Diet and climatic context of giant birds inferred from delta C-13(c) and delta O-18(c) values of Late Palaeocene and Early Eocene eggshells from southern France

Article · January 2015

7 authors, including:

Romain Amiot
French National Centre for Scientific Research

Eric Buffetaut
French National Centre for Scientific Research

François Fourel
Claude Bernard University Lyon 1

François Martineau
Claude Bernard University Lyon 1

Palaeogeography, Palaeoclimatology, Palaeoecology 435 (2015) 210–221

Contents lists available at ScienceDirect

Palaeogeography, Palaeoclimatology, Palaeoecology

journal homepage: www.elsevier.com/locate/palaeo

Diet and climatic context of giant birds inferred from $\delta^{13}C_c$ and $\delta^{18}O_c$ values of Late Palaeocene and Early Eocene eggshells from southern France

D. Angst [a,*], R. Amiot [a], E. Buffetaut [b], F. Fourel [a], F. Martineau [a], N. Lazzerini [a], C. Lécuyer [a,c]

[a] UMR 5276, Laboratoire de Géologie de Lyon, Terre, Planètes et Environnement, Université Claude Bernard Lyon 1/CNRS/École Normale Supérieure de Lyon, 69622 Villeurbanne Cedex, France
[b] Centre National de la Recherche Scientifique, UMR 8538, Laboratoire de Géologie de l'Ecole Normale Supérieure, 75231 Paris Cedex 05, France
[c] Institut Universitaire de France, 103 Boulevard, Saint-Michel, 75005 Paris, France

ARTICLE INFO

Article history:
Received 15 September 2014
Received in revised form 3 June 2015
Accepted 9 June 2015
Available online 16 June 2015

Keywords:
Stable isotopes
Bird
Diet
Climate
Eggshell
Late Palaeocene
Early Eocene

ABSTRACT

Abundant fragments from eggs laid by giant birds occur in the Palaeocene (Thanetian) and Eocene (Sparnacian) sedimentary deposits of southeastern France. In the Sparnacian, thick eggshell fragments, assigned to the oospecies *Ornitholithus arcuatus*, correspond to very large bird eggs that were most likely laid by *Gastornis*. The Thanetian thin eggshell fragments, assigned to *Ornitholithus biroi*, were presumably laid by a smaller, yet unidentified bird. In order to investigate ecology and environment of these egg-laying birds, stable carbon and oxygen isotope compositions of 125 fossil eggshell fragments were analyzed. After removing samples affected by diagenetic alteration of the calcitic shells, the measured range of $\delta^{13}C_c$ values (−11‰ to −6‰ V-PDB) is interpreted as reflecting an herbivorous diet for these birds in a context of limited annual precipitation ($\approx$500 mm y^{-1}). Stable oxygen isotope analysis of living ostrich eggshell calcite, along with that of the water extracted from their albumen and yolk, provided evidence to calculate isotopic fractionation factors between both calcite and body water ($\alpha_{calcite–body\ water} = 1.03041$) and between body water and meteoric water ($\alpha_{bw–mw} = 1.00399$), using $\delta^{18}O$ values of local meteoric waters identified as the source of the birds drinking water. Combined with the $\delta^{18}O$ values of fossil eggshells, both isotopic fractionations provided calculated $\delta^{18}O$ values of meteoric waters in the range −9.5‰ to −2.8‰ (V-SMOW) for the Thanetian, and in the range −8.9‰ to −1.7‰ (V-SMOW) for the Sparnacian. These large isotopic ranges likely reflect inter-annual temperature variations of the complete year, suggesting a year round egg-laying strategy. Corresponding Mean Air Temperatures (MAT) were comprised between 20 ± 4 °C and 22 ± 4 °C during the Thanetian, and between 23 ± 3 °C and 25 ± 3 °C during the Sparnacian. These giant birds likely lived under a warm and dry climate similar to that prevailing today in western Mediterranean islands.

1. Introduction

Tertiary giant ground birds of the family Gastornithidae are currently known from the Palaeocene to the Middle Eocene in Europe, and from the Early Eocene in North America and Asia (e.g., Andors, 1992; Martin, 1992; Mayr, 2009; Buffetaut and Angst, 2014). Although this bird group has been known for a long time, its ecology is still poorly known and remains a matter of debate, even though an herbivorous diet was recently demonstrated with the combined use of stable carbon isotope ratios of *Gastornis* bones and a morphofunctional analysis of its mandible (Angst et al., 2014). Moreover, recent study of the abundant eggshell fragments known from Southern France indicates that these eggs were laid by giant birds belonging to the family Gastornithidae on the basis of the geometrical properties of eggshell fragments (Angst et al., 2015). This study confirms the attribution of at least a part of these eggshells to *Gastornis*, skeletal remains of which are known from the Early Eocene of southern France (Saint-Papoul, Aude: Buffetaut, 2008; Laurent et al., 2010; Bourdon et al., 2014). Since the 1960s, several studies were devoted to their microstructure (Touraine, 1960; Dughi and Sirugue, 1962; Penner, 1983; Donaire and López-Martínez, 2009), their ornamentation (Dughi and Sirugue, 1959; Touraine, 1960) and their stratigraphic distribution (Touraine, 1960; Fabre-Taxy and Touraine, 1960; Corroy and Touraine, 1961; Touraine, 1961, 1962; Dughi and Sirugue, 1968; Touraine, 1978; Donaire and López-Martínez, 2009; Buffetaut and Angst, 2014). Here the stable carbon and oxygen isotope compositions of these large bird eggshell fragments were measured and analyzed in order to investigate the nesting environment of these birds and to assess whether the egg laying was seasonally controlled or not.

Avian eggshell is a biomineralised composite ceramic consisting of calcium carbonate embedded in an organic matrix, both anchored to

---

* Corresponding author.
E-mail address: angst.delphine@gmail.com (D. Angst).

http://dx.doi.org/10.1016/j.palaeo.2015.06.011
0031-0182/

the shell membranes at the base of the internal side and protected in the external side by the cuticle (e.g., Tyler, 1969). In the fossil record, only the inorganic part is potentially preserved, and corresponds to about 95% of the shell made of calcium carbonate whose carbon is inherited from the ingested food whilst oxygen derives from the bird body water. Therefore the carbonate component of the shell can be used to estimate the bird diet, i.e., the δ^{13}C value of food and the δ^{18}O value of drinking water, shortly before and during shell formation (Von Schirnding et al., 1982). Provided that the egg was led by an herbivorous bird, the carbon isotope composition of its plant diet can be calculated using the carbonate-diet ^{13}C-enrichment established for modern birds (Von Schirnding et al., 1982; Angst et al., 2014). In turn, if the ingested plants use the C_3 photosynthetic pathway, then it is possible to estimate the average amount of local precipitations using the relationship established between extant C_3 plants and Mean Annual Precipitation (MAP; Diefendorf et al., 2010; Kohn, 2010). Local air temperature can be inferred from the oxygen isotope composition of the eggshell calcium carbonate. Indeed, the δ^{18}O value of the eggshell reflects a combination of the bird body temperature (most likely ranging from 37 to 41 °C) and the oxygen isotope composition of its drinking water, the source of which is ultimately meteoric waters. In turn, oxygen isotope compositions of meteoric waters are mainly controlled by surface temperatures, as significant relationships were established between Mean Air Temperature (MAT) and mean δ^{18}O values of meteoric waters for mid to high latitudes (e.g., Dansgaard, 1964; Fricke and O'Neil, 1999). Eggshell formation in birds is very fast, lasting between 15 and 20 h (e.g., Von Schirnding et al., 1982; Schaffner and Swart, 1991; Roberts, 2004; Hincke et al., 2010). The δ^{18}O value of calcium carbonate thus reflects a short time period, the most favourable one for egg-laying. Consequently, some isotopic variability is expected in response to fluctuating meteorological conditions on a yearly scale, and this variability can be assessed by analyzing a large number of eggshell fragments. Several studies were dedicated to investigating climatic conditions that have prevailed during the early Tertiary (Fricke et al., 1998; Clyde et al., 2001; Mosbrugger et al., 2005; Schmitz and Pujalte, 2007), and a warm and rather dry environment has been proposed for central Europe based on a palaeobotanical study (Mosbrugger et al., 2005).

In order to understand some aspects of the reproductive behaviour of *Gastornis* within the climatic framework of southeastern France, we have analyzed oxygen and carbon isotope compositions of well preserved eggshell fragments and interpreted them in terms of air temperatures and mean amounts of precipitation. These climatic data should bring another piece to the complex puzzle of the early Tertiary climatic modes, with a more precise knowledge of seasonal variations that prevailed in the southeastern part of France under the influence of the western Mediterranean Sea.

Therefore, this work will lead to a better understanding of the ecology of these large Tertiary birds, but also provide some new elements about the palaeoenvironments in southern France during the end of the Palaeocene and the beginning of the Eocene.

2. Material

2.1. Geological setting

Eggshell fragments are known from about twenty Late Palaeocene and Early Eocene sites in southern France and were attributed to large birds since the 1950s (Dughi and Sirugue, 1959).

The studied fragments of fossil bird eggshells were sampled from eight Tertiary sedimentary basins in southern France (Fig. 1), where the eggshell-bearing sedimentary beds are of Late Palaeocene (Thanetian) and Early Eocene (Sparnacian) ages (Dughi et al., 1969; Plaziat, 1984; Cojan et al., 2000; Donaire and López-Martínez, 2009). Among these eight basins, twenty fossiliferous sites were previously described as hosting bird eggshells (Dughi and Sirugue, 1959; Fabre-Taxy and Touraine, 1960; Touraine, 1960, 1961; Corroy and Touraine, 1961; Dughi and Sirugue, 1962; Touraine, 1962; Villatte, 1966; Dughi and Sirugue, 1968; Dughi et al., 1969; Touraine, 1978; Plaziat, 1984; Donaire and López-Martínez, 2009). During two field trips conducted between 2011 and 2012, more than 1300 eggshell fragments were unearthed from the following fifteen sites: Saint-Antonin-sur-Bayon, Suberoque, Saint-Maurin, Les Bardouines, La Neuve, Vinon-sur-Verdon, La Baraque, Saint-Julien-le-Montagné, La Mourotte, La Roquette, Pontevès, Sillans-la-Cascade, Les Mauquiers, Lagrasse and one site that has not been clearly identified yet but is located in the Cengle basin (Cengle indet.; Fig. 1).

Among these eggshell remains, two different types of cm-sized fragments were determined by considering the thickness of the eggshells; the thin one (1 mm thick), ascribed to the oospecies *Ornitholithus biroi*, is relatively uncommon (about 5% of the total egg fragments) compared to the thick one (2 mm thick), ascribed to the oospecies *Ornitholithus arcuatus*. At the locality of Les Bardouines, the well-documented stratigraphic sequence that these two types of eggshells were not contemporaneous. Indeed, the thin eggshells are always older (Thanetian) than the thick ones, being of Sparnacian age. As noted by Dughi and Sirugue (1968), the thin eggshells always occur in the red marls underlying the 'Calcaire de Saint-Marc Formation' whereas the thick eggshells are only found in the upper part of the Calcaire de Saint-Marc and the younger overlying red marls. Following the magnetostratigraphic and chemostratigraphic study of Cojan et al. (2000), the thin eggshells are found in the Thanetian, whereas the thick ones are Sparnacian in age (Fig. 2).

2.2. Sampling strategy of eggshell fragments

Within the two oospecies *O. arcuatus* and *O. biroi*, Dughi and Sirugue (1962) proposed a subdivision into four thick eggshells oospecies and one thin eggshell oospecies on the basis of microstructural and external ornamentation criteria. However, the most recent study of eggshell microstructures, by Penner (1983), considered only one oospecies per eggshell type: *O. arcuatus* and *O. biroi*. The thick eggshell fragments, attributed to *O. arcuatus*, correspond to very large eggs that have been laid by a giant bird, most likely *Gastornis* (Angst et al., 2015). Size could not be estimated for the thin-shelled eggs and the kind of bird that laid them remains uncertain. It should be remembered, however, that a relatively small species of *Gastornis*, *Gastornis russelli*, was described from the Late Palaeocene of Berru (NE France) by Martin (1992). It cannot be excluded that the *O. biroi* eggs were laid by a small species of *Gastornis* similar to *G. russelli*, which, to judge from the length of the tarsometatarsus, was about half the size of the larger *Gastornis parisiensis*.

