

A comprehensive study of stable carbon and oxygen isotopes for *Cathaica pulveratrix* and *Metodontia yantaiensis* land snails over last two glacial cycles at Beiyao site, central China: implications for paleovegetation and climate seasonality

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November 25, 2022

Abstract

Modern investigations have shown that oxygen and carbon isotopes of land snail shells are useful indicators of climate and vegetation in monsoonal region. However, stable isotope study on snail fossil shells in strata has been seldom done, and the reliability of those indicators needs further verification. Moreover, intra-shell stable isotope analysis of individual snail is rather scarce, and seasonal variation of the glacial-interglacial monsoonal climate remains unclear. In this context, we performed $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ analyses on fossil shells of cold-aridiphilous *Cathaica pulveratrix* and sub-humidiphilous *Metodontia yantaiensis* from the loess section over the last two glacial cycles at Beiyao site in southern Chinese Loess Plateau. The $\delta^{18}\text{O}$ of fossil shells reflected monsoonal rainfall amount and more rainfall during MIS3 and MIS7. Meanwhile, the $\delta^{13}\text{C}$ of fossil shells indicated relative abundance of C3/C4 plants and more C4 biomass during MIS3 and MIS7. The $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ of the two species from the same horizon are significantly different, reflecting differences in their growing season and/or physiological habits. Intra-shell variations of stable isotopes showed that climatic seasonality was relatively strong during the glacial periods whereas seasonality became weakened during the interglacials. Our findings provide an environmental background for explaining past human activities at the Beiyao site. The investigation of stone artifacts showed that ancient human activities were relatively strong during MIS3 and MIS7. During these stages, the warm and humid climate with smaller seasonal contrast was favorable for the regional expansion of human activities.

1 **A comprehensive study of stable carbon and oxygen isotopes for *Cathaica***
2 ***pulveratrix* and *Metodontia yantaiensis* land snails over last two glacial**
3 **cycles at Beiyao site, central China: implications for paleovegetation and**
4 **climate seasonality**

5

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21 **Key Points:**

- 22 • $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ of *C. pulveratrix* and *M. yantaiensis* snails over last 20 ka were studied
23 to trace paleoclimate and paleovegetation changes.
- 24 • $\delta^{18}\text{O}$ showed more rainfall during MIS3 and MIS7 stages and an overall 1.5 times
25 stronger seasonality during glacial than interglacial period.
- 26 • $\delta^{13}\text{C}$ revealed more C_4 biomass during warm/humid MIS3 and MIS7 stages and *M.*
27 *yantaiensis* ingested more C_4 than *C. pulveratrix*.

28

29 **Abstract**

30 Modern investigations have shown that oxygen and carbon isotopes of land snail shells are
31 useful indicators of climate and vegetation in monsoonal region. However, stable isotope
32 study on snail fossil shells in strata has been seldom done, and the reliability of those
33 indicators needs further verification. Moreover, intra-shell stable isotope analysis of
34 individual snail is rather scarce, and seasonal variation of the glacial-interglacial monsoonal
35 climate remains unclear. In this context, we performed $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ analyses on fossil
36 shells of cold-aridophilous *Cathaica pulveratrix* and sub-humidophilous *Metodontia*
37 *yantaiensis* from the loess section over the last two glacial cycles at Beiyao site in southern
38 Chinese Loess Plateau. The $\delta^{18}\text{O}$ of fossil shells reflected monsoonal rainfall amount and
39 more rainfall during MIS3 and MIS7. Meanwhile, the $\delta^{13}\text{C}$ of fossil shells indicated relative
40 abundance of C_3/C_4 plants and more C_4 biomass during MIS3 and MIS7. The $\delta^{18}\text{O}$ and
41 $\delta^{13}\text{C}$ of the two species from the same horizon are significantly different, reflecting
42 differences in their growing season and/or physiological habits. Intra-shell variations of stable
43 isotopes showed that climatic seasonality was relatively strong during the glacial periods

44 whereas seasonality became weakened during the interglacials. Our findings provide an
45 environmental background for explaining past human activities at the Beiyao site. The
46 investigation of stone artifacts showed that ancient human activities were relatively strong
47 during MIS3 and MIS7. During these stages, the warm and humid climate with smaller
48 seasonal contrast was favorable for the regional expansion of human activities.

49

50 **1 Introduction**

51 Land snails are ideal materials for paleoclimate studies(Goodfriend,1992; Wang et al.,
52 2016; Wu et al., 2018). This is because they have advantages of being widely distributed,
53 abundant and well preserved in strata. And they are relatively sensitive to climate changes. To
54 date, researches on land snails include inferring the environmental conditions under which
55 land snails survived through identifying faunal assemblage and living habit of each species
56 (Gittenberger and Goodfriend., 1993; Wu et al., 2008), and reconstructing the paleoclimates
57 through analyzing stable isotopes of land snail shells (Goodfriend and Ellis, 2002; Liu et al.,
58 2006; Gu et al., 2009; Colonese et al., 2010; Rangarajan et al., 2013; Yanes and Fernández-
59 Lopez-de-Pablo, 2016; Prendergast et al., 2016; Padgett et al., 2019).

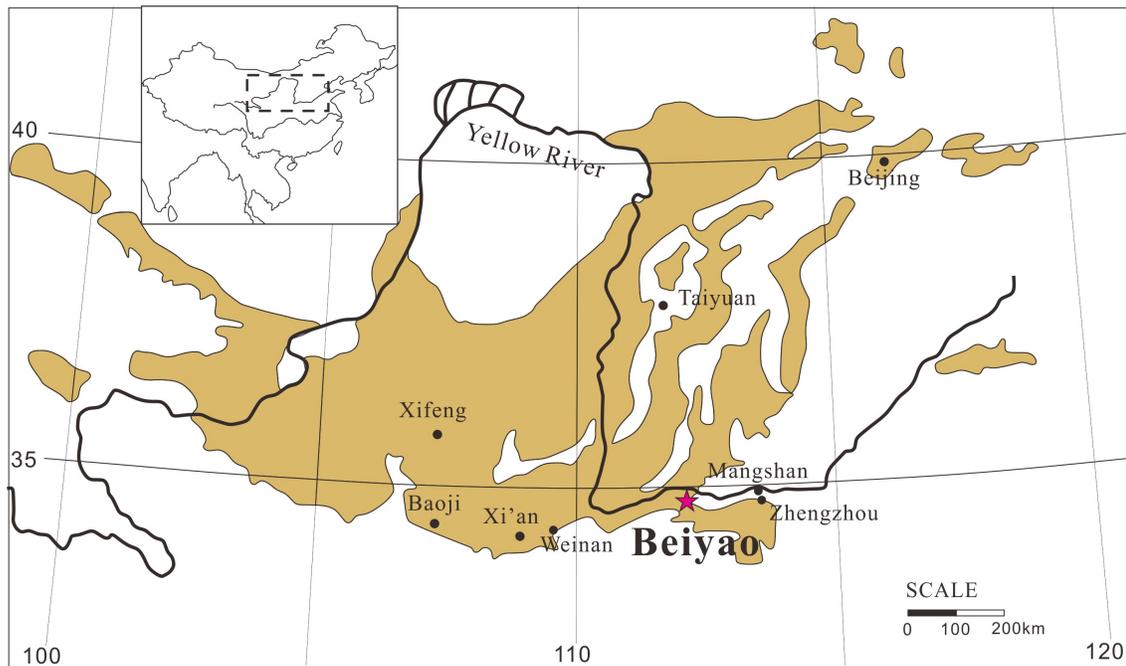
60 Theoretically, oxygen isotope in snail shell is determined by the oxygen isotope of snail
61 body water and the temperature under which shell carbonate precipitates. Although body
62 water oxygen isotopes of land snails are modified to different extents by evaporation due to
63 differences in physiological habits of various species, it can still be generally used to track
64 changes in precipitation oxygen isotopes (Zarrur et al., 2011; Zhai et al., 2019). Therefore, in
65 the case of little temperature change, oxygen isotopes of land snail shells mainly reflect
66 oxygen isotopes of rainfall (Prendergast et al., 2016; Wang et al., 2016; Milano et al., 2018;

67 Padgett et al., 2019). The snail shell carbon isotope reflects the carbon isotope composition of
68 food they intake, with a large proportion of dietary plants, e.g., organic food accounts for
69 more than 70% of carbon sources of land snail shell (Xu et al., 2010). In brief, the shell
70 carbon isotope value can provide information on the relative abundance of C₃/C₄ plants in the
71 food(Goodfriend and Ellis, 2002; Prendergast et al., 2017).

