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Determination of strontium-90 in deer bones by liquid scintillation spectrometry after separation on Sr-specific ion exchange columns

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Abstract

The activity concentration of ^{90}Sr was determined in several deer bones from Austria. Strontium specific ion exchange columns with 4′,4″(5″)-di-t-butylcyclohexane-18-crown-6 from Eichrom Industries, Inc. were used for separation. The yield of the chemical procedure was quantified with AAS. Directly after column separation, the solution containing ^{90}Sr was mixed with the scintillation cocktail HiSafe III and measured by liquid scintillation counting. Prevention of ^{210}Pb contamination and reusability of the separation columns was investigated as well as the activity distribution within the bones. Results were compared with pre-Chernobyl measurements in Austria; a correlation between activity concentration of ^{90}Sr and site altitude was found.

Keywords: ^{90}Sr; Deer bones; Sr-specific ion exchange columns; Liquid scintillation counting

1. Introduction

The determination of ^{90}Sr activity levels in environmental samples has been of considerable interest since the time of the atmospheric nuclear weapons tests in the late 1950s and early

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1960s and again after the reactor accident at Chernobyl in 1986. ^{90}Sr is a beta-emitter with a maximum β-energy of 0.5 MeV and a half-life of 28.5 years; it is chemically similar to calcium and therefore has a high transfer rate to the skeleton (Hardy et al., 1968). Due to its relatively long biological half-life in bones of about 50 years and because of its shortlived daughter ^{90}Y (half-life 64.1 h), which emits hard beta-particles with a maximum energy of 2.3 MeV, it can cause severe damage to bone and bone marrow. So, from the point of view of radiation protection, it is very important to control ^{90}Sr in the environment and in food.

Generally, deer, roe deer and reindeer as well as elk are often used as biomonitors of ^{90}Sr (Strandberg and Strandgaard, 1995; Tiller and Poston, 1999; Klevezal et al., 2001; Mietelski et al., 2001). Also in Austria, several investigations on ^{90}Sr in samples collected from the environment have been carried out: roe deer as well as snails were investigated by Tatzber et al. (1982), Tatzber and Irlweck (1984), and Schönhofer et al. (1994) measured the ^{90}Sr concentration in antlers of red deer retrospectively; Irlweck et al. (1999) determined ^{90}Sr levels of spruce needles after the Chernobyl accident. In our paper we want to show the recent environmental ^{90}Sr contamination by investigating bones of roe deer from different regions in Austria in comparison with the literature cited above.

To circumvent the time-consuming classical method for ^{90}Sr determination with fuming nitric acid for Sr separation (IAEA, 1989), we used the commercially available Sr·Spec® resin in prepacked columns. The elution containing ^{90}Sr and the inactive Sr carrier was mixed with a scintillation cocktail and measured by liquid scintillation counting (LSC); the chemical recovery of the Sr carrier was determined on aliquots by atomic absorption spectrometry (AAS). This yield determination is superior to the ''old'' gravimetric method, which was often erroneous due to low Sr carrier mass (as is required for sample preparation) and coprecipitated Ca (Bossew et al., 2000).

Horwitz and his co-workers have shown that strontium may be efficiently extracted from nitric acid by a solution of 4′,4″(5″)-di-t-butylcyclohexane-18-crown-6 in 1-octanol sorbed on an inert substrate (Horwitz et al., 1991). Nowadays this reagent is available commercially as Sr·Spec® resin in prepacked columns (2 ml of resin) from Eichrom Industries, Inc.; ^{90}Sr separation from a large variety of samples like food, milk, soil, etc. by using this resin is reported in the literature (see e.g. Jeter and Grob, 1994; Alvarez et al., 1995; Filss et al., 1998; Ware et al., 1998; Forsberg and Strandmark, 2001; Saxen, 2002; Brun et al., 2002). The solutions containing the samples as well as all solutions used for rinsing the columns and for eluting the purified Sr must be saturated with 1-octanol in order to preserve the octanol-layer in the column.

If the sample is taken up in an 8 M nitric acid solution, Sr·Spec® very effectively removes Ba and K isotopes as well as other matrix interferences. The columns show a very high affinity for strontium relative to calcium; however, the columns are reported to show breakthrough of strontium when loading more than 600–700 mg of calcium (Kwakman et al., 1996). The maximum strontium load of the columns is given with 17.8 mg Sr, but loadings of no more than 10–20% of capacity are recommended by the producer; this must be considered when fixing the amount of inactive strontium carrier used for chemical yield calculation.

