MNHN ZM AC 1880-29	H,F	78,90(Pa)
Tapiridae	<i>Tapirus pinchaque</i>	Tp	150-200	MNHN ZM AC 1982-34	H,F	80,80(M)
	<i>Tapirus terrestris</i>	Tt	180-300	MNHN ZM MO 1939-225	H,F	55,55(P)
				ZFMK 462	H,F	48,82(B)
Rhinocerotidae	<i>Ceratotherium simum</i>	Cs	1,350-3,500	MNHN ZM MO 2005-297	H,F	78,68(Pa)
				JRHRVC uncat.	H,F	684,771(L)
	<i>Dicerorhinus sumatrensis</i>	Ds	600-775 kg	MNHN ZM AC 1903-300	H,F	58,65(Pa)
	<i>Diceros bicornis</i>	Db	800-1,300	UMZC H.6481	H	518(L)
				NHMUK M92402	H	561(L)
	<i>Rhinoceros sondaicus</i>	Rs	1,200-1,500	MNHN ZM AC A7970	H,F	76,76(Pa)
	<i>Rhinoceros unicornis</i>	Ru	< 2,000 kg	MNHN ZM AC 1960-59	H,F	76,76(Pa)
Tayassuidae	<i>Pecari tajacu</i>	Pt	15-42	MNHN ZM MO 1917-263	H,F	38,38(P)
				MNHN ZM MO 2000-352	H,F	40,40(P)
Camelidae	<i>Lama guanicoe</i>	Lg	90-140	STIPB M7388	H,F	55,55(B)
Suidae	<i>Potamochoerus porcus</i>	Pp	50-115	MNHN ZM MO 1944-234	H,F	53,53(P)
				MNHN ZM MO C.G. 1971-34	H,F	61,61(P)
	<i>Sus scrofa</i>	Ss	44-320	STIPB M56	H,F	173,206(B)

Tragulidae	<i>Hyemoschus aquaticus</i>	Haq	7-16	MHNL 50.002142	H,F	54,54(P)
				MNHN ZM MO 1914-97	H,F	41,41(P)
<u>Giraffidae</u>	<i>Giraffa camelopardalis</i>	Gc	450- 1,930	JRHRVC uncat.	H,F	703,703(L)
	<i>Okapia johnstoni</i>	Oj	180- 320	UMZC H.20302	H,F	352,389(L)
Cervidae	<i>Hydropotes inermis</i>	Hi	11- 15,5	MNHN ZM MO 1874-274	H,F	74,74(P)
				MNHN ZM MO 1971-36	H,F	74,74(P)
Cervidae	<i>Alces alces</i>	Aa	280- 600	MNHN ZM MO 2013-15	H,F	60,51(P)
Cervidae	<i>Alces americanus</i>	Aam	280- 600	UMZC H.17691	H,F	352,416(B)
Cervidae	<i>Capreolus capreolus</i>	Cc	17-30	MNHN ZM AC 1993-221	H,F	Laurin <i>et al.</i> , 2011
Bovidae	<i>Bubalus bubalis</i>	Bb	400- 1000	MNHN ZM MO 1866-56	H,F	79,79(P)
Bovidae	<i>Cephalophus sylvicultor</i>	Cs	45-80	NHMUK ZD 1961.8.9.80 -1	H,F	242,316(B)
Bovidae	<i>Rupicapra rupicapra</i>	Rr	25-60	STIPB M1639	H,F	189,201(B)

Bovidae	<i>Syncerus caffer</i>	Sc	350-900	MNHN ZM MO 1936-72	H,F	56,56(P)
				NHMUK ZD 1874.11.2.4	H,F	352,545(B)
Raoellidae	<i>Indohyus indirae</i> °	Ii	~2	RR 157	H	Thewissen et al. 2007
				RR 42	F	Thewissen et al. 2007
Pakicetidae	<i>Ichthyolestes pinfoldi</i> °	Ip	~7	H-GSP 96227	H	Thewissen et al. 2007
Remingtonocetidae	<i>Remingtonocetus domandaensis</i> °	Rd	-	GSP-UM 3054	F	49(B)
Protocetidae	<i>Qaisracetus arifi</i> °	Qa	-	GSP-UM 3318	H	58(B)
	<i>Maiacetus inuus</i> °	-	-	GSP-UM 3551	H	46(B)
Microbunodontinae	<i>Microbunodon minimum</i>	Mm	~20	UP-L.M. 1967 MA 350	H	46(P)
				UP-L.M. 1968 MA 377	H	46(P)
Anthracotheriinae	<i>Paenanthracotherium bergeri</i>	Pb	< 950	UCBL-FSL 213779	H*	89(P)
Bothriodontinae	<i>Bothriodon velaunus</i>	Bv	< 150	2004-6- 1792-RON	H	44(P)
Bothriodontinae	<i>Brachyodus onoideus</i>	Bo	< 2,350	NMB S.O. 5897	F	93(P)
				MNHN Neu 75	H	57(P)
				MNHN Neu 76	H	78(P)
Bothriodontinae	<i>Elomeryx borbonicus</i>	Eb	<130	UCBL-FSL 8540	F	69(P)
				UCBL-FSL 9285	F	46(P)
				UCBL-FSL 8565	H	59(P)
				UCBL-FSL 8572	H	59(P)
Bothriodontinae	<i>Libycosaurus bahri</i>	Lb	< 1,600	TM 104- 00-003	F	93(P)
				TM 254- 02-010	F	93(P)
				TM 098- 99-001	H*	69(P)

