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Lead in Bone: Sampling and Quantitation Using K X-Rays Excited by ^{109}Cd

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Lead in bone can be measured *in vivo* using γ-rays from a ^{109}Cd source to excite lead K X-rays. Normalization of lead X-ray amplitudes to that of the elastically backscattered 88 keV γ-rays produces a determination of the concentration of lead in bone mineral that is accurate and insensitive to variations in measurement or bone geometry. For *in vivo* tibia measurements, a typical precision (1 SD) of ± 5 μg lead (g bone mineral)$^{-1}$ is achieved for an effective dose equivalent of 2.1 μSv. Measurement can be made of any superficial bone site, but precision will vary approximately as the inverse of the square root of the mass of bone mineral sampled. The apparatus required for this technique is readily transportable, and mobile laboratory facilities are easily established.

Introduction

The noninvasive measurement of bone lead humans was first reported by Ahlgren et al. in 1976 (*1*). Their X-ray fluorescence system used γ-rays from ^{57}Co to excite the K series X-rays of lead. This approach has been further developed (*2,3*) and adopted elsewhere (*4*). Subsequently, an alternative approach, which used ^{109}Cd rather than ^{57}Co to excite lead K-series X-rays, was proposed (*5*) and developed (*6,7*). A substantially different methodology, while still being X-ray fluorescence, has been employed in measuring bone lead by means of the L-series X-rays, using either radioisotope sources (*8,9*) or polarized X-rays (*10*) as the incident radiation. It is the use of ^{109}Cd to excite lead K-series X-rays that will be considered here, as this approach has considerable flexibility in being able to sample lead in a range of different bone sites and a robust normalization technique, which obviates the need to correct for bone geometry, thickness of overlying tissue, and other factors.

^{109}Cd Technique for K X-Ray Fluorescence of Lead

The equipment used in these measurements is shown schematically in Figure 1. ^{109}Cd emits γ-rays of 88.035 keV, just above the 88.005 keV K-shell absorption edge in lead, in 3.6% of its decays; it also emits silver K X-rays of 22 or 25 keV with an intensity 25 to 30 times higher. These silver X-rays are filtered out by 0.5 mm of copper in front of the source, which has little effect on the intensity of 88 keV γ-rays. The γ-rays can interact with a K-shell electron in lead, ejecting it with the resulting vacancy being filled by less tightly bound electrons and energy being released as K-series X-rays (96% of transitions) or Auger electrons.

In addition, Compton scattering of incident γ-rays can occur in the subject, resulting in scattered photons of reduced energy, the energy depending on the angle of scatter. In this case (Fig. 1), the mean angle of scatter is a little over 150°, and the corresponding energy of Compton-scattered photons is 66.5 keV. A large angle of scatter is an essential feature of the use of ^{109}Cd to measure lead because the Compton-scatter peak is then well below the X-ray peaks; for example, were a 90° scattering angle used, this would result in Compton-scattered photons of 75.1 keV, which would obscure the lead Kα X-rays.

Elastic scattering, the remaining important process characterizing this technique, results in a scattered photon, essentially of unchanged energy; the photon direction is usually only slightly altered but occasion-

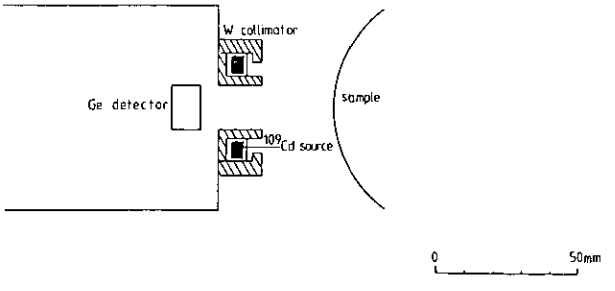

FIGURE 1. Equipment for K X-ray fluorescence of lead using ^{109}Cd.