Among the 1300 eggshells found during the field trips, 125 eggshells were analyzed for their stable carbon and oxygen isotope ratios. These analyzed eggshells belong to both oospecies and were sampled in the ten sites of Sillans-la-Cascade, Saint-Maurin, Les Bardouines, La Mourotte, Saint-Antonin, Les Mauquiers, Pontevès, Suberoque, Lagrasse and from the Cengle synclinal (Table 1).

Four extant chicken eggs and one extant ostrich egg, coming from geographic locations characterized by distinct δ^{18}O values of drinking waters (waters sampled in situ and analyzed in our laboratory or estimated using the IAEA database) were collected and their eggshell, albumen and yolk were analyzed (Table 2). These oxygen isotope compositions allowed determining the oxygen isotope fractionation between the birds body water and their drinking water.

3. Analytical techniques

3.1. Thin sections and the microstructure of the eggshells

Thin sections were cut at 'Laboratoire Transferts Lithosphériques', in Saint-Etienne, France. They consist of 30 μm thick slices of eggshell fragments cut perpendicular to the long axis of the shell, which were fixed to a glass slide with epoxy before examination with an optical microscope at 'Laboratoire de Géologie de Lyon' in Villeurbanne, France.

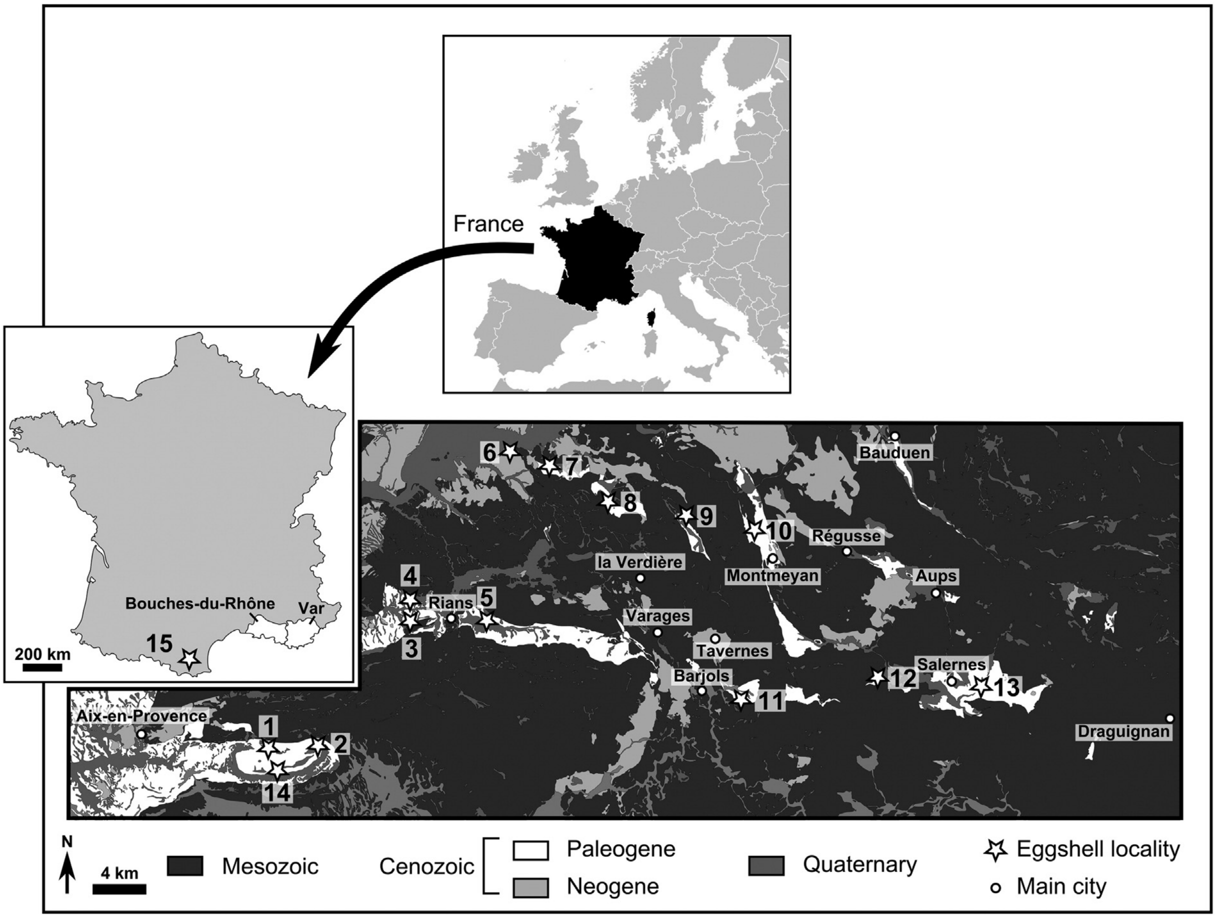

Fig. 1. Geographical and geological map of southern France showing the sites where the fossil bird eggshells were found. Site name: 1 — Saint-Antonin-sur-Bayon, 2 — Suberoque, 3 — Saint-Maurin, 4 — Les Bardouines, 5 — La Neuve, 6 — Vinon-sur-Verdon, 7 — La Baraque, 8 — Saint-Julien-le-Montagné, 9 — La Mourotte, 10 — La Roquette, 11 — Pontevès, 12 — Sillans-la-Cascade, 13 — Les Mauquiers, 14 — Cengle basin indet., 15 — Lagrasse.

Photomicrographs of the shell microstructure were taken at a 40× magnification (Fig. 3).

3.2. Oxygen isotope measurements of extant ostrich and chicken egg yolk, albumen and drinking waters

About 3 mL of albumen and yolk were pumped off with a syringe through a hole made with a drill in the thick ostrich eggshell, whereas the chicken egg yolk and albumen were delicately separated after breaking the egg and sampled with a syringe. Three aliquots of 300 μL of albumen and yolk were automatically reacted at 313 K with CO_2 and analyzed using a MultiPrep™ system on line with a GVI IsoPrime™ dual inlet isotope ratio mass spectrometer. The same analytical protocol was applied to three samples of local meteoric waters. Reproducibility of $\delta^{18}O$ values was estimated to be ±0.1‰ by normalizing raw data to the isotopic ratios of SMOW, SLAP and GISP international standards that were measured along with the samples.

3.3. Carbon and oxygen isotope measurements of bird eggshells

Bird eggshell fragments were cleaned in an ultrasonic bath with distilled water, and after drying at room temperature, the inner and outer surface layers were removed by mechanical abrasion in order to eliminate all the sedimentary residues. Each eggshell fragment was drilled with a diamond-head, throughout the complete thickness of the eggshell, and the resulting coarse powder was further ground in an agate mortar until a very fine powder was obtained.

Stable isotope ratios were determined by using an auto sampler MultiPrep™ system coupled to a dual-inlet GV Isoprime™ isotope ratio mass spectrometer (IRMS). For each sample, an aliquot of about 350 μg of calcium carbonate was reacted with anhydrous oversaturated phosphoric acid at 90 °C for 12 min. Carbon and oxygen isotope compositions are quoted in the δ notation in per mil relative to V-PDB. All samples were measured in duplicate and adjusted to the international reference NIST NBS19 analyzed during the same session. External reproducibility is ±0.1‰ for $\delta^{18}O$ values and ±0.05 for $\delta^{13}C$ values (1σ). Conversion of $\delta^{18}O$ of calcite from V-PDB to V-SMOW was performed according to Coplen et al. (1983) formula: $\delta^{18}O$ (‰V-SMOW) $= \delta^{18}O$ (‰V-PDB) $* 1.03091 + 30.91$.

4. Results

4.1. Diagenesis and microstructure of fossil bird eggshells

The photomicrographs of bird eggshell thin sections (Fig. 3) have revealed that the original structure of eggshells was fully preserved (except for one sample labelled eg#440), with the presence of two distinct layers (prismatic/palisade and mammillary) defining an organization shared by all the bird eggshells (Penner, 1983). Shell calcite from sample eg#440 form the "Cengle indet." is totally recrystallized and devoid of any remnant of its original structure (Fig. 3a), thus most likely revealing a diagenetic origin. For all the other eggshells, the preservation of the original microstructure of calcite crystals suggests that these fossils did not suffer any extensive process of recrystallization in the presence

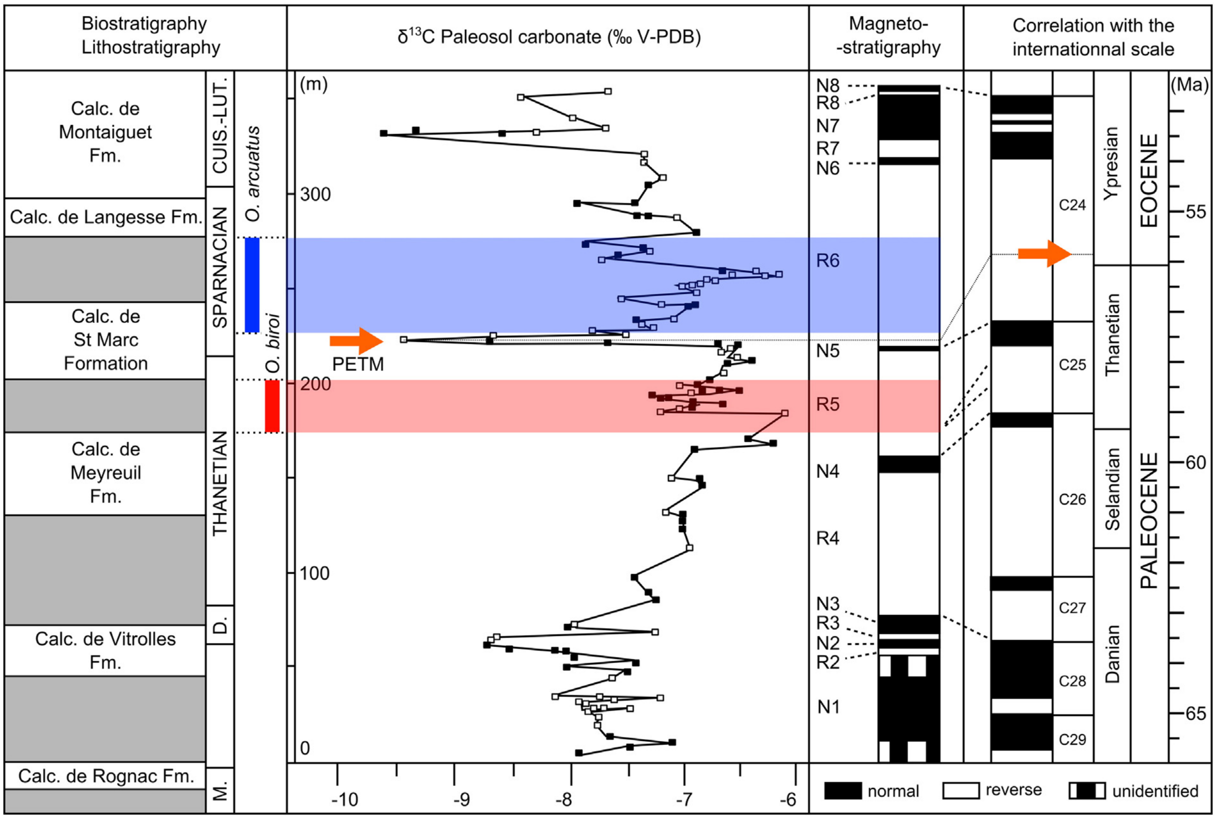

Fig. 2. Stratigraphic distribution of the *Ornitholithus biroi* and *Ornitholithus arcuatus* eggshells using a stratigraphic framework modified after Cojan et al. (2000), and correlated with the international scale.

of post-depositional crustal aqueous fluids, and are likely to have retained their pristine carbon and oxygen isotope compositions.

