PROCEEDINGS B

royalsocietypublishing.org/journal/rspb

Research



Cite this article: Cramp LJE *et al.* 2019 Regional diversity in subsistence among early farmers in Southeast Europe revealed by archaeological organic residues. *Proc. R. Soc. B* **286**: 20182347. http://dx.doi.org/10.1098/rspb.2018.2347

Received: 18 October 2018 Accepted: 14 December 2018

Subject Category:

Evolution

Subject Areas:

evolution, ecology, behaviour

Keywords:

lipid biomarkers, organic residues, neolithic, early farmer, aquatic, pottery

Author for correspondence:

Lucy J. E. Cramp e-mail: lucy.cramp@bris.ac.uk

Electronic supplementary material is available online at https://dx.doi.org/10.6084/m9. figshare.c.4346378.



Regional diversity in subsistence among early farmers in Southeast Europe revealed by archaeological organic residues

Lucy J. E. Cramp¹, Jonathan Ethier², Dushka Urem-Kotsou³, Clive Bonsall⁴, Dušan Borić⁵, Adina Boroneanţ⁶, Richard P. Evershed⁷, Slaviša Perić⁸, Mélanie Roffet-Salque⁷, Helen L. Whelton⁷ and Maria Ivanova²

¹Department of Anthropology and Archaeology, University of Bristol, 43 Woodland Road, Bristol BS8 1UU, UK ²Institut für Ur-und Frühgeschichte und Vorderasiatische Archäologie, Universität Heidelberg, Marstallhof 4, 69117 Heidelberg, Germany

³Department of History and Ethnology, Democritus University of Thrace, Tsaldari 1, Komotini 694100, Greece ⁴School of History, Classics and Archaeology, University of Edinburgh, Old Medical School, Teviot Place, Edinburgh EH8 9AG, UK

⁵The Italian Academy for Advanced Studies in America, Columbia University, 1161 Amsterdam Avenue, New York, NY 10027, USA

⁶ Vasile Pârvan' Institute of Archaeology, Romanian Academy, Henri Coandă Strada 11, Bucharest 010667, Romania

⁷Organic Geochemistry Unit, School of Chemistry, University of Bristol, Cantock's Close, Bristol BS8 1TS, UK ⁸Institute of Archaeology, Knez Mihailova 35/4, 11000 Belgrade, Serbia

IJEC, 0000-0003-2012-2753; CB, 0000-0003-0243-3232; RPE, 0000-0002-9483-2750

The spread of early farming across Europe from its origins in Southwest Asia was a culturally transformative process which took place over millennia. Within regions, the pace of the transition was probably related to the particular climatic and environmental conditions encountered, as well as the nature of localized hunter-gatherer and farmer interactions. The establishment of farming in the interior of the Balkans represents the first movement of Southwest Asian livestock beyond their natural climatic range, and widespread evidence now exists for early pottery being used extensively for dairying. However, pottery lipid residues from sites in the Iron Gates region of the Danube in the northern Balkans show that here, Neolithic pottery was being used predominantly for processing aquatic resources. This stands out not only within the surrounding region but also contrasts markedly with Neolithic pottery use across wider Europe. These findings provide evidence for the strategic diversity within the wider cultural and economic practices during the Neolithic, with this exceptional environmental and cultural setting offering alternative opportunities despite the dominance of farming in the wider region.

1. Introduction and background

Across most of Europe (with notable exceptions including the Baltic region), the transition towards an economy based upon domesticated animals and plants was accompanied by the first appearance of pottery. While the timing, pace and nature of the shift towards farming varied, the study of archaeozoological assemblages and pottery organic residues supports scenarios whereby domesticated ruminants (cattle, sheep and goat) became the predominant animals exploited, accompanied by smaller numbers of wild ruminants (e.g. cervids and aurochs) and non-ruminants (such as domesticated pig, wild boar and fish/shellfish). Alongside evidence for ruminant meat, dairy products have

© 2019 The Authors. Published by the Royal Society under the terms of the Creative Commons Attribution License http://creativecommons.org/licenses/by/4.0/, which permits unrestricted use, provided the original author and source are credited.

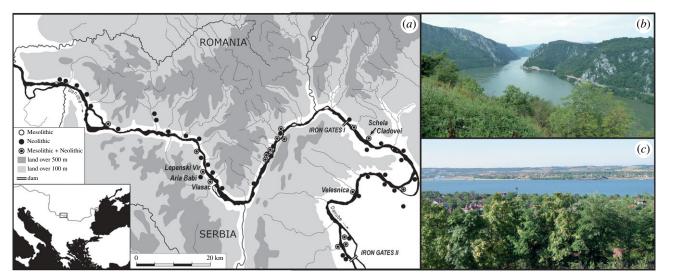


Figure 1. (*a*) Map of the Iron Gates region showing the location of important Mesolithic and Neolithic sites, including those investigated here. (*b*,*c*) The modern landscape of the gorges and downstream area, respectively.

also been identified in pottery organic residues as early as the seventh millennium BC in Anatolia and Syria [1–4], and subsequently in Neolithic pottery from Southeastern Europe [1,5–7], the Mediterranean [8], and Central [9–11], Northwestern [12,13] and Northern [14,15] Europe. While exploitation of fish and shellfish is evidenced from Early Neolithic coastal settlements in some areas including Southeastern Europe and the Bosporus (e.g. [16,17]), this ceases to comprise the staple component of the economy, and in some regions is conspicuously absent, even leading to suggestions of an ideological taboo against aquatic foods [18].

From Southwest Asia, farming spread into Southeastern Europe, reaching the interior of the Balkans by the seventh millennium BC. This marked the point at which Southwest Asian livestock was introduced beyond their natural climatic zone into temperate Europe [6,19,20], and preferences in animal and plant species exploitation have been shown to correlate with the regional bioclimatic setting as farming dispersed northwards [19]. The study of archaeozoological assemblages and organic residues from Early Neolithic pottery from the northern Balkans and Carpathian Basin demonstrate widespread and frequent evidence for dairying; the intensification of this existing practice may have been an adaptive strategy to the new environmental conditions within which animals were being raised [6].