72 Land snail fossils are abundant and widely distributed in the Asian monsoon region,
73 especially in the Chinese Loess Plateau (Wu et al., 1996 ; Wu et al., 2002; Liu et al., 2006;
74 Gu et al., 2009). However, researches on the stable isotopes of snail shells have mainly
75 focused on studying modern land snails in different climatic regions (Liu et al., 2006; Wang
76 et al., 2016; Bao et al., 2018, 2019; Wang et al., 2019; Zhai et al., 2019). In contrast, stable
77 isotope analyses of fossil snails in strata have been inadequently done, and only a few species
78 of land snails were studied (Gu et al., 2009; Huang et al., 2012). In this context, it is
79 necessary to perform stable isotope analyses on shell fossils of different land snail species
80 from strata in the different regions and compare those data with other paleoclimatic proxy
81 indicators to confirm their paleoenvironment and paleoclimate significances. Moreover,
82 stable isotope analysis of individual shell along shell ontogeny has the potential to provide
83 seasonal information (Leng et al., 1998; Goodfriend and Ellis, 2002). However, the
84 application of this type of research in paleoclimate is also less developed.

85 In this study, we systematically collected land snail fossils from loess-paleosol section over
86 last two glacial-interglacial cycles at the Beiyao site in Luoyang, central China. Carbon and
87 oxygen isotopes were measured on *Cathaica pulveratrix* (cold-aridiphilous) and *Methodontia*
88 *yantaiensis* (sub-humidiphilous) land snails. We then compared these isotopic data with
89 paleoclimate proxy indicators like grain size and magnetic susceptibility with attempt to

90 reconstruct changes in climate and vegetation (C_3/C_4 plants) in the study area. The Luoyang
91 Beiyao site is an archaeological site with human activities in the Paleolithic Age. Recent
92 studies have found some lithics in strata belonging to the late glacial period and the middle
93 and late MIS7 stage (Du et al., 2011; Du and Liu, 2014), indicating that there were human
94 activities during those time periods. However, the climate and environmental context
95 associated with the human activities is still unclear. This study will precisely analyze the
96 environmental conditions for the human activities during the late glacial period and the
97 middle and late of MIS7. At the same time, we also selected snail fossils during the typical
98 periods of the glacials and interglacials, and analyzed intra-shell isotopic variation of each
99 shell to obtain seasonal information during these periods, thereby helping us to understand
100 changes in climatic seasonality from glacial to interglacial period.



101
102 **Figure 1.** Location map of the study site (red star). The yellow shaded area is the distribution
103 range of the Loess Plateau, edited from Kukla and An (1989).

104

105 2 Geological settings and sample collection

106 2.1 Geological settings

107 The loess-paleosol section is located at the Luoyang Beiyao archaeological site
108 (34°42'24"N , 112°28'46"E) on the southeast edge of the Chinese Loess Plateau (Figure
109 1). The Beiyao site lies on the third-grade loess accumulation terrace on the south bank of the
110 Luo River in Luoyang. The terrace is about 20m higher than the modern river bed, and the
111 loess section is 16.7m long from bottom to top. Grain size and magnetic susceptibility data
112 combined with optical luminescence (OSL) and AMS ¹⁴C datings showed that the loess
113 section has covered the last two glacial-interglacial cycles (Du et al., 2011). At present, the
114 mean annual temperature and annual precipitation are 14.2°C and 546 mm, respectively. The
115 study area is located in a typical monsoonal region. Northerly wind prevails and climate is
116 cold and dry in winter, while southerly wind dominates in summer with hot and rainy
117 condition. A large number of stone artifacts were found in the Beiyao section at depth of
118 6.5~7.5m and 11~13m, indicating that there were prehistoric human activities.

119 The magnetic susceptibility and median particle size curves showed synchronous changes,
120 and had a good correspondence with the marine oxygen-isotope stage (MIS) curve (Tang et
121 al., 2017). Therefore, in this study, we sub-divided the loess section to various oxygen isotope
122 stages according to the grain size and magnetic susceptibility, referring to AMS ¹⁴C and OSL
123 datings.

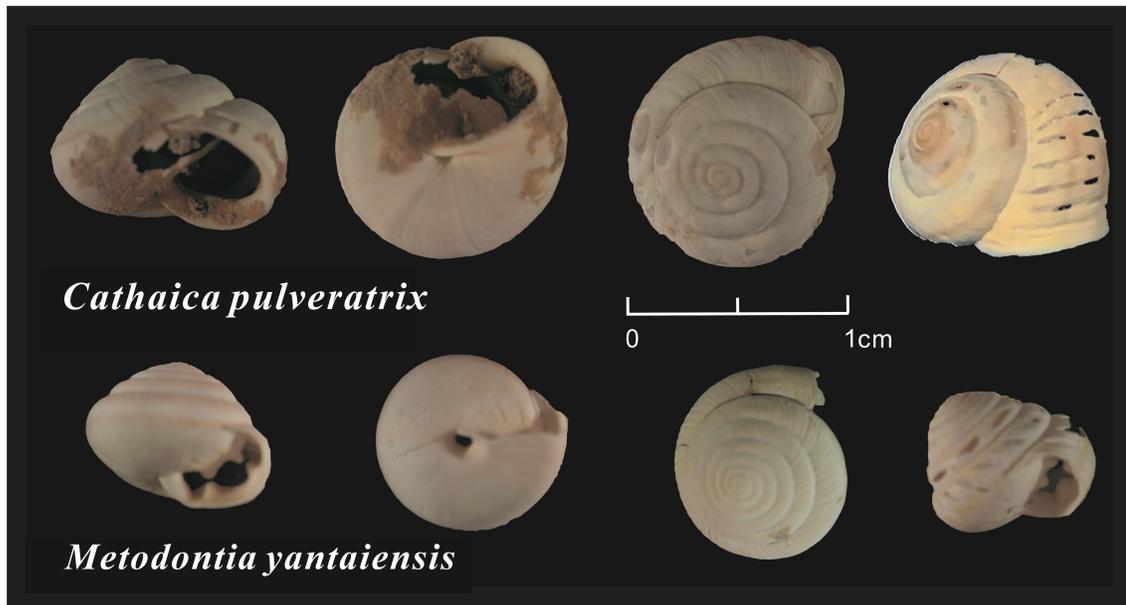
124 2.2 Collection of land snail fossils

125 During the sampling process at the Beiyao site, 1m × 1m × 10cm volume loess (or
126 paleosol) was continuously excavated downward, and the snails in each horizon were

127 collected by screening and washing using water and a 0.5 mm sieve. The identification and
128 statistics of the snail fossils used in this study were completed by Yan Wu. Throughout the
129 section, there were 1911 cold-aridiphilous *Cathaica pulveratrix* (*C. pulveratrix*) and 241 sub-
130 humidiphilous *Metodontia yantaiensis* (*M. yantaiensis*) (Wu, 2011). When the fossil
131 fragments were counted as Quaternary loess snail individuals, the calculation method
132 developed by Puisségur (1976) was used to convert the fragments into snail fossil individuals
133 and sum them as the total number of individuals. The conversion formula (Puisségur, 1976) is
134 as followed:

$$\text{Number of individuals} = \text{number of fragments}/5 - \text{number of fragments}/5 \times \text{conversion factor}$$

137 The conversion factor varies with the number of snail fossil fragments. When the number
138 of snail fossil fragments is <50, 50-75, 75-100, and >100, the conversion factor is 10%, 20%,
139 33%, and 50%, respectively. Except for few fossils due to the strong pedogenesis at 6.8-7m,
140 most of the section is rich in snail fossils. In this study, we used complete shell of land snails
141 for stable isotope analysis. Totally, there are 577 *C. pulveratrix* shells from 59 horizons, and
142 97 *M. yantaiensis* shells from 15 horizons.



