

1 **Molar wear in house mice: insight into diet preferences at an ecological time scale?**

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21 **Running head**

22 Molar wear in house mice

23

24 **Abstract**

25 In molars without permanent eruption, wear deeply modifies the geometry of the crown. To test for  
26 a signature of diet on wear dynamics, the molar geometry was compared between commensal house  
27 mice, relying on an omnivorous-granivorous diet, and Sub-Antarctic relatives, characterized by a  
28 switch towards a more “predator” behavior. Lab-bred offspring of commensal mice served as  
29 reference by providing mice of known age. Molar geometry was quantified using dense 3D semi-  
30 landmarks based descriptors of the whole molar row and the upper molar only.

31 Lab offspring displayed decreased rate of wear compared to their commensal relatives, due to  
32 reduced mastication in mice fed *ad libitum*. Sub-Antarctic mice displayed a similarly decreased rate  
33 of molar wear, in agreement with an optimization towards incisor biting to seize preys. Lab offspring  
34 and Sub-Antarctic mice were further characterized by straight molar rows, whereas in commensal  
35 mice, the erupting third molar was deviated away from the longitudinal alignment with the other  
36 molars, due to masticatory loadings.

37 Quantifying changes in molar geometry could thus contribute to trace subtle diet variations, and  
38 provide a direct insight into the constraints during mastication, shedding light on the functional role  
39 of adaptive changes in molar geometry.

40

41

42 **Keywords**

43 Geometric morphometrics – Kerguelen Archipelago – Mastication – Molar crown geometry – *Mus*  
44 *musculus domesticus* – Occlusal relief – Subantartic environment.

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## 48 Introduction

49 Mammalian teeth are very diverse in their morphology, allowing the consumption of various food  
50 items. As such, the evolution of the mammalian dental pattern participated to the radiation of the  
51 group (Hunter & Jernvall, 1995; Grossnickle et al., 2019), leaving both a phylogenetic (Cucchi et al.,  
52 2017) and an ecological signal (Gómez Cano et al., 2013) on tooth morphology. In teeth without  
53 permanent eruption, wear is the only factor modifying this geometry after dental eruption. Being the  
54 consequence of both abrasion, due to the contact of the tooth with the food items, and attrition, due  
55 to tooth-tooth contact, wear considerably affects the crown geometry. As a consequence, wear can  
56 be considered as being problematic by overprinting the original morphology and thus obliterating  
57 other signals, including genetic differences in tooth shape (Ledevin et al., 2016; Pallares et al., 2017).  
58 Conversely, wear depends on masticatory behavior and on the food items consumed along an  
59 animal's life (Teaford & Oyen, 1989) and has thus the potential to trace ecological variations on a  
60 very short time scale. Teeth are often the only fossil remain for small mammals such as rodents.  
61 Retrieving fine-scale ecological information from their geometry could be precious for understanding  
62 the selective pressure acting on their evolution (Gomes Rodrigues et al., 2013).

63 Among rodents, the house mouse (*Mus musculus domesticus*) has been associated with humans  
64 even before the Neolithic (Weissbrod et al., 2017). Being unintentional fellow-travelers of human  
65 exchanges allowed house mice to be one of the most successful invader worldwide (Cucchi, 2008;  
66 Jones et al., 2012). As such, mice have been confronted to a variety of diets depending on local  
67 resources (Miller & Webb; Le Roux et al., 2002). Being also easily bred in laboratory condition, the  
68 house mouse constitutes a good model to test for signature of diet differences in both wild and  
69 experimental conditions.

70 Experiments on a laboratory strain demonstrated that hard and soft eaters differed in the rate of  
71 molar wear (Renaud & Ledevin, 2017). Comparison between mice trapped in the wild and relatives  
72 bred in the lab further showed that the alignment of the molars on the jaw is impacted by loadings  
73 during mastication, with wild mice showing a torsion of the molar row not observed in their lab  
74 offspring (Savriama et al., 2022). Lab mice being bred with unlimited access to food, they presumably  
75 chewed less thoroughly than wild mice confronted to the cost of foraging. If so, wild populations  
76 differing in diet may display similar differences in wear trajectories and molar insertion. The present  
77 study therefore focuses on the comparison between commensal mice and Sub-Antarctic populations  
78 from the Kerguelen archipelago. In such Sub-Antarctic environments, mice shifted their diet from  
79 their usual omnivorous-granivorous diet to a larger proportion of terrestrial animal prey (Le Roux et  
80 al., 2002). This triggered an optimization of their jaw morphology for incisor biting, crucial for seizing

81 prey (Renaud et al., 2015b). Functional performance for molar mastication was concomitantly  
82 decreased, suggesting a relaxation of the functional demand compared with commensal mice  
83 feeding on anthropic resources, especially grains in agricultural settings.

84 The geometric signature of wear, and more generally of use, on the molar system may thus have the  
85 potential to trace fine-scale differences in diet between populations. Commensal mice from different  
86 populations should share similar wear rates. In Sub-Antarctic mice, for which incisor biting is  
87 predominant and masticatory loadings reduced, rate of wear and torsion of the molar row should be  
88 reduced, similarly to what is observed in lab-bred mice.

89 The signature of wear on the molar system has been quantified using a dense 3D quantification of  
90 the molar morphology (Renaud et al., 2018b; Savriama et al., 2022) on three complementary  
91 descriptors: the complete molar row (UMR), the first molar (UM1) and a truncated template of the  
92 UM1 (UM1tr), corresponding to the part of the crown affected by wear only late in life. Considering  
93 the truncated UM1 should allow to focus on genetic differences between populations (Ledevin et al.,  
94 2016; Pallares et al., 2017). The analysis of the complete crown of the first upper molar provides  
95 information relative to wear, whereas the geometry of the molar row further includes the relative  
96 arrangement of the three molars along the row. It may also vary depending on the relative size of the  
97 three molars. Molars develop according to a developmental cascade, the first molar inhibiting the  
98 subsequent ones. The increase in relative size of the first molar to the detriment of the third molar  
99 has been suggested as a signature of faunivorous diet (Kavanagh et al., 2007). This component of  
100 geometric variation of the molar row has therefore been investigated as well.

101

## 102 **Material**

103 The material is composed of three sets of the Western European house mouse subspecies (*Mus m.*  
104 *domesticus*) (Supp. Fig. 1; Supp. Table 1).

105 (1) Commensal house mice were documented by two French populations. Nineteen mice were  
106 trapped in a horse stable in Balan, nearby Lyon (group later on designed as Balan Wild). A second set  
107 of ten commensal house mice was trapped in two neighboring farms in Tournay (Brittany, France)  
108 (Renaud et al., 2017).

109 (2) Non-commensal Sub-Antarctic mice were sampled on the small Guillou Island (Kerguelen  
110 Archipelago, Indian Ocean), including nine mice from 1993 and eleven mice from 2009 (Renaud et al.,  
111 2015b). This sampling brackets a period of important human-driven environmental modifications on  
112 Guillou Island that changed the resources available to the house mice. The rabbit (*Oryctolagus*

113 *cuniculus*) constituted an important competitor until its eradication by poisoning in 1994 (Chapuis et  
114 al., 2001). During the subsequent recovery from rabbit grazing, invasive plants progressed against  
115 native vegetation in relation to climate change (Chapuis et al., 2004). Invertebrate prey constitutes  
116 an important component in the diet of Guillou mice (Le Roux et al., 2002), but earthworm availability  
117 in the litter decreased over the period due to increasing summer drought (Lebouvier et al., 2002)  
118 whereas native insects regressed due to the spread of an invasive carabid predator (*Merizodus*  
119 *soledadenus*) (Laparie et al., 2010).

120 (3) Lab-bred mice corresponded to offspring of mice trapped in Balan and brought alive to the animal  
121 facility ACSED (Lyon University). After approximately two months of acclimation, the wild-trapped  
122 mice were paired to obtain F1 offspring that were bred with standard rodent pellets (SAFE A04), with  
123 food and water *ad libitum*. Some of these F1 mice were paired to obtained F2 descendants. Breeding  
124 was conducted in accordance with animal care guidelines. This dataset included 22 laboratory  
125 descendants ranging from one to four months of age, plus one young mouse at weaning (21 days of  
126 age). This group is designed as Balan Lab.

127 Body weight data were available for all mice. Age and sex were available for all Balan Lab mice,  
128 except sex for the young mouse at 21 days.

129 All mice were killed according to the directive 2010/63/UE of the European Parliament on the  
130 protection of animals used for scientific purposes.

131

## 132 **Methods**

### 133 *Acquisition and extraction of 3D surfaces*

134 Most skulls were scanned at a cubic voxel resolution of 12  $\mu\text{m}$  using similar settings. Wild mice from  
135 Balan, Tournon and most laboratory offspring were scanned on the General Electric (GE) Nanotom  
136 microtomograph ( $\mu\text{CT}$ ) of the AniRA-ImmOs platform of the SFR Biosciences, Ecole Normale  
137 Supérieure (Lyon, France). Skulls from Guillou were scanned at the PACEA laboratory (Bordeaux,  
138 France) using a similar equipment (GE v tome x s) at a resolution of 12  $\mu\text{m}$ . The dataset was  
139 complemented by one Balan Wild scanned at 12  $\mu\text{m}$  and eight Balan Lab scanned at 17  $\mu\text{m}$  at the  
140 Mateis laboratory (INSA, Lyon, France), using a similar equipment.

141 For each mouse, the right upper molar row (UMR) was segmented using Avizo (v. 7.1—Visualization  
142 Science Group, FEI Company). In many cases, an automatic threshold was sufficient to isolate the  
143 molar row from the surrounding bone and generate a surface including the roots, but in some cases,

144 connections with the bone had to be manually delimited. Starting from the model of the complete  
145 upper molar row, the first upper molar was manually delimited by removing the contact with the  
146 second molar. In one case (G93-24), the right molar row was damaged and the mirror image of the  
147 left molar row was considered.

148 The 3D surfaces of seven Balan wild mice and 13 lab offspring are available from a previous study  
149 (Renaud et al., 2021). The molar row surfaces corresponding to Tournich and Guillou have been further  
150 deposited in MorphoMuseum (Renaud et al., 2023). Information about the scanned specimens can  
151 be found in Supp. Table 1.