Hippopotaminae	<i>Choeropsis liberiensis</i>	Cl	160-270	MNHN ZM MO 1921-309	H,F	62,73(P)
				MNHN ZM MO 1944-146	H,F	73,77(P)
				MNHN ZM MO 1978-104	H,F	69,76(P)
				ZFMK 65 570	H,F	59,66(B)
Hippopotaminae	<i>Hexaprotodon garyam</i>	Hg	< 2,200	TM 115-06-001	F	93(P)
				TM 258-01-031	F	93(P)
				TM 055-XX-05	H*	93(P)
				TM 258-01-027	H	93(P)
Hippopotaminae	<i>Hippopotamus amphibius</i>	Ha	1,000-4,500	MHNL 50.002123	H,F	93,93(P)
				MNHN ZM MO 1917-249	H,F	93,93(P)
				MNHN ZM MO 1971-308	H,F	93,93(P)
Hippopotaminae	<i>Saotherium cf. S. mingoz</i>	Sm	<1,000	KB 03-97-170	H*	93(P)

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Table 2. Supposed lifestyle of the hippopotamoids sampled based on literature and on this study. H: humerus; F: femur.

Taxon	Data from the literature	Supposed lifestyle	Microanatomical data	Mass (kg)	Inferred lifestyle
Microbunodontinae					
<i>Microbunodon minimum</i>	- Skull morphology: no elevated sense organs (Cabard 1976) - Diet: frugivorous-folivorous (micro and	Terrestrial and solitary in forested habitats	H: tubular	~20	Essentially terrestrial

	<p>mesowear; Lihoreau 2003), - Postcranial morphology: autopod showing a digitigrade stance with weekly elongated metapodials (Lihoreau 2003) - Gregarious habits: proposed to be solitary (Lihoreau 2003) - no data for subaquatic hearing - oxygene isotopic ratio: proposed to be terrestrial (Sherler 2011) and a closely related species is considered as a terrestrial forest dweller (Nelson 2007)</p>				
Anthracotheriinae					
<i>Paenanthracotherium bergeri</i>	<p>- Skull morphology: no elevated sense organs (Sherler et al. 2018) - Diet: folivorous (Sieber 1936), browser on C3 plants in a mesophytic forest (Tütken and Absolon 2015 for a close relative). - Postcranial morphology: heavily-built with short and</p>	Terrestrial forest-dweller	H: thick cortex; open medullary cavity No F	< 950	Essentially terrestrial

	<p>robust limb bones</p> <ul style="list-style-type: none"> - Gregarious habits: no data - no data for subaquatic hearing - Oxygene isotopic ratio: a close relative is proposed to be terrestrial (Tütken and Absolon 2015) -Taphonomy: relatives have historically been found in lignit deposits (Cuvier 1822) 				
Bothriodontinae					
<i>Bothriodon velaunus</i>	<ul style="list-style-type: none"> - Skull morphology: no data - Diet: supposed to have been predominantly folivorous (Lihoreau and Ducrocq 2007) - Postcranial morphology: lightly-built skeleton with gracile legs (Filhol 1881) - Gregarious habits: no data - no data for subaquatic hearing - Oxygene isotopic ratio: no data -Taphonomy: swamp deposit (Filhol 1881) 	Regularly considered to live in swamp habitat	H: tubular with spongius transition zone	< 150	Essentially terrestrial
<i>Brachyodus onoideus</i>	<ul style="list-style-type: none"> - Skull morphology: amphibious 	Terrestrial	H: thick cortex; spongius filling	< 2,350	Possibly

	<p>position of the sensory organs on the head (Orliac et al. 2013)</p> <ul style="list-style-type: none"> - Diet: no data - Morphology of the tympanic bulla potentially enabling underwater directional hearing (Orliac et al. 2013) - Postcranial morphology: terrestrial (Dineur 1981) - Gregarious habits: trackways suggest multi-aged group and important gregarism (Diaz Martinez et al. 2020) - Oxygene isotopic ratio: no data - Taphonomy: shoreline and palustrine environment 	hydrophilic and water immersive habits	of the medullary area F: thick cortex; open medullary cavity		slightly semi-aquatic (with reduced immersive habits)
<i>Elomeryx borbonicus</i>	<ul style="list-style-type: none"> - Skull morphology: low position of the superior border of the orbit and lack of specialization of the tympanic bulla and of the auditory tube (Lihoreau 2003) - Diet: no data - no data for subaquatic hearing 	Semi-aquatic lifestyle, supposedly in marshy habitats	H&F: tubular	<130	Essentially terrestrial

	<ul style="list-style-type: none"> - Postcranial morphology: hind limb more robust than forelimb and proposed occurrence of webbed feet (Geais 1934) - Gregarious habits: sexual dimorphism (Geais 1934) - Oxygene isotopic ratio: no data -Taphonomy: swamp deposit (Chateauneuf and Nury 1995) 				
<i>Libycosaurus bahri</i>	<ul style="list-style-type: none"> - Skull morphology: highly elevated orbits and external nares, compressed and dense tympanic bulla (Lihoreau et al. 2014) - Diet: fresh mixed feeder (microwear and isotopic ratio of Carbon; Lihoreau et al. 2014) - no data for subaquatic hearing - Postcranial morphology: no data - Gregarious habits: large sexual size dimorphism suggests gregarism and polygenous mating system 	Semi-aquatic	<p>H: thick cortex; extensive spongy filling of the medullary area</p> <p>F: thick cortex; open medullary cavity with spongy transition zone</p>	< 1,600	Semi-aquatic

