

## Dating the humans by radiocarbon

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**Summary.** — Radiocarbon has become a very powerful tool used for dating. This paper deals with a specific application of  $^{14}\text{C}$ , *i.e.* dating of humans. Attention is focused on those aspects that, if neglected, might lead to a misinterpretation of the results or to an unsatisfying accuracy of the measurement. After a brief description of the main principles on which the radiocarbon method is based and of Accelerator Mass Spectrometry, examples taken from the research activity of INFN-LABEC (Laboratorio di Tecniche Nucleari per i Beni Culturali) in Florence are presented. The case of the relics of St. Francis represents an example of dating not directly human remains but other objects that can be associated to them. The case of two burials from the archaeological area of Baratti-Populonia, in Tuscany, gives the possibility to show the importance of estimating the human palaeodiet when dating bone samples.

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### 1. – Introduction

Dating the humans is a key point in archaeology and history in general. Radiocarbon is definitely the best known and most used natural chronometer that can be used. Many examples can be found in the literature.  $^{14}\text{C}$  has been employed in the chronological study of a specific individual and his temporal and geographical context, as in the case of Ötzi, the mummy recovered on the Alps close to the border between Italy and Austria in 1991 [1]. It has been also employed in the study of ancient populations, both in historical and prehistorical times. An interesting example, probably not well-known yet, is the study of *Homo floresiensis* [2], a new species of hominids discovered on the island of Flores in Indonesia some years ago.

In this kind of applications, sample materials to be dated are bones and even human tissues, in order to directly date the individual, or also charcoal, textiles, leather and

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any other object that can be associated to the person. In the latter case, of course, dating is indirect and particular attention must be paid in the interpretation of results. However, also in the former case, dating might be not so straightforward, since, when dealing with humans, the carbon reservoir might be not well known at first (the idea of carbon reservoir will be introduced in sect. 2).

This paper discusses the peculiarity of dating of humans; in particular, examples taken from the activity of LABEC (Laboratorio di tecniche nucleari per i Beni Culturali), in the Florence unit of Istituto Nazionale di Fisica Nucleare, are presented.

## 2. – Basic principles of $^{14}\text{C}$ dating

There are many sources in the literature where the radiocarbon dating method is described (*e.g.* [3]); it can be useful anyway to briefly recall here the basics at least.

$^{14}\text{C}$  is one of the three carbon isotopes naturally occurring on Earth. Its origin is cosmogenic: it is continuously created in the upper atmosphere owing to neutrons — produced by the cosmic rays — that interact with  $^{14}\text{N}$  via the (n, p) reaction. Once it is formed, it oxidizes to  $\text{CO}_2$  and then enters the natural carbon cycle involving the oceans and the biosphere. However,  $^{14}\text{C}$  concentration does not increase in an indefinite way, due to the fact that this isotope is radioactive. Indeed, it decays to  $^{14}\text{N}$  by emission of  $\beta^-$  (and antineutrino), with a half life of  $5730 \pm 40$  years. The times characteristic of carbon exchanges between the different carbon reservoirs are much smaller than  $^{14}\text{C}$  half life, so that radiocarbon concentration reaches equilibrium and can be considered approximately uniform throughout atmosphere, oceans and biosphere:  $^{14}R = ^{14}\text{C}/^{12}\text{C} \sim 1.2 \cdot 10^{-12}$ . This is also the concentration in all organisms until they are alive. When they die, since no carbon uptake may continue any longer, the number of radiocarbon nuclei begins to exponentially decrease due to the radioactive decay. As  $^{12}\text{C}$  is stable,  $^{14}\text{C}$  concentration decreases as well. Assuming as known the  $^{14}\text{C}$  concentration  $^{14}R_0$  at the instant 0 (the death of the organism) and the isotope mean life  $\tau$ , and measuring the residual  $^{14}\text{C}$  concentration ( $^{14}R(t)$ ), it is possible to calculate the age  $t$ , *i.e.* how many years have elapsed from the instant 0:

$$(1) \quad t = \tau \ln \frac{^{14}R_0}{^{14}R(t)}.$$

This equation is the core of the radiocarbon dating method. The international community has assumed some conventions to determine  $t$ , the conventional radiocarbon age:  $\tau$  is chosen as the so-called Libby mean life, 8033 years;  $^{14}R_0$  is the equilibrium concentration in atmosphere in a reference year, 1950 ( $^{14}R_0 \sim 1.2 \cdot 10^{-12}$ ); concentrations are always corrected for isotopic fractionation normalizing to  $-25\%$ . The conventional radiocarbon age is expressed in years BP (Before Present, 1950). Typically, the measured  $^{14}R(t)$  is expressed as a fraction of the reference equilibrium concentration, *i.e.* in pMC (percent of Modern Carbon) or in  $F_m$  (fraction of modern).

