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Sorting the flock: quantitative identification of sheep and goat from isolated third lower molars and mandibles through geometric morphometrics

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Highlights

- Sheep and goat differ in their third lower molar and mandible size and shape
- Based on shape, 93% of the molar and 95% of the mandibles can be correctly identified
- Geometric morphometrics allow the identification of archaeological specimens
- Geometric morphometric and macroscopic identifications are only partially congruent

36 **Abstract:**

37 Sheep and goat are often herded together and show morphological similarities in their
38 skeleton. Being able to identify archaeological remains of these two taxa to species level is
39 particularly important for understanding and characterising past herding practices. Discrete
40 criteria are now available to identify a large number of their bones and teeth, and quantitative
41 approaches have been developed for post-cranial elements but not for mandible and isolated
42 teeth. In this paper we explore the discriminating potential of geometric morphometrics to
43 identify modern sheep and goat third lower molar and mandible and its application on
44 archaeological specimens. The size and shape of the mandible and the third lower molar of
45 143 modern specimens (101 sheep and 42 goats) were quantified using 2D-landmark and
46 sliding semi-landmarks geometric morphometric approaches. The results show that sheep and
47 goat differ in terms of the size, shape, and form (*i.e.* size and shape together) in both studied
48 elements. Classification accuracy of the two species reaches 93.3% (CI: 90.0-95.7%) for third
49 lower molar shape, 62.7% (CI 57.1-68.6%) for third lower molar size, 95.2% (CI: 92.0-
50 97.4%) for mandible shape and 84.0% (CI 81.6-86.8%) for mandible size. Form does not
51 provide better classification than shape alone. Sex and age appear to have little impact on the
52 ability to differentiate between sheep and goat, despite the two species displaying distinct
53 sexual dimorphism and changes through age. The same methodology was then applied on 32
54 Middle Ages third lower molars from Missignac-Saint Gilles le Vieux, Aimargues, France.
55 The identifications obtained through geometric morphometrics were only partially congruent
56 with the identifications based on visual observations calling for caution in the interpretation
57 and further investigations. Further research should include molecular identification of the
58 archaeological specimens to assess whether the geometric morphometric identification can be
59 made with confidence for all periods and all geographic areas. Nevertheless, the results
60 obtained with the newly developed geometric morphometric protocols represent an important
61 contribution toward a better understanding of past livestock husbandry practices.

62

63

64 **Keywords :** archaeozoology, discriminant analysis, classification, *Ovis aries*, *Capra hircus*,
65 GMM

66 **1. Introduction**

67 Sheep and goat were among the first animals to be domesticated (Naderi, 2007; Vigne et al.,
68 2011) about 11,000-10,500 years ago in south-eastern Anatolia, and spread across Europe
69 from approximately 8,000 years ago as part of the Neolithic revolution, expanding beyond the
70 range of their wild ancestors, *Ovis orientalis* and *Capra aegagrus* (Clutton-Brock, 1989;
71 Peters et al., 2002; Vigne et al., 2015). Since then, sheep and goat became ubiquitous;
72 regardless of the type of archaeological site and the geographic area considered, they are often
73 among the most represented species as they formed the basis of many agropastoral societies
74 and still are two inseparable emblematic species of the Mediterranean basin landscape (Altuna
75 and Mariezkurrena, 2009; Álvarez-Fernández et al., 2014; Cubas et al., 2016; Peters et al.,
76 1999; Vigne, 2011, 1988).

77
78 Sheep and goats have been selected and bred for multiple purposes, including secondary
79 products such as milk, wool/hair and manure, which can be collected during the life of the
80 animal, and final products such as skin, meat, horn and bones (*e.g.* Gillis et al., 2019).
81 Because of their morphometric similarity and the fact that they readily form mixed herds,
82 sheep and goat are often studied has a single entity (*e.g.* French, 1970; Grau-Sologestoa,
83 2015; Helmer, 2000; Payne, 1973; Salvagno, 2020). However, the two species have different
84 feeding behaviour, with sheep generally preferring to graze on grass and soft plants while
85 goats browse on leaves and bushes (Balasse and Ambrose, 2005). As a general trend, goat is
86 preferred for clothing or wineskin and milk (Bourrier, 1897; De Serre, 1600), while sheep is
87 preferred for meat and wool (Blaise, 2009; Helmer et al., 2005; Helmer and Vigne, 2004).

88
89 Differentiating and identifying sheep and goat is a very well-known longstanding challenge in
90 zooarchaeology (*e.g.* Cornevin and Lesbre, 1891) and many studies have proposed discrete
91 morphological criteria that has improved greatly our ability to differentiate between the two of
92 them (*e.g.* Balasse and Ambrose, 2005; Boessneck et al., 1964; Fernandez, 2001; Halstead et
93 al., 2002; Helmer, 2000; Payne, 1985; Prummel and Frisch, 1986; Zeder and Pilaar, 2010).
94 More recently, criteria using linear measurements, such as those traditionally collected in
95 archaeozoology (*e.g.* Von Den Driesch, 1976), have been developed for the petrous bone
96 (Mallet et al., 2019; Mallet and Guadelli, 2013) and most postcranial bones (Gron et al., 2020;
97 Salvagno and Albarella, 2017; Zedda et al., 2017).

98 In parallel, with the increased application of geometric morphometrics approaches (GMM, *i.e.*
99 a set of statistical methods and visualizations based on the analysis of landmarks, outline or

100 surface coordinates. See Rohlf and Marcus, 1993; Rohlf and Slice, 1990) to bioarchaeology, a
101 number of studies have been published focusing on the bioarchaeological history of several
102 domestic ungulate species. These include the taxonomic identification of archaeological wild
103 and domestic populations as well as the study of the spatio-temporal variation of ancient and
104 modern domestic populations of pig (*e.g.* Cucchi et al., 2011, 2009; Duval, 2015; Evin et al.,
105 2015; Harbers et al., 2020b, 2020a; Krause-Kyora et al., 2013; Neaux et al., 2020b, 2020a;
106 Ottoni et al., 2013; Owen et al., 2014), cattle (*e.g.* Csippán, 2016; Cucchi et al., 2019), and
107 horse (*e.g.* Chuang and Bonhomme, 2019; Cucchi et al., 2017; Hanot et al., 2017; Seetah et
108 al., 2014). However, only few studies have employed geometric morphometrics on caprine
109 bones. For sheep and goat, this includes studies of the talus with the aim of distinguishing
110 archaeological sheep and goat (Haruda, 2017), differentiate ancient wild sheep from modern
111 and archaeological domestic sheep (Pöllath et al., 2019, 2018), and investigate the variation
112 between archaeological sheep morphotypes (Colominas et al., 2019; Haruda et al., 2019;
113 Vuillien, 2020).

114

115 Mandibular teeth are often targeted by GMM studies because, due to their internal structure,
116 they survive very well deposition and are often found numerous in archaeological
117 assemblages (Binford and Betram, 1977). In addition, they carry a taxonomic signal. The third
118 lower molar in particular, as it is the last tooth in the jaw and is less constrained in its
119 posterior part compared to the other teeth, has been considered a phenotypic marker of
120 adaptation to natural or anthropic environment (Butler, 1939; Cucchi et al., 2019; Dahlberg,
121 1945). Nevertheless, the very few GMM studies exist on sheep and goat focus only on caprine
122 mandibles are all exclusively based on modern specimens (Demiraslan et al., 2020;
123 Demircioğlu et al., 2021; Parés-Casanova, 2013; Yalçın et al., 2010). These studies studied
124 mandibular growth (Parés-Casanova, 2013), sexual dimorphisms (Demiraslan et al., 2020;
125 Demircioğlu et al., 2021) , and differences between wild and domestic sheep (Yalçın et al.,
126 2010).

