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Dating human skeletal remains using ^{90}Sr and ^{210}Pb: Case studies

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ABSTRACT

In legal medicine, the post mortem interval (PMI) of interest covers the last 50 years. When only human skeletal remains are found, determining the PMI currently relies mostly on the experience of the forensic anthropologist, with few techniques available to help. Recently, several radiometric methods have been proposed to reveal PMI. For instance, ^{14}C and ^{90}Sr bomb pulse dating covers the last 60 years and give reliable PMI when teeth or bones are available. ^{232}Th series dating has also been proposed but requires a large amount of bones. In addition, ^{210}Pb dating is promising but is submitted to diagenesis and individual habits like smoking that must be handled carefully.

Here we determine PMI on 29 cases of forensic interest using ^{90}Sr bomb pulse. In 12 cases, ^{210}Pb dating was added to narrow the PMI interval. In addition, anthropological investigations were carried out on 15 cases to confront anthropological expertise to the radiometric method. Results show that 10 of the 29 cases can be discarded as having no forensic interest (PMI > 50 years) based only on the ^{90}Sr bomb pulse dating. For 10 other cases, the additional ^{210}Pb dating restricts the PMI uncertainty to a few years. In 15 cases, anthropological investigations corroborate the radiometric PMI. This study also shows that diagenesis and inter-individual difference in radionuclide uptake represent the main sources of uncertainty in the PMI determination using radiometric methods.

1. Introduction

To determine the post mortem interval (PMI) when only human skeletal remains are available is a difficult task in forensic sciences. In Switzerland, the statute of limitations in criminal law is 30 years. Nevertheless, for human and social reasons, forensic investigations on bones often cover a PMI of 50 years. Usually, the PMI determination relies on the skill and experience of forensic anthropologists, with few supportive techniques available [1–4]. The state of conservation of the remains depends strongly on the environment in which the body rested for years. Bodies kept for centuries or even a thousand years in glaciers or peat bogs are often excellently preserved due to freeze drying and a reductive environment, while oxidative soils are responsible for a rapid decomposition of tissues and skeletisation [5–7]. Among all the methods proposed to determine reliable PMI, those based on radionuclide determination are very promising. After death, a radionuclide incorporated in the body is only submitted to radionuclide decay, with a well-known half-life [8–10].

Several studies have proposed radionuclide analyses to determine PMI. While the half-life (5730 y) of ^{14}C is much too long to be used in forensic cases, Spalding et al. proposed determining the age at death and the nuclear bomb pulse ^{14}C in teeth to reveal the PMI [11,12]. Marzaioli et al. extended the technique to bone collagen and were able to precisely determine the PMI of an unsolved case investigated by the Rome prosecutor office [13]. However, the forensic determination of pulse bomb ^{14}C requires the use of acceleration mass spectrometry, a technique which is currently not easily available. Nevertheless bomb pulse ^{14}C dating, in combination with the determination of age at death, e.g. by the racemization method, is a promising method yielding PMI with low uncertainties. Another promising technique is based on the isotopic ratio of different radionuclides from the ^{232}Th disintegration series [14–16]. However, this method requires at least 300 g of bone, which can be a strong drawback to the application of the technique in forensic sciences. Moreover, the counting time of α and γ spectrometry involved in the measurement is at least 15 days to achieve the desired detection limit. Bomb pulse ^{90}Sr has

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also been proposed to determine PMI. The analytical technique is simpler than for bomb pulse ^{14}C but leads to larger uncertainties related to β counting [9,17]. Several authors have also reported on archeological bones having ^{90}Sr above the detection limit [9,17]. The presence of diagenetic ^{90}Sr is thus a serious disadvantage to the application of the technique to the PMI determination. Finally, ^{210}Pb is also a radionuclide of interest for PMI determination. Its physical half-life of 22 years covers the range of elapsed time required in legal investigations. Moreover, it is incorporated by ingestion and inhalation of ^{222}Rn decay products in every individual and it targets bones. In addition, it can be easily measured at very low levels using its granddaughter ^{210}Po, with which it reaches secular equilibrium about one year after death. Nevertheless, ^{210}Po diagenesis has also been observed in most of reported cases of buried skeletons [17,18].

Recently, we reported on ^{210}Po and ^{90}Sr diagenesis if the radiometric method involving both radionuclides is used for the dating of human skeletal remains [17]. We showed that ^{210}Po diagenesis can be handled satisfactorily using solubility profiling. Because of the quantity of bone ash required per analysis (5 g), ^{90}Sr solubility profiling is not recommended. Nevertheless satisfactory results are obtained by using a decontamination method involving Fe(III) reducing agent, metal-binding molecules such as EDTA and DCTA and hydroxyl-apatite abrasion by acetate buffer [17].

