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PAST CLIMATE AND AGRICULTURAL WATER MANAGEMENT IN THE MEDITERRANEAN INFERRED FROM CARBON ISOTOPE DISCRIMINATION IN ARCHAEOLOGICAL PLANT REMAINS

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SUMMARY - Knowing more about the way early farmers reacted to environmental changes may add clues to understand current water issues. The study of carbon isotope discrimination ($\Delta^{13}\text{C}$) in archaeological plant remains can provide reliable information about water availability in ancient crops. In this paper, a such methodology is described, along with some case studies aimed at the reconstruction of ancient water management practices. Archaeological samples of faba bean, barley and wheat were analysed in order to assess possible changes in water status from Neolithic to present-day in the Mediterranean coast of the Iberian Peninsula, as well as in a Neolithic site from NW Syria. Moreover, by comparing Δ values across several archaeological sites, we found evidences of differential water management practices between wheat and barley, as well as between cereal and legume crops, resembling some traditional practices that are still performed in dry areas.

Key words: carbon isotope discrimination ($\Delta^{13}\text{C}$), water use efficiency, irrigation, cereals, legumes, archaeology, origins of agriculture.

INTRODUCTION

Improving our knowledge about the way early farmers reacted to environmental changes might help us to find out alternative methods to face current water issues. Up to now, the most common way to get insight into ancient water management methods is the study of archaeological structures related to water uptake or distribution. However, this approach is limited to relatively advanced societies, and does not give any information about the actual results of these management on crops performance. In most archaeological sites, plant remains (usually carbonised) are routinely recovered during excavation. In this work, we will discuss how the study of carbon stable isotopes on these remains can provide valuable information about climate and crop water status. From such kind of data, it is possible to get a picture of water availability and its management in the beginnings of agriculture, when any other data is not available.

BRIEF THEORY OF CARBON STABLE ISOTOPES IN PLANTS

There are two stable carbon isotopes (^{13}C and ^{12}C), which occur in the molar ratio of 1:99 in the atmosphere. Nowadays, $^{13}\text{C}/^{12}\text{C}$ ratios are usually determined by mass spectrometry and referred to the PeeDee Belemnite (PDB) standard, as carbon isotope composition ($\delta^{13}\text{C}$) values:

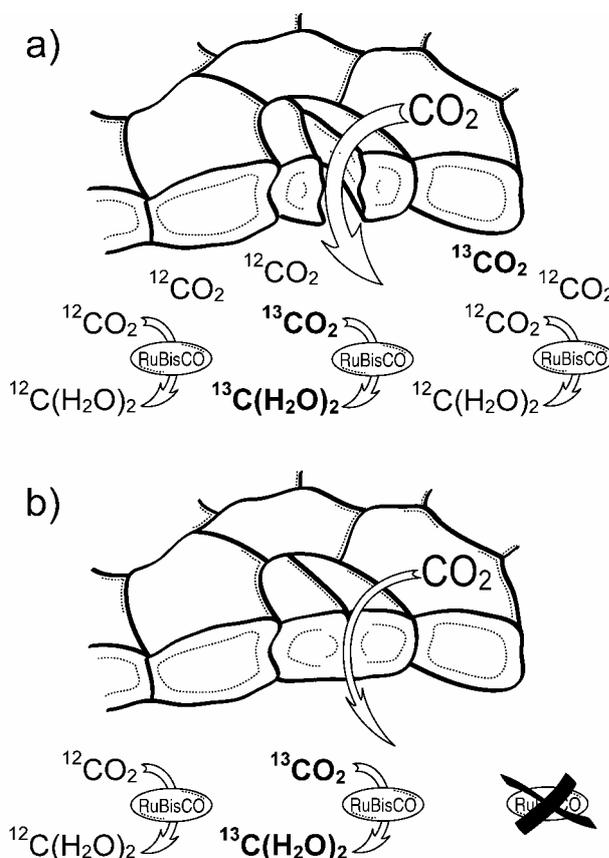
$$\delta^{13}\text{C}(\text{‰}) = \left[\left(\frac{R_{\text{sample}}}{R_{\text{standard}}} \right) - 1 \right] * 1000 \quad [1]$$