127

128 Very little attention has also been given to the measurement of teeth for species identification
129 purposes despite the fact that they are among the most commonly found anatomical elements
130 in archaeozoological assemblages (*e.g.* Buckley et al., 2010; Gerbault et al., 2016; Halstead et
131 al., 2002; Payne, 1973). This is probably due to the fact that, until recently, sheep and goat
132 teeth were considered to be too affected by occlusal wear to be used for bioarchaeological
133 studies using a geometric morphometrics approach. Such an approach however, could not

134 only help in the species identification of loose teeth, a practice which is notoriously difficult
135 (especially to a untrained eye), but it also has the potential to greatly contribute to the
136 quantitative exploration of the intra-specific diversity of sheep and goat in the past.

137

138 In that context, the major aims of this study are to: (1) establish protocols to quantify size and
139 shape of modern sheep and goat third lower molar and mandible through the use of geometric
140 morphometrics; (2) assess, for these two elements, the classification accuracy of size, shape,
141 form and allometries (size and shape relationship) to identify sheep and goat; (3) explore the
142 effect of age and sexual dimorphism on tooth and mandible morphometric variation, and their
143 impact on identification accuracy; (4) identify to species level sheep/goat medieval teeth to
144 test whether the new proposed methodology works on archaeological material.

145

146 **2. Materials**

147 *2.1 Modern reference collection*

148 A total of 102 mandibles and 133 third lower molars belonging to 143 specimens of modern
149 breeds are included in this study. Sheep (*Ovis aries*) are represented by 102 specimens from 6
150 breeds (*Préalpes, Lacaune, Merinos, Negretti, Blanche du massif Central* and *Rouge du*
151 *Roussillon*). Goats (*Capra hircus*) are represented by 42 specimens from 5 breeds (*Rove,*
152 *Cabra Catalana, Angora, Corse* and *Cachemire*) (SI table 1). All studied specimens were
153 older than 1 year. Since tooth wear is directly linked to age, and wear can potentially have a
154 significant impact on the tooth size and shape, the age of the specimens was established
155 following Payne's method (1973). Specimens were divided into the following categories: 1-2
156 years, 2-4 years, 4-6 years, 6-8 years and 8-10 years (table 1). Both sexes were present in the
157 sample however, the majority of individuals of known sex were female (table 1).

158

159 *<Table 1>: Age at death of the modern specimens for each species (third lower molar and*
160 *mandible). Sex information, when available, is provided in brackets as follows (female/male).*
161 *Mandibles with no useful teeth for age estimation are classified as 'undetermined age'*
162 *(Undet. age).*

163

Sheep		Goat	
Mandible	Third lower	Mandible	Third lower

		molar		molar
1-2 years	6 (4/1)	5 (5/0)	2 (0/1)	0
2-4 years	9 (8/2)	12 (9/1)	5 (0/1)	4 (0/1)
4-6 years	30 (22/3)	34 (25/3)	14 (9/5)	19 (8/11)
6-8 years	6 (6/0)	9 (8/0)	7 (2/1)	5 (2/1)
4-8 years	2 (1/1)	20 (1/1)	0	0
8-10 years	13 (10/2)	17 (10/2)	5 (4/3)	8 (5/3)
Undet. age	0	0	3 (0/3)	0
Total	66 (51/9)	97 (58/7)	36 (15/14)	36 (15/16)

164

165 2.2 Archaeological specimens

166 Thirty-two archaeological third lower molars from the site of Missignac-Saint Gilles le Vieux
 167 (Aimargues, Gard, France) were analysed, they dated between the end of the fifth century and
 168 the beginning of the thirteenth century AD and were either isolated teeth or teeth embedded in
 169 a mandible (SI-table 4).

170

171 The site our archaeological sample comes from is located in the South-East of France about
 172 15 km from the Mediterranean Sea (Maufras et al., in press.; Maufras and Mercier, 2002;
 173 Mercier and Barberan, 1996; Mion et al., 2019) and it was occupied from the second century
 174 BC to the thirteenth century AD. Preventive archaeology excavation carried out between 2012
 175 and 2013 concerned the heart of a medieval village and its 4 ha peripheral storage district,
 176 with 3950 silos (Maufras et al., 2018; Maufras and Mercier, 2002). While the first occupation
 177 found consists of a villa dated to antiquity, later periods attest to an increase in density of
 178 population starting from the fifth century AD and continuing in the eighth and ninth century
 179 AD. The site is then abandoned during the twelfth century AD, even though burials continued
 180 in the following century (Maufras et al., 2018).

181

182 3. Methods

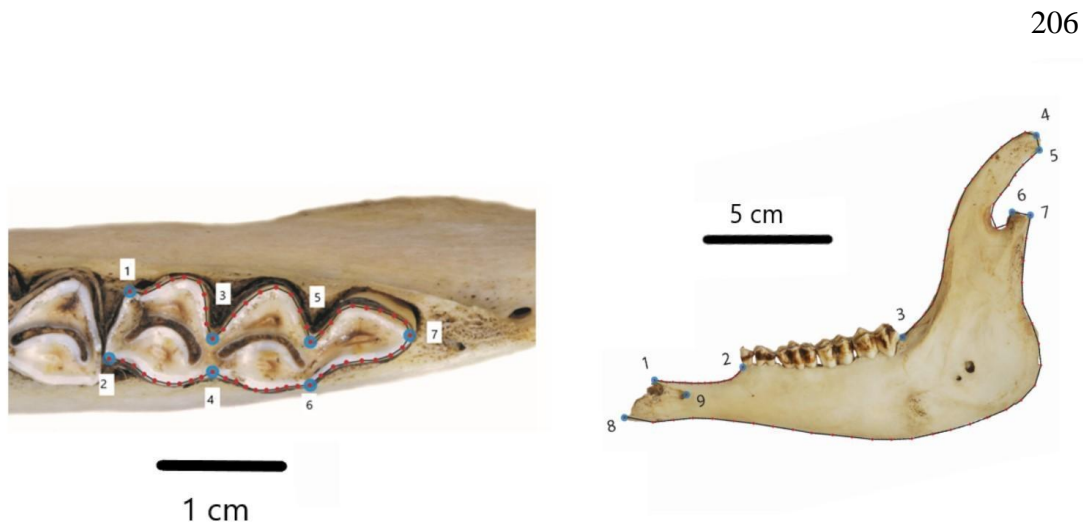
183 3.1 Data acquisition

184 Landmark coordinates were acquired from 2D-images. Third lower molars and mandibles
 185 were photographed using a Nikon d90 LSR camera paired with a 60mm macro lens (AF-S
 186 Micro NIKKOR) attached to a photographic arm (manfrotto 244RC). One ramus of each
 187 mandible was positioned with its labial side facing upward paying particular attention to the

188 flat position of the angular process. Planarity was assessed with a spirit level. Third lower
 189 molars were photographed in their occlusal view and were positioned perpendicular to the
 190 lens with the tooth root equally visible on both lateral sides. A millimetre scale was included
 191 in all pictures. Pictures were acquired both by MJ and AE with negligible inter-operator
 192 differences (lower than 11.6% following Claude, 2008). Mandibles were measured with 9
 193 landmarks (see SI table 2 for a formal description) and 70 sliding semi-landmarks (fig.1). The
 194 sliding semi-landmarks were distributed along four curves as follows: 13 points between
 195 landmarks 1 and 2; 18 points between landmarks 3 and 5; 11 points between landmarks 5 and
 196 7; and 28 points between landmarks 7 and 8. Third lower molars were measured with 6
 197 landmarks and a total of 48 sliding semi-landmarks distributed along 6 curves (8 equidistant
 198 points in each) along the outer outline of the tooth (fig. 1). Point coordinates were acquired
 199 using TpsDig (v2.32) (Rohlf, 2006) by a single operator (MJ).