Not only strontium, but also lead is retained by Sr·Spec® from nitric acid solutions (Dietz et al., 1991; Horwitz et al., 1992). In bone samples a certain amount of ^{210}Pb ($E_{\beta,\max} = 63$ keV (20%) and 16.5 keV (80%)) can be expected; its peak—easily recognizable by its energy and shape—was visible in some of our Sr spectra and had to be corrected numerically.

The aim of this work was to get information about the ^{90}Sr contamination of the natural environment by using deer bones as biomonitors. Additionally, we tried to improve the Sr·Spec®

method by searching for a procedure to get pure ^{90}Sr spectra without any ^{210}Pb contamination; different washing solutions and elutants were investigated. Also the reusability of the separation columns was investigated and found to be considerably better than cited in the literature (Brun et al., 2002). After a closer look at the activity distribution within the bones some environmental aspects will be discussed.

2. Experimental

The roe deer investigated was hunted in the years 2001 and 2002. The bones were stored deep-freezed; after unfreezing the metatarsals were sawed into three pieces: the middle part B of the bone is the tubular bone, A and C are the end parts. After ashing for 17–22 h at 450 °C, these samples were ground and weighed.

One gram of bone ash was dissolved in 5 ml of 8 M HNO_3 (saturated with l-octanol, as cited above) and 2.5 mg of Sr^{2+} carrier were added; the natural Sr content of the respective bone sample was measured in an aliquot by atomic absorption spectrometry (AAS) after generating a calibration line from a blank and five standard solutions. The sample solution was filtered; the beaker as well as the filter was washed with a few millilitres of 8 M HNO_3. The Sr·Spec® column was conditioned with 5 ml of 8 M HNO_3, then the sample solution was loaded onto it and afterwards the column was rinsed successively with 5 ml of 8 M HNO_3, 10 ml of 3 M HNO_3 (elution of Ca) and 1.5 ml of H_2O. We investigated three different elution procedures in order to find out the best procedure to obtain pure ^{90}Sr spectra without any ^{210}Pb contamination:

(1) Sr was eluted with 8.25 ml of distilled water as proposed by Kwakman et al. (1996) for milk samples;
(2) rinsing with 3 M HNO_3 was substituted by rinsing with 3 M $HNO_3 + 0.05$ M $(COOH)_2 \cdot 2H_2O$ to wash the lead from the column; elution of Sr with distilled water as in (1);
(3) Sr was eluted with 8.25 ml of 0.05 M HNO_3 as recommended by Dietz et al. (1991); here complete stripping of Sr and minimal removal of Pb from the column is expected.

Then 0.25 ml of the respective elution was used for recovery determination by AAS, and 8 ml were mixed with 12 ml HiSafe III® scintillation cocktail, cooled for 15 min and then immediately measured by liquid scintillation counting (LSC) using the ultra low-level counter Quantulus 1220 (Wallac Oy, Finland). This means that only ^{90}Sr was measured; the small amount of ingrowing ^{90}Y (only a few % within a counting time of 400 min and visible in the high energy part of the spectrum) was subtracted from the ^{90}Sr count rate. For control, the ingrown ^{90}Y was measured only in a few samples after an appropriate storage time to reach radioactive equilibrium; its activity concentration was found to equal that of ^{90}Sr within the error bars indicating pure ^{90}Sr spectra in the first run. Spectra showing also ^{210}Pb had to be corrected numerically by subtracting the ^{210}Pb peak. Fig. 1 shows the spectra of a pure ^{90}Sr sample and a ^{90}Sr sample contaminated with ^{210}Pb. The ^{210}Pb peak is relatively narrow, as it is not a pure β-peak but also consists of electrons from the highly converted γ-decay (46.5 keV); 100 decays of ^{210}Pb will generate 100 betas plus 110 conversion electrons and Auger electrons (Table de Radionucleides, 1983), so with a counting efficiency near to 50% a total efficiency of about 100% is achieved. We did not correct for the ingrowing ^{210}Bi as its contribution of 4% of the ^{210}Pb activity for our longest counting interval of 400 min directly after separation was assumed to be negligible compared with the much higher ^{90}Sr activities.