143

144 **Figure 2.** Photos showing shell morphology of the two species land snails. The sampling
 145 strategy along with the growth band was also shown.

146

147 2.3 Ecological habits of the two species

148 The two species of land snails used in this study have different living habits. *C. pulveratrix*
 149 usually lives in relatively cold and dry climates whereas *M. yantaiensis* lives in warm and
 150 sub-humid climates (Wu et al., 1996; Chen and Wu, 2008). The pictures for two species of
 151 land snails are shown in Figure 2. Both species are also living in the modern time. According
 152 to Chen (2016), *C. pulveratrix* distributes over a vast area including Shanxi, Henan, Hunan,
 153 Shaanxi, Gansu, Xinjiang provinces, and even in central Asia. The habitat for *C. pulveratrix*
 154 is usually in thick grasses or under the litter beneath trees in mountain area, on flat slope of
 155 hills as well as in ranches, orchards and crop land. *M. yantaiensis* distributes usually in
 156 northern China, i.e., Beijing, Tianjin, Hebei, Shanxi, Inner Mongolia, Shandong and Shaanxi,
 157 and also shows in area around the Yangtze River. It often lives in slightly damp bushes,
 158 grasses, under rocks and leaves in in mountainous and hilly areas.

159 Shell size comparison shows that *C. pulveratrix* is usually larger than *M. yantaiensis* (Table
 160 1). This morphology difference complies with their living environment conditions. According
 161 to previous studies, the large shell can reduce the ratio of surface area to volume, thereby
 162 limiting water evaporation and making it easier for the snails to survive in drier
 163 environments (Nevo et al., 1983; Yanes and Fernández-Lopez-de-Pablo, 2016) .

164

165 **Table 1** Snail shell sizes of the two species at various MIS stages.

	MIS3		MIS4		MIS6		MIS7	
Genus	M. yantaiensis	C. pulveratrix						
Spiral (number)	5	5	5	5	7	5	6	5
Height (cm)	0.55	1.5	0.5	1.2	0.95	1.3	0.8	1.1
Height of lip (cm)	0.3	0.85	0.3	0.7	0.4	0.7	0.4	0.7
Width of lip (cm)	0.35	0.7	0.35	0.6	0.5	0.7	0.5	0.8

166

167 3 Materials and Methods

168 3.1 Snail shells pretreatment and sampling strategy

169 The entire shell was firstly cleaned with distilled water, and the soil particles attached to
 170 the shell surface were brushed using a toothbrush, and then the shell was placed in a drying
 171 oven and heated at 60 °C for 12 hours. The relatively large shells were chosen for sampling
 172 along the growth band. Firstly, we removed the residual clay cements on the surface of shells

173 using a dental drill, then cleaned the shells using a ultrasonic utility for multiple times, and
174 finally dried the shells in an oven. The three dimension of each shell (i.e., shell height, width
175 and height of shell mouth) was measured using a ruler. For intra-shell sampling, we use a
176 micro drill to take powders from the shell lip till apex at 1-2 mm interval along the growth
177 direction of the snail (Figure 2). The drill bit was soaked in diluted hydrochloric acid solution
178 after each sample to remove residual carbonate powder on it.

179 For the carbon and oxygen isotope analyses of the whole shell, about 10 shells were
180 combined according to the availability of snail shells in each horizon. This can ensure the
181 measured data to represent a general and average environment condition under which land
182 snails lived. After the shell was cleaned and dried for the first time, it was broken into
183 fragments. The clay cement attached to each shell fragment was physically removed, and
184 then the fragments were further cleaned using an ultrasonic utility. After very clean shell
185 fragments were obtained, we dried them in an oven at 60 °C. Finally, we ground them into
186 powders and homogenized using a mortar and pestle.

187 3.2 Stable isotope analyses

188 The carbon and oxygen isotopic analyses of the snail shell powder were performed on the
189 GasBench II multifunctional gas preparation system coupled with the Delta V Plus isotope
190 ratio mass spectrometer (Thermo Fisher). A 100µg carbonate powder reacted with 100%
191 H₃PO₄ at 72 °C for 1 hour. The generated CO₂ passed through two NAFION™ water traps
192 to remove trace water and passed through a PoraPlot Q chromatography column at 45 °C to
193 separate with other impurities. After that, the CO₂ was introduced into the isotope ratio mass
194 spectrometer to measure the carbon and oxygen isotope ratios. Both carbon and oxygen
195 isotope data are reported relative to the VPDB. The standards used for data correction and
196 calibration were GBW4416 ($\delta^{13}\text{C}_{\text{VPDB}}=1.61\text{‰}$, $\delta^{18}\text{O}_{\text{VPDB}}=-11.59\text{‰}$) and NBS19

197 ($\delta^{13}\text{C}_{\text{VPDB}}=1.95\text{‰}$, $\delta^{18}\text{O}_{\text{VPDB}}=-2.20\text{‰}$). The analytical precision of carbon and oxygen
198 isotopes is 0.06‰ and 0.10‰, respectively. Detailed analytical method can be found in Wang
199 et al. (2019).

200 **4 Results**

201 4.1 Carbon and oxygen isotopes of whole shell for two species land snails

202 The variation range of $\delta^{18}\text{O}_{\text{VPDB}}$ for cold-aridiphilous *C. pulveratrix* was -2.16‰ to -
203 8.13‰, and the average value was -5.03‰. The maximum value of $\delta^{18}\text{O}_{\text{VPDB}}$ was at the depth
204 of 1.1 m in the profile, which corresponds to MIS2, while the minimum value of $\delta^{18}\text{O}_{\text{VPDB}}$
205 was at the depth of 11.7m, which belongs to MIS7. The $\delta^{18}\text{O}_{\text{VPDB}}$ value for sub-
206 humidiphilous *M. yantaiensis* ranged from -7.34‰ to -9.71‰, with an average of -8.43‰.
207 The maximum $\delta^{18}\text{O}_{\text{VPDB}}$ value was at 4.6 m (MIS4) and the minimum at 11.6 m (MIS7).

208 The $\delta^{13}\text{C}_{\text{VPDB}}$ for *C. pulveratrix* ranged from -3.17‰ to -6.62‰ with an average of -
209 4.81‰. The maximum $\delta^{13}\text{C}$ was at the depth of 10.1 m in the profile, which belongs to MIS6
210 whereas the minimum $\delta^{13}\text{C}$ was at 6.6m, which corresponds to the MIS5. The range of
211 $\delta^{13}\text{C}_{\text{VPDB}}$ for *M. yantaiensis* was between -3.05‰ and -5.03‰, and the average value was -
212 3.95‰. The maximum $\delta^{13}\text{C}_{\text{VPDB}}$ for *M. yantaiensis* showed at 3.4 m (MIS3) whereas the
213 minimum $\delta^{13}\text{C}_{\text{VPDB}}$ occurred at 12 m (MIS7).

214 4.2 Carbon and oxygen isotope changes along the growth band of individual shell

215 In the MIS3 and MIS5, intra-shell $\delta^{18}\text{O}_{\text{VPDB}}$ variation for *C. pulveratrix* was from -12.3‰
216 to 0.2‰, and the variation of $\delta^{13}\text{C}_{\text{VPDB}}$ was between -6.9‰ and -4.9‰. In contrast, of the
217 intra-shell variation of $\delta^{18}\text{O}_{\text{VPDB}}$ for *M. yantaiensis* was relatively small, i.e., from -10.1‰ to
218 -5.9‰. The intra-shell $\delta^{13}\text{C}_{\text{VPDB}}$ ranged from -7.7‰ to -4.8‰ (Table 1).

219 During the MIS4 and MIS6 stages, the intra-shell $\delta^{18}\text{O}_{\text{VPDB}}$ and $\delta^{13}\text{C}_{\text{VPDB}}$ for *C. pulveratrix*

220 varied from -12.0‰ to -3.5‰ and from -7.8‰ to -2.6‰, respectively. The corresponding
 221 intra-shell variations for *M. yantaiensis* were much larger, i.e., from -13.3‰ to -1.7‰ for
 222 $\delta^{18}\text{O}_{\text{VPDB}}$ and from -11.7 to -0.6‰ for $\delta^{13}\text{C}_{\text{VPDB}}$ (Table 1).