200
 201

202 <Figure 1>: Geometric morphometric protocols: Position of the landmarks (in blue) and
 203 sliding semi-landmarks (in red) measured on the mandible (right, specimen ISEM_926Ng)
 204 and third lower molar (left, specimen ISEM_926Nj) of a sheep. Landmark positions are
 205 described in SI table 2.



216 Coord

217 dinates were superimposed using a Generalized Procrustes Analysis (GPA) (Goodall, 1995;
 218 Gower, 1975; Rohlf and Slice, 1990). During this procedure, sliding semi-landmarks were
 219 allowed to slide by minimizing the sum of the Procrustes distances between each individual
 220 and the mean conformation (Perez et al., 2006; Sampson et al., 1996; Sheets et al., 2004).
 221 Prior to further analyses, a Principal Component Analysis (PCA) on Procrustes residuals

222 (coordinates after superimposition) was used to visualize and quantify the shape heterogeneity
223 of individuals (Bookstein, 1991; Cooke and Terhune, 2015; Dryden and Mardia, 1998;
224 Zelditch et al., 2012). Size was quantified as the log-transformed centroid size in all analyses.
225 All analyses were performed first by using only the homologous landmarks and then, by using
226 the complete protocol including the full set of coordinates, hence combining the landmarks
227 and sliding semi-landmarks. This allows us to compare performances of a simple protocol
228 with that of a more complex one, which is more time-consuming and potentially more
229 affected by wear. All raw coordinates are available in supplementary material (SI-1 for the
230 mandibles, SI-2 for the lower third molars).

231

232 *3.2 Repeatability test*

233 In order to quantify error measurement, third lower molars and mandibles from five
234 individuals of the French breed “Blanche du Massif Central (BMC)” were photographed 3
235 times, and coordinates were acquired 3 times on each picture by a single person (MJ). In
236 addition, since two different people acquired the pictures of the remaining specimens in this
237 study (MJ and AE), the differences between operators were also assessed. The measurement
238 repeatability was quantified using Procrustes ANOVAs (Claude, 2008; Evin et al., 2020).

239

240 *3.3 Inter species comparison*

241 Differences in the mandible and the third lower molar between sheep and goat were first
242 visualized using boxplot for size and principal component analysis (PCA) for shape. Linear
243 discriminant analysis (LDA), paired with a leave-one-out correct cross-validation (CVP),
244 were then performed in order to obtain classification accuracy. LDA was performed
245 separately for size and for shape. LDA on shape data were computed on the first PCA axes
246 maximizing the between species discrimination while taking into account unbalanced group
247 sample sizes (using the "mevolCVP" R function (Evin et al., 2013)) Visualisation of shape
248 changes along the discriminant axis were performed following Claude (2008). Allometry was
249 explored using Procrustes MANCOVAs within species while the homogeneity of allometric
250 trends among the two species was explored using a 2-way Procrustes MANCOVA (using
251 shape as the dependent variable, the log centroid size as a covariate and the species as a
252 factor). Allometry free shape was then calculated using a multivariate regression.

253

254 *3.4 Effect of age and sexual dimorphism*

255 Differences in size and shape between age categories and between sexes were tested using
256 Procrustes ANOVAs. First, tests were performed for each species separately using one-way
257 ANOVAs for overall and pairwise comparisons. The homogeneity of size and shape variation
258 through age and between sexes was explored using two-way Procrustes ANOVAs, utilising
259 respectively size or shape data as the dependent variable, age or sex as first factor and, species
260 as second factor. Differences between age categories were explored for categories ranging
261 from 1-2 years to 8-10 years for the mandibles and from 2-4 years to 8-10 years for the third
262 lower molar (table 1). Similarities between sheep and goat for the various age categories were
263 visualised by a neighbour-joining network computed on Mahalanobis distances.

264

265 *3.5 Identification of archaeological specimens*

266 Archaeological specimens were superimposed along the modern specimens, and a linear
267 predictive discriminant analysis was performed on shape data. The identification of the
268 archaeological specimens was based on a resampled and balanced sample size (Evin et al.,
269 2015) ('pldam' function)); this in order to avoid the effect of uneven sample size due to the
270 higher number of sheep present in the modern dataset. In addition, four experienced
271 zooarchaeologists provided a first taxonomic identification (*i.e.* sheep, goat, or unknown)
272 mainly based on the discrete morphological criteria proposed by Halstead et al. (2002). This
273 was based solely on the pictures of the occlusal and buccal views of the teeth and not on the
274 specimens themselves. We then compared the GMM identifications with those based on
275 visual observation.

276

277 When multiple comparisons were made, the p -values of the tests were adjusted according to
278 the Benjamini Hochberg method in order to avoid false recovery rate (Benjamini and
279 Hochberg, 1995). All the analyses were carried out using the R language (R Development
280 Core Team, 2012), the packages 'MASS' (Venables and Ripley, 2002), 'Morpho' (Schlager,
281 2017) and 'Geomorph' (Adams et al., 2020) and the function provided in Claude (2008).

282 **4. Results**

283 *4.1 Repeatability and measurement error*

284 Repeatability tests confirm the variation between repeated measurements to be negligible
 285 compared to the between specimens variation. The error due to both landmarking and
 286 photographing was similar for both the third lower molar and the mandible: for the third lower
 287 molar the error was 3.6% when the landmarks alone were measured and 5.1% when the
 288 complete protocol was used, while for the mandible the error was respectively 3.3% and
 289 5.7%. The inter-operator error ranged from 4.6% to 11.6% and was considered negligible.

290 *4.2 Sheep and goat differences*

291 In all comparisons, goats have smaller measurements than sheep (table 2, fig. 2). Based on
 292 size, the cross-validation percentage of the discriminant analysis is higher for the mandible
 293 than for the third lower molar (table 2). This percentage decreases slightly when only the
 294 landmarks are used (as opposed to the use of the complete protocol, though the confidence
 295 intervals overlap). Based on the size, sheep and goat can be identified with an accuracy, at its
 296 best, of 62.7% (CI: 57.1-68.6%) for the third lower molar and 84.0% (CI: 81.6-86.8) for the
 297 mandible.

298
 299 *<Table 2>: Differences in size, shape, form, allometry, and allometry free shape between
 300 sheep and goat for the third lower molar and the mandible. Procrustes ANOVA results and
 301 correct cross-validation percentages (CVP) of the discriminant analysis are provided. For
 302 allometry, only the interaction term of the 2-way ANOVA, comparing the allometric trend
 303 between the two species, is provided. The p-values in bold are significant ($p < 0.05$) after
 304 adjustment for multiple comparisons.*