4.2. Body water of modern ostrich, chicken and their drinking water

The mean $\delta^{18}O$ values of water extracted from the albumen and yolk of the ostrich egg (Table 2, sample A1) are −7.10 ± 0.02‰ and −7.13 ± 0.06‰ (V-SMOW), and −6.43 ± 0.02‰ and −6.54 ± 0.11‰ (V-SMOW) for one chicken egg (Table 2, sample C1). These values are considered as both reflecting the oxygen isotope composition of the bird body water ($\delta^{18}O_{bw}$). Because no significant differences were observed between egg yolk and albumen, only albumen $\delta^{18}O$ value of samples SEL, MtP-1 and SaoP1 were measured. In parallel, the oxygen isotope compositions of waters drunk by the birds, which are of meteoric origin ($\delta^{18}O_{mw}$), have $\delta^{18}O_{mw}$ values ranging from −11.06‰ to −6.6‰ (V-SMOW). Combining these two oxygen isotope data, the isotopic fractionation factor $\alpha_{(bw-mw)}$ defined as $(\delta^{18}O_{bw} + 1000)/(\delta^{18}O_{mw} + 1000)$ equals 1.00399 between the body water of the ostrich and its drinking water, and 1.00442 ± 0.00016 between the body water of the 4 chicken and their drinking waters. It is noteworthy that the fractionation value is not influenced by the changing composition of the drinking water. The slight difference between the ostrich and chickens $\alpha_{(bw-mw)}$ being possibly related either to natural variability or to physiological, ecological or body mass differences.

4.3. $\delta^{13}C$ and $\delta^{18}O$ values of the bird eggshells

The present-day ostrich (*Struthio camelus*) and chicken (*Gallus gallus domesticus*) eggshells have $\delta^{18}O$ values of −7.59 ± 0.1‰ and ranging from −6.93 ± 0.12‰ to −4.25 ± 0.02‰ (V-PDB), respectively (Table 1). In the case of fossil birds, stable carbon and oxygen isotope compositions of calcite were measured on 125 fragments of eggshells (Table 1, Fig. 4). The $\delta^{13}C$ and $\delta^{18}O$ values range from −16.6‰ to −6.0‰ (V-PDB) and from −10.4‰ to 1.8‰ (V-PDB), respectively. It is noteworthy, however, that these isotopic ranges include one isolated 'outlier' data characterized by the lowest $\delta^{13}C$ and $\delta^{18}O$ values of −16.6‰ and −10.4‰, respectively, corresponding to the identified recrystallized eggshell fragment (see above). Excluding this sample from the database (eggshell #eg440), isotopic ranges become narrower with $\delta^{13}C$ and $\delta^{18}O$ values comprised between −11.1‰ and −6.0‰, and between −6.0‰ and 1.8‰, respectively. These isotopic ranges measured on fossil bird eggshells are bracketed by those documented for present-day bird eggshells ($-16.1‰ < \delta^{13}C < 4.4‰$; $-20.5‰ < \delta^{18}O < 10.2‰$) that encompass various terrestrial areas characterized by specific climate modes (Erben et al., 1979; Sarkar et al., 1991; Johnson et al., 1998).

Because recovered eggshell fragments display several degrees of wear on the outer surface, statistical tests (ANOVA) were performed to potentially detect an impact of the wear rate on both $\delta^{13}C$ and $\delta^{18}O$ values of the eggshells. The statistical tests are negative when considering the $\delta^{13}C$ and $\delta^{18}O$ values of thick eggshell fragments (*O. arcuatus*) for the different classes of ornamentations ($\delta^{13}C$: ANOVA-test: F = 0.5276 and P = 0.7864; and $\delta^{18}O$: ANOVA-test: F = 0.223 and P = 0.9686).

As previously described, two different oospecies of bird eggshells constitute the corpus of analyzed eggshells, which were not contemporaneous. No significant statistical difference is observed for the carbon isotope compositions between *O. arcuatus* of Sparnacian age and *O. biroi* of Thanetian age (Kruskal–Wallis Test: P = 0.8783), whereas a statistical difference is observed for the oxygen isotope compositions (ANOVA-test: F = 0.7225 and P = 0.008231).

5. Discussion

5.1. Oxygen isotope fractionation between body water and drinking water

Using the ingested water $\delta^{18}O_{mw}$ values and the body water $\delta^{18}O_{bw}$ values of ostrich and chickens, we estimated a body water–drinking

Table 1
Oxygen and carbon isotope composition of studied eggshell fragments reported along with sample number, oo-taxon, surface ornamentation, and origin. The * indicates the samples for which a thin section was done. Site abbreviations: Sillans: Sillans-la-Cascade, St-Antonin: Saint-Antonin-sur-Bayon, St-Maurin: Saint-Maurin, Cengle indet.: Cengle basin indet. Ornamentation abbreviations: AV: average vermiculation, AS: almost smooth, H: hilly, P: pitted, S: smooth, SV: strong vermiculation, TSV: thin and spaced vermiculation.

Sample #	Taxon	Eggshell thickness	Eggshell ornamentation	Locality	Age	$\delta^{13}C_c$ (‰ V-PDB)	SD.$\delta^{13}C_c$	$\delta^{18}O_c$ (‰ V-PDB)	SD.$\delta^{18}O_c$
A1	*Struthio camelus*			Livron	Present-day	−9.89	0.22	−7.59	0.10
C1	*Gallus gallus*			St-Nizier-le-Désert	Present-day	−7.03	0.01	−6.93	0.12
SEL	*Gallus gallus*			La Clayette (France)	Present-day	−10.70	0.07	−6.74	0.33
MtP-1	*Gallus gallus*			Mont Pilat (France)	Present-day	−0.20	0.04	−4.97	0.06
SaoP1	*Gallus gallus*			Sao Paulo (Brazil)	Present-day	2.16	0.01	−4.25	0.02
eg005	*O. arcuatus*	Thick	H	Sillans	Sparnacian	−6.26	0.06	0.45	0.01
eg034	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−6.29	0.10	−0.35	0.09
eg036	*O. arcuatus*	Thick	AS	Sillans	Sparnacian	−8.17	0.02	−4.15	0.06
eg037	*O. arcuatus*	Thick	AS	Sillans	Sparnacian	−7.69	–	−0.35	–
eg039	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−7.21	0.00	−0.24	0.01
eg045	*O. arcuatus*	Thick	AS	Sillans	Sparnacian	−6.75	0.06	−1.25	0.02
eg048	*O. arcuatus*	Thick	AV	Sillans	Sparnacian	−7.98	0.01	−1.50	0.01
eg062	*O. arcuatus*	Thick	P	Sillans	Sparnacian	−7.27	–	−3.06	–
eg104	*O. arcuatus*	Thick	SV	Sillans	Sparnacian	−6.58	–	−1.79	–
eg106	*O. arcuatus*	Thick	H	Sillans	Sparnacian	−6.80	–	−1.62	–
eg107*	*O. arcuatus*	Thick	SV	Sillans	Sparnacian	−6.31	0.06	−3.12	–
eg108	*O. arcuatus*	Thick	P	Sillans	Sparnacian	−7.51	0.12	−2.21	0.10
eg109	*O. arcuatus*	Thick	SV	Sillans	Sparnacian	−6.69	0.07	−1.53	0.25
eg121	*O. arcuatus*	Thick	H	Sillans	Sparnacian	−6.02	0.04	−1.97	0.03
eg123	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−10.61	0.04	−2.18	0.13
eg129	*O. arcuatus*	Thick	AV	Sillans	Sparnacian	−6.74	0.03	−0.59	0.05
eg131	*O. arcuatus*	Thick	AV	Sillans	Sparnacian	−7.26	0.05	−3.04	0.06
eg135	*O. arcuatus*	Thick	AV	Sillans	Sparnacian	−8.23	0.00	−2.39	0.05
eg144	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−7.92	0.10	0.10	0.09
eg151	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−7.78	0.01	−0.19	0.11
eg171	*O. arcuatus*	Thick	SV	Sillans	Sparnacian	−8.78	0.00	−1.13	0.06
eg172	*O. arcuatus*	Thick	SV	Sillans	Sparnacian	−9.28	0.02	−1.68	0.06
eg173	*O. arcuatus*	Thick	AS	Sillans	Sparnacian	−8.23	0.15	−0.63	0.13
eg174	*O. arcuatus*	Thick	H	Sillans	Sparnacian	−7.01	0.11	−2.77	0.03
eg186	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−7.42	0.01	−1.03	0.03
eg188	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−7.69	0.02	−3.92	0.05
eg189	*O. arcuatus*	Thick	AV	Sillans	Sparnacian	−8.06	0.01	−2.22	0.02
eg203	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−8.32	0.05	−2.15	0.09
eg212*	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−6.03	0.02	1.44	0.16
eg216	*O. arcuatus*	Thick	P	Sillans	Sparnacian	−9.00	0.02	−1.01	–
eg219	*O. arcuatus*	Thick	S	Sillans	Sparnacian	−8.13	–	−0.39	–
eg258	*O. arcuatus*	Thick	P	Sillans	Sparnacian	−8.13	–	1.55	–
eg273	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−8.55	0.02	−2.43	0.01
eg275	*O. arcuatus*	Thick	TSV	Sillans	Sparnacian	−8.55	0.04	−2.24	0.01
eg393	*O. biroi*	Thin	AS	St-Maurin	Thanetian	−8.00	0.06	−1.62	0.04
eg394*	*O. biroi*	Thin	AS	St-Maurin	Thanetian	−8.62	0.10	−1.35	0.34
eg395*	*O. biroi*	Thin	AS	St-Maurin	Thanetian	−11.11	0.02	−6.02	0.09
eg396*	*O. biroi*	Thin	S	St-Maurin	Thanetian	−8.47	0.02	0.67	0.28
eg397	*O. biroi*	Thin	S	St-Maurin	Thanetian	−11.11	0.06	−3.63	0.17
eg398	*O. biroi*	Thin	S	Les Bardouines	Thanetian	−7.19	0.03	−2.38	–
eg399*	*O. biroi*	Thin	S	Les Bardouines	Thanetian	−8.10	0.01	−3.99	0.04
eg400	*O. arcuatus*	Thick	P	Les Bardouines	Sparnacian	−9.54	0.01	−2.31	0.00
eg401	*O. arcuatus*	Thick	P	Les Bardouines	Sparnacian	−10.20	0.13	−4.01	0.09
eg402	*O. arcuatus*	Thick	P	Les Bardouines	Sparnacian	−8.66	0.16	−3.05	0.39
eg403	*O. arcuatus*	Thick	SV	Les Bardouines	Sparnacian	−10.18	0.08	−3.23	0.35
eg404	*O. arcuatus*	Thick	H	Les Bardouines	Sparnacian	−9.40	0.42	−3.13	0.63
eg405	*O. biroi*	Thin	S	La Mourotte	Thanetian	−8.62	0.05	−4.50	0.23
eg406	*O. biroi*	Thin	S	La Mourotte	Thanetian	−7.86	0.06	−2.57	0.11
eg407	*O. biroi*	Thin	S	La Mourotte	Thanetian	−7.92	0.05	−2.27	0.24
eg408	*O. biroi*	Thin	S	La Mourotte	Thanetian	−8.57	0.17	−1.93	0.20
eg409	*O. biroi*	Thin	S	La Mourotte	Thanetian	−8.70	0.12	−4.37	0.08
eg410	*O. arcuatus*	Thick	TSV	St-Antonin	Sparnacian	−9.71	0.09	−2.38	0.17
eg411	*O. arcuatus*	Thick	TSV	St-Antonin	Sparnacian	−7.78	0.03	−0.13	0.03
eg412	*O. arcuatus*	Thick	TSV	St-Antonin	Sparnacian	−8.32	0.02	0.62	0.19
eg413	*O. arcuatus*	Thick	TSV	St-Antonin	Sparnacian	−8.16	0.03	−0.21	0.10
eg414	*O. arcuatus*	Thick	TSV	St-Antonin	Sparnacian	−10.18	0.03	−3.45	0.12
eg415	*O. arcuatus*	Thick	P	St-Antonin	Sparnacian	−9.89	0.13	−1.32	0.03
eg416	*O. arcuatus*	Thick	S	St-Antonin	Sparnacian	−10.35	0.02	−3.35	0.34
eg417	*O. arcuatus*	Thick	H	St-Antonin	Sparnacian	−6.20	0.00	−2.11	0.03
eg418	*O. arcuatus*	Thick	H	St-Antonin	Sparnacian	−8.21	0.04	−0.13	0.03
eg419	*O. arcuatus*	Thick	AV	St-Antonin	Sparnacian	−8.19	–	−0.45	–
eg420	*O. arcuatus*	Thick	AV	St-Antonin	Sparnacian	−11.10	0.02	−3.89	0.01
eg421	*O. arcuatus*	Thick	SV	St-Antonin	Sparnacian	−8.68	0.07	−1.74	0.07
eg422	*O. arcuatus*	Thick	SV	St-Antonin	Sparnacian	−7.65	0.03	−1.87	0.11
eg423	*O. arcuatus*	Thick	SV	St-Antonin	Sparnacian	−8.85	0.02	−2.91	0.02
eg424	*O. arcuatus*	Thick	AS	Les Mauquiers	Sparnacian	−8.48	0.01	−2.97	0.01
eg425	*O. arcuatus*	Thick	AS	Les Mauquiers	Sparnacian	−8.57	0.03	−0.03	0.10
eg426	*O. arcuatus*	Thick	AS	Les Mauquiers	Sparnacian	−9.79	0.02	−2.19	0.04