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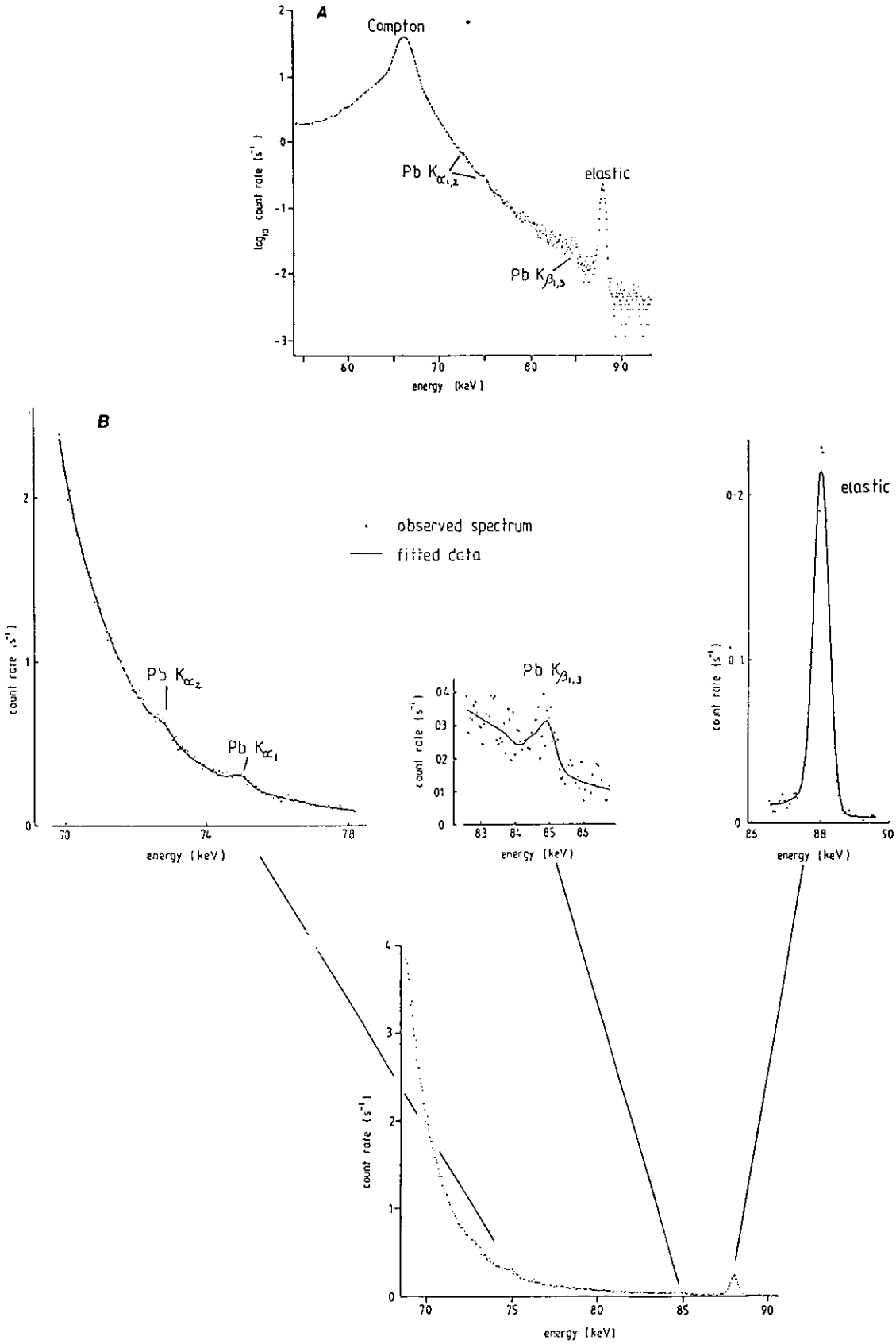

FIGURE 2. (*A*) Photon energy spectrum of tibia of a lead worker, estimated to have 123 μg lead (g bone mineral)$^{-1}$. (*B*) Expanded spectrum showing regions fitted for quantitation.

ally is changed by the large angles used for these measurements. A photon energy spectrum from a tibia measurement of a lead worker (Fig. 2) shows that Compton scattering is the dominant contributor to the observed spectrum, although the elastically scattered 88 keV peak is also clearly visible. Also shown are the three regions of the spectrum used for quantitating data, *viz*, 70 to 78 keV ($K\alpha_2$: 72.8 keV and $K\alpha_1$: 75.0 keV), 82 to 86.5 keV ($K\beta_3$: 84.5 keV and $K\beta_1$: 85.0 keV) and 86.6 to 89.5 keV ($K\beta_2$: 87.3 keV and elastic scatter: 88 keV).