152

153 *Estimates of body size: Body weight and mandible size*

154 Body size. - Body weight was available for all mice, providing an estimate of growth stage. Since  
155 weight is more related to volume than to linear size, its cubic root was considered in all analyses.

156 Mandible size. - For each mouse, a 3D model of the mandible was segmented, in most case  
157 considering the right mandible. The 3D length of the mandible was measured as the distance  
158 between two landmarks located at posteriormost extremity of the condyle and the anteriormost  
159 edge of the mandibular bone along the incisor lingual side. This measure was selected because it can  
160 be assessed even on damaged mandibles, and because mandible size has been shown to display very  
161 little sexual dimorphism and to be well correlated to body size (head + body length) (Renaud et al.,  
162 2017). Landmarks were positioned using MorphoDig (Lebrun, 2018).

163

164 *3D surfacic description of the molars*

165 Mouse molars are composed of cusps arranged in transverse chevrons (Fig. 1) that are used as rasps  
166 to grind food items. Labial, central and lingual cusps align to form three longitudinal rows that guide  
167 the forwardly directed motion (propalinal movement) during chewing. This complex geometry can be  
168 described using surfacic templates covering the erupted part of the teeth (Ledevin et al., 2016;  
169 Renaud & Ledevin, 2017). Three templates were considered, corresponding to (1) the upper molar  
170 row (UMR), with the three molars in contact; (2) the first upper molar (UM1) only and (3) a truncated  
171 “wear-free” template of the UM1 (UM1tr). These templates, used in a previous study (Savriama et  
172 al., 2022), have been empirically elaborated on a specimen of intermediate age (Balan Lab #86).  
173 “Wear-free” templates, designed on other reference molars, were successful in discarding wear  
174 effect to focus on genetic differences in various contexts (Ledevin et al., 2016; Pallares et al., 2017).

175 The young mouse at 21 days could not be included in the analysis of the molar row, since the third  
176 molar, erupting at weaning, was not yet in contact with the second one.

177 The templates were used to extract cropped surfaces limited to the zone of interest. Fixed landmarks  
178 were used to guide the application of the template on each of the initial surfaces. Five landmarks  
179 were located on the UM1, one at the anterior front of the molar, two in lateral valleys (between  
180 cusps t1 and t4 on the lingual side, and between t3 and t6 on the labial side (see Fig. 1 for  
181 nomenclature of the cusps) and two in the valleys bracketing the median cusps t5. Being located low  
182 on the crown, they could be used both for the complete UM1 and the truncated template. Six  
183 additional landmarks (three on the second molar and three on the third molar) were collected to  
184 anchor the template of the molar row. Fixed landmarks were manually positioned using MorphoDig  
185 (Lebrun, 2018). Sets of equally spaced sliding semi-landmarks were then projected on the cropped  
186 surfaces using the package Rvcg (Schlager, 2017), leading to 2186 semi-landmarks for the UMR, 2199  
187 for the UM1 and 2293 for the UM1tr (Fig. 1) (Savriama et al., 2022).

188 The sliding procedure for these semi-landmarks was done using the minimum bending energy  
189 criterion (Bookstein, 1997). A generalized Procrustes analysis (GPA) was applied to the semi-  
190 landmarks, leading to shape descriptors (the aligned coordinates) by removing the effects of location,  
191 orientation, and position. The sliding procedure and the GPA were performed using the Morpho  
192 package (Schlager, 2017).

193

#### 194 *Relative size of the first and third molars*

195 The 3D area or volume of molars includes the degree of abrasion of the cusps. The relative area of  
196 the first vs third molar was thus evaluated on 2D projections of the occlusal surface, that are less  
197 impacted by wear. The cropped surfaces of the molar row were oriented with the occlusal surface  
198 facing up. On snapshots of this view, the first, second and third molars were manually delineated.  
199 The areas of the three molars were then automatically extracted using the image analyzing software  
200 Optimas. The first / third molar area ratio estimated the relative size of the two teeth.

201

#### 202 *Statistical analyses*

203 Regarding univariate estimators, differences between groups (Balan Wild, Balan Lab, Tournich, Guillou  
204 1993, and Guillou 2009) were tested using non-parametric Kruskal–Wallis tests complemented by  
205 pairwise Wilcoxon tests. Linear regressions and Pearson correlations were used to test for

206 covariation between variables. Analyses of variance (ANOVA) were used to test for the effect of age  
207 and sex on size estimators in the Balan Lab group. Similarly, Procrustes ANOVAs were used to  
208 investigate the effect of age and sex on the molar geometry within the Balan Lab group. Since no  
209 effect of sex was documented on molar geometry, and because sexual dimorphism has been  
210 repeatedly shown to be absent or reduced when considering molar size and shape in house mice  
211 (Valenzuela-Lamas et al., 2011; Renaud et al., 2017), males and females were pooled in all  
212 subsequent analyses.

213 The shape information was first summarized by Principal Component Analyses (PCA) applied to  
214 variance-covariance matrix of the aligned coordinates using the package Morpho. Differences  
215 between the five groups were tested using permutational MANOVA performed on the Procrustes  
216 distances (Procrustes ANOVA) using the package geomorph (Adams & Otárola-Castillo, 2013).  
217 Relationship between shape (aligned coordinates) and proxies of body size were investigated using  
218 Procrustes ANOVA using a model including size, group and their interaction. In all cases, probabilities  
219 were based on 9999 permutations. These analyses also provided a regression score summarizing the  
220 shape variance along the factors of the model; reconstruction of extreme shapes along the  
221 regression scores were performed using geomorph.

222 The PCA analyses the total variance in a sample ( $\mathbf{T}$ ). This variance can be decomposed in two  
223 components: the between-group matrix  $\mathbf{B}$  and the within-group matrix  $\mathbf{W}$ . A between-group PCA  
224 corresponds to the eigenanalysis of  $\mathbf{B}$  whereas a within-group PCA corresponds to the analysis of  $\mathbf{W}$   
225 that equals  $\mathbf{T}-\mathbf{B}$  (Dolédéc & Chessel, 1987). These analyses are implemented in the package ade4  
226 (Thioulouse et al., 2018) and were used to focus on between-group differences on the one hand, and  
227 on the pattern of within-group variance on the other hand. However, between-group PCAs (later  
228 bgPCA) are sensitive to problems of “over-fitting” when too many variables are included compared  
229 to the number of specimens, leading to representations where groups appear much more distinct  
230 than they really are (Cardini et al., 2019; Thioulouse et al., 2021). To insure a realistic representation  
231 of the differentiation between groups, cross-validated factor maps were considered, validated by  
232 permutational tests of between-group differences (randtest, 9999 permutations); the index of  
233 spuriousness  $\Delta O_{ij}$  was further evaluated (Thioulouse et al., 2021).

234 A multivariate regression was used to remove the signal attributable to genetic differences from the  
235 UMR and UM1 variation. The scores on the first two axes of the PCA on the truncated UM1 were  
236 considered to summarize the shape differentiation from genetic origin; the aligned coordinates of  
237 the UMR and of the UM1 were regressed on these two UM1tr PC axes to obtained residuals

238 coordinates that should only contain information relevant to non-genetic variation (wear for the  
239 UM1 and UMR, cumulated to the relative arrangement of the molars for the UMR).

240 Finally, the relationship between inter-individual multivariate topologies based on the UM1 and the  
241 UMR was assessed using a coinertia analysis. This method evaluates the concordance between two  
242 PCAs in a multivariate way, by finding orthogonal vectors (i.e., co-inertia axes) maximizing the sum of  
243 squared covariances between two datasets (Dolédec & Chessel, 1994) and allowing their projection  
244 in a common space. This analysis was performed using ade4 (Thioulouse et al., 2018).

245 All analyses were performed under R (R Core Team, 2018).

246

## 247 **Results**

### 248 *Variation in 3D molar geometry*

249 3D geometry of the upper molar row (UMR). – The morphospace corresponding to the upper molar  
250 row (Fig. 2A) shows an important within-group variance and a differentiation between populations.  
251 The within-group variance is oblique along PC1 and PC2, but mostly expressed along PC1 (35.4% of  
252 total variance). This axis corresponds, towards PC1 positive values, to a flattening of the cusps due to  
253 increasing abrasion, and to an increasing curvature of the molar row, with the second and third  
254 molar departing from the alignment with the UM1. The differences between the wild populations of  
255 Balan Wild, Tournich and Guillou are expressed along PC2 (15.3%). This axis also describes some molar  
256 row curvature and features regarding basic cusp morphology, such as the relative development of  
257 the posterior labial cusp t9, more pronounced towards PC2 positive values.

258 Balan Wild mice display an important variation, from unworn to worn-down molar rows where the  
259 second and third molars have been pushed towards the lingual side. In contrast, Guillou mice display  
260 relatively straight and unworn molar rows, with a reduced variation in the degree of wear, to the  
261 exception of one mouse from 2009 displaying a worn-down morphology similar to the ones observed  
262 in Balan Wild. Tournich molars are intermediate in morphology and in amount of variation along PC1.  
263 Balan Lab displays a molar row morphology intermediate between Balan Wild and Guillou mice.

264 The UM1 / UM3 relative 2D area did not differ between populations ( $P = 0.6189$ ).

265

266 3D geometry of the first upper molar (UM1). – The analysis of the first upper molar (Fig. 2B) provided  
267 a pattern close to the one based on the complete molar row. The morphospace based on the first  
268 two PC axes shows an important within-group variation along PC1 (51.5%), running parallel in the

269 different populations and corresponding to a flattening of the cusps, culminating in the five molars  
270 with almost completely abraded cusps. In contrast with results based on the UMR, Balan Lab mice  
271 plot within the range of their wild relatives (Balan Wild). Guillou mice are differentiated along PC2  
272 (10.2%), due to their divergence in tooth morphology. The commensal population from Tournay  
273 appears as intermediate between Balan Wild and Guillou.