Table 1 (*continued*)

Sample #	Taxon	Eggshell thickness	Eggshell ornamentation	Locality	Age	$\delta^{13}C_c$ (‰ V-PDB)	SD.$\delta^{13}C_c$	$\delta^{18}O_c$ (‰ V-PDB)	SD.$\delta^{18}O_c$
eg429	*O. arcuatus*	Thick	TSV	Ponteves	Sparnacian	−7.22	0.02	−1.40	0.00
eg430	*O. arcuatus*	Thick	TSV	Cengle indet.	Sparnacian	−10.30	0.07	−3.43	0.17
eg432*	*O. arcuatus*	Thick	TSV	Cengle indet.	Sparnacian	−9.13	0.02	−2.56	0.12
eg433	*O. arcuatus*	Thick	TSV	Cengle indet.	Sparnacian	−11.03	0.13	−2.98	0.07
eg434	*O. arcuatus*	Thick	P	Cengle indet.	Sparnacian	−7.42	0.07	−2.56	0.19
eg435	*O. arcuatus*	Thick	P	Cengle indet.	Sparnacian	−8.58	0.06	−2.70	0.03
eg436	*O. arcuatus*	Thick	SV	Cengle indet.	Sparnacian	−10.79	0.12	−3.91	0.01
eg437	*O. arcuatus*	Thick	SV	Cengle indet.	Sparnacian	−10.95	0.03	−3.77	0.04
eg438	*O. arcuatus*	Thick	SV	Cengle indet.	Sparnacian	−10.22	0.18	−2.55	0.51
eg439	*O. arcuatus*	Thick	SV	Cengle indet.	Sparnacian	−9.31	0.00	−1.25	0.16
eg440*	*O. arcuatus*	Thick	AV	Cengle indet.	Sparnacian	−16.60	0.15	−10.40	0.17
eg441	*O. arcuatus*	Thick	AV	Cengle indet.	Sparnacian	−10.72	0.01	−3.69	0.29
eg442	*O. arcuatus*	Thick	AV	Cengle indet.	Sparnacian	−7.87	0.01	−2.46	0.25
eg443	*O. arcuatus*	Thick	AV	Cengle indet.	Sparnacian	−7.50	0.17	−0.13	0.24
eg444	*O. arcuatus*	Thick	AV	Cengle indet.	Sparnacian	−10.12	0.05	−2.55	0.01
eg449	*O. arcuatus*	Thick	S	Suberoque	Sparnacian	−10.55	0.13	−1.78	0.24
eg450	*O. arcuatus*	Thick	S	Suberoque	Sparnacian	−9.28	0.02	−1.57	0.34
eg451	*O. arcuatus*	Thick	S	Suberoque	Sparnacian	−8.94	0.15	−2.12	0.16
eg452	*O. arcuatus*	Thick	P	Suberoque	Sparnacian	−8.44	0.05	−1.72	0.04
eg453	*O. arcuatus*	Thick	TSV	Suberoque	Sparnacian	−10.87	0.05	−3.36	0.10
eg454	*O. arcuatus*	Thick	TSV	Suberoque	Sparnacian	−10.77	0.08	−2.98	0.04
eg455	*O. arcuatus*	Thick	TSV	Suberoque	Sparnacian	−9.91	0.01	−2.14	0.00
eg456	*O. arcuatus*	Thick	AV	Suberoque	Sparnacian	−10.51	0.03	−2.46	0.02
eg457	*O. arcuatus*	Thick	AV	Suberoque	Sparnacian	−9.08	0.12	−1.26	0.06
eg458	*O. arcuatus*	Thick	SV	Suberoque	Sparnacian	−8.63	0.06	−1.69	0.03
eg459	*O. arcuatus*	Thick	SV	Suberoque	Sparnacian	−8.93	0.05	−1.74	0.01
eg460	*O. arcuatus*	Thick	H	Suberoque	Sparnacian	−7.44	0.10	−1.78	0.00
eg461	*O. arcuatus*	Thick	H	Suberoque	Sparnacian	−9.66	0.06	−3.12	0.03
eg463	*O. arcuatus*	Thick	H	Lagrasse	Sparnacian	−7.17	0.02	−0.39	0.04
eg464	*O. arcuatus*	Thick	H	Lagrasse	Sparnacian	−7.15	0.14	−0.34	0.07
eg465	*O. arcuatus*	Thick	H	Lagrasse	Sparnacian	−9.07	0.03	−1.25	0.00
eg466	*O. arcuatus*	Thick	AS	Lagrasse	Sparnacian	−9.01	0.02	−2.15	0.05
eg467	*O. arcuatus*	Thick	AS	Lagrasse	Sparnacian	−7.45	0.04	−0.39	0.25
eg468	*O. arcuatus*	Thick	AS	Lagrasse	Sparnacian	−7.78	0.11	−2.26	0.01
eg469	*O. arcuatus*	Thick	AS	Lagrasse	Sparnacian	−6.79	0.02	−1.56	0.03
eg470	*O. arcuatus*	Thick	AS	Lagrasse	Sparnacian	−7.04	0.04	−1.42	0.02
eg471	*O. arcuatus*	Thick	TSV	Lagrasse	Sparnacian	−9.86	0.15	−1.54	0.13
eg472	*O. arcuatus*	Thick	TSV	Lagrasse	Sparnacian	−9.39	0.08	−0.80	0.19
eg473	*O. arcuatus*	Thick	TSV	Lagrasse	Sparnacian	−8.58	0.19	−0.27	0.26
eg474	*O. arcuatus*	Thick	TSV	Lagrasse	Sparnacian	−9.31	–	−1.83	0.09
eg475	*O. arcuatus*	Thick	TSV	Lagrasse	Sparnacian	−8.46	–	−0.21	–
eg476	*O. arcuatus*	Thick	AV	Lagrasse	Sparnacian	−9.72	0.09	−3.83	0.00
eg477	*O. arcuatus*	Thick	AV	Lagrasse	Sparnacian	−9.25	0.00	−3.24	0.25
eg478	*O. arcuatus*	Thick	AV	Lagrasse	Sparnacian	−9.09	0.06	0.00	0.20
eg479	*O. arcuatus*	Thick	AV	Lagrasse	Sparnacian	−8.16	0.04	0.11	0.19
eg480*	*O. arcuatus*	Thick	AV	Lagrasse	Sparnacian	−7.99	–	1.83	–
eg481	*O. arcuatus*	Thick	SV	Lagrasse	Sparnacian	−7.37	–	0.63	–
eg482	*O. arcuatus*	Thick	SV	Lagrasse	Sparnacian	−9.41	–	−2.08	–
eg483	*O. arcuatus*	Thick	SV	Lagrasse	Sparnacian	−9.36	–	−0.34	–
eg484	*O. arcuatus*	Thick	SV	Lagrasse	Sparnacian	−10.32	0.00	−1.17	0.04
eg485	*O. arcuatus*	Thick	P	Lagrasse	Sparnacian	−8.41	0.02	−1.96	0.04
eg486	*O. arcuatus*	Thick	P	Lagrasse	Sparnacian	−9.02	0.01	−1.36	0.05
eg487	*O. arcuatus*	Thick	P	Lagrasse	Sparnacian	−8.10	0.03	−2.28	0.05

Table 2

Oxygen isotope composition of modern bird egg yolk and albumen (chicken and ostrich), their drinking water and calculation of the fractionation between body water and meteoric water. *In situ sampling and measurement in our lab.

Sample	Location	Coordinates	Taxon	Sample type	$\delta^{18}O_{bw}$ (‰ V-SMOW)	Sources	$\delta^{18}O_{bw}$ (‰ V-SMOW)	$\alpha_{(bw–mw)}$
A1	Livron (France)	44°46′N; 4°50′E	*Struthio camelus*	Egg yolk	−7.13 ± 0.06*			
				Egg albumen	−7.10 ± 0.02*			
				Egg mean	−7.11	Pond water	−11.06 ± 0.03*	1.00399
C1	St-Nizier-le-Désert (France)	46°03′N; 5°08′E	*Gallus gallus*	Egg yolk	−6.54 ± 0.11*			
				Egg albumen	−6.43 ± 0.02*			
				Egg mean	−6.49	Pond water	−11.06 ± 0.03*	1.00463
SEL	La Clayette (France)	46°17′N; 4°18′E	*Gallus gallus*	Egg albumen	−4.90 ± 0.07*	IAEA	−9.3	1.00444
MtP-1	Mont Pilat (France)	45°22′N; 4°34′E	*Gallus gallus*	Egg albumen	−3.98 ± 0.04*	Pond water	−8.13 ± 0.01*	1.00418
SaoP1	Sao Paulo (Brazil)	23°12′S; 47°31′W	*Gallus gallus*	Egg albumen	−2.11 ± 0.03	IAEA	−6.6 ± 0.2	1.00452

