Vegetation History and Archaeobotany
https://doi.org/10.1007/s00334-026-01108-4

ORIGINAL ARTICLE

Late Neolithic barley cultivation practices at pile-dwelling sites in Slovenia

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Received: 30 December 2025 / Accepted: 14 April 2026

Abstract

Today, the area of Ljubljansko barje is used as farmland, but in the past it was covered by peat, and before that by a lake. During that time, the area was settled by pile-dwelling communities (ca. 4700–2700 BCE). Archaeobotanical crop finds and crop processing by-products point to a well-established farming society. While the pile-dwelling settlements in Ljubljansko barje are well known and have been extensively researched, the agricultural practices associated with these sites in Slovenia have received almost no research attention. We address these research gaps by carrying out stable carbon (δ^{13}C) and nitrogen (δ^{15}N) isotope analyses of barley (*Hordeum vulgare*) grains from two 4th-millennium BCE pile-dwelling sites. The mean δ^{15}N values of charred barley grains were ca. 3‰ higher than the range for unmanaged vegetation, estimated from the δ^{15}N values of bone collagen of wild herbivores. This indicates cultivation of barley in soils enriched in ^{15}N, possibly due to moderate application of manure/midden material. While barley grain δ^{13}C values differed between sites, potentially due to different environmental settings of the cultivated plots, the similarity in barley grain δ^{15}N values through time points to consistency in agricultural practice throughout the sites' occupations. Our study presents the first stable isotope analysis of plant remains from Slovenia, and the findings provide the first step toward understanding husbandry practices among early farmers in Ljubljansko barje.

Keywords Stable isotopes · Manuring · Ljubljansko barje · δ^{15}N values · δ^{13}C values · *Hordeum vulgare*

Introduction

Ljubljansko barje is a tectonic depression on the southern end of the Ljubljana basin. It is a cultural landscape with one of the largest complexes of wetland meadows in Slovenia (Zorn and Šmid Hribar 2012). In the past, the area was covered by a shallow lake, which over time transformed into a *Sphagnum*-dominated peatland. In the second half of the 18th century, the peatland began to be drained in order to obtain agricultural land. Today, a large part of Ljubljansko barje consists of a cultivated agricultural landscape (Perko and Orožen Adamič 2001).

Communicated by O. Brinkkemper

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Pile-dwelling settlements in Ljubljansko barje (Fig. 1) date to ca. 4700–2700 BCE, (Velušček et al. 2023; Velušček and Čufar 2014), corresponding to the Late Neolithic. Palynological studies of the Late Neolithic environment in Ljubljansko barje indicate some instances of fire use and landscape opening, suggesting small-scale forest clearance associated with agricultural and grazing activities (Andrič 2020; Andrič et al. 2008). The oldest finds of cultivated plants from Ljubljansko barje date to ca. 4000 cal. BCE (Tolar 2018), but despite numerous remains of crops, weeds, and edible wild plants having been recovered (Tolar et al. 2011), very little research has been done on the agricultural practices of these early farmers. It is generally assumed that husbandry and farming practices in Ljubljansko barje followed similar patterns to those observed at other contemporary sites in Europe (Andrič 2020; Budja and Mlekuž 2008; Tolar et al. 2011).

Across Europe, several studies have investigated Neolithic agricultural practices (Antolín et al. 2015; Bogaard 2004; Jakobitsch et al. 2022; Sabanov et al. 2024). Research using stable isotope analysis of cereal grains and pulses has