	(Lihoreau et al. 2014) - Oxygene isotopic ratio: aquatic signal compared to the terrestrial fauna (Lihoreau et al. 2014) -Taphonomy: peri-lacustrine deposit (Vignaud et al. 2002)				
Hippopotaminae					
<i>Saotherium cf. S. mingo</i>	- Skull morphology: no elevation of the orbit (Boisserie et al. 2003) - Diet: mixed C ₃ -C ₄ to pure C ₄ (Zazzo et al. 2000). - No data for subaquatic hearing - Postcranial morphology: not published yet - Gregarious habits: no data - Oxygene isotopic ratio: negative values compared to terrestrial fauna (notably bovids, suids – from Jacques 2007). -Taphonomy: peri-lacustrine environment	Terrestrial - Water dependence	H: spongius filling of the medullary area No F	<1,000	Terrestrial
<i>Hexaprotodon garyam</i>	- Skull morphology: moderate elevation of the sensory organs on the head	Semi-aquatic	H: (very) thick cortex; thickening around the growth center; extensive spongius filling	< 2.200	Semi-aquatic

	<ul style="list-style-type: none"> - Diet: opportunistic mixed feeder (Boisserie et al. 2005b; Jacques 2007) - No data for subaquatic hearing - Postcranial morphology: not published yet - Gregarious habits: high frequency in assemblages; moderate sexual dimorphism and agonistic weaponry (Boisserie 2002; Boisserie et al. 2005b) - Oxygene isotopic ratio: aquatic signal compared to the terrestrial fauna (Lihoreau et al. 2014) - Taphonomy: peri-lacustrine deposit (Vignaud et al. 2002) 		F: rather thick cortex; open medullary cavity		
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2 **Table 3 Values obtained for the tests of a size effect and phylogenetic signal** in the various
3 parameters used in the humerus analyses. In bold when significant at 5%

	C	CSS	Zpol	RMea nT	RSD T	S	P	PC1	PC2	R
Size	r= 0. 50 p< 0. 01	r =- 0.16 p=0. 28	r =0.92 p<0. 01	r =- 0.77 p<0.0 1	r =0.92 p<0. 01	r =0.42 p<0. 01	r =- 0.44 p=0. 03	r =0.85 p<0. 01	r =0.29 p=0. 05	-

Phylogeny (A)	K=0.79 p<0.01	K=0.47 p=0.03	K=0.67 p<0.01	K=0.56 p<0.01	K=0.84 p<0.01	K=0.74 p<0.01	K=0.56 p=0.01	-	-	K=0.71 p<0.01
Phylogeny (B)	K=0.79 p<0.01	K=0.47 p=0.03	K=0.67 p<0.01	K=0.56 p<0.01	K=0.84 p<0.01	K=0.75 p<0.01	K=0.56 p=0.02	-	-	K=0.71 p<0.01

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3 **Table 4 Values obtained for the tests of a size effect and phylogenetic signal in the various**
 4 **parameters used in the femur analyses. In bold when significant at 5%**