It is well known that the conventional radiocarbon age does not correspond to the true calendar age, because all the assumptions at the basis of eq. (1) are only valid as a first approximation. A calibration procedure is thus mandatory. Actually, thanks to the work done during many years by many laboratories worldwide, a curve in which

radiocarbon ages are plotted *vs.* true calendar ages has been built, by measuring  $^{14}\text{C}$  in thousands of samples that were also datable by other independent methods. Most of these samples are tree rings, in order to reconstruct the calibration curve for relatively recent years, back to about 12000 years ago; for older periods, corals and foraminifera in layered sediments have been used. The internationally agreed calibration curve has been recently updated and published as IntCal09 [4]: it is now extended back to about 50000 years ago, covering the full time range that can be investigated by radiocarbon (beyond 50000 years the concentration becomes so low that the sensitivity limits of the available measuring techniques are reached).

It is worth noticing that IntCal09 refers to atmospheric data and thus it can be applied in the case of those organisms that were living exchanging carbon directly either with the reservoir atmosphere or with other reservoirs exchanging directly with atmosphere itself. Organisms living in the oceans deserve a specific approach. In fact, oceanic waters are generally depleted in  $^{14}\text{C}$ , due to the delay in exchange rates between atmosphere and oceans, and to the dilution effect caused by the mixing of surface waters with upwelled deep waters which are old (even up to a few millennia). The consequence is that samples taken from the sea, if calibrated with IntCal09, can have an apparent age (reservoir age), appearing as average about 400 years older than they actually are. Even though this effect can vary depending on the place, an average marine calibration curve has been built (see, *e.g.* Marine09 [4], also extended back to about 50000 years ago as IntCal09): this can be used to calibrate conventional radiocarbon ages measured in marine samples. The reason why the topic of marine reservoir age can be of interest when dating humans will be explained in sect. 5.

### 3. – Accelerator Mass Spectrometry at LABEC, Florence

Accelerator Mass Spectrometry (AMS) has now become the leading technique to measure  $^{14}\text{C}$ , thanks to its extremely good selective sensitivity, which gives us the possibility to measure very small samples (of the order of few mg) in a reasonable short time [5]. This is possible by coupling the selective elements employed in the “traditional” Mass Spectrometry, such as magnetic and electrostatic analysers, with a particle accelerator. Typically, a Tandem electrostatic accelerator with a maximum terminal voltage of some MV is used. In AMS, the mechanisms themselves used for the injection and the acceleration of the charged beam are exploited as filters to discriminate the rare isotope from its isobars. For example, in Tandem accelerators, negative ions are extracted from the source and injected into the tube, so, in the case of radiocarbon, the elemental isobar  $^{14}\text{N}$  is already suppressed at the beginning, since nitrogen does not form stable negative ions; the molecular isobars, *e.g.*  $^{12}\text{CH}_2$  and  $^{13}\text{CH}$ , are suppressed at the high-voltage terminal thanks to the charge-exchange interactions (stripping) to convert the injected negative ions into positive ions for the further acceleration step (actually, molecules dissociate into their atomic components).

Figure 1 shows the 3 MV Tandem accelerator installed at LABEC [6].