where R stands for the $^{13}\text{C}/^{12}\text{C}$ ratio. On this basis, present $\delta^{13}\text{C}$ in air CO_2 is about -8‰. However, plants with C_3 photosynthetic pathway contain proportionally less ^{13}C than the CO_2 they fix during photosynthesis, and their typical $\delta^{13}\text{C}$ is about -29‰. The difference observed between air and plant $\delta^{13}\text{C}$ is usually expressed as carbon isotope discrimination ($\Delta^{13}\text{C}$), and can be calculated as follows (Farquhar et al. 1989):

$$\Delta^{13}\text{C}(\text{‰}) = \frac{\delta_a - \delta_p}{1 + \delta_p} \quad [2]$$

where δ_a and δ_p refer to air and plant composition, respectively. This parameter reflects the amount in which the heavier isotope ^{13}C is discriminated respect the lighter ^{12}C during the physical and chemical processes involved in the synthesis of plant organic matter (Farquhar et al. 1989). The values of $\Delta^{13}\text{C}$ in C_3 plants reflect the balance between diffusion in the intercellular space (mostly through stomata) and carbon fixation by the carboxylating enzyme RuBisCO (Ribulosa Bisphosphate Carboxylase-Oxygenase). When stomata are open (see Fig. 1a), the amount of CO_2 available for fixation is greater and the chances for discrimination against ^{13}C during carboxylation increase (which leads to higher $\Delta^{13}\text{C}$). In contrast, when CO_2 diffusion is limited by stomatal closure plants are forced to fix a higher proportion of ^{13}C , leading to lower values of $\Delta^{13}\text{C}$ (Fig. 1b).

Fig. 1. Simplified scheme of the relationship between carbon isotope discrimination ($\Delta^{13}\text{C}$) in C_3 plants and stomatal conductance. A) High stomatal conductance, high discrimination: CO_2 diffuses



easily into the intercellular space, the activity of the carboxylating enzyme (RuBisCO) is not limited by CO_2 concentration and thus it has more chances to discriminate against ^{13}C . B) Low stomatal conductance, low discrimination: the flux of CO_2 is reduced, and the limiting factor of photosynthesis is stomatal conductance. In this case RuBisCO is forced to fix a higher proportion of ^{13}C . Modified from Ferrio et al. (2003b).

RELATIONSHIPS BETWEEN $\Delta^{13}\text{C}$ OF PLANT MATERIAL AND WATER AVAILABILITY

Plants typically react against a decrease in water availability through stomata closure and thus Δ from plant tissues provide an integrated record of the water status during the time they were formed (see Farquhar *et al.*, 1989 for further details on carbon isotope theory). Many studies under growth-chamber and field conditions have shown that plants developed under water stress produced leaves with lower $\Delta^{13}\text{C}$ (see references in Hubick et al. 1993; Ferrio et al. 2003b). According to these findings, it would be expected to find significant relationships between $\Delta^{13}\text{C}$ and environmental

parameters related with water availability. Although most of the basic studies on $\Delta^{13}\text{C}$ and plant water relations were performed on leaf material (as this is the tissue directly involved in photosynthesis), further works has shown that similar relationships can be established in other plant tissues.

In Fig. 2a, for example, we can see an application of $\Delta^{13}\text{C}$ to the analysis of cereal grains. Araus et al. (1997a; 1999) reported a strong relationship between water inputs and $\Delta^{13}\text{C}$ values from barley (*Hordeum vulgare*) and wheat (*Triticum durum/aestivum*) grains across a wide range of environmental conditions. They included both irrigated and rainfed trials, and total water inputs from heading to maturity (i.e. rainfed plus supplemental irrigation, if any) were considered, as this is the time when grain tissue is formed. Both species gave nearly identical results, showing that, despite their differences, they have similar physiological responses to water stress. On the other hand, it should be noted that in both cases the observed relationship was not linear, suggesting that $\Delta^{13}\text{C}$ was more sensitive to water availability in the driest environments. This could be explained by the fact that the main factor relating $\Delta^{13}\text{C}$ with water availability is stomatal conductance, which is supposed to reach its maximum in non-stressed plants. Thus, under near-optimum water status, no further increments in stomatal conductance, and thus on $\Delta^{13}\text{C}$, would be expected (Farquhar et al. 1989; Lambers et al. 1998).