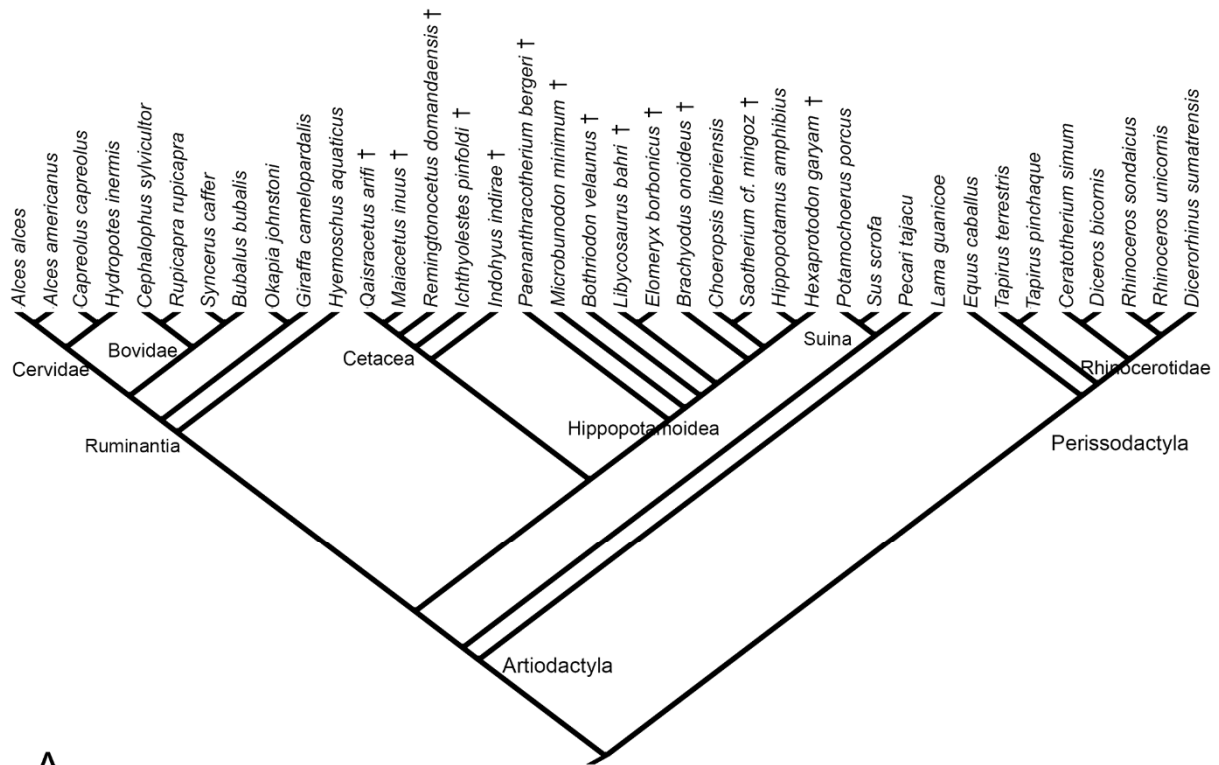
	C	CSS	Zpol	RMea nT	RSD T	S	P	PC1	PC2	R
Size	r=0.58 p<0.01	r=0.26 p=0.08	r=0.93 p<0.01	r=0.66 p<0.01	r=0.83 p<0.01	r=0.53 p<0.01	r=-0.49 p<0.01	r=0.85 p<0.01	r=0.18 p=0.23	
Phylogeny (A)	K=0.97 p<0.01	K=0.51 p<0.01	K=0.47 p=0.01	K=0.56 p<0.01	K=0.58 p<0.01	K=0.59 p<0.01	K=0.84 p<0.01	-	-	K=0.57 p<0.01
Phylogeny (B)	K=1.02 p<0.01	K=0.52 p<0.01	K=0.48 p=0.01	K=0.51 p<0.01	K=0.60 p<0.01	K=0.63 p<0.01	K=0.88 p<0.01	-	-	K=0.58 p<0.01

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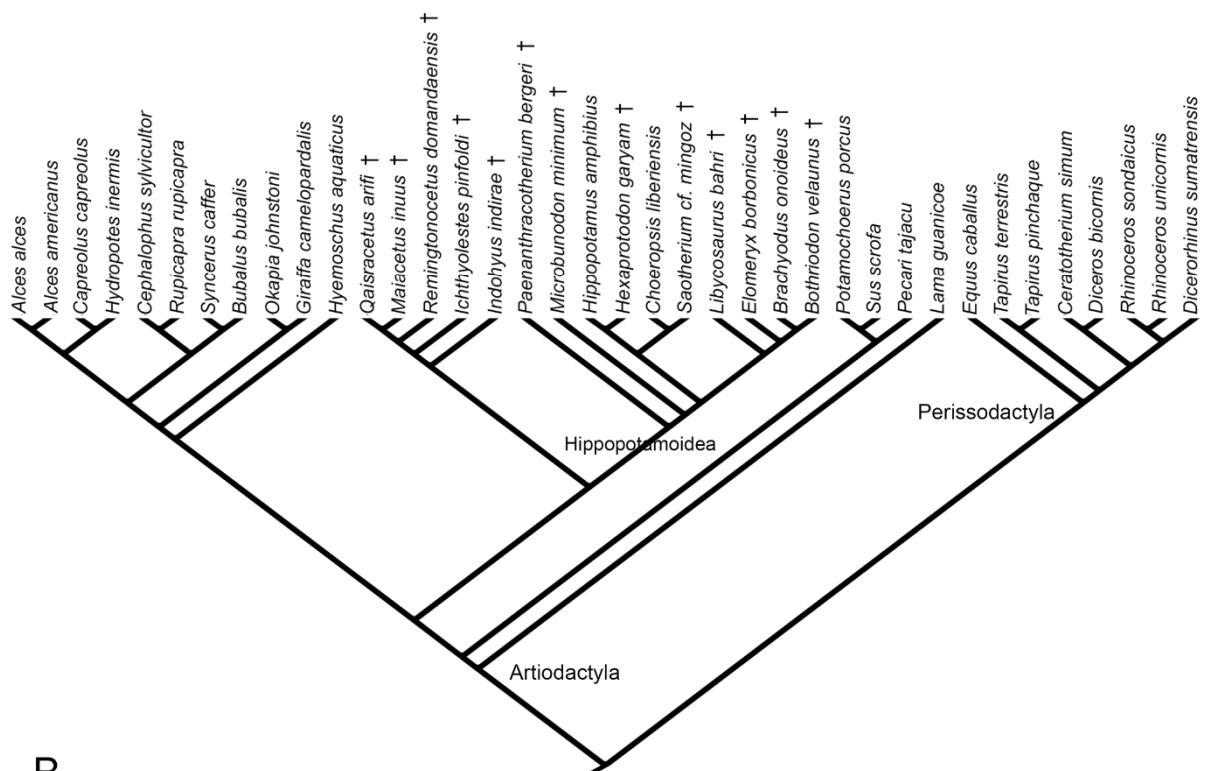
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8 **Figure legends**