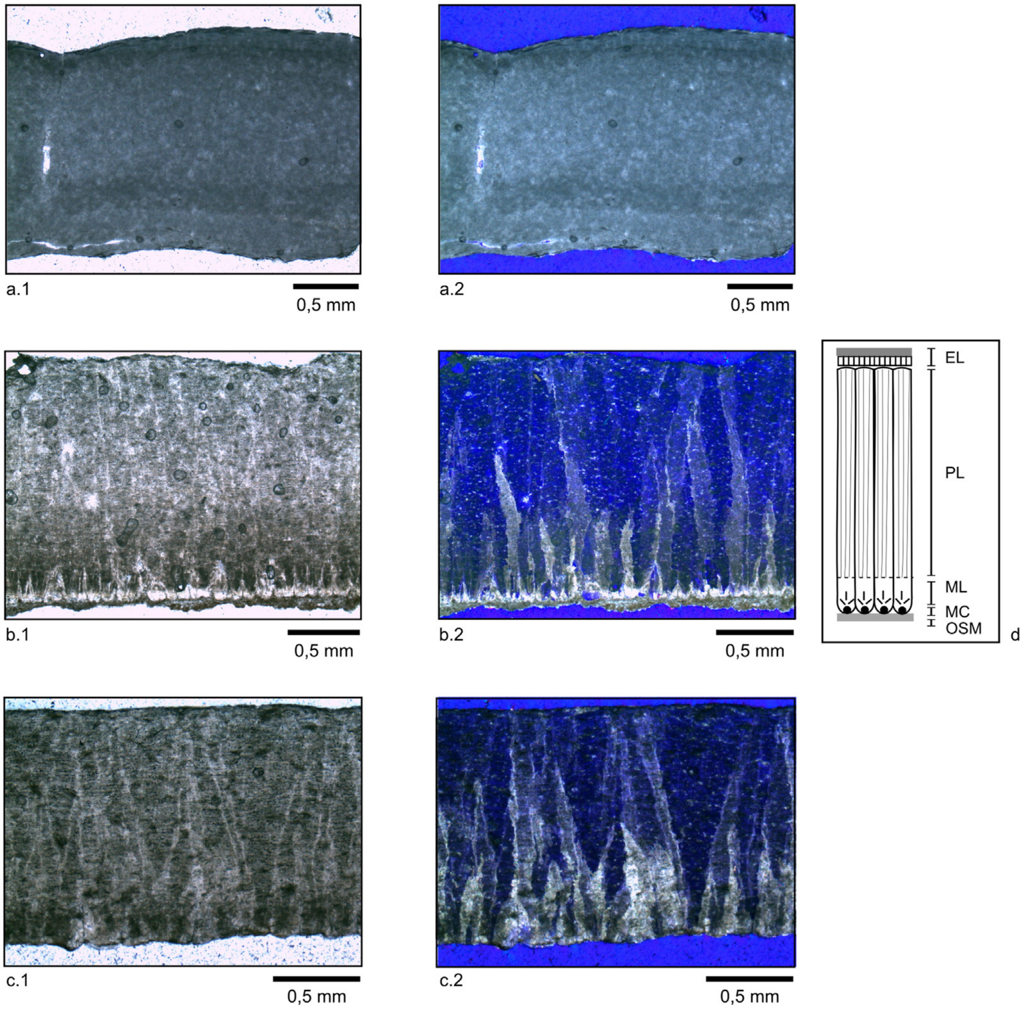

Fig. 3. Radial thin sections of two eggshells. a — Eggshell eg#440 which shows a total recrystallization, 1: observation in natural light, 2: observation in cross-polarized light; b — Eggshell eg#212 showing a good preservation of the thick bird egg microstructure without any secondary recrystallization, 1: observation in natural light, 2: observation in cross-polarized light; c — Eggshell eg#396 showing a good preservation of the thin bird egg microstructure without any secondary recrystallization, 1: observation in natural light, 2: observation in cross-polarized light; d — theoretical microstructure of a bird eggshell, EL: external layer, PL: prismatic layer, ML: mammilla layer, CL: mammilla core, OSM: organic shell membrane (not fossilized).

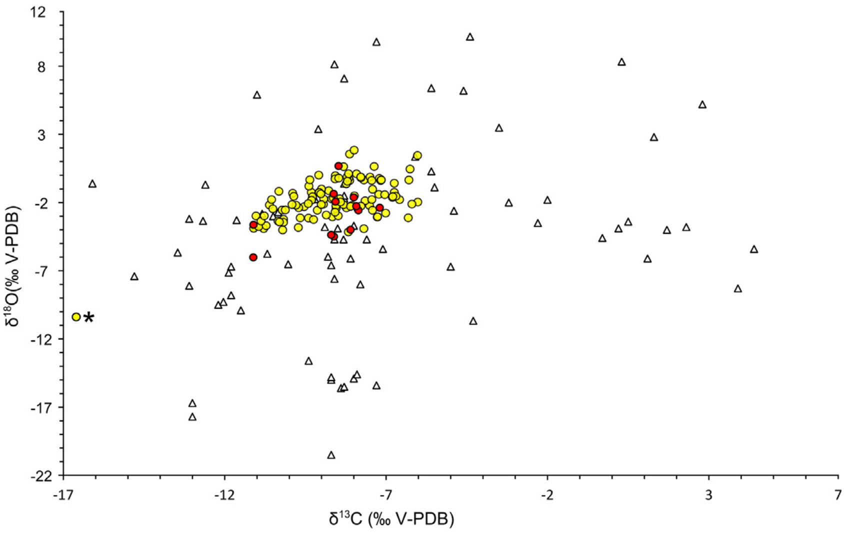

Fig. 4. Variations in $\delta^{13}C$ and $\delta^{18}O$ values of the fossil bird eggshells belonging to *O. biroi* (red dots) and *O. arcuatus* (yellow dots), compared with published isotope values of modern bird eggshells (white triangles).*: corresponds to the specimen #eg440 (Erben et al., 1979; Sarkar et al., 1991; Johnson et al., 1998).

water ^{18}O-enrichment of 3.93‰ for ostrich and 4.40 ± 0.13‰ for chicken. It is important to note that an ^{18}O-enrichment of about 3‰ to 6‰ between body and drinking water is common for terrestrial vertebrates (Kohn, 1996) including birds (Kohn, 1996), human beings (Longinelli, 1984), pigs (D'Angela and Longinelli, 1990) or cows (*Bos taurus*).

5.2. Oxygen isotope fractionation between bird egg calcite and body water

Ostrich (*S. camelus*) and chicken (*G. gallus domesticus*) have body temperatures close to 39 °C and 40 °C (Schmidt-Nielsen et al., 1969; Alexander et al., 2002). In the case of a rather constant body temperature, the oxygen isotope compositions (‰ V-SMOW) of egg calcite and body water for ostrich and chicken can be used to calculate the two corresponding isotopic fractionation factors $\alpha_{calcite–bw}$ that equal 1.03041 and 1.02951 ± 0.00066, respectively. These fractionation factors are close for the two birds within the analytical uncertainties. They are much higher than those calculated by applying to our data the oxygen isotope fractionation equation that was experimentally established by Kim and O'Neil (1997) between inorganic calcite and water ($\alpha_{calcite–bw} = 1.02569$). This result means that isotopic fractionation equations previously determined by O'Neil et al. (1969) and Kim and O'Neil (1997) between inorganic calcite and water cannot be applied to eggshells for calculating the δ^{18}O values of bird body waters. We emphasize, however, that the interpretation of such a difference is beyond the scope of this study. The oxygen isotope fractionation factor, $\alpha_{calcite–bw}$, defined for the present-day ostrich was used to interpret the $\delta^{18}O_c$ values of Palaeocene and Eocene giant bird eggshells by assuming that their body temperature was close to that of ostriches, i.e., 39 °C.

5.3. Diagenesis estimate

In addition to the obvious patterns of shell recrystallization that can be recognized by examining thin sections using optical microscopy, arguments based on geochemical data must also be considered to evaluate whether or not any kind of 'cryptic' diagenesis (i.e., the shell structure seems preserved) could have altered the samples and disturbed the original carbon and oxygen isotope compositions of the eggshell calcite. Both δ^{13}C and δ^{18}O values of bird eggshell fragments lie within the range of modern bird eggshell compositions. This observation cannot constitute a robust and definitive argument against the presence of a pervasive diagenetic alteration because under some conditions (temperature, water δ^{18}O, δ^{13}C of DIC (Dissolved Inorganic Carbon) and soil organic matter), it cannot be ruled out that some secondary calcium carbonates could have isotopic compositions that mimic those of eggshells. Diagenesis, however, is commonly responsible for two patterns of isotopic distributions in a δ^{13}C–δ^{18}O space. Under fluid-dominated conditions, isotopic compositions tend to become homogenous and appear strongly clustered when they are reported in a bivariate plot. Under mineral-dominated conditions, positive linear correlations are expected, reflecting binary mixtures between two end-members, which are on one side the original compositions of the eggshell calcite and on the other side a calcium carbonate that could result from the mineralization of soil organic matter in the presence of oxidizing fluids.

Indeed, the most likely scenario is a substitution of the initial calcium carbonate by a pedogenic carbonate mineral formed in a soil where the eggshells were buried and fossilized. As a general rule, the carbon isotope compositions of the secondary carbonates derive from the soil around the eggshell that is conditioned by the degradation of the vegetal cover. During the Palaeocene/Eocene, only a C_3-vegetation has flourished (Kohn, 2010), a degradation of the related organic matter would have generated a soil with δ^{13}C values comprised between −24‰ and −21‰ according to the carbon isotope compositions of bone carbonate from *Gastornis* and associated herbivorous mammal teeth (Angst et al., 2014). The pedogenic carbonates formed in such soils should have δ^{13}C values from −10‰ to −5‰ according to the isotopic fractionation ($14‰ < \Delta_{carbonate-organic\ matter} < 16‰$) that takes place during the oxidation of soil organic matter into carbonates (Stevenson et al., 2005). Unfortunately, the carbonate-organic matter carbon isotopic fractionation is similar to the egg carbonate-plant diet ^{13}C-enrichment, thus the calculated δ^{13}C values overlap those measured in fossil bird eggshells and cannot be used in this case to assess the original preservation of the carbon isotope composition of fossil eggshells.

In the case of oxygen isotopes, we can expect differences between the original values recorded in eggshells and those related to pedogenic carbonates. Indeed, the oxygen isotope composition of the eggshell carbonate depends on both bird body temperature and the oxygen isotope composition of the body water. Body water of terrestrial vertebrates is commonly ^{18}O-enriched by a few per mil relative to the diet water (mainly drinking water); the rate of heavy isotopic enrichment being mainly dependent on the metabolic activity of the bird and the body water turnover, itself dependent on the body mass. A large terrestrial bird such as the modern ostrich has a body mass in the 100–160 kg range and a body temperature between 38 °C and 41 °C (Schmidt-Nielsen et al., 1969). The ostrich living in southeast France, studied here, has a body water (corresponding to yolk and albumen) that is ^{18}O-enriched by 3.98‰ relative to its drinking water ($\Delta^{18}O_{bw–mw}$; Table 2). This isotopic enrichment factor between the body and drinking waters is applied to the Palaeocene giant birds assuming that the physiology and metabolism of *Gastornis* and related birds did not differ significantly from those of the present-day ostrich. As the mean annual δ^{18}O values of meteoric waters in Europe were most likely in the range −8‰ to −2‰ during the Eocene (Schmitz and Andreasson, 2001; Tütken, 2014), the δ^{18}O values of the contemporaneous fossil giant bird eggshells can be calculated by using the mean oxygen isotope fractionation factors $\alpha_{(bw–mw)}$ of 1.00399 and $\alpha_{(calcite–bw)}$ of 1.03041 that were defined above for the ostrich. In a first approximation, the fossil eggshells are expected to have δ^{18}O values ranging from −4.9 to 1.0‰ for a body temperature assumed to be 39 °C, thus matching the upper part of the range ($-6.0‰ < \delta^{18}O < 1.8‰$) of measured fossil eggshell δ^{18}O values (Fig. 4). This range of calculated δ^{18}O values of fossil eggshells, however, is much higher as a whole than that calculated for pedogenic carbonates ($-10.5‰ < \delta^{18}O < -2.7‰$) that would have been precipitated in isotopic equilibrium with the same meteoric waters at temperatures between 15 °C and 25 °C. These temperatures are considered as the minima and maxima of the range of mean annual air temperatures prevailing during the Palaeocene–Eocene (Sluijs et al., 2006).

Consequently, on that basis alone, the isotopic ranges obtained from the studied eggshell fragments suggest that at least the highest values cannot be of diagenetic origin. It cannot be totally ruled out that the studied fragments of bird fossil eggs having lower δ^{18}O values underwent diagenetic processes. Three main results, however, argue against an extensive diagenetic alteration of the studied samples. They rather support the assumption of alteration processes weak enough for allowing the interpretation of the isotopic data as original signatures of both diet and environment of the giant birds:

- In most cases, the microstructure of eggshells is preserved without any traces of calcite recrystallization or secondary carbonate deposit.
- In a bivariate plot, both δ^{13}C and δ^{18}O values of eggshell calcite constitute a large cloud of data without any positive correlation that could reflect an "isotopic contamination" through isotopic exchange or addition of carbonate derived from the mineralization of the soil organic matter.
- The highest δ^{18}O values of eggshell calcite cannot be explained by the replacement of the original calcium carbonate by pedogenetic carbonates formed by the oxidation of soil organic matter.