Elastic Scatter Normalization

As noted above, elastic scattering, particularly at these large angles (~150°), is a low probability event compared to Compton scattering. However, the cross-section at this energy and angle varies very strongly with the atomic number (z) of a target atom. This variation is illustrated in Figure 3 over a range of elements found in human tissue. Since the cross-section depends on the fifth or sixth power of z, the observed elastic scatter peak comes predominantly from the higher z elements present. In the case of a bone, such as tibia or calcaneus, 98 to 99% of the elastic scatter signal arises from the bone mineral, rather than other tissue components. Two further factors are also pertinent to the normalization process. First, the lead X-ray signal also arises from the bone as this is the principal storage site for lead and no subsidiary storage site, such as liver or kidney, is likely to be in view during a bone lead measurement. Also, although lead concentration in bone has been shown to vary sharply over distances of the order of 100 μm (*11*), it is effectively uniformly distributed on the integrated scale defined by the penetration of lead K X-rays (~30 mm). Second, effectively, only uncollided incident γ-rays can produce lead K X-rays because the 88.035 keV γ-rays from ^{109}Cd are only some 30 eV above the lead K absorption edge. Thus it is the same photon fluence that gives rise to both the elastic scatter peak and the lead K X-ray peaks.

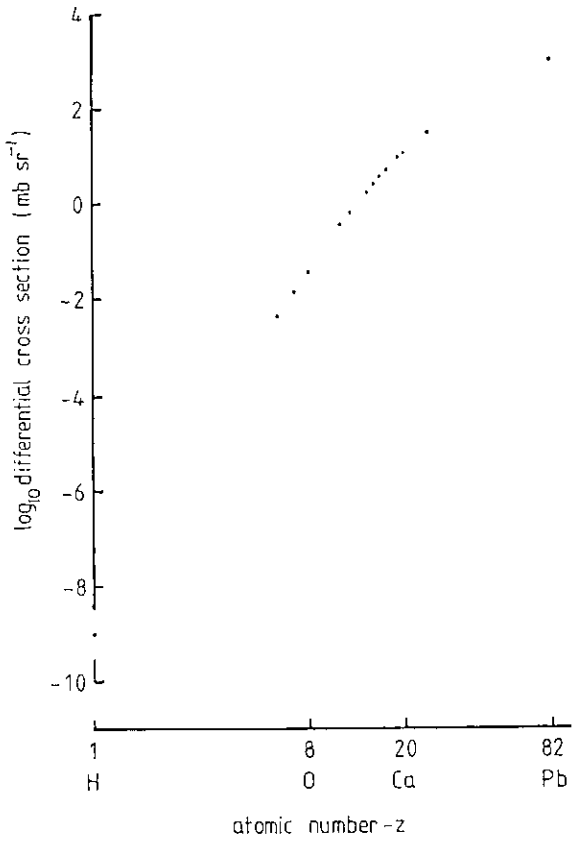

FIGURE 3. Elastic scatter cross-section at 88 keV and 150° as a function of atomic number.

The combination of these three factors produces a robust normalization of lead to bone mineral when the ratio is taken of the amplitudes of lead K X-rays to the elastic scatter peak. The effectiveness of this normalization procedure is shown by Figure 4, which shows