In the northern Balkans, straddling the modern political border between Romania and Serbia, lies the Iron Gates region of the Danube, also known as the Danube Gorges (figure 1). This is a unique landscape encompassing the mountainous topography of the gorges where the Danube cuts through the junction of the Carpathian and Balkan mountain chains, and the lower lying, more open landscape of the 'downstream area' where the river begins its journey across the Wallachian Plain towards the Black Sea (figure 1b,c). This region is home to important sites that span the crucial interface between Mesolithic fisher-hunter-gatherers and the earliest farmers in the wider landscape of the central and northern Balkans. At least 18 fisher-hunter-gatherer sites dating from the Late Glacial and early Holocene were revealed by archaeological surveys and excavations undertaken during the construction of two dams between 1964 to 1971 and 1977 to 1984, some of which may have been permanent or semipermanent settlements with semi-subterranean dwellings and formal burial grounds (e.g. [21-29]).

By the end of the seventh millennium BC, the appearance of Starčevo pottery, new crouched burial positions and domesticates in the Iron Gates region coincides with the spread of Starčevo-Körös-Criş farmers throughout the central and northern Balkans [24,30,31]. The appearance of these new cultural traits indicates the presence of, or contact with, farming communities. While faunal assemblages from early excavations in the region are affected by stratigraphic uncertainty [32], recent excavations at the downstream site of Schela Cladovei have revealed significant quantities of domesticated species during the Early Neolithic [33]. However, the gorges were arguably less well suited to farming and wild mammals continued to be important in the subsequent economy [31,34-36]. The abundance and variety of the Danube's fish resources, which included both freshwater (e.g. carp and catfish) and migratory marine (e.g. some sturgeon) species, provided a potentially rich wild resource for any groups living in the area. It seems likely therefore that farmers, who may already have incorporated some fishing into their subsistence practices, were attracted by the aquatic resources of the Danube and the fertile alluvial soils along its banks. Evidence from Sr isotopes and ancient DNA is indicative of increased mobility and genetic mixing of farmer and local forager ancestry among the populations buried in the Iron Gates [37,38] around the time Starčevo farmers are visible archaeologically in the wider landscape. Stable carbon and nitrogen isotope values of dated human bone collagen from Mesolithic and Neolithic human bones from the Iron Gates moreover indicate a broad shift towards the inclusion of more terrestrial resources in the diet, although fish continued to be a predominant component of the diets of some individuals even by the early sixth millennium BC [39-41]. That this may not have been a continuous or linear process [42,43] provides further insights into the issue of continuity of occupation and the precise nature of interactions between foragers and farmers.

Organic residues that accumulate and survive in the fabric of pottery vessels provide a means to directly determine the origins of products processed in them, such as animal fats, plant oils, leafy plants, resins, beeswax and

bitumen [44] based upon their biomolecular 'fingerprints'. Determination of the stable carbon isotope value of individual fatty acids enables further classification of animal fats, separating ruminant (e.g. cattle, sheep, goat, deer) from non-ruminant (e.g. pig, wild boar), freshwater and marine products, and fats of dairy origin [12,45,46]. The reasonably high likelihood of mixing of products in pots means that while there will be 'over-printing' of different biomolecular fingerprints, the stable isotope signatures of surviving animal fatty acids will be an integrated signature of potentially more than one origin and will reflect the major contributing source, or plot between categories.

Here we investigate preserved organic residues from over 200 Neolithic ceramic vessels from the Iron Gates and assess the wider implications for our understanding of Early Neolithic adaptations in Southeast Europe and the role of ceramic technology.

2. Material and methods

A total of 217 Neolithic sherds were selected for organic residue analysis from five settlements located in the Iron Gates region of the Lower Danube (figure 1). Two sites (Lepenski Vir and Vlasac) are located in the steep, upper gorges, positioned on narrow alluvial strips along the river bank. A third settlement, Aria Babi, is situated in the immediate hinterland of the gorges, on a hill above Lepenski Vir. The other sites, Schela Cladovei and Velesnica, are located in the more open landscape downstream of the gorges. Descriptions of individual sites, details of the sherds analysed and their archaeological contexts are provided in the electronic supplementary material (text S1, tables S1 and S2). Vessel forms sampled comprised predominantly bowls and jars, deriving from pits or 'cultural layers' (see electronic supplementary material, tables S1 and S2). Considerable debate continues regarding relative chronologies within and between sites. However, all pottery analysed here is of Starčevo-Criştype, which is broadly dateable to the late seventh to early sixth millennium BC. A summary of the archaeozoological assemblages is given in figure 3 and the electronic supplementary material, table S3.

Lipid extracts were obtained and prepared from pottery sherds using well-established protocols published elsewhere [12,47] and detailed in the electronic supplementary material, text S2. Selected lipid extracts were investigated using GC/MS-SIM for high-sensitivity detection of ω -(o-alkylphenyl)alkanoic acids (APAAs) and dihydroxy acids (DHFAs; see electronic supplementary material, text S2 and figure S1). GC/C/IRMS analysis was performed on fatty acid methyl esters from residues identified as animal fats for the determination of δ^{13} C values of individual fatty acids ($C_{16:0}$ and $C_{18:0}$). Instrumental conditions are given in the electronic supplementary material, text S2. The δ^{13} C values were derived according to the following expression and are relative to the international standard VPDB: $\delta^{\bar{1}3}C\% =$ $((R_{\text{sample}} - R_{\text{standard}})/R_{\text{standard}}) \times 1000 \text{ where } R = {}^{13}\text{C}/{}^{12}\text{C}.$ The δ^{13} C values were corrected for the carbon atoms added during methylation using a mass balance equation [48].

3. Results and discussion

(a) Evidence of aquatic products in the Neolithic pottery

From over 200 pottery sherds sampled, 45 contained preserved characterizable organic residues. The majority were dominated by saturated $C_{16:0}$ and $C_{18:0}$ fatty acids (see electronic supplementary material, table S2 and figure S1A–C), which are the most abundant fatty acids in degraded animal fats. The determination of the δ^{13} C values of these individual fatty acids enables further classification of their origins [12]. While absolute δ^{13} C values may express regional differences, plotting the $\Delta^{13}C$ ($\delta^{13}C_{18:0} - \delta^{13}C_{16:0}$) values against $\delta^{13}C_{16:0}$ values minimizes these effects, emphasizing the differences in metabolic physiologies [49] (figures 2 and 3). In contrast to nearly all previously published Early and Middle Neolithic European investigations, the stable carbon isotope determinations show that non-ruminants comprise a major source of the products processed in the pottery from this location, with non-ruminant signatures dominant in over 50% of residues. In the Iron Gates ecosystem, and taking into account the faunal records, non-ruminants could comprise both aquatic resources from the Danube, porcine fats (wild or domestic), as well as dog. By using gas chromatography/mass spectrometry operated in selected ion monitoring mode, the widespread (70%; figure 2) presence of long-chain ($\geq C_{20}$) vicinal diols and ω -(oalkylphenyl)alkanoic acids was detected in the non-ruminant residues (and in a further three residues with ruminant stable isotope signatures), which reflect the presence of precursor long-chain mono- and polyunsaturated fats respectively [46,54-58] (see electronic supplementary material, table S2 and figure S1), abundant in fresh aquatic fats. This confirms that aquatic products were regularly processed in over half of the Early Neolithic pots across all five Iron Gates settlements.