The AMS beam line (see fig. 2) is equipped with a Cs sputtering ion source to extract negative ions from the samples to be dated. The beam is first analyzed according to the energy/charge ratio by passing through an electrostatic analyzer and then according to the mass/charge ratio in a bouncer magnet. The mechanism used to sequentially inject ions of mass 14, mass 12 and mass 13 into the accelerator tube can be easily explained as follows: the magnetic field is set to transmit one of the three masses (13 in our case) and

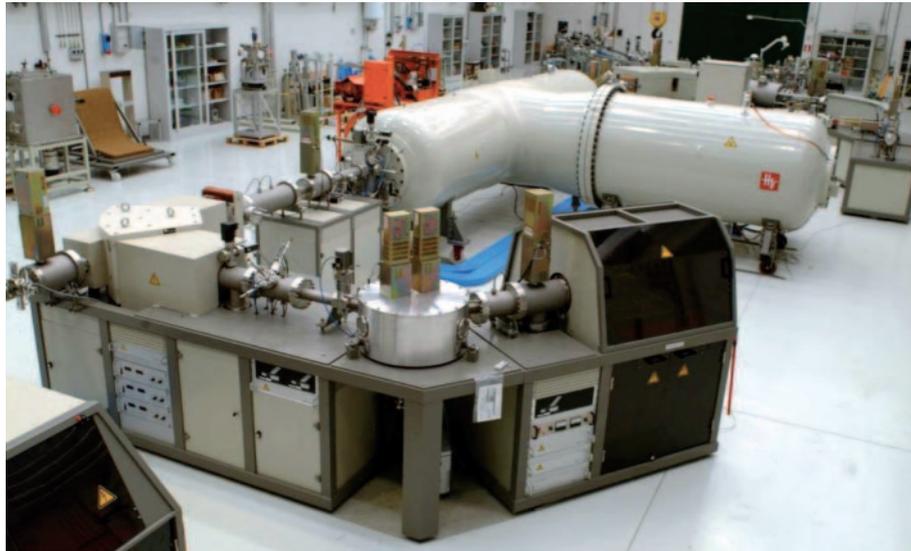


Fig. 1. – The 3 MV Tandem accelerator installed at LABEC (Laboratorio di Tecniche Nucleari per i Beni Culturali) of INFN in Florence; in the foreground, the low-energy side of the AMS beam line.

the transmission of the others is achieved by sequentially applying an appropriate voltage to the electrically insulated chamber of the magnet, so that their magnetic rigidity inside the magnet is the same of that of mass 13. Ions are accelerated up to the terminal voltage generally kept at 2.5 MV; at the terminal, passing through a narrow canal some argon gas is flown, the ions undergo stripping. On the high-energy side of the Tandem, an analysing magnet selects those  $^{14}\text{C}$  ions of a given charge state (the most abundant is  $3+$ , in these conditions) and, accordingly, of a given energy. Stable isotopes abundances ( $^{13}\text{C}$  and  $^{12}\text{C}$ ) are measured just after this analysing magnet using two off-axis Faraday cups. A further filter, an electrostatic analyser to eliminate possible residual non- $^{14}\text{C}$  ions that have however mass-charge-energy combinations such to be transmitted through the analysing magnet, is present before  $^{14}\text{C}$  ions reach the particle detector, formerly a gas ionisation chamber, now a silicon diode.

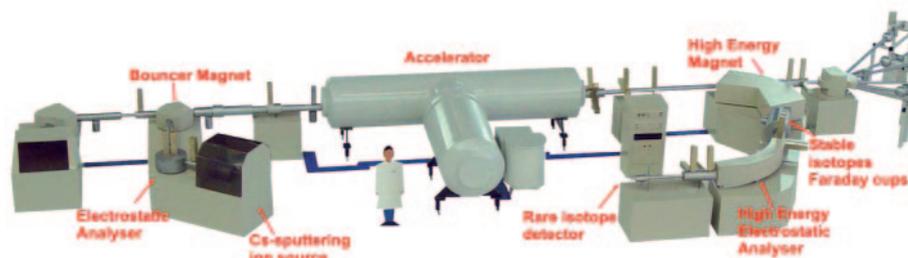


Fig. 2. – Model of the accelerator; the main components along the AMS beam line are highlighted.

LABEC is also equipped with a sample preparation laboratory, to convert any sample to be radiocarbon dated to a pellet of graphite that is the appropriate form to be used in the Tandem source for the AMS measurements.

#### 4. – Dating the humans or their context: the example of the relics of St. Francis

As already mentioned in the introduction, humans can be studied indirectly by dating their equipment or the environment that surrounded them. In this case, however, it is important to remember that radiocarbon data can only give indications whether the result is compatible or not with the attribution of the dated samples to a human in particular.

