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1 # Palaeoecological significance of the "wolf event" as revealed by

2 # skull ecometrics of the canid guilds

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12 **ABSTRACT**

13 Members of the mammalian order Carnivora are rarely considered as proxies for

14 palaeoecological reconstructions due to their broad phenotypic plasticity and high climatic

15 tolerance. However, palaeontologists have traditionally interpreted the appearance of some

16 particular carnivoran species in relation to major climatic events. The 'wolf event'

17 characterised the inset of climatic deterioration for continental Eurasian fauna, ca 2.0 million

18 years ago. It was defined as the dispersal of cursorial wild canids from East Asia to Europe

19 consequently to the spread of the steppe environment. Anecdotal interpretations were never

20 supported by rigorous testing about the palaeoecological significance of these canid taxa.

21 Here, we employ two dimensional geometric morphometrics of canid skulls to characterise

22 and interpret the structure of extant guilds and its relationship with climate and provide

inferences on the early Pleistocene Valdarno community. Averaged skull size and shape of

canid guilds are currently associated with climatic parameters that reflect annual mean

temperature, seasonality and precipitation. The canid guild from Valdarno showed a clustered

phylogenetic structure and its average shape resembles those of communities characterised by

relatively high seasonal environments. This study supports the use of canid community

structure for palaeoclimatic reconstructions and validate the early Pleistocene as a period of

climatic deterioration for Eurasian large mammal fauna.

1. Introduction

Faunal responses to environmental changes are well documented across many

mammalian groups (Blois and Hadly, 2009). Primary consumers such as ungulates or rodents

are expected to be directly influenced by vegetation showing individualistic (Jernvall et al.,

1996; Eronen et al., 2010; Damuth and Janis, 2011) and group response to major climatic

events (Fortelius et al., 2002; Mihblacher et al., 2011; Schap et al., 2021). In turn, secondary

consumers such as members of the order Carnivora (=carnivorans) have a comparatively

good record of biological trait changes (i.e., body size) in relation to climate (Klein and Scott,

1989; Meiri et al., 2004, 2009; Clauss et al., 2013), but variation at the community level

through space and time is not well understood. Recently, the employment of ecometrics

(Polly et al., 2011) has provided an explicit tool to test for the response of averaged biological

traits across animal communities through space and time. Polly (2010) and Polly et al. (2017)

47   demonstrated that calcaneal gear ratio in Carnivora is a good proxy for locomotor behaviour

48   as its changes at community level can be tracked in relation to climate on a continental scale.

49   The same holds for the temporal scale where changes in calcaneal gear ratio have been

50   identified within fossil cat communities from North America across the Neogene (Polly,

51   2020).

52   Other aspects of carnivorans postcranial morphology have been equally investigated

53   to identify habitat and climatic adaptations in extant and fossil species (Lewis, 1997; Meloro,

54   2011a; Meloro et al., 2013; Meloro and Louys, 2015; Meachen et al., 2016; Meloro and de

55   Oliveira, 2019; Tomya and Meachen, 2018) together with cranial and dental metrics. The

56   latter elements traditionally received much more attention due to their diagnostic taxonomic

57   significance. In this regard, the seminal work of Van Valkenburgh and many others (Van

58   Valkenburgh, 1985, 1988, 2007; Turner, 1990; Wesley-Hunt, 2005; Meloro, 2011b)

59   supported ecomorphological stasis of the carnivoran feeding morphologies across time.

60   Terrestrial carnivores evolved distinct feeding adaptations very early in their evolutionary

61   history (Meloro and Raia 2010; Slater and Friscia, 2019; Meloro and Tamagnini, 2021) thus

62   showing little variation at community level through space and time (Meloro, 2011a). On the

63   other hand, Dalerum et al. (2009) and Dalerum (2013) noted that modern carnivore guilds are

64   functionally depleted in several continents and their functional diversity and taxonomic

65   richness changes at large continental scale. This suggests that trophic morphology can be

66   confidently used as a proxy to detect subtle changes in carnivore community structures linked

67   to complex evolutionary processes (Dalerum, 2013).

68   The end of Pliocene/beginning of Pleistocene was traditionally identified as an important

69   benchmark for the Eurasian continental fauna due to the renewal of the carnivoran guilds

70   (defined as a group of species that exploit the same class of environmental resources in a

71   similar way, Simberloff and Dayan, 1991) concomitant with dramatic climatic changes.

72   Azzaroli (1983) proposed the term "wolf event" to characterise the dispersal of large

73   cursorial canids, i.e., *Canis etruscus*, *Canis arnensis*, and *Lycaon falconeri*, (Rook and Torre,

74   1996; Rook and Martinez-Navarro, 2010), from Asia to Western Europe and the

75   Mediterranean region occurring ca 2.0 Ma (Azzaroli et al., 1988; Masini and Torre, 1990;

76   Napoleone et al., 2001; Augustì and Antòn, 2002). Re-evaluation of the canid fossil record

77   supported members of the genus *Canis* to sporadically appear much earlier than 2.0 Ma in the

78   Chinese and European fossil record (Sotnikova and Rook, 2010; Jangzuo, 2021) and the

79   "wolf event" has now been substituted by the "*Pachycrocuta brevirostris* event" of 1.8 Ma,

80   due to the over-abundance of the giant hyena species in fossil sites (Martínez-Navarro, 2010;

81   Rook and Martínez-Navarro, 2010). The Miocene rise of canid diversity has been equally

82   linked to environmental changes in North America (Figueirido et al., 2015) due to the

83   evolution of highly cursorial forms concomitant with the spread of the grassland ecosystem.

84   Emergence of this pattern left open the question about association between canid

85   morphological diversity and climate. Several authors provided insights into the

86   ecomorphological characterisation of the Plio-Pleistocene dogs (Brugal and Boudadi-

87   Maligne, 2010; Meloro, 2011c; Cherin et al., 2013, 2014; Flower and Schreve, 2014;

88   Bartolini Lucenti and Rook, 2016; Jiangzuo et al., 2018; Koufos, 2018; Bartolini Lucenti et

89   al., 2021, 2020, 2017; Bartolini Lucenti and Spassov, 2022), but still a re-evaluation of their

90   palaeoecological significance for palaeoclimatic reconstruction is needed.

91   Here, we employ 2D geometric morphometrics (gmm) in conjunction with comparative

92   methods to test for association between canid morphological diversity and climate in modern

93   ecosystems. Recent work already highlighted for South American canids a significant

94   association between cranial morphology and climatic adaptation at species (Zurano et al.

95   2017) and community level (Bubadué et al., 2016). Such a test based on worldwide canid

96   guilds will allow us predicting climatic adaptations of Plio-Pleistocene canid community that

characterised the "wolf event". Meloro et al. (2015) already demonstrated that gmm of the

ventral cranium successfully characterise feeding ecology of extant and fossil canids and this

will be equally employed here to investigate trophic diversity at guild level.

2. Material and methods

2.1 Sample

We collected data for 105 skulls of extant canid species (see Meloro et al. 2017 for

specimen details) and four complete fossil skulls belonging to three species of the Early

Pleistocene (= Upper Villafranchian) Valdarno faunal community mainly housed in Natural

History Museum, Geology and Paleontology section, University of Florence (Italy): *Canis*

etruscus (IGF 12867), *Canis arnensis* (two specimens, IGF 601 and IGF 867) and *Lycaon*

falconeri (AMNH 97052 in Rook 1994 [Plate 1, page 73]). Although this specimen is

referred to *Canis (Xenocyon) antonii* it represents the only complete skull representative of

Lycaon falconeri (sensu Martínez-Navarro and Rook, 2003; but see Palmqvist et al. 1999 for

a deformed specimen from Venta Micena) and it compares well with fragmentary Valdarno

specimens so that Tedford et al. (2009) placed this specimen directly basal to the Valdarno

taxon. The extant canid dataset was categorised into 33 OTUs (= Operational Taxonomic

Unit) representative of 24 extant canid species. Multiple OTUs were selected when specimen

geographic location was available for the species with particularly large range size (e.g., the

red fox, *Vulpes vulpes* or the grey wolf, *Canis lupus*). In these cases, specimens were

averaged per geographic area (e.g., all the red foxes from North America vs the ones from

Eurasia) and treated as separate OTUs. This procedure allowed to account for (when

possible) intraspecific phenotypic variation related to geography.

120 To characterise worldwide morphological variation of canid communities, we selected

121 from the MAB database (http://www.ice.ucdavis.edu/bioinventory/bioinventory.html) 13

122 national parks representative of distinct climatic regions (a minimum of 3 canid species / site

123 was allowed, see Meloro, 2011a). These included: Lake Torne Area (Sweden, N = 3),

124 Daweishan Nature Reserve (China, N = 3), Kanha Tiger Reserve (India, N = 4), Azraq

125 Wetland Reserve (Israel, N = 3), Radom National Park (Sudan, N = 3), Dana Biosphere

126 Reserve (Jordan, N = 4), Amboseli (Kenya, N = 5), Yellowstone (Montana, N = 3), El Morro

127 National Monument (New Mexico, N = 4), Reserva de biosphere de Cerrado (Brazil, N = 3),

128 Iguazu National Park (Brazil/Argentina, N = 3), Estacion Biologica Beni (Ecuador, N = 4),

129 and Reserva Provincial San Guilllermo (Argentina, N = 3).