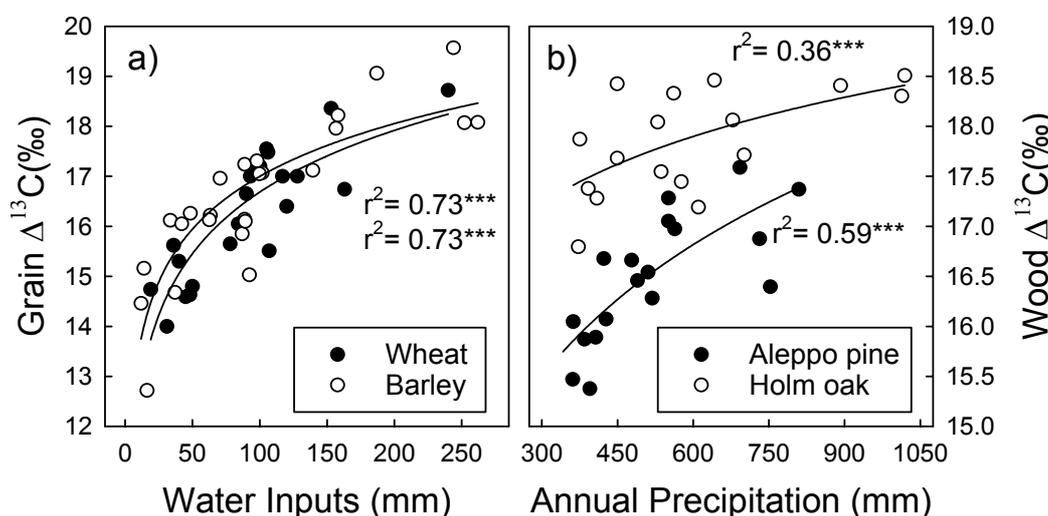


Fig. 2. Relationship between: a) water inputs (WI, rainfall plus irrigation if applied) during grain filling and $\Delta^{13}\text{C}$ of barley and wheat grains. Data from Araus et al. (1997a) for barley and from Araus et al. (1999; 2003) for wheat; $\Delta^{13}\text{C}_{\text{barley}} = 9.99 + 1.52 \cdot \ln(\text{WI})$; $\Delta^{13}\text{C}_{\text{wheat}} = 8.50 + 1.78 \cdot \ln(\text{WI})$ b) mean annual precipitation of 20-25 years and $\Delta^{13}\text{C}$ of wood from the corresponding tree-rings of Aleppo pine and holm oak. Modified from Ferrio et al. (2003a); $\Delta^{13}\text{C}_{\text{pine}} = 4.63 + 1.91 \cdot \ln(\text{WI})$; $\Delta^{13}\text{C}_{\text{oak}} = 11.72 + 0.97 \cdot \ln(\text{WI})$.

Another example of the various species and compounds where $\Delta^{13}\text{C}$ analyses are applicable is shown in Fig. 2b. Ferrio et al. (2003a) studied the $\Delta^{13}\text{C}$ from wood samples of Aleppo pine (*Pinus halepensis*) and holm oak (*Quercus ilex*). Both species are typically Mediterranean, but they display contrasting strategies against drought. The former is a clear drought avoiding species, with a fast stomatal closure, whereas the latter relies mostly on a very effective water uptake, and is partly drought tolerant. They also found a strong relationship between $\Delta^{13}\text{C}$ and annual precipitation (unlike grains, tree rings are formed throughout the year), which was also steeper among the most arid environments. Interestingly, $\Delta^{13}\text{C}$ in Aleppo pine was far more sensitive to water availability than in holm oak. Again, the explanation relies on the close relationship between $\Delta^{13}\text{C}$ and stomatal conductance. As the stomata of Aleppo pine showed faster responses to drought, the $\Delta^{13}\text{C}$ for this species have greater variations with changes in water availability. In contrast, the holm oak can extract water from drier soils, as well as from deeper water tables, showing relatively low variations in

$\Delta^{13}\text{C}$ among trees growing in contrasting environments. Thus, although the relationship between $\Delta^{13}\text{C}$ and water availability is quite clear for most of C_3 , it is species-dependent, as a consequence of the variability in the physiological responses of plants.