Owing to the relatively low content of $C_{18:0}$ in aquatic fats, their contribution to the measured stable isotope signature would readily be masked isotopically by mixing with terrestrial animal fats and could plot within the ruminant ranges (see electronic supplementary material, figure S2). Therefore, aquatic resources must have been the predominant commodity contained or processed in the pots, in order for this signature to have been retained.

The stable carbon isotope values exhibit a wide range of absolute values (figure 2), which do not fall within the more isotopically depleted range usually expected for freshwater ecosystems. However, the incorporation of more enriched values can readily be explained by likely extensive processing of anadromous fish such as sturgeon, which were available in significant quantities, especially in the vicinity of whirlpools and rapids such as those near Schela Cladovei and Lepenski Vir [39,42,59]. Since most sturgeon species do not feed once they enter the freshwater river system for spawning, a marine stable carbon isotope signature would be anticipated in tissues from larger, older sturgeon, while juveniles, non-diadromous sturgeon (e.g. sterlet) and other non-migratory fish (e.g. carp) would display more isotopically depleted freshwater signatures. In between, tissues from anadromous species that continue to feed in freshwater locations such as Huso huso (beluga) would comprise a mixture of carbon sources, indeed, as reflected in bone collagen stable carbon isotope values of sturgeon bones from Vlasac [42]. The organic residues are an accumulation of lipids across multiple uses of the pot and so the δ^{13} C values will provide an integrated signature of these potentially different sources of carbon.

(b) Evidence for terrestrial animals in Neolithic pots

While cattle, sheep/goat and deer dominate in the Neolithic archaeozoological assemblages (see electronic supplementary

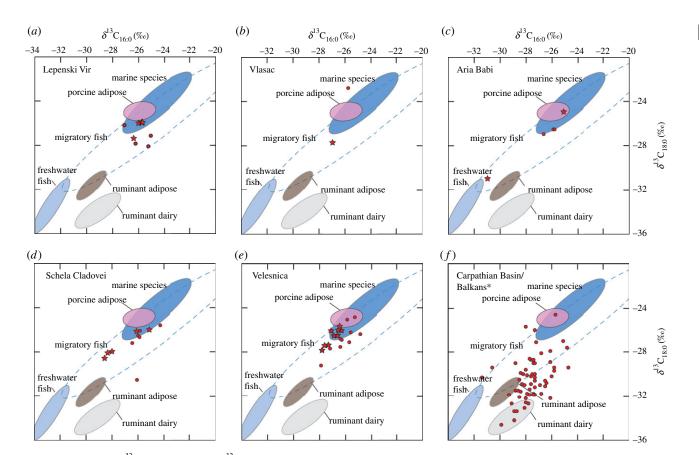


Figure 2. Scatter plot of $\delta^{13}C_{16:0}$ values against $\delta^{13}C_{18:0}$ values from Neolithic pottery residues from the Iron Gates region (a - e; n = 45) and the Carpathian Basin/northern and southern Balkans (f; n = 64; *Data from [6]). Datapoints shown as red stars indicate where APAAs and DHFAs of carbon chain length $\geq C_{20}$ were also observed in the residue. Coloured ellipses are 1σ confidence ellipses derived from modern reference datasets. Reference fats are from [12,46,50], with additional new freshwater and migratory fish data from the Severn Estuary, UK and the Irish Sea (eels and salmon). Terrestrial and freshwater modern fats have been corrected for the contribution of post-industrial carbon (+1.3‰ [51]).

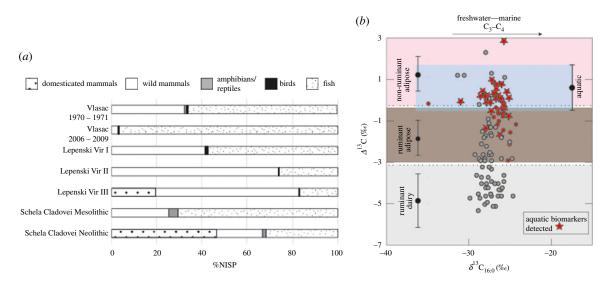


Figure 3. (*a*) Summary of archaeozoological assemblages from Vlasac [52,53], Lepenski Vir [34] and Schela Cladovei [33] based on NISP. Wild mammal data may include antler as this was not consistently quantified separately in the original reports. (*b*) Plot of $\delta^{13}C_{16:0}$ against $\Delta^{13}C$ ($\delta^{13}C_{18:0} - \delta^{13}C_{16:0}$) values, showing (red circles and stars) the data from the five sites investigated from the Iron Gates region (n = 45). By contrast, the grey circles show the stable isotope values from Neolithic pottery residues investigated from the northern and southern Balkans and Carpathian Basin, published in [6]; (n = 64). No aquatic biomarkers were detected in the lipid residues in pottery from the northern and southern Balkans. The ranges show the mean ± 1 s.d. from a database of reference values published in [49] (terrestrial fats); aquatic ranges as in figure 2.

material, table S3), fewer than half of the residues display a ruminant stable carbon isotope signature, despite the bias against the isotopic visibility of aquatic fats if products were mixed. The leanness of wild game compared with fish flesh and roe (fat comprising 1-2% of meat from modern wild deer [60,61], cf. sturgeon, catfish and carp meat in the region of 5% [62] and wild sturgeon caviar between 10 and 20% [63,64]) may have contributed to the unusually high

visibility of aquatic products here, but it is evident nonetheless that at these sites, ruminants could not have been the predominant resource processed in pottery. It is also notable that dairy (and by extension, unambiguous evidence for domesticated) products are almost entirely absent, represented by only a single residue from Schela Cladovei. In stark contrast with Starčevo pottery from the wider Balkans region, it is highly unlikely that dairying comprised a significant component of the activities taking place here, although the possibility of low quantities of dairy products being masked isotopically through mixing with very high quantities of non-ruminant fat cannot be ruled out.

In addition to ruminants, non-ruminant mammals, including pig and dog are also possible components of the economy of the Iron Gates region. Pig remains have been identified among archaeozoological assemblages in low quantities from Neolithic layers [33,34] as well as the dog from both Mesolithic and Early Neolithic assemblages [34,35,52,53,65]. While the presence of aquatic biomarkers shows that the non-ruminant stable isotope signatures from residues are associated with aquatic product processing, the additional contribution of terrestrial non-ruminants to this overall signature is quite plausible and cannot be further disentangled.

