1   **A heavyweight early whale pushes the boundaries of vertebrate morphology**

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26

Summary paragraph

28   The fossil record of cetaceans documents how terrestrial animals acquired extreme adaptations and

29   transitioned to a fully aquatic lifestyle[1,2]. In whales, this is associated with a dramatic increase in

30   maximum body size. While an elongate body was acquired early in cetacean evolution[3], the

31   maximum body mass of baleen whales reflects a recent diversification that culminated with the blue

32   whale[4]. More generally, hitherto known gigantism among aquatic tetrapods evolved within pelagic,

33   active swimmers. Here we describe *Perucetus colossus*, a new basilosaurid whale from the middle

34   Eocene of Peru. It displays the strongest degree of bone mass increase known to date, an adaptation

35   associated with shallow diving[5]. This species' estimated skeletal mass exceeds that of any known

36   mammal or aquatic vertebrate. We show that the bone structure specialisations of aquatic mammals

37   are reflected in the scaling of skeletal fraction (skeletal mass versus whole body mass) across the

38   entire disparity of amniotes. We use skeletal fraction to estimate the body mass of *Perucetus*

39   *colossus*, who proves to be a contender to the title of heaviest animal on record. Cetaceans' peak

40   body mass had already been reached ca. 30 million years before previously assumed, in a coastal

41   context where primary productivity was particularly high.

42

43   **Main text**

44   The fossil record of cetaceans provides one of the most striking documentations of an

45   evolutionary transition of lifestyle[1,2]. This transition brought a fully terrestrial group of mammals back

46   to water, over 300 million years after tetrapods first gained ground[6]. Early to middle Eocene

47   artiodactyls with a chevrotain-like morphology (e.g., *Indohyus*) are understood to be the closest

48   known relatives of cetaceans[7]. Becoming increasingly specialised to aquatic life during this global

49   greenhouse stage, cetaceans quickly evolved larger body sizes, with a first significant trend of body

50   elongation seen in late Eocene basilosaurines[8]. The true gigantism and associated body mass seen in

51   baleen whales is nevertheless a recent acquisition, probably associated with the cooling trend and

52   seasonality installed in the late Cenozoic[4]. Furthermore, the largest cetacean is the extant blue whale

53   (*Balaenoptera musculus*), which is also the heaviest animal hitherto known. More generally, the

54   largest taxa among aquatic tetrapods (which include marine reptiles such as ichthyosaurs[9]) known so

55   far are all pelagic, active swimmers.

56   As animals acquire more aquatic habits, buoyancy becomes a critical aspect of their biology.

57   Bone is dense relative to most other tissues and present in large quantities in the tetrapod body,

58   which likely explains why bone mass specialisations are documented in a myriad of tetrapod lineages

59   that independently evolved aquatic habits[10]. Shallow-diving, slow-swimming species' adaptations

60   often comprise bone mass increase (BMI). This is produced by the infilling of skeletal elements' inner

61   cavities with compact bone (i.e., osteosclerosis) and, in the more extreme cases, by additional

62   deposition of bone on their external surface[5] (i.e., pachyostosis *sensu stricto*). BMI is documented in

63   cetaceans' amphibious close relatives[11], as well as early members of the clade, the basilosaurids in

64   particular. Extant cetaceans have conversely acquired an entirely different bone microanatomy, with

65   an osteoporotic-like structure typical of pelagic, secondarily aquatic tetrapods with more active

66   swimming. Basilosaurids are therefore unique in the sense that they acquired large sizes (up to ca. 20

67   m in body length[3]) and BMI. The degree of their BMI nevertheless did not match, up until now, that

68   of some sirenians for instance, where the whole rib cage is both strongly osteosclerotic and

69   pachyostotic[5].

70   Here we describe a new basilosaurid whale that drastically pushes the upper limit of skeletal mass in

71   mammals, as well as in aquatic vertebrates in general. This early whale combines a gigantic size and

72   the strongest degree of BMI known to date. It also potentially represents the heaviest animal ever

73   described.

74

Systematic palaeontology

76

77   Cetacea Brisson, 1762

78   Archaeoceti Flower, 1883

79   Pelagiceti Uhen, 2008

80   Basilosauridae Cope, 1868

81   Subfamily indet.

82   *Perucetus colossus* gen. et sp. nov.

83

84   **Etymology.** From Peru, the country of origin of the holotype and Latin *cetus* (whale). Species epithet

85   from the Ancient Greek *kolossós* (large statue and by extension any creature of gigantic size and

86   mass).

87   **Holotype.** MUSM 3248 (Museo de Historia Natural, Universidad Nacional Mayor de San Marcos,

88   Lima, Peru), a partial skeleton including 13 vertebrae (two tentatively referred to the last thoracics

89   named here Th-a and Th-b and the others to the anterior lumbars named here L-a - L-k), four ribs,

90   and right innominate lacking the distal portion of the ilium (Fig. 1, Extended Data Figs. 1-4). The

91   animal was likely approaching skeletal maturity (see Supplementary Discussion, Bone histology).

92   **Locality and horizon.** From the upper part of the Yumaque member of the Paracas Formation

93   exposed in the Ica valley a few kilometres south of the Zamaca locality, Ica Province, southern Peru[12]

94   (Extended Data Fig. 5). Its geological age is well constrained between 39.8-37.84 million years ago

95   (Bartonian, middle Eocene) through biostratigraphy and ^{39}Ar-^{40}Ar dating on tephra layer. Coordinates

96   for the type locality are available on request through the corresponding author. See Methods,

97   Extended Data Fig. 5 and Supplementary Methods for more geological and palaeoenvironmental

98   information and detailed justification of the age.