5.4. δ^{13}C, diet of giant birds and estimates of Mean Annual Precipitations (MAP)

Angst et al. (2014) have measured the ^{13}C-enrichment between carbonate and food of present-day carnivorous birds (vultures), which is +8.3‰, and much lower than that measured for herbivorous birds (ostriches) which ranges from +13.4‰ (Angst et al., 2014) to +16.2‰ (Johnson et al., 1998). As only C$_3$ plants were present during the Palaeocene, the average δ^{13}C values of -9.0 ± 1.4‰ measured in the bone carbonate of *Gastornis* remains can only be explained by an herbivorous diet. This giant bird, indeed, ingested plants having δ^{13}C values ranging from -24.3 ± 2.1‰ to -21.5 ± 2.1‰, which are typical of C$_3$ plants whose modern range is comprised between −37‰ and −20‰ (Kohn, 2010).

It is noteworthy that the δ^{13}C values of -9.0 ± 1.4‰ obtained from the carbonate component of *Gastornis* bones compare well with those measured in both *O. biroi* and *O. arcuatus* eggshells having δ^{13}C values of -8.69 ± 1.21‰ and -8.56 ± 1.30‰, respectively. This observation indicates that the giant birds that laid these eggs had a diet similar to that of *Gastornis*. In the case of *O. arcuatus* eggshells, our results reinforce the idea that they were at least closely related to *Gastornis* or that these eggs were laid by *Gastornis* itself during the Sparnacian in southern France. Considering the *O. biroi* eggshells, they are related to a yet unidentified bird which shared at least an isotopically similar diet with *Gastornis* during the Thanetian and may in fact have been a small species of *Gastornis* (see above).

As emphasized by Kohn (2010), C$_3$ plants are characterized by a large range of carbon isotope ratios (−37‰ to −20‰, V-PDB) that mainly results from physiological responses to hydric stress (δ^{13}C increases with aridity), varying sunlight and rates of leaf litter recycling. Diefendorf et al. (2010) and Kohn (2010) have established correlations between the δ^{13}C values of plant leaves and the Mean Annual Precipitation (MAP). In our study, the δ^{13}C values of plant leaves can be inferred from those of eggshells from herbivorous birds. A limit to this method can be introduced in case these giant birds had dietary preferences, which could result in a bias between the mean δ^{13}C value of the bird diet and that of the surrounding vegetation. However, the sizeable ranges in carbon isotope composition of eggshells measured at each fossil site and reaching up to 4.6‰ (locality of Sillans) suggest that these large birds have foraged on plants having contrasted carbon isotope compositions, thus making the use of the MAP-C$_3$ plants δ^{13}C value relationship relevant. In order to minimize bias related to animal dietary preferences, Kohn's method advises to use several different plant-eating taxa to estimate an average δ^{13}C value for the global vegetation within a given landscape. Unfortunately, despite more than 40 years of fieldwork, only eggshell fragments have been found in these fossil localities. The amount of Mean Annual Precipitations (MAP) is estimated by using Eq. (1) that was proposed by Rey et al. (2013), modified from the original equation established by Kohn (2010):

$$\mathrm{Log}_{10}(\mathrm{MAP}+300(\mathrm{mm\ \ y^{-1}})) = 0.092(\pm 0.004)\Delta^{13}\mathrm{C}_{\mathrm{leaf}} + 1.148(\pm 0.074) \quad (1)$$

with $\Delta^{13}\mathrm{C}_{\mathrm{leaf}} = (\delta^{13}\mathrm{C}_{\mathrm{atm}} - \delta^{13}\mathrm{C}_{\mathrm{leaf}}) / (1 + \delta^{13}\mathrm{C}_{\mathrm{leaf}} / 1000)$. The $\delta^{13}\mathrm{C}_{\mathrm{leaf}}$ values, which were estimated using the $\delta^{13}\mathrm{C}_{\mathrm{eggshells}}$ values, range from -23.49 ± 1.86‰ to -23.36 ± 1.90‰ considering a $\delta^{13}\mathrm{C}_{\mathrm{atm}}$ value for the Palaeocene atmosphere close to −5.5‰ according to Sinha and Stott (1994). Using Eq. (1), calculated MAP were 457 ± 336 mm y^{-1} and 439 ± 314 mm y^{-1} for *O. biroi* and *O. arcuatus*, respectively. This indicates that similar arid climatic conditions prevailed during the Late Palaeocene and the Early Eocene in southeastern France whilst the aridity decreased during the Palaeocene Eocene Thermal Maximum (PETM) according to Zachos et al. (2008). By contrast, Schmitz and Pujalte (2007) have shown that the climate of northern Spain was characterized by very marked rainy seasons at the time of the PETM, which cannot be detected in the range of δ^{13}C values of analyzed eggshells. The main reason for this is probably that the analyzed eggs from southern France are either older (*O. biroi*) or younger (*O. arcuatus*) than the PETM. It must be kept in mind that in the case of eggs, the δ^{13}C value of calcite records the diet of the bird when eggs were formed, thus sampling a very short period of the bird life, i.e., a few days (Erben et al., 1979; Von Schirnding et al., 1982; Schaffner and Swart, 1991; Stern et al., 1994; Johnson et al., 1998). The relative large range of δ^{13}C values measured in the eggshells fragments, and therefore the variation of the estimated amount of precipitation, may result from the various following processes:

- Inter-annual variations in the δ^{13}C of plants, themselves resulting from climatic yearly variations (sunlight, amount of precipitations).
- Differences of diet among a population of birds reflecting individual preferences or local variations in the δ^{13}C of eaten plants.
- Seasonal variations if the birds had several periods of egg-laying over the year.

Consequently, the difference between the amounts of precipitation in southeastern France and northern Spain (Schmitz and Pujalte, 2007) could result from a bias in the δ^{13}C record of eggshells directly linked to the ecology of birds. *Gastornis* and the other large Eocene birds, may have had only one annual egg-laying period just before or at the beginning of the rainy season (spring period), as modern Ostriches have (Deeming, 1999). These birds may also have laid throughout the year as currently observed at low latitudes where climatic conditions allow a permanent access to food resources (Perrins, 1985). Depending on which assumption is correct, these birds either laid during a relatively dry season, just before the beginning of the rainy season, or within an environment mostly dry all year long. The conclusion that the eggs were laid during dry periods is also supported by Donaire and López-Martínez (2009) who studied the porosity of the *O. arcuatus* eggshells found in northern Spain. These authors concluded that these eggs were laid in an open and dry nesting environment.

Whether the climate of southern France in the Late Palaeocene and basal Eocene was continuously or seasonally dry cannot be easily determined on the basis of our isotopic results from avian eggshells, for the reason explained above, viz. that eggshells record environmental conditions only during the short period of their formation. Few data are available about the climate of southern France at the time the eggshell-bearing formations were deposited. The lack of plant remains, for instance, makes it impossible to use traditional palaeobotanical proxies of climate. Sittler (1966) studied the clay minerals in Palaeogene formations in Provence, including the eggshell-bearing marls, and concluded that they had been formed under a warm climate with alternating wet and dry seasons. This interpretation can easily be reconciled with our results if it is assumed that *Gastornis* laid its eggs during a dry part of the year, so that the wetter conditions prevailing at other times were not recorded by the eggshells. In this connection, it should be remembered that birds that lay eggs throughout the year and therefore have more than one brood, are mainly passerines, and that this behaviour is not common in non-passerines (Perrins, 1985).

5.5. δ^{18}O and estimation of the mean air temperatures

Knowledge of both fractionation factors $\alpha_{(\mathrm{bw-mw})} = 1.00399$ and $\alpha_{(\mathrm{calcite-bw})} = 1.03041$, which were defined above for modern ostriches, enables the calculation of the δ^{18}O range of meteoric waters from the δ^{18}O values of bird eggshell calcite.

In a first approximation, it can be assumed that the complete range of eggshell δ^{18}O values (from −6.0‰ to 1.8‰ V-PDB) corresponds to original isotopic compositions. Following this scenario, it means that meteoric waters drunk by the giant fossil birds had δ^{18}O values ranging from about −8.9‰ to −1.7‰ (V-SMOW) for *O. arcuatus*, and from about −9.5‰ to −2.8‰ (V-SMOW) for *O. biroi*. These values seem in agreement with previous studies. Indeed, according to Tütken (2014), the mean $\delta^{18}O_{mw}$ value was close to −5‰ in Germany during the Middle Eocene. Considering more equable climatic conditions that prevailed during the Eocene as compared to present day ones, the magnitude of seasonal $\delta^{18}O_{mw}$ variations should not have exceeded ±3‰ against about ±5‰ today (Schmitz and Andreasson, 2001). Therefore, the plausible range of −8‰ to −2‰ for Palaeocene–Eocene meteoric waters in southern France matches the range estimated from fossil eggshell $\delta^{18}O$ values. Consequently, $\delta^{18}O$ values of ingested waters higher than −2‰ may be too high for the Eocene in Europe even during the warm season and could represent evaporated residual waters formed under arid or semi-arid conditions. Indeed, around the P/E boundary, cycles of humid-dry climatic conditions have been evidenced in northwestern France on the basis of stable carbon isotope compositions of terrestrial organic matter (Storme et al., 2012; Garrel et al., 2013).

Transposing the $\delta^{18}O$ values of meteoric waters into mean annual air temperatures far back into the past is a risky challenge. Indeed, present-day relationships linking these two parameters are well operating at mid to high latitudes whilst amount effects are dominating under low latitudes (Dansgaard, 1964). Europe was located at mid-latitudes during the Palaeocene–Eocene (Butterlin et al., 1993; Scotese, 2001), however, relative air humidity and trajectory of humid air masses may have been different from those prevailing today. Moreover, during the Palaeocene–Eocene, the $\delta^{18}O$ of the global oceanic water was closer to −1‰ than to 0‰ today, which corresponds to an ice-free world (see Zachos et al., 2001), and this shift needs to be taken into account when estimating air temperatures from the average meteoric water $\delta^{18}O$ values relationship.

The interpretation of these calculated $\delta^{18}O$ values of meteoric waters also depends on the ecology of *Gastornis* and related giant birds. They may have laid their eggs all over the year as some tropical present-day birds do (e.g., *Columbina passerina*, Campbell and Lack, 1985) or during one season when the food was abundant, for example the beginning of the spring as the ostrich does nowadays.