(c) The broader picture

Farming practices spread rapidly through the Balkan peninsula, probably along coastal plains and river valleys through the mountainous Balkan interior. The archaeology of the Iron Gates region is exceptional for the survival of evidence that spans this period, with abundant evidence of Late Mesolithic and Early Neolithic settlement. However, with the loss of coastal sites due to the Holocene marine transgression and relatively unexplored archaeology of the Lower Danube, it is possible that this was not an exceptional scenario in prehistory [66,67]. This surviving evidence enables closer examination of the likely diversity of Late Mesolithic and Early Neolithic practices along the Danube, the interactions that took place between peoples from different cultural backgrounds with the local environment, and their external contacts and trade networks.

The pottery residues, which date to the first half of the sixth millennium BC, demonstrate that Starčevo-Körös-Cris traditions of Early Neolithic pottery were being used for diverse purposes within the wider Balkan region. Along the banks of the Danube in the Iron Gates, pots are strongly associated with the processing of abundant riverine resources. By contrast, some 100 km away further north in the Balkans and on the Great Hungarian Plain, broadly contemporaneous Starčevo pots were being used predominantly for terrestrial resources, even at riverine locations [6,7], and in particular for processing dairy products. This aligns with local archaeozoological assemblages dominated by domesticated cattle, sheep/goat and deer. On a wider scale, organic residues from Early Neolithic pottery from Southeastern Europe, the Southern and Central Mediterranean and Northwest Europe display an emphasis upon terrestrial, ruminant-based commodities, including varying degrees of dairy products, even at coastal or lacustrine settlements (e.g. [1-15,68,69]; figure 4) and where there is otherwise evidence for fishing and shellfish exploitation, such as Northwest Anatolia [71].

Within the Iron Gates region, the presence of a wider range and higher abundance of fish remains in the Neolithic is now becoming recognized from more recent excavations and re-analyses of older material [33,39,42,72]. In addition to carp and catfish, the exploitation of sturgeon was an important and valued source of protein for prehistoric communities inhabiting the Iron Gates Mesolithic (e.g. [32,33]), and bones have also been identified among Neolithic faunal assemblages at Schela Cladovei [33,59]. Although this was initially thought to be unusual [24,33], re-analysis of faunal remains from contexts dated to 6300-5500 BC from Lepenski Vir has also identified Acipenseridae bones further upstream [72]. It is possible that misidentification and taphonomic bias against larger (i.e. older) sturgeon due to the re-absorption of bone minerals may both have led to under-estimation of the contribution of sturgeon [32,42,59,72]. The exceptional size of sturgeon (reaching several hundred kilograms), observed frequency and likely under-representation of their remains indicate that this and other aquatic species were probably an important source of food. It is possible that sturgeon capture was undertaken on a seasonal basis, as practised in more recent times [32,59], because sturgeon tend to swim upriver for spawning in spring/summer [73], but even if so, they may have been dried, smoked or otherwise preserved for year-round consumption.

The C and N stable isotope values obtained from human bone collagen from dated individuals broadly indicate that some humans were consuming a diet with an increased terrestrial component around the time of the appearance of pottery and livestock in the Iron Gates. The evidence nonetheless indicates a continued role for aquatic resources [39,40,42,43,74–77] and a higher degree of fish consumption compared with Early Neolithic sites further from the Danube or other large rivers such as the Tisza [30,78]. While the users of pottery in the Iron Gates may have had already some familiarity with fishing practices, the high visibility of aquatic organic residues is very unusual in farmer-type pottery and more closely akin to organic residues in pottery used by hunter-fisher-foragers around the Baltic Sea in the fifth to fourth millennia BC [15,16,70]. At these locations, between 20 and 70% of residues were attributed to a non-ruminant origin based on stable isotope compositions, and between 20 and 70% of total residues contained evidence for aquatic biomarkers. In these same regions, pottery types that have stronger affinities with early farming cultures contain residues that are also indicative of continued, but usually less-visible, aquatic resource processing. This is witnessed to some extent in Neolithic TRB pottery from the western Baltic [15], and to a greater extent from residues recovered from Globular Amphora Ware and Rzucewo pottery from Lithuania [70]. In these regions, the rich resources of coastal and lacustrine habitats and environments that were challenging for farming may have combined to result in a significant degree of wild resource exploitation alongside the presence of domesticates.

In the Iron Gates, the topography of the gorge would have placed restrictions upon farming. Localized pockets of fertile land suitable for cultivation or pasture would have been available, although the more open and moderate relief of the lower Danube, where the river is flanked by fertile alluvial terraces of Pleistocene and Holocene age, would have offered better opportunities for farming. Despite terrestrial animal fats being universally widespread in prehistoric