Here we report on the use of ^{90}Sr and ^{210}Po dating and anthropological investigations to determine the PMI in 29 cases of forensic interest. We focus on the diagenesis problem that has often been neglected in previous studies on radiometric methods to determine PMI. We compare ^{90}Sr and ^{210}Po dating with anthropological analysis for 15 cases to corroborate PMI. The limitation of the use of ^{90}Sr and ^{210}Po is also presented, with an attempt to estimate the uncertainty of the method related to the location of the remains, to dietary habits influencing ^{210}Pb and ^{90}Sr intake in the skeleton in a given region and to the analytical aspects of the method.

2. Materials and methods

2.1. Sampling

Most of the human skeletal remains were recovered in different locations in western Switzerland. For the most part they came from 30 to 50 km area close to Lausanne. Two cases were found in Lake Geneva, three in different forests, three in riverbanks, five in construction sites during digging, five in relation to glaciers and one in high alpine territory. The rest of the cases were from different places such as houses and bins. Four cases had a well-established PMI, based on forensic inquiry, before measurements. Glacier cases are particularly important in Switzerland because the discovery of human remains in glaciers increased recently due to climate warming and because the diagenesis of ^{90}Sr in glacier cases is also enhanced (see Section 4).

Vertebrae of individuals who died between 2006 and 2012 have been sampled in the pathology Institutes of Lausanne and Locarno, Switzerland. Ribs were obtained from the Institut Médico-Légal of Lyon (France). These samples were used to determine the ^{210}Po reference value at the time of death for people aged 18–90 years ($n = 35$). They do not contain diagenetic ^{210}Po because they were not buried before sampling.

To probe the effect of tobacco smoke on ^{210}Po urine excretion and its possible effect on the ^{210}Po content of the bones, we collected urine from non-smoker collaborators of the Institute of Radiation Physics and one sample from a heavy smoker collaborator. 300 ml of urine were spiked with ^{209}Po and 100 ml of HNO_3 65% was added. The urine was mineralized in a large volume microwave apparatus (Milestone MLS Ultraclave IV, Germany) under pressure (initial pressure = 60 bars) for 30 min at 180 °C. Afterwards, polonium was co-precipitated on iron hydroxides. The precipitate was dissolved in 80 ml HCl 1 M, ascorbic acid (500 mg) was added and polonium was spontaneously deposited on a silver disk at 50–60 °C for at least 4 h. The alpha spectra were then recorded as described in [17].

Table 1
^{90}Sr and ^{210}Po activities (Bq/g Ca) of human skeletal remains found between 2001 and 2012 in different locations in Switzerland.