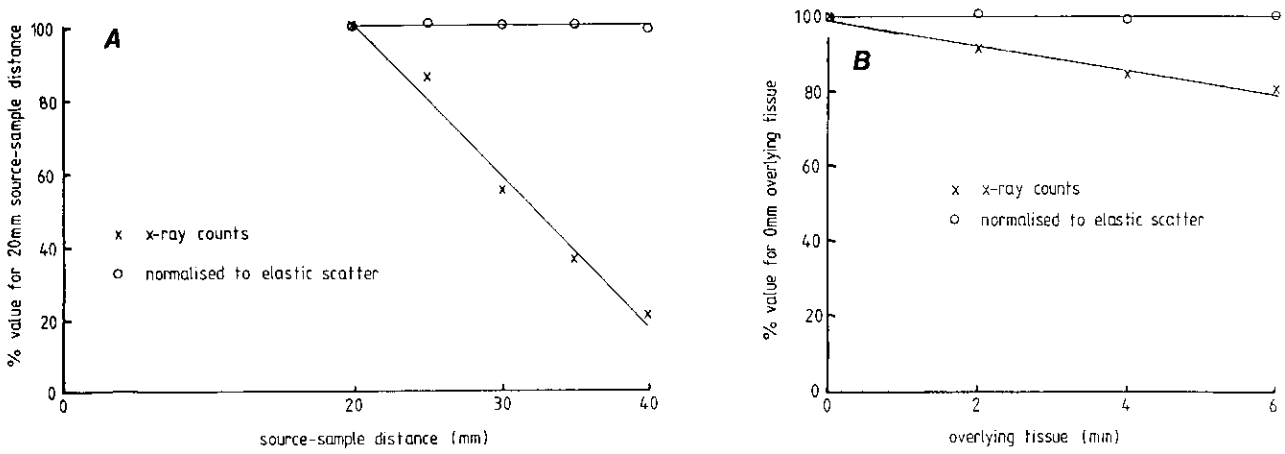

FIGURE 4. Elastic scatter normalization. (*A*) Source to sample distance; (*B*) thickness of overlying tissue.

Table 1. Elastic scatter cross-sections for bone mineral and plaster of Paris.

	Elastic scatter, $\mu m^2\ g^{-1}$		
	140°	153.25°[a]	170°
Bone mineral	7677	7420	7283
Plaster of Paris	5285	5082	4983
Correction factor	1.453	1.460	1.462

[a] Mean scattering angle for system shown in Figure 1.

how the ratio of lead X-ray to elastic amplitude is insensitive to either source to sample distance or thickness of overlying tissue, despite marked variations in lead X-ray counts alone.

The X-ray to elastic ratio observed in bones is calibrated against plaster of Paris (calcium sulfate) phantoms doped with known quantities of lead. A correction then has to be made to allow for the difference in elastic scattering cross-section between plaster of Paris and bone mineral. The cross-sections have been interpolated from the tabulated data of Hubbell and Overbo (*12*). As can be seen from Table 1, the cross-sections are a weakly varying function of angle, so the fact that a range of angles is subtended in any measurement system is unlikely to distort the normalization; thus, the mean scattering angle in a particular measurement geometry can be used.

Quantitation, Precision, and Dose

Although the lead $K\alpha_1$ and $K\alpha_2$ X-rays are the most intense, the background underlying the $K\beta_1$ and $K\beta_3$ is so much less that they can still contribute usefully to the overall precision. However, in order to extract data from the $K\beta_{1,3}$ peaks, account must be taken of the feature at 84.0 keV, which is an edge marking the upper end of the bremsstrahlung spectrum of photoelectrons ejected from the K shell of calcium by the 88 keV γ-rays (*13*). There are less prominent, but analogous features, at 85.9 keV from phosphorus, in the case of bone mineral, and at 85.6 keV from sulfur, in the case of plaster of Paris. The lead $K\beta_2$ line, although used for *in vitro* analyses, is not used for *in vivo* analyses because there is an oxygen photoelectron bremsstrahlung edge that interferes with it. The height of this latter feature varies with the amount of soft tissue overlying and surrounding the bone.

The measurement precision, although not the accuracy, varies somewhat from person to person, depending on factors such as the amount of overlying tissue and the mass of bone mineral sampled. In a typical field survey of *in vivo* tibia lead measurements, the standard deviation on an individual measurement (σ_i) varies from about ± 3.5 to ± 10.0 μg lead (g bone mineral)$^{-1}$ for a skin dose of 0.5 mGy. The corresponding root mean square standard deviation, that is,

$$\left[\left(\sum_{i=1}^{n} \sigma_i^2\right)\Big/n\right]^{1/2}$$

Table 2. Contribution of different K X-ray peaks to precision.

X-rays	Relative intensity	SD for analysis based on different X-rays, μg (g bone mineral)$^{-1}$
$K\alpha_1$	1.0	6.6
$K\alpha_2$	0.593	17.0
$K\beta_1$	0.228	9.9
$K\beta_3$	0.119	17.7
$K\alpha_1$, + $K\alpha_2$	(1.0 + 0.593)	6.2
$K\beta_1$, + $K\beta_3$	(0.228 + 0.119)	8.6
$K\alpha_{1,2}$ + $K\beta_{1,3}$	(1.0 + 0.593 + 0.228 + 0.119)	5.0

is 5.0 μg lead (g bone mineral)$^{-1}$. The contributions of the different K X-rays to the overall precision are summarized in Table 2, from which it can be seen that analysis of $K\alpha_1$ alone would result in a precision of ± 6.6 μg lead (g bone mineral)$^{-1}$ and that this figure is reduced to 5.0 μg lead (g bone mineral)$^{-1}$ when the other peaks are also analyzed. A practical detection threshold for lead in tibia can then be taken to be 10 μg (g bone mineral)$^{-1}$. In adult tibia this equates to 5.6 μg lead (g wet bone)$^{-1}$; however, results are more conveniently quoted with respect to bone mineral as this reflects the normalization process and enables straightforward comparison between bones, with no assumptions having to be made about bone mineral density.