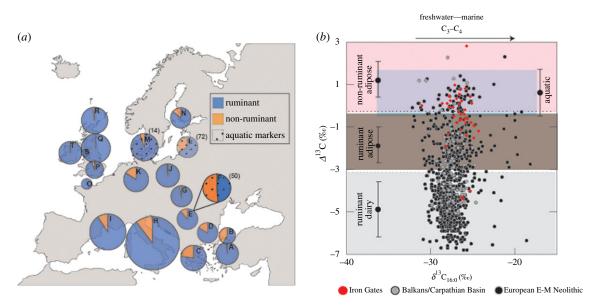


Figure 4. (*a*) Pie charts spanning distribution of sites from which organic residue analysis was undertaken, showing proportions of pottery lipids classified as ruminant (blue; $\Delta^{13}C < -0.3\%$) or non-ruminant (orange; $\Delta^{13}C \ge -0.3\%$). Numbers in brackets denote the percentage of sherds containing aquatic biomarkers, where applicable. A—northwest Anatolia, Neolithic (seventh mill. BC), n = 65 [1]; B—Yenikapı, Neolithic (mid-seventh to late sixth mill. BC), n = 12 [4]; C—northern Greece, Early – Middle Neolithic (mid-seventh to mid-sixth mill. BC), n = 100 [5]; D—southern Balkans, Karanovo I (early sixth mill. BC), n = 24 [6]; E—northern Balkans, Starčevo (late seventh to early sixth mill. BC), n = 18 [6]; F—Iron Gates, Starčevo and Criş (early sixth mill. BC), n = 45 (this paper); G—Carpathian Basin, Starčevo (early sixth mill. BC), n = 43 [8]; J—Poland, LBK (late sixth to early fifth mill. BC), n = 67 [9]; K—Germany, LBK (mid- to late sixth mill. BC), n = 25 [10]; L—Lithuania, Globular Amphora Culture and Rzucewo (late fourth to mid-third mill. BC), n = 18 [70]; M—southern Scandinavia, TRB (fourth mill. BC), n = 56 [15]; N—southern Finland, Corded Ware (third mill. BC), n = 7 [14]; O—Channel Isles, Cerny/Early Castellic, Le Pinade/VSG (fifth mill. BC), n = 8 [13]; P—southern Britain, Early Neolithic (fourth mill. BC), n = 165 [12]; Q—northern Britain, Carinated Bowl/Unstan/Early Grooved Ware (late fifth to fourth mill. BC), n = 73 [13]; R—Northern and Western Isles of Scotland, Hebridean/Unstan/Early Grooved Ware (mid-late fourth mill. BC), n = 106 [68]. Plot (b) Scatter plot of compound-specific isotope values of $\delta^{13}C_{16:0}$ values plotted against $\Delta^{13}C$ ($\delta^{13}C_{16:0} - \delta^{13}C_{16:0}$), showing data from this paper (red), the Balkans and Carpathian Basin [6] (grey) and Early –Middle Neolithic pottery from Europe (black; all other references above, except [4] not plotted). Ranges shown are as for figure 3. Total no. data points included = 904.

pottery, the pottery organic residues here do not show any greater indication of animal husbandry as a significant practice even in this downstream area. Among possible explanations for this pattern are that these pottery users had diets still drawing heavily on aquatic resources and only limited ruminant and dairy products, or pottery was being used here for a restricted purpose, with other resources prepared differently (e.g. spit roasting). A third possibility is that if the processing of a resource was frequently done close to the point of procurement, then this pattern may reflect the proximity of sampled contexts to the Danube banks which were not necessarily places where livestock was kept. A final possibility is that these sites were seasonally occupied, as may have been the case for some Neolithic sites in the Upper Gorge [24]. Despite the importance of fishing in the Iron Gates region over preceding millennia, the organic residues in pottery can be seen to represent a change in how fish was processed, with pots now being used for preparation, a technology that may have facilitated new activities such as making stews and soups or oil rendering.

To date, multiple scenarios of cultural interaction between foragers and farmers, as well as localized social transformations, have been proposed (e.g. [25,26,32,39,72,79–82]). While at sites such as Schela Cladovei, there is a hiatus between the Late Mesolithic occupation and re-occupation by groups displaying characteristically Neolithic traits [24], recent studies of both ancient DNA and strontium isotope signatures from human tooth enamel now provide evidence for the interaction of immigrant farmers with local foraging communities, including burial of non-locals in a 'Mesolithic' tradition [37,38,83,84], leading some authors to recognize a 'transformational' phase between the Late Mesolithic and Early Neolithic [26,72]. Our research has demonstrated the continued preparation of riverine resources in the Iron Gates, now using Neolithic pottery, which may relate to both the resource base encountered and cultural interactions occurring within pre-existing and newly formed social networks.

4. Conclusion

The Early Neolithic pottery residues from the Iron Gates region confirm interpretations from archaeozoological and stable isotope evidence that the use of aquatic resources continued into the early sixth millennium BC, echoing Late Mesolithic traditions, despite farming being practised in the wider region. The widespread use of Starčevo–Criş pottery in the Iron Gates for processing riverine resources is in marked contrast to its role for preparing ruminant meat and dairy products across the northern Balkans, Carpathian Basin, and indeed most Early and Middle Neolithic pottery from across wider Europe, even where fishing was probably practised. This suggests that the pottery was being used for a relatively specialized purpose at this location, whether it was being brought by farmers making use of the localized resources, incorporated into existing practices in the Iron Gates, or a combination of the two. Either way, it represents a new method for processing fish that was introduced within the Iron Gates and demonstrates a clear diversity in the function of this type of pottery within different cultural and ecological contexts.

Data accessibility. The datasets supporting this article have been uploaded as part of the electronic supplementary material.

Authors' contributions. L.J.E.C., H.L.W., M.R.-S. and J.E. carried out the laboratory work and participated in data analysis. L.J.E.C., M.I., D.U.-K., R.P.E. and C.B. participated in the conception and design of the study. L.J.E.C., M.I. and C.B. drafted the manuscript, while all others (A.B., D.B., J.E., R.P.E., S.P., M.R.-S., H.W., D.U.K.) provided interpretation, contextual information and critical revisions. All authors gave final approval for publication.

Competing interests. We have no competing interests.

Funding. This research was funded by a Natural Environment Research Council standard grant (L.J.E.C. and R.P.E.; NE/F021054/1) and an award of the German Research Council (M.I. and J.E.; DFG-IV101/ 5-1). M.R.-S. and R.P.E. are funded by a European Research Council Advanced Grant 'NeoMilk' (FP7-IDEAS-ERC/324202). The current excavations at Schela Cladovei have been supported by grants from the Carnegie Trust for the Universities of Scotland and the Munro Fund of the University of Edinburgh (C.B.). Fieldwork at the sites of Vlasac and Aria Babi was funded through grants received from the National Science Foundation of the USA (BCS-0442096; to T. D. Price and D.B.), British Academy (SG 40967 and 42170 and LRG 45589) and the McDonald Institute for Archaeological Research in Cambridge (to D.B.).

Acknowledgements. Compound-specific stable isotope analyses were performed using the Life Sciences Mass Spectrometry Facility (Bristol node) and we are grateful to Alison Kuhl and Ian Bull for technical assistance. We also thank Helen Grant of the NERC Life Sciences Mass Spectrometry Facility (Lancaster) for stable isotopic characterization of reference standards and derivatising agents.