130 These geographic locations were representative of the extant worldwide canid guild variation,

131 and they were used as a comparative sample to characterise the fossil community from the

132 Valdarno basin (ca 1.9 – 1.83 Ma).

133

134 *2.2 Phylogeny*

135 A molecular phylogeny inclusive of 33 living OTUs was constructed using the 10k tree

136 project database (Arnold et al., 2010). We selected both species and subspecies of canids for

137 which genetic data were available. When subspecies genetic data were not present (e.g., for

138 the red fox, while for the grey wolf we included the Himalayan and Indian subspecies)

139 polytomies were constructed at the basis of the species node. Time of divergences for

140 polytomies were estimated to a minimum of 1.0 Ma, based on dates obtained for other

141 subspecies with molecular record. After generating the topology with branch lengths as time

142 of divergence for extant species, we added the position of the fossil taxa (OTU = 3) from 2.0

143 Ma following Tedford et al. (2009). Branch lengths for these taxa were based on their first

144 and last occurrence (as in Raia et al., 2013). The final tree resulted in a non-ultrametric

phylogeny inclusive of 36 OTUs of which three were fossils (see Supplementary Material,

https://data.mendeley.com/datasets/42zgw9pkz2/draft?a=788baf9c-8415-40f5-b279-

1d2f551289ef).
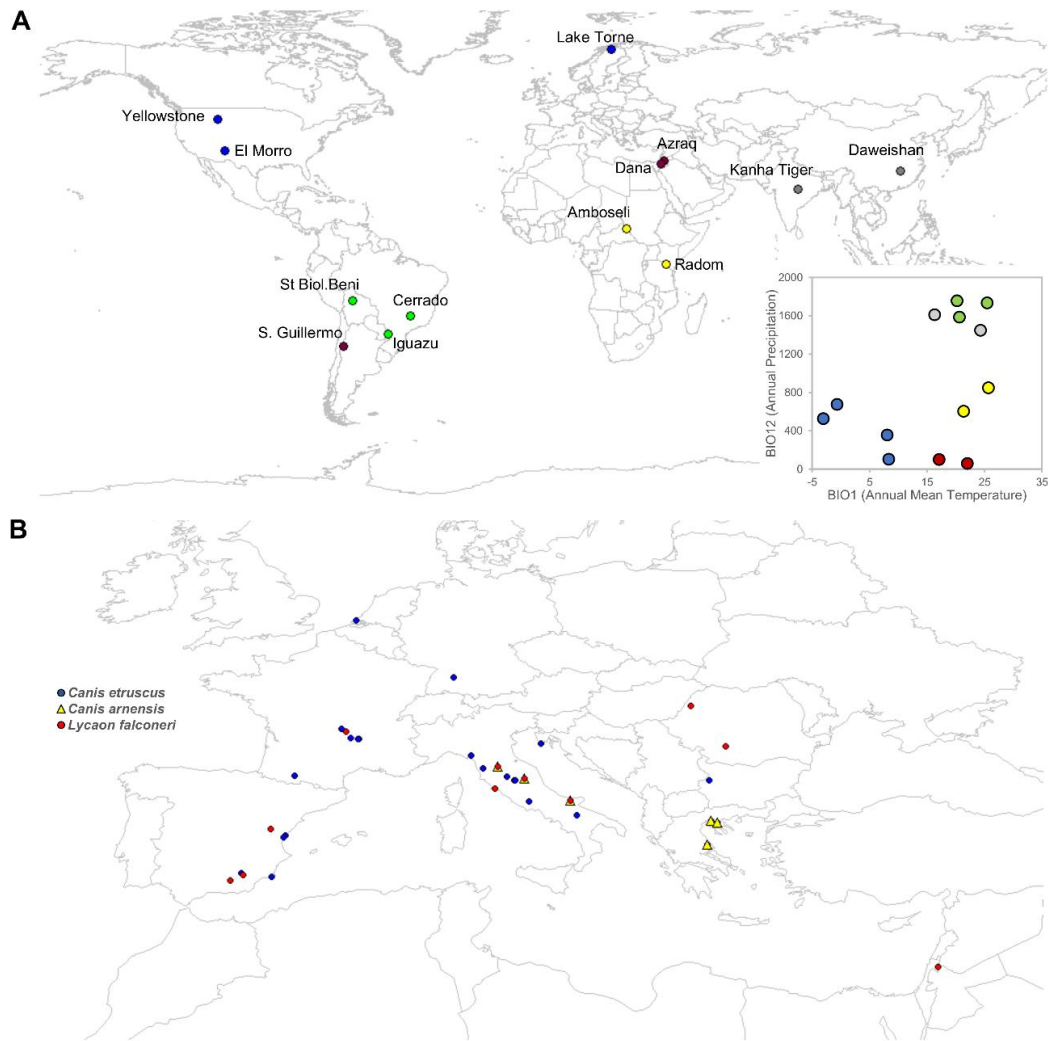
Fig. 1. Map showing the geographic locations of the 13 national parks selected to represent variation in canid guild composition worldwide (A). Locations are colour coded according to their climatic similarity. A plot of BIO1 (Annual Mean Precipitation) vs BIO2 (Annual Precipitation) indicates the general climatic cluster for the selected locations. In B the distribution of fossil sites that recorded presence of the three Pleistocene canids (*Canis etruscus*, *Canis arnensis* and *Lycaon falconeri*) based on the paleobiology database (https://paleobiodb.org/#/). Note that overlap among the three species occurs only in the Val d'Arno province (Tuscany, central Italy) and to lesser extent in the Apulia region.

2.3 Geometric morphometrics

158   The skulls were photographed in ventral view at 1 meter distance using a Nikon D40

159   attached to a Manfrotto tripod. They were placed on the floor and a spirit level was

160   positioned on the palate to ensure parallelism between the palatal plane, the floor, and the

161   camera plane (Meloro et al., 2015). The software tpsDig2 (Rohlf, 2015) was employed to

162   digitize 23 anatomical landmarks (Fig. 2) to cover palatal, dental, zygomatic and occipital

163   region. A similar configuration was employed by Bubadué et al. (2016) to investigate South

164   American canid guilds. Landmarks were digitized only by one user (CM) and a subsample of

165   three skulls was digitized three times over three different days to test for digitalisation error

166   (in all cases no differences occurred in the size and shape variables between replicas,

167   Procrustes ANOVA p = 0.99).

168
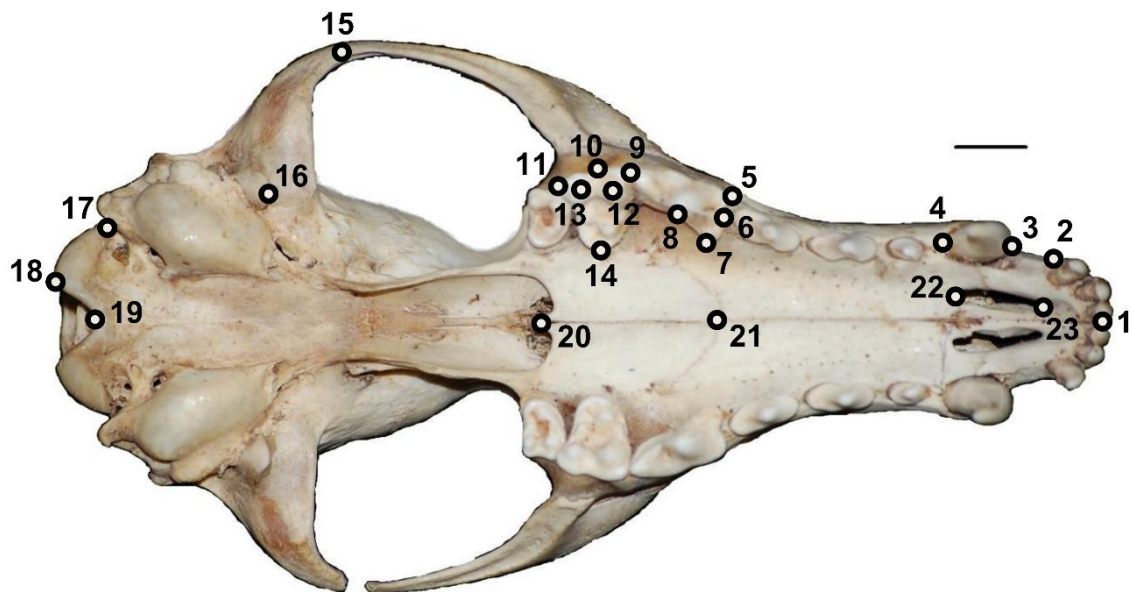
Fig. 2. Skull of *Vulpes vulpes* showing the landmark configurations employed to describe
skull size and shape in Canidae. (1) tip of the snout defined by middle point between the first
two frontal incisors, (2) posterior tip of the third incisor, (3) anterior tip of canine, (4)
posterior tip of canine, (5, 6, 7, 8, 9) outline of carnassial tooth (P4), (10-11) central and
posterior labial edges of molar one, (12, 13) cusps of molar one, (14) anterior tip of molar

174   one, (15) (17, 18, 19) outline of the occipital condyle; (20) junction of the stiff and hard

175   palate, (21) palatine-maxilla suture, (22) posterior and (21) anterior tip of the incisive

176   foramen. Scale bar equals 1 cm.