99

100   **Diagnosis.** *Perucetus colossus* differs from all other cetaceans by having an extremely

101   pachyosteosclerotic postcranium. Pachyosteosclerosis is also present in a few other cetaceans[13,14],

102   including the basilosaurid Pachycetinae[15], but to a significantly lesser degree than observed in this

103   new taxon. *P. colossus* belongs to Pelagiceti by having a high number of lumbars (at least 11), an

104   extremely reduced innominate, and centra of the last two thoracics and preserved lumbars with a

105   roughly circular cross-section (CH/CW > 0.80), not dorsoventrally compressed nor heart-shaped (as

106   observed instead in the more stemward archaeocetes). Within Pelagiceti, *P. colossus* shares with

107   *Basilosaurus*, *Chrysocetus*, *Cynthiacetus peruvianus*, *Mystacodon* and *Pachycetus wardii* a

108   plesiomorphic, well-defined acetabulum on the innominate, but differs from *Basilosaurus*,

109   *Chrysocetus* and *Mystacodon* and probably *Pachycetus* by the more robust proximal portion of the

110   ilium, and from *Basilosaurus* by the larger obturator foramen and the overall shape of the

111   innominate, which is triangular in profile view. *P. colossus* differs from all cetaceans but

112   Basilosaurinae and Pachycetinae by the great elongation of the centra of the lumbars (CL/CW = 1.25-

113   1.56), approaching the values seen in *Pachycetus* and *Antaecetus* (CL/CW = 1.30-1.68); more extreme

114   values are observed in *Basilosaurus* (CL/CW = 169-1.98) (Extended Data Fig. 6). *P. colossus* shares

115   with *Basilosaurus* the giant size (CW > 20 cm) and club-like shape of the distal end of at least some of

116   the ribs. Estimated skeletal length: ca. 17-20 m (see Supplementary Methods and Fig. S7).

117   The assignation of the vertebrae to the last two thoracic and the first 11 lumbar positions is based on

118   the ventral position of the transverse processes, the large neural canals (although reduced due to the

119   pachyostosis of the neural arches), and the lack of foveae for the capitula of the ribs. The transverse

120   processes are significantly bent ventrolaterally, similarly to the lumbars of other basilosaurids[16–18].

121   Moreover, all the distal portions of the transverse processes exhibit a peculiar wide, oval, flattened

122   area on their ventral surface (Fig. 1j; Extended Data Figs. 1-4). The two thoracic vertebrae also bear a

123   concavity at the anterolateral tip of their transverse processes, where the last ribs likely articulated.

124   These two vertebrae are also distinguished by their neural spine, which is more slender and with a

125   dorsal edge sloping posteriorly.

126   For roughly half of the recovered vertebrae, either one or both of the centrum epiphyses are missing,

127   suggesting partial epiphyseal fusion. This should not be seen as a sign of immaturity for the

128   specimen, as some large, extant cetaceans maintain their thoracic and lumbar centrum epiphyses

129   unfused late into adulthood[19].

130   The best preserved rib displays a simple proximal end without distinct tuberculum and capitulum and

131   a weak overall curvature in anterior or posterior view (Fig. 1e,f; Extended Data Fig. 6c;

132   Supplementary Data 1). This morphology (also observed in the other three preserved ribs) is

133   consistent with a rib of the posterior region (R17-20), which would entail that the whole rib cage was

134   likely pachyostotic in *P. colossus*, contrary to other basilosaurids[16,17,20].

135

136   **The heaviest mammalian skeleton**

137   The extreme skeletal morphology of the new species is not pathological. First, bone mass

138   increase (BMI) is part of the healthy phenotype expressed in many aquatic tetrapods[10]. It is common

139   in basilosaurids, and pachyosteosclerosis is even considered as a diagnostic feature of one of its

140   subfamilies, the pachycetines[15]. Secondly, in conditions involving additional bone deposition such as

141   acromegaly or osteopetrosis, both gross morphology and bone inner structure alterations are highly

142   variable, both within a single bone and across the skeleton[21,22]. In contrast, in aquatic tetrapods with

143   BMI (including *P. colossus*), skeletal elements are uniformly affected by these alterations. Finally, in *P.*

144   *colossus*, the pachyostosis affects all the bones that have been recovered (vertebrae, ribs and

145   innominate). It is most conspicuous in the strongly bloated apophyses of the vertebrae, which hence

146   strikingly differ from those of the vast majority of mammals (Figs. 1-2, Extended Data Figs. 1-4). All

147   extant cetaceans, including the largest balaenopterids and balaenids, show the usual, relatively thin

148   vertebral apophyses[23]. Pachyostosis increases significantly the volume of the new species' vertebrae,

149   which represents almost twice that of the largest vertebra of a 25-m long blue whale (Fig. 2;

150   Supplementary Table 1). The recovered ribs are also entirely pachyostotic (Fig. 1e-f, Extended Data

151   Fig. 4m). We estimate *P. colossus*' total skeletal volume (using that of the preserved elements and

152   virtually complete skeletons of other basilosaurids as well as a conservative vertebral count) to be

153   2.9-4.1 m^3. Scaling up the vertebrae of *Cynthiacetus peruvianus*, a smaller, non-pachyostotic close

154   relative, it can be estimated that the pachyostosis of *P. colossus* results in an excess of over 350 % in

155   overall volume (Supplementary Methods).