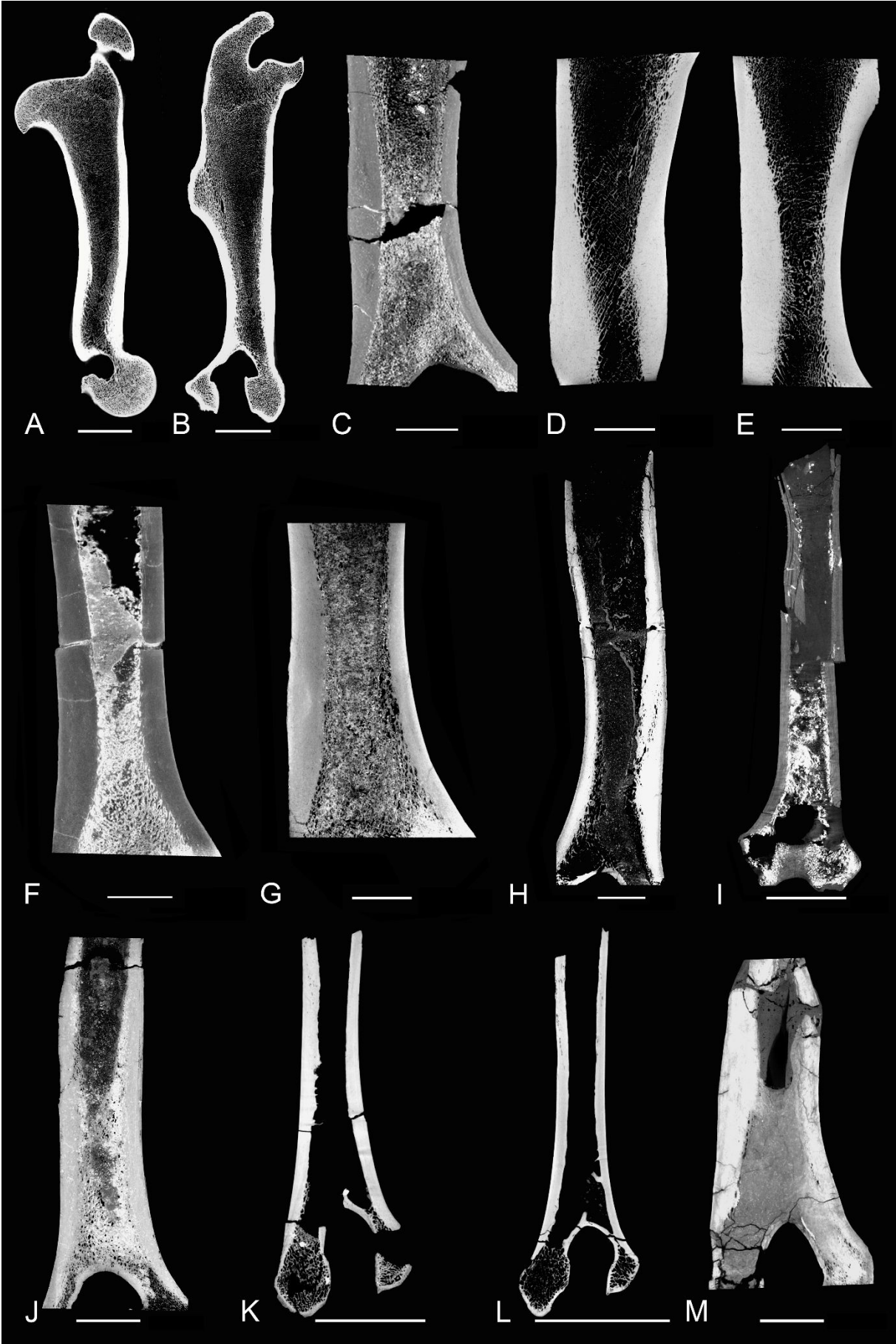
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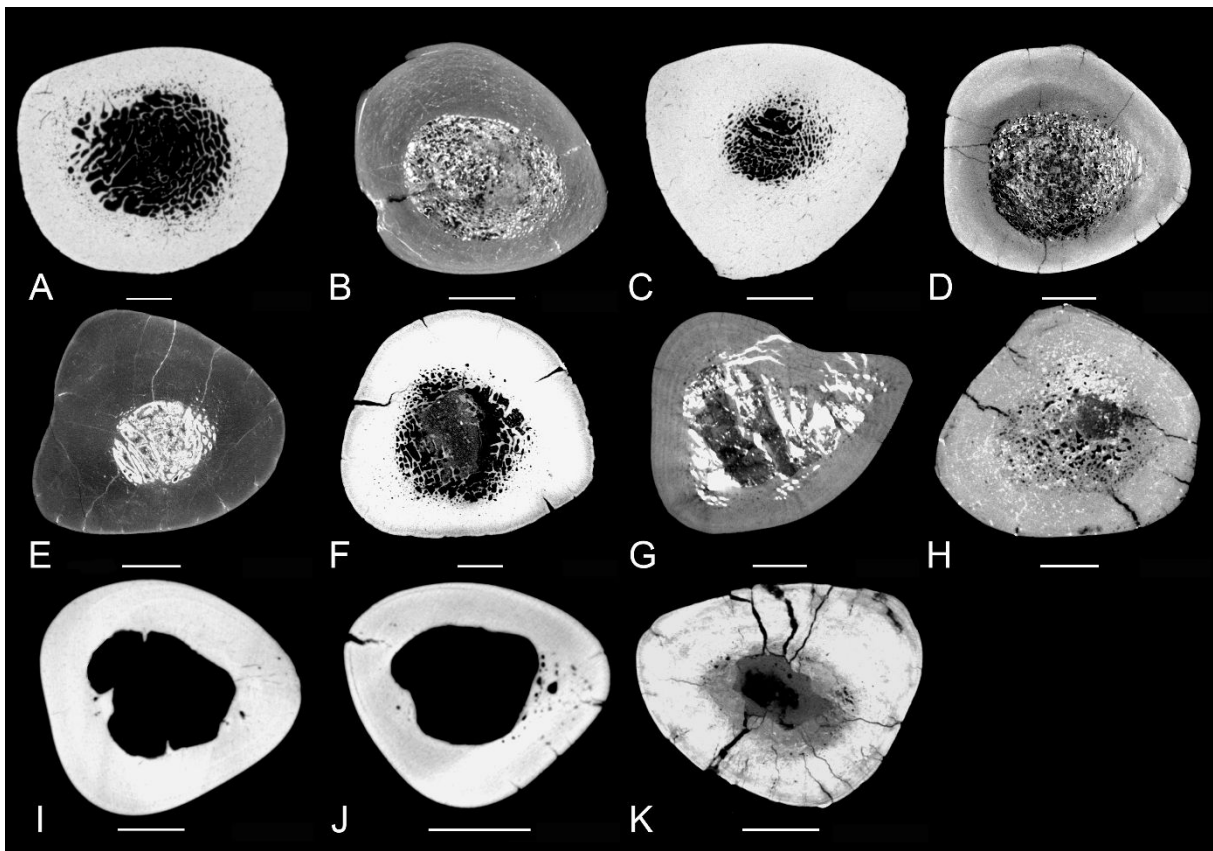
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 2 **Fig. 1 Two consensus phylogenetic trees** including the extant and extinct taxa sampled. **A**
 3 from Boisserie et al. (2011) for the hippopotamines, Lihoreau et al. (2015; SI fig. 17) and
 4 Boisserie et al. (2017) for the other hippopotamoids; **B** from Lihoreau et al. (2015, 2019) and
 5 Gomes Rodrigues et al. (2019) for the hippopotamoids, and **A,B** from Thewissen et al. (2007)