References

- Evershed R *et al.* 2008 Earliest date for milk use in the Near East and southeastern Europe linked to cattle herding. *Nature* **455**, 528–531. (doi:10.1038/ nature07180)
- Nieuwenhuyse O, Roffet-Salque M, Evershed R, Akkermans P, Russell A. 2015 Tracing pottery use and the emergence of secondary product exploitation through lipid residue analysis at Late Neolithic Tell Sabi Abyad (Syria). *J. Archaeol. Sci.* 64, 54–66. (doi:10.1016/j.jas.2015.10.002)
- Özbal H, Thissen L, Doğan T, Gerritsen F, Özbal R, Türkekul Bıyık A. 2013 Neolitik Batı Anadolu ve Marmara Yerleşimleri Çanak Çömleklerinde Organik Kalıntı Analizleri. Arkeometri Sonuçları Toplantısı 28, 105–114.
- Özbal H, Thissen L, Doğan T, Gerritsen F, Özbal R, Türkekul Bıyık A. 2014 Yenikapı, Aşağıpınar, Bademağacı ve Barcın Çömleklerinde Organik Kalıntı Analizi. Arkeometri Sonuçları Toplantısı 29, 83–90.
- Whelton H, Roffet-Salque M, Kotsakis K, Urem-Kotsou D, Evershed R. 2017 Strong bias towards carcass product processing at neolithic settlements in northern Greece revealed through absorbed lipid residues of archaeological pottery. *Quat. Int.* 496, 127–139. (doi:10.1016/j.quaint.2017.12.018)
- Ethier J *et al.* 2017 Earliest expansion of animal husbandry beyond the Mediterranean zone in the 6th millennium BC. *Sci. Rep.* 7, 7146. (doi:10.1038/ s41598-017-07427-x)
- Craig O *et al.* 2005 Did the first farmers of central and eastern Europe produce dairy foods? *Antiquity* **79**, 882–894. (doi:10.1017/ S0003598X00115017)
- Debono Spiteri C *et al.* 2016 Regional asynchronicity in dairy production and processing in early farming communities of the northern Mediterranean. *Proc. Natl. Acad. Sci. USA* **113**, 13 594–13 599. (doi:10.1073/pnas.1607810113)
- Salque M *et al.* 2012 New insights into the Early Neolithic economy and management of animals in Southern and Central Europe revealed using lipid residue analyses of pottery vessels.

Anthropozoologica **47**, 45–61. (doi:10.5252/ az2012n2a4)

- Salque M *et al.* 2013 Earliest evidence for cheese making in the sixth millennium BC in northern Europe. *Nature* **493**, 522–525. (doi:10.1038/ nature11698)
- Roffet-Salque M, Evershed R. 2015 Shifting pottery use and animal management at Kopydłowo (Poland) traced though lipid residue analyses of pottery vessels. In *Kopydłowo, stanowisko 6. Osady neolityczne z pogranicza kujaw i wielkopolski* (eds A Marciniak, I Sobkowiak-Tabaka, M Bartkowiak, M Lisowski), pp. 133–142. Pękowice-Poznań, Wydawnictwo Profil-Archeo.
- Copley M *et al.* 2003 Direct chemical evidence for widespread dairying in Prehistoric Britain. *Proc. Natl. Acad. Sci. USA* **100**, 1524–1529. (doi:10.1073/pnas. 0335955100)
- Cramp L et al. 2014 Immediate replacement of fishing with dairying by the earliest farmers of the northeast Atlantic archipelagos. Proc. R. Soc. B 281, 20132372. (doi:10.1098/rspb.2013.2372)
- Cramp L *et al.* 2014 Neolithic dairy farming at the extreme of agriculture in northern Europe. *Proc. R. Soc. B* 281, 20140819. (doi:10.1098/rspb. 2014.0819)
- Craig OE *et al.* 2011 Ancient lipids reveal continuity in culinary practices across the transition to agriculture in Northern Europe. *Proc. Natl. Acad. Sci.* USA **108**, 17 910–17 915. (doi:10.1073/pnas. 1107202108)
- Mylona D. 2014 Aquatic animal resources in Prehistoric Aegean, Greece. J. Biol. Res. Thessal. 21, 2. (doi:10.1186/2241-5793-21-2)
- Çakırlar C. 2013 Rethinking neolithic subsistence at the gateway to Europe with new archaeozoological evidence from Istanbul. In *The environmental* archaeology of subsistence, specialisation and surplus food production (eds M Groot, D Lentjes, J Zeiler), pp. 59–79. Amsterdam, The Netherlands: Sidestone Press.
- 18. Richards M, Schulting R. 2006 Touch not the fish: the mesolithic-neolithic change of diet and its

significance. *Antiquity* **80**, 444–445. (doi:10.1017/ S0003598X00093765)

- Ivanova M, de Cupere B, Ethier J, Marinova E. 2018 Pioneer farming in Southeast Europe during the early sixth millennium BC: climate-related adaptations in the exploitation of plants and animals. *PLoS ONE* 13, e0197225. (doi:10.1371/ journal.pone.0197225)
- Scheu A. 2017 Neolithic animal domestication as seen from ancient DNA. *Quat. Int.* 496, 102–107. (doi:10.1016/j.quaint.2017.02.009)
- Srejović D. 1972 Europe's first monumental sculpture: new discoveries at Lepenski Vir. London, UK: Thames & Hudson.
- Srejović D, Letica Z. 1978 Vlasac. Mezolitsko naselje u Đerdapu (I arheologija). Beograd, Serbia: Srpska akademija nauka i umetnosti.
- Boroneanţ V. 1989 Thoughts on the chronological relations between the Epi-Palaeolithic and the Neolithic of the lower Danube. In *The Mesolithic in Europe. Papers presented at the third international symposium, Edinburgh 1985* (ed. C Bonsall), pp. 475–480. Edinburgh, UK: John Donald.
- Bonsall C. 2008 The Mesolithic of the Iron Gates. In Mesolithic Europe (eds G Bailey, P Spikins), pp. 238–278. Cambridge, UK: Cambridge University Press.
- 25. Radovanović I. 1996 *The Iron Gates Mesolithic*. Ann Arbor, MI: International Monographs in Prehistory (Archaeological Series 11).
- Roksandić M. 2000 Between foragers and farmers in the Iron Gates Gorge: physical anthropology perspective. Đerdap population in transition from Mesolithic to Neolithic. *Documenta Praehistorica* 27, 1–100.
- 27. Borić D. 1999 Places that created time in the Danube gorges and beyond, c. 9000-5500 BC. *Documenta Praehistorica* **26**, 41-70.
- Borić D. 2002 The Lepenski Vir conundrum: reinterpretation of the Mesolithic and Neolithic sequences in the Danube Gorges. *Antiquity* 76, 1026-1039. (doi:10.1017/S0003598X00091833)