223 The cold-aridiphilous *C. pulveratrix* had a shell height of 1.1 to 1.5 cm, a shell lip height of
 224 0.7 to 0.85 cm, and a shell lip width of 0.6 to 0.8 cm. In contrast, the sub-humidiphilous *M.*
 225 *yantaiensis* had shell height ranging from 0.55 to 0.95 cm, shell lip height ranging from 0.3 to
 226 0.4 cm, and shell lip width ranging from 0.35 to 0.5 cm (Table 2). Obviously, the shell of *C.*
 227 *pulveratrix* was significantly larger than that of *M. yantaiensis*. As a result, the intra-shell
 228 sampling number for *C. pulveratrix* was larger than that for *M. yantaiensis*.

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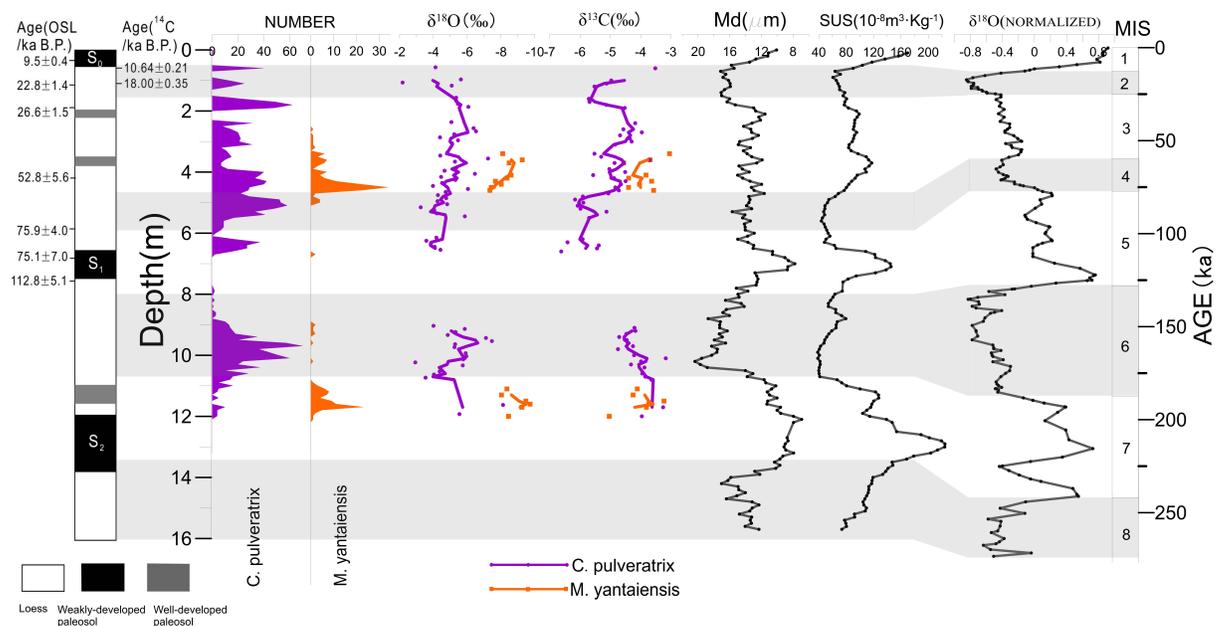
230 **Table 2** Statistics for Intra-shell $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ variations of two species at various MIS
 231 stages.

Species	Depth (m)	MIS	Number of sampling	$\delta^{18}\text{O}\%$ Average (VPDB)	$\delta^{18}\text{O}$ ‰ Max	$\delta^{18}\text{O}\%$ Min	$\delta^{13}\text{C}\%$ Average (VPDB)	$\delta^{13}\text{C}\%$ Max	$\delta^{13}\text{C}\%$ Min
<i>C. pulveratrix</i>	3.4	3	37	-9.5	-6.4	-12.3	-5.6	-4.9	-6.5
<i>C. pulveratrix</i>	4.9	4	44	-4.7	0.3	-9.7	-4.7	-2.6	-6.5
<i>C. pulveratrix</i>	9.0	6	45	-3.6	3.5	-12.0	-5.2	-4.1	-7.8
<i>C. pulveratrix</i>	11.8	7	28	-5.7	-0.2	-10.9	-6.2	-5.1	-6.9
<i>M. yantaiensis</i>	3.4	3	12	-9.8	-8.7	-11.6	-6.4	-5.1	-7.7
<i>M. yantaiensis</i>	4.9	4	12	-10.5	-7.8	-12.7	-1.6	-0.6	-3.5
<i>M. yantaiensis</i>	9.0	6	30	-5.7	-1.7	-13.3	-10.4	-8.7	-11.7
<i>M. yantaiensis</i>	11.8	7	31	-10.1	-5.9	-13.8	-6.2	-4.8	-7.4

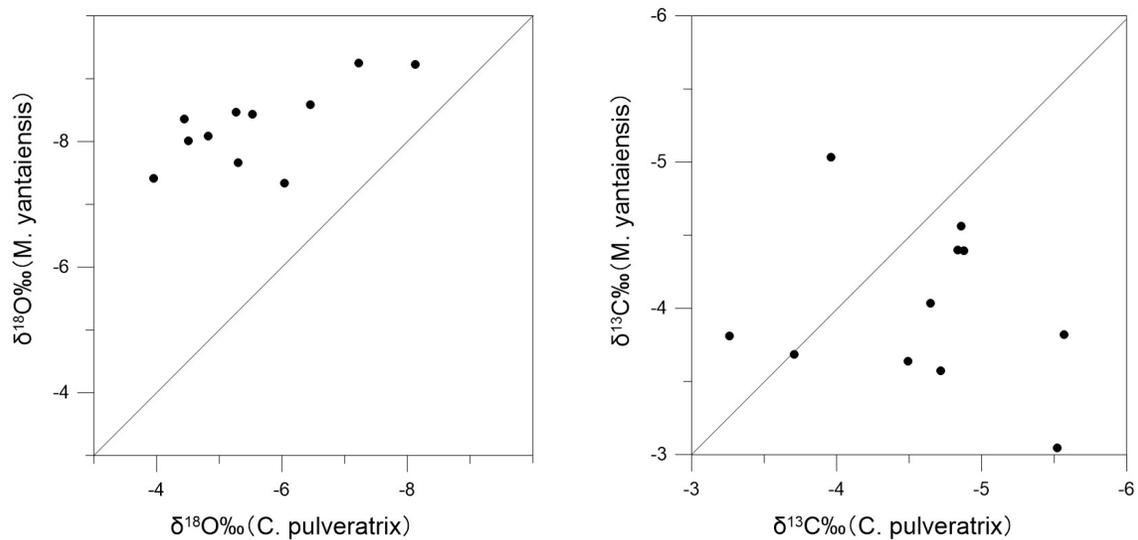
232

233 4.3 Statistics of *C. pulveratrix* and *M. yantaiensis* in loess-paleosol strata

234 In the Beiyao section, the maximum number of cold-aridiphilous snails *C. pulveratrix* was
 235 70, occurring at the depth of 9.7 m (belonging to MIS6). In contrast, the maximum number of
 236 sub-humidiphilous snails *M. yantaiensis* was 34, appearing at the depth of 4.5 m (belonging
 237 to MIS3) (Figure 3). At the bottom of the interglacial paleosol S1, very few of land snail
 238 fossils were left because of the influence of strong pedogenesis. However, the other horizons
 239 in the section were rich in snail fossils. Therefore, without considering this factor, the cold-
 240 aridiphilous species *C. pulveratrix* had a certain number distributing from MIS2 to MIS7,
 241 with two most abundant horizons (with fossil number of 58 and 70) respectively in MIS4 and
 242 MIS6. The sub-humidiphilous species *M. yantaiensis* were mainly found in MIS3 and MIS7,
 243 with maximum number reaching up to 34 and 23, respectively. Moreover, when the number
 244 of *M. yantaiensis* increased in some horizon, the number of *C. pulveratrix* in the same
 245 horizon or neighbouring horizons significantly reduced. Conversely, when the number of *C.*
 246 *pulveratrix* reached the peak of the stage, the number of *M. yantaiensis* approached the
 247 minimum or 0.