274

275 3D geometry of the “wear-free” first upper molar (truncated UM1). – The morphological variance  
276 appears to be less concentrated on PC1 (21.9% only), that does not describe within-group variance  
277 but the morphological difference between the three genetically distinct groups (Balan Wild and Lab,  
278 Tournay and Guillou) (Fig. 2C). The second axis (PC2, 13.6%) mostly corresponds to a pattern of within-  
279 group variation opposing molars with an extended vs short forepart. Despite using the truncated  
280 “wear-free” template, four out of five teeth with highly advanced wear appear as outliers in the  
281 morphospace, showing that the cusp geometry is too much erased to deliver relevant information  
282 regarding genetic differences.

283

#### 284 *Variation in body and molar size*

285 All populations displayed similar body weight<sup>1/3</sup> (Fig. 3A, Table 1) and differences are reduced  
286 regarding mandible length (Fig. 3B). The pattern was quite different regarding tooth size, with  
287 significant differences involving Guillou mice (Table 1). These mice tended to display smaller teeth  
288 than the Western European ones (Fig. 3C).

289 Mandible length increased with body weight<sup>1/3</sup> (Fig. 3D), with a significant effect of populations but  
290 no interaction (Weight<sup>1/3</sup> P < 0.0001, population P < 0.0001, interaction P = 0.327), meaning that the  
291 slopes were not different between populations. The relationship was shifted in Guillou 1993 (Fig. 3D),  
292 with smaller mandibles than expected for a given weight. Considering separate Pearson correlation  
293 per population, the relationship between mandible length and weight<sup>1/3</sup> was significant in Balan Wild,  
294 Balan Lab, Guillou 1993 and Guillou 2009 (p < 0.0001). Molar centroid size was in contrast not related  
295 to body weight<sup>1/3</sup> but differed between populations (Weight<sup>1/3</sup> P = 0.1411, population P < 0.0001,  
296 interaction P = 0.0733).

297

#### 298 *Characterizing wear trajectories*

299 In order to avoid issues due to missing data or outlying specimens, the mouse at weaning and the  
 300 five senescent mice were discarded, leading to a common sampling of 64 mice for the UMR, the UM1  
 301 and the UM1tr.

302 PCA were performed on these three reduced datasets (Fig. 4) and the variance explained by the  
 303 “population” factor was estimated using Procrustes ANOVA and between-group PCAs. Compared to  
 304 the initial sampling, the within-group variance is reduced (Fig. 4A) and remains expressed on PC1  
 305 only for the UM1. Difference between populations were highly significant ( $P = 0.0001$ ) and explained  
 306 ~30% of the total variance (% of variance related to between-group differences: UMR 32.7%, UM1  
 307 27.9%, UM1tr 29.5%; same percentages obtained using Procrustes ANOVA and bgPCAs).

308

309 Wear trajectories, age, sex, and body size in the laboratory population. - A first approach to identify  
 310 the role of wear in the geometric variation of the molars was to performed Procrustes ANOVA on the  
 311 aligned coordinates using a proxy of the animal’s growth as covariate. Such analyses provide  
 312 regression scores allowing to visualize the shape variation occurring within each group along the  
 313 animal’s growth (Figs. 5, 6). This method was first applied to the group Balan Lab alone, for which the  
 314 age of the mice was known. Effect of sex and covariation between age and body size proxies were  
 315 further investigated, taking advantage of the experimental design (Table 2, Fig. 5). Age but not sex  
 316 significantly affected molar geometry and proxies of body size, although sex appears to have a  
 317 marginal effect on body weight<sup>1/3</sup>, males reaching larger size than females in this sample (Fig. 5C).

318 The UMR geometry was highly influenced by age (Procrustes ANOVA of the aligned coordinates vs  
 319 age: effect = 15.2%,  $P = 0.0003$ ). However, the relationship was not linear, with a pronounced change  
 320 in the geometry of the molar row occurring soon after weaning (Fig. 5A). The change in the geometry  
 321 of the UM1 is more progressive (Fig. 5B), leading to a higher percentage of variance explained by age  
 322 (35.7%,  $P = 0.0001$ ). Similar results were obtained using the cubic root of body weight<sup>1/3</sup> (UMR:  
 323 17.5%,  $P = 0.0001$ ; UM1: 23.8%,  $P = 0.0001$ ) or mandible length (UMR: 19.7%,  $P = 0.0001$ , UM1: 26%,  
 324  $P = 0.0001$ ) instead of age, showing that these proxies can be used to describe wear trajectories in  
 325 wild populations, for which mouse age is unknown. However, body weight<sup>1/3</sup> (Fig. 5C) as mandible  
 326 length (Fig. 5D) do not display regular increases along age. Both display a rapid growth from weaning  
 327 until 40 – 60 days of age, then decelerating to almost reach a plateau.

328

329 Wear trajectories in the different populations. – Wear trajectories were then assessed in all  
 330 populations, using Procrustes ANOVA on the aligned coordinates vs a proxy of the animal’s growth as

331 covariate; results were overall similar when using body weight<sup>1/3</sup> or mandible length as proxy (Table  
 332 3). The impact of growth was highly significant ( $P < 0.001$ ) and explained ~7% of shape variation for  
 333 the upper molar row and the first upper molar. The relationships were not significant when  
 334 considering the truncated template of the UM1, confirming that wear effects were successfully  
 335 discarded. Models including growth proxy and population as factors indicated no interaction when  
 336 considering body weight<sup>1/3</sup>, and no or weak interaction when considering mandible length, showing  
 337 that the groups shared similar wear trajectories.

338 Wear trajectories were thus represented using body weight<sup>1/3</sup> as a proxy of age (Fig. 6). For the UMR  
 339 as the UM1, despite non-significant interaction term, the slope tended to be steeper in commensal  
 340 populations (Balan Wild, Tourch) than in Guillou mice; Balan Lab plotted together with the Sub-  
 341 Antarctic populations. UMR shape changes related to body weight<sup>1/3</sup> were represented separately in  
 342 a commensal and a Sub-Antarctic population, taking Balan Wild and Guillou 1993 as examples (Fig.  
 343 6A). In both populations, the shape variation associated with increasing body weight<sup>1/3</sup> corresponded  
 344 to an abrasion of the cusps, more pronounced along the lingual side. The lingual cusps being initially  
 345 higher than their labial counterparts, this differential abrasion tend to re-equilibrate the labial and  
 346 lingual sides of the crown. The component of molar row curvature is present in the wear trajectory of  
 347 Balan Wild, even in stages corresponding to the smallest animals; in contrast, the molar row from  
 348 Guillou remains straight whatever the life stage. The UM1 shape variation related to body weight<sup>1/3</sup>  
 349 corresponds to an abrasion of the cusps (Fig. 6B).

350

#### 351 *Wear effect as within-group variation*

352 An alternative approach was to decompose the total shape variance (Fig. 4) into its between- and  
 353 within-group components. Between-group differentiation was significant in all cases (randtest  $P =$   
 354 0.0001). Due to the important amount of between-group variation (bgPCA: UMR 32.7%; UM1 27.9%;  
 355 UM1tr 29.5%), the spurious-group effect induced by the high number of variables was moderate  
 356 (spuriousness index  $\Delta O_{ij}$  UM1 = 7.9%) to low ( $\Delta O_{ij}$  UMR = 3.2%;  $\Delta O_{ij}$  UM1tr = 2.1%). For the UMR  
 357 (Fig. 7A) as the UM1 (Fig. 7B), the bgPCA opposed the commensal population of Balan Wild to Sub-  
 358 Antarctic Guillou populations along a first bgPC axis representing ~70% of between-group variance,  
 359 with the commensal population of Tourch being intermediate. The main difference regarded the  
 360 position of Balan Lab, shifted away from their wild relatives for the UMR but plotting in the same  
 361 range of the morphospace for the UM1. The morphospace corresponding to the truncated UM1 (Fig.  
 362 7C) opposed Balan (Wild + Lab) to Guillou (1993 + 2009) along the first axis (bgPC1, 62.1%). The  
 363 population of Tourch was still intermediate along bgPC1 and differentiates along bgPC2 (23.4%).

364 The first axis of within-group variance was correlated to body weight<sup>1/3</sup> regarding the UMR (R =  
365 0.564, P < 0.0001) and the UM1 (R = 0.452, P = 0.0002) but not for the UM1tr (P = 0.8949). The  
366 resulting patterns were highly similar to those obtained with the regressions scores, with commensal  
367 populations (Balan Wild and Tournch) sharing a trajectory of higher wear for a given weight compared  
368 to Guillou mice (Fig. 7D, E). Balan Lab was close to the Sub-Antarctic trajectories. Scores along the  
369 first axis of within-group variance were highly correlated to regression scores for the UMR (R = 0.883,  
370 P < 0.0001) and the UM1 (R = 0.980, P < 0.0001), underlining that shape variation related to wear  
371 corresponded to the main component of shared within-group variance when the complete crown is  
372 considered.

373

#### 374 *Removing the “genetic” signal: regression against UM1tr*

375 A last approach was to consider that the analysis of the truncated UM1 summarized the genetically-  
376 driven shape differences among populations. A multivariate regression of the aligned coordinates of  
377 the UMR and of the UM1 was performed against the scores on the first two axes of the UM1tr PCA  
378 (Fig. 4C) to remove this “genetic” effect and concentrate on wear patterns. A PCA on the resulting  
379 residuals aligned coordinates showed, for the UMR, an overlap of Guillou 1993 with Balan Lab,  
380 whereas Guillou 2009 plotted with the two commensal populations Balan Wild and Tournch (Fig. 8A).

381 The first axis of the PCAs on the residuals coordinates was highly correlated to the regression scores  
382 (UMR R = 0.900, P < 0.0001; UM1 R = 0.969, P < 0.0001) and to the first within-group PC axes (UMR R  
383 = 0.990, P < 0.0001; UM1 R = 0.942, P < 0.0001).