1 and Vautrin et al. (2020) for cetaceans and Gilbert et al. (2006), Bibi (2013), Foley et al.
2 (2016), and Springer et al. (2019) for the remaining mammals. †: extinct taxa.



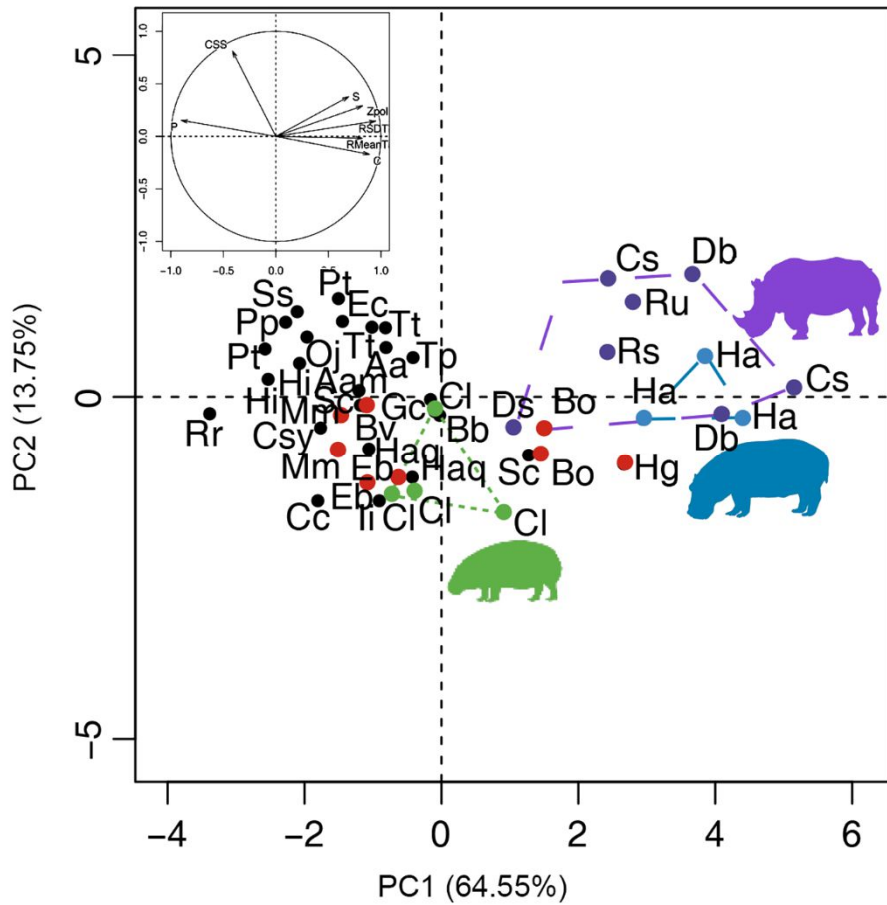
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1 **Fig. 2 Humerus longitudinal sections.** **A,B** *Choeropsis liberiensis* MNHN 1944-146 sagittal
 2 (A) and coronal (B) sections; **C** *Saotherium* cf. *S. mingo* KB 03-97-170 coronal section (CS);
 3 **D,E** *Hippopotamus amphibius* MHNL 50.002123 sagittal (C) and coronal (D) sections; **F**
 4 *Hexaprotodon garyam* TM 55-XX-05 CS; **G** *Hexaprotodon garyam* TM 258-01-27 CS; **H**
 5 *Brachyodus onoideus* MNHN Neu 75 CS; **I** *Bothriodon velaunus* 2004-6-1792-RON CS; **J**
 6 *Libycosaurus bahri* TM 98-99-01 CS; **K** *Elomeryx borbonicus* UCBL-FSL 8572 CS; **L**
 7 *Microbunodon minimum* UP-L.M. 1968 MA 377 CS; **M** *Paenanthracotherium bergeri*
 8 UCBL-FSL 213779 CS. Scale bars equal 3 cm. Partial sedimentary filling of the bone in C, F,
 9 G, H, I, J, and M.