- Vasić R. 2008 Velesnica and the Lepenski Vir culture. In *The Iron Gates in prehistory: new perspectives* (eds C Bonsall, V Boroneanţ, I Radovanović), pp. 39–54. Oxford, UK: BAR International Series 1893.
- Whittle A, Bartosiewicz L, Borić D, Pettitt P, Richards M. 2002 In the beginning: new radiocarbon dates for the Early Neolithic in northern Serbia and southeast Hungary. *Antaeus* 25, 64–117.
- Borić D, Dimitrijević V. 2007 When did the 'Neolithic package' reach Lepenski Vir? Radiometric and faunal evidence. *Documenta Praehistorica* 35, 53–72. (doi:10.4312/dp.34.5)
- Borić D. 2001 Mesolithic and Early Neolithic hunters and fishers in the Danube gorges. In From the Mesolithic to the Neolithic: proceedings of the International Archaeological Conference held in the Damjanich Museum of Szolnok, September 22–27, 1996 (eds R Kertész, J Makkay), pp. 101–124. Budapest, Hungary: Archaeolingua.
- 33. Bartosiewicz L, Boroneanţ V, Bonsall C, Stallibrass S. 2001. New data on the prehistoric fauna of the Iron Gates: a case study from Schela Cladovei, Romania. In From the Mesolithic to the Neolithic: Proceedings of the International Archaeological Conference held in the Damjanich Museum of Szolnok, September 22–27, 1996 (eds R Kertész, J Makkay), pp. 15–21. Budapest, Hungary: Archaeolingua.
- Bökönyi S. 1970 Animal remains from Lepenski Vir. Science 167, 1702–1704. (doi:10.1126/science.167. 3926.1702)
- Clason A. 1980 Padina and Starcevo: game, fish and cattle. *Palaeohistoria* 22, 142–173.
- Bartosiewicz L, Bonsall C. 2018 'Herd mentality'. In Social dimensions of food in the prehistory of the Eastern Balkans and neighbouring areas (eds M Ivanova, P Stockhammer, V Petrova, D Takorova, B Atanassov), pp. 1–13. Oxford, UK: Oxbow Books.
- Borić D, Price T. 2013 Strontium isotopes document greater human mobility at the start of the Balkan Neolithic. *Proc. Natl Acad. Sci. USA* **110**, 3298–3303. (doi:10.1073/pnas.1211474110)
- Mathieson I *et al.* 2018 The genomic history of Southeastern Europe. *Nature* 555, 197–203. (doi:10.1038/nature25778)
- Bonsall C *et al.* 1997 Mesolithic and early Neolithic in the Iron Gates: a palaeodietary perspective. *J. Euro. Archaeol.* 5, 50–92. (doi:10.1179/ 096576697800703575)
- Bonsall C *et al.* 2000 Stable isotopes, radiocarbon and the Mesolithic–Neolithic transition in the Iron Gates. *Documenta Praehistorica* 27, 119–132.
- Schulting R, Borić D. 2017 A tale of two processes of Neolithisation: South-east Europe and Britain/ Ireland. In *The Neolithic of Europe: papers in honour of Alasdair Whittle* (eds J Pollard, P Bickle, V Cummings, D Hofmann), pp. 82–105. Oxford, UK: Oxbow Books.
- Borić D, Grupe G, Peters J, Mikić Z. 2004 Is the Mesolithic-Neolithic subsistence dichotomy real? New stable isotope evidence from the Danube Gorges. *Eur. J. Archaeol.* 7, 221–248. (doi:10.1177/ 1461957104056500)

- Bonsall C *et al.* 2004 Radiocarbon and stable isotope evidence of dietary change from the Mesolithic to the Middle Ages in the Iron Gates: new results from Lepenski Vir. *Radiocarbon* 46, 293 – 300. (doi:10. 1017/S0033822200039606)
- Evershed R. 2008 Organic residue analysis in archaeology: the archaeological biomarker revolution. *Archaeometry* **50**, 895–924. (doi:10. 1111/j.1475-4754.2008.00446.x)
- Dudd S, Evershed R. 1998 Direct evidence of milk as an element of archaeological economies. *Science* 282, 1478–1481. (doi:10.1126/science.282.5393. 1478)
- Cramp L, Evershed R. 2014 Reconstructing aquatic resource exploitation in human prehistory using lipid biomarkers and stable isotopes. In *Treatise on geochemistry: archaeology and anthropology*, vol. 12, 2nd edn (eds H Holland, K Turekian), pp. 319–339. Oxford, UK: Elsevier.
- Correa-Ascencio M, Evershed R. 2014 High throughput screening of organic residues in archaeological potsherds using direct acidified methanol extraction. *Anal. Methods* 6, 1330–1340. (doi:10.1039/c3ay41678j)
- Rieley G. 1994 Derivatization of organic compounds prior to gas chromatographic – combustion – isotope ratio mass spectrometric analysis: identification of isotope fractionation processes. *Analyst* **119**, 915–919. (doi:10.1039/AN9941900915)
- Dunne J *et al.* 2012 First dairying in green Saharan Africa in the 5th millennium BC. *Nature* 486, 390–394. (doi:10.1038/nature11186)
- Outram A *et al.* 2009 The earliest horse harnessing and milking. *Science* **323**, 1332–1335. (doi:10. 1126/science.1168594)
- Friedli H, Lötscher H, Oeschger H, Siegenthaler U, Stauffer B. 1986 Ice core record of the ¹³C/¹²C ratio of atmospheric CO₂ in the past two centuries. *Nature* **324**, 237–238. (doi:10.1038/ 324237a0)
- Bökönyi S. 1978 The vertebrate fauna of Vlasac. In Vlasac: mezolitsko naselje u djerdapu, vol. 2 (ed. M Garašanin), pp. 35–65. Belgrade, Serbia: Srpska akademija nauka i umetnosti.
- Borić D *et al.* 2014 Late Mesolithic lifeways and deathways at Vlasac (Serbia). *J. Field Archaeol.* 39, 4–31. (doi:10.1179/0093469013Z.0000000070)
- Matikainen J *et al.* 2003 A study of 1,5 hydrogen shift and cyclisation reactions of isomerised methyl linoleate. *Tetrahedron Lett.* 59, 567–573. (doi:10. 1016/S0040-4020(02)01513-2)
- 55. Hansel F, Copley M, Madureira L, Evershed R. 2004 Thermally produced ω -(*o*-alkylphenyl)alkanoic acids provide evidence for the processing of marine products in archaeological pottery vessels. *Tetrahedron Lett.* **45**, 2999–3002. (doi:10.1016/j. tetlet.2004.01.111)
- Evershed R, Copley M, Dickson L, Hansel F. 2008 Experimental evidence for the processing of marine animal products and other commodities containing polyunsaturated fatty acids in pottery vessels. *Archaeometry* 50, 101–113. (doi:10.1111/j.1475-4754.2007.00368.x)