Considering the seasonality of about 3‰ proposed by Schmitz and Andreasson (2001), then the $\delta^{18}O_{mw}$ values range of about 7.2‰ (from −8.9‰ to −1.7‰) for *O. arcuatus* and 6.7‰ (from −9.5‰ to −2.8‰) for *O. biroi*, estimated from the eggshell fragments $\delta^{18}O_c$ values probably represent the whole intra-annual range of meteoric water $\delta^{18}O_{mw}$ values. Therefore, it is likely that *Gastornis* as well as the second bird species laid eggs all year round instead of only during a specific season. Assuming this hypothesis, Mean Air Temperatures (MAT) can be calculated using a proper MAT–$\delta^{18}O_{mw}$ relationship. According to the data provided by the IAEA/WMO over the whole world, MAT are related to the weighted mean annual $\delta^{18}O$ of meteoric waters as follows (Lécuyer, 2014):

$$\mathrm{MAT}(^\circ\mathrm{C}) = \left(\delta^{18}O_{mw} + 14.25\right)/0.481. \tag{2}$$

However, it is most likely that the present-day relationship does not apply to the Eocene. Secord et al. (2010) established two equations relating MAT to $\delta^{18}O_{mw}$ for the Eocene of North America (Bighorn Basin, Wyoming). These authors obtained $\delta^{18}O_{mw}$/MAT slopes of 0.36 ± 0.11 and 0.39 ± 0.24. Moreover, transposing the $\delta^{18}O_{mw}$/MAT relationship of Lécuyer (2014) to the Palaeocene–Eocene epoch requires to take into account the global oceanic value close to −1‰ instead of 0‰. Therefore, Eq. (2) becomes:

$$\mathrm{MAT}(^\circ\mathrm{C}) = \left(\delta^{18}O_{mw} + 14.25 + 1\right)/0.36 \tag{3}$$

or

$$\mathrm{MAT}(^\circ\mathrm{C}) = \left(\delta^{18}O_{mw} + 14.25 + 1\right)/0.39. \tag{4}$$

For $\delta^{18}O$ of meteoric waters comprised between −9.5‰ and −2.8‰ (average −6.3 ± 1.4‰; n = 12), these relationships result in mean air temperatures comprised between 20 ± 4 °C and 22 ± 4 °C for the Thanetian. Similarly, for $\delta^{18}O$ of meteoric waters comprised between −8.9‰ and −1.7‰ (average −5.3 ± 1.1‰; n = 112), these relationships result in mean air temperatures comprised between 23 ± 3 °C and 25 ± 3 °C for the Sparnacian. It is worth noting that these mean air temperatures matches that estimated by Tripati and Zachos (2000) who analyzed the oxygen isotope compositions of molluscs that lived in the coastal waters of Western Europe. These marine temperature estimates also compare well with those (from 20° to 24 °C) obtained by using the $\delta^{18}O$ of shark teeth from the northern Morocco phosphorite deposits (Lécuyer et al., 1993). Tindall et al. (2010) modelled the early Eocene climate by using a coupled ocean–atmosphere GCM and they obtained sea surface temperatures close to 25 °C for the palaeolatitude of 35–40° (Butterlin et al., 1993) that corresponded to that of southern France. In conclusion, both carbon and oxygen isotope compositions of the studied eggshells argue in favour of warm climatic conditions with arid episodes that prevailed during the end of the Palaeocene and the early Eocene in southern France.

As previously mentioned, the $\delta^{18}O$ values of the eggshell calcite show a statistical difference between the two types of eggshells: *O. biroi* and *O. arcuatus*, which are respectively Palaeocene and Eocene in age. However, due to the large uncertainties associated with these empirical calculations, we can only propose that the climatic conditions that prevailed during the Late Palaeocene in southeastern Europe returned after the PETM without any significant change in both air temperature and amount of precipitation.

5.6. Remarks on the ecology of Gastornis

Our results based on eggshell fragments attributed to gastornithids suggest that in southern France during the Palaeocene and basal Eocene these giant birds lived in an environment that was dry, even arid, during at least part of the year. Although this is in agreement with reconstructions based on the porosity of similar eggs from NE Spain (Donaire and López-Martínez, 2009), the postulated type of climate is different from that reconstructed for other *Gastornis* localities in Europe. This is especially striking when the results from Provence are compared with those obtained for the Middle Eocene localities of Germany (Messel, Geiseltal), for which climate reconstructions based on plant fossils (Mosbrugger et al., 2005; Grein et al., 2011; Teodoridis et al., 2012) indicate a climate that differs from that reconstructed from the French eggshells mainly by significantly higher precipitations (MAP up to 2540 mm for Messel and 2091 mm for the Geiseltal according to Teodoridis et al., 2012). For Messel, Grein et al. (2011, p. 311) suggest "a warm and humid, probably seasonal climate without extended dry seasons", which seems rather different from our reconstruction of the Late Palaeocene–basal Eocene of Provence, with at least a rather arid dry season. These discrepancies may be explained by differences in geographical location and geological age: it is not unexpected to find climatic differences between the basal Eocene of southern France and the Middle Eocene of western and central Germany. Be that as it may, the fact that the giant bird *Gastornis* could prosper in environments that seem to have been rather different in terms of climate, and presumably vegetation, is worth noting and certainly deserves further consideration within the framework of a general study of gastornithid habitats.

6. Conclusions

Carbon and oxygen isotope compositions of both Tertiary oospecies *O. biroi* (Thanetian) and *O. arcuatus* (Sparnacian) reveal that these eggshell fragments may be assigned to herbivorous birds that ate plants with a $\delta^{13}C$ values ranging from −24.3 ± 2.1‰ to −21.5 ± 2.1‰ (V-PDB). In addition, *O. arcuatus*, corresponding to the largest eggshells, could be related to the giant bird *Gastornis*. The calculated plant $\delta^{13}C$

values were used to estimate Mean Annual Precipitations (MAP) close to 500 ± 300 mm y^{-1}, assuming that the Palaeocene and Eocene giant birds did not have well-marked dietary preferences that could introduce biases in the calculation of MAP. Such low MAP values suggest that arid climatic conditions prevailed during the Late Palaeocene and the Early Eocene in southeastern France, a conclusion in agreement with that already proposed by Donaire and López-Martínez (2009) who analyzed the porosity of the eggshell *O. arcuatus* documented in northern Spain.

The combined stable isotope analyses of present-day bird eggshells (chicken and ostrich) allowed the calculation of two key isotopic fractionations for quantifying the isotopic composition of the water drunk by the Tertiary giant birds:

- The isotopic fractionation factor $\alpha_{\text{calcite–bw}}$ between ostrich eggshell calcite and body water with a value of 1.03041 instead of 1.02569, the latter value being predicted by the equation published by Kim and O'Neil (1997) established for inorganic calcite and water at a temperature of 39 °C considered as the mean body temperature of ostriches.
- The isotopic fractionation factor $\alpha_{\text{bw–mw}}$ between body water of large birds and meteoric water with a value of 1.00399.

The large isotopic ranges in estimated $\delta^{18}O$ values of meteoric waters for both the Thanetian ($-9.5‰$ to $-2.8‰$) and Sparnacian ($-8.9‰$ to $-1.7‰$) likely reflect inter-annual temperature variations of the complete year. Corresponding MAT were comprised between 20 ± 4 °C and 22 ± 4 °C during the Thanetian, and between 23 ± 3 °C and 25 ± 3 °C during the Sparnacian. These giant birds lived before and after the Palaeocene Eocene Thermal Maximum (PETM) under rather similar warm and dry conditions that resemble those prevailing nowadays in the western Mediterranean islands.

Acknowledgements

Thanks to F. Smektala, S. Giner, A. Méchin, P. Méchin, A. Amoros, L. Leroy, M. Guiomar, H. Tong and A. Martinez for their help in collecting the fossil eggshell. Thanks to M. and P. Grussenmeyer from La Ferme de l'Autruche Drômoise, Livron, for the samples of modern ostrich egg and the associated water. Thanks to S. Legendre for the samples of modern chicken egg and the associated water. Thanks to G. Blanchon-Pothet, from Le Parc aux Oiseaux, Villars-les-Dombes for the samples of modern birds eggshell. Thanks to C. Alboussière for the preparation of the thin section of the eggshells.

References

Alexander, B., de Carvalho, R.L., McCallum, H., Pereira, M.H., 2002. Role of the domestic chicken (*Gallus gallus*) in the epidemiology of urban visceral leishmaniasis in Brazil. Emerg. Infect. Dis. 8 (12), 1480–1486.

Andors, A.V., 1992. Reappraisal of the Eocene groundbird Diatryma (Aves: Anserimorphae). In: Campbell, K.E (Ed.), Papers in Avian Paleontology Honoring Pierce Brodkorb. Nat. Hist. Museum of Los Angeles County Science Series, pp. 109–125 (Los Angeles).

Angst, D., Lécuyer, C., Amiot, R., Buffetaut, E., Fourel, F., Martineau, F., Legendre, S., Abourachid, A., Herrel, A., 2014. Isotopic and anatomical evidence of an herbivorous diet in the Early Tertiary giant bird *Gastornis*. Implications for the structure of Paleocene terrestrial ecosystems. Naturwissenschaften 101 (4), 313–322.

Angst, D., Buffetaut, E., Lécuyer, C., Amiot, R., Smektalas, F., Giner, S., Méchin, A., Méchin, P.A., mouros, A., Leroy, L., Guiomar, M., Tong, H., Martinez, A., 2015. Fossil avian eggs from the Palaeogene of southern France: new size estimates and a possible taxonomic identification of the egg-layer. Geol. Mag. 152, 70–79. http://dx.doi.org/10.1017/S0016756814000077.

Bourdon, E., Mourer-Chauviré, C., Laurent, Y., 2014. The birds (Aves) from the Early Eocene of La Borie, southern France. Acta Palaeontol. Pol. 83.

Buffetaut, E., 2008. First evidence of the giant bird *Gastornis* from southern Europe: a tibiotarsus from the Lower Eocene of Saint-Papoul (Aude, southern France). Oryctos 7, 75–82.

Buffetaut, E., Angst, D., 2014. Stratigraphic distribution of large flightless birds in the Palaeogene of Europe and its palaeobiological and palaeogeographical implications. Earth Sci. Rev. 138, 394–408.

Butterlin, J., Vrielynck, B., Bignot, G., Clermonte, J., Colchen, M., Dercourt, J., Guiraud, R., Poisson, A., Ricou, L.E., 1993. Lutetian (46 to 40 Ma). In: Dercourt, J., et al. (Eds.), Atlas Tethys Paleoenvironmental Maps. Gauthier Villars, Paris, pp. 197–209.

Campbell, B., Lack, E., 1985. A Dictionary of Birds. Buteo Books, Vermillon (670 pp.).

Clyde, W.C., Sheldon, N.D., Koch, P.L., Gunnell, G.F., Bartels, W.S., 2001. Linking the Wasatchian/Bridgerian boundary to the Cenozoic Global Climate Optimum: new magnetostratigraphic and isotopic results from South Pass, Wyoming. Palaeogeogr. Palaeoclimatol. Palaeoecol. 167 (1), 175–199.

Cojan, I., Moreau, M.-G., Stott, L.E., 2000. Stable carbon isotope stratigraphy of the Paleogene pedogenic series of southern France as a basic for continental–marine correlation. Geology 28 (3), 259–262.

Coplen, T.B., Kendall, C., Hopple, J., 1983. Comparison of Stable Isotope Reference Samples.

Corroy, G., Touraine, F., 1961. Données nouvelles sur la stratigraphie du Grand-Cabri et du Cengle (Bouches-du-Rhône). Bull. Soc. Géol. Fr. 7 (3), 77–81.

D'Angela, D., Longinelli, A., 1990. Oxygen isotopes in living mammal's bone phosphate: further results. Chem. Geol. (Isot. Geosci. Sect.) 86 (1), 75–82.

Dansgaard, W., 1964. Stable isotopes in precipitation. Tellus 16 (4), 436–468.

Deeming, D.C., 1999. The Ostrich: biology, production and health. CABI, Oxon (358 pp).

Diefendorf, A.F., Mueller, K.E., Wing, S.L., Koch, P.L., Freeman, K.H., 2010. Global patterns in leaf ^{13}C discrimination and implications for studies of past and future climate. Proc. Natl. Acad. Sci. 107 (13), 5738–5743.

Donaire, M., López-Martínez, N., 2009. Porosity of Late Paleocene *Ornitholithus* eggshells (Tremp Fm, south-central Pyrenees, Spain): paleoclimatic implications. Palaeogeogr. Palaeoclimatol. Palaeoecol. 279 (3), 147–159.

Dughi, R., Sirugue, F., 1959. Sur les fragments de coquilles d'œufs fossiles de l'Eocène de Basse-Provence. C. R. Hebd. Séances Acad. Sci. 249, 959–961.

Dughi, R., Sirugue, F., 1962. Distribution verticale des œufs d'oiseaux fossiles de l'Eocène de Basse-Provence. Bull. Soc. Géol. Fr. 4 (1), 69–78.

Dughi, R., Sirugue, F., 1968. Marnes à œufs d'Oiseaux du Paléocène de Basse-Provence. Bull. Soc. Géol. Fr. 10 (5), 542–548.