177

178   A Generalized Procrustes analysis (GPA) (Rohlf and Slice, 1990) was used to rotate,

179   translate, and scale landmark configurations to unit centroid size (CS = the square root of the

180   sum of squared distances of the landmarks from their centroid; Bookstein, 1989). To

181   visualize the multivariate ordination of the aligned specimens in relation to their relative

182   reconstructed ancestral nodes, we generated a phylomorphospace (Sydlauskas, 2008;

183   Sakamoto and Ruta, 2012) as a principal component analysis (PCA) of the procrustes shape

184   coordinates. Additionally, phylogenetic signal for size and shape data was tested using the R

185   function 'physignal' (Adams, 2014). Allometry defined as the influence of size on shape

186   variation (Klingenberg, 2016) was tested in our 36 OTUs sample using the function

187   'procD.lm' and 'procD.pgls' included in the R package "geomorph" (Adams and Otarola-

188   Castillo, 2013) to account for the phylogenetic covariance matrix (Adams and Collyer, 2018).

189   Due to some species phenotype being present in multiple guilds (e.g., *Chrysocyon*

190   *brachyurus* is found in Reserva de biosphere de Cerrado, Iguazu and Estacion Biologica

191   Beni) a 'guild' morphospace (n-taxa = 48) was generated using replicas of PC scores from

192   the 36 OTU morphospace. This allowed testing size and shape differences due to guild

193   membership using non-parametric Kruskal-Wallis (for size) and non-parametric MANOVA

194   with 9,999 permutations. To test multivariate dispersion of shape data around their centroid,

195   we computed per-group (guilds) morphological disparity (=MD) as the average Euclidean

196   distance from group centroid, and then we performed a permutation test using 'betadisper'

197   and 'permutest' functions available in the R package 'vegan' (Oksanen et al., 2013) to assess

198   the significance in disparity differences between guilds. The same procedure was applied to

199   test the dispersion of size data around their centroid.

2.4 Phylogenetic structure and environmental variables

We characterized each canid guild using metrics of phylogenetic distances between the taxa (PD), net relatedness index (NRI) and nearest taxon index (NTI) (Webb et al., 2002). These metrics are descriptors of community composition based on phylogenetic relationships observed for each canid assemblage. They have been used extensively in community ecology of extant and fossil mammals and they are better descriptors of community structure than species richness (Raia, 2010; Kamilar et al., 2015). We employed the package picante that computes PD, mnpd (= mean nearest taxon phylogenetic distance, NRI = mnpd*-1) and mnptd (= mean nearest taxon phylogenetic distance, NTI = mnptd*-1) and compares their distribution with random models generated after 9,999 permutations.

Additionally, the climate theoretically experienced by each of the 13 extant canid guilds was described using nineteen bioclimatic variables as defined in Hijmans et al. (2005). Geographic location for the centroid of each national park selected was assumed to be a good spatial proxy for their climatic condition (see Meloro, 2011b). The nineteen bioclimatic variables were extracted from the centroid of each national park using the WorldClim Database (Hjmans et al., 2005) with a 2.5 arc spatial resolution.

2.5 Ecometric association and climate prediction

Each of the 14 canid guilds (13 extant plus one fossil) analysed can be phenotypically described using ecometrics (Polly et al., 2011). Morphological disparity (=MD, the morphospace volume occupied by each community) of shape and size represents a potential ecometrics that we tested for association with phylogenetic community structure and climate using non-parametric correlations (see also Meloro, 2011a). Work by Polly (2010, 2020) and Polly et al. (2017) also highlighted the potential of using averaged traits as ecometrics, so we opted to characterize each canid guild using skull shape consensus configuration. In our case,

225   the ecometric trait was multivariate (n = 46 procrustes coordinates) and we employed

226   different strategies to test for association with phylogenetic structure and climate.

227   Firstly, we applied the clustering UPGMA (=unweighted pair group method with

228   arithmetic mean) method based on the procrustes distances between averaged skull shape of

229   canid guilds. This allowed to graphically identify similarities between canid guilds based on

230   their averaged skull shape. Same procedure was applied to generate cophenetic trees based on

231   the Euclidean distance matrices of phylogenetic community descriptors (PD, NRI and NTI)

232   and selected bioclimatic variables that were associated with averaged skull shape. Each

233   bioclimatic variable was tested for association with averaged skull shape using multivariate

234   regression with permutation and retained for further analyses, all the others were discarded.

235   Mantel test was employed to verify association between procrustes and phylogenetic and/or

236   bioclimatic distance matrices. When climatic variables were concerned, the fossil community

237   was not included in the analyses.

238   Secondly, two-blocks Partial Least Squares (Rohlf and Corti, 2000) was applied to

239   test association between averaged skull shape and the previously selected bioclimatic

240   variables. This analysis is based on a singular value decomposition of the matrix of

241   covariances between two sets of variables that in our case are the block "climate" (the

242   selected bioclimatic variables per guild, appropriately standardised) and the block "averaged

243   guild shape" (the consensus configuration obtained for each guild).

244   We employed robust regression models to reconstruct bioclimatic variables for

245   Valdarno fossil community when univariate ecometric traits (MD of size and shape) and

246   phylogenetic community descriptors were used as independent variables. A projection of the

247   Valdarno community into the PLS climate-shape morphospace was also employed and based

248   on predicted PLS1 climatic scores, bioclimatic variables were extrapolated. To obtain

249   Valdarno climatic score in PLS space we opted to use linear model and robust regression.

250 Both methods were considered to explore their suitability for climatic predictions. Robust

251 regression is generally less dependent on assumptions concerning data distribution and it is

252 much less sensitive to outlier, which can be common in palaeocological studies (Gebregiorgis

253 et al. 2020). The most likely climatic variables were identified subsequently based on the

254 accuracy of each univariate or multivariate predictor.

255

3. Results

256

3.1 Shape and size analysis

257

258 Phylomorphospace identified a clear distinction between the members of Old World

259 Canini tribes and Vulpini and New World Canini. Significant overlap occurred between Old

260 and New World foxes (Fig. 3). PC1 describes shape changes in the skull related to the

261 relative proportion of the palate. On the negative scores, *Otocyon, Nyctereutes* and South

262 American genera are characterised by very short muzzle and upper dentition, while on

263 positive PC1 wolves and fossil *Lycaon falconeri* show a broader and wider palate and upper

264 dentition. On PC2 taxa are partitioned based on the relative position of the zygomatic arch

265 with hypercarnivorous species (e.g., the arctic wolf *Canis lupus arctos*, *Lycaon* and *Cuon*)

266 exhibiting negative scores, while hypocarnivores have positive scores (Fig. 3).

267 Although low, phylogenetic signal was significant in this canid shape sample (K_{multiv}

268 0.2042, P-value: 0.001). When size was analysed, a stronger and still significant signal was

269 identified (K: 0.5023, P-value: 0.001). There was a significant impact of size on skull shape

270 in the sample of 36 OTUs (33 living + 3 fossils), however this pattern was not valid when

271 accounting for phylogeny (Table 1).

272

Table 1. Procrustes ANOVA without (OLS) and with (PGLS) phylogenetic correction to test

274 for the impact of size on shape variation in 36 canid OTUs. Significant p values are

275 highlighted in bold. The test was implemented with 9,999 permutations.

			Df	SS	MS	Rsq	F	Z	Pr(>F)
Allometry	OLS	Centroid Size	1	0.020	0.020	0.159	6.435	4.702	**0.001**
		Residuals	34	0.106	0.003				
		Total	35	0.126					
	PGLS	Centroid Size	1	0.001	0.001	0.046	1.640	0.886	0.339
		Residuals	34	0.026	0.001				
		Total	35	0.028					

276

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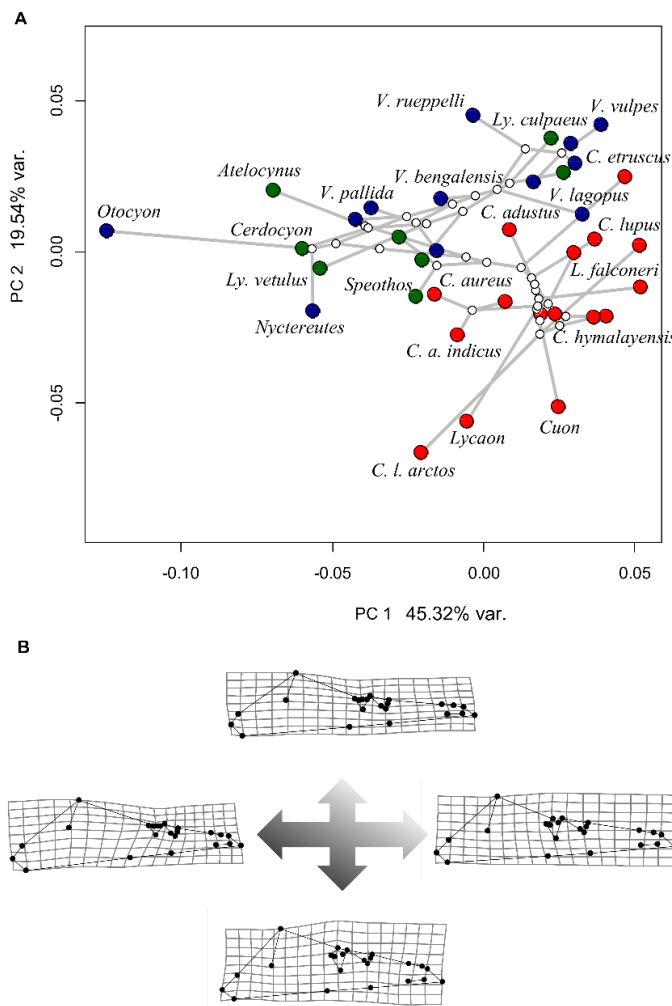
Fig. 3. A, Phylomorphospace for skulls of 33 canid taxa (OTUs). B, Thin plate spline

279 showing skull shape deformation from the most extreme negative to the positive scores along

280 PC1 (horizontal) and PC2 (vertical).

281   The 14 guilds (13 extant plus 1 fossil) did not show any difference in average skull

282   size (K-W = 10.57, p=0.647) as well as shape (Tot SS = 0.164, Within-group SS = 0.107, F =

283   1.39, p = 0.068). Similarly, the betadisper analysis returned a non-significant result when

284   performed on both size and shape variables (p-value = 0.901 and p-value = 0.246

285   respectively) showing that variances were homogeneously distributed between canid guilds

286   (Table 2).