249 **Figure 3.** Changes in carbon and oxygen isotopes of *C. pulveratrix* and *M. yantaiensis* snails
250 over the last two glacial-interglacial cycles, in comparison with median grain size (Md),
251 magnetic susceptibility (SUS) and deep-sea $\delta^{18}\text{O}$ curve. Stages partition, age data and $\delta^{18}\text{O}$
252 value(standardized) of MIS were from Martinson (1987), Md data were from Tang et al.
253 (2017), SUS data were from Du and Liu (2014), ^{14}C and OSL age data were from Du et al.
254 (2011).



255
256 **Figure 4.** Comparison of carbon and oxygen isotopes between *C. pulveratrix* and *M.*
257 *yantaiensis* from the same horizon. Note that the $\delta^{18}\text{O}$ value of *M. yantaiensis* was
258 significantly lower than that of *C. pulveratrix*, while the $\delta^{13}\text{C}$ value of *M. yantaiensis*
259 was mostly higher than that of *C. pulveratrix*.

260

261 **5 Discussion**

262 5.1 Oxygen isotopes in land snail shells and changes in summer monsoon rainfall

263 Many studies have shown that oxygen isotope in land snail shell carbonate is positively
264 related to oxygen isotope in atmospheric precipitation. (Gu et al., 2009; Prendergast et al.,

265 2016; Wang et al., 2016; Milano et al., 2018; Padgett et al., 2019; Wang et al., 2019; Zhai et
266 al., 2019). Generally speaking, the $\delta^{18}\text{O}$ values of *C. pulveratrix* were more positive than
267 those of *M. yantaiensis* (Figure 4a). This is consistent with the eco-physiological habits of the
268 two land snail species. The *M. yantaiensis* snails like to live in a relatively warm and humid
269 environment and in seasons with more abundant rainfall. Due to the rainfall effect, the
270 summer rainfall $\delta^{18}\text{O}$ will be more negative, so $\delta^{18}\text{O}$ in shell carbonate of *M. yantaiensis* is
271 also relatively low. In contrast, the active season of *C. pulveratrix* is relatively cool and dry
272 with less rainfall (such as spring and autumn), so relatively more positive oxygen isotope of
273 rainfall during this time can result in relatively high $\delta^{18}\text{O}$ in shell carbonate of *C. pulveratrix*.

274 Snail shell $\delta^{18}\text{O}$ can be combined with other paleoclimate indicators such as the median
275 grain size (Md), magnetic susceptibility (SUS) of the loess and faunal assemblages of land
276 snails to indicate the strength of the East Asian summer monsoon (Wu et al., 2018). A
277 previous study has shown that the shell $\delta^{18}\text{O}$ of *C. pulveratrix* can be used as an indicator of
278 summer precipitation to reflect the strength of the summer monsoon (Gu et al., 2009).
279 Specifically, the shell $\delta^{18}\text{O}$ of *C. pulveratrix* in the monsoon region of China decreased when
280 the summer precipitation increased. This is consistent with the $\delta^{18}\text{O}$ record of stalagmites in
281 Hulu cave in Southern China (Wang et al., 2008).

282 Generally, the shell $\delta^{18}\text{O}$ of *C. pulveratrix* showed a negative correlation with SUS and a
283 positive correlation with Md in the Beiyao loess-paleosol section (Figure 3). This is
284 consistent with the results of Gu et al. (2009). In the middle part of MIS7, the $\delta^{18}\text{O}$ of *C.*
285 *pulveratrix* exhibited a negative shift, with the minimum value being -8.13‰. Meanwhile,
286 the Md value decreased, the number of cold-aridiphilous species *C. pulveratrix* decreased,
287 and the number of sub-humidiphilous species *M. yantaiensis* increased (Figure 3). It
288 suggested that the East Asian summer monsoon intensified during this period, and the $\delta^{18}\text{O}$ of

289 precipitation became more negative due to large amount of precipitation.

290 At the beginning of MIS6, the $\delta^{18}\text{O}$ value of *C. pulveratrix* experienced a positive shift,
291 while the SUS value also became lower, indicating that the climate tended to be drier.
292 Subsequently, the $\delta^{18}\text{O}$ of *C. pulveratrix* showed a change to more negative value, with the
293 most negative value reaching -7.5‰ , and the Md value also became lower, indicating that
294 there have been a significant increase in rainfall amount during the middle part of MIS6.

295 At the end of MIS5 and during MIS4, the $\delta^{18}\text{O}$ values of *C. pulveratrix* snails were
296 generally more positive, with an average $\delta^{18}\text{O}_{\text{VPDB}}$ value of -4.2‰ . At the same time, the SUS
297 increased and the Md decreased. Collectively, it indicated a relative cold and dry climatic
298 condition.

299 From MIS4 to MIS3, the $\delta^{18}\text{O}$ of *C. pulveratrix* snail showed a significant decrease,
300 indicating that the climate has entered a humid and rainy mode. However, the oxygen isotope
301 became more positive during middle MIS3, which corresponded to the decrease in SUS. This
302 implied that the climate during MIS3 was variable and there was once a relatively cold and
303 dry climate. Despite this, the $\delta^{18}\text{O}$ of *C. pulveratrix* during the middle MIS3 was still more
304 negative than that during MIS4, indicating a slightly drying middle MIS3. The $\delta^{18}\text{O}$ values of
305 *C. pulveratrix* during the late stage of MIS3 were -0.6‰ by average more negative than those
306 during the early stage of MIS3, suggested a generally more humid climate during the late
307 MIS3. But we acknowledged that the $\delta^{18}\text{O}$ during the early MIS3 was highly variable and
308 some negative extrema that are even lower than the late MIS3 $\delta^{18}\text{O}$ also appeared during this
309 period. This may reflect some transient stages with much humid condition also occurred
310 during the early MIS3. The three-stage sub-division of MIS3 can be also envisaged on the
311 SUS curve of our loess section (Figure 3). The average $\delta^{18}\text{O}$ value of *C. pulveratrix* was $-$
312 5.3‰ during MIS3 stage. In contrast, the average $\delta^{18}\text{O}$ during MIS2 was much higher ($-$

313 4.2‰) and it showed a clear trend of increase, suggestive of a climatic transition from
314 wetness to dryness.

315 Within MIS2 stage, the $\delta^{18}\text{O}$ values of *C. pulveratrix* increased up to -2‰ at about 21.6 ka,
316 which marked extreme dryness during the last glacial period (LGM). Similarly, the $\delta^{18}\text{O}$ of *C.*
317 *pulveratrix* from Mangshan loess section in central China also showed an extremely positive
318 value (approximately -1‰) around 22 ka (Gu et al. 2009). The two study sites are about 100
319 km away. Collectively, it manifested a synchronous regional drought in central China during
320 the LGM.

321 The $\delta^{18}\text{O}$ values of *M. yantaiensis* exhibited almost the same pattern of variation as those
322 of *C. pulveratrix* did. During late MIS7 stage, the $\delta^{18}\text{O}$ of *M. yantaiensis* was more negative
323 than that of *C. pulveratrix* and attained to the most negative of -9.71‰ when the $\delta^{18}\text{O}$ of *C.*
324 *pulveratrix* dropped to its most negative one (Figure 3). In the meantime, SUS also increased
325 its peak value. These lines of evidences corroborated abundant rainfall brought by the
326 intensified summer monsoon during the late MIS7. During the early MIS3, the $\delta^{18}\text{O}$ of *M.*
327 *yantaiensis* showed a gradually decreasing trend, which was synchronous with the changes in
328 *C. pulveratrix* $\delta^{18}\text{O}$ and SUS. This further confirmed climate shifted to more humid condition
329 from MIS4 to early MIS3.