APPLICATION OF $\Delta^{13}\text{C}$ TO FOSSIL PLANT REMAINS

Reconstruction of past climate changes in the Mediterranean Basin

The interest in applying $\Delta^{13}\text{C}$ analysis to climate reconstructions was already pointed out in some of the pioneer studies on stable isotopes (e.g. Craig 1954). It soon became an useful extension for tree-ring width measurements, which have been traditionally devoted to palaeoecological studies (Robinson et al. 1990). Since then, wood $\Delta^{13}\text{C}$ has been related to changes in various climatic variables, including humidity (Stuiver & Braziunas 1987; Saurer & Siegenthaler 1989), and precipitation (Warren et al. 2001; Ferrio et al. 2003a), among others. Although most of these works were limited to the last 100 or 200 years, such relationships have been recently extended over longer tree-ring chronologies, allowing high resolution climatic reconstructions back to ca. 11000 years (see references in Heaton 1999). However, these approaches are limited to a few world regions where such long-term chronologies are available. Although they provide important keys for the understanding of global climate changes, it would be helpful to find alternative sources of information to refine climatic reconstructions at the local scale. In this context, similar approaches for the study of climatic evolution have been performed on other plant remains, such as herbarium specimens (Peñuelas & Azcón 1992), fossil leaves (Van der Water et al. 1994; Beerling 1996) or archaeological plant remains (Araus et al. 1997a; Araus et al. 1999). The latter could be specially useful, as plant remains are routinely recovered from archaeological sites, and they can help to answer specific questions related to the human settlements in which they are found.

A case study of the application of archaeological plant remains to the analysis of climate changes is shown in Fig. 3. Charred barley grains were recovered from several archaeological sites in NE and SE Spain (Araus & Buxó 1993; Araus et al. 1997b) and in a Neolithic site from NW Syria (Ferrio et al., unpublished results). $\delta^{13}\text{C}$ was determined by mass spectrometry in the archaeological samples, as well as in present barley grains grown in the same areas (Fig. 3a, below). $\Delta^{13}\text{C}$ values were then calculated as described above. However, in order to calculate $\Delta^{13}\text{C}$ values from ancient plant remains we must bear in mind temporal variations in atmospheric isotopic composition (δ_a in equation 2) and CO_2 concentration. Variations in δ_a in the past have been reported from ice-core records (Leuenberger et al. 1992; Francey et al. 1999) and organic material from C_4 plants (Marino et al. 1992). Taking these data from the literature, δ_a variations throughout the studied period were inferred, as plotted in Fig. 3a, above. Finally, applying the modelled relationship between $\Delta^{13}\text{C}$ and water inputs (Fig. 2a), it was possible to estimate water availability in the past from $\Delta^{13}\text{C}$ of ancient grain samples (Fig. 3b). We observed a consistent decrease in water availability from Neolithic to present-time in the three regions of study, whereas the differences between zones seemed to remain fairly constant. These results clearly indicate that, during early agriculture, barley was cultivated under much better water status than that expected from present-day (rain-fed) conditions in the same areas. Archaeobotanical evidence supports the possibility that environmental conditions during early agriculture were cooler and moister than today, both in the Near East (Harlan 1998; Willcox 1996) and the Iberian Peninsula (Vernet 1990; Vernet & Thiebault 1987). However, planting in naturally wet soils can not be discarded (Hillman 1996).