The effective (whole body) dose equivalent of 2.1 μSv is low and can be put in context by comparison to natural background radiation. Such exposures vary from place to place but are often in the range of 2 to 3 mSv per year. Thus, the dose delivered during a bone lead measurement is approximately equivalent to 6 to 9 hr of natural background radiation.

Comparisons between X-Ray Fluorescence and Atomic Absorption Spectroscopy

In order to validate the ^{109}Cd K X-ray fluorescence technique for bone lead measurements, sets of bone samples have been analyzed both by atomic absorption spectroscopy and by X-ray fluorescence. Atomic absorption spectroscopy measurements were performed by Wittmers et al. in the manner they have described previously (*14*). The samples used were about 20 mg of bone mineral and were removed prior to the samples being analyzed by X-ray fluorescence. The X-ray fluorescence technique sampled up to 10 to 15 g of bone mineral, less in the case of tibia fragments. Thus, the masses of the bones sampled by the two techniques were significantly different. The data comparing the two measurement techniques have been reported previously (*15,16*) and are summarized in Table 3. Table 3 shows the mean differences between X-ray fluorescence and atomic absorption measurements, their standard deviation, and the reduced χ^2 statistics, as a test of observed variance of the differences compared to their pooled measurement variances.

Table 3. Comparison of X-ray fluorescence (XRF) and atomic absorption (AA) results.

Bone sample	n	Mean (XRF-AA) ±SD, μg lead (g bone mineral)$^{-1}$	χ^2
Metatarsal	6	−3.4 ± 4.1	0.43
Tibia section	3	4.9 ± 4.3	0.74
Tibia section	16	1.0 ± 7.6	4.36
Tibia section	22	−1.0 ± 9.9	3.92
Tibia fragment	11	−3.0 ± 6.2	1.93
Calcaneus	22	2.0 ± 23.7	5.66
Combined sets			
Tibia section	41	0.2 ± 8.8	3.90
All bones	80	0.0 ± 14.1	4.01

There is clearly no evidence for a systematic difference in the results obtained by the two different techniques, and it is unlikely that a difference of more than 3 μg lead (g bone mineral)$^{-1}$ could exist. The χ^2 is highly significant, indicating that there is greater random variation between the measurements than that predicted by the known measurement precisions of the two techniques. It is probable that at least some of this increased variance is associated with the different masses of bones sampled.

Sampling Different Bone Sites

The ^{109}Cd K X-ray fluorescence technique has been used to sample different bone sites, principally tibia and calcaneus, although other bones have been measured in individuals and on surveys. The initial reason for measuring more than one bone site was to sample differing types of bone, the midshaft of the tibia being largely cortical, while the calcaneus is trabecular apart from a thin cortical sheath. Rather surprisingly, the correlation between calcaneus and tibia in a cross-sectional survey, which included retired as well as active lead workers, was very strong (r = 0.893, n = 112) (*17*). This relationship is illustrated in Figure 5.

One important factor in deciding on which bone sites to sample is the precision that will be obtained on the measurements. Precision depends on the mass of bone mineral sampled, the amount of tissue overlying the bone, and the geometry of the measurement. The resulting wide differences in the height of the elastic scatter peak are illustrated by Figure 6, which shows these portions of spectra from measurements at four different bone sites on the same person. The precision, which varies inversely with the square root of the area of the elastic scatter peak, ranged from ± 4.1 μg lead (g bone mineral)$^{-1}$ for a tibia measurement to ± 23.2 μg lead (g bone mineral)$^{-1}$ for a rib measurement.