384

#### 385 *Beyond wear only: Contrasting the response of the molar row and the first upper molar*

386 From the preceding results, the effect of wear could be characterized as shared patterns of within-  
387 group shape variation. This does not include all the “use-related” signal, however, since the analysis  
388 of the upper molar row shows between-group differences that are neither of genetic origin nor  
389 related to wear *sensu stricto*. To better characterize this non-genetic effect beyond wear, the  
390 topology obtained for the UMR (Fig. 4A) was compared to the reference topology of the UM1 (Fig.  
391 4B) using a coinertia analysis (Fig. 8B). Arrows show the difference in topology for a given specimen  
392 from the first to the second morphospace considered. Compared to the UM1 pattern, including  
393 wear, the UMR shows a divergence of Balan Wild away from their laboratory relatives (Balan Lab)  
394 and even further away from the Sub-Antarctic populations, while Balan Lab mice were shifted in the

395 direction of these Sub-Antarctic populations. UMR from Guillou 1993 tended to diverge away from all  
396 other populations, including their 2009 relatives.

397

## 398 **Discussion**

399 The three-dimensional morphometric analyses of the upper molars showed that the impact of wear  
400 was massive and pervasive, when considering the complete molar row as well as when focusing on  
401 the first upper molar. Molar wear includes two aspects: attrition due to tooth-tooth contact, and  
402 abrasion *sensu stricto* due to the contact of the tooth with food items. Attrition may vary with the  
403 masticatory behavior, with possible differences between populations in relation with their usual diet,  
404 while abrasion should depend directly from the resources ingested. Both may contribute to a  
405 signature of diet on the cusp morphology. Since mice of different age occur in wild-trapped  
406 populations, variations in the degree of molar wear overwhelmed the differentiation of genetic origin  
407 between the populations. Focusing on the lowest part of the crown by using a truncated template  
408 appeared as an efficient way to mitigate this issue, and to focus on genetic differences (Ledevin et al.,  
409 2016; Pallares et al., 2017). In contrast, focusing on the upper molar row or the complete UM1 sheds  
410 light on processes related to wear and its relationship with diet.

411

### 412 *A genetic signature on molar crown shape*

413 Focusing on the genetic signature by using the truncated descriptor of the UM1, molars of the non-  
414 commensal mice from Guillou appeared well differentiated from their Western European relatives, in  
415 agreement with an important founder effect and subsequent evolution in isolation (Hardouin et al.,  
416 2010). Subtler shape differences differentiated the two French commensal populations from Balan  
417 and Tournay. These results are in agreement with the fact that house mouse populations function as  
418 relatively isolated demes (Pocock et al., 2004), promoting morphological diversification even at a  
419 small geographic scale (Chevret et al., 2021). Within-group variation was still important when  
420 considering the truncated descriptors of the UM1, but instead of describing molar abrasion, this  
421 signal corresponded mostly to changes in the shape of the molar forepart, a signature of  
422 developmental variability in house mice (Hayden et al., 2020).

423 Similar results were already obtained using 2D analyses of the crown outline: divergence between  
424 Guillou and European mice (Renaud et al., 2013), differentiation among commensal Western  
425 European populations (Renaud et al., 2017) and importance of the molar elongation in within-group  
426 variation (Renaud et al., 2011). The facility and low cost of 2D morphometric analyses facilitate

427 access to large sample sizes, useful to assess patterns of within-group variance and their importance  
428 in channeling evolution (Renaud et al., 2015a). By suppressing large parts of the tooth geometry, the  
429 truncated descriptor of the UM1 operates an important simplification of the tooth geometry, ending  
430 close to a description of the two-dimensional molar outline. Yet, it can bring information on the  
431 central cusps on the tooth and more details on the shape changes of individual cusps, thus having the  
432 potential to provide a useful complement to extensive 2D analyses (Ledevin et al., 2016).

433

#### 434 *Dynamics of wear and molar arrangement through age*

435 The lab-bred group provided the opportunity to investigate the dynamics of wear in controlled  
436 conditions, in animals with a wild genetic background. The dynamics observed for the upper molar  
437 row was marked by a major change occurring between 26 and 30 postnatal days. In house mice, the  
438 third molar is erupting at 21 days, considered as the moment of weaning. In agreement, the young  
439 mouse at 21 days displayed an erupting third molar that was not yet in contact with the second  
440 molar. Between 22 and 26 days, the third molar was in contact with the second molar but still on the  
441 course of eruption, with very poorly developed roots. An abrupt change in geometry corresponded  
442 to the termination of the third molar eruption and the achievement of the “adult” arrangement of  
443 the three molars along the row. Thereafter, the changes in geometry were less important and  
444 followed a linear trajectory, corresponding to the progressive abrasion of the molar cusps.

445 In contrast, the first upper molar (UM1) is only affected by wear *sensu stricto*. As a consequence, its  
446 trajectory along age mostly corresponded to a linear trend describing cusp abrasion. The youngest  
447 specimens (below 30 days) however appear slightly shifted away from the trend, suggesting two  
448 phases, as for the molar row. This may correspond, shortly after weaning, to the onset of the wear  
449 facets formation.

450 The shift from milk suckling to mastication is associated with a cohort of anatomical changes,  
451 including in the composition of the tongue and masseter muscle fibers. A shift towards muscles with  
452 greater contraction speed and force (Maejima et al., 2005) leads to a rapid increase in bite force  
453 (Ginot et al., 2020). The weaning period is further associated with a stabilization of mandible shape  
454 close to the adult morphology (Swiderski & Zelditch, 2013; Ginot et al., 2020), pointing to a  
455 maturation of the whole masticatory apparatus during this critical period. The dynamics observed in  
456 the lab may however be modulated in wild mice. Weaning in lab mice was strictly completed at 21  
457 postnatal days by removing the litter from the mother. However, in natural conditions, weaning  
458 occurs more progressively, with an increasing incorporation of solid food starting at 17 days, and a

459 cessation of sucking by 23 days, but this date can be modulated depending on nutrient availability  
460 and litter size (König & Markl, 1987).

461 After the weaning phase, the wear facets will progressively expand as the cusps are worn down, up  
462 to a point of advanced abrasion resulting in outlying, senescent molar morphologies, that were not  
463 observed in the present lab-bred samples nor in laboratory mice aged of six months (Renaud &  
464 Ledevin, 2017). Such morphologies were however observed in the two wild populations of Balan and  
465 Guillou 2009.

466

#### 467 *Relationship of wear trajectories with proxies of growth*

468 Age is unknown for wild mice, making the use of indirect proxies necessary to investigate the  
469 dynamics of wear across populations. Body size, estimated by weight, and mandible size were  
470 considered here. Both continue to grow along the animal's life, without reaching a real plateau  
471 (Ginot et al., 2020), making them adequate to trace trajectories along life. However, pace of growth  
472 is not linear, slowing down between 40 and 60 postnatal days (this study, (Ginot et al., 2020)).  
473 Furthermore, body weight displays a marked sexual dimorphism (Ginot et al., 2020), whereas molar  
474 geometry and wear do not. Sexual dimorphism is very limited regarding mandible size (Renaud et al.,  
475 2010; Ginot et al., 2020). However, mandible growth can vary across populations, for instance due to  
476 nutrient quality, as exemplified by Guillou mice from 1993 displaying a mandible smaller than  
477 expected given their body weight (Renaud et al., 2015b). Furthermore, between-group differences in  
478 body size, for instance due to insular gigantism (Gray et al., 2015 ), may render the comparison of  
479 trajectories difficult. Each proxy is therefore prone to biases that may distort the image of wear  
480 trajectories across populations.

481

#### 482 *Diet modulates the arrangement of the molars along the row*

483 When comparing the pattern of shape differences obtained on the molar row to those observed for  
484 the first upper molar, a component can be isolated that does not correspond to genetic differences  
485 (assessed by the analysis of the truncated UM1) nor to wear *sensu stricto* (included in the analysis of  
486 the UM1) (Fig. 7). This component corresponds to the arrangement of the three molars along the  
487 row. The two commensal populations from Balan and Tournay appear to share a similar signature,  
488 with a third molar pushed away from the alignment with the first and second molar towards the  
489 lingual side. In contrast, the early Sub-Antarctic population (Guillou 1993) displays a straight molar  
490 row.

491 Murine rodents are characterized by a propalinal masticatory movement (Lazzari et al., 2008),  
492 meaning that they grind food by sliding their molar teeth from back to front. The longitudinal  
493 alignment of the cusps is of primary functional importance to guide this movement. The torsion  
494 observed in the molar rows of the commensal mice shows that the masticatory movement is more  
495 complex and incorporates transverse components. In agreement, analysis of jaw motion during  
496 mastication showed that the closing movement consists of an arched, forward trajectory (Utsumi et  
497 al., 2010) that may contribute to push the third molar towards the lingual side. The onset of  
498 differences in molar alignment between commensal and Guillou seems to occur as early as weaning.  
499 Since the third molar is just erupting at this time, with not fully developed roots, loadings during  
500 mastication could easily modify its insertion relative to the second molar.

501 The torsion of the molar row in the commensal populations therefore suggests important masticatory  
502 loadings, in agreement with a hard diet mostly based on cereal grains, promoting an optimization of  
503 the mandible morphology for mastication at the molars (Renaud et al., 2015b). In contrast, Guillou  
504 mice have shifted their diet towards an increased proportion of animal prey (Le Roux et al., 2002).  
505 This has favored an optimization of the jaw for incisor biting while relaxing the constraints related to  
506 mastication (Renaud et al., 2018a), thus limiting the loading on the third molar which can retain its  
507 alignment with the first and second ones. Note that, on the short time scale involved, the reduction  
508 of the third molar, interpreted as a further signature of faunivorous diet (Kavanagh et al., 2007), did  
509 not evolve in these populations.

510 Strikingly, Balan laboratory offspring display a different molar row geometry compared to their wild  
511 relatives. This difference, occurring over one or two generations only, provides evidence that plastic  
512 remodeling of the molars' insertion is involved here. The molar geometry of the laboratory offspring  
513 tends to converge towards the Guillou one. A similar decrease in masticatory demand likely occurs in  
514 lab-bred mice: being fed *ad libitum*, they probably chew their food less thoroughly than wild animals  
515 confronted to the costs of foraging (Savriama et al., 2022).