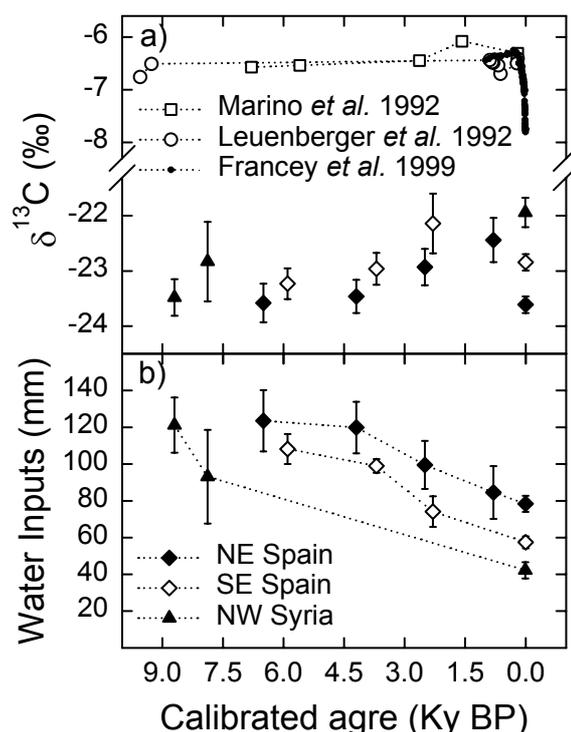


Fig. 3. Estimation of past water inputs from carbon isotope discrimination ($\Delta^{13}\text{C}$) of archaeological cereal grains (wheat and barley combined) from two regions from the Western Mediterranean Basin: A) evolution in isotopic composition ($\delta^{13}\text{C}$) of atmospheric CO_2 (squares, circles and crosses, in the upper half of the figure) and $\delta^{13}\text{C}$ of archaeological cereal grains (triangles and diamonds, below). Both variables (air and plant $\delta^{13}\text{C}$) are required to calculate $\Delta^{13}\text{C}$ of grains, as described in text. Air $\delta^{13}\text{C}$ values came from Leuenberger et al. (1992); Marino et al. (1992); Francey et al. (1999) and cereal data from Araus & Buxó (1993; 1997b). B) Evolution of cereal water inputs (estimated from $\Delta^{13}\text{C}$) from Neolithic to present times in the areas studied. See text for further explanation.

Can we discriminate between climatic and anthropogenic effects on $\Delta^{13}\text{C}$?

Once the climatic variability through time has been accounted, it is possible to find evidences of different water management strategies looking at the relationships between different species grown in the same zone. This is the case exemplified in Fig. 4, using data from the archaeological sites in SE Spain (Araus et al. 1997b). Firstly, we compared the relationship across the different sites between the $\Delta^{13}\text{C}$ of barley grains and $\Delta^{13}\text{C}$ of wheat grains and faba beans (Fig. 4a). We found that $\Delta^{13}\text{C}$ values in barley and wheat grains were well correlated, showing similar trends across the sites. This suggest that both cereals were grown under similar conditions, and responded to common (probably climatic) factors affecting their water status. In contrast, values of $\Delta^{13}\text{C}$ for faba beans were unrelated to those of cereal grains, and also significantly higher (about 1‰ in average). Differences in $\Delta^{13}\text{C}$ between cereals and grain legumes could be explained (at least in part) in terms of differences in grown pattern. Whereas cereals are determinate plants, in which grain growth coincides with the beginning of drought in the Mediterranean region, legumes are indeterminate plants, producing successive pods throughout the crop cycle. Therefore, for faba beans, most seeds may develop under a better water status. On the other hand, the possibility that legumes were irrigated, even when cereals were cultivated under rainfed conditions, can not be discarded. Indeed, Araus et al. (1997b) found that $\Delta^{13}\text{C}$ of archaeological faba beans were closer to those measured in present day irrigated crops near the sites (16.0-17.9‰) than to those found in rainfed crops (14.9‰). In addition, the relative frequencies of grains from different crop species suggest that the area under grain legumes would be very small compared with that of cereals. Therefore, such small irrigated areas would be more feasible. Intensive (i.e. under irrigation) cultivation of faba bean could ensure a basic protein

source in the human diet (Heiser 1990). The combination of extensive cereal crops expanding around human settlements with smaller plots of vegetables and legumes close to the living area seemed to be the most common pattern in subsistence and pre-industrial agriculture (Hillman 1973; 1984). The above data suggest that this kind of management of water and soil resources could have been initiated during the early phases of agriculture.