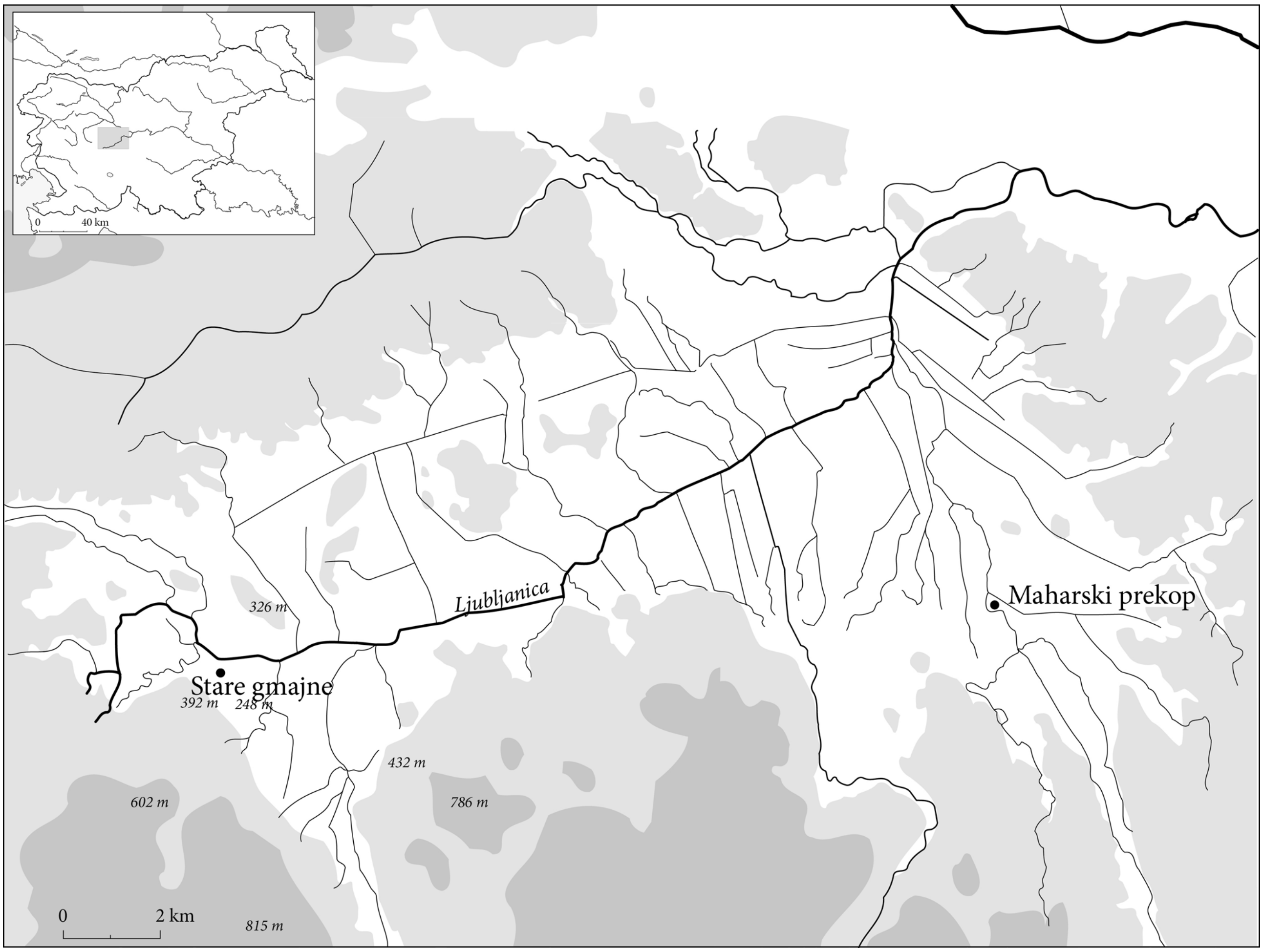

Fig. 1 Map of Ljubljansko barje with the two pile-dwelling sites, Maharski prekop (MP) and Stare gmajne (SG) marked. Today, most of the area is farmland, covered with meadows and fields and crossed by drainage ditches. Different shades of grey represent elevation. (From Tolar et al. 2011; Fig. 1b)

frequently identified the use of manure, indicating that Neolithic farmers made significant investments in their fields (Araus et al. 2024; Bogaard et al. 2013; Gillis et al. 2020; Halvorsen et al. 2023). However, the extent and intensity of manuring appears to have varied, possibly depending on livestock availability (Halvorsen et al. 2023). Studies of cultivation practices suggest that fire use contributed to landscape management, but this does not necessarily imply a "slash-and-burn" approach, whereby the location of cultivation plots moved frequently once initial soil fertility was exhausted. In most cases, weed seed and spatial analysis suggest that permanent cultivation was more likely, and crop yields could have been maintained by application of manure (Baum et al. 2016; Jacomet et al. 2016).

Evidence of cultivated crops, including (in order of ubiquity) *Hordeum vulgare*, *Triticum dicoccum*, *Triticum monococcum*, *Papaver somniferum*, *Linum usitatissimum*, and *Pisum sativum*, has been documented from pile-dwelling settlements in Slovenia (Velušček et al. 2018, 2023; Tolar 2018; Velušček 2004; Tolar et al. 2010; Jeraj 2002; Culiberg et al. 1991). The remains of wild-growing uncultivated plant species indicate wet to intermediately wet environments in the vicinity of the sites (e.g. woody *Alnus* and *Salix* sp., and herbaceous species of forest clearings, wetland and ruderal environments) (Tolar et al. 2011).

Current understanding of the past environmental and geological processes, including hydrology, that shaped Ljubljansko barje remains highly fragmented. Many aspects of its development are poorly understood, and while it is generally accepted that the area was wet, the environment was probably heterogeneous rather than uniform. Limited documentation of these local differences complicates interpretations of the pile-settlement dwellers' living conditions (Verbič 2011). Stable isotope analysis of archaeobotanical remains can offer insight into differences in environmental growing conditions between archaeological settlements, crop species, and potentially identify manuring practices.

Reconstructing past farming practices through stable isotope analysis of crop remains

Nitrogen isotope ($\delta^{15}N$) values of plants generally reflect the $\delta^{15}N$ value of the soil in which they grow. Plant $\delta^{15}N$ values can be influenced by a number of natural and anthropogenic processes, such as waterlogging, burning, and manuring with animal dung. In these cases there is preferential loss of the lighter isotope, ^{14}N, leaving the material (and soil) enriched in ^{15}N and therefore with higher $\delta^{15}N$ values (see Styring et al. 2017 for more detailed description). Soil properties, such as organic matter content, may also moderate the isotopic effect of soil amendments (Larsson et al. 2024).