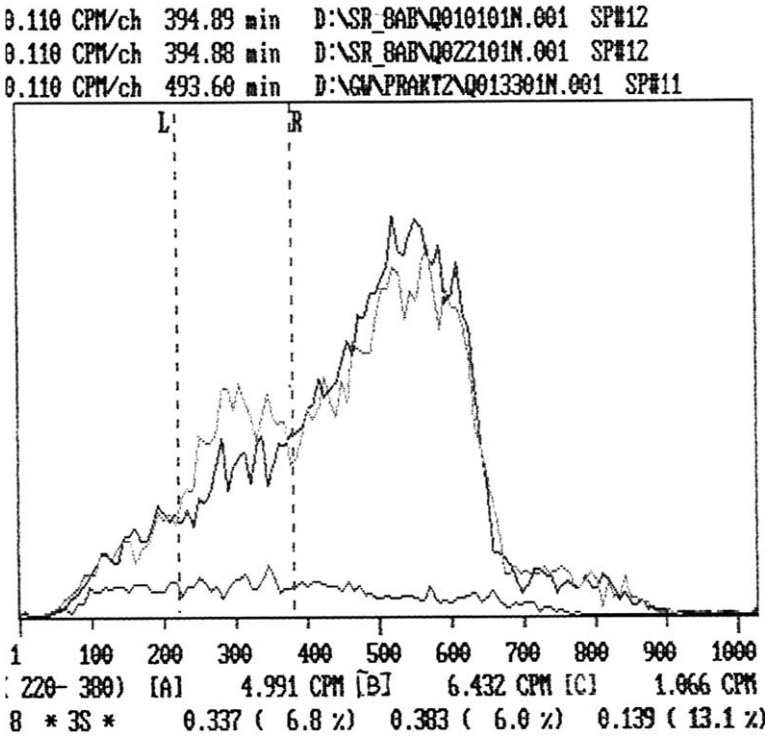

Fig. 1. LSC spectra of bone samples. (A) Spectrum of a pure ^{90}Sr sample; (B) spectrum of a ^{90}Sr sample with ^{210}Pb interference; (C) chemical blank.

The ^{90}Sr counting efficiency was found to be 100% by measuring a standard solution directly mixed with the scintillation cocktail. The chemical blank value in the ^{90}Sr region was 3.6 cpm (counts per minute) for a new Sr·Spec® column. With a maximum counting time of 400 min the lower limit of detection (LLD), given by the formula of Seymour et al. (1992) was 0.009 Bq/g ^{90}Sr (here a chemical yield of 80% was assumed). As the activity concentrations of our samples were clearly higher than the LLD, we often used shorter counting times down to 60 min.

The method was verified by measuring a solution of 1 g of hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$, the inorganic bone compound) in 5 ml of 8 M HNO_3 with a certain amount of our in-house standard added, as well as by re-measuring reindeer bone ash samples, which had for their part already been processed following the classical IAEA method (Bossew et al., 2000). The result for the standard sample was within the error bars of the standard (4%), while the real samples differed by 3–10%. This was found satisfying, especially as no one-sided trend was found.

3. Results

Table 1 summarizes the results of our measurements. The activity of the interfering ^{210}Pb is also given; if ^{210}Pb was not observed in the spectrum, a less-than-value was given for it

Table 1
^{90}Sr and ^{210}Pb activity concentrations of samples determined using methods 1–3

Sample site, altitude	Code	Method	^{210}Pb act. conc. (Bq/g)	^{90}Sr yield (%)	^{90}Sr act. conc. (Bq/g)
Salzburg (S), 425 m	1A	Water	<0.002	87	0.095 ± 0.003
	1B	Water	<0.002	95	0.067 ± 0.002
	1C	Water	<0.002	74	0.088 ± 0.003
Hornsburg (NÖ), 277 m	2A	Water	0.001	87	0.026 ± 0.003
	2B	Water	0.007	83	0.040 ± 0.003
Lassee (NÖ), 148 m	3A	Water	0.007	73	0.030 ± 0.003
	3B	Water	0.003	72	0.021 ± 0.002
Kaunertal (T), 1700 m	4A	Water	0.008	84	0.414 ± 0.005
		Oxalic acid rinse	0.005	78	0.423 ± 0.010
	4B	Water	0.027	78	0.425 ± 0.005
		Oxalic acid rinse	<0.005	50	0.409 ± 0.005
		Oxalic acid rinse	<0.005	68	0.398 ± 0.005
	4C	Water	0.015	92	0.399 ± 0.009
		Oxalic acid rinse	<0.005	80	0.429 ± 0.011
Ternitz (NÖ), 427 m	5A	Water	0.015	65	0.113 ± 0.003
	5B	Water	0.010	68	0.117 ± 0.005
Petronell (NÖ), 175 m	7A	Water	0.014	54	0.050 ± 0.004
		Oxalic acid rinse	<0.001	76	0.050 ± 0.003
Lobau (NÖ), 152 m	8B	Oxalic acid rinse	0.014	76	0.038 ± 0.003
		Oxalic acid rinse	<0.001	69	0.029 ± 0.003
Neuwaldegg (NÖ), 271 m	13A	Oxalic acid rinse	<0.002	66	0.091 ± 0.004
	13B	Oxalic acid rinse	<0.002	78	0.084 ± 0.003
	13C	Oxalic acid rinse	<0.002	73	0.083 ± 0.004
Neuwaldegg (W), 271 m	20B	HNO_3	0.001	67	0.041 ± 0.003
Neuwaldegg (W), 271 m	21B	HNO_3	<0.002	94	0.075 ± 0.003
Zwettl (NÖ), 518 m	22B	HNO_3	<0.004	67	0.151 ± 0.004