Case number	Discovery location	^{90}Sr (Bq/g Ca)	^{210}Po (Bq/g Ca)	Bone type
GRE-12-439	Alpine forest	0.020 ± 0.006	0.025 ± 0.005[b]	Trabecular bone (vertebrae)
GRE-12-415	High alpine glacier	0.019 ± 0.002	0.005 ± 0.001[c]	Trabecular bone (calcaneum)
GRE-12-414	High alpine, glacier	0.032 ± 0.003	0.005 ± 0.001 [c]	Trabecular bone (calcaneum)
GRE-12-413	High alpine, glacier	0.012 ± 0.001	0.002 ± 0.0005 [c]	Trabecular bone (calcaneum)
GRE-12-413/414/415	High alpine glacier, mixture of the three samples	<0.005[a]		Trabecular bone (calcaneum)
GRE-12-417	Lowland lake	<0.005	0.003 ± 0.001[b]	Trabecular bone (femur head)
GRE-11-363	Lowland, bed of a river	0.039 ± 0.007	0.009 ± 0.001[c]	Trabecular bone (femur head
GRE-11-320	Lowland, construction site	<0.008		Trabecular bone (vertebrae)
GRE-11-163	Alpine deep valley, rocky slope	0.019 ± 0.004	0.005 ± 0.002[b]	Cortical bone (femur)
GRE-11-162	Alpine deep valley, rocky slope	0.019 ± 0.003	0.007 ± 0.003[b]	Trabecular bone (vertebrae)
GRE-11-064	Inside, garbage bag	<0.005		Trabecular bone (vertebrae)
GRE-10-285	Sub-alpine, gravel pit	0.032 ± 0.002		Trabecular bone (jaw)
GRE-10-278	Lowland, construction site	0.032 ± 0.004		Trabecular bone (vertebrae)
GRE-10-019	Lowland, lake (fishing net)	<0.008	<0.015[c]	Cortical bone (femur)
GRE-10-204	Lowland, under 20 cm of soil	<0.005		Trabecular bone (vertebrae)
GRE-09-307	Lowland, close to ancient cemetery	0.014 ± 0.004	0.015 ± 0.003[c]	Cortical bone (skull)
GRE-09-306	Lowland, forest	0.067 ± 0.005	0.021 ± 0.003[b]	Trabecular bone (vertebrae)
GRE-08-199	Lowland, construction site	0.032 ± 0.004	0.020 ± 0.003[a]	Trabecular bone (iliac crest)
		0.028 ± 0.004		Trabecular bone (vertebrae)
GRE-08-018	High alpine, rocky slope	0.036 ± 0.005	0.008 ± 0.004	Cortical (femur)
		0.055 ± 0.005	0.050 ± 0.010	Trabecular bone (iliac crest)
GRE-07-162	Lowland, construction site	<0.005		Trabecular bone (internal ear)
GRE-07-063	Lowland, construction site	0.014 ± 0.003		Trabecular bone (vertebrae)
		<0.005		Cortical bone (skull)
GRE-07-001	Lowland, in concrete behind a kitchen wall	<0.005		Cortical bone (skull)
GRE-06-111	Lowland, bank of à river	0.008 ± 0.003		Trabecualr bone (skull)
		0.005 ± 0.003		Cortical bone (skull)
GRE-05-325	High alpine glacier	0.098 ± 0.006		Trabecular bone (iliac crest)
GRE-05-191	Alpine, inside in a box	<0.005		Trabecular bone (skull)
GRE-05-072	Lowland, on top of accumulated leaves	0.012 ± 0.004		Cortical bone (skull)
GRE-04-203	Lowland, lake, in a fish trap	<0.005		Cortical bone (skull)
GRE-04-085	Lowland, small river	0.008 ± 0.002		Cortical bone (skull)
GRE-01-256	High alpine glacier	0.107 ± 0.003		Cortical bone (femur)
GRE-01-155	Lowland, forest	0.010 ± 0.002		Cortical bone (skull)

[a] After additional decontamination steps involving iron(III)-reduction and EDTA washing, followed by acetate buffer at pH 4.5 washings.
[b] Target value after solubility profile.
[c] Decontamination by solubility profiling, only fraction corresponding to fraction 5 + 6 analyzed for ^{210}Po.

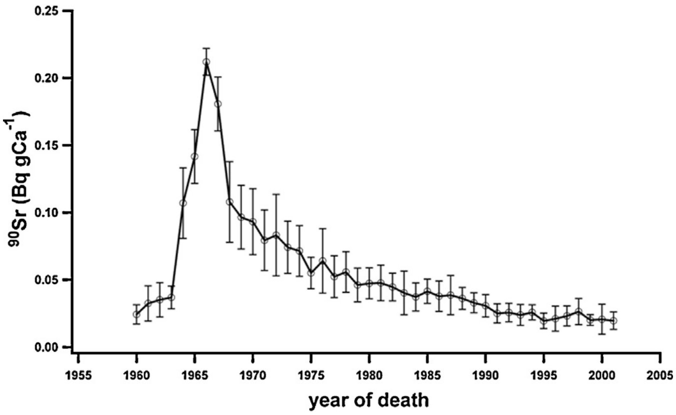

Fig. 1. Calibration curve for activity of ^{90}Sr in vertebrae based on measurements carried out on bones sampled at autopsy of individuals who died in Switzerland between 1960 and 2001. Uncertainty given for $k = 2$.

The calibration curve of bomb pulse ^{90}Sr for Switzerland has been determined previously as described in [17,19]. The samples are from individuals aged between 20 and 92 years old at the time of death. Based on 5 g of bone ash introduced in the analysis, all samples have a ^{90}Sr activity above the detection limit of 5 mBq/g Ca.

2.2. ^{90}Sr and ^{210}Po determination

The method of ^{210}Po and ^{90}Sr determination has been described in detail in [17,19]. Because our measurements from forensic cases cover the last ten years of human remains findings, different protocols of decontamination have been used, according to the state of the art and our own results. Briefly, at the beginning of the 2000s, only basic decontamination steps such as ultrasonication in water were used to remove adhering soil particles. Afterwards, EDTA and DCTA washings were introduced to decontaminate bones from metal species. Furthermore, reduction of iron hydroxide particles to soluble iron(II) with hydroxylamine 1 M was introduced to remove many interfering ions adsorbed on adhering iron hydroxides particles. At the end of the 2000s, we introduced acetate buffer washings (^{90}Sr) and acetate buffer solubility profiling (^{210}Po) to suppress diagenesis. Currently, a sequence including all of these decontamination steps is used to avoid diagenetic ^{90}Sr and ^{210}Po.