Modern field experiments across Europe have found that cereal grains that have not been manured for the last 3 + years have $\delta^{15}N$ values lower than 3‰, cereal grains manured annually or biennially at lower levels (< 20 tonnes manure per hectare) have $\delta^{15}N$ values between 3 and 6‰, and cereal grains manured annually at high levels (30 + tonnes per hectare) have $\delta^{15}N$ values over 6‰ (Bogaard et al. 2013). A more recent analysis of this modern dataset has found that cereal grains with $\delta^{15}N$ values within c. 0.5‰ of the threshold $\delta^{15}N$ values of 3 and 6‰ should be interpreted cautiously as having received low/medium or medium/high levels of manure, respectively (Ben Makhad et al. 2024).

The stable carbon isotope ($\delta^{13}C$) composition of plants is affected by stomatal conductance, which is influenced by water availability and photosynthetic activity (e.g., Farquhar et al. 1989). Periods of limited water availability lead to less negative $\delta^{13}C$ values (Farquhar et al. 1989), whereas more negative cereal grain $\delta^{13}C$ values can be a result of receiving higher levels of water (either from natural sources of precipitation or anthropogenic watering) and/or growing in soils with better water retention. In temperate regions like Slovenia, however, water is unlikely to be a major limiting factor for growth, so interpretation of cereal grain $\delta^{13}C$ values in terms of water availability is not straightforward. More negative plant $\delta^{13}C$ values can also be associated with growing in lower light levels (Ehleringer et al. 1986).

In order to interpret crop isotope values in terms of agricultural management, rather than natural environmental factors, it is useful to estimate the isotope values of unmanaged plants for the archaeological site/region in question. The approach taken by previous studies (e.g., Styring et al. 2017) is to determine the isotope values of wild herbivore bones found at the same archaeological sites and subtract the mean offsets between herbivore bone collagen and diet from the $\delta^{13}C$ and $\delta^{15}N$ values of preserved herbivore bone collagen.

This study aims to investigate the growing conditions of barley cultivated by the inhabitants of two pile-dwelling sites in Ljubljansko barje, by determining the $\delta^{13}C$ and $\delta^{15}N$ values of charred barley grains recovered from the sites. This cultivated species is present on most of the pile-dwelling sites in Ljubljansko barje and was therefore an important crop species. If we find evidence for manuring, this will imply that pile-settlement dwellers invested significant resources (time, energy, materials) in cultivating and maintaining the fertility of permanent fields (as in other LN settlements in Europe (Dreslerová et al. 2021; Gillis et al. 2020)). Variability in barley grain isotope values could indicate that fields were situated in different locations, possibly to exploit different environmental conditions and to avoid risks of crop losses (due to e.g. flooding or droughts).

Materials and methods

Study area

The barley grains in this study come from the pile-dwelling sites of Maharski prekop (MP) and Stare gmajne (SG) (Fig. 1), which have both been excavated multiple times and dated using dendrochronology (Čufar et al. 2010). MP, dated to 3489 ± 10 cal BCE, is the older site and palynological and archaeobotanical samples were collected during the 2005 excavations (Tolar 2018; Andrič 2020). SG has at least two settlement phases (Velušček 2009). Archaeological remains, including wood and bone, were recovered at SG after being accidentally unearthed during regular deepening of modern-day drainage ditches. In addition, two 15 m^2 trenches were excavated. Excavations in 2006 uncovered a single archaeological layer (SG6), dated to 3138 ± 10 cal BCE, while excavations in 2007 identified two phases: an older phase (SG8), dated to 3332 ± 10 cal BCE, and a younger phase (SG7), dated to 3138 ± 10 cal BCE. These dates indicate a hiatus in occupation of the settlement of ca. 170 years. A site plan is provided in SI 1 (Skrt Kristan et al. 2026).

Sample selection

We collected charred grains of six-rowed naked barley (*Hordeum vulgare* var. *nudum*) from the two pile-dwelling sites MP and SG, from four archaeological contexts: MP, SG6, SG7, SG8 (Table 1). Barley was the only cereal with numerous grains, while other cereals (like emmer wheat) were mostly found in the form of chaff. Of the 4 contexts, SG6 was the richest in barley grains, with more than 10,000 grains recovered (unpublished data). For the SG7, SG8 and MP contexts, approximately 100 grains were recovered from each (unpublished data). For each context, at least 20 grains with minimal external distortion were selected for isotopic analysis. After examination of their cross-sections (see Sample preparation and isotopic analysis section),

Table 1 Information about sites and samples: single barley grains were collected from 4 archaeological contexts (MP, SG6, SG7 and SG8), from the two pile-dwelling sites

Site	Context	Context date	Species	No. of analysed barley grains/bones
SG	SG6	3138 ± 10 cal BCE	Barley	9
	SG7	3160 ± 10 cal BCE	Barley	17
	SG8	3332 ± 10 cal BCE	Barley	7
	SG	3200 ± 100 BCE*	Red deer, roe deer	10
MP	MP	3489 ± 10 cal BCE	Barley	9

Dating of sites from Čufar et al. 2010. Bones were collected across the whole of SG, rather than one of the specific contexts. *Estimate, based on the duration of settlement of the site

some of the grains had to be discarded because they did not appear to be well-preserved (Stroud et al. 2023). The number of analysed grains was therefore smaller than initially planned (Table 1).