156   The bone microanatomy of *P. colossus* is also consistent with an extreme case of BMI: all

157   observed break surfaces and histological samples indicate strong osteosclerosis (Fig. 3; Extended

158   Data Fig. 7; Supplementary Discussion and Table 2). The rib is entirely made of compact bone, lacking

159   a medullary region altogether. Growth marks (annuli) indicate strong cortical drift, with the original

160   growth centre positioned in the current medial-most edge of the bone. A similar but weaker bone

161   drift is seen in *Basilosaurus*[13,24]. Histological sections of both the vertebrae and rib show a very

162   consistent reticular woven-parallel complex devoid of remodelling, making up most of the skeletal

163   elements. The network of primary osteons is extremely dense and convoluted (Fig. 3). The vascular

164   canals themselves are very narrow (most primary osteons are mature with a small lumen), increasing

165   further the compactness of the bone tissue. The same type of bone tissue is described for the rib

166   cortex of *Basilosaurus*, but in the latter various degrees of primary osteon completion are observed,

167   resulting in a less compact cortex[24]. As pachyostosis, osteosclerosis is therefore generalised, too. The

168   fact that all recovered elements (including the strongly reduced innominate) are affected by BMI

169   suggests that the responsible process acted systemically, as already argued for some other marine

170   mammals with BMI[25].

171   We estimated the total skeletal mass of *P. colossus* combining the skeletal volume (Extended

172   Data Fig. 8 and Methods) to the skeletal elements' density (based on palaeohistological samples;

173   Supplementary Methods). With estimates ranging from 5.3 to 7.6 tonnes (Supplementary Table 22),

174   the skeleton weighed 2.0-2.9 times as much as that of a 25-m long blue whale (Supplementary Table

175   6).

176   **Skeletal mass across amniotes**

177   The repeated acquisition of bone mass increase in aquatic amniotes underlines the

178   importance for these animals of body trim and overall density[26], which are defined by the

179   distribution and mass of skeletal and soft tissues. To assess how skeletal fraction (ratio of skeletal

180   and total body mass, SF) relates to lifestyle and body size, we examined the scaling of dry skeletal

181   mass in a sample of mammals with known body masses (Fig. 4; Supplementary Tables 3-4;

182   Supplementary Data 2). Compared to terrestrial mammals, cetaceans have a relatively lighter

183   skeleton, with SF values ranging from 2.2 to 5.1%. In terrestrial mammals, skeletal mass scales with

184   positive allometry, and SF ranges from 4 to 10%. The skeletal mass in amniotes actually spans nine

185   orders of magnitude (our estimates range from 26 milligrams for the dwarf chameleon *Brookesia*

186   *nana* to 9.9 tonnes for the sauropod *Argentinosaurus huinculensis*). The skeletal mass of extant

187   sirenians essentially scales as that of terrestrial mammals. These differences of scaling emphasise the

188   two types of aquatic specialisations described in extant cetaceans and sirenians, the osteoporotic-

189   like condition and BMI, respectively[5].

190   Estimating body mass in basilosaurids is challenging[27]. For *P. colossus*, methods based on

191   simple skeletal measurements would also likely be biased by the fact that its skeletal morphology

192 starkly departs from that of other marine mammals. Furthermore, the excess of skeletal mass might

193 have been compensated for by large amounts of blubber (less dense than most other soft tissues in

194 amniotes), which in turn would strongly affect the overall density of soft tissues. But the scaling

195 relationships described above, which can be used to estimate body mass based on skeletal mass, are

196 indicative of the viable SF values found in fully aquatic mammals (as for all other aquatic amniotes

197 the necessity of frequent surfacing to breathe prevents any strong deviation from neutral buoyancy).

198 Given the two patterns observed for the SF of fully aquatic mammals, we have used the extreme

199 values measured in extant sirenians and cetaceans: assuming the mean SF of adult manatees (extant

200 marine mammals with the strongest BMI) and the minimum skeletal volume estimation yields a body

201 mass of 85 tonnes for *P. colossus*. Using extant cetaceans' lowest SF and the maximum skeletal

202 volume yields an estimation of 340 tonnes (Supplementary Table 22). With the mean of the skeletal

203 mass estimates and the mean cetacean SF (3.6%), an estimated body mass of 180 tonnes is obtained.

204 These estimates fall in or exceed the body mass distribution of the blue whale[28], which hence

205 challenges the latter species' title of heaviest animal that ever existed.

206

Perucetus' lifestyle and the evolution of cetacean gigantism

208 The combination of extreme pachyostosis and osteosclerosis displayed by *P. colossus* is only

209 approached by tetrapod species that are adapted to a fully aquatic lifestyle, as it would make

210 movement on land overly problematic due to gravity[29,30]. Coupled with the greatly reduced size of

211 the innominate, this observation confirms that *P. colossus* did not return to land, not even to give

212 birth. The extreme pachyosteosclerosis of *P. colossus* is only consistent with a shallow-water lifestyle,

213 wherein it would have contributed to the buoyancy control, as in extant sirenians[26,31].

214 Such a large and heavy animal may also have been able to counteract waves in high-energy

215 waters[30,32], as also hypothesised for the giant, recently extinct Steller's sea cow, *Hydrodamalis*

216   *gigas*[29]. A coastal habitat has already been proposed for basilosaurids based on stable isotopes and

217   skeletal anatomy[33,34].

218   While drawing precise conclusions regarding the swimming style of *P. colossus* is hampered

219   by the fragmentary nature of the holotype, a tentative reconstruction can made given the elements

220   at hand. The elongate centra of the vertebrae of *P. colossus* is a character that, in other basilosaurids,

221   has been related to swimming by means of axial undulation[15,35,36]. Such a swimming style is practiced

222   by the extant manatees (*Trichechus* spp.), contrasting with the caudal oscillation seen in the dugong

223   (*Dugong dugon*), who is less affected by BMI and who occupies more open water habitats[37]. Extant

224   cetaceans' carangiform with lunate-tail swimming also involves such oscillations[38]. Extreme

225   pachyostosis would have restricted the movements between adjacent vertebrae, thus further

226   constraining the swimming style to low amplitude undulations[10]. Indeed, the extensive

227   anteroposterior widening of the transverse processes and neural spine that characterises all the

228   preserved vertebrae of *P. colossus* should have severely limited the flexural capabilities of the

229   vertebral column. To test this assumption, we followed the methodology of Molnar et al.[39],