287

288   **Table 2.** Species richness, morphological disparity for shape and size, phylogenetic distance

289   (PD), net relatedness index (NRI) and nearest taxon index (NTI) computed for each extant

290   canid guild and the fossil one from Valdarno. In bold the guild NRI and NTI values

291   significantly different from random expectation.

Guild	# Species	MD_{shape}	MD_{size}	PD	NRI	NTI
St Biol Beni	4	0.0030	0.0526	70.2970	1.7201	1.4311
Lake Torne	3	0.0040	0.0847	87.9814	-0.4364	-0.0186
Iguazu	3	0.0029	0.0681	64.0416	1.2882	1.0657
Cerrado	3	0.0041	0.0658	62.0618	1.3452	1.2552
Daweishan	3	0.0026	0.0793	110.1744	-1.2361	-2.0509
Kanha Tiger	4	0.0022	0.0800	93.2464	0.2288	0.0271
Azraq	3	0.0028	0.0811	85.1021	-0.3341	0.2489
Dana	4	0.0045	0.0822	94.6436	0.1510	0.0158
Amboseli	5	0.0056	0.0808	110.1736	0.4570	-0.7491
Yellowstone	3	0.0030	0.0820	84.5699	-0.3280	0.2968
El Morro	4	0.0018	0.0749	100.0086	-0.7041	0.9462
San Guillermo	3	0.0020	0.0677	42.8479	**2.5570**	**2.3372**
Radom	3	0.0019	0.0779	91.6090	-0.5789	-0.3578
Valdarno	3	0.0047	0.0763	61.6456	1.4894	0.9919

292

293

294   *3.2 Phylogenetic community composition and climate*

295   Table 2 reports PDI, NRI and NTI for each canid guild. Only the Patagonian canid

296   community of San Guillermo showed a significant phylogenetic clustering due to the

297   presence of three foxes all belonging to the same genus *Lycalopex*. The canid guild from

298 Sweden (arctic ecosystem) showed the lowest NRI and NTI values but was not significantly

299 over-dispersed.

300   Spearman non-parametric rank correlation was employed to explore association

301 between univariate ecometrics, phylogenetic descriptors and climate. Averaged skull size of

302 canid communities was significantly associated with several bioclimatic variables (BIO1,

303 BIO3, BIO4, BIO7, BIO15, BIO19) while morphological shape disparity correlated with PD

304 and NTI (Table 3). Among the other parameters, also NRI exhibited a significant relationship

305 with bioclimatic variables 3, 4 and 7 (Table 3).

306   Multivariate regressions allowed the selection of 10 bioclimatic variables that were

307 significantly ($P < 0.001$) associated with averaged skull shape. These included BIO1 (Annual

308 Mean Temperature) that explained 26.34% of shape variance, BIO3 (Isothermality, 31.95%

309 var.), BIO4 (Temperature Seasonality, 40.44% var.), BIO6 (Min Temperature of Coldest

310 Month, 36.32% var.), BIO7 (Temperature Annual Range , 45.86%), BIO8 (Mean

311 Temperature of Wettest Quarter, 38.67%), BIO11 (Mean Temperature of Coldest Quarter,

312 33.65% var.), BIO12 (Annual Precipitation, 32.79% var.), BIO16 (Precipitation of Wettest

313 Quarter, Precipitation of Warmest Quarter, 22.64% var.) and BIO18 (39.53% var.). All the

314 other bioclimate variables were discarded from further analyses.
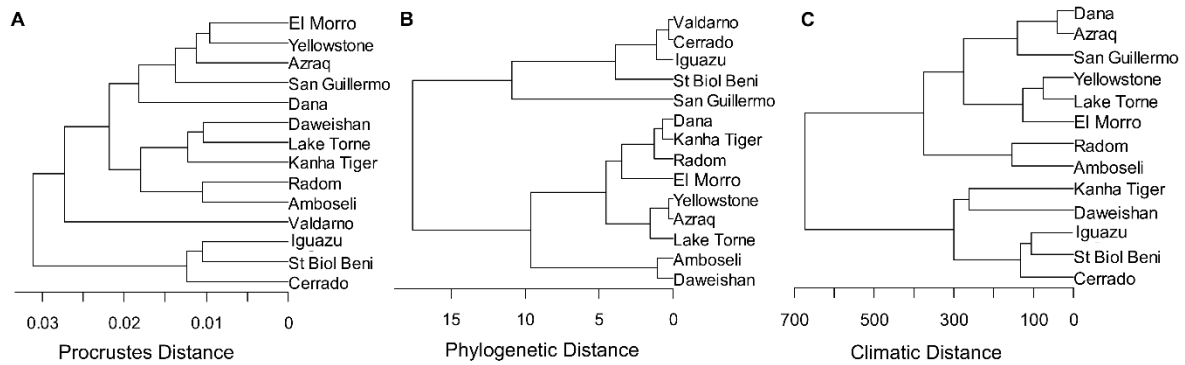
315

Table 3. Spearman correlation coefficient (below diagonal) and their respective P values (above) to test degree of univariate association between bioclimatic parameters and ecometrics of canid guilds. Only three bioclimatic indices were significantly associated with NRI. All the other associations were non-significant. BIO1 Annual Mean Temperature, BIO3 Isothermality, BIO4 Temperature Seasonality, BIO7 Temperature Annual Range, BIO15 Precipitation Seasonality, BIO19 Precipitation of Coldest Quarter. Significance is highlighted in bold.

	Ln CS	MD Shape	MD Size	PD	NRI	NTI	BIO1	BIO3	BIO4	BIO7	BIO15	BIO19
Ln CS	—	0.721	0.901	0.775	0.112	0.721	**0.027**	**0.025**	**0.024**	**0.035**	**0.049**	**0.010**
MD Shape	-0.110	—	0.803	**0.021**	0.144	**0.009**	0.482	0.831	0.817	0.802	0.748	0.364
MD Size	0.038	0.077	—	0.448	0.094	0.603	0.668	0.471	0.181	0.296	0.529	0.394
PD	0.088	**0.632**	0.231	—	**0.008**	**0.001**	0.803	0.374	0.316	0.157	0.517	0.642
NRI	-0.462	-0.429	-0.484	**-0.698**	—	**0.010**	0.344	**0.033**	**0.010**	**0.004**	0.831	0.694
NTI	-0.110	**-0.692**	-0.159	**-0.824**	0.687	—	0.775	0.255	0.364	0.425	0.494	0.517
BIO1	**-0.610**	0.214	-0.132	0.077	0.286	-0.088	—	**0.046**	**0.007**	0.077	**0.008**	0.128
BIO3	**-0.615**	0.066	-0.220	-0.269	**0.593**	0.341	**0.560**	—	**0.000**	**0.001**	0.297	0.133
BIO4	**0.621**	-0.071	0.396	0.302	**-0.687**	-0.275	**-0.703**	**-0.923**	—	**0.000**	0.162	0.128
BIO7	**0.587**	-0.077	0.314	0.416	**-0.741**	-0.242	**-0.507**	**-0.799**	0.893	—	0.522	0.343
BIO15	**-0.555**	0.099	-0.192	0.198	0.066	-0.209	**0.698**	0.313	-0.412	-0.196	—	**0.001**
BIO19	**0.681**	-0.275	0.258	-0.143	-0.121	0.198	-0.445	-0.440	0.445	0.287	**-0.808**	—

321   UPGMA trees showed distinct community associations depending on the use of

322   average skull shape, phylogenetic descriptors or climate (Fig. 4).

Fig. 4. UPGMA trees based on procrustes distance metrics (A), phylocom distance matrix (B)
and Euclidean distances obtained from 19 bioclimatic variables (C). In C the fossil Valdarno
community was not included due to the lack of climatic data.

328   Based on shape, the fossil community Valdarno is basal to a cluster that separates all canid

329   communities of Old World from those of New World (Fig. 4A). If phylocom is considered,

330   Valdarno clusters with South American communities since it includes all members belonging

331   to the same genus (Fig. 4B) while when bioclimatic variables are concerned, two clusters

332   showed differences between more seasonal (mostly Old World canid communities except

333   Chinese and Indian) and less seasonal areas (Fig. 4C).

334   Mantel test was significant only when average cranial shape distance matrix was

335   contrasted with climate distance matrix between the guilds (z =3692.987; p = 0.005). The

336   morphological distances between guilds were not associated with phylocom distances (z =

337   83.26294; p = 0.283) and the latter was equally not related to climatic distances (z=1781194;

338   p = 0.231).

339

340   *3.3 Two blocks PLS and Valdarno reconstruction*

341   Two blocks PLS returned a significant result when contrasting the averaged skull shape

342   variables against the pre-selected ten bioclimatic variables with the first pair of vectors

343   showing strong positive correlation (p-value = 0.002; r-PLS1 = 0.91; see Fig. 5).