This is the case, for instance, of radiocarbon measurements of what are probably among the most debated finds related to humans, *i.e.* relics. Sometimes, apart from their religious value, dating of relics gives important information about history and, in particular, about history of religion. In this sense, a good example is the dating of the relics of St. Francis from Assisi (1182–1226), performed at LABEC [7].

We dated some textile samples from two relics kept in the Church of St. Francis in Cortona: a woollen frock and a precious embroidered pillow (with some woollen and linen cases inside). According to tradition, these relics are connected with St. Francis' death: the frock was used to cover the body of the Saint, and the pillow used to lean his head, while he was passing away. Later, they were carried to Cortona by Friar Elia (1180?–1253), when he moved there from Assisi to establish one of the first Franciscan communities in Italy. From the historic point of view, the figure of Friar Elia has presented some controversial aspects, thus dating of the two relics has also represented the possibility to study the context of the birth of the church in Cortona: an example of how radiocarbon can help in defining the historical context in which humans lived. In addition to these relics, for comparison, we also dated the frock of St. Francis kept in the Florentine church of Santa Croce.

Details of the measurements were already discussed in the mentioned paper. Here, only the results are reported (see fig. 3). As far as the two frocks are concerned, radiocarbon ages shown in the graph are the results of weighted averages calculated over several measured samples from each of the two frocks; the two dates for the pillow are the radiocarbon ages measured for two of the inner cases found inside it (one sample for each of them). For calibration, the OxCal software was used [8, 9], exploiting the IntCal04 calibration curve [10]. In the graph, distributions of probability for calibrated ages, in black, are plotted over the calibration curve; time intervals calculated at 95.4% of probability are also indicated. The data from the Cortona frock and from the inner pillow cases are consistent with each other; moreover, they are compatible with the period of life of St. Francis, who died in 1226. In particular, at 95.4% of probability, the Cortona frock has been dated to the interval 1155–1225 AD. On the contrary, the discrepancy of the Santa Croce frock is extremely evident: actually, at 95.4% of probability, it has been dated to one of the intervals 1280–1310 AD or 1360–1388 AD. It is interesting to notice that, in this case, as can sometimes happen in radiocarbon dating, due to the peculiar trend of the calibration curve, the good precision on the measured radiocarbon age does not turn into a similarly good precision on the calibrated age. Nevertheless, such a result clearly shows that this frock cannot be compatible with the period of life of the Saint and this is a result that can be anyway useful for the study of the history of religion during the Middle Ages.

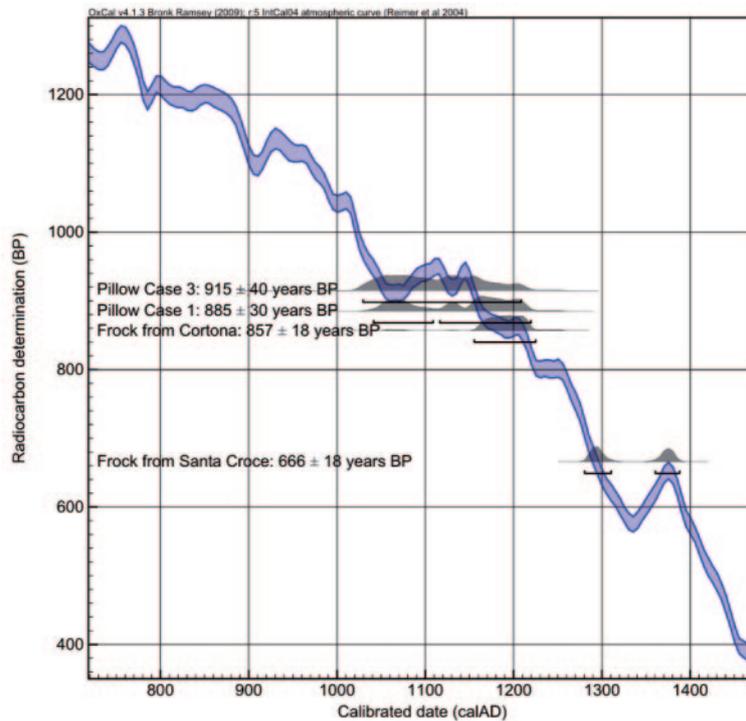


Fig. 3. – Calibration of the measured radiocarbon ages of the samples from St. Francis relics. Distributions of probability for calibrated ages, in black, are plotted over the calibration curve; time intervals calculated at 95.4% of probability are also indicated.

