DETERMINING ^{14}C CONTENT IN DIFFERENT HUMAN TISSUES: IMPLICATIONS FOR APPLICATION OF ^{14}C BOMB-SPIKE DATING IN FORENSIC MEDICINE

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ABSTRACT. Various samples extracted from human tissues (with different radiocarbon turnover rates) of a post-bomb human body were submitted to accelerator mass spectrometry (AMS) ^{14}C dating: hair; a cortical fraction of a skull bone; a trabecular fraction of a pubic symphysis; and enamel extracted from permanent teeth with different dates of formation were analyzed. The analyzed samples showed varying ^{14}C concentrations corresponding to different times of formation or different turnover rates. The implications of the results in forensics studies are discussed.

INTRODUCTION

The large increase in atmospheric radiocarbon concentration since ~AD 1950 due to the above-ground nuclear detonation tests has been used as a powerful tool for the absolute ^{14}C dating of different kinds of samples relevant in forensic disciplines (Quarta et al. 2005). The method is based on determining the ^{14}C concentration in analyzed samples, typically expressed in terms of $^{14}C/^{12}C$ isotopic ratios and in the conversion of the determined value into calendar years by using the atmospheric bomb peak as a calibration curve. The rapid variation of the atmospheric ^{14}C concentration enables very high chronological resolution on the order of a few years or even better. Several studies have shown the potential of this technique in different applications relevant to forensics, such as in the characterization of illicit drugs (Zoppi et al. 2004) and the identification of counterfeit food and wines (Tuniz et al. 2004).

This paper focuses on the application of ^{14}C bomb-peak dating in forensic anthropology and in particular on the possibility to determine the date of birth of an individual by combining the information that can be obtained by ^{14}C dating different human tissues. In fact, depending on the corresponding rates of formation and/or turnover, different tissues are expected to exhibit ^{14}C levels corresponding to different years. Several studies have shown how significant variations of the ^{14}C content can be measured in different kinds of samples extracted from the same individual. Essentially, as far as ^{14}C dating is concerned, the different kinds of samples can be grouped depending on the rates of carbon replacement and turnover in high turnover tissues, slow turnover tissues, and non-turnover tissues. In high turnover tissues such as hair, nails, blood, and bone lipids, carbon is continuously replaced and the corresponding ^{14}C levels are thus closely linked to the atmospheric trends. Previous studies have shown how these tissues can be used to determined the date of death (Wild et al. 2000; Marzaioli et al. 2011). However, these tissues are not always available, such as in skeleton remains. Among the slow turnover tissues, the most important is surely bone collagen, which is continuously replaced and remodeled during life with very long residence times (20–30 yr) and with carbon uptake rates that are variable during one's lifespan. In terms of bomb-spike dating, this means that ^{14}C levels measured in bone collagen do not reflect the value corresponding to the date of birth nor the date of death. Furthermore, different remodeling rates for the trabecular and cortical parts of the bones have been demonstrated (Ubelaker et al. 2006). Non-turnover tissues are those that do not turnover during one's lifetime. Therefore, in these kinds of tissues the ^{14}C concentration should reflect the atmospheric value at the time of their formation at a particular stage in the life of the indi-

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vidual. For instance, a method to determine the date of birth by measuring the ^{14}C concentration in the enamel of teeth formed at different times after birth has been proposed (Spalding et al. 2005). In another study, the possibility to use dentine and cementum was also suggested (Cook et al. 2006).

It is therefore important to investigate how the ^{14}C dating of different tissues allows one to obtain highly complementary information such as the dates of birth and death. Furthermore, the analysis of tissues with different turnover rates allows one to discriminate between 2 possible intervals, one before and one after the 1963–1964 maximum, which are always obtained when calibrating the measured ^{14}C concentration through the bomb-peak curve.

This paper presents the ^{14}C measurements of different tissues extracted from a post-bomb human body. The results obtained in the analysis of the enamel extracted from different teeth, hair, and cortical and trabecular bone are presented and discussed in terms of the implications for forensics studies.