344
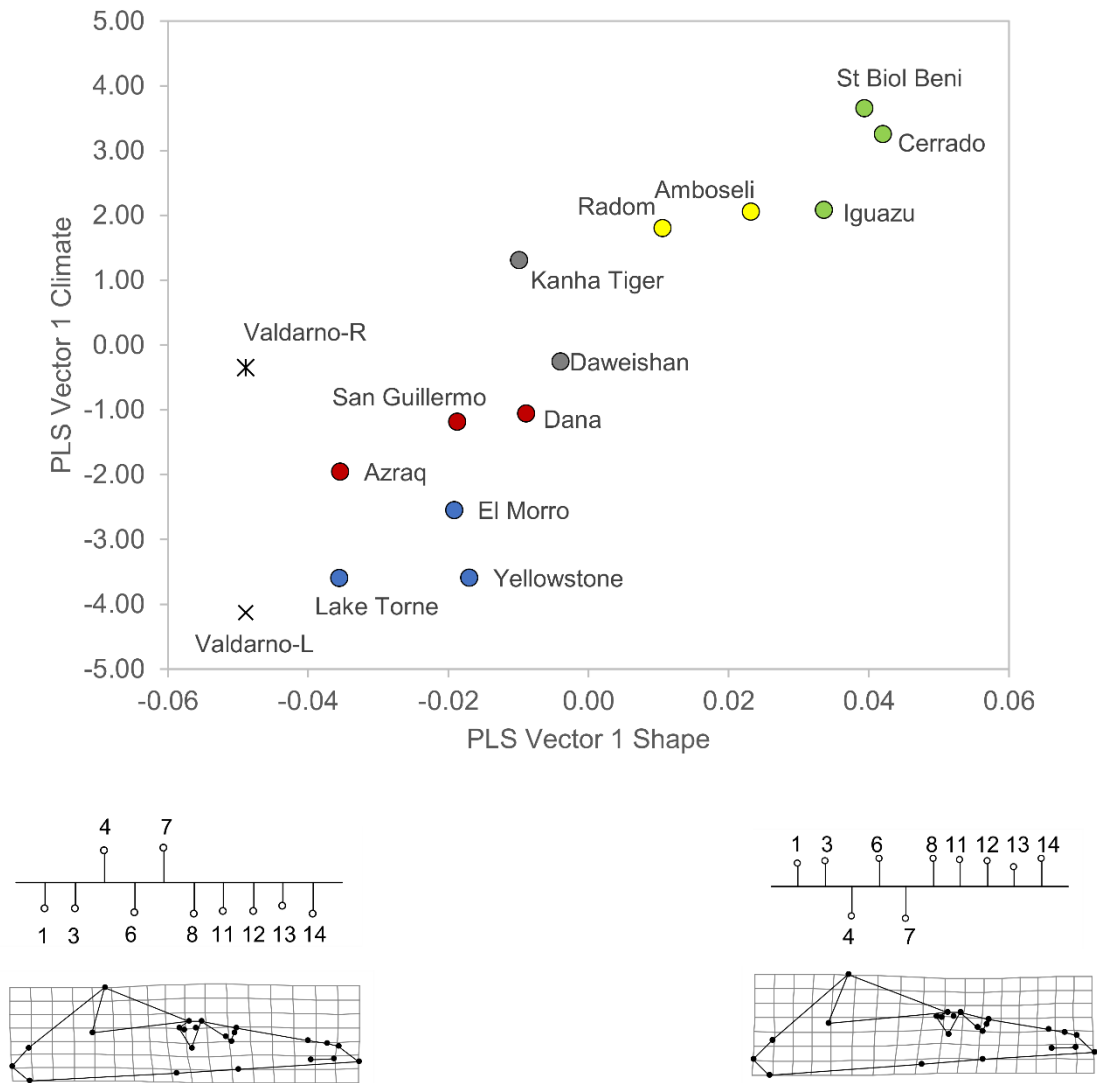
Figure 5. Scatterplot showing the first pair of Partial Least Squares vectors obtained from
testing association between averaged skull shape vs selected bioclimatic variables for each
extant guild. Deformation grids show shape changes in correspondence of the extremes of the
first PLS vector, while variable profiles give the estimate of the standardized scores of each
bioclimatic variable (including BIO1, 3, 4, 7, 8, 11, 12, 13, 14) as vertical lines extending
from a horizontal axis corresponding to the mean. Valdarno-L = projected score of fossil
guild based on linear regression; Valdarno-R=project score of fossil guild based on robust
regression.

On the negative PLS1 scores, canid communities are characterised by relatively longer

muzzle and wider teeth, traits associated with high temperature seasonality (BIO4) and

temperature annual range (BIO7) but lower temperature parameters (BIO1, 3, 6, 8 11) and

precipitation (BIO12, 13, 14). On the PLS1 positive scores, neotropical communities from

South America clusters in relation to their higher temperature and precipitation parameters.

The average shape of these communities shows a relatively shorter muzzle and wider

zygomatic arch. A projection of Valdarno into PLS morphospace based on linear regression

function (Valdarno-L, Fig. 5) clusters this community at the basis of the most seasonal extant

guild (Lake Torne). If robust regression is applied to predict PLS climatic score, the Valdarno

community show a much higher value in climate vector compatible with that of Daweishan

Reserve (China, Fig. 5).

The PLS1 climatic scores of Valdarno have a strong impact on the predictions of bioclimatic

variables depending on if linear or robust model is assumed to be correct (Table 4).

Table 4. Climatic parameters reconstructed for Valdarno canid guild based on multiple
methods (PLS = projection of Partial Least Square scores on first PLS climate vector, NRI =
net relatedness index).

	PLS-L	PLS-R	NRI
BIO1 Annual Mean Temperature	0.722	12.539	-
BIO3 Isothermality	36.575	51.823	64.369
BIO4 Temperature Seasonality	899.962	542.734	221.9
BIO6 Minimum Temp. Coldest Month	-19.941	0.4508	-
BIO7 Temperature annual range	39.662	28.965	25.301
BIO8 Mean Temperature of wettest quarter	4.592	15.213	-
BIO11 Minimum Temp. Coldest Quarter	-12.201	5.7216	-
BIO12 Annual Precipitation	473.500	1143.41	-
BIO16 Precipitation of Wettest Quarter	56.780	396.036	
BIO18 Precipitation of Warmest Quarter	130.425	91.277	-

371 Reconstructions based on lower score of linear model support Valdarno canid community to

372 be adapted to very cold and harsh conditions with average annual temperature of 0.72 degrees

373 Celsius, high temperature seasonality (BIO4) and relatively low precipitations (BIO12, 16,

374 18). The use of score obtained from robust regression model provides for Valdarno a

375 relatively higher annual temperature (12.53 degrees Celsius) although lower than those

376 experienced by extant canid communities in East Asia, and a seasonality and precipitation

377 parameter compatible (but still lower) with the Indian ecosystem of Kanha Tiger reserve.

378 NRI is also a univariate trait that allows predictions for BIO3, 4 and 7 with parameters

379 generally similar to that of PLS climatic robust score. Although the centroid size averaged by

380 community correlated with many bioclimatic parameters (Table 3), no linear model turned to

381 be significant so no predictions could be made based on this metric.

382

4. DISCUSSION

384 Palaeoenvironmental reconstruction is a challenging task that requires a multidisciplinary

385 approach. So far, mammalian community structures have been used to predict

386 palaeoenvironments (Reed, 1998; Fernández and Paláes-Campomanes, 2003; Fernández and

387 Vrba, 2006; Rodriguez et al., 2006; Louys et al., 2011, 2015; Kovarovic et al., 2018)

388 however, there is a multitude of traits and categories that investigators can employ. We

389 demonstrate that average skull shape is a reasonably good ecometric trait to characterise

390 extant canid guilds, hence this approach could also be applied to other mammalian groups to

391 improve palaeoenvironmental reconstruction.

392

393 *4.1 Palaeoecology of Valdarno canids based on skull morphology*

394   Most of extant canids are secondary consumers with a catholic diet that allowed them

395   to adapt to a multitude of environments (Sillero-Zubiri et al., 2004). Biogeographical events

396   (e.g., the canid invasion of South America) generated unique signatures in canid community

397   composition whose structural changes might reflect evolutionary history as well as

398   adaptations to local conditions (Bubadué et al., 2016). In keeping with Meloro et al. (2017),

399   the canid skull shape data exhibit a significant phylogenetic signal in both size and shape in

400   spite being indicative of dietary adaptations and bite force capabilities (Damasceno et al.,

401   2013; Meloro et al., 2015). This result did not change even if we considered subspecies as

402   separate OTUs suggesting that geographical variation, to a certain extent, could be taken into

403   account also in the analyses of guild ecometrics. Polly et al. (2017) demonstrated that the

404   American red fox varies considerably in calcaneal/gear ratio across latitudes as it might be

405   expected for mammalian body size following Bergmann's rule (Clauss et al., 2013).

406   Although this variation might not impact strongly interspecific data analyses, we noted that,

407   especially for shape, it might be relevant when genetic data support subspecies segregation

408   for taxa with a particularly broad geographical range size.

409   In our data set the grey wolf represents a good example since, based on phylomorphospace

410   occupation, the arctic subspecies (*C. lupus arctos*) separates from the rest of large canids

411   being characterised by the extreme development of hypercarnivorous traits (short muzzle,

412   wide upper carnassial, broad zygomatic arch, Fig. 3). Such genetic/phenotypic resolution was

413   not available in many other cases from our sample but the inclusion of few established

414   subtaxa appears to make a difference when interpreting phylogenetic signal in the data. This

415   trait does not hinder the identification of an adaptive signal. Indeed, our sample clearly

416   showed consistent clustering of hypercarnivorous large dogs (including the fossil *Lycaon*

417   *falconeri*, Rook, 1994) vs other more generalist taxa, further supporting the presence of a

418   pattern of convergence in the feeding apparatus driven by the adaptation to kill relatively

419   large prey (Van Valkenburgh, 2007; Slater et al., 2009).