3. Results

The results of ^{90}Sr bomb pulse dating are presented for 29 cases in Table 1. The uncertainty on the ^{90}Sr measurement was between 10% and 20% ($k = 2$). Eleven cases showed activity below the detection limit. These cases are therefore not relevant to any forensic investigation because the time of death is prior to 1955, when no ^{90}Sr was present in the environment and food. When ^{90}Sr activity in bones was above the detection limit, the result was compared to the calibration curve determined previously and presented in Fig. 1. One can see that ^{90}Sr activities in vertebrae have rapidly followed the changes appearing in environmental ^{90}Sr [17,19], reflecting a high bone turnover in the trabecuular fraction. Note that the ^{90}Sr activities are presented as a function of the year of death, as an average of at least 12 measurements of individuals who died in Switzerland. Thus these activities must be recalculated for every year of measurement of the skeletal remains using 28.8 years as the physical half-life of ^{90}Sr before comparison. Results of Table 1 show that 3 cases were compatible with a time of death corresponding to the highest ^{90}Sr activity in bones (0.067–1.07 Bq/gCa), yielding a period of death close to 1963. The results of ^{210}Pb

Table 2
Estimation of the time of death based on the ^{90}Sr and ^{210}Po dating method for 29 cases of human skeletal remains found between 2001 and 2012 in different location in Switzerland.

Case number	Discovery location	^{90}Sr (Bq/g Ca)	^{210}Po (Bq/g Ca)	Estimated time of death
GRE-12-439	Alpine forest	0.020 ± 0.006	0.025 ± 0.005[a]	After 1990
GRE-12-415	High alpine glacier	0.019 ± 0.002	0.005 ± 0.001[b]	Before 1940
GRE-12-414	High alpine, glacier	0.032 ± 0.003	0.005 ± 0.001[b]	Before 1940
GRE-12-413	High alpine, glacier	0.012 ± 0.001	0.002 ± 0.0005[b]	Before 1940
GRE-12-413/414/415	High alpine glacier, mixture of the three samples	<0.005[c]		Before 1940
GRE-12-417	Lowland lake	<0.005	0.003 ± 0.001[a]	Before 1940
GRE-11-363	Lowland, bed of a river	0.039 ± 0.007	0.009 ± 0.001[b]	After 1970
GRE-11-320	Lowland, construction site	<0.008		Before 1955
GRE-11-163	Alpine deep valley, rocky slope	0.019 ± 0.004	0.005 ± 0.002[a,d]	After 1955, before 1970
GRE-11-162	Alpine deep valley, rocky slope	0.019 ± 0.003	0.007 ± 0.003[a,d]	After 1955, before 1970
GRE-11-064	Inside, garbage bag	<0.005		Before 1955
GRE-10-285	Sub-alpine, gravel pit	0.032 ± 0.002		
GRE-10-278	Lowland, construction site	0.032 ± 0.004		
GRE-10-019	Lowland, lake (fishing net	<0.008	<0.015[b]	Before 1955
GRE-10-204	Lowland, under 20 cm of soil	<0.005		Before 1955
GRE-09-307	Lowland, closed to ancient cemetery	0.014 ± 0.004	0.015 ± 0.003[b]	After 1955, before 1980
GRE-09-306	Lowland, forest	0.067 ± 0.005	0.021 ± 0.003[b]	Close to 1965
GRE-08-199	Lowland, construction site	0.032 ± 0.004	0.020 ± 0.003[a]	After 1955, before 1963
		0.028 ± 0.004		
GRE-08-018	High alpine, rocky slope	0.036 ± 0.005	0.008 ± 0.004[b]	After 1955, before 1975
		0.055 ± 0.005	0.050 ± 0.010[b]	
GRE-07-162	Lowland, construction site	<0.005		Before 1955
GRE-07-063	Lowland, construction site	0.014 ± 0.003		After 1950, before 1963
		<0.005		
GRE-07-001	Lowland, in concrete behind a kitchen wall	<0.005		Before 1955
GRE-06-111	Lowland, bank of à river	0.008 ± 0.003		After 1950, before 1963
		0.005 ± 0.003		
GRE-05-325	High alpine glacier	0.098 ± 0.006		Close to 1965
GRE-05-191	Alpine, inside in a box	<0.005		Before 1955
GRE-05-072	Lowland, on top of accumulated leaves	0.012 ± 0.004		1990-2000
GRE-04-203	Lowland, lake, in a fish trap	<0.005		Before 1955
GRE-04-085	Lowland, small river	0.008 ± 0.002		After 1955, before 1965
GRE-01-256	High alpine glacier	0.107 ± 0.003		Close to 1965
GRE-01-155	Lowland, forest	0.010 ± 0.002		After 1955

[a] Target value after solubility profile.
[b] Decontamination by solubility profiling, only fraction 5 + 6 analyzed for ^{210}Po.
[c] After additional decontamination steps.
[d] Not buried, submitted to weathering.