We collected ten bones from wild herbivores—red deer (*Cervus elaphus*) and roe deer (*Capreolus capreolus*)—recovered from SG, to estimate the δ^{15}N values of unmanaged plants during occupation of the settlement. As it was not always possible to collect the same skeletal element from each species, bones were selected from areas of the site that were as far apart as possible, to decrease the possibility of them having belonged to the same individual. More information on the bone samples and their spatial placement can be found in SI 1 (Skrt Kristan et al. 2026).

Preparation and isotopic analysis of barley grains

All grains selected (with minimal external distortion) were weighed and photographed in dorsal and lateral views using a Leica stereo microscope with a Lumenera infinity 3–6 UR digital camera, then cross-sectioned at right angles to the ventral groove and the cross-sections photographed. Photographs of all the grains are in SI 2 (Skrt Kristan et al. 2026). Only well-preserved grains, resembling modern experimentally charred grains heated between 230 and 260 °C (Stroud et al. 2023; Fig. 5), were selected for isotopic analysis. We selected 3 grains per context to check for contamination of samples using Fourier Transform Infrared Spectroscopy with Attenuated Total Reflectance (FTIR-ATR) using an Agilent Technologies (Stockport, UK) Cary 640 FTIR instrument with a GladiATR™ accessory from PIKE Technologies (Madison, WI, USA). The spectra were acquired, the background noise subtracted, and a baseline correction was carried out using Agilent Resolution Pro to assist in the detection of contamination by carbonates, nitrates and humics following Brinkkemper et al. 2018; Vaiglova et al. 2014. The same grains were also used for isotopic analysis. As there was no contamination detected (see SI 3 Skrt Kristan et al. 2026), the grains were subsequently rinsed with distilled water, then freeze-dried. Each grain was crushed and analysed separately. Approximately 2.5 mg of each sample was weighed into tin capsules for carbon and nitrogen isotope analysis on a precisION isotope ratio mass spectrometer coupled to a vario PYRO cube high temperature elemental analyser at the Research Laboratory for Archaeology and the History of Art, University of Oxford, UK. Information on calibration, measurement uncertainty, and isotopic data of samples and reference materials are in SI 4 and SI 5 (Skrt Kristan et al. 2026).

Preparation and isotopic analysis of bone collagen

Approximately 0.5 g of bone was cleaned of any visible dirt or carbonate crusts using an aluminium oxide air abrasive and roughly crushed using a percussion mortar and pestle. Collagen was isolated using a modified 'gelatinisation method' based on the methods of Longin (1971). Briefly, bones were demineralised in 0.5 M hydrochloric acid (HCl) at 4 °C for approx. 14 days until soft. The acid was decanted and the samples rinsed to neutrality in deionised water before being heated in acidic water (pH 3) at around 70 °C for 48 h. The solution was filtered using Ezee Filters, the liquid then frozen, and then freeze-dried for 48 h. Approximately 1 mg of each bone collagen isolate was weighed into tin capsules for carbon and nitrogen isotope analysis on a Sercon 20–22 isotope ratio mass spectrometer coupled to a Sercon GSL elemental analyser at the Research Laboratory for Archaeology and the History of Art, University of Oxford, UK. Information on calibration, measurement uncertainty, and isotopic data of samples and reference materials are in SI 4 and SI 5 (Skrt et al. 2026).

Data analysis

Statistical analyses were carried out using statistical programming language R (4.4.1) (R Core Team, 2023). To account for the effect of charring, 0.31‰ was subtracted from the measured δ^{15}N values and 0.11‰ from the measured δ^{13}C values of charred barley grains following Nitsch et al. (2015). The mean offset between herbivore bone collagen δ^{15}N values and the δ^{15}N value of their plant diet has been found to be c. 4‰ (Steele and Daniel 1978) and the offset between herbivore bone collagen δ^{13}C values and the δ^{13}C value of their plant diet has been found to be c. 4.8‰ (Fernandes et al. 2012). We therefore subtracted 4‰ and 4.8‰, respectively, from the measured δ^{15}N and δ^{13}C values of the wild herbivore bone collagen to estimate the isotopic values of unmanaged vegetation. Following the approach of Styring et al. (2017), we calculated the range of estimated unmanaged (i.e., unmanured) cereal grain δ^{15}N values from