10 **Fig. 3 Humerus transverse sections** near the growth center. **A** *Choeropsis liberiensis*
 11 MNHN 1978-104; **B** *Saotherium* cf. *S. mingo* KB 03-97-170; **C** *Hippopotamus amphibius*
 12 MNHN 1971-308; **D** *Hexaprotodon garyam* TM 55-XX-05; **E** *Hexaprotodon garyam* TM
 13 258-01-27; **F** *Brachyodus onoideus* MNHN Neu 75; **G** *Bothriodon velaunus* 2004-6-1792-
 14 RON; **H** *Libycosaurus bahri* TM 98-99-01; **I** *Elomeryx borbonicus* UCBL-FSL 8572; **J**
 15 *Microbunodon minimum* UP-L.M. 1968 MA 377; **K** *Paenanthracotherium bergeri* UCBL-
 16 FSL 213779. Scale bars equal: A,G,I,J- 5 mm; F,H- 10 mm; B,C,D,E,K- 15 mm. Partial
 17 sedimentary filling of the bone in B, D,E,(F), G, H, and K.

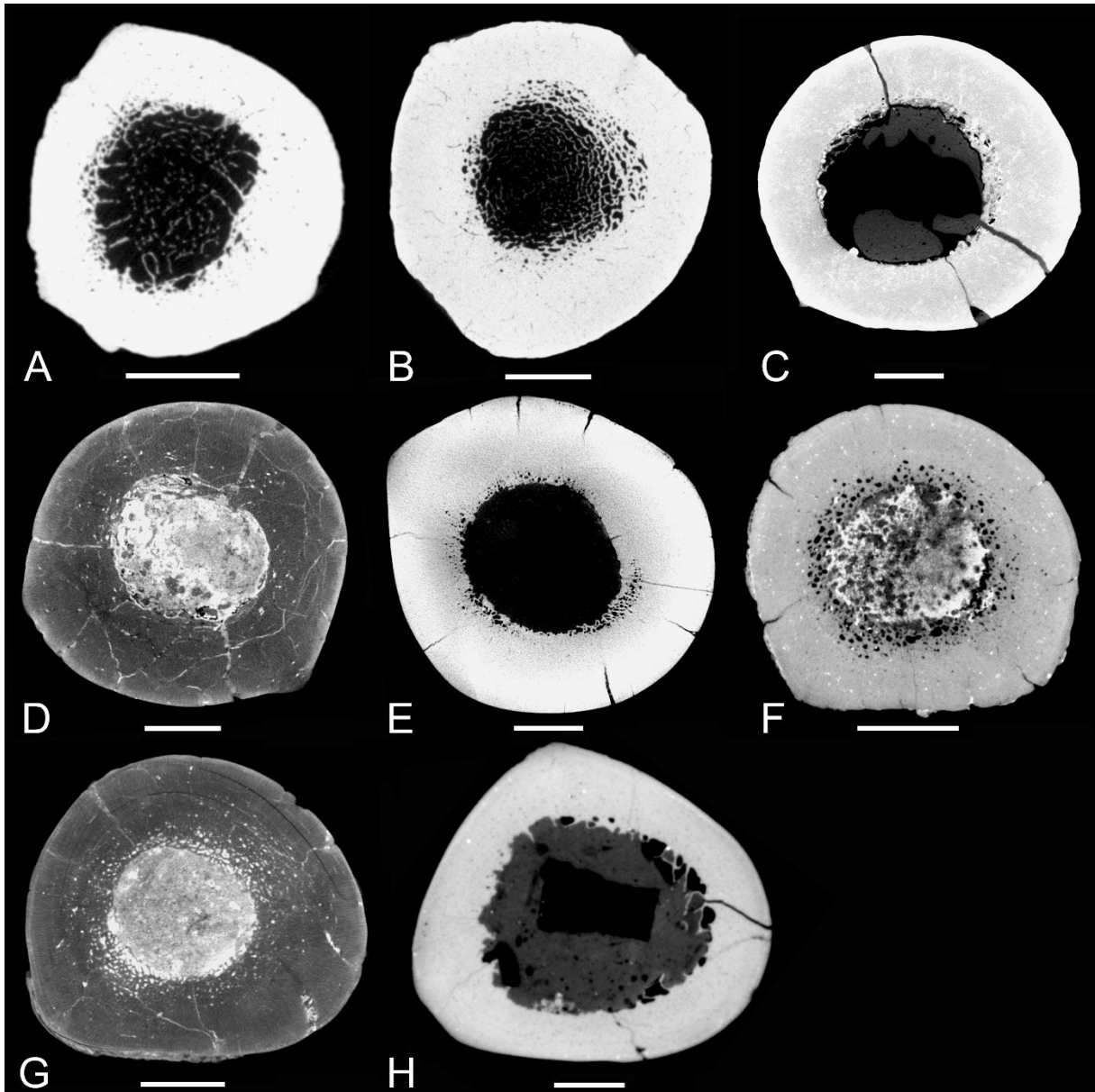
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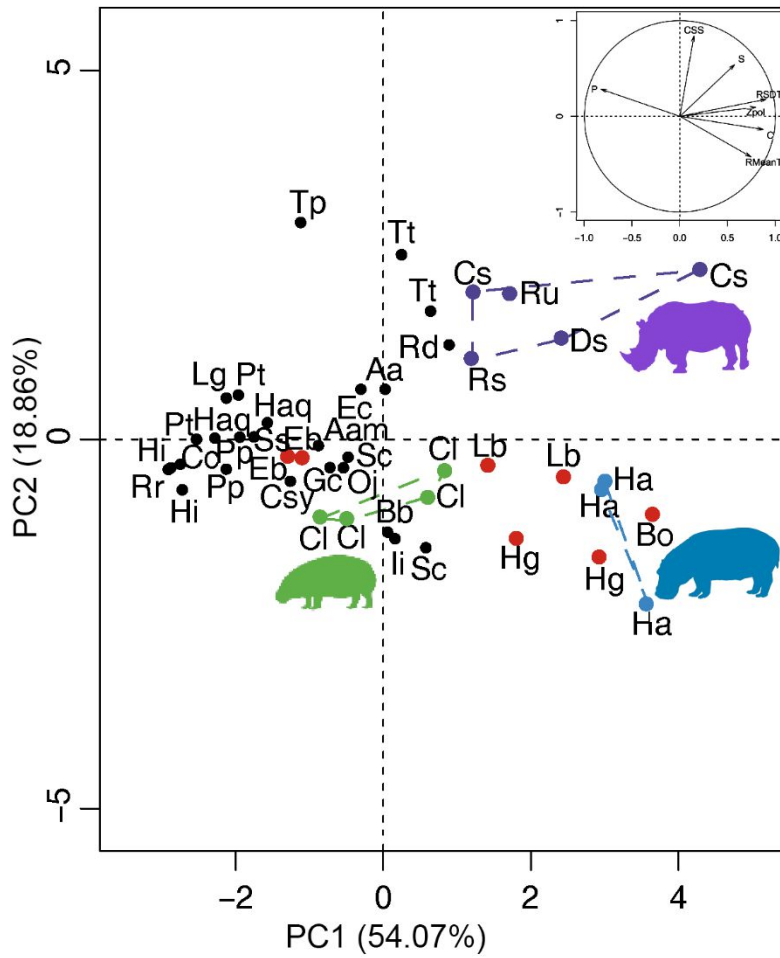
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 2 **Fig. 4 Distribution of the specimens in the morphospace** along the two first axes of the
 3 humerus PCA with the contribution of the different parameters according to the principal
 4 component (PC)1 and PC2 axes. Abbreviations as listed in Table 1



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 2 **Fig. 5 Femur longitudinal sections. A,B** *Choeropsis liberiensis* MNHN 1978-104 SS(A) and