230   evidencing that the maximum degrees of dorsal and -especially- lateral flexion were very low in *P.*

231   *colossus* compared to *Cynthiacetus peruvianus*; in turn, the capability of ventral flexion was instead

232   higher (Extended Data Fig. 9). This suggests that *P. colossus*, like *Antaecetus* and *Pachycetus*[15], swam

233   with a slow dorsoventral undulation of the posterior part of the body, without involving any degree

234   of mediolateral undulation as proposed instead for *Basilosaurus*[35,36,40]. A strong ventral flexion may

235   have helped to push the heavy body of *P. colossus* upwards from the seafloor to breathe at the

236   surface. Gigantism combined with the inferred undulatory swimming style of *P. colossus* and

237   *Basilosaurus* could represent an evolutionary adaptation analogous to that observed in Triassic

238   ichthyosaurs. Indeed, in these marine reptiles a giant size has been proposed to compensate for the

239   relatively high energetic cost of undulatory locomotion[41]. It is also noteworthy that a generalised

240   pachyostosis in the axial postcranium is otherwise found in much smaller (about 1 metre or less)

241   aquatic reptiles[42]. These taxa also purportedly used undulatory locomotion, as suggested by their

242   serpentiform body plan. There may thus be similar selective pressures resulting in the convergent

243   reduction of the limbs and acquisition of an elongate body with strong bone mass increase of the

244   axial postcranium in aquatic tetrapods. Gigantism could also have been selected to allow for longer

245   dives near the sea bottom in shallow waters. Indeed, dive duration in air-breathing vertebrates

246   displays a strong allometric relationship with body mass[43].

247   Since the skull and teeth of *P. colossus* are unknown, any hypothesis about its diet and

248   feeding strategy of this idiosyncratic early whale would be speculative. However, having

249   hypothesised slow swimming speeds as well as benthic habits in shallow and agitated waters

250   (Extended Data Fig. 10), and considering the huge quantity of food that would be needed to sustain

251   such a giant animal, several observations may be made regarding the feeding style of *P. colossus*.

252   Similar to sirenians, *P. colossus* may have fed on seagrass and/or seaweed, but that would make it

253   the sole herbivorous cetacean, a rather unlikely scenario. Alternatively, *P. colossus* may have fed on

254   benthic, sedentary or sessile fauna (e.g., crustaceans, demersal fishes, molluscs, etc.). Suction-

255   and/or filter-feeding strategies to forage benthic fauna could also be speculatively evocated, in

256   analogy with the trophic habits of the extant grey whale (*Eschrichtius robustus*). Finally, a further

257   conjectural hypothesis is that *P. colossus* fed on low-cost, high-energy food sources by means of

258   scavenging upon sunken vertebrate carcasses, similar to many extant large-bodied demersal sharks.

259   Unfortunately, the sedimentological data and the scarce associated fauna do not provide useful

260   information to better define the palaeoecology of *P. colossus*, its carcass having been probably

261   transported to relatively deeper waters, though not far away from the coast (Supplementary

262   Methods).

263

264   In summary, the morphology and microanatomy of *Perucetus colossus* extend the gamut of known

265   skeletal phenotypes, which in turn alters our understanding of mammalian maximum body mass

266   evolution. The order of magnitude of 10^5 kg has not only been reached recently, in Neoceti (crown

267   group cetaceans), but likely during the Bartonian, an age of cetacean evolution characterised by mild

268   to extreme bone mass increase. The second half of the Eocene was also a time when a peak in

269 maximum body mass was already observed in other mammalian clades[44,45]. While uncertainties

270 remain regarding the precise lifestyle and diet of *P. colossus*, it was a slow-swimming, most likely

271 coastal, benthic consumer which further suggests that the pelagic adaptations of Neoceti (and other

272 large marine tetrapods) are not a prerequisite to reach such large body masses. The ecological

273 specialisation associated with the extreme skeletal morphology of *P. colossus* indicates some further

274 degree of niche expansion[46] for Eocene cetaceans, in relation to the reduced gravitational constraints

275 on size due to life in the water[47] and the high seafloor productivity at that time[48]. This new record

276 supports the hypothesis that basilosaurids have hyper-specialised to coastal habitats during the end

277 of the Eocene, and that the subsequent major drop in the productivity of these environments[48] may

278 have preferentially impacted these whales, giving way for their relatives (the ancestors of nowadays'

279 whales and dolphins) who invaded more offshore habitats[33].

280

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478

Figure legends

Fig. 1 | Partial skeleton of *Perucetus colossus* MUSM 3248, holotype. a. Schematic body and skeletal reconstruction reporting the preserved bones (red). **b,c**. Right rib in anterior (**b**) and lateral (**c**) views. **d**-**f**. Right innominate in medial (**d**), dorsal (**e**), and lateral (**f**) views. **g-i**. Last preserved lumbar vertebra (L-k) in anterior (**g**), left lateral (**h**), and dorsal (**i**) views. **k-l**. Articulated sequence of the 13 collected vertebrae in dorsal (**j**), ventral (**k**), and right lateral (**l**) views. Scale bars, 50 cm (b,c,g-l) and 10 cm (d,e,f). All images were generated from 3D surface models.

Fig. 2 | Pachyostosis in *Perucetus colossus* MUSM 3248, holotype. a, b. Skeleton (**a**, preserved bones in dark grey), life reconstruction (**b**), and 3D models of a rib (**c**) and vertebra (L-k) of *P. colossus*. Skeletons (**e**, **h**), posterior rib (**f**, **i**) and last lumbar vertebra (**g**, **j**) of the Wexford blue whale (*Balaenoptera musculus*; **e-g**) and another basilosaurid, *Cynthiacetus peruvianus* (**h-j**). Scale bar for isolated bones, 50 cm; for skeletons and reconstruction, 2 m.