SAMPLE SELECTION AND METHODS

Different samples were obtained from a corpse found in 2010 in an artificial lake in northern Italy. An autopsy revealed that the person had been shot in the face. The body was attributed by the Italian police to a 36-yr-old male, born in 1973 and having disappeared at the end of 2009, the supposed date of death. Samples were selected and extracted from the body during the autopsy carried out at the Department of Human Morphology and Biomedical Sciences, University of Milano, Italy, according to Police Mortuary Regulations. Samples from tissues with different turnover/replacement rates such as hair (high turnover), bone collagen from trabecular and cortical bone (slow turnover), and enamel extracted from teeth with different times of formation (non-turnover) were selected (Table 1). All samples were chemically processed and ^{14}C dated by accelerator mass spectrometry (AMS) at CEDAD (Centre for Dating and Diagnostics), Department of Engineering for Innovation, University of Salento, Italy (Calcagnile et al. 2004a,b). The chemical processing employed depended on the type of sample. For the teeth (samples S2 and S3), ~100 mg of the crown were cut from the root at the level of the cervical line. Crown enamel was washed twice with H_2O_2 (30%) in a water-bath sonicator for 15 min and then dried at 60 °C for 24 hr. CO_2 was then extracted by acidification with concentrated H_3PO_4 (85%) at 85 °C.

Table 1 Summary of the selected samples.

Sample ID	Sample	Analyzed fraction
S1	Head hair	Hair keratin
S2	Mandibular right canine	Dental enamel
S3	Mandibular right second molar	Dental enamel
S4	Pubic symphysis	Trabecular bone
S5	Base of skull	Cortical bone

For the bone samples (S4 and S5), collagen was extracted following the Longin (1971) protocol, using ~500 mg of sample material. The samples were first washed in acetone. Demineralization was achieved with HCl (1%), and collagen was then filtered using 0.45-µm pore silver filters. The extracted collagen was combusted to CO_2 at 900 °C in sealed quartz tubes together with CuO and silver wool.

Hairs (sample S1) were treated, after a first attack with acetone, following the standard acid-base-acid (ABA) treatment routinely used at CEDAD (D'Elia et al. 2004; Gianfrate et al. 2007). The extracted material was then converted to CO_2 in sealed quartz tubes as for the bone samples.

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The CO_2 extracted from all the samples was reduced to graphite at 600 °C using hydrogen as a reducing agent and iron powder as catalyst. The obtained graphite was then pressed in the target holder of the AMS system installed at CEDAD for determination of the ^{14}C concentration (Calcagnile et al. 2005). The ^{14}C results were corrected for isotopic fractionation, by using the $\delta^{13}C$ values measured with the accelerator, and for sample processing and machine background.

RESULTS AND DISCUSSION

The results of ^{14}C dating are given in Table 2. Results are expressed as fraction modern with the quoted uncertainty corresponding to a 1σ confidence level. Different ^{14}C levels were observed for the different tissues. Figure 1 shows the atmospheric ^{14}C concentration in the Northern Hemisphere. For construction of the curve, the data from Levin and Kromer (2004) and Levin et al. (2008) were used. Since these curves extend up to 2006 and the last part of the curve from 2006 to the present was extrapolated, Table 2 shows also the calibrated ages obtained by using the data sets of Levin and Kromer (2004) and Levin et al. (2008) and the CALIBomb software with a smoothing of 1.0 yr and a resolution of 0.2 yr. Only the date ranges corresponding to the descending part of the curve have been reported. For sample S1 (hair), the value falls beyond the age limit of the data sets used. For this sample, a calibrated age of 2009.0 ± 2.0 was determined by intercepting the extrapolated part of the curve shown in Figure 1. We note that this value is consistent with the atmospheric ^{14}C concentration of 1.046 ± 0.12 as determined in 2009 for the United States (Norton 2011). The measured ^{14}C value for this sample would allow determination of the date of death by considering the fast replacement rate of human hair as 2009.0 ± 2.0, which fits with the time expected. The relatively large uncertainty associated with this determination is evidently due to the flattening of the curve for recent time ranges. We also observe that the analysis of just this sample would not allow to discriminate whether the sample belongs to the ascending or descending part of the curve.

Table 2 Results of AMS ^{14}C dating analyses.

Sample ID	Fraction modern (±1σ)	Calibrated ages (1σ ranges)	Dates yr AD (1σ)
S1	1.0493 ± 0.0056	2007–2011	2009.0 ± 2.0
S2	1.3632 ± 0.0079	1975.4 (May)–1976.7 (Sep)	1976.0 ± 0.7
S3	1.3182 ± 0.0063	1977.9 (Nov)–1979.3 (Apr)	1978.6 ± 0.7
S4	1.0674 ± 0.0044	2003 (Jan)–2006.7 (Sep)	2004.9 ± 1.9
S5	1.1463 ± 0.0044	1990.5 (Jun)–1992.8 (Oct)	1991.6 ± 1.1