516 However, the molar row geometry of the most recent population from Guillou, trapped in 2009,  
517 appears to share features with commensal mice. Between 1993 and 2009, Guillou Island experienced  
518 a cohort of environmental changes that decreased the availability of earthworms but increased the  
519 proportion of invasive insects and plants (Lebouvier et al., 2002; Laparie et al., 2010). This  
520 presumably led to a more omnivorous diet in mice from 2009 than in those from 1993, a hypothesis  
521 supported by changes in mandible shape and slight differences in microwear patterns (Renaud et al.,  
522 2015b). This shows that changes in the relative arrangement of the molars along the row can

523 respond to fine changes in the diet composition on a very small time scale, hardly traceable using  
524 alternate proxies of diet.

525

526 *Wear trajectories suggest reduced abrasion rate in Sub-Antarctic mice*

527 Wear massively affects molar geometry but corresponds to a pattern of within-group variation  
528 shared by all populations. Whatever the food consumed, the wear pattern on the first molar seems  
529 thus to be comparable. Differently from the early closing phase during mastication, the late closing  
530 phase occurs parallel to the sagittal plane (Utsumi et al., 2010). This may explain why the signature of  
531 wear on the UM1 is not impacted by the rotational component of jaw motion and is similar in all  
532 populations. However, the rate of wear may vary depending on the diet.

533 The commensal populations from Balan and Tournay appeared to share a rapid wear along growth, in  
534 contrast with Guillou mice and Balan laboratory offspring, displaying less advanced wear for a given  
535 growth stage, in agreement with reduced masticatory demand. The closeness between Guillou 2009  
536 and Guillou 1993 suggest a decoupled response to loadings during mastication, impacting the  
537 insertion of the molars along the row, and to the abrasiveness of the food, affecting rate of wear.

538 Few highly abraded teeth, with almost all cusps worn down, appeared as outliers. Their occurrence  
539 however provides evidences that such advanced wear, and presumably age, can be occasionally  
540 reached on the Sub-Antarctic Guillou island, despite unfavorable environmental conditions and  
541 important winter mortality (Ferreira et al., 2006).

542

## 543 **Conclusions**

544 The present study validated the hypothesis that molar wear may trace ecological differences  
545 occurring over very short time scales. Sub-Antarctic Guillou mice display modified wear dynamics  
546 compared to commensal mice, due to their shift from an omnivorous-granivorous diet requiring  
547 efficient mastication, towards a more “predator” behavior associated with an optimization for incisor  
548 biting. In these mice, the rate of wear seems to be decreased, as in mice bred in laboratory  
549 conditions. Tracing wear dynamics requires, however, proxies of the animal’s age comparable across  
550 populations. Evolution of body size, for instance in insular populations (Gray et al., 2015 ), could  
551 render the comparison of wear trajectories among populations difficult. Craniofacial measurements  
552 could provide alternate proxies, but they are equally impacted by evolution and can vary depending  
553 on local conditions affecting bone mineralization, as for instance here in the early population from

554 Guillou (Renaud et al., 2015b). Despite these potential limits, wear trajectories could be a non-  
555 invasive approach to trace diet differences in museum specimens for which body size data are  
556 available.

557 Variation in the arrangement of the three molars along the row seems to trace other differences in  
558 diet, related to loadings during mastication. They were able to trace diet shifts occurring over few  
559 decades as the results of climate warming and the spread of invasive species on Guillou Island.  
560 Quantifying the torsion along the molar row could thus contribute to trace subtle diet variations,  
561 together with other proxies such as wear or topographic indices (Pampush et al., 2016). Furthermore,  
562 this approach provides a direct insight into the constraints during mastication, which may help to  
563 better understand the functional role of changes in the molar geometry.

564

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576

#### 577 **Data Availability**

578 The 3D models of most molar rows are deposited online (Renaud et al. 2023). Morphometric data  
579 are provided as Supplementary Material.

580

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- 715
- 716

717 **Tables**

	P <sub>KW</sub>	BW/BL	BW/G09	BW/G93	BW/To	BL/G09	BL/G93	BL/To	G09/G93	G09/To	G93/To
BWeight <sup>1/3</sup>	0.0692	0.538	1.000	1.000	1.000	1.000	1.000	0.077	1.000	0.743	0.743
MdL	<i>0.0037</i>	0.079	1.000	0.364	1.000	0.454	0.012	0.079	0.267	1.000	0.394
UM1 CS	<b>&lt;0.0001</b>	0.086	<i>0.003</i>	0.341	0.526	<b>0.0002</b>	0.041	0.526	0.526	<b>0.0001</b>	0.040

718 *Table 1. Size differences between populations. PKW: Kruskal-Wallis probability; next columns:*  
719 *probabilities of pairwise differences (Wilcoxon test). BWeight<sup>1/3</sup>: cubic root of body weight; MdL:*  
720 *mandible length; UM1 CS: centroid size of the first upper molar. In bold, P < 0.001. In italics P < 0.01*  
721 *(uncorrected P-values are provided). BW: Balan Wild; BL: Balan Lab; G93: Guillou 93; G09: Guillou 09;*  
722 *To: Tournch.*

723

		Age	Sex	Age:Sex
Anova	BWeight <sup>1/3</sup>	<b>0.0001</b>	0.0836	0.1719
	MdL	<b>0.0001</b>	0.6989	0.7897
ProcD.Im	UMR	<b>0.0003</b>	0.3146	0.0857
	UM1	<b>0.0001</b>	0.4607	0.5812

724 *Table 2. Effect of age and sex on body size proxies and wear trajectories. BWeight<sup>1/3</sup>: cubic root of*  
725 *body weight ; MdL: Mandible length. In bold P < 0.001.*

726

	BW <sup>1/3</sup>		MdL	
	%	P	%	P
UMR	6.9	<b>0.0003</b>	6.6	<b>0.0005</b>
UM1	7.2	<b>0.0005</b>	10.6	<b>0.0001</b>
UM1tr	1.0	0.8316	2.5	0.0861

	CRW		Gp		W:Gp		MdL		Gp		MdL:Gp	
	%	P	%	P	%	P	%	P	%	P	%	P
UMR	6.9	<b>0.0001</b>	33.2	<b>0.0001</b>	4.8	0.1696	6.6	<b>0.0001</b>	33.5	<b>0.0001</b>	5.2	0.0686
UM1	7.2	<b>0.0001</b>	33.4	<b>0.0001</b>	4.9	0.1497	10.6	0.0001	28.9	<b>0.0001</b>	4.5	0.2970
UM1tr	1.0	0.5656	29.6	<b>0.0001</b>	5.5	0.1779	2.5	0.0171	28.6	<b>0.0001</b>	6.3	0.3820

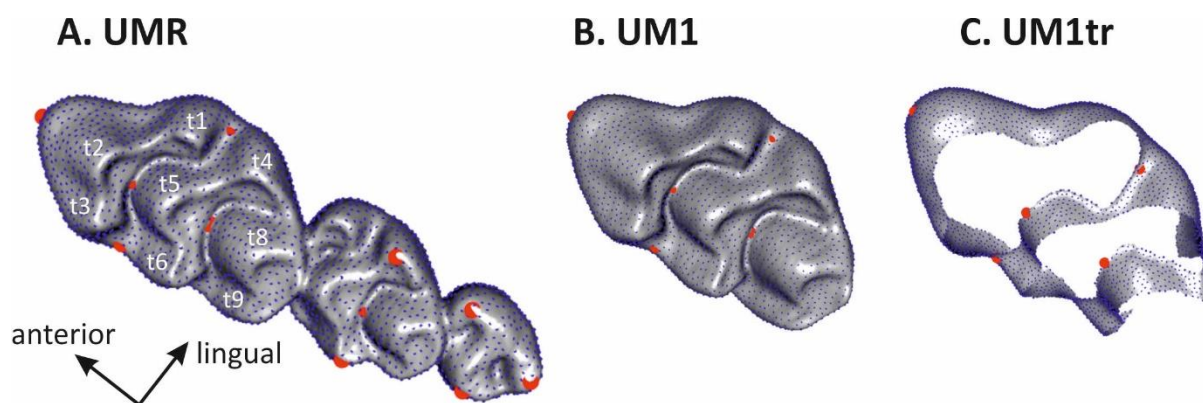
727

728 *Table 3. Relationships between shape (aligned coordinates) and proxy of animal's size and*  
729 *populations. Percentage of variance explained (%) and probabilities (P) of Procrustes ANOVA are*  
730 *given. BW<sup>1/3</sup>: cubic root of body weight. MdL: mandible length. In bold P < 0.001, in italics P < 0.01.*

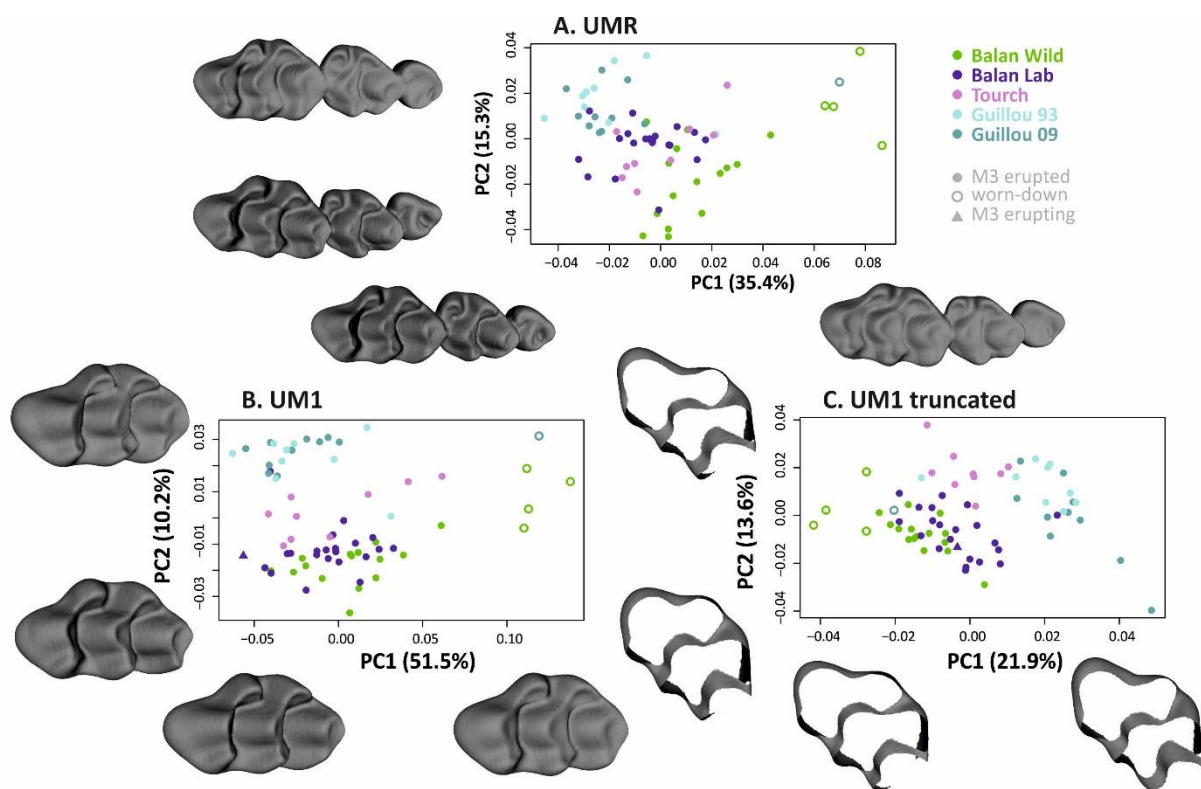
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## 733 Figures and Figure Captions