Fig. 3 | Bone histology of *Perucetus colossus* MUSM 3248, holotype. a. half rib cross-section (in three parts), featuring very sparse porosity and multiple growth marks (arrows). **b.** Detail of the above, showing a highly convoluted, woven-parallel complex, devoid of remodelling (left, natural light; right, cross-polarized light). **c**. Broken peduncle of a lumbar vertebra in ventral view (note fractures following growth lines), made of compact bone throughout. Small inset (vertebra parts in anterior view, not to scale) indicates position of the illustrated break surface. Scale bars, 1 cm (a), 0.5 mm (b), and 5 cm (c).

Fig. 4 | Scaling of the skeletal mass across amniotes' body mass range. Regressions based on extant, terrestrial mammals (grey line) and extant cetaceans (blue line). On the same graph are also plotted estimated values for extreme terrestrial amniotes, sirenians (either measured or estimated) as well as the result of our estimations for *Perucetus colossus*. Each data point corresponds to a distinct species (specific mean for the largest extant cetaceans, *Balaenoptera musculus* and *Physeter macrocephalus*), except for extant sirenians (represented by two species). Horizontal and vertical bars indicate maximum and minimum estimates (some are too small to be visible). Scales are logarithmic. Human height, 1.8 m (scale for larger silhouettes); penny's diameter, 2 cm. See Supplementary Methods and Supplementary Data 2.

511 **Methods**

512 **Geological age of *Perucetus colossus***

513 A stratigraphic section was measured at the type locality, starting from the base of the outcrop,

514 which is locally marked by two dolomite layers within the Yumaque member of the Paracas

515 Formation. The lower 18.5 m of the section are made of silt that transition upward to fine silt-clay.

516 The base of the Otuma Formation occurs at 34.5 m; it is followed by two ash layers interbedded with

517 fine silt-clay (Extended Data Fig. 5). The holotype of *P. colossus* occurs *s*ome metres below the basal

518 Otuma strata, ca. 30 m above the base of the section.

519 Samples for biostratigraphic analyses were prepared as standard smear-slides and analysed with an

520 Olympus BX50 polarised light microscope at 1000× with immersion oil. The identification of

521 calcareous nannofossils followed the taxonomy of Perch-Nielsen[49] and Young et al.[50]. Zonal schemes

522 are based on Martini[51], with zonal codes NP, and Agnini et al.[52], with zonal codes CNE.

523 For the ^{39}Ar–^{40}Ar dating analyses, a 10-cm-thick volcanic ash layer was sampled ca. 5 m above the

524 level of the fossil specimen (Extended Data Fig. 5), avoiding weathered and extraneous particles.

525 The collected tephra was petrographically analysed under a stereomicroscope and a Leica optical

526 microscope after preparing smear slides. Grain-size analyses were performed with a Malvern

527 Mastersizer 2000E™Laser Granulometer and data were processed with the grain-size analysis

528 program GRADISTAT 4.0[53] at the Università degli Studi di Milano-Bicocca. The ash sample was wet-

529 sieved through meshes of 500, 250, 125, and 63 μm, and the 250–500 μm fraction was mounted in

530 resin and polished for compositional analyses through a JEOL 8200 Superprobe™ at the Università

531 degli Studi di Milano Statale, with 15 kV accelerating voltage and 5 nA beam current. The beam

532 diameter was 3 μm for biotite phenocrysts and 10 μm for glass shards. Finally, biotite phenocrysts

533 were collected by handpicking and irradiated in the McMaster University nuclear reactor. They were

534 analysed through the NuInstruments™ Noblesse® noble gas mass spectrometer at the Università

535 degli Studi di Milano-Bicocca by stepwise heating, following Bosio et al.[54]. See Supplementary

536   Methods for details on the lithostratigraphy, biostratigraphy, palaeoenvironmental setting and ^{39}Ar–

537   ^{40}Ar dating analyses.

538

Vertebral proportions in Cetacea

540   Centrum length and width of the thoracic and lumbar vertebrae were compiled for extant[55] and

541   extinct cetaceans (Supplementary Data 3). The mean and maximum values of the length/width ratio

542   (CL/CW) were then compared to those of *P. colossus* (Extended Data Figure 6; R[56] package ggplot2[57]).

543

Bone microanatomy and histology

545   One of the recovered ribs and several vertebrae were sampled to produce palaeohistological

546   thin-sections, following standard procedures[58,59]. A half cross-section was made around the rib's mid-

547   length. For the vertebrae, we used core drillings to sample the centrum (in both the anteroposterior

548   and dorsoventral directions), neural spine, and transverse processes (see also Supplementary

549   Methods). Break surfaces were opportunistically observed for all recovered elements.

550   The obtained thin-sections were observed and photographed with a petrographic

551   microscope (Leica DM750P) equipped with a camera (Leica ICC50 W) under natural and cross-

552   polarized light for detailed views. A Keyence system (VH-Z20R objective) was used to capture general

553   views of the sections. To measure the global compactness of each section, images were thresholded

554   manually (so that bone tissue represents the foreground, and the rest the background). Compactness

555   was then measured as the ratio between the foreground and background areas within a selection

556   corresponding to the whole cross-sectional area ('Measure' routine of Fiji/ImageJ[60,61]; Supplementary

557   Table 2).

558   Extensive comparative material for rib cross-sections has already been published[5,13,14,62,63].

559   For the microanatomy of the vertebra, the regions sampled in the new specimen were examined in

23

560   extant cetaceans and sirenians through virtual thin-sections (Extended Data Fig. 7; Supplementary

561   Table 2) using high resolutions CT-scans from existing datasets[64,65]. These sections were produced

562   using a rectangular region of interest (ROI) corresponding to the regions sampled on the fossils;

563   these ROIs were extracted, thresholded, and their bone compactness was measured with

564   Fiji/ImageJ[60,61] ['Rotate', 'Orthogonal views, 'Threshold' (with stack histogram option), 'Measure',

565   routines]. The longer side of the VOI was defined from the periosteal surface to the middle of the

566   region (e.g., mid-length of the centrum); its shorter side was set proportionally to the diameter of

567   core drilled in the fossil, i.e., ca. 7.2% of the centrum's height.