420   Coherently with this pattern, the fossils *Canis etruscus* and *Canis arnensis* occupy positive

421   PC1 scores as the rest of modern *Canis* spp. (Fig. 3), although *C. etruscus* clusters closely

422   with the red fox based on PC2 score, while *C. arnensis* with *C. lupus*. Cherin et al. (2014)

423   already noted that in several cranial traits the Etruscan wolf is lesser similar to the extant grey

424   wolf than *Canis arnensis*, thus supporting patterns observed in the phylomorphospace.

425   A unique combination of cranial traits made *Canis etruscus* a flexible species that possibly

426   altered its ecological adaptations in relation to the presence of other canid competitors (see

427   Garcia and Virgos, 2007). The ecological flexibility of *C. etruscus* is equally reflected in its

428   relative abundance across Eurasia that is much higher when compared to *C. arnensis* whose

429   distribution remains limited to the Mediterranean area (Fig. 1B; Bartolini Lucenti and Rook,

430   2016; https://paleobiodb.org/#/).

431

432   *4.2 Variation between extant and fossil canid guilds*

433   No significant difference in averaged or variance skull morphometric traits was

434   detectable in the canid sample. This reinforces previous generalisations on the resilience of

435   carnivoran evolution to change in functional community structure through time. Such a lack

436   of difference (as previously identified by Van Valkenburgh, 1985, 1988; Meloro, 2011b)

437   does not prevent to detect subtle changes that can be linked with climatic adaptations (see

438   Belmaker, 2018; Koufos, 2014). This was achieved by using averaged phenotypic traits (as in

439   Polly et al., 2017), that are significantly associated with bioclimatic variables. The different

440   approaches we employed converged towards this conclusion. PLS appeared to be the method

441   suitable to maximise the degree of covariation between average skull shape and climatic

442   differences. The use of distance matrices equally confirmed a genuine association between

443   guild morphological distances and climatic distances. Interestingly, no strong pattern was

444   identified when phylogenetic distance metrics were used to test their association with climatic

445   data. The work of Kamilar et al. (2015) suggested that phylogenetic community metrics can

446   be indicative of environmental changes however, as for large scale clade sorting, such a result

447   applies to larger communities (e.g., the whole Carnivora) at larger spatial scale. On the one

448   hand, we opted to avoid pseudoreplications by analysing only selected/representative canid

449   guilds, although large spatial scale analyses might provide a stronger pattern of ecometrics

450   variation (Barr, 2017; Schap et al. 2021). On the other hand, we argue that large spatial

451   patterns in ecometrics requires spatial bias to be accounted for, since our phylomorphospace

452   clearly showed the impact of taxonomic distinctiveness. In summary, we believe that large

453   spatial scale approaches require more caution when intraspecific variation is concerned.

454

455   *4.3 Reconstruction of Valdarno palaeoenvironments*

456   Because Valdarno canid (and not only) community appears to have little in common

457   with modern analogues (see cluster analysis, Fig. 4A), it is particularly difficult to provide

458   accurate predictions for many environmental parameters. By reducing the number of

459   bioclimatic variables, we were able to reduce statistical noise in our data and provided

460   realistic parameters although quite distinct depending on the use of linear or robust

461   regression. If fossil data are projected within the PLS morphospace using linear approach, the

462   Valdarno canid guild predicts a harsh, highly seasonal environment characterised by very low

463   average annual temperature and low precipitation. In this respect, the Valdarno guild appears

464   to resemble guilds sampled in the Northern Hemisphere such as Lake Torne characterised by

465   extreme artic conditions. This similarity is partially due to the predominance of carnivorous

466   adaptations in the morphologies of the extant Arctic wolf (*C. lupus arctos*) that resembles *L.*

467   *falconeri* from Valdarno, while the red and the Arctic fox (*V. vulpes*, and *V. alopex*) equally

468   share positive PC2 scores of shape space as for *C. etruscus* (Fig. 3).

469   Alternatively, the robust regression provides an average annual temperature strongly

470   compatible with that already predicted by Bertini et al. (2010) based on the pollen record of

471   Poggio Rosso (12.5-14 degrees Celsius). Poggio Rosso (1.87-1.83 MA) is slightly younger

472   than the Tasso Faunal Unit (1.9 Ma, where all Valdarno canids co-occur) and register the co-

473   occurrence of *C. etruscus* and *C. arnensis* but no presence of *Lycaon falconeri*. Still, its fauna

474   is highly compatible with Tasso FU (Mazza et al., 2004) and predicted precipitation

475   parameters for interglacial phases (750-1200 mm) are within the range of that predicted by

476   our Valdarno data (Table 3, 1159 mm).

477   Other climatic projections for Tasso FU were obtained using cenograms by Montuire and

478   Marcolini (2002) that for 'il Tasso' locality suggested an average annual temperature of 17.36

479   degrees Celsius while for 'Pietrafitta' (another Early Pleistocene site with typical Valdarno

480   faunal assemblage) 11.9 degrees.

481   All these predictions converge in supporting previous inferences on the inset of a strong

482   glacial event that around 2.0 Ma altered flora and fauna of continental Western Europe. The

483   concomitant co-occurrence of the Val d'Arno canids in the Mediterranean region (Fig. 1B)

484   supports stability of this climatic condition in the area during this time, while more advanced

485   species (e.g., *C. mosbachensi*s, *L. lycaonoides*) were spreading from China to Europe towards

486   Dmanisi, a site coeval with Tasso FU (Qui et al., 2004; Jangzuo et al., 2018; Jangzuo, 2021;

487   Bartolini-Lucenti et al. 2020, 2021). Interestingly, Azzaroli (1983) proposed another faunal

488   event at 2.6 Ma characterised by the spreading of large grazing genera like *Mammuthus* and

489   *Equus*. Since the temporal distribution of the canid Euroasiatic record has been updated

490   multiple times with the occurrence of *Canis etruscus* earlier than expected by the '*Wolf*

491   *event*', it is likely that the spreading of steppe environments already favoured dispersal of

492 cursorial and potentially pack-hunter large dogs (Sotnikova and Rook, 2010; Cherin et al.,

493 2013).

494

495 **Credit author statement**

496 Carlo Meloro: Conceptualization, Data collection, Methodology, Formal analysis,

497 Investigation, Writing, Visualisation, Funding acquisition.

498 Gabriele Sansalone: Conceptualization, Methodology, Formal analysis, Writing.

499

500 **Declaration of competing interest**

501 The authors declare that they have no known competing financial interests or personal

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508

509 **Data availability**

510 Supplementary data to this article can be found in the Mendeley archive:

511 https://data.mendeley.com/datasets/42zgw9pkz2/draft?a=788baf9c-8415-40f5-b279-

512 1d2f551289ef

References

522 Adams, D.C., 2014. A generalized K statistic for estimating phylogenetic signal from shape

523 and other high-dimensional multivariate data. Syst. Biol. 63, 685-697.

524 Adams, D.C., Collyer, M.L., 2018. Phylogenetic ANOVA: Group-clade aggregation,

525 biological challenges, and a refined permutation procedure. Evolution 72, 1204-1215.

526 Adams, D.C., Otárola-Castillo, E., 2013. geomorph: an R package for the collection and

527 analysis of geometric morphometric shape data. Meth. Ecol. Evol. 4, 393-399.

528 Agustí, J., Antón, M., 2002. Mammoths, sabertooths, and hominids: 65 million years of

529 mammalian evolution in Europe. Columbia University Press.

530 Arnold, C., Matthews, L.J., Nunn, C.L., 2010. The 10kTrees Website: A New Online

531 Resource for Primate Phylogeny. Evol. Anthropol. 19, 114-118.

532 Azzaroli, A., 1983. Quaternary mammals and the "end-Villafranchian" dispersal event—a

533 turning point in the history of Eurasia. Palaeogeogr., Palaeoclimatol., Palaeoecol. 44, 117-

534 139.

535   Azzaroli, A., De Giuli, C., Ficcarelli, G., Torre, D., 1988. Late Pliocene to early mid-

536   Pleistocene mammals in Eurasia: faunal succession and dispersal events. Palaeogeogr.,

537   Palaeoclimatol., Palaeoecol. 66, 77-100.

538   Barr, W.A., 2017. Bovid locomotor functional trait distributions reflect land cover and annual

539   precipitation in sub-Saharan Africa. Evol. Ecol. Res. 18, 253-269.

540   Bartolini-Lucenti, S., Spassov, N., 2022. Cave canem! The earliest Canis

541   (Xenocyon)(Canidae, Mammalia) of Europe: Taxonomic affinities and paleoecology of the

542   fossil wild dogs. Quat. Sci. Rev. 276, 107315.

543   Bartolini Lucenti, S., Madurell-Malapeira, J., Martínez-Navarro, B., Palmqvist, P.,

544   Lordkipanidze, D., Rook, L., 2021. The early hunting dog from Dmanisi with comments on

545   the social behaviour in Canidae and hominins. Sci. Rep. 11, 1-10.

546   Bartolini Lucenti, S., Bukhsianidze, M., Martínez-Navarro, B., Lordkipanidze, D., 2020. The

547   Wolf from Dmanisi and augmented reality: review, implications, and opportunities. Front.

548   Earth Sci. 8, 131.

549   Bartolini Lucenti, S., Alba, D. M., Rook, L., Moyà-Solà, S., Madurell-Malapeira, J., 2017.

550   Latest Early Pleistocene wolf-like canids from the Iberian Peninsula. Quat. Sci. Rev. 162, 12-

551   25.