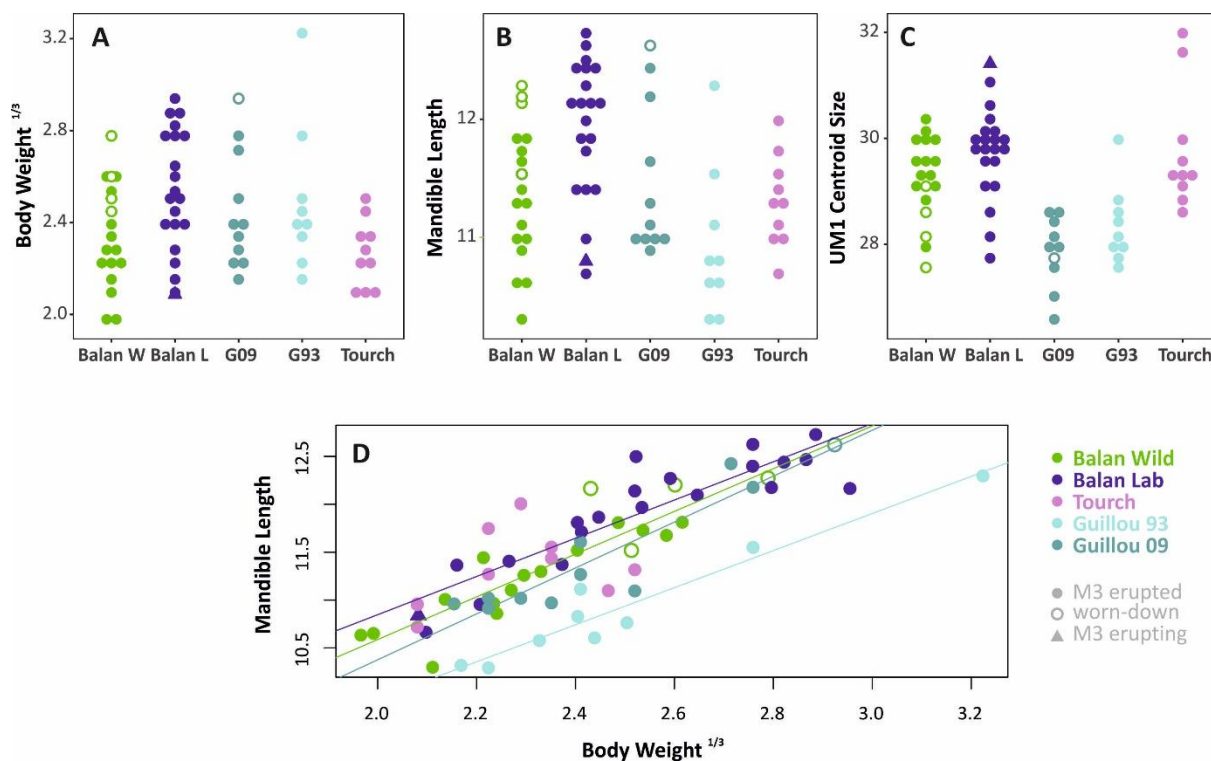


735 *Figure 1. Templates used to describe the 3D molar morphology. (A) Upper molar row (UMR), with*  
 736 *labelling of the cusps. (B) First upper molar (UM1). (C) Truncated “wear-free” first upper molar*  
 737 *(UM1tr). Red dots: fixed landmarks used to anchor the templates; blue dots: sliding semi-landmarks.*



739

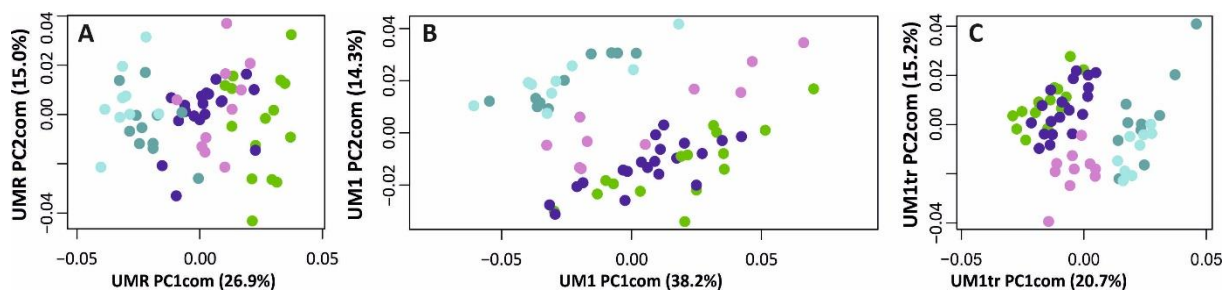
740 *Figure 2. Morphospaces depicting the shape variation of the upper molar row (A), the first upper*  
 741 *molar (B) and the truncated first upper molar (C). The first two axes of a PCA on the aligned*  
 742 *coordinates are represented. Along each axis, visualizations of molar shape corresponding to PC*  
 743 *scores = 0.05 and -0.05 are depicted. Top right: legend common to the three panels (specimen with*  
 744 *M3 erupting absent from the analysis of the UMR on panel A).*



745

746 *Figure 3. Size differences between populations and relationship between size estimators. (A) cubic*  
 747 *root of body weight. (B) mandible length. (C) centroid size of the first upper molar. (D) Relationship*  
 748 *between body weight<sup>1/3</sup> and mandible length. Full lines represent significant within-population*  
 749 *regressions ( $P < 0.05$ ).*

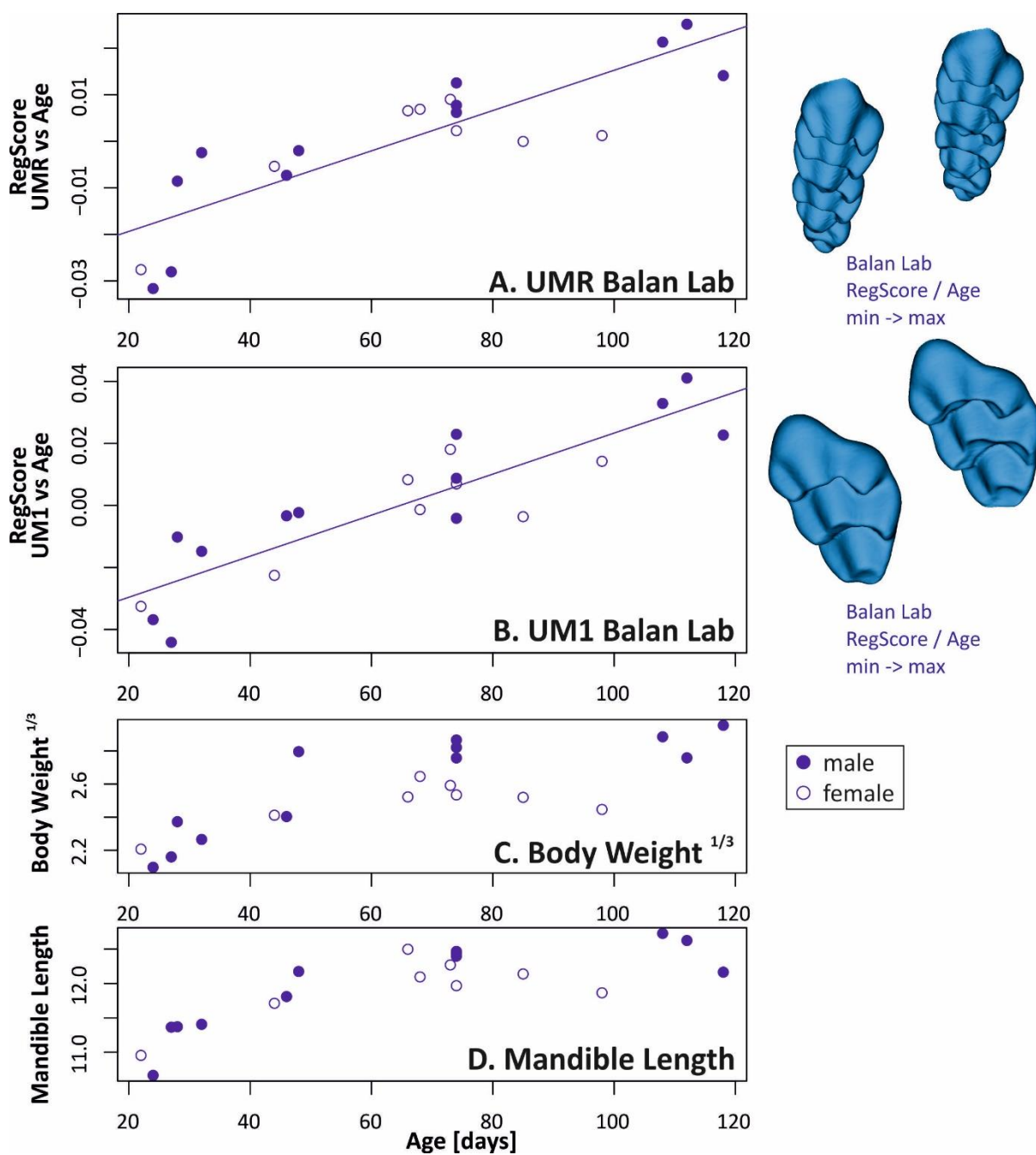
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751