Table 3
Anthropological estimates (taphonomy of time since death and ^{90}Sr and ^{210}Po activity (Bq/g Ca).

Case number	^{90}Sr (Bq/g Ca)	^{210}Po (Bq/g Ca)	Taphonomy
GRE-12-439	0.020 ± 0.006	0.025 ± 0.005	Adipocere, preserved hairs and textiles, partly bleached by weathering, not covered by algae or moss
GRE-12-415	0.019 ± 0.002		By texture and color: ancient
GRE-12-414	0.032 ± 0.003		By texture and color: ancient
GRE-12-413	0.012 ± 0.001		By texture and color: ancient
GRE-12-417	<0.005	0.003 ± 0.001	Signs of a certain period of contact with water, by texture and color: ancient
GRE-11-363	0.039 ± 0.007	0.009 ± 0.001	Signs of a certain period of contact with water, grease saturated, adipocere, cartilage preservation
GRE-11-320	<0.008		By texture and color: ancient
GRE-11-064	<0.005		By texture and color: ancient
GRE-10-285	0.032 ± 0.002[a]		By texture and color: ancient
GRE-10-278	0.032 ± 0.004[a]		By texture and color: ancient
GRE-10-019	<0.008	<0.015	Signs of a certain period of contact with water; by texture and color: ancient
GRE-10-204	<0.005		By texture and color: ancient
GER-09-307	0.014 ± 0.004	0.015 ± 0.003	Covered by plants, exposed to weathering
GRE-09-306	0.067 ± 0.005	0.021 ± 0.003	Covered by plants, exposed to weathering
GRE-08-18	0.036 ± 0.005	0.008 ± 0.004	Adipocere, grease saturated, soft tissue preservation/mummification, partly exposed to weathering, not covered by algae or moss
	0.055 ± 0.005	0.050 ± 0.010	

[a] Strong suspicion of diagenesis, based on a very low loss of mass on ignition, presence of high level of insoluble (HCl 1M) material and general aspect of fossilization.

dating for 12 cases are presented in Table 1. The uncertainty on the ^{210}Po measurement was between 10% and 20% ($k = 2$). Half of the cases had ^{210}Po activity below 15 mBq/g Ca. These values were compatible with a PMI longer than 30 years, with limitations presented in Section 4. Table 2 presents an estimation of the period of death based on ^{90}Sr and ^{210}Po dating. The results of anthropological investigations are presented in Table 3. All these results aimed to determine the PMI with a high level of confidence. In all 29 cases, a reliable PMI or a PMI limit (e.g. before 1955) was attributed (Table 2), either by ^{90}Sr bomb pulse dating alone, or in combination with ^{210}Po dating, or still with the addition of anthropological investigation.

The results of the determination of ^{210}Po at the time of death are presented in Fig. 2. The activities cover a wide range of values, from 15 to 97 mBq/gCa. The activities of the 14 ribs do not differ significantly from the activities of the 21 vertebrae and average value is 31 ± 22 mBq/g Ca ($k = 1$). Finally, we measured ^{210}Po in urine sampled from non smoker and one heavy smoker collaborators of the Institut of Radiation physic to probe the effect of smoking on the incorporation of ^{210}Pb/^{210}Po. Results showed that the non smokers have ^{210}Po activity in urine below 5 mBq/l while the heavy smoker has ^{210}Po activity in urine of 15 mBq/l (Fig. 4). This increase in ^{210}Po urine excretion is a result of an increase in incorporation of ^{210}Pb/^{210}Po that may also be reflected in ^{210}Pb/^{210}Po activity in bones.

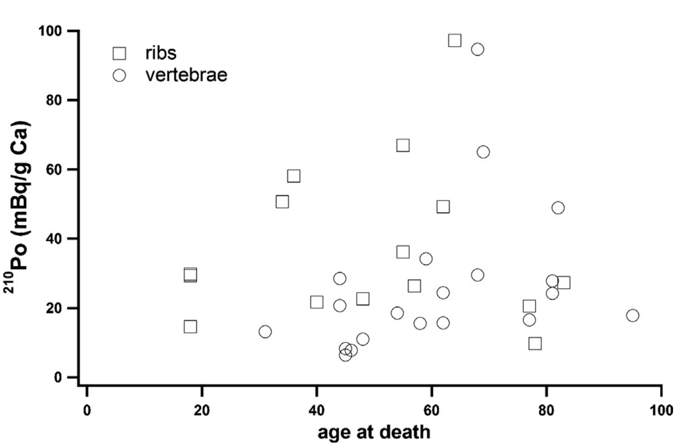

Fig. 2. Activity of ^{210}Po (Bq/g Ca) based on measurements carried out on bones sampled at autopsy of individuals who died in Switzerland (vertebrae) or were autopsied in Lyon (ribs) between 2006 and 2011 (reference date 2011).