330 5.2 Carbon isotopes in land snail shells and vegetation changes

331 The carbon isotope of land snail shell is mainly related to carbon isotopes of dietary plants
332 (Goodfriend and Ellis, 2002; Stott, 2002; Metref et al., 2003; Balakrishnan and Yapp, 2004).
333 A previous study on modern land snails in China has shown that snail shell carbonate was
334 enriched in ^{13}C by 14.2‰ relative to snail body that has on isotopic difference from organic
335 diet (Liu et al., 2006). At the same time, C_3 and C_4 plants have far different carbon isotope
336 compositions, i.e., the average $\delta^{13}\text{C}$ of C_3 plant is $-27.1 \pm 2.0\text{‰}$ whereas the average $\delta^{13}\text{C}$ of

337 C₄ plant is $-13.1 \pm 1.2\%$ (Farquhar et al., 1989; O'Leary, 1998; Cerling, 1999). Therefore, the
338 proportion of C₃ to C₄ plants in snail food can be estimated based on the shell-diet carbon
339 isotope fractionation and snail shell carbon isotope. Because there is a 1.3‰ decrease in the
340 $\delta^{13}\text{C}$ of atmospheric CO₂ since the industrial revolution due to the combustion of ¹³C-
341 depleted fossil fuels, so-called Suess effect (Marino et al., 1992), the above two $\delta^{13}\text{C}$ end-
342 members for C₃ and C₄ plants should be adjusted to -25.8‰ and -11.8‰, respectively, during
343 the last two glacial-interglacial periods in our study.

344 The maximum $\delta^{13}\text{C}$ of *C. pulveratrix* was -7.34‰ that occurred at MIS5. Considering
345 shell-diet carbon isotope fractionation of +14.2‰, the converted dietary $\delta^{13}\text{C}$ was -21.5‰
346 and the inferred proportion of C₄ plant was about 31%. The minimum $\delta^{13}\text{C}$ of *C. pulveratrix*
347 was -9.71‰ that showed at MIS7. The estimated relative C₄ abundance was about 14%. In
348 contrast, the most positive $\delta^{13}\text{C}$ of *M. yantaiensis* was -3.05‰ that occurred at MIS3,
349 corresponding to a relative C₄ abundance of 61%. The most negative $\delta^{13}\text{C}$ of *M. yantaiensis*
350 was -5.03‰ that showed at MIS7, converting to 47% of C₄ in the food. It can be seen that *M.*
351 *yantaiensis* snails consumed more C₄ plants than *C. pulveratrix*. We acknowledged that the
352 proportion of C₄ plants in snail's food was overestimated because land snails may also take in
353 a small portion of soil carbonates that have more positive $\delta^{13}\text{C}$ than C₃ and C₄ plants.
354 However, this does not influence our assessing the relative changes in C₄ abundances over
355 different MIS stages.

356 To some extent, relative abundance of C₄ plants can reflect the climate and seasonal
357 changes. At seasonal level, C₄ plants prefer to grow in the summer when there are more
358 warmth and abundant precipitation whereas C₃ plants grow in spring and autumn with
359 relatively low temperature (Sage et al., 1999; Huang et al., 2012). At glacial/interglacial time-
360 scale, C₄ biomass tended to increase during warm/humid interglacial periods whereas C₃

361 biomass dominated during the cold/dry glacial periods (Liu et al., 2005; Yang et al., 2015). As
362 shown in Figure 4, the $\delta^{13}\text{C}$ of *C. pulveratrix* was mostly more negative than that of *M.*
363 *yantaiensis* at the same horizon. This may indicate that *C. pulveratrix* was more active in
364 relatively cold/arid environments or seasons and accordingly ingested more C_3 plants. This is
365 consistent with the phenomenon observed by Huang et al. (2012).

366 In general, the $\delta^{13}\text{C}$ curve of *C. pulveratrix* has a positive correlation with the SUS curve
367 and a negative correlation with the $\delta^{18}\text{O}$ of *C. pulveratrix*. This indicates a linkage of C_3/C_4
368 abundance in dietary food of land snails to climate changes. Specifically, the $\delta^{13}\text{C}$ values of
369 *C. pulveratrix* snail shell during late MIS7 were slightly more positive than those during
370 MIS6, and the $\delta^{13}\text{C}$ of *C. pulveratrix* during MIS3 was more positive than MIS2 and MIS4 as
371 well (Figure 3). Because the feeding habits of the same snail would not largely change, the
372 above variation in C_4 abundance in the snail's food may reflect the changes of C_4 biomass in
373 natural vegetation along with climate, i.e., relative abundance of C_4 plants increased during
374 the warm/humid interglacial (or interstadial) periods. This is in accordance to the
375 aforementioned conclusion reached by previous studies (Liu et al., 2005; Yang et al., 2015).

376 5.3 The relationship between snail numbers of two species and environment change

377 During late MIS7, the number of cold-aridiphilous *C. pulveratrix* snail was relatively
378 lower than that of sub-humidiphilous *M. yantaiensis* and the land snail *M. yantaiensis* had
379 reached a peak amount. At this time, Md became finer, SUS value increased, and the shell
380 $\delta^{18}\text{O}$ values of both *C. pulveratrix* and *M. yantaiensis* shifted to more negative. These
381 multiple proxies uniformly suggested that the warm and humid climate prevailed, which was
382 suitable to the growth of sub-humidiphilous *M. yantaiensis*. In addition, a large number of
383 stone artifacts were found at the depth of 11-13 m (MIS7) in the Beiyao section (Du and Liu,
384 2014), indicating strong human activities. The inferred warm/humid climatic condition was

385 conducive to the intensified prehistoric human activities.

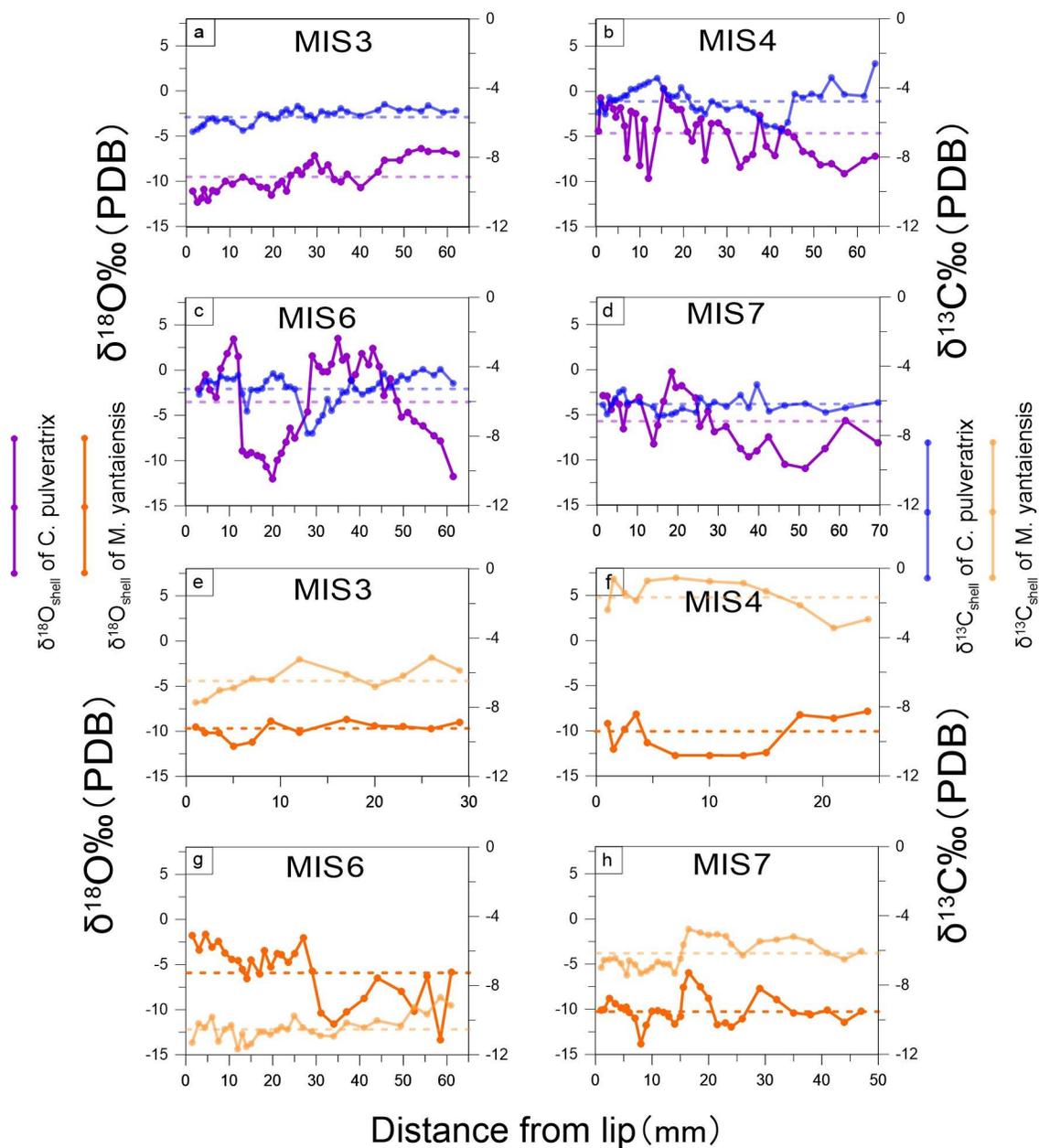
386 After entering MIS6, the number of cold-aridiphilous species increased and reached the
387 peak of the whole profile at 9.7 m whereas the sub-humidiphilous species almost
388 disappeared, which implied the climate became much colder and drier than the previous
389 stage. In the meantime, the $\delta^{18}\text{O}$ of *C. pulveratrix* shifted to more positive value, i.e., up to -
390 5.3‰, reflecting less monsoonal rainfall as well.