752 *Figure 4. Morphospaces representing shape variation of the upper molar row (A), the first upper*  
 753 *molar (B) and the truncated first upper molar (C), corresponding to PCAs on a subsampling (N = 64)*  
 754 *discarding the senescent specimens and the mouse at weaning.*

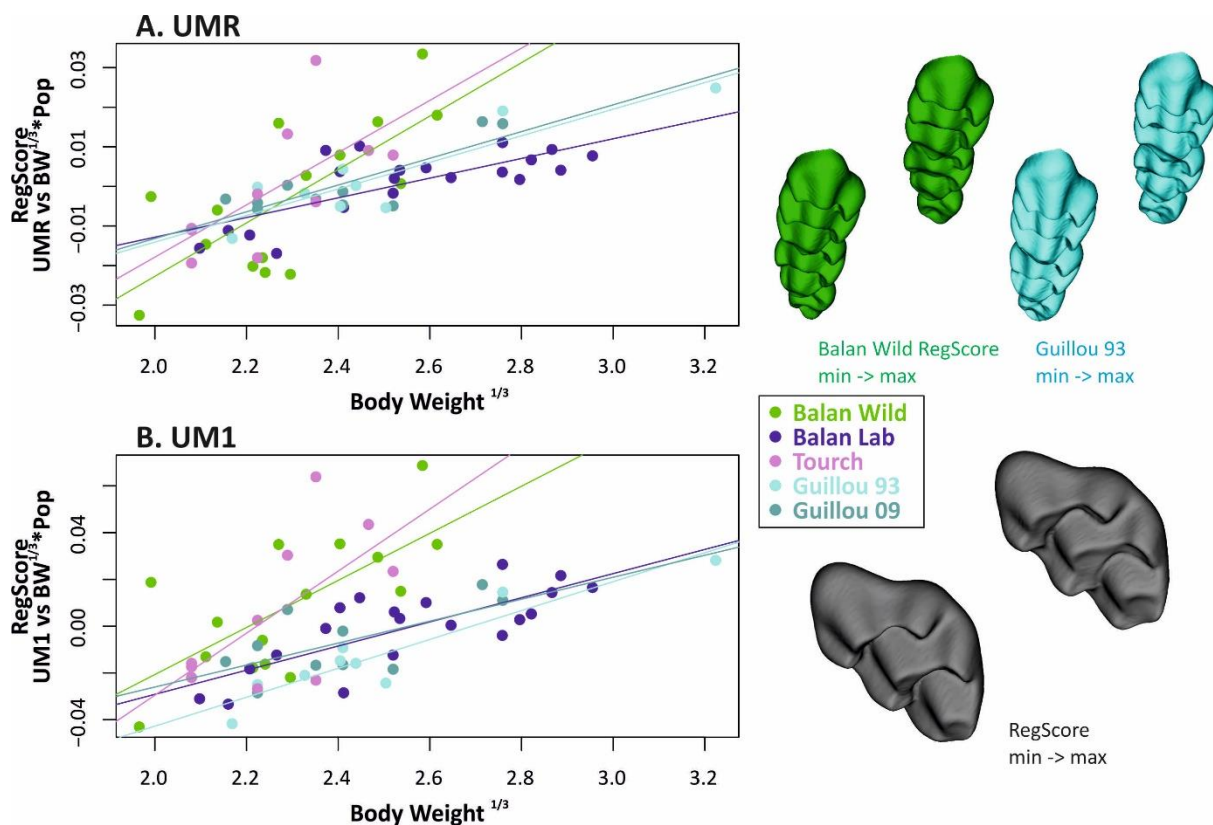
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756

757 *Figure 5. Variations with age and sex in the group of laboratory-bred mice (Balan Lab). (A,B) Wear*  
 758 *trajectories as a function of age, visualized using regression scores based on Procrustes ANOVAs of*  
 759 *aligned coordinates of the upper molar row (A) and the first upper molar (B). Full lines represent the*  
 760 *regression of regression score vs. age ( $P < 0.05$ ). (C) Body weight<sup>1/3</sup> as a function of age in the same*  
 761 *mice. (D) Mandible length as a function of age. The symbols correspond to the sex of the mice.*

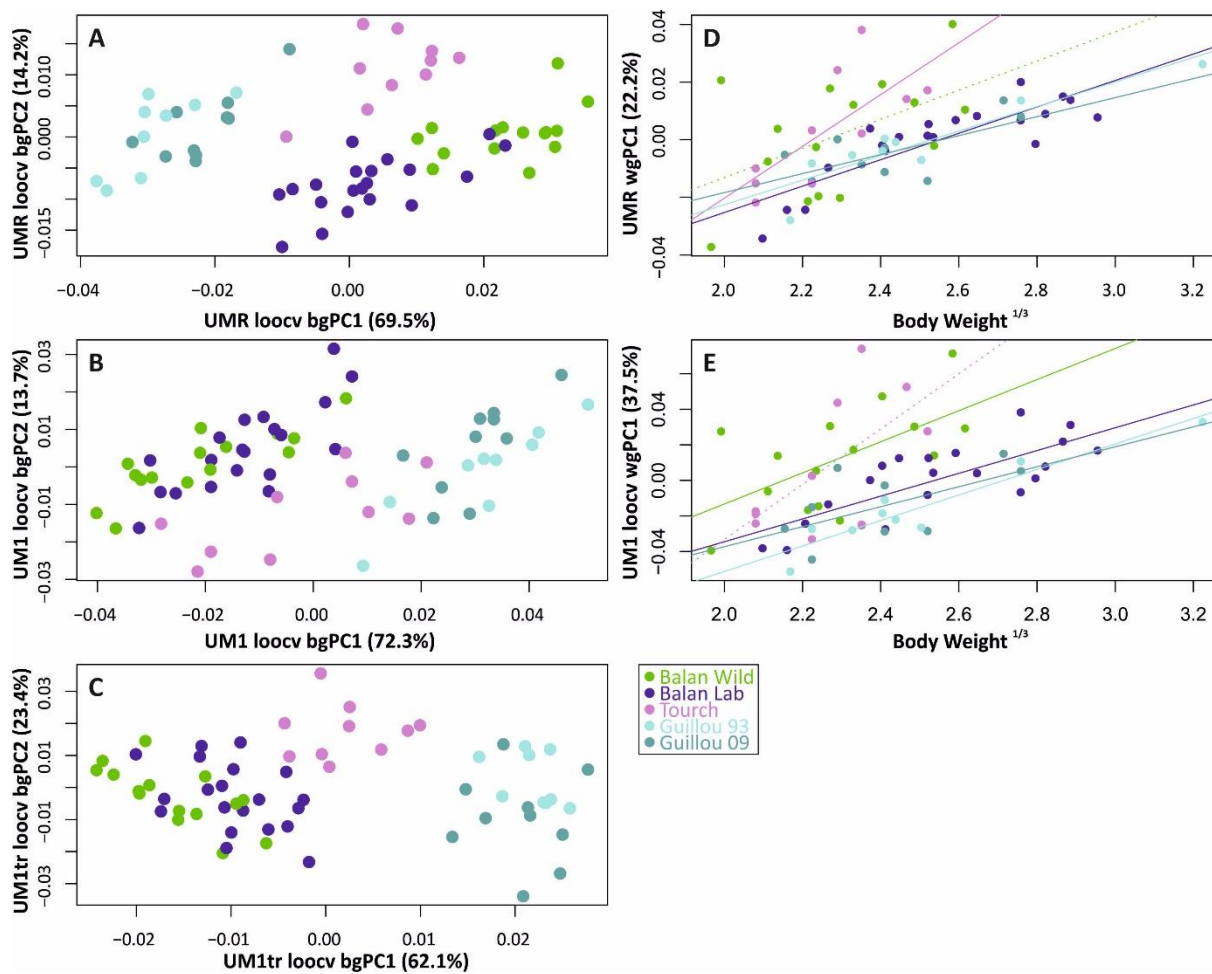
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763

764 *Figure 6. Wear trajectories for the upper molar row (A) and the first upper molar (B). Body weight<sup>1/3</sup> is*  
 765 *used as proxy of growth. Regression scores, based on Procrustes ANOVAs of aligned coordinates vs*  
 766 *body weight<sup>1/3</sup> and population, allowed to visualize shape variance related to wear within each*  
 767 *group. Full lines represent significant within-group regressions of regression score vs. weight<sup>1/3</sup>*  
 768 *(significant within all groups at P < 0.05).*