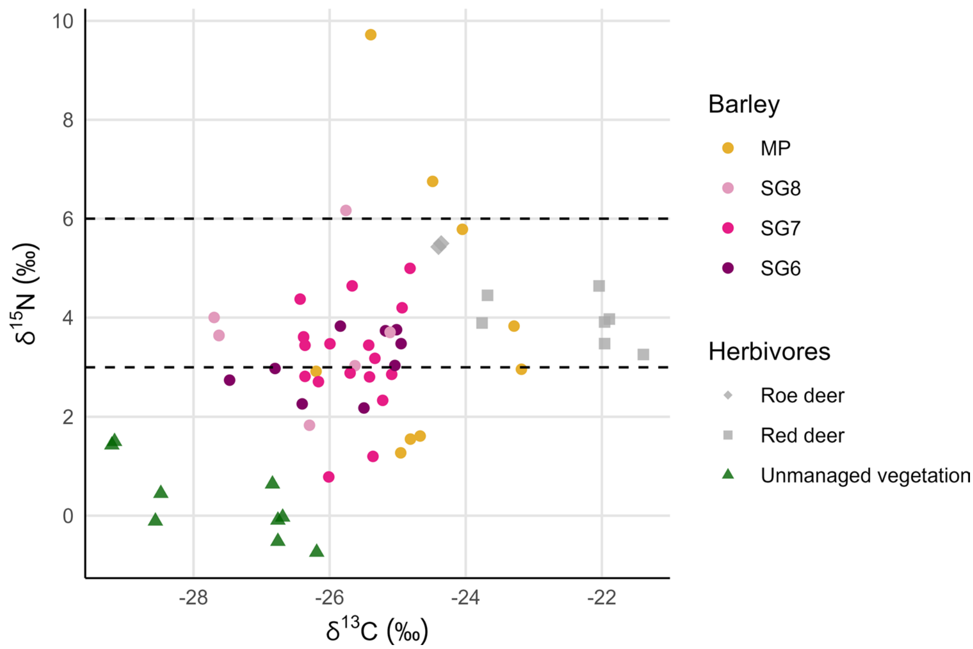

Fig. 2 δ^{12}C and δ^{15}N values of single charred barley grains from three contexts from SG and one context from MP, and wild herbivore bone collagen from SG. Barley grains are shown as coloured circles by context, while herbivore samples are distinguished by symbol according to species. Estimated unmanaged vegetation δ^{15}N and δ^{12}C values are inferred from herbivore collagen using trophic offsets of − 4‰ and − 4.8‰, respectively. Dashed horizontal lines at $\delta^{15}N$ = 3‰ and 6‰ indicate medium and high manuring intensity thresholds established from modern experimental plots (Bogaard et al. 2013)

Table 2 Summary of crop and wild herbivore bone collagen isotope results

Archaeological context	Species	n	δ^{13}C mean	δ^{13}C range	Δ^{13}C mean	Δ^{13}C range	$\delta^{15}N$ mean	δ^{15}N range
SG6	Barley	9	− 25.8	2.5	20.0	2.6	3.1	1.7
SG7	Barley	17	− 25.7	1.6	19.9	1.7	3.2	4.2
SG8	Barley	6	− 26.4	2.6	20.6	2.7	3.7	4.3
MP	Barley	9	− 24.6	3.0	18.7	3.2	4.0	8.5
SG	Red deer, roe deer	9	− 22.8	3.0	16.9	3.1	4.3	2.2
SG	Estimated unmanaged vegetation	9	− 27.6	3.0			0.3	2.2

The δ^{13}C, Δ^{13}C, and δ^{15}N (‰) values of barley are adjusted for the effect of charring

the mean wild herbivore diet δ^{15}N value ± 1 standard deviation (SD) (blue zone shading in Fig. 3). The measured δ^{13}C values of charred barley grains were normalised relative to fluctuations in the δ^{13}C value of atmospheric carbon dioxide to give Δ^{13}C values following Farquhar et al. (1982; as quoted in Wallace et al. 2013), utilising the barley grain δ^{13}C values and interpolations of the AIRCO2_LOESS system provided in Ferrio et al. (2005).

Results

Isotope values with corresponding metadata for the individual barley grains and wild herbivore bone collagen can be found in SI 5 and in Skrt Kristan et al. (2026). Key descriptive statistics for barley grains and wild herbivore bone collagen from the archaeological contexts are summarised in Table 2.

The determined δ^{15}N values of barley grains have a mean δ^{15}N value ± SD of 3.7 ± 1.6‰. One sample ($\delta^{15}N$ > 12‰) was visually and statistically identified as an outlier. This single point substantially disrupted the distribution of δ^{15}N values within context SG8, causing the group to violate normality assumptions (Shapiro-Wilk p = 0.025). After excluding the sample, the group distribution met normality (p = 0.575). The sample was excluded from analysis but is retained in supplementary data (ESM5). The determined barley grains have a mean δ^{13}C value of − 25.6 ± 1.0‰ and a mean Δ^{13}C value of 19.7 ± 1.0‰.

There is no correlation between %C or %N and isotope values, nor between the atomic C:N ratio of grains and isotope values (SI 6 Skrt Kristan et al. 2026), which means that there is no reason to suspect that patterning in the isotope values is due to compositional differences among the barley grains or exogenous contamination.

Figure 2 shows the δ^{13}C and δ^{15}N values of charred barley grains, wild herbivore bone collagen, and estimated δ^{13}C and δ^{15}N values of unmanaged vegetation. Roe deer (n = 2) have higher δ^{15}N values and lower δ^{13}C values than red deer (n = 7). The mean δ^{15}N value of unmanaged vegetation is estimated to be c. 0‰, which is consistent with the “no manuring” band in Bogaard et al. 2013. The δ^{15}N values of barley grains are notably higher than the estimated value of unmanaged vegetation and are consistent with the