391 During most MIS5, land snail fossils were not preserved due to the influence of strong
392 pedogenesis and there were only a few sub-humidiphilous snails at the depth of 6.5-7 m. At
393 the end of MIS5, a small number of cold-aridiphilous species began to appear, indicating that
394 the climate started to be relatively cold and dry, in accordance to the Md and SUS records.

395 To MIS4 stage, the number of cold-aridiphilous species significantly increased, reaching a
396 maximum of 58, while sub-humidiphilous species rarely existed and even disappeared. The
397 cold/dry climate as seen from the $\delta^{18}\text{O}$ of *C. pulveratrix*, Md and SUS accounted for the
398 flourish of the cold-aridiphilous *C. pulveratrix*.

399 During MIS3, the numbers of *C. pulveratrix* and *M. yantaiensis* showed alternative
400 increases, further testifying variable climatic conditions. It also indicated that the climate was
401 of moderate conditions so that both cold-aridiphilous and sub-humidiphilous species co-
402 existed. At the early MIS3 stage, the number of *C. pulveratrix* decreased when *M. yantaiensis*
403 reached its peak abundance. In contrast, both the numbers of *C. pulveratrix* and *M.*
404 *yantaiensis* largely reduced at the middle MIS3. To the late MIS3, *M. yantaiensis* went further
405 reduced but the number of *C. pulveratrix* increased. This assemblage change indicated that
406 the climate was warmer and more humid at the early MIS3 than at late MIS3. A faunal
407 assemblage study of land snails in central Chinese Loess Plateau also suggested that the
408 temperature and humidity were higher during the early MIS3 (Chen and Wu, 2008).

409 However, the $\delta^{18}\text{O}$ of *C. pulveratrix* was highly variable during the early MIS3 and was not
 410 as more negative as that during the late MIS3 (Figure 3). This reflected a variable summer
 411 monsoon and an overall less rainfall during the early MIS3.



412

413 **Figure 5.** Intra-shell variations of $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ for the two species at various MIS stages.

414

415 5.4 Intra-shell variation of stable isotopes and climate seasonality

416 In this study, intra-shell stable isotope analyses were performed on both *C. pulveratrix* and
417 *M. yantaiensis* snails at MIS3, MIS4, MIS6, and MIS7, respectively. The measured *C.*
418 *pulveratrix* and *M. yantaiensis* snails were chosen from the same layer (10 cm) in each MIS
419 stage. During MIS3, the $\delta^{18}\text{O}$ of *C. pulveratrix* and *M. yantaiensis* were among the most
420 negative values of the four MIS stages, with averaged $\delta^{18}\text{O}$ of -9.5‰ and -9.8‰,
421 respectively. Moreover, the intra-shell variations in $\delta^{18}\text{O}$ of the two snails were relatively
422 small. For example, the $\delta^{18}\text{O}$ of *C. pulveratrix* showed a variation magnitude of 5.9‰
423 whereas $\delta^{18}\text{O}$ of *M. yantaiensis* only changed by 2.9‰ (Figure 5a, e). This suggested a weak
424 seasonality during the warm/humid MIS3 stage. Padgett et al. (2019) also observed a steady
425 trend of $\delta^{18}\text{O}$ in land snail shell in warm and humid climate. In contrast, the magnitudes of
426 intra-shell $\delta^{18}\text{O}$ variations for *C. pulveratrix* and *M. yantaiensis* showed large increases, i.e.,
427 up to 10‰ and 4.9‰, respectively.

428 During MIS6, the average $\delta^{18}\text{O}$ values of *C. pulveratrix* and *M. yantaiensis* became more
429 positive and were around -3.6‰ and -5.7‰, respectively (Figure 5c, g). Meanwhile, the
430 intra-shell $\delta^{18}\text{O}$ of the two species exhibited largest variations during MIS6, i.e., a magnitude
431 of 15.5‰ for *C. pulveratrix* and a magnitude of 12.1‰ for *M. yantaiensis*. These magnitudes
432 were respectively 2.6 and 4 times of those for the same species during MIS3. It revealed
433 extreme seasonal contrast during the cold/dry MIS6. It is worthy of mentioning that the intra-
434 shell $\delta^{18}\text{O}$ curve of *C. pulveratrix* displayed regular seasonal changes during MIS6 (Figure
435 5c). Judging from the sinusoidal cycles, the *C. pulveratrix* snail may have a life span of about
436 two years. The snail possibly started to grow from the summer of the first year to the autumn
437 of the second year. The highest $\delta^{18}\text{O}$ values recorded in the shell growing in the spring and

438 autumn seasons attained to ca +2‰ and the lowest $\delta^{18}\text{O}$ recorded in the shell segments in
439 summer was about -12‰. The large seasonal contrast was unlikely only attributed to
440 temperature changes, which would be 56 °C offset if calculating by the carbonate oxygen
441 isotope-temperature coefficient of 1‰ per 4 °C. Obviously, seasonal changes of rainfall
442 largely contributed to the above fluctuation of $\delta^{18}\text{O}$ of *C. pulveratrix*, that is, the negative
443 values in shell $\delta^{18}\text{O}$ being caused by rainfall amount effect in summer. An intra-shell $\delta^{18}\text{O}$
444 study for the land snail collected from Ethiopia also revealed significant contribution of
445 rainfall to the shape and amplitude of shell $\delta^{18}\text{O}$ cycles (Leng et al., 1998). Except for the
446 shell lip part, the $\delta^{13}\text{C}$ of *C. pulveratrix* showed an overall opposite relationship with the shell
447 $\delta^{18}\text{O}$ (Figure 5c). When the $\delta^{18}\text{O}$ was more negative in summer, the $\delta^{13}\text{C}$ became more
448 positive, implying the snail consumed increased amount of C_4 plants in this season. In spring
449 and autumn (at 30-45 mm from shell lip), more C_3 plants were ingested by the snail. This
450 seasonal change of C_3/C_4 proportion in snail's food diet is consistent with the seasonal
451 distribution of C_3 and C_4 plants in natural vegetation (Sage et al., 1999).