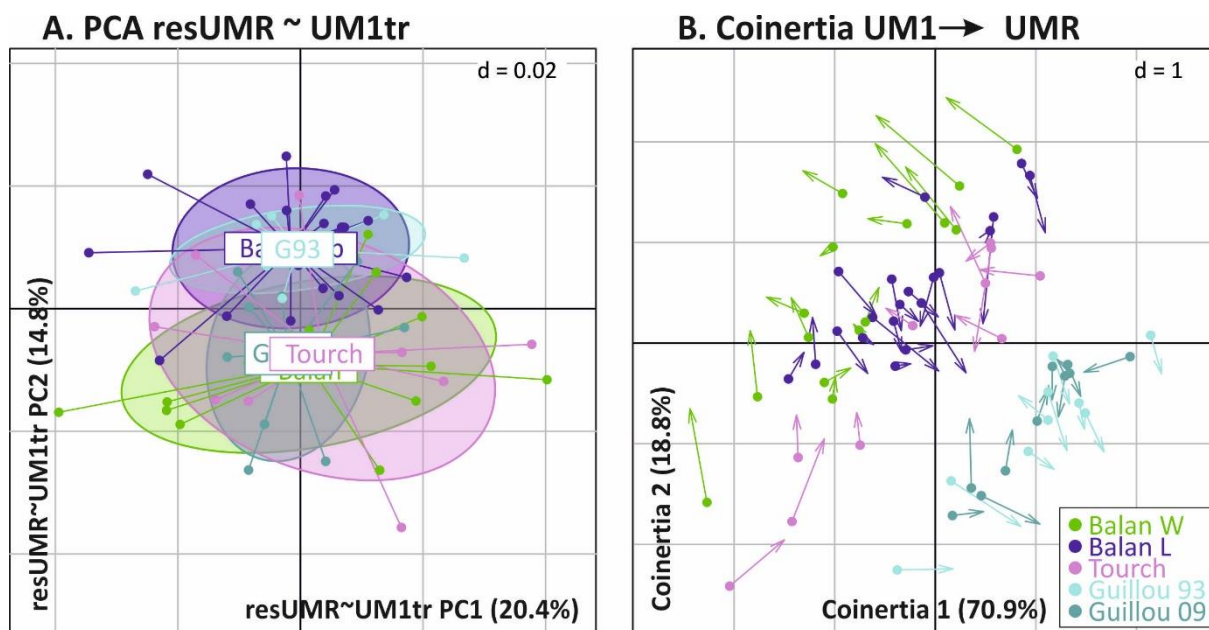
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770

771 *Figure 7. Between-group variation (A,B,C) and within-group variation (D,E) of the molar geometry.*772 *(A,D) Upper molar row; (B,E) first upper molar; (C) truncated first upper molar. (A, B, C) Cross-*773 *validated factor map of the bgPCA. (D, E) Relationship between the first within-group PC axis and*774 *body weight<sup>1/3</sup>. Full lines correspond to significant within-group regressions ( $P < 0.05$ ). The dotted line*775 *corresponds to marginally significant regression ( $0.05 < P < 0.10$ ).*

776

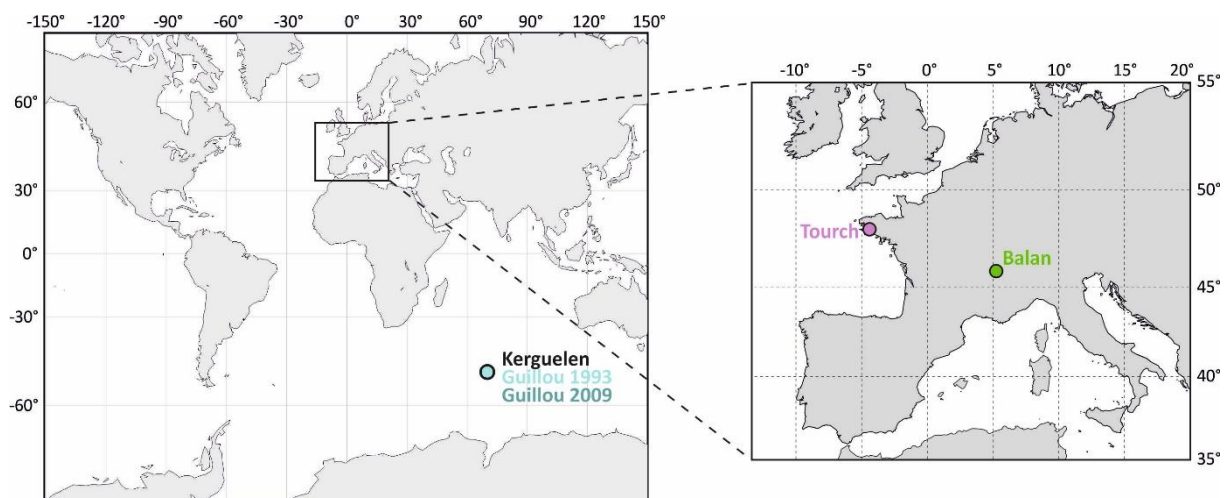


777

778 *Figure 8. (A) PCA on the residuals of a multivariate regressions of the UMR aligned coordinates vs the*  
 779 *first two axes of the PCA on the truncated UM1. (B) Coinertia between shape variation of the UM1*  
 780 *and UMR. The arrow indicates the change in topology going from the first (UM1) to the second (UMR)*  
 781 *dataset.*

782

783

784 **Supplementary Material**785 **Supplementary Figure 1.** Map of the localities considered in the study.

786

787

788 **Supplementary Table 1.** Sampling of the study: Specimen ID, population, wear stage, age for

789 laboratory specimens, weight and 3D mandible length.

Individual ID	Population	Stage	Age [days]	Sex	Weight [g]	MdL [mm]
Balan_02_2015	Balan	adult			7.6	10.6
Balan_03	Balan	adult		F	12.1	11.3
Balan_04	Balan	senescent		F	21.7	12.3
Balan_06	Balan	adult		M	17.9	11.8
Balan_07	Balan	adult		F	11.3	10.9
Balan_08	Balan	adult		F	17.3	11.7
Balan_11	Balan	adult		M	10.9	11.4
Balan_12	Balan	adult		F	11.7	11.1
Balan_BAL15	Balan	adult		M	11.2	11.0
Balan_BAL16	Balan	senescent		M	15.9	11.5
Balan_BAL17	Balan	adult		M	7.9	10.7
Balan_BAL18	Balan	adult		F	9.4	10.3
Balan_BAL19	Balan	adult		M	16.3	11.7
Balan_BAL20	Balan	adult		M	12.7	11.3
Balan_BAL21	Balan	senescent		M	17.6	12.2
Balan_BAL22	Balan	adult		F	13.9	11.5
Balan_BAL23	Balan	adult		F	15.4	11.8
Balan_BAL24	Balan	senescent		M	14.4	12.2
Balan_BAL25	Balan	adult		M	9.8	11.0
Balan_Lab_35	BalanLab	adult	98	F	14.7	11.9
Balan_Lab_46	BalanLab	adult	85	F	16.0	12.1
Balan_Lab_54	BalanLab	adult	73	F	17.4	12.3
Balan_Lab_56	BalanLab	adult	74	F	16.3	12.0

Balan_Lab_86	BalanLab	adult	108	M	24.0	12.7
Balan_Lab_82	BalanLab	adult	118	M	25.8	12.2
Balan_Lab_92	BalanLab	adult	112	M	21.0	12.6
Balan_Lab_319	BalanLab	adult	68	F	18.5	12.1
Balan_Lab_325	BalanLab	adult	74	M	22.5	12.4
Balan_Lab_329	BalanLab	adult	74	M	21.0	12.4
Balan_Lab_330	BalanLab	adult	74	M	23.6	12.5
Balan_Lab_02050407_01	BalanLab	adult	63	F		12.4
Balan_Lab_17032205_01	BalanLab	adult	66	F	16.1	12.5
Balan_Lab_BB3weeks	BalanLab	weaning	21		9.0	10.8
Balan_Lab_167	BalanLab	adult	48	M	21.9	12.2
Balan_Lab_188	BalanLab	adult	32	M	11.6	11.4
Balan_Lab_192	BalanLab	adult	28	M	13.4	11.4
Balan_Lab_194	BalanLab	adult	46	M	13.9	11.8
Balan_Lab_196	BalanLab	adult	44	F	14.0	11.7
Balan_Lab_30x17	BalanLab	adult	27	M	10.1	11.4
Balan_Lab_40x56	BalanLab	adult	24	M	9.2	10.7
Balan_Lab_47x61	BalanLab	adult	22	F	10.8	11.0
G09_06	G09	adult		F	10.0	11.0
G09_10	G09	adult		F	16.0	11.1
G09_15	G09	adult		F	11.0	11.0
G09_16	G09	adult		M	14.0	11.3
G09_17	G09	adult		M	11.0	10.9
G09_21	G09	adult		F	21.0	12.2
G09_26	G09	senescent		M	25.0	12.6
G09_27	G09	adult		F	20.0	12.4
G09_29	G09	adult		F	13.0	11.0
G09_65	G09	adult		M	14.0	11.6
G09_66	G09	adult		M	12.0	11.0
G93_03	G93	adult		F	12.6	10.6
G93_04	G93	adult		F	11.0	10.3
G93_10	G93	adult		F	15.7	10.8
G93_11	G93	adult		M	14.5	10.6
G93_13	G93	adult		F	14.0	11.1
G93_14	G93	adult		F	13.9	10.8
G93_15	G93	adult		F	10.2	10.3
G93_24	G93	adult		M	21.0	11.6
G93_25	G93	adult		M	33.5	12.3
Tourch_7819	Tourch	adult		F	12.0	12.0
Tourch_7821	Tourch	adult		F	11.0	11.7
Tourch_7839	Tourch	adult		M	9.0	11.0
Tourch_7873	Tourch	adult		M	13.0	11.6
Tourch_7877	Tourch	adult		F	11.0	11.3
Tourch_7922	Tourch	adult		M	15.0	11.1
Tourch_7923	Tourch	adult		F	16.0	11.3
Tourch_7925	Tourch	adult		M	13.0	11.4

Tourch_7927	Tourch	adult		F	9.0	11.0
Tourch_7932	Tourch	adult		M	9.0	10.7

790

791

792 **Supplementary Files (datasets)**793 UM1\_BWLGT\_BJLS\_Renaud\_et\_al\_2023.txt

794 File containing for each specimen (IndID) the population, centroid size (CS) and PC scores  
795 corresponding to the UM1 analysis.

796

797 UM1tr\_BWLGT\_BJLS\_Renaud\_et\_al\_2023.txt

798 File containing for each specimen (IndID) the population, centroid size (CS) and PC scores  
799 corresponding to the analysis of the truncated UM1.

800

801 UMR\_BWLGT\_BJLS\_Renaud\_et\_al\_2023.txt

802 File containing for each specimen (IndID) the population, centroid size (CS) and PC scores  
803 corresponding to the UMR analysis. Note that corresponding to the previous files, the specimen at  
804 weaning (Balan\_Lab\_BB3weeks) has been deleted since its molar row was not complete (eruption of  
805 the UM3 not completed).

806

807

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809