552   Bartolini Lucenti, S., Rook, L., 2016. A review on the Late Villafranchian medium-sized

553   canid Canis arnensis based on the evidence from Poggio Rosso (Tuscany, Italy). Quat. Sci.

554   Rev. 151, 58-71.

555   Belmaker, M., 2018. Insights from carnivore community composition on the paleoecology of

556   early Pleistocene Eurasian sites: implications for the dispersal of hominins out of Africa.

557   Quat. Int. 464, 3-17.

558   Bertini, A., Magi, M., Mazza, P.P., Fauquette, S., 2010. Impact of short-term climatic events

559   on latest Pliocene land settings and communities in Central Italy (Upper Valdarno basin).

560   Quat. Int. 225, 92-105.

561   Blois, J.L., Hadly, E.A., 2009. Mammalian response to Cenozoic climatic change. Annu.

562   Rev. Earth Planet. Sci. 37, 181-208.

563   Bookstein, F.L., 1989. " Size and shape": a comment on semantics. Syst. Zool. 38, 173-180.

564   Brugal, J.P., Boudadi-Maligne, M., 2011. Quaternary small to large canids in Europe:

565   taxonomic status and biochronological contribution. Quat. Int. 243, 171-182.

566   Bubadué, J., Cáceres, N., dos Santos Carvalho, R., Meloro, C., 2016. Ecogeographical

567   variation in skull shape of South-American canids: abiotic or biotic processes? Evol. Biol. 43,

568   145-159.

569   Cherin, M., Bertè, D.F., Rook, L., Sardella, R., 2014. Re-defining *Canis etruscus* (Canidae,

570   Mammalia): a new look into the evolutionary history of Early Pleistocene dogs resulting from

571   the outstanding fossil record from Pantalla (Italy). J. Mamm. Evol. 21, 95-110.

572   Cherin, M., Bertè, D.F., Sardella, R., Rook, L., 2013. *Canis etruscus* (Canidae, Mammalia)

573   and its role in the faunal assemblage from Pantalla (Perugia, central Italy): comparison with

574   the Late Villafranchian large carnivore guild of Italy. Boll. Soc. Paleontol. It. 52, 11-18.

575   Clauss, M., Dittmann, M.T., Müller, D.W., Meloro, C., Codron, D., 2013. Bergmann′s rule

576   in mammals: a cross-species interspecific pattern. Oikos 122, 1465-1472.

577   Dalerum, F., 2013. Phylogenetic and functional diversity in large carnivore assemblages.

578   Proc. R. Soc. B 280, 20130049.

579   Dalerum, F., Cameron, E.Z., Kunkel, K., Somers, M.J., 2009. Diversity and depletions in

580   continental carnivore guilds: implications for prioritizing global carnivore conservation. Biol.

581   Lett. 5, 35-38.

582   Damasceno, E.M., Hingst-Zaher, E., Astúa, D., 2013. Bite force and encephalization in the C

583   anidae (Mammalia: Carnivora). J. Zool. 290(4), 246-254.

584   Damuth, J., Janis, C.M., 2011. On the relationship between hypsodonty and feeding ecology

585   in ungulate mammals, and its utility in palaeoecology. Biol. Rev. 86, 733-758.

586   Eronen, J.T., Puolamäki, K., Liu, L., Lintulaakso, K., Damuth, J., Janis, C., Fortelius, M.,

587   2010. Precipitation and large herbivorous mammals I: estimates from present-day

588   communities. Evol. Ecol. Res. 12, 217-233.

589   Fernández, M.H., Peláez-Campomanes, P., 2003. The bioclimatic model: a method of

590   palaeoclimatic qualitative inference based on mammal associations. Glob. Ecol. Biogeogr.

591   12, 507-517.

592   Fernández, M.H., Vrba, E.S., 2006. Plio-Pleistocene climatic change in the Turkana Basin

593   (East Africa): evidence from large mammal faunas. J. Hum. Evol. 50, 595-626.

594   Figueirido, B., Martín-Serra, A., Tseng, Z.J., Janis, C.M., 2015. Habitat changes and

595   changing predatory habits in North American fossil canids. Nat. Comm. 6, 1-11.

596   Flower, L. O., Schreve, D. C., 2014. An investigation of palaeodietary variability in European

597   Pleistocene canids. Quat. Sci. Rev. 96, 188-203.

598   Fortelius, M., Eronen, J., Jernvall, J., Liu, L.P., Pushkina, D., Rinne, J., Tesakov, A.,

599   Vislobokova, I., Zhang, Z., Fortelius, M., 2002. Fossil mammals resolve regional patterns of

600   Eurasian climate change over 20 million years. Evol. Ecol. Res. 4, 1005-1016.

601 Garcia, N., Virgos, E., 2007. Evolution of community composition in several carnivore

602 palaeoguilds from the European Pleistocene: the role of interspecific competition. Lethaia,

603 40, 33-44.

604 Gebregiorgis, D., Giosan, L., Hathorne, E. C., Anand, P., Nilsson-Kerr, K., Plass, A., Lückge,

605 A., Clemens, S.C., Frank, M., 2020. What can we learn from X-ray fluorescence core

606 scanning data? A paleomonsoon case study. Geochem., Geophys., Geosyst. 21,

607 e2019GC008414.

608 Hijmans, R.J., Cameron, S.E., Parra, J.L., Jones, P.G., Jarvis, A., 2005. Very high resolution

609 interpolated climate surfaces for global land areas. Int. J. Climatol. 25, 1965-1978.

610 Jiangzuo, Q., Liu, J., Wagner, J., Dong, W., Chen, J., 2018. Taxonomical revision of fossil

611 Canis in Middle Pleistocene sites of Zhoukoudian, Beijing, China and a review of fossil

612 records of *Canis mosbachensis variabilis* in China. Quat. Int. 482: 93-108.

613 Jernvall, J., Hunter, J.P., Fortelius, M., 1996. Molar tooth diversity, disparity, and ecology in

614 Cenozoic ungulate radiations. Science 274, 1489-1492.

615 Kamilar, J.M., Beaudrot, L., Reed, K.E., 2015. Climate and species richness predict the

616 phylogenetic structure of African mammal communities. PLoS One 10, e0121808.

617 Klein, R.G., Scott, K., 1989. Glacial/interglacial size variation in fossil spotted hyenas

618 (Crocuta crocuta) from Britain. Quat. Res. 32, 88-95.

619 Klingenberg, C.P., 2016. Size, shape, and form: concepts of allometry in geometric

620 morphometrics. Dev. Genes Evol. 226, 113-137.

621 Koufos, G. D., 2018. New material and revision of the Carnivora, Mammalia from the Lower

622 Pleistocene locality Apollonia 1, Greece. Quaternary, 1: 6.

623   Koufos, G.D., 2014. The Villafranchian carnivoran guild of Greece: implications for the

624   fauna, biochronology and paleoecology. Integr. Zool. 9, 444-460.

625   Kovarovic, K., Su, D.F., Lintulaakso, K., 2018. Mammal community structure analysis. In

626   Methods in Paleoecology (pp. 351-372). Springer, Cham.

627   Lewis, M.E., 1997. Carnivoran paleoguilds of Africa: implications for hominid food

628   procurement strategies. J. Hum. Evol. 32, 257-288.

629   Louys, J., Meloro, C., Elton, S., Ditchfield, P., Bishop, L.C., 2015. Analytical framework for

630   reconstructing heterogeneous environmental variables from mammal community structure. J.

631   Hum. Evol. 78, 1-11.

632   Louys, J., Meloro, C., Elton, S., Ditchfield, P., Bishop, L.C., 2011. Mammal community

633   structure correlates with arboreal heterogeneity in faunally and geographically diverse

634   habitats: implications for community convergence. Glob. Ecol. Biogeogr. 20, 717-729.

635   Martínez-Navarro, B., 2010. Early Pleistocene faunas of Eurasia and hominin dispersals. In:

636   Fleagle J., Shea J., Grine F., Baden A., Leakey R. (Eds.) Out of Africa I. Springer, Dordrecht,

637   pp. 207-224.

638   Martínez-Navarro, B., Rook, L., 2003. Gradual evolution in the African hunting dog lineage

639   systematic implications. Compt. Rend. Palevol. 2, 695-702.

640   Masini, F., Torre, D., 1990. Large Mammal dispersal events at the beginning of the Late

641   Villafranchian. In European Neogene mammal chronology (pp. 131-138). Springer, Boston,

642   MA.

643   Mazza, P.P., Bertini, A., Magi, M., 2004. The late Pliocene site of Poggio Rosso (central

644   Italy): taphonomy and paleoenvironment. Palaios 19, 227-248.

645 Meachen, J.A., Dunn, R.H., Werdelin, L., 2016. Carnivoran postcranial adaptations and their

646 relationships to climate. Ecography 39, 553-560.

647 Meiri, S., Dayan, T., Simberloff, D., 2004. Body size of insular carnivores: little support for

648 the island rule. Am. Nat. 163, 469-479.

649 Meiri, S., Dayan, T., Simberloff, D., Grenyer, R., 2009. Life on the edge: carnivore body size

650 variation is all over the place. Proc. R. Soc. B 276, 1469-1476.