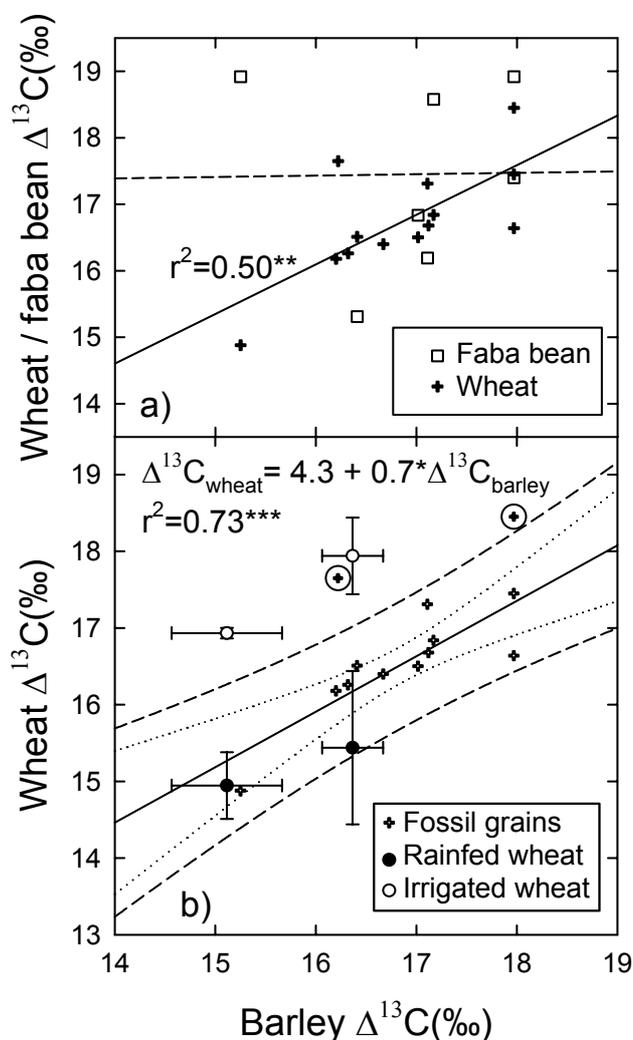


Fig. 4. Relationship between barley $\Delta^{13}\text{C}$ and either wheat or faba bean $\Delta^{13}\text{C}$ across archaeological sites in SE Spain. A) Comparison between wheat and faba bean. B) Detection of outliers for the relationship between wheat and barley $\Delta^{13}\text{C}$, taking present data for comparison. Crosses, archaeological sites from SE Spain; Full circles, present data for barley and wheat (both rainfed) growing in the same area; Empty circles, present data for rainfed barley co-occurring with irrigated wheat. Outliers from archaeological data are encircled. Present-day data are from SE Spain (Araus et al. 1997b), NE Spain (Araus et al. 2003).

Further analysis of the relationship between barley and wheat $\Delta^{13}\text{C}$ can provide additional information. At first glance, it is somewhat surprising that, despite both species followed the same relationship between water inputs and $\Delta^{13}\text{C}$ (Fig. 2a), $\Delta^{13}\text{C}$ values of wheat were consistently lower than those of barley across sites. This suggest that barley grains were grown with greater water inputs than wheat. Would this mean that barley were selectively growth under better conditions than wheat?. Apparently not: if we consider the current growth cycles of the two species, we can find that barley grows up faster than wheat, reaching maturity about 1-2 weeks before. Thus, even growing at the same site, barley grains in Mediterranean climates are generally formed under moister conditions than wheat grains, as drought has a steep increase during the last weeks of the growth cycle.

Consequently, barley grains have generally higher $\Delta^{13}\text{C}$ values than the co-occurring wheat grains. However, in Fig. 4a we can see two points where wheat have considerably greater $\Delta^{13}\text{C}$ than barley, falling far away from the fitted regression line. After removing these two “outliers” from the fitted relationship, it became stronger and more significant (Fig. 4b). It is likely that these outliers indicate some selective treatments to enhance the performance of wheat crops, but not applied in barley, including irrigation or planting in naturally wet alluvial soils (Bar et al. 1989; Araus et al. 1997b). When we plotted present values of rainfed barley, compared with either rainfed or irrigated wheat, we found that the plots for rainfed wheat fitted well within the regression line for the archaeological sites. In contrast, the plots of rainfed barley related to irrigated wheat fell out of the confidence intervals of the regression line, and showed values for wheat $\Delta^{13}\text{C}$ similar to those of the presumed outliers. This further supports the idea that the two sites showing greater wheat $\Delta^{13}\text{C}$ involved a differential water management for wheat and barley. As barley is less drought-sensitive than wheat, it is a common practice in dry areas to keep barley as a rainfed crop, reserving any additional water supplies, or the moister soils (e.g. closer to a water stream) for wheat. Currently, this practice is enhanced by the fact that most of barley harvest is addressed to animal feeding, whereas wheat is preferred for human consumption. Again, we can find evidences of water and soil managements during early agriculture that resemble those currently found in some areas. This could help us to evaluate the potential long-term impact of these practices, looking at the consequences they had in the past.

CONCLUSIONS

Despite the constraints of inferring past conditions based on present relationships, this novel approach can help us to identify events in which early farmers started to develop a conscious water management to improve their crops performance. We can also relate them with the climatic conditions in which they were developed, or look for possible changes in response of climate and/or landscape changes. The long-term consequences of these practices could be further evaluated by combining $\Delta^{13}\text{C}$ analyses with the miscellaneous data available from archaeological sites. Among others, demographic or dietary shifts might be the consequence of changes in crop management, and might provide keys of the most suitable alternatives for a given scenario.

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