452 During MIS7, two individual shells for intra-shell isotope study were taken from the depth
453 of 11.8 m, which happened to be within the period of strong prehistoric human activities (Du
454 and Liu, 2014). Based on the previous discussions on $\delta^{18}\text{O}$ of *C. pulveratrix* and *M.*
455 *yantaiensis*, the climate was generally warm and humid during this time. The intra-shell $\delta^{18}\text{O}$
456 variations for *C. pulveratrix* and *M. yantaiensis* were at amplitudes of 10.7‰ and 10.9‰,
457 respectively. The variations were smaller than those during MIS6. This overall small seasonal
458 contrast was conducive to regional spread of human activity.

459 In summary, the average amplitude of intra-shell $\delta^{18}\text{O}$ variations for *C. pulveratrix* was
460 about 8.4‰ during the interglacial periods (i.e., MIS3 and MIS7), whereas it was 12.75‰
461 during the glacial periods (i.e., MIS4 and MIS6). In the same manor, the intra-shell $\delta^{18}\text{O}$ of

462 *M. yantaiensis* varied by 10.8‰ and 16.5‰, respectively, during the interglacial and glacial
463 periods. Regardless of which species, the changing amplitude was 1.5 times larger during the
464 glacial periods. Therefore, if the intra-shell variation of $\delta^{18}\text{O}$ can be used to quantify the
465 seasonal changes, the climatic seasonality during glacial periods would be about 1.5 times
466 stronger than that during interglacial periods.

467 To explore the stable isotope differences among individual shells of each snail species
468 from the same sampling horizon (10 cm layer), we analyzed $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ on *C. pulveratrix*
469 from 7 layers and *M. yantaiensis* from 3 layers. The carbon and oxygen isotope data were
470 shown in Table 3. Firstly, within the same MIS (i.e., MIS3 or MIS7), the $\delta^{18}\text{O}$ of sub-
471 humidiphilous species (*M. yantaiensis*) showed little change, whereas the $\delta^{18}\text{O}$ of cold-
472 aridiphilous species (*C. pulveratrix*) distributed much discretely. This may indicate that sub-
473 humidiphilous species have a more strict requirement on climate conditions, i.e., only grow
474 during the period of abundant rainfall, while cold-aridiphilous species had strong adaptability
475 and can survive under large range of climate conditions. Secondly, for the cold-aridiphilous
476 species, the shell $\delta^{18}\text{O}$ changes during the even-numbered MIS (i.e., MIS2, MIS4, and MIS6)
477 were larger than those during the odd-numbered MIS (i.e., MIS3 and MIS7). Since the snail
478 shells collected each sampling layer may not strictly come from the same time year, the
479 above phenomenon may indicate that the climates within the time-span of each sampling
480 layer during glacial periods (even-numbered MIS) were very unstable, whereas the climates
481 during interglacial periods (odd-numbered MIS) had relatively stable and uniform conditions
482 within the time period of each sampling layer. Previous studies have shown that climate
483 during the last glacial period was quite unstable, with climate oscillations at centennial to
484 millennium scales (Ren et al., 1996; Ding et al., 1998). This is in accordance to the large
485 intra-species variation of shell $\delta^{18}\text{O}$ in each sampling layer.

486

487 **Table 3** Statistics for intra-species $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ variations of two species at various MIS
 488 stages.

Species	Depth (m)	MI S	Shell numbe r	$\delta^{18}\text{O}$ S.D.	$\delta^{18}\text{O}$ Max (VPD B)	$\delta^{18}\text{O}$ Min (VPDB)	$\delta^{13}\text{C}$ S.D.	$\delta^{13}\text{C}$ Max (VPDB)	$\delta^{13}\text{C}$ Min (VPDB)
<i>C. pulveratrix</i>	0.60	2	10	2.85	1.34	-7.65	0.99	-1.09	-4.54
<i>C. pulveratrix</i>	1.80	3	10	2.47	-2.51	-10.02	0.74	-3.68	-5.83
<i>C. pulveratrix</i>	3.60	3	3	1.84	-5.93	-9.33	1.52	-2.22	-5.27
<i>C. pulveratrix</i>	4.6	3	10	2.11	-2.69	-9.13	1.42	-2.62	-7.06
<i>C. pulveratrix</i>	6.60	4	10	2.56	1.10	-6.71	0.83	-4.96	-7.63
<i>C. pulveratrix</i>	9.10	6	10	1.98	-1.77	-7.18	1.27	-1.22	-5.88
<i>C. pulveratrix</i>	11.70	7	7	2.02	-5.32	-10.55	2.34	0.20	-6.29
<i>M. yantaiensis</i>	3.60	3	5	1.26	-8.08	-10.90	1.65	-1.93	-6.31
<i>M. yantaiensis</i>	4.60	3	10	1.18	-5.60	-8.92	2.20	1.18	-5.78
<i>M. yantaiensis</i>	11.70	7	10	1.13	-7.53	-11.38	0.62	-2.70	-4.80

489

490 **6 Conclusion**

491 In this study, we systematically analyzed stable carbon and oxygen isotopes on cold-
 492 aridiphilous *C. pulveratrix* and sub-humidiphilous *M. yantaiensis* snail shell fossils from the
 493 Beiyao loess-paleosol section in southeastern Chinese Loess Plateau. Stable isotopes were
 494 measured on both the mixed multiple shells and the single shell along the growth band. The
 495 obtained $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ data were compared with Md and SUS from the same profile and
 496 deep-ocean $\delta^{18}\text{O}$ curve to verify the reliability of snail shell stable isotopes for paleoclimate
 497 reconstruction. We reached the following conclusions:

498 1. $\delta^{18}\text{O}$ of snail shells in strata can be used to indicate the intensity of summer monsoon
499 rainfall. During MIS7 and MIS3 stages, the shell $\delta^{18}\text{O}$ was more negative, indicating strong
500 monsoonal rainfall, which showed a good correlation to Md, SUS, and deep-sea $\delta^{18}\text{O}$ curve.
501 Meanwhile, the shell $\delta^{13}\text{C}$ can reflect the proportion of C_4 plants in snail's food and
502 ultimately trace the relative abundance of C_4 plants in contemporary vegetation. The results
503 showed that the relative abundance of C_4 plants increased during the warm/humid MIS7 and
504 MIS3.

505 2. The stable isotopes of *C. pulveratrix* and *M. yantaiensis* from the same horizon were
506 largely different, reflecting differences in their eco-physiological habits. The $\delta^{18}\text{O}$ of *M.*
507 *yantaiensis* was significantly lower than that of *C. pulveratrix*, indicating that *M. yantaiensis*
508 lived in warmer and more humid conditions than *C. pulveratrix*. The $\delta^{13}\text{C}$ of *M. yantaiensis*
509 was mostly higher than that of *C. pulveratrix*, suggesting that *M. yantaiensis* ingested more
510 C_4 plants than *C. pulveratrix*.

511 3. Intra-shell $\delta^{18}\text{O}$ variations revealed that there was a significant difference in the climatic
512 seasonality between glacial and interglacial periods. During the glacial periods (even-
513 numbered MIS), the seasonal contrast was large, whereas the seasonal contrast was small
514 during the interglacial periods (odd-numbered MIS). Stable isotope analyses of multiple
515 shells of the same snail species within each sampling layer showed that intra-species isotope
516 data were largely scattered during the glacial periods, indicative of highly unstable climates
517 change at sub-millennial scale, whereas intra-species isotopic difference was relatively small
518 during the interglacial periods, suggestive of a steady and uniform climatic condition within
519 millennium.

520 4. During MIS3 and MIS7, there were evidences of human activities around the Beiyao site,
521 but the corresponding climate background remained unclear. By analyzing whole-shell and

522 intra-shell $\delta^{18}\text{O}$ and faunal assemblage of the two species snails, we concluded that the
523 climates were relatively warm and humid with a weak seasonality. This stable climatic
524 condition was conducive to the regional expansion of prehistoric human activities.

525 **Acknowledgement:**

526 This work was supported by National Natural Science Foundation of China (Grant No.
527 41572163), the National Key R&D Program of China (Grant No. 2017YFA0603400), and
528 National Natural Science Foundation of China (Grant No. 41872080). Thanks to Yue Jiaojiao
529 for assistance in taking photo of the snails shells. Data for producing Figures 3–5 are
530 available from the data share website and the DOI is 10.6084/m9.figshare.12190485.

531

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