##### 5. – “der Mensch ist was er isst”: the example of the tombs in the Baratti-Popolonia area

Dating of bones gives us direct information on the age of the humans; care has to be paid also in this case.

First of all, what is critical is the choice of the carbon to be dated. Bone is a very complex material, containing carbon both in an inorganic form, as calcium carbonate (that forms the mineral component of bones together with calcium phosphate), and in an organic form, as the collagen protein. Experience has demonstrated that collagen is the most reliable bone fraction that can be dated [11]; some laboratories even extract specific amino acids from the protein [12]. Nevertheless, collagen is a very perishable material: bad environmental conditions for conservation of bones can cause the contamination or the degradation/loss of collagen itself, leading to the impossibility of dating it. Some criteria, *e.g.* the measurements of the infrared absorption spectra [13] or of the C/N ratio [14], have thus been studied to have an estimation of the state of preservation of the collagen. At LABEC, for example, after extraction of collagen from bone samples to be dated, its quality is evaluated by measuring the C/N ratio; the measurement is performed by combustion of the sample into the CHN elemental analyser (then, the CO<sub>2</sub> coming out of the elemental analyser is collected and converted to graphite to be measured by AMS).

Another critical aspect of bone dating is how much the carbon reservoir from which the humans assumed  $^{14}\text{C}$  is known. In other words, humans do not uptake radiocarbon directly from the atmosphere. Their first source of carbon is food: a human is what he/she eats. Actually, food can partly have a terrestrial origin, *e.g.* vegetables and meat as beef, pork, chicken, etc. and partly a marine origin, *e.g.* fish. In sect. 2, the marine reservoir age effect has been already introduced. Thus, it can be easily understood that when a fraction of the diet of an individual is based on fish or other marine-derived foods, his/her radiocarbon concentration during life in equilibrium conditions should be lower than the concentration in atmosphere. If this individual was radiocarbon dated without taking into account his diet, we would measure an older age: a larger fraction of marine diet would correspond to a larger discrepancy between the measured age and the true age [15]. Of course, humans are omnivore; besides, eating habits have not been the same all over the world, in any geographical location, and have changed during the centuries. Accuracy of bone dating cannot thus be improved by an *a priori* knowledge, but, for every single case, an estimation of the human diet, or better palaeodiet, can be useful. This can be done by measuring the isotopic fractionation of light elements, *i.e.*  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  [16]. Indeed,  $\delta^{13}\text{C}$  varies according to whether the protein component of human food is mainly based on terrestrial or marine components and on  $\text{C}_3$  plants (and on herbivores eating  $\text{C}_3$  plants and on carnivores accordingly) or  $\text{C}_4$  plants (and on herbivores eating  $\text{C}_4$  plants and on carnivores accordingly). The two labels  $\text{C}_3$  and  $\text{C}_4$  indicate the different processes of photosynthesis:  $\text{C}_3$  characterizes trees and temperate regions plants, including, for example, wheat and barley;  $\text{C}_4$  characterizes subtropical region plants, including also few plants that can be found in our environment such as millet.  $\delta^{15}\text{N}$  reflects the so-called trophic level: the higher is the nitrogen fractionation, the higher is the step in the food chain. In this way, the consumption of more vegetables than carnivores (or *vice versa*) can be estimated.

An example of how helpful the evaluation of the palaeodiet can be is the dating performed at LABEC of two interesting sepultures found in the “Parco Archeologico di Baratti-Populonia”, in Southern Tuscany (Italy) [17].

The area of Baratti-Populonia is known as one of the most important Etruscan sites for metal working, especially iron. Moreover, the ancient town remained an active centre also when the Etruscan civilization started to decline due to the influence of the Romans. The case of these two tombs (Tomb11 and Tomb12) has been very interesting, because they were unexpectedly found in a place close to an ancient industrial area, far away from other necropolises. Tomb11 and Tomb12 hosted the skeletons of a man and of a woman, respectively; on the basis of the funerary goods (some glass objects, a coin, some gold jewels), archaeologists roughly dated back the individuals to the Roman Imperial Age. At LABEC, we dated some samples from the ribs of the two skeletons; collagen was extracted, purified and finally converted to graphite. Figure 4 gives the idea of how much material (bone) is needed in the case of a radiocarbon AMS measurement.