For the bone collagen samples (S4 and S5), the measured ^{14}C values are different for the cortical and trabecular parts of the bone. In particular, sample S4 gives a ^{14}C level lower than S5, which is consistent with the different remodeling rates. Since the studied individual was born after the 1963–1964 bomb peak, this confirms a faster turnover rate of the trabecular bone as observed by Ubelaker and Parra (2011). In particular, sample S5 can be dated to 1991.6 ± 1.1 and S4 to 2004.9 ± 1.9, with a relative offset of 13.3 yr. A lag time of 18.1 ± 1.6 yr exists between the age of cortical bone collagen and the year of birth (1973.5 ± 0.5) and of 18.2 ± 1.3 with the year of death, which was taken as 2009.8 ± 0.2 since this particular male died at the end of 2009. The time lag between the age of the trabecular bone and the years of death and birth are 4.9 ± 2.1 and 31.4 ± 2.4 yr, respectively.

For the teeth samples (S2 and S3), the 2 different ages correspond to different times of formation of the crown. For the 2 analyzed teeth, the time of crown formation can be estimated as 3–6 (4.5 ± 1.5) and 3–8 (5.5 ± 2.5) years of age for samples S2 (mandibular right canine) and S3 (mandibular right second molar) (Nolla 1960). The obtained ^{14}C results confirm the different timing of formation and

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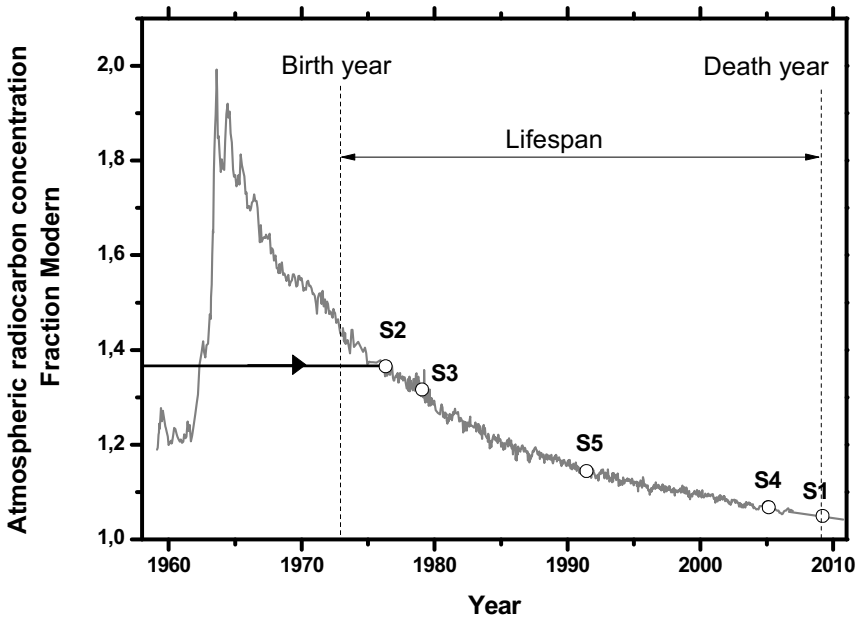


Figure 1 Bomb peak curve and measured ^{14}C concentrations for the different samples

are, again, consistent with the declining part of the bomb-peak curve. From these data, the date of birth can be estimated by subtracting the timing of formation from the measured ^{14}C age (Spalding et al. 2005). In this way, a birth year corresponding to 1971.5 ± 2.2 and 1973.1 ± 3.2 can be estimated by using the data obtained for samples S2 and S3, respectively, with the corresponding uncertainties calculated by taking into account the errors associated both with the calibrated time range and with the age of crown formation. A reduction of the uncertainty associated with the determination can be obtained by combining the birth ages calculated starting with the 2 teeth data, obtaining a weighted average of 1972.0 ± 1.8, which is consistent, at the 1σ level, with the known date of birth.

CONCLUSIONS

Different tissues extracted from a post-bomb human body were analyzed by ^{14}C AMS. Varying ^{14}C concentrations were obtained corresponding to the different formation/replacement rates of the analyzed tissues. The results indicate that tissues with a rapid turnover (hair) are indicative of the age of death. Concerning the analysis of bones, a significant offset of ~18 yr was found between the age of collagen extracted from the cortical bone and the year of death. Trabecular bones show a more rapid turnover with an age offset from the year of death of ~5 yr. Simultaneous analysis of cortical and trabecular bone can be used to establish if the analyzed samples fall on the rising or declining part of the bomb-spike curve. The results obtained on dental enamel extracted from teeth with different crown formation times confirm that this method can be effectively used to estimate the year of birth with uncertainties of ~2–3 yr. As a further step in this study, an investigation of possible different turnover rates for different bones in the body is planned.

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