- Regert M, Bland H, Dudd S, van Bergen P, Evershed R. 1998 Free and bound fatty acid oxidation products in archaeological ceramic vessels. *Proc. R. Soc. Lond. B* 265, 2027–2032. (doi:10. 1098/rspb.1998.0536)
- Hansel F, Bull I, Evershed R. 2011 Gas chromatographic mass spectrometric detection of dihydroxy fatty acids preserved in the 'bound' phase of organic residues of archaeological pottery vessels. *Rapid Commun. Mass Spectrom.* 25, 1893–1898. (doi:10.1002/rcm.5038)
- Bartosiewicz L, Bonsall C, Şişu V. 2008 Sturgeon fishing in the middle and lower Danube region. In *The Iron Gates in prehistory: new perspectives* (eds C Bonsall, V Boroneanţ, I Radovanović), pp. 39–54. Oxford, UK: British Archaeological Reports International Series 1893.
- 60. Bleier N et al. 2013 Comparison of the fat and protein content in the red deer hind meat from confined and open areas. In 2nd International Symposium on Hunting. Modern aspects of sustainable management of game populations. Novi Sad, Serbia, 7th–12th October.
- Strazdiņa V, Jemeļjanovs A, Šterna V. 2013 Nutrition value of wild animal meat. *Proc. Latv. Acad. Sci. B Nat. Exact Appl. Sci.* 67, 373–337.
- Silva J, Chamul R. 2000 Composition of marine and fresh water finfish and shell fish species and their products. In *Marine and freshwater products handbook* (eds R Martin, E Carter, E Flick, L Davis), pp. 31–46. Lancaster, UK: CRC Press.
- Wirth M et al. 2002 Fatty acid composition in sturgeon caviar from different species: comparing wild and farmed origins. International Review of Hydrobiology 87(5-6), IV Chemical and Biochemical Composition of Sturgeon Products, 629-636.
- Ovissipour M, Rasco B. 2011 Fatty acid and amino acid profiles of domestic and wild Beluga (*Huso huso*) roe and impact on fertilization ratio. J. Aquac. Res. Dev. 2, 113. (doi:10.4172/2155-9546.1000113)
- Bökönyi S. 1975 Vlasac: an early site of dog domestication. In *Archaeozoological studies* (ed. A Clason), pp. 167–178. Amsterdam, The Netherlands: North Holland and American Elsevier.
- Gurova M, Bonsall C. 2014 'Pre-Neolithic' in Southeast Europe: a Bulgarian perspective. Documenta Praehistorica XLI, 95–110. (doi:10.4312/ dp.41.5)
- 67. Clarke D. 1976 *Mesolithic Europe: the economic basis*. London, UK: Duckworth
- Smyth J, Evershed R. 2015 The molecules of meals: new insight into Neolithic foodways. *Proc. R. Irish Acad. Sect. C* 115C, 27–46.
- Thissen L, Özbal H, Türkekul Bıyık A, Gerritsen F, Özbal R. 2010 The land of milk? Approaching dietary preferences of Late Neolithic communities in NW Anatolia. *Leiden J. Pottery Stud.* 26, 157–172.
- Heron C *et al.* 2015 Cooking fish and drinking milk? Patterns in pottery use in the southeastern Baltic, 3300–2400 cal BC. *J. Archaeol. Sci.* 63, 33–43. (doi:10.1016/j.jas.2015.08.002)
- 71. Boessneck J, von den Driesch A. 1979 Die Tierknochenfunde aus der neolithischen Siedlung auf

dem Fikirtepe bei Kadiköy am Marmarameer. Munich, Germany: Institut für Palaeoanatomie, Domestikationsforschung und Geschichte der Tiermedizin der Universität München.

- Borić D, Dimitrijević V. 2005 Continuity of foraging strategies in Mesolithic – Neolithic transformations: dating faunal patterns at Lepenski Vir (Serbia). Atti della Società per la preistoria e protostoria della regione Friuli-Venezia Giulia 35, 33–107.
- Suciu R. 2008 Sturgeons of the NW Black Sea and lower Danube river countries. Mexico: NDF Workshop case studies WG 8, case study 5.
- Bonsall C *et al.* 2015 New AMS dates for human remains from Stone Age sites in the Iron Gates reach of the Danube, Southeast Europe. *Radiocarbon* 57, 33–46. (doi:10.2458/azu_rc.57.18188)
- Grupe G, Mikić Z, Peters J, Manhart H. 2003 Vertebrate food webs and subsistence strategies of Meso- and Neolithic populations of Central Europe.

In *Decyphering ancient bone* (ed. J Peters), pp. 193–221. Rahden, Germany: Leidorf.

- Borić D, Miracle P. 2004 Mesolithic and Neolithic (dis)continuities in the Danube Gorges: new AMS dates from Padina and Hajdučka Vodenica (Serbia). *Oxf. J. Archaeol.* 24, 341–371. (doi:10.1111/j.1468-0092.2004.00215.x)
- Nehlich O, Borić D, Stefanović S, Richards M. 2010 Sulphur isotope evidence for freshwater fish consumption: a case study from the Danube Gorges, SE Europe. *J. Archaeol. Sci.* 37, 1131–1139. (doi:10. 1016/j.jas.2009.12.013)
- Gamarra B *et al.* 2018 5000 years of dietary variations of prehistoric farmers in the Great Hungarian Plain. *PLoS ONE* 13, e0197214. (doi:10. 1371/journal.pone.0197214)
- 79. Voytek B, Tringham R. 1989 Rethinking the Mesolithic: the case of South-east Europe. In *The Mesolithic in Europe: papers presented at the third international symposium, Edinburgh 1985* (ed. C

Bonsall), pp. 492–500. Edinburgh, UK: John Donald.

- Chapman J. 2000 Fragmentation in archaeology. People, places and broken objects in the prehistory of south-Eastern Europe. London, UK: Routledge.
- Tringham R. 2000 South-eastern Europe in the transition to agriculture in Europe: bridge, buffer or mosaic. In *Europe's first farmers* (ed. T Price), pp. 19–56. Cambridge, UK: Cambridge University Press.
- Radovanović I, Voytek B. 1997 Hunters, fishers or farmers: sedentism, subsistence and social complexity in the Djerdap Mesolithic. *Analecta Praehistorica Leidensia* 29, 19–31.
- Borić D. 2016 Deathways at Lepenski Vir: patterns in mortuary practice. Belgrade, Serbia: Serbian Archaeological Society.
- Bonsall C, Boroneanţ A. 2018 The Iron Gates Mesolithic: a brief review of recent developments. *L'anthropologie* **122**, 264–280. (doi:10.1016/j. anthro.2018.03.003)

9