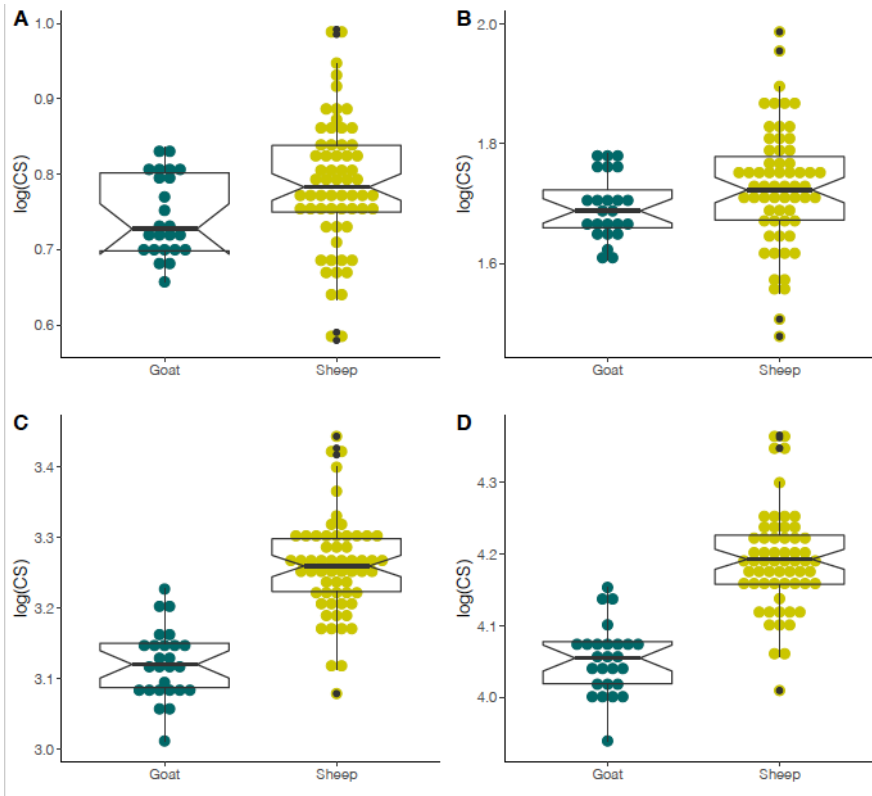
305

		landmarks		complete	
		Test	CVP	Test	CVP
Mandible	Size	F=7.3456, p=0.001	83.9 [81.6-86.8]	F=5.5993, p=0.001	84 [81.6-86.8]
	Shape	F=12.138, p=0.001	84.1 [80.3-88.6]	F=8.8521, p=0.001	95.2 [92-97.4]
	Form	F=12.248, p=0.001	84.5 [80.3-88.2]	F=9.3447, p=0.001	95.3 [92.1- 97.4]
	Allometric trend	F=1.9823, p=0.038		F=1.7404, P=0.067	
	Allometry free shape	F=4.4066, p=0.001	71.7 [67-76.4]	F=3.6787, p=0.001	75.8 [71-80.3]
Third lower molar	Size	F=63.756, p=0.001	59 [54.3-64.3]	F=63.072, p=0.001	62.7 [57.1-68.6]
	Shape	F=12.457, p=0.001	89.2 [85.6-92.9]	F=9.1407, p=0.001	93.3 [90- 95.7]
	Form	F=11.855, p=0.001	88.6 [84.3- 92.9]	F=8.6862, p=0.001	93.1 [90-97.1]

Allometric trend $F=1.5911, p=0.141$ $F=0.9588, p=0.4$
 Allometry free shape $F=11.591, p=0.001$ $88.2 [84.3-91.4]$ $F=9.1747, p=0.001$ $91.3 [87.1-95.7]$

306
 307
 308
 309

<Figure 2>: Boxplots showing the differences in size for the third lower molar (A-landmarks only, B- complete protocol) and the mandible (C and D respectively) between sheep and goat.



310

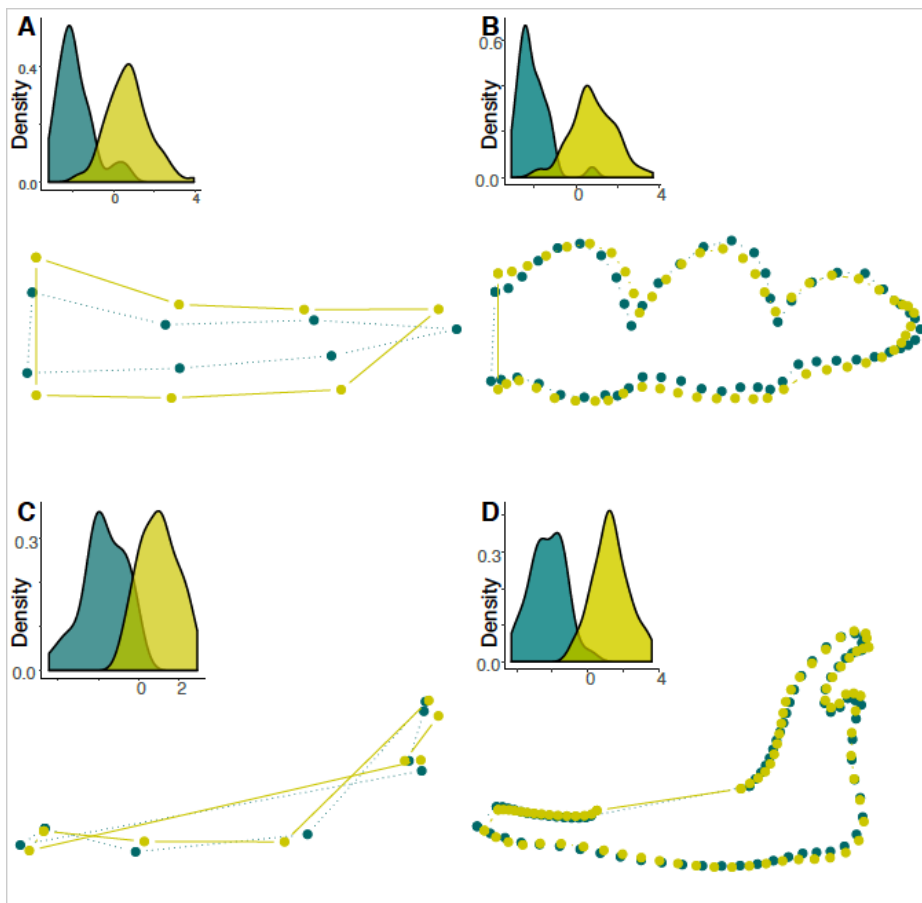
311 Principal component analysis on shape revealed a strong overlap between sheep and goat (SI-
 312 3) on the first two axes. However, the two species differ in both their mandibular and third
 313 lower molar shape regardless of the protocol used (table 2). The identification accuracy of
 314 shape data is higher than those obtained for size. An exception to this pattern is the mandible
 315 when quantified only with landmarks as it shows the same cross validation percentages for
 316 size and shape (table 2). For shape, the use of the complete protocol provides higher
 317 percentages of correct classification when compared to the results obtained when only
 318 landmarks were used, with the cross-validation percentage reaching 93.3% (CI: 90.0-95.7%)
 319 for the third lower molar and 95.2% (CI: 92.0-97.4%) for the mandible (table 2). Form
 320 analysis provides cross-validation percentages very similar to those obtained for shape (table
 321 2). Sheep and goats show similar allometric trends for all comparisons except for the third
 322 lower molar when measured only with landmarks (table 2). For both protocols, the
 323 relationship between size and shape appeared relatively weak: for the mandible, when

324 measured with landmarks, the adjusted R^2 value is 0.3% while it increases to 6.0% when
 325 measured with the complete protocol. When the third lower molar is considered, the R^2 value
 326 is 17.0%, when only landmarks are included, and of 5.0% when the complete protocol is used.
 327 Allometry-free shape provided lower correct cross-validation percentages compared to those
 328 obtained for shape (including allometry) for the mandibles but this is not the case for the third
 329 lower molar which shows similar cross validation percentages in both allometry-free shape
 330 and shape including allometry (table 2).