568

569   **Skeleton and whole body mass estimations**

570   Dry skeletal mass (SM) is here understood as the bone tissue, excluding the free water it can

571   contain (but including more tightly bound water[66]) as well as the adipose tissues. Skeletal fraction

572   (SF) is defined as the ratio between SM and total body mass (BM). Several methods of SM

573   estimations used herein will rely on compact bone density, which can vary from one species/location

574   to another. Here we used existing data for a terrestrial mammal (*Panthera leo*), a cetacean

575   (*Delphinus delphis*), and a sirenian (*Dugong dugon*)[67,68]. Regressing global compactness against bone

576   density, one can estimate (lm and predict functions, base of R) the mean dry bone tissue densities

577   (for bones of the whole skeleton) of these three animals (1.89 g.cm^{-3}, 2.07 g.cm^{-3}, and 2.12 g.cm^{-3},

578   respectively).

579   We used different methods to assess skeletal mass across a dataset that includes both extant

580   and extinct amniotes. These methods include direct weighting of the skeletons (extant terrestrial

581   mammals and some marine mammals), extrapolations based on SF for large cetaceans, based on

582   whole skeleton volume for sauropods, extremely small amniotes, extinct sirenians, and large, extinct

583   mammals. See Supplementary Methods and Supplementary Tables 6-16 for details.

584   In addition to direct body mass measurements, body masses were estimated using body

585   measurement regressions (large cetaceans, small amniotes, sirenians), published estimations

586   (sauropods, extinct terrestrial mammals), or taken from AnAge[69] (for the Etruscan shrew and for the

587   mean values of large cetaceans). See Supplementary Methods and Supplementary Tables 6, 9, and 12

588   for details.

589

590   **Skeletal mass ~ body mass scaling**

591

592   Two distinct scaling relationships were assessed, one for extant, terrestrial mammals, and one for

593   extant cetaceans (Supplementary Tables 2, 3). Skeleton data of Prange et al[70], completed for species

594   of BM ranging from 62 to 290 kg with newly acquired measurements. These new data are derived

595   from wild-caught museum specimens for which body mass is known, and for which the dry skeletons

596   were weighted with a digital scale (Supplementary Data 2). The extant cetacean data were taken

597   from Buffrénil et al.[71], Buffrénil[72], and Robineau and Buffrénil[73] (for the *Delphinus delphis* sample only

598   mature specimens were taken, i.e., with at least ten growth marks; see [71]).

599   We examined the scaling of dry skeletal mass against whole body mass through phylogenetically

600   informed linear regressions. These regressions account for the phylogenetic relatedness of

601   observations through the determination of the optimised Pagel's lambda value (bounded to [0;1]) of

602   the regression (*gls* function, nlme package[74], *corPagel* function, ape package[75]. As timetree, we used

603   the node-dated, maximum clade credibility tree from Upham et al.[76]. Sampled species were swapped

604   with closest relatives when not already present in the tree. Pseudo-R^2 values were computed with

605   the *rsquared* function (piecewiseSEM package[77]). Data were plotted with R[56], packages ggplot2[57] and

606   viridis[78]

607

P. colossus' skeletal and body masses

The skeleton volume of *P. colossus* was estimated based on surface scans of the holotype and more complete basilosaurid skeletons. The 3D surface textured models of the holotype were created using a SHINING EinScan Pro HD handheld scanner (EXScan Pro 3.7.0.3 software) for the vertebrae and an Artec Eva scanner (Artec Studio 15 Professional software) for the rib and innominate (both at the highest resolution: 0.25 mm and 0.5 mm, respectively). The volume of each model was measured (Blender 3.0.1[79]), substracting that of the sediment still filling the neural canal of some vertebrae. In addition to being used in the following steps, the vertebrae' volume was compared to those of the Wexford blue whale (NHMUK-ZD.1892.3.1.1) and to a cast of a posterior thoracic/lumbar vertebra of *Basilosaurus cetoides* (Liège University, made with an Artec Spider surface scanner).

To estimate the volume of the unpreserved skeletal elements, we used as a base the most complete and best preserved skeleton of basilosaurid, that of *Cynthiacetus peruvianus*' holotype (MNHN.F.PRU10; surface scanned with an Artec Eva scanner, Artec Studio 15 Professional software). We made several estimations based on this skeleton: 1. Scaling up and dilating *C. peruvianus*' skeleton for the corresponding bone to reach the volume of those of *P. colossus'* holotype (Extended Data Fig. 10). Because the precise position of the recovered skeletal elements is unknown, 10,000 random draws were made to select corresponding vertebrae in the posterior thoracic and lumbar regions. For the rib cage, two alternatives were assessed, considering the digitised rib of *P. colossus* to correspond to the anterior-most rib with simple proximal end morphology (R17) or last rib (R20). 2. Adapting the scaled-up, dilated models based on *C. peruvianus* to fit the skeleton composition of two other well-known basilosaurids, *Basilosaurus isis*[35] and *Dorudon atrox*[17]; a conservative skeleton composition was also used, assuming minimum counts of 12 thoracic and 15 lumbar vertebrae. See Supplementary Methods and Supplementary Tables 17 and 18 for details. Skeletal length was taken from the scaled-up models (the pachyostosis does not affect the vertebral centra anteroposteriorly). The models of *P. colossus* and *C. peruvianus* used for the estimations are accessible on MorphoMuseum[80].