651 Meloro, C., 2011a. Locomotor adaptations in Plio-Pleistocene large carnivores from the

652 Italian Peninsula: Palaeoecological implications. Curr. Zool. 57, 269-283.

653 Meloro, C., 2011b. Feeding habits of Plio-Pleistocene large carnivores as revealed by the

654 mandibular geometry. J. Vertebr. Paleontol. 31, 428-446.

655 Meloro, C., 2011c. Morphological disparity in Plio-Pleistocene large carnivore guilds from

656 Italian peninsula. Acta Palaeontol. Pol. 56, 33-44.

657 Meloro, C., de Oliveira, A.M., 2019. Elbow joint geometry in bears (Ursidae, Carnivora): a

658 tool to infer paleobiology and functional adaptations of Quaternary fossils. J. Mamm. Evol.

659 26, 133-146.

660 Meloro, C., Louys, J., 2015. Ecomorphology of radii in Canidae: Application to fragmentary

661 fossils from Plio-Pleistocene hominin assemblages. Acta Palaeontol. Pol.60, 795-806.

662 Meloro, C., Raia, P., 2010. Cats and dogs down the tree: the tempo and mode of evolution in

663 the lower carnassial of fossil and living Carnivora. Evol. Biol. 37, 177-186.

664 Meloro, C., Tamagnini, D., 2021. Macroevolutionary ecomorphology of the Carnivora skull:

665 adaptations and constraints in the extant species. Zool. J. Linn. Soc.

666 https://doi.org/10.1093/zoolinnean/zlab075

667   Meloro, C., Elton, S., Louys, J., Bishop, L.C., Ditchfield, P., 2013. Cats in the forest:

668   predicting habitat adaptations from humerus morphometry in extant and fossil Felidae

669   (Carnivora). Paleobiology 39, 323-344.

670   Meloro, C., Hudson, A., Rook, L., 2015. Feeding habits of extant and fossil canids as

671   determined by their skull geometry. J. Zool. 295, 178-188.

672   Meloro, C., Hunter, J., Tomsett, L., Portela Miguez, R., Prevosti, F.J., Brown, R.P., 2017.

673   Evolutionary ecomorphology of the Falkland Islands wolf Dusicyon australis. Mamm. Rev.

674   47, 159-163.

675   Mihlbachler, M.C., Rivals, F., Solounias, N., Semprebon, G.M., 2011. Dietary change and

676   evolution of horses in North America. Science 331, 1178-1181.

677   Montuire, S., Marcolini, F., 2002. Palaeoenvironmental significance of the mammalian

678   faunas of Italy since the Pliocene. J. Quat. Sci. 17, 87-96.

679   Napoleone, G., Albianelli, A., Azzaroli, A., Mazzini, M., 2001. The Poggio Rosso locality

680   calibrated to the end-Pliocene and its significance for dating the late Villafranchian mammal

681   faunas of the Upper Valdarno, Central Italy. Riv. Ital. Paleontol. Stratigr. 107, 287-296.

682   Oksanen, J., Blanchet, F.G., Kindt, R., Legendre, P., Minchin, P.R., O'Hara, R.B., Simpson,

683   G.L., Solymos, P., Stevens, M.H.H., Szoecs, E., Wagner, H., 2013. Package 'vegan'.

684   Community ecology package, version, 2(9), 1-295.

685   Palmqvist, P., Arribas, A., Martínez-Navarro, B., 1999. Ecomorphological study of large

686   canids from the lower Pleistocene of southeastern Spain. Lethaia 32, 75-88.

687   Polly, P.D., 2010. Tiptoeing through the trophics: geographic variation in carnivoran

688   locomotor ecomorphology in relation to environment. In: Goswami A. (Ed.) Carnivoran

689   evolution: new views on phylogeny, form, and function, 374-401.

690 Polly, P.D., 2020. Ecometrics and Neogene faunal turnover: the roles of cats and hindlimb

691 morphology in the assembly of carnivoran communities in the New World. Geodiversitas 42,

692 257-304.

693 Polly, P.D., Eronen, J.T., Fred, M., Dietl, G.P., Mosbrugger, V., Scheidegger, C., Frank,

694 D.C., Damuth, J., Stenseth, N.C., Fortelius, M. (2011). History matters: ecometrics and

695 integrative climate change biology. Proc. R. Soc. B 278, 1131-1140.

696 Polly, P.D., Fuentes-Gonzalez, J., Lawing, A.M., Bormet, A.K., Dundas, R.G., 2017. Clade

697 sorting has a greater effect than local adaptation on ecometric patterns in Carnivora. Evol.

698 Ecol. Res. 18, 61-95.

699 Raia, P., 2010. Phylogenetic community assembly over time in Eurasian Plio-Pleistocene

700 mammals. Palaios, 25, 327-338.

701 Raia, P., Carotenuto, F., Passaro, F., Piras, P., Fulgione, D., Werdelin, L., Saarinen, J.,

702 Fortelius, M. 2013. Rapid action in the Palaeogene, the relationship between phenotypic and

703 taxonomic diversification in Cenozoic mammals. Proc. R. Soc. B 280, 20122244.

704 Reed, K.E., 1998. Using large mammal communities to examine ecological and taxonomic

705 structure and predict vegetation in extant and extinct assemblages. Paleobiology 24, 384-408.

706 Rodríguez, J., Hortal, J., Nieto, M., 2006. An evaluation of the influence of environment and

707 biogeography on community structure: the case of Holarctic mammals. J. Biogeogr. 33(2),

708 291-303.

709 Rohlf, F.J., 2015. The tps series of software. Hystrix, It. J. Mammal. 26, 9-12.

710 Rohlf, F.J., Corti, M., 2000. Use of two-block partial least-squares to study covariation in

711 shape. Syst. Biol. 49, 740-753.

712   Rohlf, F.J., Slice, D., 1990. Extensions of the Procrustes method for the optimal

713   superimposition of landmarks. Syst. Biol. 39, 40-59.

714   Rook, L., Martínez-Navarro, B., 2010. Villafranchian: the long story of a Plio-Pleistocene

715   European large mammal biochronologic unit. Quat. Int. 219, 134-144.

716   Rook, L., Torre, D., 1996. The wolf-event in western Europe and the beginning of the Late

717   Villafranchian. Neues Jahrb. für Geol. Paläontol. Mh. 1996, 495-501.

718   Rook, L., 1994. The Plio-Pleistocene Old World *Canis* (*Xenocyon*) ex gr. *falconeri*. Boll.

719   Soc. Paleontol. Ital. 33, 71-82.

720   Sakamoto, M., Ruta, M., 2012. Convergence and divergence in the evolution of cat skulls:

721   temporal and spatial patterns of morphological diversity. PLoS One 7, e39752.

722   Schap, J.A., Samuels, J.X., Joyner, T.A., 2021. Ecometric estimation of present and past

723   climate of North America using crown heights of rodents and lagomorphs. Palaeogeogr.,

724   Palaeoclimatol., Palaeoecol. 562, 110144.

725   Sidlauskas, B., 2008. Continuous and arrested morphological diversification in sister clades

726   of characiform fishes: a phylomorphospace approach. Evolution 62, 3135-3156.

727   Sillero-Zubiri, C., Hoffmann, M., Macdonald, D.W., 2004. Canids: foxes, wolves, jackals,

728   and dogs: status survey and conservation action plan. Gland, Switzerland: IUCN.

729   Simberloff, D., Dayan, T., 1991. The guild concept and the structure of ecological

730   communities. Ann. Rev. Ecol. Syst. 22(1), 115-143.

731   Slater, G.J., Friscia, A.R., 2019. Hierarchy in adaptive radiation: a case study using the

732   Carnivora (Mammalia). Evolution 73(3), 524-539.

733 Slater, G.J., Dumont, E.R., Van Valkenburgh, B., 2009. Implications of predatory

734 specialization for cranial form and function in canids. J. Zool. 278, 181-188.

735 Sotnikova, M., Rook, L. 2010. Dispersal of the Canini (Mammalia, Canidae: Caninae) across

736 Eurasia during the late Miocene to early Pleistocene. Quat. Int. 212, 86-97.

737 Tedford, R. H., Wang, X., & Taylor, B. E. (2009). Phylogenetic systematics of the North

738 American fossil caninae (Carnivora: Canidae). Bull. Am. Mus. Nat. Hist. 2009(325), 1-218.

739 Tomiya, S., Meachen, J.A,. 2018. Postcranial diversity and recent ecomorphic

740 impoverishment of North American gray wolves. Biol. Lett. 14, 20170613.

741 Turner, A., 1990. The evolution of the guild of larger terrestrial carnivores during the Plio-

742 Pleistocene in Africa. Geobios 23, 349-368.

743 Van Valkenburgh, B., 1985. Locomotor diversity within past and present guilds of large

744 predatory mammals. Paleobiology 11, 406-428.

745 Van Valkenburgh, B., 1988. Trophic diversity in past and present guilds of large predatory

746 mammals. Paleobiology 14, 155-173.

747 Van Valkenburgh, B., 2007. Déjà vu: the evolution of feeding morphologies in the Carnivora.

748 Integr. Comp. Biol. 47, 147-163.

749 Webb, C.O., Ackerly, D.D., McPeek, M.A., Donoghue, M.J., 2002. Phylogenies and

750 community ecology. Ann. rev. Ecol. Syst. 33, 475-505.

751 Wesley-Hunt, G.D., 2005. The morphological diversification of carnivores in North America.

752 Paleobiology 31, 35-55.

753   Zurano, J.P., Martinez, P.A., Canto-Hernandez, J., Montoya-Burgos, J.I., Costa, G.C., 2017.

754   Morphological and ecological divergence in South American canids. J. Biogeogr. 44, 821-

755   833.

756