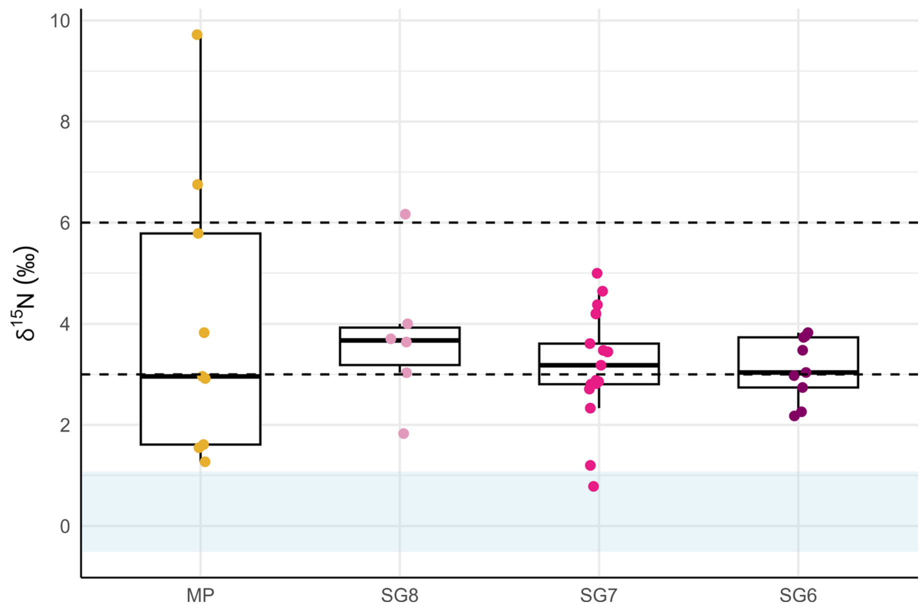

Fig. 3 Distribution of single carbonised barley grain $\delta^{15}N$ values from the three contexts from SG and one context from MP. Boxplots summarise the distribution of $\delta^{15}N$ values for each site, with individual grains shown as jittered points. The shaded blue band represents the estimated mean $\delta^{15}N$ value ± 1 SD of unmanaged vegetation, inferred from wild herbivore bone collagen $\delta^{15}N$ values

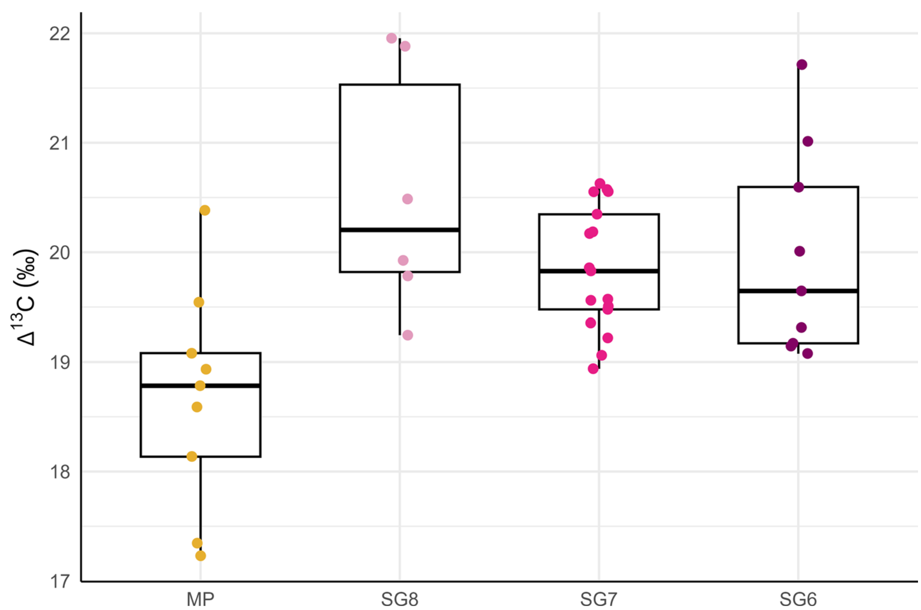

Fig. 4 Distribution of single carbonised barley grain $\Delta^{13}C$ values from the three contexts from SG and one context from MP. Boxplots summarise the distribution of $\Delta^{13}C$ values for each site, with individual grains shown as jittered points

low to medium manuring bands. Figure 3 shows the $\delta^{15}N$ values of barley grains from the three contexts from SG and one context from MP. The estimated $\delta^{15}N$ value range of unmanaged cereal grains is shaded in blue. The assumption of normality for $\delta^{15}N$ values in barley was met in all groups (Shapiro-Wilk $p > 0.17$), but Levene's test indicated significant heterogeneity of variances ($F = 3.22$, $p = 0.03$), with MP barley showing notably higher variability. A Kruskal–Wallis test revealed no significant differences in $\delta^{15}N$ values among archaeological contexts (χ^2 (3) = 1.12, $p = 0.77$).

Figure 4 shows the barley grain $\Delta^{13}C$ values for the 4 contexts. All groups met the assumptions for normality of $\Delta^{13}C$ values. Shapiro-Wilk tests indicated no significant deviation from normality ($p > 0.14$ for all groups), and Levene's test confirmed homogeneity of variances ($F = 1.21$, $p = 0.32$). A standard one-way ANOVA found a difference in $\Delta^{13}C$ between archaeological contexts (ANOVA: F (3.38) = 7.65, $p < 0.001$). Post-hoc comparisons revealed that $\Delta^{12}C$ values at MP were significantly lower than at SG6, SG7, and SG8 (Tukey HSD pairwise test: $p < 0.02$). The 95% confidence interval for MP [17.9–19.4] did not overlap with that of SG7 [19.6–20.1], and while slightly overlapping with that of SG6 [19.2–20.7] and SG8 [19.4–21.7], still had a lower mean $\Delta^{13}C$ value. There are no meaningful differences in $\Delta^{12}C$ values between the contexts of SG6, SG7 and SG8.