depending of the ^{90}Sr count rate and the counting time. The table contains all samples with a chemical recovery determination; data from this table are used below for discussion of the reusability of the columns and comparison of the activity concentrations.

3.1. Prevention of ^{210}Pb contamination

When we learned from our spectra, that ^{210}Pb was also eluted from the column together with ^{90}Sr with distilled water, 29 more samples were processed in order to determine the best method to reduce the ^{210}Pb impurities (the daughter products ^{210}Bi and ^{210}Po are not retained on the column). In this case a yield determination was not carried out. Altogether 29 samples were processed following method 2, i.e. rinsing the column with 3 M HNO_3 + 0.05 M $(COOH)_2 \cdot 2H_2O$ to wash the lead from the column before eluting Sr with distilled water. However, the lead cannot be eluted with 10 ml of 3 M HNO_3 + 0.05 M $(COOH)_2 \cdot 2H_2O$; under the described conditions it is wandering very slowly down the column. On average we observed 3–4 column runs with pure ^{90}Sr spectra, while in the following runs the ^{210}Pb peak was found again. That means, the ^{210}Pb found together with a sample's ^{90}Sr does not represent the ^{210}Pb activity of that specific sample; it originates from samples processed some runs before. Moreover, in sample 4B we found the lowest ^{90}Sr recovery (50%) of all samples investigated: ^{90}Sr had been partly eluted with the rinsing solution, where it was indicated by measurement.

Seventeen samples were then processed following method 3, i.e. Sr was eluted with 0.05 M HNO_3. These samples were processed on columns that had been used before with method 2 and had also delivered ^{210}Pb. With method 3, however, ^{210}Pb was only found—if at all—in the first run (as a remainder from the method 2 procedure); in the subsequent runs it was retained on the column. As the ^{210}Pb activities measured were small compared to the ^{90}Sr activities (<20%) and as the measurement was started shortly after sample preparation before ^{210}Bi ingrowth (^{210}Bi would overflow the ^{90}Sr region), we saw no necessity to separate the lead before loading the sample solution onto the column.

In 10 samples processed following method 3, however, quenching was observed. This is due to the fact that the recommended maximum amount of water miscible with 12 ml of the cocktail HiSafe III® is only slightly more than 8 ml; if the sample is acid as in our case, the maximum amount is even less. To circumvent the quenching, the HNO_3 solution must be heated to dryness and the residue must be dissolved in a volume of 0.05 M HNO_3 clearly smaller than 8 ml.

3.2. Chemical yield and reusability of the columns

On one certain column 19 samples were processed and the yield of the procedure was determined. Between samples, the column was washed with 10 ml of H_2O or 0.05 M HNO_3, depending on the method used. The first 12 samples were eluted with distilled water, the next four samples were prepared following method 2 with the 3 M HNO_3 + 0.05 M $(COOH)_2 \cdot 2H_2O$ rinse; the last three samples were eluted with 0.05 M HNO_3. Fig. 2 shows the results: the maximum yield of the water elution method when using a new column was 95%, then the yield decreased to 54% in experiment 12. With the 3 M HNO_3 + 0.05 M $(COOH)_2 \cdot 2H_2O$ rinse in runs 13–16 the yield showed variations between 50% (the minimum of all samples) and 80%. With the 0.05 M HNO_3 method the yields from experiments 17–19 were 70% (twice) and 67%. However, when we used this method on other columns in run 5 and 9, we found clearly higher yields of 90% (sample 5C) and 94% (sample 21B), respectively, while the 3 M HNO_3 + 0.05 M $(COOH)_2 \cdot 2H_2O$ rinse method had yields below 80% even on run 1 (i.e. a new column) to 4.