4. Discussion

The results of our study suggest that bomb pulse ^{90}Sr dating in combination with ^{210}Pb dating (through ^{210}Po measurement) can be used to reveal PMI as long as severe precautions are taken to avoid ^{90}Sr and ^{210}Po diagenesis. This was confirmed by the anthropological investigations, which gave a similar PMI to the radiometric method in all studied cases. Being able to reject all cases with ^{90}Sr activity below the detection limit of 0.005 Bq/g Ca as being of no forensic interest because of a time of death before the nuclear era represents the major advantage of using the radiometric method in forensic science. This rather simple measure will prevent a tremendous amount of forensic work, e.g. extraction of DNA. In our study, one third of all cases were archeological and not legal matters and were then rejected on the basis of ^{90}Sr dating alone. Nevertheless, it is important to note that finding ^{90}Sr above the detection limit may mislead the final PMI attribution because of two drawbacks: (i) ^{90}Sr can come from diagenesis and (ii) ^{90}Sr bomb pulse, while not symmetric (Fig. 1), has an upslope and downslope to the curve with similar activity for very different years of death.

The remains of three skeletons (cases # GRE-12-415; GRE-12-414 and GRE-12-413) found in 2012 on a Swiss glacier illustrate the first problem. Formally identified, they have been missing before 1940. Their bones should thus contain no ^{90}Sr (death before nuclear era) and should have a low ^{210}Po activity. The bones (calcaneum) received in the laboratory for radiometric dating were well preserved, with an organic matter lost on ignition of 50%. After decontamination (see [17] for details) ^{90}Sr was nevertheless found above the detection limit. This ^{90}Sr was obviously of a diagenetic origin. On the other hand, ^{210}Po activity was very low in the three bones, indicating a long PMI. This finding showed that skeletal remains from glaciers could be mixed with the ice and snow layers formed during the beginning of the sixties, when ^{90}Sr atmospheric fallout was at its highest. We measured ^{90}Sr again, adding several steps of hydroxylamine and acetate buffer washings; ^{90}Sr activity dropped below the detection limit. The case of skeletal remains found on another Swiss glacier (case # GRE-01-256) posed a similar problem; the ^{90}Sr activity was 1.07 Bq/g Ca, obtained after a very basic decontamination using ultrasonication in water that was in use in 2001, when the measurement was carried out. Based on our calibration curve of Fig. 1, the time of death should be close to 1962. However, several objects found in proximity of the remains were from the end of the 19th century, including coins from the 1880s which were no longer in use in the sixties. In this respect the relics were of archeological interest. Here again, the remains have probably been mixed in with the ice layer from the sixties due to melting, leading to ^{90}Sr diagenesis. The five cases of glacier remains showed the highest activities (e.g. case # GRE-01-256), with possibilities of diagenesis. This leads us to determine that when a positive value is observed, it should always be confirmed by a second measurement, after additional decontamination steps. When possible, ^{210}Pb dating should be added, preferably as solubility profiling, as in case # GRE-08-018. The solubility profiling method is based on the difference in solubility of the apatite of biogenic and diagenetic origin. The first washings with acetate buffer at pH 4.5 will dissolve less crystallized apatite from diagenetic origin [20]. Further washings will contain biogenic ^{210}Pb/^{210}Po [17]. In the cases of the three skeletons found on a glacier, biogenic ^{210}Po confirmed a long PMI, with ^{210}Po activities less than 5 mBq/g Ca.