Discussion

The environmental setting of barley fields in Ljubljansko barje

The relatively low $\delta^{12}C$ and $\delta^{15}N$ values of wild herbivore bone collagen indicate that at least part of the area around the SG pile-dwelling site was a forested and nutrient-poor habitat. The wild herbivore isotope values are similar to the mean $\delta^{12}C$ and $\delta^{15}N$ values of red deer from Hornstaad-Hörnle IA (−23.7‰ and 4.9‰), a contemporaneous pile-dwelling site in southwest Germany (Styring et al. 2017). According to Sykut et al. (2021), $\delta^{12}C$ and $\delta^{15}N$ values in bone collagen of red deer vary with habitat characteristics and reflect landscape openness. The $\Delta^{12}C$ values of red and roe deer from SG (from 15.4‰ to 18.5‰) correspond with the higher range of $\Delta^{12}C$ values of modern red deer (corrected for the Suess effect) which corresponds with individuals from densely forested habitats. As part of the area of Ljubljansko barje is considered to be covered by a lake during this period (Velušček 2009), it is possible that the area was a heterogeneous and diverse landscape, with wetland and other open areas (fields, grazing areas) surrounded by densely forested hillsides. This would be consistent with palynological studies from Ljubljansko barje (Andrič 2020; Culiberg et al. 1991).

The barley grain carbon isotope values indicate possible differences in barley cultivation between MP and SG, but no notable difference among the 3 SG contexts. Barley from MP could have been grown on drier soils and/or in a more open landscape, with barley from SG growing in wetter soils and/or more shaded conditions. Unmanaged plant $\delta^{13}C$ values are lower than the barley values, but this could be related to the fact that leaves tend to be depleted in ^{13}C relative to non-photosynthetic tissues, typically by 2 to 4% (Hobbie and Werner 2004).

The two settlements were potentially situated in slightly different environments: MP was possibly situated close to a river (Budja and Mlekuž 2008), while SG appears to have been located by a shallow lake (Velušček 2009; Andrič 2020). This might have had an impact on soil conditions or vegetation density in the vicinity of the river, leading to more open areas for fields or less water retentive soils compared to the lake shore.

Late Neolithic soil enrichment practices at Ljubljansko barje

A variety of conditions, most notably burning, waterlogging and manuring, can increase $\delta^{15}N$ values in the soil and consequently in plants. In our study, the barley grain $\delta^{15}N$ values are consistently higher than the estimated $\delta^{15}N$ value of unmanaged vegetation, suggesting that barley in the Late Neolithic was cultivated in plots that were ^{15}N-enriched compared to uncultivated soils.

A potential cause of ^{15}N-enrichment is burning of vegetation to clear land for cultivation, but this has been found to result in cereal grain $\delta^{15}N$ values that are still close to 0‰ (Styring et al. 2017) and slash-and-burn cultivation is therefore unlikely to have resulted in the higher $\delta^{15}N$ values of > 3‰ here. Among the recovered plant remains a significant number of annual (weed) species—in high abundances—was found, also speaking in favour of permanent field cultivation in Ljubljansko barje (Bogaard 2004; Tolar et al. 2010, 2011). The presence of certain species associated with forest edges (*Physalis alkekengi*, *Eupatorium cannabinum*) (Tolar et al. 2010, 2011) suggests proximity of forested areas to the fields and indicates the interplay of open and closed vegetation patches. People from pile-dwellings possibly burned parts of the forest to gain clearings for permanent fields and pastures, then maintained the same fields for multiple seasons. Future functional weed ecological analysis of recovered remains could help to refine the interpretation of stable isotope values by reconstructing the productivity and level of disturbance of cultivated plots (cf. Styring et al. 2017; Diffey et al. 2020).

Another potential cause of elevated crop $\delta^{15}N$ values is cultivation on soils that experience wetting and drying cycles, such as a floodplain. The archaeobotanical remains of natural and synanthropic species from the two sites *(Solanum nigrum*, *Anagallis arvensis)* indicate a wet environment. and one of the weedy species recovered from SG (*Capsella* sp.) (Tolar et al. 2011) can be found on occasionally flooded soils. However, the area of Ljubljansko barje was famously covered by peat before the intense agro-improvements in the 18th century (Perko and Orožen Adamič 2001). The presence of peat bog indicates an oligotrophic environment, arguing against the influx of nutrients with flooding waters (Rydin and Jeglum 2013). Moreover, the low $\delta^{15}N$ values of unmanaged vegetation indicate that there were areas around the sites whose soils were not ^{15}N-enriched as a result of waterlogging and so there was likely the potential to cultivate similar areas for barley.