Dughi, R., Plaziat, J.-C., Sirugue, F., 1969. La répartition stratigraphique des œufs d'Oiseaux du groupe d'*Ornitholithus arcuatus* D. et S. par rapport aux faunes d'eau douce et marines du Sparnacien et du Thanétien de l'Aude. CR Somm. Soc. Géol. Fr. 1, 9–10.

Erben, H.K., Hoefs, J., Wedepohl, K.H., 1979. Paleobiological and isotopic studies of eggshells from a declining dinosaur species. Paleobiology 5 (4), 380–414.

Fabre-Taxy, S., Touraine, F., 1960. Gisements d'œufs d'Oiseaux de très grande taille dans l'Eocène de Provence. C. R. Hebd. Séances Acad. Sci. 250, 3870–3871.

Fricke, H.C., O'Neil, J.R., 1999. The correlation between $^{18}O/^{16}O$ ratios of meteoric water and surface temperature: its use in investigating terrestrial climate change over geologic time. Earth Planet. Sci. Lett. 170, 181–196.

Fricke, H.C., Clyde, W.C., O'Neil, J.R., Gingerich, P.D., 1998. Evidence for rapid climate change in North America during the latest Paleocene thermal maximum: oxygen isotope compositions of biogenic phosphate from the Bighorn Basin (Wyoming). Earth Planet. Sci. Lett. 160 (1), 193–208.

Garrel, S., Schnyder, J., Jacob, J., Dupuis, C., Boussafir, M., Le Milbeau, C., Storme, J.-Y., Iakovleva, A.I., Yans, J., Baudin, F., Fléhoc, C., Quesnel, F., 2013. Paleohydrological and paleoenvironmental changes recorded in terrestrial sediments of the Paleocene–Eocene boundary (Normandy, France). Palaeogeogr. Palaeoclimatol. Palaeoecol. 376, 184–199.

Grein, M., Utescher, T., Wilde, V., Roth-Nebelsick, A., 2011. Reconstruction of the Middle Eocene climate of Messel using palaeobotanical data. N. Jb. Geol. Paläont. (Abh.) 260 (3), 305–318.

Hincke, M.T., Nys, Y., Gautron, J., 2010. The role of matrix proteins in eggshell formation. J. Poult. Sci. 47 (3), 208–219.

Johnson, B.J., Fogel, M.L., Miller, G.H., 1998. Stable isotopes in modern ostrich eggshell: a calibration for paleoenvironmental applications in semi-arid regions of southern Africa. Geochim. Cosmochim. Acta 62 (14), 2451–2461.

Kim, S.-T., O'Neil, J.R., 1997. Equilibrium and nonequilibrium oxygen isotope effects in synthetic carbonates. Geochim. Cosmochim. Acta 61 (16), 3461–3475. http://dx.doi.org/10.1016/S0016-7037(97)00169-5.

Kohn, M., 1996. Predicting animal $\delta^{18}O$: accounting for diet and physiological adaptation. Geochim. Cosmochim. Acta 60 (23), 4811–4829.

Kohn, M.J., 2010. Carbon isotope compositions of terrestrial C3 plants as indicators of (paleo)ecology and (paleo)climate. Proc. Natl. Acad. Sci. 107 (46), 19691–19695. http://dx.doi.org/10.1073/pnas.1004933107.

Laurent, Y., Adnet, S., Bourdon, E., Corbalan, D., Danilo, L., Duffaud, S., Fleury, G., Garcia, G., Godinot, M., Le Roux, G., Maisonnave, C., Métais, G., Mourer-Chauviré, C., Presseq, B., Sigé, B., Solé, F., 2010. La Borie (Saint-Papoul, Aude): un gisement exceptionnel dans l'Eocène basal du Sud de la France. Bull. Soc. Hist. Nat. Toulouse 146, 89–103.

Lécuyer, C., 2014. Water on Earth. Wiley, London (272 pp).

Lécuyer, C., Grandjean, P., O'Neil, J.R., Cappetta, H., Martineau, F., 1993. Thermal excursions in the ocean at the Cretaceous–Tertiary boundary (northern Morocco): $\delta^{18}O$ record of phosphatic fish debris. Palaeogeogr. Palaeoclimatol. Palaeoecol. 105, 235–243.

Longinelli, A., 1984. Oxygen isotopes in mammal bone phosphate: a new tool for paleohydrological and paleoclimatological research? Geochim. Cosmochim. Acta 48 (2), 385–390.

Martin, L.D., 1992. The status of the late Paleocene birds *Gastornis* and *Remiornis*. In: Campbell, K.E. (Ed.), Papers in Avian Paleontology Honoring Pierce Brodkorb. Nat. Hist. Museum of Los Angeles County Science Series, pp. 97–108 (Los Angeles).

Mayr, G., 2009. Paleogene Fossil Birds. Springer, Berlin (262 pp).

Mosbrugger, V., Utescher, T., Dilcher, D.L., 2005. Cenozoic continental climatic evolution of Central Europe. Proc. Natl. Acad. Sci. U. S. A. 102 (42), 14964–14969.

O'Neil, J.R., Clayton, R.N., Mayeda, T.K., 1969. Oxygen isotope fractionation in divalent metal carbonates. J. Chem. Phys. 51, 5547–5558.

Penner, M.M., 1983. Contribution à l'étude de la microstructure des coquilles d'œufs de dinosaures du Crétacé supérieur dans le bassin d'Aix-en-Provence: application biostratigraphique. Mém. Sci. Terre, Thèse Doct. Univ. Paris VI 83 (52), 1–234.

Perrins, C.M., 1985. Breeding season. In: Campbell, B., Lack, E. (Eds.), A Dictionary of Birds. T & AD Poyser, Calton, pp. 62–65.

Plaziat, J.-C., 1984. Le domaine pyrénéen de la fin du Crétacé à la fin de l'Eocène. Stratigraphie, paléoenvironnements et évolution paléogeographique. Thèse Doct. Univ. Paris Sud 2885, 1–234.

Rey, K., Amiot, R., Lécuyer, C., Koufos, G.D., Martineau, F., Fourel, F., Kostopoulos, D.S., Merceron, G., 2013. Late Miocene climatic and environmental variations in northern Greece inferred from stable isotope compositions ($\delta^{18}O$, $\delta^{13}C$) of equid teeth apatite. Palaeogeogr. Palaeoclimatol. Palaeoecol. 388, 48–57.

Roberts, J.R., 2004. Factors affecting egg internal quality and egg shell quality in laying hens. J. Poult. Sci. 41 (3), 161–177.

Sarkar, A., Bhattacharya, S.K., Mohabey, D.M., 1991. Stable-isotope analyses of dinosaur eggshells: paleoenvironmental implications. Geology 19 (11), 1068–1071.

Schaffner, F.C., Swart, P.K., 1991. Influence of diet and environmental water on the carbon and isotopic signatures of seabird eggshell carbonate. Bull. Mar. Sci. 48 (1), 23–38.

Schmidt-Nielsen, K., Kanwisher, J., Lasiewski, R.C., Cohn, J.E., Bretz, W.L., 1969. Temperature regulation and respiration in the ostrich. Condor 71 (4), 341–352.

Schmitz, B., Andreasson, F.P., 2001. Air humidity and lake $\delta^{18}O$ during the latest Paleocene–earliest Eocene in France from recent and fossil fresh-water and marine gastropod $\delta^{18}O$, $\delta^{13}C$, and $^{87}Sr/^{86}Sr$. Geol. Soc. Am. Bull. 113 (6), 774–789.

Schmitz, B., Pujalte, V., 2007. Abrupt increase in seasonal extreme precipitation at the Paleocene–Eocene boundary. Geology 35 (3), 215–218.

Scotese, C.R., 2001. Paleomap Project. PALEOMAP Project.

Secord, R., Gingerich, P.D., Lohmann, K.C., MacLeod, K.G., 2010. Continental warming preceding the Paleocene–Eocene thermal maximum. Nature 467 (7318), 955–958.

Sinha, A., Stott, L.D., 1994. New atmospheric pCO_2 estimates from paleosols during the late Paleocene/early Eocene global warming interval. Glob. Planet. Chang. 9 (3), 297–307.

Sittler, C., 1966. Le Paléogène de Provence. Vue d'ensemble sur la sédimentation argileuse. 90e Congrès des Sociétés savantes (Nice 1965). 2, pp. 25–49.

Sluijs, A., Schouten, S., Pagani, M., Woltering, M., Brinkhuis, H., Damsté, J.S.S., Dickens, G.R., Huber, M., Reichart, G.-J., Stein, R., 2006. Subtropical Arctic Ocean temperatures during the Palaeocene/Eocene thermal maximum. Nature 441 (7093), 610–613.

Stern, L.A., Johnson, G.D., Chamberlain, C.P., 1994. Carbon isotope signature of environmental change found in fossil ratite eggshells from a South Asian Neogene sequence. Geology 22 (5), 419–422.

Stevenson, B.A., Kelly, E.F., McDonald, E.V., Busacca, A.J., 2005. The stable carbon isotope composition of soil organic carbon and pedogenic carbonates along a bioclimatic gradient in the Palouse region, Washington State, USA. Geoderma 124 (1), 37–47.

Storme, J.Y., Dupuis, C., Schnyder, J., Quesnel, F., Garel, S., Iakovleva, A.I., Iacumin, P., Di Matteo, A., Sebilo, M., Yans, J., 2012. Cycles of humid dry climate conditions around the P/E boundary: new stable isotope data from terrestrial organic matter in Vasterival section (NW France). Terra Nova 24, 114–122.

Teodoridis, V., Kvaček, Z., Zhu, H., Mazouch, P., 2012. Environmental analysis of the mid-latitudinal European Eocene sites of plant macrofossils and their possible analogues in East Asia. Palaeogeogr. Palaeoclimatol. Palaeoecol. 333, 40–58.

Tindall, J., Flecker, R., Valdes, P., Schmidt, D.N., Markwick, P., Harris, J., 2010. Modelling the oxygen isotope distribution of ancient seawater using a coupled ocean–atmosphere GCM: implications for reconstructing early Eocene climate. Earth Planet. Sci. Lett. 292 (3), 265–273.

Touraine, F., 1960. Œufs d'Oiseaux de très grande taille dans l'Eocène inférieur de Provence. Bull. Soc. Géol. Fr. 2 (6), 783–789.

Touraine, F., 1961. Les œufs d'Oiseaux éocènes et la stratigraphie du Cengle (B-du-Rh). CR Somm. Soc. Géol. Fr. 4, 90–91.

Touraine, F., 1962. Le synclinal danien-éocène de la Mourotte (Var). Bull. Soc. Géol. Fr. 4 (1), 87–91.

Touraine, F., 1978. Le fossé de Montmeyan (Var). Bull. BRGM 1 (2), 89–109.

Tripati, A., Zachos, J., 2000. Paleocene and Eocene costal ocean temperatures. GFF 122, 171–172.

Tütken, T., 2014. Isotope compositions (C, O, Sr, Nd) of vertebrate fossils from the Middle Eocene oil shale of Messel, Germany: implications for their taphonomy and palaeoenvironment. Palaeogeogr. Palaeoclimatol. Palaeoecol. 416, 92–109.

Tyler, C., 1969. Avian egg shells: their structure and characteristics. Int. Rev. Gen. Exp. Zool. 4, 81–130.

Villatte, J., 1966. Découverte de fragments de coquille d'œufs d'Oiseaux dans l'Eocène inférieur de l'Aude. CR Somm. Soc. Géol. Fr. 1966, 345–346.

Von Schirnding, Y., Van Der Merwe, N.J., Vogel, J.C., 1982. Influence of diet and age on carbon isotope ratios in ostrich eggshell. Archaeometry 24 (1), 3–20.

Zachos, J., Pagani, M., Sloan, L., Thomas, E., Billups, K., 2001. Trends, rhythms, and aberrations in global climate 65 Ma to present. Science 292 (5517), 686–693.

Zachos, J.C., Dickens, G.R., Zeebe, R.E., 2008. An early Cenozoic perspective on greenhouse warming and carbon-cycle dynamics. Nature 451 (7176), 279–283.