Table I summarizes the results of the measurements performed on the two individuals. Radiocarbon concentrations are expressed as pMC: measured  $^{14}\text{C}/^{12}\text{C}$  isotopic ratios were corrected for isotopic fractionation ( $^{13}\text{C}/^{12}\text{C}$  isotopic ratio simultaneously measured in the AMS beam line during each run) and background, and then these corrected values were normalized to the isotopic ratio measured for a set of NIST Oxalic Acid II standards. Corresponding conventional radiocarbon ages calculated according to eq. (1) are also shown. Without taking into account any additional information, conversion to calendar ages would be performed using the terrestrial calibration curve (see the Cal age(IntCal04) column in table I). In this case, the additional information, as discussed above, is given

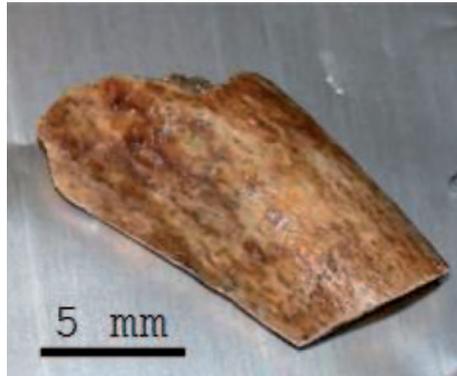


Fig. 4. – Photo of one the ribs samples from the Baratti-Populonia skeletons prepared and measured by AMS at LABEC.

by the measurement of carbon fractionation,  $\delta^{13}\text{C}$ . Indeed, for the two individuals,  $\delta^{13}\text{C}$  is lower than what is expected in the case of a 100% terrestrial diet (based on  $\text{C}_3$  vegetables). In principle, this discrepancy can be explained assuming either a fraction of marine organisms in their diet or a terrestrial supply partly based on  $\text{C}_4$  plants. The latter can however be excluded thanks to comparison of these  $\delta^{13}\text{C}$  values with data found in the literature (*e.g.*, [18,19]). By stable isotopes ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , as mentioned), it has been measured that, during the first centuries AD, the diet of Roman populations living along the Mediterranean coasts was partially based on fish or other marine food consumption. On the contrary,  $\text{C}_4$  plants, even millet, were not so used.

Hence, assuming that a fraction of the diet of the two humans is marine derived, from the measured isotopic fractionation (see table I), we can estimate the percentage by linear interpolation considering as endpoint values  $\delta^{13}\text{C} = -21\text{‰}$  for a 100% terrestrial diet and  $\delta^{13}\text{C} = -12.5\text{‰}$  for a 100% marine diet (as can be found in the literature [15]). A fraction of about 30% marine diet has been obtained. The next step is the reconstruction of a “custom” calibration curve, obtained by mixing 70% of terrestrial calibration curve (in this case, IntCal04 [10]) with 30% of marine calibration curve (in this case, Marine04 [20]). Results are shown in the last column of table I: calibrated ages are shifted of about one hundred years towards a more recent period. This result has turned out to be in agreement with the hypothesis made by the archaeologists. Not taking into account the information on the diet would have made the two skeletons seem older. Of course, as one

TABLE I. – Measured data for the two skeletons from “Parco Archeologico di Baratti-Populonia”:  $^{14}\text{C}$  concentrations (in pMC), corresponding conventional radiocarbon ages (in years BP) and  $\delta^{13}\text{C}$  measurements of bone samples are reported with experimental uncertainties quoted as 1 sigma; instead, time ranges of calibrated ages are reported at 95.4% confidence level.

	pMC	years BP	Cal age (IntCal04)	$\delta^{13}\text{C}$ (‰)	Cal age (mixed curve)
T11	$78.09 \pm 0.23$	$1987 \pm 25$	42 BC–65 AD	$-18.0 \pm 0.2$	62–210 AD
T12	$78.48 \pm 0.29$	$1946 \pm 30$	21 BC–127 AD	$-18.6 \pm 0.2$	88–238 AD

can infer from the discussion at the beginning of this section, accuracy of bones dating might be further improved by measuring  $\delta^{15}\text{N}$  too.

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