331

332 <Figure 3>: Shape differences between sheep (light green) and goat (dark blue). The
 333 distribution of the specimens along the discriminant axis and the visualisation of the shape
 334 differences between the two species are represented for both the lower third lower molar (A-
 335 landmarks only, B- complete protocol) and the mandible (C-landmarks only, D- complete
 336 protocol).

337



338

339 Along the between species discriminant axis (fig. 3), goats have, compared to sheep, a
 340 proportionally thinner third lower molar, with a proportionally reduced mesial edge and a
 341 more elongated distal lobe (fig. 3.A and 3.B). The goat mandibular ramus is proportionally

342 thinner compared to the one of sheep, this is especially visible in the mandibular condyle and
 343 notch. The angle formed by the ramus and the corpus of the mandible is also proportionally
 344 slightly acuter for goats than sheep (fig. 3.C and 3.D).

345

346 *4.3 Effect of age and sex*

347 When all age categories were compared, the analysis revealed that age has little influence on
 348 the mandible and the third lower molar size on both protocols; the only exception is the third
 349 lower molar when measured by using the complete protocol (table 3, fig. 4). Pairwise
 350 comparisons of age categories did not reveal significant differences in size (SI table 3).

351

352 Age affects the third lower molar shape of both species, but only sheep mandibular shape
 353 (table 3). Pairwise comparisons of age categories detect differences in shape that always
 354 affect the age category 8-10 years (SI table 3). Dissimilarity networks between species and
 355 age categories confirmed a closer proximity between species than age categories (fig. 5). The
 356 two species share homogenous changes through age except for their third lower molar shape
 357 when measured with the complete protocol (table 3). If the 8-10 years age category is
 358 removed from the analysis, the interaction term between shape, age and species becomes non-
 359 significant (landmarks: $F= 1.23, p=0.261$, complete protocol: $F=1.32, p=0.183$).

360

361 *<Table 3>: Influence of age on tooth and mandible size and shape. Results of one-way*
 362 *Procrustes ANOVAs for sheep and goat separately, and of two-way ANOVAs for testing the*
 363 *interaction between age and species (only the interaction term is provided). The p-values in*
 364 *bold are significant ($p < 0.05$) after adjustment for multiple comparisons.*

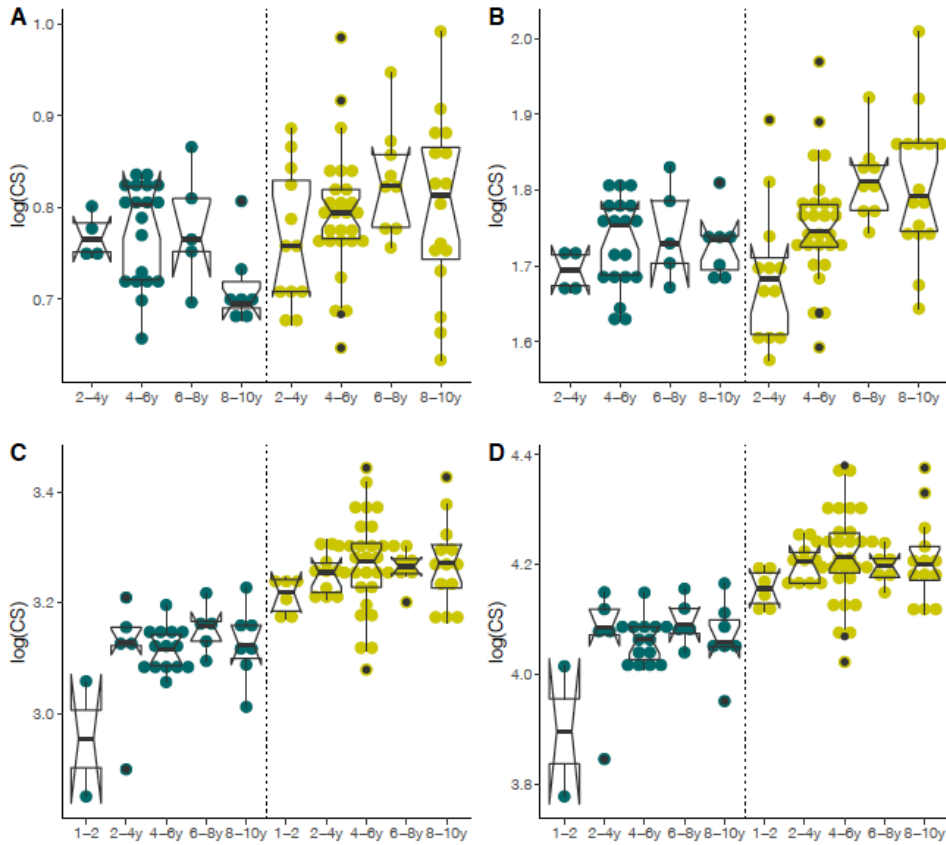
365

		Mandible				Third lower molar			
		Landmarks		Complete		Landmarks		Complete	
		F	p	F	p	F	p	F	p
Sheep	Shape	1.9321	0.006	2.1427	0.003	9.1306	0.001	12.25	0.001
	Size	0.9491	0.418	0.8754	0.468	1.1792	0.339	5.3384	0.003
Goat	Shape	1.7216	0.034	1.5906	0.026	3.0503	0.004	3.707	0.001
	Size	3.1687	0.038	3.0543	0.042	2.4466	0.081	0.6088	0.577
Interaction	Size	1.1442	0.298	1.3072	0.234	1.4376	0.225	1.1445	0.321
	Shape	1.1183	0.198	1.0870	0.243	1.7083	0.028	2.1501	0.003