Cases # 11-162/163 illustrate the limitation of ^{210}Po dating when the human remains are not buried and therefore submitted to weathering. It also illustrates the interest of conducting ^{90}Sr and ^{210}Po measurements in trabecular and cortical bone, if both are recovered. The remains from this case were found not buried, at about 1100 m above sea level, in April 2011. Formally identified, the forensic investigations established that the individual had been missing since 2008. The analyzed bones were completely bleached, with a very low content in organic matter (loss on ignition $< 10\%$). Results show that vertebrae and femur have the same ^{90}Sr activity of 0.019 Bq/g Ca. This activity in vertebrae is expected for a time of death close to 1960 or after 2000. Thus we carried out a ^{210}Po solubility profile to get a better estimate of the PMI. The solubility profile (Fig. 3) reveals no diagenetic ^{210}Po (stable values) and ^{210}Po activity below 10 mBq/g Ca. This low value of ^{210}Po alone yields a long PMI, not compatible with the forensic investigation. It is then possible that weathering, with heavy rains and snow melting, would have leached the ^{210}Pb out of the bone structure. As a matter of fact, the Ca content of the bone ash in this case was significantly higher (0.40 g/g ash) than for bone ash in our database (0.33 ± 0.04 g Ca/g ash, $n = 310$). This might express a loss of biogenic lead phosphate where lead replaces Ca in the apatite structure. In addition, weathering might have leached ^{210}Po preferentially because of the local destruction of the crystalline lattice due to nucleus recoil when ^{210}Pb and ^{210}Bi, grandmother and mother of ^{210}Po, respectively, disintegrate, as observed in the radioactive disequilibrium between ^{238}U and ^{234}U in water [21].

Often, human skeletal remains consist of only the skull and/or long bone diaphyses. On the other hand, trabecular bones found in vertebrae, ribs, etc. are less often preserved. In this case, our problem was to extend results established for trabecular bone to compact bone. Compact bone takes up ^{90}Sr and ^{210}Pb in food at the time of bone formation. In contrast, trabecular bone takes up ^{90}Sr and ^{210}Pb in food at the time of death, because of a higher bone remodeling rate. In our study, seven cases consisted only of the skull. Among them, three showed ^{90}Sr activities below the detection limit and were discarded as archeological cases. The presence of ^{90}Sr in the 4 remaining cases demonstrates that the time of death is posterior to 1955. It was nevertheless impossible to give a more precise time of death because we do not have an established calibration curve for this kind of bone. Higher ^{90}Sr activity in the skull compared to vertebrae is expected for a recent time of death only if the skull was forming during the sixties, where food contamination was at its highest. Thus it is necessary to turn to the anthropologist evaluation of the age at death to improve the determination of the time of death. For instance, human skeletal remains found in 2010 from an individual older than 60 years and who died recently should contain only traces of ^{90}Sr because the skull was already formed during the middle of the sixties, when food contamination was high. ^{210}Po/^{210}Pb dating

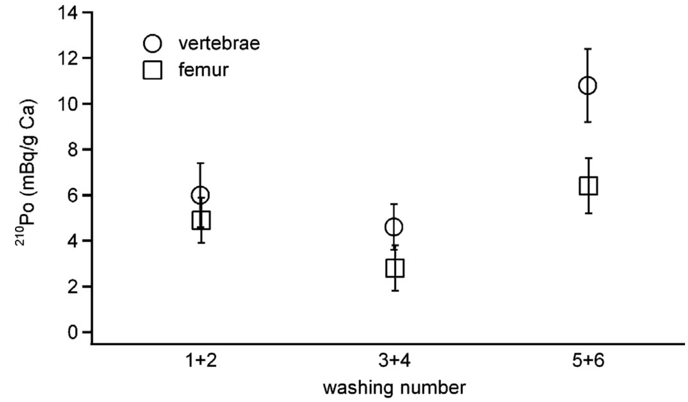

Fig. 3. Solubility profile of ^{210}Po in vertebrae (circle) and femur (square) from the #GRE-11-162 and GRE-11-163 cases.

should indicate a possible time of death but was carried out here only for the GRE-09-307 case. The activity of ^{210}Po was 0.015 Bq/g Ca. This activity is significantly lower than expected for a short PMI. Consequently we attributed a period of death after 1955 (presence of ^{90}Sr) and before 1980 (intermediate ^{210}Po value).

The uncertainties in the ^{90}Sr bomb pulse and ^{210}Pb dating are mostly the consequence of a large uncertainty on the radionuclide measurement, to the possibility of diagenesis and to the differences in individual ^{90}Sr and ^{210}Pb uptake. For both ^{90}Sr and ^{210}Po measurements at the encountered activities in human, the uncertainty ($k = 2$) is above 10% and can reach 20%. The contamination of food with ^{90}Sr can differ depending on the location and dietary habits; for instance, ^{90}Sr activity in dairy food (milk and cheese) is proportional to the altitude of cows grazing, with higher activities measured in Alpine milk than in milk from lowland regions of Europe [22]. Thus in Switzerland, where people are living between 300 and 2000 m above sea level, the contamination of food, reflected in the vertebrae, may significantly differ, depending on the living area. This will add uncertainty in estimating PMI based only on the ^{90}Sr activity found in vertebrae. In fact, our ^{90}Sr calibration curve of Fig. 1 has been established by measuring vertebrae of people having lived in the Swiss lowland (mostly Lausanne, Zürich, Basel and Lugano regions). In addition, to compare the ^{90}Sr activity found in human remains to the ones of Fig. 1 requires the activity equilibrium between vertebrae and local food, which will take about two years to establish [19]. In a country with a high immigration rate such as Switzerland, this condition may not be totally fulfilled.