A second important factor in choosing a bone site is the degree to which it might be representative of the entire bone mass in the body. Wittmers et al. (*18*) examined five different bone sites at autopsy in 134 people covering ages 0 to 98 years and concluded that of tibia, vertebra, rib, ilium, and skull, the tibia best reflected estimated total skeletal lead burden over the wide range of ages. In addition, *in vivo* tibia lead data

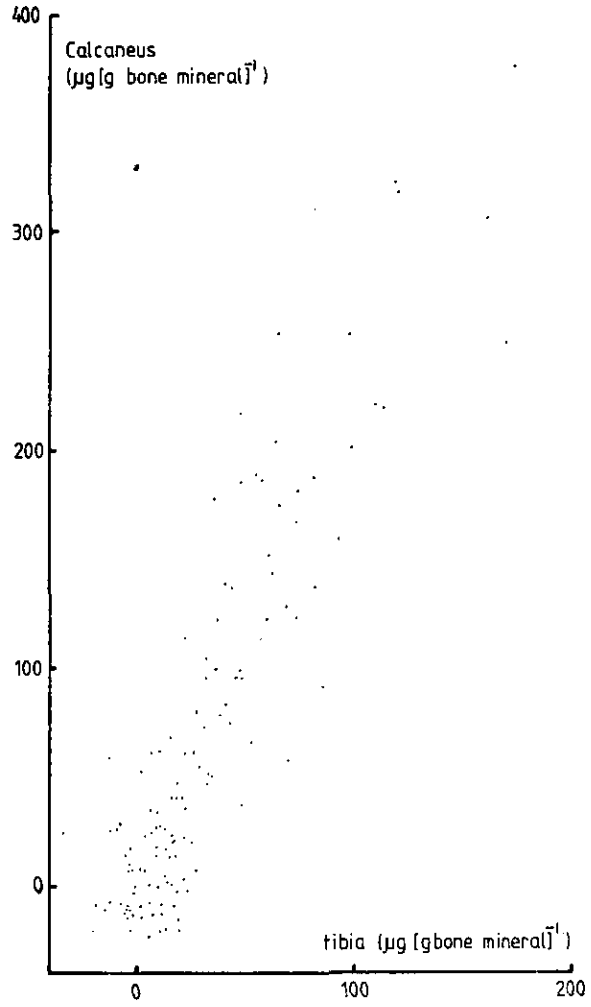

FIGURE 5. Calcaneus lead versus tibia lead in a cross-sectional survey.

have been shown to reflect cumulative exposure in that they correlate closely with an index of blood lead integrated over a worker's occupational exposure history (*19*).

The apparatus required for measuring bone lead by the ^{109}Cd K X-ray techniques is not bulky and is therefore easily transported, for example, to a factory site. We have conducted surveys at different sites within England, Sweden, and Finland and simultaneous measurements have been made of more than one bone (Fig. 7). Figure 7 shows a worker being measured in our caravan, equipped as a mobile laboratory.

Conclusions

The ^{109}Cd technique for K X-ray fluorescence of lead has been clearly demonstrated to be a practical and convenient way of monitoring lead stores in the body. The use of the elastic scatter normalization makes the measurement robust, in that it is insensitive to geo-