366

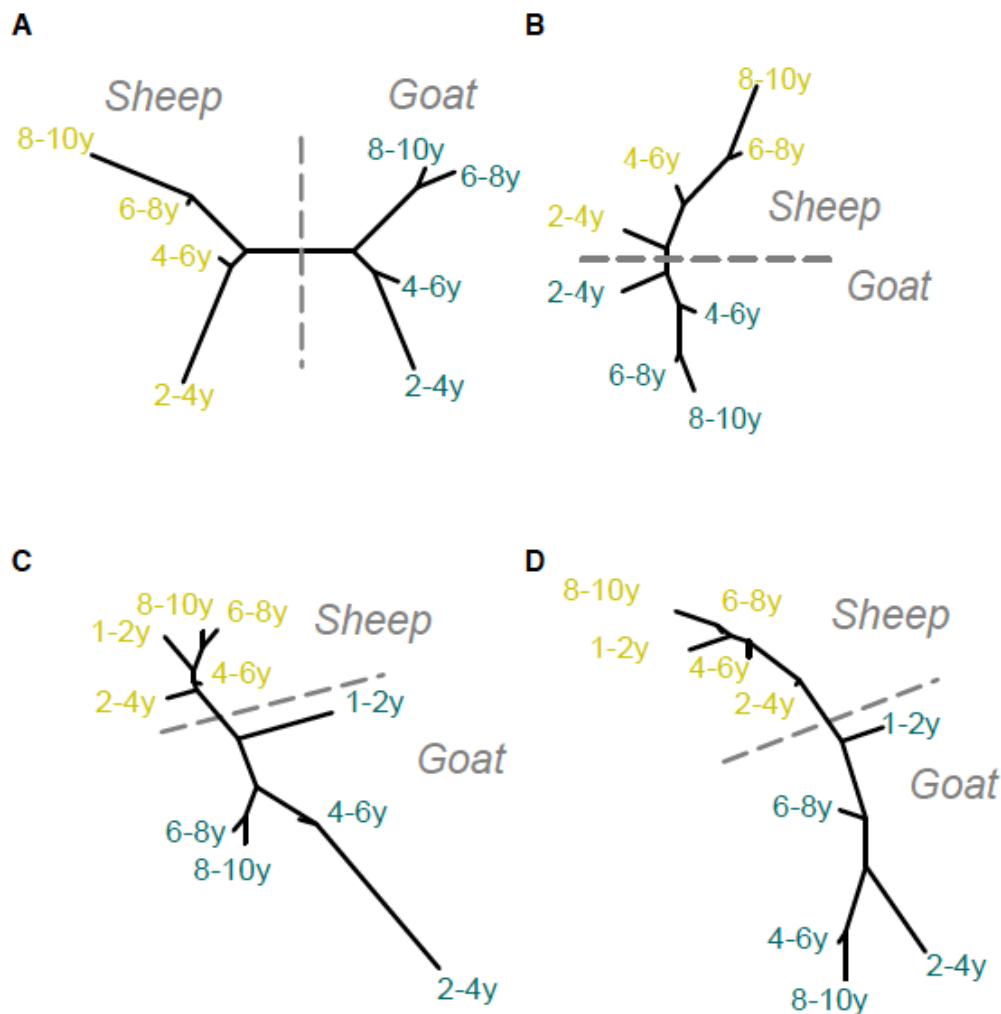
367

368 <Figure 4>: Boxplots showing size ($\log(CS)$, logarithm of the centroid size) variation
 369 through age in the mandible (C-landmarks only, D- complete protocol) and the third lower
 370 molar (A-landmarks only, B- complete protocol) of sheep and goat. Goat is in dark blue (on
 371 the left) and sheep in light green (on the right). The two species are separated by a dotted
 372 line.



373
 374 <Figure 5>: Third lower molar and mandibular shape variation through age: Dissimilarity
 375 networks between age classes for the lower third lower molar (A-landmarks only, B- complete
 376 protocol) and mandible (C-landmarks only, D- complete protocol). Goat is in dark blue and
 377 sheep in light green. The two species are separated by a dotted line.

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382 Sheep and goat show sexual dimorphism in the size of their lower third molar, with the males
383 having larger teeth than their female counterparts (table 4, fig. 6). Males and females sheep
384 also differ in their molar and mandible shape, but this sexual dimorphism impact the between
385 species differences only for the mandible when measured only with landmarks (table 4).

386

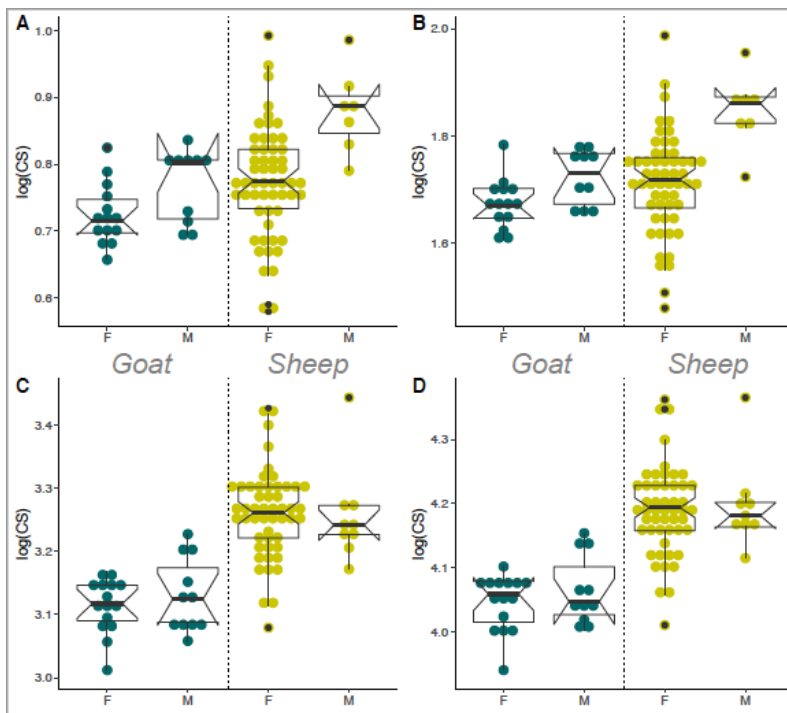
387 <Table 4>: *Sexual dimorphism in tooth and mandible size and shape. Results of one-way*
388 *Procrustes ANOVAs for sheep and goat separately, and of two-way ANOVAs used for testing*
389 *the homogeneity of sexual dimorphism (only the interaction term is provided). The p-values in*
390 *bold are significant ($p < 0.05$) after adjustment for multiple comparisons.*

		Mandible				Third lower molar			
		Landmarks		Complete		Landmarks		Complete	
		F	p	F	p	F	p	F	p
Sheep	Shape	5.2088	0.001	3.5185	0.002	3.4913	0.023	2.9595	0.04
	Size	0.0182	0.896	0.023	0.889	10.666	0.004	13.359	0.002
Goat	Shape	0.935	0.468	0.8592	0.563	0.706	0.599	0.815	0.524

	Size	0.7409	0.38	1.2001	0.269	4.7747	0.032	6.5906	0.017
Interaction	Size	0.3414	0.571	0.1145	0.739	2.0450	0.147	3.3232	0.062
	Shape	3.0138	0.003	1.7987	0.052	1.2837	0.225	1.0832	0.316

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<Figure 6>: Boxplots showing sexual dimorphism in the size of the mandible (C-landmarks only, D- complete protocol) and the lower third molar (A-landmarks only, B- complete protocol). Goat is in dark blue, and sheep in light green. The two species are separated by a dotted line. F: female, M: male.



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407 *4.4 Aimargues-Missignac: Archaeological application*

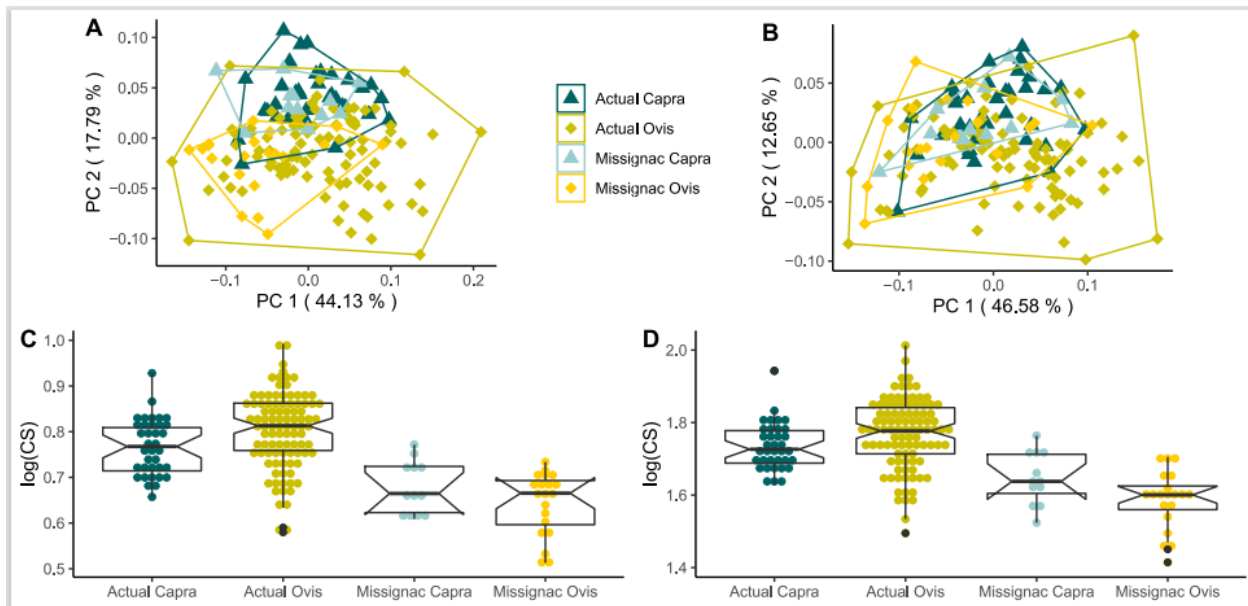
408 The geometric morphometric identification of the 32 archaeological third lower molars
409 revealed a mixed assemblage dominated by sheep (62.5%) and followed by goat (37.5%).
410 Two specimens were left unidentified since they were identified with high probability (81.5%
411 to 100%) as belonging to either sheep or goat depending of the protocol used (SI-table 4).
412 These geometric morphometric identifications match only partially (between 37.5% and
413 62.5%) the macroscopic identifications (SI table 4) that also varied depending on the
414 archaeozoologist who carried them out.