To estimate the PMI based on ^{210}Pb dating requires the knowledge of the ^{210}Pb level at the time of death. The results of 35 measurements show a large dispersion of values (min = 6.4 mBq/gCa; max = 97 mBq/gCa). The average value is 31 mBq/g Ca. The standard deviation is 22 mBq/g Ca. Thus, the ^{210}Pb content in human ribs and vertebrae varies greatly at the time of death between individuals. In a previous study, we found a target value of 45 ± 11 mBq/g Ca in vertebrae of individuals who died in 1990, excavated and measured in 2008. Recalculated to the reference date 2008, the mean activity at the time of death would be 70 mBq/g Ca, about twice the activity found in this work. So it seems that the target value at death determined in [17] is over-evaluated, possibly because the samples came from buried bodies. On the other hand, the samples in [17] were excavated from a Milanese cemetery and might illustrate differences in the place they lived, medications, age at death, and smoking and dietary habits. Fig. 2 also shows that 7 values are below 20 mBq/g Ca at the time of death. This will cause a large over-estimation of PMI based on ^{210}Pb dating for these individuals. On the other hand, two cases have values close to 90 mBq/g Ca, causing an under-estimation of PMI, should we encounter these cases in forensic

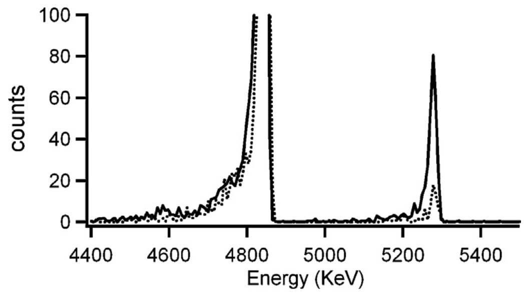

Fig. 4. ^{209}Po tracer (left peak) and ^{210}Po (right peak) spectra of urine sampled from non-smoker collaborators of the Institute of Radiation Physics (dotted line) and a heavy smoker collaborator of the Institute of Radiation Physics (black line).

evaluation. The main cause of the large inter-individual difference in the ^{210}Po activity at the time of death also probably reflects a smoking habit. Tobacco fumes are known to contain both ^{210}Pb and ^{210}Po and to significantly increase ^{210}Po excretion (Fig. 4). Thus heavy smokers are expected to have more ^{210}Pb–^{210}Po in their bones than non-smokers. In this respect, the two cases with ^{210}Po activity close to 90 mBq/g Ca at the time of the death might well have been heavy smokers.

5. Conclusions

Here we show that a third of the cases can be discarded as "of archeological interest" using ^{90}Sr determination only. This fact alone saves a lot of forensic work. In addition, the radiometric measurements yield hard data that do not rely on the investigator. In spite of large uncertainties on the model due to differences between individuals in ^{210}Pb and ^{90}Sr uptake, these hard data provide estimates which can be used to refine anthropological hints. We conclude that ^{90}Sr and ^{210}Po dating, if carried out with the necessary precautions discussed above, can be used to determine PMI. Nevertheless, anthropological investigations should always be carried out to confirm the radiometric estimate. Eventually, our study demonstrates that ^{90}Sr and ^{210}Pb dating should be carried out together to increase the power of the radiometric estimate, particularly because these two radionuclides are not submitted to the same degree of diagenesis, depending on the burial site.

Nevertheless, the method suffers drawbacks that the investigators must keep in mind; once the diagenetic problem is resolved, weathering can leach ^{210}Po from bones, yielding to unrealistic long PMI. In addition, heavy smoking habits will yield too short PMI, if based on ^{210}Po dating only. In this respect, cases of forensic interest presenting the above-described limitations could be submitted to the more precise but less available ^{14}C-AMS bomb pulse dating.

As a final remark, we propose that all the activities reported for radionuclides in human bone are normalized to the calcium content. We strongly think that a normalization to the bone wet or dry weight, as often reported in the literature, is not the right way to report activities in bones because the level of skeletisation (lost of organic matter) is not known. This is particularly true for human remains. In addition, calcium diagenesis is not susceptible to significantly change the calcium content in bone apatite.

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