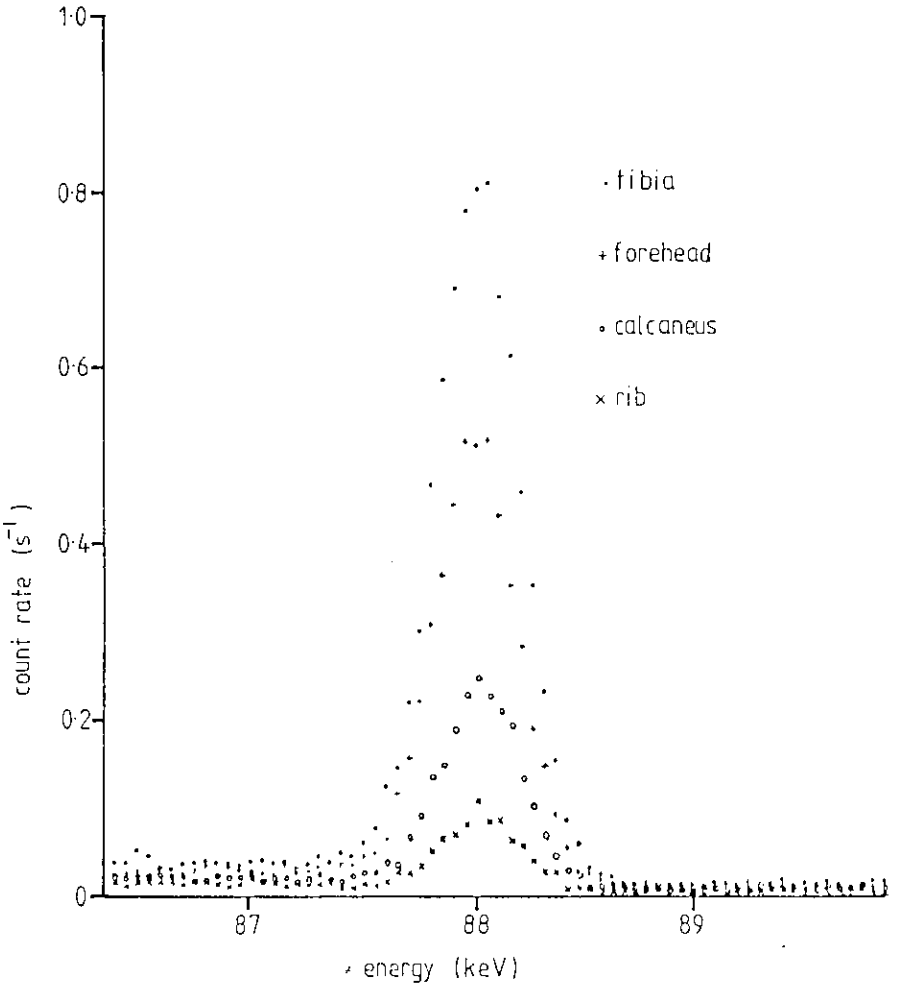

FIGURE 6. Elastic scatter peaks from measurements at four sites. Tibia, elastic amplitude, 1177, precision ± 4.1 μg lead. Forehead, elastic amplitude, 737, precision ± 5.6 μg lead. Calcaneus, elastic amplitude, 327, precision ± 10.2 μg lead. Rib, elastic amplitude, 120, precision ± 23.2 μg lead. Precision expressed as lead (g bone mineral)$^{-1}$.

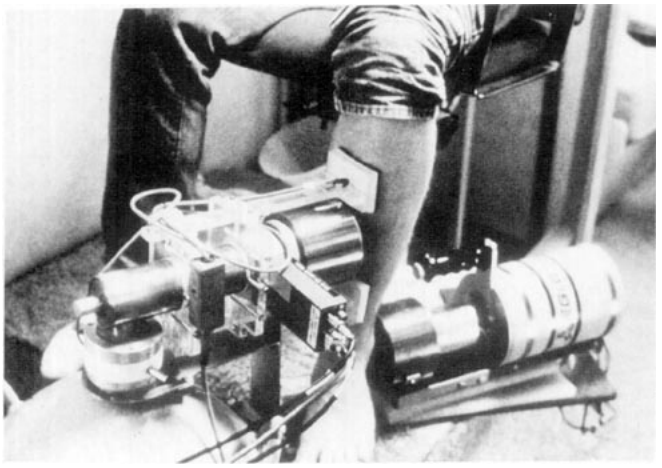

FIGURE 7. Simultaneous *in vivo* bone lead measurements of tibia and calcaneus.

metrical variations, and accurate as an assessment of lead concentration with respect to bone mineral. Quantitation based on four X-rays yields a precision of ± 5 μg lead (g bone mineral)$^{-1}$ for a very low effective dose equivalent of 2.1 μSv. The accuracy of the technique has been confirmed by intercomparison with atomic absorption spectroscopy. These points combine to underline that this methodology has many attractions for use in the study of exposure to lead and its behavior in the body.

If a single bone site is to be monitored to obtain an assessment of skeletal lead burden, then tibia is the preferred site. There remains, however, considerable interest in the comparative lead levels in different bone types and, in this context, calcaneus may be the best available way of monitoring a trabecular bone with an *in vivo* measurement. This area of bone lead metabolism clearly requires further work. It should be noted that this technique has not yet been applied to

studies of childhood lead exposure. In appropriate circumstances, there is every reason to expect it to yield valuable and quantitatively reliable information, but the precision obtained in measuring a child's tibia will be worse than that for an adult because the mass of bone mineral sampled will be considerably less. Also, interpretation of such data would be more complex than for adult studies, as a smaller and more variable proportion of a child's lead body burden is found in bone (*20*). Finally, it should be noted that these bone lead measurements can be conducted from a mobile facility, making it relatively convenient to conduct field surveys.

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