634   The skeletal mass was assessed using the minimum and maximum obtained volumes and the

635   skeletal elements' density, which derived from the histological samples made on *P. colossus'*

636   holotype (vertebrae and ribs, which represent over 97% of the skeleton's volume) or an extant

637   cetacean, *Delphinus delphis* (rest of the skeleton; Supplementary Methods and Supplementary

638   Tables 19 and 20).

639   Finally, the body mass of *P. colossus* was estimated based on the skeletal fractions (SF;

640   skeletal mass to whole body mass ratio) of extant species. These skeletal fractions inform on the

641   viable proportion of tissues in these animals, and overall body density is particularly critical for

642   aquatic vertebrates, especially amniotes who regularly have to surface to breathe. We have hence

643   used the two extreme values measured in extant, fully aquatic mammals: 2.24% *Mesoplodon*

644   *europaeus*[73] and 6.25 % for *Trichechus manatus* (mean of adults from Domning & Buffrénil[26];

645   Supplementary Table 22), as well as the mean value for cetaceans, whose SF scaling differs from that

646   of other mammals.

647

648   **Range of motion of the vertebral column**

649   Using a methodological approach similar to that applied to crocodylomorphs by Molnar et al.[39], we

650   estimated the osteological range of motion in ventral flexion, dorsal extension and lateral flexion of

651   the preserved portion of vertebral column of *P. colossus* and of an equivalent vertebral portion of

652   column of *C. peruvianus*. To take into account also the effect on flexibility of the length of the

653   vertebral centra, we selected a portion of the vertebral column of *C. peruvianus* having the centrum

654   of the middle vertebra with the same width as that of the *P. colossus*. Thus, due to the shorter

655   centra, the number of vertebrae selected for *C. peruvianus* was greater (18) than those of *P. colossus*

656   (13). The osteological range of motion was estimated by manipulating the virtual 3D models of *P.*

657   *colossus* and C. *peruvianus* holotypes in Blender 3.0.1[79]. The missing parts of the 13 vertebrae of *P.*

658   *colossus* were virtually reconstructed by copying and adapting the respective preserved parts of the

659 adjacent vertebrae. Intervertebral spacing was estimated for both species using the intervertebral

660 discs' proportions of *Delphinus delphis*[81].

661

662 **Data availability**

663 All data generated or analysed during this study are included in this published article (and its

664 supplementary information files) as well as the following public repositories: Three-dimensional

665 surface models of the holotype of *P. colossus* as well as *Cynthiacetus peruvianus* MNHN.F.PRU10 are

666 available on MorphoMuseuM[80]; newly acquired CT data are available on MorphoSource

667 (https://doi.org/10.17602/M2/M510260). The existing database AnAge[69] was also used.

668 All nomenclatural acts from this work were recorded in Zoobank: *Perucetus,*

669 urn:lsid:zoobank.org:act:E5F92709-2F65-4C50-8F46-7C005F64CE03; *Perucetus colossus*

670 urn:lsid:zoobank.org:act:CD837E76-E7B8-4E06-8F87-54AFE7AFB211.

671

672 **Code availability statement**

673 The code run for this study is provided at https://github.com/eliamson/ColossalCode.

674 **References [for Methods]**

675

676 **Extended Data Figures**

677

678 **Extended Data Fig. 1 | Three vertebrae of *Perucetus colossus* (MUSM 3248 holotype). a, g, m.**
679 Anterior view. **b, h, n.** Posterior view. **c, i, o.** Dorsal view. **d, j, p.** Ventral view. **e, k, q.** Right lateral
680 view. **f, l, r.** Left lateral view. Tentative position along the vertebral column: **a-f** = Th-a, penultimate
681 thoracic; **g-l** = Th-b, last thoracic; **m-r** = L-a, first lumbar. All images were generated from three-
682 dimensional surface models. Scale bar = 50 cm.

683

684

Extended Data Fig. 2 | Three vertebrae of *Perucetus colossus* (MUSM 3248, holotype). a, g, m.
Anterior view. **b, h, n.,** Posterior view. **c, i, o.** Dorsal view. **d, j, p.** Ventral view. **e, k, q.** Right lateral
view. **f, l, r.** Left lateral view. Tentative position along the vertebral column: **a-f** = L-b, second lumbar;
g-l = L-c, third lumbar; **m-r** = L-d, fourth lumbar. All images were generated from three-dimensional
surface models. Scale bar = 50 cm.

690

Extended Data Fig. 3 | Three incomplete vertebrae of *Perucetus colossus* (MUSM 3248, holotype).
a, g, m. Anterior view. **b, h, n.** Posterior view. **c, i, o.** Dorsal view. **d, j, p.** Ventral view. **e, k, q.** Right
lateral view. **f, l, r.** Left lateral view. Tentative position along the vertebral column: **a-f** = L-e, fifth
lumbar; **g-l** = L-f, sixth lumbar; **m-r** = L-g, seventh lumbar. All images were generated from three-
dimensional surface models. Scale bar = 50 cm.

696

Extended Data Fig. 4 | Two vertebrae (a-l) and three posterior ribs (m; R-b,c,d) of *Perucetus*
***colossus* (MUSM 3248, holotype). a, g.** Anterior view. **b, h.** Posterior view. **c, i.** Dorsal view. **d, j.**
Ventral view. **e, k.** Right lateral view. **f, l** Left lateral view**.** Tentative position along the vertebral
column: **a-f** = L-j, eleventh lumbar; **g-l** =L-k, twelfth lumbar. **a-l**. Images generated from three-
dimensional surface models. **m.** Photograph taken in the field. Scale bars = 50 cm.