At the beginning and then after run 3, 6 and so on, a blank value was measured. A slight increase of the blank value from (3.6 ± 0.1) cpm at the beginning, to (3.9 ± 0.1) cpm after 19 runs was observed; results of samples processed from run 11 forward were corrected by that slightly elevated blank value.

On a separate column we processed artificial samples (a solution of 1 g hydroxyapatite in 5 ml of 8 M HNO_3) with ^{90}Sr activities between 0.05 and 1.0 Bq, again with the above described washing procedures between successive runs. No cross-contamination was observed,

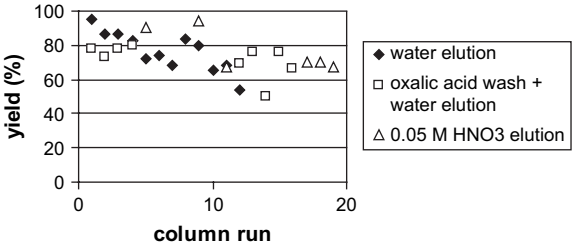

Fig. 2. Recovery for different methods and reusability of columns; the accuracy of the yield determination is ±5%.

the elution plus the washing completely removed the Sr from the column; blank levels were the same as given above.

From these results we conclude that Sr·Spec® columns can be reused for at least 15 times. While Brun et al. (2002) could reuse their resin only three times (investigation of milk samples), only a little bit lower reuse numbers than ours were given by Saxen (2002) and Jeter and Grob (1994); the latter supposed that the decreasing chemical yield is caused by release of the crown ether extractant material from the inert resin substrate after repeated acid washing.

3.3. Distribution of activity concentrations within the bone

As already mentioned above, the bones were divided into three parts by sawing; B is the midshaft, A and C are the end parts. For samples 1 and 3, the middle parts of the bones showed levels lower than the end parts, for sample 2 it was quite the contrary. The other investigated samples showed no difference within the 1σ-error bars. Momeni et al. (1975) investigated humeral bones of beagles and found a rather constant ^{90}Sr distribution in the different bone sections with only slightly lower values at the ends, when the dog had been fed continuously with ^{90}Sr enriched food. When the ^{90}Sr feeding had been stopped a few years before death and investigation of the animal, the ^{90}Sr burden per gram of dehydrated bone of the midshaft was lower by 15%, while the difference between the end parts was 75% and more. This means, that ^{90}Sr is eliminated more rapidly from the end parts due to faster bone turnover rates there relative to the midshaft. In the case of the investigated roe deer the ^{90}Sr supply was continuous and so our findings of a homogeneous distribution within the bone are in accordance with the above cited literature.

For the discussion following, the numerical mean values of the different bone parts were used.

3.4. Discussion of some environmental aspects

Although we can only present measurements of a single animal for each site with the exception of Neuwaldegg, some general conclusions can be drawn.

The value from Kaunertal (Tyrol) is by far the highest with 0.414 ± 0.011 Bq/g, followed by Zwettl (northwestern part of Niederösterreich, Lower Austria) with 0.151 Bq/g; all other samples stem—with the exception of the deer from Salzburg—from the east and south of Lower Austria and from Vienna. Strictly speaking, when comparing these data from different areas, one should take into account the different ages of the investigated animals, as Sr is accumulated over the years and therefore an older animal is expected to show higher levels than a younger one. This can also be clearly seen from our samples 13 (age 6 years), 21 (3 years) and 20 (1 year) from Neuwaldegg (Vienna). The 6-year-old deer had collected twice the amount of ^{90}Sr found in the 1-year-old animal; the 3-year-old showed a value only 14% lower than the 6-year-old deer. On the other hand, data given by Tatzber et al. (1982) indicated that the scatter of ^{90}Sr activity concentrations from animals from the same region can be a factor of 2.5 even for animals of the same age; moreover, older animals showed often lower values, especially for low activity concentrations. Therefore, when comparing our results with each other and with the above cited data by Tatzber et al. (1982), we do it irrespective of different ages.