415 The shape of the archaeological specimens overlap with those of both modern sheep and goat
416 based on the two first axes of the PCAs (Fig 7.A, 7.B) that represents respectively 61.9%
417 (landmarks data) and 59.4% (complete protocol) of the total variance in the sample. The
418 archaeological specimens were, on average, smaller than the modern sheep and goat (all
419 $p=0.001$, Fig. 7). In addition, both the archaeological specimens identified as sheep
420 (Landmarks: $F=67.59$, $p=0.001$ and complete protocol: $F=59.21$, $p=0.001$) and goat
421 (Landmarks: $F=19.93$, $p=0.001$, complete protocol: $F=15.83$, $p=0.002$) were smaller than their
422 modern counterparts. We detected no size differences between the sheep and goat
423 archaeological specimens (all $p<0.001$).

424

425

426 *<Figure 7>: Top: shape variation between the modern and the archaeological specimens*
427 *identified as sheep and goat. Two first axes of PCAs based on the landmarks only (A) and the*
428 *complete protocol (B). Bottom: boxplots showing size variation of the third lower molar (C-*
429 *landmarks, D- complete protocol).*



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432

5. Discussion

433

5.1 Repeatability of the protocols

434

Our two protocols for measuring third lower molar and mandibles were found to be repeatable, with the variation between replicates taken by the same operator being smaller than the variation between the specimens used in the repeatability test. In our study we found that the cumulative error in picturing and landmarking is, at its highest, around 11%. This is relatively similar to other protocols such as the one used by Evin et al. (2020) to measure pig third lower molars were inter-operator error averaged around 13% when both landmarks and sliding semi-landmarks were used. In addition, our study showed that the amount of error increases only marginally when pictures are acquired by multiple operators.

442

443

5.2 Sheep/Goat differences

444

Our study revealed clear differences between modern sheep and goat based on the size and shape of their third lower molar and mandible. Specimens can be correctly identified with up to 93.3% probability for the third lower molar, and 95.2% for the mandible. Modern sheep have larger measurements than goat, which is congruent with previous studies on postcranial elements (Fernandez, 2001; Haruda, 2017). As far as size is concerned, the correct cross-validation percentages range from as low as 59.0% for the third lower molar (landmarks) to up to 84.0% for the mandible (complete protocol). Sheep and goat bones are also known to

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451 vary in size diachronically (*e.g.* Davis, 2008; Espinet et al., 2021; Grau-Sologestoa, 2015).
452 From the results provided by the analysis of 143 modern specimens from 13 different breeds,
453 we conclude that the lower third molar size has a low discriminatory power and, as such, it
454 has a very limited use for the identification of sheep and goat archaeological specimens. The
455 size of the mandible, on the other hand, has provided more promising results however, it must
456 be bore in mind that this element is less likely to be found complete in high numbers in
457 archaeozoological assemblages than isolated teeth, though with some exceptions.

458

459 Importantly, sheep and goats also differ in the shape of their third lower molar and mandible.
460 Specimens can be correctly identified to species level with probability ranging from 84.1% to
461 95.2% for the mandible, and from 89.2% to 93.3% for the third lower molar (depending on
462 the protocol used). Correct cross-validation percentages of the discrete morphological criteria
463 proposed by Halstead et al. (2002) range from 63.8% to 85.4% for the criteria on the third
464 lower molar, while the two mandibular criteria provided a percentage of 84.2 and 88.3 of
465 correct identification (% derived from Halstead et al. (2002) table 2). A similar approach was
466 used by Zeder and Pilaar (2010) and provided correct cross-validations ranging from 42.9% to
467 89.5% using individual criteria (% derived from Zeder and Pilaar (2010) table 2; values were
468 adjusted to take into account the non-identified specimens). Identifications carried out by
469 using a combination of individual diagnostic criteria, practice routinely adopted in
470 archaeozoology, allowed from 82.8% to 100% correct identifications for the third lower molar
471 and from 71.3% to 78.1% correct identifications for the mandible (% derived respectively
472 from Zeder and Pilaar (2010) table 4 and Gillis et al. (2011), table 10). Although not based on
473 the same sample and statistical approach (*e.g.* our approach takes into account unbalanced
474 sample sizes), our geometric morphometric protocols performed at least as well as the discrete
475 criteria used in isolation.

476 The shape differences we observed using the geometric morphometric protocols reflect the
477 variations described by the discrete morphological criteria. Among the criteria proposed by
478 Halstead et al. (2002), three clearly mirror our observations. According to Halstead et al.
479 (2002:547) “the buccal edge of the centro-buccal cusp of third lower molar often points
480 strongly in a posterior direction in goat, while it is relatively symmetrical in sheep” (criterion
481 M3.2) and this is reflected in the fact that the distal part of the centro-buccal cusp is
482 proportionally thicker in goats than in sheep. This criterion correctly identified the specimens
483 with a probability of 76.7% (% derived from Halstead et al. (2002) table 2). The second
484 criterion on the third lower molar is that “the distal margin of the distal cusp of third lower

485 molar often has a buccally defined “flute” in sheep, rarely so in goat” (M3.5 of Halstead et
486 al. (2002:549)). This criterion provided 84.1% of correct identification (% derived from
487 Halstead et al. (2002) table 2) and it mirrors the fact that sheep have a proportionally more
488 pointed distal part of the distal cusp compared to goats. The proportionally reduced mesial
489 edge in goat compared to sheep could also reflect the Halstead et al. (2002:549) M3.6
490 criterion described as “The flange on the mesial face of third lower molar tends to be broad in
491 sheep and narrow in goat”, (70.9% of corrected identification, derived from table 2), noticed
492 also by Balasse and Ambrose (2005). Conversely to Halstead et al. observations (2002), when
493 the shape of the mesial part of the buccal edge of the mesio-buccal cusp (M3.1), and the shape
494 of mesial and central part (M3.3) were considered, we did not notice shape differences
495 between sheep and goat.

496

497 Size and shape appeared only weakly correlated, with the allometric trends being mainly
498 homogeneous between sheep and goats. While the third lower molar form and allometry-free
499 shape performed equally as well as shape in separating sheep and goat, mandibular allometry-
500 free shape was less efficient to do so than shape and form. Allometries are, therefore, at least
501 partially involved in the mandibular shape differences noticed between the two species.

502 Two different protocols were used on both the third lower molar and the mandible, one based
503 only on landmark data, the second also including sliding semi-landmarks. In both cases
504 landmark data provided lower correct cross-validation percentages and a simplified
505 description of the shape differences compared to the results obtained when the complete
506 protocol was used. However, to its advantage, the landmark protocol is quicker and simpler to
507 apply and correctly identified the two species with a high probability for both for the third
508 lower molar (84.1%) and the mandible (89.2%).