702

Extended Data Fig. 5 | Location and stratigraphic position of *Perucetus colossus* MUSM 3248. a.
Map showing the position of the Coastal Batholith and major trench-parallel structural highs along
the coast of Peru (redrawn and modified after Travis et al.[82] and Thornburg & Kulm[83]). **b.**
Stratigraphic column of the Cenozoic succession exposed in the East Pisco Basin (redrawn and
modified after Malinverno et al.[84], Bianucci & Collareta[85], and Bosio et al.[86]). **c**. Measured
stratigraphic section of the type locality of *P. colossus*, indicating the stratigraphic height of the
holotype MUSM 3248, the dated tephra layer, and the identified bioevents.

710

Extended Data Fig. 6 | Vertebral and costal morphology of *Perucetus colossus* (MUSM 3248,
holotype) compared to that of other cetaceans. a, mean and maximum ratio of vertebral centrum
anteroposterior length (CL) to mediolateral width (CW), computed with the thoracic and lumbar
vertebrae of other cetaceans (Ba = Basilosaurinae; Dor = Dorudontinae; FM = fossil Mysticeti; FO =
fossil Odontoceti; P = Pakicetidae+Ambulocetidae; Pa = Pachycetinae; Pr = Protocetidae; R =
Remingtonocetidae). **b,** profile of vertebral centrum length in *P. colossus* (Gt) compared with the
profiles of other basilosaurids (Aa = *Antaecetus aithai*; Bc = *Basilosaurus cetoides*; Bc = *Basilosaurus
isis*; Cf = *Chrysocetus fouadassii* Cp = *Cynthiacetus peruvianus*; Da = *Dorudon atrox*; data of *A. aithai*
and *B. isis* from Gingerich et al.[15], fig. 11). **c,** curvature (length/chord) of the best preserved rib (red
horizontal line) compared to those of *Cynthiacetus peruvianus* (MNHN.F.PRU10).

721

Extended Data Fig. 7 | Vertebral microanatomy illustrated with binarized sections (black = bone) obtained with physical core drills (a) or µCT data (b-f). a, *Perucetus colossus* (MUSM 3248, holotype, vertebra L-e for the centrum and transverse process and L-c for the neural spine). The global compactness (Cg) was measured in ten areas (orange outlines) to assess overall centrum compactness (see Supplementary Methods). **b,** common minke whale (*Balaenoptera acutorostrata*, LR M 523). **c,** Amazon river dolphin (*Inia geoffrensis*, ZMB_Mam_41500). **d,** common dolphin (*Delphinus* sp. ZMB_Mam_697.59). **e,** dugong (*Dugong dugon,* ZMB_Mam_69340). **f,** manatee (*Trichechus manatus*, ZMB_Mam_17377). **(1)** Centrum (anteroposterior mid-length, from its dorsal edge to its centre), external towards top; **(2)** Neural spine (dorsoventral mid-height), external towards right); **(3)** Transverse process (mediolateral mid-width), external towards top. Width of the virtual cross-sections were defined as representing 7.2% of the centrum dorsoventral height; this is the mean ratio between the centrum height of *P. colossus* and the width of the physical core drills. The core drills of the neural spine and transverse process do not reach the middle of the corresponding vertebral parts (but break surfaces indicate uniform structure throughout). See core drills' location in Fig. S6b,c. Scale bars: a = 13.9 mm, b = 23.6 mm, c = 2.2 mm, d = 2.7 mm, e = 3.2 mm, f = 3.0 mm.

Extended Data Fig. 8 | Estimating the whole skeletal volume of *Perucetus colossus* based on Cy*nthiacetus peruvianus*' holotype (MNHN.F.PRU10). a. Unmodified 3D model of Cy*nthiacetus peruvianus*' holotype. **b, c.** vertebral column scaled-up (top) and dilated (bottom). **c, d.** Rib cage scaled-up (top) and dilated (bottom) identifying the scanned rib of *P. colossus* as R17 (**c**) or R20 (**d**). Scale bars = 2 m.

Extended Data Fig. 9 Estimates of the osteological range of motion. Extension and flexion of the preserved portion of vertebral column of *Perucetus colossus* holotype (MUSM 3248) is compared with an equivalent vertebral column portion of Cy*nthiacetus peruvianus* holotype (MNHN.F.PRU10) using the respective 3D models. Intervertebral spaces were reconstructed based on the common dolphin (*Delphinus delphis*)[81] (see Methods). Scale bar = 50 cm.

Extended Data Fig. 10 | Reconstruction of *Perucetus colossus* in its coastal habitat. Because portions of the skeleton are unknown, several aspects of the reconstruction are tentative: the overall proportions of the axial postcranium are based on a close relative *Cynthiacetus peruvianus*, which was scaled-up and dilated according to the elements recovered for *P. colossus* (see Extended Data Fig. 8); the skull and limbs were only scaled-up; the tail fluke and forelimb use (bottom-walking) are based on the manatee (*Trichechus*), the extant marine mammal with the closest degree of pachyosteosclerosis in the postcranial skeleton; the hind limb of *P. colossus* was not recovered, but the anatomy of its innominate indicates the presence of a reduced, articulated leg. The associated sawfish (*Pristis*) was recovered from the same unit in the East Pisco Basin, the Yumaque Member of the Paracas Formation[87]. Reconstruction by A. Gennari.

Authors contribution

M.U. discovered and collected the fossil. E.A., G.Bi., and O.L. conceived and designed the project. G.Bo., A.C., C.Di C., E.M., I.M.V., and P.P.P. collected and analysed the stratigraphical data and wrote the corresponding methods sections. A.B., A.C., E.A., G.Bi., M.M., O.L., R.B., and R.S.-G., collected

786   phenotypic data (including CT and/or surface scans and/or palaeohistological samples). E.A., G.Bi.,

787   and O.L. analysed the phenotypic data. E.A., G.Bi., and O.L. wrote the first draft of the manuscript. All

788   authors discussed the analyses and reviewed the manuscript.

789

790   **Competing interests**

791   The authors declare no competing interests.

792   **Material and correspondence**

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