 3 CS(B); **C,D** *Hippopotamus amphibius* MNHN 1971-308 SS(C) and CS(D); **E** *Hexaprotodon*
 4 *garyam* TM 115-06-01 SS; **F** *Hexaprotodon garyam* TM 258-01-31 SS; **G** *Brachyodus*
 5 *onoideus* NMB S.O. 5897 CS; **H** *Libycosaurus bahri* TM 104-00-03 SS; **I** *Libycosaurus bahri*
 6 TM 254-02-10 SS; **J** *Elomeryx borbonicus* UCBL-FSL 8540 SS. Scale bars equal 30 mm.
 7 Partial sedimentary filling of the bone in E, F, G, H, I, and J.



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 2 **Fig. 6 Femur transverse sections** near the growth center. **A** *Choeropsis liberiensis* MNHN
 3 1944-146; **B** *Hippopotamus amphibius* MNHN 1971-308; **C** *Hexaprotodon garyam* TM 115-
 4 06-01; **D** *Hexaprotodon garyam* TM 258-01-31; **E** *Brachyodus onoideus* NMB S.O. 5897; **F**
 5 *Libycosaurus bahri* TM 104-00-03; **G** *Libycosaurus bahri* TM 254-02-10; **H** *Elomeryx*
 6 *borbonicus* UCBL-FSL 8540. Scale bars equal: A- 10 mm; B-G- 15 mm; H- 5mm. Partial
 7 sedimentary filling of the bone in (C), D, F, G, and H.



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 2 **Fig. 7 Distribution of the specimens in the morphospace** along the two first axes of the
 3 femur PCA with the contribution of the different parameters according to the principal
 4 component (PC)1 and PC2 axes. Abbreviations as listed in Table 1. Color code as in Fig. 4
 5