The similar $\delta^{15}N$ values but different $\delta^{13}C$ values of barley from MP and SG also suggest that high water retention of the soil is unlikely to be the only factor causing the relatively high $\delta^{15}N$ values of barley at both sites. It is therefore probable that the relatively high barley grain $\delta^{15}N$ values compared to unmanaged vegetation are due to addition of organic matter, such as manure/midden material, to cultivated plots in order to improve soil fertility and tractability. This has been argued to be the case at other Late Neolithic pile-dwelling sites in southwest Germany (Hornstaad-Hörnle IA and Sipplingen), supported by soil surveys and weed seed assemblages that discount the possibility of cultivating plots that would have experienced wetting and drying episodes (Styring et al. 2016, 2017). The mean δ^{13}C and δ^{15}N values of barley from these Late Neolithic sites in Slovenia are similar to the mean δ^{13}C (− 26.3‰) and δ^{15}N (4.3‰) values of cereal grains analysed from south-west Germany (Styring et al. 2016, 2017), further indicating similarities in growing conditions and cultivation practices between the two regions.

If manure input was responsible for the barley grain δ^{15}N values in this study, it would be consistent with modern cereal grains grown with the moderate addition of manure, on the order of <20 tonnes manure per hectare annually or biennially (Bogaard et al. 2013). The level of manuring can depend on the amount of available manure, labour, distance to the plots, and animals providing the manure (Bogaard et al. 2013; Jones 2012; Styring et al. 2022). Remains of cattle and ovicaprids from both MP and SG (Toškan 2009; Toškan et al. 2020; Toškan, personal communication) and a wheel of an animal-drawn cart from SG (Velušček 2009) indicate that both a source of manure and a mode of transport were available. Dreslerová et al. (2021) proposed that frequent relocation of lacustrine settlements could present an alternative way of providing long-term soil fertility, by moving fields to the site of a former settlement they would gain soils rich in nutrients for cultivation. Such frequent relocations (10–20-year intervals) were observed at SG (Velušček 2009).

Consistency in cultivation practices

Although the δ^{13}C values of barley from MP are significantly higher than those from SG, indicating a difference in the degree of shading or water retention of cultivated plots between the two sites, there is no change in barley grain δ^{13}C values among the archaeological contexts at SG, even though they span roughly 200 years. This suggests that there was consistency in the choice of plot locations throughout the occupation of the site.

Increasing distance between the settlement and fields can result in plots located at a greater distance (>500 m) receiving a decreasing level of manure (Halstead 2014). The similarity in barley grain δ^{15}N values between the two sites and among the contexts at SG (Fig. 3) indicates that the soil conditions—in terms of nitrogen source and availability—for barley plots were similar between the sites and over time at SG, probably reflecting a consistent management regime of arable fields and possibly due to similar manuring practices. The two pile-dwelling sites were located relatively close to one another (c. 15 km apart) and while MP was occupied slightly earlier than SG, the inhabitants are considered to have been part of the same cultural group (Velušček 2009). Therefore, it is perhaps unsurprising that there is no conspicuous change in barley growing conditions or management among the contexts studied. It is hard to draw concrete conclusions from relatively low numbers of barley grains, but the variability in barley grain δ^{15}N values at MP is slightly greater than among the SG contexts, which could point to more variable soil conditions at this site. Alternatively, this higher degree of variability can indicate grains harvested from multiple locations or in different years (Styring et al. 2024).

Conclusions

The δ^{13}C and δ^{15}N values of barley grains and deer bone collagen from the Late Neolithic sites of Maharski prekop (MP) and Stare gmajne (SG) in the Ljubljansko barje, Slovenia, have provided new insights into the environmental setting and crop cultivation conditions at these sites. The varied deer δ^{13}C values point to a heterogeneous landscape of open areas and dense forest, consistent with pollen data from the sites, and are similar to those of contemporaneous deer from Late Neolithic pile-dwelling sites in south-west Germany. At both sites in Ljubljansko barje, the δ^{15}N values of barley grains are notably higher than those estimated for unmanaged vegetation, suggesting that barley was cultivated in soils enriched in ^{15}N, probably due to manure application. There is a similar degree of ^{15}N-enrichment at both sites and through time, indicating consistency in cultivation practices during their occupation. This indicates that pile-settlement dwellers from Ljubljansko barje manured fields they maintained over several seasons. A difference in barley grain δ^{13}C values between Maharski prekop and Stare gmajne is consistent with their situation in different hydrological environments, likely influencing the water retention of soils used for barley cultivation. As we currently have very little reliable information on the pile-dwellings site catchment area and no information on the location of arable fields, some of our speculation is based on studies of other areas and time periods, where crop isotope values have been found to be remarkably similar (Styring et al. 2017). Applying weed ecological analysis to plant remain assemblages (e.g., Stroud et al. 2024) could contribute to obtaining additional answers in the future.

Acknowledgements We wish to thank Elizabeth Stroud, Elena Leghissa and Borut Toškan for suggestions, help and materials. We acknowledge the Wellcome Institutional Strategic Support Fund (ISSF; Grant reference number 204826/Z/16/Z) and the John Fell Fund for funding the EA-IRMS at the Oxford Stable Isotope Laboratory. We wish to thank the two anonymous reviewers for their helpful comments.

Funding The study was supported by the Slovenian Research and Innovation Agency programmes and projects: MR-56877, P6-0064, P1-

0236, and BI-VB/25-27-005.

Data availability All supplemental material (SI 1 to SI 6) is available in the Zenodo repository at: https://doi.org/10.5281/zenodo.18940376.

Declarations

Conflict of interests The authors have no relevant financial or non-financial interests to disclose.

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