509

510 *5.3 Age*

511 Because of the well-known influence of wear on ungulate hypsodont teeth, in the past, it has
512 been assumed that a geometric morphometric approach on teeth could not work. However, a
513 recent geometric morphometric study of the upper second permanent premolar and third
514 lower molar of domestic horses has revealed that age has no effect on the size and shape of
515 their occlusal folding pattern (Seetah et al., 2014). Conversely, the study by (Cucchi et al.,
516 2019) on bovid molars revealed that age-related variations in size and shape do exist, but are
517 homogeneous between species. Therefore, hypsodont teeth appear perfectly suitable for

518 geometric morphometric studies and such an approach will most likely be extensively applied
519 in the near future.

520

521 In this study, age appeared to have a limited impact on the size and shape of both the third
522 lower molar and the mandible in comparison to the differences between the two species. Most
523 of the differences noticed affected older specimens, *i.e.* 8 to 10 years old animals; such old
524 animals are usually not the most abundant in the archaeological record (*e.g.* Blaise, 2005;
525 Payne, 1973) and, even in modern husbandry practices animals are not keep alive for so long
526 (Blaise, 2006). In particular, only the third lower molar shape showed some age-related
527 differences between sheep and goats in the above mentioned age category. However, there is
528 less variation between age groups than between species, revealing that age has little impact on
529 distinction between taxa. Available discrete morphological criteria for the distinction of sheep
530 and goat teeth are highly dependent on age (Zeder and Pilaar, 2010) but can be applied to
531 younger specimens (0 to 1.5 years of age) than those composing our sample (2 to 4 years old
532 for the teeth and 1 to 2 years old for the mandible). The age-related mandibular variations we
533 observed appeared less pronounced than those observed in the study conducted by Parés-
534 Casanova (2013) where shape differences between age categories, especially visible on the
535 molar row, were due to allometry and linked to morpho-functional changes.

536

537 *5.4 Sexual dimorphism*

538 Our study confirmed that generally, despite some overlap between sexes, in both species,
539 males have larger third lower molars than females. In addition, our results suggest that the
540 molar size of male goats overlap with that of female sheep. Although further analyses and
541 additional samples are needed to confirm the observed trend, our study seem to suggest that
542 size differences between sexes may be higher for sheep than for goat.

543 According to previous literature, the ratio of body to tooth size is greater for females than
544 males (Carranza and Pérez-Barbería, 2007; Fortelius, 1985); this is the case for ungulates but
545 also other mammals (Cochard, 1987; Lucas, 2004; Lucas et al., 1986). Sheep and goat are
546 known to be sexually dimorphic species (Pérez-Barbería and Gordon, 1999), even if they
547 show less sexual dimorphism in size than their wild relatives, with an almost similar size
548 reduction in both species (Polák and Frynta, 2009). The same study also revealed that sheep
549 and goat breeds follow the Rensch's rule, according to which larger species exhibit higher
550 sexual size dimorphism (Rensch, 1959, 1950). Several studies have demonstrated that this

551 rule has exceptions depending on the species and the element considered. For example, while
552 goat shows sexual size dimorphism in the skull (Parés-Casanova, 2015), sheep do not (Parés-
553 Casanova, 2014). The opposite pattern is true for shape with male and female sheep showing
554 the most differences in the tibia, metapodials, femur, pelvis, radius and humerus (with females
555 being less variable than males) (Popkin et al., 2012), while goats showed none. Clearly, not
556 all skeletal elements display to the same extent sexual dimorphism.

557 In addition, it cannot be excluded that, the absence of significant sexual dimorphism in goats
558 is the result of a smaller sample size for this species compared to sheep. Moreover, we did not
559 explore the effect of castration known to have an impact on animal size (Davis, 2000; Popkin
560 et al., 2012), as this information was not always recorded in the collections used for this
561 study.

562

563 *5.5 Archaeological specimens*

564 During the Middle Age, inhabitants from Aimargues Missignac ate mainly cultivated and wild
565 fruits, with a meat diet based mostly on the main domestic animals. Caprine remains were
566 abundant at this site, with sheep and goat forming respectively 85% and 15% of the total of
567 the identified specimens (Bardot-Cambot et al., 2018), but with high variation between
568 skeletal elements (*e.g.* 72% of mandibles and 100% of metacarpals were identified as sheep)
569 (Bardot-Cambot et al., 2018; Mureau, 2020). The geometric morphometric identifications
570 revealed a mixed assemblage dominated by sheep (62.5% sheep and 37.5% goat), but with a
571 larger proportion of goat than originally identified. However, the initial archaeozoological
572 study focussed only on a selection of the total assemblage (Bardot-Cambot et al., 2018).
573 When we restricted our geometric morphometric analyses to the specimens coming from the
574 same contexts as analysed in (Bardot-Cambot et al., 2018), the proportion of identified sheep
575 amounted to 76%, a percentage that is in line with the previous study (72%). It should also be
576 mentioned that our study did not include the very young individuals which were part of the
577 previous archaeozoological analysis (Bardot-Cambot et al., 2018) and that might explain the
578 small discrepancy between the two studies. Regardless, the sheep-goat ratio that our study has
579 revealed fits very well into the regional pattern: during the medieval period in Languedoc,
580 sheep are usually more numerous than goat (Forest, 1997) though goat can be locally
581 particularly abundant (Forest et al., 2004; Rodet-Belarbi et al., 2002).

582 Finally, it should also be mentioned that, because of the nature of the discrete morphological
583 criteria used to identify sheep and goat third lower molars, identifications may vary from one

584 experienced archaeozoologist to another. Those macroscopic identifications also differ from
585 the ones obtained through geometric morphometrics. As a consequence, future research may
586 require the additional use of molecular identification methods based on aDNA or
587 palaeoproteomics (*e.g.* ZooMS) to confirm identification for indeterminate specimens and
588 assess the extent to which such identification can be made for all time period and areas of the
589 world.

590 **6. Conclusions and future perspectives**

591 Geometric morphometrics provide a new and efficient way of identifying third lower molars
592 and mandibles of sheep and goat. Although based on 143 specimens of 13 modern European
593 local breeds, which do not likely reflect the full diversity of ancient sheep and goat globally,
594 this study revealed clear differences between modern sheep and goat based on the size and
595 shape of their third lower molar and mandible. In particular, this work opens new perspectives
596 when it comes to the identification of isolated teeth which are abundant but largely ignored in
597 archaeozoological studies.

598 Third lower molar size appeared to have little value for the identification of archaeological
599 specimens, contrary to mandible size that appeared more promising. Shape, on the other hand,
600 was the marker of choice for identifying archaeological specimens. Geometric morphometric
601 based identifications have the advantage of being simultaneously less dependent of the
602 operator, based on quantitative data and provide high degree of confidence in the
603 identifications. Once all caprine specimens are identified to the different genus and species, it
604 will be possible to study in detail the spatio-temporal variation in taxa proportions, to explore
605 further the relationships between human populations, domestic animal species, and past
606 husbandry practices. Further studies are needed to fully explore the discrepancy between the
607 geometric morphometric based identifications and those based on discrete morphological
608 criteria. The next step forward will be to confirm archaeological identifications through
609 ancient DNA or palaeoproteomic analysis so that it will be possible to assess to what extent
610 the different methodologies can be used with confidence. Once confirmed to be accurate
611 when applied on archaeological assemblages, this 2D GMM protocol will represent an easy to
612 set up, non-destructive, repeatable, objective and quantitative identification protocol that will
613 complement discrete morphological criteria.

614

615 **Declarations of interest:** none

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640 **9. References**

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