Tatzber et al. (1982) found 1978–1980, i.e. about 15 years after the maximum of bomb fallout, ^{90}Sr activity concentrations between 14 and 60 mBq/g bone in deer samples from Lower Austria with site altitudes up to 240 m a.s.l.; our findings in this region from the years

2001–2002 were in the same range: between 25 and 91 mBq/g bone. Considering the fact that the ^{90}Sr from the bomb fallout measured by Tatzber et al. (1982) must have decreased to a proportion of 0.58 till the year 2001, one can see that the Chernobyl accident had approximately doubled the ^{90}Sr levels present in nature. This is in accordance with measurements and calculations by Mück et al. (1988), who estimated the additional deposition of ^{90}Sr in Austria from Chernobyl fallout in 1986 to be in the same range as deposition from bomb ^{90}Sr still present in the environment.

At the site of Zwettl, ^{90}Sr activity concentrations between 41 and 71 mBq/g bone were found in the years 1978–1980, while in this investigation a clearly higher value of 151 mBq/g bone was measured. This high level may be a consequence of the Chernobyl accident; elevated levels were also found 30 km west of Zwettl, were spruce needle samples were taken for ^{137}Cs and ^{90}Sr measurements (Irlweck et al., 1999). Here in 1986 the portion of Chernobyl ^{90}Sr due to uptake of airborne radioactivity was up to nearly twice the bomb fallout ^{90}Sr due to root uptake (Lenz, 1997).

The only high-altitude site Kaunertal (a valley) is between 1270 and 1770 m a.s.l., but the deer is roaming up to much higher altitudes, so the 1700 m a.s.l. given above is only a mean value. It can be compared with the site Gastein/Naßfeld (1630 m a.s.l.) given in Tatzber et al. (1982) with activity concentrations between 400 and 622 mBq/g bone in the year 1979 and corresponding values of 230 to 360 mBq/g bone for the year 2001, provided that the ^{90}Sr had not vanished from the plant–soil zone by other mechanisms than radioactive decay. The 414 mBq/g bone from the Kaunertal is higher than these decay corrected numbers. This is surprising, as this part of western Tyrol was only slightly affected by Chernobyl releases; therefore we suppose that the measured ^{90}Sr activity dates from the weapon tests fallout. While Tatzber et al. (1982) found a good correlation between amount of precipitation and fallout activity concentration in deer for ^{137}Cs, ^{90}Sr was correlated with the site altitude and so different deposition mechanisms for ^{137}Cs and ^{90}Sr were presumed. Also the Kaunertal ^{90}Sr level cannot be explained by high annual precipitation levels, as the Kaunertal is an arid alpine zone with only 835 mm precipitation in the valley at 1273 m a.s.l. (Hydrographisches Zentralbüro, 1994), compared to 1744 mm in Gastein/Naßfeld.

Fig. 3 shows our results together with the decay corrected data from Tatzber et al. (1982); the middle and high altitude data from the literature are lower than ours, probably indicating the impact of Chernobyl; the Kaunertal value may be an exception. We plan to extend sampling at higher altitudes to fill the gap between 500 and 1700 m site altitude with modern data; this together with analyses of soil samples should help to get a better understanding of the deposition mechanism of ^{90}Sr.

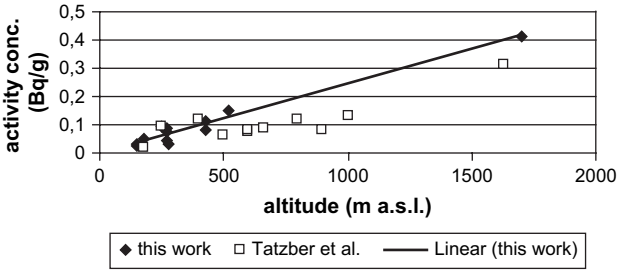

Fig. 3. ^{90}Sr activity concentration against site altitude.

4. Conclusions

Strontium specific ion exchange columns in combination with liquid scintillation spectrometry is a tool very well suited for the determination of ^{90}Sr levels in bones. ^{210}Pb, also present in the bones and interfering the ^{90}Sr measurement, is retained on the column, if Sr is eluted with 0.05 M HNO_3. The columns can be reused several times.

A comparison of the activity concentrations in the midshaft and the end parts of the bones showed no difference between the distinct parts. For animals from the same site we found the ^{90}Sr levels increasing with age.

The measured activity concentrations were between 0.025 ± 0.003 Bq/g bone in Lower Austria, and 0.414 ± 0.011 Bq/g bone in Tyrol; the values are correlated with the altitude of the site, where the animals roamed. Comparing our results with former measurements, we can confirm that generally the ^{90}Sr fallout from the Chernobyl accident in 1986 was in the same range as the ^{90}Sr burden from the nuclear bomb test fallout still present in the environment.

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